

## Water Mobility in Accelerated Ripening of UF Feta Cheese

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### ABSTRACT

The aim of this study was to investigate the effect of the addition of commercial lipase in various forms on the acceleration of the ripening of UF-Feta cheese. Commercial lipase from *Aspergillus niger* was encapsulated by using silica composition based on Sol-Gel method. Lipase was then added to cheeses in three forms, namely, free lipase, encapsulated lipase, and encapsulated lipase with Arabic gum. Lipase was added to the retentate in the ratio of 4 g:100 kg. The effects of free lipase, encapsulated lipase, and encapsulated lipase with Arabic gum on lipolysis, quantity of free water, bounded water, and chemical compounds were studied during the 60 days of ripening. Based on FTIR analysis, encapsulated lipases were completely released from the capsules during the first 15 days of ripening period. The greatest amount of lipolysis was detected in free lipase samples, compared to encapsulated and encapsulated with Arabic gum treatments. Thermal analysis of all treatments indicated that lipolysis could affect the free and bounded water content by decreasing both of them from the 15<sup>th</sup> to 60<sup>th</sup> days of ripening. The amount of salt, moisture, and fat changed during the first 15 days of ripening significantly ( $P < 0.05$ ). In comparison with free or encapsulated lipase, the encapsulated lipase with Arabic gum caused more changes in types of water and chemical compositions because the produced emulsions were uniform.

**Keywords:** Bound water, Encapsulation, Free water, Lipolysis.

### INTRODUCTION

Sol-Gel process causes high catalytic activity for the encapsulated lipase. The formation of active lipase by removing the lid results in the mechanism of high catalytic activity. Hydrogen and ionic bonds sustain this condition. A group of hydrophobic amino acids are placed along the active site and cause an exposed active site called interfacial activation (Guisan, 2006). Cheese ripening means physical, chemical, biochemical, and microbiological changes in curd, which improves its flavor and texture. Biochemical changes in cheese during ripening are grouped into primary (lipolysis, proteolysis, and metabolism of residual lactose and of lactate and citrate) and secondary (metabolism of important compounds for flavor of cheese).

Increasing the components produced by the effects of enzyme that are involve in the cheese ripening reduces the ripening time. It has the inhibiting role on further enzyme activities (Fox *et al.*, 1993). The initial periods of lipolysis in cheese is related to lipases from milk, rennet paste, starter and microflora (Rasouli Pirouzian *et al.*, 2012 ). Pasteurization of milk at 72°C for 15 seconds decreases this activation (Driessen, 1989; Collins *et al.*, 2003). Lipase includes two types of hydrolytic and synthetic reactions, which are influenced by the amount of water. This amount of water produces the content of fatty acid and alcohol. Synthetic activity is intensified in the presence of water and substrate (Gillies *et al.*, 1987). Water in cheese is categorized into three types: water placed in serum canals, water nearly firmly bound to casein, and water firmly bound to casein and connected to protein

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molecule via hydrogen bond. Protein quantity influences the type and strength of the bound (Everett and Auty, 2008). Quantity and type of water (free and bonded) in food indicate its physical, microbial, and chemical changes (Duggan *et al.*, 2008). Quality and safety eventually determine its shelf life (Duggan *et al.*, 2008). Today, different methods are used to measure free and bounded water. Thermogravimetric analysis (TG) is a method used to measure the quantity of water in some cheeses (de Angelis Curtis *et al.*, 1999), and another method is Differential Scanning Calorimetry (DSC) that has been also executed for determination of different types of water in some other cheeses (McMahon *et al.*, 1999). Thermogravimetric method, which was used to measure water quantity in Cabrales cheese, indicated that correlation coefficient was high ( $R^2 = 94.5\%$ ) in oven method of dehydration, which takes too long time (Moro Garcia *et al.*, 1993).

Based on the importance of water in the food system which plays a fundamental role in enzyme activity and the ripening period, this research is the pioneering one which works on changes of different types of water on the UF-Feta cheese produced by novel technology of production. To control lipase activity, the specific method of Sol - Gel for encapsulation was used. The objective of the present study was to evaluate the effects of encapsulated lipase by using silica compositions on accelerated ripening, lipolysis, changes of free and bounded water by TG Analysis as a new method, and chemical compositions of UF-Feta cheese during the ripening period.

## MATERIALS AND METHODS

### Encapsulation of Lipase in Sol-Gel Matrices

All materials were obtained from Sigma-Aldrich (Saint Louis, USA), Fluka (Buchs, Switzerland), Acros Organic (New Jersey, USA) and Merck (Darmstadt, Germany). Lipase from *Aspergillus niger* with activity of

$\geq 120,000 \text{ IU g}^{-1}$  provided from Sigma-Aldrich.

Lyophilized powder of microbial lipase (150 mg), 390  $\mu\text{L}$  buffer of Tris/HCl (0.1M) with the pH 7.5 and 50 mg of Celite was mixed. Then, 100  $\mu\text{L}$  of 4% polyvinyl alcohol aqueous solution, 50  $\mu\text{L}$  of 1M sodium fluoride aqueous solution, 100  $\mu\text{L}$  of Isopropanol and 100  $\mu\text{L}$  of Arabic gum was added to the mixture and was homogenized by vortex shaker to enable encapsulated lipase to gather Arabic gum. 74  $\mu\text{L}$  of 2.5 mM Methyltrimethoxysilane (MTMS) and 76  $\mu\text{L}$  of 0.5 mM Tetramethoxysilane (TMOS) were added and the resultant mixture was blended for 10 to 15 seconds. At the early stages of the process, gelatinization was observed. Drying process continued over a night. The tube cover was then removed. In order to eliminate white solid compounds, 10-15 mL of Isopropanol was added to the tube. The obtained gel was washed by 10 mL of water, 10-15 mL of Isopropanol, 10 mL of n-pentane and distilled water. Afterwards, foregoing gel was scraped by a spatula. Stabilized lipase was dried at room temperature. Finally, two types of capsule were produced whose specifications were as follows: Encapsulated lipase, Encapsulated lipase plus Arabic gum (Reetz *et al.*, 2003; Guisan, 2006).

### Cheese Making

Raw cows' milk (300 kg) and the necessary equipment were provided by Pegah Dairy Company (Shiraz, Iran, factory capacity : 430 tons per day). The used starter cultures were *Lactococcus lactis* ssp. *Cremoris* and *lactis* (DM-230) and thermophilic *Lactobacillus delbrueckii* ssp. *Bulgaricus* and *Streptococcus thermophilus* (Y-502) (Danisco Deutschland GmbH, Alemanha, Germany). Rennet was obtained from *Rhizomucor miehei* of DSM Food Specialties (Seclin, France). UF-Feta cheese making was according to the method of Hesari *et al.* (2006). Simplified flowchart for the production UF-Feta Cheese is shown in

Figure 1. Cheese treatments are shown in the Table 1.

### FTIR (Fourier Transform Infrared Spectroscopy) Analysis

Cheese treatments were dried to moisture below 1% at 37°C in Memmert Vacuum Oven-200 (Memmert GmbH + Co.KG, Germany). Then, 20 mg of the treatment mixture was prepared in powder and was mixed with 400 mg of potassium bromate. Then, it was pressed into thin and transparent tablets. To determine the penetration percentage in treatments, the tablets were analyzed in FTIR (VECTOR 22, Bruker GmbH, Germany) at wavelengths from 400 to 4,000 cm<sup>-1</sup> (Belton *et al.*, 1987).

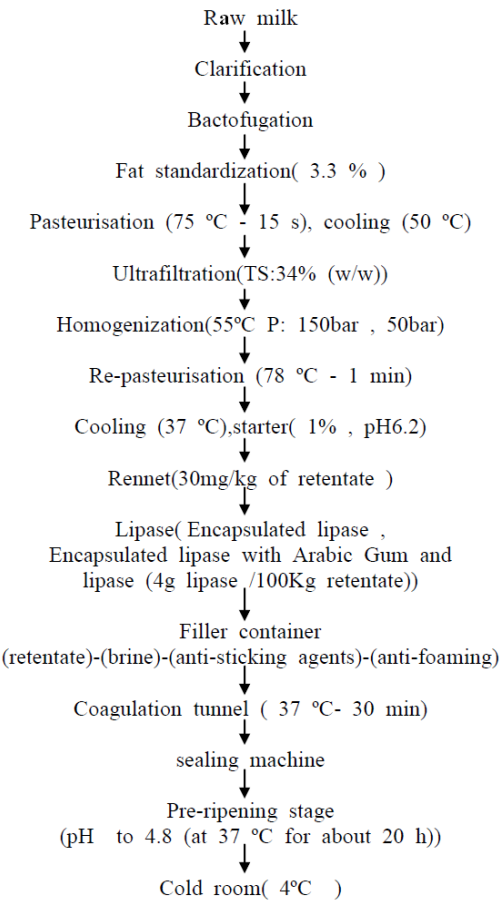
The treatments included lipase enzyme, encapsulated lipase, encapsulated lipase with Arabic gum as well as capsule ingredients, UF-Feta cheese as the control, UF-Feta cheese containing encapsulated lipase, encapsulated lipase with Arabic gum and free lipase.

### Acid Degree Value

The progress of lipolysis was evaluated with acid degree value based on Marshal's method (Marshal, 1992).

### Physicochemical Analysis

Total protein and fat were measured by Kjeldahl and Gerber method, respectively (AOAC, 1995). Salt content was also determined by Mohr method (AOAC, 2007). Cheese sample moisture content was analyzed by heating at 102 °C to reach a



**Figure 1.** Simplified flowchart for the production of UF- Feta cheese (Hesari *et al.*, 2006).

stable weight using a moisture meter (Sartorius MA35, Ltd., Epsom, UK) (IDF, 1982).

### TGA (Thermogravimetric Analysis)

TGA (TGA/DSC1 STARe (Mettler-Toledo GmbH Analytical, Switzerland)) was used to determine free water ( $\Delta W1$ ) and bound water ( $\Delta W2$ ,  $\Delta W3$ ). The cheese

**Table 1.** UF-Feta cheese treatments.

Cheese treatments	Code
UF-Feta cheese as control (no enzyme added)	(B)
UF-Feta cheese containing encapsulated lipase	(EN1)
UF-Feta cheese containing encapsulated lipase with Arabic gum	(EN2)
UF-Feta cheese containing free lipase	(F)



samples were placed at the room temperature ( $25 \pm 1$  °C). Then, an average of 16.5 mg of each treatment was put in special aluminum dishes of TGA and was set at temperature from 25 -180°C with 10°C intervals (Saldo *et al.*, 2002).

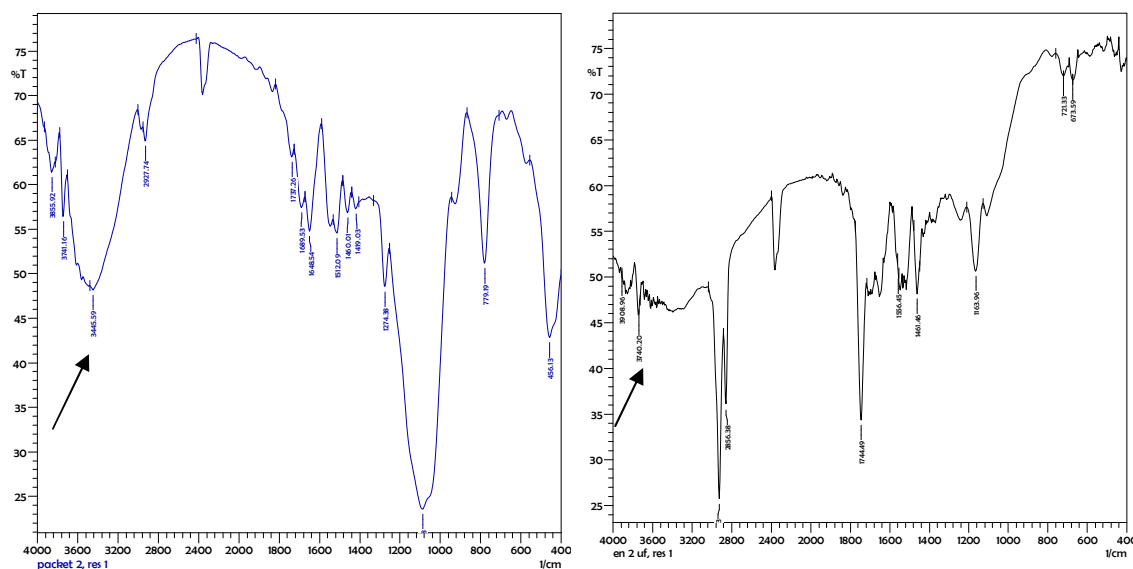
### Statistical Analysis

All measurements were performed at least in triplicate and mean values were reported. All statistical analyses were carried out using SPSS software (IBM® SPSS® Statistics 20, New York, USA). Analysis of variance (ANOVA) was performed. The least significant difference test ( $P < 0.05$ ) was used to examine the effects of type of lipase (free and encapsulated) and the ripening period (15, 30, 45, and 60 days) on the chemical components and types of water. All tables were drawn using Microsoft Excel 2007.

## RESULTS AND DISCUSSION

### FTIR Results

Functional groups of  $\text{NH}^+$  and  $\text{OH}^-$  were



**Figure 2.** FTIR spectra of encapsulated lipase on Feta cheese: (A) Before, and (B) After day 15; 400–4,000  $\text{cm}^{-1}$  range.

important. Results demonstrated that capsules got open during the 15 days. In UF-Feta cheese, encapsulated lipase with Arabic gum got open faster than other treatments. The opening of the capsule coincided with shrinkage and loss of the part related to functional groups of  $\text{NH}^+$ ,  $\text{OH}^-$  [the part is shown by arrow ( ) Figure 2]. The obtained results were in accordance with the other similar works (Belton *et al.*, 1987; Chen and Irudayaraj, 1998). The lipase was measured at the wavelength of 1,540-1,650  $\text{cm}^{-1}$ , silicon compounds at the wavelength of 110 and 810  $\text{cm}^{-1}$ , hydrogen bond at the wavelength of 3,400  $\text{cm}^{-1}$  and  $\text{NH}^+$  bond at the wavelength of 3,200-3,600  $\text{cm}^{-1}$ .

### Measurement of Lipolysis

The development of lipolysis on the treatments of UF-Feta cheese throughout the ripening is shown in Table 2. In this research, acid degree value was used to measure the development of lipolysis. There was a significant difference at the level of  $P < 0.05$  in acid degree value in treatments of UF-Feta cheese. During cheese ripening, the fat of the product was affected by lipase

**Table 2.** Changes in Acid degree value (ADV) (meq KOH 100 g fat<sup>-1</sup>), on the UF- Feta Cheese throughout ripening with different types of lipase.<sup>a</sup>

	Ripening period (day)	Treatments of UF-Feta cheese			
		B <sup>b</sup>	F <sup>c</sup>	En1 <sup>d</sup>	En2 <sup>e</sup>
ADV	1	1.3463±0.01 <sup>a</sup>	1.3463±0.01 <sup>a</sup>	1.3463±0.01 <sup>a</sup>	1.3463 ±0.01 <sup>a</sup>
	15	2.804±0.05 <sup>b</sup>	21.599±0.01 <sup>c</sup>	19.3544±0.01 <sup>d</sup>	8.977±0.01 <sup>e</sup>
	30	0.8416±0.01 <sup>f</sup>	10.660±0.01 <sup>g</sup>	1.4024±0.05 <sup>h</sup>	2.0197±0.01 <sup>i</sup>
	45	0.524±0.05 <sup>j</sup>	4.347±0.05 <sup>k</sup>	1.271±0.05 <sup>l</sup>	1.010±0.05 <sup>m</sup>
	60	30.293±0.01 <sup>n</sup>	1.123±0.01 <sup>o</sup>	1.1782±0.01 <sup>p</sup>	0.560±0.01 <sup>q</sup>

<sup>a</sup> Means with the (a-q) are significantly different (P< 0.05) type of lipase. Results are presented as average of data from three independent replicated Trials±Standard deviations.

<sup>b</sup> Control (no enzyme added); <sup>c</sup> Lipase; <sup>d</sup> Encapsulated lipase, and <sup>e</sup> Encapsulated lipase plus gum Arabic.

enzymes. Free fatty acids metabolism leads to producing flavor and smell. In all treatments, acid degree value increased from the first to 15<sup>th</sup> day of ripening and after the 15<sup>th</sup> day, it decreased. The highest amount of lipolysis was found in free lipase treatment, then, in encapsulated lipase and finally in encapsulated lipase enzyme with Arabic gum treatment. Free lipase created an intensifying lipolytic activity, but the activity of encapsulated lipase changed as a result of additives added to gel, which led to a difference in concentration of enzyme and its distribution in gel. Characteristics of covers such as size and thickness influenced the rate of activity (Fitzpatrick and Ahrné, 2005). The encapsulated lipases had less thickness than the encapsulated lipase with Arabic gum, and created a suitable gradual lipolytic activity in cheese, but the encapsulated lipases with Tetramethoxysilane and Methyltrimethoxysilane with Arabic gum produced gels with tiny pores, which limited substrate transition and created less lipolytic activity (Soares Cleide *et al.*, 2006). Lipolysis in cheese occurs by natural and microbial lipases. The activity of milk lipase was reduced by 83 to 100% due to pasteurization at 72–78°C for 15s. The basic factors of lipolysis in such cheeses were encapsulated lipases and starter enzymes. Probably, the increase in free fatty acids and salt concentration in curd had preventive effect on lipase activity and, thus, it had a

limiting impact on lipolysis (Pavia *et al.*, 2000). Irregular changes and decline in acid degree value was due to the conversion of fatty acids to compounds such as Methyl ketone (Madkor *et al.*, 1986). The cheese that is kept at low temperature has low acid degree value and lipolysis (Abd El-Salam *et al.*, 1993). The reported values of ADV in the current research are more than Teleme cheese made of cow milk (Mallatoua and Pappaa, 2003) and Urfa cheese made of cow milk (Ferit Atasoy and Turkoglu, 2009). Exposure and stabilization of lipase in the case in network caused a decline in lipase activity and by a decrease in pH, the active parts were directed toward whey proteins. Whey proteins inhibit lipase enzyme activity and reduce lipolysis (Geurts *et al.*, 2003). Physical status of milk fat during ripening (Carunchia Whetstine *et al.*, 2006) and absorption of casein and whey proteins on the surface of fat globules during cheese production limited lipolytic activity of lipase enzyme against fat substrate (Michalski *et al.*, 2001).

### Physicochemical Characteristics

The changes in protein, fat, moisture, and salt contents in UF-Feta cheese throughout the ripening are shown in Table 3. There were a significant (P< 0.05) difference between moisture (no significant differences in encapsulated lipase and encapsulated

**Table 3.** Changes in fat, moisture, salt and Protein on the Ultrafiltered Feta Cheeses throughout ripening with different types of lipase. <sup>a</sup>

Physicochemical characters	Ripening period (Day)	Treatments of UF-Feta cheese			
		B <sup>b</sup>	F <sup>c</sup>	En1 <sup>d</sup>	En2 <sup>e</sup>
Fat (%, w/w)	15	16.00±0.43 <sup>a</sup>	17.00±0.43 <sup>b</sup>	16.50±0.55 <sup>c</sup>	16.00±0.55 <sup>a</sup>
	30	17.50±1.06 <sup>d</sup>	16.00±1.06 <sup>e</sup>	16.00±1.06 <sup>e</sup>	15.50±0.78 <sup>f</sup>
	45	17.63±1.07 <sup>g</sup>	16.13±1.02 <sup>h</sup>	16.13±1.02 <sup>h</sup>	15.63±1.02 <sup>i</sup>
	60	17.38±0.68 <sup>j</sup>	15.88±1.02 <sup>k</sup>	15.88±1.02 <sup>k</sup>	15.37±1.02 <sup>l</sup>
Moisture (%, w/w)	15	64.59±0.89 <sup>a</sup>	63.29±1.39 <sup>b</sup>	65.18±1.39 <sup>c</sup>	63.17±1.33 <sup>d</sup>
	30	60.26±1.33 <sup>e</sup>	63.05±1.33 <sup>f</sup>	63.28±1.33 <sup>g</sup>	63.27±1.33 <sup>g</sup>
	45	60.34±1.33 <sup>h</sup>	63.13±1.33 <sup>i</sup>	63.36±1.39 <sup>j</sup>	63.35±1.39 <sup>j</sup>
	60	60.13±1.00 <sup>k</sup>	62.94±1.00 <sup>l</sup>	63.17±1.33 <sup>d</sup>	63.18±1.33 <sup>d</sup>
Salt (%, w/w)	15	4.44±0.59 <sup>a</sup>	5.26±0.50 <sup>b</sup>	4.68±0.50 <sup>c</sup>	4.91±0.50 <sup>d</sup>
	30	5.85±0.59 <sup>e</sup>	5.36±0.50 <sup>f</sup>	4.62±0.50 <sup>g</sup>	4.44±0.50 <sup>h</sup>
	45	5.86±0.59 <sup>e</sup>	5.37±0.59 <sup>f</sup>	4.63±0.59 <sup>g</sup>	4.45±0.59 <sup>h</sup>
	60	5.89±0.59 <sup>i</sup>	5.40±0.50 <sup>j</sup>	4.66±0.50 <sup>c</sup>	4.48±0.50 <sup>k</sup>
Protein (%, w/w)	15	11.39±1.00 <sup>a</sup>	11.65±1.10 <sup>b</sup>	10.71±1.10 <sup>c</sup>	11.73±1.10 <sup>d</sup>
	30	11.65±1.00 <sup>b</sup>	11.65±1.00 <sup>b</sup>	10.71±1.10 <sup>c</sup>	11.73±1.10 <sup>d</sup>
	45	11.46±1.01 <sup>e</sup>	11.46±1.00 <sup>f</sup>	10.52±1.10 <sup>g</sup>	11.54±1.10 <sup>h</sup>
	60	11.57±1.01 <sup>i</sup>	11.57±1.10 <sup>i</sup>	10.63±1.10 <sup>j</sup>	11.65±1.10 <sup>k</sup>

<sup>a</sup> Means with the (a-q) are significantly different (P< 0.05) type of lipase. Results are presented as average of data from three independent replicated Trials±Standard deviations.

<sup>b</sup> Control (no enzyme added); <sup>c</sup> Lipase; <sup>d</sup> Encapsulated lipase, and <sup>e</sup> Encapsulated lipase plus gum Arabic.

lipase with Arabic gum treatments on 45 and 60 days), protein (no significant differences in the control and free lipase treatments on 30 and 60 days), fat (free lipase and encapsulated lipase on 30, 45, and 60 days) and salt (no significant differences in all treatments on 30 and 45 days and encapsulated lipase treatment on 15 and 60 days) of treatments of UF-Feta.

Content of protein in all treatments decreased from 15 to 60 days. Encapsulated lipase with Arabic gum treatment had the highest protein content, which reached from 11.73%, w/w to 11.65%, w/w during ripening. During cheese ripening, proteins were converted into compounds such as water soluble nitrogen and amino acids, leading to an increase in cheese nitrogen. By adding lipase, there was a slight increase in quantity of soluble nitrogen, non-protein nitrogen (Peters and Knoop, 1974; 1975).

Similar results are reported by Omar *et al.* (1986) and Franco *et al.* (2003). Pezeshki *et al.* (2011) reported opposite results.

Content of fat in all treatments decreased from 15 to 60 days (except the control treatment which increased from 15 to 30 days and then decreased from 30 to 60 days). Free lipase treatment had the highest fat content that decreased from 17.00%, w/w to 15.88%, w/w, during ripening. Fat changed to free fatty acid and by protein hydrolysis, new bonds were created between protein networks (Karami *et al.*, 2009; 2008). Similar results were reported regarding Feta cheese (Georgal *et al.*, 2005). Similar results were reported concerning fat quantity in treatments of UF-Feta by Hagrass *et al.* (1983), Ates and Patir (2001), Yilmaz *et al.* (2005) and Karami *et al.* (2008).

There was a high moisture content in treatments of UF-Feta cheese that contained encapsulated lipase (moisture decreased during 15 to 60 days from 65.18 to 63.17%, w/w) and encapsulated lipase with Arabic gum (reaching 63.17%, w/w during ripening). Oliveira and Dourado (2011) study on Green Edam cheese revealed opposing result. During cheese ripening, salt content increased in all treatments from 15 to 60 days. From the first day of ripening to the 15<sup>th</sup> day, the decrease in curd moisture was triggered by two main factors: dehydration of curd, which was due to the emission and transmission of salt in the curd, after the first 15 days, lipolysis decreased by an increase in interactions of salt inside the curd. Adding lipase did not have any effect on the salt of cheeses (Hagrass *et al.*, 1983). Similar results were reported by Pastorino *et al.* (2003).

### Changes of Water Types

According to research by de Angelis

Curtis *et al.* (1999) and Saldo *et al.* (2002) on the basis of TGA curve, there were three stages of weight loss: first stage of free water ( $\Delta W1$ ), second stage of nearly firmly-bonded water ( $\Delta W2$ ), and third stage of firmly-bonded water ( $\Delta W3$ ) (Figure 3). The changes in the water types of UF-Feta cheese throughout the ripening are shown in Table 4. There was a significant difference between free water ( $\Delta W1$ ) and bounded water ( $\Delta W2$ ,  $\Delta W3$ ) in all treatments of UF-Feta cheese. Free and bounded water were influenced by free lipase and encapsulate lipase and decreased during ripening. The highest decrease of free water ( $\Delta W1$ ) and bounded water ( $\Delta W2$  and  $\Delta W3$ ) during ripening were 5%, w/w in the control and encapsulated lipase with Arabic gum, 26%, w/w in encapsulated lipase with Arabic gum, and 25%, w/w in free lipase treatments.

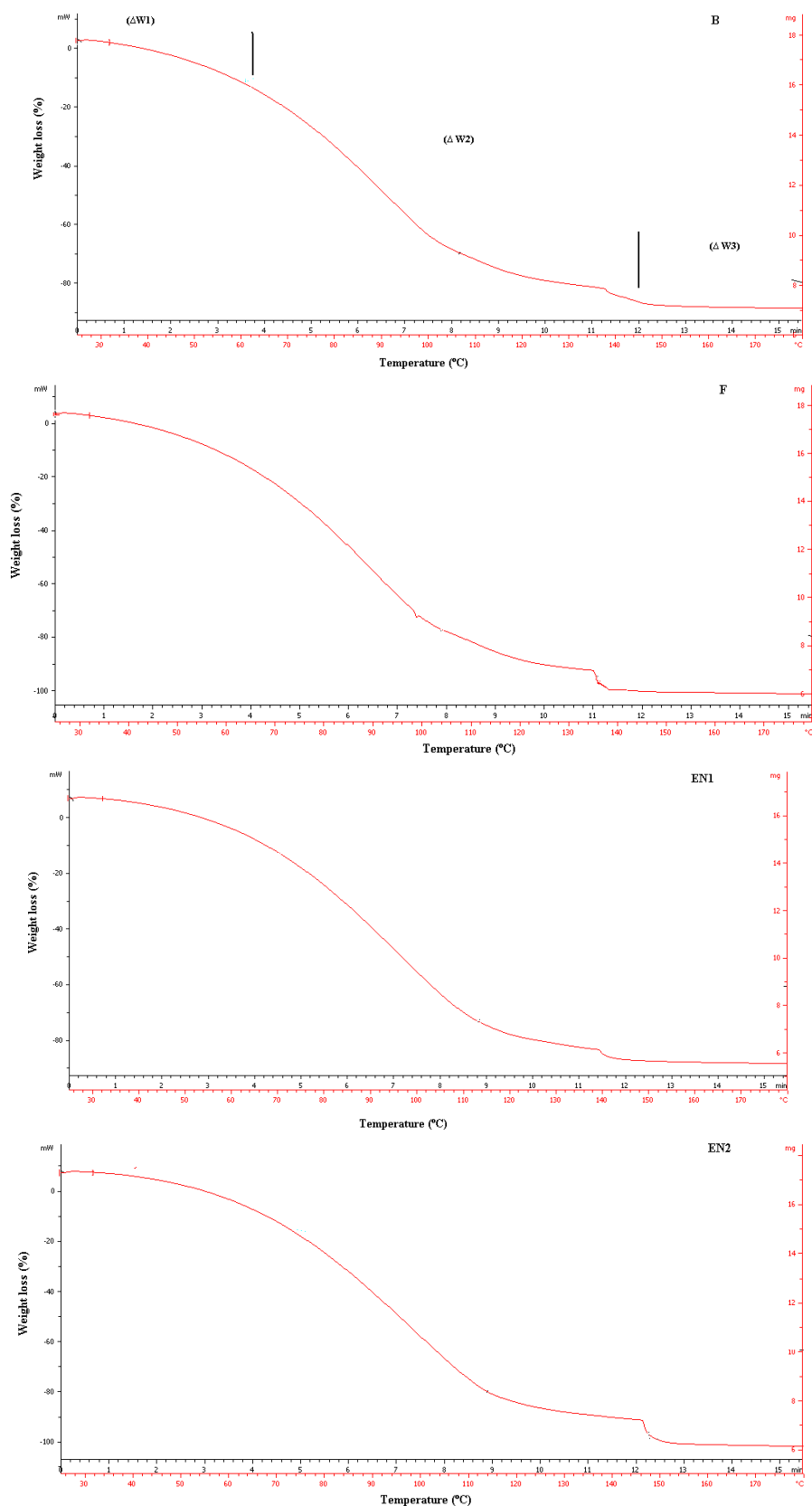
During ripening, fat was affected by gradual and selective lipolysis in the presence of encapsulated lipase. Types of water in the first ripening days had a direct relation to lipolysis products (fatty acid, glycerol, and carboxyl). However, by an

**Table 4.** Changes in free and bound water on the UF-Feta cheese throughout ripening with different types of lipase.<sup>a</sup>

Type of water	Ripening period (Day)	Treatments of UF-Feta cheese			
		B	F	En1	En2
Free water ( $\Delta W1$ ) (%, w/w)	15	90 ± 1.00 <sup>a</sup>	92 ± 0.57 <sup>b</sup>	92 ± 0.50 <sup>c</sup>	95 ± 1.01 <sup>d</sup>
	30	90 ± 1.00 <sup>c</sup>	92 ± 0.50 <sup>f</sup>	90 ± 0.57 <sup>g</sup>	95 ± 0.57 <sup>h</sup>
	45	92 ± 1.00 <sup>i</sup>	90 ± 1.00 <sup>j</sup>	91 ± 0.57 <sup>k</sup>	88 ± 1.01 <sup>l</sup>
	60	85 ± 0.57 <sup>m</sup>	88 ± 0.57 <sup>n</sup>	90 ± 1.01 <sup>o</sup>	90 ± 0.57 <sup>p</sup>
Bound water ( $\Delta W2$ ) (%, w/w)	15	28 ± 1.00 <sup>a</sup>	38 ± 1.00 <sup>b</sup>	40 ± 0.5 <sup>c</sup>	36 ± 0.57 <sup>d</sup>
	30	29 ± 0.54 <sup>e</sup>	30 ± 1.00 <sup>f</sup>	38 ± 1.00 <sup>g</sup>	28 ± 1.00 <sup>h</sup>
	45	28 ± 0.54 <sup>i</sup>	20 ± 0.54 <sup>j</sup>	19 ± 1.00 <sup>k</sup>	16 ± 1.00 <sup>l</sup>
	60	10 ± 1.00 <sup>m</sup>	10 ± 0.54 <sup>n</sup>	20 ± 1.00 <sup>o</sup>	10 ± 1.00 <sup>p</sup>
Bound water ( $\Delta W3$ ) (%, w/w)	15	20 ± 0.54 <sup>a</sup>	27 ± 1.00 <sup>b</sup>	30 ± 0.95 <sup>c</sup>	20 ± 1.00 <sup>d</sup>
	30	20 ± 1.00 <sup>e</sup>	17 ± 1.00 <sup>f</sup>	30 ± 0.95 <sup>g</sup>	14 ± 0.95 <sup>h</sup>
	45	20 ± 1.00 <sup>i</sup>	9 ± 1.00 <sup>j</sup>	10 ± 1.00 <sup>k</sup>	11 ± 1.00 <sup>l</sup>
	60	2 ± 1.00 <sup>m</sup>	2 ± 1.15 <sup>n</sup>	9 ± 1.00 <sup>o</sup>	2 ± 1.10 <sup>p</sup>

<sup>a</sup> Means with the (a-q) are significantly different ( $P < 0.05$ ) type of lipase. Results are presented as average of data from three independent replicated Trials ± Standard deviations.

<sup>b</sup> B: Control (no enzyme added); F: Lipase; EN1: Encapsulated lipase, and EN2: Encapsulated lipase plus gum Arabic.



**Figure 3.** TGA curve of the UF-Feta cheese Treatments heated at a rate of 10°C min. (B) Control (no enzyme added); (F) Lipase; (EN1) Encapsulated lipase, (EN2) Encapsulated lipase plus gum Arabic .



increase in concentration of lipolysis products during ripening (Lemay *et al.*, 1994), water activity had a positive relation to moisture and negative relation with salt (Marcos *et al.*, 1983; Mangia *et al.*, 2011). Bounded water was put under the effect of lipolysis and proteolysis products (Saldo *et al.*, 2002) and was influenced by free water through a series of reactions; however, it decreases during ripening (McMahon *et al.*, 1999). Similar findings were reported by Saldo *et al.* (2002), Zamora *et al.* (2011), and Saurel *et al.* (2004). Working on Emmental and Graviera cheese, Saurel *et al.* (2004) reported that the decrease in free water would increase the amount of nitrogen products soluble in water. Therefore, pH increased (Haas and Spillmann, 2001). Capability of whey proteins in retention of water was affected by the ionic bonds which it formed in gel substructure (Barbut, 1995). By raising the quantity of protein in the total solid, the cheese appeared drier (Lemay *et al.*, 1994). Similar results were reported by Hickey *et al.* (2013), while Saldo *et al.* (2002) reported inconsistent results regarding free water. As a result of homogenization, the interactions in rennet curd occurred on  $\alpha$ s-casein via calcium bonding and  $\beta$ -casein via calcium bonding and hydrogen and whey protein,  $\alpha$ -lactalbumin and  $\beta$ -lactoglobulin via hydrogen bonding and hydrophobic interactions in combination with water. Based on protein denaturation,  $\beta$ -lactoglobulin was influenced by the quantity of its dehydration at different degrees (Kneifel and Seiler, 1993).

## CONCLUSIONS

This study is the first attempt to investigate the effects of encapsulated lipases on development of lipolysis as well as changes of free and bonded water in UF-Feta cheese. In this research, it was shown that in the first 15 days of the 60-day ripening period, high acid degree value caused a wide lipolysis. Encapsulated

lipases gradually created lipolytic activity. Changes in chemical compositions during ripening affected lipolysis. Lipolysis and changes in the types of water were in contrast with salt, the types of water at the beginning of ripening, and had a direct relation with fat, furthermore, it was the opposite in relation to fat. Free and bounded water decreased during ripening. The greatest decline of water in treatments of encapsulated lipase with Arabic gum was because of absorption of moisture. Therefore, the rate of water accessibility in cheese decreased and the enzyme access to substrate and enzyme activity (lipolysis) decreased. At the end, we found out that regarding phenomena such as proteolysis, the extent of lipolysis was limited. But, this process has a significant role in the changes of cheese texture. In fact, lipolysis is a limited process in many cheeses including the Iranian cheeses, especially Feta cheese. Production by ultrafiltration has more limitation in this regard because whey proteins play a restricting role. The changes of free fatty acids could be the subject of further work, particularly in evaluation of unsaturated fatty acids and their related products and their role in the texture and especially flavor of the final product.

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## مطالعه جابجایی آب در تسریع رسیدن پنیر فتای فراپالایش

ص. یزدان پناه، م. ر. احسانی و م. میزانی

### چکیده

هدف از این مطالعه بررسی تاثیر اضافه کردن لیپاز تجاری در شکل های متفاوت بر تسریع رسیدن پنیر فتای فراپالایش است. لیپاز تجاری بدست آمده از آسپرژیلوس نایجر با استفاده از ترکیبات سلیکایی بر اساس روش سول - ژل اینکپسوله شد. سپس لیپاز در سه شکل لیپاز آزاد، لیپاز اینکپسوله و لیپاز اینکپسوله به همراه صمغ عربی به پنیرها اضافه شد. لیپاز به نسبت ۴ گرم : ۱۰۰ کیلوگرم رتنتیت اضافه شد. تاثیرات لیپاز آزاد، لیپاز اینکپسوله و لیپاز اینکپسوله به همراه صمغ عربی بر لیپولیز، مقدار آب آزاد، آب باندشده و ترکیبات شیمیایی در طی دوره رسیدن ۶۰ روزه مورد مطالعه قرار گرفت. بر اساس آزمون FTIR، لیپازهای اینکپسوله در ۱۵ روز اول از دوره رسیدن به طور کامل از کپسول ها آزاد شدند. بیشترین مقدار لیپولیز در نمونه لیپاز آزاد در مقایسه با لیپاز اینکپسوله و لیپاز اینکپسوله به همراه صمغ عربی تعیین گردید. آزمون حرارتی نشان داد که لیپولیز بر آب آزاد و باند شده تاثیر داشته و باعث کاهش هر دو از روز ۱۵ تا روز ۶۰، رسیدن شد. مقادیر نمک، رطوبت و چربی در ۱۵ روز اول از رسیدن تغییر معنی داری ( $P < 0.05$ ) داشت. لیپاز اینکپسوله به همراه صمغ عربی در مقایسه با لیپاز آزاد و لیپاز اینکپسوله باعث تغییر بیشتری بر انواع آب و ترکیبات شیمیایی شد زیرا امولسیون های هم شکل تولید کرده است.