

Research Article

Effects of Ethanol Extract of *Pterocarpus Erinaceus* Leaves and Potassium Bromate Administration on Some Biochemical and Haematological Indices in Albino Rats

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Abstract: This study evaluated the effect of the administrations of ethanol extract of *Pterocarpus erinaceus* leaves and potassium bromate respectively and in combination on serum electrolytes, total protein, lipid profile and haematological parameters in albino rats. The experimental animals were divided into four groups: Group A (control) was administered water, group B were administered 200 mg/kg potassium bromate, group C were administered 200 mg/kg *P. erinaceus* extract and group D were administered a combined dose of 200 mg/kg potassium bromate and 200 mg/kg of *P. erinaceus* extract, all the administrations were done orally and continued daily for 21 days. It was observed in this study that potassium bromate decreased bicarbonate ion in the serum while the administration of *P. erinaceus* extract alone and in combination with potassium bromate increased bicarbonate ion in the serum. Rats administered with potassium bromate exhibited slight symptoms of toxicity while the *Pterocarpus erinaceus* leaf extract showed a strong preventive effect against some biomarkers of cardiovascular damage. Groups administered the combined dose of potassium bromate and the extract showed a uniform percentage increase in white blood cell, lymphocytes, hemoglobin, hematocrit, mean corpuscular volume accompanied by a decrease in red blood cells and platelet. The research reaffirmed the negative food safety impact associated with the consumption of potassium bromate, the ameliorative effect of the ethanol extract of *P. erinaceus* and its potential in preventing cardiovascular diseases.

Keywords: *Pterocarpus erinaceus*, potassium bromate, serum electrolytes, total protein, lipid profile, haematological parameters.

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INTRODUCTION

Some substances called food additives are incorporated into foods during processing to enhance their quality [1]. Generally, food additives can be classified into those intentionally added and those that get into food in small amounts due to food storage and handlings practices [2]. Various additives are approved by Codex Alimentarius [35] Commission and their maximum limit has also been set based on their potential toxic effects [2]. A white crystalline salt called potassium bromate ($KBrO_3$) was listed as flour treatment agent by FAO/WHO [36], and used for several years in the baking and confectionery industry to enhance the flavor of products [3, 1]. The use of potassium bromate was challenged with its official ban having been classified as a Category 2B carcinogen by IARC [4]. Toxicological studies have revealed that potassium bromate has potential to cause disruption of the plasma membrane of cells as an oxidizing agent and cause cells to empty their internal contents to the

extracellular environment and it is also toxic to organs [5, 6]. Potassium bromate was shown to initiate kidney toxicity in experimental animals and man [7], it was also reported to ignite DNA damage membrane and lipids peroxidation [8]. In addition, there are claims that potassium bromate could induce cancer [9, 10]. An increase in the occurrence of both malignant and benign tumors was observed after administration of potassium bromate, in the membrane that lines the abdominal cavity of laboratory animals [11]. Also, administration of potassium bromate led to a major increase in cancers of the thyroid, kidneys, and other organs of the animals [12].

Pterocarpus erinaceus is commonly referred to as Bloodwood/ Barwood in English and “Maijinni” in Hausa language. It has been attributed the status of a wonder herb, since it can cure various ailments when used in traditional medicine. The ailments include fever, intestinal worms, diarrhea, gonorrhoea, dysentery, and externally to treat ulcers, eye complaints, and sores.

According to Anne [13], the phytochemicals reported to be possessed by the plant includes; saponins, tannins, triterpenoids, flavonoids and steroids. The ethanolic stem bark extract has exhibited potentials in resolving inflammatory pathologies like dermatitis, gastric ulcer, rheumatism in addition to antimicrobial and antimalarial properties [14]. The bark or roots has been used to treat toothache, bronchial infections, anaemia, menstruation complaints, post-partum haemorrhage, leprosy, ringworms, wounds, antiemetic, tumors, purgative and tonic. Leaf decoctions are applied to treat syphilis [15]. Anne [13] reported that the aqueous extract of *Pterocarpus erinaceus* stem bark possessed analgesic, antioxidant, and anti-inflammatory properties. The plant has been found to be inhibitory against malaria parasite [16], possess anti-gonadotropic properties [17], as well as having anti-helminthic properties [18]. Etuk *et al.* [19] had reported in-vivo and in-vitro anti-mycotic activity of the plant. Plants generally have formed the basis of traditional medicine system which has a rapidly growing economic importance [20]. The fact remains that a large population of Africans are unable to afford modern medicine as a result; traditional medicine is usually their recourse [21].

This research is designed to ascertain the effect of administration of the ethanol extract of *Pterocarpus erinaceus* on the electrolytes, liver total protein and serum lipid profile of albino rats treated with potassium bromate. This is to further verify the ameliorative effect of the plant extract on potassium bromate toxicity and potentially, its effect on mitigating toxicity associated

Experimental Animal Grouping

Table-1.0: Animal group treatments

Group	Number of animal	<i>Pterocarpus erinaceus</i> Extract (200mg/kg)	Potassium bromate (200mg/kg)
A	5		
B	5		√
C	5	√	
D	5	√	√

Collection of Blood/ Serum

The blood sample was collected in plain sample container and immediately placed in the centrifuge; the centrifuge was powered and timed at 3000 rpm. After the time elapsed, the serum which appeared at the top of the bottle was collected with a syringe and stored in the freezer for analysis. Whole blood was collected into EDTA bottles, for the determination of hematological parameters.

Determination of Total Protein

This was done using a method by Gornall *et al.* [22].

with toxicants of the same mode of induction as potassium bromate.

MATERIALS AND METHOD

Albino rats weighing between 120-180g were obtained from the Animal House, Department of Biochemistry, Kogi State University, and Anyigba, Nigeria. The rats were acclimatized for 2 weeks prior to the experiment and exposed to 12 hours light and darkness while allowed access to food and water at libitum. The experimental animals were handled according to the guidelines set by the Research Ethical Committee of Kogi State University, Anyigba, Nigeria.

All the reagents and chemicals used for the study were of analytical grade and all the equipment were of laboratory standard.

Preparation of Extract

The leaves of *Pterocarpus erinaceus* were harvested from Kogi State University Campus and identified in the Department of Biological Sciences of the same Institution.

The leaves were air-dried for 2 weeks and pulverized. The pulverized sample was soaked in ethanol, constituted as 1g of dried extract powder in 3 ml of solvent, left for about 72 hours for maximum extraction of sample constituent. The extract was filtered using vacuum filtration method, the filtrate was concentrated using a rotary evaporator.

Total cholesterol

This was determined using a standard method by Flegg *et al.* [23].

HDL Cholesterol

This was carried out using a method as detailed by Friedewald *et al.* [24].

Triglyceride

This was done following a method by Tietz *et al.* [25] and McGowan *et al.* [26],

Determination of sodium ion, potassium ion and chloride ion in serum

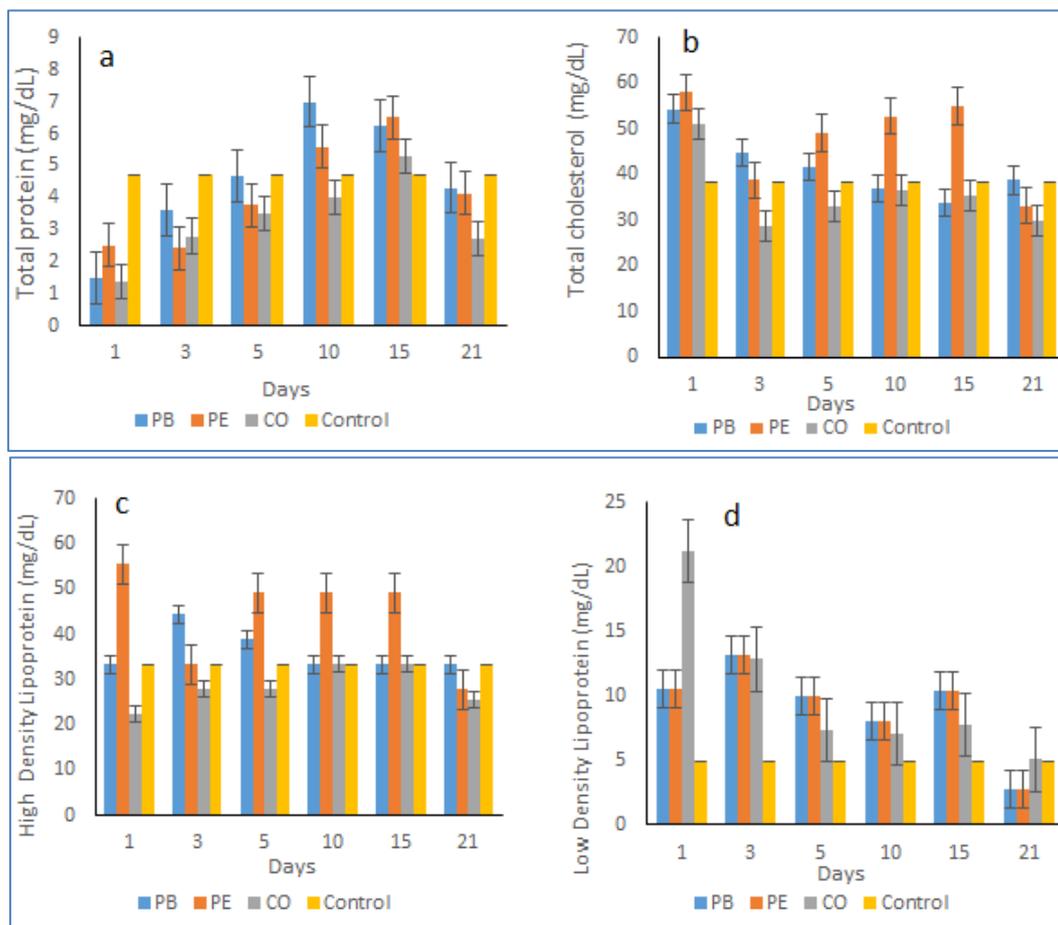
Colorimetric method was used for the determination of sodium and chloride while turbidimetric method was used for the determination of potassium in the serum.

RESULTS AND DISCUSSION

Administration of potassium bromate (PB), *Pterocarpus erinaceus* (PE) and their combined (CO) dose in albino rats showed several effects on the test animals studied. Generally resulting from all treatments, there was a decrease in total protein (Figure 1a) compared to the control, this decrease was higher with the co-administration of PB and PE (group D). The significant reduction in total protein at 21 days is indicative of a possible disruption of the protein synthesis machinery; this suggests that hepatotoxicity may have set in. This trend of increase and decrease in total protein observed during the course of treatment has been reported by Omer *et al.* [27]. An initial increase in total cholesterol (Figure 1b) levels above the control was observed in the first day of the co-administration, and first 5 days for potassium bromate and *Pterocarpus erinaceus* treatments respectively. The total cholesterol level decreased in the group administered *Pterocarpus erinaceus* and co-

administered PB and PE respectively on day 21 day. There was no significant ($p>0.05$) increase in HDL levels when compared to the control group (Figure 1c). However, for the PE treated group, there was a significant ($p>0.05$) increase in HDL concentration in the first 15 days. The PE and PB treatments resulted in an increase in TAG level in the first 15 days (Figure 1e). The increase resulting from PE was significantly higher ($p<0.05$) than the control. Co-administration of PE and PB resulted in lower TAG concentration. It was observed that the concentration of LDL was high but decreased remarkable on day 21 probably due to the increases of in HDL concentration [28]. Many studies have reported that increased level of LDL is associated with higher risk of atherosclerosis while elevated level of HDL is linked to reduced occurrences of cardiovascular disorders [29, 30].

The result of this study shows a significant increase in HDL concentration, a progressive decrease in LDL concentration and a corresponding decrease in total cholesterol (TC) and triglyceride (TAG) concentration. The study showed that the ethanol extract of *Pterocarpus erinaceus* has the potential of increasing the HDL/LDL ratio and as such will have a strong effect in preventing cardiovascular diseases.



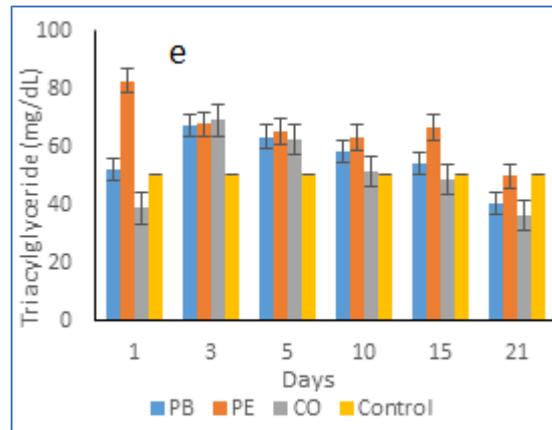


Fig-1: Effects of Potassium bromate, *Pterocarpus erinaceus* and their combination on some blood parameters in albino rats
 Note: PB = Potassium bromate, PE = *Pterocarpus erinaceus*, CO = Potassium bromate + *Pterocarpus erinaceus*

In the first 15 days of treatment, the three treatments significantly dropped the level of sodium in the blood (Figure 2a), with *Pterocarpus erinaceus* treatment having the greatest impact followed by potassium bromate. The level of bicarbonate (Figure 2b) was significantly higher than the control after 21 days for the *Pterocarpus erinaceus* and co-administered treatments, but below control for the potassium bromate treatment. Potassium level was higher than control with

the 3 treatment up to 15 days but by 21 days (Figure 2c), they had all reduced below control. There was no significant ($p > 0.05$) difference in the level of chloride ion (Figure 2d) in the blood during the treatment time. Reduction of sodium levels increases kidney excretion of water. Kurokawa *et al.* [31] had reported a lethal dose of potassium bromate in rats between 280 - 495 mg/kg bw.

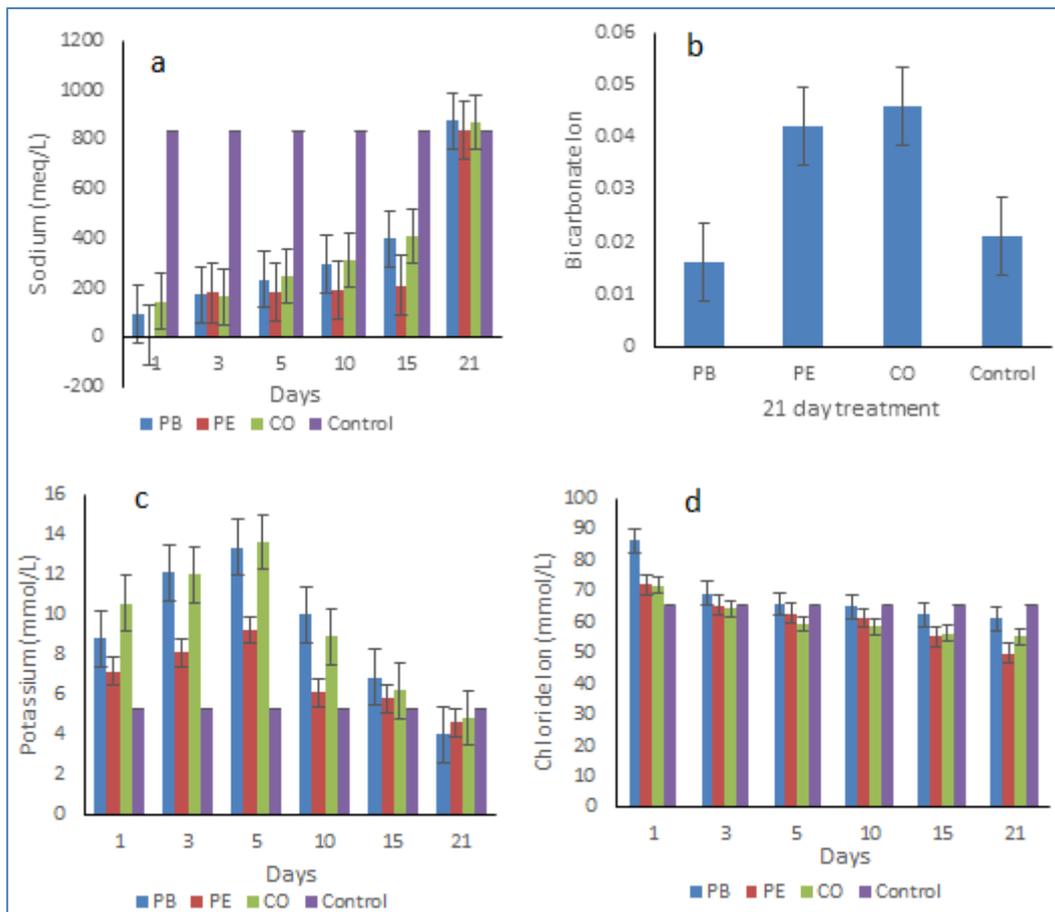


Fig-2: Effects of Potassium bromate, *Pterocarpus erinaceus* and their combination on serum electrolytes in albino rats

Table-2.0: Effects of Potassium bromate, *Pterocarpus erinaceus* and their combination on Haematological parameters

S/N	Haematological parameter	Potassium bromate (%)	<i>Pterocarpus erinaceus</i> (%)	Potassium bromate + <i>Pterocarpus erinaceus</i> (%)
1	WBC	25±2.3 ^a	23±1.1 ^a	49±2.5 ^b
2	LYM	17±1.2 ^b	9±2.2 ^a	16±2.0 ^b
3	HGB	16±1.0 ^b	14±0.8 ^b	8±1.0 ^a
4	HCT	20±2.8 ^b	8±2.5 ^a	19±2.1 ^b
5	MCV	20±0.7 ^c	14±0.0 ^a	16±1.9 ^b
6	RBC	-4±0.2 ^c	-2±0.1 ^a	-3±0.0 ^b
7	PLT	-22±2.1 ^b	-21±0.5 ^b	-11±0.0 ^a

Note: Values are presented as Mean ± SEM of duplicate determinations. Value with different superscript alphabets across a row are significantly ($p < 0.05$) different. Values represent % increase in haematological parameters as a result of treatments when compared to the control. (-) sign represents % decrease in haematological parameters as a result of treatments when compared to the control. white blood cell (WBC), lymphocytes (LYM), haemoglobin (HGB), hematocrit (HCT), mean corpuscular volume (MCV), red blood cells (RBC) and platelet (PLT).

The white blood cell increase triggered by the co-administration was significantly ($p < 0.05$) higher than that caused by the individual treatments based on comparison with the control (Table 2). The platelet count showed statistically significant ($p < 0.05$) decrease in post administration rats when compared with the control. The lymphocyte and hematocrit values recorded for *Pterocarpus erinaceus* extracts were significantly lower ($p < 0.05$) when compared to other treatments, however the difference in the levels of increase triggered for both lymphocyte and hematocrit by PB treatment and CO treatments were not significant ($p > 0.05$) (Table 2). Both red blood cell and platelet levels decreased irrespective of treatments. In both cases, potassium bromate administration triggered the most decrease. In contrast, mean corpuscular volume levels had its highest increase with the potassium bromate treatment, the lowest being associated with *Pterocarpus erinaceus* treatment. The reduction in platelet count could be due to the DNA strand breakage in these cells induced by the oxidative stress associated with potassium bromate [10, 9]. It has been established that the haematopoietic system is very sensitive to toxic compounds and serves as an important index of the physiological and pathological status in both animals and humans [32, 33]. According to Nadro and Modibbo [34], the administration of *Pterocarpus erinaceus* stem bark aqueous extract orally at over 5000 mg/kg did not induce any form of toxicity in rat, there was no mortality, weakness or any visible sign of toxicity and it is therefore safe for use in treatment of diseases [14]. The intraperitoneal LD₅₀ of the ethanolic stem bark extract of *P. erinaceus* was found to be 447.21 mg/kg, while the oral LD₅₀ was > 5000 mg/kg, hence it may be practically non-toxic through the (oral) route and may

contain some biologically active principle(s) which may be responsible for the haemostasis [14]. This research therefore validates the use of *P. erinaceus* as herb for management of several ailments especially cardiovascular diseases.

CONCLUSION

This research reaffirms the negative food safety impact associated with the consumption of potassium bromate, and the ameliorative effect of the ethanol extract of *P. erinaceus* and its potential in preventing cardiovascular diseases.

Conflict of Interest: The authors declare that there is no conflict of interest

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