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Vital Value of Coriander and Fennel Volatile Oils on Quality Beef Burger During Cryopreservation

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ABSTRACT

Effect of coriander and fennel volatile oils on quality and shelf life of beef burger were investigated. GC/MS was used to chemical components of coriander and fennel. 2-Decenal (E) accounts for 34.69 percent of total chemical components in coriander volatile oil and Estragole (60.59 percent) in fennel volatile oil. DPPH was used to evaluate antioxidant properties of volatile oils derived from fennel and coriander. Two essential oils were tested in this experiment. Fennel and coriander essential oils were examined for their antimicrobial properties against two mould strains and four bacterial strains that are most commonly found in meat and meat products. The two essential oils' inhibitory effects could be arranged as follows: Fennel > coriander essential oils. Obtained essential oils could be used in industrial food as organic antioxidants and antimicrobials. Beef burger was added with 400 ppm coriander and fennel volatile oil. Microbial examination such as total viable count, psychrotrophic counts, lipolytic and proteolytic bacteria counts, coliform group bacteria and yeasts and moulds counts and chemical evaluation namely thiobarbituric acid, total volatile basic nitrogen, and pH value at 4 day intervals throughout 16 day. Beef burger control treatments had least microbial quality and freshness properties loss during the storage period.

Keywords: Antimicrobial, Antioxidant, beef burger.

INTRODUCTION

There is a growing trend toward using natural products. Components derived from plants and animals, such as antimicrobials, antioxidants, colourants, and sweeteners, Mehdizadeh *et al.* (2020). The use of natural preservatives in foods has received widespread acclaim from customers, who increasingly seek natural and healthier products, free of synthetic additives (Sacchetti *et al.*, 2005). Customers are also used to the presence of herbs and spices that are frequently used to flavour and aromatize meats. As a result, essential oils (EOs) are a viable option for natural preservatives in fish and meat products, (Militello *et al.*, 2011).

They are perishable and contain a lot of critical nutrients, meat and meat products are especially prone to microbial degradation. If they are not adequately maintained, public health issues could develop. Recent advancements in natural and organic food ingredient systems have been employed to enhance the microbiological quality of meat and meat products. Numerous studies have demonstrated that naturally occurring antimicrobials originating from plants have antimicrobial activity in meat and meat products. Modern technology, however, has a number of ways to improve the sensory quality and microbiological stability of meat products that incorporate natural extracts and essential oils., Aminzare *et al.* (2016).

Beef burgers are a potentially dangerous product that can host pathogenic germs and allow them to thrive and create poisons if the storage temperature and time are not

managed. To avoid burger deterioration and maintain its safety, the meat industry has used different additives with antibacterial and antioxidant capabilities, Sayas-Barberá, *et al.* (2011) and Chaleshtori and Chaleshtori (2017).

Grinding Muscle structure is disrupted, and there is decreased stability as a result of this process. Meat rotting is caused by high quantities of bacteria. While aerobic conditions lead to moisture and protein content, Both of these factors can cause lipid and protein oxidation, which can compromise food safety, Rashidaie *et al.* (2019) and Tometri *et al.*(2020).

The culinary use of the annual herb coriander (*C. sativum L.*), a member of the Umbelliferae/Apiaceae family, dates back many centuries. Given that it is a source of EOs and aroma compounds with physiologically active ingredients that have antibacterial, antifungal, and antioxidant properties, *C. sativum* is helpful for preserving and flavouring food., Mandal and Mandal (2015).

For many years, the food industry has utilized fennel and fennel essential oil to give items like fish and meat dishes, baked products, liqueurs, pickles, and cheese a characteristic flavor, Pavela *et al.* (2016).

This paper studies that were carried out to find out the reasons for the ability of essential oils to prevent corruption and oxidation in meat products and knowledge of compounds responsible for this were few and insufficient. Therefore, this research was done in order to study chemical composition of coriander and fennel essential oils and add them at 400 ppm to improve quality

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of burgers in terms of microbiological quality fresh recipes during refrigeration.

MATERIALS AND METHODS

Material:

Raw material:

Coriander (*Coriandrum sativum* L.) and Fennel (*Foeniculum vulgare*) essential oils were obtained from (PHATRADE) Company, Obour City, Cairo, Egypt. Fresh beef meat was obtained from local market, Dokki Square, Giza Governorate, Egypt, and immediately transported in ice box to the laboratory, then carefully cut into fillets and finally weighed until use. Spices mixture, were obtained from the local market (Harraz) at Cairo Governorate, Egypt. Other ingredients such as texturized Soy were obtained from Food Technology Research Institute, Agricultural Research Center, Giza, Egypt. Salt, fresh eggs, bread crust, ground onion, foam plates and polyethylene film were obtained from the local market in Giza Governorate, Egypt.

Chemicals:

2, 2-Diphenyl-1-Picryl-Hydrazyl (DPPH), Tween 20 and 2-thiobarbituric acid all were obtained from sigma-Aldrich Chime, Steinheim, Germany. Hydrochloric acid, magnesium oxide, ethanol, sulfuric acid and sodium hydroxide all were obtained from El-Nasser pharmaceutical Chemical Co., Egypt.

Bacterial strains:

Faculty of Agriculture at Ain Shams University in Egypt's Cairo Microbiological Resources Center (Cairo MIRCEN) provided the bacterial strains. The test microorganisms were *Bacillus cereus* ATCC 33221, *Escherichia coli* ATCC 25922, *Staphylococcus aureus* ATCC 12600, *Salmonella typhimurium* ATCC14028. The plant pathogenic fungi strains were *Aspergillus niger* ATCC 16404 and *Aspergillus flavus* ATCC 10124, were assessed for experiments of antimicrobial activity.

Microbiological media:

Nutrient agar medium, violet red bile agar medium and chloramphenicol glucose yeast extract agar medium obtained from Biolife Company, Italy and the agent in Egypt Al-Badr Engineering Company.

Processing:

Preparation beef burger samples:

The control beef burger formula consisted of 63% lean meat, 12% rehydrated texturized soy it was rehydrated by water (at a ratio of 1:2 w / v) and minced through 3 mm plate twice, 7% fresh eggs, 5% fresh onion, 1.5% salt, 1.8% spices, 3.7% bread crust and 6% ice water, according to Feiner (2006) and modified by Nageb (2015). The mixture of spices was prepared according to Bahlol and Abd El-Aleem (2004). The other beef burger treatments were prepared by adding fennel and coriander essential oil at 400 ppm. All beef burger samples were packaged in a foam plates and stored at 4±1 °C up to 16 days. The samples were taken for analysis every 4 days periodically.

Methods:

Gas chromatography–mass spectrometry analysis (GC-MS)

The GC-MS system (Agilent Technologies) was equipped with gas chromatograph (7890B) and mass spectrometer detector (5977A) at Central Laboratories

Network, National Research Centre, Cairo, Egypt, Abd El-Motaleb et al.(2021).

Antioxidant activity of volatile oil (DPPH radical scavenging assay):

The free-radical scavenging activity of volatile oils was measured as a decrease in the absorbance of methanol solution of DPPH as reported by Sreejayan and Rao (1996). Scavenging activity was expressed as the percentage inhibition calculated using the following formula:

$$\% \text{ Anti-radical activity} = \frac{\text{Control Absorbance} - \text{Sample Absorbance}}{\text{Control Absorbance}} \times 100$$

Total volatile nitrogen and thiobarbituric acid value:

Total volatile nitrogen (T.V.N) with technique by Harold et al. (1987). T.V.N as mg/100g. The TBA as an indication for lipid oxidation was determined as reported by Kirk et al. (1991). T.B.A as mg malonaldehyde /kg sample.

pH value:

The pH of prepared sample was measured using a pH-meter (Jenway 3510 pH meter) with the technique by Fernández-López et al.(2006).

Microbiological examinations:

Antimicrobial activity of volatile oils:

The effect of different concentrations of volatile oils (1, 5, 10, 50 and 100%) on bacteria, yeasts and mold growth was studied using the paper-disc plate method, according to Loo et al. (1945) and Hassanen et al., (2015), by gauging the inhibitory zone's diameter.

Microbiological examination:

According to established procedures for total count, ten grams of each sample were put to a culture medium (1: 10-1 to 1: 10-6 and homogenized for two minutes in a stomacher), (ISO 4833:2013-1 protocol), yeast and mould count (ISO 21527-2:2008), coliforms group (ISO 21528-2:2004), proteolytic and lipolytic bacteria were counted according to Erkmen (2021).

Statistical analysis:

The obtained results were analyzed using comparison of variance (ANOVA) and least significant different (L.S.D) at the 5% level of probability; as reported by Snedecor and Cochran (1994).

RESULTS AND DISCUSSION

Fractionation and identification of coriander and fennel volatile oils:

Results observed that twenty volatile components were coriander volatile oil was extracted, separated, and identified. The identified components represented (97.81%) from the coriander volatile oil. 2-Decenal,(E), Decenal, Linalool, 2-Dodecenal, 2-Undecenal, (E)-Tetradec-2-enal and 4-Decenal, (E)- , were the chemical elements that were most prevalent in coriander volatile oil which 85.45% of the total identified chemical compounds. 2-Decenal, (E) (34.69%) of the total chemical compounds was the highest chemical compound in coriander volatile oil. However, 4-Decenal, (E)- (2.36%) of the total chemical compounds was the lowest one among the most prevalent chemical elements in volatile coriander oil. GC/MS characterizations of coriander volatile oil were presented in Table (1) and Fig. (2). These outcomes are consistent with Satyal and Setzer (2020) they reported that, coriander essential oil is dominated by linalool (62.2%-76.7%) with lesser quantities

of α -pinene (0.3%-11.4%), γ -terpinene (0.6%-11.6%), and camphor (0.0%-5.5%). Commercial cilantro essential oil is composed largely of (2E)-decenal (16.0%-46.6%), linalool (11.8%-29.8%), (2E)-decen-1-ol (0.0%-24.7%), decanal (5.2%- 18.7%), (2E)-dodecenal (4.1%-8.7%), and 1-decanol (0.0%-9.5%). The study conducted by Alsalman *et al.* (2021) showed that, the coriander leaves identified 29 chemicals totaling 94.83 % of the total composition. Other research by Laribi *et al.* (2015), showed that, Linalool (72.7%) was the predominant compound in coriander, followed by -terpinene (8.8%), -pinene (5.5%), camphor (3.7%), limonene (2.3%), geranyl acetate (1.9%), and p-cymene (1.5 percent). GC/MS characterizations of fennel volatile oil were presented in Table (1) and Fig. (1), five volatile components were identified from fennel volatile oil could be seen. The identified components represented (100%) from the fennel volatile oil. Estragole, Benzonitrile, 2-(4-methylphenyl) and I-Limonene were the most prevalent chemical compounds in fennel volatile oil, accounting for 93.84 percent of all chemical elements found. Estragole (60.59%) of the total chemical compounds was the highest chemical compound in fennel volatile oil. However, I-Limonene (10.93%) of the total chemical compounds was the lowest one among the most abundant chemical compounds in fennel volatile oil. These outcomes are consistent, Vieira *et al.* (2019) found that five volatile components were extracted and recognised from volatile fennel oil. The chemical main components of fennel were Trans-dihydrocarvone Anethole (79.62%), Fenchone (12.19%), Estragole cis-anethole (3.65%), Methyl nonadecanoate (2.89%) and 1R- α -pinene(1.65%). The study conducted by Bassyouni *et al.* (2019), revealed that chemical evaluation of the chemicals in fennel oil, Anethole was the primary substance, followed by limonene and fenchone (41.4 percent , 24.7 percent and 11.8 percent respectively).

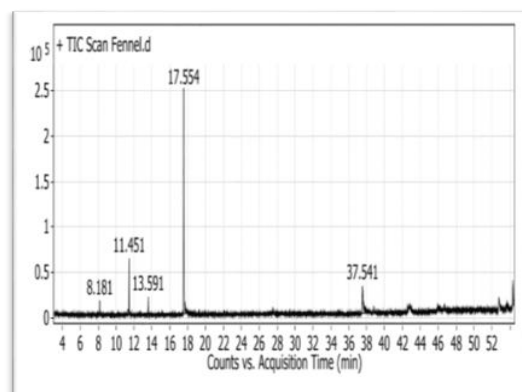


Fig. 1. Chemical components of fennel volatile oil by GC/mass.

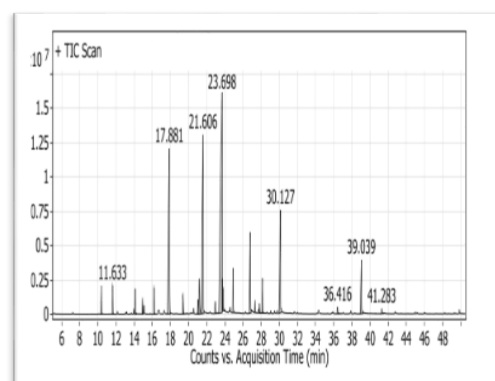


Fig. 2. Chemical components of coriander volatile oil by GC-mass.

Table 1. Essential volatile components of coriander and fennel volatile oils (%) identified by GC/MS.

Essential volatile components	Coriander volatile oils		Fennel volatile oil	
	RT*	Area %	RT*	Area %
Nonane	10.391	1.07	-	-
2,6,6-Trimethylbicyclo	11.633	1.11	-	-
Octanal	14.122	1.09	-	-
Benzene,methyl(1-methylethyl)	14.943	0.62	-	-
Gamma.-Terpinene	16.202	1.03	-	-
Linalool	17.881	15.26	-	-
2-Bomanone	19.397	0.88	-	-
4-Decenal, (E)-	21.186	2.36	-	-
Decanal	21.606	16.44	-	-
2-Decenal, (E)	23.698	34.69	-	-
Trans-2-Dodecen-1-ol	23.733	0.95	-	-
1-Decanol	23.797	0.93	-	-
Undecanal	24.91	1.90	-	-
2-Undecenal	26.799	4.92	-	-
Geranyl acetate	27.318	0.46	-	-
Dodecanal	28.134	1.50	-	-
2-Dodecenal	30.127	7.99	-	-
Tetradecanal	36.416	0.45	-	-
(E)-Tetradec-2-enal	39.039	3.79	-	-
(3S,3S)-3-Butyl-3a,4,5,6-tetrahydroisobenzofuran-1(3H)-one	41.283	0.37	-	-
Alpha.-pinene	-	-	8.181	2.37
I-Limonene	-	-	11.451	10.93
Fenchone	-	-	13.59	3.79
Estragole	-	-	17.554	60.59
Benzonitrile, 2-(4-methylphenyl)	-	-	37.541	22.32

*RT: Retention time.

Antimicrobial Activities of fennel and coriander essential volatile oil:

The antibacterial Activity of fennel and coriander essential oils against *E. coli* , *S. typhimurium* , *B. subtilis*, *S. aureus*, *A. flavus* and *A. niger* were examined their potential was evaluated by the diameter of inhibition zones were also detected. The advantages of antimicrobial essential oils measured by disk diffusion methods are shown in Fig. (3 and 4). Fennel volatile oil's antibacterial properties at various concentrations (1, 5, 10, 50 and 100%) against above-mentioned microorganisms strains, expressed as the diameters of inhibition zones (mm) are presented in Fig.(3) . From these results, it is clear that, the microbial spectra were decreased by increasing the concentration of fennel volatile oil for all microorganisms especially fungi strains which were highly sensitive for the lowest concentrations compared with yeasts and bacteria strains. It was clear that the antibacterial effects of fennel volatile oil were more pronounced on gram positive bacteria than gram negative bacteria. These outcomes are consistent with several studies which showed that gram negative bacteria had slightly higher resistance to essential oils than gram positive bacteria. This may be attributed to the variation in the cell wall structures of gram positive bacteria and gram negative bacteria. More specifically, gram negative bacteria have a high density lipopolysaccharide outer membrane that acts as a barrier to a variety of environmental chemicals, including antibiotics. (Palombo and Semple, 2001).It is evident that, different types of gram positive bacteria could be arranged descendingly according to the average diameters of

inhibition zones at different concentration of fennel volatile oil as follows: *Staphylococcus aureus* and *Bacillus subtilis* meanwhile, gram negative bacteria were as follows: *Escherichia coli*, and *Salmonella typhimurium*.

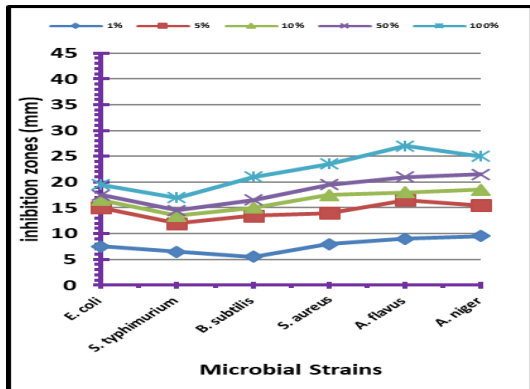


Fig. 3. Antimicrobial activity of fennel essential oil.

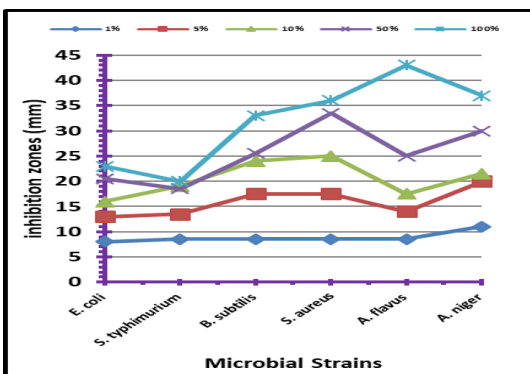


Fig. 4. Antimicrobial activity of coriander essential oil

The efficiency of fennel volatile oil against all microbial strains is related to its components such as estragole (60.59%) which was the major component of this oil and other chemical compounds such as benzonitrile, 2-(4-methylphenyl) (22.32%) and l-limonene (10.93%) which reported to exhibit antibacterial properties, Alsalman *et al.* (2021) and Ghasemian *et al.* (2020) discussed the remarkable antibacterial properties of fennel seed oil especially against *E. coli* and *S. aureus*. Mutlu-Ingok *et al.* (2021), Studies focusing on the antibacterial properties of fennel EO found that it had the lowest inhibition zones and the highest MIC values against *E. coli* O157:H7, *L. monocytogenes*, *S. Typhimurium*, *S.s aureus*, *S. albus*, *B. subtilis*, *P. aeruginosa*, and *S. dysenteriae*. Coriander volatile oil's antibacterial properties at various concentrations (1, 5, 10, 50 and 100%) against the above-mentioned microorganisms strains, expressed as the diameters of inhibition zones (mm) are presented in Fig. (4). From these results, it is show that, the microbial spectra were decreased by increasing the concentration of coriander volatile oil for all microorganisms especially fungi strains which were highly sensitive for the lowest concentrations compared with yeasts and bacteria strains. It was clear that the antibacterial effects of coriander volatile oil were more pronounced on gram positive bacteria than gram negative bacteria. The efficiency of coriander volatile oil against all microbial strains is related to its components such as 2-decenal, (E) (34.69%) which was the major component of this oil and other chemical compounds such as decanal(16.44 %) and Linalool(15.26%) which reported

to have antimicrobial properties, Mandal and Mandal (2015) revealed that *C. sativum* essential oil and extracts have promising antibacterial, antifungal, and anti-oxidative properties as diverse chemical components in different portions of the plant, which hence play a significant role in preserving the shelf-life of goods by avoiding their deterioration.. Zare-Shehneh *et al.* (2014) exhibited fungicidal activity against *P.lilacinum* and *A. niger* with MICs of 67.8 and 62.1 mg/mL, respectively, for *C. sativum* from coriander leaf extract.

Antioxidant activity of coriander and fennel volatile oils:

Coriander and fennel volatile oils ability to scavenge DPPH free radicals in comparison to conventional antioxidants (BHT and ascorbic acid) is graphically illustrated in Fig. (5). All studied samples exhibited good radical scavenging activity with different degrees. All of the examined samples' Capabilities of DPPH to scavenge free radicals are arranged descendingly as follows: BHT > coriander >fennel > ascorbic acid. Also, from the same results showed that the percentage of inhibition of DPPH free radical were increased by increasing concentration of coriander and fennel volatile oil and other antioxidants. BHT had the highest activity (12.47, 22.80, 48.59, 63.33, 72.91, and 81.55 % respectively).followed by coriander volatile oil exhibited higher scavenging activity on DPPH radicals (% inhibition 12.29, 22.13, 44.95, 61.29, 71.06 and 79.32 % at 5, 10, 25, 50, 75 and 100 µg/ml respectively) followed by fennel volatile oil had scavenging activity on DPPH radicals (% inhibition 12.07, 21.59, 43.03 , 60.77 , 70.82 and 77.88% at 5, 10, 25, 50, 75 and 100 µg/ml respectively) , than ascorbic acid (% inhibition10.33,19.08, 34.74, 58.45, 61.33, 68.41 % respectively), and slightly lower than BHT suggesting that the volatile oil of coriander and fennel possess strong electron donating capacity.

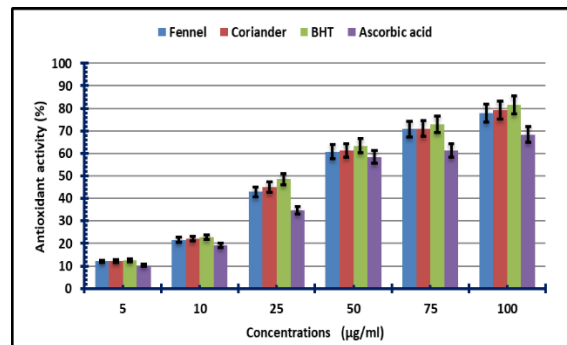


Fig. 5. DPPH-free radical scavenging activity (%) of fennel and coriander essential oil compared with those of BHT and ascorbic acid.

The antioxidant activities observed in coriander and fennel volatile oil could result from the phenolic compounds in the volatile oil or synergistic effect of two or more minor compounds found in it. Due to their different features that serve as radical scavengers, EOs from medicinal plants have displayed metabolic activities and attracted interest. (Lamiaa *et al.*, 2021).Abd ELMaged *et al.* 2012 and Misharina and Polshkov (2005), show that essential oils of laurel and coriander inhibit autoxidation of unstable components of fennel oil and, therefore, have high antioxidant activity. Most likely, other chemicals (with different functional groups and redox characteristics) and -

and - terpinenes stabilise the system of three essential oils and prevent the oxidation of reactive substances (e.g., monoterpene hydrocarbons and trans-anethol).

Microbiological quality attributes of treated beef burger with volatile oils prolonged cold storage at 4±1°C:

Total bacterial count (log cfu/g):

The total bacterial counts (TBC) of different beef burger samples as affected by addition of fennel and coriander at 400ppm were determined during cold storage period (4±1°C) results presented in Table (2). It was evident from the collected data that the initial TBC Among all preparations for beef burgers ranged from 3.83 and 3.75 log cfu/g at zero time, and the control sample had higher counts 3.87 log cfu/g as compared to alternative treatments. Furthermore, treating the samples with fennel and coriander 400ppm improved microbiological examination of the product by reducing the initial TBC by 1 log₁₀ cycle Also, from the same table, TBC of beef burger samples were affected by the cold storage period. The total bacterial counts were increased steadily in both of control and treated burger samples in compared to their initial counts with increasing storage period but at higher rates in control sample were detected, whereas their count were reached to 6.29 log cfu/g after 12 days of refrigeration at 4°C and, the samples were rejected for increasing the TBC to more than 6 log cfu/g. In addition, formulating of burger samples with fennel and coriander with 400ppm treatment can extend shelf-life of burger samples to 12 and 16 days respectively compared to 8 days for control. TBC were reached to 6.13 and 6.06 log cfu/g in burger samples with fennel and coriander, respectively at the end of cold storage. Samples were rejected because

they had counts that were greater than 6 log cfu/g. These outcomes align with those attained by (Šojić *et al.*, 2019), the product's microbiological stability would be affected by a 52-day refrigeration period with a 60 mg/kg sodium nitrite and 0.12 L/g coriander essential oil mixture, with total plate counts of 2.50 log CFU/g. Our results are consistent with research that found that using plant extracts rich in phenolic compounds increases meat and meat products expiration dates due to their higher antibacterial activity than synthetic antioxidants. (Fernandes *et al.*, 2016).

Psychrophilic bacterial counts (log cfu/g):

A key indicator of the microbiological quality of refrigerated foods is the assessment of the psychrophilic bacterial count (PBC), which is typically the main group among other bacteria groups. The psychrophilic bacterial counts (log cfu/g) of beef burger samples as affected by fennel and coriander 400ppm were determined during cold storage and the data given in Table (2). The findings revealed that the preliminary counts of PBC were 2.74 log cfu/g in control sample and the addition of fennel and coriander 400ppm had remarkable effect on the PBC of samples at zero time as their counts amounted to 1.60 and 1.54 log cfu/g, respectively. Furthermore, the treatment with volatile oils had reduced the initial count to 1 log cfu/g versus the control sample and other treatments at zero time, where its count reached to 5.14 log cfu/g at rejected time after 12 days. The addition of volatile oils at 400 ppm resulted in a reduction in the growth rate of psychrophilic bacteria versus the control sample. These outcomes concur with those attained by Osheba (2003), who found that psychrophilic bacteria count of refrigerated sausage samples were increased by storage period increment.

Table 2. Microbiological quality attributes of beef burger affected by volatile oils during cold storage at 4±1°C (log cfu/g).

Storage period (days)	Total bacterial counts log cfu/g			Psychrophilic bacterial counts log cfu/g			Lipolytic bacteria counts log cfu/g		
	Control	Fennel 400ppm	Coriander 400ppm	Control	Fennel 400ppm	Coriander 400ppm	Control	Fennel 400ppm	Coriander 400ppm
0 day	3.87	3.83	3.75	2.74	1.60	1.54	1.39	1.30	1.17
4 th day	4.84	3.95	3.90	3.68	2.48	2.43	1.97	1.87	1.77
8 th day	5.69	4.60	4.54	4.31	2.90	2.84	2.78	2.67	2.60
12 th day	6.29®	5.71	5.46	5.14®	3.78	3.76	3.99®	2.91	2.89
16 th day	®	6.13®	6.06®	®	4.70®	4.51®	®	3.66®	3.60®

Where: ®: Rejected (log cfu/g): Logarithm colony forming unit /gram.

Lipolytic and Proteolytic bacteria counts (log cfu/g):

The counts of lipolytic bacteria (log cfu/g) of beef burger samples as affected by fennel and coriander 400ppm were determined during cold storage and the data given in Table (2). These outcomes showed that the initial counts of lipolytic bacteria were 1.39 log cfu/g in control sample and the addition of fennel and coriander 400ppm had remarkable effect on the lipolytic bacteria of samples at zero time as their counts amounted to 1.30 and 1.17 log cfu/g, respectively. However, cold storage at 4±1° C induced gradual increases in the counts of lipolytic bacteria for all burger samples with higher rates in the control samples, where its count reached to 3.99 log cfu/g at rejected time after 12 days. The addition of volatile oils at 400 ppm resulted in a reduction in the growth rate of lipolytic bacteria versus the control sample. Coriander EOs can be used as a natural antioxidant in lipid-containing foods Ramadan *et al.* (2003). It was related to changes in lipids of hydrolysis by microorganisms and oxidation by

lipolytic enzymes, This outcome is consistent with those attained by El-Sherif (2006). The counts of proteolytic bacteria (log cfu/g) of beef burger samples as affected by fennel and coriander 400ppm were determined during cold storage and the data given in Table (3). The outcomes demonstrated that the preliminary counts of lipolytic bacteria were 1.65 log cfu/g in control sample and the addition of fennel and coriander 400ppm had remarkable effect on the lipolytic bacteria of samples at zero time as their counts amounted to 1.47 and 1.39 log cfu/g, respectively, where its count reached to 3.94 log cfu/g at rejected time after 12 days. The addition of volatile oils at 400 ppm resulted in a reduction in the growth rate of proteolytic bacteria versus the control sample.

Coliform group bacteria (log cfu/g):

Since coliforms are regarded as an indicator for the evaluation of the hygienic condition, the coliform group counts (CGC) were determined in all beef burger treatments under investigation as part of cold storage at

4±1°C and the results are shown in Table (3). No detected of coliform group in first storage due to hygiene and sanitation during processing samples in laboratory. The data indicated that, beef burger control counts were 1.60 and 2.02 log cfu/g after 8 and 12 days respectively. Whenever, the all treatment beef burger with fennel and coriander at 400 ppm during over all cold storage period were no detected as compared to beef burger control. It exceeded the maximal permissible limit of 2 log cfu / gm for the coliform bacterial count (Egyptian standard specifications, 2005).

Yeasts and molds counts (log cfu/g):

Total number of yeasts and molds of treated beef burger samples as affected by the addition of fennel and coriander 400ppm were presented in Table (3). From the data in this table it could be observed that, the initial TYMC sample was not detected in all treatment in zero time and after 4 days it could be observed that, TYMC in

control sample was 1.54 log cfu/g but not detected in beef burger treatments. This may be a result of high antifungal effect of volatile oils which attributed to phenolic compounds found in fennel and coriander. On the other hand, it could be observed that, TYMC were also affected by cold storage period at 4±1°C. By increasing the cold storage time, the TYMC were increased in all beef burger treatments. The highest increment in TYMC during cold storage was recorded for control sample their counts reached to 2.32 log cfu/g after 12 days of cold storage. While, the lowest increment in molds and fungi also count was recorded for the treated samples with coriander at 400 ppm 1.54 log cfu/g after 12 days of cold storage. The chemical structure of phenolic compounds influences their antimicrobial mechanism. The ability of the essential oils to penetrate and damage the intracellular membranes and cellular structure of fungi adds to the breakdown of mitochondrial cells. Singh *et al.* (2017).

Table 3. Microbiological quality attributes of beef burger affected by volatile oils during cold storage at 4±1°C (log cfu/g).

Storage period (days)	Proteolytic bacteria counts log cfu/g			Coliform group counts log cfu/g			Total yeasts and molds counts log cfu/g		
	Control	Fennel 400ppm	Coriander 400ppm	Control	Fennel 400ppm	Coriander 400ppm	Control	Fennel 400ppm	Coriander 400ppm
0 day	1.65	1.47	1.39	N.D	N.D	N.D	N.D	N.D	N.D
4 th day	1.92	1.87	1.84	N.D	N.D	N.D	1.54	N.D	N.D
8 th day	2.84	2.71	2.71	1.60	N.D	N.D	1.81	1.30	1.17
12 th day	3.94®	3.66	3.59	2.02®	N.D	N.D	2.32®	1.74	1.54
16 th day	®	4.55®	4.47®	®	N.D	N.D	®	2.01®	2.02®

Where: ®: Rejected ND: Not detected (log cfu/g): Logarithm colony forming unit /gram

Freshness properties of beef burger affected essential oils during cold storage at 4±1°C:

Total volatile basic nitrogen

Results given in Table (4) showed the effect of fennel and coriander volatile oils at 400 ppm on total volatile nitrogen of beef burger during cold storage at 4±1°C for 16 days. From statistical analysis of these data it could be observed that there were significant differences (p>0.05) in total volatile nitrogen between all beef burger treatments and untreated (control sample) at zero time, on the other hand significant differences were recorded between all above-mentioned samples during cold storage. Total volatile nitrogen of beef burger samples ranged from 9.71 to 9.84 mg/100g at zero time. These results are comparable to those of Bagheri *et al.* (2021), According to research, the largest concentration was found in the control group. of TVB-N at the conclusion of the storage period (47.65 mg/100 g) across all days of storage. The quantities of TVB-N reduced as preservative concentrations rose. All beef burger treatments' total volatile nitrogen values were significantly impacted by cold storage times as storage times increased. This could be a result of bacterial breakdown linked to the production of some alkaline substances like ammonia, which was confirmed by the rapid development of total volatile bases nitrogen, Valipour Kootenaie *et al.* (2017). Control sample had significantly higher total volatile nitrogen than beef burger at any time of storage (4 and 8 days). This may be due high antimicrobial effect of fennel and coriander volatile oils, Alsalman *et al.* (2021). At 12th day of cold storage, total volatile nitrogen the samples of beef burgers ranged from 16.35 to 22.05 mg N/100g. The highest total volatile nitrogen (22.05 mg N/100g) was recorded for control sample; while the lowest value (16.35 mg N/100g) was

observed for beef burger treated 400 ppm coriander volatile oil. Generally , total volatile nitrogen values of all samples were in the range of permissible level reported by the Egyptian standard specifications (2005), being not more than 20 mg N / 100 g, with exception of the control sample being 22.05 mg N/100 at 12th day .

Thiobarbituric acid:

Data presented in Table (4) showed the effect of volatile oils on thiobarbituric acid values of treated beef burger during cold storage at 4±1°C up to 16 days. From the statistical analysis of these data, nonsignificant difference (p≤0.05) in TBA values between different treatments either at zero time or throughout the storage periods was recorded. The thiobarbituric acid of all treatments were ranged from 0.278 to 0.282 mg malonaledhyde / kg sample at zero time. Also, from the same table, it could be observed that, TBA values of volatile oils treated beef burger were slightly lower than control. This reflected the higher antioxidant effect of volatile oils mixture. Thiobarbituric acid values of beef burger treatments were also significantly affected by storage periods. TBA of beef burger treatments significantly increased by the increase in storage time. The highest increase of TBA values was recorded for control beef burger. Meanwhile, the lowest of TBA values was found for beef burger with coriander at 400 ppm . TBA values will grow for beef burger treatments during storage indicated a continuous oxidation of lipid and consequently the production of oxidative by-products. These outcomes align with those that were noted by Bagheri *et al.* (2021) showed that, the control had the greatest levels of TBA (4.94 mg MDA/kg) at the conclusion of the storage period. The essential oil's components and its antioxidant qualities, which can stifle

free radicals and chelate iron ions, are to blame for the reduced levels of TBA in treatments including preservative. The TBA value of beef burgers treated with 400 ppm of fennel and coriander volatile oils must be within the range of allowable levels, according to the Egyptian standard specifications (2005), being not more than 0.9 mg malonaldehyde / kg sample.

pH value:

The pH value of various samples of beef burgers formulated with fennel and coriander at 400ppm were determined after processing and during storage period; results were presented in Table (4). From the results it can be noticed that, at zero time, there were no appreciable variations in pH between any of the beef burger treatments and control samples. Initial pH values for the 400ppm beef burger with fennel and coriander were 5.84, compared to 5.83 for the control sample. The pH values gradually declined in all samples during the course of the cold storage period, and there was a sizable difference between control and treated samples. The reduction rate of pH in control samples was significantly higher than those of

treated samples where reached to 4.71 after 12 days compared to 5.17 and 5.19 in treated samples with fennel and coriander at 400 ppm samples, respectively. While, the addition of fennel and coriander enhanced the pH stability depending on the concentration (400ppm) where the values reached to 5.48, 5.17 and 4.96 for beef burger samples with fennel and 5.51, 5.19 and 4.97 for beef burger samples with following 8, 12, and 16 days in the refrigerator, respectively. Similarly, Gahruie *et al.* (2017) reported that, After adding plant extracts, no discernible variations in the pH levels of beef patties and beef burgers were found. After 14 days of storage, the pH of the control burgers shifted more acidically than the treated burgers. This might be caused by microbial development and acid production, which lowers of pH during storage, Gallego *et al.* (2015) the acidity induced by bacterial activity on the increase in muscle glucose of the microbial metabolites produced by bacterial decay in beef patties may be the cause of pH variations during storage. However, during frozen and cold storage, the values gradually dropped. (EL-Mehy, 2018).

Table 4. Freshness properties of beef burger affected essential oils during cold storage at 4±1°C (Means ± SE).

Storage period	Total volatile basic nitrogen (mg /100 g sample)				Thiobarbituric acid (mg malonaldehyde/kg)				pH values			
	Control	Fennel 400ppm	Coriander 400ppm	L.S.D	Control	Fennel 400ppm	Coriander 400ppm	L.S.D	Control	Fennel 400ppm	Coriander 400ppm	L.S.D
0 day	9.84 ^{Da} ±0.058	9.71 ^{Eb} ±0.020	9.71 ^{Eb} ±0.027	0.108	0.289 ^{Da} ±0.001	0.278 ^{Ea} ±0.006	0.282 ^{Ea} ±0.001	0.011	5.83 ^{Aa} ±0.017	5.84 ^{Aa} ±0.006	5.84 ^{Aa} ±0.021	0.046
4 th day	12.56 ^{Ca} ±0.086	11.61 ^{Db} ±0.101	11.54 ^{Db} ±0.050	0.228	0.376 ^{Ca} ±0.012	0.326 ^{Db} ±0.008	0.307 ^{Db} ±0.002	0.025	5.54 ^{Bb} ±0.017	5.71 ^{Ba} ±0.017	5.68 ^{Ba} ±0.016	0.047
8 th day	16.47 ^{Ba} ±0.089	14.62 ^{Cb} ±0.047	14.41 ^{Cc} ±0.030	0.169	0.797 ^{Ba} ±0.002	0.565 ^{Cb} ±0.003	0.551 ^{Cc} ±0.005	0.010	5.18 ^{Cb} ±0.017	5.48 ^{Ca} ±0.017	5.51 ^{Ca} ±0.008	0.041
12 th day	22.05 ^{Aa} ±0.035 [®]	16.52 ^{Bb} ±0.106	16.35 ^{Bb} ±0.147	0.296	0.899 ^{Aa} ±0.008 [®]	0.730 ^{Bb} ±0.005	0.711 ^{Bc} ±0.004	0.018	4.71 ^{Db} ±0.029 [®]	5.17 ^{Da} ±0.014	5.19 ^{Ca} ±0.008	0.053
16 th day	[®]	19.37 ^{Aa} ±0.072 [®]	19.20 ^{Aa} ±0.098 [®]	0.239	[®]	0.907 ^{Aa} ±0.007 [®]	0.888 ^{Ab} ±0.008 [®]	0.022	[®]	4.96 ^{Da} ±0.005 [®]	4.97 ^{Da} ±0.008 [®]	0.020
L.S.D	0.197	0.213	0.233		0.021	0.018	0.014		0.058	0.037	0.038	

Where: Means with the same capital letter in the same columns are not significantly different, Means with the same small letter in the same rows are not significantly different, ®: Rejected L.S.D: Least significant differences at 0.05 levels.

CONCLUSION

In this study, volatile oils from coriander and fennel were used to keep beef burgers and increase refrigerator shelf life. Results showed that volatile oils increase fresh beef burgers shelf life from 8 days to 16 days. Beef burger samples treated with fennel 400ppm had adequate freshness and microbiological parameters, prolonged storage period while the other with coriander 400 ppm had the highest level of freshness qualities and microbial parameters among all samples. Essential oils may possess antimicrobial and antioxidant properties, and they can also help to improve safety of hamburgers by lowering oxidation products. Additionally, the essential. Additionally, the background information required for the use of these ingredients in a variety of meat and meat products can be offered.

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القيمة الحيوية لزيت الكزبرة والشمر العطري على جودة برجر اللحم البقري أثناء الحفظ بالتبريد
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قسم بحوث تكنولوجيا اللحوم والأسماك ، معهد بحوث تكنولوجيا الأغذية ، مركز البحوث الزراعية ، الجيزة ، مصر .
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المخلص

أجريت هذه الدراسة بهدف الاستفادة من زيوت الكزبرة والشمر العطرية كمضادة للأكسدة ولنمو الميكروبات. حيث تم دراسة التركيب الكيميائي باستخدام جهاز GC/MS وتم تقدير التأثير المضاد لنمو الميكروبات من خلال تقدير العد الكلي للبكتريا والفطريات موقع الدراسة وكذلك تقييم النشاط المضاد للأكسدة ومقارنتها بمضادات الأكسدة الصناعية بإجراء اختبار DPPH . وكذلك دراسة تأثير إضافة زيت الكزبرة والشمر العطري بتركيز 400 جزء في المليون على الجودة الميكروبيولوجية والجودة الكيميائية في برجر اللحم البقري بغرض إطالة مدة الحفظ أثناء التخزين بالتبريد على 4 ± 1 °م . وأوضحت النتائج أن المكون الرئيسي في زيت الكزبرة هو Decenal E-2 يمثل 34.69% بينما المكون الرئيسي في زيت الشمر هو Estragole يمثل 60.09% ، كذلك زيت الكزبرة له نشاط مضاد لنمو الميكروبات أعلى من زيت الشمر العطري . أظهرت النتائج أن برجر اللحم المضاد له زيت الكزبرة بتركيز 400 ppm كان أعلى في صفات الجودة الميكروبيولوجية والكيميائية يليه برجر اللحم المضاد له زيت الشمر بتركيز 400 ppm بالمقارنة بعينة الكنترول والتي فسدت في اليوم الثامن بينما عينات المعاملة بالزيوت العطرية لم تقسد حتى اليوم السادس عشر . وأخيرا يمكن التوصية باستخدام زيت الكزبرة والشمر العطري تجاريا في صناعة منتجات اللحوم مثل البرجر كمضادات لنمو الميكروبات ومضادات أكسدة طبيعية حيث أدت إلى زيادة فترة الصلاحية لهذا المنتج لتصل إلى اليوم السادس عشر مقارنة بمعاملة الكنترول على درجة التبريد.

الكلمات الدالة : مضادات لنمو الميكروبات ، مضادات الأكسدة ، و برجر اللحم .