

PATHOGENIC STREPTOCOCCI AND MILK PASTEURIZATION

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DURING the investigation of a recent epidemic of septic sore throat¹ the writers of this article took occasion to make a fairly extensive survey of the literature of the streptococci with special reference to their relation to the spread of disease through milk. The results of this survey, together with certain observations made in the course of the investigation, have impressed us with the fact that streptococci should be given special consideration in connection with the subject of milk pasteurization.

It is a well known fact that the temperature and time of heating necessary to completely sterilize a suspension of bacteria is affected not only by the specific heat resistance of the organism used but also by the number or concentration of the organisms in the suspension. As was pointed out by Phelps² the rate of killing at a given temperature follows the logarithmic curve of the physico-chemical velocity laws. That certain factors related to the nature of the suspension may also affect the thermal death rate appears evident from the results of some experiments described below.

It is the purpose of milk pasteurization, of course, to destroy by heating all of the pathogenic bacteria that may be present in the raw product. It is desirable that the temperature and the time of heating shall be kept as low as possible because of certain alterations in the milk due to heat. These changes are principally those affecting the cream line, the taste and the vitamin content. To some extent these effects bring pasteurization into conflict with business interests and there is, consequently, a more or less constant force being exerted in the direction of lower

standards. Some watchfulness is required on the part of the sanitarian, therefore, to see that these standards shall be maintained high enough to insure the accomplishment of the purpose mentioned at the beginning of this paragraph.

Direct bacteriological control of the process of pasteurization is, of course, not possible. Standards must therefore be determined by experiment. To be accepted as determining reliable standards, such pasteurizing experiments would have to be carried out on samples of contaminated milk showing such a combination of the three factors mentioned above as to represent the maximum resistance to efficient pasteurization that is likely to be encountered in actual practice. It is our purpose here to point out certain facts that lead us to believe that more severe pasteurizing treatment may be required to destroy all the pathogenic streptococci in a naturally infected milk than will be required to get rid of the organisms of any other disease known to be spread through milk.

Septic sore throat is generally considered as the only streptococcal disease occurring in widespread epidemics which is disseminated through the agency of milk. The organism causing this malady has always been found to be a hemolytic streptococcus and, judged by morphological and cultural standards, it appears to be a more or less specific type. The heat resistance of this organism under experimental conditions, as reported by various observers, varies within narrow limits and in no case has it been found to survive a temperature of 60° C. (140° F.) for thirty minutes. Some strains isolated by us last year were killed in milk at

60° C. in ten minutes. All of these recorded results, however, were carried out with milk, heated or raw, inoculated with a certain quantity of the organisms from cultures on artificial media. The organisms were present, therefore, in a free suspension unprotected by any covering of mucus or other substance. The mode of infection of milk with the organisms of this disease is now generally believed to be, in the majority of instances, through the medium of cows with infected udders. In such cases very large amounts of pus may be present in the milk and the streptococci are found closely associated with this pus. Clumps of organisms, no doubt, are frequently completely covered with mucus material and pus cells. That these coverings tend to protect the organisms from the effects of heat is indicated by the results of experiments described below.

In several epidemics of this disease, especially the earlier ones investigated in this country, no connection could be

Immediately	5 min.	10 min.	15 min.	20 min.	25 min.	30 min.
++++	+++	+++	++	++	+	+

shown between the outbreaks and infected cows. It was assumed, therefore, that direct infection of the milk had taken place from human throats. This must still be considered as a possibility though it is believed that such cases are exceedingly rare. Epidemics of considerable extent could be caused in this way only when multiplication of the inoculated organisms took place in the milk. That an occasional one of the very common cases of sporadic sore throat or tonsillitis, however, may be due to the ingestion in milk of particles of sputum or mucus containing streptococci seems very probable. Such mucus imbedded streptococci, we are convinced, may survive pasteurization at some of the lower temperatures in use. In support of this contention we offer the following experimental results. A sample of sputum from a case of asthma was obtained. It showed a slimy consistency and contained, practically in pure culture, a large number of hemolytic streptococci of the alpha type. A quantity of this

sputum was shaken with glass beads until it was well broken up and could be readily pipetted. A portion was diluted 1-10 with whole sterile milk and 1 c.c. of this mixture was then added to each of 8 tubes containing 4 c.c. of the same milk which had already been brought to the temperature of 61° C. One of these tubes was placed at once in cold water to act as a control. The rest were replaced in the carefully regulated bath at 61° C. At the end of each 5 minute period a tube was removed and placed in cold water. At the end of 30 minutes the two remaining tubes were removed. One c.c. from each tube was now plated out on horse blood agar and the plates incubated for 48 hours. The accompanying table gives the relative number of colonies on the different plates which may be taken as an approximate index of the effectiveness of the different treatments. The number on the control plate was very large, far too many to count, while only one of the two 30-minute plates showed a single colony.

The organism was now isolated from one of the plates and the next day a similar test was run, using as the source of the suspension 1 c.c. of a 24-hour broth culture. In this test none of the organisms survived longer than 15 minutes. The number of colonies on the control plate was fully as great as in the preceding test. The 15-minute plate showed only six. This organism, then, when a mere broth culture is suspended in milk, shows no greater heat resistance than has been reported by many observers for the streptococcus of epidemic sore throat, but when the suspension is made from a naturally infected sputum their resistance is markedly increased. It may be assumed that the same would hold true for a milk supply naturally contaminated with the septic sore throat streptococcus whether the contaminating organisms came from a cow with mastitis or from a human throat. A more detailed study of this question is now in progress the results of which will be published later.

The pathogenic streptococci constitute an exceedingly varied group of organisms. So widely do they differ, in fact, that as yet no features have been discovered upon which to base a usable classification. Compared with the number of investigations that have been made upon their cultural, morphological and pathogenic characteristics, studies on their resistance to heating can hardly be said to have been more than begun. Too much trust should not be placed, therefore, in the few tests that have been made, for it is quite conceivable that more extended and detailed investigations may bring to light strains differing as markedly in this respect as these are known to differ in other features. In fact a recent study by Beckwith and Eddie³ seems to be just such a case. These authors found that a virulent strain of streptococcus hemolyticus which had been transferred for several years by passage through rabbits resisted heating in milk at a temperature of 60° C. for twenty minutes and 65° C. for ten minutes. The same strain when cultivated in vitro instead of in vivo was even more resistant, some organisms remaining alive after exposure to 65° C. for thirty minutes.

As has been stated, the number of organisms in a suspension will determine to some extent the time necessary to destroy them at a given temperature, or, conversely, the temperature necessary for any given time period. This is an important point to remember in connection with the fact that milk supplies may be infected with streptococci in enormous numbers. In the case reported by us¹ at least one cow in a herd of fifty-one contributed milk containing 10,000,000 streptococci per c.c. Even supposing that this sample was diluted before it reached the consumer with the whole milk of the herd the count would still be great enough to constitute a heavy inoculation. Except under ideal conditions some multiplication is bound to take place and conditions could, and doubtless often do, exist

which would permit of very great increase in the numbers. Brown and Orcutt⁴ have reported a case in which the count from one-quarter of the infected cow's udder reached as high as 270,000,000 per c.c. Adding this milk to a large volume is comparable to the inoculation of media in the laboratory. Only immediate cooling to a temperature of 50° F. or below and maintaining that temperature would prevent rapid growth. In no other milk borne disease is the original inoculation believed to be as heavy as this.

The average user of milk has come to regard the term pasteurization as signifying a definite process which renders the milk entirely safe as regards communicable disease. Following the epidemic of sore throat in Portland last year the raw milk industry was threatened with ruin due to the desire of the people to have their milk pasteurized. No thought was given by them to the methods in use. They depended upon the health officials to see that safe standards were set. This must of necessity be the attitude of the people. The responsibility of the health official therefore is plain. It is clear, however, from a review of the standards in use in different cities that the term has not as yet been satisfactorily defined. Among scientific men who have given the question careful study it is generally agreed that a temperature of 145° F. applied for thirty minutes is a safe standard but that anything less is not safe. It is seen from the facts set forth in this paper that streptococcus infected milk may present such a combination of conditions as to make a complete destruction of the organisms by any milder treatment quite uncertain. We feel justified in urging that these standards shall constitute the minimum for all pasteurized milk.

REFERENCES

1. Benson and Sears. A Milk Borne Epidemic of Septic Sore Throat in Portland, Oregon. J. A. M. A., 1923, 80, p. 1608.
2. Jour. Inf. Dis., 1916, 8, p. 27.
3. To be published shortly in Jour. Lab. Clin. Med. We are permitted to use the article previous to publication through the courtesy of the senior author.
4. Jour. Exp. Med., 1920, 31, p. 49.