

A comparison of total bacteria counts of raw and pasteurized milk

Donald H. Holmes

Follow this and additional works at: <http://digitalcommons.butler.edu/botanical>

The Butler University Botanical Studies journal was published by the Botany Department of Butler University, Indianapolis, Indiana, from 1929 to 1964. The scientific journal featured original papers primarily on plant ecology, taxonomy, and microbiology.

Recommended Citation

Holmes, Donald H. (1950) "A comparison of total bacteria counts of raw and pasteurized milk," *Butler University Botanical Studies*: Vol. 9, Article 19.

Available at: <http://digitalcommons.butler.edu/botanical/vol9/iss1/19>

Butler University
Botanical Studies
(1929-1964)

Edited by

Ray C. Friesner

The *Butler University Botanical Studies* journal was published by the Botany Department of Butler University, Indianapolis, Indiana, from 1929 to 1964. The scientific journal featured original papers primarily on plant ecology, taxonomy, and microbiology. The papers contain valuable historical studies, especially floristic surveys that document Indiana's vegetation in past decades. Authors were Butler faculty, current and former master's degree students and undergraduates, and other Indiana botanists. The journal was started by Stanley Cain, noted conservation biologist, and edited through most of its years of production by Ray C. Friesner, Butler's first botanist and founder of the department in 1919. The journal was distributed to learned societies and libraries through exchange.

During the years of the journal's publication, the Butler University Botany Department had an active program of research and student training. 201 bachelor's degrees and 75 master's degrees in Botany were conferred during this period. Thirty-five of these graduates went on to earn doctorates at other institutions.

The Botany Department attracted many notable faculty members and students. Distinguished faculty, in addition to Cain and Friesner, included John E. Potzger, a forest ecologist and palynologist, Willard Nelson Clute, co-founder of the American Fern Society, Marion T. Hall, former director of the Morton Arboretum, C. Mervin Palmer, Rex Webster, and John Pelton. Some of the former undergraduate and master's students who made active contributions to the fields of botany and ecology include Dwight W. Billings, Fay Kenoyer Daily, William A. Daily, Rexford Daudenmire, Francis Hueber, Frank McCormick, Scott McCoy, Robert Petty, Potzger, Helene Starcs, and Theodore Sperry. Cain, Daubenmire, Potzger, and Billings served as Presidents of the Ecological Society of America.

Requests for use of materials, especially figures and tables for use in ecology text books, from the *Butler University Botanical Studies* continue to be granted. For more information, visit www.butler.edu/herbarium.

A COMPARISON OF TOTAL BACTERIA COUNTS OF RAW AND PASTEURIZED MILK

By DONALD H. HOLMES

The purpose of this study was to compare the total bacteria count in samples of raw and pasteurized milk from the Golden Guernsey Association, of Indianapolis, Ind., using both the Breed-Prescott direct microscopic method and the standard agar plate method. In addition, a record of the types of organisms observed in the direct microscopic count was made for each sample.

Producers, desirous of checking their raw milk supply, usually ask for either plate or direct counts but seldom request both counts on the same sample. Results of counts from the two methods often appear quite confusing and contradictory so that the producer is unable to decide which count to accept as correct. However, numerous research workers have made such comparison counts (5) and their results show direct count to plate count ratios varying from 44:1 to 4:1. The latter ratio is commonly accepted in dairy analysis work as a "normal" ratio. It is by no means a hard and fast one as will be observed later in this paper. Individual counts can show wide deviations from this ratio. Some reasons for the differences in counts are: (1) Factors affecting the plate count: (a) failure of all organisms to grow in media used; (b) failure of all organisms to grow at incubation temperatures used; (c) single colonies developing from clumps of organisms; (d) sampling and technique errors. (2) Factors raising or lowering direct count: (a) failure of all organisms to stain, or staining of dead organisms; (b) sampling and other technique errors. Both plate and direct methods of milk examination are subject to numerous errors in addition to those given above.

MATERIALS AND METHODS

The large supply of raw milk samples necessary was readily obtainable at the Moseley Laboratory in Indianapolis. The samples chosen were all from Guernsey cows and were produced on farms belonging to the Golden Guernsey Association. This milk has always shown a consistently low raw and correspondingly lower pasteurized count.

A raw sample, when received at the laboratory, was first smeared on a standard slide using a platinum wire loop to apply the milk and resulting in a smear of one square centimeter area. The sample was then split into two parts, one portion to be pasteurized at 143-144° F. for 30 minutes, then cooled and plated at a dilution of 1/100 and also smeared again on a different slide using the same procedure as the first smear. The other portion was plated raw immediately at dilutions of 1/100 and 1/10,000. The pasteurized sample was then placed in a cooler for approximately four hours, after which it was again smeared on a third slide. All three slides were then stained with methylene blue solution and read, using the oil immersion lens. The plated samples were allowed to cool before inverting and placing them in the incubator at 37° C. for 48 hours. At the end of the incubation period they were counted on a Quebec counter.

As each smear was counted, a record was kept of the types of organisms observed. The organisms (see table 1) were separated into individual cocci, individual rods, diplococci, rods in clumps, streptobacilli, streptococci, and staphylococci. One hundred fields were read per smear and the figure obtained multiplied by 3000 to get the total organisms present per milliliter of milk. This counting procedure was followed on all smears except a few showing very high counts on the first few fields examined. In such cases 30 fields were counted and the total multiplied by 10,000.

Each plate was counted after the incubation period and the total number of colonies found was multiplied by the dilution factor. In the case of the raw counts, when two plates were made, one at 1/1000 and one at 1/10,000, only the plate having between 30 and 300 colonies was counted and this figure multiplied by the dilution factor used. In case both plates had between 30 and 300 colonies, the average was computed and accepted as the proper total count for the sample.

OBSERVATIONS

The milk used in this study is one of the cleanest milks in use today. It is produced and handled with special care in order to keep the bacteria count low and the product clean. For this reason these samples all show quite low counts, some of the raw samples even being completely free of organisms on the Prescott-Breed smears. However, these same raw samples, when plated, showed counts from 400 to 23,000. On the smears of raw milk, staphylococci were most

abundant, then streptococci, individual cocci, rods in clumps, streptobacilli, individual rods, and diplococci, in decreasing abundance. The staphylococci also survived pasteurization better than any other organism, followed by individual cocci, streptococci, individual rods, rods in clumps, diplococci and streptobacilli. The third smear, made four hours later from the same pasteurized sample of milk, showed staphylococci predominant, streptococci next, closely followed by individual cocci, individual rods, then rods in clumps, diplococci and streptobacilli (table I).

Using the total counts (see table I) found on these samples by the plate and direct methods, the following ratios were obtained:

<u>Raw direct count</u>	3.05
Raw plate count	1
<u>Pasteurized direct count</u>	6.21
Pasteurized plate count	1
<u>Raw plate count</u>	13.38
Pasteurized plate count	1
<u>Raw direct count</u>	6.90
Pasteurized, smeared 4 hours later	1
<u>Raw direct count</u>	6.56
Pasteurized direct count	1
<u>Pasteurized direct count</u>	1.05
Pasteurized, smeared 4 hours later	1

Staphylococci were found most often on all three smears: raw, pasteurized, and the pasteurized-smeared-four-hours-later. Their presence is often an indicator of utensil contamination, (3). Streptococci were present in greater numbers than all other organisms except staphylococci on raw smears and the smears made four hours after pasteurization, but were third on the smears made immediately after pasteurization. Individual cocci were second on the pasteurized smears and third on the other two. These organisms are associated with utensil contamination and poor cooling. No particular significance can be attached to the other organisms present in these smears. The counts were low under all conditions.

The ratio of raw-direct to raw-plate count was 3.05:1. This is much better than the 4:1 ratio considered "normal" in the industry. In this study all ratios were obtained by using the pertinent averages of all counts on the total of 142 samples used. The ratio of pasteurized-direct to pasteurized-plate count was 6.21:1. This ratio is considerably different from the raw ratio and was probably influenced by the staining of dead cells in the pasteurized-direct smears.

The ratio of raw-plate to pasteurized-plate count was 13.38:1. This ratio is much higher than the pasteurized-direct to pasteurized-plate since only living cells in the pasteurized milk were to be compared with those of the raw milk. The ratio of raw-direct to pasteurized-direct count was 6.56:1 and raw-direct to pasteurized-smeared-four-hours-later was 6.90:1.

Finally, the ratio between the pasteurized-direct count and the pasteurized-smeared-four-hours-later count was 1.05:1. This ratio is insignificant, but to the extent that it may have significance, it indicates that some of the organisms stained in the smears prepared immediately following pasteurization were not stained in the smears prepared four hours after pasteurization. Immediately after pasteurization these organisms, although dead, possess the ability of taking a staining preparation, but after a period of time, approximately four hours in this case, they were no longer stainable and consequently failed to appear on the smear, (1, 2).

CONCLUSIONS

1. In raw Golden Guernsey milk, the most common organisms found are of the staphylococcus group, many of which survive pasteurization and are the organisms most commonly found in the pasteurized samples.

2. The "normal" 4:1 direct microscopic count to plate count ratio is seldom attained. The raw ratio in this case being 3.05:1 and the pasteurized ratio 6.21:1.

3. When a milk sample is smeared and stained immediately after pasteurization, numerous dead bacteria as well as bacteria which have survived pasteurization are stained and counted. When the pasteurized sample is allowed to stand for a period of time (four hours in this case) some of the bacteria which stained before fail to stain.

4. The ratio of pasteurized and smeared-immediately to pasteurized and smeared-four-hours-later was 1.05:1. This ratio would probably have been higher and more significance could have been attached to it if the original raw milk samples had shown higher counts.

LITERATURE CITED

1. HAMER, B. W. Dairy Bacteriology. John Wiley and Sons, Inc. 1948.
2. HILL, CHARLES F. A direct microscopic study of the effect of temperature on bacterial counts of milk. *Butler Univ. Bot. Stud.* 3:139-152. 1948.
3. LAZARUS, N. E. Quality Control of Market Milk. Olsen Pub. Co. 1935.
4. SOMMER, H. H. Market Milk and Related Products. Olsen Pub. Co. 1946.
5. TANNER, F. W. Microbiology of Foods. Garrard Press. 1944.

TABLE I

Treatment	Direct Counts—Averages of 142 samples							Total per ml.	Av. 142 Samples Plate Count
	Individual Cocci	Rods	Diplococci	Rods in Clumps	Streptobacilli	Staphylococci	Streptococci		
Raw	13.5	4.13	1.15	9.85	3.72	97.88	33.92	528,206	173,300
Pasteurized	4.0	1.74	0.47	0.52	0.25	16.76	3.11	80,472	12,954
Smeared 4 hours after pasteurization	3.10	1.33	0.49	0.54	0.28	16.35	3.62	76,500	—