RHEOLOGICAL PROPERTIES OF MOZZARELLA CHEESE DETERMINED BY CREEP/RECOVERY TESTS: EFFECT OF SAMPLING DIRECTION, TEST TEMPERATURE AND RIPENING TIME

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ABSTRACT

The viscoelastic properties of mozzarella cheese using a creep/recovery test considering different sampling directions (parallel and perpendicular to protein fiber orientation), test temperatures (20, 30 and 40C) and ripening times (1, 8, 15, 29 and 36 days) were studied. Creep data were interpreted by a Burger model of four parameters. A semiempirical approach was proposed to obtain the contribution of each main compliance to the total deformation of the system. Creep tests at different temperatures allowed gaining a better understanding of changes that occur in the cheese matrix during heating and ripening. Sampling direction did not affect any of the parameters studied. Finally, it was clearly observed that cheese matrix behaves as a quite different physicochemical system depending on temperature. Therefore, it is recommended to carry out the rheological tests at different temperatures to evaluate appropriately the viscoelastic properties of mozzarella cheese.

PRACTICAL APPLICATIONS

Mozzarella cheese must have certain characteristics to be used on pizzas and on other prepared foods that use the cheese in melted state. The protein chains in the mozzarella curds coalesce into large strands that are oriented in the direction of stretching. For this reason, mozzarella cheese has an anisotropic structure. Therefore, it is relevant to determine the effect of protein

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fiber orientation on the rheological properties. Valuable information may be obtained through the creep/recovery test of mozzarella cheese samples to study its rheological properties and to explain molecular mechanisms that occur during ripening or melting processes considering sampling direction.

KEYWORDS

Creep/recovery test, mozzarella cheese, rheology, viscoelasticity

INTRODUCTION

Mozzarella cheese has a special structural characteristic because the curd is subjected to a stretching process in hot water during manufacturing that promotes the formation of a fibrous structure (Lucey *et al.* 2003). The protein chains in the mozzarella curds coalesce into large strands that are oriented in the direction of stretching. For this reason, mozzarella cheese has an anisotropic structure (McMahon *et al.* 1999, 2005). Mozzarella cheese must have certain characteristics to be used on pizzas and on other prepared foods that use the cheese in melted state. In this case, rheological methods can be used as a quality control tool because the rheological parameters may be closely related to the desirable functional properties of mozzarella cheese.

Cheese is a viscoelastic material, and it can be regarded as a dispersed phase of fat globules embedded in a continuous protein matrix (Subramanian *et al.* 2006). During aging, several biochemical events are produced and structural characteristics undergo notable changes. It is well known that the number, strength and type of bonds between protein molecules (especially caseins) constitute the basis of the rheological properties of cheese (Lucey *et al.* 2003). The spatial arrangement of these bonds also influences the rheological properties; however, there is little knowledge on this aspect of mozzarella cheese.

Dynamic small amplitude oscillatory shear analysis is one of the most common rheological methods for determining viscoelastic properties of cheese (Subramanian and Gunasekaran 1997a,b; Gunasekaran and Ak 2003; Singh *et al.* 2006; Subramanian *et al.* 2006). Creep/recovery method is one of the fundamental transient tests used to characterize structural properties of viscoelastic materials. During a creep experiment, an instantaneous and constant shear stress τ_o is applied (in the region of linear viscoelasticity of the material), and a deformation γ is measured as function of time (creep time). A creep/recovery experiment also considered the measurements when the stress is removed over a likewise preestablished period of time (recovery time)

(Steffe 1996). Creep data are usually expressed in terms of a parameter called creep compliance *J*(*t*).

Typical creep data can be interpreted using mechanical models with different numbers of parameters, which are very useful for explaining time scale of molecular mechanisms contributing to the viscoelastic response (Subramanian *et al.* 2003). Although the valuable information may be obtained through the creep/recovery test, there is scarce information related to the use of this test in rheological studies of mozzarella cheese and its potential to explain molecular mechanisms that occur during ripening or melting processes. The objective of this work was to study the viscoelastic properties of mozzarella cheese using a creep/recovery test considering different sampling directions, test temperatures and ripening times.

MATERIALS AND METHODS

Cheese Sampling and Treatments

Two blocks of fresh mozzarella cheeses ($28 \times 10 \times 10$ cm^3 in size) were provided by a local factory. Cheese manufacture was carried out according to the Código Alimentario Argentino specifications (CAA 2007). The vacuum-packed cheeses were stored at 4C and sampled at different days of ripening. At each ripening time and from each cheese block, a slab of 3-cm thickness ($3 \times 10 \times 10$ cm^3 in size) was cut perpendicularly to the principal axis of the cheese (Fig. 1). Each slab was divided into two equal parts ($3 \times 5 \times 10$ cm^3 in size). One half was used to determine pH and contents of chloride, moisture, total nitrogen (TN) and soluble nitrogen in water at pH 4.6. The other half was cut parallel to the principal cheese axis in two equal parts as shown in Fig. 1, which were used to obtain samples with different protein fiber orientation for the rheological tests. Samples were cut parallel (L) or perpendicular (T) to the principal axis of the cheese block. The samples T were analyzed at 1, 8, 15, 29 and 36 days of ripening. The rheological tests using the samples L were carried out the following day of each ripening time.

Chemical Analysis

The pH was determined with an electrode for solid foods (pH spear, Oakton Instruments, Vernon Hills, IL). Chloride content was determined as suggested by Fox (1963) with an automatic titrator model DL40 RC (Mettler Instrumente AG, Greifensee, Switzerland). Moisture content was determined as suggested by AOAC (1990) in a microwave CEM AVC 80 (CEM, Mattheus, NC). Water-soluble fraction extraction at pH 4.6 was performed with a procedure developed by Kuchroo and Fox (1982) and modified by Sihufe *et al.*

(2003). TN and water-soluble nitrogen (WSN) were determined using the micro-Kjeldahl method with an automatic digestor model 430, a distillation unit model 322 and a control unit model 342 (Büchi Labortechnick AG, 1998, Flawil, Switzerland). Fat content was determined for initial composition (International Dairy Federation 1969). Chemical analyses were carried out in duplicate. Maturation index was calculated as a percentage of WSN of the cheese TN (WSN × 100/TN), and it was used to follow the proteolysis degree during ripening (Kuchroo and Fox 1982).

Rheological Measurements

Cheese slices (approximately 1.3 mm thickness) were cut L or T to the principal axis of the cheese block (Fig. 1). The first three slices were discarded, and disks (20 mm diameter) were cut using a borer. Disks were obtained by discarding the external region of approximately 10 mm from cheese surface and wrapped with plastic film to prevent dehydration. A rheometer Haake RheoStress RS80 (Haake Instrument Inc., Paramus, NJ) with parallel plates (20 mm diameter, 1 mm gap) was used for rheological measurements. The disk-shaped cheese sample was placed on the lower plate, and then the upper plate was brought in contact with the sample during 2 min to allow sample

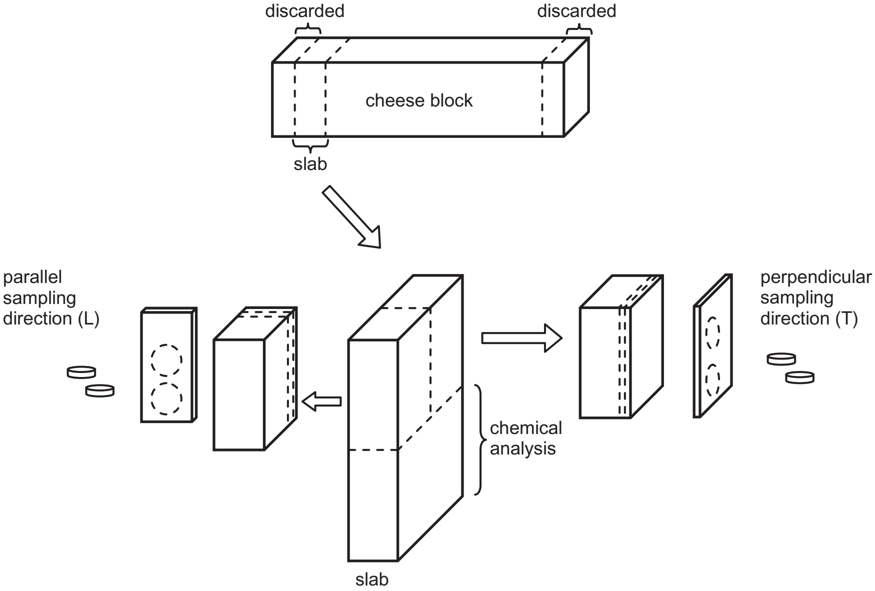

FIG. 1. SCHEMATIC VIEW OF CHEESE SAMPLING

relaxation and attain temperature equilibrium. The temperature of the lower plate was maintained constant by circulating water from a water bath.

Creep tests were carried out at 20, 30 and 40C. A constant shear stress ($\tau = 25$ Pa) was applied and the resultant strain (γ) was measured as a function of time. The results were expressed in terms of creep compliance $J(t) = \gamma(t)/\tau_o$. After 180 s, the stress was removed and sample was allowed to recover for 500 s. Linear viscoelasticity region was determined by performing creep tests at 10–100 Pa for the temperatures and ripening times analyzed. Two samples from each treatment were assayed.

Statistical Analysis

Data were analyzed by analysis of variance (ANOVA) using Statgraphics (Statgraphics Inc., Rockville, MD). When differences between treatment effects were significant ($P < 0.05$), a multiple comparison of means was performed using the least significant differences test.

RESULTS AND DISCUSSION

Chemical Characteristics

The initial fat content was 14.46 ± 1.80%. Chemical composition of mozzarella cheese at the studied ripening times is shown in Table 1. Expected values of moisture, total protein, and chloride contents and of pH were observed (Yun *et al.* 1993a,b; Guo and Kindstedt 1995; Bertola *et al.* 1996; Subramanian and Gunasekaran 1997a; McMahon *et al.* 1999; Muthukumarappan *et al.* 1999; Guinee *et al.* 2001). Maturation index increases during the ripening period studied, approximately from 3.2 to 6.0% (Fig. 2), possibly because of the proteolysis of α_{s1} and β-casein caused by the residual coagulant and plasmin (Fox and Guinee 1987; Farkye *et al.* 1991). The low level of proteolysis may be explained by the partial thermal inactivation of the

TABLE 1.
AVERAGE CHEMICAL COMPOSITION OF CHEESE SAMPLES

Ripening time (days)	pH	Moisture (%)	Total protein (%)	NaCl (%)
1	5.30	47.84	23.46	0.93
8	5.38	47.52	23.35	0.84
15	5.32	47.20	23.45	1.16
29	5.32	47.15	23.57	1.28
36	5.29	47.15	23.37	1.27

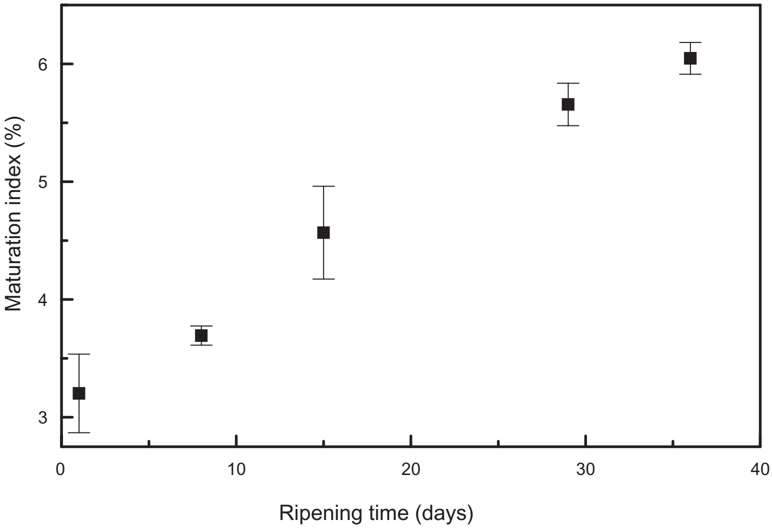

FIG. 2. MATURATION INDEX OF MOZZARELLA CHEESE CHANGING WITH RIPENING TIME
Bars indicate standard deviation.

coagulant during kneading and stretching process (Yun *et al.* 1993a; Bertola *et al.* 1996; Guinee *et al.* 2000; Feeney *et al.* 2001).

Rheological Characteristics

It was verified that the shear stress of 25 Pa ensured that the rheological tests be carried out in the linear viscoelastic region. The instantaneous creep and recovery compliance, $J(t)$, at different temperatures is shown in Fig. 3. It is observed that mozzarella cheese samples show viscoelastic behavior, and the creep and recovery compliances increase with temperature because of thermal softening. This behavior was observed at all ripening times evaluated, which is in agreement with results reported in the literature (Subramanian *et al.* 2003). There was no full recovery even after 500 s for all cases studied.

Figure 4 shows creep and recovery compliance at different ripening times. It is observed that the maximum compliance reached by the material before the constant stress was removed (J_{max}) increases as cheese ages (Brown *et al.* 2003) because of proteolysis. Moreover, the residual compliance value at the end of the recovery period (J_{res}) at the beginning of ripening was lower than the value reached after approximately the first 10 days of ripening.

Compliance data during creep test were fitted by a four-component Burger model (Steffe 1996),

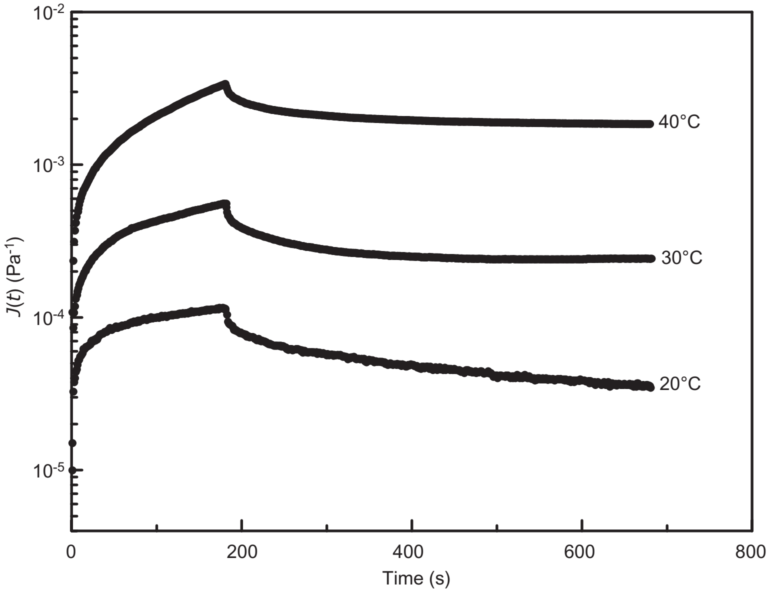

FIG. 3. TYPICAL CREEP/RECOVERY CURVES OF SAMPLES T OF MOZZARELLA CHEESE RIPENED DURING 36 DAYS EVALUATED AT DIFFERENT TEST TEMPERATURES

$$J(t) = J_o + J_1\left(1 - e^{-t/\tau}\right) + \frac{t}{\eta_N} \tag{1}$$

where J_o is the instantaneous elastic compliance (Pa^{-1}) of the Maxwell spring, J_1 is the retarded compliance (Pa^{-1}) that represents the retarded elastic region related to the Kelvin–Voigt element, τ is the retardation time (s) associated with the Kelvin–Voigt element and η_N is the Newtonian viscosity (Pa·s) associated with the Maxwell dashpot.

Figure 5 shows an example of the experimental creep compliance data and the fitted values obtained, indicating that the Burger model of four elements is adequate to describe mozzarella cheese rheological behavior. For all cases analyzed, fitting error was less than 0.3%. Similar results were obtained by Ma *et al.* (1996, 1997), Kuo *et al.* (2000), Messens *et al.* (2000) and San Martín-González *et al.* (2007), although in some cases, more than one Kelvin–Voigt element was used in the mechanical models.

The rheological parameters of the Burger model for samples T and L of mozzarella cheese are listed in Tables 2 and 3, respectively. Both samples T and L showed approximately a similar behavior. The instantaneous elastic compliance (J_o) represents the value of instantaneous shear creep compliance at initial

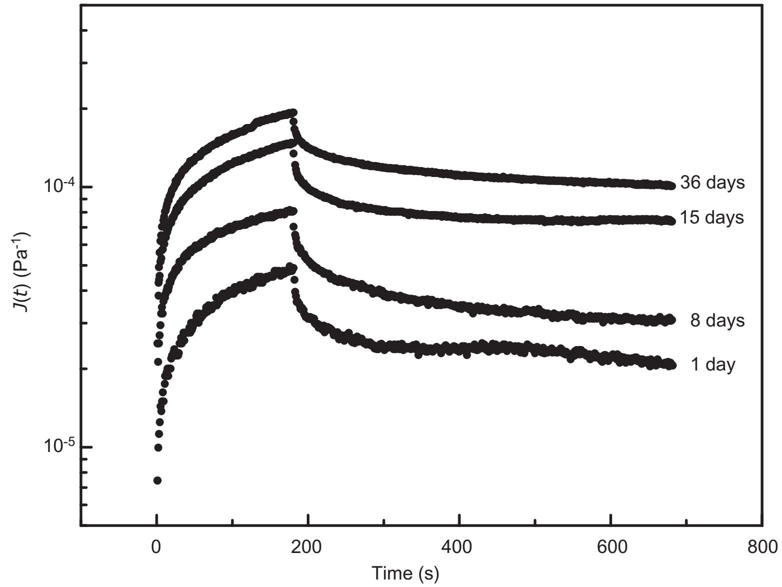

FIG. 4. TYPICAL CREEP/RECOVERY CURVES OF SAMPLES L OF MOZZARELLA CHEESE RIPENED DURING DIFFERENT TIMES EVALUATED AT 20C

time, and it may be related to the undisturbed protein network structure (Ma *et al.* 1997; Subramanian *et al.* 2003). A higher value of J_o reflects a higher degree of nonretarded Hookean-type (elastic) deformation, indicating that the polypeptide strands in the network are relatively free to rearrange between cross-links (Ma *et al.* 1996, 1997; Subramanian *et al.* 2003). In this case, J_o increased with temperature (because of thermal softening) and maturation (because of proteolysis), in agreement with the results obtained by Subramanian *et al.* (2003). The increasing J_o with ripening time indicates that the cheese becomes less rigid. It is worth mentioning that J_o showed significant differences with ripening time only when the test temperature was 30 or 40C.

According to Subramanian *et al.* (2003), the retarded compliance (J_1) represents the principal component of the viscoelastic behavior of mozzarella cheese. It was observed that J_1 increased with temperature and maturation (Tables 2 and 3). The increase of this parameter is associated with a less solid and more viscoelastic behavior. Similar to J_o, significant differences with ripening time were found only when the test temperature was 30 or 40C. Similar results were reported by Venugopal and Muthukumarappan (2003) as cited by Subramanian *et al.* (2006), who studied the dynamic rheological properties of cheddar cheese with different fat and moisture levels for up to 24 weeks of aging through oscillatory tests during heating and cooling (25 to 60C

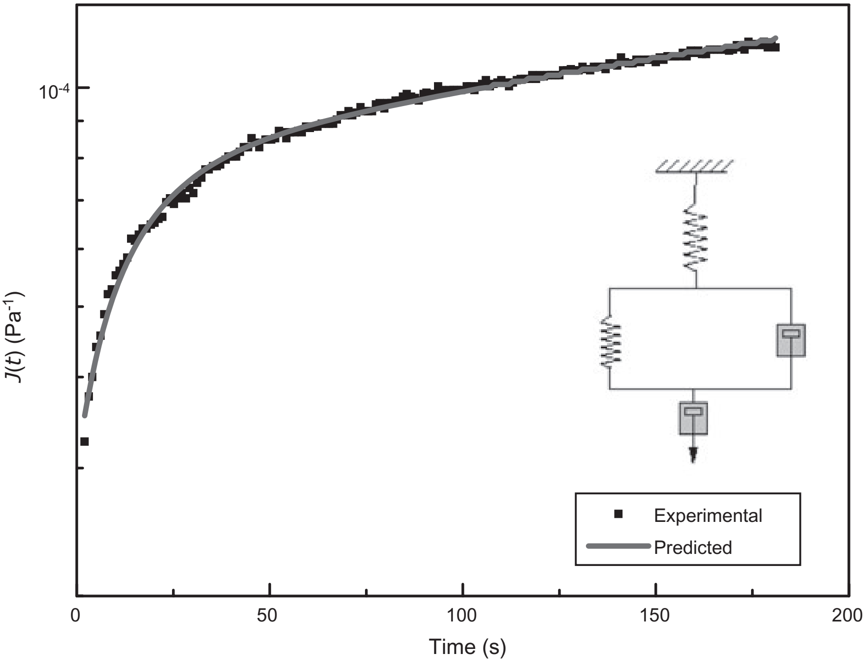

FIG. 5. PREDICTION OF CREEP COMPLIANCE OF MOZZARELLA CHEESE BY THE BURGER MODEL

Results correspond to a mozzarella cheese sample ripened during 36 days and evaluated at 20C.

and back). They found that proteolysis during ripening of cheese led to a decrease in the storage and loss modulus at higher temperatures, but the differences were not significant at lower temperatures.

The understanding of physicochemical changes during heating may help to explain the behavior of J_o and J_1. Lucey *et al.* (2003) reported an interesting analysis explaining that part of the initial softening of cheese at temperature $\leq$40C is related to the fat melting, but the major overall effects are related to protein interactions (principally casein–casein interactions). Protein matrix is stabilized by different interaction energies. Hydrophobic interactions tend to increase in strength with temperature, which may reduce the size of the contact area between casein particles; the net result is a reduction in overall gel strength. Also, various types of electrostatic interactions are likely to be important in cheese (charge repulsion between similar charges on proteins, plus minus charge interactions, salt bridges, colloidal calcium phosphate bridges). Even more, hydrogen-bonding (attractive) interactions play an important role in the stabilization of protein matrix. Hydrogen bonding

TABLE 2.
CREEP COMPLIANCE PARAMETERS FOR SAMPLES OF MOZZARELLA CHEESE CUT PERPENDICULAR TO THE PRINCIPAL AXIS (T) AT DIFFERENT TEMPERATURES AND RIPENING TIMES*

Temperature (C)	Ripening time (days)	J_o (10^{-5}/Pa)	J_1 (10^{-5}/Pa)	τ (s)	η_N (10^5 Pa·s)
20	1	0.68^{a}	5.52^{ab}	28.24^{ef}	29.08^{cde}
	8	2.65^{ab}	7.04^{ab}	26.47^{def}	27.40^{cd}
	15	2.62^{ab}	4.68^{a}	16.45^{abcd}	42.22^{ef}
	29	2.31^{ab}	3.89^{a}	18.92^{abcde}	50.32^{f}
	36	2.19^{ab}	4.84^{a}	17.18^{abcde}	39.60^{def}
30	1	1.55^{a}	12.56^{bcd}	29.00^{ef}	18.07^{bc}
	8	3.76^{abc}	7.91^{abc}	29.81^{f}	18.62^{bc}
	15	6.74^{cd}	14.29^{cd}	22.99^{cdef}	9.98^{ab}
	29	9.14^{de}	16.22^{d}	26.48^{def}	8.12^{ab}
	36	6.62^{de}	17.41^{d}	20.05^{abcdef}	6.70^{ab}
40	1	4.79^{bc}	16.17^{d}	19.74^{abcdef}	7.39^{ab}
	8	10.55^{e}	26.04^{e}	21.13^{bcdef}	2.88^{a}
	15	13.58^{f}	37.30^{f}	13.06^{abc}	1.33^{a}
	29	18.32^{g}	43.19^{f}	10.58^{a}	0.80^{a}
	36	11.22^{ef}	40.65^{f}	10.79^{ab}	0.91^{a}

* Mean values with different letters in a column indicate significant differences ($P < 0.05$).
J_o, instantaneous elastic compliance; J_1, retarded compliance; τ, retardation time; η_N, Newtonian viscosity.

decreases with increasing temperature, while electrostatic repulsion increases. This suggests that the balance would be shifted toward more repulsion and weakening of the matrix, which is the phenomenon observed. Moreover, the effect of proteolysis (breakdown of covalent protein bonds) during ripening is more detectable by the mechanical evaluation when temperature increases because polypeptide network is even less stabilized. Therefore, the phenomena described may explain the significant differences observed in J_o and J_1 during ripening at elevated temperatures.

The retardation time (τ) was significantly affected by test temperature and ripening time in the case of samples T, while in the case of samples L, no significant effect was found. In the case of samples T, higher τ values were observed at lower temperatures and ripening times. Generally, the higher the retardation time of a system network, the longer it takes to reach full deformation on application of shear stress. This also implies that retardation times are inversely related to network elasticity (Ojijo *et al.* 2004).

The Newtonian viscosity of the free dashpot (η_N) decreases significantly with temperature (Tables 2 and 3). This parameter is associated with the breakdown of the protein network structure (Ma *et al.* 1996, 1997; Messens

TABLE 3.
CREEP COMPLIANCE PARAMETERS FOR SAMPLES OF MOZZARELLA CHEESE CUT PARALLEL TO THE PRINCIPAL AXIS (L) AT DIFFERENT TEMPERATURES AND RIPENING TIMES*

Temperature (C)	Ripening time (days)	J_o (10^{-5}/Pa)	J_1 (10^{-5}/Pa)	τ (s)	η_N (10^5 Pa·s)
20	1	1.85^{a}	4.24^{a}	19.84	33.51^{cd}
	8	1.55^{a}	5.42^{a}	20.21	42.91^{d}
	15	3.18^{abc}	5.16^{a}	18.01	32.53^{cd}
	29	3.23^{abc}	5.78^{a}	18.35	31.56^{cd}
	36	2.89^{ab}	4.89^{a}	16.26	41.20^{d}
30	1	3.28^{abc}	8.26^{ab}	21.48	23.07^{bc}
	8	5.56^{cd}	11.18^{ab}	16.96	7.23^{ab}
	15	7.60^{de}	15.53^{bc}	19.33	8.39^{ab}
	29	3.82^{abc}	19.47^{c}	21.90	10.97^{ab}
	36	5.55^{bcd}	11.38^{ab}	14.42	7.38^{ab}
40	1	3.17^{abc}	14.68^{bc}	26.82	5.16^{a}
	8	9.00^{e}	19.34^{c}	23.66	3.32^{a}
	15	12.76^{f}	30.71^{d}	11.13	1.50^{a}
	29	15.57^{f}	54.50^{f}	14.66	0.76^{a}
	36	14.81^{f}	41.35^{e}	15.71	1.01^{a}

* Mean values with different letters in a column indicate significant differences ($P < 0.05$).
J_o, instantaneous elastic compliance; J_1, retarded compliance; τ, retardation time; η_N, Newtonian viscosity.

et al. 2000; Subramanian *et al.* 2003). η_N characterizes the linear region of the viscous compliance. That is to say, it measures the mechanical behavior of the fluid part of the system. As reported by Lucey *et al.* (2003), fat is the only solid in cheese that truly melts (transformation from a solid-like to a liquid-like state; milk fat is completely liquid at about 40C) in the temperature range evaluated here. The other solids in cheese are caseins and some serum proteins. Nevertheless, as discussed earlier, proteins do not melt, but their interactions with each other can change to produce a matrix relaxation effect. Hence, it may be considered that η_N embodies the fluid behavior of melted fat and the breakdown of the protein network. At elevated temperatures, fat is in liquid state and greatly contributes to η_N, the effect of the protein network breakdown because of proteolysis during ripening being masked.

The effect of temperature on Newtonian viscosity (η_N) was studied by an Arrhenius-type equation,

$$\eta_N = A_{visc} \exp\left({E_a}/{RT} \right) \tag{2}$$

where A_{visc} is the pre-exponential factor, E_a is the activation energy (cal/mol), R is the gas constant (1.9872 cal/mol K) and T is the temperature (K) (Rao

TABLE 4.
ACTIVATION ENERGIES (CAL/MOL) FOR SAMPLES OF MOZZARELLA CHEESE CUT PERPENDICULAR (T) AND PARALLEL (L) TO THE PRINCIPAL AXIS AT DIFFERENT RIPENING TIMES

Ripening time (days)	Samples T	Samples L
1	11,384	16,921
8	20,581	23,407
15	31,405	28,005
29	37,693	33,749
36	34,276	33,719

1999). Table 4 shows E_a values for samples T and L at different ripening times. It is observed that the determined values are in the order of those obtained through complex viscosity determined by dynamic oscillatory tests (Gunasekaran and Ak 2003; Ribero *et al.* 2007).

Full mechanical characterization can be established by calculating the contribution of each main compliance to the maximum deformation to which the system is subjected (Dolz *et al.* 2007). The percentage deformation of J_o and J_1 can be calculated as

$$J_o^*(\%) = \frac{J_o}{J_{max}} \times 100 \tag{3}$$

$$J_1^*(\%) = \frac{J_1}{J_{max}} \times 100 \tag{4}$$

In addition, the final percentage recovery ($R(\%)$) of the system subjected to prior deformation can be calculated as

$$R(\%) = \frac{J_{max} - J_{res}}{J_{max}} \times 100 \tag{5}$$

Tables 5 and 6 show J_{max}, $J_o^*(\%)$, $J_1^*(\%)$ and $R(\%)$ for samples T and L, respectively. Both samples T and L showed approximately a similar behavior. It is observed that J_{max} increases with temperature. Significant differences were observed between samples assayed at 40C and those assayed at 20 or 30C. It seems that when temperature is elevated, the fat melting and decrease of protein interactions allow greater deformation of the cheese matrix. Moreover, significant differences during ripening between J_{max} values were observed only

TABLE 5.
MAXIMUM COMPLIANCE AND NORMALIZED PARAMETERS FOR SAMPLES OF MOZZARELLA CHEESE CUT PERPENDICULAR TO THE PRINCIPAL AXIS (T) AT DIFFERENT TEMPERATURES AND RIPENING TIMES*

Temperature (C)	Ripening time (days)	J_{max} (10^{-5}/Pa)	$J_o^*(\%)$	$J_1^*(\%)$	$R(\%)$
20	1	12.13^{ab}	5.79^{a}	44.33^{de}	13.02^{a}
	8	16.82^{abc}	15.60^{cd}	42.84^{de}	26.53^{ab}
	15	11.55^{a}	22.73^{ef}	40.45^{cde}	84.53^{e}
	29	10.32^{a}	23.31^{f}	37.79^{cde}	67.50^{de}
	36	11.36^{ab}	19.31^{cdef}	42.52^{bcde}	65.59^{cde}
30	1	31.03^{abc}	5.27^{a}	36.79^{bcde}	65.60^{cde}
	8	31.50^{abc}	12.78^{bc}	27.31^{abcde}	45.91^{bcd}
	15	39.15^{abc}	17.04^{bcd}	36.70^{bcde}	59.37^{cd}
	29	47.80^{bc}	19.04^{def}	33.78^{bcd}	64.15^{cde}
	36	50.70^{c}	12.73^{bc}	34.46^{bcd}	62.57^{cde}
40	1	31.22^{abc}	15.68^{cd}	53.54^{e}	29.31^{ab}
	8	120.17^{d}	9.42^{ab}	24.63^{abc}	47.33^{bcd}
	15	186.20^{e}	7.20^{ab}	19.94^{ab}	42.70^{cd}
	29	290.90^{f}	6.31^{a}	14.94^{a}	43.77^{bc}
	36	257.50^{f}	4.49^{a}	16.20^{a}	41.95^{bc}

* Mean values with different letters in a column indicate significant differences ($P < 0.05$).
J_{max}, maximum compliance; $J_o^*(\%)$, percentage deformation of J_o; $J_1^*(\%)$, percentage deformation of J_1; $R(\%)$, final percentage recovery.

when creep tests were carried out at 40C. Proteolysis during ripening weakens cheese structure and greater deformations may be attained.

$J_o^*(\%)$ tends to increase with ripening at lower temperatures in samples T and L, but when samples T are evaluated at 40C, that behavior is opposite (Tables 5 and 6). We may think that, at lower temperatures, the contribution of this parameter to the total deformation (J_{max}) is larger for longer ripening times because proteolysis confers less rigidity to the material. However, at higher temperatures, the rate of increase of the total deformation is larger than the rate of increase of J_o and, hence, $J_o^*(\%)$ behavior with ripening time is inverted, which was particularly observed in the case of samples T.

$J_1^*(\%)$ was significantly affected by temperature in samples T and L, values being lower at higher temperatures (Tables 5 and 6). Moreover, it is observed that $J_1^*(\%)$ decreases with ripening in samples T and at 40C. In all cases studied, the contribution of J_1 to the total deformation was larger than the contribution of J_o, reinforcing the concept of a behavior closer to a viscoelastic material than to an elastic one.

Significant differences in $R(\%)$ with ripening were observed at lower temperatures in samples T and L (Tables 5 and 6). As cheese ages, $R(\%)$

TABLE 6.
MAXIMUM COMPLIANCE AND NORMALIZED PARAMETERS FOR SAMPLES OF MOZZARELLA CHEESE CUT PARALLEL TO THE PRINCIPAL AXIS (L) AT DIFFERENT TEMPERATURES AND RIPENING TIMES*

Temperature (C)	Ripening time (days)	$J_{\max}$ (10^{-5}/Pa)	J_o^*(%)	J_1^*(%)	R(%)
20	1	12.87^{a}	15.53^{c}	33.73^{bcdef}	15.35^{a}
	8	12.28^{a}	14.07^{bc}	44.71^{ef}	45.31^{abcde}
	15	13.77^{a}	23.46^{e}	36.58^{def}	66.46^{de}
	29	14.65^{a}	21.89^{de}	39.68^{def}	74.72^{e}
	36	13.05^{a}	21.94^{de}	38.07^{def}	70.46^{de}
30	1	21.20^{a}	15.27^{c}	39.59^{def}	51.72^{bcde}
	8	34.87^{a}	16.24^{c}	36.23^{def}	57.80^{cde}
	15	45.03^{a}	17.09^{cd}	34.51^{cdef}	60.79^{cde}
	29	41.25^{a}	8.81^{ab}	47.48^{f}	50.01^{bcde}
	36	36.01^{a}	15.86^{c}	31.58^{bcde}	65.63^{de}
40	1	55.70^{ab}	5.79^{a}	28.07^{abcd}	24.07^{ab}
	8	97.27^{b}	9.65^{ab}	22.66^{abc}	42.71^{abcd}
	15	164.67^{c}	7.93^{a}	18.68^{ab}	51.49^{bcde}
	29	308.60^{e}	5.14^{a}	17.87^{a}	36.17^{abc}
	36	264.20^{d}	6.03^{a}	17.28^{a}	48.88^{bcde}

* Mean values with different letters in a column indicate significant differences ($P < 0.05$).
$J_{\max}$, maximum compliance; J_o^*(%), percentage deformation of J_o; J_1^*(%), percentage deformation of J_1; R(%), final percentage recovery.

increases and cheese matrix can recover up to approximately 75% of its maximum deformation. This parameter gives clear evidence of cheese elasticity behavior with ripening. However, that tendency is not clear at higher temperatures (30 and 40C), because in this case, fat in melt state and weaker protein interactions may mask the cheese matrix recovery during ripening.

Kuo *et al.* (2000) proposed a mathematical expression to describe cheddar cheese meltability called viscoelasticity index (*VI*). That expression was generated combining those principal viscoelastic parameters that could provide an objective and physical indicator for cheese meltability evaluation. For this purpose, the instantaneous slope of the creep curve was calculated. For the mechanical model analyzed here, the *VI* is computed as follows

$$VI = \left.\frac{dJ}{dt}\right|_{t=0} = \frac{J_1}{\tau} + \frac{1}{\eta_N} \tag{6}$$

Kuo *et al.* (2000) found that the general trend is that the higher the *VI* parameter, the better the meltability. Values of *VI* are shown in Fig. 6. It is observed that *VI* increases with temperature for samples T and L. Significant

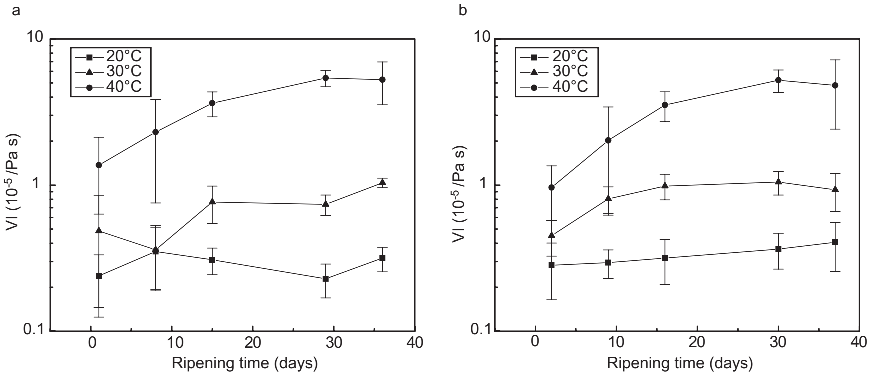

FIG. 6. VISCOELASTICITY INDEX (*VI*) OF MOZZARELLA CHEESE CHANGING WITH RIPENING TIME
(a) Samples T. (b) Samples L. Bars indicate standard deviation.

differences with ripening were found at 40C. Through this parameter, it can be seen that meltability increases with ripening as reported by Kuo *et al.* (2000). However, it is worth mentioning that the test temperature results critical for determining cheese meltability changes during ripening.

Materials with oriented structure generally exhibit anisotropic physical properties. In the case of mozzarella cheese, Cervantes *et al.* (1983) and Ak and Gunasekaran (1997) studied the effect of sampling direction on some textural and rheological properties. Ak and Gunasekaran (1997) found that the sampling direction significantly affected the fracture properties determined with tensile tests. In this case, ANOVA showed that sampling direction as main factor did not affect any of the creep compliance and normalized parameters studied. Therefore, when the creep/recovery tests are used, fiber direction might not affect the rheological properties of mozzarella cheese.

However, it is clear that cheese matrix behaves as a quite different physicochemical system at low and high temperatures. This phenomenon emphasizes the importance of evaluating cheese properties at different temperatures (principally in *pasta filata* type cheese that are consumed in melted conditions).

CONCLUSIONS

Creep/recovery tests were used to obtain information about internal structure of mozzarella cheese and physicochemical changes at different temperatures during cheese ripening. Creep curves were adequately fitted with a

Burger model of four parameters. The elastic and viscous contributions to the general viscoelastic behavior were analyzed through the obtained parameters.

Creep tests at different temperatures allowed gaining a better understanding of changes that occur in the cheese matrix during heating. The evolution of J_o, J_1 and η_N with temperature may be explained by the physicochemical changes that occur during cheese heating. Moreover, the effect of proteolysis (breakdown of covalent protein bonds) during ripening is more detectable by the mechanical evaluation when temperature increases because polypeptide network is even less stabilized.

A semiempirical approach was proposed to obtain the contribution of each main compliance to the total deformation of the system. It was observed that proteolysis during ripening weakens cheese structure and greater deformations may be attained, particularly at higher temperatures. An interesting behavior inversion was observed through the analysis of $J_o^*(\%)$ in samples T. The contribution of J_1 to the total deformation was larger than the contribution of J_o, reinforcing the concept of a behavior closer to a viscoelastic material than to an elastic one.

VI was obtained, and it is a good predictor of mozzarella cheese meltability. In addition, it was observed that test temperature to evaluate *VI* results critical for determining significant cheese meltability changes during ripening. In this case, significant differences with ripening time were found only at 40C.

ANOVA showed that sampling direction as main factor did not affect any of the parameters studied. Finally, it was clearly observed that cheese matrix behaves as a quite different physicochemical system depending on temperature. Therefore, it is recommended to carry out the rheological tests at different temperatures to evaluate appropriately the viscoelastic properties of mozzarella cheese.

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