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Effect of different level of mesophilic starter culture on sensory and bio-chemical characteristics of buffalo milk feta type cheese during ripening

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Feta cheese, a predominant variety in terms of production, qualities and worldwide acceptance, is made from sheep's milk alone or from mixture with goat's milk. Feta cheese developed from cow milk has to be bleached to result in desired white colour, which destroys valuable ß-carotene. Buffalo milk can be an appropriate substitute to cow milk, as no bleaching is required to get characteristics colour of Feta cheese. Out of 121.8 million tonnes milk produced in India, more than half (57%) of the total milk production is from buffalo milk (IDF 2008). Presently there is a great scope for manufacture of Feta cheese from buffalo milk and its export to Middle-East countries. Selection of appropriate level of starter culture for manufacture of good quality Feta cheese is important for good quality Feta cheese (Kumar *et al.* 2011a). The effect of different levels of inoculation of starter culture NCDC-149 was investigated. The Feta type cheese was manufactured from buffalo milk after standardizing the milk to casein/fat (C/F) ratio 0.70 (Kumar *et al.* 2011b, c) with using 1.0, 1.5 and 2.0% culture. The cheeses obtained from these 3 levels were analyzed for sensory and bio-chemical characteristics during ripening up to 2 months.

Fresh raw buffalo whole milk and skimmed milk were procured from the experimental dairy of the institute. The mesophilic starter culture NCDC-149 (*Lactococcus lactis* ssp. *lactis, Lactococcus lactis* ssp. *cremoris, Lactococcus lactis* ssp. *diacetylactis*) was obtained from the National Collection of Dairy Cultures, NDRI, Karnal. Commercially produced microbial rennet in granular form from *Mucor pusillus* var. *Lindt* was used.

Buffalo milk was standardized to C/F ratio 0.70. The standardized milk was pasteurized to 65°C/30 min (Kumar *et al.* 2011d) and cooled in a cheese vat to 30–32°C. Starter culture (NCDC-149) @ 1.0, 1.5 and 2.0% by weight of milk were added to the different cheese vat. Subsequently, after 40–50 min, cheese milk was rennet at 31–32°C @1.25g/ 100kg of milk and coagulation takes place in 40–50 min. Whey was collected and stored at 5°C for making brine. Moulds were turned every 2–3 h to form a firm curd. After 4–5 turns, the moulds were left undisturbed for about overnight. Subsequently, cheese blocks were cut into uniform size and transferred to the whey brine solution of 14% salt (Kumar 2009) for about 22–24 h. Cheese blocks were transfer into plastic packages for vacuum packaging. Subsequently, packaged cheeses were stored for maturation at 10 ±1°C and relative humidity of 80%. Feta cheeses were packaged under vacuum after establishing a vacuum of 20 mm of Hg (< 0.70 kpa). Samples from each cheese were taken for analysis at fresh (0), 15, 30, 45, and 60 days after production. All the analyses were made in 3 replicates. Feta cheese was analysed for its bio-chemical changes at 15 days interval — fat in milk (Gerber method) and in cheese (ISI 1981), casein (Pyne 1932), pH of cheese (digital pH meter), titratable acidity (TA, AOAC 1990), total protein content of cheeses (micro-kjeldal method), moisture (IDF 1982), soluble protein of cheeses (Kosikowski 1982), lipolytic changes in cheeses in terms of free fatty acids (Deeth and Fitz-Gerald 1976). The experimental Feta cheese at different stages of development and during ripening was evaluated by a trained panel judges for sensory characteristics. The Cheddar cheese score card using 100 point (Bodyfelt *et al*. 1988) was modified in the light of characteristics of Feta cheese to use in sensory evaluation.

The data were statistically analyzed using the SYSTAT Software (version 6.01). When significant (1 and 5% levels) differences were observed, individual means were compared by using Fisher's least significant difference model.

Sensory characteristics

Flavour: Sensory evaluation in terms of flavor during ripening (Table 1) showed that the maximum flavour score

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was in cheese manufactured using 1.5% level and minimum flavour score in cheese prepared from 1.0% culture. Flavour score increased as the ripening period progressed up to 30 days, thereafter, followed a decreasing trend towards the end of ripening period of 60 days irrespective of level of starter culture used for cheese making. Similar trends were reported by Kumar *et al*. (2011b). The interaction effect between level of starter culture and ripening period on flavour also found significant (P<0.01) and showed that the maximum flavour score was found at 30 days of ripening with cheese prepared from 1.5% culture and the minimum flavour score was found in cheese prepared from 2% culture at 60 days of ripening. The cheese made with 2% culture was acidic and slight bitter while cheese made with 1% culture lacked blend flavour. The cheese made from 1.5% culture scored highest at 30 days of ripening. Our finding was in agreement with the results of Das (1996), who recommended 1.5% culture to obtain the most acceptable Gouda cheese from buffalo milk. However, Malhotra (1996) suggested 2.0% culture for manufacture of Gouda cheese.

Body and texture

Body and texture scores differed significantly (P<0.01) with the varying level of starter culture during ripening (Table 1). The maximum body and texture score was found in the cheese with 1.5% culture and minimum score with 2.0% culture. The body and texture scores increased up to 30 days, thereafter, followed a decreasing trend up to 60 days of ripening regardless of level of culture used for cheese making. Kumar *et al.* (2011b) reported similar findings with varying level of C/F ratio of buffalo milk. The interaction effect between level of starter culture and ripening period on body and texture of cheese also was significant (P<0.01). The maximum body and texture scores in cheese was with 1.5% culture at 30 days of ripening, as it was observed with soft, smooth, mellow body and texture which is desirable for Feta cheese, while minimum body and texture scores was found at 60 days with 2.0% culture, as it was criticized for weak and pasty defects. However, the cheese prepared using 1.0% culture had hard and crumbly texture defects. Our finding was further supported by Das (1996).

Colour and appearance

The maximum colour and appearance score was with 1.5% culture and minimum with 2.0%. The colour and appearance score decreased as the ripening period progressed up to 60 days irrespective of level of starter culture. Kumar *et al.* (2011b) also reported that colour and appearance score decreased in decreased in feta cheese, with the progressive ripening period. The interaction effect between level of starter culture and ripening period was significant (P<0.01) and showed that the maximum colour and appearance score was at 0 day (fresh) with 1.5% culture and minimum at 60 days of ripening with 2.0% culture.

Biochemical changes

pH: The maximum pH (5.23) was with 1.0% culture and the minimum pH (5.03) with 2.0% culture (Fig.1). Our

Table 1. Effect of varying level of starter culture on sensory characteristics of buffalo milk Feta type cheese during ripening

Sensory characteristics	Starter culture (%)	Ripening periods (days)					Overall mean
		0	15	30	45	60	
Flavour							
	1.0	38.62 ± 0.61^{a}	39.53 ± 0.25^{b}	40.80 ± 0.11^{c}	39.73 ± 0.82^{b}	39.16 ± 0.21^{b}	39.57
	1.5	39.66 ± 0.15^{a}	41.77 ± 0.20^{a}	45.33 ± 0.15^{b}	43.13 ± 0.32^{b}	41.70 ± 0.10^{a}	42.31
	2.0	39.60 ± 0.77^{a}	40.50 ± 0.39^{a}	41.30 ± 0.58^{b}	39.74 ± 0.67^{a}	38.45 ± 0.48^{c}	39.91
	Overall mean	39.29	40.58	42.47	40.86	39.77	
		CD (SC)= 0.119; CD (RD) =0.153; C.D (SC*RD) =0.266					
Body and texture							
	1.0	31.46 ± 0.45^{a}	31.66 ± 0.78^{a}	32.12 ± 0.28^{b}	31.10 ± 0.25^{a}	30.23 ±0.35^{c}	31.31
	1.5	32.06 ± 0.12^{a}	32.53 ± 0.25^{a}	33.76 ± 0.15^{b}	32.46 ± 0.15^{b}	31.10 ± 0.10^{c}	32.38
	2.0	31.86 ±0.23^{a}	32.20 ±0.53^{b}	31.33 ±0.26^{a}	29.93 ±0.33^{c}	28.80 ±0.51^{c}	30.82
	Overall mean	31.80	32.13	32.40	31.16	30.04	
		CD (SC) =0.114; CD (RD) =0.147; CD (SC*RD) =0.255					
Colour and appearance							
	1.0	14.10 ± 0.15^{a}	13.60 ± 0.11^{b}	13.10 ± 0.09^{b}	12.61 ± 0.05^{c}	12.46 ± 0.04^{c}	13.17
	1.5	14.30 ± 0.26^{a}	13.93 ± 0.11^{a}	13.90 ± 0.10^{a}	13.70 ± 0.10^{a}	13.50 ± 0.10^{a}	13.86
	2.0	14.13 ± 0.21^{a}	13.20 ± 0.17^{b}	13.00 ± 0.12^{b}	12.78 ± 0.09^{c}	12.60 ± 0.07^{c}	13.14
	Overall mean	14.17	13.57	13.33	13.03	12.85	
		CD (SC) = 0.080; CD (RD) =0.104; CD (SC*RD) =0.180					

CD: critical difference, RD: ripening period (days), SC: level of starter culture, No. of replicates 3. The values of 100–point score were divided for flavour, body and texture, and colour and appearance, viz. 50, 35 and 15; respectively. Mean in each row with different superscripts (a,b,c,d) were significantly different (P < 0.01) from each other.

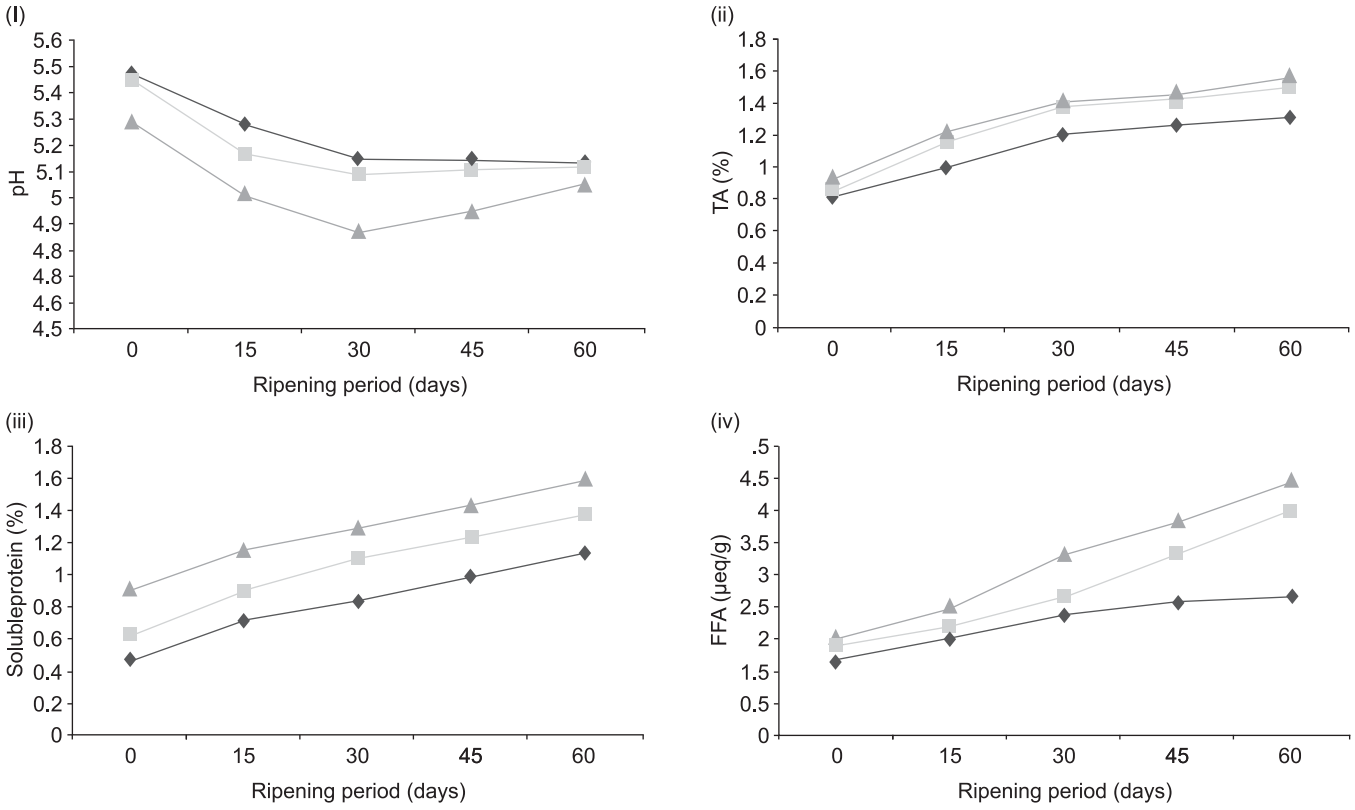

Fig. 1. Effect of different level of starters culture on biochemical characteristics of Feta type cheese during ripening: (i) pH; (ii) TA(%); (iii) Soluble protein (%); and (iv) FFA (µeq/g). —◆— 1% —■— 1.50% —▲— 20% represents level of starter culture.

finding was in agreement with the result of Najafi *et al.* (2008), who reported that pH of fresh white brined cheeses decreased with increase in concentration of starter culture. The pH of cheese decreased as the ripening period progressed up to 30 days, thereafter, followed a slight increasing trend up to 60 days of ripening irrespective of level of culture used for cheese making. Our result was in agreement with Mallatou *et al*. (1994). The increase in pH after 30 days of ripening is attributed to the utilization of lactic acid, formation of non acidic decomposition products and liberation of alkaline products of protein decomposition (McSweeney and Fox 1993). The interaction effect between levels of starter culture and ripening days revealed that the maximum pH (5.47) was at 0 day with the cheese made from 1.0% culture and the minimum pH (4.87) was at 30 days with 2.0% culture. Our finding was in accordance with the result of Azarnia *et al.* (1997) and Najafi *et al.* (2008). This may be mainly due to completion of lactose fermentation and the liberation of amino and free fatty acids. The slow solubilization of colloidal calcium phosphate during ripening causes a slow increase in pH (Lucey *et al.* 2003).

Titratable acidity

The effect of levels of starter culture on TA of Feta cheese during ripening revealed that the maximum TA (1.31%) was in cheese with 2.0% culture (Fig. 1). However, the minimum TA (1.11%) was with 1.0% culture. The TA of experimental cheeses increased with the increase in level of starter culture regardless of level of starter culture used for cheese making. Similar findings were reported by Kumar *et al.* (2011b). The interaction effect between level of starter culture and ripening period on TA also significant (P<0.01) and revealed that that the maximum acidity (1.55%) was at 60 days with 2.0% culture, while the minimum TA (0.81%) was at 0 day (fresh) with 1.0% culture. Najafi *et al.* (2008) also reported that acidity of Iraninan brined cheese increased with the increase in starter culture content during ripening and acidity of cheese increased throughout the storage period regardless of concentration of starter culture.

Soluble protein

The soluble protein was maximum (1.27%) with 2.0% culture, but minimum (0.82%) with 1.0% culture (Fig. 1). The interaction effect between level of starter culture and ripening period showed that the maximum soluble protein (1.59%) was with 2.0% culture at 60 days of ripening and the minimum soluble protein (0.47%) with 1.0% culture at 0 day (fresh). The soluble protein of cheese increased throughout the ripening period regardless of level of culture used for cheese making. Kavas *et al*. (2004) also reported

that the percent water soluble nitrogen in white pickled cheese increased throughout the ripening period up to 90 days, which supported our results. Kumar and Kanawjia (2012) reported similar findings. This may be due to higher proteolysis in higher level of starter culture as the cheese made from higher level of starter culture, retains higher moisture content with higher content of proteolytic enzyme release by starter culture, produces higher proteolysis in cheese than the cheese made with lower level of starter culture.

Free fatty acid

FFA content of cheese with respect to level of starter culture, ripening period as well as their interaction effect were found highly (P<0.01) significant (Fig. 1). The maximum FFA (3.03 μeq/g) was with 2.0% culture and minimum (1.94 μeq/g) with cheese made with 1.0% culture. FFA content increased consistently with the increase in ripening period irrespective of levels used for cheese making. Our finding was in accordance with the result of Das (1996). The interaction effect of levels of culture and ripening periods was also highly significant (P<0.01) and showed that the maximum FFA (4.41 μeq/g) was at 60 days of ripening with 2.0% culture, while the minimum FFA (2.40 μeq/g) was at 0 day (fresh) with 1.0% culture. This finding was in accordance with the result of Georgale *et al.* (2005). Similar results were reported by Kumar and Kanawjia (2012) with cow and buffalo milk.

It can be concluded that there was significant differences (P<0.01) observed in cheese made from varying level of starter culture with respect to sensory characteristics. The buffalo milk Feta type cheese made with 1.5% starter culture had relatively balanced flavour, desirable soft and smooth, body and texture, whereas cheeses with 1.0 and 2.0% culture lacked brand hard body and acidic flavour with weak body defects, respectively. The pH of cheese decreased with the increase in level of culture, while TA, soluble protein and FFA content of cheese increased with increase in level of culture. Therefore, the most acceptable quality of Feta type cheese from buffalo milk with desirable sensory and biochemical characteristics can be made by using 1.5% culture level.

SUMMARY

The role of starter culture and their level have an utmost importance in preparation of buffalo milk feta type cheese. Effect of mesophilic starter culture (NCDC-149) @ 1.0, 1.5 and 2.0% on sensory and bio-chemical characteristics of buffalo milk feta cheeses were investigated throughout the ripening period up to 2 months to select the best level of culture for manufacture of cheese. The sensory scores was significantly higher for cheese made with 1.5% level of culture. The pH of cheese significantly decreased as the level of culture increased, while titratable acidity (TA) of cheese significantly increased as the level of starter culture increased during ripening period. The proteolysis and lipolysis of cheese significantly increased as the level of culture increased during the ripening period. The cheese made with 1.5% starter level was found with comparatively better quality.

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