



Original Research Article

Protective effect of flaxseed oil and vitamin E on potassium Bromate-induced oxidative stress in male rats

Hala M. A.Wahba¹ and Thanaa A.A. Ibrahim²

¹Home Economics Department, Faculty of Specific Education, Minufiya University, Egypt

²Family and Child Sciences Department -Nutrition and Food Science, Faculty of Arts and Sciences, Qassim University, Onaizah, Saudi Arabia

*Corresponding author e-mail: hala.ali77@yahoo.com

ABSTRACT

Keywords

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The protective effect of flaxseed oil (FSO) and vitamin E (Vit E) on potassium bromate (KBrO₃)-induced oxidative stress and hepatonephrotoxicity in male rats was investigated. Forty five mature male rats were randomly distributed into 5 groups (n=9). Group (1) was negative control and the other 4 groups were injected with a single dose of KBrO₃ (125 mg/kg, i.p.) to induce oxidative stress. Group (2) was used as a positive control, while groups (3), (4) and (5) were orally given FSO (1 ml/rat, 500 mg/kg), Vit E (0.5 ml/rat, 50 mg/kg) and FSO and Vit E in combination, by the same doses daily for 8 weeks respectively. Blood samples were collected for separating the serum for assessment of hepatorenal function. Livers and kidneys were taken for preparing tissue homogenates for biochemical analysis. The results showed that oxidative stress induced by KBrO₃ in rats caused significant decreases in body weight gain and feed efficiency ratio. It also significantly increased serum biomarkers of hepatorenal function, increased tissue lipid peroxidation, and decreased the activity of hepatic and renal antioxidant enzymes glutathione peroxidase (GPx), superoxide dismutase (SOD) and catalase (CAT). Oral coadministration of FSO and Vit E increased body weight gain and feed efficiency ratio. It improved hepatorenal function, reduced tissue lipid peroxidation and normalized the activity of antioxidant enzymes in liver and kidneys of rats given KBrO₃. In conclusion, coadministration of FSO and Vit E induces good antioxidant activity and alleviates oxidative stress in rats. The study suggests that intake of flaxseed oil with vitamin E may be beneficial as a food supplement for patients who suffer from oxidative stress and hepatonephrotoxicity.

Introduction

Oxidative stress is a major predisposing risk factor of many chronic diseases. It arises from an imbalance between the production of reactive oxygen species (ROS) and the body's antioxidant defenses them, which induces tissue damage such

as hepatonephrotoxicity. The body antioxidant defenses enable the system to remove ROS, restore the prevailing reducing environment and repair the tissue damage (Halliwell and Gutteridge, 1999). Free radicals such as nitric oxide (NO) and

superoxide ions are produced as second messengers, particularly by immune cells. Superoxide reacts rapidly with nitric oxide by nitric oxide synthase to produce peroxynitrite, whereas hydrogen peroxide (H₂O₂) slowly decomposes to the highly reactive hydroxyl radicals. Both peroxynitrite and hydroxyl radicals are highly reactive oxidizing agents, capable of oxidation of proteins, lipids, and DNA (Pryor and Squadrito, 1995 and Beckman and Koppenol, 1996). Oxidative stress plays an important role in the etiology and pathogenesis of many chronic diseases such as hepatotoxicity, nephrotoxicity, atherosclerosis, hypertension, diabetes mellitus and cancers (Reuter *et al.*, 2010 and Krajcovicova *et al.*, 2012).

Potassium bromate (KBrO₃) is widely used as a food additive in the bread making processes and found in drinking water samples as a byproduct of ozone disinfection. KBrO₃ causes nephrotoxicity and renal cell cancer in rats (Kurokawa *et al.*, 1990). Administration of KBrO₃ to rats was found to induce oxidative stress and passively impair the antioxidant power of rat blood (Ahmed and Mahmood, 2012).

Dietary intake of antioxidants can inhibit or delay the oxidation of susceptible cellular substrates so prevent oxidative stress. Phenolic compounds such as flavonoids, phenolic acids, diterpenes, saponins and tannins have received much attention for their high antioxidative activity (Rice-Evan *et al.*, 1996). Therefore, it is important to enrich our diet with antioxidants to protect against many chronic diseases related to oxidative damage (Rubiolo *et al.*, 2008). In addition, antioxidants play an important role in food quality preservation due to their ability to

prevent oxidative deterioration of lipids (Erukainure *et al.*, 2012).

Flaxseed contains 35% of its mass as oil, of which 55% is alpha-linolenic acid (ALA, n-3, omega-3 fatty acid). Dietary omega-3 polyunsaturated fatty acids (PUFAs) have been shown to reduce the severity of numerous ailments. Previous studies reported that flaxseed is a source of hypocholesterolemic and anti-atherogenic agents (Prasad, 2000 and Lee and Prasad, 2003). Flaxseed oil (Omega nutrition) suppresses oxygen radical production by white blood cells and improves cardiovascular health.

Suppression of atherosclerosis by flaxseed is associated with lowering of serum lipids and decreasing oxidative stress and so it may prevent cardiovascular diseases (Prasad, 2009 and Xu *et al.*, 2012). Flaxseed oil also produces a protective effect against sodium nitroprusside-induced nephrotoxicity and oxidative damage in male rats (Khan *et al.* 2012).

Vitamin E (alpha-tocopherol) is a fat soluble vitamin which regulates different oxidation processes in the body as it acts as a powerful antioxidant. Previous studies revealed that dietary intake of vitamin E can normalize the damaging effect of oxidative stress induced by oxygen free radicals (Shalaby *et al.*, 2004; Ramachandran and Raja, 2010; Iranloye and Oludare, 2011 and Abbas and Sakr, 2013).

The present study was designed to investigate the protective effect of flaxseed oil and vitamin E on oxidative stress induced by potassium bromate in male rats.

Materials and Methods

Flaxseed oil and vitamin E

Flaxseed oil was purchased from Arab Company for Vegetable Oils Extraction and Refining (ARECO), Egypt. Vitamin E was obtained from Pharco Company for Pharmaceutical, Alexandria, Egypt. It is dispensed in the form of soft gelatinous capsules each containing 400 mg of dl-alpha tocopherol acetate.

Chemicals and kits

Potassium bromate ($KBrO_3$) was purchased from El-Gomhoryia Company, Cairo Egypt, in the form of a white powder. Biochemical kits for the determination of serum biomarkers of liver and kidney functions and activity of tissue antioxidant enzymes were purchased from Gamma Trade for Company Pharmaceutical and Chemicals, Dokki, Egypt.

Rats

Forty five mature male rats of Sprague Dawley strain weighing 175 ± 5 g each and 8-10 weeks old were obtained from the Laboratory Animals Farm, Helwan, Egypt. The rats were housed at a controlled room temperature of $23 \pm 1^\circ C$, 55 % humidity and under a 12-hr light / 12-hr dark cycles. The animals were fed on basal diet and water was provided *ad libitum* for one week before the start of experiment for acclimatization.

Preparation of basal diet

The basal diet was prepared using AIN-93 according to Reeves *et al.* (1993). It consists of 20 % protein (casein), 35 % sucrose, 7% corn oil, 2% choline chloride,

1% vitamin mixture, 3.5 % salt mixture and 5% fibers. The remainder was corn starch up to 100%.

Experiment and grouping of rats

After one week adaptation period, the rats were randomly distributed into five equal groups, of 9 animals each. Group (1) was fed on basal diet and kept without any treatment as a negative control. Rats of the other four groups were injected by a single intraperitoneal dose of potassium bromate at 125 mg/kg b.wt.

for induction of oxidative stress (Khan and Sultana 2004). Group (2) was left as a positive control, while groups (3), (4) and (5) were orally given flaxseed oil (FSO) in a dose of 500 mg/kg (1 ml/rat), vitamin E (Vit. E) in a dose of 50 mg/kg (0.5ml/rat) and FSO with Vit. E for 8 weeks, respectively. The selected dose (500 mg/kg) of flaxseed oil was previously used by Abdel Moneim (2012) and the dose (50 mg/kg) of vitamin E was selected according to Ganie *et al.* (2013).

The food intake was recorded daily and the body weight gain (g) was calculated. Feed efficiency ratio (FER) was calculated using this formula: FER = feed intake (g)/weight gain (g). At the end of experiment, the rats were anesthetized using ether anesthetic and blood samples were collected into clean centrifuge tubes to obtain the serum which used for biochemical analyses. Livers and kidneys were immediately removed, rinsed with saline, blotted on filter paper and stored at $-70^\circ C$ pending for the preparation of tissue homogenate for biochemical assays.

Blood sampling

Blood samples were withdrawn from the

orbital plexus of veins in the inner canthus of eye using capillary microtubes. Blood was left for 10 min. at room temperature to clot. Serum samples were obtained by centrifugation at 4000 rpm for 15 min. and directly frozen at -18°C till biochemical analyses.

Biochemical analyses

Activities of serum liver enzymes aspartate and alanine aminotransferases (AST and ALT) were chemically determined according to Bergmeyer *et al.* (1978). Serum total cholesterol (Ratliff and Hall, 1973) and triglycerides (Jacob and Van-Denmark, 1963) were chemically determined. Blood urea nitrogen (BUN) was determined using BioMérieux kits according to Patton and Crouch (1977). Serum uric acid was determined using the enzymatic colorimetric method as described by Fossati *et al.* (1980). Serum creatinine concentrations were colorimetrically determined by the Jaffe reaction (Husdan and Rapoport, 1968). Serum lipid peroxide malondialdehyde (MDA) and reduced glutathione (GSH) contents were estimated according to methods described by Placer *et al.* (1966) and Afzal *et al.* (2002), respectively. Measurements were performed using Spectrophotometer (Model T80, UV/visible, double beam, UK).

Preparation of tissue homogenate

One gram of frozen liver and kidney tissues was collected, washed on ice-cooled 0.9% NaCl solution and homogenized in 100 ml ice-cooled 1.15% solution of potassium chloride and 50 mM potassium phosphate buffer solution (pH 7.4) to yield 10% homogenate (W/V). Homogenization was performed using Sonicator, 4710 Ultrasonic Homogenizer

(Cole-Parmer Instrument Co., USA). The homogenate was then centrifuged at 4000 rpm for 15 minutes at 4°C and the supernatant was collected for further use.

Lipid peroxidation and tissue antioxidant enzymes

Lipid peroxidation (LPO) was determined by quantifying malondialdehyde (MDA) that formed in terms of thiobarbituric acid reactive substances (TBARS). Tissue homogenates were used for determination of tissue lipid peroxide (MDA), enzymatic (GPx, SOD and CAT) and non enzymatic (GSH) antioxidants. The reduced glutathione (GSH) content in liver and kidney homogenates was determined colorimetrically by the method modified by Bulaj *et al.* (1998). Lipid peroxide (MDA) was determined according to Ohkawa *et al.* (1979). The activities of liver glutathione peroxidase, superoxide dismutase and catalase antioxidant enzymes were determined chemically according to Paglia and Valentine (1979), Spitz and Oberley (1989) and Sinha (1972), respectively.

Statistical analysis

Data were presented as means \pm SD and statistically analyzed using one way analysis of variance (ANOVA) test followed by Duncan's multiple range test. (Snedecor and Cochran, 1986) using computerized SPSS program.

Result and Discussion

Intraperitoneal injection of potassium bromate in a single (125mg/kg b.wt.) dose to rats caused significant ($P < 0.05$ and $P < 0.001$) decreases in body weight gain, feed intake and feed efficiency ratio (FER) when compared to the negative control

group. In rats with oxidative stress induced by potassium bromate, oral administration of flaxseed oil and vitamin E, alone and in combination, for 8 weeks significantly ($P < 0.05$ and $P < 0.001$) increased body weight gain, feed intake and FER compared to the positive control group (Table 1).

Data in Table (2) showed that rats when injected intraperitoneally with potassium bromate had significant ($P < 0.05$ and $P < 0.001$) increases in serum levels of AST, ALT, total cholesterol (TC) and triglycerides (TG) when compared to the negative control group. Oral administration of flaxseed oil and vitamin E, alone and in combination, to rats with oxidative stress produced significant ($P < 0.05$ and $P < 0.01$) decreases in the elevated serum levels of AST, ALT, TC and TG when compared with the positive control group.

Rats with oxidative stress induced by potassium bromate had significant ($P < 0.05$ and $P < 0.001$) increases in blood urea nitrogen (BUN), uric acid (UA) and creatinine (Cr) levels in the serum when compared to negative control rats. Oral administration of flaxseed oil and vitamin E, alone and in combination, to rats with oxidative stress produced significant ($P < 0.01$) decreases of the elevated BUN, UA and Cr concentrations when compared with the positive control group as recorded in Table (3).

Data in Table (4) showed that intraperitoneal injection of potassium bromate to rats caused an elevation in serum level of lipid peroxide malondialdehyde (MDA) and lowering the level of reduced glutathione (GSH) as compared with negative control rats. Flaxseed oil and vitamin E when concomitantly given orally to rats with

oxidative stress induced a significant ($P < 0.05$ and $P < 0.001$) decrease in MDA and an increase in GSH levels in the serum when compared with the positive control group.

In liver tissues of rats with oxidative stress, there was a significant ($P < 0.05$ and $P < 0.001$) an increase in levels of MDA and decrease in levels of GSH when compared with the negative control group. Oral coadministration of flaxseed oil and vitamin E significantly reduced liver MDA level and increased GSH level when compared with the positive control group as shown in Table (5).

In kidney tissues of rats with oxidative stress, there was a significant ($P < 0.05$ and $P < 0.001$) increase in levels of MDA and decrease in levels of GSH when compared with the negative control group. Flaxseed oil and vitamin E significantly reduced kidney MDA level and increased GSH level when compared with the positive control group as shown in Table (6).

Concerning the activity of liver antioxidant enzymes, the rats with oxidative stress showed decreased activity of glutathione peroxidase (GPx), superoxide dismutase (SOD) and catalase (CAT) enzymes in liver tissues as compared to the negative control group. Oral coadministration of flaxseed oil with vitamin E produced significant ($P < 0.05$ and $P < 0.001$) increases in the activity of GPx, SOD and CAT enzymes in liver tissues as compared with the positive control group (Table 7).

The goal of the present study was to investigate the protective effect of coadministration of flaxseed oil (FSO)

Table.1 Effect of flaxseed oil (FSO) and vitamin E (Vit E) on body weight gain, feed intake and feed efficiency ratio (FER) in rats with oxidative stress.

Groups Parameters	Control (-ve)	Control (+ve)	FSO (500mg/kg)	Vit. E (50 mg/kg)	FSO + Vit. E
Initial weight (g)	170.00 ± 4.14 ^a	177.50 ± 6.31 ^a	175.00 ± 4.55 ^a	178.50 ± 4.36 ^a	180.00 ± 4.66 ^a
Final weight (g)	258.50 ±14.20 ^a	213.50 ±12.71 ^{b*}	240.06 ±12.98 ^a	243.19 ±14.66 ^a	252.50 ±13.28 ^{c*}
Weight gain (%)	52.05 ± 2.65 ^a	20.28 ± 3.21 ^{c***}	37.17 ± 5.11 ^{b*}	36.24 ± 4.13 ^{b*}	40.27 ± 4.11 ^{c**}
Feed intake (g/w)	18.30 ± 1.26 ^a	17.55 ± 1.31 ^{b*}	18.25 ± 1.51 ^{b*}	17.88 ± 1.61 ^{b*}	18.20 ± 1.65 ^{c*}
FER	0.206 ± 0.002 ^a	0.048 ± 0.004 ^{d***}	0.061 ± 0.002 ^{c**}	0.065 ± 0.003 ^{c**}	0.074 ± 0.003 ^{b***}

Mean ± SD values in each row with different superscripts (a, b, c, d) are significant when compared to the control groups at * $P < 0.05$ ** $P < 0.01$ *** $P < 0.001$ n= 9 rats/group.

Table.2 Effect of flaxseed oil (FSO) and vitamin E (Vit.E) on serum AST, ALT, total cholesterol (TC) and triglycerides (TG) in rats with oxidative stress.

Groups Parameters	Control (-ve)	Control (+ve)	FSO (500mg/kg)	Vit. E (50 mg/kg)	FSO + Vit. E
AST (U/dl)	62.44 ±2.33 ^c	84.22 ±4.11 ^{a***}	77.22 ±4.10 ^{b*}	75.33 ±3.66 ^{b*}	65.11 ±3.80 ^{b*}
ALT (U/dl)	34.11 ±2.11 ^c	56.33 ±3.71 ^{a***}	46.22 ± 3.71 ^{b*}	43.15 ±5.66 ^{b*}	37.34 ±4.28 ^{c*}
TC (mg/dl)	115.9 ±3.22 ^d	135.5 ±2.44 ^{a***}	129.4 ±3.15 ^{b**}	127.3 ±2.13 ^{b*}	122.4 ±1.16 ^{c***}
TG (mg/dl)	95.8 ±2.12 ^d	119.3 ±3.14 ^{a***}	114.4 ±2.52 ^{b**}	116.7 ±2.41 ^{b*}	109.5 ±1.90 ^{c***}

Mean ± SD values in each row with different superscripts (a, b, c) are significant when compared to the control groups at * $P < 0.05$ ** $P < 0.01$ *** $P < 0.001$ n = 9 rats/group

Table.3 Effect of flaxseed oil (FSO) and vitamin E (Vit. E) on blood urea nitrogen, uric acid and creatinine concentrations in rats with oxidative stress.

Groups Parameters	Control (-ve)	Control (+ve)	FSO (500mg/kg)	Vit. E (50 mg/kg)	FSO + Vit. E
Urea nitrogen (mg/dl)	34.00 ±2.34 ^d	66.63 ±2.54 ^{a***}	40.55 ±4.11 ^{b*}	38.65 ±3.13 ^{b*}	36.44 ±2.11 ^{c*}
Uric acid (mg/dl)	1.55 ±0.12 ^d	2.75 ±0.17 ^{a**}	1.80 ±0.15 ^{b**}	1.75 ±0.13 ^{b**}	1.62 ±0.16 ^{c**}
Creatinine (mg/dl)	1.10 ±0.02 ^d	1.60 ±0.04 ^{a**}	1.40 ±0.02 ^{b**}	1.30 ±0.01 ^{b***}	1.26 ±0.02 ^{b***}

Mean ± SD values in each row with different superscripts (a, b, c, d) are significant when compared to the control groups at * $P < 0.05$ ** $P < 0.01$ *** $P < 0.001$ n= 9 rats/group

Table.4 Effect of flaxseed oil (FSO) and vitamin E (Vit E) on serum malondialdehyde (MDA) and reduced glutathione (GSH) in rats with oxidative stress.

Groups Parameters	Control (-ve)	Control (+ve)	FSO (500mg/kg)	Vit E (50 mg/kg)	FSO + Vit E
MDA (mmol/ml)	34.11 ± 2.21 ^d	57.33 ± 3.61 ^{a***}	44.55 ± 2.53 ^{b*}	43.19 ± 2.46 ^{b*}	38.50 ± 2.48 ^{c*}
GSH (mmol/ml)	64.36 ± 3.71 ^b	48.11 ± 2.91 ^{d***}	59.21 ± 3.11 ^{c**}	60.31 ± 2.98 ^{b**}	66.31 ± 3.98 ^{a***}

Mean ± SD values in each raw with different superscripts (a, b, c, d) are significant when compared to the control groups at * $P < 0.05$ ** $P < 0.01$ *** $P < 0.001$ n = 9 rats/group

Table.5 Effect of flaxseed oil (FSO) and vitamin E (Vit.E) on liver malondialdehyde (MDA) and reduced glutathione (GSH) in rats with oxidative stress.

Groups Parameters	Control (-ve)	Control (+ve)	FSO (500 mg/kg)	Vit.E (50 mg/kg)	FSO + Vit.E
MDA (nmol/min/mg protein)	0.34 ± 0.002 ^d	0.71 ± 0.003 ^{a***}	0.54 ± 0.001 ^{b**}	0.51 ± 0.003 ^{b**}	0.32 ± 0.002 ^{c***}
GSH (nmol/min/mg protein)	22.50 ± 1.25 ^a	10.66 ± 1.15 ^{d***}	18.95 ± 2.71 ^{b*}	19.46 ± 2.66 ^{b**}	21.53 ± 3.28 ^{c***}

Mean ± SD values in each raw with different superscripts (a, b, c, d) are significant when compared to the control groups at * $P < 0.01$ ** $P < 0.01$ *** $P < 0.001$ n = 9 rats/group

Table.6 Effect of flaxseed oil (FSO) and vitamin E (Vit.E) on kidney malondialdehyde (MDA) and reduced glutathione (GSH) in rats with oxidative stress

Groups Parameters	Control (-ve)	Control (+ve)	FSO (500 mg/kg)	Vit.E (50 mg/kg)	FSO + Vit.E
MDA (nmol/min/mg protein)	2.02 ± 0.002 ^d	6.22 ± 0.03 ^{a***}	3.63 ± 0.01 ^{b**}	3.84 ± 0.03 ^{b**}	2.30 ± 0.02 ^{c***}
GSH (nmol/min/mg protein)	32.50 ± 1.25 ^a	20.66 ± 1.15 ^{d***}	18.95 ± 1.61 ^{b*}	17.46 ± 1.22 ^{b**}	15.53 ± 2.18 ^{c***}

Mean ± SD values in each raw with different superscripts (a, b, c, d) are significant when compared to the control groups at * $P < 0.01$ ** $P < 0.01$ *** $P < 0.001$ n = 9 rats/group.

Table.7 Effect of flaxseed oil (FSO) and vitamin E (Vit. E) on the activity of antioxidant enzymes in liver tissues of rats with oxidative stress.

Groups Parameters	Control (-ve)	Control (+ve)	FSO (500 mg/kg)	Vit. E (50 mg/kg)	FSO + Vit. E
GPx (nmol/min/mg protein)	0.60 ± 0.02 ^a	0.31 ± 0.01 ^{d***}	0.42 ± 0.003 ^{b*}	0.41 ± 0.002 ^{b**}	0.53 ± 0.001 ^{c**}
SOD (U/mg protein)	54.66 ± 1.25 ^a	31.83 ± 1.25 ^{d***}	44.42 ± 2.71 ^{b**}	42.22 ± 2.66 ^{b**}	51.42 ± 1.28 ^{c***}
CAT (nmol/min/mg protein)	0.185 ± 0.003 ^a	0.143 ± 0.002 ^{d***}	0.158 ± 0.001 ^{b**}	0.163 ± 0.002 ^{b**}	0.170 ± 0.001 ^{c***}

Mean ± SD values in each raw with different superscripts (a, b, c, d) are significant when compared to the control groups at * $P < 0.01$ ** $P < 0.01$ *** $P < 0.001$ n = 9 rats/group.

with vitamin E (Vit.E) against oxidative stress induced by potassium bromate (KBrO₃) in male rats.

Oxidative stress induced in rats in this study by KBrO₃ was manifested by reduction in body weight gain and feed efficiency, increase in lipid peroxidation, rise in total cholesterol (TC) and triglycerides (TG) as well as elevation in serum and tissue biochemical markers of hepatorenal function. These findings were partially similar to those obtained by Abd El-Ghany *et al.* (2011) and Ahmed and Mahmood (2012) who reported that the serum bilirubin level and the activity of AST and ALT enzyme and levels of TC and TG were increased, while the total proteins were significantly decreased in rats with hepatotoxicity due to oxidative stress. However, oral administration of bromobenzene induced hepatic oxidative stress manifested by significant elevation of activities of AST and ALT in male rats. These findings indicated impaired function and damage of liver cells by bromobenzene (El-Sharaky *et al.*, 2009).

Lipid peroxidation is a complex process that damages the cell structure and function. Peroxidation of lipids of cell membrane initiates a loss of membrane integrity; membrane bound enzyme activity and causes cell lyses. However, the decreased activity of tissue antioxidant enzymes is likely to cause tissue damage by lipid peroxides or protein carbonyls (Pryor and Squadrito, 1995). In the present study, the increased lipid peroxidation due to oxidative stress induced by potassium bromate was previously reported by Kurokawa *et al.* (1990), Khan and Sultana (2004), Abd El-Ghany *et al.* (2011) and Ahmed and Mahmood (2012).

Results of the current study revealed that oral administration of flaxseed oil (FSO)

to rats with experimental oxidative stress improved hepatorenal function, lowered total cholesterol and triglycerides, reduced lipid peroxidation and produced good antioxidant activity.

The hepatoprotective action of flaxseed oil reported herein was similar to that obtained by Naqshbandi *et al.* (2012) who reported that flaxseed oil ameliorated cisplatin- induced hepatotoxicity and other deleterious effects due to its intrinsic biochemical antioxidant properties. The nephroprotective activity of flaxseed oil demonstrated in this study agreed with that reported by Abd El-Ghany *et al.* (2011) and Khan *et al.* (2012). The hepatorenal protective activity of flaxseed oil could be attributed to its antioxidant effect. The lipid lowering effect and reduction of tissue lipid peroxide (MDA) of flaxseed oil were similar to those reported by Prasad, (2000), Lee and Prasad (2003) and Khan *et al.* (2012).

The antioxidant effect of FSO was in accord with that reported by Lee and Prasad (2003), Prasad (2009), Abdel Moneim *et al.* (2011) and Khan *et al.* (2012). The later author concluded that FSO is effective in reducing nephrotoxicity and oxidative damage induced by nitroprusside in rats. The antioxidant activity of FSO could be attributed to its high content of omega-3 polyunsaturated fatty acid (PUFA) as reported by Khan *et al.* (2012) who concluded that the vegetarians who cannot consume fish oil can have similar health benefits from flaxseed oil which contains omega-3 PUFA. However, Lee and Prasad, (2003) concluded that flaxseed oil (Omega Nutrition) suppresses oxygen radical production by white blood cells and improves cardiovascular health due to its powerful antioxidant activity. It was

found that dietary flaxseed may have significant health-related benefits due to its high content of the omega-3 fatty acid, alpha-linolenic acid (ALA) as mentioned by Austria *et al.* (2008).

The current study revealed that flaxseed oil significantly increased the activity of antioxidant enzyme glutathione peroxidase, superoxide dismutase and catalase in liver tissue of rats with oxidative stress. This effect of flaxseed oil was similar to that reported by Abdel Moneim *et al.* (2011) and Khan *et al.* (2012).

The present results also showed that the antioxidant effect of flaxseed oil was amplified by its coadministration with vitamin E. It is well known that vitamin E (alpha-tocopherol) is a powerful antioxidant. Previous studies showed that intake of vitamin E could normalize the damaging effect of oxidative stress induced by free radicals (Shalaby *et al.*, 2004; Ramachandran and Raja, 2010 and Iranloye and Oludare, 2011). Coadministration of flaxseed oil and vitamin E ameliorated potassium bromate-induced hepatic oxidative stress as evident by decreasing the levels of MDA, increasing hepatic reduced glutathione content and enhancing the activity of liver antioxidative enzymes (GPx, SOD and CAT). The potentiation of antioxidant activity of flaxseed oil by coadministration of vitamin E could be due to the addition effect.

In conclusion, the results suggest that oral administration of flaxseed oil to rats with oxidative stress improves hepatorenal function, normalizes serum total cholesterol and triglycerides, reduces tissue lipid peroxidation and enhances the activity of tissue antioxidant enzymes.

These effects were amplified by coadministration of flaxseed oil with vitamin E. Therefore, intake of flaxseed and vitamin E produces high antioxidant activity and may be beneficial for reducing the oxidative stress.

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