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Antimicrobial activity of bottle gourd leaf extract and its effect on the quality of food

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Abstract:

A considerable interest has developed on natural foods preservatives as extend the shelf-life. There is a strong controversy about the safety aspects of synthetic preservatives since they are considered responsible for many teratogenic attributes and carcinogenic as well as residual toxicity. Therefore, the antimicrobial activity, total phenolic and flavonoids of bottle ground leaf extract were examined. And a descriptive detection of the extract was made, where it was found that they contain (phenols, flavonoids, alkaloids, glycosides, terpenes, saponins, phlobatanins, resins, tannins And steroids). 50% ethanolic extract exhibited the highest extraction ability for such phenolic and flavonoid compounds. The total phenolic was 406 mg gallic equivalent/g and the total flavonoid was 42.6 mg quercetin/g dry weight. The antimicrobial activity was studied using Agar well diffusion method and broth dilution method. Results indicated that Gram-negative bacteria *E. Coli* was more sensitive than Gram-positive bacteria *S. aureus* and *B. subtilis* against bottle ground leaf extract, were 25, 18 and 19 mm inhibitory zone for concentration 50mg/well respectively. MIC and MLC where determined, MICs test showed that the lowest concentration was 10 mg/ml against *S. aureus*, and was equal concentration on *E. coli and B. subtilis* (20 mg/ml). On the basis of this results bottle ground leaf extract was added to beef sausage at different concentrations (1%, 2%, and 4%) and stored for 90 days at -٢٠°C to study its effect on the quality of beef sausage. Results indicated a high stability against oxidation in sausage treated with the bottle ground leaf extract compared to control samples. Also, a decrease in microbial load of the treated sausage compared to the control was observed. Finally, it was evident that the bottle gourd leaf extract has a role in reducing the number of microbes, as well as their possession of natural antioxidants that prolongs shelf life of sausage.

Key words: bottle gourd, Natural extracts, quality control, Antimicrobial activity and beef sausage.

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Introduction:

Bottle gourd is belongs to genus *Cucurbita* and family *Cucurbitaceae*. Bottle gourd *(Lagenaria siceraria)* is a green colour, longitudinal vegetable synonym calabash gourd belongs to the family *cucurbitaceae*. Bottle gourd most probably originated in tropical Africa, and occupies first place in India. It is only the crop known to have been cultivated in pre-Columbian times in both the world and new world (Lakshmaiah et al., 2018). Archaeological remains indicate that the bottle gourd was used in Egypt from 3500 to 3300 B.C (Stephens, 2018). (Lakshmaiah et al., 2018) reported that the fruits, stem, leaves, seeds and oils of "Lagenariasiceraria" are traditionally used in the treatment of diabetes, ulcer, hypertension, cardiac failure, and skin diseases, leaves are used as alternative purgative, the stem bark and rind of the fruit are diuretic, leaf juice is widely used for baldness, the flowers are an antidote to poison, extracts of the plant shows antibiotic activity, it is rich in thiamin, vitamin C, zinc, iron and magnesium thus helping in improving overall health and bottle gourd is also considered one of the best foods for weight loss.

(Handayany, 2017) Clarified that the ethanol extract of bottle ground leaves (*Lagenaria Siceraria*) has antibacterial activity against *Staphylococcus aureus*, *staphylococcus mutans*, *Staphylococcus epidermidis* and *E. Coli.* (Ammu, 2018) showed that extracts of leaves from *Lagenaria siceraria* have effects of bacteria *S.typi, S. enteritidis, E.coli , S. aureus, K. pneumonia*. (Rodge & Biradar, 2012) clearly indicates that the leaf extracts of *Lagenaria siceraria* possess potent antimicrobial activity against *Escherichia coli, and Bacillus subtilis* Comparative analysis of this study reveals that the bacterial inhibition elicited by the plant extract was higher than that of antibiotic Cephalexin. (Badmanaban et al., 2010), (Nagaraja et al., 2011) found that when compared to aqueous extract, the ethanolic (70%) extract of leaves of *Lagenaria siceraria* Mol has antibacterial and antifungal activity against the microorganisms examined. (*Escherichia coli, Pseudomonas aeruginosa, Klebsiella aeruginosa, Staphylococcus aureus, Aspergillus's niger and Candida albicans*) when compared with the standards.

In leaf, acetone and ethanol were the most suitable for the extraction of flavonoids, phenolics and tannins. In antioxidant assay of leaf, FRAP, DPPH, and ABTS assay responded significantly with ethanol and methanol. However, highest metal chelating and phosphomolybdenum activity were recorded in the aqueous extract of leaf (Patel et al., 2018). (Sharma et al., 2013) indicated that *L. siceraria* leaves extract exhibited antioxidant activity against hydroxyl, peroxide and $DPPH^{\bullet}$. Its polyphenolic content and other phytochemical constituents may be linked to the overall antioxidant activity of MELS. The results of this study indicated that L siceraria leaves may be a potential source of natural antioxidants that would be of great importance to prevent or delay development as therapeutic agents.(Badmanaban et al., 2010), (Satish et al., 2010) showed that the leaves of Lagenaria siceraria (Molina) Standl have promising antioxidant activity.

Sausage is the most appetizing and widely utilized processed food. The word "sausage" is derived from the Latin word *salsus*, which means salted. A sausage is any salted, ground meat, and there are many different types of sausages produced (Mohan, 2014). Sausage is considered one of the most beef suitable for human consumption. Beef sausage is one of the most popular meat products in Egypt and it's mostly produced from beef meat, fat tissues, dry rusk, salt and spices. (Shahin, 2016).

Objectives of the study was to evaluate antimicrobial and antioxidant of bottle gourd leaf extract. Also, to examine the addition of bottle gourd leaf extracts on sensory, proximate, physicochemical, microbiological analysis and recommend value added beef sausage enriched with bottle gourd leaf extracts.

2. Materials and Methods:

2.1. Materials

2.1.1. Raw Material

Leaves of Bottle gourd (*Lagenaria siceraria)* was collected during July 2018-2019 from various farms at Fayoum. The collected plant leaves were dried, powdered and stored in deep freeze at -20°C until analysis.

2.1.2. Chemicals, Solvents, reagents and references

Sodium carbonate, aluminium chloride (AlCl3), ethanol, sulphoric acid, hydrochloric acid, Hexane, ethanol, sodium hydroxide, boric acid, thiobarbturic acid reagent, glacial acetic acid, sodium thiosulphate, sollible starch, were from Elgamahoreyah (Egypt). Folin–Ciocalteu reagents were obtained from Sigma Chemical Co. Methanol was purchased from Aldrich Co. All other solvents and chemicals were of analytical grade.

In-vitro diagnostic discs (Pasteur LBA, Egypt) of 2 antibiotics (Penicillin and Ampicillin) were used to test the susceptibility of the used microorganisms.

2.1.3. Microorganisms

One species of gram-negative bacteria (*Escherichia coli*, O:157 ATCC 1659) and two gram-positive bacteria (*Bacillus subtilis* and *Staphylococcus aureus* ATCC 13565) were used as indicator microorganisms for detection of the antimicrobial activity. All strains mentioned above were obtained as actively growing cultures collection of the Microbiological Resources Centre Cairo (MIRCEN), Faculty of agriculture, Ain shams University, Cairo, Egypt.

2.1.4. Media

LB agar medium was used to determine the antimicrobial effect of antibiotics or plant extracts by disk diffusion method. It was also used as a completed step to determine the MLC (Minimum Lethal Concentration) in the broth dilution method for both antibiotics and plant materials. Total count agar, potato dextrose agar, baird parker agar, MacConkey agar and salmonella shigella agar were prepared according to the methods described in (Zimbro et al., 2009).

2.2. Methods

2.2.1. Preparation of plant extract

Ten grams of ground bottle leaf powder were weighed into Erlenmeyer flasks, then 100 ml of 50 % ethanol was added. Extraction was carried out by shaking at room temperature overnight. After filtration through filter paper (Whatman No.4), the residue was re-extracted twice, and then the combined extracts of were evaporated and dried in desiccators under vacuum to a constant weight.

2.2.2. Phytochemical screening tests

The color detection means were adopted according to the method mentioned (**Apsara, 2012**)

2.2.3. Determination of total phenolic and flavonoid compounds in bottle gourd leaf extract:

The total phenols were quantified using the colorimetric method of Folin–Ciocalteau (Chavan & Singhal, 2013). Total flavonoid contents were determined using the method of (Ordoñez et al., 2006).

2.2.4. Antimicrobial activity of the plant extracts

2.2.4.1. Well diffusion method:

The Antimicrobial susceptibility testing was done by using the Agar well diffusion method to detect the presence of antibacterial or antifungal activities of the

samples (Jagessar et al., 2008)**.** Three wells of 6 mm diameter were punched using the wide end of a sterile Pasteur pipette in each standard plate. The medium was previously added with 1 ml of the prepared *Bacillus subtilis*, *staphylococcus aureus* and *E. coli*. The dried extract was rehydrated in sterile distilled water, heat-sterilized by autoclaving non sterilized solution at 121°C for 15 min. Extract solution (1mg/1ml) was added into each well. The plates were incubated at 35 °C for 24 h, and 37 °C for 48 h for *Bacillus subtilis*, *Staphylococcus aureus* and *E.coli* respectively. Clear zones (mm) of inhibition were measured. Each plate contained three wells for two types of plant extracts, five concentrations of extract solution, and a distilled water control, respectively. The diameter (mm) of radial growth of each colony was measured after 48 h of incubation.

2.2.4.2. Plant antimicrobial effectiveness assessment by broth dilution method (MIC and MLC)

In this method a standard Known count of the tested microorganism must be used, which can be determined in a liquid culture by visually comparing the turbidity of the liquid medium to a standard that represents a known number of the bacteria in suspension (Eloff, 1998)

2.3. application of the plant extracts

2.3.1. Preparation of sausage

Sausage was prepared by mixing minced meat with the other ingredients which are listed in Table (1) according to the method of (Abdulla et al., 2016). Four batches of sausage were prepared according to the following addition system; Control (as described in Table. 1. (With no additives), B1 (with the addition of Bottle gourd leaves extract at 1 g/100g), B2 (with the addition of 2 g/100g of Bottle gourd leaves extract), B3 (with the addition of 4 g/100g of Bottle gourd leaves extract), the extract was thoroughly mixed with ice water before adding to the sausage formula. Formulated sausage mixtures were filled into sheep casing using filler (moulinex, France). Sausage samples of every treatment (triplicates) were stored at freeze at -20C for further use and analysis. A part of each sample was separated for the analysis at zero time, and a part of each batch was cooked for sensory evaluation.

Ingredients	Percentage	Amount (g)
Beef meat	70.9	354.5
Fat	10.9	54.5
Ice water	10.9	54.5
Starch	3.3	16.5
Skimmed milk	2.2	11
Garlic	0.3	1.5
White pepper	0.1	0.5
Ginger powder	0.1	0.5
Coriander	0.2	1
Nutmeg	0.1	0.5
Salt	1.0	5
Total	100	500

2.3.2 Chemical quality attributes:
Thiobarbituric acid (TBA) value and pH value were determined according to AOAC (2005).

2.3.3. microbiological Examination
Aerobic plate count, *Escherichia coli* count, yeast and molds counts, *Staphylococcus* count and *Salmonella* and *Shigella* count It was performed according to the guide lines recommended by ISO 4833, (2003).

2.3.4. Sensory Evaluation of Sausage
Samples for sensory evaluation were carried out in the sensory assessment facilities of the meat laboratory, twelve staff member food science and technology department and milk science department, Faculty of Agriculture, Fayoum University, were asked to evaluate the treatment effects on taste, texture, odor, appearance and juiciness of the sausage samples. Until cooking, the samples used for sensory assessment were randomly selected and thawed for 24 hours in the 4C fridge. The sausages were cooked separately for 6-10 minutes by frying them with deep fat in vegetable oil. To avoid excessive browning, sausages were turned on every three minutes. For assessment, samples were kept warm. They were put in coded dishes and served to the panellists warmly. A sample of 5 fingers was randomly positioned in a dish split into five sections under lamb light from each procedure. A hedonic five point scale was used. For use between testing samples, tap water was available.

2.4. Statistical analyses
The experiment was designed as a randomized complete block design with nine treatments and ten replicates(Gomez & Wiley, n.d.). The data analysis was made using analysis of variance following general linear model procedure by using the Info Stat computer software package (version, 2012). Means were separated using Duncan's multiple range test at the 95% level of probability.

3. Results and Discussion:

3.1. Chemical detection tests for some of the active substances in bottle gourd leaf extract:
Chemical detection tests, which the detection of various active compounds in the plant.The phytochemical screening of bottle gourd leaves extract revealed the presence of phenols, flavonoids, saponins, glycosides, alkaloids, terpenoids, phlobatanins, tannins and resins, it does not contain steroids. The results summarized in table (1), this result agree with **Badmanaban *et al.*, 2010** Who found that 70% ethanol extract and aqueous extract from the same plant

contain flavonoids, glycosides and tannins, but it did not contain alkaloids, as well as the ethanol extract was found to contain steroids, in contrast to the results obtained from this study. This difference is due to the type of solvent

These results give us good predictions of the biological activity of the plant although generally speaking, they remain especially encouraging as these compounds are known to participate in many biological activities from among them antifungal, antiviral, and antioxidant **(Athamena, 2009).**

Table (1) Screening of phytohchemical from bottle gourd leaf extract.

Test	Bottle gourd leaf extract
Saponins	+
Flavonoids	+
Phenols	+
Glycosides	+
Alkaloids	+
Tannins	+
Phlobatanins	+
Steroids	-
Terpenoids	+
Resins	+

*(+)= indicates presence, (-)= indicates absence

3.2. Determination of total phenolic and flavonoids:

The total phenolic and flavonoids of bottle gourd leaf extracts were quantified by the Folin-ciocalteau and AlCl3 method, respectively.

From the results recorded in figure (1), it is clear that the total phenolic content of bottle gourd leaf extract was (406 mg GAE / g), this result is better than obtained by (Patel et al., 2018)**,** who found that the ethanolic extract from the same plant contain 195.15mg tannic/g. While the total flavonoids content in the extract was (42.6 mg QE/ g), it is less than obtained by (Patel et al., 2018) and higher than (Badmanaban et al., 2010)**.** This difference may be due to the type of solvent used in the extraction.

3.3. Antimicrobial activity of plant extracts:

The antibiotic sensitivity test as shown in table (2) reveals that all examined bacteria were highly sensitive to the applied antibiotics except gram-negative bacteria which exhibited moderate sensitivity to ampicillin.

The inhibitory effects of different concentrations of bottle gourd leaf extract and of preparation methods of the extracts on the growth *of E. coli, S. aureus and B. subtilis* in an agar-well screening technique are presented in Table (2) and figure (2). The alcoholic extracts showed different antibacterial activities against bacteria. The extract had strong activity on *E. coli* 20, 24 and 25mm inhibitory zone for 30, 40 and 50mg/well respectively. The alcoholic extract showed lower inhibitory effect against s. *aureus, B. subtilis* than *E. coli* at concentrations 30, 40 and 50 mg/well. The results are in agreement with that reported by (Handayany, 2017).

Form these results we conclude that leaf extract of *Iagenaria siceraria* may contains the main specific components that responsible for antimicrobial effect on gram negative *E.coli.* since moderate effect was seen on gram positive bacteria *S. aureus and B. subtilis*. agree with

(Tirumalasetty et al., 2014) who stated that methanolic leaf extract of *Lagenaria siceraria* has maximal inhibitory zone on *E. coli* followed by other organisms.

MIC was defined as (the lowest concentration of the test sample that resulted in complete inhibition of growth). In table (2) MIC showed values generally ranging from 10 and 20 mg/ml. In MICs test showed that the lowest concentration has antimicrobial activity was 10 mg/ml of bottle gourd leaves extracts against *S. aureus*, it is also evident from table (2) that MIC of the extract was equal concentration on *E. coli and B. subtilis* (20 mg/ml), the results agree with(Rodge & Biradar, 2012).

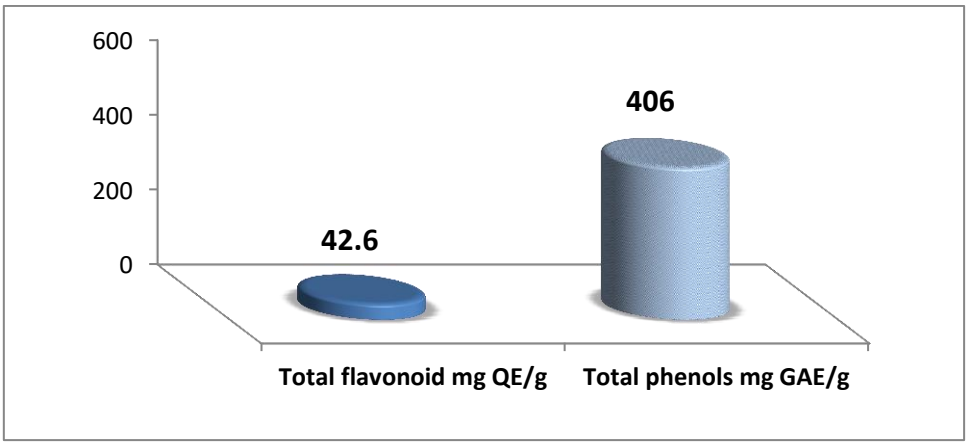

Fig. 1: Total phenols and flavonoids of bottle gourd leaf extracts.

Table (2): Inhibition zone diameter (mm) and Minimum Inhibitory Concentration (MIC) of bottle gourd leaf extract

Tasted materials	Conc. mg/well	Inhibition zone on microorganisms		
		E. coli	***S. aureus***	***B. subtilis***
Bottle gourd	**10**	11	11	10
	20	14	14	14
	30	20	15	16
	40	24	16	18
	50	25	18	19
Penicillin	**10µg**	17	14	14
Ampicillin	**10µg**	13	15	16
MIC mg/ml		20	10	20

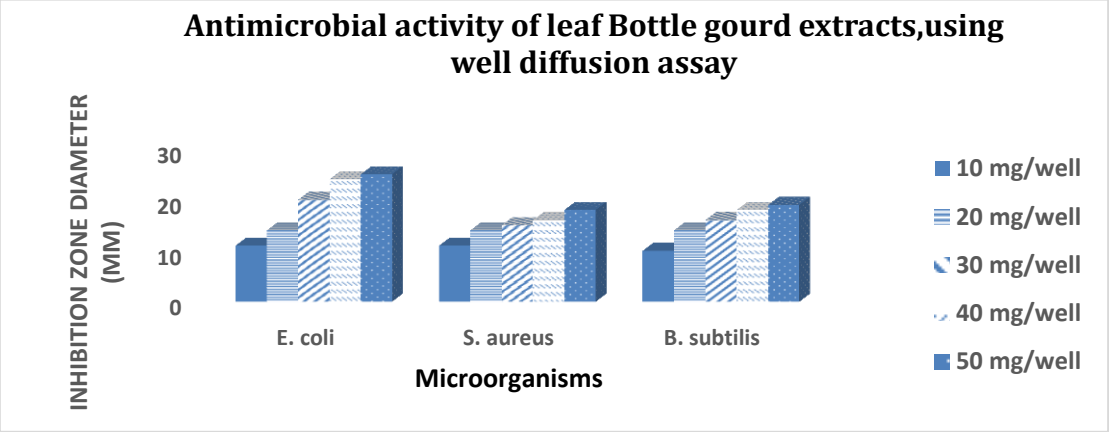

Fig. 2: Antimicrobial activity of Bottle gourd leaf extracts, using well diffusion assay (Inhibition zone diameter (mm)).

3.4. Microbiological quality criteria of experimental sausage samples during frozen storage for 12 weeks

Effects of bottle gourd leaf extract on different microbial count in beef sausage have shown in Table (3). TCB of beef sausage ranges from 6.54–6.26 (log_{10} CFU/g) at different treatment. Among four treatments, the TCB in the control (6.54 logs CFU/g) was significantly higher than in the samples treated with 1%, 2%, and 4% of bottle gourd leaf extracts. The less amount of TCB value indicates this product is most preferable for consumers' health.

The range of TYMC of beef sausage was 2.42 to 1.98 (log_{10}CFU/g), at different treatment levels. The range of TCC of beef sausage was 3.97 to 3.3 (log_{10}CFU/g), at different treatment levels. The range of TSC of beef sausage was 5.55 to 5.04 (log_{10}CFU/g), at different treatment levels. From the table (3) concluded that all the treatments were higher than the control, and the effect was increased with increasing concentration. From these it was found that the bottle gourd leaf extract can be used to extend the shelf life of sausage.*Salmonella* and shigella were not detected in samples at storage periods. **3.5. Chemical quality criteria of experimental sausage samples during frozen storage for 12 weeks:**

Changes in TBA value (expressed as mg malonaldehyde/Kg meat) of ripened sausage samples during frozen storage at -20°C for 0, 3, 6, 9, 12 weeks, were studied and the obtained results are shown in table (4). From table 4 shows that the range of overall observed TBA value at different treatment levels was 0.27 to 0.15. The TBA value in the control (0.27) was significantly higher than in the samples treated with 1%, 2%, and 4% of bottle gourd leaf extract. The result indicated that bottle gourd contains natural antioxidants reduced the oxidative rancidity in sausage samples. This result agrees with (Saba et al., 2018).

The results presented in the table (4) showed that the range of overall observed pH value at bottle gourd leaf extract treatments was 5.86 to 6.13. The result showed that the bottle gourd leaf extract was more influential on the pH values Especially the treatment containing the concentration of 2% BE. It follows from this that the additives have caused an increase in the strong and effective anti-microorganism effect and thus a decrease in their activity, which leads to a decrease in the pH as it has been proven that the smaller the number of bacteria and their activity, the greater the pH in addition to that a study (Sung & Collins, 2000) has

shown an inverse relationship between PH value and the number of microorganisms.

Table (3): Change in microbial Count of sausage samples during frozen storage for 12 weeks.

Parameters	Storage time	Treatments			
		Control	1%BE	2%BE	4%BE
Total Count bacteria log_{10} cfu/g	0	6.85±0.10	6.69±0.11	6.87±0.21	6.67±0.11
	3	6.76±2.92	6.68±2.10	6.68±1.92	6.41±2.00
	6	6.67±1.61	6.54±1.50	6.52±1.62	6.3±1.70
	9	6.34±2.05	6.33±2.10	6±1.65	6±2.00
	12	6.1±1.77	6±1.75	6±1.97	5.9±1.33
	Mean	6.54^{c}±0.31	6.45^{bc}±0.29	6.41^{b}±0.40	6.26^{a}±0.31
Total Yeasts and molds Count log_{10} cfu/g	0	2.9±019	3.2±0.17	3.3±0.20	3.3±0.22
	3	2.8±0.98	2.1±0.89	2.1±0.95	2.1±0.87
	6	2.4±0.53	2±0.55	2±0.64	1.9±0.52
	9	2.2±0.64	1.9±0.57	1.7±0.54	1.4±0.68
	12	1.8±0.46	1.7±0.43	1.3±0.35	1.2±0.44
	Mean	2.42^{b}±0.45	2.18^{ab}±0.59	2.08^{ab}±0.75	1.98^{a}±0.82
Total Coliform group Count log_{10} cfu/g	0	5±0.12	5±0.09	5.1±0.10	5±0.07
	3	4.27±1.77	4±1.65	3.9±1.34	3.77±1.72
	6	3.69±0.74	3.36±0.77	3.3±0.65	2.95±0.71
	9	3.47±1.06	3±1.03	2.9±1.10	2.77±1.09
	12	3.44±0.91	2.9±0.85	2.6±0.93	2±0.76
	Mean	3.97^{c}±0.66	3.65^{b}± 0.87	3.56^{ab}±0.99	3.30^{a}±1.14
Total Staphylococcus Count log_{10} cfu/g	0	5.84±0.10	5.82±0.13	5.85±0.06	5.81±0.08
	3	5.73±2.44	5.47±2.10	5.43±2.03	5.25±1.97
	6	5.66±1.28	5.34±1.32	5±1.08	5±1.24
	9	5.54±1.70	5±1.64	4.84±1.33	4.69±1.27
	12	5±1.39	4.9±1.51	4.69±1.16	4.47±1.36
	Mean	5.55^{c}± 0.33	5.31^{b}±0.37	5.16^{ab}±0.47	5.04^{a}±0.52

*BE= Bottle gourd leaf extract, Mean in each row having different superscript varies significantly at values $p < 0.05$.

Table (4): Change in TBA value and pH value of sausage samples during frozen storage for 12 weeks.

Parameters	Storage time (weeks)	Treatments			
		Control	1%BE	2%BE	4%BE
TBA	0	0.156	0.156	0.156	0.156
	3	0.312	0.156	0.156	0.156
	6	0.390	0.234	0.156	0.234
	9	0.238	0.117	0.156	0.156
	12	0.234	0.156	0.171	0.156
	Means	0.27^{b}±0.09	0.16^{a}±0.04	0.16^{a}±0.01	0.15^{a}± 0.02
pH	0	5.99	5.55	5.55	5.58
	3	6.15	5.89	6.2	5.92
	6	5.9	6.25	6.43	6.17
	9	5.81	5.57	6.16	5.91
	12	5.87	6.03	6.29	6.19
	Mean	5.94^{a}± 0.13	5.86^{a}± 0.30	6.13^{a}± 0.34	5.95^{a}±0.25

*BE= Bottle gourd leaf extract, Mean in each row having different superscript varies significantly at values $p < 0.05$.

3.6. Sensory evaluation of experimental sausage samples:

Sausage samples were organoleptically evaluated and the obtained results statically analyses. The obtained results are presented in table (5). The treatment with the highest number on average is the highest in determined quality characteristics. overall, the results of the statistical analysis of the sensory characteristics showed that the samples treated with bottle gourd leaf extract 1%, 2% and 4%, were equal to the degree perceptual quality of the control.

Conclusion

In conclusion, microbiological, biochemical and sensory studies shows that bottle gourd leaf extract shown better performance for beef sausage preservation. In addition the bottle gourd leaf extract contains active substances such as phenols, flavonoids, alkaloids and glycosides, and these extracts have an antimicrobial and antioxidant effect. So, it may be concluded that pumpkin leaf extract can be used in beef sausage as a source of antioxidant and antimicrobial agent to increase shelf-life of sausage.

Table (5): Sensory evaluation of experimental sausage samples:

Sausage samples	Taste	Odor	Texture	Appearance	Juiciness	overall
Control	8.58^{ab}± 1.44	8.50^{ab}± 1.45	8.25^{ab}± 1.71	8.16^{bcd}± 1.59	8.16^{abc}± 1.59	8.18^{bc} ±1.39
1%BE	8.58^{ab} ±1.24	8.66^{ab} ± 1.37	8.50^{ab} ± 1.50	8.58^{abc} ± 0.90	8.08^{bc} ± 1.83	8.35^{abc} ±1.17
2% BE	7.50^{bc} ±1.78	7.41^{cd} ± 1.67	7.83^{bc} ± 1.89	7.41^{d} ± 1.62	7.58^{bc} ± 2.06	7.55^{cd} ±1.66
4% BE	8.00^{b} ± 1.27	7.75^{bc} ± 1.13	8.25^{ab} ± 1.35	7.75^{cd} ± 1.54	7.66^{bc} ± 1.55	7.78^{bcd} ±1.20

*BE= Bottle gourd leaf extract/ Means designated with the same letter are not significantly differences ($p > 0.05$)

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النشاط المضاد للميكروبات لمستخلص أوراق اليقطين وتأثيره على جودة الغذاء

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الخلاصة:

نظرا لوجود جدل كبير حول جوانب السلامة للمواد الحافظة الاصطناعية حيث أنها تعتبر مسؤولة عن العديد من الأمراض و المسببة للسرطان هناك اهتمام كبير بالمواد الحافظة للأطعمة الطبيعية لإستخدامها فى إطالة العمر الافتراضي للأغذية. لذلك ، تم فحص النشاط المضاد للميكروبات ، إجمالي الفينول والفلافونويد لمستخلص أوراق اليقطين. وتم الكشف الوصفي عن المستخلص اليثانولى٥٠% حيث وجد أنه يحتوي على (الفينولات ، الفلافونويد ، قلويدات ، جليكوسيدات ، تربين ، صابونين ، فلوباتانين ، راتنجات ، تانينات ، منشطات). أظهر المستخلص الإيثانولي بنسبة ٥٠٪ وجود نسبة كبيرة لمركبات الفينول والفلافونويد. كان إجمالي الفينول ٤٠٦ مجم مكافئ جاليك / جم وكان إجمالي الفلافونويد ٤٢,٦ مجم كيرسيتين / جرام وزن جاف. تمت دراسة النشاط المضاد للميكروبات باستخدام طريقة Agar well diffusion method وطريقة broth dilution method. أشارت النتائج إلى أن بكتريا *E. Coli* سالبة الجرام كانت أكثر حساسية من البكتيريا موجبة الجرام *S. aureus* و *B. subtilis* ضد مستخلص أوراق اليقطين، حيث كان قطر المنطقة المثبطة ٢٥ و ١٨ و ١٩ مم لتركيز ٥٠ مجم / بئر على التوالي. عند تحديد MIC و MLC ، أظهر اختبار MICs أن أقل تركيز كان ١٠ مجم / مل ضد *S. aureus* ، وكان تركيزًا متساويًا على *E. coli* و *B. subtilis* (20 مجم/ مل). على أساس هذه النتائج تمت إضافة مستخلص أوراق اليقطين إلى سجق اللحم البقرى بتركيزات مختلفة (١٪ ، ٢٪ ، ٤٪) وتم تخزينها لمدة ٩٠ يومًا عند ـ٢٠م لدراسة تأثيرها على جودة سجق اللحم البقري. أشارت النتائج إلى ثبات عالي ضد الأكسدة في السجق المعالج بمستخلص أوراق الزجاجة مقارنة بعينات المقارنة. كما لوحظ انخفاض في الحمل الميكروبى لمعاملات السجق المحتوية على مستخلص أوراق اليقطين مقارنة بالعينة الكنترول. أخيرًا ، كان من الواضح أن مستخلص أوراق القرع الزجاجي له دور في تقليل عدد الميكروبات ، فضلاً عن احتوائه على مضادات الأكسدة الطبيعية التي تطيل العمر الافتراضي للسجق.