

# Impact of Some Essential Oils on the Quality Aspect and Shelf Life of Meat

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## Abstract

Plant essential oils (EOs) serve as a «safe» alternative to chemical or synthetic antimicrobials and antioxidants to struggle with the food borne pathogens or spoilage organisms, inhibiting lipid oxidation and thus extend shelf life of ground beef application. In our study, we investigated the effectiveness of thyme oil (*Thymus vulgaris*) at concentrations (1%, 1.5% and 2%) and cinnamon oil (*Cinnamomum zeylanicum*) at concentrations (0.5%, 1% and 1.5%) as natural preservatives as well as their ability to increase the shelf life of minced meat and also their effects on chemical, microbial analysis and sensory properties of minced meat when been stored at 2°C for 12 days storage period. thyme and cinnamon oils at different concentrations effect on improving sensory properties of minced meat, act as antioxidant, antimicrobial agent and extend shelf life of minced meat for 6 days more than control samples, thyme>cinnamon. High concentration of oil more effective in meat quality and shelf life more than lower concentrations used. thyme and cinnamon oils at different concentrations effect on improving sensory properties of minced meat, act as antioxidant, antimicrobial agent and extend shelf life of minced meat for 6 days more than control samples, thyme>cinnamon. High concentration of each of oil more effective in meat quality and shelf life more than lower concentrations used.

**Keywords:** Essential oil; Thyme and cinnamon; Shelf life; Minced meat

## Introduction

Meat has long been known for its nutritive composition which could explain why it is being the first choice sources of animal protein for many people all over the world. Meat has a short shelf life of one day or less at ambient temperature (15°C-30°C) and a few days at refrigerated temperature (0°C-10°C) due to microbial spoilage of both pathogenic and non-pathogenic microorganisms and/or lipid oxidation [1]. Once the animal has been slaughtered, the meat is fabricated into whole sale or retail cuts. Trim and other cuts of meat are then further processed and ground. This increases the surface area of the meat which allows the increased adherence and growth of the bacteria [2-5]. Meat has a complex physical structure and chemical composition that is very susceptible to oxidation [6]. Essential oils are natural, volatile liquid, complex compounds characterized by a strong odor, rarely colored, soluble in lipid and organic solvents. It could be synthesized by all plant organs, i.e. buds, flowers, leaves, stems, twigs, seeds, fruits, root, wood or bark and are stored in secretory cells, cavities, canals, epidermis cells or gland ular trichomes [7]. Thyme (*Thymus* sp.) has much attention due to its high content and wide spectrum of phenolic compounds, antimicrobial and antioxidant properties and potential for use in meat and meat products [8-12]. The volatile oil components of thyme have also been known to have antimicrobial activity against different bacteria and fungi species [13].

The bark and leaves of Cinnamon are commonly used as spices in the home kitchen and their distilled essential oils or synthetic analogs are used as flavoring agents in the food and beverage industry [14]. The major compounds in the essential oil of *Cinnamomum zeylanicum* are cinnamaldehyde, benzaldehyde, limonene, linalool and eugenol [15]. PH values showed the effect of microbial load on minced meat as activation effect of microbial load causes protein hydrolysis with appearance of alkyl group, TBARs values is routinely used as an index of lipid oxidation in minced meat in store and TVN values attributed to the breakdown of proteins as a result of activity of microbial strains and proteolytic enzymes.

The objective of the present study was the antioxidant as well as the antimicrobial effectiveness of both thyme and cinnamon oils on quality of fresh minced meat during storage at 2°C for 12 days [15-20].

## Materials and Methods

### Minced meat

Three and half Kg of fresh meat were purchased immediately after slaughter from butcher shops in Kalubiya Governorate and was directly minced and packed in clean polyethylene bags. Then they were transported directly in insulated and iced containers to Microbiology Laboratory (Animal Health Research Institute), Benha, Al-Kalubiya, Egypt [21-26].

### Essential oils

The ready-made herbal oils of thyme (*Thymus vulgaris*) and cinnamon (*Cinnamomum zeylanicum*) used in this study at pure state, free from preservatives or antioxidant substance, were purchased from El-Hawag Company, for extracting best kinds of natural seed's oils, at Badr city-Cairo-Egypt. These oils were stored in amber colored bottles at 4°C until use [27-35].

### Preparation of minced meat samples

Minced meat sample are divided to two minor groups of treated and untreated (control) ones. The treated groups were divided in to

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6 groups (300 g of each) that mixed with thyme at concentrations ((T<sub>1</sub>)1%, (T<sub>2</sub>) 1.5% and (T<sub>3</sub>) 2%) and cinnamon at concentrations ((T<sub>4</sub>)0.5%, (T<sub>5</sub>) 1% and (T<sub>6</sub>) 1.5%). All groups were stored at 2°C and examined periodically every 3 days for chemically, microbiologically and sensory. The experiment was conducted in triplicate for 12 days of storage [35-44].

#### Chemical analysis:

- Measurement of pH, according to ISO (1979) [31].
- Measurement of thiobarbituric acid (TBA) value, the thiobarbituric acid (TBA) assay was carried out according to the procedure of Vyncke (1970) [45-53].
- Measurement of total volatile nitrogen (TVN) the total volatile nitrogen (TVN) measurement was carried out according to the procedure of FAO, (1980) [18].

**Microbiological analysis:** Preparation of sample, from each group, 10g of meat were taken randomly and aseptically using sterile forceps and scissors, the removed samples were placed in a sterile homogenizer flask contained 45ml of (0.1%) peptone water. The content of each flask were homogenized at 14000 rpm for 2.5 minutes for obtaining a dilution of 10<sup>-1</sup>, from which 1ml was transferred with a sterile pipette to a sterile test tube containing 9 ml of (0.1) peptone water, from which a decimal serial dilution were prepared in a sequential manner up to 10<sup>-1</sup>, to cover all expected range of samples contamination. For microbial count, colonies were counted and recorded in colony forming units per gram (cfu/g) of meat sampled using the formula: cfu/g=level of dilution plated × number of colonies counted/volume plated. These were further expressed in mean colony forming units per gram (mean cfu/g) and converted to log<sup>10</sup> base values (log<sup>10</sup> cfu/g).

- Aerobic Plate Counts (APC), according to ICMSF (1996) [29].
- (Enterobacteriaceae Count (EBC), according to ICMSF (1996) [29].
- Coliform count, according to APHA (1985) [3].

**Sensory evaluation:** It was carried out at the third day of the experiment according to Pearson and Tauber (1984) [40]. A 9 point hedonic scale (9=Excellent, 8=Very very good, 7=Very good, 6=Good, 5=Medium, 4=Fair, 3=Poor, 2=Very poor, 1=Very very poor) was used for the evaluation of the overall acceptability.

**Statistical analysis:** ANOVA was carried out on data of the sensory, chemical and microbiological evaluations. Data are expressed as mean±SE [24].

## Results, Discussion and Conclusion

### Effect of different concentrations of thyme (*T. vulgaris*) and cinnamon (*C. zeylanicum*) oils on pH characters of minced meat during cold storage at 2°C

Table 1 showed that the pH values of untreated and treated minced meat samples were, at control samples the pH values were 5.65, 5.95, 6.10, 6.33 and 6.65 at 0, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. By using thyme oil the pH values at concentration 1% (T1) were 5.65, 5.82, 5.94, 6.01 and 6.35; concentration 1.5% (T2) pH values were 5.65, 5.76, 5.81, 5.88 and 6.22 and concentration 2% (T3) pH values were 5.65, 5.66, 5.71, 5.78 and 6.03, at 0, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. The obtained results in Table 1 showed that samples treated with thyme essential oil were lowering in pH values than pH values of control samples during different periods of analysis these may be due to activation effect of thyme oil as antimicrobial agent causing protein hydrolysis with appearance of alkyl groups. Also, by increasing concentration of thyme to 2% pH values scored highest effect in lowering pH value than lower concentration of thyme oil (1%). These results were nearly similar to results obtained by Salem et al. [46], who reported that minced beef mixed by thyme essential oil during cold storage at 4°C with different concentrations of thyme (1.5%, 1% and 0.5%) showed significant effect in lowering of pH values than those of untreated samples, also the highest oil concentration was more effective in lowering PH values than lower oil concentration [46]. Also agreement with, Kassem et al. [35], who stated that pH values of burger samples treated with high concentration of thyme (1.2%) were significantly low ( $p < 0.05$ ) comparing with control samples after 4 days of storage at refrigeration temperature, might be due to the antimicrobial effect of phenolic compound in herb oil [35]. By using of cinnamon (*C. zeylanicum*) oil the pH values at concentration 0.5% (T4) were 5.65, 5.80, 5.86, 5.94 and 6.42; concentration 1% (T5) values were 5.65, 5.76, 5.79, 5.85 and 6.26 and concentration 1.5% (T6) pH values were 5.65, 5.72, 5.74, 5.8 and 6.09, at 0, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. The obtained results indicated that PH values at different concentrations of cinnamon showed lowering pH values than control samples, also by increasing of cinnamon (*C. zeylanicum*) oil concentrations results showed more lowering in pH values than values from lower cinnamon concentrations, which may be due to antimicrobial load. These agreement with Kesavan et al. [35], who stated that pH values of raw beef meat samples measured during storage at 4±1°C for 15 days had higher pH value at control samples than samples treated by cinnamon oil, this increase in pH reflects the degree of meat spoilage through protein breakdown for the production of free amino acids, leading to the formation of NH<sub>3</sub> and amines, compounds of alkaline reactions [36]. Also, these results agree

Storage days	pH values ± SE				
	0	3	6	9	12
Control	5.65 ± 0.02 <sup>a</sup>	5.95 ± 0.01 <sup>a</sup>	6.10 ± 0.02 <sup>ab</sup>	6.33 ± 0.02 <sup>b</sup>	6.65 ± 0.03 <sup>c</sup>
Thyme 1% (T1)	5.65 ± 0.02 <sup>a</sup>	5.82 ± 0.01 <sup>a</sup>	5.94 ± 0.01 <sup>ab</sup>	6.01 ± 0.01 <sup>b</sup>	6.35 ± 0.01 <sup>c</sup>
Thyme 1.5% (T2)	5.65 ± 0.02 <sup>a</sup>	5.76 ± 0.03 <sup>a</sup>	5.81 ± 0.01 <sup>ab</sup>	5.88 ± 0.01 <sup>b</sup>	6.22 ± 0.01 <sup>c</sup>
Thyme 2% (T3)	5.65 ± 0.02 <sup>a</sup>	5.66 ± 0.04 <sup>a</sup>	5.71 ± 0.01 <sup>ab</sup>	5.78 ± 0.01 <sup>b</sup>	6.03 ± 0.01 <sup>c</sup>
<i>C. zeylanicum</i> 0.5% (T4)	5.65 ± 0.02 <sup>a</sup>	5.80 ± 0.01 <sup>a</sup>	5.86 ± 0.02 <sup>ab</sup>	5.94 ± 0.01 <sup>b</sup>	6.42 ± 0.01 <sup>c</sup>
<i>C. zeylanicum</i> 1% (T5)	5.65 ± 0.02 <sup>a</sup>	5.76 ± 0.03 <sup>a</sup>	5.79 ± 0.01 <sup>ab</sup>	5.85 ± 0.01 <sup>b</sup>	6.26 ± 0.01 <sup>c</sup>
<i>C. zeylanicum</i> 1.5% (T6)	5.65 ± 0.02 <sup>a</sup>	5.72 ± 0.02 <sup>a</sup>	5.74 ± 0.01 <sup>ab</sup>	5.80 ± 0.01 <sup>b</sup>	6.09 ± 0.01 <sup>c</sup>

Values with different letters within the same row differed significantly at ( $P < 0.05$ )

**Table 1:** Mean pH values of untreated and treated minced meat samples during storage period at 2°C (Mean ± SE).

with that obtained by Brilliana et al. [10] they stated that the addition of cinnamon essential oil on ground beef can lowering pH values of fresh ground beef during refrigerated storage compared to the control sample, these results because of the active components of the cinnamon essential oil that are cinnamaldehyde, linalool and eugenol [10].

### Thiobarbituric acid reactive substances (TBARs) values of minced meat with thyme (*T. vulgaris*) and cinnamon (*C. zeylanicum*) essential oils during cold storage at 2°C

TBA values routinely used as an index of lipid oxidation in meat products in store and the rancid flavor is initially detected in meat products when TBA values reach to 0.9 (EOS, 2005). Table 2 showed that at control samples TBA values were 0.40, 0.68, 0.92, 1.19 and 1.45 at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. Which appear rancid flavor at 6<sup>th</sup>, 9<sup>th</sup> and 12<sup>th</sup> day of storage period? By using thyme oil at concentration 1% (T1) the TBA values were 0.40, 0.58, 0.85, 0.98 and 1.77; using concentration 1.5% (T2) TBA values were 0.40, 0.53, 0.78, 0.91 and 0.99 and using concentration 2% (T3) TBA values were 0.39, 0.49, 0.60, 0.72 and 0.83, at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day, respectively. We obtained that treated samples with thyme oil at different concentrations showed lowering in TBARS values especially at 6<sup>th</sup>, 9<sup>th</sup> and 12<sup>th</sup> day of storage period than control samples and increasing of thyme essential oil concentration showed decreasing the TBARS values than lower thyme concentration used. Such findings may be attributed to the high antioxidant effect of thyme essential oil, which is related to the scavenger nature of its flavonoids and phenolic content as apigenin, naringenin, luteolin, thymonin, carvacrol and thymol [48]. Also these results were similar to that was obtained by Kassem et al [35], who mentioned that TBA values of minced meat were obtained over 9 days of storage at 4°C and TBA values of control showed a rapid increase with increasing storage time, TBA values of thyme treatment samples on day 0 were significantly lower (P<0.05) than those for the control sample [36,53-55]. Moreover, treatment samples had significantly lower (P<0.05) TBA values than control at each day of testing throughout sixth and ninth days of the storage period. Also, Ghaderi et al. [21], who mentioned that changes in TBARS value of beef burgers with different formulations during 8 days of refrigerated storage the amount of TBARS increased suddenly over time in control burgers and significant differences were evident among treated samples with thyme oil as compared to control samples [21]. By using of *C. zeylanicum* oil at concentration 0.5% (T4) TBA values were 0.40, 0.62, 0.76, 0.82 and 0.96; using concentration 1% (T5) TBA values were 0.40, 0.53, 0.71, 0.82 and 0.92 and using concentration 1.5% (T6) TBA values were 0.40, 0.50, 0.60, 0.77 and 0.88 at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. All treated samples by cinnamon oil showed lowering in TBARS values than control sample especially at 6<sup>th</sup>, 9<sup>th</sup> and 12<sup>th</sup> day of storage period also increasing concentration of *C. zeylanicum* essential oil showed decreasing in the

TBARS values than lower concentration of the same oil. This result agrees with results [41-50]. These results agree with Kesavan et al. [36] who recorded that the effect of cinnamon oil on TBARS values of raw beef samples during storage at 4°C for 15 days [36]. TBARS values of all treated samples were considerably lower (P<0.05) than those of control samples during storage, indicating high protection by the spice extracts against lipid oxidation in raw beef meat samples. These agree with, Irfiana et al. [30] they mentioned that cinnamon oil showed decreasing at TBA values of vacuum-packed ground beef during refrigerated storage (at 0, 4, 8, 12 and 16 day) when compared with TBA values of control samples, as cinnamon oil contains active compounds such as polyphenols, cinnamaldehyde leading to antibacterial action and antioxidant of meat [30].

### Total volatile nitrogen (TVN) values of minced meat formulated with thyme (*T. vulgaris*) and cinnamon (*C. zeylanicum*) essential oils during cold storage at 2°C

Total volatile basic nitrogen especially used as index of raw minced meat especially when TVBN reach 20 mg/100 mg raw minced meat [16]. Table 3 showed that at control sample TVN values were 12.46, 16.10, 17.45, 20.51 and 24.60, at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. By using thyme oil at concentration 1% (T1) TVN values scored 12.36, 13.63, 15.49, 319.56 and 21.66; at concentration 1.5% (T2) TVN values were 12.35, 13.52, 14.41, 18.59 and 20.70 and at concentration 2% (T3) TVN values were 12.25, 12.43, 12.56, 13.63 and 15.75, at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. From the obtained result which appeared that the treatment samples with thyme oil at different concentrations showed decreasing in TVN values than results were recorded by control samples especially at 9<sup>th</sup> and 12<sup>th</sup> day, also by increasing concentration of thyme (2%) was more effective in decreasing TVN values than lower concentration of thyme (1%), the degree of autolysis and bacterial proteolysis in meat can be measured as TV which actually determines the quantity of amino acids, i.e. tyrosine and tryptophan present in an extract of meat. Nearly agree with the results [46] who mentioned that preserved minced beef at 4°C during 6 days of storage period recorded the highest rate of TVN values at control samples, also the treated sample with 1.5% thyme oil was more effective in decreasing TVN values than sample treated with 0.5% thyme oil, due to the role of thyme oil on microbial population and bacterial growth as antimicrobial agents. By using cinnamon oil at concentration 0.5% (T4) TVN values were 12.38, 14.01, 16.24, 20.22 and 21.79; at concentration 1% (T5) TVN values were 12.35, 13.99, 15.26, 18.36 and 20.14 and at concentration 1.5% (T6) TVN values were 12.28, 12.46, 13.67, 14.72 and 17.79, at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. From the obtained result which appeared that the treatment samples with cinnamon oil at different concentrations caused decreasing in TVN values than control samples especially at 9<sup>th</sup>

Storage days	TBARs mg Malonaldehyde/Kg ± SE				
	0	3	6	9	12
Control	0.40 ± 0.01 <sup>a</sup>	0.68 ± 0.02 <sup>b</sup>	0.92 ± 0.01 <sup>c</sup>	1.19 ± 0.01 <sup>c</sup>	1.45 ± 0.01 <sup>d</sup>
Thyme 1% (T1)	0.40 ± 0.01 <sup>a</sup>	0.58 ± 0.01 <sup>ab</sup>	0.85 ± 0.01 <sup>b</sup>	0.98 ± 0.01 <sup>c</sup>	1.17 ± 0.01 <sup>d</sup>
Thyme 1.5% (T2)	0.40 ± 0.01 <sup>a</sup>	0.53 ± 0.02 <sup>ab</sup>	0.78 ± 0.01 <sup>b</sup>	0.91 ± 0.02 <sup>c</sup>	0.99 ± 0.01 <sup>c</sup>
Thyme 2% (T3)	0.39 ± 0.01 <sup>a</sup>	0.49 ± 0.01 <sup>a</sup>	0.60 ± 0.00 <sup>b</sup>	0.72 ± 0.01 <sup>c</sup>	0.83 ± 0.02 <sup>c</sup>
<i>C. zeylanicum</i> 0.5% (T4)	0.40 ± 0.02 <sup>a</sup>	0.62 ± 0.01 <sup>ab</sup>	0.76 ± 0.01 <sup>b</sup>	0.82 ± 0.01 <sup>c</sup>	0.96 ± 0.02 <sup>d</sup>
<i>C. zeylanicum</i> 1% (T5)	0.40 ± 0.02 <sup>a</sup>	0.53 ± 0.02 <sup>ab</sup>	0.71 ± 0.01 <sup>b</sup>	0.82 ± 0.02 <sup>c</sup>	0.92 ± 0.01 <sup>c</sup>
<i>C. zeylanicum</i> 1.5% (T6)	0.40 ± 0.01 <sup>a</sup>	0.50 ± 0.01 <sup>a</sup>	0.60 ± 0.00 <sup>b</sup>	0.77 ± 0.01 <sup>c</sup>	0.88 ± 0.02 <sup>c</sup>

Values with different letters within the same row differed significantly at (P<0.05)

**Table 2:** Thiobarbituric acid reactive substances (TBARs) values of untreated and treated minced meat samples during storage period at 2°C (Mean ± SE).

and 12<sup>th</sup> day, also by increase concentration of cinnamon (1.5%) was more effective in decreasing TVN values than lower concentration of cinnamon (0.5%) which was used. These results were agree with results were recorded by [41,50-52] who recorded that lowering TVN values of meat when treated by cinnamon oil than control samples was due to cinnamaldehyde and antimicrobial effect.

### Effect of different concentrations of thyme (*T. vulgaris*) and cinnamon (*C. zeylanicum*) essential oilson microbial growth in minced meat during cold storage at 2°C

Table 4 showed that the APC values of control minced meat samples were  $5.19 \times 10^3$ ,  $6.11 \times 10^4$ ,  $4.15 \times 10^6$ ,  $2.25 \times 10^7$  and  $2.33 \times 10^8$ , at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. By using thyme oil at concentration 1% (T1) APC values were  $5.19 \times 10^3$ ,  $5.63 \times 10^4$ ,  $6.25 \times 10^6$ ,  $1.15 \times 10^7$  and  $5.10 \times 10^8$ ; at concentration 1.5% (T2) APC values were  $5.18 \times 10^3$ ,  $4.53 \times 10^4$ ,  $5.11 \times 10^5$ ,  $4.22 \times 10^6$  and  $3.13 \times 10^7$  and at concentration 2% (T3) APC scored  $5.17 \times 10^3$ ,  $3.73 \times 10^3$ ,  $3.15 \times 10^5$ ,  $5.89 \times 10^5$  and  $2.15 \times 10^6$ , at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. From the obtained results, samples treated by different concentrations of thyme showed decreasing count of aerobic plate microorganisms than per miscible limit which is  $10^6$ cfu/g according to (EOS,2005) when compared to control samples especially at 9<sup>th</sup> and 12<sup>th</sup> day, also high concentration of thyme more effective in decreasing this count than lower concentration. These

results were similar to results were obtained by [46-52] who mentioned that the antibacterial effect of thyme oil in refrigerated minced beef as decreasing APC than control sample. High concentration of thyme oil (1.5%) was more effective on decreasing APC values than values of lower concentration of the same oil (0.5%). By using cinnamon oil at concentration 0.5% (T4) APC values were  $5.19 \times 10^3$ ,  $8.53 \times 10^4$ ,  $6.12 \times 10^6$ ,  $5.15 \times 10^7$  and  $5.14 \times 10^8$ . At concentration 1% (T5) APC values were  $5.19 \times 10^3$ ,  $7.93 \times 10^4$ ,  $6.10 \times 10^5$ ,  $5.15 \times 10^6$  and  $4.32 \times 10^7$  and at concentration 1.5% (T6) APC values were  $5.18 \times 10^3$ ,  $5.12 \times 10^4$ ,  $4.14 \times 10^5$ ,  $6.15 \times 10^5$  and  $2.32 \times 10^6$ , at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. From the obtained results, samples treated with different concentrations of cinnamon showed decreasing count of aerobic plate microorganism when compared to control samples especially at 9<sup>th</sup> and 12<sup>th</sup> day, also high concentration of cinnamon (1.5%) was more effective in decreasing this count than lower concentration of cinnamon oil (0.5%). These results were agree with results were obtained by Krishnamoorthy et al. [38], they stated that cinnamon (*C. zeylanicum*) oil consists of two chemical volatile phenols and poly phenols. Cinnamon showed as antibacterial properties against gram positive and gram negative bacteria [38]. Also, agree with Irfiana et al. [30] who recorded that cinnamon oil showed decreasing count of total plat count microorganisms of vacuum-packed ground beef during refrigerated storage (at 0, 4, 8, 12 and 16 day) when compared with count of control samples, due to cinnamon

Storage days	TVBN values				
	0	3	6	9	12
Control	12.46 ± 0.12 <sup>a</sup>	16.10 ± 0.36 <sup>b</sup>	17.45 ± 0.24 <sup>c</sup>	20.51 ± 0.67 <sup>d</sup>	24.60 ± 0.36 <sup>e</sup>
Thyme 1% (T1)	12.36 ± 0.32 <sup>a</sup>	13.63 ± 0.16 <sup>a</sup>	15.49 ± 0.11 <sup>b</sup>	19.56 ± 0.46 <sup>c</sup>	21.66 ± 0.27 <sup>d</sup>
Thyme 1.5% (T2)	12.35 ± 0.22 <sup>a</sup>	13.52 ± 0.03 <sup>a</sup>	14.41 ± 0.34 <sup>b</sup>	18.59 ± 0.42 <sup>c</sup>	20.70 ± 0.57 <sup>d</sup>
Thyme 2% (T3)	12.25 ± 0.34 <sup>a</sup>	12.43 ± 0.20 <sup>a</sup>	12.56 ± 0.24 <sup>a</sup>	13.63 ± 0.57 <sup>b</sup>	15.76 ± 0.24 <sup>c</sup>
<i>C. zeylanicum</i> 0.5% (T4)	12.38 ± 0.32 <sup>a</sup>	14.01 ± 0.34 <sup>ab</sup>	16.24 ± 0.32 <sup>b</sup>	20.22 ± 0.32 <sup>c</sup>	21.79 ± 0.21 <sup>d</sup>
<i>C. zeylanicum</i> 1% (T5)	12.35 ± 0.22 <sup>a</sup>	13.99 ± 0.16 <sup>ab</sup>	15.26 ± 0.22 <sup>b</sup>	18.36 ± 0.32 <sup>c</sup>	20.14 ± 0.45 <sup>d</sup>
<i>C. zeylanicum</i> 1.5% (T6)	12.28 ± 0.15 <sup>a</sup>	12.46 ± 0.23 <sup>a</sup>	13.67 ± 0.42 <sup>b</sup>	14.72 ± 0.43 <sup>b</sup>	17.79 ± 0.21 <sup>c</sup>

Values with different letters within the same row differed significantly at (P<0.05)

Table 3: Total Volatile Basic Nitrogen (TVBN) values of untreated and treated minced meat samples during storage period at 2°C (Mean ± SE).

Storage days	(APC) log cfu /g ± SE				
	0	3	6	9	12
Control	$5.19 \times 10^3$	$6.11 \times 10^4$	$4.15 \times 10^6$	$2.25 \times 10^7$	$2.33 \times 10^8$
Thyme 1% (T1)	$5.19 \times 10^3$	$5.63 \times 10^4$	$6.25 \times 10^6$	$1.15 \times 10^7$	$5.10 \times 10^8$
Thyme 1.5% (T2)	$5.18 \times 10^3$	$4.53 \times 10^4$	$5.11 \times 10^5$	$4.22 \times 10^6$	$3.13 \times 10^7$
Thyme 2% (T3)	$5.17 \times 10^3$	$3.73 \times 10^3$	$3.15 \times 10^5$	$5.89 \times 10^5$	$2.15 \times 10^6$
<i>C. zeylanicum</i> 0.5% (T4)	$5.19 \times 10^3$	$8.53 \times 10^4$	$6.12 \times 10^6$	$5.15 \times 10^7$	$5.14 \times 10^8$
<i>C. zeylanicum</i> 1% (T5)	$5.19 \times 10^3$	$7.93 \times 10^4$	$6.10 \times 10^5$	$5.15 \times 10^6$	$4.32 \times 10^7$
<i>C. zeylanicum</i> 1.5% (T5)	$5.18 \times 10^3$	$5.21 \times 10^4$	$4.14 \times 10^5$	$6.15 \times 10^5$	$2.32 \times 10^6$

Values with different letters within the same row differed significantly at (P<0.05)

Table 4: Changes in aerobic plate count (APC) of untreated and treated minced meat samples during storage period at 2°C (Mean ± SE).

Storage days	Enterobacteriaceae count				
	0	3	6	9	12
Control	$9.32 \times 10^3 \pm 2.25 \times 10^3$	$7.24 \times 10^4 \pm 1.1 \times 10^4$	$8.15 \times 10^5 \pm 1.89 \times 10^5$	$4.25 \times 10^6 \pm 2.09 \times 10^6$	$3.25 \times 10^8 \pm 2.19 \times 10^8$
Thyme 1% (T1)	$9.32 \times 10^3 \pm 2.25 \times 10^3$	$7.04 \times 10^4 \pm 1.1 \times 10^4$	$7.12 \times 10^5 \pm 1.17 \times 10^5$	$6.45 \times 10^5 \pm 1.09 \times 10^5$	$6.25 \times 10^6 \pm 2.19 \times 10^6$
Thyme 1.5% (T2)	$9.32 \times 10^3 \pm 2.25 \times 10^3$	$7.03 \times 10^4 \pm 1.1 \times 10^4$	$4.11 \times 10^5 \pm 1.99 \times 10^5$	$9.25 \times 10^4 \pm 2.09 \times 10^4$	$4.25 \times 10^5 \pm 1.19 \times 10^5$
Thyme 2% (T3)	$9.32 \times 10^3 \pm 2.25 \times 10^3$	$6.81 \times 10^4 \pm 1.1 \times 10^4$	$8.65 \times 10^4 \pm 2.36 \times 10^4$	$8.25 \times 10^5 \pm 2.69 \times 10^5$	$1.55 \times 10^5 \pm 2.19 \times 10^5$
<i>C. zeylanicum</i> 0.5% (T4)	$9.32 \times 10^3 \pm 2.25 \times 10^3$	$7.24 \times 10^4 \pm 1.1 \times 10^4$	$7.24 \times 10^5 \pm 1.81 \times 10^5$	$5.27 \times 10^5 \pm 1.67 \times 10^5$	$8.95 \times 10^6 \pm 2.19 \times 10^6$
<i>C. zeylanicum</i> 1% (T5)	$9.32 \times 10^3 \pm 2.25 \times 10^3$	$7.01 \times 10^4 \pm 1.1 \times 10^4$	$7.15 \times 10^5 \pm 1.29 \times 10^5$	$2.25 \times 10^5 \pm 2.09 \times 10^5$	$3.25 \times 10^6 \pm 2.01 \times 10^6$
<i>C. zeylanicum</i> 1.5% (T6)	$9.32 \times 10^3 \pm 2.25 \times 10^3$	$6.92 \times 10^4 \pm 1.1 \times 10^4$	$8.95 \times 10^4 \pm 2.34 \times 10^4$	$9.25 \times 10^5 \pm 2.43 \times 10^5$	$4.35 \times 10^5 \pm 1.17 \times 10^5$

Table 5: Changes in enterobacteriaceae count (EBC) of treated and untreated minced meat samples during storage period at 2°C (Mean ± SE).



contains active compounds such as polyphenols, cinnamaldehyde and antibacterial compounds [30].

Table 5 showed that, at control samples the enterobacteriaceae count EBC was  $9.32 \times 10^3$ ,  $7.24 \times 10^4$ ,  $8.15 \times 10^5$ ,  $4.25 \times 10^6$  and  $3.25 \times 10^8$ , at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. By using thyme oil at concentration 1% (T1) EBC was  $9.32 \times 10^3$ ,  $7.04 \times 10^4$ ,  $7.12 \times 10^5$ ,  $6.45 \times 10^5$  and  $6.25 \times 10^6$ ; at concentration 1.5% (T2) EBC was  $9.32 \times 10^3$ ,  $7.03 \times 10^4$ ,  $4.11 \times 10^5$ ,  $9.25 \times 10^4$  and  $4.25 \times 10^5$  and at concentration 2% (T3) EBC was  $9.32 \times 10^3$ ,  $6.81 \times 10^4$ ,  $8.65 \times 10^4$ ,  $8.25 \times 10^5$  and  $1.55 \times 10^5$ , at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. From the obtained results, samples treated by different concentrations of thyme oil showed decreasing count of enterobacteriaceae when compared with control sample especially at 9<sup>th</sup> and 12<sup>th</sup> day, also high concentration of thyme (2%) was more effective in decreasing this count than lower concentration (1%). These results nearly similar to results were obtained by [50-55]. Also, Salem et al. [46], stated that preserved minced beef at 4°C during 6 days storage period at the control samples had the highest counts of enterobacteriaceae when compared to other treatment of minced beef by different concentration of thyme (0.5%, 1%, 1.5%) [46]. Also, the highest concentration of thyme (1.5%) appeared more decreasing of enterobacteriaceae count than lower concentration of thyme (0.5%). Also, [1] who recorded that preserved minced beef at refrigeration temperature, with treatment by thyme oil play an important role in decreasing enterobacteriaceae count especially at 1.5% concentration.

By using cinnamon oil at concentration 0.5% (T4) EBC was  $9.32 \times 10^3$ ,  $7.24 \times 10^4$ ,  $7.24 \times 10^5$ ,  $5.27 \times 10^5$  and  $8.95 \times 10^6$ ; at concentration 1% (T5) EBC was  $9.32 \times 10^3$ ,  $7.01 \times 10^4$ ,  $7.15 \times 10^5$ ,  $2.25 \times 10^5$  and  $3.25 \times 10^6$ ; at concentration 1.5% (T6) EBC was  $9.32 \times 10^3$ ,  $6.92 \times 10^4$ ,  $8.95 \times 10^4$ ,  $9.25 \times 10^5$  and  $4.35 \times 10^5$  at 0<sup>th</sup> day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. From the obtained results, samples treated by different concentrations of cinnamon oil caused decreasing count of enterobacteriaceae when compared to control samples especially at 9<sup>th</sup> and 12<sup>th</sup> day, also high concentration of cinnamon was more decreasing of EBC than lower concentration of cinnamon (0.5%). These results were nearly similar to results were obtained by Quattara et al. [42] who mentioned that treatment cooked meat products by trans-cinnamaldehyde, which is main component of cinnamon oil, produced significant extension of cooked meat products shelf life by inhibiting growth of enterobacteriaceae [42].

Table 6 showed that, at control samples coli form count was  $6.45 \times 10^3$ ,  $4.23 \times 10^5$ ,  $3.45 \times 10^6$ ,  $5.53 \times 10^7$  and  $6.45 \times 10^8$ , at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. By using thyme oil at concentration 1% (T1) coli form count was  $6.45 \times 10^3$ ,  $3.21 \times 10^5$ ,  $2.45 \times 10^5$ ,  $6.13 \times 10^5$  and  $6.15 \times 10^6$ , at concentration 1.5% (T2) coli form count was  $6.45 \times 10^3$ ,  $3.53 \times 10^4$ ,  $3.35 \times 10^5$ ,  $3.03 \times 10^5$  and  $5.45 \times 10^6$  and coli form count at concentration 2% (T3) was  $6.25$

$\times 10^3$ ,  $3.23 \times 10^4$ ,  $2.15 \times 10^4$ ,  $2.53 \times 10^5$  and  $4.45 \times 10^5$ , at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. From the obtained results, samples treated by different concentrations of thyme oil showed decreasing count of coli form when compared to control samples especially at 9<sup>th</sup> and 12<sup>th</sup> day, also high concentration of thyme oil (2%) was more effective in decreasing this count than lower concentration (1%). These results were nearly similar to that obtained by [35-40]. Also, Emiroglu et al. [17], who mentioned that thymol is effective to destroy coli forms when used at concentration of 250,500 and 750 mg/kg of beef minced patties and stored for 16 days under ordinary or modified packaging at refrigeration temperature, also mentioned that covering surface of meat products by 0.8% thymol, the count of coli forms decreased by 2-3 log units [17]. Also, agree with Salem et al. [46], stated that preserved minced beef at 4°C a long 6 days storage period, the control sample had the highest counts of coli forms compared to other treatment of minced beef by different concentration of thyme (0.5%, 1%, 1.5%), highest concentration of thyme (1.5%) appeared more effectiveness in decreasing coliforms count especially at 3<sup>rd</sup> and 6<sup>th</sup> day of storage period than lower concentration (0.5%) [46]. Also, similar to Amani et al. [1] who recorded that preserved minced beef at refrigeration temperature, with treatment by thyme oil play an important role in decreasing coliforms count especially at 1.5% concentration [1]. By using cinnamon oil at concentration 0.5% (T4) coli form count was  $6.45 \times 10^3$ ,  $4.13 \times 10^4$ ,  $5.25 \times 10^5$ ,  $9.23 \times 10^5$  and  $6.25 \times 10^6$ ; at concentration 1% (T5) coli form count was  $6.45 \times 10^3$ ,  $2.43 \times 10^4$ ,  $4.55 \times 10^5$ ,  $7.44 \times 10^5$  and  $6.05 \times 10^6$  and at concentration 1.5% (T6) coli form count was  $6.45 \times 10^3$ ,  $4.12 \times 10^4$ ,  $3.45 \times 10^5$ ,  $5.63 \times 10^5$  and  $5.55 \times 10^5$ , at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of storage period, respectively. From the obtained results, samples treated by different concentrations of cinnamon oil caused decreasing count of coli form when compared to control sample especially at 9<sup>th</sup> and 12<sup>th</sup> day, also high concentration of cinnamon (1.5%) was more effective in decreasing this count than lower concentration (0.5%). These results were nearly similar to results were obtained by [22,25,36,50] who stated that treatment of minced beef under cool temperature 4°C by using of cinnamon (*C. zeylanicum*) oil at different concentrations (0.5%, 1%, 1.5%) showed decreasing count of coliform when compared to control sample, also high concentration of cinnamon (1.5%) more effective in decreasing this count than lower concentration (0.5%). Also, Jagadeesh et al. [32], who mentioned that cinnamon oil decreased coli form count might be due to cinnamaldehyde and the antimicrobial compound of cinnamon [32].

### Effect of different concentrations of thyme (*T. vulgaris*) and cinnamon (*C. zeylanicum*) essential oils on sensory properties in minced meat during cold storage at 2°C

Tables 7 and 8 revealed that sensory properties of different treated minced beef samples during cold storage (2°C) were enhanced by using

Storage days	0	3	6	9	12
Control	$6.45 \times 10^3 \pm 1.21 \times 10^3$	$4.23 \times 10^5 \pm 1.01 \times 10^5$	$3.45 \times 10^6 \pm 1.46 \times 10^6$	$5.53 \times 10^7 \pm 1.11 \times 10^7$	$6.45 \times 10^8 \pm 1.19 \times 10^8$
Thyme 1% (T1)	$6.45 \times 10^3 \pm 1.21 \times 10^3$	$3.21 \times 10^5 \pm 1.01 \times 10^5$	$2.45 \times 10^5 \pm 1.26 \times 10^5$	$6.13 \times 10^5 \pm 1.11 \times 10^5$	$6.15 \times 10^6 \pm 1.19 \times 10^6$
Thyme 1.5% (T2)	$6.45 \times 10^3 \pm 1.21 \times 10^3$	$3.53 \times 10^4 \pm 1.51 \times 10^4$	$3.35 \times 10^5 \pm 1.16 \times 10^5$	$3.03 \times 10^5 \pm 1.11 \times 10^5$	$5.45 \times 10^6 \pm 1.19 \times 10^6$
Thyme 2% (T3)	$6.25 \times 10^3 \pm 1.21 \times 10^3$	$3.23 \times 10^4 \pm 1.03 \times 10^4$	$2.15 \times 10^4 \pm 1.06 \times 10^4$	$2.53 \times 10^5 \pm 1.11 \times 10^5$	$4.45 \times 10^5 \pm 1.18 \times 10^5$
<i>C. zeylanicum</i> 0.5% (T4)	$6.45 \times 10^3 \pm 1.21 \times 10^3$	$4.13 \times 10^5 \pm 1.02 \times 10^5$	$5.25 \times 10^5 \pm 1.22 \times 10^5$	$9.23 \times 10^5 \pm 1.11 \times 10^5$	$6.25 \times 10^6 \pm 1.19 \times 10^6$
<i>C. zeylanicum</i> 1% (T5)	$6.45 \times 10^3 \pm 1.21 \times 10^3$	$2.43 \times 10^4 \pm 1.01 \times 10^4$	$4.55 \times 10^5 \pm 1.86 \times 10^5$	$7.44 \times 10^5 \pm 1.11 \times 10^5$	$6.05 \times 10^6 \pm 1.19 \times 10^6$
<i>C. zeylanicum</i> 1.5% (T6)	$6.45 \times 10^3 \pm 1.21 \times 10^3$	$4.12 \times 10^4 \pm 2.01 \times 10^4$	$3.45 \times 10^5 \pm 1.26 \times 10^5$	$5.63 \times 10^5 \pm 1.21 \times 10^5$	$5.55 \times 10^6 \pm 1.16 \times 10^6$

Table 6: Changes in coli form count of untreated and treated minced meat samples during storage period at 2°C (Mean ± SE).

Sensory scores					
Storage days	0	3	6	9	12
Control	Excellent	Fair	Poor	very poor	Very very poor
Thyme 1% (T1)	Excellent	Good	Medium	Fair	Poor
Thyme 1.5% (T2)	Excellent	Good	Good	Medium	Fair
Thyme 2% (T3)	Excellent	Very very good	Very good	Good	Medium
<i>C. zeylanicum</i> 0.5% (T4)	Excellent	Good	Medium	Fair	Poor
<i>C. zeylanicum</i> 1% (T5)	Excellent	Very good	Good	Medium	Fair
<i>C. zeylanicum</i> 1.5% (T5)	Excellent	Very very good	Very good	Good	Medium

**Table 7:** Mean scores of sensory characteristics of untreated and treated minced meat samples during storage period at 2°C.

Points	9	8	7	6	5	4	3	2	1
Quality	Excellent	Very very good	Very good	good	medium	fair	poor	Very poor	Very very poor

**Table 8:** Score System for Sensory Evaluation (Pearson and Tuber, 1984).

thyme and cinnamon oils compared to the untreated (control) samples at 0 day, 3<sup>rd</sup> day, 6<sup>th</sup> day, 9<sup>th</sup> day and 12<sup>th</sup> day of the storage period, respectively, as control samples were started to spoilage and appearing rancid odor at 6<sup>th</sup> day of storage period, while by using thyme and cinnamon oil sensory properties were improved till 12<sup>th</sup> day of storage period. Also, samples containing 2% thyme oil and 1.5% cinnamon oil, demonstrated the highest enhancement of sensory attributes, while the samples treated with 1% of thyme and 0.5% of cinnamon oils demonstrated lower enhancement. This result agrees with that obtained by Sasse et al. [47], who reported that many herbs and spices as thyme contain antioxidant components that improve both color and flavor stability in meat [47]. Also, Salem et al. [46] mentioned that sensory properties of minced beef samples during cold storage (4°C) were enhanced by treatment minced beef by different concentrations of thyme oil (0.5%, 1%, 1.5%) compared to the untreated (control) samples and sample contain 1.5% thyme oil revealed best enhancement of sensory properties than sample contain 0.5% of the same oil, at zero, 3<sup>rd</sup> and 6<sup>th</sup> day of the storage period, respectively [46]. The sensory properties of different treated minced beef samples during cold storage (4°C) were improved by using different concentrations (0.5%, 1% and 1.5%) of thyme oil, compared to the control samples after 3 hrs. 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> day of the storage period, also sample containing 1.5% thyme demonstrated the highest enhancement of sensory attributes than sample treated with 0.5% of the same oil. Also Amani et al. [1] they reported that the sensory properties of different treated minced beef samples by using thyme oil at concentrations (1.5%, 1%, 0.5%) and cinnamon oil at concentrations (0.5%, 1%, 1.5%) during cold storage (4°C) were enhanced at treatment samples compared to the untreated (control) samples at zero, 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> day of the storage period, also samples containing 1.5% (cinnamon and thyme) oils demonstrated the highest enhancement of sensory properties [1].

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