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Ripening Changes of Ras Cheese Made from Recombined Milk as Affected by Certain Additives

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ABSTRACT

An attempt has been made to accelerate the ripening of Ras cheese made from recombined milk (RM). RM cheese was made from curd with either a mixture of Fromase 100 (fungal rennet) and Kapalase L (an animal lipase) at concentrations of 0·025 and 0·05% or a slurry of fully ripened cheese at concentrations of 1 and 2%. These treatments enhanced flavour development, body characteristics, formation of soluble nitrogen compounds and free fatty acids. The proteinase/lipase mixture was the most effective. A rancid flavour and bitter taste were developed in 3–4-month-old RM cheese made with the higher concentration enzyme mixture.

INTRODUCTION

The manufacture of hard cheese from dried milk is complicated by technological problems such as slow coagulation and weak curd. Such characteristics lead to a cheese of low moisture content and a retarded rate of ripening which results in a product with inferior flavour and weak body and texture (Peters & Williams, 1964; Abdel-Gawad, 1970; Theo *et al.*, 1970; Abdel Salam *et al.*, 1974; Hofi *et al.*, 1978).

Hofi *et al.* (1983) showed that the most suitable manufacturing conditions for the production of Ras cheese from recombined milk with

properties similar to those of fresh milk curd were: coagulation temperature of 40 °C with the addition of 0·05 % calcium chloride. The authors suggested the addition of either 1 % NaCl, 0·2 % disodium phosphate, 0·2 % sodium hexametaphosphate or 0·1 % sodium citrate at renneting. The suggestions of Hofi *et al.* (1983) were applied by El-Ghandour *et al.* (1983) in the production of Ras cheese from recombined milk. They showed that cheese body was improved but the ripening process occurred at a relatively slow rate compared with fresh milk cheese.

It is generally recognized that protein and in some cases fat breakdown are good indices of the progress of ripening and the products of these processes may contribute to background taste and provide a pool of flavour precursors. Several investigators have reported on the effectiveness of selected exogenous proteinases and lipases as well as cheese slurry in enhancing protein breakdown and fat hydrolysis during the ripening of cheese made from fresh milk (Von Bockleman & Lodin, 1974; Kosikowski & Iwaski, 1975; Abdel Salam *et al.*, 1979; Abdel Baky *et al.*, 1982).

The present work was carried out to evaluate the effect of a selected proteinase/lipase mixture or a slurry of ripened cheese on the quality and ripening changes of Ras cheese (national hard type in Egypt) made from recombined milk.

MATERIALS AND METHODS

Materials

Skim milk powder produced in Holland was obtained from the Misr Milk and Food Company, Egypt. Butter oil, produced in France, and fully ripened good quality Ras cheese were obtained from the local market. Fresh cow's milk for the manufacture of control Ras cheese was obtained from the herd of the Faculty of Agriculture, Zagazig University, Egypt. A rennet powder (1:100 000) was obtained from the L. C. Glad Company AS, Copenhagen, Denmark.

Commercial enzyme preparation

Fromase 100, a fungal rennet derived from *Mucor meihei*, was obtained from Rapidase seclin, France. Kapalase L, a pregastric lipase, was

obtained from Dairyland Food Laboratories, Waukesha, Wisconsin, USA. A mixture consisting of 2 parts of proteinase and one part of lipase was prepared and used in cheesemaking.

Cheese slurry

Two parts of good quality Ras cheese were blended with one part of 0·5% sodium citrate solution (Von Bockleman & Lodin, 1974).

Cheesemaking

Recombined cream containing 25% fat was prepared at El Mansoura Factory, Misr Milk and Food Company, Egypt. Skim milk powder was reconstituted in warm water (40 °C) at a ratio of 1:8. The fat content of cheese milk was standardized to 4% using the recombined cream. Ras cheese curd was prepared as described by Abdel Tawab (1963) with the modification suggested by Hofi *et al.* (1983). The curd was divided into five equal parts. The proteinase/lipase mixture was added to the first and second parts at concentrations of 0·025 and 0·05% respectively.

The third and the fourth parts were mixed well with cheese slurry at concentrations of 1% and 2% of curd weight respectively. The fifth part was left without additives. Control cheese was made from fresh milk (Abdel Tawab, 1963). All cheeses were ripened at 12 ± 2 °C for 4 months. Treatments were carried out in triplicates.

Chemical analysis

Cheese samples were analysed for moisture, fat, NaCl, total nitrogen (TN), and soluble N as described by Ling (1963). Amino acid N was determined as described by Stadhouders (1959). Cheese fat acidity was determined by the method described by Abdel Kader (1971). The slurry of ripened cheese was chemically analysed as described for cheese.

Preparation of the samples for gas–liquid chromatography

Sodium soaps of the free fatty acids were prepared from the 4-month-old cheese samples according to the method of Kuzdzal & Kuzdzal-Savoie (1966). Methyl esters of the free fatty acids were prepared as described by Kuzdzal-Savoie & Kuzdzal (1967). They were separated in a Pye Unicam,

Series 104, gas chromatograph (Pye Unicam, Cambridge, England) equipped with a dual flame ionization detector. Columns of 3·6-m length and 2-mm inner diameter were used with 80–100 mesh silanized Chromosorb W carrier coated with 10% polyethylene glycol adipate as the stationary phase. Temperature programming at a rate of 5 °C min^{-1} was applied in the range of 130–180 °C. The temperature of the injection port was 200 °C and that of the detector was 300 °C. Carrier gas (He) flow was adjusted to 35 ml min^{-1}. Chart speed was 5 mm min^{-1}. Peak areas were calculated by multiplying the peak height at the maximum by the width of the peak at half of its height. Results are expressed as mg per 100 g cheese.

Organoleptic properties

Ras cheese samples were examined organoleptically according to the method described by Abdou *et al.* (1977) with maximum score points of 40, 50 and 10 for body characteristics, flavour and appearance, respectively.

Bacteriological examination of the cheese slurry

Cheese slurry was examined for total bacterial, proteolytic and lipolytic counts according to the methods of Marth (1978).

RESULTS

Gross chemical composition

It appears from the results in Table 1 that recombined milk cheese without additives differed from fresh milk cheese in showing lower moisture content, slightly lower fat and higher pH values during the ripening period. The salt and total N contents of both cheeses were very similar. Addition of the proteinase/lipase mixture or slurried ripened cheese to cheese curd did not considerably affect the above properties except that the pH of the cheese was lower in samples containing additives.

TABLE 1
Gross Chemical Composition of Ras Cheese Made from Recombined Milk (RM) as Affected by Some Additives

Property	*Ripening period (months)*	*Fresh milk cheese (control)*	*RM cheese*				
			Without additives	*A*[a]		*B*[a]	
				0·025%	*0·05%*	*1%*	*2%*
Moisture, %	Fresh	39·31	37·6	37·40	37·35	37·60	37·83
	2	38·08	35·8	34·98	35·20	36·00	36·20
	4	37·46	34·2	33·97	34·10	34·50	34·90
Fat (DM), %	Fresh	47·00	45·6	45·60	45·70	45·40	45·50
	2	47·92	46·3	46·40	46·48	46·30	46·32
	4	48·40	47·0	47·00	47·20	47·00	47·10
Salt (DM), %	Fresh	3·30	3·32	3·26	3·30	3·32	3·30
	2	4·50	4·58	4·52	4·50	4·54	4·56
	4	4·68	4·70	4·66	4·68	4·70	4·70
TN (DM), %	Fresh	3·60	3·62	3·58	3·60	3·62	3·62
	2	3·62	3·66	3·64	3·68	3·66	3·67
	4	3·68	3·69	3·70	3·70	3·72	3·72
pH	Fresh	5·40	5·39	5·38	5·36	5·38	5·38
	2	5·20	5·36	5·00	5·00	5·01	5·02
	4	4·99	5·20	4·90	4·90	4·92	4·90

[a] A, Cheese from curd with added proteinase/lipase mixture. B, Cheese from curd with added slurry of ripened cheese.

TABLE 2
Ripening Indices of Ras Cheese Made from Recombined Milk (RM) as Affected by Some Additives

Property	*Ripening period (months)*	*Fresh milk cheese (control)*	*RM cheese*				
			Without additives	*A*[a]		*B*[a]	
				0·025%	*0·05%*	*1%*	*2%*
SN[b]	Fresh	7·00	6·8	7·00	7·00	7·10	7·10
	2	14·20	10·4	13·50	16·40	12·50	13·40
	4	17·60	12·60	20·60	23·90	18·40	18·60
NPN[b]	Fresh	1·56	1·00	1·40	1·40	1·45	1·46
	2	3·33	2·52	3·15	3·75	2·94	2·91
	4	6·40	3·72	5·18	6·58	4·31	4·85
AN[b]	Fresh	0·96	0·64	0·64	0·68	0·65	0·67
	2	1·51	0·84	2·40	2·24	1·29	1·50
	4	3·44	1·16	4·20	4·20	2·79	3·13
Cheese fat acidity (0·1 N NaOH per 10 g of cheese)	Fresh	0·80	0·40	0·50	0·56	0·48	0·46
	2	2·18	0·72	1·92	2·56	1·68	1·70
	4	3·08	1·74	5·00	6·80	4·00	4·40

[a] A, Cheese from curd with added proteinase/lipase mixture. B, Cheese from curd with added slurry of ripened cheese.
[b] Percentage of total N.

Ripening indices

Soluble N (SN), non-protein N (NPN), amino acid N (AN), cheese fat acidity and free fatty acids were taken as indices for cheese ripening.

SN, NPN, AN and cheese fat acidity

Table 2 shows that the SN, NPN, AN and cheese fat acidity of recombined milk cheese without additives were lower than those of fresh milk cheese. Addition of the proteinase/lipase mixture or slurried ripened cheese to the curd enhanced the formation of soluble nitrogen compounds and cheese fat acidity. The proteinase/lipase mixture was more effective in this respect.

Free fatty acids

Free fatty acids (FFA) were determined in the 4-month-old cheeses. Table 3 shows that the pattern of FFAs in the different treatments was found to be similar. The cheese made with added enzymes contained a higher concentration of FFAs than that made from curd mixed with the slurry of ripened cheese, but both contained higher concentrations of FFAs than the recombined milk cheese without additives.

The formation of volatile fatty acids (C_2–C_8) and non-volatile fatty acids (C_{10}–$C_{18:3}$) was stimulated in recombined milk cheese with added enzymes or cheese slurry. Concentrations of FFAs in cheese with added enzymes or slurry were at least as high as those in fresh milk cheese and in some cases were higher.

Organoleptic properties

Table 4 shows that recombined milk cheese, at 1 month without additives, had low flavour intensity and acid taste. The flavour of the cheese did not improve greatly during ripening. Some bitterness was noted in most samples of recombined milk cheese without additives after 2 and 4 months of ripening. The cheese body was crumbly, weak, tough and hard compared with fresh milk cheese.

Addition of enzymes to the curd influenced flavour intensity but a rancid flavour and a bitter taste were observed in cheese made with the

TABLE 3
Free Fatty Acids of 4-month-old Ras Cheese Made from Recombined Milk (RM) as Affected by Some Additives (mg per 100 g cheese)

Free fatty acid	*Fresh milk cheese (control)*	*RM cheese*				
		Without additives	*A*[a]		*B*[a]	
			0·025%	*0·05%*	*1%*	*2%*
C_2	4·0	3·0	5·3	5·6	2·4	2·7
C_3	1·3	0·7	4·6	4·3	2·6	2·0
C_4	5·8	1·6	5·7	11·5	5·3	5·3
C_5	0·4	0·1	0·1	0·2	0·2	0·1
C_6	1·9	0·8	1·9	3·9	1·7	1·6
C_8	1·6	1·2	2·0	2·5	1·0	1·7
C_{10}	5·9	4·0	5·3	6·4	3·1	4·1
C_{11}	0·0	0·4	0·5	0·7	0·6	0·5
C_{12}	10·7	3·5	7·6	8·8	7·8	10·7
C_{13}	2·6	0·6	2·1	1·1	1·0	1·7
C_{14}	23·5	10·2	14·7	22·5	17·1	20·8
C_{15}	5·3	1·1	4·1	2·7	2·2	3·0
C_{16}	28·0	15·4	30·6	43·2	23·6	28·4
C_{17}	0·0	0·5	0·7	0·9	1·0	1·1
C_{18}	2·3	2·9	6·8	0·4	3·3	5·1
$C_{11:1}$	2·6	0·2	2·4	3·3	0·3	0·3
$C_{14:1}$	6·1	2·0	3·5	4·6	3·0	4·8
$C_{16:1}$	4·0	2·4	6·0	6·5	4·0	4·9
$C_{17:1}$	0·0	0·5	0·7	1·0	0·9	1·1
$C_{18:1}$	3·8	9·5	24·0	34·7	18·10	19·7
$C_{18:2}$	1·50	Trace	3·5	4·1	7·2	7·4
$C_{18:3}$	0·80	Trace	1·0	2·0	0·8	1·0

[a] A, Cheese from curd with added proteinase/lipase mixture. B, Cheese from curd with added slurry of ripened cheese.

high concentration of enzymes, particularly at the late stage of ripening. Also flavour development was enhanced by the use of slurried ripened cheese and although its effectiveness in this respect was lower than that of the enzyme mixture, no bitterness or rancidity was detected in the slurry-containing cheese with continued storage.

Body characteristics of the recombined milk cheese were similarly enhanced by the incorporation of enzymes or slurried ripened cheese into the curd and were comparable to those of fresh milk cheese.

TABLE 4

Organoleptic Properties of Ras Cheese Made from Recombined Milk (RM) as Affected by Some Additives

Ripening period (months)	*Property (maximum points)*		*Fresh milk cheese (control)*	*RM cheese*				
				Without additives	*A*[a]		*B*[a]	
					0·025%	*0·05%*	*1%*	*2%*
1	Appearance	(10)	8	7	8	8	8	8
	Body & texture	(40)	33	26	32	33	32	32
	Flavour	(50)	43	27	42	42	41	41
2	Appearance	(10)	8	7	8	8	8	8
	Body & texture	(40)	35	28	34	35	34	34
	Flavour	(50)	44	28	42	43	42	42
3	Appearance	(10)	9	7	8	8	8	8
	Body & texture	(40)	36	29	35	36	35	35
	Flavour	(50)	46	30	44	40	43	44
4	Appearance	(10)	9	7	8	8	8	8
	Body & texture	(40)	37	30	36	36	36	35
	Flavour	(50)	46	30	45	37	44	44

[a] A, Cheese from curd with added proteinase/lipase mixture. B, Cheese from curd with added slurry of ripened cheese.

DISCUSSION

In this study an attempt was made to accelerate the ripening of Ras cheese made from recombined milk by using either a proteinase/lipase mixture or a slurry of ripened cheese. Introducing either additive into Ras cheese curd improved flavour as well as body and texture. Cheese made from curd with added enzymes or slurry acquired strong flavour and good body characteristics after 3–4 months of ripening. El-Ghandour *et al.* (1983) showed that Ras cheese made from recombined milk showed low flavour intensity even after 4 months of ripening.

Slurry-containing cheese did not develop flavour defects with continuous storage but a rancid flavour and bitter taste were detected with the higher concentration of proteinase/lipase mixture. Abdel Baky *et al.* (1982) showed that Ras cheese made from milk or curd with added slurry had good flavour and satisfactory body during ripening. Also, Abdel Salam *et al.* (1979) reported that Ras cheese, made from curd to which large quantities of proteolytic and lipolytic enzyme preparations were added, developed strong flavour after 45 days of storage but exhibited defects on prolonged storage.

The results obtained can be explained on the basis that the addition of proteinase and lipase to cheese curd enhanced both proteolysis and lipolysis during cheese ripening (Abdel Salam *et al.*, 1979). In addition, cheese slurry contains several factors relating to cheese ripening, e.g. simple nitrogenous compounds, proteolytic and lipolytic bacteria (Table 5) and probably proteinases, peptidases and lipases also.

In conclusion, the ripening of Ras cheese made from recombined milk could be enhanced by incorporating a proteinase/lipase mixture (2 parts

TABLE 5
Chemical Composition and Bacterial Examination of Slurry of Ripened Cheese

Chemical composition (%)		*Bacterial examination*	
Moisture	60·80	Total count	128×10^6
Fat	16·50	Proteolytic count	36×10^6
Salt	2·10	Lipolytic count	24×10^6
SN[a]	22·70		

[a] Percentage of total N.

of Fromase 100:1 part of Kapalase L) into the curd at a concentration of 0·025% or slurried ripened cheese at a concentration of 2%.

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