

Full Length Research Paper

Physico-chemical and rheological properties of prato cheese during ripening

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The influence of storage temperature (6, 12 and 18°C) on texture parameters of Prato cheese were evaluated during 22 days of ripening. A reduction of firmness, gumminess and elasticity was observed; however, cohesiveness and adhesiveness increased. With the increasing temperature, firmness and gumminess reduced, whereas the elasticity, cohesiveness and adhesiveness did not change. It can be concluded that storage temperature influences the texture profile of Prato cheese which present differences in the evaluated parameters during 22 days of ripening.

Key words: Cheese ripening, hard cheese, colour, texture profile analysis.

INTRODUCTION

Prato cheese is the cheese of semi-cooked dough, and it began to be manufactured in Brazil in the 1920s by Danish immigrants in an attempt to produce cheeses similar to Danish Danbo and Dutch Gouda cheeses (Furtado et al., 1994). Prato is a matured cheese with average moisture, is semi-rigid, and exhibits a mild flavour and yellow hay colour provided by the natural dye urucum (Kubo et al., 2013). Prato cheese is produced in all regions of the country, with an average annual production of approximately 150,000 tons (ABIQ, 2010). Maintenance of the identity and quality standards of Prato cheese is of fundamental importance to meet the expectations of the consumer. Texture is an important parameter for the identification of quality, as the

consumer usually evaluates the colour and aroma prior to other characteristics (Santos Júnior et al., 2012). Many parameters influence the texture of cheeses because the lower the fat content of cheese is, the denser the casein network and the firmer the cheese (Tunick et al., 1993).

Rheological methods that study material deformation and flow are used to measure the texture properties of materials. Furthermore, these methods are used to determine the elastic and viscous components as well as to elucidate short-reach interactions, such as casein conformation and structure (Steffe, 1996). One of the methods used to evaluate the texture of cheeses is a texture profile analysis (TPA), which simulates the compression of molar teeth during mastication, allowing

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the prediction of texture characteristics even before product consumption (Delgado et al., 2011). A TPA generates values of firmness, cohesiveness, elasticity, adhesiveness and gumminess that enable the definition of the texture parameters of cheese (Amar and Surono, 2012).

The objective of this study was to evaluate the physical and chemical characteristics of the cheese plate, in addition to studying the variations of texture of Prato cheeses by TPA at different temperatures (6, 12 and 18°C) at the 1st, 8th, 15th and 22nd day of maturation.

MATERIALS AND METHODS

The study was developed during the period from June 9th to July 3rd of 2014, in a dairy industry facility located in the city of Rio Verde, GO, Brazil, and the Post-Harvest of Vegetable, Fruits, Greenery and Products of Animal Origin Laboratories of the Federal Institute of Goiás, Rio Verde Campus, GO, Brazil. Prato cheeses were used that were produced by a dairy industry facility registered at the Federal Inspection Service of the Ministry of Agriculture, Animal Farming and Supply of Brazil.

Quality of refrigerated milk used to process prato cheese

The quality evaluation of the milk used to process the Prato cheeses was performed in the Milk Quality Laboratory from the Centre for Food Research of the School of Veterinary and Animal Science (Centro de Pesquisa em Alimentos da Escola de Veterinária e Zootecnia) of the Federal University of Goiás, Goiânia, GO, Brazil. Prior to cheese fabrication, milk samples were packaged in flasks containing Bronopol[®] preservative. After the collection of milk samples, the flasks were transported in isothermal boxes with ice for the evaluation of fat, protein, lactose, total dry matter and degreased dry extract contents using Milkoscan 4000 equipment (Foss Electric A/S. Hillerød, Denmark) (International Dairy Federation, 2000). Before the analysis, the flasks containing the milk samples were heated in a water bath at 40°C for 15 min for fat dissolution. The results are expressed in percentage (%).

Preparation of prato cheese

The Prato cheeses were produced with refrigerated milk, standardised at 3.3% fat, pasteurised under constant agitation at 75°C for 16 s, followed by cooling at the temperature of 35°C. Next, lactic acid cultures were added (mesophilic cultivation based on *Lactococcus lactis* and *Lactococcus* subsp. *cremoris* and thermophilic cultivation based on *Lactobacillus delbrueckii* subsp. *Bulgaricus* and *L. delbrueckii* subsp. *helveticus*) at a proportion of 100 g of lactic acid cultures to 5,000 L of milk, calcium chloride (solution at 40% m/m) at a proportion of 2.5 to 5,000 L of milk, urucum natural dye (500 mL to 5,000 L of milk) and industrial rennet (300 mL for 5,000 L). Next, the milk was allowed to rest for 40 min until complete coagulation. Then, the cheese curd was cut using a 2 cm blade curd knife and allowed to rest for 10 min, followed by the removal of 30% of the milk whey. The second mass mixing took place 20 min afterwards, followed by pre-pressing in a pneumatic press and moulding in 4 kg moulds. After moulding in dewheying moulds, the Prato cheese was pressed for 120 min in a pneumatic press at 40 lb/pol², and the cheeses were immersed in brine at 10°C (21% of salt and pH 5.3) for 24 h. After brining, the cheeses were remained in the drying chamber for 24 h at 8°C, were packaged in thermo-shrinkable packaging and were stored in a BOD incubator oven at 10°C for maturation (for 22 days).

Physical and chemical analysis of prato cheeses

Instrumental analysis of the texture profile of prato cheese

The instrumental texture of Prato cheese was measured over 22 days (1st, 8th, 15th and 22nd day of maturation). Thirty-six cheese bars of ± 4 kg, divided into three equal parts to improve sampling, were used. A 2 cm piece of the outer layer of cheese (crust) was discarded so as not to interfere with the analysis. The cheese was cut in 2 cm cubes with a calliper. Prior to the analysis of texture, the 2 cm Prato cheese cubes were conditioned at three temperatures (6, 12 and 18°C). During the maturation period, three repetitions were performed for each evaluation temperature, for the three sections of the cheese, giving a total of nine repetitions per temperature. The TPA was analysed using a Brookfield texturometer and a 5 g trigger, probe displacement speed of 1 mm/s and compression rate of 50%. The firmness, elasticity, adhesiveness, gumminess and cohesiveness parameters were obtained.

Instrumental colour parameters of prato cheese

The colour was analysed with a Hunter Lab digital colourimeter during the Prato cheese maturation period. For this step, the cheese was cut into 2 cm cubes, and cubes from the middle part and extremities of the cheese were selected to provide better sampling. The cubes were placed on the reader to obtain L*, a* and b* values, which were converted into Chroma ($\sqrt{(a^*^2 + b^*^2)}$) and Hue ($\tan^{-1}(b^*/a^*)$) (Venturini et al., 2011). Sampling was performed as described by the TPA, giving a total of 27 colour analyses per day of evaluation.

Physico-chemical parameters of prato cheese

The pH, titratable acidity and moisture of Prato cheese were analysed according to the methodology proposed by the AOAC (1995) during the maturation period. The fat content was analysed by Gerber's method (Furtado, 1975), the total protein by Kjeldahl's method (AOAC, 1995) and the ash content by the gravimetric method (AOAC, 1995) at the 22nd maturation day.

Statistical analysis

The statistical analysis was performed using a completely randomised design with Tukey's test for the comparison of textures, at the temperatures of 6, 12 and 18°C, between the 1st, 8th, 15th and 22nd maturation days. The average results of pH, titratable acidity and moisture between the maturation days were compared by Tukey's test. For the statistical analysis, a Sisvar computer program (Ferreira, 2008) was used. Differences between treatments were considered significant at p<0.05. The average fat, protein and ash results were presented descriptively from the statistical analysis.

RESULTS AND DISCUSSION

The milk used in the preparation of Prato cheeses presented average values of 3.67 ± 0.01% fat, 3.17 ± 0.01% protein, 4.32 ± 0.01% lactose, 12.12% ± 0.01% total dry matter and 8.45 ± 0.02% non-fat dry matter. These values are considered adequate for industrial cheese processing in Brazil, where the legislation determines a minimum content of 3.0% fat, 2.9% total protein, 11.4% total dry matter and 8.4% degreased dry

Table 1. Firmness (g) of Prato cheese at 6, 12 and 18°C during the maturation period.

Storage (days)	Temperature		
	6°C	12°C	18°C
1	10603.67±789.72 ^a	9281.11±530.83 ^a	7852.67±487.06 ^a
8	9860.89±498.22 ^a	7974.66±1065.25 ^{ab}	7684.56±246.89 ^a
15	7928.50±374.08 ^b	7401.22±1204.25 ^b	6512.33±234.06 ^b
22	7691.00±563.56 ^b	6599.55±640.43 ^c	6344.00±388.53 ^b
CV (%)	6.41	11.58	5.09

Averages in the columns followed by different letters represent significant differences according to Tukey's test at 5% significance.

Table 2. Cohesiveness of Prato cheese evaluated at 6, 12 and 18°C during the maturation period.

Storage (days)	Temperature		
	6°C	12°C	18°C
1	0.62±0.02 ^b	0.65±0.02 ^a	0.63±0.02 ^b
8	0.63±0.02 ^{ab}	0.65±0.02 ^a	0.65±0.01 ^{ab}
15	0.65±0.02 ^a	0.66±0.02 ^a	0.65±0.02 ^{ab}
22	0.65±0.02 ^a	0.66±0.01 ^a	0.66±0.02 ^a
CV (%)	2.79	2.47	2.33

Averages in the columns followed by different letters represent significant differences according to Tukey's test at 5% significance.

Table 3. Adhesiveness (mj) of Prato cheese evaluated at 6, 12 and 18°C during the maturation period.

Storage (days)	Temperature		
	6°C	12°C	18°C
1	0.82±0.04 ^a	0.79±0.03 ^a	0.79±0.06 ^a
8	0.83±0.01 ^a	0.83±0.10 ^a	0.79±0.03 ^a
15	0.84±0.05 ^a	0.84±0.05 ^a	0.80±0.07 ^a
22	0.85±0.05 ^a	0.86±0.05 ^a	0.84±0.05 ^a
CV (%)	5.96	7.77	6.94

Averages in the columns followed by different letters represent significant differences according to Tukey's test at 5% significance.

extract (Brasil, 2011). There are no current standards for lactose. During the maturation period, the firmness of Prato cheese evaluated at the temperatures 6, 12 and 18°C decreased with increasing temperature (Table 1).

The cohesiveness of Prato cheese evaluated at the temperatures of 6 and 18°C increased during the maturation period; however, at 12°C, it remained constant (Table 2). The adhesiveness of Prato cheese evaluated at the temperatures 6, 12 and 18°C remained constant during the maturation period (Table 3). The elasticity of Prato cheese evaluated at 6, 12 and 18°C Table 4. Elasticity (mm) of Prato cheese evaluated at the temperatures 6, 12 and 18°C

Storage (days)	Temperature		
	6°C	12°C	18°C
1	8.58 ±0.03 ^a	8.69 ±0.14 ^a	8.45 ±0.21 ^a
8	8.47 ±0.03 ^a	8.35 ±0.17 ^b	8.44 ±0.21 ^a
15	8.18 ±0.08 ^b	8.25 ±0.10 ^b	8.25 ±0.10 ^{ab}
22	8.16 ±0.15 ^b	8.01 ±0.22 ^c	8.11 ±0.12 ^b
CV (%)	1.93	1.95	2.04

Averages in the columns followed by different letters represent significant differences according to Tukey's test at 5% significance.

during the maturation period. decreased during the maturation process (Table 4). The gumminess of Prato cheese evaluated at 6, 12 and 18°C decreased during the maturation period (Table 5). Regardless of the different evaluation temperatures, during the maturation period, the Prato cheese had reduced firmness and increased cohesiveness, and the adhesiveness remained constant. The same pattern was reported by Sant'ana et al. (2013) for the texture evaluation of fresh Minas cheese during 21 days of storage. Guinee et al. (2001) also reported that the firmness of Mozzarella cheese was reduced during maturation. At the 8th day of maturation at 18°C, Prato cheese presented a firmness of 7,684.56 g, a cohesiveness of 0.65 and an elasticity equal to 8.44 mm. Instrumental texture values lower than the values obtained in the present study were reported by Trancoso-Reyes et al. (2014), who evaluated the texture of traditional Mexican cheese 10 days after production. They verified a firmness of 5,810 g, a cohesiveness of 0.22 and an elasticity of 4.5 mm. At the 22nd maturation day, Prato cheese evaluated at 6°C presented a firmness of 7,691 g. Higher firmness values than those obtained for the present study were reported by Bayarri et al. (2012), who evaluated the viscoelasticity and texture of cream cheese refrigerated at 10°C. These authors reported firmness values of 13,054 g.

The Prato cheese in the present study presented constant firmness during the first eight days of evaluation; according to De Jong et al. (1976) during this phase,

Table 5. Gumminess (g) of Prato cheese evaluated at 6, 12 and 18°C during the maturation period.

Storage (days)	Temperature		
	6°C	12°C	18°C
1	6971.67±652.12 ^a	6257.78±398.13 ^a	5049.56±502.41 ^a
8	6368.00±368.85 ^a	5073.33±575.33 ^b	4851.00±677.13 ^{ab}
15	5096.63±443.78 ^b	4851.00±677.13 ^{bc}	4795.78±518.81 ^{ab}
22	4737.88±552.82 ^b	4193.33±371.44 ^c	4193.33±371.44 ^b
CV (%)	8.84	10.23	11.20

Averages in the columns followed by different letters represent significant differences according to Tukey's test at 5% significance.

Table 6. Firmness (g), cohesiveness, adhesiveness (mj), elasticity (mm), and gumminess (g) of Prato cheese at the 22nd maturation day at different temperatures.

Texture	Temperature (°C)			CV (%)
	6%	12%	18%	
Firmness	10603.67±789.72 ^a	9281.11±530.83 ^b	7852.67±487.06 ^b	7.89
Cohesiveness	0.65±0.02 ^a	0.66±0.01 ^a	0.66±0.02 ^a	2.51
Adhesiveness	0.85±0.05 ^a	0.86±0.05 ^a	0.84±0.05 ^a	6.23
Elasticity	8.16±0.15 ^a	8.01±0.22 ^a	8.11±0.12 ^a	2.09
Gumminess	4737.88±552.82 ^a	4193.33±371.44 ^b	4193.33±371.44 ^b	9.97

Averages in the same row followed by different letters represent significant differences according to Tukey's test at 5% significance.

there are no significant alterations in cheese texture, which is the result of degradation at the protein matrix that occurs after the lipid phase, a discontinuous process. From the 8th to 15th day of maturation, Prato cheese had reduced firmness. According to De Jong et al. (1976), in the first seven to 14 days of maturation, rubbery cheese texture converts into the characteristics of the variety under analysis. It is believed that during this phase, proteolysis of the casein network stops and hydrolysis by residual coagulation of approximately 20% of casein- α 1 starts, producing peptide α 1-I and causing weakening of the casein network. At the maturation period from the 15th to 22nd day, the reduction of firmness in Prato cheese was noticeable. De Jong et al. (1976) reported that after 14 days of maturation, gradual texture changes occur in cheese. It is during this time period that the remaining α 1-casein and other caseins are hydrolysed. The cohesiveness of Prato cheese during the maturation period remained constant. According to Bourne (2002), this occurs when the proportion of the compression area of the second area relative to the first area is the same in the TPA analysis.

During the TPA analysis, Prato cheese presented high elasticity, almost returning to the same initial state when it is compressed 50%. Creamer and Olson (1982) observed that cheeses with higher pH are plastic and elastic. At a higher pH, the casein molecules have a

negative charge, causing the protein aggregates to absorb water to solvate the non-neutral ionic charges. The elastic characteristic demonstrated by Prato cheese in the present study can be explained by the high content of fat (28.43%). According to Adda et al. (1982), cheeses of higher fat content are firmer and more elastic. The gumminess of Prato cheese decreased in the maturation period. According to Bourne (2002), gumminess is directly proportional to firmness, and therefore, gumminess decreases with reduced firmness. This pattern was observed in the present study. At the end of Prato cheese maturation (22 days), firmness and gumminess were reduced with the increase in temperature up to 12°C, whereas elasticity, cohesiveness and adhesiveness were not influenced by the different temperatures at the moment of analysis (Table 6).

Hydrophobic interactions played an important role in the determination of the conformation and interaction of the protein molecules (Nakai and Li-Chan, 1988). They manifested as strong attraction forces between the non-polar groups separated by water (Ben-Naim, 1980). When dealing with molecule arrangement, the molecules are grouped as a function of temperature. At low temperatures, the molecules of a material do not move or move too little, as the result of low energy. Thus, the molecules move closer. However, at high temperatures, the molecules move further apart, and because of the

Table 7. Colour parameters of Prato cheese evaluated during 22 days of maturation.

Colour parameters	Storage (days)				CV (%)
	1	8	15	22	
Chroma	22.65±0.75 ^d	24.47±0.57 ^c	27.82±0.31 ^b	30.42±1.22 ^a	2.91
H ^o	1.42±0.01 ^c	1.44±0.00 ^b	1.45±0.01 ^b	1.47±0.01 ^a	0.42

Table 8. The pH, titratable acidity (5 of lactic acid) and moisture (%) of Prato cheese during maturation.

Parameter	Storage (days)				CV (%)
	1	8	15	22	
pH	5.32±0.01 ^a	5.30±0.02 ^b	5.16±0.01 ^c	5.10±0.02 ^d	0.31
Titratable acidity	0.94±0.10 ^c	1.01±0.16 ^c	1.40±0.15 ^b	1.71±0.37 ^a	17.97
Moisture	43.61±1.24 ^a	43.60±1.23 ^a	43.65±1.24 ^a	43.61±1.24 ^a	2.85

Averages in the same row followed by different letters represent significant differences according to Tukey's test at 5% significance.

high energy, the molecules are in constant movement and need more space to be organised, a relation that is visible in the characteristics of water. In water at a solid state, the molecules contain lower energy and are closer to each other compared to those of water at a gas state (Malenkov, 2009). In the present study, the lower the temperature is, the higher the firmness of cheese, which can be explained by the closer proximity of molecules caused by the low molecular energy at colder temperatures. The same viscoelastic pattern was reported by Bayarri et al. (2012). When evaluating cheese at temperatures of 10 and 22°C, they reported that at lower temperatures, the samples presented higher firmness and rigidity values. The researchers attributed this factor to the formation of fat as a function of the evaluation temperature. By studying the effects of temperature on the viscoelasticity of cream cheeses, Brighenti et al. (2008) reported that the lower the temperature is at the moment of analysis, the higher the firmness of the product. During the maturation period, the colour parameters of Prato cheese (Chroma and H^o) increased with the maturation time (Table 7). Averages in the same row followed by different letters represent significant differences according to Tukey's test at 5% significance. The colour parameters of Prato cheese, Chroma and H^o, increased during the maturation period. The same behaviour was observed by Buffa et al. (2001) in the evaluation of cheese colour during the maturation period. In turn, when evaluating the colour of Asiago cheese during the maturation period, Marchesini et al. (2009) reported the reduction of Chroma values and the increase of H^o. According to Nollet and Toldrá (2010), lipid and protein degradation causes colour changes in products.

The pH decreased and the titratable acidity increased

during the maturation period of Prato cheese; however, the moisture remained constant (Table 8). At the 8th day of storage, Prato cheese presented a pH of 5.30, an acidity of 1.01% of lactic acid, and a moisture of 43.60%. Spadoti et al. (2005) reported values lower than those of the present study in the evaluation of Prato cheese at the 10th day of maturation, with a pH value of 5.12, an acidity of 0.97% lactic acid, and a moisture of 41.70%; the cheese was characterised as average moisture. The relationship between pH and acidity was inversely proportional in the present study. While the pH decreased, acidity increased; the same pattern was observed by Sauer-Leal and Okada (2008), who evaluated the physico-chemical characterisation of Prato cheese by infrared spectroscopy. According to Fox (1993), during cheese maturation, the pH is reduced because lactic acid is produced by microorganisms responsible for fermentation. In turn, by evaluating the effect of pH on Mozzarella cheese, Guine et al. (2002) reported an increase during the storage period. This pH increase is associated with the reduction of the lactate:protein ratio (Fox and Wallace, 1997); when this ratio decreases, curd loses its buffering capability (Czulak et al., 1969).

The moisture remained constant during the maturation period because the thermo-shrinkable packaging prevented moisture loss or gain from the environment. Moisture values higher than those of the present study were reported by De Rensis et al. (2009), who evaluated the physico-chemical, rheological and sensorial characterisation of Prato cheese, with 50.73% moisture. In turn, Roig et al. (2003) reported a moisture of 48.4% for the evaluation of Prato cheese obtained by ultrafiltration. The fat content of Prato cheese at the end of maturation was on average 28.43±0.79% in wet basis

and $50.4 \pm 1.34\%$ fat in the dry extract, which is characterised as fat cheese. By analysing Prato cheese obtained by ultrafiltration, Roig et al. (2003) reported values for fat in wet basis and dry extract similar to those of the present study. Values lower than those of the present study were found by Spadoti et al. (2005), who evaluated Prato cheese at the 10th day of maturation, with fat values of 25.33%. A fat value slightly higher than that of the present study was reported by Sauer-Leal and Okada (2008), who evaluated the physico-chemical characterisation of Prato cheese by infrared spectroscopy, with an average of 28.68%.

The protein content of Prato cheese evaluated 22 days after maturation was on average $24.06 \pm 0.98\%$. A protein content similar to that of the present study was reported by Sauer-Leal and Okada (2008). Values lower than those of the present study were found by Vianna et al. (2008) and Roig et al. (2003), with protein contents of 22.7 and 18.42%, respectively. The ash content of Prato cheese at the end of maturation presented an average of $3.11 \pm 0.67\%$; higher values were reported by Sauer-Leal and Okada (2008) and Alves et al. (2013), who evaluated the physico-chemical characterisation of Prato cheese, with averages of 4.04 and 4.03%, respectively. Evaluating which parameters influence the texture of Prato cheese is of fundamental importance to establishing patterns that meet the expectations of consumers. Knowing that the packaging temperature of Prato cheese influences the texture parameters, it is necessary to determine the ideal packaging temperature to perform a texture analysis of cheese. According to this study, a temperature of 12°C is indicated because at this temperature, the cheese presents good firmness and is within the limits of storage temperatures.

Conclusions

During the maturation period of Prato cheese, a reduction of firmness, gumminess and elasticity was observed; however, cohesiveness and adhesiveness increased. With an increasing temperature of evaluation, Prato cheese had reduced firmness and gumminess, whereas the elasticity, cohesiveness and adhesiveness did not change. The pH of Prato cheese decreased as the maturation stage advanced, acidity increased, and the moisture remained stable. At the end of maturation, Prato cheese was characterised as fat cheese, with protein and ash contents compatible to those of other studies. According to the present study, the temperature at the moment of analysis influences the texture parameters of Prato cheese.

Conflict of interests

The authors did not declare any conflict of interest.

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