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Effect of *Moringa oleifera* leaves powder as antioxidant on quality of meat-rice Kofta during frozen storage

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ABSTRACT

Meat-rice kofta is the famous, delicious and common local dish in Egypt. Meat, rice powder and green herbs are the main components of meat-rice kofta. So, dark green color of moringa leaves powder had no effect in appearance, color and acceptability of samples. Different levels of moringa leaf powder (MLP) were used to process meat-rice kofta. Chemical composition (moisture, fat, protein and minerals content) were determined at zero time. Antioxidant activity, physical properties (cooking loss, color and shearing force) and sensory evaluation of samples were studied during 90 days storage at –18 °C. Protein and fat content of meat-rice kofta at zero time ranged from (18.01 to 19.52 %) and (8.22 to 9.62 %), respectively. Also, results show that MLP exhibited high phenolic content (55 mg/gm Gallic acid equivalent per gm). Good antioxidant activity as determined by radical scavenging activity of 1,1-diphenyl-2-picrylhydrazyl (DPPH), it was 96.038 % at 200 µg/ml concentration. Thiobarbituric acid decreased very fast after 45 days storage, then increased after 90 days of storage but still lower than zero time. Peroxide value generally decreased during different storage periods.The results indicated that the MLP can be used as natural antioxidant to extend shelf life of meat-rice product without loss in quality and nutritional value.

Key words: *Maringa oleifera*, antioxidant activity, physical properties, sensory evaluation, DPPH

Introduction

Considering the need of expanding shelf life of meat products isa point of view for its export potentiality, varied items of antioxidant, antibacterial activity and preservatives have been tested with different degrees of success (Suchandra *et al.,* 2012).

Nowadays, meat products represent a large percentage of foods consumed to be easily attainable and inexpensive comparing to fresh meat cut (de Oliveiraa *et al.,* 2012). During processing most of ground or minced beef are exposed to lipid and pigment oxidation. So, the use of natural antioxidants from plant extract to preserve meat products has attracted the consumer's attention due to their potential health advantage, safety (Jung *et al.,* 2010) and ability to prolong shelf life (Falowo *et al.,* 2017).

Maringa oleifera has very high nutritional properties such as, food supplement, potentiality to be a source of new drugs, the micro-nutrient content in dried leaves is more ten times vitamin A of carrots, 17 times calcium of milk, 15 times potassium of bananas, and 25 times iron of spinach (Singh *et al.,* 2010 and Manzoor *et al.,* 2007).

Moringa oleifera is the most widely cultivated specie of the genus Moringa (Fuglie, 2001). It is not only a good source of naturally antioxidant (Dillard and German, 2000), but also the leaves have important medicinal properties which contains a variety of bioactive substances, such as ascorbic acid, carotenoids and phenolic compounds (Siddhuraju and Becker, 2003), and have been assessed as natural preservative for various meat products (Al-Juhaimi *et al.,* 2016 and Falowo *et al.,* 2016). *Moringa oleifera* leaves have been used successfully in its dried state or powdered for preservation, packaging, transportation (Alakali *et al.,* 2015).

Generally, leaves dried under shade is the way to preserve nutrient content (Olushola, 2006), which retained about 50 – 70 % of vitamin A.

Therefore, shade dry is recommended and should be completely dry within a maximum of four days, then stored for up to 6 months in protected clean containers. The aim of this work is to

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investigate the utilization of moringa leaf powder as a source of natural antioxidant in reducing peroxidation of fried meat-rice kofta during freezing storage.

Materials and Methods

Materials:

Fresh boneless meat, rice, black pepper, onion, garlic, fresh herbs and sunflower oil were burchased from local market of Giza, Egypt.
Folin-Cicalteus, thiobarbituric acid and 2,2-diphenyl-1-picryhydrazyl (DPPH) were purchased from Sigma Aldrich Chemical Co. (St. Louis, MO, USA) and Merck (Darmstadt, Germany). All other chemicals were of analytical grade. Moringa leaves were cleaned, washed and dried at room temperature until reached 7.5 % moisture content; leaves were grinded and sieved to obtain moringa leaf powder (MLP).

Methods:

Kofta formulation:

Beef meat was minced and divided to four groups; each group was thoroughly mixed by hand with other ingredients as in Table (1). Parsley leaves were substituted by MLP at levels 10, 30 and 50 % (samples 1, 2 and 3, respectively). Control sample didn't contain MLP. Samples were formed in similar finger shape (10 g) then fried in sunflower oil and turned upside down to avoid color difference. When samples reached room temperature, they were packaged in polyethylene pouches and stored at –18 °C until analyzed.

Table 1: Formula of meat-rice kofta samples with MLP

Ingredients	Samples (g/1000 g)			
	Control	(1)	(2)	(3)
Mined meat	600	600	600	600
Rice flour	200	200	200	200
Water	100	100	100	100
Black pepper	5	5	5	5
Onion	30	30	30	30
Garlic	20	20	20	20
Salt	10	10	10	10
Cumin	10	10	10	10
Green coriander	5	5	5	5
Green dill	10	10	10	10
Green parsley	10	9	7	5
*MLP	--	1	3	5

**MLP: Moringa Leaves Powder*

Analysis of samples at zero time:

Proximate analysis of meat-rice kofta samples:

Moisture, protein and fat were analyzed according to A.O.A.C. method (A.O.A.C. 2010).

Antioxdant properties of moringa leaves powder (MLP):

Total phenolic compounds were determined by the Folin-Cicalteus (Escarpa and Gonzalez, 2001), radical scavenging activity DPPH determined by (Aromatic *et al.,* 2013).

Minerals Content:

The minerals content were determined using the atomic absorption spectrophotometer as described by A.O.A.C. (2005).

Cooking loss:

Cooking loss was calculated as follows by A.M.S.A., (1995).

$$\text{Cooking loss \%} = \frac{F - C}{F} \times 100$$

Where, F: fresh sample; C: cooked sample

Analysis of samples during storage:

Thiobarbituric acid (TBA) value:

Thiobarbituric acid value was estimated as described by Ohkawa, (1979).

Peroxide value (PV):

Peroxide value was determined by iodometric titration method as described by official standard method (A.O.A.C.1990).

Shear force value (Tenderness):

The Warner Bratzler Shear force apparatus Ametek / Mansfield & Green div. Largo, Florida was used to measure the tenderness of samples according to Herring (1976). Samples (at room temperature) were sheard for three times at different positions, and average of shear force was calculated in Newton (N).

Color:

The instrumental color of samples was analyzed using Hunter lab color (Model, CIE lab colour scale, Scan XE - Reston VA, USA). Color parameters: L* (lightness), a* (redness), and b* (yellowness) according to Hunter, (1958).

Sensory evaluation:

Sensory evaluation was done by 10 staff members in Food Technology Dept., National Research Centre; Egypt. Panelists evaluated samples according to Gelman and Benjamin (1989).

Statistical analysis:

The data obtained from sensory evaluation were analyzed statistically by analysis of variance (ANOVA) and least significant difference (L.S.D.) at 5 % level of significance as reported by Snedecor and Cochran, (1980).

Results and Discussion

Proximate analysis of meat-rice kofta samples:

The chemical composition of meat-rice kofta samples at zero time are presented in Table (2). Protein and fat content ranged between (18.01 to 19.52 %) and (8.22 to 9.71 %), respectively. This results showed that no difference between control and others samples. Similar results were obtained

by Suchandra *et al.,* (2012) who reported that moisture, total protein and fat contents did not differ in all samples.

Table 2: Chemical composition of kofta samples at zero time

Chemical composition	Control sample %	Sample (1) %	Sample (2) %	Sample (3) %
Moisture	45.93	44.75	43.65	41.16
Protein	18.01	18.82	18.97	19.52
Fat	9.62	9.59	9.71	8.22

Antioxidant properties of MLP:

Total phenolic content of MLP are 55 mg/gm, which polyphenols are accountable of antioxidant activity, high total phenolic content in MLP indicates its high antioxidant activity according to Das *et al.,* (2012) who found that the total phenolics of MLP were 48.36 mg/gm.
Radical scavenging activity (DPPH) has been used to evaluate the free radical scavenging ability of plant. In Figure (1), increasing the concentration of the MLP extract (µg/ml) increase the radical scavenging activity from 67.647 to 96.038 %, similar results were reported by Arun *et al.,* (2012).

The DPPH scavenging activity of MLP m

ay be attributed to its hydrogen donating ability as antioxidants, on interaction with DPPH, either transfer on electron or hydrogen atom to DPPH, thus neutralizing its free radical character (Naik *et al.,* 2003).

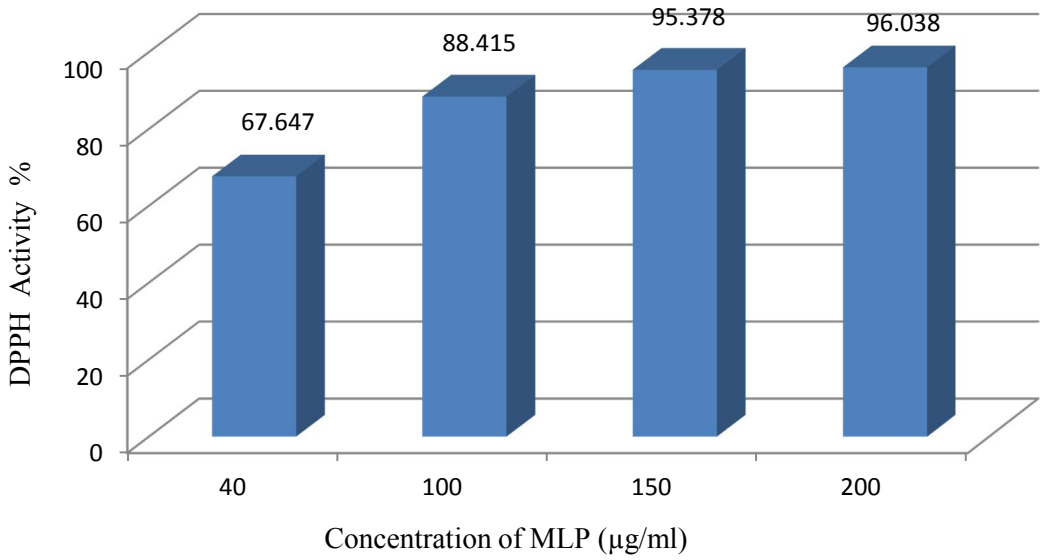

Fig. 1: DPPH activity of MLP

Minerals content of meat-rice kofta samples:

Table (3) presents the minerals composition of meat-rice kofta samples; calcium content of sample (3) was higher than the control sample 13.17, 7.52 mg/100gm, respectively. The percentage of increase was 75 %, as calcium builds healthy, strong bones and teeth and also assists blood clotting (Gordon, 1999).

Also, Table (3) reveals that sample (3) was higher in Zn, Mn, Cu, K than control sample. Iron of sample (3) was 13.71 mg/100 gm, which is more than 200 % in control sample.
In general, minerals content were above recommended daily allowance (Gamman and Sherrington, 1990).

Table 3: Minerals content of kofta samples at zero time

Samples	Minerals (mg/ 100 gm sample)					
	Fe	K	Cu	Mn	Zn	Ca
Control	3.51	409.80	0.14	0.14	2.96	7.52
Sample (1)	5.55	433.76	0.22	0.16	3.22	8.65
Sample (2)	9.63	481.81	0.38	0.20	3.74	10.91
Sample (3)	3.71	529.70	0.54	0.24	4.26	13.17

Cooking loss of meat-rice kofta samples:

As shown in Table (4), cooking loss percent of meat-rice kofta samples 1, 2 and 3 decreased slightly than control sample (24.57, 24.53, 24.46 and 25.60 %), respectively. Brugiapaglia and Destefanis, (2012) explained that cooking loss is a combination of liquid soluble matter lost during cooking and with increasing temperature (frying) water content decreases while, fat and protein contents increase indicating that the main part of cooking loss is water.

Table 4: Cooking loss of kofta samples at zero time

	Samples			
	Control	(1)	(2)	(3)
Cooking loss %	25.60	24.57	24.53	24.46

Thiobarbituric acid (TBA) and Peroxide values (PV) of kofta samples:

TBA and PV values of meat-rice kofta samples were determiend during storage. Lipid oxidation is the main non-microbial cause of quality deterioration in meat products during storage (Falowo *et al.,* 2014) it was monitored by measuring TBA nmol/g and PV (meq/kg), results are showen in Table (5). Samples 1, 2 and 3 had lower TBA values than control sample during 45 days storage. This may be due to polyphenols in Moringa leaf powder (MLP) which can play an important role in adsorbing and neutralizing free radicals, which lead to the inhibition of lipid peroxidation (O'Byrne *et al.,* 2002). With the increase in storage time (90 days), there was an increase in TBA values in the same samples but still lower than zero time. At the end of storage time (90 days) samples containing MLP showed delay in lipid oxidation compared to the control sample.

Table 5: Thiobarbituric acid (TBA) and peroxide value (PV) of kofta samples during storage.

Samples	Storage period	TBA (nmol/g)	PV (meq/kg)
Control	Zero time	6.04	4.36
	45 days	0.06	2.75
	90 days	6.39	0.14
Sample (1)	Zero time	5.53	3.63
	45 days	0.57	1.94
	90 days	4.35	0.13
Sample (2)	Zero time	6.73	2.53
	45 days	1.03	0.66
	90 days	6.01	0.02
Sample (3)	Zero time	6.82	0.95
	45 days	0.96	1.05
	90 days	6.13	0.06

Reduction of peroxide values during storage may be due to inhibition lipid peroxidation by MLP, which contains polyphenols that have antioxidant activity effects (Jayawardana *et al.,* 2015). Also, several reports revealed that substitution of MLP in meat product inhibited lipid oxidation (Al Juhami *et al.,* 2016; Das *et al.,* 2012; Muthukumar *et al.,* 2014; Shah, *et al.,* 2015).

Finally, phenols of MLP have antioxidant activity which could led to the observed peroxide value reduction of meatkofta during storage period.

Shear force (Tenderness) of kofta samples during storage periods:

Texture of meat is one the most important characteristics that affects consumer acceptability.(Aktas and Kaya, 2001). Besides, shear force is one of the methods which measures texture properties (tenderness).

Results in Table (6) showed that shear value of control sample at zero time had the lowest value at all investigated samples.

Extending storage time was accompanied by a gradual increase in shear force- values for all samples. Sample 3 had the highest value after 90 days storing (28.28N).

Results in Table (6) also illustrated that tenderness of sample (1) - containing 1gm MLP- had a lower values than samples (2) or (3) at all storage periods. Dreeling *et al.* (2000) found that shear force of cooked (grilled 12-16 min.) low-fat beefburger (5.2% fat) was 18 N.

Table 6: Shear force (N) values of kofta samples during storage periods.

Storage time (days)	Shear force (N) of samples			
	Control	Sample (1)	Sample (2)	Sample (3)
0	10.89	14.97	16.09	17.52
45	15.67	20.28	22.14	23.80
90	24.27	26.07	27.64	28.28

Color of meat-rice kofta samples:

Color of meat-rice kofta samples 1, 2 and 3 during storage were measured. Color is an important sensory attribute, which determines the products acceptability rate. The effects of substituted with MLP and storage time on Hunter lab color are presented in Table (7).

Table 7: Color of kofta samples during storage periods

Samples	Storage period	L^*	a^*	b^*
Control	Zero time	22.01	3.61	8.59
Sample (1)		22.98	3.28	8.74
Sample (2)		24.01	3.45	7.28
Sample (3)		24.72	2.14	6.02
Control	After 45 days	23.49	3.96	12.00
Sample (1)		24.79	4.11	13.45
Sample (2)		25.76	5.42	14.17
Sample (3)		25.97	3.36	14.08
Control	After 90 days	23.81	4.02	12.26
Sample (1)		24.40	4.50	13.22
Sample (2)		25.47	5.55	13.62
Sample (3)		25.85	3.35	12.90

Control samples had lower L^*(lightness) values compared to other samples owing to addition of MLP powder. Both control and treated samples, upon storage showed an increase in b^*(yellowness) values from 8.59 to 12.26 in control sample, 8.74 to 13.22 for sample (1), 7.28 to13.62 for sample (2) and 6.02 to 12.90for sample (3) after 90 days of storage .Increasing MLP lowers a*(redness) values as it were 3.28, 3.45and 2.14for samples 1,2and 3 respectively at zero time. Extending storage time had an increase in a*values for control sample from 3.61 to 3.96 and 4.02 after 45 and 90 days of storage respectively. Sample (3) had a* values lower than other samples at zero time,45 and 90 days of storage as it contained much more MLP than other samples.

Sensory evaluation:

Sensory evaluation of meat-rice kofta samples were done during different storage periods. Results in Table (8) showed that all samples had no significant differences in color, odor and appearance of fried meat-rice kofta after all storage periods. This may be due to more effective inhibition of lipid peroxidation (O'Byrne *et al.,* 2002). Also, mouth-feel scores seem no significant affect among substitution percentage or days of storage, the scores ranged from 6.91to 8.00.
After 45 and 90 days of frozen storage, sample (3) was sensorially acceptable and substitution of MLP did not have any negative effect on sensory attribute. Therefore, MLP at 50 % substitution has the potential to reduce oxidative rancidity and elongate shelf life of freezed fried meat-rice kofta.

Table 8: Sensory evaluation of kofta samples during storage periods.

Samples treatments	Taste	Oder	Color	Mouth-feel	Appearance	Overall acceptability
Zero time						
Control	7.73±1.27	7.55±1.21	7.18±0.98	7.18±0.98	7.36±1.12	7.45±0.52
Sample (1)	7.64±1.43	7.45±1.44	7.18±1.07	7.36±1.02	7.64±0.81	7.54±0.52
Sample (2)	7.64±1.21	7.64±1.36	7.72±1.19	7.36±1.36	7.73±1.19	8.09±1.04
Sample (3)	7.36±1.02	7.18±1.25	7.45±1.04	6.91±1.30	7.64±1.12	7.45±1.13
LSD	NS	NS	NS	NS	NS	NS
After 45 days						
Control	8.21±0.49	8.23±0.63	7.81±1.15	7.81±0.80	7.88±0.87	8.07±0.86
Sample (1)	8.21±1.03	8.00±1.00	7.77±1.01	7.38±1.19	7.73±0.73	7.61±0.76
Sample (2)	8.16±0.83	7.92±0.95	7.85±1.14	7.62±1.04	7.62±1.19	7.85±0.80
Sample (3)	7.71±1.17	7.50±1.00	7.96±0.87	7.73±1.05	8.11±0.63	7.76±0.95
LSD	NS	NS	NS	NS	NS	NS
After 90 days						
Control	7.91±1.22	8.18±0.98	8.18±0.60	7.91±1.04	7.73±0.78	7.91±0.94
Sample (1)	8.18±0.41	8.55±0.69	8.09±0.54	7.73±0.46	8.00±0.63	8.18±0.60
Sample (2)	8.18±0.98	8.27±1.00	8.09±0.94	7.82±1.17	8.09±0.83	7.73±1.10
Sample (3)	7.64±1.42	8.09±1.04	8.09±1.04	8.00±1.34	8.18±1.08	8.00±1.00
LSD	NS	NS	NS	NS	NS	NS

Conclusion

The present study has revealed that *Moringa oleifera* leaves has substantial amounts of phenolic compounds with free radical activity. The application of MLP at 5 g/kg meat-rice kofta can delay lipid oxidation during freezing storage.

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