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Effect of temperature during early stage of curing upon cheddar cheese characteristics

Md. Abdul Hamid Miah
Iowa State University

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EFFECT OF TEMPERATURE DURING EARLY STAGE OF CURING UPON CHEDDAR CHEESE CHARACTERISTICS

by

Md. Abdul Hamid Miah, D.V.M.S.

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In Charge of Major Work

Signature was redacted for privacy.

Head of Major Department

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Dean of Graduate College

Iowa State University
Of Science and Technology
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DEDICATION

TO MY DAUGHTER "BEETHY"

INTRODUCTION

For centuries cheesemaking has been considered as a craft or art. In the beginning, people knew little about the changes occurring during the conversion of milk to cheese. This was only natural, as cheesemaking encompasses physico-chemical, enzymatic, and microbial changes at every stage of the operation. Even with the advancement of science and technology, many facets of cheesemaking and curing are still not fully understood. "Proper" cheesemaking involves a multiplicity of factors which are usually empirically controlled by experienced cheesemakers. With growing demand and a competitive economy leading to the development of larger manufacturing units, it has become necessary to standardize processes to obtain a uniformly high-grade product. The evolution of cheese technology is by no means complete and only in recent years has a real beginning been made in gaining insight into areas hitherto accepted by blind faith. Investigations have been conducted on the effect of different types of microorganisms, milk quality, heat treatment of milk, and various curing temperatures on Cheddar cheese quality. Research emphasis thus far has been primarily devoted to the role and contribution of microorganisms to the desirable characteristics of Cheddar cheese. Controlled growth of microorganisms during Cheddar cheese curing is important for proper flavor development.

Cheddar cheese is normally cured at 5-7.5 C. Cheese blocks are usually placed in the curing room while the blocks are still at pressing temperatures. In a large commercial operation, thousands of pounds of cheese are made every day. Cheese blocks are frequently stacked close

to one another in the curing room without any effort to hasten cooling to curing room temperature.

Though not fully recognized by many cheesemakers, it has been empirically shown that the rate of cooling of cheese curd on the press and during the first few days of curing will have a definite effect upon the flavor, body, and texture of Cheddar cheese. A survey of pertinent literature has failed to reveal any previous attempt to study effect of temperature during the early part of curing on the characteristics of cured Cheddar cheese.

A sizable amount of commercial Cheddar cheese is criticized for lack of flavor, high acidity, defective body and texture, fruitiness, and other off-flavors. In many cases, definite differences between blocks from the same vat have been noticed. This probably could be due to ununiformity in temperature control during post-hooping operations. The present study was conducted to explore the probable contribution of temperature control during this period to the differences noted between blocks.

This investigation was designed to determine the effect of curd cooling rates after pressing upon:

(a) The numbers and types of microorganisms in the curd.

(b) Microbial metabolism and chemical changes during curing in terms of sugar dissipation, proteolysis, and lipolysis.

(c) Organoleptic changes during curing.

Cheeses were made on a pilot plant scale simulating commercial practice and blocks from individual vats were subjected to variable

temperature control after hooping. The bacterial population in the blocks was followed along with chemical tests for sugar disappearance, proteolysis, lipolysis, pH, and lactic acid at predetermined intervals. At the end of 3 months and 6 months all cheeses were graded organoleptically to determine differences.

LITERATURE REVIEW

Role of Microorganisms in Development of Cheddar Cheese Characteristics

Cheddar cheese flavor development appears to be related to milk conditions, ripening temperature, and cheese microflora. Microorganisms are believed to produce both desirable and undesirable flavor components in Cheddar cheese (3). Because of the great complexity in their cultural characteristics and other variables, it is difficult to identify the bacteria responsible for the development of the characteristic flavor of Cheddar cheese.

Cheese flora commonly found in Cheddar cheese may be grouped into "starter organisms" and "non-starter organisms".

Starter organisms

Vedamuthu and Reinbold (82) and Reiter et al. (72) emphasized the role of starter organisms in determining the final body, texture, and flavor of cheese.

Dawson and Feagan (21) investigated the fate of starter organisms during manufacture and curing of Cheddar cheese by using a mixed culture of Streptococcus lactis, Streptococcus cremoris, and Streptococcus diacetilactis. Maximum counts attained by S. cremoris were lower than those by S. lactis and S. diacetilactis. Streptococcus cremoris reached its maximum count at milling whereas S. lactis and S. diacetilactis reached this point at half cheddaring. During curing, S. lactis produced the highest count while S. cremoris was the lowest. In curing, S. cremoris disappeared more rapidly than the other two

species. "Non-starter" organisms in Cheddar cheese were found to consist of 70% lactobacilli and 30% micrococci.

Russell (74), in 1896, reported that the bacterial flora of cheese differed markedly from that of milk. Milk always contained a great number of liquifying or peptonizing bacteria, although lactic acid bacteria predominated. In cheese ripening, proteolytic bacteria were found to disappear rapidly while gas-producing bacteria disappeared slowly. But there was rapid growth of lactic acid bacteria until the cheese was partially ripened. After this stage, they gradually diminished in numbers.

Evans, Hastings, and Hart (27) reported four groups of organisms were present in 21 raw milk cheeses sampled. They classified them as Bacterium lactis acidi, Streptococcus, Micrococcus, and Bacterium casei. Considerable variation in numbers and flora was observed, but all good cheeses contained all four groups of organisms listed. Some of the cheeses also contained a small number of spore-forming and liquifying organisms, which had an appreciable detrimental effect on ripening. Pasteurized-milk cheese flora was dependent upon the type of starter used.

A simple method for manufacture of "normal" and "starter-free" Cheddar cheese under controlled bacteriological conditions were reported by Perry and McGillivray (64). A vat was specially designed to follow an aseptic system of operation in making cheese. Milk drawn from cows, which when tested gave no more than 100 colonies per ml was selected. The fresh milk was heat treated at 155 F (68.3 C) for 5 min.

All equipment coming in contact with milk and curd was properly sterilized. The starter used comprised three cultures of S. lactis and two cultures of S. cremoris. Effect of the addition of Lactobacillus plantarum-casei and Micrococcus species also was studied. A few batches of cheese also were made with starter with and without the use of aseptic conditions. Bacteriological examination showed no difference between control cheeses and aseptically-made cheeses. Streptococcus lactis was found to survive longer in cheese than S. cremoris. Addition of micrococci or lactobacilli did not produce any significant difference from the corresponding control cheeses made under non-aseptic conditions. Survival of L. plantarum was not affected by the adventitious flora. In aseptically-made cheeses, the best flavor resulted from the addition of L. plantarum-casei. It was significant that starter alone did not produce Cheddar flavor in aseptic cheeses in the normal time.

In a study of the influence of S. lactis and S. cremoris starter cultures on Cheddar cheese flavor, Perry (63) reported that a S. lactis single strain culture imparted a characteristic abnormal flavor but three single strain S. cremoris cultures produced normal flavor in Cheddar cheese. Intensity of abnormal flavor produced by the S. lactis culture increased with ripening. The "lactis" flavor probably was a direct effect, due to flavor substances produced by S. lactis cells present in young Cheddar cheese.

Vedamuthu, Sandine, and Elliker (83) studied the role of mixed strain lactic starter cultures in Cheddar cheese. Of three commercial

starter cultures, two were found to produce cheeses with open texture and fermented flavor. Significant differences in the rates of protein and sugar degradation between normal and defective cultures were not found. Defective cultures, however, were found to consist of high carbonyl-producing S. lactis and S. diacetilactis strains. The normal culture consisted of low carbonyl-producing S. cremoris strains and a few Leuconostoc strains.

Vedamuthu et al. (84), in their subsequent report, indicated that there were differences in the amounts and types of aldehydes produced by normal and defective mixed strain cultures in 11.0% nonfat milk at 30 C for 24 hr. Both starter cultures produced acetaldehyde, acetone, pyruvic acid, diacetyl, glyoxal, and an alphaketo alkanal. In addition, the normal cultures produced propionaldehyde which was replaced by formaldehyde in the defective cultures. The defective cultures contained more than twice the amount of acetaldehyde, diacetyl, and pyruvic acid than the normal culture.

Non-starter organisms

Robertson and Perry (73) indicated that the strains of microorganisms that are numerically important in mature cheese need not necessarily be responsible for flavor development. They indicated that the distinct flavor of Cheddar cheese could be enhanced as a result of the addition of a small amount of a culture of lipolytic Micrococcus to cheese milk.

Irvine, Beach, and Burnett (48) studied the relationship between numbers of bacteria present in milk supplies, subsequent microbiological

changes, and flavor quality of raw milk Cheddar cheese. Nineteen of 20 vats of cheese were found to be of first grade when first examined but after 20 to 26 weeks of ripening, seven declined to second grade. The total count on the original milk producing grade 1 cheese averaged 3.38 million per ml while milk averaging 35.8 million per ml produced defective cheese. No significant differences were observed in the Mannitol Salt Agar count (staphylococci), and Violet Red Bile Agar count (coliforms) in good and defective cheeses. Organisms tolerant to 3.75 ppm of brom cresol purple (nonlactic group) increased to an average count of 70.2 million in good cheese but to 310.8 million in defective cheeses at 3 weeks of age. The poor flavored cheese also contained a considerable number of alkaline-producing bacteria when examined at 1 week of age.

Bacterial flora trends during manufacture and ripening of White cheese were reported by Rašic (67). After 1 day of manufacture, streptococci comprised 96% of the total bacterial flora, 60% after 4 weeks, and 50% after 6 weeks. Lactobacilli were maximal in number after 4 weeks, constituting 30% of the bacterial flora. After 6 weeks, lactobacilli composed 50% of the flora. Micrococci and gram-negative bacteria were found to multiply during manufacture but disappeared rapidly after curing.

During a flavor study of 38 Cheddar cheeses, Johns and Cole (49) determined the number of lactobacilli in milk and cheese at various stages of ripening. Lactobacilli were found to multiply rapidly during the first few days of curing and a maximum level was attained at 3-6

months which declined appreciably at 1 year. Flavor intensity was found to be positively correlated with the number of lactobacilli in milk and cheese during ripening. Commercial cheese with the highest degree of flavor contained large numbers of this genus.

A similar observation was made by Hucker (46), in 1922, in his experiment with 37 samples of Cheddar cheese from 25 different factories. Better grades of cheese contained a different and distinct flora than poor cheese. Better grade cheese contained large numbers of lactobacilli and S. lactis while the poorer grade cheese contained large numbers of spore formers and gram-negative rods.

Harding and Prucha (38) isolated over 300 cultures from agar plates of nine Cheddar cheeses representing four first-class commercial factories. Qualitative studies indicated that Bacterium lactis acidi was the only form constantly found in Cheddar cheese. In addition, they also found four other groups of microorganisms: acid liquifiers, gas-producing forms, an inert group, and yeasts. Hastings, Evans, and Hart (44) described two types of organisms which occurred in large numbers in Cheddar cheese. These were Bacterium lactis acidi which grew in large numbers until the sugars were completely fermented and Bacillus bulgaricus, which grew after the fermentation of sugars had been completed. In the early part of ripening, Bacterium lactis acidi predominated while in the late ripening period, Bacterium bulgaricus, which in many instances constituted over 90% of the lactic flora, predominated. Coccus types also were found to predominate at one time or other in 11 of the 13 cheeses sampled.

Franklin and Sharpe (31) studied the bacterial flora of cheese made from different milk and the use of different heat treatments, starters, and their effect on flavor. The non-starter lactic acid flora consisted of Lactobacillus casei, Lactobacillus plantarum, Lactobacillus brevis, Lactobacillus buchnerii, Pediococcus species, and Leuconostoc species. Lactobacillus casei were found to be numerically most important, occurring much more frequently than any other species, especially in severely heat-treated milk. The groups of cheeses with lowest average flavor scores contained the lowest number of non-starter lactic acid bacteria. The severely heat-treated milk (160 F for 17 sec), being low in non-starter lactic acid bacteria, led to lower scores than that of less severely heat-treated milk (145 F for 17 sec), containing high non-starter lactic acid bacteria. A positive correlation was found between the number of lipolytic bacteria and desirable flavor.

Clark and Reinbold (12) obtained a total of 1,162 isolates from 50 7-day-old cheese from 11 Iowa cheese-manufacturing plants. They characterized these microorganisms as: 578 enterococci (50%); 149 micrococci (13%); 126 lactic streptococci (10.5%); 110 miscellaneous gram-positive rods (9%); 93 miscellaneous associated bacteria (8%); 67 lactobacilli (6%); 34 miscellaneous gram-negative rods (3%); and 5 other miscellaneous groups. It was not possible to trace the source of enterococci back to lactic starter cultures used.

Dacre (18) reported that a month-old Cheddar cheese, made from flash pasteurized milk contained 6% streptococci, 61% lactobacilli,

and 33% pediococci. He did not find any streptococci in cheese ripened for 2 months at room temperature. By the end of the third month the cheese was found to contain 75% lactobacilli and 25% pediococci. This flora persisted through the ninth month of ripening.

Crossley (14) and Yale and Marquardt (89) studied the coliform organisms associated with commercial manufacture and storage of Cheddar cheese. Crossley (14) isolated 116 cultures from 2-day to 12-month-old Cheddar cheeses of commercial factories which used pasteurized milk. Of the 116 cultures, 99 were Escherichia coli types, 13 were Aerobacter aerogenes types, and 4 were intermediate types. Yale and Marquardt (89) observed that coliforms survived over 12 months in cheese made from poor quality milk but with better quality milk, containing fewer coliforms, these bacteria survived for 3-6 months only.

In his subsequent report, Crossley (15) indicated that coliform organisms were found in 36.5% of samples of pasteurized milk delivered to the cheese vats and 35.1% of all samples of bulk factory starters. One hundred per cent of the cheeses tested contained coliform after 1 week, 93.3% after 1 month, 78.9% after 2 months, and 58.8% after 3 months.

Chemical Aspects of Cheddar Cheese Ripening

Excellent comprehensive reviews on Cheddar cheese ripening were reported by Mabbitt (55) and Marth (56). Reiter et al. (72) made a critical assessment on the studies on cheese flavor with respect to

defined bacterial flora and enzymatic breakdown of cheese curd such as: glycolysis, proteolysis, and lipolysis.

Flavor with respect to bacterial flora

Flavor components of Cheddar cheese include carbonyl, nitrogenous and sulfur compounds, fatty acids, alcohols, esters, salts, and unidentified cheese fractions. Many efforts have been made to duplicate Cheddar flavor by adding some of these compounds individually or in mixtures to fresh cheese curd with little real success. Evidently, Cheddar flavor development is related to conditions of milk, ripening temperature, and cheese microflora. Marth (56) indicated that homofermentative lactic streptococci contribute flavor to finished cheese by producing acids and enzymes which liberate nitrogenous compounds from protein. The streptococci also may degrade amino acids ultimately producing aldehydes.

Reiter et al. (71) compared the flavor, free fatty acids, and bacterial flora of commercial Cheddar cheese and experimental cheese made under aseptic conditions. Experimental cheeses were made (i) with ∂-gluconic acid lactone instead of starter, (ii) with starters only, and (iii) with starters and added flora derived from the curd of commercial cheeses. In all cheeses, lactobacilli increased in numbers during maturation; the amount of acetic acid produced was influenced by lactobacilli and the starter organisms. Commercial strains containing Streptococcus diacetilactis produced more acetic acid. There were variations in the initial levels of butyric acid related to season and milk source. Further increase in the fatty

acids occurred in cheese made with starters but no increase in fatty acids content was observed in the cheese made with ∂-gluconic acid lactone. This indicated that the lactic acid bacteria were weakly hydrolyzing the milk fat. Flavor trials indicated that Cheddar cheese flavor was only present in the commercial cheese, cheese made with reference flora, and starters only. No flavor was developed in cheese made with ∂-gluconic acid lactone.

Protein degradation and Cheddar flavor

Studies on the effect of acido-proteolytic organisms and curing temperatures on ripening of pasteurized milk Cheddar cheese were conducted by Deane (23). No significant differences were observed in the rate of production of soluble nitrogen or volatile fatty acids between Cheddar cheeses made with a 0.75% inoculum of commercial starter plus 0.5% of a pure culture of acido-proteolytic micrococcus AP5A or AP No. 1 and Cheddar cheese made from commercial starter alone during the 24-week ripening period. Addition of a 0.1-0.5% inoculum of Streptococcus liquifaciens resulted in the development of a bitter flavor, making the cheese quite unpalatable. Ripening at 65 F, instead of 50 F, increased ripening and rate of flavor development.

Dahlberg and Kosikowsky (19) reported that increased development of tyramine in Cheddar cheese produced good characteristic Cheddar flavor. Pasteurized milk was used to make American Cheddar cheese with starters practically free from tyramine-producing bacteria. After ripening for 6 months at 40 (4.4), 50 (10), and 60 F (16 C) the tyramine content of the cheeses was 3, 12, and 17 µg/g. The flavor

of the cheese was considered to be mild to mild-medium lacking characteristic Cheddar flavor. When Streptococcus faecalis starter (a special rapid acid-producing strain) was used alone, tyramine content after 6 months of ripening at 40 (4.4), 50 (10), and 60 F (16 C) was 18, 108, and 315 µg/g. Flavor intensities were mild, medium, and medium +, respectively. The use of commercial lactic cultures and Streptococcus faecalis in pasteurized milk caused the greatest development of tyramine. After 6 months of ripening at 40 (4.4), 50 (10), and 60 F (16 C), the tyramine content was 85, 428, and 1172 µg/g, and flavor intensity was mild +, medium +, and sharp -, respectively, with good characteristic Cheddar flavor.

Vakaleris et al. (79) measured the soluble nitrogen and formol nitrogen content of Dariworld and Cheddar cheese during a 5-month ripening period to see if the rate of proteolysis affected cheese consistency. Each variety of cheese was made with normal (pH 5.45) and abnormally high pH 6.45. Proteolysis rates were the same in the normal lot of both varieties and in Cheddar with high pH. Both the cheeses with high pH were hard and woody in consistency and did not improve as the amount of soluble and formol nitrogen increased with ripening. The rapid loss of curdiness in sweet Dariworld cheese was not considered to be due to unusually fast protein break down. It was concluded that physico-chemical changes caused by prolonged exposure of the curd to sodium chloride brine during the making operation of this cheese and the relatively high moisture content

were responsible for its characteristic consistency. Increase in nitrogen content was alike in all cheeses.

Stadhouders (77) investigated the possible source of proteolytic enzymes which hydrolyze protein during the ripening of Dutch cheese. Rennet did not form amino acids directly from protein but was found to stimulate the production of amino acid by breaking down proteins into polypeptides. An overdose of rennet during cheesemaking did not affect the production of amino acid. Enzymes from the starter streptococci were found to hydrolyze protein to amino acids. The protease enzyme of milk and the enzymes from Achromobacteriaceae, Pseudomonas species, lactobacilli, and enterococci were shown to be of minor importance in the break down of protein to amino acids in cheese.

Hlynka, Hood, and Gibson (45) reproduced rancid and other less accurately defined flavors in Cheddar cheese by addition of commercial lipase to cheese milk. Greater amounts of rennet, pepsin, or other proteolytic enzymes retarded rancidity and developed a better flavor in cheese compared to corresponding cheese with less rennet.

Vakaleris and Price (80) reported a rapid method for measuring cheese ripening. They indicated that the flavor intensity of cheese had a linear relationship with the soluble tyrosine content of cheese. The degree of proteolysis and flavor intensity was measured by the concentration of soluble tryptophane and soluble tyrosine content.

Bullock and Irvine (9) also estimated the progress of protein breakdown in cheese ripening. They identified 18 amino acids in

Cheddar cheese. The concentration of amino acids increased with age. Raw milk cheese contained higher amounts of amino acids than the pasteurized milk cheese.

Emmons, McGugan, and Elliott (26) made 99 vats of Cheddar cheese with 11 single strain starter cultures in a series of three replicates of three treatments for each culture; sweet, normal, and acid cheese with pH 6.41, 6.04, and 5.63, respectively, at whey draining. Seven of the cultures produced bitterness of varying degrees. The manufacturing procedure did not have any significant effect on bitterness. The remaining four did not produce bitterness under any treatment.

Czulak and Shimmin (16) obtained cell-free extracts from two single strain cheese starter cultures to compare proteolytic activity. They observed an increase in production of amino nitrogen by the culture which was reported as "non-bitter" over the culture which was reported as "bitter culture". They supported the hypothesis that the bitter flavor was due to the insufficient break down of polypeptides to amino acids.

Pette (65), in his review on the flavor development of Cheddar cheese, reported that casein break down was affected by bacteria and rennet. In addition to bacterial enzymes, rennet proteinase was important in this respect, but it did not decompose casein further than to polypeptides. The formation of amino acid then must be due to bacterial activity.

Sugar Breakdown in Cheese

A survey of literature indicates that sugar disappears in cheese quickly within 2 weeks after manufacture (57, 58, 66, 76, 81, 88).

Van Slyke and Price (81) and McDowall and Dolby (57) indicated that at initiation of pressing, lactose concentration in the cheese varied from 1.7 to 0.77% and after 2 weeks only a trace quantity of lactose was present if at all.

Dolby, McDowall, and McDowell (24) investigated the effect of addition of lactose in milk and curd and its effect on body and flavor. In most cheese batches, lactose almost completely disappeared in 2-3 weeks. Increasing lactose produced an acid cheese.

Anderson, Nilsson, and Sjöström (2) reported that cooking at 47 C, rather than 37 C, retained more sugar in Svecia type cheese.

Sjöström (76) investigated the velocity of lactose break down in Herrgård-cheese, Prästost, Svecia, Cheddar, and Camembert cheese. Glucose was not detected in any of the cheeses. Two cheeses with the highest cooking temperature, Herrgård and Prästost, contained galactose but not lactose. Galactose in the cheese remained a relatively long time (22 days). In hard cheese made with a lower cooking temperature, the sugar break down was found to go faster and all sugar disappeared in about 3 days.

Raadsveld (66) also investigated the break down of lactose and the presence of galactose and glucose in Dutch type cheese. After 1 day of standing (Gouda type cheese), amounts less than 0.1% lactose + galactose were present. After brining, the lactose had disappeared

and after 7-day brining, the last trace of galactose also had disappeared. Warming the contents of vats to 38 C or above resulted in higher sugar content. Neither the amount of starter (0.3 to 0.6%) added to cheese milk nor the temperature during standing (13-20 C) influenced the sugar content of cheese.

Fagen, Stine, and Hussong (28) used a paper chromatographic method for the quantitative determination of sugar(s) in cheese. In raw milk cheeses, reducing sugars disappeared in 25 days but in pasteurized milk cheese the sugars were retained up to 53 days. Galactose remained for a longer period in pasteurized milk cheese. Concentration of salt was found to influence the amount of sugars retained in the cheese. At 0.78% salt, the sugar disappeared in 7 days and at 2.14% salt, the sugars were observed up to 63 days.

Nilsson and Guldstrand (58) separated lactose, glucose, and galactose as borate sugar complexes on a Dowex-1 borate column by elution with 0.22 M potassium tetraborate. They observed that lactose had disappeared after 1 day in Herrgård cheese.

Mabbitt reported (55) that sugars remaining in Cheddar cheese after development of starter bacteria might control the growth of the adventitious flora of cheese. He suggested that fermentation of residual sugars by heterofermentative lactobacilli such as Lactobacillus brevis might lead to the production of acetic acid, ethanol, glycerol, and mannitol. Under normal conditions, the quantities of such products accumulated would be minute, due to the limited quantity of substrate,

but the level of alcohol may be enough to form esters by chemical combinations with fatty acids.

Acidity or pH of Cheddar Cheese During Curing

Too much or too little acidity may result in serious defects in Cheddar cheese. Proper manufacturing technique for Cheddar cheese requires controlled acid production in all stages of manufacture. The hydrogen ion concentration of cheese furnishes an indication which can not be measured in terms of titratable acidity.

Van Slyke and Price (81) and Wilster (88) indicated that the pH was lowest in cheese on the third or fourth day of pressing. A mild, low-acid cheese several days old might have a pH from 5.1 to 5.3. A medium-acid cheese several days old might have a pH of about 5.0.

Brown and Price (8) reported the best cheese had the highest pH when fresh but lowest after curing. In general, there was a gradual decrease in pH during the early stages of curing. In this study, conducted over 2 years, it was observed that the pH of good quality cheese at pressing was 5.38 which decreased rapidly to pH 4.99 on the third day and then gradually increased to pH 5.5 in 24 months.

Vakaleris et al. (79) in a comparison of Cheddar cheese with Dariworld cheese indicated that the pH of normal (pH 5.45) Dariworld and Cheddar cheese decreased during the first 10 days from 5.45 to 5.1 and then gradually decreased until the end of the observation (150 days). The low-acid Cheddar lot (pH 6.45) decreased slowly in pH after making until 60 days of curing. After this period, they were

only 0.1 to 0.2 pH units higher than that of normal Cheddar or normal Dariworld cheese.

Dolby, McDowall, and Riddet (25) reported that in most cases, since lactose in Cheddar cheese was fermented in 7-10 days, there was no further acid production thereafter. Comparisons of grading score with pH value showed a definite falling off in average grading score with cheese of high pH value and a less sharp decline at lower pH values. With cheese of low titratable acidity there was a rapid decrease in average grading score as acidity decreased. But very little decrease in grading score was observed with high acidity. A direct comparison was made between acidity and pH. It was found that as the pH rose above 4.9, there was a rapid fall in acidity but with a decrease in pH value below 4.9, there was little corresponding increase in acidity.

So, the value of titratable acidity determination in characterizing cheese was considered to be limited because of the buffer capacity of cheese.

Irvine (47) determined the pH value of 375 samples of Cheddar cheese from the different cheesemaking districts of Ontario, Canada. Except for a small number of samples, the range of pH was between pH 5.1-5.25. Cheeses were graded higher within this range of pH than those outside this range. Color of the cheeses was found to be low in the case of high-acid cheese.

Relation of Fatty Acids and Other Volatile Compounds to Flavor of Cheddar Cheese

Grag and Verm (34) determined various constants (Reichert-Meissl and Krisner Values) in fats extracted from cows' milk and buffalo milk cheese. Presence of lower molecular fatty acids was considered to be important in producing flavor compounds in cheese.

Ohren (59) and Ohren and Tuckey (60) compared the flavor and free fatty acids content of Cheddar cheese prepared from milk containing 0-4.5% fat and with varying bacterial counts. Normal pasteurization of milk with a high total plate count resulted in cheese with a better flavor compared to same milk when used raw. A typical Cheddar flavor was associated with the presence of limited amounts of free fatty acids and acetic acid. Fermented, unclean, and whey-taint flavor was associated with high contents of C_{10}, C_{12}, and C_{14} fatty acids. Free fatty acids were increased with an increasing bacterial count of the raw milk from 3,000 to 10^7/ml but the quality of flavor decreased.

There have been many efforts to identify and characterize the compounds responsible for characteristic Cheddar flavor. Dacre (17) separated volatile fatty acids, ethyl alcohol, butyraldehyde, ethyl acetate, and ethyl butyrate to be main components from the steam distillate of Cheddar cheese. He failed to reproduce the typical Cheddar flavor by adding the above components to fresh cheese curd.

Calbert and Price (10) concluded that diacetyl in amounts smaller than .05 mg/100 g cheese was an essential part in the flavor complex

of Cheddar cheese, although beyond this level it was associated with adverse criticism.

Patton, Wong, and Forss (62) identified volatile flavor components to be dimethyl sulfide, ethanol, acetone, diacetyl, 2-butanone, 2-heptanone, and 3-hydroxy butanone.

Kristoffersen, Mikolajcik, and Gould (54) reported they could reproduce fuller Cheddar flavor by the addition of glutathione (10-100 ppm) to fresh curd slurries.

Two Cheddar cheeses were analysed for aldehydes and ketones during ripening by Harvey and Walker (43). One-day-old cheese contained acetaldehyde and acetone together with traces of butan-2-one. As the cheese matured, heptane-2-one undecan-2-one appeared and the concentration of all these compounds progressively increased.

Kristoffersen, Gould, and Purvis (50, 51, 52) observed that the concentration of active SH-groups in cheese manufactured from raw milk or from milk heated at 143 F (61.7 C) for 5 and 30 min, and 155 F (68.3 C) for 15 min, were related inversely to the severity of the heat treatment of the milk. The intensity of characteristic Cheddar flavor was related to the concentration of active SH-groups of the cheese. Kristoffersen, in a later study (54), indicated that free fatty acids and hydrogen sulfide were important as ripening products of Cheddar cheese. Desirable flavor occurred only when these compounds were present in a definite interdependent concentration. It also was found that when active SH-groups failed to appear early in the ripening

period and to reach a relatively high concentration during ripening, the cheese lacked flavor.

Temperature with Relation to Cheddar Cheese Quality

Temperature during the early stage of manufacture

Call and Price (11) indicated that the quality and uniformity of Cheddar cheese were improved by pasteurization of milk at 160 F (71.1 C) for 15.6 sec.

Overcast, Jarman, and Albrecht (61) and Babel (5) compared different cooking temperatures for manufacture of Cheddar cheese. The rate of acid development was twice in the curd cooking at 98 F (36.7 C) compared to 102 F (38.9 C) from draining to milling. The average time from draining to milling was 65 min when cooked at 98 F (36.7 C) and 125 min when cooked at 102 F (38.9 C). Flavor quality of cheese did not show any difference because of the cooking temperature. Babel (5) suggested the time required to attain desired milling acidity could be reduced if a longer period of ripening was allowed before setting.

Bevan et al. (6, 7) conducted a study on the texture of cheese manufactured at high temperature (43.3 C) using *Streptococcus thermophilus* or *Streptococcus durans*. This cheese was compared with cheese manufactured by a conventional method using *S. lactis* or *S. cremoris*. Cheese made by high temperature method eventually developed a open texture.

Dawson and Feagan (22) found cheese to be unsatisfactory (open texture) when cooked at high temperature (43.3 C) during manufacture.

It was suggested that this openness was due to the fermentation of residual sugars by non-starter organisms.

Temperature during curing

Little information is available concerning the effect of temperature on cheese quality during the early stage of curing. Cheese blocks are usually taken out of the press between 70 (21.1) to 90 F (43.3 C) and placed in the curing room. Conochie and Sutherland (13) indicated that cheese graders commented on the uneven characteristics of different cheeses from the same vat. In a particular instance, it was observed that there were some differences which could be attributed to the block stacking of the cheese on the pallets and block stacking of the pallets soon after the warm cheese was packed. The temperature of the warm pressed cheese varied from 70 (21.1) to 85 F (40.6 C) and the block stacked cheese, even at 70 F (21.1 C), took 11 days for any inner temperature to come to a curing room temperature of 50 (10 C) ± 2.5 F. If a spacing of 1¼ in. was provided between the blocks, the cheese reached room temperature in 4 days. Although no attempt was made to demonstrate any differences in Cheddar cheese characteristics due to these discrepancies in cooling rates between blocks of the same vat of cheese, the authors indicated that such differences could attribute to subsequent differences in maturing rates.

Effect of temperature on the mechanical properties of cheese during ripening was studied by Wearmouth (85, 86). The Standard Ball Compressor revealed variations in physical properties of a cheese when the temperature of that cheese was changed. Influence of temperature

variation on the firmness of cheese was marked. When the ripening temperature was kept constant, the firmness of the cheese increased sharply during the early stages of ripening and thereafter, firmness continued to increase at a uniform but less rapid rate. This effect was observed on cheese made from milk of normal fat content and also on cheese which had been made from milk deliberately reduced in fat content. A rise of temperature caused a decrease in firmness of the cheese. In a comparison, cheese ripened at constant temperature was preferred by a panel of trained judges over cheese ripened at fluctuating temperatures.

The literature is quite voluminous on the effect of different curing temperatures upon maturing. Previous to 1900, factory practice was to cure Cheddar cheese at room temperature without refrigeration (81). Cheese cured below 55 F (12.8 C) invariably gave good flavor. It was recognized in 1900 that cheese cured below 60 F (15.4 C) received at least 5 points higher in flavor scores and 2.5 points higher in texture compared to cheeses which were cured at 65 F (18.5 C) or above.

Babcock et al. (4) in 1901 concluded that curing cheeses from 33-50 F (0.75-10 C) would control bitter and other undesirable flavor defects. More uniform flavor, however, was obtained when cured from 40-50 F (4.5-10 C). They also indicated that the improvement in body and texture was marked in cold-cured cheese compared to conventional high temperature curing. In the initial stage, the cold-cured cheese had a tendency to be curdy but as the ripening progressed, curdiness disappeared. Keeping quality of cheese also was greatly improved by

cold-curing. Color in cold-cured cheeses was found to be more uniform. In general, cheeses cured from freezing point to 50 F (10 C) were of better quality than those ripened at higher temperatures.

There have been many attempts to shorten the ripening period by increasing the curing temperatures. Freeman (32, 33) using 5-lb., wax-coated Cheddar cheese blocks observed that higher ripening temperatures than those normally used increased the rate of flavor development. Best results were obtained at a temperature of 60 F (16 C) for the first 4-6 weeks or 75 F (24 C) for the first 3 weeks, followed by 40 F (4.4 C) for the remainder of the 12-week ripening periods. The rate of flavor development was improved when a mixed starter of S. lactis and S. faecalis was used.

Hansen (37) indicated that Cheddar cheese made from good quality milk developed more flavor in 3 months at 60 F than did cheese at 40 (4.4) and 50 F (10 C) for 6 months. But increasing the ripening temperature caused more loss in weight.

Reichert and Downs (68) reported that cheese ripened better at 45 F (7.5 C) than at a higher temperature (18.3 C). The body of the cheese was better when cured at 45 F (7.5 C) rather than 65 F (18.3 C).

Foster et al. (30) have indicated that the rate of flavor development can be increased by increasing the temperature up to a certain limit, but at the same time the flavor defects are also accelerated.

Hammer and Babel (35) have indicated that curing temperature of cheese made from raw milk should be kept low to prevent the action of adventitious flora causing undesirable flavor defects. The use of

high curing temperature in pasteurized milk cheese may prove useful in accelerating ripening process but underheated or recontaminated pasteurized milk might cause rapid development of defects in cheese. They have pointed out some definite advantages in curing cheese below 45 F (7.5 C):

> (1) There is no loss from insects, (2) Shrinkage is kept at a minimum, (3) Special humidity control in commercial storage is seldom necessary, (4) Maximum use of storage space is obtained, and (5) Cheese can be held 1 to 2 years without handling with minimum deterioration in grade.

Reinbold (69) emphasized the importance of curing temperature to the final quality of cheese. He suggested that any changes in the physical and chemical factors involved in the curing could upset the normal bacterial, chemical, and enzymatic action in cheese.

Davis (20) also has given a great emphasis on the temperature control of curing room. A better result is obtained when cheese is cured at low temperatures (5-7 C).

EXPERIMENTAL METHODS

Survey of Commercial Plants

Before designing the experiment, nineteen commercial Cheddar cheese plants located in Iowa, Wisconsin, New York, and South Dakota were surveyed to determine curd-handling practices immediately beyond the hooping stage. A letter with an accompanying questionnaire (Appendix, p. 131) was sent to each plant manager. After analyzing the answers that were received, an experimental design was established to include what appeared to be extremes in curd-handling procedures in commercial practice.

Manufacture of Cheese

More than 16 batches of Cheddar cheese were manufactured on a pilot plant scale in a 5000-lb. vat, or in small 400-lb. vats, in the Dairy Products Laboratory at Iowa State University. A total of nine vats of Cheddar cheese were made in 5000-lb. vats simulating commercial conditions. Six of these vats were made from Iowa manufacturing-grade milk[1] of which 3 had milling acidities above 0.60 per cent and 3 with normal-acidity i.e. below 0.60 per cent. Hereafter these cheeses will be designated as high acid and normal acid cheeses respectively. Three were made from grade-A milk as defined in the U. S. Public Health Grade 'A' Pasteurized Milk Ordinance, 1965 (78).

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[1]Iowa grading law for milk used for manufacturing purposes, Chapter 194, Iowa Department of Agriculture.

Time and operation schedules for the manufacture of cheese were followed according to practices outlined by Wilson and Reinbold (87). The milk was heated to 62.8 C for 17 sec and stored at 4.4 C, usually overnight, before use.

Starters used

A commercial lactic culture (No. 253, Hansen's Dri-Vac, Chr. Hansen's Laboratory, Milwaukee, Wisconsin) was used in the manufacture of the first six lots of cheese made in a 5000-lb. capacity vat with manufacturing-grade milk and three lots in 400-lb. capacity vats with grade-A milk. Three more lots of cheese were made from grade-A milk in a 5000-lb. capacity vat with mixed-strain culture obtained from Oregon State University. This culture was known to produce fruity flavor defects (83, 84). Hereafter these cheeses will be referred to as experimental fruity cheese. Three lots of cheese also were made in 400-lb. vats with a culture obtained from Dr. Emmons of Canada. This culture produced a bitter flavor defect in their investigations (26). These cheeses will be designated as bitter cheese in later discussions. Still one more lot was made in the small vats with Hansen's lactic-culture No. 253 plus an equal amount of culture made with coliform organisms isolated from natural Cheddar cheese.

Cheese treatment

Post-hooping temperature changes occurring in each lot of cheese were closely followed from the time of pressing with a thermocouple connected to a continuous recorder system[1].

[1]Honeywell, Inc., Philadelphia, Pa.

The experiment was designed as a 2 x 2 factorial with two pressing times (4 hr and 20 hr) and two cooling rates (rapid and slow). After 4 hr of pressing, four 40-lb., rectangular blocks were taken from the press, wrapped in Marathon foil-cello-foil wrappers[1] and sealed with a Flexpress, model R.L.100[2]. Two of the blocks were submerged in a brine tank at 4.4-7.2 C for rapid cooling. The other two were placed in the curing room in hardboard boxes at 7.2 C. Temperature changes were recorded continuously for the first 8 hr and then at certain intervals based on earlier studies. After 20 hr of pressing, four more blocks of cheese were taken from the press and were treated in the same way (Appendix, Fig. 10).

Microbiological and Chemical Examination of Cheese

Sampling

Samples for bacteriological analysis were taken aseptically and plated immediately. Trier holes in the cheese blocks were properly sealed to prevent mold growth. These samples were taken immediately after the two pressing periods, on the 4th day, 8th day, 12th day, 21st day, 30th day, 60th day, and 90th day.

The pH also was determined immediately after each sampling. Portions for lactic acid and reducing sugars were collected at the same intervals but were frozen and stored at -10 C until analyses were made. For determination of proteolysis and total free fatty acids, samples

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[1]Marathon, Division of American Can Company, Neenah, Wisconsin.

[2]D. L. Manufacturing Company, W. De Pere, Wisconsin.

were collected after the two pressing periods and at monthly intervals for 3 months. In this case also, samples were stored at -10 C until use.

Cheese judging

All cheeses were judged by a panel of trained judges after 3 and 6 months of curing. The cheeses were judged in two ways. Since it was the object of the experiment to determine if there was any difference in cheese characteristics due to different pressing times and cooling rates, a paired comparison was made among four different treatments (Appendix, p. 133). All four treatments of the same vat of cheese were arranged in pairs of six possible combinations. Each pair was placed at random in a row with necessary code numbers. This ensured unprejudiced judging of samples. The judges were asked to record any difference in flavor, body and texture, and color between individuals in pairs. This comparison on all batches of cheese was done only after 3 months of curing. The cheeses also were scored after 3 and 6 months according to the American Dairy Science Association Cheddar cheese score card. All judges were asked to indicate their preferences among four different samples representing four treatments in each batch of cheese.

Microbiological analysis of cheese

Bacterial counts included: (a) total bacterial count, (b) enterococcus count, and (c) gram-negative bacterial count. Eleven grams of cheese were weighed into a sterile, tempered (45 C) Waring

blendor and macerated for 2 min with 99 ml of 2.0% sterile, tempered (45 C) sodium citrate as recommended by Standard Methods for the Examination of Dairy Products (1). The next higher dilution (10^{-2}) was made with 11 ml of well mixed sample from the Waring blendor to a 99 ml ± 2-ml dilution blank (1). Further dilutions were made as usual with a 1-ml sample. A series of dilutions in duplicates were plated for the total bacterial count in Eugonagar[1] and incubated at 21 C for 7 days; for the gram-negative bacterial count in Violet Red Bile Agar[2] at 32 C for 18-24 hr; and for the enterococcus count in the Citrate Azide Agar of Saraswat, Clark, and Reinbold (70, 75) at 37 C for 72 hr.

pH determination

Ten grams of cheese were weighed into a mortar and ground into a homogeneous paste with 2 ml deionized, double distilled water. The pH was determined using a Beckman[3] model H2 glass electrode pH meter.

Lactic acid determination

Lactic acid was determined by a method adopted from Harper and Randolph (41). Ten grams of cheese were weighed into a Waring blendor and were macerated for 6 min with 90 ml of distilled water. Two 10-ml portions of sample were separately diluted to 100 ml in volumetric

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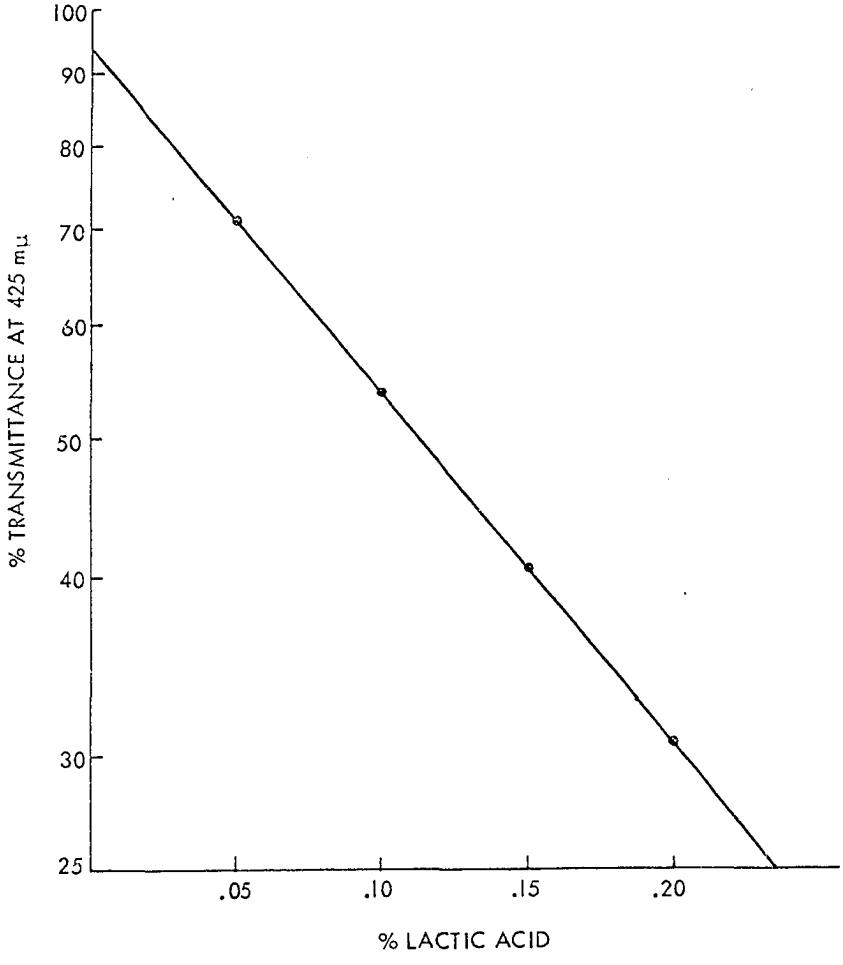
[1]Baltimore Biological Laboratory, Baltimore, Maryland.

[2]Difco Laboratories, Detroit, Michigan.

[3]Beckman Instrument Inc., Fullerton, California.

Fig. 1. Relationship between per cent transmittance and concentration of lactic acid


100
90
80
70
60
50
40
30
25
% TRANSMITTANCE AT 425 mμ
.05
.10
.15
.20
% LACTIC ACID

flasks in duplicates. Twenty-five milliliters of sample from each volumetric flask were placed in separate 125-ml Erlenmeyer flasks. The following reagents were added, in order, with thorough mixing after each addition:

(a) Ten milliliters of barium chloride solution (98.8 g c.p. crystals per liter).

(b) Five milliliters of zinc sulfate solution (225 g c.p. crystals per liter).

(c) Five milliliters of 0.66 N sodium hydroxide.

Thoroughly mixed samples were filtered through Whatman No. 40 filter paper. Then, 10 ml of filtrate were transferred into a clean, standardized spectrophotometer cuvette. One milliliter of 1.0% freshly prepared ferric chloride was added to develop color and was mixed with a Vortex-Genie[1] for 5 sec. The per cent transmittance was determined at 425 mμ using a Coleman Model 11 spectrophotometer[2], against a reagent blank prepared in the same way using 25 ml of water for samples.

The amount of lactic acid was determined directly from a standard curve (Fig. 1). A standard curve was prepared by adding a known amount of lithium lactate (c.p. grade) in distilled water and then calculating total lactic acid.

Measurement of proteolysis

The extent of proteolysis was determined by measuring total protein by the dye binding technique of Hammond, Seals, and Reinbold (36), using

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[1]Scientific Industries, Inc., Queens Village 29, New York.

[2]Coleman Electric Co., Inc., Maywood, Illinois.

orange G[1]. The dye solution (1 mg/ml) was prepared by weighing 1.0638 g orange G (assayed 94% dye, dried for 3 hr) and 6.3020 g oxalic acid into a 1000-ml volumetric flask and diluting to volume with distilled water.

One hundred and fifty mg cheese were weighed from the interior of a plug into a 15 x 2 cm test tube. Fifteen milliliters of dye solution were added with a volumetric pipette. The sample was then homogenized in a specially designed test tube homogenizer for 1 min. The tubes and their contents were held overnight and were centrifuged at 2,500 rpm for 15 min. The absorbence was read in a Beckman model D. U. spectrophotometer using a "flo-thru" curvette at 475 mμ against a blank. The blank was prepared for the entire experiment by diluting the original dye solution with an equal volume of distilled water. Results were expressed as per cent proteolysis. The value at 4 hr pressing time was taken as showing no measurable proteolysis.

Determination of total free fatty acids

Total free fatty acids were determined by the silica gel column method described by Harper, Schwartz, and Hagarawy (39, 40, 42).

Preparation of reagents

(a) Chloroform (technical grade) was washed four times by shaking with distilled water to remove all traces of alcohol.

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[1]Matheson Coleman and Bell, Matheson Co., Norwood, California.

(b) Five per cent n-butanol was prepared by adding 5 ml n-butyl alcohol in 95 ml of washed chloroform.

(c) Silicic acid (Mallinckrodt No. 2847) was prepared by washing with distilled water, allowing to stand for 10 min and decanting the supernatant. This procedure was again repeated and was followed by drying at 100 C for 48 hr.

(d) Buffer (pH 6.5) was prepared by mixing 2 M KH_2PO_4 and 2 M K_2HPO_4.

(e) Alcoholic 0.01 N KOH was prepared in absolute alcohol.

(f) Phenol red indicator was prepared by grinding 100 mg phenol red with 0.1 ml 1 N KOH and was then diluted to 100 ml with absolute alcohol.

(g) Silicic acid stock solution. Five grams of dry silicic acid, mixed in 3 ml of 2 M pH 6.5 phosphate buffer were slurried with 20 ml of washed chloroform. The stock solution was stored in a tightly stoppered brown bottle.

Preparation of column and extraction of fatty acids

The column consisted of two sections:

I. Lower section Twenty-five milliliters of well mixed silicic acid stock solution were added to the lower half of the column for each analysis immediately before adding the upper half.

II. Upper section Sufficient 20.0% H_2SO_4 (0.2 to 0.4 ml) was added to 5 g cheese to adjust the pH to 1.7 to 2.0. This was determined experimentally for each batch at each sampling period. The final volume was adjusted to 3.0 ml in a 500-ml mortar with distilled

water. Then, 10 g dry silicic acid were added and ground thoroughly with the sample. The well mixed sample was then slurried with 5.0% n-butyl alcohol in chloroform and was transferred quantitatively on the top of the lower column. The column was then attached to a 250-ml suction flask and vacuum was applied such that the solvent would flow through the column at the rate of 25 ml/min. One hundred and fifty milliliters of eluant were collected.

The total free fatty acids in the eluant were titrated with 0.01 N alcoholic KOH after adding 0.3 ml phenol red and 15 ml neutral absolute ethanol.

Sugar Determination

A method for determination of glucose, galactose, and lactose was developed during the course of research. The method consisted of the enzymatic determination of glucose by glucose oxidase, galactose by galactose oxidase, and lactose as its hydrolysis products using specific enzymes.

Regents

1. DOWEX 1-X10 (ionic form Cl^-, mesh 200-400).
2. DOWEX 50W-X8 (ionic form H^+, mesh 200-400).
3. 3 N HCl.
4. 6 N NaOH.
5. 12.5% trichloroacetic acid (TCA).

6. Glucostat[1] (a coupled enzyme system for glucose).
7. Galactostat[2] (a coupled enzyme system for galactose).
8. Glycine buffer, pH 9.7.
9. 0.15 M phosphate buffer.
10. 4 M HCl.

Preparation of resin

The cationic resin DOWEX 1-X10 was converted to the hydroxyl form with 8.0% sodium hydroxide. The resin was then washed thoroughly with distilled water until the sodium hydroxide was removed. This was determined by adding a drop of phenolphthalein to the wash water and observing color change. Both DOWEX 1-X10 and DOWEX 50W-X8 were then dried under reduced pressure at 45 C for 18 hr (29).

Resin mixture I One part of DOWEX 50W-X8 was mixed with 7 parts of DOWEX 1-X10 using a mortar and pestle.

Resin mixture II One part of DOWEX 50W-X8 was mixed with 4 parts of DOWEX 1-X10 using a mortar and pestle.

Preparation of filtrate

Fifty grams of cheese were weighed into a Waring blendor. With the help of a volumetric pipette, 100 ml of distilled water were added and the mixture was blended for 7 min. The pH of the homogenate was adjusted to 4.6 with 12.5% TCA while still in the Waring blendor container.

[1,2]Worthington Biochemical Corporation, Freehold, N. J.

The samples were then transferred into centrifuge tubes and centrifuged for 15 min at 2,000 rpm. The supernatant was collected with a probe needle attached to a syringe to avoid the fat layer at the top of the centrifuge tube. Ten milliliters of sample thus collected were transferred into screw-capped centrifuge tubes. Then, 5 ml of 3 N HCl were added, mixed, and centrifuged for 15 min at 2,000 rpm. The clear supernatant in the centrifuge tubes was used for different sugar analyses. For determination of glucose and galactose, 10 ml of the supernatant were placed in a 30-ml beaker and neutralized to pH 7.0 with 6 N NaOH using a microburette. For the lactose determination, an aliquot of the supernatant was poured to another screw-capped test tube and heated for 4.5 hr in a silicone or mineral oil bath at 90 C. This treatment was found to induce maximum hydrolysis in preliminary experiments using different time-temperature combinations.

Galactose determination

Ten milliliters of neutralized samples were pipetted into 50-ml Erlenmeyer flasks containing 2.6 g previously weighed DOWEX resin mixture II to remove interfering materials (29). Samples were filtered through Whatman No. 40 filter paper. The "Galactostat" was prepared according to manufacturer's directions (Worthington Biochemical Corporation). The "Chromogen" was dissolved in 0.5 ml absolute methanol and was added to 0.15 M phosphate buffer (pH 7.0). Then, the "Galactostat" was dissolved in 0.15 M phosphate buffer and transferred quantitatively into the graduated cylinder. The final volume was restored with 0.15 M phosphate buffer to 50.0 ml. Two milliliters of filtrate were pipetted

into a standardized spectrophotometer tube and incubated in a water bath at 37 C. At zero time, 2.0 ml of enzyme preparation were added. At exactly 60 min, 6.0 ml of glycine buffer (pH 9.7) were added to stop the reaction. The developed color was read in a Coleman model 11 spectrophotometer at 425 mμ against a reagent blank using 2 ml of distilled water instead of sample. The amount of galactose present was determined directly from a standard curve (Fig. 2).

Glucose determination

Ten milliliters of neutralized sample were pipetted into a 50-ml Erlenmeyer flask containing 2.6 g DOWEX resin mixture I, mixed, and filtered through Whatman No. 40 filter paper. The "Glucostat" was prepared according to manufacturer's directions. The "Chromogen" and the "Glucostat" were dissolved in distilled water and transferred quantitatively into a graduated cylinder containing 60 ml of distilled water. The final volume was restored to 90 ml with distilled water. Nine milliliters of glucostat were placed in a cuvette. One milliliter of filtrate was added to the cuvette at zero time; then, after exactly 10 min, 1 drop of 4 M HCl was added to stop the reaction. The developed color was read in a Coleman model 11 spectrophotometer at 400 mμ. The amount of glucose was determined from a standard curve (Fig. 3).

Lactose determination

Samples were cooled and neutralized after lactose hydrolysis at 90 C for 4.5 hr. The amount of glucose in an aliquot was determined as described previously. The amount of lactose was expressed on the basis

Fig. 2. Relationship between absorbence and concentration of galactose

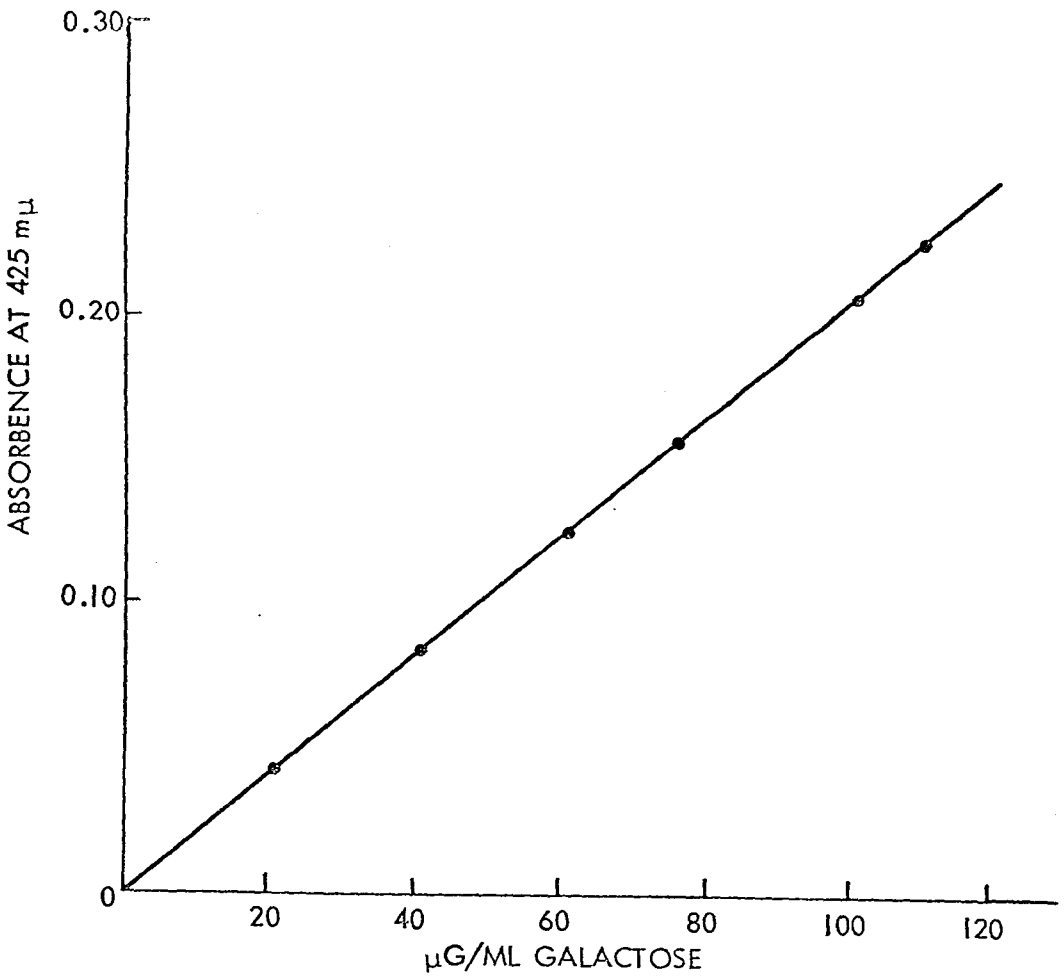
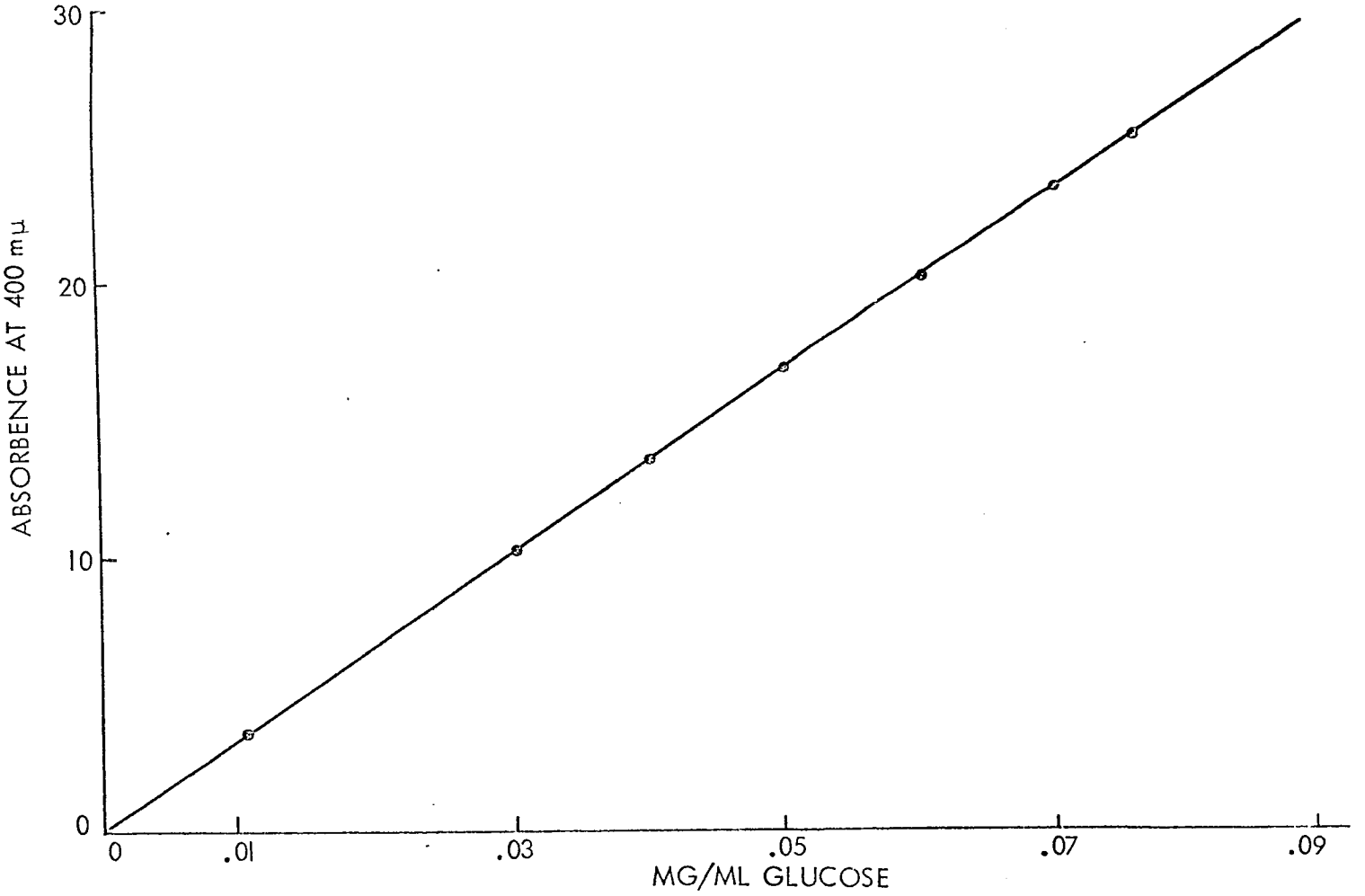

0.30
0.20
0.10
0
ABSORBENCE AT 425 mμ
20
40
60
80
100
120
μG/ML GALACTOSE

Fig. 3. Relationship between absorbence and concentration of glucose


30
20
10
0
ABSORBENCE AT 400 mμ
0
.01
.03
.05
.07
.09
MG/ML GLUCOSE

of the amount of glucose in the hydrolyzed sample of cheese. A correction factor was applied on the basis of 80% lactose hydrolysis. Dilution of the aliquot was made when lactose concentration was higher than the standard range required to obey the Beer's Law.

Preparation of standard curves

Known amounts of glucose (range 0-100 µg/ml in 0.5% (w/v NaCl) and galactose (range 0-60 µg/ml H_2O) were used to prepare the standard curves (Fig. 2 and 3). Samples for the standard curves were treated with resins in same way as the samples.

Measurement of color

Color measurements were made after 3 months on cheeses made in the large vats. The color was determined by reflectance measurement on a small plug of cheese using a Beckman DK-2A ratio recording spectrophotometer[1]. Reflectance curves were obtained on four different treatments of a batch of cheese on the same chart paper at wavelengths from 350 to 700 mµ to obtain a comparison between different treatments.

[1]Beckman Instrument Co., Fullerton, California.

RESULTS AND DISCUSSION

Rate of Cooling

Complete and precise information on the rate of cooling of freshly pressed curd to the curing room temperature is lacking in the literature. This phase of study was designed to determine the cooling rates of cheese when handled according to commercial practice, and when induced to cool rapidly in a brine tank. Rate of cooling of different stacks of cheeses were determined by a multi-channel continuous recording system. Tables 1 and 2 show the rates of cooling of 40- and 20-lb. blocks in brine and in air at 7.5 C. When 40-lb. blocks were stacked in a curing room held at 7.5 C and cooled by air, the cheeses approximately required 150 to 480 hr to reach the curing room temperature. But, by immersing blocks after pressing and sealing in brine at 7.5 C, only 25 to 70 hr were required to reach 7.5 C. The rate of cooling also was found to depend on the number of warm blocks stacked together. From lot A, only one block of cheese was placed on a pallet in the curing room after each pressing time. The cooling rates of lot A cheeses were faster than other lots-i.e. lots B, C, D, E, F, G, H, and I-which were stacked 2 to 4 blocks high close together on a pallet in the same curing room. The cooling rate also was affected by the size of the blocks. Because of the smaller size and consequent more rapid temperature equilibration, 20-pound blocks cooled more rapidly both in curing room and in brine. Most 20-pound blocks required only 20 to 25 hr (brine cooling) or 140 to 185 hr (air cooling) to come to 7.5 C.

Table 1. Approximate time required to cool 40-lb. blocks to 8 C

Lots	Treatment			
	Pressed 4 hr		Pressed 20 hr	
	Brine cooled	Air cooled	Brine cooled	Air cooled
	hr			
A[a]	25	65	45	150
B[b]	45	480	115	480
C	40	265	50	265
D	90	310	90	310
E	45	290	65	290
F	70	165	70	190
G	70	220	70	220
H	50	170	70	170
I	45	165	45	165

[a]A single block was placed on a pallet after each pressing time.

[b]Lots B to I: Two or more blocks were stacked tightly on a pallet in the curing room after each pressing time.

Table 2. Approximate time required to cool 20-lb. blocks to 8 C

Lots	Treatment			
	Pressed 4 hr		Pressed 20 hr	
	Brine cooled	Air cooled	Brine cooled	Air cooled
	hr			
M	25	185	40	185
N	20	140	40	140
O	20	120	40	120
P	20	150	55	150
Q	20	140	45	140
R	25	140	40	140

Fig. 4. Cooling rate of 40-lb. blocks from lot B in brine (7.5 C) and in curing room (7.5 C)

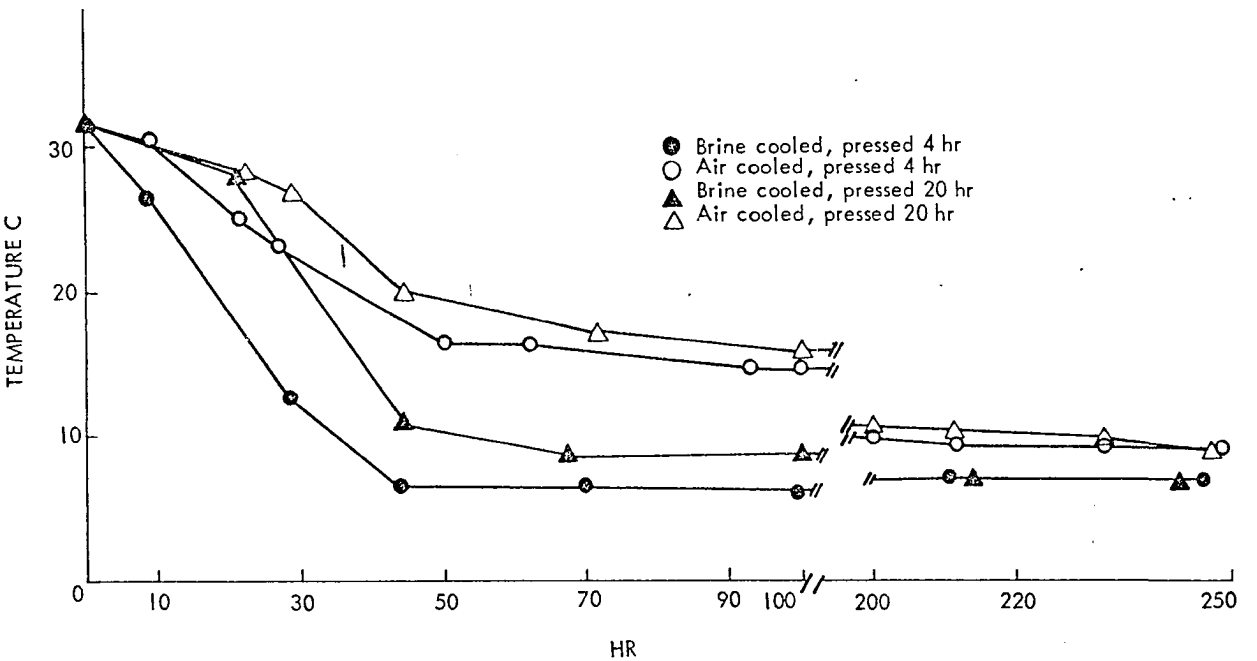

Brine cooled, pressed 4 hr
Air cooled, pressed 4 hr
Brine cooled, pressed 20 hr
Air cooled, pressed 20 hr
TEMPERATURE C
30
20
10
0
10
30
50
70
90
100
200
220
250
HR

Fig. 5. Cooling rate of 40-lb. blocks from lot H in brine (7.5 C) and in curing room (7.5 C)

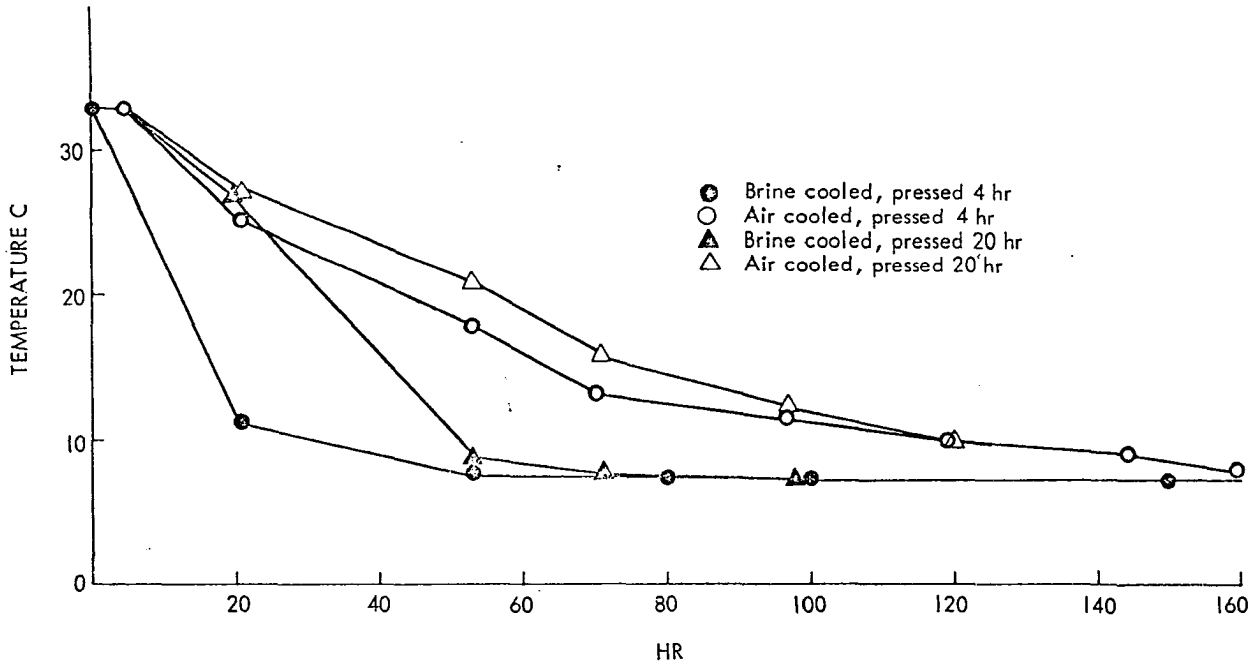
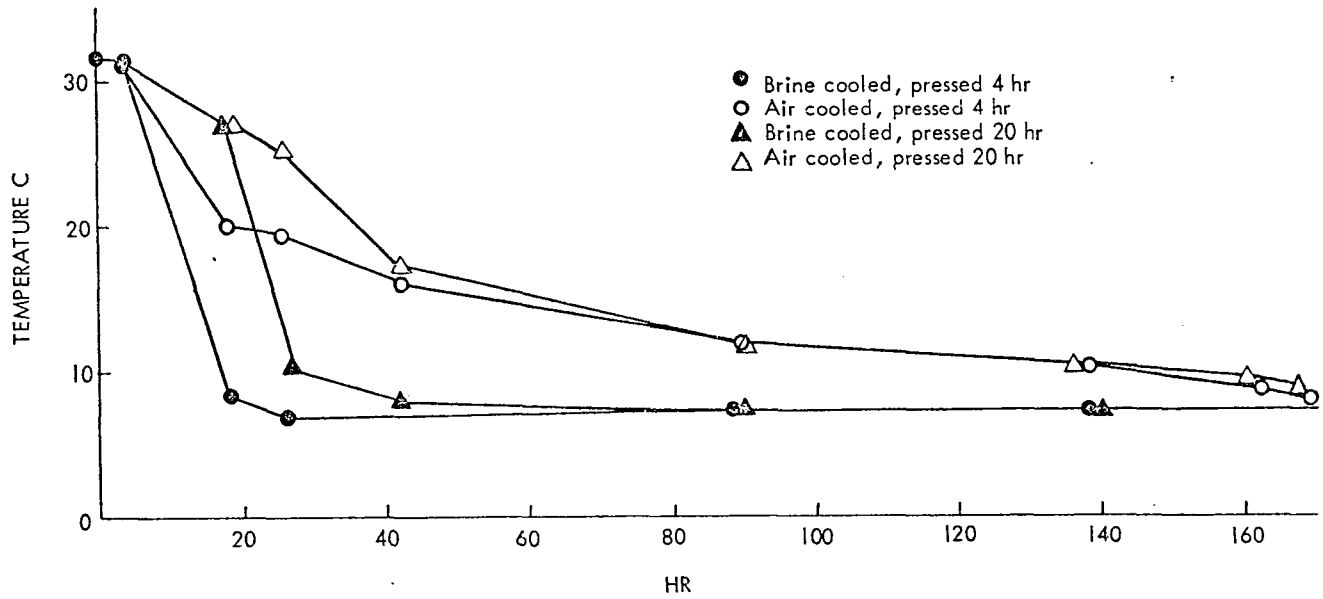

TEMPERATURE C
30
20
10
0
20
40
60
80
100
120
140
160
HR
Brine cooled, pressed 4 hr
Air cooled, pressed 4 hr
Brine cooled, pressed 20 hr
Air cooled, pressed 20 hr

Fig. 6. Cooling rate of 20-lb. blocks from lot M in brine (7.5 C) and in curing room (7.5 C)


Brine cooled, pressed 4 hr
Air cooled, pressed 4 hr
Brine cooled, pressed 20 hr
Air cooled, pressed 20 hr
TEMPERATURE C
30
20
10
0
20
40
60
80
100
120
140
160
HR

Temperature changes in press also were observed over a 20-hr period. Depending on temperature fluctuation in the room where the press was installed, up to 4 C drop in temperature of the cheese in the press was noted. The plots of temperature changes in the various blocks from several experimental lots are shown in Fig. 4, 5, and 6. Temperature of brine-cooled blocks fell rapidly to 8 C or below within 48 hr. The temperature of air-cooled blocks fell gradually down to 14-18 C in 90 to 100 hr and remained within this range for over 70 hr until the attainment of 8 C or lower (Fig. 4).

These results are in full agreement with the information received from the cheese manufacturing plants surveyed as a part of this study and that of Conochie and Sutherland (13). Confidential information received from a commercial cheese manufacturer also indicates that under usual commercial conditions with a large number of blocks stacked together, the cooling rate is much slower. This company studied the cooling rate of 40-lb. blocks stacked eight high against an outside wall of a curing room held at 30-32 F (-1 to 0 C). The temperature of the fourth block dropped from 85 F (29.5 C) to 54 F (12.5 C) in 7 days. When the blocks were stacked five high with 6 inches of space in between blocks, the temperature fell from 78 F (25 C) to 33 F (0.75 C) within the same period. The same manufacturer also indicated that blocks on the top of a twelve high stack (with initial block temperature of 74 F) took 28 days to cool to 34 F (1.5 C) in a room held at same temperature. But the fourth block from the floor cooled only to 45 F (7.5 C) in 28 days.

The data presented in this experiment in no way represent an exaggerated condition. According to a confidential source it takes at least one month to cool 40-lb. blocks to curing room temperature of 40-45 F (4.5-7.5 C).

Bacterial count of experimental cheese

Bacterial counts were made to observe the relationship of temperature during early stage of curing to the population of starter and non-starter organisms. Since enterococci were reported to be the predominant flora of young Cheddar cheese (12), this was also included in this study. Importance of bacterial flora on the development of characteristic Cheddar flavor has been recognized by many investigators. Flavor development in cheese has been known to be due to controlled fermentation and breakdown of milk components. These changes in cheese are produced not only by starter microorganisms but also the various adventitious types that are added to cheese at various stages of manufacture and handling. For this study total count, enterococcus count, and gram-negative bacterial count were chosen.

Total count The data presented in Tables 3, 4, and 5 show the total bacterial count of different lots of cheese at different sampling periods. Lots A, B, and C (Table 3) were made from three batches of Iowa manufacturing-grade milk. All these cheeses were milled at acidities above 0.60%. The total bacterial count of raw milk ranged from 11,000,000 to 130,000,000/ml with an average of 73,000,000/ml; however, after heat treatment at 62.8 C for 17 sec the bacterial population was greatly reduced. In cheeses made from manufacturing-grade

Table 3. Total bacterial count of high acid cheeses[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots A	B	C	Avg	Pressed 4 hr, Air cooled, Cheese lots A	B	C	Avg	Treatment: A
	X 10^6 per g								
1	130.0	89.0	11.0	73.0					
2	0.8	53.0	3.8	19.0					
3	29.0	12.0	17.0	19.0					
4	1400.0	530.0	1200.0	1000.0					
5									490.0
6	320.0	870.0	500.0	560.0	540.0	23.0	620.0	400.0	510.0
7	230.0	580.0	120.0	310.0	100.0	400.0	120.0	210.0	370.0
8	470.0	1800.0	14.0	440.0	130.0	320.0	180.0	210.0	130.0
9	310.0	310.0	380.0	330.0	620.0	330.0	260.0	400.0	610.0
10	110.0	22.0	500.0	210.0	67.0	43.0	400.0	170.0	49.0
11	1.8	17.0	92.0	37.00	1.7	12.0	55.0	21.0	1.5
12	1.0	27.0	62.0	30.0	3.1	12.0	58.0	24.0	3.1

[a] Made from manufacturing-grade milk.

[b] 1-Raw milk; 2-Vat milk; 3-Milling; 4-Pressed 4 hr; 5-Pressed 20 hr; 6-4th day; 7-8 12-3 months.

Treatment									
		Pressed 20 hr							
		Brine cooled				Air cooled			
Cheese lots									
	Avg	A	B	C	Avg	A	B	C	Avg
X 10^6 per g									
		490.0	770.0	110.0	460.0				
.0	400.0	510.0	820.0	380.0	570.0	650.0	1000.0	110.0	590.0
.0	210.0	370.0	430.0	120.0	310.0	400.0	470.0	160.0	340.0
0	210.0	130.0	950.0	580.0	550.0	160.0	310.0	280.0	250.0
.0	400.0	610.0	310.0	290.0	400.0	75.0	110.0	120.0	100.0
0	170.0	49.0	90.0	460.0	200.0	37.0	45.0	190.0	91.0
0	21.0	1.5	15.0	34.0	17.0	6.6	20.0	39.0	22.0
0	24.0	3.1	33.0	81.0	39.0	3.0	16.0	58.0	26.0

20 hr; 6-4th day; 7-8th day; 8-12th day, 9-21st day; 10-1 month; 11-2 months;

Table 4. Total bacterial count of normal acid cheeses[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots D	E[c]	F	Avg	Air cooled, Cheese lots D	E	F	Avg	Br, Cheese lots D	Br
	X 10^6 per g									
1	61.0	0.6	34.0	32.0						
2	15.0	300.0	-3.6	110.0						
3	150.0	3900.0	9500.0	4500.0						
4	920.0	2700.0	1600.0	1700.0						
5									550.0	210
6	910.0	2600.0	---[d]	1800.0	660.0	1500.0	---	1100.0	590.0	160
7	640.0	1900.0	---	1300.0	320.0	1200.0	---	760.0	120.0	230
8	68.0	1900.0	1000.0	990.0	10.0	1700.0	1300.0	1000.0	60.0	240
9	500.0	---	---		350.0	---	---		600.0	
10	600.0	1400.0	560.0	850.0	410.0	870.0	530.0	600.0	430.0	160
11	110.0	300.0	230.0	210.0	130.0	620.0	460.0	400.0	73.0	6
12	42.0	27.0	370.0	150.0	32.0	23.0	380.0	150.0	72.0	1

[a]Made from manufacturing-grade milk.

[b]1-Raw milk; 2-Vat milk; 3-Milling; 4-Pressed 4 hr; 5-Pressed 20 hr; 6-4th day; 7-8th d
12-3 months.

[c]Raw milk cheese.

[d]Not run.

Treatment									
		Pressed 20 hr							
)oled		Brine cooled				Air cooled			
Cheese lots									
F	Avg	D	E	F	Avg	D	E	F	Avg
X 10^6 per g									
		550.0	2100.0	9400.0	4000.0				
---	1100.0	590.0	1600.0	---	1100.0	330.0	1400.0	---	860.0
---	760.0	120.0	2300.0	---	1200.0	110.0	1600.0	---	810.0
1300.0	1000.0	60.0	2400.0	1300.0	1300.0	65.0	1900.0	1200.0	1100.0
---		600.0	---	---		150.0	---	---	
530.0	600.0	430.0	1600.0	510.0	850.0	180.0	1600.0	900.0	890.0
460.0	400.0	73.0	63.0	42.0	59.0	58.0	180.0	280.0	170.0
380.0	150.0	72.0	18.0	210.0	100.0	100.0	15.0	250.0	120.0

essed 20 hr; 6-4th day; 7-8th day; 8-12th day; 9-21st day; 10-1 month; 11-2 months;

Table 5. Total bacterial count of experimental fruity cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled: Cheese lots G	H	I	Avg	Pressed 4 hr, Air cooled: Cheese lots G	H	I	Avg	Cheese lots G
	X 10^6 per g								
1	---	0.4	(0.04)	(0.22)					
2	(.0028)	0.4	(0.002)	(0.13)					
3	910.0	23.0	141.0	360.0					
4	170.0	420.0	480.0	360.0					
5									650.0
6	33.0	300.0	440.0	260.0	250.0	180.0	360.0	260.0	11.0
7	36.0	62.0	18.0	39.0	57.0	120.0	8.9	62.0	3.1
8	0.6	4.0	31.0	12.0	3.0	12.0	39.0	18.0	0.1
9	21.0	22.0	81.0	41.0	16.0	84.0	27.0	42.0	14.0

[a]Made from grade-A milk with a culture producing fruity flavor.

[b]1-Raw milk; 2-Vat milk; 3-Milling; 4-Pressed 4 hr; 5-Pressed 20 hr; 6-12th day; 7-1 mo

Treatment									
		Pressed 20 hr							
ed		Brine cooled				Air cooled			
Cheese lots									
I	Avg	G	H	I	Avg	G	H	I	Avg
X 10^6 per g									
		650.0	670.0	450.0	590.0				
360.0	260.0	11.0	130.0	260.0	130.0	12.0	90.0	300.0	130.0
8.9	62.0	3.1	6.7	66.0	25.0	31.0	18.0	400.0	150.0
39.0	18.0	0.1	1.6	48.0	17.0	0.1	37.0	51.0	29.0
27.0	42.0	14.0	22.0	21.0	19.0	24.0	28.0	46.0	33.0

avor .

ed 20 hr; 6-12th day; 7-1 month; 8-2 months; 9-3 months.

milk there was a slight increase in bacterial count during the first 21 days; then it declined sharply. The cooling rate and pressing time did not influence the total bacterial count. On the fourth day, the brine-cooled high acid cheese blocks pressed 4 hr and 20 hr had an average count of 560,000,000 and 520,000,000/g, respectively and the air-cooled blocks, 400,000,000 and 590,000,000/g, respectively showing no appreciable differences.

The experimental fruity cheese (lots G, H, and I) and normal acid (milling acidities below 0.60%) cheese (lots D, E, and F) did not show any significant differences in the total bacterial count at this period.

Data collected on the 12th day, and after 1, 2, and 3 months on all cheeses were statistically analyzed. Cooling rates or pressing times did not have any significant effect on total counts.

From statistical treatment of the data, a highly significant F value was obtained for the interaction between sampling period X type of cheese. This indicated that the total bacterial count of different types of cheese (normal acid, high acid, and fruity) declined at a different rates. It is obvious from Tables 4, 5, and 6, that considerably large numbers of microorganisms (150,000,000/g) persist for 3 months in normal acid cheese but in fruity (average total initial count 360,000,000/g) and high acid (average total initial count 19,000,000/g) cheeses the total bacterial count drops down to 42,000,000/g or below after 1 month. The number of bacteria present during the curing period was relatively higher in normal acid cheese (Table 4) and lower in experimental fruity cheese (Table 5).

Enterococcus count The enterococcus count was determined only on normal acid cheese made from Iowa manufacturing-grade milk (lots D, E, and F) and experimental fruity cheese made from grade-A milk. The enterococcus count in manufacturing-grade raw milk ranged from 130,000 to 18,000,000/ml with an average count of 12,000,000/ml. The grade-A milk contained less than 10 to 200/ml with an average of 100/ml. The heat treatment reduced the enterococcus count to one-third or less of the original numbers. The initial numbers of enterococci in the raw milk seemed to influence the extent of their survival until later stages of curing. Data presented in Table 6 indicated that a large number of enterococci were present in normal acid cheese made from the manufacturing-grade milk. Their numbers increased rapidly from the time of milling up to 1 month. The air-cooled blocks seemed to have a greater number of enterococci during the later part of the curing period. In normal acid cheese (lot E) made from raw manufacturing-grade milk, a greater increase occurred in the numbers of enterococci during curing. Experimental fruity cheese (Table 7), made from grade-A milk, did not show any significant increase in the number of enterococci during the sampling period.

Statistical analysis of the data showed a significant difference at the 1% level in the number of enterococci present in cheeses made with different milk. No significant differences were observed in the number of enterococci at any stage of curing due to cooling rates or pressing time.

Table 6. Enterococcus count of normal acid cheese[a]

Sampling period[b]	Treatment									
	Pressed 4 hr									
	Brine cooled				Air cooled					
	Cheese lots									
	D	E[c]	F	Avg	D	E	F	Avg	D	
	X 10^4 per g									
1	18.0	(0.13)	17.0	12.0						
2	6.4	65.0	2.2	25.0						
3	290.0	1800.0	85.0	720.0						
4	270.0	2200.0	14.0	830.0						
5									89.0	2
6	1800.0	1500.0	---[d]	1700.0	2200.0	1500.0	---	1900.0	1700.0	
7	1800.0	1300.0	---	1600.0	2400.0	1100.0	---	1800.0	800.0	1
8	2400.0	1300.0	92.0	1300.0	1100.0	1100.0	140.0	780.0	3000.0	1
9	2100.0	---	---		2800.0	---	---		3100.0	
10	3200.0	5200.0	88.0	2800.0	800.0	3400.0	91.0	1400.0	440.0	5
11	660.0	75.0	98.0	280.0	1200.0	530.0	130.0	620.0	680.0	
12	53.0	52.0	89.0	65.0	490.0	110.0	110.0	240.0	1500.0	

[a] Made from manufacturing-grade milk.

[b] 1-Raw milk; 2-Vat milk; 3-Milling; 4-Pressed 4 hr; 5-Pressed 20 hr; 6-4th day; 7-8th
12-3 months.

[c] Raw milk cheese.

[d] Not run.

Treatment									
		Pressed 20 hr							
ooled		Brine cooled				Air cooled			
Cheese lots									
F	Avg	D	E	F	Avg	D	E	F	Avg
X 10^4 per g									
		89.0	2100.0	15.0	740.0				
---	1900.0	1700.0	740.0	---	1200.0	2100.0	1200.0	---	1700.0
---	1800.0	800.0	1500.0	---	1200.0	1200.0	1200.0	---	1200.0
140.0	780.0	3000.0	1300.0	180.0	1500.0	2200.0	1900.0	160.0	1400.0
---		3100.0	---	---		4000.0	---	---	
91.0	1400.0	440.0	5700.0	130.0	2100.0	810.0	8500.0	54.0	3100.0
130.0	620.0	680.0	38.0	49.0	260.0	1100.0	110.0	29.0	410.0
110.0	240.0	1500.0	46.0	95.0	550.0	1900.0	140.0	81.0	710.0

ressed 20 hr; 6-4th day; 7-8th day; 8-12th day; 9-21st day; 10-1 month; 11-2 months;

Table 7. Enterococcus count of experimental fruity cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots G	H	I	Avg	Pressed 4 hr, Air cooled, Cheese lots G	H	I	Avg	Bri: Cheese lots G	H
	X 10^4 per g									
1	<10	0.01	0.02	0.01						
2	<10	<10	<10	<10						
3	<10	0.01	0.01	0.01						
4	<10	0.05	0.01	0.02						
5									<10	0.
6	0.03	0.04	0.02	0.03	0.05	0.05	0.01	0.04	0.03	0.
7	0.07	0.03	0.03	0.04	0.05	0.07	0.02	0.05	0.06	0.
8	0.05	0.03	0.04	0.04	0.04	0.07	0.04	0.05	0.05	0.
9	0.05	0.04	(0.003)	0.03	0.03	0.02	<10	0.02	0.03	0.

[a]Made from grade-A milk with a culture producing fruity flavor.

[b]1-Raw milk; 2-Vat milk; 3-Milling; 4-Pressed 4 hr; 5-Pressed 20 hr; 6-12th day; 7-1 mon

Treatment									
		Pressed 20 hr							
oled		Brine cooled				Air cooled			
Cheese lots									
I	Avg	G	H	I	Avg	G	H	I	Avg
X 10^4 per g									
		< 10	0.04	< 10	0.01				
0.01	0.04	0.03	0.03	0.01	0.02	0.03	0.02	0.01	0.02
0.02	0.05	0.06	0.02	0.10	0.06	0.04	0.06	0.09	0.06
0.04	0.05	0.05	0.04	0.03	0.04	0.03	0.09	0.03	0.05
< 10	0.02	0.03	0.02	0.01	0.02	0.05	0.02	0.01	0.03

flavor.

essed 20 hr; 6-12th day; 7-1 month; 8-2 months; 9-3 months.

Gram-negative bacterial count The gram-negative bacterial count showed the same trend as the enterococcus count (Tables 8 and 9). Cheese, made from poor quality milk, harbored more gram-negative bacteria than cheese made from grade-A milk. Manufacturing-grade raw milk contained from 74,000 to 1,100,000/ml with an average of 470,000/ml of these bacteria. Heat treatment reduced the gram-negative bacterial content by 99.9 per cent. Grade-A milk contained from 2,800 to 20,000/ml with an average of 11,000/ml; heat treatment reduced this by almost 100 per cent. Survival of gram-negative bacteria was very low in all the cheeses made from grade-A milk. Gram-negative bacteria in cheeses made with manufacturing-grade milk showed an increase in count from the time of milling (39,000/g) to 1 month (410,000/g) of curing. This increase was even greater in lot E which was made from raw manufacturing-grade milk. Brine-cooled cheeses, pressed 4 hr and 20 hr had higher gram-negative bacterial counts upto twelfth-day compared to corresponding air-cooled cheeses.

Although there was a great difference in time required to reach the desired curing temperature between the brine-cooled and the air-cooled cheese blocks, no corresponding significant differences in total count, enterococcus count, and gram-negative bacterial count were observed.

The decrease in total bacterial counts after 2-3 weeks was probably the result of gradual death of starter organisms due to accumulation of metabolites (21, 48, 67). In raw milk cheese, however, the total count was quite high up to 2 months of maturation and then the count dropped. This may have been due to the presence of lactobacilli in raw milk as

Table 8. Gram-negative bacterial count of normal acid cheeses[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots D	E[c]	F	Avg	Air cooled, D	E	F	Avg	Treatment, Cheese lots D
	X 10^2 per g								
1	2900.0	740.0	11000.0	4700.0					
2	3.6	240.0	< 10	120.0					
3	910.0	240.0	4.2	390.0					
4	780.0	1400.0	7.2	730.0					
5									210.0
6	530.0	14000.0	---	7300.0	180.0	6800.0	---	3500.0	120.0
7	220.0	12000.0	---	6100.0	320.0	7900.0	---	4100.0	7.6
8	150.0	120000.0	10.0	40000.0	150.0	20000.0	25.0	6700.0	630.0
9	300.0	---	---		310.0	---	---		330.0
10	590.0	3000.0	4.6	1200.0	560.0	1000.0	5.0	520.0	440.0
11	160.0	60.0	2.6	74.0	170.0	2300.0	2.5	820.0	69.0
12	< 10	13.0	0.3	4.4	< 10	25.0	0.7	8.6	< 10

[a] Made from manufacturing-grade milk.

[b] 1-Raw milk; 2-Vat milk; 3-Milling; 4-Pressed 4 hr; 5-Pressed 20 hr; 6-4th day; 7-8t
12-3 months.

[c] Raw milk cheese.

[d] Not run.

es[a]

Treatment									
		Pressed 20 hr							
ooled		Brine cooled				Air cooled			
Cheese lots									
F	Avg	D	E	F	Avg	D	E	F	Avg
X 10^2 per g									
		210.0	750.0	8.4	320.0				
---	3500.0	120.0	750.0	---	430.0	140.0	240.0	---	190.0
---	4100.0	7.6	45000.0	---	23000.0	63.0	13000.0	---	6500.0
25.0	6700.0	630.0	62000.0	40.0	21000.0	570.0	46000.0	32.0	16000.0
---		330.0	---	---		81.0	---	---	
5.0	520.0	440.0	4600.0	10.0	1700.0	290.0	12000.0	14.0	4100.0
2.5	820.0	69.0	13.0	(0.01)	27.0	26.0	57.0	0.2	28.0
0.7	8.6	< 10	3.3	0.2	1.2	< 10	35.0	0.7	12.0

essed 20 hr; 6-4th day; 7-8th day; 8-12th day; 9-21st day; 10-1 month; 11-2 months;

Table 9. Gram-negative bacterial count of experimental fruity cheese[a]

Sampling period[b]	Treatm: Pressed 4 hr, Brine cooled G	H	I	Avg	Air cooled G	H	I	Avg	Cheese Avg
									X 10^2 p
1	---	200.0	28.0	100.0					
2	< 10	<10	< 10	<10					
3	39.0	<10	0.4	13.0					
4	0.9	0.2	61.0	21.0					
5									
6	< 10	0.4	22.0	7.5	<10	0.4	11.0	3.8	
7	< 10	0.1	< 10	(0.03)	<10	0.1	<10	(0.03)	
8	<10	0.6	1.0	(0.53)	<10	1.6	1.4	1.0	
9	<10	<10	0.4	(0.13)	<10	<10	0.8	(0.27)	

[a] Made from grade-A milk with a culture producing fruity flavor.

[b] 1-Raw milk; 2-Vat milk; 3-Milling; 4-Pressed 4 hr; 5-Pressed 20 hr; 6-12th

y cheese[a]

Treatment									
		Pressed 20 hr							
oled		Brine cooled				Air cooled			
Cheese lots									
I	Avg	G	H	I	Avg	G	H	I	Avg
X 10^2 per g									
		2.4	8.0	13.0	8.0				
11.0	3.8	<10	<10	11.0	3.7	<10	<10	5.1	1.7
<10	(0.03)	<10	<10	3.3	1.1	<10	<10	13.0	4.3
1.4	1.0	<10	0.9	0.4	1.1	<10	3.0	1.4	1.5
0.8	(0.27)	<10	<10	0.3	0.1	<10	<10	0.5	(0.17)

flavor.

essed 20 hr; 6-12th day; 7-1 month; 8-2 months; 9-3 months.

reported by Johns and Cole (49). Lactobacilli are known to survive in large numbers in Cheddar cheese for prolonged periods (31).

Enterococci were present in large numbers only in cheese made from manufacturing-grade milk. In recent studies, Clark and Reinbold (12) reported frequent incidence of enterococci in young commercial Cheddar cheese. Initial presence of enterococci in milk probably influences their presence in cheese during curing.

Results of this experiment are in full agreement with those presented by Yale and Marquardt (89) who indicated coliform organisms survived for 6 to 12 months in cheese made from poor quality milk. With better quality milk these organisms survived for only 3 to 6 months. Crossley (14, 15), on the other hand, observed that the overall sanitation of plant and the personnel during making was contributory to high coliform count rather than poor quality of milk.

From this study it is difficult to arrive at any definite conclusion regarding the influence of curd cooling rates on the overall bacterial population, and the individual groups enumerated. However, the relative proportions of the various microbial groups in the raw cheese and their growth and metabolic response to the different cooling rates at this period appears to influence the characteristics of the final products as seen by judging scores.

pH The pH of all cheeses were determined at the same intervals as the bacterial counts. The pH values of high acid, normal acid, and experimental fruity cheeses are presented in Tables 10, 11, and 12, respectively. The pH range of high acid cheeses at milling was from

Table 10. Comparison of pH of high acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots A	B	C	Avg	Air cooled, Cheese lots A	B	C	Avg	Cheese lots A
1	---[c]	5.20	5.10	5.15					
2	4.70	5.20	5.20	5.03					
3									4.65
4	4.90	4.90	5.30	5.03	4.75	5.00	5.20	5.32	4.90
5	4.70	4.99	5.35	5.01	4.65	4.80	5.20	4.88	4.70
6	---	4.80	5.40	5.10	---	4.70	5.28	4.99	---
7	5.00	5.00	5.25	5.08	5.00	5.00	5.20	5.07	4.90
8	4.50	5.25	5.30	5.02	4.45	5.10	5.20	4.92	4.50
9	4.90	5.20	5.45	5.18	4.90	5.02	5.40	5.11	4.88
10	5.10	5.54	5.75	5.44	5.10	5.40	5.65	5.38	5.10

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day; 7-2

[c]Not run.

Treatment									
		Pressed 20 hr							
led		Brine cooled				Air cooled			
Cheese lots									
C	Avg	A	B	C	Avg	A	B	C	Avg
		4.65	5.10	5.20	4.98				
5.20	5.32	4.90	5.10	5.25	5.08	4.70	5.00	5.10	4.93
5.20	4.88	4.70	4.90	5.35	4.98	4.70	4.75	5.25	4.90
5.28	4.99	---	4.80	5.35	5.08	---	4.70	5.30	5.00
5.20	5.07	4.90	5.00	5.32	5.07	4.90	4.90	5.18	4.99
5.20	4.92	4.50	5.30	5.25	5.02	4.50	5.15	5.15	4.93
5.40	5.11	4.88	5.25	5.48	5.20	4.90	5.20	5.39	5.16
5.65	5.38	5.10	5.45	5.70	5.42	5.10	5.40	5.65	5.38

8th day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 11. Comparison of pH of normal acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots D	E[c]	F	Avg	Pressed 4 hr, Air cooled, Cheese lots D	E	F	Avg	Cheese lots D
1	5.55	5.40	5.50	5.48					
2	5.52	5.50	5.60	5.54					
3									5.55
4	5.58	5.55	---[d]		5.50	5.55	---		5.58
5	5.55	5.60	---		5.55	5.48	---		5.55
6	5.88	5.65	5.70	5.74	5.75	5.58	5.65	5.66	5.82
7	5.85	---	---		5.75	---	---		5.82
8	5.72	5.20	5.90	5.71	5.68	5.40	5.80	5.63	5.75
9	5.80	5.75	5.90	5.82	5.72	5.75	5.80	5.77	5.78
10	5.71	5.75	5.70	5.72	5.75	5.65	5.62	5.67	5.75

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day; 7-21s

[c]Raw milk cheese.

[d]Not run.

Treatment								
	Pressed 20 hr							
	Brine cooled				Air cooled			
Cheese lots								
Avg	D	E	F	Avg	D	E	F	Avg
	5.55	5.55	5.60	5.57				
	5.58	5.60	---	5.59	5.58	5.45	---	5.52
	5.55	5.52	---	5.54	5.50	5.38	---	5.44
5.66	5.82	5.52	5.65	5.66	5.78	5.42	5.60	5.60
	5.82	---	---		5.75	---	---	
5.63	5.75	5.52	5.75	5.67	5.70	5.38	5.75	5.61
5.77	5.78	5.70	5.88	5.79	5.65	5.65	5.85	5.72
5.67	5.75	5.70	5.68	5.71	5.60	5.68	5.68	5.65

6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 12. Comparison of pH of experimental fruity cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots G	H	I	Avg	Pressed 4 hr, Air cooled, Cheese lots G	H	I	Avg	Cheese lots G
1	5.50	5.50	5.60	5.53					
2	5.55	5.65	5.75	5.65					
3									5.1
4	5.65	5.65	5.72	5.67	5.65	5.60	5.75	5.67	5.6
5	5.55	5.70	5.75	5.67	5.45	5.70	5.75	5.63	5.6
6	5.62	5.78	5.82	5.74	5.55	5.68	5.70	5.64	5.6
7	5.85	5.62	5.75	5.74	5.85	5.68	5.70	5.74	5.8

[a]Made from grade-A milk with a culture producing fruity flavor.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-12th day; 5-1 month; 6-2 months;

Treatment									
		Pressed 20 hr							
ed		Brine cooled				Air cooled			
Cheese lots									
I	Avg	G	H	I	Avg	G	H	I	Avg
		5.45	5.60	5.68	5.58				
5.75	5.67	5.65	5.58	5.80	5.68	5.55	5.58	5.80	5.64
5.75	5.63	5.60	5.72	5.70	5.67	5.50	5.60	5.65	5.58
5.70	5.64	5.60	5.78	5.82	5.73	5.49	5.60	5.70	5.60
5.70	5.74	5.80	5.68	5.75	5.74	5.75	5.68	5.70	5.71

avor.

-1 month; 6-2 months; 7-3 months.

5.10 to 5.20 with an average of 5.15. The pH of normal acid and fruity cheeses at milling ranged from 5.40 to 5.60 with averages of 5.48 and 5.53, respectively. The pH of all cheeses decreased gradually up to 1 month and then increased during the rest of the observation period. The average pH values of brine-cooled cheeses pressed 4 hr and 20 hr measured at 1 month were 5.02 and that of air-cooled cheeses 4.94 and 4.93, respectively. At the end of 3 months, the pH of brine-cooled cheeses increased to 5.44 and that of air-cooled cheeses to 5.38 (Table 10). The average values of pH for normal acid cheeses, pressed 4 hr and 20 hr were 5.71 and 5.67 respectively after 1 month, and that of air-cooled cheeses 5.63 and 5.61, respectively. The pH value after 3 months curing increased to 5.72 in brine-cooled cheese and to 5.67 in air-cooled cheese. At 1 month, 4-hr and 20-hr pressed air-cooled experimental fruity cheeses, had an average pH of 5.63 and 5.58, respectively. On the other hand the average pH of brine-cooled cheeses was 5.67 at both pressing times. At the end of 3 months the average pH of both 4-hr pressed brine- and air-cooled cheeses, and 20-hr pressed brine-cooled cheeses was 5.74. But the air-cooled cheeses, pressed for 20 hr, had an average pH of 5.1.

Statistical analysis of the data showed that there was a significant difference at the 5% level between the pH of high acid, normal acid, and fruity cheese at all sampling periods. There also was a significant difference at the 1% level due to cooling of cheese in brine. Rapidly cooled cheeses had significantly higher pHs compared to air-cooled cheeses. Significant differences at the 1% level also

were found in the pH value of each sample at different sampling periods. There was, however, no significant effect of pressing time on pH.

Lactic acid Lactic acid contents of high acid, normal acid, and experimental fruity cheeses are presented in Tables 13, 14, and 15, respectively. Average lactic acid contents of high acid, normal acid, and experimental fruity cheese at milling were 0.84, 0.52, and 1.03 per cent, respectively. Lactic acid content of only one sample of fruity cheese was determined at milling. Lactic acid concentration was the highest from the 21st day to 1 month of curing in high acid cheese. Average lactic acid contents of brine-cooled high acid cheeses pressed 4 hr and 20 hr were 1.28 and 1.31 per cent, respectively after 1 month curing, and those of air-cooled cheese 1.39 and 1.40 per cent, respectively (Table 13). Lactic acid content of experimental fruity cheese was maximum on the 12th day of curing; whereas in normal acid cheeses the highest values were observed in the 3-month sample. The average lactic acid contents of experimental fruity, brine-cooled 4-hr and 20-hr pressed cheeses were 0.67 and 0.74 per cent, respectively.

Statistical analysis of the data indicated that the lactic acid concentration in air-cooled cheeses were significantly higher than those of brine-cooled cheeses at the 1% level.

Van Slyke and Price (81) and Brown and Price (8) indicated that the lowest (4.99) pH in cheese was reached on the third day and then gradually increased up to pH 5.58. In this investigation, pH was found to decrease gradually up to 1 month and then increased slowly. Vakaleris et al. (79) reported that the pH of low acid Cheddar (pH 6.45 at milling)

Table 13. Lactic acid content of high acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots A	B	C	Avg	Pressed 4 hr, Air cooled, Cheese lots A	B	C	Avg	Brine cooled, Cheese lots A	B
	%									
1	1.22	0.65	0.64	0.84						
2	1.22	0.85	0.75	0.94						
3									1.40	1.(
4	1.35	1.07	0.58	1.00	1.45	1.25	1.20	1.30	1.50	1.3
5	1.75	1.20	0.89	1.28	1.80	1.40	1.05	1.42	1.60	1.1
6	1.67	1.00	0.89	1.19	1.70	1.38	1.12	1.40	1.60	1.3
7	1.68	1.22	0.90	1.27	1.85	1.17	1.03	1.35	1.70	1.3
8	1.63	1.22	1.00	1.28	1.63	1.40	1.15	1.39	1.67	1.1
9	---[c]	1.10	0.93	1.01	---	1.20	1.15	1.18	---	1.1
10	---	1.08	0.95	1.01	---	1.24	1.15	1.20	---	1.1

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day; 7-21st da

[c]Failure to run the test properly.

Treatment								
	Pressed 20 hr							
	Brine cooled				Air cooled			
Cheese lots								
Avg	A	B	C	Avg	A	B	C	Avg
%								
	1.40	1.05	0.64	1.03				
1.30	1.50	1.22	0.85	1.19	1.55	1.37	0.90	1.27
1.42	1.60	1.13	0.89	1.21	1.90	1.15	1.00	1.35
1.40	1.60	1.85	0.80	1.08	1.74	1.25	0.85	1.28
1.35	1.70	1.20	0.97	1.29	1.75	1.40	1.05	1.40
1.39	1.67	1.10	1.15	1.31	1.65	1.38	1.15	1.40
1.18	---	1.15	0.90	1.03	---	1.24	1.16	1.20
1.20	---	1.13	0.92	1.03	---	1.13	0.98	1.06

6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 14. Lactic acid content of normal acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots D	E[c]	F	Avg	Pressed 4 hr, Air cooled, Cheese lots D	E	F	Avg	Br, Cheese lots D	Br
	%									
1	0.40	0.78	0.38	0.52						
2	0.45	0.83	0.35	0.54						
3									0.44	0
4	0.58	0.68	0.75	0.67	0.74	0.80	0.87	0.80	0.60	0
5	0.58	0.62	0.87	0.67	0.55	0.74	1.06	0.78	0.70	0
6	0.60	0.70	0.98	0.76	0.63	0.82	1.09	0.85	0.64	0
7	0.70	0.25	0.85	0.60	0.71	0.23	0.94	0.63	0.74	0
8	0.52	0.35	0.74	0.53	0.64	0.40	0.98	0.67	0.55	0
9	0.60	0.44	0.45	0.50	0.65	0.50	0.68	0.61	0.70	0
10	0.90	0.95	1.09	0.98	1.00	1.11	1.20	1.10	0.97	0

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day; 7-21st d

[c]Raw milk cheese.

Treatment									
		Pressed 20 hr							
		Brine cooled				Air cooled			
Cheese lots									
	Avg	D	E	F	Avg	D	E	F	Avg
%									
		0.44	0.80	0.53	0.59				
57	0.80	0.60	0.74	0.84	0.73	0.68	0.75	0.97	0.80
06	0.78	0.70	0.75	0.93	0.79	0.73	0.90	1.14	0.92
09	0.85	0.64	0.88	0.95	0.82	0.67	0.98	1.10	0.92
04	0.63	0.74	0.48	1.00	0.74	0.70	0.63	0.88	0.74
08	0.67	0.55	0.38	0.87	0.60	0.81	0.58	1.03	0.81
58	0.61	0.70	0.48	0.23	0.47	0.87	0.50	0.37	0.58
10	1.10	0.97	0.65	1.10	0.91	1.15	0.85	1.06	1.02

day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 15. Lactic acid content of experimental fruity cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled: Cheese lots G	H	I	Avg	Air cooled: Cheese lots G	H	I	Avg	Treatment: Cheese lots G
	%								
1	1.03	---[c]	---	1.03					
2	1.00	---	0.38	0.69					
3									0.83
4	0.81	0.83	0.55	0.73	0.83	0.88	0.71	0.81	0.98
5	0.49	0.70	0.27	0.49	0.71	0.62	0.37	0.57	0.68
6	0.92	0.68	0.44	0.67	1.15	0.55	0.62	0.77	1.06
7	0.55	0.55	0.48	0.53	0.62	0.50	0.68	0.60	0.52
8	0.17	0.57	0.30	0.35	0.25	0.62	0.50	0.46	0.38
9	0.58	0.36	0.45	0.46	0.68	0.35	0.55	0.53	0.62
10	0.62	0.58	0.70	0.63	0.65	0.72	0.75	0.71	0.67

[a]Made from grade-A milk with a culture producing fruity flavor.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day; 7-21st

[c]Not run.

Treatment									
		Pressed 20 hr							
d		Brine cooled				Air cooled			
Cheese lots									
I	Avg	G	H	I	Avg	G	H	I	Avg
%									
		0.83	---	0.38	0.61				
0.71	0.81	0.98	0.86	0.45	0.76	0.76	0.60	0.68	0.68
0.37	0.57	0.68	0.83	0.62	0.71	0.82	0.44	0.70	0.65
0.62	0.77	1.06	0.68	0.48	0.74	1.09	0.48	0.60	0.72
0.68	0.60	0.52	0.38	0.36	0.42	0.48	0.52	0.81	0.60
0.50	0.46	0.38	0.49	0.35	0.41	0.24	0.44	0.52	0.40
0.55	0.53	0.62	0.38	0.60	0.53	0.72	0.37	0.57	0.55
0.75	0.71	0.67	0.70	0.70	0.69	0.69	0.71	0.77	0.72

vor.

th day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

decreased slowly to 5.3 until 60 days of curing but in the normal and high acid Cheddar, pH values were found to decline only during first 10-12 days. Dolby, McDowall, and Riddet (25) indicated that since in most cases lactose disappeared in 7-10 days there was no further acid production and consequently no lowering of pH. In this investigation, however, lactose was found to persist for longer periods. So the pH values also continued to decrease over a longer period.

There was a good agreement with pH and lactic acid contents of both brine- and air-cooled cheeses. Dolby, McDowall, and Riddet (25) indicated that there was close agreement of pH and titratable acidity if the pH was above 4.9. In this investigation, however, the acidity was measured colorimetrically as lactic acid, and the buffering action of cheese did not influence the lactic acid measurement. The reason for high pH and high lactic acid at 3 months in normal acid cheese was not clearly understood. The total bacterial count and enterococcus count were found to be relatively higher in these cheeses from 2-3 months period. This may account for the higher amount of lactic acid at 3 months.

Although no significant differences could be established in numbers of different bacterial groups, between the rapidly cooled and conventionally cooled blocks, the pH values and lactic acid content exhibited significant differences. All air-cooled cheeses had lower pHs and higher lactic acid than the brine-cooled blocks. Rapid cooling probably controlled the growth and microbial metabolism and as a result rapid conversion of lactose to lactic acid could not take place. On

the other hand, in warm temperature during early stage of curing, it is probable that certain other groups of microorganisms not determined in this experiment converted the available sugars to lactic acid. In later stages of ripening the increase in pH may have been caused by the neutralizing effect of proteolysis (liberation of amino, guanidyl, and other basic groups and free ammonia). Uncontrolled fermentation of sugars and production of excessive amounts of lactic acid may also contribute to high acid flavor and acid cut or ununiformity of color.

Sugar content of experimental cheese

Sugar determinations were done to measure the effect of cooling rates and pressing times on the fermentative metabolism of cheese flora. Lactic acid is reported to be the principal product of sugar fermentation. Controlled fermentations of sugars would produce lactic acid and other by-products in right proportions to give characteristic flavor to cheese. Holding cheese blocks at warm temperature may cause excessive production of lactic acid which may affect flavor, body and texture and color of cheese.

Lactose content Lactose was found to persist in cheese for longer periods of time than usually reported in the literature (53, 75, 82). The data are presented in Tables 16, 17, and 18. Average lactose contents at milling for high acid, normal acid, and experimental fruity cheese, were 4,200.0, 4,100.0, and 3,300.0 μmole/100 g cheese, respectively at milling. As ripening continued, the lactose content gradually fell. A substantial amount of lactose was still present after 3 months of curing. A higher amount of lactose was present in the brine-cooled

Table 16. Lactose content of high acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled: Cheese lots A	B	C	Avg	Air cooled: A	B	C	Avg	Cheese lots A
									μmole per 100 g
1	4300.0	4900.0	3300.0	4200.0					
2	1900.0	4400.0	2400.0	2900.0					
3									1900.0
4	1500.0	2200.0	2600.0	2100.0	980.0	1600.0	2000.0	1500.0	1400.0
5	740.0	2000.0	2300.0	1700.0	510.0	960.0	1800.0	1100.0	720.0
6	540.0	2000.0	2000.0	1500.0	300.0	1200.0	1200.0	900.0	730.0
7	290.0	1600.0	1700.0	1200.0	140.0	940.0	1400.0	810.0	270.0
8	170.0	1800.0	1600.0	1200.0	100.0	1100.0	1200.0	800.0	150.0
9	180.0	1700.0	1600.0	1200.0	100.0	1200.0	950.0	800.0	190.0
10	170.0	1200.0	1500.0	1000.0	97.0	970.0	1200.0	800.0	150.0

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day; 7-2

Treatment								
	Pressed 20 hr							
	Brine cooled				Air cooled			
Cheese lots								
Avg	A	B	C	Avg	A	B	C	Avg
μmole per 100 g								
	1900.0	3600.0	2400.0	2600.0				
0 1500.0	1400.0	2200.0	2200.0	1900.0	630.0	1900.0	2200.0	1600.0
0 1100.0	720.0	2100.0	2200.0	1700.0	330.0	1400.0	2300.0	1300.0
0 900.0	730.0	2100.0	1900.0	1600.0	320.0	1000.0	2100.0	1200.0
0 810.0	270.0	1700.0	1400.0	1100.0	140.0	740.0	2300.0	740.0
0 300.0	150.0	1500.0	1200.0	920.0	140.0	1300.0	1100.0	850.0
0 800.0	190.0	1600.0	1500.0	1100.0	140.0	1300.0	1400.0	920.0
0 800.0	150.0	1400.0	1300.0	950.0	110.0	1200.0	1200.0	840.0

ay; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 17. Lactose content of normal acid cheese[a]

Sampling period[b]	Treatme							
	Pressed 4 hr							
	Brine cooled				Air cooled			
	Cheese l							
	D	E[c]	F	Avg	D	E	F	Avg
	μ mole pe							
1	4200.0	4100.0	4000.0	4100.0				
2	3500.0	3400.0	2800.0	3200.0				
3								
4	3000.0	2800.0	2400.0	2700.0	2400.0	2100.0	2200.0	2300.0
5	3000.0	2600.0	2200.0	2600.0	2100.0	1900.0	1900.0	2000.0
6	2900.0	2800.0	2200.0	2600.0	2700.0	2100.0	1800.0	2200.0
7	2400.0	2100.0	1700.0	2100.0	2000.0	1600.0	1500.0	1700.0
8	2200.0	1900.0	1900.0	2000.0	1900.0	1300.0	1500.0	1600.0
9	1800.0	1400.0	1800.0	1700.0	1700.0	1000.0	1400.0	1400.0
10	1700.0	390.0	1800.0	1300.0	1600.0	99.0	1600.0	1100.0

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th

[c]Raw milk cheese.

Treatment									
		Pressed 20 hr							
d		Brine cooled				Air cooled			
	Cheese lots								
F	Avg	D	E	F	Avg	D	E	F	Avg
	μ mole per 100 g								
		2700.0	2500.0	2600.0	2600.0				
200.0	2300.0	2500.0	2400.0	2200.0	2400.0	2100.0	1900.0	2000.0	2000.0
900.0	2000.0	2400.0	2000.0	2200.0	2200.0	1900.0	1400.0	1600.0	1600.0
800.0	2200.0	2800.0	2300.0	2000.0	2400.0	1900.0	1400.0	1700.0	1600.0
500.0	1700.0	2500.0	2000.0	1400.0	2000.0	1700.0	1600.0	1300.0	1500.0
500.0	1600.0	2200.0	1800.0	1800.0	1900.0	1800.0	1300.0	1400.0	1500.0
400.0	1400.0	1800.0	1500.0	1600.0	1700.0	1400.0	900.0	1300.0	1200.0
600.0	1100.0	1500.0	160.0	1600.0	1000.0	1000.0	74.0	740.0	610.0

8th day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 18. Lactose content of experimental fruity cheese[a]

Sampling period[b]	Treatment								
	Pressed 4 hr								
	Brine cooled				Air cooled				
	Cheese lot								
	G	H	I	Avg	G	H	I	Avg	G
	μmole per 100 g								
1	3300.0	3200.0	3300.0	3300.0					
2	3300.0	3100.0	3100.0	3100.0					
3									3000
4	3100.0	2800.0	2800.0	2900.0	2300.0	2500.0	2300.0	2400.0	2700
5	2700.0	2400.0	2800.0	2700.0	2100.0	2000.0	2400.0	2200.0	2500
6	2600.0	2500.0	2700.0	2600.0	2400.0	1800.0	2400.0	2200.0	2100
7	2100.0	1900.0	2100.0	2000.0	1800.0	1700.0	1800.0	1800.0	1400
8	1100.0	1800.0	1800.0	1600.0	1300.0	1400.0	1200.0	1300.0	1600
9	2000.0	1600.0	1700.0	1800.0	1300.0	1200.0	1200.0	1200.0	1600
10	1400.0	1600.0	1800.0	1600.0	1400.0	1400.0	1500.0	1400.0	1500

[a]Made from grade-A milk with a culture producing fruity flavor.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day;

Treatment									
		Pressed 20 hr							
ed		Brine cooled				Air cooled			
Cheese lots									
I	Avg	G	H	I	Avg	G	H	I	Avg
= μmole per 100 g									
		3000.0	2900.0	3000.0	2900.0				
2300.0	2400.0	2700.0	2500.0	2900.0	2700.0	2200.0	2400.0	2400.0	2400.0
2400.0	2200.0	2500.0	2800.0	2600.0	2600.0	1700.0	1700.0	2200.0	1900.0
2400.0	2200.0	2100.0	2400.0	2700.0	2400.0	2100.0	1700.0	2100.0	1900.0
1800.0	1800.0	1400.0	1900.0	2300.0	1900.0	1400.0	1400.0	2000.0	1600.0
1200.0	1300.0	1600.0	1800.0	1800.0	1700.0	1300.0	1500.0	1900.0	1600.0
1200.0	1200.0	1600.0	1700.0	1700.0	1600.0	1900.0	1400.0	1400.0	1500.0
1500.0	1400.0	1500.0	1600.0	1700.0	1600.0	1000.0	1000.0	1400.0	1200.0

avor.

8th day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

cheese than in air-cooled cheese during all stages of ripening. Higher concentrations of lactose also were observed at all stages of curing in cheeses pressed 4 hr than the 20-hr pressed cheeses with the same temperature treatment.

The lactose contents of 20-hr pressed brine-cooled cheeses were much higher than both 4-hr and 20-hr pressed air-cooled cheeses.

Statistical analysis of the data indicated that the pressing time and cooling rate produced a significant difference at the 1% level in the rate of lactose disappearance and in the amount present during the curing period. The highest value for the rate of lactose disappearance was observed in the high acid cheese made from manufacturing-grade milk and the lowest in the experimental fruity cheese made from grade-A milk. In the normal acid cheese the rate was intermediate. From Table 17, it appears that lactose dissipates more rapidly in raw milk cheese (lot E) than in heat treated milk cheese. Lot E made from raw milk had 4,000.0 μmole/100g at milling. After 3 months of curing, the 4-hr and 20-hr pressed, brine-cooled cheeses contained 390.0 and 160.0 μmole/100g respectively and air-cooled cheeses contained only 99.0 and 74.0 μmole/100g respectively. But cheeses made under the same conditions from heat treated manufacturing-grade milk had 1800.0 and 1600.0 μmole/100g when brine cooled, and 1600.0 and 700.0 μmole/100g when air cooled. These discrepancies may be attributed to differences in the number of the adventitious flora in the raw milk cheese.

The results of this investigation contradict the earlier belief that lactose disappears within the first 2 weeks after manufacture of

cheese (57, 58, 66, 76, 81, 88). The average lactose content of all cheeses was 2200.0 µmole/100g (ranging from 630.0 to 3,000.0 µmole/100g). At the end of 3 months, the average lactose content of all cheeses was 1,100.0 µmole/100g (ranging from 74.0 to 1800.0 µmole/100g). It appears that the rate of lactose breakdown is greatly influenced by the type of milk and the number of microorganisms present. The raw milk cheese (lot E) had a high total count (Table 4), enterococcus count (Table 6), and gram-negative bacterial count (Table 8). Consequently lactose breakdown was rapid in raw milk cheese. The same observation also holds true for the high acid cheeses.

Fagen, Stine, and Hussong (28) observed that cheese sugars were retained up to 53 days in pasteurized milk cheese but had disappeared within 25 days in raw milk cheese. In this investigation, however, lactose breakdown was greater in raw milk cheese but did not disappear completely at the end of 3 months.

High cooking temperature was reported to influence the retention of sugars in Svecia type cheese (2, 76). This was explained to be due to destruction of adventitious flora and slowing down the rate of growth of added culture. In this investigation, statistically significant differences could not be shown in the numbers of different types of organisms between different rates of cooling or periods of pressing, but the sugar fermentation and acid production were found to be significant. It appears that high temperature during the initial stage of cheese curing may be responsible for the rapid acid production and consequently rapid disappearance of sugar. Rapid cooling, on the other

hand, slows down the rate of bacterial metabolism and consequently less fermentation occurs. In essence the temperature seems to affect the metabolic rate, but not the growth rate.

Glucose content Since glucose is a breakdown product of lactose, the disappearance of glucose is similar to lactose. The glucose contents of the high acid, normal acid, and experimental cheeses at different sampling periods are reported in Tables 19, 20, and 21 respectively. The maximum glucose concentrations were observed at milling; 110.0, 200.0, and 170.0 μmole/100g, respectively, in high acid, normal acid, and experimental fruity cheese. The amounts of glucose after milling were not generally higher than 50.0 μg/100g cheese. Although rate of hydrolysis of lactose in air-cooled cheese was greater, the amount of glucose detected at different sampling periods was found to be lower than that found in brine-cooled cheeses. This indicated that the rate of glucose breakdown was much faster in the air-cooled cheese.

Statistical analysis of the data indicated that the amount of glucose present in the brine-cooled cheese was significantly higher at the 5% level than in the air-cooled cheese. Pressing time, however, did not have any significant effect on the amount of glucose present.

In contrast to the present finding, Raadsveld (66), Sjöström (76) and Anderson, Nilsson, and Sjöström (2) could not demonstrate glucose in cheese tested 1 day after manufacture. The lack of agreement was obviously due to the insensitivity of their methods in the detection of glucose, galactose, and lactose in cheese. Increase in the amount

Table 19. Glucose content of high acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lot A	B	C	Avg	Air cooled, Cheese lot A	B	C	Avg	Cheese lot
	μ mole per								
1	130.0	62.0	140.0	110.0					
2	39.0	25.0	48.0	37.0					
3									3
4	31.0	74.0	41.0	49.0	40.0	24.0	29.0	31.0	2
5	41.0	19.0	20.0	27.0	37.0	34.0	20.0	31.0	4
6	28.0	8.9	26.0	21.0	37.0	2.8	42.0	27.0	4
7	12.0	8.9	180.0	68.0	19.0	19.0	110.0	51.0	2
8	16.0	44.0	180.0	80.0	6.0	23.0	170.0	66.0	1
9	43.0	68.0	64.0	59.0	25.0	29.0	48.0	34.0	4
10	33.0	96.0	97.0	75.0	33.0	46.0	58.0	44.0	2

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th da

Treatment								
	Pressed 20 hr							
	Brine cooled				Air cooled			
Cheese lots								
Avg	A	B	C	Avg	A	B	C	Avg
μ mole per 100 g								
	37.0	23.0	27.0	29.0				
31.0	28.0	32.0	27.0	29.0	31.0	22.0	20.0	24.0
31.0	41.0	22.0	22.0	28.0	37.0	24.0	20.0	27.0
27.0	41.0	14.0	30.0	28.0	28.0	8.9	38.0	25.0
51.0	22.0	17.0	170.0	70.0	25.0	50.0	140.0	72.0
66.0	12.0	17.0	170.0	67.0	12.0	19.0	110.0	48.0
34.0	43.0	44.0	90.0	59.0	40.0	32.0	64.0	46.0
44.0	28.0	55.0	87.0	57.0	22.0	19.0	64.0	35.0

day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 20. Glucose content of normal acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled — Cheese lots: D	E[c]	F	Avg	Pressed 4 hr, Air cooled — D	E	F	Avg	Cheese lots: D
	μmole per 100 g								
1	110.0	210.0	270.0	200.0					
2	44.0	43.0	70.0	52.0					
3									38.0
4	49.0	28.0	73.0	50.0	41.0	26.0	49.0	39.0	38.0
5	46.0	26.0	54.0	42.0	31.0	11.0	52.0	31.0	32.0
6	55.0	23.0	34.0	38.0	54.0	25.0	31.0	37.0	58.0
7	68.0	26.0	34.0	43.0	48.0	21.0	27.0	32.0	57.0
8	59.0	44.0	18.0	40.0	76.0	15.0	28.0	40.0	100.0
9	68.0	28.0	33.0	43.0	71.0	28.0	32.0	43.0	65.0
10	54.0	18.0	27.0	33.0	43.0	28.0	21.0	30.0	68.0

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day; 7-21s

[c]Raw milk cheese.

Treatment								
	Pressed 20 hr							
	Brine cooled				Air cooled			
Cheese lots								
Avg	D	E	F	Avg	D	E	F	Avg
μ mole per 100 g								
	38.0	33.0	49.0	40.0				
39.0	38.0	21.0	43.0	34.0	52.0	21.0	41.0	38.0
31.0	32.0	14.0	64.0	37.0	34.0	4.0	55.0	31.0
37.0	58.0	31.0	33.0	41.0	63.0	31.0	33.0	42.0
32.0	57.0	18.0	32.0	35.0	68.0	28.0	22.0	39.0
40.0	100.0	43.0	28.0	57.0	91.0	28.0	22.0	47.0
43.0	65.0	32.0	37.0	45.0	68.0	40.0	22.0	43.0
30.0	68.0	19.0	25.0	37.0	84.0	11.0	31.0	42.0

y; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 21. Glucose content of experimental fruity cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled — Cheese lots G	H	I	Avg	Pressed 4 hr, Air cooled — Cheese lots G	H	I	Avg	Cheese lots G
	μ mole per 100								
1	140.0	140.0	230.0	170.0					
2	48.0	88.0	76.0	71.0					
3									73.(
4	53.0	31.0	48.0	44.0	43.0	28.0	50.0	40.0	65.(
5	62.0	31.0	33.0	42.0	54.0	38.0	50.0	47.0	53.(
6	65.0	44.0	28.0	46.0	54.0	42.0	28.0	41.0	65.(
7	50.0	81.0	65.0	65.0	48.0	65.0	56.0	56.0	39.(
8	77.0	24.0	25.0	42.0	56.0	21.0	28.0	35.0	53.(
9	34.0	50.0	40.0	42.0	28.0	53.0	31.0	37.0	34.(
10	27.0	43.0	33.0	34.0	27.0	33.0	25.0	28.0	28.(

[a]Made from grade-A milk with a culture producing fruity flavor.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day;

Treatment									
		Pressed 20 hr							
ł		Brine cooled				Air cooled			
	Cheese lots								
I	Avg	G	H	I	Avg	G	H	I	Avg
	μ mole per 100 g								
		73.0	90.0	65.0	76.0				
50.0	40.0	65.0	31.0	41.0	45.0	38.0	14.0	44.0	32.0
50.0	47.0	53.0	44.0	35.0	44.0	53.0	49.0	32.0	45.0
28.0	41.0	65.0	56.0	28.0	50.0	50.0	56.0	24.0	43.0
56.0	56.0	39.0	51.0	50.0	47.0	33.0	21.0	56.0	36.0
28.0	35.0	53.0	31.0	32.0	38.0	28.0	33.0	40.0	34.0
51.0	37.0	34.0	46.0	50.0	43.0	31.0	41.0	43.0	38.0
25.0	28.0	28.0	39.0	23.0	30.0	18.0	28.0	28.0	25.0

avor.

8th day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

of glucose present at different sampling periods of this experiment could be due to increase in the rate of lactose breakdown.

Galactose content Disappearance of galactose also followed the same pattern as glucose and lactose. Tables 22, 23, and 24 show the galactose contents of high acid, normal acid, and experimental fruity cheese. Like glucose, the galactose content was higher at milling; 190.0, 120.0, and 230.0 μmole/100g cheese in high acid, normal acid and experimental fruity cheese, respectively. Galactose content during the remaining period ranged from 50-100 μmole/100g. Statistical analysis of the data indicated that all 4-hr pressed cheeses contained higher amounts of galactose than 20-hr pressed cheeses at the 5% level of significance. All brine-cooled cheeses also contained higher amounts of galactose than the air-cooled at the 1% level of significance. The variation in the amount of galactose present in different types of cheese was not statistically significant. The rate of galactose disappearance was slower compared to glucose. Sjöström (76) reported that galactose remained for a relatively long time (22 days) in Herrgård, Prastost, Svecia, Cheddar, and Camembert cheese although lactose and glucose had disappeared within 3 days. Raadsveld (66) also indicated that galactose remains relatively longer than glucose or lactose in cheese. None of these investigators could demonstrate the presence of galactose beyond 3 weeks of ripening. From this investigation it appears that galactose, glucose, and lactose contents are relatively higher during the first 3 weeks. These anomalies may be due to the application of relatively insensitive colorimetric and/or chromatographic methods by those investigators. The enzymatic method used in this

Table 22. Galactose content of high acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots A	B	C	Avg	Pressed 4 hr, Air cooled, Cheese lots A	B	C	Avg	Treatment, Cheese lots A
	μmole per 100 g								
1	180.0	160.0	220.0	190.0					
2	100.0	94.0	120.0	110.0					
3									69.0
4	130.0	108.0	66.0	100.0	67.0	32.0	54.0	51.0	65.0
5	82.0	54.0	92.0	76.0	49.0	22.0	45.0	39.0	43.0
6	70.0	33.0	92.0	65.0	43.0	27.0	41.0	37.0	49.0
7	14.0	33.0	150.0	64.0	22.0	27.0	92.0	47.0	20.0
8	7.0	69.0	150.0	74.0	16.0	69.0	140.0	74.0	7.0
9	51.0	64.0	29.0	48.0	33.0	30.0	26.0	30.0	37.0
10	47.0	51.0	44.0	47.0	28.0	28.0	29.0	29.0	36.0

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day; 7-2

Treatment									
		Pressed 20 hr							
cooled		Brine cooled				Air cooled			
Cheese lots									
C	Avg	A	B	C	Avg	A	B	C	Avg
μmole per 100 g									
		69.0	65.0	62.0	65.0				
54.0	51.0	65.0	23.0	57.0	48.0	56.0	23.0	58.0	46.0
45.0	39.0	43.0	82.0	52.0	59.0	45.0	44.0	49.0	46.0
41.0	37.0	49.0	36.0	57.0	47.0	42.0	30.0	48.0	40.0
92.0	47.0	20.0	30.0	140.0	62.0	19.0	54.0	110.0	62.0
140.0	74.0	7.0	51.0	134.0	64.0	15.0	36.0	91.0	47.0
26.0	30.0	37.0	52.0	24.0	38.0	33.0	46.0	21.0	33.0
29.0	29.0	36.0	28.0	32.0	32.0	28.0	31.0	19.0	26.0

ay; 5-8th day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 23. Galactose content of normal acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots D	E[c]	F	Avg	Pressed 4 hr, Air cooled, Cheese lots D	E	F	Avg	Br D	Br
	μ mole per 100 g									
1	85.0	140.0	140.0	120.0						
2	47.0	36.0	88.0	57.0						
3									33.0	2
4	52.0	84.0	93.0	77.0	77.0	22.0	100.0	67.0	41.0	2
5	52.0	41.0	140.0	76.0	36.0	22.0	100.0	53.0	22.0	3
6	55.0	49.0	100.0	68.0	47.0	28.0	100.0	58.0	52.0	3
7	47.0	52.0	100.0	68.0	11.0	36.0	50.0	32.0	13.0	4
8	31.0	36.0	72.0	46.0	22.0	28.0	57.0	35.0	13.0	1
9	36.0	33.0	66.0	45.0	31.0	31.0	53.0	38.0	31.0	3
10	31.0	33.0	36.0	33.0	28.0	31.0	28.0	29.0	25.0	3

[a]Made from manufacturing-grade milk.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day; 7-21st

[c]Raw milk cheese.

Treatment									
	Pressed 20 hr								
	Brine cooled				Air cooled				
Cheese lots									
	Avg	D	E	F	Avg	D	E	F	Avg
μ mole per 100 g									
		33.0	22.0	88.0	48.0				
.0	67.0	41.0	22.0	100.0	55.0	49.0	22.0	74.0	49.0
.0	53.0	22.0	31.0	120.0	58.0	14.0	22.0	104.0	47.0
.0	58.0	52.0	33.0	100.0	63.0	39.0	31.0	80.0	50.0
.0	32.0	13.0	47.0	50.0	37.0	31.0	38.0	44.0	38.0
.0	35.0	13.0	19.0	63.0	32.0	19.0	47.0	44.0	37.0
.0	38.0	31.0	31.0	53.0	38.0	42.0	11.0	44.0	32.0
.0	29.0	25.0	31.0	66.0	41.0	13.0	28.0	28.0	23.0

h day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

Table 24. Galactose content of experimental fruity cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled G	H	I	Avg	Air cooled G	H	I	Avg	Cheese lot
	μmole per								
1	300.0	158.0	240.0	230.0					
2	160.0	150.0	210.0	170.0					
3									1(
4	91.0	78.0	150.0	110.0	110.0	47.0	140.0	97.0	1
5	140.0	56.0	180.0	130.0	91.0	47.0	140.0	92.0	1
6	120.0	30.0	140.0	97.0	99.0	11.0	120.0	77.0	1
7	110.0	56.0	100.0	88.0	58.0	30.0	120.0	70.0	6
8	110.0	89.0	110.0	110.0	83.0	67.0	86.0	79.0	1(
9	22.0	30.0	110.0	55.0	33.0	22.0	120.0	58.0	2
10	36.0	22.0	160.0	73.0	44.0	36.0	140.0	74.0	3

[a]Made from grade-A milk with a culture producing fruity flavor.

[b]1-Milling; 2-Pressed 4 hr; 3-Pressed 20 hr; 4-4th day; 5-8th day; 6-12th day

Treatment									
	Pressed 20 hr								
	Brine cooled				Air cooled				
Cheese lots	Avg	G	H	I	Avg	G	H	I	Avg
μmole per 100 g									
		100.0	140.0	180.0	140.0				
)	97.0	140.0	36.0	190.0	120.0	100.0	44.0	150.0	98.0
)	92.0	120.0	47.0	140.0	100.0	67.0	36.0	160.0	89.0
)	77.0	120.0	42.0	120.0	94.0	72.0	11.0	130.0	70.0
)	70.0	67.0	53.0	140.0	85.0	83.0	33.0	130.0	81.0
)	79.0	100.0	67.0	120.0	96.0	78.0	47.0	110.0	78.0
)	58.0	28.0	36.0	100.0	55.0	31.0	30.0	120.0	60.0
)	74.0	36.0	36.0	160.0	76.0	36.0	11.0	150.0	65.0

)r.

day; 6-12th day; 7-21st day; 8-1 month; 9-2 months; 10-3 months.

investigation is very specific and highly sensitive. As low as 0.001% galactose, 0.002% glucose, and 0.004% lactose could be determined by this method with a reasonable accuracy.

The reason for lower amount of lactose, glucose, and galactose in air-cooled cheeses was obviously due to faster rate of sugar utilization either by starter bacteria or other adventitious microorganisms. However, from bacterial counts, it appears that the rate of growth of microorganisms was unaffected by the different cooling rates.

Free fatty acids of experimental cheese

Fatty acids in cheese are formed as a result of milk fat breakdown to a large extend by microorganisms. Starter bacteria are non-lipolytic and hence the major portion of lipolysis is effected by adventitious flora such as lactobacilli and microccocci (71, 72). Fatty acids in right proportions are considered essential for characteristic Cheddar flavor but excessive amounts can also impart undesirable flavor defects (55, 59, 60).

Total free fatty acid content of the cheese was expressed as ml of 0.01 N alcoholic KOH required to neutralize the free fatty acids in 5 g cheese. The results appear in Tables 25, 26, and 27. The normal acid cheese (lots D, E, and F) contained higher amounts of total free fatty acids than did the high acid (lots A, B, and C) cheese. The experimental fruity cheeses contained the lowest amounts. These differences were statistically significant at the 1% level. The average total free fatty acids at 4 hr pressing in high acid, normal acid, and experimental fruity cheese were 3.25, 4.82, and 2.75, expressed as ml

Table 25. Free fatty acid content[a] of high acid cheese[b]

Sampling period[c]	Treatment: Pressed 4 hr, Brine cooled A	B	C	Avg	Air cooled A	B	C	Avg	Cheese lot
1	3.88	3.09	2.79	3.25					
2									1
3	7.70	4.13	4.72	5.52	7.25	4.49	4.72	5.49	6
4	7.65	6.42	5.00	6.36	7.25	7.08	4.79	6.37	
5	7.45	6.91	4.76	6.37	8.29	6.45	4.72	6.49	8

[a]Expressed as ml 0.01N KOH required to neutralize fatty acids from 5 g of che

[b]Made from manufacturing-grade milk.

[c]1-Pressed 4 hr; 2-Pressed 20 hr; 3-1 month; 4-2 months; 5-3 months.

Treatment								
	Pressed 20 hr							
	Brine cooled				Air cooled			
Cheese lots								
Avg	A	B	C	Avg	A	B	C	Avg
	4.32	3.12	3.21	3.55				
5.49	6.94	4.11	4.95	5.33	7.45	5.13	5.05	5.88
6.37	7.42	5.17	4.71	5.77	7.40	5.81	4.93	6.05
6.49	8.24	7.01	6.22	7.17	6.48	7.14	6.27	6.63

rom 5 g of cheese.

nths.

Table 26. Free fatty acid content[a] of normal acid cheese[b]

Sampling period[c]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots D	E[d]	F	Avg	Air cooled, Cheese lots D	E	F	Avg	Cheese lots D
1	4.50	3.55	6.41	4.82					
2									4.52
3	5.92	5.18	8.24	6.45	6.34	6.87	7.71	6.97	8.10
4	6.26	5.91	11.19	7.79	7.48	7.11	9.71	8.10	6.50
5	10.47	6.41	10.18	9.02	12.73	7.19	12.30	10.74	11.69

[a]Expressed as ml 0.01N KOH required to neutralize fatty acids from 5 g of cheese.

[b]Made from manufacturing-grade milk.

[c]1-Pressed 4 hr; 2-Pressed 20 hr; 3-1 month; 4-2 months; 5-3 months.

[d]Raw milk cheese.

Treatment								
	Pressed 20 hr							
	Brine cooled				Air cooled			
Cheese lots								
Avg	D	E	F	Avg	D	E	F	Avg
	4.52	3.56	5.62	4.57				
6.97	8.10	6.37	9.93	8.13	6.26	7.91	10.56	8.24
8.10	6.50	6.90	11.83	8.41	8.81	10.75	11.26	10.27
10.74	11.69	7.07	10.54	9.77	14.55	12.68	11.78	13.00

rom 5 g of cheese.

nths.

Table 27. Free fatty acid content[a] of fruity cheese[b]

Sampling period[c]	Treatment: Pressed 4 hr, Brine cooled G	H	I	Avg	Air cooled G	H	I	Avg	Cheese lots G
1	3.73	2.58	2.43	2.91					
2									3.57
3	3.77	2.85	2.49	3.04	3.91	3.36	3.23	3.50	3.98
4	3.70	3.91	4.78	4.13	4.08	4.41	4.72	4.40	4.07
5	3.85	4.68	4.74	4.42	3.41	6.02	5.53	4.99	4.88

[a]Expressed as ml 0.01N KOH.

[b]Made from grade-A milk with a culture producing fruity flavor.

[c]1-Pressed 4 hr; 2-Pressed 20 hr; 3-1 month; 4-2 months; 5-3 months.

Treatment									
		Pressed 20 hr							
)oled		Brine cooled				Air cooled			
Cheese lots									
I	Avg	G	H	I	Avg	G	H	I	Avg
		3.57	2.24	2.44	2.75				
3.23	3.50	3.98	3.00	2.86	3.28	5.03	2.77	3.60	3.80
4.72	4.40	4.07	4.56	4.70	4.44	3.97	3.66	4.74	4.12
5.53	4.99	4.88	6.25	4.78	5.30	5.21	6.75	4.90	5.62

flavor.

; 5-3 months.

of 0.01 N KOH, respectively. As the ripening progressed, the free fatty acid content increased significantly. The brine-cooled cheeses contained significantly lower amounts of free fatty acids than the air-cooled cheeses. Pressing time also had a highly significant effect on the amount of fatty acids, 20 hr pressed being higher in fatty acids than the 4 hr pressed cheeses.

Ohren and Tuckey (60) indicated that free fatty acids increased with an increasing bacterial count in cheese. In this investigation, all bacterial counts appeared to be higher in normal acid cheese than high acid cheese or fruity cheese. The fatty acid contents also followed the same pattern as the bacterial counts. Raw milk cheese, however, did not contain high free acids as reported by Ohren and Tuckey (60).

High amounts of free fatty acids in normal acid cheese compared to high acid cheese were probably due to the presence of large numbers of adventitious flora in normal acid cheese. High acid cheese containing high lactic acid did not probably favor the growth of lipolytic organisms.

Fatty acids along with alcohol, may form esters, and may adversely effect the flavor. This may also be a reason for lower amounts of fatty acids in experimental fruity cheese.

The higher amount of fatty acids in air-cooled cheeses may be due to the favorable condition for the growth of lactobacilli during early stage of curing. Ohren and Tuckey (60) observed fermented, unclean, and whey taint flavor defects in cheese containing high contents

of C_{10}, C_{12}, and C_{14} fatty acids. Air-cooled cheeses were criticised for these defects at 3-and 6-month judging.

Measurement of proteolysis

Degree of proteolysis has been considered as the indicative of cheese ripening (19, 23, 56, 80). Protein is broken down by milk protease, rennet, and bacterial proteinases. Besides other adventitious microorganisms, starter organisms itself are slightly proteolytic. Proteolysis was determined to observe the relationship between temperature during early stage of curing and maturation of cheese. Excessive proteolysis or incomplete proteolysis, on the other hand, impart objectionable flavor in cheese (16, 26, 45).

Proteolysis was measured by using an orange G dye binding technique (32); results were expressed as per cent proteolysis. Data are presented in Tables 28, 29, and 30. The extent of proteolysis increased with maturing of the cheese. The average per cent proteolysis in high acid cheese (lots A, B, and C), after 1 month ranged from 6.3 to 7.8% among brine- and air-cooled cheeses. After 6 months it ranged from 30.1 to 32.9%. The average proteolysis index of normal acid cheese (lots D, E, and F, Table 29) after 1 month ranged from 11.7 to 12.8% among brine- and air-cooled cheeses and after 6 months they ranged from 25.6 to 27.9%. The extent of proteolysis was lowest in experimental fruity cheese. After 1 month average proteolysis of this cheese ranged from 5.8 to 8.6% and after 6 months ranged from 15.8 to 19.8% among brine- and air-cooled cheeses.

Table 28. Comparison of proteolysis of high acid cheese[a]

Sampling period[b]	Treatment								
	Pressed 4 hr								
	Brine cooled				Air cooled				
	Cheese lots								
	A	B	C	Avg	A	B	C	Avg	A
	% Proteolysis								
1	9.2	4.4	9.3	7.6	1.7	8.0	9.3	6.3	10.0
2	15.3	9.7	9.9	11.6	15.9	6.0	9.5	10.5	19.7
3	22.6	10.6	11.9	15.0	18.9	10.2	13.9	14.3	16.6
4	42.0	32.3	22.8	32.4	40.5	32.4	25.7	32.9	38.0

[a]Made from manufacturing-grade milk.

[b]1-1 month; 2-2 months; 3-3 months; 4-over 6 months.

Treatment									
		Pressed 20 hr							
i		Brine cooled				Air cooled			
Cheese lots									
C	Avg	A	B	C	Avg	A	B	C	Avg
% Proteolysis									
).3	6.3	10.0	5.1	8.7	7.9	8.8	6.2	8.4	7.8
).5	10.5	19.7	9.2	11.0	13.3	14.6	12.4	11.4	12.8
).9	14.3	16.6	10.6	13.3	13.5	17.6	6.0	14.3	12.6
).7	32.9	38.0	35.1	21.5	31.5	37.4	28.8	24.2	30.1

Table 29. Comparison of proteolysis of normal acid cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled, Cheese lots D	E[c]	F	Avg	Air cooled, Cheese lots D	E	F	Avg	Cheese lots D
	% Proteolysis								
1	13.0	11.7	10.5	11.7	15.2	12.8	10.5	12.8	16.
2	16.3	13.7	13.1	14.4	15.8	19.3	11.6	15.6	11.
3	21.9	19.6	16.3	19.3	23.3	20.1	17.0	20.1	24.
4	26.9	29.7	21.7	26.1	25.0	19.1	29.6	27.9	27.

[a]Made from manufacturing-grade milk.

[b]1-1 month; 2-2 months; 3-3 months; 4-over 6 months.

[c]Raw milk cheese.

Treatment									
		Pressed 20 hr							
led		Brine cooled				Air cooled			
Cheese lots									
F	Avg	D	E	F	Avg	D	E	F	Avg
% Proteolysis									
10.5	12.8	16.0	11.0	10.1	12.4	14.0	12.8	11.6	12.8
11.6	15.6	11.2	16.6	10.7	12.8	17.5	15.0	12.3	14.9
17.0	20.1	24.2	21.0	16.6	20.6	25.2	18.9	17.1	20.4
29.6	27.9	27.8	29.1	20.7	25.6	28.8	33.8	19.2	27.3

Table 30. Comparison of proteolysis of fruity cheese[a]

Sampling period[b]	Treatment: Pressed 4 hr, Brine cooled: Cheese lots G	H	I	Avg	Air cooled: G	H	I	Avg	Pressed 4 hr: G
	% Proteolysis								
1	7.6	6.6	6.3	6.8	8.1	2.4	7.0	5.8	11.8
2	17.8	8.9	13.6	13.4	17.9	8.3	14.8	13.7	11.9
3	16.4	15.1	14.5	15.3	18.0	14.4	12.0	14.8	17.8
4	17.8	17.1	13.6	16.2	29.3	16.4	13.8	19.8	18.4

[a]Made from grade-A milk with a culture producing fruity flavor.

[b]1-1 month; 2-2 months; 3-3 months; 4-over 6 months.

Treatment								
	Pressed 20 hr							
	Brine cooled				Air cooled			
Cheese lots								
Avg	G	H	I	Avg	G	H	I	Avg
% Proteolysis								
5.8	11.8	6.0	7.8	8.5	11.0	6.6	8.1	8.6
13.7	11.9	7.4	8.9	9.4	20.2	12.1	10.6	14.3
14.8	17.8	12.1	13.6	14.5	20.6	14.2	13.2	16.0
19.8	18.4	15.6	13.5	15.8	19.7	15.6	13.6	16.3

Statistical analysis indicated no significant difference due to cooling rates or pressing times. The F value for the type of cheese and sampling period being highly significant, indicated that rate of proteolysis at different sampling periods was influenced by the type of cheese. The rate was greater towards the later part of the ripening in high acid cheese.

Vakaleris et al. (79) reported the increase of formol nitrogen in Dariworld and Cheddar cheese with the progression of ripening period. They did not find any significant effect of the initial pH of Cheddar cheese on the amount of formol nitrogen. But sweet Dariworld cheese (initial pH 6.4) contained 2 to 3% more soluble nitrogen than normal Dariworld cheese (initial pH 5.4) after 60 days ripening. These differences were considered to be due to the action of bacterial proteases. Total bacterial count of high acid cheese in this investigation was reduced greatly during this 2- to 3-months period and the bacterial endo-enzymes probably were responsible for high proteolysis. Deane (23) and Dahlberg and Kosikowsky (19) reported that the presence of Streptococcus faecalis (an enterococcus) increased the rate of proteolysis. Tyramine content, produced by S. faecalis, also was found to be directly related to the characteristic Cheddar flavor. The normal acid cheese (lots D, E, and F), however, with relatively high enterococcus count did not show any significant difference in the rate of proteolysis in this investigation.

Color measurement of experimental cheese

A comparison of color in cheese in terms of their reflectance spectrum was performed after 3 months of curing. Fig. 7 shows the reflectance spectra of cheese (lot B) made from manufacturing-grade milk. The color differences between cheeses made with different cooling rates and pressing times are apparent from this figure. Fig. 8 shows the reflectance spectra for cheese made from grade-A milk (lot I). Figures 7 and 8 show that color differences among treatments are greater in high acid cheese made from manufacturing-grade milk (Fig. 7) than in experimental fruity cheese (Fig. 8). The reflectances of all cheeses were measured at their maximum absorption at 465 mμ wavelength and are presented in Table 31. These data were analysed statistically. Brine-cooled cheeses showed more pronounced color than the air-cooled cheeses. These differences were highly significant at the 1% level. There was, however, no significant difference in color due to pressing time.

The color differences noted are explainable in the light of sugar breakdown and acidity of cheese. The rate of sugar breakdown, amount of lactic acid, fatty acids, and the hydrogen ion concentration were always higher in air-cooled cheeses. In many instances, the air-cooled cheeses were criticized for acid cut and high acid taste in judging. Irvine (47) reported that high acid cheese always had a lower intensity of color than low acid cheese.

Fig. 7. Reflectance spectra (400-700 mμ) of high acid cheese from lot B made from manufacturing-grade milk

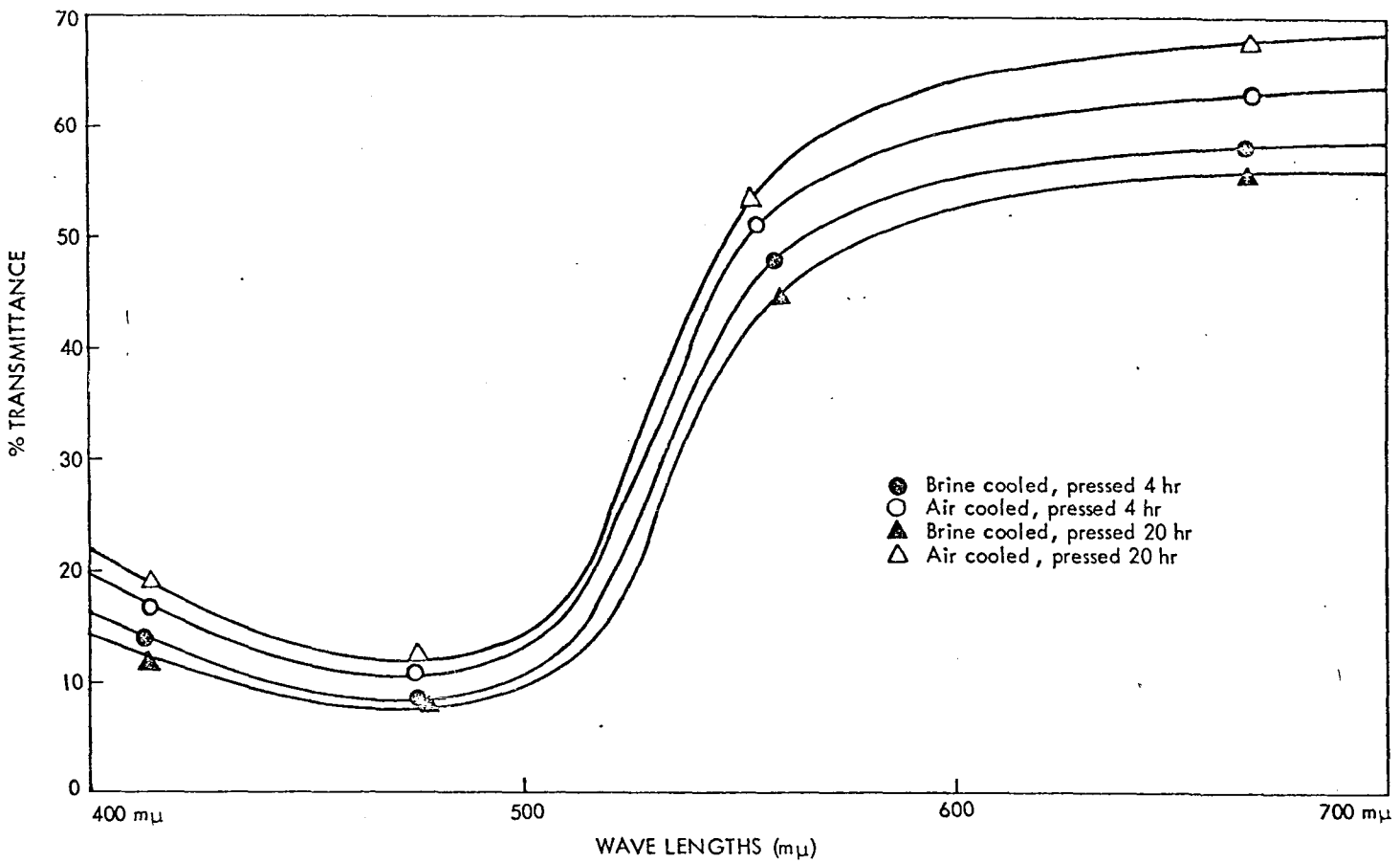

70
60
50
40
30
20
10
0
% TRANSMITTANCE
400 mμ
500
600
700 mμ
WAVE LENGTHS (mμ)
Brine cooled, pressed 4 hr
Air cooled, pressed 4 hr
Brine cooled, pressed 20 hr
Air cooled , pressed 20 hr

Fig. 8. Reflectance spectra (400-700 mμ) of experimental fruity cheese from lot I made from grade-A milk

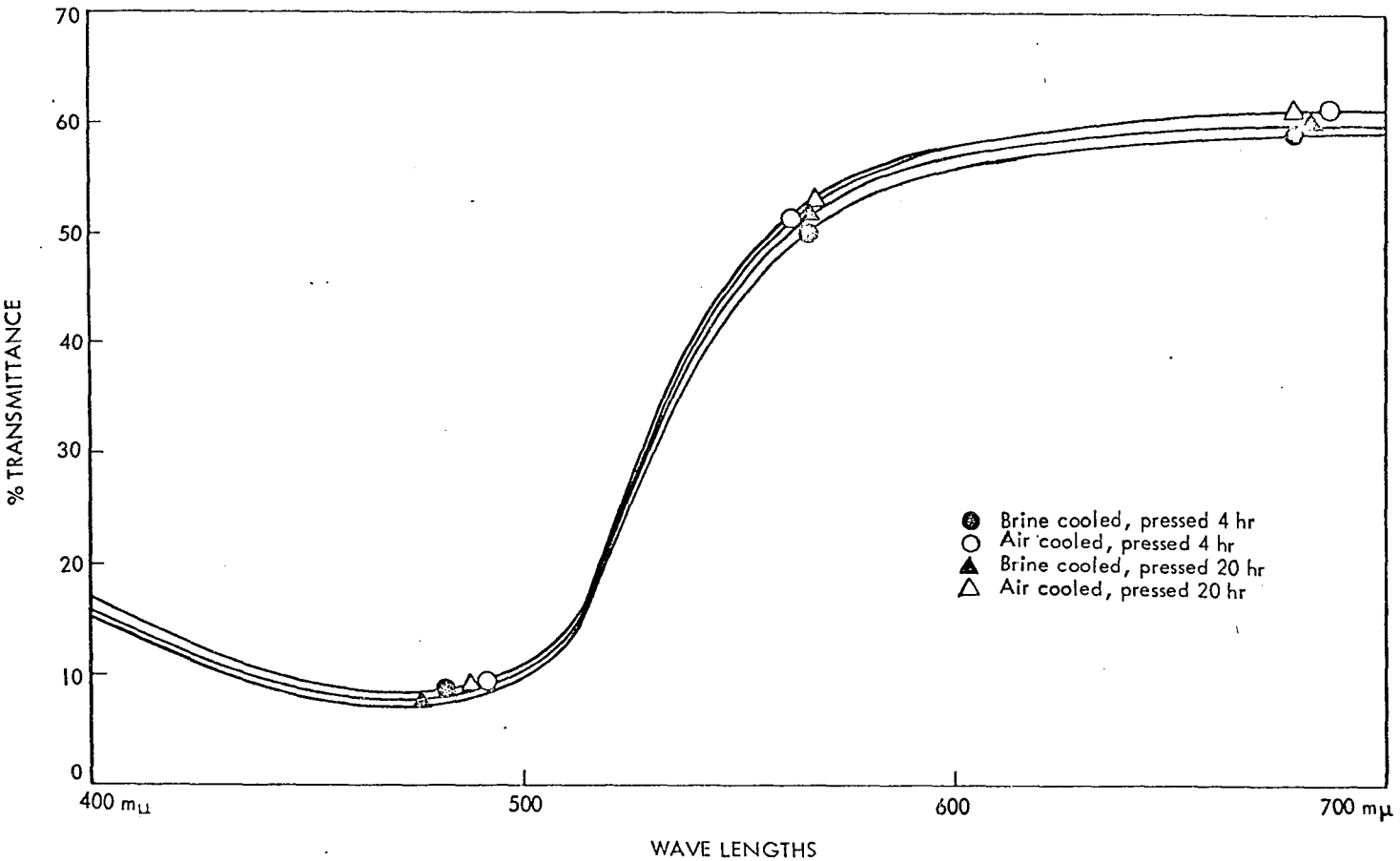

70
60
50
40
30
20
10
0
% TRANSMITTANCE
400 mμ
500
600
700 mμ
WAVE LENGTHS
Brine cooled, pressed 4 hr
Air cooled, pressed 4 hr
Brine cooled, pressed 20 hr
Air cooled, pressed 20 hr

Table 31. Intensity of color measured by reflectance of cheese at 465 mμ after 3 months of curing

Lots	Treatment			
	Pressed 4 hr		Pressed 20 hr	
	Brine cooled	Air cooled	Brine cooled	Air cooled
	Absorbence			
B	1.083	0.989	1.140	0.921
C	1.126	1.112	1.126	1.083
D	0.979	0.969	0.989	0.912
E	1.083	1.059	1.204	1.022
F	1.000	0.989	1.046	1.046
G	1.022	1.022	1.046	1.000
H	1.046	0.989	1.155	1.022
I	1.112	1.083	1.140	1.071

Judging of Cheese

All cheeses were judged at 3 and 6 months of curing using the American Dairy Science Association Intercollegiate score cards. At 3 months in addition to this method, paired comparisons were made among the four treatments (Appendix, p. 133).

Scoring

Judging scores for flavor and body and texture were statistically analysed for all batches of cheese. Cooling rates or pressing times did not have any significant influence in judging scores. There was a significant difference at the 1% level in body and texture scores among normal acid, experimental fruity, normal grade-A milk, and experimental bitter cheese. The normal grade-A milk cheese and bitter experimental cheese had firm and smooth body than normal acid and experimental fruity cheese. Each type of cheese, judged after 6 months had a higher score for flavor and body than at 3 months of curing. These differences were statistically significant.

Since there was a great variation among judges in scoring cheeses subjected to different treatments, the flavor score became inappliable to evaluate their differences. On the basis of the criticisms marked on the score cards, an attempt was made to categorize the defects of different cheeses.

Lot A (high acid cheese) was considered to be of poor quality with high acid and bitter flavor at preliminary judging and was not placed before the panel of judges for judging. But at 6 months a great improvement was noticed; the flavor score then ranged from 36.5 to 39.5 and

body and texture scores ranged from 28 to 29 among the judges. Five of the six judges criticised the 4-hr and 20-hr pressed air-cooled cheeses for high acid and bitter flavor. The brine-cooled cheeses did not receive such criticisms from three judges; three others described the acid or bitter flavor in this cheese to be slight compared to the air-cooled cheeses. In lot C air-cooled cheeses were criticised more severely for high acid than the brine-cooled cheeses by four judges.

LotsD and E (normal acid cheese) made with manufacturing-grade milk, were criticised by four judges for high acid and by 3 judges for fruity flavor defects in all blocks at 6 months. Lot F showed a remarkable difference in flavor, both at 3 and 6 months. Both the 4-hr and 20-hr pressed air-cooled cheeses were severely criticised by all judges for high acid and fruity defects but none of the judges criticised the brine-cooled cheeses of the same pressing periods for such defects.

With regard to cheeses made with "fruity culture", at 3 months none of these cheeses were found to be fruity. At 6 months, three of the six judges indicated that the 4-hr and 20-hr pressed, air-cooled cheeses developed fruity flavor in lot G but not in the brine-cooled cheeses. In lot H, none of the brine-cooled cheeses developed any appreciable fruity flavor but the air-cooled cheeses of both the pressing times were criticised for high acid at 3 and 6 months. Similar to lot G, lot I also received criticisms for high acid when air cooled at both the judging periods. Fruity defects were observed in all treatments (both brine-cooled and air-cooled cheeses) at 6 months only.

Three of the lots of cheese were made in 400-lb. capacity vats under ideal conditions with grade-A milk. All treatments showed equal criticisms for high acid in all lots except for the 4 hr pressed brine-cooled blocks. All these cheeses received the highest scores for flavor and body compared to other lots.

Lots P, Q, and R were made with a "bitter culture" obtained from Dr. Emmons of Canada. Brine-cooled, 4-hr pressed cheese from lot P at 3 months was criticised for bitterness by one judge. Cheeses subjected to the same treatments from lots Q and R were criticised for bitterness by only two and one judges respectively. At 6 months, 4-hr pressed brine-cooled cheese from lot P was criticised for bitterness by two judges and from lots Q and R by four judges. The remainder of the cheeses (4-hr pressed, air-cooled and 20-hr pressed air- and brine-cooled) were criticised for bitterness by four of the seven judges. The number of judges who criticised for high acid flavor was not more than three in lots P and R. But all the blocks of lot Q were criticised for high acid by more than four judges.

The last lot of cheese was made with grade-A milk contaminated with coliform organisms. After 3 months, all blocks received fairly low scores both for flavor and body. All cheeses were considered to have a gassy, weak, and pasty body at this time. The brine-cooled cheese did not have high acid taste according to five of the six judges, but the flavor was considered to be flat. Four of the six judges criticised the air-cooled blocks as high acid and two of them considered these also to be fruity. At 6 months, except 4-hr pressed, brine cooled block,

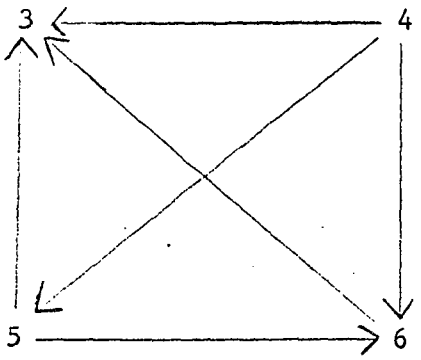
rest of the blocks were criticised for high acid by more than five of the six judges. The body and texture scores did not improve much at 6 months.

Paired comparisons

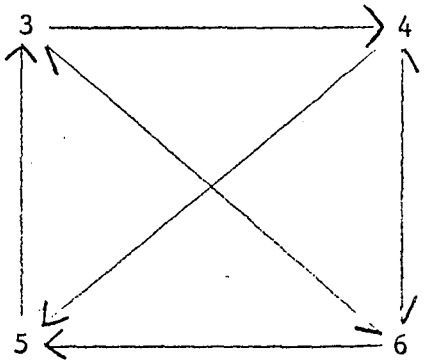
Paired comparisons were made as described in the experimental methods section of this thesis. Samples of cheese, treated in four different ways (4-hr pressed, air- and brine-cooled and 20-hr pressed, air- and brine-cooled), were arranged in pairs in six possible combinations. Each pair was randomized at judging to determine if there were any apparent differences in flavor, body and texture, and color within the pair. The judges also were asked to indicate their preferences within each pair. Most of the judges noted the differences in flavor and body within a pair made of air- and brine-cooled cheeses. These differences ranged from slight to moderate. But differences within a pair made of both air-cooled or brine-cooled cheeses (pressed 4 and 20 hr) ranged from none to slight. Each judge had to indicate his preference within a pair if he could find any difference in flavor. Statistical analyses were made on the basis of preference only within each pair. Four cheeses (no. 3-four-hr pressed, brine-cooled; 4-four-hr pressed, air cooled; 5-twenty-hr pressed, brine-cooled; and 6-twenty-hr pressed, air-cooled) were placed on the four corners of a rectangle as shown in Fig. 9. All cheeses were connected with a straight line running from one to another. In a paired comparison, if one cheese is preferred over the other, a double-winged arrow head was drawn pointing towards the preferred cheese. If the judge could not find any

Fig. 9. Mapping of paired comparisons with a rectangle

Lot A



Lot B



3 - Brine cooled, pressed 4 hr

4 - Air cooled, pressed 4 hr

5 - Brine cooled, pressed 20 hr

6 - Air cooled, pressed 20 hr

difference and indicated no preference, both ends of the arrow received one-winged arrow head pointing each other. In this way, all possible pairs were examined for each judge and preferred cheeses were marked with an arrow head. In lot A shown in Fig. 9, between samples 3 and 4, the former was preferred over the latter, and similar preference was noted between samples 3 and 5. If a judge failed to indicate his preference as shown between the figures 3 and 6 and 4 and 6 in lot B, Fig. 9, both the ends of the straight line connecting the two samples of a pair received one-winged arrow. In this manner, all possible pairs were evaluated for all lots of cheese for each judge. For statistical analysis and evaluation of preference, 1 point was assigned to each cheese for receiving each double-winged arrow head. In lot A of Fig. 9 samples 3, 4, 5, and 6 received the scores of 3, 0, 1, and 2, respectively. Likewise samples 3, 4, 5, and 6 of lot B received the scores 1.5, 1.5, 2, and 1, respectively. From the paired comparison cards of each judge on each lot of cheese, the score for each treatment was calculated and multiplied by 2 to remove fractional points. Scores on treatment for each judge on all cheeses were added and reported in Table 32. From this table it can be seen that brine-cooled cheeses had higher scores in most cases. Some exceptions can be noted with judges B, D, and F. With judges B and F, 4-hr pressed, air-cooled cheeses received slightly higher scores and with judge F 20-hr pressed air-cooled cheeses received a high score compared to brine-cooled cheeses. Statistical analysis of the data over the individual lots showed a

Table 32. Paired comparison scores of judges on individual treatment over 15 lots of cheese

Judge	Total score per judge on all cheeses	Treatment			
		Pressed 4 hr		Pressed 20 hr	
		Brine cooled	Air cooled	Brine cooled	Air cooled
	Judging score				
A	108	56	40	48	36
B	180	43	45	51	41
C	156	50	39	38	29
D	144	38	29	36	41
E	157	38	36	48	35
F	120	24	33	33	30
G	108	33	21	32	22

significant difference at the 1% level only with judge A. The other judges were not very consistent in giving their preferences.

Results of judging are considered consistent with the results of chemical and bacteriological tests. The air-cooled cheeses had higher lactic acid, relatively low pH, high fatty acids, and high degree of proteolysis. The brine-cooled cheeses, on the other hand, containing high sugars, less lactic acid, relatively high pH, less fatty acids, and less proteolysis, were criticised less severely. So, differences in the rate of cooling could attribute serious problems in the cheese flavor as reported by Conochie and Sutherland (13). It is evident from the judging results of high acid and normal acid cheeses made with poor quality milk containing millions of adventitious flora, the fermentative metabolism of the organisms goes faster if the temperature is suitable. In the air-cooled cheeses, the organisms could grow faster to produce unbalanced fermentations leading to serious defects. Although the bacterial counts did not show any appreciable difference between air- and brine-cooled cheeses during the sampling period, the bacterial metabolism was conspicuous in the light of chemical and organoleptic tests. The warm temperature of air-cooled cheeses could cause growth and death of cheese flora at an equal rate and bacterial enzymes could be more active under the conditions prevailing in the warm cheese. But rapid cooling could keep the organisms and their enzymes relatively less active and consequently the counts could remain the same.

The high acid and normal acid cheeses containing large number of adventitious microorganisms gave an uncontrolled growth in air cooled

cheeses. Irvine, Beach, and Burnett (48) indicated that milk quality greatly influenced the flavor quality of cheese. It appears from this observation that bacterial flora in cheese made from poor quality milk can be controlled to some extent if cooled rapidly to curing room temperatures after pressing.

Vedamuthu et al. (83, 84) reported that fruity flavor defects were produced due to high amount of aldehydes and other carbonyl compounds by starter organisms. The same culture from the above investigators was used in experimental fruity cheese. Fatty acid contents of these cheeses were significantly lower than the other experimental cheeses. Only the air-cooled cheeses were more severely criticised for fruity flavor defects than the brine-cooled cheeses. One of the probable reasons for the flavor defect could be due to the formation of esters from alcohol and fatty acids. Mabbitt (55) suggested that residual sugar fermentation by heterofermentative lactobacilli produced acetic acid, ethanol, glycerol, and mannitol. If these are produced in excessive quantity, there could be formation of enough esters with fatty acids. The holding temperature of cheese may again play an important role in these reactions.

No improvement was noticed in bitter cheese due to rapid cooling. It is the belief that insufficient breakdown of proteins and polypeptides to amino acids may lead to the development of bitter flavor in cheese (16, 26, 65). Emmons, McGugan, and Elliott (26) concluded that the manufacturing procedure would not have any significant effect on the prevention of bitter flavor development in Cheddar cheese.

SUMMARY

Nineteen commercial dairy plants were surveyed to determine the post-hoop curd handling procedures. Experiments for this investigation were designed to include what appeared to be extremes in the curd handling procedures. Sixteen lots of cheese were manufactured on a pilot plant scale simulating commercial conditions. A total of 6 vats were made with Iowa manufacturing-grade milk in 5000 lb. vats, of which three had milling acidities above 0.60% and three with normal acidity; i.e. below 0.60% at milling. Three lots were also made in 5000 lb. vats using grade-A milk with a culture producing fruity flavor defect. Rest of the lots were made in 400-lb. vats using grade-A milk. Three lots were made under "ideal" conditions and three were made using cultures producing bitter flavor. The last lot was manufactured with a starter consisting of a commercial lactic culture and a heavy inoculum of coliform organisms isolated from natural Cheddar cheese.

Each lot of cheese was treated in 4 different ways with two pressing times and two cooling rates. After 4-hr pressing, two sets of blocks were removed, wrapped, and sealed. One set of blocks was immersed in a brine tank at 7.5 C for rapid cooling and the other was stacked on the pallet in the curing room. After 20-hr pressing, two more sets of blocks were treated in the same way. Change in temperature in the interior of each block was followed with a continuous recording system.

Samples were collected at milling, after 4-hr and 20-hr pressing, on the 4th, 8th, 12th, 21st, 30th, 60th, and 90th day of curing for

total bacterial count, enterococcus count, and gram-negative bacterial count. Lactose, glucose, galactose, pH, and lactic acid were determined at the same intervals. Measurements of proteolysis and total free fatty acids were made at monthly intervals. Bacteriological and chemical tests were made only on the first 9 lots of cheese made in 5000 lb. capacity vats. All cheeses were judged after 3 and 6 months.

Forty-pound blocks stacked in the curing room at 7.5 C took 150 to 480 hr to cool to 8 C or below, but by immersing blocks in brine at 7.5 C, 25 to 60 hr were required to cool to 7.5 C. Twenty-pound blocks required only 20 to 25 hr in brine and 135 to 185 hr in the curing room to cool to 8 C or below.

No significant differences were found in total bacterial count, enterococcus count, and gram-negative bacterial count due to cooling rates or pressing times. Total bacterial count was found to decline at different rates in different cheeses. The enterococci were present in large numbers in cheese made from manufacturing-grade milk. Initial presence of enterococci in large numbers in raw milk favored their presence in cheese during ripening even though heat treatment of milk greatly reduced their numbers.

Lactose, glucose, and galactose were found to persist beyond 90 days in substantially high concentrations. The brine-cooled and 4-hr pressed cheeses had higher amounts of lactose during all stages of curing than the corresponding air-cooled and 20-hr pressed cheeses. Brine cooled cheeses also had significantly higher amounts of glucose and galactose than the air-cooled cheeses. The air-cooled cheeses

contained significantly higher lactic acid, fatty acids, and had lower pH values at all sampling stages. The cooling rates or pressing times did not significantly affect the amount of proteolysis.

The air-cooled cheeses were more severely criticised for high acid, fruity, and other flavor defects than the brine-cooled cheeses. Brine-cooled cheeses were awarded higher scores based on preference indexes. Rapid cooling of cheese definitely proved to be better in producing uniform flavor, body, texture, and color, especially when poor quality milk was used. Further, it was shown that rapid cooling would control the rate of bacterial metabolism and other changes in the maturing of cheese, and check the undesirable changes during early stages of curing.

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APPENDIX

Questionnaire Used in the Survey of Cheese Plants

1. Do you make Barrel Cheddar _____ or 40-lb. Block Cheddar _____? Other style? _____
2. What is the temperature of the curd at hooping or at the beginning of pressing? _____° F.
3. How long do you press? _____ hrs.
4. What is the curd temperature immediately after pressing? _____° F.
5. What is the average temperature of the room where the curd is pressed? _____° F.
6. At what temperature do you hold the curd after pressing? _____° F. (Does the cheese go immediately into the cooler after pressing or does it receive some other treatment?_____)
7. Do you wrap the blocks immediately after pressing? _____
8. What type of wrapper do you use and how is it sealed? _____

   _____
9. What type of box do you put the wrapped blocks into? _____
10. If you make Barrel cheese, how long do you hold the barrels in the make room before putting them into the cooler? _____
11. What is the average temperature of your cooler? _____
12. Do you use fans? Is there direct air circulation of any type?

    _____
13. How are the blocks stacked? Are they separated in any way? _____

    _____

14. How long does it take the center of the block or barrel to get to:

    70^{o} F. _____

    60^{o} F. _____

    50^{o} F. _____

    40^{o} F. _____

15. How long do you hold the cheese before shipping? _____

16. As you can see, we are primarily concerned with the length of time it takes the curd to cool from hooping temperature to cooler temperature. Do you have reason to believe that your cheese always gets the same treatment from block to block from day to day or could significant variations occur? _____

    _____

    _____

    _____

17. Comments:

This information will be held confidential and will not be revealed by plant name or location. I am interested in variations from one plant to another only and will consider plant processes by plant name as confidential information.

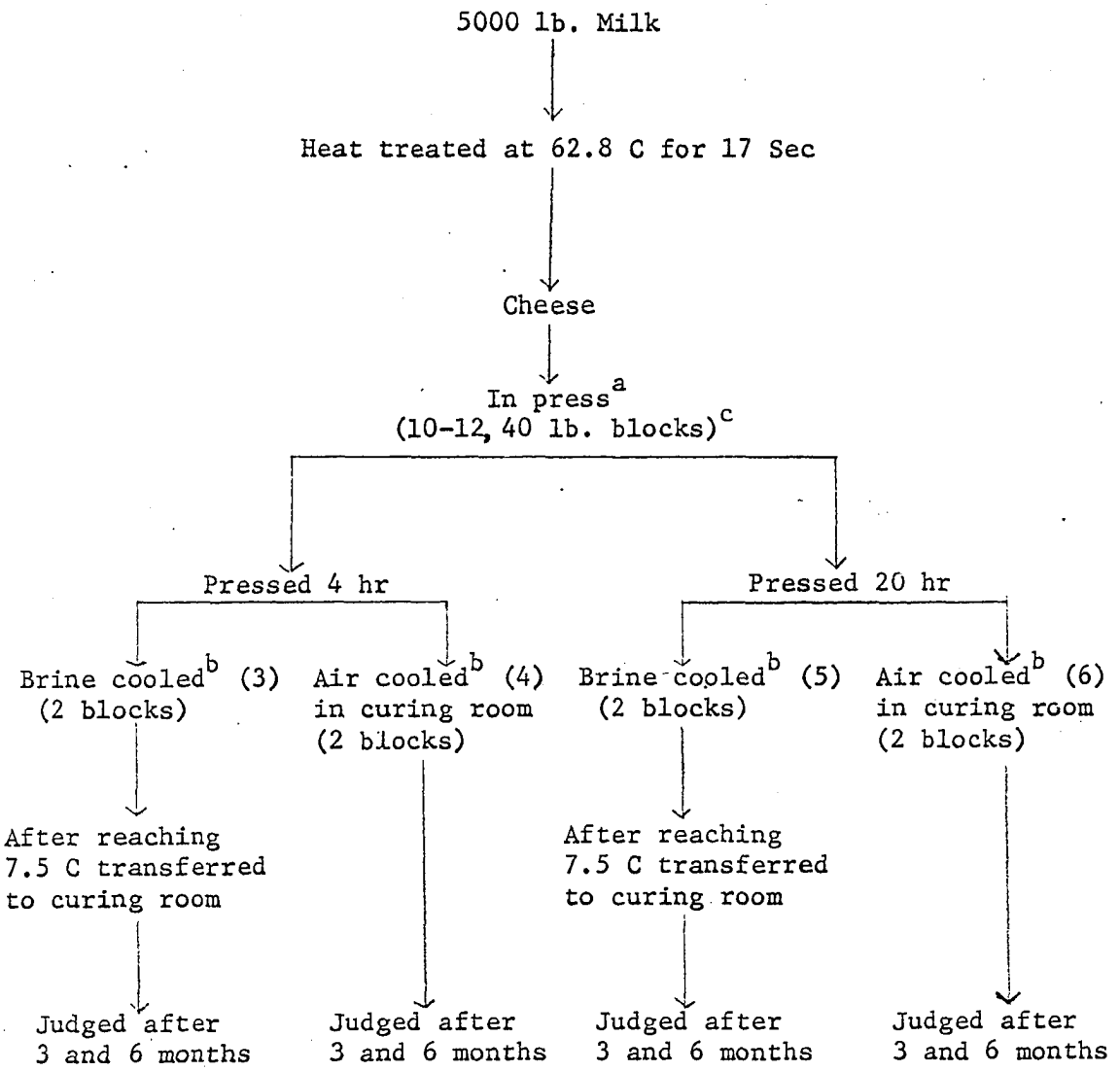
Used for Comparison of Cheddar Cheese Samples

Name _____ Vat No. _____

Date _____

Comparison Pairs	Flavor difference					Body and Texture difference					Color difference					Preference and remarks
	None	Slight	Moderate	Marked		None	Slight	Moderate	Marked		None	Slight	Moderate	Marked		
1.																
2.																
3.																
4.																
5.																
6.																

Fig. 10. Flow diagram of the experimental set up for the manufacture and treatment, testing, and judging of cheese



[a]Recording of cooling rate commenced at this point.

[b]Samples collected periodically for bacterial and chemical tests.

[c]Size: 14" x 11" x 7".

Make Report

Vat. No. F - "Normal acid" Made by R. J. and Miah Date Sept. 6, 1967

MILK: Lbs. 4300 Percent fat

HEAT TREATMENT: Date 9/6 Time 17 Sec. Temp. 62.8 C

STARTER: Culture Ns. 253 Date set 9/5

Acidity 1.03 Amount 21 lbs.

Color 159 ml. Rennet 360 ml.

Salt 15 lbs.

Make Procedure

Operation	Time	Temp.	Acidity
Received in vat	6:30	40	.17
Starter added	8:00	86	.175
Rennet added	8:35	86	.175
Cut	9:05	86	.10
Cook - begin	9:35	86	.11
end	10:05	102	.115
Whey drawn	10:50	102	.12
Curd packed	11:20	--	.155
Curd milled	1:15	--	.50
Hooped	1:50		
Pressed	2:15		

Laboratory report:	Moisture	37.10
	Fat (FDB)	52.46
	Salt	1.83

Make Report

Vat. No. H - "Fruity" Made by R. J. and Miah Date Sept. 18, 1967

MILK: Lbs. 5300 Percent fat 3.4

HEAT TREATMENT: Date 9/18 Time 16 sec. Temp. 62.8 C

STARTER: Culture No. Date set 9/17

Acidity A - 1.0, B - 1.16 Amount 53 (26½ lbs. each)

Color 201 ml. Rennet 444 ml. (2.8 per 1000)

Salt 18½ lbs. (3.5 per 1000)

Make Procedure

Operation	Time	Temp.	Acidity
Received in vat	6:30	40	.17
Starter added	8:15	86	.17
Rennet added	9:15	86	.18
Cut	9:40	86	.11
Cook - begin	10:10	86	.115
end	10:43	102	.13
Whey drawn	11:13	102	.145
Curd packed	11:35	--	.20
Curd milled	1:30	--	.54
Hooped	2:30	--	--
Pressed	2:45	--	--

Laboratory report:	Moisture	36.06
	Fat (FDB)	49.16
	Salt	1.92

Make Report

Vat. No. C - "High acid" Made by R. J. and Miah Date June 1, 1967

MILK: Lbs. 4900 Percent fat 3.5

HEAT TREATMENT: Date 6/1/67 Time 17 Sec. Temp. 62.8 C

STARTER: Culture No. 253 Date set 5/31/67

Acidity 1.11 Amount 49 lb.

Color 147 ml. Rennet 426 ml.

Salt 18 lbs.

Make Procedure

Operation	Time	Temp.	Acidity
Received in vat	7:15	80	.16
Starter added	7:30	86	.165
Rennet added	8:30	86	.17
Cut	8:50	86	.11
Cook - begin	9:20	86	.115
end	9:55	102	.135
Whey drawn	10:45	102	.16
Curd packed	11:15	102	.26
Curd milled	12:40	--	.70
Hooped	1:30	--	--
Pressed	1:45	--	

Laboratory report:	Moisture	34.36
	Fat (FDB)	47.98
	Salt	1.95

Make Report

Vat. No. M - "ideal" Made by R. J. and Miah Date Oct. 3, 1967

MILK: Lbs. 400 Percent fat

HEAT TREATMENT: Date 10/3 Time 15 sec. Temp. 62.8 C

STARTER: Culture No. 253 Date set 10/2

Acidity 1.08 Amount 4 lbs.

Color 15 ml. Rennet 33 ml.

Salt 1.4 lbs.

Make Procedure

Operation	Time	Temp.	Acidity
Received in vat	8:10	52	.15
Starter added	8:45	86	.16
Rennet added	9:45	86	.175
Cut	10:07	86	.105
Cook - begin	10:37	86	.11
- end	11:07	86	.125
Whey drawn	11:50	102	.14
Curd packed	12:15	--	.24
Curd milled	2:00	--	.53
Hooped	2:15	--	--
Pressed	2:30	--	--

Laboratory report

Not run

Make Report

Vat. No. Q - "bitter" Made by R. Carr and R. J. Date Oct. 19, 1967

MILK: Lbs. 390 Percent fat 3.4

HEAT TREATMENT: Date 10/18/67 Time 17 Sec. Temp. 62.8 C

STARTER: Culture No. F8DBE Date set 10/18/67

Acidity .80 Amount 4 lbs.

Color 15 ml. Rennet 33 ml.

Salt 1.4 lbs.

Make Procedure

Operation	Time	Temp.	Acidity
Received in vat	8:30	48	.150
Starter added	9:00	86	.160
Rennet added	10:00	86	.170
Cut	10:30	86	.115
Cook - begin	11:00	86	.125
end	11:30	102	.13
Whey drawn	12:05	102	.14
Curd packed		--	
Curd milled	2:50	--	.54
Hooped			
Pressed			

Laboratory report

Not run