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Sensory evaluation and consumer acceptability of buffalo curd prepared at 37 °C incubation temperature with various lactic starter cultures under refrigerated storage conditions

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Abstract
The major objective of this work is to determine the sensory properties of lactic cultures inhabiting buffalo curd. Commercially available indigenous starter cultures were used to prepare buffalo milk curd. Buffalo Milk was adjusted to contain 5% fat and 9% SNF levels using the Pearson square method. All the above samples were incubated at 37 °C separately to study the effect of incubation temperature on the sensory properties of buffalo curd. All curd samples were compared with the control curd sample by sensory evaluation to select the best starter culture combination with higher sensory score.
The sensory scores for buffalo milk curd samples prepared using Buffalo milk curd prepared by *Lactococcus lactis* subsp. *lactis* + *Lactococcus lactis* subsp. *cremoris* + *Lactococcus lactis* subsp. *lactis biovar diacetylactis* inoculation and incubated at 37 °C combination of starter cultures were higher than the control and other treatments in terms of flavour, body and texture, colour and appearance, overall acceptability (8.88 ±0.01, 8.76 ±0.01, 8.84 ±0.01, 8.87 ±0.01) on day 0 and lowest flavour, body and texture, colour and appearance, overall acceptability scores (7.52 ±0.01, 7.13 ±0.01, 8.03 ±0.01, 7.50 ±0.01) were observed for buffalo milk curd sample prepared using Buffalo milk curd prepared by *Lactococcus lactis* subsp. *cremoris* inoculation and incubated at 37 °C starter culture on day 0. Results showed that starter cultures, and incubation temperatures significantly influenced the sensory characteristics of buffalo curd.

Keywords: Buffalo milk curd, sensory properties - flavour, body and texture, colour and appearance, overall acceptability

Introduction
Starter cultures and fermentation temperature are key factors affecting sensory characteristics of curd. The starter cultures confer distinct textural and flavour properties and are also used to promote acid development during curd manufacturing process. (McSweeney and Sousa, 2000) [14]. Composition of milk is influenced by species, breed, individuality, nutritional status, health, and stage of lactation of milch animals. (Fox *et al*., 2000) [9]. Buffalo milk is rich in fat, lactose, caseins, calcium, magnesium, and phosphate compared with cow milk (Fundora *et al*., 2001; Ahmad *et al*., 2008) [10, 1] and, because of its chemical composition, buffalo milk offers excellent opportunities for the development of different dairy products (Murtaza *et al*., 2008) [15]. Curd is a fermented dairy product that undergoes a complex series of chemical, bacterial, and enzymatic reactions during ripening (Singh *et al*., 2003; Farkye, 2004) [20, 8], which are responsible for the development of sensory characteristics (Pollard *et al*., 2003; Smit *et al*., 2005; Azarnia *et al*., 2006) [17, 21, 2].
Fermentation of curd is time-consuming lengthy process (Law, 2001; Murtaza *et al*., 2012) [13, 16] use of variety of starter cultures in different combinations (Lee *et al*., 1990), and incubation temperatures (Rehman *et al*., 2000) [18]. Attempts to shorten the fermentation time using a range of ripening systems have had varying degrees of success (Wilkinson, 1993; Law, 2001) [22, 13].
Grading and judging are used extensively for sensory evaluation of dairy products (Bodyfelt *et al*., 1988) [3]; Assessment of the sensory characteristics of curd and various sensory characteristics is an important part of cheese quality evaluation and currently involves the

use of trained sensory panelists or individuals (Downey *et al*., 2005) [6]. A product is evaluated based on the presence or absence of specific attributes and on overall quality score. These quality scores are usually based on the opinions of one individual, and the quality score is subjective rather than specifically defined (Drake *et al*., 2005; Caspia *et al*., 2006) [7, 4].
The study was designed with the objective determine the sensory properties of lactic cultures inhabiting buffalo curd prepared at 37 °C incubation temperature under refrigerated storage conditions and its consumer acceptability.

Materials and Methods
Buffalo milk curd were prepared and incubated at 37 °C using 5 per cent fat and 9 per cent SNF combination by means of pearson square standardization using the starter culture combination listed in table 1.

Sensory evaluation of curd
The panel of judges assessed the coded curd samples at random, according to the methodology described by Indian standards (IS, 1971). Sensory evaluation of samples was carried out with a 12-member and the panelists had a good knowledge on the sensory evaluation of dairy products and participated previously in such evaluations. Curd samples, in the plastic containers were conditioned at room temperature for 15 min before testing. Sensory evaluation of the samples was carried out in the sensory evaluation room under appropriate fluorescent lighting. Each panellist was asked to taste the curd samples labelled with code and rate the sensory parameters on a 9-point hedonic scale.
According to the 9-point structured hedonic scale, the acceptance test was carried out for the attributes of colour and appearance, body and texture, flavour and overall acceptability. For all the attributes, 9-point scale was defined as that the highest value indicates the highest degree of preference. Panellists were also requested to give the scores and comments on a sensory evaluation score card. Water was provided to rinse the palate before and after tasting the sample.
The various starter cultures and their combinations above were used to prepare buffalo milk curd samples with 5% fat and 9% SNF combinations. The sensory scores of buffalo curd samples prepared with various combinations of starter cultures by incubating at 37 °C and stored at refrigeration temperature are shown in Tables 1, 2, 3 and 4 respectively.

Results
The sensory scores of buffalo milk curd samples prepared with various combinations of starter cultures by incubating at 37 °C and stored at refrigeration temperature 5.00 ± 0.5 ºC are shown in Tables 2, 3, 4 and 5, respectively. The various starter cultures and their combinations as in Table 1 were used to prepare buffalo milk curd samples with 5% fat and 9% SNF combinations by incubating at 37 °C.
All buffalo milk curd samples were compared with the control curd sample by sensory evaluation to select the best starter culture combination with extended shelf life.
The sensory scores for buffalo milk curd samples prepared using B1Ll+Lc+Ld *Lactococcus lactis* subsp. *lactis* + *Lactococcus lactis* subsp. *cremoris* + *Lactococcus lactis* subsp. *lactis biovar diacetylactis* inoculation and incubated at 37 °C were higher than the control and other treatments in terms of flavour, body and texture, colour and appearance, overall acceptability (8.88 ±0.01, 8.76 ±0.01, 8.84 ±0.01, 8.87 ±0.01) on day 0 and lowest flavour, body and texture, colour and appearance, overall acceptability scores (7.52 ±0.01, 7.13 ±0.01, 8.03 ±0.01, 7.50 ±0.01) were observed for buffalo milk curd sample prepared using *Lactococcus lactis* subsp. *cremoris* inoculation and incubated at 37 °C (B1Lc) starter culture on day 0. (Table 2-5).
The flavour, body and texture, colour and appearance, overall acceptability scores (6.92 ±0.01, 6.67 ±0.01, 7.05 ±0.01, 7.04 ±0.01) were higher than the control and other treatments for the buffalo milk curd samples prepared using B1Ll+Lc+Ld (Table 2-5) starter culture combinations and incubated at 37ºC on day 6 of storage at 5.00 ± 0.5ºC and whereas flavour, body and texture, colour and appearance, overall acceptability scores were lower than the control and other treatments for the buffalo milk curd sample prepared using Buffalo milk curd prepared by (B1St) *Streptococcus salivarius* subsp. *thermophilus* inoculation and incubated at 37 °C starter culture (3.89 ±0.01, 3.65 ±0.01, 6.16 ±0.01, 4.04 ±0.02) on day 6 of storage at 5.00 ± 0.5 ºC.

Discussion
Buffalo Curd sample containing *Lactococcus lactis* subsp. *lactis*, *Lactococcus lactis* subsp. *cremoris* and *Lactococcus lactis* subsp. *lactis* biovar *diacetylactis* starter cultures had significantly high sensory scores compared to other starter cultures. These findings are in close agreement with Shekhar *et al.* (2013) [19] who investigated about the effect of different cultures on physico-chemical and sensory properties of buffalo curd incubated at 30ºC and 42ºC found that the sensory scores of buffalo curd ranged as 6.76 to 7.78.
Khedar *et al.* (2016) reviewed that homofermentative *L. lactis* subsp. *lactis* is able to produce free amino acids, which are believed to stimulate the growth of *Lac.lactis* subsp. *cremoris* strains. Another important species in the mesophilic starter category is the homofermentative *L. lactis* subsp. *lactis biovar diacetylactis*, which is able besides lactic acid production of fermenting citric acid into a number of other compounds such as diacetyl and carbon dioxide. However, at a pH below 5, carbon dioxide and a number of compounds similar to diacetyl, but non-aromatic, are produced.
Homofermentative thermophilic starter microorganisms, like *L. delbrueckii* subsp. *bulgaricus* and *Str. salivarius* subsp. *thermophilus* are used to obtain mildly sour dahi. These starters play an important role in producing a good quality dahi having a firm and uniform consistency with a sweet aroma and acidic taste. (Chatterjee *et al*.,2022) [5]
The lower sensory scores for some starter culture is explained by Ghosh and Rajorhia (1990) [11, 12] recorded that certain curd samples scored low flavor ratings because of the heterofermentative nature of the bacterial culture and known to produce small amounts of formic acid and CO_2 imparts a sharp taste to the product.

Table 1: Starter cultures and their combinations

B1C	Control buffalo curd prepared by curd culture incubated at 37 °C
B1Ll	Buffalo milk curd prepared by *Lactococcus lactis* subsp. *lactis* inoculation and incubated at 37 °C
B1Lc	Buffalo milk curd prepared by *Lactococcus lactis* subsp. *cremoris* inoculation and incubated at 37 °C
B1Ld	Buffalo milk curd prepared by *Lactococcus lactis* subsp. *lactis biovar diacetylactis* inoculation and incubated at 37 °C
B1Ll+Ld	Buffalo milk curd prepared by *Lactococcus lactis* subsp. *lactis and* + *Lactococcus lactis* subsp. *lactis biovar diacetylactis* inoculation and incubated at 37 °C
B1Lc+Ld	Buffalo milk curd prepared by *Lactococcus lactis* subsp. *cremoris* + *Lactococcus lactis* subsp. *lactis biovar diacetylactis* inoculation and incubated at 37 °C
B1Ll+Lc+Ld	Buffalo milk curd prepared by *Lactococcus lactis* subsp. *lactis* + *Lactococcus lactis* subsp. *cremoris* + *Lactococcus lactis* subsp. *lactis biovar diacetylactis* inoculation and incubated at 37 °C
B1Lnl	Buffalo milk curd prepared by *Leuconostoc lactis* inoculation and incubated at 37 °C
B1Lnm	Buffalo milk curd prepared by *Ln.mesenteroides* subsp. *cremoris* inoculation and incubated at 37 °C
B1Lnl+Lnm	Buffalo milk curd prepared by *Leuconostoc lactis* + *Ln.mesenteroides* subsp. *cremoris* inoculation and incubated at 37 °C
B1Lp	Buffalo milk curd prepared by *Lactobacillus plantarum* inoculation and incubated at 37 °C
B1Lpp	Buffalo milk curd prepared by *Lactobacillus paraplantarum* inoculation and incubated at 37 °C
B1St	Buffalo milk curd prepared by *Streptococcus salivarius* subsp. *thermophilus* inoculation and incubated at 37 °C

Table 2: Flavour scores of buffalo milk curd with various starter cultures incubated at 37 °C during refrigerated storage

Storage period in Days	Starter cultures							
	B1C	**B1Ll**	**B1Lc***	**B1Ld**	**B1Ll+Ld**	**B1Lc+Ld**	**B1Ll+Lc+Ld**	**B1St**
0	8.74^{cA}±0.10	8.76^{bA}±0.10	7.52^{eA}±0.10	8.77 ^{bA}±0.10	8.88^{aA}±0.10	7.57^{dA}±0.10	8.88^{aA}±0.10	7.57^{dA}±0.10
2	8.05^{dB} ±0.10	8.04^{dB}±0.10	7.07^{eB}±0.10	8.38^{bA}±0.10	8.28^{cB}±0.10	7.07^{eB}±0.10	8.47^{aA}±0.10	7.07$^{eB\ A}$±0.10
4	6.55^{cC}±0.10	6.05^{dC}±0.10	6.07^{dC}±0.10	7.08^{bB}±0.10	7.09^{bC}±0.10	6.08^{dC}±0.10	7.58^{aB}±0.10	6.57^{cC}±0.10
6	4.97^{bD}±0.10	4.96^{cD}±0.10	4.98^{cD}±0.10	4.97^{cC}±0.10	4.96^{cD}±0.10	4.98^{cD}±0.10	6.92^{aC}±0.10	3.89^{dD}±0.10
8	3.92^{bE}±0.10	3.93^{bE}±0.10	3.92^{bE}±0.13	3.93^{bD}±0.10	3.99^{bE}±0.13	3.99^{bE}±0.13	5.17^{aD}±0.13	2.96^{cE}±0.10
10	2.94^{bF}±0.10	3.00^{aF}±0.10	2.95^{bF}±0.10	2.96^{bE}±0.10	3.00^{aF}±0.10	2.94^{bF}±0.10	3.00^{aE}±0.10	3.01^{aE}±0.10

Mean ± Standard error values from six trials.
Mean values bearing different superscripts in a column differed significantly ($p < 0.01$)
* indicates curd sample with long setting time/poor coagulation effect

Table 3: Body and texture scores of buffalo milk curd with various starter cultures incubated at 37 °C during refrigerated storage

Storage period in Days	Starter cultures							
	B1C	**B1Ll**	**B1Lc***	**B1Ld**	**B1Ll+Ld**	**B1Lc+Ld**	**B1Ll+Lc+Ld**	**B1St**
0	8.68^{cA}±0.10	8.69^{cA}±0.10	7.13^{dA}±0.10	8.70 ^{bA}±0.10	8.72^{aA}±0.10	7.15^{dA}±0.10	8.76^{aA}±0.10	7.15^{dA}±0.10
2	7.63^{dB} ±0.10	7.63^{dB}±0.10	6.65^{eB}±0.10	7.96^{bB}±0.10	7.86^{cB}±0.10	6.65^{eB}±0.10	8.05^{aB}±0.10	6.65^{eB}±0.10
4	6.13^{cC}±0.10	5.54^{dC}±0.10	5.55^{dC}±0.10	6.66^{bC}±0.10	6.67^{bC}±0.10	5.67^{dC}±0.10	7.16^{aC}±0.10	6.15^{cC}±0.10
6	4.54^{dD}±0.10	4.63^{cD}±0.10	4.65^{bD}±0.10	4.66^{aD}±0.10	4.65^{bD}±0.10	4.66^{aD}±0.10	6.67^{aD}±0.10	3.65^{eD}±0.10
8	3.63^{bE}±0.10	3.63^{bE}±0.10	3.65^{bE}±0.13	3.66^{bE}±0.10	3.67^{bE}±0.13	3.67^{bE}±0.13	4.87^{aE}±0.13	2.87^{cE}±0.10
10	3.05^{bF}±0.10	2.69^{cF}±0.10	2.65^{cF}±0.10	2.66^{cF}±0.10	2.68^{cF}±0.10	2.66^{cF}±0.10	3.72^{aF}±0.10	2.69^{cE}±0.10

Mean ± Standard error values from six trials.
Mean values bearing different superscripts in a column differed significantly ($p < 0.01$)
* indicates curd sample with long setting time/poor coagulation effect

Table 4: Colour and appearance scores of buffalo milk curd with various starter cultures incubated at 37 °C during refrigerated storage

Storage period in Days	Starter cultures							
	B1C	**B1Ll**	**B1Lc***	**B1Ld**	**B1Ll+Ld**	**B1Lc+Ld**	**B1Ll+Lc+Ld**	**B1St**
0	8.75^{cA}±0.10	8.77^{bA}±0.10	8.03^{dA}±0.10	8.78 ^{bA}±0.10	8.83^{aA}±0.10	8.14^{dA}±0.10	8.84^{aA}±0.10	8.07^{dA}±0.10
2	8.48^{dB} ±0.10	8.68^{cB}±0.10	7.99^{eA}±0.10	8.66^{cA}±0.10	8.77^{bA}±0.10	8.08^{eA}±0.10	8.77^{aA}±0.10	8.02^{eA}±0.10
4	7.52^{bC}±0.10	7.02^{dC}±0.10	7.05^{dB}±0.10	7.03^{dB}±0.10	7.04^{dB}±0.10	7.03^{dB}±0.10	7.53^{aB}±0.10	7.22^{cB}±0.10
6	6.87^{bD}±0.10	5.74^{cD}±0.10	5.81^{dC}±0.10	5.80^{dC}±0.10	5.79^{dC}±0.10	5.85^{dC}±0.10	7.05^{aB}±0.10	6.16^{cC}±0.10
8	5.28^{cE}±0.10	5.25^{cD}±0.10	5.31^{cC}±0.13	5.24^{cD}±0.10	5.26^{cD}±0.13	5.25^{cD}±0.13	6.84^{aC}±0.13	5.83^{bD}±0.10
10	5.14^{cF}±0.10	3.17^{cE}±0.10	5.21^{cD}±0.10	5.15^{cD}±0.10	5.08^{cD}±0.10	5.11^{cD}±0.10	6.06^{aD}±0.10	5.54^{bD}±0.10

Mean ± Standard error values from six trials.
Mean values bearing different superscripts in a column differed significantly ($p < 0.01$)

Table 5: Overall acceptability scores of buffalo milk curd with various starter cultures incubated at 37 °C during refrigerated storage

Storage period in Days	Starter cultures							
	B1C	**B1Ll**	**B1Lc***	**B1Ld**	**B1Ll+Ld**	**B1Lc+Ld**	**B1Ll+Lc+Ld**	**B1St**
0	8.72^{cA}±0.10	8.83^{bA}±0.10	7.50^{dA}±0.10	8.82^{bA}±0.10	8.85^{aA}±0.10	7.52^{dA}±0.10	8.87^{aA}±0.10	7.51^{dA}±0.10
2	8.02^{dB} ±0.10	8.02^{dB}±0.10	7.00^{eA}±0.10	8.33^{bB}±0.10	8.23^{cB}±0.10	7.03^{eA}±0.10	8.43^{aA}±0.13	6.96^{fB}±0.10
4	6.52^{cC}±0.10	6.03^{dC}±0.10	6.01^{dB}±0.10	7.03^{bC}±0.10	7.05^{bC}±0.10	6.03^{dB}±0.10	7.54^{aB}±0.10	6.54^{cC}±0.10
6	6.04^{bD}±0.10	5.03^{cD}±0.10	5.02^{cC}±0.10	5.02^{cD}±0.10	5.03^{cD}±0.10	5.04^{cC}±0.10	7.04^{aC}±0.10	4.04^{dD}±0.13
8	4.02^{bE}±0.10	4.04^{bE}±0.10	4.02^{bD}±0.13	4.03^{bE}±0.10	4.05^{bE}±0.13	4.04^{bD}±0.13	5.26^{aD}±0.13	3.02^{cE}±0.10
10	3.01^{bF}±0.10	3.04^{aF}±0.10	3.01^{bE}±0.10	3.01^{bF}±0.10	3.03^{aF}±0.10	3.01^{bE}±0.10	3.04^{aE}±0.10	3.02^{bF}±0.10

Mean ± Standard error values from six trials.
Mean values bearing different superscripts in a column differed significantly ($p < 0.01$)

Conclusion

Conclusively, the results of the present study suggest that the buffalo milk curd prepared with *Lactococcus lactis* subsp. *lactis*, *Lactococcus lactis* subsp. *cremoris* and *Lactococcus lactis* subsp. *lactis* biovar *diacetylactis* incubated at 37 ºC could be stored at refrigeration temperature for up to 6 days had good consumer acceptability.

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