COME-UP TIME METHOD OF MILK PASTEURIZATION.

II. INVESTIGATION OF MILK PROPERTIES AND SOME PRELIMINARY BACTERIOLOGICAL STUDIES\(^1,\ 2,\ 3\)

R. B. Read, Jr., N. L. Norcross, D. J. Hankinson and Warren Litsky

Departments of Bacteriology and Public Health and Dairy Industry, University of Massachusetts, Amherst

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In recent years, there has been increased interest in higher temperatures for processing milk with a corresponding reduction in heating time. Simplicity of operation, improved flavor and keeping quality have been objectives in the utilization of these higher temperatures. With the use of higher temperatures, it appears practical to depend entirely on the lethality of the come-up and cooling portions of the cycle to achieve adequate pasteurization. This would enable one to completely eliminate the holding time requirement normally contained in the definition of high-temperature short-time (HTST) pasteurization. Since the amount of lethality from the cooling portion is relatively minor in this study, the investigation has been confined to the effect of the come-up portion of the heating cycle, thus the term “come-up time pasteurization” is used to describe this process in which there is no intended holding time.

The object of this study is to evaluate the effect of rapid heat treatment on the phosphatase enzyme, creaming, curd formation, bacterial destruction and flavor using the instrument described in a previous paper (1).

**Review of the Literature**

In recent years, new terms have been introduced which have replaced the term “Flash pasteurization” for the designation of any process involving pasteurization where the fluid is heated at rapid rates. Newer terms to designate this rapid heating to higher temperatures are “quick-time”, “no hold”, “no intended hold”, and “come-up time” pasteurization.

“Quick-time” pasteurization was a term introduced by Dahlberg, Holland, and Miner (2) in 1941 in which pasteurization of milk was carried out from \(169^\circ\) to \(177.5^\circ\) F. with the time interval above \(140^\circ\) F., varying from 5 to 24 seconds. It was found that a slightly better milk was produced by “quick-time” pasteurization than by vat pasteurization. No data were secured using pathogenic bacteria.

Tobias, Herreid and Ordal (3) heated milk in the Mallory small-tube heat exchanger and found that a temperature of \(188.34^\circ\) F. with a holding time of 2.36 seconds was found to give destruction of *M. freudenreichii* (MS66) equivalent to laboratory pasteurization at \(143^\circ\) F. for 30 minutes.

Barber (4) used the term “no hold” pasteurization in discussing various problems connected with the pasteurization of dairy products in short time periods.

Ball (5) emphasized the lethality contained within the come-up time portion of the heating curve. For this reason, the designation “come-up time” pasteurization was used in a previous paper (6) which described a type of pasteurization wherein virtually all the lethality from the heating cycle was derived from the heating portion of the cycle.

Specific types of equipment have been developed to accomplish rapid heating such as the Vacreator, Mallorizer, and the Roswell heater. Roberts, Blanton, and Warren (7), using the Vacreator, reported that phosphatase was destroyed at \(185^\circ\) F. and temperatures of \(195^\circ\) and \(200^\circ\) F. produced a milk comparable in keeping quality to vat pasteurized milk.

**Experimental**

Methods used for evaluation of rapid heat treatment of milk

Raw milk was obtained from the university herd, pumped into the holding tank of the instrument and forced through the small bore stainless steel tubing by air pressure. As the fluid passed through the tube, it was heated to the desired temperature by electrical resistance. Samples were cooled by collection of this fluid in bottles containing glass marbles which were refrigerated prior to their use. In all cases, the final temperature of the fluid was below \(185^\circ\) F. after collection in bottles. The bottles were immediately placed in ice water and refrigerated until used in the various tests.

Milk properties studied after heating were: (a) phosphatase, (b) pH, (c) creaming, (d) curd

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\(^2\) Contribution No. 1017 of the University of Massachusetts College of Agriculture Experiment Station.

\(^3\) Presented at the 42nd Annual Meeting of the International Association of Milk and Food Sanitarians, Inc., at Augusta, Georgia, October 2-4, 1955.
formation, and (e) flavor.

The phosphatase test was performed immediately after heating. A second test was performed after 24 hours to determine possible enzyme reactivation. The phosphatase determinations were made using the modified Scharer Laboratory method (8). Phenol standards were prepared according to the procedure outlined. A standard curve was prepared using a Cenco photometer with a red filter for measuring light transmission through the colored samples. A boiled milk control was used to adjust the light transmittancy to 100 percent.

The pH of the milk was determined immediately before and after heating. The pH determinations were made using a Beckman Model H pH meter. After heat treatment, the samples under test were placed in 100 ml. graduates and refrigerated for 24 hours. At this time, the cream line readings were taken and recorded as per cent decrease in cream line as compared to the unheated control.

A sample of remnin obtained from Chr. Hansen Laboratories, Milwaukee, Wisconsin, was diluted 1:25. One cc. of this dilution was added to each 6" x 3/4" test tube in a series and 5 ml. of heat treated milk were added. Incubation was carried out at 86°F. At intervals of 1 min., a tube was inverted and checked for clotting. The curd formation end point was taken as that time when the clot would not slip down the barrel of the inverted tube. Controls were run at 148°F, for 30 minutes and 160°F for 30 minutes. The experimentally heated samples were compared with the controls.

The samples were examined organoleptically at room temperature for evidence of heated flavor. Flavor was evaluated as follows: no heated flavor; slight heated flavor; definite heated flavor; pronounced heated flavor.

Methods used to evaluate destruction of vegetative cells of bacteria

The organism MS 102 is an unidentified micrococcus which has been used in several heat resistance studies. This organism was grown on N-Z Case medium of the following composition:

<table>
<thead>
<tr>
<th>Component</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yeast extract</td>
<td>1.0 gram</td>
</tr>
<tr>
<td>N-Z Case (Sheffield)</td>
<td>5 gram</td>
</tr>
<tr>
<td>Glucose</td>
<td>5 gram</td>
</tr>
<tr>
<td>K2HPO4</td>
<td>4 gram</td>
</tr>
<tr>
<td>KH2PO4</td>
<td>1 gram</td>
</tr>
<tr>
<td>Agar</td>
<td>1.5 gram</td>
</tr>
<tr>
<td>Distilled water</td>
<td>100.0 gram</td>
</tr>
<tr>
<td>pH 7.0, sterilization at 15 lbs.</td>
<td>20 minutes</td>
</tr>
</tbody>
</table>

The transfer schedule for this organism was as follows. Daily transfers of the culture were prepared and incubated at 37°C for 24 hours; the slants were then stored in the refrigerator for 48 hours. A transfer was made from the refrigerated slant, and incubated at 37°C for 24 hours and then used as the test culture. After growth, the organisms were washed from the medium with approximately 10 ml. of sterile distilled water. The culture suspension was then aseptically filtered through Reeve Angel Filter paper No. 235, to remove large clumps of bacteria. The suspension was then checked for optical density and a suitable aliquot used to inoculate the suspending fluid.

MS 102 was found too heat resistant to accomplish 100 percent kill below 100°C, at 0.25 sec. heating time. With the instrument set to heat to 97°C in 0.25 second, it was found that 19 percent of the samples taken demonstrated growth of MS 102. Accordingly, the 99.9 percent destruction level was investigated. This was done by inoculating 10 ml. of fluid as it left the instrument into 90 ml. of sterile water. Dilutions of 1:10, 1:100, and 1:1000 were prepared. All samples were taken in triplicate. The recovery medium was the N-Z Case medium. Counts were done after 48 hours of incubation at 37°C. The 99.9 percent destruction point was computed from the initial count.

Determination of heating curve

Multiple thermo couples were installed along the heating tube so that the thermocouple was exposed directly to the fluid in the tube. The heating curve of the tube was determined by taking temperatures of the fluid at intervals of one foot while the tube was under actual operating conditions. Heating curves were determined for intervals of 10°F. from temperatures of 150 to 200°F, using 0.25 and 0.50 sec. come-up time. All determinations were made using milk as the fluid heated.

Results

Phosphatase

Phosphatase studies were performed on samples immediately after heat treatment and were repeated after 24 hours at 46.4°F, to check for reactivation. (9, 10, 11) Using 2.3 micrograms of phenol as a standard, it was found that with a preheat temperature of 135°F, and a heating time of 0.25 sec., phosphatase was inactivated at a mean temperature of 182.4°F. Results are shown in Table 1.

Samples which showed 2.3 micrograms of phenol immediately after heating were re-examined after 24 hrs. at 46.4°F. It was found that reactivation did occur in these samples. However, results were rather erratic and are shown in Table 1.

A similar study was conducted using a heating time of 0.5 seconds. The mean temperature of phos-
of Mattick and Hallet (12) who found that a moderate amount of heating (up to 141°F, for 30 min.) reduced the coagulation time when compared to raw milk. On the other hand, in their studies, heat treatment of 145 to 151°F. for 30 minutes yielded coagulation times about the same as for raw milk. Furthermore, heat treatments above 155°F. for 30 min. gave coagulation times which increased progressively with higher temperatures when compared with raw controls. In this study, with a heating time of 0.5 sec., it was found that in no case did the coagulation time exceed that of raw milk. Results of a typical series are summarized in Table 5.

**Flavor**

With heating times varying from extremes of 0.14 to 0.9 sec., it was found that no heated flavor was detected in milk heated to as high as 212°F., provided that the milk was cooled rapidly. Results of many tests have indicated that milk processed in this manner is comparable or better than vat pasteurized milk from a flavor standpoint.

**Bacteriological studies using MS 102**

The temperature necessary for 99.9 percent destruction...
The heating curve in the stainless steel heating tube. Averages of these trials were determined and the results for heating times of 0.25 and 0.5 sec. are shown in Figures 1 and 2.

The heating curves are essentially linear for the 0.25 and 0.5 sec. heating times.

**DISCUSSION**

Results of this study indicate that laboratory pasteurization of milk can be accomplished using heating rates as short as 0.25 sec. heating time with no intended holding period. Calculations of the holding time in this study involve a consideration of the time required for the milk to pass through the stainless steel valve at the terminal end of the holding tube. This time is approximately 0.05 sec., and is therefore part of the total heat applied and is in addition to

Many trials were made to determine the nature of an unidentified micrococcus* was determined for heating times of 0.25 and 0.5 sec. using milk as the vehicle. The mean temperature for a series of seven trials with 0.25 sec. heating time was 191.9°F, whereas a series of twelve trials using a 0.5 sec. heating time yielded a mean temperature of 190.6°F for the 99.9 percent destruction level. Results are shown in Tables 6 and 7.

**Heating curve determination**

Many trials were made to determine the nature of

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*Obtained from Dr. Franklin Barber of the National Dairy Products Research Laboratories, Oakland, L. I.
the reported 0.25 or 0.5 sec. come-up time. Following heating, the fluid is collected and cooled either by passing over pre-cooled glass marbles or by inoculation into a pre-cooled fluid. Since thermocouple studies have shown that the temperature of the fluid as measured in the collecting flask has never been above 135°F., the lethal effect of the cooling portion of the cycle was considered to be much less than the heating time and therefore was not evaluated separately. However, any lethality in the cooling cycle is in addition to the reported 0.25 or 0.5 sec. come-up time.

Phosphatase inactivation was found to occur at a point below the 99.9 percent destruction level of MS102. Studies on bacteria other than MS102 completed in this laboratory suggest, however, that the 99.9 percent destruction level of MS102 requires a temperature considerably higher than most other organisms commonly associated with milk. These studies on other bacteria will be published in a subsequent paper. Accordingly, if the phosphatase test is used as an indicator of adequate pasteurization, the observed reactivation of the phosphatase enzyme must be taken into consideration, using this type of heating.

Creaming studies indicate a severe loss in cream line especially with higher temperatures. The heating tube used is of small bore and the fluid velocity is such that highly turbulent flow is produced as calculated by Reynolds' Number. The turbulence probably produces a homogenizing effect on the milk. Perhaps creaming would not be reduced so dramatically in a standard HTST machine modified for a come-up time process, since the milk would not be subjected to the same forces between the conventional plates of the HTST unit as in the small bore tube.

Curd formation, flavor, and pH determinations indicate that this rapid heating process can produce a milk either as good or perhaps slightly better than the standard vat process.

The instrument used in this study is intended only for laboratory investigation of the effects of rapid heat on milk properties and the thermal resistance of bacteria. It is felt that data obtained from this machine may be applied to the commercial HTST unit which is modified by the elimination of the holding tube. Actual studies on the commercial HTST pasteurizer will be necessary to show the feasibility of more rapid heating and the extent of modification necessary.

**Conclusions**

1. Phosphatase is inactivated at temperatures of 182.3°F. and 178.5°F., for heating times of 0.25 and 0.5 sec., respectively.
2. No significant changes in pH were produced by this process.
3. Creaming was markedly impaired by this process at temperatures over 185°F.
4. Curd formation tests demonstrated no marked protein denaturation from this heating.
5. Flavor tests demonstrated a heated flavor either comparable to or less than vat pasteurized milk.
6. The organism MS102 was 99.9 percent destroyed at temperatures of 191.9°F. and 190.6°F., for heating times of 0.25 and 0.5 sec., respectively, using milk as the vehicle.
7. The heating curve for the instrument used is essentially linear.

**References**