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Determination of Optimum Dosage of Milk Coagulants for Cheddar Cheesemaking

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The coagulant traditionally used for cheesemaking in most parts of the world is rennet extract obtained from calves. Its short supply, increased production of cheese the world over and, to certain extent, non-acceptability for religious or ethical reasons have stimulated interest in the search for suitable substitutes for use as coagulants in cheesmaking.

Amongst different substitutes, those derived from fungi, namely, *Endothia parasitica*, *Mucor miehei* and *Mucor pusillus* var. *Lindt* have shown real promise (Sternberg, 1976). An array of cheese varieties including Cheddar cheese from cow milk have been manufactured using preparations derived from these sources (Lawrence *et al.* 1972; Wigley, 1974; Poznanaki *et al.* 1974; Dinesen *et al.*, 1975). However, the informatin on technological suitability of these coagulants to manufacture such varieties from buffalo milk is lacking. A study was, therefore, planned and conducted to evaluate the suitability of two fungal milk coagulants, viz., Rennilase from *Mucor miehei* and *Meito* from *Mucor pusillus* var. *Lindt* and their mixtures for the manufacture of buffalo milk Cheddar cheese and preliminary observation on clotting time, curd firmness and optimum dosage are reported in this communication.

The fungal rennets were tried as full or part replacement of the standard calf rennet or as a mixture of both, evolving following combinations.

1. Calf rennet	C. Control
2. Meito rennet	M. alone
3. *Rennilase*	R. alone
4. Calf+Meito	CM.1:1 W/W
5. Calf+Rennilase	CR. 1:1 W/V
6. Meito+Rennilase	MR. 1:1 W/V

For each of the six rennets arbitrarily four levels of addition were selected. The rennets were weighed/measured accurately and stock solutions, 100 ml each, were prepared using distilled water. Suitable working solutions were obtained by dilluting the stock solution with distiled water 1 ml of which when added to 100 ml milk, represented the required final concentration of respective rennet in cheese milk. The solutions were prepared fresh for each trial when required.

Fresh, standardized (casein : fat ratio adjusted to 0.7±0.01), pasteurized (72° C, 15 to 16 Sec) buffalo pooled milk was used as substrate.

Coagulation time was determined by the Macrofilm technique of Sharma and Bhalerao (1964) using a pair of big and small diameter test tubes instead of a test tube and 12.5 ml. Kjeldhal flask.

Curd firmness for the various rennets was evaluated using the same substrate

and at similar concentrations, as used in determination of coagulation time.

Glass bowls, containing one litre of milk each, were placed in a thermostatically controlled water bath at 32±0.1° C and 10 ml of respective rennet solutions were added to each pair of bowls. The contents were mixed well and allowed to set for 30 min. After 30 minutes, curd firmness was judged by subjective and penetrometer methods.

In subjective method, the firmness of curd was ascertained by the sense of touch feel and by the type of cut obtained when a glass rod was inserted at an angle of 45° and the curd lifted straight up. A clean sharp cut revealed the desired degree of firmness at cutting.

The penetrometric measurements expressed in millimeters were made two minutes after setting, on AIMIL make universal penetrometer, type FPN-3. The measurements were made using a cone and test rod weighing 150 g for a fix time of 5 seconds. For the same sample, the readings were recorded at three different spots and average of two closest values was used for ascertaining the degree of curd firmness.

The optimum rate of addition of each of the six rennets was determined on the basis of results of tests for coagu lation time and curd-firmness. Out of the four levels of each rennet tested, the one that gave the desired curd strength at minimum level was considered optimum and selected for cheesmaking.

TABLE 1

Influences of type and amount of rennet on coagulation time and curd firmness.

Type of rennet	*Rate of addition g/ml per 100 liters*	*Time for coagulation at 32° C.* Min	Sec.	*Penetrometer reading in mm*	*Firmness of curd after 30 minutes*	*Optimum Dosage per 100 liters*
C	3.00	20	12	54.0	Very weak	
	3.50	17	03	45.5	weak	3.75 g
	3.75	16	06	41.7	firm	
	4.00	15	24	39.4	firm	
M	1.50	17	19	51.4	weak	
	1.75	14	12	42.8	firm	1.75 g
	2.00	13	16	41.0	firm	
	2.25	11	40	40.5	firm	
R	1.50	17	14	51.0	weak	
	2.00	14	50	42.0	firm	2.0 ml
	2.50	11	03	40.7	firm	
	3.00	9	29	40.5	firm	
CM	2.00	19	15	54.0	weak	
(50 : 50,	2.50	15	50	42.7	firm	1.25 g +
W/W)	3.00	14	22	41.5	firm	1.25 g
	3.50	11	52	40.3	firm	
CR	2.00	18	30	52.6	weak	
(50 : 50,	2.50	15	10	42.4	firm	1.25 g +
W/V)	3.00	12	04	41.0	firm	1.25 ml
	3.50	11	50	39.5	firm	
MR	1.50	17	15	54.0	weak	
(50 : 50,	2.00	14	05	43.0	firm	1.0 g +
W/V)	2.50	10	25	41.5	firm	1.0 ml
	3.00	9	16	40.5	firm	

Average of 3 trials.

The data presented in Table 1 show that out of four levels of each rennet tested the minimum concerntration that gave curd of suitable firmness in 30 min. at 32° C. for Cheddar cheesemaking from buffalo milk, per 100 litres was 3.75 g for rennet C., 1.75 g for rennet M, 2.0 ml for rennet R., 2.5 g (1.25 g C+1.25 g M) for rennet CM, 2.5 (1.25 g C+1.25 ml. R) for rennet CR and 2.0 (1.0g M+ 1.0 ml R) for rennet MR.

At the above rate of addition of each rennet, the curd firmness as revealed by extent of penetration, ranged between 41.7 mm for rennet C to 43.0 mm for MR rennet. The other coagulants showed intermediate values. The results of penetrometric measurements as well as subjective evaluation of the curds obtained with different coagulants, showed that all milk coagulants had almost similar curd firmness. This is in accordance with the results obtained with cow milk by Stavlund and Kiermeir (1973), Reps. *et al.* (1974 a,b,c, 1975), Dolezalek and Havlova (1974) and Dennien (1977).

The coagulation time (i.e. first sign of appearance of curd flakes) recorded for different coagulants at the rate of addition found most suitable for cheesemaking from buffalo milk, ranged between 14 min and 5 seconds for MR rennet to 17 min and 3 seconds for C rennet. The order of coagulation time observed with different coagulants was C > CM > CR > R > M > MR. The shorter coagulation time observed with fungal rennets and their mixtures compared to calf rennet (C) could be ascribed to their greater sensitivity to CA^{++} concentration (Iwasaki *et al.* 1967; Richardson *et al.* 1967; Alais and Lagrange, 1972; Reps *et al.* 1975), which is usually higher in buffalo milk (Verma and Anantakrishnan, 1946; Prakash, 1965; Dastur *et al.* 1971; Hamdy *et al.* 1972; Ganguli, 1973).

Irrestpective of differences in coagulation time all the coagulants developed similar curd firmness and gave a setting time of 30 min. Nelson (1969) and Richardson *et al.* (1967) also observed similar behaviour in cow milk for calf rennet, *Mucor pusillus Lindt* rennet and their combinations. However, Richardson *et al.* (1967) noted longer coagulation time and softer curd formation with Mucor rennet compared to calf rennet, which is contrary to the findings of the present study and the same could be attributed to differences in the behaviour of the two milks towards rennet action.

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