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PROCEEDINGS

PRESENTED BY MARSCHALL DAIRY PRODUCTS
ELKHART, INDIANA

IN CONJUNCTION WITH SEMINAR EXHIBITORS
AND NON-EXHIBITING HOSTS

SEPTEMBER 13 & 14, 1989

THE FORUM ■ DANE COUNTY EXPOSITION CENTER
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PROCEEDINGS
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26TH ANNUAL MARSCHALL
ITALIAN CHEESE SEMINAR

September 13 & 14, 1989

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Elkhart, Indiana

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CHANGES IN DAIRY PRODUCTION AND CHEESEMAKING . . . ARE YOU READY FOR THE FUTURE?

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Abstract

Fewer and bigger dairy farms, new milk production technology and changing farm organizations are just a few of the challenges signaling an era of dramatic change in the basic milk production industry.

Add ever-more efficient cheese making techniques, processor mergers and buyouts, legislation, changing consumer wants and public activism and emotionalism journalism.

The result . . . new and accelerated challenges to the cheesemaker, the cheese marketer and the entire Italian Cheese Industry. Are you ready?

Question?

Can you remember when "going out to eat" meant hamburgers or steak? When pizza was not a main part of the menu for teenage parties, after football game get-togethers or for supper on weekends?

If you do, don't tell anyone -- they may want to lead you back to the old folks home. Try to tell anyone under thirty (even forty) years of age that there was a time when pizza, lasagna and a whole host of other Italian foods containing Italian cheese were not readily available in restaurants, supermarkets and delis. Better yet, don't even try. They'll never believe you.

Ask a young friend about that "new" string cheese. You'll get a blank look, not because they don't know about and eat string cheese, but because of the word "new." (Note -- string cheese, has become a popular food item in just the past ten years and is truly a product of the 80's).

Your hosts -- Marschall Dairy Products -- sent out the 1988 Italian Cheese production report. The headline -- "37th Consecutive Record Year" tells it well. The copy cites 1988 production of 1,937,118,000 pounds of Italian Cheese, 8% above a year ago. Without doubt, production will exceed the two billion pound figure this year.

In Wisconsin -- still fondly the "dairy state" -- where 34.3% of the Italian Cheese is made, production has soared from 153.7 million pounds to 664.1 million pounds in the past 20 years. Without a doubt Wisconsin will more than keep pace with the nation's hunger for Italian Cheese in years to come.

So much for the past, but we are here to look at the future.

Surely the success of ever-increasing production in Italian Cheese for 37 straight years makes for a rosy future. And -- no one sees anything but increasing consumer demand in years to come.

Yet -- consumer demand is only one measure of success for your industry and demand in itself does not insure success for individual processors, marketers or suppliers.

Fact of the matter, there are some hidden booby traps, marked minefields and widely proclaimed obstacle courses offering challenges to the cheese industry as a whole and of course, to the Italian Cheese segment.

Let's talk about these challenges as seen through my eyes but formulated as a result of close relationships with most segments of the cheese industry from farm to processor, to marketer to consumer.

The Challenge of The Changing Farm Scene

Logically we start on the farm where most all the Italian cheese in this land has its beginnings. In Wisconsin -- still the biggest state in terms of milk production - - milk is produced on 36,000 farms where some 1.8 million cows reside.

Over the years ever-fewer farmers annually produce more milk from each cow than the year before. Why? Advances in production management in terms of labor, feed, equipment, herd health and money have brought milk production and efficiency to record levels. And it continues.

As a result, milk supplies have been more than adequate -- actually in surplus for long periods -- for the needs of both processor and consumer.

Not all U.S. dairy farmers are happy with supply/demand pricing of their milk and have long discussed but never united behind a production control program. Be assured that supply/management does have a following among some farmers, in certain farm organizations and with some influential legislators.

Currently this subject is one of non-discussion, but it will arise when milk production increases beyond needs and prices to the farmer drop. Should that happen at the same time the next Farm Bill is being written, milk production controls will be discussed. Bet on it!

The changing farm scene may make a control program potentially more probable because of:

1. fewer and bigger farm units
2. increasing control of farm milk supplies by dairy cooperatives
3. rapid changes in farm milk pricing
4. governmental desire to have a farm program

As the milk supply is controlled by fewer and bigger producers and cooperatives, chances for unity appear to be better. Congress respects direction from unified groups. If a supply/management program was initiated, the cheese industry would face the challenge of limited milk supply at potentially higher prices.

You could be in a whole new ball game!

Challenge of Consumer Involvement

On the other end of the Italian Cheese chain are consumers, the folks who can't get enough of your products. This happy group -- it's all of us -- could be one of the major challenges to your industry.

For many years the nation's dairy farmers have depended on University, government and private monies to research new ways to produce milk. The results of individual research worked or didn't but long-term we benefitted in terms of cheap food, long life and full stomachs. Increased production through research was the pride and joy of agriculture and America.

Times have changed! You know the story but briefly in review . . .

Bovine growth hormone (BGH) or bovine somatotropin (BST) -- same product different names -- is in the test stage by four companies. The University of Wisconsin planned on-farm testing to determine health and dollar merit on cows in private herds. A small group of activists objected to the tests because the milk from the test herds was to enter normal marketing channels.

Even though the FDA approved the disposition of milk the resulting hue and cry aroused consumers to question milk purity and safety. Every dairy farmer, processor and marketer has had to (or will) face the issue. An issue of facts became an issue of emotions . . . and . . . right or wrong the consumer is the key on which all others must decide.

BGH/BST is the subject of the moment, but it will not be the last that the industry will face. Must cows be raised without benefit of modern veterinary medicine, feed void of modern medicinal or technology-based additives and milked by hand? Silly? Of course. But, there are so-called "environmental" groups who advocate some unusual approaches to raising cattle.

How might this impact you? Where it hurts, in the supply and price of raw materials -- milk, in your methods of producing your product and in your relations with the consumer and producer.

By the way -- the New York City consumer may see dairy farms as an idyllic place where big-eyed, floppy-eared, contented animals are tended by happy folks with bib-overalls, a straw in their teeth and a pitch fork in hand. I am rather sure than many consumers actually know very little or want to know much about cows or farms or milk.

But -- they are learning and are about to learn more -- a lot more. And -- that might not be so good, at least in early stages, for the entire dairy industry.

Challenges of Success Bringing Competition

The very success of Italian Cheese has changed that smallish, family oriented and rather specialized industry of a few years ago into one that's now automated, big, finance driven and very competitive.

Processors of all sizes are concerned about their future.

Small processors (independents and cooperatives) see big cooperatives getting bigger, foreign companies expanding their U.S. holdings and new facilities and equipment everywhere. And they wonder.

Mid-size processors look at the labor efficiencies, automation and marketing strength of the bigger processors and wonder if they should get bigger or if they should have stayed small.

The bigger processors have the things that their smaller competitors might see as the ultimate in equipment and facilities but find that value-added marketing, product specialization, fast direction-changes and management doesn't come easy.

All find that the competition in terms of processing and marketing has and will continue to pose challenges in:

- a) Available milk supplies . . . Cooperatives have for many years controlled the milk supply at the farm level; mergers may intensify the situation.
- b) Marketing of commodity cheese . . . Bigger operations can have price advantages.
- c) Specialized markets . . . Large, commodity-oriented operations are entering the specialty areas through acquisitions.

It should be noted that the challenges presented by cooperative mergers and acquisitions of smaller processors by foreign or multinationals resulting in mega-processors/marketers does not take away opportunity for the smaller operation to specialize and innovate for success.

The New Product, New Consumer Challenge

What will Mr. and Mrs. and Ms. America want to eat? What will they buy and what will they pay? Questions that everyone wants to answer.

For sure consumers want pizza and the now-traditional foods using Italian Cheese, the question is; what other foods or specialties can expand the market for your products and services? Some thoughts . . .

--Away from home dining . . . McDonalds is testing full-size pizza to get the family into their establishments. A few years ago they also tested mini-pizza in Madison and the product failed. However, I personally see someone producing pizza slices or mini-pies in fast food outlets.

Why? Because in my case I wanted something that didn't drip on my tie or shirt as I hurriedly ate while driving. Pizza fits that bill, unfortunately the test product left much to be desired in taste. But, someone will do it. The away-from-home food industry is ever-changing and the future is unlimited.

--Refrigerated dishes . . . This new class of cold, but not frozen product is just hitting the market and lots of folks are getting into it.

--Healthy foods . . . Do you know anyone who just eats without selecting for calories, sodium, cholesterol, carbohydrates, fiber, energy, vitamins or whatever? Enough said . . .

The Challenge of Business

Ask a hundred Italian Cheese makers, marketers or eaters to list major challenges facing their corner of the industry and you may get two hundred suggestions. Here are some that will be fairly unanimous . . .

--Waste disposal . . . what goes in comes out and government and the public are becoming more concerned with disposal of water, chemicals, organic matter and solids. Legislators usually act fast in crisis situations and waste is fast becoming an issue.

--Speeding technology . . . offers gains and problems. It can be expensive and not everyone wants to be , or can be, "state of the art" all the time. We are impacted everyday in many ways. Handling it is the question.

--Crisis management . . . as consumers become more involved and technology advances, chances of a crisis may increase and the scope becomes wider. The question is . . . what do you do when the reporters call? The answer -- be ready.

--Staying ahead of the market . . . what is your niche? There are few absolute answers here, rather it is a combination of technology, skill, intuition and luck.

We have been considering "challenges" to your industry . . .note a challenge is often just another word for opportunity. And . . . YOU DO HAVE OPPORTUNITY.

But -- opportunity only comes to those who seek it.

And the rewards can be great.

SUCCESS in your search.

MILK FROM bST-TREATED COWS: COMPOSITION AND MANUFACTURING PROPERTIES

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ABSTRACT

Treatment of lactating dairy cows with bovine somatotropin has been extensively studied to determine the impact on milk composition and the safety of milk for human consumption. The FDA has concluded that the milk produced by cows treated with bST is safe for human consumption. Extensive evaluation of milk composition and other dairy product manufacturing characteristics (i.e. flavor, culture growth, etc.) have found that characteristics of milk from bST treated cows are within the normal range of biological variation in these milk characteristics for milk produced by untreated cows.

INTRODUCTION

The dairy product manufacturing sector of the dairy industry provides a vital link in the chain starting with milk production at the farm and ending with a variety of wholesome dairy foods for the consumer.

New technology has been and continues to be developed to improve the efficiency of food production. The cost effective manufacture of large quantities of bovine somatotropin (bST) by fermentation technology will provide the dairy industry with a technology to be used at the farm that has the potential to increase the efficiency of milk production. The dairy industry has a long history of implementation of new technologies to improve milk production efficiency (e.g. ration balancing, feed supplements, genetic selection, improved milking equipment, etc.).

Why would a dairy farmer want to use bST? bST improves the efficiency of conversion of feed to milk. A portion of the energy and other nutrients consumed by the dairy cows is used for body maintenance. In business terms, this is the fixed operating energy cost of the dairy cow whether or not the cow is producing milk. When a lactating dairy cow is treated with bST, the cow voluntarily increases feed consumption. Since the energy and nutrient requirements for body maintenance are not changed, the additional nutrient

intake is available for increased milk synthesis. This increases the amount of milk produced per unit of feed consumed.

How much will milk production increase when a dairy farmer treats his cows with bST? The increase in milk production will vary from farm to farm, but an estimate of the average response is in the range of 10 to 15%. Some farms will obtain higher response, while other farms will obtain lower or no response.

Why would there be no increase in milk production and milk production efficiency on some farms? If a herd of dairy cows is not provided with an adequate quantity and quality (properly balanced ration) of feed to meet the nutrient requirements for milk production, then that herd will not be able to produce a quantity of milk that reflects their genetic potential for milk production. Thus when bST is used, a dairy herd with inadequate quantity or quality of feed will have a low or no milk production response to bST. On a farm with a low average milk production per cow, that particular farm could probably benefit more (i.e. improve milk production) by properly balancing the ration and using other available technologies/services (e.g. DHI, AI, etc.) prior to considering implementation of bST technology.

IMPACT OF bST ON SAFETY OF MILK FOR HUMAN CONSUMPTION

The safety of milk from bST-treated cows has been evaluated by the Food and Drug Administration and is not an issue. The FDA has determined that the milk and meat from bST-treated cow is safe. Why would the FDA decide that milk from bST treated cows is safe for human consumption?

To address this question, I find it useful to review some basic information that identifies what bST is and is not. Bovine somatotropin is a hormone that is produced by the pituitary gland of the dairy cow. Humans produce a similar hormone called human somatotropin (hST) in their pituitary glands. Is there a difference between bovine somatotropin and bovine growth hormone? No, they are the same thing. This hormone functions during both the active growth stage of the animal and during the stage of life when an animal is not actively growing. Therefore, the name growth hormone only relates the function of this hormone during the active growth phase of an animal. In later life the same hormone functions in concert with other hormones in the body to regulate metabolism and nutrient utilization. Therefore, the name somatotropin applies more correctly to the overall function of the hormone throughout the life of an animal.

What is a hormone? A hormone is a substance produced by an organ in one part of the body and then it is transmitted through the circulatory system to start, stop, or change the rate of a metabolic activity in another part of the body. Hormones are mother nature's form of a FAX message system within the body. Are there different types of hormones? Yes, there are many different types of hormones produced in the body and there are two very different types of chemical structures of hormones; protein hormones and steroid hormones.

Insulin (used for treatment of diabetes) is an example of a protein hormone and estrogen (used in oral contraceptives) is an example of a steroid hormone. From a chemical perspective, protein hormones are very large structures and steroid hormones are relatively small structures. Bovine somatotropin is a protein, not a steroid.

The difference in chemical size and structure of proteins and steroids is very important. Estrogen (a steroid) can be taken orally and absorbed in the stomach because of its compact chemical structure. Insulin (a protein) however is not effective when taken orally because it is large and is digested in the stomach. Thus to be effective, insulin has to be injected. Bovine somatotropin also has to be injected into dairy cattle to be active and cannot be given orally.

Thus, several factors are important when considering the food safety aspects of bST. First, because bST is a protein, it cannot be absorbed in the intact form when consumed as a component of milk. It is digested in the human stomach just like any other protein present in the food that we eat. Second, bST has no biological activity in humans. The chemical structure of somatotropin is different from one species of animal to another. Even if bST is injected into a human, it has no activity. Finally, bST is normally found in very low concentrations in bovine milk. When dairy cows are treated with bST to improve efficiency of milk production, there is no measurable increase in the bST content of milk. For these reasons, milk from bST-treated cows is safe for human consumption.

OTHER COMMON QUESTIONS ABOUT bST

Is the level of bST increased in milk when cows are treated with bST? No. When cows are treated at the recommended levels of bST (level that will maximize financial return to the dairy farmer), there is no change in the bST content of milk. bST is normally present in milk at an average level of 1 ng/ml (1 part in 1 billion parts of milk). One study designed to determine the impact of high doses of bST on the dairy cow, reported a bST content of milk of 3ng/ml. However, the dose used in this study was 214 bST/day. Use of bST at this dose level would not be economical for the dairy farmer (i.e. above the optimum dose level the production response does not continue to increase at a favorable rate). The normal dose of bST for dairy cows will probably be in the range of 25 to 35 mg/day.

Is the bST that would be marketed identical to the natural bST produced by the dairy cow? No, however the one commercial product (sometribove) that I am most familiar with differs by just 1 out of the 191 amino acids found in the bST structure produced by the dairy cow. Thus, 190 out of 191 amino acids in the sequence of this commercial product are identical to that produced by the cow. There are at least 4 different commonly found structures of bST that are produced by the dairy cow. Two of the structures contain 190 amino acids and two contain 191 amino acids. Much like Holstein cows vary from cow to cow in their black and white markings and the proportion of black and white, a cow may also produce the four different types of bST structures in different proportions. One commercial product (sometribove) contains 191 amino acids

in its structure, 190 out of 191 amino acids in the chain are identical to the bST produced by the cow. The last amino acid on the N-terminal end of the amino acid chain in the commercial product is methionine, instead of the amino acid alanine.

All proteins (including bST and milk proteins) are chains of amino acids. There are 20 different amino acids. All proteins are made from different combinations of these amino acids. The reason that one protein is different from another protein is due to the fact that each protein contains different proportions of each of the 20 amino acids and has them organized in a different linear sequence. Thus, bST and milk casein (and all other proteins) are made of the same amino acid building blocks.

Is this difference in structure of a commercially produced bST of any practical significance? No. It is correct that the medical studies of the impact of injection of bST into humans that were conducted in the 1950's were done using "natural" bST, not the commercially produced bST that is available today. It is highly unlikely that the difference of one amino acid in the sequence at the N-terminal end would change the activity of bST when injected into humans, because it is well documented that approximately 30% of the amino acid sequence is different (i.e. about 50 amino acids) when bST and hST are compared directly. To bring this issue back into proper perspective, we need to remember that we eat milk not inject it into our bodies. The bST present in milk from treated and untreated cows will be digested just like any other protein in the diet. The additional methionine added to the structure of the commercially produced bST is of no practical significance because methionine is a normal part of our diet. Methionine is normally present as part of the structure of milk protein. Normal bovine milk protein is approximately 2.4% by weight methionine.

IMPACT OF bST ON MILK COMPOSITION

It is well established that changes in raw milk composition result in changes in the manufacturing properties of milk. Milk composition varies normally due to diet, breed, genetics, mastitis, stage of lactation, environmental temperatures, and many other factors. The detailed chemical composition of milk and specific composition of the major milk constituents is very important to dairy product manufacturers and consumers. For example, large changes in fatty acid composition of milk fat will influence flavor and texture of high fat dairy products (i.e. butter, ice cream, and high fat cheeses). Changes in the milk protein fraction (i.e. types of caseins or whey proteins) can influence moisture, texture, flavor and yield of various cheeses. Changes in milk mineral balance can influence heat stability of milk proteins and protein coagulation in cheese making. Therefore, changes in detailed chemical composition of milk components can have as much or more impact on dairy product manufacturing and product characteristics than changes in total percentage of individual milk solids components. In addition, changes in milk composition could influence nutrient content of milk. Thus, it is important to determine if bST technology will change milk composition.

GENERAL MILK COMPOSITION: PERCENT FAT, PROTEIN, LACTOSE, TOTAL SOLIDS, AND SOLIDS-NOT-FAT

The effect of bST on general milk composition is dependent upon the nutritional status of the cows both before and during bST treatment (20). In the early stages of bST treatment and in trials where bST is administered for short intervals (i.e. less than 28 days), increases in percent milk fat and decreases in percent milk protein can occur (7,9,11,17,18,21). These changes in milk composition occur only when the milk yield increases cause changes in energy and protein balance in the cow during the initial stages of bST treatment, as body fat and protein stores are mobilized to meet the increased nutrient demands. With prolonged bST administration (e.g. full lactation) cows voluntarily adjust their feed intake to meet their increased nutrient requirements, and nutrient balance is restored. These changes in energy balance are similar to, but smaller than, the changes in energy balance that normally occur at the onset of lactation. In general, the percentage of milk fat, protein and lactose was not different for bST and untreated cows when bST was administered over a full lactation (3,19,23). In a full lactation study (2,4) the concentration of fat, lactose, ash, solids-not-fat, total solids and calcium content of milk was not affected by bST. In the same study (2,4), milk protein was increased slightly. On a practical level, the use of proper feed management practices combined with the use of bST should result in normal fat, protein and lactose content of milk. Milk total solids and solids-not-fat were not influenced by bST administration (2,19,23).

MILK CASEIN, WHEY PROTEIN AND NONPROTEIN NITROGEN

Increases in percent nonprotein nitrogen and whey protein, and decreases in casein as a percent of true protein have been observed with long term bST administration, although the differences were not always statistically significant (1,2,12). It is unclear if the production of whey proteins actually increases relative to casein synthesis, or whether the changes observed in the whey and casein fractions are a result of a slight increase in proteolysis of casein (13). A separate analysis of individual milk proteins normally present in milk (α _S-casein, β -casein, κ -casein, α -lactalbumin, and β -lactoglobulin) found that the relative proportions of these proteins were not different in milks produced by cows treated with bST and those not treated (14). With milk composition data from that study, it was predicted that average cheese yields would be unaffected by bST.

More work needs to be done to determine if previously observed changes in casein as a percent of protein (1,12) are directly related to bST administration. These changes may reflect normally occurring differences in protein composition or level of native milk protease activity that exists between milk from low producing and high producing cows without bST treatment. In addition, the relative impact of level of milk production and/or bST treatment needs to be put in perspective with the magnitude of impact of other factors (such as mastitis and age of cow) which can have a much larger influence on

milk protease activity and milk casein as a percent of protein than any influences that have been reported in milk produced by bST-treated cows.

MILK ASH AND MAJOR MINERALS

Hard et. al. (10) summarized the data from long term studies conducted at four different locations involving a total of 364 cows and concluded that the percentage of total ash, phosphorus and calcium in milk was not altered by bST administration. In one study where bST was administered for 10 days, percent milk sodium, iron, copper and manganese did not change, while percent magnesium increased and zinc concentrations varied with bST dosage (8).

FATTY ACID COMPOSITION AND THERMAL PROPERTIES OF MILK FAT

The fatty acid composition of milk fat from cows treated with bST would be expected to reflect the energy status of the cows. Dairy cows are normally in negative energy balance during early lactation, and the increased energy needs of the cow caused by the onset of lactation are met initially by the mobilization of body fat stores. As a result, the relative percentage of long chain fatty acids is higher and the relative percentage of the short and medium chain fatty acids is lower in milk fat produced in early lactation compared to milk fat produced in late lactation in milk fat (14). Similarly, in studies where bST treatment was of short duration (these studies essentially reflect the effects of the early stages of bST treatment), the increased energy demands of the cow were met initially by the mobilization of body fat stores. This caused the relative percentages of long chain fatty acids to increase while the relative percentages of short and medium chain fatty acids decreased (5,8). These results are consistent with the idea that, with prolonged bST administration, cows adjust their feed (energy) intake to meet increased production demands, and fatty acid composition normalizes once the energy balance in the cow is restored. Normal changes in milk fatty acid composition with stage of lactation are relatively large compared to any small short term changes in milk fatty acid composition that occur at the beginning of bST treatment of dairy cows.

Similar changes in fatty acid composition of milk fat occur in both bST treated and untreated cows with stage of lactation (14). As expected, the substantial change in fatty acid composition due to stage of lactation had an influence on the relative proportions of high and low melting fat, but had no influence on the initial and final melting temperature of the milk fat (15). Bovine somatotropin had no influence on the melting properties of milk fat (15).

In one study when bST was administered for a full lactation, the relative percentages of the short, medium and long chain fatty acids were not significantly affected (14). In another full lactation study (1), a small change in fatty acid composition was reported; short chain 11.6% & 10.5%, medium chain 58.5% & 56.0%, and long chain 26.9% & 30.4%, control & bST respectively. However, when one places these changes in the context of their nutritional significance, there was a slight decrease in saturated fat level, an increase in

the level of monounsaturated fat, and no change in polyunsaturated fat. Thus, the small change in fatty acid composition reported in this study would be considered to be a move in a favorable direction from a diet and health perspective, however the magnitude of the observed shift would not be large enough to be of any practical significance in the total diet.

MILK FREE FATTY ACIDS AND MILK FLAVOR

No differences in the level of free fatty acids in fresh milk from either cows receiving bST or untreated cows have been observed (13). Similarly, no effect of bST on the relative activity of native milk lipase, as indicated by the release of free fatty acids from a susceptible milk fat substrate, has been noted (13). These conclusions are supported by results from milk flavor analysis at one and eight days after pasteurization where panelists were unable to tell the difference between milk from bST treated and untreated cows (16). No flavor differences or off-flavors were detected between milk from bST treated and untreated cows, nor did the milk differ in susceptibility to oxidation after the addition of copper (1).

CHOLESTEROL CONTENT OF MILK

Cholesterol content of food is of concern to individuals that desire to control or restrict their dietary intake of cholesterol. It is recommended by nutrition professionals that daily dietary intake of cholesterol be 300 mg or less. Average cholesterol content of milk produced by untreated and bST treated cows was .388 and .405 g/100g of milk fat, respectively (15). Cholesterol content of milk fat increased similarly with stage of lactation for both bST treated and control cows (15). The average cholesterol content of a one cup serving of whole (3.5% fat) milk from control and bST treated cows was 33.1 mg and 34.6 mg, respectively. Food composition tables (USDA Handbook No. 8-1) estimate the cholesterol content of a one cup serving of whole milk as 33 to 35 mg.

STARTER CULTURE GROWTH

Normal growth of lactic acid producing starter culture bacteria in milk is critical for the manufacture of fermented milks and cheeses. Trace amounts of inhibitory substances can prevent the growth of these bacteria and cause problems in cheese manufacture. The growth of five commercial lactic acid producing starter cultures used for Cheddar cheese manufacturing was evaluated in milk from control and bST treated cows. Inoculation percentages were as recommended by the culture supplier. Each culture strain gave similar amounts and rates of change in milk pH and titratable acidity (30°C for 6 hours) in milk from bST treated and untreated cows (16). Treatment of lactating dairy cows with bST has no influence on the growth of lactic acid producing starter culture bacteria commonly used in the dairy industry.

CONCLUSIONS ABOUT THE IMPACT OF bST ON MILK COMPOSITION

Nutritional status of cows influences milk composition, but bST appears to have no direct influence on milk composition. Both cows receiving bST treatments and untreated cows need to be provided with adequate dietary energy and protein to maintain milk composition. The requirements of bST supplemented cows appear to be similar to those of high producing untreated cows, in that both require appropriate nutritional management in order to obtain maximal production benefits (6,22,24). Published reports to date on the effect of bST on milk composition indicate that there would be no significant impact on the dairy product manufacturing characteristics of milk.

OTHER COMMON QUESTIONS AND ISSUES RAISED ABOUT bST

What is IGF-I and is it related to bST? IGF-I is insulin-like growth factor I. When a dairy cow is treated with bST, the bST does not act directly on the mammary tissue. There are receptors in the liver that sense the increased concentration of bST in the blood. The liver responds by releasing more IGF-I into the blood. The mammary tissue does have receptors that sense changes in the level of IGF-I in the blood. The exact mechanism by which IGF-I stimulates increased synthesis of milk fat, protein, and lactose is not understood.

Does the level of IGF-I change in milk when cows are treated with bST? Yes. Milk levels have been reported to increase up to two fold (untreated cows 3.22, 2.62, and 3.78 ng/ml and bST treated cows 3.80, 5.39, and 4.98 ng/ml, respectively). IGF-I concentration in bovine milk from untreated cows normally starts out high (up to 30.5 ng/ml) at the beginning of lactation and then decreases. Unlike bST and hST which are different in structure, the structure of IGF-I in bovine and human milk are the same. It is important to note that levels of IGF-I normally found in human breast milk are reported to be higher (7 to 30 ng/ml) than the level of IGF-I in bovine milk. Thus, the modest rise in IGF-I concentration in milk produced by bST-treated cows is well within the normal range of concentrations found currently in bovine and human milks.

Will bST put small dairy farmers out of business? In my opinion, this technology will be equally attractive and adoptable by both a well managed small or large farm. In 1955 there were 2.76 million dairy farms and 21 million dairy cows, in 1975 there were 444 thousand dairy farms and 11.1 million dairy cows, and in 1985 there were 272 thousand dairy farms and 11 million dairy cows. During that same time period the average milk production per cow has increased from 5,842 lbs/cow/year to 12,994 lbs/cow/year. These changes in number and productivity of dairy farms have resulted from changes in technology at the farm. The two technologies that probably put more small dairy farmers out of business during the history of our dairy industry are the milking machine and the refrigerated bulk milk tank. Both of these technologies and many other new technologies offered to the dairy farmer have a high capital cost associated with them. New technologies with relatively high capital cost barriers and slower rates of pay back are usually a problem for the small farmer. Adoption of bST technology does not have a high capital cost barrier. If a dairy farmer buys and successfully uses bST, the pay back is very rapid. If the farmer (small or large) uses a bST product, the response (or lack of response) to bST will be evident

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very rapidly (a few days). If it does not work, the farmer can stop using it immediately and minimize any short term loss.

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THE MANUFACTURING PROCEDURE FOR FONTINA AND BEL PAESE TYPE CHEESES FROM NON-U.F. AND U.F. MILK

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Justification

Italian Cheese consumption has increased during the last several years. The bulk of this growth has been in Mozzarella cheese, but there are other cheeses which also have potential. Many of the procedures and practices, such as the use of nitrates, multiple mixed cultures and shelf curing of cheese, are used in Europe, but are not common in the United States or, in some cases, are illegal.

Bel Paese Type

Bel Paese is a soft to semi-soft cheese which was first made in Italy about 1920. It is important to note that the name, Bel Paese, is trademarked necessitating the use of other names for cheese resembling this variety. Similar types of cheese, Konigkase, Bella Alpina, Vittoria, Fleur des Alps and Butter, are made in various parts of Europe. Bel Paese type cheeses have a moisture content of 44-47%, fat content of 28-29%, and a salt content of 2.3-2.5%.

Fontina Type

Fontina is a cooked-curd, whole milk, semi-soft to hard, slightly yellow cheese with a delicate, nutty flavor and a pleasing aroma. It is made from ewe's milk in the Aosta Valley in Piedmont, Italy and from cow's milk in the United States and Sweden. Fontina type cheeses require the use of special cultures, such as mixed mesophilic lactic cultures for flavor and propionic bacteria for proper eye development. Generally, special equipment for the washing and pressing of the curd is needed. This cheese traditionally has sparse eye development and is similar to Gruyere in appearance. Manufacturing methods are similar to Danbo. Fontina is a 6 million pound per year market and currently growing at 12%. Fontina type cheeses have 38-42% moisture and a fat content of not less than 50 FDB.

The second phase of this project will be the production of these cheeses from milk concentrated via ultrafiltration.

There was a diversity in the examples of cheese that were purchased as references for this study. This was evident in the Bel Paese type samples which had a wide range of flavor despite the fact they had one country of origin and one supplier. Several countries produce Fontina type cheese. Cheeses from each of these countries has slightly different flavor, texture and physical characteristics. Therefore, a wide range of flavors was encountered. The

differences in flavor may have been compounded by mistreatment during shipping and retailing. It was evident from all the samples that were purchased that the people marketing these cheeses, especially at the retail level, need to be educated about how to handle and sell them.

One of the main objectives of this study was to develop manufacturing procedures which would allow these cheeses to be produced alongside existing cheese production. Reduction of the use of specialized equipment and the ability to produce additional specialty varieties or variations of these cheeses was also a concern. Therefore, all the equipment used was manufactured in the United States and modifications needed were made by the Cal Poly staff.

Shelf curing, which involves high labor and/or capital cost, is used extensively in Europe. The cheese is frequently turned and washed, which results in the formation of a rind. This practice does not appear to be compatible to cheesemaking practices in the United States. The identification of the proper packaging material for this cheese was another concern. It is important that these cheeses have a limited exchange of gases during aging. The Cryovac barrier bag allows this exchange. The multilayer laminated bags were not as successful.

One difficulty in the production of the cheeses in this study has been identifying culture combinations and ripening practices that will be acceptable. Multiple mixed cultures from Europe were most successful. Cheeses made with American lactic cultures did not have proper flavor development and had a tendency to weep, or whey-off after packaging.

Bel Paese Type

Three potential methods for the production of Bel Paese type cheese were identified. The selection of the starter cultures was one of the most critical steps. The cheese is formed without pressing and is turned in a hoop, which is similar to brick cheese production in the United States of America. Salt is applied by brining in a saturated salt solution. The brine treatment is critical for the development of proper body and texture. The production of a Bel Paese type cheese from retentate has been initiated and a possible processing procedure outlined.

The procedures evaluated were Richelieu-St. Hyacinthe, the Wilster method and one Italian procedure. A modified procedure was developed, which gave better control of milk ripening and reduced the amount of excess whey in the final package.

Cal Poly Bel Paese type Cheese Procedure

A flow chart for the processing of this cheese is found in Appendix 1.

One percent (1%) starter containing a mixture of Streptococcus thermophilus and Streptococcus lactis or Streptococcus cremoris was used. These cultures were grown separately, then mixed upon addition to the vat. The starter cultures were added to pasteurized milk which had been warmed to 104°F (40°C).

Changes in the percentages of S. thermophilus culture to lactic culture produced a flavor variation ranging from that of a Feta cheese when 100% S. thermophilus culture was used, to that of a mild Bel Paese type when a 50:50 mixture was added.

Rennet was added at a rate of 90 ml per 450 kg milk, approximately 45 minutes after culture addition or when the pH of the ripened milk reached 6.54. The identification of the pH at which rennet was added was very critical. If the addition of rennet was based on elapsed time from starter addition, the pH of the milk at this point varied. If the pH at rennet addition was not controlled to pH 6.54, differences in the firmness of the coagulum, final pH of the cheese and body of the cheese resulted. These differences often produced unacceptable cheese. The pH of the milk before ripening and at the time of rennet addition was closely monitored to avoid defects. Adjustments, such as the amount of starter added or amount of ripening time allowed, were made to control the pH of the milk at this crucial point. When the pH of the ripened milk was standardized at 6.54 to 6.5, coagulation occurred in 20-30 minutes. The vat was cut with 3/8 inch (9.5 mm) or 1/2 inch (13 mm) wire knives, depending on the desired moisture level of the cheese.

The curd was allowed to rest for 10 minutes after cut, then gently stirred for 30 minutes. The temperature of the cook was 104°F (40°F). No additional heat was applied. The curd firmed properly and was dipped or placed into open-ended stainless steel hoops placed on a plastic draining mat. The cheese was turned every thirty minutes for a total of 4 hours. The temperature was held at 104°F (40°C). When the pH of the cheese reached 5.3, the cheese, still in the hoop, was removed from the vat and placed in a cooler at 45°F (7°C) for 40 minutes.

At the end of the cool air drying, the cheese was removed from the hoop and placed in a saturated salt brine at 50°F (10°C). After 18 hours, the cheese was removed from the brine, dried, and vacuum-packed in heat shrinkable plastic bags.

The cheese evaluated at the end of one month had a mild, pleasant taste. Sharper flavors and more acid taste was achieved by increasing the percentage of S. thermophilus culture used.

In this study, a 10.5% yield was achieved from milk with 3.6% fat, making a cheese with a 44% moisture. The cheese could be sold after 30 days of aging.

A Bel Paese type cheese has been made using retentate concentrated 2.5 fold by ultrafiltration and following the processing procedure used for regular milk.

The body and texture were very good, but flavor development was not acceptable.

Fontina Type

European Fontina, Gouda, Jarlesburg type and American Baby Swiss manufacturing procedures were evaluated as possible bases for production. A modified procedure, which was a combination of all of these methods, was used to make cheese with the equipment installed at the Cal Poly plant. With minor modifications to this procedure, other styles of cheeses which are similar to Fontina could be made. By changing cultures, pressing procedures and warm room treatments, Gouda, Lacey Swiss, Farmers, Jarlesburg type and Baby Swiss could be made.

One important step in the processing of this cheese is the removal of whey and the addition of hot water to perform the cooking of the curd. This step influences the body and texture, final pH and flavor of the cheese.

The pressing procedures used, both under the whey and after molding, have a marked influence on the appearance and texture of the cheese. Changes in the pressing protocol often result in different cheeses.

The warm room treatment is a critical step for the development of proper flavor and appearance of this cheese. Treatments vary from a constant temperature of 50°F (10°C) for two to three months to combinations of warm and cold room times and temperatures.

Cal Poly Fontina Procedure

A flow chart for the processing of this cheese is found in Appendix 2.

The milk was inoculated with 1.5-2% mesophilic mixed multiple strain lactic starter culture. In order to have moderate eye development, 1 gram *Propionibacterium shermanii* per 440 pounds (200 kg) of milk was added. Culture was added to pasteurized milk warmed to 88°F (31°C). The milk was ripened for one hour or until a pH of 6.54 was attained. The pH of the milk at renneting appears to be a critical control point since it influenced the quality of the cheese.

Single strength calf rennet was added at a rate of 3 ounces per 1000 pounds (90 ml per 453 kg) of milk. A firm coagulum occurred in 20-30 minutes. The coagulum was cut with 3/8 inch (9.5 mm) wire curd knives. The curd was gently stirred for 15 minutes, during which time the curd firmed. No additional heat was applied at this time.

At the end of the 15 minutes, whey was removed until 30% reduction of the original milk volume was attained. This took approximately 5 minutes. Water at 130°F (54°C) was added until the original volume of the vat was reached. At this time, the temperature of the vat should have been 102°F (39°C). If the hot

water addition did not raise the temperature of the curds and whey to the desired temperature, the steam jacket was used. The addition of water should not exceed the amount of whey which had been drained. The amount of whey removed in combination with the amount of water that was added had a marked effect on the final pH, flavor and body of the cheese. The watered curd and whey were stirred for 1 hour, during which time the curd firmed.

The gathering and pressing of the curd under the whey, which is referred to as prepressing, is an important step. The vat must be measured and the curd must be pushed back and pressed so that portions of prepressed cheese can be cut from the mass that are the proper size and weight. If a solid piece of prepressed cheese is not used, eye development is poor and the cheese does not have the proper appearance. Pressing under the whey was performed by using a solid plastic sheet and applying 5 pounds per square foot (224.4 kg per m²) of force.

After 30 minutes prepressing, the whey was drained and the curd was pressed for an additional 30 minutes. The prepressed cheese was cut into 10 pound (4.5 kg) blocks, which approximated the size of the hoops, and placed into metal hoops lined with plastic cheesecloths. The cheese was pressed overnight at 5-8 psi in a conventional cheddar cheese press.

After pressing, the cheese was removed from the hoop and placed in a saturated salt brine at 50°F (10°C). Brine time was dependent on the size, shape and finish of the pressed cheese. In this case, the cheese was brined for 24 hours. The cheese was airdried and vacuum-packaged in heat shrinkable plastic bags.

The cheese was stored at 50°F (10°C) for 7 days. This was done to allow salt and moisture equilibration. The cheese was placed in a warm room at 65°F (18°C) and cured for 1 to 3 weeks. The shorter cure times resulted in cheese with little eye development. The 2-3 week treatments produced cheese with more eyes. These cheeses had an appearance similar to Danbo or Baby Swiss. After the warm room, the cheeses were placed in a cooler at 45°F (7°C) and stored for 1-2 months.

Moisture in the finished cheeses was 40-42% with a 50% FDB. Using this procedure, a 10.5% yield was attained. The cheese was ready for market 1 to 2 months after manufacture. By using the breathable plastic bag, repackaging of the cheese was not necessary.

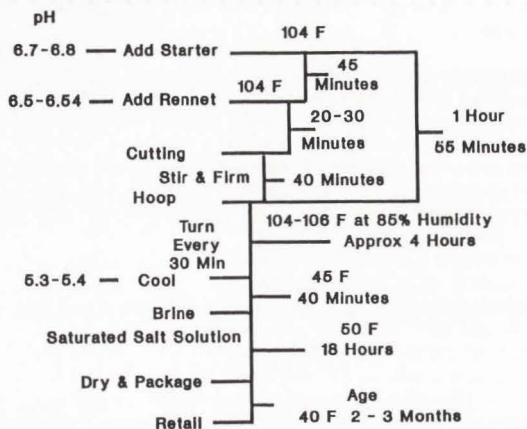
CONCLUSION

Bel Paese and Fontina type cheeses can be made using cheesemaking equipment normally used in America, with only minor modifications. The types of cultures, as well as, the combinations of starter cultures used have marked effect on the quality of the cheese. Cultures normally used for cheddar cheese production do not work well in Bel Paese and Fontina type cheeses. Cheese cultures used must be a combination of acid producing and flavor producing organisms to make cheese with the proper body, texture, and flavor.

APPENDIX 1

CHEESEMAKING CHART

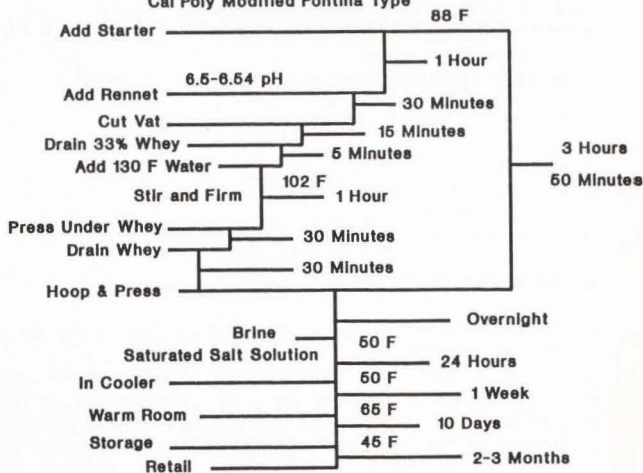
Cal Poly Modified Bel Paese Type



APPENDIX 2

CHEESEMAKING CHART

Cal Poly Modified Fontina Type



REMOVING CHOLESTEROL FROM MILKFAT USING SUPERCRITICAL CARBON DIOXIDE

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This research is the result of a grant from the dairy farmers of the State of Wisconsin through the Wisconsin Milk Marketing Board. The physical separation of cholesterol was conducted at PhaseX Corporation in Lawrence, MA, by Dr. Val Krukonis while the analytical and applications effort was at the University of Wisconsin.

One should also consider the impact on the consumer of "cholesterol-free" advertising on sales of animal-based products. One hard hit product is butter with a loss of 50% of its market in 25 years.

Another contributing factor is that Americans consume on an average of 500-600 milligrams of cholesterol per day. This is diametrically opposed to the dietary requirements set forth by the American Heart Association. They recommend no more than 300 milligrams of dietary cholesterol per day. This emphasis and awareness concerning dietary intake is continuing with advertising and with mass cholesterol screening programs scheduled for shopping malls regardless of the fact that there is insufficient evidence to link dietary intake of cholesterol and blood serum levels in the general public.

A valid picture of the thinking of a typical consumer can be realized from a bar graph in the Wall Street Journal from 6/23/88 (Figure 1). In caption was "Fear of Food:." Consumers were asked how much they were concerned about constituents in food; fat - 61%, cholesterol - 59%, thought these a serious health hazard while 35% thought both were somewhat of a health hazard. Thus, removal of cholesterol from products like milkfat without altering the composition or properties of the fat would appear to place milkfat in an enviable position in the marketplace. This figure expresses the doubt of the consumer over a broad spectrum of the food industry.

The process used for this extraction is a simple distillation that is conducted under pressure. Carbon dioxide in the supercritical state, thus a liquid, is the extractive solvent. The theory underlying this practice is not new; it is only the practical application that is. In fact, the subject of the solubility of solids in gases was reported in 1879 by Hannay and Hogarth.

Supercritical extraction using principally carbon dioxide is in use in many laboratories across this country, Europe and Japan. These bench top units are capable of handling up to about 200 grams of feedstock and separation of components is through an increasing pressure profile. There are very few pilot

units that truly simulate commercial scale in that large quantities of product can be processed and descending pressure profiling is used.

Commercially, supercritical extraction is in use by the food industry for coffee decaffeination, General Foods plant in Houston \$93,000,000, removal of bitter flavor from hops, Foster beer in Canada and Pfizer in the U.S., isolation of essential oils from spices and purification of omega-3 fatty acids from fish oils. However, more effort has been spent on coffee decaffeination than on any other food product to date.

A supercritical fluid is any fluid at a temperature above its critical value. A phase diagram (Figure 2) for a pure component such as carbon dioxide is shown. The solid-liquid and gas-liquid temperature/pressure lines ascend from the triple point to the critical point. At the critical point the properties of both liquid and gas are the same. As a supercritical fluid it has enhanced and unique solvent powers with high density, low viscosity and no surface tension to provide for easy penetration and diffusion into the food material to be extracted.

Another unique feature of supercritical fluid is that it can show a wide spectrum of solvent characteristics so that a supercritical fluid can be fine tuned to a specified solubility behavior as shown by the curves (Figure 3). In fact at low pressures the solubility of triglycerides in supercritical CO₂ is less at high temperatures than at low temperatures. This is contrary to our usual chemical reactions.

There are a number of gases and liquids that can be used as supercritical fluids; even water can be a supercritical fluid. These are shown in Figure 4 along with their critical temperatures and pressures. Clearly, only CO₂ is unique in the respect that it is inexpensive, nearly as abundant as water, nontoxic in any concentration, non-flammable, non-corrosive and with its low critical temperature, an attractive solvent.

Processing

Various milkfat products have been evaluated to find the ideal fat source: whole milk, 40% cream, 80% plastic cream, butter and butteroil made from these creams. The results of supercritical extraction indicate that plastic cream, butter and butteroil are near ideal sources. Another consideration is that the pH in the extractor is about 3.0, thus casein will coagulate. A cheese manufacturer would need to double separate incoming milk to obtain a plastic cream at 80% fat or higher. After extraction the milkfat could be re-incorporated back into skim milk to produce a milk of desired fat content.

Data (Table 1) used to calculate the distribution coefficients and the selectivities were from these results. Cholesterol values are from analysis of extracts produced by ascending profile separation. Pressures used as well as volume of CO₂ used are shown. The method used for cholesterol assay is the digitonin procedure for β -3-OH sterols as shown in A.O.A.C. (14th edition).

Two pieces of information (Table 2) are necessary in order to develop a process flow pattern and preliminary equipment sizing and configuration; the information needed is the distribution coefficient of cholesterol in supercritical carbon dioxide and the selectivity of carbon dioxide for cholesterol relative to the triglycerides in butter. These can be calculated given enough data. The distribution coefficient influences the recycle ratio of fluid, the solvent-to-feed ratio, and the diameter of the extraction column; while the selectivity influences the number of transfer stages or theoretical plates and the height of the column. Furthermore, there are many similarities between butter separation and the fractionation of fish oil ethyl esters. The information obtained in the fish oils study is directly applicable to the separation of cholesterol from milkfat. The separations of fish oil ethyl esters is practical and can be done commercially.

Equipment

Bench top equipment (Figure 4) is designed with flexibility to allow a wide variety of sample fractionation. The sample is held in the extractor at a preset temperature while the supercritical fluid, adjusted to the correct pressure with a diaphragm pump, is allowed to pass through. Fractions of the product in the extraction vessel are collected in the tubes which are at atmospheric pressure and temperature. The rotameter monitors gaseous flow/unit of time while the test meter monitors total flow.

Pilot scale equipment (Figure 5) to fractionate cholesterol from milkfat may be designed in more than one manner. This equipment requires two stage processing, i.e., the lower pressure first stage is to separate flavors and low melting triglycerides. The remainder with cholesterol is conveyed to the second stage higher pressure unit where cholesterol is separated from the high melting triglycerides. Note that provisions for recycling are a part of the equipment. Cholesterol and a small fraction of milkfat will be removed through the collection part marked "product." By subsequently blending appropriate amounts of light extract and heavy raffinate the manufacturer has the potential of producing a fat with a melting point range equivalent to butter or to produce a soft-spread butter. Both products with low cholesterol content. A cheese manufacturer can double separate, extract, then reincorporate with a sour cream valve at 200-400 psi to obtain one hour of fat emulsion stability before the enzyme-directed clotting of milk in the cheesemaking process.

It is difficult before scale-up to set a dollar value for this processing to extract cholesterol. However, knowing the cost of similar processing, one could establish a processing cost of \$.15 - .20 per pound of milkfat. For example, at \$.15 per pound of milkfat for a processing cost to reduce the cholesterol 90% of its original content, the added processing cost per pound of Cheddar cheese would be \$.05, i.e. $1/3 \# \text{ fat} / \# \text{ cheese}$. This increased cost probably would be lost in marketing. A similar scenario could be constructed for other dairy foods.

Supercritical extraction costs to produce a low cholesterol butter or cheese can be offset by the value of the by-product. Cholesterol has value as an ingredient in the manufacture of steroids and emollients. The current demand for

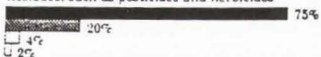
cholesterol suggests a price of \$.04/gram. If an operator of a commercial sized supercritical extraction unit could produce a 10% concentrate of cholesterol in milkfat, the value of that product would be \$1.77/pound of milkfat. Thus, a value greater than the parent compound.

For a cheese manufacturer taking in 500,000 pounds of milk per day the equipment cost to extract cholesterol from this milkfat is about \$1,000,000. Costs to install and test run the equipment will be about 2.5 to 4 times this cost.

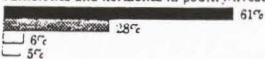
Fear of Food

When consumers were asked how much they are concerned about the following items in food, they said:

Residues, such as pesticides and herbicides



Antibiotics and hormones in poultry/livestock



Fats



Cholesterol

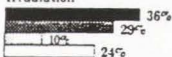


Salt



- Serious health hazard
- Somewhat of a health hazard
- Not a hazard
- Not sure

Irradiation



Nitrites



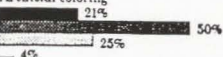
Additives and preservatives



Sugar



Artificial coloring



NOTE: Total percentages vary because of rounding
Source: Food Marketing Institute

Figure 1. Fear of Food (Wall Street Journal June 23, 1988)

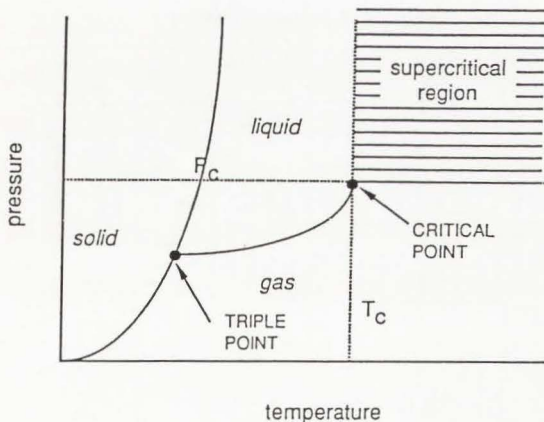


Figure 2. A Phase Diagram for Carbon Dioxide

Figure 3. Solubility of Fish Oil Triglycerides in Supercritical Carbon Dioxide.

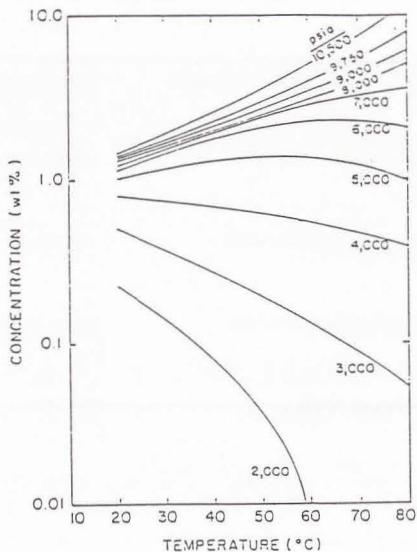
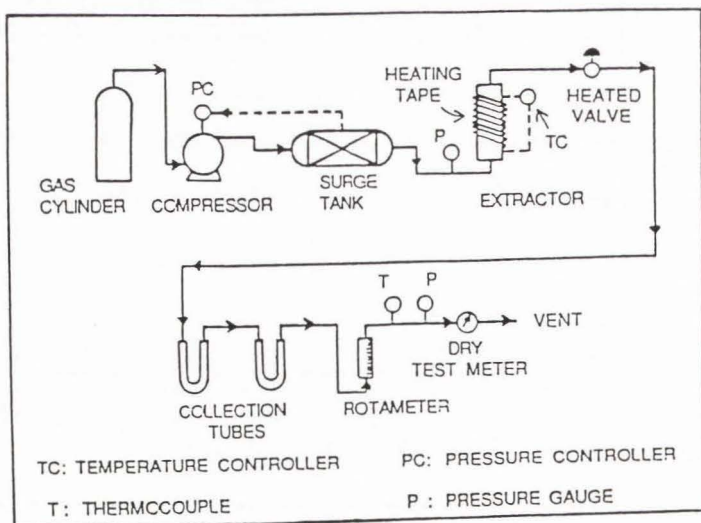


Figure 4. A Benchtop Supercritical Extraction Set-up.



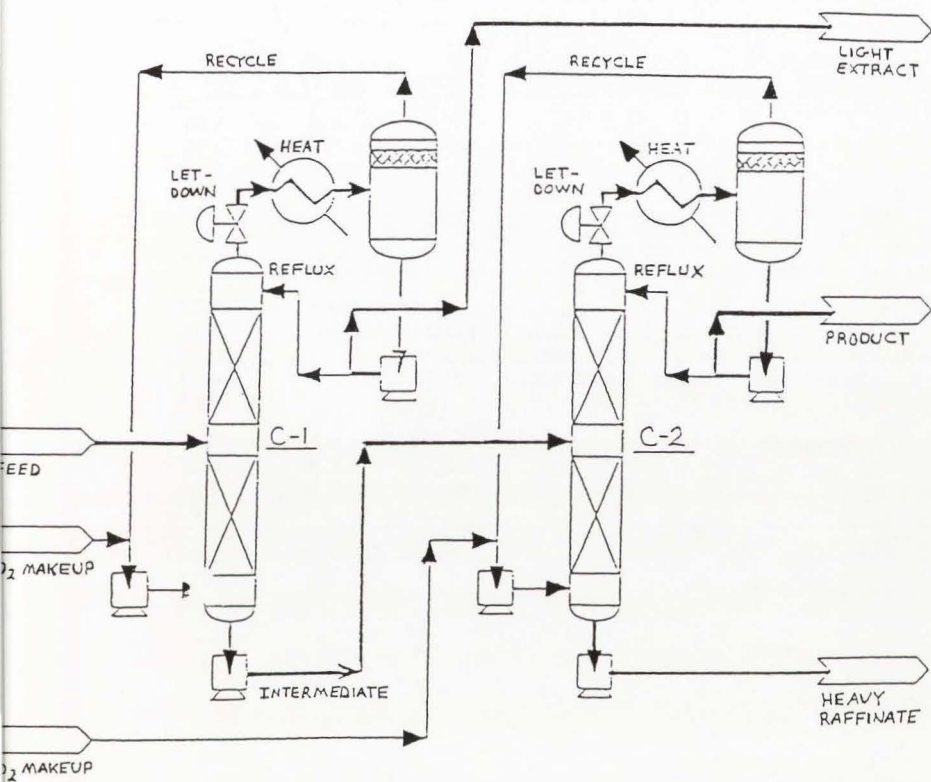


Figure 5. An Example of a Commercial Supercritical Extraction Layout

Table 1. Response of Cholesterol in Butter to Supercritical Carbon Dioxide

| <u>Butter</u> | | | | | |
|---------------------------|----------|---------------|--------------------------------------|---------------------|---------------------------|
| Trial No. and temperature | Fraction | Pressure psig | Std. liter CO ₂ /fraction | Fraction weight, g. | mg Cholesterol /g milkfat |
| WM-14 80C | 1 | 2300 | 500.00 | 0.86 | 0.00 |
| | 2 | 2500 | 500.00 | 0.13 | 11.66 |
| | 3 | 2800 | 300.00 | 0.35 | 10.62 |
| | 4 | 3050 | 300.00 | 0.51 | 5.65 |
| | 5 | 3350 | 300.00 | 1.01 | 2.68 |
| | 6 | 3750 | 300.00 | 0.99 | 1.43 |
| | 7 | 6000 | 300.00 | 2.05 | 0.25 |
| | | | | 6.10 | 2.02 |
| WM-21 80C | 1 | 2300 | 500.00 | 0.65 | 0.16 |
| | 2 | 2500 | 1500.00 | 0.51 | 12.42 |
| | 3 | 2800 | 1000.00 | 0.85 | 4.93 |
| | 4 | 3050 | 300.00 | 0.68 | 1.44 |
| | 5 | 3400 | 300.00 | 0.54 | 0.31 |
| | 6 | 6000 | 300.00 | 2.65 | 0.00 |
| | | | | 6.01 | 2.14 |
| WM-22 65C | 1 | 2100 | 150.00 | 0.56 | 0 |
| | 2 | 2350 | 1400.00 | 0.43 | 6.37 |
| | 3 | 2700 | 500.00 | 0.80 | 3.51 |
| | 4 | 3150 | 400.00 | 1.34 | 0.83 |
| | 5 | 3450 | 400.00 | 1.44 | 0.04 |
| | 6 | 6000 | 300.00 | 1.20 | 0.66 |
| | | | | 6.00 | 2.12 |

Table 2. Calculated Selectivities and Distribution Coefficients for Selected Supercritical Extraction Trials Involving Butter.

| Trial no and fraction | Selectivity | Distribution coefficient (X10 ³) |
|--------------------------|-------------|---|
| WM14-1 0 | .66 | |
| 2 | 5.84 | 3.60 |
| 3 | 5.31 | 3.90 |
| 4 | 2.81 | 6.40 |
| 5 | 1.32 | 10.0 |
| 6 | .13 | -- |
| WM21-1 0.77 | .06 | |
| 2 | 6.27 | 1.50 |
| 3 | 2.92 | 4.40 |
| 4 | .72 | 15.10 |
| 5 | .16 | 0 |
| 6 | 0 | 0 |
| WM22-1 0 | 0 | |
| 2 | 3.16 | 1.80 |
| 3 | 1.74 | 6.80 |
| 4 | .41 | 3.30 |
| 5 | .02 | 0 |
| 6 | .33 | 0 |
| Fish Oil | | |
| C18/C20 | 1.82 | |
| C20/C22 | 1.65 | |

**TOTAL QUALITY SYSTEM IN ITALIAN CHEESE PRODUCTION:
A COMPREHENSIVE QUALITY ASSURANCE PROGRAM**

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ABSTRACT

"Quality" is a customer determination based on the customer's actual experience with the product, judged against his or her requirements and expectations. The purpose of any quality assurance program is to provide a product into which quality is designed, built and maintained to allow the maximum customer satisfaction at the most economical cost. Traditionally, the Italian Cheese industry, like most other segments of the food industry, tries to achieve this goal by controlling the quality of raw material manufacturing process and extensive testing of the finished products for compliance with standards and specifications. However, the traditional approach to product quality has been shown to be inadequate, particularly with regards to microbiological contamination of the product.

The Total Quality System (TQS) is a company wide comprehensive system for coordinated actions of the management, labor, processing operations, distribution and marketing to achieve the goal of providing quality products to the customer. The TQS involves planning and establishing all elements necessary to assure safety and quality of cheese manufactured by your company. It involves designing and implementing a comprehensive Hazard Analysis Critical Control Points (HACCP) program emphasizing strict compliance with the Good Manufacturing Practices (GMPs), product recall plan policies, QA auditing program and employee and supervisory training program.

The TQS is voluntary. Successful implementation of the TQS approach results in improvement in product quality, production flow, employee morale and quality consciousness. It also leads to reduction in cost of quality which, in turn, increases cost effectiveness and productivity.

This presentation is designed to describe the Total Quality System approach for the Italian Cheese industry.

The U.S. Italian Cheese Industry has shown a steady growth in recent years (Table 1) (1). Nearly 1.8 billion pounds of Italian Cheese was produced in 1987 - a 10% increase over the 1986 production. About 6 million pounds of Italian cheese was produced in Wisconsin during 1987. This figure represented a 17.5% increase over 1986 production. The number of plants manufacturing Italian cheese increased from 177 in 1986 to 185 in 1987 (Table 2). The production trends reflect increased popularity of Italian cheeses, e.g. per capita consumption of Italian cheeses increased from about 1.0 pound in 1960 to over 6 pounds in 1985 (2). Several Italian cheeses, particularly Mozzarella have found a unique niche in specialty cheese market (3). Growing popularity of pizza and consumer trends toward take-out, Deli and convenience foods are likely to continue creating increased demand for an excellent quality product suitable for the varied tastes of consumers.

However, rising expectations of customers - both internal and external - along with a changing regulatory climate and competitive pressures from other food industry segments have greatly increased the demands upon the management for quality products. The Italian cheese manufacturers and distributors must be able to provide superior quality, value-added products consistently to compete successfully in the market place.

Quality Expectations

Although "quality" is a concept in every day use in our industry, it is often elusive. "Quality" means different things to different people. To some it is a vague and abstract notion. They can't quite put their fingers on it but say: "I know it when I see it" (5).

There are several definitions of food quality (Table 3) (12). Simply stated, quality product is one that meets the following criteria:

- Presence of desirable attributes
- Freedom from defects and impurities
- Compliance with regulations
- Standards and specifications
- Functionality
- Safety following the use

Quality of Italian cheeses, like many other dairy products, is comprised of two different aspects: Attributes and Excellence.

Attributes include factors such as temperature, pH, moisture content, weight, color, consistency, flavor, etc. that can be described objectively in terms of °C, % H₂O, grams, hue, chroma and value; flow behavior, etc. Excellence, on the other hand, is a value judgment based on subjective evaluation expressed in terms of grade, preference rating, acceptability, etc.

Quality is a customer determination judged against his or her requirements and expectations. Consequently there are very few, if any, widely accepted standards of Italian cheese quality.

A national survey of mozzarella cheese quality revealed variations in compositional parameters such as fat, FDM, moisture, salt and pH of cheese produced in various parts of the country (9). Similarly, differences in flavor, body and texture scores and melting quality were evident. However, 52/60 (87%) samples analyzed were within the compositional standards set by the FDA. Only 5 of 60 (8%) samples were rejected based on flavor alone. Since mozzarella is usually consumed in the melted state, e.g. on pizza and related foods, functional properties of the cheese viz. meltability, stretchability and elasticity as well as browning and oiling-off tend to dominate quality specifications prescribed by distributors and buyers (2,7).

Microbiological aspects of Italian cheese quality

Microorganisms in milk and those contaminating cheese and milk from the environment during processing(s), handling, storage and distribution play an important role in determining cheese quality, e.g. psychrotrophic organisms in milk can cause a variety of flavor and texture defects. They can also degrade milk protein which results in loss of yield. The presence of large numbers of coliforms in milk may indicate poor quality milk, inadequate pasteurization, undesirable cleaning and sanitation or general carelessness in milk and cheese handling. Microbiological quality considerations in Italian cheeses are often limited to determination of coliforms and yeasts and molds counts in finished product (6).

A number of pathogens viz. Staphylococcus aureus, E. coli, Salmonella, Listeria monocytogenes, Clostridium perfringenes, etc. have been detected in our milk supply (6,11). Although proper pasteurization of milk can destroy all but a few heat resistant species of enterococci and spore forming organisms, the potential health hazard posed by pathogenic organisms is a matter critical to economic survival of the industry.

A recent Salmonella javiana outbreak in Minnesota in which mozzarella and other cheeses were alleged to be vehicles of transmission of the organism caused major concerns among the consumers and cheesemakers alike. The merit of the public warning issued against the consumption of uncooked cheese notwithstanding, the incident stressed the importance of the most fundamental criterion of food quality viz. safety.

The Quality System

Italian cheese manufacture, like any other modern food processing, is a collection of activities and functions within the production system. Purchasing, product design, process development, manufacturing, inspection, warehousing and distribution and sales, are all different functions within the system. Each of the functions must include activities devoted to product quality. Thus quality

control represents a collection of activities within a production system coordinated to produce a product that consistently meets customer expectations and therefore is "... fit for its intended use."

In a small, single plant operation, where all production activities may be carefully controlled, the quality function is relatively simple. However, in today's multi-plant, giant corporations, with extended distribution and marketing territories, the task of producing a quality product is very complicated. Since every operating unit of the company influences the quality of the company's product(s) and since the company can remain in business as long as its products are acceptable to the consumer, a system approach to quality is desirable.

A system is defined as "... a physical or conceptual entity comprised of interdependent parts that interact within boundaries established to achieve some common goal(s)" (8). A simple production system consists of the input (material, labor, etc.), the production (manufacturing/processing), and the output (products/services) (Figure 1). Most systems incorporate some feedback information regarding process, compliance, actual production and other parameters to provide some control mechanism (Figure 1). The significance of the system approach is that it involves an orderly assessment and determination of compliance with requirements of the product.

Figure 2 shows an example of a quality system which is the network of administrative and technical operations required to manufacture a product of specified quality. The consumer's quality requirements and expectations are assessed and production, processing and distribution methods are planned to meet consumer needs (subsystem S₁).

Subsystem S₂ is a feedback system designed to provide prompt and continuing feedback important in making quality decision. The quality planning and manufacturing provide communications inputs to quality assurance and quality control. On the other hand, quality control operations provide feedback regarding the quality status of the product.

Traditional Quality Control Programs vs. The Quality System Approach

Traditional quality control programs in food industry are based on the following three principles (4):

1. Raw material control
2. Process control
3. Finished product inspection

The emphasis on testing of the finished product to determine product compliance with quality specification as practiced in traditional quality control program is ineffective. Once a food product has been through a manufacturing process, very little can be done to change its quality. The finished product

evaluation designed to permit acceptance or rejection of the product does not allow any preventative approaches for assuring the quality of the product. Also, traditional quality control tests for finished product evaluations are inadequate in providing any indication of foodborne pathogens. A carefully designed Hazard Analysis and Critical Control Point (HACCP) program as a part of the Total Quality System of product safety and quality assurance is highly recommended.

Planning a HACCP Program

The Hazard Analysis Critical Control Point (HACCP) program is an organized, documented QA procedure for the production of safe, high quality food product. It has been successfully employed in the low-acid canning industry for more than 20 years.

The HACCP involves two main aspects:

1. Hazard Analysis - A critical examination of entire food manufacturing process to determine every step, or point, where a possibility of physical, chemical or microbiological contamination may enter the food and render it unsafe or unacceptable for human consumption, and
2. Critical Control Points - A point in a food process where there is a high probability that the lack of control may cause, allow, or contribute to a hazard or to filth in the final food, or to decomposition of the final food.

As pointed out at this conference last year (10), the HACCP should be a priority for the Italian cheese industry. However, that is another topic, worthy of separate discussion in detail. The main steps in developing a comprehensive HACCP system are listed in Table-4.

Some of the major elements of the HACCP system are as follows:

1. Develop an up-to-date plant flow diagram indicating clearly various streams - raw materials, processed products, CIP-lines, etc. The process flow diagram may consist of several sub-systems with an overall flow diagram showing integrated systems. The product/process flow diagram must be accurate and match with plant engineering blue prints. An example of a process diagram for mozzarella cheese operation is shown in Figure 3.
2. Monitor quality of raw products and ingredients to ensure compliance vendor agreements and specifications. This is particularly important for minimizing the potential hazard of microbial contamination, metal fragments, filth and other impurities. Raw material quality control is the first line of defense against quality problems in finished products.

3. Determine process compliance by frequent, if possible, on-line monitoring of critical parameters such as temperature, pH, salt content, etc. It can be claimed that if quality control of raw material and ingredients is perfect and manufacturing process is in compliance with set specifications for that process, the final product will be a quality product requiring very little end-product inspection and testing.
4. In addition to cleaning and sanitation of processing equipment, control of plant environment is critical to product safety and quality. Many organisms can be transmitted through airborne contamination. Therefore, monitoring heating, ventilating and air conditioning system, drains, screen traps, etc. is essential for a successful HACCP. Results of dairy plant surveillance by the industry and the FDA has indicated that organisms such as *Listeria* may indeed be isolated from plant environment. Isolating critical areas from main traffic flow and minimizing employee movement from raw to finished area is critical in reducing the risk of pathogenic contamination.
5. Keep accurate records of critical control point monitoring and other process variable. Designate a specific location for these records and person(s) responsible for maintaining records of the critical control point monitoring.
6. Finally, plan a good product recall (retrieval program that is adequately tested). Designate a "response team" and a plan of action to be followed in the event of product contamination.

Summary and Conclusion

Traditional quality control programs in Italian cheese industry emphasizing subjective quality criteria and finished product testing may not always be adequate to assure quality of our products. The Total Quality System (TQS) approach based on a well designed HACCP program can help meet consumer expectation for safe, quality product. The TQS approach can accomplish the following:

- A product made right the first time, minimizing scrap, rework and customer complaints
- Uniform and reliable processing resulting in consistent superior quality
- Prompt effective and proper response in the event of pathogenic contamination, such action can diffuse consumer concerns and maintain the positive image of the company

Acknowledgement

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Table 1. Italian Cheese Production*, Selected States, 1986-1988

| States | Production (1000 lbs.) | | |
|--------------|------------------------|-----------|-----------|
| | 1986 | 1987 | 1988 |
| Wisconsin | 510,138 | 599,765 | 684,114 |
| New York | 286,097 | 289,785 | 293,831 |
| California | 224,938 | 244,461 | 293,831 |
| Pennsylvania | 167,312 | 171,850 | 179,403 |
| Minnesota | 31,445 | 35,065 | 38,061 |
| Illinois | 78,147 | 40,807 | 34,925 |
| US Total | 1,632,921 | 1,799,770 | 1,937,118 |

*Source: Reference 1

Table 2. Italian Cheese Plants, Selected States, 1988-1989

| States | Plants | | | Changes | |
|--------------|--------|------|------|-------------|-------------|
| | 1986 | 1987 | 1988 | '86 vs. '87 | '87 vs. '88 |
| Wisconsin | 56 | 58 | 53 | +2 | -5 |
| New York | 21 | 21 | 18 | 0 | -3 |
| California | 17 | 21 | 18 | +3 | -3 |
| Pennsylvania | 13 | 12 | 13 | -1 | +1 |
| Illinois | 8 | 8 | 8 | 0 | 0 |
| Minnesota | 5 | 5 | 5 | 0 | 0 |
| US Total | 177 | 185 | 176 | +8 | -9 |

*Source: Reference 1

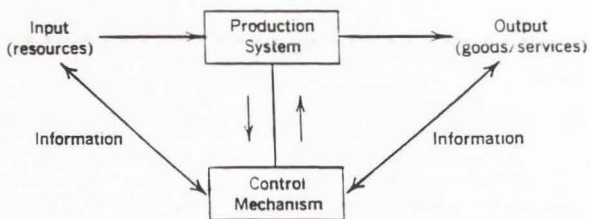


Figure 1. A simple production system with control mechanism. (Source: Reference 8)

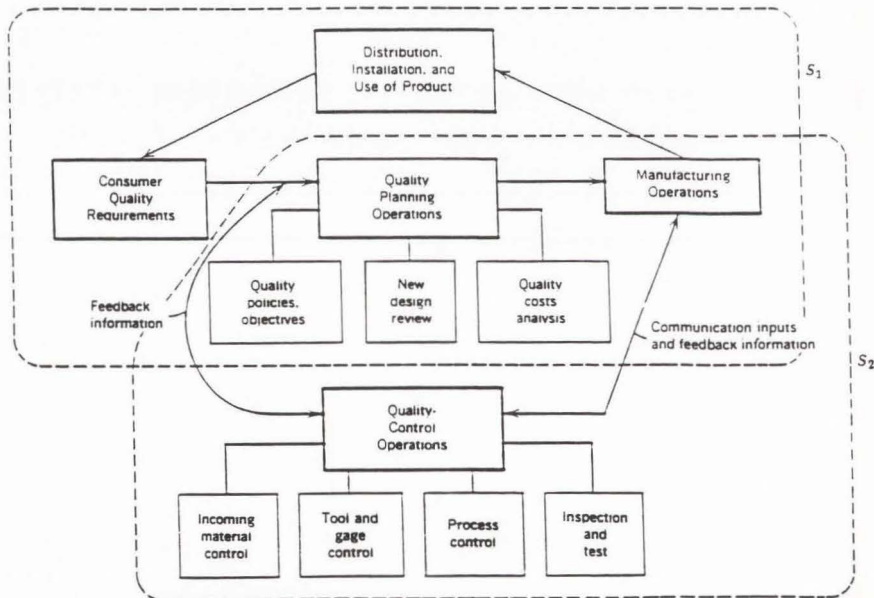


Figure 2. The Total Quality System - showing communication system dealing with consumer quality requirements (Subsystem S1) and quality planning function (Subsystem S2). (Source: Reference 8)

Table 3. Definitions of Food Quality

1. "... a multicomponent measure of the extent to which the units of a product, which a seller is willing and able to offer at a price, consistently meet the requirements and expectations of the group of buyers willing and able to buy that product at that price."
 2. "... a peculiar and essential character of the product. It has distinctive properties or characteristics, it is a degree of excellence, it is a fitness for use."
 3. "Quality is the extra component that distinguishes a product in its field."
 4. "Quality in the food industry includes: purity, safety, economics, taste and a variety of other issues. Above all quality must meet the requirements of the customer and consumer."
 5. "Quality is 100% conformance to consumer (Warren Schwecke).
 6. "... a combination of attributes or characteristics of a product that have significance in determining the degree of acceptability of the product to a user."
 7. "... the totality of features and characteristics of a product or service that bear upon its ability to satisfy a given need." (ASQC)
 8. "Quality is ... a product's fitness for use" (J.M. Juran).
 9. "Quality ... the presence of value as defined by the customer" (John Guaspari).
 10. "Quality is one that fulfills its intended purpose but does so at the least cost to the society" (Genechi Taguchi).
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Source: Reference 12

Table 4. Steps in developing a HACCP system

1. Designate a HACCP coordinator
2. Conduct a Hazard Analysis of ingredients and process
3. Develop accurate product and process flow chart
4. Identify the Critical Control Points for each hazard on the flow chart
5. Establish inspection and monitoring of controls and each CCP
6. Document and record results of all control procedures
7. Develop product retrieval and recall plan and policy
8. Develop an effective employee training program

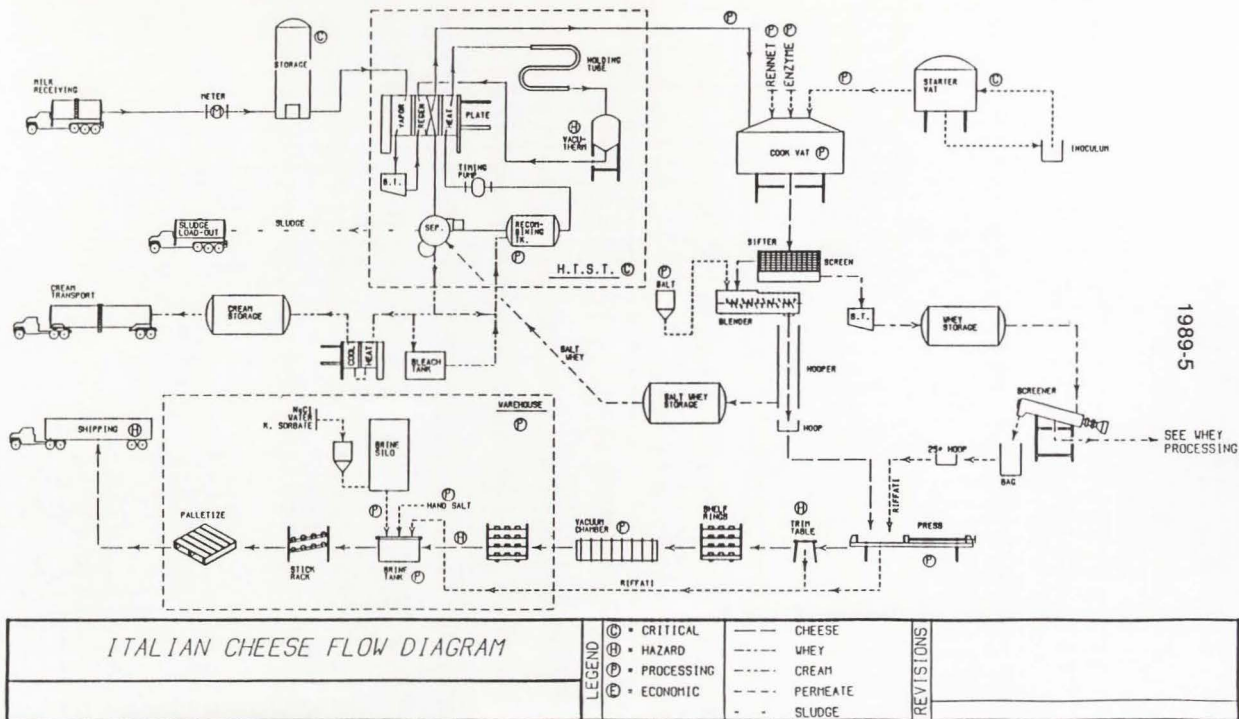


Figure 3. A product/process flow diagram for Italian cheese manufacturing showing Hazard and Critical Control Points.

EVALUATING CRITERIA FOR MOZZARELLA CHEESE MANUFACTURING UTILIZING MEMBRANE PROCESSING TECHNOLOGY

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Mozzarella Cheese Production using U.F. Milk

It might be best to start with a review of the Federal Standards for Mozzarella cheeses.

| | <u>Moisture %</u> | <u>FDB %</u> |
|------------------------------------|-------------------|--------------|
| Mozzarella | >52 - <60 | ≥45 |
| Low Moisture Mozzarella | >45 - <52 | ≥45 |
| Part Skim Mozzarella | >52 - <60 | >30 - <45 |
| Low Moisture, Part Skim Mozzarella | >45 - <52 | >30 - <45 |

The latter is the most popular.

It is easy to see from the Federal Standards for Mozzarella cheese that the nomenclature (Mozzarella) is misleading. All categories are usually lumped into one (Mozzarella). The total percent of cheese manufactured as "Mozzarella" is very small. In most instances, there are few manufacturers of this particular cheese. There are even fewer who can routinely manufacture it (>52 - <60% moisture) and attain over 52% moisture. It is easy to realize that two of the standards allow a total solids content of 40% with a fat (FDB) of 45% and >30 - <45%. With this low total solids content, the cheese could be manufactured from full concentration U.F. milk. There are a few products on the market in both the United States and Europe that contain near the maximum, 60% moisture.

In Italy, much of the Mozzarella cheese is manufactured and marketed as Water Mozzarella. The cheese is manufactured using direct acidophication, requiring no starter. Italians use primarily citric acid as the acidulant. Milk is standardized, pasteurized, acidified, set with coagulator, cut, cooked little or not at all above the set temperature, whey removed, firming of the curd, mixing-stretching of the curd in hot water, molding in balls (100-150 grams), packaged in sterilized-cooled saline solution (below 40°F) and marketed.

In the United States, this procedure is performed as described above, except acetic acid (vinegar from apple fermentation) is more commonly used. In certain parts of the United States, Eastern), there is also cheese manufacture that falls into the Mozzarella and Part Skim Mozzarella standards with the 52 - 60% moisture levels. These cheeses are manufactured using acetic acid (apple vinegar) or starter cultures as the acidulant. It is difficult to attain 52% or

more moisture using starter as the acidulant. The procedure is similar to that described above, except that the cheese is molded in various retail sized (4 - 32 oz.) direct salted (0.6 - 1.0%), molded and then the finish salted (1.5 - 1.8%) and cooled in refrigerated, saturated salt brine solution (40°F or below). The cheese is then packaged with continuous cooling, if necessary, before boxing. It might be possible to manufacture this product with a moisture content of near 60% utilizing U.F. Research is needed in this area.

The majority of Mozzarella falls into the pizza cheese category (low moisture, part skim Mozzarella). To my knowledge, there is little or no direct acidified pizza cheese manufactured in the United States. This would be an interesting research project for someone.

There has appeared and continues to appear "Light" or "Lite" Mozzarella cheese that could fall into any of the four Mozzarella standards. Let's discuss some of the product characteristics required for an acceptable pizza cheese, since the majority of Mozzarella cheese produced in the United States falls into this category (Low Moisture Part Skim Mozzarella cheese).

1. Flavor

- a. Flavor must be clean, typical of this particular cheese variety.
- b. Lack of flavor is not a strong objective.
- c. Cheese flavor is somewhat masked by the array of condiments added to the pizza.
- d. Plain cheese pizza would be affected by an off flavor, or lack of flavor.

2. Color

- a. White to yellowish in color.
- b. Bleaching is permitted.
- c. Slightly off color may not be a critical problem.

3. Body and Texture

- a. Firm to hard body.
- b. Must be shreddable.
- c. Must be grindable.
- d. Must be sliceable.

4. Meltability and Stretchability

- a. Must completely melt and cover pie.
- b. Must stretch when heated
- c. Must not reveal the shape of the cheese (shreds, etc.) when baked.

5. Butterfat Retention

- a. Little or no fat leakage (free fat) on pie during baking.
- b. Little is known why this happens.
- c. One of the most critical problems for the pizza parlour.

The next question might be "Can Mozzarella cheese (pizza) be manufactured using U.F. milk, and if so, which strategies could be used?" Let's define U.F. strategies.

Strategies

1. Full Concentration - Milk concentration by U.F. to the total solids content of fresh cheese. Little or no whey is released. Limited to cheeses with less than 50 -52% total solids.
2. U.F. and Whey Drainage - Concentrate to 4 - 6 X with some whey drainage depending on concentration of milk and total solids of cheese variety.
3. U.F. and Evaporation - U.F. to approximately 40% total solids and then evaporate to desired total solids level of product to be produced (58 - 70% T.S.).
4. Protein Standardization - With this approach, whole milk is concentrated two-fold using U.F., then traditional equipment is used for cheesemaking.

Total solids of Mozzarella cheese, according to the Federal Standards, are as follows: Mozzarella 40 - 48%, Low Moisture 48 - 55%, Part Skim 40 - 48% and Low Moisture Part Skim 48 - 55%. It is, therefore, obvious that Mozzarella and Part Skim Mozzarella can be manufactured from any of the U.F. strategies. Low Moisture Mozzarella and Low Moisture Part Skim Mozzarella cheese could be manufactured by all but the full concentrate strategy (U.F. and whey drain, U.F. and evaporation or protein standardization). Now let's discuss research data that will help us determine problems that could be encountered from utilizing any of the above U.F. strategies and its effect on the acceptable characteristics of pizza cheese (low moisture part skim).

1. Flavor

Data has suggested that casein is degraded slower (3,15,14), free amino acids are formed more slowly (10), β -casein is broken down at a much slower rate (3,14,16), and degradation of α -1 seems to be slower (16). This has been attributed to the inhibition of native milk proteinase, plasmin, and by the increased content of β -lactoglobulin (17). Recent evidence suggests that the proteolytic action of rennet is inhibited by whey proteins increased due to U.F. (2). Other possible causes include use of less rennet due to the increase in total solids (8). Less starter microorganisms or the rate of autolysis of these microorganisms are probably a factor (7). It is well understood that native whey proteins are not affected by rennet, starter enzymes, or plasmin in cheese (3,15). With the substantial increase of the whey proteins due to U.F., there will be a general slowing of the breakdown of the caseins. Flavor development in Mozzarella manufacture from U.F. milk will be curtailed. There are some who are not as concerned about flavor, due to the addition of many flavorful condiments to the pizza. The flavor cannot be bad or harsh, but the lack of

flavor is not a great concern. I disagree with this and would require the best flavor possible.

2. Color

It has been suggested that using U.F. milk for cheese production will cause a slight color change from the traditional yellowish to a more grayish color (15, 16). While this could be a problem in certain instances, the addition of food grade bleaches could be used to overcome this defect.

3. Body and Texture

It seems that when discussing this characteristic of pizza cheese, the industry needs and requires the best of both worlds. They expect a cheese loaf that is firm enough to be shredded or ground, and have a texture that will melt and stretch with little or no free fat (fat leakage on pie when baked). This further complicates the manufacturing process using U.F. If one removes the calcium to enhance the melting properties and at the same time causes the cheese to become too soft, in some cases liquid whey leaks inside the bag (13). Both of these defects are unacceptable. It has been reported that Mozzarella cheese can be manufactured from 2 - 5 fold milk and that the cheese has adequate melting properties (5,6,11). This data has been questioned and needs additional study.

4. Meltability - Stretching

One of the main characteristics associated with pizza cheese is the ability of the cheese to melt smoothly and evenly when heated, and its stretching to an acceptable length. Melting is even more critical than stretching. Cheese made from U.F. concentrated milk has impaired stretching and melting properties (1). It has been reported that the severity of the problem worsens as the concentration of the milk increases. Whey protein seems to be the cause of this problem. This problem seems to be due to whey proteins precipitation on the casein network. The denaturation of the whey proteins during heating, and their fixing on the casein makes it difficult or impossible for casein strands to move relative to each other (18). Another factor to consider is the high water binding properties of denatured whey protein reducing the free water in the cheese, thereby reducing the flow properties of the cheese when heated. It has been argued that cheese made from U.F. milk will produce curd having a coarser protein network than that of traditional cheese. It is believed that this affects the melting properties of the cheese. Another factor that might affect melting is the calcium balance. Denatured β -lactoglobulin is known to expose calcium binding free carboxyl groups. Regulation of the calcium content of the retentate has been reported to have a beneficial influence on melting and stretching of the U.F. cheese. It has been reported that by reducing the pH of the milk at the time of U.F. processing to a pH 5.8 - 6.0 and/or doing the diltation with a brine solution, there will be a significant improvement in the melting properties of the finished cheese. However, even utilizing these processing procedures, less than one half of the average melt-stretch is obtained when using full concentration of the milk (4,12).

5. Butterfat Retention

This is one of the most critical defects associated with pizza cheese. If the fat leakage (free fat) appears on the surface of the pie during cooking, it creates enormous problems for the pizza parlour. It has been reported (5,6) that generally more fat leakage is found with cheese manufactured from U.F. milk during cooking, even when the cheese is made from milk concentrated less than two-fold. It has been reported that this may be due to the effects of a coarser protein and fat distribution in the cheese (9,19).

Summary

1. Standards for Mozzarella cheese are broad and should be adequate for the production of any formula desired.

Moisture 45 - 60%

FDB 30 - 45%

2. Characteristics required for an acceptable pizza cheese:
 - a. Flavor
 - b. Color
 - c. Body and Texture
 - d. Meltability
 - e. Stretchability
 - f. Butterfat retention

3. Problems related to production of pizza cheese from U.F. milk:
 - a. Flavor - Greatly influenced by U.F. Could or could not be a problem depending on the desire of the individual consumer.
 - b. Color - Slight change from yellowish to grayish color. Could be corrected by the addition of a food grade bleach.
 - c. Body and Texture - Body is usually softer which causes problems when grinding or shredding.
 - d. Melting and Stretching
 1. Worsens as the concentration of the milk increases.
 2. Whey proteins precipitated on the casein network retard stretching and melting.
 3. High water binding properties of denatured whey protein.
 4. Coarser protein network.
 5. Calcium balance, denatured β -lactoglobulin is known to expose calcium binding free carboxyl groups.
 - e. Butterfat Retention
 1. One of the most severe problems for the pizza parlour.
 2. Generally more severe when U.F. milk is used to manufacture the cheese.
 3. Due to coarser protein and fat distribution in the cheese.
 - f. It is obvious that additional research is necessary to solve many of the above listed problems.

MOZZARELLA CHEESE STANDARDS

| | <u>Moisture %</u> | <u>FDB %</u> |
|------------------------------------|-------------------|--------------|
| Mozzarella | >52 - <60 | ≥45 |
| Low Moisture Mozzarella | >45 - <52 | ≥45 |
| Part Skim Mozzarella | >52 - <60 | >30 - <45 |
| Low Moisture, Part Skim Mozzarella | >45 - <52 | >30 - <45 |

**ULTRAFILTRATION
STRATEGIES**

1. Full Concentration
2. U.F. and Whey Drainage
3. U.F. and Evaporation
4. Protein Standardization

**CHARACTERISTICS
OF AN ACCEPTABLE
PIZZA CHEESE**

1. Flavor
2. Color
3. Body and Texture
4. Meltability
5. Stretchability
6. Butterfat Retention

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POTENTIAL FOR THE DEVELOPMENT OF BACTERIOPHAGE RESISTANT THERMOPHILIC CULTURES

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INTRODUCTION

Production of cheese and cultured dairy products relies largely on the fermentation of milk by the lactic acid bacteria. Of this group, the lactococci, lactobacilli, and thermophilic streptococci (*Streptococcus salivarius* subsp. *thermophilus*) are the primary bacteria used in both mesophilic and thermophilic milk fermentations for the production of a variety of hard and soft cheeses, buttermilk, and yogurt. Selection of lactic acid bacteria for specific roles in dairy products and processing is based on the fundamental knowledge that the particular strain or combinations of strains dictate not only the type and quality of the final product, but also the efficiency of the process.

In forthcoming years it is anticipated that increasing markets for dairy products and technology innovations in dairy processing will stimulate creation of new products and expand biological processing applications for fluid milk. The availability of lactic cultures with new, enhanced, or diversified capabilities will be essential to implement processing and product innovations. Thermophilic and mesophilic milk fermentations employing lactic acid bacteria occur in a non-sterile medium, pasteurized milk. As such these processes are highly susceptible to contamination with bacteriophages. Experience with mesophilic cultures makes it very clear that the longevity of any starter culture is dictated by the phage resistance or sensitivity of that culture (Heap and Lawrence, 1976, Klaenhammer, 1984). It is now well established that collective expression of different mechanisms of phage defense is responsible for the phage insensitive state of mesophilic lactococci. In this light, use of genetic technologies to engineer cultures that are insensitive to phage attack offers a long term solution to the phage problem in dairy fermentations. Genetic programs will be designed to engineer phage insensitive cultures by construction of plasmid and gene combinations that assemble multiple phages defenses which target and disrupt different points of the bacteriophages lytic growth cycle.

It is widely acknowledged that biotechnology will play a significant role in the growth of all fermentation industries over the next decade. With the recent accumulation of fundamental genetic information on the mesophilic and thermophilic streptococci, we are now in an excellent position to employ these technologies for the improvement and expansion of dairy products and fermentation processes. The following presentation will briefly discuss,

- A. the genetic tools and systems that are currently available to engineer dairy starter cultures,

- B. phage defense systems defined in lactococci and genetic approaches to engineer phage-insensitive cultures.
- C. phages characterized in *S. thermophilus*,
- D. strategies that can be applied to thermophilic starter cultures to engineer phage-resistant strains.

GENETIC TRANSFER SYSTEMS IN DAIRY LACTOCOCCI AND THERMOPHILIC STREPTOCOCCI

Any effort to apply genetic technologies to the improvement of dairy cultures requires the following. First, knowledge of the genetic, physiological, and metabolic properties of the bacterium targeted for manipulation. Second, the availability of efficient genetic transfer systems through which one can deliver plasmids or DNA carrying the genes of interest. Third, vehicles (plasmids, cloning vectors, phage) must be constructed to introduce, maintain, and express the desired genes and gene sequences. In the dairy streptococci (i.e., both the lactococci and *S. thermophilus*), we currently have at our disposal genetic transfer systems and cloning/expression vectors to deliver and maintain genes of interest. Gene transfer systems that have been developed include the following.

Transduction: phage mediated gene transfer.

Conjugation: exchange of genetic information via cell to cell fusion.

Transformation: electrotransformation of intact cells with plasmid DNA.

A number of cloning vectors and vehicles for conjugative mobilization are also now available for *S. thermophilus*. For references and discussion of gene transfer mechanisms and vectors the reader is referred to Fitzgerald and Gasson, 1988; Romero et. al., 1987; and Mercenier and Chassy, 1988; and Somkuti and Steinberg, 1988. Development of the genetic tools essential for manipulation and molecular analysis of the dairy streptococci has occurred only in recent years. We are now in a position where these technologies can be exploited for improvement of starter cultures used in dairy products and processing.

PHAGE DEFENSE SYSTEMS IN LACTOCOCCI

Bacteriophage infection of starter cultures is widespread among numerous commercial food and dairy fermentations (Sanders, 1987). The starter culture bacterium serves as a terminal host in the lytic cycle of the phage. After infection, the phage replicates its genome and assembles new particles (heads and tails) within the cell. The cycle is completed when the host cell bursts open and hundreds of progeny phage are released into the medium. The phage increases in population faster than the bacteria and eventually the starter culture is destroyed and the fermentation slowed or stopped. The major steps of the phage lytic cycle are illustrated in Figure 1 (Sanders, 1987).

Bacteria that are resistant to phage attack circumvent some point of the lytic phage cycle. Disruption of phage adsorption, DNA injection, DNA replication,

phage-directed transcription and translation, particle assembly and packaging, or cell lysis can halt phage development and prevent phage populations from increasing to levels that threaten starter culture activity and performance. Genetic studies with lactococci have identified three different defense systems that occur naturally in these bacteria and can function to inhibit phage attack, via interruption of some point in the phage lytic cycle (Figure 2; Klaenhammer, 1987; Sanders, 1988).

1. Interference with phage adsorption.
2. Phage restriction and modification.
3. Plasmid-induced abortive phage infection.

Genetic determinants for these phage defense systems are commonly associated with plasmid DNA in lactococci. Each of these mechanisms can provide a first line of phage defense, but as independent mechanisms they can be too easily overcome by phage. In the prototype phage insensitive strain Lactococcus lactis ME2, combinations of all three mechanisms are present and function cooperatively to confer a defense system that is relatively impermeable to phage (Sanders and Klaenhammer, 1984). The genetic evidence which has accumulated strongly indicates that the phage insensitive condition reflects the collective expression of multiple mechanisms of phage defense. Such naturally occurring strains are rare and not widely available to the dairy industry. Consequently, efforts to provide such strains must include genetic strategies to combine multiple phage defense within single strains of lactic streptococci.

One of the plasmid-encoded defenses identified from Lactococcus lactis ME2 has been used successfully to improve the phage resistance of commercial starter cultures. The plasmid pTR2030 (Figure 3) provides the bacterial cell with two important characteristics; resistance to phage and the ability to conduct a sexual mating process for exchange of genetic material with other bacteria. Phages species that attack lactococci can be defined in different morphology groups. These include small isometric-headed phages, large isometric-headed, and prolate-headed phages. Small isometric phages are the most common type of virulent phage encountered in cheese plants. When dairy fermentations are disrupted by phage, 80% of the time a small isometric type phage is involved. Prolate phages are not the most common phage encountered, but they usually have a wide host range and can attack numerous strains. Once established in a cheese plant these phages can be devastating to culture rotation programs. Although rare, large isometric phages are also virulent and can induce starter failures. pTR2030 interferes with the lytic cycle of all these phages, albeit to different degrees, by aborting the infection after injection of phage DNA. In each case, the phage adsorbs and injects its DNA. At some point thereafter, the plasmid interferes with the lytic replication cycle of the phage. In cells that contain pTR2030, the number of progeny phage released per infected cell is reduced significantly and consequently, the growth rate of phage population is slowed. For small isometric phages, the most common species responsible for failures in dairy fermentations, pTR2030-induced resistance provides complete protection (Jarvis and Klaenhammer, 1986).

The plasmid pTR2030 is "self-transmissible" and encodes determinants that provide for gene transfer via conjugation. A donor cell, carrying pTR2030, forms mating complexes with other bacterial cells (recipients). After physical contact, a fusion "bridge" is formed where genetic information can be transferred from the conjugal donor to the recipient strain. Conjugation processes are widespread in dairy streptococci and can be used to direct the transfer of important genes and plasmids to strains of commercial significance. In this regard, we took advantage of the conjugative ability associated with pTR2030 in order to direct transfer the plasmid into commercial cultures that were susceptible to attack by virulent phages in cheese plants.

In a collaborative effort with Mary Ellen Sanders, at Miles Inc., Biotechnology Products Division, Elkhart IN, conjugal strategies were developed for transfer pTR2030 into industrial strains (Figure 4, Sanders et al., 1986). It was important to design the manipulation so as not to alter desirable properties (acid production, proteolytic activity, flavor development) of the starter strains or add undesirable genetic markers (antibiotic resistance) typically used to select genetic recombinants from mixed populations. Fast-acid producing recipients were mated with a *Lactococcus lactis* donor which did not ferment lactose, was proteinase negative, and carried only pTR2030. Transconjugants were selected by challenge of the mating mixture with phage virulent for the commercial starter strain. Colonies that were phage-resistant and fast-acid producing were selected and scored for acquisition of the plasmid by hybridization with pTR2030-specific DNA probes. In cases where the rates of mutation and frequency of plasmid transfer are similar, use of a confirmation test is essential to distinguish between true pTR2030 transconjugants and phage resistant mutants.

Recombinant strains carrying pTR2030 were isolated in this manner and evaluated to determine whether or not,

- I. acquisition of the plasmid altered the acid producing ability of starter culture, and,
- II. if phage resistance was effective and maintained under the temperature conditions encountered during commercial cheesemaking.

The pTR2030 transconjugants showed no loss of acid producing ability in the presence or absence of phage during Heap-Lawrence starter culture activity tests. Furthermore, the transconjugants survived repeated cycles of the SAT test, typical of strains able to resist phage attack for extended periods in cheese plants.

pTR2030 transconjugants have been used successfully as single strain starters in dairy plants where the parental strains were rapidly attacked by phage. After a number of years of use in the U.S. and Europe, as DVS-frozen culture concentrates, failures due to phage attack remain to be reported. The approaches used in this study clearly demonstrated that the phage sensitivity of

existing commercial strains can be reduced or eliminated through genetic strategies which introduce plasmid-encoded defense systems.

In spite of the commercial success of these cultures, phages capable of circumventing the pTR2030-encoded defense have been detected through routine screening of whey samples. It should be anticipated, however, that starter cultures with single barriers of phage defense would be eventually attacked under the dynamic phage pressure of the cheesemaking environment. Phage capable of circumventing a single phage resistance mechanism should be expected to appear or adapt to any host that is used continuously. We are currently studying these phages to understand how they have adapted to circumvent inhibition by pTR2030, and to identify points in their development that may be susceptible to other defense mechanisms at our disposal. Numerous examples of the coevolution between phage and bacteria exist where constantly changing defenses and counterattacks ensures survival of both species. Consequently, our challenge is to engineer the bacteria to higher orders of resistance and, thereby, create an "evolutionary step" for the phage which is complex and highly improbable. In this light, genetic programs that are designed to develop phage-insensitive starter cultures for dairy fermentations must emphasize gene combinations for multiple defenses that can act in concert to prevent phage infection and proliferation. Application of molecular technologies to genetically stabilize important genes and improve their expression will undoubtedly contribute to the evolutionary distance that might be engineered between the phage and lactococcal host.

PHAGES OF STREPTOCOCCUS THERMOPHILUS

In comparison to the lactococci, phages attacking the thermophilic streptococci are not as well characterized or as frequently linked to fermentation failures. Phage attack of rod-coccus starters is usually attributed to infection of the *S. thermophilus* component of the culture. Failure of *S. thermophilus* disrupts the growth synergism with *Lactobacillus bulgaricus* resulting in slow or incomplete acid development (Veringa et al., 1968). A recent study of Neve et al., (1989) concluded that phages of *S. thermophilus* are all closely related and members of one major group. The phages were isolated from Swiss cheese and yogurt samples at different geographical locations in France, Switzerland, and Germany. Electron micrographs of 12 phages evaluated in the study revealed the same basic morphology; isometric heads and long, non-contractile tails (Figure 5). Biochemical and genetic analysis of the phages distinguished three phage subgroups with the following characteristics.

- Subgroup I: Genome size 41.2 - 44.2 kb, major proteins of 39.8, 24.0, and 14.8 kDal in SDS-PAGE; Two phages classified - 100% homologous.
- Subgroup II: Genome size 38 kb, major proteins of 30.9 and 20 kDal, but < 40% homology with phages from subgroups I and III; One phage classified.

Subgroup III: Genome size 33.8 - 36.7 kb pairs, main proteins of 30.9 and 24 or 26 kDal; Nine phages classified, > 50% homology between phages within subgroup III.

For additional information on S. thermophilus phages see Kivi et al. (1987) and Mata and Ritzenthaler (1988). Phages within all three subgroups are highly related. Neve et al. (1989) suggested that evolution of S. thermophilus phages was the result of a variety of recombinational events. The genetic rearrangements giving rise to the different phage strains must, therefore, be inclusive to a single progenitor group. Consequently, defense mechanisms that interfere with the lytic cycle of one S. thermophilus phage would be expected to operate equally against most, if not all, phages attacking the thermophilic streptococci.

GENETIC STRATEGIES TO ENGINEER PHAGE-RESISTANT THERMOPHILIC STREPTOCOCCI

Two genetic approaches are currently available to develop phage resistant S. thermophilus cultures. Through a traditional or classic approach, spontaneous phage resistant mutants can be selected directly from a sensitive population. For every 10^6 phage sensitive cells, one cell may be resistant because the phage cannot adsorb, infect, or proliferate on the mutant. If the sensitive population is infected with the lytic phage at multiplicities of infection of greater than 1, phage resistant mutants can be selected from the surviving colonies. Considering the close relatedness of S. thermophilus phages, resistant mutants should be recalcitrant to attack by other streptococcal phages and recombinational routes minimized where new lytic phages might develop. Therefore, classical selection of naturally-occurring mutants would be expected to be a viable approach for the development of phage resistant thermophilic streptococci. It must be emphasized, however, that any resistant mutant must be thoroughly characterized to assure that the strains desirable fermentative properties (growth, acid production, proteinase activity, flavor development, co-cultivation interactions) have not been altered. Historically, selection and use of phage-resistant mutants in mesophilic starters has been problematic because the mutants often revert to phage sensitivity, may be attacked by a new phage, or exhibit undesirable fermentative characteristics. Current multiple-strain starter programs for lactococci have overcome these problems by development of effective methods to select fast-acid producing mutants and accurately predict their longevity in the cheesemaking environment (Heap and Lawrence, 1976; Huggins, 1984; Klaenhammer, 1984). Similar approaches should be equally successful in S. thermophilus.

Second, directed genetic improvements through engineering of phage resistance mechanisms offers an alternative to the above classical approach. Gene transfer mechanisms (conjugation and electrotransformation) and vehicles (cloning and mobilization vectors) are available for delivery and expression of gene/plasmid-encoded phage defense systems in S. thermophilus. However, the following questions remain to be answered. Are there native phage resistance mechanisms available in S. thermophilus? Will

the defense systems defined in lactococci function in S. thermophilus and which of these might be most effective?

There have been no reports of studies on native phage defense systems in S. thermophilus, even though restrictions and modification systems most certainly exist in this bacterial species. In a study of DNA transfection (phage DNA transformation) in S. thermophilus, Mercenier et al. (1987) reported operation of a DNA restriction system. The frequency of transfection was reduced 10 fold on strain AO23 when non-homologous phage DNA was used in the assay. These data implicate operation of a phage restriction and modification system, but biochemical, genetic, or operational evidence (i.e. host-dependent phage replication) for R/M systems in S. thermophilus phages have yet to be presented. Native gene-directed mechanisms that might interfere with phage adsorption or abort infection of S. thermophilus phages have also not been investigated to date. Therefore, immediate efforts to introduce such mechanisms in S. thermophilus would require use of the appropriate heterologous defense systems defined in the mesophilic lactococci.

Of the three major classes of resistance mechanisms (restriction - modification, abortive infection, and interference with phage adsorption), R/M systems are universal among prokaryotes and should function given the proper replicons to maintain heterologous R/M genes and suitable expression/regulatory signals that are recognized by the target host, S. thermophilus. In this regard, preliminary efforts to introduce and express lactococcal R/M systems in S. thermophilus have not been successful in our laboratory. Although the vectors and transfer systems are operating, lactococcal plasmids carrying R/M determinants were either not expressed or stably maintained in S. thermophilus. Similarly, it has not been determined if lactococcal mechanisms that interfere with adsorption or abort phage infection will function in S. thermophilus. Ultimately, delivery of lactococcal R/M or other phage defenses to thermophilic starters may require more sophisticated approaches to maintain and express these gene systems. These could include placing the lactococcal structural genes under S. thermophilus regulatory signals or cloning (in vivo or in vitro) the defense systems on replicons functional in S. thermophilus.

An additional source of genetic material that might interfere with phage infection could be isolated from the genome of the phage targeted for inhibition. DNA sequences of S. thermophilus phages could be cloned and used to express antisense RNA or present phage regulatory signals in a manner that disrupts normal phage development. Expression of phage antisense RNA has been shown to interfere with the development of coliphages (Coleman et al., 1985).

CONCLUSION

Future developments in starter cultures for dairy fermentations will be driven largely by the events now occurring in biotechnology. Looking backwards through 50 years, the dairy industry evolved from undefined to defined culture systems and realized substantial improvements in quality, efficiency and uniformity. The next evolutionary step that will occur in the dairy industry will

establish the competitive advantage to the cheesemaker with defined genes and gene systems in highly specialized starter cultures. If thermophilic fermentations require phage resistant S. thermophilus, such starter cultures can be developed. The biological technologies are currently available and the genetic strategies established to minimize phage infection and proliferation in dairy starter cultures that are susceptible to phage attack.

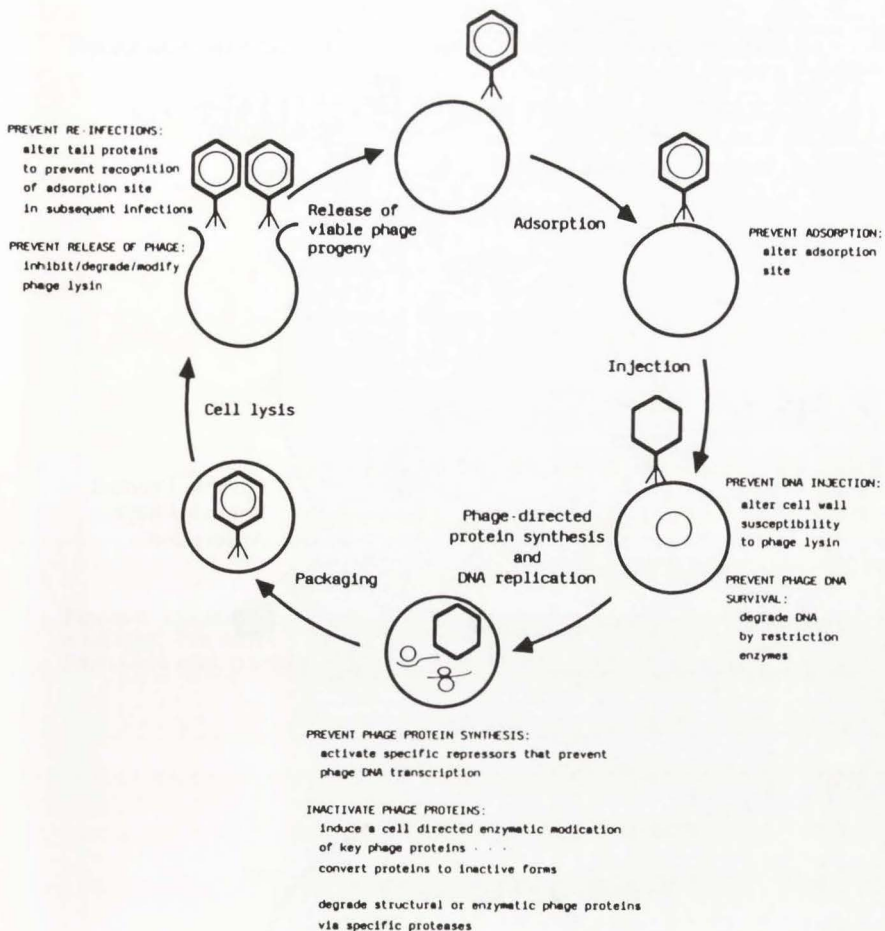


Figure 1 Fact and fantasy of bacterial interference with phage development. Described are some potential mechanisms whereby a cell could defend itself against phage. In some cases, these mechanisms have been experimentally verified (e.g., restriction/modification systems, alteration of cellular adsorption sites). In most cases, with industrially important phage-host crosses, however, reports suggest an inhibition of phage development, but mechanistic studies have not been pursued. From Sanders, M.E. (1987) Phage Ecology.

Figure 2
 PLASMID-ENCODED DEFENSES IDENTIFIED IN LACTIC STREPTOCOCCI

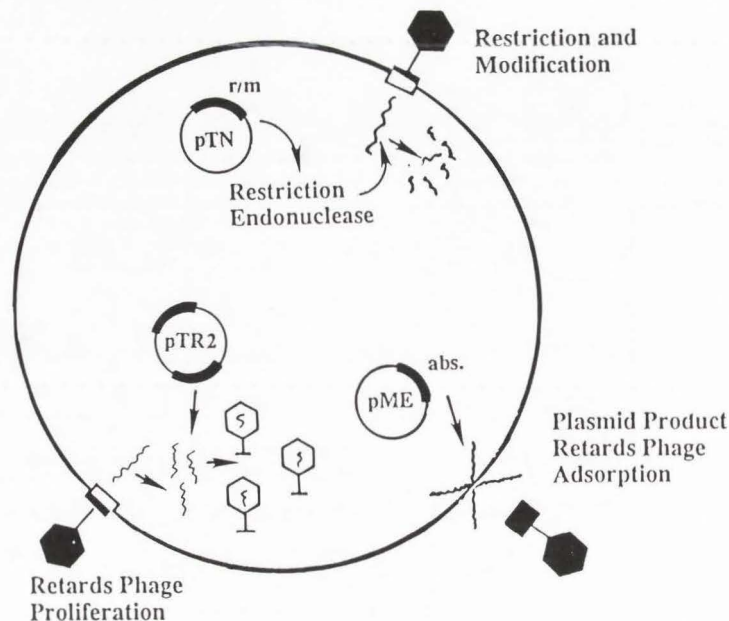


Figure 3

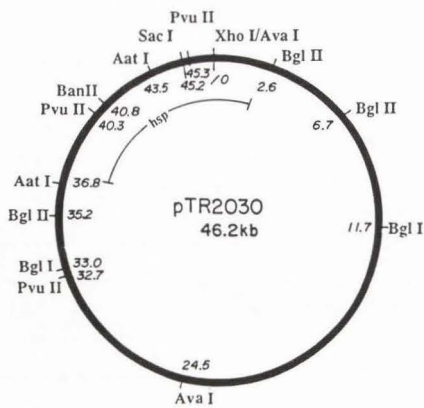


Figure 4

GENETIC STRATEGY FOR CONSTRUCTION OF
STARTER CULTURES TARGETED FOR FOOD FERMENTATIONS

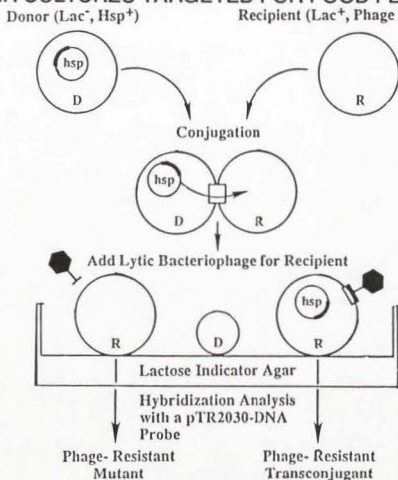
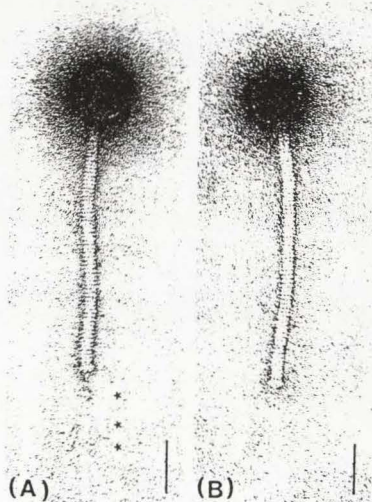


Figure 5 Electron micrographs of bacteriophage P53 with (A) and without (B) a tail fibre. The nodes on the tail fibre are indicated by asterisks (bars: 50 nm). From Neve et al. (1989)



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CULTURE PERFORMANCE IN MOZZARELLA CHEESEMAKING

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While some mozzarella cheese is made in the U.S. using direct acidification methods, most of the mozzarella cheese produced here today is manufactured with starter cultures. These starter cultures, along with rennet, and cheese milk itself, constitute a dynamic, ever-changing, biological system for the cheesemaker to deal with. It is therefore vital that the cheesemaker understands the parameters within which the starter bacteria grow and function, and what effects each manufacturing step can have upon the starter bacteria, and their associated functions in the cheesemaking process.

Let us begin our discussion by comparing some of the characteristics of thermophilic starter bacteria (Tables 1 and 2). It should be noted that these are general characteristics of the respective bacterial genera, and the characteristics may and will vary some from strain to strain.

Table 1. Some Characteristics of Thermophilic Rods and Cocci

| Rods | Cocci |
|--|----------------------------------|
| Longer generation times | Shorter generation times |
| Greater total acid production | Less total acid production |
| Lower final pH | Higher final pH |
| Galactose variable | Galactose negative |
| Sucrose negative | Sucrose positive |
| Acetaldehyde production | Acetaldehyde production |
| Strong proteolysis | Weak proteolysis |
| Strong aminopeptidases <Lh>Lb=St) | Strong aminopeptidases |
| Weak dipeptidases | Strong dipeptidases |
| Provides stimulatory amino acids | Stimulated by amino acids |
| Stimulated by formate, CO ₂ | Produce formate, CO ₂ |
| Optimal pH's <6.0 | Optimal pH's <7.2 |
| Optimal temp. approx. 45°C | Optimal temp. approx. 45°C |
| Sl. narrower growth temp. range | Sl. wider growth temp. range |
| Less antibiotic sensitive | Greatest antibiotic sensitivity |
| Less NaCl sensitivity | Greatest NaCl sensitivity <2% |
| Less phage sensitivity | Greater phage sensitivity |
| High phosphate sensitivity | Lower phosphate sensitivity |

Table 2. Galactose Utilization and Proteolytic Activities of Several DSS Thermophilic Strains

| Strain | Galactose Utilization | Casein Hydrolysis PAU/10 g (DWB) | Amino peptidase PEP/10 g (DWB) | Ratio * PEP/PAU |
|--------|-----------------------|----------------------------------|--------------------------------|-----------------|
| R110 | Weak | 9.9 | 11.7 | 1 |
| R120< | Weak/Moderate | 2.5 | 62.0 | 25 |
| R130< | Strong | 0.7 | 13.8 | 20 |
| R140 | Strong | --- | ---- | -- |
| R150< | Strong | 1.2 | 69.6 | 58 |
| R160 | Strong | 3.4 | 41.0 | 12 |
| R170 | Weak | 3.8 | 4.3 | 1 |
| C110 | Negative | 0.3 | 4.5 | 15 |
| C120 | Delayed/Strong | 1.5 | 9.8 | 7 |
| C130 | Negative | 0.1 | 3.4 | 34 |
| C140 | Negative | 0.1 | 7.9 | 79 |
| C150 | Negative | 0.5 | 12.5 | 25 |
| C160 | Negative | 0.1 | 15.9 | 159 |
| C170 | Delayed/Moderate | 0.4 | 6.9 | 17 |
| C180 | Negative | 0.1 | 0.2 | 2 |
| C190 | Delayed/Moderate | 1.8 | 0.3 | <1 |
| C200 | Negative | 2.5 | 8.3 | 3 |

< = *Lactobacillus helveticus*

* = Those strains exhibiting highest PAU/PEP ratios generally do not produce bitter flavors.

Let us now apply some of the above bacterial traits and functions to mozzarella cheesemaking steps.

RIPENING/SETTING

The length of ripening time is influenced directly by the methods of starter propagation. If the starter has been grown up under traditional methods, i.e. acid ripening, the cocci are stressed by the acidic conditions and will perform sub-optimally until the bacteria repair their acid-damage. This is expressed as increased lag time, and acid-damaged starter usually requires higher inoculum rates. By allowing acid-damaged starter to grow in the higher-pH conditions of cheese milk, acid-damage is eventually overcome and normal acidification proceeds (log phase). In the case of acid-ripened starter, the cocci are most severely impaired by acid. Therefore, under conventional starter conditions it is

important to have higher ratios of rods so that some acidification by the rods takes place, while the cocci are recovering from acid-induced lag phase.

If starter has been grown under some form of pH-control the bacteria are not subjected to acid-damage, and thus cheesemaking requires lower inoculum rates, less ripening time, and generally lower rod numbers.

Temperatures for setting the vat usually vary from plant to plant, and can range from 31°C (88°F) to 38°C (100°F). Increases in temperature not only speed up the enzymatic action of rennet, but also move into temperature ranges more favorable for thermophilic bacteria. Although thermophiles will grow and produce acid at temperatures in the low 90's, optimal growth and acid producing temperatures are closer to 45°C (113°F). For most strains, increases in acid production, due to increasing temperature, are almost linear from 30° to 40°+ C.

We have noted through field experience, and also in the literature, that the thermophilic cocci seem more prone to bacteriophage attack than are the rods. We do occasionally see a rod attacked by phage, but phage incidence is much greater for the cocci. The ripening period represents the most phage-susceptible stage of cheesemaking. Because phage are non-motile, their contact with starter cells is through random collision. In this case, random collisions are aided by agitation of the cheese milk. Extended ripening times increase chances for phage attack through prolonged agitation, high surface-to-volume ratios in the vat during filling, and the lengthy exposure time. Once the vat is set, phage adsorption does not easily occur.

COOKING

As mentioned above, optimal growth and acid-producing temperatures for thermophilic rods and cocci are near 45°C (113°F). These bacteria, depending on the strain will produce acid up to temperatures of 48-52°C (119-125°F). In some instances acid production is uncoupled from growth at these high temperatures. Above temperatures of 48-52°C, acid production drops off dramatically as seen in data from Martley (Figure 1). Characterization data gathered in our lab substantiate these findings. As temperatures increase from set temperature to final cook temperature, acid production by the starter bacteria increases almost linearly. Cook temperatures reached during mozzarella make are usually not high enough to inhibit rods and cocci; instead they move into more favorable temperatures for the bacteria, and speed up acid production.

It should be noted that the conversion of lactose to lactic acid differs between rods and cocci, and also between rod species. *Streptococcus salivarius* ssp. *thermophilus* (coccus) converts only the glucose portion, (one-half of each lactose molecule), to lactic acid. The galactose half of the lactose molecule is excreted into the surrounding medium. This is significant since the relative numbers of reducing sugars remain almost constant, even though their mass is decreasing by half. These reducing sugars are available to react in the Maillard browning reaction, when the cheese is placed in an oven. Galactose utilization

by the rods is variable from strain to strain. Most strains of Lactobacillus bulgaricus are almost or totally galactose negative, whereas strains of Lactobacillus helveticus are galactose positive, i.e. they are capable of converting the entire lactose molecule to four molecules of lactic acid.

The significance of the above information becomes apparent when one examines acidification curves for rods and cocci (Figure 2). The galactose negative rods and cocci produce acid exponentially until they reach pH values around 5.3 at which point the slope of the acidification curve flattens sharply. Acid production is greatly slowed, and will continue for a while, but at ever-decreasing rates. However, due to the high buffering capacity of cheese curd in this pH range, very little pH drop will be observed in the cheese. This point will be discussed further under mixing/molding.

WASHING

The starter bacteria rely on a constant supply of lactose to continue acid production. It is common practice, in many plants, to predraw a portion of the whey and replace it with water, or to add water to the curd during the "cheddaring" stage of acid development. Cold water addition results in higher moisture cheese, but might also result in temperature decreases which will slow acid production. The amount of water added, along with the degree and time of agitation, will tend to wash lactose and lactic acid from the curd particles into the surrounding whey, as solutes seek to reach equilibrium between curd and whey. Since the majority of the starter bacteria are entrapped in the curd matrix, the degree of washing can dramatically limit the availability of fermentable carbohydrate, and thus slow acid production rates. While this step can help reduce browning, it must be carefully controlled so as to not disrupt or slow acid production. Again, optimal temperatures for these starter bacteria are well above 40°C (104°F).

MIXING/MOLDING

The "stretch" in mozzarella cheese is almost entirely dependent upon cheese pH. Optimal stretch occurs in a pH range from 5.4 down to 5.1. In this pH range the rennet-modified casein is converted from the dicalcium to the monocalcium form, which in turn reduces calcium bridging and allows cheese fibers to stretch. Until proper pH conditions are met, no stretch occurs. Mozzarella cheese made with cocci alone loses its "drive" (logarithmic acid production) as it approaches the stretch pH range. The pH may slowly creep into the desirable range, but the make process is slowed down. The addition of rods helps to "finish off" the pH drop into the stretch-range, as well as smooth out the cheese body and texture. Where pH optima for cocci occur well above pH 6, the pH optima for rods lie in the mid to low pH 5 range. As cocci begin to wane in this pH range, the rods are growing in exponential phase (particularly galactose-positive rods), and are stimulating some acid production in the cocci.

Temperatures of 63°C (145°F) and above, encountered in the mixing/molding process, are high enough to stop acid production by the thermophilic cultures.

However, depending on the time/temperature relationship, some of the bacteria (especially cocci) may survive up to 30 minutes of heat treatment. As surface temperatures of the cheese decrease, coming out of the mixer/molder, starter bacteria will resume fermentation as long as lactose is available and other conditions are favorable. Acid production in the center of the cheese, where temperatures decrease more slowly, will be delayed in relationship to the cheese surface. Acid will be produced only at temperatures which are favorable and where nutrients are available. Further cooling of the cheese, prior to brining, slows down microbial metabolism and acid production can slow to almost nothing.

BRINING

The brining step quickly cools the outside surfaces of the cheese, so it will retain its shape, and also slows acid production on the outer surfaces of the cheese. As the center of the cheese cools to more favorable temperature ranges, some acid production resumes. However, as the cheese cools further, starter metabolism in the center of the cheese is slowed and acid production soon stops.

Strains of *S. thermophilus* exhibit greater sensitivities to salt than any of the other lactic acid bacteria. Salt concentrations of less than 2% are sufficient to completely stop acid production by cocci. It should be noted that salt penetration of brined cheese is fairly slow, and requires several days to reach equilibrium within the cheese block. Salt concentrations on the outside surfaces of the cheese are high enough to inhibit acid production by starter bacteria, but acid production within the cheese block would be unaffected. Availability of fermentable carbohydrate and temperature of the cheese play greater roles in controlling acid production than does salt.

STORAGE

Because mozzarella cheese is generally consumed as a fresh cheese within a few days of manufacture, proteolytic breakdown of the cheese is not usually a cause for concern. However, in cases where excessively high numbers of rods were used, or non-starter lactobacilli survived pasteurization, soft-body defects have appeared. Generally, proteolysis in mozzarella is lower than in Cheddar cheese of the same age, even though thermophilic starters are usually more proteolytic than mesophilic starters. Most, if not all, of the rennet has been inactivated or diluted by the mixing/molding step. Also, total bacterial populations are lower than those found in Cheddar cheese. According to Nilson and LaClair (1975), lower bacterial numbers in Mozzarella cheese, are a result of low setting temperature, optimal growth temperatures only being approached during cook-out, high steam/water temperatures encountered during mixing and molding, high temperatures being maintained in the cheese block after molding, and immediate cool down of the cheese to sub-optimal growth temperatures after molding. In short, growth conditions remain optimal for only a brief portion of the total make time.

CONCLUSION

Successful cheesemaking relies not only on the careful control of the biochemical and microbiological interactions occurring simultaneously in the cheese vat, but also on the knowledge of microbial control points, and what effect each control point has on the current, as well as subsequent cheesemaking steps.

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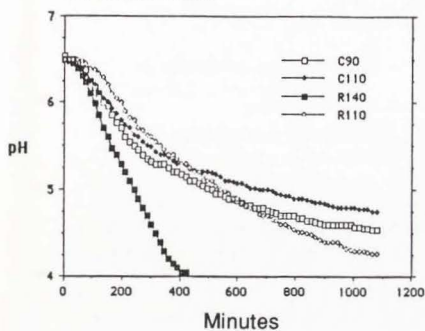
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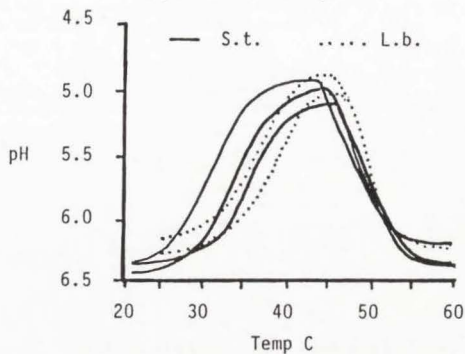
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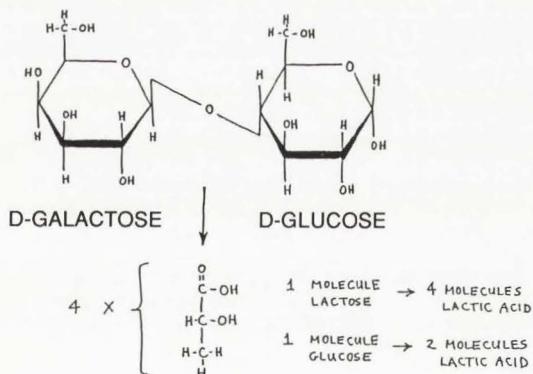
Growth Curves for Selected
Rods and Cocci



Adapted from Martley, 1983.



LACTOSE



SOME ASPECTS OF ITALIAN GRANA CHEESE TECHNOLOGY

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Grana is the general common name given to a large round cheese with extremely hard rind on account of its granular appearance when broken.

Grana-type cheeses have been made in the Po Valley under different local names as long as many centuries ago.

At present, according to Italian standards, there are two main types of Grana cheeses: Parmigiano-Reggiano, which is made in the territorial framework of the town of Parma and Reggio; and Grana Padano, which is made in the upper Po Valley and some of the Italian Alpine areas.

Both types are summing up the biggest outlet of the Italian single cheese milk utilization and require continuous penetrating knowledge to keep them in a solid balance within the domestic dairy industry. The two types, which are produced almost in the same size, shape and quantity, differ principally, besides the milk quality related to the respective production areas, in details of manufacture, in milk setting time, in fat content, in ripening time and in the use of additives.

Both types are now produced all year long, but in the past Grana cheesemaking was mainly a seasonal activity devoted to the conversion of surplus milk into a stable product. The remarkable biological stability of Grana cheese lies in the achievement, in the course of manufacture, of a set of condition which provide both a steady barrier to adverse micro-organisms, and a unique long ripening time.

The ancient Grana cheese prototypes differed noticeably from the modern one.

The factors which have controlled the survival of the present day varieties are mainly the modern facilities for the cheese milk standardization and the easiness with which the manufacturing process can be carried out.

From early centuries onward there have been no significant advances in Grana cheese technology until about the end of the last century when natural whey starter was introduced, commercial rennet preparations became available, and steam heated copper kettles appeared.

Before that time the evening milk, after standing overnight in shallow pans or trays for creaming, developed some lactic acid fermentation, which acted as a starter. After the removal of the cream layer, the evening milk was mixed with

the morning one, also shortly settled for creaming, and heated in copper kettles with wood fire to about 34°C; the exact temperature was depending both on the season and on the milk acidity. The kettle was then taken off the fire and the milk renneted. The usual setting time was from 20 to 60 minutes. The curd was then cut very fine and scalded with stirring to 46° to 52°C for 30 to 60 minutes with some time interval.

It was early recognized that, depending on the milk acidity, even different time/temperature ratio of making Grana cheese were conductive almost to the same product. At that time the control of milk production was rather difficult and the cheesemaker had to be very skillful to coordinate the basic technological factors.

The adoption of the natural whey starters, coupled with steam heated copper kettles, from about the end of last century has brought substantial advantages to the standardization of the Grana cheese technology.

At the beginning the natural whey starter was mainly a mixed lactic acid culture weakly acidified, and it was used in rather small quantities according to the milk acidity (1).

At this time the farmhouse Grana cheesemaking was found to be suitable for factory cheesemaking. The factory system development, which took place mainly in the fertile irrigated plain of the upper Po Valley, naturally encouraged those methods which could be less subject to the uncertainty of the milk and curd behaviors. Among those methods a twice a day cheesemaking was introduced: one for the evening milk and the other for the morning milk. After standing in larger basins and deeper layers for 6 to 8 hours for creaming, the evening milk went into the cheese vat early morning and the morning milk followed the same cheesemaking process early in the afternoon. This method prevented milk acidification. As a consequence the cheese milk had to receive more starters and the cheesemaking time was shortened. Natural whey starter went up from .60 to 1.30 percent or more acidity with a final dominance of thermophilic lactobacilli.

This factory system has been extensively adopted in the Grana Padano cheese area. On the other side Parmigiano-Reggiano cheese type is still based on a once a day cheesemaking mixing together evening and morning milk.

With the exception of those factory methods which could be better standardized and reproduced than the old farmhouse Grana cheesemaking, little advance was made in the first part of this century. During recent decades great changes and developments have influenced and are still moving the milk production side. Concerning the Grana cheesemaking we can consider the following points:

- 1) the consistent increase of cow herd size and high milk yield extend the time elapsing from milking to the reception of the warm milk at the Grana cheese factory;

- 2) milk casein content is generally decreased from 2.60 to 2.40%;
- 3) the ration casein/total milk nitrogen is in many instances closer to 77% than to 78% as it was before;
- 4) also the ratio between the colloidal calcium phosphate and casein appear to be declining;
- 5) the buffer value of milk and whey is also decreased;
- 6) mais silage is now largely used for feeding dairy cattle in the Grana Padano area;
- 7) besides the butyric acid bacteria and the other gas forming bacteria in milk originating from feeding the cows with silage in the Grana Padano area, the overall picture of the microbiological quality of the present milk is changed.

Taking into account that the old Grana cheese varieties arised from an area in which many differences in the form of crop and animal husbandry, in soil from hills to fertile irrigated plains, influenced the milk quality and local preferences for different flavors, we can understand that with the recent extensive changes in animal husbandry, in breed and feeding, the Grana cheesemaking technology had to adjust the methods of manufacture in order to meet the changing cheese milk and curd behavior and the new market requirements for traditional Grana cheeses.

The Grana cheese technology is still following the basic principles of cooked cheeses obtained from raw milk collected twice a day, partially skimmed by natural cream rising, but some modifications have been introduced and new fine traits in both Parmigiano-Reggiano and Grana Padano cheesemaking are emerging in spite of all the old art and craft still lasting in the face of the environment changes recently occurred.

Good quality constitutes the most valuable asset of any cheese. Grana cheese is not an exception. In this respect the cheese milk quality is certainly of primary importance. One of the most interesting aspects of the general question of cheese quality in relation to milk origin is the statement that the real Grana cheese quality can only spring from certain areas. The opinions expressed on this subject are still controversial and a search for the evidence of the different assertions does not bring to a sound conclusion (2).

However recently (3) some fine points of the Parmigiano-Reggiano cheese composition and proteolysis have been evaluated and recognized as closely related to the combined effect of the milk source and the cheesemaking process.

But the great number of variables involved in Grana cheese milk quality are rather difficult to control. The microbiological quality, combined with the absence of pasteurization and the long milk setting for creaming and ripening, still remain a crucial point both in Parmigiano-Reggiano and Grana Padano cheesemaking. This aspect is difficult to evaluate because of the influence of the ripening process on the wide variation in the type and number of bacteria in raw milk.

During the natural creaming of the milk which is laying in thin strata (10 to 20 cm) under 20°C from 6 to 10 hours, more than 80% of the original bacteria present in the milk are entrapped in the fat globule clusters of the rising cream and with the same cream carefully separated. This process can be considered from a general microbiological standpoint a cold pasteurization which eliminates also large part of the heat-resistant bacterial spores like butyric acid bacteria.

Meanwhile, during the milk standing time some groups of bacteria, mainly mesophilic, can develop. The milk undergoes some biochemical and biophysical changes during this slow, long ripening process. Even experienced Grana cheesemakers with good technological background have expressed the opinion that milk can't be either too clean or too fresh for the best Grana cheese quality. For a long time it has been recognized that a large number of micro-organisms, which grow in milk and produce taints and gas in mild acid conditions can be present in the Grana cheese milk vat and yet the cheese turn out to be of excellent quality. As a matter of fact all those bacteria are nullified by the huge number of strong lactic-acid thermophilic bacteria added to the cheese milk with the natural whey starter. Nevertheless a low count milk as free as possible from cheese fault-producing bacteria is going to be imposed and preferred. It can be said that nowadays it is easier to obtain a clean milk from the farm and may be also that the bacteria groups in today's milk are rather different than those found in the past.

A special problem is posed by silage feeding and butyric acid bacteria in the Grana Padano cheese area in which maize silages belong to the common diet of dairy cows; while in the Parmigiano-Reggiano cheese area, in order to avoid late blowing, a silage free diet is prescribed.

At the early stage of the silage feeding late blowing used to be controlled by adding approximately 30 ppm formaldehyde to the standing milk for natural creaming. This practice coupled with an extreme curd drying, lower pH and more salt in the cheese, has been pursued for at least half a century.

In the last 20 years, a steady intensification of corn silage feeding and a general weakening of the cheese milk casein content have been observed.

Milk with a poor casein content had to be manufactured in a shorter time to avoid structural faults in the cheese (mainly split defects). But a rapid manufacturing process is leading to an increased final pH in cheese which promote the butyric acid fermentation. This trend required new approaches to prevent late blowing caused mainly by Clostridium tyrobutyricum in Grana Padano cheese.

One has been the purification of cheese milk by bactofugation. Extensive experiments (4) revealed that the spore level in contaminated milk can be reduced by a factor of ten (about 90%).

The other approach has been the use of lysozyme. After an extensive investigation (5) the use of lysozyme at a 20-25ppm level in cheese milk for Grana Padano cheese came to be a general practice in silage feeding areas.

The use of the bactofugation process is still restricted to a few factories mainly because of its cost compared to lysozyme use. A combination of bactofugation and lysozyme would be better, but very expensive.

Nevertheless, the use of lysozyme to prevent butyric fermentation in Grana cheese requires to be fully effective, very clean milk containing approximately less than 500 of clostridia spores per liter.

The action of lysozyme can also be strongly aided by proper acidification process with which the pH of the fresh cheese body should drop as low as about 5.0.

Recent unpublished investigations seem to confirm this last point. The use of two consecutive milked milks cooled and stored at 8-10°C at the farm and then treated in the usual way for creaming at the cheese factory, appear to be a very promising new line to keep Italian Grana cheesemaking totally on fresh unpasteurized milk (6).

But the new and already common way for Grana cheesemaking is the short time manufacturing process carried out in the traditional 2,500 pound steam-jaket, cone-shaped, copper vat, delivering two forms of approximately 80 pounds each at the ripened point.

In the old time the curd formation took 20 to 30 minutes or even more, and the cooking time was usually kept 35 to 50 minutes. Around 1950 the coagulation and the cooking times were approximately 15 and 25 minutes, respectively. Now with the short time manufacturing process two divergent routes can be observed: one over 20 minutes (usually 22 to 30) with a fine granulation of the coagulum (size of a wheat kernel); the second one below 20 minutes (usually 15 to 18) in which the curd is carefully but coarsely cutted in shattered, irregular pieces of the average size of half nut. In both cases, but especially in the second one, the coagulum is cut in a very soft state. The softer and shattered curd grains increase curd surface area and whey expulsion rate. As soon as the milk coagulum appears, the whey expulsion takes place in a very short time indeed and also the cooking temperature is quickly risen from 32-33°C (90°F) to about 55°C (130°F) in more or less 10 minutes time. Cooking the curd immediately after cutting results in a drier cheese and higher fat loss in the whey (7). In the case of the fine granulation of the coagulum the actual cut to cook heal time will increase.

The actual Grana cheesemaking has become a full speed process that would be impossible to implement without the real play maker instrument: the high flexible conic shaped copper vat. Such a quicker Grana cheesemaking process will be hardly absorbed in elaborate mechanical equipments.

The value of scientific control in Grana cheesemaking is now only gradually appreciated because of the limitations of the techniques available and also for the great number of variables involved. Grana cheesemakers are very skilled technicians by virtue of their own experience and of their training under other experienced Grana cheesemakers, but usually have neither the time nor training to make complicated scientific tests. Even today some of the most experienced Grana cheesemakers disdain the value of the scientific quality assurance because they find themselves in a better position to control the behavior of the milk, starter and curd from day to day or from vat to vat. However in the last time scientific trained people and specialized labs are cooperating with Grana cheese factories and more and more cheesemakers are willing to accept scientific knowledge even if it is incomplete.

The new short time manufacturing process has been developed to counteract the gradual weakening of the cheese milk behavior due mainly to a poor casein content. The firmness of the coagulum and the syneresis forces are reduced by milk with a poor casein and colloidal calcium phosphate content. Starting with the right pH (usually around 6.40) in the milk cheeses vat just before renneting, the combined effect of the short processing times (curdling, cutting, heating and cooking) accelerate syneresis to an enormous rate: the curd moisture decrease from about 90% to about 50% in 10 minutes time. It is like a superefficiency whey permeation process.

With the short time manufacturing process the development of acidity in the cheese vat is practically inexistent and a more pliant and rubbery cheese texture is obtained even from milk with a low casein content.

As a matter of fact the processing time and the lactic acid fermentation temperature profile are so short into the cheese vat that the acidity development will take place only into the molded and already dried cheese. This technology is supported by the use of both a high quantity of rennet, usually in powder form and with a low content of bovine pepsine, and great quantities of a strong acid whey starters.

Natural whey starter is still the most important Grana cheesemaking factor. No other aspect deserves more attention.

In the old time the natural whey starter used to be a mixed culture of streptococci and lactobacilli weakly acidified and used in rather small quantities according to the cheese milk acidity. Today is a strong acid culture based almost exclusively on vigorous thermophilic lactobacilli producing lactic acid at a quick and steady rate, without appreciable proteolysis or lipolysis, in a wide range of temperatures (from 48° to 35°C). The whey starters are renewed daily just by setting the fresh hot whey from the cheese vat at a slow cooling temperature for about 18 hours. After 8-10 hours a good whey starter should produce at least 1.40% at the end point with the desired clean lactic flavor. Not always the natural whey starter can adapt itself to unsuitable environment like milk composition variations, high scalding temperature, use of lysozyme, etc.

Studies of biotype distribution have been made (8) in order to isolate the best suitable strains of thermophilic lactobacillus. But until now, any real and complete replacement of the natural whey starter has been found profitable. Selected cultures are now used only to complement or reinforce the natural one and the future outlook is probably for a mixed solution.

As we have seen in the actual Grana cheese manufacturing process the starters bacteria will activate the lactic acid production only inside the dried cheese already molded. The whey starter with about 1.30% acidity is added to the milk at about 3% on milk volume, which will introduce approximately 20 million lactic acid bacteria per ml of cheese milk. These bacteria will concentrate further, almost tenfold, inside the cheese curd. But the large mass of Grana cheese does cool very slowly and does not cool uniformly because of its poor thermal conductivity. Temperature measured at the center (15cm depth) remains around 50°C for about 10h after manufacture (9) whereas near the surface (3cm depth) temperature drops quickly. As a consequence the starter bacteria should be able to resist and to grow even at the high Grana cheese internal temperature and also to be capable to lower quickly the cheese pH and to ferment all the sugar content at the same time.

As a future outlook for Grana cheese we can consider that sooner or later the fat content of cheese will attract the unfavorable attention of the consumer. The production of low fat cheese which still taste good may offer great market opportunities. Old style Grana cheese had a protein/fat ratio around 2, almost like a creamed cottage cheese, and also retained the highest content of calcium and phosphorus among all cheese types.

At the present state Grana cheese is made with a protein/fat ratio closer to 1.4 still retaining its highest calcium and phosphorous content. But the old low fat Grana cheese with its fine, sweet, mellow, nutlike flavor could be easily resumed to fit better in the modern dietary guidelines.

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U. S. CHEESE MARKET VS. EUROPEAN CHEESE MARKET: SITUATIONS AND OPPORTUNITIES

By Gerald Dryer, President
The Jerry Dryer Group
2085 Charter Point Drive
Arlington Heights, IL 60004

It is truly a pleasure to be with you this afternoon to discuss the future of the cheese business and, in fact, the future of the entire dairy business -- here in the United States and worldwide.

Marschall Dairy Products is to be commended once again for hosting an excellent conference and show. And an extra thank you to Marschall for being foresighted enough to include a discussion of the world marketplace on the agenda.

Today, the dairy foods business certainly is operating in a world market. Yes, numerous barriers -- quotas, tariffs, health standards and subsidies -- still stand in the way of free trade, but there are a number of major changes in progress and the world will look a lot different in just a few years.

This afternoon, I would like to address some of these changes -- the situations mentioned in the title of my speech and, more importantly, the opportunities being created. However, the title is a little misleading. In today's economic, political and social climate, we can no longer afford to talk about the U.S. market versus the European market. And we cannot continue to talk about the cheese business alone. The cheese business is too closely interwoven with the fabric of the total dairy foods business.

To understand where the cheese and dairy business is headed internationally, we need to first step back and look at domestic agricultural policy here and abroad. Tremendous changes have transpired on both sides of the Atlantic during the past decade.

First, U.S. policy: for nearly 40 years, the U.S. price support program dictated ever-increasing prices for milk and finished dairy products. That all changed when Congress began to deregulate the farm milk price in 1983.

The price setting mechanism and a healthy dose of politics had raised the support price beyond reason. Add productivity gains on the farm and we were drowning in a sea of surplus milk. It was costing taxpayers too much just like a lot of other government programs. The budget axe fell and the support price was cut from more than \$13 per hundredweight in 1983 to about \$10.50 today.

That's price deregulation and it will continue. There will be further deregulation in the next farm bill -- if the surplus goes up, the minimum price will go down.

This deregulation is not without problems. At the plant level, you have all been experiencing roller coaster milk prices. The cheese price also has been on a roller coaster, and not always headed in the same direction as the milk price. That has caused more than enough problems at the plant level. Just like other industries that have been deregulated; the dairy business is in the midst of changing times and instability.

Business is further complicated by re-regulation -- USDA's tinkering with the internal workings of the price support program. By maintaining or increasing the powder price and lowering the butter price, USDA has changed the rules of the game.

Some are questioning USDA's tactics, but the net result is correct. We must lower the value of butterfat and increase the value of the nonfat solids in milk. That's what consumers are telling us. That's what the marketplace is saying. Government regulation is being replaced by the rules of the marketplace.

The net effect of all of this change is lower milk prices. And productivity gains at the farm level will mean lower milk prices in the future. In real 1989 dollars, the U.S. milk price is as high as it will ever be. That means the United States is quickly becoming the world's low-cost producer of milk. Yes. Some countries like New Zealand can produce milk for less, but their volume is not significant in terms of total world supplies.

As the United States becomes the low-cost producer, we will gain access to markets in other countries. It also means the end of import quotas that now protect U.S. cheese manufacturers from lower-priced foreign products. The quotas won't be eliminated during this round of trade talks, they probably will be weakened.

There will be a steady movement toward fewer artificial barriers aimed at keeping foreign cheese and other dairy products out of the U.S. market. Instead, our lower costs and prices will become a natural barrier. As the production of European-type cheese varieties increases in the United States, another natural barrier to imports is being built.

Next, let's look at European policy. In much of Europe, dairy/agricultural policy has also undergone dramatic changes. Just like American consumers/taxpayers, Europeans got fed up with the price tag for support programs.

However, European policymakers approached the problem differently. They froze the support price and established quotas to control the supply of milk sent to market.

In 1984, the support price was locked in through 1992. No increase for nearly 10 years.

At the same time, quotas were set to equal the average production of 1981 through 1983. The net effect was an almost immediate 4.5 percent reduction in milk marketings. The quotas have been reduced further since then and milk marketings are now 8.5 percent below the 1981-83 average.

When Europe had a huge surplus, much of the excess product was sold overseas by using export subsidies. In other words, the government helped exports sell into the world market where prices were well below the prevailing prices in Europe. European traders had developed an excellent customer base and suddenly there was no product to export.

The traders wanted to keep their international customers and turned to the United States for a supply of product -- especially milk proteins -- nonfat dry milk and whey. The net result: a powder shortage that has driven our price to record highs.

With the powder price so high, butter/powder plants have been able to compete very effectively with cheesemakers for the available supply of milk. There is enough milk being produced right now for the domestic market. In fact, butter is moving to the government as surplus. Cheesemakers should be able to get enough milk for cheese, but they can't because of the world's appetite for nonfat dry milk.

U.S. cheesemakers are deeply involved in the world dairy market -- like it or not. Cheesemakers really have no choice in the matter and the degree of involvement will continue to grow. It is time to learn more about the international marketplace.

In Europe, the support program has always focused on butter and powder -- the two products the government purchased as part of its support program. With the support program curtailed, more of the available milk has been shifted to the production of cheese, fresh dairy products and whole milk powder. Cheese output is up 11 percent and whole milk powder, up 32 percent. That says to me: Europe will be a larger force in the world cheese market. However, European traders will be in the market with fewer subsidies available to them. The playing field is being leveled.

With European milk production and prices frozen until at least 1992, there are opportunities for U.S. butter and powder in world markets.

There is tremendous potential for the United States and Europe to significantly increase dairy product sales in several regions of the world. Look at the marketplace:

In the United States: there is still good steady growth for specialty cheeses. We've barely scratched the surface of reduced-fat product sales. A steady parade of new products are using cheese as an ingredient. The pizza business is still growing.

In the Middle East: the Europeans have been building markets there for some time and the growth potential is huge.

On the Pacific Rim: there is a huge and untapped market. Twenty years ago, the Japanese didn't eat steak. Today, they pay 10 times the U.S. price for red meat. Cheese can follow the same track. Ice cream and frozen yogurt sales are booming in the Far East. Australia and New Zealand are "wheying" their appetite for dairy products; but those two produce less milk than the dairy farmers of Wisconsin. We can help develop and capitalize on that potential.

In Latin America: ditto the Pacific Rim. Most of these economies are improving and they want to eat better. We should be encouraging foreign aid in the form of food.

I know you can't find enough milk right now for your domestic customers. But six months or a year from now we may have milk running out of our ears.

It takes lots of development and relationship building to get into the international market. Get started now.

Encourage your trade associations to get involved. Learn from your neighbors who are selling whey and nonfat dry milk overseas. Check out government assistance and visit trade shows overseas.

In the United States and Europe, it is very clear: milk production will not be increased and milk prices will not be increased at the expense of taxpayers. Improved financial rewards can only be achieved through increases in productivity and in the marketplace. Fortunately, the world is becoming our marketplace.

Happy marketing.

**FOUNDING AND OPERATING A SPECIALTY CHEESE FACTORY --
THE MOZZARELLA COMPANY**

By Paula Lambert,
Founder and President
Mozzarella Company
2944 Elm Street
Dallas, TX 75226

Paula Lambert was born in Fort Worth, Texas. She attended universities in Virginia and Texas, and completed her graduate studies in Italian and Art History in Perugia, Italy. In 1973 she married Jim Lambert, a landscape architect, and moved back to Texas.

Five years in Perugia gave her a deep appreciation for Italian foods, especially their fresh cheeses, none of which were available at the time in the southwestern United States.

Determined to duplicate the subtleties of fine cheese production, Paula returned to Italy to work and study in cheese factories. The Mozzarella Company opened in 1982 under the guidance of an Italian cheese professor.

During the first few years her fledging company lost money while producing small quantities of mozzarella and ricotta cheeses. A second trip to Italy for additional training enabled Paula to expand her production to include other cheeses such as the mascarpone and caciottas - as well as to perfect her cheesemaking skills. Soon thereafter she began producing goats' milk cheeses.

Expanding her selection and adding various herbs and chilies indigenous to the southwest gave her cheeses a unique character. Recognition and widespread distribution followed - and so did profits!

The company now produces over two thousand pounds of cheese weekly and has won numerous awards for excellence from competitions in the American Cheese Society.

Mozzarella Co. a big cheese in awards

FOR THE THIRD year in a row, the Mozzarella Co. of Dallas garnered top awards for its cheeses at the 1987 annual judging of the American Cheese Society held in Boston recently.

The tiny company, located on Elm Street in downtown Dallas, was awarded six ribbons, including a blue ribbon for its Goat Caciotta laced with Mexican marigold mint. This semisoft cheese, made from goat's milk and aged two to six months, has a robust flavor enhanced by the anise flavor of the mint.

Also singled out by the judges were Caciotta, Texas Basil Cheese and Ancho Chile Cheese, all made from cow's milk, as well as Montasio, a firm goat's milk cheese.

The company's fresh mozzarella, made daily in the traditional Italian manner from fresh milk, won a ribbon for the third consecutive year.

The Mozzarella Co. was founded five years ago by Paula Lambert, who remains the Southwest's only specialty cheesemaker.

Lambert says her cheeses "reflect modern culinary trends as well as our Southwest region."

Lambert's cheeses are served at top-rated gourmet restaurants in Dallas and across the country.

All of the cheeses can be purchased at the factory at 2944 Elm St., or at various Dallas specialty food stores. Gift baskets also are available.

For more information, call 741-4072.

A ton of cheese

After spending five years in Italy, Paula Lambert returned to the United States to find that fresh, Italian-style cheese was scarce here. She decided to capitalize on its absence and make cheese in Dallas.

In 1982, she went back to Italy to work and study in cheese factories. After learning many of the subtleties of fine cheese production, Lambert returned and opened the Mozzarella Co. under the guidance of a cheese expert from Italy.

Several years of barely making a profit at making mozzarella and ricotta cheese convinced Lambert to return to Italy for more training. During the second trip, she expanded her knowledge to include several more types of cheese.

Expanding her selection and adding ingredients from the Southwest paid off for the cheese company. It now produces about a ton of cheese a week and has won several ribbons from the American Cheese Society, which holds domestic specialty-cheese competitions.



Paula Lambert

The majority of her business comes from restaurants and hotels but she also sells to gourmet shops and specialty grocery stores. Few sales come from walk-in customers, she says.

About 50 percent of her business is outside of Dallas and requires shipping the cheese via overnight mail. Her six employees try to keep the business growing gradually and stay ahead of the competition — European cheese makers.

The six-figure gross revenue business has grown gradually over the years, Lambert says, and sticking to word-of-mouth advertising keeps it that way.

— Floyd Whaley

From the Dallas Times Herald, August 17 & 19, 1987.

LISTERIA - SAFETY AND GOVERNMENT REGULATION OF CHEESE

By Dr. Catherine W. Donnelly,
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ABSTRACT

A major outbreak of listeriosis which occurred in California in 1985 was linked to the consumption of contaminated Mexican-style cheese. Subsequent surveys analyzing the incidence of Listeria in cheese have revealed the presence of Listeria not only in finished ready-for-sale cheeses, albeit at low incidence rates, but more importantly the widespread presence of Listeria within cheese processing environments. Numerous studies have confirmed the ability of Listeria to grow and multiply in cheeses once introduced as a contaminant. Recent studies which have explored the heat resistance of Listeria suggest that failure to properly pasteurize milk may lead to the survival of Listeria in milk destined for processing into aged cheddar cheese, for example. Are current governmental regulations adequate to assure the safety of cheese? Are existing methods sufficient in their ability to detect the presence of Listeria in processed cheeses? Are these concerns actually warranted? This paper will examine these and other related issues pertinent to safety and governmental regulation of cheese.

INTRODUCTION

A review of basic concerns related to Listeria which impact governmental regulation of cheese is warranted due to the adverse economic and health consequences associated with the presence of Listeria. This will be accomplished through a review of documented listeriosis outbreaks which have been linked to cheese. Secondly, the growth potential of Listeria in cheese and the elimination of this organism via pasteurization will be reviewed. Sources of Listeria in processing environments will be examined, and current detection procedures and their concomitant limitations will be examined. Through this analysis, areas of concern pertaining to governmental regulation of cheese will be revealed.

The first documented link between cheese consumption and an outbreak of listeriosis was reported in California in 1985. This well publicized outbreak received much attention from the news media. In this outbreak, consumption of

Jalisco-brand Mexican style cheese was linked with onset of 142 cases of listeriosis. The outbreak occurred during the period January 1-August 15, 1985. Ninety-three (65.5%) of the cases were recorded in pregnant women or their offspring. Forty-nine cases were recorded in nonpregnant adults. In all, 48 deaths resulted from this outbreak. The outbreak strain was identified as a serotype 4b L. monocytogenes (Linnan, et al. 1988).

A second outbreak of foodborne listeriosis linked to cheese was reported by Bille (1988). The outbreak occurred in Vaud, Switzerland and was linked to consumption of Vacherin Mont D'or cheese. A total of 122 cases occurring during the period 1983-1987 were reported. The normal endemic rate of listeriosis in Switzerland is 5-10 cases per million population. During the outbreak period, the rate of listeriosis rose to 50 cases/million population. Sixteen cases were reported in 1983, 24 cases in 1984, 13 cases in 1985, 28 cases in 1986 and 41 cases in 1987. Associated with these cases was a 28% mortality rate. In reviewing this outbreak, it is interesting to note the length of time over which this outbreak took place. It is also interesting to question whether the U.S. Food and Drug Administration or Centers for Disease Control would have issued a quicker response to the problem.

Even though very few outbreaks of human illness and listeriosis specifically have been linked with consumption of contaminated cheese, cheeses are nevertheless well documented sources of Listeria. In 1985, the U.S. Food and Drug Administration conducted a domestic soft and semi-soft cheese survey in order to estimate incidence rates of Listeria in domestic soft and semi-soft cheeses. Of 542 samples tested, 10 were found to be positive for L. monocytogenes. In addition to the presence of Listeria, contaminated cheeses were found to have other microbiological deficiencies, such as containing high levels of Escherichia coli per gram. The FDA also conducted an imported soft cheese survey between February and October of 1986. In this survey, 1699 samples were analyzed for the presence of Listeria. Sixty-nine samples were found to be positive for L. monocytogenes.

Soft and semi-soft cheeses possess characteristics which favor the proliferation of Listeria, thereby explaining the incidence rates revealed in random surveys. Studies conducted by Ryser and Marth (1987) at the University of Wisconsin document the tremendous growth potential of L. monocytogenes in soft-ripened cheeses such as Camembert. Low population levels ($2.6-2.9 \log_{10}$) of Listeria intentionally added during the cheesemaking process show a rapid increase during ripening, reaching maximum population levels of $1 \times 10^6-5 \times 10^7$ CFU/g. The pH conditions in Camembert average approximately 6.25, favoring the rapid growth and multiplication of Listeria. Unlike the rapid growth observed for Listeria in soft ripened cheeses, a rapid decline in viable populations is observed when Listeria is added during the manufacture of Cheddar cheese. Although populations show a rapid decline, detectable levels still persist up until 300 days of ripening at 4°C. The current U.S. regulation calls for ripening of cheddar cheese manufactured from raw milk for at least 60 days at 1.7°C (35°F). This issue is currently of concern as this process will not assure a safe product with respect to Listeria.

It is now generally accepted that Listeria is unable to survive proper milk pasteurization. The largest threat to the safety of milk and dairy products, including cheese, is due to post-pasteurization contamination from the manufacturing environment. Numerous surveys have been recently designed to identify sources of pathogenic contamination within the cheese and dairy processing environment. One such survey was conducted by our laboratory at the University of Vermont (Klausner et al., 1989). In this survey, each of Vermont's 34 dairy processing plants were examined for contamination due to the pathogens Listeria and Yersinia. It was the objective of this survey to identify sources of Listeria and Yersinia in Vermont dairy plants. Secondly, results were statistically analyzed to determine whether contamination was most frequently linked to the type of plant being analyzed; the conditions existing within plants; and specific sites within plants. Of 34 plants analyzed, samples taken from cheese plants accounted for the majority of samples (168 sites; 46.5%) analyzed in our survey followed by fluid plants which accounted for 115 sites (31.9% of samples) and finally non-cheese plants, consisting of whey, infant formula, ice cream and yogurt plants, accounting for 78 sites or 21.6% of samples.

When the distribution of Yersinia and Listeria positive samples were examined as a function of plant type, fluid plants accounted for 66.7% of the Listeria positive samples and 51.1% of the Yersinia positive samples. Non-cheese plants accounted for 23.8% of the Listeria-positive samples and 12.8% of the Yersinia positive samples. Cheese plants, from which the majority of samples were taken, accounted for only 9.5% of Listeria positive samples, illustrating that sanitation measures which eliminate bacteriophage are also effective in eliminating Listeria contamination.

When analyzed statistically, our results indicate that fluid plants showed a significantly greater prevalence for Listeria and Yersinia than did the other plants. For Listeria, cheese plants showed a significantly lower prevalence than did non-cheese plants, but cheese plants did not show any significant difference for Yersinia. Specific areas of concern revealed in our survey included milk cases and casewashers, beds of paper fillers and whey drainage pans of cheese presses. These latter areas are of specific concern due to the close proximity of the sample site with finished product. Clearly, these are areas for which aggressive monitoring and sanitation are a necessity. In conclusion, our survey revealed that contamination of wet areas was significantly greater than dry areas. Further, incidence of Listeria was significantly greater for floors than non-product contact surfaces and importantly, sanitizing floor mats and foot baths can be a source of contamination when used incorrectly.

In order for the cheese manufacturer to assure the absence of Listeria in the manufacturing environment, a testing method which is both rapid and sensitive is of necessity. Currently, there are a number of current concerns regarding rapid detection schema for Listeria. The first of these concerns relates to the lower limits of detectability for Listeria. All rapid methods developed to date

require a minimum of 5×10^4 - 1×10^5 Listeria/ml for detection. Failure of the utilized enrichment medium to support growth of a Listeria population to this level will result in the recording of a negative sample. Secondly, most, if not all, commercially available rapid detection procedures reveal the presumptive presence of Listeria species, and not the pathogenic L. monocytogenes. Confirmation of L. monocytogenes requires several days beyond initial presumptive results. Thirdly, no detection scheme developed to date, with the exception of cold enrichment, allows the detection of injured Listeria. Finally, most rapid methods developed for use to date are expensive, limiting their widespread employment.

Currently, the U.S. Food and Drug Administration imposes a zero tolerance for Listeria in dairy products. What exactly does zero tolerance mean or imply? Related to cheese, analyzed product must test negative for Listeria. However, because of limitations in detection methods, cheeses which contain low levels of Listeria could test negative when they are actually positive for Listeria. Work conducted by Dr. Michael P. Doyle and colleagues at the University of Wisconsin confirms this. A lot consisting of 90 soft ripened cheese samples was tested for presence of Listeria (Doyle and Schoeni, 1987). Using a combination of three methods, 41 of 90 (46%) of cheese samples actually tested positive for Listeria. When performance of each of three methods was examined individually, the cold enrichment technique detected only 21 positive samples, followed by the FDA procedure which detected only 16 positive samples and finally the SEP procedure of Doyle and Schoeni (1986) which detected only 13 positive samples. Subsequent work by the U.S. Food and Drug Administration revealed that the FDA method recorded a positive result only when the population of Listeria in soft cheese was 1100 Listeria/g or greater. Thus, existing methods must be improved in order to make the policy of zero tolerance meaningful and applicable to a variety of food commodities.

The fact that Listeria can be injured by a variety of processing treatments which include heating, freezing and exposure to acid poses a major public health threat. Clearly, Listeria in cheese would be subjected at different times to potentially all of these treatments. When Listeria are heat-injured, for instance, they are no longer able to grow in the presence of compounds such as potassium tellurite, phenyl ethanol and acriflavin. These constituents are found as ingredients in most media selective for Listeria. Thus, injured Listeria cultivated in the presence of these constituents would escape detection. A future challenge to food microbiologists is the development of repair/detection procedures which allow for the recovery of healthy as well as injured Listeria populations from processed food products and food manufacturing environments.

Although the presence of Listeria in all food products is potentially significant, relative significance is dictated by inherent parameters possessed by the food commodity in question. For instance, in Brie cheese, conditions exist which support the growth and proliferation of high levels of Listeria. Likewise, in ready-to-eat meats, initial low level Listeria contamination could result in growth of high levels of Listeria due to the absence of inhibitors or growth-suppressive

conditions in most ready-to-eat meats. The case is much different in a product such as ice cream, however. Provided that ice cream remains and is consumed in a frozen state, it is difficult to envision that sufficient temperature abuse could occur to permit the multiplication of high levels of Listeria. Secondly, Listeria in ice cream most likely exist in a freeze-injured state. Previous studies from our laboratory (O'Brien and Donnelly, 1988) show that freeze-injured Listeria are sensitive to physiological concentrations of bile. Thus, it is unlikely that freeze-injured Listeria would survive passage and implantation in the gastrointestinal tract of humans following ingestion.

As seen previously, low levels of Listeria initially present in Cheddar cheese would not increase, but would in fact decrease over time due to the unfavorable growth environment which exists in Cheddar cheese. Similarly, Listeria populations initially present in a product such as yogurt show rapid declines due to the adverse, low pH environment which supports neither growth nor proliferation of Listeria. Thus, when assessing significance of Listeria in food products, it is clear that numerous factors must be taken into consideration, as must the reliability of employed detection procedures to assess presence of the organism.

Prevention of contamination of cheeses and other food commodities is necessary in order to retain consumer confidence in the unequivocal safety of processed foods. Meeting this challenge should be a primary goal of all cheese manufacturers. However, before this goal is met, it is clear that major improvements in the testing and risk assessment, which impacts governmental regulation of cheese, is in order.

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