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THE PERPETUATION OF PURE CULTURES FOR BUTTER STARTERS.

By E. F. PERNOT.

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THE PERPETUATION OF PURE CULTURES FOR BUTTER STARTERS.

BY E. F. PERNOT

The introduction and adoption of the cream separators has opened a new field of possibilities for controlling the flavor of butter, by the use of pure cultures of selected varieties of organisms, to such a degree of certainty that many of the leading dairies now use starters to assure a uniformity of flavor in their products. Some of the methods in vogue at present are crude, but as we progress new methods of perpetuating pure cultures will be introduced and modified to meet the requirements. For a starter most of the dairies procure a culture of organisms from firms who prepare them for the market. Upon receipt of the culture the content of the bottle is emptied into about two to three quarts of pasteurized skimmed milk. This is kept at a temperature of 60 to 70 degrees F. until about .4 of 1 per cent acidity is reached; it is then added to five gallons of pasteurized skim milk, and kept at a temperature of 60 to 70 degrees F. until an acidity of .3 to .4 per cent is reached, and then added to cream in an amount equalling from five to ten per cent; the cream is then ripened to an acid reaction of .5 to .6 per cent. To perpetuate, two quarts are taken of the starter and added to skimmed milk as before.

It is claimed that in about one week, starters handled in this way require being renewed by fresh cultures, but this may be obviated by the simple method herein described, and a pure culture can be perpetuated so long as reasonable care is exercised.

Before entering upon a description of a new method, it is well to consider some of the weak points in the old ones. If we continue replanting a culture in milk which is only pasteurized, it will eventually become so contaminated with undesirable germs as to overcome the flavor producing qualities of the starter, because pasteurized milk is but partially sterilized, and contains more or less spores and germs which have resisted the single pasteurizing temperature or even a higher one. They multiply and soon become as

numerous as those originally planted. In this way the starter loses its virtue and is but little or no better than milk soured in the open air. The first time that the starter is used, it is comparatively pure, but after that, every time it is added to fresh pasteurized milk more foreign germs develop in it and prevent the desirable ones from exerting their influence until finally the starter becomes useless. In the preparation of anti-toxin, vaccine, and serum; in fact all bacteriological work, the strictest precautions are exercised to prevent contamination or mixtures of germs, lest the products become valueless. Most of the dairy work is of a bacteriological nature and if the highest results are to be obtained by the use of pure cultures, then at least some bacteriological methods must be employed. The following is a simple and effective way of perpetuating pure cultures for starters and is within the capabilities of any modern dairyman.

Take twelve or more one pint milk bottles, wash thoroughly with hot water and soap, steam them out to remove all traces of the soap, then fill half full with fresh milk, skimmed with a separator. Take some cotton batting and plug the necks of the bottles with good substantial plugs, neither too loosely nor too tightly. The plug should reach at least one and one-half inches below and one-half inch above the top of the bottle neck. Never wet the cotton or it will become useless.

Through the center of one of these plugs, pass a glass tube sufficiently long to reach to within one inch of the bottom of the bottle and extend an inch or more above the cotton plug. The lower end of this tube is partly closed so as to hold milk in it by capillarity. The upper end is filled with cotton for one inch in length and then the rubber bulb from a medium dropper is slipped on the tube. (See Fig. 1). The cotton in the tube is to prevent any germs from entering the tube although they may find their way into the bulb.

A well made wooden box with a closely fitting cover is then provided and connected with a steam pipe and acts as a steam chest; the bottles are then placed in the box, lid replaced and steam turned on. To ascertain the temperature, place another bottle in the box, fill it with water or milk and place a thermometer in it. The temperature of the milk may thus be obtained and should be raised to 180 degrees F. or higher, and maintain this heat for twenty minutes. The cover is then taken off the box and the cotton plugs will dry



FIGURE 1

immediately. This operation should be repeated three times, allowing twenty-four hours to elapse between heatings; the milk will then be sterile and in an ideal condition for germs to grow in when planted. Milk so treated will remain sterile for an indefinite time. Another means of sterilizing the milk intermittently, is to use a steam cooker or Arnold steam sterilizer. Either of these may be used to good advantage in dairies where steam is not available, as they may be placed on a stove and require no further attention other than to note the time that the bottles remain in them, which ought not to be more than ten minutes after the milk has acquired the temperature of the sterilizer, which is 212 degrees F. A fixed time cannot be given here for the bottles to remain in the sterilizer, as it will vary with the amount of heat applied. Still another method is to sterilize the milk in one operation by the use of steam under a pressure of ten pounds, which gives a temperature of 240 degrees F. It should be kept at this heat for from five to ten minutes, but the milk is not as suitable for culture purposes as when sterilized at a lower temperature, besides, an autoclave or steam chest capable of withstanding such a pressure is required and is seldom found in a dairy. While sterilizing the milk bottle with the pipette in it, the lower end of the pipette should be raised out of the milk to avoid pressure sending the milk up the tube and wetting cotton in the upper end or filling the bulb. After sterilizing the milk of course, the pipette must be lowered into it again.

The next step is to procure a suitable culture. If bought from a dealer, the cork must be carefully removed without allowing any particles of wax to fall into the bottle; the neck is then passed through the flame of an alcohol lamp to sterilize it, and contents of the bottle emptied into the bottle containing the pipette. To do this properly, twist the cotton plug to loosen it, draw it out gently and just high enough to insure its not touching the other bottle while it is being emptied; replace it immediately and shake the bottle slightly to mix the culture with the milk. After twenty-four hours this gives the first starter. To perpetuate it, withdraw the cotton plug from a sterile bottle of milk and at the same time withdraw the cotton plug and pipette together and transfer them to the other bottle quickly and absolutely without touching them to anything. When in place the pipette bulb is squeezed, which empties the milk which it has carried from the first bottle into the fresh milk, thus inoculating a fresh supply of milk with the organisms

desired. The contents of the bottle which had the pipette will then be ready with a pure culture, to be emptied into the required amounts of pasteurized milk or cream as it is customarily done.

There are a few points about this method which must be rigidly observed to insure success, otherwise it is much more simple than it appears in print.

The inoculation of one bottle from another must be quickly done and *positively* without touching the pipette or plug with anything else. Particles of cotton or dust *must not* fall into the bottles. The milk *must* be sterile before inoculating. Cultures of reliable purity must be used for the first inoculation. As fresh milk as possible should be used for culture media lest the products of bacteria which had acted upon older milk should act as enzymes and cause disturbances later. Very fresh milk should be used for culture media, not only because it contains less germs, but less germ products which may have a peptonizing action. Fresh milk also contains its full food value for germ cultures. The mixed skim milk from patrons of a dairy will not give such good results, because it is much more difficult to sterilize and is robbed of some of its normal constituents.

During the dairy short course last winter, this method was carried on by the students with good success. Plate tests were made at regular intervals and the starters were found to maintain their original pure cultures throughout the term. The butter which was made with the use of these cultures, was carefully scored by Professor Kent, of the dairy department. The data relative to the several churnings is given in the following table:

Churnings of February 18, 1904.
Each consisting of 17 pounds of Cream containing 27 per cent Fat.

Culture used.	Acidity. Per cent.	Churn Temp. Degrees	Time of Churning. Minutes.	Per cent Fat in But- termilk.	Flavor Score.	
					At 1 week.	At 3 weeks.
Check.	.60	59	16	.3	40	35
A69	59	25	.2	42	37
B57	59	23	.2	41	35
C59	59	30	.2	41	38
D58	59	28	.2	41½	41
E58	59	19	.3	40½	41
F57	59	27	.15	39	40

Churnings of February 20.
Each consisting of 19 pounds of Cream containing 27 per cent Fat.

A60	58	23	.2	41	33
B57	58	23	.3	40	38
C60	58	27	.2	42	35
D61	58	24	.2	43	41
E60	58	23	.2	40	41
F60	58	30	.2	41	40

Churnings of February 23.
Each consisting of 10 pounds of Cream containing 25 per cent Fat.

Check.	.66	62	14	.4	40	40
A70	62	15	.5	41	36
B69	62	15	.4	41½	40
C70	62	16	.4	41½	40
D69	62	17	.5	42½	39
E67	62	12	.5	40	39
F70	62	18	.6	41½	39

Churnings of February 25.
Each consisting of 18 pounds of Cream containing 28 per cent Fat.

Check.	.68	60	15	.4	40½	32
A64	60	18	.4	39	32
B66	60	21	.4	41	34
C63	60	30	.2	42	39
D67	60	23	.4	41½	33
E61	60	24	.2	41	35
F66	60	28	.3	40	36

45 is considered a perfect score for flavor in this table. As indicated by the table the several lots of butter were scored at one week and again at three weeks after making. It is interesting to note that in two cases there was a slight improvement in flavor at the time of the second scoring. The low scores for the second scoring of the churning of Feb. 25 are probably due to the samples being examined when four, instead of three weeks old as in the other cases.

The samples were kept in a refrigeration box but without ice, the room temperatures varying from 50 to 70 degrees F.