Milk and the National Defense

As we engage in the immediate task of strengthening our defenses, we may be inclined to think entirely in terms of airplanes, naval craft, guns, and other such armaments. Without doubt, these are necessary. But there are other weighty considerations about which we do not seem to hear much, if anything. These are concerned with the morale and strength of the people themselves.

An epidemic in a community may seriously handicap the defense program. The recent series of sit-down strikes demonstrated that when an important power house or some key plant that produced a necessary part was closed down, a huge industry stopped production. We recall the case where an epidemic in a manufacturing town necessitated the closing of a large factory because so many of the employees were sick. As new plants are erected in what are now open fields, as great numbers of workmen create correspondingly new communities, and as the tempo of production is stepped up, there must be an attendant creation of an adequate and safe food supply. As our Dr. Jones colorfully says, "Where they're rushing night and day turning out airplanes and ships and guns and food and what not—stuff that's needed to meet an emergency—well, it don't take any great imagination to see what a handicap it (an epidemic)’d be . . . An epidemic’s an emergency in itself and one emergency at a time’s about enough . . ."

Military centers themselves are one of best (!) breeders of epidemics. Dr. W. S. Leathers, President of the American Public Health Association, states, "The protective measures of general sanitation around army camps and cantonments can be greatly facilitated by thorough coordination between the state and local health agencies and the military population. Well prepared and experienced sanitation officers are required in the enforcement of regulations, in providing a pure water supply, in the proper supervision of milk, and in the safe disposal of sewage. A careful inspection of food supplies to prevent contamination, and general cleanliness of the vicinity in cooperation with the officials of military camps must be strictly enforced."

But the situation has still another important aspect. Dr. George W. Cox, Texas State Health Officer, says:

"Defense in its broadest sense means security. It is a great satisfaction to have a feeling of security against foreign aggression, internal revolution, unemployment,
need, hunger, and communicable disease. We treasure freedom of speech, freedom of action, and freedom to exercise initiative. The sole defense program means more than air power and armament—it includes an enormous development that has been so guided that they can meet and survive the pressures brought upon them whether in war or peace. It includes food, clothing, shelter, drugs, and thousands of articles from mine and field...

"Some of our statesmen fear disturbance from within our borders more than aggression from abroad. These statesmen are far-seeing men who have looked down into the mass of humanity and recognized these individuals who, through poverty, disease, and undernourishment have become warped either mentally, emotionally, or physically... If relatively simple diseases such as cold and hayfever may bring about bad tempers, how much more may chronic illness, malnutrition, and starvation lead the individual to react violently to his surroundings. We must maintain a high morale in our population...

But we have more than our present "home fires" to consider. New fires are being lighted as industrial centers spring up and flourish. Our former president, Mr. V. M. Eliers, writes: "There is work to be done by the milk sanitarians—milk to be provided for the new centers of population that are being created, milk to be properly supplied to keep down disease, milk for the young, and milk for the building of man power."

These new centers of population, arising out of our increased defense program, will require the opening of new milk-producing territory, the employment of new help, the erection of new plants. When such enterprises are undertaken in a hasty manner, there is a tendency for important but unrecognized principles of public health to be neglected. A corollary of such a development would be the diversion of necessary milk supplies from a given community to some large cannery or other newly constructed series of plants or community.

Will the new payrolls be used entirely for the purchase of new cars, new radios, and new clothes—or will the necessary part go to providing the family table with not only the proper quantity but also the proper quality and variety of food? Unless there is an effective educational program for these groups who have suddenly come into greater prosperity than they have heretofore enjoyed, it is probable that current nutritional deficiencies will not be corrected. With such a background to the faster tempo of living with its accompanying increased output of work, we might expect decreased resistance to disease, an undermining of morale, and a curtailment of production.

So we see that national needs are facing us. We should tighten our defenses against the inroads of epidemics. We should improve our nutritional health, so to speak. We should provide for a safe supply of milk to the new communities, and also to the present ones now well supplied. We should educate the public on the necessity of buying the proper food. In addition to these, we should provide the government with information as to what type of training should be given the officers of the sanitary service of the army and the other officials who will be responsible for administering the newly established communities.

To insure that the above aspects of the defense program get off to a good start with a minimum of misdirection, we suggest that the President of the INTERNATIONAL ASSOCIATION OF MILK SANITARIANS appoint a Committee on Defense. This committee would make the necessary contacts with governmental and other professional and civic agencies concerned, to the end of marshalling our technical knowledge for the prevention of epidemics from infected milk, for supervising the production of a safe supply, and for educating the public on the health value of an adequate diet.

J. H. S.
to methods of freeing such utensils from pathogenic microorganisms.

PROCEDURE FOR THE DETECTION OF ORAL CONTAMINATION OF DRINKING GLASSES

During the progress of the present investigation, the following procedure has been developed as a test for oral streptococci found on lips or on the rims of drinking glasses.

An ordinary test tube containing a small, tightly wound cotton swab made on a 7-inch wood applicator which extended through the cotton plug of the tube, was sterilized at 15 pounds pressure for 15 minutes. Prior to taking the sample, the swab may be moistened in sterile broth; however, if the glass to be examined is moist, this step can be eliminated. Samples were taken by swabbing three times around the inner and outer rim of the glass approximately one-half inch from the rim, using a slow rotary motion. The swab was then placed in a test tube containing an enrichment medium (10 ml of 1 percent lactose yeast infusion broth containing crystal violet in a concentration of 1-400,000, adjusted to pH 7.6), and incubated for 24 to 48 hours at 37°C.

The type of growth which is important was observed at the end of 24 hours incubation. A flocculent growth attached to the swab proved through many instances to be the species to which they belonged.

Of particular significance in the incubation of swabs is the type of growth which appears when inoculations have been made from drinking glasses known to carry an oral contamination. The characteristic growth of streptococci in these cultures serves as a most acceptable presumptive index of the presence of oral streptococci.

THE PRESENCE OF STREPTOCOCCUS SALIVARIUS ON HUMAN LIPS

Early in the investigation it was felt that if a study was to be made of oral contamination of drinking utensils, information should be available on the type of organisms which predominate on human lips, hence the lips of 100 individuals selected at random were examined. The outer portion of the closed lip was swabbed with moistened swabs and the organisms present were determined by the procedure outlined above. Of 100 individuals (Table 1) it was found that all carried streptococci on the outer portions of the lips. The 340 strains isolated from the various lips appeared to be of one type. All of the strains failed to produce acid from mannitol and sorbitol, and practically all reduced lactose subsequent to the curdling of the milk. The fermentation of raffinose and trehalose was somewhat variable. With this information at hand, it would appear that these streptococci were all of the Streptococcus salivarius Andrews and Horder type.

No information was obtained as to the relative numbers of Streptococcus salivarius on the individual lips. The use of the selective enrichment medium precluded making estimates of this type. From these data it is evident that the lips of practically all individuals carry streptococci identified as Streptococcus salivarius or closely related types.

THE DEPOSITION OF STREPTOCOCCUS SALIVARIUS ON DRINKING GLASSES

If the presence of Streptococcus salivarius is to be used as an index of oral contamination of drinking glasses, it is important that definite information be available as to the length of time that this organism will survive on the rims of drinking glasses, providing the utensil has received no washing or sterilizing treatment. A study of 150 drinking glasses which received no treatment after use (Table 3) indicated that Streptococcus salivarius will usually survive on the rims of drinking glasses at least for 48 hours when held at room temperature (22°C). This was particularly true when the glass was used for dispensing water. In some instances, however, the glass was used for dispensing milk, the streptococci appeared to survive for a somewhat shorter length of time.

From these results it would appear that the presence of Streptococcus salivarius on the rims of drinking glasses would indicate that it has been orally contaminated within 48 hours and has received little or no washing or sterilizing treatment.
Further studies (Table 3) on the effect of a rinse in cold tap water subsequent to use indicated that such a treatment may reduce in part the survival of *Streptococcus salivarius* but not the universal presence of the organisms on the glass following its use. A study of 160 cases in which both water and milk were dispensed from the glasses, indicated that *Streptococcus salivarius* could be found on drinking glasses immediately after such use but that the number of organisms began to decrease following 4 to 8 hours when held at room temperature. However, *Streptococcus salivarius* could still be recovered after 24 to 30 hours.

A study of the effect of a double rinse as well as washing in soapy water at 120° F. followed by various types of rinses indicated (Table 4) that when such procedures were used the number of instances in which *Streptococcus salivarius* could be recovered was materially reduced. It was found that in a series of examinations, washing and rinsing in cold tap water did not reduce the incidence of *Streptococcus salivarius* on the rims of drinking glasses when examined immediately following this treatment. However, in over 33 percent of the cases, micrococci were found following such a treatment, whereas in the case of the Gram-negative rods of the coliform type only 16 percent of those originally showing these organisms were still found to contain the coliform types of organisms following washing and rinsing in cold tap water. If subsequent to use drinking glasses were washed in water at 120° F. without soap and subsequently rinsed in water with the same temperature, 30 percent of the glasses which originally contained streptococci were found to be free of this organism following such a treatment. However, where soap was added to the original water (pH 8.6) at 120° F., 70 percent of the glasses on which streptococci were originally found proved to be free of this organism after such treatment. It is of particular interest also to note that washing in a soapy water at 120° F. followed by a rinse for five minutes at 165° F. completely removed, in all cases studied, streptococci, micrococci and Gram-negative rods from the rims of drinking glasses. This temperature is of interest insofar as glasses are concerned. By examination it was found that tap "hot water" rarely exceeds 165° F., while the report (1956-1957) of the sub-committee of the American Public Health Association suggests a rinse water of 170 to 180° F.

The results further would indicate that *Streptococcus salivarius* is an acceptable index for the oral contamination of drinking glasses and as a further aid to the use of soapy water for washing together with 165° F. rinse would serve as an effective procedure for eliminating this streptococcus.

### Table 3

**Survival of lip organisms on drink glasses held at room temperature (22° C)**

<table>
<thead>
<tr>
<th>No. of glasses</th>
<th>0</th>
<th>2</th>
<th>4</th>
<th>8</th>
<th>24</th>
<th>48</th>
<th>72</th>
<th>100</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of glasses</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Time in hours</td>
<td>0</td>
<td>2</td>
<td>4</td>
<td>8</td>
<td>24</td>
<td>48</td>
<td>72</td>
<td>100</td>
</tr>
</tbody>
</table>

### Table 4

**The effect of washing on survival of organisms on orally contaminated glasses.**

<table>
<thead>
<tr>
<th>Percent of glasses showing organisms after treatment</th>
<th>Washed in water at 120° F.</th>
<th>Washed in soap water at 120° F.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Organisms on lip of glass</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Streptococcus</em></td>
<td>100</td>
<td>70</td>
</tr>
<tr>
<td><em>Micrococcus</em></td>
<td>55</td>
<td>35</td>
</tr>
<tr>
<td>Gram-negative rods</td>
<td>16</td>
<td>10</td>
</tr>
</tbody>
</table>

**GLASSES USED FOR DISPENSING WATER**

- Washed in water at 120° F.
- Washed in soap water at 120° F.
- With 5 min. rinse in 120° F. water

Further studies (Table 3) on the effect of a rinse in cold tap water subsequent to use indicated that such a treatment may reduce in part the survival of *Streptococcus salivarius* but not the universal presence of the organisms on the glass following its use. A study of 160 cases in which both water and milk were dispensed from the glasses, indicated that *Streptococcus salivarius* could be found on drinking glasses immediately after such use but that the number of organisms began to decrease following 4 to 8 hours when held at room temperature. However, *Streptococcus salivarius* could still be recovered after 24 to 30 hours.

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**ORAL CONTAMINATION OF DRINKING GLASSES AS ENCOUNTERED IN PUBLIC EATING ESTABLISHMENTS**

On the assumption that the presence of *Streptococcus salivarius* is an index of the oral contamination of drinking glasses, a survey was made of about 200 (Tables 5 and 6) eating establishments to determine the presence of this organism as well as micrococci, Gram-negative rods and Gram-positive rods on the rims of drinking glasses. It was found that a wide variation occurred among various types of eating establishments (Table 5) and even...
of drinking glasses in public eating establishments.

<table>
<thead>
<tr>
<th>Number of establishments</th>
<th>City</th>
<th>Streptococci</th>
<th>Micrococci</th>
<th>Gram-Neg.</th>
<th>Gram-Pos.</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>50</td>
<td>A</td>
<td>39</td>
<td>81</td>
<td>12</td>
<td>8</td>
<td>28</td>
</tr>
<tr>
<td>58</td>
<td>B</td>
<td>64</td>
<td>30</td>
<td>24</td>
<td>8</td>
<td>32</td>
</tr>
</tbody>
</table>

between different municipalities. This variation obviously is to be expected because of a general lack of sanitary control over this matter in eating establishments.

A detailed study (Table 6) of 100 eating establishments indicated that the greatest oral contamination of drinking glasses in these studied was found in tap rooms and bars. In the 100 establishments of this type which were examined, only 4% were found in which glasses were free of streptococci. In 63% of the tap rooms and bars, indicated by the Streptococcus salivarius test, the glasses were found to have been previously subjected to oral contamination without subsequent washing or cleaning. Soda fountains were found to be the most acceptable source of oral contamination concerned, as 30% of this type of establishment had glasses available upon the rims of which no organism could be found.

In the survey of 100 eating establishments it is interesting to note that roughly one-half of these establishments had drinking glasses available for public use which showed evidences of oral contamination not removed by subsequent washing or sterilizing.

The presence of micrococci and Gram-negative rods on a large percentage of drinking glasses in public eating establishments no doubt indicates contamination other than that of an oral nature. If the condition of drinking glasses is to be dependent entirely upon oral contamination, neither the presence of staphylococci (micrococci) nor Gram-negative rods will serve as an index. Micrococci may indicate excessive handling without subsequent washing and the same may be true of the presence of Gram-negative rods, although in such instances indications are that the presence of these organisms may be an indirect index of unsanitary contamination.

CONCLUSIONS

1. A study of 100 individuals indicates that a streptococcus identified as Streptococcus salivarius could be recovered from the closed lips of all persons tested.

2. A study of 100 controlled cases indicated that without exception Streptococcus salivarius is deposited on the rims of glasses during use.

3. When not cleaned or sterilized, Streptococcus salivarius will survive on the rims of drinking glasses for at least 48 hours following use.

4. A cold tap water rinse following use will not remove Streptococcus salivarius from the rims of glasses.

5. The use of a double rinse of milk with good quality raw milk approximated to a plate count of 140,000,000 per gm., while the methylene blue reduction test figures varied from 3 to 14 hours. The mean figures calculated as reconstructed milk approximated to a plate count of 400,000 per ml. and compared unfavorably with good quality raw milk. Testing 1 ml. quantities of spray-dried milk showed that of raw milk about 10 per cent gave positive results to the presumptive cohn test. However, a material proportion was due to the growth of anaerobic spore-formers and not to cohn test strains.

6. A soap water wash (pH 8.6) at 120°F. following by rinsing for five minutes at 165°F. in clear water will remove traces of oral contamination provided the deposition made on the glasses during the act of drinking is not too great.

7. The presence of Streptococcus salivarius on the rims of drinking glasses is a presumptive index which indicates previous oral contamination of these utensils. A procedure for making such a presumptive test is presented.

BIBLIOGRAPHY


7. The presence of Streptococcus salivarius on the rims of drinking glasses is a presumptive index which indicates previous oral contamination of these utensils. A procedure for making such a presumptive test is presented.
The Early Detection of Bovine Mastitis by an Electrometric Method

Ernest C. McCulloch
Division of Veterinary Science
Agricultural Experiment Station,
State College of Washington, Pullman, Washington

The detection of cows yielding abnormal milk presents a serious problem to the veterinary inspector whose duty it is to exclude all but "the lacrimal secretion obtained by the complete milking of one or more healthy cows." (1)

Examinations for udder health should have two definite objectives. The first must be to protect the consumer against receiving the products of unhealthy udders. Milk inspection which does not include this service cannot be worthy of security given.

The multiplicity of mastitis tests advocated by various research workers, the difficulties and uncertainties in the interpretation of these tests, together with the lack of agreement as to their accuracy, have led to such general confusion that some health officials feel that extensive mastitis control measures in their milk-sheds must await more research work and the presentation of more lucid data. As is the result of the unavailability of the milk which is used, the finding that mastitis is not detected by bacteriological methods.

Bacteriological Methods. Samples for bacteriological examination must be correctly drawn, and examinations made in a sanitary laboratory by trained personnel. This limits the method to very valuable herds or to research work. The mastitis streptococi do not grow well on ordinary media, and an enriched medium usually must be employed. Changes due to the action of a virus are not detected by bacteriological methods.

Horsit Test. In many herds the horsit test has given satisfactory results. The quarter samples are drawn into a solution of brom cresol purple and then incubated. Mastitis streptococi, if present, tend to agglutinate on the surface of the tube where they multiply to form yellow flakes which are readily visible on the purple background.

Indirect Tests. The so-called indirect tests for the detection of mastitis are in more general use. All are based upon the alteration in the milk which follow inflammatory changes in the udder. These changes are positive at the degree of acidity, with milk in which much controversy exists as to their value.

More accurate determinations of the degree of acidity or alkalinity can be made by means of a potentiometer. This, however, involves the drawing and labeling of quarter samples and taking them to a laboratory, which greatly adds to the labor and expense.

Rennet Clotting Time. Partly because of the decrease in acidity, mastitic milk clots much more rapidly. This test has not been extensively used as a method of testing individual quarter samples, because other methods are equally reliable and much more rapid. It is used, however, in some cheese factories on the mixed milk from individual herds.

Leucocyte Determination. Mastitic milk contains more leucocytes or white blood cells than does normal milk. Smears of quarter samples are made on glass
slides, properly stained, and examined under the microscope. Some have considered the presence of over 50,000 leucocytes per cubic centimeter as an indication of udder involvement. Others have set the figure at 100,000, 500,000 or even 1,000,000. The first figure probably is too low, and would result in normal cows being regarded as mastitic, while the last figure probably is so high as to allow many mastitic cows to go undetected.

Sediment. Mastitic milk yields more sediment when centrifugalized than does normal milk, probably due almost entirely to the greater number of leucocytes it contains.

Catalase. With an increase of leucocytes there is also an increase in the enzyme, catalase. This enzyme decomposes hydrogen peroxide with the liberation of oxygen gas. Field tests have been made with this method. A small amount of the milk sample and hydrogen peroxide solution are mixed on a piece of dark glass, and the intensity of the liberation of bubbles of oxygen observed. This test is so difficult to interpret quantitatively that it is but little used at the present time. More frequently, the milk sample is drawn into a test tube, hydrogen peroxide solution added, the tube filled to the top with water and stoppered with a one-hole stopper and then inverted. The oxygen gas liberated by the catalase collects in the upper portion of the inverted tube where the amount readily can be estimated. This method has given variable results. Some investigators have found it very sensitive in detecting obscure infections. Others have found it to give many false positives. Milk from normal udders shortly after parturition and near the end of lactation frequently gives positive results.

Chloride Content. Mastitic milk almost always is high in chlorides. The chloride content of milk may be measured by titration with silver nitrate solutions in the presence of potassium chromate or other indicators such as dichlorofluorescein.

This method is the most widely used of the indirect tests and has given satisfactory results, although it is known that the chloride content of milk from normal udders is very high during the first week following parturition. It then falls, only to rise gradually after the third month, normally being rather high after the eighth or ninth month in milk. Due to the absorption of some of the silver nitrate by milk constituents other than chlorides and the masking of the color changes by the opacity of the milk, the results given are high, usually by approximately 0.025 percent. Field modifications of this test also have been developed. They indicate whether the chloride content is above or below a predetermined amount.

Electrical Conductivity. Mastitic milk conducts electrical currents more readily than does normal milk. Direct current measurements of the electrical conductivity of milk are not practical because of the rapidity with which milk breaks down with the passage of a continuous electrical current. Until the present time, measurements with alternating current have not been used extensively for the detection of abnormal milk because samples had to be drawn, taken to a laboratory, remixed to distribute the cream, brought to a constant temperature, and then the conductivity determined by means of rather expensive equipment.

A Portable Electrometric Device. In order to provide a rapid, convenient, and reasonably accurate method for obtaining quantitative measurements of the degree of abnormality of milk, without the necessity of drawing samples into test tubes which must then be labeled, taken to a laboratory and the results known only at a later time, the writer has developed a portable electrometric mastitis detector (4). This is shown in Figure 1. Essentially, this consists of a Wheatstone's bridge in which the milk cell forms one arm. The milk cell of the present model consists of a one-half inch hole in a block of the thermoplastic, "Flexiglas." The upper portion of the cell is enlarged
to facilitate the drawing of milk into it. Two nickel electrodes pass through the lower portion of the milk cell. The other arms of the bridge consist respectively of a 200, a 200 and a 400 ohm resistor with a 200 ohm potentiometer. A diagram of the electrical circuit is shown in Figure II. The source of the alternating electrical current is an audio oscillator, which uses six "Penlight" cells as "B" batteries and one flashlight cell as an "A" battery. The drain on the "B" batteries is almost negligible and the drain on the "A" battery is approximately one-fifth that of a flashlight. These batteries can be purchased for a few cents at any hardware or drug store. Radio earphones are worn to detect the current.

Testing by means of this device is very rapid. The operator dons the earphones,
Early Detection of Mastitis

**Figure 2.** Diagram of electric circuit of electrometric device.

Quarter samples can be made on 30 cows in an hour without undue haste. Unless the previous reading was unduly high, there is no necessity for rinsing the milk cell. If rinsing is necessary, a single stream from the next quarter to be tested suffices. Bacteriological sterility is not necessary.

Since the results are known immediately while the examining veterinarian is seated at the cow, opportunity is given for confirmation of high readings by physical examination. Samples of milk can also be drawn for such other tests as may seem advisable. Since thorough physical examination and other tests need be made only on those quarters showing high conductivity, the examination of the herd can be made in much less time. Also, the inspector is in a position to supervise the isolation or removal of the animal and to suggest such sanitary measures as he may deem necessary.

The accuracy of the device, as compared with the estimation of the chloride content of the milk by means of a silver nitrate solution in the presence of potassium chromate as an indicator, is shown in Figure III. Those samples, showing high chloride content in relation to the electrical conductivity, are, for the most part, from cows just fresh or in the latter stages of lactation. To date, approximately 5,000 quarter samples have been compared.

Like all of the previously described methods, high readings are obtained on milk from normal udders immediately following parturition and during the latter stages of lactation. A correction factor for this is provided on the dial.

In order to prevent unethical exploitation of this method, patent rights have been applied for and assigned to the Research Foundation of the State College of Washington.

The writer wishes to express appreciation to the National Youth Administration for student help in collecting samples.

**REFERENCES**

A Modified Resazurin Test for the More Accurate Estimation of Milk Quality *

C. K. Johns and R. K. Howson

Dominion Department of Agriculture, Ottawa, Canada

Among its other attributes, good milk should be the product of healthy udders and relatively free from contaminating bacteria. While the desirability of a minimum bacterial content is generally accepted, less attention has hitherto been given to the udder condition. It is well known that many herds contain cows with udders seriously affected by mastitis. Milk from such udders is generally changed in composition and contains large numbers of leucocytes. Studies on the leucocyte content of normal milk have shown (3, 6, 15) that this rarely exceeds 100,000 per ml., while counts in excess of 500,000 per ml. indicate that the milk has come from an udder containing an appreciable amount of fibrosis (6). The leucocyte content, as indicated by either the direct microscopic count (1) or the catalase test (6, 8, 18), therefore furnishes a useful indication of the degree of udder abnormality in a herd. Where this is high, the milk can scarcely be regarded as an entirely acceptable product for human consumption.

If a routine test were available which in addition to reflecting the bacterial content also placed these “abnormal” milks in a lower grade, there would be a stronger stimulus to the dairyman to eliminate advanced cases of mastitis. While the direct microscopic count (1) is very useful here, many who have had experience with it prefer to use one of the simpler dye reduction tests for routine grading, especially where large numbers of samples must be examined. The widely used method, the methylene blue (M. B.) reduction test (1), is of little value in detecting mastitis when applied to herd milks (13). The newer resazurin “one-hour” test is more sensitive to both physiologically and pathologically abnormal milks (2, 5, 11, 16, 17). Nevertheless, a fair proportion of milks with high leucocyte counts fails to cause an appreciable color change within one hour (10, 16), while high counts of dormant bacteria frequently escape detection (10).

The accuracy of the resazurin test as an index of bacterial numbers is considerably improved by continuing incubation to the pink end-point (10). However, the resazurin “pink” test is less effective than the “one-hour” test in the detection of “abnormal” milks. Such milks usually show considerable color change during the first few hours, after which the rate of change is so slow that, in the absence of sufficient bacterial activity, the pink stage may not be reached within the time limit adopted. The following data (Table 1) illustrate the difference between color changes with such milks and with those where changes are mainly due to bacterial activity.

Our studies, supplemented by plant experience, indicate that the greatest amount of information is obtainable where the number of samples is recorded hourly. This, however, is impracticable in the routine testing of large numbers of samples. We have therefore subjected our data on 275 market milks to additional careful study to determine whether it would be possible to develop a simpler method for routine grading which would combine the sensitivity of the “one-hour” test for “abnormal” milks with the greater accuracy of the “pink end-point” for bacteria.

Table 2 shows the distribution of samples on the basis of color number after 1, 2 and 3 hours. The samples being grouped according to bacteria and leucocyte counts. It will be observed that within each bacterial group the higher leucocyte counts are reflected in a higher color number. The number of high-count samples showing little change after one hour should also be noted. As the incubation period is lengthened, the proportion of such discrepancies decreases until the third hour these are relatively few. The value of further observations beyond the first hour has been recognized by Ramsdell et al. (16) and by Keenan (12).

Just where the leucocyte count limit for herd milk should be set is a problem with which we are not particularly concerned. The important thing is that the degree of color change with resazurin, especially by the third hour, is closely related to the leucocyte count. If a color number not greater than 8 at 3 hours is taken as the limit for a first grade milk, it will be seen from Table 2 that the great majority of milks with high leucocyte counts fails to meet this standard. This is true even where the bacteria count is low. The color limit indicated also reflects the bacteria count very satisfactorily.

These results suggested the possibility of using this same color reading at one, two, and three hours as a simple method of grading a selection of shippers' milks into four grades. In the proposed method, all samples showing a color number in excess of 8 at 1 hour would go into 4th grade; those showing a similar change by the 2nd hour would be 3rd grade; those doing the same by the 3rd hour would be 2nd grade; while the remainder would constitute the 1st grade. In passing, it should be noted that the “creaming error” is minimized by inverting the tubes after each hourly reading (9, 10).

To compare the grading by the proposed “triple reading” method with that by several other methods, we have prepared Table 3 in which are shown the percentages of samples in the various classes placed in the different grades by each method. For the modified methylene blue reduction test (10) we have taken 2, 4 and 6 hours as the class limits; for the resazurin “pink” test, 11/2, 3 and 4½ hours (10); and for the resazurin “one-hour” test, the color limits suggested by Ramsdell et al. (16) but expressed in terms of the color notation employed by us.

While with the low bacteria count milks the resazurin “one-hour” test is more sensitive to high leucocyte counts than are either the resazurin “pink” or M. B. tests, it is much less so than is the “triple reading” method. Furthermore, the “one-hour” test, using Ramsdell’s standards, places an unduly large percentage of high bacteria count milks in the first and second grades; they being practically no differentiation between bacteria counts below 2,000,000 per ml. The triple reading method reflects the bacterial and leucocyte contents of the milk much more satisfactorily than any of the other three methods.

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* Contribution No. 114 (Journal Series) from the Division of Bacteriology and Dairy Research, Science Service, Department of Agriculture, Ottawa.

† Color numbers range from 0 for initial color to 16 for pink end-point, and 24 for decolorized. Changes from the initial blue color are quantitatively related to the action of bacteria, or to that of leucocytes or reducing substances present in abnormal milk.

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>Breed count (thousands)</th>
<th>Bacteria, Leucocytes</th>
<th>Resazurin color no. after hours</th>
<th>Reduction time, Modified M.B.</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>64</td>
<td>1,100</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>2</td>
<td>413</td>
<td>169</td>
<td>6</td>
<td>9</td>
</tr>
<tr>
<td>3</td>
<td>514</td>
<td>193</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>4</td>
<td>564</td>
<td>220</td>
<td>5</td>
<td>10</td>
</tr>
<tr>
<td>5</td>
<td>435</td>
<td>120</td>
<td>16</td>
<td>9</td>
</tr>
<tr>
<td>6</td>
<td>324</td>
<td>150</td>
<td>10</td>
<td>11</td>
</tr>
<tr>
<td>7</td>
<td>243</td>
<td>200</td>
<td>5</td>
<td>12</td>
</tr>
<tr>
<td>8</td>
<td>123</td>
<td>250</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>9</td>
<td>94</td>
<td>180</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>10</td>
<td>60</td>
<td>130</td>
<td>1</td>
<td>4</td>
</tr>
</tbody>
</table>

---

1 For full yearly and 24 for decolorized. Changes from the initial blue color are quantitatively related to the action of bacteria, or to that of leucocytes or reducing substances present in abnormal milk.
DISCUSSION

The presence of excessive numbers of leucocytes in milk is definitely indicative of abnormal udder conditions. Such milk can scarcely be regarded as being on a par with that containing small numbers. A routine testing method which grades milk on the basis of leucocyte, as well as bacterial content furnishes a more comprehensive index of the true quality of the milk. As a supplementary test for determining whether or not dye reduction is due to non-bacterial factors, the direct microscopic examination (1) has been recommended (2, 5, 12). In our experience, this information can be obtained much more simply and rapidly by determining the catalase content, using one of the simpler modifications (4, 8). We hope to report upon this in the near future.

The possible role played by the leucocytes themselves in the reduction of methylene blue in milk has been studied recently by Wilson (21) and Strynadka and Thornton (20). Both groups of workers showed that the addition of washed leucocytes had little effect on reduction time. The latter workers suggest that "the abnormal udder conditions responsible for milk of high leucocyte content are also responsible for abnormally high concentrations of reducing substances in the milk. The presence of reducing substances in abnormally high concentrations would explain the observations under discussion without soliciting aid from the leucocytes." Regardless of the correctness of this hypothesis, there is abundant evidence that these abnormal milks (especially quarter samples) do cause color changes with resazurin (and to a less extent with M. B.) out of all proportion to the bacterial content. Furthermore, potentiometric studies with such milks have shown that considerable color change with resazurin may occur with little or no concurrent change in redox potential, even when air is bubbled through the tube during the test (9). This phase is receiving further study.

Although the new method involves three readings at hourly intervals, this demands considerably less time and effort than is required with the resazurin "pink" and M. B. tests. The relative times required for the evaluation of first grade milks of equivalent quality are as follows:

- Resazurin "Triple Reading" Test: 3 hours
- Resazurin "Pink" Test: 4½ hours
- M. B. (modified): 6 hours
- M. B. (standard): 8 hours

The extra time and work involved in the "triple reading" test as compared with the "one-hour" test is more than compensated for by the improved accuracy, while a single color standard is also a definite advantage.

The use of the direct microscopic count as the yardstick in comparing the relative accuracies of the several reduction tests is admittedly open to criticism. Even though an attempt was made to improve the accuracy by counting 60-80 fields for bacteria and 40-60 fields for leucocytes, the experimental error, especially with low count milks, is distressingly large (19). Nevertheless, our experience has convinced us that this method presents a truer picture of the degree of bacterial contamination than can be obtained by the plate count. The quite close agreement between our results for the low bacteria and leucocyte count milks suggests that some, if not all, of the samples placed in second grade by all four methods actually contained many more bacteria than the Breed count indicated.

The color standard selected (No. 8) is described as P 15/4 according to the Munsell system of color notation (14). Uniform end-points in different laboratories may be assured by obtaining standard color sheets of this particular shade from the manufacturer. The provision of a daylight lamp also facilitates accurate color recognition. In our work we have used a 15-watt Mazda fluorescent daylight lamp (General Electric) with a neutral grey background. This has been found...
modified resazurin test for milk quality

superior to any other type of daylight lamp we have tried.

It may be objected that the grade standards selected in these studies are too high for use in some communities. [The same objection holds true in regard to the recently suggested standards for the M. B. reduction test (1).] In such cases the same color standard (No. 8) may be used and readings made at shorter intervals until the quality of the supply has improved to the point where the suggested standards can be introduced.

Summary

Using a single color standard (P 7/4 Munsell notation) approximately half way between the initial blue and the full pink color, and making hourly readings up to the third hour, milk may be graded simply and accurately by means of the resazurin test.

The "triple reading" test is superior to the "one-hour" resazurin test in the detection of high bacteria and leucocyte counts, and reflects high leucocyte counts much more definitely than do either the Munsell notation or M. B. reduction tests.

REFERENCES


18. Seelemand, M. Die Streptokokkeninfek­


Mr. Jennings Now in Health Work

The many friends of Mr. J. R. Jennings will be glad to know that he is back in the field of public health. He is now in charge of the supervision of milk sanitation for the Iowa State Department of Health at Des Moines. For two years he has been associated with one of the commercial companies which sold supplies in the public health field.

Mr. Jennings has been an active member of the International Association of Milk Sanitarians for many years. He has regularly attended the meetings of the Association, and has actively contributed to the advancement of milk sanitation. We are glad to see him back in the field of public health where he can devote his full time and capabilities to this field of public need.
Factors Affecting the Survival of Streptococcus pyogenes in Cheese*

M. W. Yale and J. C. Marquardt

New York State Agricultural Experiment Station, Geneva, New York

INTRODUCTION

Milk-borne outbreaks of septic sore throat and scarlet fever are most frequently traced to raw milk. Hucker and Marquardt (1) pointed out that inasmuch as a large percentage of cheese is made from raw milk, these milk sources are as liable to infection as are milk supplies that are subjected to little or no sanitary supervision. These milk sources are as liable to contamination with Streptococcus pyogenes as raw milk supplies that are used for fluid consumption. They were the first to study the possibility of cheddar cheese being incriminated in septic sore throat epidemics, presenting their work at the Schenectady meeting of the New York State Association of Dairy and Milk Inspectors in 1936.

These authors found in a brief study that Streptococcus pyogenes could live in cheddar cheese for more than 160 days when cured at 40°F. (4.4°C) and for more than 85 days when cured at 60°F. (15.5°C). In addition, an examination of cheese known to have been made at a factory from milk containing Streptococcus pyogenes showed that this organism was present in the cheese.

Information on cheese-borne epidemics is meager. Swammer (2) summarized 31 outbreaks of septic sore throat and scarlet fever in Europe in the literature which occurred throughout the world between 1883 and 1939. While none of these are reported as due to Streptococcus pyogenes, these milk sources are as liable to infection as are milk supplies that are subjected to little or no sanitary supervision. These milk sources are as liable to contamination with Streptococcus pyogenes as raw milk supplies that are used for fluid consumption. They were the first to study the possibility of cheddar cheese being incriminated in septic sore throat epidemics, presenting their work at the Schenectady meeting of the New York State Association of Dairy and Milk Inspectors in 1936.

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Table 1

Survival of Streptococcus pyogenes in Cottage Cheese made with 1, 5 and 10 percent Lactic Starter

<table>
<thead>
<tr>
<th>Sample Source</th>
<th>Hours after inoculation</th>
<th>Lot A 1% lactic starter</th>
<th>Lot B 5% lactic starter</th>
<th>Lot C 1% lactic starter</th>
<th>Lot D 10% lactic starter</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH</td>
<td>pH</td>
<td>pH</td>
<td>pH</td>
<td>pH</td>
</tr>
<tr>
<td>Uninoculated milk</td>
<td>&lt;10</td>
<td>6.60</td>
<td>&lt;10</td>
<td>6.60</td>
<td>&lt;10</td>
</tr>
<tr>
<td>S. pyogenes culture</td>
<td>0</td>
<td>86,000,000</td>
<td>5.18</td>
<td>86,000,000</td>
<td>5.18</td>
</tr>
<tr>
<td>Inoculated milk</td>
<td>1</td>
<td>950,000</td>
<td>6.20</td>
<td>1,840,000</td>
<td>6.10</td>
</tr>
<tr>
<td>Milk</td>
<td>3</td>
<td>1,200,000</td>
<td>6.17</td>
<td>1,030,000</td>
<td>5.23</td>
</tr>
<tr>
<td>Milk (A, C);</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Curred (B, D)</td>
<td>6</td>
<td>1,480,000</td>
<td>5.70</td>
<td>300,000</td>
<td>4.75</td>
</tr>
<tr>
<td>Re inoculated curred*</td>
<td>28</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Re inoculated curred*</td>
<td>48</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Fifty grams of the salted curd inoculated with 25 ml. of pasteurized whole milk containing 670,000 S. pyogenes colonies per ml.

---

Table 2

Survival of Lactic Streptococci in Ladle Milk

<table>
<thead>
<tr>
<th>Sample Source</th>
<th>High Moisture</th>
<th>Low Moisture</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH values</td>
<td>pH values</td>
</tr>
<tr>
<td></td>
<td>Colonies per gm</td>
<td>Colonies per gm</td>
</tr>
<tr>
<td>Cheese, 1 day</td>
<td>7.3</td>
<td>7.0</td>
</tr>
<tr>
<td>Cheese, 2 days</td>
<td>7.0</td>
<td>6.7</td>
</tr>
<tr>
<td>Cheese, 3 days</td>
<td>6.8</td>
<td>6.5</td>
</tr>
<tr>
<td>Cheese, 4 days</td>
<td>6.5</td>
<td>6.3</td>
</tr>
<tr>
<td>Cheese, 5 days</td>
<td>6.3</td>
<td>6.1</td>
</tr>
<tr>
<td>Cheese, 6 days</td>
<td>6.1</td>
<td>5.9</td>
</tr>
</tbody>
</table>

---

The survival of S. pyogenes in cottage cheese was investigated by inoculating the cheese with one percent of an S. pyogenes culture. The pH values were measured at the end of six hours and then every hour for six hours. The results indicated that S. pyogenes could survive for up to six hours in cottage cheese made from pasteurized milk. The cheese with a pH below 5.5 was found to be unsuitable for eating. The limiting hydrogen ion concentration for growth of S. pyogenes was found to be pH 5.5. The cheese made from raw milk had a higher moisture content than the cheese made from pasteurized milk, which may have contributed to the survival of S. pyogenes.
state. Control lots of cheese were made from equal amounts of un inoculated milk. The cheeses were held at 60°F (15.5°C) for 14 days for development of a smear, then wrapped and placed in storage at 50°F (10°C).

The inoculated milk in lot A contained more beta hemolytic organisms than that in lot B, 360,000 per ml. as compared to 300,000 per ml. This relation also held true for the curd at cheddaring. Beta hemolytic colony counts being 1,800,000 per gram in lot A compared to 1,500,000 per gram in lot B. The maximum number of beta hemolytic colonies in the cheese of lot A, 14,400,000 per gram, was obtained on the seventh day while the maximum number in lot B, 13,900,000 per gram, was obtained when the cheese was but one day old.

S. pyogenes survived considerably longer in the cheeses of lot A than in those of lot B, beta hemolytic colonies being recovered from lot A at 28 days but not at 51 days and from lot B at 9 days but not at 14 days. The more rapid increase in numbers in lot B than in lot A during the first 24 hours suggested that S. pyogenes might die off at an earlier age in lot B which proved to be the case. The hydrogen ion concentrations of the first 5/16th inch of the cheese beneath the surface and of the center portion were determined in lot B. Since in limburger and other surface-ripened types of cheese there is a more rapid shift in pH toward neutrality near the surface than at the center, there is a possibility that S. pyogenes might survive longer in one part of the cheese than in another. Comparative blood agar plate counts of outer and inner portions showed no significant differences in count. The pH values (Table 2) are in line with those reported for commercial limburger by Kelly and Marquardt (13), and dropped as low as pH 4.75 in the interior of the cheese. These values are not as low as those for cottage cheese (Table 1), which dropped to pH 4.32 in one instance.

Lot A contained 42.8 percent moisture and 1.7 percent salt, and lot B 49.3 percent moisture and 2.8 percent salt. These differences in composition probably afford the chief explanation for the longer survival time in lot A than in lot B. In the absence of lactic acid, S. pyogenes would die off more rapidly in the high moisture cheese with more lactose present to be converted into acid resulting in the limiting hydrogen ion concentration being attained more quickly.

Since the moisture content of commercial limburger falls within the above range of 43 to 49.3 percent and since pH values vary considerably in market cheese, S. pyogenes when present may survive considerably longer under some conditions than others. These experiments indicate that S. pyogenes will not survive longer than five or six weeks in limburger cheese. Since the great bulk of commercial limburger is 6 to 12 weeks of age when purchased by the consumer, it appears that the chance of septic sore throat being transmitted through limburger is negligible.

**Survival in Cheddar Cheese**

Only a small percentage of cheddar cheese is made from pasteurized milk in this country. While pasteurization of milk for cheddar cheese making is increasing each year, it is being accepted more slowly in the rest of the world. Much of the high-moisture, quick-curing type of cheddar cheese made from raw milk is placed on the market when only about five weeks of age.

Two lots of cheddar cheese were made from raw Station milk (Table 3). Lot A was made from 157 pounds of milk inoculated with one percent of a lactic starter and with one percent of the Miniature strain of S. pyogenes. Lot B was made from 55 pounds of raw Station milk inoculated with one percent of lactic starter and one percent of the Medina strain of S. pyogenes. Controls were made using equal amounts of milk. In the case of lot B, this control was made by a separate worker in a separate room to avoid any possibility of accidental inoculation with S. pyogenes. The cheeses were made as Young Americans and weighed about 5.5 pounds each. In addition, some of the cheese in Lot A was canned. Results obtained from the canned cheese (vented cans) are similar to those for the Young Americans and are not reported for this reason.

One of the Young Americans of lot A was cured at 45°F (7.2°C), the other at 62°F (16.6°C) while the Young America in lot B was held at 50°F (10°C). S. pyogenes survived for over 18 weeks in the cheddar cheese held at 45°F, but could not be detected in the cheese held at 62°F. At the end of 11 weeks, S. pyogenes died within 18 weeks in the cheese of lot B cured at 45°F. This indicates that the temperature of curing is an important factor affecting the length of survival of S. pyogenes in cheddar cheese. The maximum number of S. pyogenes in lot A was 560,000 per gram in the second day old cheese, while the maximum count in lot B of 2,000,000 per gram was obtained from the curd at cheddaring.

Hucke and Marquardt (1) found maximum counts at 160 days and 85 days in the case of two lots of cheddar cheese ripened at 40°F (4.4°C). Since S. pyogenes does not grow at this temperature (14, 15), it should be pointed out that apparent increases in counts were probably due to limitations of sampling and plating methods rather than to growth of the organisms.

The pH values for lot B were normal and did not drop below pH 5.0, which was obtained at three days. Although the hydrogen ion concentration increases more rapidly during the first few hours in cheddar cheese than in limburger, pH values do not drop as low as they do in the case of limburger and remain at pH 5.0 or above unless the cheese is abnormally high in acid.

**Confirmation as Pyogenes**

Broad zone beta hemolytic streptococci which do not belong to Lancefield’s group A are sometimes present in raw milk (16, 17). Hucker (18) found that streptococci of hemolytic streptococci which Avery and Cullen (8) isolated from cream cheese belonged to group D. Obviously, beta hemolytic colonies on blood agar plates prepared from experimental cheese need confirmation before acceptance as S. pyogenes.

The raw milk from the Station supply

---

**Table 3**

<table>
<thead>
<tr>
<th>Sample Source</th>
<th>45°F (7.2°C)</th>
<th>62°F (16.6°C)</th>
<th>50°F (10°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S. pyogenes</td>
<td>S. pyogenes</td>
<td>S. pyogenes</td>
</tr>
<tr>
<td>Uninoculated milk</td>
<td>&lt;10</td>
<td>&lt;10</td>
<td>&lt;10</td>
</tr>
<tr>
<td>Inoculated milk</td>
<td>152,000,000</td>
<td>152,000,000</td>
<td>12,500,000</td>
</tr>
<tr>
<td>Why at cheddaring</td>
<td>6,800</td>
<td>6,800</td>
<td>2,800</td>
</tr>
<tr>
<td>Cured at cheddaring</td>
<td>400,000</td>
<td>400,000</td>
<td>3,000,000</td>
</tr>
<tr>
<td>Cheese, 2 days</td>
<td>560,000</td>
<td>560,000</td>
<td>160,000</td>
</tr>
<tr>
<td>Cheese, 1 week</td>
<td>350,000</td>
<td>125,000</td>
<td>22,000</td>
</tr>
<tr>
<td>Cheese, 3 weeks</td>
<td>250,000</td>
<td>34,000</td>
<td>145,000</td>
</tr>
<tr>
<td>Cheese, 5 weeks</td>
<td>92,000</td>
<td>16,300</td>
<td>129,000</td>
</tr>
<tr>
<td>Cheese, 7 weeks</td>
<td>29,500</td>
<td>28,000</td>
<td>31,000</td>
</tr>
<tr>
<td>Cheese, 9 weeks</td>
<td>28,000</td>
<td>14,000</td>
<td>17,000</td>
</tr>
<tr>
<td>Cheese, 11 weeks</td>
<td>6,800</td>
<td>&lt;10</td>
<td>18,000</td>
</tr>
<tr>
<td>Cheese, 13 weeks</td>
<td>1,350</td>
<td>&lt;10</td>
<td>20,000</td>
</tr>
<tr>
<td>Cheese, 18 weeks</td>
<td>2,400</td>
<td>&lt;10</td>
<td>5,200</td>
</tr>
<tr>
<td>Cheese, 39 weeks</td>
<td>&lt;10</td>
<td>Not examined</td>
<td></td>
</tr>
</tbody>
</table>

*Moisture content, Lot A, 37.0 percent, Lot B, 35.0 percent. Salt content, Lot A, 1.7 percent, Lot B, 1.8 percent.
used in most of the experiments was free from broad zone beta hemolytic colonies in the 1:10 dilution and usually developed only a few narrow zone beta hemolytic colonies. The presence of broad zone beta hemolytic colonies on plates from cheese made from milk inoculated with S. pyogenes and absence of such colonies on plates from control cheese were accepted as presumptive evidence that the colonies were S. pyogenes. Several broad zone beta hemolytic colonies were picked from plates during the course of each experiment, purified by replating on blood agar, and picked again into veal infusion broth. If growth and morphology were typical of S. pyogenes, one culture was selected for Lancefield grouping with group A serum. Both the Lancefield (18) and Coffey (19) techniques employing this serum were used with satisfactory results.

In the case of the cottage cheese, confusing types of beta hemolytic colonies were absent, but this was not true of some of the experiments with limburger and cheddar cheese. Beta hemolytic colonies of gram negative rods were found on plates prepared from limburger cheese, lot B, at 28 and 35 days of age.

In the case of the cheddar cheese, the type of beta hemolytic colony most easily visible in cheddar cheese were grown from plates from cheese made from milk inoculated with S. pyogenes. Colonies on plates from cheese made from broad zone beta hemolytics in the milk, the limiting hydrogen ion concentration of the strain of S. pyogenes, the type of cheese, and the curing temperature. Cheese is subject to great variation in manufacturing methods and in composition so that it is reasonable to assume that there is considerable variation in the length of time during which S. pyogenes will survive in the cheeses of any given variety. Moreover, abnormal colonies of any variety are sometimes manufactured which do not show normal acid development. The length of survival of S. pyogenes in cheese of abnormal quality may be quite different from that in cheese of normal quality.

Not all cheese which is consumed passes through regular trade channels. Fresh curd is frequently eaten by cheese makers and there is a limited practice of selling curd only a few hours old to customers who bring their own container.

Young cheeses which may be only a few days old are regularly examined at factories by inspectors, buyers and cheese makers who taste the cheese as part of the scoring procedure.

There is also a limited amount of farm cheesemaking, where the raw milk used comes from single herds. If infection of the milk supply should occur, greater numbers of S. pyogenes would likely be present under farm than under factory conditions due to greater dilution in the latter case with milk from non-infected herds.

The important question whether Streptococcus pyogenes retains its virulence and pathogenic properties during its existence in cheese remains to be answered. Just as cultures of S. pyogenes lose virulence under laboratory conditions, so S. pyogenes organisms in cheese may gradually lose their virulence. This is likely in view of the unfavorable acid condition of many cheeses. Furthermore, no cheeseborne outbreaks of septic sore throat or scarlet fever are reported in the literature.

Cottage cheese and limburger cheese were made from raw milk appear to be safer from the standpoint of transmitting S. pyogenes than does cheddar cheese. S. pyogenes may survive in cheddar cheese as long as 18 weeks if the cheese is cured at a low temperature. This confirms results previously obtained by Hacker and Marquardt (1).

**DISCUSSION**

There appear to be many factors which influence the length of survival of Streptococcus pyogenes in cheese such as the original number of S. pyogenes organisms in the milk, the limiting hydrogen ion concentration of the strain of S. pyogenes, the type of cheese, and the curing temperature. Cheese is subject to great variation in manufacturing methods and in composition so that it is reasonable to assume that there is considerable variation in the length of time during which S. pyogenes will survive in the cheeses of any given variety. Moreover, abnormal colonies of any variety are sometimes manufactured which do not show normal acid development. The length of survival of S. pyogenes in cheese of abnormal quality may be quite different from that in cheese of normal quality.

**SUMMARY**

Streptococcus pyogenes Rosenbach was not recovered from a high-acid, non-genetic type of cottage cheese several hours after manufacture. Lots of cheeses were made using 1, 5, and 10 percent amounts of lactic starter. When the curd was inoculated, as might occur in the creaming of cottage cheese with raw cream, S. pyogenes again died off rapidly due to the high hydrogen ion concentration. Minimum pH values in the different lots of cottage cheese were about pH 4.5.

S. pyogenes survived for 28 and 51 days in one lot of limburger cheese with a moisture content of 42.8 percent and between 9 and 14 days in another lot with 49.3 percent moisture. The minimum pH value as determined for the second lot of limburger was pH 4.75.

S. pyogenes survived for over 18 weeks in cheddar cheese cured at 45°F (7.2°C) and between 9 and 11 weeks in a duplicate cheese cured at 62°F (16.6°C). In a second lot of cheddar cheese, S. pyogenes survived more than 18 weeks when the cheese was cured at 55°F (13°C). The pH values as determined for the second lot of cheddar cheese did not drop below pH 5.0.

The variety of cheese, its moisture and salt content and the curing temperature are some of the important factors affecting the length of survival of S. pyogenes in cheese.

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11. Cheese Reporter, 64, No. 8 (1939).


Application of the Resazurin Test in Determining the Quality of Pasteurized Cream

W. H. Chilson and M. A. Collins

INTRODUCTION

Resazurin has been used in quality tests by the United Farmers' Co-operative Creamery Association, Inc., laboratories for nearly three years and has proven very useful.

The results discussed in this paper were secured by the Quality Control Department of United Farmers' in its laboratory in Boston, Massachusetts.*

The data were obtained from the daily routine samples of pasteurized cream received by truck and rail from Vermont plants.

PURPOSE OF THE INVESTIGATION

The purpose of carrying on further work with resazurin dye as an indicator in quality tests with cream was:

(a) To compare the resazurin test with the incubation test and the standard plate count as a rapid method for determining the quality of pasteurized cream.

(b) To ascertain whether or not the resazurin test alone could be used as an index to (1) initial quality, and (2) keeping quality, in order to secure the information within a few hours following processing and before the cream is shipped, or to enable the city plants to quickly evaluate the quality of cream they receive from outside plants.

HISTORICAL

Although several investigators, both institutional and commercial, have studied resazurin in comparison to methylene blue, plate counts, and microscopic examinations on raw milk and cream, we are aware of only one report of work with pasteurized cream. This work was reported by Jenkins (2) at the Dairy Science Convention held at Columbus, Ohio, in June, 1938. Jenkins concluded that resazurin was very useful as an indicator in quality studies of pasteurized cream.

METHODS

(1) The strength of resazurin used was 0.005 percent aqueous solution as suggested by Moldavan (3), and used by Collins, White, Chilson, Turner, and Rice (1) in their work on the resazurin test as applied to raw milk and cream.

(2) The end point read was a uniformly pronounced pink color. The end point rather than a set time was the standard.

(3) The incubation test comprised (a) the initial acidity (made with a 9 ml. sample of cream diluted by means of 9 ml. of water and titrated against N/10 NaOH using 5 to 6 drops of phenolphthalein as an indicator) when the fresh cream reached the city, (b) incubation at a constant temperature of 72° F. for 15 hours, after which time the second acid titration was made. A rise of 1/100 percent in acid is discussed as a one-point rise.

(4) The plate count was made with tryptone-glucose-skim milk agar, according to Standard Methods of Milk Analysis, the samples being incubated at a temperature of 35-37° C. instead of 32° C. as is now the practice in Boston. The detailed procedure of the resazurin test was as follows:

Ten milliliter quantities of cream were placed in sterile test tubes and 1 ml. of 0.005 percent resazurin solution added. The tubes were then tempered to approximately 98° F. The resazurin dye solution was made up weekly and stored in brown glass bottles. The samples were prepared and read in a medium light, but away from direct or reflected sunlight. Observations for color changes were made at fifteen minute intervals. If the reduction appeared to be at the pink end point but the color had developed irregularly from top to bottom, the tube was inverted once to bring the entire contents to a uniform color before recording the final reading.

RESULTS

Initial Quality of Fresh Pasteurized Cream

The first studies were made on 20 or 40 percent cream pasteurized during the months of June, July, and August, 1939. These shipments were at least 36 hours old when they were sampled in the plant at Boston where all the tests were made. The results secured are recorded in Table 1, using the resazurin reduction time as the standard for comparison. Routine work in United Farmers' laboratories for several months prior to June, 1939, had indicated that pasteurized cream which would not reduce 0.005 percent resazurin to pronounced pink within six hours had exceptionally fine keeping quality. An acid rise of 0.02 percent has been accepted as appreciable and one which can be read accurately. A plate count not exceeding 40,000 bacteria per ml. has been accepted as the top standard because it is the current maximum set by the Boston Board of Health for pasteurized cream.

The data in Table 1 shows that 122 of the 179 samples had resazurin tests of five or more hours and 111 of these gave not more than 0.02 percent acid increase in incubation. Also 86 of the 122 samples had plate counts not exceeding 40,000 bacteria per ml. There are sufficient samples in both A and B to give a fairly true picture and since there was little difference in the relationship of resazurin tests to incubation tests or plate counts between the two groups, we may conclude that the cream in Group B was almost as good as that in Group A and that a five hour resazurin-pink test indicates a good quality cream and one which will conform to the new Boston plate count standards at least seven times in ten. Twenty-two samples of the 122 had plate counts above 40,000 but under 100,000 which, until January 1st, 1940, was the Boston Board Department maximum. In Group C, we find a closer correlation between the resazurin time and the plate count than between the resazurin time and the incubation test. Additional results taken on winter shipments are recorded in Table 2.

The data in Table 2 show that practically all the winter samples fell into Group A having resazurin tests above six hours. However, it is significant that in this table also, all the samples in group B (5-5% hrs.) showed a marked

* Paper completed in June, 1940.

Table 1

<table>
<thead>
<tr>
<th>Reduction Number</th>
<th>Acid Rise During Incubation</th>
<th>Standard Plate Counts</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time Of Resazurin</td>
<td>No. %</td>
<td>No. %</td>
</tr>
<tr>
<td>A-6 hrs. or more</td>
<td>40,000</td>
<td>41,000</td>
</tr>
<tr>
<td>B-3 to 5% hrs.</td>
<td>46</td>
<td>42</td>
</tr>
<tr>
<td>C-3 to 4% hrs.</td>
<td>76</td>
<td>69</td>
</tr>
<tr>
<td>Total</td>
<td>179</td>
<td></td>
</tr>
</tbody>
</table>
correlation with those in Group A (6 hours or over). More than 80 percent of the 450 samples compiled with the Boston Board of Health standard of 40,000 or less bacteria per ml. Seventy-eight samples which had counts above 40,000 came within the 40,000-100,000 class, while only 11 of the 450 samples had plate counts above 100,000 per ml. The results in Table 2 lend support to the statement regarding Table 1 that a five hour or longer resazurin test on pasteurized cream indicates good initial quality and the ability to remain sweet through the fifteen hour incubation period at 72° F.

Summarizing the results recorded in Tables 1 and 2, we note that of a total of 572 samples, which had resazurin tests of five hours or more, 451 (78.8 percent) gave less than 2 points (0.02 percent) acid rise on the incubation test and 547 (95.6 percent) complied with the Boston Board of Health standards (100,000 or less bacteria per ml.) which were in effect for pasteurized cream when the samples were studied. Four hundred and forty-seven of the 572 samples (78.1 percent) conformed with the new Boston Board of Health standards (40,000 or less bacteria per ml.) which became effective early in 1940. From the foregoing results shown in Tables 1 and 2, it would seem justifiable to conclude that a reduction time to pronounced pink of 0.005 percent resazurin solution in five hours or longer is a satisfactory criterion of good quality pasteurized cream.

Initial and Keeping Quality of Pasteurized Cream

The relationship of the resazurin test to the incubation test and standard plate count on pasteurized cream stored at 40-45° F. for about four days was studied with 73 samples of 20 or 40 percent cream, first sampled at from 36 to 48 hours and on two successive days. Large samples were taken from the 40-quart cans when they arrived in the city, and these were held in a refrigerator regulated between 40 and 45° F. All the samples were from 84 to 96 hours old when the last tests were made. This meant that the cream was at least 100 hours old at the end of the third incubation test.

The samples were divided into two classes based on the results of the resazurin tests recorded on the incubation tests, and these two divisions in turn were divided on the basis of the resazurin reduction time, with 6 hours again being chosen as the standard. The classification is summarized in Table 3.

Attention is drawn to the fact that 69 of 75 samples (92 percent) failed to reduce resazurin to pink in less than 6 hours and of these 69 samples, 47 or 62.6 percent of the total number, showed plate counts of 40,000 or less through the fourth day.

Summarizing the results secured on the 69 samples in more detail, there were 207 plate counts made during the three days. Fifty-seven of the 69 initial counts (36-48 hour old cream) were 40,000 or less: fifty-seven of the counts made on the 60-72 hours old were 40,000 or less, and fifty-two samples conformed to the standards at ages of 84 to 96 hours. That is, 52 of 69 samples (77.9 percent) aged four days conformed to the latest standard of the Board of Health. All of these samples had resazurin tests above six hours. A summary of the 22 samples which had plate counts above 40,000 is included at the bottom of Table 3 and the results of tests on these samples are discussed in the following paragraphs.

The data on the 22 samples indicate clearly that only seven of these samples showed an appreciable increase in count between the 2nd and 4th days (between the first and third tests). In five of these seven samples the resazurin reduction time was lowered sufficiently to indicate a definite change in quality. On the contrary, with two samples the change would not have been predicted from the resazurin tests. Six samples had relatively high counts, ranging from initial counts of 80,000 to 120,000 and final counts of 72,000 to 120,000 bacteria per ml. through the three days. None of these samples showed appreciable increases between the first and third tests, while some of the counts were reduced. Microscopic examinations of these samples revealed appreciable numbers of thermophilic organisms. Also, it is significant that only one sample did the resazurin show an appreciable reduction. It would seem reasonable to conclude that in view of the long reduction time of resazurin and the relatively unimportant changes from the initial count during the 48 hour storage period, the types of bacteria constituting the high counts did not possess strong reducing powers and probably were types which would not increase in number significantly at a low temperature, although they were largely responsible for the high counts at the 37° C. incubation temperature.

In order to secure more information about the relationship of resazurin reduction times to both incubation tests and standard plate counts, it was decided to hold samples of pasteurized 20 or 40 percent cream at rather high cold storage temperatures (45-50°F.) until incubation tests on these samples showed an acid increase over the initial titration of more than two points (0.02 percent). The final resazurin tests and plate counts recorded on the samples which, on the corresponding incubation tests, had shown more than two points acid rise. The results from the examination of these samples are tabulated in Table 4.

The data in Table 4 show that only 3
of 19 samples gave more than two point acid rise on incubation until the end of the fourth day. At the corresponding ages, 10 of the remaining 16 samples showed appreciable drops in reduction times of resazurin while 11 of the 16 samples appeared definitely inferior on the basis of the plate count. These results correspond with the agreements shown in the last column of the table. This column gives our conclusion as to which tests agree most closely in picking out definite quality deterioration. Only in sample 10 did it appear that the seven point acid rise on the fourth day predicted more truly the high count than did the 51/2 hour resazurin test and agreed more closely with the plate counts. Samples 1, 3, 12, 15, 17 and 18 are the best examples of close agreement between the resazurin test and the plate count.

Considering again the relationship between five or six hour resazurin tests and corresponding plate counts, it would appear from samples 13 and 14 that when cream is stored below 40° F., the resazurin may be less sensitive to the deterioration in quality than is the plate count. More samples with six hour resazurin tests had plate counts over 40,000 than in previous tables. This can be explained by the 32° C. incubation temperature which is known definitely to increase the plate count over the 37° C. temperature.

We may conclude from the foregoing discussion on Table 4 that there is a closer correlation between the time of reduction of resazurin to pink and the standard plate count than there is between the acid rise on samples incubated at 72° F. for 15 hours and the standard plate count.

DISCUSSION OF RESULTS

It is readily understood why creamery organizations in Vermont which are processing cream for the large urban centers of population such as Boston, Worcester, Providence, etc., should seek a quality test which will give reliable information within a few hours after pasteurization. We believe that the resazurin test as discussed in this paper fulfills this purpose. Cream pasteurized in the Vermont plants of the United Farmers' Co-operative Creamery Association, and no doubt the same applies to that processed in other commercial plants, is aged at approximately 40° F. for several hours before shipment. Even a six hour holding period in country plants enables the laboratory to send the resazurin results with the shipment to the city customers or distributors.

Any batches of cream which are not sold immediately upon arrival in the city are sampled at once and again checked by the resazurin test, incubation test, and plate count. Since most of the cream is distributed the first day after arrival, it is not possible to wait for an incubation test or plate count. Here again the resazurin test gives very valuable information in as little as three hours and dependable information within six hours.

The results in the first four tables were secured before the Boston Board of Health recommended that 32° C. be used as the standard incubation temperature for all plate count work. However, the tryptone-glucose-skimmilk agar was used throughout. The plate counts recorded in Table 4 were secured at an incubation temperature of 32° C.

It should be mentioned also that the plate counts recorded in Tables 1 to 3, inclusive, were made while the Boston Board of Health Standard for pasteurized cream was less than 100,000 bacteria per ml. Nevertheless the plate counts were classified and discussed from the standpoint of the new regulations (40,000 or less) which became effective January 1, 1940.

There was some question as to what acid rise should be considered the standard. In this work we have called a two point (0.02 percent) acid rise significant although it is questionable if this much increase during a 15 hour incubation test at 72° F. is sufficient to predict poor keeping quality. Although the results have shown that a resazurin reduction time to pronounced pink of more
than five hours indicates good quality cream, we feel that it would be wise for commercial plants to accept six hours as the minimum standard for "top grade" quality, especially for 20.0 percent pasteurized cream.

**SUMMARY AND CONCLUSIONS**

1. A six hour reduction time of 0.005 percent resazurin dye to a pronounced pink color is a criterion of fine quality in pasteurized 20 or 40 percent cream.

Such cream will generally show not more than 2 points (0.02 percent) acid rise upon incubation and standard plate counts not exceeding 40,000 bacteria per ml. when aged four days at 40-45° F.

2. A five hour resazurin test indicates quality of pasteurized 20 percent cream than is a six hour reduction time of 0.02 percent resazurin used on pasteurized 20 or 40 percent cream. However, it cannot be depended upon always to pick out high counts in cream that has been stored at 40° F. or lower for four or more days.

3. A reduction time of five hours for 0.005 percent resazurin to pronounced pink is a more accurate criterion of satisfactory quality in pasteurized 20 or 40 percent cream than is a 0.02 percent acid rise upon incubation at 72° F. for 15 hours.

4. A reduction time of over five hours for 0.005 percent resazurin used on pasteurized 20 or 40 percent cream indicates a standard plate count of less than 100,000 bacteria per ml. in 9 of 10 samples and of 40,000 or less in at least 7 of 10 cases.

5. When the pasteurized cream was stored for several days at less than 40° F., the resazurin reduction time did not indicate fully the increase in plate counts which occurred.

6. There was a much closer agreement between the resazurin test and the standard plate count than between the incubation test and the standard plate count in the indication of poor quality.

7. The resazurin test as applied in the work reported herein is a satisfactory and rapid criterion of both initial and keeping quality of pasteurized 20 or 40 percent cream. However, it cannot be depended upon always to pick out high counts in cream that has been stored at 40° F. or lower for four or more days.

**REFERENCES**


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**The Leucocyte Count and the Chloride Content of Milk from Bovine Udders with Mild Streptococcic Infections**

J. Frank Cone

Bureau of Dairy Industry, United States Department of Agriculture, Washington, D.C.

**INTRODUCTION**

The general practice, in using the chloride content and the leucocyte count of milk as indicators of mastitis, is to establish some arbitrary level for each test as a standard for interpreting the results. The standards, as used by different investigators, have varied widely. In a brief review of the literature, Little (4) pointed out that one group of investigators recommended as a criterion of mastitis a leucocyte count of 3,000,000 per ml. or more, whereas another group of investigators considered a count of 100,000 per ml. as indicating mastitis. In the case of direct titration values for chlorides, the usual criterion employed is 0.14.

Little (4) reports an experiment in which 95 percent of 1,010 quarter samples of foremilk from 8 young cows contained fewer than 300,000 leucocytes per ml., and 99.3 percent of the 1,010 samples contained less than 0.135 percent of chlorides. The 8 cows in the experiment were then exposed to or inoculated with a hemolytic streptococcus. Of 2,165 samples from quarters that developed subclinical mastitis, 94.5 percent contained leucocytes in excess of 300,000 per ml., but only 34.2 percent gave chloride titration values greater than 0.14 percent. Similar though generally less clear-cut results have been obtained in this laboratory and by other investigators.

It is generally recognized that the normal chloride content and leucocyte count of milk from different cows may differ widely. Some of the factors influencing these values are breed, age, and stage of lactation of the cow. These wide normal variations probably account for the poor definition of results that are obtained when one is dealing with a mixed herd of all ages and in various stages of lactation. In the mastitis investigations in the Bureau of Dairy Industry herd at Beltsville, Maryland, such a problem exists. In this work individual quarter samples are taken at intervals of two or three weeks. A comparison of the results from each quarter sample with the previous history of the quarter and also with results from other quarters of the same cow on the same day has proved to be of more value than arbitrary standards. The purpose of this paper is to show the results of such a comparison.

**METHODS**

Strip-cup tests are made daily on all of the cows. Samples for laboratory examination are collected at intervals of two or three weeks. Sampling is done in the afternoon just prior to the milking of each cow. Each test is wiped off with a pledget of cotton saturated with alcohol. After the first two streams of milk are discarded, about one-fourth of one pint is milked into a sterile, half-pint, screw-cap jar. These jars are collected in insulated shipping cases and iced for transport to the laboratory, where the samples are examined on the following day.

The examination consists of the Hotte test, as modified by Cone and Grant (1); plating 0.1 ml. in Edwards' (2) ascumin blood agar; leucocyte counts by direct microscopic examination of Breed smears stained with Newman's (3) stain; and...
direct titration for chlorides with silver nitrate according to the method of Hammer and Bailey (3).

RESULTS BEFORE AND AFTER INFECTION

Eleven cows have developed subclinical streptococcal infections in 22 quarters while under observation. For 12 of these quarters complete or partial histories are available for the lactation period prior to the one in which infection occurred. Eleven quarters were found to be infected at the time of the first sampling after calving. The other 11 quarters developed infections several weeks after calving and after at least 2 samples had been taken in that lactation period. The data obtained before and after the beginning of infection are shown in Table 1.

With 2 exceptions, the average of the leucocyte counts obtained after the onset of the infection is significantly higher than the average of those obtained before infection. The four samples from the left rear quarter of cow 1235 taken before the infection started were obtained at the end of a lactation period. At that time all quarters of the cow were giving milk high in chloride and leucocyte content. Since no infection occurred in the other 3 quarters in the next lactation period, concurrent figures for those quarters do not appear in Table 1. In the case of the left front quarter of cow 1469, the first 2 of the 6 samples taken after the beginning of the streptococcus infection gave leucocyte counts that were inexplicably high — 22,500,000 and 2,600,000 per ml, respectively. In this case, however, the corresponding chloride values were low, and the average leucocyte count of the last 3 samples before the beginning of infection was significantly lower than the average for the infected samples.

In general the average chloride content of the samples taken before the onset of infection is lower than the average for the samples taken after the beginning of infection. There are some exceptions, however, and in addition the case of the leucocyte counts. This probably is due to the greater tendency for the chloride content to change as the result of factors other than infection.

When infection takes place the rise in the chloride and leucocyte values is usually rather abrupt. This is shown when the results of the last 3 samples before infection are contrasted with the results from the first 3 samples after the infection has become established. The average of 3 samples rather than the results of a single sample has been used in each case in this comparison to rule out possible aberrant results that occasionally occur with a single sample. In the eleven comparisons shown, there was always a rise, usually marked, in the leucocyte count. With one exception the chloride values also increased at the beginning of infection.

COMPARISON OF RESULTS FROM SAMPLES TAKEN AT THE SAME TIME FROM INFECTED AND UNINFECTED QUARTERS

In a comparison of the uninfected quarters with the quarters of the same cow that were infected with streptococci, it seemed advisable to eliminate from the study all samples that were taken in acute stages of the disease. No sample from any quarter that showed positive strip-cup reactions within one week of the sampling date was included. Furthermore, all samples having a leucocyte count of 10,000,000 or more per ml were arbitrarily excluded. A total of 954 samples from 19 cows are included in this study. The results are shown in Table 2.

Without exception, the average leucocyte count of the samples from infected quarters was higher than that from uninfected quarters of the same cow. Seventeen cows showed a higher average chloride content in samples from infected quarters than in those from the uninfected quarters. In the other 2 cases the chloride values in the uninfected quarters were higher.

The 19 cows listed in Table 2 were sampled a total of 198 times. With each sampling the results of the infected
of uninfected quarters. In all but 12 of these 198 comparisons, the counts were higher in the infected quarters than in the uninfected quarters. Of 11 of the 12 aberrant cases, the leucocyte counts of the infected quarters were less than 11,000 per ml. In the other case, the chloride count was 19,600,000.

In all but 12 tests considered in this study, the average chloride values of the uninfected samples were 0.138 or higher. Thus in almost every instance in which this sort of comparison failed to reveal the infected quarters, the commonly employed arbitrary criteria based on the chloride titration and the leucocyte count would also have failed to distinguish between infected and uninfected quarters.

**DISCUSSION**

Most of the infections considered in this study are obviously very mild. A large number of them yielded average chloride values lower than the usually accepted standard of 0.14 percent. That is true of 14 of the 22 infected quarters listed in Table 2, and of 13 of the 22 infected quarters listed in Table 1. In almost every instance in which this sort of comparison failed to reveal the infected quarters, the commonly employed arbitrary criteria based on the chloride titration and the leucocyte count would also have failed to distinguish between infected and uninfected quarters.

On the other hand, a large number of the uninfected samples would have been included with the mastitis group if they had been considered in comparison with the arbitrary standards commonly employed. Three of the 22 uninfected quarters listed in Table 1 showed average chloride values greater than 0.14 percent and two others gave values greater than 0.15 percent. The samples from uninfected quarters of 5 of the cows listed in Table 2 gave average chloride titration values greater than 0.14 percent and the uninfected samples of two other cows gave average chloride values greater than 0.15 percent.

The average leucocyte counts of uninfected samples from 8 of the 22 quarters listed in Table 1, and the average counts for the uninfected quarters of 3 of the 19 cows listed in Table 2, were greater than 500,000 per ml.

The results show clearly that in an unselected group of cows, such as the one reported here, no arbitrary values can be selected for the chloride and leucocyte tests that will distinguish with any degree of reliability between samples from infected and those from uninfected quarters. Comparisons of the results from any cow with the previous history of that cow, or with the results from other quarters of the same cow, usually give a fairly reliable clue to the condition of the quarter. But neither of these indirect tests is as reliable as an index of infection as bacteriological tests. When used in conjunction with bacteriological tests, they add supporting information.

7. It can fairly be said that such mild infections have no significance, especially where an effort is being made to eliminate streptococcal mastitis by segregation of the infected animals. These mild cases, if left in an otherwise clean herd, might serve as foci for spreading the infection to other cows. It is possible, too, that the long continued, though slight, inflammation will adversely affect the production of the involved quarter.

**CONCLUSIONS**

1. In a group of cows of different breeds of various ages and in all stages of lactation, the chloride content and the leucocyte count of the milk failed to distinguish reliably between quarters with very mild streptococcal infections and the uninfected quarters.

2. In conjunction with bacteriological tests a comparison of the chloride content and leucocyte count of the samples from different quarters of the same cow gave valuable information in support of the culture methods.

3. When a cow is being tested periodically, an abrupt rise in the chloride content and leucocyte count in the milk strongly indicates the beginning of infection, and these values may not exceed the arbitrary values usually accepted as indicating mastitis.

4. The leucocyte count is a more reliable index of infection than the direct chloride titration values, probably because of the greater tendency for the chloride content to be affected by factors other than infection.

**REFERENCES**


Determining Riboflavin in Dried Milk Products
II. Seasonal Variations

Royal A. Sullivan and Evelyn Bloom
Kraft Cheese Company, Chicago, Ill.

The average amount of riboflavin in various dried milk products has been reported (1) and slight differences in the riboflavin content of milk from different breeds of cows has been noted (2, 3). In one locality it was found that summer milk contained more riboflavin than winter milk (2) while in another locality no difference was observed between milk obtained in May and that obtained in November (4). A study in one state of commercial milk as compared with that obtained from the experiment station farm, has demonstrated a 20 percent seasonal variation for the former as compared with a 13 percent variation for the latter (5). This has been attributed to differences in the silage fed, a factor which has already been shown to be of great importance in controlling the concentration of riboflavin (5).

Since dried milk products are among the most important sources of riboflavin for animal and poultry feeding, the question of a seasonal variation in concentration is of considerable practical significance. Before much of the above information became available, it was decided in the early part of 1937 to carry on an extended study of this problem. Instead of limiting the observations to a single herd of cows or even to an isolated area, a survey was made of several different states which were representative of the main sources of dried whey. The results would therefore indicate whether, throughout the country, there was sufficient seasonal variation in this factor to influence the value of milk products as feed supplements.

The material most readily available for this study was dried whey from cheese. Although the results obtained could not be interpreted directly in terms of liquid whole milk, any changes which were observed in dried whey would be an indication of corresponding changes in the original milk. The validity of such an interpretation is based upon the common observation that the major portion of the total riboflavin content of whole milk is found in the whey after the cheese-making operation.

EXPERIMENTAL

Six drying plants were selected for this study, and these were located in five different states. It was thought that in this manner the influence of any unusual local climatic conditions would be eliminated. Several of these plants combined their whey with that from other nearby cheese factories, so that the final product represented a composite sample from a very large number of dairies.

Bi-monthly samples were obtained from these plants over a period of two years beginning in July, 1937. The volume of milk which these individual samples represented may be judged from the total annual production of 5,300 tons of dried whey from these six drying plants.

The riboflavin concentration of the dried whey samples was determined by the photometric procedure which was described in the first paper of this series (6). The method is based upon the extraction of riboflavin with acid-acetone and the destruction of colored impurities by mild oxidation. After filtration, the riboflavin concentration is measured by the determination of light absorption before and after reduction to the leuco form. It was shown that when this method is applied to dried milk products, the results are reproducible to within one microgram per gram.

Each sample was run in duplicate and the average of several readings was taken. In Table 1 each horizontal row represents the values obtained from a single plant at three-month intervals. The mean value for each season together with its probable error is given at the foot of the table.

<table>
<thead>
<tr>
<th>TABLE 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ribo­flavin concentration of bi-monthly samples of dried whey obtained from five states over a two-year period.</td>
</tr>
<tr>
<td>Summer Fall Winter Spring</td>
</tr>
<tr>
<td>23.4 23.6 23.0 23.6</td>
</tr>
<tr>
<td>25.4 25.6 25.0 25.2</td>
</tr>
<tr>
<td>27.4 27.6 27.0 27.4</td>
</tr>
<tr>
<td>29.4 29.6 29.0 29.4</td>
</tr>
</tbody>
</table>

The only significant differences which were observed were for the mean value for winter as compared with those for the other three seasons. Even in this case the average value for winter was only seven percent lower than that for summer. This is in good agreement with the value of thirteen percent for an individual herd and considerably better than the twenty percent variation found in one locality for commercial milk (3).

In Figure 1, the results from the two-year survey have been averaged in order to demonstrate the general trend of the riboflavin concentration throughout the year. Several factors appeared to have influenced the riboflavin concentration. The most obvious one was the effect of green pasture in the spring. Since the concentration did not continue to increase uniformly up to its maximum value, it may be concluded that for a time the volume of milk produced was increasing too rapidly for the riboflavin to keep pace. In confirmation of this explanation, it should be pointed out that a negative correlation between riboflavin concentra-
The riboflavin content of dried whey from cheese. A total of 244 samples, collected from five different states, was analyzed by a photometric procedure. The average concentration of all samples was 25.1 micrograms of riboflavin per gram, a value which is in excellent agreement with the published value of 25 for dried whey from cheese (7). A statistically significant difference was found between the mean value for winter and that for any other season of the year. Contrary to the belief of many people, the maximum deviation amounted to only seven percent of the total concentration. This small seasonal variation would therefore be insignificant for practical feeding purposes. The graphical representation of the results gave indications that the seasonal variation was not a simple phenomenon but depended upon several factors.

**REFERENCES**


**Mere Possession of Unwholesome Food Not Illegal**

Conviction of violation of city sanitary code in keeping unwholesome canned grapes reversed.—(New York Court of Appeals; People v. Wallace & Co., 26 N.E.2d 959; decided April 16, 1940.)

The defendant company was convicted of a violation of section 163 of the New York City Sanitary Code. This section provided, among other things, that no vegetables not being then wholesome or safe for human food should be brought into the city or kept, offered for sale, or sold as such food, or kept or stored anywhere in the city, and that any vegetables packed in cans, the contents of which had become fermented as evidenced by swelling or bulging, should be deemed not wholesome or safe for human food. The term "vegetables" included any article used as and for human food other than milk or meat. The case against the defendant, a candy manufacturer, was that a health department inspector had found, in a storeroom of defendant's factory, 12 cans that were swollen and bulging and which contained grapes that had become unwholesome. These cans had been in the storeroom for at least a month. In defense the proof was that the defendant made no use of the foodstuffs kept under lock in its storeroom without first inspecting them and that any article found on inspection not to be wholesome was put aside for return to the seller. There was no proof in respect of the time when the grapes had become fermented, nor was it shown that anything in respect of the time of fermentation could have been validly inferred from the swollen and bulging shape of the cans.

In considering the case on appeal by the defendant, the court of appeals said that, on such record, the judgment of guilt must mean that the defendant's mere possession of the containers made it answerable as for a crime once the enshealed grapes became unwholesome, and that the broad text of the section—that no unwholesome food should be "kept or stored anywhere in the said city"—appeared to go a long distance in that direction. "But a penal statute," stated the court, "is not necessarily to be liberally applied in all circumstances." It was said to be the court's best judgment that no considerations of expediency required such unfairness as would result were section 163 to be so freely construed as to force its application to the facts which in the instant case were found below. The judgments were reversed and the information dismissed.

*Pub. Health Reports, September 20, 1940, p. 1744.*
Dairy Herd Management Practices Affecting the Quality of Milk *

A. C. Ragsdale
Chief, Dairy Husbandry Department
University of Missouri

A successful sanitation program calls for the coordinated efforts of many individuals, officials, and groups. The chief objective is safeguarding the public health. This is the economic problem of the milk producer who is receiving the only incidental consideration. Some may disagree, but it is my judgment that it has been is the inclination of public health and milk inspectors, and local officials, to place reliance chiefly on the owner and herd manager, but not just in a place in which to keep cows.” The intelligent owner or herd manager knows that without a high quality product that commands a good price in a competitive market, his chances of economic success are reduced. Where is there a good dairy farmer who does not take satisfaction and pride in building up a good herd and marketing a healthful product of high food value? It is not natural for a man to do a good job of breeding, feeding, and managing his herd and who is economically successful to be indifferent and careless about milking and caring for the milk of the cows under his supervision. He has a right to expect an intelligent understanding of his problems and his business on the part of public health officials.

Good quality milk must be clean, free from pathogenic bacteria, high in food value, have no unpleasant odors or tastes, and have good keeping qualities. Milk of poor sanitary or nutritive quality results from:

1. Disease organisms transmitted to the milk from cows infected with certain diseases, the most important of which are mastitis, Bang's disease, and tuberculosis.
2. Disease organisms introduced by persons handling the milk or from other sources, among the more common of which are typhoid and paratyphoid fevers, streptococcus infections such as septic sore throat, diphtheria, and scarlet fever, diarrhea in children, and diphtheria.
3. Milk utensils and equipment of poor construction or not properly cleaned and sterilized.
4. Infected water supply or sewage contamination.
5. Barn and milk rooms poorly located or constructed and inconvenient in arrangement.
6. Improper feeding, herd management, and unsanitary practices.

The six points just named may usually be controlled in such a manner as to insure high quality milk and milk products where there is:

1. An intelligent understanding and viewpoint on the part of the herd manager, assuring his economic success.
2. An intelligent understanding and common sense viewpoint on the part of the inspector and public health officials, particularly their showing ability to secure cooperation in all real essentials.
3. Cooperation, careful planning and educational work on the part of milk producers and public health officials in providing a safe milk supply, his viewpoint and methods, have a very direct and significant influence upon the quality of the milk, and the goal of the good inspector is to arouse his interest and secure his active cooperation.

The chief responsibility for the care and management of the dairy herd and the care of the milk produced rests first and foremost on the farmer or herd manager. Whether a small producer where economic problems of the milk producer ordinarily receiving little testing was done, it decreased by 76 percent from 1917 to 1936.

In other words, the tuberculosis eradication program in cattle once it got under way on a large scale has proven more effective than the best that medical science has been able to do in the case of tuberculosis of human origin.

Bang's disease, because of its relation to undulant fever in man and because of its great economic importance to the dairy farmer, needs special consideration. Fortunately, it is possible to detect animals infected with this disease by means of the "blood test" with a high degree of accuracy, especially when regular tests are conducted at short intervals. The economic aspects of this disease have special regard for the farmer. The incidence in infected animals (a) the milk yield is reduced 20 to 25 percent, (b) the loss of calves averages 30 to 40 percent greater than in healthy animals, (c) the calfng interval is increased from a normal of 12 months to approximately 20 months, (d) one out of five aborting cows becomes sterile and replacements in the herd are increased by 30 percent. Control measures consist chiefly of regular blood tests, removal of infected cows, and a carefully planned sanitation program.

Mastitis is a major dairy problem from both the economic and hygienic standpoint. Methods of detecting this disease and control measures are not so well worked out as in the case of tuberculosis or Bang's disease. Nevertheless, reasonably efficient control is possible with the help and cooperation of skilled bacteriologists, veterinarians, and herdsmen. The careful bacteriological examination of milk, use of the Hotis test, determination of leucocytes, titration for chlorides, and the catalase test give the bacteriologist a reasonably satisfactory means of diagnosis. Likewise, a physical examination of the udder by an experienced veterinarian gives fairly satisfactory results. Watching the herd closely for injured and abnormal udders and abnormal milk, the use of the strip cup and regular use of one or more of the bacteriological or chemical tests just mentioned, coupled with the physical examination, gives the herd
manager the principal information he needs for establishing management prac-
tices to obtain the elimination of the
disease. Isolation of infected cows, milking them last or better still by a dif-
f erent milker and by hand is suggested. A care ful and strict sanitation program is always essential.

DISEASES TRANSMITTED BY MAN AND
MISCELLANEOUS SOURCES
When the herd man age sters the importance of personal hygiene, that no person who is sick or just recovering from any disease should act as a milker, care for the milk or handle milk utensils, the danger of introducing disease organisms is reduced to a minimum. His co-
operation in proper cleansing and sterilization of all utensils and equipment, con-
trol of flies and their exclusion from milk and milk utensils, and avoidance of water contamination of the water supply eliminates the last possibility of introducing harmful organisms.

FEEDS, FEEDING AND MANAGEMENT
Nutritive Value and Color of Milk. The kind of feed is important in supply-
ing the vitamin A and D requirements of cattle and influences the vitamin con-
tent and color of the milk. The rela-
tionship of feed to the vitamin A content of the milk deserves special mention. All green pasture crops, green colored, prop-
ely cured hays, and properly preserved fresh green silage crops provide liberal amounts of vitamin A. Much of the caro-
etene and vitamin A value of hay is de-
stroyed by oxidation from excessive expo-
sure to sunlight and weathering in field curing. Conse quently, hays that are cut in the early blosom stage, cured without exposure to rain or too much sun, retain a larger proportion of their carotene con-
tent than those cut in the later stages and exposed for long periods to sun or rain. If hay heads severely in the mow, there is always a slight loss of carotene. In the artificial curing of hay there is only a slight loss of carotene and consequently it is high in vitamin A value. In general, the amount of green color in hay is in direct relation to the amount of carotene. Among the vegetable oils, cod liver oil, fish oils, egg yolk, and milk fat are ordinarily rich sources of this vi-
tamin.

Among cattle there are marked breed differences in their ability to convert caro-
etene into vitamin A. This is reflected in the color of the body fat and milk fat of the several breeds. It is important to note that the yellow color of carotene, and consequently yellow color, present in the milk is not a true measure of its vitamin A value since it does not give a measure of the vitamin present as such. For example, in the milk of Guernseys and Jer-
seys most of the vitamin A value is due to carotene, whereas Holstein, Brown Swiss, and Ayrshire milk contains less of the pigment and more of the colorless vitamin A as such. As a result, hays of the various breeds, when fed the same rations, may produce milk and fat of equal vitamin A value although differing in yellow color. The extent of the con-
version of carotene into vitamin A also varies with individuals and the species. In fact, the occurrence of carotene in milk is limited primarily to the bovine species. The milk of the goat and of women, for exam-
ple, is nearly colorless because of the very complete conversion of carotene into vitamin A, and thus the vitamin A value of their milk may be high even though no color is present.

Flavor and odor of milk. Fresh clean milk has a rich and pleasing taste. Slight de-
defects in flavor may prevent full enjoy-
ment of milk and thus curtail consump-
tion. During the spring months in par-
ticular when the cows are on pasture, complaints concerning undesirable flavors in milk and its products are most com-
mon.

Flavors in milk may originate at vari-
ous stages in its production and handling. Bad flavors are not necessarily associated with the safety of the milk for food pur-
poses nor is its bacterial content always high. Such flavors may be present when the milk is drawn and these are largely dependent upon two factors—the physical

condition of the cow, and the kind of feed consumed. Abnormal conditions of the udder, such as edema or mastitis, may cause an "off" flavor. Abnormal conditions of the udder and edema or mastitis, may cause an "off" flavor. Among the many of the more suscep-
tient green feeds have strong flavors which are transmitted to milk. A change from dry feeding to grass always results in a different flavor appearing in the milk, which sometimes causes complaints from customers. These flavors can be prevented by following a well planned routine of feeding. Strong flavored feeds should be fed either several hours before or dist-
ally after milking. When fed at milking time, or even one hour before, such feeds as silage, green alfalfa, sweet clover, green rye, and turnips affect the flavor and odor of milk. Wild onions, garlic, and bitterness affect the flavor as soon as eaten and continue to do so for at least 4 to 7 hours. In early spring and particularly when pastures are short and weed-infested, weed flavors are quite common since cows are forced to eat herbage they might otherwise refuse. The various flavors imparted by weeds can be held to a minimum by removing the cows 3 to 6 hours before milking time.

The exposure of milk to the rays of the sun seriously affects its flavor. The presence of iron or copper salts causes a speedier action of sunlight than would otherwise occur. The "off" flavor developing in this instance is usually described as "allowy", "cardboard", "metallic", or "astringent". The use of poorly tinned milk cans, buckets, coolers, vats, etc. is responsible for many "off" flavors, and also adds copper and iron salts to the milk. This aids in developing the flavors associated with exposure to sunlight.

Washing compounds and chemical dis-
si nfectants, if care lessly used, may be re-
sponsible for the addition of foreign flavors. Only readily soluble cleaners free from odors should be used for milk utensils. Chemical disinfectants such as chlorine solutions used according to dire-
tions cause little trouble. Poor drain-
ing of cans and utensils after sterilization, or use of excessive amounts of this agent may, however, cause trouble.

After the milk is drawn, unless the utmost care and cleanliness is exercised, it may acquire an "off" flavor. Odors absorbed from the barn, milk house, and general surroundings often affect the flavor and destroy the pleasing taste of milk. Oils, fly sprays, and medi-
cines used about the barn often impart flavors. Removing milk from the barn immediately after it is drawn, prompt cooling to 50° F. or lower, and holding it at this temperature in a clean place, largely eliminates the dangers from "off" flavors of the absorbed type.

In conclusion, may I point out that compared with other types of busi-
nessmen, dairy farmers are efficient. The percentage of failures falls well below the national average for all business. Education on the dairy farm is progress-
ing. The successful milk inspector, health officers, and others who occupy places of public trust should never lose sight of their subject. They should know more than the average person with whom they work—yes, they must stand out above the crowd. The more the milk inspector uses the educational method and the less conspicuous he makes his authority, the more he believes in the value of milk and its products, the grea t er his enthusiasm and the better his qualities of salesmanship, the more likely he is to succeed. We need ever-increasing intelligence, under-
standing, better planning, and team-work on the part of the milk producer—yes— and likewise on the part of public health officials. Let the milk inspector and public health officials really join forces with the strong educational agencies now working with dairy farmers, as educators, if you please, and the public will be assured of a safe milk, high in nutritious value, and pleasing to the taste.
The Need of Milk in the South

O. D. Abbott

Agricultural Experiment Station, Gainesville, Fla.

Approximately 28 percent of the population of the United States live in the 13 states called Southern. In this section, there are 35 percent of the children under 15 years of age. Moreover, in their search for jobs the productive middle age groups leave the South in greatest numbers. This exodus tends to make this section the land of the very old and very young. Now both of these groups, the old and young, are the greatest users of milk. It is the first food for the young and the last for the old. The U. S. census for 1930 shows that 16,666,668 cows were milked daily with a production of 11,053,034,357 gallons; but here in the South where we find 35 percent of the children, there are 22 percent of the cows and 17 percent of the milk. This comparison can be carried still farther. In Florida according to the same census there are 1,468,211 people and an annual production of 26,283,944 gallons of milk. If everyone in the state had a quart of milk a day the supply would last only 72 days, if a pint of milk 144 days and if a half pint the amount Streibling puts in the lowest cost protective diet for adults, then there would still be 77 milkless days. It is true of course that many farms did not report on milk production. Moreover, considerable evaporated and dried milks and also ice cream are used, but evaporated milk is used mainly in infant feeding and in seasoning, very little is used in Florida as a beverage, while ice cream is still considered a luxury.

If we examine the people of this state, what evidence is found that indicates that milk consumption is too low? In other words, can specific conditions be traced to a deficiency of milk?

Before determining this, it is necessary to know what milk contributes to the diet and the effect of a lack of it on the health of the people. Milk furnishes proteins of high quality, a carbohydrate, lactose, and butter fat, an easily emulsified fat. But the outstanding nutritional advantage of butter over other fats lies in the fact that butter fat is associated with the fat soluble vitamin A. Milk is also an important source of vitamin G, and if handled so as to conserve its vitamin C value, is a good source of this vitamin.

In addition to these constituents, milk contributes a very well balanced mineral mixture to the dietary. This is especially true in regard to calcium. As a rule the calcium content of the diet depends mainly upon the amount of milk used. In the family dietary where limited amounts of milk are used, the diet is more often deficient in calcium than in any other element. A liter of milk furnishes approximately one-half the daily calcium requirement. Milk also furnishes phosphorus compounds, and while the amount of iron in milk is very small it appears to be well utilized. But of all these constituents the chief value of milk appears to lie in its protein, calcium, and vitamin A.

What effects then would a lack of these food factors have upon the health of the individuals? In young children milk furnishes the greater part of the protein, and in the absence of it the protein is apt to be too low for optimum growth. The addition of meat or vegetable protein in amounts to furnish adequate protein often brings digestive disturbances. On the other hand, milk protein is particularly adapted for growth and also for the repair and maintenance of adult tissues. The proteins of milk and eggs are well suited for conversion into body proteins and for this reason should be used extensively in the diet of children and any one who needs to be built up.

In the absence of adequate protein, growth would be retarded. Many investigators have shown the marked influence of milk upon the growth of children. They found that children getting milk showed more of that lassitude peculiar to well nourished animals, greater alertness and buoyancy of spirits, and greater height.

To be normal and healthy the full grown human body must be richer in calcium than in any other mineral element, yet every one is born calcium poor. This calcium poor conditions of an animal is due to the fact that the bones are soft and this facilitates birth. Therefore, after birth the diet should contain an abundance of calcium to insure bone and tooth development. During pregnancy and lactation a mother's diet should be high in calcium else her bones and teeth will be sacrificed to supply this essential element. Then with a deficit of calcium poor conditions of the teeth and poorly formed bones would probably result. According to Sherman an absence of calcium or phosphorus in the diet of the young is apt to result in a permanent loss of the long, lithe form, the skeleton becoming unduly stocky, if not actually distorted.

As mentioned before, whole milk is one of the best sources of vitamin A. While green and yellow vegetables and fruits are high in carotene, the precursor of vitamin A, and egg yolk is a good source of vitamin A, it is the experience of many that a diet low in milk and butter means a diet low in vitamin A. The clinical symptoms in children attributed to a lack of vitamin A are retarded growth, eye and ear defects, conjunctivitis, dry hair and skin, and increased susceptibility to infections; in the adult, dry hair and skin, and eye defects. It would then appear that a deficit of protein, calcium, and vitamin A would tend to inhibit growth. Nutritionists now recognize the difference between adequate and optimum nutrition just as physicians consider good health as more than freedom from disease.

Now let us examine the health records and see if we can find conditions which might indicate a lack of these constituents. In a nutritive study of approximately 4000 rural white school children in Florida, it was found that the average 8-year old girl was 2½ inches shorter than the age-height standard given by Rose and Holt, while the girl at 13 was 3 inches shorter than either standard. Similar observations were made on the boys. The 16-year old boy was 3 inches shorter than Rose's standard and 4 inches by Holt's.

In later nutritional studies of approximately 5000 children, the prevalence of vitamin A deficiency among a large number of children was established by blood studies and clinical examination. Recently studies were made on University students. Only those students using neither milk nor butter in the diet were examined. In 60 percent of cases symptoms of vitamin A deficiency were found. More recent examinations of women on reducing and restricted diets showed symptoms of vitamin A deficiency. In the women and students the outstanding symptoms were red, itching, and burning eyes, and dry hair and skin. Most of the women and about half the students wore glasses but in spite of this the eye defects prevailed. Among the children, retarded growth, dry skin and hair, and conjunctivitis were not. That the diagnosis was correct was confirmed by giving large doses of vitamin A to selected subjects. In 4 to 6 weeks the gross symptoms of avitaminosis A had disappeared and in a few more weeks the blood picture returned to normal. Of course during that time no changes in growth were noted. After this intensive treatment the students and children were given one quart of whole milk per day and urged to eat large amounts of butter. The women on the reducing diets, however, continued to take vitamin A concentrates.

In the study of defects of 4000 rural school children, carious teeth were found in 45 percent of them. In many cases...
of hookworm. From this work it was concluded that in any community where parasitic infestation was endemic and where reinfection was common, the maintenance of children on a high plane of nutrition was imperative.

Several months ago in making recommendations for the food supply for humans and livestock in the state it was brought out that in estimating the amount of corn necessary to keep a horse or cow, double the necessary amount must be raised because the weevil ate half. Therefore, in planning the milk supply for Florida children, it is evident that after giving the child his quart, enough should be allowed to take care of the hookworm.

SUMMARY

Data have been presented on 10,000 school children, approximately 50 university students, and 400 women. Among these subjects, abnormal height-age relationships, vitamin A deficiency, and carious teeth were the symptoms which may be related to a lack of milk in the diet. Moreover, it has been shown that milk production and milk consumption in the state is much below even the lowest amount recommended as barely protective. Even with only one-half pint a day for the most part, no doubt, to inadequate diets, perhaps calcium. And in the case of these women the old saying, "Every child takes a tooth" is an actuality. Later a study was made of the relation of diet to teeth. In this study 406 children were examined. It was found that 33 percent of the children using milk had teeth which were defective, while a little more than 10 percent of those not using milk had good teeth. The incidence of carious teeth was twice as common for the children who did not use milk as for those who did. Moreover, chalyte teeth were also more prevalent in the former group. Of late many investigators have shown that dental caries has been reduced by improving the diet, and an increased use of milk is a part of the dietary regime.

While the effects of parasitic infestation, especially hookworm on the physical development of children is not to be minimized, in the study of hookworm in man it has been noted that a well-nourished adult may often have a heavy infestation of hookworm and show neither a reduction in hemoglobin nor other noticeable clinical symptoms. Investigations conducted in Brazil and in this laboratory on hookworm infested subjects show the beneficial effect of milk. Smillie found that a group of hookworm infested milkers who drank large amounts of milk were better nourished and showed fewer symptoms of hookworm infestation than workers of the same age infested with a comparable number of hookworms but who did not use milk. In this laboratory a study of the effects of diet on children with a moderate number of worms, both hookworm and ascaris, showed that the symptoms usually associated with hookworm infestation were alleviated if large amounts of milk were included in the diet. Moreover, ovacounts made throughout the experimental period showed a reduction of 100 percent for ascaris and a trend towards reduction of hookworm. From this work it was concluded that increased consumption of milk is a part of the dietary regime.

An amendment to the Constitution, reducing membership dues, was adopted at the last meeting of the Association. This was done with the thought that many persons interested in milk sanitation would find it possible and desirable to enter the Association and participate in its activities. This belief was well founded. Five hundred and twenty-five new members have joined since our last meeting. At present we have members from 41 states, the District of Columbia, Alaska, Canada, British Columbia, Cuba, Mexico, British West Indies, South America, England, Ireland, and India. Impressive as these figures are, it is the interest that is being shown by all members which is significant. There are 284 Active Members and 622 Associate Members, making a total of 907 persons.

The Journal of Dairy Technology has continued its progress in interest, value, and circulation. It is now well established, and there is every indication that the Association will be able to recompose the editor, the managing editor, and the necessary clerical assistants to a greater degree than has been possible heretofore. During the past year, the California Association of Dairy and Milk Inspectors, the Pacific Northwest Association of Dairy and Milk Inspectors and the Pennsylvania Association of Dairy Sanitarians have designated the Journal as their official publication.

There are three provisions which are now under consideration and study by the Association:

1. Awards for meritorious and outstanding service in the field of milk sanitation.
2. Student training in relation to the Dairy Industries Exposition, and
3. Affiliations of local milk sanitarian organizations with this association.

Each of these projects has interesting and valuable possibilities, and merits serious study and consideration. Various individuals and committees have been working on these proposals and much has been accomplished. In the matter of affiliations—all local and International associations—constitutional and financial obligations must be considered. Studies so far indicate that a plant for affiliation can be devised and successfully operated.

However, it is believed that further studies should be given to these three proposals. As soon as this has been done, definite plans will be laid before the Association. It appears as if this could be done during the year and that definite action could be taken at our next meeting.

To the many individuals, committees, and organizations which have cooperated with the Association in its work, to the President for his mature counsel and advice, to the other members of the Executive Committee, and to the Managing Editor and Editor of the Journal for their interest and work, your secretary is deeply indebted. Any success which has been gained has been the result of splendid cooperation. This same cooperation will carry through successfully the problems and projects which are before us, with benefit not only to the Association but to the entire dairy industry.

Respectfully submitted,
C. Sidney Leete, Secretary.
Oct. 15, 1940.
New Books and Other Publications

Veterinary Bacteriology, by I. A. Merchant. The Iowa State College Press, Ames, Iowa. 1940. 628 pages, $7.00.

This book was written as a textbook and served as an introduction to bacteriology to students in veterinary medicine. It emphasizes the general morphological and physiological characteristics of bacteria, and is concerned primarily with the species of bacteria, yeasts, molds, and filamentous viruses which are pathogenic to animals. It acquaints the student with bacteria which are pathogenic to animals, those which are pathogenic to both man and animals, and with certain of those which are pathogenic to man only. The book contains no section on the pathogenic protozoa nor on laboratory methods of milk analysis.

The first 128 pages deal with the general biology of microorganisms. The second section of 82 pages deals with infection, resistance, and immunity. The third section of 318 pages deals with the classification and characteristics of pathogenic bacteria. The fourth section of 65 pages covers filamentous viruses. At the conclusion of each chapter, the author appends a list of references for further study.

Numerous illustrations and tables clarify the text. The index is adequate and well arranged. The printing is clear and well organized.

This book will be valuable to dairy farm inspectors. It will refresh the minds of veterinarians with much of what they have learned, and will bring to their attention some of the newer developments in the field. The long experience of the author in the application of veterinary practice to milk sanitation gives the book authoritative and usefulness in a field of practical importance. J. H. S.


The author has compiled authoritative information on milk distribution. He develops the subject under the following headings: Part I—Historical and introductory background (in which he compares the characteristics of public utilities with the fluid milk business); Part II—Cost and profits of distributing milk and savings through unification (in which he analyzes operating costs and the possible savings through unification); Part III—Legal considerations control; Part IV—Methods and difficulties of regulation, and Part V—Economic effects of regulation.

He concludes that a unified system of milk distribution could bring about a reduction in milk distribution costs by amounts varying from about 1/2c to even 2/3c per quart of milk handled. However, he emphasizes that there is no proof or certainty that they actually would be carried out, because of the difficulty of securing either an efficient management or freedom from political interference. The author raises many very practical and pertinent questions which are often ignored when the uninformed public endeavors to dictate how the milk business should be run. In view of the fact that the subject of the distribution of milk is such a recurring problem in so many communities, this book will be useful to those milk control officials and industrialists who want to be kept informed on this important question. The book is well-printed, easy to read, well-organized, thoroughly indexed, and adequately indexed. J. H. S.


This book embodies the results of the authors' study of the isolations of streptococci during the past ten years of over 25,000 samples of milk drawn directly from the cow's udders and plated on blood agar. The book describes in detail the methods used and the results found. Among the interesting data presented is information concerning their discovery of a new species which they call Streptococcus zooepidemicus. They also cultured the throats of about 3,000 dairy plant employees working on five different farms. Ten species of hemolytic streptococci were identified. The most common enzymes were pyrogenes, which was to be expected, mastitis, which has not been reported before and was not expected, and infrequent, which has been reported as occurring infrequently. The most significant findings were the two species zooepidemicus and epidermis, for which differentiation is given in detail. The authors append a bibliography of about 622 references. J. H. S.


The combined ordinance and code for the control of frozen desserts has been issued by the United States Public Health Service in mimeographed form, accompanied by the "Frozen Desserts Plant Inspection Form". The general arrangement, form, and scope follow closely those now in use in the well-known milk ordinance and code. The long study that has been devoted to the drawing of this ordinance and code, preceded by the experience in the application of the milk ordinance and code, would seem to give this document an immediate and wide applicability.

The subject comprehends not only the commonly accepted frozen desserts but also the partially frozen ones which are similar to these, including frozen custard, ice milk, and ices. Both the ordinance and code are arranged conveniently for adoption of either the grading or the non-grading type by merely deleting certain phrases.

Pasteurization is prescribed at a minimum heat treatment of 155°F for 30 minutes (or a corresponding effective method). Both the new tryptone-glucose-extract-milk agar and the old beef extract-peptone agar are allowed "where the new medium is found to yield considerably higher counts", Grade A bacterial standards are 50,000, Grade B 100,000, and Grade C no limit, per gram. The requirements are reasonable and practical. Some parts, such as the medical check-up, might be made more specific for the guidance of the enforcement officer as well as the dealer. J. H. S.


In the ten years that have elapsed since the appearance of the first edition of this book, the field of dairy bacteriology has developed extensively. This new edition brings these fields into agreement with the present day concepts. Much new material has been added, and yet the size of the book has been kept very close to that of the first edition by the judicious elimination of some good text that was informative but not strictly necessary in a book of this type. The sections on spread of diseases through milk and the bacteriology of butter have particularly gained in amount of treatment. The index has been enlarged and improved. The format of the earlier edition has been maintained with its wealth of useful information. J. H. S.
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Charleston, W. Va.
Association News

California Association of Dairy and Milk Inspectors

Legislation favored by the Association will be handled differently in the future from what it has been in the past. In order to eliminate confusion, the members have appointed a legislative committee with Mr. H. E. Erikson of Santa Barbara as chairman. With the aid of suggestions from the committee, Mr. Erikson will clarify resolutions and represent the Association in matters of legislation.

LEONARD E. NISSON, Secretary-Treasurer.

Central States Milk Sanitarians

The meeting of the Central States Milk Sanitarians, held on November 4, was attended by about one hundred members. Mr. L. E. Bober gave an interesting and enlightening address on "Mastitis." Dr. W. H. Haskel spoke at length, and among other things stated that the Chicago milk supply is one of the finest in quality of any in the country. The feeling was strongly apparent that it is most beneficial for men interested in the quality production of milk to be united in an association, such as this, and to attend the meetings where subjects in this field are discussed with such profit.

D. V. FITZGERALD, Secretary.

Chicago Dairy Technology Society

On November 19, 1940, the Chicago Dairy Technology Society met and heard Dr. N. E. Fabricius of Iowa State College speak on "New Developments in the Butter Industry, with Special Reference to the Vacreation of Cream for Improving the Quality of Butter, also for Improving the Quality of Ice Cream Mix." He described the Vacreator, a device for pasteurizing and treating cream and other products under vacuum, and discussed experiments connected with its performance.

Mr. Bonewitz, of the P. W. Bonewitz Chemical Company, summarized the research on control of proteolytic organisms in milk cans by treating with steam acidulated with gluconic acid.

J. T. THORNE.

Massachusetts Milk Inspectors' Association

Legislation favored by the Association meeting in Springfield, Massachusetts, on November 1, President Enright introduced Dr. Edwin M. Knight, who gave a paper on "The Milk Serum Test used in Detecting Bang's Disease."

Professor J. H. Frandsen of Massachusetts State College spoke on the new equipment for milk plants and ice cream manufacturing which he saw at the Dairy Show in Atlantic City. Dr. Carl Fellers, also of Massachusetts State College, gave a talk on "Food Poisoning."

Nomination of officers for the year 1941 was held at the business meeting, and election will take place at the meeting in Worcester, Massachusetts, on January 8 and 9. Dr. Wolman from Philadelphia will talk on "Homogenization," and Dr. Workman of Connecticut, on "High-Temperature-Short-Time Pasteurization."

The Association extends a cordial invitation to all to attend its meetings in connection with the Union Agricultural meeting and display of equipment at the Memorial Auditorium, Worcester, on January 8 and 9.

ROBERT E. BEMIS, Secretary-Treasurer.

Michigan Association of Dairy and Milk Inspectors

In conjunction with the Michigan Allied Dairy Association Convention and Machinery show, the annual meeting of the Michigan Association of Dairy and Milk Inspectors will be held in Grand Rapids on March 12 and 13, 1941. The program will consist of reports and discussions on the progress and work of the Committees on standardization of requirements for dairy farms, dairy plants, ice cream, and butter plants; new tests for quality in the dairy field; can and bottle washing problems; and a question and answer period.

H. J. BARNUM, Secretary-Treasurer.
## INDEX TO VOLUME 3

### Authors

<table>
<thead>
<tr>
<th>Author</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abbott, O. D.</td>
<td>The need of milk in the South</td>
</tr>
<tr>
<td>Abele, C. H.</td>
<td>Flavors and odors of milk</td>
</tr>
<tr>
<td>Babcock, C. J.</td>
<td>Flavors and odors of milk</td>
</tr>
<tr>
<td>Bloom, E.</td>
<td>Necessity for and some difficulties of public health milk control</td>
</tr>
<tr>
<td>Butterworth, T. H.</td>
<td>The training of personnel for the field of milk sanitation</td>
</tr>
<tr>
<td>Caulfield, W. J., Nelson, F. E., and Martin, W. H.</td>
<td>Measuring the sanitary quality of market cream</td>
</tr>
<tr>
<td>Chilson, W. H. and Collins, M. A.</td>
<td>Application of the reazurin test in determining the quality of pasteurized milk</td>
</tr>
<tr>
<td>Cohen, L.</td>
<td>Obtaining good results with Broadhurst-Paley stain for milk smears</td>
</tr>
<tr>
<td>Collins, M. A.</td>
<td>See Chilson, W. H.</td>
</tr>
<tr>
<td>Cone, J. E. and Grant, F. M.</td>
<td>Improvement of the Hots Test for the detection of mastitis streptococci</td>
</tr>
<tr>
<td>Guthrie, E. S.</td>
<td>See Sharp, P. F.</td>
</tr>
<tr>
<td>Hall, A. H.</td>
<td>Training in the public service in New York State</td>
</tr>
<tr>
<td>Hand, D. B.</td>
<td>See Sharp, P. F.</td>
</tr>
<tr>
<td>Howson, R. K.</td>
<td>See Johns, C. K.</td>
</tr>
<tr>
<td>Hucker, G. J.</td>
<td>The elimination of mastitis</td>
</tr>
<tr>
<td>Also, see Dick, L. A.</td>
<td></td>
</tr>
<tr>
<td>Irwin, R. E.</td>
<td>The effect of the inspector system program on uniformity?</td>
</tr>
<tr>
<td>Johns, C. K. and Howson, R. K.</td>
<td>A modified reazurin test for more accurate estimation of milk quality</td>
</tr>
<tr>
<td>Kemp, M.</td>
<td>See Scales, F. M.</td>
</tr>
<tr>
<td>Knapp, J. V.</td>
<td>The effect of the</td>
</tr>
<tr>
<td>Dixon, D.</td>
<td>Symposium on tryptone-glucose-extract-milk agar</td>
</tr>
<tr>
<td>Downs, F. H., Jr.</td>
<td>The voluntary grading of milk supplies in Alabama</td>
</tr>
<tr>
<td>Ehlers, V. M.</td>
<td>Syllabus on milk sanitaritians' associations</td>
</tr>
<tr>
<td>Syllabus on an educational program for the production of safe milk</td>
<td>255</td>
</tr>
<tr>
<td>Fabian, F. W.</td>
<td>Chairman—Report of the committee on ice-cream sanitation</td>
</tr>
<tr>
<td>Fisher, M. R.</td>
<td>Common causes for intermittent high bacterial count and positive phosphate tests</td>
</tr>
<tr>
<td>Gilcreas, F. W. and Davis, W. S.</td>
<td>Precision in reading the results of the phosphate test</td>
</tr>
<tr>
<td>Grant, F. M.</td>
<td>See Cone, J. F.</td>
</tr>
<tr>
<td>Graves, F. W.</td>
<td>Determination of mastitis control under several plans</td>
</tr>
<tr>
<td>Grenier, T. J.</td>
<td>How to overcome defective Babcock cream tests</td>
</tr>
<tr>
<td>Griffith, R. L.</td>
<td>Comment on licensing of pasteurizer operators from Oakland, California</td>
</tr>
<tr>
<td>Guthrie, E. S.</td>
<td>See Sharp, P. F.</td>
</tr>
<tr>
<td>Hall, A. H.</td>
<td>Training in the public service in New York State</td>
</tr>
<tr>
<td>Hand, D. B.</td>
<td>See Sharp, P. F.</td>
</tr>
<tr>
<td>Howson, R. K.</td>
<td>See Johns, C. K.</td>
</tr>
<tr>
<td>Hucker, G. J.</td>
<td>The elimination of mastitis</td>
</tr>
<tr>
<td>Also, see Dick, L. A.</td>
<td></td>
</tr>
<tr>
<td>Irwin, R. E.</td>
<td>Has the approved inspector system program uniformity?</td>
</tr>
<tr>
<td>Johns, C. K. and Howson, R. K.</td>
<td>A modified reazurin test for more accurate estimation of milk quality</td>
</tr>
<tr>
<td>Kemp, M.</td>
<td>See Scales, F. M.</td>
</tr>
<tr>
<td>Knapp, J. V.</td>
<td>The effect of the</td>
</tr>
<tr>
<td>Bang's disease control program on milk production in Florida dairies</td>
<td>35</td>
</tr>
<tr>
<td>Krauss, W. E.</td>
<td>Nutritional aspects of milk</td>
</tr>
<tr>
<td>Layson, V. F.</td>
<td>Familiarizing milk plant personnel with sanitary requirements</td>
</tr>
<tr>
<td>Levowitz, D.</td>
<td>Closures employed for dairy products containers</td>
</tr>
<tr>
<td>Lind, H. E.</td>
<td>Symposium on tryptone-glucose-extract-milk agar</td>
</tr>
<tr>
<td>Little, L.</td>
<td>Comparative studies upon the methyl blue and reazurin tests</td>
</tr>
<tr>
<td>Little, R. B.</td>
<td>A discussion of the international classification of the streptococci of bovine mastitis</td>
</tr>
<tr>
<td>Maack, A. C., and Tracy, P. H.</td>
<td>A method for the accurate sampling of ice cream</td>
</tr>
<tr>
<td>McCulloch, E. C.</td>
<td>The early detection of bovine mastitis by an electrometric method</td>
</tr>
<tr>
<td>Marquardt, J. C.</td>
<td>Determining Lactobacillus bacteria in milk with spectrophotometry at 32° C</td>
</tr>
<tr>
<td>Marquardt, J. C.</td>
<td>Detecting cow's milk mixtures</td>
</tr>
<tr>
<td>Also, see Yale, M. W.</td>
<td></td>
</tr>
<tr>
<td>Martin, W. H.</td>
<td>See Caulfield, W. J.</td>
</tr>
<tr>
<td>Miller, H. E., Chairman</td>
<td>Report of the committee on education and training</td>
</tr>
<tr>
<td>Moss, F. J.</td>
<td>Milk investigations of the U. S. Public Health Service</td>
</tr>
<tr>
<td>Nelson, F. E.</td>
<td>See Caulfield, W. J.</td>
</tr>
<tr>
<td>Parker, M. E., Reported by—Abstracts of technical papers presented at thirty-first annual meeting of the American Butter Institute</td>
<td>113</td>
</tr>
<tr>
<td>Safranek, W. E.</td>
<td>Nutritional aspects of milk</td>
</tr>
<tr>
<td>Bang’s disease control program on milk production in Florida dairies</td>
<td>33</td>
</tr>
<tr>
<td>Krauss, W. E.</td>
<td>Nutritional aspects of milk</td>
</tr>
<tr>
<td>Layson, V. F.</td>
<td>Familiarizing milk plant personnel with sanitary requirements</td>
</tr>
<tr>
<td>Levowitz, D.</td>
<td>Closures employed for dairy products containers</td>
</tr>
<tr>
<td>Lind, H. E.</td>
<td>Symposium on tryptone-glucose-extract-milk agar</td>
</tr>
<tr>
<td>Little, L.</td>
<td>Comparative studies upon the methyl blue and reazurin tests</td>
</tr>
<tr>
<td>Little, R. B.</td>
<td>A discussion of the international classification of the streptococci of bovine mastitis</td>
</tr>
<tr>
<td>Maack, A. C., and Tracy, P. H.</td>
<td>A method for the accurate sampling of ice cream</td>
</tr>
<tr>
<td>McCulloch, E. C.</td>
<td>The early detection of bovine mastitis by an electrometric method</td>
</tr>
<tr>
<td>Marquardt, J. C.</td>
<td>Determining Lactobacillus bacteria in milk with spectrophotometry at 32° C</td>
</tr>
<tr>
<td>Marquardt, J. C.</td>
<td>Detecting cow's milk mixtures</td>
</tr>
<tr>
<td>Also, see Yale, M. W.</td>
<td></td>
</tr>
<tr>
<td>Martin, W. H.</td>
<td>See Caulfield, W. J.</td>
</tr>
<tr>
<td>Miller, H. E., Chairman</td>
<td>Report of the committee on education and training</td>
</tr>
<tr>
<td>Moss, F. J.</td>
<td>Milk investigations of the U. S. Public Health Service</td>
</tr>
<tr>
<td>Nelson, F. E.</td>
<td>See Caulfield, W. J.</td>
</tr>
<tr>
<td>Parker, M. E., Reported by—Abstracts of technical papers presented at thirty-first annual meeting of the American Butter Institute</td>
<td>113</td>
</tr>
</tbody>
</table>

### Subjects

<table>
<thead>
<tr>
<th>Subject</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Agar Slice Method for Detection of Mold and Yeast on Utensils</td>
<td>162</td>
</tr>
<tr>
<td>Agar, Tryptone-Glucose-Extract-Milk</td>
<td>208</td>
</tr>
<tr>
<td>Air Infection by Staphylococcus</td>
<td>268</td>
</tr>
<tr>
<td>Alabama, Grading Milk Supplies</td>
<td>97</td>
</tr>
<tr>
<td>Associations, Milk Sanitarians</td>
<td>194</td>
</tr>
<tr>
<td>Bacterial Count and Positive Phos-</td>
<td>165</td>
</tr>
</tbody>
</table>
"Doctor Jones" Says—

"One of the military commentators, here awhile ago—he was discussing the proposal about turning over those destroyers to Great Britain and so on and he said it'd be unfortunate if the decision in a matter like that should become, as he expressed it, 'the subject of ill-informed and irresponsible agitation'. Then he went on to say that matters of policy were quite properly the subject of public debate but when it came to an executive decision in a war situation, that was something else.

"It struck me, as I was reading that: encouraging 'the voice of the people' to express itself and still get the things done that ought to be—it's one of the real problems in a democracy. If they don't have their say, within reason—if the people don't, it ain't democracy. But there's times when too much voice is a serious handicap. Like Louie, the tailor down here: over to the Firemen's Ball he came off the floor and somebody thought he was sick or something. 'No', Louie says, 'it was just that woman's voice: it was so strong it made me dizzy. Ven they eat onions', Louie says, 'they ought to stay home'. Of course, if he hadn't wanted to take a chance on onions he could've stayed home himself. That's democracy.

"But take it in our public health line: there's things we know'd benefit the public if they were done—like cleaning up tuberculosis and pasteurizing milk and so on—us health officers, but it's pretty well agreed you can't move much ahead of public sentiment. And it's awful irritating, sometimes, when you're trying to get something done and you run up against what looks to you like some of that 'ill-informed and irresponsible agitation': people that don't know what it all about leading the opposition and others following 'em like a flock of sheep. I'm a great believer in the freedom of the press but I do know a good health officer: one of the newspapers in his town—I don't suppose they've missed an opportunity in twenty years to oppose or criticise what he did. On the other hand, we can look back and see some things we thought were awful necessary at the time, that it'd have been just as well if they hadn't been done—like building some of those small tuberculosis hospitals. I've been sure I was right, before now, when I wasn't.

"Ill-informed agitation against health measures—the best way to avoid it, the way it looks to me—there's two things: have a full-time health officer that's so competent everybody'll have confidence in him and make a business of keeping folks well-informed. Bad leadership and ignorance—they're termites you've got to look out for in the underpinning of a democracy." *

PAUL B. BROOKS, M.D.

* Health News, New York State, August 26, 1940.