FLAVOR DEVELOPMENT AND MICROBIOLOGY OF SWISS CHEESE—A REVIEW

II. STARTERS, MANUFACTURING PROCESSES AND PROCEDURES

T. LANGSRUD AND G. W. REINBOLD
Department of Food Technology
Iowa State University, Ames, Iowa 50010
(Received for publication January 31, 1973)

Abstract
This paper, the second of four, discusses the starters used in Swiss cheese manufacture. Propionibacteria are essential for development of characteristic Swiss cheese flavor; high-temperature lactic acid and Streptococcus thermophilus are necessary for control of body, texture, acidity, moisture, and other related factors.

The effects of pH, salt, temperature, metals, and media composition on growth of propionibacteria are first discussed. Since growth inhibition and stimulation of propionibacteria are closely related to Swiss cheese quality, these factors also are considered. Maintenance of cultures in the plant is mentioned with some bacteria are discussed. Slow-acid production caused by metallic inhibitors, antibiotics, and bacterialpoles also is reviewed.

Relationships between manufacturing processes and procedures and Swiss cheese quality are discussed. In this portion of the review, we are primarily concerned with growth and changes of bacterial flora in the milk and curd from the vat to the warm room. This includes the influence of such factors as preripening, cutting, cooking, pressing, block size, brining, and prestorage before the warm room.

Normal ripening of Swiss cheese involves the combined action of various bacteria to produce resultant texture, flavor, and body changes. Although a wealth of data concerning the major (lactate) fermentation has been accumulated, there is still much to be learned. Von Freudenreich and Orla-Jensen (123) isolated propionic-acid producing bacteria from Swiss cheese in 1906 and assumed that these bacteria were important for Swiss cheese flavor. It took until 1921, however, for Sherman (393) to prove that propionic-acid bacteria were essential for characteristic flavor development in Swiss cheese. It was later shown that Streptococcus thermophilus and Lactobacillus species assisted in the development of high-quality Swiss cheese and limited abnormal fermentations (141, 394). In this section we will discuss Swiss cheese starters in some detail.

Starters for Swiss Cheese

Propionibacteria

Classification. Propionibacteria are nonmotile, non-sporoforming, gram positive, irregularly shaped short rods, and are generally catalase-positive. They ferment lactic acid, carbohydrates, and polyhydroxy alcohols to propionic and acetic acids and carbon dioxide. Their growth is slow, and they prefer anaerobic conditions. Nutritional requirements are complex, and they grow best in a medium containing yeast extract with tryptone as the nitrogen source plus lactate or a simple carbohydrate as an energy source (8). For a thorough discussion of the growth, metabolism, and miscellaneous metabolic activities of the propionibacteria, see the recent review by Hettinga and Reinbold (156, 157, 158).

In 1906 von Freudenreich and Orla-Jensen (123), isolated three species, Bacterium acidi propionici a (Propionibacterium freudenreichii), Bacterium acidi propionici b (Propionibacterium jensenii), and Bacillus acidi propionici (Propionibacterium pentosaceum) from Emmentaler and Limburger cheese. Propionibacteria were later isolated from other cheeses, but only in Swiss-type cheeses were they ever found in the majority of all isolates (8). Propionibacterium were isolated from milk, soil, and silage by Hitchner (167) and also have been found in fecal matter from cows and pigs (93).

The genus Propionibacterium was first proposed in 1909 by Orla-Jensen (312). It was only after van Niel’s (290) classical work The Propionic Acid Bacteria, published in 1928, that propionibacteria were accepted as a genus in the family Lactobacillaceae in the 1930 3rd edition of Bergey’s Manual of Determinative Bacteriology (29). Only eight species were recognized at first on a few morphological and biochemical differences. Later work led to recognition of three new species which were included in the 1957 7th edition of Bergey’s Manual (48).

Primary emphasis in classification of propionibac-
teria is placed on carbohydrate metabolism, although Kurmann (229, 230) found his data on carbohydrate fermentation were inconsistent with earlier data (48). In 1963, Antila and Gyllenberg (11) made a study of the interrelationships between different species according to a taxonomic key proposed by Janocek (178). They concluded that species consolidation was necessary. Numerical taxonomy was used by Malik et al. (264) to re-evaluate speciation of Propionibacterium. Their results suggested that P. shermanii and P. freudenreichii ought to be consolidated, relegating P. shermanii to varietal basis. Propionibacterium arabinosum and P. pentosaceum also were shown to be closely related. The fermentation of xylose and rhamnose by P. pentosaceum, and the development of orange-yellow pigment by P. arabinosum were not constant characteristics. Close relationship also was found between Propionibacterium rubrum, Propionibacterium peterssonii, and P. jensenii. All these results seem to indicate re-evaluation of the genus Propionibacterium is necessary and that the number of species should be reduced.

A recent development has been a suggestion for reclassification of Corynebacterium acnes to Propionibacterium acnes (274, 278). This suggestion was initiated because of similarities between C. acnes and propionibacteria (274, 278) in the production of propionic and acetic acids from lactate and in serological analyses. Fatty acid composition of C. acnes and propionibacteria also was similar. Dissimilarities, however, do exist; Werner (459) noted that there is a difference in DNA analysis and that C. acnes produces lecinthinase which is not produced by the propionibacteria. These dissimilarities support the present separation of this species from the propionibacteria.

Effect of pH on growth. The optimum pH for growth of propionibacteria lies between pH 6.0 and 7.0 with a pH maximum at 8.5 and minimum at 4.8 (216).

In a detailed study of P. shermanii, Tiitttsler and Sanders (431), found that the initial pH of the medium within the range pH 4.7 to 5.5 greatly influenced growth. The critical pH was about pH 5.0, but this value varied with the degree of anaerobiosis and the size of inoculum. The bacteria did not grow at pH values lower than the critical value. This sensitivity to pH emphasizes the importance of pH control during Swiss cheese production, because slight variations, as small as 0.1 to 0.2 pH unit, in acidity may determine if the desired fermentation will proceed.

Effect of salt. Propionibacteria show low tolerance to salt. Experiments by Orla-Jensen (319) showed that concentrations of sodium chloride as low as 0.5% were enough to reduce growth in a substrate containing calcium lactate. A possible change from sodium chloride to calcium chloride was the suggested cause of growth reduction. Later experiments with sodium lactate-containing media showed that inclusion of 3% sodium chloride was necessary to reduce the growth rate (8). Rollman and Sjöström (363) used a normal and a fast lactate-fermenting strain of propionibacteria in two media, one medium being adjusted to pH 7 and the other to pH 5.2. The normal strain tolerated sodium chloride better and formed more gas at pH 5.2 than at pH 7. With a sodium chloride concentration above 3% the fermentation was impeded at pH 5.2 for the fast fermenting strain, but at pH 7, 6% sodium chloride was necessary to retard fermentation. This demonstrates a combination effect between pH and salt concentration. Exposure of strains of P. shermanii and P. freudenreichii to 6 to 8% sodium chloride for 100 hr was very destructive (179).

With a sodium nitrate concentration of 80 g/l in a medium at pH 7, propionibacteria are able to grow (363). At pH 5.2, a normal strain grew at all concentrations from 10 to 80 g/l, while a fast lactate-fermenting strain could not grow at sodium nitrate concentrations greater than 10 g/l.

Effect of temperature on growth. The optimum growth temperature of propionibacteria is 30 C (77 F) (8). In a review article, Krane (216) states that Orla-Jensen found propionibacteria grow between 15 and 40 C (59 and 104 F), but Foster et al. (116) reported growth between 15 and 45 C (59 to 113 F). Growth at 45 C (113 F) was extremely slow. Antila (8) mentions that growth occurs as low as 12 to 13 C (53.6 to 55.4 F) and the ability for growth at 12 C (53.6 F) was used by Antila and Gyllenberg (11) as one of their taxonomic determinative tests. In a detailed study, Park et al. (319) found that 16 of 33 strains of propionibacteria tested grew at 7.2 C (45 F), while 31 grew at 12.8 C (55 F). Scanty growth was found at 2.8 C (37 F). Another significant growth feature noted was the greater production of CO2 at 12.8 C (55 F) rather than 27.2 C (81 F). There was little difference in CO2 production after 4 weeks, but differences were significant after 8 weeks.

The effect of storage temperature on the activity of propionibacteria was studied by Ritter (353). At 5 C (41 F) the propionibacteria retained activity and high viability up to 8 weeks; at 25 C (77 F) biochemical activity was higher, but the total number of bacteria decreased, possibly because of inhibition by the propionic and acetic acids produced. The keeping time at 25 C (77 F) was about 2 weeks.

Antila (8) reports that Thöni and Allemann found that propionibacteria were killed at 70 C (158 F) in 15 min, but destruction was negligible at 60 C (140
production as compared with the negative control. Rollman and Sjöström (363) found that all propionibacteria survived 70°C (158°F) for 10 to 20 sec. In their study of the heat resistance of propionibacteria, Thomé and Lindgren (429) found strains which survived 76°C (168.8°F) for 10 sec but never 20 sec. Nearly half of the strains survived 72°C (161.6°F) for 20 sec. This indicates that propionibacteria found in cheese made from milk pasteurized at 72°C (161.6°F) or lower could originate from the raw milk. Ten-day-old cultures also are usually more heat resistant than 2-day-old cultures (94, 429). The media also have an influence on survival rate. In milk the bacteria can tolerate about 7°C (12.6°F) higher heat treatment than in broth. So, propionibacteria are relatively heat-resistant and can easily survive the cook temperature (ca. 53°C or 127.4°F) of Swiss cheese.

**Effect of metals on growth.** Originally, Swiss cheese was made in copper kettles, which suggested the necessity of studying the effect of copper on propionibacteria since Swiss cheese is now made in the U.S. almost exclusively in stainless steel vats (345). Following this idea, Mueller et al. (280) found that copper in amounts from 2 to 8 mg/l of lactate broth increased the lag phase and decreased the growth rate of three strains of *P. shermanii*. At the same time, a lag in volatile acid and CO₂ production was observed. After incubation for 13 days, however, cell mass, volatile acid, and CO₂ production were about equal for the various tested levels of copper. Maurer and Reinbold (unpublished results), using copper levels from 2 to 16 mg/l of broth, found that 16 mg/l strongly inhibited two strains of *P. shermanii*: lower concentrations caused slight growth inhibition and a reduced level of CO₂ production as compared with the negative control. One strain of *P. shermanii* showed no inhibition at 16 mg/l. At pH 5.4, CO₂ production was lower than at pH 7 with a copper concentration of 16 mg/l. High levels of copper (18 mg/kg) deleteriously influence the ripening of cheese (280), but propionic acid development and eye formation are normal in Swiss cheeses containing 5 mg/kg of copper (470). *Propionibacterium shermanii* formed pigments when grown in the presence of copper-oxidized milk-fat at suboptimal growth conditions (387).

Metallic iron and its chlorides promoted propionic acid fermentation with *P. peterssonii* (200). Gas production by *P. shermanii* was retarded in a medium containing only small quantities of iron, but, eventually, the amount of CO₂ equalled that in the control medium (340).

Magnesium and manganese accelerate decarboxylation of succinate and growth of *P. arabinosum* (216). A manganese concentration of 0.01% also was found to change the propionic acid:acetic acid ratio in favor of propionic acid without affecting growth (452). Concentrations of cobalt up to 3 mg/l of broth stimulated the growth of *P. freudenreichii*, but amounts of 4 to 5 mg/l decreased growth (374).

Since some trace elements in the diet of the cow are readily transferred into the milk, these observations may help to explain the occasional variations in growth rate, CO₂, and flavor development by propionibacteria observed among lots of cheese, seasons, and manufacturing plants.

**Effect of media on growth.** For a more detailed study of the influence of media composition on growth of propionibacteria, the reader is referred to a review article by Hettinga and Reinbold (156). The nutritional requirements of propionibacteria are complex. Growth is best in a complex medium containing yeast extract; a nitrogen source, peptone or some other casein hydrolysate; and an energy source, such as lactate or a carbohydrate. Propionibacteria usually grow rather slowly in milk. Van Niel (290) found only two strains that grew fairly rapidly in milk. Adding amonium chloride, peptone, or casein-amino acids to the milk increased the growth rate (162). Autoclaving the carbohydrate together with other media constituents enhanced propionibacterial growth (109, 110, 111).

**Inhibition of propionibacteria.** Most of the work on the action of antibiotics against propionibacteria has been done in conjunction with the butyric-acid fermentation of Swiss cheese. Nisin, an antibiotic produced by strains of *S. lactis*, was first shown to have little influence on eye formation and ripening of Swiss cheese (216). Later experiments by Galesloot (128) showed that nisin hindered growth of propionibacteria, as well as growth of anaerobic sporeformers. Winkler and Fröhlich (464) found that the chief ripening microorganisms in Emmental cheese were inhibited by nisin. *Lactobacillus helveticus* and propionibacteria were especially sensitive. The inconsistent results obtained by different workers may probably be explained on the basis of use of different starter strains, as well as procedures and materials.

The growth of *P. shermanii* (116) is totally inhibited by use of 0.1 unit penicillin and 5 μg streptomycin per ml of milk, and these antibiotics completely inhibit the normal ripening of Swiss cheese (8). Tylosin inhibited eight strains of *Propionibacterium*. This inhibition was greatly influenced by the pH of the medium, inoculum size, and intrinsic strain specificity (43). A recent study of the activity of 40 antibiotics and antimicrobial agents on 30 strains of *Propionibacterium* has been conducted by Reddy and Reinbold. Their, as yet, unpublished results show
that these bacteria as a group are relatively resistant to cloxacillin, nafcillin, oxacillin, and nalidixic acid in addition to numerous sulfonamides.

The metabolic products of some other bacterial genera and species can adversely influence the growth of propionibacteria. For example, Winkler (462) found that Pseudomonas aeruginosa inhibited 16 of 17 strains of propionibacteria. Strains of Bacillus subtilis, Bacillus mesentericus, and Bacillus mycoides also reduced growth of strains of Propionibacterium. A heat labile and a heat stable compound were found in Lactobacillus lactis, L. helveticus, and Lactobacillus acidophilus which inhibited the growth of propionibacteria (216).

In a study by Vedamuthu et al. (441, 442) 6 of 8 species of Propionibacterium were inhibited by a filtrate from Swiss cheese whey; the two unaffected species were the pigmented P. rubrum and Propionibacterium thoenii. Heat treatment produced inactivation of the inhibitory principle; partial inhibition occurred on soft agar lawns at a 10-fold dilution, and complete inhibition was shown in broth cultures containing 2% undiluted whey. An acid solution adjusted to or below the pH of the whey did not inhibit susceptible strains. Later, it was shown that the inhibitor was an immunoglobulin (444). We have since demonstrated that P. rubrum and P. thoenii also are inhibited by this immunoglobulin but to a lesser extent than the unpigmented species.

Some of the metabolites in the propionic-acid fermentation display inhibitory capabilities. Lactate in high concentration was shown by Antila and Hiettaranta (13) to inhibit growth of propionic-acid bacteria in certain media. Sodium lactate proved more inhibitory in smaller concentrations than calcium lactate. The inhibitory effect did not depend on the presence of undissociated lactic acid. It was also found that levorotatory lactic acid was fermented slower than the dextorotatory form. Acetic and propionic acids also inhibited the growth of propionibacteria. According to Antila (8), acetate, in certain instances, stimulates growth but, conversely, may lead to a strong inhibition at rather low concentrations. Growth of propionibacteria was inhibited in various degrees in media containing 4% sodium or calcium propionate. Sodium propionate was more inhibitory than calcium propionate and inhibition was more pronounced at pH 5.5 than at pH 6.6 (12). Propionic acid is a more effective inhibitor than acetic acid and inhibition is not additive because, with both acids present, the acid at the highest relative concentration is the growth-controlling factor (289).

**Associative effect of bacteria on growth of Propionibacterium.** In 1921 Sherman and Shaw (395) showed that growth of propionibacteria together with S. lactis or L. casei increased production of propionic acid six to eight times in comparison to that in media fermented only with propionibacteria. The number of Propionibacterium in the medium increased with addition of L. helveticus (8). Lactobacillus casei culture added to vat milk improved the quality of Emmentaler cheese with respect to both flavor and eye formation (329).

Klimovskii and Alekseeva (206) investigated the effect of lactic-acid bacteria on the growth of P. shermanii at pH 5.2 to 5.5 by addition of sodium caseinate in sterilized skim milk. Milk acidity was maintained. Propionibacteria (4 X 10⁴/ml initially) grew slowly during lactose breakdown, then increased rapidly in media containing L. helveticus to 7 X 10⁶/ml, but in media containing L. casei remained at 7 X 10⁴/ml. No differences in the development of propionibacteria were noted when cultivated in dilute cultures of different lactic-acid bacteria (streptococci ± L. helveticus or L. casei) that had been killed by autoclaving. When pH 7 was maintained during growth of propionibacteria, their numbers rose to 2.3 to 2.8 X 10⁶/ml after 10 days; at pH 5.4, they numbered about 1.2 to 1.7 X 10⁶/ml. Cheeses were made by Nieuwenhof et al. (292) using a mixed culture of P. freudenreichii and P. pentosaceum, and with the same culture containing L. casei. After ripening for 3 weeks the cheese containing L. casei had developed larger eyes than the other cheese. The number of propionibacteria was greater, as was the amount of CO₂ and acetic and propionic acid, thereby showing stimulation of propionibacteria by L. casei.

Gas production by P. shermanii was markedly stimulated in skim milk previously fermented by Lactobacillus bulgaricus, L. helveticus, and L. lactis while S. thermophilus, S. lactis, and Streptococcus faecalis had little effect (169, 170). Gas production was shown to be slower when D(-) lactic acid was fermented. Inoculation of lactobacilli and propionibacteria together markedly stimulated gas production. The stimulatory factors were partly dialyzable, heat- and acid-stable substances, possibly of peptide nature.

In Switzerland, P. Ritter and coworkers have studied the symbiotic effect of micrococci on the growth of propionibacteria (335, 336). By addition of a milk culture of Micrococcus before renneting these results occurred: the experimental cheeses reached their maximum pH 2 weeks before the controls; CO₂ production increased about 20%; and the experimental cheeses contained higher numbers of propionibacteria than the controls. The authors suggested that this symbiotic activation of propionibacteria could cause a “secondary fermentation” defect in Swiss cheese.
It also is conceivable that this, or similar stimulation of CO₂ production, could be responsible for instances of the split defect. (See reference 345 for a complete discussion of this defect).

**Lactic-acid bacteria**

*Effect of cultures on quality.* In earlier times, manufacturers of Swiss cheese prepared their own rennet from calves' stomachs. The resulting product contained both coagulating enzyme and acid-forming bacteria which served as starter (116). As the knowledge of cheese bacteriology increased, it became clearer that lactic-acid bacteria play a principal role in cheese ripening since normal ripening only took place when the lactic-acid bacteria were used alone or in combination with other microorganisms (18). Doane (88) reported that L. bulgaricus could be used to repress undesirable fermentations in cheese. Gas production especially was repressed but without harmful effect on eye formation and L. bulgaricus did not interfere with flavor and texture characteristics. *Lactobacillus bulgaricus* is the most commonly used high-temperature lactobacillus in the U. S., but in Europe *L. helveticus* is more generally used. *Lactobacillus casei* has been shown to control overswelling of Swiss cheese at times (394), but its use was not always successful. Before 1930, *S. thermophilus* usually was added to cheese milk either by contamination or by use of mixed rennet-whey culture. The quality of Swiss cheese made with either *L. casei* alone or *L. casei* and *S. thermophilus* together was once studied by Frazier et al. (118). *Streptococcus thermophilus* nearly always improved the quality of the cheese, especially improving eye-formation, texture, and flavor. If either was used alone, however, more glass and checking were present which possibly may be because *S. thermophilus* stimulates gas production by *Clostridium tyrobutyricum* (109).

For successful use in Swiss cheese production, a starter organism must be heat resistant and able to grow at relatively high temperatures because it must survive the cooking process and then continue to grow during pressing. Iowa Swiss-type cheese originally used *S. lactis* as the lactic acid-producing starter and, therefore, used a lower cooking temperature [37.8 C (100 F)] (136). Recent changes in the make procedure include the use of high-temperature starters (345).

Comparative studies were made between starters (*S. thermophilus* and *L. helveticus*)—either cultured in sterilized milk and added in amounts of 0.2% to the cheese milk or as concentrated frozen suspensions [2-5 × 10⁷ organisms/g at -30 C (-22 F)] added at levels of 0.001% for *S. thermophilus* and 0.01% for *L. helveticus* for Gruyère cheese. Acid production during pressing was faster and more uniform when concentrated starters were used. These cheeses scored better than cheeses made with conventional starter culture because of more uniform eye distribution and better flavor (365).

Usually, heat-resistant organisms are used for Swiss cheese manufacture. Demeter et al. (81) used a culture of *L. acidophilus* plus *S. lactis* var. *taetted* instead of the usual Swiss cheese starters. The rate of acid development in vat and in press varied slightly from the control, but appearance, texture, and flavor of the cheese were excellent. The cheese body was of smoother texture than is usual in Swiss cheese made from pasteurized milk. Olsanský et al. (308) used 0.25 to 0.50% of “cream culture,” 0.061 to 0.12% of “Emmentaler” culture, 0.03 to 0.06% of *L. casei* culture, and 1.0 to 2.5 ml of propionic culture/1000 liters for Swiss cheese manufacture. These cultures used in the given amounts ensured correct lactic and propionic acid fermentation and protein breakdown. Influence on cheese quality by *L. casei* when added to a mixed starter culture of *S. thermophilus* and *L. helveticus* was studied by Bijok and Domańska (33, 34). The two cultures were mixed in equal amounts and a total of 0.1 to 0.2% was added to the cheese milk. These workers stated that addition of *L. casei* was necessary, in addition to the normal “Emmental” culture and propionic-acid bacteria, to produce Swiss cheese with a typical sweet, walnut taste.

Bijok (30) isolated "new" strains of *S. thermophilus* and *L. helveticus* from whey used as starter in Gruyère cheese production in Switzerland. Starter for a Swiss-type cheese was made by combining two parts of a culture of *L. casei* var. *thermophilus* with one part of a 1:1 mixture of cultures of *S. thermophilus* and *L. helveticus*. Cheeses produced using this starter had few, small eyes and a sweet, nutty flavor. They were considered of better quality than the control cheeses made with normal cultures (31).

Throughout this discussion, propionibacteria, high-temperature lactobacilli, and *S. thermophilus* have received most attention. Many readers will be aware that the mesophilic streptococci *S. lactis* and *S. cremoris* are frequently used in Swiss cheese production. Since their use in conventional make procedures is truly optional, they have not been discussed to any extent. If information on their use is desired see the monograph by Reinbold (345).

Kurmann (234) found that *S. faecalis*, inoculated in low-count raw or pasteurized milk had a favorable effect on Swiss and Gruyère cheese by improving eye formation and body and flavor of the cheese. *Streptococcus faecalis* was antagonistic against the coliformes group and, therefore, suppressed early
blowing of cheese. The influence on flavor and body of cheese is due to inhibition of unwanted flora, activation of normal growth of lactic-acid bacteria and lactic-acid fermentation, and provision of necessary proteolytic enzymes.

Lactic starter cultures used in Swiss cheese production have an important effect on cheese quality. To ensure characteristic organoleptic properties for Swiss cheese the selection of a starter, according to Dilanyan (84), should be based on sufficient acid formation in cultures after 24 hr and 7 days; proteolytic activity; presence of acetoin and diacetyl in the coagulum, syneresis of the coagulum, and formation of free amino acids as related to their proportions in high-quality cheese.

**Growth and heat resistance of Swiss cheese starter organisms.** Mother cultures of *S. thermophilus* and lactobacilli usually are carried in sterile milk which is incubated at 37 C (98.6 F) until curdling of the milk occurs. Rapid cooling is then advisable. Transfer should be daily or at least three times per week.

Incubation temperature of the starter is important. *Lactobacillus helveticus* grown at 30 C (86 F) for 24 hr was shown to have definitely less activity in cheese than when grown at 37 C (98.6 F) (105). Peppler and Frazier (326) studied the effect of temperature and time on starter activity. Cultures of *S. thermophilus* and *L. helveticus* transferred at 37 C (98.6), 40 C (104), or 42 C (107.6 F) showed no harmful effect on activity with variation of the incubation period from 12 to 24 hr in bulk culture. Continuous incubation of *S. thermophilus* at 45 C (113 F) with transfer every 12 or 24 hr produced cultures with activities equivalent to that incubated at 37 C (98.6 F). *Lactobacillus helveticus* grew irregularly and poorly after transfers at 45 C (113 F) in bulk culture. After growth at 37 C (98.6 F), *L. helveticus* and *S. thermophilus* could be stored at 0 C (32) to 20 C (68 F) for at least 96 hr without reduction in activity. This shows that active starter cultures can be prepared with considerable latitude in incubation temperature and time (326). Some mixed cultures of *L. bulgaricus* and *S. thermophilus* may grow at temperatures as low as 15 C (59 F) (171).

As all cheesemakers know, the activity of a starter may decrease. To overcome this problem, Bijok and Dománska (32), who used a mixed culture of *L. helveticus, S. thermophilus,* and *L. casei* which had been incubated for 20 hr at 37 C (98.6 F) with a titratable acidity close to 70° SH, activated the culture by dilution (1:2) with sterilized skim milk and incubation at 36 C (96.8 F) for 2 hr. (SH or Soxhlet-Henkel units express acidity as the ml of 0.25 N sodium hydroxide required to neutralize 100 ml of milk or whey to the phenolphthalein endpoint). Swiss-type cheese made from activated starter had better body and flavor than cheeses made from unactivated starter but was inferior in eye pattern.

So-called “thermophile” starter cultures are most frequently subcultured in sterile milk. Heat treatment of milk used in the bulk culture also is quite rigorous. In Switzerland, where most Swiss cheese is made from raw milk, this may lead to a problem. *Streptococcus thermophilus* and lactobacilli adapted to sterilized milk will grow poorly in raw milk. *Streptococcus thermophilus* may be readapted by subculturing in raw milk but lactobacilli must grow jointly with *S. thermophilus* to be readapted (354).

Schwab (388) investigated the influence of rancid milk on high-temperature lactic starters. Milk lipase and bacterial lipases were used to produce rancidity. High-temperature starters, particularly *S. thermophilus,* were inhibited in milk containing free fatty acids. Lauric acid was most inhibitory, while myristic acid slightly stimulated growth of lactobacilli. Therefore, lipolytic fat breakdown may lead to shifting of the bacteriological equilibrium in mixed starter and growth in the vat milk if rancidity is present.

In a study of the heat resistance of 60 lactobacilli isolated from Swiss cheese, Bassett and Slatter (24) found that 50 of these strains were killed in 16 min at 65.7 C (150.3 F). Only four were killed at 71 C (159.8 F) for 18 sec, whereas 25 did not survive 74 C (165.2 F) for 18 sec.

Time and incubation temperature may affect activity and heat resistance of high-temperature starter bacteria. Cultures of *L. helveticus* carried at 37 C (98.6) and 40 C (104 F) were more active following a heat treatment of 60 C (140 F) for 30 min than were cultures carried at 30 C (86), 35 C (95), and 47 C (116.6 F) (105). After the first 12-hr transfer of *L. helveticus,* the cultures grown at 37 C (98.6 F) were more heat resistant than those grown at 40 C (104 F), but after numerous 12-hr transfers the 40 C (104 F) cultures were more resistant than the 37 C (98.6 F) culture. Cultures of *S. thermophilus* after numerous successive transfers made by the same methods as for *L. helveticus* showed greater heat resistance [60 C (140 F) for 30 min] when grown at 30 C (86), 35 C (95), and 37 C (98.6) than at 40 C (104) or 42 C (107.6 F).

In regard to the importance of incubation time, *L. helveticus* was far more heat resistant when grown at 37 C (98.6) and 40 C (104 F) for 12 to 16 hr than cultures grown at the same temperatures for 7 to 8 hr. No marked differences in activity following heat treatment of cultures of *S. thermophilus* grown at 37 C (98.6 F) for 6 to 16 hr were observed (105). The influence of storage time and temperature upon heat resistance of *L. helveticus* initially incubated...
at 37 C (98.6 F) for 12 hr was shown by storage at 12 C (53.6 F) resulting in cultures with more activity after heat treatment than in cultures stored at 4 (39.2) and 20 C (68 F). These cultures could be stored at 12 C (53.6 F) for 60 hr and would still show the same heat resistance as cultures grown at 37 C (98.6 F) for 12 hr. Streptococcus thermophilus strains showed essentially the same effects as L. helveticus, but were more heat resistant after storage at 4 (39.2) and 20 C (68 F) than L. helveticus (327).

Freeze-drying of high-temperature lactic-acid bacteria. Lactic-acid starters are usually transferred at least three times a week to keep them at their highest activity. For commercial preparation and storage of starters, methods such as freezing and freeze-drying have been used. Experiments have been performed to examine the consequences of freeze-drying on L. helveticus and S. thermophilus (133). These microorganisms best survived freeze-drying when they were propagated in 1% sterilized reconstituted skim milk with incubation at 40 C (104 F) for 12 to 18 hr. Cultures showed optimum activity on reconstitution when they had been freeze-dried in reconstituted dried skim milk containing 0.5% yeast extract. Lactobacillus helveticus was more sensitive to freeze-drying than S. thermophilus. Swiss cheese produced with pure or mixed freeze-dried cultures, in comparison with conventional liquid-starter cultures, showed no difference in quality grade. The only difference seen during manufacture was that the freeze-dried cultures produced slightly less acid (133, 134). Freeze-drying mixtures of L. bulgaricus and S. thermophilus disturbed the balance between these organisms. Peptides formed by the proteolytic activity of L. bulgaricus stimulated growth of S. thermophilus upon reactivation of freeze-dried cultures. When cultures were first separated from the growth medium and then freeze-dried in concentrated milk, thereby excluding the peptides and amino acids formed by the proteolytic activity of L. bulgaricus, species balance could be maintained (132). Jabbarit (173) found that, after freezing a mixed culture at -25 C (-13 F), the proportion of L. bulgaricus and S. thermophilus changed from 51 and 49% in fresh yogurt to 60 and 40%, respectively, in frozen yogurt. This ratio was less affected by freeze-drying. With a mixed culture of L. helveticus and S. thermophilus, changes in proportion of the two organisms were less marked than with L. bulgaricus and S. thermophilus. In this instance, a greater proportion of L. helveticus survived (174). The greater resistance of L. helveticus to freezing and freeze-drying is attributed to its physiological properties and the interaction between L. helveticus and S. thermophilus.

Symbiotic effects. Symbiotic growth of L. helveticus and S. thermophilus with the film yeast, Candida krusei, increased the activity of the starters after heating. The bacteria isolated from areas in the medium close to the film were more active after heat-treatment than organisms growing in the medium at lower levels more distant from the yeast (327). Even L. helveticus and S. thermophilus growing together without film yeast show a symbiotic relationship (132, 174), which, possibly, is why neither of these organisms used by itself can produce a high-quality Swiss cheese.

This symbiotic relationship can be of value in Swiss cheese making. When S. thermophilus was used as the sole starter in the cheese, the maximum amount of lactic acid produced was reached after 10 days (407). Only half the total amount of acid was present, however, as compared with the cheese made with the mixed culture in which maximum acid levels were reached in 25 days. Using only L. helveticus, maximum acid levels were higher and were reached later than in the control cheese with mixed culture (407).

There are many reports that lactobacilli stimulate growth of S. thermophilus. For example, Pette and Lolkema (332) showed that L. bulgaricus stimulated growth of S. thermophilus by liberation of essential amino acids from milk proteins, and, possibly, peptides. Accolas et al. (1) observed that high-temperature lactobacilli, L. bulgaricus, L. helveticus, and L. jugurti, as well as mesophilic lactic streptococci stimulated acid production by S. thermophilus in milk heated at 80 C (176 F) for 30 min and in autoclaved milk. Bautista et al. (26) identified these growth-promoting factors as the amino acids glycine and histidine. In a later study, Accolas et al. (1) found that three strains of S. thermophilus were stimulated by a mixture of leucine, valine, and histidine, and one of these strains also was stimulated by a mixture of leucine, valine, and isoleucine. This indicates that the stimulatory factors may be different for different strains of S. thermophilus. Streptococcus thermophilus grown in association with L. bulgaricus also produced more acid per cell than when grown alone (332). Peptides formed by L. bulgaricus were shown by Gavin (132) to stimulate growth of S. thermophilus.

Streptococcus lactis stimulates growth of L. casei (141). A stimulatory factor has been isolated from cell extracts of S. lactis by Brannen and Keenan (46), and was shown to be a small peptide with a molecular weight of 4,500 Daltons. Streptococcus faecalis also stimulates the growth of L. helveticus (234).

On the other hand, fewer studies have been performed on the reverse effect, the influence of S. thermophilus on high-temperature lactobacilli. Acco-
Las et al. (1) observed that \textit{L. lactis} and \textit{L. bulgaricus} are stimulated by certain strains of \textit{S. thermophilus} in milk heated to 80 C (176 F) for 30 min, but no growth enhancement occurred in autoclaved milk. The stimulatory effect of \textit{S. thermophilus} on \textit{L. helveticus} and \textit{L. jungurti} varies with the strains and, when it exists, is not very marked. Formic acid and adenine exerted a definite stimulatory influence on \textit{L. helveticus} and \textit{L. jungurti} was not very marked, and varied from strain to strain (1). \textit{Lactobacillus helveticus} and \textit{S. thermophilus} also stimulate acid production by some strains of mesophilic lactic streptococci (1).

\textit{Streptococcus lactis} and \textit{S. thermophilus} usually stimulate the growth of \textit{L. bulgaricus} and \textit{L. helveticus}, but Politi (334) studied inhibition of acid production by different strains of \textit{S. lactis} and \textit{S. thermophilus} on \textit{L. helveticus} and \textit{L. bulgaricus}. One normal strain of \textit{S. lactis}, as well as two antibiotic-producing strains, strongly inhibited \textit{L. helveticus} after incubation for 40 hr at 35 C (95 F), but only the antibiotic-producing strains showed inhibition after 16 hr at 45 C (113 F). Only the antibiotic-producing strains inhibited \textit{L. bulgaricus}, and at 45 C (113 F) the rate of inhibition was considerably decreased. Six strains of \textit{S. thermophilus} decreased acid production of \textit{L. helveticus} for 40 hr at 35 C (95 F), and for 16 and 24 hr at 40 C (104 F); no such reduction was observed on \textit{L. bulgaricus} at 40 C (104 F) and only two strains of \textit{S. thermophilus} were inhibitory at 35 C (95 F) (334).

Micrococci isolated from Swiss cheese, were shown by Ritter (352) to be stimulatory to one test-strain each of \textit{L. helveticus}, \textit{L. lactis}, and \textit{S. thermophilus} in artificial media. These organisms also were grown in sterile milk, and the micrococi stimulated the two lactobacilli but had no effect on \textit{S. thermophilus}. It may be that milk is a more favorable medium for \textit{S. thermophilus}. The micrococci tested included \textit{Micrococcus luteus}, \textit{Micrococcus freundreich}, \textit{Micrococcus candidus}, and \textit{Micrococcus conglomeraus}. Desmazeaud and Devoyod (82) isolated caseolytic micrococci strains from aseptically drawn milk. These organisms stimulated acid production by \textit{S. thermophilus} but not by lactobacilli. The isolated stimulatory substances were shown to be peptides (82). Stimulatory substances in the symbiotic growth of these starter organisms, therefore, usually seem to be peptides and amino acids produced by proteolytic breakdown of casein.

In a later study, Nath and Ledford (287) observed that capsular material from micrococci isolated from Cheddar cheese stimulated acid production by a \textit{Lactobacillus} isolate, by lactic streptococci, and by \textit{S. thermophilus}. Capsular preparations contained sialic acid, N-acetylhexosamine, riboflavin, and certain amino acids in small amounts.

Symbiosis between lactic-acid bacteria is complex. Amino acids and peptides produced by \textit{L. bulgaricus} and \textit{L. helveticus} can stimulate growth of \textit{S. thermophilus} but the stimulatory value of these substances may vary between different strains. Further identification of these materials would help to explain the phenomenon of stimulation and might be of value to formulate new starter media. Recognition of the variation of degree or, even, absence of stimulation between different strains of starter bacteria suggests the need for careful selection of compatible strains for use in cheese making. It is known that strongly proteolytic strains of \textit{L. bulgaricus} are more stimulatory than less proteolytic strains. Perhaps a study of the proteolytic enzymes of \textit{L. bulgaricus} and \textit{L. helveticus} and determination of the specificity of their proteinases would help to further explain stimulation and/or physical and chemical changes occurring during cheese ripening. Amino acids and peptides have been designated as the primary stimulatory compounds in a majority of reported studies. This does not preclude the fact that nucleosides, formic acid, and other, as yet, unidentified factors might not be implicated.

\textit{Inhibition of growth}. Influence of metals on growth of high-temperature lactic-acid starters will be discussed briefly and only in regard to copper because of the traditional relationship between Swiss cheese and copper kettles. Mueller et al. (280) found that copper in amounts of 2 to 16 mg/l only slightly inhibited growth and acid production of \textit{S. thermophilus} R. Growth of \textit{L. casei} (280) was slightly stimulated by addition of 2 mg/l of copper, but higher amounts were slightly inhibitory. Growth and acid production of \textit{L. bulgaricus} K was unaltered by copper concentrations up to 20 mg/l. These experiments were done with artificial media, and growth was estimated by turbidimetry. Since turbidimetry measures both living and dead cells, Mauer and Reinbold (unpublished results) tested the influence of copper on the plate count of three strains each of \textit{S. thermophilus} and \textit{L. bulgaricus} grown in sterile milk with added copper. In the early growth stage, \textit{S. thermophilus} was slightly inhibited by amounts of 2 to 8 mg/l, but this inhibition was overcome after 24 hr. Inhibition was found at 16 mg/l. Two strains first decreased in number of viable cells but then, evidently following adaptation, grew more vigorously. \textit{Lactobacillus bulgaricus} was slightly inhibited at the higher copper levels.

Antibiotics are important inhibitors of starter organisms and may be found in cheese milk as a result.
of cow medication. Antibiotics also may be produced by added or adventitious bacteria. Nisin is produced by some strains of S. lactis and inhibits growth of L. helveticus and L. casei (65, 351). Reports on the reaction of S. thermophilus to nisin vary. For example, Cziszár and Pulay (65) report that it inhibits S. thermophilus, while Alifax and Chevalier (3) found that it had no effect. This difference may be the result of strain variation. Nisin added to cheese milk in Swiss-cheese manufacturing reduced lactic-acid production. Penicillin in amounts of 0.01 unit/ml inhibited S. thermophilus and 0.1 unit/ml was found to inhibit L. bulgaricus (145). In another study (402), from 0.3 to 0.6 unit penicillin/ml was necessary to inhibit growth of L. bulgaricus, L. acidophilus, and L. casei. Five μg/ml of streptomycin was enough to reduce growth of S. thermophilus, and 1 μg/ml retarded L. bulgaricus (145). A detailed study on the effect of 10 different antibiotics on pure cultures and mixed cultures of S. thermophilus and L. helveticus was reported by Winkler (463). After studying the sensitivity of strains of L. bulgaricus and S. thermophilus to 30 antibiotics, Reddy and Reinbold (unpublished data) observed that S. thermophilus was resistant to nalidixic acid and colistin. Lactobacillus bulgaricus was resistant to nalidixic acid. Usually, L. bulgaricus was sensitive to the same antibiotics as S. thermophilus but to a lesser degree.

Antibiotics have been implicated in many failures in the manufacture of Swiss cheese. Their presence usually interferes with acid production and results in high-moisture cheese of low market value. Continued testing of the milk supply for presence of antibiotics and other inhibitory compounds is necessary.

Bacteriophages

Few reports have been published concerning bacteriophages for high-temperature Swiss cheese starters. Most of the work on bacteriophage for these starters has been in association with their use as yogurt cultures. Deane et al. (77) tested 81 whey samples from U. S. Swiss cheese factories, many of which had experienced slow acid development; bacteriophage against S. thermophilus, however, was isolated from only one sample. This bacteriophage was quite specific and lysed only one of four strains of S. thermophilus. In Finland, Kiuru and Tybeck (203) isolated phages lysing S. thermophilus, L. lactis, and L. helveticus from starter cultures which were used for Swiss cheese manufacture and showed slow acid production. Streptococcus thermophilus phage was widespread, but only one L. helveticus phage was found. The isolated bacteriophages showed great strain specificity. They were destroyed by a heat treatment of 85°C (185°F) for 3 to 5 min, by strong acid (pH 3.0), or by basic solutions (pH >11.0) (203). Activity of these phages in the calcium-free, phage-resistant medium “Cockade” was almost completely inhibited, but growth of L. helveticus, L. lactis, and S. thermophilus was slow, so this medium could not be used to prepare Emmen-taler cheese starters (437).

Usually, bacteriophages against S. thermophilus have been isolated from yogurt (57, 59, 135, 331), but they also have been isolated from “soft-cheese” whey (402), and from whey samples taken during Gorgonzola production (403). A phage active against S. thermophilus has been isolated in a dairy plant; the same phage also was isolated from dust, dairy utensils, and leaves of beetroots and sugar beet (59). The isolate contained DNA. The phage showed an absolute requirement for calcium ions for replication (adsorption) (59). The concentration of calcium determined plaque size and plaque number (344). Electron microscopy showed that the head of the phage had a diameter of about 70 nm and an undifferentiated tail about 237 nm long (59). Gelin et al. (135) observed that their isolate had an exceptionally long tail, and the same unusual tail length was observed in our laboratory (Reddy and Reinbold, unpublished results). The latent period was about 80 min and burst-size was about 34 particles per infected bacterium (59). In a study of 17 different phages for S. thermophilus, Sozzi (401) observed that all phage strains survived 74°C (165.2°F) for 20 sec, so usual heat treatments in the dairy industry will not inactivate this phage.

So far, all isolated bacteriophages against S. thermophilus show a narrow host range. Sozzi and Prella (409) studied 20 strains of S. thermophilus and their bacteriophages and found that eight strains were sensitive only to their specific phage, six strains showed sensitivity to more than one strain, usually two or three, and the six remaining strains showed evidence of mutation for phage resistance. During investigation of phage-resistant strains of S. thermophilus, Ciblis (58) observed that the resistant strain was lysogenic for Phage 63. This resistant strain did not adsorb the phage. Light microscopy showed that phage-sensitive cells formed colonies with smooth edges, but colonies of the phage-resistant strain were smaller and had slightly rough edges. Electron microscopy showed that cells of the phage-sensitive strain had a rough surface, but surfaces of cells of the phage-resistant strain were smooth. No difference in cell-wall composition or in physiology was found between these strains (58).

As mentioned, most of the work with bacteriophage
against *S. thermophilus* has been done in connection with yogurt manufacture. The work of Deane et al. (77) seemed to indicate that these phages were of little significance regarding slow-acid development in Swiss cheese. These researchers sampled whey from the vat, however, and that may not be the proper time to test for *S. thermophilus* phage. Frazier et al. (120) observed that *S. thermophilus* grows slowly during the time in the vat. It was only after 3 to 4 hr after dipping that *S. thermophilus* started to grow (logarithmic growth phase) (117). It would probably be more correct to test for phage in whey samples taken during optimum growth of *S. thermophilus*. Preliminary results in our laboratory confirm this assumption (Reddy and Reinbold, unpublished results). Interference with growth of *S. thermophilus* in cheese curd during pressing would result in the presence of residual, unfermented sugar after pressing. The pH of the curd could conceivably be too high at this time, signifying production of poor quality cheese. It is our belief that many of the failures in Swiss-cheese making (as well as Italian-cheese making) attributed to poor growth of lactobacilli during pressing would, probably, be more correctly attributed to presence of *S. thermophilus* bacteriophage.

**MANUFACTURING PROCESSES AND PROCEDURES**

For a recent, detailed description of the manufacture of wheel or drum and rindless, block Swiss cheese, consult the monograph by Reinbold (345).

The reader is cautioned that much of the research data and observations presented throughout this review have been drawn from European and not too recent American literature. This implies that the cheeses studied would have been wheels with rinds. The cheeses would have been pressed separately in wooden hands and would normally contain somewhat less moisture than the rindless counterpart. Copper kettles and whey starters would have been used as would longer brining periods and different make and curing temperature combinations. Probably only those articles published in the United States after the early 1950s, or even later European literature, would have referred to our now conventional rindless block Swiss cheese. This does not invalidate these data or references but it does suggest that the reader must be prepared to compare, interpret, and interpolate this information according to his specific needs and purposes.

**Growth in the vat**

The usual time for curd-making, from setting to dipping, is approximately 2.5 to 3 hr depending on the bacterial content (ripeness) of the milk. A well-ripened milk, in which considerable bacterial growth has taken place, requires a much shorter make period because this milk already contains a low amount of preformed acid. Earlier cheesemakers preferred milk with a methylene-blue reduction time of 3 to 4 hr so that the make process would proceed rapidly (116). Starter is added at the same time as rennet or within 20 to 30 min before rennet addition. If added before this time period, *S. thermophilus* will most usually be through its lag phase before the milk has coagulated.

As the quality of cheese milk became better, preripening of milk became more common. Frazier et al. (119) preripened cheese milk with *S. thermophilus* and found that preripening improved cheese quality if the kettle milk had a methylene-blue reduction time of 5 to 6 hr or longer. In their experiments, one-fifth of the cheese milk was first ripened with *S. thermophilus* and was then added to the rest of the cheese milk in the kettle. The increase of *S. thermophilus* was greater in the kettle than if it had been added as normal starter, but it grew more slowly during pressing. The pH in the interior of cheese made from milk ripened with *S. thermophilus* was lower during the early hours of pressing, but after 21 hr, the pH values became higher than those of the control cheeses (119).

The propionibacteria are added at the same time as the other starter organisms. The first worker to add pure cultures of propionibacteria to cheese milk was Sherman (393) and this procedure then became customary in the United States. Addition of pure cultures of propionibacteria was uncommon in Switzerland until Dorner et al. (92), in 1938, did experiments in which they made high-quality Emmentaler cheese with the addition of 1/8 to 1/4 drop of propionibacteria culture per cheese. Nevertheless, practical cheesemakers in Switzerland asserted as late as 1958 that a large inoculum of propionibacteria was harmful to the quality of Swiss cheese and led to irregular eye formation. Experiments by Oehen et al. (298) showed that large inoculations did not cause defects whereas too small an inoculum led to cheese with inferior flavor. An excess of propionibacteria led to a "stagnation" in pH after 10 weeks presumably because the bacteria attained their maximum number too rapidly. Production of volatile fatty acids and carbon dioxide paralleled the increase in number of propionibacteria. Babel and Hammer (18) found that an inoculum of 25 ml of *propionibacterium* culture per 43.5 kg (100 lb) of cheese, produced cheese with the desired sweetness, but larger amounts produced too-sweet cheese. An inoculum of 5 ml produced cheese lacking in sweetness but with a slightly more desirable eye number and...
size. Related studies showed that Swiss cheese milk must contain a minimum number of propionibacteria to yield high-quality cheese (289, 297) and that the propionic-acid fermentation was more affected by the initial pH after pressing than by the size of the inoculum of the propionic culture (91).

Growth of bacteria during the manufacturing process in the kettle has been investigated by Frazier et al. (120). Streptococcus thermophilus increased slowly during the entire process but more rapidly during the later phase at higher temperatures. Lactobacillus casei and L. bulgaricus decreased slightly in number as did P. shermanii. Streptococcus lactis increased during the initial portion of the process, but growth was arrested by the cooking temperature [ca. 53°C (127°F)]. The pH decreased from 6.57 to 6.48 during this part of the process (120).

After setting the milk, the coagulum is cut. Finely harped Swiss cheese curd contains about 1% more moisture than coarsely harped curd (430). Curd particles from a mechanical vat were of more uniform size than curd from a, then, conventional vat. The number of eyes in cured Swiss cheese decreased with decreasing curd particle size (208). Cheese dust tends particularly to cause retention of whey during pressing and may lead to defective fermentation, oversetting, and gassiness of the cheese (430).

Removal of a portion of the whey and addition of water after cutting lead to an improvement in organoleptic properties, eye pattern, and consistency of the cured Swiss cheese in comparison with controls (32, 35, 469). The cooking temperature of the Swiss cheese is usually between 50°C (122°F) and 54°C (129.2°F) (430), although in some Swiss-type cheeses with S. lactis as the principal lactic-acid starter a lower cooking temperature [40 to 46°C (104 to 114.8°F)] is used (469). Too-low cooking temperature may result in survival of an adverse flora, which may lead to faulty fermentation.

Rogers et al. (362) found that, with milk lacking in ripeness and with a whey pH greater than 6.51 at dipping, about 12% of the Swiss cheese manufactured was of good quality. With the pH at dipping between 6.35 and 6.51, most of the cheeses were of high quality (362). If the pH was lower than 6.30 at dipping, surfaces of eyes became rough. Using a dip pH between 6.30 and 6.35 is risky (447) and may lead to Swiss cheese of low quality.

One major difference between Swiss cheese manufacture in Europe and the United States is that copper vats are used much more often in Europe. Only a very few copper-lined kettles may still be in domestic use. Use of stainless steel vats is believed to lead to an increase in production of acetic and propionic acid and a decrease in lactic-acid content during ripening, which, ostensibly, reflects the unfavorable effect of copper on propionibacteria (114). Mature cheese from copper vats contains 13.84 mg copper/kg vs. 0.94 mg copper/kg in cheese from steel vats (190). The copper content increases particularly during cooking and totally oxidizes the ascorbic acid. The protein breakdown in cheeses is delayed, but in organoleptic tests, cheese from steel vats was generally rated lower (190). In the United States, Mueller et al. (280) found that copper concentrations of 8 mg/kg did not affect the quality, but 16 mg/kg resulted in abnormal cheese. In a study on the effect of copper on organoleptic properties of Swiss cheese made by a commercial plant and Iowa-style Swiss cheese made at Iowa State University, the taste panel preferred the copper-containing cheeses (3 to 7 mg/kg) from the commercial plant. The opposite result was obtained on Iowa-style Swiss cheese (Mau rer and Reinbold, unpublished results). This indicates that make-procedures (345), and also the starters used, will influence the flavor and quality of Swiss cheese. Liberation of copper from the vat was greater from oxidized than from shining surfaces (112).

Growth in the press

Draining and pressing of curd regulates moisture content and allows acid production to assist in knitting curd particles together. The area just beneath the rind cools more rapidly than the interior of the curd mass. Initiation of bacterial growth and acid production corresponds generally with the decrease in temperature. Close to the surface of the cheese, the high-temperature lactic starters start to multiply during the first few hours of pressing (51). Frazier et al. (117) found that S. thermophilus started to grow 3 to 4 hr after dipping and increased rapidly to the 6th or 8th hr. Thereafter, the increase in number of S. thermophilus was slow, and sometimes there was even a decrease in number. The pH drop was fastest the first 7 to 8 hr and was mainly due to S. thermophilus which produced cells of a diameter 3 to 4 times as large as cells grown at ordinary temperatures. This possibly explains the large fermentative activity per cell. The buffer value of the cheese also is low at this early pH range, so low acid production may produce relatively large pH changes (117). The high acid production of S. thermophilus also is important because it facilitates moisture drainage from the cheese. If there are large differences in pH between the interior and the area just beneath the rind, the result is insufficient drainage and a high moisture content, which may cause checking or splitting (51). Lactobacillus helveticus decreased slowly in number to the 6th to 8th hr in the press and then l-
creased fairly rapidly. *Lactobacillus bulgaricus* decreased until about the 5th hr, and then a rapid increase in number took place (117). These species reach their maximum number after 12 to 14 hr and then slowly decrease (79). Growth of the thermophilic rods is stimulated by the acid production of *S. thermophilus* (432). *Propionibacterium shermanii* did not grow in the press (117) and the number usually decreased slightly (79). Bacteria of the *L. casei* type usually increased in numbers. The high temperature of the cheese in the press, and the high activity of starter organisms, effectively suppresses growth of other bacteria (117).

Acidity of the curd should be below pH 5.3 at the end of pressing to obtain good flavor and texture in the cheese. Higher pH values will cause lower flavor scores and also will lead to soft texture (447). Too many eyes will be produced in the cheese, a defect known as oversetting (430). The acidity must not be lower than pH 5.0. Excessive acidity is very inhibitory to growth of propionibacteria as discussed earlier, and thereby, on propionic acid and eye formation (431). There is a significant relationship between pH after pressing and growth of *P. shermanii* (236).

Even the height of the forms (or curd blocks) has been shown to strongly influence Swiss cheese quality. Uotila (438) found that cheese made in hoops 17 cm (6.7 inches) in height were of better quality than those in hoops 18 (7.1) or 19 cm (7.5 inches) in height. The cheese was of better texture, contained a higher percentage of solids and sodium chloride, and was less liable to defects in eye formation and flavor. With increasing height of cheese hoops, it was necessary to prolong the salting periods to obtain cheese of equal quality. This is another point of disparity between domestic and imported Swiss cheese. Domestic Swiss cheese usually is made to cure at slightly less than 20.3 cm (8 inches) and is most frequently brined for only 1 day. Imported Swiss cheese is not as thick and is brined longer.

**Effect of salting**

Salt is usually added to Swiss cheese to improve flavor and control ripening. Salt also has a desiccating effect and, thereby, increases firmness so cheese is less susceptible to damage from handling (430). The propionibacteria are usually very sensitive to salt; therefore, Swiss cheese must be salted lightly if eyes are to develop normally (116). Demeter et al. (79) found that the number of propionibacteria reached their lowest level during brining. The number of *S. thermophilus* and *L. helveticus* also decreased during brining primarily because of the sharp fall in temperature. Oversalting leads to few eyes, and too-little salt is one of the causes of the "oversetting" defect (430).

The effect of salt on Swiss cheese quality has been investigated by Ołanski et al. (306). Cheeses were salted for 1 to 9 days in brine of 21°C Be at 12°C (53.6 F) and contained from 0.64 to 1.70% salt. Cheeses containing 0.76 to 1.70% salt were of the best quality because of better eye formation, consistency, flavor, and, to some extent, color. Therefore, cheese should be brined for about 3 days. Babel and Hammer (18) found that cheese that was salted 2 days after manufacture had more sweet flavor and a larger number of propionibacteria than when it was salted normally, that is, 1 day after pressing. Brine tanks in 20 different Swiss cheese factories contained more than one million bacteria/ml as shown by Kurmann (231), but the primary flora was of a harmless nature.

**Wrapping**

Because of the combined influences of lower moisture and higher salt content, as well as other factors, propionibacteria are not present in the rind of wheel Swiss cheese in any appreciable number (347). Unpublished data obtained in this laboratory indicate that rindless block Swiss cheese has essentially equivalent numbers of propionibacteria present throughout the curd mass during curing.

**Pre-storage of cheese before ripening**

During brining cheese is held at 12 to 14°C (53.6 to 57.2 F). In the United States, cheese usually is pre-stored at 7 to 14°C (44.6 to 57.2 F) for 7 to 10 days before it is moved to the hot room (345). In an experiment in Germany, Hanusoh and Bösiger (144) found that pre-storage of Swiss cheese at 14 to 17°C (57.2 to 62.6 F) up to 90 days before ripening usually improved the quality of the cheese. In Switzerland, Oehen and Bolliger (295) found that pre-storage improved eye formation, but only at some loss of characteristic texture and flavor, and in a second study of Swiss cheese from low bacterial count milk, these results were confirmed (296). Oehen and Bolliger (295), therefore, did not recommend pre-storage. At this pre-storage temperature of 12 to 17°C (53.6 to 62.6 F) very little growth of the bacteria in the Swiss cheese takes place.