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The Effect of Salt Concentration on Some Characteristics in Herby Cheese

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Abstract: Herby cheese is a salted traditional cheese manufactured from sheep's and cow's milk in the Eastern and South-eastern of Turkey. The aim of this study was to assess the influence of different salt concentrations (4, 5 and 6%) on the ripening characteristics of Herby cheese. Pasteurized whole cow's milk (3.7% fat) was used for Herby cheese manufacture. The cheese samples were ripened under soil at 7±1°C for 90 days. The samples were characterized in terms of microbiological, chemical and sensory properties. The ripening time had a significant ($P<0.05$) effect on all parameters except for protein, fat, total aerobic bacteria (TAB), appearance and colour, body and texture, and flavour. In addition, salt concentration had a significant influence on dry matter, salt, titratable acidity, micrococci and staphylococci, proteolytic bacteria, yeasts and moulds, appearance and colour, and saltiness. As a result, we concluded that increasing the salt concentration in cheese samples contributed to reducing the number of microorganisms and increased the acceptability in terms of appearance and colour, and body and texture from sensory properties.

Key words: Herby cheese, salt concentration, ripening

Introduction

Herby cheese, a semi-hard, salty and herb added, is manufactured in small family businesses for their needs and commercial purposes from raw sheep's and cow's milk between May and June in the Eastern and South-eastern of Turkey.

Milk used in manufacture of Herby cheese is coagulated with rennet and added different herbs (*Allium* sp., *Ferula* sp., etc) in curd. The cheeses are ripened in a cool place without temperature control because cheese samples were generally stored under soil in containers throughout ripening. Since the production method of Herby cheese is not standardized, it is availability different quality cheese in the markets.

The purpose of salt added in cheese is to control the indigenous microflora, particularly pathogens. However, salt has a major influence on acid development and rennet coagulation. In order to avoid use of excessive salt, pasteurization has been recommended for eliminating pathogenic bacteria (Guinee and Fox, 1993). Salting of curd is an important step in the manufacture of most cheese varieties. Salt fulfills many important functions in cheese: it contributes directly to cheese flavour, it controls the growth of starter and nonstarter bacteria, it regulates the activity of rennet and other enzymes, and it promotes curd syneresis (Morris *et al*., 1985).

Several aspects of Herby cheese have already been studied (Coskun, 1998; Coskun and Tuncturk, 2000; Tarakci and Akyuz, 2001), but there is no study about the effect of salt concentrations during ripening. Thus, the aim of this study was to determine the influence of different salt ratios on microbiological, chemical and sensorial characteristics of Herby cheese.

Materials and Methods

Whole cow's milk (3.7% fat) was supplied from Van Agriculture High School. Commercial animal rennet was obtained from Mayasan Company, Istanbul. Herbs known as "sirmo" (*Allium* sp.) in region were obtained from Van cheese shopping district. Yogurt culture (YC-180) obtained from Christina Hansen's Company (Denmark) was used as starter culture (*Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus*).

Herby cheesemaking and sampling: Cheese samples were manufactured at Food Hygiene and Technology Laboratory of Veterinary Faculty of Yuzuncu Yil University. Raw cow's milk was pasteurized at 65°C for 30 min and cooled to 32°C. Starter culture was added at the level of 1% and held for 60 min. Then, milk was coagulated with rennet for 75 min. After coagulation, curd was cut into 8-10 mm cubes with a wire knife and then sprinkled with herb at the ratio of 2% of milk weight. The herbs were thoroughly mixed into the curds. After the curd was pressed for 90 min, it was divided into three parts. Each part of curd was salted at the levels of 4, 5, and 6% (C1, C2 and C3). After salting, the curd was held for an additional 48 h. At the end of this time, cheeses were

Table 1: Chemical properties of cheese samples during ripening

		Ripening time (day)				
Property	Cheese	2	15	30	60	90
Dry	C1	44.22±0.18^{B,b}	45.54±0.39^{B,b}	45.66±0.49^{B}	47.47±0.52^{A}	47.55±0.76^{A}
matter	C2	45.55±0.37^{a}	46.16±0.64^{b}	47.27±0.97	47.13±0.50	48.36±0.23
(%)	C3	46.00±0.66^{C,a}	48.01±0.27^{B,a}	48.11±0.22^{B}	48.19±0.21^{B}	50.28±0.58^{A}
Protein	C1	18.44±0.09	18.73±0.32	18.54±0.13	19.56±0.40	19.27±0.36
(%)	C2	18.79±0.23	18.63±0.07	18.98±0.49	18.69±0.27	19.56±0.32
	C3	18.54±0.05	19.46±0.45	19.21±0.18	19.27±0.54	20.13±0.31
Fat (%)	C1	20.25±0.35	20.50±0.00	20.50±0.00	21.25±0.35	21.25±0.35
	C2	20.50±0.00	20.75±0.35	21.00±0.70	20.75±0.35	21.25±0.35
	C3	20.25±0.35	21.25±0.35	21.00±0.00	21.25±0.35	21.75±0.35
Salt (%)	C1	3.99±0.08^{C,c}	4.62±0.08^{B,c}	4.98±0.18^{A,c}	4.99±0.06^{A,b}	5.10±0.08^{A,b}
	C2	4.84±0.05^{D,b}	5.41±0.04^{C,b}	5.86±0.13^{B,b}	5.98±0.18^{B,a}	6.59±0.20^{A,a}
	C3	5.88±0.04^{C,a}	6.47±0.21^{B,a}	6.67±0.09^{AB,a}	6.74±0.23^{AB,a}	6.98±0.06^{A,a}
Titratable	C1	0.74±0.01^{D}	0.88±0.01^{C,a}	1.06±0.02^{B}	1.19±0.02^{A}	1.09±0.02^{B,a}
acidity	C2	0.77±0.03^{C}	0.85±0.01^{C,b}	1.01±0.03^{B}	1.19±0.02^{A}	0.95±0.01^{B,b}
(LA%)	C3	0.76±0.01^{D}	0.83±0.01^{C,b}	0.92±0.01^{B}	1.17±0.03^{A}	0.96±0.02^{B,b}
WSN/TN	C1	14.88±1.54^{D}	21.64±0.36^{C}	23.58±0.07^{BC}	26.44±1.47^{B}	30.96±0.59^{A}
(%)	C2	15.11±0.42^{E}	19.52±0.58^{D}	22.02±0.34^{C}	26.46±0.63^{B}	28.55±0.69^{A}
	C3	16.01±0.69^{E}	19.34±0.01^{D}	21.10±0.43^{C}	24.85±0.70^{B}	26.95±0.87^{A}
TCA-SN/TN	C1	6.75±0.21^{C}	11.59±0.19^{B}	12.05±0.58^{B}	12.90±0.50^{AB}	13.92±0.73^{A}
(%)	C2	6.45±0.40^{D}	10.96±0.43^{C}	11.78±0.78^{BC}	13.15±0.69^{A}	12.89±0.02^{AB}
	C3	6.72±0.72^{C}	10.50±0.25^{B}	10.89±0.47^{B}	11.92±0.13^{A}	12.37±0.19^{A}
PTA-SN/TN	C1	2.08±0.50^{B}	3.75±0.42^{A}	4.25±0.55^{A}	4.48±0.04^{A}	4.48±0.33^{A}
(%)	C2	2.37±0.45^{C}	3.26±0.26^{BC}	4.37±0.36^{A}	4.27±0.18^{AB}	4.24±0.40^{AB}
	C3	2.24±0.24^{B}	3.29±0.54^{A}	4.15±0.19^{A}	3.98±0.11^{A}	3.97±0.16^{A}
Lipolysis	C1	0.62±0.01^{E}	1.17±0.03^{D}	1.74±0.03^{C}	2.23±0.04^{B}	2.62±0.06^{A}
(ADV)	C2	0.62±0.04^{E}	1.12±0.04^{D}	1.69±0.01^{C}	2.20±0.03^{B}	2.66±0.04^{A}
	C3	0.64±0.01^{E}	1.08±0.04^{D}	1.70±0.01^{C}	2.17±0.06^{B}	2.60±0.04^{A}

C1, C2, C3 Salting with 4, 5, 6%, respectively. ^{ABCDE}Different letters indicate P< 0.05 between ripening times. ^{abc}Different letters indicate P<0.05 between salt concentration of cheese

well packed in plastic bags. The samples were ripened under soil at 7±1EC for 90 days. Two batches of cheeses from different lots of milk were used for this study. Samples were taken from two batches of Herby cheese after ripening for 2, 15, 30, 60, and 90 days.

Chemical analysis: Cheese samples were analyzed for dry matter, protein (Kjeldahl method), fat (Gerber method), salt (Mohr method) and titratable acidity as lactic acid (LA%) were determined according to the methods described by Case *et al.* (1985). Water-soluble nitrogen (WSN) was determined by the Kjeldahl method as described follows; 10 g sample was homogenized with 100 ml distilled water and filtered. The nitrogen content of the extracted cheese was expressed as a percentage of total nitrogen (WSN/TN, %), which was described as a ripening index. Trichloroacetic acid-soluble nitrogen (TCA-SN) was determined in the same cheese extract described above. 10 ml 24% TCA was added to 10 ml cheese extract and after mixing was incubated at 4°C for 2 h. Then, precipitate was filtered through Whatman no 40 paper. Filtrate of the nitrogen was determined according to the Kjeldahl method and TCA-SN was expressed as a percentage of total nitrogen (TCA-SN/TN, %). Water-soluble extract (10 ml) was mixed with 7 ml 3.95 M H_2SO_4 and 3 ml 33% (w/v) phosphotungstic acid (PTA). The mixture was held at 4°C for 12 h and then filtered through Whatman no 40 paper. Filtrate of the nitrogen was determined according to the Kjeldahl method and PTA-SN was expressed as a percentage of total nitrogen (PTA-SN/TN, %) (Kamaly *et al*., 1989; Butikofer *et al*., 1993). Lipolysis as acid degree value (ADV) was measured according to the method described by Case *et al*. (1985). Each measurement was done in duplicate and results reported were average values of the two batches.

Microbiological analysis: Representative 10 g cheese samples were homogenized with 90 ml of sterile 2% (w/v) sodium citrate solution at 45°C in a Stomacher Blender (2300/400, Barcelona, Spain) for 2 min. Decimal dilutions were prepared in 0.1% sterile peptone water and plated in duplicate. Total aerobic bacteria (TAB) counts were made on Plate Count Agar (PCA, Oxoid) at

Table 2: Microbiological properties of cheese samples during ripening (log cfu/g)

		Ripening time (day)				
Property	Cheese	2	15	30	60	90
TAB	C1	6.80±0.21	7.75±0.52	7.86±0.06	7.69±0.13	6.63±0.32
	C2	6.57±0.04	7.61±0.18	7.13±0.92	7.25±0.42	6.39±0.44
	C3	6.59±0.13	7.53±0.33	6.92±1.16	7.20±0.28	5.97±1.04
Psychrotrophics	C1	$5.89±0.06^{B}$	$7.16±0.41^{A}$	$6.16±0.04^{B}$	$4.73±0.18^{C}$	$4.54±0.34^{C}$
	C2	$5.72±0.08^{B}$	$7.57±0.04^{A}$	$5.89±0.06^{B}$	$5.57±0.24^{B}$	$5.03±0.21^{C}$
	C3	$5.78±0.11^{B}$	$7.23±0.35^{A}$	$5.64±0.34^{BC}$	$5.28±0.46^{BC}$	$4.70±0.22^{C}$
Coliforms	C1	$3.04±0.62^{A}$	$2.56±0.13^{AB}$	$2.15±0.21^{B}$	Nd	Nd
	C2	$2.88±0.04^{A}$	$2.16±0.16^{A}$	$1.09±1.54^{B}$	Nd	Nd
	C3	$2.85±0.05^{A}$	$2.37±0.11^{A}$	$1.00±1.41^{B}$	Nd	Nd
LAB	C1	$7.17±0.33^{B}$	$8.19±0.30^{A}$	$7.98±0.29^{A}$	$6.67±0.26^{B}$	$6.48±0.25^{B}$
	C2	7.21±0.74	7.61±0.41	7.39±0.62	6.28±0.60	5.75±0.63
	C3	$6.51±0.04^{B}$	$7.79±0.16^{A}$	$7.24±0.12^{A}$	$6.29±0.58^{B}$	$6.26±0.36^{B}$
Micrococci and staphylococci	C1	5.42±0.17	$5.64±0.05^{b}$	5.57±0.04	5.10±0.28	5.57±0.23
	C2	$5.65±0.07^{B}$	$6.74±0.20^{A,a}$	$6.56±0.36^{A}$	$4.25±0.08^{C}$	$5.50±0.28^{B}$
	C3	5.79±0.55	$6.57±0.34^{a}$	6.17±0.33	4.38±0.32	5.18±0.62
Proteolytic bacteria	C1	$5.05±0.44^{C}$	$6.45±0.01^{AB,a}$	$6.63±0.21^{A}$	$6.02±0.24^{AB}$	$5.72±0.25^{BC}$
	C2	$4.37±0.04^{C}$	$6.26±0.03^{AB,b}$	$6.48±0.25^{A}$	$6.15±0.35^{AB}$	$5.63±0.21^{B}$
	C3	$4.36±0.07^{D}$	$6.05±0.01^{AB,c}$	$6.20±0.28^{A}$	$5.69±0.13^{BC}$	$5.33±0.38^{C}$
Lipolytic bacteria	C1	$5.71±0.24^{C}$	$6.70±0.22^{B}$	$7.52±0.16^{A}$	$6.38±0.61^{BC}$	$6.51±0.46^{BC}$
	C2	5.48±0.25	6.17±0.66	6.99±0.08	6.19±0.41	6.45±0.31
	C3	$5.68±0.04^{C}$	$6.13±0.28^{BC}$	$7.31±0.17^{A}$	$6.50±0.28^{B}$	$6.02±0.24^{BC}$
Yeasts and moulds	C1	$5.97±0.05^{a}$	6.63±0.67	7.34±0.11	6.30±0.91	6.56±0.21
	C2	$5.64±0.02^{b}$	6.33±0.68	7.18±0.32	6.30±0.42	5.43±0.67
	C3	$4.87±0.12^{B,c}$	$6.17±0.33^{A}$	$6.78±0.25^{A}$	$6.17±0.45^{A}$	$5.71±0.24^{A}$

C1, C2, C3 Salting with 4, 5 and 6%, respectively. Nd Not detected, ^{ABCD}Different letters indicate P< 0.05 between ripening times.
^{abc}Different letters indicate P<0.05 between salt concentration of cheese.

30°C for 48 h, and psychrotropics were grown on PCA at 7°C for 10 days. Coliforms were determined on Violet Red Bile Agar (Oxoid) at 30°C for 24 h. Lactic acid bacteria (LAB) were grown anaerobically on MRS Agar (Oxoid) with its pH reduced to 5.7 at 30°C for 5 days (Pichhardt, 1998) and cycloheximide (Sigma) was added (100 mg/l) to prevent the growth of yeasts (Kandler and Weiss, 1986). Micrococci and staphylococci were grown on Baird-Parker Agar (Oxoid) at 37°C for 48 h. Proteolytic bacteria were made on PCA supplemented with reconstituted 10% skim milk and lipolytic bacteria were made on Tributyrin Agar (Oxoid) at 30°C for 48h. Yeasts and moulds were made on Potato Dextrose Agar (Oxoid) adjusted to pH 3.5 with 10% lactic acid (Merck) at 25°C for 3 days (Pichhardt, 1998). After incubation, colonies were counted and results were expressed as log cfu/g of cheese.

Sensorial analysis: Cheese samples ripened for 30, 60, and 90 days were organoleptically assessed by a group of five panelists. The samples were presented to panelists in randomized order after having stood for 4 h at room temperature and were graded between 1-10 (1: very bad and 10: very good) for appearance and colour, body and texture, flavour and saltiness (Larmond, 1987).

Statistical analysis: Statistical analysis of data for effects of salt concentrations and ripening times on microbiological, chemical and sensory properties were performed by ANOVA procedures using SAS® PROC GLM/STAT (SAS, 1998). Differences among means were identified using Duncan multiple range test.

Results and Discussion

Chemical changes during ripening in cheese samples are presented in Table 1. Dry matter, protein, fat and salt contents in all cheese samples gradually increased during ripening. However, only dry matter and salt contents were found significant (P<0.05). As salt concentration increased from 4 to 6%, dry matter, protein, fat and salt contents increased. Only salt content from these changes were significantly (P<0.05) affected. Titratable acidity was significantly (P<0.05) increased until 60 days of ripening and then was decreased (P<0.05). The initial increase in acidity is due to lactic acid and hydrogen formation (Dervisoglu and Yazici, 2001). The decrease of afterward in acidity may happen as a consequence of acid assimilation by yeasts and moulds (Coskun, 1998). Increase in the salt content in cheese samples was slightly decreased acidity. This decrease of acidity may be due to bacteria activities (see

Table 3: Sensory properties of cheese samples during ripening

		Ripening time (day)		
Property	Cheese	30	60	90
Appearance and colour	C1	6.20±0.84	6.21±1.48	6.21±1.10^{b}
	C2	6.80±0.45	6.80±1.92	6.81±0.84^{ab}
	C3	7.00±0.71	7.00±1.58	7.20±0.84^{a}
Body and texture	C1	6.00±1.23	6.20±1.30	7.00±1.00
	C2	6.40±0.89	6.40±0.55	7.60±1.14
	C3	6.70±0.71	6.80±1.30	7.20±1.64
Flavour	C1	5.80±1.79	6.40±1.52	7.00±1.23
	C2	6.00±0.71	6.20±1.30	7.00±0.71
	C3	5.60±1.82	5.60±1.14	5.80±0.84
Saltiness	C1	7.00±0.71	6.20±1.30	6.60±0.55^{a}
	C2	6.60±1.14^{A}	6.60±0.89^{A}	5.80±0.84^{B,b}
	C3	5.60±1.14	5.60±1.14	5.00±0.71^{c}

C1, C2, C3 Salting with 4, 5 and 6%, respectively. ^{AB}Different letters indicate P< 0.05 between ripening times.
^{abc}Different letters indicate P<0.05 between salt concentration of cheese.

Table 2).

Ratios of WSN/TN, TCA-SN/TN, and PTA-SN/TN of cheese samples were significantly (P<0.05) increased during ripening. Ripening time, as already observed by Freitas and Malcata (1996) and Freitas *et al*. (1997) was the dominant factor in terms of proteolytic evolution. But salt concentrations in cheese samples played negative role on the mentioned parameters. These values are similar to those reported by Dervisoglu and Yazici (2001) for Kulek cheese.

Lipolysis levels of all cheese samples increased significantly (P<0.05) from 0.62 to 2.66 during ripening. The effect of salt concentration on lipolysis was not found significant (P>0.05).

The microbiological changes of ripening times versus salt concentrations in cheese samples are presented in Table 2. Ripening times have no a significant effect on TAB counts. Counts of psychrotrophics increased until 15 days and then decreased significantly (P<0.05). Coliforms decreased rapidly until 30 days of ripening and then it was not determined.

Availability of coliforms in cheeses emphasizes the importance of contamination during cheesemaking. Similar results were reported by other researchers (Coskun, 1998; Pérez Elortondo *et al*., 1999; Hatzikamari *et al*., 1999). Evolution of LAB in the C1 and C3 were significantly (P<0.05) founded during ripening times. These data are in accordance with the results found by Coskun (1998) and Dervisoglu and Yazici (2001) for Herby cheese and Kulek cheese, respectively. Micrococci and staphylococci counts increased until 15 days of ripening. They decreased until 60 days and approximately reached counts of beginning at 90 days. These changes have only a significant (P<0.05) effect in C2 samples. Proteolytic and lipolytic bacteria counts were observed similarity changes during ripening times. These bacteria counts were founded a somewhat decrease during ripening. These results are in agreement with those reported by others (Hatzikamari *et al*., 1999; Dervisoglu and Yazici, 2001). Yeasts and moulds counts increased until 30 days and afterward decreased (P<0.05). It was generally observed that salt concentrations had no influence on the microbiological parameters.

The mean scores of the sensory panel's assessment of the cheese quality after ripening for 30, 60 and 90 days are shown in Table 3. These data show that all the sensory scores except for saltiness scores increased during ripening times. The cheese samples added with higher salt received higher the appearance and colour scores but these scores were significantly (P<0.05) found at the only 90 days. The saltiness scores during ripening times had a significant (P<0.05) effect in C2 samples.

Conclusion: In generally, the results of the present study indicated that ripening times had a significant effect (P<0.05) on the microbiological and chemical parameters of Herby cheeses. The salt concentrations in the cheese samples decreased the microbiologic counts and sensory scores (except saltiness score) but these decreases were not statistically found significant (P>0.05).

It was concluded that resistant microorganisms, such as micrococci and staphylococci, yeasts and moulds, seemed to be stimulated by the higher salt content in cheese samples.

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