

THE EFFECT OF AQUEOUS EXTRACT OF *XYLOPIA AETHIOPICA* (NIGRO PEPPER) ON SOME HAEMATOLOGICAL PARAMETERS IN ALBINO RATS

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ABSTRACT

The effect of *xylopiya aethiopica* on some haematological parameters in albino rats was studied. Thirty albino rats were randomly assigned into 3 groups, n =6. Group one served as control, group 2 received low dose of extract (100mg/kg body weight), while group 3 received high dose of extract (200mg/kg body weight). All animals had free access to feed and water *ad libidum*. The feeding lasted for 28 days and was done with the aid of an orogastric cannula. At the end of the 28 days, blood samples were collected and analyzed. Results shows no significant change in red blood cell count, packed cell volume, mean corpuscular haemoglobin, and MCV. WBC, Hb and MCHC was significantly lower ($p < 0.05$ and $p < 0.01$) in the low dose group, while clotting and bleeding time were significantly higher ($p < 0.05$ and $p < 0.01$) in the high dose group. In conclusion, aqueous extract of *xylopiya aethiopica* lower haemoglobin concentration at low dose, lowers white blood cell count, lowers mean corpuscular haemoglobin, increases bleeding time

and clotting time, hence care should be taken when using *xylopiya aethiopica* as it may be detrimental to our blood system.

KEY WORDS: *Xylopiya aethiopica*, blood cell, clotting time, bleeding time, white blood cell.

INTRODUCTION

Many medicinal plants are said to contain substances that are either of therapeutic value or are precursors for synthesis of useful drugs.^[1] One of such plants is *Xylopiya aethiopica* (Negro pepper) of the family Annonaceae. It is commonly known as “udo” in igbo (Nigeria).

It has been found to have antioxidant, hyperlipidemic and hypoglycemic properties. It is also for treatment of bronchitis, febrile pain, and rheumatism.^[2]

In some areas in Nigeria, fruits of *Xylopiya aethiopia* are used in the spicing in food; Fruit extract of *Xylopiya aethiopia* have been established to be helpful in management of a range of medical conditions such as dysentery.^[3] malaria.^[4] bronchitis, febrile pain, and rheumatism^[5], Oils of *X. aethiopia* have been used collectively with cosmetic products like creams and perfumes, they have also been used as insecticide.^[5] and as a preservative.^[6] Its extracts have also been reported to improve steroid hormones levels and sperm count, and to serve as an analgesic.^{[7][8]}

Methanolic fruit extract of *xylopiya aethiopia* have been reported to be beneficial in management of gastric ulcers.^[9]

Blood is a connective tissue containing cells floating in extracellular matrix called plasma. The cells found in blood exist at reasonably constant levels, due to close monitoring provided by a number of regulatory mechanisms in the body.^[10] The red blood cells' (RBC) primary role is conveying substances such as respiratory gases, nutrients and waste materials all over the body. The white blood cells (WBC) guard the body against pathogens and other foreign disparaging substances, while the platelets play the role of preventing blood loss (haemostasis). Deviation in the concentration of any of the above mentioned haemopoietic mechanism may be harmful to health. As a connective tissue, blood may be exposed to several exogenous insults for a number of days. Therefore evaluation of haematological indices could be vital information on what every system is exposed to in the use of *xylopiya aethiopia*.

MATERIALS AND METHODS

Extract preparation

The dried fruits of *Xylopiya aethiopia* were bought from Ugep market in Yakurr Local government area, Cross River state, Nigeria. Under the direct guide and identification of the traditional herbarium, the dried fruits were washed and grinded to coarse powder using an engine grinder. The powder was dried again and grinded to fine powder, using an electric blender. 800g of the powdered *Xylopiya aethiopia* were soaked in distilled water and stirred at intervals. The liquid extract was filtered with glass wool and the filtrate was then

evaporated to dryness using a rotary evaporator. A stock solution of 25mg/ml of water was prepared from the extract and the refrigerated until used.

Experimental Animals

Thirty albino rats from the animal house, department of Pharmacology university of Calabar were used for this study. The animals were grouped into three groups, control, low dose and high dose treated groups, n=6 each. The animals were housed in big plastic cages with wired gauge cover and are kept in the animal room department of Physiology university of Calabar. The rats were fed with growers feed a daily basis. All animals had unlimited access to water and feed before the commencement of the experiment, the animals were allowed to acclimatize for one week after which their body weight were measured accordingly before the start of the feeding regiment.

Administration of extract

Animals in the low and high dose treated groups received extracts of *xylopia aethiopica* at a dose of 100mg/kg and 200mg/kg body weight respectively for 28 days. Feeding was done orally with the aid of an orogastric feeding.

Collection of blood samples

At the end of the feeding period of extract administration, the animals were anaesthetized with chloroform after an overnight fast. Blood samples were collected into screw cap sample bottles containing heparin for assessment of the various haematological parameters. Red blood cells (RBC), Haemoglobin concentration (Hb), packed cell volume (PCV), Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH), Mean corpuscular haemoglobin concentration (MCHC) and White blood cell (WBC) count were determined using standard methods.

Estimation of bleeding time

Bleeding time was estimated following the method used by.^{[11][12]} It is briefly described as follows; the tail tip (0.5cm) of the animal was pricked under aseptic condition, a filter paper was then used to tap the blood every 30seconds, using a different portion of the paper each time. This was continued until bleeding stops. The total blood stained spots on the filter paper was counted and divided by two, and was recorded as the bleeding time in minutes. Timing was done with the aid of a stop watch.

Determination of clotting time

The tail tip of the animal was pricked and a drop of blood was placed on a clean glass slide. A pin was then passed through the drop of blood at intervals of 15 seconds to note when the fibrin thread starts to appear. Timing was done with the aid of a stop watch.

Statistical analysis

The results were expressed as mean \pm SEM. The One – way analysis of variance (ANOVA) was used to analyze the data. Values of $p < 0.05$ were considered significant. Computer software SPSS and excel analyzer were used for the analysis.

RESULTS

Table 1: Comparison of RBC parameters and clotting time between the control and test groups.

Parameter	Control	Low Dose	High Dose
RBC (millions/mm ³)	6.53 \pm 0.10	6.24 \pm 0.23	6.40 \pm 0.18 ^{NS}
PCV (%)	49.33 \pm 2.40	47.60 \pm 0.68	47.60 \pm 0.81 ^{NS}
Hb (g/dL)	11.85 \pm 0.25	10.85 \pm 0.06**	11.58 \pm 0.13
MCV (μ^3)	75.60 \pm 3.56	76.70 \pm 3.30	74.46 \pm 1.45 ^{NS}
MCH (pg)	18.16 \pm 0.25	17.49 \pm 0.75	18.13 \pm 0.45 ^{NS}
MCHC (%)	24.36 \pm 1.51	22.81 \pm 0.25	24.34 \pm 0.34 ^b
Bleeding time (mins)	7.33 \pm 0.77	8.26 \pm 0.09**	10.04 \pm 1.40 ^c
Clotting time (mins)	1.58 \pm 0.09	1.36 \pm 0.01*	2.08 \pm 0.03** ^c
WBC count (x1000cell/ μ L)	3716.6 \pm 87.22	2950 \pm 336.68*	3320 \pm 225.21

Values are mean \pm SEM, n = 6.

NS = not significant;

*** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$ vs. control. c = b = $p < 0.01$ vs control

From table 1 above, there was no significant difference in red blood cell (RBC), packed cell volume (PCV), mean corpuscular volume (MCV) and mean corpuscular haemoglobin values in the different experimental groups. However, the haemoglobin concentration (Hb) in the low dose treated group (10.85 \pm 0.06 g/dL) was significantly ($p < 0.01$) lower than control, no significant change was observed in the high dose treated group. (Table 1).

Mean corpuscular haemoglobin concentration (MCHC) in the high dose (24.34 \pm 0.34 %) was significantly ($p < 0.01$) higher compared with low dose treated group (22.81 \pm 0.25%) (Table 1).

The mean values for the bleeding time are 7.33 ± 0.77 mins, 8.26 ± 0.09 mins and 10.04 ± 1.40 mins for control, low dose and high dose treated groups respectively. Bleeding time was significantly higher ($p < 0.01$ and $p < 0.01$) in the high dose treated group compare with control and low dose treated groups (table 1).

The results for the clotting time for control, low dose and high dose treated groups are as follows; 1.58 ± 0.09 mins, 1.36 ± 0.01 mins and 2.08 ± 0.03 mins respectively. Clotting time was significantly ($p < 0.05$) lower in the low dose treated group compare with control. On the other hand, the high dose treated group had significant ($p < 0.01$ and $p < 0.01$) increase in clotting time compare with control and low dose.

White blood cell (WBC) count in the low dose treated group ($2950 \pm 336.68 \times 10^3$ cell/ μ L) was significantly ($p < 0.05$) lower than control, also, there was a non-significant decrease in WBC count compared with control (table 1).

DISCUSSION

This study investigated the effect of aqueous extract of *xylopiya aethiopica* on some haematological parameters using albino wistar rats. They was no significant change in red blood cell (RBC) count, packed cell volume (PCV), mean corpuscular volume (MCV) and mean corpuscular haemoglobin (MCH) among all the groups irrespective of the doses, this is in contradiction the report by Johnkennedy *et al.*^[13] where the authors reported a dose dependent increase in RBC, Hb, PCV and platelets.

Haemoglobin (Hb) concentration was significantly lower ($p < 0.01$) in the low dose treated group in contrast to the report by Onyebuagu *et al.*^[14] where the authors also reported an increase in Hb concentration.

Haemoglobin is one of the important haematological indices as it aids in the transport of oxygen to all tissues. Therefore a decrease in its concentration will imaginably lead to decrease oxygen transport and subsequent to tissue hypoxia.

White blood cell (WBC) was significantly lower ($p < 0.05$) in the low dose treated group and also lower in the high dose treated groups even though it wasn't significant against a report by Taiwo *et al.*^[15] where the authors reported an immunostimulatory properties of *xylopiya aethiopica* consequent upon its WBC count increasing property.

White blood performs a vital role in the body's immune system as they tend to fight and destroy foreign bodies such as disease causing bacteria, viruses as well as fungi. As long as white blood cells count remains in a physiologically normal state, the body's immune system is also normal. However, a decrease in WBC count as observed in this study might lead to decrease immunity thereby predisposing the body to every possible infection. This is in contradiction to a report by Iwu.^[16] where the author reported that *xylopia aethiopica* has an antimicrobial effect on gram positive and gram negative bacteria.

Mean corpuscular haemoglobin concentration (MCHC) was significantly lower ($p < 0.01$) in the low dose treated group compare with control and high dose treated group. Evaluation of MCH and MCHC are influential in identifying the morphology of RBC. Hence a decrease in MCHC clinically might confer conditions of hypochromic anaemia.

There is a paucity of information on the effect of *xylopia aethiopica* on both bleeding time and clotting time. This study hereby showed that bleeding time was prolonged after administration of aqueous extracts of *xylopia aethiopica*, irrespective of the dose. This infers that the extract may displace some clotting factors, which is a clinical diagnostic tool for haemophilia. This result however opposes the previous report from Burkhill.^[17] who stated that *xylopia aethiopica* arrests post partum bleeding. But in this study, *Xylopia aethiopica* prolonged bleeding time and may rather be responsible for clearing clots after delivery than arrest bleeding.

In conclusion, aqueous extract of *xylopia aethiopica* lower haemoglobin concentration at low dose, lowers white blood cell count, lowers mean corpuscular haemoglobin, increases bleeding time and clotting time, hence care should be taken when using *xylopia aethiopica* as it may be detrimental to the blood system.

REFERENCES

1. Sofowora, A., 1982. Medicinal plants and traditional medicine in Africa. John Wiley and sons LTD.
2. Ameyaw, Y. and E. Owusu-Ansah, 2005. Morphohistological studies of two plant species used in ethnomedicine. Ethnobot. Leaf lets, 2005; 1.
3. Oliver-Bever, B., Medicinal plants in tropical West Africa III. Anti-infection therapy with higher plants. Journal of ethnopharmacology, 1983; 9(1): 1-83.

4. Odugbemi, T.O., O.R. Akinsulire, I.E. Aibinu and P.O. Fabeku, Medicinal plants useful for malaria therapy in Okeigbo, Ondo State, Southwest Nigeria. *African Journal of Traditional, Complementary and Alternative Medicines*, 2006; 4(2): 191-198.
5. Adewoyin, F.B., A.B. Odaibo and C.O. Adewunmi, Mosquito repellent activity of *Piper guineense* and *Xylopiia aethiopia* fruits oils on *Aedes aegypti*. *African Journal of Traditional, Complementary and Alternative Medicines*, 2006; 3(2).
6. Kouninki, H., E. Haubruge, F.E. Noudjou, G. Lognay and F. Malaisse *et al*., Potential use of essential oils from Cameroon applied as fumigant or contact insecticides against *Sitophilus zeamais* Motsch. (Coleoptera: Curculionidae). *Commun. Agric. Applied Biol. Sci.*, 2004; 70: 787-792.
7. Woode, E., A. Alhassan and C.S. Abaidoo, Effect of ethanolic fruit extract of *Xylopiia aethiopia* on reproductive function of male rats. *Int J Pharm Biomed Res*, 2011; 2(3): 161-165.
8. Woode, E., E.O. Ameyaw, E. Boakye-Gyasi and W.K. Abotsi, Analgesic effects of an ethanol extract of the fruits of *Xylopiia aethiopia* (Dunal) A. Rich (Annonaceae) and the major constituent, xylopic acid in murine models. *Journal of pharmacy & bioallied sciences*, 2012; 4(4): 291.
9. Archibong, A.N., A.O. Obembe, C.C. Mfem, D.E. Ikpi and V.U. Nna, Effect of methanolic extract of *Xylopiia aethiopia* fruits on cytoprotection in cold stress-induced gastric ulcer in albino Wistar rats. *Res. Rev.: J. Med. Health Sci.*, 2014; 3: 155-160.
10. Guyton, A.C. and J.E. Hall, Red Blood Cells, Anemia and Polycythemia. In: *Textbook of Medical Physiology*, Guyton, A.C. and J.E. Hall (Eds.). 12th Edn., WB Saunders Company, Philadelphia, PA., USA., 2011; 13: 978-1416045748. 425-426.
11. Obembe, A. O., Omini, G. C., Okon, U. A., Okpo-ene, A. I., & Ikpi, D. E. Hematological and Immunological Effect of *Cannabis sativa* on Albino Wistar Rats. *British Journal of Medicine and Medical Research*, 2015; 7(1): 52.
12. Obembe A. O1, Okon, V. E., Ofutet E.O, Okpo-ene, A.I. Effect Of Chronic Consumption of *Cannabis Sativa* on Bleeding Time, Prothrombin Time and Platelet Count In Albino Rats. *International Journal of Science and Research (IJSR)*, 2015; 4(7): 2562-2565
13. Johnkennedy, N., Adamma, E., Austin, A., & Chukwunyere, N. E. Influence of *xylopiia aethiopia* fruits on some hematological and biochemical profile. *American J Med Sci*, 2011; 4: 191-6.

14. Onyebuagu, P. C., Pughikumo, D. T., & Aloamaka, C. P. Effects of Dietary *Xylopi*a *aethi*opica on Hematological Parameters and Plasma Lipids in Male Wistar Rats. *International Journal of Basic, Applied and Innovative Research*, 2014; 3(1): 29-34.
15. Taiwo, I. A., Oboh, B. O., & Francis-Garuba, P. N. Haematological properties of aqueous extracts of *Phyllantus amarus* (Schum and Thonn.) and *Xylopi*a *aethi*opica (Dunal) A. Rich in albino rats. *Ethno-Med*, 2009; 3(2): 99-103.
16. Iwu, M.M., 2014. *Handbook of African medicinal plants*. CRC press.
17. Burkhill, H.M., *The Useful Plants of West Tropical Africa, Volume 1: Families A-D*. 2nd Edn., Royal Botanic Garden, Kew, UK., 1985; 094764301X. 130-132.