WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 5.990
Volume 4, Issue 12, 215-246. Research Article ISSN 2277– 7105

ANTIOXIDANT EFFECT OF RADISH (*RAPHANUS SATIVUS L.*) AND LEEK (*ALLIUM PORRUM L.*) JUICES AGAINST HEPATOTOXICITY AND NEPHROTOXICITY INDUCED BY DIMETHOATE IN MALE ALBINO MICE

Gehan Salah Eldin Moram*, Tahany El-sayed Kholief, Aziza Tarik Fathy Ahmed

Department of Biochemistry and Nutrition, Women's College Ain Shams University, Egypt.

Article Received on
02 Oct 2015,

Revised on 27 Oct 2015,
Accepted on 20 Nov 2015

***Correspondence for Author**
Dr. Gehan Salah Eldin Moram
Department of Biochemistry and Nutrition, Women's College Ain Shams University, Egypt.

ABSTRACT

Objectives: This study aimed to investigate the ameliorative effect of radish and leek (leaves and roots) juices against the damage induced by dimethoate (DM) on liver and kidney functions and oxidative stress in mice. **Methods:** DM was administered orally at a dose of 10mg/kg b.wt /day. Radish and leek (leaves and roots) juices were administered orally at a dose of 8ml/kg b.wt /day. Mice were divided into 10 groups: G1: healthy control, G2: radish leaves juice (healthy RLJ), G3: radish roots juice (healthy RRJ), G4: leek leaves juice (healthy LLJ), G5: leek roots juice (healthy LRJ), G6: dimethoate group (DM), G7: RLJ plus DM, G8: RRJ plus DM, G9: LLJ plus DM, G10: LRJ plus DM. **Results:** The results indicated that DM depleted the oxidative stress markers including reduced glutathione content (GSH), superoxide dismutase activity (SOD), and total thiol content (T.SH) and also significantly elevated malondialdehyde level (MDA), nitric oxide concentration (NO) and advanced oxidation protein products content (AOPP) in liver and kidney tissues. However, radish and leek (leaves and roots) juices significantly elevated the depleted oxidative stress markers including GSH, SOD and T.SH and also significantly reduced the marked elevation of MDA, NO and AOPP. Moreover, radish and leek juices significantly improved liver, kidney functions and hematological parameters in mice administered dimethoate. These results were confirmed by histopathological examination of liver and kidney. **In conclusion,** Radish and leek juices administration attenuated the severity of oxidative damage accompanying dimethoate toxicity with hepatoprotective and nephroprotective properties.

KEYWORDS: Radish, leek, dimethoate, antioxidant, hepatoprotective, nephroprotective.

INTRODUCTION

Organophosphorus insecticides (OPI) represent one group of pesticides that is widely used and has proved to have toxic effects for humans and animals.[1] Residual amounts of OPI have been detected in the soil, vegetables, grains and other food products.[2]

Dimethoate (O, O'-dimethyl S-methyl carbamoyl phosphorodithioate) is one of the most important OPI used extensively on a large number of crops against several pests. [3] It has been classified as moderately hazardous. [4] Numerous studies indicate that dimethoate intoxication can cause oxidative stress and induce hepatic lipid peroxidation in mice [5] and rats. [6] Liver plays a central role in the detoxification process and along with kidney faces the threat of maximum exposure to xenobiotics and their metabolic by-products. [7] Also studies showed that the primary site of action of most chemicals is presumed to be the proximal tubule and glomeruli in kidney. [8]

Natural antioxidants in fruits and vegetables, such as vitamins and polyphenols, are considered to be responsible for health benefits. As an important category of phytochemicals, phenolic compounds universally exist in plants, and have been considered to have high antioxidant ability and free radical scavenging capacity, with the mechanism of inhibiting the enzymes responsible for reactive oxygen species (ROS) production and reducing highly oxidized ROS. [9]

Accordingly, interest has recently grown in the role and usage of natural antioxidants as a strategy to prevent oxidative damage in various health disorders as a factor in their pathophysiology. [10, 11]

Radish (*Raphanus sativus L.*) is one of the members of the family Brassicaceae that contains broccoli, cauliflower, cabbage, and kale. [12] Radish contains wide array of polyphenolics such as catechin, protecatechuic acid, syringic acid, vanillic acid, ferulic acid, sinapic acid, o-coumaric acid, myricetin, and quercetin in the leaves and stem of R. sativus, thus it should be regarded as a potential source of natural antioxidant and have potential to be developed as an ingredient in health or functional food. [13]

Raphanus sativus is used worldwide for its culinary and medicinal properties especially as a laxative. There have been reports of good hypoglycemic potential coupled with antidiabetic

efficacy [14] and anti-inflammatory as well as immunomodulatory activity. [15] Also, it has been attributed to possess other pharmacological and therapeutic properties. [16, 17, 18]

Leek (*Allium porrum L.*) belongs to the Alliaceae family, which contains garlic, onions, shallots, and scallions. Although less well-researched than their follow allium vegetables (especially garlic and onions), leeks nevertheless contain many sulfur compounds that are similar to, or identical with, sulfur compounds in these better researched vegetables. The considerable amount of sulfur found in leeks may play an important role in support of our body's antioxidant and detoxified systems. [19, 20] They also contain an impressive amount of polyphenols, carotenoids and chlorophyll present mainly in the green tops. [21]

Leek have been shown to modify certain pathways associated with inhibtion the growth of malignant tumors. [22] The roots have been reputedly used in the traditional medicine for treating inflammatory symptoms. The crushed roots are used to treat initial stages of cough, mucous secretion and sore throat. The fresh juice is taken orally as antispasmodic and also possesses digestive properties. [23]

This study aimed to investigate the ameliorative effect of radish and leek (leaves and roots) juices against the damage induced by DM on liver and kidney functions and oxidative stress in male albino mice.

MATERIALS AND METHODS

1. MATERIALS

1.1. Plant Materials

Radish and Leek samples were obtained from local market (Cairo, Egypt). Leaves separated from roots, washed, cutted, squeezed using juicer and filtered. The fresh filterate was given orally at a dose of 8ml/kg b.wt /day. [24]

1.2. Animals

The experimental animals used throughout this study were one hundred adult male Swiss albino mice weighing 25±5g and were obtained from the breading unit of Egyptian organization for biological product and vaccines (Helwan, Egypt). Mice were maintained on standard commercial pellet diet [25] and tap water *ad libitum*, and kept individually in stainless steel cages in constant environmental conditions.

1.3. Chemicals

- Dimethoate was purchased from Sigma chemical company, Cairo, Egypt, with molecular formula $C_5H_{12}NO_3PS_2$ and purity of 99.5%.
- Kits used for the determination of biochemical parameters were obtained from Biodiagnostic company, Cairo, Egypt.
- All other chemicals were purchased from El-Gomhouria Company, Cairo, Egypt and were of analytical grade.

2. METHODS

2.1. Determination of the bioactive components derived from radish and leek (leaves and roots) juices

High Performance Liquid Chromatography (HPLC) was performed to identify the phenolic compounds [26] and flavonoids [27] content in radish and leek (leaves and roots) juices.

2.2. Experimental design

One hundred adult male albino mice were used in this study. Animals were divided into ten groups (10 mice /group) as follows.

- **Group 1**: (healthy control): Mice in this group were orally administered with 0.5 ml saline solution daily.
- **Group 2**: (healthy RLJ): Mice in this group were orally administered with radish leaves juice (8ml/kg b.wt) daily.
- **Group 3**: (healthy RRJ): Mice in this group were orally administered with radish roots juice (8ml/kg b.wt) daily.
- **Group 4**: (healthy LLJ): Mice in this group were orally administered with leek leaves juice (8ml/kg b.wt) daily.
- **Group 5**: (healthy LRJ): Mice in this group were orally administered with leek roots juice (8ml/kg b.wt) daily.
- **Group 6**: (DM group): Each mouse in this group administered with a daily oral dose of dimethoate (10mg/kg b.wt) dissolved in 0.5 ml saline. [28]
- **Group 7**: (RLJ plus DM): Mice in this group were orally received radish leaves juice (8ml/kg b.wt/day) then administered with dimethoate (10mg/kg b.wt/day) dissolved in 0.5 ml saline.

- **Group 8**: (RRJ plus DM): Mice in this group were orally received radish roots juice (8ml/kg b.wt/day) then administered with dimethoate (10mg/kg b.wt/day) dissolved in 0.5 ml saline.
- **Group 9**: (LLJ plus DM): Mice in this group were orally received leek leaves juice (8ml/kg b.wt/day) then administered with dimethoate (10mg/kg b.wt/day) dissolved in 0.5 ml saline.
- **Group 10**: (LRJ plus DM): Mice in this group were orally received leek roots juice (8ml/kg b.wt/day) then administered with dimethoate (10mg/kg b.wt/day) dissolved in 0.5 ml saline.

2.3. Biological Evaluation

The experimental period was 6 weeks during which food intake and body weight of mice were recorded weekly to monitor the body weight changes and feed efficiency ratio (FER).

2.4. Blood sample collection

At the end of experimental period, all mice were sacrificed after 12 hours fasting with water ad libitum. Blood samples were collected in two clean dry sterile and labeled centrifuge tubes. The first one contained the anticoagulant ethylene diamine tetra acetic acid (EDTA) for collecting blood immediately used for the determination of hematological parameters. In the second tube, blood was allowed to stand for 15 minutes at temperature of 37^{o}C, then centrifuged at 4000 rpm for 20 minutes by EBA8 centrifuge (obtained from china) for the separation of serum. Serum was removed and kept in plastic vials at -20^{o}C until used for biochemical analyses.

2.5. Tissue sampling

Liver and kidney were separated and cleaned, rinsed and washed by saline solution then plotted on filter paper to remove water residue, then weighed to calculate the absolute and relative organs weight that were calculated by dividing the absolute weight of organs on the final body weight of mouse then multiplying by 100 to obtain g % value. Part of the liver and kidney were stored frozen at -20^{O}C until used for biochemical analyses. Another portion of the liver and kidney were kept in 10% formalin for the purpose of histopathological examination.

2.6. Biochemical measurements

2.6.1. Assessment of oxidative stress markers

Oxidative stress markers measured in liver and kidney tissues included nitric oxide (NO) concentration [29], malondialdehyde (MDA) level as one of the main end products of lipid peroxidation by the thiobarbituric acid test [30], reduced glutathione (GSH) content [31] ,superoxide dismutase (SOD) enzyme activity [32], advanced oxidation protein products (AOPP) content [33] and total thiol groups (T-SH) content [34].

2.6.2. Hematological Parameters

Complete blood picture and measurements of blood indices were assessed immediately after mice were sacrificed. [35]

2.6.3 Assessment of liver function

Liver function tests included aspartate aminotransferase (AST), alanine aminotransferase (ALT) activities [36], alkaline phosphatase (ALP) enzyme activity [37], total bilirubin [38], total protein (TP) [39], albumin, globulin levels and A/G ratio in serum [40].

2.6.4 Assessment of kidney function

Kidney function tests included determination of urea [41], creatinine [42] and uric acid [43] concentrations in serum.

2.7. Histopathological Examination

Liver and kidney morphology were assessed by light microscopy. Part of the liver and kidney were sliced and tissues were fixed in 10% buffered-neutral formalin for 6 hours. Fixed liver and kidney tissues were processed and embedded in paraffin. Sections of 4 mm in thickness were subjected to Haemtoxylin and Eosin (H&E) staining before examination.

2.8. Statistical analysis

The data were statistically analyzed by Statistical Package for Social Science (SPSS) version 17.0 statistical packages. Values were presented as mean ± standard deviation (S.D.). Statistical differences between groups were performed using one way ANOVA, the mean difference was significant at the ($p<0.05$) level.

3. RESULTS

General observation

During the experiment, no death was observed in any of the experimental groups. Mice treated with radish and leek juices did not show any sign of toxicity. However, dimethoate treated mice showed varying degrees of clinical signs few minutes after dosing. The signs included huddling, mild tremor, hyperirritability and aggressive behavior. The observed signs were related to the cholinergic crisis; a consistent sign in organophosphate poisoning. Except for the huddling, no other significant clinical manifestation was observed following dimethoate plus juices supplementation. Dimethoate also didn't induce any mortality.

3.1.Bioactive components derived from radish and leek (leaves and roots) juices

Figures (1) and (2) show the percentage of the main phenolic compounds and flavonoids in radish and leek (leaves and roots) juices. We found that total phenolic compounds in RLJ, LLJ, RRJ and LRJ were 12820, 6670, 1430 and 1130 µg GAE /100g juice, respectively. While total flavonoids were 10910, 9560, 350 and 550 µg CE /100g juice, respectively (Table 1).

3.2. Nutritional effect of radish and leek (leaves and roots) juices

Table (2) showed that the administration of dimethoate at a dose of 10 mg/kg b.wt/day caused a marked decrease in food intake, FER and body weight change by 36.83%, 73.81% and 83.33%, respectively as compared to healthy control group. The treatment of dimethoate intoxicated mice in groups 7, 8, 9 and 10 with radish and leek (leaves and roots) juices caused significant elevation in food intake in comparison with G6. However, the decrease in FER and body weight change in the same groups was non-significant in comparison with G1. There was a significant increase in food intake in G2 as compared to G1 ***(P<0.05)***.

In this study, there was a significant increase in the relative weight of liver (5.43±0.69) and kidney (2.17±0.49) for G6 as compared to G1. Mice in groups 7, 8, 9 and 10 showed a significant decrease in liver and kidney relative weights ***(P<0.05)*** as compared to dimethoate treated group.

3.3. Effect of radish and leek (leaves and roots) juices treatment on oxidative stress markers

Tables (3) and (4) display the oxidative stress markers measured in this study. Nitric oxide (NO) level was found to be significantly increased in G6 by 176.44% and 66.74% in liver and kidney tissue respectively as compared to G1. In liver tissue, NO level was expressively

reduced in groups 7, 8, 9 except 10 by 18.56%, 9.50%, 15.74% and 5.16%, respectively as compared to G6. Similarly, NO level was significantly decreased in kidney tissue of groups 7, 8, 9 and 10 by 41.72%, 33.24%, 35.64% and 32.19%, respectively as compared to G6 ***(P<0.05)***.

A significant elevation of free radicals in DM group was observed and evidenced by increased lipid peroxidation in sample tissues. The most remarkable result was the upsurge of MDA level in G6. The elevation in lipid peroxidation- exemplified by MDA content- as a result of DM toxicity reached 157.32% and 75.63% in liver and kidney, respectively as compared to healthy control. Predictably, hepatic MDA in groups receiving radish and leek (leaves and roots) juices has fallen by 47.76%, 37.75%, 44.75%, and 36.91% for groups 7, 8, 9 and 10, respectively when compared with G6. Similarly, in kidney tissue, the MDA concentration was significantly lowered by 23.06%, 17.93%, 16.38%, and 14.28% in the same order ***(P<0.05)***.

In liver tissue, the nonenzymatic antioxidant GSH was dramatically decreased in G6 when compared with G1 by 67.90%. Administration of Radish and leek (leaves and roots) juices resulted in a substantial elevation of GSH content in groups 7, 8, 9 and 10 as compared to G6 by 117.76%, 112.77%, 117.81%, and 109.41%, respectively**.** Likewise, in kidney tissue of G6 the measured GSH content was significantly lowered ***(P<0.05)*** by 53.13% in comparison with G1. However, radish and leek (leaves and roots) juices were able to promptly correct this demotion in G7, G8, G9 and G10 by 64.25%, 59.02%, 61.90%, and 56.99%, respectively as compared to G6 ***(P<0.05)***.

Liver of mice in G6 exhibited a significant decrease in superoxide dismutase (SOD) activity by 45.85% when compared with G1. Mice in groups 7, 8, 9 and 10 showed significant increment in SOD activity by 28.85%, 21.46%, 22.34%, and 14.34%, respectively as compared to G6. Likewise, in kidney of G6 the measured SOD activity was significantly lowered ***(P<0.05)*** by 43.64% in comparison with G1. However, radish and leek (leaves and roots) juices in groups 7, 8, 9 and 10 were able to improve this lowered activity by 42.00%, 32.82%, 38.62%, and 30.30%, respectively compared to G6.

As for advanced oxidation protein products (AOPP) content, there was a significant rise in DM group that reached 121.95% in liver and 47.37% in kidney when compared with G1. Conversely, administration of juices in groups 7, 8, 9 and 10 significantly decreased ***(P<0.05)***

the content of AOPP in liver by 26.37%, 17.58%, 20.33%, and 13.74%, respectively and in kidney by 21.43%, 17.86%, 19.05%, and 17.86%, respectively when compared with G6.

There was a significant fall in the content of T.SH in G6 that reached 36.77% in liver and 50.06% in kidney when compared with G1. On the other hand, groups 7, 8, 9 and 10 exhibited a significant increase ***(P<0.05)*** in the content of T.SH in liver by 47.48%, 45.98%, 46.99%, and 36.37%, respectively. Similarly, in kidney tissue, our results showed a significant increased T.SH content for groups 7, 8, 9 and 10 reached 58.29%, 45.47%, 53.15%, and 40.42%, respectively when compared with G6.

3.4. Effect of radish and leek (leaves and roots) juices treatment on hematological parameters

Table (5) demonstrates the effect of radish and leek (leaves and roots) juices administration on the hematological parameters in healthy and dimethoate treated groups. In the group administered dimethoate, there were significant decreases ***(P<0.05)*** in Hb concentration, HCT, RBC_S count, MCV, MCH, and MCHC. The percent of decreases were 21.10%, 23.28%, 35.86%, 2.18%, 9.51%, and 10.44%, respectively, while there was a significant increase in total WBC_S count by 101.25% when compared with healthy control group.

However, in groups 7, 8, 9 and 10, radish and leek (leaves and roots) juices were able to raise significantly RBC_S count, Hb concentration, HCT, MCV, MCH, MCHC and restored the elevated total WBC_S in comparison with G6 ***(P<0.05)***.

3.5. Effect of radish and leek (leaves and roots) juices treatment on liver function

Table (6) showed that treatment with DM significantly increased the activities of liver enzymes AST, ALT and ALP by 43.75%, 49.25%, and 68.97%, respectively. In addition to increased total bilirubin concentration by 438.46% as compared to control animals. Also dimethoate caused a significant reduction in the level of total protein, albumin and A/G ratio by 28.26%, 40.17%, and 30.38% but it caused a non-significant decrease ***(P<0.05)*** in globulin compared to G1.

Whereas groups administered radish and leek (leaves and roots) juices caused very significant improvement in serum AST activity by 10.73%, 8.20%, 8.95%, and 8.16% in groups 7, 8, 9 and 10, respectively as compared to G6. Also in these groups a significant reduction in serum ALT activity by 5.17%, 5.22%, 4.05%, and 3.51%, respectively were shown. In case of ALP activity, there was a significant decrease by 36.01%, 32.80%, 33.87%, and 30.97%, respectively as compared to G6. Total bilirubin was significantly reduced ***(P<0.05)*** by the

administration of radish and leek (leaves and roots) juices by 56.43%, 45.71%, 47.86%, and 37.14% in groups 7, 8, 9 and 10, respectively as compared to G6.

Total protein was significantly increased in groups 7, 8, 9 and 10 by 21.78%, 10.89%, 18.33% and 4.90%, respectively as compared to G6. There was a significant increase ***(P<0.05)*** in albumin level in groups 7, 8, 9, and 10 by 45.71%, 25.36%, 28.93%, and 11.43% respectively as compared to G6. There was no significant difference in globulin level in groups 7, 8, 9 and 10 as compared to G6.

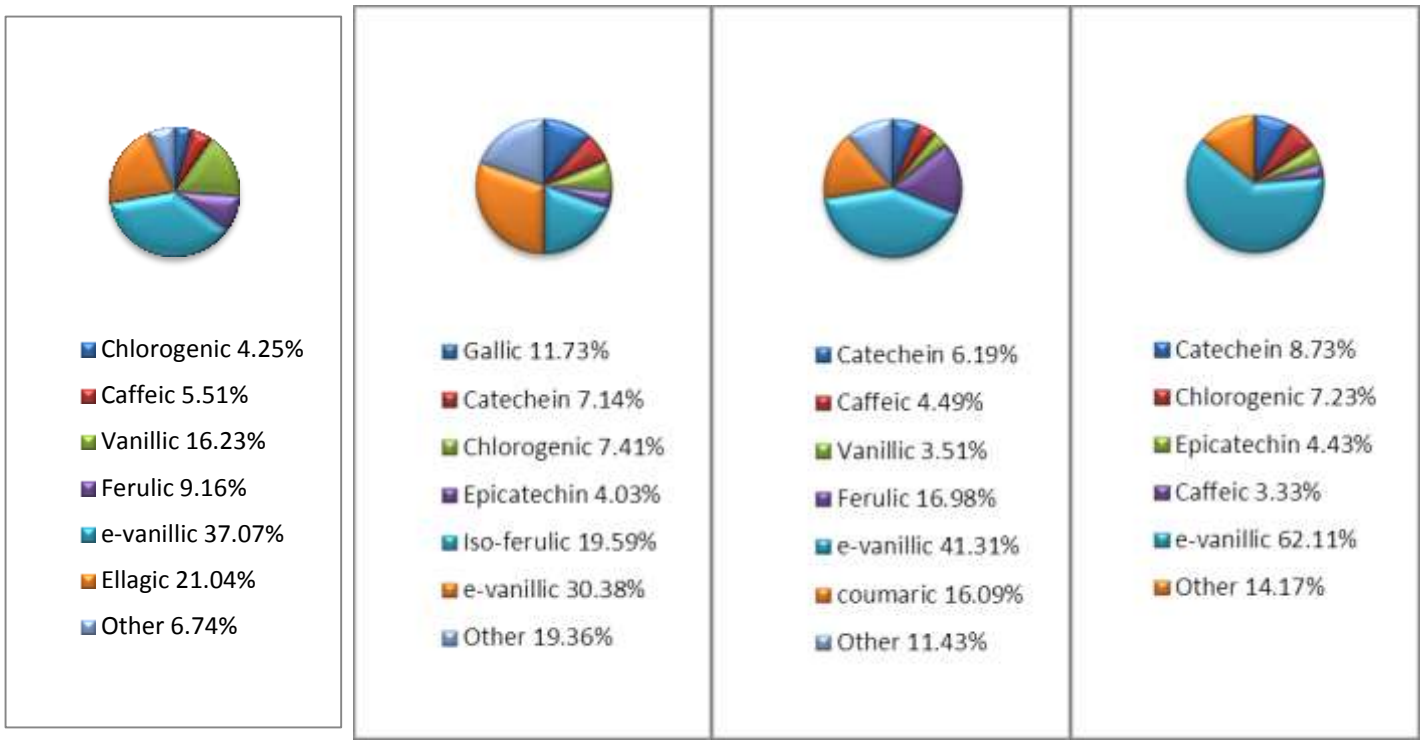
3.6. Effect of radish and leek (leaves and roots) juices treatment on kidney function

Dimethoate-intoxicated rats showed a constellation of disorders in renal function witnessed by increased urea, creatinine and uric acid levels (Table 7) as compared to controls. The percent of change in urea, creatinine and uric acid were 76.55%, 153.33% and 58.08%, respectively as compared to G1***(P<0.05).***

Administration of radish and leek (leaves and roots) juices in groups 7, 8, 9 and 10 caused a significant decrease in urea level by 34.83%, 24.04%, 27.13%, and 23.32%, a significant reduction in creatinine level by 35.96%, 24.56%, 29.82%, and 19.30% as well as a marked decrease in uric acid concentration by 22.88%, 21.14%, 21.84%, and 14.21%, respectively as compared to dimethoate treated group ***(P<0.05).***

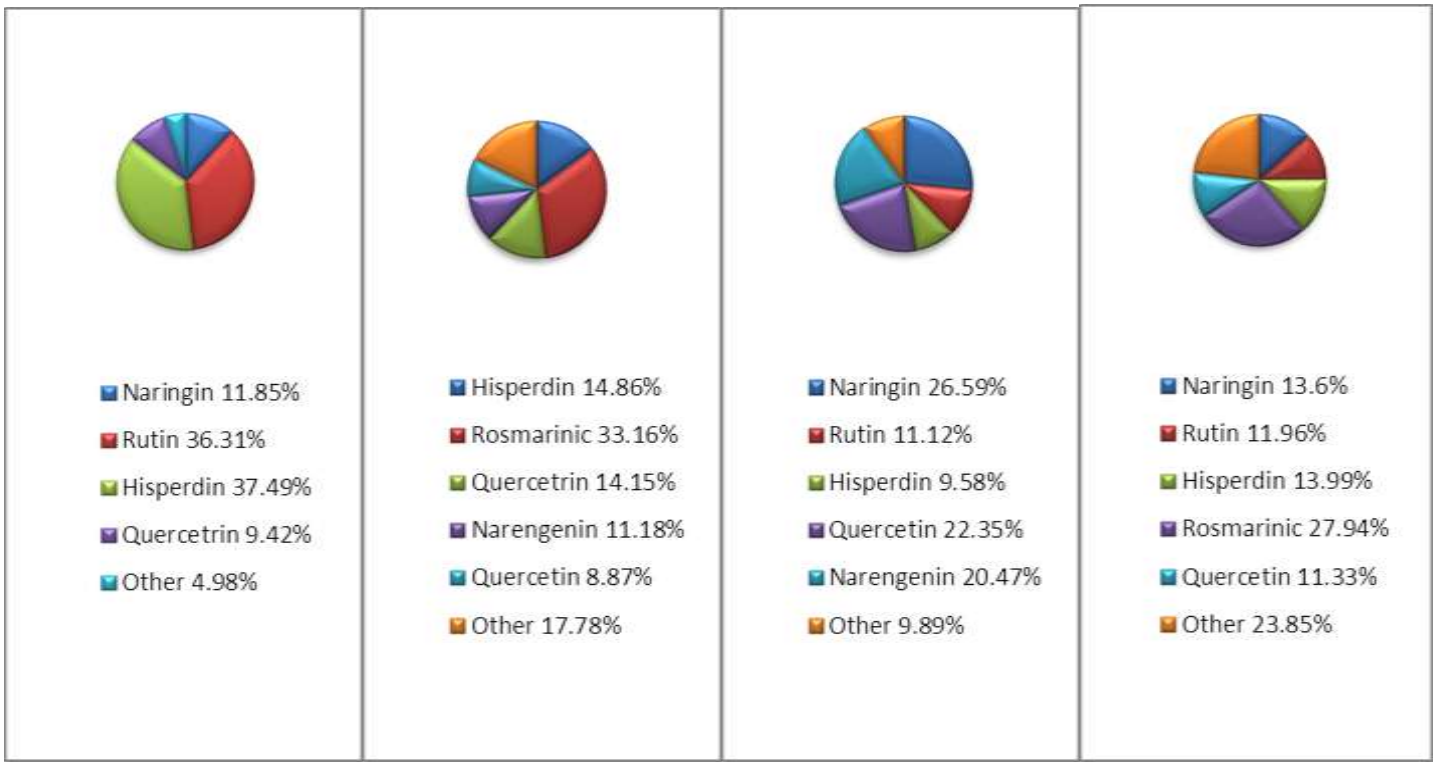
3.7. Histopathological Examination of liver and kidney

Histopathological examination of livers of healthy control and healthy radish and leek (leaves and roots) juices groups' revealed normal histological picture of hepatic lobule as shown in Fig. 3 (A-E). Examination of liver of the DM intoxicated mice showed dilated central vein (D), interstitial hemorrhage (R) and lymphocytic infiltration (L) (Fig. 3F). Livers of RLJ + DM, RRJ + DM, LLJ + DM, LRJ + DM showed almost normal histology of the hepatic lobule Fig. 3 (G-J). In histopathological examination of kidneys of healthy control and healthy radish and leek (leaves and roots) juices groups' showed normal glomerular and tubular histology as shown in Fig. 4 (A-E) whereas DM was found to cause glomeruli atrophy (G), congestion (C) and tubular degeneration (D) (Fig. 4F). Treatment with radish and leek (leaves and roots) juices were found to have a protective effect on DM induced kidney damage as shown in Fig. 4 (G-J).



(1-A) RLJ **(1-B) RRJ** **(1-C) LLJ** **(1-D) LRJ**

Fig (1): Percentage of main phenolic compounds derived from radish and leek (leaves and roots) juices



(2-A) RLJ **(2-B) RRJ** **(2-C) LLJ** **(2-D) LRJ**

Fig (2): Percentage of main flavonoids derived from radish and leek (leaves and roots) juices

Table (1): Total phenolics and total flavonoids in radish and leek (leaves and roots) juices

Juice	Total phenols (µg GAE/100g juice)	Total flavonoids (µg CE/100g juice)
RLJ	12820	10910
RRJ	1430	350
LLJ	6670	9560
LRJ	1130	550

GAE = Gallic Acid Equivalent; CE = Catechein Equivalent

Table (2): Nutritional effect of radish and leek (leaves and roots) juices in healthy and dimethoate treated groups

Parameters / Groups	Food intake (g)	Body weight Change (g)	Feed efficiency ratio	Relative weight of liver (g %)	Relative weight of kidney (g %)
G1: Normal control	143.10±28.41 ^{bc}	6.00± 2.54 ^{a}	0.042±0.029 ^{a}	4.05±0.44 ^{d}	1.32±0.43 ^{c}
G2: Healthy RLJ	176.00±38.94 ^{a}	5.53±2.79 ^{a}	0.026±0.019 ^{ab}	4.08±0.82 ^{d}	1.36±0.43 ^{bc}
G3: Healthy RRJ	163.80±28.41 ^{ab}	5.10±3.90 ^{a}	0.021±0.017 ^{ab}	4.09±0.42 ^{cd}	1.36±0.39 ^{bc}
G4: Healthy LLJ	145.00±18.28 ^{bc}	5.00±3.02 ^{a}	0.024±0.010 ^{ab}	4.14±0.38 ^{cd}	1.37±0.22 ^{bc}
G5: Healthy LRJ	136.60±24.72 ^{cd}	5.00±3.80 ^{a}	0.024±0.018 ^{ab}	4.17±0.61 ^{cd}	1.37±0.22 ^{bc}
G6: DM	90.40±5.17 ^{f}	1.00±4.81 ^{b}	0.011±0.056 ^{b}	5.43±0.69 ^{a}	2.17±0.49 ^{a}
G7: RLJ + DM	128.40±20.80 ^{cde}	3.70±2.75 ^{ab}	0.020±0.020 ^{ab}	4.60±0.66 ^{bc}	1.64±0.58 ^{bc}
G8: RRJ + DM	117.90±10.73 ^{de}	3.40±2.07 ^{ab}	0.019±0.008 ^{ab}	4.74±0.54 ^{b}	1.71±0.50 ^{b}
G9: LLJ + DM	120.60±7.85 ^{de}	3.50±4.62 ^{ab}	0.017±0.022 ^{ab}	4.69±0.56 ^{b}	1.67±0.33 ^{bc}
G10: LRJ + DM	113.80±30.31^{e}	3.30±4.37 ^{ab}	0.016±0.023 ^{ab}	4.80±0.54 ^{b}	1.73±0.43 ^{b}

Values are expressed as means ± S.D, n=10, (P<0.05)
There is no significant difference between means have the same letter in the same column.

Table (3): Effect of radish and leek (leaves and roots) juices treatment on oxidative stress markers in liver of healthy and dimethoate treated groups

Parameters / Groups	GSH (mg/g tissue)	MDA (nmol/g tissue)	NO (µmol/g tissue)	SOD (U/g tissue)	AOPP (µmol/g tissue)	T.SH (nmol/g tissue)
G1: Normal control	64.90±4.87 ^{a}	52.25±5.73 ^{d}	31.90±6.97 ^{de}	136.72±2.29 ^{a}	0.82±0.113 ^{ef}	134.63±3.50 ^{a}
G2: Healthy RLJ	65.50±2.37 ^{a}	49.37±5.96 ^{d}	28.95±4.20 ^{e}	136.23±3.39 ^{a}	0.80±0.059 ^{f}	136.64±3.06 ^{a}
G3: Healthy RRJ	64.54±3.50 ^{a}	50.62±7.57 ^{d}	34.68±4.39 ^{d}	134.62±2.44 ^{a}	0.83±0.050 ^{ef}	135.64±2.92 ^{a}
G4: Healthy LLJ	65.47±1.99 ^{a}	50.03±8.84 ^{d}	30.30±6.48 ^{de}	135.17±2.63 ^{a}	0.81±0.092 ^{ef}	137.11±2.89 ^{a}
G5: Healthy LRJ	63.44±3.71 ^{a}	51.86±4.95 ^{d}	35.00±2.91 ^{d}	134.43±3.39 ^{a}	0.86±0.014 ^{e}	134.00±2.75 ^{a}
G6: DM	20.83±4.24 ^{c}	134.45±2.28 ^{a}	86.42±7.09 ^{a}	74.04±2.42 ^{e}	1.82±0.088 ^{a}	85.13±2.62 ^{d}
G7: RLJ + DM	45.36±2.77 ^{b}	70.24±8.88 ^{c}	70.38±7.98 ^{c}	95.40±3.06 ^{b}	1.34±0.049 ^{d}	125.55±2.41 ^{b}
G8: RRJ + DM	44.32±2.14 ^{b}	83.69±7.70 ^{b}	78.21±5.93 ^{b}	89.93±3.27 ^{c}	1.50±0.020 ^{c}	124.27±2.73 ^{b}
G9: LLJ + DM	45.37±3.26 ^{b}	74.29±3.35 ^{c}	72.82±6.13 ^{c}	90.58±4.00 ^{c}	1.45±0.054 ^{c}	125.13±2.38 ^{b}
G10: LRJ + DM	43.62±2.83 ^{b}	84.82±2.90 ^{b}	81.96±2.86 ^{ab}	84.66±3.07 ^{d}	1.57±0.038 ^{b}	116.09±2.80 ^{c}

Values are expressed as means ± S.D, n=10, (P<0.05)
There is no significant difference between means have the same letter in the same column

Table (4): Effect of radish and leek (leaves and roots) juices treatment on oxidative stress markers in kidney of healthy and dimethoate treated groups

Parameters / Groups	GSH (mg/g tissue)	MDA (nmol/g tissue)	NO (µmol/g tissue)	SOD (U/g tissue)	AOPP (µmol/g tissue)	T.SH (nmol/g tissue)
G1: Normal control	47.32±1.20 ^{ab}	50.55±2.74 ^{e}	75.11±5.29 ^{f}	224.97±3.09 ^{ab}	0.57±0.035 ^{c}	62.32±3.45 ^{a}
G2: Healthy RLJ	49.46±4.72 ^{a}	47.83±4.57 ^{e}	73.75±4.22 ^{f}	229.07±22.80 ^{a}	0.55±0.027 ^{c}	63.70±4.87 ^{a}
G3: Healthy RRJ	44.45±3.10 ^{b}	49.46±2.81 ^{e}	75.28±3.18 ^{f}	223.93±2.45 ^{ab}	0.55±0.022 ^{c}	62.24±1.97 ^{a}
G4: Healthy LLJ	45.90±3.15 ^{b}	48.03±3.02 ^{e}	74.15±3.40 ^{f}	227.92±5.59 ^{ab}	0.55±0.024 ^{c}	63.39±8.65 ^{a}
G5: Healthy LRJ	44.29±2.45 ^{b}	50.39±3.07 ^{e}	75.67±2.70 ^{ef}	221.20±5.89 ^{b}	0.57±0.018 ^{c}	61.92±4.48 ^{a}
G6: DM	22.18±3.76 ^{d}	88.78±5.99 ^{a}	125.24±2.66 ^{a}	126.80±3.85 ^{f}	0.84±0.109 ^{a}	31.12±0.89 ^{e}
G7: RLJ + DM	36.43±4.96 ^{c}	68.31±1.61 ^{d}	72.99±5.84 ^{de}	180.05±6.90 ^{c}	0.66±0.035 ^{b}	49.26±0.73 ^{b}
G8: RRJ + DM	35.27±2.87 ^{c}	74.24±2.48 ^{bc}	83.81±3.35 ^{bc}	168.41±5.38 ^{de}	0.69±0.035 ^{b}	45.27±3.79 ^{cd}
G9: LLJ + DM	35.91±3.58 ^{c}	72.86±5.18 ^{c}	80.61±5.96 ^{cd}	175.77±5.02 ^{cd}	0.68±0.041 ^{b}	47.66±1.94 ^{bc}
G10: LRJ + DM	34.82±3.06 ^{c}	76.10±1.21 ^{b}	84.92±3.01 ^{b}	165.22±5.08 ^{e}	0.69±0.023 ^{b}	43.70±2.73 ^{d}

Values are expressed as means ± S.D, n=10, (P<0.05)
There is no significant difference between means have the same letter in the same column.

Table (5): Effect of radish and leek (leaves and roots) juices treatment on hematological parameters in healthy and dimethoate treated groups

Parameters / Groups	Hemoglobin concentration (g/dl)	HCT (%)	RBC_S count ($x10^6$/µl)	WBC_S count ($x10^3$/ µl)	MCV (fl)	MCH (pg)	MCHC (g/dl)
G1: Normal control	13.60±0.09 ^{a}	42.70±0.09 ^{a}	4.29±0.38 ^{a}	8.45±0.64 ^{f}	93.74±0.51^{a}	30.73±1.37 ^{a}	32.70±1.62 ^{a}
G2: Healthy RLJ	13.62±0.34 ^{a}	42.70±0.53 ^{a}	4.32±0.48 ^{a}	8.43±0.05 ^{f}	93.62±0.51 ^{ab}	30.73±0.65 ^{a}	32.65±0.47 ^{a}
G3: Healthy RRJ	13.61±0.07^{a}	42.72±0.51 ^{a}	4.29±0.47 ^{a}	8.46±0.14 ^{f}	93.59±0.56 ^{ab}	30.71±0.47 ^{a}	32.54±0.31 ^{a}
G4: Healthy LLJ	13.61±0.54 ^{a}	42.70±0.67 ^{a}	4.32±0.10 ^{a}	8.43±0.57 ^{f}	93.60±0.46 ^{ab}	30.71±0.99 ^{a}	32.62±0.26 ^{a}
G5: Healthy LRJ	13.59±0.45 ^{a}	42.73±0.93 ^{a}	4.29±0.15 ^{a}	8.49±0.17 ^{f}	93.35±0.20 ^{ab}	30.68±0.60 ^{a}	32.50±0.22 ^{a}
G6: DM	10.73±0.07 ^{e}	32.76±0.13 ^{e}	2.88±0.06 ^{e}	17.67±0.15 ^{a}	92.07±0.62 ^{e}	28.35±0.71 ^{c}	28.39±1.49 ^{c}
G7: RLJ + DM	13.42±0.09 ^{a}	37.34±0.12 ^{b}	3.90±0.05 ^{b}	12.58±0.10 ^{e}	93.27±0.51 ^{bc}	29.85±0.36 ^{b}	29.91±0.87 ^{b}
G8: RRJ + DM	12.80±0.06 ^{c}	36.65±0.13 ^{c}	3.50±0.04 ^{c}	15.59±0.17 ^{c}	92.59±0.59 ^{d}	29.50±0.37 ^{b}	29.68±0.94 ^{b}
G9: LLJ + DM	13.11±0.07 ^{b}	36.57±0.18 ^{c}	3.69±0.09 ^{b}	14.23±0.09 ^{d}	92.89±0.50 ^{cd}	29.81±0.59 ^{b}	29.71±0.96 ^{b}
G10: LRJ + DM	12.32±0.07^{d}	35.30±0.21 ^{d}	3.26±0.05 ^{d}	16.54±0.30 ^{b}	92.58±0.47^{d}	29.43±0.33 ^{b}	29.59±0.97 ^{b}

Values are expressed as means ± S.D, n=10, (P<0.05)
There is no significant difference between means have the same letter in the same column.

Table (6): Effect of radish and leek (leaves and roots) juices treatment on liver function of healthy and dimethoate treated groups

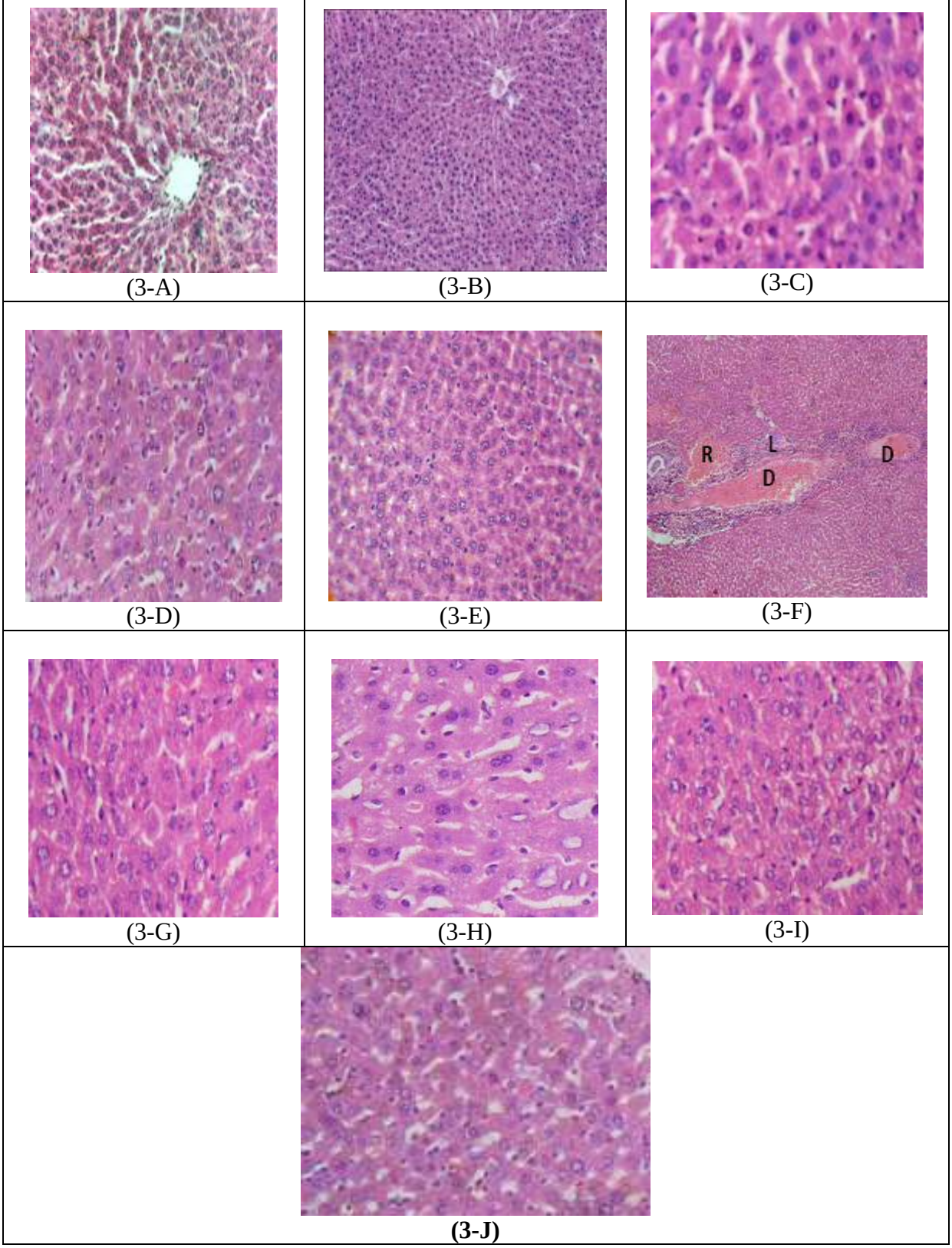
Parameters / Groups	AST (U/ml)	ALT (U/ml)	ALP (IU/L)	Total bilirubin (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)	A/G ratio
G1: Normal control	53.03±4.17^{c}	46.36±3.13^{c}	75.18±5.74^{e}	0.26±0.064^{e}	7.68±0.54^{a}	4.68±0.44^{a}	2.97±0.24^{a}	1.58±0.17^{abcd}
G2: Healthy RLJ	52.56±5.91^{c}	46.44±3.10^{c}	74.63±2.66^{e}	0.27±0.049^{e}	7.64±0.28^{a}	4.77±0.12^{a}	2.87±0.34^{a}	1.65±0.41^{abc}
G3: Healthy RRJ	52.77±5.16^{c}	46.67±0.22^{c}	74.70±2.70^{e}	0.29±0.066^{e}	7.46±0.32^{a}	4.63±0.48^{a}	2.83±0.61^{a}	1.63±0.24^{ab}
G4: Healthy LLJ	52.67±5.38^{c}	46.48±3.40^{c}	74.68±3.05^{e}	0.29±0.032^{e}	7.62±0.33^{a}	4.69±0.63^{a}	2.93±0.42^{a}	1.60±0.79^{a}
G5: Healthy LRJ	53.05±2.32^{c}	46.67±0.03^{c}	75.30±2.86^{e}	0.35±0.071^{e}	7.45±0.32^{a}	4.61±0.80^{a}	2.84±0.16^{a}	1.62±0.97^{a}
G6: DM	76.23±3.12^{a}	69.19±0.58^{a}	127.03±1.89^{a}	1.40±0.298^{a}	5.51±0.33^{d}	2.80±0.32^{d}	2.71±0.61^{a}	1.03±0.34^{e}
G7: RLJ + DM	68.05±0.39^{b}	65.61±1.31^{b}	81.29±2.95^{d}	0.61±0.092^{d}	6.71±0.42^{b}	4.08±0.30^{b}	2.63±0.40^{a}	1.55±0.32^{abcde}
G8: RRJ + DM	69.98±0.47^{b}	66.39±0.28^{b}	85.37±2.95^{bc}	0.76±0.150^{b}	6.11±0.22^{c}	3.51±0.14^{c}	2.60±0.30^{a}	1.35±0.17^{bcde}
G9: LLJ + DM	69.41±0.17^{b}	65.58±0.18^{b}	84.00±2.70^{cd}	0.73±0.108^{c}	6.52±0.34^{b}	3.61±0.10^{c}	2.92±0.30^{a}	1.25±0.12^{cde}
G10: LRJ + DM	70.01±0.47^{b}	66.76±0.12^{b}	87.69±2.80^{b}	0.88±0.155^{b}	5.78±0.21^{d}	3.12±0.35^{d}	2.66±0.46^{a}	1.17±0.33^{de}

Values are expressed as means ± S.D, n=10, (P<0.05)
There is no significant difference between means have the same letter in the same column.

Table (7): Effect of radish and leek (leaves and roots) juices treatment on kidney function of healthy and dimethoate treated groups

Parameters / Groups	Urea (mg/dl)	Creatinine (mg/dl)	Uric acid (mg/dl)
G1: Normal control	25.89±1.30 ^{d}	0.45±0.027 ^{d}	3.65±0.21 ^{d}
G2: Healthy RLJ	24.74±0.84 ^{d}	0.36±0.038 ^{d}	3.65±0.16 ^{d}
G3: Healthy RRJ	25.94±2.01 ^{d}	0.41±0.017 ^{d}	3.67±0.37 ^{d}
G4: Healthy LLJ	25.78±1.37 ^{d}	0.36±0.012 ^{d}	3.66±0.44 ^{d}
G5: Healthy LRJ	26.48±1.17 ^{d}	0.43±0.037 ^{d}	3.67±0.47 ^{d}
G6: DM	45.71±2.62 ^{a}	1.14±0.381 ^{a}	5.77±0.48 ^{a}
G7: RLJ + DM	29.79±3.92 ^{c}	0.73±0.276 ^{c}	4.45±0.10 ^{c}
G8: RRJ + DM	34.72±2.76 ^{b}	0.86±0.235 ^{bc}	4.55±0.72 ^{c}
G9: LLJ + DM	33.31±2.33 ^{b}	0.80±0.075 ^{bc}	4.51±0.28 ^{c}
G10: LRJ + DM	**35.05±3.95** ^{b}	**0.92±0.185** ^{b}	**4.95±0.51** ^{b}

Values are expressed as means ± S.D, n=10, (P<0.05)
There is no significant difference between means have the same letter in the same column.



(3-A) (3-B) (3-C)

(3-D) (3-E) (3-F)

(3-G) (3-H) (3-I)

(3-J)

Fig. 3 (A-J): Liver sections of mice in experimental groups, stained with (H&E X200)

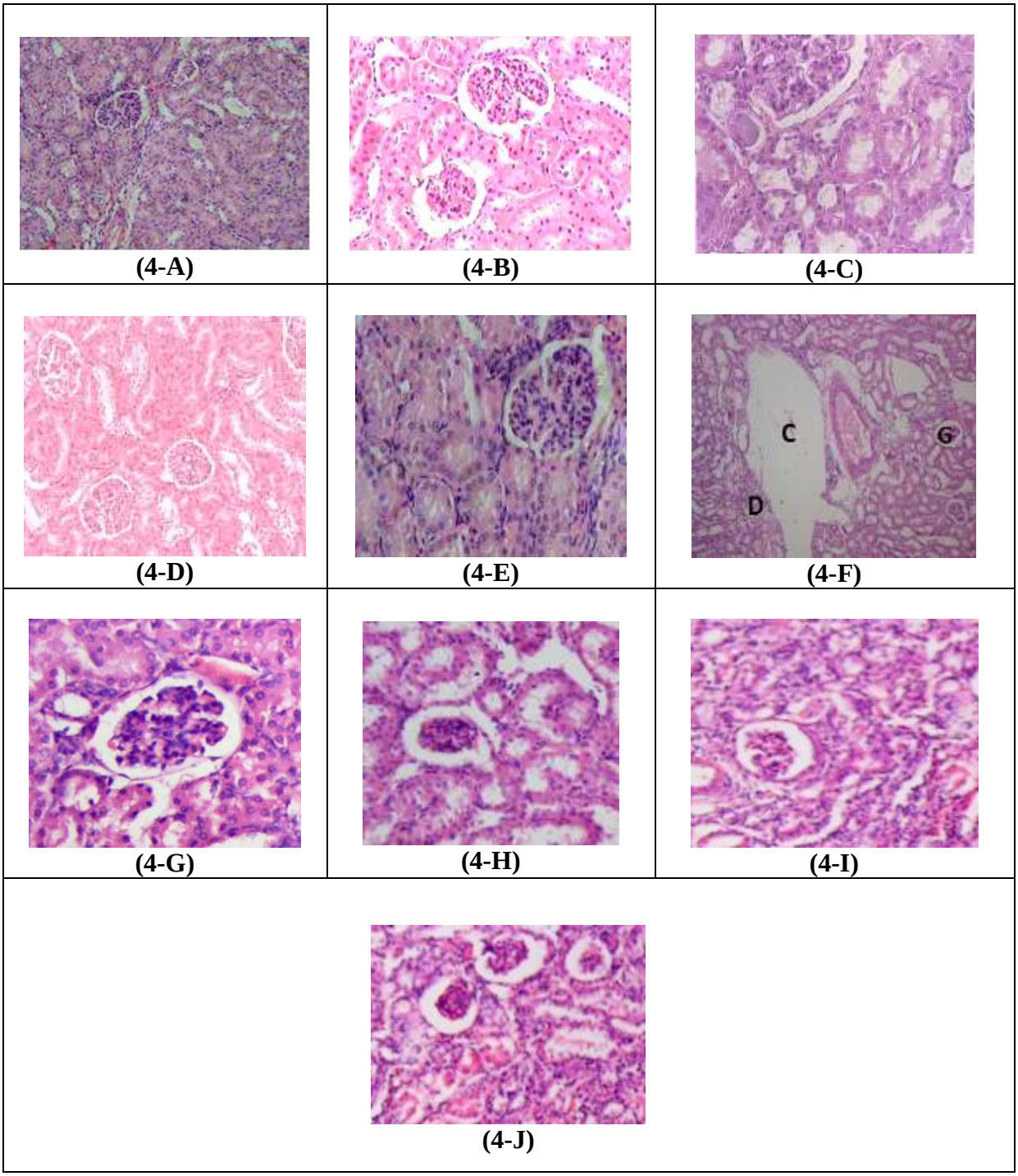


Fig. 4 (A-J): Kidney sections of mice in experimental groups, stained with (H&E X200) (A): Normal control, (B): Healthy RLJ, (C): Healthy RRJ: (D): Healthy LLJ, (E): Healthy LRJ, (F): DM, (G): RLJ + DM, (H): RRJ + DM, (I): LLJ + DM, (J): LRJ + DM

4. DISCUSSION

4.1. Bioactive components derived from radish and leek (leaves and roots) juices

Radish and leek (leaves and roots) juices contain many bioactive components with varying amounts. e-vanillic, ellagic, vanillic, ferulic, caffeic, chlorogenic, iso-ferulic are the main phenolic compounds found in RLJ, while hisperdin, rutin, quercetrin, hespertin, apegenin and 7-OH flavone are the main flavonoids found in this juice. p-coumaric acid, catechein, gallic acid, coumarin, resversetrol, and epicatechein are the main phenolic compounds found in LLJ, while naringin, quercetin, narengenin, rosmarinic, and kampherol are the main flavonoids found in the same juice. [13, 44] RLJ was found to contain the higher concentration of bioactive components followed by LLJ then RRJ and finally LRJ. The presence of these bioactive components may be associated with decreased oxidative stress and injury in mice groups administered DM.

4.2. Nutritional effect of radish and leek (leaves and roots) juices

In the present study, oral administration of dimethoate caused a significant reduction in food intake, body weight change, and an increase in relative weight of liver and kidney. The reduction in food intake in the dimethoate treated group may be due to the toxic effects of pesticide that resulted in the loss of body weight or as a result of oxidative stress. These observations are consistent with the previous findings.[45, 46]

In toxicological studies, organ and relative organ weights are important criteria for evaluation of organ toxicity. Similar increase of the liver and kidney weight after pesticide administration was also reported. [47, 48] The intake of radish and leek (leaves and roots) juices caused an improvement on the relative weight of liver and kidney. This may be due to the presence of flavonoids and the phenolic compounds in these juices which have high antioxidant ability, thus reducing oxidative stress accompanied with dimethoate intoxication.

4.3. Antioxidant Effect of radish and leek (leaves and roots) juices

A common consequence of most stresses is that they result in an increased production of reactive oxygen species. The successive reduction of molecular oxygen to H_2O yields the intermediates which are potentially toxic because they are relatively reactive than O_2. Reactive oxygen species (ROS) may lead to unspecific oxidation of proteins and membrane lipids or may cause DNA injury. [49]

The mechanism by which dimethoate and OPI in general, could promote oxidative stress is that dimethoate acts as an inducer of P450 isoenzyme. This induction of P450 enzyme system may be responsible for dimethoate increased biotransformation to P=O analogue. The dimethoate induced enhancement in liver microsomal Cytochrome P450 content and oxygen radicals production together with lipid peroxidation which detected by the significant increase in thiobarbituric acid reactive substances (TBARS) in the liver and kidney. The increased levels of TBARS indicate an enhanced lipid peroxidation leading to tissue injury and failure of the antioxidant defense mechanisms to prevent the formation of excess free radicals. [50]

Nitric oxide (NO), the end-metabolic product of peroxinitrite anion (nitrites and nitrates), is determined as a biomarker for reactive nitrogen species (RNS). The increments observed in NO level following dimethoate induced toxicity reflected the systemic impact in the production of NO caused by the oxidative insult of dimethoate. NO also reacted with O_2•− to form peroxynitrite anion (ONOO−) which is a high reactive radical responsible for oxidative damage to macromolecules. [51] Nitric oxide (NO) has been implicated in the mechanisms of cell injury and long-term physiological changes in cellular excitability suggested that NO has an important role in modulating oxidant stress and tissue damage. [52] This confirmed by several studies. [51, 53]

Dimethoate induced enhancement in liver microsomal cytochrome P450 content and oxygen radical production is paralleled by an augmented lipid peroxidation as evidenced by the significant increase in thiobarbituric acid reactive substances (TBARS) detected in liver homogenates.

Highly reactive oxygen metabolites, especially hydroxyl radicals, act on polyunsaturated fatty acids of phospholipid components of membranes to produce malondialdehyde, a lipid peroxidation product. Lipid peroxidation caused by ROS has been suggested as one of the molecular mechanisms involved in organophosphorous pesticide. This was in agreement with our results since dimethoate has been reported to induce oxidative stress, as shown by increased MDA production leading to tissue injury and failure of the antioxidant defense mechanism that prevent the formation of excess free radical.

Results have shown that radish and leek juices has a high potent protective effect against oxidative stress; as demonstrated by the significant decrease of lipid peroxidation, as well as

the amelioration of enzymes' antioxidant status. Polyphenols are also demonstrated by its ability to inhibit the production of nitric oxide.

The ability of radish and leek juices to ameliorate the increased levels of MDA and NO could be due to their content of polyphenols which have mechanisms by which they reduce oxidation besides direct role as antioxidants: (1) Binding of metal ions such as iron and copper and preventing their participation in oxidation reactions (leading to the formation of hydroxyl radical). (2) Suppression of oxidation stimulants such as induced nitric oxide synthase (iNOS), cyclooxygenase 2 (COX- 2), lipoxygenase 2 (LOX-2) and xanthine oxidase. (3) Induction of antioxidant enzymes such as glutathione S-transferase and superoxide dismutase. Significant decreases in the levels of MDA and NO and activation of antioxidant enzymes activities as a result of radish and leek juices administration were previously reported. [54, 55, 56]

Biological systems have evolved with endogenous defense mechanisms to help the protection against free radicals induced cell damage. SOD is an important endogenous antioxidant enzyme that acts as the first line defense system against ROS, metabolizes toxic oxidative intermediates and catalyzes superoxide to H_2O_2 and O_2. [57, 58]

In this study, SOD activity and GSH level significantly decreased in dimethoate toxicated mice due to their consumption and exhausting antioxidant agents present in the body. GSH is a tripeptide and a powerful antioxidant present within the cytosol of cells and is the major intracellular non protein thiol compound (NPSH). It is important in maintaining –SH groups in other molecules including proteins, regulating thiol-disulfide status of the cell, and detoxifying foreign compounds and free radicals. [58]

The depletion of the enzymatic antioxidant SOD and non-enzymatic antioxidant GSH in the dimethoate administration could be either the result of their increased utilization for conjugation and/or their participation in achieving free radical products induced by dimethoate toxicity. This supported by several researches. [59, 60]

The present study revealed that SOD activity and GSH level significantly increased by radish and leek juices. The antioxidant activity of these plant juices may be due to an inhibition of the reactive oxygen species (ROS) inducing a chain reaction mediated by several antioxidant enzymes including SOD and non-enzymatic antioxidant as reduced glutathione (GSH) for

dealing with these toxic substances suggesting that natural antioxidants constitute efficient treatment of toxicity induced by xenobiotics.

A study showed that combined treatment with radish extract and zearalenone succeeded in restoring the antioxidant enzyme activities since it caused a significant increase in GSH and SOD activity in the liver and kidney which may be due to the higher content of isothiocyanate, kaempherol glycosides and L-tryptophan compounds in radish extract and their ability to scavenge free radicals. [61] Another study revealed that Raphanus sativus methanolic extract reversed the decreased levels of reduced GSH and SOD which may be due to the presence of polyphenolic compounds. [62]

Also, the presence of S-allylcysteine and S-allylmercaptocysteine which are extracted from Allium porrum exert an antioxidant action by scavenging the ROS, enhancing the cellular antioxidant enzyme; superoxide dismutase and increasing glutathione in the cells. [63]

Both GSH and SOD are considered to be free-radical scavengers in the cells. Thus, the decrease in GSH level and SOD activity leading to an indirect increase in oxidative DNA damage, which suggests that SOD plays a role in the suppression of oxygen free-radical formation and the decrease of NO generation. [64]

Since free radicals have very short half-lives, the clinical assessment of oxidative stress is based on the measurement of different stable oxidized products of modified proteins, lipids, carbohydrates and nucleic acids. Proteins are susceptible to oxidant-mediated injury, forming cross-linkage and aggregation products that are resistant to proteolysis. [65]. Accumulation of modified proteins disrupts cellular function either by loss of catalytic and structural integrity or by interruption of regulatory pathways. Markers of protein oxidation named as advanced oxidation protein products (AOPPs) which are defined as dityrosine containing cross linked protein products, and are considered as reliable markers to estimate the degree of oxidant-mediated protein damage.[66]

In this study, AOPP content significantly increased in dimethoate toxicated mice. This may be due to the ability of dimethoate to oxidize and modify proteins. In previous studies, dimethoate resulted in a significant increase in AOPP levels in the heart tissue and cerebral cortex tissue of adult rats by (+50%) and (74%) respectively suggesting that dimethoate activated the formation of free radicals in these tissues. [67, 68]

The protective effect of radish and leek (leaves and roots) juices may be explained depending on the fact that these juices contain polyphenolic compounds which may scavenge free radicals offering protection and their antioxidant potential mechanism suggesting that the juices of these plants may be useful to prevent the oxidative stress inducing damage and protein oxidation.

Plasma thiol groups are physiological free radical scavengers. The -SH moiety of cysteine is highly prone to oxidative attack by several mechanisms, leading to the formation of disulfide bonds and thiyl radicals. Oxidation of cysteine-SH groups can also give rise to intra- or inter-protein cross-linked derivatives.

It has been estimated that proteins can scavenge the majority (50% –75%) of reactive species generated and much of this function is attributed to the thiol groups present in them. The serum levels of protein -SH in the body indicate antioxidant status and low levels of protein –SH correlated positively with the increased levels of lipid peroxides and of advanced oxidation protein products (AOPPs). [69]

In our study, dimethoate toxicity has been postulated to have multiple effects including generation of ROS and induction of intracellular oxidative stress, thereby disrupting normal cellular development and caused depletion in the thiol content due to the usage in detoxification.

In a previous study, there was a significant decrease in the level of total thiols in acute organophosphorus poisoned patients and that organophosphate induces renal tubular epithelial cell toxicity through elevation of reactive oxygen species. [70] Also, total SH groups of pesticide workers were significantly lower than that of controls suggesting that there is suppression in total antioxidant capacity of the body. [71]

A number of other dietary antioxidants exist beyond the traditional vitamins present in plants collectively known as phytonutrients or phytochemicals which are being increasingly appreciated for their antioxidant activity, one example is flavonoids which are a group of polyphenolic compounds. Flavonoids exhibit several biological effects such as anti-inflammatory, anti-hepatotoxic, anti-allergic, and anti-ischemic. Many of the biological activities of flavonoids are attributed to their antioxidant properties and free radical scavenging capabilities. The antioxidant activities of flavonoids vary considerably depending

upon the different backbone structures and functional groups. A number of flavonoids efficiently chelate trace metals, which play an important role in oxygen metabolism. [72] This may be the mechanism by which radish and leek juices ameliorate the depleting effect of dimethoate upon total thiol groups.

4.4. Effect of radish and leek (leaves and roots) juices on hematological parameters

In hematological analysis, results showed tendency toward a decrease in haemoglobin concentration, hematocrit and red blood cells count in group administered dimethoate. The decrement in RBC_S count may be due to interference of Hb biosynthesis, disruption of erythropoiesis and shortening of the life span of circulating erythrocytes. The decrease in the haematocrit percent may be due to either a decrease in the size of RBC_S or a decrease in the number of erythrocytes. Decrease in the values of blood indices can be correlated with the decrease in the erythrocyte count and hemoglobin concentration. Also the results showed that there was stimulation in leukocyte production in dimethoate group may be due to the action of dimethoate as chemical stressors causing an increase in adrenaline level and consequently lymphatic leucocytosis. These results are confirmed by several studies. [73, 74, 75]

The administration of radish and leek (leaves and roots) juices normalized these hematological parameters. The protective effects are most likely due to their natural constituent of antioxidants potential. [61, 76]

4.5. Effect of radish and leek (leaves and roots) juices on Liver function

Liver is one of the major organs for detoxification of xenobiotics. A large number of xenobiotics caused oxidative stress by generation free radicals in biological systems. There is a considerable importance of the investigation into free radical-mediated damage to biological systems due to pesticide exposure. [77]

In clinical diagnosis increased AST, ALT and ALP activities indicates affected liver. In the present study, oral administration of DM caused oxidative stress and elevation of hepatospecific enzyme activities. This elevation in liver enzymes may be due to degeneration and necrosis of hepatocytes which attributes an increased permeability of the cell membrane that results in the release of transaminases and ALP into the blood stream as a result of the oral administration of DM. The present study is in a good agreement with the previous study in which exposure to dimethoate caused a significant increase in the serum AST, ALT and ALP activities. [78]

Treatment with radish and leek juices was able to alleviate the liver damage caused by DM exposure as revealed by remarkable decrease in these enzymes. This could be due to the impressive amount of polyphenols present in radish and leek juices which decrease the oxidative stress resulting in restoration the functions of the plasma membrane and decreasing release of enzymes into serum.

In a previous study, water and ethanol extracts of Raphanus sativus leaves (2 g/kg b.wt.) have shown a reduction in the elevated transaminases, ALP activity and total bilirubin in paracetamol induced toxicity in rabbits. [79] In another study, co-administration of Tunisian radish extract (TRE) at 5, 10, or 15 mg/kg b.wt., lead to a restoration of serum activities of ALT and AST, and a significant amelioration of the cadmium related effect on serum total protein levels. [80]

The reduced activities of AST, ALT and ALP as a result of ethanolic leek leaves extract (200 and 400 mg/kg b.wt.) administration in rat group administered CCl_4, point towards an improvement in the secretory mechanism of the hepatic cell and is a clear manifestation of anti-hepatotoxic effect of leek extract. [76] This effect was similar to that reported in a previous study. [81] Liver is central place for protein synthesis and damage to liver reduces the rate of protein synthesis and hence the level of proteins in the circulation. Low levels of total protein in DM treated group may be due to increased oxidative stress leading to elevated MDA and NO levels which attributed to liver dysfunction and inability to synthesize different proteins. In addition, the observed decrease in serum proteins could be attributed in part to the damaging effect of dimethoate on liver cells, as confirmed by the increase in activities of serum AST and ALT.

The elevation of total bilirubin levels could be as a result from degeneration in liver tissues and perturbation of the biliary system. Our results are in agreement with previous research. [3] The resulted increase in serum total protein and albumin after the administration of radish and leek juices may be due to the prevention of protein oxidation.

4.6. Effect of radish and leek (leaves and roots) juices on kidney function

The kidneys are the major detoxification organs for many xenobiotics, are frequently susceptible to the nephrotoxic effects. Kidney is one of the targets organs of experimental animals attacked by acute, sub-chronic and chronic exposure to OPI compounds. [5]

Increase in the serum urea and creatinine levels were taken as the index of nephrotoxicity induced by DM which caused elevation in ROS production thus increasing the oxidative stress in kidney tissue leading to impairment of the glomerular function, tubular damage in the kidneys and decrease in filtration rate of the kidney. This is confirmed by several studies.[46, 82]

Uric acid is the end product of the catabolism of tissue nucleic acids, i.e. purine and pyrimidine bases metabolism. In the present work, the serum uric acid level exhibited significant increment in dimethoate treated mice. This may be due to inability of excretion as a result of kidney oxidative damage. A rise in blood urea nitrogen, creatinine and uric acid levels was observed in dimethoate treated group indicating its oxidative effect. [59]

Interestingly, our results indicated that administration of radish and leek juices to dimethoate intoxicated mice restored these altered biochemical parameter levels to within normal limits and improved kidney dysfunction. This could be due to the phytoconstituents detected in the plant materials of radish and leek juices which may be responsible for their nephroprotective activity. In addition, these bioactive molecules also possess antioxidant activity. These results are confirmed by several studies. [62, 83, 84]

4.7. Histopathological Examination of liver and kidney

The administration of radish and leek juices to dimethoate intoxicated mice reversed some amelioration of liver and kidney lesions induced by dimethoate treatment. Histopathological observations were in correlation with biochemical measurements carried out in our study that further support the hepatoprotective and nephroprotective effects of these juices.

In the present study, radish and leek juices minimized the histopathological alterations in mice toxicated with dimethoate. It is concluded that this may be due to reduced lipid peroxidation processes and/or enhancement of anti-oxidant action by radish and leek juices.
In a previous study, Raphanus sativus exhibits nephroprotection against nephrotoxicity induced by gentamicin. They concluded that this may be due to its potent antioxidant effect.[62] Also Raphanus sativus was found to reduced necrosis or inflammation on CCl_4 induced liver toxicity in rats. [85]

Furthermore, it was indicated that liver of CCl_4-intoxicated rats fed on 200mg/kg b.wt. leek extract showed little vacuolar degeneration of hepatocytes while livers of CCl_4 intoxicated

rats fed on 400mg/kg b.wt. leek extract showed almost normal histology of the hepatic lobule. Also, liver of groups gives only leek extract showed as negative control rats. The higher concentrations of leek extract the improvement in liver histopathology. [76]

CONCLUSION

In conclusions, this study revealed that radish and leek (leaves and roots) juices are multi-protective agents that can protect humans against the oxidative stress and reduce consequently the risk of hepatotoxicity, nephrotoxicity and cellular damage in rat liver and kidney after oral exposure to dimethoate. However, the results revealed that radish leaves juice is most effective juice followed by leek leaves juice then radish roots juice and finally leek roots juice.

REFERENCES

1. Hagar HH, Fahmy AH. (A biochemical, histochemical, and ultrastructural evaluation of the effect of dimethoate intoxication on rat pancreas). Toxicol Lett, 2002; 133(2-3): 161-170.
2. John S, Kale N, Pathone N, Bahatnagar D. (Protective effect of vitamin E in dimethoate and malathion induced oxidative stress in rat erythrocytes). J Nutr Biochem, 2001; 12: 500-504.
3. Saafi EB, Louedi M, Elfeki A, Zakhama A, Najjar MF, Hammamia M, Achour L. (Protective effect of date palm fruit extract (Phoenix dactylifera L.) on dimethoate induced-oxidative stress in rat liver). Exp Toxicol Pathol, 2011; 63(5): 433-441.
4. IPCS/WHO. (Classification of pesticides by hazard and guidelines to classification). Switzerland: Geneva., 2001.
5. Sivapiriya V, Jayanthisakthisekaran, Venkatraman S. (Effects of dimethoate (O, O-dimethyl S-methyl carbamoyl methyl phosphorodithioate) and Ethanol in antioxidant status of liver and kidney of experimental mice). Pestic Biochem Physiol, 2006; 85(2): 115-121.
6. Kamath V, Joshi AKR, Rajini PS. (Dimethoate induced biochemical perturbations in rat pancreas and its attenuation by cashew nut skin extract). Pestic Biochem Phys, 2008; 90: 58–65.
7. Khan SM, Sobti RC, Kataria L. (Pesticide-induced alteration in mice hepatooxidative status and protective effects of black tea extract). Clinica Chimica Acta, 2005; 358: 131-138.

8. Yasin M, Sharma P. (Dimethoate 30EC induced histopathological effects on some organs of albino mice following oral exposure). International Journal of Recent Scientific Research, 2013; 4: 1327-1331.
9. Li HB, Wong CC, Cheng KW, Chen F. (Antioxidant properties in vitro and total phenolic contents in methanol extracts from medicinal plants). J Food Sci and Technol, 2008; 41: 385-390.
10. Shireen KF, Pace RD, Mahboob M, Khan AT. (Effects of dietary vitamin E, C and soybean oil supplementation on antioxidant enzyme activities in liver and muscles of rats). Food Chem Toxicol. 2008; 46:3290–4.
11. Poljsak B. (Strategies for reducing or preventing the generation of oxidative stress). Oxid Med Cell Longev, 2011; 2011: 1-15.
12. Nakamura Y, Iwahashi T, Tanaka A, Koutani J, Matsuo. T, Okamoto S, Sato K, Ohtsuki K. (4-methylthio-3-butenyl isothiocyanate, a principal antimutagen in daikon (Raphanus sativus; Japanese white radish). J Agric Food Chem, 2001; 49(12): 5755–60.
13. Beevi SS, Narasu ML, Gowda BB. (Polyphenolics profile, antioxidant and radical scavenging activity of leaves and stem of Raphanus sativus L). Plant Foods Hum Nutr, 2010; 65: 8-17.
14. Shukla S, Chatterji S, Mehta S, Rai PK, Singh RK, Yadav DK, Watal G. (Antidiabetic effect of Raphanus sativus root juice). Pharm Biol, 2011; 49(1): 32–7.
15. Salah-Abbès JB, Abbès S, Abdel-Wahhab M, Oueslati R. (Immunotoxicity of zearalenone in Balb/c mice in a high subchronic dosing study counteracted by Raphanus sativus extract). Immunopharmacol Immunotoxicol, 2010; 32(4):628–36.
16. Jan M and Badar A. (Effect of crude extract of Raphanus sativus roots on isolated trachea of albino rat). Pak J Physiol, 2012; 8(1): 23-26.
17. Beevi SS, Mangamoori LN, Subathra M, Edula JR. (Hexane extract of Raphanus sativus L. roots inhibits cell proliferation and induces apoptosis in human cancer cells by modulating genes related toapoptotic pathway). Plant Foods Hum Nutr, 2010; 65(3): 200–9.
18. Ghayur MN, Gilani AH. (Gastrointestinal stimulatory and uterotonic activities of dietary radish leaves extract are mediated through multiple pathways). Phytother Res, 2005; 19(9): 750–5.
19. Chun OK, Chung SJ, Song WO. (Estimated dietary flavonoid intake and major food sources of U.S. adults). J Nutr, 2007; 137(5): 1244-52.

20. Nimni ME, Han B, Cordoba F. (Are we getting enough sulfur in our diet?). Nutr Metab (Lond), 2007; 6(4): 24-36.
21. Onyeagba RA, Ugbogu OC, Okeke CU, Iroakasi O. (Studies on the antimicrobial effects of garlic (Allium Sativum Linn), ginger (Zingiber Officinale Roscoe) and lime (Citrus Aurantifolia Linn). Afr J Biotechnol, 2004; 3: 552-554.
22. Xiao HB, Fang J, Lu XY, Chen XJ, Tan C, Sun ZL. (Protective effects of kaempferol against endothelial damage by an improvement in nitric oxide production and a decrease in asymmetric dimethylarginine level). European Journal of Pharmacology, 2009; 616(1-3): 213-222.
23. Adão CR, da Silva BP, JPParente. (A new steroidal saponin with anti-inflammatory and antiulcerogenic properties from the bulbs of Allium ampeloprasum var. porrum). Fitoterapia, 2011; 82: 1175–1180.
24. Sadeek E. (Protective effect of fresh Juice from red beetroot (Beta vulgaris L.) and radish (Raphanus sativus L.) against carbon tetrachloride induced hepatotoxicity in rat models). African J Biol Sci, 2011; 7(1): 69-84.
25. National Research Council. Nutrient requirements of laboratory animals.4th ed. National Academy Press, Washington, DC, chapter 1995.
26. Goupy P, Hugues M, Boivin P, Amiot MJ. (Antioxidant composition and activity of barley (Hordeum vulgare) and malt extracts and of isolated phenolic compound). J Sci Food Agric, 1999; 79: 1625-1634.
27. Mattila P, Astola J, Kumpulainen J. (Determination of flavonoids in plant material by HPLC with diode-array and electro-array detections). J Agric Food Chem, 2000; 48: 5834-5841.
28. Wu QE, Ban TT, Chang XL, Wu Q, Zhou ZJ. (Effects of acute and subchronic exposures to dimethoate on rat cerebral cortex GABAergic system). J Health Sci, 2010; 56(3): 267-274.
29. Montgomery HA, Dymock JF. (Determination of nitrite in water). J Analyst, 1961; 86: 414-416.
30. Ohkawa H, Oshini W, Yogi K. (Assay for lipid peroxide in animal tissues by thiobarbituric acid reaction). Biochem, 1979; 95: 351-358.
31. Beutlar E, Duron O, Kelly BM. (Improved method for the determination of blood glutathione). J Lab Clin Med, 1963; 61: 882-888.

32. Nishikimi N, Roa NA, Yogi K. (The occurrence of superoxide anion in the reaction of reduced phenazine methosulphate and molecular oxygen). J Biochem Biophys Res Commun, 1972; 46: 849-853.
33. Witko V, Nguyen AT, Descamps-Latscha B. (Microtiter plate assay for phagocyte derived taurine-chloramines). J Clin Lab Anal., 1992; 6(1): 47–53.
34. Sedlak J, Lindsay RH. (Estimation of total, protein-bound, and non-protein sulfhydryl groups in tissue with Ellman's reagent). Anal Biochem, 1968; 25(1): 192–205.
35. Dacie JV, Lewis SM. (Practical Haematology). Churchil Livingstone, 1984; 1-453.
36. Reitman A, Frankel S. (A colorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminases). Amer J Clin Path, 1957; 28: 56- 63.
37. Belfield A, Goldberg DM. (Colorimetric determination of alkaline phosphatase activity). Enzyme, 1971; 12: 451.
38. Walter M, Gerade H. (A colorimetric method for determination bilirubin in serum and plasma). Micro Chem J, 1970; 15: 231–236.
39. Gornal AC, Bardawill CJ, David MM. (Determination of serum proteins by means of the biuret reaction). J Biol Chem, 1949; 177: 751–766.
40. Doumas BT, Watson WA, Biggs HG. (Albumin standards and the measurements of serum albumin with bromocresol green). Clin Chim Acta., 1971; 31(1): 87-96.
41. Fawcett JK, Soctt JE. (Urea determination after enzymatic hydrolysis). J Clin Path, 1960; 13: 156-159.
42. Schirmeister J. (Determination of creatinine in serum). Dtsch Med Wschr, 1964; 89: 1940.
43. Barham D, Trinder P. (Enzymatic determination of uric acid). Analyst, 1972; 97: 142-145.
44. Soininen TH, Jukarainen N, Soininen P, Auriola S, Riitta Julkunen-Tiitto R, Oleszek W, Stochmal A, O. Karjalainene R, Vepsäläinen JJ, (Metabolite profiling of leek (Allium porrum L) cultivars by 1H NMR and HPLC–MS). Phytochem. Anal, 2014; 25: 220–228.
45. Heikal TM, Mossa ATH, Nawwar GAM, El-Sherbiny M, Ghanem HZ. (Protective effect of a synthetic antioxidant acetyl gallate derivative against dimethoate induced DNA damage and oxidant/antioxidant status in male rats). Environmental and Analytical Toxicology, 2012; 2(7): 155.
46. El-Damaty MA, Farrag AH, Rowayshed G, Fahmy HM. (Biochemical and histopathological effects of systemic pesticides on some functional organs of male albino rats). Journal of Applied Sciences Research, 2012; 8(11): 5459-5469.

47. Baconi DL, Barca M, Manda G, Ciobanu, AM, Balalau C. (Investigation of the toxicity of some organophosphorus pesticides in a repeated dose study in rats). Rom J Morphol Embryol, 2013; 54(2): 349-356.
48. Balkan S, Akta T. (Study on the liver functions in rats exposed tobenomyl). J Biol Sci, 2005; 5(5): 666-669.
49. Schutzendubel A, Polle A. (Plant responses to abiotic stresses heavy metal-induced oxidative stress and protection by mycorrhization). Journal of Experimental Botany, 2002; 53: 1351-1365.
50. Sharma Y, Bashir S, Irshad M, Nagc TC, Dogra TD. (Dimethoate-induced effects on antioxidant status of liver and brain of rats following subchronic exposure).Toxicology, 2005b; 215: 173–81.
51. Astiz M, de Alaniz MJT, Marra CA. (Antioxidant defense system in rats simultaneously intoxicated with agrochemicals). Environmental Toxicology and Pharmacology, 2009; 28: 465–473.
52. Alp H, Aytekin I, Hatipoglu N.K, Alp A, Ogun M (Effects of sulforophane and curcumin on oxidative stress created by acute malathion toxicity in rats). European Review for Medical and Pharmacological Sciences, 2012; 16: 144-148.
53. Alp H, Aytekin I, Atakisi O, Hatipoglu NK, Basarali K, Ogun M, Buyukbas S, Altintas L, Ekici H, Alp A. (The effects of caffeic acid phenethyl ester and ellagic acid on the levels of malondialdehyde, reduced glutathione and nitric oxide in the lung, liver and kidney tissues in acute diazinon toxicity in rats). Journal of Animal and Veterinary Advances, 2011; 10(11): 1488-1494.
54. Gaona-Gaonaa L, Molina-Jiona E, Tapiab E, Zazuetac C, Hernandez-Pandod R, Calderon-Olivera M, Zarco-Marqueza G, Pinzone E, Pedraza-Chaverria J. (Protective effect of sulforaphane pretreatment against cisplatin-induced liver and mitochondrial oxidant damage in rats). Toxicology, 2011; 286: 20–27.
55. Syed SN, Rizvi W, Kumar A, Khan AA, Moin S, Ahsan A.(In vitro antioxidant and in vivo hepatoprotective activity of leave extract of Raphanus sativus in rats using CCl4 model). Afr J Tradit Complement Altern Med, 2014; 11(3): 102-106.
56. Tsai TH, Tsai PJ, Ho SC. (Antioxidant and anti-inflammatory activities of several commonly used spices). Journal of food science, 2005; 70(1): 93-97.
57. Halliwell B. (Role of free radicals in the neurodegenerative diseases, therapeutic implications for antioxidant treatment). J Drugs and Aging, 2001; 18(9): 685-716.

58. Demir S, Yilmaz M, Koseglu M, Aydin A. (Role of free radicals in peptic ulcer and gastritis).Turk J Gastroenterol, 2003; 14: 39-43.
59. Nazam N, Lone MI, Sharma M, Khan AA, Kelany AM, Ahmad W. (Biochemical and cytoarchitechtural evaluation of dimethoate intoxication in rat liver and kidney: An in vivo study). Indo American Journal of Pharm Research, 2015; 5(03): 1127-1137.
60. Ben Amara I, Soudani N, Troudi A, Bouaziz H, Boudawara T, Zeghal N. (Antioxidant effect of vitamin E and selenium on hepatotoxicity induced by dimethoate in female adult rats). Ecotoxicology and Environmental Safety, 2011; 74: 811–819.
61. Salah-Abbès JB, Abbès S, Ouanes Z, Houas Z, Abdel-Wahhab MA, Bacha H, Oueslati R. (Tunisian radish extract (Raphanus sativus) enhances the antioxidant status and protects against oxidative stress induced by zearalenone in Balb/c mice). J Applied toxicology, 2008; 28: 6–14.
62. Kishor Kumar S, Sridher Rao K, Sridhar Y, Shankaraiah P. (Effect of Raphanus sativus Linn. against gentamicin induced nephrotoxicity in rats). J Advanced Pharmaceutical Sciences, 2013; 3: 355-365.
63. Borek C. (Antioxidant health effects of aged garlic extract). The Journal of nutrition, 2001; 131(3): 1010S.
64. Abdel-Wahhab MA, Aly SE. (Antioxidant property of Nigella sativa (black cumin) and Syzygium aromaticum (clove) in rats during aflatoxicosis). J Appl Toxicol, 2005b; 25: 218–223.
65. Zuwała-Jagiełło J, Pazgan-Simon M, Simon K, Warwas M. (Elevated advanced oxidation protein products levels in patients with liver cirrhosis). Acta biochemica polonica, 2009; 56(4): 679–685.
66. Loeckie Meerman LZ JHN, Commandeur JNM, Vermeulen NPE. (Biomarkers of free radical damage: applications in experimental animals and in humans). Free Radic Med, 1999; 26: 202-226.
67. Ben Amara I, Soudani N, Hakim A, Troudi A, Zeghal KM, Boudawara T, Zeghal N. (Protective effects of vitamin E and selenium against dimethoate induced cardiotoxicity in vivo: biochemical and histological studies). Wiley Online Library, 2011a; 1-14.
68. Ben Amara I, Soudani N, Hakim A, Troudi A, Zeghal KM, Boudawara T, Zeghal N. (Selenium and vitamin E, natural antioxidants, protect rat cerebral cortex against dimethoate-induced neurotoxicity). Pesticide Biochemistry and Physiology, 2011b; 101: 165–174.

69. Prakash M, Upadhya S, Prabhu R. (Protein thiol oxidation and lipid peroxidation in patients with uremia). Scand J Clin Lab Invest, 2004; 64: 599-604.
70. Kale B. (Correlation of oxidative stress and antioxidant status with cholinesterases in different grades of organophosphorus toxicity). International Journal of Scientific Research in Environmental Sciences, 2013; 1(5): 85-91.
71. Ranjbar A, Pasalar P, Abdollahi M. (Induction of oxidative stress and acetylcholinesterase inhibition in organophosphorous pesticide manufacturing workers). Human & Experimental Toxicology, 2002; 21: 179 – 182.
72. Sun AY, Simnyi A, Sun GY. (Neuroprotective effects of polyphenols). J Free Rad Biol Med, 2002; 32(4): 314-318.
73. Lone MY, Baba BA, Raj P, Shrivastava1 VK, Bhide M. (Haematological and hepatopathological changes induced by dimethoate in Rattus rattus). Indo American Journal of Pharmaceutical Research, 2013; 3(6): 4360: 4365.
74. Lu wang Q, Zhang Y, Zhou C, Zhang J, Dou Y, Li Q. (Risk assessment of mouse gastric tissue cancer induced by dichlorvos and dimethoate). Oncology Letters 5, 2013; 1385-1389.
75. Jain N, Sharma P, Sharma N, Joshi SC. (Haemato-biochemical profile following subacute toxicity of malathion in male albino rats). Pharmacologyonline, 2009; 2: 500-506.
76. Nasir AS. (Hepatoprotective and some haematological parameters effect of Allium ampeloprasum against carbon tetrachloride induced liver toxicity in albino rats). Kufa Journal for Veterinary Medical Sciences, 2012; 3(2): 117-126.
77. Salim AB, Abou-Arab AAK, Mohamed SR, Eldesouky TA. (Influence of pomegranate (Punica granatum L.) on dimethoate induced hepatotoxicity in rats). International Journal of Biological, Food, Veterinary and Agricultural Engineering, 2014; 8(8): 896- 901.
78. Attia, AM, Nasr, HM. (Dimethoate-induced changes in biochemical parameters of experimental rat serum and its neutralization by black seed (Nigella sativa L.) oil). Slovak Journal of Animal Science, 2009; 42(2): 87–94.
79. Anwar R, Ahmed M. (Studies of R. sativus as hepatoprotective agent). Journal of medical sciences, 2006; 6(4): 662-665.
80. Salah-Abbe`s J, Abbe`s S, Zohra H, Oueslati R. (Tunisian radish (Raphanus sativus) extract prevents cadmium-induced immunotoxic and biochemical alterations in rats).J Immunotoxicol. Early Online, 2014; 1–8.

81. Durak L, Kavutcu M, Aytac B, Avci E, Devrim, H. (Effect of leek extract consumption on blood lipid and oxidant/antioxidant parameters in humans with high blood cholesterol). J Nutr Biochem, 2004; 15: 373-377.
82. Mahjoubi-Samet A, Fetoui H, Zeghal N. (Nephrotoxicity induced by dimethoate in adult rats and their suckling pups). Pesticide Biochemistry and Physiology, 2008; 91: 96–103.
83. Abed SA, El-Shazely MO, Ahmed KA, Abdel-mawla EM, Ibrahim AK. (Pathological, immunohistochemical and biochemical studies on the therapeutic effect of Raphanus Sativus oil on streptozotocin induced diabetic rats). Egypt J Comp Path &Clinic Path, 2015; 28(1): 1- 17.
84. Badary OA, Yassinb NAZ, El-Shenawy SMA , Abd EL-Moneemc M, AL-Shafeiyb HM. (Study of the effect of Allium porrum on hypertention induced in rats). Rev Latinoamer Quím, 2013; 41(3): 149-160.
85. Lee SW, Yang KM, Kim JK, Nam BH, Lee CM, Jeong MH, Seo SY, Kim GY, Jo1 WS. (Effects of white radish (Raphanus sativus) enzyme extract on hepatotoxicity). Official journal of Korean society of toxicology, 2012; 28(3): 165-172.