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Technology and Physicochemical Characteristics of Traditional Dhaka Cheese

¹S. Parvin, ²M.M. Rahman, ²K. Shimazaki and ¹I. Kato

¹Graduate School of Dairy Science, Rakuno Gakuen University,
Ebetsu, Hokkaido 069-8501, Japan

²Laboratory of Dairy Food Science, Research Faculty of Agriculture,
Hokkaido University, Hokkaido 060-8589, Japan

Abstract: This is the first report describing traditional technology of Dhaka cheese preparation upon surveying the study area and also some physicochemical properties of the product. Although Dhaka cheese is usually sold in unripened (fresh) condition; in the present study, an attempt was taken to investigate and compare ripening effects on physicochemical properties including proteolysis pattern of Dhaka cheese either collected from two different places of Bangladesh or prepared in the laboratory (control type Dhaka cheese). The nitrogen distribution pattern, analyzed by the Kjeldahl method showed an increase in nitrogen content and ripening index during ripening. Proteolysis pattern, carried out by Sodium dodecylsulfate polyacrylamide gel electrophoresis (SDS-PAGE) displayed a higher degradation of protein in Dhaka cheese during ripening. Loss of moisture content and increase of pH and titratable acidity during ripening was also observed. However, changes of physicochemical characteristics during ripening significantly vary among the places from where Dhaka cheese collected. This result strongly indicates the microbial biodiversity and generation of short peptides in traditional Dhaka cheese during ripening.

Key words: Dhaka cheese, physicochemical characteristics, proteolysis pattern, ripening

INTRODUCTION

Several types of fermented milk products have been reported to exist throughout the world (Stanley, 1998; Tamime and Marshal, 1997). Among them, cheeses constitute one of the families of milk-derived fermented products having a number of advantages over fresh fermented products. There are many traditional cheeses from different corner of the world have been reported. The traditional food products are often considered as an indication of the cultural heritage of a nation, have persisted over centuries and they often evolved from home manufacture in an artisan and traditional way to large-scale industrial production using specific starter cultures and modern equipment (Oberman and Libudzisz, 1998). However, empirical manufacturing practices and poor sanitary conditions associated with such practices are incompatible with the tighter control enforced by Public Health Offices, a situation that will eventually hamper the global trade of such products (Tavaria *et al.*, 2006).

Dhaka cheese is such type of traditional fermented dairy products in Bangladesh. The cheese was originated from a small village named Austagram of Bangladesh around 100 years ago. Dhaka cheese is manufactured from raw milk of indigenous cow using cattle abomasum as a source of rennet, is produced at home in small scale and is sold in unripened (fresh) condition. Special type of bamboo made basket is used for Dhaka cheese preparation. Like typical traditional fermented dairy products, for the preparation of Dhaka cheese fermentation process relies on the natural microbiota of the milk and the environment. Backslopping, a process wherein a portion of a traditionally fermented milk or sour cream from a earlier batch is used as an inoculum for the new batch, is also practiced sometimes to expedite the fermentation process.

Corresponding Author: Isao Kato, Graduate School of Dairy Science, Rakuno Gakuen University, Ebetsu,
Hokkaido 069-8501, Japan Tel/Fax: +81-11-388-4721

Although Dhaka cheese has long been popular in Bangladesh, no scientific report has been found on this traditional product. Moreover, standardization of this traditional product is yet to be defined. Microbial biodiversity has recently been reported in many traditional cheeses (Ouahghiri *et al.*, 2005; Zamfir *et al.*, 2006; El-Baradei *et al.*, 2007, 2008). Thus, bioactive peptides are expected to generate by the action of natural microbiota present in traditional cheese. Since Dhaka is sold in unripened condition; it would be of great importance to study the biochemical changes in this traditional fermented dairy product during ripening. Therefore, the present study was carried out to introduce traditional Dhaka cheese for the first time by describing traditional technology and also by analyzing the physiochemical characteristics including proteolysis pattern of Dhaka cheese during ripening. In addition, a control type Dhaka cheese was prepared in the laboratory and compared to traditional Dhaka cheese to observe the diversification of biochemical changes in this traditional product during ripening.

MATERIALS AND METHODS

Survey on Dhaka Cheese

The information on Dhaka cheese technology was collected by surveying the cheese producing area in Bangladesh in the year of 2005. Interview on several cheese makers was carried out to get the reliable data. Moreover, to learn the know-how of Dhaka cheese preparation and also to check the application of protocol obtained during interview, active participation during the preparation of Dhaka cheese was followed.

Collection of Cheese Sample

Cheese samples were collected from two different places of Bangladesh as shown in Fig. 1. The place Austagram (Fig. 1A) was selected considering origin of the cheese and named here as DA. On the other hand, the place Manikgonj (Fig. 1B) is known as one of the milk pocket area in Bangladesh and also a large number of families of this area are involved with cheese making. Therefore, Dhaka cheese sample was also collected from Manikgonj and named here as DM. Cheese makers were requested to

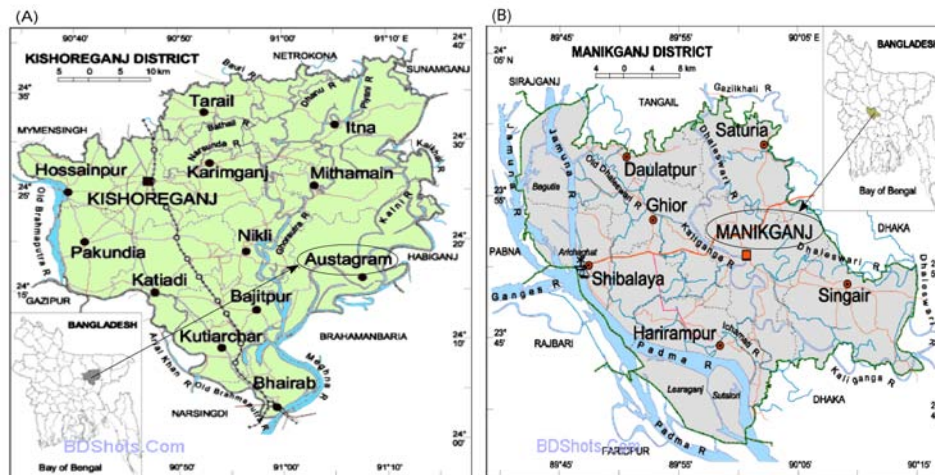


Fig. 1: Dhaka cheese collection area, (A) Austagram, the origin of Dhaka cheese and (B) Manikgonj, one of the milk pocket area in Bangladesh. Source of image: <http://bdshots.com/v/Bangladesh/atlas/>

prepare cheese and samples were collected soon after preparation. A control type Dhaka cheese, named as DC was also prepared in the laboratory using raw milk obtained from Dairy Farm of Rakuno Gakuen University, Ebetsu, Japan, commercial starter culture (Chr. Hansen's, YO-FLEX YC-350) and rennet (Chr. Hansen's, Rennet powder). The protocol followed by the cheese makers was applied for the manufacture of DC. Special type of bamboo made basket usually is used for Dhaka cheese preparation and was obtained from Bangladesh for the preparation of DC. In order to investigate the physicochemical changes occurred by cheese microbes, all samples were kept at 25°C (as this is the average temperature in Bangladesh) for a period of 28 days. At every 7 days interval samples were taken for further analysis.

Physicochemical Properties of Cheese

The moisture percentage of all type cheeses was obtained by drying the samples at 100°C for 90 min using infrared moisture determination balance (FD-600, Kett Electric Laboratory, Tokyo). The ash content was determined by ashing the sample to constant weight at 550°C. The Total Nitrogen (TN) and Water-Soluble Total Nitrogen (WSTN) content was determined using the Kjeldahl method as described by Nihonyakugakkaihen (1999). The Non-Protein-Nitrogen (NPN) and Low Molecular Peptide Nitrogen (LMP-N) content in WSTN was also measured using the Kjeldahl method. High Molecular Peptide Nitrogen (HMP-N) content was measured by subtracting LMP-N content from NPN content. Water Soluble Protein Nitrogen (WSP-N) content was determined by subtracting NPN content from WSTN content. The ripening degree was estimated using the formula $[(WSTN/TN) \times 100]$. The total titratable acidity (SH: Soxhlet Henkel) was determined according to Nihonyakugakkaihen (1999). The pH of the cheese samples was measured using a pH meter (HM-5S, TOA Electronics, Tokyo).

Analysis of Proteolysis Pattern

Sodium dodecylsulfate polyacrylamide gel electrophoresis (SDS-PAGE) (Laemmili, 1970) was carried out to analyze the proteolysis pattern of cheese samples during ripening. The concentration of separation gel was 15%. Each cheese sample (0.001 g) was dissolved with 2×sample buffer (0.2 mL) and then applied (10 µL) on gel. Proteins separated were stained with Coomassie brilliant blue R250.

RESULTS AND DISCUSSION

Technology of Dhaka Cheese

A village of Bangladesh, named Austagram located 250 km North-East of Dhaka city is the origin of Dhaka cheese. Now-a-days, Dhaka cheese is prepared in many places of Bangladesh. The technology was diffused gradually by migrating the cheese makers family members to the other part of the country especially where milk production is abundant as well as price is cheap. However, consumers still prefer to buy Dhaka cheese prepared in Austagram.

The cheese is produced at home using the raw milk of indigenous/crossbred cows sold by the smallholder farmers in the local market. The fourth stomach (abomasum) of cow is collected from local slaughter house followed by washing and sun drying with table salt and is kept until used as a source of rennet. As shown in the Fig. 2, after filtration using indigenous towel raw milk is coagulated with the addition of indigenous coagulating solution. Coagulating solution is prepared by mixing previously prepared abomasum, raw milk and water followed by 12 h incubation at room temperature (Fig. 2). This coagulating solution is locally known as MAWA or medicine water. Before adding, quantity of coagulating solution is determined by checking coagulation ability of the solution by mixing one drop of solution and around 10 drops of milk to be coagulated on the palm. The solution is then slowly added to the milk with gentle stirring by hand and kept undisturbed for 30 min for coagulation. After examining the coagulation condition by entering a bamboo knife, the curd is allowed to stand for

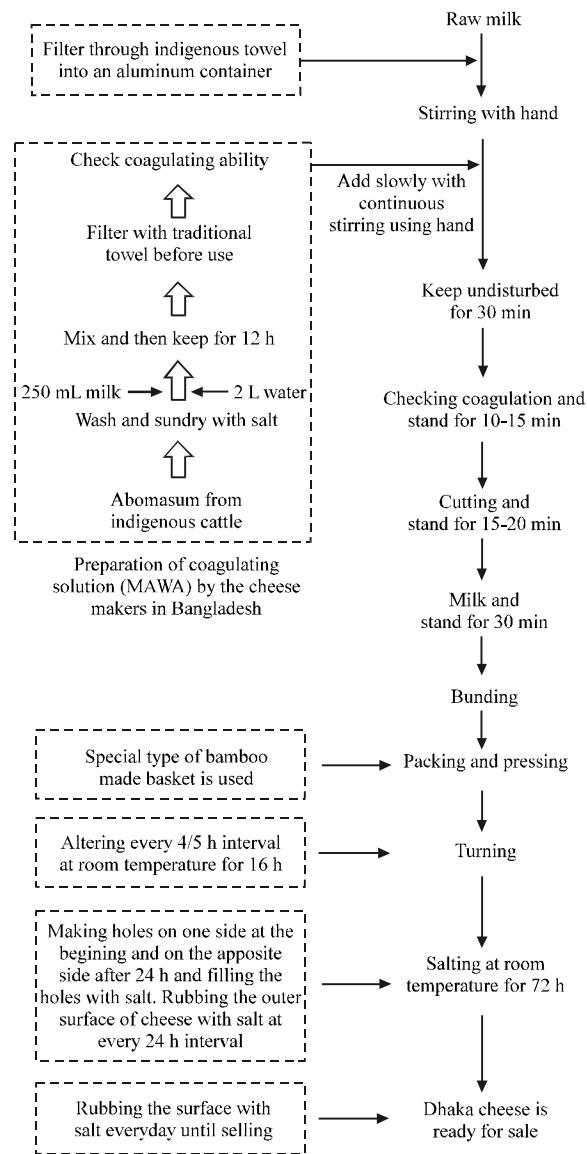


Fig. 2: Schematic diagram of traditional technology for Dhaka cheese preparation

10-15 min. The curd is then cut in square size using a bamboo knife and settled for another 15-20 min. After milling using hand, the curd is kept for further 30 min to drain out the whey. The curd is banded using hand and is separated from whey by curving the container. The curd is then cut with knife and packed in to a special type of bamboo made basket by applying pressure using hand for draining of whey. For proper shape and size turning is made at every 4/5 h interval for a period of 16 h. After sufficient hardening of the curd, salting process using table salt is started. Three holes of 7.5 cm diameter each are made on the center of the upper and lower surface of cheese at an interval of 24 h and salt is plugged into these holes. The outer surface of cheese is also rubbed by salt at every 24 h interval

Table 1: Physicochemical properties of Dhaka cheese during ripening

Type of cheese	Days of ripening	Nitrogen distribution (%)						Chemical composition			
		TN*	WSTN	NPN	LMP-N	HMP-N	WSP-N	Moisture (%)	Ash (%)	Acidity (%)	pH
Austagram	0	3.0	0.36	0.03	0.02	0.009	0.33	41.1	5.72	1.11	5.58
	7	3.1	0.48	0.05	0.04	0.005	0.43	35.9	5.88	1.30	5.59
	14	3.8	0.64	0.07	0.06	0.01	0.57	27.3	6.30	1.83	5.94
	21	4.8	1.40	0.15	0.14	0.002	1.30	23.1	7.32	1.98	6.22
Manikgonj	28	4.5	1.19	0.13	0.12	0.01	1.10	20.9	7.15	3.13	5.88
	0	3.8	0.18	0.02	0.01	0.001	0.16	40.8	5.63	1.19	5.26
	7	3.3	0.22	0.02	0.02	0.002	0.21	39.2	5.07	1.20	5.35
	14	4.7	0.32	0.04	0.03	0.002	0.29	18.3	5.78	1.37	5.59
Control	21	5.3	0.58	0.06	0.06	0.0005	0.52	22.2	6.49	1.94	5.35
	28	5.7	0.46	0.05	0.04	0.002	0.41	17.6	6.74	2.12	5.34
	0	3.3	0.03	0.03	0.02	0.01	0.26	45.0	4.42	0.92	5.62
	7	3.4	0.05	0.05	0.04	0.007	0.45	39.7	4.97	1.08	5.55
Control	14	4.6	0.06	0.06	0.04	0.02	0.53	38.9	6.10	1.44	5.60
	21	5.2	0.11	0.11	0.08	0.02	0.95	28.9	6.47	1.71	5.55
	28	5.2	0.12	0.12	0.09	0.03	0.95	23.5	6.15	1.48	5.84

*TN: Total Nitrogen, WSTN: Water-Soluble Total Nitrogen, NPN: Non-Protein-Nitrogen, LMP-N: Low Molecular Peptide-Nitrogen, HMP-N: High Molecular Peptide Nitrogen, WSP-N: Water Soluble Protein Nitrogen

for 72 h. The cheese is then ready for sale. However, cheese surface is rubbed by salt everyday until selling. It should also be noted that cheese is washed with water before is being appeared to the consumer for sale.

Changes in Chemical Composition

No significant changes of pH were observed in DM and DC type cheese. Conversely, pH value of DA type was significantly increased after 21 days and was again decreased after 28 days of ripening (Table 1). This increase of pH may be due to high degradation of proteins. Presence of mold may also be the cause of such increase of pH. It is reported that presence of *Brevibacterium linens* in cheese caused an increase in pH (5.5-6.0) during ripening (Kato and Ando, 1994). In an artisanal Mexican Fresco cheese, pH values near to 6.3 and 6.0 were also reported (Torres-Llanez *et al.*, 2005). However, the pH values at the beginning and end of ripening of all cheese samples were in the range of 5.26 and 5.88 which was similar to those observed in other cow's milk cheeses (Marcos *et al.*, 1981, 1983; Coulon *et al.*, 1998; Prieto *et al.*, 2000). On the other hand, the titratable acidity was gradually increased with the ripening time in all cheese samples. The evolution of titratable acidity was less in DC and high in DA and DM cheese throughout the ripening period. However, owing to the high titratable acidity values observed, lower pH values would be expected in cheese samples. The titratable acidity-pH relationship is not constant under all conditions and might be influenced by such factors as types of organism, initial apparent acidity and buffer capacities. The apparent discrepancy in the presented data can be explained by any of these factors. During ripening, all Dhaka cheese samples lost moisture with an increase in ash content; however, rapid loss of moisture was observed after 14 days of ripening in DM and DA type and after 21 days in DC type cheese (Table 1).

Proteolysis Pattern

Nitrogen content in Dhaka cheese during ripening is also shown in Table 1. All the nitrogen contents such as TN, WSTN, NPN, LMP-N, HMP-N and WSTN were found to be increased with ripening period. The TN content of DM and DC type cheese samples was little bit higher than that of DA type. Conversely, WSTN content in DA was higher than DM and DC. The LMP-N content of DA type was closer to DC type but higher than that of DM type cheese sample. These data indicate the high degradation of proteins in DA and DC type than that of DM type cheese samples.

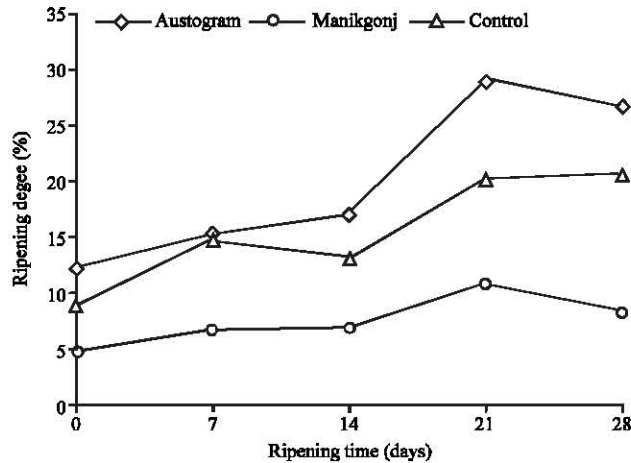


Fig. 3: Changes of ripening index of Dhaka cheese during ripening

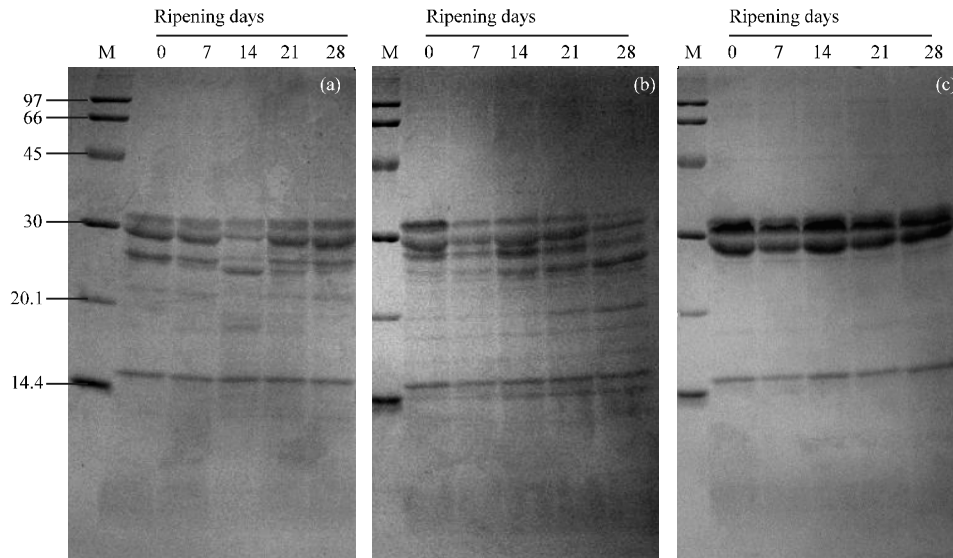


Fig. 4: SDS-PAGE analysis of Dhaka cheese samples at different ripening period, (a) Control, (b) Austogram and (c) Manikgonj

The ripening index of DA type was estimated to be higher than DC type and DC type was higher than that of DM type cheese samples (Fig. 3). This result is correspondence to the findings by SDS-PAGE analysis as shown in Fig. 4. Degradation of protein with the increase of ripening time was observed in DC and DA type Dhaka cheese (Fig. 4a, b) whereas very little degradation was observed in DM type Dhaka cheese (Fig. 4c). The data presented here indicated a higher proteolysis in DA type cheese and also a distinct difference with DM type cheese samples. Proteolysis is one of the main biochemical events that occur during cheese ripening and plays a vital role in the development of flavour (Sousa *et al.*, 2001). Proteolysis is catalyzed by enzymes from the coagulant, milk, starter bacteria, the adventitious nonstarter microflora and in certain varieties, from the secondary microflora (Poveda *et al.*, 2003). Since Dhaka cheese is prepared using the microflora present in raw milk and the source of milk is different between DA and DM type cheese; microbial biodiversity is expected.

CONCLUSIONS

It is generally considered that environmental conditions such as temperature, origin of the milk, processing and sanitary conditions, etc., might have a significant influence on the microbial composition of traditionally made dairy products. High degradation of proteins also is an indication of generating short peptides in DA type cheese. Therefore, existence of some bioactive peptides in Dhaka cheese may be possible. Thus, the present study is expected to encourage more research such as microbial diversity or bioactive peptides in traditional Dhaka cheese.

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