

EFFECT OF THE CALF RENNET PASTE ON SOME PHYSIOCHEMICAL AND MICROBIOLOGICAL PROPERTIES OF BUFFALO'S PICKLED SOFT CHEESE

Hassan, Z.M.R and O. A. Aita

Food Sci. Dept., Fac. of Agriculture, Ain Shams University, Cairo, Egypt.

ABSTRACT

The effect of rennet paste on soft cheese making and quality was investigated. Two different calf rennet pastes (CRP1 and CRP2) were used. The rennet clotting time (RCT) increased as the fat content of the salted milk increased. Skim milk samples containing 12 % NaCl with CRP2 decreased, compared with the CRP1. The cheeses made by using the different rennets did not differ in their moisture, protein and fat content or in the total nitrogen, which resulted from proteolysis. The cheese produced with rennet CRP2 had the highest lipolysis than the cheese made with CRP1. The FFAs in the soft cheese made with CRP2 rennet paste was higher than the cheese made with CRP1. The value of TVFA increased during cheese ripening. The higher concentration of TVFA might be attributed to higher degree of fat hydrolysis being occurred by residual activity of enzyme and microflora. Fresh cheese showed the lowest thiobarbituric acid (TBA) value compared to ripened cheese. Cheese produced by CRP2 showed higher TBA values than that of CRP1 cheese sample. There was a remarkable inhibition in the growth of spore forming bacteria and yeasts & moulds. The α_1 -I- casein band was found in all cheese samples. Sensory quality attributes of cheese were improved with the prolongation of the ripening period. Ripening of cheese resulted in better flavor as well as body and texture.

Keywords: Rennet pastes; Soft cheese; Lipolysis; Proteolysis; Hygienic quality

INTRODUCTION

Cheese is the most popular dairy products in the worlds, produced in a wide range of types and forms throughout the world countries (Fox, 1993). Pickled white soft cheese, namely Domiati cheese, is the most popular soft cheese to the Egyptian consumer. After pickling, such cheese gains more preferable organoleptic properties. Soft cheeses are usually made in Egypt from buffalos and cows milks as well as their mixture.

The clotting of milk by rennet is a key passage in cheese making, which markedly could affect the characteristics of the produced cheese. Nowadays different types of rennet are available. They differ both in their origin (animal, vegetable, microbial and recombinant from genetically modified microorganism), and their physical state (liquid, powder or paste). The most used rennet derives from the abomasa (fourth stomach or vell) of unweaned calves. It is available as liquid or powder form. In Mediterranean countries, where sheep and goat breeding is largely widespread, it is common to use of lamb or kid rennet paste. The most important difference between calf rennet and lamb or kid rennet paste is due to the presence of lipolytic enzymes in the last one. This is due to the lipolytic enzymes are denatured during the activation process of chymosin and pepsin zymogens when calf rennet is produced. (Addis *et al.*, 2008).

The methods used for artisanal lamb rennet paste preparation depends on the region. In Sardinia, the manufacturing process provides that lambs are slaughtered at 25–30 days old. The abomasa are collected and the perivisceral fat removed, then they are salted and stratified in containers suitable to drain the organic liquid. Afterward, the abomasa are dried and ripened for at least 3 months. The abomasa finally are ground and blended into a paste (Pettinau *et al.*, 1977). In other regions of Southern Italy, the abomasa, removed from lambs slaughtered at 20–40 days old, are filled with milk (50–100 ml) before salting, then they are dried, ripened for 2 months and ground into a paste (Santillo *et al.*, 2005). The method of preparation of the lamb rennet paste is different in Spain (Bustamante *et al.*, 2000). Lamb stomachs are inspected immediately after that animals are slaughtered (4 weeks old), and selected on the basis of their appearance indicating that animals fed only milk. Stomachs are air dried in a ventilated room until a constant weight (45 days). External fat is then removed and the stomachs are cut open to remove any wool found inside. They are then ground, mixed with salt and ground again to obtain a paste. Paste is kept at 4 °C in covered glass jars for up to 1 year. Artisanal liquid lamb rennet is used in the traditional Feta cheese making. This rennet is prepared by cutting, mixing and extracting (with NaCl solution for 24 hr) dried and salted abomasa of lambs slaughtered before weaning (Georgala *et al.*, 2005). No activation procedures of chymosin are described for artisanal lamb rennet paste. However, most of extracts of rennet paste, particularly, those prepared from the stomachs of animals slaughtered after feeding, had pH values < 4.7. This could replace the activation by acidification of bovine rennet extracts. In the recent years, a great increase in the industrial rennet paste production has been reported. The industrial process did not differ substantially from the artisanal one. It provides that the abomasa, salted and stratified in containers, are ripened, in refrigerated rooms, for 3 months at least. The abomasa are then ground and blended to obtain a paste. In some areas of Upper Egypt (*fayoum* and *Mallawy*), the traditional dairies use a kind of rennet preparation locally called *Baladi* past rennet. This paste rennet is prepared from curdled milk collected from slaughtered suckling calves. The curdled milk is salted and packed in 16 kg metal tins, and kept at room temp until use when fresh and up to one year.

The aim of this work is to evaluate the effect of commercial local buffalo calf rennet pastes with different enzymatic characteristics on the proteolysis and Lipolysis of buffalo milk soft cheese with the same ripening period. Also, the specificity of the lipolytic and proteolytic system of such rennet paste was investigated.

MATERIALS AND METHODS

Fresh buffalo milk was obtained from the herd of the Faculty of Agricultural Ain Shams University, commercial buffalo calf rennet pastes were obtained from south Egypt, rennet paste (CRP1) from *Al-fayoum* and (CRP2) from *Mallawi*. Fine grade salt (sodium chloride) was obtained from El-Nasr Salines Company, Alexandria. Calcium chloride was obtained from El-Nasr Pharmaceutical Company.

Rennet was prepared by dispersing 1.0 g of rennet paste in 10 mL of water. The suspensions were stirred and the volume was made up to 100 mL with water and the suspension was employed for cheese making. This was first suitably diluted to about 110 milk clotting unit for each vat so that the clotting time was the same.

The enzyme milk-clotting activity (MCA) was measured by the method of Arima *et al.*, (1970), and proteolytic activity (PA) was determined through the estimation of soluble nitrogen and total nitrogen by the semi-micro-Kjeldahl method as described by Ling (1963).

Clotting time (s) indicated as time from rennet addition to the formation of the first visible floccules was visually measured. The enzyme solution (0.5 mL) was added to 10% solution of skim milk powder containing 10 mM calcium chloride (2.5 mL) at 35°C, the solution time elapsing between the mixing of reagents and the first appearance of solid material against the background was recorded.

For the preparation of the buffalo milk cream substrate, buffalo milk cream, obtained by centrifuging of fresh raw milk, was mixed with reconstituted skimmed milk to obtain a final fat concentration of 25% (w/w) and heated at 80 °C for 15 sec. to inhibit milk and microbial enzymes. The substrate was divided into samples of 25 g and kept at -20 °C.

For measuring of the lipolytic activity in each rennet, 30 g of paste was suspended in 70 mL of deionized water and stirred for 20 min at 20 °C. The rennet extract was then filtered through two layers of cheese cloth and the volume was made up to 100 mL with deionized water. An aliquot of the rennet extract (2.5 mL) was added to 25 g of buffalo milk cream substrate, and incubated at 37 °C for 24 h. A blank was prepared using 25 g of the same substrate without rennet extract being added. Two incubation trials were made for each rennet, and each was tested for lipolytic activity.

The lipolytic activity of each rennet was determined by titration of the FFA in the total volume of the incubated trails with NaOH 0.1 N until a pH of 8.5 was reached. Before titration, the incubated trail, without the fat being extracted, was stirred for 2 min to be homogenized. The rennet extract (2.5 ml) and the blank were also titrated in the same way. The lipolytic activity of each assay was calculated by subtracting the NaOH added to the blank and to the rennet extract from the NaOH volume added to the incubated trail. A lipolytic unit (LU) was defined as the amount of enzyme producing, on the buffalo milk cream substrate, after 24 h at 37 °C, (the amount of FFA titratable with 1 µeq of NaOH, until a pH of 8.5 was reached). The values are given as LU g⁻¹ for the rennet pastes described by Addis *et al.*, (2005)

Cheese was made by the method adopted by Fahmi and Sharara (1950). Fresh buffalo milk (6.5% fat, 15.83 %TS) was heated at 73 °C for 15 seconds then rapidly cooled to 45 °C. The milk was salted with sodium chloride at rate the rennet paste was added. The complete coagulated curd was scooped into wooden frame lined with cheese cloth for whey draining. After 24 h of whey drainage, the cheese curd was cut into pieces (~500g), and transferred to plastic containers. The obtained cheeses were pickled in their respective whey, and stored at room temperature. Samples were taken for analysis when fresh and after 60 days.

Cheese samples were examined when fresh and after 60 days for moisture, titratable acidity, soluble nitrogen (SN), total nitrogen (TN), ash and fat contents (AOAC, 2005), salt content (Bradly *et al* 1992) and total volatile fatty acids (TVFA) as ml 0.1N NaOH/100g cheese (Kosikowski,1982). Lactose content was determined according to Barnett and Tawab (1975). Values of pH were measured with a digital pH meter (Chemcadet, Cole-Palmer, and Chicago, IL). Thiobarbituric acid test (TBA) was colourimetrically determined according to the method described by Pearson (1977) as optical density (OD) at 532 nm wavelength using Spekoll II colorimeter. Distilled water was taken as a reference blank.

A method described by Laemmli (1970) was adopted for the polyacrylamide-gel electrophoresis qualitative study. The obtained SDS-PAGE patterns were identified as described by Basch *et al* (1985) and Farrell *et al.*, (2004).

Cheese samples were examined for the total bacterial count (TBC) according to Difco manual (1984). Yeasts and Moulds were counted on Malt-Extract Agar medium as suggested by Harrigan and McCance (1966).

Cheese samples were organoleptically scored for flavor (60 points), body and texture (30 points) and appearance (10 points) according to El-Koussy, (1966). Samples were judged by 10 staff members of Food Science Department, Faculty of Agriculture, Ain Shams University.

RESULTS AND DISCUSSION

Differences in the enzymatic composition of the calf rennet pastes CRP1&CRP2 may result in differences in the composition of the cheeses. The evolution during ripening of the physical and chemical parameters of the cheeses is reported in Table 1. There were no significant differences in pH values due to the type of rennet, but a significant difference was observed between the fresh cheese and the ripened cheese. No differences due to rennet type were observed in moisture, fat and protein contents.

Table (1): Physiochemical composition of calf rennet paste

Component %	Calf Rennet Past	
	CRP1	CRP 2
Moisture	77.55	75.79
Fat (In Dry Matter)	44.56	41.32
Protein (In Dry Matter)	37.38	34.74
Ash	5.99	5.06
Salt	6.25	6.20
Lactose	2.62	3.10
pH	4.18	4.18

CRP1= Calf rennet paste from *Al-fayum*, CRP 2= Calf rennet paste from *Mallawi*

The effect of sodium chloride concentration and fat content of skim and whole milk on the rennet clotting time is reported in Table 2. The results showed that RCT decreased with increasing NaCl concentration in skim milk compared with the whole milk in CRP1. The RCT increased as the fat content of the salted milk increased. Skim milk samples containing 12 % NaCl with

CRP2, RCT decreased as compared with the CRP1. RCT of skim milk samples without NaCl with both CRP1 and CRP2 decreased compared with the whole milk. This finding is in agreement with the known theory of the partial salting out of rennet enzyme as well as the cationic exchange taking place between add sodium and calcium ions of milk, thus extending collating time.

Table (2): Clotting activity of calf rennet paste as influenced by salt level and fat content in milk.

Clotted milk	RCT (min)	
	CRP 1	CRP 2
Skim milk		
0.0 % salt	3.37	2.34
12% salt	7.90	6.73
Whole milk		
0.0 % salt	3.85	3.40
12% salt	9.51	8.40

CRP1= Calf rennet paste from *Al-fayum*, CRP 2= Calf rennet paste from *Mallawi*

The data on proteolysis and lipolysis indices in the two rennet pastes are shown in Table 3. The results show that the SN/TN of CRP2 was higher than CRP1. The lipolytic activity of the two rennet pastes was determined using a natural substrate (buffalo milk cream substrate). The results were expressed in LU g⁻¹ (80 LU ml⁻¹ of extract) in CRP2 and (60 LU ml⁻¹ of extract) in CRP1. These results show that the lipolytic activity of CRP2 was 1.2 times higher than CRP1. The FFAs in the buffalo milk cheese made with CRP2 were higher than in the cheese made with CRP1.

Table (3): Proteolysis and lipolysis indices of calf rennet paste.

Calf Rennet Past	Proteolysis index			Lipolysis index		
	TN/DM	SN/DM	SN/TN	TVFA *	FFA **	TBA ***
CRP 1	5.44	4.07	74.89	48	8.46	0.723
CRP 2	5.86	4.94	84.30	52	10.01	0.664

CRP1= Calf rennet paste from *Al-fayum*, CRP 2= Calf rennet paste from *Mallawi*,
 SN= soluble nitrogen, TN= total nitrogen, DM= dry matter, * ml of 0.1 N Na OH / 100 g,
 ** FFA= free fatty acid, *** TBA= Thiobarbituric acid *** O. D. 532 nm

Table (4): Hygienic quality of calf rennet paste.

Microorganisms (CFU/g)	Calf Rennet Paste	
	CRP 1	CRP 2
Total count	1.85 + 5	1.60 + 5
Yeast & Moulds	1.6 + 3	2.4 + 3
<i>Enterococcus sp</i>	1.8 + 4	2.2 + 4
Coliform bacteria	absent	absent
<i>Staphylococcus aureus</i>	absent	absent

CRP1= Calf rennet paste from *Al-fayum*, CRP 2= Calf rennet paste from *Mallawi*

Differences in the enzymes activity of the rennet pastes may result in cause differences in the composition of the cheeses. The evolution of the

physical and chemical parameters of the cheeses during ripening is reported in Table 5. There were no differences in pH values due to the type of rennet, but a difference was observed between the fresh cheese and the ripened cheese. No obvious differences due to rennet type were observed in the moisture, fat and protein contents. The moisture content varied between the fresh cheese and the ripened cheese. The lactose content decreased in fresh cheese than the ripened cheese. During cheese ripening, the lactose content decreased in all cheese samples. The acidity development in the ripened cheese is mainly due to the hydrolysis of lactose content in the cheese by the resistant microorganisms.

Table (5): Physiochemical composition of soft cheese made with calf rennet paste.

Component %	Cheese samples		
	Fresh	CRP 1	CRP 2
Moisture	59.63	57.96	58.01
Fat (In Dry Matter)	50.78	47.58	54.78
Protein (In Dry Matter)	20.79	19.97	19.60
Ash	8.75	8.35	9.39
Salt	8.40	7.55	9.35
Lactose	4.95	2.42	2.81
pH	5.04	3.95	4.29

CRP 1= cheese sample with calf rennet paste from *Al-fayum*, CRP 2= cheese sample with calf rennet paste from *Mallawi*.

The data of the quantitative proteolysis indices, expressed as ratios between nitrogen fractions, are reported in Table 6. No differences were observed in the SN/TN ratio among the cheeses made with the different rennets. The greatest difference in SN/TN was at the end of 60 days ripening (61.2–64.9%) for all cheeses. The differences in the levels and types of proteolytic enzymes between rennets resulted in only slight differences in proteolysis (Piredda & Addis, 1998). The data of lipolysis in cheeses are shown in Table (6). The levels of the different FFAs at the end of ripening compared with the fresh cheese are reported in Table (6). The greatest difference in FFAs was in cheese CRP2 than in cheese with CRP1. The TVFA content was the highest in all cheese. The value of TVFA increased during cheese ripening. The higher concentration of TVFA might be attributed to higher degree of fat hydrolysis being occurred by the residual activity of enzyme. The general trend of the obtained results agreed with Akin *et al.*, (2003), who showed that the volatile free fatty acids (VFFAs) and free fatty acids (FFAs) increased with increased levels of lipase. Thiobarbituric acid (TBA) value of fresh cheese and ripened cheeses was also showed in Table (6). Fresh cheese contained the lowest TBA value, compared to the ripened cheese. Cheese with CRP2 contained higher TBA values than that of CRP1cheese. The changes in TBA value during storage period were mainly due to oxidation of unsaturated fatty acids, which apparently involved in the formation of malonaldehyd that reacts with TBA.

Table (6): Proteolysis and lipolysis indices of soft cheese made with calf rennet paste.

Cheese samples	Proteolysis index			Lipolysis index		
	TN/DM	SN/DM	SN/TN	TVFA *	FFA **	TBA ***
Fresh	3.25	2.11	64.92	74	1.97	0.485
CRP 1	3.13	2.03	64.85	76	9.87	0.805
CRP 2	3.07	1.88	61.23	78	11.56	0.824

CRP1= Calf rennet paste from *Al-fayum*, CRP 2= Calf rennet paste from *Mallawi*, SN= soluble nitrogen, TN= total nitrogen, DM= dry matter, * ml of 0.1 N Na OH / 100 g, ** FFA= free fatty acid, *** TBA= Thiobarbituric acid *** O. D. 532 nm

Table (7): Hygienic quality of soft white cheese made with calf rennet paste.

Microorganisms (CFU/g)	Cheese samples		
	Fresh	CRP 1	CRP 2
Total count	2.12 + 6	2.0 + 5	1.5 + 5
Yeast & Moulds	4.0 + 2	2.0 + 3	1.6 + 3
Enterococcus sp	absent	absent	absent
Coliform bacteria	absent	absent	absent
Staphylococcus aureus	absent	absent	absent

CRP 1= cheese sample with calf rennet paste from *Al-fayum*, CRP 2= cheese sample with calf rennet paste from *Mallawi*.

Table (8): Sensory evaluation of soft white cheese made with calf rennet paste.

Characteristics	Cheese samples		
	Fresh	CRP 1	CRP 2
Appearance (10)	8	9	9
Body & texture (30)	26	28	29
Flavor (60)	54	57	58
Total (100)	88	94	96

CRP 1= cheese sample with calf rennet paste from *Al-fayum*, CRP 2= cheese sample with calf rennet paste from *Mallawi*.

Patterns of electrophoresis were performed to monitor soft cheese and calf rennet pastes used in manufacturing are shown in Fig (1) and Table (9) . After 60 days of ripening. SDS-profiles of soft cheese samples could be divided into two zones. Zone A contains high molecular weight components (MW. 43.000-94.000), whereas zone B corresponds to 40 -14 kDa fractions (Fig 1). Zone B contains the most important milk proteins including α -, β -, κ -casein, β -Lg and α -La. The electrophoretic patterns for CRP1and CRP2 cheese samples (colum2, 3, 4, 5) seemed to be divided into eight to eleven regions including β - casein and α -casein. The intensity of the bands, either native or newly formed, mainly depended upon the ripening time. The α -casein and β - casein bands in the electrophoretograms of cheese are labelled according to Chianese *et al.* (1992), as indicated for the reference sample of bovine casein. On the whole, a decrease was observed throughout cheese ripening due to continuous proteolysis of these two casein. The intensity of the β - casein band was the highest in all cheese samples. The α_1 -I- casein band was present in all cheese samples.

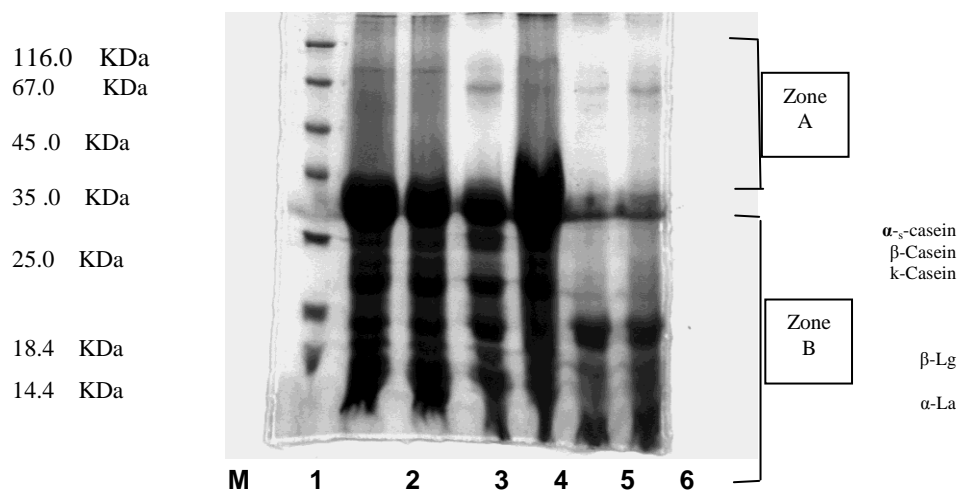


Figure 1: SDS - polyacrylamide gel electrophoresis and molecular weight determination of soft cheese and calf rennet pastes used in manufacturing. The electrophoretic analysis of the calf rennet paste (1, 2) and soft cheese CRP 1 (3, 4) and CRP 2 (5, 6).

Table (9): Determination of molecular weight (MW) of hydrolysed protein bands from rennet paste as well as respective ripened soft cheese by SDS- polyacrylamide gel electrophoresis.

Band No	Marker		CRP				Cheese samples			
			1		2		CRP 1		CRP 2	
	RF	Mol. Wt. KDa	RF	Mol. Wt. KDa	RF	Mol. Wt. KDa	RF	Mol. Wt. KDa	RF	Mol. Wt. KDa
1	0.055	116	0.12	79.486	0.126	79.486	0.128	79.486	0.13	79.486
2	0.152	66.2	0.385	33.708	0.393	32.833	0.176	64.698	0.172	64.698
3	0.263	45	0.537	24.787	0.532	24.787	0.212	57.121	0.206	57.121
4	0.366	35	0.579	23.064	0.582	23.064	0.416	31.164	0.413	31.164
5	0.528	25	0.617	21.747	0.618	21.747	0.529	24.787	0.429	30.052
6	0.705	18.4	0.697	18.784	0.702	18.593	0.588	23.064	0.469	27.435
7	0.817	14.4	0.743	16.976	0.744	16.976	0.62	21.747	0.595	21.747
8			0.773	15.742	0.794	14.967	0.691	19.141	0.647	19.709
9			0.872	11.923	0.874	11.923	0.742	16.976	0.731	16.595
10							0.798	14.967	0.802	14.353
11							0.878	11.923	0.872	11.157

CRP 1= cheese sample with calf rennet paste from *Al-fayum*, CRP 2= cheese sample with calf rennet paste from *Mallawi*.

Total bacterial count (TBC) of rennet paste and 12% salt soft cheese manufactured from heat treated milk during ripening period with calf rennet paste were illustrated in Tables (4 &7). From the data in Table (4) the CRP2

decreased TBC compared, with the CRP1. It is clear from these data that the count in control cheese (fresh cheese) was comparatively higher than cheese treatments. Moreover, lower numbers of TBC, were detected in cheese treated with CRP2, followed by that made with CRP1.

Yeasts and Moulds count in rennet paste and cheese were shown in Tables (4&7). From the results in Table (4) the CRP1 decreased the yeasts & moulds than CRP2. Yeasts and moulds continued to appear in fresh cheese. While the yeasts and moulds count decreased rapidly in all cheese samples during ripening period. This might be attributed to unsuitable anaerobic pickling conditions for yeasts and moulds growth. Increasing the amount of salt added to cheese milk resulted in a pronounced decrease in yeasts and moulds counts in resultant cheese. This probably due to the effect of high salt in preventing growth of yeasts and moulds (El-Sissi and Neamat-Allah, 1996).

Sensory evaluation of cheese are shown in Table (8). Addition of calf rennet paste remarkably enhanced the sensory quality attributes of flavour, intensity, body & texture and consequently the total acceptability of the resultant product. This might be attributed to the higher acidity being developed in cheese, which could enhance the texture of resultant cheese. Body & texture as well as flavour and consequently cheese acceptability improved during cheese ripening. The flavour enhancement in cheese with calf rennet paste is due to the role of enzyme to hydrolyze the milk component involved in cheese flavour such as proteins, fat, lactose, citrates and phosphates. Appearance of cheese showed no differences among all cheese samples. The fresh cheese sample was hard with less aromatic compound. Both soft cheeses CRP1& CRP2 samples were preferred to panelists due to its aroma, smooth soft body and homogenous texture as compared with the fresh sample.

REFERENCES

- Addis, M.; Piredda, G and Pirisi, A., (2008). The use of lamb rennet paste in traditional sheep milk cheese production. *Small Ruminant Research* 79: 2–10
- Addis, M.; Pirisi, A.; Di Salvo, R.; Podda, F., and Piredda, G.; (2005). The influence of the enzymatic composition of lamb rennet paste on some properties of experimentally produced PDO Fiore Sardo cheese. *Intern. Dairy J.* 15:1271–1278.
- Akin, N ; Aydemir, S.; Koçak, C. and Yildiz, M. A. (2003). Changes of free fatty acid contents and sensory properties of white pickled cheese during ripening. *Food Chem.* 80: 77–83.
- AOAC (2005). *Official Methods of Analysis. 18th Ed., Ch. 33,pp. 7-10-14.* Association of official Analytical Chemists, Benjamin Franklin Station Washington, D.C., USA.
- Arima, K.; Yu, J., and Iwasaki, S. (1970). In *Methods in Enzymology*, vol. 19, Perlmann, G. E., and Lorand, L., eds., Academic, NY, pp. 446-459.

- Basch, J.J.; Douglas, F.W.; Procino, L.G.; Holsinger, V.H. and Farrell, H.M.,(1985). Quantitation of caseins and whey proteins of processed milks and whey protein concentrates application of gel electrophoresis, and comparison with Harland-Ashworth procedure. *J. Dairy Sci.*, 68: 23-31.
- Barnett, A.J. and Tawab, G.A. (1975). Determination of lactose in milk and cheese. *J. Sci. Food Agric.* 8:437-441.
- Bustamante, M.A.; Chavarri, F.; Santisteban, A.; Ceballos, G.; Hernandez, I.; Miguelez, M.J.; Aramburu, I.; Barron, L.J.R.;Virto, M.; and Renobales, M., (2000). Coagulating and lipolytic activities of artisanal lamb rennet paste. *J. Dairy Res.* 67: 393–402.
- Bradly, R.L.; Arnold J.E. and Barbano, D.M. (1992). Chemical and physical methods. In: *Standard Methods for the Examination of Dairy Products (16th Ed)*, pp. 433-531. Robert, T. Marchal (ed.), American Public Health Association. Washington, DC, USA.
- Chianese, L.; Mauricillo, R.; Moio, L.; Intorcia, N., and Addeo, F. (1992). Determination of bovine casein heterogeneity using gel electrophoresis and immunochemical techniques. *J. Dairy Res.*, 59: 39-47.
- Collins, Y. F.; McSweeney, P. L. H. and Wilkinson, M. G. (2003). Lipolysis and free fatty acid catabolism in cheese: A review of current knowledge. *Intern. Dairy J.* 13:841- 866.
- Difco (1984). *Difco manual of Dehydrate Culture Media and Reagents*, Difco Laboratories, Detroit 1, Michigan, U.S.A.
- El-Koussy, L.A. (1966). *Studies on Soft Cheese Manufactured from Pasteurized Milk*. Ph.D. Thesis, Fa. Agric. Ain Shams Univ., Cairo, Egypt.
- El-Sissi, M.G. and Neamat-Allah, A.A. (1996). Effect of salting levels on ripening acceleration of Domiati cheese. *Egyptian J. Dairy Sci.*, 24:265-27.
- Fahmi, A.H. and Sharara.H.A. (1950). *Studies on Egyptian Domiati cheese*. *J. Dairy. Res.*, 17:312.
- Farrell, H. M.; Jimenez-Flores R.; Bleck, G.T.; Brown, E.M.; Butler, J. E; Creamer, L.K.; Hicks, C.L.; Hollar, C.M.; Ng-Kwai-Hang, K.F. and Swaisgood, H.E. (2004). Nomenclature of the proteins of cows' milk-sixth Revision. *J. Dairy Sci.*, 87: 1641-1674.
- Fox, P. F. (1993). *Cheese: An overview*. In P. F. Fox (Ed.), *Cheese: Chemistry, physics and microbiology*, Vol. 1 (pp. 1–36). London, UK: Chapman & Hall.
- Georgala, A.; Moschopoulou, E.; Aktypis, A.; Massouras, T.; Zoidou, E.; Kandarakis, I., and Anifantakis, E., (2005). Evolution of lipolysis during the ripening of traditional Feta cheese. *Food Chem.* 93: 73–80.
- Harrigan, W.F. and McCance, M. E. (1966). *Laboratory Methods in Microbiology*.Academic Press London and New York.
- Kosikowski, F.V. (1982). *Cheese and Fermented Milk Foods*. 2nd Ed.pp. 560-597.F.V. Kosikowski and Association, Brooktonal, New York.
- Laemmli, U. K., (1970). Cleavage of structural proteins during the assembly of the head of bacteriophage T4. *Nature*, 227: 680-685.
- Ling, E.R. (1963). *Text Book of Dairy Chemistry*. 3rd Vol .2Chapman and Hall Ltd., London.

- Pearson, D. (1977). The Chemical analysis of food. Chem. Puk. Comp. Inc., New York, U.S.A.
- Pettinau, M. G.; Nuvoli, F. P. and Ledda, A. (1977). Influenza del metodo di preparazione del caglio d'agnello sulla conservazione delle sue proprietà coagulanti, proteolitiche lipolitiche, (preparation methods and clotting, proteolytic and lipolytic activities preservation of lamb-rennet). Sci. Tech. Latt. Cas. 28:101-114.
- Piredda G. and Addis, M. (1998). Influenza dell'alimentazione sulle caratteristiche enzimatiche del caglio in pasta d'agnello, (diet influence on enzymatic activities in lamb rennet paste). Sci. Tech. Latt. Cas.49:129-138 .
- Santillo, A.; D'Angelo, F.;Caroprese, R.; Marino, R. and Albenzio, M. (2005). Influence of diet and of lamb slaughtering age on the coagulating properties of rennet paste. Ital. J. Anim. Sci. 2 : 336-338.

**تأثير عجينة منفحة العجول الرضية على الخواص الفيزيوكيميائية
والميكروبيولوجية للجبن الطرى الخزير المصنع من لبن جاموسى
زكريا محمد رزق حسن و عثمان عبد العظيم عيطة
قسم علوم الاغذية - كلية الزراعة - جامعة عين شمس - شبرا الخيمة - القاهرة - مصر**

يهدف هذا البحث الى دراسة تأثير عجينة منفحة العجل على الخواص الفيزيوكيميائية والميكروبيولوجية للجبن الطرى المصنع من لبن جاموسى والمخزن لمدة ٦٠ يوم. لذلك تم استخدام نوعين مختلفين من عجينة المنفحة وفحصت من حيث بعض الخواص مثل قوة المنفحة ووقت التجبن سواء كان اللبن منزوع الدسم أو كامل الدسم وسواء كان بدون ملح أو مضاف اليه ١٢% ملح الطعام كما تم تقدير التركيب الكيماوى. كما ان الجبن المصنع تم تحليله كيماويا سواء طازج أو بعد التسوية. و تم تقدير مقدار التحلل الدهنى عن طريق تقدير الاحماض الكلية الطيارة والاحماض الدهنية الحرة وقيمة TBA. وتم تقدير مقدار التحلل البروتينى عن طريق تقدير النتروجين الذائب والنتروجين الكلى. وتم تقدير الجودة الميكروبيولوجية للجبن الطرى المصنع من اللبن الجاموسى. وقد أوضحت النتائج أن وقت التجبن لعجينة المنفحة CRP2 كان أقل من المصنعة CRP1 مما يدل على تركيز المنفحة سواء كان اللبن منزوع الدسم أو كامل الدسم وسواء كان بدون ملح أو مضاف اليه ١٢% ملح الطعام. ولا يوجد اختلاف فى التركيب الكيماوى للجبن الطرى المصنع من لبن جاموسى من الرطوبة والبروتين ومحتوى الدهن والنتروجين الكلى. كما ان مقدار التحلل الدهنى للجبن الطرى المصنعة باستخدام عجينة المنفحة CRP 2 اكبر من المصنعة CRP1. كما ان الاحماض الدهنية الحرة فى الجبن المصنعة باستخدام عجينة المنفحة CRP 2 اكبر من المصنعة CRP1. كما تزداد الاحماض الكلية الطيارة بزيادة درجة التحلل. كما نجد ان الجبن الطازج تحتوى على قيمة منخفضة من TBA بالمقارنة بالجبن المسواه. لذلك نجد ان قيمة TBA مرتفعة فى الجبن المصنعة باستخدام عجينة المنفحة CRP2 عن المصنعة CRP1. ونجد من الفصل الكهربى خلال جل الاكربلاميد ظهور الببتيد α_s كازين فى كل عينات الجبن الطرى المصنعة باستخدام عجينة المنفحة CRP2 و CRP1.

من خلال التحكيم الحسى نجد حدوث تحسن فى الطعم والقوام والتركيب بزيادة فترة التسوية.

قام بتحكيم البحث

كلية الزراعة - جامعة المنصورة
كلية الزراعة - جامعة القاهرة

أ.د / طه عبد الحليم نصيب
أ.د / منير محمود العبد