Ripening process of Cascaval cheese: compositional and textural aspects

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Abstract Two textural characteristics, elasticity modulus and firmness, were determined during the ripening process of Cascaval cheese, using both instrumental and sensorial techniques. Uniaxial compression was used to determine the textural characteristics and the results were compared with the ones obtained by sensorial analysis, revealing a good correlation. The chemical composition of cheese was also determined, including the nitrogen fractions (total nitrogen, water soluble nitrogen, non-protein nitrogen and phosphotungstic acid soluble nitrogen). The data thus obtained were statistically processed in order to find the differences between the samples, as well as to find the correlation between the techniques of analysis. The study showed that the ripening process of the Cascaval cheese is similar to the ripening of other pasta filata cheese. The moisture content decreases during maturation as a result of water evaporation. The concentration of nitrogen fractions increases during the ripening stage, and so do the firmness and elasticity modulus. The biochemical processes that occur during maturation largely influence the textural parameters and this is proved by both instrumental and sensorial analyses.

Keywords Cascaval cheese · Texture characteristics · Nitrogen fractions · Ripening

Introduction

Consumer behaviour and the decision to purchase a product are closely related to the product quality. Food quality is a complex notion that includes several aspects: legal,

nutritional, sensorial and hygienic. Out of the aspects mentioned above, the sensorial one is very important to the consumer (Foegeding et al. 2011; Gunasekaram and Ak 2003) due to the fact that his first contact with the product is sensorial. Texture is a quality attribute that is closely related to the structural and mechanical properties of a food product (Lu and Abbott 2004).

Texture is one of the food characteristics that was traditionally evaluated by panellists, who, despite advanced training, may make subjective assessments (Kealy 2006; O’Callaghan and Guinee 2004). Moreover, sensorial analysis involves considerable costs (Guinee and O’Callaghan 2010). In these conditions, one of the researchers’ concerns refers to finding certain scientific methods to measure texture characteristics and to correlate the mechanical to the sensorial characteristics.

Cascaval is a cheese type with a specific texture, part of the category of *pasta filata* cheese, together with Mozzarella, Provolone and Ragusano (Kindstedt et al. 2003). The defining feature of all pasta filata cheeses is the unique thermising and texturising process that occurs at the end of cheese making, when the curd is immersed in hot water, whey or salt brine and worked mechanically into a hot plastic consistency, followed by moulding into a desired shape and size (Kindstedt et al. 2010). The texturising process makes the cheese curd reorganize into a unidirectional fibrous structure, due to the calcium phosphate that is available to crosslink the amorphous *para*-casein matrix at the moment when heat is applied to the curd (Kindstedt et al. 2003).

Another important process in the Cascaval cheese manufacturing technology is ripening (Johnson and Law 2010), a complex process that defines its textural properties and specific flavour.

The specific texture of the Cascaval cheese makes it very appreciated by consumers, both in its unprepared state, as well as part of some dishes.

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Scanning electron microscopy micrographs of the Cascaval cheese and other cheese varieties showed distinct differences in the microstructure of the cheeses, which helped to explain their textural properties (Kindstedt et al. 2003; Hassan 1988). Cheese hardness was correlated with its chemical composition: increased protein and NaCl resulted in higher cheese hardness, while higher contents of water, fat and pH, as expected, reduced hardness (Kindstedt et al. 2003; Hassan 1988).

Specialized literature abounds in information about the physico-chemical parameters of cheese ripening (Delgado et al. 2012; Zaharia 2012; Karami et al. 2009; Madadlou et al. 2006; O'Reilly et al. 2002), and the texture parameters of cheese ripening (Delgado et al. 2012; Madadlou et al. 2006; Muir et al. 1996), but there are few references to the correlation between them (Imm et al. 2003; Sheehan and Guinee 2004; Sheehan et al. 2003).

Under these circumstances, the aim of this paper is to find a correlation between the physico-chemical parameters and the textural characteristics of the Cascaval cheese during ripening. This correlation is important in modelling and optimizing the technological process of the Cascaval cheese. The data obtained in the study contribute to elucidating some topical problems related to obtaining cheese texture in general, and to the pasta filata cheese texture in particular. Likewise, these data are useful for the technologists, who want to obtain a proper texture of the Cascaval cheese, suitable for consumer's demands. On the other hand, the inconveniences of sensorial analysis (subjectivity, high costs) may be eliminated by investigating the instrumental methods of texture evaluation.

Materials and methods

Sample preparation

The Cascaval cheese was obtained using the traditional method described by Costin 2003 (Fig. 1). The raw cow milk, with 3.5 % fat and 15°T acidity (pH=6.5), was heated at 35 °C. A commercial mixed culture of lactic bacteria (*Streptococcus thermophilus*, *Lactobacillus delbrueckii*ssp.*Bulgaricus* and *Lactobacillus helveticus*) was added 0.5 % (w/w). When the acidity of 25°T (pH=6.4) was reached, rennet (1/4 tablet of Fromase 50 - DSM Food Specialties per 12 l of milk) was added so that the milk coagulated in 40 min. After pressing, the curd was acidified at 185°T (pH=4.8) and texturised in hot brine, at 73 °C for 60 s. Then the curd was moulded, dried and ripened for 56 days. All the analyses were carried out in triplicate after 0, 14, 28, 42 and 56 days of ripening.

Compositional analyses

It is considered that cheese texture is influenced by factors that include the pH and the state of calcium (ionic or bound to the

casein matrix), the sodium chloride content, and the extent of proteolysis (Everett and Auty 2008). The pH was measured directly, by immersion of the pH meter electrode (IQ-SCIENTIFIC) into the samples. The state of the calcium was evaluated by determining the total calcium and the soluble calcium content, using a 1 UV VIS UNICAM UV 2 (Italy) Spectrophotometer at 185–900 nm wavelength. The sodium chloride content was determined using the method shown in SR EN ISO 5943:2007. The extent of proteolysis was assessed by analysing the nitrogen fractions. The total nitrogen (TN) and the water soluble nitrogen (WSN) were determined by the Kjeldhal method, according to SR ISO/TS 17837:2009. The non-protein nitrogen (NPN) was determined by means of SR EN ISO 8968–4:2002. The content of proteins (TP) was determined by the Kjeldahl method with conversion to protein content using a factor of 6.38. The phosphotungstic acid soluble nitrogen (PWN) was determined by the Kjeldahl analysis, from the post-precipitation filtrate of N-components from the solution with phosphotungstic acid.

In order to complete the compositional analysis of the samples, acidity (according to SR 2418:2008), dry matter (according to SR ISO 5534:2004) and fat content (according to SR ISO 3433:2009) were determined as well.

Textural analyses

Out of the textural parameters, firmness and the modulus of elasticity were determined by uniaxial compression. The cheese was cut in cylindrical pieces, 10 mm in diameter and 10 mm in length. The samples were compressed at 54.5 % (deformation) using a Testometric machine, M350-5AT model. The crosshead speed was 5 mm/s. The firmness expressed as the maximum compression force (N) and elasticity modulus expressed as the ratio between the normal stress and strain on the material (N/mm²) were measured. The method was elaborated taking into account the recommendations of O'Callaghan and Guinee 2004, and Gunasekaram and Ak 2003.

The sensorial analysis was performed with the help of 10 trained panellists, 8 female and 2 male, aged between 25 and 50, who were asked to evaluate firmness (the extent of resistance offered by the cheese, assessed during the first five chews using the front teeth) - Delahunty and Drake 2004 and elasticity (the extent to which the cheese returns to its initial form after biting, assessed during the first 2–3 chews) Delahunty and Drake 2004 – by giving a score between 0 (very low) and 5 (very high). The sensorial analyses took into account the recommendations of Lawless and Heymann 2010. The analysis took place at room temperature, in a room fitted with white fluorescent lighting. The samples were cut in cube shape (2x2x2 cm) and presented to panellists on white plates. Water and bread were supplied.

Fig. 1 The flowchart for manufacturing Cascaval cheese

Statistical analyses

The data were statistically processed using STATISTICA 8 programme. The differences among the samples were evaluated by means of the one-way analysis of variance (ANOVA).

Results and discussions

Cheese composition

The main physico-chemical characteristics of the Cascaval samples during ripening are shown in Table 1.

Dry matter increased during maturation by approximately 13.2 % (from 51.45 % at the beginning of the process to 58.28 % at the end of the process), due to evaporation of water, the samples subjected to ripening being unpackaged. By analysing the values of dry matter in Table 1 it can be noticed that the greatest quantity of water (7 %) was lost in the initial 14 days and during the intervals this quantity decreased: 4 %, 1 % and 0.6 % respectively. This evolution can be explained by the formation of a rind at the surface of the sample, which obstructs water evaporation. An increase of

dry matter was reported by (Simov and Ivanov 2005) but the percentage was smaller (0.4 %) due to ripening of samples while packaged. There were no significant differences ($P < 0.05$) in the dry matter content of the tested samples.

In order to get an objective conclusion, the fat content was compared to dry matter and the results are shown in Table 1. During maturation, a slight decrease of fat in dry matter can be noticed, i.e. 0.25 % (from 48.48 % at the beginning of the process to 48.36 % at the end of the process). This decrease, more noticeable in the final ripening interval, is due to the lipolysis process. For the Cascaval cheese, the intensity of the lipolytic process is moderate, because of the enzymatic systems' inactivation during the immersion of the curd into hot brine during the texturising process (Kindstedt et al. 2003).

The salt content registered an increasing with 14 % (from 2.62 % at the beginning of the process at 3 % at the end of the process) during maturation, simultaneous with the increasing of dry matter. There were no significant differences ($P < 0.05$) in salt content of tested samples. The values are smaller than those reported by (Simov and Ivanov 2005), probably, because of the more concentrated brine they used in manufacturing process.

During the ripening process, the accordance between the increase of the titratable acidity and the decrease of the pH

Table 1 Variation of the main physico-chemical characteristics of Cascaval cheese during ripening process

Parameter	Ripening stage				
	0 days	14 days	28 days	42 days	56 days
Dry matter, %	51.45±0.009*	55.12±0.025	57.33±0.018	57.92±0.057	58.28±0.039
Fat in dry matter, %	48.48±0.079	48.47±0.121	48.45±0.091	48.42±0.109	48.36±0.086
Protein in dry matter, %	42.04±0.023	41.25±0.438	40.17±0.521	39.43±0.193	39.07±0.687
NaCl content, %	2.62±0.210	2.81±0.165	2.94±0.743	2.86±0.381	3±0.492
Acidity, °T	148±0.235	174±0.354	185±0.187	192±0.243	197±0.215
pH	5.4±0.004	5.1±0.017	5±0.008	4.9±0.011	5±0.024
Soluble calcium, %	0.31±0.003	0.51±0.002	0.54±0.006	0.55±0.003	0.55±0.001

* mean±standard deviation

may be noticed. The acidity rose by 33 % (from 148°T at the beginning to 197°T at the end of the process) during the whole maturation process, more visibly (with 17 %) in the initial interval, due to the lactose content which was higher than in the final intervals. The pH values are similar to those reported by (Simov and Ivanov 2005; Kindstedt et al. 2010).

The content of proteins reported to dry matter decreased by 7.6 % during the ripening time, mainly on account of the proteolysis process. This evolution was also reported by (Simov and Ivanov 2005; Kindstedt et al. 2003).

Protein degradation occurs gradually during ripening. One method to evaluate this degradation is to determine the nitrogen soluble in, or extractable by, various solvents or buffers (Upadhyay et al. 2004). The evolution of nitrogen fractions are shown in Fig. 2.

The water-soluble nitrogen (WSN) is important in determining the primary proteolysis index (PPI) as the ratio between WSN and TN (Costin 2003). Fig. 1 shows that at the end of the maturation interval the WSN is almost triple (1.06 %) than at the beginning of the interval (0.41 %). In these conditions, PPI ranged between 6.05 and 15.9 %. The results are similar to those reported by (Zaharia 2012) taking

into account the fact that in the current study the cheese was obtained from unpasteurized milk. There are also similarities with the results presented by (Simov et al. 2006).

Non-protein nitrogen (NPN) is an indicator of secondary proteolysis, as long as the secondary proteolysis index (SPI) is defined as the ratio between NPN and TN (Costin 2003). NPN increased by 312 % during ripening (from 0.16 to 0.58 %). An increase in NPN is reported by many other authors that studied the Cascaval cheese ripening (Zaharia 2012; Zaharia and Rotaru 2011; Simov et al. 2006; Simov and Ivanov 2005), but the increase rate varies with many factors (the coagulant enzyme, the lactic bacteria used for milk acidification, the enzymes used to accelerate ripening). In the case of the present study, in the first 2 weeks of ripening, NPN increased by 225 % (almost 2/3 of the total increase). The SPI registered values from 2.3 %, at the beginning of the interval, up to 8.46 %, at the end of the interval.

NPN is associated with casein-derived peptides which are either tasteless or bitter and do not contribute directly to the typical taste or flavour of cheese, but their production, particularly in the initial weeks of cheese maturation, is essential to the softening of the rubbery, elastic cheese curd texture (Law 2010).

Fig. 2 Nitrogen fractions variation

Table 2 Values of firmness and Young's modulus for Cascaval cheese during ripening process

Parameter	Ripening stage				
	0 days	14 days	28 days	42 days	56 days
Firmness, N	4.401±0.143*	4.884±0.182	5.202±0.626	5.256±0.851	7.888±0.497
Young's modulus, N/mm ²	0.067±0.009	0.083±0.002	0.084±0.001	0.087±0.006	0.089±0.006

* standard deviation

The phosphotungstic acid-soluble nitrogen (PWN) is an indicator of free amino acids and small peptides, with a smaller molecular weight than 2000 Da (Costin 2003). For the Cascaval cheese samples under analysis, the PWN content ranged between 0.017 % at the beginning of ripening, and 0.11 % at the end of the process (Fig. 2). The most notable increase was registered in the last 2 weeks, due to the increase of the proteolysis process. The PWN values are comparable to those presented by (Zaharia and Rotaru 2011).

The soluble calcium content rose from 0.31 % at the beginning of ripening, to 0.51 % in 2 weeks of ripening and 0.55 % at the end of the process. This behaviour is explained by the increase of the lactic acid amount, which favours the calcium solubilisation (Kindstedt et al. 2003; Costin 2003).

Cheese texture

Firmness expressed as the maximum compression force (N) and elasticity modulus expressed as the ratio between the normal stress and strain on the material (N/mm²) were measured.

The values of firmness and elasticity (Young's) modulus are given in Table 2.

Firmness expressed as maximum compression force, is 1.1 times greater at 14 ripening days, comparing to the beginning of maturation. In the next 2 weeks the firmness increased approximative 1.2 times comparing to 0 days ripening; at the end of the process it is 1.79 times bigger than at the beginning. This evolution is consistent to the earlier results according to which firmness increases with the increasing of dry matter and protein content and decreases with the increasing of fat content. The results are comparable with those reported by Juan

et al. 2008 for ewes' cheese and by Madadlou et al. 2006 for Iranian White Cheese. The differences come from the different technological processes applied.

Elasticity modulus increased with 33 % during whole ripening interval, the more accentuated increasing (23 %) registering in the first 14 days. The explanation of this behaviour is given by Gunasekaram and Ak 2003 "as the moisture content increases, the resistance of cheese to deformation, and hence its modulus of deformability decreases".

The evolution of firmness and elasticity determined by compression tests is similar to that determined by sensorial analysis (Fig. 3).

Statistical analysis shows no significant differences ($P < 0.05$) as far as the sensorial score for firmness is concerned. Figure 3a shows a good correlation between the sensorial score and the compression values for firmness $R^2 = 0.9288$. A good correlation was also found for the elasticity modulus and the sensorial score for the sample elasticity, $R^2 = 0.8903$.

In order to find a correlation between textural parameters (firmness and elasticity modulus) and the physico-chemical composition of the Cascaval cheese, STATISTICA 8 software was used to generate equations that describe their dependence.

Figure 4 shows the graphical representation of the firmness and Young's modulus against the fat content and the protein content. The equations generated by the software are:

$$F = 23.35 + 1.17 \cdot P - 2.84 \cdot Fa \quad (1)$$

$$YM = -13.6 + 4.79 \cdot P + 3.89 \cdot Fa \quad (2)$$

where F is firmness, in Newtons, YM is Young's Modulus, in kiloPascals, P is the content of proteins, in % and Fa is the content of fat, in %. Equation (1) shows that firmness is

Fig. 3 Correlation between sensorial score for firmness and compression firmness (a) and sensorial score for elasticity and elasticity modulus (b)

Fig. 4 The influence of fat content (*Fa*) and protein content (*P*) on firmness (**a**) and on Young's Modulus (**b**)

directly influenced by protein content and indirectly influenced by fat content, which is in accordance with (Kindstedt et al. 2003; Hassan 1988). The relationship between the coefficient of protein content (1.17) and fat content (2.48) shows that the firmness of the Cascaval cheese is influenced more by fat content than by protein content. Young's Modulus (Eq. 2) increases with both protein and fat content and this is accountable as long as the casein matrix provides the elastic character of cheese texture (Madadlou et al. 2006). It can be noticed that the influence of protein content on the Young's Modulus is greater than the influence of fat content.

$$F = 4.69 - 0.07 \cdot \frac{CaS}{CaT} + 0.43 \cdot \frac{WSN}{TN} \quad (3)$$

$$YM = 39.15 + 0.56 \cdot \frac{CaS}{CaT} + 0.88 \cdot \frac{WSN}{TN} \quad (4)$$

Equation 3 shows the correlation between firmness, soluble calcium (CaS)/total calcium (CaT) ratio and the water soluble nitrogen (WSN)/total nitrogen (TN) ratio. Firmness increases with the decrease of the CaS/CaT ratio and this is in accordance with (Gunasekaram and Ak 2003). This relationship

between firmness and CaS/CaT ratio can be explained by the fact that the protein matrix is destabilised during calcium solubilisation. The increase of firmness with the WSN/TN ratio shows that the degree of proteolysis is lower than the degree of water evaporation (Fig. 5).

In Eq. 4, that shows the correlation between Young's Modulus, CaS/CaT ratio and WSN/TN ratio, it can be noticed that YM is positively influenced by the two parameters. The value of the free term in Eq. 4 indicates that the considered parameters are not significant for the Young's Modulus.

Conclusions

This study revealed that the Cascaval cheese ripening process follows the pattern of the pasta filata cheese ripening.

The biochemical processes that occur during maturation largely influence its textural parameters, sometimes in agreement and sometimes in disagreement with other reports, which is due to the specific technological stages in the Cascaval cheese manufacturing process.

Fig. 5 The influence of WSN/TN and CaS/CaT ratios on firmness (**a**) and on Young's Modulus (**b**)

The same may be said about the Cascaval cheese in point of textural parameters (firmness and elasticity modulus) and the content of dry matter. Both sensorial and instrumental techniques were used in support of this statement.

The sensorial method for firmness and elasticity determination can be successfully replaced by instrumental methods.

Thus, it is obvious that the results obtained offer new research vistas in the field, including other techniques of texture analysis, microstructure analysis and modelling.

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