



## Assessment of the Effects of Fresh Palm Oil, Natron (*Kanwa*) and Natron-Treated Palm Oil on Some Hematological Parameters in Wistar Rats

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### ABSTRACT

The effect of fresh palm oil, natron (*kanwa*) and natron-treated palm oil on some hematological parameters were assessed in wistar rats. Twenty- four wistar rats of different sexes weighing between 123-217g were divided into four groups of six (6) rats each. Group A served as a normal control and received normal saline and feed as placebo. Groups B, C and D received 5ml/kg body weight (BW) of fresh palm oil, 5ml/kg bw of 3% natron solution only and 5ml/kg of 3% natron-treated palm oil respectively. They were provided with commercial rats' chow and water ad libitum. Their body weights and some hematological parameters (RBC, WBC, HB, PCV and PLATELETS) were assessed prior to the commencement of the experiment and weekly for three weeks (21 days). Result of the study shows that the control group ( $6.03 \pm 0.17$ )  $\times 10^{12}/L$  was significantly ( $P < 0.05$ ) higher than all the groups B ( $5.5 \pm 0.17$ )  $\times 10^{12}/L$ , C ( $4.93 \pm 0.69$ )  $\times 10^{12}/L$  and D ( $4.37 \pm 0.28$ )  $\times 10^{12}/L$  respectively. The packed cell volume (PCV) of group A ( $43.5 \pm 0.76$ ) % was significantly ( $P < 0.05$ ) increased more than group B ( $31.0 \pm 1.46$ )%, C ( $30.83 \pm 0.43$ )% and D ( $28.0 \pm 1.07$ )% respectively. Also, the hemoglobin (HB) concentration of the control group A ( $14.46 \pm 0.28$ ) g/dl was significantly ( $P < 0.05$ ) higher group D ( $11.5 \pm 0.25$ ), B ( $11.15 \pm 0.45$ ) and group C ( $10.36 \pm 0.45$ ) g/dl respectively. The white blood cell (WBC) of the control group A ( $4.38 \pm 0.26$ ) was significantly ( $P > 0.05$ ) lower than all the groups and there was no significant difference between the control group A and other groups. The group that received 5ml/kg body weight of 3% natron only had the highest mortality rate before the termination of the experiment than other groups and increased body weights more than other groups while group B had no mortality and had the lowest body weights. Therefore, the result of this study shows that chronic consumption of natron (*kanwa*) in our diets can result in increase in body weight, coronary heart failure due to atherosclerosis caused as a result of plaque deposition and should be discouraged. Therefore, the study suggests that the adverse effects of this diet be properly studied in other physiological indices in human models.

**Keywords:** Natron (*kanwa*), fresh palm oil, natron-treated palm oil, blood parameters, body weights.

## INTRODUCTION

Natron (kanwa), sesquicarbonate or hydrated carbonate of sodium is generally referred to as "kanwa" in the northern part of Nigeria and "akanwu" by the Igbos of the south-eastern part of Nigeria. It is the second most commonly consumed salt in Nigeria, hence the next important table salt<sup>[1]</sup>. It mostly occurs in the northwestern part of the country and erroneously called "potash" although it contains very low amounts of potassium compared to sodium<sup>[1]</sup>. This syndicates that the chemistry is misunderstood by those describing it as "potash". The effect of kanwa in cooking time was determined by<sup>[2]</sup> as it's commonly used by many Nigerians in the rural/urban areas for cooking legumes including the preparation of tapioca. It was observed that it increases the green color and texture of vegetables as well as reduces the cooking time of many legumes. When used in the cooking of cowpea which is noted for its prolonged cooking time of 45-65 minutes was reduced to 10-15 minutes when cooked with high concentration of kanwa (0.1-0.5%). It was observed by<sup>[3]</sup> that kanwa can be administered as medicine for all sorts of ailments. As medicine in its grounded form. It is mixed with tobacco and used as snuff. In northern Nigeria, it is also administered in large doses by the Hausa majority in the form of porridge of guinea corn and millet in what is popularly called "kunun kanwa". This is administered to their women immediately after birth as medicine to increase the quality and quantity of breast milk<sup>[4]</sup>. Natron has several uses. It is used in cooking as food tenderizer especially in pulses<sup>[5,2]</sup> to curdle milk, tanning industry and in the preparation and enhancement of flavor of local beverages and snuff<sup>[3]</sup>. It is used extensively in ethno-veterinary practices for the treatment of skin diseases and digestive problems. It also serves as salt lick and mineral supplements in ruminants<sup>[6]</sup> and used in decoction for the treatment of reproductive ailments such as retained placenta and difficulty in urination. It's uncontrolled use in livestock may lead to abortion in pregnant animals<sup>[7]</sup>. Ethnomedicinal potency

has been documented for ailments such as stomach ache, constipation and tooth ache<sup>[3]</sup>. This historical use of natron as an aid in child birth has been documented. In Kanuri tribe of Borno State in Nigeria, it is used to increase uterine contractility and motility for safer child birth<sup>[8]</sup>. In the Hausa- Fulani tribe of northern Nigeria, it is used in post natal care of puerperium as nursing mothers consume large quantities of natron (About 40g equivalent of 450mg of Na) daily in pap of guinea corn as part of forty day post partum practice in the believe that it increases the quantity of breast milk<sup>[9]</sup>. Consequently, this has been implicated in the incidence of peripartum cardiac failure (PPCF) among nursing mothers of this region<sup>[4]</sup>. Traditionally, it is used to treat various ailments related to the endocrine and reproductive systems such as treatment of painful uterus, including uterine contraction and abortions, management of retained placenta and post partum bleeding, infection and, infertility, colic pains and treatment of irregular and painful menstruation. One of its folkloric claims is its usage in abortifacient manda, which has to be substantiated scientifically, Therefore In spite of all the effects of kanwa stated above, there is paucity of information on the effects of kanwa and kanwa treated palm oil on hematological parameters and this suffixed our study as the diet is mostly consumed by the south-eastern part of Nigeria as a local ingredient in the preparation of tapioca diet.

## MATERIALS AND METHODS

### PLANT COLLECTION AND IDENTIFICATION

The natron (kanwa) and fresh palm oil were purchased from Railway Market in Makurdi North LGA of Benue State and were identified and authenticated at the Department of Chemistry, Faculty of Science, Benue State University Makurdi. Ethical Clearance for this study was also obtained from Animal Research Ethical Clearance Committee of the University of Nigeria Teaching Hospital Ituku – Ozalla, Enugu State.

### EXPERIMENTAL ANIMALS

Twenty- four wistar rats of different sexes each weighing between 123-217g were used in this study, The animals were procured from the Animal House Unit of the College of Health Sciences, Benue State University Nigeria. They were allowed to acclimatize for seven days in clean plastic cages in a fly-proof house with 12hr day/light cycle and were fed with commercial rat's chow (Top feeds Nigeria Ltd Jos) and were provided with clean drinking water *ad libitum* throughout the experimental period which lasted for three weeks (21 days).

### PREPARATION OF THE 3% NATRON SOLUTION

A lump of natron (kanwa) was grounded into fine powder with mortar and pestle after which it was sieved and solid particles removed. 6g of the fine powdered natron (kanwa) was weighed using an electronic weighing balance (Ohaus Instruments USA) and was divided into two portions of 3g each. The first portion (3g) was dissolved in approximately 97ml of distilled water as vehicle making the 3% (<sup>w</sup>/v) natron solution. The second (3g) portion also dissolved in approximately 97ml of fresh palm oil to obtain the 3% (<sup>v</sup>/v) solution respectively. They were kept in a warm room at a temperature of about 27<sup>0</sup>C till they were ready for use.

### EXPERIMENTAL DESIGN

They experimental animals were divided into four groups A, B, C and D of same sex in a group. Group A received 10ml/kg normal saline daily as placebo. Group B received 5ml/kg body weight of fresh palm oil daily, Group C and D received 5ml/kg of 3% natron solution and 5ml/kg natron-treated palm oil daily via oral route of administration for twenty one days.

### MEASUREMENT OF BODY WEIGHTS

The weekly body weights of the experimental animals were determined before and weekly during the experimental period using a digital top loading weighing balance by Harvard apparatus Company Ltd USA). Thus, transparent cylindrical glass restrainer was weighed and the weight tarred

to zero mark before introducing the animals individually and their weights recorded in gram.

### METHOD OF BLOOD COLLECTION AND ANALYSIS OF THE HEMATOLOGICAL PARAMETERS.

About 1.5 milliliter of blood were collected weekly through a cardiac puncture and transferred into EDTA anticoagulant bottles. The samples of blood were analyzed in the hematology laboratory by the use of Automated hematology autoanalyzer-sysmex-2IN. All the samples were analyzed within one hour of blood collection and the results recorded.

### METHOD OF DATA ANALYSIS

The data were presented as Mean  $\pm$  Standard Error of Mean and were analyzed using one –way Analysis of variance (one-way ANOVA) to compare their means using computer software (SPSS version 21.0 and Excel for windows.  $P \leq 0.05$  was considered to be statistically significant.

### RESULTS AND DISCUSSION

This research sought to assess the effect of fresh palm oil, natron (kanwa) and natron-treated palm oil on some hematological parameters in wistar rats. Fig i. This figure shows the result of mean  $\pm$  standard error of mean of the weekly body weights of the animals. In this figure, it was observed that group A (control group) had a steady increase in the body weights from the onset of the experiment to the final week. The increase in the body weight was more pronounced in the groups that received 5ml/kg fresh palm oil with different of  $39.8 \pm 7.47$ g between the baseline body weights and the week three (day21) and this was followed by group D ( $32.9 \pm 6.02$ )g, C ( $27.3 \pm 1.63$ )g and group A ( $13.6 \pm 6.5$ )g respectively. The body weights of the respective groups were compared statistically with the control group A and found that there was a significant difference ( $P < 0.05$ ) between the groups compared with group A. In this study, the increase in their body weights could be an indication of the increased extracellular fluid volume and this is in agreement with the work of Soladoye and Oyeleke <sup>[15]</sup>

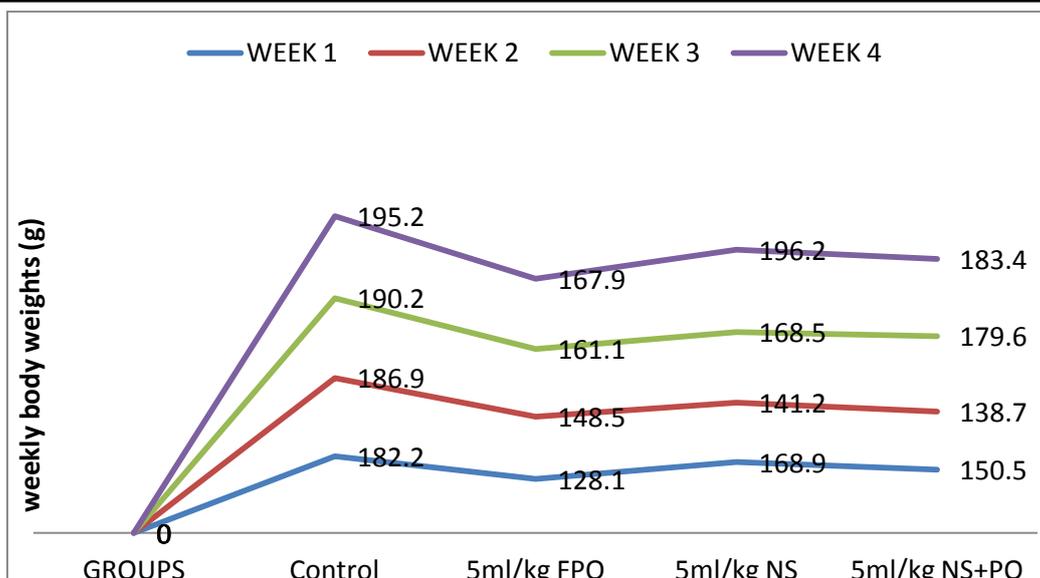


Fig i. Result of the Mean ± SEM of the weekly body weights of the animals

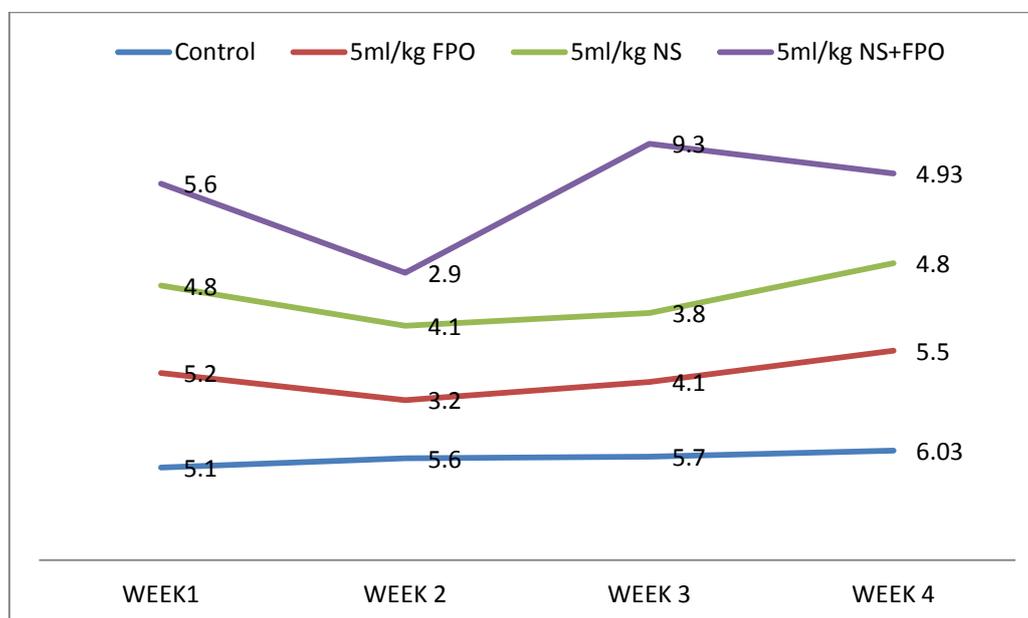


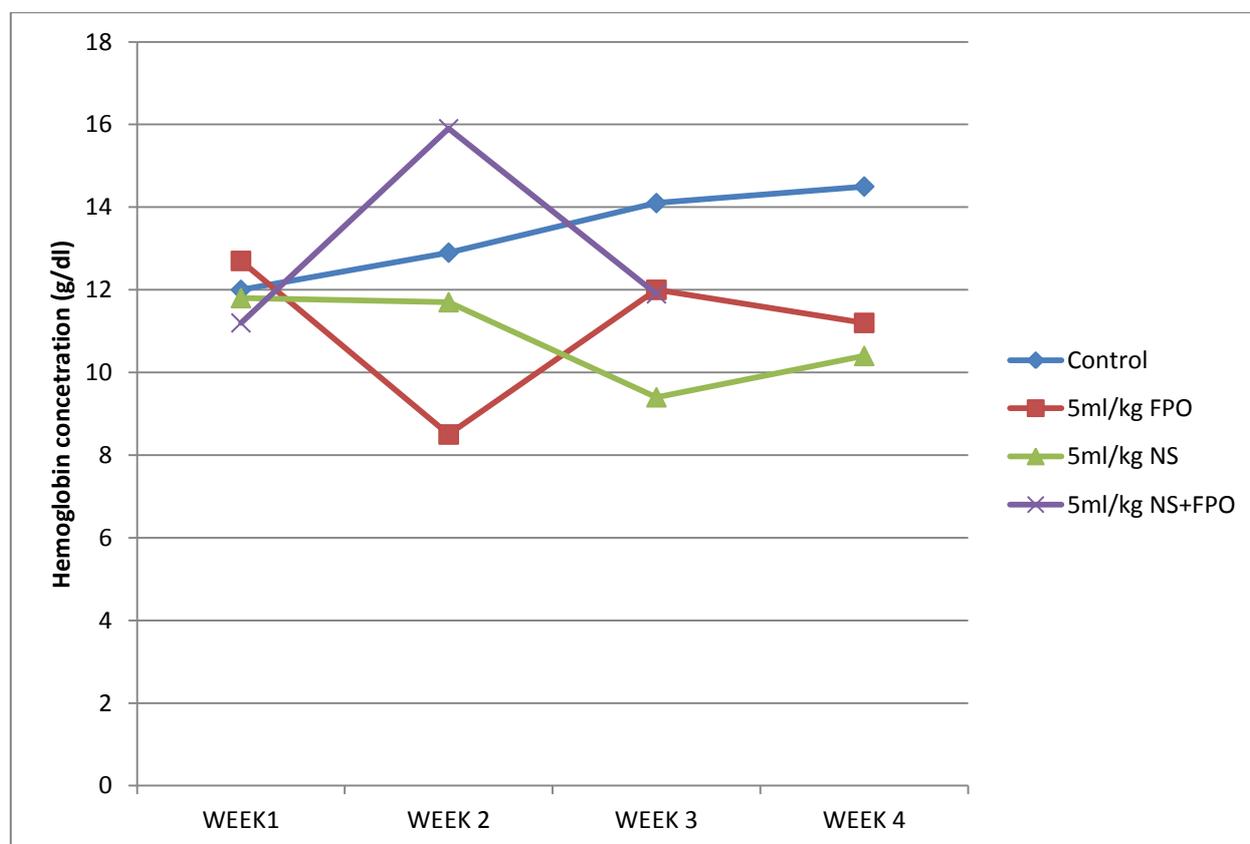
Fig ii ; Result of the Mean ± SEM of the weekly Red Blood Cell Counts x 10<sup>12</sup>/L)

Fig ii. Depicts the result of the weekly Red blood cell counts of the animals .The result shows some significant increase in groups A, B C and D. with differences between the baseline values and the week 3 values lying between  $0.93 \pm 0.06$ ,  $0.3 \pm 0.05$  and  $0.02 \pm 0.11 \times 10^{12}/L$  respectively. While group D decreased by  $0.67 \pm 0.36 \times 10^{12}/L$  respectively at the end of the study. This result shows that the RBC was significantly lower in the group that received 5ml/kg body weight of 3% natron-treated palm oil. Meanwhile, the decrease in the RBC of the group D may be due to the suppressive effect of the constituents of natron

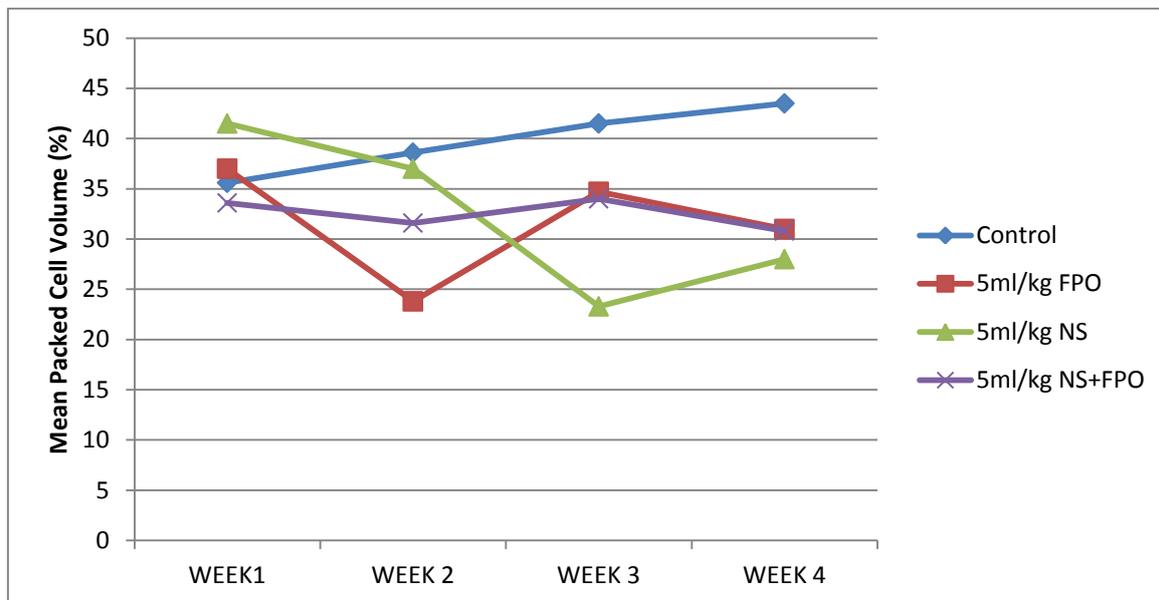
(kanwa) on the bone marrow and could also be as a result of failure of erythropoietin production. The kidney is the main source of erythropoietin which is produced by interstitial peritubular cells [4]. The group that received 5ml/kg of 3% natron had a decrease in the first two weeks of treatment. All the decrease in the RBC counts of groups D could be as a result of antioxidant property of fresh palm oil or as a result of the unsaturated nature of fresh palm oil, low level of low density lipoprotein cholesterol in the body which increases metabolism and promotes health. The RBC counts of the various groups were used to

compare the control group A and found that there was no significant difference ( $P>0.05$ ), despite the decrease in group D compared with the control that increased throughout the experimental periods. Meanwhile, figure iii shows the result of the weekly hemoglobin concentration (HB) of all the test substances. There was a significant increase in the hemoglobin concentration of the control group A and a slight increase/ negligible increase in group D. The hemoglobin concentration was significantly lower in the group that received 5ml/kg of 3% natron solution compared to the control group and other groups respectively. The decrease in the HB concentration may be a consequence of reduced uptake of iron by the damaged intestinal mucosa resulting in a reduced bioavailability of iron in the system<sup>[10]</sup>. The decrease in the HB concentration may also be attributed to the decreased storage of iron in the liver as a result of damage to the liver. The liver stores iron as ferritin and hemosiderin

<sup>[11]</sup>. The damage to the liver may have depleted the amount of iron stores and therefore resulted in reducing the HB concentration. Comparably low HB observed in the group C shows that the rats might have been anemic. The mean concentration of group A was significantly higher than group D ( $11.5\pm 0.25$ ), B ( $11.2\pm 0.45$ ) and C ( $10.4\pm 0.45$ ) but there was no statistical significant difference ( $P>0.05$ ) between the control group and groups B, C and D respectively. Similar results were also seen in the figure 4 of the result of the mean PCV otherwise known as hematocrit. The PCV is a function of RBC counts. It represents the percentage of RBC in blood<sup>[12]</sup>. Therefore the decrease in the PCV observed in the study is in agreement with the observed RBC. The mean PCV of the control group was significantly higher than that of group B which is higher than groups D and C respectively. There was no significant difference ( $P>0.05$ ) between the groups compared with control group.



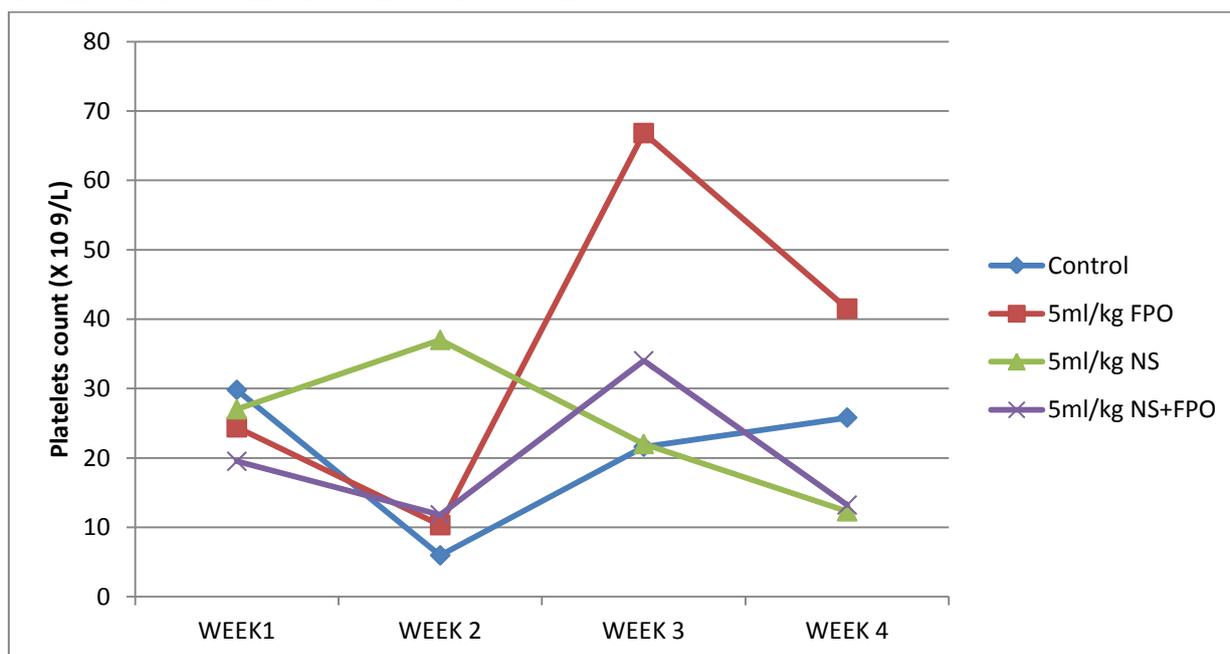
**Fig iii.** Results of the Mean  $\pm$  SEM of the weekly Hemoglobin Concentration of the animals (g/dl)



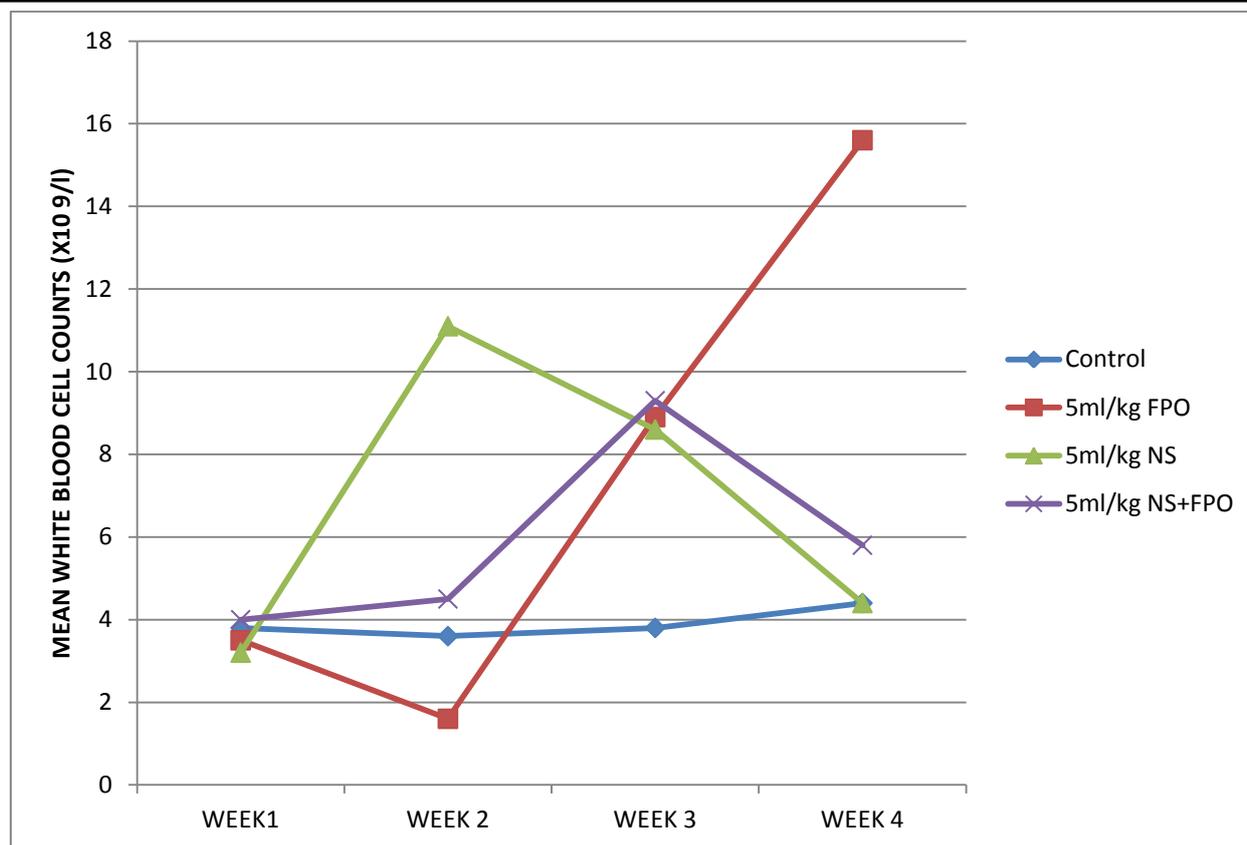
**Fig iv:** Results of the Mean± SEM of the weekly packed cell volume (%).

Fig v. Depicts the result of the mean platelet counts of the test substances. Groups A ( $25.8 \pm 1.02$ )  $\times 10^9/L$  was significantly lower than group C ( $12.3 \pm 1.02$ ), D ( $13.2 \pm 3.00$ )  $\times 10^9/L$ . But the platelet count of group B was significantly higher than that of all the groups and there was no significant difference ( $P > 0.05$ ) among the groups compared with control group A. The white blood cell (WBC) was observed to have increased in all the groups. This subsequent increase in the WBC of all the groups could be as a result of building up of its immunity against the damage that could have been done to other organs by the natron solution. The increase in the white blood cell

could be in line with normal sudden attack of the normal body cells (Simbulingham, 2006). Although, the increase did not have any significant difference ( $P > 0.05$ ) between group C and group D just like in the control group A and group B in their platelet counts. But there was tendency of the hematological parameters of the rats fed with 5ml/kg fresh palm oil and natron solution to be different from control because of other forms of oxidative stress processes, photolytic, enzymatic, hydrolytic and hemolytic which might be going on in fresh palm oil and natron<sup>[13]</sup>



**Fig v:** Result of the Platelet counts of the experimental animals ( $\times 10^9/L$ )



**Fig vi:** Result of the mean white blood cell counts of the experimental animals ( $\times 10^9/L$ )

The mean WBC of the control group A ( $4.38 \pm 0.26$ )  $\times 10^9/L$  was significantly lower than all the groups B ( $15.6 \pm 0.98$ )  $\times 10^9/L$ , D ( $5.8 \pm 0.8$ ) and C ( $4.4 \pm 0.22$ )  $\times 10^9/L$ . They were compared statistically with the control group A and found no statistical significant difference ( $P > 0.05$ ). Therefore, this study indicates that there is need to protect palm oil from various sources of oxidation. Therefore, this work is in agreement with Soladoye and Oyeleke<sup>[15]</sup> which showed that chronic intake of "kanwa" has adverse effect on the growth rate and blood parameters in rats even when diarrhea was absent as earlier reported<sup>[14]</sup>. This habitual consumption of natron (kanwa) was reported by Davidson et al, 1974 to have contributed to the periparturient cardiac failure in Zaria and Malumfashi areas of Northern Nigeria. Therefore, the aim of this study which is to assess the effect of chronic intake of kanwa in our tapioca (abacha) diets which is orchestrated by the local tapioca vendors within the south eastern Nigeria as a food tenderizer should be discouraged due to the

inability to have the accurate quantity or dosage of the natron to be added to their diet (jollof -tapioca) which could be detrimental to their health as seen in the group that received 5ml/kg of 3% natron solution only. Hence, this study recommends that further studies be conducted on the adverse effect of the natron (kanwa) on other health indices.

## CONCLUSION

If this research is applicable to man, the researcher suggests that there is reason for concern regarding the adverse hematological consequences of chronic consumption of natron (kanwa) in our diets as its adverse effect from indication outweighs the benefits. Chronic consumption of natron should be discouraged and an awareness campaign be carried out to inform the women vendors about the adverse effect of chronic consumption of natron (kanwa) in our local diets and encourage more of fresh palm oil than natron.

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**CONFLICT OF INTEREST**

No conflict of interest was declared by the authors.

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