

Effect of Potassium Bromate on Some Hematological and Biochemical Parameters and Protective Role of Vitamin C on Laboratory Rats (*Rattus Rattus*)

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Summary:

This study was conducted to investigate the toxic effect of potassium bromate on Wistar albino rats; twenty-four rats were used in this study and divided into four groups. Each group consists of six animals. First group the control G: which treated with (0.1)ml from normal saline orally for 21 days, second group: treated with potassium bromate 200mg/kg b.w orally for 21 days, third group: treated with vitamin C 400mg/kg b.w which given intraperitoneal (I.P.) for 21 day and fourth group, first treated with potassium bromate and after one hr treated with vitamin C also four 21 days, the results showed a significant decrease in all blood parameters (R.B.C.s, WBCs, Hb and PCV) in the group which treated with potassium bromate as compared with control and vitamin C group. Also, showed a significant decrease in total protein and a substantial increase in the liver enzyme (A.L.T. and A.S.T.) when treated animal with potassium bromate as compared with the control group, and from the results can be seen the vital role of vitamin C in return the blood and biochemical parameters for average level.

Keywords: *potassium bromate, vitamin C, hematological and biochemical parameters. Rats*

Introduction:

A white crystal, granules, or powder is potassium bromate, which is colorless and odorless. It has no therapeutic benefit, but it is applied as a maturing agent to rice, dough, fish paste as a conditioner, and to beer or cheese as well (1). It is also used in the cold wave hair solution as a component (2).

Potassium bromate is a material with a vapor density of 5.8 (air=1) and a density of 3.27 (g/cm³) and can cause a fire when it comes into contact with gasoline. It is incompatible with organics, aluminum and finely ground metals, reducing agents, and (3).

It has infinite solubility in water, and at drinking water pH, it should exist almost exclusively in the ionic form (4). The potassium bromate problem started with the ozonation of drinking water to form bromate as a significant by-product (5). When the research was done to confirm ozonated water's safety, it was found that potassium bromate caused renal cancer in rats when they drank water with potassium bromate. Following these discoveries, many countries, Health Organizations, and Agencies started banning the use of potassium bromate (6). Nonetheless, several experiments have been performed in various parts of the world to show that potassium bromate is harmful to health if ingested in food or drink. In both men and laboratory animals, it is nephrotoxic (7). Furthermore, potassium bromate caused renal cell tumors, mesotheliomas of the peritoneum, and follicular cell tumors of the thyroid (8).

Potassium bromate, which was given to male rats in drinking water, was shown to result in accumulative occurrence of adenomas and kidney carcinoma (9). The potassium bromate induces renal oxidative stress at the kidney, which is known to cause renal failure, methemoglobinemia, and kidney cancer (10). There have not been many reports on potassium bromate's effect on hematological indices; thus, this research is designed to experimentally investigate the impact of potassium bromate on some blood parameters using Wistar rats.

Potassium bromate (KBrO₃) is used widely in food and cosmetic industries to induce oxidation. It is used in bakeries as a food additive and flavor improver, giving strength and sponge-like characters to the dough in baking bread; potassium bromate is reduced to potassium bromide (KBr), a more soluble compound. However, this reaction can be incomplete, with some KBrO₃ remaining and contributing to possible oxidative damage. In certain countries, however, the use of KBrO₃ in bread production is banned. However, the manufacturers of flour do use potassium bromate (11). It is also used in treating barley in beer making and is commonly added to some Japanese fish pastes. Also, the use of ozone as a disinfectant in drinking water transforms bromide, which is typically found in water, into bromate, causing potassium bromate degradation of the water (Ueno et al., 2000). The LD₅₀ of potassium bromate (median lethal dose, 50 percent) was 150-385 mg/kg body weight. In humans, oral doses of 185-385 mg/kg body weight lead to irreversible renal damage and deafness, while lower doses lead to gastrointestinal irritation and stomach pain (12). Vitamin C is a water-soluble and highly effective antioxidant. Vitamin C even given in small amounts; it can protect the indispensable molecules in the body, like nucleic acid lipids, proteins, and carbohydrates, from reactive oxygen species and free radical undesirable effects that can be provoked during the normal metabolic process as well as after exposure to toxins (13).

Previous studies have related vitamin C to hepatoprotective properties and have also reported synergistic hepatoprotective effects with other antioxidants (13,14).

Aim of the study:

This article's objectives were to investigate the toxic effects of potassium bromate on hematological and biochemical parameters and the protective function of vitamin C in albino rats.

MATERIALS AND METHODS:

Study Design:

Twenty-four adult male albino rats, their weights were ranging between 200 and 210 gm, were included in this study. The experiment was performed at the University of Basrah's Veterinary Medicine Student Animal Building. They were kept in clean, well-ventilated polypropylene cages under identical conditions and had a sufficient supply of food and water during the experiment.

They were acclimatized to the laboratory setting at least two weeks before the start of the research. In this study, rats were divided roughly into four experimental groups:

Group I: acted as a monitoring group and received 0.1 ml of normal saline orally for 21 days.

Group II: Rats received potassium bromate (KBrO₃) 200 mg/kg b.w orally for 21 days. (potassium bromate, which obtains from the department chemistry College of Science University of Basrah Iraq.

Group III: Rats received vitamin C 400 mg/kg b.w in intraperitoneal injection for 21 days. (vitamin C, local production Iraq).

Group IV: Rats received (KBrO₃) 200 mg/kg b.w orally and after 1 hr received vitamin C 400 mg/kg (I.P.) for 21 days.

After the end of the experiment, the rats were sacrificed under general anesthesia. Placing them in tightly closed glass containers cotton soaked with chloroform as inhaled anesthesia; after that, the abdominal cavity was opened, and a Y-shaped cut the rat abdomen was done. Blood collected via direct heart by using a 5 ml disposable syringe; some of the blood samples then transferred into plain tubes centrifuged at (3000 rpm for 15 minutes) to obtain the serum, which then transferred into several Eppendorf tubes to use in analyses of different parameters and stored at -4°C, and 2 ml of the blood were poured into anti-coagulant containing tubes to be used later for hematological examinations.

Study parameters

Hematological parameters

A. Red blood cells count (R.B.C.) (cell/mm³)

The red blood cell count was obtained by the use of a haematocytometer (Neubauer improved double) and (Hayme's solution) and a special pipette for dilution (15).

B. Hemoglobin concentration (Hb) (g/dl)

Hb concentration is estimated by Sahli apparatus, where an amount of 10 ml of HCl of 0.1 normality

is poured into Sahli tube, and 10 µl of the blood specimen is added and mixed, then the mixture is let for 10 minutes, and later on, distilled water is added gradually till the color of the blood is rendered similar to that of the standard tube color(16).

C. Packed cells volume(PCV.)

Used the micro hematocrit method to calculate the percentage of PCV by using capillary tubes that contain heparin, where one end is closed by an elasto plastic after being filled to 3/4 of its length with blood. It is put in micro centrifuge on a 1200 rotation/minute velocity for five minutes; then, the hematocrit value is obtained by Service device (17).

D. White blood cells count (WBC)(cell/mm³)

The WBC was obtained using Haemocytometer (Neubauer improved double) and Thoma's solution and a special pipette for dilution(18).

Biochemical parameters:

Serum transaminases activity determination:

Serum transaminases activity ALT or AST is determined by using a special kit (BIOMÉRIEUX®sa Transaminases-Kit, France) (19).

Results:

Hematological results:

Table(1) showed a significant decrease in the number of (R.B.C., WBC count, hemoglobin concentration, and packed cell volume) after administration of potassium bromate alone to the animals when compared to the control and vitamin C group, which also can be seen significantly return in all blood parameters to the normal value in group four when giving a combination of potassium bromate and vitamin C compared to control group. But the values still less considerably from those of the control group value.

Table(1). The effect of potassium bromate and vitamin C's role on malerats' blood parameters for 21 days

PARAMETERS GROUPS	RBC ($\times 10^6/\text{mm}^3$)	Hb(g/dl)	PCV(%)	WBC ($\times 10^3/\text{mm}^3$)
Control\G1	A 5.00±0.00	A 13.30± 0.26	A 36.01±0.46	A 5.83±0.40
G2 potassiumbromate	D 3.00±0.00	D 9.66±0.45	C 30.50±0.30	C 4.08±0.10
G3 vitaminC	B 4.33±0.51	B 12.31±0.27	A 35.61±0.37	B 5.17±0.15
G4 Potassiumbromate &vitamin C	C4.00 ±0.00	C11.0 3±0.61	B33.8 8±0.80	D4.65 ±0.31
LSD	0.33	0.98	1.73	0.51

*Different letters denote significant differences among groups ($p \leq 0.05$).

2-Biochemical parameters:

table (2) shows a significant decrease ($p \leq 0.05$) in total protein and a substantial increase in A.L.T. and A.S.T.) enzyme when treated animals with potassium bromate as compared to the control group, while when treated animals with vitamin C after one hour from treated animals with potassium bromate there's improved in

all biochemical parameters as compared with the control group.

Table(2).The effect of potassium bromated and vitamin C's role on malerats' bio chemical parameters for 21 days.

PARAMETERS GROUPS	Total protein (g/dl)	ALT(U/L)	A.S.T.(U/L)
G1 Control	A7.08 ±0.07	C151.5 3±2.08	D31.30 ±0.20
G2 Potassium bromate	C 5.11±0.11	A 234.21±7.00	A 93.90±0.61
G3 Vitamin C	A 6.88±0.44	C 151.16±1.94	C 32.36±0.39
G4 Potassium bromate & vitamin C	B6.35± 0.24	B188.8 6±6.68	B42.1 6±0.15
LSD	0.53	37.33	1.06

*Different letters denot to significant differences among groups(p≤0.05).

DISCUSSION:

The present study has shown, when treated animals with potassium bromated (200mg/kg) from body weight, there is a decrease in all blood parameters (R.B.C.s, WBCs, HB, and PCV) as compared with the control and vitamin C group and decrease the total protein with an increase in A.L.T. and A.S.T. enzyme. From this study, it observed that leucocytes decreased in rats administered with potassium bromated in water. The decrease in the leucocyte count is in agreement with (20).

Decreased leukocyte count has also been reported due to the consumption of chemicals like Bromate (21). These decreases in the number of leucocytes may be due to D.N.A. strand breakage in these cells caused by oxidative stress associated with potassium bromate.(22).

Also, bone marrow suppression with selective megacaryocyte depression may have occurred (21). Besides, a study in mice found little improvement in blood parameters other than a temporary decrease in R.B.Ccount(23).

The temporary decrease in Hb and PCV values in rats received 200 mg/kg B w, potassium bromate at the third week is an anemia indicator. Similar results reported by. (1) and. (24), who claimed that potassium bromate was known to induce methemoglobinemia and cyanosis that resulted from oxidation of ferrous ion to ferric by reactive oxygen species (R.O.S.) generated from potassium bromate. The reduction of total protein may be due to liver damage, which resulted in a reduction of protein synthesis.

The increase of A.L.T. and A.S.T. enzymes may be attributed to the high permeability of the hepatocyte membranes or its destruction in the liver cell. This finding is in agreement with Kurokawa *et al.*(1990). Increased serum A.S.T. activity and A.L.T. activity are also used to predict hepatic damage, signaling cellular leakage of intracellular enzymes and lack of liver cell membrane stabilization (25). Also, maybe due to destruction of the hepatocytes induced by KBrO₃ Causes a substantial improvement (P<0.05) in A.S.T. and A.L.T. in Group 2. Conversely, a significant decrease in vitamin C-treated rats' enzyme activity indicates its potential to enhance the harmful effects of KBrO₃. Decreased transaminases propose maintaining the plasma membrane's stability and shielding the hepatocytes from damage incurred by hepatotoxin. The frequently-recognized view is that serum transaminase levels return to normal with the healing of hepatic parenchyma and hepatocyte regeneration(26). Potassium bromate (KBrO₃) has many dangerous effects. KBrO₃ exerts nephrotoxic and ototoxic effects in experimental animals as well as in men. It is a carcinogen inducing renal cell tumors, mesotheliomas, and thyroid follicular cell tumors in rats. Active oxygen radicals are probably involved in these effects leading to D.N.A. damage. Efforts to stop the use of potassium bromate in food processing should be stepped up.

conclusion:

From this study can be conclude the toxic effect of potassium bromated in hematological and biochemical parameters of laboratory animals (rats) for 21 day and the important role of vitamin C in return the all parameters to normal levels.

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