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Synergic effect of natural herbal spices on the quality of refrigerated beef Kofta

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ABSTRACT

Spices such as paprika and frankincense extracts possess a potent antimicrobial potential against a wide range of foodborne microorganisms, which may shorten the shelf life and cause public health hazards. The current study aimed to investigate the synergic effect of paprika (PE) and frankincense extracts (FE) (1.0 and 2.0%) on the sensory and bacteriological quality of home-made beef kofta during refrigeration (4 ±1°C). Sensory evaluation revealed a significant enhancement in the color, odor, texture, and overall acceptability characteristics of beef kofta among all treated and control samples with an extension of organoleptic acceptability up to 12 days. Nevertheless, control samples showed spoilage within 6 days of cold storage. Additionally, the used extracts showed a significant antibacterial effect and appeared as considerable retardation in the APC, *E. coli* and *S. aureus* counts in the treated groups compared to the control group, which seemed to be maximized at the high concentration of the added substance. Aerobic plate mean counts (CFU/g) were 7.0, 6.6, 6.5, 5.8, and 4.8 log CFU/g for paprika extract (1 and 2%), frankincense extract (1 and 2%) and PE-FE mixed treatment at the 12th day of cold storage, respectively. On the other hand, frankincense extract showed higher levels of *S. aureus* inhibition than paprika extract. After all, the obtained results revealed that the paprika and frankincense extracts could be used as new ingredients that could control spoilage and/or foodborne pathogens and prolong the shelf life of kofta.

1. INTRODUCTION

High-grade raw meat and meat products are an important source of vitamins and minerals as well as a useful supply of protein (Geiker et al., 2021). However, it may be loaded with a wide range of foodborne pathogens due to unhygienic conditions during manufacture, packing, and storage that may pose threats to consumer's health (Hassan et al., 2019). Since meat products are regarded as reliable indicators of possible fecal and environmental contamination, the aerobic plate count, Enterobacteriaceae, and *S. aureus* may be used to qualify the bacteriological quality and hygienic procedures followed during meat production (Hamed et al., 2015). Foodborne diseases, e.g., Salmonella species, *E. coli*, and *S. aureus,* are the main causes of infection and death, particularly in underdeveloped nations (Todd, 2014). The presence of such bacteria in meat and meat products causes major health hazards since eating contaminated food is the primary source by which these illnesses spread (Zafar et al., 2016).

Customers are more interested in healthy food products because they are worried about the use of chemical additives, mostly because of their carcinogenic potential (Inguglia et al., 2023). In this context, meat industry search for natural additives is pushed by the risk associated with synthetic food additives combined with the consumer's preferences. Some herbal and medicinal plants extracts have been shown to have a potential use in food preservation, which is primarily attributed to their high concentrations of phenolic compounds and other antioxidants, which may delay oxidative processes and sensory changes in food (Ribeiro et al., 2019).

Plants are the source of a broad family of bioactive compounds known as phytochemicals, which may have anti-microbial characteristics (Abdullah et al., 2024). Herbs and spices, and their extracts, have been used as natural food preservatives, flavorings, and organoleptic enhancers because of its ability to prolong the shelf life and keeping quality through improvement of the color stability and reduction of lipid oxidation and microbiological growth (Kumar et al., 2023).

As a member of the capsicum family, paprika has been reported to contain carotenoids and capsaicin, which are potent antioxidants. Additionally, paprika significantly can reduce the bacterial contamination of food items because of its content of phenols, flavonoids, and capsaicinoid chemicals (Hu et al., 2021). Furthermore, different types of frankincense (*Boswellia* sp.) extracts have been used globally because of their recorded medicinal benefits and biological properties. According to Woolley *et al.* (2012), 68 % of the bioactive compounds in frankincense extract are α-pinene, which has a potent antibacterial action (Abbood et al., 2022).

As the level of contamination of beef and its products with different foodborne pathogens constitutes serious problems for consumers, the present study was conducted to evaluate the beneficial effect of paprika and frankincense aqueous extracts (1.0 and 2.0%) on the sensory and bacteriological quality of beef kofta during refrigeration storage.

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2. MATERIAL AND METHODS

All techniques used in this study were approved by Scientific Research Ethics Committee, Faculty of Veterinary Medicine, Benha University (BUFVTM02-10-24)

2.1. Preparation of paprika and frankincense extracts
Non-smoked powder of paprika (*Capsicum Annuum* L) and frankincense (*Boswellia* sp.) were obtained from Agricultural Research Center (ARC, Giza, Egypt), followed by preparation, extraction and dryness processes in Food Hygiene Department, Animal Health Research Institute (Benha branch), ARC, Egypt according to Dent *et al.* (2013). Paprika and frankincense resin powder were mixed with boiled distilled water for one hour, followed by sieving and dryness in an electric oven at 55 °C overnight. Working solutions (1.0 and 2.0%) of the used extracts were prepared by dissolving them in distilled water.

2.2. Preparation of beef kofta samples
A total of 2.25 kilograms of fresh minced meat were purchased from a butcher shop in Benha city, Qalubiya Governorate, Egypt, and stored in the icebox during transportation to the Animal Health Research Institute-Benha laboratory. Raw meat was mixed with 7.5 g of table salt and 2.5g of ground black pepper (Egyptian kofta recipe, 2022).

2.3. Experimental design
Under sterile conditions, samples were divided equally into six groups (125 g / each) representing control untreated (G1), treated samples with paprika extract (PE: 1% v/wt) and (2% v/wt) (G2 and G3), treated samples with frankincense extract (FE: 1% v/wt) and (2% v/wt) (G4 and G5), and treated minced meat with paprika extract (2% v/wt) plus frankincense extract (1% v/wt) (G6). The treated groups were mixed well and distributed by hand as beef kofta model. Control and treated groups were packed individually in a sterile polyethylene bag and kept in refrigeration storage at 4 ± 1 °C and were examined for their sensory and bacteriological quality every three days until the appearance of spoilage signs.

2.4. Sensory evaluation of examined meat samples
Meat samples were assessed for their sensory characteristics (color, odor, texture) following Mörlein (2019) in scores (1 to 5), where 1- represented the worst while 5- represented the excellent mark.

2.5. Bacteriological examination
2.5.1. Preparation of samples (ISO 6887-2: 2017):
Tenth fold serial dilutions were prepared on sterile peptone water (0.1%); from which the following parameters were examined:

2.5.2. Aerobic plate count "APC" according to ISO 4833-1 (2013)
This was performed on APC agar incubated at 30 ± 1 °C for 72 h. The Aerobic Plate Count (APC) per gram was calculated on plates containing 15-300 colonies and each count was recorded separately

2.5.3. Escherichia coli count "EC" according to ISO 16649-2 (2018)
This was performed on TBX agar incubated at 44 ± 1 °C for 24 h. Suspected colonies, which showed bluish-green colonies, were enumerated to obtain *E. coli* count /g.

2.4.4. Staphylococcus aureus count
This was performed by plating 0.1 ml on *Baird Parker agar*. Black colonies with hallow precipitation were counted and recorded following ISO 6888- 1 (2021).

2.3. Statistical analysis
The data obtained from triplicate trials was statistically treated by one-way ANOVA using SPSS software for Windows (Version 16). Results were presented as mean ± standard error. Duncan's post hoc analysis was used to analyze the data, with a p-value of 0.05 or less being regarded as statistically significant. Acceptability and shelf-life justification were referred to the Egyptian standard No. 2613-2 (EOS, 2008).

4. RESULTS

Table (1) showed that the treatment with PE and FE extended the acceptable sensory characters significantly ($P \leq 0.05$) when compared with a control group which showed spoilage signs (Noticeable changes in color, odor, and texture of the beef kofta samples) after the 6th day of chilling storage. Regarding the acceptability overall scoring, the treated kofta samples with PE-FE combination (G6) showed higher acceptability scores up to the 12th day of the experiment (2.7: acceptable) than the other treated samples; where G2 and G4 showed organoleptic acceptability up to the 9th days of the experiment with mean acceptability scores of 2.3 and 2.6, while G3 and G4 were still acceptable up to the 12th day of the experiment with acceptability scores of 2.3 and 2.4, respectively.

Table 1 Sensory profile of the examined groups in cold storage (4±1°C).

Groups	Tested parameter	G1	G2	G3	G4	G5	G6
Zero day	Color	4.8±0.1	4.8±0.16	4.8±0.16	4.7±0.16	4.6±0.16	4.7±0.16
	Odor	4.9±0.3	4.9±0.3	4.9±0.3	4.9±0.3	4.9±0.3	4.9±0.3
	Texture	4.9±0.1	4.8±0.4	4.8±0.4	4.8±0.4	4.8±0.4	4.8±0.4
	Overall	4.9±0.03^{a}	4.8±0.03^{a}	4.8±0.03^{a}	4.8±0.06^{a}	4.8±0.08^{a}	4.8±0.1^{a}
3rd day	Color	3.5±0.3	4.1±0.4	4.5±0.1	4.2±0.1	4.1±0.2	4.5±0.2
	odor	3.7±0.4	4.0±0.3	4.2±0.2	4.4±0.3	4.5±0.3	4.7±0.3
	Texture	3.5±0.2	3.6±0.3	4.0±0.3	4.3±0.2	4.5±0.4	4.4±0.4
	Overall	3.6±0.1^{b}	3.9±0.2^{b}	4.2±0.1^{ab}	4.3±0.1^{a}	4.3±0.1^{a}	4.5±0.1^{a}
6th day	Color	2.8±0.2	3.7±0.11	4.2±0.1	3.9±0.2	4.0±0.1	4.0±0.1
	odor	2.9±0.3	3.5±0.2	3.8±0.2	3.7±0.5	4.1±0.3	4.2±0.3
	Texture	2.5±0.2	3.2±0.3	3.5±0.3	3.5±0.3	3.9±0.2	4.0±0.2
	Overall	2.7±0.1^{d}	3.5±0.1^{c}	3.8±0.2^{b}	3.7±0.1^{b}	4.0±0.1^{a}	4.1±0.1^{a}
9th day	Color	0.9±0.01	2.5±0.2	3.7±0.1	2.8±0.3	3.5±0.2	3.7±0.2
	odor	1.2±0.2	2.3±0.2	3.0±0.3	2.5±0.2	3.2±0.3	3.5±0.3
	Texture	0.8±0.05	2.0±0.3	3.2±0.3	2.5±0.4	3.5±0.2	3.5±0.2
	Overall	1.0±0.1^{e}	2.3±0.1^{d}	3.3±0.2^{b}	2.6±0.1^{c}	3.4±0.1^{b}	3.6±0.1^{a}
12th day	Color	S.	1.5±0.2	2.3±0.1	1.7±0.3	2.5±0.2	2.8±0.2
	odor	S.	1.2±0.2	2.1±0.3	1.5±0.2	2.2±0.3	2.5±0.3
	Texture	S.	1.2±0.3	2.4±0.3	1.5±0.4	2.5±0.2	2.8±0.2
	Overall	S.	1.3±0.1^{d}	2.3±0.1^{b}	1.6±0.1^{c}	2.4±0.1^{b}	2.7±0.1^{a}

The values represent Mean ± SE of three replicates. Acceptability and shelf life was referred to the Egyptian standards (EOS, 2008) Means of overall score within the same row (a,b,c,d) followed by different superscript letters are significantly different ($P \leq 0.05$). Zero time: 30 min after inoculation. 4.0-5.0 very good (VG). 3.1-3.9 good (G). 2.1-3.0 Acceptable (A). 1.1-2.0 Unacceptable (U). 0.0-1.0 spoiled (S). G1: Control untreated beef kofta. G2: 1% PE treated samples. G3: 2% PE treated samples. G4: 1% FE treated samples. G5: 2% FE treated samples. G6: 1% FE+2% PE treated samples.

Referring to the recorded results in figs (1-3), significant retardation in the bacterial growth was recorded in the treated samples either with PE or FE compared with the control samples, which showed a concentration-dependent reduction manner; where higher concentrations had higher antibacterial activity; while, it worth noted that significant reductions were recorded in the treated group (G6) that was treated with FE2%+PE1% in the first 6 days of storage, followed by gradual raising up to the 12th day of storage. Additionally, the frankincense extract showed a higher antibacterial effect than the paprika extract. It is clear that the used extracts had a higher antibacterial effect against Gram-negative bacteria, although FE had a superior anti-staphylococcus effect than PE.

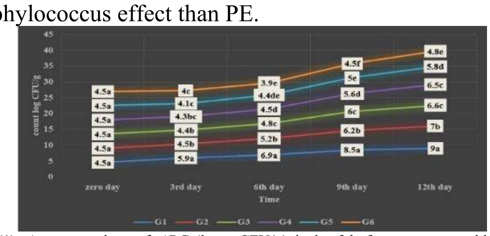

Fig. (1). Average values of *APC* (log_{10} CFU/g) in beef kofta groups at cold storage (4±1^{O}C). Means with different superscript letters (abcd) are significantly different ($p \leq 0.05$).

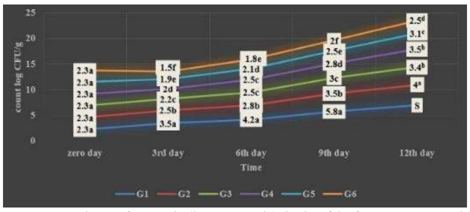

Fig. (2). Average values of *E. coli* (log_{10} CFU/g) in beef kofta groups at cold storage (4±1^{O}C). Means with different superscript letters (abcd) are significantly different ($p \leq 0.05$)

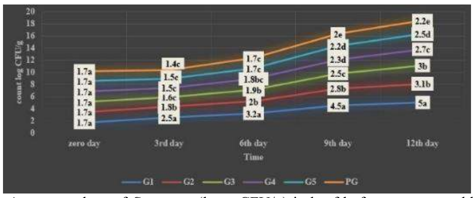

Fig. (3). Average values of *S. aureus* (log_{10} CFU/g) in beef kofta groups at cold storage (4±1^{O}C). Means with different superscript letters (abcd) are significantly different ($p \leq 0.05$).

4. DISCUSSION

Meat and meat products' shelf life may be affected by several variables, such as handling and storage conditions, that alter their color, flavor, odor, and nutritional value, compromising consumer approval (de Carvalho et al., 2019). Significant alterations in the sensory characteristics of meat products may lead to their rejection, especially after obvious discoloration that has a detrimental effect on customers' intentions to purchase as it is mainly associated with the product's safety and freshness (Agregán et al., 2019).

Referring to the Egyptian Standard (EOS, 2008), the shelf life of different chilled meat products should not exceed seven days and never be less than 4 days. On the other hand, oxidation processes are the primary cause of meat degradation (Domínguez et al., 2019); Therefore, the meat industry has thus searched for a way to prevent these oxidation events and preserve the quality of fresh foods through the application of either natural and\or synthetic antioxidants.

Food deterioration causes undesirable aromas, texture, and visual changes, which alter the food's quality and render it unfit for human ingestion. Microbial contaminations are characterized as sessile structures adhering to food processing, packing, and equipment surfaces that exhibit distinctive social and cooperative behaviors. They are primarily responsible for contaminating processed goods in the food business (Galié et al., 2018).

Because of its significance, sensory traits of meat and meat products, such as color, odor, texture, and overall acceptability, are the main factors in investigating the customer's approval and shelf life of meat (Fiorentini et al., 2020).

Regarding the results of the overall shelf life of the treated kofta samples (Table, 1), and about the Egyptian Standards, higher PE and FE (2.0%) and PE-FE combination treatments (G3, G5, G6, respectively) could extend the treated samples organoleptic acceptability up to twelve days, that means five days over the recommended keeping time by the Egyptian standard. The recorded results reflected the potent preservative efficacy of the used paprika and frankincense extracts. In addition, significant differences ($P \leq 0.05$) were noticed in the features of the treated samples since the 3rd day of storage. Also, the results revealed certain improvements in the characteristics of kofta in all treated samples in relation to the control group. These findings indicated that the natural extracts used can be added to kofta without any negative effects on consumer perception.

The currently obtained results matched with those of Jokanović *et al.* (2011) who investigated the effect of paprika extract (1.0%) on chicken breast bacteriological quality, Sabeeh (2023) who investigated the effect of frankincense extract (100-400 mg/ml conc.) on chicken meat samples, and Taha *et al.* (2024) who investigated the effect of frankincense extract (1.0%) on the sensory, bacteriological quality and shelf life of chicken drumsticks; they recorded significant enhancement in the treated meat products with paprika and frankincense extracts with considerable elongation in the shelf life of the treated samples; while Zaher *et al.* (2023) revealed non-significant difference among the treated samples with paprika extract and the control samples.

Enhanced sensory characteristics of the treated beef kofta may be linked with the bioactive contents of the used additives that play powerful antimicrobial and antioxidant effects (Riyad et al., 2020); which came in line with Lu *et al.* (2018) who recorded that paprika as a member of the Capsicum family was reported to include carotenoids and capsaicin compounds that have remarkable antioxidant activity which may be attributed to their phenolic and polyphenolic molecules that provided stable free radicals. Moreover, Celep *et al.* (2012) and Sabeeh (2023) reported that the phenolic compounds present in the frankincense extracts have been proven to be better antioxidants than vitamins E and C because they can chelate metal ions and be electron donors.

According to the principle, newly produced meat should be largely free of pathogenic microbes. However, if the animal is stressed or diseased, meat and meat products may become even more contaminated, especially through the gastrointestinal and skin microbes, which are the most often source of contamination for raw freshly dressed meats (Khalalfalla et al., 2017).

It has been demonstrated that the extracts of herbal spices and medicinal herbs, such as frankincense and paprika, have strong antibacterial properties against bacteria and germs that cause food spoiling (Al-Kharousi et al., 2023; Zaher et al., 2023).

The data shown in Figs. (1-3) revealed that the additives used have a strong antibacterial effect appeared as a considerable bacteriostatic bacterial growth inhibition in relation to control samples, which may be attributed to their bioactive compounds represented by phenols, flavonoids, and capsaicinoid compounds of paprika, and α-pinene of frankincense extracts; which have the ability of cell wall disruption and adverse its selective permeability (Hu et al., 2021; Abbood et al., 2022).

5. CONCLUSIONS

In conclusion, paprika and frankincense extracts can contribute to the development of safe and new ingredients that could be used to control spoilage microbes and foodborne pathogens in meat as well as prolong shelf life with a reduction of chemicals used.

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