

## Effect of Adding Cardamom, Thyme and Clove Essential Oils on Some Properties of White Soft Cheese Made from Goats' Milk

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### ABSTRACT

This study aimed to improve the properties of white soft cheese made from goats' milk by adding spice essential oils at concentrations of 75 & 90 ppm cardamom, 50 & 75 ppm thyme and 40 & 60 ppm clove to goats' milk. All resultant cheeses were compared against that made from buffalos' milk (control I) and goats' milk (control II). The quality of resultant cheeses was evaluated for their chemical composition, microbiological and organoleptic properties during storage at 6±1°C for 45 days. The results revealed that additives at two concentrations of essential oils insignificantly affected percentages of moisture, salt/ moisture and fat/dry matter. Moisture % significantly decreased ( $P \leq 0.05$ ) while fat and protein % increased during storage period at 6±1°C. Values of titratable acidity (TA), water soluble nitrogen/total nitrogen (WSN/TN), non protein nitrogen/total nitrogen (NPN/TN) and total volatile fatty acids (TVFA) for all treated cheeses were significantly lower than control II during pickling period. Increasing the proportion of spice essential oils had a marked effect on the same properties. The TA, WSN/TN, NPN/TN and TVFA in cheese samples increased, while the pH values decreased continuously during storage period ( $P \leq 0.05$ ).

The hysteresis area, expressed as  $\int \alpha \text{d}p\text{H}$  values, of control II was lower than goats' cheese samples treated by cardamom, thyme and clove essential oils during the pickling period. The rate of decrease in the hysteresis area of control II was higher than that of control I. Statistically, there were significant differences due to treatments, pickling period and the interaction between treatments and pickling period ( $P \leq 0.05$ ). Addition of spice essential oils decreased clearly counts of different tested microbial groups (total viable count, psychrophilic bacteria, yeasts & moulds and coliforms) throughout storage period as compared to control II and control I. Spice essential oils added improved the flavour and quality of white soft cheese made from goats' milk. Therefore, white soft cheese could be successfully made from goats' milk with adding cardamom, thyme or clove essential oils, especially clove or cardamom at concentrations of 40 and 75 ppm, respectively and with storage at 6±1°C for 45 days.

**Key words:** goats' milk, soft cheese, cardamom, thyme, clove, organoleptic properties, potentiometric acid-base titration.

### INTRODUCTION

Goats were probably among some of the first animals to be domesticated, and can utilize poor quality feeds such as shrubs and "left over" plants unsuitable for other milk animals. They have astonishing adaptability to adverse climatic and geophysical conditions (Devendra, 1987, Riekeman, 2005). Often the dairy goat has been called the "poor man's cow", because good dairy goats don't cost near as much as good dairy cows do. It is well known that goats' are early regarded as a source of milk production. A good dairy goat can produce up to 10% of its body weight in milk (Peterson, 2005). Goats' milk is also a healthier alternative to cow milk, because cow milk has to be homogenized to be more easily digested. However, this is not necessary with goats' milk because it is naturally homogenized. Moreover, the goats' milk protein gel

formed in the stomach is softer than that formed by cows' milk. This is also beneficial to the digestibility. Therefore, goats' milk is much more easily digested than cows' milk; also goats' milk has 13% less lactose than cows' milk and most people who are allergic to cows' milk tend not to be allergic to goats' milk. Allergies are more common in very young children (Einsiedel, 2005, Riekeman, 2005). On the other hand, goats' milk has more of the essential vitamins that we need.

Goats' milk has been used since ancient times for the manufacture of different types of cheeses, throughout the world. Many famous varieties of cheese are made from goats' milk, while in Egypt goat's milk products are not highly accepted because fresh goat's milk has a mild "goaty flavour", this flavour is mainly due to the presence of short-chain fatty acids, the breed of goats', unhygienic



milking of the goats and the diet of the animal has a lot to do with the taste of the milk. If goats and cows are managed similarly, the smell and the taste of both milks are quite comparable (Gates, 2005).

Since ancient times, peoples have used herbs and spices for preventing food deterioration and foodborne diseases. At the end of last century, antimicrobial activities of herbs and spices had already been examined and their oils were known to retard microbial spoilage in dairy products (Conner & Beuchat, 1984; Abou Dawood, 1996, Abd-El Kader *et al.*, 2001). In recent years, there are growing interests in using natural antimicrobial compounds, especially those extracted from plants for the preservation of foods and dairy products, which leads to increase the shelf life of these products. On the other hand, the species and herbs give a good flavour and mask the undesirable flavours, such as "goaty flavour" of goats' milk. In Egypt, many investigators used natural flavouring additives in dairy products as flavours and used as preservatives (antiviral, antibacterial and antifungal compounds) such as aqueous cayenne pepper and marjoram extracts in ultrafiltered cheese (Abd-Alla *et al.*, 2000); tolu balsam extracts in Karish cheese (El-Nemer *et al.*, 2003); twenty different essential oils (such as black cumin, shallots, cardamom, capsicum ... etc.) were used in manufacturing of flavoured Tallaga cheese (Hussein, 2004) and essential oils of garlic, cumin, coriander, clove, dill and parsley were added to Labneh (Ismail *et al.*, 2006). Therefore, this work aims to use goats' milk in making white soft cheese with satisfactory properties by adding cardamom, thyme and clove essential oils. At the same time the effect of these additives on chemical, microbiological and organoleptic properties of the resultant cheese during cold storage ( $6\pm 1^\circ\text{C}$ ) was studied as compared to control buffalos' and goats' cheese.

## MATERIALS AND METHODS

### Materials

Fresh buffalos' milk was obtained from the herd of animal production farm of the Faculty of Agriculture, Fayoum University, while fresh Egyptian goats' milk was obtained from a commercial private farm in Fayoum district. The gross chemical composition of both buffalos' and goats' milk is presented in Table (1). Standard microbial rennet powder (Maxiren 1800 MG, 100% chymosin purified from *Kluyveromyces lactis*, storage tem.

$4-5^\circ\text{C}$ ) was obtained from Hansen's Laboratories. Commercial edible grade salt (sodium chloride) produced by El-Nasr Company for Salt, Alex., Egypt was obtained from the local market and calcium chloride was obtained from Solvay, Italy. Spices (thyme, cardamom and clove) essential oils and Emulgum were obtained from Perfumes and Essences Factories, El-Hawamdia, Giza, Egypt. Essential oils were extracted by steam distillation according to the Food Chemical Codex, purity 100% and free from fatty acids.

### Methods

#### Preparation of essential oils

The essential oils (cardamom, thyme and clove) were emulsified at the rate of 4% (v/v) in Emulgum emulsion in water (10% w/v) using magnetic stirrer.

#### Manufacturing procedure

White soft cheese was made in the dairy processing pilot plant, Dairy Dept, Faculty of Agric, Fayoum Univ. using fresh whole buffalos' and goats' milk according to the method described by Fahmi & Sharara (1950). The cheese was manufactured using 10kg heated ( $75^\circ\text{C}$  for 15 sec and cooled to  $43^\circ\text{C}$ ) buffalos' milk as control (I) and 70kg heated goats' milk which was divided into seven portions where spice essential oils (the ratios of cardamom, thyme and clove were used based on the preference of the panelists as established in preliminary studies) were added as follow:

- 1- Goats' milk without additives (control II)
- 2- Treatment (1) and (2) goats' milk with the addition of cardamom essential oil at concentration of 75 and 90 ppm, respectively.
- 3- Treatment (3) and (4) goats' milk with addition of thyme essential oil at concentration of 50 and 75 ppm, respectively.
- 4- Treatment (5) and (6) goat's milk with addition of clove essential oil at concentration of 40 and 60 ppm respectively:

Calcium chloride, edible salt and rennet were added at the rate of 0.02, 4.0 and 0.003%, respectively, stirred well and set for 1 hr, the curd was filled in a special Domiati cheese moulds (perforated cylindrical stainless steel moulds, 7 cm diameter and 12 cm high) and drainage the whey takes 24 hrs at  $25\pm 1^\circ\text{C}$ . When the curd became firm enough, it is cut into pieces each 5 cm square. All resultant cheese



pieces are arranged in layers into plastic containers, covered with salted whey (4%) and the containers are tightly closed and pickled at  $6\pm 1^\circ\text{C}$  for 45 days. Cheese samples were withdrawn when fresh and after 15, 30 and 45 days for chemical, microbiological analysis and organoleptic evaluation.

All experiments were performed in triplicates and each analysis in triplicates and average results were recorded.

### Methods of analysis

#### Chemical analysis

Samples of white soft cheese were thoroughly ground in a clean and dry mortar and analyzed for some of their chemical components. The following methods were adopted: The moisture and salt contents were determined using a thermostatically controlled oven at  $105^\circ\text{C}$  for 3 hrs and direct titration was made, respectively as mentioned by Bradley *et al.* (1992). The standard Gerber method for fat determination in milk and cheese samples was used as described by Ling (1963). Titratable acidity (TA %) of milk and cheese samples were determined by titrametric method as described in the A.O.A.C. (2000). The pH values of milk and cheese (samples prepared by homogenization of 10 g cheese with 5 ml distilled water) were determined using a digital pH meter (540-GLP, Multical., Germany) The total nitrogen content (TN), water-soluble nitrogen (WSN) and non-protein nitrogen (NPN) were estimated using macro-Kjeldahl method as described by the A.O.A.C. (1998). Total volatile fatty acids (TVFA) in cheese samples were determined by the direct distillation method as described by Kosikowski, (1978), values were expressed as ml NaOH 0.1N /100g cheese

Cheese samples were prepared to determine Potentiometric acid-base titration by a modified method of Kirchmeier (1977) as follow: \*

- 1- Ten g cheese were kneaded in a mortar with 6 ml ( $3 \times 2$  ml) NaOH (0.25 N) and 5 ml saturated urea solution and diluted to 100 ml with saturated urea solution and then filtered, using filter paper whatman No 42.
- 2- Potentiometric titration was carried out by the following procedure: Twenty ml of the filtrate were added with 80 ml NaCl (0.01N). The pH value of cheese sample was adjusted to the required step with drops of NaOH (3N) or HCl (3N). Cheese samples were titrated from pH 7.0

to pH 5.0 in 0.2 pH-intervals using HCl (0.01N) at a constant temperature and constant stirring speed. The time until each (seeming) equilibrium was fixed for 40 sec. After reaching pH 5.0, back titration to pH 7.0 was done using NaOH (0.01N), using the same procedure.

Results of the obtained potentiometric titrations were plotted in form of integral curve, in terms of  $\alpha$  as a function of pH. A single curve was obtained by the potentiometric acid-base titration of true solution from pH 7.0 to 5.0. In contrast, by the acid-base titration of colloidal solution (such milk) from pH 7.0 to 5.0, two different curves for the forward and back titration were obtained. The lower curve is always the acid branch and the upper curve is the base branch. It can be seen that the acid titration curve and the base curve forms a hysteresis loop.

The total consumption of titrants in milliliter is considered as the degree of proteonation  $\alpha = 1.0$ . The mean degree of proteonation  $\alpha$  is defined as the ratio of the concentration of bound protons [HA] to the total concentration of partners [HA] + [A<sup>-</sup>], (Neumann, 1973).  $\alpha = [\text{HA}]/[\text{HA}] + [\text{A}^-]$ . The quantitative measurement of hysteresis curve is performed with the closed integration of ( $\oint \alpha \text{d pH}$ ) which represents the hysteresis area. This value of  $\oint \alpha \text{d pH}$  is obtained by a simple method through measuring the hysteresis loop and ( $1.0\alpha \times 1.0 \text{ pH}$ ) unit using planimeter (digital planimeter, model planix 7).

#### Microbiological analysis

Total viable count was determined using plate count agar (PCA) medium as described by Bridson (1990). The plates were incubated at  $37\pm 2^\circ\text{C}$  for 24 hrs. Psychrophilic bacterial count was estimated by using PCS medium (Bridson, 1990). The plates were incubated at  $5-7^\circ\text{C}$  for 10 days. yeasts and moulds were estimated as described in the Standard Methods for the Examination of Dairy Products (1992). The plates were incubated at  $25\pm 2^\circ\text{C}$  for 4 days. Most probable number (MPN) of coliform bacteria was estimated using MacConkey broth as described by Bridson (1990). The tubes were incubated at  $37\pm 2^\circ\text{C}$  for 48 hrs.

#### Organoleptic evaluation

Cheese samples were evaluated during the pickling period by a 15 panelists of the experienced staff members of Microbiology, Food Science & Technology and Dairy Science Departments, Faculty of Agriculture, Fayoum University. Cheese samples



were evaluated for flavour (50 points), body & texture (35 points) and colour & appearance (15 points) as described by Nelson & Torut, (1981).

### Statistical analysis

All experiments were performed in triplicate and the results obtained were analyzed statistically using General linear Models (GLM) according to SPSS (1999), Version 9. Significant differences among treatments, pickling periods and the interaction mean between them were compared ( $P \leq 0.05$ ) according to Duncan's multiple range test (Duncan, 1955).

## RESULTS AND DISCUSSION

### Gross composition of buffalos' and goats' milk

Data presented in Table (1) show the chemical composition of buffalos' and goats' milk used in white soft cheese manufacture. It is clear that buffalos' milk had percentages of fat, protein, lactose, total solids (TS), solids not fat (SNF), and ash higher than their corresponding values of goats' milk, while both types contained similar values of TA (0.17 & 0.18) and pH (6.7 & 6.6) for buffalos' and goats' milk, respectively. These values are in accordance with those obtained by El-Almy *et al.* (1990). In contrast, Abo-Dawood *et al.* (1980a & b) found that fat, TS and protein contents of Egyptian goats' milk were higher than results obtained in the present study.

**Table 1: Gross composition of buffalos' and goats' milk used in white soft cheese manufacture**

Parameters (%)	Buffalos' milk	Goats' milk
Fat	6.78	3.80
Protein	3.99	3.20
Lactose	5.43	4.80
Total solids	17.31	13.15
Solids not fat	10.53	9.35
Ash	0.90	0.76

### Chemical composition of cheese

Table (2) summarizes the moisture, fat and salt contents of white soft cheese made from buffalos' and goats' milk as affected by adding some spice oils during the pickling at  $6 \pm 1^\circ\text{C}$  for 45 days. It is clear that control I retained higher moisture than control II, being 63.68 and 61.45 % in the fresh samples, which reached 62.91 and 59.90% at the end of the storage period for control I and II, respectively. This could be attributed to the differences in the

physical and chemical properties between buffalos' and goats' milk. On the other hand, the moisture of all cheese samples increased slightly throughout the first 15 days of the pickling period. These findings could be attributed to some absorption of pickling solution by low salt cheese that stored at low temperature ( $6 \pm 1^\circ\text{C}$ ). After that, moisture (%) significantly ( $P \leq 0.05$ ) decreased in all cheeses up to the end of the storage period. The loss in moisture could be attributed to the increase of acidity, which had a contractive effect on the cheese causing expulsion of moisture. Similar results were reported by Hamed *et al.* (1992) and Salama (2004). Addition of cardamom, thyme or clove essential oils to the cheese milk at different ratios resulted in a significant increase in the moisture of the all samples of the resultant cheese as compared to control II. This could be attributed to the significant continuous increase in acidity until the end of storage in the control II (Table 3) as compared to other treatments.

As it was expected, the fat (%) of control I was higher than that made from goats' milk. This result is in accordance with that reported by Sevda *et al.* (2004) that the lower fat was closely related to the fat content of milk used in manufacturing of cheese and to the whey drainage. On the other hand, fat of goat cheeses treated with cardamom, thyme and clove were slightly higher than the control II.

It is noteworthy to mention that the fat contents of all cheese samples (Table 2) are complying with the Egyptian legal standards (EOSQC, 2000) for fresh or pickled white soft cheese made from buffalos' milk and that made from goats' milk. Buffalos' cheese gained the highest values of fat either when being fresh or during the pickling period. The results indicate that fat contents did not show considerable differences between different treatments and control II at zero time, with the same trend of cheese fat content throughout the pickling period. The  $T_2$  and  $T_3$  treatments showed the highest fat contents after 15 days. Statistically, the fat content showed a significant increase between control I and II, whereas pickling period and the interaction between treatments and the pickling period were not affected significantly ( $P \leq 0.05$ ).

As shown in Table (2), at the beginning of the pickling period, there were no significant differences between the treatments, the percentage of salt of all cheeses slightly decreased at 15 days, then continued to increase till the end of pickling period. There was a significant difference for salt %



**Table 2: Effect of cardamom, thyme and clove essential oils on the moisture, fat / dry matter, salt and salt / moisture of goats' white soft cheese during the pickling period**

Treatment	Pickling period (days) × Treatment interaction				Treatment effect
	Fresh	15	30	45	
<b>Moisture (%)</b>					
Control I	63.68 <sup>a</sup>	66.18 <sup>a</sup>	64.61 <sup>a</sup>	62.91 <sup>a</sup>	64.35 ± 0.78 <sup>A</sup>
Control II	61.45 <sup>a</sup>	63.25 <sup>a</sup>	61.61 <sup>a</sup>	59.90 <sup>a</sup>	61.55 ± 0.54 <sup>A</sup>
T <sub>1</sub>	61.34 <sup>a</sup>	63.26 <sup>a</sup>	61.71 <sup>a</sup>	60.13 <sup>a</sup>	61.61 ± 0.75 <sup>A</sup>
T <sub>2</sub>	61.39 <sup>a</sup>	63.79 <sup>a</sup>	61.80 <sup>a</sup>	60.32 <sup>a</sup>	61.83 ± 0.66 <sup>A</sup>
T <sub>3</sub>	61.41 <sup>a</sup>	63.64 <sup>a</sup>	61.82 <sup>a</sup>	60.24 <sup>a</sup>	61.78 ± 0.76 <sup>A</sup>
T <sub>4</sub>	61.43 <sup>a</sup>	63.72 <sup>a</sup>	61.85 <sup>a</sup>	60.43 <sup>a</sup>	61.86 ± 0.71 <sup>A</sup>
T <sub>5</sub>	61.42 <sup>a</sup>	63.42 <sup>a</sup>	61.65 <sup>a</sup>	59.97 <sup>a</sup>	61.62 ± 0.78 <sup>A</sup>
T <sub>6</sub>	61.40 <sup>a</sup>	63.40 <sup>a</sup>	61.78 <sup>a</sup>	60.38 <sup>a</sup>	61.74 ± 0.74 <sup>A</sup>
Pickling period effect	61.69 ± 0.5 <sup>BC</sup>	63.83 ± 0.46 <sup>A</sup>	62.10 ± 0.44 <sup>B</sup>	60.54 ± 0.47 <sup>C</sup>	
<b>Fat / dry matter (%)</b>					
Control I	51.21 <sup>a</sup>	51.45 <sup>a</sup>	51.43 <sup>a</sup>	51.50 <sup>a</sup>	51.40 ± 0.55 <sup>A</sup>
Control II	45.65 <sup>a</sup>	45.71 <sup>a</sup>	45.06 <sup>a</sup>	44.39 <sup>a</sup>	45.20 ± 0.34 <sup>B</sup>
T <sub>1</sub>	45.53 <sup>a</sup>	46.81 <sup>a</sup>	45.42 <sup>a</sup>	45.02 <sup>a</sup>	45.69 ± 0.55 <sup>B</sup>
T <sub>2</sub>	45.58 <sup>a</sup>	47.22 <sup>a</sup>	45.68 <sup>a</sup>	45.11 <sup>a</sup>	45.90 ± 0.47 <sup>B</sup>
T <sub>3</sub>	45.61 <sup>a</sup>	46.62 <sup>a</sup>	46.10 <sup>a</sup>	45.27 <sup>a</sup>	45.90 ± 0.52 <sup>B</sup>
T <sub>4</sub>	45.63 <sup>a</sup>	45.86 <sup>a</sup>	45.61 <sup>a</sup>	45.11 <sup>a</sup>	45.55 ± 0.46 <sup>B</sup>
T <sub>5</sub>	45.62 <sup>a</sup>	46.22 <sup>a</sup>	45.38 <sup>a</sup>	44.59 <sup>a</sup>	45.45 ± 0.54 <sup>B</sup>
T <sub>6</sub>	45.60 <sup>a</sup>	46.98 <sup>a</sup>	45.79 <sup>a</sup>	45.18 <sup>a</sup>	45.89 ± 0.53 <sup>B</sup>
Pickling period effect	46.30 ± 0.53 <sup>A</sup>	47.11 ± 0.47 <sup>A</sup>	46.31 ± 0.51 <sup>A</sup>	45.77 ± 0.50 <sup>A</sup>	
<b>Salt (%)</b>					
Control I	2.97 <sup>a</sup>	2.80 <sup>a</sup>	3.07 <sup>a</sup>	3.26 <sup>a</sup>	3.01 ± 0.06 <sup>C</sup>
Control II	3.04 <sup>a</sup>	2.93 <sup>a</sup>	3.18 <sup>a</sup>	3.34 <sup>a</sup>	3.12 ± 0.05 <sup>A</sup>
T <sub>1</sub>	3.04 <sup>a</sup>	2.90 <sup>a</sup>	3.10 <sup>a</sup>	3.30 <sup>a</sup>	3.09 ± 0.05 <sup>AB</sup>
T <sub>2</sub>	3.08 <sup>a</sup>	2.83 <sup>a</sup>	3.05 <sup>a</sup>	3.20 <sup>a</sup>	3.04 ± 0.04 <sup>BC</sup>
T <sub>3</sub>	3.04 <sup>a</sup>	2.88 <sup>a</sup>	3.00 <sup>a</sup>	3.25 <sup>a</sup>	3.04 ± 0.04 <sup>BC</sup>
T <sub>4</sub>	3.06 <sup>a</sup>	2.85 <sup>a</sup>	3.00 <sup>a</sup>	3.15 <sup>a</sup>	3.02 ± 0.04 <sup>BC</sup>
T <sub>5</sub>	3.08 <sup>a</sup>	2.90 <sup>a</sup>	3.15 <sup>a</sup>	3.20 <sup>a</sup>	3.08 ± 0.04 <sup>BC</sup>
T <sub>6</sub>	3.04 <sup>a</sup>	2.90 <sup>a</sup>	3.08 <sup>a</sup>	3.35 <sup>a</sup>	3.09 ± 0.05 <sup>AB</sup>
Pickling period effect	3.04 ± .01 <sup>B</sup>	2.88 ± 0.02 <sup>C</sup>	3.07 ± 0.02 <sup>B</sup>	3.26 ± 0.02 <sup>A</sup>	
<b>Salt in water phase (%)</b>					
Control I	4.66	4.23	4.75	5.18	
Control II	4.95	4.63	5.16	5.58	
T <sub>1</sub>	4.96	4.58	5.02	5.49	
T <sub>2</sub>	5.02	4.44	4.94	5.31	
T <sub>3</sub>	4.95	4.53	4.85	5.40	
T <sub>4</sub>	4.98	4.47	4.85	5.21	
T <sub>5</sub>	5.02	4.57	5.11	5.34	
T <sub>6</sub>	4.95	4.57	4.99	5.55	

A, B and C: Means within the same effect having different capital superscripts are significantly different ( $P \leq 0.05$ ).

a,b,...and o: Means within the same interaction having different small superscripts are significantly different ( $P \leq 0.05$ ).

Control I and II = white soft cheese made from buffalos' and goats' milk, respectively.

T<sub>1</sub> & T<sub>2</sub> = goats' white soft cheese contained 75 and 90 ppm cardamom essential oil, respectively.

T<sub>3</sub> & T<sub>4</sub> = goats' white soft cheese contained 50 and 75 ppm thyme essential oil, respectively.

T<sub>5</sub> & T<sub>6</sub> = goats' white soft cheese contained 40 and 60 ppm clove essential oil, respectively.

between control I and II, also during the pickling periods (from 15 up to 45 days), but the interaction between treatments and the pickling period was not affected significantly ( $P \leq 0.05$ ).

#### Titratable acidity (TA) and pH values

Changes occurred in the TA and pH values of various white soft cheese samples during the pickling period are presented in Table (3). The TA of all cheeses was found to increase progressively during the pickling period. These results are in agreement with those of Mehanna & Hefnawy (1991) and Hamed *et al.* (1992). The gradual increase of the TA of all cheese samples could be attributed to the fermentation of lactose to lactic acid by lactic acid bacteria. On the other hand, the results did not reveal any significant differences between all treatments at zero time. Fresh cheese of control I had the lowest TA (0.21%), while control II had the highest value of TA either when fresh or stored for 45 days (0.23 and 1.05%, respectively). The T<sub>4</sub> treatment had the lowest value of TA followed by

T<sub>2</sub> and T<sub>6</sub> treatments in an ascending order as compared with control II and other treatments at the end of 45 days pickling period. This could be attributed to inhibition of the growth of cheese microflora by added essential oils especially at the high concentrations. These results are in agreement with those of Abd-Alla *et al.* (2000). Statistically, TA was significantly influenced by treatments, pickling period and the interaction between treatments and the pickling period ( $P \leq 0.05$ ).

It is clear from the results given in Table (3) that the pH decreased during the pickling period in all samples, with an opposite trend of TA. These results are similar to those found by Mallatou *et al.* (1994). There were no differences between goats' cheese treatments at the first stage of pickling (fresh cheese), while at 15 days T<sub>2</sub> treatment had the highest pH value (5.90) followed by 5.80, 5.75, 5.70, 5.65, 5.60, 5.50 and 5.45 in a descending order for T<sub>4</sub>, T<sub>3</sub>, T<sub>6</sub>, T<sub>5</sub>, T<sub>1</sub>, control I and control II, respectively. At 30 days, the pH values were in the order of T<sub>4</sub> > T<sub>3</sub> > T<sub>6</sub> = T<sub>2</sub> > T<sub>1</sub> > T<sub>5</sub> > control II >

**Table 3: Effect of cardamom, thyme and clove essential oils on the titratable acidity and pH values of goats' white soft cheese during the pickling period**

Treatments	Pickling period (days) × Treatment interaction				Treatment effect
	Fresh	15	30	45	
	<b>Titratable acidity (%)</b>				
Control I	0.21 <sup>n</sup>	0.45 <sup>klm</sup>	0.65 <sup>g</sup>	0.90 <sup>b</sup>	0.55 ± 0.08 <sup>B</sup>
Control II	0.23 <sup>n</sup>	0.53 <sup>ij</sup>	0.70 <sup>f</sup>	1.05 <sup>a</sup>	0.63 ± 0.09 <sup>A</sup>
T <sub>1</sub>	0.22 <sup>n</sup>	0.49 <sup>jk</sup>	0.58 <sup>g</sup>	0.86 <sup>c</sup>	0.54 ± 0.07 <sup>B</sup>
T <sub>2</sub>	0.22 <sup>n</sup>	0.40 <sup>m</sup>	0.56 <sup>gh</sup>	0.82 <sup>d</sup>	0.50 ± 0.07 <sup>C</sup>
T <sub>3</sub>	0.22 <sup>n</sup>	0.45 <sup>klm</sup>	0.54 <sup>ij</sup>	0.84 <sup>d</sup>	0.51 ± 0.07 <sup>C</sup>
T <sub>4</sub>	0.22 <sup>n</sup>	0.43 <sup>lm</sup>	0.54 <sup>ij</sup>	0.79 <sup>e</sup>	0.50 ± 0.06 <sup>C</sup>
T <sub>5</sub>	0.22 <sup>n</sup>	0.47 <sup>kl</sup>	0.60 <sup>g</sup>	0.92 <sup>b</sup>	0.55 ± 0.08 <sup>B</sup>
T <sub>6</sub>	0.22 <sup>n</sup>	0.45 <sup>klm</sup>	0.56 <sup>gh</sup>	0.82 <sup>d</sup>	0.51 ± 0.07 <sup>C</sup>
Pickling period effect	0.22 ± 0.003 <sup>D</sup>	0.46 ± 0.008 <sup>C</sup>	0.59 ± 0.01 <sup>B</sup>	0.88 ± 0.02 <sup>A</sup>	
	<b>pH</b>				
Control I	6.45	5.50	4.95	4.70	-
Control II	6.30	5.45	5.00	4.80	-
T <sub>1</sub>	6.35	5.60	5.35	5.05	-
T <sub>2</sub>	6.35	5.90	5.40	5.20	-
T <sub>3</sub>	6.35	5.75	5.50	5.15	-
T <sub>4</sub>	6.35	5.80	5.60	5.30	-
T <sub>5</sub>	6.35	5.65	5.20	4.95	-
T <sub>6</sub>	6.35	5.70	5.40	5.20	-

A, B, C, D and E: Means within the same effect having different capital superscripts are significantly different ( $P \leq 0.05$ ).

a, b, ... and o: Means within the same interaction having different small superscripts are significantly different ( $P \leq 0.05$ ).

Control I and II = white soft cheese made from buffalos' and goats' milk, respectively.

T<sub>1</sub> & T<sub>2</sub> = goats' white soft cheese contained 75 and 90 ppm cardamom essential oil, respectively.

T<sub>3</sub> & T<sub>4</sub> = goats' white soft cheese contained 50 and 75 ppm thyme essential oil, respectively.

T<sub>5</sub> & T<sub>6</sub> = goats' white soft cheese contained 40 and 60 ppm clove essential oil, respectively.



control I. On the other hand, control II had the lowest value of pH while T<sub>4</sub> treatment had the highest value of pH at the end of the pickling period. Addition of cardamom, thyme and clove essential oils to cheese milk, especially at high concentrations, led to increase the pH values of the resultant cheese as compared to other treatments.

Table (4) illustrates the changes in total nitrogen (TN) and total nitrogen/ dry matter (TN/DM) of white soft cheese produced from buffalos', untreated and treated goats' milk. The TN decreased in all cheese samples throughout the first 15 days of the pickling period. This decrease was associated with an increase of moisture content in this stage. There was a noticeable gradual increase in TN of cheese produced from all treatments after 15 days and until the end of the pickling period. This is in accordance with that reported by Ahmed & Abd El-Razig (1998) and Kandeel *et al.* (1991). The TN of fresh and stored cheese for 45 days of control I had a relatively higher value than that in control II and other treatments. Such greater values could be attributed to higher TN of buffalos' milk (Table 1). On the other hand, there were no noticeable differences in the TN among samples from all treatments at zero time whereas the values ranged between 2.43 to 2.45% during the pickling period. Control I at 15 days of the pickling period had the highest value of TN (2.32%), but T<sub>2</sub> treatment had the lowest value (2.02%) as compared with other goats' cheeses. At 30 days of the pickling period, the values of TN ranged between 2.23 to 2.60 %, whereas it ranged between 2.6 to 2.80% at the end of the pickling period. Significant differences ( $P \leq 0.05$ ) were found between cheeses of control I and that of control II during the pickling period. The interaction between treatments and pickling period did not affect significantly TN content of all cheese treatments.

#### Pickling degree indices

Water-soluble nitrogen/total nitrogen (WSN/TN) and non-protein nitrogen/total nitrogen (NPN/TN) as affected by using buffalos' or goats' milk either alone or mixed with two concentrations of cardamom, thyme and clove essential oils are presented in Table (4). It is clear that WSN/TN and NPN/TN ratios increased with prolonged the pickling period of all cheeses samples. These results are in harmony with those reported by many workers (Ahmed & Abd-El Razig, 1998, Salama ,2004 , Ismail *et al.*, 2006 ). Also, the results showed that WSN/TN and NPN/TN were significantly ( $P \leq 0.05$ )

lower in fresh and stored cheese of control I as compared to cheese of control II. These high values of WSN/TN and NPN/TN in fresh or stored cheeses of control II and other treatments are due to the nature of each buffalo and goat casein; also coincide with the high numbers obtained from the total viable counts and psychrophilic counts (Table 6), which are responsible for protein breakdown. At the beginning of the pickling period, there were no significant differences among control II and different treatments (T<sub>1</sub> to T<sub>6</sub>). Also, WSN/TN and NPN/TN ratios of control II were significantly ( $P \leq 0.05$ ) higher than cheeses from all treatments, this could be attributed to the inhibition action of essential oils towards proteolytic activity which is responsible for protein degradation. It can be noted that as the WSN and NPN content decreased, the ratio of essential oil increased. The T<sub>2</sub> treatment showed the lowest value of WSN followed by T<sub>4</sub> and T<sub>6</sub> in an ascending order after 15 days of the pickling period. On the other hand, T<sub>4</sub> treatment showed the lowest value of WSN, while control II had the highest value of WSN after 45 days of pickling; this could be attributed to the effect of thyme essential oils (75ppm) on the microbial growth and activity.

#### Titration curves (hysteresis indicator)

During cheese processing and pickling period, proteins are physically and chemically changed through the action of rennet and microbial enzymes. More direct information about the changes of cheese proteins during the different periods of pickling (fresh, 15, 30 and 45 day) can be derived from the potentiometric results.

#### Hysteresis size

The obtained results of acid-base potentiometric titration of goats' white soft cheese as affected by adding some spice essential oils as compared to control buffalos' cheese during the pickling period are integrally plotted in the form of pH-values as a function of protonation degree ( $\alpha$ ) (Figs. 1 to 8). The total consumption of milliliters titrants are considered as a degree of protonation  $\alpha = 1.0$ . It can be shown that acid titration curve and the base curve are a form of hysteresis loop. The size of hysteresis loop is given by  $\oint \alpha d\text{pH}$ . The results showed that the shape of hysteresis loop of buffalos' cheese was greater than that of goats' cheese. These results confirm those reported by El-Shobery (1988), while the hysteresis loop of control goats' cheese was smaller than that of all treated goats' cheese throughout the



**Table 4: Effect of cardamom, thyme and clove essential oils on the total nitrogen, total nitrogen / dry matter, water soluble nitrogen / total nitrogen and non protein nitrogen / total nitrogen content of goats' white soft cheese during the pickling period**

Treatments	Pickling period (days) × Treatment interaction				Treatment effect
	Fresh	15	30	45	
<b>Total nitrogen (%)</b>					
Control I	2.60 <sup>a</sup>	2.32 <sup>a</sup>	2.60 <sup>a</sup>	2.80 <sup>a</sup>	2.58 ± 0.06 <sup>A</sup>
Control II	2.44 <sup>a</sup>	2.28 <sup>a</sup>	2.41 <sup>a</sup>	2.75 <sup>a</sup>	2.47 ± 0.07 <sup>B</sup>
T <sub>1</sub>	2.45 <sup>a</sup>	2.23 <sup>a</sup>	2.38 <sup>a</sup>	2.70 <sup>a</sup>	2.44 ± 0.06 <sup>BCD</sup>
T <sub>2</sub>	2.43 <sup>a</sup>	2.02 <sup>a</sup>	2.32 <sup>a</sup>	2.65 <sup>a</sup>	2.36 ± 0.07 <sup>DE</sup>
T <sub>3</sub>	2.43 <sup>a</sup>	2.15 <sup>a</sup>	2.27 <sup>a</sup>	2.72 <sup>a</sup>	2.39 ± 0.07 <sup>CDE</sup>
T <sub>4</sub>	2.44 <sup>a</sup>	2.11 <sup>a</sup>	2.23 <sup>a</sup>	2.60 <sup>a</sup>	2.35 ± 0.06 <sup>E</sup>
T <sub>5</sub>	2.44 <sup>a</sup>	2.20 <sup>a</sup>	2.47 <sup>a</sup>	2.72 <sup>a</sup>	2.46 ± 0.06 <sup>BC</sup>
T <sub>6</sub>	2.44 <sup>a</sup>	2.18 <sup>a</sup>	2.35 <sup>a</sup>	2.63 <sup>a</sup>	2.40 ± 0.06 <sup>CDE</sup>
Pickling period effect	2.46 ± 0.02 <sup>B</sup>	2.19 ± 0.02 <sup>D</sup>	2.38 ± 0.03 <sup>C</sup>	2.69 ± 0.03 <sup>A</sup>	
<b>Total nitrogen / Dry matter (%)</b>					
Control I	7.16	6.86	7.35	7.55	
Control II	6.33	6.20	6.28	6.86	
T <sub>1</sub>	6.34	6.07	6.22	6.77	
T <sub>2</sub>	6.29	5.58	6.07	6.68	
T <sub>3</sub>	6.30	5.91	5.95	6.84	
T <sub>4</sub>	6.33	5.82	5.85	6.57	
T <sub>5</sub>	6.32	6.01	6.44	6.79	
T <sub>6</sub>	6.32	5.96	6.15	6.64	
<b>Water soluble nitrogen / total nitrogen</b>					
Control I	5.75 <sup>t</sup>	7.24 <sup>s</sup>	7.69 <sup>q</sup>	8.57 <sup>p</sup>	7.29 ± 0.32 <sup>F</sup>
Control II	7.58 <sup>q</sup>	12.28 <sup>hi</sup>	14.52 <sup>b</sup>	17.09 <sup>a</sup>	12.87 ± 0.10 <sup>A</sup>
T <sub>1</sub>	7.35 <sup>qrs</sup>	11.66 <sup>k</sup>	12.61 <sup>g</sup>	14.07 <sup>c</sup>	11.42 ± 0.76 <sup>B</sup>
T <sub>2</sub>	7.24 <sup>s</sup>	9.90 <sup>o</sup>	12.07 <sup>ij</sup>	13.21 <sup>e</sup>	10.61 ± 0.69 <sup>D</sup>
T <sub>3</sub>	7.41 <sup>qrs</sup>	10.23 <sup>n</sup>	11.89 <sup>jk</sup>	12.50 <sup>gh</sup>	10.51 ± 0.60 <sup>D</sup>
T <sub>4</sub>	7.38 <sup>qrs</sup>	10.43 <sup>mn</sup>	11.66 <sup>k</sup>	11.92 <sup>j</sup>	10.35 ± 0.54 <sup>E</sup>
T <sub>5</sub>	7.38 <sup>qrs</sup>	10.91 <sup>l</sup>	12.96 <sup>f</sup>	14.71 <sup>b</sup>	11.49 ± 0.82 <sup>B</sup>
T <sub>6</sub>	7.38 <sup>qrs</sup>	10.55 <sup>m</sup>	12.34 <sup>h</sup>	13.69 <sup>d</sup>	10.99 ± 0.71 <sup>C</sup>
Pickling period effect	7.17 ± 0.12 <sup>D</sup>	10.40 ± 0.29 <sup>C</sup>	11.97 ± 0.38 <sup>B</sup>	13.22 ± 0.45 <sup>A</sup>	
<b>Non protein nitrogen / Total nitrogen</b>					
Control I	0.08 <sup>m</sup>	0.10 <sup>kl</sup>	0.12 <sup>ij</sup>	0.18 <sup>bcd</sup>	0.12 ± 0.01 <sup>C</sup>
Control II	0.08 <sup>m</sup>	0.13 <sup>hi</sup>	0.17 <sup>de</sup>	0.22 <sup>a</sup>	0.15 ± 0.02 <sup>A</sup>
T <sub>1</sub>	0.08 <sup>m</sup>	0.12 <sup>ij</sup>	0.15 <sup>fg</sup>	0.19 <sup>bc</sup>	0.12 ± 0.01 <sup>C</sup>
T <sub>2</sub>	0.09 <sup>m</sup>	0.11 <sup>jk</sup>	0.13 <sup>hi</sup>	0.17 <sup>de</sup>	0.14 ± 0.01 <sup>B</sup>
T <sub>3</sub>	0.08 <sup>m</sup>	0.10 <sup>kl</sup>	0.13 <sup>hi</sup>	0.18 <sup>bcd</sup>	0.12 ± 0.01 <sup>C</sup>
T <sub>4</sub>	0.08 <sup>m</sup>	0.10 <sup>kl</sup>	0.12 <sup>ij</sup>	0.15 <sup>fg</sup>	0.11 ± 0.01 <sup>D</sup>
T <sub>5</sub>	0.08 <sup>m</sup>	0.12 <sup>ij</sup>	0.16 <sup>ef</sup>	0.20 <sup>b</sup>	0.14 ± 0.01 <sup>B</sup>
T <sub>6</sub>	0.08 <sup>m</sup>	0.11 <sup>jk</sup>	0.14 <sup>gh</sup>	0.17 <sup>de</sup>	0.12 ± 0.006 <sup>C</sup>
Pickling period effect	0.08 ± 0.002 <sup>D</sup>	0.11 ± 0.003 <sup>C</sup>	0.14 ± 0.004 <sup>B</sup>	0.18 ± 0.004 <sup>A</sup>	

A, B and C: Means within the same effect having different capital superscripts are significantly different ( $P \leq 0.05$ ).  
a,b,...and o: Means within the same interaction having different small superscripts are significantly different ( $P \leq 0.05$ )  
Control I and II = white soft cheese made from buffalos' and goats' milk, respectively.

T<sub>1</sub> & T<sub>2</sub> = goats' white soft cheese contained 75 and 90 ppm cardamom essential oil, respectively.

T<sub>3</sub> & T<sub>4</sub> = goats' white soft cheese contained 50 and 75 ppm thyme essential oil, respectively.

T<sub>5</sub> & T<sub>6</sub> = goats' white soft cheese contained 40 and 60 ppm clove essential oil, respectively.



**Table 5: Effect of cardamom, thyme and clove essential oils on the total volatile fatty acids\*\* of goats' white soft cheese the during the pickling period**

Treatments	Pickling period (days) × Treatment interaction				Treatment effect
	Fresh	15	30	45	
Control I	3.0 <sup>o</sup>	5.0 <sup>n</sup>	7.0 <sup>m</sup>	9.0 <sup>k</sup>	06.00 ± 0.68 <sup>E</sup>
Control II	7.9 <sup>l</sup>	13.7 <sup>h</sup>	20.0 <sup>cd</sup>	24.5 <sup>a</sup>	16.53 ± 1.9 <sup>A</sup>
T <sub>1</sub>	7.8 <sup>l</sup>	12.0 <sup>ij</sup>	17.0 <sup>fg</sup>	21.0 <sup>bc</sup>	14.45 ± 1.5 <sup>C</sup>
T <sub>2</sub>	7.8 <sup>l</sup>	11.5 <sup>ij</sup>	16.0 <sup>g</sup>	20.50 <sup>bcd</sup>	13.95 ± 1.4 <sup>CD</sup>
T <sub>3</sub>	7.9 <sup>l</sup>	12.5 <sup>i</sup>	18.5 <sup>e</sup>	21.50 <sup>b</sup>	15.10 ± 1.6 <sup>B</sup>
T <sub>4</sub>	7.8 <sup>l</sup>	11.5 <sup>ij</sup>	17.5 <sup>f</sup>	21.0 <sup>bc</sup>	14.45 ± 1.6 <sup>C</sup>
T <sub>5</sub>	7.8 <sup>l</sup>	12.0 <sup>ij</sup>	17.0 <sup>fg</sup>	20.0 <sup>cd</sup>	14.20 ± 1.4 <sup>C</sup>
T <sub>6</sub>	7.8 <sup>l</sup>	11.0 <sup>j</sup>	16.5 <sup>fg</sup>	19.5 <sup>de</sup>	13.70 ± 1.4 <sup>D</sup>
Pickling period effect	7.23 ± 0.4 <sup>D</sup>	11.15 ± 0.5 <sup>C</sup>	16.19 ± 0.8 <sup>B</sup>	19.63 ± 0.9 <sup>A</sup>	

**Table 6: Effect of cardamom, thyme and clove essential oils on the total viable count, psychrophilic count, total yeast & mould count of goats' white soft cheese during the pickling period**

Treatments	Pickling period (days) × Treatment interaction				Treatment effect
	Fresh	15	30	45	
<b>Total viable count (count × 10<sup>6</sup> cfu*/gm)</b>					
Control I	3.0 <sup>gh</sup>	17.0 <sup>fg</sup>	81.0 <sup>c</sup>	310 <sup>b</sup>	102.8 ± 37.2 <sup>B</sup>
Control II	36 <sup>e</sup>	59.0 <sup>d</sup>	98.0 <sup>c</sup>	460 <sup>a</sup>	163.3 ± 52.1 <sup>A</sup>
T <sub>1</sub>	27 <sup>fg</sup>	6.8 <sup>gh</sup>	2.0 <sup>h</sup>	9.6 <sup>gh</sup>	11.6 ± 2.97 <sup>D</sup>
T <sub>2</sub>	30 <sup>ef</sup>	0.98 <sup>h</sup>	1.3 <sup>h</sup>	8.2 <sup>gh</sup>	10.1 ± 3.58 <sup>D</sup>
T <sub>3</sub>	24 <sup>ef</sup>	7.1 <sup>gh</sup>	0.9 <sup>h</sup>	9.0 <sup>gh</sup>	10.8 ± 2.82 <sup>D</sup>
T <sub>4</sub>	25 <sup>ef</sup>	4.8 <sup>gh</sup>	0.91 <sup>h</sup>	5.5 <sup>gh</sup>	8.8 ± 2.70 <sup>D</sup>
T <sub>5</sub>	23 <sup>ef</sup>	6.3 <sup>gh</sup>	3.2 <sup>h</sup>	36.0 <sup>e</sup>	17.13 ± 4.00 <sup>C</sup>
T <sub>6</sub>	25 <sup>ef</sup>	4.2 <sup>gh</sup>	1.2 <sup>h</sup>	8.0 <sup>gh</sup>	9.6 ± 2.87 <sup>D</sup>
Pickling Period effect	24.38 ± 1.88 <sup>B</sup>	13.27 ± 3.72 <sup>C</sup>	23.56 ± 7.99 <sup>B</sup>	105.8 ± 34.58 <sup>A</sup>	
<b>Psychrophilic count (count × 10<sup>4</sup> cfu /gm)</b>					
Control I	2.08 <sup>r</sup>	12 <sup>r</sup>	160 <sup>l</sup>	800 <sup>i</sup>	243.5 ± 98.7 <sup>G</sup>
Control II	400 <sup>j</sup>	900 <sup>h</sup>	6000 <sup>c</sup>	38000 <sup>a</sup>	11325 ± 4690 <sup>A</sup>
T <sub>1</sub>	350 <sup>k</sup>	130 <sup>l</sup>	70 <sup>mnpq</sup>	5600 <sup>d</sup>	1538 ± 708 <sup>C</sup>
T <sub>2</sub>	300 <sup>k</sup>	96 <sup>lmn</sup>	25 <sup>qr</sup>	1400 <sup>f</sup>	180.3 ± 36.9 <sup>H</sup>
T <sub>3</sub>	300 <sup>k</sup>	91 <sup>lmno</sup>	40 <sup>opqr</sup>	3400 <sup>e</sup>	957 ± 426 <sup>D</sup>
T <sub>4</sub>	310 <sup>k</sup>	72 <sup>mnpq</sup>	30 <sup>pqr</sup>	970 <sup>g</sup>	345.5 ± 113 <sup>F</sup>
T <sub>5</sub>	320 <sup>k</sup>	110 <sup>lm</sup>	80 <sup>lmnop</sup>	9600 <sup>b</sup>	2528 ± 1232 <sup>B</sup>
T <sub>6</sub>	350 <sup>k</sup>	95 <sup>lmn</sup>	55 <sup>nopqr</sup>	1700 <sup>f</sup>	550 ± 203.1 <sup>E</sup>
Pickling Period effect	291.5 ± 23.8 <sup>C</sup>	213.8 ± 56.4 <sup>D</sup>	816.4 ± 409 <sup>B</sup>	7512 ± 2481 <sup>A</sup>	
<b>Total yeast &amp; mould (count × 10<sup>2</sup> cfu /gm)</b>					
Control I	1.5 <sup>klm</sup>	3.5 <sup>hi</sup>	6.5 <sup>g</sup>	20.0 <sup>d</sup>	7.88 ± 2.2 <sup>C</sup>
Control II	3.3 <sup>hi</sup>	8.6 <sup>f</sup>	31.0 <sup>c</sup>	91.0 <sup>a</sup>	33.48 ± 10.5 <sup>A</sup>
T <sub>1</sub>	3.0 <sup>hijk</sup>	3.5 <sup>hi</sup>	3.6 <sup>h</sup>	21.0 <sup>d</sup>	7.78 ± 2.3 <sup>C</sup>
T <sub>2</sub>	3.1 <sup>hij</sup>	0.8 <sup>m</sup>	3.3 <sup>hi</sup>	8.5 <sup>f</sup>	3.93 ± 0.85 <sup>E</sup>
T <sub>3</sub>	2.5 <sup>hijkl</sup>	1.9 <sup>ijklm</sup>	2.3 <sup>hijkl</sup>	12.0 <sup>c</sup>	4.68 ± 1.3 <sup>D</sup>
T <sub>4</sub>	2.9 <sup>hijk</sup>	1.1 <sup>lm</sup>	2.5 <sup>hijkl</sup>	5.8 <sup>g</sup>	3.08 ± 0.52 <sup>F</sup>
T <sub>5</sub>	2.6 <sup>hijkl</sup>	2.4 <sup>hijkl</sup>	3.8 <sup>h</sup>	58.0 <sup>b</sup>	16.7 ± 7.2 <sup>B</sup>
T <sub>6</sub>	2.4 <sup>hijkl</sup>	1.7 <sup>ijklm</sup>	3.2 <sup>hij</sup>	7.9 <sup>f</sup>	3.8 ± 0.73 <sup>E</sup>
Pickling Period effect	2.66 ± 0.11 <sup>C</sup>	2.94 ± 0.49 <sup>C</sup>	7.03 ± 1.91 <sup>B</sup>	28.03 ± 5.95 <sup>A</sup>	

\*\* Expressed as ml NaOH 0.1 N/100gm cheese sample

\* cfu = colony forming unit

A, B, C, D and E: Means within the same effect having different capital superscripts are significantly different (P&lt;0.05).

a, b, ... and o: Means within the same interaction having different small superscripts are significantly different (P&lt;0.05).

Control I and II = white soft cheese made from buffalos' and goats' milk, respectively.

T<sub>1</sub> & T<sub>2</sub> = goats' white soft cheese contained 75 and 90 ppm cardamom essential oil, respectively.T<sub>3</sub> & T<sub>4</sub> = goats' white soft cheese contained 50 and 75 ppm thyme essential oil, respectively.T<sub>5</sub> & T<sub>6</sub> = goats' white soft cheese contained 40 and 60 ppm clove essential oil, respectively.



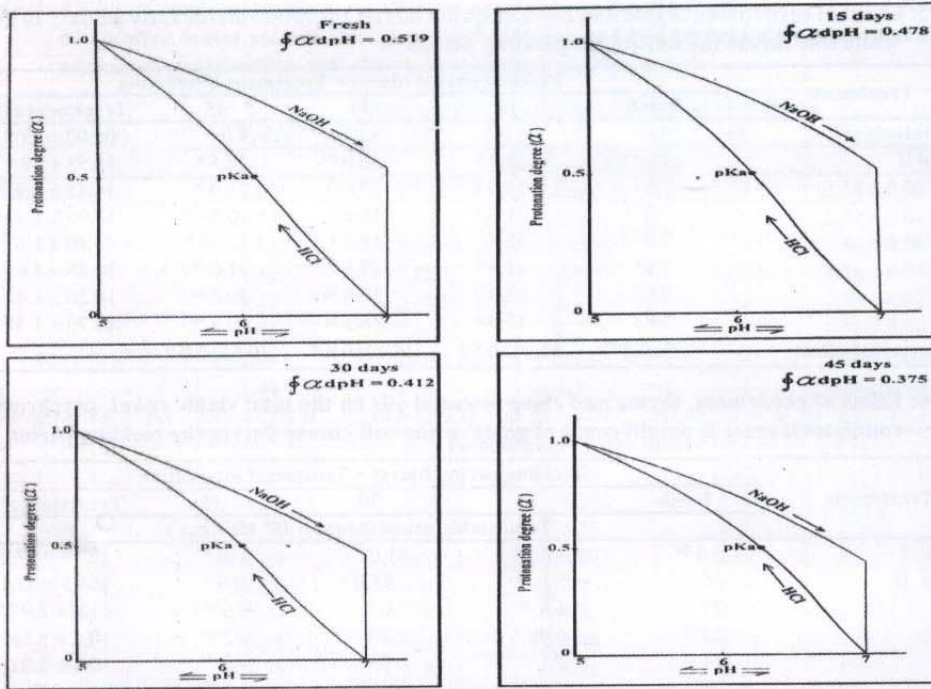


Fig. 1: Changes in the hysteresis size of buffalos' white soft cheese during the pickling period

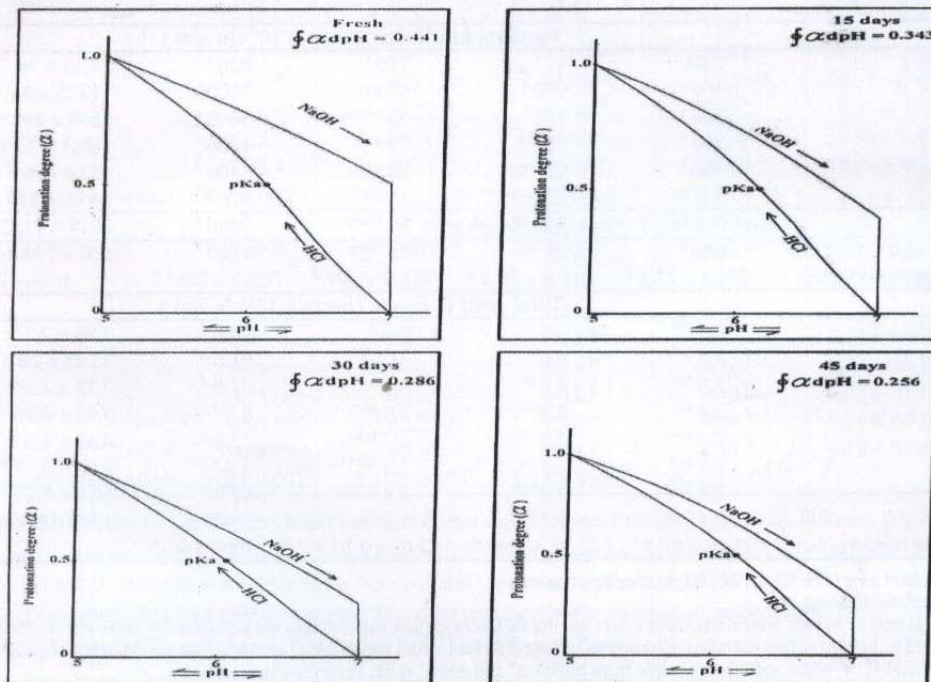


Fig. 2: Changes in the hysteresis size of goats' white soft cheese during the pickling period



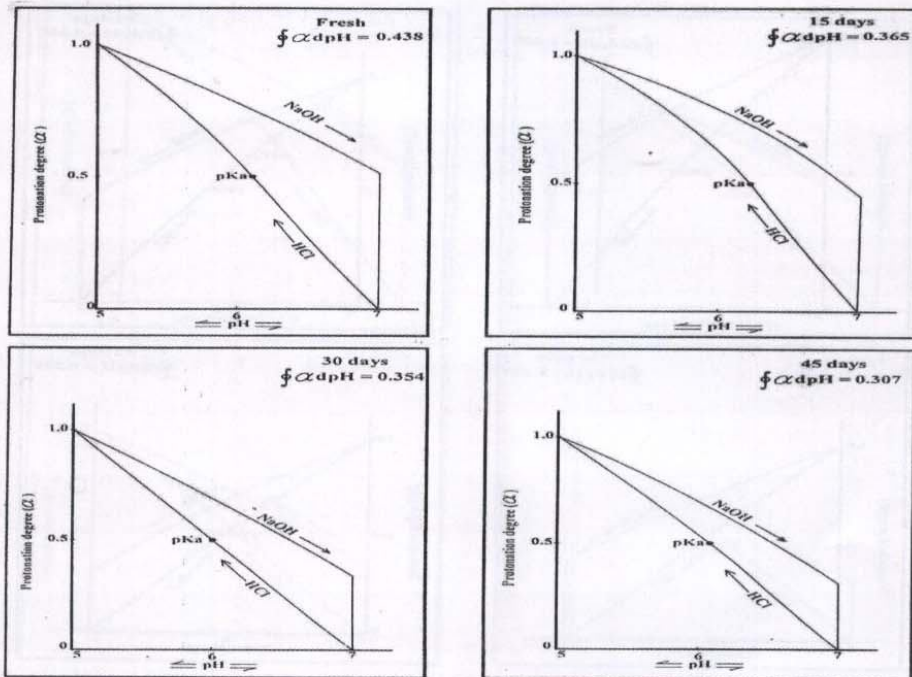


Fig. 3: Changes in the hysteresis size of goats' white soft cheese treated with 75 ppm cardamom during the pickling period

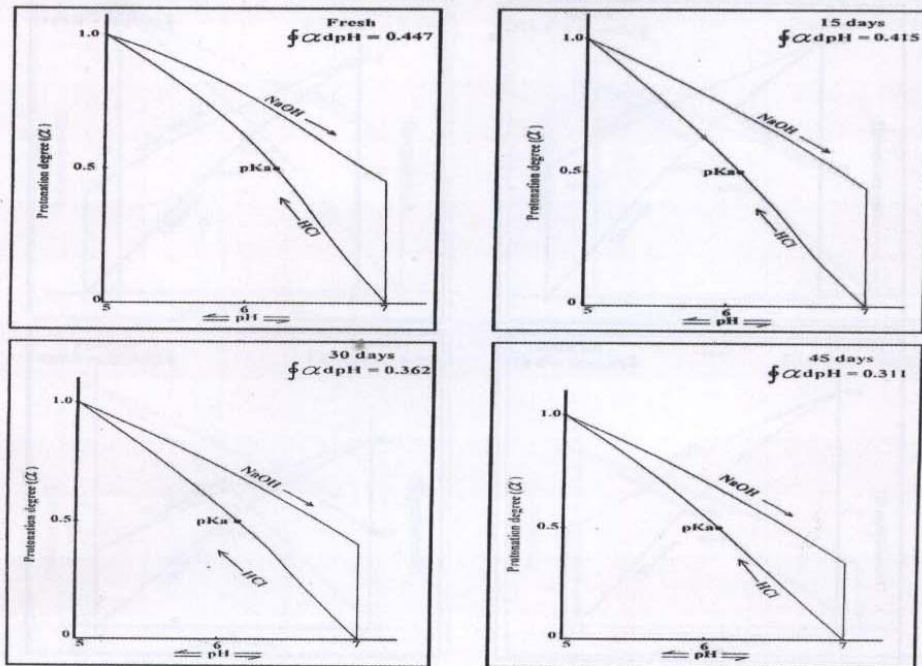


Fig. 4: Changes in the hysteresis size of goat's white soft cheese treated with 90 ppm cardamom during the pickling period



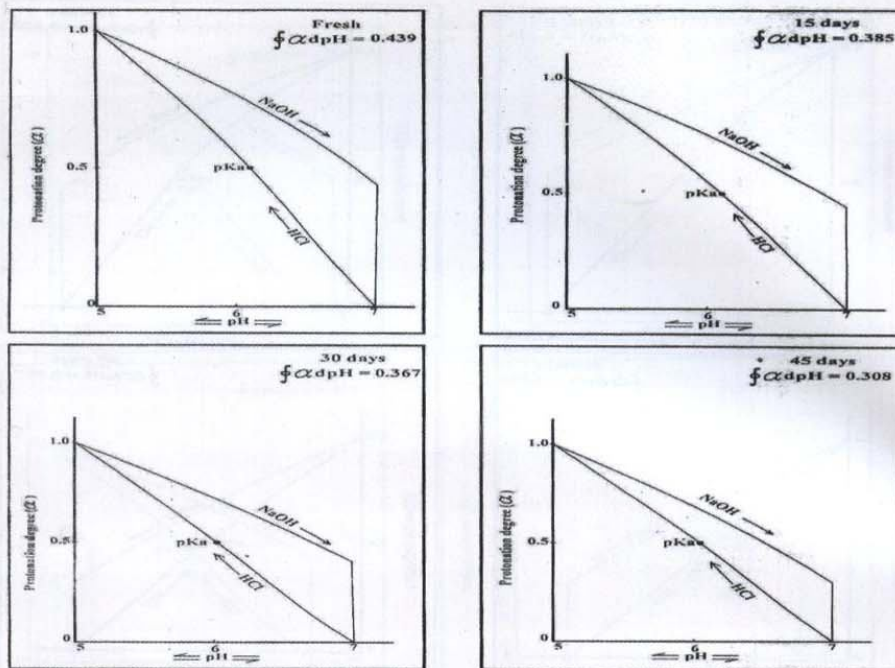


Fig. 5: Changes in the hysteresis size of goats' white soft cheese treated with 50 ppm thyme during the pickling period

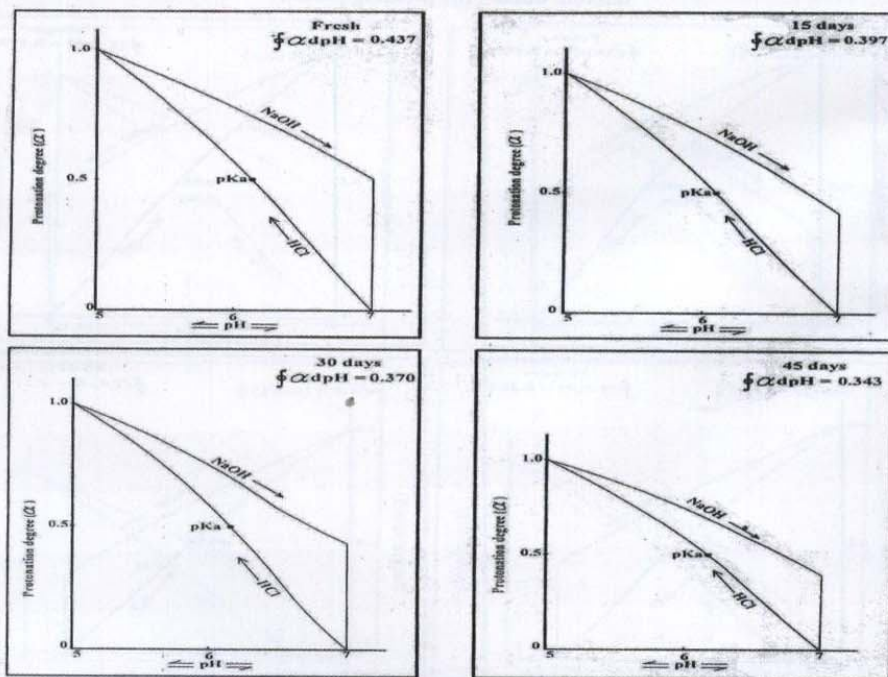


Fig. 6: Changes in the hysteresis size of goats' white soft cheese treated with 75 ppm thyme during the pickling period



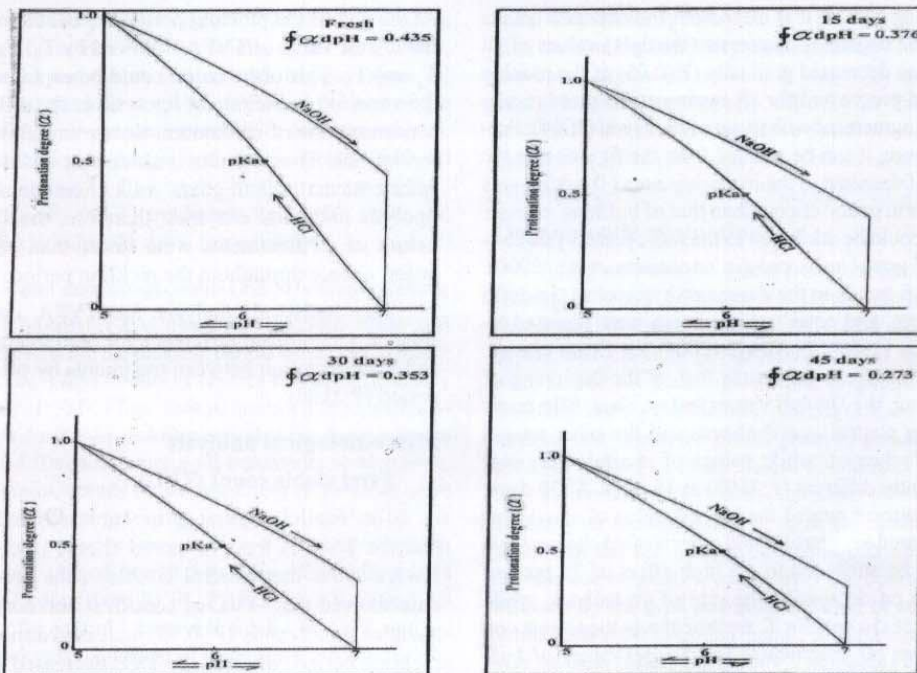


Fig. 7: Changes in the hysteresis size of goats' white soft cheese treated with 40 ppm clove during the pickling period

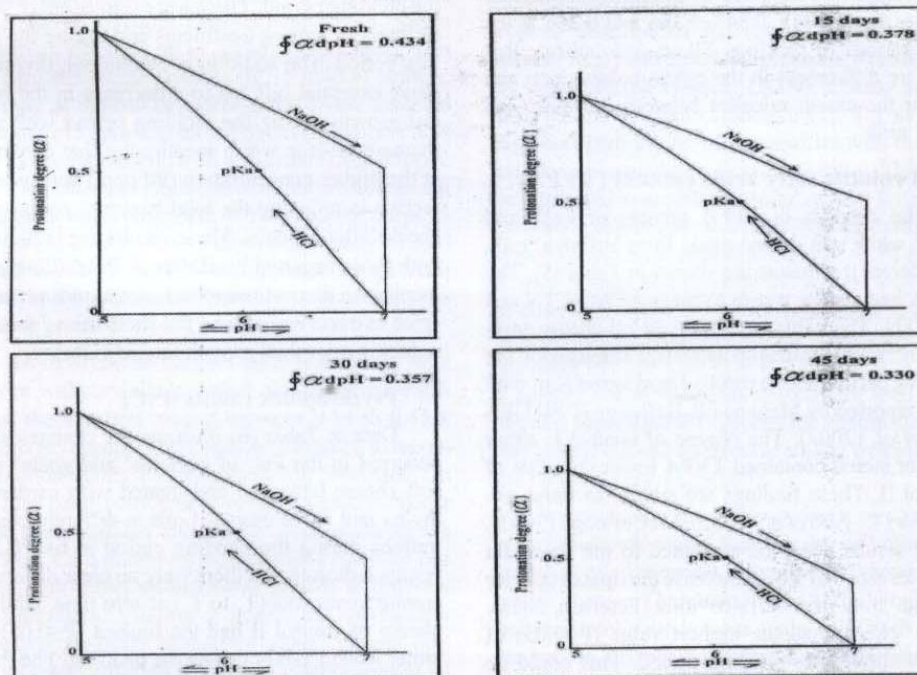


Fig. 8: Changes in the hysteresis size of goats' white soft cheese treated with 60 ppm clove during the pickling period



pickling period. It is clear from the obtained results that the degree of hysteresis ( $\oint \alpha \text{d pH}$ ) values of all cheeses decreased gradually ( $P \leq 0.05$ ) as the pickling period progressed for all treatments, so these results are in agreement with those of El-Tawel (2004). Furthermore, it can be noticed from the figures that the rate of decrease in the hysteresis area ( $\oint \alpha \text{d pH}$ ) was higher in goats' cheese than that of buffaloes' cheese. This could be attributed to the more proteolysis ability of goats' milk protein (Antunace *et al.*, 2000). Similar trends in the degree of hysteresis ( $\oint \alpha \text{d pH}$ ) of goats' and cows' white cheese were reported by Shendy (1989), El-Tawel (2004) for Edam cheese. It is clear from the results that, at the beginning of pickling, the  $\oint \alpha \text{d pH}$  values had no clear differences among control goats' cheese and the other treated goats' cheeses, while values of  $\oint \alpha \text{d pH}$  were significantly different ( $P \leq 0.05$ ) at 15 days. At 30 days,  $T_4$  treatment gained the highest value of  $\oint \alpha \text{d pH}$  as compared to control II and other treated cheeses; this could be attributed to the high effect of 75 ppm of thyme on decreasing the rate of proteolysis, while value of  $\oint \alpha \text{d pH}$  for  $T_5$  treatment was the lowest one between all treatments. The  $\oint \alpha \text{d pH}$  values of buffaloes' cheese samples were 0.519, 0.478, 0.412 and 0.375 for fresh, 15, 30 and 45 days of pickling, respectively, while the  $\oint \alpha \text{d pH}$  values of goats' cheese samples were 0.441, 0.343, 0.286 and 0.256 at the same periods of pickling. These data indicate that there are differences in the casein composition and size of the casein micelles between buffaloes' and goats' milk.

#### Total volatile fatty acids content (TVFA)

The changes in TVFA content of fresh and stored white soft cheese made from buffaloes' milk or different treatments are shown in Table (5). The TVFA had similar trends to those of WSN/TN and NPN/TN. The values of TVFA in all cheese samples increased gradually ( $P \leq 0.05$ ) throughout the pickling period. These results are in agreement with those reported by Mehanna & Hefnawy (1991), El-Abd *et al.* (2003). The cheese of control I, either fresh or stored contained TVFA lower than that of control II. These findings are similar to those obtained by El-Abd *et al.* (1992), McCullough (2003). These results could be attributed to the lower fat globules size that would increase the lipases activity in goats' milk than buffaloes' milk (Peterson, 2005). Goats' cheese had the highest value ( $P \leq 0.05$ ) of TVFA all over the pickling period. This could be attributed to the activity of certain lipolytic enzymes and activity of psychrophilic (PsC) bacteria.

At the end of the pickling period,  $T_6$  treatment had the lowest value of TVFA followed by  $T_5$ ,  $T_2$ ,  $T_1$ ,  $T_4$  and  $T_3$ . This observation could be explained by the variation in the rate of lipolysis at the different concentrations of cardamom, thyme and clove essential oils. These results indicate that addition of spice essential oils to goats' milk cheese inhibited lipolytic microbial enzymes; therefore, the TVFA values of all treatments were lower than control goats' cheese throughout the pickling period.

The statistical analysis for TVFA content showed a significant difference for both treatments and the interaction between treatments by pickling period ( $P \leq 0.05$ ).

#### Microbiological analysis

##### Total viable count (TVC)

The results illustrated in Table (6) indicate that the TVC of fresh or stored cheese of control I were lower than control II. Also, the obtained data showed that TVC of control II and control I samples significantly ( $P \leq 0.05$ ) increased during the pickling period, whereas the TVC decreased in the other cheese treatments ( $T_1$  to  $T_6$ ), up to 30 days of pickling, then the trend was vice versa at 45 days of the pickling period. There were significant ( $P \leq 0.05$ ) differences among treatments and during the pickling period. The addition of cardamom, thyme and clove essential oils led to a decrease in the bacterial growth during the pickling period with some fluctuation. It is worth mentioning that cardamom at the higher concentration (90 ppm) was more effective in reducing the total bacterial count of the cheese after 15 days. These results are in harmony with those reported by Holley & Patel (2005), who mentioned that antimicrobial compounds present in spice extracts can extend the shelf-life of foods by reducing microbial growth rate or viability.

##### Psychrophilic counts (PsC)

Data in Table (6) illustrate the changes which occurred in the PsC of buffaloes' and goats' white soft cheese untreated and treated with cardamom, thyme and clove essential oils at different concentrations during the pickling period at  $6 \pm 1^\circ\text{C}$ . The results indicated that there were no clear differences among treatments ( $T_1$  to  $T_6$ ) at zero time, while the cheese of control II had the highest ( $P \leq 0.05$ ) PsC either when fresh or during the pickling. The PsC in all treated cheese samples significantly ( $P \leq 0.05$ ) decreased as the pickling period extended up to 30 days



after that increased at the end of the pickling process which coincides with the results of the TVC. Also, control I and cheese treated with 75 ppm thyme oil ( $T_4$ ) had the lowest counts of PsC at the end of the pickling period as compared with other treatments. Meanwhile, control II and cheese treated with 40 ppm clove oil ( $T_5$ ) had the highest counts of PsC. The  $T_3$ ,  $T_5$  and  $T_6$  treatments had 3400, 9600 and  $1700 \times 10^4$  after 45 days, respectively.

#### Yeast and mould counts (Y&M) coliform counts

The total Y&M counts of different white soft cheeses during the pickling period are shown in Table (6). The Y&M counts of control II was significantly higher ( $P \leq 0.05$ ) than that in control I or other treatments during the pickling period. Also, there were no clear differences among all treatments at zero time. Generally, counts in the beginning of pickling were low and then gradually increased ( $P \leq 0.05$ ) as the pickling period progressed. The Y&M counts in fresh cheeses ranged from 1.5 to  $3.3 \times 10^2$ , while counts after 45 days were 20, 91, 21, 8.5, 12, 5.8, 58 and  $7.9 \times 10^2$  for control I, control II,  $T_1$ ,  $T_2$ ,  $T_3$ ,  $T_4$ ,  $T_5$  and  $T_6$ , respectively. Addition of some spice essential oils to goats' milk led to a reduction in the number of Y&M in the resultant goats' cheese as compared with control II. These results are accordance with the findings of Serrano *et al.* (2005). Cheeses treated with 75 ppm cardamom essential oil ( $T_1$ ) had higher counts than other treated cheese samples till 15 days of storage, whereas, at the end of pickling, the thyme essential oil at ratio 75 ppm was more effective towards Y&M than cardamom essential oil at the ratio 75 or 90 ppm. There was no clear inhibition of treatment containing 40 ppm clove oil on growth of Y&M during the pickling period except after 15 days. Statistically, Y&M counts showed a significant difference for all treatments, and the interaction between treatments by the pickling period ( $P \leq 0.05$ ).

All fresh cheese samples made from goats' milk with or without spice essential oils contained coliforms, the numbers ranged between 0.14 to  $0.45 \times 10^2$  cfu/gm cheese. This could be attributed to recontamination during the manufacturing processes. On the other hand, coliforms were not detected in cheese made from buffalos' milk either fresh or during the pickling. Cardamom, thyme and clove essential oils had a high effect on coliforms growth at different concentrations. Counts of coliforms significantly increased ( $P \leq 0.05$ ) in control goats' cheese during pickling, whereas coliforms disappeared in all cheese treatments beginning of 15 days up to the end of the pickling period. These results are in accord-

ance with those of Farag *et al.* (1989) who found that, minimum inhibitory concentration (MIC) of clove and thyme oil on *E. coli* was 1.25 and 0.75 mg/ml medium, respectively. Also Smith-Palmer *et al.* (2001), concluded that, clove and thyme oil had the highest effect on *E. coli* with a bacteriostatic concentrations of 0.04 and 0.05 mg, respectively.

#### Organoleptic evaluation

White soft cheese made from buffalos' milk (control I), goats' milk (control II) and goats' milk containing different essential oils of cardamom, thyme and clove at 2 concentrations of each oil were scored for their organoleptic properties when fresh and during the pickling periods (15, 30 and 45 days) at  $6 \pm 1^\circ\text{C}$  and the points given by panelists are given in Table (7). Fresh cheese of control I gained the maximum total scores as compared with control II and other treatments. In general, the results indicated that the use of spice essential oils improved the flavour of goats' cheese, also the scores were affected by the type and concentration of spice essential oils. On the other hand, at zero time there was no clear difference among treatments ( $T_1$  to  $T_6$ ) in colour & appearance and body & texture, while addition of clove at 40 ppm ( $T_5$ ), thyme at 50 ppm ( $T_3$ ) and cardamom at 75 ppm ( $T_1$ ) improved flavour of these cheeses than control II and other treatments ( $T_2$ ,  $T_4$  and  $T_6$ ). Similar results were reported by Ayar (2002), Hussein (2004). Cheese samples with 75 and 90 ppm cardamom ( $T_1$ ,  $T_2$ ) showed a high acceptability by most panelists with the highest flavour scores at 15 days of pickling. This could be attributed to accumulation of volatile fatty acids, carbonyl compounds and other flavour compounds which are essential to cheese flavour. At 30 days  $T_1$ ,  $T_3$  and  $T_5$  treatments gained the highest total scores as compared with other treatments, while at the end of the pickling total score of all treatments was in the order of:  $T_1 > T_3 = T_5 > T_4 > \text{control II} = T_2 > T_6 = \text{control I}$ . The statistical analysis for total scores showed significant differences for treatments, pickling period and the interaction among treatments by the pickling period ( $P \leq 0.05$ ).

In conclusion, from the previous results it can be conclusively stated that the use of some spice essential oils improved the quality of the resultant goats' cheese. Therefore, white soft cheese could be successfully made from goats' milk with adding cardamom, thyme or clove essential oils, especially clove or cardamom at concentration of 40 and 75 ppm, respectively and stored at  $6 \pm 1^\circ\text{C}$  for 45 days.



**Table 7: Organoleptic properties of goats' white soft cheese as affected by adding some essential oils during the pickling period**

Treatments	Parameters	Pickling period (days)				Treatment effect
		Fresh	15	30	45	
Control I	Flavour (50)	45	46	45	35	86.5 ± 1.68 <sup>A</sup>
	Body & texture (35)	30	32	33	34	
	Colour & appearance (15)	14	13	11	8	
	Total scores (100)	89 <sup>bc</sup>	91 <sup>a</sup>	89 <sup>bc</sup>	77 <sup>o</sup>	
Control II	Flavour (50)	40	43	36	38	81.5 ± 0.83 <sup>D</sup>
	Body & texture (35)	31.5	29	31	28.5	
	Colour & appearance (15)	11.5	13	11	11.5	
	Total scores (100)	83 <sup>hi</sup>	85 <sup>fg</sup>	80 <sup>lm</sup>	78 <sup>no</sup>	
T <sub>1</sub>	Flavour (50)	42	44	40	39	84.13 ± 0.73 <sup>B</sup>
	Body & texture (35)	31	31	32	30	
	Colour & appearance (15)	12	12	12	11.5	
	Total scores (100)	85 <sup>fg</sup>	87 <sup>de</sup>	84 <sup>gh</sup>	80.5 <sup>kl</sup>	
T <sub>2</sub>	Flavour (50)	40	44	36	38	81.75 ± 0.93 <sup>D</sup>
	Body & texture (35)	31.5	29	31	28.5	
	Colour & appearance (15)	11.5	13	15	11.5	
	Total scores (100)	83 <sup>hi</sup>	86 <sup>ef</sup>	80 <sup>lm</sup>	78 <sup>no</sup>	
T <sub>3</sub>	Flavour (50)	42.5	40	38	37	83.13 ± 0.71 <sup>C</sup>
	Body & texture (35)	31.5	32.5	32	32	
	Colour & appearance (15)	12	12	12	11	
	Total scores (100)	86 <sup>ef</sup>	84.5 <sup>g</sup>	82 <sup>ij</sup>	80 <sup>lm</sup>	
T <sub>4</sub>	Flavour (50)	36.5	38.5	35	34	80.63 ± 0.61 <sup>E</sup>
	Body & texture (35)	31.5	32.5	32	33.5	
	Colour & appearance (15)	11.5	13	13	11.5	
	Total scores (100)	79.5 <sup>lm</sup>	84 <sup>gh</sup>	80 <sup>lm</sup>	79 <sup>mn</sup>	
T <sub>5</sub>	Flavour (50)	42.5	41	39	38.5	84.5 ± 0.91 <sup>B</sup>
	Body & texture (35)	31.5	34	32	30.5	
	Colour & appearance (15)	12	13	13	11	
	Total scores (100)	86 <sup>ef</sup>	88 <sup>cd</sup>	84 <sup>gh</sup>	80 <sup>lm</sup>	
T <sub>6</sub>	Flavour (50)	38.5	39.5	33	34.5	80.4 ± 0.83 <sup>E</sup>
	Body & texture (35)	31	32	33	32.5	
	Colour & appearance (15)	12	12.5	13	10	
	Total scores (100)	81.5 <sup>jk</sup>	84 <sup>gh</sup>	79 <sup>mn</sup>	77 <sup>o</sup>	
Pickling period effect		84.13±0.59 <sup>B</sup>	86.19±0.49 <sup>A</sup>	82.25±0.66 <sup>C</sup>	78.69±0.29 <sup>D</sup>	

A, B, C, D and E: Means within the same effect having different capital superscripts are significantly different ( $P \leq 0.05$ ).

a, b, ... and o: Means within the same interaction having different small superscripts are significantly different ( $P \leq 0.05$ ).

Control I and II = white soft cheese made from buffalos' and goats' milk, respectively.

T<sub>1</sub> & T<sub>2</sub> = goats' white soft cheese contained 75 and 90 ppm cardamom essential oil, respectively.

T<sub>3</sub> & T<sub>4</sub> = goats' white soft cheese contained 50 and 75 ppm thyme essential oil, respectively.

T<sub>5</sub> & T<sub>6</sub> = goats' white soft cheese contained 40 and 60 ppm clove essential oil, respectively.



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