Effect of Sodium Chloride and Milk Preacidification on Serum Phase and Chemical Composition of Soft White Pickled Cheese

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ABSTRACT

The objective of the present work was to study the effect of milk salting and preacidification with citric or acetic acid on residual coagulant activity, expressible serum and chemical composition of soft white cheese. Also, to produce soft white cheese from unsalted milk with characteristics comparable to those of cheese made from salted milk. The results showed that cheese made from salted milk contained the lowest activity of residual coagulant, while cheese made from milk preacidified with citric acid contained the highest activity. Cheese moisture and soluble proteins in expressible serum were lower in cheese made from unsalted milk than in that made from salted milk. The expressible serum decreased during pickling in all treatments. Preacidification with citric acid to pH 6 increased the moisture content in cheese and reduced the amount of expressible serum. Reverse-Phase High Performance Liquid Chromatography (RP-HPLC) analysis showed that the peptides in the expressible serum from salted milk-cheese were present at higher relative concentrations than in that from the unsalted milk-cheese. The late-eluting peptides of 90 days old cheese were higher in preacidified milk-cheese compared to that in none preacidified milk-cheese.

Keywords: soft white cheese, residual coagulant activity, expressible serum, preacidification.

INTRODUCTION

Domiati cheese is the most popular soft white cheese produced in Egypt. This cheese is unique among cheese varieties in terms of addition of large quantities (8-15%) of NaCl to milk before renneting (Abd El-Salam *et al.*, 1993). Calcium plays an important role in cheese manufacturing as well as texture properties of Domiati cheese. Addition of NaCl to milk solubilizes part of the colloidal calcium (Zoon , *et al.*, 1989, Casiraghi & Lucisano, 1991, Gatti & Pires 1995, Gaucheron, *et al.*, 2000). In buffalo's and cows' milk, about 23 – 25 % of the colloidal calcium can be solubilized by addition of NaCl (Abd El-Salam *et al.*, 1993). The colloidal calcium phosphate (CCP) dissociates from the casein micelle as a function of NaCl added or pH reduction (Creamer 1985, Gatti & Pires, 1995, Gaucheron, *et al.*, 2000, Lucey, *et al.*, 2003). The calcium content in Mozzarella cheese and the calcium as a percentage of protein (Ca : protein) decreased significantly with milk preacidification, and citric acid caused larger reductions in cheese calcium content than acetic acid (Metzger, *et al.*, 2001).

The level of expressible serum has been used as an indirect measure of the water-holding capacity (WHC) of cheese, with a low level indicating a high WHC (Kindstedt, 1995, Kindstedt & Guo, 1997). The expressible serum of soft and semi soft cheeses can be removed by centrifugation (Lucey, *et al.*, 2003). Diffusion of salt into cheese increases the ionic strength and osmotic pressure of the aqueous phase (Geurts, *et al.*, 1974), which could make it more difficult to express the aqueous phase by low pressure. Guinee, *et al.* (2002) found that reducing the calcium-to-casein ratio from ~ 28 to ~ 21 mg / g, by preacidification of milk prior to renneting, resulted in higher level of moisture in Mozzarella cheese and a lower protein level of the expressible serum.

The high salinity level of whey obtained during Domiati cheese manufacture has no any benefit and makes its disposal a problem. So, modification in Domiati cheese processing using unsalted milk is required.

The objectives of the present work were to study the effect of milk salting and preacidification on expressible serum, residual coagulant activity and chemical composition of cheese.

MATERIALS AND METHODS

Cheese making

Raw milk was obtained from the dairy farm, Faculty of Agriculture, Alexandria University. Three

replicates of white soft cheese were manufactured from full fat cow milk (fat 3.2%). Cheese milk was assigned to two double stainless steel cheese vats (Department of Dairy Science, Faculty of Agriculture, Alexandria University). The following four treatments of cheese were employed, T_1: cheese made by the traditional method using salted milk (12 % NaCl, w/w), T_2: cheese made from unsalted or preacidified milk (untreated milk), T_3: cheese made from unsalted milk preacidified with acetic acid to pH 6.0, and T_4: cheese made from unsalted milk preacidified with citric acid to pH 6.0. Soft white cheese was made according to the method described below, which was adapted from Hayaloglu *et al.* (2002). The raw milk was pasteurized at 63°C for 30 min and then cooled to 4°C. The milk was transferred to four cheese vats, 10% solutions of citric acid and acetic acid were added individually to vats 3 and 4 to reduce the pH to 6. The milk was heated to 40°C and the pH was measured after 30 min equilibration. Thermophilic starter culture (*Sterptococcus thermophilus*, *Lactobacillus bulagricus* and *Lactobacillus helveticus*, direct in vat set, FRC60, Chr. Hansen, Denmark) was added at level of 0.03% (w/w), and $CaCl_2$ at level of 0.02% (w/w). The inoculated milk was held for 1 hr, and the NaCl was added to vat 1 at level of 12% (w/w). Suitable amount of commercial calf rennet was added to coagulate the milk throughout 90 min. The curd was then transferred to moulds, which varied in sizes and were lined with cheese cloth. The surface of the cheese was covered with cheese cloth. After 2–3 hr, a plate and weights (20-25kg for each 100kg of cheese milk) were placed to compact the curd. The weights were removed after 4-6 hr and the cheese mass was divided with a knife into blocks of about 9 × 9 × 9cm, weighing 450–500 g. The cheese blocks were then arranged in the cans that were filled with pasteurized (65°C/30 min) brine (15% NaCl for unsalted milk-cheese and 12% for salted milk-cheese). The cans were closed and stored at room temperature (20–25°C) for 90 days. The salt concentration in the brine was checked weekly and adjusted to 12% in all treatments during pickling.

Cheese composition

Cheese samples were analyzed for moisture by oven method (AOAC, 2000), salt by chloride analyzer (model 926, Nelson Jameson Inc., Marshfield, WI), fat by the Gerber method (AOAC, 2000) and total protein by the macro-Kjeldahl (AOAC, 2000). The pH was measured, using pH meter model Jenway 3505, in slurry prepared by macerating 20g of grated cheese in 20mL of deionized water.

Expressible water

Water that is not impeded by the protein matrix and may be expressed by centrifugation is known as expressible water (McMahon, *et al.*, 1999). Cheese samples were centrifuged at 19,500 xg for 60 min at 25°C. The expressed serum and fat were collected in a preweighed graduated cylinder that was held at 4°C until the surface layer of fat had solidified. The solidified fat layer was punctured to release the supernatant serum, which was poured off and weighed. The NaCl concentration and dry matter of serum were measured. The expressible water was calculated as the expressed fluid minus dry matter. Because samples contained different salt concentrations at the first day of manufacture, the non-salt expressible serum was calculated as the expressed fluid minus NaCl concentration.

Reverse-Phase-High Performance Liquid Chromatography (RP-HPLC)

The profile of peptides in expressible serum were separated using the C18-Lichrospher analytical (column 250 × 4.6mm, 5µm, Perkin Elmer, Norwalk, CT, USA). Samples were eluted with a four-step linear gradient over a period of 75 min according to Awad, *et al.* (1998). Separation was conducted at 21°C and peptides were monitored at 214 nm.

Determination of residual chymosin activity

The residual coagulant activity was determined by the method described by Hurley, *et al.* (1999) with some modifications. Fifty mg of cheese sample were weighed into a 1.5ml microcentrifuge tube to which 1ml of 0.05M trisodium citrate was added. The tubes were agitated at room temperature using a vortex mixer, to disperse the cheese. The peptides resulting from hydrolysis of the substrate (Pro-Thr-Glu-Phe-[NO_2-Phe]-Arg-Leu, Bachem, Switzerland), by the action of coagulant, were separated by HPLC according to Awad *et al.* (2005). The concentration of residual coagulant activity was expressed as RU kg^{-1} of cheese using a standard curve of chymosin activity (Fig. 1).

Statistical analysis

Data reported were the average of three measurements. The Statistical Analysis Software Package (SAS, 1999) was used for analysis of variance. Differences were considered significant at $P<0.05$.

RESULTS AND DISCUSSION

Chemical composition of the cheese

The chemical composition of the cheese samples is shown in Table (1). Fresh cheese made from salted milk had a higher moisture content ($P<0.05$) and lower fat and protein than the cheese made from untreated milk (T_2), but there were no significant differences in fat and protein in dry matter among treatments. Displacement of calcium from casein micelles by adding salt to milk may cause increased hydration or salvation of casein (Creamer, 1985). Furthermore, Abd El-Salam *et al.* (1993) reported that the moisture content of Domiati cheese, fresh or pickled, increased with increasing the concentration of salt added to milk. A high salt content weakens the cheese curd and it retains more moisture. Preacidified milk fresh cheeses had higher moisture ($P<0.05$), and lower fat and protein than untreated milk-cheese. After 30 days of pickling, the moisture content of all treatments significantly decreased, while the fat and protein in dry matter did not change. Salting the cheese normally promotes syneresis and decreases the moisture content of cheese (Kindstedt, *et al.*, 1992, Guinee & Fox, 1993, Pastorino, *et al.*, 2003). During pickling at room temperature, cheese made from untreated milk (T_2) lost moisture at higher rate than the other three treatments (T_1, T_3 and T_4). Solubilization of CCP from casein micelles by adding salt or acid to milk may cause increased hydration of casein (Creamer, 1985, Guo, *et al.*, 1997), and may exhibit syneresis during pickling at room temperature.

Cheese pH

The pH of cheese samples was significantly influenced ($P<0.05$) by treatments and the cheese age (Table 1). There were significant differences ($P<0.05$) among all treatments at the first day of manufacture, the highest pH was in salted milk-cheese while the lowest was in preacidified milk-cheese with citric acid. However, the pH decreased in all treatments during pickling. The pH of all aged cheese was in the normal range of Domiati cheese. Abd El-Salam *et al.* (1993) reported that the pH of pickled Domiati cheese closes to the isoelectric point of caseinate and partially solubilizes the colloidal calcium which causes shrinkage of the cheese matrix and exudation of cheese serum into the pickle.

Residual coagulant activity in soft white cheese

The residual coagulant activity for fresh cheese made from salted milk was 25.68 RU kg^{-1}, while it was 31.46 in cheese (T_2) made from unsalted milk (Table 1). When the cheese was made from preacidified milk, the residual coagulant activity increased to 37.69 and 44.36 RU kg^{-1} for acetic acid

Table 1: Chemical composition (%), pH value and residual coagulant activity of soft white cheese during pickling

Cheese code	Pickling time (days)	Moisture	Fat (WB)	Total protein (WB)	Fat (DB)	Total protein (DB)	Salt	pH	RCA
T_1	1	60.83^{b}	18.50^{h}	11.89^{ed}	47.23	30.35	9.01	6.22^{a}	25.68^{d}
	30	57.63^{d}	20.40^{f}	12.18^{cd}	48.15	28.75	9.28	5.86^{b}	12.86^{g}
	90	54.92^{f}	21.60^{e}	12.84^{b}	47.91	28.48	9.65	5.42^{d}	ND
T_2	1	58.85^{cd}	19.70^{ef}	12.72^{b}	47.87	30.91	0.00	5.89^{b}	31.46^{c}
	30	53.92^{f}	22.00^{b}	13.61^{a}	47.74	29.54	8.16	5.45^{d}	18.93^{f}
	90	49.58^{g}	22.60^{a}	13.89^{a}	44.82	27.55	9.25	5.25^{e}	ND
T_3	1	62.83^{a}	17.20^{i}	10.97^{d}	46.27	29.51	0.00	5.70^{c}	37.69^{b}
	30	59.94^{bc}	19.40^{g}	12.05^{cd}	48.43	30.08	8.05	5.33^{e}	21.75^{e}
	90	56.08^{e}	21.05^{de}	12.37^{c}	47.93	28.16	9.78	5.17^{f}	ND
T_4	1	63.40^{a}	17.10^{i}	10.78^{e}	46.72	29.45	0.00	5.51^{d}	44.36^{a}
	30	59.25^{c}	19.30^{g}	12.08^{cd}	47.36	29.64	7.36	5.24^{e}	27.34^{d}
	90	56.22^{e}	21.10^{d}	12.41^{c}	48.20	28.35	9.12	5.06^{g}	ND

T_1: Cheese made by the traditional method using salted milk (12%, NaCl, w/w), **T_2**: Cheese made using untreated milk, **T_3**: Cheese made from unsalted milk preacidified with acetic acid to pH 6.0, and **T_4**: Cheese made from unsalted milk preacidified with citric acid to pH 6.0.
WB = Wet basis **DB** = Dry basis
RCA= Residual Coagulant Activity (RU kg^{-1} cheese) **ND** = Not determined
a, b, c, d, e, f, h. i means within the same column with different subscripts are significantly different ($P<0.05$).

and citric acids, respectively. At lower pH values, the number of positive charges on all caseins and on chymosin would be larger, yielding a greater probability for association between positive groups on the one protein and negative ones on the other (Larsson, *et al.*, 1997). This would imply that at low pH, chymosin can associate with casein, which agrees with the marked increase in adsorption of chymosin, with decline pH.

The residual coagulant activity decreased in all cheeses after 30 days of pickling at room temperature. The reduction was in the range of 38 to 50%, being at higher rate in cheese made from salted milk.

The obtained results showed that there were great effects of NaCl and pH on chymosin adsorption of soft white cheese. It may also be considered that the size and voluminosity, i.e. the surface, of the casein micelles may affect the possibilities for chymosin adsorption, and these properties may well vary with pH and casein composition (Larsson *et al.*, 1997, Larsson & Andrén, 1997).

Expressible serum of soft white cheese

Table (2) shows the expressible serum composition of soft white cheese during pickling. A large amount of non NaCl expressible serum was obtained from fresh cheeses (17.5 to 21.5% of cheese), and this expressible serum contains approximately 2–3% protein (Table 2). The free NaCl expressible serum was lower for cheeses made with salted milk or preacidified milk compared to that made with untreated milk, and the expressible serum was lower for cheese made from preacidified milk with calcium chelating (citric acid) than that made from preacidified milk with acetic acid.

The non NaCl dry matter and protein content of expressible serum were higher in cheese made from salted milk than that made from untreated milk. The amount of protein and non NaCl dry matter in expressible serum were also higher in cheeses made from preacidified milk than the other two cheeses. The ratio of non-expressible water to cheese protein (NEWP) was higher in cheeses made from salted or preacidified milk than those made from untreated milk, indicating that the water holding capacity was lower in untreated milk-cheese.

After pickling for 30 days at room temperature, the amount of moisture content and non NaCl expressible water significantly decreased (Table 2), while the ratio of non-expressible water to cheese protein increased for all treatments (Table 2). An increase in salt in cheese moisture replaces calcium ions with sodium (Lawrence, *et al.*, 1987), thereby increasing the water holding capacity of casein (Ruegg *et al.*, 1991). When the CCP in milk is solubilized by acidification, the phosphate ions that are released from the nanocluster are rapidly proto-

Table 2: Composition of expressible serum of soft white cheese during pickling

Cheese code	Pickling time (days)	Parameters								
		ES	NSES	EWC	EWM	NEWP	Salt	DM	NSDM	Protein
T_1	1	19.82^{b}	17.50^{c}	16.73^{c}	27.50^{c}	3.71^{cd}	11.72	15.58	3.86^{e}	2.32^{g}
	30	9.76^{g}	8.54^{f}	8.16^{f}	14.15^{e}	4.06^{b}	12.55	16.43	3.88^{e}	2.57^{e}
	90	3.04^{j}	2.65^{i}	2.52^{i}	4.60^{h}	4.08^{b}	12.82	17.08	4.26^{c}	2.85^{d}
T_2	1	21.55^{a}	21.55^{a}	20.78^{a}	35.31^{a}	2.99^{f}	0.00	3.58	3.58^{f}	2.05^{h}
	30	11.17^{e}	9.88^{e}	9.45^{e}	17.52^{d}	3.27^{e}	11.57	15.44	3.87^{e}	2.25^{g}
	90	4.25^{i}	3.72^{h}	3.54^{h}	7.15^{f}	3.31^{e}	12.54	16.67	4.13^{d}	2.41^{f}
T_3	1	20.01^{b}	20.01^{b}	19.11^{b}	30.42^{b}	3.65^{d}	0.00	4.49	4.49^{c}	2.57^{e}
	30	12.75^{d}	11.34^{d}	10.72^{d}	17.88^{d}	4.08^{b}	11.03	15.93	4.90^{b}	2.89^{d}
	90	5.11^{h}	4.46^{g}	4.21^{g}	7.50^{f}	4.19^{a}	12.78	17.73	4.95^{b}	3.07^{c}
T_4	1	18.09^{c}	18.09^{c}	17.20^{c}	27.12^{c}	3.92^{c}	0.00	4.94	4.94^{b}	2.96^{cd}
	30	10.60^{f}	9.50^{e}	8.90^{ef}	15.02^{e}	4.17^{ab}	10.37	16.03	5.66^{a}	3.41^{b}
	90	4.52^{i}	3.92^{h}	3.66^{h}	6.51^{g}	4.24^{a}	13.18	18.96	5.78^{a}	3.52^{a}

T_1: Cheese made by the traditional method using salted milk (12%. NaCl, w/w). **T_2**: Cheese made using untreated milk, **T_3**: Cheese made from unsalted milk preacidified with acetic acid to pH 6.0, and **T_4**: Cheese made from unsalted milk preacidified with citric acid to pH 6.0.
ES= expressible serum % of cheese, **NSES** = non-salt expressible serum % of cheese, **EWC** = expressible water % of cheese, **EWM** = expressible water % of cheese moisture, **NEWP** = Ratio of non-expressible water to cheese protein, **DM** = dry matter % of expressible serum, **NSDM** = non salt dry matter.
a, b, c, d, e, f, g, h, i, j means within the same column with different subscripts are significantly different ($P<0.05$).

nated, which results in an increase in the hydrogen ion buffering in milk (Lucey, *et al.*, 1993b). The solubilization of CCP in cheese, the serum absorption occurs and the functional properties change (Lucey, *et al.*, 1993a). The inability of the cheese to express serum after extended storage indicates that the water phase contains enough protein and other constituents to prevent its release from the cheese. During refrigerated storage of Mozzarella cheese, the insoluble casein matrix appears to start to solublizie and interact with water phase of the cheese (Guo & Kindstedt, 1995, Guo *et al.*, 1997). Metzger *et al.* (2001) hypothesized that the interaction of solubilized casein with the water phase of Mozzarella cheese results in the observed decrease in the amount of expressible serum and the increase in the concentration of protein in the expressible serum.

The expressible water was lower in salted milk-cheese than in other three treatments at 90 days of pickling. There were no significant differences ($P<0.05$) between untreated milk-cheese and preacidified milk-cheese with citric acid in the expressible serum, non-salt expressible serum and expressible water, while the expressible water of cheese moisture was higher ($P<0.05$) and the ratio of non-expressible water to cheese protein was lower ($P<0.05$) for untreated milk-cheese than preacidified milk-cheese with citric acid. These suggested that the water holding capacity was higher in citric acid cheese than in untreated milk-cheese. The constituents of the water phase appear to progressively reach concentrations that allow them to form a gel that resists removal of water under the conditions of the test (i.e., 25°C and 19,500 xg).

Peptides profile analysis of expressible serum

The RP-HPLC profile analysis of expressible serum from cheese at 1 and 90 days of pickling are shown in Figs. (2 and 3). For convenience, the chromatograms were divided into four zones I, II, III, and IV, each of which contained one or more peaks. Major differences were evident between the peptide profiles of the expressible serum of untreated milk-cheese (T_2) and those from the other three treatments at the first day of manufacture (Fig. 2). Peaks II and III in the chromatograms of expressible serum from salted milk and preacidified milk-cheeses (Fig. 2, T_1, T_3 and T_4) were absent in the chromatogram of expressible serum from untreated milk-cheese (Fig. 2, T_2), suggesting that peptides corresponding to those peaks were solubilized by salt or acids. Peak II in the chromatogram of the expressible serum of the cheeses made from salted milk and preacidified milk with acetic acid (Fig. 2, T_1 and T_3) was present at very low relative concentrations in expressible serum of cheese made from milk preacidified with citric acid (Fig. 2, T_4), and this peptide was absent in expressible serum from cheese made from untreated milk (Fig. 2, T_2). Some differences were evident between the expressible serum from salted milk-cheese and preacidified milk-cheeses (Fig. 2, T_1, T_3 and T_4). Marked quantitative differences were apparent between the expressible serum from the acetic acid and citric acid cheeses (Fig. 2, T_3 and T_4). Late eluting peak (IV) in the chromatograms of expressible serum of cheeses made from untreated milk and preacidified milk (Fig. 2, T_2, T_3 and T_4) was absent in expressible serum from salted milk-cheese (Fig. 2, T_1).

Differences between the cheeses with respect to the relative concentrations of peptides (Fig. 2, T_1 and T_2) presumably reflect differences between these two cheeses with regard to the salt added to the milk. Most of the peptides in the expressible serum from salted milk-cheese were present at higher relative concentrations than in the expressible serum from the untreated milk-cheese. Guo & Kindsted (1995) concluded from increases in protein concentration in the expressed serum that a continual solublization of casein (mainly β-casein) from the protein matrix into the expressed serum had occurred. Subsequently, they proposed that the solublization of casein resulted from changes to the protein brought about by the addition of salt to the cheese, and that the serum in the fat-serum channels became converted into a hydrated paracasein gel (McMahon *et al.*, 1999, Lucey *et al.*, 2003). The larger area of late-eluting peaks in the unsalted milk-cheeses may reflect the accumulation of rennet-produced peptides, which are produced from β-casein by chymosin in absence of NaCl (Awad *et al.*, 1998).

The total peaks area, and in particular that of the middle eluting peaks increased after 90 days of pickling in all cheeses (Fig. 3). The results indicated that the accumulation of new proteolysis products during ripening and an increasing contribution of proteolytic enzymes to overall proteolysis, especially at low pH. After 3 months of pickling, the expressible serum of cheeses made from preacidified milk (Fig. 3, T_3 and T_4) had some late-eluting peaks (retention time >40 min), and which probably correspond to high MW and hydrophobic peptides, these peaks were absent in expressible se-

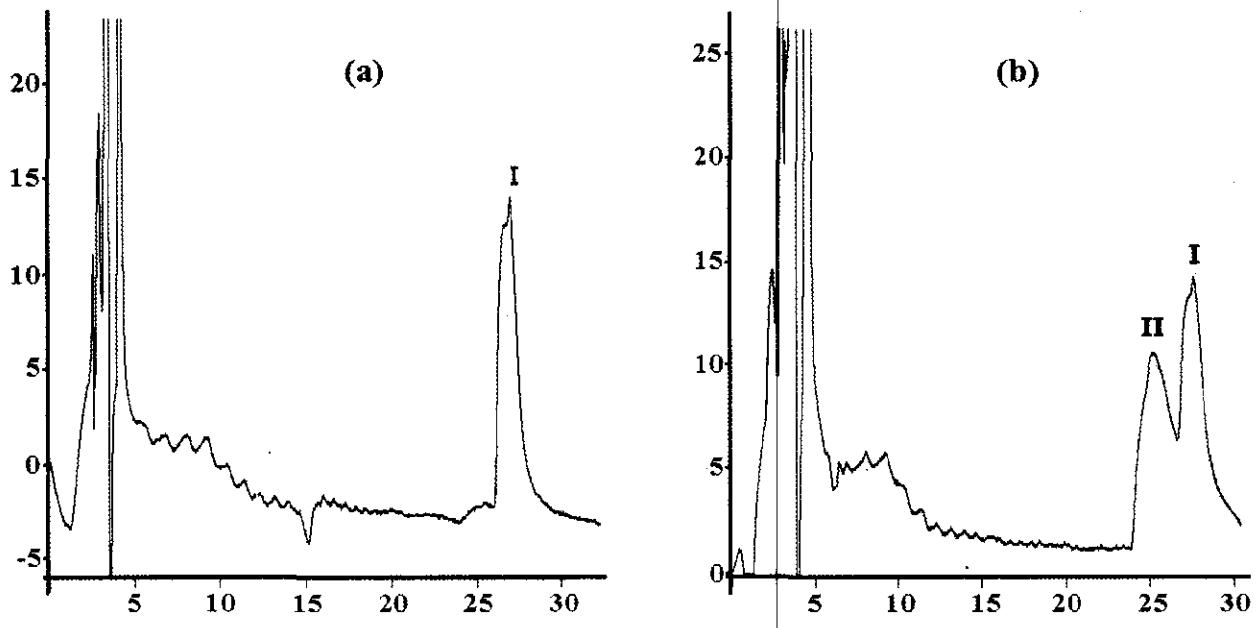

Fig. 1a,b: RP-HPLC chromatogram of substrate without addition of chymosin (a) and after addition of chymosin (b)
I: The substrate **II: The peptide resulting from hydrolysis of the substrate**

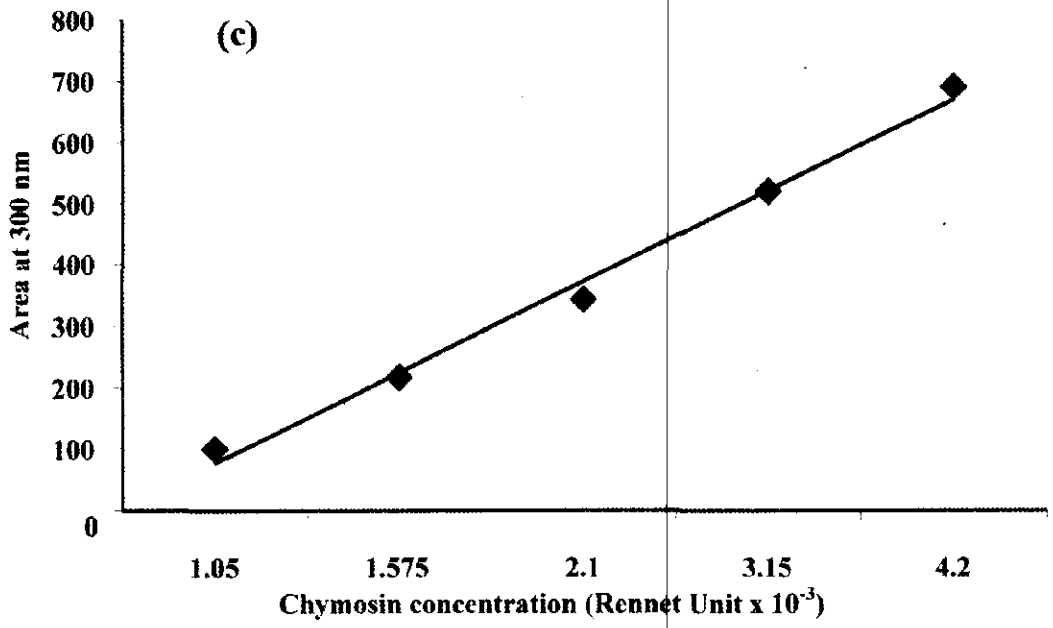

Fig. 1c: Standard curve of chymosin activity

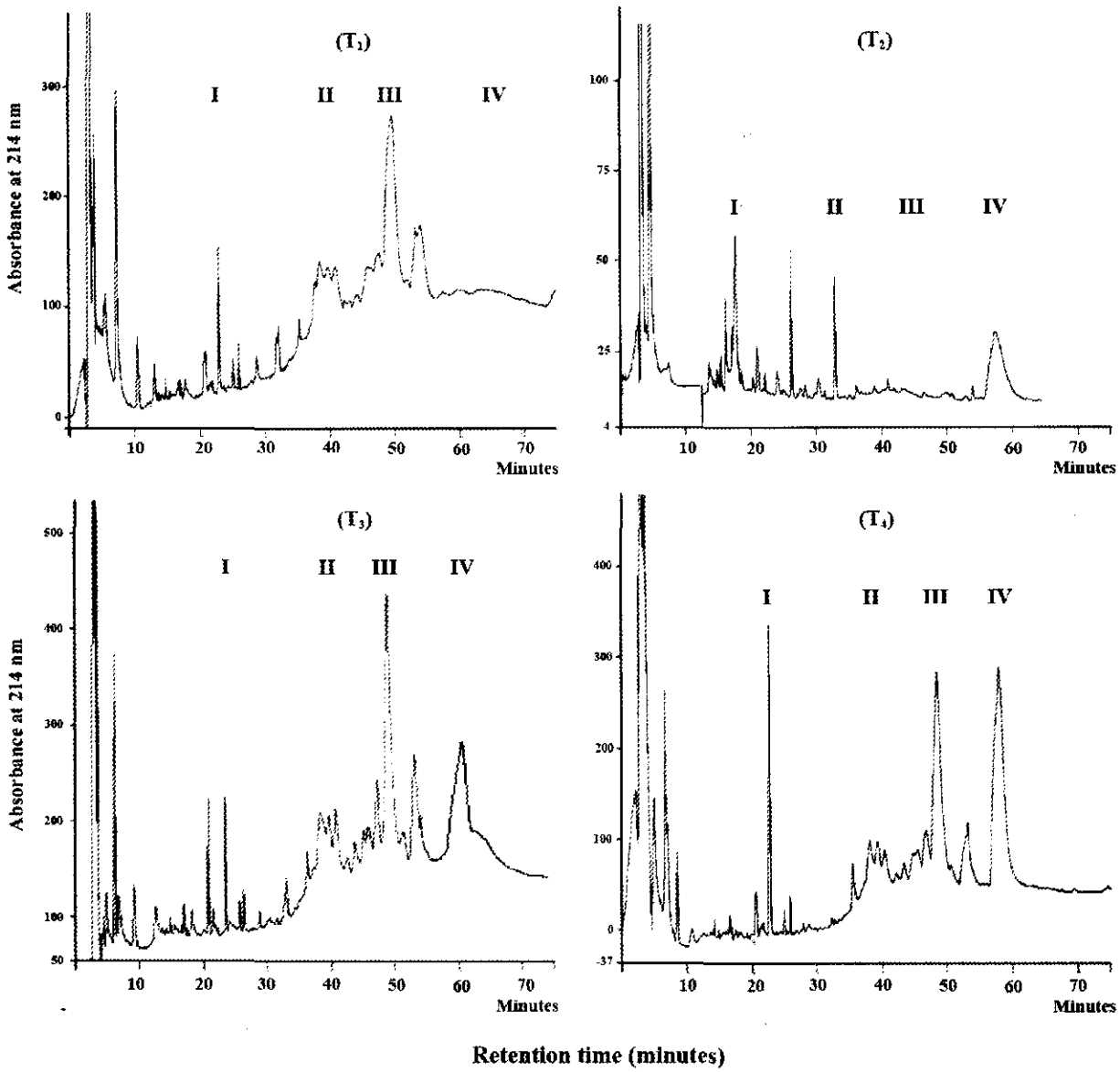

Fig. 2: RP-HPLC profile of expressible serum extract of soft white cheese at 1 day of manufacture

T_1: Cheese made by the traditional method using salted milk (12 % NaCl, w/w).
T_2: Cheese made from untreated milk.
T_3: Cheese made from unsalted milk preacidified with acetic acid to pH 6.0.
T_4: Cheese made from unsalted milk preacidified with citric acid to pH 6.0.

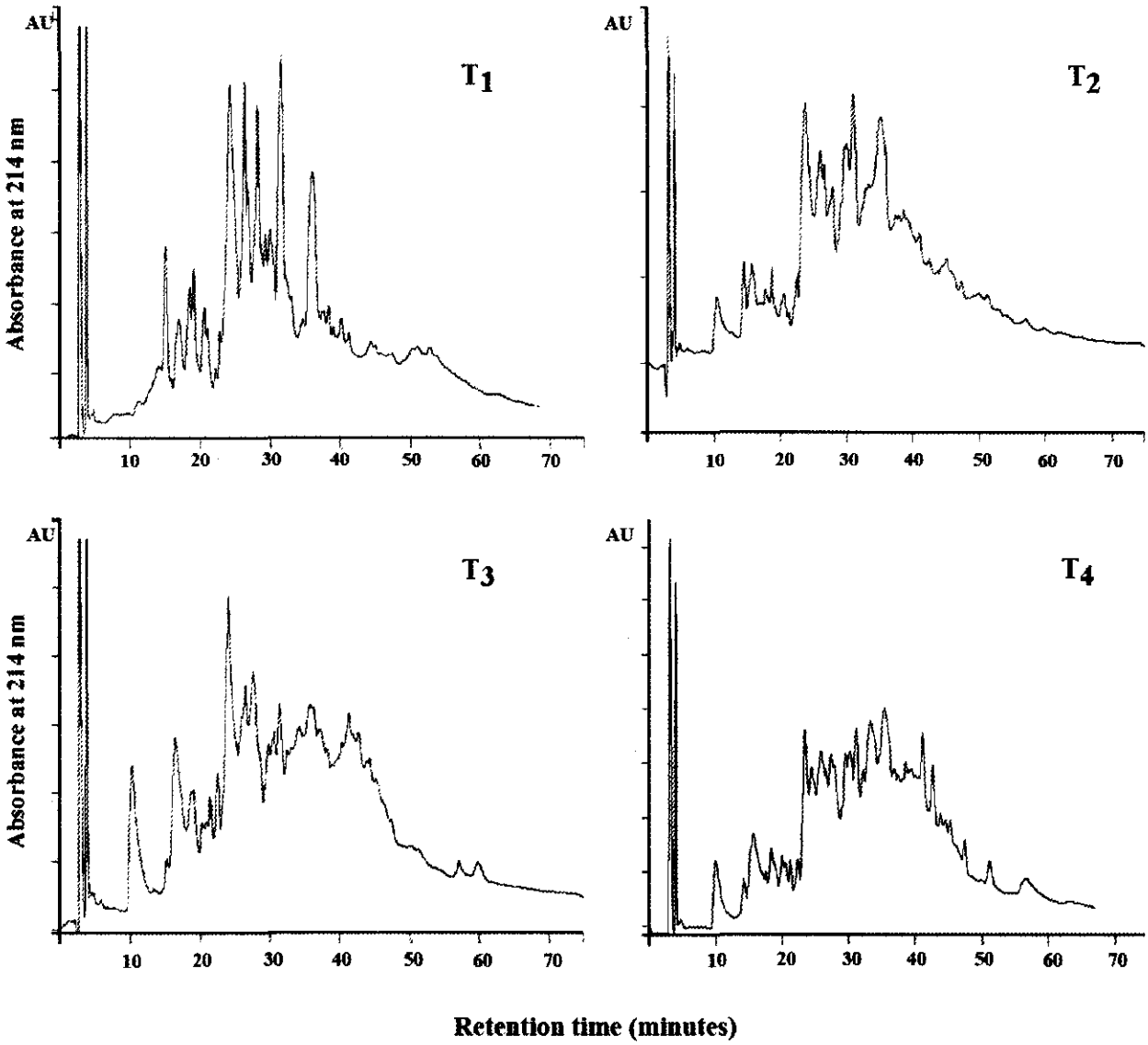

Fig. 3: RP-HPLC profile of expressible serum extract of soft white cheese at 90 days of pickling

T_1: Cheese made by the traditional method using salted milk (12 % NaCl, w/w).
T_2: Cheese made from untreated milk.
T_3: Cheese made from unsalted milk preacidified with acetic acid to pH 6.0.
T_4: Cheese made from unsalted milk preacidified with citric acid to pH 6.0.

rum from the other two cheeses (Fig. 3, T_1 and T_2). It is possible that solubilization of CCP, at lower pH value, exposes the phosphoserine residues and makes them more susceptible to hydrolysis (Fox, 1970). Cleavage of phosphopeptides would also reduce electrostatic repulsion between casein molecules in the cheese matrix (Lucey *et al.*, 2003), and would increase the peptides in expressible serum. The total peaks area was generally smaller in salted milk-cheese than in unsalted milk-cheese at 90 days of pickling, this is related to lower residual coagulant activity in salted milk-cheese than in the other three cheeses (Table 1).

CONCLUSION

Milk preacidification and salt addition reduced the moisture losing of white soft cheese during pickling at room temperature. The chemical and functional properties during pickling of resultant cheese were influenced by milk preacidification and salting. Preacidification of unsalted milk reduced expressible serum and produced cheese with characteristics comparable to those of cheese made from salted milk. Residual coagulant activity decreased in salted milk-cheese, while it increased in preacidification milk-cheese. The soluble proteins of expressible serum in the preacidification milk-cheese were higher than in that of control. The impact of preacidification on the proteolysis, rheological and sensorial aspects through pickling of resultant cheese is currently under preparation.

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تأثير كلوريد الصوديوم والتحميض الأولي للبن على الماء المرتبط وتركيب الجبن الأبيض المخلل الطري

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تمت دراسة تأثير تمليح اللبن وكذا تحميض اللبن الأولى باستخدام حمضي الخليك أو الستريك على الماء المرتبط والتركيب الكيماوي ومتبقيات الإنزيمات المجبنة فى الجبن الأبيض الطري خلال فترة التخليل، وذلك بهدف دراسة إمكانية تصنيع جبن أبيض طري من لبن غير مملح للحصول على قوام جيد والاستفادة من الشرش غير الملح. وقد أظهرت النتائج أن الجبن المصنع من لبن مملح لا يفقد الرطوبة بسرعة خلال فترة التخليل مقارنة بالجبن المصنع من لبن غير مملح، ولكن تحميض اللبن إلى pH ٦ باستخدام حمض الستريك أدى إلى زيادة قدرة الجبن على الاحتفاظ بالرطوبة وبذلك يعطي قواماً طرياً مرغوباً.

ولقد أظهرت النتائج أيضا ارتفاع نسبة متبقيات الإنزيمات المجبنة فى الجبن المصنع من اللبن غير الملح وغير المحمض مقارنة بذلك المصنع من لبن مملح وقد أدى التحميض إلى ارتفاع نسبة متبقيات الإنزيمات المجبنة وقد انخفض نشاط الإنزيمات المجبنة فى جميع المعاملات خلال فترة التخليل. وأظهرت النتائج ارتفاع نسبة البروتينات الذائبة فى سيرم الجبن المصنع من لبن مملح مقارنة بالجبن المصنع من لبن غير مملح وغير محمض. ولقد زادت نسبة البروتينات الذائبة فى سيرم الجبن عند تحميض اللبن، وهو ما تأكد بالتحليل باستخدام جهاز RP-HPLC.

كذلك فقد أدى تحميض اللبن إلى زيادة نسبة الببتيدات في سيرم الجبن المخلل لمدة ٩٠ يوماً ويرجع السبب في ذلك إلى زيادة نشاط متبقيات المنفحة.