University of Zawia

Faculty of Science

Biology Department – Zoology division

Protective Effect of Red Onion on Potassium Bromate-Induced Toxicity in Male Albino Rats

Thesis Submitted in Fulfilment for the Degree of Master of Science

by:

Amera Mohamed Aldaek

Supervised by:

Dr. Nadia M Kermani

Dr. Fikry A Abushofa

2020

ABSTRACT

Potassium bromate is an oxidizing agent and one of the best and cheapest dough improvers in the baking and food industry. Although negative effects are not evident in animals fed diets made from flour treated with $KBrO_3$, the agent is classified as carcinogenic in rats and nephrotoxic in both man and experimental animals when given orally. Red onion that is *Allium cepa* used in daily diet for taste. Onions also have number of medicinal properties, such as anticancer, antifungal, antioxidant, antiulcer, anti-inflammatory, etc. This study was designed to investigate the protective effects of aqueous extract of red onion against $KBrO_3$-induced toxicity. This can be achieved by monitoring changes in hematological and biochemical parameters and histopathological observation. 36 Wistar male rats divided into 6 batches. During a 4 week period, (group I) served as a control. Group II received (100 mg/kg b.w of $KBrO_3$) on the 24^{th} and 27^{th} days. Group III received 1 ml of red onion juice/ 100 g b. w every day and (100 mg/kg b.w of $KBrO_3$) on the 24^{th} and 27^{th} days of the experiment. The group IV received (50 mg/kg b.w of $KBrO_3$) twice a week. Group V received 1ml of red onion juice/100 g b. w /day and (50 mg/kg b.w of $KBrO_3$) twice a week. The (group VI) received (30 mg/kg $KBrO_3$) every day till the last. The rats were weighed and sacrificed after completion of the treatment, Blood samples for hematological and biochemical analysis. Kidney and small intestine tissues were collected and fixed in 10% formalin for histopathological studies. The results showed significant ($p< 0.05$) increase in the WBC in groups received (30&50 mg/ kg b.w of $KBrO_3$) and significant ($p< 0.05$) decrease in PLT in group that received (100 mg/ kg b.w of $KBrO_3$). Additionally, results showed significant ($p< 0.05$) increase in urea, uric acid creatinine, ALP and AST in all treated groups with $KBrO_3$ compared to the controls. However, administration of red onion juice with KBrO3 showed improvement in the studied parameters. The histopathological analysis of kidney showed dilatation of Bowman's capsule, hemorrhage, degeneration, congestion, necrosis as well as the loss of brush border in renal tubules. $KBrO_3$-treated small intestine showed degenerative villi, an increase in the goblet cells in epithelium and the mucosa of the intestine is infiltrated by inflammatory cells. From these results it is clear that the toxic effect of $KBrO_3$ were pronounced more in the group that received single dose of 100 mg/ kg b. w. This indicates that the higher the dose, the greater the effect. The daily dose of (30 mg/kg $KBrO_3$) also indicates the risk of daily exposure to potassium bromate. Also, the red onion juice works as protective agent against $KBrO_3$ induced damage by reducing the physiological and histological alterations. These results suggested that the protective effect of red onion may be attributed to their antioxidant activities.

Acknowledgements

In the name of ALLAH, I would like to express my special thanks to Allah, who gave me the necessary health, strength and patience to conduct this work.

I would like to express my sincere gratitude to my supervisors, Dr. Nadia Mohamed Kermani and Dr. Fikry Ali Abushofa, for their continuous support, encouragement and immense knowledge in this challenging research. Their guidance helped me throughout the duration of the research and writing this thesis.

I would like to thank all the staff at the Animal House of the National Medical Research Centre in Al-Zawia City, especially Dr. Khaled and Dr. Akram for their kind help, supplying me with rats for the research and support in completing my work.

I would like to thank technicians Ahmed Alqadeery, Akram Abbashar, Warda Alkhitony and all the team at the histopathology laboratory at Sabratha Teaching Hospital in providing me with needed equipments. Without they precious support it would not have been possible to conduct this research. Also, I would like to sincerely thank Mr. Ibrahim Ammar who drives me to Sabratha Teaching Hospital to carry out my histology part.

My sincere thanks also go to the technical staff at Al-Raed medical analysis laboratories, for providing me with the opportunity to join their team as intern and allowing access to laboratory and research facilities. I would like to thank Dr. Magda Hasan for providing us with the microscope and the camera.

My deepest appreciation also extends to all lecturers and staff in the Department of Zoology, Faculty of Science at Al-Zawia University for their kind help, generous advice and support during the study.

With the love and support of my family, this project has certainly not been carried out singlehandedly. Special thanks go to my mum and dad, my husband and beloved kids. They have always been proud of me and believed in me in striving to achieve my goals.

DEDICATION

To:

MY FATHER AND MY MOTHER

MY HUSBAND

MY SON AND MY DAUGHTER

Declaration

I hereby declare that the work in this thesis is my own except for quotations and summaries which have been duly acknowledged

Amera Mohamed Aldaek 12 November 2020

Table of Content

CHAPTER III

CHAPTER IV

CHAPTER V

LIST OF FIGURES

LIST OF TABLES

ABBREVIATIONS

AcE	*Allium cepa* Extract
ALP	Activity Alkaline Phosphatase
ALT	Activity Alanine Aminotransferase
AO	Antioxidants
AST	Activity Aspartate Aminotransferase
BUN	Blood Urea Nitrogen
CBC	Complete Blood Count
CSE	Centre for Science and Environment
H&E	Hemotoxyline and Eosin
HB	Hemoglobin
Hct	Hematocrit
HDL	High Density Lipoprotein
IARC	International Agency for Research on Cancer
KBr	Potassium Bromide
$KBrO_3$	Potassium Bromate
LDL	Low Density Lipoprotein
LPO	Peroxidation
PCT	Plateletcrit
PLT	Blood Platelets

Ppb	part per billion
RBC	Red Blood Corpuscles
ROS	Reactive Oxygen Species
WBC	White Blood Corpuscles

CHAPTER I

1. INTRODUCTION

1.1 Research Background

A food additive is a substance or mixture of substances present in a food as a result of any aspect of production, processing, storage, or packaging. More than 2500 different additives are intentionally added to foods to produce desired effects and to modify flavor, colour, stability or texture (Branen *et al.* 2001). The use of these additives is a well-accepted practice but is not without controversy.

Potassium bromate ($KBrO_3$) is a chemical additive mixed in flour to improve the action of gluten. Gluten is a protein found in wheat flour that gives the dough of the bread elasticity during kneading and thereby encourages the fermentation of the dough by retaining the gases produced by yeast. By strengthening the gluten, potassium bromate causes the bread to increase in volume and hold its shape (Chauhan & Jain 2016). Hence, potassium bromate helps to strengthen and soften the dough and gives the finished bread an appealing white colour. When it is used within prescribed limits of 15-30 ppm, it changes during the baking process and leaves no trace in the end product (Shanmugave *et al*. 2020). Ideally, therefore, the end product should contain no potassium bromate, which has been broken down during the baking

process into potassium bromide (KBr), a harmless byproduct (McMinn 2017). However, if the mixture includes higher amounts of potassium bromate, or if the bread is not fully cooked or has been baked at insufficiently high temperature, then residual amounts of $KBrO_3$ remaining in the bread would be harmful to health when consumed (Oyekunle *et al*. 2014; Ben Saad *et al*. 2016).

Bromate can be also found as a disinfection byproduct of the ozonation of drinking water (Ahmad *et al*. 2012). The bromide in the source water is oxidized to bromate by the ozone. Furthermore, besides its use in the baking industry, potassium bromate is also widely used in cosmetic, especially in hair lotions, in treating barley in beer making, and in fish paste and cheese production (Ahmad *et al*. 2015). Despite the benefits of this chemical, toxicological studies have shown that bromates cause kidney, thyroid and gastrointestinal cancer in experimental animals, and the International Agency for Research on Cancer (IARC) classifies bromates as a group 2B carcinogen (a possible human carcinogen) with renal tumour risks at high doses (IARC 1986). As a result of extensive investigations into the potential risks associated with $KBrO_3$, this food additive is banned in many countries such as the United Kingdom, France, Nigeria, and Canada (Fawell & Walker 2006; Qin *et al.* 2018).

Potassium bromate is a powerful oxidizing agent that forms free radicals in cells (Khale *et al*. 2018) which are more generally known as reactive oxygen species (ROSs). ROSs play important roles in the mechanism of bromate toxicity (via lipid peroxidation, oxidative stress, etc.) and are considered one of the key factors that promote chemical carcinogenesis (Ahmad *et al*. 2012; Ahmad & Mahmood 2014). At high concentrations of $KBrO_3$, increased levels of ROSs will be produced which can damage

all major cellular elements such as protein, lipids, and DNA (Murata e*t al.* 2001). The ROSs react with DNA and induce oxidative damage such as strand breaks, and consequent gene mutation and chromosomal aberrations (Starek & Starek-Swiechowicz 2016).

Indeed, several reports indicate toxicological effects and several diseases, including cancer (Kurokawa *et al*. 1990; Ahmad *et al*. 2013). $KBrO_3$ has been shown to be nephrotoxic in both human and experimental animals (Uchida *et al* 2006). Although the kidney is considered to be the main organ affected by $KBrO_3$, it also causes severe tissue damage to many other organs of treated rats and mice including the liver, thyroid, testes and intestine (Delker *et al*. 2006. Ahmad *et al.* 2012; Ahmad *et al.* 2013). Also, it induces kidney cancer, peritoneal mesotheliomas, and follicular cell tumours of the thyroid (Kurokawa *et al*. 1990; Parsons & Chipman 2000; Umemura & Kurokawa 2006; Starek & Starek-Swiechowicz 2016).

Several antioxidants (AOs) have shown protection against bromate-induced toxicity. These antioxidants could reduce or prevent free radical formation. The antioxidants disrupt the oxidation chain reaction and neutralize free radicals, leading to delays to or the inhibition of cellular damage. The antioxidant activity of several plant materials has recently demonstrated potential protective effects against $KBrO_3$ toxicity (Khan 2003; Bao *et al*. 2008; Ahmad *et al*. 2013; Ali *et al*. 2018). The search for newer effective natural antioxidants, especially of plant origin, has since intensified.

Onion (*Allium cepa L.*) is one of the most widely consumed and cultivated vegetable crops in the world (Santas *et al.* 2010). It is one of

the vegetables richest in flavonoids, mainly quercetin and its derivatives (Zhang *et al*. 2016), which are effective antioxidants due to their capability to scavenge free radicals. Traditionally, onions have been used as an herbal remedy for a wide range of disorders due to their association with various pharmacological effects (Rose *et al*. 2005). Scientific reports have confirmed their functional properties, which include: antioxidant activity (Nasri *et al*. 2012); a hepatoprotective effect (Ozougwu & Eyo 2014); anti-carcinogenic, anti-infection properties (Corzo Martínez *et al*. 2007) and sundry other biological actions (Ashwini & Sathishkumar, 2014).

1.2 Significance of the Study

Bread is a staple food of humans worldwide (Pagewise 2002) and one of the main and most popular foods in Libya. Unfortunately, the basic ingredient of bread is flour which usually contains a flour improver such as potassium bromate to improve the quality of the product. In addition, most bakeries use $KBrO_3$ (legally and illegally) via brominated flour to prepare a variety of bread, pastas and biscuits which are commonly consumed in the region. Elssaidi *et al.*(2013) detected concentrations of $KBrO_3$ in bakery products in the southern region of Libya as well as in flour in local markets flour, and also in other products such as sweets and potato crisps. Moreover, it is also known that bromate is generated as a by-product during the ozone disinfection of drinking water (Cavanagh *et al*. 1992). Although the concentration of bromate in drinking water is usually very low, it is still likely to be associated with human health problems (Wei *et al*. 2009). Therefore, there is an urgent need to explore methods to manage the toxicity caused by $KBrO_3$ in natural resources so as to avoid serious side effects.

Red onion is used as a protective source in this study, based on the known use of this species in foodstuffs and medicines for a wide range of disorders.

1.3 Objectives of the Study

The present research studies the ameliorative potential of red onion on the adverse effects of $KBrO_3$ in Wister albino rats. Its objectives are as follows:

1. To study the effect of $KBrO_3$ on hematological and biochemical parameters in Wistar rats; and to observe any protection provided by red onion extract against $KBrO_3$ toxicity.

2. To investigate the nephrotoxic effect of oral doses of $KBrO_3$ in rats and the possible ameliorative effect of pre-treatment with red onion extract.

3. To examine the intestinal histopathological change associated with $KBrO_3$-induced toxicity and its reversal using red onion extract.

CHAPTER II

2. LITERATURE REVIEW

2.1 Background

The use of food additives has been practised for thousands of years. More than 3000 food additives are currently listed by the USA Food and Drug Administration (FDA), including flavourings, colourings, preservatives, dough strengtheners, and conditioners, among others (Andreozzi *et al.* 2019). The usage of food additives has, however, changed dramatically and clearly throughout history (Wilson & Bahna 2005). Since the early 19th century, food additives that are incorporated into almost all processed foods has increased dramatically (Jansen *et al*. 2020).

Due to improvements and developments in preservation and processing technology, the numbers of various additives and the scale of their use have considerable increased. In recent years there has been some concern about safety and the adverse effects of additives used in food. Therefore, food additives are now more rigorously managed and regulations insist that each substance must be safe and within certain limits before use (Fennema 1987). Some food additives, however, have been prohibited because of their toxicity

(Abdel-Reheim *et al*. 2014). Many types of toxicological studies are carried out to provide information on types of toxicity such as neurotoxicity and cardiotoxicity and to identify target tissues and organs in order to eliminate potential risks to humans (Pressman *et al*. 2017).

2.2 Potassium Bromate

2.2.1 Physicochemical Properties

Potassium bromate ($KBrO_3$) takes the form of white crystals or powder. It is readily soluble in water and slightly soluble in alcohol (Budavari *et al*. 1989; Lewis 1991), with a melting point of 350 °C and a decomposition temperature of 370 °C. It has a density of 3.27 g/cm^3 and a molecular weight of 167.01 g/mol, and its molecular formula is $KBrO_3$. Bromic acid and potassium salt are synonyms for potassium bromate. Its molecular shape is almost like a trigonal pyramid (Cotton *et al*. 1987), with the three oxygen atoms each at a vertex fanning away from the bromine atom (Fig. 2.1).

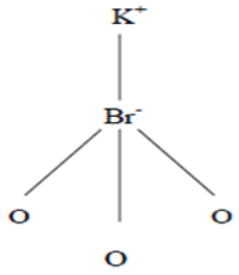

Figure 2.1 Structure of the Potassium Bromate Molecule

This harmful substance can be chemically synthesized by passing bromine into a solution of potassium hydroxide. Potassium bromate is a chemical substance which is added to flour to enhance the action of gluten, Gluten is a protein existing in wheat flour that gives the dough of the bread elasticity through kneading, and hence ferments the dough by retaining the gases produced by yeast. The result of enhancing the gluten is that the bread rises in volume and holds its shape. Therefore, potassium bromate is a flour enhancer which renders dough stronger and softer, so that it rises higher and is more voluminous, and with faster mixing features. In addition, it gives the final bread product an appealing white colour (Chauhan & Jain 2016).

2.2.2 Uses and Action

Potassium bromate ($KBrO_3$) is an oxidizing agent that had been used as a flour treatment agent for over 80 years (Kurokawa *et al*. 1990). Potassium bromate is used widely as an enhancing agent in flour (E924) and a dough strengthener in industrial bakeries (IARC 1986; Chipman *et al*. 1998; Chauhan & Jain 2016). When it is used within prescribed limits of 15-30 ppm, it becomes cooked up during baking operations and leaves no trace in the end product. Ideally, that end product should contain no potassium bromate, which breaks down during baking into potassium bromide (KBr), a harmless by-product (McMinn 2017). However, if the mixture contains higher amounts of potassium bromate, and if the bread is baked at low temperatures or for an indefinite period, quantities of potassium bromate may remain in the bread and will be hazardous to health when consumed (Oyekunle *et al*. 2014; Ben Saad *et al*. 2016; Chauhan & Jain 2016).

A report by the Centre for Science and Environment (CSE) in New Delhi, India, found that about 84% of 38 commonly accessible brands of pre-packaged breads, pav and buns tested positive for potassium bromate and potassium iodate. CSE stated that potassium bromate usually increases dough strength resulting in the higher rising of the bread, whereas potassium iodate is a flour-treatment agent (Chauhan & Jain 2016).

Meanwhile, reports by the CSE and International Agency for Research on Cancer (IARC) define potassium bromate as a chemical in the category of 2B carcinogens (possible human carcinogens) with renal tumour risks to humans at high doses (IARC 1986; Chauhan & Jain 2016). In 2011 the CSE quoted the prescribed level of usage in the Food Product Standards and Additives of potassium bromate in bread set at 50 ppm. The extreme upper limit of this chemical in wheat flour or refined wheat flour for baking is 20 ppm (Chauhan & Jain 2016; Fisher *et al*. 1979). As a result of extensive investigations into the potential risks of $KBrO_3$, this food additive is banned in most countries, including in the European Union, China, United Kingdom, France, Nigeria, and Canada (Fawell & Walker 2006; Chauhan & Jain 2016; Qin *et al*. 2018).

Bromate can be also found as a disinfection by-product of the ozonation of drinking water (Cavanagh *et al*. 1992; Ahmad *et al*. 2012). Bromide in source water is oxidized to bromate via exposure to ozone (Ahmad *et al*. 2015) during disinfection and is frequently detected in tap and bottled water. Ozone has been employed for the disinfection of water because it is more effective than chlorine in eliminating microbes and results in much lower levels of carcinogenic trihalomethanes (THMs) (Park *et al*. 2016). Its currently specified limit in

treated drinking water at a contamination level of 10 mg/l in the USA and Europe (EPA 2001).

Potassium bromate is also widely used in cosmetics, especially hair lotions, as well as in treating barley in beer making, and fish paste and cheese production. Therefore, it can affect human health through occupational and environmental exposure (Ahmad & Mahmood 2014; Ahmad *et al*. 2015).

2.2.3 Metabolism of Potassium Bromate in Animals and Humans

There are no data available for humans, but a lot is known from animal experiments (IARC 1986). Chemically, bromate is very stable in the body and only small amounts change into bromide by glutathione processes in the liver and kidney (Kutom *et al*. 1990). Fisher *et al* (1979) suggested that bromine did not disclose in the lipid tissue of Wistar rats fed for 104 weeks on a diet consisting of 79% bread crumbs made from flour treated with 75 mg/kg potassium bromide.

According to Fujie and others (1988), $KBrO_3$ is quickly absorbed from the gastrointestinal tract after oral administration to rats at a level of 100 mg/kg bw $KBrO_3$. Animals were sacrificed at different times after handling, and bromate was assayed in the stomach, small intestine contents, plasma and bladder urine. Bromate was found to be quickly absorbed and eliminated after 2 h from administration, when it was no longer detected in the plasma, and after 4 h from treatment bromate was no longer found in bladder urine or the small intestine. In addition, after twenty-four hours from the administration of potassium bromate at a dose of ≤ 2.5 mg/kg bw, bromate was not detected in urine. But at

doses of 5-100 mg/kg bw, increased concentrations of bromate were detected in urine after 24 hours and the increase was proportional to dose (Fujie *et al*.1988). Similarly, Tanaka and coworkers reported that high concentrations of bromate are rapidly degraded in blood and tissues in vitro (Tanaka *et al*. 1984). Also, they stated that all of the bromate was degraded after 30 min incubations in red blood cells (RBC), liver and kidney homogenates and about 80 to 90 percent degradation in spleen, stomach and small intestine homogenates. Bull *et al*. (2012) showed there was a substantial reduction of bromate in whole blood. It should be noted that these in vitro assays may not be useful for estimating the in vivo degradation of bromate.

2.3 Toxicity and Safety

Potassium bromate (PB) is an extremely interactive substance which decomposes to inactive bromide during dough fermentation and baking, although this reduction is sometimes incomplete and residual PB persists in the bread. This is considered a source of potential carcinogens to humans (Zhang *et al*. 2011). Therefore, the Joint FAO/WHO Expert Committee on Food Additives (JECFA) (1989) stated that there should be no remaining $KBrO_3$ in food after processing so as to avoid toxicity. Analytical techniques are now available to detect bromate at low levels of parts per billion (ppb) (IARC 1986).

In many countries $KBrO_3$ is still used legally or illegally as a bread improver even though it is related to the development of damage to several organs (EPA 2001; Oloyede & Sunmonu 2009). The acute toxicity of $KBrO_3$ in experimental animals and in humans can result in renal failure, neuropathological disorders,

congestion of the central veins in the hepatocytes, infiltration of the interstitial cells, mucosal dysfunction in the small intestine and cell necrosis in the renal proximal tubule (Kurokawa *et al.* 1990; Akanji *et al.* 2008; Ahmad *et al.* 2011a; Ahmad & Mahmood 2012; Sultan *et al.* 2012). Furthermore, the development of various renal and nonrenal tumors in male and female F344 rats has been associated with the chronic toxicity of $KBrO_3$ (Kurokawa *et al.* 1990; Kurata *et al.* 1992; DeAngelo *el al.* 1998).

Potassium bromate is a strong nephrotoxic agent which has been found to be related to congestion of the central vein in the hepatocytes, further infiltration of the interstitial cells accompanied with acute nephritis in the nephrons and mild mucosal failure in the function of the small intestine, and eventual cell necrosis at selected sites inside the renal proximal tubule (Kurokawa *et al*. 1990; Akanji *et al.* 2008). In addition to effects on the kidney, the structure and function of major tissues such as in the gastrointestinal tract, hepatic cells and blood have also been reported to be affected (Akanji *et al.* 2008; Ahmad *et al.* 2011b; Ahmad & Mahmood 2012; Sultan *et al.* 2012). Induced nephrotoxicity of the bromate can be detected by raised serum creatinine and blood urea nitrogen, which are biomarkers of nephrotoxicity (Khan & Slutana, 2005b; Oloyed & Sunmonu, 2009; Khan *et al*. 2012a).

2.3.1 Toxicity in Humans

Norris (1965) described several cases of accidental poisoning in children between 1.5-3 years of age after the ingestion of 57-133 g of a 2% solution of potassium bromate which caused nausea and vomiting, usually with epigastric

or abdominal pain, and diarrhea and haematemesis occurs in some cases (Parker & Barr 1951). In both children and adults (hairdressers), oliguria and death from renal failure have been reported (Dunsky 1947; Ohashi *et al*. 1971; Gradus *et al*.1984). Bromate may also induce hypotension, depression of the central nervous system, thrombocytopenia (Campbell 2006; Health Canada 2016), and partial hearing loss and complete deafness have also been cited (Matsumoto 1973; Quick *et al*. 1975; Gradus *et al*.1984).

Moreover, the toxic or lethal dose of potassium bromate in humans has not been accurately determined (Kurokawa *et al*. 1990). But a dosage of 500 mg of potassium bromate caused serious symptoms in a 15-month-old child (Quick *et al*. 1975). Oral doses of $KBrO_3$ (185-385 mg/kg b.w) lead to irreversible renal damage and deafness, while lower doses cause gastrointestinal disturbances and abdominal pain (WHO 2005). Around 10 female hairdressers exposed to hair-curling solution for 10-30 years were found to suffer from nonspecific dizziness and vague symptoms and experienced rotational vertigo attacks, nausea, vomiting, headaches and tinnitus (Young *et al*. 2001).

De Vriese *et al*. (1997) described the clinical symptoms of a patient who ingested 300 ml of a cold-wave neutralizer containing 10% potassium bromate. In addition, they reviewed 49 other cases of human exposure reported since 1947. Common symptoms after the ingestion of different solutions containing bromate included nausea, vomiting, abdominal pain and diarrhea shortly after ingestion. Acute renal failure varying from mild to severe anuric form have been indicated in both children and adults. Also 33% of cases of poisoning resulted to death. Severe sensorial hearing loss was recorded within 4-16 hours of ingestion in all adults, but only in a small number of children. Bromate

toxicity is strongly associated with deafness which occurred in 18 of 31 cases, usually within 4-16 hours of exposure (Matsumoto *et al*.1980; Hamada *et al*. 1990).

Several cases of acute poisoning in children have resulted in kidney disorder effects, and a review of the renal toxicity of bromate found that kidney failure occurred in 26 of 31 reported cases (Matsumoto *et al*.1980). It was observed that urea continued for several days or more after exposure to 20 mg $KBrO_3$/ kg (Quick *et al*. 1975). In addition, the histological examination of kidney biopsies of children indicated that the renal effects included the appearance of interstitial edema, interstitial fibrosis, and tubular atrophy (Watanabe *et al*.1992). The length of time required to restore kidney function ranged from 7 days to 5 weeks and, in two cases, renal function was never restored (Hamada *et al*. 1990; Kutom *et al*. 1990).

The treatment of human erythrocytes with $KBrO_3$ under in vitro conditions results in significant increases in protein oxidation, lipid peroxidation, hydrogen peroxide levels, and decreases in total sulphydryl content, which is an indication of oxidative stress in the erythrocytes. The exposure of erythrocytes to $KBrO_3$ also causes decreases in the activity of catalase, glutathione peroxidase, thioredoxin reductase, glucose 6-phosphate dehydrogenase and glutathione reductase (Ahmad *et al*. 2014).

2.3.2 Toxicity in Laboratory Animals

Several studies have evaluated the toxicity of bromate in lab animals following oral exposure in rats, mice and Syrian golden hamsters. Many signs and symptoms of poisoning have been confirmed, including signs of sedation, suppression of locomotion, ataxic gait, polypnoea, hypothermia, diarrhea, lacrimation and piloerection (Fujie *et al*. 1988; Kurokawa *et al*. 1990; Starek & Starek-Swiechowicz 2016). Histological effects have also been indicated in numerous studies, such as degenerative, necrotic, nephropathic and regenerative changes in kidneys which were also reported in F344 rats to which potassium bromate was administered in drinking water (Kurokawa *et al*. 1983).

Several sub-chronic or chronic studies in rats and mice show that the kidney is the primary target organ following long-term oral exposure to bromate, such as when rats received a 13-week exposure to doses of 63 mg $KBrO_3$/kg/day (Kurokawa *et al*. 1990). The kidney is a critical organ significantly affected by exposure to $KBrO_3$. Its administration to the kidney leads to acute renal failure, and several reports have documented nephrotoxicity when administered via subcutaneous or intraperitoneal (Khan & Sultana, 2005a; Bao *et al*. 2008) and oral (Ahmad *et al*. 2012; Khan *et al.* 2012b) routes of administration. For example, when rats were given a single oral dose of $KBrO_3$ (100 mg/kg body weight) and sacrificed at different times after this treatment, the results suggest that a single nephrotoxic dose inhibits brush border membrane enzymes, induces oxidative stress and alters the energy metabolism of the renal system in a reversible manner (Ahmad *et al*. 2012). Furthermore, the oral administration of $KBrO_3$ (20 mg/kg bw) causes a marked increases in the serum levels of creatinine, urobilinogen, blood urea nitrogen (BUN), total bilirubin and direct

bilirubin (Khan *et al*. 2012a). Also, marked histological changes were observed in the cortex of kidneys in $KBrO_3$-treated rats, including tubular degeneration, congestion, and dilation and glomerular injuries.

Various non-neoplastic effects have also been observed, such as the inhibition of body weight gain, significant increases in several serum parameters including BUN and droplets of different sizes, and regenerative changes in the renal tubules. Similar effects were observed in chronic studies of oral bromate exposure (Nakano *et al*. 1989; DeAngelo *et al.* 1998). Non-neoplastic effects reported following long-term exposure include increased BUN and severity of nephropathic change, and degenerative and necrotic kidney lesions such as hyaline casts in the tubular lumen, hyaline droplets, eosinophilic bodies, and brown pigments in the tubular epithelium, as well as urothelial hyperplasia of the transitional epithelium of the renal pelvis. Non-neoplastic effects have also been reported in tissues other than the kidney (Kurokawa *et al*. 1990).

2.4 Acute Toxicity

Many long-term bioassays have examined the effect of potassium bromate administered in drinking water to rodents. It causes renal cell tumours, thyroid follicular carcinomas and mesotheliomas predominantly in rats, although renal cell tumours have also been observed in male mice and Syrian hamsters. Administration of a dose of $KBrO_3$ (0, 250 or 500 mg/l) in drinking water to male and female F344 rats for 110 weeks reduced survival rates and increased body weight in males but not in females, and substantially increased cases of renal cell tumours observed in each treated group in both sexes. Male

rats also showed significantly increased cases of peritoneal mesothelioma (Kurokawa *et al.* 1983). In a further study, male F344 rats were administered potassium bromate at 0, 15, 30, 60, 125, or 500 mg/l in drinking water for 104 weeks. The results showed that the occurrence of adenomas was significantly increased at doses higher than 125 ppm $KBrO_3$, adenocarcinomas were observed only in 3 of 20 (15%) animals at a dose of 500 ppm $KBrO_3$ (Kurokawa *et al.* 1986). Also, treated males showed decreased body weight and survival rates.

2.4.1 Oral Administration

DeAngelo *et al*. (1998) administered to male B6C3F1 mice potassium bromate doses of 0, 0.08, 0.4 or 0.8 g/l (corresponding to 0, 7, 32.6 and 59.9 mg/kg-bw per day as bromate) in drinking water for 100 weeks. Significantly increased renal cell tumour incidence was reported in the 7 mg/kg bw per day group. Although renal tumours were also observed in the 32.6 and 59.9 mg/kg bw per day groups. Takamura *et al*. (1985) conducted a long-term bioassay study using male Syrian hamsters to which were administered potassium bromate doses of 0, 125, 250, 500 or 2000 mg/l (equivalent to 0, 20.1, 40.2, 80.4 and 321.6 mg/kg bw per day as bromate) in drinking water for 89 weeks. Cases of renal cell tumours increased in the 80.4 and 321.6 mg/kg bw per day groups, but the effect was not dose- dependent or statistically significant. The authors stated that the spontaneous incidence of renal cell tumours in Syrian hamster was very low.

Kurokawa *et al* (1990) reported that the potassium bromate LD50 is significantly lower at approximately 160-180 mg/kg body weight for both sexes of Wistar rats which received high doses of $KBrO_3$ of 150, 300, 600, 1250 and 5000 mg/l in drinking water for 13 weeks. All animals given doses greater than 1250 mg/l died after 7 weeks. In addition, significant increases in alanine transaminase (ALT) aspartate transaminase (AST), and blood urea nitrogen (BUN) were found in rats dosed with 600 mg/kg bwt.

In many recent scientific studies, important changes in blood parameters and tissue alterations have been noted, and the most prominent findings are as follows. Potassium bromate induces oxidative stress, as reflected in haematological parameters in European rabbits which orally received a dose of (2.2 mg/kg bw) for 28 days (Dhembar & Dale 2017). Decreases were also shown in red blood corpuscles (RBC), haemoglobin (HB), haematocrit (Hct), and mean corpuscular haemoglobin (MCH), while white blood corpuscles (WBC), platelets (PLT), and plateletcrit (PCT) significantly increased in rabbits treated for 28 days. On the other hand, Achukwu *et al*. (2009) reported that oral administration with graded doses of $KBrO_3$ solution (30, 50, 70, and 90 mg/kg bw) for six weeks led to significant declines in platelet counts but no differences in hematocrit, cellular HB content and leukocyte counts.

In another study, mice were orally administered potassium bromate at the rate of 150 mg/kg body weight daily in single doses for 30, 60 and 120 days. The findings suggest that the chemical significantly reduced the RBC count, Hb%, platelet count and total protein and albumin levels while significantly increasing urea and creatinine levels. Histopathological examination indicated degenerative changes in tubular cells, cellular infiltration, cytoplasm

vacuolation, and tubular dilation with eosinophilic debris, and clear cell cytoplasm was observed (Stuti & D'Souza 2013).

Another study confirmed the side effects of $KBrO_3$ administration in Swiss mice and rats which received high doses of 200 mg/kg or low doses of 100 mg/kg every day. The administration of $KBrO_3$ led to decreased white blood corpuscle (WBC), red blood corpuscle (RBC) and platelet counts in the animals in both the high and low dose groups. Furthermore, altered lipid profiles were also observed in plasma samples of mice in both treated groups, represented by low density lipoprotein (LDL), high density lipoprotein (HDL), and increased levels of cholesterol and LDH in the plasma. Histological investigations showed impaired renal and hepatic histology that was concomitant with increased plasma creatinine levels. Nevertheless, decreased glutathione levels in both renal and hepatic tissue of the mice was also observed (Altoom *et al*. 2018).

2.5 Pathophysiology of $KBrO_3$ in the Kidney: Morphological and Biochemical Effects.

Previous research has shown that $KBrO_3$ is highly toxic with renal effects being among the early symptoms of bromate poisoning (Umemura *et al*. 2004; Khan & Sultana 2005a; Zhang *et al.* 2011). $KBrO_3$ induces nephrotoxicity, which is usually distinguished by increases in serum creatinine and blood urea nitrogen associated with a marked decrease in the tubular transport of electrolytes, calcium and magnesium in addition to glucose, protein and organic anion transport (Gin *et al*. 1999; Khan & Sultana 2005b; Ahmad *et al*. 2012).

$KBrO_3$ causes histopathological changes in the kidney including irregular tubular structure, hyperplasia, hyaline droplet degeneration, additional necrotic changes and stratified squamous cell metaplasia, and studies support the conclusion that tubular necrosis is the major cause of $KBrO_3$ toxicity (Kurokawa *et al*. 1983; Wolf *et al*. 1998; Umemura *et al*. 2006).

The superstructural changes in proximal tubular cells include the loss of the brush border, the diffusion and multimembranous restructuring of lysosomes, formation of myeloid bodies, endoplasmic retinal bulges and the swelling of the mitochondria (Niwa *et al*. 1974; Hamada *et al*. 1990), resulting in the total disintegration and disabling of cellular organelles along with cellular necrosis (Kurokawa *et al*. 1983; Hamada *et al*. 1990). The passive interaction of $KBrO_3$ with one or more critical processes within the cell leads to impaired renal function and the disruption of the functioning of organelles such as mitochondria, lysosomes, microsomes, BBM and the basal membrane (Gin *et al*. 1999, Umemura *et al*. 2004; Ahmad *et al*. 2012).

2.6 Pathophysiology of $KBrO_3$ in Other Tissues

Although the kidney is an important organ in the body, $KBrO_3$ has also been proven to accumulate in various other tissues, including the intestines, liver, heart and ear, causing multiple harmful effects (Kurokawa *et al*. 1990; Campbell 2006; Oloyede & Sunmonu, 2009; Josiah *et al*. 2011). A dose of $KBrO_3$ of 20 mg/Kg/day twice daily for 4 weeks caused damage to the liver of male albino rats, such as distortion, degeneration, and cellular infiltration, altering the tissue architecture of the liver as well as its biochemical properties

and inducing hepatotoxicity (Bayomy *et al*. 2016). Similar results have been documented in Wistar rats to which 100 mg/kg of $KBrO_3$ was orally administered for 2 weeks (Awoniran & Adeyemi 2018). It has been reported by El-Sokkary (2000) and Khan *et al*. (2003) that $KBrO_3$ causes a significant increase in lipid peroxidation of the liver tissue due to free radical damage in the degenerative livers of rats. A significant deficiency of antioxidant enzymes and an increase in lipid peroxidation were observed when rats were treated with $KBrO_3$ at a dose of 100 mg/kg bw intraperitoneally. Furthermore, $KBrO_3$ is known to interact with pulmonary tissues and to contribute to causing malignant tumours associated with bromates in the lung (Kurokawa *et al*. 1990).

The treatment of rats with 20 mg/kg bw of $KBrO_3$ for 4 weeks has been found to significantly decrease the hormonal levels of testosterone, luteinizing hormone and follicle-stimulating hormone while markedly increasing testis weight and the secretion of estradiol and prolactin (Khan *et al*. 2012b). The degeneration of the seminiferous tubules, loss of germ cells, abnormality of the germinative epithelium, interruption in meiosis, and sperm with abnormal shapes were also visible after $KBrO_3$ administration.

$KBrO_3$, affects the intestinal morphology and induces adenomas of the small intestine in experimental animals (Kurokawa *et al*. 1990). The exposure of the gastric mucosa to damaging factors results in the disturbance of protective mechanisms and the disruption of the mucosal barrier (Kwiecien *et al*. 2002). As a by-product of the disinfection of water, bromate alters the intestinal microbial flora in rats and may results in irreversible physiological consequences (George *et al*. 2004). The administration of $KBrO_3$ (at 100 mg/kg bw) induces DNA degradation in the intestinal tissue of treated rats, leading to

extensive intestinal damage such as to the mucosal cells and especially the membrane, so that the lumen becomes full of debris and the intestinal villi lose their form (Ahmad *et al.* 2015). Also, the activities of intestinal brush border membrane enzymes decreased while those of antioxidant defence and carbohydrate metabolism were also severely altered (Ahmad *et al*. 2012; 2013).

Exposure to $KBrO_3$ results in multiple organ toxicity, and numerous toxicity studies have been summarized in a toxicology review of bromates by the EPA (2001) and by Starek-Swiechowicz (2016).

2.7 $KBrO_3$ Oxidative Stress

Although a number of studies have documented the toxic effects of bromate, its modus operandi remains unclear. It has been proposed that reactive oxygen species (ROSs) and oxidation stress (OS) may contribute to this toxicity. Once the bromate enters the cell, it is reduced to the more stable bromide ion. This process is accompanied by the production of free radicals and ROSs that may contribute to renal carcinogenicity and tissue toxicity (Sai *et al*. 1994). The formation of ROSs following the reduction of bromate to bromide as explained earlier (Rahman *et al*. 1999; Zhang *et al*. 2010).

Furthermore, $KBrO_3$ mediates toxicological effects via the induction of oxidative stress, causing the modification of DNA bases, LPO and protein oxidation in the kidneys (Karbownik *et al*. 2006). Many modified bases are produced from continuous oxidative stresses. 8-oxodG, a base lesion in DNA that is usually removed by base excision repair (McDorman *et al*. 2005).

Moreover, 8-oxodG has also extensively been used as an indicator of oxidative DNA damage and various diseases (Cellai *et al.* 2020). $KBrO_3$ induces the production of the superoxide anion (O_2), hydrogen peroxide (H_2O_2), and the hydroxyl radical (OH) in renal tissue (Sai *et al*. 1994; Murata *et al*. 2001; Khan & Sultanah 2005b). The reduction in total SH content, an increase in LPO and the formation of 8-hydroxy-2-dioxiguanosin (8-oxodG) have also been reported in the proximal neural tube of isolated rat (Sai *et al*. 1994).

Umemura *et al.* (2004) have suggested that $KBrO_3$ in drinking water may have a carcinogenic effect through production of bromine radicals (Br) or oxide radicals (BrO, BrO_2, etc) are the species directly responsible for the damage to cellular and cell-free DNA.

2.8 Chemoprotective Effects of Antioxidants

Many attempts have been made to protect or promote recovery from experimental toxicity. These studies are useful not only for the amount of protection they may offer, but also for insights into the working mechanisms of toxic substances, many of which act at the instigation of the OS. The affected tissues/cells have several ways to mitigate the effects of the OS, either by repairing the oxidative damage or by directly minimizing its occurrence by enzymatic and non-enzymatic means. Antioxidants (AOs) are capable of attenuating free radicals and ROS-induced oxidative damage (Ahmad 2013).

Additionally, human exposure to $KBrO_3$ is a common occurrence in both occupational and environmental situations. The increased use of $KBrO_3$, in the

food industry and the generation of bromate ions during the water purification process have been associated with risks to human health. The effective management of this compound is necessary to avoid adverse health effects on the population, and possible therapeutic approaches to reverse the toxicity caused by $KBrO_3$ would be of immense clinical significance in increasing safety and protection against exposure to bromate (Ahmad 2013).

The health problems that an affected individual may face are greatly influenced by eating unhealthy food, which results in many problems. Unhealthy food could be produced which increases exposure to several chemicals used in industrial food manufacture. For example, the use of food additives such as $KBrO_3$ has increased enormously in recent years. This additive has been shown to cause various health disorders and histopathological lesions in treated animals (Moubarak *et al*. 2020; Shanmugavel *et al*. 2020). Unfortunately, $KBrO_3$ is added to bread dough by some bakeries in levels above its permitted limits, and bromate can also be found as a by-product of the disinfection of drinking and waste- water. Consequently, it is necessary to identify potential therapeutic agents which may ameliorate its damaging effects (Ahmad 2013).

As $KBrO_3$ is primarily an oxidizing agent, lipid peroxidation (LPO) and free radicals generated from bromate appears to be the basis of bromate-induced kidney carcinogenesis and other toxic effects (Chipman *et al*. 1998; Kawanishi & Murata 2006). Several studies have shown that protective agents such as from plant ingredients are sources of diverse antioxidant properties which can protect the body against bromate-induced toxicity (Khan *et al*. 2001). Some of these plants have been scientifically evaluated for their potential protective effects against $KBrO_3$ toxicity. For example, *Curcuma longa* can be a potent

chemopreventive agent against $KBrO_3$-induced liver injury (Awoniran & Adeyemi 2018). Also, Khan (2017*)* suggested that a methanol extract of *Launaea procumbens* can protect thyroid tissue against oxidative damage, possibly via the antioxidant effects of its bioactive compounds. Curcumin could potentially be used as a protective agent against the carcinogenicity of $KBrO_3$ due to its antioxidant, anti-inflammatory, and free-radical scavenging activity (Obaidi *et al*. 2018). The leaves of some plants such as *Thaumatococcus daniellii* have shown a protective property for the testicular functions of rats administered with $KBrO_3$ (Nwonuma *et al.* 2016).

Although the toxicity and other toxic effects of $KBrO_3$ have been extensively investigated, only limited studies have been carried out to find protection and prevent $KBrO_3$ toxicity. In animal experiments, several approaches have been employed in attempting to reduce the toxicity of $KBrO_3$ (Ahmad 2013).

In addition, numerous medicinal plants or their derivatives have already been investigated for their antioxidant properties and their possible role in the prevention of $KBrO_3$ nephrotoxicity. Some of these exhibit protective effects against $KBrO_3$-induced renal damage, such as *Tephrosia purpurea* (Khan *et al*. 2001), *Nigella sativa* (Khan & Sultana 2005a), *Nymphaea alba* (Khan & Sultana 2005b), bilberry (Bao *et al*. 2008), *Hibiscus sabdariffa* (Josiah *et al*. 2010), rutin (Khan *et al*. 2012a), taurine (Ahmad *et al*. 2013) and gum acacia (Ali *et al.* 2018).

2.8.1 Antioxidants

An antioxidant (AO) is a molecule which is able to inhibit the oxidation of other molecules by removing free radical intermediates and preventing other oxidation reactions. AO ingredients are widely used in dietary supplements and have been investigated for the prevention of broad spectrum diseases such as cancer, coronary heart disease and even altitude disease (Gulcin 2020).

Antioxidants can be categorized in many ways, based for example on their origin (natural or synthetic), activity (enzymatic and nonenzymatic), structure (flavonoids, polyphenols, etc.), or solubility (water-soluble or lipid-soluble). (Vertuani *et al*. 2004). Examples of natural antioxidants are vitamin E and ascorbic acid (vitamin C), while examples of synthetic antioxidants are butylated hydroxytoluene(BHT), butylated hydroxyanisole (BHA), octyl gallate (OG), and ethoxyquin. The enzymatic defence system consists mainly of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx), and glutathione reductase, while non-enzyme molecules include gluthathione, α-tocopherol (vitamin E), ascorbic acid, uric acid, bilirubin and melatonin (Atta *et al*. 2017; Neha *et al*. 2019). Numerous other antioxidants are abundant in fruits and vegetables as well as other foods such as nuts, wholegrains and some meats, poultry and fish (Sarker & Oba 2020).

A number of investigations have demonstrated that a diet supplemented with antioxidants can have profoundly beneficial health effects against various diseases, such as asthma and inflammatory disease, and also immunological disorders of the kidney and intestines (Valko *et al*. 2007). Some of the AO

content in food is incorporated into cellular membranes, and especially plasma membranes which affect the fluidity and regulation of various enzymes and transport proteins. This changes the functional aspects of different organs, especially the kidney, liver and intestines, leading to positive outcomes related to health and disease issues (Halliwell 1996; Young & Woodside 2001; Seifried *et al*. 2007). Generally, a water-soluble AO reacts with oxidants in the cell cytoplasm and blood plasma, whereas lipid-soluble AOs protect cell membranes from LPO (McDermott 2000).

Considering the profoundly helpful health effects of AOs against different pathologies, the work described in this thesis was carried out to study protection against or prevention of $KBrO_3$ toxicity by the use of dietary AOs. The most important and effective source of antioxidant found so far is the onion (*Allium cepa* L), which is one of the most economically important agricultural crops grown in the world (Lanzotti 2006). It has been used in herbal medicine for thousands of years (Gokce *et al*. 2010), and exhibits a wide range of biological and medicinal properties including immunomodulatory, hepatoprotective, antioxidant, antimutagenic and anticarcinogenic effects (Corzo-Martínez *et al*. 2007; Pareek *et al*. 2017). Due to the plant's substantial flavonoid content, the valuable nutritious and antioxidants presence in onion, numerous studies have been carried out to investigate its role in preventing or ameliorate hepatotoxicity, nephrotoxicity and various types of diseases such as cardiovascular disease and cancer (Nile & Park 2013).

2.8.2 *Allium cepa* and Phytochemistry

Onion is an important crop in the north-western region of Libya (Fennir 2014) and Libyans are among those who consumes the highest amounts of onion (FAO 2015). It belongs to the family Alliaceae, and the different varieties such as red, yellow, white, and green onions each have their own unique flavour (Slimestad *et al*. 2007). It has considerable economic importance as one of the most valuable vegetable crops, and it also has significant medicinal, nutritional, and functional value (FAO 2009; Nile & Park 2013). Recently, several review articles have been published which summarize the nutritional composition and phytochemical analyses of onions (Lanzotti 2006; Pareek *et al*. 2017; Teshika *et al*. 2018).

To summarize, onions contain three main chemicals that are believed to have beneficial effects on human health, which are flavonoids, organosulphur compounds and fructans (Pareek *et al*. 2017). Flavonoids are responsible for a variety of pharmacological activities (Mahomoodally *et al*. 2005; Rezaei-Sadabady *et al*. 2016), and hence the potential health benefits arising from the antioxidant activities of these polyphenolic compounds are of great interest (Kumar & Pandey 2013).

Onions have been reported to be a major dietary source of flavonoids, and particularly of flavonols and anthocyanins (Ko *et al*. 2014), the latter being found only in red onions (Pérez-Gregorio *et al*. 2010). Red onion has higher radical scavenging activities than yellow onion (Lanzotti 2006), containing 415-1917 mg of flavonols per kilogram of fresh onion (Slimestad *et al*. 2007).

Sixteen different flavonols have been identified and quercetin derivatives are considered the most important in the red onion cultivar, including 3,4'-*O*-diglucoside and 4'-*O*-monoglucoside (Perez-Gregorio *et al.* 2011). Other phenolics have also been detected, included catechin, epigallocatechin gallate, myricetin, resveratrol, naringenin. apigenin and kaempferol (Kim *et al*. 2013; Hur *et al*. 2013). At least 25 different anthocyanins found in red onions have been reported (Slimestad *et al*. 2007), such as cyanidin 3-glucoside, cyanidin 3-laminaribioside, cyanidin 3-(6''-malonylglucoside), and cyanidin 3-malonylaminaribioside (Pérez-Gregorio *et al*. 2010).

In addition, onion is rich in sulphur compounds such as cysteine, dipropyl disulphide, dipropyl sulphide, dipropyl trisulphide, allyl propyl disulphide (Corzo-Martínez *et al*. 2007). The organosulphur compounds in *A. cepa* have been well documented in many reviews (Breu 1996; Rose *et al*. 2005; Suleria *et al*. 2015). Yamazaki *et al.* (2011) identified 11 sulphur-containing flavour precursors in onion, such as S-alk(en)ylL-cysteine derivatives, methiin, alliin, isoalliin, cycloal- liin, deoxyalliin and N-(gamma-glutamyl)-S-methyl-L-cysteine.

Furthermore, recent studies reveal that the nutritional composition of onion is very complex, due to the presence of several compounds. Among these are saponins, fructans, proteins, amino acids, carbohydrate, fat and minerals as well as various vitamins including thiamin, riboflavin, niacin, folate, vitamin E and ascorbic acid (Ugwoke & Ezugwe 2010; Suleria *et al*. 2015; Ogbonna *et al.* 2016).

2.8.3 Antioxidant Activity of *Allium cepa*

The compounds found in onions are involved in significant biological activities significant for the maintenance of health, including antioxidant, antimicrobial, anti-inflammatory, antithrombotic, anticancer, antidiabetic and anti-allergic effects due to the varieties of bioactive phytochemicals present. However, the present research focuses on the antioxidant activity of onions. Red onions are bountiful in phytochemical content, with huge amounts of flavonoids and organosulphur compounds that have antioxidant properties (Vlase *et al*. 2013; Pareek *et al*. 2017). These antioxidant compounds have radical scavenging potential for preventing or slowing down oxidation by free radicals before they act to damage the cells and tissues of the human body (Kumar & Pandey 2013). In their comparison of the antioxidant capacity and phenolic content of green, white, red and violet onions, Prakash *et al.* (2007) found that red onions have the highest quercetin and kaempherol content. Similar results have been reported by Gokce *et al*. (2010). Quercetin and kaempferol are both considered effective antioxidants against the non-enzymatic lipid peroxidation and oxidation of low-density lipoproteins (LDL) (Gulsen *et al*. 2007). In addition, kaempferol is considered to be a strong antioxidant because its higher concentration accelerates the formation of antioxidant enzymes like catalase and superoxide dismutase (Calderon-Montaño *et al*. 2011).

The antioxidant activities of the methanol extracts of selected varieties and parts of garlic and onion have been investigated by the inhibition of lipid peroxidation induced by tert-butyl hydroperoxide in isolated rat hepatocytes (Nuutila *et al*. 2003). It was found that onions had higher radical scavenging activities than garlic, red onion being more active than yellow onion. The

extracts from onion skin exhibited the highest activity. Also, the radical scavenging activity correlated positively with the total phenolics content of the extract.

An aqueous extract of *A. cepa* was reported to significantly reduce blood glucose levels in alloxan-induced rabbit diabetes (Ogunmodede *et al*. 2012). Also, *A. cepa* vastly improved the enzymatic antioxidants include superoxide dismutase, catalase and glutathione peroxidase. Also, it increased malondialdehyde which is a product of lipid peroxidation. So, *A. cepa* may be effectively ameliorated diabetes-induced hepatotoxicity and harmful effect of bromate on hematological and biochemical parameters. Chyun *et al*. (2013) investigated the hepatoprotective effects of *A. cepa* extract on liver injuries induced by CCl_4 in rats. Their results indicated that the extract protects liver injuries by reducing lipid peroxidation and enhancing antioxidant properties as well as improving blood cholesterol profiles. Similar results were obtained when rats were given a single dose of 100 mg/kg b w of aluminum chloride ($AlCl_3$) and 1 mL/0.1 kg body weight of *A. cepa* extract (AcE) for four weeks (Ige *et al.* 2017). AcE was found to ameliorate the hepatotoxic effect of the administration of $AlCl_3$ through the regulation of the hepatic oxidant/antioxidant system.

The ethyl acetate fraction of an aqueous extract of onion flesh and peel could be effective in improving cognitive function through acetylcholinesterase (AChE) inhibition and antioxidant activity in mice brains (Park *et al*. 2015). Due to the content of phenolics such as isoquercetin, quercetin, and isorhamnetin 4'-glucoside, onion flesh and peel may be used as natural potential resources in

attempting to reduce the learning and memory dysfunctions caused by ageing and neurodegenerative disease.

Abdel Gadir *et al*. (2007) suggested that a combination of 2% onion, 2% garlic or 1 ppm selenite in the rat's diet is chemoprotective and suppresses the development of the hepatonephrotoxicity induced by $KBrO_3$, which was demonstrated by the reduction of nephrotoxicity and hepatotoxicity, along with improvements in growth, pathological, serum-biochemical and hematological parameters.

Several experimental studies have reported the beneficial effect of *Allium cepa* on cadmium-induced hepatic damage (Ige *et al.* 2011) and renal dysfunction (Suru 2008; Ige *et al.* 2009) in male rats. Furthermore, *A. cepa* mitigates the toxic effects of $AlCl_3$ and improves the antioxidant status and sperm quality of male rats (Ige & Akhigbe 2012). Similarly, extracts of onion and garlic could provide protection against cadmium-induced testicular oxidative damage and spermiotoxicity by reducing lipid peroxidation and increasing antioxidant activity in rats (Ola-Mudathir *et al*. 2008).

Lee *et al*. (2012) argued that red onion may enhance antioxidant defense mechanisms by the induction of the activity of plasma superoxide dismutase (SOD) and glutathione peroxidase (GPx) and decreased lipid peroxidation in the liver. Therefore, red onion may exert important protective effects against oxidative stress-related disease.

2.9 Kidney Anatomy and Physiology

The kidney is a vital organ that plays a crucial role in health and disease. The essential function of the kidney is to maintain the total liquid volume of the body, and also its composition and pH within the physiologically appropriate range. This is achieved by the collective activity of thousands of the structural and functional units of the kidney known as nephrons. A nephron is composed of the glomerulus, which has an extended tubular structure (Pfaller 2012). The rat kidney contains 30,000-35,000 nephrons, while the human kidney is composed of approximately 1 million of them (Delaney *et al*. 2018). All of these nephrons help in the maintenance of kidney function by the selective reabsorption of different ions and solutes (Hu *et al*. 2019)

2.9.1 The Structure and Function of the Kidney

The structure of the kidney in most mammals seems to be very homogeneous (Costantini & Kopan 2010). The rodent kidney is located retroperitoneally, often surrounded by white fatty tissue with pockets of brown fatty tissue inside the pelvis and sometimes surrounding the renal capsule (Figure 2.2). Both kidneys are located in the middle of the upper abdomen. The kidney structure has a bilateral structure, is unipapillate and smooth and is larger in the male than in the female.

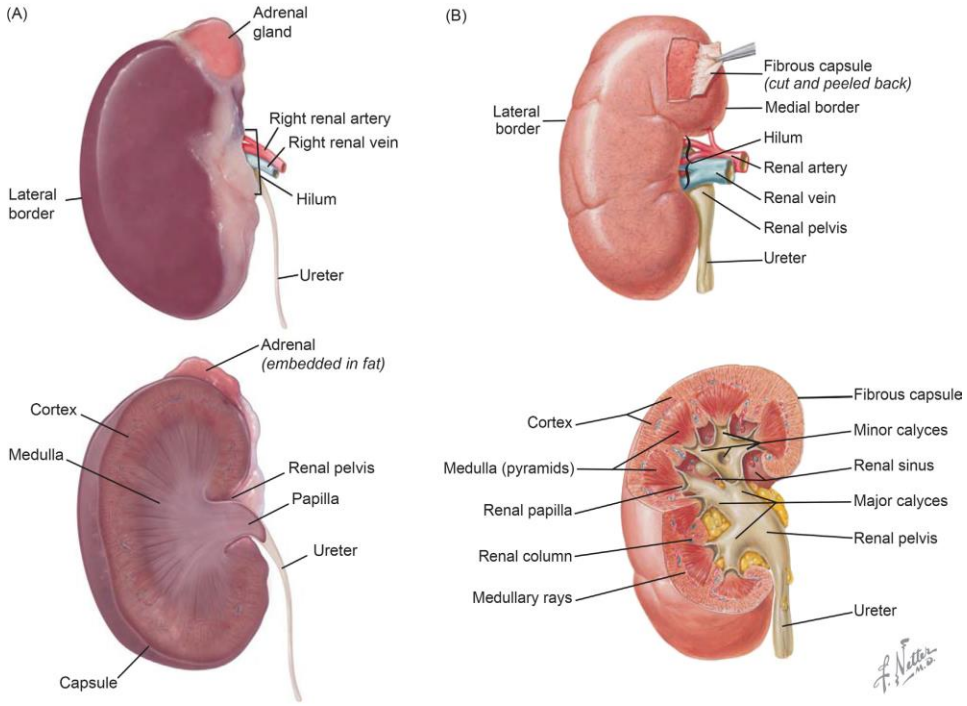

Figure 2.2 Anatomical Diagram Showing the Structure of the Kidney: (A) Rodent (Mouse); (B) Human

Histologically, the kidney is composed of two regions: the outer cortex and the inner medulla. The cortex and medulla are arranged into a more pyramidal shape called renal pyramids, and the apex of each pyramid is called the renal papilla (Sands & Verlander 2010). The main functional unit of the kidney is the nephron. Each nephron consists of a glomerulus surrounded by the Bowman's space, a proximal tubule (convoluted and straight), descending and ascending loops of Henle, and a convoluted distal tubule (Al-Samawy 2012; Brown *et al.* 2016). Nephrons are classified by site in the cortex: superficial or juxtamedullary in mice; and superficial, mid-cortical, or juxtamedullary in rats and humans (Sands & Verlander 2010).

The main portion of the nephron is the renal corpuscle (Malpighian corpuscle). It consists of a renal glomerulus and Bowman's capsule. The corpuscle has two

poles: the vascular and urinary poles. At the vascular pole, the afferent arteriole enters, and the efferent arteriole leaves the corpuscle. The urinary pole is the beginning of the proximal convoluted tubule, and it is continuous with the Bowman's space. The glomerulus is a tuft of epithelial cells (fenestrated endothelium) and capillaries where blood is filtered into urine filtrate (Maurya *et al*. 2018). Bowman's capsule is a double-walled envelope that holds the glomerulus. The parietal layer of the capsule is composed of simple squamous epithelial cells that are continuous with the cuboidal cells of the proximal tubules. Meanwhile the visceral layer is made up of podocytes whose feet surround the glomerulus capillaries (Krinke 2000).

The medulla of the kidney is subdivided into an outer and an inner zone. The inner zone forms the papilla and the outer zone can be further subdivided into inner and outer stripes. The outer stripe of the outer zone extends into the cortex, forming so-called medullary rays. These anatomic subdivisions comprise specific parts of the nephron (Seely *et al*. 2018).

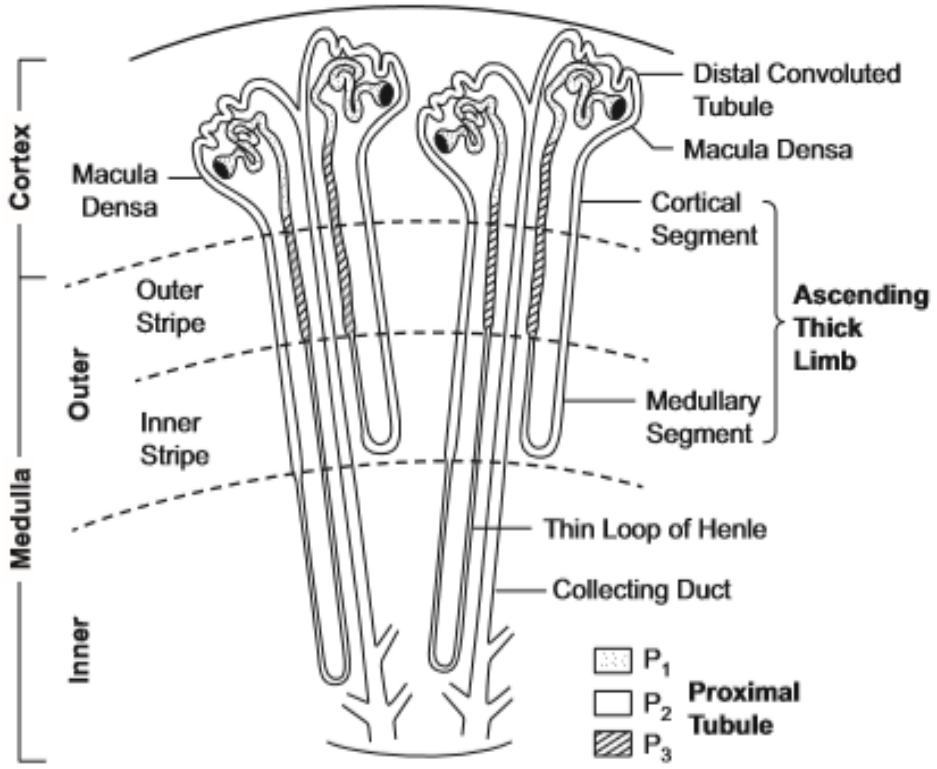

Figure 2.3. Schematic showing location of nephron segments within renal medulla and cortex. (adopted from Montgomery & Seely 1990)

The proximal tubule exhibits a small, uneven lumen and a single layer of cuboidal cells with eosinophilic, granular cytoplasm. A brush border lines the cells. The proximal tubule plays an important role in maintaining homeostasis through the reabsorption of water, sodium and chloride ions, and also by a number of absorptive and secretory mechanisms (Zhang 1999). The distal convoluted tubule is the second type of tubule in the cortex, which is different from the proximal tubule in that the cells of its lining are of the cuboidal type of epithelia with round and large nuclei. Distal convoluted tubules exist within the medullary rays, where they empty into collecting ducts and drain to the medulla. They can be differentiated histologically due to their pale staining and

their lack of brush borders (Al-Samawy 2012). The medulla of each kidney is formed from collecting tubules, and the thick and thin parts of the loops of Henle, where the thin limb has a distinct rounded lumen. The Henle's loop consists of a thick descending limb which is very similar in structure to the proximal convoluted tubule, a thin descending limb, a thin ascending limb, and a thick ascending limb which is very similar in structure to the distal convoluted tubule (Krinke 2000; Seely *et al*. 2018).

2.10 Intestinal Anatomy and Physiology

The intestines are the most active tissue in the body in mammals. The intestinal mucosa is the major site for the digestion and absorption of nutrients, water and electrolytes. Most absorption occurs in the duodenum and the proximal half of the jejunum (DeSesso & Jacobson 2001). Many complex compounds are analyzed as pass through the small intestine into simple compounds that cross the intestinal epithelium before reaching the different organs of the body. Consequently, the intestinal epithelium not only regulates various absorption and excretion processes, but also processes the substances which pass through it (Ahmad 2013).

2.10.1 The Structure and Function of the Intestine

According to many publications describing the rats gastrointestinal structure is histologically very similar to the human's. Therefore, rats are used as subjects in the experimental modelling of pathological conditions of the digestive system (DeSesso & Jacobson 2001; Hryn *et al*. 2018, 2019). The

small intestine of albino rats is a transitional canal connecting the stomach and caecum which are located close to each other (Fig 2.4), It is about one metre long, and in terms of its ratio to body weight is significantly higher than the corresponding part in humans (Hryn *et al*. 2019).

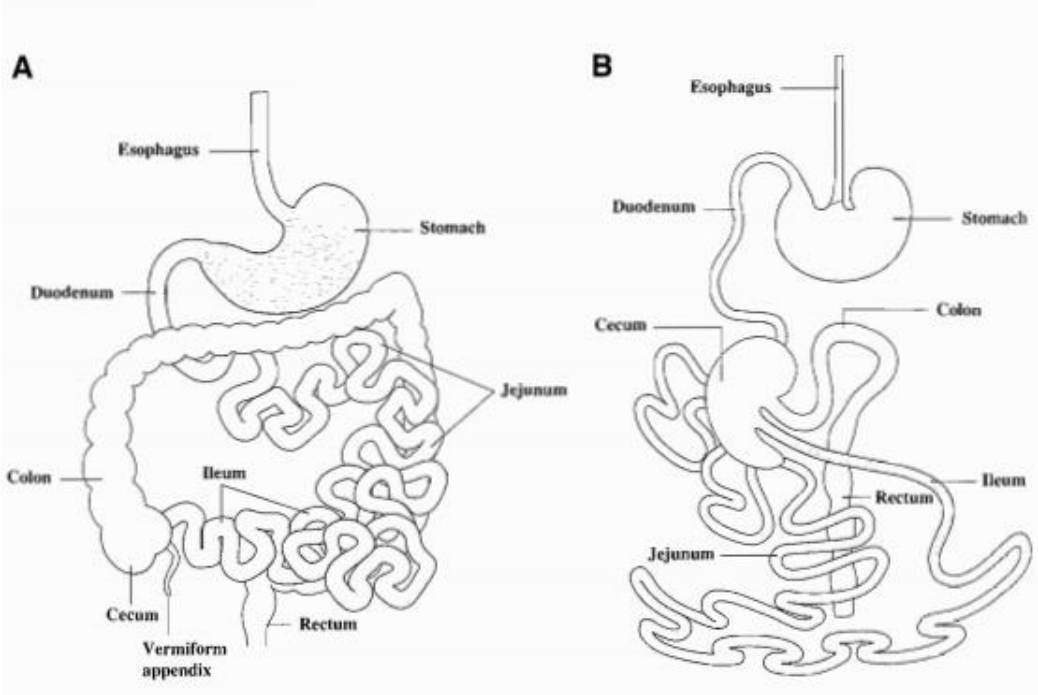

Figure 2.4. Overview of digestive tracts of (A) humans and (B rats). The small intestine is divided into the duodenum (the short initial part), jejunum and ileum, which empties into the large intestine. (Source: DeSesso & Jacobson 2001.)

The rats were chosen as animal subjects for the current study. The anatomy and functioning of digestive canal in humans is in considerable agreement with rat species except for some distinct differences. Morphologically, the small intestine is made up of the duodenum, the jejunum and the ileum (Reed & Wickham 2009; Rao &Wang 2010). Histologically, it has all the typical tunics or layers, including mucosa, submucosa, muscularis externa, and

serosa/adventitia. The mucosa is the more significant layer for absorption, secretion, and defense against orally ingested xenobiotics, as it is exposed to food and other substances in the intestinal lumen (Rao & Wang 2010; Collins & Badireddy, 2019). The surface area of the mucous membrane of the intestine is increased by circular folds known as villi, which are finger-like projections 0.5-1.5 mm in length. The villi are filled with a layer of epithelial cells and also contain networks of capillaries and lymphatic vessels (lacteals). Furthermore, the free edges of the cells of the epithelium of the villi are divided into minute microvilli. The microvilli maximize nutrient digestion and absorption by greatly increasing the surface area (Treuting *et al*. 2018). Several cell types are present in the intestinal epithelium, including absorptive cells, goblet (mucous) cells, enteroendocrine cells and paneth cells (Nakatsuji 2018).

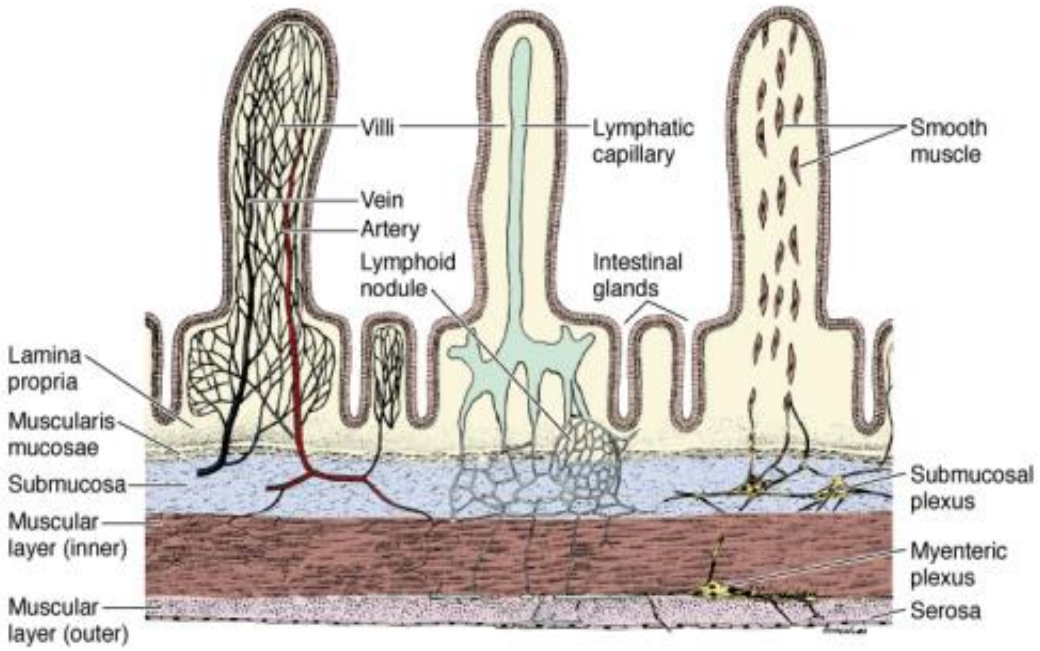

Figure 2.5: Schematic cross –section of the small intestine (adopted from Ganong 2010).

The small intestine is divided into three sequential structural and functional parts as shown in Figure 2.3:

A. Duodenum: The rat duodenum arises from the stomach and runs dorsally forming a wide loop with descending, transverse and ascending sections before continuing as the jejunum. The duodenum is approximately 95–100 mm long, and it is covered in part by the mesothelium of the visceral peritoneum.

The mucosa is arranged as a series of shallow folds that run transversely to the long axis of the gut. The rat has no folds of Kerckring (plicae circulares) (Hosoyamada & Sakai. 2005). The mucosa projects into the lumen of the duodenum as leaf-shaped villi, which greatly increase the surface area of the gut. Villi are found throughout the small intestine (Foligne *et al*. 2001). The submucosa at the first part of the duodenum is characterized by the presence of Brunner's glands which are branching, coiled, tubular glands (Nakatsuji *et al*. 2018) and are the only submucosal glands in the rat intestine. Lymphatic follicles (Peyer's patches) are present in the submucosa from around the middle of the small intestine, whereas in man they are limited to the ileum (Maynard & Downes 2019).

B. Jejunum: The jejunum is the longest segment of the gastrointestinal tube. The shape of the villi is ridge-like, which are mucosa projections with abundant intestinal glands (crypts of Lieberkuhn) that open into pits between the bases of the villi and penetrate the mucosa as far as the muscularis (Nzalak *et al*. 2015). The submucosa contains the autonomic plexus of Meissner and a plexus of blood and lymphatic vessels. The muscularis consists of an outer longitudinal layer and a thicker inner circular layer of smooth muscle separated by the

myenteric (Auerbach's) plexuses located in the connective tissue (Maynard & Downes 2019).

C. Ileum: The ileum has villi and intestinal glands which are part of the mucosa. The villi are short and cylindrical (finger-like). There are Peyer's patches in the submucosa, while the tunica muscularis has longitudinal muscles. The goblet cells are more numerous in the ileum (Maynard & Downes. 2019). However, the jejunum and ileum are similar in the rat. The ileum is a distal continuation of the jejunum with the same diameter, although the ileum lacks folds (Nzalak *et al*. 2015).

CHAPTER III

3 MATERIALS AND METHODS

3.1 Experimental Animals

Albino Wistar rats (12–13 weeks old, weighing 150–200g each) were obtained from the Animal House of the National Medical Research Centre in the city of Al-Zawia, Libya. 30 male Wistar rats were randomly allocated to 5 groups of 6, where each group was subjected to one of the treatments described in section 3.3 below. They were housed in standard clear plastic cages and maintained under standard animal housing conditions. They were allowed to acclimatize for 2 weeks before the experiments were conducted, and were given free access to standard laboratory food and water.

3.2 Preparation of *Allium cepa* Extract (AcE)

Fresh onion, *Allium cepa*, was obtained from the local market in Al-Zawia. The bulbs were rinsed thoroughly with water and cut into small pieces. Extract (AcE) was prepared daily following the procedures used in previous studies (Suru 2008; Jaiswal *et al*. 2013). In brief, about 100 ml of distilled water per 100g of onion were crushed in a mixing machine, and the resulting paste was filtered through a fine cloth or filter paper. The filtrate/juice was used on the day of preparation.

3.3 Potassium Bromate ($KBrO_3$)

Potassium bromate was purchased in powder form from the Sigma-Aldrich Company. Stock of potassium bromate ($KBrO_3$) solution was prepared by dissolving 25 g of $KBrO_3$ in 1000 ml distilled water, followed by storage at 4°C (Dimkpa *et al*. 2013). Bromate working solutions were prepared by diluting the stock solution to the required concentrations as needed.

3.4 Experimental Design

After the acclimatization period, the experimental groups received regular rat feed and water, and were grown for the four-week period of the experiment. The groups were each subjected to one of the following treatments:

Group 1 (control): Rats were fed with a normal diet and drinking water only.

Group 2 ($KBrO_3$): Rats were given $KBrO_3$ by oral gavage (100 mg/kg bw) at days 24 and 27 of the experiment.

Group 3 (AcE + $KBrO_3$): Rats were treated with 1 mL/100 g bw/day of AcE extract via gavage for 28 days, and were given $KBrO_3$ (100 mg/kg b w) at days 24 and 27 of the experiment.

Group 4 ($KBrO_3$): rats were given $KBrO_3$ by oral gavage in doses of 50 mg/kg bw twice per week.

Group 5 ($KBrO_3$ + AcE): rats were treated with (1 mL/100 g bw/day) of AcE extract via gavage for 28 days, and were given $KBrO_3$ (50 mg/kg bw) twice a week.

Group 6 ($KBrO_3$): rats were given $KBrO_3$ by oral gavage in doses of 30 mg/kg bw/day) for 4 weeks.

3.5 Body and Organ Weights

The body weights of the rats were recorded throughout the experiment. Change in body weight was calculated by subtracting the final body weight from the initial body weight. Additionally, the liver, kidneys, testes and lungs were weighed after sacrifice.

3.6 Sample Collection and Biochemical Analysis

At the end of the 28-day experimental period, the rats were fasted overnight and sacrificed 48h after the last treatment. Rats were sacrificed under chloroform anaesthesia and quickly dissected. Blood samples were collected directly from the animals by heart puncturing prior to the excision of the required organs. Blood was withdrawn from the heart into 2 tubes: heparinized tubes for the complete blood count (CBC), and plain tubes for biochemical tests. The levels of alanine transaminase (ALT), aspartate transaminase (AST) and alkaline phosphatase (ALP), as well as urea, uric acid and creatinine, were determined.

3.7 Tissue Processing

Portions of the kidney and duodenum were immediately fixed in 10% neutral buffered formalin for histological study. Tissues were dehydrated through a series of ethanol solutions, cleared in xylene, embedded in paraffin and routinely processed for histological analysis (Slaoui & Fiette 2011). Sections of 5 µm thickness were cut using a rotary microtome and stained with haematoxylin-eosin .

3.8 Haematoxylin-Eosin Staining Procedure

The plan for Haematoxylin-Eosin staining procedure shows the arrangement for de-waxing with xylene, dehydration, clearing and staining prior to mounting. The slides have to go through several consecutive steps as listed in Figure 3.1:

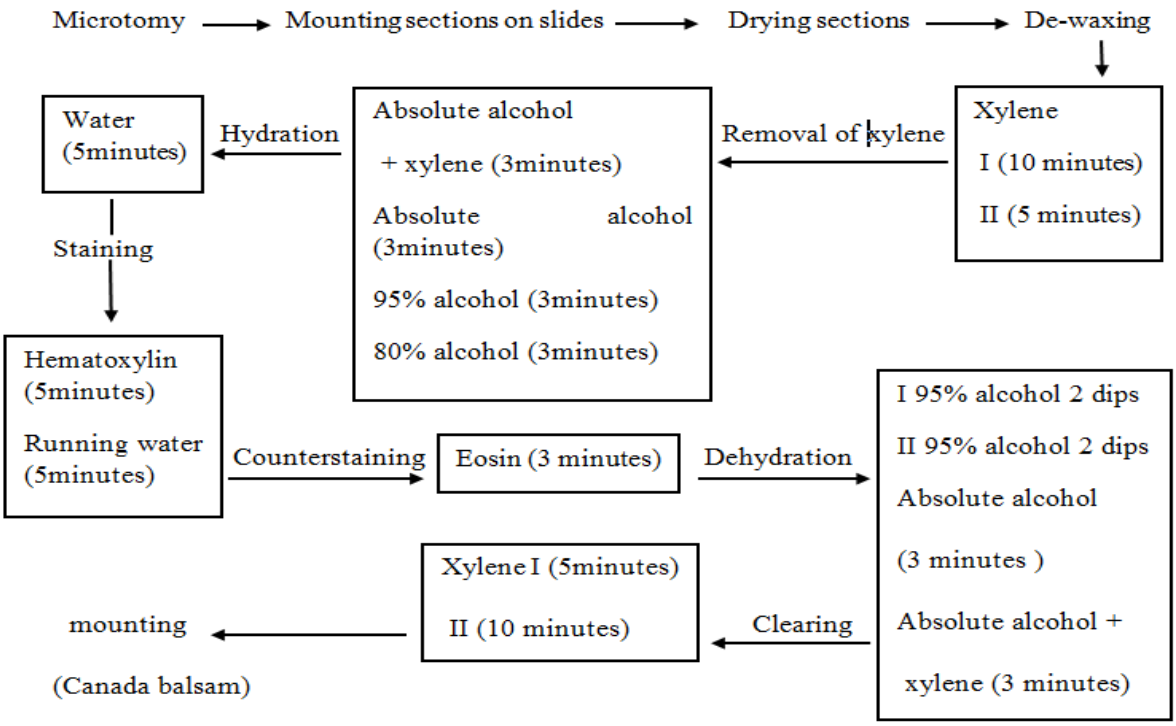

Figure 3.1 Haematoxylin-Eosin Staining Procedure

3.9 Statistical Analysis

Data were subjected to one-way ANOVA (analysis of variance) using GraphPad Prism. The data are presented as mean ± SEM, and the threshold for statistical significance is set at $P = 0.05$. A p value of < 0.05 is treated as statistically significant and indicated with an asterisk.

CHAPTER IV

4. Results

4.1 Haematological and Serum Biochemical Parameters in Rats

The first objective of this study was to evaluate the effect of $KBrO_3$ on the haematological characteristics of Wistar rats and to observe any protection provided by red onion juice against $KBrO_3$ toxicity in experimental rats. Thirty rats were randomly divided into six groups, each of which were given different concentrations of $KBrO_3$ and red onion juice. Serum biochemical analysis was then performed to determine the haematological characteristics of the subject animals in order to evaluate the extent of damage caused.

4.1.1 Haematological Parameters

The mean±SE values for haematological properties from different samples of each group are presented in Table 4.1 and Figure 4.1. Although there were variations in RBC and HGB, the differences were not statistically significant compared to normal values. On the other hand, the results showed a statistically significant ($P < 0.05$) increase in WBC when 50 or 30 mg/kg bw of $KBrO_3$ had been administered to rats as compared with normal controls

However, when the $KBrO_3$ was administered concurrently with juice of red onion, decreases in WBC levels towards normal control values occurred.

The results also show a significant decrease in PLT in the group treated with 100 mg/kg bw of $KBrO_3$ compared with the control group (Table 4.1). No significant difference was observed between the PLT values of all other groups and the controls.

Table 4.1 Effects of $KBrO_3$ on haematological parameters, and the protection provided by red onion juice against $KBrO_3$ toxicity in male albino rats.

Parameter / Group	**WBC** (mean±SE)	**RBC** (mean±SE)	**HGB** (mean±SE)	**PLT** (mean±SE)
Control	4.42± 1.23	7.56± 0.16	13.6±0.15	563.8±31.38
$KBrO_3$ (100 mg/kg bw)	6.46±061	8.11± 0.55	14.06±1.02	348.4±16.57*
AcE + $KBrO_3$ (100 mg/kg bw)	8.74±1.38	8.05± 0.42	13.44±0.75	414±50.77
$KBrO_3$ (50 mg/kg bw)	14.12±2.51***	8.52± 0.38	14.94±0.60	689.2±106
AcE + $KBrO_3$(50 mg/kg bw)	4.26±0.48	8.20± 0.18	14.06±0.27	606±5.74
$KBrO_3$ (30 mg/kg bw)	10.48±1.71*	8.16± 0.14	14.14±0.15	689±17.17

One-way analysis of variance (ANOVA) followed by Duncan's Multiple Range test was used for statistical analysis. The values shown are means ± SE.

*** $P < 0.001$ Very highly statistically significant difference between control and treatment group.

* $P < 0.05$ Statistically significant difference as compared to the control group.

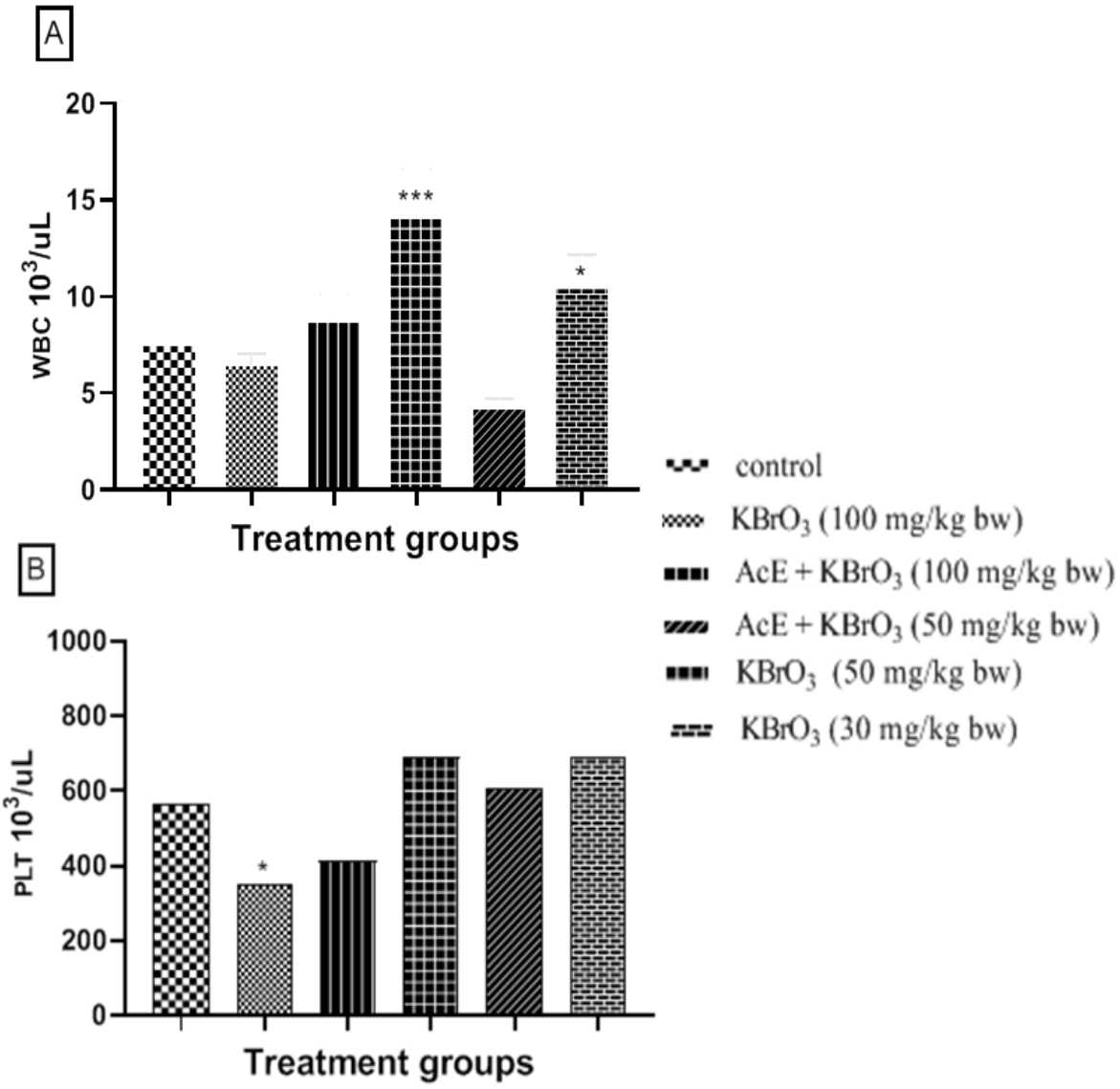

Figure 4.1 Effects of $KBrO_3$ on haematological parameters and of pretreatment with red onion juice against $KBrO_3$ toxicity in male albino rats compared with the control group.

4.1.2 Effect of $KBrO_3$ and Red Onion on Serum Biochemical Parameters

Biochemical tests were conducted to provide information on the status of the rat liver to assess the extent of damage, and the results are shown in Table 4.2. No significant differences were found in alanine aminotransferase (ALT) concentration between control and treatment groups, while significant increases were observed in alkaline phosphatase (ALP) and aspartate aminotransferase (AST) in those treated with 100 mg/kg bw of $KBrO_3$ at days 24 and 27 of the experiment and in the group treated with 30 mg/kg bw of $KBrO_3$ daily throughout the experimental period. When rats received red onion juice plus $KBrO_3$, the activity levels of ALP, ALT and AST were no different from those of the controls, as indicated in Figure 4.2.

Table 4.2 Effects of $KBrO_3$ on ALP, ALT and AST and the protection provided by red onion juice against $KBrO_3$ toxicity in male albino rats

Parameter / Croup	**ALP** (mean±SE)	**ALT** (mean±SE)	**AST** (mean±SE)
Control	205.4±27.38	38.4±2.16	87±5.3
$KBrO_3$ (100 mg/kg bw)	315.6±21.25**	38.4±5.34	154.4±11.8*
AcE + $KBrO_3$ (100 mg/kg bw)	211.2±24.49	39.4±4.38	129.2±6.85
$KBrO_3$ (50 mg/kg bw)	149.6±16.92	40.4±3.75	112±17.44
AcE + $KBrO_3$(50 mg/kg bw)	143.4±4.49	33±4.53	113.2±32.47
$KBrO_3$ (30 mg/kg bw)	296.8±16.73*	29.8±10.43	167±11.87*

All groups were tested using ANOVA in comparison with control group.
** $P < 0.01$ Highly statistically significant difference compared to the control group.
* $P < 0.05$ Statistically significant difference compared to the control group.

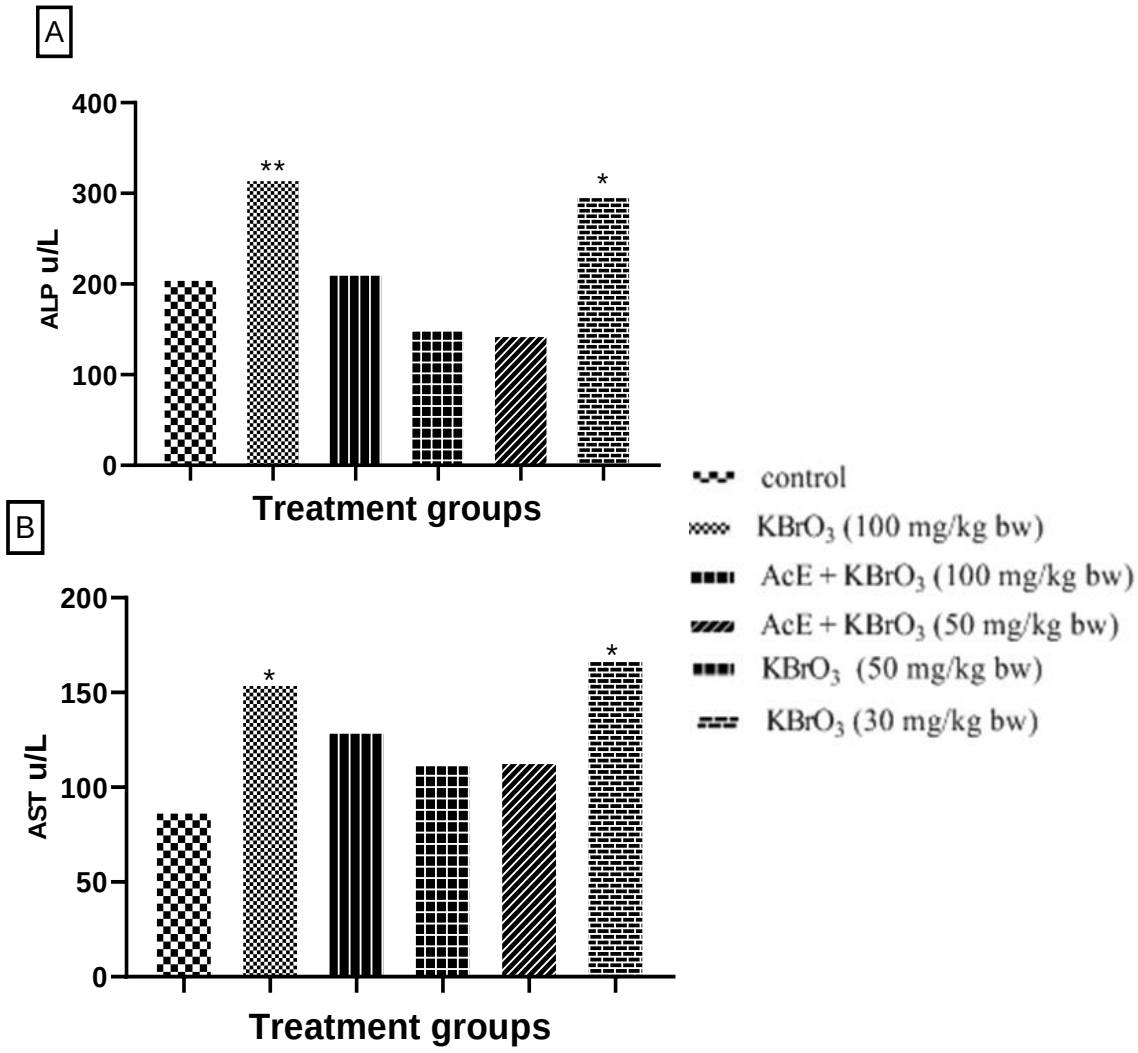

Figure 4.2. Changes in liver function markers in different experimental groups: (a) serum alkaline phosphatase; (b) serum aspartate aminotransferase.

** $P < 0.01$ Highly statistically significant difference compared to the control group.

* $P < 0.05$ Statistically significant difference compared to the control group.

4.2 Biomarkers of Renal Function

For the assessment of kidney functioning, serum urea, creatinine and uric acid content were estimated. The results for the biochemical tests carried out on the serum and urine of the rats are highlighted below:

4.2.1 Serum Urea, Creatinine and Uric Acid

Changes in the levels of renal functional markers in the experimental rats are given in Table 4.3. In $KBrO_3$-treated rats, renal functional markers such as creatinine, urea and uric acid increased markedly compared to the control rats ($P < 0.05$), indicating clear dysfunction caused by exposure to $KBrO_3$. Levels of these biochemical parameters improved after treatment with the red onion juice, and were lower in the rats treated with AcE + $KBrO_3$ compared with the $KBrO_3$ groups as shown in Figure 4.3.

Table 4.3 Biochemical markers of kidney.

Parameter / Group	**Urea** (mean±SE)	**Creatinine** (mean±SE)	**Uric Acid** (mean±SE)
Control	1.16±0.05	3.04±0.25	28.8±1.36
$KBrO_3$ (100 mg/kg bw)	1.62±0.06*	4.62±0.33*	58.2±5.19*
AcE + $KBrO_3$ (100 mg/kg bw)	1.2±0.14	3.16±0.29	40.8±1.88
$KBrO_3$ (50 mg/kg bw)	1.5±0.13	4.22±0.38	57±9.63*
AcE + $KBrO_3$ (50 mg/kg bw)	1.36±0.13	3.68±0.41	44±7.32
$KBrO_3$ (30 mg/kg bw)	1.62±0.13*	4.82±0.54*	60±11.1*

The relationships between group treatment and results for biochemical markers were assessed using one-way ANOVA.

* $P < 0.05$ Statistically significant difference compared to the control group.

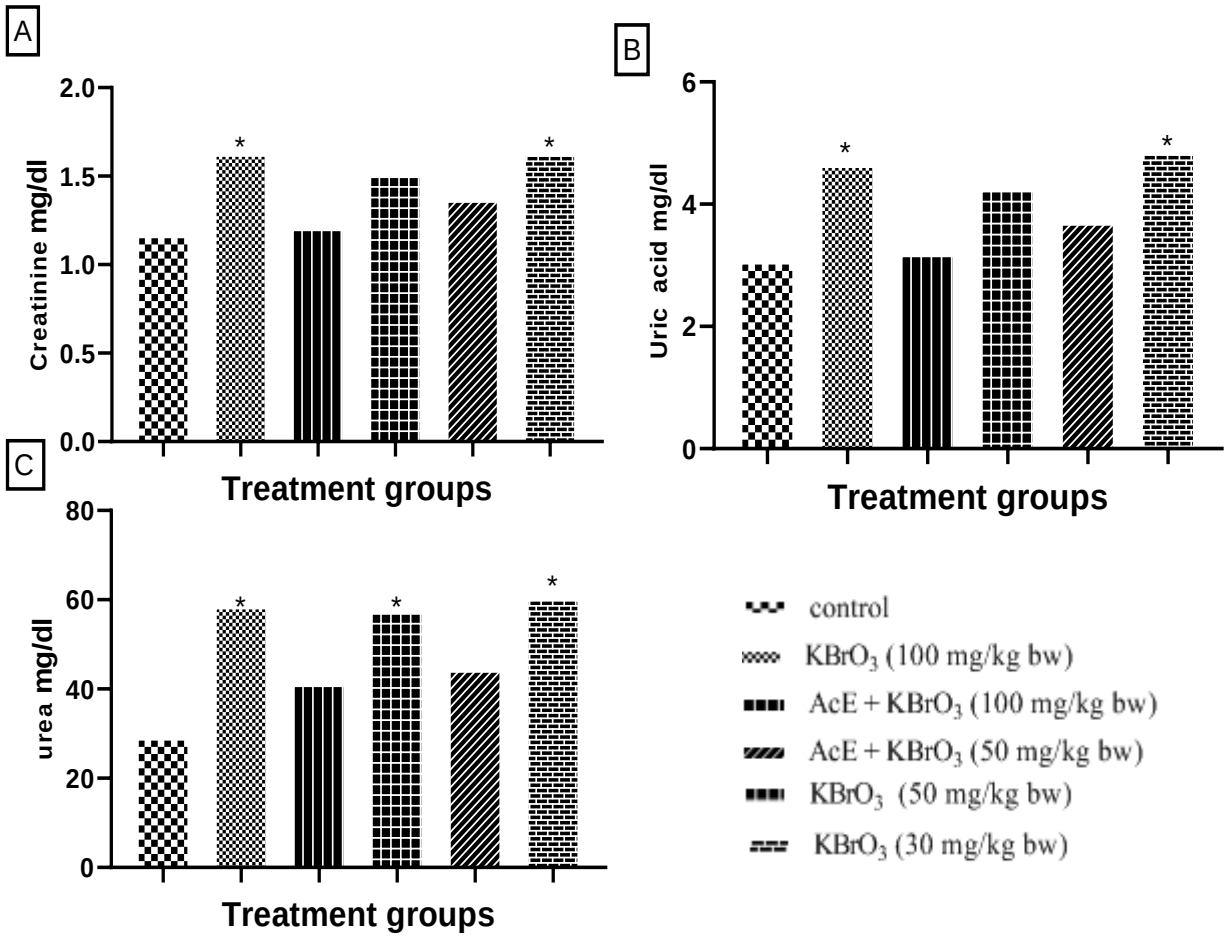

Figure 4.3. Effects of $KBrO_3$ on kidney function in terms of serum urea, creatinine and uric acid concentrations, and the protection provided by red onion juice against $KBrO_3$ toxicity in male albino rats compared with the control group.

* $P < 0.05$ Statistically significant difference compared to the control group.

4.2.2 Concentrations of Sodium and Potassium Ions

The results show a significant increase in sodium and potassium concentrations only when rats were treated with 30 mg/kg bw of $KBrO_3$ daily throughout the experimental period compared with the control group, as shown in Table 4.4, and Figure 4.4. The mean values of the electrolytes (Na^+ = 158.8±8.8; K^+ = 7.22±0.52) increased when rats were treated with 100 mg/kg bw of $KBrO_3$; however, the difference was not significantly different compared to the control value (Na^+ = 138±2.59; K^+ = 5.52±0.11). Pretreatment with red onion juice seemed to shift these parameters toward normal values.

Table 4.4. Effect of $KBrO_3$ on concentrations of sodium (Na^+) and potassium (K^+) ions, and the protection provided by red onion juice against $KBrO_3$ toxicity in male albino rats.

Parameter / **Group**	**Sodium** (mean±SE)	**Potassium** (mean±SE)
Control	138±2.59	5.52±0.11
$KBrO_3$ (100 mg/kg bw)	158.8±8.8	7.22±0.52
AcE + $KBrO_3$ (100 mg/kg bw)	138.3±1.94	5.55±0.37
$KBrO_3$ (50 mg/kg bw)	138.8±0.74	6.02±0.37
AcE + $KBrO_3$ (50 mg/kg bw)	139±0.81	6.91±0.32
$KBrO_3$ (30 mg/kg bw)	162±11.14*	8.09±0.72*

The relationships between groups and results for electrolyte levels were assessed using one-way ANOVA.
* $P < 0.05$ Statistically significant difference compared to the control group.

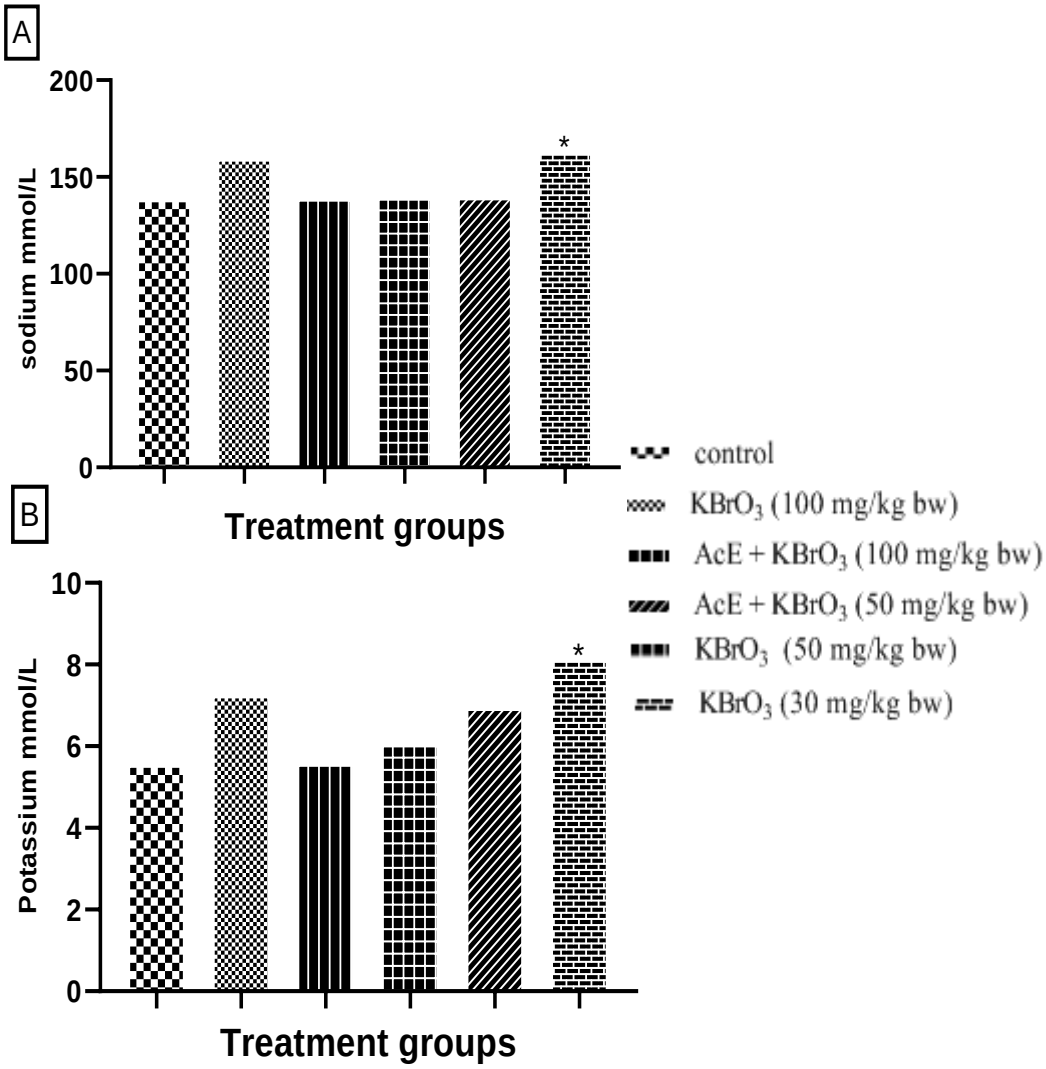

Figure 4.4 Effect of $KBrO_3$ on kidney serum concentrations of sodium and potassium ions, and the protection provided by red onion juice against toxicity $KBrO_3$ in male albino rats compared with the control group.

* $P < 0.05$ Statistically significant difference compared to the control group.

4.3 Effects of $KBrO_3$ and AcE on Body and Organ Weight

In order to investigate the protective effects of red onion juice against KBrO3-induced toxicity, the body weights of all experimental rats were measured. There were no significant changes in the body weights of rats that had survived to the end of the experiment nor in the relative weights of selected organs, as shown in Table 4.5. The weights of the kidney, testis, lung and liver in the treated groups were slightly lower compared to those in the control group, but the differences were not statistically significant.

Table 4.5 Effect of red onion juice on body and organ weight in $KBrO_3$-treated experimental rats.

Parameter / Group	Body (mean±SE)	Kidney (mean±SE)	Testis (mean±SE)	Liver (mean±SE)	Lung (mean±SE)
Control	222.4±7.34	0.72±0.02	1.10±0.15	7.01±0.25	1.24±0.09
$KBrO_3$ (100 mg/kg bw)	206.6±17.01	0.73±0.03	1.08±0.14	7.24±0.23	1.46±0.21
AcE + $KBrO_3$ (100 mg/kg bw)	191.4±9.19	0.80±0.07	1.16±0.11	6.45±0.43	1.34±0.15
$KBrO_3$ (50 mg/kg bw)	184.4±12.69	0.70±0.03	1.11±0.11	6.8±0.52	1.26±0.04
AcE + $KBrO_3$ (50 mg/kg bw)	213.6±4.82	0.74±0.05	1.32±0.07	6.82±0.23	1.46±0.11
$KBrO_3$ (30 mg/kg bw)	214.2±12.06	0.74±0.4	0.96±0.15	6.48±0.40	1.44±0.10

One-way analysis of variance (ANOVA) followed by Duncan's Multiple Range test were used for statistical analysis. The values shown are means ± SE.

4.4 Histopathological Observations

Generally, histological staining is performed with H&E to examine differences between tissue components under normal and pathological conditions.

4.4.1 Effects of Treatment on Histology of Kidney Tissues

The kidney is a target organ greatly affected by exposure to $KBrO_3$. The administration of $KBrO_3$ is toxic to the kidney and results in acute renal failure. Many published reports document the nephrotoxicity effect of $KBrO_3$. Renal tissue from all the experimental and control rats was examined using a light microscope. At least five fields from each section of kidney were assessed for the appearance of the glomerulus and the related tubules and compared with control sections taken at the same time using the same method of processing.

In the non-treated group, the microscopic observation of kidney sections displayed the normal architecture of both the renal corpuscles and tubules, as shown in Figure 5.1A. In addition, the renal histological examination showed no tissue degeneration, inflammation, necrosis, or tubular dilation. However, the histology of the kidneys of rats treated with 100 mg/kg of $KBrO_3$ shows degenerative changes in the renal tubules indicated by tubular destruction and the detachment of tubular epithelial cells. In addition, glomerular changes included the dilation of the Bowman's space and haemorrhaging (Figure 5.1B).

The administration of 50 mg/kg of $KBrO_3$ twice per week caused irregular glomerular morphology, hemorrhages, degeneration and necrosis. The desquamation of epithelial tubular cells was also observed, with a loss of the brush border as indicated with arrowheads in Figure 5.1D. Significant changes in the group treated with 30 mg/kg/day of $KBrO_3$ are apparent in Figure 5.1F, showing dilation in Bowman capsules and tubules (black arrows), shrinkage of the glomerular tuft (red arrow), capillary congestion (blue arrow) and severe

necrosis of the tubular cells (yellow arrow). In contrast, the KBrO3 + red onion juice group showed apparently mild to moderate tubular epithelial changes and less damage in histological architecture of renal corpuscles and glomerular tuft surrounded by Bowman's space (Figure 5.1C and 5.1E). There was also little tubular damage compared to group treated with $KBrO_3$ alone.

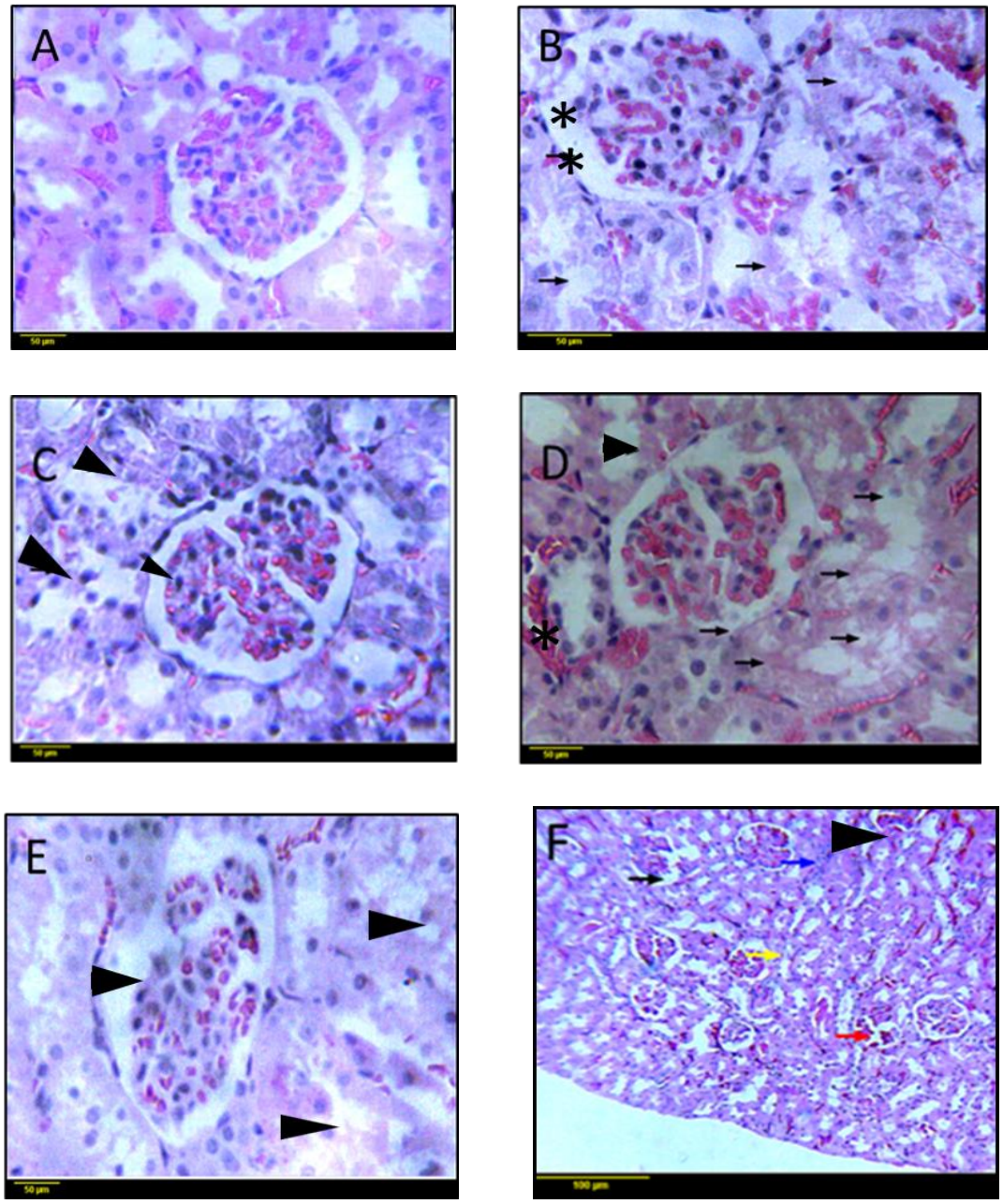

Figure 5.1. Light micrographs of hematoxylin and eosin staining of kidney tissues of control and experimental groups of rats. (A) Control group shows normal histology (x400)**. (B)** Rats intoxicated with $KBrO_3$ (100 mg/kg bw) show degenerated glomeruli (black arrows) and dilation of Bowman's space (Asterisk) (x400). (**C)** treated group by AcE + $KBrO_3$ (100 mg/kg bw) shows less damage (arrowheads) in glomerulus and tubules (x400). (**D)** irregular glomerular morphology, hemorrhage (Asterisk) and degenerated tubular (arrows) with loss of brush border (arrowheads) are seen in rats receiving $KBrO_3$ (50mg/kg bw) (x400). **(E)** AcE + $KBrO_3$ (50mg/kg bw) treated group (x400) shows moderate damage (arrowhead). (**F)** dilatation in Bowman capsule (black arrows), shrinkage of glomerular tuft (red arrow), capillary congestion (arrowhead), infiltration(blue arrow) and severe necrosis of tubular cells (yellow arrow) are seen in rats receiving $KBrO_3$ (30 mg/kg bw) (x100).

4.4.2 Effects of Treatment on Histology of Intestinal Tissues

The small intestine is the major site for the digestion and absorption of nutrients, and a large number of circumstances may affect its function, such as the administration of $KBrO_3$. Such a toxic chemical may cause damage to the small intestine, and especially the duodenum.

A histological evaluation of tissue samples from control rats demonstrated a normal architecture of the intestine, as shown in Figure 5.2.A, with the appearance of a prominent mucus layer. The administration of KBrO3 induced considerable changes observed in the small intestine. Microscopic examinations of intestinal tissue from rats treated with 100 mg/kg of bromate revealed the loss of some superficial epithelium, the necrosis of the upper villi and decreased numbers of goblet cells. Also, cellular debris was observed in the lumen of the crypt (Figure 5.2B).

In the group treated with 50 mg/kg bromate, there were indications of damage to the microvillar structure on the apical surface of the crypt epithelium. Decreased numbers of goblet cells can easily be detected in these areas (Figure 5.2D). Furthermore, decreases in villus height and crypt depth are marked in the group treated with 30 mg/kg/day of bromate. The shortening of the villus or fusion of villi, and desquamation of the surface epithelium were also observed. Decreased numbers of goblet cells, crypt loss and cell infiltration in the epithelial lining were also detected (Figure 5.2.F). Meanwhile, microscopic examination of intestinal tissue from rats treated with AcE + KBrO3 revealed negligible lesions on the surface epithelium and in histological characteristics.

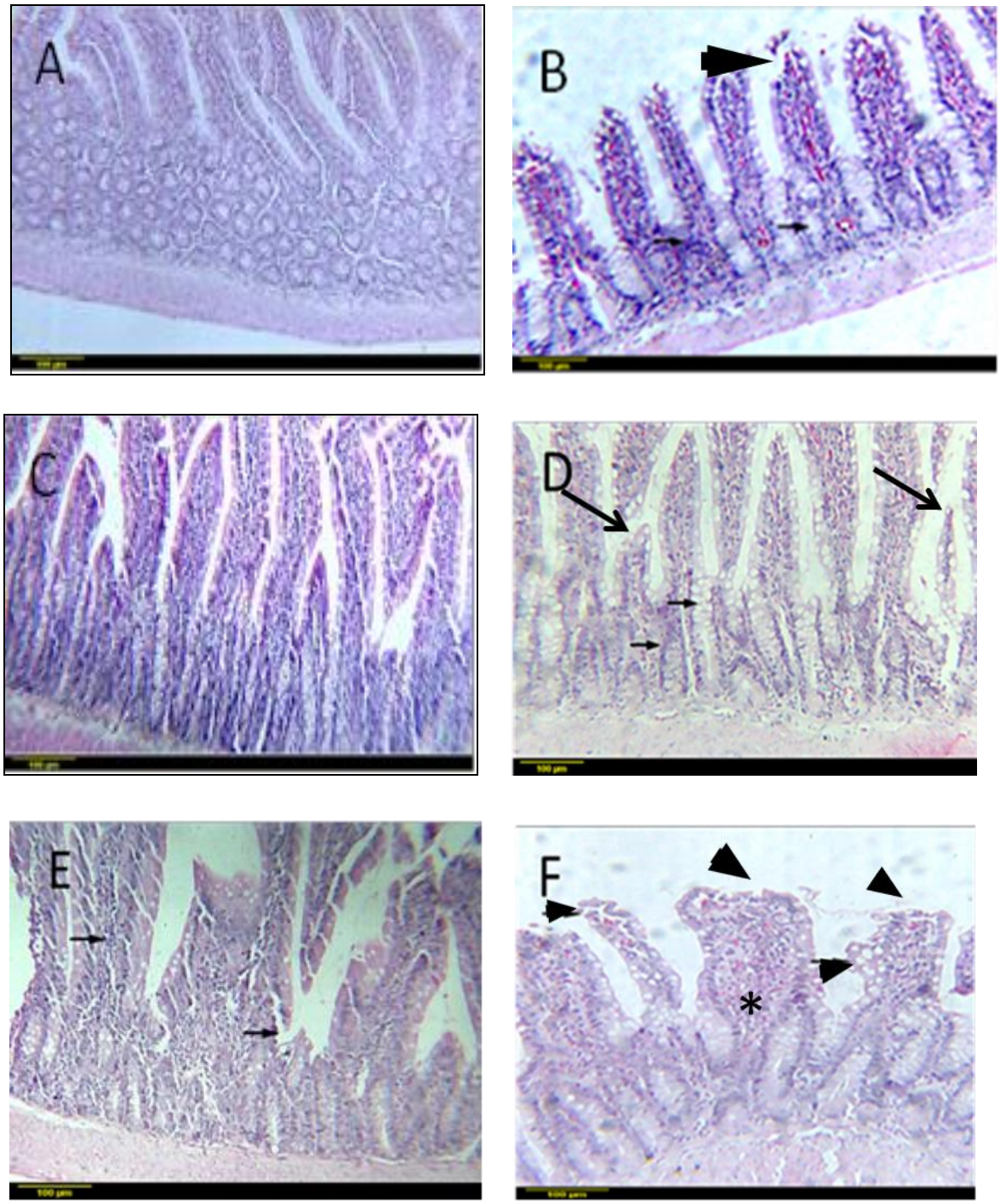

Figure 5.2 Micrograph of the small intestine, harvested from control and treated rats. (**A)** Normal architecture of duodenum from control rats (**B)** loss of some superficial epithelium (arrowhead), decreased number of goblet cells and cellular debris (arrow) was observed in the lumen of the crypt in rats treated with $KBrO_3$ (100 mg/kg bw). (**C)** Rats received AcE + $KBrO_3$ (100 mg/kg bw) shows less damaged tissue. (**D)** damaged microvillar structures on the apical surface of crypt epithelium (arrows) and decreased number of goblet cells are seen in rats receiving $KBrO_3$ (50 mg/kg bw). **(E)** treated group section with AcE + $KBrO_3$ (50 mg/kg bw) shows negligible lesions of surface epithelium. (**F)** Shortening of villus or fusion of villi, desquamation of surface epithelium (arrowheads), and cell infiltration (Asticks) in the epithelial lining treated are seen in rats treated with $KBrO_3$ (30 mg/kg bw). (sections stained with H&E, x100).

CHAPTER V

5. Discussion

5.1 Effect of Potassium Bromate on Haematological Characteristics

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. The present study was designed to investigate any toxic effects of $KBrO_3$ on the haematological characteristics of Albino Wistar rats and to observe any protection provided by red onion juice against $KBrO_3$ toxicity in these rats. A biochemical analysis of serum was carried out to assess haematological parameters and thus to evaluate the extent of any damage occurring.

5.1.1 Effect of $KBrO_3$ on Blood Parameters

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 inflammation. The results of the present study showed a significant increase in WBC, which might associated with inflammatory conditions due to the effect of $KBrO_3$ and similar chemicals as suggested by Ullah *et al.* (2020). It is also observed that $KBrO_3$ does not affect the red blood cell count (RBC) or haemoglobin level (Hb%), which contrasts with the findings of Mohamed and Saddek (2019) who observed significant decreases in these parameters after rats were treated with a 100 mg/kg bw dose of $KBrO_3$

Rats in $KBrO_3$-administered groups showed a notable decline in platelets (PLT) as compared to the controls. PLT play an important role in blood clotting and prevent blood loss from haemorrhaging. Potassium bromate may thus adversely affect platelet levels. Similar results were reported by Achukwu *et al.* (2009) and Akinola *et al*. (2020). These reductions in PLT could be due to the DNA strand breaking in cells, induced by the oxidative stress associated with $KBrO_3$ (Chipman *et al.* 1998)

However, the pre-treatment of animals with red onion juice improved the haematological parameters and PLT content improved, while a significant reduction in the WBC count down to a normal level was noted. These findings are in agreement with earlier studies, including those by Akinola *et al*. (2020), Dhembare and Dale (2017), and others. This effect may indicate an activation of the animal's immune system in response to tissue damage caused by any toxicant (Milan and Gyan 2016).

5.1.2 Effect of $KBrO_3$ on the Biochemical Parameters of the Kidney

The kidney is involved in the secretion of many of the products of toxic metabolic waste. In the current study, treatment with $KBrO_3$ also induced significant increases in some renal function biomarkers compared with other groups, including urea, creatinine, uric acid, and sodium and potassium ions. This is an indication of severe kidney damage, which was also observed in terms of pathological differences in kidney parameters. Similar results were reported by Khan *et al.* (2003). Increases in serum creatinine and urea are damage indicators concerning poor glomerular filtration and have been recognized as major biomarkers of kidney dysfunction and the loss of integrity of the renal tubules (Gowda *et al*. 2010). Also, Akomolafe *et al*. (2020) stated that rising levels of urea have been linked with renal failure. The increase in uric acid levels may also be due to the degradation of purines and pyrimidines which are strongly linked to a surge in xanthine oxidase activity, causing the overproduction of uric acid and also the production of reactive oxygen species (ROSs) (Mohamed and Saddek 2019). However, the administration of red onion juice reversed the abnormal amounts of creatinine, urea, and NA^+ and K^+ electrolytes in rats in the $KBrO_3$ + AcE group whose levels were not significantly different from the controls. This suggests a possible protective effect of red onion against $KBrO_3$-induced renal damage. This finding is in line with those of Haidari *et al.* (2008), where the total antioxidant status of hyperuricemic rats increased after receiving 5 g/kg/day of onion for 2 weeks, as well as Rahmat *et al*. (2019) where a larger dose of 10.5 g/kg/day of onion juice decreased the serum uric acid level in hyperuricemic rats. Rahmat *et al*.

(2019) concluded that the hypouricemic effects of onion juice might be due to the presence of flavonoids which exert inhibitory effects on XO and xanthine dehydrogenase enzyme activities. Moreover, the rich flavonol content of red onion, with 32 phenolic compounds, is believed to be responsible for antioxidant activity such as the reduction of uric acid in the blood (Abouzed *et al*. 2018). However, despite the potential benefits of red onion, some experiments have identified possible negative effects in mice after the consumption of aqueous red onion extract over 56 days (Spataru *et al*. 2019).

5.1.3 Effect of $KBrO_3$ on the Biochemical Parameters of the Liver

The liver is the main organ of detoxification and plays a central role in severe metabolic processes in general, and it is prone to various disorders due to exposure to toxins (Nikmaneshi *et al*. 2020). Measurements of serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) have been used extensively in the evaluation of liver damage due to the release of large quantities of these enzymes into the bloodstream (Jaffar *et al*. 2020). The present study has confirmed that a significant ($P < 0.05$) increase was observed in the activities of selected liver enzymes when rats were treated with either 100 or 30 mg/kg bw $KBrO_3$. Similar results have been obtained in mice treated with 100 mg/kg/day of KBrO3 for 15 days, and the observed increase in plasma AST and ALT suggests the leakage of these enzymes from the liver to the plasma, leading to hepatocyte damage (Ben Saad *et al*. 2016). Oloyede and Sunmonu (2009) also showed a significant increase in alpha amylase (ALP), ALT and AST activity levels, which indicates the adverse effect on the livers of rats fed on a diet containing potassium bromate. This may be attributed to the

hepatocellular damage induced by superoxide anions, hydrogen peroxide, and hydroxyl radicals, which cause oxidative stress to the cell membrane resulting in increases in liver enzyme activity (Ben Saad *et al*. 2016). However, according to the present study, treatment with AcE at a dose of 1 ml/kg significantly ameliorated $KBrO_3$-induced liver injury. This protective effect is clearly evident from the reduction of the elevated marker enzymes ALP and AST to levels similar to those of the control group. A possible mechanism by which AcE could exhibit protection against $KBrO_3$-induced hepatotoxicity may be due to the presence of active phytomolecules like phenolic acids, flavonoids, cepaenes, thiosulphinates, and anthocyanins (Singh *et al*. 2009; Metrani *et al*. 2020). Results obtained in comparable investigations suggest that *A. cepa* plays a protective role in cadmium-induced testicular oxidative damage and sperm toxicity (Ola-Mudathir *et al*. 2008), and it provides regenerative potential against aluminum-provoked toxicity in male mice (Ara *et al*. 2020) as well as possibly also protecting against the hyperglycaemia and dyslipidemia arising from diabetes (Ülger and Çakiroglu 2020).

5.2 Histopathogical Observations

The investigation of tissue alterations in selected internal organs and tissues could be an important aid in the diagnosis of the adverse effects of chemicals like potassium bromate in rats. In the current research, the potentially harmful effects of $KBrO_3$ on the kidney and intestine were examined histologically as well.

5.2.1 Histopathological Observations of Kidney Tissue

The kidneys are important organs responsible for the excretion of xenobiotics and their metabolites, and are susceptible to damage caused by external substances. So, a second objective of this study was investigate the nephrotoxic effect of oral doses of $KBrO_3$ in rats and the possible ameliorative effect of pre-treatment with red onion extract. The histopathological results observed in the present study support the biochemical results and indicate that $KBrO_3$ induces severe histological alterations in kidney tissue. Similar changes have been recorded by other investigators (Mohamed and Saddek 2019; Rashmi and Negi 2020) The histopathological results in this study revealed that oxidative damage to kidney tissues was evident in rat groups treated with different doses of potassium bromate. This included the dilatation of the Bowman capsule, glomerular shrinkage, and haemorrhage associated with tubular degeneration, as well as marked necrotic changes and loss of the brush border. In contrast, pretreatment with red onion led to significant amelioration of the significant pathological changes induced by $KBrO_3$ administration. This could be attributed to the antioxidant and anti-inflammatory characteristics of red onion, which significantly reduces oxidative stress leading to a restoration of the normal physiological state of kidney cells (Mohamed and Saddek 2019). Moreover, bioactive compounds in red onion may provide further improvements to these pathological changes in the renal tissue of $KBrO_3$-treated rats (Rashmi and Negi 2020). Similar observations have been reported in an earlier study by Ben Saad *et al*. (2016), which confirmed that treatment with 100 mg/kg/day of vanillin for 15 days restored the histopathological changes in $KBrO_3$-induced kidney damage.

The histopathological changes seen in the renal tissues of $KBrO_3$ induced nephrotoxic untreated group further confirm the renal injury which might be activated by oxidative damage. Past studies have indicated that $KBrO_3$ may initiate glomerular injury, tubular necrosis, and other damage (Mohamed and Saddek 2019; Akomolafe *et al*. 2020; Alhazza *et al*. 2020). This is in line with the findings of the current study which indicate the degeneration of corpuscular tissues after $KBrO_3$ administration in comparison with the control.

5.2.2 Histopathological Observations of Intestine Tissue

Antioxidant characteristics of red onion have been previously reported in different organs, such as the liver (Ige *et al*. 2011; Ahmed *et al*. 2017), kidney (Ige *et al*. 2009; Ara *et al*. 2020) and testis (Ola-Mudathir *et al*. 2008). Therefore another objective of the present research work has been to explore the possible protective effect of red onion against the injuries to rats caused by $KBrO_3$ in other organs such as the intestine through histological study. This study shows that the histological features of the intestinal mucosa of rats treated with 50 or 100 mg/kg bw of $KBrO_3$ were characterized by epithelial desquamation, loss of the crypt, and inflammatory infiltration along with decreases in goblet cells. These histopathological changes again correlate with the biochemical findings. Similar results presented by Ahmad *et al.* (2015) were that the administration of 100 mg/kg bw of $KBrO_3$ induced DNA degradation in the intestinal tissue of treated rats and led to extensive intestinal damage such as mucosal cell damage, especially to the membrane. Also the lumen became full of debris and the intestinal villi lost their form. Kurokawa *et al.* (1990) conducted an extensive study of the effects of $KBrO_3$ on rats, and concluded that

it affects intestinal morphology and induces adenomas in the small intestine in experimental animals.

Potassium bromate has been reported to induce tumours in the small intestine of the mouse after oral administration (Isoda *et al.* 2014; Piao *et al.* 2014*;*). In addition, a more recent study by Aoki *et al.* (2020) investigated tumour formation in the small intestine of *gpt* delta mice induced by the oral administration of $KBrO_3$. They showed that the formation of the oxidative DNA base modification 8-oxo-deoxyguanosine (8-oxo-dG) was significantly increased at doses of 0.6 and 2 g/L in these mice.

It is well known that the small intestine is the site at which the digestion and absorption of 90% of ions and molecules occurs. On the other hand, this may cause undesirable results; for example, when food contain drugs, toxic pollutants or harmful chemicals such as $KBrO_3$ (Circu and Aw 2011). Thus, seems likely that any alterations in metabolic pathways caused by toxicants could affect the function of the small intestine (Ahmad *et al.* 2013). The intestinal brush border membrane (BBM) is one of the most important cellular membranes, because it is a major site of antioxidant action besides its role in nutritional absorption and digestion (Shahid *et al*. 2018). The administration of $KBrO_3$ to rats has been found to induce oxidative stress (OS) and to lower the activity levels of several enzymes in the BBM and to cause extensive damage to the villi and intestinal gland cells, with the lumen being filled by debris (Ahmad *et al.* 2013). The decrease in the activity of BBM enzymes may be linked to $KBrO_3$-generated free radicals and ROSs which lead to leakage or loss of the enzymes after ROS-induced damage to the epithelial cells lining the intestine, and especially to the cell membrane (Shahid *et al*. 2017). In addition, Ahmad *et*

al. (2015) concluded that increased lipid peroxidation may affect the structure of the intestinal membrane and its function, leading to the decreased activity of these enzymes.

In this study, rat body weight decreased after $KBrO_3$ treatment compared with those in the control group, although the difference was not statistically significant. This finding is consistent with research carried out by Rezq (2017) which showed that the oxidative stress induced by $KBrO_3$ in rats caused significant decreases in body weight. The reduction in body weight could be attributed to the direct toxic effect of $KBrO_3$ on the gastrointestinal tract, which perhaps results in the poor digestion of food or malabsorption of nutrients (Rezq 2017).

The small intestine is exposed continuously to high levels of ROSs and requires digestive and absorptive functions to be tightly controlled by a series of antioxidant substances (Wang *et al.* 2018). In this study, the protective effect of AcE on $KBrO_3$ induced intestinal damage could have been due to its effectiveness in inhibiting of $KBrO_3$ generated free radicals before they attacked their cellular target. The subsequent reduction in lipid peroxidation and oxidative modification of BBM enzymes might have contributed to the efficacy of the antioxidants in ameliorating the effects of $KBrO_3$ (Rezq 2017). The present results are in accordance with those of Rezq (2017), who showed that the intake of sesame oil or jojoba oil may be useful in improving liver and kidney function and might protect against $KBrO_3$-induced oxidative stress in rats by providing stronger antioxidant activity. Similarly, Ahmad *et al*. (2015) showed that taurine alleviates $KBrO_3$-induced tissue toxicity and oxidative

damage by improving antioxidant defence, tissue integrity and energy metabolism.

Finally, red onion exhibits important biological activity for health maintenance due to its huge content of antioxidant compounds, which have radical scavenging potential which can help to prevent or slow down the oxidation of free radicals before damage to cells and tissues can occur. It is one of the vegetables richest in flavonoids, mainly comprising quercetin and its derivatives (Zhang *et al*. 2016). Meanwhile bread is a staple food of humans worldwide (Pagewise 2002) and a basic element of bread is flour, which usually contains flour enhancers such as potassium bromate to improve the quality of the product's texture. In addition, most bakeries use $KBrO_3$, whether legally or illegally, through the use of brominated flour in producing a variety of types of bread commonly consumed in our region. So, the regular consumption of red onion in the diet can be recommended in order to avoid some of the harmful effects of $KBrO_3$ in bread and other products containing food additives.

Conclusion

Potassium bromate ($KBrO_3$) causes toxicity in humans and experimental animals. It is a class IIB carcinogen, and its application in food processing is restricted in many countries. Due to long exposure to $KBrO_3$ in water disinfection and as a food additive, antidotes need to be available for use in protection against its hazards. The current study has attempted to examine the protective role of pre-treatment with red onion juice in ameliorating the deleterious effects of $KBrO_3$-induced oxidative stress in haematology, the kidney and small intestine. In this investigation, bromate incited nephrotoxicity in rats led to changes in renal tissues, as demonstrated by changes in some kidney biomarkers and biochemical parameters. $KBrO_3$ caused increases in the levels of urea, uric acid, creatinine and sodium in the serum, which reflect reductions in glomerular filtration and tubular damage. The results demonstrate that $KBrO_3$ also induced haematological changes such as a significant increase in WBC and a decrease in PLT, which occur due to the harmful effects of $KBrO_3$ on the bone marrow and haematopoietic organs. The activity of the aspartate aminotransferase (AST) and alkaline phosphatase (ALP) enzymes in the plasma also significantly increased, which might be attributed to the hepatotoxicity induced by $KBrO_3$. The histopathology results for $KBrO_3$-treated rats showed pathological alterations in the renal capsule, the Bowman's space, and the epithelial tubules. Histological observations of the small intestine (duodenum) strongly support the biochemical results. Intestines from $KBrO_3$-treated rats showed various types of intestinal damage. The lumen was full of

debris and inflammatory cells, the intestinal villi had lost their contours, and goblet cells had decreased.

However, the administration of red onion juice was shown to have a marked nephroprotective role against the deleterious effects induced by $KBrO_3$ in rats. *Allium cepa* (AcE) significantly improved haematological, biochemical and histological parameters while attenuating inflammatory and oxidative stress markers. Furthermore, this treatment was associated with no observable toxicity in the animals. Thus, based on the current study, AcE may have the potential to counter the nephrotoxic effects of $KBrO_3$. Onion is a well-known antioxidant-rich raw vegetable. Vegetables such as red onion have been found to contain substances providing higher antioxidant activity compared to other vegetables. Thus, it is believed that the consumption of red onion increases the total antioxidant status of rats. Red onion is also inexpensive and non-toxic and can be administered safely and significantly to people who are exposed to $KBrO_3$ and related compounds. We suggest that the red onion or its active compounds have a protective role to maintain accurate renal damage and intestinal toxicity caused by potassium bromate. In addition, red onion has been confirmed to have beneficial effects on human health via multiple different functions, including antioxidant, anti-inflammatory, and antibacterial properties, and it is recommended to be taken frequently in the diet.

Furthermore, there is a need to study the effect of all the metabolites formed as a result of the thermal processing of potassium bromate in bread and their toxicity levels after digestion, in order to ensure the safety of bread and other bakery products if potassium bromate is added, or else to find a healthier alternative that can be used as a safe replacement for it. Henceforth, further

investigations are necessary to elucidate the mechanism of nephroprotective impact caused by red onion against potassium bromate. Also, random samples of bread products commonly sold to and consumed by people should be analyzed to determine their levels of safety for human consumption with respect to bromate and trace metallic content.

REFERENCES

Abdel Gadir E, Gadir WA, Adam S. 2007. Toxicologic interaction of potassium bromate and *Allium cepa, Allium sativum* or sodium selenite in wistar rats. *Journal of toxicology and pharmacology*, 2:496-501.

Abdel-Reheim E.S, Abdel-Hafeez H A, Mahmoud BM, Abd-Allah EN. 2014. Effect of food additives (monosodium glutamate and sodium nitrite) on some biochemical parameters in albino rats. *International Journal of Bioassays*, 3 (8): 3260-3273.

Abouzed TK, Contreras MDM, Sadek KM, Shukry M, H Abdelhady D, *et al*. 2018. Red onion scales ameliorated streptozotocin-induced diabetes and diabetic nephropathy in Wistar rats in relation to their metabolite fingerprint. *Diabetes Research and Clinical Practice*, 140:253-264.

Achukwu PU, Ufelle S, Ukaejiofo EO, Ejezie FE, Nwachukwu DN. *et al*. 2009. The effect of potassium bromate on some haematological parameters of Wistar rats. *Nigerian Journal of Physiological Sciences*, 24: 59-61.

Ahmad MK, Khan AA, Ali SN, Mahmood R. 2015. Chemoprotective effect of taurine on potassium bromate-induced DNA damage, dna-protein cross-linking and oxidative stress in rat intestine. *Public Library of Science ONE*, 10 (3).

Ahmad MK, Amani S, Mahmood R. 2014. Potassium bromate causes cell lysis and induces oxidative stress in human erythrocytes. *Environmental Toxicology*, 29: 138-145.

Ahmad MK, Mahmood R. 2014. Protective effect of taurine against potassium bromate-induced hemoglobin oxidation, oxidative stress, and impairment of antioxidant defense system in blood, *Environmental Toxicology*, 31: 304-313.

Ahmad MK. 2013. Effect of potassium bromate on human blood and some rat tissues. Thesis, Aligarh - 202 002 (INDIA)

Ahmad MK, Khan AA, Mahmood R. 2013. Taurine ameliorates potassium bromate-induced kidney damage in rats. *Public Library of Science ONE*, 45(5): 1109-21.

Ahmad MK, Naqshbandi A, Fareed M, Mahmood R. 2012. Oral administration of a nephrotoxic dose of potassium bromate, a food additive, alters renal redox and metabolic status and inhibits brush border membrane enzymes in rats, *Food chemistry*, 134: 980-985

Ahmad MK, Amani S, Mahmood R. 2011a. Potassium bromate causes cell lysis and induces oxidative stress in human erythrocytes. *Environmental Toxicology*, 29: 138-145.

Ahmad MK, Syma S, Mahtnood R. 2011b. Cr (V1) induces lipid peroxidation, protein oxidation and alters the activities of antioxidant enzymes in human erythrocytes. *Biological Trace element Res*, 144: 426-435.

Ahmed AF, Al-Yousef HM, Al-Qahtani JH, Al-Said MS. 2017. A hepatonephro-protective phenolic-rich extract from red onion (*Allium cepa L.)* peels. *Pakistan Journal of Pharmaceutical Sciences*, 30(5):1971-1979.

Akanji MA, Nafiu MO, Yakubu MT. 2008. Enzyme activities and histopathologp of selected tissues in rats treated with potassium bromate. *The Journal of Biomedical Research*, 11: 87-95.

Akinola BK, Olawuyi TS, Ogunmokunwa AE. 2020. The Protective Effects of *Telfairia occidentalis* on potassium bromate induced hepatotoxicity in adult Wistar rats. *African Journal of Biological Sciences*, 2(3): 51-61.

Akomolafe, SF, Olasehinde, TA, Adewale, OO, Ajayi OB. 2020. Curcumin improves biomolecules associated with renal function and attenuates oxidative injury and histopathological changes in potassium-induced toxicity in rats' kidney. *Biological Trace Element Research*. 199(1), 197-204.

Alhazza IM, Hassan I, Ebaid H, Al-Tamimi J, Alwasel SH. 2020. Chemopreventive effect of riboflavin on the potassium bromate-induced renal toxicity in vivo. *Naunyn Schmiedebergs Arch Pharmacol*, 14.

Ali BH, Al Za'abi M, Karaca T, Al Suleimani Y, Al Balushi KA, *et al*. 2018. Potassium bromate-induced kidney damage in rats and the effect of gum acacia thereon. *American Journal of Translational Research,* 10: 126-137.

Al-Samawy ERM. 2012. Morphological and Histological study of the kidneys on the Albino rats. *Al-Anbar Journal of Veterinary Sciences*, 5(1):115-119.

Altoom NG, Ajarem J, Allam AA, Maodaa SN, Adel-Maksoud MA. 2018. Deleterious effects of potassium bromate administration on renal and hepatic tissues of Swiss mice. *Saudi Journal of Biological Sciences*, 25: 278-284.

Atta EM, Mohamed NH, Abdelgawad AAM. 2017. Antioxidants: An overview on the natural and synthetic types. *European Chemical Bulletin*, 6: 365-375

Andreozzi L, Giannetti A, Cipriani F, Caffarelli C, Mastrorilli C, *et al*. 2019. Hypersensitivity reactions to food and drug additives: Problem or myth? *Acta BioMedica*, 90: 80-90

Aoki Y, Taniguchi Y, Matsumoto M, Matsumoto M, Ohno M, *et al*. 2020. Oxidative-stress-driven in vivo mutagenesis induced by oral administration of potassium bromate in the small intestines of gpt delta mice. *Mutation Research/ Genetic Toxicology and Environmental Mutagenesis*, 503136.

Ara C, Asmatullah SH, Andleeb S, Zahid B, Arshad M. *et al*. 2020. Red Onion Extract Attenuates Aluminum Induced Toxicity in Swiss Albino Mice. *Pakistan Journal of Zoology*, 1-4.

Ashwini M, Sathishkumar R. 2014. Onion (*Allium cepa*) Ethnomedicinal and therapeutic properties. *Handbook of Medicinal plants and their bioactive compounds*. 2734. Ed. Nidhi Gupta, Bharathiar University, India.

Awoniran, PO, Adeyemi DO. 2018. Ethanol extract of *Curcuma longa* rhizome mitigates potas-sium bromate-induced liver changes in the Wistar rat: Histological, histochemical and immunohistochemical assessments. *Morphologie*, 102: 276-288.

Bao L, Yao XS, Tsi D, Yau CC, Chia CS, Nagai H, *et al*. 2008. Protective effects of bilberry (*Vaccinium myrtillus L.)* extract on $KBrO_3$-induced kidney damage in mice. *Journal of Agricultural and Food Chemistry*, 56: 420-425.

Bayomy NA, Soliman GM, Abdelaziz EZ. 2016. Effect of potassium bromate on the liver of adult male albino rat and a possible protective role of Vitamin C: Histological,

immunohistochemical, and biochemical study. *The Anatomical Record*, 299: 1256-69.

Ben Saad H, Driss D, Ben Amara I, Boudawara O, Boudawara T.*et al.* 2016. Altered hepatic mRNA expression of immune response-associated DNA damage in mice liver induced by potassium bromate: Protective role of vanillin. *Environmental Toxicology*, 21: 10-22181.

Branen AL, Davidson PM, Salminen S, Thorngate J. 2001. Food additives. New York: Taylor & Francis.

Breu W. 1996. *Allium cepa* (onion). Part 1. Chemistry and analysis. *Phytomedicine*, 3: 293-306.

Brown DL, Walling BE, Mattix ME. 2016. Urinary System. In: Atlas of Histology of the Juvenile Rat. GA Parker, and CA Picut (eds). Elsevier, *Amsterdam*, 395-421.

Budavari S, O'Neil MJ, Smith A, Heckelman PE. 1989. The Merck index: an encyclopedia of chemicals, drugs and biologicals. 11th ed. Rahway, NJ: Merck and Company, Inc.

Bull RJ, Muralidhara S, Quinones O, Lim KY, Guo ZX, *et al*. 2012. Absorption and disposition of bromate in female F344 rats. *Toxicology*, 300:83-91

Calderon-Montaño JM, Burgos-Moron E, Perez-Guerrero C, Lopez-Lazaro M. 2011. A review on the dietary flavonoid kaempferol. *Journal of Medicinal Chemistry*, 11(4): 298-344.

Campbell K. 2006. Bromate-induced ototoxicity N, *Toxicology*, 221: 205-211.

Cavanagh JE, Weinberg HS, Gold A, Sangalah R, Marbury D. *et al*. 1992. Ozonation by-products: Indentification of bromohydrins from the ozonation of natural waters with enhanced bromide levels. *Environmental Science & Technology*, 26: 1658-62.

Cellai F, Bonassi S, Cristaudo A, Bonotti A, Neri M, *et al*. 2020. Chromatographic detection of 8-hydroxy-2′-deoxyguanosine in leukocytes of asbestos exposed workers for assessing past and recent carcinogen exposures. *Diagnostics*, 10(4): 239

Chauhan D, Jain P .2016. A scientific study of genotoxic-carcinogenic impacts of potassium bromate as food additive on human health.

International Research *Journal of Engineering and Technology*, 3: 1136-1139.

Chipman JK, Davies JE, Parsons JL, Nair J, O'Neill G, *et al*. 1998. DNA oxidation by potassium bromate, a direct mechanism or linked to lipid peroxidation. *Toxicology*, 126: 93-102.

Chyun JH, Park HJ, Yim JE. 2013. Onion peel extracts have hepatoprotective effects and decrease blood levels of LDL cholesterol in liver injured rats. *Federation of American Societies for Experimental Biology*, 27.

Circu ML, Aw TK. 2011. Redox biology of the intestine. *Free Radical Research*, 45: 1245-1266.

Collins J, Badireddy M. 2019. Anatomy, Abdomen and Pelvis, Small Intestine. Treasure Island: StatPearls Publishing,.

Corzo-Martínez M, Corzo N, Villamiel M. 2007. Biological properties of onions and garlic. *Trends in food science & technology*, 18: 609-625.

Costantini F, Kopan R. 2010. Patterning a complex organ: branching morphogenesis and nephron segmentation in kidney development. *Development Cell*, 18: 698-712.

Cotton FA, Wilkinson G, Gaus PL. 1987. *Basic Inorganic Chemistry*, 2nd edition. Wiley, New York, NY.

De Vriese A, Vanholder R, Lameire N. 1997. Severe acute renal failure due to bromate intoxication: report of a case and discussion of management guidelines based on a review of the literature. *Nephrology Dialysis Transplantation*, 12(1): 204-9.

DeAngelo AB, George MH, Kilburn SR, Moore TM, Wolf DC. 1998. Carcinogenicity of potassium bromate administered in the drinking water to male B6C3F1 mice and F344/N rats. *Toxicologic Pathology*, 26(5): 587-594.

Delaney MA, J Kowalewska, PM Treuting. 2018. Urinary system. In: Treuting PM, Dintzis SM, editors. Comparative anatomy and histology A mouse and human atlas. 2nd ed. Oxford. *Academic Press*, 275–30.1

Delker D, Hatch G, Allen J, Crissman B, George M, *et al*. 2006. Molecular biomarkers of oxidative stress associated with bromate carcinogenicity. *Toxicology*, 221(2-3): 158-165.

DeSesso JM, Jacobson CF. 2001. Anatomical and physiological parameters affecting gastrointestinal absorption in humans and rats. *Food and Chemical Toxicology*, 39: 209-228.

Dhembare AJ, Dale PG. 2017. Potassium bromate induced hematological alteration in European rabbit. *The Journal of Zoology Studies*, 4(3): 01-05

Dimkpa U, Ukoha U, Anyabolu E, Uchefuna R, Anikeh L, *et al*. 2013. Hepatotoxic Effects of Potassium Bromate on Adult Wistar Rats. *Journal of Biology, Agriculture and Healthcare*, 3(7): 111-115

Dunsky I. 1947. Potassium bromate poisoning. *The American Journal of Diseases of Children*, **7**4: 730-734.

El-Sokkary G H. 2000. Melatonin protects against oxidative stress induced by the kidney carcinogen $KBrO_3$. *Neuroendocrinology Letters*, 21(6): 461-468.

Elssaidi MA, Alhadad AS, Mohamed MY. 2013. Detection of potassium bromate in some bakery products in southern region of Libya. *Journal of Pure & Applied Sciences*, 12(3): 16-66

EPA US. 2001. Toxicological review of bromate, EPA/635/R-01/002. Washington, DC: United States Environmental Protection Agency.

FAO .2009. Food and agriculture organization. Production and trade statistics.

FAO 2015. Food and agriculture organization statistical pocketbook on world food.

Fawell J, Walker M. 2006. Approaches to determining regulatory values for carcinogens with particular reference to bromate. *Toxicology*, 221(2-3): 149-153.

Fennema OR. 1987. Food additive-an unending controversy. *The American Journal of Clinical Nutrition*, 46: 201-203.

Fennir MA. 2014. Potential of unrefrigerated storage of onions in the western mountain (wm) region of Libya. 2nd International Conference on Sustainable Environment and Agriculture *IPCBEE*, 76: 26-30, IACSIT Press, Singapore.

Fisher N, Hutchinson JB, Hardy J, Ginocchio AV, Waite V. 1979. Long-term toxicity and carcinogenicity studies of the bread improver potassium bromate. 1. Studies in rats. *Food and Cosmetics Toxicology*, 17: 33-39.

Foligne B, Aissaoui S, Senegas-Balas F, Cayuela C, Bernard P, *et al*. 2001. Changes in cell proliferation and differentiation of adult rat small intestine epithelium after adrenalectomy: kinetic, biochemical, and morphological studies. Digestive diseases and sciences, 46: 1236-1246

Fujie K, Shimazu H, Matsuda M, Sugiyama T. 1988. Acute cytogenetic effects of potassium bromate on rat bone marrow cells in vivo. *Mutation Research*, 206: 455-458.

Ganong WF, 2010. Ganong's review of medical physiology. 23rd edition. Kim Barrett, Heddwen Brooks, Scott Boitano, Susan Barman. Mc Graw Hill. New York, Chicago, San Francisco. PP: 430

George SE, Wolf DC, Brooks IR, Bailey KC, Hooth MJ, *et al*. 2004. Changes in cecal microbial metabolism of rats induced by individual and a mixture of drinking water disinfection by-products. *Cancer Letters*, 204: 15-21.

Gin L, Iqbal M, Athar M. 1999. Potassium bromate ($KBrO_3$) induces renal proliferative response and damage by elaborating oxidative stress. *Cancer Letters*, 135: 181-188.

Gokce AF, Cemal K, Serçe S, Özgen M. 2010. Effect of scale color on the antioxidant capacity of onions. *Scientia Horticulturae Amsterdam,* 123: 431-435.

Gowda S, Desai PB, Kulkarni SS, Hull VV, Math AAK, *et al*. 2010. Markers of renal function tests. *North American Journal of Medicine and Science*, 2:170-173

Gradus (Ben-Ezer) D, Rhoads M, Bergstrom LB. Jordan. S.C.1984. Acute bromate poisoning associated with renal failure and

deafness presenting as hemolytic uremic syndrome. *American Journal of Nephrology,* 4: 188-191.

Gulcin I. 2020. Antioxidants and antioxidant methods: an updated overview. *Archives of Toxicology*, 1-65.

Gulsen, A, makris DP, kefalas P. 2007. Biomimetic oxidation of quercetin: Isolation of naturally occurring quercetin heterodimer and evaluation of its in vitro antioxidant properties, *Food Research International*, 40: 7-14.

Haidari F, Rashidi MR, Eshraghian *MR, et al*. 2008. Hypouricemic and antioxidant activities of *Allium cepa* Lilliaceae and quercetin in normal and hyperuricemic rats. *Saudi Medical Journal*. 29(11):1573-1579.

Halliwell B. 1996. Antioxidants in human health and disease. *Annual review of nutrition,* 16: 33-50.

Hamada F, Ohzono S. Yamada S, Baba Y, Tokuda Y, Yamashita W, *et al*. 1990. A case of acute potassium bromate intoxication. *Fukuoka Igaku Zasshi*, 81: 271-276.

Health Canada 2016. Guidelines for canadian drinking water quality: Guideline Technical Document Bromate. Water and air quality bureau, healthy environments and consumer safety branch, Health Canada, Ottawa, Ontario.

Hosoyamada Y, Sakai T. 2005. Structural and mechanical architecture of the intestinal villi and crypts in the rat intestine: integrative reevaluation from ultrastructural analysis. *Anatomy and Embryology (Berl),* 210:1-12.

Hryn VH, Kostylenko YP, Bilash VP, Ryabushko OB. 2019. Microscopic structure of albino rats' small intestine. *Wiadomosci lekarskie (Warsaw, Poland: 1960)*, 5(72), 733-738.

Hryn V, Kostylenko Yu, Yushchenko Yu. *et al*. 2018. Comparative histological structure of the gastrointestinal mucosa in human and white rat: a bibliographic analysis. Wiadomości Lekarskie, 7:1398-1403

Hu R, McDonough A.A, Layton AT. 2019. Functional implications of the sex differences in transporter abundance along the rat

nephron: modeling and analysis. *American Journal of Physiology*, 317 (6): F1462-F1474

Hur SJ, Lee SJ, Kim DH, Chun SC, Lee SK. 2013. Onion extract structural changes during in vitro digestion and its potential antioxidant effect on brain lipids obtained from low- and high-fat-fed mice. *Free Radical Research*, 47(12): 1009-1015.

IARC 1986. IARC Monographs on the evaluation of the carcinogenic risk of chemicals to humans, Some naturally occurring and synthetic food components, Furocoumarins and Ultraviolet Radiation, Lyon, 40: 207-220

Ige SF, Adeniyi MJ, Iyalla GO. 2017. *Allium cepa* mitigates aluminum chloride-induced hepatotoxicity in male Wistar rats. *Journal of Biomedical Science*, 6(4): 27.

Ige SF, Akhigbe RE, Edeogho O, Ajao FO, Owolabi OQ, *et al*. 2011. Hepatoprotective activities of *Allium cepa* in cadmium-treated rats. *International Journal of Pharmacy and Pharmaceutical Sciences*, 3: 60-63.

Ige SF, Akhigbe RE. 2012. The role of *Allium cepa* on aluminuminduced reproductive dysfunction in experimental male rat models. *Journal of Human Reproductive Sciences*, 5(2): 200-205.

Ige SF, Salawu EO, Olaleye SB, Badmus J, Adeleke AA. 2009. Onion (*Allium cepa*) extract prevent cadmium induced renal dysfunction. *Indian Journal of Nephrology*, 19: 140-4.

Isoda T, Nakatsu Y, Yamauchi K, Piao J, Yao T, Honda H, Nakabeppu Y, Tsuzuki T, 2014. Abnormality in Wnt signaling is causatively associated with oxidative stress-induced intestinal tumorigenesis in MUTYH-null mice. *International journal of biological sciences*, 10(8): 940.

Jaffar SK, Khasim SM, Prasad MSK, 2020 Protective Effect of *Mimusops elengi L*. on Renal and Hepatic Markers in STZ-Induced Diabetic Rats. In: Khasim SM, Long C, Thammasiri, Lutken H. (eds) *Medicinal Plants:* Biodiversity, Sustainable Utilization and Conservation, 509-520.

Jaiswal N, Kumar D, Rizvi SI, Wellness H. 2013. Red onion extract (*Allium cepa L*) supplementation improves redox balance in oxidatively stressed rats. *Food Science,* 2: 99-104

Jansen T., Claassen, L., van Kamp, I., & Timmermans, D. R. 2020. All chemical substances are harmful. Public appraisal of uncertain risks of food additives and contaminants. Food and Chemical Toxicology, 136, 110959.

JECFA 1989. FAO/WHO Joint Expert Committee on Food Additives Toxicological evaluation of certain food additives and contaminant. Series No. 24 Cambridge University Press, Cambridge.

Josiah SJ, Nwangwu SCO, Akintth VA, Vivian A, Eiifeta GO, *et al*. 2011. Protective role of water extract of unripe pulp *of Canca papaya* (fruit) against potassium bromate induced tissue damage in Wistar rats. *British Journal of Pharmacology*, 12: 205-208.

Karbownik M, Stasiak M, Zygmunt A, Zasada K, Lewiński A. 2006. Protective effects of melatonin and indole-3-propionic acid against lipid peroxidation, caused by potassium bromate in the rat kidney. *Cell Biochemistry and Function: Cellular biochemistry and its modulation by active agents or disease*, 24(6): 483-489.

Kawanishi S, Murata M. 2006. Mechanism of DNA damage induced by bromate differs from general types of oxidative stress, *Toxicology*, 221: 172-178.

Khale S, McMahon B, Otto-Hitt S. 2018. Effect of potassium bromate on OXR1 gene expression and cell growth in tetrahymena thermophila. *Life and Environmental Sciences Course-based Research Projects*, 18.

Khan N, Sharma S, Alam A, Saleem M, Sultana S. 2001. *Tephrosia purpurea* ameliorates N-diethylnitrosamine and potassium bromate-mediated renal oxidative stress and toxicity in Wistar rats, *Journal of toxicology and pharmacology*, 88: 294-299.

Khan N, Sharma S, Sultana S. 2003. Nigella Sativa (black cumin) ameliorates potassium bromate–include early events of carcinogenesis: diminution of oxidative stress". *Toxicology and Industrial Healthl*, 22(4): 193-203.

Khan N, Sultana S. 2005a. Modulatory effect of ficus racemosa: diminution of potassium bromate-induced renal oxidative injury and cell proliferation response. *Basic and Clinical Pharmacology and Toxicology,* 97: 282-288.

Khan N, Sultana S. 2005b. Inhibition of potassium bromateinduced renal oxidative stress and hyperproliferative response by *Nymphaea alba* in Wistar rats, *Journal of Enzyme Inhibition and Medicinal Chemistry*, 20:3, 275-283.

Khan N. 2003. *Nigella sativa* (black cumin) ameliorates potassium bromate-induced early events of carcinogenesis: diminution of oxidative stress. *Human & Experimental Toxicology* 22:193-203.

Khan RA, Khan MR, Sahreen S. 2012a. Protective effects of rutin against potassium bromate induced nephrotoxicity in rats. *BMC Complementary and Alternative Medicine*, 12:204.

Khan RA, Khan MR, Sahreen S. 2012b. Protective effects of Sonchus asper against KBO_3 induced lipid peroxidation in rats. *Lipids in Health and Disease*, 11: 367-374.

Khan RA. 2017. Effect of launaea procumbens on thyroid glands lipid peroxidation and hormonal dysfunction: a randomized control trial. *Lipids In Health And Disease*.16:168-173.

Kim JM, Kim JS, Park EJ .2013. Antioxidative and antigenotoxic effects of onion peel extracts in non-cellular and cellular systems. *Food Science and Biotechnology*, 22(5): 1-8

Ko EY, Nile SH, Sharma K, Li GH, Park SW. 2014. Effect of different exposed lights on quercetin and quercetin glucoside content in onion (*Allium cepa L.*). *Saudi Journal of Biological Sciences,* 22(1): 398-403.

Krinke GJ. 2000. The handbook of experimental animals: The laboratory rat. In: Bullock G, Bunton TE, editors. New York: Academic Press.

Kumar S, Pandey AK. 2013. Chemistry and biological activities of flavonoids: an overview. *The Scientific World Journal*, 162750, 16.

Kurata, Y, Diwan BA, Ward JM. 1992. Lack of renal tumor-initiating activity of single dose of potassium bromate, a genotoxic renal carcinogen in male Fischer 344/NCr rats. *Food and Chemical Toxicology*, 30: 251-259.

Kurokawa Y, Aoki S, Matsushima Y, Takamura N, Imazawa T, Hayashi Y. 1986. Dose-response studies on the carcinogenicityof potassium bromate in F344 rats after long-termoral administration. *Journal of the National Cancer Institute*. 77: 977-982.

Kurokawa Y, Havashi Y, Maekawa A, Takahashi M, Kokubo T, Odashuna S. 1983. Carcinogenicity of potassium bromate administered orally to F344 rats. *Journal of the National Cancer Institute*, 71: 965-972.

Kurokawa Y, Maekawa A, Takahashi M, Hayashi, Y. 1990. Toxicity and carcinogenicity of potassium bromate - a new renal carcinogen. *Environmental Health Perspectives*, 87: 309-335.

Kutom A, Baziliski NG, Magana L, Dunea G. 1990. Bromate intoxication: Hairdresser's anuria. *American Journal of Kidney Diseases*.15: 84-85.

Kwiecien S, Brzozowski T, Konturek SJ. 2002. Effects of ROS action on gastric mucosa in various models of mucosal injury. *Journal of Physiology and Pharmacology*, 53: 39-50.

Lanzotti V. 2006. The analysis of onion and garlic. *Journal of chromatographya*, 1112(1-2): 3-22.

Lee B, Jung J-H, Kim H-S. 2012. Assessment of red onion on antioxidant activity in rat. *Food Chemical Toxicology*, 50: 3912-3919.

Lewis R, J Sr. 1991. Hazardous Chemicals Desk Reference, 2nd Edition. New York: Van Nostrand Reinhold.

Mahomoodally M, Gurib-Fakim A, Subratty AH. 2005. Antimicrobial activities and phytochemical profiles of endemic medicinal plants of Mauritius. *Pharmaceutical Biology*, 43: 237-42.

Matsumoto I. 1973. Clinical and experimental studies on the toxicity of bromate. *Hotel. Fukuoka*, 19: 220-236.

Matsumoto I, Morizono T, Paparella MM 1980. Hearing loss following potassium bromate: two case reports. *Otolaryngology–Head and Neck Surgery*, 88:625-629.

Maurya H, Kumar T, Kumar S. 2018. Anatomical and physiological similarities of kidney in different experimental animals used for basic studies. *Journal of Clinical & Experimental Nephrology*, 3:9-14.

Maynard RL, Downes N. 2019. Anatomy and Histology of the Laboratory Rat in Toxicology and Biomedical Research. Academic Press, London. 359.

McDermott JH. 2000. Antioxidant nutrients: current dietary recommendations and research update. Journal of the American Pharmaceutical Association (Wash), 40(6):785-99.

McDorman KS, Pachkowski BF, Nakamura J, Wolf DC, Swenbcrg JA. 2005. Oxidative DNA damage from potassium bromate exposure in Long-Evans rats is not enhanced by a mixture of drinking water disinfection by-products. *Chemico-Biological Interactions*, 152:107-117.

McMinn S. 2017. The Asylum. Lulu.com. P 223

Metrani R, Singh J, Acharya P, K Jayaprakasha G, S Patil B. 2020. Comparative Metabolomics Profiling of Polyphenols, Nutrients and Antioxidant Activities of Two Red Onion (*Allium cepa L*.) Cultivars. *Plants*, 9: 1077.

Milan Chandal, Gyan CJ. 2016. Manganese-induced hematological alteration in Wister rats. *Journal of Environmental and Occupational Science,* 5(4): 77-81.

Mohamed EAK, Saddek EA. 2019. The protective effect of taurine and/or vanillin against renal, testicular, and hematological alterations induced by potassium bromate toxicity in rats. *The Journal of Basic and Applied Zoology*, 80(1):3.

Montgomery CA, Seely JC. 1990. Kidney. In Pathology of the Fisher Rat. Reference and Atlas (GA Boorman, SL. Eustis MR. Elwell CA. Montgomery, WF. MacKenzie, eds.). Academic press, San Diego, CA, 127-53.

Moubarak HS, Essawy TA, Mohammed SS. 2020. Carcinogenic effect of potassium bromate on tongue of adult male albino rats. *Journal of Radiation Research and Applied Sciences*, 13(1): 121-131.

Murata M, Bansho Y, Inoue S, Ito K, Ohnishi S, *et al*. 2001. Requirement of glutathione and cysteine in guanine-specific oxidation of DNA by carcinogenic potassium bromated. *Chemical Research in Toxicology*, 14: 678-685.

Nakano K. 1989. Renal changes induced by chronic oral administration of potassium bromate on ferric nitrilotriacetate in Wister rats. *Japanese archives of internal medicine*, 36: 41-47.

Nakatsuji S, Szabo KA, Elmore SA. 2018. Small and large intestine. In: Suttie AW, ed. Boorman's Pathology of the Rat. 2nd ed. Boston, MA: Academic Press, 51-69. Chap 7.

Nasri S, Anoush M, Khatami N. 2012. Evaluation of analgesic and anti inflammatory effects of fresh onion juice in experimental animals. *African Journal of Pharmacy and Pharmacology*. 6(23): 1679 -1684.

Neha K, Haider MR, Pathak A, Yar MS. 2019. Medicinal prospects of antioxidants: a review. *European Journal of Medicinal Chemistry*, 178: 687-704

Nikmaneshi MR, Firoozabadi B, Munn LL. 2020. A mechanobiological mathematical model of liver metabolism. *Biotechnology and Bioengineering*, 117(9): 2861-2874.

Nile SH, Park SW. 2013. Total phenolics, antioxidant and xanthine oxidase inhibitory 531 activity of three colored onions (*Allium cepa L.). Frontiers in Life Science*, 7: 224-228.

Niwa T, Ito T, Matsui E. 1974. Serial renal biopsy in potassium bromate intoxication. *Japanese circulation journal,* 38: 387-392.

N0rris JA.1965. Toxicity of home permanent waving and neutralizer solutions. *Food and cosmetics toxicology*, 3: 93-97

Nuutila AM, Puupponen-Pimiä R, Aarni M, Oksman-Caldentey KM. 2003. Comparison of antioxidant activities of onion and garlic extracts by inhibition of lipid peroxidation and radical scavenging activity. *Food chemistry*, 81(4): 485-493.

Nwonuma C, Irokanulo E, Iji C, Alejolowo O, Adetunji C. 2016. Effect of thaumatococcus daniellii leaf rat-feed on potassium bromate induced testicular toxicity. *Asian Pacific Journal of Reproductio,* 5: 500-5

Nzalak JO, Wanmi N, Imam J, Ali M, AD. 2015. Anatomical and Histological Studies of the Small Intestine of the African Giant Rat (Cricetomys gambianus-Water house)-II. Annals of Experimental Biology, 3 (4):20-26.

Obaidi I, Higgins M, Bahar B, Davis JL, McMorrow T. 2018. Identification of the multifaceted chemopreventive activity of curcumin against the carcinogenic potential of the food additive, $KBrO_3$. *Current pharmaceutical design,* 24: 595-614.

Ogbonna OJ, Udia PM, Abe PN, Omoregha CU, Anele EI. 2016. Phytochemical and proximate analysis, mineral and vitamin compositions of *Allium Cepa* bulb extract. *Advances in Biomedicine and Pharmacy*, 3(4): 181-186.

Ogunmodede O, Saalu L, Ogunlade B, Akunna G, Oyewopo A. 2012. An evaluation of the hypoglycemic, antioxidant and hepatoprotective potentials of onion (*Allium cepa L*.) on alloxan-induced diabetic rabbits. *International journal of pharmacology*, 8: 21-29.

Ohashi N, Shiba T, Kamiya K, Takamura T. 1971. Acute renal failure following potassium bromate (‘cold wave’ neutralizer) poisoning (recovery from prolonged oliguria. *The Japanese Journal of Urology*, 62: 639-646.

Ola-Mudathir KF, Suru SM, Fafunso MA, Obioha UE, Faremi TY. 2008. Protective roles of onion and garlic extracts on cadmium-induced changes in sperm characteristics and testicular oxidative damage in rats. *Food and Chemical Toxicology*, 46(12): 3604-3611.

Oloyede OB, Sunmonu TO. 2009. Potassium bromate content of selected bread samples in Ilorin, Central Nigeria and its effect on some enzymes of rat liver and kidney *Food and chemical toxicology*, 47: 2067-2070.

Oyekunle JAO, Adekunle AS, Ogunfowokan AO, Olutona GO, Omolere OB. 2014. Bromate and trace metal levels in bread loaves from outlets within Ile-Ife Metropolis, Southwestern Nigeria. *Toxicology*, 1:224-230.

Ozougwu JC, Eyo JE. 2014. Hepatoprotective effects of *Allium cepa* (onion) extracts against paracetamol-induced liver damage in rats. *African Journal of Biotechnology*, 13(26): 2679-2688.

Pagewise I. 2002. The history of bread: A recipe for wonderful bread making. *Truester Health Encyclopedia*, 3: 1-5.

Pareek S, Kumar V, Sharma S, Sagar NA. 2017. Onion (*Allium cepa L.*). *Food Chemistry*, 52(5): 1174-1131.

Park KY, Choi SY, Lee SH, Kweon JH, Song JH. 2016. Comparison of formation of disinfection by-products by chlorination and ozonation of wastewater effluents and their toxicity to Daphnia magna. *Environmental Pollution*, 215: 314-321.

Park SK, Jin DE, Park CH, Seung TW, Guo TJ, *et al*. 2015. Ameliorating effects of ethyl acetate fraction from onion (*Allium cepa L.*) flesh and peel in mice following trimethyltin-induced learning and memory impairment. *Food Research International*, 75: 53-60.

Parker WA, Barr JR. 1951. Potassium bromate poisoning, *British medical journal*, 16:1(4719):1363-1364.

Parsons J.L, Chipman JK. 2000. The role of glutathione in DNA damage by potassium bromate in vitro. *Mutagenesis*, 15: 311-316.

Perez-Gregorio M.R, Regueı`ro J, Gonza´lez-Barreı´ro C, Rı´alOtero R, Sı´mal-Ga´ndara J. 2011. Changes in antioxidant flavonoids during freeze-drying of red onions and subsequent storage. *Food Control*, 22: 1108-1113.

Pérez-Gregorio RM, García-Falcón MS, Simal-Gándara J, Rodrigues AS, Almeida DP. 2010. Identification and quantification of flavonoids in traditional cultivars of red and white onions at harvest. *Journal of Food Composition Analysis*, 23: 592-8.

Pfaller W. 2012. Structure Function Correlation on Rat Kidney: Quantitative Correlation of Structure and Function in the Normal and Injured Rat Kidney. New York: Springer Science & Business Media.

Piao J, Nakatsu Y, Ohno M, Taguchi K.I, Tsuzuki T, 2014. Mismatch repair deficient mice show susceptibility to oxidative stress-induced intestinal carcinogenesis. *International journal of biological sciences*, 10(1):73.

Prakash D, Singh BN, Upadhyay G, 2007. Antioxidant and free radical scavenging activities of phenols from onion (*Allium cepa*). *Food Chemistry,* 102: 1389-1393.

Pressman P, Clemens R, Hayes W, Reddy C. 2017. Food additive safety: A review of toxicologic and regulatory issues. *Toxicology Research*, 1: 1-22

Qin J, Kim MS, Chao K, Dhakal S, Cho BK.2018. Non-targeted and targeted raman imaging detection of chemical contaminants in food powders, Proc. *Sensing for Agriculture and Food Quality and Safety X*, 10665: 106650G

Quick CA, Chole RA, Mauer SM. 1975. Deafness and renal failure due to potassium bromate poisoning. Arch. *Otolaryngology*, 101: 494-495.

Rahman A, Ahmed S, Khan N, Sultana S, Athar ill. 1999. Glycerol trinitrate, a nitric oxide donor, suppresses renal oxidant damage caused by potassium bromate. *Redox Report,* 4: 263-269.

Rahmat RF, Adnan S, Anugrahwaty R, Alami EPS, Siregar B. 2019. Red onion growth monitoring system in hydroponics environment. *In Journal of Physics: Conference Series*, 1235(1): 012117.

Rao JN, Wang JY. 2010. Intestinal Architecture and Development. In Regulation of Gastrointestinal Mucosal Growth. San Rafael (CA): Morgan & Claypool Life Sciences.

Rashmi HB, Negi PS. 2020. Phenolic acids from vegetables: A review on processing stability and health Benefits. *Food Research International*, 136: 109298

Reed KK, Wickham R. 2009. Review of the gastrointestinal tract: from macro to micro. *Seminars in Oncology Nursing*, 25(1): 3-14.

Rezaei-Sadabady R, Eidi A, Zarghami N, Barzegar A. 2016. Intracellular ROS protection efficiency and free radical-scavenging activity of quercetin and quercetin-encapsulated liposomes. Artificial cells, nanomedicine, *biotechnology* 44: 128-34.

Rezq A. 2017. Potential protective and ameliorate effects of sesame oil and jojoba oil against potassium bromate ($KBrO_3$)-induced oxidative stress in rats. *Journal of Studies and Searches of Specific Education*, Faculty of Specific Education, Zagazig University. 3 (1):58-61.

Rose P, Whiteman M, Moore PK, Zhu ZY. 2005. Bioactive S-alk(en)yl cysteine sulphoxide metabolites in the genus Allium: The chemistry of potential therapeutic agents. *Natural Product Reports*, 22: 351-368.

Sai K, Tyson CA, Thomas DW, Dabbs JE, I Iasegawa R, Kurokawa Y .1994. Oxidative DNA damage induced by potassium bromate in isolated rat renal proximal tubules and renal nuclei. *Cancer Cell,* 87: 1-7.

Sands JM, Verlander W. 2010. Functional anatomy of the kidney. *Comprehensive Toxicology*, 7: 1-22

Santas J, Almajano MP and Carbo R. 2010. Antimicrobial and antioxidant activity of crude onion (*Allium cepa, L*) extracts. *International Journal of Food Science*, 45: 403-409.

Sarker U, Oba S. 2020. Nutritional and antioxidant components and antioxidant capacity in green morph Amaranthus leafy vegetable. *Scientific Reports*, 10, 1336.

Seely JC, Hard GC, Blankenship B. 2018. Kidney. In: Maronpot RR, Boorman GA, Gaul BW (eds) Pathology of the mouse. Reference and atlas. *Academic Press San Diego,* CA: 125-166

Seifried H E, Anderson DE, Fisher EI, Milner JA. 2007. A review of the interaction among dietary antioxidants and reactive oxygen species. *The Journal of Nutritional Biochemistry,* 18: 567-579.

Shahid F, Farooqui Z, Khan AA, Khan F. 2018. Oral Nigella sativa oil and thymoquinone administration ameliorates the effect of long-term cisplatin treatment on the enzymes of carbohydrate metabolism, brush border membrane, and antioxidant defense in

rat intestine. *Naunyn-Schmiedeberg's Archives of Pharmacology*, 391: 145-157.

Shahid F, Farooqui Z, Rizwan S, Abidi S, Parwez I. *et al*. 2017. Oral administration of Nigella sativa oil ameliorates the effect of cisplatin on brush border membrane enzymes, carbohydrate metabolism and antioxidant system in rat intestine. Exp. *Toxicologic Pathology*. 69: 299-306.

Shanmugavel V, Komala Santhi K, Kurup AH, Kalakandan S, Anandharajm A, *et al*. 2020. Potassium bromate: Effects on bread components, health, environment and method of analysis: A review. *Food Chem*, 311:125964.

Singh, BN, Singh BR, Singh RL, Prakash D, Singh DP, Sarma BK, *et al*. 2009. Polyphenolics from various extracts/fractions of red onion (*Allium cepa*) peel with potent antioxidant and antimutagenic activities. *Food and Chemical Toxicology*, 47: 450-452.

Slaoui M, Fiette L. 2011. Histopathology procedures: from tissue sampling to histopathological evaluation. *Methods in Molecular Biology,* 91:69-82

Slimestad R, Fossen T, Vagen IM. 2007. Onions: a source of unique dietary flavonoids. *Journal of agricultural and food chemistry*, 55: 10067-10080.

Spataru MC, Spataru C, Trofin AE, Hritcu LD, Zamfir CL, *et al*. 2019. Evaluation of the effect of red onion extract consumption in mice (Mus musculus). *Revista de Chimie*, 70(7): 2506-2510

Starek A, Starek-Świechowicz B. 2016. Toxicological properties of potassium bromate. *Journal of Pharmacological Reports*, 1, 1-9.

Stuti, M, D'Souza D. 2013. Effects of potassium bromate on the kidney and haematological parameters of Swiss albino mice. *The Bioscan. Supplement on Toxicology*, 8(3): 1011-1014.

Suleria HAR, Butt MS, Anjum FM, Saeed F, Khalid N. 2015. Onion: Nature protection against physiological threats. Critical reviews in food science nutrition, 55: 50-66.

Sultan MT, Butt MS, Ahmad RS, Pasha I, Ahmad AN, ayy'uIn MM. 2012. Supplementation of *Nigella sativa* fixed and essential oil mediates potassium bromate induced oxidative stress and

multiple organ toxicity. *Pakistan Journal of Pharmaceutical Sciences,* 25: 175-181.

Suru SM. 2008. Onion and garlic extracts lessen cadmium-induced nephrotoxicity in rats. *Biometals*, 21(6): 623-633.

Takamura N, Kurokawa Y, Matsushima Y, Imazawa T, Onodera H, *et al*. 1985. Long-term oral administration of potassium bromate in male Syrian golden hamsters. *The science reports of the research institutes*, 32(1-4): 43-46.

Tanaka K, Oikawa K, Fukuhara C, Saito H, Onosaka S, Min, K. S, *et al.* 1984. Metabolism of potassium bromate in rats II. In vitro studies. Chemosphere, 13(11): 1213-1219.

Teshika JD, Zakariyyah AM, Toorabally Z, Zengin G, Rengasamy KRR. *et al*. 2018. Traditional and modern uses of onion bulb (*Allium cepa L*.): A systematic review. *Critical reviews in food science and nutrition*, 1549-7852.

Treuting, PM, Dintzis SM, Montine K. 2018. Upper gastrointestinal tract. In Comparative Anatomy and Histology (Second Edition), A Mouse, Rat, and Human Atlas; Academic Press, Elsevier: London, UK, 190-211.

Uchida HA, Sugiyama H, Kanehisa S, Uchida HA, Sugiyama H, *et al*. 2006. An elderly patient with severe acute renal failure due to sodium bromate intoxication. *Internal medicine*, 45: 151-154.

Ugwoke CEC, Ezugwe CO. 2010. Phytochemical screening and proximate composition and onion bulb (*Allium cepa L*.). *Journal of Pharmaceutical and Allied Sciences*, 7: 1596-8499.

Ülger TG, Çakiroglu FP. 2020. The effects of onion (*Allium cepa L*.) dried by different heat treatments on plasma lipid profile and fasting blood glucose level in diabetic rats. *Avicenna journal of phytomedicine*, 10(4): 325-333

Ülger, T. G., & Çakiroglu, F. P. 2020. The effects of onion (*Allium cepa L*.) dried by different heat treatments on plasma lipid profile and fasting blood glucose level in diabetic rats. Avicenna journal of phytomedicine, 10(4), 325–333

Ullah H, Khan A, Baig MW, Ullah N, Ahmed N, *et al*. 2020. Poncirin attenuates CCL4-induced liver injury through inhibition of

oxidative stress and inflammatory cytokines in mice. *BMC Complementary Medicine and Therapies*, 20(115):1-14

Umemura T, Kitamura Y, Kanki K, Maruyama S, Okazaki K, *et al*. 2006. Dose-related changes of oxidative stress and cell proliferation in kidneys of male and female F344 rats exposed to potassium bromate. *Cancer Science*, 95: 393-398.

Umemura, T, Kurokawa Y. 2006. Etiology of bromate-induced cancer and possible modes of action-studies in Japan. *Toxicology*, 221(2-3): 154-157.

Umemura T, Kitamura V, Kanki K, Maruyama S. Okazaki K, Imazawa T, *et al*. 2004. Dose-related changes of oxidative stress and cell proliferation in kidneys of male and female 1:344 rats exposed to potassium bromate. Cancer Science, 95: 393-398.

Valko M, Leibfritz D, Moncol J, Cronin NIT, Mazur NI, Telser J. 2007. Free radicals and antioxidants in normal physiological functions and human disease. *The International Journal of Biochemistry & Cell Biology*, 39: 44-84.

Vertuani S, Angusti A, Manfredini S. 2004. The antioxidants and pro-antioxidants network: an overview. *Current Pharmaceutical Design*, 10:1677-94.

Vlase L, Parvu M, Parvu EA, Toiu A. 2013. Chemical constituents of three allium species from Romania. *Molecules,* 18: 114-127.

Wang Y, Zhao H, Liu J, Shao Y, Li J, *et al*. 2018. Copper and arsenic-induced oxidative stress and immune imbalance are associated with activation of heat shock proteins in chicken intestines. *International Immunopharmacology*. 60: 64-75.

Watanabe T, Abe T, Satoh M, *et al*. 1992 Two children with bromate intoxication due to ingestion of the second preparation for permanent hair waving. *Acta Paediatrica Japonica,* 34(6): 601-605.

Wei M, Hamoud A.S, Yamaguchi T, Kakehashi A, Morimura K, *et al*. 2009. Potassium bromate enhances N-ethyl-N-hydroxyethylnitrosamine-induced kidney carcinogenesis only at high doses in Wistar rats: Indication of the existence of an enhancement threshold. *Toxicologic Patohology*, 37: 983-991.

WHO. 2005. Bromate in drinking-water: background document for development of WHO guidelines for drinking-water quality, WHO/SDE/WSH/05.08/78, World Health Organization. 85.

Wilson BG, Bahna SL. 2005. Adverse reactions to food additives. *Annals of Allergy, Asthma & Immunologyl*, 95(6):499-507.

Wolf DC, Crosby LM, George MH, Kilburn SR, Moore TM, *et al*. 1998. Time and dose-dependent development of potassium bromate-induced tumors in male Fischer 344 rats. *Toxicoloic phathology,* 26: 724-729.

Yamazaki Y, Iwasaki K, Mikami M, Yagihashi A, 2011. Distribution of eleven flavor precursors, S-Alk(en)yl-L-cysteine derivatives, in seven allium vegetables. *Journal of Food Science and Technology*, 17: 55-62.

Young IS, Woodside IS. 2001. Antioxidants in health and disease. *Journal of Clinical Pathology*, 54*:*176-186.

Young Y, Chuu J, Liu S, Lin-Shiau S. 2001. Toxic effects of potassium bromate and thioglycolate on vestibuloocular reflex systems of guinea pigs and humans, *Toxicology and Applied Pharmacology*, 177103-111.

Zhang SL, Deng P, Xu YC, Lü SW, Wang JJ. 2016. Quantification and analysis of anthocyanin and flavonoids compositions, and antioxidant activities in onions with three different colors. *Integrated Agricultur*e, 15(9): 2175-2181.

Zhang Y, Jiang I, Mang I, Gcng C, I.i I, *et al*. 2011. Possible involvement of oxidative stress in potassium bromate-induced genotoxicity in human I lepG2 cells. *Chemico-Biological Interactions,* 189: 186-191.

Zhang X, De Silva D, Sun B, Fisher J, Bull RJ, *et al*. 2010. Cellular and molecular mechanisms of bromate-induced cvtotoxicity in human and rat kidney cells. *Toxicology*, 269: 13-23.

Zhang S. 1999. An atlas of histology, First edition.0-387-94954-2.

الملخص

يعتبر برومات البوتاسيوم $KBrO_3$ عاملا مؤكسدا قويا يدخل في العديد من الصناعات أهمها إستخدامه كمحسن غدائي للخبز. على الرغم من أن الآثار السلبية ليست واضحة في الحيوانات التي تتغذى على الوجبات الغذائية المصنوعة من الدقيق المعالج بـالبرومات، إلا ان العامل يصنف على أنه مادة مسرطنة لحيوانات التجارب ومسبب للسمية الكلوية في كل من الإنسان والحيوانات عند إعطائه عن طريق الفم. يتم توجيه اهتمام كبير إلى العديد من النباتات بسبب احتوائها على مضادات الأكسدة، من بينها البصل الأحمر، الذي يستخدم على نطاق واسع كغذاء وفي الطب التقليدي. البصل الأحمر له تأثيرات معروفة مفيدة مثل مضادات الأكسدة، ومضادات الأورام، ومضادات الالتهاب ومضادات الكوليسترول. تهدف الدراسة لاختبار فعالية عصير البصل الأحمر في تعديل التأثيرات السمية لبرومات البوتاسيوم على بعض التغيرات الدموية والنسيجية في ذكور الفئران البيضاء. استخدمت في هذه الدراسة ستة وثلاثين فأرا بالغا، قسمت عشوائيا إلى ستة مجاميع متساوية، حيث كانت المجموعة الأولى كمجموعة ضابطة. المجموعة الثانية عولجت بالبرومات (100 ملجم /كجم) عن طريق التجريع بالفم يومي 24 و27 من الاسبوع الرابع. والمجموعة الثالثة جرعت (1ملل/100جم) من عصير البصل الأحمر كل يوم طيلة التجربة بالإضافة لجرعة البرومات (100ملجم /كجم) يومي 24 و27 من الاسبوع الرابع. أما المجموعة الرابعة تلقت البرومات (50ملجم /كجم) مرتين كل أسبوع. المجموعة الخامسة استقبلت (1ملل/100جم) من عصير البصل الأحمر كل يوم و(50ملجم /كجم) من $KBrO_3$ مرتين كل أسبوع، أما المجموعة السادسة والأخيرة استقبلت (30 ملجم /كجم) من $KBrO_3$ كل يوم من التجربة التي استمرت لمدة أربعة أسابيع. وفي نهاية التجربة شرحت الحيوانات بعد وزنها وتم إستخراج كل من الكبد والكلى والقلب والرئتين وتسجيل أوزانهم، وإعداد مقاطع نسيجية لكل من الكلى والامعاء. ولقد أوضحت النتائج أن وزن هذه الأعضاء لا يختلف إختلاف كبير عن وزن الأعضاء بالمجموعة الضابطة. بينما أظهرت النتائج زيادة معنوية ($P < 0.05$) في خلايا الدم البيضاء WBC في المجموعات التي استقبلت البرومات (30&50 ملجم /كجم). ونقص معنوي ($P < 0.05$) في الصفائح الدموية PLT في المجموعة التي استقبلت (100 ملجم /كجم $KBrO_3$). أوضحت النتائج أيضا زيادة معنوية ($P < 0.05$) في اليوريا، الكيراتنين، حمض اليوريك، البوتاسيوم والصوديوم للمجموعات المعالجة بالبرومات مقارنة بمجموعة السيطرة. وأشارت النتائج الى وجود إرتفاع معنوي في فعالية الأنزيمات ALP و AST في المجموعات المعالجة بالبرومات مقارنة بمجموعة السيطرة .الا أن المجموعات التي عولجت بعصير البصل الأحمر قبل جرعة البرومات أظهرت تحسن في قراءات هذه الانزيمات وكذلك في

وظائف الكلى. أما نتائج الدراسة النسيجية للكلية فأظهرت إتساع في محفظة بومان، نزف (hemorrhage)، تحلل (degeneration)، إحتقان (congestion)، تنخر(necrosis) كذلك فقد (brush border). كذلك أوضحت نتائج التغيرات النسيجية في الأمعاء الدقيقة عدم وجود الزغب (villi) وزيادة في الخلايا الكأسية (goblet cells) في البطانة الظاهرية. كما لوحظ تسلل (infiltration) خلايا التهابية في معظم المجموعات التي عولجت ببرومات البوتاسيوم. من نتائج الدراسة الحالية يتضح أن الأثر السمي لبرومات البوتاسيوم كان أكثر في المجموعة التي استقبلت جرعة واحدة (100 ملجم /كجم $KBrO_3$) يومي 24 و27 من الاسبوع الرابع. وهذا يدل على أنه كلما زادت الجرعة زاد التأثير. كذلك فإن تأثير الجرعة اليومية (30 ملجم/كجم) من برومات البوتاسيوم تدل على خطورة التعرض اليومي للبرومات. كما أن للبصل الأحمر خصائص وقائية ضد الضرر المستحث ببرومات البوتاسيوم وذلك عن طريق تقليص التغيرات الفسيولوجية والنسيجية المرضية. وتقترح هذه النتائج أن التأثيرات الوقائية لهذه المواد يمكن أن تعزى إلى أنشطتها المضادة للأكسدة.

Publications

N. M. Kermani., **A. M**. **Aldaek**, F. A. Abushofa & Jaat, F. G. 2020. Protective Effect of *Allium cepa* L. (onion) against potassium bromate-induced hematological, biochemical and histopathological alterations in rats. *International Journal of Innovative Science and Research Technology*, 5 (11): 201-207.

N. M. Kermani, F. A. Abushofa, Jaat, F. G & **A. M**. **Aldaek**. 2021. Ameliorative Effect of *Allium Cepa* L. (Red Onion) Extract on Potassium Bromate Induced Intestinal Mucosal Injury in Experimental Rats. (Unpublished).