PROCESS ALTERATION
IN
SHRIKHAND TECHNOLOGY

THESIS
SUBMITTED TO THE KURUKSHETRA UNIVERSITY
FOR THE DEGREE OF
Doctor of Philosophy
IN THE FACULTY OF DAIRYING, ANIMAL HUSBANDRY
AND AGRICULTURE

By

R. S. Patel
M.Sc. Dairying (D.T.)

DIVISION OF DAIRY TECHNOLOGY
NATIONAL DAIRY RESEARCH INSTITUTE
(I. C. A. R.)
KARNAL (Haryana) INDIA
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I certify that the work reported in the thesis entitled, "PROCESS ALTERATION IN SHRIKHAND TECHNOLOGY" was carried out by Shri Revabhai Somabhai Patel, under my guidance as requirement for the degree of DOCTOR OF PHILOSOPHY, in the faculty of Dairying, Animal Husbandary and Agriculture, Kurukshetra University, Kurukshetra.

( BIJOY KUMAR CHAKRABORTY )
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( R.S. PATEL )
Dedicated to

my parents,

wife Bharti, and

daughter Sonal
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APPENDIX

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II Compositional information of various shrikhand formulations.

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VIII Changes in sensory quality of shrikhand obtained from different MSNF systems.
CHAPTER 1

INTRODUCTION
1. INTRODUCTION

Shrikhand is an acid coagulated and sweetened milk product, which is a popular delicacy in the states of Gujarat, Maharashtra and partly in Karnataka. This indigenous dairy product is prepared by lactic coagulation of milk and expulsion of whey from the curd, followed by blending of sugar, flavour and spices. Until recently shrikhand production has been largely of a small scale nature. Recently, mechanized production of Sugam shrikhand, a patented process, has been taken up by the Baroda dairy on a large scale (about 1,000 tonnes/year). The basic advantage of this mechanized process appears to be in the elimination of contamination from human hands, and an overall reduction in the process time. However, there appears to be further scope for improving shrikhand making technology particularly in terms of using alternative sources of milk solids, techniques of acidifications and blending approaches to increase versatility, shorten process time and extend the keeping quality.

Shrikhand being a summer speciality, its manufacture from fluid milk in summer implies diversion of milk from fluid consumption during the lean season. Therefore, preparation of shrikhand from other sources of milk solids which can be conveniently manufactured during flush season, would enable its economic production during summer. Sources of milk solids
like reconstituted skim milk, reconstituted unsweetened condensed milk, and buttermilk etc. appear to be promising in this regard.

In shrikhand making technology, preparation of dahi by lactic fermentation of milk takes about eight hours, a time period too long to promote the development of continuous shrikhand making processes. Application of faster and continuous milk coagulation technology may be of great practical utility in mechanized shrikhand production system.

It would be interesting to study the feasibility of applying fast acid producing cultures and direct acidification of milk with coagulants such as lactic acid, hydrochloric acid and slowly hydrolyzing lactones.

Replacing manual kneading of chakka with the mechanical blending in a planetary mixer has already reduced contamination, extending shelf life of the finished product. Ground sugar used for blending may still be a major source of contamination. Therefore, heat treatment of sugar and/or addition of preservatives may be beneficial for extending the shelf life of shrikhand.

A detailed study on the alteration of certain manufacturing aspects of shrikhand has been studied in order to reduce the manufacturing time and increase the keeping quality of shrikhand.
CHAPTER 2

REVIEW OF LITERATURE
2. REVIEW OF LITERATURE

Shrikhand belongs to the group of fermented and coagulated milk products. The traditional technology of shrikhand making involves (1) the coagulation of milk by fermentation with starter culture to obtain acid curd or dahi, (2) preparation of chakka by the drainage of whey from the curd suspended for about 6 to 8 hours in muslin cloth bags, and (3) blending of additives like sugar, colour, flavouring materials and spices, with chakka to a desired composition and consistency. Published work on the technology of shrikhand is rather limited. However, considerable information is available on the unit process such as preparation and drainage of curd that are relevant to the technology of shrikhand and quarg making. This review briefly discusses (a) the technology of curd formation by lactic fermentation and direct acidification for shrikhand and other similar products, (b) drainage of whey, (c) manufacture of shrikhand, (d) quality of shrikhand, and (e) methods of enhancing the shelf-life.

2.1 CURD FORMATION

Curd for shrikhand making is traditionally prepared by lactic fermentation. However, in recent years acid curd required for several other dairy products, particularly cottage cheese, has also been prepared by the method of
direct acidification. The basic aim of both the methods is to attain a typical smooth gel structure rather than an amorphous proteinaceous precipitation. Several operational parameters control the quality of curd formed by either of these processes.

2.1.1 CURD FORMATION BY LACTIC CULTURE

Lactic fermentation essentially implies the process of a gradual increase in lactic acidity as a result of lactose metabolism of starter bacteria in milk. Formation of acid curd is often a prerequisite to the manufacture of a large number of dairy products such as cottage cheese, yoghurt, dahi and shrikhand. However, the type of lactic acid curd differ on the basis of their use in various dairy products. For example, acidity of the curd set for the preparation of American cottage cheese do not usually exceed a value of 0.49 to 0.60% Lactic Acidity (LA), whereas curd for yoghurt usually has a Titrateable Acidity (TA) 1.0% or more. While high heat treatment of milk for cottage cheese curd is considered undesirable, such a treatment is necessary for the setting of yoghurt curd. For the preparation of dahi and shrikhand, the traditional practice has been to use boiled or similarly high heat treated milk for the development of mixed type multiple strains of lactic cultures to an acidity of 1.0% and above. Although the various aspects of manufacturing Indian dahi have been reported (Srinivasan and Banerjee, 1946; Laxminarayana and Iya, 1952; Rangappa and Achaya, 1974; Gandhi and Jain, 1977;
Chakraborty et al., 1981), relatively less attention has been paid to the formation of lactic acid curd for shrikhand making.

The basic requirement of lactic acid curd for the manufacturing of shrikhand must be satisfied in terms of attaining a desired level of curd firmness, acidity, and aromatic traits along with an optimum rate of whey drainage. These qualities of lactic curd are influenced by the type, composition and treatment of milk, type of starter culture and incubation conditions.

2.1.1.1 Type, composition and treatment of milk

Although cow, buffalo and goat milk is produced in large amount in our country, generally cow and buffalo milks are used for shrikhand. The quality of the curd prepared from cow or buffalo milk depends on their inherent characteristics.

2.1.1.1.1 Type and composition of milk

The compositional differences between cow and buffalo species have been reviewed (Laxminarayana and Dastur, 1968; Ganguli, 1974). It has been well established that the curd from buffalo milk is firmer than the curd obtained from cow milk. The high curd tension of buffalo milk dahi is due to the higher levels of SNF (Ganguli and Menon, 1971), and calcium (Yadav and Singh, 1973; Sindhru and Roy, 1973).

Buffalo milk contains 1 to 2 times higher fat content than cow milk. As a result of this the curd acquire some
mellowness. However, the effect of higher fat level on curd gets offset by high calcium, larger micellar size of casein and more SNF content. In addition, the proportion of micellar casein and β-casein is more in buffalo milk than cow milk. Higher rate of syneresis has been observed in rennet curds prepared from buffalo milk (Ganguli, 1974). This may also hold true for lactic curd from buffalo milk.

It is a common practice to make use of whole or standardized milk either from cow or buffalo, singly or in combination for curd preparation, and its subsequent use in shrikhand making. Upadhaya and Dave (1977) reported that use of standardized milk for dahi making yielded smooth body but noticed higher fat losses in whey. Patel and Chakraborty (1981) observed a loss of 0.20 to 0.35% fat in whey obtained from standardized milk (5% fat) curd.

Aneja et al. (1977), Upadhyay (1981) and Miyani (1982) preferred the use of skim milk to whole milk for making dahi for the manufacture of shrikhand. By using skimmilk, not only fat losses were eliminated but faster moisture expulsion and less moisture retention in the curd were achieved.

Considerable attention has been given to the effect of solids content of skimmilk upon the quality of acid curds as required for cottage cheese (Angevine, 1953; Hedrick, 1953; Cordes, 1959; Silanteva et al., 1976) and yogurt (Galesloot, 1968; Ashton, 1963; Emmons and Tuckey, 1967; Davis, 1973; Dordevic et al., 1973; Zmarlicki et al., 1974; Robinson and Tamine, 1975). Higher SNF in milk gives a curd of firmer
body, reduces curd loss in whey resulting in a higher yield. Aneja et al. (1977) reported 2% increase in chakka yield when SNF level was increased from 10 to 11%. However, further increase in SNF level resulted into powdery flavour in the finished product.

2.1.1.1.2 Heat treatment of milk

Heat treatment of milk usually improves its value as a medium for starter organisms and other lactic acid bacteria and also reduces the curd tension. The application of heat to milk results in the destruction of certain heat labile inhibitors as well as many of the competing micro-organisms that are present in raw milk (Babel, 1959; Speck, 1962; Parry, 1974; Feldstien and Westhoff, 1979). Severe heat treatments can result in alterations of proteins in milk to favour the growth and activity of starter culture. One such alteration which will favour the growth and action of starter cultures is the production of free sulphydryl groups (Davies et al., 1978).

The heating of milk at 90°C for 16 seconds, increased the yield of chakka by 5% as compared to that when the milk was heated at 70°C for 16 sec, which was attributed to the precipitation of heat denatured whey proteins. Aneja et al. (1977) and Ott et al. (1979) observed that heating of milk at 90°C/30 min increased the tvorog yield by 15-20% due to co-precipitation of α-lactalbumin and β-lactoglobulin, and it was claimed that the consistency as well as the nutritive value and hygienic qualities of tvorog were improved thereby. Kroger (1981) observed that double heating of
cheese milk at 95-98°C and the coagulum to 60°C, before separating the curd resulted in 50% recovery of whey proteins. Heating of milk to 80-87°C or 96°C for 0.5-1.0 min considerably increased the yield of curd with a corresponding reduction of about 25-30% in whey protein content of the curd whey. The β-lactoglobulin was more readily denatured than α-lactalbumin and β-lactoglobulin-β than β-lactoglobulin A (Puhan and Plueter, 1974).

2.1.1.2 Starter culture

Traditionally small amount of previous day's curd or whey used to be the source of lactic culture required for setting the curd. However, this method of curd setting may not suit industrial requirements as there is a considerable risk of uncontrolled fermentation. The use of contamination-free lactic culture in a desired state of activity is an essential prerequisite for the manufacture of fermented milk products including Shrikhand. The basic role of starter culture is to supply a large number of desirable microorganisms in their optimum state of activity. This is attained in a starter culture by promoting the growth of culture microorganisms under optimum conditions of incubation time and temperature, and nutrition supply in the culture media (Hammer and Bable, 1957; Foster et al., 1958; Mocquot and Hurel, 1970; Lloyd, 1971; Stadnouders, 1975; Tamime and Dieleman, 1980). For this purpose modern dairy laboratories obtain and propagate dairy cultures as mother culture, intermediate culture and bulk culture for ultimate utilization in the
manufacture of fermented products. Although through this practice it has been possible to obtain a desired level of activity of the starter culture, propagation of starter culture in the laboratory is not entirely free from problems of contamination, loss of activity, mutation of strains, loss of strain balance and bacteriophage attack (Lloyd, 1971; Cox and Lewis, 1972; Christensen, 1972; Lawrance et al., 1976). In recent years several advances in the technology of starter manufacture have been aimed at producing culture concentrates for direct application in the vat, thus eliminating the need for culture propagation. A number of workers have dealt with the advances in starter technology including those concerned with starter concentrates, frozen as well as freeze dried (Baumann and Reinbold, 1964, 1966; Lattey, 1968; Lloyd, 1971; Lawrance and Pearce, 1972; Cox and Lewis, 1972; Osborne and Mabb, 1974; Speakman et al., 1974; Chapman, 1978; Cox et al., 1978; Jespersen, 1977; Wigley, 1977).

For lactic fermentation in milk systems, starter culture microorganisms are selected on the basis of their ability to utilize lactose and produce lactic acid and/or flavour compounds in a manner that is the characteristic of a particular fermented product. In general, streptococci and lactobacilli are the two major groups of microorganisms that are used in fermented milk products for acid production (Bautista et al., 1966; Iyengar et al., 1967; Davis, 1975; Hard and Ramirez, 1975; Stadhouders, 1975; Sharpe, 1979; Tamime and Grig, 1979). For the purpose of diacetyl-type aroma,
Leuconostoc cultures are often used (Hammer and Bable, 1957; Lindsay et al., 1965; Keenan and Bills, 1968).

Starter cultures are available either as single strain or as multiple strains. The inherent problem of bacteriophage attacking a single strain culture is often circumvented by practising a scheme of using several different single strain cultures in rotation (Lloyd, 1971; Lawrance et al., 1976) and propagating culture in phage resistant media (Anderson and Meanwell, 1942; Whitehead and Hunter, 1947; Lewis, 1956; Czulak and Keogh, 1957; Lawrance and Pearce, 1972). The success of using multiple strain cultures, on the other hand, often depends upon the compatibility of various strains in the culture, their stability, and relative dominance during subsequent propagation. For many applications, multiple strain culture is considered ideal due to its little susceptibility to bacteriophage attack.

Fermented products such as dahi or shrikhand requires the presence of both acid and flavour producing organisms in the starter culture. Several mixed type multiple strains culture satisfactorily meet this requirement (Lindsay et al., 1967; Keenan and Bills, 1968; Sandine et al., 1972; Coğan, 1974; Robinson and Tamime, 1975).

The quality and flavour characteristics of curd formed are influenced by the type and activity of microorganisms present in the starter culture. The activity of starter culture in terms of pH attained, the extent of gas formed, and proteolytic activity exhibited influences the
quality of curd formed (Law, 1981). Similarly, the relative production of diacetyl and other aromatic compounds by the flavour producing culture organisms influence the overall flavour traits of the fermented products (Lloyd, 1971; Law, 1981; Jonson and Petterson, 1977).

The starter cultures exhibit varying rate and extent of lactic acid production. Most of the culture of strepto-cocci variety can produce lactic acid up to a level of 1.0%. Lactobacillus species, on the other hand, exceed this level of lactic acid production and often at a much faster rate. L. bulgaricus and L. acidophilus are typical examples of fast and high acid producing culture organisms (Thomas et al., 1966; Iyengar et al., 1967; Dutta et al., 1971). Increase in rate of acid production has been observed in mixed strains of S. thermophilus and L. bulgaricus in comparison to their individual ability (Accolas et al., 1977).

Besides the type of starter organisms, several other factors also contribute to the rate and extent of acid production. Among these, T5 level in milk, heat treatment given to substrate, incubation conditions and type of media are some of the significant factors. The inhibition of yoghurt culture, particularly L. bulgaricus have been reported by Tramer (1973) in milk system containing above 21% solids. Also, heat treatment of milk sufficient to denature whey proteins and release of free S-S compounds have been shown to stimulate the growth of starter organisms. In addition, heat can destroy many of the competing microorganisms in milk which help to grow starter culture
freely (Green and Jezeski, 1957; Dutta et al., 1973; Davies et al., 1978). The ability to produce lactic acid in the given medium by starter culture is also governed by temperature of incubation. Yoghurt cultures produce acid at a faster rate at 42°C than at 37°C. Also, the lactic cultures produced acid more rapidly in the whole milk and in a mixture of skim milk and buttermilk than either of them alone (Gilliland and Olson, 1963). Yoghurt cultures have been observed to produce more acid in buffalo milk than in cow milk (Thomas et al., 1966; Singh and Ranganathan, 1978).

Although the lactic acid bacteria are usually considered to be only weakly proteolytic (Dutta et al., 1971), they cause a significant degree of proteolysis in many fermented dairy products, adversely affecting their texture and flavour (Koznanski et al., 1960). Proteolysis in yoghurt culture, particularly at 38°C incubation temperature has been found to contribute bitterness in yoghurt while no bitterness was reported at 44°C (Renz and Fuhan, 1975). Some of the flavour producing cultures such as S. diacetylactis have a tendency to produce CO₂ gas. If this gas remains entrapped in the curd it may cause floating of curd, thereby affecting adversely the curd texture characteristic (Law, 1981).

Heterofermentative lactic cultures characteristically produce aroma compounds during the fermentation of milk. Several review articles have been published on different aspects of flavour production by starter bacteria (Lloyd, 1971;
Cogan, 1974; Robinson and Tamime, 1975; Law, 1981). The major flavour components in fermented milk products are acetaldehyde, acetone, acetoain and diacetyl. Main flavouring compounds found in yoghurt is acetaldehyde which is produced by *S. thermophilus* (Bottazzi and Dellaraglio, 1967; Gorner et al., 1968; Shankar, 1977; Lees and Jago, 1977; Robinson et al., 1977; Rasic and Kurmann, 1978; Shankar and Davies, 1978). However, in case of cottage cheese it is attributed to diacetyl. For a balanced flavour in cottage cheese, only 2 ppm of diacetyl is required which is mainly produced by *S. diacetylactis*. The ratio between diacetyl to acetaldehyde should be between 5:1 and 3:1 for a balanced flavour. On the other hand, if relatively more acetaldehyde is produced than diacetyl, "grain" or harsh flavour defect is detected (Lindsay et al., 1965).

Traditional method of using previous days materials for setting fresh shrikand curd has generally resulted in the propagation of a naturally balanced mixed types multiple strain culture that is required to produce about 1% lactic acidity within a period of 12 to 15 hrs at an incubation temperature of 25 to 30°C. Futambekar (1968) made use of *S. lactis* or *S. cremoris* at the rate of 1 to 2%. While Gandhi and Jain (1977) used mixed culture containing *S. lactis, S. diacetylactis* and *Leu. cremoris* in a ratio of 1:1:1. Wagmair et al. (1978) tried six different strains, i.e. *L. bulgaricus, L. casei, S. diacetylactis, L. acidophilus, S. lactis* and *S. cremoris* for shrikhand making.
The product prepared with former three strains was reported to have superior flavour, taste and body characteristics to the product obtained by using latter three cultures. Aneja et al. (1977), Upachayay (1981) and Miyani (1982) used mixed culture for the preparation of shrikhand.

Although rapid production of acidity in curd set at higher temperature should be useable for manufacturing shrikhand, the use of yoghurt cultures for shrikhand has not been reported so far.

2.1.2 AMOUNT OF INOCULUM, TIME AND TEMPERATURE OF INCUBATION

The conditions during incubation should be so selected so as to maintain a desirable balance between the lactic acid and the aroma producing bacteria (Hammer and Sable, 1957). Active culture added at the rate of 1.0% and incubated at 21 to 22°C yields a firm curd with an acidity of 0.8 - 1.0% LA and delicate flavour in 14 to 16 hrs (Nelson et al., 1958; Baisya and Bose, 1975). However, the above parameters are varied to suit individual process. In cottage cheese manufacture, curd can be set either by long, medium or short process. The amount of starter culture, temperature and time of setting in short, medium and long set processes are 5% (32.2°C, 4 hrs), 3% (26.7°C, 8 hrs) and 0.5% (22.2°C, 14-16 hrs), respectively. For the manufacture of shrikhand different workers have reported various time temperature combinations. Milk was inoculated by Ingle and Joglekar (1974) with 2.0% mixed starter culture and incubated at
34°C for 16 hrs. Dahi so formed was firm with a good body and texture. Gandhi and Jain (1977) inoculated milk having 4.0% fat with 5.0% mixed starter culture and incubated it at 30°C for 12 to 14 hrs. Aneja et al. (1977) inoculated 0.5 to 5% mixed starter culture to the milk, incubated it at 30°C for 8 hrs and obtained an acidity 1.0% in dahi. Upadhyay (1981) added 2.0% mixed starter culture to the skim milk and incubated it at 28 ± 2°C for 9 hrs. Dahi so obtained had a 0.85% LA. Addition of 1.5 to 2.0% mixed culture to the skim milk and incubation at 30 ± 1°C for 9 to 10 hrs resulted dahi with a titratable acidity of 0.9 to 1.0% LA (Miyani, 1982).

2.1.3 SYNTERESIS OF CURD

Expulsion of whey from curd is essential for the desirable body and texture of shrikhand. Expulsion of whey is a function of moisture binding tendency of casein, which (Cheeseman, 1962) is influenced by the type and composition of milk, heat treatment and level of Ca++. Whey expulsion is also a function of pH of the coagulum (Fatel et al., 1972).

In traditional shrikhand making technology, milk is placed in muslin cloth bags and left for whey drainage that takes anywhere from 8 to 10 hrs. Ingle and Joglekar (1974) drained whey from dahi by hanging it in muslin cloth for about 5 to 6 hrs. Rapid whey drainage has an added advantage of better control on acidity. Although rate of whey expulsion is known to be a function of temperature, there is no report in the literature concerning the effect of various
temperatures on the rate of whey separation from dahi. However, techniques employing mechanical separation of whey had presumably been performed at a temperature nearer to that at which curd was incubated (Aneja et al., 1977). The whey separation in muslin cloth bag, on the other hand, may undergo varying extent of cooling depending on the size of coagulum in the cloth bag and the ambient temperature.

2.1.4 DIRECT ACIDIFICATION

The quality of fermented dairy products is somewhat unpredictable even when manufactured under optimum conditions. It is more so because of the risk of infections and contaminations of milk supply with the antibiotics. Further, problems associated with the use of starters are time and care required for their maintenance and propagation, and the difficulties encountered in completely automating and rendering continuous the processes which are dependent on starter activity. For these reasons, there has been interest in exploring the possibility of developing chemical and/or biochemical alternatives to starters for the manufacture of acidified dairy products.

The chemicals to be used for direct acidification should be non-toxic, must not react with the constituents of milk to form toxic products or reduce its nutritive value, must dissolve readily in milk, must produce acid at a reasonable rate, must not alter the properties of curd obtained, must not impart off flavour to the finished
product, and should be inexpensive and readily available
(Anon, 1973).

Replacing part or all microbial lactic acid produc-
tion by chemical means or direct acidification is claimed
to have certain advantages - (a) decreased production time,
(b) improved product consistency, and (c) curd formed in a
controlled manner without fermentation, thus eliminating
every day problems associated with microorganisms (Mulder
and Kadema, 1948; Satterness et al., 1978; Gerson, 1978).

Several attempts have been made for manufacturing a
large variety of soft and hard cheeses applying direct acidi-
fication techniques, using hydrochloric, lactic, citric and
phosphoric acids as acidulants. The cheeses manufactured
with the above techniques included cheddar (Mabbitt et al.,
1955; Breen et al., 1964a; Law et al., 1974, 1976), blue
cheese (Shehata and Olson, 1966; Shenata et al., 1967), cottage
cheese (Deane and Hammond, 1960; McNurlin and Ernstrom, 1962;
Ernstrom, 1965; Anon, 1973; Ernstrom and Kale, 1975) and
Mozzarella cheese (Breen et al., 1964b; Larson et al., 1966;
Shehata et al., 1966; Kosikowski, 1977).

To obtain direct acidified curd, the following appro-
aches have been attempted by several authors:

1. Pre-acidification followed by addition of starter
culture.

2. Complete acidification by acids.

3. Use of GDL that slowly hydrolyses to increase
acidity.

4. Combination of acids and GDL.
2.1.4.1 Pre-acidification followed by addition of starter culture

Boddicker et al. (1967) studied pre-acidification by using 2 N HCl followed by addition of starter culture. They reported 25 and 50% reduction in time to reach a pH of 4.7 by the starter culture at 31°C when the pH of reconstituted skim milk was pre-adjusted to 6.0 and 5.0, respectively with 2N HCl at 5°C. Leeder et al. (1965) recommended the use of HCl (to pH 5.4) followed by addition of starter culture in reconstituted skim milk containing 10-12% total solids to achieve 20-25% reduction in setting time. In the manufacture of satisfactory quality of cottage cheese, a reduction of 42% on setting time was achieved by pre-adjusting the acidity of milk to 0.4 to 0.55% LA with phosphoric acid prior to adding 25% low acid non-coagulated starter culture (Martin et al., 1973). Shehata and Olson (1966) reported that blue cheese could be prepared by acidifying milk at 4.4°C to pH 5.6 with HCl followed by heating to 30°C and adding 0.5% lactic starter culture. The coagulation of milk was attained by rennet at the rate of 50 ml/454 kg of milk.

2.1.4.2 Complete acidification by acids

Acidulants like hydrochloric, phosphoric and lactic acids were used by Ernstrom (1965), Martin et al. (1973) and Green (1977) in the preparation of various cheeses.

Quarne et al. (1968) prepared Mozzarella cheese without starter culture by direct acidification of milk to
pH 5.6. They showed that the use of phosphoric acid as acidulant increased SNF recovery slightly as compared to that with hydrochloric acid or lactic acid. Similar results were observed by Breene et al. (1964b) and Green (1977). Shehata et al. (1967) and Quarne et al. (1968) observed that lactic acid tends to give a high moisture in blue and pizza cheeses, while hydrochloric acid and phosphoric acid retained less moisture but better retention of calcium in the curd with consequently higher yields. Shehata et al. (1967) considered lactic acid and citric acid unsuitable for the manufacture of these types of cheeses.

Ernstom (1962) noted that the use of hydrochloric acid or lactic acid at 40°F followed by warming to 70-80°F without agitation yielded a firm curd for the preparation of cottage cheese. Soddicker et al. (1967) noticed 50% reduction in setting time by acidification to pH 5.0 but the curd produced shattered on cutting. However, the shelf life of cottage cheese increased by 30-35% with substantial increase in the yield. Similar results were reported by Gerson (1978).

Pizza cheese of satisfactory quality was prepared using direct acidification techniques as reported by Larson et al. (1966, 1967) too.

Ricotta type cheeses were made by heating whole, partly skimmed or skimmilk or skim/whey mixtures to approximately 88°C and acidifying to approximately pH 5.0 with lactic, acetic or citric acid (Kosikowski, 1977).

Mabbitt et al. (1955) concluded that hydrochloric or
acetic acids as sole acidulants gave unsatisfactory Cheddar cheese which lacked typical cheese flavour. A sour cream of very good quality was prepared using direct acidification techniques by Little (1967) and Anon (1973). Garson and McDermott (1963), Little (1967) and Reddy et al. (1976) reported the production of yoghurt or cultured buttermilk by direct acidification.

2.1.4.3 Use of slowly hydrolysed chemical to increase acidity

Mabbitt et al. (1955) reported that glucono-δ-lactone (GDL) when dissolved in milk hydrolysed slowly to produce gluconic acid and formed a suitable curd for making Cheddar cheese. Dodson et al. (1965) reported that satisfactory Cheddar cheese curd could be made by substituting D-glucono-δ-lactone for lactic starter.

Deane et al. (1960) observed that addition of GDL to milk at the rate of 12.3% of SNF at 20°C and 40°C required 15 hrs and 3.5 hrs respectively for coagulation, while meso lactide added at the rate of 9.8% of SNF at 25°C required 2 hrs and at 37.5°C only 47 min. At higher coagulation temperature the curd was firm. The coagulation temperature above 37°C caused excessive matting of the curd. O'Keeffe et al. (1975) observed the considerable effect of temperature on the rate of hydrolysis of GDL. They also reported that the addition of 3% GDL to milk decreased the pH to 3.75 within 4 hrs at 30°C.

Satterness et al. (1978) used vitex 750 and vitex 850
for preparing cottage cheese from skimmilk. The use of this chemical decreased production time and improved product consistency. The yield of cottage cheese was greater than that with yielding conventional culture process.

2.1.4.4 Combination of acid and slow hydrolysing agents

Mabbitt et al. (1955) demonstrated that sole acidulants (hydrochloric or acetic) gave unsatisfactory cheese. Partial acidification with acid to approximately pH 5.8 coupled with the use of the acidogen GDL (which hydrolyses to gluconic acid in aqueous media) to reduce the pH to approximately 5.2 gave better results. Best quality curd was produced when the pH of milk was reduced to 6.4 by addition of 12% HCl and then to a pH of 3.2 by solid GDL.

O'Keeffe et al. (1975) modified the acidification schedule of Mabbitt et al. (1955). Milk was acidified to pH 6.5 with concentrated lactic acid and after cutting the curd/whey mixture was titrated with lactic acid to simulate the pH decline in a control starter cheese. After complete drainage 13 g GDL/450 g curd was added to reduce the pH to approximately 5.3. A further 3 g GDL/450 g curd was added with the salt for each 0.1 unit by which it was desired to decrease the pH. Ernststrom and Kale (1975) observed that cottage cheese can be prepared by acidification of warm milk to approximately pH 5.1 with food grade acid followed by final acidification to pH 4.5 with GDL.

Coagulation does not occur when the pH of pasteurised skimmilk is adjusted to 5 - 5.05 at 5°C with lactic or
phosphoric acid and heated in a jacketed vat to 31°C. However, coagulation is induced by the addition of sufficient GDL to decrease the pH to approximately 4.7 (Anon, 1973). Use of acid in combination with GDL was also reported by Little (1967), Reddy et al. (1976) and Anon (1973).

2.2 QUALITY OF CHAKKA

The curd mass known as chakka is the base material for shrikhand. It is obtained by the removal of whey from dahi. The quality of shrikhand is largely influenced by the physical and chemical properties of chakka.

Ganguly et al. (1959) analysed the laboratory made chakka and found that it contained 59.50% moisture, 22.4% fat, 1.03% ash, 2.32% acidity (LA), 1.62% nitrogen and 10.30% protein. They recommended that moisture in the chakka should not exceed 65% and fat should not be less than 15%. Another group of workers reported that the laboratory made chakka contained 52.70% water, 8.5% fat, 13-15.6% protein, 0.69-0.78% ash and 2.6% lactose. The corresponding figures for market chakka were 59-81, 12.41-20.8, 10.4-18.4, 0.68-0.97 and 0.3 to 2.1% (Sharma et al., 1975).

The average composition of chakka prepared both from cow and buffalo milk was reported by Mahajan (1971). In cow milk chakka, the fat, moisture, total solids, acidity, pH and volatile acids contents were 3.92, 68.82, 33.18, 0.84, 4.6 and 2.76%, respectively. The corresponding figures for buffalo milk chakka were 14.70, 61.18, 38.82, 0.80, 4.7 and 3.0%.
Upadhyay and Dave (1977) reported that chakka made from whole milk and standardized milk has smooth body but noticed higher fat losses in whey. Skimmilk chakka was rough and dry. Aneja et al. (1977) reported that the yield of chakka depends on heat treatment and total solids content of the skimmilk. The heating of milk to 85°C and 90°C for 16 sec yielded 24 and 25% chakka, respectively. The skimmilk containing 11% total solids produced highest yield.

2.3 MANUFACTURE OF SHRIRKHAND

It is essentially blending of chakka, sugar, cream and flavouring agents such as spices, fruits etc. Quantity of this blend depends on the final composition required in the shrirkhand.

Ingle and Joğlekar (1974) suggested addition of 36% sugar by weight of chakka along with saffron colour (2%), cardamon (1.6%) and charoli (0.8%). The quantity of sugar to be added depends on the acidity of the chakka. The addition of equal quantity of sugar to chakka along with other additives like cream, flavour, colour, fruits and nuts and kneading the contents, resulted in a homogeneous consistency of the shrirkhand (Upadhyay and Dave, 1977).

In traditional shrirkhand making technology, only wire mesh is used for kneading chakka, fat and sugar to obtain desired composition in the final product for getting silky texture in shrirkhand, more kneading of blend through a fine shieve should be used.

Upadhyay (1981) prepared shrirkhand of smooth
consistency by kneading chakka with 80% sugar and calculated quantity of pasteurized cream having 80% fat over a cleaned and sanitized wire mesh. Miyani (1982) also used wire mesh for kneading of chakka, sugar and cream to obtain 6% fat and 40% sugar in the finished shrikhand. However, in an industrial method, Aneja et al. (1977) employed a planetary mixer for kneading or blending of chakka with other additives for preparation of shrikhand.

2.3.1 COMPOSITION OF SHRIKHAND

A wide variation in the composition of shrikhand has been reported by various workers.

Bhattacharya et al. (1972) reported the composition of shrikhand as fat 6.8%, moisture 55-60%, total solids 40-45% and sugar 18.20%. Whereas Joglekar and Manjrekar (1978) reported the composition of shrikhand as 10% fat, 7.5% protein, 28.59% sugar and 39% moisture.

Upadhayay (1981) prepared laboratory samples of shrikhand containing on an average fat 5.16%, protein 6.59%, reducing sugar 1.64%, non-reducing sugar 39.37% and moisture 47.24%.

The shrikhand samples collected from different manufacturers in Gujarat contained fat 1.93%-5.56%, proteins 5.33-6.13%, reducing sugar 1.56-2.18%, non-reducing sugar 52.55-53.76%, total solids 64.34-64.52% and moisture 34.48-35.66% (Upadhayay et al., 1975). Sharma and Zarivala (1978) observed large variation in the composition of market sample
of shrikhand collected in Bombay. The samples were found to contain fat 4.5-11.4%, proteins 3.4-15.70%, lactose 0.66-2.79%, sucrose 38.8-57.1% and moisture 25.4-40.8%.

Shrikhand prepared by Aneja et al. (1977) had on an average 5% fat, 42% sugar and 60% TS. While Upadhyay and Dave (1977) reported the composition of Sugam shrikhand as 40 to 43% moisture, 5 to 6% fat and 45% non-reducing sugar.

2.3.2 SHELF LIFE OF SHRIKHAND

Though shrikhand is a fermented milk product like dahi or yoghurt, it has a higher keeping quality than latters due to higher sugar content (which work as a preservative). The keeping quality of shrikhand largely depends on its initial microflora like yeast and mould and other foreign organisms.

The microbiological examination of market samples of shrikhand conducted by Upadhyay et al. (1975) revealed large variations in total viable count ($2.4 \times 10^4$ to $1.6 \times 10^6$ g), coliform count (0 to 930/g), psycrotropic count (0 to $1 \times 10^4$ g) and yeast and mould count (0 to $10^5$ g). Sharma and Zariwala (1980) reported that during storage of laboratory made shrikhand at 37°C, SPC, spores, lipolytic and proteolytic count decreased, while an increase in YM was observed. The coliform count and staphylococci count remained zero during storage. The same authors reported an increase only in staphylococci count during storage of market shrikhand at 10°C for 45 days. All other types of microbes followed a declining trend in the count. The storage of market samples
at 37°C for 21 days caused an increase in Y&M, spores, staphylococci and proteolytic counts, whereas SPC, coli, lipolytic and acid produces showed a declining trend.

According to Upadhayay (1981) the shrikhand stored at 7 ± 2°C registered an increase in total psychrotrophic acid formers, lactobacilli, proteolytic, lipolytic and yeast and mould counts. Gandhi and Jain (1977) reported the keeping quality of shrikhand to be about 12-14 days under refrigeration. Thereafter mould growth and increase in acidity to 1.02% were observed.

Shrikhand stored at 10 ± 3°C developed off flavour and unpleasant odour in about 40 days, whereas that stored at 37°C spoiled within a period of one week. Deterioration of shrikhand was attributed to the chemical and bacteriological changes in shrikhand during storage (Sharma and Zariwala, 1980).

Upadhayay (1981) observed a decreasing trend in sensory evaluation scores of shrikhand samples during storage at -7 ± 2°C and +7 ± 2°C. The total scores out of 18 of the samples decreased from 16.4 to 13.52 and 16.0 to 11.94 at storage temperature of -7 ± 2°C and 7 ± 2°C, respectively. He reported storage life of shrikhand as 50 days at -7 ± 2°C and as 40 days at 7 ± 2°C. Deterioration in the organoleptic quality of shrikhand was attributed to proteolysis.

2.4 ENHANCING THE KEEPING QUALITY OF FERMENTED PRODUCTS

In spite of low pH, cultured products like yoghurt,
dahi, acidophilus milk and others have a rather short life. However, use of heat treatment or so called thermization and chemical preservatives such as sorbic acid and its salts have been found to extend the shelf life of such products to an appreciable extent.

2.4.1 HEAT TREATMENT OF YOGHURT

Spoilage of dahi, yoghurt, shrikhand and quary samples on storage have been attributed to non-lactic contaminants such as spore formers, micrococci, coliform, yeasts and moulds (Singh et al.,1970; Lang and Lang,1973; Sebele,1978). These undesirable organisms rapidly increase in number when the starter is weak and the ratio of non-lactic to lactic organisms is high. Also containers having a large surface of air in contact with the fermented milk accelerate the process of spoilage. To eliminate these types of problems, thermization of cultural milks were studied (Schulz,1966; Luck and Mostern,1971; Bake,1971; Klupsch,1972; Voss,1973; Lang and Lang,1973; Kroger,1975; Puhan,1976; Davies,1976; Egli and Egli,1977; Obert et al.,1978).

Pasteurization conditions similar to those used for milk are suitable for yoghurt. However, it has been reported that a shorter-time of treatment may be required because of the lower pH of yoghurt and whey separation problem (Stadhouders and Dijk,1974; Dellaolio,1977).

The whey separation problem can be eliminated either by incorporation of stabilizers like pectin, CMC and
Eggwhite or heating yoghurt at 70°C for 30-40 sec and packing it at 55-60°C (Stadhouders and Dijk, 1974; Schulz and Voss, 1977; Della Glio, 1977). However, in the industrial production of pasteurized yoghurt, the product is subjected to flash treatment at 60-70°C after incubation (Rakshy, 1966; Bake, 1971; Klupsch, 1972). Further, Mulcahy (1972) and Obert et al. (1978) observed that pasteurization of yoghurt at 70-75°C for 15-45 sec followed by hot packaging increased the keeping quality of the product at room temperature.

2.4.2 ROLE OF POTASSIUM SORBATE

Sorbic acid and its potassium and sodium salts have found wide application in the food industry as antimycotic agents for the preservation of meats, wine, margarine, butter, cheese products and khoa (Smith and Rollin, 1954; Geminder, 1959; Chakraborty and Kristofferson, 1963). This is the only preservative which has been permitted by U.S. Food and Drug Administration for use in 27 varieties of natural and processed cheese up to a maximum concentration of 0.2% by weight. Smith and Rollin (1954) listed the merits of adding sorbic acid as follows: (a) It is effective inhibitor of mould growth. (b) It is harmless as a component of food because of its outstanding quality in comparison to other fungistatic agents. (c) The mobility or diffusion rate of sorbic acid in cheese is relatively low and its water solubility is only about 0.21 at 20°C. (d) Sorbic acid is light coloured, crystalline material and easy to handle. (e) It does not create unusual difficulties in application to
the wrappers. (f) Cheese stored in sorbic acid wrappers does not change the colour, texture, odour or taste. Under unfavourable conditions, sorbic acid is metabolized by moulds to CO₂ and water. (g) It does not interfere with a selective action against certain bacteria and yeasts. (h) It is non-toxic in dietary constituents. (i) It is resistant to oxidation and sublimation. (j) There is availability of suitable assay technique for determining residual sorbic acid in the samples.

2.4.2.1 Mechanism of sorbate degradation by mould

By and large, the action of sorbic acid is directed primarily against yeasts and moulds including aflatoxin-forming microorganisms (Wallhauber and Luck, 1970). Bacteria are only partially inhibited, the catalase-positive more so than the catalase-negative (Lillard and Vaughn, 1952), the strictly aerobic bacteria the most (York, 1950), and both lactic acid bacteria and clostridia the least (York and Vaughn, 1955). The mechanism of sorbate degradation by moulds has been explained by Mukherjee (1952) as follows:

\[
\begin{align*}
\text{CH}_3\text{-CH}_2\text{-CH}_2\text{-COOH} & \quad \text{CH}_3\text{-CH=CH-COOH} \\
\text{\textup{\uparrow\, butyric acid}} & \quad \text{\textup{\uparrow\, crotonic acid}} \\
\text{\textup{\downarrow\, \beta-oxidation}} & \quad \text{\textup{\downarrow\, \beta-oxidation}} \\
\text{CH}_3\text{-CH}_2\text{-CH}_2\text{-CH}_2\text{-COOH} & \quad \text{CH}_3\text{-CH=CH=CH- COOH} \\
\text{Capric acid} & \quad \text{Sorbic acid}
\end{align*}
\]

It is well established that \(\beta\)-oxidation of capric acid will yield butyric acid, and it is expected that sorbic acid already containing \(\beta\)-unsaturated acids, would be
changed to crotonic acid via β-oxidation. On the other hand, Marth et al. (1966) indicated that a hydrocarbon like odour appeared in sorbate fortified mould culture after 24-36 hrs of incubation. The volatile matter was identified as 1,3-pentadiene by gas chromatographic infrared spectroscopic analysis. The following reactions are believed to occur in the medium:

(i) \[
\text{Potassium sorbate + tartaric acid} \rightarrow \text{potassium tartrate + sorbic acid}
\]

(ii) \[
\text{Sorbic acid decarboxylase} \rightarrow \text{1,3-pentadiene + CO}_2
\]

The results of the above reactions suggest that penicillin decarboxylase convert the inhibitory sorbic acid to the inactive 1,3-pentadiene, and make the growth of mould possible because the effect of inhibiting substance has been neutralized. The authors believed that degradation of sorbic acid follow the same pathway as degradation of fatty acids since mould degrade sorbate as they do with other fatty acids.

2.4.2.2 Inhibitory action of sorbic acid

The inhibitory mechanism of sorbic acid was shown by Melnick et al. (1954) to be due to the inhibition of the dehydrogenase enzyme system of moulds. However, because of continued production of dehydrogenase, the inhibitory effect was overcome ultimately and sorbic acid was metabolized by moulds. The inhibitory effect is thus a function of initial mould contaminations and of the concentration of sorbic acid.
However, Bell et al. (1959) observed that the inhibitory action of sorbic acid on the growth of Y&M was closely related to pH which indirectly determined the quantities of undissociated active form of sorbic acid. These workers concluded that at pH 7.0, less than 1% of the sorbic acid was in undissociated form and many microorganisms accordingly grew well. At pH 4.4, more than 70% of the sorbic acid remained in undissociated form and was inhibitory to the growth of many microorganisms. Similar conclusion was also drawn by Samson et al. (1955) who reported that decrease in pH was also associated with increase in the inhibitory activity.

2.4.2.3 Sorbic acid in cottage cheese

Bonner and Harmon (1956) observed that increasing concentration of sorbic acid from 0.05 to 0.025% and decreasing the pH from 5.2 to 4.8, decreased the viability of 20 organisms, associated with cheese spoilage. Bradley et al. (1962) demonstrated that potassium sorbate caused a definite lag in the growth of the following organisms: _B5. fragi_, _Geotrichum candidum_, _Fenicillium frequentons_, _Tetulopsis candida_ and _Rhodotorula mucilagenosla_. The lag period extended from 1 to 8 days depending upon the sorbate concentration (0.05 to 0.1%) and the sensitivity of the organisms. The extension of the shelf-life of creamed cottage cheese was approximately equivalent to the length of the lag period.

Gemindes (1959) and Chakraborty and Kristofferson
(1963) reported that both sorbic acid and sorbates retarded the growth of yeasts and mould and slime-forming organisms when added to cottage cheese at a concentration of .05 and 0.75%.

A 17% increase in keeping quality of cottage cheese was obtained on addition of 0.05% sorbic acid (Chakraborty and Kristofferson, 1963).
CHAPTER 3

SCOPE AND PLAN OF WORK
3. SCOPE AND PLAN OF WORK

With the aim of reducing the total time of manufacturing shrikhand, and increasing its keeping quality in a manner suitable for large scale production, the present investigation was conducted to study:

(a) the effect of utilizing different types of milk solids other than fresh milk on the quality of shrikhand;

(b) the feasibility of direct acidification technique on the quality of shrikhand; and

(c) the effect of heat treatment of sugar and use of preservatives for extending the keeping quality of shrikhand.

Within this scope of investigation, the study was performed in following five phases:

(i) Establishing a standardized shrikhand making technology using buffalo skimmilk and cream, to be used as control in subsequent phases of investigation.

(ii) Use of different acidulants such as lactones, hydrochloric, citric and lactic acids with buffalo skim milk under various curd setting conditions for shrikhand making.

(iii) Use of different sources of milk solids, such as
skimmilk powder, condensed skimmilk, and butter-
milk for preparing shrikhand.

(iv) Using heat treated sugar for extending the keeping
good quality of shrikhand.

(v) Adding antimycotic agent such as potassium sorbate
for prolonging the keeping quality of shrikhand.
4. MATERIALS AND METHODS

This section deals with the methods and materials used in the preparation and characterisation of shrikhand.

4.1 SELECTION OF RAW MATERIALS

4.1.1 SKIM MILK

Raw buffalo milk was taken from the herd maintained at the National Dairy Research Institute, Karnal. The milk preheated to 40-45°C in a plate heat exchanger (ALFA LAVAL), was clarified and separated in a TITAN separator (Denmark) having a capacity of 3,000 litres per hour. The skimmilk contained on an average 0.05% fat, 3.93% proteins, 10.14% total solids and 0.12 to 0.14% lactic acidity.

4.1.2 BUFFALO CREAM

Buffalo cream was obtained from the Experimental Dairy of the Institute. The average composition of the cream was 77 to 80% fat, 2.17 to 2.45% SNF and 17.83 to 20.55% moisture.

4.1.3 BUTTER MILK

The buffalo cream was obtained from the Experimental Dairy of the Institute. The standardized buffalo cream (40% fat) was pasteurized at 70°C for 30 min, cooled to 10°C and stored for 12 hrs before churning. The cream was then churned in a metal butter churn (Silkeborg top verge metal churn). The butter milk thus obtained was collected in a
pre-sterilized aluminium milk can and heated to 55°C in a steam jacketed kettle. The fat in the buttermilk was removed by using TITAN cream separator. The buttermilk used for the experiment contained on an average 9.1 to 9.2% total solids, 3.45 to 3.47% proteins, 0.1 to 0.2% fat and 0.12 to 0.13% lactic acidity.

4.1.4 MAIN CONDENSED MILK

The buffalo skim milk was preheated to 80°C without holding in a Silkeborg plate heat exchanger (capacity 500 litres/hr) using steam as a heating medium. The preheated buffalo skim milk was then condensed to 40-43% total solids in a vacuum pan (Murray Deodorizers Ltd., New Zealand) at 61-63.5 cms vacuum. The condensed milk was drawn in sterilized milk cans and used for subsequent experiments.

4.1.5 SKIM MILK POWDER

The condensed skim milk as obtained in 4.1.4 was spray dried in an Anhydro (Denmark) spray dryer (35 kg water evaporation per hour capacity), keeping the outlet and inlet air temperatures at 95 ± 2°C and 190 ± 2°C, respectively. The atomizer speed was maintained at 20,000 ± 1,000 rpm.

4.1.6 STARTER CULTURE

Pure lactic starter culture of LF-40, NDRI1, NDRI2, YH, S+L, B, Mixed and Acidophilus were obtained from Dairy Bacteriology Division of the National Dairy Research Institute, Karnal.
4.1.7 SUGAR

Food grade sugar was obtained from the Experimental Dairy of the National Dairy Research Institute, Karnal.

4.1.7.1 Heat treatment of sugar

The commercial grade sugar was subjected to heat treatment to reduce the microbial load. For this, sugar was spread in an aluminium tray (thickness of layer, 4 to 4.5 cms) and heated in a hot air oven at different time-temperature combinations as given below:

120°C - 10, 20, 30, 40 and 50 min
140°C - 10, 15, 20, 25 and 30 min
160°C - 5, 10, 15, 20 and 25 min
180°C - 5, 7\text{\frac{1}{2}}, 10, 12\text{\frac{1}{2}} and 15 min
200°C - 2\text{\frac{1}{2}}, 5, 7\text{\frac{1}{2}} and 10 min

4.1.8 SALT

The commercial grade fine grain salt obtained from M/s. Tata Chemicals was used.

4.1.9 GLUCONIC-DELTA-LACTONE (GDL)

The anhydrous GDL (D-gluconic acid lactone) having molecular weight 178.10 was obtained from Sigma Chemical Company, USA.

4.1.10 ACIDS

The I.I/B.P grade hydrochloric lactic and citric acids were obtained from Sarabhai M. Chemicals, Baroda.
4.1.11 CHEMICALS

Analytical grade chemicals were used for chemical analysis.

4.1.12 DIACETYL

Diacetyl having molecular weight 86 was obtained from Sigma Chemical Company, USA.

4.1.13 PACKAGING MATERIALS

Polyethylene bags (300 gauge of 500 g capacity) and plastic cups (60 g capacity) were used.

4.1.14 MEDIA

The dehydrated media obtained from Hindustan Dehydrated Media, Bombay was used for microbiological analysis.

4.2 PREPARATION OF DAHI/CHAKKA/SHRIKHAND

4.2.1 DAHI

Dahi was prepared separately from buffalo skim milk, butter milk, reconstituted skim milk, concentrated skim milk and dilute condensed skim milk. These were heated to 85°C for 30 min. The milk was then cooled to 42 ± 2°C and YH culture (mixture of \textit{s. thermophilus} and \textit{L. bulgaricus}) was added at the rate of 2.0% and incubated for 3½ to 4 hrs at the same temperature so as to get a pH of 4.3 to 4.4 with the corresponding 0.85 to 0.9% lactic acidity of dahi. In another set of experiment, 1.0% LF-40 culture was added and the samples were incubated at 30 ± 1°C for 12 to 16 hrs to get a pH of 4.4 to 4.5 with the corresponding acidity.
of 0.85 to 0.9% lactic acid.

4.2.2 DIRECT ACIDIFIED CURD

The buffalo skim milk was heated to 85°C for 20 min and cooled to 5 ± 0.5°C. The pH of this milk was then adjusted to 5.2 by adding required quantity of chilled (5°C) lactic acid (1:9 dilution). This finally resulted a pH of 4.8 at 35°C of the acidified milk when the curd was formed.

4.2.3 CHAKKA

4.2.3.1 Chakka from dahi

The dahi as obtained in 4.2.1 was centrifuged in a Remi basket centrifuge (T 300) capacity 500 g/batch to separate out the whey. The resultant chakka contained 22.80-23.0% total solids, 13.60-13.95% total protein, 3.18-3.21% reducing sugar and 2.12-2.8% TVFA. The lactic acidity and pH of the chakka varied from 2 to 2.1% and 4.2 to 4.3% respectively.

4.2.3.2 Chakka from direct acidified curd

The whey from direct acidified curd was separated using T-300 Remi basket centrifuge. The chakka thus obtained was used for shrikhand making.

4.2.4 SHRIKHAND

4.2.4.1 Conventional shrikhand

The calculated quantities of chakka sugar and cream were mixed in a planetary mixer (Metrex, New Delhi;
capacity 2.0 kg/batch) at 30 to 35 rpm for half an hour to
get 41.0% sucrose and 6.0% fat in the finished product.
The finished product was then packaged in plastic cups.

4.2.4.2 Shrikhand from direct acidified curd

The ground sugar and ripened cream (with LF-40) test-
ing 80% fat were mixed in the direct acidified chakka as
in 4.2.4.1.

4.3 STORAGE STUDIES

The shrikhand made from standardized procedure was
studied for its keeping quality. The product with and
without potassium sorbate (0.01, 0.025, 0.05 and 0.1%) and
heat treated sugar was stored in plastic cups (60 ml) with
lid at 10 and 30°C. The samples were analysed for their
acceptability, microbiological quality and chemical chara-
cacteristics at definite intervals and compared with the
control.

4.4 ANALYTICAL METHODS

4.4.1 ANALYSIS OF RAW MATERIALS

The methods used for analysis of skim milk, butter
milk, reconstituted skim milk, diluted condensed milk,
plain condensed skim milk, whey and curd are given below:

4.4.1.1 Organoleptic test/acidity

As per IS:1479 (Part I) 1960.

4.4.1.2 Fat

As per IS:1224 (Part I) 1977.
4.4.1.3 **Total solids** (by gravimetric methods)

As per IS:1479 (Part II) 1961.

4.4.1.4 **Proteins** (T.P.)

As per IS:1479 (Part II) 1961.

4.4.1.5 **Acidity**

As per IS:1479 (Part I) 1960.

4.4.2 **CURD TENSION OF DAHI/CURD**

4.4.2.1 **Curd tension of dahi**

The curd tension of dahi was measured by the method of Chandrashekhar et al. (1957) with the following modifications:

50 ml portion of milk in a 100 ml beaker (7 x 4.5 cm) inoculated with starter culture was incubated separately for a specified period of time at different temperatures. The curd tension was then measured and expressed in g.

4.4.2.2 **Curd tension of direct acidified curd**

Curd tension of direct acidified curd was measured by the method given in section 4.4.2.1. The curd was set using different acidulant at various temperatures and the curd tension measured after 35-45 min of incubation.

4.4.3 **STARTER**

Fresh starter having clean flavour and smooth body and texture was used. The acidity of starter was 0.85 to 0.9% lactic acidity.
4.4.3.1 Acidity

As per the method given in the Laboratory Manual "Methods of Analysis of Milk and Milk Products" of the Milk Industry Foundation (1959).

4.4.4 CHEMICAL ANALYSIS OF CHAKKA

4.4.4.1 Acidity

Titratable acidity of chakka was determined as follows:

Ten grammes of sample was diluted with 20 ml of lukewarm distilled water. After thorough mixing, it was titrated against 0.1N NaOH, using phenolphthalein (1% in 75% alcohol) as an indicator. The results were expressed as percentage lactic acid.

4.4.4.2 pH

The pH of chakka was determined with a combined electrode (CK-61) Elico pH meter, Model LI-15 by making a paste of 10 g of sample with about 10 ml of glass distilled water as per IS:1479 (Part II) 1961.

4.4.4.3 Protein fraction

The nitrogen fractions, namely total proteins and soluble nitrogen were determined as per the method of El-Sokkary et al. (1952) and then following semi-micro kjeldahl method of Menefee and Overman (1940) for digestion and distillation. In case of soluble nitrogen, the method was modified only in terms of sample size. Twenty grammes of the sample were taken and 5 ml of the filtrate was
digested and distilled.

4.4.4.4 Reducing sugar

As per IS:2802 (1964).

4.4.4.5 Total volatile fatty acids (TVFA)

The volatile fatty acids were determined by the method of Hempenions and Liska (1968) with slight modifications. Fifty grammes of the sample was weighed and transferred to a 800 ml kjeldahl flask and 3.0 ml of 1N H₂SO₄ was added. The contents were then steam distilled and about 100 ml of the distillate was collected and titrated using 0.01N NaOH and the total volatile fatty acids content was expressed as ml of 0.01N NaOH per 50 g of the sample.

4.4.4.6 Total solids/moisture

Moisture in chakka was determined gravimetrically using Mojonnier test assembly as per the method of Milk Industry Foundation (1959). A representative sample of 2 to 3 g was weighed into total solids dish with acid washed sand and glass rod and dried to constant weight in the hot air oven at 110 ± 5°C. The percentage moisture was calculated as follows:

\[ \% \text{ Moisture} = \frac{\text{Loss of weight}}{\text{Weight of sample}} \times 100 \]

4.4.5 Chemical analysis of shrirhsand

4.4.5.1 Sensory evaluation

The fresh as well as stored samples of shrirhans were subjected to sensory evaluation by a panel of trained
judges. The stored samples were evaluated at the interval of 10 days (for the samples stored at 10 ± 2°C) and 2 days (for samples stored at 30°C). The score cards used for judging are given in Figs.1(a) and 1(b).

4.4.5.2 Acidity

The acidity was determined as per 4.4.4.1.

4.4.5.3 pH

The pH was determined as per 4.4.4.2.

4.4.5.4 Total proteins

The total proteins were determined as per 4.4.4.3.

4.4.5.5 Soluble nitrogen

The soluble nitrogen was determined as per 4.4.4.3.

4.4.5.6 Reducing sugar

The reducing sugar was determined as 4.4.4.4.

4.4.5.7 TVFA

The TVFA was determined as per 4.4.4.5.

4.4.5.8 Moisture and total solids

The moisture and total solids were determined as per 4.4.4.6.

4.4.5.9 Fat

The fat content was determined by modified Gerber test for milk as suggested by Puntambekar (1968). Shrikhand was diluted 1:2.5 w/v and 80% sulphuric acid (sp. gravity, 1.730 at 20°C) then followed IS:1224 (Part I) 1977.
EVALUATION CARD FOR HEDONIC RATING TEST

Name ____________________ Date ____________________

Product ____________________ Time ____________________

Instruction: (The given sample of shrikhand is prepared by using 'Y1' culture). You are requested to assess the product in terms of general acceptability with special reference to flavour, colour, appearance and body texture on the basis of 9 point hedonic scale given below:

- Liked extremely 9
- Liked very much 8
- Liked moderately 7
- Liked slightly 6
- Neither liked nor disliked 5
- Disliked slightly 4
- Disliked moderately 3
- Disliked very much 2
- Disliked extremely 1

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Sample No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Colour</td>
<td>1  2  3  4  5</td>
</tr>
<tr>
<td>Appearance</td>
<td></td>
</tr>
<tr>
<td>Body &amp; texture</td>
<td></td>
</tr>
<tr>
<td>Flavour</td>
<td></td>
</tr>
<tr>
<td>Overall acceptability</td>
<td></td>
</tr>
</tbody>
</table>

Remarks

Signature

Fig. 1(a)
**SCORE CARD FOR SHRINKHAND**

Date ____________  Time ____________  Name ____________

**Scoring:** Assign score for each sample for different characteristics after deducting for defects.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Defect</th>
<th>Degree of defect</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Slight</td>
<td>Definite</td>
</tr>
<tr>
<td><strong>Colour</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dull</td>
<td>0.5-1.0</td>
<td>1.5-2.5</td>
</tr>
<tr>
<td>Lack of uniformity</td>
<td>0.25-0.5</td>
<td>1.0-1.5</td>
</tr>
<tr>
<td></td>
<td>Dull</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Silk-finished</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Wet look</td>
<td>1</td>
</tr>
<tr>
<td><strong>Appearance</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leaky</td>
<td>2-3</td>
<td>4-5</td>
</tr>
<tr>
<td>Grainy</td>
<td>3-4</td>
<td>5-7</td>
</tr>
<tr>
<td><strong>Body and texture</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Weak</td>
<td>2-3</td>
<td>4-5</td>
</tr>
<tr>
<td>Hard</td>
<td>2-3</td>
<td>5-7</td>
</tr>
<tr>
<td>Acidic/sour</td>
<td>4-6</td>
<td>7-10</td>
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<tr>
<td>Cooked/heated</td>
<td>3-6</td>
<td>7-11</td>
</tr>
<tr>
<td><strong>Flavour</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Too strong</td>
<td>4-6</td>
<td>7-10</td>
</tr>
<tr>
<td>Flat</td>
<td>3-6</td>
<td>7-11</td>
</tr>
<tr>
<td>Yeasty</td>
<td>4-7</td>
<td>8-12</td>
</tr>
<tr>
<td>Mouldy</td>
<td>5-7</td>
<td>8-11</td>
</tr>
<tr>
<td>Rancid</td>
<td>3-4</td>
<td>5-7</td>
</tr>
</tbody>
</table>

**SCORE CARD**

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Ideal score</th>
<th>Sample score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Colour</td>
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<td>1  2  3  4  5  6  7  8</td>
</tr>
<tr>
<td>Appearance</td>
<td>10</td>
<td></td>
</tr>
<tr>
<td>Body &amp; texture</td>
<td>30</td>
<td></td>
</tr>
<tr>
<td>Flavour</td>
<td>55</td>
<td></td>
</tr>
</tbody>
</table>

**Remarks:**

Fig.1(b)
4.4.5.10 ** Sucrose  

The sucrose content was determined as per IS:2802 (1964).

4.4.6 MICROBIOLOGICAL ANALYSIS OF SHRIKHAND  

The product was analysed for TVC, YM, proteolytic coliforms and lactic count as per the method given in the Laboratory Manual "Methods of Analysis of Milk and Milk Products" of Milk Industry Foundation (1959).

4.4.6.1 Dilution blanks  

The dilution blank consisted of 2% sodium chloride solution in 99 ml and 9 ml portion in screw capped dilution bottle and screw capped culture tubes. Those were autoclaved at 121°C for 20 min. The dilution blanks were warmed to 45°C before use for preparation of samples.

4.4.6.2 Sampling of shrikhand  

Random samples were taken from the storage lot and 11 g of shrikhand was weighed in a sterilized beaker. This was mixed thoroughly with dilution blank (99 ml). Further dilutions were made with the use of 9 ml dilution blanks.

4.4.6.3 Total viable counts (TVC)  

For total viable counts (TVC), lactose yeast phosphate agar media having the following composition was used. 
Peptone 0.5%, yeast extract 0.5%, beef extract 1.0%, 
Na₂HPO₄ 0.3%. The media was sterilized by autoclaving at 120°C for 20 min and cooled to 40°C. Serial dilutions were
made from 1:10 dilutions of the suspension of the sample and the petridishes were poured in duplicates. These petri-
dishes were then incubated at $37 \pm 1^\circ C$ for 48 to 72 hrs and the colonies for TVC were counted.

4.4.6.4 Coliform count

Violet red bile agar media was used for coliform count. Before using this media it was autoclaved at $120^\circ C$
for 20 min and cooled to $35^\circ C$ and then the sample was taken into a petridish and violet red bile agar was poured in the petridish. After solidification, second layer was made using the same agar. The petridishes were incubated at $37 \pm 1^\circ C$
for 24 to 48 hrs.

4.4.6.5 Proteolytic counts

For proteolytic counts, tryptone glucose extract agar media was used. This media was autoclaved at $120^\circ C$ for 20
min and cooled to $40^\circ C$. Five percent sterilized skim milk was mixed with the media and then it was poured into the petri-
dishes. These petridishes were incubated at $37^\circ C$ for 4 days. The colonies having a digested zone surrounding each colony, represented proteolytic one.

4.4.6.6 Lactic count

Lactic count was taken by using tomato juice agar media, autoclaved at $121^\circ C$ for 20 min and cooled to $45^\circ C$, and it was poured into sample containing petridishes. The petri-
dishes were incubated at $37^\circ C$ for three days and then counted for the colonies.
4.4.6.7 Yeast and mould count (YM)

Potato dextrose agar media was used to determine the yeast and mould count of the samples. The media was sterilized by autoclaving at 120°C for 20 min. The pH of the media was adjusted to 3.5 pH before plating by using sterile 10% tartaric acid. The media was poured into the sample containing petridishes and then incubated at 22°C for 2 to 3 days and the colonies of yeasts and moulds were counted.

4.4.7 RHEOLOGY OF SHRIKHAND

4.4.7.1 Curd tension

An 'I' shape curd tension knife (Chandrashekhara et al., 1959) was placed in a 100 ml beaker and filled with shrikhand by gentle trapping up to 50 ml. The beakers were then held overnight at 10 ± 2°C and then the curd tension (the force required to pull the knife through shrikhand) of the shrikhand at different temperatures was measured in gramme as per the Chandrashekhara et al. (1959) method.

4.4.7.2 Coming up time

The time required to pull the knife through shrikhand was determined and expressed as coming up time in seconds. For this, the time was recorded from the moment the curd tension knife started moving upwards till it cut the surface of the shrikhand.

4.4.7.3 Penetrometer reading

Firmness of the product was measured with a precision
Penetrometer (The Central Ignition Company, London). The
shrikhand samples were filled in 60 g plastic cups and kept
overnight at 10 ± 2°C. The tempered sample was then placed
on the platform of the penetrometer in such a way that the
penetration point was at least one inch away from the edges.

The core of the penetrometer was lowered over the sample
after removing the lid of the cup so that the tip just
touched the sample. The core was released and allowed to
descend exactly for 5 seconds and reading in 0.1 mm was
taken.
CHAPTER 5

RESULTS AND DISCUSSION
5. RESULTS AND DISCUSSION

Studies on the process alteration in shrikhand making were carried out in five distinct stages. A standard method of manufacturing shrikhand with the lactic fermentation of buffalo skimmilk was established as the first step in order to provide a basis of comparing all other subsequent stages of shrikhand making against these control standards. In the next step, the utilization of different acidulant for the direct acidification of buffalo skimmilk was studied for shrikhand making. The feasibility of using milk solids from sources other than fresh skimmilk was studied in the third step. The fourth and fifth steps of this study were concerned with the effect of heat treating sugar and using potassium sorbate on the shelf-life of shrikhand. In this section, the results of these studies are presented in that order.

5.1 STANDARDISATION OF SHRIKHAND MANUFACTURING PROCESS BY LACTIC FERMENTATION

Although traditional method of shrikhand making begins with the lactic fermentation of whole milk, the benefits of using a skimmilk curd in terms of saving fat losses in whey and faster syneresis, have been reported (Upadhayay and Dave, 1975; Aneja et al., 1977). The yield and quality of shrikhand are further influenced by the selection of starter culture, heat treatment of the milk, time and temperature
of incubation, final acidity of curd, and the compositional aspects of shrikhand in terms of moisture, fat and sugar levels. Effect of these factors on the quality of shrikhand is presented here.

5.1.1 SELECTION OF STARTER CULTURE

In shrikhand making technology, preparation of dahi by conventional starter culture takes about 12-15 hrs, a time period too long to promote the development of a continuous shrikhand making process. However, it is known that the incubation time is largely governed by the incubation temperature, so the cultures capable of producing lactic acid at a relatively high temperature (yoghurt culture), can coagulate the milk in shorter times (Wilkowske, 1954; Accolas et al., 1977). With a view to select a culture suitable for fast acid production, several lactic acid cultures, viz. LF-40, NDRI1, NDRI2, DRC1, YH, L.acidophilus, 'B', mixed and S+L were used for curd forming at temperatures optimum for these cultures. For these studies, 1% inoculum was used in buffalo skimmilk (10% TS and heated to 85°C for 10 min). The hourly changes in pH and T.A. of various cultures are presented in Appendix I. Flavour, curd strength, the rate of acid development and formation of total volatile fatty acids are summarized in Table 1.

It can be seen from Table 1 that as expected, the time required to reach a pH level of 4.6 is related to the incubation temperature. The longest time of 11 hrs was taken by LF-40 and DRC1 at 30°C, while shortest time of 4 hrs
Table 1. Curd forming characteristics of selected lactic cultures

<table>
<thead>
<tr>
<th>Curd forming characteristics</th>
<th>LF-40</th>
<th>NDRI$_1$</th>
<th>NDRI$_2$</th>
<th>DRC$_1$</th>
<th>YH</th>
<th>Acido-philus</th>
<th>'B'</th>
<th>Mixed</th>
<th>S + L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Incubation temp (°C)</td>
<td>30</td>
<td>37</td>
<td>37</td>
<td>30</td>
<td>42</td>
<td>37</td>
<td>37</td>
<td>37</td>
<td>37</td>
</tr>
<tr>
<td>Time required to reach pH 4.6 (hrs)</td>
<td>11</td>
<td>9</td>
<td>10</td>
<td>11</td>
<td>4.0</td>
<td>9</td>
<td>9</td>
<td>9</td>
<td>6</td>
</tr>
<tr>
<td>Flavour$^2$</td>
<td>+++</td>
<td>++</td>
<td>++</td>
<td>+++</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td>TVFA (0.01 ml NaOH/50 g of curd)</td>
<td>10.10</td>
<td>6.00</td>
<td>5.70</td>
<td>10.80</td>
<td>2.40</td>
<td>4.20</td>
<td>5.90</td>
<td>7.00</td>
<td>4.50</td>
</tr>
<tr>
<td>Curd tension (g)</td>
<td>86.90</td>
<td>73.60</td>
<td>78.30</td>
<td>66.30</td>
<td>98.70</td>
<td>69.80</td>
<td>71.80</td>
<td>81.30</td>
<td>83.40</td>
</tr>
</tbody>
</table>

1 Buffalo skim milk of 10% TS and heated to 85°C for 10 min were inoculated with 1% active cultures

2 +++ pronounced culture flavour; ++ moderate culture flavour; + slight culture flavour

* Average of three trials
was required by YH culture working at 42°C. However, the culture varies markedly among them, even when the same incubation temperatures were used.

At 37°C temperature, the pH reached by S+L culture in 6 hrs was attained by the other cultures in 9 to 10 hrs. Among all the cultures studied, Y1 exhibited the fastest rate of acid development.

The flavour quality of the lactic cultures, LF-40 and DRC₁, the slowest acid producers, was judged to be best in term of the development of a desirable mild aroma. In this regard YH, acidophilus, 'B' and S+L exhibited only slight aromatic flavour. The level of TVFA exhibited a general relationship with the flavour traits, being highest for LF-40 and DRC₁, and lowest for the YH. Although fast acid producing culture are known to be associated with the development of bitter taste, no such defect was detected in curd prepared from YH culture. Curd tension properties of this culture did not show any relationship with the other parameters such as the rate of acid development, flavour or TVFA. YH exhibited the highest curd tension value of 98.70 g, while DRC₁ had a lowest value of 66.30 g.

In general Y1 culture could be considered most desirable as this would permit most rapid development of acidity without adversely affecting the curd strength and the flavour aspects. For these reasons Y1 culture was selected for shrikhand manufacture and other subsequent studies.
5.1.2 HEAT TREATMENT OF MILK

A Yh culture is basically a yoghurt culture, containing a mixture of \textit{S. thermophilus} and \textit{L. bulgaricus}. Heat treatment of milk prior to inoculation plays a significant role in acid development, particularly with lactobacillus cultures. This might be because of possible destruction of heat labile inhibitory compounds and also the heat induced formation of growth stimulants such as peptides, amino acids and formic acids which have been reported to enhance acid production (Miller \textit{et al.}, 1964). The effect of four different heat treatments of skimmilk, viz. 80°C/1 min, 85°C/1 min, 80°C/30 min and 85°C/30 min on the pH and acid development is presented in Fig. 2(a). As the heat treatment of milk increased, the rate of acid development as well as the final acidity also increased. The effect of increased holding period in the range studied appeared to cause more pronounced differences in the rate than that associated with the differences in temperature. The effect of heat treatment on reducing the pH of various curd systems were similar to the effect on acidity development as can be seen from Fig. 2(b). As the incubation time increased, the differences with level of pH were more pronounced among the various samples. Very similar to the case of acidity development, differences were more due to time of treatment and less due to temperature differences.

In addition to an enhanced production of acidity,
FIG. 2. EFFECT OF HEAT TREATMENT OF MILK ON THE RATE OF ACID DEVELOPMENT AND CHANGES IN pH USING YH CULTURE
high heat treatments resulting in denaturation of whey proteins, and their subsequent co-precipitation during acid coagulation had been observed to increase the yield of the chakka (Aneja et al., 1977). Thus, the highest heat treatment, viz. 85°C for 30 min giving a pH of 4.0-4.20 and acidity 0.9-0.95% LA was adopted for further studies.

5.1.3 EFFECT OF INCUBATION TEMPERATURE AND AMOUNT OF STARTER CULTURE ON CURD FIRMNESS

Firmness of the curd used for chakka making is an important property mainly from the point of view of solids losses in whey. The firmer the curd, the less will be solids losses. The effect of adding Yd culture to buffalo skim milk at the rate of 1.0, 1.5 or 2.0% and incubating the milk at 37°C and 42°C for 4 hrs can be seen from Table 2. The curd tension was somewhat higher with the higher incubation temperature. For all the three rates of inoculation, with the increasing inoculation rate, the curd tension also increased. However, 1.5% level of inoculation gave a substantially firmer curd as compared to that obtained at 1.0%. There was only a slight difference between 1.5 and 2.0% levels. Accordingly, the inoculum levels of 1.5 to 2.0% were used in shrikhand making trials subsequently.

5.1.4 EFFECT OF CURD ACIDITY ON SHRIKHAND FLAVOUR

The flavour quality of shrikhand is greatly influenced by the level of the developed acidity at which curd is converted to chakka, and chakka in its turn to
Table 2. Effect of incubation temperature and amount of inoculum on curd firmness*

<table>
<thead>
<tr>
<th>Inoculum level %</th>
<th>Curd tension (g) at 37°C</th>
<th>42°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>83.56 (81.60 - 83.60)</td>
<td>92.80 (90.50-94.60)</td>
</tr>
<tr>
<td>1.5</td>
<td>90.82 (89.90 - 91.50)</td>
<td>98.30 (94.20-103.50)</td>
</tr>
<tr>
<td>2</td>
<td>91.70 (90.60 - 93.20)</td>
<td>99.35 (95.20-101.70)</td>
</tr>
</tbody>
</table>

* Average of three trials

Figures in parentheses indicate range
shrikhand. Three levels of acidity were, therefore, developed in the curds, viz. 0.81, 0.90 and 0.98% LA for their conversion to maska or chakka. The chakka samples were then blended with sugar and cream to a final compositional level of approximately 40% sugar and 6% fat in shrikhand. The data on the organoleptic evaluation of these shrikhand samples are presented in Table 3. The titratable acidity of chakka exhibited a corresponding increase as the curd acidity increased.

On the basis of flavour score, a curd acidity of 0.81% LA was considered too low as the shrikhand was criticized as mildly acid and too sweet. A titratable acidity of curd in the range of 0.9 to 0.98% LA appeared to result in most desirable shrikhand flavour, as was also reported by Aneja et al. (1977).

5.1.5 FACTORS AFFECTING THE CONSISTENCY AND SENSORY PROPERTIES OF SHRIKHAND

Shrikhand had a typical semisolid consistency showing a characteristic firmness and pliability contributing to its suitability for consumption with "puree" as traditionally done. The consistency is influenced to a great extent by the moisture, fat and sugar levels in shrikhand. These last two factors also have an influence on the flavour of the product.

Effect of varying the levels of moisture, fat and sugar on the rheological properties of the resulting shrikhand was studied in terms of consistency measurements
Table 3. Effect of curd acidity on the sensory quality of shrikhand*

<table>
<thead>
<tr>
<th>Acidity of curd</th>
<th>Acidity of chakka</th>
<th>Acidity of shrikhand</th>
<th>9 point hedonic score</th>
<th>Criticism</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.81</td>
<td>1.94</td>
<td>0.99</td>
<td>6.93</td>
<td>too sweet lacking mild acidic flavour</td>
</tr>
<tr>
<td>0.90</td>
<td>2.11</td>
<td>1.08</td>
<td>7.34</td>
<td>mild acidic flavour, optimum sugar level</td>
</tr>
<tr>
<td>0.93</td>
<td>2.70</td>
<td>1.390</td>
<td>7.36</td>
<td>slight acidic flavour</td>
</tr>
</tbody>
</table>

* Average of three trials
using curd tension apparatus as well as penetrometer. In this study attempt was made to vary one component at a time keeping the relationship of other component affected to minimum. The Appendix II lists the various ingredients, their composition and their properties used for preparing various shrikhand blend at three levels of moisture, fat and sugar.

A curd tension apparatus was used to measure the curd tension value and time required (sec) for the knife to come up through the given depth of the sample at 15, 30 and 40°C. It can be seen from Fig. 3 and Appendix III that within the compositional range studied, there was a general reduction in the so called curd tension values of shrikhand samples along with an increase in the level of moisture, fat and sugar. Effect of increasing temperature from 15 to 40°C on the curd tension value, was also in the similar direction and more pronounced than the effect due to compositional changes.

From Fig. 4 and Appendix IV, it can be seen that a similar reduction in the coming up time values due to increasing levels of moisture, fat, sugar and temperature was exhibited.

Penetration readings taken with the same series of samples at 5, 10 and 15°C, as shown in Fig. 5 and Appendix V gave increasing penetration values along with an increasing level of moisture, fat and sugar levels.
EFFECT OF MOISTURE, FAT, SUGAR AND TEMPERATURE ON THE CURD TENSION VALUE OF SHRIKHAND
FIG. 4. EFFECT OF MOISTURE, FAT, SUGAR AND TEMPERATURE ON THE COMING UP TIME OF SHRINHANH
FIG. 5. EFFECT OF MOISTURE, FAT, SUGAR AND TEMPERATURE ON THE PENETRATION VALUE OF SHRIKHAND
However, the compositional differences in fat had the least effect on the penetrometer values in comparison to the effects due to the variation in moisture and sugar levels. In general, most pronounced effect was due to the temperature differences.

Studies with curd tension apparatus and penetrometer confirm the major role of the protein systems towards the consistency building properties of shrikhand. The effect of increasing the nonprotein constituents of the shrikhand solids such as fat and sugar was similar to the reduction of the total solid levels of shrikhand due to the increasing levels of moisture, in terms of the weakening of shrikhand consistency.

On the basis of organoleptic evaluation (vide Appendix VI), most desirable shrikhand in this study was obtained from a system containing 6% fat, 41% sugar and 40% moisture. It is, therefore, decided to keep the standard composition for subsequent study.

5.1.6 EFFECT OF DIACETYL ADDITION ON THE FLAVOUR OF SHRIKHAND OBTAINED WITH THE USE OF YH CULTURE

Earlier it was noticed that the curd obtained with the YH culture had a less desirable aromatic property than that obtained with cultures known to produce diacetyl (vide Table 1). In order to improve the flavour of the curd from YH culture, addition of diacetyl at 10, 15, 20 and 25 ppm levels in shrikhand was investigated. The experimental product was compared with the reference product
obtained by using LP-40, a diacetyl producing culture. From Table 4 it can be seen that diacetyl added at the rate of 10 ppm improved the flavour score from 6.66 to 7.26 (on nine point hedonic scale), which was fairly comparable with the reference sample score of 7.42. Further increase in the diacetyl level decreased the flavour score, the product flavour tended to be harsh and unnatural.

It is not worthy, however, that the favourable influence of the diacetyl addition (10 ppm), shrikhand flavour was no longer perceptible when cardamom was used for flavouring the product (as usually is the practice in traditional shrikhand making), apparently because of the masking effect of cardamom. Therefore, flavour enrichment of the Y.I culture product with diacetyl was thought unnecessary. However, a comparison of cardamom containing samples, prepared from LP-40 and Y.I cultures, without added diacetyl, revealed little difference in the reference score, indicating an effective masking action of cardamom flavour which predominates the system. Therefore, flavour enrichment with diacetyl was thought unnecessary.

5.1.7 STANDARDIZED METHOD OF SHRIKHAND MAKING

On the basis of studies conducted so far, a standard method was established for preparing a control shrikhand sample (Fig.6) for comparison in various subsequent studies. The method involves use of buffalo skimmilk (10% TS), subjected to heat treatment of 85°C/30 min, followed by
Table 4. Effect of diacetyl addition on the flavour of shrikhand obtained with YH culture***

<table>
<thead>
<tr>
<th>Reference sample*</th>
<th>Diacetyl level in experimental sample**</th>
<th>Experimental sample with cardamom @ 1 g/1 kg of shrikhand</th>
<th>Reference sample + cardamom @ 1 g/1 kg of shrikhand</th>
</tr>
</thead>
<tbody>
<tr>
<td>7.42 (7.39-7.47)</td>
<td>6.66 7.26 7.15 7.11 6.95 7.38 7.54</td>
<td>6.91-7.02 7.02 7.32-7.46 7.49-7.62</td>
<td>7.02 7.06</td>
</tr>
<tr>
<td>pleasant aromatic</td>
<td>flat pleasant slightly harsh definite-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>flavour</td>
<td>flavour</td>
<td>ly harsh</td>
<td>un-</td>
</tr>
<tr>
<td></td>
<td></td>
<td>flavour</td>
<td>natural</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>flavour</td>
</tr>
</tbody>
</table>

* Reference sample was prepared with LF-40 culture (1% inoculum at 30°C)
** Experimental sample was prepared with YH culture (2% inoculum at 42°C)
*** Average of three trials

Figures in parentheses indicate range
FIG. 6. SHRIKHAND MANUFACTURE

BUFFALO MILK
Pasteurized (85 °C for 30 min)

Cream separator

10% TS

Skim milk

Curd setting vat
temp. 42 °C, 4 hrs.

0.9 to 1.0% L.A.

Whey drainage

Chakka
22-23% T.S., 2.11 to 2.70% L.A.

Cream 80% fat

Sugar @ 80% of chakka

Shrikhand

Moisture 40%
Fat 6%
Protein 6.6%
Sugar 41%

FIG. 6. SHRIKHAND MANUFACTURE
1.5 to 2.0% inoculation of an active YH culture at 42 ± 2°C for obtaining a curd (0.9% LA) at the end of 4 hrs. The chakka obtained from this curd was then blended with required quantity of 80% fat pasteurized (85°C/10 min), plastic cream and sugar to represent a final composition of 40% moisture, 6% fat and 41% sugar. Thus, in the standard process itself it was possible to reduce the curd formation time from 8 hrs to 4 hrs by selecting an appropriate culture and a higher setting temperature.

5.2 SHRikhAND MANUFACTURING BY DIRECT ACIDIFICATION

Although it was possible to reduce the curd setting time considerably by an appropriate selection of starter culture and incubation temperature, application of direct acidification technique for curd formation was attempted to gain additional advantages in the form of eliminating biological variation due to the use of starter culture on a day to day basis and easier adaptation to continuous coagulation system similar to those reported by Ernstrom (1963), Satterness et al. (1978) and Fox (1978).

For this purpose the feasibility of using slowly hydrolysing glucono-delta-lactone (GDL) as well as inorganic and organic food grade acids, such as hydrochloric, lactic and citric were studied.

5.2.1 USE OF GDL FOR SHRikhAND MAKING

GDL has been used as an acidulant in milk (Mabbitt et al., 1955; Deane et al., 1960; O’Keeffe et al., 1975; and
Satterness et al., 1978). Hydrolysis of the product results in a gradual release of gluconic acid which, by reducing the pH of milk, ultimately brings about its coagulation. In the first stage, the curd forming properties of GDL were determined to select a set of curd-forming conditions for preparing shrikhand in the second stage.

5.2.1.1 Curd formation by GDL

Rate of hydrolysis of GDL is influenced by its concentration in milk and the temperature of hydrolysis. Effect of seven different levels of GDL (0.5 to 3.0%) in buffalo skim milk, set at 20, 30 and 40°C, on the lowering of pH is presented (vide Appendix VII).

Rate of change in pH for selected concentration levels are presented in Fig. 7(a), (b) and (c). In general, hydrolysis rate, apparent on the basis of pH changes, was greatest during the first half hour. Thereafter, the rate slowed down considerably.

The extent of acid development, proportional to lowering of the pH value, increased along with the rising level of GDL in milk. Temperature was also responsible for a faster hydrolysis rate. At the coagulation temperature of 30°C, a 2% level of GDL resulted in distinct curd formation after about 2 hr of holding. At 2.5% concentration level and 40°C, curd formation was achieved within 90 min. It was therefore, decided to use GDL at 2% and 2.5% levels at 30 and 40°C in the shrikhand making studies.
FIG. 7. RATE OF CHANGES IN pH OF SKIMMILK AT DIFFERENT TEMPERATURE AND CONCENTRATION OF GDL
5.2.1.2 Shrikhand making trials

Shrikhand prepared from the curd derived by mixing GDL at 2.0 and 2.5% levels at 30 and 40°C (corresponding to a pH 4.8 and 4.6 after 150 and 90 min of hydrolysis time, respectively) was assessed for its texture characteristics. It was noticed that there was no appreciable difference in the product texture with regard to the pH of coagulation or temperature of coagulation. When compared with the control, all the samples were considered unsatisfactory on account of perceptible "graininess" defect.

Direct acidification with GDL was thus found unsatisfactory for shrikhand making under the present experimental conditions.

5.2.2 USE OF FOOD GRADE ACIDS

Using food grade acids for the coagulation of milk must be attempted in such a way that a desirable curd strength is obtained through the formation of a gel structure instead of a mean precipitate of proteins. The temperature of milk and the dilution of acids are two important parameters for attaining direct acidification of milk in a desirable manner (Mabbitt et al., 1955). It has already been established that cold acidification (at a temperature below 5°C) followed by a gradual warming (30°C or higher) is desirable for obtaining a smooth and firm coagulum for direct acidification. Acid must be diluted to a desirable level, as over concentration might result in spot
coagulation due to excessive heat generation and insufficient mixing. Overdilution, on the other hand, might create difficulty associated with handling a larger volume of liquid.

On the basis of preliminary trials with a sample of lactic acid diluted to 1:6 and 1:9, the latter was found to promote a formation of better curd. Accordingly, it was decided to undertake direct acidification study with different food grade acids diluted to the extent of 1:9 with tap water and mixed at a temperature of 4°C or below with milk chilled to similar temperature range. For these studies, required quantity of diluted acid used (Fig.8), added to cold milk, mixed for 2 to 3 min and the beaker containing 400 ml of this mixture was then placed in a water bath at 37°C. A thermometer placed in a beaker indicated that the temperature of 35°C was usually attained by the quiescent samples within a period of 25 to 30 min. Curd formation was judged by the expulsion of clean whey bids from freshly cut curd surface. Average coagulation time (min) plotted for different acids in Fig.9 indicated that, in general, faster coagulation was attained at low pH values. Coagulation times were similar for the curd formed with the organic acids, i.e., lactic acid and citric acid. Coagulation time with hydrochloric acid was lower than those required with other two acids.

5.2.2.1 Effect on curd tension

Curd tension data recorded for the three acid
FIG. 8. QUANTITIES OF DIFFERENT TYPES OF ACID REQUIRED TO ACIDIFY SKIMMILK TO VARIOUS pH LEVELS
FIG. 9. EFFECT OF TYPES OF ACID ON THE COAGULATION TIME (MIN) AT VARIOUS pH LEVELS
containing systems at different pH levels, graphically
plotted in Fig.10, indicated that the firmness of curd (in
terms of curd tension) increased with the decreases in pH
from 5.2 to 4.6 and pH 4.5, for all the three types of acids
studied. It thus appeared that coagulation near the iso-
electric point of casein was conducive to greater firmness
of the curd as compared to either side of the isoelectric
point. However, at the same pH, the curd tension was in
increasing order for lactic acid, citric acid and hydrochlo-
ric acid, particularly in the pH range of 4.6 to 4.9.

5.2.2.2 Effect on chakka texture

Various curd samples were drained off whey with the
aid of a laboratory basket centrifuge (1,000 to 1,2000 rpm)
for 30 to 40 min. The resultant chakka samples were subjec-
ted to sensory evaluation for textural property. Results are
presented in Table 5. It can be seen from the results that
at pH levels of 5.2 and 5.1 no chakka was obtained with the
use of any of the three acids. The presence of grainy tex-
ture defect in all the samples was obtained at pH 5 to 4.3.
However, this defect was less pronounced at pH 5.0.

Among the three acids, lactic acid resulted in curd
containing the least amount of the grainy texture defect and
the hydrochloric acid the most. On the basis of this curd
forming and chakka textural quality, lactic acid was chosen
as the coagulant for further experiments on direct acidifica-
tion.
FIG. 10. EFFECT OF THE TYPE OF ACIDS AND pH OF COAGULATION ON CURD TENSION AT VARIOUS pH LEVELS.
Table 5. Effect of the type of acids and pH of coagulation on texture of chakka

<table>
<thead>
<tr>
<th>pH</th>
<th>5.2</th>
<th>5.1</th>
<th>5.00</th>
<th>4.90</th>
<th>4.80</th>
<th>4.70</th>
<th>4.60</th>
<th>4.50</th>
<th>4.40</th>
<th>4.30</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lactic acid (1:9 dil)</td>
<td>Texture</td>
<td>-</td>
<td>-</td>
<td>x</td>
<td>x</td>
<td>xx</td>
<td>xx</td>
<td>xxx</td>
<td>xxx</td>
<td>xx</td>
</tr>
<tr>
<td>Citric acid (1:9 dil)</td>
<td>Texture</td>
<td>-</td>
<td>-</td>
<td>x</td>
<td>x</td>
<td>xx</td>
<td>xx</td>
<td>xxx</td>
<td>xxx</td>
<td>xx</td>
</tr>
<tr>
<td>HCI acid (1:9 dil)</td>
<td>Texture</td>
<td>-</td>
<td>-</td>
<td>xx</td>
<td>xx</td>
<td>xxx</td>
<td>xxx</td>
<td>xxx</td>
<td>xxx</td>
<td>xx</td>
</tr>
</tbody>
</table>

1 x slightly grainy; xx moderately grainy; xxx very grainy
5.2.2.2.1 Effect of curd setting temperature and pH

As the curd forming properties and subsequent behaviour of chakka can be influenced by the pH and temperature chosen for curd setting, it was decided to study the effects of these parameters on lactic acid curd with the ultimate aim for selecting setting conditions that would improve the quality of curd. Consequently, to study these, three water bath temperatures, viz. 30, 35 and 40°C were selected along with the coagulation pH of 4.8, 4.9 and 5.0 (on the basis of previous observations). Data obtained for this curd in terms of coagulation time, curd tension, solids losses in whey, texture quality of chakka and texture score of shrikhand are presented in Table 6.

It can be seen from the table that the coagulation time was inversely related to the heating temperature, while the curd tension increased along with increasing heating temperature. With the temperature range studied, the decreased coagulation time and increased curd strength coincided with a drop in the solids content of whey. It was observed that the lower heating temperature, especially at the higher pH values, resulted in turbid whey possibly due to casein particles escaping into the whey, increasing the solids losses.

The texture became coarser at the higher heating temperature with the coagulation pH of 4.8, whereas there was no perceptible change with the higher pH values of 4.9 and 5.0. Shrikhand obtained from different chakka,
Table 6. Effect of heating temperature and pH on coagulation behaviour of skim milk with lactic acid

<table>
<thead>
<tr>
<th></th>
<th>30°C</th>
<th>35°C</th>
<th>40°C</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>4.8 pH</td>
<td>4.9 pH</td>
<td>5.0 pH</td>
</tr>
<tr>
<td>Coagulation time (in min)</td>
<td>53.00 (51-55)</td>
<td>56.30 (55-58)</td>
<td>67.60 (66-69)</td>
</tr>
<tr>
<td>Curd strength (in g)</td>
<td>27.63 (27.10-28.40)</td>
<td>26.16 (25.9-26.4)</td>
<td>24.40 (23.7-24.9)</td>
</tr>
<tr>
<td>Solids losses in whey</td>
<td>6.74 (6.60-6.82)</td>
<td>7.22 (7.15-7.30)</td>
<td>7.36 (7.26-7.44)</td>
</tr>
<tr>
<td>Sensory quality 4</td>
<td>Text-</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td></td>
<td>ture of</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>chakka 2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Texture score of shrikhand 3</td>
<td>25.2</td>
<td>26.8</td>
<td>26.3</td>
</tr>
</tbody>
</table>

1 Using 1:9 dil lactic acid
2 x slightly grainy; xx moderately grainy
3 Body texture score out of 30 points
4 Average of three trials

Figures in parentheses indicate range
exhibited a texture score ranging from 25.20 to 26.80 out of 30, indicating little differences due to setting temperatures or pH.

Considering all parameters, a combination of 35°C curd setting temperature and 4.8 coagulation pH appeared to give a fairly firm curd of relatively low graininess. The solids losses were not excessively high under these coagulation conditions. The quality of shrikhand obtained from this curd also exhibited highest texture score of 26.8 (However, all the samples were criticized for graininess). Hence, this pH-temperature combination was adopted in further studies on direct acidification.

5.2.3 FURTHER ATTEMPTS ON IMPROVING CURD FORMING PROPERTIES

Using lactic acid for cold acidification at pH 4.8 and a setting temperature of 35°C, it was possible to obtain curd for shrikhand making within a period of 39 to 40 min. However, the shrikhand prepared from this curd was not entirely free from grainy defect. It was, therefore, decided in further attempts to alleviate this problem of graininess in direct acidified curd. For this, various modifications, viz. (a) temperature of heat treatment, (b) type of skimmilk, (c) addition of sodium chloride, (d) slow addition of acids, and (e) slow heating of the acidified skimmilk were tried out.

The curd obtained on draining of whey was prepared into shrikhand using ripened cream and sugar so as to
obtain 6% fat and 36% sugar in the finished product, and examined for sensory quality.

5.2.3.1 Heating temperature of skimmilk

It was thought that high heat treatment of buffalo skimmilk (85°C/20 min) might be responsible for poor textural characteristics of the resulting curd and, therefore, three different heat treatment of skimmilk were investigated, viz. 63°C/30 min, 72°C/16 sec, 85°C/10 min. The curd so obtained was used for shrikhand making and subjected to sensory evaluation. There was no improvement in the body or texture of the resulting product.

5.2.3.2 Type of skimmilk

It was postulated that the high Ca content of buffalo skimmilk and the nature of buffalo milk protein might be responsible for poor textural characteristics of the resulting curd. Hence, buffalo skimmilk was compared with cow: buffalo (50:50) and cow skimmilk.

It was noticed that shrikhand prepared from all the three types of milk were also grainy in texture. However, the cow milk produced slightly less graininess than that obtained with mixed milk, which was slightly better than the buffalo milk product.

5.2.3.3 Addition of sodium chloride

In order to counteract a possible influence of high Ca content of buffalo skimmilk which might be responsible for textural characteristics of the resulting curd, attempts
were made to add common salt in milk with the aim of replacing divalent (Ca\(^{++}\)) ions with monovalent (Na\(^{+}\)) ions (Fahmi and Sharana, 1950; Mahmoud et al., 1973) in systems like milk. Therefore, the effect of adding 0.1, 0.2 and 0.5% common salt to buffalo skimmilk, on the graininess in curd, was examined. Although slight improvement was recovered in the texture of the product when 0.2% or more salt was used, it was criticized for saltiness. Washing of chakka with water (amount equal to the whey expelled from the curd) removed the saltiness defect only partially. Thus, simple addition of common salt to buffalo skimmilk was not considered practicable.

5.2.3.4 Slow addition of acid

Instant addition of acid might be responsible for inferior texture of shrikhand. Therefore, attempts were made to add acid in such a way that it takes 30, 60 and 120 min under constant agitation. The mix was slowly warmed to 35\(^\circ\)C for curd setting. The curd so obtained was used for shrikhand making and subjected to sensory evaluation. There was no improvement in body and texture. All the three samples rated as grainy in texture.

5.2.3.5 Slow tempering of acidified milk

Slow tempering of the acidified skimmilk (4\(^\circ\)C) at various water bath temperatures was studied earlier (5.2.2.2.1) wherein the final temperature of acidified skimmilk decreased with the decrease in the water bath
temperature and so the heating rate. With a view to decrease the rate of heating while keeping the final temperature desirably at 35°C, the warming up was carried out in several stages such as first 5 min at 5°C, then 10 min each at 10°C and 15°C, 15 min each at 20°C and 25°C, and finally 20 min at 35°C. However, there was no perceptible difference in the texture of the product obtained by this method with that from the single stage heating process.

5.2.4 LIMITATION OF DIRECT ACIDIFICATION

The application of direct acidification technique was investigated with the basic aim of enhancing the rate of acid production so that skim milk curd can be obtained within a time period of 30 to 60 min. Although cold acidification with lactic acid at pH 4.8 and setting at 35°C could result in curd setting within 39-40 min, the resulting chakka or shrikhand from this system had a characteristic graininess.

Applying several modifications to induce the formation of softer coagulum with buffalo skim milk, partial replacement of ionic calcium, simulating a gradual curd forming effect of acid by slow raise of temperature etc. did not remove the grainy defects completely.

5.3 USE OF ALTERNATE SOURCES OF MILK SOLIDS FOR SHRIKHAND

Shrikhand, being a summer speciality, its manufacture from fluid milk in summer implies its diversion from fluid consumption during the lean season. Therefore, preparation of shrikhand from other sources of milk solids,
that can be conveniently manufactured during the flush season, would enable its economic production during summer.

Although skimmilk powder would be the product of choice for this purpose, it was decided to also include plain condensed skimmilk and fresh buttermilk in the study. Since the solids level of condensed milk and the chakka is within the range of 22 to 25%, it was tempting to develop a technology that would enable the development of acidity, by passing the whey drainage step. Since unsweetened evaporated buffalo skimmilk is not commercially available under Indian conditions, plain unsweetened condensed buffalo skim milk was used as a nearest candidate. Varying amounts of fresh buttermilk is often available in modern dairies manufacturing creamery butter from pasteurized cream. 

Shreshtha and Gupta (1979) used buttermilk for preparing an acceptable quality dahi by lactic fermentation. It was, therefore, decided to extend the use of buttermilk to shrikhand making.

Initial studies were concerned with the quality of curd that could be obtained from skimmilk powder, plain condensed milk and buttermilk. Shrikhand was then prepared from these systems for organoleptic evaluation and storage studies.

5.3.1 RATE OF ACID DEVELOPMENT IN DIFFERENT MILK SOLIDS SYSTEMS

To study the progress of fermentation with Yi culture, various sources of milk solids were given an appropriate
heat treatment, tempered to 42°C and were inoculated with 2% active culture of YH. The development of acidity and changes in pH were recorded at an interval of one hour.

5.3.1.1 Effect of total solids level in plain condensed milk

Condensed skim milk was adjusted to different TS levels ranging from 30 to 43% by adding calculated quantities of water into concentrated milk of higher TS levels. Due to the problem of heat stability and browning, high TS containing samples were given a heat treatment of just 85°C/10 min instead of 30 min given to 10% TS containing skim milk.

The rate of acid development and pH changes brought about by the YH culture in condensed skim milk containing 30, 35 and 43% solids have been depicted in Fig.11. It is evident that with the increasing solids level, the rate of acid production as well as the final acidity attained after 6 hrs incubation decreased considerably. Solids content above 35% appeared to be far less conducive to the acid development, as the acidity difference after 6 hrs between 35 and 38% solid levels was appreciably greater than that between 30 and 35% or 38 and 43% solids level. Although the highest acidity achieved (1.35% LA) was attained by 30% condensed skim milk after 6 hrs, it was much lower than that of the skim milk chakka where it is normally in the range of 1.8 to 2.0%.

There was a corresponding trend in pH values as a
FIG. 11. EFFECT OF CONCENTRATION OF MILK ON THE RATE OF ACID DEVELOPMENT AND CHANGES IN pH USING YH CULTURE
result of variation in TS level. At the end of 6 hrs, a minimum pH value of 4.8 was attained at 30% TS level, whereas the 43% TS containing system attained a pH value of only 6. It may be recalled (Fig.2), that in a 10% TS containing system, the YH culture brought down the pH to the level of 4.1 to 4.2 within 4 hrs incubation at 42°C.

With the hope of enhancing the fermentation rate, plain condensed milk containing 30 and 35% TS were also incubated with 5% YH culture. The comparative rates of acid development and pH changes of 2 and 5% YH culture-containing systems are presented in Fig.12. The changes in acidity and pH indicate only slight higher acidity levels and slight lower pH values effected, as the culture level increased from 2 to 5%. However, in comparison to chakka, these acidity and pH values are far from the desirable ranges. The hampered acidity development by yoghurt cultures at higher solids concentrations have been attributed to the increased buffering capacity (Tramer, 1973) as well as the retarded growth of the culture organisms on account of the higher osmotic pressure.

5.3.1.2 Rate of acid development in different sources of milk solids at 10% TS level

As the activity of starter culture is affected by higher TS level of the substrate, it was decided to use the YH culture in reconstituted skimmilk, buttermilk and condensed milk, at a much lower level of 10% TS for comparing with its activity in 10% TS containing skimmilk control
FIG. 12. EFFECT OF AMOUNT OF INOCULUM ON THE RATE OF ACID DEVELOPMENT AND pH CHANGES IN CONCENTRATED MILK
system. After standardizing the various milk systems to 10% TS, these as well as the skim milk were heat treated at 85°C/30 min and were inoculated with 2% active YH culture at 42°C. Acidity development and pH changes studied over a period of 4½ and 5 hrs are depicted in Fig.13(a). The data indicate that in comparison to the skim milk system, the rate of acidity development and pH changes in buttermilk, reconstituted skim milk and plain condensed milk were more similar at a 10% TS level than those experienced by the same condensed milk at three different TS levels (section 5.3.1.1). Within a 4½ hrs period, all the samples attained an acidity level of 0.7% LA or more. However, the acidity development in fresh skim milk was slightly better than that in the other milk systems. There was no perceptible difference between diluted condensed skim milk and reconstituted skim milk. The acid development in buttermilk was a little faster initially, but later it declined considerably, ending at the lowest final acidity of 0.75% after 5 hrs.

The trend in pH changes in the different milks were similar to the effect on acid development (Fig.13b), the drop in pH was maximum (3.9) in skim milk, followed by a value of 4.1 in reconstituted milk and dilute condensed milk, and 4.4 in buttermilk. Of the three sources of milk solids tested at 10% TS level for the activity of YH culture, buttermilk exhibited a somewhat lower cultural activity.

It is not certain as to what extent this reduced level of starter activity related to the differences in the processing treatments, particularly heating, which the
FIG. 13. RATE OF ACID DEVELOPMENT AND CHANGES IN pH IN DIFFERENT SOURCES OF MILK SOLIDS AT 10% TS LEVELS
products such as skim milk powder, plain condensed milk and buttermilk have to undergo during manufacture.

5.3.2 CHARACTERISTICS OF CURD PREPARED FROM 10% TS CONTAINING MILK SYSTEMS

The data on the quality of curd prepared from 10% TS containing milk system in terms of curd tension, colour and flavour are presented in Table 7. In the order of decreasing curd strength, skim milk systems were followed by those prepared from dilute condensed milk, reconstituted skim milk (RSM) and buttermilk. The differences in the colour and taste attributes of various samples were only slight. The slightly yellowish colour noticed in the curds prepared from systems other than skim milk, may possibly be due to the varying degrees of heat treatment undergone by those systems during manufacture. RSM curd also had a slightly powdery flavour.

On the basis of curd forming qualities, the buttermilk system was rated lowest among the three alternative sources of milk solids, in comparison to skim milk systems. In addition to the formation of a weak coagulum, buttermilk curd also exhibited poor whey drainage properties and was criticized for somewhat unnatural flavour. TVFA content of buttermilk curd was the highest (5.28) as compared to other curd systems. The unnatural flavour in buttermilk systems may be related to this high TVFA content as well as the presence of phospholipids.
Table 7. Characteristics of curd obtained from different MSNF systems

<table>
<thead>
<tr>
<th>Properties</th>
<th>Skim milk</th>
<th>Reconstituted skim milk</th>
<th>Butter milk</th>
<th>Dilute condensed milk</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>-10% TS</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Curd tension (g)</td>
<td>98.78</td>
<td>59.00</td>
<td>47.60</td>
<td>82.05</td>
</tr>
<tr>
<td></td>
<td>(96.40-101.20)</td>
<td>(58.30-59.80)</td>
<td>(46.60-48.40)</td>
<td>(75.90-86.30)</td>
</tr>
<tr>
<td>Colour</td>
<td>slight greenish, white</td>
<td>slight yellowish dull, white</td>
<td>V.slightly yellowish white</td>
<td>slightly yellowish white</td>
</tr>
<tr>
<td>Flavour</td>
<td>typical acidic taste</td>
<td>slightly powdery flavour</td>
<td>slightly unnatural taste</td>
<td>acidic</td>
</tr>
<tr>
<td>TVFA 0.01 ml/50 g curd</td>
<td>2.4 (2.3-2.50)</td>
<td>2.30 (2.2-2.40)</td>
<td>5.28 (5.16-5.33)</td>
<td>2.70 (2.60-2.85)</td>
</tr>
</tbody>
</table>

* Average of three trials
Figures in parentheses indicate range
5.3.2.1 Effect of Ca and skim milk powder (SMF) addition on buttermilk-curd

Improvement in the firmness of buttermilk-curd obtained in the previous study (section 5.3.2) was attempted through simple approaches. Increasing the availability of ionic calcium and the total solids level of the butter milk could be practical under plant conditions.

5.3.2.1.1 Effect of Ca addition

Curd strength of milk can be enhanced by incorporating Ca\(^{++}\) ion in the form of CaCl\(_2\) solution (Robertson and Gilles, 1969; Thomasow and Schmanko, 1968), as often practiced in case of American cottage cheese.

In order to study the effect of increased levels of ionic calcium on the quality of buttermilk curd, a calculated amount of 4% aqueous CaCl\(_2\) solution was added to incorporate Ca\(^{++}\) ions at the rate of 8 g, 12 g and 16 g per 100 litres of milk, at the end of 85°C/30 min heat treatment. As can be seen from Fig.14(a), the curd tension (after 4 hrs incubation at 42°C, and 4.1-4.2 pH) increased with the increased level of added calcium. However, the marginal increase in the curd tension (from 45 to about 52 g) did not cause any perceptible improvement in the whey drainage properties.

5.3.2.1.2 Effect of SMP addition

Since the curd firmness is related to the TS level of milk, effect of increasing the TS content of buttermilk
FIG. 14. EFFECT OF Ca\(^{++}\) AND SMP FORTIFICATION ON CURD TENSION OF BUTTERMILK CURD
to levels higher than 10% on the quality of curd formed was studied by fortifying the system by SMF. In this study, SMF was dissolved into buttermilk at 1, 2 and 3% levels, and the system was then given a heat treatment of 85°C/30 min, prior to YH culture inoculation at 42°C. Since these milk systems, fortified with different quantities of SMF had different levels of TS, it was decided to study the rate of acid development and pH changes in these systems, along with the measurement of curd tension values.

It can be seen from Fig. 14(b) that an increasing level of fortification with SMF was accompanied by an increase in the curd firmness. However, increase in curd firmness was more between 1 and 2% level of SMF addition than between 2 and 3% addition levels.

The rate of acidity development as well as pH changes showed little differences in the activity of the YH culture due to the increasing TS levels within the range studied (Fig. 15a, b). However, the whey drainage properties of the SMF fortified system showed improvement over that could be attained with simple addition of ionized calcium.

A comparison of the two methods of enhancing the firmness of buttermilk curd, placed the SMF fortification approaches in a position somewhat more favourable than the calcium chloride addition method. Increasing the milk solids level earned to be more attractive in terms of better whey drainage properties and yield of chakka than a mere gain in curd firmness. Since there was not much
FIG. 15. CHANGES IN pH AND ACIDITY UPON SMP FORTIFICATION IN BUTTER MILK.
difference between the 2 and 3% level of SMF addition, 
fortification of buttermilk with 2% SMF was accepted for 
进一步 studies on shrikhand making.

5.3.3 CONVERSION OF CURD FROM DIFFERENT SOURCES OF 
MILK SOLIDS INTO CHAKKA

Although lactic curd can be prepared from different 
sources of milk solids using Yι cultu re, there exist a 
variation in the quality of curd formed from these sources. 
Differences in curd tension are known to be associated with 
a variation in the moisture retention capacity of the 
protein system. Relatively weaker curds cause higher losses 
of solids in whey. Thus, any variation in the moisture 
holding properties of the curd as well as solids losses in 
whey might result in a variation in the quality and yield of 
chakka, obtained at the end of whey drainage steps. In 
order to study the recovery of solids and yield of chakka 
obtained from various milk systems, curd was set according 
to the procedure developed (section 5.1.7) and subjected to 
basket centrifuging (at 1,000 to 1,2000 rpm for 40 min) for 
obtaining chakka. The data presented in Table 8 show that 
the TS level of various milk systems remained close to 10% 
TS level, except in the case of buttermilk where a 2% 
addition of SMF resulted in a TS level of about 12%. The 
percentage yield of chakka was highest with SMF-fortified 
system, followed by the skim milk. On the basis of % TS in 
chakka, the moisture retention behaviour of these two curd 
systems also appeared to be similar. RSM and diluted 
condensed milk systems exhibited slightly higher level of
<table>
<thead>
<tr>
<th>Particulars</th>
<th>Skim milk</th>
<th>Reconstituted skim milk</th>
<th>Dilute plain condensed skim milk</th>
<th>Butter milk</th>
</tr>
</thead>
<tbody>
<tr>
<td>TS of milk</td>
<td>10.14</td>
<td>10.05</td>
<td>10.10</td>
<td>11.98</td>
</tr>
<tr>
<td></td>
<td>(9.99-10.31)</td>
<td>(9.97-10.12)</td>
<td>(10.02-10.14)</td>
<td>(11.80-12.18)</td>
</tr>
<tr>
<td>% protein in milk</td>
<td>3.87</td>
<td>3.83</td>
<td>3.81</td>
<td>4.12</td>
</tr>
<tr>
<td></td>
<td>(3.83-3.92)</td>
<td>(3.79-3.85)</td>
<td>(3.76-3.86)</td>
<td>(3.98-4.21)</td>
</tr>
<tr>
<td>Time (min) required to obtain a pH 4.3-4.2</td>
<td>220</td>
<td>250</td>
<td>245</td>
<td>282</td>
</tr>
<tr>
<td>Acidity of curd</td>
<td>0.914</td>
<td>0.911</td>
<td>0.894</td>
<td>0.845</td>
</tr>
<tr>
<td></td>
<td>(0.909-0.918)</td>
<td>(0.909-0.913)</td>
<td>(0.855-0.869)</td>
<td>(0.805-0.889)</td>
</tr>
<tr>
<td>% yield of chakka</td>
<td>24.10</td>
<td>23.12</td>
<td>23.58</td>
<td>26.11</td>
</tr>
<tr>
<td></td>
<td>(23.90-24.40)</td>
<td>(22.95-23.31)</td>
<td>(23.33-23.72)</td>
<td>(25.89-26.28)</td>
</tr>
<tr>
<td>% TS of chakka</td>
<td>23.23</td>
<td>22.85</td>
<td>22.83</td>
<td>23.08</td>
</tr>
<tr>
<td></td>
<td>(23.18-23.31)</td>
<td>(21.78-22.98)</td>
<td>(22.79-22.92)</td>
<td>(22.98-23.20)</td>
</tr>
<tr>
<td>% proteins of chakka</td>
<td>13.90</td>
<td>13.64</td>
<td>23.58</td>
<td>13.66</td>
</tr>
<tr>
<td>% TS recovery</td>
<td>54.520</td>
<td>52.540</td>
<td>53.289</td>
<td>50.308</td>
</tr>
<tr>
<td></td>
<td>(53.620-55.05)</td>
<td>(50.92-54.272)</td>
<td>(52.734-53.881)</td>
<td>(49.58-51.12)</td>
</tr>
<tr>
<td>% TS in whey</td>
<td>5.96</td>
<td>6.34</td>
<td>6.16</td>
<td>7.89</td>
</tr>
<tr>
<td></td>
<td>(5.92-5.99)</td>
<td>(6.30-6.38)</td>
<td>(6.10-6.21)</td>
<td>(7.29-7.92)</td>
</tr>
<tr>
<td>% protein in whey</td>
<td>0.298</td>
<td>0.340</td>
<td>0.315</td>
<td>0.451</td>
</tr>
<tr>
<td></td>
<td>(0.294-0.301)</td>
<td>(0.335-0.348)</td>
<td>(0.312-0.319)</td>
<td>(0.453-0.465)</td>
</tr>
</tbody>
</table>

* Average of three trials

Figures in parentheses indicate range
moisture retention. The highest level of TS recovery was in the case of skimmilk-chakka, and lowest in case of buttermilk-chakka.

Analysis of total solids content of the whey also indicated that higher solids loss in whey corresponded to the lower TS recovery in the chakka. Curd tension data, obtained earlier in the section 5.3.2, clearly indicated that weaker curd forming properties were associated with higher solids losses in whey in case of buttermilk system, and higher moisture retention in case of the systems containing RMS and dilute condensed milk.

5.3.4 CHARACTERISTICS OF SHRINKHAND PREPARED FROM VARIOUS MSNF SYSTEMS

Shrikhand was prepared with the chakka samples made from the four different MSNF systems, viz. fresh skimmilk, diluted condensed skimmilk, reconstituted skimmilk and buttermilk (containing 2% SMF) and from the curd obtained with 30% TS condensed skimmilk according to the procedure outlined in Fig.6 (section 5.1.7). These samples were filled in 60 g size plastic cups (fitted with snap-in lids) for organoleptic evaluation, compositional analysis and storage studies.

5.3.4.1 Sensory characteristics

For sensory evaluation, the samples were tempered overnight at 10°C before presenting to the trained judges. It is apparent from Table 9 that there were no significant variations in the colour score of the product prepared from
Table 9. Sensory characterisation of shrikhand prepared from various MSNF systems*

<table>
<thead>
<tr>
<th>Attributes</th>
<th>Colour</th>
<th>Appearance</th>
<th>Body and texture</th>
<th>Flavour</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>Skim milk shrikhand</td>
<td>4.12 (4.08-4.15)</td>
<td>7.50 (7.41-7.65)</td>
<td>26.62 (26.51-26.79)</td>
<td>51.52 (51.28-51.73)</td>
<td>good aroma, smooth body and texture</td>
</tr>
<tr>
<td>Shrikhand from condensed milk (30% TS)</td>
<td>3.97 (3.90-4.10)</td>
<td>6.90 (6.80-7.08)</td>
<td>23.21 (23.18-23.41)</td>
<td>46.51 (46.38-46.75)</td>
<td>wet body, salty taste, powdery flavour, less acidic, too sweet</td>
</tr>
<tr>
<td>Shrikhand from reconstituted milk</td>
<td>3.90 (3.82-4.02)</td>
<td>7.42 (7.33-7.56)</td>
<td>26.25 (26.19-26.39)</td>
<td>50.92 (50.83-51.28)</td>
<td>slight dull colour, very smooth body and texture, slight weak body</td>
</tr>
<tr>
<td>Shrikhand from dilute condensed milk (4.05-4.16)</td>
<td>4.10 (4.05-4.16)</td>
<td>7.40 (7.28-7.52)</td>
<td>25.91 (25.75-26.05)</td>
<td>51.43 (51.29-51.71)</td>
<td>pleasant aroma, smooth body and texture</td>
</tr>
<tr>
<td>Shrikhand from butter milk</td>
<td>4.00 (3.95-4.06)</td>
<td>7.09 (6.98-7.18)</td>
<td>26.13 (25.98-26.25)</td>
<td>51.10 (50.99-51.32)</td>
<td>slight dull colour, very smooth texture, weak body</td>
</tr>
</tbody>
</table>

* Average of three trials

Figures in parentheses indicate range
the various types of chakka. However, it was noticed that condensed skim milk and buttermilk gave a slightly yellowish product, while reconstituted skim milk resulted in a dull yellowish shririkhand. The buttermilk product was observed to have a slightly more silky appearance than other samples with regard to the body and texture characteristics. The condensed skim milk product definitely lacked the firmness of the control samples. It had a distinctly weak and soggy body. The texture of shririkhand from reconstituted skim milk or buttermilk was notably smoother than other samples, presumably because of the relatively weaker curd formation.

Shrihakd obtained from dilute condensed milk, reconstituted skim milk and buttermilk had nearly the same scores in the flavor as the skim milk shririkhand. However, the condensed skim milk product was definitely less desirable than the control. It was criticized for saltish taste and low acidity. On these grounds it was found unsuitable for shririkhand making. One more implication of low acid production was that insufficient sugar could be incorporated into the final product which affected consistency and yield of the product.

5.3.4.2 Composition of shririkhand

The compositional characteristics of various shririkhand samples analysed in terms of total solids, total proteins, reducing sugar, fat, acidity, and pH are
presented in Table 10. The TS content varied from 59.43 to 60.04%, except for shrikhand made from concentrated milk, having substantially lower solids (52.08%), because inadequate acid development did not permit sufficient addition of sugar. For the same reason, other constituents were somewhat lower in shrikhand from condensed milk, although reducing sugar was substantially higher in it (obviously on account of elimination of the whey drainage step).

In general, the composition of shrikhand made from the different systems except that from condensed milk agree with the values reported earlier (Aneja et al., 1977; Upadhyay and Dave, 1975; Miyani, 1982).

5.3.4.3 Shelf-life of shrikhand

Further studies were conducted to assess the shelf-life of shrikhand made from the four different MSNF systems. The product, in 60 ml plastic cups, was stored at 10 ± 2°C for 60 days. The samples were subjected to sensory evaluation and analysed for moisture, total protein, sucrose, fat, soluble nitrogen, reducing sugar, TVFA, acidity and pH. Microbiological quality of the samples was determined in terms of total viable count (TVC), lactic count, proteolytic count and yeast and mould count (YM).

5.3.4.3.1 Changes in sensory properties

The shrikhand samples were scored for flavour
Table 10. Average composition of shrikhand obtained from different MSIP*

<table>
<thead>
<tr>
<th>Particulars (%)</th>
<th>Shrikhand from skim milk</th>
<th>Shrikhand from condensed milk</th>
<th>Shrikhand from reconstituted milk</th>
<th>Shrikhand from dilute condensed milk</th>
<th>Shrikhand from butter milk</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total solids</td>
<td>59.879 (58.893-60.987)</td>
<td>52.083 (51.890-52.263)</td>
<td>59.883 (58.873-60.886)</td>
<td>60.0401 (60.401-59.690)</td>
<td>59.435 (60.085-58.980)</td>
</tr>
<tr>
<td>Sucrose</td>
<td>40.923 (40.797-41.051)</td>
<td>32.32 (32.18-32.49)</td>
<td>40.892 (40.68-41.20)</td>
<td>40.690 (40.521-40.831)</td>
<td>41.167 (40.98-41.34)</td>
</tr>
<tr>
<td>Reducing sugar</td>
<td>1.621 (1.612-1.628)</td>
<td>7.13 (7.05-7.23)</td>
<td>1.809 (1.72-1.877)</td>
<td>1.690 (1.63-1.720)</td>
<td>2.173 (1.959-2.310)</td>
</tr>
<tr>
<td>Fat</td>
<td>5.98 (5.65-5.99)</td>
<td>5.96 (5.92-6.05)</td>
<td>6.09 (5.98-6.20)</td>
<td>6.16 (6.10-6.20)</td>
<td>6.13 (6.098-6.210)</td>
</tr>
<tr>
<td>Acidity (% LA)</td>
<td>1.054 (1.044-1.066)</td>
<td>0.732 (0.723-0.745)</td>
<td>1.013 (1.010-1.016)</td>
<td>1.040 (1.012-1.088)</td>
<td>1.021 (1.017-1.044)</td>
</tr>
<tr>
<td>pH</td>
<td>4.10 (4.1-4.1)</td>
<td>4.80 (4.7-4.90)</td>
<td>4.20 (4.15-4.30)</td>
<td>4.20 (4.10-4.30)</td>
<td>4.25 (4.15-4.40)</td>
</tr>
</tbody>
</table>

* Average of three trials
Figures in parentheses indicate range
(55 point max), body and texture (30 point max), appearance (10 point max) and colour (5 points max) for a period of 40 days and evaluated at 10 days intervals. The results have been presented in Appendix VIII_a,b. The flavour scores depicted in Fig.16 tended to decline during storage, the decline being slow for first 20 days followed by a relatively rapid drop, particularly during the last 10 days. While there was no perceptible difference in the rate of change of flavour scores among the shrikhands made from skim milk, reconstituted skim milk and dilute condensed milk; flavour of shrikhand made from buttermilk, showed somewhat faster deterioration. Considering a score of 40 as the minimum desirable one, the first three types of product kept well for about 35 days, and the buttermilk product for about 28 days.

The body and texture score of the product did not show any definite trend during storage. In general, slight graininess and tough body were the criticisms expressed for all types of stored product. As storage progressed, the product became dull in appearance, mainly at the surface because of loss of moisture. Towards the end of storage, there appeared green discolorations due to mould growth in samples.

5.3.4.3.2 Changes in moisture, pH and acidity

The storage period influenced the moisture content, pH and acidity among other factors. The moisture content was expected to change during storage for the reason of
FIG. 16. CHANGES IN FLAVOUR SCORE OF DIFFERENT SHRIKHAND DURING STORAGE AT 10°C
evaporation losses attributed to unsealed packaging of the product.

Fig. 17 shows that there was a steady drop in the moisture content of all the samples. Apparently there was no definite influence of the type of shrikhand on the moisture, although slightly enhanced moisture losses were observed in reconstituted and buttermilk shrikhand samples towards the end of storage.

The acidity of the product increased during storage in all types of shrikhand. The increase was highest in buttermilk followed by reconstituted skimmilk, dilute condensed skimmilk and skimmilk shrikhand, respectively (Fig. 18a). Correspondingly, there was a decline in pH (Fig. 18b), although no definite pattern with respect to the type of starting material was noted. The lactic counts were reduced in all the four types of shrikhand (Fig. 19). The highest decline was in reconstituted skimmilk-shrikhand, whereas almost the same trend was observed in other types of shrikhand.

Since lactic counts declined along with an increasing acidity, the development of acidity during storage can be attributed to the activity of the non-starter organisms. Increased acidity during the storage is largely due to the fermentation activity of YM. It is quite apparent from Fig. 20 that an increase in the counts of YM represented about 100-fold increase during 50 days storage at 10°C.
FIG. 17. CHANGES IN MOISTURE CONTENT OF SHRIKHAND DURING STORAGE AT 10 °C
FIG. 18. CHANGES IN ACIDITY AND pH OF SHRIKHAND DURING STORAGE AT 10°C
FIG.19. CHANGES IN LACTIC COUNTS OF SHRIKHAND DURING STORAGE AT 10°C
FIG. 20. CHANGES IN YM COUNTS OF SHRIKHAND DURING STORAGE AT 10°C
The growth of YM in various shrikhand samples was essentially similar. However, coliform was not found in shrikhand during storage.

5.3.4.3.3 Changes in carbohydrates and TVFA

It is seen from Fig. 21(a) that during storage there was an increase in reducing sugar with a simultaneous decrease in sucrose as shown in Fig. 21(b). The increase being rapid after first 20 days. Also buttermilk product had a high reducing sugar content from beginning to end.

The sucrose content decreased during storage in all the samples. The decrease being slightly faster in the case of buttermilk than in other samples. Decreasing sucrose level, along with an increasing level of reducing sugar during the storage of shrikhand samples is largely due to utilization of sucrose by the rapidly growing YM as their energy source. Similar observations were reported by Sharma and Zariwala (1980) and Upadhyay (1981).

The TVFA level can be considered as an indicator of lipolytic activity in system containing butter fat. During the storage at 10°C, TVFA content of all shrikhand samples registered gradual increase as shown in Fig. 22. The rate of increase in TVFA level appeared to be somewhat lower for the reconstituted skim milk system than the other three. Buttermilk system, however, represented a very much higher level of TVFA, right from the beginning. Increasing TVFA content of shrikhand is also due to the lipolytic activity of YM.
FIG. 21. CHANGES IN REDUCING SUGAR AND SUCROSE OF SHRIKHAND DURING STORAGE AT 10°C
FIG. 22. CHANGES IN TVFA OF SHRIKHAND DURING STORAGE AT 10°C
5.3.4.3.4 Changes in soluble nitrogen and proteolytic counts

Proteolytic behaviour of shrikhand was measured in terms of soluble nitrogen and proteolytic counts. The YM culture is known for proteolysis in a system like milk (Dutta et al., 1971; Formisano et al., 1971; Luca, 1975).

It is clear from the Figs. 23(a) and 23(b) that during storage, soluble nitrogen and proteolytic counts increased. The rate of increase being more or less same in all cases although in the buttermilk shrikhand it was higher than the other samples throughout storage.

5.3.4.3.5 Shelf life

Data on organoleptic evaluation, changes in acidity, pH, soluble nitrogen, TVFA and microflora, particularly in terms of lactic count, YM and proteolytic counts indicated an average shelf life of these shrikhand samples within the range of 28 to 35 days at 10°C. Most samples were within the range of 30 to 35 days. However, the buttermilk shrikhand samples had a shelf-life of about 28 days.

Deterioration in the keeping quality was usually associated with an increase in activity of YM and proteolytic microflora under refrigerator (10°C) conditions of storage. Increasing YM activity was associated with a reduction in sucrose levels, and an increase in the levels of reducing sugar, titratable acidity, TVFA and soluble nitrogen. The spoiled samples exhibited characteristics, such as fruity, fermented, rancid and aromatic traits along
FIG. 23. CHANGES IN SOLUBLE NITROGEN AND PROTEOLYTIC COUNTS DURING STORAGE AT 10°C
with surface discolouration due to mould and yeast growth. All the shrikhand samples were characterized essentially by the similar spoilage behaviour during storage, although in buttermilk shrikhand system this was somewhat faster than the others.

5.4 Enhancing the Keeping Quality of Shrikhand

It was quite apparent from the storage behaviour of shrikhand that extending the shelf-life would essentially depend on effectively controlling the YM activity during storage. This could be attempted in principle by minimizing the YM contamination during the various stages of manufacturing shrikhand and by retarding the activity of YM during storage. Under normal conditions of shrikhand manufacture, involving heat treated milk, incubated with contamination-free active culture, in a commercially sanitized curd tank, little YM activity could be expected up to the chakka making stage. However, during the vigorous mixing and kneading of chakka with other ingredients such as pasteurized cream, sugar and flavouring agent, contamination might occur. Of the various ingredients added to chakka, initial quality of sugar, a major ingredient of shrikhand required particular attention. Variation in the YM counts in sugar might result in contamination of shrikhand with different load of YM. It was, therefore, decided to study the effectiveness of heat treatment of sugar on the YM counts and keeping quality of shrikhand.
In an attempt to check the YM activity in shrikhand during storage, a permissible food grade antimycotic agent, viz. potassium sorbate (ps) was used.

5.4.1 EFFECT OF HOT AIR TREATMENT OF SUGAR

To eliminate YM contamination through sugar, different time-temperature combinations were used for hot air treatment of commercial grade crystalline and ground sugar. Data presented in Fig. 24 reveal that the time required to reduce the YM count to zero was 30, 20, 10, 7.5 and 5 min at oven temperature of 120, 140, 160, 180 and 200°C, respectively. However, temperatures above 140°C tended to damage the sugar colour because of non-uniform heating. Hence, 140°C, a temperature requiring relatively short time (20 min) was selected for the sugar treatment.

Further studies were conducted to assess the shelf-life of shrikhand made from buffalo skim milk using the standardized method as outlined in Fig. 6 with and without the sugar treatment. The product was stored at 10°C for 60 days. The samples were analysed for sensory characteristics, chemical properties like pH, acidity and also for YM count.

5.4.1.1 Changes in sensory properties

The shrikhand samples were scored for flavour, colour, body and texture, and appearance using score card (Fig. 1b).

The flavour score depicted in Fig. 25 tended to
FIG. 24. EFFECT OF DIFFERENT TEMPERATURE AND TIME ON YM COUNTS OF SUGAR
FIG. 25. CHANGES IN FLAVOUR SCORE OF CONTROL AND HOT AIR TREATED SUGAR SHRIKHAND SAMPLES DURING STORAGE AT 10°C
decline during storage, the decline being slow for first 20 days followed by relatively rapid drop during the last 10 days. There was no perceptible differences in the rate of change of flavour scores between the control sample and hot air treated sugar sample.

The body and texture score of the product also decreased during storage. The product appeared to deteriorate also with respect to colour. There was no perceptible difference in the changes in colour between the samples during storage.

5.4.1.2 Changes in acidity and pH

As shown in Fig.26(a), the acidity was recorded to increase during storage and there was no perceptible difference in the rate of acid development between the samples. The rise in acidity was accompanied by drop in pH 26(b), the trend in declining of pH is nearly same in both the samples.

There was an increase in YM counts in both the samples (Fig.27). No perceptible difference was noted in YM counts.

From the data on organoleptic evaluation, changes in pH and acidity and YM counts, it is clear that there was no advantage of using hot air treated sugar for the purpose of increasing the keeping quality of shrikhand. Therefore, the hot air treated sugar was excluded from further studies.
FIG. 26. CHANGES IN ACIDITY AND pH OF SHRIKHAND DURING STORAGE AT 10°C
FIG. 27. CHANGES IN YM COUNTS OF SHRIKHAND DURING STORAGE AT 10°C
5.4.2 USE OF POTASSIUM SORBATE

In order to minimize the spoilage of shrikhand due to 
YM growth, potassium sorbate added at the rate of .01, 
.025, .05 and 0.1% was used as preservative. The required 
amount of sorbate was dissolved in a small quantity of 
water and the solution was added at the last stages of 
kneading operation. The samples stored in 60 ml plastic 
cups at 10°C ± 2°C were examined for organoleptic proper-
ties, microbial quality and chemical parameters viz. 
acidity, pH, reducing sugar, soluble nitrogen and TVFA at 
intervals of 10 days.

5.4.2.1 Changes in sensory property

It was observed that as the storage period increased, the flavour scores decreased (Fig. 28). The control 
sample kept well for 35 days, although it lost its freshness 
after 20 days. Samples treated with .01, .025, .05 and 0.1% 
level were acceptable for 37, 44, 53 and 55 days, 
respectively. Thus, the sorbate addition appeared to impro-
ve the keeping quality of the product. The favourable 
influence of the preservative increased with increasing 
levels of addition, although the 0.1% level was not appre-
ciably better than the 0.05% one. Hence, sorbate addition 
at 0.05% rates was chosen for subsequent storage studies. 
Higher level of potassium sorbate had an influence on the 
initial flavour score of the shrikhand samples.

With respect to colour, there was no definite trend
FIG. 28. CHANGES IN FLAVOUR SCORE OF SHRINKHAND TREATED WITH DIFFERENT PS LEVELS AT 10°C
noticed during storage although the spoilage was generally slower in the samples containing the preservatives.

5.4.2.2 Effect on acidity, pH and YM counts

The acidity increased gradually during storage in all the samples of shrikhand, the increase being less with the sorbate-added samples (Fig. 29a). There was a slight drop in pH during storage (Fig. 29b) and again the rate of decrease was rather low in the preservative containing product.

The YM counts increased during storage (Fig. 30). The increase being highest in control shrikhand samples followed by .01%, .025%, .05% and 0.1% ps treated samples. Thus, potassium sorbate substantially retarded the growth of YM in shrikhand.

5.4.2.3 Changes in carbohydrates and TVFA

As shown in Fig. 31, the reducing sugar content of shrikhand increased perceptibly during storage, thereby indicating breakdown of sugar (sucrose) through microbial activity. While the rate of increase was considerably lower in samples treated with ps as compared to control, the rate of increase in reducing sugar was observed to be inversely proportional to the concentration of ps. It is also observed that lower levels of the preservative (.01 and .025%) were not much effective in inhibiting the sugar breakdown. The TVFA content was found to increase as is clear from Fig. 32. Use of sorbate, especially at levels of .025% and above, helped to reduce the TVFA rise to a
FIG. 29. CHANGES IN ACIDITY AND pH OF SHRIKHAND TREATED WITH DIFFERENT PS LEVELS AT 10°C
FIG. 30. CHANGES IN YM COUNTS OF SHRIKHAND TREATED WITH DIFFERENT LEVELS OF PS AT 40°C
FIG. 31. CHANGES IN REDUCING SUGAR OF SHRINKHAND SAMPLES TREATED WITH VARIOUS LEVELS OF PS AT 10°C.
FIG. 32. CHANGES IN TVFA OF PS TREATED SHRIKHAND STORAGE AT 10°C
measurable extent. It indicated that the microbial action resulting in TVFA production was retarded by the preservative.

5.4.2.4 Changes in soluble nitrogen and proteolytic counts

It is clear from Fig.33(a) and Fig.33(b) that soluble nitrogen and proteolytic count increased with the advancing storage period. The increase was faster in the control as compared to that in samples containing 0.01, .025, .05 and 0.1% ps. The preservative retarded the changes leading to increase the soluble nitrogen.

It is thus clear from foregoing results on microbiological, chemical and sensory examination of the product during storage that potassium sorbate had a definitely beneficial effect on enhancing the keeping quality of the product. The extension of shelf-life was related essentially to the action of ps on retarding the activity of YM, the major spoilage microflora in shrikhand. By adding 0.05% or more potassium sorbate, keeping quality of the product was substantially increased from 35 to 53 days at 10°C. At all levels of 0.1%, potassium sorbate showed slightly adverse effect on the initial flavour of the product, presumably because of its own flavour. Hence, the level of 0.05% potassium sorbate was adopted for the future experiments.
FIG. 33. CHANGES IN SOLUBLE NITROGEN AND PROTEOLYTIC COUNTS OF SHRIKHAND DURING STORAGE AT 10°C
5.4.3 SHELIF LIFE OF POTASSIUM SORBATE ADDED SHRikhAND PREPARED FROM DIFFERENT SOURCES OF MILK SOLIDS

Spoilage behaviour of shrikhand prepared from various sources of milk solids (section 5.3.4.3) indicated YM as the major cause for limiting shelf-life with shrikhand prepared from skim milk. Potassium sorbate was effective in extending the shelf-life at 10°C up to 50 days. Studies were undertaken to observe the effect of 0.05% added ps on the shelf-life of shrikhand prepared from other sources of milk solids.

5.4.3.1 Changes in sensory properties

It can be visualized from Fig.34 that the keeping quality of shrikhand was highest with fresh skim milk (50 days) followed by reconstituted skim milk (46 days), dilute condensed milk (43 days) and buttermilk (36 days). However, their shelf-life was appreciably higher than that observed earlier without the preservative.

The changes in body and texture as well as colour and appearance were almost similar for all systems as observed earlier without the use of preservative. Although the rate of change was slightly slower with preservatives.

5.4.3.2 Changes in acidity, pH and YM counts

It can be seen from Figs.35(a) and 35(b) that the acidity of shrikhand samples increased and simultaneously pH decreased. The YM counts also increased as the storage period progressed (Fig.36). These changes were, however,
FIG. 34. CHANGES IN SENSORY PROPERTY OF SHRUKHAND TREATED WITH (0.05 PS) DURING STORAGE AT 10°C
FIG. 35. CHANGES IN ACIDITY AND pH OF SHRIKHAND DURING STORAGE AT 10°C
FIG. 36. CHANGES IN YM COUNTS OF SHRIKHAND DURING STORAGE AT 10°C
much slower as compared to those observed in samples without preservatives.

5.4.3.3 Changes in carbohydrates and TVFA

It can be seen from Figs. 37(a) and 37(b) that as the reducing sugar content increased, the sucrose content decreased during storage of shrikhand from the different MSNF systems. These changes, however, appeared to be related by the addition of ps. Fig. 38 indicates that the TVFA content increased during storage although the extent of this change was not as great as that observed without adding the preservatives. The rise was somewhat faster towards the end of storage.

5.4.3.4 Changes in soluble nitrogen and proteolytic counts

As can be seen from Figs. 39(a) and 39(b) that the soluble nitrogen as well as proteolytic counts increased during storage, although the extent of this change was not as great as that observed without adding the preservative. The soluble nitrogen content ran nearly parallel to the proteolytic counts of the shrikhand samples.

It was thus evident that the shelf-life of different shrikhand samples could be extended by the use of ps as the antimycotic agent, the relative difference between the samples was, however, almost unaltered when the preservative was used.

Due to the addition of ps, the average shelf-life of 28, 36, 37, 37 were extended to 36, 46, 44 and 50 days in
FIG. 37. CHANGES IN REDUCING SUGAR AND SUCROSE OF SHRIKHAND DURING STORAGE AT 10°C
FIG. 38. CHANGES IN TVFA OF SHRINKHAND
DURING STORAGE AT 10°C
FIG. 39. CHANGES IN SOLUBLE NITROGEN AND PROTEOLYTIC COUNTS OF SHRINKHAND DURING STORAGE AT 10°C
shrikhand prepared from buttermilk, dilute condensed milk, reconstituted skim milk and skim milk, respectively.

5.4.4 STORAGE AT ROOM TEMPERATURE

With a view to find out the storage stability of shrikhand at ordinary temperature, samples prepared from the different MSNF systems were stored at 30°C and examined for different chemical and sensory properties.

5.4.4.1 Changes in sensory score

As depicted in Figs. 40(a) and 40(b), the flavour score dropped rapidly for all the samples. The flavour deterioration was only slightly slower when ps was employed, the shelf-life being about 3 days without preservative and 4 days with preservative. The stored samples of shrikhand showed comparatively less changes in colour and appearance at 30°C as compared to low temperature storage.

There was slight surface drying on all samples and slightly mouldy appearance on the untreated samples. Similarly, there were comparatively smaller degree of changes with regard to body and texture too.

5.4.4.2 Changes in pH, acidity and YM

The acidity of shrikhand samples increased, whereas the pH decreased during storage. The changes were less pronounced when ps was used. Similarly, the YM counts showed appreciable increase but the increase was somewhat higher in the untreated samples (Figs. 41a, b; 42a, b and 43a, b).
FIG. 40. CHANGES IN FLAVOUR SCORE OF CONTROL AND PS TREATED SHRIKHAND DURING STORAGE AT 30°C
FIG. 41. CHANGES IN ACIDITY AND pH OF CONTROL SHRIKHAND DURING STORAGE AT 30°C
FIG. 42. CHANGES IN ACIDITY AND pH OF PS TREATED SHRINKHAND DURING STORAGE AT 30°C
Fig 43. Changes in YM counts of control and PS treated shrikhand during storage at 30°C
5.4.4.3 Changes in carbohydrates and TVFA

The reducing sugar and TVFA contents increased in the samples (Figs. 44a,b and 45a,b). This increase was slightly more in shrikhand stored without the addition of ps.

5.4.4.4 Changes in soluble nitrogen and proteolytic counts

It is clear from the Figs. 46(a)(b) and 47(a)(b) that soluble nitrogen and proteolytic counts increased during storage. This increase was slightly more in control shri-khand as compared to ps treated samples.

5.5 DISCUSSION

This investigation concerning the process alteration in shrikhand making technology was essentially aimed at developing a faster method of manufacturing shrikhand of a longer shelf-life and from milk solids that would be easily available in summer when the product is in great demand. Attempts of this study was also to select and to develop a set of methods that could be used to the advantage of large scale manufacturing of shrikhand such as being practiced successfully at the Baroda Dairy for many years (Aneja et al., 1977).

In the patented process of Aneja et al. (1977), the curd setting step of 8 hrs at 37°C appears to be the longest unit process among all other steps considered. This study, therefore, explored the possibility of cutting
FIG. 44. CHANGES IN REDUCING SUGAR OF CONTROL AND PS TREATED SHRIKHAND DURING STORAGE AT 30°C
FIG. 45. CHANGES IN TVFA OF CONTROL AND PS TREATED SHRIKHAND DURING STORAGE AT 30°C
FIG. 46. CHANGES IN SOLUBLE NITROGEN OF CONTROL AND PS TREATED SHRIKHAND DURING STORAGE AT 30°C
Fig. 47. Changes in proteolytic counts in control and PS treated shrikhand during storage at 30°C.
down further the curd setting time. For this purpose, curd setting with a yoghurt culture, YH, at 42°C gave the most encouraging result. With this culture it was possible to bring about a 50% reduction in the curd setting time.

A further reduction in curd setting time through a direct acidification did not yield encouraging results. Although cold acidification, followed by a slow warm up, gave within 1 hr skim milk curd comparable to that prepared with lactic culture. A distinct grainy defect marred the subsequent conversion of direct acidified curd into chakka and shrikhand. Attempts to remove this defect by manipulating the rates of cold acidification and warming up time for curd settings, and levels of ionic calcium through heat treatment and salt addition provided little relief. Future attempts in solving this problem of grain formation may have to be considered in terms of a more detailed structural and physico-chemical study of the 'grains' material isolated from chakka and shrikhand in order to understand the mechanism of their formation and, hopefully, find suitable means of controlling their formation. Nevertheless, this study has demonstrated a possibility of setting curd within 4 hrs using YH culture.

Shrikhand, like several other fermented milk products, is a summer time favourite. Fresh milk being in short supply during the summer season, shrikhand manufacturers attempt to freeze chakka or shrikhand produced in flush season and store these for use in lean season. However,
considering the total milk solids levels in chakka (about 21 to 25%) and shrikhand (about 21%) freezing these products would involve storing a larger amount of water and non-perishable items such as sugar under an expensive frozen condition. An alternative to this approach of storing chakka and shrikhand was attempted in terms of using skim milk powder, unsweetened condensed skim milk and buttermilk for shrikhand making. The choice of condensed milk was also with the intent of developing a way of preparing chakka of a desired solids level, bypassing the whey drainage step. Buttermilk was chosen in our study not so much as a source of milk solids that would be available in lean season but more as a way of by-product utilisation for a remunerative market.

Experience with these sources of milk solids make buttermilk the least promising candidate for shrikhand making. Although the weaker coagulum formation and slower syneresis could be overcome by skim milk powder fortification, the flavour quality of buttermilk shrikhand and its shelf-life was the least desirable of all, which may possibly be due to the presence of phospholipids in buttermilk. However, in this study no flavouring such as cardamom, kesar or fruits was used. Therefore, it is not certain if the slight flavour criticism of the buttermilk shrikhand could have been masked by these flavouring agents. It is also necessary to establish in future studies the possibilities of using buttermilk as a partial replacement of milk solids rather than its sole source for preparing shrikhand.
Using unsweetened condensed milk at a high TS level of 30 to 43% created difficulties in terms of insufficient development of acidity that created problems in incorporating sufficient sugar, which in turn affected the body and texture of shrikhand. In the absence of the whey drainage step, a slightly saltish taste was also detectable in high solids condensed milk shrikhand. Although membrane concentrating of milk could provide potential relief from the salty defect, inability of the starter culture to develop sufficient level of acidity appears to be the limiting factor for using condensed milk at a higher TS level.

At the level of 10 to 13% TS, shrikhand prepared from condensed milk and skim milk powder were comparable to the fresh skim milk system in terms of curd setting properties, sensory quality and shelf-life. Thus, this study has established the feasibility of using skim milk powder as an effective alternative to skim milk in the lean season. Use of condensed skim milk by diluting to about 10% TS level offered no particular advantage in terms of overall technology. However, long distance transport of liquid milk as refrigerated and concentrated products has been considered as an economically preferred alternative under certain circumstances (Bhanumurthy et al., 1972). The present study establishes the feasibility of using such product for shrikhand making after a dilution to 10% TS level. Heat sterilized evaporated milk or UHT sterilized concentrated milks were not available for our experimentation. As these
products undergo different types of heat treatments, it is difficult to anticipate their useability for shrikhand on the basis of the experiments with unsweetened condensed skim milk.

Experiments on the standardization of skim milk shrikhand in terms of the gross composition indicated varying roles played by moisture, fat, MSNF and sugar on the consistency of shrikhand. MSNF appeared to be the most important determinant of the curd strength of shrikhand. The firmness of the shrikhand was also influenced by levels of moisture. Data on curd tension, coming up time and penetrometer reading indicated a somewhat lesser role played by the sugar and fat levels within the ranges studied. The most acceptable gross composition of shrikhand, i.e. 40% moisture, 6% fat, and 41% sugar is within the acceptable limits of ISI standards.

Storage of the various shrikhand samples at the 10°C refrigerator temperature indicated a normal shelf-life of 5 weeks or more. However, buttermilk shrikhand samples kept under similar conditions were good for only 4 weeks. The spoilage of these high acid and high sugar level products was typically due to the YM activity. Minimizing the initial levels of YM and their subsequent activities have been achieved through suitable heat treatments such as in stirred yoghurt (Bake, 1971; Lang and Lang, 1973; Obert et al., 1978) and by incorporating food grade YM inhibiting agents such as sorbic acids and its salts. The heat treatment of acidified product, however, also causes damage to some of the useful
products that are elaborated in the cultured milk systems during lactic fermentation (Shahani and Chandan, 1979). For this reason, addition of antimycotic agents was preferred to heat treatment in this study. As the results indicated, potassium sorbate extended the shelf-life or refrigerated shrikhand to about 7 weeks. With the exception of butter milk shrikhand, storageability exhibited with or without potassium sorbate was essentially similar for shrikhand prepared from different milk solids.

On the basis of this investigation, it is possible to recommend the following set of process alterations that would be potentially beneficial to the shrikhand making industry:

(a) Curd setting time of 4 hrs can be achieved by using YLI culture at 2% level and 42°C.

(b) Skimmilk powder can be used by reconstitution at 10% TS level as an effective alternative to fresh skimmilk for shrikhand making.

(c) Considerable extension of shelf-life is possible by incorporating 0.05% potassium sorbate in shrikhand.
CHAPTER 6

SUMMARY AND CONCLUSIONS
6. SUMMARY AND CONCLUSIONS

6.1 The conventional shrikhand making technology requires a relatively long time because of slow setting of milk through lactic fermentation. Moreover, this product being a summer speciality, its manufacture from fresh milk in the summer implies diversion of milk from fluid consumption during the lean season. Also, shelf-life of the product is limited. Hence, a study on making certain process alterations in shrikhand technology was carried out in five different parts, viz. (i) standardization of technology of shrikhand making using buffalo skimmilk, (ii) direct acidification of buffalo skimmilk to obtain curd, (iii) use of alternative sources of milk solids for shrikhand making, (iv) observing organoleptic, chemical and microbiological changes during storage of shrikhand at refrigerator temperature (10°C) and at 30°C, and (v) use of hot air treated sugar and potassium sorbate to enhance the keeping quality of shrikhand.

6.2 Various cultures of lactic acid bacteria consisting of streptococci and lactobacilli were examined with respect to time required for curd formation and properties of the product. Yoghurt culture (YH) was the most desirable as it produced satisfactory curd within 4 hrs, thus reducing the total processing time from 8 to 12 hrs to 4 hrs for curd preparation.
6.3 A standard method was established for preparing control shrikhand samples. The method involved using buffa-lo skimmilk (10% TS), subjected to heat treatment of 85°C/30 min, followed by 1.5 to 2.0% inoculation of an active YH culture at 42°C for obtaining a curd (0.9% LA) at the end of 4 hrs. The chakka obtained from this curd was then blended with required quantity of 80% fat pasteurized (85°C/10 min) plastic cream and sugar to represent a final composition of 40% moisture, 6.0% fat and 41% sugar.

6.4 The levels of moisture, fat and sugar were found to influence the consistency measured in terms of curd tension, coming up time and penetration value. There was a general reduction in the so called curd tension values of shrikhand samples along with an increase in the level of moisture, fat and sugar. A reduction in the coming up time values due to increasing level of moisture, fat, sugar and temperature was exhibited.

Increasing penetration values were associated with an increasing level of moisture, fat and sugar. Studies with curd tension apparatus and penetrometer confirmed the major role of the protein system towards the consistency building properties of the shrikhand. The effect of increasing the non-protein constituents of the shrikhand solids, such as fat and sugar were similar to the reduction of the total solids levels of shrikhand due to the increasing levels of moisture in terms of the weakening of shrikhand consistency.
6.5 Although it was possible to reduce the curd setting time considerably by an appropriate selection of starter culture and incubation temperature, application of curd forming by direct acidification was attempted to further reduce the curd setting time from 4 hrs to 2 hrs by employing glucono-D-lactone, organic and inorganic acids.

Shrikhand prepared from the curd derived by mixing GDL at 2.0 and 2.5% levels at 30° or 40°C (corresponding to a pH 4.8 and 4.6 after 150 and 90 min of hydrolysis time, respectively), was assessed for its texture characteristics. All the samples were considered unsatisfactory on account of a perceptible graininess defect.

Three different food grade acids were studied, viz. lactic, hydrochloric and citric acids in terms of (i) curd setting temperature and pH, (ii) effect on curd tension, and (iii) effect on chakka texture. Lactic acid as an acidulant (1:9 dilution) in milk at 30°C, 4.8 coagulation pH, and 35°C curd setting temperature appeared to give the most satisfactory curd. However, all the samples were criticized for graininess. Hence, further attempts on the improvement of curd forming properties were carried out in terms of (i) various heating temperatures of milk, (ii) type of skim milk, (iii) addition of sodium chloride, (iv) slow addition of acid, and (v) slow tempering of acidified milk. However, there was no perceptible improvement in body and texture of shrikhand. Hence, direct acidification of milk was considered unsuitable for shrikhand making.
6.6 Different types of milk solids, such as plain condensed skimmilk, skimmilk powder (SMF) and buttermilk were compared with fresh skimmilk with respect to acid development by VH culture, characteristics of the resulting curd, and recovery of solids and proteins. The acid development in condensed skimmilk, particularly above 35% total solids was much less than that in other systems. Fresh skimmilk showed slightly better acid development than buttermilk with or without added SMF, reconstituted skimmilk and diluted condensed skimmilk.

Solids and protein recovery appeared to be directly proportional to curd firmness. Fresh skimmilk showing maximum curd strength resulted in the highest recovery of solids as well as protein in chakka followed by dilute condensed milk, reconstituted milk and buttermilk system. Flavour wise there was no perceptible difference between the curds obtained from fresh skimmilk, dilute condensed skimmilk and reconstituted skimmilk, although the buttermilk curd tasted somewhat 'unnatural' or 'abnormal'.

The condensed milk curd was, however, definitely saltish, primarily because of the absence of whey drainage and the saltiness persisted in shrikhand prepared therefrom. Also the condensed milk product was not sufficient acidic in taste. Hence, condensed milk was considered unsuitable for the preparation of curd intended for shrikhand making. The texture of the shrikhand from the reconstituted skimmilk was smoother than that from other milk systems. On the
basis of these findings, fluid skimmilk can be success-
fully replaced by reconstituted skimmilk.

6.7 Data on organoleptic evaluation, changes in acidity, pH, soluble nitrogen, TVFA and microflora, particularly in terms of lactic, YM and proteolytic counts indicated an average shelf-life of these shririkand samples, at 10°C storage, was within the range of 28 to 35 days. Most samp-
les were within the range of 30 to 35 days. However, the buttermilk shririkand samples had a shelf-life of about 28 days.

Deterioration in the keeping quality was usually associated with an increased activity of YM and proteolytic microflora under refrigerated (10°C) conditions of storage. Increasing YM activity was associated with a reduction of the sucrose level and an increase in the levels of reducing sugar, titratable acidity, TVFA and soluble nitrogen. The spoiled samples exhibited characteristic fruity fermented and rancid, aromatic traits, along with surface discoloura-
tion due to mould and yeast growth.

6.8 In order to minimize the yeast and mould contamina-
tion in shririkhand through sugar, the latter was heat treated under different time-temperature conditions. A temperature of 140°C for 20 min was found to reduce the YM count to zero. From the data on organoleptic evaluation, changes in pH and acidity and YM counts, there was no advantage of using hot-air-treated sugar for the purpose of increasing the keeping quality of shririkhand.
6.9 Potassium sorbate added to shrikhand samples retarded the changes in soluble nitrogen, reducing sugar, total volatile fatty acids (TVFA), acidity and pH as well as bacteriological and sensory quality of the product. At 0.01, 0.025, 0.05 and 0.1% levels of potassium sorbate additions, products kept well for 37, 44, 53 and 55 days, respectively. Thus, 0.05% and 0.1% levels were substantially more effective than lower levels. There was no appreciable difference between these two levels and hence addition of 0.05% ps was considered most desirable.

6.10 From the sensory evaluation, chemical parameters and microbiological analysis of the samples, ps definitely enhanced the keeping quality of shrikhand. The changes in the sensory score ran parallel to chemical changes in the product. The moisture content, sucrose, pH and lactic count decreased with the storage period, while soluble nitrogen, reducing sugar, TVFA, acidity, YM counts and proteolytic counts increased. The increase being slower in samples with ps as compared to control at 10°C.

The chemical, sensory and bacteriological status of shrikhand changed during storage in a more or less similar fashion as in the case of reconstituted milks, buttermilk and dilute condensed milk used as the source of MSNF. The storage life of shrikhand was observed to be 36, 43, 46 and 49 days in buttermilk, dilute condensed milk, reconstituted milk and skimmilk shrikhands.

6.11 Flavour score declined faster at 30°C than that at
10°C. The keeping quality of shrikhand was 3 and 4 days in controls and ps treated samples, respectively. There was decrease in pH and sucrose, while there was an increase in acidity, soluble nitrogen, reducing sugar, TVFA, proteolytic counts and YM the increase being faster in shrikhand samples without ps as compared to ps treated shrikhand.

6.12 It was thus concluded from the present investigation that the use of a yoghurt type (YM) culture for the preparation of curd intended for shrikhand making could considerably reduce the total processing time without affecting the quality of the product.

Reconstituted skim milk could successfully replace skim milk for shrikhand making.

Potassium sorbate at 0.05% level substantially enhanced the storage stability of shrikhand.
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## Appendix I

**Effect of types of starter culture on the rate of acid development in buffalo skim milk**

<table>
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<th>Culture</th>
<th>pH</th>
<th>Acidity</th>
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<tr>
<td><strong>pH/ Acidity</strong></td>
<td>0.0</td>
<td>1.0</td>
</tr>
<tr>
<td>LP-40</td>
<td>6.60</td>
<td>6.50</td>
</tr>
<tr>
<td>1.0%, 36°C</td>
<td>0.126</td>
<td>0.135</td>
</tr>
<tr>
<td>NDRI1</td>
<td>6.60</td>
<td>6.40</td>
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<tr>
<td>1.0%, 37°C</td>
<td>0.126</td>
<td>0.130</td>
</tr>
<tr>
<td>NDRI2</td>
<td>6.60</td>
<td>6.30</td>
</tr>
<tr>
<td>1.0%, 37°C</td>
<td>0.126</td>
<td>0.170</td>
</tr>
<tr>
<td>DRC1</td>
<td>6.60</td>
<td>6.40</td>
</tr>
<tr>
<td>1.0%, 36°C</td>
<td>0.126</td>
<td>0.171</td>
</tr>
<tr>
<td>YH</td>
<td>6.60</td>
<td>5.90</td>
</tr>
<tr>
<td>1.0%, 42°C</td>
<td>0.126</td>
<td>0.149</td>
</tr>
<tr>
<td>Acidophilus</td>
<td>6.60</td>
<td>6.40</td>
</tr>
<tr>
<td>1.0%, 37°C</td>
<td>0.126</td>
<td>0.171</td>
</tr>
<tr>
<td>B</td>
<td>6.60</td>
<td>6.40</td>
</tr>
<tr>
<td>1.0%, 37°C</td>
<td>0.126</td>
<td>0.170</td>
</tr>
<tr>
<td>Mixed</td>
<td>6.60</td>
<td>6.40</td>
</tr>
<tr>
<td>1.0%, 37°C</td>
<td>0.126</td>
<td>0.170</td>
</tr>
<tr>
<td>S + L</td>
<td>6.60</td>
<td>6.30</td>
</tr>
<tr>
<td>1.0%, 37°C</td>
<td>0.126</td>
<td>0.144</td>
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### Appendix II

Compositional information of various shrikhand formulations

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<th>Sugar variation</th>
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<td></td>
<td>5.0</td>
<td>6.0</td>
<td>7.0</td>
<td>38.5</td>
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<tr>
<td><strong>Quantity of ingredients</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cream (g)</td>
<td>7.14</td>
<td>8.59</td>
<td>10.00</td>
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<tr>
<td>Chakka (g)</td>
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<td>49.27</td>
<td>47.83</td>
<td>50.34</td>
</tr>
<tr>
<td>Sugar (g)</td>
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<td>41.44</td>
<td>40.48</td>
<td>42.04</td>
</tr>
<tr>
<td>Whey (g)</td>
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<td>0.70</td>
<td>1.69</td>
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<td><strong>Composition of shrikhand</strong></td>
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<td>Sugar %</td>
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<td>41.44</td>
<td>40.48</td>
<td>42.04</td>
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<tr>
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<td>6.00</td>
<td>7.00</td>
<td>6.14</td>
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<td>40.42</td>
<td>39.44</td>
<td>39.65</td>
<td>38.76</td>
</tr>
<tr>
<td>Fat/SNF</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.462</td>
</tr>
<tr>
<td>Sugar/SNF</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>3.161</td>
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<tr>
<td>Fat/Moisture</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
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<tr>
<td>SNF/Moist.</td>
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<td>0.332</td>
<td>0.324</td>
<td>-</td>
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<tr>
<td>Sugar/Moist.</td>
<td>1.025</td>
<td>1.050</td>
<td>1.021</td>
<td>-</td>
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* Cream having 26.77% moisture, 70% fat and 3.23% SNF
Chakka having 74.0% moisture and 26.0% SNF
Sugar having 100% total solids
Whey having 94% moisture and 6.0% SNF
# APPENDIX III

Effect of fat, moisture, sugar and temperature on the curd tension(g) of shrikhand

<table>
<thead>
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<th>Fat level (%)</th>
<th>Temperature 0°C</th>
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<td></td>
<td>15</td>
<td>30</td>
<td>40</td>
</tr>
<tr>
<td>5</td>
<td>52.86 (51.50-53.80)</td>
<td>42.20 (40.30-43.80)</td>
<td>34.40 (33.30-35.80)</td>
</tr>
<tr>
<td>6</td>
<td>50.26 (49.30-51.30)</td>
<td>39.40 (38.60-40.50)</td>
<td>28.80 (28.50-29.10)</td>
</tr>
<tr>
<td>7</td>
<td>48.80 (46.90-51.10)</td>
<td>35.23 (34.20-36.20)</td>
<td>26.06 (25.70-26.30)</td>
</tr>
<tr>
<td>38.5</td>
<td>57.36 (56.30-58.50)</td>
<td>46.60 (45.90-47.80)</td>
<td>33.60 (32.20-34.80)</td>
</tr>
<tr>
<td>40.0</td>
<td>50.28 (49.50-51.13)</td>
<td>40.90 (39.30-42.30)</td>
<td>28.00 (26.90-28.60)</td>
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<tr>
<td>42.0</td>
<td>46.76 (46.20-47.30)</td>
<td>37.66 (37.30-38.20)</td>
<td>26.59 (25.30-28.30)</td>
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<tr>
<td>39.0</td>
<td>55.33 (53.80-55.40)</td>
<td>40.73 (30.20-41.20)</td>
<td>33.06 (32.50-33.80)</td>
</tr>
<tr>
<td>41.0</td>
<td>48.70 (47.30-50.30)</td>
<td>38.10 (37.30-38.90)</td>
<td>28.03 (26.70-28.90)</td>
</tr>
<tr>
<td>43.0</td>
<td>44.40 (43.90-44.80)</td>
<td>33.80 (33.20-34.30)</td>
<td>26.23 (25.10-26.90)</td>
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### APPENDIX IV

Effect of fat, moisture, sugar and temperature on the coming up time (sec) of shrikhand

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<th>40</th>
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<td></td>
<td>(15-20)</td>
<td>(16-22)</td>
<td>(15-17)</td>
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<td>18.66</td>
<td>14.33</td>
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<td>(75.0-79.0)</td>
<td>(18-20)</td>
<td>(14-15)</td>
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<tr>
<td>6</td>
<td>76.33</td>
<td>16.33</td>
<td>13.86</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(75-78)</td>
<td>(16-17)</td>
<td>(13.5-14.10)</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>71.00</td>
<td>14.83</td>
<td>13.33</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(70-72)</td>
<td>(14.5-15.00)</td>
<td>(13-14)</td>
<td></td>
</tr>
<tr>
<td>Moisture levels (%)</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>38.5</td>
<td>83.00</td>
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<td>16.33</td>
<td></td>
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<tr>
<td></td>
<td>(81-85)</td>
<td>(21-22)</td>
<td>(16-17)</td>
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<tr>
<td>40.0</td>
<td>75.33</td>
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<td>13.66</td>
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<td>(17-18)</td>
<td>(13-14)</td>
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<td>71.00</td>
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<td>8.66</td>
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<td></td>
<td>(70-72)</td>
<td>(15-16)</td>
<td>(8-10)</td>
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<td></td>
<td>(85-90)</td>
<td>(20-21)</td>
<td>(16-17)</td>
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</tr>
<tr>
<td>Sugar levels (%)</td>
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<td></td>
</tr>
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<td>75.66</td>
<td>16.83</td>
<td>13.66</td>
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</tr>
<tr>
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<td>(75-76)</td>
<td>(16-17)</td>
<td>(13-14)</td>
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<tr>
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<td>71.33</td>
<td>15.33</td>
<td>11.00</td>
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<td>(70-73)</td>
<td>(15-16)</td>
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**APPENDIX V**

Effect of fat, moisture, sugar and temperature on the penetration value (0.1 mm) of shrikhand

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<th></th>
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<td>15</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>194</td>
<td>207.66</td>
<td>231.33</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(194-195)</td>
<td>(205-210)</td>
<td>(230-233)</td>
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</tr>
<tr>
<td>6</td>
<td>195</td>
<td>210</td>
<td>234.33</td>
<td></td>
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<tr>
<td></td>
<td>(193-198)</td>
<td>(209-211)</td>
<td>(234-235)</td>
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</tr>
<tr>
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<td>211.30</td>
<td>235.30</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(196-198)</td>
<td>(210-213)</td>
<td>(234-236)</td>
<td></td>
</tr>
<tr>
<td>38.5</td>
<td>190.66</td>
<td>200.66</td>
<td>221.66</td>
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<tr>
<td>Moisture levels (%)</td>
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<td></td>
<td></td>
</tr>
<tr>
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<td>199.33</td>
<td>209.66</td>
<td>234.33</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(199-200)</td>
<td>(209-210)</td>
<td>(233-235)</td>
<td></td>
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<tr>
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<td>228.00</td>
<td>254.66</td>
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</tr>
<tr>
<td></td>
<td>(215-219)</td>
<td>(227-229)</td>
<td>(253-256)</td>
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<td>226.33</td>
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<td>Sugar levels (%)</td>
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</tr>
<tr>
<td>41.0</td>
<td>198.00</td>
<td>211.66</td>
<td>235.66</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(197-199)</td>
<td>(211-213)</td>
<td>(235-237)</td>
<td></td>
</tr>
<tr>
<td>43.0</td>
<td>203.66</td>
<td>216.33</td>
<td>241.66</td>
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</tr>
<tr>
<td></td>
<td>(203-205)</td>
<td>(215-217)</td>
<td>(241-243)</td>
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## Effect of Different Fat, Moisture and Sugar Levels on the Sensory Score of Shrikhand

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<tr>
<th>Particulars</th>
<th>Appearance</th>
<th>Body and Texture</th>
</tr>
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<tbody>
<tr>
<td>Shrikhand with 5% fat</td>
<td>3.9-4.0</td>
<td>(7.63-7.67)</td>
</tr>
<tr>
<td>Shrikhand with 6% fat</td>
<td>4.0-4.1</td>
<td>(7.62-7.70)</td>
</tr>
<tr>
<td>Shrikhand with 7% fat</td>
<td>4.0-4.1</td>
<td>(7.83-7.91)</td>
</tr>
<tr>
<td>Shrikhand having 38.5% moisture</td>
<td>3.9-4.0</td>
<td>(7.58-7.64)</td>
</tr>
<tr>
<td>Shrikhand having 40% moisture</td>
<td>3.9-4.0</td>
<td>(7.98-8.00)</td>
</tr>
<tr>
<td>Shrikhand having 42% moisture</td>
<td>3.9-4.0</td>
<td>(7.60-7.62)</td>
</tr>
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<td>Shrikhand having 39% sugar</td>
<td>4.0-4.1</td>
<td>(7.66-7.68)</td>
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<tr>
<td>Shrikhand having 41% sugar</td>
<td>4.0-4.1</td>
<td>(7.81-7.85)</td>
</tr>
<tr>
<td>Shrikhand having 43% sugar</td>
<td>4.0-4.1</td>
<td>(7.63-7.67)</td>
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APPENDIX VII

Effect of concentration of GDL and temperature on the rate of hydrolysis of GDL in buffalo skim milk

<table>
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<tr>
<th>Time (min)</th>
<th>Concentration of GDL</th>
<th>Water bath temp. 40°C</th>
<th>Water bath temp. 30°C</th>
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<td>0.8%</td>
<td>1.0%</td>
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<tr>
<td>30</td>
<td>5.70</td>
<td>5.55</td>
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</tr>
<tr>
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<td>5.60</td>
<td>5.40</td>
<td>5.30</td>
</tr>
<tr>
<td>90</td>
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</tr>
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<td>120</td>
<td>5.50</td>
<td>5.20</td>
<td>5.00</td>
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<td>150</td>
<td>5.40</td>
<td>5.15</td>
<td>5.00</td>
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<td>180</td>
<td>5.40</td>
<td>5.15</td>
<td>5.00</td>
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<td>210</td>
<td>5.40</td>
<td>5.15</td>
<td>5.00</td>
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</table>

contd......
contd...... APPENDIX VII

<table>
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<th>Time (sec)</th>
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<td>6.40</td>
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<td>60</td>
<td>6.10</td>
</tr>
<tr>
<td>90</td>
<td>5.90</td>
</tr>
<tr>
<td>120</td>
<td>5.80</td>
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<td>150</td>
<td>5.80</td>
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<td>180</td>
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<tr>
<td>210</td>
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<td>300</td>
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* 10% TS level; heat treatment of milk (85°C/20 min)
## APPENDIX VIII(a)

Changes in sensory quality of shrikhand obtained from different MSNF systems

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<th>Storage period (days)</th>
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<th>Reconstituted milk</th>
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<td>Appearance</td>
<td>Body &amp; texture</td>
<td>Flavour</td>
<td>Colour</td>
<td>Appearance</td>
<td>Body &amp; texture</td>
<td>Flavour</td>
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<td>(25.01-</td>
<td>(51.10-</td>
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<td>25.90)</td>
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<td>(25.08-</td>
<td>(44.11-</td>
<td>(3.95-</td>
<td>(7.13-</td>
<td>(25.03-</td>
<td>(44.03-</td>
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<td>(6.98-</td>
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<td>(35.48-</td>
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<td>7.33)</td>
<td>23.30)</td>
<td>37.72)</td>
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<td>7.19)</td>
<td>24.98)</td>
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APPENDIX VIII(b)

Changes in sensory quality of shrikhands obtained from different MSNF systems

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<tr>
<th>Storage period (days)</th>
<th>Buttermilk</th>
<th>Diluted condensed milk</th>
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<td>Appearance</td>
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<td>(7.39-</td>
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<td>7.51</td>
</tr>
<tr>
<td></td>
<td>(4.23)</td>
<td>7.63</td>
</tr>
<tr>
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<td>4.11</td>
<td>7.44</td>
</tr>
<tr>
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<td>(4.03-</td>
<td>(7.35-</td>
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<tr>
<td></td>
<td>4.21)</td>
<td>7.61</td>
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<td>4.12</td>
<td>7.11</td>
</tr>
<tr>
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<td>(6.97-</td>
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