



Effect of Smoked and Oven-dried Catfish (*Clarias gariepinus*) on Haematological Parameters, Liver and Antioxidant Enzymes of Wistar Rats

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Catfish is a traditional part of the diet of a large section of the world's population. This study compared the effect of smoked and oven-dried catfish on hematological parameters, liver and antioxidant enzymes of wistar rats. Catfish samples were processed by smoking and oven-drying and used for formulation of the experimental feeds. Twenty one wistar rats were acclimatized for seven (7) days, weighed and allotted into three dietary treatments; control (standard feed), smoked and oven-dried fed groups. The study was conducted for a period of 21 days. Haematological analysis was carried out using haematology auto-analyzer. Liver enzymes (Alanine aminotransferase (ALT), Aspartate aminotransferase (AST), alkaline phosphatase (ALP) and Gamma-glutamyltransferase (GGT)) were assayed using standard assay kits while antioxidant enzymes were assayed using spectrophotometric method. The result revealed a significant increase ($P < 0.05$) in the body weights of rats maintained on experimental feeds; oven-dried catfish fed group ($240.83 \pm 6.13g$), smoked catfish fed group ($246.83 \pm 4.97g$). There were no significant difference ($P > 0.05$) in the haematological parameters of the treatment groups except in their total red blood cell counts, mean cell volume and mean cell hemoglobin. A non-significant difference ($p > 0.05$) was observed in the antioxidant enzymes (SOD and CAT) of the test groups, indicating

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the absence of oxidative stress. The results of this research showed that both drying methods (oven and smoke drying) did not affect the palatability of the diets as the experimental diets were accepted by the experimental animals and their weight significantly improved. However, both diets have deleterious effects on the blood; hence, individuals with severe cases of anaemia and other blood disorders are encouraged to avoid them.

Keywords: Catfish; Haematology; antioxidant; anaemia.

1. INTRODUCTION

Fish is a highly nutritious food and it is particularly valued for its protein which is of high quality compared to meat and egg [1]. It contains high quality protein, amino acids and absorbable dietary minerals [2]. Fish contributes to the world protein and it is being used as a good tool for food therapy and source of therapeutic substances for the treatment of various diseases such as coronary diseases, auto-immune diseases and protein energy malnutrition [3]. According to Olayemiet al. [4], fish contains 16.24% protein and 0.50% fat [5]. The major constituents of fish are moisture, protein and fat with minerals occurring in trace amount. Generally, fish contains very little carbohydrate while the moisture content is very high. In most fish species the moisture content is between 60-80 % [6]. The fat content of fishes varies with species, age, size and also season [6]. It also contains some bioactive compounds with therapeutic properties that are beneficial to human health [3], with vast increase in its consumption among Nigerians.

However, it is highly perishable because it provides a favorable medium for the growth of macro and microorganisms after death [1]. Fish spoilage is a metabolic process that makes it to be undesirable for human consumption due to changes in its sensory and nutritional characteristics, therefore, it has become increasingly important to ensure that they are fully and efficiently utilized to avoid deterioration.

Since fish is not normally consumed raw, various processing methods are employed in preparing them for consumption [6]. These processing methods are also employed to extend its shelf life.

Thus, the processing and preservation of fresh fish becomes imperative in order to maintain product quality, reduce wastage and prevent economic losses [7]. To prolong the shelf life of fish, it is preserved by many processes including oven drying, solar drying, canning and smoking among others [6]. Some of these processes,

though important for preservation, have various effects on the physical and nutritional quality of fish because it has been observed that different processing and drying methods have different effects on the nutritional compositions of fish [2].

African catfish (*Clarias gariepinus*) is highly relished and considered to be the most farmed tropical catfish species in Nigeria and other African countries [8]. *C. gariepinus* is a good aquaculture candidate due to its hardy nature, high tolerance to poor water quality conditions, easy adaptation in captivity and high growth rate [9]. The need to compare the effect of smoking and oven-drying methods on the haematological parameters, and its continuing effect on liver and antioxidant enzymes in Wistar rat is imperative.

2. MATERIALS AND METHODS

2.1 Sample Collection

A total of fifty (50) adult *C. gariepinus* of about 550 g each were procured from a private fish farm in Awka, Anambra State capital and transported to the Department of Fisheries and Aquaculture Laboratory, Nnamdi Azikiwe University, Awka, Anambra State in 250 L capacity plastic container containing aerated water. Fish were conditioned at the Departmental fish pond till they were used for the experiment.

2.2 Preparation of Sample

Fish samples were washed with distilled water and divided into two groups. One group was smoked with firewood. The fish was cut into pieces and spread out on smoking trays. The trays were stacked on smoking oven fired with firewood and left for 15 hours to obtain a dry-smoked product. It was then homogenized to powder and used for feed formulation while the other group was oven-dried at initial temperature 70°C and was increased to 105°C after 3 hours for another 12 hours [10].

2.3 Experimental Animals

Twenty one albino rats weighing between 100 – 140 g were purchased from Chris farm,

Mgbakwu, Anambra state and used for this study. The animals were housed in standard rat's cages in the animal facility of Applied Biochemistry Department, Awka, Anambra state. They were allowed to acclimatize for 7 days and fed with standardized rat pellets and water *ad libitum*.

2.4 Animal Grouping and Treatment

The rats were randomly distributed into three groups consisting seven rats in each group. The rats in Group 1 served as the control group and were fed 100% standard rat pellet while Group 2 and Group 3 served as treatment groups and were fed with formulated diet as shown in the table below:

2.5 Sacrifice and Blood Collection

The animals were fed for 21 days and sacrificed after a 24-hour fast. They were anaesthetized using chloroform soaked in cotton wool and blood collected by heart puncture into coagulated and non-coagulated universal blood sample tubes.

2.6 Haematological Analysis

Haematological parameters include packed cell volume (PCV), haemoglobin concentration (Hb), red blood cells count (RBC), MCV, MCH, MCHC, white blood cells (WBC) and its differentials and Platelet count were analysed using haematology autoanalyser (BC 6800 Mindray, Canada).

2.7 Assay of Liver Enzymes

The serum enzymes assay was carried out using serum and assayed with a Mindray kit (China).

2.8 Assay of Antioxidant Enzymes

2.8.1 Assay of superoxide dismutase activity

The activity of superoxide dismutase was carried out on the serum using the method of Sun and Zigma [10].

2.8.2 Assay of catalase activity

The activity of superoxide dismutase was carried out on the serum using the method of Beers and Seizer [11].

2.9 Data Analysis

Results of the study were expressed as mean \pm standard error of mean. Differences between mean of the treated groups and their control in this animal studies were analyzed using Analysis of variance (ANOVA) followed by Tukey's post-hoc tests of SPSS 16.0 spreadsheet statistical package. Values were taken to be significant at ($P < 0.05$).

3. RESULTS AND DISCUSSION

Fig. 1 showed the effect of smoked and oven-dried catfish on the body weight of Wistar rats. There was a significant increase ($P < 0.05$) in weight of the smoked catfish group after 7 days. A significant increase ($P < 0.05$) in weight was also observed in the oven-dried catfish group after 7 days. This significant increase in weight was also observed after 14 days and 21 days in both treatment groups. However, there was no significant difference ($P > 0.05$) in the weights of the control group after 7, 14 and 21 days. There was also no significant difference ($P > 0.05$) between the treatment groups after 7, 14 and 21 days.

The result for the change in weight of the various rat groups in percentage is presented in Fig. 2. The control group had a 5.81% increase in body weight after 7 days which decreased to 0.81% after 21 days. A 61.62% increase in body weight was observed in the smoked catfish group after 7 days but decreased to 3.64% after 21 days. The oven-dried catfish group had a 53.17% increase in body weight after 7 days but decreased to 5.47% after 21 days.

The results of the effects of the formulated feeds on haematological parameters are presented in Figs. 3 and 4. The results showed that the Hb, PCV and Neutrophils were increased in the treatment groups but statistically not significant ($P > 0.05$) when compared to the control group. Increase in hemoglobin concentration may be as a direct consequence of the high content of iron in smoked catfish that may stimulate synthesis of hemoglobin. Iron is important for the production of hemoglobin. Sayadet al. [12] reported that smoked catfish is a good source of lean meat and trace elements especially iron and zinc.

There was, however, a statistically significant decrease ($p < 0.05$) in TRBC in the treatment groups with the significant difference occurring when the smoked catfish group was compared

with the control group and also when the oven-dried catfish group was compared with the control group. The significant reduction ($p < 0.05$) in the RBC count in the treatment groups suggests an indication of severe anaemia caused by constituents of the smoke and oven-drying. The experimental animals gained weight at the same time probably because of underactive thyroid gland. The anaemic response could be as a result of the destruction or inhibition of erythrocyte production [13] or haemodilution as reported by Sampathet al [14].

A significant increase ($P < 0.05$) was observed in the MCH level of the smoked group when compared to the control group. A significant increase was also observed when the oven-dried catfish group was compared to the control group. The most common reason for high MCH is macrocytic anemia [15] which is a blood disorder

in which the body fails to produce enough red blood cells. In macrocytic anemia, red blood cells that are produced are larger than usual, each carrying more hemoglobin than normal-sized cells would. This condition can be caused by deficient levels of vitamin B-12 or folic acid in the body; nutrients found in foods like fish, liver, green leafy vegetables and fortified cereals.

There was a reduction in the platelet levels of the test groups, though not significant ($P > 0.05$). The control group had the highest platelet level while the oven-dried group had the least. Thrombocytopenia is a condition in which one has a low blood platelet count. This agrees with Akoh and Hearnberger [16], who reported that the consumption of processed catfish diet caused a decrease in platelet count and prolonged blood clotting time.

Table 1. Animal grouping and feed composition

Group	Feed Composition
1	100% standard feed
2	50% standard feed + 50% smoked catfish
3	50% standard feed + 50% oven-dried catfish

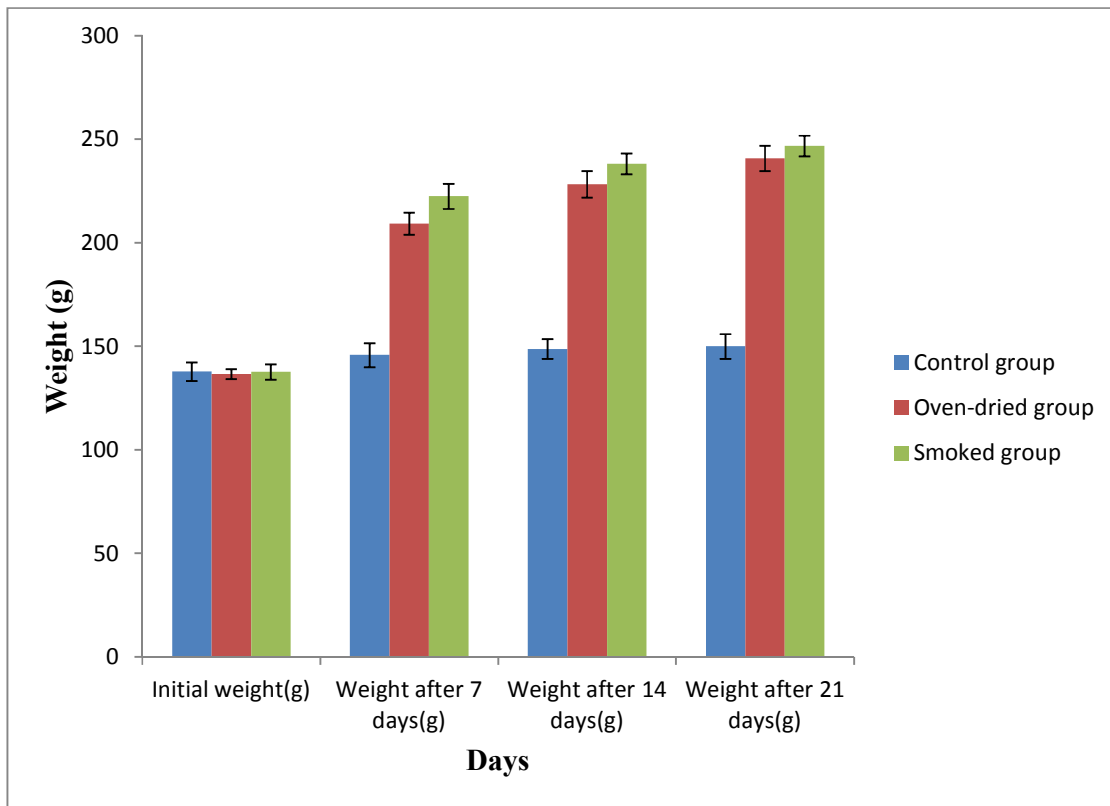


Fig. 1. Effect of smoked and oven-dried catfish on body weight of wistar rats

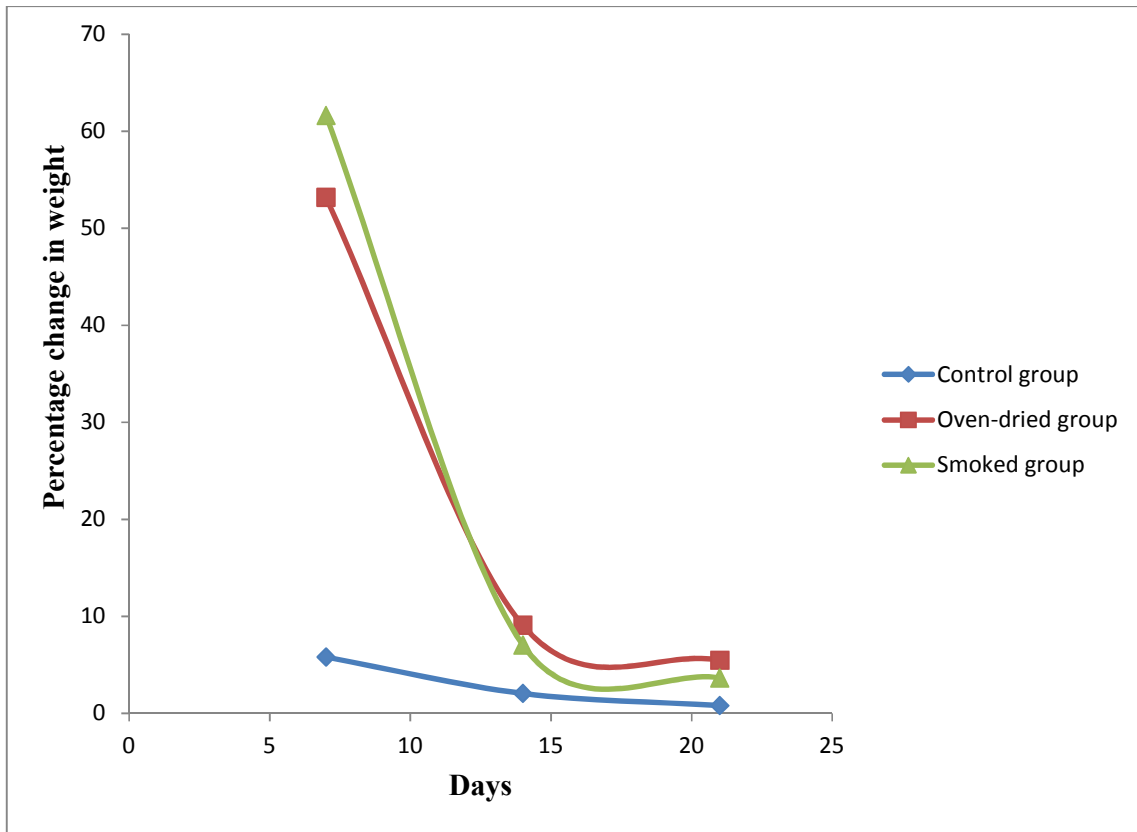


Fig. 2. Percentage change in weight of the various rat groups

There was a significant increase ($P < 0.05$) in the MCV levels of the test groups with the significant difference occurring when the control group was compared with the smoked catfish group and also when the control group was compared with the oven-dried catfish group.

Mean corpuscular volume (MCV) is the average volume of a red blood cell. This is a calculated value derived from the hematocrit and red cell count. An increase in MCV value may be due to swelling of RBC and/or disturbance of osmoregulation and reduction in erythrocytes. Increase in MCV values seems to be correlated with decline in RBC count [17].

The Effects of the formulated feeds on liver enzymes is presented in Fig. 5. There was a statistically significant increase ($P < 0.05$) in the AST level of the smoked fish group compared to the control group. No significant change was observed in the oven-dried group. Smoked catfish group had the highest value while the control group had the least. The AST level also

increased in the oven-dried group, though not significant. Increases in transaminases are markers of hepatocellular damage and leakage of enzymes through damaged cell membranes [18]. It indicates that the hepatic intracellular enzymes have leaked into circulation [19]. The significant increase observed when the group fed the smoked fish diet was compared with the control indicates hepatic damage resulting from the toxicants which could probably be present in the smoked fish diet.

There was a decrease in the ALT level in the treatment groups, though not statistically significant ($P > 0.05$). Alanine aminotransferase (ALT) is the most widely used clinical biomarker of hepatic health [20]. Damaged hepatocytes leak the ALT into the extracellular space and ultimately plasma, so that ALT activity and/or quantity will be increased in animals with damaged hepatocytes when compared with normal. The decrease in ALT concentration in the serum of the test rats may be an indication that there may not be liver damage.

The difference in the ALP level of the various groups were not statistically significant ($P>0.05$).

The smoked catfish group had the highest value while the oven-dried catfish group had the least.

There was an increase in the GGT level in the treatment groups, though not statistically significant ($P>0.05$). Gamma-glutamyltransferase (GGT) is a relatively non-specific enzyme whose activity can increase with hepatic disease, cardiac injury and muscular injury [21]. GGT is a specific biomarker of hepatobiliary injury, especially cholestasis and biliary effects. Increased serum activities of gamma-glutamyltranspeptidase (γ GT), alkaline phosphatase, total bile acids, 5'-nucleotidase (5'NT) and bilirubin have been associated with

cholestatic disease in rats. Observations support the use of serum GGT in the rat as diagnostic of bile duct necrosis when increases are detected.

Antioxidant enzymes are proteins involved in the catalytic transformation of reactive oxygen species and their by-products into stable nontoxic molecules therefore representing the most important defense mechanism against oxidative stress-induced cell damage [22]. A non-significant increase ($p>0.05$) in superoxide dismutase (SOD) and catalase (CAT) activities were noted in the test groups (Fig. 6). Increased SOD and CAT activity is a mechanism among antioxidant enzymes in response to increased oxidative stress. SOD and CAT combines to protect cells from superoxides and hydrogen peroxide [19].

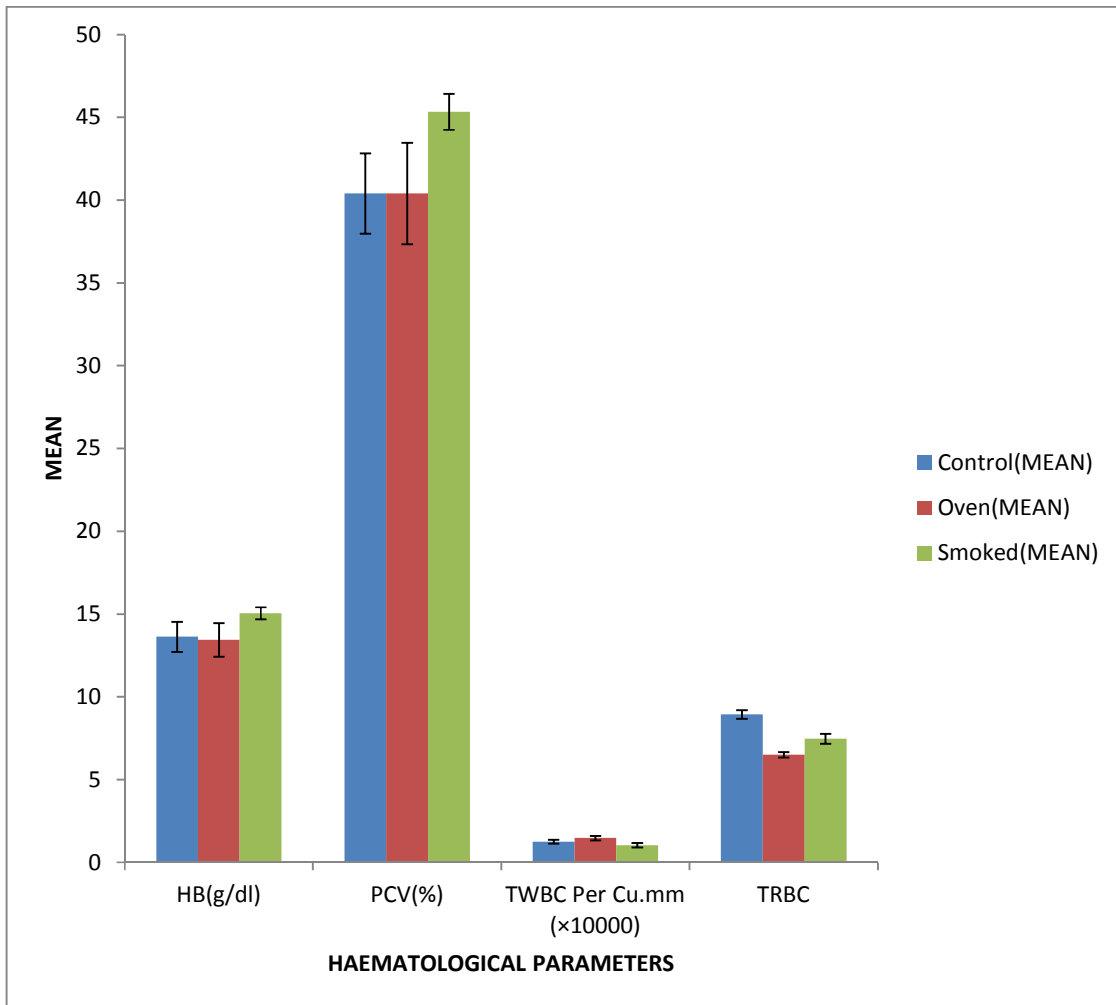


Fig. 3. Effects of the formulated feeds on haematological parameters

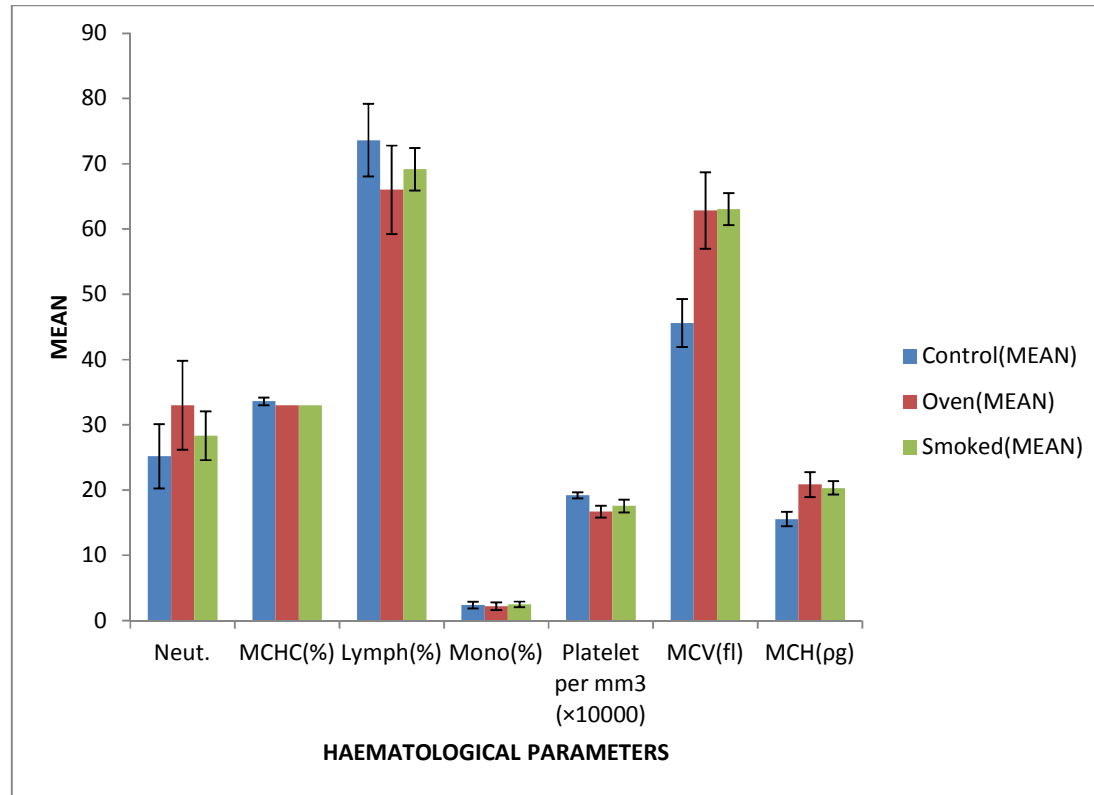


Fig. 4. Effects of the formulated feeds on haematological parameters

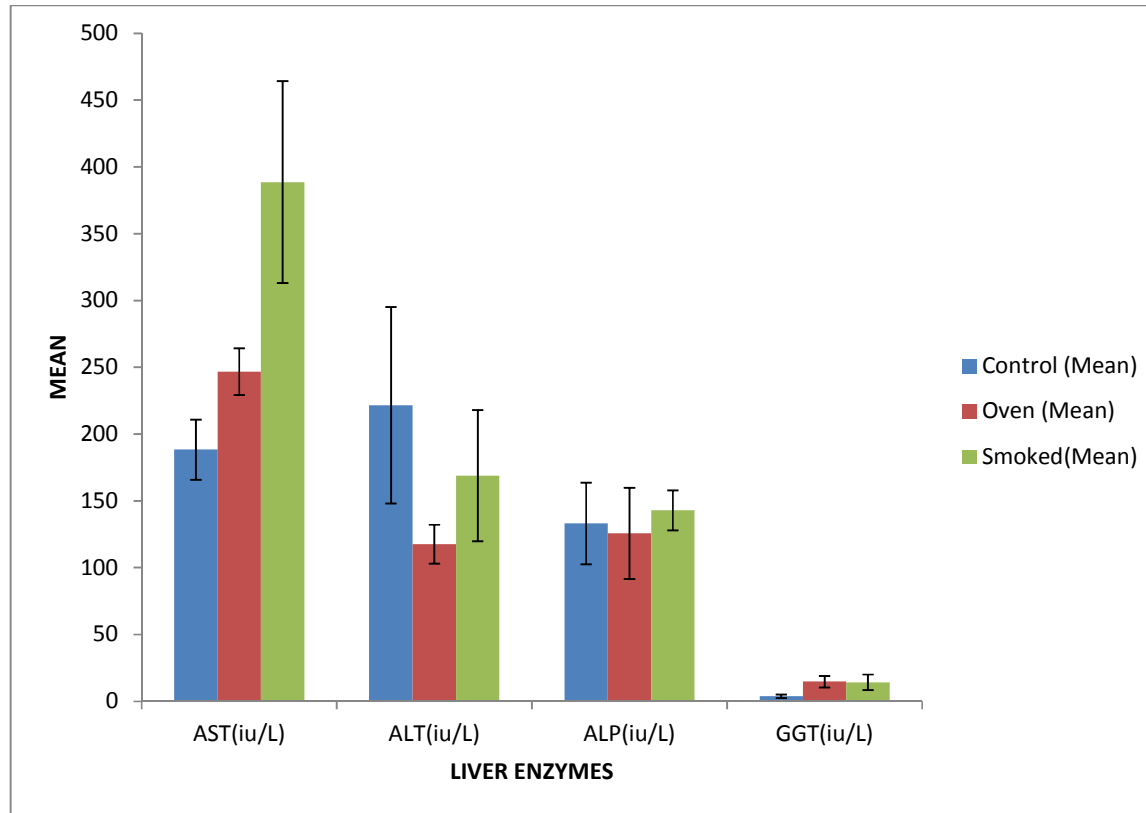


Fig. 5. Effects of the formulated feeds on serum enzymes

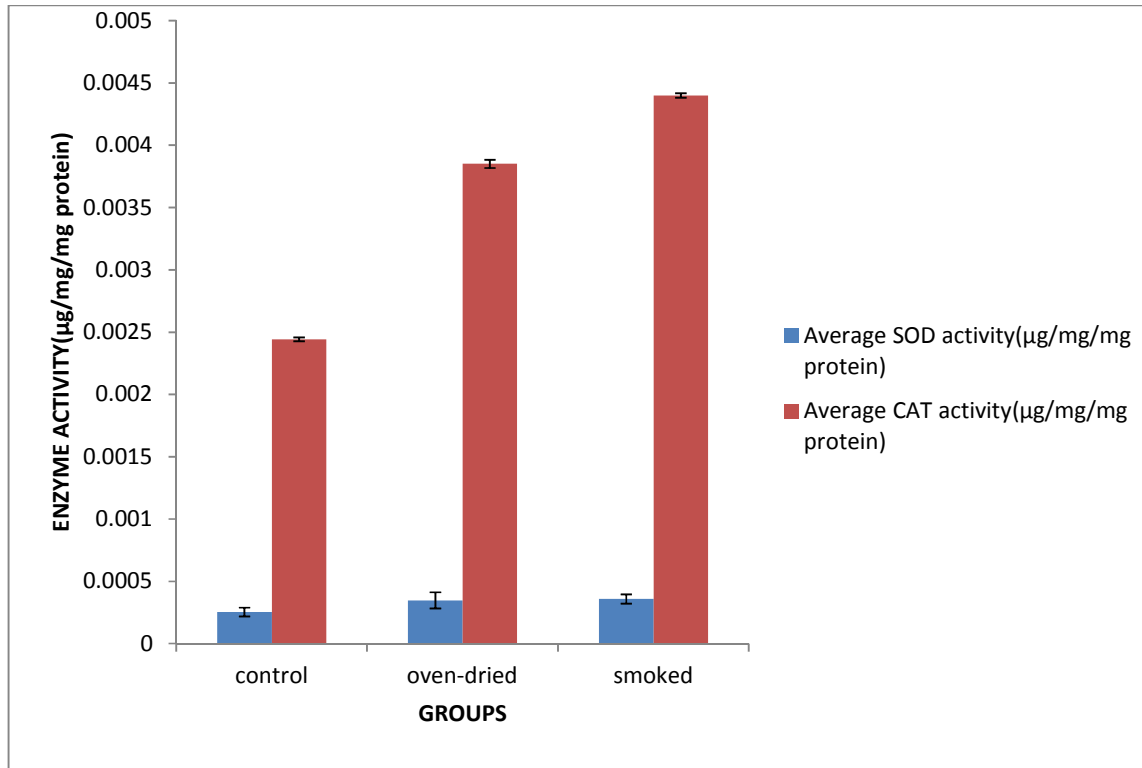


Fig. 6. Effects of the formulated feeds on anti-oxidants enzymes

4. CONCLUSION

The results of this research provide the comparative knowledge of the effect of commonly used drying methods (oven and smoke drying) on haematological parameters, liver and antioxidant enzymes of Wistar rats fed with African Catfish feed additive. Results noted that both drying methods (oven and smoke drying) did not affect the palatability of the diets as the experimental diets were accepted by the experimental animals and their weight significantly improved. However, both diets have deleterious effects on the blood as can be seen from the haematological parameters. Individuals with severe cases of anaemia and other blood disorders should therefore, avoid both diets. Elevated transaminase activities, as observed from this study, are considered as an index marker of hepatotoxicity, linked to oxidant stress. Both processing methods are good in extending the shelf-life of catfish.

ETHICAL APPROVAL

As per international standard or university standard written ethical approval has been collected and preserved by the authors.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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