Composition of Ragusano Cheese During Aging

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ABSTRACT

Ragusano cheese is a brine-salted pasta filata cheese. Composition changes during 12 mo of aging were determined. Historically, Ragusano cheese has been aged in caves at 14 to 16°C with about 80 to 90% relative humidity. Cheeses (n = 132) included in our study of block-to-block variation were produced by 20 farmhouse cheese makers in the Hyblean plain region of the Province of Ragusa in Sicily. Mean initial cheese block weight was about 14 kg. The freshly formed blocks of cheese before brine salting contained about 45.35% moisture, 25.3% protein, and 25.4% fat, with a pH of 5.25. As result of the brining and aging process, a natural rind forms. After 12 mo of aging, the cheese contained about 33.6% moisture, 29.2% protein, 30.0% fat, and 4.4% salt with a pH of 5.54, but block-to-block variation was large. Both soluble nitrogen content and free fatty acid (FFA) content increased with age. The pH 4.6 acetate buffer and 12% TCA-soluble nitrogen as a percentage of total nitrogen were 16 and 10.7%, respectively, whereas the FFA content was about 643 mg/100 g of cheese at 180 d. Five blocks of cheese were selected at 180 d for a study of variation within block. Composition variation within block was large; the center had higher moisture and lower salt in moisture content than did the outside. Composition variation within blocks favored more proteolysis and softer texture in the center.
(**Key words:** Ragusano, composition, proteolysis, lipolysis)

Abbreviation key: FDB = fat on a dry basis, **HSD** = Tukey's Studentized Range Test Honestly Significant Difference, **PDO** = Protected Denomination of Origin.

INTRODUCTION

In 1996, the European Union gave Ragusano cheese the designation of Protected Denomination of Origin (**PDO**) (9). Ragusano cheese is one of 118 European PDO cheeses (9). The PDO recognition is designed for food products whose qualities derive exclusively from human and natural factors and whose production, processing, and manufacture take place in a defined geographic area (8). This type of recognition is a consequence of a European Union agricultural policy that encourages the diversification and characterization of product in Europe with the objective of obtaining a better market equilibrium, reducing surplus, and stimulating an extensive versus intensive agricultural production system. This strategy links environmental protection (e.g., prevention of erosion) and reduction of sociological problems (e.g., decreased risk of depopulation of less favored rural areas) with the heritage and cultural diversity of traditional agricultural products (e.g., specialty cheese production).

Ragusano cheese is a pasta filata cheese that is made by traditional cheese manufacturing procedures that were recently described (11). Ragusano cheese can be aged up to 12 mo; however, it is common to sell the cheese at 4 to 6 mo as semiaged and at greater than 6 mo as an aged cheese. The objectives of this research were to determine the average initial chemical composition (before brining) of traditionally produced Ragusano cheese and the changes in chemical composition that occur during aging and to determine the variation in chemical composition within a block of cheese induced by brine salting and aging without packaging.

MATERIALS AND METHODS

Cheese Composition During Aging

Twenty farms (Brown Swiss and Modicana cattle breeds) with the best cheese quality were selected for this study from those farms that participated in the previous cheese-making technology survey (11). One hundred thirty-two blocks (ca. 14 kg/block) of cheese were analyzed before brining. Each block of cheese was sampled (d 0) at the end of stretching and just before forming the block. Generally, the cheese blocks were kept in brine at 14 to 16°C for an average of 2 d/kg of cheese. Once the brine salting was completed, the cheese was removed from the brine and sampled. A cheese trier was used to remove four cores from each block of Ragusano cheese. The 1 cm of cheese from the

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cheese trier position nearest the surface of the block was removed from the core sample and placed back in the hole in the block of cheese. The hole was sealed with paraffin before the cheese was returned to the aging room. The cheese was aged in a ventilated room at 14 to 16°C with a humidity of about 80 to 90%. Two blocks of cheese were tied together on opposite ends of a rope and then hung in pairs over a beam to allow air circulation during 12 mo of aging. The cheese was sampled seven times during 12 mo of aging.

All analyses were done in duplicate. Cheese analyses were as follows: 24 h of forced-air oven drying at 100°C for total solids (12), total nitrogen by Kjeldahl using a 1-g sample size [(1); AOAC method 33.2.11, 991.20] with conversion to protein content by using a factor of 6.38, and salt by the Volhard method (12).

The fat content of the cheeses was determined by the Gerber method using a butyrometer with a calibrated range from 0 to 40% fat. Three grams of grated cheese were weighed into a small, perforated, glass sample holder that was attached to a rubber stopper. The stopper with the glass sample holder plus the cheese was inserted into the bottom end of the butyrometer to close the tube. Next, enough sulfuric acid (density 1.5) was added through the opening at the top of the butyrometer tube to cover glass sample holder that contained the cheese. The butyrometer containing the sample plus sulfuric acid was placed into a water bath at 65°C and mixed periodically to achieve complete dissolution of the cheese. In a hood, 1 ml of amyl alcohol (density 0.815) was added, and then sulfuric acid was added until the level of the sulfuric acid reached the 30% mark on the calibrated portion of the neck. A rubber stopper was inserted into the top of the butyrometer tube, and the tube was inverted four or five times to achieve complete mixing. The butyrometers were placed in a heated centrifuge for 15 min at 700 rpm. After centrifugation the butyrometers were placed in a 65°C water bath for 2 or 3 min before reading the percentage fat from the calibrated portion of the butyrometer.

During cheese aging, pH 4.6 acetate buffer- and 12% TCA-soluble nitrogen were determined (4) and expressed as a percentage of the total nitrogen content of the cheese measured at each stage of aging. Proteolysis during aging may contribute to both texture and flavor development in Ragusano cheese.

The statistical analyses of the change in composition (y) with (x) days of aging were done for the 132 blocks of cheese from the 20 farms by using two models: one linear equation, $y = a + bx$, for the pH parameter and one exponetial equation, $y = a + be^{-x/c}$, for the other parameters (16). Figures are shown with solid lines representing the best fit equation, with the dotted lines representing 95% confidence intervals (i.e., the interval within which the true regression line exists with 95% confidence), and the line of boxes representing 95% prediction intervals (i.e., interval within which 95% of future individual block observations would be expected).

A group of 10 blocks was selected from the 132 blocks of Ragusano cheese. They were sampled at 90 and 180 d of aging for FFA content. The quantity of total FFA and individual FFA were determined using a resin binding technique to remove FFA from an acidic ether extract of the cheese, followed by quantitative GLC analysis (10). Changes in FFA content of Ragusano cheese may be related to flavor development.

Composition Variation Within Block

Five blocks of Ragusano cheese made using traditional cheese-making procedures (11) that had been aged for 180 d at a commercial aging center were se-

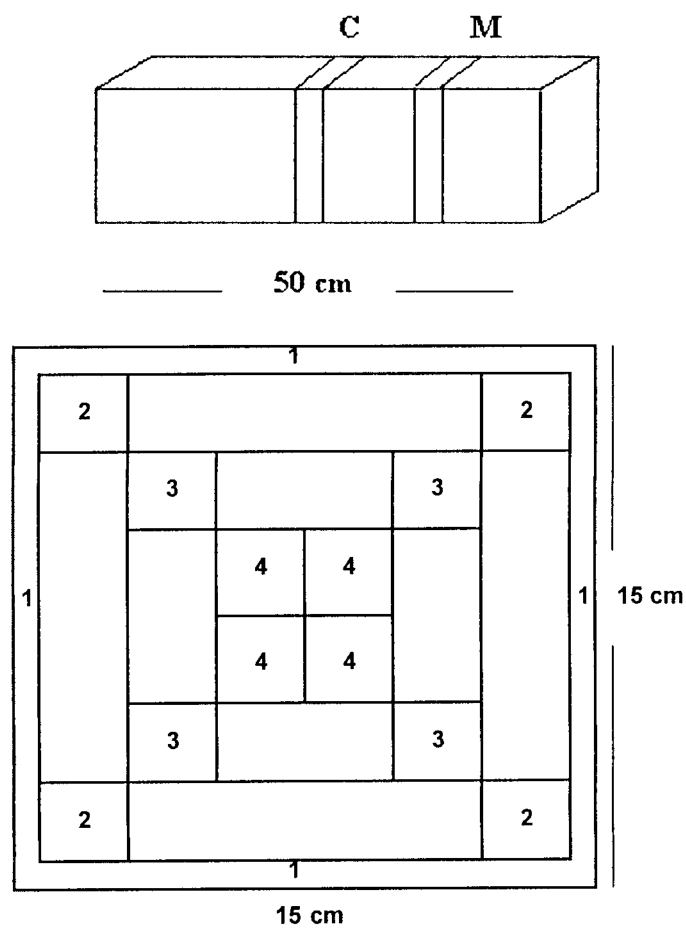

Figure 1. a) Position of center (C) and middle (M) slices within block; b) sampling pattern used within the center and middle slices of cheese taken from blocks of Ragusano cheese. Numbers for Positions 1, 2, 3, and 4 represent the area of the slice where the cheese sample was collected.

Table 1. Mean cheese composition during aging of Ragusano cheese.

	Months							
Component	0	1–2	2–3	3–4	4–6	6–8	8–10	10–12
Moisture, %	45.35	40.39	38.34	36.32	36.74	34.61	33.33	33.58
Fat, %	25.45	27.23	28.17	28.51	28.45	29.02	29.24	29.97
Protein, %	25.30	26.45	27.16	27.76	27.85	28.33	29.10	29.24
Salt, %	0.10	2.34	2.99	3.61	3.73	4.31	4.59	4.44
pH	5.25	5.32	5.16	5.21	5.25	5.34	5.29	5.54
FDB,[1] %	46.29	45.68	45.68	44.77	44.97	44.38	43.86	45.12
S/M,[2] %	0.23	5.88	7.94	9.98	10.34	12.48	13.76	13.31
pH 4.6/TN,[3] %	3.19	9.80	13.29	14.69	16.03	18.64	21.72	23.38
12% TCA/TN,[4] %	0.71	5.44	7.67	9.44	10.68	13.69	17.16	17.37

[1]FDB = fat on a dry basis.

[2]S/M = ((salt content)/(moisture content)) × 100.

[3]pH 4.6/TN = pH 4.6 acetate buffer-soluble nitrogen as a percentage of total nitrogen.

[4]12% TCA/TN = 12% TCA-soluble nitrogen as a percentage of total nitrogen.

lected for sampling. The sampling of each rectangular block of cheese was done by removing a 5-cm thick slice from the center of the block and a second 5-cm thick slice taken halfway between the center and one end of each block. The locations of the slices and the samples from within each slice are shown in Figure 1a. Composition data are reported for positions 1, 2, 3, and 4 within each slice, as identified in Figure 1b. Position 1, 2, 3, and 4 represent 5.3, 52.7, 31.5, and 10.5%, respectively, of the weight of cheese within a slice. The mean composition of a slice was calculated using these weighting factors for position within slice. The methods of analyses were as described above for moisture, fat, protein, salt, and soluble nitrogen fractions. Degradation of α_s- and β-CN was determined using urea-PAGE (5, 6). Calcium was determined using an atomic absorption method (3).

The GLM procedure within SAS (15) was used to determine whether there were differences in composition between and within slices of cheese removed from blocks of Ragusano cheese. The Tukey's Studentized Range Test Honestly Significant Difference (**HSD**) was used to determine whether differences ($P \leq 0.05$) existed between weighted mean composition of the center versus the middle slice and to determine whether there were differences ($P \leq 0.05$) in composition within slice. The Tukey's HSD test is more conservative than Fishers LSD, so that it is more difficult to reject the null hypothesis (13, 14).

RESULTS AND DISCUSSION

Cheese Composition During Aging

The average composition of the cheese at d 0 and at various times during 12 mo of aging is shown in Table 1. The global variation in these components is shown in Figures 2 to 8. At the end of cheese making, the moisture content was about 45%. The salt content of the cheese was very low at time 0 (i.e., 0.10%) prior to brine salting. Because Ragusano cheese is brine salted and aged without packaging, the blocks of cheese form a rind and lose moisture during 12 mo of aging with a final moisture of about 33.6%. This pattern of change in average moisture content of a block of Ragusano cheese during aging is similar to that reported for Parmigiano Reggiano during ripening (2).

The decrease of about 11.8% moisture during 12 mo of aging caused observed increases in fat, protein, and salt content on a wet basis (Table 1). However, the fat on a dry basis (**FDB**) remained relatively constant

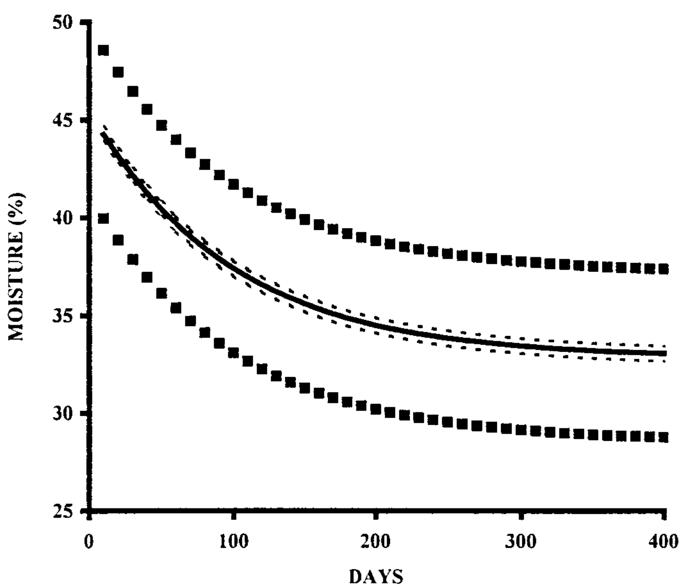

Figure 2. Best fit regression $y = a + be^{-x/c}$ (solid line), 95% confidence interval (dotted line ± 0.385), and prediction interval (line of boxes ± 4.23) for change in moisture content of Ragusano cheese during aging. a = 32.90, b = 12.58, and c = 97.10; $r^2 = 0.836$.

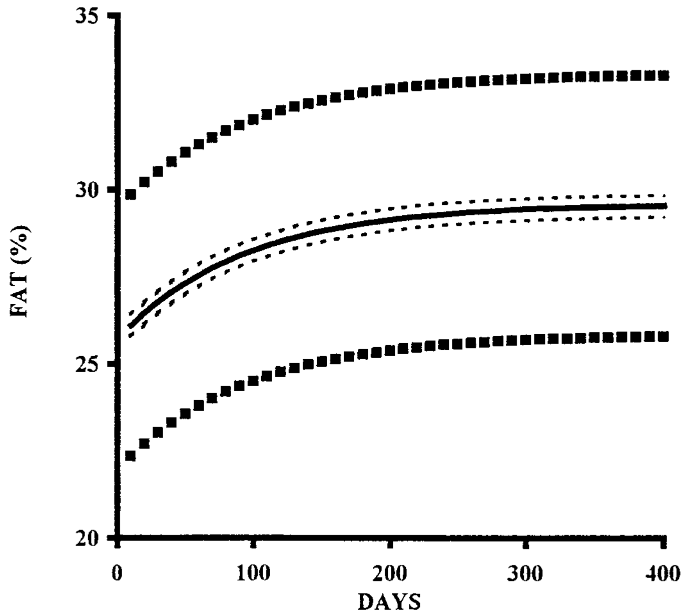

Figure 3. Best fit regression y = a + be$^{-x/c}$ (solid line), 95% confidence interval (dotted line ± 0.313), and prediction interval (line of boxes ± 3.75) for change in fat content of Ragusano cheese during aging. a = 29.62, b = -3.91, and c = 95.00; r^2 = 0.34.

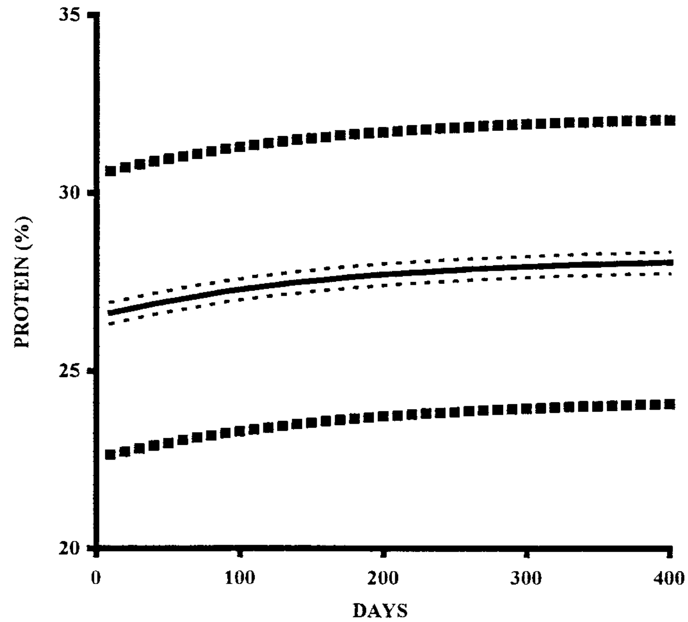

Figure 4. Best fit regression y = a + be$^{-x/c}$ (solid line), 95% confidence interval (dotted line ± 0.30), and prediction interval (line of boxes ± 4.0) for change in protein content of Ragusano cheese during aging. a = 28.24, b = –1.72, and c = 168.51; r^2 = 0.06.

during the 12 mo of aging (Table 1). A slight downward trend in FDB may be caused by a small amount of fat loss from the surface of the block as it shrinks in size due to the moisture decrease. The salt content of Ragusano cheese was high because the managers of commercial cheese aging centers know that high salt content will reduce the risk and frequency of development of quality defects (e.g., gas production). The pH of the cheese starts out about 5.25 before brine salting and remains relatively stable during aging.

Because the nature of the small-scale cheese making with traditional methods (11), the variation in composition from batch to batch within and between farms was large. Figures 2, 3, 4, and 5 demonstrate the temporal changes and degree of variation in moisture, fat, protein, and salt as a function of aging time. The rate of change of general composition was faster during the first 150 d of aging and then slowed after that time. The pH of the cheese was relatively stable with time of aging, but there was a high degree of variation from batch to batch and farm to farm (Figure 6). The mean salt in the moisture content increased from about 5.9% after brining to an average of 13.3% at 10 to 12 mo of aging (Table 1). This relatively high salt content would be expected to slow down or stop proteolysis during aging.

Lipolysis and Proteolysis During Aging

Rennet was prepared at the farms from a goat or sheep stomach and was used to coagulate the milk for Ragusano cheese making. These rennets contained lipase activity, which may contribute to release of FFA from triglycerides during cheese aging. It is thought

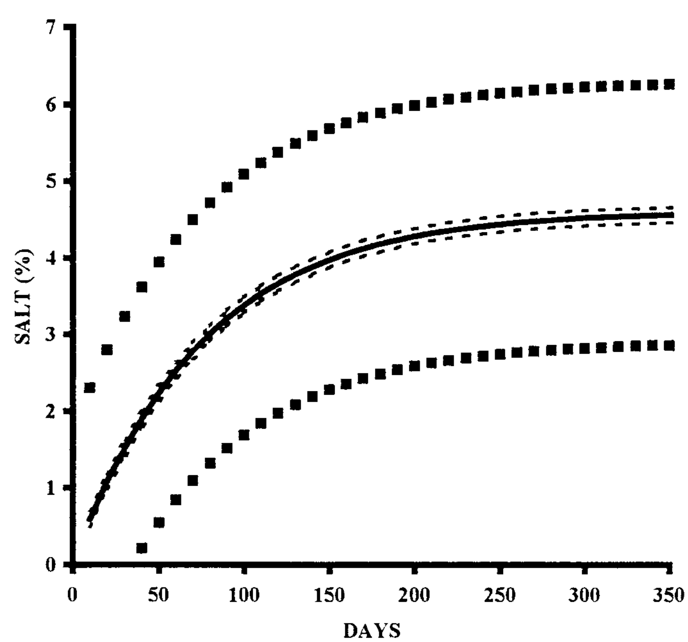

Figure 5. Best fit regression y = a + be$^{-x/c}$ (solid line), 95% confidence interval (dotted line ± 0.1%), and prediction interval (line of boxes ± 1.7%) for change in salt content of Ragusano cheese during aging. a = 4.62, b = –4.58, and c = 76.03; r^2 = 0.81.

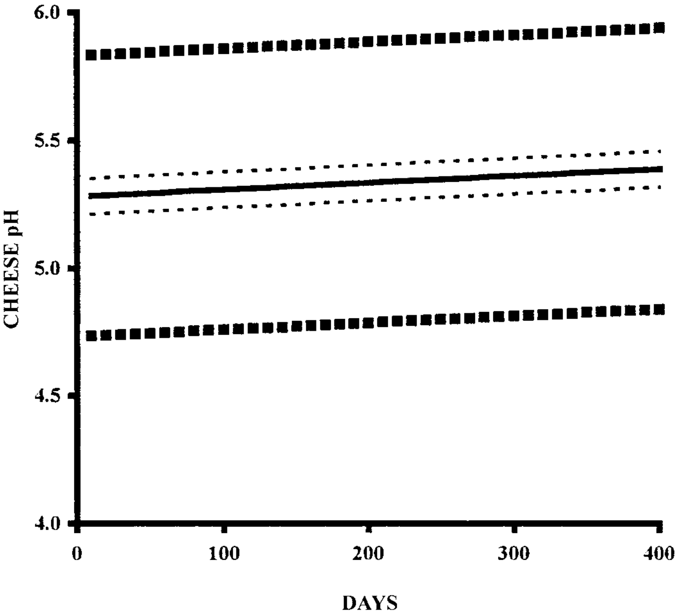

Figure 6. Best fit regression equation y = a + bx (solid line), 95% confidence interval (dotted line ± 0.03), and prediction interval (line of boxes ± 0.57) for change in pH of Ragusano cheese during aging. a = 5.28, and b = 0.0002; r^2 = 0.010.

that FFA make a significant contribution to the typical flavor characteristics of Ragusano cheese. Ten blocks of cheese out of the 132 were randomly selected for determination of FFA content at 90 and 180 d of aging. The mean FFA content of the 10 blocks of cheese was 312.9 mg/100 g at 90 d and 642.8 mg/100 g of cheese at 180 d of age (Table 2). The average FFA content of another aged Italian pasta filata cheese, Provolone, has been reported to be 635 mg/100 g of cheese (7). Fatty acids with a chain length of 12 carbons or less made up 36% of the FFA present in the 180-d-old cheese (Table 2). In a report on the mean FFA composition in Provolone cheese (7), the relative percentages of individual fatty acids reported are similar (i.e., C_4 to C_{12} equal to 35.5% of total FFA) to those in Table 2 for 180-d-old Ragusano cheese. In future work, it will be useful to determine whether there is a gradient of FFA content from the surface to the interior of a block of cheese.

Soluble nitrogen content of cheese expressed as a percentage of total nitrogen was used as an index of proteolysis during aging. The nitrogen soluble in pH 4.6 acetate buffer reflects the action of the coagulant producing medium molecular weight peptides during cheese aging, and the amount of 12% TCA-soluble nitrogen reflects the action of starter culture peptidases producing low molecular weight peptides during aging. The rate of increase in soluble nitrogen content of the cheeses with time of aging (Table 1, Figures 7 and 8) did not show as much leveling off after 150 d of aging as the general composition did. However, the global variation in soluble nitrogen was large (Figures 7 and 8).

It may seem surprising that the pH 4.6 and 12% TCA-soluble nitrogen both continued to increase after 4 mo of aging even though the mean concentration of salt in the moisture phase was 10% and increasing. At this point it is important to remember that composition data in Table 1 represent the average of the block and that there could be systematic variation of composition within every block of cheese. The moisture content of the cheese at the outside surface (i.e., hard rind) of the block was expected to be much lower than the average for the block, and the moisture content of the cheese in the soft center of the block was expected to be much higher than average for the block. These results would produce a gradient of salt in the moisture within each block of cheese, which might allow proteolysis to con-

Table 2. Mean FFA content (mg/100 g) of Ragusano cheese and relative FFA composition after 90 and 180 d of aging.

	90 days			180 days		
FFA	Mean	SD	Relative percentage	Mean	SD	Relative percentage
C_4	68.2	20.4	21.8	109.3	32.3	17.0
C_6	21.8	1.4	7.0	43.7	6.3	6.8
C_8	7.1	0.3	2.3	16.1	2.5	2.5
C_{10}	15.0	0.9	4.8	30.9	5.3	4.8
C_{12}	15.3	1.1	4.9	31.7	7.7	4.9
C_{14}	25.6	1.2	8.2	59.7	20	9.3
$C_{14:1}$	4.7	0.4	1.5	12.3	3.1	1.9
C_{16}	49.5	3.5	15.8	128.5	51.5	20.0
$C_{16:1}$	11.8	5.1	3.8	9.6	4.1	1.5
C_{18}	15.3	1.4	4.9	37.1	15.1	5.8
$C_{18:1}$	51.3	7.5	16.4	123.2	56.3	19.2
$C_{18:2}$	26.9	4.8	8.6	40.7	15.9	6.3
Total	312.9	34.1	100	642.8	192.7	100

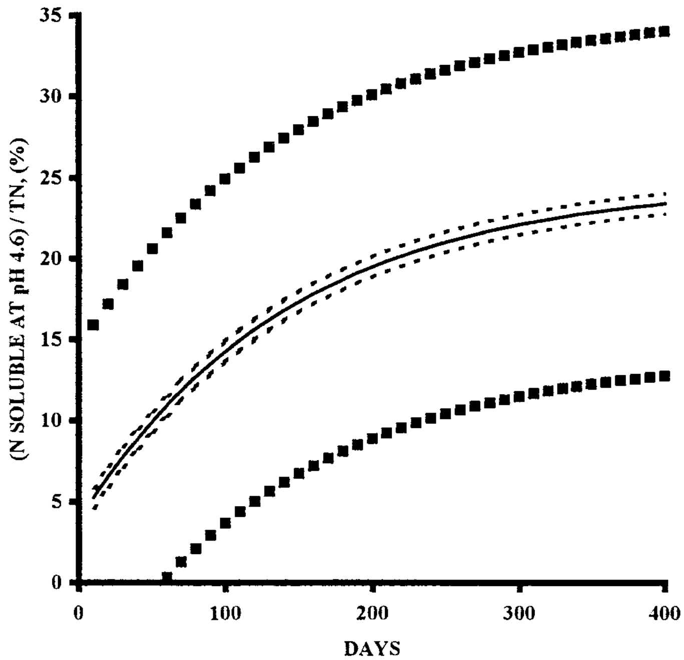

Figure 7. Best fit regression y = a + be$^{-x/c}$ (solid line), 95% confidence interval (dotted line ± 0.625), and prediction interval (line of boxes ± 10.62) for change in pH 4.6 acetate buffer-soluble nitrogen as a percentage of total nitrogen content of Ragusano cheese during aging. a = 24.71, b = –20.86, and c = 144.44; r^2 = 0.61.

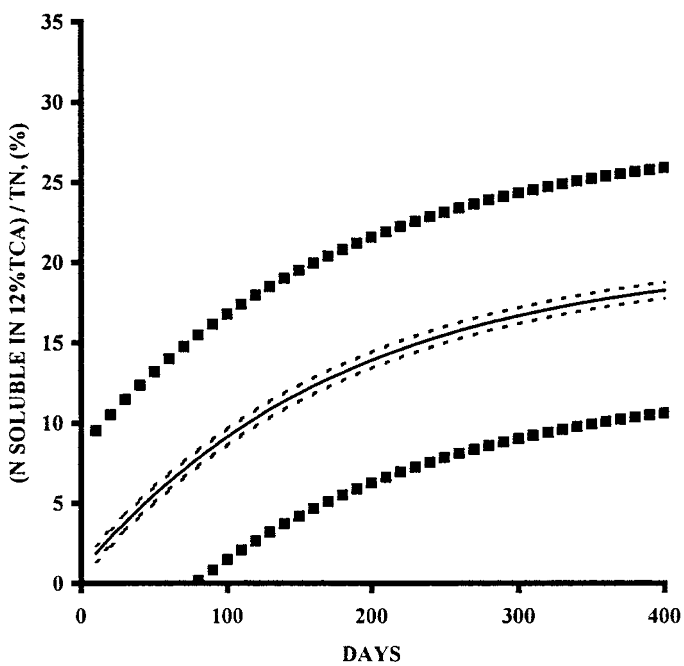

Figure 8. Best fit regression y = a + be$^{-x/c}$ (solid line), 95% confidence interval (dotted line ± 0.50), and prediction interval (line of boxes ± 7.65) for change in 12% TCA-soluble nitrogen as a percentage of total nitrogen content of Ragusano cheese during aging. a = 20.52, b = –19.70 c = 182.41; r^2 = 0.66.

Table 3. Comparison of mean values for composition of the center and middle slice taken from blocks (n = 5) of Ragusano cheese at 180 d.

	Slice			
	Center	Middle	Significance	HSD[1]
Component, %				
Moisture	33.59	31.92	P < 0.050	1.67
Fat	33.48	34.09	NS	2.75
Protein	28.33	28.73	NS	0.97
Salt	3.37	4.33	P < 0.022	0.78
pH 4.6/TN[2]	11.91	9.56	P < 0.040	2.21
12% TCA/TN[3]	8.34	6.41	P < 0.001	0.90
NaCl/M[4]	10.19	13.81	P < 0.023	2.98
NaCl/DM[5]	5.09	6.39	P < 0.019	1.03
FDB[6]	50.40	50.05	NS	3.58
PDB[7]	42.65	42.20	NS	1.71
Ca	0.99	0.84	P < 0.001	0.04
Ca/TN[8]	3.50	2.95	P < 0.001	0.16

[1]HSD = Tukey's Studentized Range Test Honestly Significant Difference at α = 0.05.

[2]pH 4.6/TN = pH 4.6 acetate buffer-soluble nitrogen as a percentage of total nitrogen.

[3]12% TCA/TN = 12% TCA-soluble nitrogen as a percentage of total nitrogen.

[4]NaCl/M = ((sodium chloride)/(moisture content)) × 100.

[5]NaCl/DM = ((sodium chloride)/(dry matter)) × 100.

[6]FDB = fat on a dry basis.

[7]PDB = protein on a dry basis.

[8]Ca/TN = calcium as a percentage of total nitrogen.

tinue in the center of the block, while proteolysis near the outside surface of the block stopped.

Composition Variation Within Block

Data for composition variation within blocks of cheese after 180 d of aging are shown in Tables 3 and 4. The data are presented separately for between slice variation (Table 3) and within slice variation (Table 4). The center slice contained significantly more moisture, pH 4.6 acetate- and 12% TCA-soluble nitrogen, calcium, calcium as a percentage of protein, and lower salt and salt in the moisture than the middle slice (Table 3). Thus, the cheese would be expected to decrease in moisture, increase in salt, decrease in proteolysis, and increase in firmness as one moves from the center slice to either end of the block of cheese.

The magnitude of systematic differences in composition within slice of cheese was much larger than between slice variations. There was approximately a 17% lower moisture content (Table 4) at the outside surface (i.e., P1) than in the center (i.e., P4) of the blocks of cheese after 180 d of aging. As a result, the concentration of fat and protein on a wet basis decreased from the outside surfaces to the center of each block of cheese. However, FDB and protein on a dry basis were calculated, and it was found that the protein on a dry basis

Table 4. Variation in composition (%) of Ragusano cheese at 180 d at four positions (i.e., P1, P2, P3, P4) within a 5-cm thick slice taken from the center of the block (n = 5) or the slice at half of the distance from the center to one end of the block (n = 5).

		Center					Middle				
		P1	P2	P3	P4	HSD[1]	P1	P2	P3	P4	HSD
Moisture	Mean	21.92^{c}	31.30^{b}	37.62^{a}	38.92^{a}	2.32	21.09^{c}	29.33^{b}	36.17^{a}	37.67^{a}	2.44
Fat	Mean	39.5^{a}	34.80^{b}	31.15^{c}	30.80^{c}	3.22	38.80^{a}	35.30^{ab}	32.30^{b}	31.00^{b}	5.78
Protein	Mean	34.76^{a}	29.37^{b}	26.31^{c}	25.97^{c}	1.90	34.37^{a}	29.83^{b}	26.82^{c}	26.15^{c}	1.81
Salt	Mean	2.31^{b}	3.48^{a}	3.47^{a}	3.01^{ab}	0.96	3.29^{b}	4.41^{ab}	4.47^{a}	4.05^{ab}	1.14
pH 4.6/TN[2]	Mean	10.81^{b}	9.55^{b}	14.58^{a}	16.28^{a}	4.05	7.11^{b}	7.01^{b}	12.52^{a}	14.76^{a}	4.09
12% TCA/TN[3]	Mean	7.91^{bc}	6.60^{c}	10.53^{ab}	10.73^{a}	2.80	4.42^{c}	4.86^{c}	8.18^{b}	9.93^{a}	1.97
NaCl/M[4]	Mean	10.53^{a}	11.19^{a}	9.28^{a}	7.75^{a}	NS	15.58^{a}	15.07^{a}	12.40^{ab}	10.80^{b}	4.25
NaCl/DM[5]	Mean	2.97^{b}	5.06^{a}	5.55^{a}	4.91^{a}	1.29	4.18^{b}	6.23^{a}	6.99^{a}	6.48^{a}	1.55
FDB[6]	Mean	50.60^{a}	50.65^{a}	49.95^{a}	50.44^{a}	NS	49.18^{a}	49.93^{a}	50.51^{a}	49.70^{a}	NS
PDB[7]	Mean	44.50^{a}	42.76^{ab}	42.20^{b}	42.52^{ab}	2.17	43.55^{b}	42.21^{cd}	42.04^{d}	41.98^{d}	NS
Calcium	Mean	1.01^{b}	1.09^{a}	0.88^{c}	0.82^{c}	0.08	0.83^{a}	0.85^{a}	0.84^{a}	0.81^{a}	NS
Ca/TN[8]	Mean	2.90^{c}	3.72^{a}	3.34^{b}	3.16^{bc}	0.30	2.42^{c}	2.85^{b}	3.14^{a}	3.11^{a}	0.25

[a,b,c]Means within the same row within slice not sharing a common superscript are different ($P < 0.05$).

[1]HSD = Tukey's Studentized Range Test at Honestly Significant Difference at $\alpha = 0.05$.

[2]pH 4.6/TN = pH 4.6 acetate buffer-soluble nitrogen as a percentage of total nitrogen.

[3]12% TCA/TN = 12% TCA-soluble nitrogen as a percentage of total nitrogen.

[4]NaCl/M = ((sodium chloride)/(moisture content)) × 100.

[5]NaCl/DM = ((sodium chloride)/(dry matter)) × 100.

[6]FDB = fat on a dry basis.

[7]PDB = protein on a dry basis.

[8]Ca/TN = calcium as a percentage of total nitrogen.

and FDB were much more constant from the center to the outside surface of a block of Ragusano cheese than fat or protein on a wet basis. Thus, most of the systematic variation in fat and protein on a wet basis within slice was due to moisture variation. The salt concentration on a wet basis and on a dry basis were slightly lower at the exterior surface (i.e., P1) and the center (i.e., P4) of each slice but were relatively uniform across Positions 2 to 3 within the block (Table 4). The mean salt content in the middle slice from a block of cheese was higher than the center slice (Table 3) because the middle slice took up salt from all four side faces of the block plus the end face of the block. There was a significant ($P < 0.05$) difference in salt concentration in the moisture phase of the cheese within the middle slice, and the same trend ($P < 0.07$) was observed in the center slice (Table 4). The salt in the moisture phase of the cheese at the outside surfaces of the slice was higher, whereas in the center of the slice it was lower. Both pH 4.6 acetate buffer- and 12% TCA-soluble nitrogen had the opposite trend; more soluble nitrogen was in the center, and less soluble nitrogen was on the outside position of a slice. The high salt concentration in the aqueous phase of the cheese near the surface of the block would be expected to inhibit proteolysis. Similar results were found using urea-PAGE (Figure 9). There was less proteolysis in total for Position 1 for both slices, and the amount of degradation of both α_s- and β-CN was greater in Position 4 (i.e., the center) of each slice. This finding is consistent with the observation that the outside surfaces of the blocks of cheese were hard, whereas the cheese in the center was soft.

CONCLUSIONS

The mean moisture content of Ragusano cheese decreased from about 45.3 to 33.5% during 12 mo of aging. As a result of this moisture decrease, the concentration

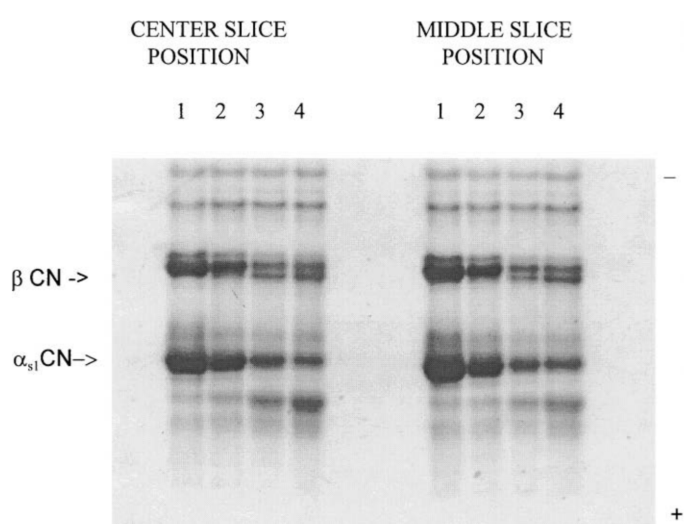

Figure 9. Urea-PAGE gel of cheese samples taken from Positions 1, 2, 3, and 4 of the center and middle slices of Ragusano cheese after 180 d of aging.

of other milk components increased proportionately. The mean salt in the moisture content increased from about 0.23% before brine salting to 13.3% after 12 mo of aging. The pH 4.6 acetate buffer- and 12% TCA-soluble nitrogen as a percentage of total nitrogen increased from 3.19 to 23.4% and from 0.71 to 17.4%, respectively, during 12 mo of aging. The mean FFA content of the cheese at 180 d was 642.8 mg/100 g of cheese. Within-block variation was very large; moisture was approximately 17% higher in the center of each block than at the outside surface. This large range of moisture produced a very large difference (up to 4.8%) in the salt in moisture concentration from the center to the outside surface of a block within slice and favored more proteolysis in the center of the block. It is not clear whether this large variation in composition within slice is produced mostly by moisture loss during aging after brining or by moisture loss during the brining process. Understanding the time sequence of development of these differences and the factors that influence them will provide a knowledge base that may allow development of strategies to improve the consistency of cheese composition and quality of brine-salted cheeses.

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