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# The Effects of Persea americana leaves on the Enzymes and Organosomatic indices of Pseudomonas aeruginosa Infect Clarias gariepinus

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## 1. Abstract

Enzymatic activities (AST, ALT, ALP and ACP) and organosomatic indices were examined in *P. aeruginosa* infected *Clarias gariepinus*. 120 (one hundred and twenty) of *C. garipienus* of mean weight (120±13g) were intramuscularly injected with 1.0ml of $4.1x10^4$cfu *P. aeruginosa* using 2ml injection syringe and observed for disease presence/signs. After disease presence, they were distributed into four groups in triplicate and expose/treated via immersion with *P. americana* aqueous leaves extracts at 0.0ml, 1.0ml, 1.5ml and 2.0ml. Blood samples were collected before infection, after disease presence, and after day 2, 5, and 7 of treatment/exposure and taken to the laboratory to ascertain the therapeutic effects of the experimental extract on infected *C. gariepinus.* To this end the following enzymes were analysed: *aspartate transaminase* (AST), *alanine transaminase* (ALT), *alkaline phosphate* (ATP), and *acid phosphate* (ACP).The organosomatic indices were analysed at the day 7 of treatment period to ascertain the therapeutic effects of the extract on the organs of the infected *C. gariepinus.* The results shows that all the analysed enzymes (AST, ALT, ALP and ACP) were significantly higher in *C. gariepinus* after the infection compared to the activities before infection. The activities after day 2, 5 and 7 of the treatment shows that enzymes activities in the treated fish (1.0ml – 2.0ml of extracts) reduced ($P<0.05$) compared to the untreated (0.0ml). The organosomatic indicies assessment shows that hepatosomatic index (HSI), vescerasomatic index (VSI), spleenosomatic index (SSI) and intraperitoneal fat (FI) were higher ($P>0.05$) in the untreated (0.0ml) fish compared to the treated (1.0 – 2.0ml of experimental extract) fish. While the cardiosomatic index (CSI) was significantly the same across the treatments.

Keywords: Aquaculture, Enzymatic changes, *P. aeruginosa,* Organosomatic indices, *P. americana, C. gariepinus.*

## 1. INTRODUCTION

Aquaculture is one of the fastest growing food-producing sectors around the world (Harikrishnan *et al.,* 2011). The Worlds total production of fish and shellfish was 99 (ninty-nine) metric tonne in 1990 and it increased to 122 (one hundred and twenty-two) Metric tonne in 1997 (Hill, 2010), and the global aquaculture production has increased from about 28.3 million tonnes to 40 metric tonnes as at 2009 (FAO, 2009). According to Harikrishnan *et al.,* (2011), cultivating organisms, such as finfish and shellfish species constitute an important industry with yearly increase in production.

The importance of aquaculture in the world over, can never be over emphasized. The high demand for aquacultural products has led to employment opportunities in both developed and developing societies (Ukwe *et al.,* 2018). Aquaculture products such as fish are open to a wide range of bacterial pathogens, which have the capacity to cause diseases. (Schmdit *et al.,* 2000).

In aquaculture, disease control using chemotherapeutic agents has been difficult and most time harmful due to complicated instructions provided to the farmers by feed and chemical companies regarding the use of antibiotics and other therapeutic agents (FAO, 2003a). Application of antibiotics and other chemicals as prophylactic and therapeutic measures have been widely criticized for their negative impacts like immunosuppression and residue accumulation in tissues (Rijkers *et al.,* 1980; FAO, 2003b; Harikrishnan *et al.,* 2009a, 2009b) and the development of drugs resistant pathogens and environmental pollution (Smith *et al.,* 1994). International agencies recommends that the use of antibiotics should be restricted to therapeutic purposes only, and that preventive approaches should be preferred in fish disease management over costly post disease treatments (GESAMP, 1997; FAO, 2005). Commercial vaccines are expensive for fish farmers and are specific against particular pathogens (Raa *et al.,* 1992; Ukwe and Gabriel 2019).

It is widely demonstrated that farmed fish are more susceptible to diseases as they are caused by infection and stress (FAO, 2003a).

Medicinal herbal extracts are potential alternatives to synthetic drugs in aquaculture as they provide useful phytochemicals with various benefits such as immune system modulation (Zanuzzo *et al.,* 2015a; Yang *et al.,* 2015), growth promotion, antioxidants enhancement, appetite - stimulating effects, among others (Citarasu 2010; Zahran *et al.,* 2014; Abdel -Tawwab *et al.,* 2010) when properly administered. Medicinal herbal extracts are also easily available and inexpensive, and tend to be more biodegradable in nature compared to synthetic drugs (Olusola *et al.,* 2013; Reverter *et al.,* 2014). Ukwe and Gabriel (2019) state that the waste water from aquaculture involving herbs/herbal extracts can be used as growth enhancers in crop farming.

Herbs like neem leaves containing *nimbin*, *azadirachtin* and *meliantroil* have been reported to possess a variety of properties, including insecticidal and antiviral from ancient time (Biswas *et al,* 2002; Das *et al,* 2002). Indian almond *(terminalia catappa)* and garlic *(Allium sativum)* have been studied as an alternative to chemicals to treat fish ectoparasites, *trichodina sp*. infections in tilapia *(O. niloticus)* fingerlings. (Chitmanal *et al*, 2004).

Advantages of herbal plant extracts includes: readily available, cheap, environmental friendly etc. (Ukwe and Gabriel, 2019). Herbs/plants parts act as supplement in feed growth promoters, immune stimulants and antimicrobial agents (Ukwe et al., 2020a; Ukwe et al., 2020b; Ukwe and Gabriel, 2019). Use of medicinal plant is an alternative to antibiotics in fish heath management (Chakraborty and Chattopadhya, 1998). Many studies have proved that herbal additives enhanced the growth of fishes and also protection from the diseases (Sasmal *et al.,* 2005; Ukwe and Jamabo, 2020).

The genus *Clarias* belongs to the family Clariidae commonly known as African catfish (Teugels, *et al*., 1990). *C. geriepinus* as fresh water fish found in the tropical regions of West African. Although some species are distributed in Syria, Southern Turkey and throughout Southeast Asia, their diversity is greatest in Africa (Teugels, 1996; Teugels and Adriaens, 2003). Some of the generalized, fusiform species in Africa, such as *C. gariepinus* are broadly distributed, while the anguilliform species are restricted to swampy areas in the Nile Sudan, the Lower Guinea and the Zaire (Congo River basin) ichthyological provinces (Poll, 1957; Roberts, 1975; Teugels, 1986; Teugels et al., 1990). These adaptations lie at the base of the vast distribution of certain species, like the African species *C. gariepinus*, which has an almost Pan-African distribution and even occurs into the Middle East (Teugels, 1986).

The avocado tree belongs to the family Lauraceae, classified as *Persea americana* (Chen *et al.,* 2008). They are widely cultivated throughout the tropics and subtropics of the world for their edible fruits (Purseglove, 1977). Avocado are rich source of soluble phenolics, ascorbic acid compared to most common fruits and vegetables (Garcia-Alvarado *et al.,* 2001). It is recommended for gastritis, gastroduodenal ulcer, hypertension, anaemia and exhaustion (Pamplona and Roger, 1998) previous studies by Adeboye *et al.,* (1999) and Adeyeme *et al.,* (2002), have shown the pharmacological activity of *Persea americana.* The bark, leaves, stem and roots are used as local medicine against diseases (Neuwinger, 2000). In fish culture, phytochemicals in avocado pear powdered leaves have been proven to enhance immunastimuslant and growth in African catfish (*C. gariepinus)* (Ukwe *et al,* 2020a; Ukwe *et al*, 2020b).

## 2. MATERIALS AND METHODS

### Experimental Fish

One hundred and twenty (120) healthy *C. gariepinus* of mean weight 120 ± 13g were purchased from Aquaculture Centre of the Department of Fisheries and Aquatic Environment, Faculty of Agriculture. Rivers State University Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria. They were observed for two weeks to evaluate disease presences or bruises and were fed to satiation with blue crown commercial diets twice daily.

### Source of Pathogen

*Pseudomonas aeruginosa* was ordered from the National Veterinary Institute, Vom in Jos, Plateau State, Nigeria and was transferred to the Microbiology Department of the Rivers State University for preservation.

### Preparation of Experimental Herb

The *P. americana* aqueous leaf extract was prepared using the method of (Ukwe and Jamabo 2020). *P. americana* leaf was washed, clean, dried for four hours, pounded to paste and was soaked in tap water ($50^{o}$c) at the concentration of one hundred (100) grams per litre (100g/l) for twenty-four (24) hours. It was filtered and the filtrate was used immediately.

### Experimental Design

A complete randomized method (CRD) was used. There were four treatment in triplicates.

### Experimental Procedure

One hundred and twenty (120) *C. gariepinus* were injected intra muscularly with 1.0ml of 4.1 X $10^{4}$*cfu* overnight grown of *P. aeruginosa* using a 2ml injection syringe and 21 guage hypodemic niddle at day 1, 2, and 4, 5 and observed for disease presence/signs.

After disease presence/sign, the infected fish were distributed into four (4) groups of ten (10) fish each in triplicate and was treated via immersion with *P. americana* aqueous leaf extracts at 0.00ml/L, 1.0ml/L, 1.5ml/L and 2.0ml/L for eight (8) hours daily. Blood samples were collected before infection, after disease presence and after day 2, 5 and 7 of treatment/exposure and was taken to the laboratory to ascertain the therapeutic effect of the *P. amerciana* extracts on the enzyme activities of the infected fish (*C. gariepinus*) as a means of treatment. The organosomatic indices were also determined on the liver, heart, spleen, visceral and intraperitoneal fat after day 7 of the treatment/exposure to ascertain the medicinal effect of the *P. americana* aqueous leaf extract on the organs of the infected fish.

**Blood Extraction**

The fish was blindfold by covering the head with a thick cloth, to attain calmness, and blood was extracted via kidney puncture through the genital opening using 5ml injection syringe.

**Enzymes Analaysis**

The collected blood samples were transferred into LITHUM HEPARIN tube and sent to the laboratory for biochemical analysis. The blood was assayed for aspartate amino transferase (AST), alanine amonitransferase (ALT), alkaline phosphate (ALP) acid phosphate (ACP) This was done by the use of “Evolution 3000 Machine” an auto-analyzer, the screen master model, manufactured by Biochemical system, China. It was used according to manufacturers instructions.

**Organosomatic Indices (OSI)**

This was calculated as:

$$\text{OSI} = \frac{Weight\ of\ \ Organ}{Weight\ of\ \ Fish} \times 100 \text{(Dekie } et\ al.\text{, 2016)}$$

**Data Analysis**

The data were analysed using SPSS Statistics software 17.0 for Windows, a one-way analysis of variance was used to determine if there were difference in the variables among treatments. Turkeys multiple comparison test was used to compare the means of the treatments. (Wahua, 1999).

# 3 RESULTS

## 3.1 Enzyme Activities in Plasma biochemistry of *C. gariepinus* infected with *P. aeruginosa* and exposed to aqueous *P. americana* leaves extract.

The enzyme activities of the experimental *C. gariepinus* before and after the infection is shown in Table 4.1. All the analyzed enzymes increased in activities after the infection compared to the activities before infection. Tables 3.1 – 3.4 shows the enzyme activities in experimental fish after day 2, 5 and 7 of treatment/exposure to herb respectively. Though they all had reduced activities compared to the activities before the treatment, the activities were lower ($P<0.05$) in the treated groups (1.0ml – 2.0ml) compared to the activities in the untreated groups (0.0ml).

The comparative enzymatic activities of the experimental *C. gariepinus* within the period of the experiment is shown in Figures 3.1 – 3.5.

## 3.2 Organosomatic Indices of *C. gariepinus* infected with *P.aeruginosa* and exposed to various concentrations of *P.americana* leaf aqueous extract for seven (7) day

The result of the organosomatic indices in *C.gariepinus* after 7days of treatment are contained in Table 3.5. The values of HSI, SSI, VSI and IF were significantly higher in the 0.0ml aqueous *P. americana* leave extract compare to the other treatments that were significantly the same; except in the CSI that there was no significant difference acron the treatments.

**Table 3.1: Enzyme activities in the plasma biochemistry of *C. gariepinus* before and after infection (BI and AI) with *P.aeruginosa***

| Enzymes (IU/L) | *Before (BI) | Range | | *After (AI) | Range | |
|---|---|---|---|---|---|---|
| | | Min | Max | | Min | Max |
| AST | 22.00±2.00[a] | 20.00 | 24.00 | 39.67±8.62[b] | 32.00 | 49.00 |
| ALT | 3.53±0.51 [a] | 3.10 | 4.10 | 6.03± 2.16 [b] | 4.50 | 8.50 |
| ALP | 6.67±2.60 [a] | 4.00 | 9.20 | 10.80±0.72 [b] | 10.00 | 11.40 |
| ACP | 0.54±0.32 [a] | 0.35 | 1.00 | 1.08±0.15 [a] | 0.94 | 1.23 |

***Means within the same row with different superscripts are significantly different (P<0.05)**

**Key:** ALT -*Alanine transminase*; AST - *Aspartate transaminase* ; ALP -*Alkaline phosphatase* and ACP-*Acid phosphatase*

**Table 3.2: Enzymes activities in plasma biochemistry of *C. gariepinu*s infected with *P. aeruginosa* and exposed to different concentrations of *P. amaricana* leaf aqueous extracts for two (2) days**

| Concentration (ml) | Enzymes (IU/L) | | | |
|---|---|---|---|---|
| | AST | ALT | ALP | ACP |
| 0.0 | 33.67±7.57[b] | 5.00±1.77[c] | 9.03±1.70[c] | 1.12±0.11[b] |
| 1.0 | 26.67±5.50[a] | 4.50±0.96[b] | 5.98±0.96[a] | 0.71±0.38[a] |
| 1.5 | 25.00±13.89[a] | 3.83±0.85[a] | 7.63±3.40[b] | 0.70±0.40[a] |
| 2.0 | 25.66±4.04[a] | 4.30±0.40[b] | 7.93±2.87[b] | 0.85±0.33[a] |

**Means within the same column with different superscripts are significantly different (P<0.05)**

**Key:** ALT -*Alanine transaminase* ; AST - *Aspartate transaminase* ; ALP -*Alkaline phosphatase* and ACP-*Acid phosphatase*

**Table 3.3: Enzymes activities in plasma biochemistry of *C. gariepinus* infected with *P. aeruginosa* and exposed to different concentrations of *P. amaricana* leaf aqueous extracts for Five (5) days**

| Concentration (ml) | Enzymes (IU/L) AST | ALT | ALP | ACP |
|---|---|---|---|---|
| 0.0 | 30.33±9.71 [b] | 4.33±1.19 [b] | 8.33±2.25 [b] | 0.73±0.36 [a] |
| 1.0 | 26.66±11.93 [a] | 3.63±1.11 [a] | 6.76±2.88 [a] | 0.66±0.24 [a] |
| 1.5 | 25.00±11.00 [a] | 3.06±1.76 [a] | 6.66±2.38 [a] | 0.69±0.39 [a] |
| 2.0 | 25.33±12.85[a] | 3.43±1.25 [a] | 6.10±2.32 [a] | 0.66±0.34 [a] |

**Means within the same column with different superscripts are significantly different (P<0.05)**

**Key:** ALT -*Alanine transaminase* ; AST - *Aspartate transaminase* ; ALP -*Alkaline phosphatase* and ACP-*Acid phosphatase*

**Table 3.4: Enzymes activities in plasma biochemistry of *C. gariepinus* infected with *P. aeruginosa* and exposed to different concentrations of *P. amaricana* leaf aqueous extracts for Seven (7) days**

| Concentration (ml) | Enzymes (IU/L) | | | |
|---|---|---|---|---|
| | AST | ALT | ALP | ACP |
| 0.0 | 26.66±13.01 [b] | 4.05±1.70 [b] | 7.43±2.70 [b] | 0.69±0.30 [a] |
| 1.0 | 24.33±8.50 [b] | 4.80±0.80 [b] | 6.36±2.92 [a] | 0.63±0.40 [a] |
| 1.5 | 21.33±10.96 [a] | 4.13±3.69 [b] | 6.36 ±1.90 [a] | 0.61±0.32 [a] |
| 2.0 | 23.33±15.37[a] | 3.03±0.77 [a] | 6.40±1.21 [a] | 0.68±0.27 [a] |

**Means within the same column with different superscripts are significantly different (P<0.05)**
**Key:** ALT -*Alanine transminase*; AST - *Aspartate transaminase*; ALP -*Alkaline phosphatase* and ACP *Acid phosphatase*

**Table 3.5: Organosomatic Indices of *C. gariepinus* infected with *P. aeruginosa* and exposed to different concentration of *P.americana* leaf aqueous extracts for seven (7) days**

| CONC (ml) | HSI (%) | SSI (%) | CSI (%) | VSI (%) | IF (%) |
|---|---|---|---|---|---|
| 0.00 | 0.0578±0.02[b] | 0.0059±0.00[b] | 0.0024±0.00[a] | 0.2408±0.02[b] | 0.0941±0.02[b] |
| 1.00 | 0.0357±0.01[a] | 0.0035±0.00[a] | 0.0024±0.00[a] | 0.1855±0.02[a] | 0.0734±0.01[a] |
| 1.50 | 0.0328±0.02[a] | 0.0037±0.00[a] | 0.0020±0.00[a] | 0.1795±0.01[a] | 0.0709±0.01[a] |
| 2.00 | 0.0399±0.01[a] | 0.0038±0.00[a] | 0.0029±0.00[a] | 0.1721±0.02[a] | 0.0749±0.02[a] |

**Means within the same column with different superscripts are significantly different**

**Key:** HSI-Hepatosomatic Index; SSI- Spleenosomatic Index; CSI- Cardiosomatic Index; VSI- Viscerasomatic Index; IF- Intraperitoneal Fat

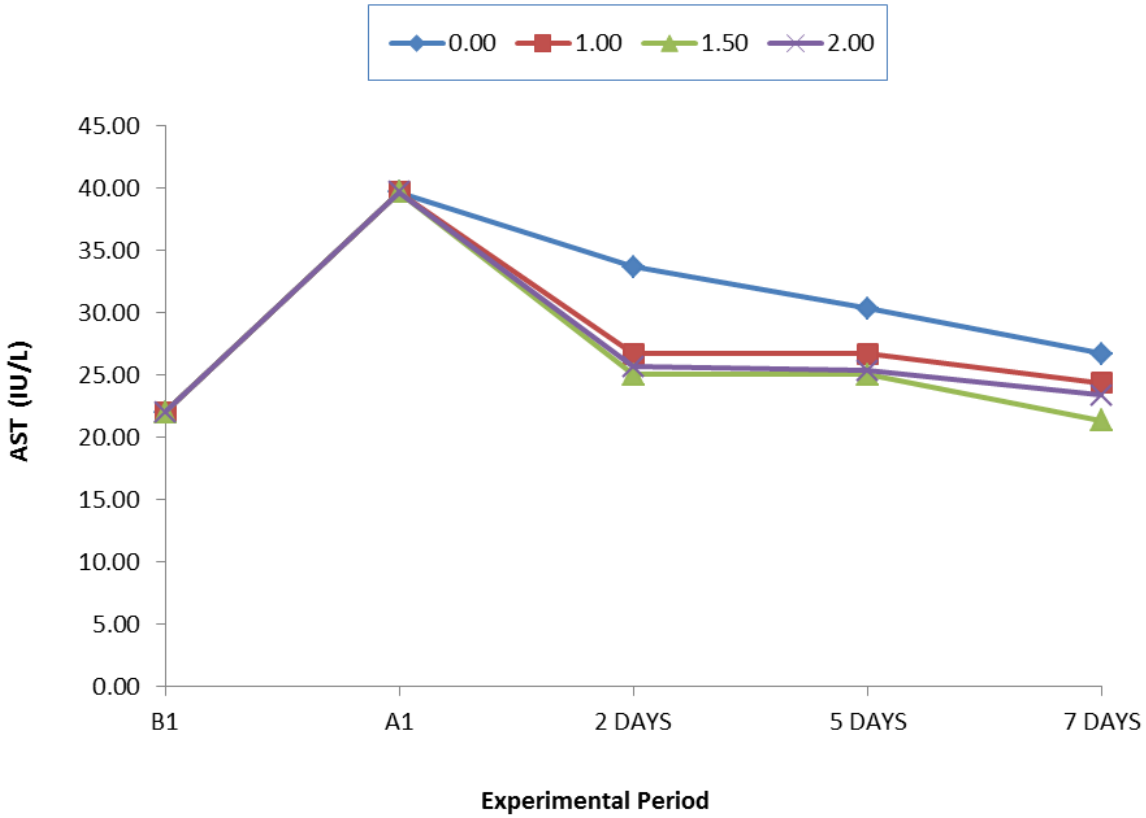


**Figure 4.1: Comparative Values of AST in the Plasma of *C.gariepinus* Infected with *P.aeruginosa* and exposed to *P.americana* Extracts at different Experimental Period**

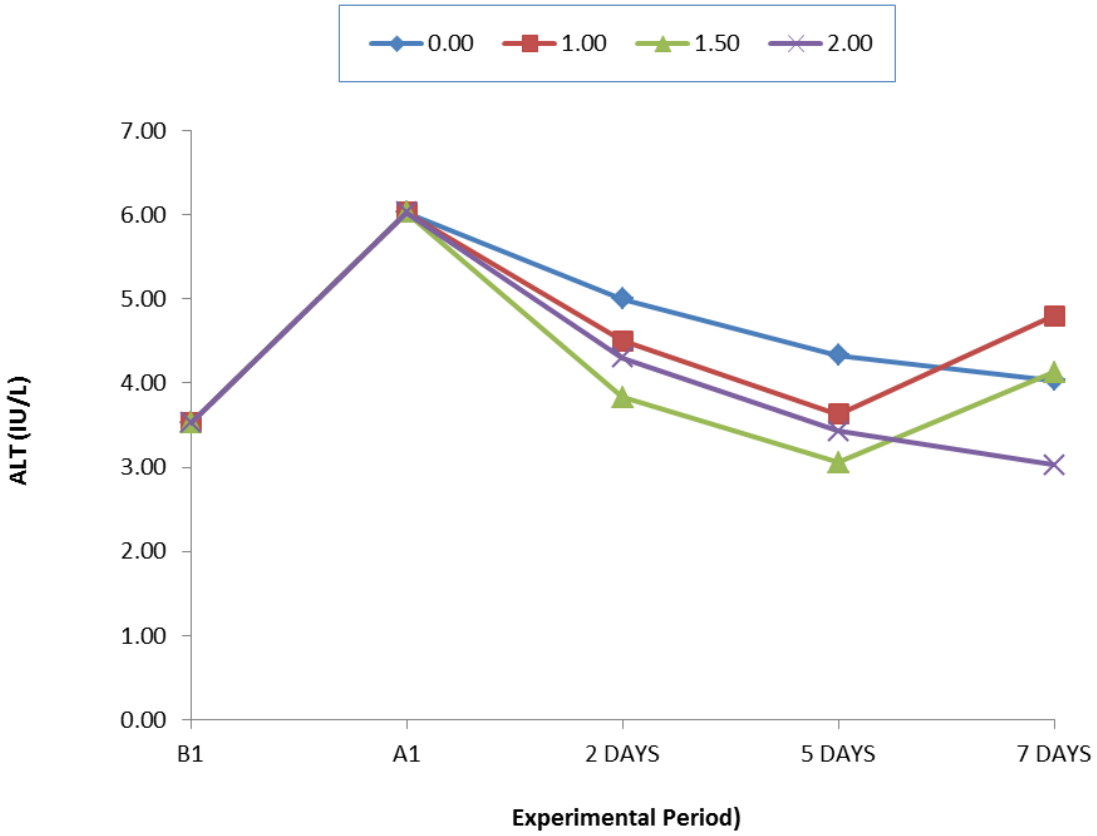


**Figure 4.2: Comparative Values of ALT in the Plasma of *C.gariepinus* Infected with *P.aeruginosa* and exposed to *P.americana* Extracts at different Experimental Period**

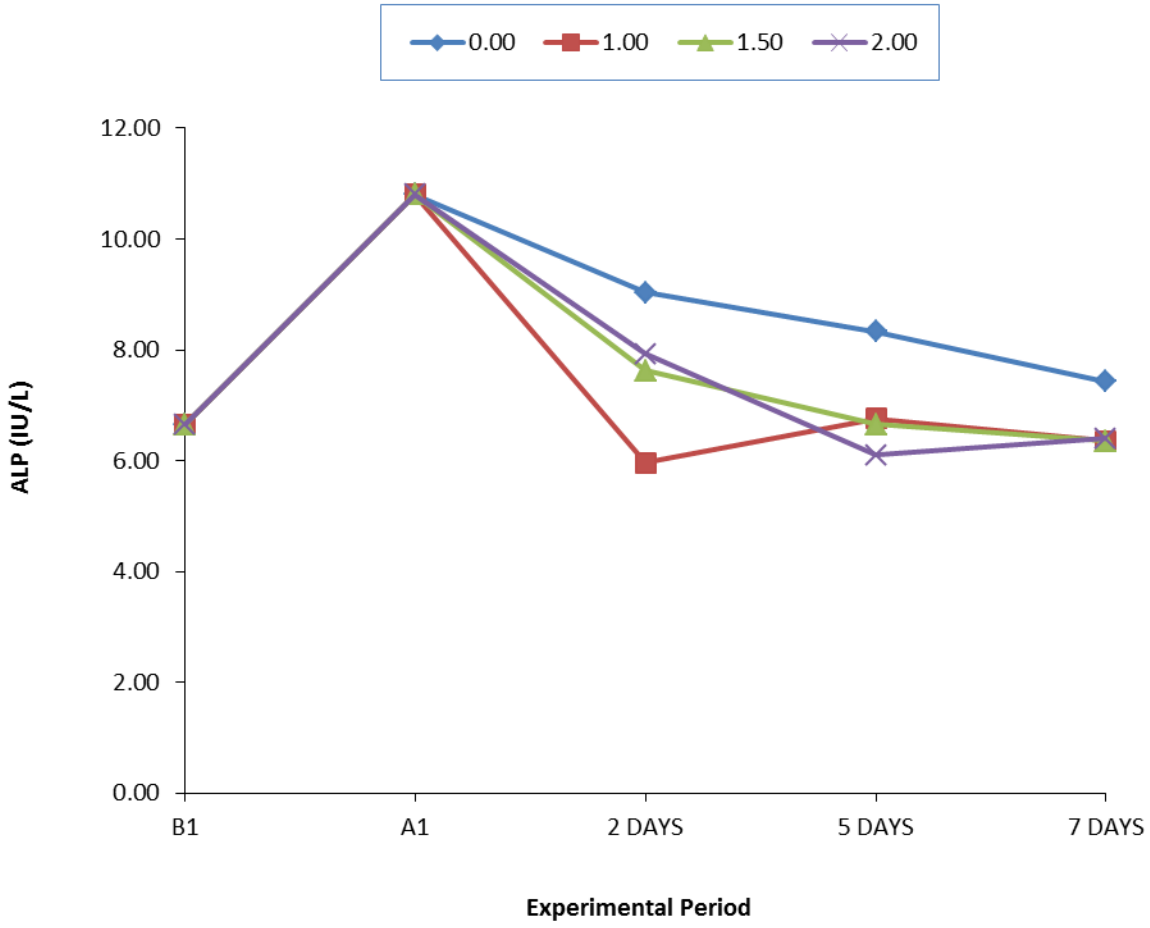


**Figure 4.3: Comparative Values of ALP in the Plasma of *C.gariepinus* Infected with *P.aeruginosa* and exposed to *P.americana* Extracts at different Experimental Period**

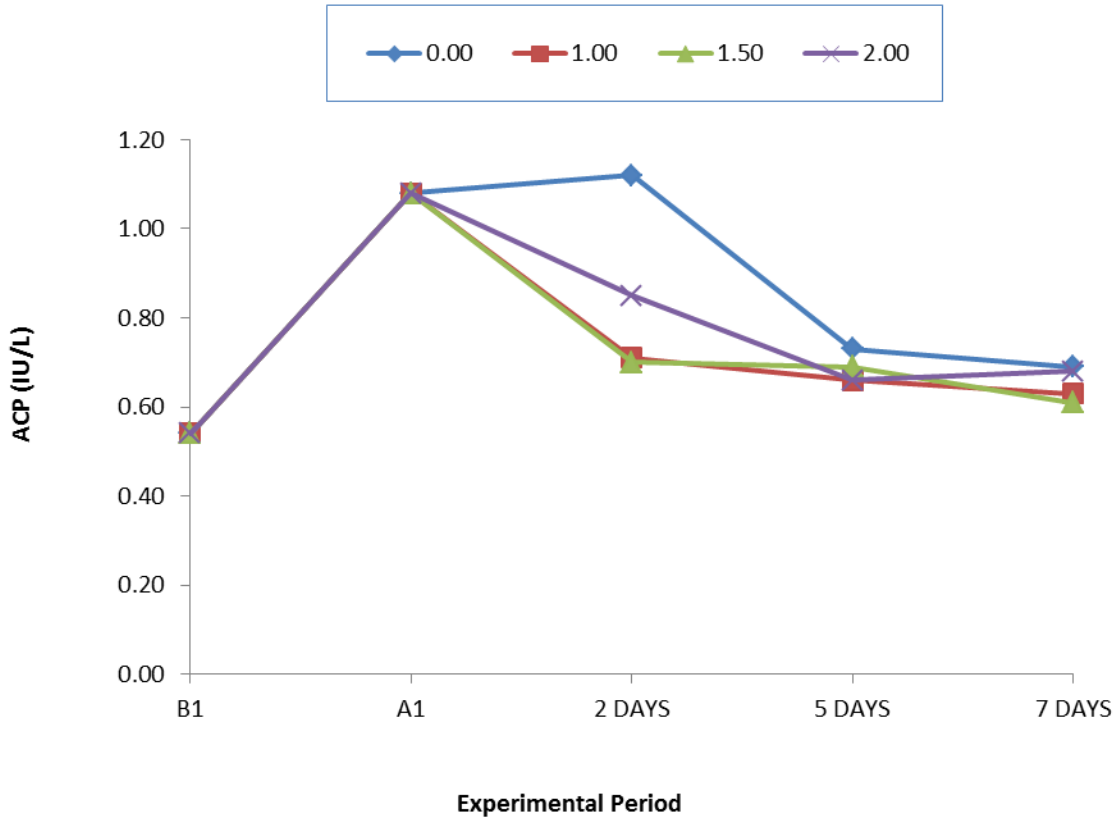


**Figure 4.4: Comparative Values of ACP in the Plasma of *C.gariepinus* Infected with *P.aeruginosa* and Exposed to *P.americana* Extracts at Different Experimental Period**

## 4 . DISCUSSION

### 4.1 Enzymatic activities of the experimental fish exposed to *P. americana* aqueous extracts

Biochemical parameters have been used by many researchers to evaluate the health of fish and it's reactions to pathogens, feeds, intoxicants and environmental stress (Shalaby *et al*., 2006; Ukwe and Oladapo – Akinfolarin, 2019). Changes in serum/plasma AST, ALT, and ACP are noted as the most sensitive biomarkers in the diagnosis of hepatic damage (Fadi et al., 2013; Ukwe and Oladapo-Akinfolarin, 2019) because they are released into circulation after cellular damage (Pari, 2005). They also indicate the health status of the tissue parenchyma and tissue necrosis which is consider as the major source of their increase in the blood of animals (Zaki *et al.,* 2007). Enzymes assays such as AST, ALT, ALP and ACP are parts of standard laboratory test to detect abnormalities in animals (Ayalagu *et al.,* 2001; Saka et al., 2011). Changes in these enzymes activities resulting from exposure to therapeutic application in plasma of fish have been reported (Ribas *et al*., 2007; Shalaby et al., 2006). In this study activities of these enzymes decreased as the concentration of *P. americana* increase in the infected fish, hence the inhibition appears to be dose dependent. This is in line with the report of Fadi *et al.*, (2013) who observed reduction in these enzymes when Nile tilapia fed *cyanobacteria* was infected with *A. hydrophila.* The transaminanse (AST and ALT) plays an important role in the utilization of amino acid for the oxidation activities and gluconeogenesis. They function as links between carbohydrate and proten metabolism under altered physiological, pathological and stress induce environment conditions (Daniel, 2009). The elevations in AST and ALT activities after the infection and throughout the period of exposure to 0.0ml (untreated) compared to the treated group (1.0 – 2.5m) could be due to threat on the hematopoetic organs such as liver and kidney (Ukwe and

Oladapo – Akinfolarin, 2019). This is in agreement with Khalil *et al*., (2011) who postulated that increased in AST and ALT levels in plasma was associated with hematopoietic organs damage when *Anguilla anguilla* was experimentally infected with *Vibrio anguillarum.* Furthermore, Ukwe and Oladopo - Akinfolarin (2019) also reported that the presence of *P.*aeruginosa and A. *hydrophila* increased enzymes activities such as AST, ALT, ATP and ACP in *C.gariepinus*. The enzyme phosphatases (ALP and ACP) are present practically in all tissues especially in the cell membrane, where active transport normally takes place and has hydrolases and transphospholyase functions. Gabriel *et al* (2015) reported increase in the values of AST and ALT in the control (unprotected) when *Oreochronuis miloticus* fed dietary *Aloe vera* was challenged with *streptococous iniae,* similar observation was reported by Saka *et al* (2011) when alcohol – induced rats were treated with *aloe vera.* Increase in AST and ALT is associated with biliary distruction (Hill, 2004). In line with the findings of this work, Zodepe (2010) also reported increase of ACP and ALP in the blood of the control fish when *Labeo rohita* induced with chromium was treated with *aloe vera* juice. The increased ALP and ACP in the infected fish, indicates alterations in the function of kidney and liver (Ukwe and Oladapo-Akinfolarin, 2019). In all the periods of the treatment/exposure, the activities of the enzymes in the treated groups (1.0ml, 1.5ml and 2ml of *P. americana*) were lower than the activities in the untreated group (0.0ml), this could be as a result of the presence of medicinal phytochemicals in the *P. americana* aqueous leave extracts (Ogundare and Oladejo, 2014) that may have reduced the pathogenicity of the *P.aeruginosa* by boosting the fish immune system (Sales et al., 2013) or thicken the cell membrane of the organs of the fish to stop further penetration/attack from the *P.aeruginosa* (Davi and Patrono, 2007). Several authors have reported the therapeutic effects of other medicinal herbs on fish hematopoietic organs; *Achyranthes aspera* in Indian major carp

infected with *A. hydrophila* (Vasudeva et al, 2006) and dietary *aloe vera* in O. noliticus infected with streptococcus iniae (Gabriel *et al,* 2015).

### 4.2 Organosomatic indices of the experimental fish exposed to *P. aeruginosa.*

Organosomatic indices are used to evaluate the health status of fish and other organisms (Ronald and Bruce, 1990). The analysed organosomatic indices HSI, SSI, VSI and IF were significantly higher in 0.0ml untreated group compare to the other treatments (1.0m – 2.0ml) that were significantly the same. This results is in agreement with the result of Gupta *et* al., (2016) who observed the enlargement of the liver (HSI) when *rattus rattus* was parasitized, and Ukwe and Jamabo (2020), who reported the medicinal importance of mango bark extract on the organosmatic indices of *C.gariepinus* infected with *P.aeruginosa.* This could be as a result of loss of glycogen due to restlessness encountered after infection (Bandsman et al 2008) which may have cause liver enlargement (Verman, 2017; Ukwe and Jamabo, 2020). The increase in the spleen and liver in the untreated fish could also be as a result of the presence of the pathogen instigating the forceful release of white blood cells (Kumar et al, 2011), this may stress the liver and the spleen which could lead to enlargement (Plumb and Hanson, 2011; Ukwe and Jamabo, 2020).

Though the implication of the intraperitoneal fat (IF) also known as visceral adipose fat (VAT) have not been fully studied in fish, Meza-Perez and Randall (2018) state that adipose fat eliminates pathogens and maintain immune homeostasis. The increase in the intraperitoneal fat in the infected and untreated fish could be the fish innate immune response against the pathogen *P.* aeruginosa (Jenab *et al,* 2021). Increase in intraperitoneal fat (IF) as seen in the infected and untreated fish could also be an indication that muscle fiber in the heart (left ventricular) is getting stressed and may lead to the heart getting stressed disorders (Chughtai *et al,* 2011) and Coronary

artery disease (Nemes *et al,* 2007), and this can lead to sudden death of the fish. The increase in the visieral could be as a result of the increase associated with the organs of the fish as a result of the presence of the pathogen, since organs like liver, spleen, and interperitoneal fat have been increased due to the presence of the pathogen (*P. aeruginosa).* The presence of the medical phytochemicals in the *P.americana* could be the reasons why there was no increase in the organs of the fish in the treated groups (1.0ml, 1.5ml and 2.0ml) (Ogundare and Oladejo, 2014). Some of the phytochemicals present in avocdo pear leave extracts includes alkaloids, tannias, saparines etc. (Reverter et al, 2014; Citarasu, 2010). These phytochemicals are bactericidal, and have improve the phagocytic activity of the experimental fish exposed to the various concentration of the experimental herb (Ardo *et al*, 2008; Sivaram *et al*, 2008), as a result the organs of the fish infected with the experimental pathogen and expose to the various concentrations of the *P. americana* aqueous extracts were protected from inflammation.

## 5. CONCLUSION AND RECOMMENDATIONS

Though the infected fish had reduced enzymatic activities even in the non-treated groups at the end of day two, five and seven of the treatment, the rate of reduction was higher in the infected fish exposed to the various levels of aqueous *P. americana* leaves extracts especially with the 1.5ml/L concentrations within the days of treatment. This is an indication that the *P. americana* aqueous leaves extracts possesses some therapeutic phytochemcials that has the capacity to restore enzymatic distortions in fish that are diseased as a result of pathogenic attack. The result of the organosomatic indices show that the *P. americana* aqueous leaves extracts is therapeutic against fish organ inflammation especially diseased fish, and it is also a strong antibacterial agent against *P. aeruginosa.* The result of this research has also shown that enlargement in fish organs

such as the liver, spleen, intraperitoneal fat etc, can be attributed to the presence of bacterial pathogens such as *P. aeregunosa*

Avocado pear leaf is cheap readily available all year round, eco-friendly and non-poisonous to both fish and the fish consumer. It is therefore recommended that avocado pear trees be planted within and around our fish farms, and be used at will for the disease treatment in aquaculture.

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