

Antioxidant Activities of Marjoram (*Origanum majoranum* L.) Added to Frozen Beef Kofta and its Therapeutic Effect Against Kidney Damage in Rats

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Abstract: Besides certain medicinal effects aromatic plants prolonged the storage life of food by their antioxidant properties. Marjoram powder 5% and 0.5% oil were used during preparation of beef kofta. Beef kofta meat stored at (-18°C) for 6 months was analyzed for peroxide value (PV), P-anisidine value (AV), antioxidant effectiveness (AE), oxidation rate ratio (ORR), thiobarbituric acid (TBA) and 1,1 diphenyl-2-picrylhydrazyl (DPPH) radical. The results showed a significant decrease in (PV), P-anisidine value and (TBA) in beef kofta meat treated with different marjoram concentration of during storage at (-18°C) for 6 months compared with control. Therefore, the results of this study showed that marjoram to maintain quality the of frozen beef kofta during storage and can be marjoram proposed to therapeutic the kidney against (KBrO₃)-induced kidney damage in rats. The therapeutic effect of Marjoram powder and its oil were investigated to therapeutic against (KBrO₃) induced kidney damage in rats. This study was conducted on twenty five albino male rats and classified into five groups (n=5). The first group kept as normal group (control -ve), the second group (n=20) received (KBrO₃) through intragastric (20mg/kg B.W.) twice/week until the end experiment period. Then it classified into four groups as: untreated group (control positive) and three treated groups (ALA group, 5% powder of marjoram group and 0.5ml oil marjoram group). The results revealed that the rats treated with marjoram powder and oil showed significant decreases in serum ALT, AST, urea, creatinine, uric acid and bilirubin levels, cholesterol, triglycerides (TG), (LDL-C) and (VLDL-C). While, there was an increase in (HDL-C) compared to untreated group (+ve).

Key words: Antioxidant • ALA • DPPH • KBrO₃ • Marjoram • Meat products

INTRODUCTION

Not only the lipid oxidation can cause a loss in nutritional value but there are another too factors such as free radicals which led to various undesirable, chemical reactions. Also, oxidation can cause other degradation effects such as discoloration, vitamins destruction and polymerization [1]. Some modern cultures still consume wild plants as a normal spice and herb source for obtaining fairly good amounts of several nutrients and it is widely accepted that herbs are good nutritional sources of minerals. Furthermore, other nutrients, such as carotenoids and phenols, are found of larger quantities in these plants [2, 3]. Marjoram is a member of the mint family Lamiaceae. Typically, products identified marjoram

as the dried leaves and flowering tops of *Origanum majorana* L. which is found throughout the world. It contains phenolics terpenoids (thymol, carvacrol), flavonoids (diosmetin, luteolin, apigenin), tannins, hydroquinone, phenolic glycosides (arbutin, methyl arbutin, vitexin, orientin, thymonin), triacontan, sitosterol, acids (oleanolic acid) and cis-sabinene hydrate [4]. Crude extracts of spices rich in phenolics are now the mean interest in the food industry because they retard oxidative degradation of lipids and thereby improve the quality and nutritional value of food [5-7].

Beef kefta is a food which is made by grinding meat and mixing it with an assortment of spices. The resulting seasoned meat can be shaped into meatballs or cylinders of meat which can be cooked in a wide variety of ways.

Variations on kefta are common street foods in the Middle East and they may also be served as appetizers or as a more central part of a meal. It is easy to make kefta at home and many cooks enjoy making this simple dish at home since it allows them to adjust the spices as desired [8]. The total chemical composition of the samples of kofta collected from local market was 56.7% moisture, 15.72% total protein, 19.75% total fat, 0.90% crude fiber, 4.98% ash, 1.95% carbohydrates [9]. Potassium bromate (KBrO₃) has been used widely for water disinfection. KBrO₃ has been reported to be a potent nephrotoxic agent which can mediate kidney damage, toxicity and tumor response in rats, it also increase kidney lipid peroxidation and hydrogen peroxide formation with reduction in kidney antioxidant enzymes [10, 11]. This study was carried out to investigate the organoleptic properties and stability of raw fat beef kofta meat during storage at -18°C for 6 months by adding marjoram at concentrations 5.0% powder and 0.5ml oil on and also study its Therapeutic effects against KBrO₃ induced kidney damage in rats.

MATERIALS AND METHODS

Materials:

Plants: Marjoram (*Origanum majoranum* L.) and its oil were obtained from the Agricultural Research Centre, Giza, Egypt.

Beef Kofta: Fresh beef meat was obtained from a butcher shop, onion, salt, spices were obtained from the local market at El-Mansoura city.

Potassium Bromate (KBrO₃): Potassium bromate[®] is a white powder odorless, purchased from El-Gomhoria Co. Cairo, Egypt.

Alpha Lipoic Acid: α-lipoic acid[®] is an organosulfur compound derived from octanoic acid, purchased from El-Gomhoria Company.

Rats: Twenty-five male albino rats of Sprague Dawley strain were purchased from Laboratory Animal Colonies, Helwan, Egypt. The average weight was 80 ± 10g [12].

Methods:

Biochemical Analysis:

Scavenging Effect on DPPH Radicals: The effect of marjoram oil on 1,1-diphenyl-1-picrylhydrazyl DPPH radical was studied, employing the modified method described earlier by Yamaguchi *et al.* [13]. Briefly, oils (5, 10, 15 and 20 ul) in methanol (1ml) were mixed with 4 ml of

0.004% methanolic solution of DPPH. The reaction mixture was shaken well and incubated for 30 min at room temperature and the absorbance of the resulting solution was read at 517 nm against a blank. The radical scavenging activity was measured as a decrease in the absorbance of DPPH and was calculated using the following equation:

$$\text{Scavenging effect \%} = \frac{\text{Ab. Control} - \text{Ab Sample (517nm)}}{\text{Ab Control (517nm)}} \times 100$$

Preparation of Kofta: Kofta blend was prepared using the formula given by Youssef *et al.* [14] as follows: Meat 90% was mixed into house mincer, flour 3%, fresh onion 3% were peeled and minced before the addition, 2% salt and 2% spices. These ingredients were mixed together, divided to three equal portions, the first portion was remained without any addition (control) and the two reminder other portions were individually mixed with marjoram at concentrations of 5.0% powder and 0.5ml oil to give the treatments. Finger shaped pieces were prepared to formulate kofta. All samples were freeze stored at -18±2°C up to 6 months.

Extraction of Fat from Prepared Kofta: One hundred gram of crushed frozen kofta after 2, 4 and 6 months were placed in a closed stopper flask (500ml) using petroleum ether. The flask was shaken for 30 min using horizontal shaker at room temperature and left for 24h. The homogenized mixture was filtered following method according to AOAC. [15].

Chemical Analysis: Peroxide and P-anisidine values were determined according to the AOAC [15]. Thiobarbituric acid value (TBA) was determined according to the method described by Sidwell [16]. The percentage antioxidant effectiveness (AE) was calculated according to method of Adegoke and Krishna [17]. The third method of expression based on the oxidation rate ratio (ORR) was calculated according to method of Marinova *et al.* [18].

Organoleptic Evaluation: Organoleptic properties of appearance, color, odor, juiciness and texture of prepared products was carried out by aid of 10 panelists according to Klein and Bardy [19], who recommended the following judging scale:

Very good	8-9 score
Good	6-7 score
Fair	4-5 score
Poor	2-3 score
Very Poor	0-1 score

Experimental Animal Design: The experimental rats were fed on basal diet for five days before starting the experiment for adaptation then the rats were allocated into five equal groups. Normal control group fed on the basal diet only while the other five groups were administered a single dose of freshly prepared KBrO_3 (20 mg/kg B.W) intragastric twice/week [20] all over period of the experiment, then rats classified into four groups which were control positive (+ve) (untreated) and treated groups that were Alpha lipoic acid (ALA) (10 mg/kg B.W daily) [20] and two treated groups with marjoram at concentration (5.0% powder from standard diet.) and (0.5 ml/kg B.W. oil by stomach tube). The study was assigned for eight weeks. The food intake was calculated daily and the body weight gain was recorded weekly. Food efficiency ratio (FER) was calculated according to Chapman *et al.* [21].

The Analytical Methods of Blood Serum: At the end of the experiment, the rats were sacrificed to obtain blood samples. Heparinized blood was analyzed for estimation of total cholesterol (TC), triglyceride (TG) and high density lipoprotein cholesterol (HDLc) in serum which were determined according to the methods of Richmond [22], Fassati and Principe [23] and Gordon [24], respectively. Low density lipoprotein cholesterol (LDLc) and very low density lipoprotein cholesterol (VLDLc) were calculated using the method of Hatch and Lees [25] and Friedewald *et al.* [26], respectively. Atherogenic indices were obtained by dividing TC/HDLc or dividing LDLc/HDLc according to Castelli and Levitar [27] Bilirubin, serum alanine aminotransferase (ALT) and aspartate aminotransferase enzymes (AST) were determined according to the method described by Reitman and Frankel [28] ALP was determined according to Bergmeyer and Horder [29]. Serum creatinine was determined according to the method described by Bartles *et al.* [30], respectively. Serum uric acid was estimated by an enzymatic method according to Trinder [31].

Statistical Analysis: The obtained data were statistically analyzed using computerized SPSS (Statistic Program Sigmastat, Statistical Soft-Ware, SAS Institute, Cary, NC). Effects of different treatments were analyzed by one way ANOVA (Analysis of variance) test using Duncan's multiple range test and $p < 0.05$ was used to indicate significance between different groups as described by Snedecor and Cochran [32].

RESULTS AND DISCUSSION

Scavenging Effect on 1,1-diphenyl-2-picrylhydrazyl Radical (DPPH) (%) of Marjoram Oil: The 1,1-diphenyl-2-picrylhydrazyl radical (DPPH) is a stable radical with a maximum absorbance at 517 nm that can readily undergo reduction by an antioxidant. Because of the ease and convenience of this reaction, it now has widespread use in the free radical scavenging activity assessment Brand Williams [33]. In addition at concentration of 5 $\mu\text{g/ml}$, the results in Table 1 revealed that the marjoram oil effect was (62.4%). Marjoram oil effect addition at concentrations 10, 15 and 20 $\mu\text{g/ml}$ were 80.7, 83.3 and 93.2, respectively.

Effect of Treating by Marjoram of Different Concentrations on the Organoleptic Properties of Cooked Treated Beef Kofta: Data presented in Table 2 showed the effect of treating by marjoram of different concentrations on the organoleptic properties (appearance, color, odor, juiciness and texture) of cooked treated beef kofta meat during storage at -18°C for 6 months. It could be concluded that no obvious differences obtained between kofta meat which treated by marjoram (5% powder and 0.5% marjoram oil) during storage at -18°C for 6 months. Samples (5% marjoram powder and 0.5% marjoram oil) were of more score than control during storage at -18°C for 6 months. Changes in eating habits arising from the development of society in recent decades have led people to search for affordable and healthier foods with satisfactory taste and pleasant appearance. Thus, the food industry continually seeks to adapt and develop new formulations designed to increase shelf life and to improve quality and food safety as reported by Naif [34].

Inhibitory Effect of Marjoram Addition on Peroxide Value of Lipids Extracted from Frozen Beef Kofta: The peroxide value is known as indicator for the extent of forming the hydroperoxides which are consider as the primary products of lipid oxidation in food stuffs. The oxidation of lipid during frozen-storage leads to the development of rancidity and formation the undesirable off-flavor in food. Therefore, it seems reasonable to determine the concentration of hydroperoxides as a measure of the oxidation extent of lipids. Data present in Table 3 show the peroxide value changes in lipids extracted from frozen beef kofta. Peroxide value is a widely used measure of the primary lipid oxidation, indicating the amount of peroxides

Table 1: Scavenging effect (%) of marjoram oil on (DPPH) radical.

Marjoram oil level	5 (µg/ml)	10(µg/ml)	15(µg/ml)	20(µg/ml)
Scave	62.4±1.6	80.7±1.7	83.3±2.2	93.2±1.5*

Mean values ± standard deviation

Table 2: Effect of treating by marjoram powder and oil of different concentrations on the organoleptic properties of treated beef kofta meat during storage at -18°C for 6 months.

Time of storage (months)	Samples	Organoleptic properties					
		Appearance 20*	Color 20	Odor 20	Juiciness 20	Texture 20	Total score 100
0	Control	18.6±1.2	18.6±0.4	18.6±0.4	16.6±0.4	18.6±0.4	91
	5%marjoram powder	18.6±0.4	17.6±0.4	18.6±0.4	17.0±0.8	18.6±0.4	90.4
	0.5% marjoram oil	19.0±0.8	18.3±0.4	18.6±0.4	16.6±1.2	18.0±0.8	90.5
2	Control	17.6±0.4	17.0±0.8	17.6±1.2	16.0±0.8	18.0±0.8	86.2
	5% marjoram powder	18.0±0.8	17.0±0.8	18.0±0.8	16.3±0.4	18.0±0.8	87.3
	0.5% marjoram oil	17.0±0.8	17.6±0.4	17.6±0.4	16.0±0.8	17.6±0.4	85.8
4	Control	17.0±0.8	16.6±0.4	18.0±0.8	15.6±0.9	17.6±0.4	84.8
	5% marjoram powder	17.0±0.8	16.6±1.2	18.0±0.8	15.6±0.9	17.0±2.1	84.2
	0.5% marjoram oil	17.3±0.4	17.0±0.8	18.6±1.2	15.0±0.8	17.0±0.8	84.9
6	Control	16.0±0.8	16.3±0.4	17.0±0.8	15.0±0.8	16.0±0.8	80.3
	5% marjoram powder	16.0±0.8	16.6±1.2	17.6±0.4	15.6±0.9	16.0±0.8	81.8
	0.5% marjoram oil	16.6±1.2	16.6±0.4	17.3±0.4	15.6±0.9	16.6±1.2	82.7

Control= sample was prepared without *marjoram*, * = Max. score, ± = Means standard deviation.

Table 3: Inhibitory effect of marjoram powder and oil addition on peroxide value (meq. /kg fat) of lipids extracted from frozen beef kofta during storage at -18°C for 4 months.

Samples	Time of storage (months)			
	Zero time	2	4	6
Control	2.3±0.36	4.7±0.3 a	5.4±0.55 a	6.6±0.6 a
5.0% Marjoram powder	2.3±0.36	3.6±0.4 b	4.9±0.15 b	5.1±0.3 b
0.5% Marjoram oil	2.3±0.36	3.4±0.33 b	4.53±0.25 b	4.9±0.4 c

Control= sample was prepared without marjoram

Mean values in each column having different superscript (a, b, c, d) are significantly different

Means with the same letter are insignificantly different.

formed in the fats during oxidation. Fat oxidation was measured after 2, 4, 6 months of storage. The results revealed that marjoram reduced peroxide value significantly after 2 months of storage. The maximum reduction which reached 4.9 after 6 months was noticed in the sample treated with 0.5% of marjoram oil followed by 5.0% powder of marjoram which was 5.1 compared to control sample which was 6.6. Peroxide value reflecting fat oxidation of beef kofta meat progressively increased during frozen-storage and at any giving time of storage, control samples had higher (PV) than the beef kofta treated with different concentrations of marjoram (Table 3). These results are in agreement with those obtained by Soumia *et al.* [35].

Data in Table 4 illustrate the percentage of antioxidant effectiveness (AE) and oxidation rate ratio (ORR) of fat beef kofta treated with different concentrations of marjoram during storage at -18°C for 6 months. Data showed that antioxidant effectiveness (AE)

decrease for (25.7 to 16.46) during storage from 2-6 months in raw beef kofta meat while oxidation rate ratio (ORR) increased from (0.74 to 0.83) for treating by 5.0% marjoram powder. On the other hand (AE) were decrease from 30.30 to 22.56, while (ORR) were increase from (0.70 to 0.77) for treating by 0.5% marjoram oil during storage respectively. It could be noticed that beef kofta meat treating by marjoram at 5.0% powder or 0.5% oil concentrations were more effective as natural antioxidant.

Inhibitory Effect of Marjoram Addition on the Formation of 2-alkenes of Lipids Extracted from Frozen Beef Kofta Measured by P-anisidine (Av) Method: P-anisidine value is extensively used to measure the secondary oxidation products mainly non- volatile carbonyls, which are formed during lipid oxidative degradation-P-anisidine values reflect the inhibitory effect of the marjoram under study on the formation of 2-Alkenes in frozen kofta was recorded in Table 5. The results revealed that alkenes

Table 4: Percentage of antioxidant effectiveness (AE) and oxidation rate ratio (ORR) of fat beef kofta treated with different concentrations of marjoram in beef kofta meat during storage at -18°C for 6 months.

Samples	Time of storage (months)					
	2		4		6	
	AE	ORR	AE	ORR	AE	ORR
5.0% Marjoram powder	25.75	0.74	22.80	0.77	16.46	0.83
0.5% Marjoram oil	30.30	0.70	29.56	0.70	22.56	0.77

AE= antioxidant effectiveness and ORR= oxidation rate ratio.

Table 5: Inhibitory effect of marjoram powder and oil addition on the formation of 2-Alkenes of lipids extracted from frozen beef kofta measured by P-anisidine method.

Time of storage (months)	Zero time	2	4	6
Control	11.3±0.36	18.8±0.55 a	26.3±0.83 a	31.4±0.76 a
5.0% Marjoram powder	11.3±0.36	15.3±0.56 b	23.6±0.51 b	26.1±0.98 b
0.5% Marjoram oil	11.3±0.36	14.0±0.35 b	22.0±0.55 c	23.9±0.75 c

Mean values in each column having different superscript (a, b, c, d) are significantly different.

Means with the same letter are insignificantly different.

formation increased by increasing storage time in control and all concentration (5.0% powder and 0.5% oil) of marjoram samples. While, the increase of P-anisidine value was lower in the samples treated with different concentration of marjoram comparing to control. P-anisidine was determined after 2, 4, 6 months, the effects of samples under study on alkenes formation in frozen kofta was carried out. After 2 months, the P-anisidine value of marjoram oil 0.5% group was 14.0 while it was 15.3 in marjoram powder 5.0% group. While, after 6 months, the P-anisidine value of marjoram oil 0.5% group was 23.9 while it was 26.1 in marjoram powder 5.0% group. Food industries use synthetic additives with antioxidant properties. However, due to reports of possible toxic effects from synthetic antioxidants and to increasingly demanding consumer preferences for natural products and health benefits, the interest for alternative methods to retard lipid oxidation in foods, such as the use of natural antioxidants, has increased. These methods include spices and herbs which have many phytochemicals which are potential sources of natural antioxidants, e.g. phenolic diterpenes, flavonoids, tannins and phenolic acids as reported by Dawidowicz [36].

Inhibitory Effect of Marjoram Addition on Malonaldehyde Formation of Lipids Extracted from Frozen Beef Kofta Measured by Using Tba Value Method: Malonaldehyde formation was determined after 2, 4, 6 months, the effects of marjoram addition on malonaldehyde formation for frozen beef kofta results are shown in Table 6. After 2 months the reductions in TBA values caused by marjoram

addition as compared to control, the highest reduction (0.36) was caused by marjoram oil addition at concentration 0.5% followed by marjoram powder which was 0.40. After 6 months, the reduction in the malonaldehyde formation were significant as comparing to control sample which increased to 1.29 with storage time, the highest reduction (0.74) was caused by marjoram oil addition at concentration 0.5%, followed by marjoram powder addition at concentration 5.0% (0.77). Thiobarbituric acid (TBA) test is used as an index for measuring oxidative rancidity (malonaldehyde formation). The TBA test is a sensitive test for the decomposition products of highly unsaturated fatty acids which do not appear in peroxide value determination [37]. These results were also in agreement with those obtained by Zheng and Wang [38], who noticed that marjoram showed high antioxidant effect for reduction T.B.A values in samples, the effectiveness of decreasment followed the sequence: Thyme > sage > rosemary > marjoram > black seeds.

Effect of Marjoram Treatment on Body Weight Gain, Food Intake and Food Efficiency Ratio in Kbro₃ Induced Kidney Damage in Rats: As shown in Table 7, body weight gain, food intake and food efficiency ratio (FER) showed significant decreases in KBrO₃ group (untreated) compared to normal control group. On the other hand, the treatment with ALA group at three levels were increased significantly body weight gain, food intake and food efficiency ratio the groups treated with 5.0% powder marjoram and 0.5% oil marjoram compared to positive control group. These results were in agreement with those

Table 6: Inhibitory effect of marjoram powder and oil addition on malonaldehyde formation of lipids extracted from frozen beef kofta measured by using TBA value method.

Time of storage (months)	Zero time	2	4	6
Control	0.38±0.01	0.53±0.07 a	0.78±0.06 a	1.29±0.30 a
5.0% Marjoram powder	0.38±0.01	0.40±0.04 b	0.61±0.03 b	0.77±0.03 b
0.5% Marjoram oil	0.38±0.01	0.36±0.04 c	0.52±0.02 c	0.74±0.04 b

Mean values in each column having different superscript (a, b, c, d) are significantly different.

Means with the same letter are insignificantly different.

Table 7: Effect of marjoram treatment on body weight gain, food intake and food efficiency ratio in KBrO₃ induced kidney damage in rats.

Groups	Parameters					
	Initial weight	Final weight	Weight gain (g/day)	%	Food intake (g/day)	FER
- control	86.4±5.41a	152.6±12.32a	66.2±7.08a	76.42±6.07a	15.36±0.78a	0.077±0.51a
+ control	86.2±7.5a	130.1±10.6b	43.8±8.73b	51.14±9.99b	13.48±1.16b	0.058 ±0.45b
ALA	82.3±7.98a	147.2±10.61a	65.0±3.39a	79.42±6.11a	15.0±0.94a	0.077±0.18a
5.0% Marjoram powder	84.2±7.19a	149.2±8.07a	64.8±3.37a	77.56±7.12a	15.08±0.72a	0.77±0.23a
0.5% Marjoram oil	82.8±9.04a	146.8±11.71a	64.0±6.44a	77.92±1.06a	14.96±0.83a	0.076±0.33a

Mean values in each column having different superscript (a, b, c, d) are significantly different.

Means with the same letter are insignificantly different.

Table 8: Effect of marjoram treatment on lipid profile in KBrO₃ induced kidney damage in rats.

Groups	Parameters				
	TC (mg/dl)	TG (mg/dl)	HDL-c (mg/dl)	LDL-c (mg/dl)	VLDL-c (mg/dl)
- control	150.4±36.16d	83.01±12.72c	43.31±7.47a	90.49±5.31c	16.6±2.54b
+ control	285.20±40.1a	265.8±76.92a	31.21±4.54b	200.83±23.39a	53.16±15.38a
ALA	180.01±31.62cd	91.8±24.18c	39.8±5.17a	121.85±12.54ab	18.36±4.84b
5.0% Marjoram powder	261.02±43.92ab	150.41±37.09 b	42.2±4.91a	188.74±15.07b	28.12±9.86b
0.5% Marjoram oil	220.0±28.43bc	128.21±22.97bc	44.6±5.17a	149.76±15.13bc	25.64±4.59b

Mean values in each column having different superscript (a, b, c, d) are significantly different.

Means with the same letter are insignificantly different. TC: Total cholesterol TG: Triglycerides. HDL: High density lipoprotein cholesterol LDL: Low density lipoprotein cholesterol. VLDLc: Very low density lipoprotein cholesterol

obtained by Oyewo *et al.* [39], who reported that the intake of potassium bromate caused some characteristic physical changes in adult wistar rat as evident in the reduction of physical activity and feebleness.

Effect of Marjoram Treatment on Lipid Profile in KBrO₃ Induced Kidney Damage in Rats: Administration of (KBrO₃) for positive control group (untreated) resulted in a significant increase in serum total cholesterol, triglyceride and LDL-cholesterol levels as shown in Table 8 compared to normal group (control -ve). Table 8 showed the effect of marjoram supplement at two levels (5.0% powder, 0.5 % oil /kg diet) and ALA groups on serum lipid profile in KBrO₃ induced kidney damage. The kidney damage induced by KBrO₃ caused a significant rise in total cholesterol, triglycerides, LDL-C and VLDL-C compared to positive control group (untreated). While, there was a significant decrease in HDL-C compared to negative control group (untreated). These results were agreed with Soltan and Abdel Wahab [40], who found

that the turmeric, mustard, marjoram, rosemary, ginger and red pepper powders have hypocholesterolaemic, hypolipidemic and antioxidant effects properties when studied in rats, a significant decrease in serum total cholesterol, triglyceride, LDL-cholesterol levels and VLDL cholesterol occurred compared to hypercholesterolaemic group. The hypolipidaemic activity of marjoram in rats could be attributed to the presence of valuable polyphenolic compounds, terpenoids, flavonoids, tannins, hydroquinone, phenolic glycosides and sabinene.

In Table 9 the treatment with marjoram at two levels (5.0% power, 0.5 % kg diet) and ALA group showed a significant decreased deterioration in atherogenic indexes (TC/HDLc and LDLc/HDLc) due to decrease HDLc and increased of LDLc and TC compared to positive control group (untreated) than those of control group. These results are in parallel with those obtained by Faleiro *et al.* [41], who noticed that the essential oil obtained from marjoram has aromatic smell and contain a high

Table 9: Effect of marjoram treatment on atherogenic indexes in KBrO₃ induced kidney damage in rats.

Groups	Parameters	
	TC/LDL	LDL/HDL
- control	3.47±1.24 c	2.09±0.39 cd
+ control	9.14±2.44 a	6.44±0.69 a
ALA	4.52±1.05 bc	3.06±0.54b
5.0% Marjoram powder	6.19±1.24b	4.47±0.65Bc
0.5% Marjoram oil	4.93±0.79bc	3.36±0.45cd

Mean values in each column having different superscript (a, b, c, d) are significantly different.

Means with the same letter are insignificantly different.

Table 10: Effect of marjoram treatment on liver and kidneys function in KBrO₃ induced kidney damage in rats.

Groups	Parameters					
	ALT (μ/ml)	AST(μ/ml)	ALP (μ/ml)	Creatinine (mg/dl)	Uric acid (mg/dl)	Bilirubin (mg/dl)
- control	30.60±3.51c	26.03±5.57d	2.52±0.37bc	0.43±0.16c	2.48±0.31b	0.48±0.07b
+ control	49.07±6.82a	40.00±3.32a	3.66±0.53a	2.14±0.56a	3.94±0.56a	0.77±0.24a
ALA	34.41±4.93bc	29.02±4.30cd	2.80±0.25b	0.72±0.26bc	2.92±0.43b	0.49±0.04b
5.0% Marjoram powder	39.83±6.46b	36.61±3.51ab	2.74±0.24bc	0.92±0.22b	3.04±0.40b	0.62±0.03b
0.5% Marjoram oil	38.64±6.02 b	34.80±3.35ab	2.42±0.33bc	0.83±0.14bc	3.04±0.41b	0.62±0.05b

Mean values in each column having different superscript (a, b, c, d) are significantly different.

Means with the same letter are insignificantly different.

ALT: alanine aminotransferase enzymes. AST: aspartate aminotransferase enzymes ALP: alkaline phosphatase

percentage of polyphenols and monoterpenes which have antioxidant properties. Amarowicz *et al.* [42] found that marjoram ethanolic extract contain considerable amounts of total phenolics compounds and have antioxidant activity and free radical-scavenging capacity. The hypocholesterolemic effect of marjoram could be attributed to presence of isoflavones which prevent intestinal absorption of cholesterol by competition for its absorption sites [43]. These results are also in accordance with Nagm [44], who found that marjoram extract lead to significantly lowering in TG ($p<0.01$) than may be due to lower fatty acids synthase.

Effect of Marjoram Treatment on Alt, Ast, Alp, Creatinine, Uric Acid and Bilirubin in KbrO₃ Induced Kidney Damage in Rats: Data in Table 10 showed that liver and kidney function tests values which were elevated by KBrO₃ in positive control group (untreated) administration. The level of serum aspartate and alanine amino transferase (AST&ALT) enzymes, alkaline phoshatase (AP) creatinine and uric acid were found to be significantly lowered in ALA, marjoram powder and marjoram oil groups compared to positive control group(untreated). Therefore, it is possible to suggest that powder and oil are safe and may therapeutic against hepato cellular damage as evidenced by normal control group (-ve) in serum levels of AST and ALT in treated groups. The positive control group showed a significant increase in serum aspartate and alanine amino transferase

(AST&ALT) enzymes, alkaline phoshatase (AP), creatinine and uric acid compared to normal control group. The present results were also supported by Kanjil *et al.* [45] histopathological investigation where potassium bromate caused congestion of the central vein with blood cells in the hepatocytes, infiltration of the interstitial cells accompanied with acute nephritis in the nephrons and mild mucosal dysfunction in the small intestine. On the other hand, it was found the significant decrease in serum uric acid and bilirubin by marjoram treatment (5.0% power, 0.5 % oil kg diet) and ALA groups compared to positive control group (untreated). These results were in agreement with those reported by of Rodriguez *et al.* [46] who found that volatile oil and extract of marjoram reduce in serum uric acid bilirubin this might be due to its isoflavones, polyphenols and other antioxidants. El Ashmawy *et al.* [47] found that of administration of marjoram induced a significant decrease in serum activities of transaminases (ALT, AST), ALP, urea and creatinine in comparison with lead acetate treated in mice.

CONCLUSION

Based on the evidence available it appeared Marjoram (*Origanum majoranum* L.) was beneficial to organoleptic properties and stability of raw fat beef kofta meat during storage at -18°C for 6 months. This herb is beneficial to renal function and eliminated oxidative stress by virtue of its antioxidant properties.

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