Journal of Experimental Research

DECEMBER 2023, Vol 11 No 4

Email: editorinchief.erjournal@gmail.com
editorialsecretary.erjournal@gmail.com

Received: Nov., 2023
Accepted for Publication: Dec., 2023

HAEMATOLOGICAL EFFECT OF *ANTHOCLESTA NOBILIS* ON POTASSIUM BROMATE-INTOXICATED RATS

Chukwu U.E[1]. Aloh G. S[1], and Eze-Steven P.E[2].

[1]Department of Biochemistry, Michael Okpara University of Agriculture, Umudike. Abia State, Nigeria
PMB 7267, Umudike, Abia State, Nigeria.
[2]Department of Applied Biochemistry, Faculty of Applied Natural Sciences, Enugu State University of Science and Technology, Enugu State, Nigeria.

***Author for Correspondence:** ugblinks13@gmail.com

ABSTRACT

Potassium bromate used widely in foods has been associated with various complications and much attention has been drawn to the toxic effect of bromate on the organs like liver, kidney etc. In this study, the effect of the ethanol extract of *Anthocleista nobilis* leaves on kidney and liver damages induced by exposure to an acute dose of potassium bromate was measured using appropriate biomarkers. *Anthocleista nobilis*, known commonly as *Okpokoro*, is extensively used in traditional medicine to treat many disorders. Potassium bromate was induced through oral administration followed by oral administration of ethanol extract of *Anthoclesta nobilis* at doses 200mg/kg body weight, 400mg/kg body weight for 4 weeks, while the control group received distilled water and the positive control group received the ascorbic acid and distilled water. At the end of the experiment, the rats were sacrificed, body weight, relative organ weight, the parameters of the lipid. Treatment with ethanol extract of *Anthoclesta nobilis* at doses of 200mg/kg body weight, 400mg/kg body weight and 800mg/kg body weight showed significant increase of the body weight in treated groups. Result shows a significant increase in RBC, PCV, Hb and a significant decrease in WBC and PLT. Result also showed a significant decrease in urea uric acid, creatinine, sodium, potassium, chlorine and bicarbonate at ($p<0.05$). This study revealed that *Anthoclesta nobilis* was efficient in reducing and management of potassium bromated induced systemic toxicity and be a potential source of control for oxidative stress caused some environmental oxidants.

Keywords: Haemorrhage,*Anthocleista nobilis* (Okpokoro), haematology, potassium bromate

INTRODUCTION

Food additives are substances added directly and intentionally to food, usually in small quantities. Potassium bromate is a chemical compound used as additive in baking especially bread in many countries including Nigeria, to strengthen the dough and make it rise when added to fish paste as a conditioner; to beer or cheese and also, as a constituent in cold wave hair solution (Ueno, 2013; Oseni *et al*. 2015). Bromate was found to be genotoxic and carcinogenic (Sai *et al*, 1992) a discovery made while evaluating potassium bromate for acceptable level in baking flour meant for consumption by man (FAO/WHO,1992). Single doses of $KBrO_3$ have been reported to induce toxicity and the deleterious effect can be ameliorated by natural antioxidants (Ahmad *et al*. 2012). Furthermore, researchers found potassium bromate to have the potential of inducing cancer, liver failure, kidney failure, deafness, pains, redness of the eye and skin (Quick *et al*. 1976). This led to the ban of potassium bromated as food additives in several developed and developing countries (WHO (1993).

Plant determined items have been utilized for restorative purposes for quite a long time, it is assessed that 80% of the total populace depends on natural arrangements as drugs to meet their wellbeing needs (Macia *et al*. 2005) *Anthocleista nobilis* roots and stem are usually utilized for the treatment of diabetes, hypertension, intestinal sickness, typhoid fever, weight, the runs, looseness of the bowels, stomach torment, ulcer, jaundice, asthma, haemorrhoids, hernia, malignant growth, wounds, chest torments, irritations, ailment,

sexually transmitted diseases, barrenness and skin illnesses(Ngwoke *et al.* 2015).

The aim of this study is to identify the effect of continuous consumption of methanol extract of *Anthocleista nobilis* on potassium bromated intoxicated Wistar rats while the objective is to access the haematological effects of methanol extract of *Anthocleista nobilis* on potassium bromated intoxicated Wistar rats.

MATERIALS AND METHODS

Plant Materials and Extraction:

This work was carried out at Biochemistry Department of Michael Okpara University of Agriculture Umudike (MOUAU).

The leaves of *Anthocleista nobilis* were collected from the Forest reserve in Michael Okpara University of Agriculture, Umudike and properly identified at the Herbarium Unit of the Department of plant source and Environmental Biology where a voucher specimen: No 18 was kept**.**

Soxhlet extraction technique described by (Jensen 2022) was adopted. The leaves of *Anthocleista nobilis* were air-dried for 14days in the laboratory, and chopped into pieces before being pulverized into fine powder by grinding. The pulverised plant material (200g) was soaked in one (1) litre of 70% crude ethanol (solvent) and the conical flask was covered with an aluminium foil and allowed to stand for 72 h with constant agitation (Zirihi and Kra, 2003).

Animal Studies

A total of 30 adult male albino rats aged 8-10 weeks and weighing 80 to 120g and 21 mice were used for the study. Twenty-one mice were used for acute toxicity or lethal dose (LD_{50}) evaluation of the extract while 30 rats were used for the bromated toxicity studies. The animals were obtained from the Animal House of the Department of Zoology and Environmental Biology, Michael Okpara University of Agriculture, Umudike, housed in iron cages (5 rats per cage) and allowed to acclimatize for two weeks to allow for proper adaptation to their new environment and living conditions before commencement of the studies.

Experimental Design

Potassium Bromate intoxication and treatment

Thirty rats were used for this study. The animals were acclimatized for two weeks and were randomly divided into 5 groups with all groups comprising of six (6) rats each.

Group1: served as control (was given distilled water and feed *ad libitum*) (Negative control).While the other four groups were injected with a single intraperitoneal dose of potassium bromate at a dose of 50 mg/kg.b.wt., to induce oxidative stress. Group II: treated daily with oral dose of *Anthocleista nobilis* leaves extract (200 mg/kg b.wt) for 4 weeks. Group III: treated daily with oral dose of *Anthocleista nobilis* leaves extract (400 mg/kg b.wt) for 4 weeks. Group IV: treated daily with oral dose of *Anthocleista nobilis* leaves extract (800 mg/kg b.wt) for 4 weeks. Group V: treated daily 100 mg/kg dose of Vit. C + $KBrO_3$+ feed and water *ad lib* for 4 weeks positive control).

Table 1: Effect of ethanol extract of ANE on selected haematological parameters

Treatment groups	RBC x $10^6/mm^3$`	PCV %	Hb (g/dl)	WBC x $10^3/mm^3$	PLT x $10^3/mm^3$	MCV (fl)	MCH (pg)	MCHC (g/dl)
Normal control	7.17 ± 0.39^{d}	45.00 ± 2.55^{c}	17.92 ± 0.88^{d}	9.30 ± 0.86^{a}	110.00 ± 7.18^{a}	62.77 ± 2.38^{a}	25.00 ± 0.92^{a}	$39.84\pm0.68^{a,b}$
50 mg/kg KBrO3	4.72 ± 0.44^{a}	32.40 ± 2.61^{a}	13.24 ± 0.82^{a}	16.27 ± 1.37^{c}	129.20 ± 6.91^{c}	69.03 ± 7.09^{a}	23.01 ± 1.35^{a}	40.96 ± 2.33^{b}
200 mg/kg ANE+$KBrO_3$	5.85 ± 0.36^{b}	40.20 ± 2.17^{b}	16.04 ± 0.72^{b}	12.12 ± 0.88^{b}	$123.80\pm8.96^{b,c}$	68.76 ± 1.64^{b}	27.49 ± 1.84^{a}	$39.95\pm1.88^{a,b}$
800 mg/kg ANE+$KBrO_3$	6.45 ± 0.15^{c}	43.20 ± 2.17^{c}	$17.24\pm0.46^{c,d}$	11.60 ± 1.32^{b}	$120.60\pm4.56^{b,c}$	$66.95\pm2.17^{a,b}$	26.74 ± 0.85^{a}	$39.97\pm1.79^{a,b}$
100 mg/kg Vit. C+$KBrO_3$	5.84 ± 0.20^{b}	43.20 ± 0.84^{c}	$16.54\pm0.46^{b,c}$	12.17 ± 1.02^{b}	120.00 ± 5.24^{b}	74.01 ± 2.74^{c}	28.33 ± 1.11^{a}	38.29 ± 0.69^{a}

Results are presented as mean ± standard deviation (n = 5). The results with different letter superscripts are significantly different ($P < 0.05$) from any paired mean with the column.

RESULT AND DISCUSSION

Effects of ethanol extract of ANE on selected haematological parameters

The haematological effect Anthocleista nobilis leaf extract on the rats exposed to $KBrO_3$ is shown in Table 4.10. A significant decrease ($p<0.05$) in RBC, PCV MCV and MCH was observed when compared with the normal control group. A significant increase in WBC and PLT were observed in the $KBrO_3$ mixture co-treated with Anthocleista nobilis leaf extract compared to the control rats at varying doses. The effect shown in rats co-treated with Anthocleista nobilis leaf were dose-dependent.

The increased use of food additives such as potassium bromate ($KBrO_3$) has resulted in concerns over both the toxicity and safety of products such as, for example herbal medicines which are widely used by the public who perceive them as being natural, healthy, and free from side effects. A biochemical analysis of serum was carried out to assess haematological parameters and thus to evaluate the extent of any damage occurring. The measurement of haematological parameters can be helpful in determining the effects of chemicals and drugs on the health status of humans and animals. White blood cells (WBC) help to fight against infection and protect the body by phagocytosis against invasion by foreign organisms, and so their level is considered to be an important biomarker of information. The result of the present study showed a significant increase in WBC, which might be associated with inflammatory conditions due to the effect of $KBrO_3$ and similar chemicals as suggested by (Uchida et al.2006). Potassium bromate induces oxidative stress in human erythrocytes through the generation of reactive oxygen species and alters the cellular antioxidant defence system (Ahmad et al. 2014b).The increased level of lymphocyte showed lymphocytosis in rats administered with $KBrO_3$, which could be due to the depletion of the intracellular GSH by the diethylmalate in lymphocytes, which decreases the amount of strand breakage induced by potassium bromate (Parson and Chipman, 2000).However, on administration with Anthocleista nobilis extract decreased the WBC level.

It is also observed that $KBrO_3$ decreased the red blood cell count (RBC), haemoglobin level (Hb%), which agrees with the findings of Mohamed and Saddek (2019) who observed significant decrease in these parameters after rats were treated with a 100 mg/kg bur dose of $KBrO_3$. This decrease in RBC could bring about hemopoiesis effects resulting in anaemia. Anaemia is a condition that has multiple origins. One such origin is the destruction of red blood cells (RBCs) membrane induced by free radicals. Treatment of anaemia could therefore be enhanced by the use of free radicals' scavengers potentially found in some medicinal plants (Kuwara et al. 2019).

However, treatment of the rats with Anthocleista nobilis extract significantly increased the RBC count indicating its ability to correct anaemia, hemorrhage and blood cancer conditions

Rats in $KBrO_3$ administered groups showed a notable increase in platelets (PLT) as compared to the controls. PLT play an important role in blood clotting and prevent blood loss from hemorrhaging. Potassium bromate may thus adversely affect platelet levels. This result is in contrast with reports by Akinola et al. (2020). The administration of Anthocleista nobilis extract improved the PLT to a normal level. This effect may indicate an activation of the animal's immune system in response to tissue damage caused by any toxicant (Leeuwenberg et al. 2022). This study generally establishes the assertion that Anthocleista nobilis extract could improve haematological parameters. The chemical composition of the leaves might be connected with the increase in the haematological parameters observed in this study. Some of the components are well-known hemopoietic factors that have direct influence on blood production in the bone marrow.

CONCLUSION

This study reaffirms the negative food safety impact associated with continuous consumption of potassium bromate, and the ameliorative effect of ethanol extract of Anthocleista nobilis and its potential in preventing cardiovascular diseases. Increase in RBC PCV, Hb and decrease WBC count and PLT indicates that Anthocleista nobilis leave extract has a protective effect on blood cells.

REFERENCES

Ahmad MK, Khan AA, Mahmood R. (2013). Taurine ameliorates potassium bromate-induced kidney damage in rats. Amino Acids. 45: 1109-1121.

Ahmad S, Latif A, Qasmi IA. (2013). Aphrodisiac activity of 50% ethanolic extracts of Myristica fragrans Houtt. (nutmeg) and Syzygium aromaticum (L) Merr. and Perry.(clove) in male mice: a comparative study. BMC Complementary and alternative Medicine. 3(1): 1-5.

Kuwahara T, Ikehara Y, Kanatsu K, Doi T, Nagai H, Nakayashiki H, Kawai C. (2019). 2 cases of potassium bromate poisoning requiring long-term hemodialysis therapy for irreversible tubular damage. Nephron. 37(4): 278-280.

Leeuwenberg JF, Smeets EF, Neefjes JJ, Shaffer MA, Cinek T, Jeunhomme TM, Buurman WA. (2022). E-selectin and intercellular adhesion molecule-1 are released by activated human endothelial cells in vitro. Immunology.77(4): 543.

Macía MJ, García E, Vidaurre PJ. (2005). An ethnobotanical survey of medicinal plants commercialized in the markets of La Paz and El Alto, Bolivia. Journal of Ethnopharmacology. 97(2): 337-350.

Mack RB. (1988). Round up the usual suspects. Potassium bromate poisoning. North Carolina Medical Journal. 49(5): 243-245.

Ngwoke K, Anusi I, Eze P, Okezie U, Abba C, Abonyi D. (2015). Phytochemical and antioxidant properties of stem bark extracts of Anthocleista nobilis. European Journal of Medicinal Plants. 8(2): 107-111.

Ogbuehi IH, Ebong OO, Asuquo EO, Nwauche CA. (2014). Evaluation of the antiplasmodial activity of the methanolic root extracts of Anthocleista nobilis G. Don, Nauclea latifolia Smith and Napoleona imperialis P. Beauv. British Journal of Pharmacology and Toxicology. 5(2): 75-82.

Oseni OA, Olagboye SA, Idowu ASK. (2015). Potassium bromate induced renal toxicity in wistar albino rats: effects of aqueous extract of nutmeg (Myristica fragrans Houtt). British Journal of Medicine and Medical Research. 5(12): 1547-1556.

Parsons JL, Chipman JK. (2000). The role of glutathione in DNA damage by potassium bromate in vitro. Mutagenesis.15(4): 311–316.

Quick CA, Chole RA, Mauer SM. (1975). Deafness and renal failure due to potassium bromate poisoning. Archives of Otolaryngology. 101(8): 494-495.

Uchida HA, Sugiyama H, Kanehisa S, Harada K, Fujiwara K, Ono T, Makino H. (2006). An elderly patient with severe acute renal failure due to sodium bromate intoxication. Internal Medicine.45(3): 151-154.

Ueno H, Oishi K, Sayato Y, Nakamuno K. (2013). Oxidative cell damage in kat-sod assay of oxyhalides as inorganic disinfection byproducts and their occurrence by ozonation. Archives of Environmental Contamination and Toxicology.38: 1–6.

Zirihi GN, Kra AKM. (2003). Evaluation de l'activitéantifongique de Microglossa pyrifolia (Larmarck) O. Kuntze (Asteraceae) « PYMI » sur la croissancein vitro de Candida albicans. Rev Med Pharmacol Afr.17: 11-8