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Title: Some Microbiological and Sensory Characteristics of Cheddar Cheese Manufactured from Conventional and Ultrafiltered-Concentrated milk.

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Cheddar cheese making from ultrafiltered (UF) concentrated milk potentially increases yield, saves raw materials, cuts transportation costs, minimizes waste pollution, and stream-lines production. However, the resulting UF-cheese has been found to differ from conventionally made cheese (CM-cheese) in its casein matrix and ripening process.

UF and CM Cheddar cheeses were compared in regard to their overall bacterial numbers, non-starter lactic acid bacterial population, potentially pathogenic bacteria, flavor, body and textural characteristics. Standard microbiological sampling procedures were followed. Two plating methods were used and concurrently compared; the manual spread plate, and the automated spiral plate.

No significant differences were noted in the over-all bacterial numbers of the experimental cheeses. The non-starter lactic acid bacteria from both cheese types showed comparable carbohydrate fermentation patterns and similar metabolic activities in milk. No significant counts of potential pathogenic bacteria were detected in the UF cheese. The textural scores showed UF cheese to have a slightly higher quality than that of the CM cheese, but there was no difference in the flavor scores .

Results obtained lead to the conclusion that the UF and CM Cheddar cheeses used in this study appear not to differ in overall quality. The spiral plating method, as used in this study, was found to give comparable counts and it can potentially save time and costs when the media preparation and counting techniques are done properly.

Some Microbiological and Sensory Characteristics of Cheddar Cheese Manufactured from Conventional and Ultrafiltered-Concentrated milk.

by

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INTRODUCTION

It has been estimated that 150 countries in the world manufacture cheese. Made in numerous ways, these cheeses result from traditional, labor-intensive, low yielding practices (1). Depending on the variety, for example, it takes from 8-10 pounds of milk to make 1 pound of cheese (2). Unaware of manufacturing inefficiencies however, consumers worldwide have made cheese a significant part of their diets; more than 26 billion pounds of cheese are produced annually in the world (1). The United States alone consumes over 5 billion pounds of cheese, 3 billion pounds of one variety alone, Cheddar (3). An increase in cheese consumption in recent years has been reported. The U.S. Department of Agriculture reports the per capita consumption of hard cheese for 1987 as 23.2 pounds, more than double the 9.6 pounds per year in 1965 (4).

Despite its worldwide importance, cheesemaking has been, as previously mentioned, a traditional, batch process of multiple steps and low yields (5) ;however, in 1969, a major advance took place in the development of a new cheese makingmethod, which was expected to offer the potential of increased yields,

greater throughput, more continuity in processing and more flexibility in disposal ofunwanted milk components (6).

Three French scientists, Maubois, Mocquot and Vassal fractionated and concentrated milk into its components. The separation was done on the basis of molecular size, using high pressure and small-pore diameter membranes (6). This separation process, which became known as ultrafiltration (UF), is described as a pressure-driven, membrane filtration process which allows lactose, non-protein nitrogen fractions and soluble salts to freely pass through the membrane. These components, along with water, form the *permeate*, usually fed to livestock at the farm itself. The casein, fat, lactoalbumin and lactoglobulin, on the other hand, concentrate in a stream flow, refered to as the *retentate,* which is taken to the creamery and made into cheese (7, 8).

Maubois et al. used the highly concentrated milk solids, or pre-cheese, to succesfully make a specialty soft cheese with no whey drainage, in a more continuous manner and with significantly higher yields than the conventional method allowed. This new cheesemaking method was named MMV, after the scientists' names (6). Use of this technology in the dairy industry brought numerous advantages; the most significant one of economical importance was an increase in product yield. This greater yield, along with a higher nutritional quality, resulted from an increased retention (up to 18-20%) of the whey proteins , usually lost in traditional cheese-making (9,10). Reduced losses of insoluble casein, fat and fines also contributed to the yield increase (11).

Transportation costs were reported cut by one half when UF -concentrate was taken from the farm to the creamery instead of the whole milk (87% water) (12). Other advantages mentioned included a higher capacity of existing cheese vessels, smaller ammounts of rennet used, less pollution (BOD of permeate is reduced about 20%) and the possibility of continuous cheese production (13).

The use of UF in the dairy industry grew rapidly and became firmly established for soft and feta cheese production; but its use in the production of high solids cheese ,such as Cheddar, proved to be a challenge (14). Initial Cheddar cheesemaking trials gave poor results. Milk concentrated higher than two-fold resulted in fat losses, abnormal composition, flavor and texture(15). The concentration step was thought to cause a reduction in fat globule size and denaturation of whey proteins (16).

Adaptations of the traditional method and changes in the new MMV process eventualy made Cheddar cheese production from UF-milk possible. The Cheddar cheese obtained from ultrafiltered concentrated milk had a slightly different composition when compared with that of Cheddar chees made from conventional milk (1). Whether or not the UF-cheese would have a normal microbial flora and whether or not it would ripen normally became important questions.

Determination of possible major quality differences between conventionlly-made and cheese made from UF-concentrated milk became the goal of numerous studies. Chemical and nutritional composition, texture and flavor and microbiological safety of the cheese were extensively studied.

The objective of this study was also a partial determination of the quality of Cheddar cheese made from milk concentrated by ultrafiltration. A comparision of Cheddar cheese made from conventional cheese-milk and cheese made from milk concentrated 1.65-1.90 fold was done. The objectives included some microbiological observations of the lactic acid bacteria and possible pathogens, a limited sensory analysis and a partial characterization of lactic isolates from both cheese types. Standard methodology was used. No evidence of significant differences in the microbiology between cheese types was found. Furthermore, there was no meaningful difference in the average flavor. However, a slight difference in texture was detected; UF-milk cheese had lower average scores, i.e. superior texture.

LITERATURE REVIEW

ULTRAFILTRATION AND ADAPTATIONS FOR ITS USE IN THE DAIRY INDUSTRY

Ultrafiltration in cheesemaking was first successfuly used in France in 1969. The MMV method, so named after Mabouis, Mouquot, and Vassal, was originally used for production of Brie and Camembert cheeses. The "pre-cheese", obtained by concentrating milk with addition of cream, had the same approximate composition as the final cheese. Addition of starter, rennet and salt was followed by pouring and setting the mixture in molds with little or no whey drainage. The concentration factor required was about five-fold (6).

In 1974 Maubois and Mocquot reported on the successful application of their MMV process to the manufacture of fresh soft cheese, soft cheese of the camembert type, and cheese made from goat's milk. They saw the principal advantage of the process as the increase in cheese yield.

All the milk proteins, whether casein or soluble proteins, remained in the retentate and finally in the cheese. Less rennet (80%) was used, and the whey contained no proteins (80% less polluting power) (17). By 1981, 120,000 tons of

cheese, mainly feta cheese, had been produced using the ultrafiltration technology (18). Difficulties in reaching the appropriate concentration, however, imposed limits to the application of MMV in the manufacture of other higher solid cheeses. For instance, to reach the solids level equivalent to that of Cheddar cheese would require a nine-fold concentration, which was practically not feasible (14). Adaptations of cheese making by the ultrafiltration technology were then developed. Unlike the MMV method, which required highly concentrated retentates (up to 10:1) or precheese and non-traditional manufacturing steps, the LCR method, Low Concentrated Retentate, required a 2:1 maximum volume concentration and used traditional cheesemaking equipment. Chapman et al. in 1974 were the first to report on the manufacture of Cheddar, Cheshire, soft cheese and yoghurt using LCR. However, no increase in yield was achieved (19).

Another modification to the MMV principle consisted of a 3-6 fold concentration, adjustment of the milk to the composition of the cheese to be made, followed by modified cheesemaking with some whey drainage (14).

APPLICATIONS OF UF TECHNOLOGY IN THE DAIRY INDUSTRY

Numerous dairy products have been made using the modified processes of ultrafiltration technology, in general, with lower concentration factors. Low concentrated retentate (LCR) cottage cheese making on a laboratory scale was reported early in 1976 by Matthews et al. . They used a 2:1 concentrated

retentate (20). Later Kosikowski (21) and Atar et al. (22) made cottage cheese using UF retentates from 1:1 to 1:2 total protein. More recently, Kosikowski et al. (23) concluded that optimum qualities in cottage cheese were attained from retentates concentrated to 1.7:1 total protein . Kealey and Kosikowski (24) wrote on the optimization of cottage cheese manufacture from UF retentates, and demonstrated the possibility of production of highly concentrated skim milk retentate for shipment to cheese plants (25).

Cream cheese was also made early on via UF. Covacevich and Kosikowsi (26) used a maximum of 27.6% solids concentration to obtain a cream cheese of excellent shelf life and smoothness. Mozarella cheese adapted easily to the ultrafiltration concentration step. Non-traditional cheesemaking methods were used by several authors: Kovacevich and Kosikowski (27,28) Maubois and Kosikowski (29), Covacevich (30), Friis (31), and Fernandez and Kosikowsi (32). Constant improvements in the process and the product were made. Commercial production of mozzarella via UF process is now common in the United States. The product is said to be the same as mozzarella from conventional methods in its physical and chemical qualities (33).

Semihard cheese varieties also have been produced with UF-milk retentates and non-traditional cheesemaking methods. DeBoer and Nooy produced a full fat semihard cheese with composition nearly identical to that of Gouda cheese (34). Bush et al. (35) described Brick and Colby cheeses manufactured from milk concentrated by UF; the rennet level used was 45% less than in control cheeses.

Also, Emmental cheese made from milk concentrated by a factor of 1.2-1.7 was reported by Rosseaux et al. (36).

Other dairy products besides cheese have been manufactured making use of UF technology. Chapman et al. made excellent yoghurt directly from milk concentrated 2-fold by UF, without fortification with milk powder (19). To the benefit of those allergic to cow's milk, the use of UF has improved the quality of yoghurt made from goat's milk, as reported by Marshall and El-Bagoury (37).

The Danish specialty product Ymer has also been succesfully produced from ultrafiltered skim-milk concentrated 2-fold. Rubin and Werner reported higher yields of Ymer due to increased retention of the soluble proteins (38). Increased yields were also reported by Puhan and Gallmann (10) in the manufacture of Quark. Nutritionally the product became more valuable because ultrafiltration allowed the ratio of casein to whey to be the same as that of milk (4:1). Conventionally made Quark has a ratio of 16:1 (10).

Striving towards the production of higher solids cheese, such as Cheddar via UF methods, Ernstrom et al. (39) made a cheese-like product called cheese-base. This product was thought to potentially replace the immature natural cheese component of processed cheese blends. Improvements on the Ernstrom method were made by Madsen and Bjerre (40) who made the process practically applicable for the dairy industry.

Nineteen percent more cheese-base was produced than Cheddar cheese , from the same amount of milk. Kumar and Kosikowski (41) suggested the manufacture of a process cheese from ultrafiltered, freeze-dried, skim-milk retentates with addition of plastic cream.

A number of retentate powder products, milk powder produced by drying skim milk retentate, and their practical applications were investigated by Madsen and Bjerre (42). Retentate powder containing less lactose than common skim milk powder, was expected to be used as "cheese powder". Reduction in transport costs, waste water and elimination of problems connected with seasonal changes are some benefits of using retentate powder. Retentate powder without lactose and with maltose additive were also produced. Applications of ultrafiltration in the dairy industry are numerous and varied. The removal of penicillin G for example has been studied. Concentrations from 0.05 to .20 IU/ml of this antibiotic were removed to undetectable levels by a combination of ultrafiltrations and permeate washes(43)

CHEDDAR CHEESE BY ULTRAFILTRATION

The Difficulties

Cheddar cheese production using UF has represented a challenge. Many difficulties had to be overcome; first, there was the intrinsic difficulty of manipulating the high solids content cheese, using the MMV principle without the release of any whey. As a solution to this problem the LCR principle was developed

which required lower concentration factors and modification of the traditional cheesemaking procedure (1). In concentrating the protein and fat by ultrafiltration, the retentate acquired a higher ratio of protein to dry matter, so its properties differed greatly from those of the milk from which it was prepared.

It was determined, for example, that the clotting time increased as the protein content increased (44); but once renneted, the coagulates were so firm that several men were needed to pull the knife through the coagulum. Calcium, concentrated in the retentate was thought responsible for this abnormality (45). Three solutions were adopted: acidifying the milk before ultrafiltration, diafiltering the concentrate, and adding sodium chloride during ultrafiltration to displace the calcium (14).

Further on, it was shown that the ratio of minerals to casein required adjusting. In making Cheddar cheese traditionally, the ratio of mineral to casein (basic structure), depends only upon the extent of acid production in the vat; in making Cheddar cheese from UF-concentrated milk, however, there are four separate stages at which mineral losses were found to occur: preacidification of milk, ultrafiltration, diafiltration and acid production in the coagulum. Adjustment of minerals to casein ratio,therefore, had to be done at each of these stages (46).

One of the more serious difficulties in making Cheddar cheese from UF-milk was that concentration of the milk increased the buffering capacity of the retentate to acid (47).

This, in turn, increased cheese-making time, and the final pH value of the cheese. It was suggested that holding the curd longer in the whey, along with measures to increase moisture, such as lower scalding temperatures and coarser cutting the curd, could counteract the higher buffering capacity (48,49).

Retention of the fat was also difficult to attain. Serious losses in the whey were observed when highly concentrated milks were used. This loss was thought to be due to the inability of the curds to retain the fat effectively, which in turn was believed to be the result of a lower degree of casein micelle aggregation at the time of curd-cutting. An increase in the length of the set was found to be of help in this case (50).

It was also observed that as the concentration factor of the milk increased, the rate of casein break down, the intensity of Cheddar flavor (29), and the levels of hydrogen sulfide and methanethiol in the cheese decreased. All of these perhaps were due to a reduced concentration of active rennet retained in the curd. Increasing the amount of rennet and addition of small amounts of proteinases were tried (50)

UF-CHEDDAR CHEESE MAKING TRIALS

Chapman et al. (19) were the first to report on the manufacture of Cheddar cheese from whole milk concentrated by UF by a factor of 2. No increase in yield was achieved. However, use of only half the normal amount of rennet and lower scald temperatures were considered potential advantages.

Covacevich and Kosikowski (27) added freeze-dried retentate to fresh retentate in an effort to minimize whey production while making Cheddar cheese from retentates. Milk concentrated to the maximum protein content of Cheddar cheese (60.5% solids) was used. The result was a Cheddar cheese crumbly and corky in body, and lacking the typical cheese flavor of conventional Cheddar.

Kosikowski attempted again to make Cheddar cheese using water reconstituted milk retentates and conventional cheesemaking techniques. After 3 months of ripening the cheese had a resemblance to Gouda-Swiss type cheese, untypical of Cheddar cheese, with a pleasing, sweet, mild flavor and small eyes (51,52).

Using a series of cheese making modifications, Kealey and Kosikowski (53) made Cheddar cheese from retentate-supplemented milks. The adaptations included a reduction in rennet, 5/8" knives and a cooking temperature of 96 ^{0}F. The resulting cheese was acceptable and of normal composition. Industrial cheesemaking trials employing this supplementation and modifications were done to confirm the process. The cheese yields were high and the cheese body and flavor acceptable (54)

Sutherl and and Jameson (55) evaluated different degrees of preacidification and diafiltration of whole milk concentrated 4.8-fold by UF. The lactose and mineral levels were varied by adjustment of degree of diafiltration and milk pH. The resulting cheeses had normal fat content, slightly high moisture levels and varying pH, Ca, P and lactose. It was concluded that ultrafiltration should be carried out at pH 6.2-6.4 with sufficient diafiltration to yield a 3.3% lactose content in the retentate.

Green et al. (50) evaluated the effect of using milk concentrated 1.7-4 fold on the manufacture and ripening of Cheddar chees. They found concentration factors higher then 2-fold gave a cheese of low fat content and concentration factors above 3 gave cheese with too high a moisture content. It was also reported that as the concentration factor increased, the rate of casein break down, the intensity of Cheddar flavor and the levels of hydrogen sulfide in the cheese decreased.

Another study by Green et al. (56) found that Cheddar cheese made from milk concentrated by UF had abnormal texture due to poor curd fusion, was slightly harder, more granular and drier than control cheese. In 1985 Green reported again on the effect of UF-milk pretreatment and Cheddar cheese making conditions on the properties of the resulting cheese (57). Concentration factors of 3 to 6-fold were used along with a light homogenization before concentration, lower coagulation temperature and addition of the bacterial proteinase, neutrase. The composition of all cheeses were similar to those of control cheese.

In the same year Kosikowski et al. (58) reported on the manufacture of Cheddar cheese with retentate supplemented whole milk. Traditional cheese making techniques were used. The optimal cheese was obtained from milk supplemented with 4.5:1 retentates. Supplementation was found to increase total solids, fat, protein and ash, and also to improve flavor and texture. High concentrations ,10:1 (33% protein), and MMV concepts were also tried. A stable pH of 5.2 was attained in 6 to 12 h.

The flavor was excellent and the texture smooth with soft to medium firm bodies (59).

Once the possibility of Cheddar cheese manufacture using UF retentates and LCR or MMV principles was established on a laboratory scale, industrial trials then were done. Kealey and Kosikowski (60) determined that it was feasible to concentrate milk at the farm or collecting station and transport the retentate to a distant large industrial cheese making site. Similarly, Zall et al. carried out large scale studies in which more than 14 million pounds of milk were ultrafiltered and converted into cheese. Their studies demonstrated that it is not necessary to have special equipment to handle low concentrated retentates of UF milk at the farm or cheese plant, and that UF cuts costs for both the dairy farmer and cheesemaker (61,62,63).

LACTIC ACID STARTERS AND UF CHEDDAR CHEESE

The effect starter organisms have on the quality of the cheese previously has been established for conventional cheese. Lowrie and Lawrence for example, concluded that bitterness in cheese depends upon the influence of manufacturing conditions on particular starter strains (64). The selection of starter is equally important for retentate ripening because, as Coton concluded, not all commonly used Cheddar cheese starters perform equally well in the high concentration of protein (14).

Hickey et al. (65) studied the growth and acid production of starters in ultrafiltered milk. It was concluded that the concentration of milk caused an apparent stimulation of growth and acid production of S. cremoris and S. lactis starter organisms. This stimulation was thought to be due to the release and concentration of compounds in the milk during ultrafiltration. This may explain the higher starter activity in retentate than in milk equivalent (retentate blended with permeate). The increased acid yield was believed to be enhanced by the higher buffering capacity of the retentate. These results suggest that retentate is an improved growth medium for starter organisms .

Mistry and Kosikiwski (66) also concluded that the high total solids or ash of retentates did not affect growth of the starter organism, although overcrowding of cells was believed responsible for the longer doubling times in fast cultures. The authors also examined the lactic acid producing capacity of direct-set mesophilic starters and their influence on pH changes in UF concentrated milk. Their study showed that lactic fermentation of UF-retentes placed greater demands on starter bacteria for lactic acid production than fermentation of milk. The higher buffering capacity previously mentioned was thought responsible for the difficulty in pH reduction. A larger inoculum size was found to partially fulfill the demand for increased lactic acid (46).

Mistry and Kosikowski (66) also showed that in skim milk UF-retentates, this high buffering capacity combined with uncoupling of growth from acid production permits lactic acid bacteria to grow to higher numbers, despite continued lactose

metabolism. Turner and Thomas described uncoupling as the phenomenon of continued lactic acid production despite insignificant bacterial growth at low pH (67).

Experimenting with bulk retentate starters, the same two authors concluded, on the other hand, that the naturally built-in buffering system enabled lactic starter grown in milk retentates to maintain a steady activity for longer time than when grown in milk. A higher cell population was obtained in retentates and keeping the pH at high levels for long times minimized bacterial cell injury. The starter grown in this naturally buffered system was more active and steady for long periods (68).

Mistry and Kosikowski (69) then applied the retentate starter produced from ultrafiltered milk to the manufacture of Cheddar cheese. The retentate starter eliminated the need for milk ripening, thereby reducing the total cheese making time. They concluded that retentate bulk starters can successfully make good quality Cheddar cheese at specific inoculum levels.

Kosikowski (1) commented on the future use of various combinations of mesophilic with thermophilic starters for UF Cheddar cheese manufacture. A more extensive use of starter systems which produce large numbers of bacteria and increased use of microbial flavor enzymes are expected.

PROPERTIES OF UF RETENTATES

Overall, investigators have found that UF milk retentates display different properties from those of standard or condensed milk. Sweetsur and Muir (70) observed that at similar protein concentrations, UF milk retentates were more stable to sterilization temperatures than condensed milks. Kosikowski et al. reported that when held chilled, UF milk retentates were also more stable to oxidation than raw and heated milks (71).

Mistry and Kosikowski (72) examined the influence of milk ultrafiltration on bacteriophages of lactic acid bacteria. They determined that phages present in milk are retained during UF due to their small molecular weights (23,000-72,000); also no phages were observed in the permeate. Phage concentration was, however, less than that of protein concentration, and phage destruction was obtained at 85C for 30 min., a treatment said to normally be given during starter preparation. They concluded that concentrating phage during UF would not be a drawback during starter manufacture. These observations agree with results obtained by Zottola et al. (73). A 20,000 molecular weight cut off was used by this group to do the ultrafiltration. It was concluded that phage particles do not pass through the membrane, but become entrapped in the polarization concentration layer or in the membrane.

QUALITY OF UF CHEDDAR CHEESE

Four factors are believed to determine the quality of Cheddar cheese: chemical composition, rate and extent of acid production, type of curd matrix before salting, and temperature of cheese during cheese curing (48). Of these four, the most important factor in producing cheese of uniform quality is thought to be the rate and extent of acid production in the vat; this determines the characteristic basic structure of the cheese and its pH (46). As shown by numerous UF Cheddar cheesemaking trials, these factors, and the degree of acid production in particular, are all affected by the concentration factor used during the ultrafiltration step and the cheesemaking process, whether traditional, MMV or LCR.

As previously mentioned, a selective concentration of proteins and calcium phosphate exponentially increase the buffering capacity with increasing total solids of the UF retentate affecting the bacterial lactic acid production (74). Research done by Green et al. (15) pointed out the three major problems causing quality defects when using milk concentrated by UF for Cheddar cheese production: a large loss of fat into the whey, abnormal cheese texture, and decreased maturation rate. Light microscopy observations showed the protein packed in large, more compact areas and the fat more segregated. The firmness, crumbliness, granularity and dryness of the cheese increased as the concentration factor of the milk increased. The concentration factor of the milk, as determined by Dalgleish (75) as well as the rennet concentration affected the amount of soluble casein at clotting time. Clotting

time itself was said not to be affected by concentration.

Despite the numerous difficulties with UF Cheddar cheese manufacture and its quality, equally numerous reports suggest that the production of acceptable cheese should be possible utilizing UF technology(76). As of June, 1986, it was reported that the U.S. Food and Drug Administration (FDA) would consider approval of the process for cheese on a case-by -case basis. The UF cheese must display the same physical and nutritive properties as the conventionally made cheese (77).

Challenges remain ; it is said that a major long-term goal of ultrafiltration research is the development of excellent commercial grade Cheddar cheese from MMV precheese in a continuous cycle. A full scale prototype plant of 32,000 kg. whole milk/hr. is reported under construction in Australia to prove the new technology (1).

MATERIALS AND METHODS

Cheddar Cheese

Thirteen Cheddar cheese longhorn style samples were analyzed monthly for a period of eleven months. The cheeses were provided by two different creameries in California. They were made from the same milk, using approximately the same manufacturing conditions and within a period of 4 months. Five of the thirteen samples were produced using regular milk and the conventional Cheddar cheese making process; these will be referred to as "CM-cheese". The other eight cheeses were made from milk concentrated from 1.65 to1.90 fold using low concentrated retentate filtration technology. These will be referred to as "UF-cheese". Weights and dates of manufacture are given in Table 1.

The moisture of the cheese at the time of manufacture, for both types, was given by the creameries as ranging from 37.6% to 37.8%, the salt content from 1.7 to 1.8% and the pH from 5.07 to 5.09. The cheese samples were stored at 40 ^{0}F in their original package and moved to room temperature for intervals of 15 min. maximum at testing periods only. Development of heavy mold growth, however, prevented complete sampling of some of the cheeses in the last months. Pinhole defects in the plastic wraper were thought to be the cause.

SAMPLE PREPARATION

Each cheese block was plugged monthly using a sterile cheese trier. The upper plug hole was filled with the outer 2 cms. of the cheese plug and immediately covered with hot paraffin wax to prevent or minimize mold growth. The paraffin wax was soon replaced with "amber-gel" (a petroleum derivative), because the wax seal broke easily upon transportation of the cheese blocks, allowing mold contamination.

Eleven grams from each cheese block were weighed, blended with 99 ml of sterile 2% sodium citrate solution and diluted, using 0.1% peptone water up to 10^9. The samples were then plated in duplicate and incubated at appropiate temperature and time. Preparation of the samples for microbiological analysis was done following standard procedures given in the Standard Methods for the Examination of Dairy Products (78).

Sensory evaluation of the Cheddar cheese samples was undertaken concurrently with the collection of samples for bacteriological testing. Coded samples of the cheese at room temperature were presented to the evaluator. Analysis of flavor, body and texture were done using the American Dairy Science Association score card and the USDA approach to scoring Cheddar cheese.

MICROBIOLOGICAL ANALYSES

Cheeses were analyzed for total bacterial content (aerobic plate count) on Standard Plate Count medium, staphylococcus count on Baird-Parker medium, coliform count on violet red bile agar, Salmonella count on Salmonella-Shigella medium, and enterococcus count on KF streptococcal medium. Specific media preparation, incubation times and temperatures were followed as indicated in the Standard Methods for the Examination of Dairy Products (78).

Additionally, the cheese was analyzed for total and heterofermentative lactobacilli using the three tube Most Probable Number method through six dilutions with vaspar overlay. Vaspar is a mixture of 50% vaseline petroleum jelly and 50% wax. Modified Elliker broth was the medium used (D.W. Weddle, personal communication). Vancomycin resistant bacteria,(pediococci, lactobacilli, Leuconostoc and some streptococci), were enumerated by plating cheese samples on modified Rogosa medium containing tomato juice and 10 ppm vancomycin

IDENTIFICATION OF ISOLATES

After each incubation period and during colony counting, representative colonies were selected from those growing on the modified Rogosa-vancomycin plates. Colonies were isolated from both cheese types. Of the 175 isolates obtained, 40 were further characterized, 23 from UF cheese and17 from CM

cheese. After reinoculating the isolates, the following observations were made: colony morphology, cell morphology, catalase test, litmus milk reaction at 30C and 37C, pH and carbohydrate fermentation pattern.

Determination of these characteristics was done following the methods given in the Standard Methods for the Examination of Dairy Products and The Compendium of Methods for the Microbiological Examination of Foods (78,79). The carbohydrate fermentation patterns were obtained via API Rapid CH identification kits for Lactobacillus species. Production of acid by anaerobic fermentation of 49 different carbohydrates is the basis for this test. A suspension of the microorganism to be tested is made in the medium provided, grown and inoculated in the strip of cupules. The degree of color change (acid production) is read at 3, 6 , 24 and 48 hours. Numbers from 1 through 5 are given according to the intensity of the reaction.

PLATING METHODS

At the onset of this project a new spiral plater (model D from Spyral Systems, Inc.) became available for use. In an effort to gain familiarity with this plating system, inoculations were done using both the manual spread plate method and the automated spiral plate method, both in duplicate. Colony counting was done using a Quebec colony counter for the spread plates and a viewing-grid (model MV) colony counter for the spiral plates.

The spread plates were counted according to the guidelines cited in The Compendium of Methods for the Microbiological Examination of Foods (79). The "counting rule of 20", along with constants provided in the spiral plater user manual were applied in obtaining the spiral plate counts.

SENSORY EVALUATION

An expert judge in the evaluation of Cheddar cheese carried out the analysis of flavor, body and texture. A penalty point deduct basis was used to score the cheese. Over-all, the lower the score given, the higher the quality of the sample. A score of 0 meant no defect or criticism, 0.5 or 1.0 points deducted for slight defects (criticism); 1.5 or 2.0 points were deducted for moderate to indefinite defect(s); and more than 2.5 points were deducted for more pronounced or combined defects. A perfect score is 0 (no defects); a lower score represents a higher sensory quality and a higher score a lower sensory quality. Defect characterzations were those specified in the official American Dairy Science Association score card for Cheddar cheese.

STATISTICAL ANALYSIS

The data obtained were analyzed assuming that the cheeses acted as a random sample of cheeses from each cheese treatment. Statistical analysis of the cheese bacterial plate counts was done by repeated measures MANOVA using PC-SAS.

Sensory analysis data were not of a continuous nature, therefore, the values were simply averaged over time to get a single number for each cheese block. These averages were analyzed using both a standard t-test and a non-parametric test (Wilcox).

RESULTS

Eight Cheddar cheeses made from ultrafiltered concentrated milk and five made from conventional milk were evaluated for microbiological and sensory characteristics in an effort to establish whether or not differences exist between the two cheese types. Two enumeration methods; spread and spiral plate counting, were also used and a comparison of the methods was done.

AEROBIC PLATE COUNT

Aerobic plate counts (total viable bacteria) are presented in Tables 2 and 3. From Table 2 it can be seen that cheese block "E" (UF-cheese) showed the highest bacterial load at 22 weeks and the lowest bacterial numbers were found in cheese blocks "I" (CM-cheese) and "K" (UF-cheese) at the end of 57 weeks of storage. Other cheese samples showed a wide variation in bacterial population. In general, both cheese types showed higher bacterial counts during earlier weeks of storage and decreased with increasing storage time. Some exceptions were noted, however. When the bacterial counts were determined by the spiral plate method (Table 3) the same trend was noticed, except that bacterial numbers were slightly lower.

Average viable bacterial numbers were compared. The results are presented in Figures 1 and 2. There was not much variation in the over-all trend between the data

obtained using the spiral plate technique and the data obtained using the traditional spread plate method.

The bacterial population in UF-cheese was initially slightly lower than in CM-cheese and reached its maximum around 35 weeks of age, continuing with a gradual decline. On the other hand, although the conventional milk cheese had a slightly higher bacterial population initially (13 weeks), its counts were much lower than in UF-cheese at the end of ripening (57 weeks). A greater variation in bacterial population between UF-cheese and CM-cheese was noticed at 35 weeks of age than at any other time.

VANCOMYCIN RESISTANT BACTERIA

Spread and spiral plate methods were also used in the enumeration of these bacteria. Results are presented in Tables 4 and 5. As in the aerobic plate count, vancomycin resistant bacteria in general were higher at 13 weeks of ripening than at 57 weeks. The $\log_{10}$ of bacterial counts ranged from 7.82 colony forming units (CFU) per gram in the case of cheese block "E" (UF-cheese) at 22 weeks to 3.30 CFU/gram in samples "G" (CM-cheese), "M" and "K" (UF-cheese) at 38 and 47 weeks (Table 4). Figures 3 and 4 show the average number of vancomycin resistant bacteria in the cheeses. Similar to the observations made with aerobic plate counts, the lactic acid bacteria (LAB) of the UF-cheese were lower than that of the CM-cheese in the initial weeks (13 weeks) and slightly higher at the end of 57 weeks.

At 35 weeks UF-cheese showed the highest count of vancomycin resistant bacteria, and CM-cheese the lowest.

In the case of UF-cheese there was an over-all gradual increase in vancomycin resistant bacteria up to 35 weeks of ripening and then a steady decline. The trend observed in case of CM-cheese was almost reversed. However the counts at 13 weeks were not much different from those at 57 weeks. Here too the counts obtained using the spiral plater seem slightly lower than those given by the spread plate method

HETEROFERMENTATIVE LACTIC ACID BACTERIA

Modified Elliker broth was used to enumerate these bacteria. Table 6 shows the total lactic acid bacteria as determined by the Most Probable Number method and also identifies the cheeses containing heterofermentative bacteria. Out of 13 cheeses sampled, 9 were positive for gas production at least once during the testing period. Four of the CM-cheeses positive for heterofermentative LAB (F,G,H,I) and 3 of the UF-cheeses (J,L,M) were all manufactured at the same creamery. Some cheeses were positive for gas forming lactobacilli only occasionally (cheeses D,E,G,I and J), whereas other samples were positive on a more regular basis.

Figure 5 compares the average lactic acid bacterial numbers of UF and CM cheeses. Unlike aerobic plate and vancomycin resistant counts, the cheese made

from UF milk showed higher lactic bacterial numbers at 13 weeks than the CM-cheese. The lactic acid bacterial counts showed a much greater fluctuation between the two cheese types than previous counts

COLIFORM, STAPH, SALMONELLA, AND ENTEROCOCCUS COUNTS

Efforts to enumerate these potential pathogens in the cheeses yielded no significant counts with one exception, cheese block "E" (UF-cheese). This block of UF-cheddar cheese gave aerobic and LAB counts much higher than the other 12 cheese samples from the initial analysis and steadily high thereafter. Following the same trend, it also yielded detectable numbers of staphylococci and E. coli. All other samples, UF and CM cheeses gave no countable colonies of these bacteria.

CHARACTERIZATION OF LACTIC ACID BACTERIA

The 40 isolates chosen (23 from UF cheese and 17 from CM cheese) had typical colony and cell morphologies for lactic acid bacteria as described in Burgey's Manual for Determinative Bacteriology (80). They also gave a negative reaction for the catalase test. Subcultures from colonies of gram-positive, non-spore-forming, rods in pairs or chains, presumptive positive lactobacilli, were done in litmus milk and incubated at 30 ^{0}C and 37 ^{0}C.

Tables 9 and 10 present litmus milk reactions and pH measurements at these temperatures. Table 11 shows the carbohydrate fermentation reaction results of

the isolates. Observations from these tables permit the conclusion that the bacterial populations in both cheese types were quite similar in their activity in litmus milk and in carbohydrate fermentation capability.

SENSORY ANALYSIS RESULTS

When the cheese samples were tested for microbiological qualities, they were also analyzed for their rheological and sensory characteristics such as flavor, body and texture. The results are presented in Tables 7 and 8. Cheese samples were scored for flavor characteristics on a scale of 0-10, where 0 was best and 10 was worst (unacceptable) based on American Dairy Science Association guidelines (Table 7). Flavor scores of cheese ranged from 1.0 to 6.0. Most of the cheeses were well within the acceptable range.

Figure 6 shows the average flavor scores of cheeses made from both UF and conventional milk. At 13 weeks of age the cheese from UF milk had much better flavor than the cheese from conventional milk. However, at 57 weeks the difference in flavor between the two cheese types was much more narrow. UF-cheese showed more fluctuation in flavor characteristics from week to week than the CM-cheese.

Another rheological property of cheese samples tested was body and texture. The body and texture of the samples was scored on a scale of 0-5, 0 being the best and 5 unacceptable, based on ADSA guidelines (Table 8). The body and texture scores ranged from 0.0 to 3.0, indicating that the samples were better than the average with

respect to their body and texture. The average body and texture scores of both cheese types are compared in Figure 7. The body and texture characteristics of the cheeses differed much more widely than the flavor characteristics (Figures 6 and 7).

DISCUSSION

It is held that the success of Cheddar cheesemaking is assessed in terms of the yield and quality of the matured cheese. Yield and quality in turn depend on the materials and processes used in the manufacture of the cheese (50). With any one variety, alterations of either the materials or manufacturing conditions have been shown to alter the final cheese structure or texture (15).

Cheddar cheese made from ultrafiltered concentrated milk, therefore, should be expected to differ from Cheddar cheese made from regular milk, since its manufacture requires a change in both the raw material and the process. However, as the literature review shows, the degree of change appears to depend on the concentration factor used during the ultrafiltration of the milk. No significant changes in the process, for example, seem to be required for satisfactory cheesemaking from milk concentrated by UF up to 2 fold (as in the cheese used for this study) (19). On the other hand, the use of a conventional cheese making procedure with higher concentrations of milk results in losses of fat, and in the production of cheese of abnormal composition, flavor and texture (15,55).

The bacteriological data presented here indicate that the Cheddar cheese made from milk concentrated between 1.65-1.90, as used in this study, had non-starter lactic acid bacteria (NSLAB) quite similar to that of the Cheddar cheese made from conventional milk. NSLAB isolates from both cheese types showed comparable

carbohydrate fermentation patterns and similar metabolic activities in milk.

The importance of NSLAB in the ripening of conventionally made cheese has been established. Unlike starter bacteria, NSLAB remain viable at higher densities for long periods and perform activities which may be relevant in flavor development. One such activity is the fermentation of lactate to acetate and carbon dioxide, which lowers the redox potential, (a reducing environment stabilizes reduced sulphur compounds for optimum flavor development) (81,82). It is important, therefore, that the NSLAB be able to survive and carry out the metabolic activities in the ultrafiltered Cheddar cheese as well.

The low pH ($\sim$5) ,low redox ($\sim$-200 mv), low moisture, high salt concentration and low storage temperature make conventionaly made cheese a highly restrictive environment for the growth of contaminating bacteria (83); however, in ultrafiltered concentrated milk cheese, a strong buffering capacity can affect cheese quality adversely by making it more difficult to attain optimum pH and by creating an opportunity for growth of food poisoning organisms (1).

Rash and Kosikowski, for instance, found profuse growth and high survival of enteropathogenic Escherichia coli in Camembert cheese made from highly concentrated UF retentates (84,85). In the case of the UF-Cheddar cheese involved in this study, however, no significant counts of possible pathogens were detected. UF- cheese block "E" was the exception; and in this case, the probability of an isolated case of initial contamination during the manufacture is likely. A persistent unclean aroma was

noted on its flavor score card and all other bacterial counts also were comparatively higher than the rest of the cheeses.

The texture of cheese is said to be important because it is this property by which the consumer first identifies and judges the specific variety. Overall appearance and mouth feel are noted before flavor is assessed (86). One of the objectives of UF cheesemaking is to incorporate whey proteins into the cheese to increase yields (16); but it has been shown that cheese texture may be significatly changed by the incorporation of these whey proteins (87). de Koning, et al., for example, established that cheese made from UF milk generally had a smoother consistency than the cheese made from whole milk. The degree of smoothness presumably depended upon the proportion of whey proteins incorporated (88). Lawrence et al. on the other hand suggested that UF-cheese ripens more slowly than traditionally made cheese, because B-lactoglobulin, the major whey protein, inhibits plasmin activity and that undenatured whey proteins are resistant to the action of chymosin and other proteinases (86).

In this study, the texture scores showed UF cheeses to have a lower average texture scores (mean of 0.87 UF vs. mean of 1.04 CM-cheese) which means that they had a slightly higher textural quality than the conventionally made Cheddar cheese.

Full Cheddar flavor development is said to be ensured by the use of appropriate starter strains and manufacturing conditions. The response to these selected manufacturing conditions, rather than any single difference between particular starter strains, is thought to determine the likelihood of bitterness development

(88,88,90). In this respect, again UF Cheddar cheese would be expected to differ from CM-cheese in its flavor qualities because the lactic starter is exposed to different manufacturing conditions. Tolerance of lactic starter bacteria to osmotic pressure is said to be of particular importance in UF technology (91). However, Cheddar cheese flavor is also attributed to a complex association of chemical compounds produced during the manufacturing and ripening processes by the degradation of protein, fat and lactose (92).

If whey proteins incorporated into the UF cheese were accidentally denatured because of inappropiate UF equipment and conditions, they would be expected to behave like heat denatured material, undergoing proteolysis during ripening and giving rise to flavor defects (16). However, in this study, there was no significant overall difference in the flavor scores of the two cheese types.

UF cheeses presently made are marketed under traditional names even though casein breakdown, ripening, texture and flavor are somewhat different from those of traditionally made cheeses. In this regard, Lawrence et al., suggest that these new characteristics of the cheese may well prove to be very acceptable commercially, and that it may be more logical to develop a new range of cheese varieties rather than to attempt to duplicate exactly the properties of traditional cheese (86).

In this study there was no statistical evidence of an average difference over time between the two methods (method main effect) or evidence of a difference in time trend between the two methods (time by method interaction). This lack of evidence of

difference leads to the conclusion that Cheddar cheese manufactured from ultrafiltered concentrated milk (1.65-1.90 fold) and Cheddar cheese manufactured from conventional milk do not differ greatly in bacteriological or sensory parameters.

Use of both the spiral plater and the the manual spread plate in the testing of the cheeses allowed for a comparision of the methods. In the individual bacterial counts of the cheeses, the spiral plate method had a slight tendency to give lower counts than the spread plate method. However, Gilchrist et al. found in their study of the spiral plate method that this system gave counts higher than those obtained by the pour plate method. A greater breaking up of clumps of bacteria resulting from spreading a small volume of liquid on a surface was thought responsible for the difference (93).

Several advantages of spiral plating instead of spread plating were observed; first, it is easy to work with the system, it saves time, the work of preparing dilutions and the use of extra pipettes, dilution bottles and dilution fluid. It also allows visual differentiation of species because the bacteria are deposited along fixed lines and the growth rate of the same species is equal. Another advantage is that bacteria are dispensed only on the clear, level part of the agar which facilitates the counting. Cross contamination from sample to sample was not a problem. A quick rinse with 5% sodium hypochlorite was used between platings.

A few disadvantages were also noted; the agar plates prepared for use with the spiral plater must have a perfectly level, smooth and dry surface. Equal amounts of

media have to be dispensed in each plate, which calls for automated plate pouring, otherwise extremely careful and time consuming preparation of the plates is required. The counting technique was found to be completely different from traditional counting, and even though the colonies were equal in size when using the spiral plate, the counting was initially somewhat difficult to master because counting is limited to small delineated areas. In this respect, further automation of the system by having an electronic laser or another automatic counting system is expected (94). Another disadvantage is that this method does not permit counting bacteria in numbers $<2x10^2/g$.

Liberski (94) cocluded that the spiral plate apparatus can be recommended for the determination of the number of microorganisms in chilled cured meat products, even though other authors (95) have mentioned problems with blocked styluses in spiral platers caused by small particles from meat. Donnelly, et al. (96) used raw and pasteurized milk. Their study indicated the spiral plate system could be substituted for the Standartd Plate Method in the bacteriological examination of milk. From the observations made throughout this study, it can be concluded that the spiral plate system can be successfully used for the determination of the lactic acid bacterial population and the aerobic plate count of Cheddar cheese, but not for the detection of enteric species since their numbers are low. If care is excersized when manually pouring the media plates and during the colony counting, savings in time, space and costs are possible.

Fig. 1. TOTAL PLATE COUNT, spread plate

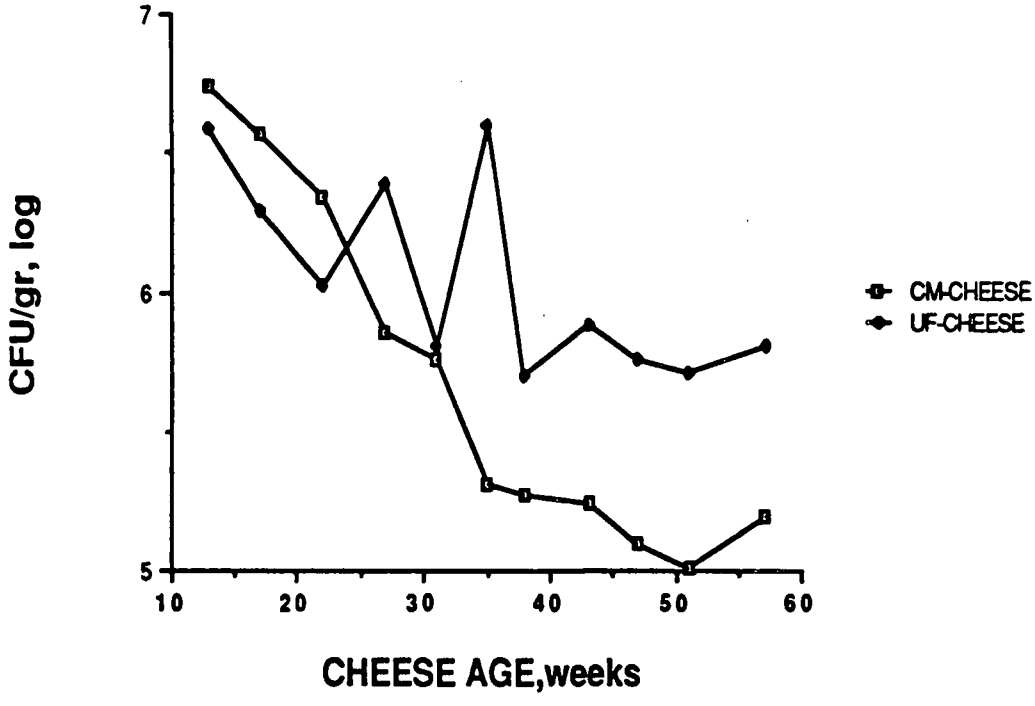

Fig. 2. TOTAL PLATE COUNT, spiral plate

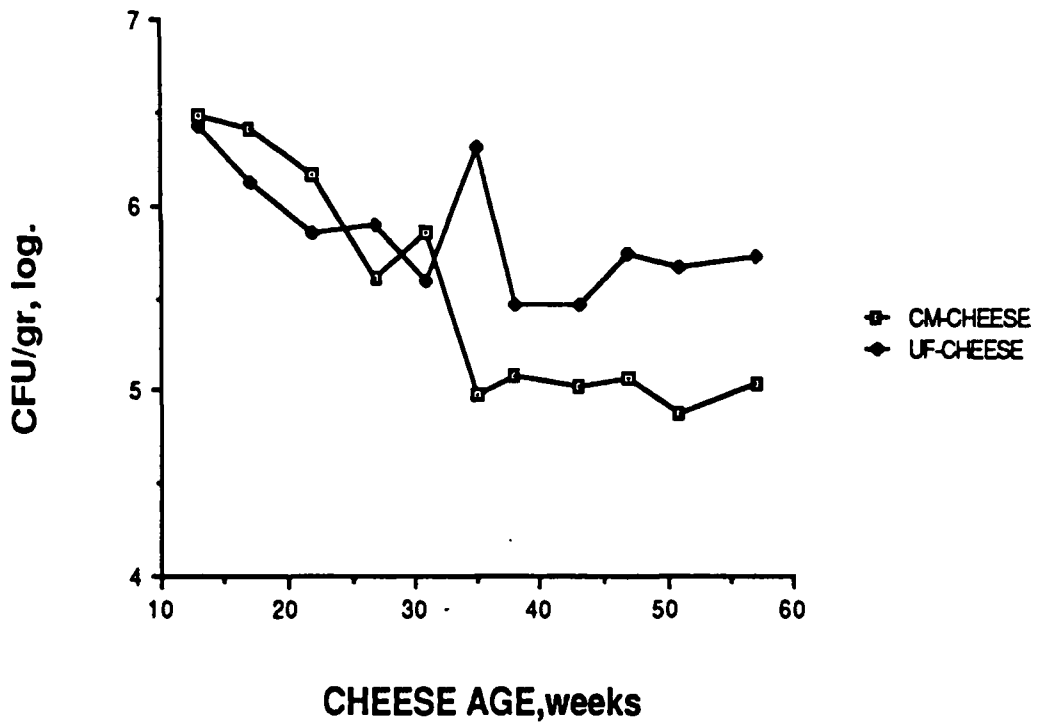

Fig. 3. VANCOMYCIN RESISTANT BACTERIA, spread plate

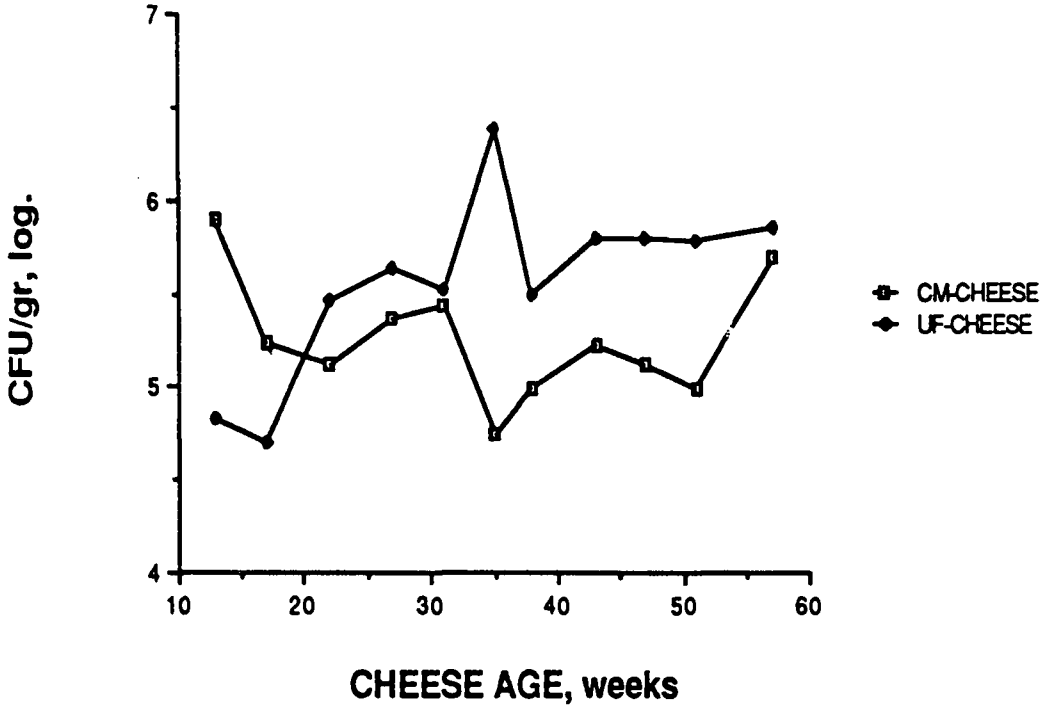

Fig. 4. VANCOMYCIN RESISTANT BACTERIA, spiral plate

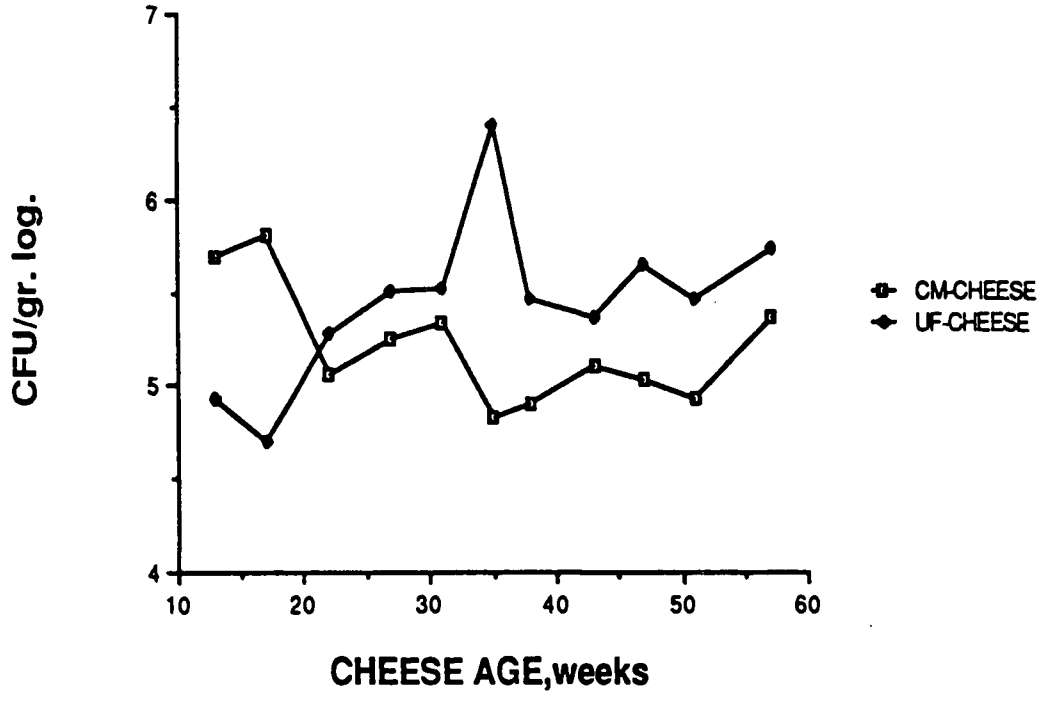

Fig.5 TOTAL LACTIC ACID BACTERIA, MPN

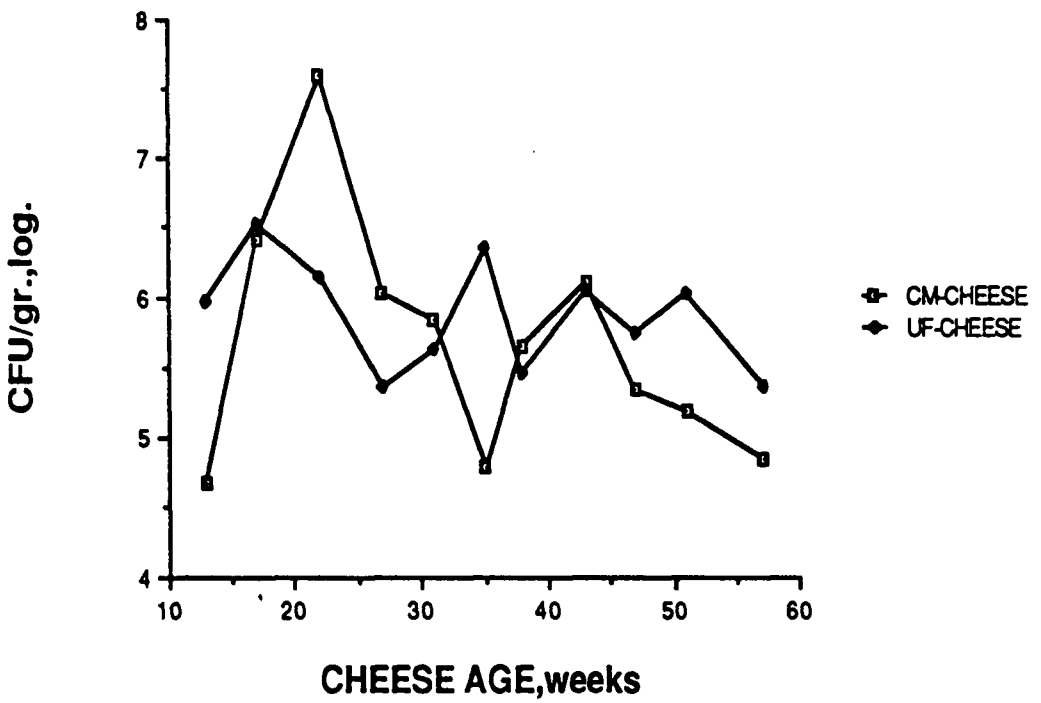

Fig. 6. CHEESE FLAVOR SCORES

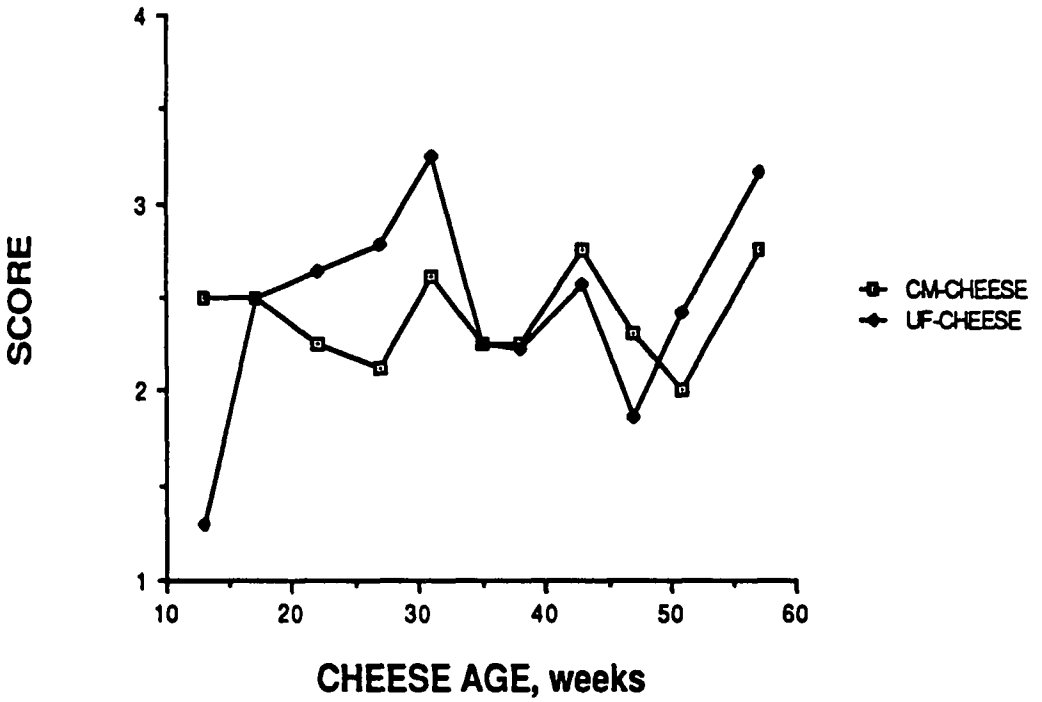

Fig. 7. CHEESE BODY AND TEXTURE SCORES

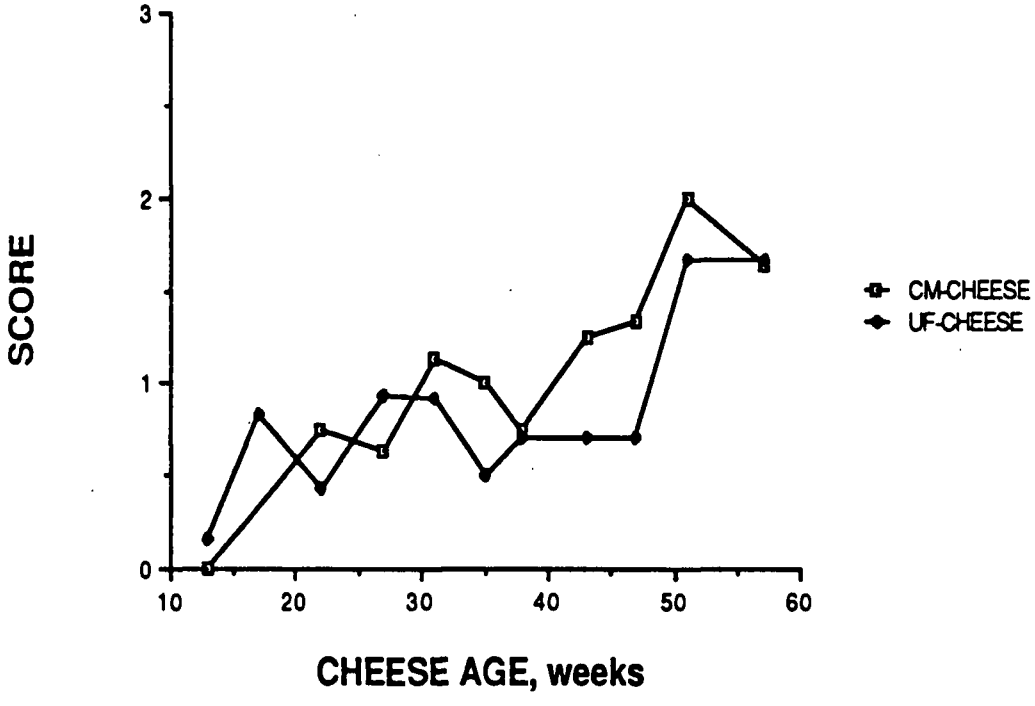

Table 1. **EXPERIMENTAL CHEDDAR CHEESES**

CHEESE SOURCE	CHEESE LAB. I.D.	CHEESE TYPE	DATE OF MANUFACTURE	CHEESE WT.(Lbs.)
1	A	CM	1- 3- 85	40
1	C	UF	1- 9- 85	40
2	E	UF	1- 7- 85	40
2	B	UF	2-12-85	40
2	D	UF	2-17-85	40
2	F	CM	3-20-85	12
2	G	CM	3-26-85	12
2	J	UF	3-27-85	12
2	H	CM	4- 5- 85	12
2	K	UF	4-10-85	12
2	L	UF	4-12-85	12
2	M	UF	4-18-85	12
2	I	CM	4-19-85	12

CM conventionally made Cheddar cheese
UF ultrafiltered-milk Cheddar cheese
1 creamery
2 creamery

Table 2. VIABLE BACTERIAL COUNTS (Log_{10} CFU/g) IN CHEESES MADE FROM ULTRAFILTERED-CONCENTRATED MILK AND CONVENTIONAL MILK AS DETERMINED BY THE SREAD PLATE METHOD.

sample	cheese type	cheese age (weeks)										
		13	17	22	27	31	35	38	43	47	51	57
A	CM	-	-	-	5.00	-	5.00	5.60	5.70	5.00	5.37	5.25
B	UF	-	-	5.98	7.11	6.89	7.64	7.56	7.82	-	-	-
C	UF	-	-	-	7.65	-	6.74	7.17	7.08	7.09	6.77	7.50
D	UF	-	-	5.60	6.02	5.30	5.78	6.20	5.30	5.73	6.43	6.00
E	UF	-	-	8.00	7.79	7.57	7.15	6.94	7.42	7.27	6.99	7.46
F	CM	-	7.14	6.02	5.60	5.95	5.93	5.61	5.68	5.74	4.96	4.81
G	CM	-	6.41	6.08	6.61	5.30	5.00	3.78	3.88	4.04	4.72	6.31
H	CM	6.71	6.47	6.45	6.28	6.79	-	6.96	6.44	6.26	5.53	6.00
I	CM	6.76	6.24	6.98	5.78	5.00	-	4.42	4.52	4.48	4.45	3.60
J	UF	-	6.53	6.30	6.10	5.00	5.70	4.87	5.20	6.67	5.48	5.49
K	UF	6.19	5.90	5.70	5.00	5.00	-	3.74	4.43	4.00	4.42	3.60
L	UF	6.43	6.30	5.70	6.16	5.85	-	5.38	5.08	5.00	5.32	5.62
M	UF	7.14	6.43	6.40	5.30	5.00	-	3.74	4.72	4.56	4.53	4.90

- data not available

UF ulftrafiltered-concentrated-milk cheese

CM conventionally made cheese

Table 3. VIABLE BACTERIAL COUNTS (Log_{10} CFU/g) IN CHEESES MADE FROM ULTRAFILTERED-CONCENTRATED MILK AND CONVENTIONAL MILK AS DETERMINED BY THE SPIRAL PLATE METHOD.

sample	cheese type	cheese age (weeks)										
		13	17	22	27	31	35	38	43	47	51	57
A	CM	-	-	-	4.85	-	4.90	5.00	5.20	5.60	5.13	5.18
B	UF	-	-	5.93	6.50	6.91	7.28	7.40	7.41	-	-	-
C	UF	-	-	-	6.50	-	6.91	6.81	7.01	7.03	6.70	7.26
D	UF	-	-	5.60	5.28	5.46	5.53	4.70	5.39	5.76	6.52	5.81
E	UF	-	-	7.83	7.58	7.35	7.11	6.82	7.20	7.10	7.37	7.49
F	CM	-	6.58	6.40	5.81	5.72	5.75	5.58	5.52	4.95	4.53	4.60
G	CM	-	6.54	5.18	6.00	5.38	4.30	4.10	3.79	3.91	5.00	6.04
H	CM	6.60	6.52	6.90	6.18	7.61	-	6.61	6.29	6.16	5.59	5.84
I	CM	6.38	5.72	6.19	5.20	4.70	-	4.13	4.30	4.70	4.17	3.49
J	UF	-	6.34	5.76	6.04	5.11	4.70	4.67	5.12	5.20	4.89	5.27
K	UF	6.51	5.76	4.95	4.30	4.30	-	3.72	4.61	5.08	4.68	3.95
L	UF	6.20	6.00	5.78	5.83	5.60	-	5.49	4.20	5.44	5.46	5.58
M	UF	6.58	6.38	5.70	5.18	5.38	-	4.10	2.70	4.07	4.53	4.66

- data not available

UF ulftrafiltered-concentrated-milk cheese

CM conventionally made cheese

Table 4. **VANCOMYCIN RESISTANT BACTERIAL COUNTS (Log_{10}CFU/g) IN CHEESES MADE FROM ULTRAFILTERED-CONCENTRATED MILK AND CONVENTIONALMILK AS DETERMINED BY THE SPREAD PLATE METHOD.**

sample	cheese type	cheese age (weeks)										
		13	17	22	27	31	35	38	43	47	51	57
A	CM	-	-	-	5.11	-	4.30	5.06	5.32	4.85	5.51	6.03
B	UF	-	-	5.30	7.00	6.91	7.29	7.25	7.25	-	-	-
C	UF	-	-	-	6.00	-	6.55	6.96	7.09	7.13	6.91	7.46
D	UF	-	-	5.04	5.49	5.34	5.76	5.11	5.33	5.82	6.70	5.48
E	UF	-	-	7.82	7.39	7.52	7.38	7.00	7.48	6.99	6.99	7.39
F	CM	-	6.10	5.90	5.33	5.04	5.91	5.61	6.15	5.90	5.12	5.81
G	CM	-	4.30	4.00	5.26	5.64	4.00	3.30	3.81	4.00	4.51	6.40
H	CM	6.70	6.50	6.29	6.41	7.05	-	6.83	6.48	6.20	5.76	6.47
I	CM	5.10	4.00	4.30	4.74	4.00	-	4.19	4.36	4.66	4.06	3.80
J	UF	-	4.30	5.13	4.78	5.20	4.90	4.92	5.21	6.90	5.60	6.36
K	UF	4.00	4.00	4.00	4.00	4.00	-	4.00	4.61	3.30	4.51	3.84
L	UF	6.20	5.77	5.91	6.00	5.68	-	5.43	5.21	5.95	5.44	5.57
M	UF	4.30	4.78	5.00	4.48	4.00	-	3.30	4.11	4.52	4.30	4.83

- data not available

UF ulftrafiltered-concentrated-milk cheese

CM conventionally made cheese

Table 5. **VANCOMYCIN RESISTANT BACTERIAL COUNTS (Log_{10}CFU/g) IN CHEESES MADE FROM ULTRAFILTERED-CONCENTRATED MILK AND CONVENTIONAL MILK AS DETERMINED BY THE SPIRAL PLATE METHOD.**

sample	cheese type	cheese age (weeks)										
		13	17	22	27	31	35	38	43	47	51	57
A	CM	-	-	-	5.50	-	4.30	4.79	5.42	5.10	5.30	5.18
B	UF	-	-	5.30	6.45	6.80	7.24	7.34	7.30	-	-	-
C	UF	-	-	-	5.52	-	6.96	6.98	7.00	7.00	6.57	7.26
D	UF	-	-	4.30	5.40	5.18	5.53	4.95	5.90	5.59	6.58	5.91
E	UF	-	-	7.84	7.55	7.39	7.20	6.86	7.20	7.14	6.92	7.33
F	CM	-	5.95	5.62	5.15	4.78	5.89	5.43	5.72	5.48	5.00	5.64
G	CM	-	5.95	4.48	5.04	4.95	4.30	3.90	3.69	3.78	4.70	5.98
H	CM	6.60	6.54	5.85	6.25	7.33	-	6.60	6.34	6.25	5.69	6.46
I	CM	4.78	4.78	4.30	4.30	4.30	-	3.83	4.31	4.60	4.03	3.52
J	UF	-	4.30	5.15	4.78	5.00	5.00	4.83	5.09	5.28	4.89	5.24
K	UF	4.30	4.30	4.30	4.30	4.30	-	3.36	4.59	4.30	4.54	3.86
L	UF	5.90	5.89	5.80	5.77	5.52	-	5.51	3.20	5.74	5.20	5.57
M	UF	4.60	4.30	4.30	4.30	4.48	-	3.94	2.60	4.53	3.57	4.95

- data not available

UF ulftrafiltered-concentrated-milk cheese

CM conventionally made cheese

Table 6. TOTAL LACTIC ACID BACTERIA COUNTS (Log_{10} CFU/g) IN CHEESES MADE FROM ULTRAFILTERED-CONCENTRATED MILK AND CONVENTIONAL MILK AS DETERMINED BY THE MOST PROBABLE NUMBER METHOD.

sample	cheese type	cheese age (weeks)										
		13	17	22	27	31	35	38	43	47	51	57
A	CM	-	-	-	4.63	-	4.36	6.60	4.36	4.36	4.63	4.97
B	UF	-	-	6.60	7.36	7.36	7.36	7.36	7.63	-	-	-
C	UF	-	-	-	5.38	-	7.36	7.36	6.38	7.36	7.36	7.36
D	UF	-	-	6.60	5.38	6.60	5.38	4.63	5.38	7.15	7.36	6.60
E	UF	-	-	6.95	5.38	6.95	7.36	7.36	7.20	7.36	7.36*	6.60
F	CM	-	5.66	7.38*	7.97*	7.36	5.66*	6.60*	7.36*	6.85*	4.63	4.36
G	CM	-	4.95	7.38	7.04*	4.36	4.36	4.36	4.36	4.36	4.36	4.36
H	CM	5.38	7.66*	7.88	6.85*	7.63*	-	6.85*	7.36*	4.63	5.38	6.95
I	CM	3.95	7.38	7.71	3.60	3.95	-	3.85*	7.04	6.48*	6.95	3.60
J	UF	-	3.95	7.38	6.38*	4.36	4.36	4.36	5.38	5.38	3.60	4.63
K	UF	6.95	7.38	3.60	3.60	3.95	-	3.95	4.36	3.60	4.97	3.60
L	UF	6.60	7.38*	5.38*	5.38*	6.60	-	4.63	6.60	5.38*	6.60	4.81
M	UF	4.36	7.38	6.48*	3.95	3.60	-	3.95	5.38	3.95	4.97	3.95

* gas production

\- data not available

UF ulftrafiltered-concentrated-milk cheese

CM conventionally made cheese

Table 7. **FLAVOR SCORES OF CHEESES MADE FROM ULTRAFILTERED-CONCENTRATED MILK AND CONVENTIONAL MILK**

sample	cheese type	cheese age (weeks)										
		13	17	22	27	31	35	38	43	47	51	57
C	UF	-	-	3.0	1.0	-	3.0	3.0	4.0	1.0	1.0	2.0
D	UF	-	2.0	2.0	3.0	3.0	1.5	2.0	2.0	1.5	2.5	2.5
E	UF	-	4.0	3.0	6.0	5.0	3.0	1.5	3.0	2.0	-	-
F	CM	-	2.0	2.0	3.0	2.5	2.0	2.5	3.0	3.0	2.0	2.5
G	CM	-	2.0	2.0	3.0	2.0	2.5	2.5	2.0	2.0	2.5	3.0
H	CM	3.0	2.0	3.0	1.5	3.0	-	2.0	3.0	-	2.5	2.5
I	CM	2.0	4.0	2.0	1.0	3.0	-	2.0	3.0	2.0	1.0	3.0
J	UF	-	3.0	3.5	2.0	3.0	1.5	2.5	2.5	2.0	2.0	2.5
K	UF	2.0	3.0	3.0	2.0	3.0	-	2.5	2.5	2.0	3.5	4.0
L	UF	1.0	1.0	2.0	2.5	2.5	-	2.0	2.5	2.0	2.5	4.0
M	UF	1.0	2.0	2.0	3.0	3.0	-	2.0	1.5	2.5	3.0	4.0

1-10 lower score= better flavor

\- data not available

UF ulftrafiltered-concentrated-milk cheese

CM conventionally made cheese

Table 8. **TEXTURE SCORES OF CHEESES MADE FROM ULTRAFILTERED-CONCENTRATED MILK AND CONVENTIONAL MILK**

sample	cheese type	cheese age (weeks)										
		13	17	22	27	31	35	38	43	47	51	57
C	UF	-	-	0.0	0.0	-	1.0	1.0	0.5	0.5	0.5	1.5
D	UF	-	1.0	0.0	1.0	1.0	0.0	0.5	0.5	0.0	2.0	1.5
E	UF	-	2.0	1.0	2.0	1.0	0.5	1.0	0.5	0.5	-	-
F	CM	-	0.0	1.0	1.0	1.0	1.0	1.0	0.5	1.5	3.0	2.0
G	CM	-	0.5	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.5	1.5
H	CM	0.0	0.5	0.0	0.0	1.5	-	0.5	1.5	-	2.5	1.5
I	CM	0.0	1.0	1.0	0.5	1.0	-	0.5	2.0	1.5	1.0	1.5
J	UF	-	0.0	1.0	1.0	1.0	0.5	0.5	1.5	1.0	1.5	1.5
K	UF	0.0	0.5	1.0	1.0	1.0	-	1.0	0.5	1.0	2.0	1.5
L	UF	0.0	0.5	0.0	1.0	0.5	-	0.5	1.0	1.0	2.0	2.0
M	UF	0.5	1.0	0.0	0.5	1.0	-	0.5	0.5	1.0	2.0	2.0

0-5 lower score= better flavor

- data not available

UF ulftrafiltered-concentrated-milk cheese

CM conventionally made cheese

Table 9. **LITMUS MILK REACTION OF CONVENTIONAL MILK CHEDDAR CHEESE NON-STARTER LACTIC ACID BACTERIAL ISOLATES**

CM-CHEESE ISOLATE #	PH/LITMUS MILK REACTION 30°C	37°C
2	4.81 /r,nc	4.55 /a,r,c
14	4.52 /a,r,nc	4.65 /a,r,c
26	4.15 /a,r,c	4.89 /a,nc
33	4.29 /a,r,c	4.42 /a,r,c
37	3.46 /a,r,c	3.46 /a,r,c
41	3.99 /a,r,c	3.78 /a,r,c
51	4.88 /a,nc	4.85 /a,nc
60	5.20 /a,nc	4.39 /a,r,c
72	3.91 /a,r,c	3.69 /a,r,c
79	4.35 /a,r,c	3.90 /a,r,c
90	3.86 /a,r,c	3.52 /a,r,c
106	4.12 /a,r,c	6.08 /k,nc
132	4.59 / a,r,sc	3.73 /a,r,c
141	5.91 /k,nc	5.61 /a,nc
155	4.41/a,r,sc	4.03 /a,r,sc
172	4.83 /a,r,c	4.02 /a,r,c
175	3.75 /a,r,c	3.79/ a,r,c

a acid
r reduced
k alkaline
c hard curd
sc soft curd
nc no curd

Table 10. **LITMUS MILK REACTION OF ULTRAFILTERED-CONCENTRATED-MILK CHEDDAR CHEESE NON-STARTER LACTIC ACID BACTERIAL ISOLATES**

UF-CHEESE ISOLATE #	PH/LITMUS MILK REACTION 30°C	37°C
4	4.48 /a,r,c	5.16 /r,nc
25	4.08 /a,r,sc	3.68 /a,r,c
29	4.97 /a,nc	4.42 /a,r,c
35	4.80 /a,r,nc	4.31 /a,r,c
46	3.68 /a,r,c	3.47 /a,r,c
56	4.84 /a,r,nc	4.45 /a,r,c
64	5.11 /a,nc	4.87 /a,nc
74	4.12 /a,r,c	3.53 /a,r,c
83	3.86 /a,r,c	4.23 /a,r,c
88	4.05 /a,r,c	3.73 /a,r,c
86	3.97 /a,r,c	3.68 /a,r,c
93	3.97 /a,r,c	3.71 /a,k,c
99	5.66 / k,r,nc	5.63 /k,r,nc
105	5.13 /a,nc	5.25 /a,nc
113	4.10/a,r,c	5.10 /a,nc
123	3.81 /a,r,sc	3.93 /a,r,c
129	5.26 /a,nc	3.79 / k,nc
134	4.22 /a,r,c	3.72 /a,r,c
150	5.52 /k,nc	5.63 /k,nc
164	6.06 /k,nc	6.01 /k,nc
166	4.08 /a,r,c	4.17 /a,r,c
167	5.42 /k,nc	5.48 /k,a,nc

a acid
r reduced
k alkaline
c hard curd
sc soft curd
nc no curd

Carbohydrates Fermented (+) or Not Fermented by Lactic Acid Bacterial Isolates from CM and UF Cheddar Cheese

Carbohydrate	CM CHEESE																
	2	14	26	33	37	41	51	60	72	79	90	106	132	141	155	172	175
L-Arabinose	-	+	+	+	-	+	+	+	+	+	+	+	-	+	+	+	-
Ribose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-Xylose	-	-	-	-	-	+	-	-	+	+	+	-	-	+	+	+	-
Adonitol	-	-	-	+	-	+	-	+	-	-	+	-	+	-	-	-	-
D-Glucose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Galactose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-Fructose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-Mannose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
L-Sorbose	-	+	-	+	-	+	-	+	-	+	+	-	+	-	-	-	+
Rhamnose	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Dulcitol	-	+	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-
Mannitol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Sorbitol	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	+
α-Methyl D-Mannoside	-	-	-	-	-	-	-	-	+	+	-	-	-	-	+	+	+
D-Glucoside	-	-	-	-	-	+	-	-	-	-	+	-	-	+	+	-	-
N-Acetyl-Glucosamine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Amigdalin	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	+
Arbutin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Esculin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Salicin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Cellobiose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Maltose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-
Lactose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Melibiose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Saccharose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trehalose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Inuline	+	+	+	-	+	-	+	-	+	+	-	-	-	-	-	-	+
Melezitose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-
Raffinose	+	-	-	-	-	+	-	+	+	+	+	+	+	+	-	-	+
β-Gentiobiose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-
D-Turanose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-
D-Tagatose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-Arabitol	-	-	+	-		-	-	-	+	+	-	+	-	-	-	-	+
L-Arabitol	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Gluconate	-	+	-	+	+	+	+	-	+	+	+	+	-	+	-	-	-

Carbohydrate	UF CHEESE																						
	4	25	29	35	39	46	56	64	74	83	88	86	93	99	105	113	123	129	134	150	164	166	169
L-Arabinose	-	+	+	+	+	-	+	-	+	+	-	-	-	-	-	-	-	-	-	-	+	+	+
Ribose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+
D-Xylose	-	-	-	+	+	-	-	-	-	-	-	-	-	-	+	+	-	+	-	+	+	+	+
Adonitol	-	-	-	-	-	+	-	+	-	-	-	-	+	-	-	-	-	-	+	-	-	-	-
D-Glucose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Galactose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-Fructose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-Mannose	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
L-Sorbose	-	+	+	-	+	+	+	+	+	+	-	+	+	-	-	-	-	-	+	-	-	-	-
Rhamnose	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	+	-	-	-	-	-	-
Dulcitol	-	-	-	-	-	-	-	-	-	+	-	+	-	-	-	-	+	-	-	-	-	-	-
Mannitol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	-	+	-	+	+	+
Sorbitol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	-	+	-	-	+	-
α-Methyl D-Mannoside	-	-	-	-	+	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	+
D-Glucoside	-	-	-	+	-	+	+	+	-	-	-	+	-	-	+	-	+	+	-	+	+	-	-
N-Acetyl-Glucosamine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Amigdalin	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	-	+	-	+	-	-
Arbutin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	-	+	+	+
Esculin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Salicin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+
Cellobiose	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	-	+	+	+
Maltose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lactose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Melibiose	+	+	-	+	+	+	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	+
Saccharose	+	+	+	+	+	+	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	+
Trehalose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Inuline	+	+	+	-	+	-	+	-	+	-	-	+	-	-	-	-	+	-	-	-	+	+	-
Melezitose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	-	+	+	+
Raffinose	+	+	-	-	+	+	+	-	+	-	+	-	-	-	+	+	-	-	+	-	+	-	-
β-Gentiobiose	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	-	+	-	-	-	+
D-Turanose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	+	-	+	+	+	+	+
D-Tagatose	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	-	+	-	+	+	+	+	+
D-Arabitol	-	-	-	-	+	-	-	-	-	-	+	-	-	-	-	+	-	-	-	-	-	-	+
L-Arabitol	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	+	-	-	-	-	+	-
Gluconate	+	+	+	+	+	-	+	-	+	+	+	+	+	+	-	+	-	-	-	-	-	-	+

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