

## NATURAL PEPPERMINT-FLAVORED CHEESE

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### ABSTRACT

**Background.** The essential oils of edible and medicinal plants, herbs and spices are natural biologically active agents. Peppermint has a special position in Egyptian culture and is used as fresh or dried leaves. One of the main aims of the food industry is to manufacture products with good sensory acceptance. The present study aims to produce a novel attractive cheese with a low concentration of natural peppermint (*Mentha piperita*) essential oil (PEO). Moreover, PEO volatile composition, total phenolic content (TPC) and antioxidant activity extracted by the hydro-distillation of peppermint fresh leaves were investigated.

**Material and methods.** Volatile components of PEO were identified using a GC-MS instrument. Total phenolic content and antioxidant activity were determined using the DPPH radical scavenging method for PEO. Moreover, PEO-flavored cheeses at a level of 20, 40, 60, 80 and 100 ppm compared to plain cheese were analyzed for sensorial, chemical and rheological properties during storage at 5°C.

**Results.** Identification of PEO's volatile compounds using GC-MS with a flame ionization detector analysis showed that the main components were menthol (37.62%), menthone (20.98%), carvone (11.76%), dihydro carveol acetate (11.23%), cineol (5.89%),  $\beta$ -caryophyllene (2.94), limonene (2.78%) and iso-menthone (2.39). Moreover, the antioxidant activity of PEO was 56.03 (%) with a TPC of 0.299 (mg/ml). Sensory evaluation of cheese showed the flavor and body and texture of PEO-flavored cheeses were higher than plain cheese during the storage period. There were no significant ( $p \leq 0.05$ ) differences in the cheese's appearance nor in the chemical composition of plain and PEO-flavored cheeses. However, the TVFA and SN of plain cheese was significantly ( $p \leq 0.05$ ) lower than PEO cheeses at the level of 60, 80 and 100 ppm during the storage period.

**Conclusion.** Hydro-distillation of Egyptian peppermint leaves revealed a lot of volatile compounds and antioxidant activity for the resulting essential oil; Moreover, the presence of PEO in cheese leading to enhanced TVFA and SN nitrogen contents compared to plain cheese may offer a novel flavored cheese with desirable multifunctional health effects in humans. Peppermint-flavored cheese was more accepted than plain cheese and survived without any defects until the end of the cheese storage period.

**Keywords:** peppermint, natural, essential oil, GC-MS, cheese

### INTRODUCTION

Essential oils are the main class of aromatherapy substances, which are obtained from various aromatic parts of plants by different methods, such as

hydro- and steam-distillations or cold pressing. Edible and medicinal plants, herbs and spices extracts and their essential oils are very potent natural biologically

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active agents (Nychas et al., 2003). Essential oils are volatile plant components that have been used in medicine, food and drinks flavoring, perfumery, cosmetics, incense and in household cleaning products (Bakkali et al., 2008). A few preservatives containing essential oils are already commercially available, such as Apopka, FL, USA, and are generally classified in USA as safe (GRAS) food additives (Burt, 2004).

Among aromatic and medicinal herbs, peppermint (*Mentha piperita*), whose ethnic Arabic name is nana al-fulfuli, like most species of mint, derives from the Latin *mentha*, meaning “mint”, which in turn originated from the Greek *Minthē*, the name of a nymph in ancient Greek mythology. The “Pepper” component derives from the genus name *Piperita*, referring to the strong, peppery taste (Singh et al., 2015). In traditional and folk medicines, peppermint was used for its antimicrobial, anti-inflammatory and antioxidant properties (Tsai et al., 2013); it has a special position in Egyptian culture, which uses it as fresh or dried leaves. It has traditionally been used for gastrointestinal disorders, colds, headache and cramps (Spirling and Daniels, 2001).

One of the main objectives of the food industry is to manufacture products with good sensory acceptance. To achieve this objective and overcome strong competition, a product must meet consumer expectations. In this context, using essential oils in the production of cheese could be a promising alternative addition for dairy industries, because cheeses are viewed positively by consumers (Kresic et al., 2010). Amirdivani and Baba (2011) studied the influence of peppermint (*Mentha piperita*), dill (*Anethum graveolens*) and basil (*Ocimum basilicum*) on yoghurt fermentation. Flavored-yoghurts had faster rates of pH reduction than plain yoghurts. Peppermint, dill and basil may be used to modify milk microbial fermentation with the intention of dairy products producing with higher antioxidant capacity. Furthermore, the effect of mint (*Mentha spicata*), bee balm (*Mentha longifolia*), eucalyptus (*Eucalyptus camaldulensis*) and ziziphora (*Ziziphora tenuior*) on probiotic drinking yoghurt characteristics were investigated by Shahdadi et al. (2015). These essential oils inhibited pH reduction during storage except for eucalyptus essential oils, which did not decrease the viability of probiotic bacteria. All showed higher viability than the control yoghurt.

Therefore, the aim of this study was to produce a novel, attractive, white soft cheese with a low concentration of natural peppermint (*Mentha piperita*) essential oil (PEO) extracted by hydro-distillation of Egyptian peppermint leaves. Their effect on the sensorial, chemical and rheological properties of cheese during storage was studied. In addition, PEO's volatile composition, antioxidant activity and total phenolic content were investigated.

## MATERIALS AND METHODS

### Materials

Fresh buffalo milk was obtained from the herd at the Faculty of Agriculture, Cairo University, Egypt. Microbial rennet powder (RENIPLUS) from *Mucor miehei* was purchased from Gaglio Star, Spain. Gallic acid, 2,2-diphenyl-1-picryl-hydrazyl (DPPH) and Folin-Ciocalteu phenol reagent were purchased from Sigma Aldrich, Germany. The other chemicals used were of analytical grade.

### Extraction of peppermint essential oil

Fresh peppermint (*Mentha piperita*) leaves, purchased from a local market in Egypt were charged into a 5-liter round-bottomed flask and mixed with distilled water at a ratio of 1:10 (plant material to water) followed by hydro-distillation using a Clevenger-type apparatus for 2.5 h. At the end of distillation, the separated essential oil was collected from the side arm of the apparatus, dried over anhydrous sodium sulfate and stored in dark-glass bottles at  $-4^{\circ}\text{C}$  until it was used in the manufacture of the flavored cheese. The distillation procedure was repeated several times with a new fresh charge of peppermint leaves each time until the required amount of peppermint essential oil had been collected.

### Chromatographic analyses of peppermint essential oil

Analysis of peppermint essential oil GC was performed using a Perkin Elmer Auto System XL model equipped with a flame ionization detector (FID). A fused silica capillary column ZB-5 (60 m  $\times$  0.32 mm i.d.) was used. The oven temperature was programmed from  $50^{\circ}\text{C}$  to  $240^{\circ}\text{C}$  at a rate of  $3^{\circ}\text{C}/\text{min}$ . Helium was used as the carrier gas, at a flow rate 1.1 ml/min. The injector and detector temperatures were  $230^{\circ}\text{C}$

and 250°C. The retention indices (kovats index) of the separated volatile compounds were calculated with reference to the retention time of the n-paraffin (C6–C20) series. The isolated peaks were identified by comparison with those of authentic compounds.

### Total phenolic content of peppermint essential oil

The total phenolic content of PEO was determined colorimetrically using a Folin-Ciocalteu reagent, in accordance with the modified method described by Lafka et al. (2007). Total phenolic content was calculated by a calibration curve prepared with Gallic acid as a standard and expressed as mg gallic acid equivalent (GAE/ml).

### Antioxidant activity of peppermint essential oil

The antioxidant activity of PEO was evaluated by using the stable 2,2-diphenyl-1-picryl-hydrazyl (DPPH, Sigma Aldrich, Germany) radical scavenging method, in accordance with Matthus' modified method (2002). The radical scavenging activities of the samples tested, which were expressed as a percentage inhibition of DPPH, were calculated according to the following formula:

$$\text{Inhibition, \%} = ((A^{\text{control}} - A_0^{\text{sample}}) / A^{\text{control}}) \times 100$$

where:

- A – the absorbance at 515 nm of the control sample,
- A<sub>0</sub> – the final absorbance of the test sample at 515 nm.

### White soft cheese preparation

Fresh buffalo milk was heated to 72°C/15 s, and after this was cooled to 37°C calcium chloride and sodium chloride were added at the rate of 0.02% and 4.0% (w/w), respectively. Milk was divided into four equal portions as follows: the first portion was used as the control, and the other five portions were flavored with peppermint (*Mentha piperita*) essential oil at the level of 20, 40, 60, 80 and 100 ppm (served as T1, T2, T3, T4 and T5, respectively). All the portions of milk were coagulated with microbial rennet powder (RENIPLUS), then the resulting curds were packed into small plastic cups and stored at 5°C ±1. White soft cheese was made from each portion in accordance

with Mehanna and Rashed's method (1990), except for the fact that the milk was not treated with CO<sub>2</sub>. Cheese samples were taken weekly for analysis during one month of the storage period. Plain and the five cheeses flavored with peppermint essential oil were prepared on the same day. The whole experiment was repeated two times.

### Preliminary experiments

White soft cheeses flavored with peppermint (*Mentha piperita*) essential oil were manufactured at the level of 20 and 100 ppm. The sensorial properties of the cheese were evaluated by staff from the Dairy Science Department at the National Research Centre, in accordance with Pappas et al. (1996), with a maximum score of 50 points for flavor, body and texture (40 points) and 10 points for the cheese's appearance, in order to select the proper concentration range of peppermint essential oil.

### Cheese chemical analysis

White soft cheese samples were chemically analyzed according to AOAC (2000) for total solids, fat, total protein, ash and soluble nitrogen contents. The pH of the cheese samples was measured using a pH-meter (Hanna Instruments Model 170300, Ingold, Knick, Germany). Acidity as lactic acid was determined by the titration method with 1/9 N NaOH, according to Ling's method (1963). Total volatile fatty acids were determined according to Kosikowski's procedure (1982) with 0.1 N NaOH per 100 g cheese.

### Texture profile analysis of cheese

Texture profile analysis (TPA) was carried out using at least three samples for each cheese treatment with a Universal Testing Machine (Cometech, B type, Taiwan) complete with software. A back extrusion cell with 35 mm diameter compression disc was used. Two cycles were applied at a constant crosshead velocity of 1 mm secG1 to 35% of the sample depth then returned. From the resulting force-time curve the values for texture attributes, i.e., hardness, chewiness, cohesiveness, gumminess and springiness were calculated.

### Sensory properties of cheese

The sensory properties of the cheese samples after 1, 2, 3 and 4 weeks of cold storage at 5 ±1°C were

evaluated according to Pappas et al. (1996). Cheese was assessed by 15 panelists from the staff of the Dairy Science Department at the National Research Centre, with a maximum score points of 50 points for flavor, body and texture (40 points) and 10 points for the cheese's appearance.

### Statistical analysis

Statistical analysis of the results obtained was performed using the Statistical Analysis System (SAS) using the ANOVA procedure for analysis of variance, and the general linear model (GLM) procedure for SAS software (SAS, 1990). The results were expressed as mean  $\pm$  standard error and the differences between means were tested for significance using Duncan's multiple range test at ( $p \leq 0.05$ ).

## RESULTS AND DISCUSSION

Volatile composition, antioxidant activity and total phenolic content of PEO extracted by hydro-distillation in using Clevenger-type apparatus were investigated. Moreover, the effect of natural PEO on the sensorial, chemical and rheological properties of white soft cheese during cold storage was investigated.

### Volatile composition of peppermint essential oil

Fifteen volatile components of PEO were identified by GC-MS instrument using a flame ionization detector as shown in Table 1. The principal constituents identified in PEO were menthol (37.62%), carvone (11.76%), menthone (20.98%), limonene (2.78%), di-hydrocarveol acetate (11.23%), cineol (5.89%) and  $\beta$ -caryophyllene (2.94%). In addition, other minor volatile compounds were identified in PEO, including iso-menthone,  $\alpha$ -pinene, sabinene,  $\beta$ -myrcene, trans-menthyl acetate,  $\beta$ -bourbonene,  $\beta$ -elemene and  $\beta$ -muurolene. Similar results were obtained by Tsai et al. (2013) who found that the major components of PEO were menthol (30.35%), menthone (21.12%) and trans-carane (10.99%). Khan and Abourashed (2010) reported that the peppermint volatile oil yield was 0.1–1.0% and the main components were menthol (29–48%), menthone (20–31%) and menthyl acetate (3–10%). The components of mint oil varied with plant maturity, variety, geographical region and processing conditions (Rohloff et al., 2005; Scavroni et al., 2005).

**Table 1.** Volatile composition of peppermint (*Mentha piperita*) essential oil

Volatile compound	Concentration, %
Menthol	37.62
Carvone	11.76
Menthone	20.98
Limonene	2.78
Di-hydro carveol acetate	11.23
Cineol	5.89
$\beta$ -caryophyllene	2.94
Iso-menthone	2.39
$\alpha$ -pinene	0.61
Sabinene	0.46
$\beta$ -myrcene	0.91
Trans-menthyl acetate	1.20
$\beta$ -bourbonene	0.09
$\beta$ -elemene	0.62
$\beta$ -muurolene	0.52

Menthol is the primary component of PEO and is mostly responsible for antispasmodic and antimicrobial effects (Balakrishnan, 2015; Chouhan et al., 2017). Menthol is a bactericidal against more strains, such as *Staphylococcus pyogenes*, *S. aureus*, *Streptococcus pyogenes*, *Escherichia coli*, and *Mycobacterium avium* (El-Kady et al., 1993; Pattnaik et al., 1997). Menthol is also a fungicidal against *Candida albicans*, *Aspergillus albus* and *dermatophytic* fungi (El-Kady et al., 1993). In addition, the pharmacologically active ingredient of PEO is menthol, through its ability to act as a calcium antagonist, menthol appears to have a spasmolytic effect in the gastrointestinal tract. Consequently, peppermint oil was used to treat abdominal ailments in ancient Egypt, Greece and Rome (Ulbricht et al., 2010). It seems that peppermint oil relaxes the gastrointestinal smooth muscles by reducing calcium influx in both the large intestine and jejunum (Nissen and Lau, 2016). Peppermint oil and menthol are inhibitors for calcium channel activity in rats and guinea

pig atrial and papillary muscle, rat brain synaptosomes and chick retinal neurons (Harris, 2016).

D-limonene is one of the most common terpenes in nature. It is another of the most abundant active compounds of peppermint oil (1–5%) and several citrus oils such as orange, lemon, mandarin, lime and grapefruit. D-limonene has also been used clinically as an excellent solvent of cholesterol-containing gallstones due to its gastric acid neutralizing effect and its support for normal peristalsis. It has also been used to relieve heartburn. Moreover, it is well established that limonene has chemopreventive activity against many types of cancers (Sun, 2007). Limonene was among the examples of some constituents of essential oils with antimicrobial activity (Chouhan et al., 2017).

Several reports have shown that PEO has significant antiviral activity against HSV-1 and HSV-2 viruses (Edris, 2007; Reichling et al., 2009). It seems that peppermint helps the immune system and protects the body against viruses (Loolaie et al., 2017).

#### Total phenolic content and antioxidant activity of peppermint essential oil

Table 2 shows the PEO total phenolic content (TPC) and their antioxidant activity. The TPC of PEO was  $0.299 \pm 0.04$  (mg/ml) and its antioxidant activity was  $56.03 \pm 3.23$  (%), which could be due to the presence of indigenous phytochemical compounds in PEO (Mairapetyan et al., 2016; Riachi and Maria, 2015). It could be mean that the peppermint flavored cheeses had higher TPC and greater antioxidant activity than the control cheese due to the phytochemical content of PEO and cheese starter culture metabolic activity. This was in accordance with the findings of Ishikawa et al. (2002), Thompson et al. (2007) and Amirdivani and Baba (2011), who reported that the higher TPC and antioxidant activity in flavored yoghurts than plain yoghurt were most likely influenced by phytochemical

compounds in herbs (e.g., flavonoids and phenolic compounds) compared to plain yoghurt and microbial metabolic activities. Özer et al. (2007), also reported that herbal essential oils have antimicrobial and antioxidant effects. The herbs' antioxidant activities have been attributed to various mechanisms such as prevention of chain initiation, binding of transition metal ion catalysts, decomposition of peroxides, prevention of continued hydrogen abstraction and radical scavenging (Tsai et al., 2013).

#### Sensory evaluation of cheese

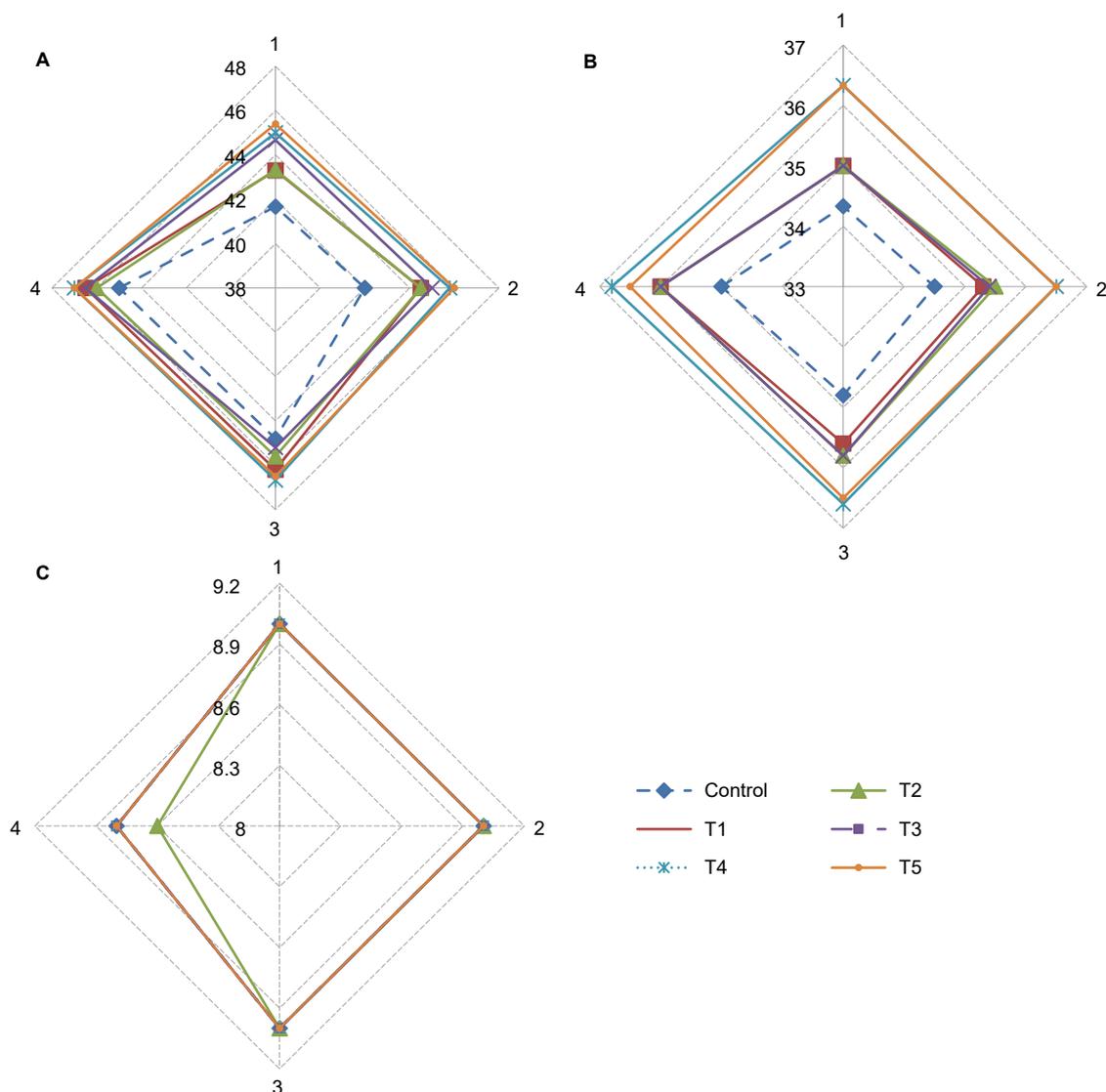
The sensorial properties of preliminary experimental cheeses showed that the higher concentration (100 ppm) of PEO added to white cheese without salt had higher scores in flavor, body and texture compared to the control cheese. On the other hand, the panellists preferred the salted white cheese (5%) flavored with a lower level (20 ppm) of natural PEO in all sensorial properties compared to plain cheese. Therefore, the PEO level ranged from 20 to 100 ppm and the salt level was reduced to 2%.

The flavor, body and texture and appearance of all cheese samples were evaluated during the cheese's one-month cold storage. Figure 1 (a, b) shows the flavor and body and texture of peppermint-flavored cheeses were higher than the control cheese during the storage period, which could be due to the addition of natural PEO imparting unique organoleptic properties to the experimental cheeses, where the herbal essential oils were used to improve the sensory characteristics and extend the shelf life of food (Özer et al., 2007). Moreover, PEO contains more volatile compounds, including menthol, menthone, limonene, iso-menthone, trans-menthyl acetate, isomenthone,  $\alpha$ -pinene, sabinene,  $\beta$ -myrcene,  $\beta$ -bourbonene,  $\beta$ -elemene and  $\beta$ -muurolene. El-Zaeddi et al. (2016) reported that mint samples have the highest number of volatile compounds and the higher concentration of these compounds in their essential oils, and were characterized by high intensity of mint (9.5), green grass (7.5, green aromatics associated with newly cut-grass and leafy plants), citrus (8.5, aromatics associated with commonly known citrus fruits, such as lemons, limes and oranges), and spicy (5, sharp aromatics with a physically penetrating sensation in the nose reminiscent of radish and horseradish). Moreover, it should be noted

**Table 2.** Total phenolic content and antioxidant capacity of peppermint (*Mentha piperita*) essential oil

Total phenolic compounds, mg/ml	Antioxidant capacity, %
$0.299 \pm 0.04$	$56.03 \pm 3.23$

Each value is a mean of duplicates  $\pm$  standard error.



**Fig. 1.** Sensory properties of white soft cheese flavored with peppermint (*Mentha piperita*) essential oil during storage at  $5 \pm 1^\circ\text{C}$ : A – flavor, B – body and texture, C – appearance; Control – cheese without peppermint oil, T1 – cheese flavored with 20 ppm of peppermint oil, T2 – cheese flavored with 40 ppm of peppermint oil, T3 – cheese flavored with 60 ppm of peppermint oil, T4 – cheese flavored with 80 ppm of peppermint oil, T5 – cheese flavored with 100 ppm of peppermint oil

that the flavor score of T4 and T5 flavored with 80 and 100 ppm of PEO respectively were significantly ( $p \leq 0.05$ ) higher than the plain cheese. No significant ( $p \leq 0.05$ ) differences in the appearance of control and experimental cheeses flavored with PEO during the storage period as shown in Figure 1 (c).

### Chemical composition of cheese

The chemical composition of the control and experimental cheeses flavored with PEO are presented in Table 3. The moisture contents of peppermint-flavored cheese were slightly lower than plain cheese. Furthermore, Table 3 shows that the total protein/dry matter, fat/dry

**Table 3.** Chemical composition of white soft cheese flavored with peppermint (*Mentha piperita*) essential oil

Parameter	Cheese treatments, %					
	C	T1	T2	T3	T4	T5
Moisture	57.78 ±0.33 <sup>a</sup>	57.09 ±0.43 <sup>ab</sup>	55.55 ±0.95 <sup>b</sup>	55.93 ±0.85 <sup>ab</sup>	55.33 ±0.70 <sup>b</sup>	56.04 ±0.55 <sup>ab</sup>
TP/DM	22.12 ±0.68 <sup>c</sup>	22.77 ±0.15 <sup>b</sup>	22.18 ±0.38 <sup>bc</sup>	24.89 ±0.5 <sup>a</sup>	24.63 ±0.35 <sup>a</sup>	25.09 ±0.06 <sup>a</sup>
F/DM	54.48 ±0.29 <sup>c</sup>	58.26 ±0.15 <sup>a</sup>	56.24 ±0.12 <sup>b</sup>	56.73 ±0.09 <sup>b</sup>	55.97 ±0.58 <sup>b</sup>	56.87 ±0.06 <sup>b</sup>
Ash	2.25 ±0.05 <sup>a</sup>	2.29 ±0.01 <sup>a</sup>	2.32 ±0.12 <sup>a</sup>	2.34 ±0.01 <sup>a</sup>	2.3 6±0.01 <sup>a</sup>	2.37 ±0.01 <sup>a</sup>

All parameters are represented as a mean of replicates ±standard error. Averages in the same line with different superscript letters are significantly different at  $p \leq 0.05$ .

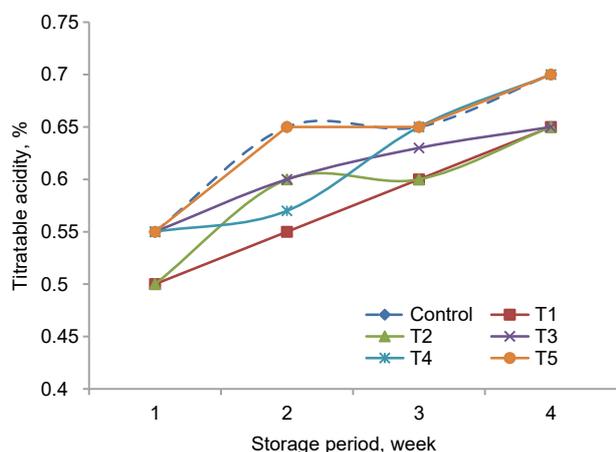
C – control cheese without peppermint oil, T1 – cheese flavored with 20 ppm of peppermint oil, T2 – cheese flavored with 40 ppm of peppermint oil, T3 – cheese flavored with 60 ppm of peppermint oil, T4 – cheese flavored with 80 ppm of peppermint oil, T5 – cheese flavored with 100 ppm of peppermint oil. DM – dry matter, TP – total protein, F – fat content.

matter and ash contents of cheese treatments flavored with PEO had higher values than the control cheese.

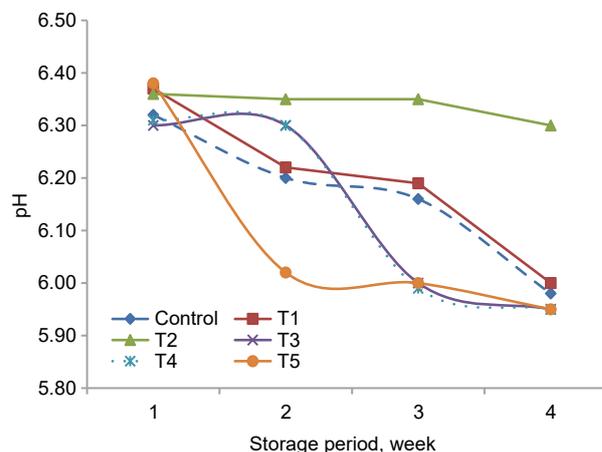
### Acidity and pH changes of cheese

The acidity changes of white soft cheese during storage at  $5 \pm 1^\circ\text{C}$  for 4 weeks are presented in Figure 2. It can be noted that the acidity of the control cheese was slightly higher than the experimental cheese

which contains PEO with opposite trends in pH values without any significant ( $p \leq 0.05$ ) differences between the control and cheese treatments as shown in Figure 3. These findings could be due to the presence of PEO which slight affects the cheese starter culture and acid production. It appeared that the essential oil compounds prevent the starter cultures growth and acid production (Khodaparast et al., 2007). These



**Fig. 2.** Acidity changes of white soft cheese flavored with peppermint (*Mentha piperita*) essential oil during storage at  $5 \pm 1^\circ\text{C}$ : Control – cheese without peppermint oil, T1 – cheese flavored with 20 ppm of peppermint oil, T2 – cheese flavored with 40 ppm of peppermint oil, T3 – cheese flavored with 60 ppm of peppermint oil, T4 – cheese flavored with 80 ppm of peppermint oil, T5 – cheese flavored with 100 ppm of peppermint oil



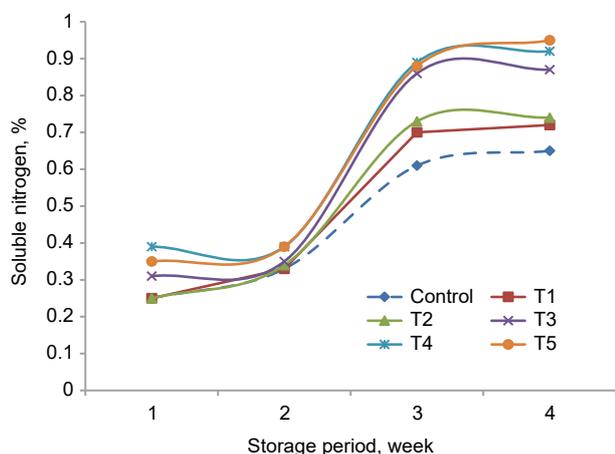
**Fig. 3.** pH variation of white soft cheese flavored with peppermint (*Mentha piperita*) essential oil during storage at  $5 \pm 1^\circ\text{C}$ : Control – cheese without peppermint oil, T1 – cheese flavored with 20 ppm of peppermint oil, T2 – cheese flavored with 40 ppm of peppermint oil, T3 – cheese flavored with 60 ppm of peppermint oil, T4 – cheese flavored with 80 ppm of peppermint oil, T5 – cheese flavored with 100 ppm of peppermint oil

results were in accordance with those of Shahdadi et al. (2014; 2015), who reported that the acidity of mint drinking yoghurt had a higher pH and lower acidity than plain yoghurt. On the other hand, Amirdivani and Baba (2011) observed that the acidity for herbal yoghurts was higher than plain yoghurt at all time points during the incubation time and these differences may be due to the herbal oil enhancing the metabolic activity of the cheese starter culture during the storage period. Moreover, the acidity of all cheese samples increased throughout the storage period.

### Changes of SN content in cheese

Water-soluble nitrogen content in cheese is primarily formed by coagulating enzymes, plasmin or cell-wall envelope proteases at the early stage of proteolysis. It is commonly known that during cheese ripening, protein breakdown is an important factor for the development of both flavor and texture (Sousa et al., 2001).

Figure 4 shows the SN content of control and peppermint-flavored cheese during storage at  $5 \pm 1^\circ\text{C}$  for one month. It can be noted that the SN content of cheeses flavored with PEO was higher than the plain cheese without PEO. Moreover, the SN content of all

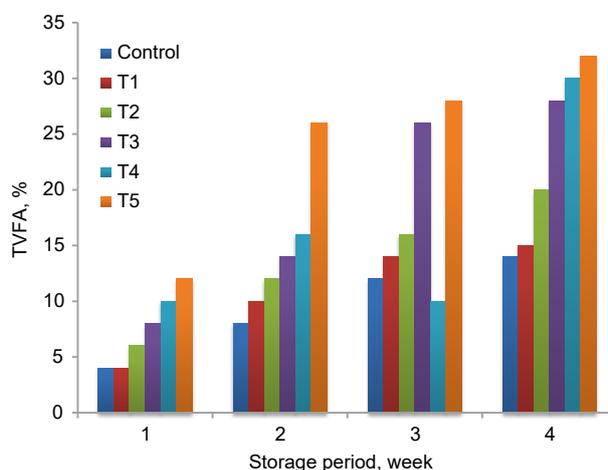


**Fig. 4.** Changes of soluble nitrogen content of white soft cheese flavored with peppermint (*Mentha piperita*) essential oil during storage at  $5 \pm 1^\circ\text{C}$ : Control – cheese without peppermint oil, T1 – cheese flavored with 20 ppm of peppermint oil, T2 – cheese flavored with 40 ppm of peppermint oil, T3 – cheese flavored with 60 ppm of peppermint oil, T4 – cheese flavored with 80 ppm of peppermint oil, T5 – cheese flavored with 100 ppm of peppermint oil

the cheese samples increased with as the storage period progressed. Statistical analysis of results shows that the SN content of control cheese was significantly ( $p \leq 0.05$ ) lower than cheese treatments of T3, T4 and T5 flavored with PEO at a level of 60, 80 and 100 ppm respectively during the cold storage period. This could be due to the proteolysis extended by the cheese starter culture being enhanced by the presence of peppermint, as shown by a substantial increase in SN values in experimental cheese compared to plain cheese without PEO during storage. This may lead to changes in the protein breakdown with potentially higher production of bioactive peptides (Shahidi and Zhong, 2008). These findings were more similar to Amirdivani and Baba (2011), who reported that herbal yoghurts had higher o-phthalaldehyde peptides values than plain yoghurt during the storage period. The proteolysis extent and peptide sizes produced during the cheese storage are expected to be different in the presence of herbs.

### TVFA content changes of cheese

Figure 5 shows TVFA changes to control and experimental cheeses flavored with PEO during storage at  $5 \pm 1^\circ\text{C}$  for 4 weeks. The TVFA content of plain cheese



**Fig. 5.** TVFA changes of white soft cheese flavored with peppermint (*Mentha piperita*) essential oil during storage at  $5 \pm 1^\circ\text{C}$ : Control – cheese without peppermint oil, T1 – cheese flavored with 20 ppm of peppermint oil, T2 – cheese flavored with 40 ppm of peppermint oil, T3 – cheese flavored with 60 ppm of peppermint oil, T4 – cheese flavored with 80 ppm of peppermint oil, T5 – cheese flavored with 100 ppm of peppermint oil

**Table 4.** Rheological properties of white soft cheese flavored with peppermint (*Mentha piperita*) essential oil

Parameter	Cheese treatments					
	C	T1	T2	T3	T4	T5
Hardness, N	0.70	3.0	4.7	5.2	3.1	4.9
Cohesiveness	0.67	0.51	0.51	0.43	0.46	0.45
Springiness, mm	0.65	0.42	0.51	0.37	0.39	0.41
Gumminess, N	0.45	1.25	2.39	1.91	1.22	1.99
Chewiness, N/m	0.30	0.64	1.23	0.82	0.56	0.91

C – control cheese without peppermint oil, T1 – cheese flavored with 20 ppm of peppermint oil, T2 – cheese flavored with 40 ppm of peppermint oil, T3 – cheese flavored with 60 ppm of peppermint oil, T4 – cheese flavored with 80 ppm of peppermint oil, T5 – cheese flavored with 100 ppm of peppermint oil.

was significantly ( $p \leq 0.05$ ) lower than peppermint-flavored cheeses (T2, T3, T4 and T5) at a level of 40, 60, 80 and 100 ppm respectively during the cold storage period. This could be due to the addition of PEO, which contains more volatile compounds, such as menthol, menthone, limonene, iso-menthone, trans-menthyl acetate, isomenthone,  $\alpha$ -pinene, sabinene,  $\beta$ -myrcene,  $\beta$ -bourbonene,  $\beta$ -elemene and  $\beta$ -muurolene. Moreover, the TVFA content of all the cheese samples increased with as the storage period progressed, which might be due to the reduction in moisture content during the storage period (Kebary et al., 1996).

#### Texture profile analysis of cheese

Table 4 shows the texture profile of the control cheeses and those flavored with PEO. It can be noted that the hardness, gumminess and chewiness of PEO-flavored cheese were higher than plain cheese. The hardness of the cheeses flavored with PEO was higher than the control cheese due to the low moisture content of the experimental cheeses compared to the control cheese. Calvo et al. (2007) reported that hardness significantly increased as a consequence of low moisture content. The texture profile of the cheese samples also shows that the springiness and cohesiveness of the control cheese was higher than that of peppermint-flavored cheeses. Springiness of flavored cheeses was lower than plain cheese could be due to the higher acidity and F/DM ratio compared to control cheese. Lower cohesiveness values of flavored cheeses with PEO

could be related to higher TP/DM ratio compared to plain cheese without peppermint essential oil.

#### CONCLUSION

Hydro-distillation of Egyptian peppermint leaves revealed a lot of identifiable volatile compounds and antioxidant activity in the resulting essential oil. Moreover, the presence of peppermint (*Mentha piperita*) essential oil in white soft cheese leads to enhanced total volatile fatty acid and soluble nitrogen content compared to plain cheese and may offer a new range of flavored cheese with desirable multifunctional health effects to consumers. The sensory properties of peppermint-flavored cheese showed that the flavor and body and texture values were higher than plain cheese during the storage period with no effect on the cheese's appearance without any defects until the end of the storage period. Although peppermint plays a great beneficial and economic role in human society, research must consider its minor side effects and toxicity. Future studies are required to determine the molecular mechanism of peppermint essential oil in human health.

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