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EFFECT OF MORINGA LEAVES (*Moringa oleifera* Lam.) EXTRACT ADDITION ON LUNCHEON MEAT QUALITY

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ABSTRACT: The effect of addition moringa leaves extract (MLE) on physical, chemical and sensory properties of luncheon was investigated. Luncheon samples were supplemented with 0.5, 1.0 and 1.5% of MLE. Results showed that supplementation of luncheon with MLE increased its content of protein, fiber and antioxidants. Protein content ranged from 8.49% in control to 10.90% in luncheon sample with 1.5% MLE. Total phenolic content of MLE was ranged from 65 to 67 mg/100g (as Gallic acid equivalent) while, the scavenging effects of 100 µl of MLE were ranged from 79.51 to 88.57%. The thiobarbituric acid number (TBA) values increased for all the investigated luncheon samples gradually during storage period. The highest value for TBA was noticed in L1 (control sample 1) (0.5239) after four weeks of cold storage while, the lowest value was observed with luncheon meat containing 0.01% butylated hydroxytoluene (BHT) (L2) (0.1251) at zero storage. The total polyphenol contents and antioxidants activity increased in luncheon meat after the addition of moringa leaves extract. All the luncheon meat samples containing moringa leaves extract were generally acceptable.

Key words: Moringa leaves, extract, luncheon, antioxidant, sensory evaluation.

INTRODUCTION

Moringa (*Moringa oleifera* L.) is a kind of local medicinal Indian herb which has turn out to be familiar in the tropical and subtropical countries. The other expressions used for Moringa are Horseradish tree, Mulangay, Mlonge, Benzolive, Drumstick tree, Sajna, Kelor, Saijihhan and Marango. *Moringa oleifera* is shown in scientific division to become from Kingdom: Plantae, Division: Magnoliophyta, Class: Magnoliopsida, Order: Brassicales, Family: Moringaceae, Genus: *Moringa*, Species: *M. oleifera* (Fahey, 2005).

Moringa belonging to the family of Moringaceae is an efficient remedy for malnutrition. Moringa is rich in nutrition owing to the presence of assortment of essential phytochemicals present in its leaves, pods and seeds. In fact, moringa is said to provide 7 times more vitamin C than oranges, 10 times more

vitamin A than carrots, 17 times more calcium than milk, 9 times more protein than yoghurt, 15 times more potassium than bananas and 25 times more iron than spinach (Rockwood *et al.*, 2013).

Ozumba (2011) reported that moringa is an exceptionally nutritious vegetable tree with varieties of potential value. The leaves are eaten as vegetable in many cultures, either fresh or packaged. In Ghana, they are cooked and eaten like spinach or used to make soups, sauces or salads.

Lockett and Calvert (2000), Newton (2007), Mukunzi *et al.* (2011) and Satya *et al.* (2012) stated that the leaves, seeds and flowers of Moringa all have great nutritional and curative values. The seeds are eaten like peas or roasted like nuts whilst the flowers are eaten when cooked and taste such as mushrooms.

Ozumba (2011) noticed that in many tropical and subtropical countries, different parts

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of moringa (leaves, fruits, immature pods, and flowers) are integrated into the traditional food of humans. Moringa is one of the nature's gifts to humanity because of its several wealth in vitamins and minerals as well as natural anti-oxidants.

Moringa is rich in phytosterols such as stigmasterol, sitosterol and kampesterol which are precursors for hormones. These compounds increase the estrogen production, which in turn stimulates the proliferation of the mammary gland ducts to produce milk. It is used to treat malnutrition in children younger than 3 years (**Mutiara *et al.*, 2013**).

Each part of this plant is a storehouse of important nutrients and antinutrients. The leaves of *M. oleifera* are rich in minerals such as calcium, potassium, zinc, magnesium, iron and copper (**Kasolo *et al.*, 2010**). Vitamins like vitamin A, vitamin B like folic acid, pyridoxine and nicotinic acid, vitamin C, D and E are also present in *M. oleifera* (**Mbikay, 2012**). Phytochemicals like tannins, sterols, terpenoids, flavonoids, saponins, anthraquinones, alkaloids and reducing sugar are present along with anticancerous agents such as glucosinolates, isothiocyanates, glycoside compounds and glycerol-1-9-octadecanoate (**Berkovich *et al.*, 2013**).

Moringa is considered one of the most beneficial trees, in the world, as almost each part of the moringa tree can be used for food, medication and industrial purposes (**Khalafalla *et al.*, 2010**).

Moringa leaves have been reported to be a rich source of β -carotene, protein, vitamin C, calcium and potassium and act as a good source of natural antioxidants; and thus enhance the shelf-life of fat containing foods due to the presence of different types of antioxidant compounds like ascorbic acid, flavonoids, phenolics and carotenoids (**Dillard and German, 2000; Siddhuraju and Becker, 2003**).

Luncheon meat is an important industrial meat product, one of the most agreeable food products, widely consumed and used for fast food. It is usually be formed of finely chopped meat and fat with or without some added cereals, cured with salt and nitrite and heat

processed (**Ranken, 1984**). Minced meat undergo oxidative changes and develop rancidity more quickly than intact muscle since grinding exposes more of the muscle surface to air and microbial contamination (**Mitsumoto *et al.*, 2005**).

Much attention has been concentrated on extracts from herbs and spices which have been utilized traditionally to improve the sensory characteristics and extend the shelf-life of foods (**Botsoglou *et al.*, 2003**).

Refrigeration storage is usually the most common preservative method of meat and meat products. In order to increase refrigerated storage time, antimicrobial and antioxidant additives are added to foods (**Solomakos *et al.*, 2008**). So, the use of natural antioxidants from plants to produce luncheon roll meat of high quality and safety is important.

Thus, the aim of this paper is to study the chemical composition of moringa leaves and examine the effect of adding moringa leaves extract to luncheon roll meat during refrigerate storage on its physical, chemical and sensorial properties.

MATERIALS AND METHODS

Materials

Moringa leaves powder (*Moringa oleifera lam*) was obtained from Agricultural Research Center, Giza, Egypt.

All ingredients used in the preparation of luncheon meats were purchased from local super market at Zagazig District, Sharkia Governorate, Egypt.

Methods

Preparation of Extracts from Moringa Leaves

Moringa powder was extracted according to the method of **Vongsak *et al.* (2013)**. About 50 g of moringa leaves powder was mixed with 1 L of 70% ethanol (1:20 *W/V*) for 72 hr., at room temperature with occasional shaking. The mixture was centrifuged at 3000 X g for at 20°C 10 min then; it was filtered through Whatman No. 1 filter paper. The ethanolic mixture was

concentrated under vacuum at 45°C using a rotary evaporator (Buchi Waterbath B-480 with Buchi Rotavapor R-124, Germany) to obtain the crude extract. The extract was freeze dried (Vacuum freeze dryer model: FDF 0350; Korea). The extract was stored in an air-tight container at -18°C until use.

Luncheon Rolls Meat Preparation

The luncheon roll meat batter was prepared according to the following formula listed in Table 1. The procedure used in preparation of luncheon samples was carried out according to the protocol described by **Zhanc et al. (2004)** with some modifications. Processing of the luncheon samples involved blending the minced meat and fat with the other ingredients. Five batches of luncheon were manufactured: sample (L1) prepared as control without any additives; Sample (L2) was prepared by adding 0.01% BHT. Samples (L3), (L4) and (L5) were prepared by adding 0.5, 1.0 and 1.5% moringa leaves extract, respectively. Each sample was mixed well to be homogenous and packaged in sealed food bags and fibrous casings (about 500g each) and sealed. Also wrapped in aluminum foil and sealed well. Luncheon rolls were cooked in boiled water for an hour and a half then it let to cool down. The luncheon roll meats were stored in the refrigerator until analysis which was performed at different periods of cold storage.

Chemical Composition

Moisture, ash, crude protein, crude lipids, crude fiber, and ash of moringa leaves and luncheon samples were determined according to the methods recommended by **AOAC (2005)**, while total carbohydrate content was calculated by difference. All analyses were conducted in central lab for soil, food and feed staff (CLSFF), Faculty of Technology and Development, Zagazig University, Egypt.

Determination of Total Phenolic Content (TPC)

The concentration of total phenols was measured by spectrophotometer (Jenway-UV-VIS Spectrophotometer) based on a colorimetric oxidation/reduction reaction, as described by **Skerget et al. (2005)** using Folin-Ciocalteu as oxidizing reagent (**AOAC, 2005**). To 0.5 ml of diluted extract (10 mg in 10 ml solvent), 2.5 ml of Folin-Ciocalteu reagent (diluted 10 times

with distilled water) and 2 ml of Na₂CO₃ (75 g/l) were added. The sample was incubated at 50°C for 5 min then cooled. For the control sample, 0.5 ml of distilled water was used. The absorbance was measured at 760 nm. Quantification of TPC was based on a Gallic acid standard curve generated by preparing 0, 5, 10, 15, 20, 30 ml/l. of Gallic acid equivalent (GAE) and calculated using the following linear equation based on the calibration curve:

$$y = 0.015x + 0.0533$$

$$R^2 = 0.9966$$

Where (y) is the absorbance

(x) is the concentration (mg GAE g⁻¹ extract).

R²=Correlation Coefficient.

Determination of Antioxidants Activity

Antioxidants activity was determined by DPPH (2,2-diphenyl-1-picryl hydrazyle) method, scavenging effect (DPPH method) was adopted to assess antioxidative potential of the moringa extract as follows: The electron donating ability of the obtained extract was measured by bleaching of the purple coloured solution of DPPH according to the method of **Hanato et al., (1988)**. One hundred µl of each extracts (10 mg extract/ 10 ml solvent) was added to 3 ml of 0.1 mM DPPH dissolved in ethyl acetate and ethanol according to the solvent used for extraction. After 30, 60, 90, and 120 min incubation periods at room temperature, the absorbance was estimated against a control at 517 nm (**Gulcin, 2012**). Percentage of antioxidant activity of free radical DPPH was calculated as follows:

Antioxidant activity (inhibition %) =

$$\frac{A \text{ control} - A \text{ sample}}{A \text{ control}} \times 100$$

Where:

A control is the absorbance of the control reaction. A sample is the absorbance in the presence of plant extract. Tert-butylhydroquinone (TBHQ) was used as positive control.

Determination of Thiobarbituric Acid Test (TBA)

Thiobarbituric acid value was measured according to the method described by **Fernandez et al. (2005)**. About ten grams of

Table 1. Formula of control luncheon sample

Ingredient	(%)	Weight (g)
Minced lean beef meat	65	325
Back beef fat	15	75
Egg	5	25
Salt	2.5	12.5
Spices	1	5
Garlic powder	0.5	2.5
Onion powder	2	10
Potato starch	4	20
Ice	5	25
Total	100	500

sample was blended with 100 ml distilled water for 2 min. The pH of the sample was adjusted to 1.5 by adding few drops of 4N HCl and then transferred to a distillation tube. The mixture was distilled and 50 ml distillate was collected. Five ml of 0.02 M 2- thiobarbituric acid in 90% acetic acid (TBA reagent) were added to a vial containing 5 ml of the distillate and mixed well. The vials were capped and heated in a boiling water bath for 30 min to develop the chromogen and cooled to room temperature. The absorbance was measured at 538 nm, against a blank, using JENWAY 6705 UV/VIS spectrophotometer. The TBA values were calculated as mg malondialdehyde/kg sample according to the following equation:

$$\text{TBA value (/kg)} = \text{absorbance at 538 nm} \times 7.8$$

Colour Determination

Colour properties of luncheon were performed using Hunter Lab colour analyzer (Hunter Lab Colour Flex EZ, USA) according to **Singh *et al.* (2008)**. The L value (lightness index scale) ranges from 0 (black) to 100 (white), while a value indicates the redness (+a) or greenness (-a) and the b value refers to the yellowness (+b) or blue (-b).

Sensory Evaluation

Sensory evaluation of luncheon meat

The sensorial criteria (taste, flavour, texture, appearance and colour) of under investigation luncheon samples were evaluated by twenty five

untrained panelists. Luncheon samples were cut into 2mm thick slices and served in numerically-coded glass petri dishes. Each panelist received five coded samples (one from each tested samples) then independently evaluated the luncheon meat for texture, flavour, colour, appearance and taste using a 5-point hedonic scale (1 = extremely poor, 2 = poor, 3= acceptable, 4= good, 5= excellent), according to the described method of **Lavrova and Krilov (1975)**.

Statistical Analysis

The results were reported as mean \pm standard deviation (SD) (n = 3) and were statistically investigated using one-way analysis of variance (ANOVA) with Duncan by SPSS for Windows 16.0. A statistical probability (p value) less than 0.05 indicated a statistically significant difference between groups (**Steel and Torrie, 1980**).

RESULTS AND DISCUSSION

Chemical Composition of Luncheon Samples and Moringa Leaves

The chemical composition of luncheon samples is presented in Table 2. The moisture content of luncheon samples ranged between 52.97 and 57.60%. From the results it was noticed that, moisture content was decreased with the addition of moringa leaves extract. Protein content was increased in luncheon samples fortified with MLE, it was ranged

Table 2. Chemical composition of the prepared luncheon meat treatments

Sample	(%)					
	Moisture	Crude protein	Crude fat	Ash	Crude fiber	Carbohydrate
L 1	57.60 ^a ±0.08	8.49 ^e ±0.01	8.04 ^e ±0.05	3.27 ^e ±0.05	3.04 ^e ±0.01	19.56 ^c ±0.02
L 2	57.03 ^b ±0.03	8.45 ^e ±0.02	8.02 ^d ±0.02	3.19 ^e ±0.05	3.06 ^e ±0.04	20.25 ^b ±0.04
L 3	54.12 ^c ±0.04	9.61 ^d ±0.01	8.21 ^d ±0.01	3.92 ^d ±0.05	3.42 ^d ±0.02	20.72 ^c ±0.05
L 4	53.64 ^d ±0.04	10.08 ^c ±0.02	8.75 ^c ±0.01	4.26 ^c ±0.02	3.76 ^c ±5.29	19.51 ^c ±0.03
L 5	52.97 ^e ±0.04	10.90 ^b ±0.01	9.070 ^b ±0.03	4.57 ^b ±0.01	3.91 ^b ±0.01	18.58 ^d ±0.02
Moringa leaves	5.90 ^e ± 0.01	22.16 ^a ±0.01	10.70 ^a ±0.06	11.77 ^a ±0.01	9.91 ^a ±0.01	39.56 ^a ± 0.03

* The results are presented as the mean value ± SD. Values expressed with different treatments are significantly different at P<0.05 L1: Control (Luncheon meat free from moringa leaves extract)- L2: Luncheon meat containing 0.01% BHT- L3: Luncheon meat containing 0.5% moringa leaves extract- L4: Luncheon meat containing 1% moringa leaves extract- L5: Luncheon meat containing 1.5% moringa leaves extract.

between 8.49% in control sample L1 to 10.90% in L5 (luncheon with 1.5% MLE). Table 2 show also that there is a gradual increase in ash and crude fiber contents with the addition of MLE. These results are consistent with the findings of **Abdullah (2007)**.

The chemical composition of moringa leaves is tabulated in Table 2. From the results it could be noticed that the moisture, protein, fat, ash and fiber contents were 5.90, 22.16, 10.70, 11.77 and 9.91%, respectively. These results are in agreement with the findings of **Satwase et al. (2013)**, **Offor et al. (2014)** and **Mansour (2017)**.

Total Phenolic Compounds in Moringa Leaves (TPC)

Phenolic compounds and flavonoids are very important constituents due to antioxidants activity. Moringa leaves regarded as a promising rich source of phenolic compounds. The Polyphenol contents of moringa leaves extract was determined. The total phenolic content was ranged from 65 to 67 mg/100g (as Gallic acid equivalent). These result are in agreement with those reported by **Mansour (2017)** who found that total polyphenol content (65.5 mg/100g) was recorded for the oven dried samples at 70°C, and **Singh and Prasad (2013)** who found that the total polyphenol content of moringa leaves ranged from 23.57 to 155.15 mg/100g

depending on the drying temperature, particle size and blanching process. **El-Masry et al. (2013)** reported that total polyphenol contents were reduced as a result of drying process in comparison with fresh samples.

Antioxidant Activity of Moringa Leaves Extract

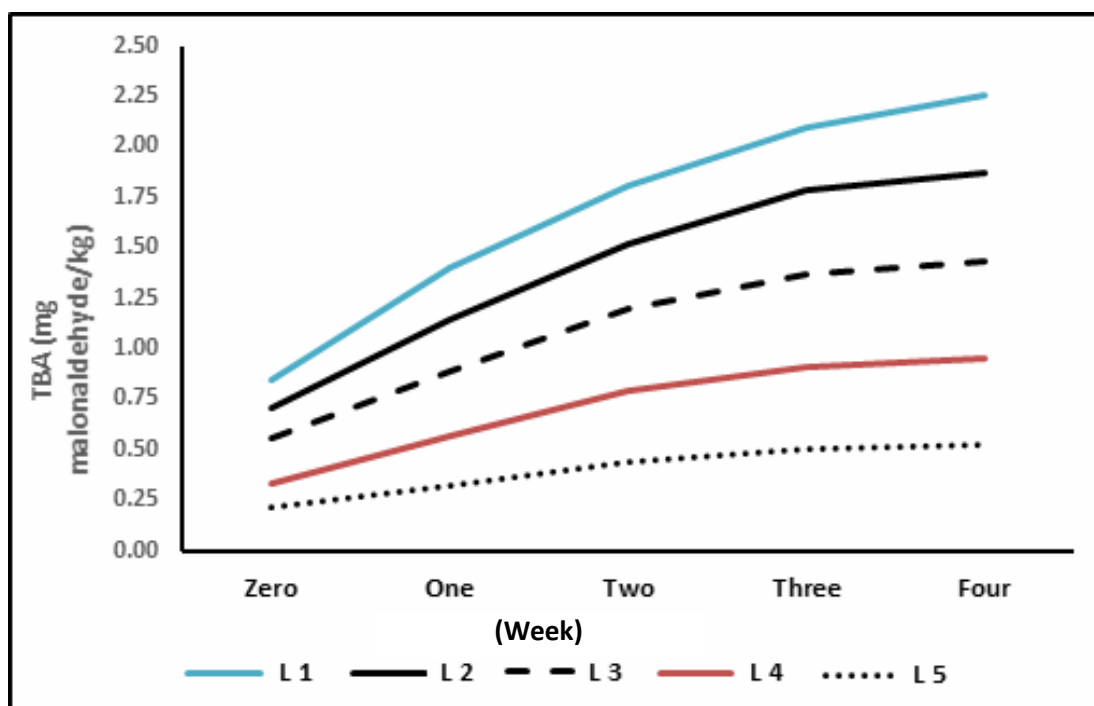
Table 3 show the antioxidants activity of moringa leaves extract. The antioxidants activity determined by DPPH of moringa leaves powder was ranged from 79.51 to 88.57% (zero time to 120 min.). This result agree with that of the antioxidants activity reported by **Mansour (2017)** who found that dried moringa leaves powder was 92.46%. **Pakade et al. (2013)** found that the antioxidants activity of moringa was in the range from 59.8 to 40.4%. In view of the above, Moringa leaves can be considered as a good source of natural antioxidants.

Changes in Thiobarbituric Acid (TBA) of Luncheon Meat During Cold Storage

The TBA of luncheon meat during cold storage is shown in Fig. 1. From the results, it was found that moringa leaves extract decreased the formation of TBA in luncheon samples during the cold storage period (four weeks). Results indicated also that TBA values of the luncheon samples showed no great differences between all prepared samples at zero time.

Table 3. Antioxidant activity of Moringa leaves extract

Sample	DPPH method (time, min)				
	Zero	30	60	90	120
Moringa leaves extract	79.51%	87.10%	87.95%	88.33%	88.57%

**Fig. 1. Changes in TBA (mg malonaldehyde/kg) of luncheon meat treatments during cold storage**

These values increased for all the investigated samples gradually during storage period. The highest value for TBA was noticed in control sample (0.5239) after four weeks of cold storage while, the lowest value was observed with L2 (0.1251) at zero storage. The highest value of TBA was determined after four weeks of cold storage with each of the following treatments L1, L2, L3, L4 and L5 (0.524, 0.428, 0.484, 0.437 and 0.382, respectively). Furthermore, when frozen fat is used, especially at high levels, oxidative rancidity resulting from excessively long storage commonly occurs (Pearson and Tauber, 1984).

Colour Evaluation of Luncheon Meat

Colour analysis of food product is an important parameter defining consumer's choice

and acceptability and controlling the first impression of the food product. The colour values of luncheon samples are presented in Table 4. Lightness (L^*) was 39.15 in zero time and 38.82 after 15 days (control luncheon meat). Lightness (L^*) was decreased in L4 after 15 days and increased in L2, L3 and L5 (zero time and after 15 days). Redness (a^* value) was increased to 15.36 and 17.86 in L4 (zero time and after 15 days) as compared to that observed in control (L1); 13.66 and 14.73 (zero time and after 15 days), and decreased in L2, L3 and L5. Treatments on the other hand, the yellowness (b^* value) was no great differences between all prepared samples where ranged from 12.73 to 14.43 but increased to 15.09 in L5 (after 15 days) and 16.31 in L3 (after 15 days).

Table 4. Colour evaluation of luncheon meat

Sample	L*	a*	b*	
L 1	0 time	39.15 ^f	13.66 ^d	13.64 ^e
	15 days	38.82 ^g	14.73 ^c	12.84 ^f
L 2	0 time	42.09 ^d	3.86 ⁱ	13.85 ^d
	15 days	42.09 ^d	3.77 ^j	14.43 ^c
L 3	0 time	40.75 ^e	4.62 ^g	13.98 ^d
	15 days	43.02 ^b	5.45 ^e	16.31 ^a
L 4	0 time	35.29 ⁱ	15.36 ^b	10.79 ^g
	15 days	38.42 ^h	17.86 ^a	12.73 ^f
L 5	0 time	42.66 ^c	5.22 ^f	14.42 ^c
	15 days	45.57 ^a	4.32 ^h	15.09 ^b

(L*):- Lightness (a*):- Redness (b*):- yellowness

Changes in Total Phenolic Compounds and Antioxidant Activity of Luncheon Meat

The polyphenolic contents of luncheon meat were determined, and the obtained results are presented in Table 5. The total polyphenol contents were 38.82, 72.7, 75.51, 76.89 and 79.9 mg/100g in L1, L2, L3, L4 and L5 treatments, respectively.

Table 5 show the antioxidants activity of luncheon meat during cold storage. The antioxidants activity determined by DPPH of luncheon meat was 20.4 in L1, then increased with the addition of moringa leaves extract, from 53.9 in L2 to 62.8 in L5 treatments.

Sensory Evaluation of Luncheon Meat

Results of sensory evaluation indicated that all the luncheon meat samples containing moringa leaves extract were generally acceptable for all tested parameters as non-scored below the minimum acceptable rating of three (Table 6). However, these luncheon meats were significantly differ from content. Results in Table 6 shows the results of L1, L2 and L3 are approximated. The results of sensory tasting showed that the taste was acceptable with good score for all luncheon meat samples. Flavor score was found to be the highest in L1 compared to other studied luncheon samples; while L5 was of worst score. The highest colour

score was for L1 and L4, followed by L2, meanwhile the L3 and L5 showed a relative low score. However, all the investigated samples (including the control) realized good colour scores. With regard to texture, L1 was found to be of highest texture score; while L5 was of lowest score. The highest score in appearance was 4.5 in L4 while, the lowest was 3.7 in L5. **Abu-Salem et al. (2011)** showed that the texture ranged from 3.7 to 4.0, flavour ranged from 3.7 to 4.2, colour ranged from 4.1 to 4.4 and taste ranged from 3.8 to 4.1.

Conclusion

This investigation shows the potential value of moringa leaves extract (MLE) as a good natural source of nutritive components, total phenolic compounds and antioxidants. Based on its total phenolic compounds and antioxidants, moringa leaves extract could be very suitable as a natural additive or substituted material in the production of many foodstuffs. The information obtained in the present investigation is useful for characterizing moringa leaves extract and for the industrial utilization in luncheon meat preparation. The addition of different concentrations of moringa leaves extract improved the quality criteria of luncheon and increased the values of total phenolic compound and antioxidants activity of luncheon after added moringa leaves extract.

Table 5. Changes in total phenolic compounds and antioxidant activity of luncheon meat as affected by treatments preparteion

	Polyphenol contents (mg/100g)	Antioxidant activity (DPPH)
L 1	38.82 ± 0.055	20.4 ± 0.02
L 2	72.70 ± 0.042	53.9 ± 0.013
L 3	75.51 ± 0.045	55.9 ± 0.068
L 4	76.89 ± 0.071	59.1 ± 0.014
L 5	79.90 ± 0.063	62.8 ± 0.07

Table 6. Sensory evaluation of luncheon meat

Luncheon meat sample	Sensory evaluation				
	Colour	Appearance	Flavour	Texture	Taste
L 1	4.3 ^a	4.1 ^{ab}	4.5 ^a	4.6 ^a	4.7 ^a
L 2	4.1 ^{ab}	4.1 ^{ab}	4.1 ^{abc}	4.3 ^{ab}	4.3 ^{ab}
L 3	3.8 ^{ab}	3.9 ^b	4.0 ^{bc}	4.2 ^{ab}	4.1 ^b
L 4	4.3 ^a	4.5 ^a	4.4 ^{ab}	4.3 ^{ab}	4.3 ^{ab}
L 5	3.7 ^b	3.7 ^b	3.7 ^c	4.0 ^b	3.5 ^c

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تأثير إضافة مستخلص أوراق المورينجا على جودة لانشون اللحم

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تم دراسة تأثير مستخلص أوراق المورينجا (MLE) على الخواص الفيزيائية والكيميائية والحسية للانشون اللحم، تم تدعيم عينات اللانشون مع 0,5 ، 1 و 1,5% من MLE، أظهرت النتائج أن تدعيم اللانشون ب MLE زاد محتواه من البروتين والألياف ومضادات الأكسدة حيث تراوحت نسبة البروتين من 8,49% (الكنترول) إلى 10,90% في عينة اللانشون المدعمه ب 1,5% MLE، تراوح محتوى المركبات الفينولية في الـ MLE من 65 إلى 67 ملجم/100جم بينما تراوح مضادات الأكسدة من 79,51% إلى 88,57%، زادت قيم TBA لجميع عينات اللانشون تدريجيا خلال فترة التخزين، لوحظت أعلى قيمة لـ TBA في L1 (0,5239) بعد أربعة أسابيع من التخزين البارد، بينما لوحظت أقل قيمة مع L2 (0,1251) عند صفر تخزين، زاد إجمالي محتوى مركبات البوليفينول ونشاط مضادات الأكسدة في اللانشون بعد إضافة مستخلص أوراق المورينجا، كانت جميع عينات اللانشون التي تحتوي على مستخلص أوراق المورينجا مقبولة بشكل عام لجميع المعاملات التي تم اختبارها حيث أنها غير مسجلة دون الحد الأدنى المقبول وهو 3 بالرغم من اختلاف محتويات اللانشون، لذلك يستنتج من هذه الدراسة أهمية إضافة مستخلص أوراق المورينجا إلى اللانشون.

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