



## EFFECT OF CARBON DIOXIDE TREATMENT ON THE SHELF-LIFE OF KAREISH CHEESE

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### ABSTRACT

To determine the influence of milk pre-acidification with CO<sub>2</sub> on Kareish cheese shelf-life, cheese was manufactured from milk with and without added CO<sub>2</sub>. This is accomplished by direct injection of CO<sub>2</sub> into the milk prior to coagulation. Four different carbonation levels ranging from approximately 10 to 35 mg/100g were achieved by adjusting the flow rate of CO<sub>2</sub> at 3, 5, 7, and 9 L/min for 12, 20, 28, and 36 min respectively while keeping the weight of milk constant (4 kilograms per treatment). This treatment decreased the pH of milk from 6.6 to levels between 5.8 and 6.4 at 40°C in the cheese vat. Kareish cheese was manufactured from CO<sub>2</sub>-treated and untreated milk by the conventional method. Cheese samples were stored at 4°C for 28 d. Shelf-life of the cheese was measured by microbial counts and by taste panel scoring. Microbial analysis revealed that, CO<sub>2</sub> effectively delayed microbial growth in CO<sub>2</sub>-treated cheese. The carbonated cheeses had significantly lower total bacterial count compared with that of control. Psychrotrophic, and molds and yeast counts were significantly lower in carbonated cheese samples. Coliforms were not detected in all cheese samples. The use of CO<sub>2</sub> significantly improved the organoleptic quality of cheese as compared to the corresponding control cheese up to 28 d. The CO<sub>2</sub> provided the same quality after 28 d that accomplished at 21 d. High

quality cheese with 35 mg/100g added CO<sub>2</sub> could successfully keep at 4°C for 28 d with SPC < 3 x 10<sup>5</sup> cfu/g. The control cheese exhibited spoilage after 21 d at 4°C, while the carbonated cheeses remained fresh for as long as 28 d.

### INTRODUCTION

The shelf-life of refrigerated non-sterile dairy products, including cheese is generally limited to 1-3 wk (Muir, 1996 and Salvador & Fiszman, 2004), depending upon the quality of the raw materials, processing conditions, and post processing handling.

Kareish cheese is a mild acid-coagulated white soft cheese made from skim milk with a slight salt. It is the most popular type of cheese in Egypt and Arabian countries. The popularity of this cheese is mainly due to its fresh taste and good digestibility, but unfortunately, shelf-life does not paint such a picture. The shelf-life of Kareish cheese, produced by major dairy processors, is generally limited between 14 and 21 d at refrigerator temperatures. For this reason, there is an interest in extending shelf-life of this type of cheese to meet market demands for longer shelf-life products.

Milk and dairy products are excellent growth media for pathogenic and spoilage microorganisms (Muir 1996). The composition of most dairy products provides a favorable environment for the growth and propagation of a broad spectrum of microorganisms. Microbiological deterioration of refrigerated non-sterile dairy products such as Cottage cheese, and similar products is often caused

by the growth of psychrotrophic gram-negative bacteria, yeasts and molds, causing flavor, textural, and visual spoilage (Ternstrom *et al* 1993).

Development of ways to prevent or slow microbiological growth in food therefore can extend shelf life, improve marketability and overall quality, and make a safer product for the consumer. Carbon dioxide has been used extensively in extending the shelf life of a wide variety of cold-stored dairy products, including cheese and, more particularly cottage cheese (Eliot *et al* 1998; Gonzalez-Fandos *et al* 2000; Fornasari *et al* 2004; Nelson *et al* 2004; Hotchkiss *et al* 2006). The physicochemical changes that occur as milk pH changes due to CO<sub>2</sub> content may be as important to cheese manufacture as the antimicrobial effect (Ruas-Madiedo *et al* 2003).

Therefore, the object of the present study was to determine the potential addition of CO<sub>2</sub> as a shelf-life extender in Karish cheese and to provide cheese having an increased margin of safety for the consumer without sacrificing desired properties.

## MATERIALS AND METHODS

### Milk Treatments

Raw whole cow's milk was received from the Mehalet Moussa Experimental Station, Kafr El-Sheikh Governorate, Egypt. The milk was defatted, pasteurized at 72°C for 15 s and cooled to 4°C. Twenty kilograms of pasteurized skim milk (pH 6.6) was then divided into five batches (four kilograms for each). The first batch was served as control whereas the other four batches were separately carbonated in a tightly closed hermetic pan. Food-grade CO<sub>2</sub> gas (Industrial Gas Co., Cairo, Egypt) was injected in the bottom of the pan to minimize the loss of CO<sub>2</sub> and to insure more homogeneous distribution of CO<sub>2</sub> in the product. The procedure was performed through a flow meter at the rates of 3, 5, 7, and 9 l./min for 12, 20, 28, and 36 min for the first, second, third, and fourth batch respectively to achieve carbonation levels of approximately 10, 20, 25, and 35 mg/100g, which caused a decrease in pH-values of milk to 6.40, 5.95, 5.88 and 5.83 respectively.

### Cheese Manufacture

Four kilograms of each batch were weighed and used for cheese manufacture by the conventional method. The milk was inoculated with about 1.5% of the lactic acid starter culture (i.e., *Streptococcus thermophilus* and *Lactobacillus delbrueckii*

*ssp. bulgaricus*) and incubated at 40°C until complete coagulation. After coagulation, curds were transferred to cheese cloth to drain excess whey and salt was added at 3% of the curd weight. Milk treatments and cheese manufacture were completed on the same day. The cheese curd was packaged in sterile, sealable containers to provide the highest margin of safety and stored at 4°C for 28 d. Samples were taken, at 0-time, 7, 14, 21 and 28 d of storage for microbiological tests and sensory evaluation. Cheese manufacture trials were repeated in duplicate and each analysis in quadruplicate. The values reported are the means of the two cheese manufacture trials.

### Determination of pH

Milk and cheese samples were examined for pH-value using a pH-meter Jenway 3020 (Jenway Ltd. Gransmore Green, Felsted, Dunmow, England).

### Determination of CO<sub>2</sub> concentrations

Concentrations of CO<sub>2</sub> in milk and cheese samples were estimated according to the method of AOAC (1990). CO<sub>2</sub> was released from the sample by the addition of sulphuric acid (1N) accompanied with gentle heating and absorbed in barium hydroxide (0.1N) to form precipitate of barium carbonate. The excess alkali was back titrated with oxalic acid (0.1N) solution. A blank was carried out. The concentration of CO<sub>2</sub> in an individual sample was calculated by extrapolation of the following equation: CO<sub>2</sub> (mg/100 g) = (b-a) x 10000/ weight of sample, where: a = ml of oxalic acid require for test and b = ml of oxalic acid required for blank.

### Microbiological examination

Microbiological analysis included determination of standard plate count (SPC), psychrotrophic bacteria count (PBC), coliform count (CC), and mold and yeast counts (MYC) according to Houghtby *et al* (1992). Actual counts of the microorganisms were reported as log 10 cfu/g.

### Sensory evaluation

Cheese samples were sensory scored using score card for flavor (60 points), body and texture (30 points), appearance (5 points) and saltiness (5 points) as described by Nelson and Trout (1981). The scores were averaged by five panelists.

### Statistical analysis

A one way ANOVA was used for statistical analysis. All tests were carried out in quadruplicate. Duncan test was used to compare treatment means at probability ( $p \leq 0.05$ ). The PROC GLM procedure of SAS was used for all data analysis.

## RESULTS AND DISCUSSION

### Carbon dioxide content

The mean value of CO<sub>2</sub> content in untreated milk was about 3.78 mg/100 g (Table 1). This value is too lower than that previously reported by many investigators. Carbon dioxide occurs naturally in milk, but most of it is lost in the course of processing. A proportional increase was found in carbonated milks by increasing the CO<sub>2</sub> addition rates.

A portion of the CO<sub>2</sub> present in milk pre-acidified with CO<sub>2</sub> remained in the cheese. The CO<sub>2</sub> levels of carbonated cheeses were significantly higher ( $p \leq 0.05$ ) than that of control (Table 2). The highest concentration was found in the cheese treated with the highest level of CO<sub>2</sub>, which was approximately 9 times higher than that measured in control (34.45 vs. 3.78 mg/100g). The CO<sub>2</sub> contents of the carbonated cheeses were decreased throughout the storage period due to the gradual diffusion of the CO<sub>2</sub>. At any time of storage, the CO<sub>2</sub> content for each treatment was lower than the initial value. CO<sub>2</sub> is a byproduct of bacterial respiration and responsible for the increase of CO<sub>2</sub> content in the control sample during storage (data not presented). Carbonated cheeses with low increases in bacterial growth did not show an increase in CO<sub>2</sub> concentration over the storage period. The CO<sub>2</sub> content of the carbonated cheeses decreased rapidly during the first week of storage, remained practically constant during the second week then decreased slowly until the end of the storage period (28 d). The higher CO<sub>2</sub> content of carbonated cheeses was maintained throughout 28 d of storage.

### Changes in pH

Levels of CO<sub>2</sub> added to the used milk decreased the pH of the milk by about 0.2 to 0.8 units, depending upon the level of CO<sub>2</sub> added. The higher the level of CO<sub>2</sub>, the lower was the pH value. Initial pH of milk (6.6) was decreased to 6.4, 5.95, 5.88 and 5.83 with the injection rates of

3, 5, 7, and 9 L/min respectively. The decrease in pH values may be due to CO<sub>2</sub> bound to protein and formation of carbonic acid and release of H<sup>+</sup> as CO<sub>2</sub> dissolves in the water of the milk (Ma *et al* 2001; Ma and Barbano, 2003). The extent of pH decrease is related to the amount of CO<sub>2</sub> dissolved, hydrated, and protonated in the aqueous phase and, thus, depends on the intrinsic properties of the aqueous phase, such as buffering capacity and initial pH (Devlieghere *et al* 1998). This decrease in pH values was then resulted in shorter coagulation time during cheese manufacture. Several studies have showed that addition of CO<sub>2</sub> to milk used to cheese manufacture reduced the processing time (Montilla *et al* 1995; Gevaudan *et al* 1996; Ruas-Madiedo *et al* 2002; Gueimonde and de los Reyes-Gavilan, 2004).

At 0-time of storage, the mean pH of the control cheese was significantly higher ( $P \leq 0.01$ ) than those of the carbonated cheese samples (Table 3). This trend was observed till reach the minimum pH at the end of storage period. The pH decreased in both the control and carbonated cheese samples over the storage period. The decrease of pH in the control cheese was probably caused by the high level of microbial growth rate during the storage period.

Decreases in the pH values of carbonated cheese samples were potentially attributed to either lactic acid produced by starter culture or to CO<sub>2</sub> dissolved into the sample during milk treatment. After injection, carbonated milks had pH values of 6.4, 5.95, 5.88, and 5.83 while after inoculation and a 3 hr incubation, sufficient lactic acid was produced to decrease the pH of cheeses to 4.44, 4.31, 4.21, and 4.1, respectively. The drop in pH was due to the production of lactic acid by the starter culture and not due to the incorporation of CO<sub>2</sub> into the sample during milk treatment. Several studies have showed that milk treated with CO<sub>2</sub> and inoculated with cheese starter cultures produced curds contained sufficient numbers of viable starter culture to continue lowering the curd pH suggesting that CO<sub>2</sub> dissolved in milk does not affect the growth and metabolism of cheese starter cultures (Ruas-Madiedo *et al* 1998 and van Hekken *et al* 2000).

### Changes in microbial counts

Changes in microbial counts of the control and carbonated cheeses during storage at 4°C for 28 d are shown in Table (4).

Table 1. Carbon dioxide content\*\* (mg/100g) of CO<sub>2</sub>-treated and untreated milk

	Treatments				
	Control*	3L/min	5L/min	7L/min	9L/min
Milk	3.775 <sup>e</sup>	10.950 <sup>d</sup>	20.175 <sup>c</sup>	25.375 <sup>b</sup>	34.450 <sup>a</sup>

\*Control: Skim milk without adding CO<sub>2</sub>

\*\*Mean of four replicates

Table 2. Carbon dioxide content\*\* (mg/100g) of the control and carbonated cheeses during 28 d of storage at 4°C

Storage period (d)	Treatments				
	Control*	3L/min	5L/min	7L/min	9L/min
0	3.78	10.95	20.18	25.38	34.45
7	-	7.90	11.58	11.98	15.33
14	-	6.05	9.65	10.00	14.05
21	-	4.68	6.33	8.03	12.65
28	-	3.95	5.20	6.55	11.25

\*Control: Cheese sample made without adding CO<sub>2</sub>

\*\*Mean of four replicates

Table 3. Changes in pH values\*\* of the control and carbonated cheeses during 28 d of storage at 4°C

Storage period (d)	Treatments				
	Control*	3L/min	5L/min	7L/min	9L/min
0	4.60 <sup>a</sup>	4.44 <sup>b</sup>	4.31 <sup>c</sup>	4.21 <sup>d</sup>	4.10 <sup>e</sup>
7	4.46 <sup>a</sup>	4.29 <sup>b</sup>	4.16 <sup>c</sup>	4.04 <sup>d</sup>	3.94 <sup>d</sup>
14	4.29 <sup>a</sup>	4.14 <sup>b</sup>	4.00 <sup>c</sup>	3.91 <sup>c</sup>	3.76 <sup>d</sup>
21	4.16 <sup>a</sup>	4.00 <sup>b</sup>	3.85 <sup>c</sup>	3.76 <sup>c</sup>	3.63 <sup>d</sup>
28	4.03 <sup>a</sup>	3.84 <sup>b</sup>	3.66 <sup>c</sup>	3.55 <sup>c</sup>	3.39 <sup>d</sup>

\*Control: Cheese sample without adding CO<sub>2</sub>

\*\*Mean of four replicates

- Means with the same letter in the same row are not significantly different.

### Standard plate count (SPC)

The initial SPC was significantly lower ( $p \leq 0.05$ ) in carbonated cheeses than those of the control (Table 4). The initial SPC was almost similar ( $p \geq 0.05$ ) in different carbonated cheeses, while final SPC decreased as CO<sub>2</sub> level increased. The SPC in the control cheese increased rapidly to reach their double initial count between 14 and 21 d of storage and further increase was continued. The SPC in the carbonated cheeses also increased but at a substantially lower rate, thus extending shelf-life. The rate of increase in the carbonated cheeses was reversely proportional with the level of CO<sub>2</sub> added. This was probably due to the inhibitory effect of CO<sub>2</sub> which drastically reduced the growth rate of microorganisms than that of control. The greater the amount of CO<sub>2</sub> dissolved in milk the greater was the magnitude inhibition. It was reported that the antimicrobial effect of CO<sub>2</sub> was related to the amount of CO<sub>2</sub> dissolved in the aqueous phase of food and was independent of the pH reduction effect of CO<sub>2</sub> (Devlieghere *et al* 1998). The results also showed that carbonated cheeses contained lower ( $p \leq 0.05$ ) SPC than the control at any time of storage. At the end of storage, carbonated cheese showed the lowest values of population. SPC at 28 d of storage ranged between  $0.6 \times 10^6$  and  $0.95 \times 10^6$  cfu/g with an average of  $0.74 \times 10^6$  compared with  $1.95 \times 10^6$  in control. Nearly similar findings were reported by Lee and Hotchkiss, (1997).

### Psychrotrophic bacteria

A similar trend was observed for psychrotrophic bacterial counts. Psychrotrophs grew in all cheese samples but were less numerous in carbonated cheese over entire storage period (Table 4). The inhibitory effect of CO<sub>2</sub> treatment on the growth and survival of psychrotrophic bacteria in various types of cheese has been extensively investigated (Moir *et al* 1993; Eliot *et al* 1998; Gonzalez-Fandose *et al* 2000). McCarney *et al* (1995) concluded that addition of 30 mM of CO<sub>2</sub> to milk used to cheese manufacture reduced the time to reach psychrotrophic counts of  $10^6$  cfu/mL<sup>-1</sup> and that in turn improved grading scores of cheese. Hotchkiss *et al* (1999) reported that, added CO<sub>2</sub> can effectively control growth of psychrotrophic bacteria in raw milk and final dairy products during refrigerated storage.

### Coliform bacteria

Over 28 d of storage at 4°C, coliform bacteria, did not detected in any of carbonated or control cheese samples (data not presented). Similar findings were reported by Ballestra *et al* (1996). A simultaneous increase in the lag phase and decrease in exponential growth rate due to CO<sub>2</sub> treatment has been demonstrated for coliform bacteria (Kimura *et al* 1999 and Martin *et al* 2003).

### Mold and yeast

Mold and yeast counts were significantly ( $P \leq 0.05$ ) lower in carbonated cheeses in comparison with the control cheese over storage period despite the higher acidity of the former cheeses which may enhance their growth (Table 4). CO<sub>2</sub> acts both directly on molds and indirectly by displacing O<sub>2</sub>; molds have an absolute requirement for O<sub>2</sub>. Chen and Hotchkiss (1991) reported that CO<sub>2</sub> had an inhibitory effect on the yeast and mould populations in Cottage cheese and the inhibitory effect was not attributed to the pH-reducing effect of CO<sub>2</sub> as the controls and treated samples had almost similar pH values.

Although, the inhibitory effect of CO<sub>2</sub> on certain microorganisms, have been extensively studied, the direct and indirect mechanisms by which CO<sub>2</sub> affects microbial growth and metabolism are not entirely clear. There is evidence that there are at least 3 general mechanisms. The 1st and simplest is by the displacement of O<sub>2</sub>. The 2nd mechanism is a reduction of the pH due to the dissolving of CO<sub>2</sub> in the aqueous phase and formation of carbonic acid, which dissociates to form the bicarbonate and H<sup>+</sup> ions as the following equilibrium:



Carbon dioxide and the H<sup>+</sup> ions are mainly responsible for the inhibitory effect; however, bicarbonate and carbonate ions have also been shown to have an inhibitory effect. The 3rd mechanism is a direct effect on the metabolism of microorganisms as opposed to the indirect effects of displacement of O<sub>2</sub> and pH reduction. The direct inhibition of metabolic pathways, including membrane damage, enzyme inactivation, decarboxylation reactions and DNA replication (Stretton & Goodman 1998; Hong & Pyun 2001).



Table 4. Changes in microbial counts\*\* of the control and carbonated cheeses during 28 d of storage at 4°C

Microbial counts	Storage period (d)	Treatments				
		Control*	3L/min	5L/min	7L/min	9L/min
SPC 10 <sup>6</sup> (cfu/g)	0	0.75 <sup>a</sup>	0.25 <sup>b</sup>	0.25 <sup>b</sup>	0.23 <sup>b</sup>	0.20 <sup>b</sup>
	7	0.85 <sup>a</sup>	0.48 <sup>b</sup>	0.43 <sup>b</sup>	0.30 <sup>b</sup>	0.30 <sup>b</sup>
	14	1.35 <sup>a</sup>	0.58 <sup>b</sup>	0.53 <sup>b,c</sup>	0.38 <sup>c,d</sup>	0.30 <sup>d</sup>
	21	1.65 <sup>a</sup>	0.73 <sup>b</sup>	0.60 <sup>b,c</sup>	0.55 <sup>b,c</sup>	0.43 <sup>c</sup>
	28	1.93 <sup>a</sup>	0.95 <sup>b</sup>	0.75 <sup>b</sup>	0.67 <sup>b</sup>	0.60 <sup>b</sup>
PBC 10 <sup>4</sup> (cfu/g)	0	0.58 <sup>a</sup>	0.20 <sup>b</sup>	0.20 <sup>b</sup>	0.18 <sup>b</sup>	0.20 <sup>b</sup>
	7	0.83 <sup>a</sup>	0.30 <sup>b</sup>	0.28 <sup>b</sup>	0.20 <sup>b</sup>	0.20 <sup>b</sup>
	14	1.10 <sup>a</sup>	0.38 <sup>b</sup>	0.33 <sup>b,c</sup>	0.28 <sup>b,c</sup>	0.23 <sup>c</sup>
	21	1.38 <sup>a</sup>	0.55 <sup>b</sup>	0.43 <sup>b,c</sup>	0.33 <sup>c</sup>	0.28 <sup>c</sup>
	28	1.63 <sup>a</sup>	0.83 <sup>b</sup>	0.73 <sup>b</sup>	0.48 <sup>c</sup>	0.35 <sup>c</sup>
MYC 10 <sup>4</sup> (cfu/g)	0	0.48 <sup>a</sup>	0.15 <sup>b</sup>	0.15 <sup>b</sup>	0.15 <sup>b</sup>	0.15 <sup>b</sup>
	7	0.73 <sup>a</sup>	0.25 <sup>b</sup>	0.23 <sup>b</sup>	0.20 <sup>b</sup>	0.20 <sup>b</sup>
	14	0.95 <sup>a</sup>	0.35 <sup>b</sup>	0.30 <sup>b</sup>	0.20 <sup>b</sup>	0.18 <sup>b</sup>
	21	0.98 <sup>a</sup>	0.50 <sup>b</sup>	0.45 <sup>b,c</sup>	0.30 <sup>b,c</sup>	0.20 <sup>c</sup>
	28	1.55 <sup>a</sup>	0.60 <sup>b</sup>	0.58 <sup>b</sup>	0.40 <sup>b,c</sup>	0.28 <sup>c</sup>

\*Control: Cheese sample made without adding CO<sub>2</sub>

- SPC: Standard plate count, PBC: Psychrotrophic bacterial count, MYC: Mold and yeast count

\*\*Values are log 10 cfu/g means, based on four replicates.

- Means with the same letter in the same row are not significantly different.

### Sensory properties

The results of the sensory assessment of cheese are given in Table 5. The carbonated cheese treated with the highest level of CO<sub>2</sub> always had the highest flavor score compared to the control. The flavor of carbonated cheese treated with the higher CO<sub>2</sub> levels was improved as storage period progressed. All carbonated cheeses ranked higher flavor scores than the control at 21 up to 28 d. This may be due to the lower levels of proteolysis, which related to lower bacteria counts in those cheeses (Table 4). Montilla *et al* (1995) demonstrated that cheeses produced from CO<sub>2</sub>-treated milk showed less proteolysis than control cheeses, but no significant differences in sensory characteristics between cheeses were detected. At 21 d of storage, the control cheese exhibited a slight bitterness, and developed off-flavour and putrid aroma, a common defect in such types of cheese which, may be attributed to the greater degree of

proteolysis that associated with the high moisture and low salt content (Mistry, 2001; Ma *et al* 2003).

The CO<sub>2</sub> addition to cheese milk decreased proteolysis via at least two mechanisms: the reduction of microbial proteases due to a reduced microbial growth and the possible reduction of endogenous protease activity due to a lower milk pH. Because of the low levels of CO<sub>2</sub> used in the present study, the common tactile sensation associated with CO<sub>2</sub>-containing beverages was not detected in any of carbonated samples. Chen *et al* (1992) found that the lowest flavor threshold for CO<sub>2</sub> in milk was between 4.54 and 9.10 mM. Similarly, Moir *et al* (1993) found that 10 mM CO<sub>2</sub> injected into Cottage cheese cream dressing could significantly increase shelf life without affecting flavor.

At both, 7 and 14 d, all carbonated cheeses had body and texture scores not significantly ( $P \geq 0.05$ ) different from those of the control. However, the

Table 5. Sensory properties\*\* of the control and carbonated cheeses during 28 d of storage at 4°C

Attributes	Storage period (d)	Treatments				
		Control*	3L/min	5L/min	7L/min	9L/min
Flavour (60)	0	52.00 <sup>a,b</sup>	45.00 <sup>c</sup>	46.25 <sup>c</sup>	48.00 <sup>b,c</sup>	54.25 <sup>a</sup>
	7	50.75 <sup>b</sup>	50.50 <sup>b</sup>	51.50 <sup>b</sup>	52.50 <sup>a,b</sup>	55.50 <sup>a</sup>
	14	45.25 <sup>c</sup>	48.75 <sup>b</sup>	48.25 <sup>b,c</sup>	50.75 <sup>b</sup>	55.75 <sup>a</sup>
	21	43.00 <sup>c</sup>	45.00 <sup>b</sup>	48.25 <sup>a,b</sup>	50.50 <sup>a,b</sup>	55.00 <sup>a</sup>
	28	27.50 <sup>d</sup>	37.50 <sup>c</sup>	41.25 <sup>c</sup>	50.75 <sup>b</sup>	56.00 <sup>a</sup>
Body & Texture (30)	0	25.75 <sup>b</sup>	25.50 <sup>b</sup>	25.25 <sup>b</sup>	26.75 <sup>a,b</sup>	28.50 <sup>a</sup>
	7	24.75 <sup>a</sup>	23.25 <sup>a</sup>	23.25 <sup>a</sup>	24.75 <sup>a</sup>	25.75 <sup>a</sup>
	14	22.50 <sup>a</sup>	23.00 <sup>a</sup>	23.50 <sup>a</sup>	25.00 <sup>a</sup>	25.00 <sup>a</sup>
	21	22.50 <sup>b</sup>	23.00 <sup>b</sup>	23.50 <sup>a,b</sup>	25.00 <sup>a,b</sup>	26.00 <sup>a</sup>
	28	21.25 <sup>b</sup>	22.50 <sup>a,b</sup>	22.50 <sup>a,b</sup>	25.00 <sup>a</sup>	25.00 <sup>a</sup>
Salt ness (5)	0	4.25 <sup>a</sup>	3.75 <sup>a</sup>	3.50 <sup>a</sup>	4.00 <sup>a</sup>	4.25 <sup>a</sup>
	7	3.75 <sup>a</sup>	3.50 <sup>a</sup>	3.25 <sup>a</sup>	3.75 <sup>a</sup>	3.75 <sup>a</sup>
	14	4.00 <sup>a</sup>	3.75 <sup>a</sup>	3.50 <sup>a</sup>	4.00 <sup>a</sup>	4.50 <sup>a</sup>
	21	3.50 <sup>a</sup>	3.50 <sup>a</sup>	3.50 <sup>a</sup>	3.75 <sup>a</sup>	3.75 <sup>a</sup>
	28	3.75 <sup>a</sup>	3.75 <sup>a</sup>	3.75 <sup>a</sup>	3.75 <sup>a</sup>	3.75 <sup>a</sup>
Appearance (5)	0	4.25 <sup>a</sup>	4.00 <sup>a</sup>	4.00 <sup>a</sup>	4.25 <sup>a</sup>	4.25 <sup>a</sup>
	7	4.00 <sup>a</sup>	3.75 <sup>a</sup>	3.75 <sup>a</sup>	4.00 <sup>a</sup>	4.00 <sup>a</sup>
	14	4.00 <sup>a</sup>	3.75 <sup>a</sup>	3.75 <sup>a</sup>	4.00 <sup>a</sup>	4.25 <sup>a</sup>
	21	3.75 <sup>a</sup>	3.75 <sup>a</sup>	4.00 <sup>a</sup>	4.00 <sup>a</sup>	4.00 <sup>a</sup>
	28	3.25 <sup>b</sup>	3.00 <sup>b</sup>	3.00 <sup>b</sup>	3.50 <sup>b</sup>	4.25 <sup>a</sup>
Total (100)	0	86.25 <sup>b</sup>	78.25 <sup>d</sup>	79.00 <sup>d</sup>	83.00 <sup>c</sup>	91.25 <sup>a</sup>
	7	83.25 <sup>b,c</sup>	81.00 <sup>c</sup>	81.75 <sup>c</sup>	85.00 <sup>b</sup>	89.00 <sup>a</sup>
	14	75.75 <sup>d</sup>	79.25 <sup>c</sup>	79.00 <sup>c</sup>	83.75 <sup>b</sup>	89.50 <sup>a</sup>
	21	72.75 <sup>d</sup>	75.25 <sup>d</sup>	79.25 <sup>c</sup>	83.25 <sup>b</sup>	88.75 <sup>a</sup>
	28	55.75 <sup>e</sup>	66.75 <sup>d</sup>	70.50 <sup>c</sup>	83.00 <sup>b</sup>	89.00 <sup>a</sup>

\*Control: Cheese sample made without adding CO<sub>2</sub>

\*\*Mean of four replicates

-Means with the same letter in the same row are not significantly different.

body and texture scores of the carbonated cheeses became significantly ( $P \leq 0.05$ ) higher than those of the control cheese at 21 up to 28 days old (Table 5). The panelist's comments indicated that the control cheese had a rubbery, dry and hard body and texture while carbonated cheeses were much less rubbery and softer than the control. The use of the CO<sub>2</sub> improved the body and texture of the carbonated cheeses over the control up to 28 d. Also, these results could be due to the lower proteolysis levels in carbonated cheeses compared to the control. The body and texture score of all cheese samples tended to decrease as storage period increased but the rate of this decrease was faster in control cheese than that in carbonated ones. A similar

trend was reported by Katsiari and Voutsinas (1994) for low-fat Kefalograviera cheese.

Table (5) shows also that the carbonated cheeses had saltiness scores that were not significantly ( $P \geq 0.05$ ) different from those of the control cheese throughout the storage period. This may be due to that the used rates of salt were the same for all cheeses. The appearance of the carbonated cheeses was considered good over storage period and they were comparable ( $P \geq 0.05$ ) to that of the control. Cheese samples were examined at both 21 and 28 d of storage for visible microbial growth on the product surface. These results show a considerable reduction in the number of spoiled samples using CO<sub>2</sub>, while the control cheese ex-

hibited a spoilage in 21 d at 4°C, which indicated by a mold growth, discoloration and slime formation. The overall quality of karish cheese was significantly ( $P \leq 0.05$ ) affected by the level of the CO<sub>2</sub> over entire storage period (Table 5). All carbonated cheeses gained significantly ( $P \leq 0.05$ ) higher scores for total assessment than the corresponding control cheese at 14 d up to 28 d.

Many authors reported that, organoleptic properties of CO<sub>2</sub>-treated cheese were as good as (Ruas-Madiedo *et al* 1998; Ruas-Madiedo *et al* 2002) or better than (Maniar *et al* 1994; McCa-rney *et al* 1995; Montilla *et al* 1995) control samples made from untreated milk.

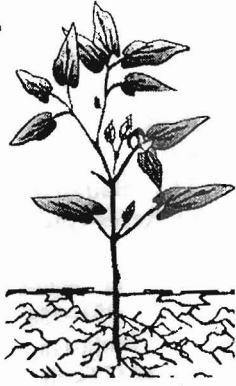
In conclusion, CO<sub>2</sub> can be used as an effective tool of extending shelf-life of Kareish cheese without affecting sensory properties. The shelf-life of Kareish cheese of the present study at normal refrigeration temperatures was in the range of about 21 to 28 d or longer. In other words, Karish cheese of this study had a reduce risk of spoilage as compared to conventional Karish cheese of the same age and, therefore, it had an increased safety margin for the consumer.

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## تأثير المعاملة بغاز ثاني أكسيد الكربون على فترة الصلاحية في الجبن القريش

[٣٥]

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عنه في جبن المقارنة بينما لم يتم اكتشاف بكتريا للقولون سواء في جبن المعاملات أو المقارنة. وقد كان التأثير المثبط أكثر وضوحا مع استخدام المستويات الأعلى من الغاز. كما أظهرت نتائج التحكيم الحسي ثباتا في الخواص الحسية في جبن المعاملات على مدار فترة التخزين وحصلت على درجات تحكيم أعلى من نظيرها في جبن المقارنة بداية من اليوم الرابع عشر من التخزين والذي بدأ عنده ظهور بعض مظاهر التدهور في جبن المقارنة بلغت ذروتها بحلول اليوم الحادي والعشرين بينما ظلت جبن المعاملات محتفظة بحالتها الطازجة ومستوى جودتها حتى بعد ٢٨ يوما من التخزين مما يدل على أن حقن اللبن المعد لصناعة الجبن القريش بغاز ك<sup>٢</sup> يمكنه إطالة فترة الصلاحية إلى أكثر من ٢٥% دون الإضرار بالخواص الحسية للجبن الناتج.

استهدف هذا البحث دراسة إمكانية إطالة فترة صلاحية الجبن القريش المخزن داخل الثلجة باستخدام غاز ك<sup>٢</sup> نظرا لقصر فترة الصلاحية في هذا النوع من الجبن. حيث تم حقن اللبن الفرز المستخدم بالغاز بمعدل ٣، ٥، ٧، ٩ لتر/ق للحصول على مستويات مختلفة من الغاز في اللبن تراوحت بين ١٠-٣٥ مج/١٠٠ جم مما أدى إلى خفض رقم الـ pH في اللبن من ٦.٦ إلى ٥.٨-٦.٤. ثم صنعت الجبن القريش بالطريقة التقليدية من كل من اللبن المعامل واللبن غير المعامل وتم تخزين الجبن الناتج على درجة حرارة ٤م<sup>٥</sup> لمدة ٢٨ يوما لإجراء الاختبارات الميكروبيولوجية والحسية. وقد أشارت النتائج إلى أن حقن اللبن بغاز ك<sup>٢</sup> أدى إلى انخفاض العدد الكلى للبكتريا وأعداد البكتريا المقاومة للبرودة وأعداد الخمائر والفطريات في الجبن الناتج