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BLOOD INDICES OF AFRICAN CATFISH (*CLARIAS GARIEPINUS*) FOLLOWING DIETARY ADMINISTRATION OF *TALINUM TRIANGULAR*

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Abstract

The effects of *Talinum triangulare* on the haematological indices of *Clarias gariepinus* was investigated. *T. triangulare* leaf powder at 0, 0.5, 1.0, and 3.0% concentrations were fed to *Clarias gariepinus* juveniles (n=84; 117.3 ± 1.57 g; 26.70 ± 0.26 cm) as feed additive for a period of six (6) weeks. The packed cell volume (PCV) and Haemoglobin (Hb) concentration values, 34.67 ± 0.67%, 12.03 ± 0.26g/dl, respectively observed in the group fed with 1.0% were significantly higher (P<0.05) compare with the control group. Red blood cell counts (RBCs), white blood cell counts (WBCs), lymphocytes, MCV, MCH and MCHC recorded were marginally different (P>0.05) among the groups, were within standard for *Clarias gariepinus*, therefore adjudged not to indicate a negative physiological effect on the experimental fish. Blood indices values observed revealed that the varying additive levels of *T. triangulare* used had no negative physiological stress on the health status of the fish studied and haematological values observed were seen to be best in the group fed 1.0% *T. triangulare*. It could be recommended that 10g/kg of *Talinum triangulare* leaf powder be included in the diet of *Clarias gariepinus* for boosting of the animal blood and treatment of disease conditions such as anaemia.

Keywords: *Clarias Gariepinus*; Dietary Inclusion Levels; Haematology; *Talinum Triangulare*.

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1. Introduction

FAO recognizes the important contribution that fishery and aquaculture make to global poverty alleviation and food security. In the bulletin, state of world fisheries and aquaculture, 2016 report, it was stated that more than 800 million people worldwide suffer from chronic malnourishment and the global population is expected to grow by another 2 billion to reach 9.7 billion people by

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2050. The huge challenge now according to FAO, (2016) is how to feed our planet and at the same time safeguarding its natural resources for future generations. Aquaculture is a source not just of health but also of wealth. The sector provides jobs to tens of millions and supports the livelihoods of hundreds of millions. Billions of people, mostly in developing countries depend on fish as a primary source of animal protein (FAO, 2016).

In Nigeria, fish farming continues to make substantial contributions to source of animal protein requirements for her teaming population. Fish products are relatively cheaper compared to beef, pork and other animal protein sources in the country ((FDF, 2008). The culture of fish is receiving a lot of attention in Nigeria with the result that new cultivation techniques are being introduced and adopted. One can hardly speak about aquaculture today in Nigeria without the mentioning of artificial propagation of a culturable species, African Catfish (*Clarias gariepinus*) for commercial purposes. This fish species is most often chosen for its fast growing nature and its acceptability to consumers especially in Africa, Nigeria included. Adewumi & Olaleye (2011) pointed out that the story of the Nigerian aquaculture is essentially that of catfish farming.

The African catfish (*Clarias gariepinus*) is appreciated by consumers for the quality of its meat (Pruszyński, 2003), widely accepted among urban dwellers for pepper soup and is mostly smoked and used in soups as well. African sharp tooth catfish *Clarias gariepinus* is a typical air-breathing catfish with a scaleless, bony elongated body with long dorsal and anal fins, and a helmet like head (Plate 1).

Plate 1: African catfish (Clarias gariepinus) (Photo by Dr. Rutaisire, 2005)

It is recognized by its long dorsal and anal fins, which give it a rather eel-like appearance. The catfish has a slender body, a flat bony head, and a broad, terminal mouth with four pairs of barbells. Its prominent barbells give it the image of cat-like whiskers. The fish is mostly cultured in earthen ponds. However, it can be cultured in other systems such as tanks and hapas. In the wild and riverine systems, the fish reproduces naturally but considerable effort is required to induce spawning under culture conditions. The African catfish is an excellent species for aquaculture as it is omnivorous, grows fast, and tolerates relatively poor water quality **(**Rad *et. al*., 2003).

Despite the increasing effort channeled toward production of finfish in Nigeria, the demand of the ever increasing population in the country for fish and fish products has not been met (Ojutiku,

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2008). The challenges of nutritionally deficient diets which led to poor growth, losses associated with diseases were responsible for this observation. Disease causes economic losses because of fish mortality, treatment expenses, postponement or loss of the opportunity to sell the fish and contraction of zoonotic diseases by the handler and final consumer of the affected fish. The usage of antibiotics among other number of approaches applied to address these issues have been criticized because of the potential of antibiotics to enhance microbial resistance and the accumulation of residues in the tissues of the fish (Siwicki, 1989, Adedejt *et al*., 2011)

A number of plants have been continued to be investigated for their potential in supplementing animal diets in order to avoid or minimize usage of chemicals. Aletor and Adeogun, (1995) reported that several vegetable species abound in Nigeria and most West African countries where they are used partly as condiments or spices in human diets or as supplementary feeds to livestock such as rabbits, poultry, swine and cattle. These vegetables are harvested at all stages of growth and fed either as processed, semi-processed or fresh to man while they are usually offered fresh to livestock (Aja, 2007). One of such plant is Water leaf (*Talinum triangulare*) (Plate 2).

Plate 2: Water leaf (Talinum triangulare)

Talinum triangulare is a cosmopolitan weed belonging to the Talinum genus, family Portulacaceae that grows best under humid conditions (Burkil *et al*., 1994). It is one of the vegetables widely cultivated and consumed in Africa especially Southern Nigeria (Imoh *et al.*, 2000). (Ezekwe *et al.*, 2001) reported that *T. triangulare* is a rich source of vitamins, β-carotene, minerals (such as calcium, potassium and magnesium), pectin, proteins and vitamins. It has also been found to possess useful medical potentials such as laxative, purgative, treatment of diarrhea, gastro-intestinal diseases (Oguntona, 1998; Mensor *et al*, 2001) as well as in the management of cardiovascular diseases such as: stroke and obesity (Aja *et al*., 2010).

Since immense benefits have been derived by man from using medicinal herbs in management of health because they are relatively safer, more affordable and sometimes offer better therapeutic value than synthetic drugs, the increasing discovery of more medicinal plants has necessitated increased scientific scrutiny of their bioactivity in order to provide data that will help veterinarians, farmers, physicians and patients make wise decisions before using them (Oyewole and AKingbala, 2011). According to Fedato *et al.* (2010), analysis of haematological parameters are used to monitor health status and to diagnose diseases caused by various factors namely heavy metals, environ-mental stress, parasitic infections, genotoxic effect of pollutants, nutrition, and pollution.

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Many studies concerning the hematology of different fish species (Adeyemo, 2007; Adedeji & Adegbile 2011; Suleiman & Abdullahi, 2016) had been carried out and they had been used among the few tools available to monitor health status in different species of fish to detect acute and chronic patho-physiologic changes attributable to nutrition, water quality, toxicants, and disease. Oyawoye and Ogunkunle, (1998) pointed out that haematological components of blood are valuable in monitoring feed toxicity especially with feed constituents that affect the formation of blood in culture fisheries. Therefore, the aim of this study was to investigate the effect of *Talinum triangulare* powder fed as additive on the hematological parameters of experimental fish.

2. Materials and Methods

Eighty four African Catfish juvenileb of both sex (mean weight 117.3 ± 1.57 g; mean length 26.70 ± 0.26 cm) were sourced from a private fish farm at Olodo town in Oyo State, Nigeria. The fish were acclimatized for 14 days in 500L capacity circular tank fed with a locally formulated diet of 35% crude protein that did not contain herbal extract and no history of herbal feeding from the farm where fish were sourced. Afterward, fish were randomly divided into four sets in triplicates labelled (A, Control), B, C, and D, and fed with diet containing 0.0%, 0.5%, 1.0% and 3.0% of *Talinum Triangulare* powder as feed additives respectively. The experimental fish were fed at their 5% body weight twice daily for forty two days. The water quality parameters were maintained within recommended limits according to recommendation of Boyd, (1999) during this study. Blood sampling was conducted at the expiration of 42 days. Blood samples were collected from three fish randomly selected from each replicate with heparinized plastic syringe, fitted with 21 gauge hypodermic needle and preserved in disodium salt of ethylene-diaminetetraacetic acid (EDTA) bottles for analysis. The Blaxhall and Daisley (1973), Brown (1980) and Wedemeyer *et al. (*1983) haematological methods were adopted for this study. The cyano-haemoglobin method was used to determine haemoglobin (Hb) using diagnostic kits from Sigma diagnostics USA, and packed cell volume (PCV) was determined by the microhaematocrit method. Red blood cell (RBC) and thrombocyte count were determined with the improved Neubauer haemocytometer according to (Dacie and Lewis, 1991). White blood cells (WBC) was determined with the improved Neubauer counter, while differential counts such as neutrophils, lymphocytes and monocytes were determined on blood film stained with Giemsa stain. Mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were derived from the RBC, PCV and Hb using standard formulae (MAFF, 1984). MCV was calculated in femtoliters = PCV/RBC x 10, MCH was calculated in picograms = Hb/RBC x10 and MCHC = (Hb in 100mg blood / Hct) x 100.

3. Data Analysis

All statistical analyses were performed with Graph Pad Prism statistical software Version 5.1. All data are presented as means with standard error (SEM). The data were analyzed with one-way ANOVA, and differences among mean values were considered significant at $\alpha < 0.05$ with Tukey test used to compare differences among individual means.

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4. Results

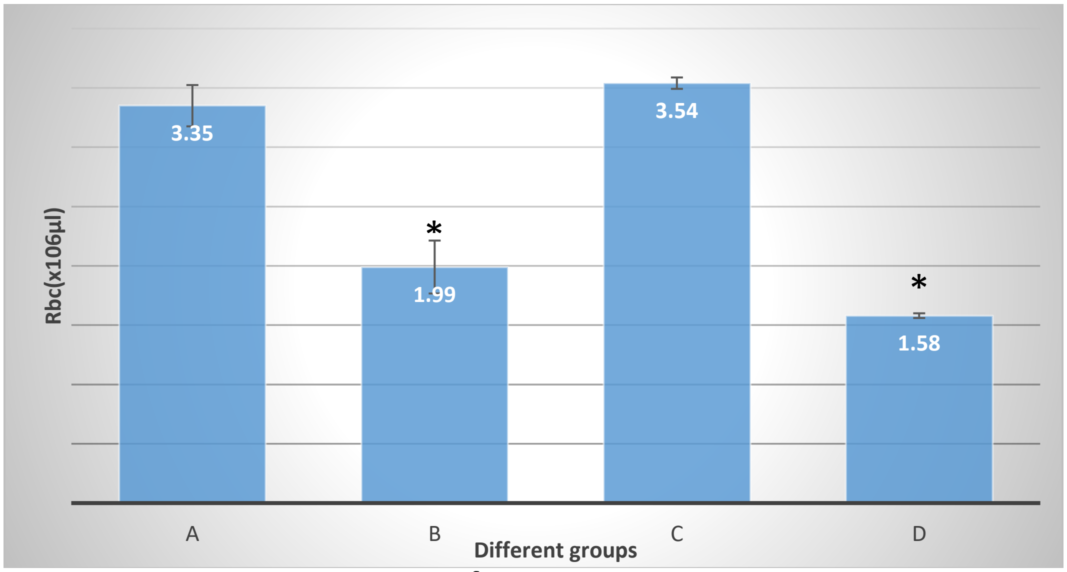

Figure 1: Red Blood Cells – RBC (cell x10^6µl) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period. *Significantl at P<0.05.

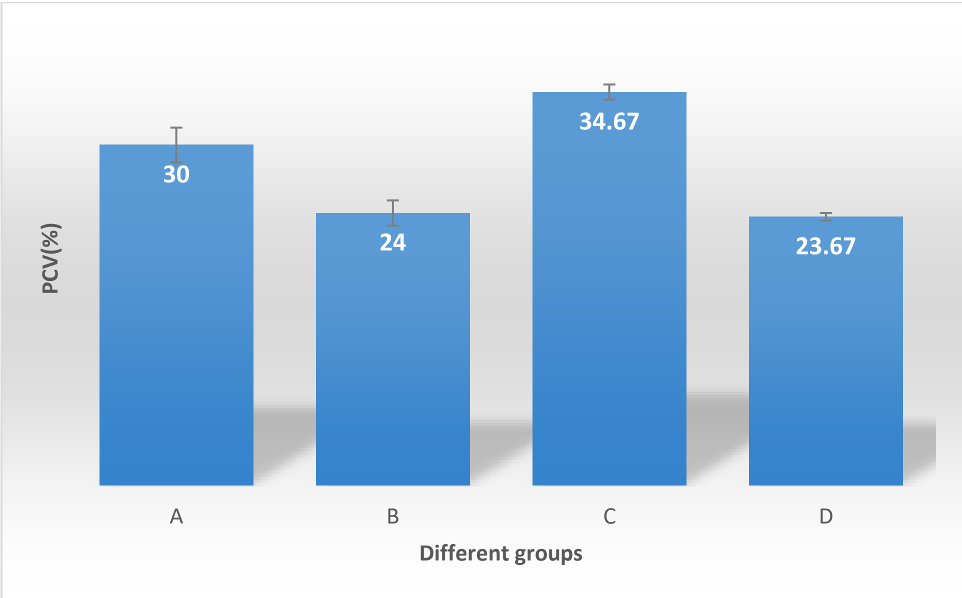

Figure 2: Packed cell volume -PCV (%) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period. *Significantl at P<0.05.

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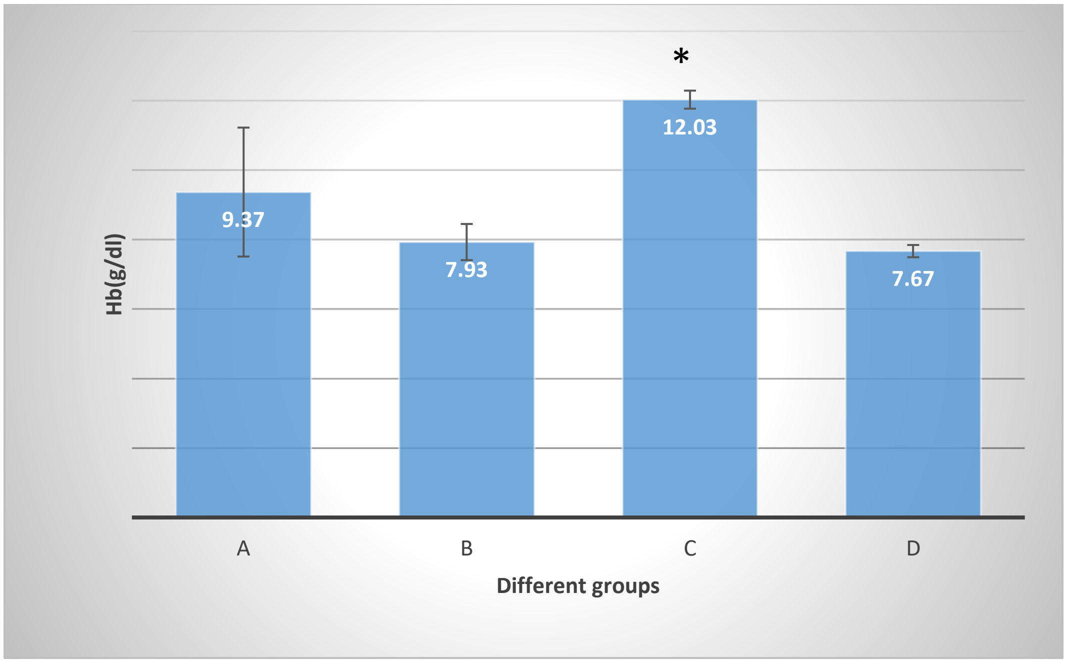

Figure 3: Haemoglobin -Hb (g/dl) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period.
*Significantl at P<0.05.

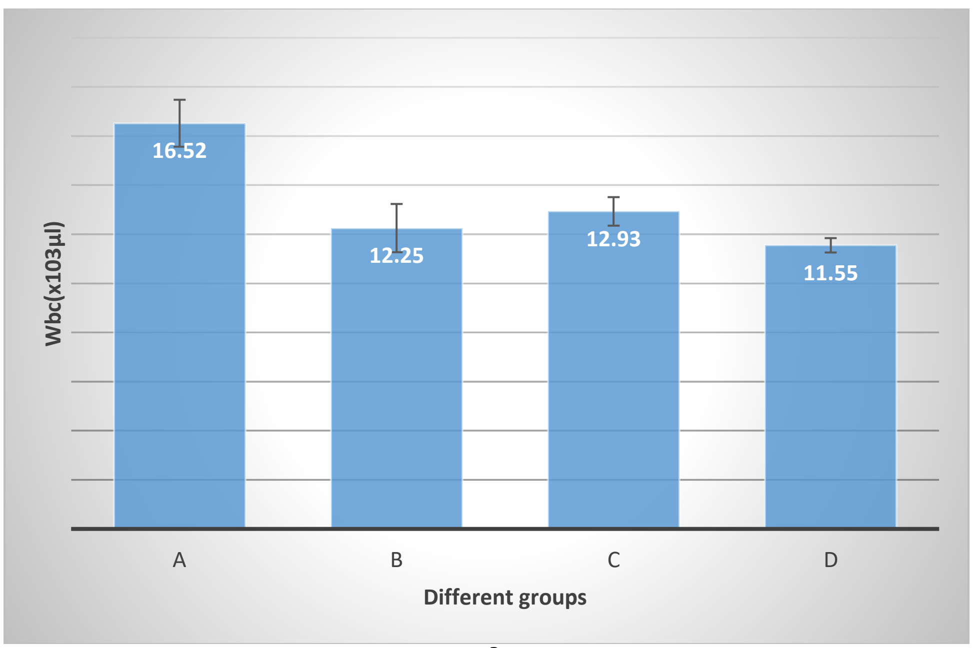

Figure 4: White blood cells -WBC (cells x 10^3µl) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period

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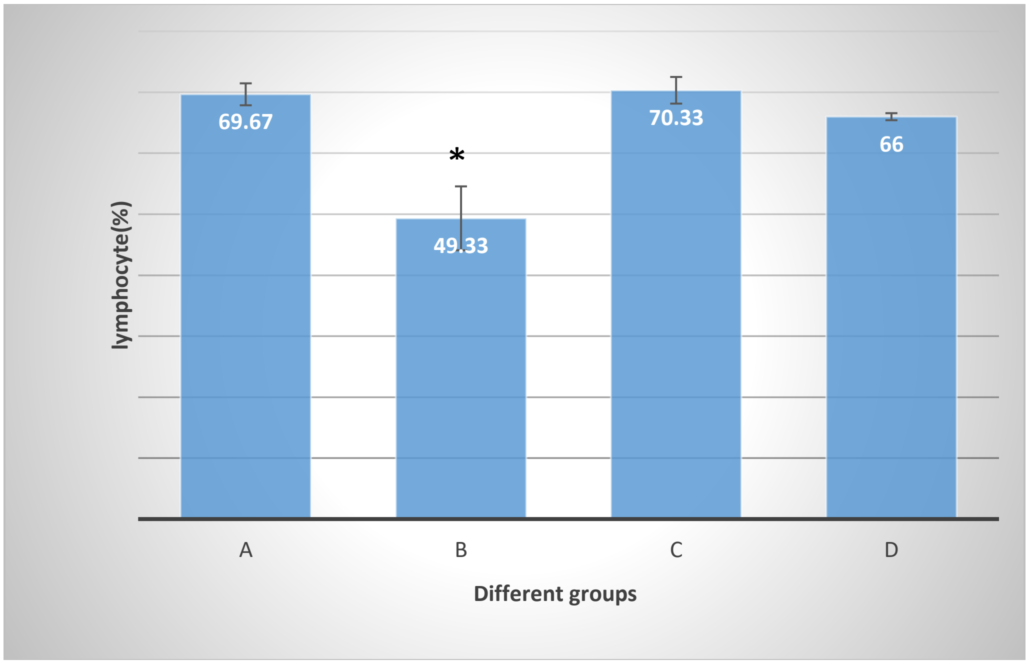

Figure 5: Lymphocytes (%) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period.
*Significantl at P<0.05.

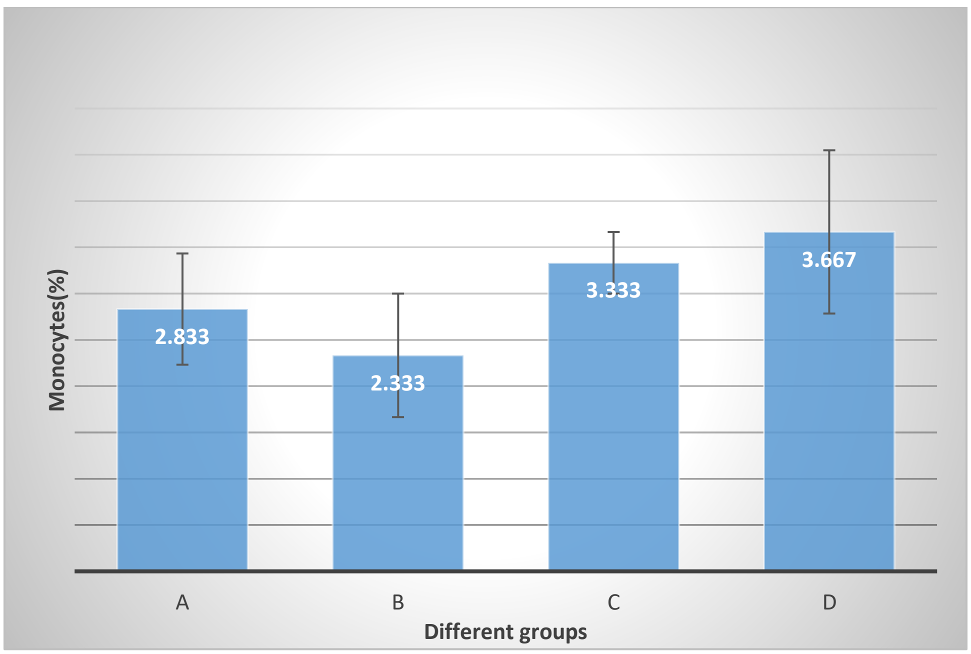

Figure 6: Monocytes (%) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period

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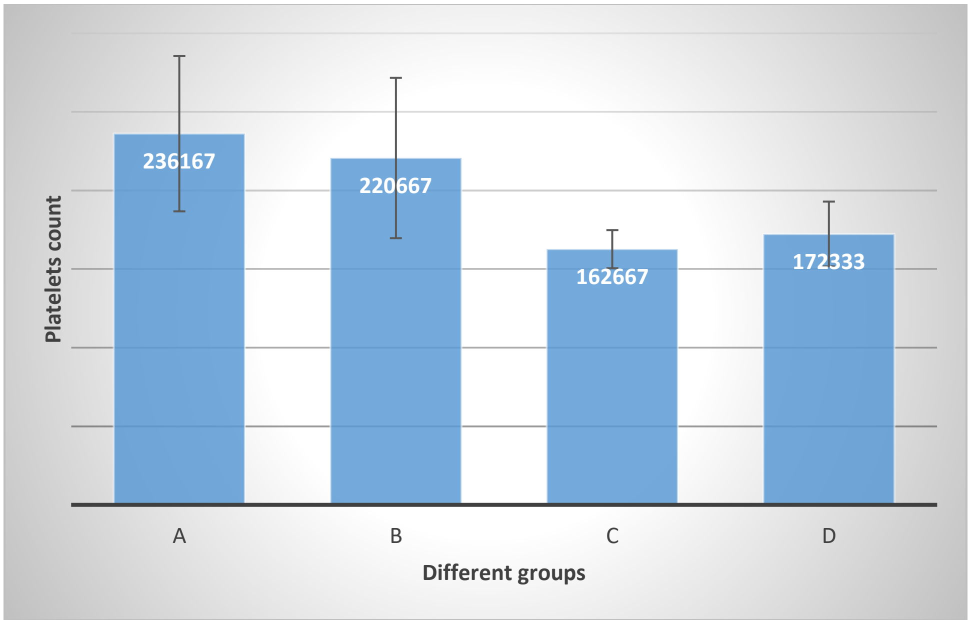

Figure 7: Platelet count of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period

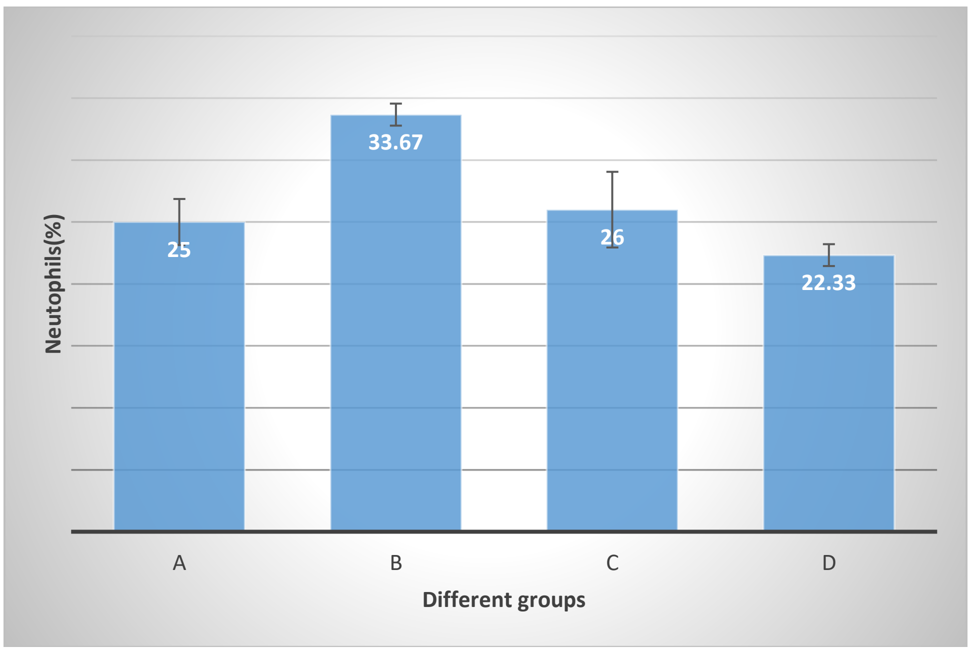

Figure 8: Neutrophil (%) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period

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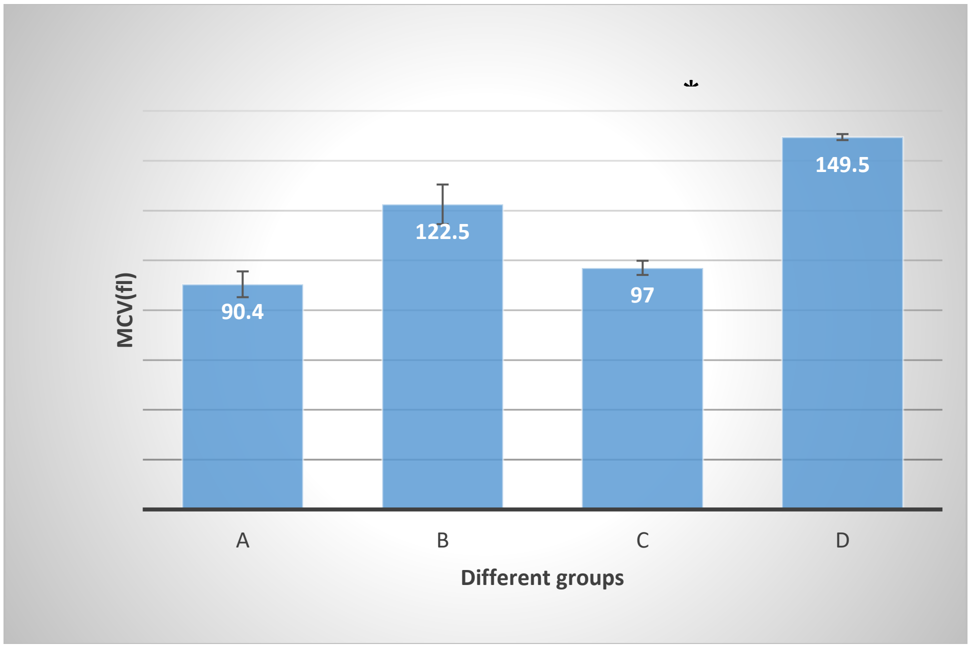

Figure 9: MCV (fl) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period.
*Significantl at P<0.05.

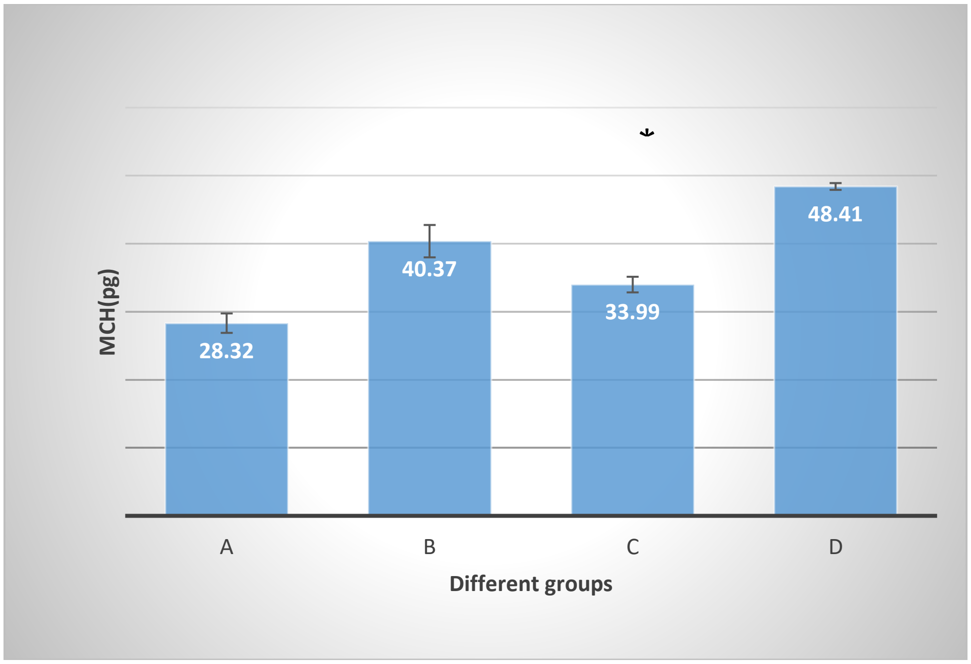

Figure 10: MCH (pg) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period.
*Significantl at P<0.05

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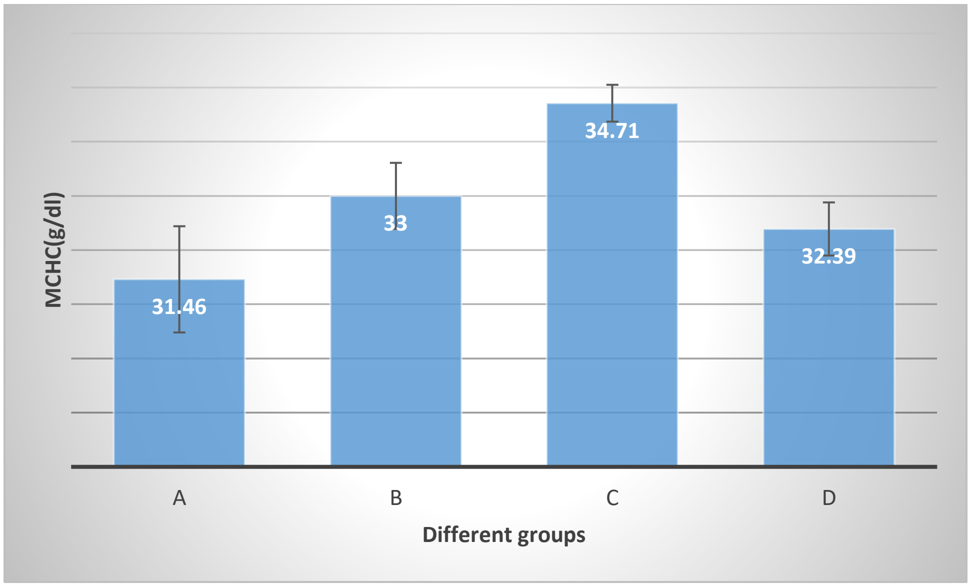

Figure 11: MCHC (g/dl) of groups of fish fed with *T. triangulare* powder as feed additive at inclusion rate of 0.0% (A), 0.5% (B), 1% (C) and 3% (D) for 42 day-period.

Figures 1 to 11 showed the results obtained for haematological indices of *African catfish (*Clarias gariepinus*) fed with locally formulated feed that was supplemented with various concentrations of Talinum triangulare* powder as feed additive for a period of 42 days. Statistical analysis conducted with the recorded values of packed cell volume (PCV) and hemoglobin (Hb) concentration from group of fish fed with 1.0% *T. triangulare* were significantly higher $P < 0.05$ than the control. The values of MCV and MCH recorded in the group of fish fed with 3.0% *T. triangulare* were equally significantly higher than the control group. Erythrocytes count (RBCs) recorded in the group of fish fed with 0.5% and 3.0% of *T. triangulare* were observed to be significant lower $P < 0.05$ than the control. Furthemore, a significant lower value $P < 0.05$ was recorded in the lymphocytes of group of fish fed with 0.5% *T. triangulare* when compared with control group. However, the results recorded for leukocytes (WBCs), monocytes, platelet count, neutrophils and MCHC recorded in the groups of fish fed with 0.5%, 1.0% and 3.0% *T. triangulare* were marginally different ($P>0.05$) when compared with the control group.

5. Discussion

Blood is an active transport medium in higher animals, especially in the vertebrates and it is explained to be a medium that constantly bathes all the organs and tissues of the body, enabling exchange of materials between the internal and external environment of the organs and tissues (Ramalingam, 2011). All the haematological parameters measured in this study were within the recommended physiological ranges reported for *C. gariepinus* according to the previous studies carried out by some workers (Erondu *et al*, 1993; Adeyemo *et al*., 2014, Okorie- Kanu *et al*., 2014). Aletor and Egberongbe (1998) reported that red blood cell counts and packed cell volume (PVC) are mostly affected by dietary treatment. PCV ranging from 23.67 ± 0.33% to 34.67 ± 0.67% observed in this study were within the range of 20 to 50% reported for African Catfish. The value of 7.67 ± 0.18 to 12.03 ± 0.26g/dl recorded for Hb concentrations were within the normal range reported by some researchers (Omitoyin, 2006; Osuigwe *et al*., 2007) for African Catfish. PCV and Hb are major and reliable indicators of various sources of stress (Rainza-Paiva *et al*, 2000)

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and these parameters decrease in the presence of anti-nutritional factors (Osuigwe *et al.,* 2007). Reduction in the concentration of the PCV in the blood usually suggests the presence of toxic factor example of which is haemagglutin which has adverse effect on blood formation (Oyawoye and Ogunkunle, 1998). This present study revealed the absence or reduction of toxic factor in the feed hence the experimental fish were not stressed. A significant increase in the values of PCV and Hb recorded in the group of fish fed with 1.0% of *T. triangulare* indicated that 10g/kg of water leaf added to the feed improved the blood level of the experimental fish. Blaxhall and Daisley (1973) reported the essence of using haematocrit to detect anaemic condition in fishes. The experimental fish were not anaemic as suggested by the improved values of PCVs recorded. According to Atamanalp and Yanık (2002) the low Hb levels may impair oxygen supply to the various tissues and result in slow metabolic rate and low energy production. (Satheeshkumer, *et al,* 2011) also reported low activity associated with low Hb value. Higher haemoglobin values indicate higher rate of transportation of oxygen to and removal of carbon (iv) oxide from the body tissues. This results in higher metabolism and growth. Low haemoglobin suggests a predisposition to anaemia. Values of erythrocytes (RBC) counts observed in the group of fish fed with 0.5 % and 3.0% *T. triangulare* were significantly lower compared with control whereas the value obtained in in the group of fish fed with 1.0% of *T. triangulare* was marginally different to the group of fish fed with 0.0% *T. triangulare* (Control). However the values obtained in all groups ranging from 1.58± 0.02 to 3.54± 0.05 x 10^6 µl with higher value recorded in the group of fish fed with 1.0% of *T. triangulare* were within the of 1.5 x 10^6 µl and 2.3 - 2.9 x 10^6 µl described for catfish by Adeyemo *et al*., 2003 and Gabriel *et al*., 2004 respectively. Higher value 3.54 (x 10^6µl) recorded for the group of fish fed with 1.0% of *T. triangulare* was similar to the finding of Dada and Ikuerowo (2009) who recorded the value of 3.50 ± 0.35 x 10^6 µl when ethanoic extracts of *Garcinia kola* seeds were fed to *Clarias gariepinus* brood stock.

White blood cells (WBC) and lymphocytes are reported to be the defense cells of the body and Douglas and Jane (2010) demonstrated that the amount present in the body of the animal has implication in immune responses and the ability of the animal to fight infection. High WBC count is usually associated with microbial infection or the presence of foreign body or antigen in the circulating system (Oyawoye & Ogunkunle, 1998). The results obtained for WBC ranged from 11.55 ± 0.29 to 12.93 ± 0.58 x 10^3 µl in treated groups were lower though not significantly lower compared with the control, an indication that no stimulation of the immune system in response to toxicity of feed additive, or otherwise could be explained that *T. triangulare* powder added to the feed is not toxic. Often the increase in WBC may be due to recruitment of more cells to combat the stressor (Ajani *et al.,* 2007). In the present study, the lymphocytes, mean value 49.33 ±5.24% to 70.33±5.24%, were observed, adjudged to be numerous than any other differential cells but typical of most fishes according to Owolabi, (2011). The abundance of neutrophils recorded in this fish species compared with the value of monocyte is also typical of most fishes (Owolabi, 2011). Monocytes range from 2.33± 0.67 to 3.67± 0.88% recorded in this study is also typical of most fish species and could be adjudged not to be influenced by the feed additive.

The MCV, MCH and MCHC increased in all treatments compared to the control with the significantly higher value ($P<0.05$) observed in MCV and MCH for the groups of fish fed with 3.0%*T. triangulare.* The values obtained ranging from 90.40±5.18 to 149.5±1.21 fl, 28.32±1.43 to 48.41±0.50pg and 31.46±0.98 to 34.71±0.34g/dl for MCV, MCH and MCHC respectively were similar to Peruzzi *et al*., (2005) findings.

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6. Conclusion

The results of this research provide the knowledge of the characteristics of haematological parameters of African Catfish fed with *T. triangulare* powder as feed additive. And from our findings it was noted that incorporation of *T. triangulare* feed additive did not affect the palatability of the diets as all the experimental diets were accepted by *Clarias gariepinus* juveniles and blood of the experimental fish improved. It can therefore be concluded that *T. triangulare* leaves can contribute significantly to the health management of animal. *T. triangulare* leaf could be used for treatment of disease conditions such as anaemia and boosting of blood in animal and Man. In addition, with the recent wind of economic depression and its resultant effect on the purchasing power of commodities for people living in less developed nations, it has become obvious that the local food stuffs and vegetables which prices are easily affordable, will play increasing roles in the food, nutrition and health security of the impoverished people living in the rural areas.

7. Recommendation

Since most of the haematological values obtained in this study were seen to be best in the fish fed 1.0% *T. triangulare* diet, it could be recommended that 10g/kg of *T. triangulare* leaf powder be included in the diet of African Catfish, the additive level had no negative physiological stress on the health status of the fish studied and this invariably will reduce production cost of the conventional additive and prevent incidence of emergence of bacterial resistance strains and antibiotic residue in food fish. Further investigation should be carried out on other fish species under different culture conditions either in earthen and concrete ponds.

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