

### 3. MATERIALS AND METHODS

### 3.1. REAGENTS USED

#### Ammonium molybdate solution

6% w/v  $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}\cdot 4\text{H}_2\text{O}$  in distilled water

#### Barium chloride solution

9.88% w/v  $\text{BaCl}_2$  in distilled water

#### p-Dimethylaminobenzaldehyde solution

4% w/v p-dimethylaminobenzaldehyde in chloroform

#### Ferric chloride solution

10% w/v  $\text{FeCl}_3\cdot 6\text{H}_2\text{O}$  in distilled water - used the fresh preparation

#### Glycine-sodium hydroxide buffer

150 ml solution containing 2.4768 g glycine and 1.9359 g NaCl was mixed with 850 ml 0.385 N NaOH to pH 12.8.

#### Methylamine solution

5% w/v methylamine-HCl in distilled water

#### Mixed indicator solution

One part of 0.2% w/v ethanolic methyl red was mixed with five parts of 0.2% w/v ethanolic bromocresol green.

#### Phosphate-phthalate buffer

0.3 g  $\text{KH}_2\text{PO}_4$  and 5.1 g potassium acid phthalate were dissolved in 158.1 N NaOH and diluted to 250 ml to pH 5.3. A few drops of toluene was added.

#### Sodium carbonate-sodium tetrphosphate solution

75 g anhydrous  $\text{Na}_2\text{CO}_3$  and 10 g sodium tetrphosphate are dissolved in water and diluted to 500 ml.

**Sodium hydroxide-sodium thiosulphate solution**

60 g NaOH and 5 g  $\text{Na}_2\text{S}_2\text{O}_3$  were dissolved in distilled water and diluted to 100 ml.

**Sodium sulphite solution**

1% w/v  $\text{Na}_2\text{SO}_3$  in distilled water

**Zinc acetate-phosphotungstic acid reagent**

25.0 g zinc acetate and 12.5 g phosphotungstic acid were dissolved in distilled water. After addition of 20 ml glacial acetic acid, the mixture was diluted to 100 ml.

**Zinc sulphate solution**

22.5% w/v  $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$  in distilled water

**TBA reagent**

0.67 g 2-thiobarbituric acid was dissolved in 75 ml glacial acetic acid. 2 ml concentrated HCl was added in it. Final volume was made to 100 ml with distilled water.

All the chemicals used were of the highest purity grade.

**3.2. EXPERIMENTAL****3.2.1. Survey on churpi**

A survey was conducted in the villages of Bhutan, Sikkim and Darjeeling hills to get detailed information on the types, traditional methods of preparation, modes of consumption, cost of production and market prices of churpi used by the local people.

### 3.2.2. Collection of market samples

Samples of churpi were purchased from different shops of Phuntsholing (Bhutan), Gangtok (Sikkim) and Darjeeling (West Bengal). They were packed in clean stainless steel containers with tightly closed lids. The containers were transported immediately to the laboratory for analyses.

### 3.2.3. Physico-chemical analysis of market churpi

#### 3.2.3.1. Reflectance

Colour of churpi was measured in terms of reflectance at 450 nm using a Reflectometer (Elico, type CL-28, India) against magnesium block which gives 100% reflectance.

#### 3.2.3.2. Chemical analysis

Samples of churpi were broken into small pieces with the help of heavy cutter and pestle. The pieces were then powdered by using a grinder (Bajaj, India).

##### 3.2.3.2.1. Moisture

A sample of ground churpi (ca 2 g) was accurately weighed into a cooled and weighed tared dish, previously heated to  $130 \pm 1^{\circ}\text{C}$ . The sample was allowed to dry for 1 h at  $130 \pm 1^{\circ}\text{C}$  in a hot air oven. The dish was covered while still in oven, transferred to a dessicator, and weighed soon after reaching room temperature. The process of drying, cooling and weighing was repeated until the two successive weights reached a constant value. Moisture content was calculated by subtracting the final weight from the initial weight (AOAC 1990).

### 3.2.3.2.2. Total fat

The method as described in SP : 18 (Part XI) (1981) was followed. Approximately 5 g accurately weighed ground sample was placed in about 10 ml concentrated hydrochloric acid containing in a beaker. Ten ml hydrochloric acid was further added to wet and wash down any particles of the material that might have adhered to the sides. The contents were heated over a burner so that all particles of the material adhering to the sides of the beaker are dissolved in the acid. The contents were finally boiled for 10 min and allowed to cool to room temperature (ca 25°C). The contents were quantitatively transferred to a R'ose-Gottlieb fat extraction tube using about 10 ml of acid as wash liquid. Twenty-five ml of diethyl ether was added to the beaker and transferred the contents to the tube. Twenty-five ml of petroleum ether was added to the beaker and transferred to the tube which was then stoppered with a bark cork, shaken vigorously and allowed to stand until the upper liquid was clear. The ether solution was decanted into a preweighed Erlenmeyer flask. The extraction was repeated using both the solvents in equal parts. The solvent was evaporated completely on a steam bath. The fat was dried in an oven at 100±1°C to constant weight.

### 3.2.3.2.3. Free fat

The procedure recommended by Hall and Hedrick (1971) for milk powder was followed for churpi. Ten g of ground churpi was taken in a 250 ml Erlenmeyer flask. Hundred ml petroleum ether was added in it and shaken with a vertical motion for 10 times. The contents were allowed to settle for 15 min and filtered through

Whatman No. 42 filter paper catching the solvent in a tared Mojonnier fat dish. A second extraction was also exercised following the same procedure. The ethereal layer was evaporated in a hot air oven at  $100 \pm 1^\circ\text{C}$  and the sample weight was determined.

#### 3.2.3.2.4. Free fatty acid

The free fatty acid content in churpi was determined as per IS : 3508 (1966) with minor modifications. An accurately weighed (ca 5 g) ground sample was mixed with 25 ml 95% ethanol neutralized previously by 0.1 N NaOH, using phenolphthalein (0.01% w/v in 95% ethanol). The solution was heated to boiling on water bath and filtered through Whatman No. 1 filter paper. The filtrate was titrated with 0.1 N NaOH until a pink colour persisted.

$$\% \text{ Fatty acid content (as oleic acid)} = \frac{100 \times \text{ml of NaOH} \times \text{N of NaOH} \times 2.82}{\text{Weight of sample (g)}}$$

#### 3.2.3.2.5. Total protein

The micro-Kjeldahl method as described in IS : 4079 (1967) was followed. Approximately 1 g accurately weighed ground sample, taken in a digestion flask, was added with 10 g potassium sulphate, 0.5 g mercuric oxide and 20 ml concentrated sulphuric acid (Sp. gr. 1.84, nitrogen-free). The flask was heated gently until frothing ceased, boiled briskly until the solution became clear, and then continued the boiling for about 1 h. The contents of the flask were cooled to room temperature and made up to volume in a 100 ml volumetric flask with distilled water. Ten ml of the aliquot of the solution was transferred to micro-Kjeldahl distillation flask and made the solution alkaline by 8 ml sodium hydroxide-sodium thiosulphate solution. The flask

was immediately connected to a distillation apparatus and the tip of the condenser was immersed in a saturated solution of boric acid containing 2-3 drops of mixed indicator solution. The distillation was continued until about 50 ml of the distillate was collected which was then titrated against 0.02 N hydrochloric acid till violet colour appeared. A blank was carried out using all reagents except the test material.

$$\text{Protein, \% w/w} = \frac{89.32 (V_2 - V_1) N}{W}$$

Where

$V_2$  = ml of hydrochloric acid used in distillation,

$V_1$  = ml of hydrochloric acid used in the blank test,

N = normality of hydrochloric acid, and

W = weight of sample (g) taken for analysis.

#### 3.2.3.2.6. Water-dispersible protein

Approximately 5 g of accurately weighed ground sample was dispersed in about 50 ml warm (40°C) water. The mixture was transferred to a 100 ml volumetric flask and the volume made upto the mark. The contents were thoroughly mixed and filtered through Whatman No.2 filter paper and the nitrogen content in the known volume of the filtrate was determined using the micro-Kjeldahl procedure as described in section 3.2.3.2.5.

#### 3.2.3.2.7. Tyrosine value

The method as described by Hull (1947) was followed. One g sample was taken in a test tube and 1 ml of distilled water was added followed by 0.72 N trichloroacetic acid while agitating the tube to mix the sample. The tube was stoppered, shaken vigorously and allowed to stand for 10 min before filtering the

contents through Whatman No. 42 filter paper. Five ml of the filtrate was added to a 50 ml Erlenmeyer flask and 10 ml of sodium carbonate reagent was added and mixed thoroughly before 3 ml of phenol reagent was added. The blue colour was measured in a spectrophotometer (Systronics, type 106, India) at 650 nm. A standard tyrosine curve was prepared to convert the absorbance into tyrosine equivalent.

#### 3.2.3.2.8. Lactose and glucose-galactose

The method as described by Nickerson et al. (1976) was followed. One ml of zinc acetate-phosphotungstic acid reagent was added to about 4 g of accurately weighed ground sample. Twenty ml distilled water was added to it and after 10 min the well-mixed content was filtered through a Whatman No. 1 filter paper. The filtrate (0.5 ml) was mixed with 0.5 ml 1 N sodium hydroxide, diluted to 10 ml by adding distilled water, and filtered through a Whatman No. 1 filter paper. Five ml of the filtrate was mixed thoroughly with 5 ml glycine-sodium hydroxide buffer, 0.5 ml methylamine solution and 0.5 ml sodium sulphate solution. The content was heated in a water bath at 65°C for 25 min and cooled immediately in an ice-water bath. Absorbance was read against blank (using water in place of sample) at 540 nm.

For the determination of glucose-galactose, 5 ml phosphate-phthalate buffer was mixed thoroughly with 1.0 ml standard unknown or water (blank) and 5 ml ammonium molybdate solution. The content was heated in a boiling water bath for exactly 15 min and cooled immediately with tap water to stop reaction. Absorbance was read against blank (using water in place of sample) at 710 nm.



### 3.2.3.2.9. Lactic acid

The method recommended by Harper and Randolph (1960) for cottage cheese was used for determining lactic acid in churpi. Ten g ground churpi was macerated with 90 ml distilled water for 5 min in a waring blender at high speed. Twenty-five ml of the mixture was pipetted into a 100 ml Erlenmeyer flask. Ten ml barium chloride solution, 5 ml zinc sulphate solution and 5 ml 0.66 N sodium hydroxide were added in order and mixed. The contents were filtered through Whatman No. 40 filter paper, and 10 ml of the filtrate was taken in a clean dry test tube. Two ml of freshly prepared ferric chloride solution were added and mixed by inversion. The absorbance was recorded in a spectrophotometer at 425 nm against a blank prepared in the same manner replacing churpi with 10 ml cold distilled water. A standard curve was prepared by adding known concentrations of lithium lactate to distilled water to convert the sample reading into its lactic acid equivalent.

### 3.2.3.2.10. pH

A sample (10 g) was mixed with 90 ml carbon dioxide-free distilled water in a waring blender (Bajaj, India) for 1 min. The temperature of the mixture prepared was equilibrated at 25°C and the pH was noted (AOAC 1990) using a pH meter (Systronics, type 335, India).

### 3.2.3.2.11. Titratable acidity

A ground sample (ca 10 g) was accurately weighed and blended with 90 ml carbon dioxide-free hot distilled water for 1 min. The mixture was filtered through Whatman No. 1 filter paper, and 25 ml of the filtrate was titrated with 0.1 N sodium hydroxide

to end point of phenolphthalein (0.1% w/v in 95% ethanol) (AOAC 1990).

$$\% \text{ Titratable acid content (as Lactic acid)} = \frac{100 \times \text{ml of NaOH} \times \text{N of NaOH} \times 0.09}{\text{Weight of sample (g)}}$$

### 3.2.3.2.12. Ash

A ground sample (ca 3 g) was accurately weighed into a previously dried and weighed silica crucible and placed in a hot air oven at  $100 \pm 1^\circ\text{C}$  for 1 h. The dried sample was then ashed in a muffle furnace at  $550 \pm 20^\circ\text{C}$  for 3 h. The crucible was transferred directly to a dessicator, allowed to cool to room temperature and weighed immediately. The process of heating at  $550 \pm 20^\circ\text{C}$  for 30 min, cooling and weighing was repeated until the difference between two successive weighings was  $< 1$  mg. The lowest mass was recorded (IS : 1167 1965).

### 3.2.3.2.13. 2-Thiobarbituric acid

The method described by Keeney and Bassette (1959) was followed. One g of ground sample of churpi was reconstituted with 10 ml of distilled water in a 25 ml volumetric flask. One ml 40% trichloroacetic acid solution was added with gentle rotation. The flask was placed in a water bath at  $70^\circ\text{C}$  for 25 min and cooled immediately in ice bath. The contents were filtered through Whatman No. 42 filter paper. Eight ml of filtrate was transferred to a clean dry test tube and 2 ml of TBA reagent was added. The tubes were placed in  $40^\circ\text{C}$  water bath for 50 min and were cooled to room temperature. The absorbance was measured using a spectrophotometer at 425 nm.

### 3.2.3.2.14. 5-Hydroxymethylfurfural (HMF)

Free plus potential HMF were measured following the method as described by Keeney and Bassette (1959). One g of ground churpi was taken in a 50 ml test tube, 5 ml of 0.3 N oxalic acid was added in it and mixed thoroughly. The tube was covered with inverted 20 ml beaker and placed in boiling water bath for 1 h, after which it was removed and cooled with cold water to room temperature. This heating step was omitted while estimating the free HMF in the sample. Five ml 40% trichloroacetic acid was added in it, mixed and filtered through Whatman No. 42 paper. Four ml of the filtrate was taken into a test tube and 1.0 ml of 0.05 M 2-thiobarbituric acid was added. The tube was placed in a water bath at 40°C for 35 min, and then cooled to room temperature. The absorbance of the solution was measured at 443 nm against blank prepared same as sample, substituting water for churpi in the spectrophotometer. The following regression equations were obtained from the standard curves using standard HMF (Sigma, USA) solution.

$$\text{HMF } (\mu\text{mol/g}) = R \times 250.57 - 7.07$$

(for free HMF)

$$\text{HMF } (\mu\text{mol/g}) = R \times 183.67 - 3.06$$

(for total HMF)

where, R = absorbance

### 3.2.3.2.15. Para-dimethylaminobenzaldehyde reactivity

The method of Kumar and Hansen (1972) was followed. Two ml para-dimethylaminobenzaldehyde solution and 4 ml distilled formic acid were taken in screw capped test tubes. Ground sample (0.1 g) of churpi was added slowly into the reaction tube. The caps were closed and mixed thoroughly. Tubes were

incubated at  $37 \pm 1^\circ\text{C}$  for 30 min and immediately cooled at  $5^\circ\text{C}$ . The cold contents were then centrifuged at  $1000 \times g$  for 10 min. Absorbance of the clear pink supernatant was recorded at 545 nm against a blank prepared from unheated milk. Data were expressed in terms of absorbance.

#### **3.2.3.2.16. Energy value**

The energy value of a sample determined by multiplying its per cent protein, fat and carbohydrate contents by the factors 4.1, 9.3, and 4.1 respectively, and adding all the multiplication values to get kcal ( $4.184\text{cal} = 1\text{J}$ ) per 100 g (De 1980).

#### **3.2.4. Sensory evaluation of churpi**

The representative samples of market churpi procured from Bhutan, Sikkim and Darjeeling were served to a panel of seven judges. The judges were first trained in the assessment of quality attributes and the associated defects in churpi. The samples were presented to the panelist for sensory evaluation using a partially modified 100 point score card (Table 2) of Patil and Gupta (1986). The sensory texture was assessed using a rating scale (Table 3) in the form of a 14 cm horizontal dotted straight line with its left end indicating one parametric extreme (0) and the opposite end indicating the other extreme (100). Scoring was done by indicating the perceived intensity/acceptability by means of small vertical line along the 100 point scale.

#### **3.2.5. Instrumental evaluation of churpi**

Cubical (5 mm x 5 mm x 5 mm) samples of churpi were subjected to texture profile analysis using an Instron Universal testing

**Table 2.** Sensory score card for organoleptic evaluation of churpi

Attributes	Defects	Intensity			Samples			
		Slight	Difinite	Pronounced	A	B	C	D
Flavour (35)	High acid/sour	33	31	29				
	Bitter	25	18	5				
	Rancid	18	10	5				
	Oxidized	21	14	7				
	State	28	21	18				
	Mouldy/yeasty	18	7	0				
	Unclean	21	14	4				
	Fermented/fruity	18	10	4				
Body and texture (30)	Weak/soft	15	7	0				
	Mealy/grainy	24	18	12				
	Open	24	18	12				
	Soggy	0	0	0				
Gumminess and chewiness (25)	Rubbery	15	10	5				
	Brittle	12	6	2				
Colour and appearance (10)	Brown	6	4	2				
	Mouldy	0	0	0				
	Unnatural	2	0	0				
Total score (100)								

Date :

.....

Signature of the Judge

**Table 3.** Unstructured scale for texture evaluation of churpi

To the panelist:

Kindly evaluate the given samples for different properties using the scales given below. To indicate your judgement, make a vertical line along the scale and give the respective sample number against the line.

1. Elasticity

Least elastic ..... Extremely elastic

2. Firmness

Too soft ..... Too hard

3. Crumbliness

Least crumbly ..... Extremely crumbly

4. Smoothness

Grainy/rough ..... Extremely smooth

5. Gumminess

Least gummy ..... Extremely gummy

6. Chewiness

Least chewy ..... Extremely chewy

7. Overall textural quality

Undesirable ..... Most desirable

Remarks (if any):

Signature:

Date:

Name:

machine (Model 4301, Instron Limited, UK) fitted with a hundred N load cell and operated in a two bite compression mode with a crosshead speed of 50 mm/min and chart speed of 250 mm/min. All instron measurements with full scale deflection of strip chart recorder were carried out at 15°C. The parameters measured were hardness (maximum force recorded during the first compression cycle, N), cohesiveness (area under curve  $A_2$ /area under curve  $A_1$ ), springiness (width of the downstroke in curve  $A_2$ , mm), gumminess (hardness x cohesiveness, N) and chewiness (gumminess x springiness, N.mm).

### 3.2.6. Preparation of laboratory samples

Based on the idea of traditional methods of its preparation, churpi was made in the laboratory.

#### 3.2.6.1. Materials

##### 3.2.6.1.1. Milk

Pooled cow milk was obtained from the Himalayan Co-operative Milk Producers' Union Limited, Matigara. All raw milk samples (4 to 5 h-old) had pH of 6.6 to 6.7, fat content of 3.5% to 4.0% and total solids of 11.5% to 12.0%.

##### 3.2.6.1.2. Skim-milk powder

For standardization of milk to desired solids-not-fat(SNF) content, the skim-milk powder produced by the Kaira District Co-operative Milk Producers' Union Limited, Anand, Gujarat was used.

### 3.2.6.1.3. Coagulants

Citric acid (SD Chemicals, Bombay), lactic acid (SD Chemicals, Bombay) and tartaric acid (Glaxo laboratories, Bombay) were used as coagulants in desired concentrations.

### 3.2.6.2. Processing conditions

#### 3.2.6.2.1. Standardization

Cow milk was separated by a mechanical cream separator (Kamdhenu, Sinhal Metal Industries, Bombay; capacity 60 l/h) (Fig.2). Skim milk obtained was standardized to desired fat and SNF levels by adding fresh cream and skim milk powder. Total solids content was determined with 5 g milk following the method as described in section 3.2.3.2.1. Fat in milk was determined by the Gerber method as described in SP : 18 (Part XI) (1981). Ten ml of sulphuric acid (density 1.807 - 1.812 g/ml at 27°C) was taken into a Gerber butyrometer for milk. A well-mixed sample (10.75 ml) of milk and 1 ml of amyl alcohol were added in it. The butyrometer was stoppered and the content was mixed by shaking at an angle of 45° until all the curd was dissolved. The butyrometer was kept in water bath at 65±2°C for 5 min. The butyrometer was then centrifuged at 1000 x g for 5 min and placed again in the water bath at 65±2°C for another 5 min. The fat content was directly read from the scale on the butyrometer. Titratable acidity of milk or whey was determined by titrating 10 ml of well-mixed sample against 0.1 N sodium hydroxide after adding 1 ml of 0.5% phenolphthalein indicator.





**Fig. 2** A mechanical cream separator in operation

#### 3.2.6.2.2. Heating of milk

Milk taken in a stainless steel container was heated to the desired temperature using a water bath.

#### 3.2.6.2.3. Coagulation

Coagulation of milk was effected within 60 s by the addition of coagulant solution of the required strength and stirring the content continuously. The coagulum-whey mixture was held for required time before removing the whey by straining through a muslin cloth.

#### 3.2.6.2.4. Cooking

The coagulated mass, held in muslin cloth, was transferred to a stainless steel container and cooked with constant stirring to the desired level of moisture (Fig. 3) content using a water bath.

#### 3.2.6.2.5. Pressing

The hot cooked mass wrapped in muslin cloth (Fig. 4) and transferred to a wooden hoop (10 cm x 10 cm x 10 cm) (Fig. 5) and pressed under desired dead weight (Fig. 6) for required time period.

#### 3.2.6.2.6. Drying

The pressed mass was cut into four equal pieces hanged (Fig. 7) over wooden fire, and dried at  $30 \pm 5^\circ\text{C}$  for 40-50 days.

### 3.2.7. Optimization of process parameters

#### 3.2.7.1. Fat level

Skim milk was standardized to 0.1, 1.0, 1.5 and 2.0% w/w fat and



Fig. 3. Cooking of green curd to a stringy mass



Fig. 4. Wrapping of hot cooked mass in muslin cloth

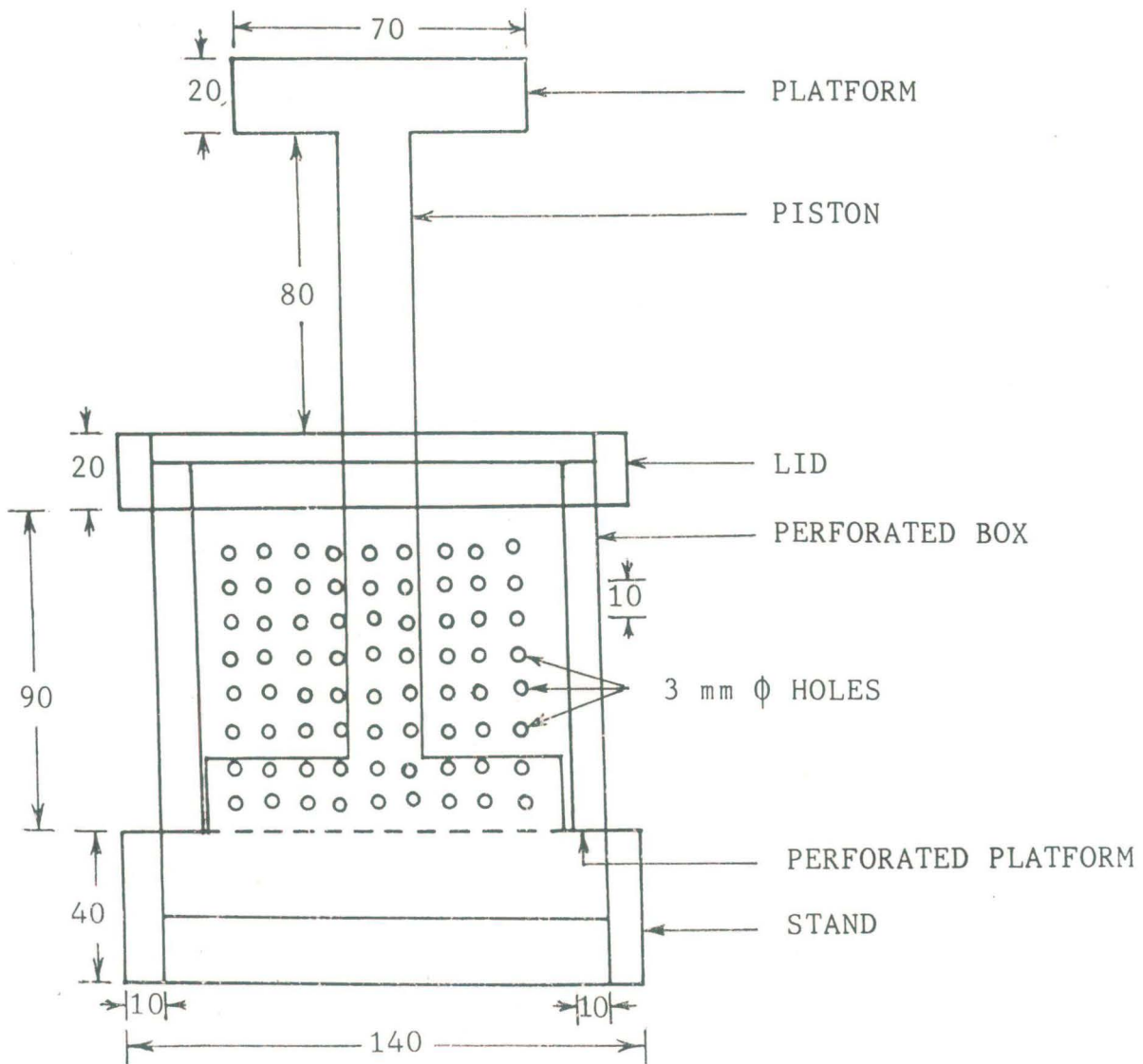
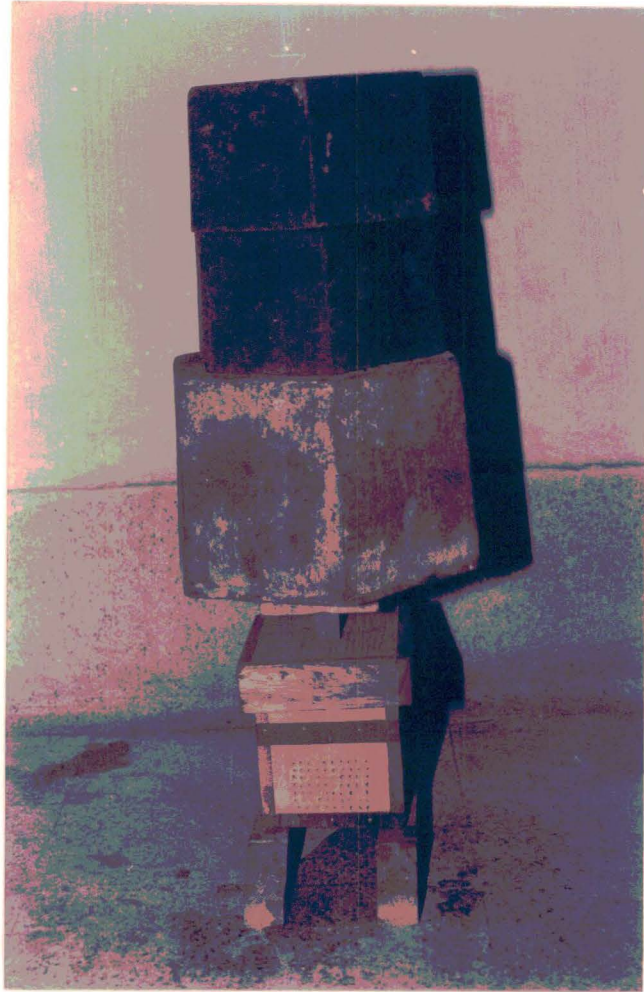
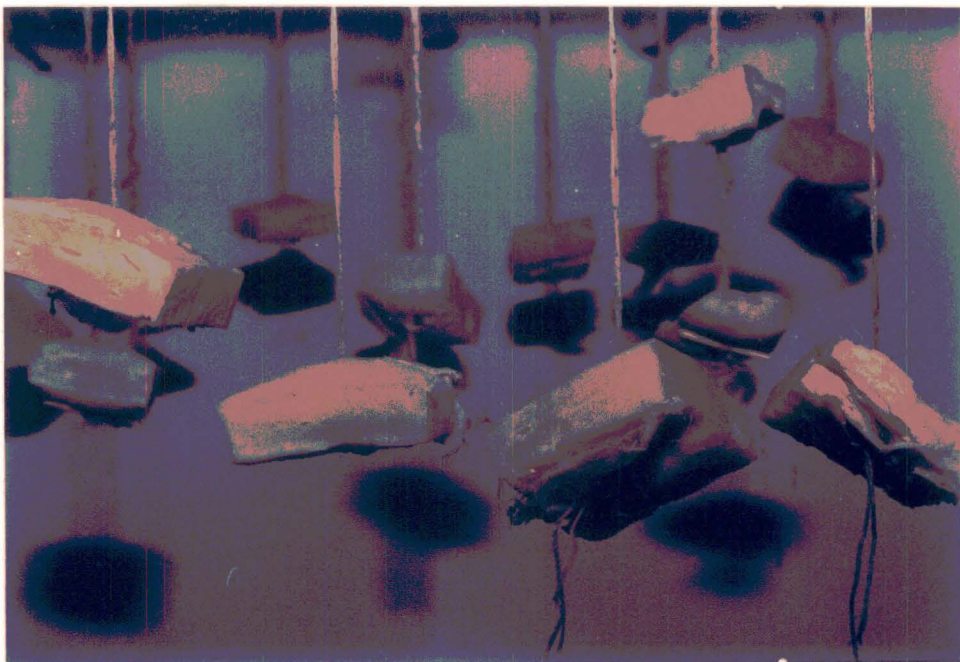


Fig. 5. The churpi press. (all dimensions are in mm)



**Fig. 6.** Wrapped mass placed in a wooden hoop pressed under 90 kg dead weight



**Fig. 7.** Quarters of pressed mass hanging over wooden fire

8.7% w/w SNF using fresh cream and skim milk powder. Milk was heated to 70°C and the coagulation of milk was effected within 60 s using 2% w/v hot (70°C) citric acid solution. Whey was removed by straining through a muslin cloth without holding. The coagulated mass was cooked in a stainless steel container, using a boiling water bath, till the disappearance of free moisture followed by the appearance of an oily surface. The hot cooked coagulum was wrapped in a muslin cloth and pressed at 9 kg.cm<sup>-2</sup> pressure for 12 h. The pressed mass was cut into four equal cubical pieces and hanged over wooden fire (30+5°C) for 40-50 days. The churpi samples prepared from four different fat levels were subjected to various chemical, sensory and instrumental analyses.

#### 3.2.7.2. Temperature of coagulation

Milk standardized to 1.0% w/w fat and 8.7% w/w SNF was heated to 40, 50, 60, 70 and 80°C and coagulated with 2.0% w/v citric acid solution. Rest of the procedure was same as described in section 3.2.7.1. Samples of churpi thus prepared were subjected to instrumental analysis. Total solids in whey, and per cent yield and moisture content of churpi were also determined.

#### 3.2.7.3. Method of straining

Milk standardized to 1.0% w/w fat and 8.7% w/w SNF was heated to 70°C and coagulated at this temperature with hot (70°C) 2.0% w/v citric acid solution. The coagulum-whey mixture was held for 0, 5, 10 and 15 min before removing the whey by straining through a muslin cloth. The rest of the procedure was same as described in section 3.2.7.1. The samples of churpi thus prepared were subjected to instrumental analysis. Moisture content in churpi, per cent yield of churpi, total solids in whey and total solids

recovery were also determined.

#### 3.2.7.4. Strength of citric acid

Milk standardized to 1.0% w/w fat and 8.7% w/w SNF was heated to 70°C and coagulated at this temperature with hot (70°C) 1.0, 2.0, and 3.0% w/v citric acid solution. Rest of the procedure was same as described in section 3.2.7.1. The samples of churpi thus prepared were subjected to instrumental analysis. Moisture content and per cent yield of churpi and total solids in whey were also determined.

#### 3.2.7.5. Type of coagulant

Milk standardized to 1.0% w/w fat and 8.7% w/w SNF was heated to 70°C and coagulated with hot (70°C) 2.0% w/v lactic acid, 2.0% w/v citric acid, 1.5% w/v tartaric acid and 1.0% w/v citric acid in sour whey. After coagulation, the whey was strained immediately through a muslin cloth. The rest of the procedure for preparation of churpi was same as described in section 3.2.7.1. The samples of churpi thus prepared were subjected to sensory and instrumental analysis. Moisture content in churpi, per cent yield and total solids in whey were also determined.

#### 3.2.7.6. Cooking of green curd

Milk standardized to 1.0% w/w fat and 8.7% w/w SNF was heated to 70°C and coagulated with 2.0% w/v citric acid solution (70°C) and strained immediately through a muslin cloth. In the first case of this standardizing procedure, the coagulum was not heated but pressed at  $9 \text{ kg.cm}^{-2}$  pressure for 12 h and the pressed mass was hanged over wooden fire for drying. In subsequent cases, coagulum was heated in a stainless steel container by using boiling water

bath for 5, 10, 15, 20 and 25 min. The hot coagulum was pressed at  $9 \text{ kg.cm}^{-2}$  for 12 h, the pressed curd was cut into four equal pieces and allowed to dry over wooden fire. The moisture ratio was calculated by the formula

$$\text{MR} = \frac{\% \text{ moisture after cooking}}{\% \text{ moisture before cooking}}$$

The samples of churpi thus prepared were subjected to subjective and objective textural analysis and p-DMAB reactivity.

### 3.2.7.7. Pressing condition

Experiments were conducted at 7, 9 and  $11 \text{ kg.cm}^{-2}$  pressures, and pressing time intervals of 2, 4, 6, 8, 10 and 12 h with the hot cooked mass as described in section 3.2.7.1. Moisture ratio at the onset of drying of the samples was determined as follows :

$$\text{MR} = \frac{\% \text{ moisture after pressing}}{\% \text{ moisture before pressing}}$$

Samples were analysed for instrumental parameters.

### 3.2.7.8. Drying behaviour as effected by size of churpi

Churpi prepared as described in section 3.2.7.1. was cut into different sizes (10 cm x 10 cm x 2.5 cm; 5 cm x 10 cm x 2.5 cm and 5 cm x 5 cm x 2.5 cm) which were dried under identical condition. The maximum and minimum drying temperatures and relative humidity at different days of drying were recorded.

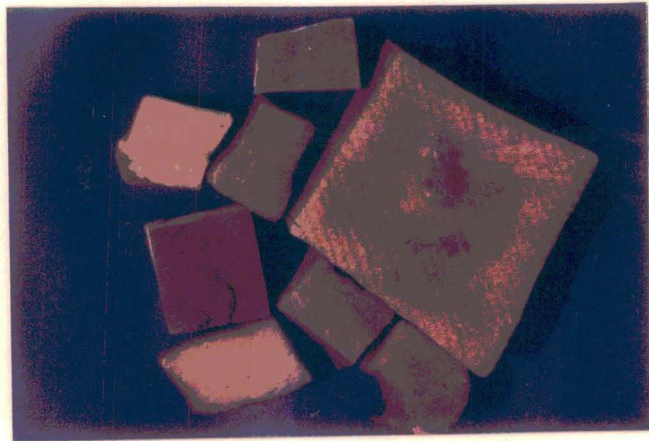
### 3.2.8. Consumer response to laboratory-made churpi

Samples of churpi prepared under optimized conditions (Fig. 8) and the best quality market samples (Fig. 9) were served to (approximately 10 g of each sample) each of 200 consumers included in the study. Individuals, habituated in eating churpi, were randomly selected from among the staff of the institute. Score card used for this study is presented in Table 4.





**Fig. 8.** Churpi prepared under optimized conditions



**Fig. 9.** Market churpi

**Table 4.** Score card for consumer survey of churpi

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Name: \_\_\_\_\_

Age: \_\_\_\_\_

Sex: Male \_\_\_\_\_ Female \_\_\_\_\_

Please answer the following questions:

1. How often do you eat this product?

Several times a week \_\_\_\_\_

Several times a month \_\_\_\_\_

Several times a year or never \_\_\_\_\_

2. When do you eat this product?

With meal \_\_\_\_\_

Between meals \_\_\_\_\_

As and when desired \_\_\_\_\_

On specific occasion \_\_\_\_\_

(please specify)

Testing procedure

You will receive two samples numbered "1" and "2". Taste both the samples and indicate your preference.

Preference: (Check only one of the following)

I prefer sample 1 over 2

Extremely \_\_\_\_\_

Very much \_\_\_\_\_

Moderately \_\_\_\_\_

Slightly \_\_\_\_\_

I prefer sample 2 over 1

Extremely \_\_\_\_\_

Very much \_\_\_\_\_

Moderately \_\_\_\_\_

Slightly \_\_\_\_\_

Reasons for preference: I prefer sample \_\_\_\_ because

It has better flavour \_\_\_\_\_

It has better texture \_\_\_\_\_

It has better colour \_\_\_\_\_

It has better appearance \_\_\_\_\_

Overall sensory quality is better \_\_\_\_\_

Signature \_\_\_\_\_

### 3.2.9. Changes in sensory attributes and physico-chemical parameters during manufacturing churpi

Churpi was prepared under optimized process conditions. The observations were started immediately after removal of the pressed curd and proceeded at every 7 days interval till the desired quality of the product was reached. A sample of curd was prepared by mixing the representative portion of the product in a mortar and pestle. The mass was then subjected to various sensory and intrinsic chemical analyses.

### 3.2.10. Cost of churpi production

The cost of production of churpi was calculated for 100 l of cow milk per day. The factors included in the costing are the cost of raw material like milk, variable costs such as labour, fuel, testing, packaging, detergents, rent on building, depreciation on fixed costs, interest on capital investment, interest on variable costs and miscellaneous expenditure. The output of churpi in kg and the cost of surplus fat realized from standardization were taken into consideration.

### 3.2.11. Statistical analysis

Data on compositional parameters, sensory scores and subjective and objective textural properties were analysed using the randomized block design (Snedecor and Cochran 1967). The chemical, sensory and instrumental data were subjected to analysis on Wipro 386 Computer System in order to develop correlations and multivariate linear and log-linear (power function) relationship (Kapsalis and Moskowitz 1979; Moskowitz 1981) as follows:

$$S = k_0 + k_1 I_1 + k_2 I_2 + \dots \dots \dots k_n I_n$$

$$S = K_0 \cdot I_1^{k_1} \cdot I_2^{k_2} \dots \dots \dots I_n^{k_n}$$

where,

S = dependent variables,  $I_1, I_2 \dots \dots \dots$  = independent variables, and  $k_0, k_1 \dots \dots \dots$  are constants compounded from the data. Exponential relationships were described by the model  $y = \exp(-k_1 x^{k_2})$ , where  $k_1$  and  $k_2$  are constants and were determined by least square regression of experimental data (Raghavarao 1983). The model suggested by Page (1949) was used to express the rate of drying of churpi.