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MILK FERMENTATIONS

AND

THEIR RELATIONS TO DAIRYING.

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U. S. DEPARTMENT OF AGRICULTURE,
OFFICE OF EXPERIMENT STATIONS,
Washington, D. C., June 15, 1892.

SIR: A popular summary of Prof. H. W. Conn's review of the investigations on the fermentations of milk, which was originally issued as Experiment Station Bulletin No. 9 of this Office, is herewith transmitted for publication as a Farmers' Bulletin. The practical importance of the results of recent bacteriological investigations of milk and its products to an intelligent practice of improved methods of dairying, calls for the wide distribution of a condensed statement of the outcome of these researches among dairymen and farmers.

Respectfully,

A. W. HARRIS,
Director.

Hon. J. M. RUSK,
Secretary of Agriculture.

CONTENTS.

	Page.
Composition of milk	5
Causes of fermentation	6
Fermentation of milk by rennet	7
The souring of milk	8
Number of bacteria in milk	8
Relation of electricity to the souring of milk	9
Other forms of fermentation of milk	10
Alkaline fermentation of milk	10
Butyric acid	11
Bitter milk	12
Alkaline curdling of milk	12
Blue milk	13
Alcoholic fermentation of milk	14
Slimy fermentation	14
Miscellaneous forms of fermentation	15
Practical bearing of the subject upon dairying	15
Handling milk	15
Butter-making	18
Cheese-making	21

MILK FERMENTATIONS AND THEIR RELATIONS TO DAIRYING.

In the following pages the word fermentation will be used in its broadest sense. It will include not only the fermentations produced by yeast and rennet, but also all of the numerous destructive changes to which milk is subject, and will cover, therefore, all of the changes which occur in milk, such as curdling, souring, and putrefaction.

COMPOSITION OF MILK.

It will be necessary at the outset to notice briefly the chemical composition of milk, since upon this are based all of its fermentative changes. While the composition of samples of milk obtained from different cows and produced under different conditions may show wide variation, a fair average composition may be given as follows: Water, 87 per cent, and solids, 13 per cent. The solids include fat, 3.6; casein, 3.3; albumen, 0.7; milk sugar, 4.7, and ash, 0.7 per cent. The casein and albumen are the materials containing nitrogen and are of special importance in cheese-making. In general the ash, sugar, and albumen are in solution, the casein in partial solution, and the fat in suspension, being mixed with the milk, but not dissolved in it. Milk when freshly drawn from the cow is a thin liquid, but after standing for a short time becomes slightly viscous or ropy through the formation in it of a small amount of fibrin.

Milk fat consists of a mixture of several fats. As its composition begins to undergo changes almost immediately after the milk is drawn, its exact condition at any moment is very uncertain. It is distributed throughout the milk in the form of minute globules varying in size. The belief that there is an albuminous membrane around the globules which keeps them from combining readily has been abandoned. The whiteness of milk has usually been attributed to the presence of these globules of fat, but is probably due in part to the phosphate of lime which milk contains.

In regard to the nature of the *casein* of milk there has been much discussion. In the first place, it seems certain that casein does not exist in milk in a state of complete solution, but rather in the form of finely divided particles. Probably some of it is in actual solution, while the greater part is in a state of suspension. When milk is filtered through porcelain a clear liquid called milk serum passes

through the filter, the casein and fat being left behind. In this serum there is a certain amount of albumen and usually part of the casein also, which has been in solution in the milk; but the fact that most of the casein does not pass the filter shows that it is not in a state of complete solution. The relation of the soluble and insoluble portions of the casein is a matter of much importance in the study of milk fermentations. The coagulation of the casein, or curdling, as the process is commonly called, is readily effected by rennet and also by various chemicals, among which are lead acetate, sulphate of copper, alum, corrosive sublimate, tannic acid, sulphate of magnesium, and the mineral acids, but is not effected by boiling.

The *albumen* of milk is in complete solution and seems to differ only slightly from the serum albumen of the blood. Considering the intimate relation which the milk while in the mammary gland must have with the blood, or rather with the lymph, *i. e.*, the blood deprived of its red corpuscles, it is not surprising to find one of the constituents of milk so similar to this albumen of the blood. We may probably look upon it as derived directly from the albumen of the lymph. The presence of this serum albumen is perhaps an important factor in explaining the changes which take place in milk upon standing.

Milk sugar exists in milk in a state of complete solution. It may be obtained by evaporating and crystallizing whey.

The *ash* of milk contains potash, soda, lime, iron, and magnesia, in combination with muriatic, phosphoric, and sulphuric acids, and other constituents. These are all in solution except, perhaps, some of the lime compounds.

Besides the above-mentioned constituents, minute quantities of several other compounds are found in milk, but these have not yet entered into the study of milk fermentations and at present may be neglected. Milk is a very complex body, and a complete study of its fermentations would, of course, take into consideration all of its constituents. At present, however, our knowledge is confined to the study of the fermentations as affecting milk sugar and the casein, albumen, and fat of milk.

CAUSES OF FERMENTATION.

The organisms and substances concerned in the fermentation of milk may be divided into two distinct classes, namely, organized and unorganized ferments. The former include the minute living organisms (microorganisms), such as bacteria, yeasts, etc., which by their growth cause changes or fermentation.

The unorganized or chemical ferments, on the other hand, are substances devoid of life which are capable of causing certain chemical changes in other substances without themselves being changed. Rennet and pepsin are familiar examples of unorganized ferments.

Bacteria proper, which have most to do with milk and cream, are

found in immense numbers everywhere, and play an important part in nature. They are all extremely minute. In shape they show three chief varieties, which may be compared, respectively, to a lead pencil (*bacillus*), a ball (*coccus*), and a corkscrew (*spirillum*). With the highest powers of the microscope they appear as scarcely more than simple dots and lines. They are to be classed with plants rather than animals, in spite of the fact that many of them are endowed with motion.

The isolation and cultivation of a single kind of bacteria is a matter requiring the greatest care. Although imperfectly studied as yet, many different forms are known which are distinguished by their habits of growth the substances in which they thrive, and the changes which they produce in various substances as a result of their growth. Bacteria are cultivated in beef broth, gelatin, and other substances which, when used for these purposes, are called *cultures*. What is known as a *pure culture* contains only one kind of bacteria.

Yeasts are also plants of a lower order, which grow very rapidly in certain substances and thus cause changes which are commonly called fermentations. The most common kind of yeast is that used in making beer and raising bread.

FERMENTATION OF MILK BY RENNET.

Rennet is a preparation usually made from the stomach of a calf, and has the power of coagulating the casein of milk in a very short time. This curdling of milk by rennet is the only form of fermentation of milk known which is produced by an unorganized ferment.

Many investigations have been made to determine how rennet acts on milk and causes it to curdle. The results are certainly not yet very conclusive nor very satisfactory, but the following general summary may serve to bring together the conclusions which are to be drawn from the facts thus far observed. Casein appears to be kept in partial solution by the alkaline condition of the milk, for it is easily separated from the solution by the presence of a small quantity of acid; but when thus separated it seems to be simply thrown from its solution without being altered in its nature, while the active principle of rennet has a very different effect upon it. Under the action of rennet the casein is chemically changed. It is broken up into two nitrogenous bodies (proteids), one of which is easily coagulated, while the other is coagulated only with great difficulty. The former is readily thrown from its solution by salts of calcium (lime), and since these are always present in the milk, the result of rennet action is always to throw down the curd. This portion of the original casein is then manufactured into cheese, while the other portion, being soluble, goes into the whey and is lost to the cheese maker. The amount of protein thus lost may be still further increased through the action of bacteria, which have the power of making even curdled casein soluble, and this fact teaches the advisability of using rennet in a manner which will produce the coagulation

as quickly as possible. The rapidity of the action will depend upon the relative amount of rennet and the temperature, and may be lessened by alkalies and increased by various salts. The active principle of rennet is a chemical ferment which is distinct from the other digestive ferments in the stomach juices. It seems to be somewhat widely distributed in nature among animals and plants, and it is a common product of bacteria growth. It is destroyed by a temperature of 158° F., and it acts best at about 95° F. It is undoubtedly to be regarded as one of the digestive ferments.

THE SOURING OF MILK.

The normal souring of milk has until quite recently been regarded as a characteristic of milk itself, unassisted by any outside influences. To-day, however, there is such a uniformity of results on the part of all experimenters that it is no longer possible to question the fact that the souring of milk is a fermentative process produced by organisms which get into the milk after the milking is done.

After much investigation bearing upon the subject had been carried on by Pasteur and others, Lister, about 1873, found in milk several forms of bacteria which at first he thought were all of one species, but afterwards found to belong to independent species. It appeared, moreover, that while there is one species of bacteria which produce the lactic acid accompanying the souring of milk, there are others which have different effects. The lactic organism, he determined, is common around the dairy, but not common elsewhere in nature, not even in the barn. He found that sterilized milk, if exposed to the air in different places, in his laboratory, in a barn, or in the open air, or if inoculated with water would ferment after a time, but would not sour; in fact, the souring of milk was found to be rare except in milk which had come directly from the dairy. This somewhat surprising observation has been confirmed, and hence the conclusion is forced upon us that the lactic organism is peculiar to the dairy, but is not especially abundant elsewhere in nature. Careful experimenters now have no difficulty in obtaining milk free from bacteria.

Later investigations have shown that there are several kinds of bacteria which may cause the souring of milk. It is doubtful, however, whether any two of the organisms of this class act on milk in precisely the same way. It is probable that the decomposition of milk may take place in a number of different ways. While we are certain that the fermentation of milk, commonly known as souring, is caused by bacteria, we have yet much to learn regarding the details of the process.

NUMBER OF BACTERIA IN MILK.

These organisms all get into the milk from external sources, such as the air, the hands of the milker, the hair or udder of the cow, and especially from the vessels into which the milk is drawn. It is plain

that the number present in the milk will vary with the cleanliness used in the dairy and barn. If the udder of the cow be carefully cleaned and the milk be drawn into a glass tube which, by heating, has been made free from all living germs, and which can be closed so as to keep from the milk all unfiltered air, it is easy to get milk so free from bacteria that it will remain unaffected for two weeks, even though kept all the time in a warm oven.

The number of bacteria present in milk depends chiefly upon the length of time the milk has been standing and upon the temperature. Estimates based upon milk under different conditions have shown them to be almost innumerable. In one instance a specimen of milk which had been standing for four days in a cold place was found to contain about ten millions of bacteria per quart. The same milk was then allowed to stand in a warm room for seven hours, and during this time the bacteria increased a hundredfold. So far as the practical side of this discovery is concerned it only makes more evident the value of keeping milk as cool as possible from the very outset, if we wish to avoid the troublesome growth of bacteria. For a day or two the bacteria increase with great rapidity, then their multiplication is checked, and finally they entirely cease to grow. This can not, of course, be due to a lack of food, for there is plenty of food in the milk at all times. It is rather to be attributed to the accumulation of the products of their action. Those growths which produce an acid will soon be checked by this, for bacteria can not grow in an acid medium. The amount of acid, however, will vary, for some species of bacteria are very sensitive to acid, while others will endure a larger amount without injury.

RELATION OF ELECTRICITY TO THE SOURING OF MILK.

A consideration of the subject of the souring of milk would not be complete without reference to the effect of electricity. The popular belief that thunderstorms will sour milk is so widespread that it would seem as if there must be some foundation for it. It has been asserted by many that the ozone produced in the air by electricity causes the milk to become sour. In experiments in which electric sparks were discharged over the surface of the milk, however, little or no effect has been produced upon it. The conclusion is that electricity is not of itself capable of souring milk or even of materially hastening the process. Nor can the ozone developed during the thunderstorm be looked upon as of any great importance. It seems probable that the connection between the thunderstorm and the souring of the milk is one of a different character. Bacteria certainly grow most rapidly in the warm, sultry conditions which usually precede a thunderstorm, and it frequently happens that the thunderstorm and the souring occur together, not because the thunder has hastened the souring, but rather because the climatic conditions which have brought the storm have at the same time been such as to cause unusually rapid bacteria growth. Milk

deprived of bacteria will certainly keep well during thunderstorms. Dairymen find no difficulty in keeping milk if it is cooled immediately after being drawn from the cow and is kept cool. Milk submerged in cool water is not affected by thunder. Dairymen find that during "dog-day" weather, even when there is no thunder, it is just as difficult to keep milk as it is during thunderstorms; and they also find that scrupulous cleanliness in regard to the milk vessels is the best possible remedy against souring during a thunderstorm. It is safe to conclude, therefore, that in all cases it is the bacteria which sour the milk, and if there seems to be a casual connection between the thunder and the souring it is an indirect one only. Climatic conditions have hastened bacteria growth and have also brought on the thunderstorm. The same conditions would affect the milk in exactly the same way, even though no thunderstorm were produced, and this effect, our dairymen tell us, is frequently observed during the warm, sultry autumn days.

OTHER FORMS OF FERMENTATION OF MILK.

Students have not recognized until in recent years that a great variety of fermentations may occur in milk. The reason for the tardiness of this discovery is easily seen. Under ordinary conditions milk always undergoes some sort of lactic fermentation (souring). Only under rare conditions is this absent. The production of lactic acid soon curdles the milk and immediately obscures all other forms of fermentation which have occurred during the process. The acid also stops the growth of all bacteria, so that no subsequent effect can be seen. Hence in normal milk clear evidence of fermentation of any other sort than souring is rarely noticed. But the study of bacteria from the time of the earlier investigations of Pasteur has shown the existence of a large number of species of organisms, and though for a time the fact was disputed, it soon became definitely demonstrated. It became evident, therefore, that with a variety in the species of organisms a variety in the fermentations of milk could be expected. Our knowledge of the newly discovered fermentations is by no means complete as yet. We are beginning to recognize that each species probably has its own distinct effect on milk, and the matter thus becomes very complex. Still we are already able to divide these many varieties of the fermentations of milk into classes, each characterized by a single general action, but comprising many varieties. The first class has been noticed, its general characteristic being the production of lactic acid. The second class is characterized by the production of an alkaline reaction instead of an acid.

ALKALINE FERMENTATION OF MILK.

The fermentation of milk is not always accompanied by the production of an acid. It has been ascertained that no lactic acid appears in spontaneously fermenting boiled milk. The milk may become coagu-

lated into a soft, slimy mass, which usually possesses a bitter taste. The taste is never sour, and the milk, instead of having an acid reaction, is either alkaline or neutral. After a day or two the curd begins to dissolve into a somewhat clear liquid, and, if the action is allowed to continue long enough, may become completely dissolved into a semi-transparent liquid having no resemblance to milk. The chemical study of this liquid shows a variety of ingredients, among which are peptones, leucin, tyrosin, and ammonia. To the peptones the bitter taste may be at least partly attributed, and to the ammonia the alkaline reaction.

Many varieties of fermentation are accompanied by the alkaline reaction, but three distinct features of the general class may be conveniently selected for discussion. The formation of butyric acid, the formation of a bitter taste, and the curdling of the milk, with the subsequent dissolving of the curd. are all striking characters which may be considered separately.

BUTYRIC ACID.

As early as 1843 the formation of butyric acid in milk was observed, but it was not until 1861 that Pasteur associated this fermentation with a distinct organism. His cultures of it were not pure ones, however, and his results were not satisfactory to us of to-day. But the work was of the utmost value, since it was a departure in a new direction. Pasteur studied the organism enough to discover the very important fact that it grows best out of contact with air.

In 1880 Prazmowski made a careful study of the organism and named it *Bacillus butyricus*. The species described under this name grows rapidly in the absence of oxygen. Boiling will not kill the germs of this butyric organism, and hence milk after boiling will be pretty sure to undergo the butyric fermentation. Its growth in normal milk is not very great for the first few days, nor, indeed, until the milk sugar has been largely turned into lactic acid by the ordinary lactic organisms. The butyric species will certainly grow in sterilized milk, where there is no possibility of the production of lactic acid, but where the growth seems rather to be due to the presence of the oxygen dissolved in the milk.

After the work of Prazmowski others studied the butyric organisms, and their descriptions, which were found to differ considerably, caused much confusion. More recently other species have been found, and the butyric-acid class seems to be as large as the lactic-acid class of organisms. Some of the butyric organisms produce alkaline reactions, while others render the milk acid. In the ordinary handling of the milk this class of organisms is of little importance, but it has been supposed that they have an important effect upon the keeping qualities of butter. Rancid butter contains considerable quantities of butyric acid, and the development of the rancidity is simultaneous

with the appearance of the butyric acid. When it became known that many species of bacteria produce butyric acid and that there are many bacteria in butter, it was a natural inference that the rancidity is produced by them. But it now appears very doubtful whether bacteria have much to do with the change. Rancidity may be hastened by them, but will occur when they are entirely absent, and is probably of the nature of a direct chemical oxidation closely connected with the agency of sunlight.

BITTER MILK.

Out of the general confusion which has surrounded the subject of butyric fermentation have gradually crystalized some definite ideas in regard to the phenomenon known as bitter milk. The milk of old milch cows is said to have a tendency to be bitter, and various foods which the cow may eat are also thought to have the effect of producing bitter milk. Probably these and other causes may be occasionally at work, but in addition it is certain that microorganisms are frequently the cause of the trouble. At times it is associated with the production of butyric acid, and at other times it is produced by organisms which do not produce butyric acid, but in all cases the bitterness seems to be independent of the acid. Evidently there is quite a number of species which produce this effect. Three have been described with care, and several others incidentally noticed, but we have no means of knowing how numerous they are.

ALKALINE CURDLING OF MILK.

One of the common effects of the alkaline bacteria, though by no means a universal one, is the curdling of the milk. The alkaline reaction of the milk proves, of course, that the curdling can not be due to the formation of an acid as in the ordinary souring. From the investigations thus far made we are forced to the conclusion that certain species of bacteria give rise simultaneously to two distinct forms of fermentation in milk, one producing a rennet-like curdling and the other a digestion or dissolving of the casein. So far as is known to-day, the curdling power is always accompanied by a digesting power, but some instances are known in which bacteria have the digesting power without the curdling property.

The curdling of milk by these organisms is very similar to that produced by rennet. In the ordinary handling of milk the class of organisms included under the head of alkaline ferments is of little importance. They grow slowly and the lactic-acid-forming species usually get the start of them, producing their own marked effect on the milk, so that the action of the alkaline species is entirely obscured. Moreover the acid-forming species soon produce so much acid that the growth of all bacteria is checked, and thus the alkaline species have

no chance to produce much effect on the milk. At the same time these species are of the greatest importance in dairy matters. In the first place many of them form resisting germs which will endure high temperature and render it very difficult to sterilize milk by heat. They are always present in milk which has been standing for a short time, and sometimes their abundance is great enough to produce noticeable effect. Everyone who has had an extended experience with milk has seen instances of milk curdling without the usual acid taste, and it is a familiar fact that curdled milk is by no means constant in character. There is the greatest variety in the stiffness of the curd, the amount of the whey, the taste, odor, etc., and all of these differences are due to varying numbers and species of bacteria other than the lactic acid class.

Among the numerous species of bacteria affecting milk those producing the rennet-like curdling are abundant, and their share in the ordinary fermentations of milk is not a small one, especially in cool weather. In the keeping property of butter they doubtless play a part, though they are not the sole cause of rancidity. In the ripening of cream for churning their part is still greater, and in the ripening of cheese they are of the utmost importance. Undoubtedly we may trace many of the difficulties of the butter and cheese maker to bacteria of this class. A further knowledge of their action will be of great value to the dairy interest. We are as yet only on the threshold of the study of these organisms, for while the lactic-acid organisms have been quite carefully studied in past years, the rennet-forming class has only recently come into notice.

BLUE MILK.

This fermentation, characterized by the deep blue color which has given it its name, occurs sometimes as an isolated trouble in individual dairies, and sometimes it has become so prevalent in certain localities as to be almost an epidemic. The explanation now given for blue milk is a double one. Ordinary milk contains some of the lactic-acid organisms, and these, acting in connection with another species of bacteria known as *Bacillus cyanogenus*, produce the brilliant blue color which characterizes this infection. When growing in ordinary milk the effect of this organism is very marked. For a few hours no change is noticed, but just about the time when the milk begins to become acid some intense blue patches make their appearance. The faster the acid forms the quicker the coagulation appears and the smaller are the blue patches, while if the acid is produced more slowly the blue patches are larger and of a better color.

Where the blue-milk organism comes from is unknown, nor have we any knowledge of the causes of the occasional epidemics of blue milk. There can be little doubt that the cause is always from some unknown source of filth. In some cases the trouble has been traced to a single cow in a large

dairy, and has been easily stopped by isolating the individual found to be the cause, or by carefully washing the cows's teats with a little weak acetic acid solution. Blue milk is always an infection due to outside contamination, and its remedy is always to be found in care and cleanliness. It does not occur in the carefully kept dairy.

Blue milk appears to be harmless. It has been fed to animals, which eat it readily and without harm. Within a few years blue cheese has been brought to the attention of scientists, and has been attributed to the same organism which produces the trouble in milk.

ALCOHOLIC FERMENTATION OF MILK.

Milk does not readily undergo alcoholic fermentation. Koumiss, a beverage prepared from mares' milk from time immemorial by the nomadic tribes of Tartary, contains alcohol produced by alcoholic fermentation of the milk. Within the last few years this drink has become somewhat common as a beverage for invalids and for that purpose is prepared from cows' milk by adding a little sugar to induce alcoholic fermentation. Another alcoholic drink obtained from milk is the "kefir," of the Caucasus. In both of these beverages the alcoholic fermentations of milk are accompanied by various other fermentations. Within recent years it has been discovered that there are numerous organisms capable of producing an alcoholic fermentation of milk sugar when acting alone. Among these are at least two or three species of yeast. It appears that small amounts of alcohol are produced in some of the common forms of lactic fermentation, and finally we have found that almost any species of yeast can produce alcohol from milk if the process be first started by adding to the milk a little of the easily fermentable cane sugar. So well known is this alcoholic fermentation to-day, that it has recently been suggested that the use of whey in the manufacture of alcohol on a large scale would be a profitable undertaking.

SLIMY FERMENTATION.

A slimy fermentation of milk is a somewhat common occurrence and occasionally produces great trouble in dairies. Slimy milk has an important bearing upon the manufacture of Edam cheese. Elsewhere, too, this fermentation is a troublesome one, since it destroys the milk for all ordinary uses. Such milk will furnish no cream. It can not be churned and is ruined for drinking purposes.

There have been the greatest variety of theories as to the cause of slimy milk. Diseases of the mammary gland, variations in the food of the cow, and differences in conditions surrounding the dairy have all come in for a share in the explanation. But the slimy fermentation of milk has been found to be connected with a large variety of organisms. Some of them give to the milk only a slight sliminess, while others render it tenacious almost beyond belief. One, described by Conn,

renders the milk and other solutions so slimy that it can be drawn into threads 10 feet long, and so small as to be hardly visible. Some of the organisms render milk slimy in their early growth, others only after several days, and some do not render the fresh milk slimy at all, but first curdle it and then dissolve the curd into a slimy solution. So far as their chemical side is concerned, the fermentations are also widely different from each other, although not sufficient is known to enable us to classify them all at present.

MISCELLANEOUS FORMS OF FERMENTATION.

In addition to the above well-marked classes of fermentation there are various others not so well known or so carefully studied. Among them are some organisms that produce especially striking effects from the production of pigments. Blue milk has already been noticed, but several other pigment-forming species of bacteria cause milk to turn violet, yellow, green, or red.

Of other miscellaneous forms of fermentation, very little is known beyond the mere fact of their existence. The fact is that milk is an excellent medium for bacteria growth. It furnishes proper food for all of the bacteria connected with decomposition, and the various organisms of the air or the water may grow in it to almost any extent. The study of its fermentative changes resolves itself, therefore, into the study of fermentation in general. Fermentation, decomposition, putrefaction, etc., will all run into each other in the study of the changes occurring in milk, and it is impossible to draw any line separating them. For a complete knowledge of the fermentations of milk, we must wait until we understand thoroughly the process of fermentation and decomposition in general. At present this is an almost unknown field. We can pick out a few of the simpler, more striking types of fermentation and group them into classes as we have done, but we must leave for future study the miscellaneous forms of decomposition and fermentation whose existence we recognize, but of whose nature we are entirely ignorant.

PRACTICAL BEARING OF THE SUBJECT UPON DAIRYING.

After this review of the fermentations of milk, the question of their practical bearing forces itself upon our attention. It is becoming more and more evident every year that their bearing upon dairying is of the utmost importance. The practical application of our knowledge of the fermentations of milk will concern each of the three chief dairy products, milk, butter, and cheese.

HANDLING MILK.

To those dealing with milk itself in any form, the various fermentations are especially undesirable and are constant sources of trouble. Such persons want the milk pure and sweet, and any of the various

forms of fermentation injure the milk for their purposes. Now, so far as these matters are concerned, our study of milk fermentation has taught us first of all *that all fermentations of milk, even the common souring, are due to the contamination of the milk with something from the exterior after it is drawn from the cow.* If they are thus all due to contamination from without, all that is needed to prevent them is to treat the milk in such a way that no such contamination is permitted. But simple as this is in theory our study has shown us that it is a matter of practical impossibility. The various organisms affecting milk are so numerous and so common everywhere that no practical method can be devised for keeping them out of the milk. The person who handles milk must therefore recognize their presence in the milk as inevitable, and he must simply turn his attention to means of reducing them to the smallest number and keeping their growth within the smallest possible compass. This has been shown to be accomplished best by two precautions, absolute cleanliness and low temperatures. The great source of these organisms is in the unclean vessels in which the milk is drawn and in the filth which surrounds the cow. By scrupulous cleanliness in the barn and dairy the number of organisms which get into the milk may be kept comparatively small. The statement of a dairyman that "one should make as careful a toilet for the milking yard as for the supper table" is no exaggeration. Of equal value in preserving milk, is the use of low temperature, and to be of the most use it should be applied *immediately* after the milk is drawn. When drawn from the cow, milk is at a high temperature, and indeed at just the temperature at which most bacteria will grow the most rapidly. Under the influence of the atmospheric temperature, especially in the summer, the milk will become cool very slowly, but never becomes cooler than the air. The bacteria which have gotten into the milk will therefore have the very best opportunity for rapid multiplication and the milk will sour very rapidly. If, however, the milk is cooled to a low temperature immediately after it is drawn, the bacteria growth is checked at once and will not begin again with much rapidity until the milk has become warmed once more. This warming will take place slowly, and therefore the cooled milk will remain sweet many hours longer than that which is not cooled. It frequently happens from this cause that a milkman finds that his morning milk will sour earlier than the milk of the night before. The milk drawn in the evening is put in a cool place at once and becomes quite cool during the night, while the morning's milk is at once put in cans and taken away for delivery. It will thus happen that the older milk will actually keep longer than the newer milk, simply because it has been cooled and must be warmed before bacteria can begin to grow very rapidly. A practical knowledge of this fact will be of great value to every person handling milk. Early cooling to as low a temperature as is practicable is the best remedy for too rapid souring of milk.

A second lesson of no less importance has been taught. All of the *abnormal* fermentations of milk, such as blue milk, red milk, slimy milk, etc., are also due to the growth of organisms in the milk, and *all* of these *may be prevented by care*. While the lactic organisms are so common and so abundant as to make it hopeless to try to keep them out of the milk, this is not true of the organisms producing the abnormal fermentations. These organisms are not so abundant, and by the exercise of care they may all be prevented from getting into the milk in such a way as to cause trouble. If a dairy is suddenly troubled with slimy milk or any other abnormal trouble, the dairyman may feel sure that the cause is to be found in some unusual contamination of his milk and that the remedy must be extra cleanliness. He need not look for the cause in the food that the cow has eaten, but may perhaps find it in the hay which the milker has handled or in the dust which has been stirred up in the milking shed. He must look for the trouble in something apart from the cow, and usually in his own carelessness, either in the barn or the dairy. Search in this direction will usually enable him to remove the trouble, while experiments upon the food or conditions of the cow will usually be worthless. Sometimes such troubles may be traced to an individual cow among a large herd. This will of course indicate that this cow has in some way become contaminated with a source of filth which produces the trouble. We must always remember that with a healthy cow all contamination of the milk must come from the outside, and the remedy is seen at once. Such a cow should be cleaned, and especial care should be taken to carefully wash her teats with a weak solution of acetic acid for the purpose of removing whatever bacteria may be clinging to them. Such methods will soon remove the trouble. The second lesson for the dairyman is, therefore, that *all abnormal fermentations may be prevented by sufficient care and cleanliness*.

It is well to notice that certain abnormal odors and tastes in milk may be produced directly by the food eaten by the cow. If a cow eats garlic or turnip the flavor of the milk is directly affected. Various other foods may in a similar manner affect the taste of milk, but this class of taints may be readily distinguished from those due to bacteria growth. The odors and taints due to the direct influence of the food are at their maximum as soon as the milk is drawn, never increasing afterwards. But the taints due to bacteria growth do not appear at all in the fresh milk, beginning to be noticeable only after the bacteria have had a chance to grow. If, therefore, a dairyman has trouble in his milk, which appears immediately after the milking, he may look for the cause in something that the cow has eaten; but if the trouble appears after a few hours and then grows rapidly worse until it reaches a maximum, he may be assured that the cause is some form of fermentation and that the remedy is to be sought not in changing the food of the cow, but in greater care in the management of the dairy or barn.

Various methods have been devised for destroying the organisms after they have found their way into the milk. Numerous chemicals have been used, and several methods of using heat have been devised. Into the details of this subject we can not go at present. The methods have been devised for the consumer of the milk rather than the dairyman, and the latter need not concern himself with them. The lessons for the dairyman to learn from the study of the fermentations of milk are scrupulous cleanliness in all affairs relating to milk care in the dairy, thorough washing with boiling water of all milk vessels, and low temperatures applied to the milk immediately after it is drawn from the cow.

BUTTER-MAKING.

To the butter-maker the bacteria of milk present a different aspect. To him they prove friends instead of enemies. After the cream is separated from the milk it proves of advantage to the butter-maker to allow bacteria to grow in it before churning. It is the custom of butter-makers to allow their cream to "sour" or "ripen" for a number of hours before churning. This is accomplished by allowing it to stand in a warm place for twelve to twenty-four hours. During this time the bacteria in it are multiplying rapidly and, of course, producing the first stages of the various forms of fermentation of which they are the cause. Prominent among them will be some of the lactic-acid organisms, and these will produce the souring of the cream. But the changes which occur are not confined to the lactic-acid organisms, for the warm temperature will hasten the growth of various other organisms which happen to be present in the cream.

The butter-maker finds certain advantages in such ripening. He finds that the cream will churn more easily and that he can get a larger amount of butter from a given amount of cream if it is ripened than he could if it were churned while fresh. He finds, further—and this is perhaps the chief value of ripening cream—that the butter made from ripened cream has a flavor superior to that of butter made from sweet cream. To obtain the proper flavor or aroma is one of the chief objects of the butter-maker.

Taking up the last matter first, we notice that the aroma is undoubtedly connected with the decomposition products of the bacteria growth. The volatile acids supposed to give flavor to the butter are not present in fresh milk, but only appear after standing, *i. e.*, after the fermentations have begun. For a time it was thought that the aroma of butter was due to some alcohol-like product formed during the ripening or to the presence of lactic acid itself. In accordance with this last idea lactic acid has been used artificially to ripen cream, but without much success. Of course, after we have learned that micro-organisms have been forced to grow in the cream during the ripening, and when we combine this with the facts which we have learned of fermentation

products of micro-organisms, we are led to believe that the ripening of cream is a more complicated process than the simple production of lactic acid. The first person to investigate this matter, in the light of modern discoveries, was Storch, a Swedish scientist. He assumed that the butter aroma was due to the growth of organisms and made a study of the bacteria in butter and cream for the purpose of finding, if possible, the proper species of organism for producing the aroma. After considerable search he finally succeeded in isolating from ripening cream a single bacillus, which seemed to produce the proper butter aroma when it was used in pure culture to ripen cream. Shortly after this Weigmann studied the same phenomenon and also succeeded in obtaining cultures of an organism which produced a normal ripening and gave rise to a proper aroma. This ferment is coming into use in some of the creameries in Germany, the claim being made for it that it insures certainty in the result of the ripening process. It has not yet been introduced into this country for practical purposes.

The value of using such a ferment, if it can be supplied in a practical manner, is easily seen. It will introduce improvements into the creameries similar to those introduced into breweries by means of the study of yeasts. In normal butter-making as practiced to-day there is no way of obtaining any control of the bacteria present in the cream. A given specimen of cream will contain a large variety of bacteria. Conn has shown that there may be a score of different species of bacteria growing in cream which has been collected in the usual way. The butter-maker has no means of regulating this assortment or even of knowing anything about it, but must depend upon what has been brought to him. During the ripening process there will ensue a conflict of the different organisms with each other, and the result will depend upon a variety of circumstances. The result will be influenced by temperature, variety of species, quality of the cream, and length of time of ripening, as well as by the advantage which certain species of organisms may get from an earlier start. In such a conflict it will be a matter of accident if the proper species succeeds in growing rapidly enough to produce its own effect on the cream unhindered by the others. Now it certainly makes a great difference in the product what species of bacteria happen to grow most rapidly. Storch found only a single species that produced the proper aroma, and Conn has found that cream ripened with improper species of bacteria produces very poor butter.

The proper aroma of butter is a very intangible matter for study. It is not due to the volatile acids, as was formerly supposed, for the butter aroma has been found to be produced in solutions containing no butter fat. Evidently this aroma is in some way connected with the first products of decomposition which are set up in the cream as the result of bacteria growth. But these decomposition products are very numerous and not at all desirable. The bacteria which grow in ripening cream have been found to produce all sorts of disagreeable

flavors and tastes in milk or cream if allowed to act unhindered. It seems to be only the first products of the decomposition that have the pleasant flavor, the later stages of the decomposition giving rise to products of a very different character. Too long a ripening results in the production of a butter containing too strong flavors, and one of the difficulties of butter-makers is to determine the right length of time for proper ripening. Indeed, the greatest difficulty which the butter-maker has to meet is in obtaining a uniform product. Proceeding according to rules which his experience has taught him, he can usually obtain a good product; but even the best butter-makers will sometimes fail from causes not explained.

Now, while the trouble is of course not entirely due to difficulties in the ripening, there is no question that this is one of the prominent sources of difficulty. The butter-maker can have no certainty that his cream is supplied with the proper species of bacteria to produce the proper aroma, and can never be sure as to the result of the ripening. If he could be furnished with a ferment for the purpose, as the brewer is furnished with yeast, one of his chief difficulties would be overcome. It is in this direction that experiments are being directed to-day. The bacteriologists have offered the butter-maker in Europe even now a ferment with which to ripen his cream, and by using fresh milk and separating the cream with the centrifugal machine there is nothing to prevent ripening cream with the ferment offered, unhindered by the other organisms which are usually present. But the work as yet is only preliminary. While there has been found a species of bacteria which produces a good result, we do not yet know enough of the effect of the ordinary species of bacteria. We have no knowledge as to whether more than a single species can produce good results, nor do we know whether the species used in creameries in this country and in Europe are one and the same.

The matter of the production of the proper butter aroma as the result of the use of artificial ferments in ripening cream is at present too uncertain for definite conclusions. We may be confident that the flavor of the butter is largely dependent upon the decomposition products of the bacteria that grow in the cream, and we have positive evidence that some organisms will produce much better quality of butter than others. We may hope that the further study of the decomposition products of different organisms and their relation to cream and butter will offer to the butter-maker the solution of this difficult problem in the future. If that occurs we may hope not that the butter-maker will be able to make better butter than the best that is made to-day, but that he will be able to obtain the best product with uniformity; and we may also expect that the creameries which at present make an inferior quality of butter will be able to improve it so as to compete with the best.

As for the other purposes of ripening, it is not possible to say much at present. Evidently the greater ease of churning and the larger

product obtained from ripened cream are matters closely related to each other. The simple fact is that fat is more easily collected into masses of sufficient size to be removed mechanically from the butter-milk; but why the ripening makes them thus more easily collected is not yet fully explained. The difficulty of an explanation lies in the fact that we do not know exactly the condition of the fat in the milk.

The treatment of cream for butter-making needs, therefore, to be very different from the treatment of milk and cream for ordinary purposes. The milkman desires his milk to be as free from microorganisms as possible, but the butter-maker uses them and takes pains to cultivate them, but he wants the proper species if the bacteriologists can furnish them. He desires their action on the albuminoids of the milk, which renders the churning easier for him, and he desires still more the early products of their decomposition, which give him the desirable butter aroma.

CHEESE-MAKING.

If bacteria are desirable allies of the butter-maker, they are absolute necessities to the cheese manufacturer. Without their agency in ripening cream the butter, though it may taste flat, is still usable, but cheese is worthless without them. New cheese is not palatable; it tastes like fresh-milk curd and is not at all pleasant. The proper flavor of cheese appears only as a result of a ripening process which is allowed to continue for several weeks or months, the flavor slowly growing stronger all the while.

The ripening of cheese has been conclusively proved to be a matter of the action of microorganisms. In 1875 Cohn first found bacteria in cheese. But it was Duclaux who first connected the ripening with the growth of these organisms. His first paper in 1877 gave the results of a chemical study of the ripening process and showed that it consists chiefly in the transformation of insoluble casein into soluble albuminoids and that the process is associated with the production of several ferments. Three years later he made a study of the bacteria in such cheese and determined that they are very numerous and comprise several species. In general the process of ripening is quite similar to the digestion by the digestive fluids of the stomach and alimentary canal. At this time Duclaux first suggested that certain of the bacteria produce ferments as the result of their growth similar in their characters to the digestive ferments, a discovery which has been well established by later work.

The ripening of cheese was now studied by others, but the most systematic work was done by Adametz. That ripening is due to bacteria growth was proved by this observer by treating fresh cheese with a disinfecting agent, which would prevent bacteria growth without affecting the chemical condition of the cheese. Under these conditions the cheese did not ripen. He also made quantitative estimates of the num-

ber of organisms present, finding from 25 to 165 millions per ounce, and this number was found to increase slowly during the ripening process. He also tried to determine whether ripening is due to the combined action of many species of organisms or to a single species. For this purpose he studied many specimens, and studied the cheese at intervals during the ripening. He found many species of bacteria present, but as the ripening went on one species was found to increase at the expense of the others and was much more abundant at the close of the ripening than any of the others. This species he always found, while the others were more variable, and hence he concluded that this species is the cause of the ripening. These results have since been confirmed.

At this point the knowledge of the normal ripening of cheese rests at the present time. But few observations have been made in regard to abnormal ripening. The greatest difficulty that the cheese manufacturer has to contend with lies in this direction. He can not be sure of a uniform product. In spite of all precautions his cheese will sometimes undergo abnormal troubles and become worthless by changes taking place during the ripening process. These troubles have been attributed to every sort of difficulty, including health and condition of the cow, the condition of the barn, the food of the cow, etc. In some cases they have actually been traced to filth connected with the management of the cows. Recent experiments have indicated that the direct result is in all cases to be attributed to the action of abnormal species of microorganisms which get into the milk, and hence have a share in the ripening of the cheese. Certain it is that black cheese, bitter cheese, and cheese flecked with red spots are all thus caused, and several other troublesome infections have with certainty been traced to microorganisms. But while abnormal ripening is undoubtedly due to growth of improper species of organisms, we can not at present determine how far the variations in the ripening are due to different species of organisms planted in the curd and how far to different conditions of the ripening. Each, doubtless, has its effect, and much further study is needed in this direction.

It is evident that the presence of bacteria in cheese is inevitable. The milk from which it is made always contains them, and when made into cheese part of the bacteria at least will be inclosed in the cheese. Here they find proper conditions for growth. The conditions are not very favorable, it is true, for the density of the cheese prevents ready access of air, and the organisms which require air for their growth suffer in consequence, except at the surface. The lack of moisture is also doubtless a disadvantage. But in spite of these disadvantages the bacteria grow slowly and soon produce profound chemical changes. They give rise to the peptonizing ferment, which acts upon the casein, rendering it partly soluble. Besides this, they induce numerous other decomposition changes, the total result of which is the

production of the rich, delicately flavored cheese for the market. But though many chemical studies have been made of ripening cheese, we are not in condition at the present time to follow the process beyond stating the few salient facts already mentioned. The cheese-maker thus forces the bacteria to give him products for which he obtains a high price. Of course, so far as the food value of cheese is concerned, it is the casein and the fat which render cheese valuable, but its market price depends not upon the quantity of casein, but upon the flavor, and this flavor is supplied by microorganisms. To a certain extent also it is true that the different flavors of different cheeses are due to the action of different species of organisms in the ripening, although we know little in regard to this matter at the present time.

What the practical application of these results will be in the future, it is impossible to say. We have as yet only learned that there is a causal connection between the ripening and the microorganisms, but the conditions affecting their growth, the variety of species which can produce a normal ripening of cheese, whether different species of organisms will produce differently flavored cheeses, whether the cheeses of the markets are due to different organisms used in the ripening or chiefly to different conditions under which they are grown, are all problems to be settled before any practical results can be expected. But we can confidently predict one result: If we ever succeed in reducing the ripening of cheese to a systematic process and become able to use the proper species of organisms to produce it, we may expect an end of the cases of poisonous cheese of which so many instances are on record. The poisons in these cheeses are due to the growth of mischievous organisms, and will be avoided when we learn to ripen cheese with pure cultures of the proper species of bacteria.

We may then, perhaps, predict a time in the not distant future when both the butter-maker and cheese-maker will make use of fresh milk. The butter-maker will separate the cream by the centrifugal machine in as fresh a condition as possible and will add to the cream an artificial ferment consisting of a pure culture of the proper bacteria, and then ripen his cream in the normal manner. The result will be uniformity. The cheese-maker will in like manner inoculate fresh milk with an artificial ferment, and thus be able to control his product. Perhaps he will have a large variety of such ferments, each of which will produce for him a definite quality of cheese. To the dairy interest, therefore, the bacteriologist holds out the hope of uniformity. The time will come when the butter-maker may always make good butter and the cheese-maker will be able in all cases to obtain exactly the kind of ripening that he desires.

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