OUTLINES OF

Dairy Bacteriology

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OUTLINES
OF
DAIRY BACTERIOLOGY

A Concise Manual for the Use of
Students in Dairying

BY
H. L. RUSSELL
PROFESSOR OF BACTERIOLOGY
University of Wisconsin

FOURTH EDITION, THOROUGHLY REVISED

MADISON, WIS.
H. L. RUSSELL
1899

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PREFACE TO FOURTH EDITION.

The cordial reception with which these Outlines continue to be received shows that the effort to keep them abreast of bacteriological advance is appreciated by students of dairying. An exposition of the principles of bacteriology as applied to dairy problems is now recognized as an essential part of dairy science, for modern dairy practice is rapidly adopting the methods that have been developed through bacteriological study.

Although a thoroughly satisfactory explanation has not as yet been given for all the phenomena connected with milk and its products, still a better understanding of these problems has been rendered possible through the light that has been thrown upon these questions by scientific study from a bacteriological point of view.

A brief glossary is appended, embracing those technical terms that are necessarily employed. My acknowledgments are due to the following parties for the loan of cuts: Wisconsin Agricultural Experiment Station; Western Creamery, San Francisco, Cal.; Cornish, Curtis and Greene, Ft. Atkinson, Wis.; and A. H. Reid and Co., Philadelphia, Pa.

H. L. RUSSELL.

*Wisconsin Dairy School,
Madison, Wis., Dec., 1898.*

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INTRODUCTION.

Bacteriology is the youngest member of the sisterhood of those biological sciences that deal with life in its varied manifestations and functions. While it has a strictly scientific side, this subject has a greater practical bearing than is found in many of its sister sciences. For a long time it was merely a protégé of medicine; even now, the term bacteria is nearly always associated in the minds of many with some dread contagious disease, but with further study, the effect of bacteria in many other practical lines is becoming better known, and the circle of its influence is steadily widening.

Its methods have revolutionized the brewing industries; on the presence of bacteria depends the success or failure of many of the industrial arts, such as butter and cheese-making, many of the operations in tanning, the manufacture of vinegar, of wines, etc. Modern agriculture recognizes the effect of germ life in the various processes of fertilization by natural manures; in the accumulation of nitrogenous food in the soil as a result of the action of nitrification, and in the fixation of free nitrogen of the air by members of the clover family.

Sometimes the bacteria are to us a scourge, but often, indeed, they come in the character of a friend and helper. More especially is this true of that class that are associated with dairy products, for in both the butter and cheese industries, success is only assured through the activities of certain favorable forms.

The following pages attempt to show this group of organisms—so infinitely small yet almost infinitely powerful—in their relation to the dairy and dairy products.

Knowledge in dairying, like all other technical industries has grown mainly out of experience. The facts have been learned by observation, but the *why* of each is frequently shrouded in mystery.

Modern dairying is attempting to build its more accurate knowledge upon a broader and surer foundation, and in doing this is seeking to ascertain the cause of well established processes. In this, bacteriology is destined to play an important rôle. Indeed, it may be safely predicted that future progress in dairying will, to a large extent, depend upon bacteriological research. As Fleischmann, the eminent German dairy scientist, says: "The gradual abolition of uncertainty surrounding dairy manufacture is the present important duty which lies before us, and its solution can only be effected by bacteriology."

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PART I.

GENERAL PRINCIPLES OF BACTERIOLOGY.

CHAPTER I.

STRUCTURE AND FORM.

BEFORE one can gain any intelligent conception of the manner in which bacteria affect dairying, it is first necessary to know something of the life history of these organisms in general, how they live, move, and react toward their environment.

1. What are bacteria? Toadstools, smuts, rusts, and mildews are known to even the casual observer, because they are of such evident size. Their plant-like nature can be readily understood from their general structure and habits of life. The bacteria, however, are so small that, under ordinary conditions, they only become evident to our unaided senses by the by-products of their activity.

When Leeuwenhoek (pronounced Lave-en-hake) in 1675 first discovered these tiny, rapidly moving organisms, he thought they were animals. Indeed, under a microscope, many of them bear a close resemblance to those minute worms found in vinegar that are known as "vinegar eels." The idea that they belonged to the animal kingdom continued to hold ground until after the middle of the present century; but with the improvement in microscopes, a more thorough study of these tiny structures was made possible, and their vegetable nature

demonstrated. The bacteria as a class are separated from the fungi (smuts, molds, etc.), mainly by their method of growth; from the lower green algæ by the absence of chlorophyll, the green coloring matter of vegetable organisms.

2. Structure of bacteria. As far as structure is concerned the bacteria stand on the lowest plane of vegetable life. They are single-celled plants. In its inner structure, the cell does not differ essentially from that of many other types of plant life. It is composed of a protoplasmic body which is surrounded by a thin membrane that separates it from neighboring cells that are alike in form and size.

3. Form and size. Where a plant is composed of a single cell but little difference in form is to be expected. While there are intermediate stages that grade insensibly into one another, the bacteria may be grouped into three main types (so far as form is concerned). These are

FIG. 1.—Different forms of bacteria. *a*, *b*, *c*, represent different types as to form: *a*, coccus, *b*, bacillus, *c*, spirillum; *d*, diplococcus or twin coccus; *e*, staphylococcus or cluster coccus; *f* and *g*, different forms of bacilli, *g* shows internal endospores within cell; *h* and *i*, bacilli with motile organs (cilia).

spherical, elongated, and spiral, and to these different types are given the names, respectively, *coccus*, *bacillus*, and *spirillum* (plural, *cocci*, *bacilli*, *spirilla*), (fig. 1).

A ball, a short rod, and a corkscrew serve as convenient models to illustrate these different forms.

In size, the bacteria as a class are the smallest organisms that are known to exist. Relatively there is considerable difference in size between the different species, yet in absolute amount this is so slight as to require the highest powers of the microscope to detect it. As an average diameter, one-thirty-thousandth of an inch may be taken. If a hundred individual germs could be placed side by side, their total thickness would not equal that of a single sheet of paper upon which this page is printed.

4. Manner of growth. As the cell increases in size as a result of growth, it usually elongates in one direction, and finally a new cell-wall is formed, dividing the so-called mother-cell into two equal-sized daughter-cells. This process of cell division, which is called *fission*, is continued almost indefinitely until growth ceases as a natural sequence.

5. Cell arrangement. If this process of fission goes on in the same plane it results in the formation of a *cell-row*. A species forming such a chain of cells of the coccus type is called *strepto-coccus* (chain-coccus). If the second division plane is formed at right angles to the first, a *cell-surface* is formed. If growth takes place in three dimensions of space, a *cell-mass* is produced as in the *sarcina* group. In some cases these cell aggregates cohere so tenaciously that this character is of value in distinguishing different species.

6. Reproduction. The process of cell division known as fission enables the bacterial form to reproduce itself rapidly. Some species possess another method of vegetative reproduction, viz., spore-formation (fig. 1, *g*). The spores are usually formed within a mother-cell; hence,

they are called *endospores*. The protoplasm of the endospore contracts into a small ball, and finally acquires a thickened outer part that enables it to resist unfavorable surroundings; therefore, they are adapted to the perpetuation of the species under severe conditions, and are analogous in this respect to the seeds of higher plants. Many of the bacilli form spores, the cocci do not. Spores do not germinate and grow as readily as the ordinary cells.

7. Movement. Many bacteria are unable to move about from place to place. They have, however, a vibratory movement known as the *Brownian* motion, that is purely physical. Many other forms possess an ability to move about. This they do by means of fine thread-like processes on the edge of the cell known as *cilia* (sing. *cilium*). Coccus forms in general are non-motile, while bacilli may or may not possess these locomotor organs.

8. Classification. In arranging bacteria according to their various relationships, difficulties are experienced that do not obtain with the higher forms of plants. There exists so little difference between the various species as regards form and size that it becomes necessary to employ other characters of a physiological nature, as for instance the way in which the germ develops in culture media, the by-products that are formed as a result of growth and other characters of a similar nature.

CHAPTER II.

PHYSIOLOGY.

9. Conditions essential for growth. The growth of bacteria, like all other living organisms, bears a direct relation to their external surroundings. Certain conditions are absolutely essential before life can develop. Other conditions though often advantageous are not of such vital importance.

10. 1. Food-supply. Concerning the character of the food-supply necessary for different species there is a very great difference. Many of the disease-producing forms are very particular in the selection of their food. With those forms that are usually found in milk, such delicacy of choice is not common.

A food substance to be available for bacteria must be in solution, as it must pass through the cell-wall by absorption. Bacteria can live upon solid substances, but they form chemical substances that enable them to render soluble the necessary food material.

The essential food elements that must be present are nitrogen, carbon, and oxygen, together with minute quantities of mineral elements. The nitrogen and carbon are more available when in the form of organic compounds than as simple inorganic salts. Albuminous or proteid substances are best adapted for the nitrogen supply, while sugars are available for the carbonaceous part of the food. The nitrogenous element is, however, the most indispensable.

Concentration of medium. If the fluid is too dense, bacteria cannot grow. Condensed milk or syrup is too

concentrated to permit of bacterial growth, but if diluted considerably, rapidly undergoes fermentation. The keeping quality of these products is dependent upon this property.

Chemical reaction of medium. As a rule, bacteria prefer a slightly alkaline to an acid medium, but there are so many exceptions to this rule, that the force of it as a general statement is not very strong. Those organisms that are normal inhabitants of milk are, as a rule, less susceptible to slight variations in the reaction of the food medium than many others.

11. 2. Temperature. A certain degree of heat is absolutely necessary before the spores of bacteria can germinate, just as seed grain will not sprout when the ground is too cold. As the temperature of a fluid increases, the rapidity with which the bacteria multiply also increases for a time. Beyond a certain point, however, a heat rigor sets in that destroys the activity of the protoplasm. There is therefore, an *optimum* or best temperature for growth, and *minimum* and *maximum* points as well, below and above which, development is impossible. These three cardinal growth points vary considerably with different germs.

The temperature limits of growth, i. e., the range between the maximum and minimum points of development, are much wider with bacteria than with almost any other forms of living matter. For this reason, bacteria are more widely distributed than any other class of living organisms.

Some forms thrive at 32° F., while others are able to grow at temperatures approximating 140° F. With the great majority of bacteria, especially those growing in milk, this range is not so great. Most milk bacteria fail to develop at a point below 40°–50° F., while the maximum growth point does not exceed 105°–110° F., the optimum

ranging from 80°–100° F. Parasitic forms living in the animal or human body have a high optimum, usually approximating the blood temperature (98°–100° F.).

12. 3. Gaseous environment. To most forms of life, atmospheric air is a necessity, in order to supply the oxygen used in growth. With the bacteria, the great majority require the free access of air the same as other living organisms, and if denied this, fail to grow. Such bacteria are called *aerobic*. Toward some, however, oxygen acts as a direct poison. Only when they are surrounded with an atmosphere other than air, such as hydrogen or nitrogen gas, can they grow. These forms are called *anaerobic*. All bacteria are not divided sharply into these two classes. While some of them grow strictly under one condition or the other, hence are called *obligate* aerobes or anaerobes, other forms are seemingly indifferent to their gaseous surroundings. To this class, the name of *facultative* or optional aerobe or anaerobe is given, depending upon the relation of the germ to the oxygen supply (fig. 1, *d*, *e*).

Most milk bacteria belong to the aerobic class, but there are quite a number that are able to grow without free oxygen.

13. 4. Moisture. On a dry medium, bacteria can not grow, neither can the spores germinate. A certain amount of moisture is, therefore, necessary before bacterial growth can take place. Organic substances such as vegetable or animal tissue contain sufficient water to permit growth.

14. Rate of growth. The rate of growth of actively vegetating bacteria is in many instances perfectly astounding. With many species under favorable conditions, a single cell will divide in twenty minutes, and each of the daughter-cells will repeat this operation in the same

time. This rapid rate cannot be maintained indefinitely, for the bacteria soon limit their own development by the production of by-products that are unfavorable to their own growth. The sour milk bacillus thrives readily in milk until the lactic acid that is formed by it exceeds a certain amount, then growth ceases. If this acid is neutralized by the addition of chalk, the lactic bacteria will start again to grow, and produce acid to the point of excess. There is a marked difference with different forms in their rate of growth, and the conditions under which development best occurs.

15. Effect of external conditions. Bacteria, even in a vegetating stage, possess a much greater resistance toward external forces than other forms of animal or vegetable life. When they are in a spore stage, this resistance is still further increased. A thorough knowledge of the effect of these external forces is essential, for it is by their action that we are often able to destroy undesirable forms of germ life.

16. Physical forces. 1. *Heat.* The bacteria possess a high power of resistance toward heat; but this relation varies, depending upon the condition in which it is applied, whether it is in a dry or moist state. At some temperatures moist heat, on account of its greater penetrative power, is much more effective than dry in destroying germ life. The temperature at which any form is killed is called its *thermal death point*. For the majority of germs in a spore-free, developing condition, a temperature from 130°–140° F. for ten minutes in a liquid medium is fatal. A shorter exposure necessitates a somewhat higher temperature.

When in the more resistant spore-stage, many of them are able to withstand moist heat in the form of steam (212° F.) from one to three hours. To destroy spores

by dry heat, a temperature varying from 260° – 300° F. is necessary for an hour or more. Germ life may be more rapidly destroyed in super-heated steam under pressure, a temperature of 230° – 240° F. for fifteen to twenty minutes being sufficient to kill most species. Many of the milk bacteria, like the sour milk germ, are very easily destroyed by heat, as they do not form spores. Other forms like the hay and potato bacilli are difficult to eradicate on account of the great resistance that their spores offer toward heat. High temperatures are by far the most efficient means that can be used to render any substance germ-free and are most often employed for this purpose.

2. *Cold.* While the maximum temperature that the different spore-bearing species can stand has been accurately determined in many cases, the minimum temperature has in most instances never been reached. But little reliance is to be placed upon this method in attempting to free any substance completely from bacterial life, although freezing is able to destroy a large percentage. Even an artificial degree of cold as low as -220° F. for a day has been found to be insufficient to destroy some forms, especially when in the spore stage. Bacterial growth is held in abeyance when a liquid is congealed, but development occurs very soon after it is melted.

3. *Desiccation.* Different bacteria behave very differently when subjected to drying. The cholera germ dies in three hours if it is dried, while anthrax retains its virulence unimpaired for decades. Tuberculous sputum withstands drying and is often found to be infectious after the lapse of eight or nine months. Those species that form spores naturally resist desiccation much better than those that do not form these latent structures.

4. *Light.* The influence of light on bacterial growth has not been generally appreciated until within a few years. Exposed to the rays of direct sunlight, many forms are killed in a few hours. Even diffused daylight often exerts a powerful inhibiting effect, if the exposure covers any considerable length of time. Bacterial spores are not as readily destroyed as the vegetative forms.

17. Chemical substances. Different chemical substances exert a powerful toxic action on bacterial life with which they come in contact. Those that destroy or kill bacterial forms are known as *disinfectants*; those that merely inhibit, or retard growth are known as *antiseptics*. All substances possessing disinfecting power must of necessity be antiseptic in their action, but not all antiseptics are disinfectants even when used in strong doses. Most of the disinfectants, under ordinary circumstances, have no use in creamery practice except as preservatives for samples. Of these corrosive sublimate and potassium bichromate are most frequently used. In some countries antiseptics, principally of boracic acid or formalin, are employed to "keep" milk, but in general, their use is prohibited by law.

18. The rôle of bacteria in nature. The great majority of bacteria like animal life, belong to a class whose function it is to disintegrate organic matter and resolve it into its constituent elements. The value of this breaking down process is evident at a glance when we consider what would be the result, if all decay, putrefaction, and decomposition were at once arrested. Not only would the supply of carbonic acid gas be very soon absorbed, stopping the development of chlorophyll-bearing plants, but all nature would be clogged and soon buried under its own *debris*. Dead and dying vegetable and animal

matter, unable to rot and decay would soon bury all under the constantly accumulating mass. Owing to the energies of the lower types of plant and animal life, all of this dead effete matter is slowly consumed. The splitting of these materials into the simpler elements is largely due to the activities of the bacteria. Almost without exception, they prey upon organized matter, absorbing sufficient energy for their maintenance, and at the same time, giving off the unused elements to form new combinations that are again absorbed in different forms.

19. Specialization of bacteria. With the higher plant and animal forms, a certain specialization is in progress. The plant or the animal adapts itself to its environment, and in doing so, each group as a whole acquires certain characteristics that mark it in a greater or lesser degree. Usually, specialization accompanies a complexity of structure, so that the higher developed plants and animals show this characteristic more fully than those that belong to the more primitive types.

In the bacteria is seen a curious anomaly to this rule. Simple in structure as they appear to be, they surpass in functional variability many of the higher forms of plant and animal life. Ubiquitous in their distribution throughout the realm of nature, they have gradually adapted themselves to the different habitats in which they have found themselves, and as a result, more or less well marked groups are to be found. Thus, there is a normal bacterial flora of the mouth, another of the skin, and still another of the intestines. Again, there is a class known collectively as water bacteria, still another that might with propriety be classified as milk bacteria. Those forms that have become habituated to certain conditions, may be said to be *indigenous* or native to that habitat, while the presence of uncommon forms from some other source

would be considered as *adventive* or introduced. Thus, the tubercle bacillus is adventive in milk, even though it may be derived from the udder direct, while the sour milk bacillus is a natural and therefore indigenous organism.

20. Distribution of bacteria. If one classifies bacteria according to the habitat in which they are found, they may be divided into various groups such as soil, air or water forms.

1. *Soil bacteria.* The superficial layers of the soil teem with myriads of forms of bacteria. The amount of food present and other favorable growth conditions enable many forms to thrive luxuriantly. At a depth of a few feet most of them are filtered out, and the conditions are likewise unsuitable for their growth, so in deep layers the soil is practically sterile.

2. *Air bacteria.* The bacteria found in the air are originally from the soil beneath. In the atmosphere they are unable to develop, but exist in a latent condition. Their prevalence in the air is measured by the condition of the soil below and the movement of dust particles. For this reason they are more numerous in summer than in winter; the atmosphere of cities contains a larger number of them than country air. They are very prevalent in ill-ventilated houses and out-buildings, particularly barns and stables, where dust from hay and dried manure particles fill the air.

3. *Water bacteria.* Water when exposed to the air invariably contains a sufficient amount of organic matter to serve as bacterial food. Some of its germ life is derived from dust, or the washings of the land, but many species exist in this element which are not to be found in soil. Stagnant pools rich in organic matter always teem with bacteria, and even running water often has large numbers. The ground water layer is normally free, as

the bacteria are filtered out by passing through the intervening soil layers. As a consequence spring water as it issues from the soil is relatively poor in bacteria, but quickly becomes contaminated after it reaches the surface. Some of the highly infectious diseases, such as cholera and typhoid fever are often transmitted by means of a contaminated water-supply.

21. Saprophytic bacteria. If bacteria are considered from the manner in which they live, they may be divided into two very unequal divisions, known as *saprophytes* and *parasites*. Those belonging to the first class subsist on dead organic matter, while the parasitic forms are able to thrive in living tissues of either vegetable or animal nature. The great majority of different forms belong to the first class, and it is undoubtedly true that this condition is more fundamental than the parasitic method of life. The saprophytes find their food-supply in the vegetable and animal matter that has already ceased to live. They are the organisms concerned in the tearing down processes of nature and their beneficent function in the world about them is in their scavenger character as they make way with the offal and *debris* of organic life.

22. Parasitic bacteria. There is no sharp line separating the parasitic bacteria from those that live on lifeless matter. In all probability these forms that are now able to thrive in living tissue came originally from ancestors of a saprophytic type. Some, as the tubercle bacillus, have gradually adapted themselves to the more restricted parasitic method of growth, just as many parasitic plants have been produced, and as a result of this specialization, they have often lost the power to thrive under conditions generally favorable to saprophytic forms.

In parasitism a marked variation in degree is to be

noted. Some species, like leprosy, grow with only the greatest difficulty outside of their proper host. Such as this may be said to be an *obligatory parasite*. Then again, other forms like the colon or feces bacillus are normally saprophytes, but under certain conditions, assume a semi-parasitic mode of life, becoming therefore, *facultative parasites*, i. e., they possess at times the faculty of developing under parasitic conditions.

23. Fermentation. Most of the saprophytic bacteria are concerned in the breaking down of organic matter, and consequently, they are often associated with the many different phases that this process assumes as seen in putrefaction, fermentation, decomposition or decay. These changes are of a complex character and vary much from one instance to another.

The changes embraced under the special term, fermentation, are distinguished by such a prominent characteristic that they may well be considered separately. In fermentation, complex substances are transformed by regular steps into simpler compounds.

In this class are to be included a large number of fermentative changes that occur in milk, such as the production of lactic acid in the souring of milk, the formation of butyric acid, and others that will be mentioned later. In fermentation, there are often large quantities of the fermentable substances changed, and, while a certain amount of the energy released in the breaking down of this material may be utilized by the bacterial cells, yet the disruptive changes set in motion by the living germs are out of all proportion to the results that are seen in the end.

Fermentation, although one of the best known processes that occurs in nature is even yet a partial mystery. Our knowledge of the changes that fermentable solutions

undergo during this process has been vastly augmented during the latter half of this century, but even yet, the problem has not been completely solved. The results of fermentation have been made use of from time immemorial, but no adequate conception of the changes involved was ever recognized until this century. The earlier theories of this action were mainly chemical, and it was not until the important studies of Pasteur were made that the relation of these changes to the action of living organisms was fully proven. He showed that fermentation was closely connected with the growth and multiplication of minute forms of organic life, and that the process was usually inaugurated by vital forces rather than by purely chemical activity.

Previous to his work the action of rennet upon milk had been known, and the difference between this fermentation and other types had been recognized by Schroeder and Dusch. They showed that the rennet ferment was unaffected by alcohol and other chemicals, while other fermentations due to organisms were checked in these fluids. Pasteur emphasized the distinction between these two sets of fermentative action and soon was able to classify these changes into two distinct types.

1. *Organized or living ferments*—those in which the fermentative change takes place as a result of the activity of a living organism.

2. *Unorganized or chemical ferments*—those in which the change is caused by a chemical substance, devoid of vitality, that is itself unchanged in the fermenting process. These unorganized, non-vital ferments are known as *enzymes*. Among the better known of these are rennet, that has the power of coagulating milk; diastase, the enzyme that converts starch into sugar; pepsin and trypsin, the digestive ferments of the animal body. In

their relation toward external influences such as heat, etc., many of these are affected in a manner similar to the vital ferments.

Among the organized class of ferments are those that directly affect the character of sugar-containing fluids, such as the yeasts and most of the bacteria. A great many of these organized ferments accomplish their effect indirectly through the agency of the enzymes that they themselves excrete. For instance, many of the abnormal fermentations in milk are caused by bacteria that are able to form various enzymes.

CHAPTER III.

METHODS OF STUDYING BACTERIA.

24. Necessity of bacterial masses for study. Bacteria are so infinitesimally small that it is impossible to study individual germs separately without the aid of first-class microscopes. For this reason, but little advance was made in the knowledge of these lower forms of plant life, until the introduction of culture methods, whereby a single organism could be cultivated and the progeny of this cell increased to such an extent in a short course of time, that they would be visible to the unaided eye.

25. Culture methods. The system of cultivating bacteria, known as the pure culture method, is based upon the supposition that the food medium in which the organism is grown is first freed completely from all pre-existing forms of life, or in other words, is perfectly sterile. The pure culture processes of the bacteriologist may be said to be in a sense, refined methods of seeding, such as the agriculturist employs. Just as the seed grain will in due season bring forth a harvest after its kind, so any kind of bacteria planted in a favorable food medium will produce a crop of its own. If the farmer's seed is foul, it shows in this crop, and the same is true with bacterial farming.

Bacteria, however, are so universally distributed that it becomes an impossibility to grow any special kind, unless the soil is first freed from all existing forms of germ life. To accomplish this, it is necessary to subject the nutrient medium used for a culture to some method of sterilization, such as by heat or filtration, whereby all

forms of organic life are thoroughly eliminated. Germ-free culture material is kept in sterilized glass tubes and flasks, and is protected from outside infection by plugs of sterile cotton. Material thus prepared, if protected from evaporation, will keep indefinitely, as the cotton acts as an effectual filter against the passage of any particles of matter.

26. Culture media. For culture media, many different substances are employed. In fact, bacteria will grow on almost any organic substance whether it is solid or fluid, provided the essential conditions of growth are furnished. The food substances that are used for culture purposes are divided into two classes; solids and liquids.

Solid media may be either permanently solid like potatoes or they may retain their solid properties only at certain temperatures like gelatin or agar. These last are of utmost importance in bacteriological research, for their use, which was introduced by Koch, permits the separation of the different forms that may happen to be in any mixture. Gelatin is used advantageously because the majority of bacteria present wider differences in their appearance upon this medium than upon any other. It remains solid at ordinary temperatures, becoming liquid in the neighborhood of 70° F. Agar, a gelatinous product derived from a Japanese sea-weed, has a much higher melting point, and can be successfully used, especially with those organisms whose optimum growth point is above the melting point of gelatin.

Besides these solid media, different liquid substances are extensively used, such as beef broth, milk, and infusions of various vegetable and animal tissues. Skim-milk is of especial value in studying the milk bacteria and may be used in its natural condition, or a few drops of litmus solution may be added in order to detect any change in its chemical reaction due to the bacteria.

27. Methods of isolation. Suppose for instance one wishes to isolate the different varieties of bacteria found in milk. The method of procedure is as follows: Sterile gelatin in glass tubes is melted and cooled down so as to be barely warm. To this gelatin which is germ-free a drop of milk is added. The gelatin is then gently shaken so as to thoroughly distribute the milk particles, and poured out into a sterile flat glass dish and quickly cov-

FIG. 2. A gelatin plate culture showing appearance of different organisms from a sample of milk. Each mass represents a bacterial growth (colony) derived from a single cell. Different forms react differently toward the gelatin, some liquefying the same, others growing in a restricted mass. *a*, represents a colony of the ordinary bread mold; *b*, a liquefying organism; *c*, and *d*, solid forms.

ered. This is allowed to stand on a cool surface until the gelatin hardens. After the culture plate has been left for twenty-four to thirty-six hours at the proper temperature, tiny spots will begin to appear on the surface, or in the depth of the culture medium. These

patches are called *colonies* and are composed of infinite numbers of individual germs, the result of the continued growth of the single organism that was in the drop of milk which was firmly held in place when the gelatin solidified. The number of these colonies represent in general the number of germs that were present in the milk drop. If the plate is not too thickly sown with these germs, the colonies will continue to grow and increase in

FIG. 3. Profile view of gelatin plate culture. Shaded part represents the gelatin medium in the covered glass dish; on the surface, different bacteria are developing; *b*, is a liquefying form that dissolves the gelatin while *c* and *d* grow on surface only and do not render gelatin soluble.

size, and as they do, minute differences will begin to appear. These differences may be in the color, the contour and the texture of the colony, or the manner in which it acts toward gelatin. (See fig. 3.) In order to make sure that the seeding is not too copious so as to interfere with continued study, an *attenuation* is usually made. This consists in taking a drop of the infected gelatin in the first tube, and transferring it by means of a sterile needle into another tube of sterile media. Usually this operation is repeated again so that these culture plates are made with different amounts of seed with the expectation that in at least one plate the seeding will not be so thick as to prevent further study.

To further study the peculiarities of different germs, the separate colonies are transferred to other sterile tubes

of culture material and thus *pure cultures* of the various germs are secured. These cultures then serve as a basis for continued study and must be planted and grown upon all the different kinds of media that are obtainable. In this way, the slight variations in the growth of different

FIG. 4. Pure cultures of different kinds of bacteria in gelatin tubes. *a*, growth slight in this medium; *b*, growth copious at and near surface. Fine parallel filaments growing out into medium liquefying at surface; *c*, a rapid liquefying form; *d*, a gas-producing form that grows equally well in lower part of tube as at surface (facultative anaerobe); *e*, an obligate anaerobe, that develops only in absence of air.

forms are detected and the peculiar characteristics are determined, so that the student is able to recognize this form when he meets it again.

These culture methods are of essential importance in bacteriology, as it is the only way in which it is possible to secure a quantity of germs of the same kind.

28. Use of the microscope in bacterial investigation. The microscope is in constant demand throughout all the different stages of the isolating process in order to verify the purity of the cultures. For this purpose, it is essential that the instrument used shall be one of strong magnifying powers (600–800 diameters) combined with sharp definition, so that these tiny organisms shall stand out clear and distinct.

The microscopical examination of any germ is quite as essential as the culture characteristics; in fact, the two must always go hand in hand. This examination reveals not only the form and size of the individual germ, but the manner in which they are united with each other, and any peculiarities of movement that they may possess.

In carrying out the microscopical part of the work, not only is the organism examined in a living condition, but stained preparations are made by using solutions of anilin dyes as staining agents. These are of great service in bringing out almost imperceptible differences. The art of staining has been carried to the highest degree of perfection in bacteriology, especially in the detection of germs that are found in diseased tissues in the animal or human body.

In studying the peculiarities of any special organism, not only is it necessary that these cultural and microscopical characters should be closely observed, but special experiments must be carried out along different lines, in order to determine any special properties that the germ may possess. Thus, the ability of any form to act as a fermentative organism can be tested by fermentation experiments; the property of causing disease, studied by

the inoculation of pure cultures into animals. A great many different methods have been devised for the purpose of studying special characteristics of different bacteria, but a full description of these would necessarily be so lengthy that in a work of this character they must be omitted.¹

¹ The following general works contain more or less complete descriptions of the various processes employed in studying bacteria:

Sternberg, *Manual of Bacteriology*, 1898; Fränkel, *Bacteriology*, 1891; Woodhead, *Bacteria and their products*, 1893; Abbott, *Principles of Bacteriology*, 1896; Pearmain and Moor, *Applied Bacteriology*, 1897; McFarland, *Text-Book upon Pathogenic Bacteria*, 1898.

PART II.

BACTERIA IN RELATION TO MILK.

CHAPTER IV.

CONTAMINATION OF MILK.

29. Milk as a food for bacteria. The fact that milk so readily undergoes decomposition changes shows that it is well suited for the nourishment of bacterial life. Its high content in organic matter, and the dilution of the same in a watery medium makes it an excellent food for germ as well as mammalian life. Its different constituents, however, possess different nutritive values. Of most importance are the nitrogen-containing compounds.

The albumen which is in solution is readily available. Casein can not be appropriated on account of its insoluble nature, unless it is first rendered soluble, a process which occurs with those bacteria that secrete enzymes that act on proteids.

Of the constituents of the milk that belong to the carbon-containing compounds, only one can be utilized by bacteria. The fat possesses but little food value for these organisms, because it cannot be decomposed by them. The milk sugar, however, is an admirable food for many species, more especially those that are known as the lactic acid producing or natural milk bacteria.

The bacterial cell contains so little mineral matter that the requirements of the cell for its growth are very limited, yet the mineral elements of the milk are needed for the growth of any protoplasm, and are used by the bacteria in the formation of new cell matter.

30. Milk, germ-free in udder. Under ordinary conditions, when examined in the proper manner, milk always reveals bacterial life. This germ content, however, is due to infection from without, for in the udder of a healthy animal, as secreted, the milk like the other secretions and tissues of the body is normally sterile.

31. Contamination of milk. In withdrawing the milk from the udder, it invariably comes in contact with germ life. The same is true after it is milked. From the time of milking, until it is consumed in one form or another, it is continually subject to contamination from exterior sources. In the main, germ life gains access while the milk is on the farm, but even in the factory

FIG. 5.—Microscopic appearance of milk showing relative size of fat globules and bacteria. Group of bacteria on left are lactic acid organisms.

the opportunities for infection are present in greater or lesser degree. Those forms that gain an early entrance generally predominate in the milk, as their early introduction enables them to develop for a longer period of time.

A. INFECTION OF MILK ON THE FARM.

32. Sources of contamination. The bacterial life that finds its way into the milk while it is yet on the farm may be traced to several sources, which may be grouped under the following heads: Unclean dairy utensils, fore milk, coat of animal, and general atmospheric surroundings. The relative importance of these various factors fluctuates in each individual instance.

33. Dairy utensils. Of first importance, are the vessels that are used during milking, and also all storage cans and other dairy utensils that come in contact with the milk after it is drawn. By unclean utensils, actually *visible* dirt need not always be considered, although its presence in cracks and joints of pails and cans is often evident. Unless cleansed with especial care, these places are apt to be filled with foul and decomposing material that suffice to abundantly seed the milk. Soxhlet¹ found that the addition of 0.1 per cent. of sour milk to fresh milk decreased the keeping quality of the latter from 15-30 per cent.; the addition of 1.5 per cent. diminished it 80 per cent. Where cans are not well cleaned the above amount could easily be added to the milk from the material that adhered to the walls of the can.

Through negligence, vessels are often used that are either unfit or are in an improper condition for handling milk. A rusty milk-can often spoils more milk than sufficient to purchase a new vessel. Wooden pails are no longer to be tolerated in a well-regulated dairy. Where possible, vessels should be made of pressed tin. If joints are necessary, they should be well flushed with solder so that they may be easily and thoroughly cleaned. In much of the cheap tinware that is now to be found on

¹Soxhlet, Ber. d Wanderversammlung bayer. Landwirthe, Oct., 1894.

the market, there is altogether too thin a veneer of solder to effectually cover the joints.

34. Use of milk-cans for transporting factory by-products. The general custom of using the milk-cans to carry back to the farm the factory by-products (skim-milk or whey) has much in it that is to be deprecated. These by-products are generally rich in bacterial life,

FIG. 6. The wrong and the right kind of a milk-pail. A, the ordinary type of pail showing sharp angle between sides and bottom; B, the same properly flushed with solder so as to facilitate thorough cleaning. The lower figure represents a joint as ordinarily made in tinware. The depression *a* affords a place of refuge for bacteria from which they are not readily dislodged. This open joint should be filled completely with solder.

more especially where the closest scrutiny is not given to the daily cleaning of the vats and tanks. Too frequently the cans are not cleaned immediately upon arrival at the farm, so that the conditions are favorable for rapid fermentation. This danger can be entirely obviated by a thorough cleansing process, but the patron who is shiftless in regard to his cans will generally be lax in his treatment of these vessels, and thus the opportunity for infection is present.

Many of the taints that bother factories are directly traceable to such a cause. A few dirty patrons will thus seriously infect the whole supply.

The responsibility of this defect should, however, not be laid entirely upon the shoulders of the producer. The factory operator should see that the refuse material does not accumulate in the waste vats from day to day and be transformed into a putrid mass. A dirty whey tank is not an especially good object lesson to the patron to keep his cans clean.

It is possible that abnormal fermentations may thus be disseminated from one farm to another in this way.

Suppose there appears in the dairy of A an infectious milk trouble, such as bitter milk. This milk is taken to the factory and passes unnoticed into the general milk-supply. The skim-milk from the separator is of course infected with the germ, and if conditions favor its growth, the whole lot soon becomes tainted. If this waste product is returned to the different patrons in the same cans that are used for the fresh milk, the probabilities are strongly in favor of some of the cans being contaminated and thus infecting the milk-supply of other patrons. If the organism is endowed with spores so that it can withstand unfavorable treatment, this disease may spread from patron to patron simply through the infection of the vessels that are used for the transportation of the by-products.

It would be possible to obviate any trouble arising from this source if a separate receptacle was used for this purpose, but the objection is frequently urged that this is impractical, yet many of the more progressive factories are following this practice with excellent results.

35. Effect of steaming milk-pails. Even where utensils are in good condition and well cleaned, the germ content of the milk may be reduced, and therefore, the keeping quality enhanced by a brief steaming of the receiving cans. For this experiment¹ two cans were taken,

¹ Russell, 11th Rept. Wis. Agr'l. Expt. Stat., p. 152, 1894.

one of which had been cleaned in the ordinary way, while the other was sterilized by steaming. Before milking, the udder of animal was thoroughly cleaned, and special precautions taken to avoid raising of dust; the fore milk was also rejected. Milk was then drawn directly into these two cans and bacterial determinations made:

Number of bacteria per cc. in steamed pail, 165.

Number of bacteria per cc. in ordinary pail, 4265.

Time before milk in steamed pail soured, $28\frac{1}{2}$ hours.

Time before milk in ordinary pail soured, 23 hours.

If an imperfectly cleaned pail had been used for this purpose, the difference in souring would have undoubtedly been more apparent.

To illustrate the varying germ content of cans cleaned in different ways, Harrison¹ rinsed out cans with 100 cc. of sterile water, and then determined the germ content of the same. The following data represents the number of bacteria per cc. in the rinsing water from cans improperly cleaned (series A), from cans washed in tepid water and then scalded — the usual factory method — (series B) and from cans washed in tepid water and steamed for five minutes (series C.).

Effect of steaming on germ content of cans.

Series A (Dirty cans) ..	238,525,	342,875,	215,400,	618,200,	806,320,
	510,270,	230,100,	610,510,	418,810,	317,250.
Series B (Ordinary cleaning method) ...	89,320,	84,750,	26,800,	24,000,	38,400,
	76,800,	15,200,	13,080,	44,160,	93,400.
Series C (Approved method)	1,170,	1,792,	890,	355,	416.

36. Cleaning dairy utensils. Milk vessels should never be allowed to become dry when dirty, for dried particles of milk residue are extremely difficult to remove.

¹ Harrison, 22nd Rept. Ont. Agr'l. Coll., p. 113. 1896.

In cleaning dairy utensils they should first be rinsed in lukewarm instead of hot water, so as to remove organic matter without coagulating the milk. Then wash thoroughly in hot water, using soap or weak alkali. A borax solution is sometimes recommended for cleaning bottles. Strong alkalies should not be used. After washing, rinse thoroughly in clean hot water; then invert over a steam jet for a few minutes. A momentary application of hot or even boiling water is insufficient to destroy germ life that lurks in joints of vessels. Live steam is especially efficient as a germ-destroying agent. If steamed, the cans will dry more quickly.

It is not often that steam is available on the farm, but under such conditions, it is possible to acquire practically the same results by using boiling water, although the length of exposure must be increased.

Not only should the greatest care be paid to the condition of the cans and milk-pails, but all dippers, strainers, and other utensils that come in contact with the milk, must be kept thoroughly clean. Cloth strainers, unless attended to, are objectionable, for the fine mesh of the cloth retains so much moisture that they become a veritable hot-bed of bacterial life, unless they are daily boiled or steamed.

FIG. 7. Section of udder showing relation of milk-secreting tissue to milk duct (after Thanhofer). *a*, exterior opening of milk duct; *b*, milk cistern; *c*, milk sinus; *d*, glandular secreting tissue; *e*, sphincter muscle of teat.

37. Influence of first or fore milk. A leading factor in the contamination of milk, the importance of which is rarely recognized, comes from the bacteria that gain access to the milk, by mixing the first or fore milk with

the remainder of the milk. Even when the milking is most thoroughly done, there remains in the milk ducts of the udder, a few drops of milk that afford sufficient nutriment for the development of any germs that may gain access through the opening of the teat. According to Gernhardt, it is possible that they may penetrate the udder as far as the milk cisterns or gland tissue itself, but the evidence on this point is not decisive. The relatively high temperature of the teat facilitates a rapid growth.

Under these conditions, a small number of organisms are able to increase in such numbers that the first few spurts of milk contain many more than those which are subsequently drawn. The following data by Harrison¹ strikingly illustrates this point:

Number of bacteria per cc. in milk.

Fore milk	26,070,	25,630,	38,420,	18,110,	54,800,	32,700,
	43,520,	27,830,	18,500,	29,400,	45,630,	48,700.
Milk after removal						
of fore milk	1,246,	1,150,	1,430,	3,420,	1,560,	890,
	2,575,	4,820,	3,270,	1,285,	1,350.	

If the fore milk is received in a separate vessel and kept protected from the air, it will generally be noted that it sours more rapidly than the remainder of the milk.

As a rule the number of different species found in the fore milk is usually small, not more than one or two forms being present at any time. As to the character of these forms data is conflicting. Harrison² reports finding peptonizing bacteria in the same, and Marshall³ states that organisms are found that resist pasteurizing,

¹ Harrison, 22d Rept. Ont. Agr'l. Coll., p. 108, 1896.

² l. c., p. 108.

³ Marshall, Mich. Expt. Stat., Bull. 147, p. 42.

a characteristic not usually associated with the lactic acid class.

Bolley¹ in thirty experiments found twelve out of sixteen species to belong to the lactic acid class. In no case were gas-generating species present. This fact is important in the selection of a milk from a single animal for the cultivation of a starter.

This condition represents the milk of perfectly healthy animals; where the udder is diseased, as in the case of infectious garget or inflammation, bacteria may be present in much larger numbers and affect the milk seriously for food purposes.

Not all species seem to be able to maintain themselves in the udder even if they should be introduced. Dinwiddie² injected into the milk cistern a lactic acid producing facultative anaerobe that grew luxuriantly at 99° F. An examination of the milk several times afterwards failed to show the presence of this organism in any case, although other species were isolated. .

38. Dirt from animal. By many it is believed that much of the germ life that gets into the milk comes from the food of the animal. For this reason fermenting food-stuffs of any kind are regarded as unfit for use. While material of this class is not a suitable nutrient for any animal, the danger to the milk is not due to the bacteria ingested with the food, but to a large extent to those that adhere to the animal's coat, and subsequently fall into the milk. Of course the udder may be infected if the animal is diseased as in inflammation (mammitis),³

¹ Assoc. Ag. Coll. and Expt. Stat., 1895, also Cent. f. Bakt. II Abt. 1: 795, 1895.

Second article, Bull. No. 21, N. D. Expt. Station.

² Dinwiddie, Ark Expt. Stat., Bull. 45, p. 57.

³ Guillebeau, Landw. Jahr. d. Schweiz, 1892, p. 27.

or tuberculosis, in which case bacteria find their way directly into the milk. Marshall¹ succeeded in isolating direct from the udder, a pure culture of Nocard's streptococcus, the cause of infectious udder inflammation.

The hairy coat of the animal offers exceptional facilities for the harboring of dust and dirt. Cows wading in stagnant pools cover the udder with slime and dirt that is readily dislodged when dried. The hairy coat is, therefore, extremely rich in the various forms of bacterial life that are derived from the particles of excreta that stick to the flanks and under parts of the animal. Where peat is used for bedding, favorable reports are made as to its value.

The amount of actual impurities that are to be found in milk, even after it is strained, will surprise the casual observer, although it should be noted according to Backhaus,² that about one-half of fresh manure dissolves in milk and thus does not appear as sediment. From a large number of determinations of the solid impurities found in the market milk of different European cities, Renk³ deduces the following rule: If a sample of milk shows any evidence of impurity settling on a transparent bottom within two hours, it is to be regarded as containing too much solid impurities. These solid particles, composed largely of manure and dirt, are always teeming with bacteria, especially with putrefactive and decomposition organisms. It has recently been estimated that the city of Berlin consumes daily 300 pounds of dirt and filth in its milk-supply.

Not only is the number of bacteria thus introduced into the milk very considerable, but the character of the same

¹ Marshall, Mich. Expt. Stat., Bull. 146, p. 6.

² Backhaus, Milch Ztg., 26: 357, 1897.

³ Renk, Cent. f. Bakt., 10: 193.

is a question of even greater importance. Those species that are derived primarily from manure particles are, as a rule, the peptonizing or digestive species that cause a decomposition of the casein, and should, therefore, be avoided if possible.

Improper stable conditions greatly favor the amount of filth that may adhere to the animal. The more highly nitrogeaneous feeding that is practiced at present produces a softer manure and one in which putrefactive bacteria are much more likely to be abundant.

Wüthrich and Freudenreich¹ have studied the influence of feeding on the bacterial content of manure, and they find a markedly higher content in manure where animals are given dry feed than where kept on grass. The character of the manure, however, is different, it being much more liquid with moist than dry feed, and therefore, they believe more likely to find its way into the milk. They found as many as 375,000,000 bacteria per gram in fresh manure, the majority of which consisted of *B. coli communis*, the hay bacillus, and other species able to peptonize the casein.

39. Influence of the milker. The condition of the milker is by no means an unimportant factor. If he performs the milking in the dust-laden garments that he has worn all day, he himself is covered with particles that are readily dislodged when he comes in contact with the cow.

Particular attention should be paid to the hands of the milker. The habit of moistening the hands with a few drops of milk just before milking is to be deprecated from every standpoint, but especially so, when considered from our present point of view. After having

¹ Cent. f. Bakt., II Abt., 1: 873, 1895.

washed his hands in clean water, a pinch of vaseline on his hands will enable him to obtain a firmer grasp, and at the same time, any scales or dirt rubbed from the teat would be held by the vaseline. Its healing effect on chapped or sore teats would also be helpful. Freudenreich¹ reports some experiments in which the germ content of milk was reduced from several thousand to 200 where the hands were well rubbed with vaseline before milking. Where the best of conditions are carried out it is worth while to have the milker clothed in a suit kept for this purpose, especially the upper portion of the body. An outer garment could easily be slipped over the regular working clothes. This garment should be laundered at frequent intervals.

40. Exclusion of dirt. A large amount of filth and dirt can be prevented from falling into the milk. Carding the udder and flanks to remove the loose hairs will remove a considerable source of dirt. So long, however, as the coat of the animal is dry, dust particles with their adherent bacteria are readily dislodged. If the coat of the animal is moist, this deposition can be almost effectually prevented. The surface should, however, not be dripping wet. The objection has been urged by some that washing the udder starts the milk secretion, and unless the animal is milked at once, the yield of milk is diminished thereby. Eckles² has recently reported a number of experiments from which he concludes that when the animal is accustomed to the treatment, no noticeable effect is produced either in amount of milk or butter fat.

In order to show the effect of dirt and dust, the experiment described below teaches a valuable lesson. A cow

¹Freudenreich, *Die Bakteriologie*, p. 30.

²Eckles, *Hoard's Dairyman*, Aug. 8, 1898.

that had been pastured in a meadow was partially milked out of doors. During the operation a covered glass dish containing sterile gelatin was exposed for sixty seconds underneath the belly of the cow in close proximity to the milk pail. The udder, flank, and legs of the cow were then thoroughly cleaned with water, and all of the precautions referred to before were carried out, and the milking then resumed. A second plate was then exposed in the same place for an equal length of time; a control also being made at the same time at a distance of ten feet from the animal and six feet from the ground to ascertain the germ contents of the surrounding air.

From this experiment the following instructive data were gathered. Where the animal was milked without any special precautions being taken, there were 3,250 bacterial germs *per minute* deposited on an area equal to the exposed top of a ten-inch milk pail. Where the cow received the precautionary treatment as suggested above, there were only 115 germs per minute deposited on the same area. In the plate that was exposed to the surrounding air at some distance from the cow, there were sixty-five bacteria. This indicates that a large number of organisms from the dry coat of the animal can be kept out of milk if such simple precautions as these are instituted.

Another method of exclusion is to use a milk pail having a partially closed top that will prevent the introduction of a large part of the dirt.

Still another method that has much to recommend it, is passing the milk through a hand separator immediately. This not only removes nearly all of the suspended particles of foreign matter, such as dirt and filth of various kinds,¹ but it eliminates a large part of the bacteria

¹ Backhaus (Milch Ztg., 26: 358, 1897) found that 95.6 per cent. of impurities were removed by centrifugal separation.

with the same, if it is done immediately after the milk is drawn. The only objection to this process is that the cream does not rise so thoroughly as where it is not centrifuged, but the other advantages where an especially clean milk is wanted more than compensate for this defect.

41. Influence of air. It is impossible to separate the influence of the air entirely from that of the animal, as the dust particles from the coat of the animal must of necessity pass through the air.

FIG. 8.—Effect of contaminated air. Each spot on this surface represents a developing colony that has grown from a germ that was deposited on surface of a sterile gelatin plate (3 inches in diam.) in 30 seconds. This exposure was made at time the cows were fed.

Germ life cannot develop in the air, but in a dried condition, organisms retain their vitality for long periods of time. The use of dry fodder, the bedding of animals with straw adds greatly to the amount of dust particles, and consequently the germ life floating in the air, as seen in fig. 8. Taints in milk have frequently been traced to infection arising from this source.

While the stable air cannot be freed entirely from dust, much can be done with a little forethought. Feeding before milking adds materially to the germ content, especially if feed is of dry character. If moistened feed is given just before or during milking, the same objection does not inure. Harrison¹ gives some striking results on this point. The numbers cited indicate the number of bacteria that were deposited per minute on a surface equal to that of a twelve-inch milk pail. In Series A, the exposure was made during bedding; in B, this operation was performed an hour before.

Influence of dusty air on germ life.

Series A.....	16,000,	13,536,	12,216,	12,890,	15,340,
	19,200,	23,400,	27,342,	42,750,	18,730.
Series B.....	483,	610,	820,	715,	1,880,
	2,112,	1,650,	990,	1,342,	2,370.

These results indicate that the bacteria are for the most part attached to particles of considerable weight as they settle readily to the floor.

42. The relative importance of foregoing factors.

The relative importance of these various factors differs so much in different cases that no uniform rule can be given as to the effect of their presence in different cases. In those dairies where no especial care is exercised over the methods of handling the milk, the factor of unclean utensils and filth from the animal are usually the greatest. Where a careful supervision is given to these details, the influence of the fore milk is of first importance. The effect of these various contaminating factors can be largely minimized, and in some instances eliminated without great difficulty.

A goodly number of the bacteria present in the fore milk can be prevented from gaining access to the milk by rejecting the first few streams that are milked from each

¹ Harrison, 22d Rept. Ont. Ag. Coll., 1896, p. 111.

teat, although this factor can be controlled to a less degree than any of the others. The actual loss occasioned in doing this is very slight, as the first part of the milking is very poor in butter fat.

Dust in the air of the barn is a minor factor, yet this can be materially reduced by exercising care relative to feeding dry fodder, or bedding the animals just previous to, or during the milking.

The number of germs derived from the animal itself and the milker can be largely decreased by keeping the animal thoroughly clean, and having the milker milk with clean hands.

The effect of contamination arising from imperfectly cleaned milk vessels can be practically excluded by sterilizing all such utensils in steam, or even in scalding water for a short time.

To determine what reduction could be accomplished by drawing and handling the milk under as clean conditions as possible, the following experiment was carried out: The udder was thoroughly carded, and then moistened; the milk received in steamed pails, the fore milk being rejected.

The milk from a cow treated in this way contained 330 bacteria per cc., while that of a mixed herd taken under usual conditions was 15,500 bacteria for same volume. This carefully handled milk kept over twenty hours longer at room temperatures than the ordinary product. Backhaus estimates that the germ life in milk can be easily reduced to $\frac{1}{2000}$ of its original number by using care in milking. Methods of this sort must be instituted in those dairies that are furnishing the highest grade of product. Where such methods are in vogue, bacteria are excluded for the most part and pasteurization becomes unnecessary. Such milks are frequently known as

“sanitary” or “certified” as the method of handling the herd is often under the control of outside parties (physicians or health board) who certify to the regulations that are followed.

FIG. 9. Bacterial content of milk handled in ordinary way. Each spot represents a colony growing on gelatin plate. Compare with fig. 10, where same quantity of milk is used in making culture. Over 15,000 bacteria per cc. in this milk.

The question may arise as to the necessity of these precautionary measures if the milk is to be used for ordinary factory purposes. While certain bacteria are essential in the butter and cheese industry to secure a normal and characteristic fermentation, yet it is better to have the germ life reduced to the lowest numbers than to have the raw milk infected through slovenly methods of handling. If this is done, the maker has the fermentation under his control, and by the addition of a starter which he can choose, he can vary the character of the product to suit the demands of the trade, which he cannot do, if the raw milk is brought to him in a dirty condition, and in an advanced stage of fermentation.

Dairymen have learned many of these lessons in the severe school of experience, but the reason for the same is so palpably plain in the light of bacteriological explanation, that further discussion would seem unnecessary. It

FIG. 10. Bacterial content of milk drawn with care. Diminished germ content is shown by smaller number of colonies (330 bacteria per cc.). Compare this culture with that shown in fig. 9.

remains to be seen whether the words of the eminent German authority, Prof. Fleischmann will much longer prove true, when he says that "all the results of scientific investigation which have found such great practical application in the treatment of disease, in disinfection and in the preservation of various products, are almost entirely ignored in milking."

43. Effect of temperature on bacterial growth. After milk is once seeded with bacterial life, no one factor exerts so potent an effect upon the rate of change that takes place as that of temperature, for the rapidity of bacterial growth in milk is determined mainly by this factor as is seen in fig. 11.

Although different species vary in their rate of development, yet moderately warm temperatures from 75° to 90° F., encourage rapid growth. Unless the milk is

FIG. 11—Showing the effect of cooling milk on the growth of bacteria. The beneficial results of early chilling are readily apparent.

quickly deprived of its original heat, the rate of the fermentative changes will be much increased as is shown in following results obtained by Cnopf and Escherich:

Rate of growth of single germ

	2 hrs.	3 hrs.	4 hrs.	5 hrs.	6 hrs.
54° F.....	4	6	8	26	435
97° F.....	23	60	215	1830	3800

If a can of milk is allowed to cool naturally, it will take several hours before it reaches the temperature of the surrounding air. During this time, the organisms in the fore milk are continuing their rapid growth, while those forms that come from dust, and are presumably in a latent state on account of their condition, awake from their lethargy under the influence of these favorable surroundings. If bacteria once gain an entrance and begin to germinate, a considerably lower temperature is required to successfully check development than to hold

latent organisms, like spores, in a condition where germination will not occur.

To hasten this lowering of temperature, artificial cooling is a necessity. Coolers using cold running water or ice water are efficient in reducing the temperature to a point where development is much checked.

44. Tainted milks. Not all taints in milk can be traced to the development of bacterial causes. In many cases they are produced by the direct absorption of odors in a purely physical way or to some unusual condition of the system of the animal. In some cases with animals old in lactation, the milk becomes abnormal in that the cream does not rise readily. Such milks are known as "lazy" or "dead" milks. Then again, the dairyman may experience difficulty in churning, but more often these difficulties are attributable to the non-performance of important manufacturing details rather than to a perverted condition of the milk itself. Milk is very prone to absorb volatile odors, the fat especially having a great affinity for many of these substances.

45. Direct absorption from animal. Odors of this sort may be absorbed by the milk previous to or after the milking has occurred. The peculiar "cowy" or "animal odor" of fresh milk is an illustration of an inherent peculiarity of the milk. If certain strong flavored substances as onions, certain root crops, or other vegetables are consumed by the cow, the odor-yielding substances in the same may reappear in the milk, especially if the animal partakes of these a short time before milking. According to Kober and Busey, the milk of swill-fed cows has a peculiar taste, and is said to produce a highly acid urine and eczema. Brewers' grains and distillery slops when fed in large quantities frequently induce an abnormal chemical reaction of the milk. Taints

of this sort can easily be prevented by taking care that dairy stock is not fed such feed.

46. Straining milk in the barn. For convenience, milk is often strained in the barn, but the danger of tainting the same where this is done is so great that the custom is not to be recommended. In straining the milk so much surface is exposed to the air that considerable bacterial infection can occur, unless this is carried out in a room free from all dust particles. Where the process is done in the barn, the can with its strainer is often left uncovered. Under these conditions, a constant deposition of germ life is taking place, and with every pailful of milk strained, this is washed through the meshes of the strainer into the milk below.

47. Absorption of odors in fresh milk. Not only is straining in the barn to be deprecated from the above standpoint, but the possibility of direct physical absorption of existing taints in the stable should warn one against this custom. It is a commonly accepted idea that milk evolves odors and cannot absorb them so long as it is warmer than the surrounding air, but from experimental evidence,¹ the writer has definitely shown that the direct absorption of odors takes place much more rapidly when the milk is warm than when cold, although under either condition, it absorbs volatile substances with considerable avidity. In testing this, fresh milk was exposed to an atmosphere impregnated with odors of various essential oils and other peculiar aromatic substance, whose odors could be readily identified. Under these conditions the cooled milk was tainted very much less than the milk at body temperatures, even where the exposure was for a half hour.

¹ Russell, 15th Rept. Wis. Expt. Stat., p. 104, 1898.

48. Aeration. Practical experience has long demonstrated the advantage of aerating the milk as soon after milking as possible. This is accomplished in a variety of ways. In some cases, air is forced into the milk; in others, the milk is allowed to distribute itself in a thin sheet over a broad surface and fall some distance so that it is brought intimately in contact with the air. The benefit claimed for aeration is that foul odors and gases which may be present in the milk are thus allowed to escape by bringing the finely divided milk into contact with the air. As ordinarily practiced, aeration is usually combined with cooling, and it is noteworthy that the most effective aerators are those that cool simultaneously. Under these conditions, the keeping quality of the milk is increased, but where milk is simply aerated without cooling, no material benefit in keeping quality is observed. A satisfactory scientific explanation of the advantages of aeration has not yet been made. It is difficult to see how the process can have any effect on the bacterial life in the milk. Its influence, undoubtedly, is on the odors directly absorbed by the milk.

49. Distinction between bacterial and non-bacterial defects in milk. In fresh milk it is relatively easy to distinguish between taints caused by the operation of external biological forces and those due to direct absorption.

If the taint grows more pronounced as the age of the milk increases, it is probably due to the living organisms, as the taint-producing bacteria usually gain an entrance after the milk is drawn from the cow, and require a certain period of incubation before undesirable products are formed. Taints due to errors in feeding are more pronounced when the milk is first drawn.

The absorption of flavors by old milk is sometimes puzzling to distinguish from a bacterial trouble, but if the surroundings are closely noted, the cause can usually be traced to delayed absorption of bad odors from the room where milk is stored.

If the trouble is of bacterial origin, it can frequently be detected by transferring a small quantity of the suspected milk to a fresh lot, preferably that which has first been boiled. If due to a living organism, the abnormal fermentation will be propagated and so reproduce the difficulty in the infected milk.

B. INFECTION OF MILK IN THE FACTORIES.

50. Treatment of milk in factories. Where milk is taken to a creamery or cheese factory, the method of treatment often modifies the bacterial life of the milk to a marked degree.

Butter and cheese-making both require the presence of bacteria, but the germ life must be of the right sort to be beneficial in its action. For this reason the maker wishes the milk to be as free as possible from all contaminating influences. If the foregoing suggestions are intelligently followed, the raw material will be turned over to the factory in prime condition; but the rational treatment must be continued here if the best of products is to be expected. Numerous influences are at work in the factory that add their mite to the milk in different stages of its manufacture, and ultimately affect in a serious way the final product. Of course, very much depends upon the proper observance of physical conditions, but a brief outline of some of the possible dangers will also be pertinent.

The cardinal precept in the factory as well as on the farm should be cleanliness. Cleanliness here must not

be taken to mean a mere absence of dirt and filth, but all utensils that come in actual contact with the milk should be rendered as germ-free as possible.

From the time that the milk enters the weigh-can until the butter is in the tub, and the cheese on the curing-shelf, it should be remembered that many opportunities for infection are always present.

51. Factory utensils. In the factory, part of the same set of factors of infection are at work as are found in the farm dairy. The condition of factory utensils is always a point of prime importance. Where steam is accessible, as it is in the majority of cases, there is no excuse for uncleanness of any sort, as most pieces can be steamed directly. Open vats can be thoroughly scalded if covered with a heavy canvas cloth. Separator bowls, churns, cans, and dippers should always receive a daily treatment. The rational nature of these methods is to be seen in those cases where the same utensil, say a dipper, is employed indiscriminately in handling all kinds of dairy products. All cans with rusty seams should be discarded. Permit no vat to be repaired by putting in a false covering over the old one. If a minute leak is established, such places become a harbor of refuge for all kinds of putrefactive organisms. In a number of cases ill-smelling factory odors have been traced to such a cause.

52. Infection from air. The influence of the air on the germ content of the milk is, as a rule, overestimated. If the air is quiet, and free from dust, the amount of germ life in the same is not relatively large. In a creamery or factory, infection from this source ought to be much reduced, for the reason that the floors and walls are, as a rule, quite damp, and hence germ life cannot easily be dislodged. The majority of organisms

found under such conditions comes from the person of the operators and attendants. Any infection can easily be prevented by having the ripening cream-vats covered with a canvas cloth. The clothing of the operator should be different from the ordinary wearing-apparel. If made of white duck, the presence of dirt is more quickly recognized, and greater care will therefore be taken than if ordinary clothes are worn.

The surroundings of the factory have much to do with the danger of germ infection. Many factories are poorly constructed and the drainage is poor so that filth and slime collect about and especially under the factory. The emanations from these give the peculiar "factory odor" that indicates fermenting matter. Not only are these odors absorbed directly, but germ life from the same finds its way into the milk, contaminating the same. Connell¹ has recently reported a serious defect in cheese that was traced to germ infection from defective factory drains. According to Robertson, sometimes it becomes necessary to remove cheese factories to new locations before the bad conditions can be controlled.

53. Water and ice-supply of factories. These supplies should be carefully controlled. Water, and to a less extent ice, always contain bacteria in varying numbers, so that it is possible that undesirable organisms may be introduced from this source. Most creameries derive their water-supply from private wells, and in using these, care should be taken that they are arranged so as not to receive any surface drainage. A deep well from which the water is used in large quantities, if properly arranged, will contain the minimum number of germs as the ground water is practically free from them.

¹Connell, Rept. of Com. of Agric., part xvi, p. 15, 1897.

Harrison¹ has recently traced an off-flavor in cheese in a Canadian factory to an infection arising from the water-supply. He found the same germ in both water and cheese and by inoculating a culture into pasteurized milk succeeded in producing the undesirable flavor. Milk can easily be infected through using such water for washing utensils. Some well waters containing iron in solution are a source of trouble in factories on account of the development of iron bacteria that cause the soluble iron to be precipitated in the form of rusty flakes of ferric oxid.

In ice the majority of organisms (60-90%) are destroyed, but enough remain, so that if ice is secured from a highly polluted source, the living germ life in it may be an element of danger. It is not considered advisable to add ice directly to milk or cream, but as storage vessels are indiscriminately used to contain either ice or milk, the possibility of infection should be kept in mind.

54. Numbers of bacteria in milk. The germ content of milk varies so greatly that unless the conditions are all known, it is impossible to foretell what may be found therein. An examination of milk will often reveal a difference in numbers, ranging from a few score of germs to hundreds of millions per cc.. The presence of such a varying number is dependent upon certain factors, as the age of the milk, the care taken during the milking, and also the way in which it has been handled since that time. Disregarding milk of different ages, the number of germs present in any sample bears a general relation to the amount of dirt and filth with which it has come in contact since it was drawn from the cow. Bacteria and

¹Harrison, Hoard's Dairyman, Mch. 4, 1898.

filth of all kinds are so intimately associated with each other that the presence of one rightly presupposes that of the other.

As to the numerical bacterial content of any milk, there is such a wide variation under different conditions that figures are of little worth. No exact relation can be maintained between number of bacteria in milk and the development of fermentative products.

The studies by different observers have been carried on under such diverse conditions that no comparison of the results can well be made. Under American conditions but little work has been done in this direction, yet milk as it is sold here to the consumer usually contains less bacteria than that retailed in European cities, although as Conn has pointed out, it is materially older. As he intimates this fact is explained by the relatively free use of ice in this country. A few determinations of the bacterial contents of European milks that have been analysed biologically will illustrate this point.

Renk¹ found in Halle milk-supply 6-30,000,000 germs per cc.; Cnopf² in Munich milk-supply 200,000-6,000,000 per cc.; Uhl³ in Giessen milk 83,000-170,000,000 per cc.; Clauss⁴ in Wurzburg 222,000-23,000,000 per cc.; Bujwid in Warsaw an average of 4,000,000 per cc. and Knochensteirn⁵ in Dorpat 25,000,000 per cc.

Sedgwick and Batchelder⁶ report fifty-seven samples of Boston milk as containing from 30,000-4,220,000 per cc. In the country, they found in the milk fresh from

¹ Renk, *Cent. f. Bakt.*, **10**: 193.

² Cnopf, *Ibid.* **6**: 553.

³ Uhl, *Zeit. f. Hyg.*, **12**: 475 (1892).

⁴ Clauss, *Diss. Wurzburg*, 1889.

⁵ Knochensteirn, *Chem. Cent.*, **11**: 62.

⁶ Sedgwick & Batchelder, *Boston Med. Surg. Journ.*, Jan. 14, 1892.

the cow 30,000 and in the milk as used on the table, about 70,000 organisms per cc. Loveland and Watson¹ found in the supply of Middletown, Conn., from 11,000 to 85,500,000 per cc. McClatchie² found in Los Angeles, Cal., supply, an average of 11,700 per cc. in twenty-six trials. In my experience, the mixed milk of a herd that is kept with any reasonable degree of cleanliness, if examined immediately after it is milked, usually will not contain more than 5-20,000 germs per cc. The number present in any milk is due to the influence of so many factors that it is practically impossible to establish any number as a *normal*, although Bitter³ sets 50,000 germs per cc. as a maximum limit for a milk intended for human food. The milk as delivered by the milkmen to their private customers in the city of Madison, Wis., ranges from 15,000-2,000,000 organisms per cc., varying mainly with the season of the year.

The presence of such large numbers in a food product need not necessarily occasion alarm from a hygienic standpoint, although it is quite certain that putrefactive forms may have an irritating effect upon a deranged digestive tract, and thus produce intestinal disturbances, especially with infants during the summer months.

If we compare the bacterial flora of milk with that of sewage, a fluid that is popularly and rightly supposed to be teeming with germ life, it will almost always be observed that milk when it is consumed, is richer in bacteria by far than the sewage of our large cities. Sedgwick⁴ found that the sewage of Lawrence, Mass., contained at

¹ Loveland and Watson, 7th Report, Storrs Sta. (Conn.), 1894, p. 72.

² McClatchie, Bull. 3, Agr. Expt. Stat. (So. Cal. Acad. Sc.), Aug. 1897.

³ Bitter, Zeit. f. Hyg., 8: 240.

⁴ Sedgwick, Rept. Mass. Bd. Health, 1890, p. 60.

the lowest, 100,000 germs, while the maximum number was less than 4,000,000 per cc. This range in numbers is much less than is usually found in the milk-supply of our large cities.

55. Kinds of bacteria in milk. The number of bacteria in milk is not of so much consequence as the kind present. While milk may contain forms that are injurious to man, still the great majority of them have no apparent effect on human health. In their effect on milk, the case is much different. Roughly, we may divide them into three classes, depending upon their action in milk.

1. Bacteria that exert no appreciable effect in milk.
2. Bacteria that are beneficial by reason of the products which they form.
3. Bacteria that are injurious on account of the effect which they produce in milk.

A suprisingly large number of bacteria that are found in milk belong to the first class. Undoubtedly they affect the chemical characteristics of the milk somewhat, but not to the extent that it becomes physically perceptible.

Those species that are concerned in the production of proper flavor and aroma in butter, and which are also concerned in the development of acid and possibly associated with formation of cheese flavor represent the second type. Many of these organisms are lactic acid producing, but in addition to these, some of the casein ferments are also associated with aroma production in butter.

The third class includes those species that are able to produce deleterious and undesirable flavors in milk and milk products. The majority of the abnormal fermentations of milk referred to in this chapter come under this

head. Most of these gain access to the milk through slovenly and careless methods of handling. Those species associated with animal excreta are particularly dangerous. The number of different kinds that have been found in milk is quite considerable, something over 200 species having been described more or less thoroughly. In all probability, however, many of these forms will be found to be identical when they are subjected to critical study.

CHAPTER V.

MILK FERMENTATIONS AND THEIR TREATMENT.

56. Classification of milk fermentations. If any sample of milk is allowed to stand at any ordinary temperature for several days, a profound physical and chemical change inevitably takes place. As a rule the milk will sour. In this process, certain acids are formed at the expense of the milk-sugar. If it is still allowed to stand for some time after it has become thoroughly sour, a series of subsequent changes usually occur. Often it will evolve foul-smelling gases and undergo putrefactive changes. Sometimes, however, other ferment organisms gain the ascendancy over the ordinary souring process, in which case the fermentation is said to be abnormal. These peculiar changes are the cause of various taints in milk. They are of great importance because a small quantity of milk tainted in this way is liable to infect a much larger quantity and spoil the same through the rapid development of the obnoxious organism. In some cases, the abnormal condition may not become marked until the milk is made up into some other product as butter or cheese. The difficulties that occur under these conditions will be discussed under their appropriate heads.

It is impossible in the present state of our knowledge to classify these fermentations in any other than a most provisional way. While our knowledge of some of them from a bacteriological standpoint is fairly complete, so many, as yet, have been studied only superficially that we are not in a position to classify them as we would other chemical changes.

The various fermentations may be grouped according to the substances in the milk upon which they chiefly act, such as those affecting milk-sugar, or casein. This arrangement is followed in this connection, but it should be borne in mind that many organisms act upon several of the milk constituents, and hence, it is difficult to correctly classify the same.

Another method of classification is based upon the products manufactured during the fermentation, but even here confusion is liable to enter to some extent, because in many cases there are several distinct substances formed.

Milk is such a complex substance that the changes produced by a single germ are often so numerous that the processes can not be separated in their reactions. It must be remembered then, in referring to the different types of fermentations, that perhaps a distinct by-product is being formed, but it is more than probable that there are a series of changes, in which the most marked decomposition process is alone taken into consideration. For example, there is a fermentation classed under the head of the butyric changes, a decomposition process in which butyric acid is the chief product formed, but this may be associated with an alkaline condition of the milk and the production of a bitter substance in the same. Thus, the subdivision followed here will of necessity be imperfect and occasional instances will be noted where some changes in milk might well be described under several heads.

Some of these fermentation changes occur so constantly that they are to be regarded as normal or natural in their character. Normal, however, only in the sense that the bacteria that cause them are so widely distributed that the change is inevitable and not that the milk itself would undergo any of these changes if it were not for the presence of the different germs that bring them

about. Of such a nature is the lactic acid fermentation, by far the most common change that occurs in milk.

57. Souring of milk. Milk naturally undergoes a change known as souring, if allowed to stand for several days at ordinary temperature. This is due to the formation of lactic acid, which is produced by the decomposition of the milk-sugar. While this change is wellnigh universal, it does not occur without a pre-existing cause, and that is the presence of certain living bacterial forms. These organisms develop in milk with great rapidity, and the decomposition changes that are noted in souring are due to the by-products of their development.

The milk-sugar undergoes fermentation, the chief product being lactic acid, although various other by-products as other organic acids (acetic, formic, and succinic), different alcohols, and gaseous products, as CO_2 , H, N, and methane (CH_4) are produced in small amounts.

In the souring of milk, the formation of acid does not continue until the sugar is all exhausted. When the acidity reaches about 0.4%, milk begins to taste sour. Incipient curdling takes place at about 0.6%, and soon after this, bacterial development is checked altogether. The acid formation goes on, however, until from 0.8 to 1.0% is reached. The amount of acid formed varies considerably in different cases.¹

If the acid produced in any case is removed by neutralizing it with a carbonate, its development will begin anew, showing that it is suspended by the inability of the organisms to grow in such a medium.

Cream never contains as much acid as milk for the reason that a considerable proportion of its volume is occupied by the butter-fat which is not subject to this decomposition.

¹ Warrington, Journ. Chem. Soc., 53: 727, 1888.

It is a wide-spread belief that thunder-storms cause the premature souring of milk. Numerous experiments have been conducted along these lines, and the general conclusion is that neither the electric discharge nor shock due to thunder exert any effect on the development of acid, but that the atmospheric conditions usually incident to a thunder-storm are such as permit a more rapid bacterial growth.

The lactic acid fermentation is produced by a large number of different kinds of bacteria, although in the spontaneous coagulation of milk, it is now believed that a very widely distributed species is responsible for the most of it. This organism was first described in a complete manner by Hüppe¹ and called by him *B. acidi lactici*. Günther & Thierfelder² working on the spontaneous souring of milk in the neighborhood of Berlin found what they think is the same germ. Esten³, in this country, studied milks from thirty different localities in New England and the Middle States. He found a germ in all but two cases that agreed in general with Günther's description. Dinwiddie⁴, studying the same question in Arkansas, arrives at the same conclusion. This preponderance of evidence makes it quite probable that there is a widely distributed germ that is concerned in this change although there are numerous other forms⁵ that are associated with this type of decomposition. Conn and Aikman refer to the fact that over 100 species are already known. It is fair to presume, however, that a careful comparative study of these would show that

¹ Hüppe, Mitt. a. d. k. Gesund. Amte, **2**: 309, 1884.

² Günther & Thierfelder, Arch. f. Hyg., **25**: 164.

³ Esten, 9th Rept. Storrs Expt. Stat., p. 44, 1896.

⁴ Dinwiddie, Ark. Expt. Stat., Bull. 45, May, 1897.

⁵ Kayser, Ann. de l'Inst. Past., **10**: 737.

simply racial differences exist in many cases, and therefore, that they are not distinct species.

As a rule this class of bacteria is unable to liquefy gelatin or develop spores. On account of this latter characteristic they are easily destroyed when milk is pasteurized or heated to a higher temperature. They live under aerobic or anaerobic conditions, many of them being able to grow in either environment. According to Höft¹ spontaneous souring occurs more rapidly in vessels having a small surface exposed to the air, indicating that anaerobic activity was more pronounced.

The temperature conditions as to growth vary somewhat with different species. With most species growth occurs at 50° F., but appreciable amounts of acid are not produced until a higher temperature is reached².

While the souring of milk is a very wide-spread phenomenon, still lactic acid germs do not abound everywhere. Esten finds them abundantly in the milk-ducts but not present on hay. From the milk dealer's standpoint, this fermentation like all others, is undesirable, as it destroys the food value of milk for direct consumption. The fermented product, sour milk, has some value for cooking purposes. From this fermented product, the Armenians make a palatable drink, *matzoon*, that has considerable dietetic value in certain stomach troubles.

While these bacteria are undesirable in milk-supplies, they are essential to the production of butter and cheese. The aroma and flavor of butter is, as a rule, dependent upon their presence, and in cheese they are absolutely essential in various stages of its manufacture.

¹ Höft, *Milch Ztg.*, 26: 212, 1897.

² Kayser, *Cent. f. Bakt.*, II. Abt., 1: 436.

58. "Gassy" milks. This very common and undesirable type of fermentation is caused by the presence of a large number of different bacteria, the majority of which belong to the lactic acid group. The amount of acid formed is always considerably less than that which is present in the typical lactic formation. Besides this, there is also produced as a result of the decomposition of the milk-sugar, various gases such as CO_2 , H_2 , and methane. Accompanying these are also to be noted various

FIG. 12. A cheese made from "gassy" milk. A severe type of this fermentation.

other decomposition substances that impart to milk and its products an undesirable flavor and odor. These fermentations are particularly undesirable in cheese-making, as they are the cause of "floating" and "pin-hole" curds.¹ The swelling or huffing of green cheese is invariably due to this type of ferment action. Organisms producing this abnormal change are particularly numerous in manure particles², the colon bacillus (*B. coli communis*),

¹ Freudenreich, Landw. Jahr. d. Schweiz, p. 17, 1890.

Russell, 12th Rept. Wis. Expt. Stat., p. 139, 1895.

² Bolley, N. D. Expt. Stat., Bull. 21.

the common fecal inhabitant of the intestinal canal being able to produce large quantities of gas. In some cases gas-generating bacteria belong to the casein-dissolving group, but for the most part they attack the milk-sugar.

59. Slimy or ropy milk. The viscosity of milk is often markedly increased as a result of bacterial fermenta-

FIG. 13. Slimy milk. This milk would "string out" several feet in length.

tions. This condition varies much in intensity, in some cases the milk merely becoming viscous, when it is known as sticky or slimy; then again the viscosity may be so increased, that particles of milk, when touched, will cohere and string out in long threads, in which condition it is known as ropy, thready, or stringy. This condition in milk generally occurs during warm weather. Its presence prevents perfect creaming, as the globules of butter-fat are unable to rise, owing to the viscous nature of the fluid. Naturally it also has an injurious effect upon the quality of the cream, owing to the by-products that are formed. While a number of different species of bacteria have been more or less thoroughly studied which possess the property of rendering milk slimy, these various species are not of equal importance in affecting milk under natural conditions. The manner in which the slime-forming organisms are first

introduced into milk is of considerable importance, as a knowledge of this often enables restrictive measures to be applied directly. Marshall¹ reports an outbreak in Michi-

¹ Marshall, Mich. Expt. Stat., Bull. 140.

gan which he traced to an external infection from the udder. In another case, he found the slimy germ in the dust on the floor of a barn. Guillebeau¹ has reported a number of cases in which slime-forming organisms were traced to a diseased condition of the udder.

In Switzerland, the chief cause of ropy milk seems to be *Mic. Freudenreichii*, a large, immotile, non-liquefying coccus that grows well at ordinary temperatures. This organism is readily killed by heat, two minutes at 212° F. being sufficient, but in a dried condition it has great resisting powers.

The slimy substance formed in milk comes from various ingredients of the milk, and the chemical character of the slime produced also varies with different germs. In some cases the slimy material is merely the swollen outer cell membrane of the bacteria themselves; in others it is due to the decomposition of the proteids, but in general, the chief decomposition product appears to come from a viscous fermentation of the milk-sugar.

Normally this class is repressed by the development of the lactic acid bacteria; in fact, putrefactive processes seldom occur where the lactic organisms are in the ascendancy. In sterilized or pasteurized milk this competition is removed as the lactic forms are unable to withstand this treatment, while this enzyme-forming group survive by virtue of the spores which they possess. They gain access to the milk not infrequently from manure particles² that are derived from the coat of the animal.

60. Favorable slimy fermentations. While in this country slimy milks are considered as undesirable, yet in certain parts of Europe changes of this character are put to good use. In Holland it has long been the custom to

¹ Guillebeau, Landw. Jahr. d. Schweiz, p. 27, 1890.

² Backhaus, abs. in Expt. Stat. Rec., 10: 89.

add a starter known as "lange wei" (long or stringy whey), in the manufacture of Edam cheese to control the gassy fermentations. Weigmann has isolated from this material a slime-producing organism known as *Streptococcus Hollandicus*, which renders milk stringy and acid in a few hours at ordinary temperatures.

The clotted or thickened milk known as "taettemjöl," that is a favorite beverage in Norway, and the filmjöl (ropy milk) of Finland, is produced by adding leaves of common butterwort (*Pinguicula vulgaris*) to the milk. Weigmann¹ has isolated a bacterial form from the leaves of this plant that is similar to *St. Hollandicus* that is able to cause a slimy change in milk, so it is probable the change is due to the germs on the surface rather than to the plant itself.

61. Alcoholic fermentations. While fluids containing ordinary sugar or glucose are very apt to undergo alcoholic fermentation if exposed to the air, milk-sugar does not decompose readily in this way. The alcohol-producing ferments are mainly yeasts which class of organisms do not thrive readily in milk, although Duclaux² reports a serious outbreak in a dairy due to an organism of this class.

Among some of the Oriental tribes, alcoholic beverages are used that are made from milk. Kumiss (spelt also koumiss, kumys), is made by a fermentation of mare's milk induced by the addition of old kumiss.

A similar preparation intended for invalids is now made in this country from cow's milk by the addition of a small amount of cane-sugar and the subsequent introduction of yeast.

¹ Weigmann, Milch Ztg., Beilage, No. 48, 1889.

² Duclaux, Principes de Laiterie, p. 60.

Another alcoholic beverage that is in common use among the people of Caucasus is *kephir* (also *kefyr*, *kefir*). This is a sour, effervescent alcoholic fluid prepared from the milk of goats, cows, or sheep. The direct cause of the fermentation is the so-called kefir grain, a yellowish mass about as large as a walnut that is added to the milk. These grains are left in the milk for about a day; the milk is then poured off, and the grain dried and preserved for future use. This milk after being mixed with fresh milk is kept in leather flasks and soon a mixed fermentation sets in.

This alcoholic change has been studied considerably from a biological point of view¹, but even yet is not thoroughly understood. It is evidently a mixed fermentation, and one in which no single organism can produce all the essential ingredients. The sugar of milk is partially converted into alcohol, but at the same time the casein is coagulated and digested to a certain extent, so that the process is quite complicated. Some of the organisms isolated have been found to be able to produce a change allied to that seen under natural conditions. A yeast form is probably the main cause of the alcoholic fermentation, while bacteria change the other constituents of the milk.

62. Butyric acid fermentations. The fermentation characterized by the production of butyric acid is also a class fermentation that is caused by the action of a number of different aerobic and anaerobic bacterial species. This decomposition process is common in boiled milk and as a secondary fermentation in sour milk.² For a long time it was thought that the butyric acid change in sour milk was a continuation of the lactic fermentation, but

¹ Freudenreich, Landw. Jahr. d. Schweiz, 10: 1, 1896.

² Hofmann, Ann. de Sci. Nat., 11: 5, 1869.

it is now believed that these organisms find more favorable growth not so much on account of the lactic acid formed as in the absence of dissolved oxygen in the milk which is consumed by the sour milk organisms.

Most of the butyric class of bacteria are spore-bearing, and hence, they are frequently found in boiled or sterilized milk. The by-products formed in this series of changes are quite numerous. In most cases, butyric acid is prominent, but in addition to this, other organic acids as lactic, succinic, and acetic are produced, likewise different alcohols. Concerning the chemical origin of butyric acid there is yet some doubt. Duclaux¹ affirms that the fat, sugar, and casein are all decomposed by various forms. In some cases, the reaction of the milk is alkaline, with other species it may be neutral or acid. This type of fermentation has not yet received the study it deserves.

In milk these organisms are not of great importance, as this fermentation does not readily gain the ascendancy over the lactic bacteria. It has been supposed until recently, that the rancidity of butter was attributable to the action of these ferments, but the general belief now is that this is a purely chemical change that may be influenced but not caused by these organisms.²

63. Bitter milk. A bitter taste may be imparted to milk in a variety of ways. In some cases it is due to improper feeding caused by eating herbs such as lupines, wormwood, or chicory. Then again, at certain periods of lactation, a bitter salty taste is occasionally noted in the milk that is peculiar to individual animals.

A considerable number of cases of bitter milk have, however, been traced to bacterial origin. For a number of years the bitter fermentation of milk was thought to

¹ Duclaux, *Principes de Laiterie*, p. 67.

² Duclaux, *Compt. rendu*, **102**: 1077.

be associated with the butyric fermentation, but Weigmann¹ showed that the two conditions were not dependent upon each other. He found that the organism which produced the bitter taste acted upon the casein.

Conn² found a coccus form in bitter cream that was able to impart a bitter flavor to milk. The writer separated a lactic acid species from milk that also possessed a similar property. Sometimes a bitter condition does not develop in the milk, but may in the milk products later. Freudenreich³ separated a micrococcus from cheese that was found to be the cause of bitterness.

Cream ripened at low temperatures not infrequently develops a bitter flavor, showing that the optimum temperature for this type of fermentation is below the typical lactic acid change.

It has long been a question how to account chemically for the bitter taste in milk. Various ideas have been advanced, but Freudenreich has demonstrated in one case that a bitter substance is formed in the milk that can be isolated by adding alcohol.

Milk that has been cooked is likely to develop a bitter condition. The explanation of this is that the bacteria producing the bitter substances usually possess endospores and that while the boiling or sterilizing of milk easily kills the lactic acid germs, these forms on account of their greater resisting powers are not destroyed by the heat.

64. "Sweet curdling" and digesting fermentations. Not infrequently milk instead of undergoing spontaneous souring curdles in a weakly acid or neutral condition, in which state it is said to have undergone

¹ Weigmann, *Milch Ztg.*, 1890, p. 881.

² Conn, *Rept. Storrs' Expt. Stat.*, p. 158, 1890.

³ Freudenreich, *Fühl. Landw. Ztg.*, 43: 361.

“sweet curdling.” In some cases the curdled casein may remain intact; in others it steadily diminishes in volume, a turbid and somewhat colored watery fluid separating from it. In this stage, the milk is said to have “wheyed off.”

The physical appearance of milk undergoing these fermentations is materially different from that where normal souring occurs. The curd of sour milk is hard and breaks with a fractured surface, while the coagulum in these cases is soft and somewhat slimy. In the later stages of the digestive process, the milk assumes a watery appearance.

These fermentations assume two phases: 1. Curdling followed by a subsequent digestion of the casein; 2. Digestion or peptonization of the casein without any apparent previous coagulation.

A great variety of bacteria are able to participate in these changes, particularly those belonging to the class represented by the hay and potato bacilli. This group of bacteria as a rule are able to liquefy gelatin, a fermentative change of a similar nature to the digestion of the casein.¹ In fact they are frequently referred to as casein ferments. The characteristic of these fermentations is the production of certain unorganized ferments or enzymes that have the power of acting on the proteid molecule independent of vital activity. The two ferments that are best known are the rennet or curdling enzyme, and the tryptic or digesting enzyme. As a rule any organism that possesses the digestive power, first² causes a coagulation of the casein in a manner comparable to rennet. Conn² has separated this enzyme in a relatively pure condition, and Fermi³ has isolated the digestive

¹ Sterling, *Cent. f. Bakt.*, II. Abt., 1: 473.

² Conn, 5th Rep't. Storrs' Expt. Stat., 196, 1892.

³ Fermi, *Arch. f. Hyg.*, 14: 1, 1892.

principle from a number of different species. Duclaux¹ has given to this digesting enzyme the name *casease* or cheese ferment. These isolated ferments when added to fresh milk possess the power of causing the characteristic curdling and subsequent digestion quite independent of cell development. The quantity of ferment produced by different species differs materially in some cases, the amount of rennet ferment being so imperceptible as to be obscured in the reaction by the digestive process.

In these fermentations, the chemical transformations are profound, the complex proteid molecule being broken down into albumoses, peptones, amido-acids (tyrosin and leucin), and ammonia as well as fatty acids.

Not infrequently these fermentations gain the ascendancy over the normal souring change, but under ordinary conditions they are repressed, as they are unable to tolerate the lactic acid group. They are, however, present in all milks to a greater or less extent, as can be seen from the fact that boiled, sterilized, or pasteurized milks invariably undergo this type of fermentation on account of the resistance of the spore-bearing species that remain in the milk after the lactic forms are killed.

They are present in milks to a larger extent in summer than winter, on which account it is much more difficult to sterilize milk thoroughly during this season. Germs of this class are not only undesirable if they gain the ascendancy in milk, but where milk is made up into cheese, considerable loss occurs from the digestion of the casein, the peptonized portions being lost in the whey.

65. Soapy milk. Weigmann and Zirn² isolated from a milk having a soapy flavor, a specific germ, *B. lactis saponacei*, capable of imparting a taste of this sort. Milk

¹ Duclaux, *Le Lait*, p. 121.

² Weigmann and Zirn, *Milch Zeit.*, 22: 569.

affected in this way foams readily, and is abnormally slow in souring. They traced the cause of the trouble in one case to the infection of the milk from the straw that was used for bedding; in another, it was present in the hay. Marshall¹ in this country has also isolated an organism of this sort that acts upon casein and albumen.

66. Bloody or red milk. This condition often arises from the actual presence of blood in the milk due to some wound in the udder. The ingestion of certain plants, as sedges and scouring rushes, is said to cause a bloody condition in milk; madders impart a reddish tinge on account of a coloring matter absorbed. These instances can always be separated from bacterial troubles, because if due to these sources the color will be noted at the time of milking.

There are several chromogenic or pigment-bearing bacteria that have been isolated from milk, that have the power of turning milk red, although this change is so slow that it does not amount to much in dairy practice. The most widely known form able to bring about this change is *Bacillus prodigiosus*, the so-called "bleeding bread" bacillus. It is interesting from a historical standpoint, inasmuch, as it has been found to be the cause of the bloody bread that has been the source of much superstitious fear. Its growth in milk is marked by the production of a coloring matter that is diffused throughout the milk, especially near the upper surface. Free contact with oxygen is required to produce the characteristic pigment. *Bacillus lactis erythrogenes* (bacillus of red milk), is another form that grows easily in milk, producing a red color, but it has the curious property of being able to form the color only in the dark, and in milk

¹ Marshall, Mich. Expt. Stat., Bull. 146, p. 16.

that is not strongly acid in its reaction. When grown in the light, this germ forms a yellow pigment. In milk, the casein is slowly precipitated and gradually dissolved. It thrives at a high temperature, from 80°–95° F.

Other cases of red milk have been reported that have been traced to other germs.¹ Menge² found a red *sarcina* that was the cause of trouble in a milk-supply. Here in this country, it is not at all uncommon to find on old milk, red patches that are caused by the growth of a yeast-like germ, *Saccharomyces glutinis*, that is often found in the air.

67. Blue milk. Blue milk is historically a better known disease than almost any other trouble in milk. As long ago as 1838, Steinhoff showed that the trouble was communicable from one lot of milk to another. It manifests itself in the course of one to three days by the appearance of isolated flecks of bluish or grayish color on the surface of the milk. In fresh milk that is only slightly acid, the gray tints prevail, but as the amount of lactic acid increases in the milk, the blue coloration becomes more marked. So far only one form (*Bacillus cyanogenus*) is known that is able to produce this change. It does not materially affect the milk, but the butter made from infected cream has very poor keeping qualities. In Mecklenburg an outbreak of this disease once persisted for a period of several years³. Heim⁴ found that this bacillus was especially resistant toward drying, or the influence of chemical agents like soda and potash, but a temperature of 176° F. for a moment sufficed to kill it.

¹ Keferstein, Cent. f. Bakt. I Abt., 21: 177.

² Menge, Cent. f. Bakt., 6: 596.

³ Fleischmann, Book of the Dairy, p. 51.

⁴ Heim, Arb. a. d. Kais. Ges. Amte, 5: 518.

68. Other kinds of colored milk. Two or three chromogenic forms producing still other colors have been found in milk. Schroeter discovered in a sample of cooked milk, a peculiar form (*Bacillus synxanthus*) that produced a citron yellow appearance and which precipitated and finally dissolved the casein. Adametz, Conn, and List have described other species that confer tints of yellow on milk. Some of these are bright lemon, others orange, and some amber in color.

Still other color-producing bacteria, such as those that produce violet or green changes in the milk have been observed. In fact, almost any of the chromogenic bacteria are able to produce their color changes in milk as it is such an excellent food medium. Under ordinary conditions, these do not gain access to milk in sufficient numbers so that they modify the appearance of it except in occasional instances.

69. Treatment of milk fermentations. Attention has already been drawn to the distinction that should be made between taints due to fermentative action caused by the absorption of some pre-existing odor. In treating any abnormal fermentation, the attempt should first be made to locate the cause of the trouble. In most instances where the difficulty is due to bacteria, it is caused by foreign matter gaining access to the milk. Scrupulous cleanliness will therefore in most cases eliminate the trouble and the suggestions made in Chapter IV as to the methods of preventing bacterial infection will be helpful in this connection. So efficacious is this course that cleanliness in every detail in dairy pursuits is almost a panacea for troubles and taints of all sorts that occur in milk.

If the taint is recognized in the mixed milk of the herd, it is necessary to ascertain, first, whether it is a general

trouble, or whether it is restricted to one or more animals. For this purpose the fermentation or curd test in some form or other is invaluable. Often the whole milking may be infected from the milk of a single animal, as is frequently the case in udder inflammation.

To prove whether the trouble is a general or incidental one, can easily be done by separating the milk of the different cows, or if this is not feasible by massing that of a few together, and so gradually narrowing down the number.

Where the trouble is a general one, and is not due to the spreading of the infection from a local source, the fault is usually to be traced to some error in handling the whole mass of milk. Imperfectly cleaned cans that are used in setting the milk often contaminate the entire lot; then, too, noxious germs derived from the coat of the animal often gain access to the milk. The herd, especially in the late summer, when the upland pastures are dry and the grass is short, are generally pastured on marsh or lowland fields. The stock seek the low places—frequently slime-covered mud-holes, and in passing through these, their coats are fouled with the scum and slime that is filled with putrefactive forms of bacteria.

Sometimes the source of the filth may be in the barn itself. Dirty stalls filled with moist and decaying matter may be the means by which the milk is often seeded, or it may come from manure particles that contain putrefactive organisms in abundance. Generally where the source of contamination can be discovered, it will be an easy matter to get rid of the obnoxious fermentation by using physical means of disinfection such as steam or hot water. In some cases pasteurizing the milk is of material help. Chemical disinfection can sometimes be employed advantageously, but the application of these agents should

be confined to the treatment of surroundings, rather than used in the milk itself.

70. Overcoming taints by use of starters. Another method that is often fruitful, is that which rests upon the inability of one kind of bacteria to grow in the same medium in competition with other species.

Some of the undesirable taints in factories can be in large part controlled by the introduction of starters made from certain organisms that are able to obtain the ascendancy over the taint-producing germ. Such a method is commonly followed when a lactic ferment, either a commercial pure culture, or a home-made starter, is added to milk to overcome the effect of gas-generating bacteria.

A similar illustration is seen in the case of the "lange wei" (slimy whey), that is used in the manufacture of Edam cheese to control the character of the fermentation of the milk.

This same method is sometimes applied in dealing with certain abnormal fermentations that are apt to occur on the farm. It is particularly useful with those tainted milks known as "sweet curdling." The ferment organisms concerned in this change are unable to develop in the presence of lactic acid bacteria, so the addition of a clean sour milk as a starter restores the normal conditions by giving the ordinary milk bacteria the ascendancy.

71. Chemical disinfection. In only exceptional instances will it be necessary to employ chemical disinfectants to restore the normal conditions. Of course with such diseases as tuberculosis, very stringent measures are required, as they are such a direct menace to human life, but with these abnormal or taint-producing fermentations, care and cleanliness, well directed, will usually overcome the trouble.

In case it becomes necessary to employ chemical substances as disinfecting agents, their use should always be preceded by a thorough cleansing with hot water so that the germicide may come in direct contact with the surface to be disinfected.

It must be borne in mind that many chemicals act as deodorants, *i. e.*, destroy the offensive odor, without destroying the cause of the trouble.

Sulfur is often recommended as a disinfecting agent, but its use should be carefully controlled, otherwise the vapors have but little germicidal power. The common practice of burning a small quantity in a room or any closed space for a few moments, has little or no effect upon germ life. The effect of sulfur vapor (SO_2) alone, upon germ life is relatively slight, but if this gas is produced in the presence of moisture, sulfurous acid (H_2SO_3) is formed, which is very much more efficient. To use this agent effectively, it must be burned in large quantities in a moist atmosphere (three lbs. to every 1,000 cubic feet of space) for at least twelve hours. After this operation, the space should be thoroughly aired.

Formalin, a watery solution of a gas known as formaldehyde is a new disinfectant that recent experience has demonstrated to be very useful. It may be used as a gas where rooms are to be disinfected, or applied as a liquid where desired. It is much more powerful in its action than sulfur, and it has a great advantage over mercury and other strong disinfectants, as it is not so poisonous to man as it is to the lower forms of life.

Bleaching powder or *chloride of lime* (CaCl_2) is often recommended where a chemical can be advantageously used. This substance is a good disinfectant as well as a deodorant, and if applied as a wash, in the proportion of four to six ounces of the powder to one gallon of water,

it will destroy most forms of life. In many cases it is inapplicable on account of its odor.

Corrosive sublimate (HgCl_2) for most purposes is a good disinfectant, but it is such an intense poison that its use is dangerous in places that are at all accessible to stock.

For the disinfection of walls in stables and barns, common thin *whitewash* (CaOH) if made from freshly burned quicklime is admirably adapted. It possesses strong germicidal powers, increases the amount of light in the barn, is a good absorbent of odors, and is exceedingly cheap.

Carbolic acid, creosote, and such products, while excellent disinfectants, cannot well be used in factories on account of their odor.

For gutters, drains, and waste-pipes in factories, *vitriol salts* (sulfates of copper, iron, and zinc) are often used. These are deodorants as well as mild disinfectants.

CHAPTER VI.

DISEASE-PRODUCING BACTERIA IN MILK.

73. Milk a medium for pathogenic bacteria. Not only is milk an excellent food for fermentative bacteria, but a not inconsiderable number of disease organisms also find in it a suitable substratum for development. In so far as these occur in milk under natural conditions, they become a serious menace to public health. Statistical evidence shows that quite a large number of epidemics have been traced to a contaminated milk-supply, which has served as a vehicle for the transmission of different disease organisms. The following table, prepared by Freeman, represents the outbreaks that have occurred since 1880.

Number of disease epidemics traced to contaminated milk-supply.

Typhoid fever	53	Foot and mouth disease.....	2
Scarlet fever.....	26	Throat trouble.....	3
Diphtheria	11	Cholera ..	1

While such evidence can only be approximately correct, still the data at hand is overwhelming as to the relation of disease bacteria to milk.

These organisms may be considered under the following heads:

1. Pathogenic bacteria causing diseases common to man and beast, which may be transmitted directly to man through the medium of the milk, as in tuberculosis.

2. Pathogenic bacteria that can thrive in milk under saprophytic conditions, and which gain access to it subsequent to its withdrawal, as in typhoid fever.

3. Saprophytic bacteria that can form toxic or poisonous substances in the milk itself or in the intestine after it is ingested, as in certain types of cholera infantum.

A. DISEASE BACTERIA DERIVED DIRECTLY FROM AFFECTED ANIMAL.

74. Tuberculosis. Of those diseases that are communicable from the animal to man by means of the milk, tuberculosis is by far the most common. This term is now used to indicate a number of maladies that have heretofore been claimed as separate diseases and which affect warm-blooded animals. It is now known that these different manifestations are all caused by the growth of the tubercle bacillus, which was discovered by Koch in 1882. In this connection, reference can only be made to the bovine type of the disease, and the relation that this bears in milk and dairy products to the human race. The disease is caused by the same germ whether it is present in the human being or the lower animals,¹ and the danger of infection exists in the transmission of the virus from one to the other.

The organism causing this disease is remarkable for the narrow temperature limits within which growth will take place, the minimum being, according to Koch, 86° F., while the maximum is 104° F.

This fact is of importance as it indicates that the tubercle bacillus is unable to develop, under normal temperature conditions, in milk after it is drawn. The organism withstands drying readily; in fact, by virtue of this property, it is most widely disseminated, as the tubercular material is discharged from the diseased animal and is distributed in a dried condition in the dust.

¹ Th. Smith has recently determined that there are certain varietal differences between these two types of tuberculosis.

Putrefaction and decomposition even, will not quickly destroy its vitality. Direct sunlight is, however, an efficient disinfecting agent, and, even in diffused light the bacilli are destroyed in a few days.

75. Prevalence among cattle. Tuberculosis not infrequently affects many species of warm-blooded animals, but it is particularly prevalent among cattle in certain regions of the world.

The recent introduction of the tuberculin of Koch as an aid in the diagnosis of the disease in cattle has shown the trouble to be more widely spread than was at first believed, but sufficient data have not yet been collated to enable an accurate estimate to be made.

In Denmark the percentage of affected animals has been shown by Bang to be very high, ranging from 30-40%. Concerning the distribution of the disease in this country, data is still very meager. In 15,000 head tested under the auspices of the U. S. Dept. of Agriculture, 19% showed a reaction. Unquestionably it is more widely disseminated, and a larger percentage of stock are affected in the older dairy regions of the east than in the west. The amount of disease among the range stock of the western plains is relatively small.

76. Tuberculin test. The tuberculin test is made by injecting into the animal a small quantity of a liquid known as tuberculin, which is a sterile glycerine extract of the growth products of the tubercle bacillus. The results obtained by the use of this test show that it is far superior to the physical methods of examination, it being possible to detect the disease in its earliest stages. When tuberculin is inoculated into an animal it causes a febrile reaction in those cases affected with this disease; the rise in temperature usually exceeding the average normal

temperature three or four degrees; in healthy animals only a slight rise occurs. A positive reaction should be at least from $2.2-2.5^{\circ}$ F. above average normal, and should be maintained with some fluctuations for several hours. Animals in the later stages of the disease sometimes do not respond, but when the test is properly applied, it is on the whole a most satisfactory aid in diagnosis.

77. Relation of disease organism to milk-supply.
While tuberculosis is widely distributed among cattle, it

FIG. 14. Side view of tuberculous udder, showing extent of swelling in single quarter.

must not be supposed that the milk of all reacting animals is infected with the specific organism. In a small percentage of cases, the udder and related tissues are affected, in which case the milk generally possesses in-

fectious qualities. In the early stages of the disease, the milk appears perfectly normal; as the disease progresses the milk assumes a watery appearance and changes in color. A diseased condition of the udder can usually be diagnosed where a hard, painless swelling of the udder occurs that is confined to one quarter. Udder inflammations other than tubercular are generally accompanied with pain. Milk from animals having udder tuberculosis should be unconditionally rejected for food purposes.

Woodhead¹ found in fourteen out of nineteen cases that milk from a tuberculous animal or the sediment from it was sufficiently infectious to produce the disease in guinea-pigs inoculated with small quantities of it.

The writer² found one case in which a single cc. of milk from a diseased animal sufficed to kill a rabbit inoculated with it. In this same case, the bacilli were also demonstrated microscopically in the milk.

Sometimes the udder contains tubercle bacilli and still does not show any external symptoms of the disease. Bang³, Ernst⁴, and others have demonstrated that in quite a percentage of animals with apparently healthy udders, the milk possesses infectious properties.

Where the disease is localized in the lungs, the danger from the milk is probably but slight, but it is often impossible to determine the exact condition of the disease in the animal. Even where the disease is local there is a danger that a previous chronic condition may suddenly become acute.

With adults in normal health, the danger from an infected milk-supply is undoubtedly greatly minimized, as

¹ Woodhead, Trans. 7th Intern. Hyg. Cong., London, 1891.

² Russell, 11th Rept. Wis. Expt. Stat., p. 196, 1894.

³ Bang, Cong. for Tuberculosis, 1888, p. 70.

⁴ Ernst, Hatch Expt. Stat., Bull. 8, 1890.

the healthy digestive tract is relatively insusceptible, but with infants and invalids the case is far different. The difficulty of proving an infection in this way is very great, as it is impossible to exclude the more common infection by the air; yet a number of well authenticated cases have been traced to this source. The presence of the disease in many cases in the digestive organs alone is inexplicable except in this way. It is impossible to determine what percentage of human tuberculosis is acquired in this way, but even though the amount be small, it devolves upon us to restrict this possibility in every conceivable way. Not alone should it be considered from the human standpoint, but from the point of view of successful animal industry must every measure be taken to repress and extirpate this disease from our herds.

Tubercle bacilli in other milk products. If milk containing numerous tubercle bacilli is made up into different products the specific organism still is able to exist in such for a considerable length of time. Heim¹ found that tubercle bacilli were able to live in butter for a month and in cheese for a fortnight, but the probability of infection occurring in this way is very slight as the quantity of these materials consumed at any one time is not large.

78. Methods of treating milk. The possible danger from tuberculosis being spread by a contaminated milk-supply may be greatly diminished or entirely eliminated in the following ways:

1. *Dilution.* To produce infection it requires the simultaneous introduction of a number of organisms. Bollinger and Gebhardt² showed that milk from tuberculous animals that would produce the disease in guinea-

¹ Heim, Arb. a. d. kais. Gesundh., 5: 294.

² Bollinger, Tagebl. d. 62 Versamm. Deutsch. Naturf., Sept. 1889.

pigs and rabbits was innocuous when diluted with healthy milk to 50–100 times its volume. Therefore, there is less danger in mixed herd milks, than in that of a single cow, unless it is positively known that she is unaffected with the disease.

2. *Destruction by heat.* If milk containing tubercle bacilli is boiled, the disease organism is destroyed. The same result may be accomplished by pasteurizing if this process is correctly carried out. A temperature of 155°–160° F. for fifteen to twenty minutes is sufficient for this purpose.

3. It has been suggested that inasmuch as centrifuging seems to throw out many of the tubercle bacilli in the slime¹ that this method of purification could be used, but Moore² has shown that no reliance can be placed on this method.

79. Foot and mouth disease. This disease is distinctively an animal malady, but it is also transmissible to man, although it is not often fatal. Hertwig³ records some experiments made on himself. After drinking milk from an animal suffering from this disease, the mucous membrane of the mouth became swollen, small vesicles appearing in the mouth. Not an inconsiderable number of epidemics have been reported that have been traced to this source.⁴ Calves readily acquire the disease suckling affected mothers. The law in Prussia since 1894 requires that the milk from all infected herds shall be heated to 194° F., for fifteen minutes before it is taken from the dairy.

¹ Bang, *Milch Zeitung*, 1893, p. 672.

² Moore, *Year Book*, Dept. of Agriculture, p. 432, 1895.

³ Ziemssen, *Cyclop. of Prac. of Med.*, 3: 521.

⁴ Freeman, *Med. Record*, March 28, 1896.

80. Other diseases. There are a number of other bovine diseases such as anthrax,¹ lockjaw,² and hydrophobia in which it has been asserted that the virus of the disease is transmissible to man, but in most cases of these diseases the secretion of the udder soon becomes affected, so that no danger need be apprehended unless such milks are wilfully sold for human food. The only safe rule is to reject milk coming from animals that show any signs of sickness, even though the disease may be one that is not common to man.

B. INFECTION OF MILK BY DISEASE BACTERIA AFTER IT IS DRAWN.

81. Typhoid Fever. This disease stands next to tuberculosis in importance in its relation to milk. The organism producing this fever does not develop in the animal itself, consequently, no danger need be apprehended from milk, if it is properly cared for after it comes from the cow. The typhoid fever bacillus, however, finds in raw milk such favorable conditions for development, that if it is once introduced, it is often able to thrive for a considerable length of time. The disease usually spreads by means of the water-supply, yet a large number of epidemics have been traced directly to milk as the original and only source of infection. A striking case is the Stamford, Conn., epidemic of 1895. 386 cases of the disease developed in six weeks, and of this number, over 97% came from a single milk-supply. The milk was infected by rinsing out the cans with water from a shallow contaminated well.

The contamination of milk by this disease has been

¹ Stohmann, *Milch u. Mol. produkte*, p. 382.

² Marx, *Deutsche Viertelsjahr. f. öffentl. Gesundheits. Pflege*, 20: 444, 1890.

traced to numerous causes, among which the following are most important:

1. Direct transmission to the milk from person convalescing from the disease.

2. Indirect transmission by milker also serving as nurse to patient.

3. Indirect transmission through polluted water-supply where same is used for cleansing milk vessels.

All of these sources of infection can be readily governed with a little care. If typhoid fever is present in the family of a milk-handler, especial care should be taken that no one has any access to the patient that has anything to do with the milk. In case of disease in family, all water used in cleaning cans should first be boiled or else secured from a source in which contamination is impossible. Wells in vicinity of dwellings are often infected with disease germs that are derived from the excreta, and if such water is used, infection of milk-supply is possible.

In some cases, the disease has been spread by infecting a general creamery supply, the contaminated skim-milk distributing the virus of the disease. In the Welly epidemic in England in 1893, twenty-three cases appeared among the patrons of a single factory.

The typhoid fever organism is able to thrive in milk for a considerable time (20-35 days according to Heim)¹, on account of its tolerance toward weak acids. Even in butter and cheese these organisms are able to live, but the conditions are so unfavorable and the probability of infection so slight as to practically eliminate such a source.

82. Cholera, diphtheria, scarlet fever, etc.

1. *Cholera.* Milk also functions as a medium for the

¹ Heim, Arb. a. d. k. Ges. Amte, 5: 303.

transmission of cholera, although the danger from this source is somewhat minimized on account of the inability of this germ to thrive luxuriantly in acid fluids. Kitasato¹ found that the cholera germ could live in raw milk from one to four days, depending upon the amount of acid that is present in the milk. In boiled, or sterilized milk, cholera can grow more freely as the acid-producing organisms are destroyed by the heat used. In butter it has been found after four or five days, but the acid reaction restrains its growth and soon kills it.

Cholera epidemics in India have more than once been traced directly to contaminated milk. Simpson² records a case of infection where ten sailors out of twenty-four partook of some milk that had been purchased from a milkman, in whose district cholera had recently broken out. Of these ten, four died, five were severely sick, and one that used the milk sparingly was slightly ill. Investigation proved that the milk had been adulterated with water that had been taken from an open pool in the infected district.

2. *Diphtheria*. This is another contagious disease, the specific organism of which finds in milk favorable conditions of growth, and there is abundant evidence to indicate that milk functions as a transmitter of contagion if it once becomes contaminated with the same. Whether the animal can actually serve directly to spread the disease is yet a question. Klein³ affirms as a result of animal inoculations that diphtheria will develop in the cow, attacking among other organs the udder, and so infecting the milk, but both Abbott⁴ and Vladimirow⁵ failed to confirm these observations.

¹ Kitasato, Arb. a. d. Kais. Gesund. Amte, 1: 470.

² Simpson, London Practitioner, 39: 144 (1887).

³ Klein, 19th Rept. Soc. Gov. Bd. (Great Britain), 1889, 167.

⁴ Abbott, Vet. Mag., 1: 17.

⁵ Vladimirow, Arch. sci. biol. Inst. Med. St. Petersburg, p. 84, 1892.

3. *Scarlet Fever*. The exact relation of scarlet fever to milk is still harder to study, inasmuch, as the specific organism of this disease has not yet been isolated. Nevertheless, the evidence in a constantly increasing number of epidemics is so strong in favor of the view that dissemination of disease occurs by means of polluted milk, that the relation may be considered as practically established. Klein¹ holds that a certain eruptive udder disease of cattle is the same as scarlet fever, and that it is communicable to man. His conclusions, however, are not generally accepted, although it is regarded as thoroughly proven that the disease may be propagated in the milk after it is drawn.

C. POISON-FORMING BACTERIA IN MILK.

83. Toxic or poisonous milk. Milk not infrequently acquires poisonous properties by virtue of the development of various putrefactive bacteria that form poisonous by-products as a result of decomposition processes. In some cases the toxic products are formed in the milk outside of the body. When such milk is ingested, symptoms of poisoning soon appear. In other cases, certain bacteria found in the milk develop in the intestine producing a toxic effect.

Milk contaminated with filth frequently contains putrefactive bacteria that form substances that cause gastric and intestinal troubles, especially in infants. Vaughan asserts that the larger number of cases of summer diarrhoea are due to putrefactive organisms in the milk. The much higher mortality of bottle-fed, compared with breast-fed infants is undoubtedly attributable to the action of these microbes in milk².

¹ Klein, 7th Intern. Hyg. Cong. (London), p. 130, 1891.

² Baginsky, Hyg. Rund., p. 176, 1895.

Many of the cases of poisoning due to ice cream, cheese, and milk come from such a source. Improperly handled milk may be made up into these various products, and in this condition, the poisonous principle remain unchanged for a considerable time. Vaughan¹ has isolated a poisonous substance from these materials, calling it *tyrotoxicon* (cheese poison). Our knowledge concerning the way in which it is produced is, as yet, very vague, but from his most recent researches², it is quite probable that the organisms causing these troubles are putrefactive forms that gain access to the milk where it is improperly kept.

¹ Vaughan, 9th Intern. Hyg. Cong., (London), p. 118, 1891.

² Vaughan and Perkins, Arch. f. Hyg., 27: 308.

CHAPTER VII.

PRINCIPLES OF MILK PRESERVATION.

84. Keeping quality. Where milk is consumed in the form of milk or cream, it is desirable to retard or inhibit the fermentative changes as much as possible. In the preceding chapters, the direct relation which exists between the keeping quality of the milk and the germ life of the same has been indicated, so that it is evident, in order to perfectly preserve milk, the bacteria must either be prevented from gaining an entrance, or destroyed after they have once established themselves. Bacteria are so widely distributed, and milk is such a nutritious medium for their development, that to *entirely* prevent their entrance and growth is not feasible. Scrupulous care in the dairy both before and after the milking, will however, do much to reduce the germ content of milk as is seen in the "sanitary" or "certified" milk that is being put on the market in some cities. These milks are drawn under the most careful conditions, and then held during distribution in such a way as to retard the course of the fermentative changes in a marked degree. Milk so treated will remain sweet for several days, which period is sufficiently long for practical purposes. Such milk cannot well be produced in herds that are not under one management as the degree of care necessary to success is not often exercised unless under the direct supervision of a competent manager.

85. Preservation of milk. In considering methods of milk preservation, reference will only be made to

those that are applicable to milk intended for food. Samples destined for analytical use, such as for fat tests, etc., would be treated in a different manner. The various methods that have been suggested may be classified under two heads, those attained by the use of chemical, or by physical agents. In some methods the preservation is accomplished by the operation of both physical and chemical factors.

86. Chemical agents. Under the head of antiseptics and disinfectants, the action of different chemicals on bacterial life has been discussed¹. Those substances that are inimical to the development of bacteria are usually too strong for use in a food product as preservatives, because the protoplasm of bacteria as a rule is more resistant than other forms of living matter.

For this reason, the application of strong disinfectants like carbolic acid, mercury salts, strong acids, or alkalis is excluded. The chemical agents that are used in the preservation of milk fall into two classes.

1. Those that unite chemically with certain products of bacterial growth to form more or less inert substances in the milk.

2. Those that restrain or inhibit the development of fermentative organisms in the milk.

To this first class belong those alkaline salts such as bicarbonate of soda, etc., that combine with the acids that are formed in the milk. While salts of this sort neutralize the acidity produced by the sour milk bacteria, they do not kill these organisms, their growth again being renewed after the milk is neutralized.

As representatives of the second class, salicylic acid, boracic acid, and their derivatives may be mentioned.

¹Lazarus, *Zeit. f. Hyg.*, 8: 207.

Formalin has been extensively advertised of late, but even though this agent does not exert as prejudicial an effect on human tissues as some of the other disinfectants, yet it must be regarded with the remainder of chemical preservatives as undesirable.

These substances can be added to milk in quantities not recognizable to the taste, and they will materially increase the time that milk will remain sweet, but their general use in milk is to be strongly deprecated. In many European countries their use is prohibited entirely. A large number of the preservatives are sold under proprietary names, and as a rule for extremely high prices, but they all depend for their efficiency upon the antiseptic action of chemicals.

87. Detecting preservaline. The detection of these various preservatives in milk does not come within the general province of this work, but Farrington¹ has suggested such a simple means for the determination of the presence of preservaline and other boracic acid compounds that is so intimately connected with the development of acid that it is given here. When the acidity of a normal milk reaches 0.3–0.4%, it tastes sour, but when boracic acid is added to fresh milk, the acidity is much increased without affecting the taste. In fact, for some reason not yet explained, the acidity of the milk is increased considerably more than that due to the chemical added. Therefore milks having a high acid reaction (exceeding 0.3–0.4%) and not tasting perceptibly sour have in all probability been treated with preservatives. Failure to sour, especially when milk is kept at room temperatures for several days, is pretty conclusive evidence that chemicals have been used. The sale of these preparations for use in milk finds its only outlet with those dairymen who

¹ Farrington, Journ. Amer. Chem. Soc., Sept., 1896.

are anxious to escape the exactions that must be met by all who attempt to handle milk in the best possible method. Dirty milk drugged with these chemicals is frequently able to falsely pass as an extra good product.

88. Physical agents. The physical methods of milk preservation as a rule are much more feasible than the chemical, as they do not injure the nutritive value of the fluid to such an extent. The efficiency of physical methods of preserving milk is dependent upon the elimination or destruction of bacterial life, or at least an inhibition of their powers of growth. Some methods, such as the use of electricity, or of compressed air, or different gases have been suggested, but a practical treatment of milk with these agents has not as yet been devised. Different methods of filtration have been tried. Gravel or sand filters have been recommended for purifying milk-supplies. In these the suspended foreign matter, and a part of the bacteria can undoubtedly be removed, but the inconvenience of sterilizing such a filter would seem to be considerable. Filters of this character have, however, been satisfactorily used by the Copenhagen Dairy Co., for a number of years. Germ-proof filters, such as the Pasteur filter that are so efficient in water purification, are not applicable to milk as they clog so quickly. Recently the use of cellulose as a filtering substance has been proposed, and according to Backhaus and Comlien,¹ this is satisfactory from a bacteriological as well as a mechanical stand-point.

89. Freezing. Chilling the milk has always been practiced as a means of enhancing the keeping quality, but recently the attempt has been made to transport milk for long distances in a frozen condition, in which state bacterial growth is entirely suspended. Milk can be pre-

¹ Ber. Landw. Inst. Univ. Königsberg, 2: 12, 1897.

served best by first pasteurizing it, then freezing it and shipping in this condition.¹ Another process is Casse's method in which a quantity of milk is frozen and placed in large cans. The intervening space is then filled with fresh milk and the whole closed air tight. It is claimed this milk keeps for weeks in this condition.² One of the difficulties of simple freezing is that the milk constituents separate out so thoroughly that it is difficult to perfectly re-incorporate them upon melting.

90. Condensed milks. Milks may be preserved for an indefinite period of time by condensing them. The keeping quality of such milk often depends, however, upon the action of another principle, viz., the inhibition of bacterial development by reason of the concentration of the medium. This degree of concentration is reached in either of two ways, by the addition of sugar, or by evaporating the watery solution by boiling it down either in open air or preferably in a vacuum pan. In the milks preserved by addition of sugar, the bacteria are not necessarily destroyed but they are unable to grow on account of the concentrated condition of the medium. Such milks diluted with several volumes of water, even sterile water, not infrequently undergo the ordinary decomposition changes. In some cases, the heating process also destroys in part the contained germ life.

91. High temperatures. Heat has long been used as a preserving agent. Milk has been scalded or cooked from time immemorial to keep it. Heat may be used at different temperatures, and when so applied exerts a varying effect, depending upon temperature employed. All methods of preservation by heat rest, however, upon the use of the two following principles:

¹ *Milch Zeit.*, No. 9, 1895.

² *Milch Zeit.*, No. 33, p. 527, 1897.

1. A temperature above the maximum growing-point ($42-45^{\circ}\text{C.}$) and below the thermal death-point ($52-68^{\circ}\text{C.}$) will prevent further growth, and consequently fermentative action.

2. A temperature above the thermal death-point destroys bacteria, and thereby stops all changes. This temperature varies, however, with the condition of the bacteria being much higher for spore-bearing species.

Attempts have been made to employ the first principle, viz., prolonged heating above growing temperature, but when milk is so heated, its physical appearance is changed. The methods of heating most satisfactorily used are

known as sterilization and pasteurization, in which a degree of temperature is used approximating the boiling- and scalding-points respectively.

FIG. 15. Microscopic appearance of normal milk. The fat globules are grouped together into tiny clots. The consistency or body of cream is in part due to this characteristic.

92. Characteristics of heated milk. When milk is subjected to the action of heat, a number of changes take place in its reactions.

1. *Thinner body.* Milk, but more especially cream, heated to a temperature exceeding 150°F. , becomes much thinner, a condition due to a change in the grouping of the fat globules.¹ In normal milk, the butter-fat is massed together in microscopic clots as in fig. 15. In heated

¹ Babcock & Russell, 13th Rept. Wis. Stat., p. 73, 1896.

cream these clumps are broken down and the globules are homogeneously distributed as in fig. 16.

2. *Cooked flavor.* If milk is heated to 160° F., it acquires a cooked taste that becomes more pronounced as the temperature is further raised. The cause of this change is not well known. Usually it has been explained as being produced by changes in the nitrogenous elements in the milk, particularly in the albumen. Recently, Thoerner¹ has pointed out the coincidence that exists between the appearance of a cooked taste and the loss of certain gases that are expelled by heating. He finds that the milk heated in closed vessels from which the gas cannot escape has a much less pronounced cooked flavor than if heated in an open vessel.

3. *Fermentative changes.* Normally milk undergoes the lactic acid fermentation. If, however, it is heated to a temperature above the thermal death-point of these non-spore-bearing organisms, as is the case in pasteurizing, it does not sour but curdles by means of a rennet ferment which is secreted by those bacteria that resist the heating process.

4. *Action toward rennet.* The action of heat causes the soluble salts of lime in the milk to be precipitated, and

FIG. 16. Microscopic appearance of milk heated to 150° F. or above. The aggregation of fat globules that is present in normal milk is broken down, the globules being homogeneously distributed throughout the milk serum. This lessens the consistency of cream and makes it appear thinner.

¹ Thoerner, Chem. Ztg., 18: 845.

as the curdling of milk by rennet is dependent upon the presence of these salts, their absence in heated milks retards greatly the rennet action. The character of the coagulum is also considerably different from what it is with raw milk, being much softer and less tenacious.

5. *Hydrogen peroxid test* (Storch's test). When a small quantity of potassium iodid and starch is added to milk and the same then treated with a few drops of dilute hydrogen peroxid, normal milk will turn blue, while that heated to 175° F. or above, will not change. This reaction is due to the fact that the inherent enzymes normally found in milk are destroyed by this degree of heat, and therefore the hydrogen peroxid is not decomposed¹.

93. Sterilization. To thoroughly sterilize milk, it is necessary to render it germ free. So far as the vegetating bacteria are concerned, this can be done by an application of heat such as is used in pasteurizing, but on account of the presence of spore-bearing forms, a much higher temperature is necessary. To render milk perfectly sterile, it is necessary to heat it for about two hours at 250° F., or thirty minutes at 270° F. At the temperature of boiling-water, milk often withstands, especially during the summer months, a prolonged steaming for five to six hours.

Another method of sterilizing is that used in what is known as intermittent sterilization, a method employed by Dahl, of Norway. This consists in heating the milk to a temperature fatal to the contained bacteria, 158° F. or above, for one-half hour in order to kill the vegetating bacteria. The milk is then placed under conditions that will cause the latent spores to germinate. After this oc-

¹ V. Storch, 40th Rept. Danish Expt. Stat., Copenhagen, 1898.

Babcock and Russell, 15th Rept. Wis. Expt. Station, p. 84, 1898.

curs, a second application of heat is made on the succeeding day, and so on for three or four days. By this intermittent process, all spores are given a chance to germinate, and thus become susceptible to a relatively low heat.

These methods which necessitate so protracted a treatment are manifestly out of the question for commercial purposes unless a product is desired that will keep for a considerable period of time.

When milk is sterilized commercially, it is usually treated for a single time at a high temperature, and in some of the better types of apparatus is rendered nearly sterile, or at least the small amount of germ-life remaining is so weakened by the treatment given that development is suspended, unless the conditions are very favorable. In most cases sterilized milks acquire a more or or less pronounced cooked taste. This is more of an objection in America than in Europe, where in most cases milk is usually heated before being consumed. Apparatus of this class is usually expensive, so that the cost of sterilized milk is higher than the normal product. Machines for this purpose have not been readily adopted in this country, the tendency being to employ the following process for milk preservation.

94. Pasteurization. In this method the degree of heat ranges from 140°-175° F. and the application is made for only a limited length of time. The process was first extensively used by Pasteur (from whom it derives its name) in combatting the various maladies of beer and wine. Its importance as a means of increasing the keeping quality of milk was not generally recognized until a few years ago; but the method is now growing rapidly in favor as a means of purifying milk for commercial purposes. The method does not destroy all

germ-life in milk; it affects only those organisms that are in a growing, vegetative condition, but if the same is quickly cooled, it enhances the keeping quality very materially.

The experiments of Bitter¹ indicate that when stored at 86° F.; properly pasteurized milk will remain sweet from six to eight hours longer than raw milk. At 77° F., ten hours; at 73° F., twenty hours; at 58° F., from fifty to seventy hours. In our experience, pasteurized cream when kept in an ordinary refrigerator usually remains sweet for a period of four to six days, and sometimes even longer.

95. Requirements of Pasteurized milks. Pasteurized products should possess the following requirements:

1. Absolute freedom from disease bacteria. Fortunately the disease bacteria that are apt to be transmitted by means of the milk (tuberculosis, typhoid, etc.), do not form spores. Therefore, a proper pasteurization will destroy the virus of these diseases.

2. Ordinary milk bacteria should be diminished. Proper pasteurizing will destroy all non-spore-bearing or vegetative bacteria.

3. Improved keeping quality. This destruction of the normal milk flora will result in lengthening the time that milk will remain sweet.

4. Normal in taste and appearance. Pasteurized products should have no perceptible cooked taste. They will, however appear thinner in consistency, a condition that often leads to the belief that they contain less fat. This is not a serious objection in the case of milk, but in cream it is more apparent. It can, however, be easily remedied by the use of viscogen (98).

¹ Bitter, Zeit. f. Hyg., 8: 240, 1890.

96. Pasteurized vs. sterilized milk. For consumption within a few days, pasteurized products are as well adapted as sterilized. For export and long-keeping milk, sterilized is better. Pasteurized is less objectionable than sterilized on account of cooked taste and flavor. As to the relative digestibility, authorities disagree. For the healthy child there is probably but little choice; for infants, pasteurized is believed to be better adapted. Pasteurized products can be prepared and sold at less expense than sterilized.

97. Children's milk. Attention has already been called to the relation which exists between infant mortality and the use of cows' milk. The cause of this lies in the fact as shown by Soxhlet that breast milk is consumed directly and in a condition practically sterile, although the researches of Knochensteirn show that it is quite as impossible to secure germ free mother's milk as it is cow's milk. The method of handling cow's milk is such that germ life finds ready access and opportunity to grow, with the result that infantile disturbances are common where ordinary milk is used. Soxhlet introduced a method of sterilizing designed for family use that has proven very successful in Germany, where it is widely used.

In sterilizing or pasteurizing milk for infant feeding, it is desirable that only a sufficient quantity for each day's use be prepared at once, for any milk that is not perfectly sterile will gradually develop the latent germ life that is in the same, and as these forms belong to the peptonizing class of organisms, their presence is not desirable, as has been shown by Flügge¹.

¹ Flügge, Zeit. f. Hyg, 17: 272.

7—B.

98. Restoration of "body" of pasteurized cream.

The action of heat causes the tiny groupings of fat globules in normal milk (fig. 15) to break up, and with this change which occurs in the neighborhood of 150° F., the consistency of the liquid is diminished, notwithstanding the fact that the fat-content remains unchanged. Babcock and the writer¹ devised the following "cure" for this apparent defect. If a strong solution of cane-sugar is added to freshly slaked lime and the mixture allowed to stand, a clear fluid can be decanted off. The addition of this alkaline liquid, which they call "viscogen," to pasteurized cream in proportions of about one part of sugar-lime solution to 100-150 of cream, restores the consistency of the cream, as it causes the fat globules to cluster together in small groups.

The relative viscosity of creams can be easily determined by the following method (fig. 17):

Take a perfectly clean piece of glass (plate or picture glass is preferable, as it is less liable to be wavy). Drop on one edge two or three drops of cream at intervals of an inch or so. Then incline piece of glass at such an angle as to cause the cream to flow down surface of glass. The cream having the heavier body or viscosity will move more slowly. If several samples of each cream are taken, then the aggregate lengths of the different cream paths may be taken, thereby eliminating slight differences due to condition of glass.

99. Pasteurizing details. While the pasteurizing process is exceedingly simple, yet in order to secure the best results, certain conditions must be rigidly observed in the treatment before and after the heating process. It is a mistaken idea that any milk is fit for pasteurizing.

¹ Babcock and Russell, Bull. 54, Wis. Expt. Stat., also 13th Rept. Wis. Expt. Stat., p. 81, 1896.

The fresher and better the milk, the less likely it is to contain deleterious spore-bearing bacteria which are not destroyed by pasteurizing. The unhindered development of these germs in the milk may sometimes give rise to the production of decomposition products that are toxic or at least highly irritating in their character¹.

FIG. 17. Relative consistency of pasteurized cream, before (A) and after (B) treatment with viscogen. Cream flowing down inclined glass plate.

100. Selection of milk. Milk for pasteurizing should be as clean as possible, for in general terms, the bacteria associated with filth and dirt, more especially manure particles, belong to the spore-bearing putrefactive class.

¹ Flügge, *Zeit. f. Hyg.*, 17: 272.

For high grade pasteurizing, it is advisable to purify the milk by passing it through a separator, unless the raw product is secured under conditions that exclude the admission of dirt.

The age of the milk and the conditions under which it has been kept should also be carefully noted. Old milk is almost always richer, not only in bacterial germs but in the latent spore forms as well; so the fresher the milk the fewer bacteria it will have, and therefore the pasteurizing process will be the more complete.

101. Selecting milk by acid test. The true standard for selecting milk for pasteurization should be to determine the actual number of bacterial *spores* that are able to resist the heating process, but this method is impracticable under commercial conditions.

The following method while only approximate in its results will be found helpful. Assuming that the age or treatment of the milk bears a certain relation to the presence of spores, and that the acid increases in a general way with an increase in age or temperature, the amount of acid present may be taken as an approximate index of the suitability of the milk for pasteurizing purposes. Biological tests were carried out in the author's laboratory¹ on milks having a high and low acid content, and it was shown that the milk with the least acid as a rule was the freest from spore-bearing bacteria.

This acid determination can be made at the weigh-can by employing the Farrington alkaline tablet which is used in cream-ripening. Where milk is pasteurized under general creamery conditions, none should be used containing more than 0.2% acidity. If only perfectly fresh milk is used the amount of acid will generally be about 0.15% with phenolphthalein as indicator.

¹ Shockley, Thesis, Univ. of Wis., 1896.

Circumstances may arise that might lead to an error if this method is blindly followed. It has been pointed out that if milk is allowed to stand in rusty cans for some time its acid content is diminished materially. Biddick¹ has reported the following interesting observations. The average of nine samples of milk brought to a factory in clean cans contained .228% acidity, while that of nine other patrons brought in rusty cans contained only .134% acid. Milk kept in rusty cans is sure to contain large quantities of bacteria, even though its acid content may be low.

FIG. 18. Apparatus used in making rapid acid test.

102. Making the acid test. Fig. 18 gives the apparatus necessary to make an approximate determination of the acidity of the milk. A solution of the alkaline tablets is first prepared by dissolving same in clean soft water, one tablet for each ounce of water; thus eight

¹ Biddick, Hoard's Dairyman, July 30, 1897.

tablets to an eight ounce bottle of water. In determining the acidity in each patron's milk, a number of common white cups are used, one for each patron. Two measures full of the alkali solution are placed in each cup, and then as the milk is received at the weigh-can, one-half as much milk is added to the alkali solution in the cup,¹ and the whole gently, but thoroughly shaken. If the pink color of the alkali solution persists even faintly, it shows that there is not enough acid in the milk to neutralize the same; if it disappears altogether, leaving the milk white in color, it indicates that there is more acid in the milk than can be neutralized by the alkali of the tablet solution. Any standard desired can be chosen, but where the relation of the milk to the alkali solution is maintained in above ratio (two to one), it indicates that 0.2% acidity is present, if the alkali is completely neutralized. Extended experience has shown the necessity of selecting milks for pasteurizing that come within this standard.

103. Temperature and time limits. The time and temperature limits in pasteurizing are subject to considerable variation. The *minimum* temperature, however, must exceed the thermal death-point at which the milk bacteria are destroyed. As the tubercle bacillus is sometimes found in milk, and as it is one of the most resistant organisms in its vegetative state that is known, the thermal death-point of this germ serves as a minimum standard for efficient pasteurizing.

The determination of this point depends on the following conditions:

1. The temperature of heat used.
2. The length of exposure to the heat.

¹ Farrington, Wis. Expt. Stat., Bull. 52.

With either condition fixed, the same result is accomplished more rapidly by an increase either in temperature or duration of exposure, so that an exposure for a longer time at a lower temperature is quite as effective as a shorter exposure at a relatively higher temperature.

With the great majority of bacterial species that have been individually tested as to their thermal death-point, 140° F. for ten minutes has been found fatal. This degree of heat suffices to kill all the disease-producing bacteria that are found in milk, with the exception of the tubercle bacillus.

According to Forster, heating thirty minutes at 149° F., fifteen minutes at 155° F., or ten minutes at 167° F. suffices to destroy this germ.

The *maximum* temperature that can be employed is just below the point at which the milk will *permanently* acquire a cooked flavor. The appearance of this peculiar flavor cannot be detected with absolute accuracy, but it is not far from 158° F. It appears in milk to some extent before this temperature is reached but upon chilling disappears. It is not enough to heat milk to this temperature, but it should be maintained for the requisite length of time.

104. Subsequent chilling. The heating process destroys only the vegetative organisms, the spores resisting this temperature. To prevent the germination of these latent forms the milk should be quickly chilled, for if germination once occurs, they can develop at even low temperatures.

The following experiments by Marshall¹ are of interest as showing the influence of refrigeration on germination of spores.

¹ Marshall, Mich. Expt. Stat., Bull. 147, p. 47.

Cultures of organisms that had been isolated from pasteurized milk were inoculated into bouillon. One set was left to grow at room temperature, another was pasteurized and allowed to stand at same temperature,

FIG. 19. Diagram showing temperature changes in pasteurizing, and the relation of same to bacterial growth.

Shaded zone represents limits of bacterial growth, 10–43° C. (50–109° F.); the intensity of shading indicating rapidity of development. The solid black line shows temperature of milk during the process. The necessity for rapid cooling is evident as the milk falls in temperature to that of growing zone.

while another heated set was kept in a refrigerator. The unheated cultures at room temperature showed evidence of growth in thirty trials in an average of 26

hours; 29 heated cultures at room temperature all developed in an average of 50 hours, while the heated cultures kept in refrigerator showed no growth in 45 days with but four exceptions.

105. Preparation of utensils. After the milk is pasteurized, it must of necessity be stored and handled

FIG. 20. Steam sterilizer for sterilizing utensils (cans, bottles, etc.). St. p., steam pipe; St. p. v., vent for steam pipe; w. s., wire shelf; c, outlet cock for condensed water.

in germ free receptacles. All utensils such as cans, dip-pers, bottles, etc., must be thoroughly sterilized. For this purpose a sterilizing oven should be had which is steam fitted, so that direct steam can be used. Material of this sort after being thoroughly cleansed, should be steamed for one-half to three-quarters of an hour. Sterilized bottles should be kept protected from dust until they are used.

106. Bottling and handling the product. In bottling the product it is necessary to keep the milk protected from reinfection. It may be bottled from a large can with a bottom faucet, or, on a large scale, with commercial bottling machines that fill several bottles at once. If "viscogen" is added to restore consistency of cream, it should be done before bottling, but not before the cream is thoroughly cooled. The best bottles for the purpose are those that have a plain pulp cap. All metal fastenings or stoppers are dirt catchers and are likely to get out of order. It is our practice to heat pulp caps in paraffin, thereby rendering them more pliable and at the same time sterilizing them. Bottles sealed with hot caps in this way are tightly closed.

In delivering pasteurized products, it is always necessary to use care in handling to prevent the cream and milk from being warmed up, and thus inciting into activity the latent spores.

107. Pasteurizing apparatus. If it is desired to pasteurize milk or cream for direct consumption, the treatment differs somewhat from that when pasteurizing for butter-making is the object in view. The equipment necessary for the first type of pasteurizing may be divided into two general classes.

1. Apparatus of limited capacity designed for private family use.
2. Apparatus of sufficient capacity to pasteurize on a commercial scale.

108. Domestic apparatus. Pasteurization can be easily and efficiently done in a limited way with the addi-

FIG. 21. Sectional view of family pasteurizer, showing milk bottles immersed in water.

tion of an ordinary dairy thermometer to the common utensils found in any kitchen. Fig. 22 indicates a simple contrivance than can be readily arranged for this purpose.

FIG. 22. A home-made pasteurizer.

The following suggestions indicate the different steps of the process:

1. Use only fresh milk for this purpose.
2. Place milk in clean bottles or fruit cans, filling to a uniform level. If pint and quart cans are used at the same time, an inverted dish or piece of wood will equalize the level. Set these in a flat-bottomed tin pail and fill with warm water to same level as milk. An inverted pie tin punched with holes will serve as a stand on which to place the bottles during the heating process.
3. Heat water in pail until the temperature of same reaches 160° F.; then remove from source of direct heat,

cover with a cloth or tin cover and allow the whole to stand for half an hour.

4. Remove bottles of milk and cool them as rapidly as possible without danger to bottles and store in a refrigerator.

109. Apparatus for commercial pasteurizing. Pasteurizing as applied to the preservation of milk, originated in Germany and Denmark, where it is used largely in the treatment of skim-milk, and the heating of cream in butter-making. The type of machinery that has been devised for this purpose has therefore naturally been constructed on somewhat different principles than would have been employed where milk is treated for direct consumption. For the treatment of milk for direct consumption, it is believed that the sanitary phase of the problem demands more attention than the economic operation of the apparatus, and, therefore, it is more necessary to use a machine that pasteurizes so thoroughly that all possible disease bacteria are destroyed, than it is to secure apparatus that will permit of the handling of large quantities of milk. Pasteurizing involves considerable time and trouble, and it is better not to have the process done at all than to have it imperfectly performed.

The various types of machinery that have been suggested for this use may be grouped as follows, depending upon their method of operation¹.

1. Continuous flow machines.

2. Intermittent machines.

110. Continuous flow pasteurizers. Apparatus of this class varies much in detail, but they possess this common principle that the milk enters the machine in a

¹ For the detailed description of pasteurizing machinery, reference should be made to Monrad's *Pasteurization of Milk*, or Weigmann's *Conservierung der Milch*.

continuous stream and is generally discharged in the same way. The objection to this type of apparatus is that the time of heating cannot be regulated with any certainty, although the temperature can be controlled

FIG. 24. Hochmuth's combined heater and cooler. Cold water enters at *k* and circulates through coil, cooling the milk which is heated in upper part of ribbed surface. This warm water is used to warm the milk, and then steam is introduced through *d*.

in part by varying the speed of flow. Another objection is that the rapid heating often necessitated by these machines scalds the proteids of the milk, making them adhere to the walls of the pasteurizer.

In some of these machines (Thiel, Kuehne, Lawrence, De Laval, and Hochmuth), a ribbed surface is employed over which the milk flows, while the opposite surface is heated with hot water or steam. Monrad, Lefeldt and Lentsch, employ a centrifugal apparatus in which a thin layer of milk is heated in a revolving drum.

FIG. 25. Monrad's centrifugal pasteurizer. Milk is introduced into center through *p*, and is thrown out on the walls of centrifuge *d*, the steam being applied to the outside of the revolving drum. Reservoir *m* catches and holds the milk for a moment. Milk finally flows at *h*.

Some of these machines are suitable for heaters, if the milk was held in vats that would retain the temperature at a pasteurizing point for a sufficient length of time.

Other machines of the continuous flow type employ a

reservoir that is filled with milk and surrounded with an outer shell that contains the heating agent, steam or hot water.

Some of this class are provided with an agitator in the milk reservoir so as to hasten the equalization of temperature in the inner chamber, and at the same time keep the milk in motion in order to prevent the coagulation of the proteids.

Most of them are arranged for a continuous delivery, the milk flowing in at the lower end and displacing that already pasteurized, which flows out above into a cooler. In some of them the agitator even mixes the fresh supply with that which has already been heated, so that the efficiency of the process where milk is being treated for direct consumption, is much lessened.

Where it is constructed for continuous delivery, the length of exposure must necessarily be quite limited, and as the temperature of the milk ought not to exceed 160° F. for fear of scalding, it very often happens that the pasteurizing process is not efficient. In the case of the sour milk organisms, from the hygienic standpoint, it is of little moment, but to insure absolute freedom from disease germs, the temperature and the time of exposure must be thoroughly under control. Very few of the machines intended for this purpose have been subjected to a rigid bacteriological test, and the lack of this has allowed the introduction of many designs that may be adapted for the pasteurization of by-products intended for animal food, for which purpose many of them were originally designed, but they certainly do not deliver a product that can be relied upon for human food.

111. Intermittent pasteurizers. Inasmuch as the biological and physical requirements as to pasteurizing, necessitate milk being heated between the temperatures

of about 145° – 160° F., it is desirable that the temperature should be under absolute control. Moreover the time limit must also be known. This requires the treatment of a *definite quantity* of material for a *definite length of time* at a *definite temperature*. A fulfillment of these conditions necessitates the use of the intermittent type of apparatus, or continuous apparatus arranged so as to practically conform to the discontinuous process.

The simplest way in which these conditions can be carried out is to employ a number of shot-gun cans immersed in a tank of hot water. By means of this crude device milk or cream can be pasteurized more effectually than in many of the specially designed pieces of apparatus. Tanks surrounded with water spaces can also be used quite successfully, although from a commercial point of view apparatus of this sort is imperfect, unless it is specially designed for this purpose.

The use of the Boyd cream ripening vat has been suggested, and this fulfills the necessary conditions as to a commercial pasteurizer. The cream in this is heated by means of a swinging coil immersed in the same, through which hot water circulates.

In some of the pasteurizers, steam is introduced directly into the milk or cream, as in Bentley's apparatus. It is obvious that while such a method may be a cheaper way in which to heat the milk, still the proteids of the fluid must be scalded in part, although the temperature of the whole mass may not exceed the proper pasteurizing point.

The writer¹ in 1894 devised a tank pasteurizer that was made to conform to the bacteriological requirements. It consists of a long, narrow vat, surrounded by a water chamber, on the bottom of which is placed a row of per-

¹ Russell, Wis. Expt. Stat., Bull. 44.

forated steam pipes. To facilitate the heating of the milk, both the milk and water reservoirs are supplied with agitators having a to and fro movement. The milk stirrers are perforated so as to give an up and down

FIG. 26. Russell's pasteurizing vat.

movement, thus preventing the cream from rising to the top of the vat during the operation. The milk is withdrawn from the vat by means of a stop-cock that is placed inside of the water chamber. This cock has a circular bore so that when open, the outlet tube presents a continuous passage that can be easily and thoroughly cleaned. Placing the stop-cock within the water chamber prevents the accumulation of unpasteurized milk in the outlet tube at a point not reached by the heated water, and where it would contaminate the whole vat when the milk was withdrawn.

The Potts pasteurizer is another machine of the intermittent type that has recently been introduced that conforms to the necessary biological conditions. This apparatus has a central milk chamber that is surrounded with an outer shell containing hot water. The whole machine revolves on a horizontal axis, and the cream or milk is thus thoroughly agitated during the heating process.

112. Coolers. A speedy cooling of the heated product is essential to success in pasteurizing. Some of the machines have been devised for a combination purpose, being used for the heating and subsequent cooling of the

FIG. 27. Pott's pasteurizer.

milk. This is an evident advantage in some ways, as it lessens the amount of apparatus necessary, also the work involved in cleaning the same, but at the same time the problems of quick heating and cooling involve somewhat different principles, so that for the most economical manipulation of the product, separate pieces of apparatus are advisable where the business warrants such expense.

The simplest method of treatment in cooling is to draw off the milk in shot-gun cans and place these first in water, then in ice-water.

To cool milk most economically, two coolers should be provided. One of these can use cold water, and by the aid of good well water, the temperature can be reduced to nearly that of the water in a short time. In order, however, to lower the temperature below a point where spore germination will readily occur, milk should be chilled by the aid of ice. This may be applied in the same cooler as is used for running cold water, by supplying ice-water for the latter part of the cooling process.

To use ice economically, the ice itself should be applied as closely as possible to the milk to be cooled, for the larger part of the chilling value of ice comes from the melting of the same. To convert a pound of ice at 32°

FIG. 28. Water cooler, using either running cold water, or water and crushed ice. The milk is introduced at *i. m.* and is spread out in *m. c.* in a thin cylindrical sheet, flowing out at *o. m.* Cold water circulates through inside of milk cylinder in *w. c.*, while ice may be used in outer water chamber, *o. w. c.*

F. into a pound of water at the same temperature requires as much heat as would suffice to raise 142 pounds of water one degree F., or one pound of water 142° F. The absorptive capacity of milk for heat is not quite the same as it is with water. It fluctuates with the amount of solids in the milk, but for ordinary milk is about .85 while water is taken as a standard 1.0. Hot milk would therefore require somewhat less ice to cool it than would be required by an equal volume of water at same temperature.

In the mere melting of a pound of ice, a large part of the heat in a pound of pasteurized milk will be absorbed. To take advantage of this, the ice should be brought in close contact with the milk rather than to spend all of this absorptive capacity on cooling water which is later applied to the milk. If broken ice is used directly, it should be arranged so that the milk surrounds it, as in this way the specific capacity for heat that is latent in the ice acts on the milk instead of being radiated in part to the outside.

113. Bacteriological study of pasteurized milk.

An extended bacteriological examination of milk and cream pasteurized on a commercial scale in the Russell vat at the Wisconsin Dairy school showed that over 99.8

A

FIG. 29. Effect of pasteurizing on germ content of milk. Black square represents bacteria of raw milk; small white square, those remaining after pasteurization.

% of the bacterial life in raw milk or cream was destroyed by the heat employed, i. e., 155° F. for twenty minutes duration¹. In nearly one-half of the samples of milk, the germ content in the pasteurized sample fell below 1,000 bacteria per cc., and the average of twenty-five samples contained 6,140 bacteria per cc. In cream the germ content was higher, averaging about 25,000 bacteria per cc. When the high initial content of either milk or cream is taken into consideration, it indicates that the spore-bearing bacteria are relatively few in properly selected milk that

¹ Russell, 12th Wis. Expt. Stat. Rept., p. 160, 1895.

is efficiently pasteurized. The kinds that are destroyed are mainly the lactic acid species; those that resist the pasteurizing heat are generally of the enzyme-forming class, which in many cases are also able to liquefy gelatin. The acidity of pasteurized products always increases somewhat, but even when it is curdled, it rarely contains over 0.3–0.4 % of acid. The organisms that remain belong then to the sweet curdling type of bacteria.

While these organisms capable of resisting the pasteurizing temperature are fermentative germs, it is also important to determine if they have the power of forming toxic substances that exert a harmful effect on the animal body. Fluegge¹ has found several forms in sterilized milk that are able to produce highly toxic substances. A similar study has been made under the writer's direction on the bacteria isolated in pasteurized milk under commercial conditions. Shockley² studied thirteen different forms found in pasteurized milk, but all of them with a single exception failed to cause any disturbance when inoculated into white mice and rabbits. In the single instance, fatal results were only obtained when large quantities were inoculated.

¹ Fluegge, *Zeit. f. Hyg.*, 17: 272, 1894.

² Shockley, Thesis, Univ. of Wis., 1896.

PART III.

RELATION OF BACTERIA TO MILK PRODUCTS.

CHAPTER VIII.

BACTERIA IN CREAM AND FACTORY BY-PRODUCTS.

114. Bacteria in cream due to mechanical causes. Cream whether secured by the gravity process or by a cream separator is invariably richer in bacteria than the skim-milk of the same age. A sample of milk might contain less than 100,000 germs per cc. in the skimmed part, while the bacterial contents of the cream layer in the same would be several millions for the same unit of volume. This is largely due to the filtering out of the microbes during the process of creaming. It is a well-known fact in sewage filtration that the addition of some chemical that will cause the precipitation of certain organic elements always present in the sewage, will take out the majority of bacteria in the settling of this precipitate. The same principle seems to be operative in the process of creaming, except that the fat globules instead of sinking to the bottom rise to the surface. Gravity-raised cream is usually richer in bacteria than separator cream, because it is materially older when gathered.

In full milk separated by the centrifugal method, there are three well-marked products,—the skim-milk, the cream, and the slime that adheres to the separator bowl.

The bacterial content of each of these will vary materially in different cases.

The slime which is composed of particles having a greater specific gravity than the milk serum is thrown out on the edge of the revolving fluid by centrifugal force. This material, if examined microscopically, will be found to contain large quantities of foreign matter as well as innumerable bacteria. The fact that it rapidly undergoes decomposition is evidence of its high germ content.

The cream will almost always contain a larger number of bacteria than the skim-milk. Popp and Becker found in a sample of whole milk containing 73,000 germs per cc., the following germ contents after it had been separated: Cream, 58,275 germs; skim-milk, 21,700 germs, and the separator slime 43,900 per cc. This centripetal movement of a large number of germs with the cream indicates that they adhere to the tiny fat globules, for this peculiarity in distribution can hardly be explained on the ground of their specific gravity, as they remain in the skim-milk in considerable numbers in spite of the great centrifugal pressure.

115. Tubercle bacillus and separator slime. According to Scheurlen¹ and Bang², tubercle bacilli, if present in a milk are largely thrown out with the slime in the separating process. Moore³ found in milk artificially infected with tubercle bacilli that the separating process diminished them to such an extent that they could not be determined microscopically, but when this separated milk was inoculated into guinea-pigs, infection occurred. This indicates that while the removal was considerable, yet it was not complete enough to justify the

¹ Scheurlen, *Arb. a. d. k. Ges. Amte*, 7: 269, 1891.

² Bang, *Land. Woch. f. Sch. Hol.*, 1894, p. 47.

³ Moore, *Year-book of U. S. Dept. of Agri.* 1895, p. 432.

use of this method for the purification of infected milk. Coupled with this peculiar relation of the tubercle germ to centrifuge slime, is the fact that tuberculosis among swine is much more prevalent in Denmark and North Germany where the centrifugal process in creaming is extensively used, and where, until recently, the swine were fed the uncooked separator slime. Ostertag¹ has pointed out this condition, and has drawn attention to the numerous cases of intestinal tuberculosis in hogs.

116. Bacterial changes in cream. Although cream is numerically much richer in bacteria than milk, yet the changes due to bacterial action are so much slower that the latter product usually spoils sooner than cream. For this reason, cream will sour sooner when it remains on the milk than it will if it is separated as soon as possible. This fact indicates the necessity of early creaming, so as to increase the keeping quality of the product, and is another argument in favor of the separator process.

117. Bacteria in different creaming methods. The method used in creaming has an important bearing on the kind as well as the number of the bacteria that are to be found in the cream. The difference in species is largely determined by the difference in ripening temperature, while the varying number is governed more by the age of the milk.

1. *Primitive gravity methods.* In the old shallow pan process, the temperature of the milk is relatively high, as the milk is allowed to cool naturally. This comparatively high temperature favors especially the development of those forms whose optimum growing-point is near the air temperature. By this method the cream layer is exposed to the air for a longer time than any

¹ Ostertag, *Milch Ztg.*, 22: 672.

other, and consequently, the contamination from this source is greater. Usually, cream obtained by the shallow pan process will contain a larger number of species and also have a higher acid content.

2. *Modern gravity methods.* In the Cooley process, or any of the modern gravity methods where cold water or ice is used to lower the temperature, the conditions do not favor the growth of a large variety of species. The bacterial numbers in the cream will depend largely upon the way in which the milk is handled previous to setting. If milked with care, and kept so as to exclude outside contamination, the cream will be relatively poor in bacteria. Only those forms will develop in abundance that are able to grow at the low temperature at which the milk is set. Cream raised by this method is less frequently infected with undesirable forms than that which is creamed at a higher temperature.

3. *Centrifugal method.* Separator cream should be freer from germ-life than that which is secured in any other way. It should contain only those forms that have found their way into the milk during and subsequent to the milking, for the cream is ordinarily separated so soon that there is but little opportunity of infection, if care is taken in the handling. As a large part of the infection of fresh milk is due to the contamination from the fore milk, which usually has but a few species, separated cream, if handled with caution, will contain mainly those species that are to be found in abundance in the milk while it is still fresh.

Where milk is separated, it is always prudent to cool the cream, as the milk is generally heated before separating in order to skim efficiently.

118. Factory by-products. While the by-products in the manufacture of butter and cheese are in a certain

sense waste products, yet, all of them possess sufficient nutrient value to warrant their use as human food or in animal feeding. When handled carelessly, however, their nutritive value is much lessened by the continued fermentations that occur in them. All of these products are rich in bacteria owing either to their age or the treatment they have undergone in the manufacture of butter or cheese. It is, therefore, all the more essential that they should be kept in such a manner as to check the continued development of germ-life within them.

119. Skim-milk. Skimmed milk varies much in its bacterial content, depending upon the way in which the full milk has been treated. Milk from which the cream has been removed by the shallow setting process is usually very rich in germs, and often has so much acid that it is easily recognized by the taste. Where the cream is gathered by the aid of ice-water, the temperature is reduced to such an extent that the skimmed part is relatively poor in bacteria. Separator skim-milk is treated in such a radically different way that it is bacteriologically quite a different product. The skimmed part is separated from the fat when the milk is only a few hours old so that the opportunity for germ growth is relatively slight.

120. Buttermilk. Buttermilk contains a large amount of casein and sugar, and is, therefore, of considerable value for feeding purposes. It is usually very rich in bacteria, sometimes containing even more than ripened cream. Pammel¹ found 1,700,000 germs per cc. in buttermilk, while the organisms in butter were less than half a million. This high content is due to the age of the milk and temperature at which the cream is previously ripened.

¹ Pammel, Iowa Expt. Stat., Bull. 21.

121. Whey. This by-product of cheese-making also has considerable value for feeding purposes. Like separator skim-milk, it is drawn at a temperature that greatly favors bacterial development. Compared with the germ content of the raw milk, whey possesses fewer organisms than skim-milk or buttermilk kept under similar conditions, as a larger part of the bacteria present in the milk are caught in the coagulated curd. The presence of these in whey, if left to cool naturally, soon sours the product, as the milk-sugar is further converted into various acids. Both whey and skim-milk for feeding purposes should be carefully handled in order to get the most out of them. If the ferments are allowed to develop, the sugar is changed into various acids, the albumen undergoes putrefactive changes, and much of the feeding value is lost.

122. Treatment of whey and skim-milk tanks. The vats for factory by-products are often made of wood; consequently, they are difficult to clean thoroughly, even if that procedure is attempted. If not carefully cleansed and sterilized by steam each day, the particles of milk or whey that adhere to the walls, quickly sour, and so infect the material that is stored in the vat on the succeeding day. In this way the vat becomes a center of bacterial infection. Often, too, this already contaminated waste product is allowed to stand in cans after it is taken back to the farms until it is thoroughly soured. Such fermented food has only a minimum value, as much of its nutritive worth is gone. Vats for these by-products should be constructed from galvanized iron and arranged to empty by gravity. The vats and supply pipes should be carefully cleaned each day as much as any other part of the factory.

The trouble arising from sour whey and sour milk is

made still worse by the practice of returning these highly contaminated fluids to the farm in the same set of cans that are used for the transportation of milk. Filthy whey-vats and cans are responsible for much of the tainted milk that is seen in factories. It is necessary that the cheese-maker should have his waste-vat free from sour and half-fermented whey before he can charge *all* of the blame upon the patron who brings bad milk.

FIG. 30. A Swiss cheese factory, showing careless way in which the whey is handled. Each patron's share is placed in a barrel, from which it is removed by him. No attempt is made to cleanse these receptacles.

Fig. 30 illustrates the manner in which the whey is distributed and cared for in many factories that make Swiss cheese particularly. The hot whey is run out through the trough from the factory into a large trough that is placed over a row of barrels, as seen in the foreground. Each patron thus has allotted to him his proper share, which he removes day by day. No attempt is made to clean out these receptacles, and the inevitable

result is that they become a foul, stinking mass in hot weather. This material is usually carried home in the same set of cans in which the milk is brought and as the cans are often insufficiently cleaned, the new milk is infected with undesirable organisms. That such a custom should have grown up in the Swiss cheese industry, where on account of the sweet curd nature of the cheese, all possible precautions should be taken to insure the best quality of milk, illustrates the necessity of bacteriological principles being thoroughly instilled into the dairy industry.

123. Preserving factory by-products. Factory by-products on account of their chemical constitution and their bacterial flora soon decompose, and a considerable proportion of the nutrients are lost. This has led to the introduction in some factories of measures that tend to prolong the keeping quality of the whey and skim-milk, although the value of these products do not warrant much expense. The conditions under which skim-milk and whey are produced facilitate the rapid development of bacteria. The continued fermentation of these materials may be inhibited in part as follows:

1. Quick cooling to a temperature unfavorable for germ development.

2. Pasteurizing or scalding.

Where fed immediately, it is hardly worth while to treat these by-products, as they will remain sweet for several hours. In some factories the skim-milk is heated by introducing live steam. Where milk is pasteurized for butter-making, the keeping quality of the skim-milk is increased 24-48 hours thereby.

124. Skim-milk a distributor of disease. In some countries, notably in Denmark and Germany, tuberculosis is so common among cattle that skim-milk from

factories becomes a serious menace when fed to young stock. To prevent distribution in this manner, compulsory legislation requires that the skim-milk shall be pasteurized at a temperature of 176° F., in order to destroy the bacilli. Storch has recently devised a test that can be easily applied to milk to determine whether such treatment has been carried out. It rests upon the following principle: Milk contains a certain substance, (presumably an enzyme,¹) that decomposes hydrogen peroxid. If milk is heated to 80° C. (176° F.,) or above, this reaction ceases. When potassium iodid and starch are added to unheated milk and the same treated with dilute hydrogen peroxid, the fluid assumes a blue color due to the action of released iodine upon the starch.

A striking illustration of the danger that may come from a skim-milk-supply that is infected with disease bacteria is seen in the Welply epidemic of typhoid fever in England in 1893, where twenty-three cases of this disease developed in the families of patrons of a single factory, the milk-supply of which became infected and was spread by means of the skim-milk.

Foot and mouth disease of cattle is often disseminated in the same way. During the last decade, this disease has been very severe in certain parts of Europe. The virus of the disease can, however, be destroyed by the use of heat. The regulation of the Prussian government require milk of all diseased animals to be heated to 212° F. or to 194° F. for fifteen minutes.

¹ Storch believes it to be the unorganized ferment discovered in milk by Babcock and Russell, 14th Wis. Expt. Stat., p. 77, 1897.

CHAPTER IX.

BACTERIA IN BUTTER-MAKING.

125. Sweet and ripened cream butter. The peculiar qualities of ordinary butter are so dependent upon the relation of bacteria to cream, that in order to understand the subject aright, it is necessary to consider, first, their effect in cream. If butter is made from fresh sweet cream, it has a delicate, although unpronounced flavor that differs considerably from the usual product. With the great bulk of the commercial product, the cream is allowed to stand for a certain length of time, during which it undergoes a series of fermentations technically known as "ripening."

American consumers have become accustomed to the peculiar properties of acid cream butter, and as yet, there is but little demand for the sweet cream product. The keeping quality of the latter is relatively poor compared with that made from ripened cream, so that it is only adapted for immediate consumption.

The germ content and the quality of ripened cream butter varies materially, depending upon the way the cream is handled.

In dairy butter-making, the cream is usually gathered by the gravity process, and is ripened in various ways. In creamery butter two methods of securing cream are in vogue. The more primitive way is where the cream is separated by gravity and is taken to the factory when it is nearly ready for churning. The more modern method is to bring the whole milk to the creamery where the cream is removed by centrifugal separation. In the one

case, the fermentative changes are well under way when it reaches the central station, and as the cream is secured from a number of different persons in a partially ripened condition, it is far from being uniform in character. Where centrifugal cream separators are used, the cream is secured sweet, thereby permitting a supervision of the ripening process at the central station where the butter is made.

This control of the ripening process exerts a profound influence on the character of the fermentation that takes place, and naturally modifies the product. The uniformity of methods employed necessarily has a tendency to render the product more uniform.

A. BENEFICENT ACTION OF BACTERIA IN BUTTER.

126. Ripening of cream. The ripening changes that occur in cream are exceedingly complex, and our knowledge concerning the same is as yet far from satisfactory. The present belief is that these fermentations are largely the result of bacterial action, as germ growth is very abundant. Conn¹ found in fresh cream 4,060,000 bacteria, while the ripened cream contained 346,000,000.

In the ripening of cream at least three different factors are to be taken into consideration, the development of acid, flavor and aroma. Much confusion in the past has arisen from a failure to discriminate between these factors. While all three of these qualities are produced simultaneously in ordinary ripening, it does not necessarily follow that any single organism is able to develop all three qualities. If this ripening is allowed to go too far, undesirable rather than beneficial decomposition products are produced. These greatly impair the value of butter, so that in a sense the commercial value of this

¹ Conn, Storr's Agric. Expt. Stat., 7: 72, 1894.

product is dependent upon the character and the extent of the ripening changes. A careful study of large numbers of dairy bacteria as has been done by Conn, shows that these problems can be separated, if pure cultures are used.

127. Development of acid. In the ripening of cream acid is almost invariably developed. This development is essential, as it renders churning easier, and increases the yield of butter. The acid formed is largely lactic, and is produced by a decomposition of the milk-sugar. So characteristic of cream-ripening is this production of acid that the process is frequently spoken of as souring, although it must be kept in mind that the process involves much more than mere acid production. Normal butter made from ripened cream always has a characteristic flavor, which quality is distinct from the acid formation. This difference is seen in the results obtained by Tiemann¹ when cream is ripened by the addition of hydrochloric acid. When so treated, cream can be easily churned, but the product is lacking in the aromatic qualities found in normally ripened cream.

128. Flavor. The flavor of butter is that quality that is judged by the sense of taste. Good, bad, or indifferent flavors may be produced in butter as a result of bacterial action. In fact, the production of flavor in ripened cream butter is in large part due to the fermentative products of microbes, although it is to some extent modified by the inherent qualities of the milk that are induced by the character of the feed which is consumed by the animal.

129. Aroma. The aroma of butter is often confounded with that of flavor, but this quality is dependent upon

¹ Tiemann, *Milch Ztg.*, **23**: 701.

the presence of volatile decomposition products that appeal to the sense of smell rather than taste. As a rule butter is judged by this characteristic, a good flavor accompanying a desirable aroma, but while these two qualities are frequently present in the same butter made from naturally ripened cream, it does not by any means follow that one is directly dependent upon the other.

130. Origin of flavor and aroma. The source from which these delicate and evanescent qualities are derived is not yet definitely known. Two opposing views have been advanced. Storch¹ holds that the flavors are produced from the decomposition of milk-sugar and the absorption of the volatile flavors by the butter fat. Conn² believes that the nitrogenous elements in the cream function as food materials from which are formed various decomposition products, among which is the desired aromatic substance. The change is unquestionably a complex one, and cannot be explained as a single fermentation.

There is no longer much doubt but that both acid-forming and casein-digesting species are concerned in the production of proper flavors as well as aromas. The researches of Conn³, who has studied this question most exhaustively, indicate that both of these types of decomposition participate in the production of flavor and aroma. He has shown that both flavor and aroma production are independent of acid, that many good flavor-producing forms belong to that class which renders milk alkaline, or does not change the reaction at all. Some of these species liquefied gelatin and would therefore belong to the casein-dissolving class. Those species that produced bad flavors are also included in both fermentative types.

¹ Storch, Nogle. Unders. over Floed. Syrning, 1890.

² Conn, 6th Storrs Expt. Stat., p. 66, 1893.

³ Conn, 9th Storrs Expt. Stat., p. 17, 1896.

Conn has found a number of the organisms that are favorable flavor-producers; in fact they were much more numerous than desirable aroma-yielding species. None of the favorable aroma forms were lactic acid species¹.

131. Effect of bacteria on ripening. The majority of bacteria in ripening cream do not seem to exert any particular influence in butter. A considerable number are positively beneficial, inasmuch as they produce a good flavor and aroma, so far as these qualities are pronounced. A more limited number are concerned in the production of undesirable ripening changes². A knowledge of the conditions that control the development of these respective types of ripening is of greatest practical value to the butter-maker. If it were possible to have these conditions under exact control, then the butter industry would be reduced to the terms of an exact science.

132. Methods of cream-ripening. 1. *Natural ripening.* The simplest and oldest method is where the cream is allowed to ripen without any special control, the only artificial aid being a regulation in a crude way of the temperature. The whole process is a let-alone one. The ripened product varies much in degree of ripeness; also in the kind of fermentation, depending upon the various species of bacteria that have happened to gain access to the milk.

The results obtained by these cruder methods are usually fair, but simply because the majority of the organisms normally present in ordinarily clean milk are such as are unable to produce marked flavors of an undesirable character.

2. *Natural starters.* In the above method the rate of ripening is often irregular, and to overcome this, starters

¹ Weigmann has reached these same results (Milch Ztg., p. 793, 1891).

² Eckles, Cent. f. Bakt., II. Abt., 4: 730, 1898.

have been introduced as a means of hastening and rendering the ripening process more uniform. In the use of these, bacterial growths are added to assist in the ripening of cream, although generally no knowledge of the bacterial processes involved has until recently been taken into consideration. These methods are a great improvement over the previous let-alone policy of cream-ripening as one is often able to exclude in this way, undesirable fermentative changes. For starters of this sort, several different materials are used, as sour-milk, buttermilk, or whey, all of which are liquids rich in bacteria.

3. *Artificial starters (bacterial cultures)*. A much more scientific method of ripening cream has been introduced within the last few years and is now being extensively used in Scandinavian, Danish, and German dairies. This method dates from the labors of Storch,¹ who in 1890 proposed and introduced the use of pure cultures that have been selected on account of their ability to produce a desirable ripening change in cream. His investigations opened up a new field of research which has since been diligently prosecuted in both Europe and America.

133. Principles of pure culture cream-ripening. This method rests on the principle that a selected culture that has been chosen on account of its favorable ripening qualities will be able to produce a similar fermentation in cream, if the proper conditions are present. To get the best results, it is evident that the selected culture will have favorable conditions for its development, if the pre-existing bacteria in the cream are first destroyed. In this way competition is reduced to a minimum. This preparation of the cream is accomplished by pasteurizing

¹ Storch, *Milch Zeit.*, 1890, p. 304.

the same. This destroys for the most part the vegetative bacteria, leaving the latent spore forms in the cream. The addition then of a properly propagated starter gives the selected organism such an impetus that it should be able to overcome any other organism in the cream. Where pure cultures are employed extensively, as in Denmark and Germany, this method of cream-ripening is followed.

The attempt has been made to use these culture starters in raw sweet cream, but it can scarcely be expected that the most beneficial results will be attained in this way. This method has been justified on the basis of the following experiments. Where cream is pasteurized and no starter is added, the spore-bearing forms frequently produce undesirable flavors. These can almost always be controlled if a culture starter is added, the obnoxious form being repressed by the presence of the added starter. This condition is interpreted as indicating that the addition of a starter to cream which already contains developing bacteria will prevent those originally present in the cream from growing.¹ This repressive action of one species on another is a well-known bacteriological fact, but it must be remembered that such an explanation is only applicable in those cases where the culture organism exercises a direct prejudicial influence on the existing flora of the cream.

If the culture organism is added to raw milk or cream which already contains a flora that is eminently adapted for development in this medium, it is quite doubtful whether the culture organism would gain the supremacy in the ripening cream. The above method of adding a culture to raw cream renders cream-ripening details less burdensome, but at the same time Danish experience,

¹ Conn, 9th Storrs Expt. Stat., p. 25, 1896.

which is entitled to most credence on this question¹, is opposed to this method.

134. Pasteurizing milk or cream for butter. To pasteurize for butter-making, it is not necessary to carry out the process in the same stringent manner as when it is done to preserve milk or to free it from possible disease-breeding bacteria. A temperature of 140° F. for ten minutes is fatal to most of the lactic acid group, but in pasteurizing for butter-making, it is customary to heat the milk to a higher temperature (155-175° F.). This is done for two purposes. First, the milk is generally heated in a continuous-flow machine; consequently, the maximum temperature is not maintained for more than part of the time required to go through the machine. Second, the wide-spread prevalence of tuberculosis, and foot and mouth disease in some of the dairy regions of Europe makes it necessary to heat the skim-milk high enough to destroy the seeds of these diseases.

In pasteurizing for butter-making, it is also possible to drive off many taints that have been absorbed directly from the cow or through exposure to foul odors². Some states of the Atlantic sea-board are so infested with wild garlic that the milk-supply is rendered practically worthless for dairy purposes. If this is treated by pasteurization, it is often possible to eliminate in part such taints, so that a fairly good product of butter can be made.

In pasteurizing for butter-making, two methods are in vogue in Denmark where the greatest activity exists in regard to this matter. There, either the whole milk is

¹ 99% of the Danish creameries have pasteurizers, and over 90% pasteurize the cream.

² McKay, Ia. Expt. Stat., Bull. 32, p. 477.

handled or the cream is pasteurized after separation. The necessity of heating the skim-milk to destroy the seeds of tubercle in the milk-supply makes it advisable to heat the whole milk, thus subserving two purposes by the same operation.

135. Pasteurizing apparatus for butter-making.

Most of the European pasteurizing machinery has been devised from the butter-maker's standpoint. They are, therefore, machines that have a large capacity, as this factor is generally regarded as of chief importance in pasteurizing for butter-making. To fulfill this condition, most of them are arranged with a continuous inflow and outflow. Such an arrangement militates considerably

against the efficiency of any machine as a bacteria-destroying agent. In a test made by the writer¹ of Reid's pasteurizer (fig.31), which is a modification of a Danish machine, it was found that the bacterial content of the pasteurized milk varied greatly, ranging from 50–99%, although on the whole the germicidal effect was very marked.

136. Attributes desirable in pure culture starters.

In adding a pure culture starter to cream, it is desirable, if possible, to use a ferment that possesses the following characteristics:

¹ Wis. Expt. Stat., Bull. 69, p. 11.

1. Vigorous growth in milk at ordinary ripening temperatures.
2. Ability to form acid so as to facilitate churning and increase the yield of butter.
3. Able to produce a clean flavor and desirable aroma.
4. Impart a good keeping quality to butter.
5. Not easily modified in its flavor-producing qualities by artificial cultivation.

These different conditions are difficult to attain, for the reason that some of them seem to be in part incompatible. Weigmann¹ found that a good aroma was generally an evanescent property, and therefore, opposed to good keeping quality. Conn has shown that the functions of acid-formation, flavor and aroma production are not necessarily related, and therefore, the chances of finding a single organism that possesses all the desirable attributes are not very good. It may well be asked if the conditions are so peculiar, how is it that a good product is ever made where no special attention is given this matter of culture starters? The reason for this is that the ripening of cream is not a single type of fermentation, but a complex process in which various organisms may participate.

In all probability no one germ possesses all of the desirable qualities, but the resultant of the action of several forms may produce a good product². This idea has led to the attempt of mixing selected organisms that have been chosen on account of certain favorable characteristics which they might possess. The difficulty of maintaining such a composite culture in its correct proportions when it is being propagated in the creamery is seemingly well nigh insuperable, as one organism is very apt to develop more or less rapidly than the other.

¹ Weigmann, Landw. Woch. f. Schl. Hol., No. 2, 1890.

² Weigmann, Cent. f. Bakt., II. Abt., 3: 497, 1897.

137. Reputed advantages of culture starters.

1. *Flavor and aroma.* These are factors of greatest importance, as the market value is determined largely by this quality. Ferments of this sort rarely produce any higher or more pronounced flavor or aroma than the best of the natural product, but they possess the reputation of producing a clean, mild flavor that is very desirable when the market becomes accustomed to it.

2. *Uniformity of product.* Culture starters produce a more uniform product, because the type of fermentation does not vary from day to day. Even the best of butter-makers sometimes fail to make a standard product, owing to their inability to fully control the character of the ripening process.

3. *Keeping quality of product.* Butter made from pasteurized cream to which a good, pure starter has been added will keep much better than the ordinary product, because the diversity of the flora is less and the milk is not likely to contain those organisms that produce an "off" condition.

4. *Elimination of butter defects.* One of the greatest advantages is its application to factories that are unable to make good butter on account of some hidden trouble due to bacteria.

138. Pure cultures vs. home-made starters. The question as to the relative merits of pure or domestic ferments as starters should be answered in the light of varying conditions. In the present development of the pure culture system, no culture on the market possesses such superior advantages over a first-class domestic starter especially as to flavor and aroma, that it can be unqualifiedly recommended as better. The main advantage of the culture system lies in the uniformity of the product. The elimination of the risk that always attends in a greater

or smaller degree the use of an ordinary starter, is of material worth. If great care, however, is taken in the selection of the domestic starter, and it is propagated under as equally favorable conditions for the retention of its original purity, as can easily be done with the commercial starter, then there is no reason why practically as good results cannot be obtained in the use of one as the other.

139. Imperfections in system. The commercial value of butter is so dependent upon the character of the flavor that the effect of a starter on this factor will always be of supreme importance. At the present time no pure culture starter is known that is capable of imparting a much higher flavor than is obtained in the regular way¹. Consequently, the maker is unable to secure a higher price for his butter than in the better creameries where the older method is used.

Another decided disadvantage that occurs where the cream is pasteurized in conjunction with the use of a pure culture, is the effect on the grain or body of the product. As judged by our present American standards, the grain of the butter is materially lowered where the cream has been heated.² This undoubtedly would not be so considered in the foreign, especially the English or Danish markets, but so long as our market standards exact the qualities now demanded on grain, this will to some extent handicap pasteurized butter.

140. Propagation of starter for cream-ripening. The preparation and propagation of a starter for cream-ripening is a process involving considerable bacteriological knowledge, whether the starter is of domestic origin or prepared from a pure-culture ferment. In any event,

¹ This statement is confined to American conditions.

² Farrington and Russell, Wis. Expt. Stat., Bull. 69, p. 28.

it is necessary that the starter should be handled in a way so as to prevent the introduction of foreign bacteria as far as this is possible. The following points should be kept in mind in manipulating the starter:

FIG. 32. Apparatus for sterilizing and propagating the starter

1. If a pure-culture ferment is used, see that it is fresh and that the seal has not been disturbed.
2. If a home-made starter is employed, use the great-

est possible care in selecting the milk that is to be used as a basis for the starter.

3. For the propagation and perpetuation of the starter from day to day, it is necessary that the same should be grown in milk that is as germ-free as it is possible to secure it. For this purpose sterilize some fresh skim-milk in a covered can that has previously been well steamed. This can be done easily by setting cans containing skim-milk in a vat filled with water (fig. 32 A) and heating the same to approximately the boiling point. The temperature should be maintained for an hour or more. This destroys all but the most resistant spore-bearing organisms¹.

4. After the heated milk is cooled down to about 70° F., it can be inoculated with desired culture. Sometimes it is desirable to "build up" the starter by propagating it first in a smaller volume of milk, and then after this has developed, adding it to a larger amount.

This method is of particular value where a large amount of starter is needed for the cream-ripening.

5. After the milk has been inoculated, it should be kept at a temperature that is suitable for the rapid development of the contained bacteria, 60–70° F., which temperature should be kept as constant as possible.

6. This can best be done by filling water-vat (A) with water and heating the same to desired temperature, covering the vat with a wooden cover or heavy cloth during the night to maintain proper temperature.

7. The starter should not be thoroughly curdled and solid when it is needed for use, but should be well soured

¹ A number of tests made by the writer at the Wis. Dairy School Creamery, showed that the bacteria in skim-milk were practically destroyed, only 7–30 bacteria per cc. remaining after this sterilizing process.

and partially curdled. This point is of importance for the following reasons:

a. It is difficult to thoroughly break up curd particles if the starter is completely curdled. If these curd masses are added to ripening cream, white specks may appear in the butter.

b. The vigor of the starter is undoubtedly stronger when the milk is on the point of curdling than it is after the curd has been formed some time. The continued formation of lactic acid kills many of the bacteria and thus weakens the fermentative action.

8. The starter should be propagated from day to day by adding a small quantity to a new lot of milk. For this purpose two propagating cans should be provided (fig. 32 B) so that one starter may be in use while the other is being prepared.

9. After the starter has been used for a few days, the same should be emptied, and the can cleaned and sterilized by steam before being used again.

141. How long should a starter be propagated?

No hard and fast rule can be given for this, for it depends largely upon how carefully the starter is handled during its propagation. If the starter is grown in sterilized milk kept in steamed vessels and is handled with sterile dippers, it is possible to maintain it in a state of relative purity for a considerable period of time; if, however, no especial care is given it, it will soon become infected from the air and the retention of its purity will depend more upon the ability of the contained organism to choke out foreign growths than upon any other factor. While it is possible by bacteriological methods to determine with accuracy the actual condition of a starter as to its germ content, still such methods are inapplicable in creamery practice. Here the maker must rely largely

upon the general appearance of the starter as determined by taste and smell. Where it is possible to propagate the starter as has been described, it is hardly worth running any risk in using an old starter when a fresh one can be so easily kept in condition.

142. Bacteria in butter. As ripened cream is necessarily rich in bacteria, it follows that butter will also contain germ life in varying amounts, but as butter-fat is not well adapted for bacterial food, the number of germs in butter is usually less than in ripened cream.

Sweet cream butter is naturally poorer in germ life than that made from ripened cream. Grotenfelt¹ reports in sweet cream butter, the so-called "Paris butter" only 120-300 bacteria per cc., while in butter from sour cream 2,000-55,000 germs per cc. were found. Pammel² found from 125,000-730,000 per gram, while Lafar³ found in butter sold in Munich from 10-20,000,000 organisms per gram.

The germ content of butter on the outside of a package is much greater than it is in the middle of a mass; this doubtless being due to the freer access of air favoring the growth of aerobic forms.

143. Changes in germ content. The bacteria that are incorporated with the butter as it first "comes" undergo a slight increase for the first few days. The duration of this period of increase is dependent largely upon the condition of the butter. If the buttermilk is well worked out of the butter, the increase is slight and lasts for a few days only, while the presence of so nutritious a medium as buttermilk affords conditions much more favorable for the continued growth of the organisms.

¹ Grotenfelt-Woll, Prin. Mod. Dairy Practice, p. 244.

² Pammel, Bull. 21, Iowa Expt. Stat., p. 801.

³ Lafar, Arch. f. Hyg., 13: 1, 1891.

While there may be many varieties in butter when it is fresh, they are very soon reduced in kind as well as number. The lactic acid group of organisms disappear quite rapidly; the spore-bearing species remaining for a somewhat longer time. Butter examined after it is several months old is often found to be almost free from germs. This fact is important, for in considering the after changes in "storage" butter, the relation of these changes to the germ life would naturally be considered.

In the manufacture of butter there is much that is dependent upon the mechanical processes of churning, washing, salting, and working the product. These processes do not involve any bacteriological principles other than those that are incident to cleanliness. The cream, if ripened properly, will contain such enormous numbers of favorable forms that the access of the few organisms that are derived from the churn, the air, or the water in washing will have little effect, unless the conditions are abnormal.

144. Rancid change in butter. Fresh butter has a peculiar aroma that is very desirable and one that enhances the market price, if it can be retained; but this delicate flavor is more or less evanescent, soon disappearing, even in the best makes. While a good butter loses with age some of the peculiar aroma that it possesses when first made, yet a gilt-edged product should retain its good keeping qualities for some length of time. All butters, however, sooner or later undergo a change that render them worthless for table use. This change is usually a rancidity that is observed in all stale products of this class. The cause of this rancid condition in butter has been attributed to the action of living organisms, particularly those that form butyric acid, to the influence of light, of air, etc.

In rancid butter, butyric and allied acids are always found, and it was supposed for a long time that the change was a butyric fermentation inaugurated by some of the butyric organisms that are found so commonly in milk and cream.

Ritsert¹ found that sterile butter became rancid in three days if exposed to the action of the light, while unsterilized, normal butter exposed under similar conditions, did not change in five months, if the air was excluded from it. Duclaux² has proven that the rancid change is largely a chemical action that takes place in butter-fat where it is exposed to light and oxygen; that it is not necessarily inaugurated by the vital functions of any special kind of bacteria. While the change goes on in most cases in a purely chemical way, there are, however, certain organisms that are able to hasten this process if they are present in the butter³. For this reason a soft butter containing considerable quantities of buttermilk, and therefore rich in nitrogenous material, undergoes a rapid change and quickly becomes rancid.

145. Defects due to manufacturing methods.

There are other defects in butter that are also attributable to other than bacterial causes. These are for the most part due to errors in manufacture. Thus, mottled or wavy butter is generally caused by the uneven distribution of the salt. White specks in butter are often produced where the cream is allowed to ripen for too long a time, or where curdled milk is used as a starter. In such cases, the curd particles remain undissolved, and are held in the butter, where they appear as white specks.

¹ Ritsert, Inaug. Diss. Berne, 1890.

² Duclaux, *Le Lait*, p. 34.

³ Von Klecki, *Cent. f. Bakt.*, 15: 354.

B. BACTERIAL DEFECTS IN BUTTER.

146. Lack of flavor. Often this may be due to improper handling of the cream in not allowing it to ripen far enough, but sometimes it is impossible to produce a high flavor. The lack of flavor in this case, is due to the absence of the proper flavor-producing organisms. This condition can usually be overcome by the addition of a proper starter. The relation between flavor and desirable bacteria is very intimate; and troubles of this kind usually arise, because the proper forms commonly found in the cream have been supplanted by other species that do not possess the ability of forming these aromatic substances so necessary in sour cream butter.

147. Putrid butter. This specific butter trouble has been observed in Denmark, where it has been thoroughly studied by Jensen¹. Butter affected by it rapidly acquires a peculiar putrid odor that ruins it for table use. Sometimes, this flavor may be developed in the cream previous to churning. Jensen found the trouble to be due to several different putrefactive bacteria. One form which he called *Bacillus fætidus lactis*, a close ally of the common feces bacillus, produced this rotten odor and taste in milk in a very short time. Fortunately, this organism was easily killed by a comparatively low heat, so that pasteurization of the cream and use of a culture starter quickly eliminated the trouble, where it was tried.

148. Turnip-flavored butter. Butter sometimes acquires a peculiar flavor recalling the odor of turnips, rutabagas, and other root crops. Often this trouble is due to feeding, there being in several of these crops, aromatic substances that pass directly into the milk, but in some instances the trouble arises from bacteria that

¹ Jensen, Cent. f. Bakt., 11: 409, 1891.

are able to produce decomposition products¹, the odor and taste of which strongly recalls these vegetables.

149. "Cowy" odor in butter. Frequently there is to be noted in milk a peculiar odor that resembles that of the cow stable and sometimes the odor of the animal itself. Usually this defect in milk has been ascribed to the absorption of impure gases by the milk as it cools, although the gases and odors naturally present in fresh milk have this peculiar property that is demonstrable by certain methods of aeration. Occasionally, it is transmitted to butter, and recently, Pammel² has isolated from butter, a bacillus that produced in milk the same peculiar odor so commonly present in stables.

150. Lardy and tallowy butter. The presence of this unpleasant taste in butter may be due to a variety of causes. In some instances, improper food seems to be the source of the trouble; then again, butter exposed to direct sunlight bleaches in color and develops a lardy flavor.³ In addition to these, cases have been found in which the defect has been traced to the action of bacteria. Storch⁴ has described a lactic acid form in a sample of tallowy butter that was able to produce this disagreeable odor.

151. Oily butter. Jensen has isolated one of the causes of the dreaded oily butter that is reported quite frequently in Denmark. The specific organism that he found belongs to the sour-milk bacteria. In twenty-four hours it curdles milk, the curd being solid like that of ordinary sour milk. There is produced, however, in addition to this, an unpleasant odor and taste resembling

¹ Jensen, *Molk. Ztg.*, **6**: Nos. 5 and 6, 1892.

² Pammel, *Bull.* 21, Iowa Expt. Stat., p. 803.

³ Fischer, *Hyg. Rund.*, **5**: 573.

⁴ Storch, 18th Rept. Danish Agric. Expt. Stat., 1890.

that of machine oil, a peculiarity that is transmitted directly to butter made from affected cream.

152. Bitter butter. Now and then butter develops a bitter taste that may be due to a variety of different bacterial forms. In most cases, the bitter flavor in the butter is derived primarily from the bacteria present in the cream or milk. Several of the fermentations of this character in milk are also to be found in butter. In addition to these defects produced by a biological cause, bitter flavors in butter are sometimes produced by the milk being impregnated with volatile, bitter substances derived from weeds.

153. Moldy butter. This serious defect in butter is caused by the development of the common mold fungus (*Penicillium glaucum*) on the inside of butter packages. Where tubs are made from green, sappy wood, the mold-spores that are practically everywhere, find favorable conditions for growth, especially, where moisture exists. The result is that as the wood molds, the outer layers of the butter are stained and at the same time acquire a moldy taste. This defect can be easily obviated by steaming the tubs thoroughly before packing. Butter tubs should always be kept in a cool, *dry* place under conditions unfavorable for the growth of the mold-spores.

CHAPTER X.

BACTERIA IN THE CHEESE INDUSTRY.

154. Relation of bacteria to cheese. The processes of cheese-making are much more affected by the operation of bacterial causes than almost any other dairy industry. Cheese contains so large a proportion of nitrogenous matter that, so far as food is concerned, bacteria find better conditions for development than in butter. The ripening or curing of cheese is a fermentative process in which bacteria are intimately concerned. Our knowledge of the actual changes induced by these organisms is, as yet, quite meager, but enough has already been determined to indicate that the whole industry is based largely on phenomena of ferment action, and that the application of bacteriological principles and ideas to this phase of dairying is sure to yield more than ordinary results, in explaining in a rational way, the reasons underlying many of the operations of this industry.

155. Principles of cheese-making. The general principle of cheese-making is to precipitate the casein, and then press this into a more or less solid mass, allowing it to stand for varying periods of time, and under various conditions, so that a wide range is presented, in which various fermentative actions may operate to produce the widely different kinds of cheese. From the same kind of milk, a great variety of different cheeses can be prepared, depending upon the treatment of the milk during the manufacture, and the way in which the cheese is handled after it is made.

In precipitating the casein, the fat is caught in the curd particles, and is thus incorporated in the cheese. This curdling of the casein is brought about in two ways.

1. Addition of rennet, a chemical ferment extracted from calves' stomachs¹, which has the specific property of rendering the casein of milk insoluble, *e. g.*, cheddar and Swiss varieties of cheese.

2. Development of acid until the casein is precipitated, *e. g.*, sour milk, cottage cheese.

By far the larger amount of cheese is made in the first way. The various kinds manufactured may be divided into two general classes, soft and hard cheese, depending upon the character of the finished product, which is produced by the kind of treatment that is given the milk in the manufacture of the cheese and its subsequent curing. By far the larger amount of cheese made in this country belongs to hard, firm type of cheese.

156. Summary of manufacturing details. In making cheese, it is necessary to concentrate the casein and adherent fat into a compact mass, containing only a limited amount of water. The addition of the rennet precipitates the casein and causes it to shrink in volume. In order to facilitate this process, the coagulated casein is cut into small pieces, thereby giving a better opportunity for the continued shrinking action, and so expel the milk-serum or whey. This change is greatly affected by the presence of acid and heat. In order to make the process uniform from day to day, the milk is held or "ripened" until a certain degree of acidity is reached from the development of the bacteria that are invariably present in the milk. The curds are then heated to a varying temperature, the action of the heat being to further expel

¹ In Italy this ferment is sometimes secured from artichokes and is used in cheese manufacture.

the moisture which is separated from the curd in the whey. If a rather dry, slow-curing cheese is wanted, the curds are cooked in the whey at a higher temperature, and for a longer time, than if a more rapidly curing product is desired.

The bacteria originally present in the milk are able to grow with very great rapidity under these conditions, and the result is, that if the normal milk bacteria (lactic acid forms) are present, there will be a steady development of the acid produced by the decomposition of the milk-sugar. This development of the acid in the curd has a marked effect upon the character of the green cheese.

157. Influence of bacteria in making cheese. Inasmuch as the proper development of acid in the milk before and after the casein is precipitated is determined largely by the kind of bacteria that may happen to be present, it necessarily follows that the success of the process is governed somewhat by the original condition of the milk as to its bacterial life. In pure, clean milk, the native species of bacteria seem to be present in sufficient numbers, so that the proper development of the acid in the vat is rendered possible, but it not infrequently happens that the milk may be "off" in flavor, *i. e.*, the normal milk organisms may be supplanted by other species of a less desirable character that have gained access to the milk through improper handling. There is considerable competition among different species of bacteria when growing in same fluid, and sometimes they antagonize each other so strongly by reason of the by-products which are formed that where one kind is in abundance, other species are unable to grow. So, if certain kinds are introduced, say from dirt or filth gaining access to the milk, these species may repress the growth of the normal milk

forms to such an extent that the necessary acid is not produced.

If bacteria are present in the milk that are able to peptonize or digest the same, a not inconsiderable amount of the cheese-producing solids may be dissolved and be lost in the whey. Organisms of this class are usually derived from dust and animal filth. By the use of the fermentation or curd test (182), it is possible to detect a condition of this sort in the milk.

158. Starters in cheese-making. Where the milk reaches the factory in an extra sweet condition, it is sometimes advisable to add a starter to hasten the ripening of the milk. Usually sour milk is employed, but with any starter, great care should be taken to add nothing that will introduce into the milk any undesirable taint.

Where "gassy" milks prevail, the proper development of the acid is also interfered with, and in such cases, the addition of a lactic ferment is of great aid in repressing these abnormal fermentations.

While the ordinary starter used is one of domestic origin, as skim or sour milk, within recent years the attempt has been made to introduce pure cultures¹ of various lactic acid-producing forms. These cannot be used in pasteurized milk as is the case in butter-making, but even where added to the raw milk, they exert a material effect. By their use the necessary impetus is given to the formation of acid, and the development of this retards the growth of the gas-forming organisms. The period of manufacture is also materially shortened. Their use should, however, be carefully governed, as sour cheese is apt to develop if employed too freely.

Campbell² has also used the lactic starter to overcome

¹ Russell, 13th Rept. Wis. Expt. Stat., p. 108, 1896.

² Campbell, North Brit. Agric., May 12, 1897.

a serious cheese defect that occurs frequently in Scotland, viz., the discoloration of cheese.

The use of stringy or slimy whey has been advocated in Holland for some years as a means of overcoming the tendency toward gas formation in Edam cheese which is made from practically sweet milk. This fermentation, the essential feature of which is produced by a culture of *Streptococcus Hollandicus*¹, develops acid in a marked degree, thereby inhibiting the production of gas.

159. Bacteria in rennet. The addition of rennet to milk has no appreciable effect on the development of the bacteria in the same. The idea has been advanced that the bacteria in the rennet extract exert a considerable influence on the cheese; this view is, however, hardly tenable, in spite of the fact that the rennet usually contains large numbers of organisms, for the relative amount of rennet that is added is so small that this does not perceptibly increase the germ content of the milk.

It is possible that undesirable bacteria may be introduced into the milk by means of the rennet where the same is contaminated with noxious forms; but these cases are relatively rare. This might apply particularly to those cases in which the natural rennets are used. These are preserved in salt, alcohol, or boric acid, but they are never free from bacteria. Adametz² found ten different species and from 640,000–900,000 bacteria per cc. in rennets.

160. Green cheese. If the proper amount of acid has been allowed to develop, a cheese fresh from the press will present a close uniform texture, that in the "green" condition is tough and elastic. This rapidly changes as the cheese ripens, and with this change there is a marked increase in the amount of bacterial life.

¹ Weigmann, Milch Ztg., No. 50, 1889.

² Adametz, Landw. Jahr., 18: 256.

Where but little acid is developed as in a sweet curd cheese the texture is open and porous. The numerous cavities that are to be noted arise from the fact that the curd particles have not been closely matted together. This matting or cementing of the curd masses is due to their partial solution under the liquefying influence of the acid produced by the bacteria. Where these "mechanical holes" are present, gas is almost invariably developed, as the gas-producing bacteria that are quite susceptible to acid are not inhibited under these conditions. This gas finds its way into the ready-made mechanical holes, greatly distending them as in fig. 33.

FIG. 33. Showing "mechanical holes" in a sweet curd cheese and the distension of the same by gas.

L shows appearance of a sweet curd cheese immediately after it is taken from the press. The "mechanical holes" are produced by the failure of the curd particles to cement, owing to the absence of acid.

P shows appearance of a duplicate cheese four days after removal from press. The gas produced by bacterial germs fills up and distends these "mechanical holes," thus causing a marked distortion in shape of cheese (huffing).

161. Physical changes in ripening. When a green cheese is taken from the press, the curd is tough, firm, but elastic. It has no value as a food product for imme-

diate use, because it lacks a desirable flavor and is not readily digestible. It is nothing but precipitated casein and fat. In the course of a short time, a deep-seated change occurs. Physically this change is demonstrated in the modification that the curd undergoes. Gradually it breaks down and becomes plastic, the elastic, tough curd being changed into a softened mass. This change in texture of the cheese is also accompanied by a marked change in flavor. The green cheese has no distinctively cheese flavor, but in course of time, with the gradual change of texture, the peculiar flavor incident to ripe cheese is developed.

The characteristic texture and flavor are susceptible of considerable modification that is induced not only by variation in methods of manufacture, but by the conditions under which the cheese are cured. The amount of moisture incorporated with the curd materially affects the physical appearance of the cheese, and the rate of change in the same. The temperature at which the same is ripened, likewise the moisture content of the surrounding air, also exert a marked influence on the ripening product. To some extent the action of these forces is purely physical, as in the gradual loss by drying, but they also affect the course of the biological changes to a certain extent.

162. Chemical changes in ripening. The marked physical change that occurs during ripening also records a profound chemical action in the composition of cheese. Coincident with the softening of the curd, comes a change in the condition of the casein. The hitherto insoluble casein is gradually transformed into soluble substances (*caseone* of Duclaux, or *caseogluten* of Weigmann). This chemical phenomenon is a breaking down process that is analogous to the peptonization of proteids, although in

addition to peptones, numerous other intermediate products are formed ranging from albumens to ammonia. Amido acids (leucin and tyrosin) are present in an old cheese. The chemical reaction of cheese is generally slightly acid to phenolphthalein, although in an old cheese where putrefactive changes are going on, the reaction may be alkaline.

The changes that occur in a ripening cheese are for the most part confined to the proteids. The fat remains practically unchanged. In the green cheese considerable milk-sugar is present, but as a result of the fermentation that occurs, this is rapidly converted into acid products.

163. Bacterial flora of cheese. It might naturally be expected that the green cheese, fresh from the press would contain practically the same kind of bacteria that are in the milk, but a study of cheese shows a peculiar change in the character of the flora. In the first place, fresh cottage cheese made by the coagulation of the casein through the action of acid, has a more diversified flora than cheese made with rennet, for the reason, as given by Lafar,¹ that the fermentative process is farther advanced.

When different varieties of cheese are made from milk in the same locality, the germ content of even the ripened product has a marked similarity, as is illustrated by Adametz's work² on Emmenthaler or Swiss hard cheese, and Schweitzer Hauskäse, a soft variety. Of the nine species of bacilli and cocci found in mature Emmenthaler, eight of them were also present in ripened Hauskäse.

Different investigators have studied the bacterial flora of various kinds of cheese, but as yet no systematic comparative work has been done in an extended manner.

¹ Lafar, *Technical Mycology*, p. 216.

² Adametz, *Landw. Jahr.*, **18**: 228.

Freudenreich,¹ and the writer,² independently, have determined the character and numbers of bacteria in Emmenthaler and American cheddar cheese. Their results practically agree as to the course of the bacterial changes in these two kinds of cheese.

First, there is a marked decrease in numbers lasting for a day or so. This is then followed by an enormous numerical increase that is entirely produced by the marked development of the lactic acid forms. Synchronous with this increase, the peptonizing and gas-producing bacteria gradually disappear, so that in a short period of time, the bacterial flora seems to be made up very largely of this lactic type of organisms. This rapid development is followed by a gradual decline that continues in an old cheese for two years or more. As the cheese dries out, the conditions become unfavorable, and as the lactic acid species are not able to form spores they soon succumb to their unfavorable environment. The following data illustrate this:

Bacteria per gram in cheddar cheese at different stages of ripening.

Date of analysis.	Total number.	Lactic acid bacteria.
Curd	26,532,000	22,560,000
1st day	21,060,000	20,176,000
5th day	43,716,000	39,680,000
10th day	98,080,000	97,280,000
19th day	48,140,000	48,000,000
36th day	11,171,000	11,000,000

This condition has been found to exist not only in American cheddar cheese but in Canada³ and England⁴.

¹ Freudenreich, Landw. Jahr. d. Schweiz, 4: 17; 5: 16.

² Russell, 13th Wis. Expt. Stat., p. 95, 1896.

³ F. C. Harrison (unpublished data).

⁴ Lloyd, Bath, and West of Eng. Soc. Rept., 2: 180, 1892.

164. Relation of curing temperature to bacterial flora. It has long been known that a variation in temperature, particularly if the temperatures exceeded 70° F., had a detrimental effect on the quality of the cheese ripened under these conditions. The cause of this difference is not thoroughly apprehended at the present time, although the general belief is that it is due to the growth of various kinds of bacteria that are able to develop under these various temperature conditions.

A detailed study made by the writer¹ of the characteristics of the bacterial flora of cheese ripened at different constant temperatures shows that the bacterial life is considerably influenced by temperature conditions. The rise and fall of bacteria in cheese ripened at normal temperatures has just been stated. In cheese ripened at refrigerator temperatures (50–56° F.), the bacterial flora retains for a considerable period the same general aspect as in the milk. In this respect it differs somewhat from that of normal cheese, in which a differentiation soon takes place, the lactic acid bacteria gaining the ascendancy. In cheese cured at a high temperature (80–86° F.), the number of bacteria per gram is greatly diminished, and the organisms fail to persist for as long a period of time as under normal conditions. Some difference in character of organisms is found in cheese kept at low and high temperatures, but this is not always constant nor as well marked as has been supposed.

165. Theories of cheese curing. The curing of cheese is far from being a simple problem, yet, within the last few years definite knowledge of the processes concerned has been greatly increased.

In the earlier theories of cheese-ripening it was thought to be purely a chemical change, but with the growth of

¹ Russell, 14th Wis. Expt Stat., p. 203, 1897.

bacteriological science, evidence was forthcoming that indicated that it was due to the activity of organisms. Schaffer¹ showed that if milk was boiled and made into cheese, it failed to ripen. Adametz² added to greek cheese various disinfectants as creolin, thymol, etc. He found that this practically stopped the curing process. From these experiments, the conclusion was drawn that bacteria must be the cause of the ripening. As to the nature of the organisms that were supposed to be concerned in this process, the evidence is not convincing.

166. Digestive or peptonizing theory. From the chemical products determined in a matured cheese, there can no longer be any question, but that the general character of the cheese-ripening process is a peptonization or digestion of the casein. Duclaux³ in 1887 propounded the theory that this change was due to that type of bacteria that are able to liquefy gelatin, peptonize milk, and cause a hydrolytic change in proteids. To this widely spread group that he found in cheese, he gave the name *tyrothrix* (cheese hairs). According to him, these organisms do not function directly as ripening agents, but they secrete an enzyme or unorganized ferment to which he applies the name *casease*. This ferment acts upon the casein of milk, converting it into a soluble product known as *caseone*. These organisms grow in liquid milk with great rapidity and if they function as casein transformers, one would naturally expect to find them at least frequently, if not predominating in the ripening cheese, but such is not the case. In typical cheddar cheese, however, they rapidly disappear, although in the moister, softer varieties, they persist for consider-

¹ Schaffer, *Milch Ztg.*, p. 146, 1889.

² Adametz, *Landw. Jahr.*, **18**: 261.

³ Duclaux, *Le. Lait*, p. 213.

able time. Even where added in large numbers to the curd, they soon perish.

While the by-products of proteid decomposition induced by this class of bacteria are very similar to those found in a well-ripened cheese, still the above facts have seemed an insuperable barrier to the general acceptance of this explanation.

167. Lactic acid theory. It has already been shown that the lactic acid species seem to find in the green cheese, the optimum conditions of development, that they increase enormously in the cheese for a short period, and then finally decline in numbers. This marked development coincident with the breaking down of the casein, has led to the view that has been so ably expounded by Freudenreich¹ that this type of bacterial action is concerned in the ripening of cheese. This group of bacteria are unable to liquefy gelatin, or digest milk, or, in fact, to exert under ordinary conditions any proteolytic or peptonizing properties. This has been the stumbling-block to the acceptance of this hypothesis as an explanation of cheese-ripening. Freudenreich has recently carried on experiments that he believes solves the problem. By growing cultures of these organisms in milk to which sterile, freshly precipitated chalk had been added, he was able to prolong the development of bacteria for a considerable period of time, and as a result of this, he finds that an appreciable part of the casein is digested. Weigmann² inclines to the view that the lactic acid bacteria are not the true cause of the peptonizing process, but that their development guides or directs the character of the ripening by giving favorable conditions for the development of other species.

¹ Freudenreich, Landw. Jahr. d. Schweiz, p. 85, 1897.

² Weigmann, Cent. f. Bakt., II. Abt., 4: 593, 1898.

168. Enzyme theory. In 1897, Babcock and the writer¹ were able to demonstrate that milk contains certain inherent unorganized ferments or enzymes that have the power of digesting the casein of milk, when kept under the influence of chemicals that would suppress bacterial action, but which would not materially interfere with enzyme activity. This ferment they find present in the milk of different species of mammalia (burro, horse, sheep, goat, pig, buffalo, and human), and in cows' milk in such quantities that they can isolate it from centrifuge slime. The effect of this chemical ferment which they call *galactase*, is shown in the breaking down of the nitrogenous matter in boiled milk, forming albumoses, peptones, amido acids, and ammonia, products that agree in every way with those present in a normally ripening cheese. This ferment is allied to trypsin, the pancreatic enzyme, and Jensen² has recently shown that pancreatic extracts when added to cheese accelerate the digestive changes.

169. Present status of theory of cheese-ripening. Comparative experiments on milk and cheese seem to show beyond all question that the breaking down of casein into peptone and related products is due to the action of this inherent ferment of the milk which is called galactase, and not to the action of bacteria as heretofore believed. Previous investigators have failed to recognize the presence of this ferment in milk because they have sterilized the same by heat, and under these conditions galactase is destroyed. A brief exposure of milk to 176° F. is sufficient to accomplish this result, and even exposures to considerably lower temperature weakens its activity considerably, especially if the reaction of medium

¹ Babcock and Russell, 14th Wis. Expt. Stat., p. 161, 1897.

² Jensen, Cent. f. Bakt., II. Abt., 3: 752.

is acid. This undoubtedly explains the contradictory results obtained in the ripening of cheese made from pasteurized milk, such cheese occasionally breaking down but in an abnormal manner.

The previous results as to the failure of cheese to ripen when treated with disinfectants,—experiments which were supposed to be the foundation of the bacteriological theory of cheese-ripening—are now explainable on a new basis. The casein is not peptonized because these strong disinfectants destroy the activity of the enzyme as well as the bacteria. Where chemicals such as benzol, chloroform, ether, and toluol are used, bacterial growth is inhibited, but enzyme action is still possible. In the presence of these, green cheese undergoes the normal peptonization of the casein.

170. Flavor of cheese. While the action of this ferment is exerted on the decomposition of the casein, it is not yet known what its relation is to the production of proper flavors in cheese. Acting as it does on the nitrogenous compounds of the milk, it has a marked effect on the texture of the ripened cheese.

The manner in which the peculiar flavors that characterize the various kinds of cheese are produced is yet an unsolved problem. The view that is most generally accepted is that this most important phase of cheese curing is dependent upon bacterial activity, but the organisms that are concerned in this process have not as yet been satisfactorily determined. In a number of cases, different species of bacteria have been separated from milk and cheese, that have the power of producing aromatic compounds that resemble in some cases, the peculiar flavors and odors that characterize some of foreign kinds of cheese; but an introduction of these into curd has not

resulted in the production of the peculiar variety, even though the methods of manufacture and curing were closely followed. The similarity in germ content in different varieties of cheese made in the same locality has perhaps a bearing on this question of flavor as related to bacteria. Of the nine different species of bacteria found in Emmenthaler cheese by Adametz, eight of them were also present in ripened Hauskäse. If specific flavors are solely the result of specific bacterial action, it might naturally be expected that the character of the flora would differ.

The present state of our knowledge does not permit any definite statement being made with reference to the cause of flavor-production in the hard type of cheese, such as cheddar and Emmenthaler.

171. Ripening of moldy cheese. In a number of foreign cheese, the peculiar flavor obtained is in part due to the action of various fungi which grow in the cheese, and there produce certain by-products that flavor the cheese. Among the most important of these are the Roquefort cheese of France, Stilton of England, Gorgonzola of Italy.

Roquefort cheese is made from goat's or cow's milk, and in order to introduce the desired mold, which is nothing more than *Penicillium glaucum*, the ordinary bread mold, carefully prepared moldy bread crumbs are added to the curd. The cheese are ripened in limestone caverns where the temperature and moisture content are quite constant. When partially cured, they are placed in a machine that punches them full of small holes. These slender canals allow the mold organism to penetrate the whole mass more thoroughly; the moldy straw matting upon which the ripening cheese are placed helping to furnish an abundant seeding of the desired germ.

When new factories are constructed it is of advantage to introduce this necessary germ in quantities, and the practice is sometimes followed of rubbing the walls and cellars of the new location with material taken from the old established factory. In this custom, developed in purely an empirical manner, is to be seen a striking illustration of a bacteriological process crudely carried out.

In the Stilton cheese, one of the highly prized moldy cheese of England, the desired mold fungus is introduced into the green cheese by exchanging plugs taken with a cheese trier from a ripe Stilton.

In all probability, the breaking down of the casein is attributable in part, at least, to the action of the inherent ferments of the milk, although with some of the softer cheese, this may be aided by bacterial action.

172. Ripening of soft cheese. The type of ripening which takes place in these softer cheese is materially different from that which occurs in the first class. In many cases, the peptonizing action does not go on uniformly throughout the cheese but is hastened on the exterior by the development of organisms that exert a solvent effect on the casein. For this reason, soft cheeses are usually made up in small sizes, so that this action may be facilitated. The bacteria that take part in this process are those that are able to form unorganized ferments (similar in their action to trypsin, galactase, etc.), and these soluble ferments gradually diffuse from the outside through the cheese.

Most of these peptonizing bacteria are hindered in their growth by the presence of lactic acid, so that in many cases, the appearance of the digesting organisms on the surface is delayed until the acidity of the mass is reduced to the proper point by the development of other organisms, principally molds, which prefer an acid sub-

stratum for their growth. In Brie cheese a blue coating of mold develops on the surface. In the course of a few weeks, a white felting appears which later changes to red. This slimy coat below the mold layer is made up of diverse species of bacteria and fungi that are able to grow after the acid is consumed by the blue mold. The red coating acts upon the casein, also producing an alkaline reaction that is unfavorable for the growth of the blue mold. Two sets of organisms are therefore, essential in the ripening process, one preparing the soil for the ferment that later produces the requisite ripening changes. The process as carried on is largely empirical and if the red coat does not develop at the proper time, the maker resorts to all sorts of devices to bring out the desired ferment. The appearance of the right form is dependent, however, upon the proper reaction of the cheese, and if this is not suitable, the wished-for growth will not appear.

In those cases where the ripening change takes place from outside toward the center, there is always an outer layer that is changed to such an extent as to be unfit for food.

B. BACTERIA IN ABNORMAL CHEESE PROCESSES.

173. Susceptibility of cheese to abnormal changes. Cheese more than butter is subject to detrimental fermentations incited by bacteria, because it is so much better adapted for germ growth on account of its better and more available food-supply. Many abnormal ferments that find their way into the milk before it is made up develop these peculiarities, especially in cheese. Then too, the method of curing is so often without control, especially with many of those methods that employ fungi as agents in the curing process. Inability to check the

ripening at the proper moment often results in the decomposition processes being carried too far, and in this way unpleasant flavors are developed.¹

174. Indefinite faults in cheddar cheese. Besides a number of more or less distinct fermentations of diverse character, ill-defined defects in flavor and odor are often to be noted. Cheese possessing these may be thoroughly ripened and therefore digestible, but their market value is much diminished by reason of these improper flavors. In many cases, troubles of this sort are undoubtedly traceable to faulty manufacture, but more often, the defects arise from the presence of some fermenting organism that is the cause of the unpleasant flavor. Our knowledge of the influence that the different species have in the ripening changes of cheese is still too meager to enable us to trace, in all cases, these undesirable conditions to their proper source. The great majority of the taints observed in the factory are due to the abnormal development of some of these forms capable of evolving unpleasant or even putrid odors. Most of them are seeded in the milk before it comes to the factory and are due to careless manipulation of the milk while it is still on the farm. Others gain access to the milk in the factory, owing to unclean conditions of one sort or another. Sometimes the cheese-maker is able to overcome these taints by vigorous treatment, but often they pass on into the cheese only to detract from the market value of the product.

In studying the effect of the different organisms found in milk in their relation to cheese-making, we have isolated a number of different species that are concerned in

¹ Adametz has brought together in his little book entitled, *The causes of the abnormal ripening of cheese*, a large amount of data that pertains especially to European conditions.

the production of these "off" flavors in the curd. If these organisms are seeded in pasteurized milk and the infected milk made into cheese, they develop odors that are sometimes noted in factories troubled with taints. These indefinite "off" flavors and odors are often insufficiently pronounced to be given a name, consequently, it will be impossible to describe them under any well defined head.

175. "Gassy" fermentations in cheese. One of the worst and at the same time most common troubles in cheese-making is where the cheese undergoes a fermentation marked by the evolution of gas. The presence of gas is recognized by the appearance either of spherical or lens-shaped holes of various sizes in the green cheese; often they appear in the curd even before it is put to press. Usually in this condition, the curds look as if they had been finely punctured with a pin, and are known as "pin-hole" curds. Sometimes the gas holes are larger, even approaching the large round so-called "Swiss holes." When the gas is abundant, the holes are more apt to be restricted in size. The formation of gas may continue to such an extent that the curd even floats on the surface of the whey before it is removed. These "floating curds" are permeated through and through with gas bubbles, giving the curd a "spongy" appearance.

If "gassy" curds are put to press in this condition, an abnormal change usually occurs within a few days. The fermentation goes on in the green cheese causing it to swell or "huff," until it may be considerably distorted. The gas diffuses with some difficulty in the new cheese so that the mass rapidly swells. The fermentation may be so energetic as to actually cause the cheese to crack, owing to the pressure of the contained gas. In the severe types of this gaseous fermentation, the product is

rendered worthless, but even where the development is not so marked, the flavor of the cheese is impaired and the market value diminished. The difficulty may occur at almost any season of the year, but the trouble is most frequently observed in the late summer months. So common are these difficulties that they may be regarded as the worst cheese troubles with which our cheese-makers have to contend.

This same defect occurs in other kinds of cheese, but it is particularly marked in those that are made from relatively sweet curd. In Emmenthaler, or Swiss cheese, the formation of large gas holes is called "blähen." A closely related phenomenon is seen in "nissler" formation, where very numerous small holes are formed (thousand eyes).

The cause of the difficulty has long been charged to various sources, such as lack of aeration, improper feeding, retention of animal gases, etc., but in all these cases it was nothing more than a surmise. Very often the milk does not betray any visible symptom of fermentation when received, and the trouble is not to be recognized until the process of cheese-making is well advanced.

Recent studies from a biological standpoint have, however, thrown much light on this troublesome problem; and, with the *cause* of these fermentative changes more fully recognized, it is quite probable that improved methods of handling milk will be rapidly introduced that will enable the maker to exclude these troubles.

176. Relation to living germs. The formation of gas, either in the curd or after it has been put to press, is due entirely to the breaking down of certain elements, such as the sugar of milk, under the influence of various living germs. This trouble is then a type fermentation, and is therefore, much more widely distributed than it

would be if it was caused by a single specific organism. There are present in all milks a few bacterial forms that are able to produce a varied series of fermentations in which different gases may be given off. Among these organisms are a large number of the bacteria, although yeasts and allied germs are often present in milk and are likewise able in some cases to set up fermentative changes of this sort. In these cases the milk-sugar is decomposed in such a way as to give off CO_2 and H_2 , and in some cases, alcohol.

According to Guillebeau a close relation exists between those germs that are able to produce an infectious inflammation (mastitis) in the udder of the cow and some forms capable of gas evolution. Several outbreaks of "gassy" milk have been traced directly to animals suffering from an acute inflammatory condition of the udder in which it has been shown that the organisms producing this disease were the direct cause of the gas production in the milk.

Gas-producing bacteria are so numerous that they are almost always present in any sample of milk, especially if it is a little old. Bolley and the writer isolated six different species from a single sample of summer milk. Even in winter we have found them present in milk in such numbers as to require special care in the manufacture of cheese. Under normal conditions, where care is taken in the handling of the milk, they are not usually found in large numbers, and when thus numerically restricted, are doubtless kept under subjection by the competition of numerous other bacteria in the milk.

These fermentations are very often observed in foreign kinds of cheese, especially those that are made after the sweet-curd process. Adametz¹ has collated data on the

¹ Adametz, Die Ursachen u. Erreg. d. abnorm. Reif. Vorg. b. Kaese, p. 27.

European forms that have been isolated, and has found nineteen bacterial species (five cocci, fourteen bacilli) and eight varieties of yeast-like fungi that are gas-producing organisms.

If pasteurized milk is seeded with a pure culture of any of these gas-forming organisms and made into cheese, an intense fermentation is always to be noted. This often appears in the curd, sometimes even before the whey is drawn. The cheese when taken from the

FIG. 34. "Gassy" cheese. The milk from which this cheese was made was first pasteurized, and then infected with a gas-generating bacillus. Note not only the "spongy" texture, but the distorted form.

press develops gas rapidly, causing it to swell and a cross section of it at this stage will show a spongy structure. If the cheese is left intact, the fermentation may progress to such an extent that the pressure of the contained gas will cause the rind to split open. This fermentation does not last for more than a few days; then the swelled cheese sinks as the gas slowly diffuses throughout its mass, but the flavor of the product is generally impaired.

177. Gaseous fermentations in Swiss cheese.

From the researches of v. Freudenreich, Baumann, and Weigmann, it is known that certain gas-producing forms invariably play a rôle in the normal ripening of Emmenthaler cheese. Sometimes Swiss cheese is devoid of these gas holes, or "eyes," in which case, they are said to be "blind." Such cheese may ripen well but their market value is diminished on account of the absence of these round holes.

The size of the gas holes as they appear in abnormal cheese is by no means constant. Neither is the character of the hole dependent upon any specific germ. If the gas-forming bacteria are plenty and quite evenly distributed the holes will be small and numerous as in

FIG. 35. A block Swiss cheese, showing "gassy" fermentation.

"nissler" cheese. Where the organisms are less frequent and develop in small groups, then the "eyes" are much enlarged. As the different organisms vary in their intensity of gas formation, this too, modifies the size of the gas holes to a marked degree.

178. Treatment of "pin-hole" curds. When this type of fermentation appears during the manufacture of the cheese, the maker can control it in part within certain limits. These methods of treatment are, as a rule, purely mechanical; in other cases they are to place the curds or the green cheese under such conditions as to prevent the development of gas. One of the methods that is purely mechanical in its nature is the piling and turning of the curds and then the subsequent grinding of the same in a curd mill. This breaks up the curd layers and allows the gas to escape. After the gas has been forced out, the curds are then put to press and the whole mats down into a compact mass.

Another method of treatment based upon bacteriological principles is the addition of a starter to induce the formation of acid. Where acid is developed as a result of the growth of the lactic acid bacteria, the gas-producing species do not readily thrive; therefore, the development of gas may be controlled in part in this way. Another reason why acid aids in repressing the development of gas is that the curd particles are partially softened or digested by the action of the acid. This causes them to mat together more closely and there is not left in the cheese, the irregular openings where the curd particles fail to unite and in which the developing gas finds lodgment.

Another method that is also useful with these curds is to employ salt. This represses gaseous fermentations and the use of more salt than usual in making the cheese will very often restrain the production of gas. Tendency to form gas in Edam cheese is controlled by the addition of a starter prepared from slimy whey (*lange wei*) which is caused by the development of an acid-forming organism.

The temperature at which the cheese is cured also ma-

terially affects the production of gas. At high curing temperatures, the gas-producing bacteria are able to develop more rapidly; therefore, more trouble is experienced in summer than at other seasons. The relation of temperature of curing-room to the ripening of cheese is a question of very great importance. We may, roughly speaking, divide normal temperatures into three zones more or less well marked, safe, hazardous, and dangerous. Where the temperature exceeds 70° F., the ripening of the cheese is apt to be impaired. At these high temperatures, gas-generating forms are apt to become prominent.

179. Detection of tainted milks. Taints in milk may be due to two sets of causes. They may be produced by bacterial agency or absorbed directly from some pre-existing source. Those belonging to the first class are extremely difficult to detect at the weigh-can, for the taint itself may not be pronounced, while the organism causing it may exist in the milk in large numbers. Taints of this class are especially dangerous, for they are produced by vital causes which may further develop, and thereby greatly intensify the undesirable condition. Taints due to absorption can, if prominent, be recognized by sense of smell. A detection of these is facilitated by warming the milk, thus permitting the contained odors to escape more readily.

It is extremely desirable that some method of recognizing these taints of microbic origin should be employed, but thus far, no rapid way has been devised that is applicable at the weigh-can. It is possible, however, to treat the milk by making what is known as a fermentation test, and thus encourage the early development of the contained bacteria, but these methods are only of use in preventing future troubles.

180. Fermentation tests. Fermentation tests are based upon the principle that if milk is held at a moderately high temperature, approximating the blood heat, the bacteria in the same will develop rapidly. Particularly is this true with taint-producing organisms as these seem to develop more rapidly at the higher temperature.

A number of tests have been devised that are based upon the operation of the same general bacteriological principle. In Walther's lacto-fermentator the milk is allowed to stand in bottles or glass jars until it sours¹. To hasten this process it is incubated in a warm place. If the curd formed is homogeneous and has a pure acid smell, the milk is regarded as all right. If it floats in a muddy serum, is full of gas or ragged holes, it shows abnormal conditions. As usually carried out, no special attempt is made to free the vessels used for this purpose from various kinds of bacteria. Often, too, the samples are left uncovered, thus permitting infection to occur from without. The utility of this method could be materially increased by using sterile tubes and having same protected by a plug of cotton from external contamination.

A few drops of rennet are sometimes added to the milk that is incubated in order to enable a more ready detection of the gas that is evolved. The rents and splits in the curdled casein are rendered more evident in this way.

181. Casein or rennet tests. The action of rennet on milk can be utilized in determining its value for cheese purposes. If a given quantity of rennet solution is added to a definite quantity of milk at a certain temperature, the time of coagulation will give some indication as to the character of the milk. Good milks ought to curdle

¹ This same principle is employed in Gerber's test.

quickly, and the curd should be uniform and firm. If it is soft or tender, and does not curdle within the usual time (which will vary, of course, with the proportion of rennet and milk used) it shows an abnormal condition, due generally to the addition of water or chemical substances. Milk from cows advanced in lactation (strip-pers) not infrequently reacts abnormally with the rennet test. This test rests upon the chemical rather than the biological characteristics of the milk.

182. Wisconsin curd test. The curd test in some respects is a modification of the fermentation test, yet the method is sufficiently different in detail to merit separate mention. As here outlined it originated at the Wisconsin Dairy School in 1895, as an experimental method of studying the influence of gas-generating bacteria in cheese-making¹.

In the curd test, a small pat of cheese is made in a glass jar from each sample of milk. The advantage of this method over the ordinary fermentation tests lies in the fact that the samples of milk are treated in a way so as to conform more closely to cheese conditions, inasmuch as the whey is removed, thus permitting the ready recognition of gas-production in the curd. Nearly all milks contain some bacteria capable of decomposing the milk-sugar and producing gas. In the curd test this slight evidence does not appear, but if the milk contains an abnormally high content of gas-generating bacteria, sufficient to injure the quality of the milk for cheese-making, then such a condition becomes evident when a curd test is made.

183. Improvised apparatus. A simple, inexpensive apparatus can be readily made by any operator in the

¹ 12th Rept. Wis. Expt. Stat., p. 148, 1895. A full description of this test is given in Bull. 67, Wis. Expt. Stat., June, 1898.

following way: Take a number of pint glass fruit jars and sterilize the same with the covers in boiling water. Fill the bottles two-thirds full with milk from the individual patron's supply. Place the bottles in a wash tub and fill the same with warm water. The temperature of the water after the tub is filled should be from 95–100° F. When the milk reaches 98° F., add to each sample by means of a pipette, ten drops of rennet extract and mix thoroughly. Allow the jars to remain undisturbed until the milk is thoroughly curdled, then cut the curd into

FIG. 36. Improved bottles for making curd test. *A*, test bottle complete; *B*, test bottle showing construction of cover; *s*, sieve to hold back the curd when bottle is inverted; *c*, outer cover with *d h* drain holes to permit of removal of whey.

small particles with a case knife, stirring the same to better expel the whey. Pour off the whey as soon as the curd settles, repeating this process at frequent intervals until the curd mats. The excess of fermentable sugar is thus removed. Keep the temperature of water at blood heat (98° F.) for six to eight hours, and then examine the cut surface of the curds.

184. Improved apparatus. A more convenient form of apparatus has been devised by various dairy manufacturers. This consists of a water-tight box, with cover, which is provided with a faucet for the withdrawal of the water; also a rack to hold the bottles. The test bottles are made with parallel sides, the large top permitting of the easy removal of the curd. The tops are provided with a sieve of such construction that the bottles will drain thoroughly if inclined in an inverted position (fig. 36).

FIG. 37. Curd from a good milk. The large irregular holes are mechanical.

185. Interpretation of results of test. The curd from a good milk has a firm, solid texture, and should contain at most only a few small pin holes. It generally has a few large, irregular, "mechanical" holes where the curd particles have failed to cement as is seen in fig. 37. If gas-producing bacteria are very prevalent in the milk, the conditions under which the test is made cause such a rapid growth of the same that the evidence of the abnormal fermentation may be readily seen in the spongy texture of the curd. If the undesirable organisms are not very abundant and the conditions not especially suit-

able to their growth, only small "pin holes" may appear; if, on the contrary, the fermentative process takes a more violent course, such as would be the case in a "floating" milk, then the test curd will be spongy and full of holes as in fig. 38. Sometimes the curds will show no evidence of gas, but their abnormal condition can be recognized from the "mushy" texture and the presence of "off" flavors that are materially intensified by keeping them in closed bottles. The odor should always be noted with care, as this is of more importance than the gas which can be eliminated to a large extent by proper manipulation during the manufacture of the cheese.

FIG. 38. Curd from a badly tainted milk. Large ragged holes mechanical small "pin holes" due to gas.

186. Bitter cheese. Bitter flavors are sometimes developed in cheese especially where the ripening process has not been fully completed, or where improper temperatures have been maintained for a considerable length of time. Several organisms associated with this abnormal fermentation have been noted.

Guillebeau¹ isolated several forms from Emmenthaler

¹ Guillebeau, Landw. Jahr., 1890, p. 27.

cheese which he connected with udder inflammation that were able to produce a bitter substance in cheese.

Von Freudenreich¹ has recently described a new form, *Micrococcus casei amari* (micrococcus of bitter cheese) that was found in a sample of bitter Swiss cheese. This germ is closely related to Conn's micrococcus of bitter milk. It develops lactic acid rapidly, coagulating the milk and producing an intensely bitter taste in the course of one to three days. When milk infected with this organism is made into cheese, there is formed in a few days, a decomposition product that imparts a marked bitter flavor to the cheese.

FIG. 39. "Floating" or "spongy" curd from a very bad milk.

It is peculiar that some of the organisms that are able to produce bitter products in milk do not retain this property when the milk is worked up into cheese.

187. Putrid or rotten cheese. Sometimes cheese undergoes a putrefactive decomposition in which the texture is profoundly modified and various foul-smelling gases are evolved. These often begin on the exterior as small circumscribed spots that slowly extend into the

¹ Freudenreich, Fuehl. Landw. Ztg., **43**: 361.

cheese changing the casein into a soft slimy mass. Then, again, the interior of the cheese undergoes this slimy decomposition. The soft varieties are more prone toward this fermentation than the hard, although the firm cheeses are by no means exempt from the trouble. The "verlaufen" or "running" of limburger cheese is a fermentation allied to this. It is where the inside of the cheese breaks down into a soft semi-fluid mass. In severe cases, the rind may even be ruptured, in which case, the whole interior of the cheese flows out as a thick slimy mass having sometimes a putrid odor. The conditions favoring this putrid decomposition are usually associated with an excess of moisture, and an abnormally low ripening temperature.

188. Pigment changes in cheese. Occasionally with hard cheeses, but more often with the softer foreign varieties, abnormal conditions are noted that express themselves in the production of various pigments in the different sorts of cheese. The production of these variously colored pigments are due mainly to the action of bacteria, yeasts, and molds. More frequently they are merely superficial, and affect only the exterior layers of the cheese.

1. *Rusty cheese.* This difficulty has been the source of considerable trouble to makers of white or uncolored cheddar cheese in some sections of America. It is characterized by the appearance, especially in July and August, of rusty red or orange spots that are noted throughout the interior of the cheese, particularly along the edge of the curd particles. Connell¹ traced an outbreak of this sort in a Canadian factory to defective drains, and succeeded in isolating a red chromogenic

¹ Connell, Rept. of Com. of Agric. and Dairying, part xvi; p. 14, 1898.

bacillus that when inoculated into milk, and cheese made therefrom, produced the rusty discoloration. A similar defect has been studied by Campbell¹ in Scotland. He used a pure lactic ferment with good results in overcoming the tendency to discoloration.

2. *Blue cheese.* Blue discoloration in cheese sometimes arises from copper and iron salts derived from manufacturing utensils. Especially is this liable to occur with foreign cheese where utensils of such a character are used in the manufacture of these cheese. De Vries² has described a blue disease in Edam cheese that he ascribes to the action of bacteria. It appears first as a small blue spot on the inside, increasing rapidly in size until the whole mass is often affected. By the use of slimy whey, this abnormal change is controlled.³

3. *Black cheese.* Black cheese have now and then been noted, especially with limburgger products. This appearance is caused by the copious growth of different forms of low fungi, mainly those that spread out in tiny threads, like the molds. In one instance a yeast-like form has also been isolated. So far as known troubles of this sort are not caused by bacteria.

189. Molding of cheese. With some varieties of cheese, more especially foreign kinds, the presence of mold on the exterior of cheese is not regarded as detrimental; in fact the limited development of this condition is much desired. In hard rennet cheese, such as Emmenthaler and Cheddar, the market demands a product free from mold, although it must be said that this condition is imposed by the desire to secure a good looking cheese rather than to any injury in flavor that the mold might cause.

¹ Campbell, North Brit. Agric., May 12, 1897.

² De Vries, Milch. Ztg., 1888, Nos. 44, 45.

³ Weigmann, Cent. f. Bakt., II. Abt., 4: 607, 1898.

Development of mold invariably takes place in a moist atmosphere, either at high or low curing temperatures. These are frequently the conditions that are required in curing certain types of hard American cheese, so that mold is apt to appear.

The use of a double bandage or rubbing the cheese when ready for market will remove this blemish. Recent experiments with certain disinfectants like formalin show that a 2 % solution rubbed over surface will retard the development of mold.

190. Poisonous cheese. The production of poisonous ptomaines in cheese is perhaps more common than with almost any other food product. Vaughan¹ to whom the most of the work on this subject is to be credited, reports over 300 cases of cheese-poisoning in two years. It seems to be very much more common here in America than it is in Europe. Vaughan has isolated from numerous samples a highly poisonous alkaloid that he calls *tyrotoxicon*. This ptomaine has also been frequently demonstrated in milk, cream, and ice cream. The poisonous substances formed in the milk are probably produced through the agency of putrefactive organisms that gain access to the milk.

The poisons secreted in the milk are transferred to the cheese with their virulence often unimpaired, so that serious complications follow where the infected material is used for human food. Vaughan states that cheese which reddens litmus paper rapidly should be regarded as suspicious. In normal cheese, this reaction occurs very slowly. Danger from this source is to be noted particularly with the so-called sour milk cheese; as the methods by which these are made give the most favorable oppor-

¹ Vaughan, Zeit. f. physiol. Chemie, 10: 146.

tunity for the development of poisonous decomposition products.

191. Prevention of cheese defects. In attempting to treat the various defects or diseases found in cheese, no uniform method can be applied in all cases. Excluding faults due to manufacturing methods, most of the troubles traceable to bacteria originate in the milk before it comes to the factory.

In some instances the maker can repress or subdue the influence of these undesirable ferments by changing his methods somewhat to suit the varying conditions. But what he must insist upon under all circumstances is that the milk shall be handled by the patron in such a way as to exclude as far as possible all conditions favoring the access of any organisms into it. If this is done and pains are taken to point out to the individual patron the way in which his milk becomes infected, then the maker may expect to reap the reward for his teachings in the improved condition of the milk.

GLOSSARY.

- Adventive.—Species introduced from foreign sources.
- Aeration.—The intimate mixing of air with the milk.
- Aerobes.—Bacterial organisms requiring free oxygen for growth.
- Algæ.—Green water plants of simple organization.
- Amido-acids.—Organic acids formed in proteid decomposition.
- Anaerobes.—Bacteria growing without free oxygen.
- Anthrax (splenic fever).—A contagious animal disease characterized by blood poisoning.
- Antiseptics.—Chemicals capable of restraining bacterial growth.
- Bacillus (plural bacilli).—Straight, rod-like bacteria.
- Brownian movement.—A physical movement independent of vital force.
- Carbohydrates.—Organic substances like sugars and starches.
- Casease.—The enzyme capable of converting casein into soluble compounds.
- Caseone.—Casein that has been rendered soluble by enzymes.
- Cell.—The simplest form unit of vegetable or animal life.
- Chlorophyll.—The coloring matter in leaves of green plants.
- Chromogenic.—Color-producing.
- Cilia (singular cilium).—Tiny, whip-like protoplasmic appendages of cells, serving as locomotor organs.
- Coccus (plural cocci).—Bacteria having a spherical shape.
- Colony.—The progeny of a single germ growing in an isolated mass.
- Culture medium.—A food substance on, or in which bacteria can grow.
- Culture starters.—Starters for use in butter or cheese-making that are developed from a selected bacterial culture.
- Deodorants.—Chemicals that destroy odors.
- Diastase.—The starch-converting enzyme.
- Disinfectant.—Any chemical substance able to destroy germ life.
- Endospores.—Spore formed within a mother cell.
- Enzymes.—Unorganized chemical ferments not endowed with life.
- Facultative.—Forms that possess the faculty of growing under varied conditions.
- Filmjölkk.—Ropy milk of Finland.

- Fission.—Division of a cell by direct partition.
- Formalin.—A watery solution of formic aldehyde gas possessing powerful disinfecting properties.
- Galactase.—An unorganized ferment inherent in milk which digests casein in ripening cheese.
- Germicide.—Any substance capable of destroying germ life.
- Hydrophobia.—A highly contagious disease, often occurring in dogs, wolves, etc., but communicable to man.
- Hydrocarbons.—Organic material like the fats and oils.
- Indigenous.—Bacteria normal to any given habitat.
- Lange wei (slimy whey).—A viscous fermentation of whey used as a starter in Edam cheese.
- Mammalia.—Animals that suckle their young.
- Mammitis.—Inflammation of the udder.
- Matzoon.—A fermented milk used largely in Armenia.
- Mother cell.—A vegetable cell capable of reproduction.
- Obligate.—Bacteria that are obliged to grow under certain conditions.
- Optimum growth temperature.—The best temperature for the growth of any form.
- Parasites.—Organisms subsisting on living matter.
- Pasteurization.—The use of heat from 140° to 165° F. as a germ destroyer.
- Pathogenic.—Bacteria able to produce disease in living tissue.
- Pepsin.—An enzyme capable of digesting proteids in acid solution.
- Peptones.—Soluble products of nitrogenous digestion.
- Preservaline.—A proprietary antiseptic composed mainly of boric acid.
- Proteids.—Complex nitrogenous substances of an insoluble nature.
- Protoplasm.—The living substance of organic tissues.
- Ptomaine.—A complex nitrogenous product of bacterial growth.
- Pure Culture.—A bacterial growth of a single species in a sterile medium.
- Rennet.—The curdling enzyme of milk.
- Saccharomyces.—A genus of plants embracing the yeasts.
- Saprophytes.—Organisms subsisting on dead organic matter.
- Sarcina.—Spherical bacteria forming packets of cells.
- Spirillum (plural, spirilla).—Curved or bent cylindrical bacteria.
- Spores.—The latent or resting stage of certain types of bacteria.
- Sterilization.—The use of heat in the neighborhood of 212° F. as a germ destroyer.

Streptococcus.—Spherical bacteria growing in chains.

Tættemjök.—The clotted milk of Norway.

Thermal death-point.—The temperature at which the cell is destroyed.

Toxic.—Poisonous.

Trypsin.—The active principle of the pancreatic secretion.

Tyrotoxin.—A poisonous ptomaine isolated from milk or cheese.

Tyrothrix.—Digesting bacteria supposedly concerned with cheese-ripening.

Viscogen.—A solution of lime dissolved in sugar used in restoring the consistency of cream that has been heated above 145° F.

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