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Bacteriology of Cheese

V. Defects of Blue (Roquefort-Type) Cheese

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SUMMARY

A black discoloration and a musty flavor in blue cheese were attributed to the growth of *Hormodendrum olivaceum*, particularly in punch holes and cracks in the surface.

Gas formation is of relatively little importance in blue cheese, presumably because of the open texture, which permits the gas to escape, and the unfavorable conditions in the cheese for growth of the common gas-forming organisms. Trials with a culture of *Aerobacter aerogenes* recently isolated from gassy cheddar cheese showed that inoculations (of the milk) which resulted in very gassy cheddar cheese caused no gas holes or only insignificant numbers in blue cheese.

A defect of blue cheese in which a portion of the edges became soft appeared to be caused by excessive moisture in the softened parts of the cheese. The defect was readily reproduced by placing cheese near a humidifier where free moisture could strike it. Various conditions encountered in curing rooms favor the accumulation of moisture on the cheese and thus may be involved in the defect.

In the outbreak studied the lack of mold growth in blue cheese apparently was caused by the use of a mold powder in which an atypical strain of *Penicillium roqueforti* predominated. The variation in the mold may have been caused by the long continued cultivation on an artificial medium of the culture used to prepare the powder, although there also is the possibility of contamination of the culture.

A defect in which a gray discoloration and a mousy, ammoniacal flavor, that later became soapy, developed in blue cheese was accompanied by an increase in pH. The variation from the normal ripening mechanism which caused the defect presumably involved the formation of basic products from protein. Extensive development of *Penicillium roqueforti* or growth of contaminating organisms could be responsible for an abnormal protein decomposition. Contaminating organisms capable of reproducing the defect could not be isolated.

Bacteriology of Cheese

V. Defects of Blue (Roquefort-Type) Cheese¹

BY H. W. BRYANT AND B. W. HAMMER

The ripening of any of the important types of cheeses is a complicated microbiological process, and variations in the results are to be expected. In blue cheese, as in most other types, some of the variations are of minor nature, and the cheese showing them are still considered satisfactory. In other cases the variations are of more importance, the cheese involved being definitely defective.

The objectionable conditions encountered in blue cheese vary widely, and some of them are difficult to classify. However, a number of rather specific defects have been noted and, in general, these are much the same as the defects encountered in other cheeses.

An improvement in the general quality of blue cheese requires a reduction in the number of cheese showing minor variations from the most desirable qualities and elimination of the cheese showing definitely objectionable conditions. As a basis for this, the causes of the various defects should be determined.

The studies reported herein deal with some of the defects in blue cheese that have been encountered in the attempts of the Iowa Agricultural Experiment Station to assist in the development of a blue cheese industry in the United States. The defects considered are: (a) Black discoloration, (b) gas formation, (c) soft edge defect, (d) lack of mold growth and (e) gray discoloration.

METHODS

MANUFACTURE OF BLUE CHEESE

All milk used for the manufacture of blue cheese was homogenized at 92°F. and a pressure of 2,200 pounds per square inch. Two-percent cheese culture was added to the milk and the milk ripened to an acidity of 0.19 to 0.20 percent. Rennet was added at the rate of 3 ounces per 1,000 pounds of milk; it was diluted to 20 times its volume with water before the addition. The milk was set at 88°F. A setting period of 1 hour was used, after which the curd was cut with one-half-inch knives. The curd was held for

¹ Project 119 of the Iowa Agricultural Experiment Station.

1 hour with occasional stirring and then dipped into swiss-cheese cloths, where it drained. After draining for several minutes the mold powder was added and the curd hooped. The cheese were turned for the first time 15 to 20 minutes after hooping. They were turned again four or five times at increasing intervals during several hours and then drained over night. The cheese were dry salted at the rate of 6 pounds of salt per 100 pounds of green cheese. After salting the cheese were punched and placed in the curing room.

MANUFACTURE OF CHEDDAR CHEESE

The milk used to make cheddar cheese was adjusted to 86°F. in a vat. Two-percent cheese culture was added and the milk ripened to an acidity of 0.16 percent. Cheese color was added at the rate of 1 ounce per 1,000 pounds of milk and rennet extract at the rate of 3 ounces per 1,000 pounds of milk, the rennet being diluted to 20 times its volume with water before the addition. The milk was set for 25 minutes and then cut with three-sixteenth-inch knives. The curd was cooked at 102°F. until the acidity in the whey reached 0.16 percent and the desired firmness of the curd was obtained. After dipping, the curd was cheddared until about 0.5 percent acid in the whey was reached. Upon completion of the milling, the curd was forked for about 15 minutes and then 3-percent salt was added. As soon as the salt had completely dissolved, the curd was placed in 5-pound hoops and pressed over night. The cheese were dried at 50°F. for 2 days and then par-affined. They were then placed in a 50°F. curing room to ripen.

PLATING CHEESE

One gm. of cheese was weighed on a sterile paper, transferred to a mortar and ground to a homogeneous suspension with 9 ml. of 2-percent aqueous sodium citrate solution. In making the subsequent dilutions, 1 ml. of the suspension was used in 100 ml. water blanks.

The media employed varied with the organisms that were of particular interest. Czapek's and acidified tomato juice agars were used in plating for molds, tomato juice and acidified tomato juice agars in the detection of yeasts and beef infusion agar, often with skim milk or fat emulsion or both added, for total, proteolytic or lipolytic bacterial counts.

DETERMINATION OF pH

Two gm. of cheese was placed in a mortar and ground to a thick paste. Ten ml. of boiled and cooled distilled water was added and the mixture ground to a homogeneous suspension. Measurements were made with a potentiometer, using a quinhydrone electrode and saturated calomel cell.

DETERMINATION OF MOISTURE AND SALT

The methods employed were those recommended by the Subcommittee Report on "Determination of fat, moisture, and salt in hard cheese," (2).

PREPARATION OF MOLD POWDER

Mold powder was prepared with the procedure suggested by Hussong and Hammer (19).

EXPERIMENTAL

BLACK DISCOLORATION OF BLUE CHEESE

Blue cheese is characterized not only by its flavor but also by the blue veins of mold growth through it. Contaminating molds occasionally develop in this type of cheese. If the color produced by them is rather similar to that of the normal blue portions it is overlooked, but if it is not similar it constitutes a defect. One of the most noticeable of the color defects is a black discoloration.

GENERAL OBSERVATIONS

Several lots of cheese made in a commercial plant showed a black discoloration and also had a musty flavor in the discolored areas. There was little evidence of normal slime formation, the cheese being dry at the surface and cracked in several places. The cracks were filled with a black mold growth, and most of the surfaces of the cheese were darker in color than normal. Cut surfaces of the cheese showed that there had been an invasion of the cheese through the punch holes and that the discoloration extended beyond the areas of black mold growth. There were many areas of normal mold growth, and in these the flavor of the cheese was fair.

HISTORICAL

Microorganisms are known to cause various color defects in cheeses. Investigations indicate that bacteria often are responsible.

Gruber (15) found that brownish red spots in hard and soft cheeses were due to *Bacterium casei fuscum*. Thöni and Allemann (32) attributed red spots in emmenthal cheese to a propionic acid organism which they named *Bacillus acidii propionici* var. *ruber*. Burri and Staub (7) described an organism responsible for a similar discoloration and designated it *Bacterium subrufum*.

Connell (9) isolated an organism causing red or rust-colored spots in cheddar cheese and named it *Bacillus rudensis*. This organism also was found in defective cheese by Harding and Smith (16). Davis and Mattick (12) isolated an organism causing rusty spot in cheddar cheese which they regarded as a true lactic acid organism that produced pigment under favorable conditions. Later, Mattick and Davis (22) noted that the most important factor in growth of the organism in milk is the presence and growth of certain other bacteria.

Stocker (30) reported black spots and black discoloration of the rinds of cheeses. For the most part *Monilia nigra* was responsible. When it was absent a bacterial species producing a black color was isolated.

Discoloration in cheddar cheese was studied bacteriologically by Morgan (26); he noted that discoloration occurred near cracks or openings in the cheese and concluded that it was caused by entrance of molds already established on the surface. Leitch (20) reported that the factor causing mottling and bleaching in cheddar cheese is of bacterial origin and that certain organisms of the coliform group invariably accelerate discoloration.

EXPERIMENTAL

Small portions of cheese showing the black discoloration were plated on Czapek's, tomato juice and acidified tomato juice agars. The plates were divided into three lots and incubated at 10°, 21° and 37°C., the incubation time being arbitrarily set at 1 week. The plates incubated at 37°C. showed very few molds and none suggestive of causing a black discoloration in cheese. Many of the plates incubated at 10° or 21°C. developed dark mold colonies in addition to colonies of *Penicillium roqueforti*. Of the three media, Czapek's agar appeared the most favorable for growth of the dark mold. Several colonies of this mold were picked to Czapek's agar slants and spotted on Czapek's agar plates for further study. Plating of the original cheese on Czapek's agar was repeated several times, and in all cases plates incubated at 10° or 21°C.

developed a significant number of dark mold colonies. Samples of normal blue cheese from various sources, when plated on Czapek's agar and the plates incubated at 10° or 21°C. for 1 week, showed an occasional contaminating mold, but none was dark in color or in any way resembled the mold present in the defective cheese.

Preliminary examination indicated that all the colonies of the dark mold were similar and were probably the same species. Its presence in significant numbers in the defective cheese but not in normal cheese from various sources suggested that it was responsible for the black discoloration. Mold powder was prepared with the dark mold (19), and trials were carried out in an attempt to duplicate the defect.

In the first trials, in which cheese weighing about 2.5 pounds each were used, addition of the powder made with the black mold to the curd resulted in a defect essentially like the original. The surfaces of the cheese eventually became brown to black; cutting the cheese showed there was black mold growth in the punch holes and a darkening near the surfaces, especially next to punch holes with the black growth in them. A slight musty flavor developed in the cheese.

Later trials, using normal-size cheese, confirmed the early observations. The general effects of the mold are illustrated by the following trial:

Sufficient blue cheese curd to make three cheese of normal size was divided into three portions. One portion was inoculated with powder made from the dark mold, one portion with a mixture of the powder made from the dark mold and normal mold powder and one portion with only normal mold powder. The lots of curd were then handled in the usual manner. After ripening 3 months the cheese were cut and observed. The surface of the cheese made with the dark mold suggested nothing abnormal, but when the surface slime and mold were removed many of the punch holes appeared as conspicuous, regularly spaced, black pits in the surface of the cheese. Near these punch holes the cheese had begun to turn dark. The outside appearance of the cheese made with the mixed powder and of the control cheese was normal. The cut surface of the cheese made with the dark mold showed black streaks following the punch holes, and the cheese surrounding these streaks was dark. The dark mold had not spread through the cheese and was confined to the punch holes and openings leading from them.

The flavor of the cheese was musty and did not suggest blue cheese. The cut surface of the cheese made with the mixed powder showed normal blue areas, black areas and areas in which the mold appeared dark gray; the gray areas probably were caused by the two molds growing in close proximity. The flavor of the cheese suggested a combination of blue cheese and musty flavors. The cut surface of the control cheese was normal, and the cheese had developed a fine flavor.

IDENTIFICATION OF DARK MOLD

Various cultures of the dark mold were studied in considerable detail. The organism was identified, according to the classification of Gilman and Abbott (13), as *Hor-modendrum olivaceum*.²

ADDITIONAL OBSERVATIONS

A blue cheese showing black discoloration was encountered on the market. It was from a plant other than the one supplying the cheese investigated. There were dark brown areas on the surface, and the entire surface was darker than normal. The cut surface showed black streaks corresponding to the punch holes and a discoloration which was most intense beneath the dark surface portions. No areas of normal mold growth were observed. The cheese had a musty, peppery flavor which was not at all typical of blue cheese.

DISCUSSION

Under natural conditions the invasion of blue cheese by *H. olivaceum* would be expected to occur through cracks and punch holes. If the texture of the cheese is open, the mold has an opportunity to spread through the cheese and produce extensive black discoloration. Under experimental conditions the growth of the mold was confined to the outside of the cheese and to punch holes. This indicates that *H. olivaceum* requires a good oxygen supply. From their investigations, Thom and Currie (31) concluded that the dominance of *P. roqueforti* in the interior of roquefort cheese is due partly to the reduced oxygen content which favors the growth of the normal mold over other types. For this reason *H. olivaceum* would not be expected to cause a great deal of spoilage in normal cheese. It is conceivable, however, that under certain manufacturing conditions, in which the cheese had

² The identification was confirmed by Dr. J. C. Gilman.

a body particularly susceptible to cracking, the mold could cause extensive damage.

The musty flavor produced by *H. olivaceum* is of less importance than the discoloration. In experimental cheese made with a mixture of *H. olivaceum* and *P. roqueforti*, the flavor of the cheese was not typical but was close enough to normal to satisfy most consumers.

If the proper procedure is followed in making blue cheese, and particularly when homogenized milk is used, the cheese will have a body that is not susceptible to cracking, and if the proper humidity is maintained in the curing room, the cheese will not dry and crack. Suitable manufacturing methods should do much to control black discoloration.

H. olivaceum is not the only contaminating mold that grows in blue cheese under certain conditions. The color produced by such a mold is of special importance; colors which do not contrast sharply with the normal colors of a cheese are not conspicuous and, accordingly, are less objectionable than sharply contrasting colors.

GAS FORMATION IN BLUE CHEESE

In certain cheeses gas-forming organisms, including those of the genus *Propionibacterium*, are necessary to produce the characteristic eyes, but in other cheeses gas-siness and off flavors often associated with it are definitely objectionable. Gas formation occurs in blue cheese but is of much less significance than in cheddar cheese. The organisms most commonly responsible for undesirable gas formation in various cheeses are those of the *Escherichia-Aerobacter* group.

GENERAL OBSERVATIONS

Gas formation has been observed in only a few blue-veined cheeses, all of which were imported. None of the cheeses were bulged, and the outside appearance did not suggest the presence of the gas holes. The holes were practically round, varied in size and were not numerous. The mold growth was normal, and apparently the flavor was not affected.

HISTORICAL

The importance of *Escherichia-Aerobacter* organisms as a cause of gas formation in cheddar cheese was emphasized by the early studies of Russell (27), Moore and Ward (25), Marshall (21) and Harrison (17). Whitehead

(34) found that when colon organisms were added to milk the resulting cheddar cheese developed unclean flavors but no gas holes. Leitch (20) reported that production of gas in cheddar cheese curd was due primarily to the colon group but sometimes was due to *Bacillus welchii* or certain yeasts.

In swiss cheese, gas formation apparently is due to various organisms; Russell and Hastings (28) reported lactose-fermenting yeasts, Burri (6) reported butyric acid organisms, Albus (1) reported *Clostridium welchii* and Babel and Hammer (3) reported *Escherichia-Aerobacter* organisms.

EXPERIMENTAL

Three gassy, imported blue-veined cheeses (one roquefort, one French blue and one Danish blue) were examined for gas-producing organisms by preparing plate and shake cultures, but the organisms were not found in significant numbers. The cheeses were well ripened, and the long exposure of the gas-forming organisms to acid, salt and low temperatures presumably resulted in their destruction. Commonly, gas production in cheese occurs during the manufacture or early part of the ripening. With gassy cheddar cheese that has been aged for some time it often is difficult to isolate the causative organism.

Although the general observations indicated that gas production is of relatively little significance in blue cheese, trials were carried out in which a gas-producing organism was inoculated into the milk used for blue cheese; cheddar cheese, made with milk from the same lot and inoculated in the same way, was used as a control.

TRIAL 1

Raw milk of fair quality was divided into two portions of about 100 pounds each. The milk for blue cheese was homogenized, cooled to 60°F. and placed in a refrigerator. The milk for cheddar cheese was divided into two equal lots, and each lot was placed in a compartment of a five-section experimental cheese vat. Two-percent cheese culture was added to each lot. One lot was used as a control, and to the other was added 10 ml. of a milk culture of *Aerobacter aerogenes* which recently had been isolated from gassy, raw milk, cheddar cheese. The cheese were manufactured in the usual manner. When they had reached the cheddaring stage the homogenized milk was taken from the refrigerator, and manu-

facture of the blue cheese was begun. This procedure was necessary, because different temperatures are employed in the manufacture of the two types of cheese, and all five compartments of the vat are heated by the same jacket. The milk for blue cheese was divided into two equal lots, and each lot was placed in one of the unused compartments of the vat. After adding 2-percent cheese culture to each lot, one lot was kept as a control while to the other was added 10 ml. of the culture of *A. aerogenes* used with the cheddar cheese. The manufacture of the cheese was completed in the normal manner.

The cheddar cheese curd from each compartment was pressed into a loaf of approximately 5 pounds; the two cheese were dried, paraffined and placed in the curing room. The blue cheese curd from each compartment made one normal size cheese; the two cheese were salted, punched and placed in the curing room.

After 1 month the experimental cheddar cheese was very slightly bulged, and the cut surface showed large numbers of small gas holes. The flavor and odor were unclean and suggestive of gassy cheese. The control cheddar cheese had a normal outside appearance, and while the cut surface was solid for the most part, it had a small number of openings suggestive of gas holes. The flavor and odor, however, were mild and clean. The blue cheese were examined at this time also but were not cut. The outside appearance of each was normal, and several plugs drawn from each indicated that the mold growth and texture of both cheese were good, no off flavors or gas holes being noted. After 2 months both the cheddar and blue cheeses were cut. The observations made at the end of 1 month were confirmed.

TRIAL 2

Good-quality, raw milk was obtained and trial 1 repeated, with the exception that to each experimental lot of milk 20 ml. of a milk culture of *A. aerogenes* was added instead of 10 ml. The cheese were ripened 2 months before being cut. The experimental cheddar cheese was normal in shape, but the cut surface showed large numbers of gas holes, and the cheese had an unclean flavor. The control cheddar cheese was close textured and had a mild, clean flavor, but there were a few questionable gas holes in it. The experimental blue cheese showed a few small gas holes, but the number and size were such that they would be noted only on very detailed examina-

tion; for the most part the holes were in the outer portion of the cheese. As is usual, the center of the cheese was more open than the outer portion. The mold growth in the cheese was normal and some of the characteristic flavor had developed; no unclean flavor was detected.

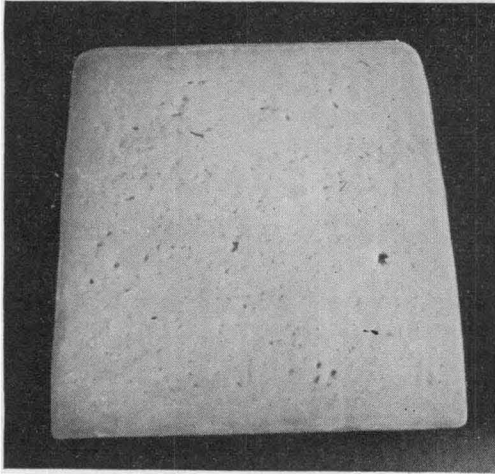


Fig. 1. Cheddar cheese made from raw milk inoculated with *A. aerogenes*. Gas holes evident.

The control blue cheese was free of gas holes; the mold growth was normal and the cheese had begun to develop desirable flavor.

Figures 1 and 2 show the cut surfaces of the experimental and control cheddar cheese.

TRIAL 3

Milk similar to that used in trial 2 was pasteurized at 143°F. for 30 minutes, and trial 2 was repeated.

After ripening 2 months the cheese were cut. The experimental cheddar cheese was not definitely bulged; however, it was filled with many small gas holes and had an unclean, slightly bitter flavor, with no typical cheddar cheese flavor. The control cheddar cheese was close textured and free of gas holes but was slightly bitter and lacked cheddar cheese flavor. Both blue cheese appeared normal and showed no gas holes; with each there was a more compact and brittle body than is normally found in raw milk cheese, and although there was good mold growth the cheese lacked flavor.



Fig. 2. Cheddar cheese made from raw milk not inoculated with *A. aerogenes*. Few questionable gas holes evident.

The cut surfaces of the experimental and control cheddar cheese are shown in figs. 3 and 4 and of the experimental and control blue cheese in figs. 5 and 6.

TRIAL 4

Regular pasteurized market milk was used to repeat trial 3 with results essentially the same as those obtained in trial 3.

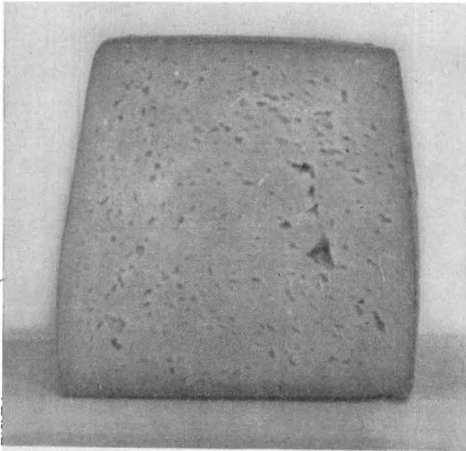


Fig. 3. Cheddar cheese made from pasteurized milk inoculated with *A. aerogenes*. Gas holes evident.

DISCUSSION

The much greater production of gas holes by *A. aerogenes* in cheddar cheese than in blue cheese presumably was due to several factors. Perhaps the most important was the difference in texture between the two types of

cheeses. In cheddar cheese the texture is close and retards the escape of gas, while in blue cheese the texture is open, and gas should escape readily.

In milk used for blue cheese, homogenization results in an early production of fatty acids which have an inhibitory effect on various bacteria. While the cheese culture organisms apparently develop rapidly in the milk, they are present in relatively large numbers, and less effect would be expected on them than on species present in much

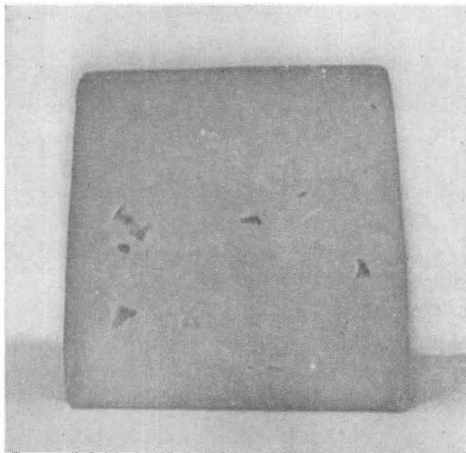


Fig. 4. Cheddar cheese made from pasteurized milk not inoculated with *A. aerogenes*. No gas holes evident.

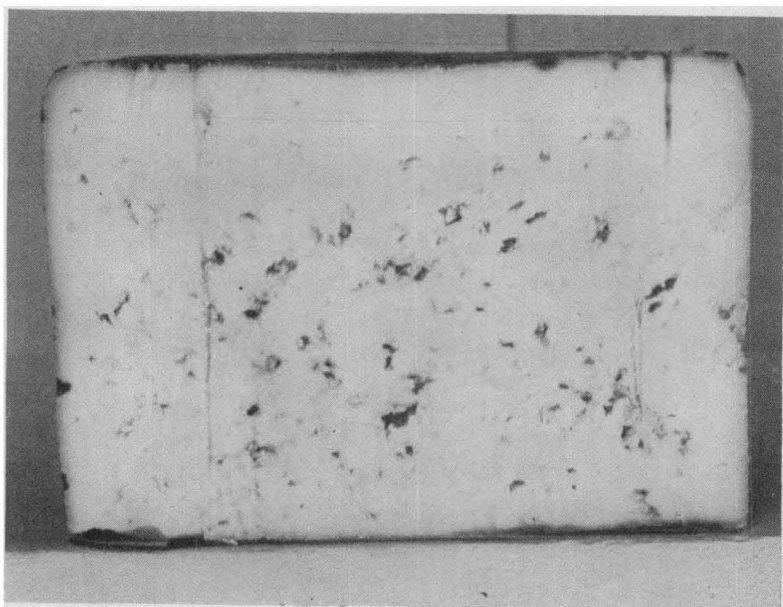


Fig. 5. Blue cheese made from pasteurized milk inoculated with *A. aerogenes*. No gas holes evident.

smaller numbers. The temperatures employed in making blue cheese are not as high, especially during the cooking process, as those used in cheddar cheese and would tend to limit the development of *A. aerogenes*. The delayed salting with blue cheese apparently does not compensate for the other factors involved.

Presumably, the results obtained with *A. aerogenes* are essentially the same as those that would be obtained with other species of the *Escherichia-Aerobacter* group.

SOFT EDGE DEFECT OF BLUE CHEESE

Blue cheese occasionally develops a defect in which a portion or all of the edges of a cheese become soft. The high humidity at which blue cheese must be ripened may favor the development of undesirable conditions not encountered in most other ripened cheeses. The soft edge defect is of practical importance because the defective edge must be removed before the cheese is marketed.

GENERAL OBSERVATIONS

The defect in a soft edge cheese is confined to the edges and immediate vicinity. It usually penetrates to a depth of 0.25 to 1 inch. The cheese appears normal in shape and color and has the usual slime formation on the

surface, but when the firmness of the cheese is tested by pressing with the thumb or forefinger, the top, sides and bottom of the cheese are firm while the edge is soft. There is no gradual softening as the defective edge is approached but rather a sharp dividing line between the firm cheese and the soft edge. The surface of a soft edge is very easily slipped off, exposing soft cheese beneath. The soft material has the color and consistency of well-ripened camembert cheese. When it first develops, the soft material does not have an off flavor or odor, but after a period of about 1 month it may have the flavor and odor of limburger cheese; the flavor of the normal portion of the cheese is not influenced.

Detailed observations on the defect were limited to one plant. The first signs of soft edge were noted on cheese which had been ripened for 4 to 6 weeks. In some of the racks of ripening cheese the defect had developed only on one or two end cheese. With others the defect involved the edges on one side of each of several cheese. In some instances the defect extended completely around each cheese, but when this occurred it often was much more pronounced on one side of a cheese than on the other. The defect was not present in all the cheese of a day's make, but frequently occurred in some of them

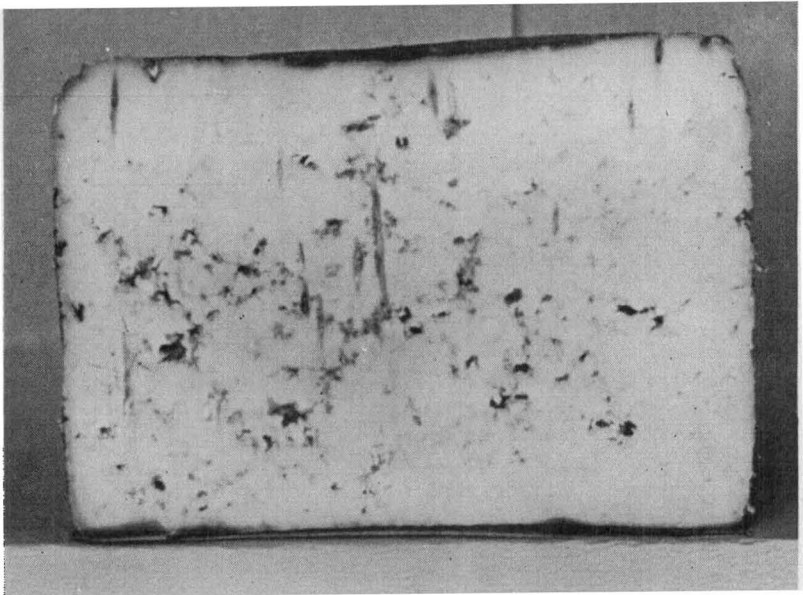


Fig. 6. Blue cheese made from pasteurized milk not inoculated with *A. aerogenes*. No gas holes evident.



Fig. 7. Blue cheese showing soft edge defect.

and not in others. It was more prevalent during the spring and summer than during the fall and winter.

Investigation showed that cheese near the humidifier or the refrigeration pipes in the curing room developed soft edges most often. This suggested that certain moisture conditions favored the defect. Other evidence to support this was the fact that if the position of defective

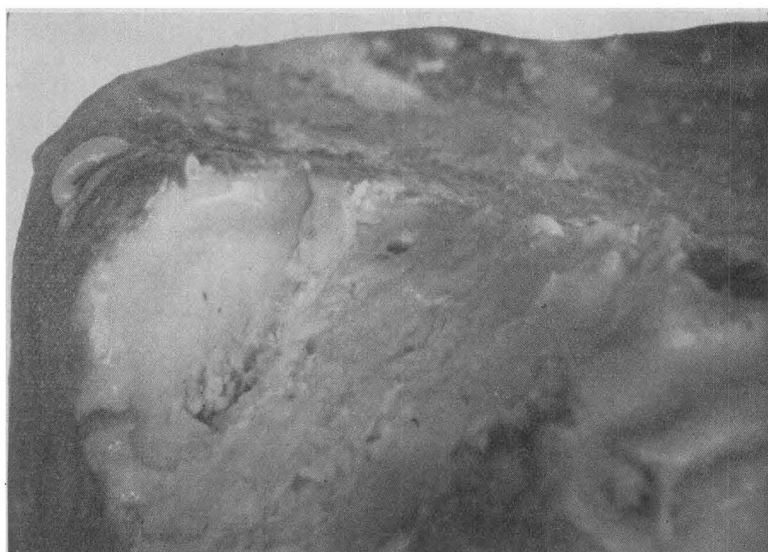


Fig. 8. Blue cheese showing deep penetration of soft edge defect.

cheese in the curing room was changed, certain locations seemed to permit a partial recovery.

Figures 7 and 8 illustrate the soft edge defect in blue cheese; fig. 8 shows particularly the depth to which the defect may penetrate.

EXPERIMENTAL

Since the general observations indicated that the moisture conditions in certain locations in the curing room are closely related to the development of the soft edge defect in blue cheese, attempts were made to reproduce the defect under experimental conditions.

TRIAL 1

Two 2.5-pound cheese, which were normal except for size, were placed in the curing room, one near the humidifier and the other as far as possible from any source of free moisture. After 1 month the cheese near the humidifier had a more luxuriant slime formation than the control cheese. The edges of both cheese were normally firm. After 6 weeks the cheese near the humidifier had developed soft edges, whereas the control cheese had not; when the cheese had ripened 2 months the former showed the soft edge defect in an advanced stage. The edges of the cheese had broken away in spots revealing smooth, creamy material having a limburger cheese odor. In contrast, the control cheese was firm and appeared to be ripening normally. The cheese near the humidifier was moved to a position near the control cheese and held another month. This resulted in a general firming of the outside of the cheese. At this time both cheese were cut. The mold growth in them was good, but neither had developed a satisfactory flavor. The body of the soft edge cheese was not as firm as that of the control, probably indicating a higher moisture content. In the defective cheese there was a distinct division between the soft edge and the normal cheese. The defect had penetrated to a depth of about 0.75 inch.

TRIAL 2

After salting and punching three normal-size cheese, a part of each cheese was covered with dry absorbent cotton, and the cheese were placed in different parts of the curing room. The positions chosen represented the average ripening conditions in the room. The cheese were examined at 2-week intervals. The exposed por-

tion of each cheese developed a normal slime, whereas the covered portion was slow in developing slime, and the amount was less than usual. The absorbent cotton remained dry for about 1 month, after which a gradual moistening took place. When the cheese had ripened 2 months the portions covered with absorbent cotton had developed soft edge. With all three cheese the soft edges had a tendency to penetrate deeper than usual. The defect was not confined entirely to the edges of the cheese but had a tendency to spread back from the edges and follow the outline of the cotton. The consistency of the soft edge material was characteristic of the defect, but it had not developed an off odor. When the cheese were finally cut, the normal portions showed good mold growth and flavor development.

TRIAL 3

During the making of two normal-size cheese, a 2-inch cube of raw potato was placed in the curd used for one of them in such a position that it was near the center of the resulting cheese. After salting and punching, the cheese containing the potato (the experimental cheese) was partly covered with pliofilm, while the other cheese (the control cheese) was not. In the curing room the cheese were placed near a humidifier. After 6 weeks the unprotected portion of the experimental cheese and the control cheese had developed soft edge. The pliofilm was not removed from the experimental cheese, but the cheese under it appeared normal and was firm to the touch. After 2.5 months the unprotected portion of the experimental cheese and the control cheese showed the soft edge defect in an advanced stage. The defect had spread back from the edge, and the soft material had a limburger cheese odor. The cheese under the pliofilm still was normal. When cut, both cheese had normal mold growth and good flavor. The soft edge defect had penetrated about 1 inch. There was a sharp dividing line between the normal and defective cheese. The half of the experimental cheese left unprotected appeared the same as the control cheese, while the protected half was normal in all respects.

When the cheese containing the potato was cut, there was a cavity in it about the size of the original piece of potato. In this cavity a shrunken, brown mass was all that remained of the potato. Evidently, the cheese had dehydrated it. The results show how readily cheese takes up moisture.

TRIAL 4

After salting and punching three normal-size cheese, they were placed in a refrigerated room to dry. Three days later half of each was paraffined. The cheese were then placed near the humidifier in the curing room. At intervals the cheese were turned in the rack so that all parts of each cheese would be subjected to the same general conditions, especially exposure to free moisture. After 2 months the unparaffined portion of each cheese had developed soft edge, while the paraffined portion was firm to the touch. The cheese were cut 2 weeks later, and the unprotected half of each cheese showed a penetration of the soft edge defect of 0.25 to 0.75 inch; the mold growth was normal, and the cheese had developed good flavor. The cut surface of the protected portion of each cheese showed that there had been no softening under the paraffin. On removing the paraffin, the surfaces of the cheese appeared to be the same as when they were covered. The mold growth in the protected parts of the three cheese varied. In two cheese the texture was fairly close, and there was little mold growth; the flavor was lacking. In the third cheese, however, the texture was more open, and no difference in mold growth and flavor could be detected between the paraffined and the unparaffined portions.

TRIAL 5

A normal cheese was used to study the possibility of hygroscopic materials causing the soft edge defect. After salting and punching, the cheese was placed on a board in the center of the curing room. Two aluminum rings, 0.5 inch deep and 2 inches in diameter, were used to keep the hygroscopic materials in definite areas. One ring was filled with calcium chloride and the other with sodium chloride. As these salts took up moisture and in turn were taken up by the cheese, additional materials were placed in the rings. It was necessary to replace the calcium chloride more often than the sodium chloride. The trial was continued for 2 months, and although there was free moisture in the aluminum rings most of the time, the cheese did not develop soft edges. Apparently, the hardening effect of the salts was greater than the softening effect of the moisture.

COMPARATIVE ANALYSES OF NORMAL AND DEFECTIVE CHEESE

Five experimental cheese, representing different trials in which the soft edge defect had been developed, were

TABLE 1. COMPARATIVE ANALYSES* OF NORMAL BLUE CHEESE AND MATERIAL FROM A SOFT EDGE

Percent moisture		Percent NaCl		pH		Bacteria, millions per gram					
Normal cheese	Soft edge	Normal cheese	Soft edge	Normal cheese	Soft edge	Normal cheese			Soft edge		
						Total	Proteolytic	Lipolytic	Total	Proteolytic	Lipolytic
43.50	52.37	4.44	2.89	6.91	7.23	1,240	1.2	0.0**	3,616	4.1	0.0
35.16	52.07	3.70	3.16	6.09	7.32	106	0.0	0.0	1,910	0.0	0.0
45.21	53.40	4.68	3.28	5.46	7.23	5	0.0	0.0	128	0.0	0.0
39.37	51.32	3.85	2.56	6.00	6.84	61	0.0	0.0	1,380	0.1	0.0
37.58	50.98	3.38	2.49	6.13	6.78	9	0.0	0.0	880	0.0	0.0

* Values given are the averages of duplicates.
 ** No colonies on the dilutions poured

analyzed to compare the normal cheese with the soft edge material. The comparison included moisture and salt contents, pH and counts of total, proteolytic and lipolytic bacteria. The soft material was very soft and represented the defect in an extreme stage. The data are presented in table 1.

The moisture contents in different parts of a normal cheese are known to vary. The general tendency is for the outer portions to be the lower in moisture. The analyses show that in every case there was a significantly higher moisture content in the soft edge than in the normal portion of the same cheese, the differences varying from 8.19 to 16.91 percent. The unusual relationship suggests that the outer portion of the cheese had taken up and retained considerable moisture.

Variations in salt content are noted in comparing different portions of the same cheese or different cheese from a lot. The variation in a cheese is greatest during the salting period, when the salt content of the outer portion is relatively high, and the content tends to become more uniform as the cheese ages because of the diffusion. In the analyses, the soft material regularly had a lower salt content than the normal cheese, the differences ranging from 0.54 to 1.55 percent. The lower salt content in the soft edge presumably was related to the taking up of moisture by this material.

The data show that the soft edge regularly had a higher pH than the normal cheese, the differences ranging from 0.32 to 1.77 pH units. There was considerable variation in pH between the different normal cheese and also between the different soft edges.

The numbers of bacteria in different parts of a cheese normally vary. With a decrease in salt content, a higher bacterial count would be expected, and this is shown by the data on total bacterial counts. A direct microscopic examination not only confirmed the difference in numbers between the soft material and normal cheese but also showed that the soft cheese contained a variety of bacteria, whereas the normal cheese contained largely cocci. The great variety of organisms in the soft material suggested the many morphologic types commonly found in slime of normal cheese.

The softening of a cheese and the production of an objectionable odor suggest a protein breakdown. The analyses do not show significantly more proteolytic or lipolytic bacteria in the soft edge than in the normal cheese.

If the air were saturated with moisture and fresh cheese were brought into the cave, air currents could carry moisture from the fresh cheese to the older cold cheese.

LACK OF MOLD GROWTH IN BLUE CHEESE

Lack of mold growth occasionally is noted in blue cheese. In certain cases it probably is caused by too short a ripening period. Presumably, this cause applies with some of the foreign cheese, especially when there is a marked increase in demand, which results in the sale of relatively young cheese, or a shipping deadline. With further ripening such cheese commonly develops normal mold growth and flavor. In other cases cheese lacks mold growth even after extended ripening. Undoubtedly, there are various causes for this, such as unsatisfactory manufacturing methods, inadequate curing facilities or use of a strain of mold which does not develop properly in the cheese.

GENERAL OBSERVATIONS

An opportunity to investigate a lack of mold growth in blue cheese occurred when a commercial cheese plant experienced an outbreak of this defect. The outbreak occurred suddenly. High-quality cheese was produced up to and including the lot manufactured Dec. 24, 1938. Cheese manufactured on and after Dec. 25, 1938, did not develop normal mold growth or flavor. From the daily plant records it was noted that a fresh supply of mold powder had been obtained from a laboratory on Dec. 24, 1938. It was used to make cheese the following day and thereafter at the rate of about three times a week. The cheese were handled in the usual manner and, on the basis of slime formation, appeared to be ripening normally; the surfaces developed good slime and often showed considerable mold growth also. When the first few lots of cheese made with the new mold powder were about 2 months old, they were examined for mold growth by plugging two or three cheese in each lot. The plugs showed very little mold growth. However, since the outside appearance of the cheese was satisfactory, the plant operator concluded that the mold growth was slow and that it would develop later. The manufacture of cheese was continued. About 1 month later another routine examination showed additional cheese lacking in mold growth. The cheese previously plugged were re-examined, and the plugs showed no improvement in mold growth.

A systematic investigation, in which several cheese in each lot were plugged and at least one cheese was cut, showed that normal mold growth had not developed in any of the cheese made after Dec. 24, 1938. Certain of the cut cheese showed small areas of normal growth, while others were perfectly white. In some cases it appeared that the mold had invaded the cheese through punch holes. From the observations the plant operator concluded that the lack of mold growth was due to defective mold powder and discontinued the manufacture of cheese until a new supply of powder could be obtained. When such a supply was received, several lots of cheese were made and kept under observation. The cheese were plugged several times, beginning after 3 weeks and ending after 6 weeks of ripening. Normal mold growth was not observed in any of the plugs.

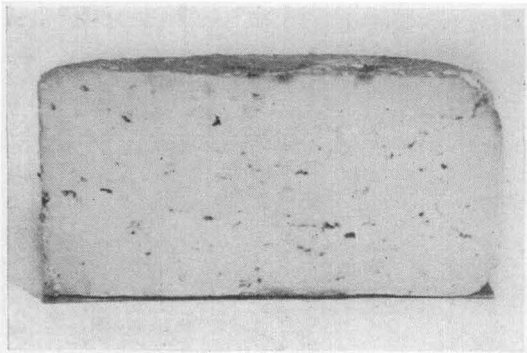


Fig. 9. Blue cheese showing lack of mold growth. Curd inoculated with original defective powder.

The second apparent failure of the mold powder was called to the attention of the laboratory supplying the powder. The laboratory reported that another cheese plant had used a shipment of the same mold powder and had not encountered difficulty in obtaining normal mold

growth in cheese. A new supply of mold powder was sent to replace that which was questioned. Cheese manufactured with this powder developed normal mold growth.

When the defective cheese were from 4 to 6 months old some of them were repunched, the punch being dipped into a water suspension of normal mold spores. This was successful in that many cheese later developed considerable mold growth and flavor. It was estimated that the outbreak involved some 5,000 pounds of cheese and that about 1,000 pounds eventually was marketed.

An experimental cheese, made with the original defective powder and lacking in mold growth, is shown in fig. 9.

EXPERIMENTAL

A sample of the questionable mold powder was obtained from the cheese plant. The powder was somewhat lighter in color than normal. It was plated on Czapek's agar and the plates incubated at 21°C. Mold colonies developed rapidly on this medium and had a wide margin of sterile mycelium. The spores were blue-green in color and spread from the center to the outside of the colony with age. While the colonies of the mold were not typical of *P. roqueforti*, they were near enough to be accepted as one of the species of *Penicillium* used in making blue-veined cheeses. When the questionable mold powder was plated on acidified tomato juice agar under the same conditions, a different type of colony developed. The colonies were more compact, slower growing, white in color and failed to produce a blue-green color even after weeks of growth; they did not resemble colonies of the penicillia commonly used in blue-veined cheeses. Flasks of whole wheat bread were inoculated with the questionable mold powder and incubated at 10°C. The resulting mold growth was blue-green in color and appeared to be identical with that of normal *P. roqueforti*.

TRIAL 1

An attempt was made to reproduce the defect in experimental cheese by inoculating with the original questionable mold powder. A quantity of blue cheese curd, sufficient to make three cheese of 2.5 pounds each, was divided into three portions. To one portion the questionable mold powder was added, to the second portion a mixture of questionable and normal mold powder and to the third portion normal mold powder. The curd was then handled in the usual manner. After salting and punching the cheese, the surface of the cheese made with the questionable powder only was treated with calcium propionate, and the cheese was wrapped in parchment to limit mold contamination from the outside. The three cheese were then placed in the regular curing room. After ripening 2 months the cheese were cut. The cheese made with the questionable powder did not show any mold growth, the cheese made with the mixed powder showed areas of normal mold growth and other areas in which no mold growth could be detected, and the control cheese had normal mold growth. When the cheese had ripened 3 months they were again cut. In the cheese made with the questionable powder there was no mold growth or

flavor development. The cheese made with the mixed powder did not show any marked improvement in mold growth, although there was some increase; very little flavor had developed. The control cheese had about the same mold growth as when first examined and had less flavor than is desirable.

TRIAL 2

Trial 2 was a repetition of trial 1 with the exception that the cheese were of normal size and the one made with the questionable powder was not treated with calcium propionate or wrapped in parchment. After ripening 2 months, the cheese were cut. The cheese made with the questionable powder showed an occasional small area of normal mold growth; these areas were near the surface of the cheese and appeared to have been due to an invasion of the cheese through the punch holes. The cheese made with the mixed powder showed a fair but rather irregular growth of mold; it had not developed any appreciable flavor. The control cheese showed a good mold growth and had developed some flavor. After 3 months the cheese were again cut. The cheese made with the questionable powder appeared about the same as at 2 months; however, in the small areas of normal mold growth some flavor had developed. The cheese made with the mixed powder had improved somewhat in mold growth and had developed a fair flavor. The control cheese had about the same mold growth as at 2 months and had developed a fine flavor.

TRIAL 3

Several samples of the original commercial cheese showing a lack of mold growth and several samples of experimental cheese showing the defect were plated in the usual manner on Czapek's and acidified tomato juice agars. Characteristic colonies of the questionable mold were noted on plates poured with each cheese. Some of the colonies were picked and purified by repeated platings. Powder A was prepared with a culture obtained from the commercial cheese and powder B with a culture isolated from the experimental cheese.

Sufficient blue-cheese curd to make three normal-size cheese was divided into three portions. To the first portion powder A was added, to the second portion powder B and to the third portion normal powder. The cheese were handled in the usual manner and after salting and punch-

ing were placed in a curing room. When the cheese had ripened 2 months they were cut. The cheese made with powder A or powder B showed very little mold growth and had no flavor. The control cheese had normal mold growth and had developed considerable flavor. After 3 months the cheese were again cut. The cheese made with powder A or powder B were still lacking in mold growth and flavor. The control cheese was about the same as when previously examined.

IDENTIFICATION OF MOLD IN DEFECTIVE POWDER

A number of mold cultures that had been isolated from the original defective mold powder or from cheese made with it, as well as cultures from the experimental cheese that lacked mold growth, were investigated in more or less detail. Identification studies were carried out with cultures grown on Czapek's agar. Microscopic observations and measurements suggested that the organism was *Penicillium echinatum*, according to the classification of Gilman and Abbott (13), while cultural characteristics suggested that the organism was *P. roqueforti*, according to Biourge (5). The cultural characteristics were accepted rather than the microscopic observations, and the mold was considered to be an atypical *P. roqueforti*.³

DISCUSSION

Several species of the genus *Penicillium* have been used successfully in the ripening of blue cheese, but other species are not satisfactory. Although the organism present in the defective mold powder was regarded as an atypical strain of *P. roqueforti*, it did not develop well in blue cheese. The culture used to prepare the powder had been carried on a synthetic medium for an extended period, and this may have resulted in a variation; however, there remains the possibility of contamination of the culture with an organism which outgrew the original type.

Because of the very close relationship between certain species of the genus *Penicillium*, it might be difficult to detect variation or contamination in a culture used to prepare mold powder. In case there was variation or contamination, the first indication might be failure to produce normal mold growth in cheese.

Although the questionable mold powder caused diffi-

³Dr. J. C. Gilman concurred in the conclusion.

culty in one plant, in another plant it did not. The plant having difficulty received and used several pounds of the powder, while the other plant received only 1 pound. At the plant having no difficulty, the cheese normally had an unusually extensive mold growth, although the rate of inoculation of powder was the same as in plants making cheese with less abundant mold growth. This indicates that the cheese made in the one plant either was receiving a natural inoculation of mold while being manufactured and ripened, or conditions were exceptionally favorable for mold growth in the cheese. The experimental results support the former assumption; cheese inoculated with the questionable mold powder, treated with calcium propionate and wrapped in parchment failed to show normal mold growth, whereas cheese not treated with calcium propionate or wrapped in parchment showed some areas of normal growth. It appears that under certain conditions natural inoculation results in fair mold growth and flavor development in cheese.

In order to avoid slow mold development in blue cheese, the mold powder must be carefully controlled. It would seem advisable to establish the effectiveness of a new lot of powder by using it in one or two runs of cheese before it is employed in more extensive operations. There may be an advantage in occasionally isolating a fresh culture of *P. roqueforti* from a good blue cheese and using it in the preparation of powder.

GRAY DISCOLORATION OF BLUE CHEESE

In blue cheese which has ripened for some time, gray discoloration is a rather common defect. At first the color varies only slightly from normal, but it darkens as the cheese is held until a gray brown is reached. The defect is particularly serious, because when once started it tends to spread through the entire cheese.

GENERAL OBSERVATIONS

Gray discoloration of blue cheese commonly can be noted on the surface of the cheese when the slime is removed by scraping or washing. In the early stages there may be only a few discolored areas, while in the extreme condition the entire surface is involved. When defective cheese are cut they usually show normal mold growth. The defect appears to originate on the surface and then spread through the cheese. Because of this the discoloration evident on a cut surface varies from a few small areas

to the entire surface. In most cases extensive discoloration is accompanied by a mousy, ammoniacal flavor which has a tendency to become soapy with age; the off flavor does not appear until the cheese are several months old. The defect seems to be confined to certain lots of cheese and may or may not involve all the cheese in a lot. Apparently, there is no tendency for the defect to spread from one cheese to another.



Fig. 10. Blue cheese showing extreme gray discoloration.

Figure 10 shows extreme gray discoloration in a blue cheese.

HISTORICAL

Discolorations in cheese have been noted by various investigators.

Golding (14) investigated color defects in stilton cheese. The cheese first turned yellow but upon aging, especially if the cheese were cut, turned red and even black. Under practical conditions the use of relatively large amounts of salt on the curd tended to favor the defect. Oxygen had a tendency to increase the darkening, which suggested that an oxidase might be responsible. This was further substantiated when heated cheese failed to turn yellow, while chloroform did not stop the change. The addition of a solution of tyrosine to the cheese caused it to turn dark; from this, Golding concluded that under certain conditions tyrosine may be a limiting factor.

Cornish and Williams (10), working on discoloration in stilton cheese, isolated many types of microorganisms and studied two groups. The first group, identified as *Bacillus proteus vulgaris*, produced a brown color, both in solutions of tryptophane and in tryptophane agar; in tyrosine media, however, it produced only a slight dis-

coloration. The second group, composed of gram negative, alkali-producing bacilli, gave a slight discoloration in tryptophane media and turned tyrosine media a dark brown or black. Further studies were carried out by Venn (33) and by Mattick and Williams (23). Venn used the gram negative, alkali-producing bacilli isolated by Cornish and Williams in studying the effect of pH on color production in tyrosine media and found that color was produced over a wide pH range. The color was deep brown or reddish brown from pH 5.83 to pH 9.47. Mattick and Williams reported that in tryptophane solutions *Bacillus proteus vulgaris* isolated by Cornish and Williams produced a color ranging from deep orange to light yellow at pH values from 8.95 to 9.41.

Discoloration in cheddar cheese was investigated biochemically by Moir (24) and bacteriologically by Morgan (26). Moir found that the pH of the muddy areas was much higher than that of normal areas in the same cheese. From this and other studies he reported that the muddy discoloration appears to be produced by enzymes, possibly including tyrosinase, which diffuse into the cheese from the centers of mold growth and act on substances present in mature cheese. Morgan noted that the muddy discoloration occurred near cracks or openings in the cheese and concluded that it was caused by growth of mold.

Skinner (29) reported that nearly one-third of the hundreds of strains of *Actinomyces* he had isolated were capable of producing a dark color and noted that cultures studied by other investigators were much the same in this respect.

Clark and Smith (8) found that *Bacillus niger* produced a black pigment in protein media which contained metabolically available tyrosine. *Bacillus atterimus* blackened media containing fermentable carbohydrates, either in the presence or absence of tyrosine, but did not blacken sugar-free peptone media which were readily blackened by *Bacillus niger*. Carbohydrate media containing mineral nitrogen were also blackened by *Bacillus atterimus* but not by *Bacillus niger*.

Dark discoloration of cheese due to metals has been reported by a number of investigators. Hood and White (18) found that light brown to yellowish brown areas in cheddar cheese were caused by fragments of the steel

wool used to clean the vats. Leitch (20) noted that black discoloration in cheddar cheese was due to lead sulfide, while a gray-black discoloration was due to iron sulfide formed under neutral or alkaline conditions. Davies (11) found traces of tin in darkened areas of cheddar cheese but considered the lead constituent of the solder to be responsible for the color defect. Barnicoat (4) added various metals to cheddar cheese milk at the rate of 3 to 7 parts per million. The discolorations noted with copper and iron were considered to be due to the atmospheric oxidation of a colorless metal:protein complex, while the discoloration with lead was due to sulfides.

EXPERIMENTAL

Several investigators have concluded that discolorations in cheeses were due to melanins produced by the action of tyrosinase upon tyrosine. This suggested that perhaps there are microorganisms present in gray discolored cheese which are capable of producing the defect.

The surfaces and interiors of several blue cheese showing gray discoloration were plated on Czapek's, tomato juice, acidified tomato juice and beef infusion agars. The plates were divided into three lots and incubated at 10°, 21° and 37°C. When the plates were well developed, they showed a variety of colony types which included bacteria, yeasts and molds. The total numbers of microorganisms in the cheese differed greatly, and there was a marked variation in the flora of the different cheese, with *P. roqueforti* the only conspicuous microorganism consistently present. Representative colonies from each plate were picked into a medium consisting of skim milk saturated with *l* tyrosine. The cultures were divided into two lots and incubated at 10° and 21°C. Observations were made each week to determine whether any of the cultures were capable of darkening the medium. An occasional culture produced a slight discoloration, but the discolorations were not considered significant, and after 1 month the cultures were discarded. Additional samples of blue cheese gave similar results.

The work of Skinner (29) indicates the possibility of *Actinomyces* species being involved in the discoloration of blue cheese. Since blue cheese is usually made from raw milk, several samples of such milk were plated on beef infusion agar; some of the plates were incubated at 21°C. and others at 37°C. *Actinomyces* colonies were

noted on plates from an occasional sample of milk and were of two types; one type produced a brown discoloration in the medium while the other did not. Colonies of *Actinomyces* were picked on beef infusion agar slopes, and when a number of cultures had been collected they were streaked on plates of beef infusion agar to which had been added 5 ml. of the skim milk tyrosine medium per plate. Tubes of the skim milk tyrosine medium also were inoculated with the organisms. Some of the cultures were incubated at 10°C. and others at 21°C. The cultures of *Actinomyces* which did not produce a discoloration in the beef infusion agar failed to produce a discoloration in the tyrosine media, and all cultures producing a discoloration in beef infusion agar produced a much darker discoloration in the tyrosine media. On plates the discoloration extended beyond the limits of the colonies. In the skim milk tyrosine medium, the color ranged from a black at the surface to a dark gray at the bottom. The dark gray discoloration was similar to that in a defective blue cheese.

Since the gray discoloration in cheese appears to originate at the surface and progress into the cheese, an attempt was made to reproduce the defect by inoculating pieces of cheese with pigment-producing *Actinomyces* cultures. Portions from the surfaces of several normal cheese were placed in wide-mouth glass containers and inoculated heavily with the cultures. The containers were held in a refrigerator comparable to a curing room. After 2 months no growth of the *Actinomyces* cultures could be detected, and the cheese showed no discoloration.

The failure of the *Actinomyces* cultures to grow and produce gray discoloration in cheese suggests that conditions for growth in the cheese may not have been satisfactory. Since the organisms grew on agar and in milk of approximately the same pH as that found at the surface of a cheese, it appears that from this standpoint cheese was a favorable medium. The high salt content of the cheese, however, might be a factor in preventing the growth. Several plates were prepared, using in each beef infusion agar plus 5 ml. of the skim milk tyrosine medium to which had been added varying amounts of salt. The medium in the plates contained approximately 0.00, 0.01, 0.1, 2.0, 3.5 and 7.0 percent salt. The plates were streaked with pigment-producing *Actinomyces* cultures, incubated at 21°C. and observed each day. The organisms grew on all the plates, and salt concentrations up to and including

TABLE 2. pH DETERMINATIONS ON NORMAL AND GRAY DISCOLORED BLUE CHEESE.

Cheese no.	Normal cheese	Gray discolored cheese
1	6.07	6.38
2	6.11	6.25
3	6.15	7.12
4	6.16	7.18
5	6.09	7.12
6	6.72	7.02
7	6.50	7.03
8	6.15	6.85
9	6.32	6.83
10	6.00	6.65
11	5.25	7.29
12	5.56	7.22
13	5.77	7.28

2.0 percent seemed to accelerate their growth. Higher concentrations definitely limited growth. In all cases the organisms produced a dark discoloration, and this seemed to vary in direct proportion to the amount of salt. The results indicate that the salt in the inoculated cheese did not prevent growth of the *Actinomyces* cultures.

The pH was determined on 13 samples of gray discolored cheese; in each case the pH also was determined on a normal portion of the same cheese. The data are presented in table 2.

The striking thing shown by the data is that in each instance the pH of the discolored cheese was higher than that of the corresponding normal cheese, and in most cases the difference was significant. A variation in pH occurred with both normal and discolored cheese; the pH in the normal cheese varied from 5.25 to 6.72 and in the discolored cheese from 6.25 to 7.29. The greatest difference in pH between normal and discolored portions of the same cheese was 2.04 pH units, and the least difference was only 0.14 unit.

Because of the pH relationship noted, pieces of normal blue cheese in the petri dishes were treated with sodium

TABLE 3. SODIUM CHLORIDE CONTENTS OF NORMAL AND GRAY DISCOLORED BLUE CHEESE FROM THE SAME PLANT.

Normal cheese		Gray discolored cheese	
No.	% NaCl	No.	% NaCl
1	4.30	1	5.05
2	3.60	2	4.55
3	3.95	3	6.06
4	4.08	4	5.99
5	3.70	5	5.10
6	3.90	6	5.32
Av.	3.92	Av.	5.34

hydroxide, ammonium hydroxide, trimethylamine and dimethylamine, as well as with hydrochloric, nitric and sulfuric acids. In no case did the cheese darken in color, even after rather extended holding. Apparently, if a high pH is important in gray discoloration, some other factor also is involved.

Golding (14) noted that large amounts of salt favored discoloration in stilton cheese. This suggests that perhaps salt has something to do with the gray discoloration in blue cheese.

Six discolored and six normal blue cheese were analyzed for salt. Each cheese represented a different lot, and all were obtained from the same plant. Table 3 presents the results. The analyses show that in most cases the salt content of the defective cheese was considerably higher than of the normal cheese. There was a variation in the salt content of both the normal and the defective cheese. The average salt content of the normal cheese was 3.92 percent, while that of the defective cheese was 5.34 percent.

In an attempt to produce the gray discoloration, four cheese of normal size were prepared, salted in the usual manner and then held in a saturated salt solution for 72 hours. Finally, the cheese were punched and placed in the curing room. After ripening 3 months the cheese were cut. With each cheese the body was firmer than usual, the mold growth was limited, and the flavor was fair, but the salt tended to mask it. The cheese were not discolored in any way and even appeared whiter than normal.

ADDITIONAL OBSERVATIONS

It appears that gray discoloration is most common in blue cheese which has ripened for rather extended periods. It was especially serious in cheese made at the Iowa Agricultural Experiment Station before homogenization of the milk became a regular procedure; such cheese required extended ripening because of the slow flavor development, and it frequently showed gray discoloration and a soapy flavor. The defect also has been noted in imported cheese which has the general appearance of having been held for long periods.

DISCUSSION

The ripening of blue cheese involves a protein breakdown. Normally this does not proceed to a point where the resulting products affect the flavor of the cheese in an

objectionable way. In cheese showing extensive gray discoloration, a mousy, ammoniacal odor commonly developed, and the cheese tended to become soapy with age. This suggests a relatively large change in reaction, which is further indicated by the pH measurements on the cheese. Presumably, certain lower disintegration products of proteins which are basic in character are involved in the reaction change.

The variation in the normal ripening mechanism which yields cheese showing gray discoloration may be due to extensive growth of *P. roqueforti* since this organism actively attacks milk protein. Another possibility is that contaminating organisms are involved, although none that would produce the defect could be isolated. Certain *Actinomyces* cultures darkened tyrosine media, but they apparently failed to grow on cheese.

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