

SEPTEMBER, 1971

Vol. 34 P. 423-466 No. 9

Journal of

MILK and FOOD TECHNOLOGY

Official Publication

International Association of Milk, Food and
Environmental Sanitarians, Inc.

Flash in the pan.



If your pans look like
this after cleaning



...and you want them
to look like this,

we'll show you how it's done...in a flash.

We surprised a food processor in California by removing stubborn stains from aluminum food tubs like those shown here.

Our man out there cleaned them in a flash with one of the special cleaners from our B-K® line of cleaners and sanitizers. A quick soak and rinse did the job.

Your local Pennwalt representative can show you how to make even the most stubborn stains vanish from your plant equipment and utensils. If you've got an especially tough cleaning problem that baffles him (possible, but not likely), our scientists at Pennwalt's Technological Center will find the answer. Without charge.

Contact us now. You have nothing to lose but your stains. Dairy and Food Department, Pennwalt Corporation, Three Parkway, Philadelphia, Pa. 19102.

 **PENNWALT**
CLEANERS & SANITIZERS

To Our Contributors

The success of this 58th annual meeting could not have been achieved without your generous assistance.

Alta Dena Dairy
Alpha Beta Acme Markets
Associated Dairymen
Avoset Food Corporation
Babson Bros. Co. (Surge)
Beaver Mfg. Co.
Borden's
Bou-Matic Milkers
Brinderson DeLaval Sales
California Milk Producers Advisory Board
Challenge Cream & Butter Ass'n.
Chace C. Hoffman (Neilson Creamery)
Chino Valley Cattle Co.
Clay Equipment Co.
Coast Grain Co.
Consolidated Dairy Prod. Co.
Continental Dairy Supply
Creamery Package Division St. Regis
Dairy Council of California
Dairymen's Coop. Creamery Ass'n.
Dairy Research Prod., Inc.
Danish Creamery Ass'n.
Dean Milk Co.
DeLaval Separator Co.
Diversey Chemical Co.
Ex Cello Corp.
Food Filters Corp.
Fults Chemical, Inc.
Globe Veterinary Supply
Golden Arrow Dairy
Hites Dairy
International Paper Co.
Jerseymaid Milk Products
California Cheese Assn.
California Cheese Co.
Standard Cheese Co.

Vella Cheese Co.
Kendall-Fiber Products Div.
Kings Country Creamery Ass'n.
Kings Country Truck Lines
Klenzade
Kennel K 999 (L.A. Yellow Dogs)
Kruse, O. H. Grain & Milling)
Knudsen Corp.
League of Calif. Milk Producers Ass'n.
Lily-Tulip Div.
Los Angeles Mutual Dairymen's Ass'n.
Lucky Stores, Inc.
Los Banos Dairymen's Ass'n.
Meyenberg Milk Products
Mulligan Sales
Penny-Newman Grain Co.
Protected Milk Prod. Ass'n.
Real Fresh Milk, Inc.
Ralph's Grocery Co.
Rehrig Pacific Co.
Ross-Holm
Safeway Stores Milk Dept.
San Francisco Milk Industry
Schweitzers Metal Fabricators
Sep-Ko Chemical, Inc.
Sunshine Dairy Sales Yard
Superior Milk Products, Ass'n.
The Dairyman
Thrifty Drug Stores, Inc.
Top-Line Corp.
Transflow Tubing & Inflatons
United Dairymen's Ass'n.
West-Argo Chemical, Inc.
Western Dairy Journal
Weyerhaeuser Co.
Wilsey Bennett Foods, Inc.
Zan Mfg. Co.

DAIRY INSTITUTE OF CALIFORNIA

Adohr Milk Farms
Arden Farms Co.
Berkeley Farms
Brookside Dairy
Carnation Company, Los Angeles
Carnation Company, Oakland
Crystal Cream & Butter Co.
Driftwood Dairy
Edgemar Farms
Favorite Foods, Inc.
Foremost Dairies, Inc.
Foster Farms All-Jersey Dairy
Green Glen
Meadow Gold Dairies

McCull's Dairy Products Co.
Pellissier Dairy Farms
Peninsula Creamery
Petaluma Cooperative Creamery
Producers Dairy Delivery Co.
Quaker Maid Dairy
Scott Brothers
Shady Grove Dairy
Spreckels Dairies, Inc.
Sunshine Farms
Sun Valley Dairy
Wayne's Dairy
Western Holstein Farms, Inc.

MILK and FOOD TECHNOLOGY

INCLUDING MILK AND FOOD SANITATION

Official Publication

International Association of Milk, Food and Environmental Sanitarians, Inc.
Reg. U. S. Pat. Off.**OFFICERS AND EXECUTIVE BOARD***President*, ORLOWE M. OSTEN, Minn.
Dept. of Agric., 517 State Office
Bldg., St. Paul, Minn. 55101.*President-Elect*, WALTER F. WILSON,
County Los Angeles Health Dept.,
313 N. Figueroa St., Los Angeles,
Calif. 90012.*First Vice-President*, EARL O. WRIGHT,
116 Dairy Industry Bldg., Iowa
State U., Ames, Ia. 50010.*Second Vice-President*, P. J. SKULBOR-
STAD, 2100 South York Rd., Oak-
brook, Ill. 60521.*Secretary-Treasurer*, RICHARD P. MARCH,
118 Stocking Hall, Cornell Univ.,
Ithaca, N. Y. 14850.*Junior Past-President*, DICK B. WHITE-
HEAD, Miss., State Board of Health,
P. O. Box 1700, Jackson, Miss.
39205.*Senior Past-President*, MILTON E. HELD,
910 Lupin Way, San Carlos, Calif.
94070.**Editors**DR. ELMER H. MARTH, *Editor*, Dept.
of Food Science, University of Wis-
consin, Madison, Wis. 53706.H. L. THOMASSON, *Executive Secretary
and Managing Editor*, Box 437,
Shelbyville, Indiana 46176.**Editorial Board**

C. A. ABELE -----Evanston, Ill.
H. S. ADAMS -----Indianapolis, Ind.
J. A. ALFORD -----Beltsville, Md.
E. F. BAER -----Washington, D. C.
F. W. BARBER -----Glenview, Ill.
F. L. BRYAN -----Atlanta, Ga.
W. J. DYER -----Halifax, N. S.
J. C. FLAKE -----Washington, D. C.
H. R. GRONINGER -----Seattle, Wash.
L. G. HARMON -----East Lansing, Mich.
N. F. INSALATA -----Battle Creek, Mich.
C. K. JOHNS -----Ottawa, Ont.
H. KOREN -----Terre Haute, Ind.
R. T. MARSHALL -----Columbia, Mo.
S. A. MATZ -----Villa Park, Ill.
E. M. MIKOLAJCIK -----Columbus, Ohio
J. C. OLSON, JR. -----Washington, D. C.
R. L. OLSON -----Albany, Calif.
Z. J. ORDAL -----Urbana, Ill.
J. W. PENCE -----Albany, Calif.
H. J. PEPLER -----Milwaukee, Wis.
D. S. POSTLE -----Ithaca, N. Y.
W. D. POWRIE -----Vancouver, B. C.
R. B. READ, JR. -----Washington, D.C.
G. W. REINBOLD -----Ames, Iowa
G. H. RICHARDSON -----Logan, Utah
R. L. SAFFLE -----Athens, Ga.
W. E. SANDINE -----Corvallis, Oregon
F. M. SAWYER -----Amherst, Mass.
D. F. SPLITTSTOESSER -----Geneva, N. Y.
C. E. SWIFT -----Philadelphia, Pa.
B. A. TWIGG -----College Park, Md.
C. VANDERZANT -----College Station, Texas
W. G. WALTER -----Bozeman, Mont.
H. B. WARREN -----Omaha, Nebr.
K. G. WECKEL -----Madison, Wis.
J. C. WHITE -----Ithaca, N. Y.
H. WISTREICH -----Chicago, Ill.
E. R. WOLFORD -----Puyallup, Wash.
E. A. ZOTTOLA -----St. Paul, Minn.

The Journal of Milk and Food Technology is issued monthly beginning with the January number. Each volume comprises 12 numbers. Published by the International Association of Milk, Food and Environmental Sanitarians, Inc. with executive offices of the Association,

Vol. 34

September, 1971

No. 9

Isolation of Salmonellae from Naturally Contaminated Dried Milk Products II. Influences of Storage Time on the Isolation of Salmonellae <i>Bibek Ray, J. J. Jezeski, and F. F. Busta</i>	423
Sanitation and Water Usage in the Processing of Sour Cherries <i>Y. D. Hang, D. L. Downing and D. F. Splittstoesser</i>	428
Temperature Profiles in Holding Tubes of Milk and Egg Pasteurizers <i>R. W. Dickerson, Jr., and R. B. Read, Jr.</i>	431
Immunoglobulins in Skimmilk and Model Systems and their Effect on <i>Bacillus Cereus</i> <i>E. M. Mikolajcik and A. K. Choudhery</i>	435
Testing Milk for Cleanliness of Production <i>Michael Roman</i>	438
Microwave Ovens and their Public Health Significance <i>Robert L. Elder and Walter E. Gundaker</i>	444
<i>Vibrio Parahaemolyticus</i> - A Review <i>R. Nickelson and C. Vandersant</i>	447
Amendment to 3-A Sanitary Standards for Multiple-Use Plastic Materials Used as Product Contact Surfaces for Dairy Equipment	452
Performance in Microscopic Counting of Somatic Cells in Milk. I. Effects of Procedural Variations on Accuracy and Precision <i>W. D. Schultze, J. W. Smith, D. E. Jasper, O. Klastrup, F. H. S. Newbould, D. S. Postle and W. W. Ullmann</i>	453
Amendment to the 3-A Accepted Practices for the Sanitary Construction, Installation, Testing and Operation of High-Temperature Short- Time Pasteurizers, Revised	457
Holdings of 3-A Symbol Council Authorizations on August 20, 1971	458
The National Conference on Interstate Milk Shipments: Twenty Years in Retrospect <i>J. C. McCaffrey</i>	461
Association Affairs	465
News and Events	466
Index to Advertisers	III
Classified Ads	III

Blue Ridge Rd., P. O. Box 437, Shelbyville,
Ind.
2nd Class postage paid at Shelbyville, In-
diana 46176.

EDITORIAL OFFICES: Dr. Elmer H. Marth,
Dept. of Food Science, University of Wiscon-
sin, Madison, Wis. 53706. H. L. Thomasson,
Managing Editor, P. O. Box 437 Shelby-
ville, Indiana 46176.

Manuscripts: Correspondence regarding man-
uscripts and other reading material should
be addressed to Dr. Elmer H. Marth, Dept. of
Food Science, University of Wisconsin, Mad-
ison, Wis. 53706.

"Instruction to Contributors" can be ob-
tained from the editor for the use of con-
tributors of papers.

Page Charge: Effective January 1, 1969 a
charge of \$25.00 per printed page will be made
for all research papers which are published.
See Volume 31, issues 10, 11, or 12 for details.

Business Matters: Correspondence regarding
business matters, advertising, subscriptions,
orders for single copies, etc., should be ad-
dressed to H. L. Thomasson (address above).

Subscription Rates: One volume per year.
Individual non-members. Governmental and
Commercial Organization subscription.

1 yr.	\$14.00
Public and Education Institution Libraries, 1 yr.	\$12.00
Single Copies	\$ 1.50

Orders for Reprints: All orders for reprints
should be sent to the executive office of the
Association, P. O. Box 437, Shelbyville, Ind.

Membership Dues: Members in the In-
ternational Association of Milk, Food and En-
vironmental Sanitarians, Inc., is \$14.00 per
year, and Special Certified Student mem-
bership is \$4.00 per year, which includes annual
subscription to the *Journal of Milk and Food
Technology*. All correspondence, regarding
membership, remittances for dues, failure to
receive copies of the *Journal*, changes in ad-
dress and other such matters should be ad-
dressed to the Executive Secretary of the
Association, H. L. Thomasson, Box 437, Shel-
byville, Indiana 46176.

ISOLATION OF SALMONELLAE FROM NATURALLY CONTAMINATED DRIED MILK PRODUCTS

II. INFLUENCE OF STORAGE TIME ON THE ISOLATION OF SALMONELLAE¹

BIBEK RAY², J. J. JEZESKI³, AND F. F. BUSTA

Department of Food Science and Industries
University of Minnesota
St. Paul 55101

(Received for publication March 29, 1971)

ABSTRACT

Qualitative tests for the presence of salmonellae in naturally contaminated dried milk products and tailings obtained from five plants indicated rapid decreases in numbers of positive samples. After storage of up to 10 months, <23% of the 49 product samples and <36% of the 93 tailings samples exhibited viable salmonellae. In product fractions of daily production, storage survival was greatest in the first product through the system (48.9%) when compared to an average of 23% survival in the food grade fractions. Quantitative determinations of salmonellae in nine samples indicated a significant reduction from 2.2 to 0.9 Most Probable Number per 100 g sample. These data indicated that samples which contained viable salmonellae immediately after manufacture might test negative after extended storage. However, extended storage did not free all samples of contamination. Therefore, storage cannot be based as a method of freeing contaminated dried milk products of salmonellae.

Detection of salmonellae in contaminated dried milk products may depend upon the interval between production and testing, i. e., storage time. In artificially contaminated dry milk, the survival of salmonellae, as determined by the reduction in number, was affected by storage (4, 5). In general, salmonellae were destroyed initially at a rapid rate followed by a relatively reduced rate (4). A rapid initial reduction in number of salmonellae in meat meal during storage also was reported (8). Bacterial death rates in dry milk, as measured by total number, were high during the first 2 months of storage (1, 6). This reduction in total number during storage of dry milk was extremely high at 22 C. Also, reduction in total bacterial number in dry milk stored in air was greater than that stored under nitrogen (6).

MATERIAL AND METHODS

Samples

Detection of salmonellae from naturally contaminated dried milk products was done by isolation procedures (2) and by

most probable number (MPN) procedures. For isolation studies, different product fractions and tailings samples from daily production runs from several dry milk plants were tested for the presence of salmonellae within 7 days after manufacture. All these plants had a history of producing powder contaminated with salmonellae. The different product fractions used were the first product (animal feed), and portions from the first, middle, and last bags of food grade products. All the samples from which salmonellae were isolated initially were stored in polyethylene bags at room temperature and were tested again during 2 to 12 months of storage. Limited quantities of each sample did not permit more frequent and long term testing of all the samples from these plants.

For MPN determinations, "bulk samples" in 50 and 100 lb. bags and drums were procured about 2 months after manufacture and tested for the presence of salmonellae. Samples that showed the presence of salmonellae were used. Numbers of salmonellae were determined by the MPN technique, initially and again 4 months later. Products were stored in the same manner as described previously. A total of 9 bulk samples from three different plants were used. The products tested were: two roller dried nonfat dry milk (NDM), three spray dried NDM, one spray dried whole milk, one spray dried buttermilk, and one spray dried caseinate mix.

Methods of testing

Both in the isolation studies and MPN determination, enrichment, plating on selective agar medium, biochemical identification, and serological confirmation were done according to the recommended procedure (2) and also by the procedure described in a previous paper (7).

For the isolation of salmonellae from different product fractions and tailings, samples in 100-g quantities were pre-enriched in lactose broth pre-enrichment (1000 ml in a 2-quart fruit jar.) After 20 to 24 hr incubation at 35 C, a 10-ml portion from the thoroughly mixed pre-enriched sample was used for enrichment.

For MPN determinations, lactose broth was used as the pre-enrichment medium. The method used for reconstitution and rehydration (first hour after reconstitution) prior to pre-enrichment incubation is presented in Table 1. Two temperatures (25 and 45 C) and two solid to liquid ratios (1:10 and 1:2.5) were used during reconstitution and the first hour of rehydration. In general, for any particular test condition, a total of 333 g of sample was reconstituted in the required amount of lactose broth and mixed vigorously for 2 to 3 min with a stainless steel whip (18-inch piano wire balloon type). The reconstitution and mixing were done in a stainless steel beaker of 4 to 5 liter capacity. After the rehydration period of 1 hr, each 333-g portion of sample

¹Scientific Journal Series Paper No. 7577, Minnesota Agricultural Experiment Station, St. Paul.

²Present address: Department of Food Science, North Carolina State University, Raleigh, N. C. 27607.

³Present address: Department of Botany and Microbiology, Montana State University, Bozeman, Montana 59715.

TABLE 1. RECONSTITUTION PROCEDURES FOR MOST PROBABLE NUMBER (MPN) DETERMINATION OF SALMONELLAE FROM DRIED MILK PRODUCTS

		1336 g ¹	
333 g ¹	333 g ¹	333 g ¹	333 g ¹
(A)	(B)	(A')	(B')
Reconstituted in 3,330 ml of medium, rehydrated for 1 hr, mixed and divided into 3 × 1000 ml, 3 × 100 ml, and 3 × 10 ml and incubated at 35 C.	Reconstituted in 835 ml of medium, rehydrated for 1 hr, mixed and divided into 3 × 250 ml, 3 × 25 ml, and 3 × 2.5 ml. Enough medium was added to make the volumes 1000, 100, and 10 ml, respectively, and incubated at 35 C.		
1:10 solid to liquid ratio	1:2.5 solid to liquid ratio	1:10 solid to liquid ratio	1:2.5 solid to liquid ratio
Reconstituted and rehydrated for first hour at 25 C		Reconstituted and rehydrated for first hour at 45 C	

¹Samples were dry blended in a bag by shaking 100 times.

Lactose broth was used in all phases of reconstitution and pre-enrichment.

A' and B' were the same as A and B, respectively, except for reconstitution and rehydration temperature.

At the end of the rehydration period and before pre-enrichment incubation at 35 C all the subsamples were adjusted to a 1:10 solid to liquid ratio.

TABLE 2. EFFECT OF STORAGE ON CONTAMINATED SAMPLES ON THE ISOLATION OF SALMONELLAE

Product samples	Number of samples positive for salmonellae/Number of samples tested						Tailing samples						
	(Storage time, months)												
	0	2	4	6	8	10	0	2	4	6	8	10	12
Plant 1	10/10	5/10	- ¹	10/25*	5/16	2/13	27/27	-	-	-	5/27	4/27	4/27
Plant 7	13/13	1/13	-	-	-	-	9/9	3/9	-	-	-	-	-
Plant 8	-	-	-	-	-	-	33/33	13/33	11/33	12/33	8/33	10/33	-
Plant 3	11/11	-	7/11	-	4/10	-	9/9	-	-	4/9	6/9	4/9	5/9
Plant 11	15/15	4/15	-	-	-	-	15/15	11/15	-	-	-	-	-

¹- not tested.

All the samples used in the storage study were positive for salmonellae in the first test.

Serotypes isolated from different plants: Plant 1 - *S. typhimurium*, *S. java*, and *S. blockley*

Plant 3 - *S. montevideo*; Plant 7 - *S. oranienburg*; Plant 8 - *S. minnesota*, *S. kentucky*, and *S. tennessee*; Plant 11 - *S. anatum*.

*25 samples included eight from the two months test (including all five positive) and 17 new samples.

Product samples tested at 6, 8, and 10 months from Plant 1 and at 8 months from Plant 3 included all the positive samples from the preceding tests.

was divided into nine subsamples containing three 100-g, three 10-g, and three 1-g portions. The containers used for pre-enrichment incubation were 2-quart fruit jars for the 100-g samples, wide mouth screw cap dilution bottles for 10-g samples, and screw cap test tubes (150 × 25 mm) for 1-g samples. The subsamples were incubated for 20 to 24 hr at 35 C and then enriched by the procedure described above. Each of the nine subsamples was considered as a separate unit and was tested separately according to the recommended procedure for the presence of salmonellae (2). The total number of subsamples out of the total number tested in each of the 100, 10, and 1 g levels that showed the presence of salmonellae were added together. *Salmonella* population levels were then calculated by MPN techniques according to the method suggested by Halvorson (3).

RESULTS

Survival of salmonellae

Salmonellae in naturally contaminated dried milk

products may die off during storage, possibly at a rate equal to or much faster than that in artificially contaminated products. To establish the effect of storage, a series of different fractions of products samples and tailings, which showed the presence of salmonellae when first tested within 7 days after manufacture, were retested at later dates. A reduction in number of samples from which salmonellae could be isolated after storage was considered to result from death of these organisms during storage. The results of these tests are presented in Table 2 and Fig. 1.

Product samples consisting of animal feed and first, middle, and last bags of food-grade products from four different plants were retested during 2 to 10 months of storage (Table 2). Within 2 months of storage, salmonellae were isolated from only 8

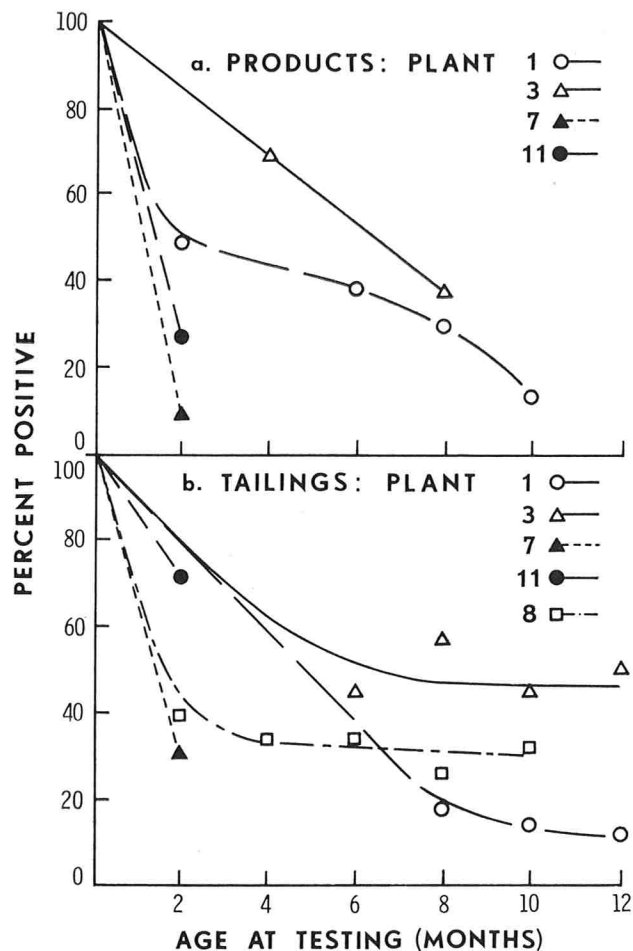


Figure 1. Effect of storage on the isolation of salmonellae from contaminated dried milk samples.

to 50% of the samples from plants 1, 7, and 11 (Fig. 1). After the initial rapid decline in incidence of isolation, *Salmonella* isolation percentages in samples from plant 1 remained essentially constant up to 10 months. However, more than 70% of the product samples from plant 3 were found to contain salmonellae after 4 months of storage. This was reduced to about 40% after 8 months. The high incidence of *Salmonella* isolation from stored plant 3 product samples contaminated with *S. montevideo* could result from one or more of the following factors: difference in serotype, difference in initial level of contamination, mechanisms of contamination, or differences in the processing conditions.

Tailings samples from five different plants were tested during a storage period up to 12 months (Table 2). A rapid reduction in the incidence of *Salmonella* isolation in tailings from plants 7 and 8 was observed within 2 months of storage (Fig. 1). Only 15% of the samples from plant 1 were positive after 8 months, the time when the samples from this plant were first retested. From these results it is impossible to determine the time during storage

where most of the reduction might have actually occurred. A relatively high percentage of tailings samples from plant 3 showed the presence of salmonellae (45 to 55%) during 6 to 12 months of storage. The product samples from this plant also showed high isolation frequencies during storage. Tailings from plant 11, which were actually powder from a central vacuum fines collection system, were positive for salmonellae in about 70% of the samples after 2 months. Products from the same plant, however, showed the presence of salmonellae in only 30% of the samples on retest after the same period of storage. This could be caused by differences in the original populations or any of the reasons mentioned before. In some of the tailings from plants 3 and 8, salmonellae were not isolated in one test but were isolated in later tests.

Survival in different product fractions

Data on *Salmonella* test results observed during storage were related to different product fractions from two plants (Table 3). The first product (animal feed) provided more isolations of salmonellae than did the other samples taken later during the production of a lot. Furthermore, contamination persisted longer in the animal feed. About 49% of the total animal feed samples tested during 2 to 10 months of storage were found to contain salmonellae. This high isolation rate could be direct evidence

TABLE 3. EFFECT OF STORAGE ON THE SURVIVAL OF SALMONELLAE IN DIFFERENT PRODUCT FRACTIONS FROM TWO DRY MILK PLANTS¹

Type of samples	Number positive/Number tested			
	Plant 1	Plant 3	Total	%
First product (animal feed)	21/59	22/29	43/88	48.9
Food grade products				
First bag	6/27	3/8	9/35	25.7
Middle bag	0/6	0/0	0/6	0.0
Last bag	4/14	0/2	4/16	25.0
Total food grade	10/47	3/10	13/57	23.0

¹All samples tested during storage were positive initially. The samples were retested during storage up to 10 months.

TABLE 4. EFFECT OF STORAGE ON SURVIVAL OF *Salmonella* SEROTYPES IN CONTAMINATED DRIED MILK PRODUCTS FROM PLANT 1¹

Number of samples positive	Fresh samples	Stored samples up to 6 months
	43	18
<i>S. typhimurium</i>	143 (67%)	95 (55%)
<i>S. java</i>	69 (33%)	74 (42%)
<i>S. blockley</i>	0 (0%)	6 (3%)

¹All 43 samples were tested during storage. From each positive sample, four or more colonies were isolated on the basis of difference in colony morphology and serologically typed.

TABLE 5. MOST PROBABLE NUMBER (MPN) OF SALMONELLAE IN CONTAMINATED DRIED MILK PRODUCTS¹

	25 C						45 C						Total				
	1:10			1:2.5			1:10			1:2.5						MPN/ 100 g	C.I. (95%)
	100	10 (A)	1	100	10 (B)	1	100	10 (A')	1	100	10 (B')	1	100	10	1		
Number of samples tested in each test	27	27	27	27	27	27	27	27	27	27	27	27	108	108	108		
Number positive in:																	
First test	19	9	3	21	12	0	19	11	3	24	11	4	83	43	10	2.2	1.67-3.09
Retest	12	6	0	16	5	1	11	4	0	12	8	2	51	23	3	0.9	0.68-1.26

¹A total of nine samples from three different sources were tested by the method shown in Table 1. Two of these samples were contaminated with *S. bredeny*, one with *S. anatum*, and two with *S. minnesota*, and four with *S. minnesota*, *S. kentucky*, and *S. tennessee*.

At the time of the first test, the samples were at least two months old. Retesting was done four months after the first test. MPN's for the total positive samples were determined by the method of Halvorson (3).

of elevated survival rate or could be caused by a high level of initial population of salmonellae. The incidence of *Salmonella* isolation was higher in animal feed from plant 3 than from plant 1. This could result from a difference in serotypes or other factors mentioned earlier. Incidence of *Salmonella* isolation from the different fractions of food grade products was relatively low, the lowest being in the sample from the middle bag.

Survival of various serotypes

To study the effect of storage on individual *Salmonella* serotypes, dried milk products contaminated with multiple serotypes were tested (Table 4). These product samples from plant 1 were contaminated with *Salmonella typhimurium*, *Salmonella java*, and *Salmonella blockley*. Four to 16 colonies were randomly selected from each plate containing typical *Salmonella* colonies (after 20 to 24 hr incubation at 35 C) and these isolates were serologically typed. Of the 212 colonies typed from 43 positive samples on the first test, about 67% were *S. typhimurium* and 33% were *S. java*. These samples were retested after 2 to 6 months of storage and salmonellae could be isolated from only 18 samples. Of the 175 colonies, about 55% were *S. typhimurium*, 42% *S. java*, and 3% *S. blockley*. Though *S. blockley* was isolated only in the stored samples, no gross differences in percentages of isolation between *S. typhimurium* and *S. java* were observed. Probably the initial levels of contamination for *S. blockley* were low in comparison to *S. typhimurium* and *S. java* and thus were not detected in mixed populations. However, *S. blockley* might have been more tolerant of storage and thus was isolated. Also, the other serotypes seemed to differ very little in their survival rates during storage.

Effect of storage on apparent populations

Salmonellae in naturally contaminated dried milk

products apparently die off during storage. The rate of this death may be fairly rapid during the early stage of storage. An estimation of the rate of reduction in population over a period of 4 months was done by MPN determination.

A total of nine bulk samples was tested at two initial rehydration temperatures and two initial rehydration solid to liquid ratios in lactose broth pre-enrichment medium. The total number of subsamples that showed the presence of salmonellae out of the total number of subsamples tested in each of the 100, 10, and 1-g levels were used to determine MPN values (Table 5). Although two rehydration temperatures and two rehydration solid to liquid ratios were used, the results did not show any major differences between these variables. Values in the first test and retest were calculated only on the compiled data. The MPN of salmonellae in 100 g of product was 2.2 in the first test and 0.9 in the retest. Even though the samples were at least 2 months old during the first test, about 60% reduction in population occurred within the next 4 months of storage. Reduction in percentage of population might have been much higher during the early period of storage.

DISCUSSION

Salmonella populations in naturally contaminated dried milk products appeared to die off rapidly during storage. In general, the presence of salmonellae was not demonstrable in about 60 to 70% of the contaminated samples after storage. Initial rapid reduction may have occurred early in storage and possibly within the first few days after manufacture, particularly when the samples are stored at room temperature and without any humidity control. Similar rapid reduction in total count as well as in *Salmonella* population in dried milk products has been observed by other workers. Crossley and John-

son (1) reported a 70% reduction in original total counts within 4 weeks of storage of dry milk powder at room temperature. In artificially contaminated dry milk, McDonough and Hargrove (5) observed about 50% survival of original *Salmonella* population after 1 week and 2% after 4 weeks of storage at 26.6 C. Riemann (8) also observed about a 100-fold reduction of salmonellae in meat meal during the first few days of storage.

Death of salmonellae in naturally contaminated dried milk products during storage appeared rapid initially and slowed in later phases of storage. Even in samples that were 2 months old, there was about a 60% reduction in population over a period of 4 months storage. The reduction rate during the initial stage of storage is probably much higher. Supplee and Ashbaugh (9) observed that bacterial death rate in dry milk powder determined by total count was high during the first 2 months of storage and reduced later in storage. LiCari and Potter (4) reported that initially *Salmonella* destruction occurred rapidly, and after about 2 weeks of storage, destruction occurred at a reduced rate. Similarities exist in the death pattern of salmonellae during storage in the naturally or artificially contaminated dried milk product.

Survival of salmonellae in dried milk products during storage may be dependent upon several factors such as the numbers of organisms present initially, serotype involved, type of processing, kind of product, conditions, and duration of storage, etc. With the samples stored at room temperature and without humidity control, the initial number of salmonellae was a major factor in determining whether or not a sample exhibited the presence of salmonellae on retest during storage. Initial high populations, therefore, may have been the reason for the first product processed during a day's operation demonstrating higher percentages of positive tests during storage.

Different serotypes of salmonellae may have different survival rates during storage of the contaminated products. *Salmonella montevideo* isolated from the samples from one plant appeared to be somewhat more resistant to storage conditions than several serotypes isolated from other plants. This also could result from a difference in initial population. In a multiserotype contaminated product, the frequency of isolation of a particular serotype may depend on the relative proportion of these serotypes in the population. The isolation of *S. blockley* only upon retest may have been due to its initial number in comparison to *S. typhimurium* and *S. java* and relatively better resistance to storage. Existence of

different survival rates during storage among *Salmonella* serotypes also was reported by LiCari and Potter (4).

Though salmonellae in naturally contaminated products appeared to die off during storage, this method cannot be adopted to free all samples from contamination. In a majority of samples, the presence of salmonellae could not be detected within two months. However, in some samples the presence of salmonellae was detected after 1 year of storage. A similar suggestion has also been made by LiCari and Potter (4).

ACKNOWLEDGMENT

This investigation was supported in part by Food and Drug Administration Contract 67-61 and Contract 68-46 and by a grant from the American Dry Milk Institute. We thank the Consumer and Marketing Service, USDA, for their cooperation in obtaining samples, the Minnesota Department of Health for assistance in serologically typing and identifying isolates, and local food industries laboratories who assisted in supplying certain samples. We also thank Miss Gretchen Saecker for technical assistance and H. Orin Halvorson and F. B. Martin for consultations on statistical evaluations. We are grateful to a group of people associated with the plants involved, who must remain anonymous, but without whose cooperation and active assistance, these data could not have been collected.

REFERENCES

1. Crossley, E. L., and W. A. Johnson. 1942. Bacteriological aspects of the manufacture of spray-dried milk and whey powders, including some observations concerning moisture content and solubility. *J. Dairy Res.* 13:5-44.
2. Galton, M. M., G. K. Morris, and W. T. Martin, 1968. Salmonellae in foods and feeds. Review of isolation methods and recommended procedures. U. S. Dept. of Health, Education, and Welfare, Public Health Service, C.D.C., Atlanta, Georgia. p. 17-25.
3. Halvorson, H. O. 1958. Some elements of statistics for microbiologists. Dept. of Biochemistry, The Technical University of Norway, Trondheim. p. 19-22.
4. LiCari, J. J., and N. N. Potter. 1970. *Salmonella* survival during spray drying and subsequent handling of skim-milk powder. III. Effects of storage temperature on *Salmonella* and dried milk properties. *J. Dairy Sci.* 53:877-882.
5. McDonough, F. E., and R. E. Hargrove. 1968. Heat resistance of *Salmonella* in dried milk. *J. Dairy Sci.* 51:1587-1591.
6. Nichols, A. A. 1939. Bacteriological studies of spray-dried milk powder. *J. Dairy Res.* 10:202-249.
7. Ray, B., J. J. Jezeski, and F. F. Busta. 1971. Isolation of salmonellae from naturally contaminated dried milk products. I. Influence of sampling procedures on the isolation of salmonellae. *J. Milk Food Technol.* (*In press*)
8. Riemann, H. 1968. Effect of water activity on the heat resistance of *Salmonella* in "dry" materials. *Appl. Microbiol.* 16:1621-1622.
9. Supplee G. C., and V. J. Ashbaugh. 1922. Bacterial content of milk powder. *J. Dairy Sci.* 5:216-228.

SANITATION AND WATER USAGE IN THE PROCESSING OF SOUR CHERRIES¹

Y. D. HANG, D. L. DOWNING, AND D. F. SPLITTSTOESSER

New York State Agricultural Experiment Station,
Cornell University, Geneva, New York 14456

(Received for publication March 22, 1971)

ABSTRACT

Surveys were made of four red sour cherry processing plants to obtain information regarding microbial contamination and water usage. With the exception of some long conveyor belts, no single sites were found to be major sources of contamination and the mean count at most stages was under 50,000 per gram.

The primary sources of waste water were the soak tanks, sprays, and flumes. From 600 to over 800 gal of water were discharged per ton of fruit processed; the BOD per ton ranged from 6 to over 10 lb. The study pointed to the soak tanks and pit flumes as possible areas where waste water generation could be reduced.

Pure waters programs are forcing many members of the food industry to improve their treatment of aqueous wastes that are to be discharged into classified waters. Treatment costs usually are sizeable for fruit and vegetable processors because of the high volume and strength of their wastes. One of the more promising approaches for reduction of these costs is to minimize water consumption and consequently effluent production (6). Although a prerequisite for this is an understanding of the effect of different variables on waste water generation; only a limited number of reports describe in-plant studies dealing with this aspect of fruit and vegetable processing (2, 4, 7, 10).

The purpose of this research was to study water usage and sanitation in the processing of red sour pitted (RSP) cherries. It was felt that the two should be considered together to avoid alterations for reducing waste water that might sacrifice sanitation and thus product quality.

MATERIALS AND METHODS

Waste water flow rates were measured by two methods. Discharges from various equipment and pipes were estimated by the calibrated container-stop watch procedure in which the time required to fill a container was determined (3). The flow through open channels was calculated on the basis of cross section dimensions and stream velocity. The latter, measured with a cork float and stop watch, was corrected to establish the mean velocity (5).

Samples of waste water and fruit were sealed in one

pint polyethylene bags and transported to the laboratory in an insulated ice chest over ice. Bacteriological analyses were performed as soon as the samples reached the laboratory, the same day they were collected. The samples to be analyzed for BOD, total solids, and volatile solids were frozen immediately and stored at -23 C until the tests could be made.

The microbial populations on fruit were determined by blending 22 g in 198 ml of sterile water for 2 min in a Waring blender. Appropriate decimal dilutions were plated on potato dextrose agar, pH 5.6. Colonies were counted after an incubation of 4 days at 20 C. Water samples were handled similarly except that mechanical blending was omitted.

Standard methods (1) were used for determining the BOD, total and volatile solids.

RESULTS AND DISCUSSION

During the 1970 processing season 22 surveys were made of the lines of four cherry processors. Three of the plants prepared the fruit for freezing while the fourth was a cannery; all were quite similar with respect to the processing steps that were followed. The general procedure was that the fruit was transported to the factory in water contained in half-ton pallet tanks. After weighing it was transferred to soak tanks where it was held 8 or more hr in 10 C water to effect firming. The soak tanks were part of a closed recirculated-water system in which the water also was pumped through a reservoir containing ice for cooling and through different flumes used for conveying the fruit. Following the soak tanks the cherries were conveyed via flume and belt through the stemmer, size and color graders, an inspection table, and the pitter. They then were ready for canning or freezing.

Microbiology

Microbial counts were made on 120 samples of fruit collected at the different processing stages. The data are presented as geometric means (Table 1) because of the considerable variation in counts from survey-to-survey. In 8 studies of Plant B, for example, the counts on cherries taken from the inspection belt ranged from 6 to 240×10^3 per gram.

In general, no specific sites were found to be the primary sources of contamination. This was unexpected since it had been anticipated that certain

¹Approved by the Director of the New York State Agricultural Experiment Station for publication as Journal Paper No. 1877.

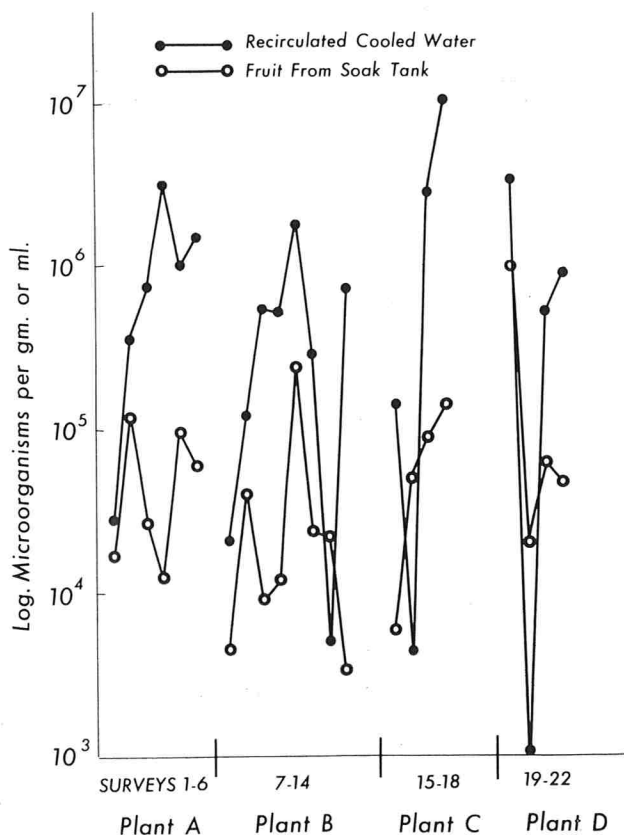


Figure 1. Individual survey results comparing microbial counts of recirculated water with that of fruit from the soak tanks.

TABLE 1. MICROBIAL CONTAMINATION OF CHERRIES DURING THEIR PREPARATION FOR FREEZING.

Sampling site	Processing plant	Microorganisms/g - geometric mean $\times 10^{-3}$			
		A	B	C	D
Receiving station		43	17	20	45
Following soak tank		40	17	44	87
After stemmer		12	35	8.9	19
Following grader		—	10	19	—
Inspection belt		16	20	—	47
After pitter		15	14	78	25
Filler		8.9	12	35	160

equipment such as the pitters would be areas of significant microbial build-up. These results, which differ from our observations of vegetable lines (8, 9) are thought to be due to the low pH of cherries, under 4.0, which selects for yeasts as the predominant contaminant. The periodic line clean-ups, usually limited to shutdown periods, appear to have been sufficiently frequent to control the relatively slow growing yeasts.

Plant D used a number of long belts to convey the fruit from pitters to fillers. This is believed to explain the significantly heavier contamination of cherries at this latter stage. Cherries at Plant D were

given a thermal process and, therefore, this viable count would not be found in the preserved product.

The microbial populations of the recirculated 10 C water that is pumped through the soak tanks and flumes was compared with the level of contamination on fruit taken from the soak tanks (Fig. 1). There was considerable variation in counts; the soak water at Plant B, for example, yielded 5×10^5 to 1.8×10^6 organisms per milliliter. These differences undoubtedly reflected the time that had lapsed since the system had been completely drained and refilled—it was not uncommon for this to be done weekly or at even longer intervals.

It can be seen (Fig. 1) that the concentration of organisms on the fruit paralleled that in the water to only a limited extent and that the number per milliliter of water was almost always higher than the number per gram of fruit. The latter would be expected since most of the organisms on the cherry would be restricted to its surface. The rather poor correlation between microbial densities on the fruit and in the water suggest that other factors have a marked effect on the microbiology of the cherry. It is likely that an important variable was the incidence of surface fractures on the fruit that was cultured.

Waste water

Soak tanks, sprays, and flumes were the main sources of waste water in the four plants (Table 2). The gallons of water and pounds of BOD per ton of cherries shown here probably underestimated the true values because of additional water used for clean-ups, and because the soak tank-flume-cooled water systems were completely drained and refilled periodically. Surprisingly, the different processors could not provide figures as to the volumes of water in these systems.

Total cherry production and water consumption data for the entire processing season, available for Plants A and B, permitted an evaluation of these per-ton figures. Based on seasonal totals, Plant A used 1800 gal of water per ton, a value considerably higher than the 790 gal estimated from in-plant flow measurements. However, this factory completely emptied the water from its soak tanks daily which could account for the difference. The seasonal calculations for Plant B, which emptied its recirculated water weekly, indicated a consumption rate of 780 gal per ton, a figure comparable to the 869 gal in Table 2.

It would appear that the amount of waste water generated in the soak tank-flume operation varied considerably from factory-to-factory. In addition to the frequency that different plants emptied and refilled these systems, considerable variation in wa-

TABLE 2. SOURCES AND PROPERTIES OF WASTE WATER

Plant	effluent source	Av. flow gal/hr	Av. Strength (ppm)			Total/ton of fruit ²	
			BOD	TS ¹	VS ¹	gal lb	BOD
A.	Soak tank	2190	1230	3126	2770	790	10.5
	Pit flume	1060	2600	5910	5560		
B.	Soak tank	1590	137	482	392	869	6.1
	Spray rinse	1580	334	1020	852		
	Flume	2460	1100	3020	2800		
	Pit flume	3110	1070	2750	2540		
C.	Spray rinse	1220	814	2050	1910	611	7.1
	Flume	781	68	306	219		
	Pit flume	4110	1570	3690	3460		
D.	Spray rinse	759	829	2140	1870	847	9.6
	Flume	4900	1340	3260	2920		
	Pit flume	2810	1120	2710	2420		

¹Total and volatile solids.

²Approximate figures based on estimated hourly production and effluent flow at the different discharge points.

ter discharge and make-up rates was observed. Factory A, for example, discharged almost twice as much water per hour as did Plant B although its cherry processing output was only about one-half as great. Plants C and D did not discharge water at this point although some of the discharge from their flumes represented soak tank water.

Our conclusions were that there may be little technological basis for the water management practices presently followed in cherry processing soak systems. It would seem that an ideal approach might be to establish make-up rates and drainage schedules that would keep microbial populations below a certain level. Practical methods for accomplishing this would be difficult, however, without some rapid method for estimating microbial numbers. Our attempts to correlate pH or the optical densities of recirculated water with viable counts were not successful.

All four of the plants were found to use fresh water for the fluming of cherry pits. The amount of waste generated by this practice was sizeable, both as to volume and strength (Table 2). Since the pits were to be discarded as solid waste, here was an area where a significant reduction in waste water could be effected without any threat of reducing product sanitation. Measures that might be adopted include extended recirculation of the flume water or the use of belts or some other means to convey the pits.

ACKNOWLEDGEMENT

John Peck ably assisted in the surveys and waste water analyses.

REFERENCES

1. Anon. 1965. Standard methods for the examination of water and waste water. 12th ed. pp. 244, 247, 406. Amer. Public Health Ass., New York, N. Y.
2. Anon. Cannery waste treatment, utilization and disposal. Publ. No. 39, State Water Resources Control Board, Sacramento, Calif. p. 274.
3. Anon. 1969. Guide for waste management, p. 3.3. National Canners Ass., Berkeley, Calif.
4. Eckenfelder, W. W., Jr., C. Woodward, J. P. Lawler, and R. J. Spinna. 1958. Study of fruit and vegetable processing waste disposal methods in the eastern region. Final report. Contract 12-14-100-482(73). Manhattan College, New York, N. Y. p. 99.
5. Kent, W. 1950. Mechanical engineers' handbook, 12th ed. Sect 4, Art. 8. John Wiley and Sons, Inc. New York, N. Y.
6. Mercer, W. A. 1970. Pollution problems and solutions for the vegetable processing industry. pp. 4-7. In Proc. Conference on the treatment and disposal of wastes from vegetable processing. ARS 72-80. US. Dept. of Agr., Southern Util. R & D Div., New Orleans, La.
7. Rambo, R. S. 1968. Vegetable canning process wastes. Ph.D. thesis. Univ. of Wis., Madison, p. 157.
8. Splittstoesser, D. F., W. P. Wettergreen, and C. S. Pederson. 1961. Control of microorganisms during preparation of vegetables for freezing. I. Green beans. Food Technol. 15:329-331.
9. Splittstoesser, D. F., W. P. Wettergreen, and C. S. Pederson. 1961. Control of microorganisms during preparation of vegetables for freezing. II. Peas and corn. Food Technol. 15:332-334.
10. Weckel, K. G., R. S. Rambo, H. Veloso, and J. H. von Elbe. 1968. Vegetable canning process wastes. Research Report 38, College of Agric. and Life Sci., Univ. of Wis., Madison. p. 20.

TEMPERATURE PROFILES IN HOLDING TUBES OF MILK AND EGG PASTEURIZERS

R. W. DICKERSON, JR. AND R. B. READ, JR.

U. S. Department of Health, Education, and Welfare
Public Health Service, Food and Drug Administration
Bureau of Foods, Office of Food Sanitation
Division of Microbiology, Food Process Evaluation Branch
Cincinnati, Ohio 45226

(Received for publication March 5, 1971)

ABSTRACT

Product temperatures in a cross-section of the holding tube of milk and egg pasteurizers are required to be within 1 F, and this condition is assumed to be met when tube diameter is < 7 inches and product velocity is at least 1 ft/sec. Because of the viscous nature of some milk products and most egg products, the 1 F requirement might not be met, and therefore product temperatures were measured at the entrance and exit cross-sections of both milk and egg holding tubes.

Product temperatures were measured with 0.010-inch-diameter thermocouples and a 24-point recorder. With the pasteurizer operating with product in forward flow, holding tube temperatures were monitored for about 0.5 hr. Except for condensed skim milk, milk product temperatures in a cross-section did not vary by more than 0.8 F. Although the temperature variation of condensed skim milk exceeded the 1 F requirement, it was within 2 F. For plain yolk and for salted yolk (10% salt by weight) the variation within a cross-section was 1.2 and 4.8 F respectively. The variation for the other egg products was < 1 F.

Holding tubes are required on milk and egg pasteurizers to ensure that every particle of product is held for a minimum time at some minimum temperature. If there were large differences in product temperature in the holding tube, pathogens might survive in the cooler portions of product, reducing the effectiveness of pasteurization. To prevent this, there is a requirement that product temperatures in a cross-section of the holding tube cannot differ by more than 1.0 F (8, 9). It has been generally assumed that this condition is met when the tube size is less than 7 inches in diameter and product velocity is 1 ft/sec or more (8). For milk and water, the flow is turbulent (1, 2) and no temperature differences are expected.

For viscous products, however, the flow is not turbulent (1, 3, 4, 6). Furthermore, the requirement of a minimum product velocity of 1 ft/sec cannot be met with a product such as salted yolk (10% salt by weight). With a normal plating arrangement of the pasteurizer, Scalzo et al. (7) have shown that a pressure of 64 psig was required to pump salted yolk through the pasteurizer at a capacity yielding an average velocity of 0.4 ft/sec in the holding tube.

An attempt to operate the pasteurizer at a velocity of 1 ft/sec in the holding tube resulted in significant leakage from the heater section. Consequently, we questioned whether the 1.0 F requirement could be met with viscous products such as egg and ice cream mix. Finally, some milk particles have been observed moving as much as 3.8 times faster than average through the heater section of a pasteurizer (10), and we wondered if uneven heating would cause significant temperature variations in product at the inlet to the holding tube.

The objective of this work was to measure product temperature differences at the inlet and exit cross-sections of holding tubes used on pasteurizers for both egg and milk products.

MATERIALS AND METHODS

Products and equipment

The pasteurizers, holding tubes, and products used in this study were those described by Dickerson et al. (1) for milk products and Scalzo et al. (6) for egg products. The holding tube used for milk was size 1.5 inches O.D. stainless-steel tubing, 20 ft long, consisting of 3 unequal straight sections, one 180-degree bend, and one 90-degree bend. For the other milk products, the holding tube was the same diameter and 30 ft long, consisting of 5 unequal straight sections, three 180-degree bends, and one 90-degree bend. Neither milk product holding tube was insulated. The holding tube used for salted yolk (10% salt by weight) was size 1.5 inches O.D. stainless-steel tubing, 84 ft long. The holding tube used with the other egg products was the same diameter and 210 ft. long. Piping arrangements were as described previously (6). Both egg holding tubes were insulated with ARMAFLEX¹ (trade name of the Armstrong Cork Co.) moisture-proof pipe insulation. The insulation was 0.5 inch thick. The inside diameter of all holding tubes was 1.40 inches.

Test procedure

The pasteurizer was operated as described previously (1, 6). Flow rate was 300 gal/hr for all products except salted yolk, (10% salt by weight), and this yielded an average holding tube velocity of 1 ft/sec. Flow rate for salted yolk was 114 gal/hr with an average velocity of 0.4 ft/sec.

The small thermocouples were susceptible to breakage and we tested them for continuity prior to each temperature de-

¹Mention of a commercial product implies neither endorsement nor criticism by the U. S. Department of Health, Education, and Welfare.

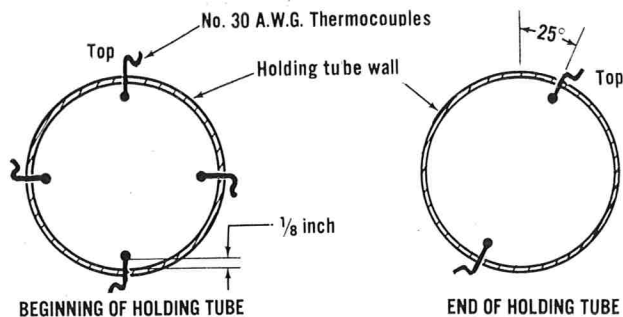


Figure 1. Position of thermocouples in the holding tube.

termination. This was done by pushing the indicating head of the recorder off the balance point and observing a restoring force. To achieve steady-state conditions, the pasteurizer was operated with product in forward flow for about 20 min before harvesting of data. With the exception of one trial on ice cream mix, in diverted flow (8), all tests were performed in the forward flow condition.

Instrumentation

Thermocouples with insulation but without a metallic sheath were fabricated from 0.010-inch-diameter copper-constantan duplex wire and installed as shown in Fig. 1. The thermocouple junctions were located diametrically 1/8 inch from the tube wall. These locations were selected as the ones believed to yield the largest temperature variation. At the beginning of the holding tubes, temperature variations were expected to be the result of uneven heating in the heater section, and because of a 90-degree sharp turn at the heater exit, individual streamlines of flow should have been selectively segregated between the inside and outside radii of the turn. Thermocouples at the beginning of the holding tube were located in a cross-section 7 inches downstream from the exit port of the heater. At the end of the holding tube there were no changes in flow direction, and temperature differences were expected to be the result of differences in fluid density with the coldest product flowing along the bottom of the tube; therefore, we measured temperatures at the top and bottom of the tube. The thermocouples were shifted slightly from the true vertical position to ensure that they would always be immersed in product. Preliminary investigations with a glass holding tube indicated a continuous flow of air bubbles (about 3/8 inch in diameter and 1/8 inch thick) along the top of the holding tube. Thermocouples at the end of the holding tube were located in a cross-section 14 inches upstream from the flow diversion valve for milk products and 20 inches upstream for egg products. Temperatures were recorded with a 24-point recorder (Bristol Co. Model 24P12G591). The smallest chart division was 2 F, and readings were interpolated to 0.2 F.

The thermocouples were calibrated with an NBS thermometer at 180 F for milk products and 145 F for egg products. All bare wire thermocouples were immersed in a water bath, and all readings were within 0.2 F at both temperatures cited above.

RESULTS AND DISCUSSION

Holding tube temperatures were recorded automatically and a complete set of temperatures were obtained about 2 times per minute during each test (about 80 min). Temperature differences within

a cross-section did not vary significantly with time and the data reported here are those occurring approximately 40 min after the changeover from water to product operation.

Milk products

Except for condensed skim milk, product temperatures in a cross-section did not vary by more than 0.8 F, and the existing requirement of 1.0 F was met. Conversely, condensed skim milk exhibited temperature variations that exceeded 1.0 F but were within 2.0 F (Table 1). The variation was greatest at the beginning of the holding tube; it differed by less than 0.8 F at the end of the holding tube for all products.

As expected, product temperatures at the end of the holding tube were higher at the top of the tube. Despite the absence of insulation, temperature drop along the holding tube was < 2.4 F for all products.

With the exception of ice cream mix, all tests were performed in the forward flow condition. For ice cream mix, tests were performed in both forward and diverted flow, and no significant differences were detected. During diverted flow, data were taken after multi-pass heating had raised heater inlet temperature from the forward flow condition of 136 F to 173 F.

To determine if the temperature differences would be more severe under non-steady-state conditions, a test on ice cream mix (16% milk fat) was conducted wherein the steam and hot water recirculating pump were shut off and holding tube temperatures were recorded during the transient. The flow-diversion valve was held in forward flow to prevent the flow disturbances that would otherwise occur. Despite the sharp drop in temperatures, the temperature differences within the cross-section at the beginning of the holding tube were about the same as those observed during steady-state operation (Fig. 2).

For all milk products, temperature differences across and along the holding tube were small, even at the beginning of the holding tube. This suggests that the holding tube can begin immediately at the heater exit port. Furthermore, it is acceptable to locate the temperature sensing element of the recorder-controller at the beginning of the holding tube as recommended by Read et al. (5) for Ultra-High-Temperature (UHT) processes. Our data show even temperature distributions in a cross-section for products at about 180 F, and product temperatures at 190 to 212 F (the UHT processes) are expected to be even more uniform because of the lower fluid viscosities at the higher temperatures.

Egg products

With the exception of salted yolk (10% salt by

TABLE 1. VARIATION OF TEMPERATURE OF MILK PRODUCTS AND WATER IN THE HOLDING TUBE OF A MILK PASTEURIZER

Product ^a	Holding tube inlet				Holding tube exit	
	Top	Side	Bottom	Side	Top	Bottom
(Temperature, F)						
Water	176.2	176.4	177.0	176.8	176.2	176.0
Raw milk	163.2	163.0	162.6	163.2	163.2	163.2
Ice cream mix (16% milk fat)	177.0	177.6	177.4	177.4	176.4	176.0
Diverted flow	176.8	177.4	177.2	177.2	176.6	176.0
Condensed skim milk (40% total solids)	175.2	176.2	177.2	176.8	175.4	174.8

^aIn forward flow except as otherwise noted.

weight), temperatures of egg products in holding tubes were uniform (Table 2), despite the long holding tube (210 ft). The temperature drop along the holding tube was small (0.016 F/ft), but the insulation was required to maintain these temperature distributions. A test with an uninsulated holding tube was not conducted on the egg pasteurizer; however, the temperature drop of 0.053 F/ft for ice cream mix suggests a temperature drop of 11.1 F for an uninsulated 210-ft holding tube. Consequently, holding tubes of egg pasteurizers must be insulated.

For salted yolk (10% salt by weight), the difference in product temperature in a cross-section was significant at both the beginning (3.6 F) and end (4.8 F) of the holding tube. For the entire holding tube, the lowest product temperature occurred at the end of the tube, near the bottom. Rather than attempt to meet the 1.0 F requirement within a cross-section for salted yolk, it appeared more reasonable to locate the temperature-sensing element at the zone of lowest temperature. An off-set mounting ferrule is commercially available (Taylor Instrument Companies) and may be used to position the temperature-sensing element at the bottom of the tube. At the end of the holding tube, the zone of highest temperature occurred at the top of the tube and this is the worst location for the temperature-sensing element.

For the other egg products, and milk products as well, the specific location of the temperature-sensing element within a cross-section is not significant.

Although the lowest temperature occurred at the end of the holding tube, the temperature-sensing element may be located upstream from the flow-diversion valve within the limits recommended by the Public Health Service (9). For the holding tube and flow rate we used with salted yolk, this location is between 8 and 16 ft upstream from the flow diversion

valve. Since the temperature drop along the bottom of the tube was approximately 0.03 F/ft for salted yolk, the maximum upstream position would have yielded a process control error no greater than 0.5 F, provided the temperature-sensing element was located at the bottom of the tube cross-section. The longitudinal temperature drop of 0.03 F/ft is not precise; it does not include the effect of residence time in the holding tube. The process controller exhibited a slight drift (± 0.1 F/min) and because of the 3.5-min average residence time in the holding tube, a longitudinal temperature change of ± 0.35 F can be directly attributed to the process controller.

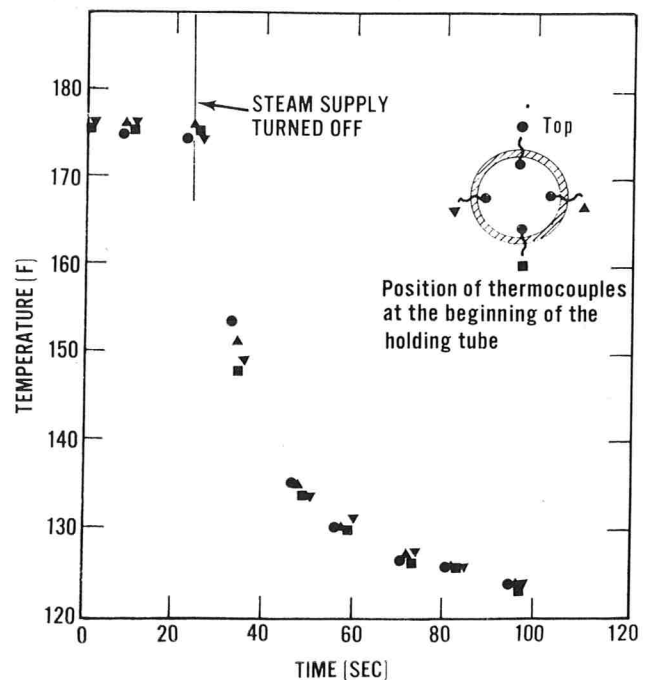


Figure 2. Temperatures of ice cream mix in a holding tube during heat supply failure.

TABLE 2. VARIATION OF TEMPERATURE OF EGG PRODUCTS AND WATER IN THE HOLDING TUBE OF AN EGG PASTEURIZER

Product ^a	Holding tube inlet				Holding tube exit	
	Top	Side	Bottom	Side	Top	Bottom
	(Temperature, F)					
Water	145.8	146.2	146.2	146.2	145.2	144.8
Liquid whole egg	144.2	144.4	144.4	—	143.4	143.0
Stabilized egg white (pH 7)	144.4	145.6	145.0	145.2	143.8	143.2
Plain yolk	144.6	145.8	145.1	144.6	143.2	142.4
Salted yolk (10% salt by weight)	147.6	144.0	144.0	147.0	146.2	141.4

^aIn forward flow.

In comparison with the longitudinal temperature drop of salted yolk (2.6 F), however, the effect is not significant.

REFERENCES

- Dickerson, R. W., Jr., A. M. Scalzo, R. B. Read, Jr., and R. W. Parker. 1968. Residence time of milk products in holding tubes of high-temperature, short-time pasteurizers. *J. Dairy Sci.* 51:1731.
- Jordan, W. K., R. F. Holland, and J. C. White. 1952. The efficiency of holding tubes used in high-temperature short-time pasteurizers. *J. Milk Food Technol.* 15:155.
- Kaufman, V. F., K. Ijichi, and G. W. Putnam. 1968. Flow characteristics in holding tubes of commercial egg pasteurizers. *J. Milk Food Technol.* 31:269.
- Kaufman, V. F., G. W. Putnam, and K. Ijichi. 1968. Test methods for measuring minimum holding time in continuous egg pasteurizers. *J. Milk Food Technol.* 31:310.
- Read, R. B., Jr., R. W. Dickerson, Jr., and H. E. Thompson, Jr. 1968. Time-temperature standards for the ultra-high temperature pasteurization of Grade A milk and milk products by plate heat exchange. *J. Milk Food Technol.* 31:72.
- Scalzo, A. M., R. W. Dickerson, Jr., R. B. Read, Jr., and R. W. Parker. 1969. Residence times of egg products in holding tubes of egg pasteurizers. *Food Technol.* 23:678.
- Scalzo, A. M., R. W. Dickerson, Jr., and R. B. Read, Jr. 1970. Pressure relationships in the regenerator of an egg pasteurizer. *J. Food Sci.* 35:411.
- U. S. Public Health Service. 1965. Grade "A" pasteurized milk ordinance - 1965 recommendations of the U. S. Public Health Service. Public Health Service Publication No. 229. U. S. Government Printing Office, Washington, D.C.
- U. S. Public Health Service. 1970. Ordinance and code regulating the processing of eggs and egg products - 1970 recommendations of the U. S. Public Health Service. Public Health Service Publication No. 1943. U. S. Government Printing Office, Washington, D. C.
- Windau, T. D., R. W. Dickerson, Jr., and R. B. Read, Jr. 1969. Residence times of milk in a flow passage of a plate-type pasteurizer. *J. Dairy Sci.* 52:898. (Abstr.)

IMMUNOGLOBULINS IN SKIMMILK AND MODEL SYSTEMS AND THEIR EFFECT ON *BACILLUS CEREUS*¹

E. M. MIKOLAJCIK AND A. K. CHOUDHERY²

Department of Dairy Technology, The Ohio Agricultural
Research and Development Center and
The Ohio State University Columbus 43210

(Received for publication April 2, 1971)

ABSTRACT

Immunoglobulins isolated from milk by ammonium sulfate fractionation were added to skimmilk and the rate of *Bacillus cereus* spore germination and subsequent vegetative cell growth was determined. At 1 hr incubation at 35 C, 96.6, 96.8, 91.4, and 74.3% of the spores had germinated in pasteurized (62.8 C - 30 min) skimmilk without added immunoglobulins, skimmilk pasteurized with immunoglobulins added, pasteurized skimmilk supplemented with unheated immunoglobulins, and nonheated unsupplemented skimmilk, respectively. With respect to vegetative cell growth, the number of generations at 4 hr in these systems was 5.9, 5.6, 4.4, and 2.3. Nonheated skimmilk without added immunoglobulins depressed *B. cereus* spore germination and vegetative cell growth to a greater extent than did pasteurized skimmilk supplemented with unheated immunoglobulins. In model systems composed of protein-free skimmilk dialysate and heated or unheated immunoglobulins, the system with unheated immunoglobulins lowered spore germination by 20% and subsequent cell growth by one-half in comparison with the heated system indicating that vegetative cells of *B. cereus* 7 are more susceptible to the immunoglobulins than spores.

We (5) have observed that unheated sterile skimmilk was a less satisfactory germination and growth medium for *Bacillus cereus* 7 than the same skimmilk heated at 62.8 for 30 min. This study was undertaken to determine if the effect was related to bovine lacteal immunoglobulins.

EXPERIMENTAL PROCEDURE

Our previous paper (5) describes the procedures followed for preparation of unheated *B. cereus* 7 spores and for determination by the agar plate method of spore germination and vegetative cell growth. Spores of *B. cereus* 7 were heat-shocked at 80 C for 12 min in sterile distilled water prior to inoculation of the skimmilk or model systems.

Sterile unheated skimmilk was prepared by Seitz filtration (7). The protein-free skimmilk dialysate used for model systems was obtained by dialyzing sterile water against unheated sterile skimmilk (2). Pasteurization of skimmilk and model systems was at 62.8 C for 30 min.

The immunoglobulins were prepared from mixed herd raw milk by ammonium sulfate fractionation procedure of Smith (8). The D fraction was used. The protein content was determined by the method of Lowry et al. (4). Analysis of the D fraction by polyacrylamide gel disc electrophoresis (1) at pH 8.9 and gel strength of 7.5% revealed the presence of

a single major band just below the spacer gel. Upon immunoelectrophoresis (3) of the D fraction against anti-bovine serum (rabbit), precipitin arcs corresponding to IgG1, IgG2, and IgM were evident. IgA was not detected.

RESULTS

To study the effect of immunoglobulins on *B. cereus* spore germination and vegetative cell growth in skimmilk, the rate of germination and vegetative cell growth was determined in four systems: unheated skimmilk, pasteurized skimmilk, pasteurized skimmilk fortified with unheated immunoglobulins, and skimmilk pasteurized after fortification with immunoglobulins. The immunoglobulins were added to a final concentration of 200 µg protein/ml. Each system was inoculated with approximately 50,000 heat-shocked *B. cereus* spores/ml and incubated at 35 C.

Spore counts of the systems are shown in Fig. 1 with the percent germination indicated at 1 and 8 hr. Upon incubation, spore counts decreased in all skim-milks with the decrease being more pronounced in the heated than nonheated systems. At 1 hr, germination was 74.3% in the unheated skimmilk, 91.4% in the pasteurized skimmilk fortified with unheated immunoglobulins, and over 96% in the fully heated systems. Whereas at 8 hr >99% of the spores had germinated in the heated systems, the unheated system continued to suppress germination and only 90.5% of the spores had germinated. Initially, the unheated immunoglobulins depressed spore germination, but, the effect was dissipated upon continued incubation or upon heat treatment of the immunoglobulins. In unheated skimmilk, agents other than the immunoglobulins had a more pronounced and sustained effect on spore germination.

The rate of multiplication of *B. cereus* following incubation in the various systems is shown in Fig. 2. The number of generations at 4 hr was determined from the total count. In all skim-milks, the number of vegetative cells increased as incubation progressed. However, rates of increase were more rapid and the number of cells was higher in the completely heated systems than in the unheated skimmilk or the heated skimmilk fortified with unheated immunoglobulins. At 4 hr, the number of generations was 2.25, 4.35, 5.62, and 5.89 in unheated skimmilk, pasteurized skimmilk with unheated immunoglobulins, and pas-

¹Approved as Journal Series Article No. 29-71 of The Ohio Agricultural Research and Development Center, Wooster. This investigation was supported in part by Public Health Services Grant EF - 00161 from the Office of Research and Training Grants, Food and Drug Administration.

²Present address: Amul Dairy, Anand, India.

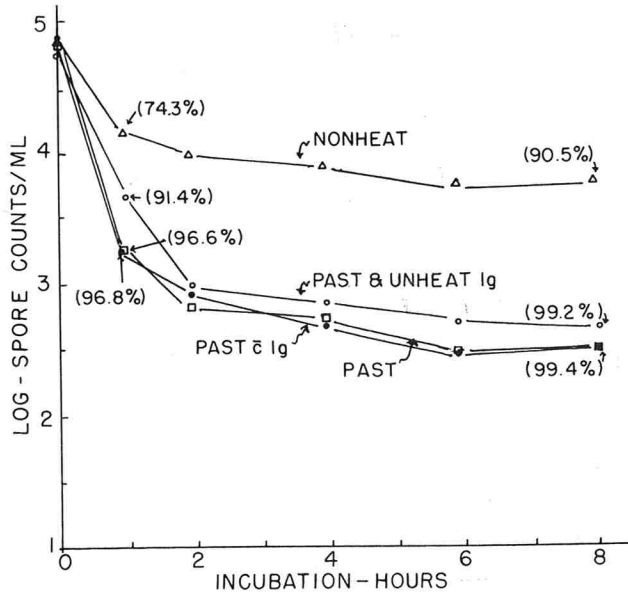


Figure 1. Spore counts of *Bacillus cereus* 7 in unheated (nonheat) and pasteurized (past) skim milk, and in pasteurized skim milk with added unheated immunoglobulins (past & unheat Ig), and in skim milk pasteurized following addition of immunoglobulins (past \bar{c} Ig). Pasteurization was at 62.8 C - 30 min and immunoglobulins added at 200 μ g protein/ml. Percent germination at 1 and 8 hr is shown in parentheses.

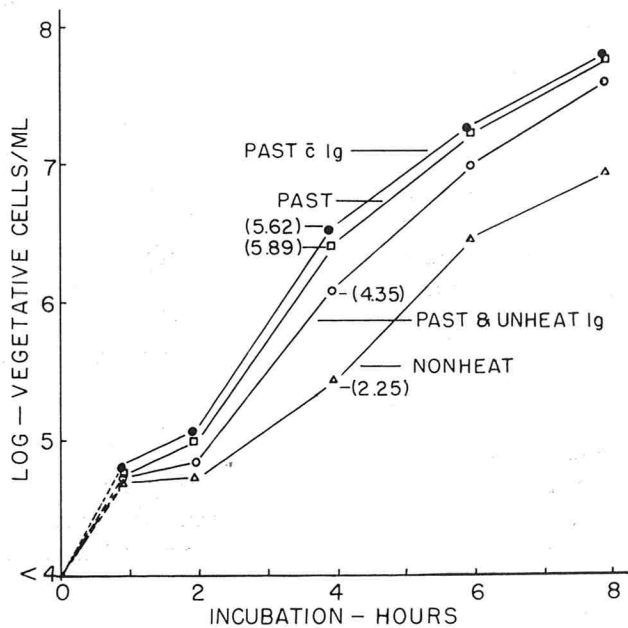


Figure 2. Vegetative cell counts of *Bacillus cereus* 7 in the systems described in Fig. 1. The number of generations at 4 hr is shown in parenthesis.

teurized skim milk. The data present a similar trend to those for germination rates, in that, unheated immunoglobulins had an intermediate depressing effect on cell multiplication. However, unheated skim milk without added immunoglobulins was the most inhibitory to growth.

In an attempt to ascertain the effect of immunoglobulins *per se* on *B. cereus* 7 spore germination

and cell multiplication, a model system was developed. This consisted of 4 ml of protein-free skim milk dialysate (pH 6.5), 8 ml of sterile distilled water, 1 ml of an aqueous suspension of heat-shocked *B. cereus* 7 spores, and a weighed amount of lyophilized D Fraction immunoglobulins to yield 1500 μ g protein/ml. Six systems were prepared: heated (62.8 C - 30 min) and unheated dialysate controls, unheated dialysate fortified with unheated immunoglobulins, dialysate and immunoglobulins heated together, dialysate and immunoglobulins combined after separate heat treatment, and unheated dialysate combined with heated immunoglobulins.

The log of spore counts in the systems with the percent germination at 6 hr is shown in Fig. 3. For the dialysate controls, spore counts were relatively unchanged during the first 6 hr of incubation. In the systems with immunoglobulins, spore counts decreased with incubation and at 6 hr, percentage of germinated spores ranged from 62 in the unheated system to 82 in the totally heated system. Thus, unheated immunoglobulins depressed spore germination when compared with heated samples. Heat treatment of the dialysate *per se* did not influence spore germination: 79% of the spores germinated in

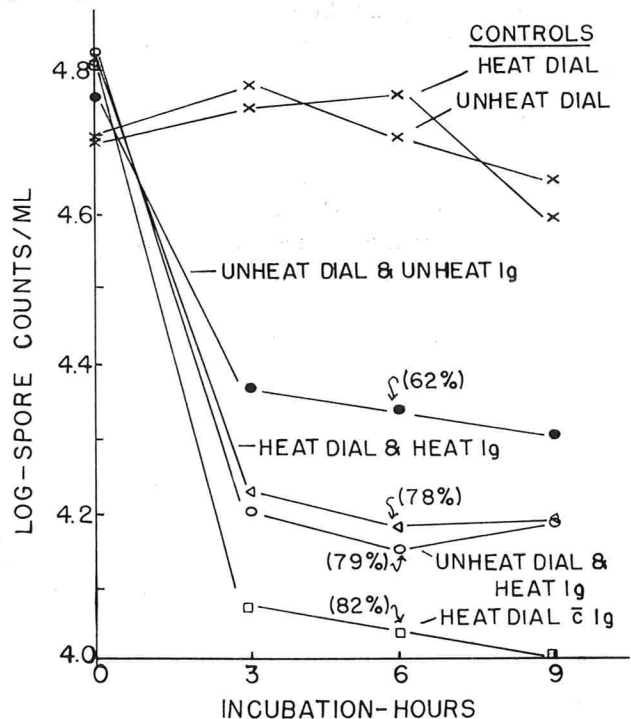


Figure 3. Spore counts of *Bacillus cereus* 7 in model systems composed of unheated or heated protein-free skim milk dialysate (unheat dial or heat dial), unheated dialysate with unheated immunoglobulins (unheat dial & unheat Ig), heated dialysate combined with separately heated immunoglobulins (heat dial & heat Ig), unheated dialysate combined with separately heated immunoglobulins heated together (heat dial \bar{c} Ig). Heat treatments were at 62.8 C - 30 min and immunoglobulins were added at the rate of 1500 μ g protein/ml. The percent germination at 6 hr is shown in parentheses.

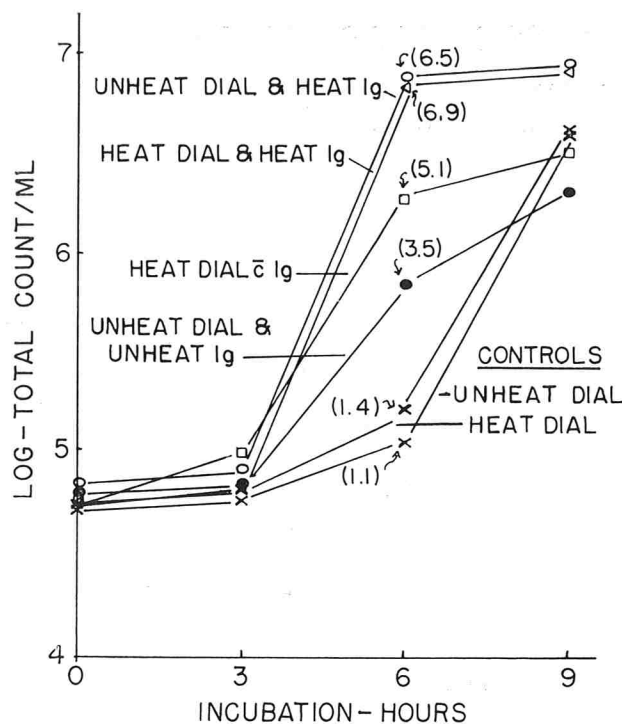


Figure 4. Multiplication of *Bacillus cereus* 7 in the various model systems described in Fig. 3. The number of generations at 6 hr is shown in parenthesis.

the unheated dialysate with heated immunoglobulin system as compared with 82% in the heated dialysate with immunoglobulins and 78% in the separately heated system. In all instances, model systems were less effective germinating media than the native skimmilk.

Rate of multiplication and number of generations of *B. cereus* in model systems is shown in Fig. 4. In systems supplemented with immunoglobulins, the total *B. cereus* counts increased rapidly after an initial 3-hr lag period, whereas in the dialysate control lots, the lag period was longer. The number of generations at 6 hr ranged from 1.1 for the unheated dialysate control to 6.9 for the separately heated dialysate and immunoglobulin system. The number of generations in the unheated dialysate and unheated immunoglobulin system was approximately one-half that observed in the separately heated system, indicating that unheated immunoglobulins depressed the growth of *B. cereus*. Heat treatment of the dialysate did not alter significantly its properties as a growth medium for the organisms.

DISCUSSION

Immunoglobulins in normal, mixed raw skimmilk play only a minor role as inhibitors of *B. cereus* spore germination and subsequent proliferation. The limited bacterial inhibitory properties of the immunoglobulins are further negated upon pasteurization. For *B. cereus*, at least, one must look to other heat-labile agents or factors in skimmilk which will depress

germination and growth of sporeformers. Reiter and Oram (6) have suggested that lactotransferrin is one such agent, being an effective inhibitor of *Bacillus subtilis* and *Bacillus stearothermophilus*. However, lactotransferrin is highly heat stable (90 C - 60 min) and would thus be expected to manifest its activity in pasteurized milk; an effect we have not observed. Smith (9) has shown that lactoferrin is present in dry mammary gland secretions of dairy cows and in some analytical procedures it may be confused with IgG2. It is doubtful that lactoferrin would be present in concentrations necessary to manifest its inhibitory activities in mixed herd milk.

In model systems, where unheated immunoglobulins were the only protein source, spore germination and subsequent growth of the organisms were reduced. However, upon heat treatment of the immunoglobulins *per se*, the inhibitory property of the systems diminished. It is possible that the increased germination and more rapid growth of the organisms resulting from heat treatment of immunoglobulins may relate to a denaturation of these proteins making them more accessible to enzymatic breakdown by the organisms. In some unpublished work, we have observed that heat treatment of blood serum albumin alters its electrophoretic patterns: one major band plus a multiplicity of minor bands appear in the gels. The proteins represented by the minor bands were selectively degraded by *B. cereus* 7 with the major band remaining essentially unchanged. Regardless of effect, unheated immunoglobulins appear to influence multiplication of *B. cereus* to a greater extent than spore germination.

REFERENCES

1. Choudhery, A. K., and E. M. Mikolajcik. 1970. Activity of *Bacillus cereus* proteinases in milk. *J. Dairy Sci.* 53: 363-366.
2. Koka, M., and E. M. Mikolajcik. 1967. A microbial-free milk dialysate. *J. Dairy Sci.* 50:762.
3. Kumar, S., and E. M. Mikolajcik. 1971. Potency of commercial antiovine sera. *J. Dairy Sci.* 54:913-915.
4. Lowry, O. H., N. J. Rosenbrough, A. L. Farr, and R. J. Randall. 1951. Protein measurement with the Folin phenol reagent. *J. Biol. Chem.* 193:265-275.
5. Mikolajcik, E. M., and M. Koka. 1968. Bacilli in milk. I. Spore germination and growth. *J. Dairy Sci.* 51: 1579-1582.
6. Reiter, B., and J. D. Oram. 1967. Bacterial inhibitors in milk and other biological fluids. *Nature* 216:328-330.
7. Singh, V. K., and E. M. Mikolajcik. 1971. Influence of heat treatment of skimmilk upon growth of enteropathogenic and lactic bacteria. *J. Milk Food Technol.* 34:204-208.
8. Smith, E. L. 1946. Isolation and properties of immune lactoglobulins from bovine whey. *J. Biol. Chem.* 165:655-676.
9. Smith, K. L. 1970. The IgG immunoglobulin of bovine lacteal secretion and a possible role of estrogen on IgG concentration. Ph.D. dissertation. Ohio State University, Columbus.

TESTING MILK FOR CLEANLINESS OF PRODUCTION¹

MICHAEL H. ROMAN

New York State Department of Agriculture and Markets
Building 8, State Campus
Albany, New York 12226

ABSTRACT

Clean milk production is an important requisite for high quality. The *mixed* sample sediment test of producer's milk is indicative of production practices and should be renamed "Cleanliness of Production Test." Cleanliness of milk in the farm tank should relate to dairy farm inspection ratings. Standards of < 1.5 mg/gal are practical to meet. Milk can be kept clean if milking preparation and procedures as outlined in regulatory codes are followed. Unclean milk can be only partially "cleaned" by filtration because of fragmentation and solubility of some filths. The test disk is likely to be fouled and may show yellow color when testing highly abnormal milk and thus testing serves a dual purpose. Before-straining testings indicate degree of sediment contamination. Thorough washing and drying of teats with single service material with *no* reuse of solution cannot be overemphasized.

A universal milk sample taken from each milk collection simplifies monitoring for cleanliness using 4 oz sample and 0.2 inch diameter test. Milk cleanliness should be regulated since voluntary programs are subject to many variables. Producers should assume responsibility for clean milk production by conducting their own mixed sample test using sediment test disk.

Numerous articles have been written on the subject of clean milk production and the importance of sediment testing of milk by well versed men from all parts of the country. To quote a few:

Ben Luce, Washington State Department of Agriculture: "I believe sediment in milk is one of the two major problems of the dairy industry today. Ignoring it as we have done for the most part since the advent of the bulk tank does not correct it or make it go away" (8).

Harold J. Barnum, Denver Department of Health and Hospitals: "I consider routine sediment testing a must in any quality and sanitation program. Dirt is an adulterant and is not relished by anyone" (2).

Dr. James C. White, Cornell University: "There is a segment of the dairy industry that does not feel that sediment in milk is a problem. They have come as close to achieving that impossibility, hoping that if they shut their eyes and refuse to recognize it, the problem will disappear" (12).

Dr. John Flake, Evaporated Milk Association: "It is puzzling how such a simple test can cause so much controversy as to its interpretation and merits" (5).

Earl Wright, Iowa State University: "The sediment test is an excellent indication on how well the dairyman cleaned the cows' udders before milking" (13).

Professor B. J. Liska, Purdue University: "There should

be a good practical sediment testing program on the farm before milk is pumped into the tank truck in order to maintain high quality milk supply" (6).

Elmer Kihlstrum, Chicopee Mills: "Sediment tests offer visible evidence that a quality problem exists" (9).

I shall discuss sediment testing of milk and more especially the testing of bulk supplies such as milk in farm tanks, tank trucks, storage tanks and shall include the testing of milk at milking time before straining.

It should not be necessary to discuss why we should strive towards production of sediment-free milk since sanitation is a part and parcel of the American way of life, as evidenced by our many pure food laws.

HISTORY OF SEDIMENT TESTING

Sediment testing of milk is not new. A brief review of history would show that even in the days of "dip" milk, consumers were watchful of the cleanliness of milk dipped out for them by the driver of a horse drawn milk wagon on which were one or more large 40 or 50 gal cans uncovered and exposed to the elements during the dipping operation. The invention of the glass milk bottle made sediment visible on the bottom of the bottle and was perhaps the greatest step forward in milk sediment control. The glass bottle was first accepted with reservation by those selling milk since it made every consumer a potential milk inspector. Cream lines and milk cleanliness could now be observed. The glass milk bottle must have led to the earliest sediment testing of milk. One of the early testing devices consisted of a metal funnel into which was fitted a loosely formed cotton wad through which the test sample of milk was filtered. As more efficient and commercially made test disks became available, the funnel shaped tester was provided with a cover and a hand operated pressure bulb to create air pressure and force milk through the test disk. Next a milk thief-type sample procurer followed. This was equipped with a spring operated foot valve so that the tube would fill with milk from the bottom of the can. The milk in the tube was then poured into a funnel cup for gravity filtration. The milk thief method was improved by development of a suction type pump. This provided for suction removal of the sample and pressure filtration for off-bottom testing of milk in cans. To meet today's need for testing mixed samples of bulk milk,

¹Presented at the 57th Annual Meeting of the International Association of Milk, Food, and Environmental Sanitarians, Cedar Rapids, Iowa, August 17-20, 1970.

various types of pressure and vacuum operated devices have been perfected.

As an aid in classifying milk as to its cleanliness, official photographed test standards (1) have been made available for this purpose so that milk cleanliness may be referred to in terms of milligrams per gallon, such as 0.2 mg, 1.5 mg, or 6.0 mg, etc. These standards are comparative means which can be used for more uniform grading of sediment tests. An electronic densitometer is presently under trial and if it proves satisfactory, it will remove the variable of the human element in estimating from photos the sediment levels of tests. This, in brief, is a history of testing milk for cleanliness and the tools that were used.

A NEW NAME?

Consideration should be given to using the name "Cleanliness of Production" test to designate the common sediment test. This is a more complete description of what a mixed milk sample test of a producer's milk reveals. In milking cows with healthy udders and using clean production methods, the test area of the disk for such a milk sample is almost indiscernible since it is free of sediment or stain. Milk is clean and sediment-free as it leaves the cow's teats, however from this point it is subject to contamination with extraneous matter unless proper milking procedures and care are exercised. Thus if milk is clean in the udder but not clean in the bulk tank or milk can then contamination with extraneous matter occurred during the milking or handling process.

The degree of cleanliness of production can be measured. Contaminants such as manure, barnyard soil, and silt may be fragmented into tiny particles and may have high moisture contents, the liquid portion going into solution in the milk. This liquid along with the fragmented particles cannot be effectively strained out by farm filtration. An unclean test result is a good indication that proper milking and milk handling practices were not followed. One cannot observe actual milking and handling practices during inspections when milking is not in progress, thus the sediment test can serve as an indicator or guide as to compliance by the dairyman with required procedures for clean milk production.

A STANDARD METHOD

It is obvious that cleanly produced milk is not only obligatory to consumers but is also a requisite for high quality. *Standard Methods (1)* recognizes the sediment test as one of the quality tests for milk. Chapter 15 of the 12th edition is devoted to this subject and spells out (a) methods and procedures to be used for sediment testing by the mixed sample and

off-bottom methods, (b) the required features of sediment testing devices to assure that no by-pass occurs while testing the sample, (c) specifications relative to sediment retention for test disks and (d) directions for preparation of grading standards.

It is interesting to note that *Standard Methods (1)* lists the presence of abnormal milk as one of the factors which will influence the rate of flow through a test disk. Our many mixed sample testings have shown this to be a fact. The mixed sample sediment test does serve the dual purpose of not only determining cleanliness but also revealing highly abnormal milk supplies. A milk quality problem likely exists if a properly warmed mixed milk sample cannot be passed through a test disk with vacuum, milk which fouls the disks may be exceedingly unclean or have a high abnormal milk reaction. The sediment level must be very high in the test sample before it fouls the disk and resists passage.

EXPERIENCES IN NEW YORK

I would like to share with you my experiences in New York State during the past 35 years in testing milk for cleanliness. In the late 1930's we started to check on milk cleanliness by actual restraining of a full can of morning's milk as delivered by producers to the plant. A 20 quart strainer with a flannel square clamped to its bottom was used. The flannel squares would then be displayed on a clothes line for producer observation. This method, although very cumbersome, was quite effective in attaining some improvement in milk cleanliness.

Restraining of a whole can was discontinued when the plunger type off-bottom tester was invented. This tester made it possible to test many more cans of milk and thus milk cleanliness programs were given more attention. Food and Drug Administration participation in testing of supplies added further stimulus to the program. The off-bottom testing was practiced for many years and did much to further improve on milk cleanliness. Rejections, exclusions, and penalties had to be resorted to in some instances to effect an improvement. Sediment testing of milk was and continues to be disturbing to some producers and this group needs the sanitarian's assistance in clean milk production.

Studies were conducted in the late 1940's on effectiveness of straining materials and on construction of strainers; thus it is significant the concentration at the time was on cleaning of milk rather than on clean milk production.

During the early days of bulk milk handling, many studies were conducted as to how to test such milk for cleanliness. Experiments were conducted by drawing 5 gal of milk off the bottom of tanks, ob-

TABLE 1. VOLUME-AREA FOR MIXED SAMPLE SEDIMENT TESTING

Volume	Filter area (Diameter in inches)
1 gal	1.125
3 quarts	1.0
2 quarts	0.80
1 quart	0.56
1 pint	0.40
4 oz	0.20

TABLE 2. MILK RESISTANCE TO FILTRATION THROUGH SEDIMENT TEST DISK AND M. W. T. RESULTS¹

No. samples resisting filtration	M.W.T.	
64	Neg.	2
	Trace	5
	1	9
	2	24
	3	24

¹299 individual cow's bucket samples (mixed test sample—1 pint—0.4 inch disc), vacuum used—15 inches.

taining unstirred samples from outlet valves of tanks, and using the off-bottom double length tester. It was about this time that Watson (11) and Calbert and Liska (7) established that the amount of sediment obtained from testing of 1 gal of mixed milk through an 1 1/8th inch diameter test area of a disk approximated that obtained by the 1-pint unstirred method in the off-bottom testing of the same milk when collected in 10 gal cans and allowed to remain quiescent for several hours. This was a most important observation and it established the basis for mixed sample testing. Liska and Calbert also suggested use of smaller milk samples for the mixed sample method and compensated for this by adjusting the size of the test area. Thus if 1-pint of mixed milk is to be tested, the filter area should be 1/8th of that used for a 1 gal sample or an area 0.4 inch in diameter, or if a 4 oz universal milk sample is to be tested, the filter area should be 0.2 inch in order to maintain a constant volume-area relationship (Table 1).

NEW YORK STUDIES

As a result of establishing a method for testing any bulk milk supply for cleanliness, much work has been done by various sanitarian committees and others in New York State to obtain further data on the problem. I would now like to describe the design and results of a few of these studies:

(a) Studies to ascertain adequacy of mixing milk in bulk tanks by mechanical agitators for representative sampling. It has been concluded that if milk is sufficiently mixed for a representative butterfat sample then mixing is adequate for a

sediment sample.

- (b) Determining the effect of the temperature of milk sample at testing on the resulting sediment test. It was found that bulk farm milk must be warmed to 95 to 100 F in order to effect its passage through the test disk and eliminate the minute fat flakes which may foul or mask the disk. However, if a sample has been properly warmed and still resists passage it should be suspected of being abnormal unless it has a very high sediment level. Fouling of disks when applied to clean, freshly produced milk of individual cows relates highly to abnormal milk. Studies have shown that in many instances it is possible to pass several quarts of an individual cow's freshly produced milk through a 0.4 inch diameter sediment test disk, whereas in other instances only a small fraction of 1-pint of clean milk will pass before fouling the disk. Producers have long known that a cow's milk is likely to be abnormal when it fouls the strainer pad. The sediment test disk, being much denser, makes for a much more sensitive test (Table 2).
- (c) Recent studies have been made regarding use of a 4 oz universal milk sample and a 0.2 inch diameter test area to screen milk for cleanliness. It has been found that such a test is very practical for quickly determining the cleanliness of supplies and for rechecks when needed, in that the hauler brings in a sample with each milk collection. This procedure holds much promise for milk quality improvement and for pinpointing sources where improvement is needed. It should place much inspection work on a selective and productive rather than random basis.
- (d) Studies have been made on the behavior of sediment in milk. Contrary to common thinking, sediment does not necessarily settle to the bottom, but depending upon its nature and density, much of it may remain in suspension for hours. In some instances sediment concentration has been found to be greater in the cream portion than in the bottom of the tank or can.
- (e) Since much can milk still exists and the off-bottom test is still an official test in *Standard Methods* and in some codes, comparisons have been made and it has been found that the mixed sample test reveals more instances of unclean milk than does the off-bottom method. Maloney and Armstrong (10) have made a similar observation. This is caused by suspended fine sediment and the fact that most off-bottom tests are conducted on milk which has been agitated

during transportation and unloading when can milk is delivered to a plant and thus are not truly off-bottom unstirred milk tests (Tables 3, 4, 5, and 6).

- (f) Observations have been made regarding the significance of a definite yellow color observed on the back-side of some test disks in mixed sample testing. Choi and Forster (3) and Cole,

TABLE 3. INSTANCES OF WIDE VARIATIONS BETWEEN OFF-BOTTOM AND MIXED SAMPLE TESTS OF CAN MILK SEDIMENT TESTS MADE IMMEDIATELY UPON DELIVERY TO PLANT

Sample No.	Off bottom	Mixed sample
1	0.2 mg	2.0 mg/gal
2	0.2	1.5
3	0.2	3.0
4	0.1	2.0
5	0.2	3.5
6	1.5	6.0

TABLE 4. BEHAVIOR OF SEDIMENT IN MILK CAN MILK UPON DELIVERY TO PLANT MIXED SAMPLE TESTS

Sample	No stirring dipped	3-sec stir	10-sec stir
1	0.5 mg	0.5 mg	0.5 mg
2	3.0	4.0	4.0
3	0.2	0.2	0.2
4	4.0	3.0	3.0
5	0.5	0.2	0.2
6	2.5	3.0	3.0

TABLE 5. SEDIMENT TESTS OF 195 INDIVIDUAL CANS OF MILK BY TWO METHODS. TESTS MADE IMMEDIATELY UPON DELIVERY TO PLANT.

Grade	Off-bottom (1-1/8 inch disc)	Mixed sample after O.B. test (0.4 inch disc)
	----- (Number of tests) -----	
0.5 mg	131	80
0-0.9 mg	151	113
0-1.4 mg	168	139
1.5 or more	27	56
2.5 or more	13	26
3.0 or more	10	15

et al. (4) indicated that yellow color is associated with abnormal milk. Yellow color on disks is a good clue to suggest conducting an abnormal milk test. It becomes even more significant when applied to milk of individual cows or quarters, however all abnormal milk does not show yellow color on disks (Table 7).

- (g) Use of screened inserts when pumping milk out of tanks as a means for determining cleanliness was studied. It has been found that although the inserts have merit and serve a good purpose

TABLE 6. VARIATIONS IN CLEANLINESS OF MILK IN INDIVIDUAL 10 GAL CANS (TESTED IMMEDIATELY UPON DELIVERY TO PLANT)

Producer No.	Off-bottom	Mixed weigh-vat test of all cans
	(mg)	(mg)
1	2.0	6.0
	.2	
	.75	
	9.0	
	2.5	
2	.75	4.5
	1.0	
	.75	
3	2.0	
	1.0	3.0
	.5	
	.5	
	1.0	

in demonstrating careless methods, they may not reveal those instances of unclean milk where sediment nature is of minute character and readily passes through the screen. They should be considered a handy means to check for course types of matter but not a replacement for the sediment test.

- (h) A study was made to determine uniformity in degree of cleanliness of production by individual producers. A study on a group of 50 producers with a surprise monthly mixed sample sediment test over a 6-month period which included pasture and stabling seasons, showed that some produce acceptably clean milk continuously whereas this is not true of others. This is a reasonable conclusion for if a producer is following certain production practices one day, he is likely to follow the same practices each day. The fact that producers are generally consistent with either clean or unclean milk production makes it possible to concentrate on that group needing attention (Table 8).

- (i) A study was made on use of prepared actual milk sediment tests in comparison with photo standards for grading purposes. It has been found that when actual prepared standards are used there is much more uniformity in grading of tests. The newly developed densitometer should make it possible to accurately prepare standards at various levels. Since the color of soil varies in different areas of the country, the densitometer could be used to prepare standards suitable for a particular region. Since we were experiencing some difficulty in attaining uniformity in grading when using photo standards, we prepared an actual set for each of our

TABLE 7. SIGNIFICANCE OF YELLOW COLOR ON SEDIMENT TEST DISCS TO ABNORMAL MILK (INDIVIDUAL COW'S MILK)^{1, 2, 3}

M. W. T.	Samples
1+	4
2+	4
3+	17

¹Number of cow's milk tested—160.

²Discs with yellow color—25

³Average somatic cell count of above 25—2,758,000.

TABLE 8. 6-MONTH STUDY OF 50 PRODUCERS. MIXED SAMPLE TEST METHOD USED

Number of producers	Number of the 6 monthly tests at 1.5 mg/gal or more
Same 24	0
Same 1	1
Same 4	2
Same 5	3
Same 5	4
Same 2	5
Same 2	6

TABLE 9. MILKING TIME INSPECTION—INDIVIDUAL COWS MILK MIXED SAMPLE BEFORE-STRAINING TESTS

Grade	4 selected clean milk producers	4 selected unclean milk producers
----- (Number of samples) -----		
<1.0 mg	67	6
1.0-2.9 mg	26	11
3.0-6.0 mg	6	4
6.0 mg or more	6	67

TABLE 10. SEDIMENT—BEFORE AND AFTER STRAINING PRODUCERS, SELECTED ON BASIS OF CLEAN AND UNCLEAN TESTS IN EACH GROUP

Farms	Average sediment grade ¹	
	Before-straining	After-straining
	(mg)	(mg)
4 Farms (105 cows)	1.0	0.2
4 Farms (88 cows)	7.0	2.2

¹Mixed sample tests.

inspectors using a blend of milk from several sources. The standards were prepared at estimated 1.5 and 3.0 mg levels which are the two critical points in New York State milk cleanliness regulations. When checked with the densitometer, these two disks showed a 1.5 and 3.2 mg level. Accurate grading problems arise only in those instances where the tests are at the cut-off point between acceptable and unacceptable. Disks which are clean or considerable beyond the acceptable grade present no grading problem.

(j) Before-straining testing of milk at milking time has been one of our most important studies.

This was a real breakthrough in clean milk production for we found that some producers were able to produce practically sediment-free milk by good milking practices, whereas this was not true where practices did not follow codes. Many milking time before-straining tests have been conducted and it is obvious that milk must be produced clean if a clean test of the milk after straining is to result. Incomplete an inadequate washing and drying of teats with reuse of the solution for many cows were found to be the greatest contributing sources of sediment. After a number of cows had been prepared for milking with the reuse of the washing solution it became so soiled that a number of test disks would be required to test 1-pint of it for cleanliness. The single use of the cleaning solution preferably using white single service towels soaked in the tepid germicidal solution should be encouraged. If a cow's teats are badly soiled and the white shows soil on drying the cow's teats, washing should be repeated. Keeping udders clipped will facilitate cleaning. The more modern practices for dairy farm operation such as use of loose housing, especially where inadequate bedding exists and the use of drylots for feeding which become muddy from rains and other conditions allowing cows to become badly soiled places greater emphasis on the importance of proper preparation of the cow for clean milk production. Work with before-straining testing has shown that the visible sediment test of strained milk is equivalent to about one-fourth of the original visible sediment contamination plus the moisture content of the contaminants which may be very high. Thus a 2 mg test would mean that the milk originally had about 8 mg of visible sediment plus the contaminants' moisture before straining. There have been instances at our milking time inspections when producers would start milking with high levels of sediment, would show interest in the testing and would soon be producing milk at practically a 0 mg level. If cow's teats are as clean before milking as they are after milking, milk cleanliness problems would be greatly reduced. Tables 9 and 10).

Although the major source for contamination of milk is from improper cleaning and drying of teats and udders, there are other sources which could contribute contamination, such as improper or careless use of the milking machine allowing it to draw in dust and dirt off the floor, dusty stables, unclean farm water,

and dust and dirt settling into cans and farm tanks.

- (k) Another study that was conducted was a pipeline filter survey. Cleanliness tests were made of bulk milk and water supplies at 237 farms with such installations. Information was tabulated as to make of filter holders, type of material used, and milk and water cleanliness results. It was concluded that cleanliness of milk was directly related to cleanliness of production and care at cowside.
- (l) Monitoring cleanliness of milk in farm tank truck loads was another one of the studies. It was found that the universal milk sample makes it possible to quickly monitor the supply contained in the tanker and pinpoint the sources contributing to sediment.

Cleanliness of milk in New York State is regulated. Prior to adoption of regulations in 1961 and amending them in 1968, cleanliness testing was voluntary but suitable progress was not being made either in effecting improvement or bringing about uniform programs in industry. Briefly, the regulations relating to adulteration and cleanliness of milk are as follows: (a) Producer acceptable milk cleanliness level is set at less than 1.5 mg/gal level by the mixed sample test. (b) Monthly testing is required except if a producer has acceptable tests for three consecutive months, his milk may be tested quarterly. (c) Plants must institute additional sediment testing as soon as blended supply is at the 1.0 mg level and continue it until reduced to less than 1.0 mg. (d) The regulations spell out mixed sample volumes and test area diameters: 4 quarts—1.125 inches, 3 quarts—1.0 inch, 2 quarts—0.8 inch, 1 quart—0.56 inch, and 1 pint—0.40 inch. (e) The mixed sample test is the official test for both can and bulk milk. Can milk may be tested by sampling out of weigh vat. (f) Off-bottom tests may be made on can milk and unclean milk rejected on the basis on this test, but mixed sample test must be made as the official test. A clean off-bottom test on can milk is not accepted as an official test. (g) If milk is at 1.5 mg but less than 3.0 mg level, producers must be notified, the milk re-sampled at next collection and if still at 1.5 mg level or above, no further milk accepted until less than 1.5 mg. If test is at 3.0 mg or above, no further milk is

accepted until less than 1.5 mg. With bulk farm milk, this necessitates testing the milk at the farm before recheck acceptance. (h) Tests must be held for 1 month and records for 1 year.

Regulations also have been promulgated which require sediment testing of cream including gathered cream shipped for manufacture. Cleanliness level is established at less than 2.0 mg/gal. The procedure calls for filtering of a mixed 2 oz cream sample through a 0.4 inch diameter area. The cream is liquified and made filterable by mixing the test sample with 2 oz of a 10% sodium citrate solution and warming to 120 F before testing. A rapid improvement in gathered cream cleanliness was noted after just a few tests and producer notification of unsatisfactory results.

REFERENCES

1. American Public Health Association Inc. 1967. Standard methods for the examination of dairy products, 12 ed. American Public Health Association, New York.
2. Barnum, H. J. 1961. Sediment testing—then and now. Colorado Dairy Fieldmen's Annual Conference. Feb. 27.
3. Choi, S. S., and T. L. Forster. 1966. The relationship of yellow color on milk sediment pads to mastitis. *J. Milk Food Technol.* 29:53-56.
4. Cole, E. J., G. MacKay, and H. J. Barnum. 1965. Observations on the yellow color that appears on milk sediment pads. *J. Milk Food Technol.* 28:372-374.
5. Flake, J. C. 1964. University of Wisconsin conference for dairy fieldmen, plant managers and laboratory technicians. Jan. 30-31.
6. Liska, B. J. 1962. Sediment testing of bulk tank milk. *J. Milk Food Technology* 25:48-50.
7. Liska, B. J., and H. E. Calbert. 1954. A study of mixed sample methods of sediment testing using off-the-bottom standards and equipment for possible use with farm bulk holding tanks. *J. Milk Food Technol.* 17:83.
8. Luce, B. 1966. The problem of sediment in milk. *J. Milk Food Technology.* 29:29.
9. Kihlstrum, E. E. 1963. Sediment testing for quality. *Amer. Milk Rev.* June.
10. Maloney, P., and J. G. Armstrong. 1969. Mixed sample tests for extraneous matter in milk. *J. Milk Food Technol.* 32:455-457.
11. Watson, N. E. 1952. A sediment pump for farm tanks. *J. Milk, Food Technol.* 15:43.
12. White, J. C. 1964. Sediment in milk gets a closer scrutiny. *Amer. Milk Rev.* July.
13. Wright, E. 1968. "Fingerprinting" quality milk. *Hoards Dairyman.* Feb. 10.

MICROWAVE OVENS AND THEIR PUBLIC HEALTH SIGNIFICANCE

ROBERT L. ELDER AND WALTER E. GUNDAKER

*Public Health Service
Bureau of Radiological Health
Rockville, Maryland 20852*

(Received for publication March 29, 1971))

ABSTRACT

The number of microwave ovens sold in the United States is expected to increase greatly in the next 2 to 3 years. Recent field surveys have indicated that proper maintenance on the part of the owner or operator and improved servicing play an important role in controlling microwave oven leakage. The Department of Health, Education, and Welfare performance standard for microwave ovens, which will apply to ovens manufactured after October 6, 1971, cannot be truly effective unless the ovens are conscientiously maintained after purchase. The sanitarian has an extremely important role in promoting microwave oven safety, and State and local health workers are urged to take an active part in convincing owners to implement proper maintenance procedures and to practice good sanitation.

A microwave oven is an electronic product which produces energy in the nonionizing microwave region of the electromagnetic spectrum for the purpose of heating or cooking food. The exact number of microwave ovens in use in homes and restaurants around the United States is difficult to pinpoint exactly, but we estimate that there are currently 40 to 50 thousand units in homes and 50 to 80 thousand units in commercial food establishments. It is expected that the number of microwave ovens sold in the United States will increase greatly in the next 2 to 3 years, particularly if the cost to the owner becomes less.

Because microwave radiation does not carry sufficient energy per photon to break chemical bonds, the principal end result of microwave exposure is heating. The detrimental biological effects are associated with both thermal and nonthermal (field) effects. Some researchers believe that microwave absorption in humans raises the temperature and produces an observable effect when the body's thermal compensating mechanism is unable to remove the increased thermal load. Cataracts, temporary sterility, and subcutaneous burns are commonly referred to as thermal effects (2). Nonthermal effects, those which cannot be accounted for by thermal rise in the tissue, are mainly reported in the Russian literature on the subject and are associated with psychological, physiological, and psychoneural responses. Fatigue, headaches, and overall slowness of reaction are typical symptoms reported (2).

The Department of Health, Education, and Welfare has published a performance standard for mi-

crowave ovens which becomes effective on October 6, 1971, for ovens manufactured after that date. The standard limits leakage to 1 mW/cm² prior to acquisition by a purchaser and allows leakage to increase to no greater than 5 mW/cm² throughout the life of the oven. The allowable increase in leakage acknowledges that some degradation in the quality of the oven with wear and tear seems to be technologically unavoidable. The leakage is to be measured at 5 cm by means of a microwave measurement instrument with specific characteristics. The standard also requires two interlocks, one of which must be concealed, and specifies that no object may be inserted into the oven while the oven is "on," when this would allow microwave levels outside the oven to exceed the applicable limits. This particular section of the standard is directed primarily to door screens through which an object, such as a pipe cleaner, can be inserted. An insulated metallic object could act as a receiving antenna on the inside and a transmitting antenna on the outside, thus producing externally high levels of microwave radiation.

FIELD SURVEYS OF MICROWAVE OVENS

An early Bureau of Radiological Health report (7) summarized results of a number of surveys in which radiation leakage from microwave ovens was measured with food loads or water loads placed in the ovens to simulate actual operating conditions. The tests were made under a variety of conditions. Leakage apparently occurred through the oven door seal and also as a result of conditions which permitted the oven to operate when the door was not completely closed. The data in this report were collected by different organizations, and 109 of the 494 units surveyed, or approximately 22%, were reported to emit microwave leakage in excess of 10 mW/cm².

In the fall of 1969, with the availability of a microwave measurement instrument much more suited to surveying microwave ovens, the Narda 8100, the Bureau of Radiological Health conducted a survey of microwave ovens in cooperation with the State Health Departments of Mississippi and Massachusetts and with the Nassau County Health Depart-

ment in New York. Approximately 72 facilities were visited, and 155 ovens made by six manufacturers were surveyed under closely controlled test conditions. Fifty-one of the 155 ovens surveyed, or about 33%, were found to emit radiation in excess of 10 mW/cm² which was stated to be the maximum design leakage standard recognized by the industry. Of the 51 ovens which emitted more than 10 mW/cm², 36 exceeded this amount with the oven door closed and 15 exceeded it only during the act of opening the door. The surveyors noted that 19 ovens had observable defects, such as loose hinges, broken hinges, failed interlocks, and defective doors, which probably contributed to excessive radiation leakage.

In 1970, a combined Federal-State-industry survey was conducted on 4,033 ovens produced by 17 manufacturers. The results confirmed the fall 1969 study, with approximately 20% of the ovens emitting leakage radiation in excess of 10 mW/cm². In those instances in which a specific feature of the oven's design, construction, or adjustment capability was identified as the cause of microwave radiation leakage in excess of the voluntary industry standard of 10 mW/cm² measured at 5 cm, the manufacturer was requested to arrange for corrective servicing or modification of the ovens. In every instance in which a manufacturer was requested to submit a corrective action plan to the Bureau, the manufacturer complied with the request and implemented the plan.

A review of the data from the 1970 survey indicates that:

1. Excessive radiation leakage from microwave ovens seems to be a function of oven design or construction, interlock maladjustment, user maintenance, and frequency of service.
2. The most frequently observed cause of excess leakage was interlock maladjustment or lack of ability to adjust the interlock satisfactorily. In almost all of the oven models surveyed, this condition was readily detected by simple test procedures and could be corrected by manufacturer's or owner's representatives.
3. Oven design or construction appears to contribute more to excessive leakage from ovens manufactured prior to 1970.
4. Proper maintenance on the part of the owner and improved servicing, including a microwave emission measurement, seem to be significant factors in maintaining control over excessive microwave oven leakage.

There is a need for continued surveillance of microwave ovens to monitor the thoroughness of the manufacturers' design, construction, and corrective

action programs and their efforts to improve their field service capability in the area of microwave emission control.

IMPORTANCE OF OVEN SANITATION AND MAINTENANCE

A microwave oven is designed to contain microwave energy and prevent its escape from the oven cavity. Three types of door seals are commonly used by the industry for this purpose. One type, the choke seal, consists of a fractional wavelength trap at the frequency of the oven which absorbs energy leaking from the oven. A second type is the metal-to-metal seal which causes reflection of the energy back into the oven. A third type contains materials commonly called "lossy." These nonreflective absorbers are used to coat surfaces and generally serve as a backup for one of the other types of door seals. There are combinations of these, and each has advantages, according to the overall design of the individual oven.

Any object which accumulates on the seal, such as dirt, grease, or metal, can defeat the purpose of the seal and allow microwave leakage to increase. Hence, basic sanitation is vital to microwave oven safety. The microwave manufacturer is responsible for designing an oven that does not leak, whose seals are easily and safely cleaned, and which will not leak excessively from dirt buildup. Scouring should not be necessary, since any abrasive item used to scrub a seal could reduce the efficiency of the seal to absorb and/or reflect the energy into the cavity. Metal scouring pads in particular should not be used, unless recommended by the manufacture, because residual metal particles can cause arcing and pitting of the seals. Manufacturers are continuing to design improved seals which effectively trap the energy and can be easily cleaned.

A local health official visiting a restaurant does not need an instrument to tell a proprietor that his microwave oven is dirty. If an official observes that an oven has dirty seals, he can assume that the potential for leakage has been increased. Therefore, owners should be encouraged to keep microwave oven seals clean. Costs of instruments to measure microwave leakage at present vary from inexpensive for unreliable devices, such as those with fluorescent bulb detectors, to expensive for highly sophisticated instruments costing nearly \$1000. Instrument costs, however, are expected to drop below \$200 in 1971. A DEP report has been published which describes the design of a simple and inexpensive instrument that can be used by technicians to detect radiation leakage from microwave ovens (3). Several other DEP reports describe a comparison of microwave detec-

tion instruments (5) and related work on microwave measurements (1, 6, 8, 9).

The Bureau of Radiological Health has published a pamphlet entitled *Facts About Microwave Oven Radiation* (4) which lists safety tips for the microwave oven owner and operator. The following list summarizes some of these tips as they apply to the sanitarian.

1. Read the instruction manual for the manufacturer's recommendations for safe operation of the oven.
2. Examine the oven for evidence of shipping damage.
3. Instruct the owner to arrange the installation, if possible, to allow the user to stay at least a full arm's length away from the front of an operating oven.
4. Train the user to switch the oven off before opening the door.
5. Instruct the user never to insert objects (for example, a fork prong, aluminum foil, wire) through the door grill (if there is one) or around the door seal.
6. Instruct all users and maintenance personnel never to tamper with or inactivate the oven safety interlocks.
7. Instruct the owner or user to clean the oven cavity, door, and seals frequently with water and mild detergent, and to refrain from using pads, steel wool, or other abrasives unless recommended by the manufacturer.
8. Instruct the owner to have each oven examined at regular intervals by a qualified serviceman for signs of wear, damage, or tampering, and to have it adjusted, repaired, and measured for excessive leakage.

During the joint industry/Bureau survey of microwave ovens, we discovered that many cases of excessive leakage could be caused by poor sanitation. One hospital, for example, reported that 26 ovens which leaked more than 10 mW/cm² before proper cleaning, emitted no measurable radiation after cleaning; this situation is typical of the metal-to-metal seal. In addition to sanitation, there are other readily observable problems which will indicate the potential for unnecessary leakage. If an oven door appears to be loose, indicating excessive hinge wear, the door may not be giving a tight seal. If the door interlock or cooking indication light appears to be malfunctioning, there may be an interlock failure. In all instances only specially trained repairmen should make tests or adjustments or repair ovens. The Division of Electronic Products is encouraging manufacturers to increase their repair capabilities for safety and encourages the support of every sanitarian in helping these same manufacturers transmit information to the repairman and oven owner concerning basic sanitation.

During the recent industry/Bureau survey of microwave ovens, about 23% of one manufacturer's ovens were found to be leaking microwave radiation in excess of 10 mW/cm². However, when 100 of the

same models were surveyed in a single city, only 6% were found to leak in excess of 10 mW/cm². Further investigation showed that these were owned and operated by a vendor who had a well-run maintenance program. Most of the ovens surveyed had been serviced about 2 or three months before the survey as part of a routine program.

Microwave ovens can be designed, constructed, and used safely and are practically a necessity in the rapid vending of food. Food vendors are making a major effort to purchase good, durable ovens and reliable microwave measurement instruments to make certain that the microwave ovens they use in their food delivery service are safe for public use. These efforts should continue to increase in order that continued, safe use of microwave ovens can be realized. Sanitation, maintenance, and continual safety monitoring can and will accomplish this for ovens which are designed and constructed to operate safely.

REFERENCES

1. Bendix Corporation Research Laboratories. 1970. Development of liquid crystal microwave power density meter. BRH/DEP 70-8. USDHEW, PHS, BRH, Rockville, Md. (Accession No. PB 191 396).*
 2. Cleary, S. F. 1970. Biological effects of microwave and radiofrequency radiation. Critical Reviews in Environmental Control, The Chemical Rubber Company, Vol. 1, Issue 2.
 3. Cloke, R. L. 1970. Inexpensive readout for a commercial thermocouple microwave power density probe. USDHEW, PHS, BRH, Rockville, Md. BRH/DEP 70-31. (Accession No. PB 196 440).*
 4. Facts About Microwave Oven Radiation. Pamphlet BRH/P-4. USDHEW, PHS, BRH, Rockville, Md. 20852.
 5. Moore, R. L., and S. W. Smith. 1970. A comparison of microwave detection instruments. BRH/DEP 70-7. USDHEW, PHS, BRH, Rockville, Md. (Accession No. PB 191 395).*
 6. Radiation Measurements and Calibration Branch. 1970. The effect of instrument averaging time on microwave power density measurements. BRH/DEP 70-12. USDHEW, PHS, BRH, Rockville, Md. (Accession No. PB 192 322).*
 7. Rosenstein, M., W. A. Brill, and C. K. Showalter. 1969. Radiation exposure overview-microwave ovens and the public. OCS 69-1, USDHEW, PHS, BRH, Rockville, Md. (Accession No. PB 188 690).*
 8. Rudge, A. W., and R. M. Knox. 1970. Near field instrumentation. BRH/DEP 70-16. USDHEW, PHS, BRH, Rockville, Md. (Accession No. PB 192 748).*
 9. Swicord, M. L. 1971. Microwave measurements and new types of detectors for evaluation of health hazards. BRH/DEP 71-1. USDHEW, PHS, BRH, Rockville, Md. (Accession No. PB 197 715).*
- *Available from National Technical Information Service, Springfield, Va. 22151. \$3.00 for paper copy, \$0.95 for microfiche. The accession number should be cited when ordering.

VIBRIO PARAHAEMOLYTICUS—A REVIEW¹

R. NICKELSON AND C. VANDERZANT

Animal Science Department
Texas A&M University, College Station 77843

(Received for publication February 26, 1971)

ABSTRACT

This review presents current information on the taxonomic position, biochemical characteristics, distribution, isolation and identification procedures, pathogenicity, and serology of *Vibrio parahaemolyticus*. In the past, *V. parahaemolyticus* was associated primarily with outbreaks of gastroenteritis in Japan caused by consumption of seafoods and other salted foods. In recent years, this organism has been isolated from marine environments and seafoods in many countries, including the United States. In addition to gastroenteritis, some strains may cause localized tissue infections in humans and cause death of crab and shrimp. In the United States, *V. parahaemolyticus* has been incriminated in unconfirmed outbreaks of foodborne illness associated with consumption of shellfish. Isolation procedures based on direct plating of food homogenates on selective media with or without prior enrichment in broth media are available. Suspect colonies are confirmed by biochemical tests and fluorescent antibody technique. Although antisera (7 polyvalent and 47 monovalent) for serological grouping of strains of *V. parahaemolyticus* are available, their usefulness in diagnostic procedures is at the present uncertain.

Vibrio parahaemolyticus is the causative agent of an infectious food poisoning syndrome associated with warm summer months and consumption of raw seafood, particularly in Japan (31). Recent isolations of this organism from marine environments and seafoods in the United States and other countries have prompted many studies on the isolation, identification, and public health significance of this organism. The purpose of this review is to present current information on various aspects of *V. parahaemolyticus*.

TAXONOMICAL HISTORY

Vibrio parahaemolyticus as it is now known was first isolated by Fujino et al. (11) in 1950 and designated *Pasteurella parahemolytica*. The organism caused gastroenteritis in 272 persons resulting in 20 deaths. "Shirasu" (the fry of sardine boiled in salt water and sold and eaten in the half-dried state) was the contaminated food product eaten by all who had acute gastroenteritis. The major symptoms observed were abdominal pains, vomiting, diarrhea, chills, headache, and dyspnea. The organism was placed

in the genus *Pasteurella* because of its bipolar staining and fastidious nature on ordinary culture media.

In 1955, "Asazuke" (salted chopped cucumber) was incriminated in an outbreak of gastroenteritis involving 120 persons with no deaths. Takikawa (28) classified the causative agent as *Pseudomonas enteritis*. He noted that the characteristics of his isolate were similar to the culture isolated by Fujino et al. (11), but changed the genus designation to *Pseudomonas* based on biochemical properties and growth on media containing salt. This was the first indication of the halophilic nature of this organism.

Because of serological differences between this organism and *Pseudomonas*, Miyamoto et al. (18) proposed the generic name *Oceanomonas*. They made a detailed study of the characteristics of their cultures isolated from mackerel incriminated in foodborne illness and those isolated by Fujino et al. (11) and Takikawa (28). All cultures were able to utilize glucose fermentatively with or without production of gas. This suggested that they should be placed in the genus *Vibrio* or *Aeromonas*. *Vibrio* was not considered since pathogens such as *Vibrio cholerae* grow on 3% NaCl and on ordinary culture media. A survey indicated that the natural habitat of this organism was a marine environment. Three species were recognized based on the ability to utilize chitin and alginate, resistance to desoxycholate, citrate utilization, and sucrose fermentation. *Oceanomonas parahaemolytica* (*Pasteurella parahaemolytica* Fujino) was negative for all the above tests. *Oceanomonas enteritidis* (*Pseudomonas enteritis* Takikawa) was positive for chitin, desoxycholate, and citrate. *Oceanomonas alginolytica* (*Pseudomonas enteritis* Takikawa's serotype XII) was positive for all tests.

Sakazaki et al. (24) discarded the generic designation *Pasteurella* on the basis of halophilism, *Pseudomonas* on the basis of fermentative glucose utilization, and *Oceanomonas* because it would incorporate many heterogenous organisms possessing different properties. In an attempt to clarify the taxonomical position of these organisms, they made an extensive study of many of the halophilic organisms in question. Among those studied were the strains of Fujino et al. (11) and of Takikawa (28). They reported (24) that these organisms were closely related to the genus *Vibrio*. They recognized three subgroups

¹Published with the approval of the Texas Agricultural Experiment Station, College Station. This work was funded in part by the NSF Sea Grant Program Institutional Grant GH-101 made to Texas A&M University.

based on differences of growth in peptone water containing 7 and 10% NaCl; Voges-Proskauer reaction; and fermentation of sucrose, arabinose, and cellobiose. Subgroup 3 resembled *Vibrio anguillarum*, which did not grow in 7 or 10% NaCl. Subgroups 1 and 2 were designated as *V. parahaemolyticus*. Cultures of subgroup 2 grew in 7 and 10% NaCl, whereas those of subgroup 1 grew in 7% NaCl only. Almost all cultures of subgroup 1 were isolated from feces of human patients affected with gastroenteritis, those of subgroup 2 primarily from sea fish and seawater.

The differences between subgroups 1 and 2 were confirmed by Zen-Yoji et al. (43). They reported differences in pathogenicity between the two groups. Subgroup 1 was isolated frequently from patients with unidentifiable enteritis. The occurrence of subgroup 2 in patients with gastroenteritis was not significantly higher than in persons without this illness. They concluded that subgroup 2 was not pathogenic to man.

Prompted by the investigation of Zen-Yoji et al. (43), Sakazaki (23) re-examined 100 cultures of each subgroup. He confirmed the results reported by Zen-Yoji et al. (43). Cultures of subgroup 2 grew in 10% NaCl, fermented sucrose and produced acetoin, whereas those of subgroup 1 did not. Because of these differences, he proposed the specific name *Vibrio alginolyticus* for biotype 2 (subgroup 2). The organisms of biotype 1 (subgroup 1) continued to be classified as *V. parahaemolyticus*.

MICROBIOLOGICAL AND BIOCHEMICAL PROPERTIES

Sakazaki et al. (24) were the first to present a detailed description of *V. parahaemolyticus*. Many reports contain scattered information concerning the properties of this organism (3, 4, 5, 6, 9, 11, 24, 28, 36, 38, 40). The most recent and comprehensive investigation of Japanese strains and related cultures isolated in the United States was made by Twedt et al. (36). The following list of characteristics is compiled from reports of many investigators. Morphologically the organism is a short, gram-negative rod exhibiting pleomorphism. Slightly curved, straight, coccoid, and swollen forms can be observed. All strains of *V. parahaemolyticus* are motile by means of a single polar flagellum. On agar plates most cultures appear as smooth, moist, circular, opaque colonies with entire edges. Some rough variants have been reported in pure cultures (36). A swarming phenomenon occurs in some instances when low concentrations of agar are used. This diminishes with increased concentrations of agar.

Vibrio parahaemolyticus grows readily at temperatures from 22 to 42 C, but fails to grow at 2 C.

Growth has been observed at pH values from 5 to 11 and at NaCl concentrations from 1 to 7%. Temmyo (30) noted that *V. parahaemolyticus* was killed by heating at 55 (10 min) and 60 C (5 min) in a liquid medium.

The following biochemical characteristics have been reported for *V. parahaemolyticus*.

- Starch hydrolysis + (4, 5, 6, 9, 11, 28, 36)
- Acetoin production - (5, 9, 28, 36, 38, 40); variable (24)
- Hemolysis + (4, 5, 9, 11, 36, 38, 40); slight (28)
- Triple Sugar Iron Agar alk/acid, gas-, H₂S- (9, 38, 40)
- Growth in 0% NaCl - (4, 5, 9, 36, 38)
- 3% NaCl + (4, 5, 9, 36, 38)
- 7% NaCl + (4, 5, 9, 24, 36, 38, 40)
- 10% NaCl - (4, 5, 9, 24, 38); variable (36)
- Indole + (6, 9, 11, 24, 28, 36, 38, 40)
- Methyl Red + (5, 6, 9, 24, 38, 40); - (28)
- Citrate utilization + (6, 9, 28, 36, 38, 40); - (24)
- Phenylalanine deaminase - (9, 38)
- Nitrate reduction + (4, 5, 6, 9, 11, 24, 28, 36, 38, 40)
- Urease - (6, 9, 24, 36, 38, 40)
- Gelatin liquefaction + (3, 4, 5, 6, 9, 11, 24, 28, 36, 38, 40)
- Catalase + (9, 24, 28, 36, 38, 40)
- Malonate - (6, 9, 24, 36, 38, 40)
- Oxidase + (4, 5, 9, 24, 36, 38, 40)
- Cholera-Red - (9, 24); + (36); variable (38)
- Fermentation of glucose (Hugh-Leifson) + Gas-(3, 4, 9, 11, 24, 28, 36, 38, 40)
- Alginate - (24)
- Luminescence - (24, 40)
- Pigment - (24)
- Decarboxylase lysine + (3, 36)
- Arginine - (36)
- Ornithine - (40)
- Sensitivity to pteridine 0/129 + (3, 5, 24, 40)
- Chitin hydrolysis + (4, 5, 24)
- Sensitivity to penicillin - (4, 5, 24)
- NH₃ from arginine - (3, 40)
- Fermentation of: (Acid no gas)
- Cellobiose - (4, 5, 9, 24, 38); variable (36)
- Sucrose - (4, 5, 6, 9, 11, 28, 36, 38, 40); variable (24)
- Arabinose + (11, 36, 40); variable (24, 28, 38)
- Maltose + (4, 9, 11, 24, 28, 36, 38)
- Mannitol + (4, 6, 9, 24, 28, 36, 38, 40)
- Trehalose + (9, 24, 28, 36, 38)
- Lactose - (4, 6, 9, 11, 24, 28, 36, 38, 40)
- Rhamnose - (9, 11, 24, 28, 38)
- Xylose - (6, 9, 11, 24, 36, 38)
- Adonitol - (6, 9, 11, 24, 36, 38)
- Inositol - (3, 6, 9, 11, 24, 28, 36, 38)
- Sorbitol - (9, 11, 24, 36, 38)
- Salicin - (6, 9, 24, 28, 36, 38); +(11)
- Galactose + (11, 28, 36)
- Mannose + (6, 11, 28, 36)
- Raffinose - (11, 28, 36)
- Fructose - (11, 28); + (36)
- Dulcitol - (11, 24, 28, 36, 40)
- Inulin - (28)
- Melibiose (36)
- Utilization as sole source of carbon:
- Glucose + (36)
- Fructose + (36)
- Sorbitol variable (36)
- Mannitol + (36)
- Sorbose - (36)

Mannose + (36)
 Galactose + (36)
 Dulcitol - (6, 36)
 α - Methyl-D-glucoside - (36)
 Potassium gluconate + (36)
 Arabinose variable (36)
 Adonitol - (36)
 Xylose - (36)
 Rhamnose - (36)
 Erythritol - (36)
 Glycerol + (36)
 Maltose + (36)
 Lactose - (36)
 Sucrose - (36)
 Cellobiose - (36)
 Melibiose - (36)
 Raffinose - (36)
 Salicin - (36)
 Inositol - (36)
 Trehalose + (36)
 Melezitose - (36)
 Lactate + (24, 36)
 Gluconate + (36)
 Pyruvate + (24); - (36)
 Acetate + (24); variable (36)
 Formate - (24, 36)
 Citrate + (36)
 Succinate + (24, 36)
 Malonate - (24, 36)
 Malate + (24, 36)
 Oxalate - (24, 36)
 DL-Phenylalanine - (24, 36)
 L-Tryptophane - (24, 36)
 Benzoate - (36)
 Phenol - (24, 36)

Utilization in complex medium:

Alginate - (36)
 Malonate variable (36)
 Tartrate variable (36)
 Citrate variable (36)
 Mucate variable (36)

More sophisticated procedures also have been used to characterize *V. parahaemolyticus*. Colwell (8) subjected various marine *Vibrio* species, *V. cholerae*, *V. parahaemolyticus*, and other related organisms to 200 different morphological, physiological, and biochemical tests. Using numerical taxonomy she was able to separate *V. parahaemolyticus* from the other organisms. *Vibrio parahaemolyticus* species clustered at $S \geq 80\%$. DNA base composition for all *Vibrio* species was 40 to 48% guanine plus cytosine. Little relation existed between *V. cholerae* and *V. parahaemolyticus*. The lack of relationship between these two human pathogens was confirmed by Citarella and Colwell (7) with DNA reassociation reactions.

DISTRIBUTION OF *V. parahaemolyticus*

For many years isolations of *V. parahaemolyticus* seemed to be limited to Japan where it is responsible for 70% of the gastroenteritis (22). Miyamoto et al. (19) investigated the seasonal distribution of this organism in Sagami and Tokyo Bays. They observed

a "winter-summer type" distribution. The avirulent stenohaline type was predominant in the winter and the virulent euryhaline type in the summer season. Euryhaline types withstood wider ranges of salinity and therefore were more predominant in coastal waters.

In addition, the organism has been isolated in Germany, the Far East, Hawaii, Hong Kong, Taiwan, Singapore, the Phillipines, and in the Netherlands (9, 12, 36). *Vibrio parahaemolyticus* has been studied more extensively in the United States in recent years. Organisms resembling *V. parahaemolyticus* were isolated from Gulf and South Atlantic Coast water and sediment samples (41). Baross and Liston (4) isolated *V. parahaemolyticus* from seawater, sediments, and shellfish of Puget Sound in the Northwest Pacific. They reported a seasonal distribution similar to that reported in Japan by Miyamoto et al. (19). Large numbers were isolated in the summer, but counts were very low in early spring and early fall. It also has been isolated from moribund and lethargic blue crab in Chesapeake Bay (14), shrimp from the Gulf of Mexico (40), Pacific oysters (5), processed meat of Chesapeake Bay blue crab (9), Gulf Coast oysters, and cooked crab meat sold in the Dallas area (15). In the United States in 1969, *V. parahaemolyticus* was incriminated in two unconfirmed outbreaks of food-borne illness associated with the consumption of shellfish (37). Seventy-one persons were involved in these outbreaks. Twedt et al. (36) also studied human isolates (CDC cultures) from wound infections associated with marine bathing. The distribution of *V. parahaemolyticus* appears ubiquitous in marine environments.

ISOLATION PROCEDURES FOR *V. parahaemolyticus*

Isolation protocols for *V. parahaemolyticus* are almost as numerous as reports of its isolation. Earlier isolations were on media such as Salmonella-Shigella or nutrient agar containing 4% NaCl (11, 28). In 1965 Zen-Yoji et al. (43) used a brom thymol blue teepol agar for the isolation of *V. parahaemolyticus*. The Food and Drug Administration now uses a combination of Japanese methods and methods proposed by Liston and Baross (38). The Japanese phase employs two enrichment broths, Glucose Salt Teepol Broth (GSTB) and Salt Colistin Broth (SCB) which are then streaked on Thiosulfate Citrate Bile Salts Sucrose agar (TCBS). The Liston-Baross method is more quantitative in that it is a direct plating medium. In addition to common nutrients, this medium contains salt water and starch and is incubated anaerobically. In the identification protocol, the Japanese method emphasizes differential tests such as reactions in Triple Sugar Iron Agar, Voges-Pro-

kauer reaction, nitrate reduction, etc. The Liston-Baross method stresses type of hemolysis and pleomorphism. Kampelmacher et al. (12) studied five different enrichment broths and three different isolation media. The best enrichment was a meat broth with 5% NaCl. This medium then was streaked on Teepol Brom Thymol Blue agar (BTB) and TCBS agar. Twedt et al. (35) developed a selective plating medium consisting of 2% peptone, 0.2% yeast extract, 0.5% corn starch, 3% NaCl, 1.5% agar, and Penicillin G (5 $\mu\text{g}/\text{ml}$). The medium was adjusted to pH 8.0. Average recovery of *V. parahaemolyticus* from buffered suspensions was 82%. Recently, Vanderzant and Nickelson (39) reported a direct plating procedure with or without preliminary enrichment. Enrichment was in Trypticase Soy Broth with 7% NaCl with incubation at 42 C for 18 hr. The plating medium consisted of 0.2% peptone, 0.2% yeast extract, 1% corn starch, 7% NaCl, and 1.5% agar. The pH was adjusted to 8.0. Plates were incubated aerobically at 42 C for 48 hr. Smooth, white-creamy, circular, amylase-positive colonies were picked as suspect *V. parahaemolyticus*. Confirmation of gram-negative, fermentative, oxidase-positive, pleomorphic rods was made by fluorescent antibody technique.

PATHOGENICITY OF *V. parahaemolyticus*

Vibrio parahaemolyticus is the causative agent of a food poisoning syndrome, related primarily to the consumption of seafood. Several case histories are described (1, 11, 26, 28, 42). The usual symptoms in order of frequency are: diarrhea, abdominal pain, nausea, pyrexia, vomiting, and chills. Incubation period before onset of symptoms is reported from 9 to 25 hr (28, 42). In an experimental case, onset of illness was observed in 3.5 hr (28) and in an accidental laboratory infection, the time was 6 hr (26). The speed with which symptoms occur suggests the presence of an exotoxin, however, only live cells have produced the disease syndrome making it an infectious disease (28, 42). Hemolytic activity ("Kanagawa phenomenon") has been closely correlated with human pathogenicity (17, 26). This hemolytic activity results from a direct thermolabile hemolysin (10).

Pathogenicity for mice has been demonstrated in several instances (1, 11, 24, 28, 40). Death in mice seems to be caused by a septicemia. The "De-Test," using ligated rabbit gut inoculated with *V. parahaemolyticus* produced enteritis at the loop of the gut (2, 24). In experimental infections, *V. parahaemolyticus* is also reported as pathogenic for dogs and cats (1, 28). Marine *Vibrio* species are known causes of marine animal diseases (2, 13, 21, 27, 34). *Vibrio parahaemolyticus* has caused deaths in the blue

crab (*Callinectes sapidus*) (14) and Gulf of Mexico shrimp (*Penaeus aztecus*) (40). Organisms similar to *V. parahaemolyticus* also have been isolated from human skin infections. These patients had a history of marine bathing (36).

SEROLOGY OF *V. parahaemolyticus*

Fujino et al. (11) and Takikawa (28) observed no reaction between their isolates and sera from recovering patients. Sakazaki et al. (24) noted that H antigens might be a better indication of phylogenetic relationships. O-Antigen relationships were noted among the genera *Escherichia*, *Salmonella*, *Proteus*, *Citrobacter*, and *Enterobacter*.

It had been noted that living cultures were not agglutinated by homologous O antiserum, but if the cultures were heated at 100 C for 1 to 2 hr strong agglutination was observed (1, 24). This was interpreted by Omori et al. (20), to be caused by a K-antigen that interfered with the O-antigen reaction. Immunochemical and biological properties of this K-antigen were studied by these investigators.

The first extensive study of all antigens was made by Sakazaki et al. (25). With 2,720 cultures of *V. parahaemolyticus* they established 11 O and 41 K antisera. Production of H-antigen and antiserum was difficult. A close H-antigen relationship between vibrios was noted. With the cultures studied, no one K antiserum occurred in two or more O groups. The determination of O antigen was time-consuming. For these reasons, it was recommended that only K antigen determinations be made in routine diagnostic work. No significant antigenic relationships between *V. parahaemolyticus* and *V. cholerae* including the so called NAG vibrios was found.

Preparation, specificity, and chemical nature of *V. parahaemolyticus* O-antigen were reported by Torii et al. (32, 33). This antigen was a lipopolysaccharide. The envelope fraction of *V. parahaemolyticus* is reported to contain an unusual α -1,4-D-glucan (29). This glucan (V-glucan) differs from glycogen in that it is not sensitive to α - or β -amylase.

Twedt et al. (36) made a serological comparison of Japanese strains, isolates from human sources, and some non-pathogenic marine vibrios. The serological comparisons and biochemical tests strongly suggested that the wound isolates (human sources) were *V. parahaemolyticus* species.

A common antigenic substance of *V. parahaemolyticus* was isolated and purified by Miwatani et al. (16). This antigen was designated as A-substance and was common to strains of *V. parahaemolyticus* but not to *V. cholerae* or *V. alginolyticus*. The substance was found to be different from O, K, or H antigens of *V. parahaemolyticus*. A serological kit

with 7 polyvalent and 47 monovalent K antisera is commercially available (Nichimen Company, New York, manufactured at Toshiba Inst. of Biological Science, Japan). The usefulness of antisera in diagnostic procedures is at the present questionable.

This review of literature indicates that much recent information is available related to the morphological and biochemical characteristics of *V. parahaemolyticus*. Although several isolations have been reported from marine environments and seafoods in the United States, little is known about its presence in other salted foods and about its significance in foodborne illnesses in this country. Additional information in these areas is needed.

REFERENCES

1. Aiso, K., and M. Matsuno. 1961. The outbreaks of enteritis-type food poisoning due to fish in Japan and its causative agent. *Jap. J. Microbiol.* 5:337-346.
2. Akazawa, H. 1968. Bacterial disease of marine fishes. In Symposium on bacterial fish diseases. *Bull. Jap. Soc. Sci. Fish.* 34:271-272.
3. Bain, N., and J. M. Shewan. 1968. Identification of *Aeromonas*, *Vibrio*, and related organisms. pp. 79-84. In B. M. Gibbs and D. A. Shapton (ed.) Identification methods for microbiologists. Part B. Academic Press. New York.
4. Baross, J., and J. Liston. 1968. Isolation of *Vibrio parahaemolyticus* from the Northwest Pacific. *Nature (London)* 217:1263-1264.
5. Baross, J., and J. Liston. 1970. Occurrence of *Vibrio parahaemolyticus* and related hemolytic vibrios in marine environments of Washington State. *Appl. Microbiol.* 20:179-186.
6. Carpenter, K. P., J. M. Hart, J. Hatfield, and G. Wicks. 1968. Identification of human vibrios and allied organisms. pp. 9-18. In B. B. Gibbs and D. A. Shapton (ed.) Identification methods for microbiologist. Part B. Academic Press. New York.
7. Citarella, R. V., and R. R. Colwell. 1970. Polyphasic taxonomy of the genus *Vibrio*: Polynucleotide sequence relationships among selected *Vibrio* species. *J. Bacteriol.* 104:434-442.
8. Colwell, R. R. 1970. Polyphasic taxonomy of the genus *Vibrio*: Numerical taxonomy of *Vibrio cholerae*, *Vibrio parahaemolyticus* and related *Vibrio* species. *J. Bacteriol.* 104:410-433.
9. Fishbein, M., I. J. Mehlman, and J. Pitcher. 1970. Isolation of *Vibrio parahaemolyticus* from the processed meat of Chesapeake Bay blue crabs. *Appl. Microbiol.* 20:176-178.
10. Fujino, T., T. Miwatani, Y. Takeda, and A. Tomaru. 1969. A thermolabile direct hemolysin of *Vibrio parahaemolyticus*. *Biken J.* 12:145-148.
11. Fujino, T., Y. Okuno, D. Nakada, A. Aoyama, K. Fukai, T. Mukai, and T. Ueho. 1953. On the bacteriological examination of Shirasu-food poisoning. *Med. J. Osaka Univ.* 4:299-304.
12. Kampelmacher, E. H., D. A. A. Mossel, L. M. van Noorle Jansen, and H. Vincentie. 1970. A survey on the occurrence of *Vibrio parahaemolyticus* on fish and shellfish, marketed in the Netherlands. *J. Hygiene* 68:189-196.
13. Kiehn, E. D., and R. E. Pacha. 1969. Characterization and relatedness of marine vibrios pathogenic to fish: deoxyribonucleic acid homology and base composition. *J. Bacteriol.* 100:1248-1255.
14. Krantz, G. E., R. R. Colwell, and E. Lovelace. 1969. *Vibrio parahaemolyticus* from the blue crab *Callinectes sapidus* in Chesapeake Bay. *Science* 164:1286-1287.
15. Landry, W. L., B. Wentz, and M. Fishbein. 1970. Recovery of *Vibrio parahaemolyticus* during a market survey of Gulf Coast seafoods in the Dallas area. Paper presented at the Fall Meeting of the Texas Branch, ASM. Dallas.
16. Miwatani, T., S. Shinoda, H. Nishimune, M. Okada, Y. Takeda, and T. Fujino. 1969. A common antigenic substance of *Vibrio parahaemolyticus*. I. Isolation and purification. *Biken J.* 12:97-106.
17. Miyamoto, Y., T. Kato, Y. Obara, S. Akiyama, K. Takizawa, and S. Yamai. 1969. In vitro hemolytic characteristic of *Vibrio parahaemolyticus*: Its close correlation with human pathogenicity. *J. Bacteriol.* 100:1147-1149.
18. Miyamoto, Y., K. Nakamura, and K. Takizawa. 1961. Pathogenic halophiles. Proposals of a new genus "*Oceanomonas*" and of the amended species names. *Jap. J. Microbiol.* 5:477-486.
19. Miyamoto, Y., K. Nakamura, and K. Takizawa. 1962. Seasonal distribution of *Oceanomonas* spp., halophilic bacteria, in the coastal sea. Its significance in epidemiology and marine industry. *Jap. J. Microbiol.* 6:141-158.
20. Omori, G., M. Iwao, S. Iida, and K. Kuroda. 1966. Studies on K antigen of *Vibrio parahaemolyticus* I. Isolation and purification of K antigen from *Vibrio parahaemolyticus* A55 and some of its biological properties. *Biken J.* 9:33-43.
21. Pacha, R. E., and E. D. Kiehn. 1969. Characterization and relatedness of marine vibrios pathogenic to fish: physiology, serology, and epidemiology. *J. Bacteriol.* 100:1242-1247.
22. Sakazaki, R. 1965. *Vibrio parahemolyticus*. A non-cholero-genic enteropathogenic vibrio. In Proceedings of the Cholera Research Symposium, 1965, (U. S. Dept. Health, Education and Welfare, Washington, D. C.).
23. Sakazaki, R. 1968. Proposal of *Vibrio alginolyticus* for the biotype 2 of *Vibrio parahaemolyticus*. *Jap. J. Med. Sci. Biol.* 21:359-362.
24. Sakazaki, R., S. Iwanami, and H. Fukumi. 1963. Studies on the enteropathogenic, facultatively halophilic bacteria, *Vibrio parahaemolyticus*. I. Morphological, cultural, and biochemical properties and its taxonomical position. *Jap. J. Med. Sci. Biol.* 16:161-188.
25. Sakazaki, R., S. Iwanami, and K. Tamura. 1968. Studies on the enteropathogenic, facultatively halophilic bacteria, *Vibrio parahaemolyticus*. II. Serological characteristics. *Jap. J. Med. Sci. Biol.* 21:313-324.
26. Sakazaki, R., K. Tamura, T. Kato, Y. Obara, S. Yamai, and K. Hobo. 1968. Studies on the enteropathogenic, facultatively halophilic bacteria, *Vibrio parahaemolyticus*. III. Enteropathogenicity. *Jap. J. Med. Sci. Biol.* 21:325-331.
27. Smith, I. W. 1961. A disease of finnock due to *Vibrio anguillarum*. *J. Gen. Microbiol.* 24:247-252.
28. Takikawa, I. 1958. Studies on pathogenic halophilic bacteria. *Yokohama Med. Bull.* 9:313-322.
29. Tamura, T., T. Fujino, H. Miyaji, A. Misaki, and S. Kotani. 1969. A highly branched α -D-glucan of *Vibrio parahaemolyticus* A 55. I. Its isolation and structure. *Biken J.* 12:231-244.
30. Temmyo, R. 1966. Studies on the prevention of outbreaks of food poisoning caused by *Vibrio parahaemolyticus*. *Bull. Tokyo Med. Dent. Univ.* 13:489-510.
31. Thatcher, F. S., and D. S. Clarke. 1968. Microorganisms in foods: Their significance and methods of enumeration, p. 14, 15. University of Toronto Press, Canada.

32. Torii, M., T. An, K. Igarashi, T. Sakai, and K. Kuroda. 1969. Immunological studies on O-antigen of *Vibrio parahaemolyticus*. Preparation, specificity, and chemical nature of the antigens. *Biken J.* 12:24-39.
33. Torii, M., and K. Igarashi. 1969. Immunochemical studies on O-antigens of *Vibrio parahaemolyticus*. Effect of alkali and acid on specificity. *Biken J.* 12:149-159.
34. Tubiash, H. S., R. R. Colwell, and R. Sakazaki. 1970. Marine vibrios associated with bacillary necrosis, a disease of larval and juvenile bivalve mollusks. *J. Bacteriol.* 103:272-273.
35. Twedt, R. M., B. E. Ivins, R. M. E. Novelli, R. G. Thompson, and H. E. Hall. 1970. Selective and differential medium for *V. parahaemolyticus*. *Bacteriol. Proc.* p. 6.
36. Twedt, R. M., P. L. Spaulding, and H. E. Hall. 1969. Morphological, cultural, biochemical and serological comparison of Japanese strains of *Vibrio parahemolyticus* with related cultures isolated in the United States. *J. Bacteriol.* 98:511-518.
37. U. S. Department Health, Education, and Welfare. 1969. Foodborne outbreaks. Annual Summary. Center for Disease Control. Atlanta, Ga.
38. U. S. Dept. Health, Education, and Welfare. 1969. Bacteriological analytical manual. Food and Drug Administration, Washington, D. C.
39. Vanderzant, C., and R. Nickelson. 1971. Isolation and enumeration procedure for *Vibrio parahaemolyticus*. *Bacteriol. Proc.* p. 20.
40. Vanderzant, C., R. Nickelson, and J. C. Parker. 1970. Isolation of *Vibrio parahaemolyticus* from Gulf Coast shrimp. *J. Milk Food Technol.* 33:11-162.
41. Ward, B. Q. 1968. Isolations of organisms related to *Vibrio parahaemolyticus* from American estuarine sediments. *Appl. Microbiol.* 16:543-546.
42. Zen-Yoji, H., S. Sakai, T. Terama, Y. Kudo, and H. Hitogoto. 1963. Studies on the enteropathogenic halophilic bacteria. I. On the epidemiology of the food poisoning due to halophilic bacteria in Tokyo district during 1961 and its bacteriological examination (English summary). *Jap. J. Infec. Dis.* 39:195-204.
43. Zen-Yoji, H., S. Sakai, T. Terayama, Y. Kudo, T. Ito, M. Benoki, and M. Nagasaki. 1965. Epidemiology, enteropathogenicity, and classification of *Vibrio parahaemolyticus*. *J. Infec. Dis.* 115:436-444.

**AMENDMENT TO
3-A SANITARY STANDARDS FOR MULTIPLE-
USE PLASTIC MATERIALS
USED AS PRODUCT CONTACT SURFACES
FOR DAIRY EQUIPMENT**

Serial #2005

*Formulated by
International Association of Milk, Food and Environmental Sanitarians
United States Public Health Service
The Dairy Industry Committee*

The "3-A Sanitary Standards for Multiple-Use Plastic Materials Used as Product Contact Surfaces for Dairy Equipment, Serial #2000", are hereby further amended as indicated in the following:

Section I. Standards for Acceptability, Sub-paragraph (2):
Add the following material to the list of Generic Classes of Plastics:

Polysulfone Resin	.05	0.1	0.1	
-------------------	-----	-----	-----	--

This Amendment is effective Oct. 23, 1971.

PERFORMANCE IN MICROSCOPIC COUNTING OF SOMATIC CELLS IN MILK

I. EFFECTS OF PROCEDURAL VARIATIONS ON ACCURACY AND PRECISION¹

W. D. SCHULTZE², J. W. SMITH², D. E. JASPER³,
O. KLAstrup⁴, F. H. S. NEWBOULD⁵, D. S. POSTLE⁶,
AND W. W. ULLMANN⁷

(Received for publication March 8, 1971)

ABSTRACT

The effects of three changes in the optical equipment used for Direct Microscopic Somatic Cell Count in milk (National Mastitis Council method) on performance of the method have been measured and subjected to statistical analysis of probable significance. Replicate slides of six milk samples were counted by eight technicians among the authors' laboratories. Three levels of cell concentration were represented among the samples. No consistent differences in estimated cell concentration were demonstrated in the comparisons of narrow versus wide reticle band, wide field versus Huyghenian eyepieces, or low (high dry) versus high (oil immersion) magnification. In a few instances within each comparison, individual counters produced markedly different results when the optical equipment was modified. These individual biases were large and consistent with regard to change in objective magnification. No evidence was found to preclude use of wide field eyepieces in performing the DMSCC.

In developing the Direct Microscopic Somatic Cell Count in milk (DMSCC) (1), the need to specify appropriate optical equipment forced us to make certain restrictive decisions. If, for example, reticles of the same band width were to be used with both Huyghenian and uncorrected wide field eyepieces, counts made with the latter equipment would be derived from a considerably smaller area of milk film and thus in theory would be expected to exhibit a greater variance. We decided to specify only Huyghenian eyepieces rather than introduce the confusion

of a multiplicity of reticles (2) in describing a procedure with defined precision.

Since cells are difficult to recognize, there was no question that the high magnification achieved with oil immersion objectives should be required for general use in the DMSCC. Several participating researchers subsequently had occasion to make counts in parallel at lower magnifications and found a marked discrepancy in estimated cell concentration which seemed to be a function of objective magnification. Similarly, counts made using both wide and narrow bands of the two-band reticle seemed for some operators to show a consistent discrepancy. Optical physics offered no theoretical explanation. The latter difference was of immediate concern, for it would constitute a bias in our recommended procedure to minimize differences in expected coefficient of variation by varying the number and width of strips to be counted (3). Consequently, a collaborative study was designed to test the effects of these three procedural variations through counting replicate sets of milk films.

MATERIALS AND METHODS

Milks containing approximately 0.5, 0.75, and 1.0 million somatic cells/ml were prepared by admixing bucket milks from individual cows. Each sample was subdivided into two portions. These were labeled I and IV, II and VI, and III and V to correspond in duplicate with the three desired levels of cell concentration. Sets of DMSCC slides containing two milk films of each of the six samples were prepared by a single technician. The order of film preparation among sets was randomized. Film preparation and staining were performed as specified for the DMSCC (1).

One set of slides was mailed to each of the participating research laboratories with instructions for counting according to eight procedural variations, as shown in Table 1. The two-band reticle originally designed by the Subcommittee on Screening Tests, National Mastitis Council, and described by Schultz (2) was used throughout. Counts were recorded separately for each of the four strips examined for each cell count as specified in the DMSCC.

Because there was good reason not to assume normality of distribution among the cell count data in this study we used nonparametric methods of statistical analysis, in which distributions rather than means are compared. To compare counts produced on a given milk sample by any two procedures we

¹A contribution from the Subcommittee on Screening Tests, National Mastitis Council, Inc.

²United States Department of Agriculture, ARS, Animal Science Research Division, Beltsville, Maryland 20705.

³Department of Clinical Pathology, School of Veterinary Medicine, University of California, Davis, California 95616.

⁴Statens Veterinære Serumlaboratorium, 4, Odinsvej, DK 4100 Ringsted, Denmark.

⁵Department of Veterinary Microbiology and Immunology, Ontario Veterinary College, University of Guelph, Ontario, Canada.

⁶Department of Large Animal Medicine, Obstetrics and Surgery, New York State Veterinary College, Cornell University, Ithaca, New York 14850

⁷Laboratory Division, State Department of Health, P. O. Box 1689, Hartford, Connecticut 06101.

TABLE 1. VARIANT COUNTING PROCEDURES USED IN COLLABORATIVE STUDY.

Count type code	Objective	Eyepieces	Reticle band
1	Oil-immersion	Huyghenian	Wide
2	Oil-immersion	Huyghenian	Narrow
3	High-dry	Huyghenian	Wide
4	High-dry	Huyghenian	Narrow
5	Oil-immersion	Wide field	Wide
6	Oil-immersion	Wide field	Narrow
7	High-dry	Wide field	Wide
8	High-dry	Wide field	Narrow

computed count differences as percentage, as follows:

$$\text{Difference (count b from count a)} = b/a - 1$$

This corrects for inequality of cell concentration and permits pooling of results across samples.

Wilcoxon's Signed Rank Test for Paired Replicates (4, 5) is a nonparametric method which considers medians rather than means of distributions and makes some use of the magnitudes of difference. N rank numbers are assigned to the differences between pairs and each rank number is given a sign according to the sign of the difference. The positive and negative ranks are summed separately, and the smaller sum in absolute value is designated "T". The expected total of one sign, \bar{T} , is equal to $N[(N+1)/4]$ and the variance of the total is equal to $2N\bar{T}/6$. The totals corresponding to the 0.05% and 0.01% levels of significance are computed as follows:

$$T_{.05} = \bar{T} - 1.96(2N\bar{T}/6)^{1/2} \quad T_{.01} = \bar{T} - 2.576(2N\bar{T}/6)^{1/2}$$

In each comparison, the null hypothesis to be tested is that the difference has a median value of zero.

Three variations were included in the experimental design, each a function of the other two. Thus, analysis of any one procedural variation required the comparison of four pairs of counts, selected from among the count types shown in Table 1. For the comparison of narrow versus wide reticle band we used pairs 2:1, 4:3, 6:5, and 8:7; for wide field versus Huyghenian eyepieces we used 5:1, 6:2, 7:3, and 8:4; for high dry versus oil immersion magnification we used 3:1, 4:2, 7:5, and 8:6.

RESULTS AND DISCUSSION

All estimated cell concentrations (factored counts) obtained in the study are shown in Table 2. Counts were not made with wide field eyepieces by Counter 2 of Laboratory C nor by Counter 1 of Laboratory E. Because preliminary work led us to suspect that individual counters might not respond similarly to changes in the DMSCC procedure and equipment all such comparisons were made in parallel within counter.

As an example of the computations involved in the Signed Rank Test, all desired differences which were determined for Counter 1 of Laboratory A are shown in Table 3. Table 4 illustrates the assignment of signed rank numbers to the computed signed differences, using the comparison of narrow versus wide reticle band for counts made with oil-immersion objectives and Huyghenian eyepieces. Only the cumulative

ranking used to quantitate the comparison across all counters is shown. Ranking was also done within each counter's data. The computed values of "T" obtained for all three procedural comparisons are tabulated in Table 5.

The overall analysis indicated high probability that counts made using a narrow reticle band were higher than counts made on the same milk film using a wide reticle band (984 - and 1010 -, as shown in Table 5a). It is instructive, however, to examine individual counter performance. Only three of eight counters produced deviations from count equivalence significant at even the 5% level, and in several instances the direction of trend was reversed. These observations suggest that we are dealing with a problem of psychological bias rather than of physical optics. The magnitude of mean differences in factored cell count was not great, averaging 5.9%.

No evidence was found that counts made using wide field eyepieces might be expected to differ consistently either above or below counts on the same material using Huyghenian eyepieces (Table 5b). Some counter individuality was apparent in the results, however. In both Laboratories A and B, two technicians trained by the same researcher tended toward opposite biases. Subject to investigation of the respective count variances, which will be dealt with in a companion publication, we see no reason to discourage the use of wide field eyepieces for the DMSCC.

The overall analysis of pooled cell counts made with high dry and oil-immersion objectives appeared to establish their nonequivalence (1333 + and 671 -, as shown in Table 5c). Counts made with the high-dry objective might be expected to be lower with greater than 99% probability. But again it was necessary to consider the counters as individuals. The overall bias was contributed largely by two technicians, C-1 and E-1. Since for $N = 12$, the expected total for one sign (T) is 39 and the critical point for $P = 0.05$ is 14, it is apparent that among the other technicians only B-1 shared this strong counting bias.

Originally, only one technician from Laboratory C took part in the investigation. At a meeting of the Subcommittee on Screening Tests held at Laboratory C a preliminary analysis of these data was offered for comment. It was suggested that the uniqueness of count relationships produced by C-1 might relate to the microscope used, an unusually fine instrument of European manufacture. To test this, two of the authors recounted sample I by procedures 1 and 3 at this same microscope. The factored cell counts in thousands were, respectively, 1,146 versus 1,310 and 1,156 versus 1,263. Thus, these two checks produced

TABLE 2. FACTORED CELL COUNTS ($\times 10^{-5}$ PER ML) OBTAINED BY ALL PARTICIPATING COUNTERS.

Milk sample	Count type	Lab A		Lab B		Lab C		Lab D	Lab E	Mean
		Ctr 1	Ctr 2	Ctr 1	Ctr 2	Ctr 1	Ctr 2	Ctr 1	Ctr 1	
I	1	1,128	1,022	1,312	1,247	1,398	1,304	1,308	1,511	1,279
	2	1,243	1,089	1,463	1,257	1,470	1,262	1,199	1,390	1,297
	3	1,174	1,125	1,270	1,423	1,137	1,162	1,263	1,025	1,197
	4	1,160	1,162	1,248	1,266	1,291	1,038	1,196	1,326	1,211
	5	1,203	1,123	1,264	1,364	1,430	—	1,335	—	1,287
	6	1,156	1,062	1,568	1,245	1,693	—	1,372	—	1,349
	7	1,141	1,100	1,161	1,232	1,181	—	1,247	—	1,177
	8	1,299	1,184	1,270	1,375	1,210	—	1,452	—	1,298
II	1	432	480	506	488	590	475	491	592	507
	2	484	470	516	449	706	386	490	635	517
	3	472	455	470	461	412	385	449	296	425
	4	481	440	525	493	470	390	515	493	476
	5	474	440	457	429	483	—	510	—	466
	6	457	423	511	486	656	—	494	—	505
	7	450	469	443	396	413	—	447	—	436
	8	438	462	432	473	490	—	432	—	455
III	1	724	700	802	729	953	731	793	1,042	809
	2	759	847	923	710	1,009	909	752	839	844
	3	733	694	818	838	712	772	759	584	739
	4	741	654	852	863	752	908	840	831	805
	5	696	625	739	784	1,002	—	1,065	—	819
	6	732	517	818	733	1,410	—	1,202	—	902
	7	708	670	743	730	694	—	855	—	733
	8	714	673	691	684	738	—	879	—	730
IV	1	1,089	1,280	1,299	1,218	1,384	1,209	1,209	1,334	1,253
	2	1,236	1,290	1,324	1,245	1,820	1,347	1,374	1,563	1,400
	3	1,189	1,155	1,369	1,425	1,005	1,177	1,292	834	1,181
	4	1,242	1,149	1,369	1,309	1,267	1,101	1,268	1,169	1,234
	5	1,232	1,154	1,270	1,395	1,457	—	1,254	—	1,294
	6	1,304	1,277	1,227	1,287	1,640	—	1,355	—	1,348
	7	1,093	1,179	1,248	1,248	1,056	—	1,304	—	1,188
	8	1,201	1,151	1,360	1,352	1,068	—	1,357	—	1,248
V	1	730	704	858	783	991	760	796	987	826
	2	719	941	880	704	1,056	619	821	922	833
	3	723	722	750	887	652	740	753	860	761
	4	706	730	781	937	788	777	727	352	725
	5	712	631	779	796	938	—	842	—	783
	6	780	726	716	852	1,197	—	869	—	857
	7	719	667	716	835	730	—	819	—	748
	8	692	640	744	849	807	—	757	—	748
VI	1	391	442	482	443	677	427	479	588	491
	2	430	585	540	492	589	421	443	581	510
	3	434	483	469	511	426	437	425	326	439
	4	440	459	454	451	494	438	466	507	464
	5	483	385	469	480	668	—	429	—	486
	6	578	376	520	486	798	—	443	—	534
	7	446	424	465	516	432	—	478	—	460
	8	490	416	500	500	545	—	439	—	482

somewhat higher counts at high dry magnification. Following this, a second technician at Laboratory C was assigned to the study and also failed to confirm the strong bias of Counter C-1. We conclude that there is no evidence of a necessary difference in cell counts referable to the level of objective magnification. Other difficulties, in particular the problem of cell recognition, are stronger arguments against departure from oil-immersion objectives for the DMSCC. Idiosyncracies among individual techni-

cians can produce such strong biases in cell counting, however, that we are cautioned against accepting radical innovations as free options in counting procedure. Procedural variations should be minimized, and first evaluated in a collaborative study before their acceptance.

Through this study it became apparent that individual counter biases can be an important source of error in quantitative microscopic procedures. Even though minor changes in the optical equipment or

TABLE 3. COMPUTED SIGNED DIFFERENCES FOR COUNTER I, LABORATORY A

Count pair (b:a)	Milk sample					
	I	II	III	IV	V	VI
2:1	+10.20	+12.04	+4.83	+13.50	- 1.51	+ 9.97
4:3	- 1.19	+ 1.91	+1.09	+ 4.46	- 2.35	+ 1.38
6:5	- 3.91	- 3.59	+5.17	+ 5.84	+ 9.55	+19.67
8:7	+13.85	- 2.67	+0.85	+ 9.88	- 3.76	+ 9.87
5:1	+ 6.65	+ 9.72	-3.87	+13.13	- 2.47	+23.53
6:2	- 7.00	- 5.58	-2.56	+ 5.50	+ 8.48	+34.42
7:3	- 2.81	- 4.66	-3.41	- 8.07	- 0.55	+ 2.76
8:4	+11.98	- 8.94	-3.64	- 3.30	- 1.98	+11.36
3:1	+ 4.08	+ 9.26	+1.24	+ 9.18	- 0.96	+11.00
4:2	- 6.68	- 0.62	-2.37	+ 0.49	- 1.81	+ 2.33
7:5	- 5.15	- 5.06	+1.72	-11.28	+ 0.98	- 7.66
8:6	+12.37	- 4.16	-2.46	- 7.90	-11.28	-15.22

apparent magnification can in theory be compensated for by calculation of a revised microscope factor, they can have a large and unpredictable effect on the counting performance of individual technicians. This observation argues strongly for the precise definition of detail of any quantitative microscopic procedure, and also for the selection of only a single reference method. The DMSCC (National Mastitis

TABLE 5. VALUES OF "T" COMPUTED FOR THE SIGNED RANK TEST FOR PAIRED DIFFERENCES¹

a. Comparison of NARROW vs. WIDE RETICLE BAND

Counter	Used in combination with:			
	Oil immersion		High dry	
	N	"T"	N	"T"
A-1	12	6 - **	12	24 -
A-2	12	23 -	12	23 +
B-1	12	9 - °	11	13 -
B-2	12	35 +	12	35 -
C-1	12	5 - **	12	0 - **
C-2	6	9 +	6	9 -
D-1	12	30 +	12	29 -
E-1	6	8 +	6	5 -
All	84	984 - **	83	1010 - **

b. Comparison of WIDE FIELD vs. HUYGHENIAN EYEPIECES

Counter	Used in combination with:			
	Oil immersion		High dry	
	N	"T"	N	"T"
A-1	12	18 -	12	26 +
A-2	12	5 + **	12	25 +
B-1	12	6 + **	12	13 + °
B-2	12	13 - °	12	15 +
C-1	12	29 -	12	29 -
D-1	11	10 -	12	21 -
All	71	1229 -	72	1029 +

TABLE 4. ASSIGNMENT OF RANK NUMBERS TO COMPUTED SIGNED DIFFERENCES: COMPARISON OF NARROW VERSUS WIDE RETICLE BANDS USED WITH OIL-IMMERSION OBJECTIVES

Counter	Count pair		Milk sample					
			I	II	III	IV	V	VI
A-1	2:1	CSD ¹	+10.20	+12.04	+ 4.83	+13.50	- 1.51	+ 9.97
		Rank ²	+49	+58	+25	+63	-7	+47
A-1	6:5	CSD	- 3.91	- 3.59	+ 5.17	+ 5.84	+ 9.55	+19.67
		Rank	-24	-22	+27.5	+30	+46	+67
A-2	2:1	CSD	+ 6.56	- 2.08	+21.00	+ 0.78	+33.66	+32.35
		Rank	+33	-10	+76	+2	+82	+81
A-2	6:5	CSD	- 5.43	- 3.86	-17.28	+10.66	+15.06	- 2.34
		Rank	-29	-23	-68	+50	+65	-12
B-1	2:1	CSD	+11.51	+ 1.98	+15.09	+ 1.92	+ 2.56	+12.03
		Rank	+55	+9	+66	+8	+13	+57
B-1	6:5	CSD	+24.05	+11.82	+10.69	- 3.39	- 8.09	+10.87
		Rank	+77	+56	+51	-21	-43	+52
B-2	2:1	CSD	+ 0.80	- 7.99	- 2.61	+ 2.22	-10.09	+11.06
		Rank	+3	-40	-14	+11	-48	+53
B-2	6:5	CSD	- 8.72	+13.29	- 6.51	- 7.74	+ 7.04	+ 1.25
		Rank	-45	+62	-32	-39	+36	+5
C-1	2:1	CSD	+ 5.15	+19.66	+ 5.88	+31.50	+ 6.56	-13.00
		Rank	+26	+74	+31	+80	+34	-61
C-1	6:5	CSD	+18.39	+35.82	+40.72	+12.56	+27.61	+19.46
		Rank	+69	+82	+84	+59	+79	+72
C-2	2:1	CSD	- 3.22	-18.74	+24.35	+11.41	-18.55	- 1.41
		Rank	-19	-71	+78	+54	-70	-6
D-1	2:1	CSD	- 8.33	- 0.20	- 5.17	+13.65	+ 3.14	- 7.52
		Rank	-44	-1	-27.5	+64	+16.5	-38
D-1	6:5	CSD	+ 2.77	- 3.14	+12.86	+ 8.05	+ 3.21	+ 3.26
		Rank	+15	-16.5	+60	+42	+18	+20
E-1	2:1	CSD	- 8.01	+ 7.26	-19.48	+17.17	- 6.59	- 1.19
		Rank	-41	+37	-73	+67	-35	-4

¹Computed signed difference

²Rank among counts from all counters

*c. Comparison of HIGH DRY vs.
OIL IMMERSION OBJECTIVES*

Counter	Used in combination with:			
	Huyghenian		Wide field	
	N	"T"	N	"T"
A-1	12	26 -	12	14 + *
A-2	12	21 +	12	19 -
B-1	12	15 +	12	16 +
B-2	12	7 - °	12	31 +
C-1	12	0 + °°	12	0 + °°
C-2	12	30 +	-	-
D-1	12	28 +	12	17 +
E-1	12	0 + °°	-	-
All	96	1333 + °°	72	671 + °°

Significance at P = .05 designated *

Significance at P = .01 designated °°

Council method) is uniquely susceptible to detailed performance analysis, and counter idiosyncracies can

be identified and corrected through retraining.

REFERENCES

1. National Mastitis Council, Subcommittee on Screening Tests. 1968. Direct Microscopic Somatic Cell Count in milk. *J. Milk Food Technol.* 31:350.
2. Schultze, W. D. 1968. Design of eyepiece reticles for use in the Direct Microscopic Somatic Cell Count method. *J. Milk Food Technol.* 31:344.
3. Schultze, W. D., and J. W. Smith. 1969. A procedure to minimize differences in expected coefficient of variation in the Direct Microscopic Somatic Cell Count. *J. Milk Food Technol.* 32:477.
4. Wilcoxon, F. 1945. Individual comparisons by ranking methods. *Biometrics* 1:80.
5. Wilcoxon, F., and Roberta A. Wilcox. 1964. Some rapid approximate statistical procedures. Lederle Laboratories Division, American Cyanamid Company, Pearl River, New York.

AMENDMENT TO THE 3-A ACCEPTED PRACTICES FOR THE SANITARY CONSTRUCTION, INSTALLATION, TESTING AND OPERATION OF HIGH-TEMPERATURE SHORT-TIME PASTEURIZERS, REVISED

Serial #60304

Formulated by

*International Association of Milk, Food and Environmental Sanitarians
United States Public Health Service
The Dairy Industry Committee*

The "3-A Accepted Practices for the Sanitary Construction, Installation, Testing and Operation of High-Temperature Short-Time Pasteurizers, Revised, (Effective January 22, 1967)", as amended, Serial #60303, are hereby further amended by substituting the following for definition B.1:

B.1

HTST Pasteurization: Heating every particle of (1) milk or milk product to a temperature of at least 161° F, and holding it continuously at or above this temperature for at least 15 seconds in a holding tube, provided that milk products which have a higher milkfat content than milk and/or contain added sweeteners, and concentrated milk products

to be repasteurized before drying, shall be heated to at least 166° F, and held continuously at or above this temperature for at least 15 seconds in a holding tube, or (2) frozen dessert mix to at least 175° F, and holding at or above this temperature continuously for at least 25 seconds in a holding tube.

This Amendment is effective October 23, 1971.

HOLDERS OF 3-A SYMBOL COUNCIL AUTHORIZATIONS ON AUGUST 20, 1971

"Questions or statements concerning any of the holders of authorizations listed below, or the equipment fabricated, should be addressed to Earl O. Wright, Asst. Sec'y-Treas., Dept. of Food Technology, 116 Dairy Industry Bldg., Iowa State University, Ames, Iowa 50010."

0102 Storage Tanks for Milk and Milk Products As Amended

116	Jacob Brenner Company, Inc.	(10/ 8/59)
	450 Arlington, Fond du Lac, Wisconsin 54935	
28	Cherry-Burrell Corporation	(10/ 3/56)
	575 E. Mill St., Little Falls, N. Y. 13365	
102	Chester-Jensen Company, Inc.	(6/ 6/58)
	5th & Tilgham Streets, Chester, Pennsylvania 19013	
1	Chicago Stainless Equipment Corp.	(5/ 1/56)
	5001 No. Elston Avenue, Chicago, Illinois 60630	
2	CP Division, St. Regis	(5/ 1/56)
	100 C. P. Ave., Lake Mills, Wisconsin 53551	
117	Dairy Craft, Inc.	(10/28/59)
	St. Cloud Industrial Park St. Cloud, Minn. 56301	
76	Damrow Company	(10/31/57)
	196 Western Avenue, Fond de Lac, Wisconsin 54935	
115	DeLaval Company, Ltd.	(9/28/59)
	113 Park Street, So., Peterborough, Ont., Canada	
207	The DeLaval Separator Co.	(7/23/69)
	Duchess Turnpike, Poughkeepsie, N. Y. 12602	
109	Girton Manufacturing Company	(9/30/58)
	Millville, Pennsylvania 17846	
21	The J. A. Gosselin Co., Ltd.	(9/20/56)
	P. O. Box 280, Drummondville, Quebec, Canada	
114	C. E. Howard Corporation	(9/21/59)
	9001 Rayo Avenue, South Gate, California 90280	
127	Paul Mueller Company	(6/29/60)
	P. O. Box 828, Springfield, Missouri 65801	
197	Paul Mueller (Canada), Ltd.	(9/ 9/67)
	84 Wellington St., South, St. Marys, Ont., Canada	
213	Sanitary Processing Equipment Corp.	(3/25/70)
	Butternut Drive E. Syracuse, N. Y. 13057	
31	Walker Stainless Equipment Co.	(10/ 4/56)
	Elroy, Wisconsin 53929	

0204 Pumps for Milk and Milk Products Revised, as Amended

214R	Ben H. Anderson Manufacturers	(5/20/70)
	Morrisonville, Wis. 53571	
212R	Babson Bros. Co.	(2/20/70)
	2100 S. York Rd., Oak Brook, Ill. 60621	
29R	Cherry-Burrell Corporation	(10/ 3/56)
	2400 Sixth St., S. W. Cedar Rapids, Iowa 52406	
63R	CP Division, St. Regis	(4/29/57)
	100 C. P. Ave., Lake Mills, Wisconsin 53551	
205R	Dairy Equipment Co.	(5/22/69)
	1919 So. Stoughton Road, Madison, Wis. 53716	
180R	The DeLaval Separator Co.	(5/ 5/66)
	Duchess Turnpike, Poughkeepsie, N. Y. 12602	
65R	G & H Products Corporation	(5/22/57)
	5718 52nd Street, Kenosha, Wisconsin 53140	
145R	ITT Jabco, Incorporated	(11/20/63)
	1485 Dale Way, Costa Mesa, Calif. 92626	
26R	Ladish Co., Tri-Clover Division	(9/29/56)
	2809 60th Street, Kenosha, Wisconsin 53140	

148R	Robbins & Myers, Inc.	(4/22/64)
	Moyno Pump Division 1345 Lagonda Ave., Springfield, Ohio 45501	
163R	Sta-Rite Products, Inc.	(5/ 5/65)
	P. O. Box 622, Delavan, Wisconsin 53115	
72R	L. C. Thomsen & Sons, Inc.	(8/15/57)
	1303 53rd Street, Kenosha, Wisconsin 53140	
219	Tri-Canada Limited	(2/15/71)
	21 Newbridge Road, Toronto 18, Ont., Canada	
175R	National Milkinc Machine Div.	(10/26/65)
	Universal Cooperatives, Inc. First Avenue at College, Albert Lea, Minn. 56007	
52R	Viking Pump Div.—	
	Houdaille Industries, Inc. (12/31/56) 406 State Street, Cedar Falls, Iowa 50613	
5R	Waukesha Foundry Company	(7/ 6/56)
	Waukesha, Wisconsin 53186	

0402 Homogenizers and High Pressure Pumps of the Plunger Type, As Amended

87	Cherry-Burrell Corporation	(12/20/57)
	2400 Sixth Street, S. W., Cedar Rapids, Iowa 52404	
37	CP Division, St. Regis	(10/19/56)
	Fort Atkinson, Wis. 53538	
75	The Inc.	(9/26/57)
	44 Garden Street, Everett, Massachusetts 02149	

0506 Stainless Steel Automotive Milk Transportation Tanks for Bulk Delivery and/or Farm Pick-up Service, As Amended

131R	Almont Welding Works, Inc.	(9/ 3/60)
	4091 Van Dyke Road, Almont, Michigan 48003	
98R	Beseler Steel Products, Inc.	(3/24/58)
	417 East 29th, Marshfield, Wisconsin 54449	
70R	Jacob Brenner Company	(8/ 5/57)
	450 Arlington, Fond du Lac, Wisconsin 54935	
40	Butler Manufacturing Co.	(10/20/56)
	600 Sixth Ave., S. E., Minneapolis, Minn. 55114	
118	Dairy Craft, Inc.	(10/28/59)
	St. Cloud Industrial Park St. Cloud, Minn. 56301	
66	Dairy Equipment Company	(5/29/57)
	1818 So. Stoughton Road, Madison, Wisconsin 53716	
123	DeLaval Company, Ltd.	(12/31/59)
	113 Park Street, South Peterborough, Ont., Canada	
45	The Heil Company	(10/26/56)
	3000 W. Montana Street, Milwaukee, Wisconsin 53235	
201	Paul Krohnert Mfg., Ltd.	(4/ 1/68)
	West Hill, Ontario, Canada	
80	Paul Mueller (Canada), Ltd.	(11/24/57)
	84 Wellington Street, So., St. Marys, Ont., Canada	
85	Polar Manufacturing Company	(12/20/57)
	Holdingsford, Minn. 56340	
144	Portersville Equipment Company	(5/16/63)
	Portersville, Pennsylvania 16051	
71	Progress Industries, Inc.	(8/ 8/57)
	400 E. Progress Street, Arthur, Illinois 61911	
47	Trailmobile, Div. of Pullman, Inc.	(11/ 2/56)
	701 East 16th Ave., North Kansas City, Mo. 64116	
189	A. & L. Tougas, Ltée	(10/ 3/66)
	1 Tougas St., Iberville, Quebec, Canada	

25 Walker Stainless Equipment Co. (9/28/56)
New Lisbon, Wisconsin 53950

**0808 Fittings Used on Milk and Milk Products
Equipment, and Used on Sanitary Lines Conducting
Milk and Milk Products, Revised**

79R Alloy Products Corporation (11/23/57)
1045 Perkins Avenue, Waukesha, Wisconsin 53186

138R A.P.V. (Canada) Equipment, Ltd. (12/17/62)
103 Rivalda Rd., Weston, Ont., Canada

82R Cherry-Burrell Corporation (12/11/57)
2400 Sixth Street, S. W. Cedar Rapids, Iowa 52406

124R DeLaval Company, Ltd. (2/18/60)
113 Park Street, South, Peterborough, Ont., Canada

184R The DeLaval Separator Co. (8/ 9/66)
Duchess Turnpike, Poughkeepsie, N. Y. 12602

67R G & H Products Corporation (6/10/57)
5718 52nd Street, Kenosha, Wisconsin 53140

199R Grayco, Inc. (12/ 8/67)
60 Eleventh Ave., N.E., Minneapolis, Minn. 55413

203R Grinnell Company (11/27/68)
260 W. Exchange St., Providence, R. I. 02901

204R Hills McCanna Company (2/10/69)
400 Maple Ave., Carpentersville, Ill. 60110

34R Ladish Co., Tri-Clover Division (10/15/56)
2809 60th St., Kenosha, Wisconsin 53140

200R Paul Mueller Co. (3/ 5/68)
P. O. Box 828, Springfield, Mo. 65601

149R Q Controls (5/18/64)
Occidental, California 95465

89R Sta-Rite Industries, Inc. (12/23/68)
P. O. Box 622, Delavan, Wis. 53115

73R L. C. Thomsen & Sons, Inc. (8/31/57)
1303 43rd Street, Kenosha, Wisconsin 53140

191R Tri-Canada Fittings & Equipment, Ltd. (11/23/66)
21 Newbridge Road, Toronto 18, Ontario, Canada

151R Tubular Components, Inc. (11/18/64)
Butternut Drive, East Syracuse, New York 13057

215R Universal Milking Machine Div., (7/31/70)
National Cooperatives, Inc.,
First Avenue at College, Albert Lea, Minn. 56007

86R Waukesha Specialty Company (12/20/57)
Walworth, Wisconsin 53184

218 Highland Equipment Corporation (2/12/71)
175 Stockholm St. Brooklyn, N.Y. 11237

**0902 Thermometer Fittings and Connections Used
on Milk and Milk Products Equipment and
Supplement 1, As Amended**

32 Taylor Instrument Companies (10/ 4/56)
95 Ames Street, Rochester, New York 14611

206 The Foxboro Company (8/11/69)
Neponset Ave., Foxboro, Mass. 02035

**1002 Milk and Milk Products Filters Using Disposable
Filter Media, As Amended**

35 Ladish Co., Tri-Clover Division (10/15/56)
2809 60th Street, Kenosha, Wisconsin 53140

**1102 Plate-Type Heat Exchangers for Milk and Milk
Products, As Amended**

20 A.P.V. Company, Inc. (9/ 4/56)
137 Arthur Street, Buffalo, New York 14207

30 Cherry-Burrell Corporation (10/ 1/56)
2400 Sixth Street, S.W., Cedar Rapids, Iowa 52404

14 Chester-Jensen Co., Inc. (8/15/56)
5th & Tilgham Streets, Chester, Pennsylvania 19013

38 CP Division, St. Regis (10/19/56)
Fort Atkinson, Wisconsin 53538

120 DeLaval Company, Ltd. (12/ 3/59)
113 Park Street, South, Peterborough, Ont., Canada

17 The DeLaval Separator Company (8/30/56)
Duchess Turnpike, Poughkeepsie, N. Y. 12602

15 Kusel Dairy Equipment Company (8/15/56)
100 W. Milwaukee Street, Watertown, Wisconsin 53094

**1202 Internal Return Tubular Heat Exchangers,
for Milk and Milk Products, As Amended**

103 Chester-Jensen Company, Inc. (6/ 6/58)
5th & Tilgham Street, Chester, Pennsylvania 19013

96 C. E. Rogers Company (3/31/64)
8731 Witt Street, Detroit, Michigan 48209

152 The DeLaval Separator Co. (11/18/69)
350 Duchess Turnpike, Poughkeepsie, N. Y. 12602

217 Girton Manufacturing Co. (1/23/71)
Millville, Pa. 17846

**1303 Farm Milk Cooling and Holding Tanks—
Revised, As Amended**

11R CP Division, St. Regis (7/25/56)
Lake Mills, Wisconsin 53551

4R Dairy Equipment Company (6/15/56)
1919 S. Stoughton Road, Madison, Wisconsin 53716

92R DeLaval Company, Ltd. (12/27/57)
113 Park Street, South Peterborough, Ontario, Canada

49R The DeLaval Separator Company (12/ 5/56)
Duchess Turnpike, Poughkeepsie, N. Y. 12602

10R Girton Manufacturing Company (7/25/56)
Millville, Pennsylvania 17846

95R Globe Fabricators, Inc. (3/14/58)
7744 Madison Street, Paramont, California 90723

179R Heavy Duty Products (Preston), Ltd. (3/ 8/66)
1261 Industrial Road, Preston, Ont., Canada

12R Paul Mueller Company (7/31/56)
P. O. Box 828, Springfield, Missouri 65801

58R Schweitzer's Metal Fabricators, Inc. (2/25/57)
806 No. Todd Avenue, Azusa, California 91702

134R Universal Milking Machine Division (5/19/61)
National Co-operatives, Inc.
First Avenue at College, Albert Lea, Minn. 56007

216R Valco Manufacturing Company (10/22/70)
3470 Randolph St., Huntington Pk., Calif. 90256

42R VanVetter, Inc. (10/22/56)
2130 Harbor Avenue S.W., Seattle, Washington 98126

18R Whirlpool Corporation, St. Paul Division (9/20/56)
850 Arcade Street, St. Paul, Minnesota 55106

55R John Wood Company (1/23/57)
Superior Metalware Division
509 Front Avenue, St. Paul, Minnesota 55117

170R The W. C. Wood Co., Ltd. (8/ 9/65)
5 Arthur Street, South, Guelph, Ont., Canada

16R Zero Manufacturing Company (8/27/56)
Washington, Missouri 63090

**1400 Inlet and Outlet Leak Protector Plug Valves
for Batch Pasteurizers, As Amended**

- | | | | |
|------|----------------------------------|------------|---|
| 122R | Cherry-Burrell Corporation | (12/11/59) | 2400 Sixth St., S.W. Cedar Rapids, Iowa 52406 |
| 69 | G & H Products Corporation | (6/10/57) | 5718 52nd Street, Kenosha, Wisconsin 53140 |
| 27 | Ladish Co. - Tri-Clover Division | (9/29/56) | 2809 60th Street, Kenosha, Wisconsin 53140 |
| 78 | L. C. Thomsen & Sons, Inc. | (11/20/57) | 1303 43rd Street, Kenosha, Wisconsin 53140 |

**1603 Evaporators and Vacuum Pans for Milk and
Milk Products, Revised**

- | | | | |
|------|--|------------|--|
| 132R | A.P.V. Company, Inc. | (10/26/60) | 137 Arthur Street, Buffalo, New York 14207 |
| 11R | Blaw-Knox Food & Chemical Equip., Inc. | (2/12/59) | P. O. Box 1401
Buffalo, N. Y. 14210 |
| 110R | Arthur Harris & Company | (11/10/58) | 210-218 North Aberdeen Street, Chicago, Illinois 60607 |
| 164R | Mora Industries, Inc. | (4/25/65) | 999 E. Maple Street, Mora, Minnesota 55051 |
| 107R | C. E. Rogers Company | (8/1/58) | 8731 Witt Street, Detroit, Michigan 48209 |
| 186R | Marriott Walker Corporation | (9/6/66) | 925 East Maple Road, Birmingham, Mich. 48010 |

**1702 Fillers and Sealers of Single Service Containers,
For Milk and Milk Products, As Amended**

- | | | | |
|-----|--|------------|--|
| 221 | Bertopack Limited | (5/25/71) | 75 Ardelt Place, Kitchener, Ontario, Canada |
| 192 | Cherry-Burrell Corporation | (1/3/67) | 2400 Sixth St., S. W., Cedar Rapids, Iowa 52404 |
| 139 | Exact Weight Scale Company | (4/15/68) | 944 W. 5th Ave., Columbus, O. 43212 |
| 137 | Ex-Cell-O Corporation | (10/17/62) | P. O. Box 386, Detroit, Michigan 48232 |
| 220 | Haskon, Inc., Package Equipment Division | (4/27/71) | 2285 University Ave., St. Paul, Minnesota 55114 |
| 142 | Polygal Company | (4/15/63) | Div. of Inland Container Corp.
P. O. Box 68074, Indianapolis, Indiana 46268 |
| 211 | Twinpak, Inc. | (2/4/70) | 1133 Avenue of the Americas, New York, N. Y. 10036 |

**1901 Batch and Continuous Freezers, For Ice Cream,
Ices and Similarly Frozen Dairy Foods, As Amended**

- | | | | |
|-----|----------------------------|------------|--|
| 141 | CP Division, St. Regis | (4/15/63) | 100 C. P. Avenue, Lake Mills, Wisconsin 53551 |
| 146 | Cherry-Burrell Corporation | (12/10/63) | 2400 Sixth Street, S. W., Cedar Rapids, Iowa 52404 |

**2201 Silo-Type Storage Tanks for Milk and
Milk Products**

- | | | | |
|-----|----------------------------|-----------|---|
| 168 | Cherry-Burrell Corporation | (6/16/65) | 575 E. Mill St., Little Falls N. Y. 13365 |
| 154 | CP Division, St. Regis | (2/10/65) | 100 C. P. Ave., Lake Mills, Wisconsin 53551 |
| 160 | Dairy Craft, Inc. | (4/5/65) | St. Cloud Industrial Park
St. Cloud, Minn. 56301 |

- | | | | |
|-----|--------------------------------|-----------|---|
| 181 | Damrow Company | (5/18/66) | 196 Western Ave., Fond du Lac, Wisconsin 54935 |
| 156 | C. E. Howard Corporation | (3/9/65) | 9001 Rayo Avenue, South Gate, California 90280 |
| 155 | Paul Mueller Co. | (2/10/65) | P. O. Box 820, Springfield, Missouri 65801 |
| 195 | Paul Mueller (Canada), Ltd. | (7/6/67) | 84 Wellington St., So., St. Marys, Ont., Canada |
| 165 | Walker Stainless Equipment Co. | (4/26/65) | Elroy, Wisconsin 53929 |

**2300 Equipment for Packaging Frozen Desserts,
Cottage Cheese and Milk Products Similar to Cottage
Cheese in Single Service Containers**

- | | | | |
|-----|--------------------------------|-----------|--|
| 174 | Anderson Bros. Mfg. Co. | (9/28/65) | 1303 Samuelson Road, Rockford, Illinois 61109 |
| 209 | Doughboy Industries, Inc. | (7/23/69) | Machine Division
869 S. Main Ave., New Richmond, Wis. 54017 |
| 193 | Triangle Package Machinery Co. | (1/31/67) | 6655 West Diversey Ave., Chicago, Illinois 60635 |

2400 Non-Coil Type Batch Pasteurizers

- | | | | |
|-----|----------------------------|-----------|---|
| 161 | Cherry-Burrell Corporation | (4/5/65) | 575 E. Mill St., Little Falls, N. Y. 13365 |
| 158 | CP Division, St. Regis | (3/24/65) | 100 C. P. Avenue, Lake Mills, Wisconsin 53551 |
| 187 | Dairy Craft, Inc. | (9/26/66) | St. Cloud Industrial Park
St. Cloud, Minn. 56301 |
| 208 | The DeLaval Separator Co., | (7/23/69) | Duchess Turnpike, Poughkeepsie, N. Y. 12602 |
| 177 | Girton Manufacturing Co. | (2/18/66) | Millville, Pennsylvania 17846 |

**2500 Non-Coil Type Batch Processors for Milk and
Milk Products**

- | | | | |
|-----|--------------------------------|-----------|---|
| 162 | Cherry-Burrell Corporation | (4/5/65) | 575 E. Mill St., Little Falls, N. Y. 13365 |
| 159 | CP Division, St. Regis | (3/24/65) | 100 C. P. Avenue, Lake Mills, Wisconsin 53551 |
| 188 | Dairy Craft, Inc. | (9/26/66) | St. Cloud Industrial Park
St. Cloud, Minn. 56301 |
| 196 | Paul Mueller (Canada), Ltd. | (7/6/67) | 84 Wellington St., So., St. Marys, Ont., Canada |
| 202 | Walker Stainless Equipment Co. | (9/24/68) | New Lisbon, Wis. 53950 |

2600 Sifters for Dry Milk and Dry Milk Products

- | | | | |
|-----|-----------------------------|-----------|--|
| 171 | Entoleter, Inc. | (9/1/65) | Subsidiary of American Mfg. Co.
1187 Dixwell Avenue, Hamden, Connecticut 06514 |
| 173 | B. F. Gump Division | (9/20/65) | Blaw-Knox Food & Chem. Equip. Inc.,
750 E. Ferry St., P. O. Box 1041
Buffalo, New York 14240 |
| 185 | The Orville-Simpson Co. | (8/10/66) | 1230 Knowlton St., Cincinnati, Ohio 45223 |
| 176 | Sprout, Waldron & Co., Inc. | (1/4/66) | Munsey, Pennsylvania 17756 |
| 172 | SWECO, Inc. | (9/1/65) | 6111 E. Bandini Blvd., Los Angeles, California 90022 |

THE NATIONAL CONFERENCE ON INTERSTATE MILK SHIPMENTS: TWENTY YEARS IN RETROSPECT

J. C. McCAFFREY

*National Conference on Interstate Milk Shipments
1800 West Fillmore Street
Chicago, Illinois 60612*

(Received for publication March 10, 1971)

ABSTRACT

The National Conference on Interstate Milk Shipments has developed, during the past 20 years, from a shaky beginning, into a solid organization that has benefited all phases of the dairy industry. The gradual increase in and diversification of attendance, by disciplines as well as by geographical entities, has proven that a voluntary organization can work. The gradual changes in the format of the program have resulted from the changing demands of the industry.

Since discussion of the "generation gap" seems to occupy the minds of both the "older" and "younger" generations today, this might be an appropriate time to summarize the progress that has been made by the National Conference on Interstate Milk Shipments. The writer has been familiar with many "starts and stops" that have taken place during the past two decades. Many of the individuals who were active participants during the early years of the Conference have either retired or departed this earth. Many of the new participants in Conference deliberations were still in high school and/or college during the formative years of the organization. It is hoped, therefore, that this article will help to bridge the so-called "generation gap".

EARLY HISTORY

The sanitary quality of milk shipped both interstate and intra-state has been a matter of concern to receiving areas for many years. Lack of uniform standards of inspection, differences in interpretation of regulations, and simple economics of trade barriers were prime causes for the great concern over the low quality of some milk supplies. The situation was brought to a head by the events of World War II. The outbreak of hostilities brought about extensive shifts in population and a tremendous increase in military personnel. This resulted in procurement officers scouring the country for satisfactory milk supplies. Both military and health officers soon found that there was an insufficient supply of clean, safe milk to meet the demands.

In 1946 the Conference of State and Territorial Health Officers requested the United States Public Health Service to develop a workable plan for certification of milk supplies that might be shipped interstate. On December 31, 1946 (1), the Surgeon General sent a letter to all state milk control authorities outlining the proposed plan. The response

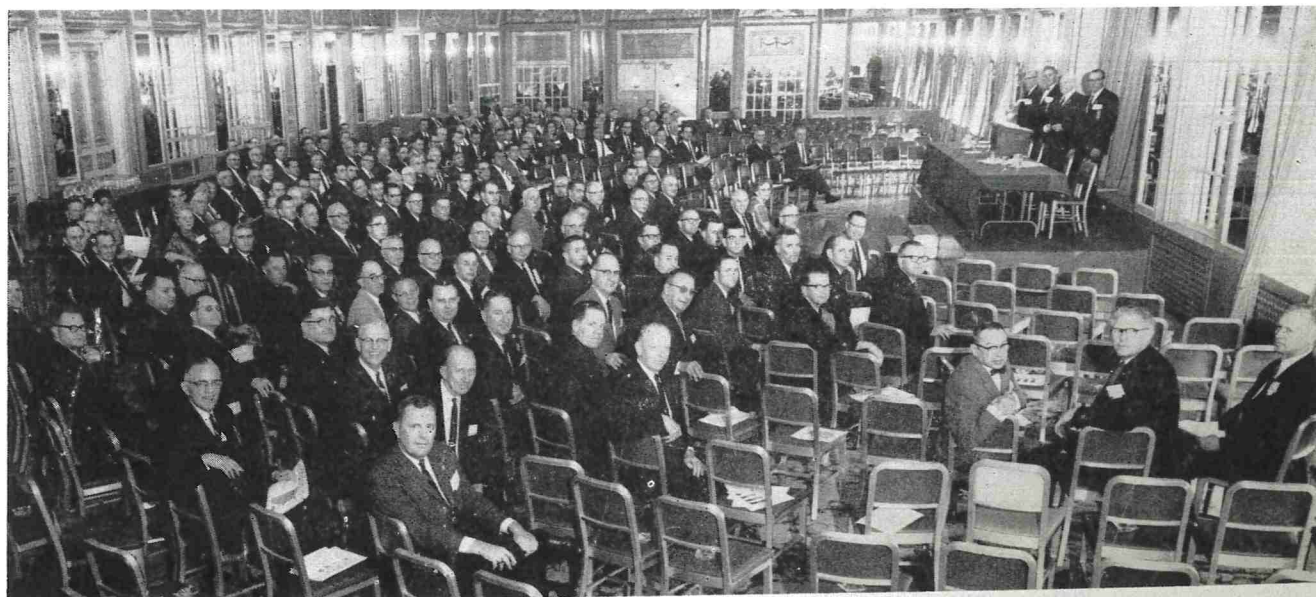


Figure 1. Opening session of the 1963 Conference which met in Memphis.



Figure 2. Park Livingston, Chairman, addressed the opening session of the 1965 Conference in Louisville.

from state authorities was very spotty. This apparent lack of interest, plus a lack of funds and personnel in the Public Health Service, prevented the plan from being implemented at that time. In 1949, therefore, the Association of State and Territorial Health Officers again requested the Public Health Service to assist the states in implementing the program. At this time, similar requests were made by state departments of agriculture, local health officials, and representatives of the milk industry.

In December, 1949, representatives of nine midwestern states met at the Indiana State Board of Health, Indianapolis, Indiana, to study the problem and to determine whether a workable plan could be developed whereby the interstate shipment of milk might be facilitated. In February, 1950, a second meeting was held in Chicago in which representatives of 11 midwestern states participated. At this time, a committee composed of representatives from Wisconsin, Illinois, Indiana, Missouri and Ohio, was appointed to arrange the details for a National Conference.

After considerable deliberation, this committee requested the Surgeon General to call a National Conference to be held at St. Louis, Missouri, June 1-3, 1950. The Surgeon General wrote an invitational letter to all states requesting them to have representatives at this meeting. Representatives of 22 states and the District of Columbia, numbering 70 individuals, participated in the first meeting. As a result of group discussions and joint planning, certain basic conclusions were established, to be used in developing and administering state milk control programs that would bring about reciprocity. Several states immediately began to implement the interstate

program, using the report of the 1950 Conference as guidance.

SUBSEQUENT CONFERENCES

By the time the second Conference was held in St. Louis, June 4-6, 1951, 21 states were participating in the program (1). The purpose of the second meeting was to evaluate the fledgling interstate plan and to make constructive improvements where needed. Seven task forces were set up as study groups to handle problems submitted by the delegates. Historically, the names of these task forces were: (a) Certification, (b) Supervision, (c) Laboratory, (d) Education, (e) Promotion of Interstate Programs, (f) Manufactured Milk Products, and (g) Channels and Forms for Reporting.

Chairman J. L. Rowland announced the following rules to govern the operations of the task forces and the general assembly.

1. Task Force Rules
 - a. Task forces will be appointed by the Conference chairman.
 - b. Each task force will select its own chairman.
 - c. Each task force will select a subcommittee of three to prepare the report of the task force.
 - d. The chairman of the task force will present the report to the general assembly.
2. General Assembly Rules
 - a. In general assembly, each state will be entitled to one vote.
 - b. Representatives of municipalities, industry, Public Health Service, and other federal agencies will not be entitled to a vote in the general assembly.

The third National Conference was held in St. Louis, Missouri, June 10-12, 1952. During this Conference Chairman Rowland appointed a parliamentary procedures committee, under the chairmanship of C. K. Luchterhand of Wisconsin. The committee was directed to study the organizational problems and outlined a procedure for selection of responsible officials to direct the activities of the Conference in the future. The committee recommended selection of an Executive Board to be composed of 12 members including the preceding chairman *ex officio* (1). The remaining 11 members were to be selected as follows: from shipping states as a group and from receiving states as a group, each group having one member from a state department of health, one from a state department of agriculture, one from a municipal health agency, and one from industry; one from the U. S. Public Health Service; one from the U. S. Department of Agriculture; one from an educational institution. The Executive Board was to elect a chairman from among its own membership.

In 1955 the constitution was revised and membership of the Executive Board was increased from 12 to 17 (2). States were divided geographically,

TABLE 1. ANNUAL ATTENDANCE BY STATE

State	1950	1951	1953	1955	1957	1959	1961	1963	1965	1967	1969
Alabama	1		1	7	7			1	3	5	2
Arizona								2	3	2	4
Arkansas	1	3	2	5	2	1	3	12	7	6	3
California						1	1	2	6	5	10
Colorado	1	1	1	2		2	3	4	5	5	32
Connecticut	1	1	2	3	3	4	3	3	2	3	3
Delaware		1			1	3		1	1		1
Dist. of Col.	3	10	6	11	4	6	11	14	18	15	14
Florida	2	1	1	1	1			4	3	12	8
Georgia	4	8	1	5	4	2	7	8	11	12	3
Idaho	1	1	1				1		1	1	6
Illinois	10	11	17	19	16	20	22	28	33	29	32
Indiana		1	4	5	4	5	6	7	17	7	8
Iowa	2	2	1	4	6	10	13	9	14	8	9
Kansas	4	2		4	4	2	3	3	6	5	8
Kentucky		1	1	3	4	1	9	14	41	9	10
Louisiana	1	1	1	1	2	2	1		4	5	1
Maine				1		1	1	1	1	1	
Maryland		1		1	1	1	2	2	3	13	4
Massachusetts			1	1		1	1	2	3	4	3
Michigan				2	2		4	6	12	7	10
Minnesota	4	12	6	12	6	6	4	6	9	10	7
Mississippi		1	1	3	3	4	3	6	6	5	4
Missouri	17	18	17	9	11	21	22	17	17	17	17
Montana										1	5
Nebraska	2	2	2	1	1	1	1	1	1	3	4
Nevada											1
New Hampshire										1	
New Jersey	2	1		1		1		2	2		1
New Mexico	1	1	1		1		1		1	1	
New York	2	4	4	6	4	2	5	11	19	22	16
No. Carolina	2		2	2	1	1	1	2	5	8	9
No. Dakota				1	1	3	2	2	4		2
Ohio	3	4	2	5	4	6	8	16	38	24	26
Oklahoma	1	1	1	6	2	1		2	3	2	4
Oregon								1	1	1	1
Pennsylvania	1	4	5	7	6	5	5	9	18	13	10
Puerto Rico											1
Rhode Island				2				1			
So. Carolina				2					2	3	1
So. Dakota			3	1						1	3
Tennessee	1			22	14	4	5	28	13	7	12
Texas	4	4		8	5	5	5	6	7	10	15
Utah			1				1	2		1	1
Vermont			1	1		1	1	1	1	2	2
Virginia			1	2	5	4	6	7	10	11	10
Washington								1	2	2	4
W. Virginia		1	1	1	1	1	1	2	2	4	1
Wisconsin	2	6	11	19	18	14	11	14	15	19	15
Wyoming											3
Total	70	104	101	186	144	142	173	260	371	322	346

TABLE 2. REGISTRANTS BY FIELD

State	1950	1951	1953	1955	1957	1959	1961	1963	1965	1967	1969
Health	30	33	41	76	65	51	63	98	133	93	87
Agriculture	13	22	12	24	17	26	20	27	30	29	44
P.H.S.	13	13	7	9	9	9	19	27	36	33	26
Industry	13	25	26	58	39	44	49	88	130	111	123
U.S.D.A.			1	3	1	1		2	6	3	2
University	1	2	4	5	4	5	7	5	11	12	19
Comm. labs.				2	1	2	4	2	5	3	2
Suppliers		5	2	5	5	3	9	3	15	12	15
News media			3		2			2	2	3	3
Miscellaneous		4	5	4	1	1	2	6	3	23	25
Total	70	104	101	186	144	142	173	260	371	322	346

from north to south, into 3 groups: Group I consisting of the eastern states, Group II the midwestern states, and Group III the western states. Each group was to have one member from a state health department, one from industry, one from a state department of agriculture, and one from a municipal health department. The representatives from the U. S. Public Health Service, U. S. Department of Agriculture, and an educational institution were to be elected "at large," meaning that the individual could be selected from any part of the country. At this time, a representative of laboratory was added to the Board, also to be elected "at large." The Public Health Service representative was assigned to Group I, representatives of an educational institution and laboratory were assigned to Group II, and representatives from the U. S. Department of Agriculture were assigned to Group III. The make-up of the Executive Board has not changed since 1955. The format of task forces remained essentially the same through all Conferences up to 1969, but the titles varied somewhat from year to year depending upon the problems involved.

ORGANIZATIONAL CHANGES

The Executive Board, during an interim meeting held at the O'Hare Ramada Inn, Chicago, on March 8, 1966, authorized the Secretary to publish a Newsletter to keep all registrants advised of future Conference plans. The first issue was published on May 16, 1966. At the same meeting the Board voted to select future meeting places 4 years in advance, as well as to extend the Conference through a fourth day so that individuals participating would have more time to study task force reports.

In 1965 the Executive Board voted to establish a standing Credentials Committee to arbitrate disputes concerning the voting rights of official delegates. The committee was to be composed of three members, one from each of the Regions, with the Secretary-Treasurer as a non-voting ex-officio member. The members of the original committee were: Roger Holkesvik, North Dakota; Roy Perkins, Tennessee; and Earle Borman, Connecticut as Chairman. At the same time the Board voted to make the Task Force reports available to registrants at the Conference on the evening before the final business session, so that interested persons and/or delegates would have the opportunity to study the reports.

The task force alignment for the 1969 (4) Conference was completely reorganized in an attempt to develop better working conditions. Each task force consisted of 5 members who were appointed in advance by the Program Committee, with the approval of the Executive Board. The individuals selected

represented the five different disciplines involved in the Conference: state department of health, state department of agriculture, local health department, industry, and educational institutions. Registered participants were allowed to participate in any task force that they wished. After the conclusion of deliberations, the five members of the task force prepared a short statement regarding the problems and the actions taken. Any individual (or group) who disagreed with the task force statement was authorized to make a 3-min presentation of a minority report at the business session. While some individuals complained about task force reorganization, the procedure operated much more smoothly than at some times in the past. The former practice of "packing" task forces was largely eliminated.

LOCATION

The first four Conferences (1950, 1951, 1952, and 1953) were held at the Statler Hotel, St. Louis, Missouri, with attendance averaging slightly over 100. The Executive Board shifted the 1955 and 1957 meetings to the Hotel Peabody in Memphis, with a tremendous increase in attendance. St. Louis hosted the meeting in 1959 and 1961. The 1963 meeting returned to Memphis and an all-time high registration of 260 was reached. The Board voted to meet in Louisville, Kentucky in 1965 and attendance skyrocketed to 371. The famous Deauville Hotel in Miami Beach, Florida, was the scene of the 1967 Conference and the New Albany Hotel, Denver, Colorado, was the headquarters for the 1969 meeting. The 1971 Conference was held at the Chase-Park Plaza Hotel, St. Louis, Missouri.

ATTENDANCE BY STATES

As was mentioned previously, only 22 states and the District of Columbia were represented at the first Conference in 1950. The attendance figures, both by state and by individuals, for all conferences are listed in Table 1. The Secretary's files are, with the exception of 1952 registration, complete from 1950 through 1969. The author has been unable to obtain a registration list for the 1952 Conference. Only 12 states and the District of Columbia have been represented at every one of the Conferences. Illinois has the largest average representations of the above mentioned group with 21.5. Missouri is second with an average of 17, followed by Ohio with an average of 13.5, and Wisconsin with an average of 13.

TYPE OF REGISTRANTS

The type of registrants by profession is shown in

Year	Chairman	Secretary	Treasurer
1950-3	J. L. Rowland	None	None
1955	Dr. K. G. Weckel	C. K. Luchterhand	H. J. Barnum
1957-9	H. L. Hortman	Harvey Weavers	H. J. Barnum
1961	H. J. Barnum	J. C. McCaffrey	M. P. Baker
1963	Park Livingston	J. C. McCaffrey	M. P. Baker
1965	Park Livingston	J. C. McCaffrey-Secretary-Treasurer	
1967	Dr. Howard K. Johnston	J. C. McCaffrey	
1969	Shelby Johnson	J. C. McCaffrey	

Table 2. It is interesting to note that health, agriculture, industry, and educational institutions have been well represented at every Conference. While registration from health and agriculture has remained relatively constant throughout the years, dairy industry representation has markedly increased. Educational institutions likewise are showing a greater interest in the deliberations of the Conference, as are representatives of the dairy industry supply organizations.

OFFICERS

From 1950 through 1953, the only elected Conference officer was the Chairman, J. L. Rowland, Associate Professor of Preventive Medicine and Public Health, Kansas City College of Osteopathy and

Surgery, Kansas City, Missouri. From 1955 through 1963, the elected officers consisted of (a) Chairman, (b) Secretary, and (c) Treasurer. In 1963 the constitution was changed to combine the officers of Secretary and Treasurer. The officers who have guided the Conference through its first 20 years are listed below:

REFERENCES

1. 1952 Conference report. 1953. J. Milk Food Technol. 16:232-237.
2. 1957 Conference report. 1958. J. Milk Food Technol. 21:164-171.
3. 1967 Conference report. 1967. J. Milk Food Technol. 30:208-212.
4. 1969 Conference report. 1969. J. Milk Food Technol. 32:290-294.

ASSOCIATION AFFAIRS

NOTICE TO MEMBERS IAMFES, INC.

At the Annual Meeting of the Executive Board San Diego, Calif. August 15, 1971—the Executive Board in accordance with the By-Laws, Article I, Section 2 raised the Annual dues to \$12.00 affiliate, and \$14.00 Direct, beginning Jan. 1, 1972. It was determined by the Board that the raise was absolutely necessary for the following reasons:

1. The last dues raise was Jan. 1, 1966 and we all know costs have been consistently rising, making it increasingly difficult each year to meet expenses.
2. An increase in May 1971 of over 20% in postal costs.
3. An increase of over 20% in printing costs as of August 1971.
4. A greatly expanded Journal over the past two years which has provided considerably more for the money.
5. The need to build sufficient margin to hire a competent replacement for "Red" when he retires, with pension plan for the new Executive Secretary.
6. The need to meet our obligation to "Red" for

his pension—we have two more payments of \$6120.00 each.

7. It is estimated that our expenses will increase by \$8500 this fiscal year July 1971-June 30, 1972. Allowing for a net income of \$6000.00 we will need to increase our income by \$11,500. We average about \$3000.00 net income now. We would still be short on our pension obligation. However the Board felt that a \$4.00 raise in dues plus increases in subscription rates, reprints, advertising etc. would increase our income over \$12,000.00. We will be able to meet the \$6120.00 payment this year from the reserve and only one more payment will then be due next year so we will be in a position to handle the situation at the end of this period.

The Executive Board earnestly solicits your support and understanding of the situation and truly hopes that each and every member appreciates the contribution IAMFES makes to them and to the field of milk and food sanitation. To anyone worthy of the name "Professional Sanitarian" the Journal alone is worth the membership dues.

Sincerely,
Orlowe M. Osten, President

NEWS & EVENTS

15TH ANNUAL EDUCATIONAL SEMINAR OF NSPS

The 15th Annual Educational Seminar and Convention of the National Society of Professional Sanitarians will be held at the Sands Motel, Tucson, Arizona, November 3, 4, and 5, 1971.

The theme of the seminar is "Food: From Seed to Man." Instructors from the College of Agriculture, University of Arizona, will be instructing the seminar.

All sanitarians and other persons interested in the seminar are invited to attend. Please make advanced reservations and mail to: Bailey E. Battiste, R.P.S., President, N.S.P.S., 151 West Congress Street, Pima County Health Department, Tucson, Arizona 85701.

NATIONAL RESTAURANT ASSOCIATION IN RECENT MESSAGE URGES MEMBERS TO IMPROVE BY VOLUNTARY SELF-INSPECTION

The image of your foodservice establishment that greets your customer determines, to a considerable extent, his patronage of your establishment.

Very important to you is what the customer sees during his visit . . . hopefully, a bright, clean and attractive place in which to dine.

More important to you is what your customer, for the most part, does not see. The personal hygiene practices of your employees; the manner in which food is handled during storage preparation and serving; and the manner in which equipment and utensils are cleaned and sanitized.

The extent to which your employees practice good personal hygiene, prepare and serve food in a safe manner and maintain equipment and utensils in a clean and sanitary condition, measures the extent to which you protect your customers from contaminated food and possible foodborne illness.

You cannot rely on the visits and counsel of your health department's sanitarian to greatly aid you in presenting a safe eating place for your customers. Although your customers visit your restaurant every day, the sanitarian sees it infrequently . . . once a month; once every 2 or 3 months; once or twice a year or perhaps not at all.

As in many other situations, a businessman has to do it himself . . . in other words, the effective protection of your customer can be achieved by requiring that your managers and supervisors do a continual job of self-inspection of both facilities and

practices . . . by checking department by department and function by function for unsafe procedures and unsanitary conditions which can lead to contamination of food and growth of harmful bacteria.

Your health officer can assist you in setting-up an effective self-inspection program for your establishment. The National Restaurant Association can supply you with reference charts and guidance material to supplement the assistance given you by the health department.

PLANS UNDERWAY FOR NATIONAL DAIRY HOUSING CONFERENCE

Providing practical information to dairymen and others interested in dairy facilities will be the goal of the National Dairy Housing Conference that will be held February 6-8, 1973, at Michigan State University. Noted authorities from all areas of dairy housing will be on hand to discuss new ideas and relate their field experiences. Topics will include housing, feeding, milking, and construction systems; building materials, environmental control, manure and waste management, regulations, economics, sanitation, and herd health.

The meeting will combine the efforts of the American Society of Agricultural Engineers, American Dairy Science Association, Canadian Society of Agricultural Engineering, Farmstead Equipment Association, Milking Machine Manufacturers Council, International Association of Milk, Food and Environmental Sanitarians, Agricultural Research Service, and the Agricultural Extension Service. Conference organizers are looking for program material from anyone interested in dairy cattle housing and facilities. Either 200-word abstracts or completed papers should be sent to NDHC Program Chairman T. J. Brevik, Agricultural Engineering Department, University of Wisconsin, Madison, Wisconsin 53706, by November 30, 1971.

PROFESSOR F. E. NELSON HONORED

F. Eugene Nelson, Professor of Dairy and Food Sciences and Professor of Microbiology and Medical Technology at the University of Arizona, was the recipient of the American Dairy Science Association Award of Honor during the Annual Meeting of ADSA at Michigan State University, June 20-23. He has served as President of the Association, a member of the Board of Directors and member and chairman of numerous committees. He was editor of the

Journal of Dairy Science from 1947 to 1952 and 1953 recipient of the Borden Award of ADSA in recognition of his research on dairy products. Many of his former graduate students hold responsible positions in commercial and academic phases of the dairy and food industries.

LABORATORY EXAMINATION OF DAIRY PRODUCTS - I

This course is designed for technicians who are responsible for the microbiological examination of raw or pasteurized milk and other dairy products. Enrollment is limited to 24 to enable instructors to deal with individual needs of the less experienced technician and to provide consultation with the more experienced worker.

Laboratory procedures described in the current "Standard Methods for the Examination of Dairy Products" and AOAC as they support the 1965 Grade A Pasteurized Milk Ordinance and the National Conference on Interstate Milk Shipments are emphasized.

Considerable time is spent in the laboratory. Each technique is accompanied by a lecture to present the test in detail; manipulations or factors which are

likely to cause variations in the results are pointed out to enable the technician to achieve a greater degree of standardization.

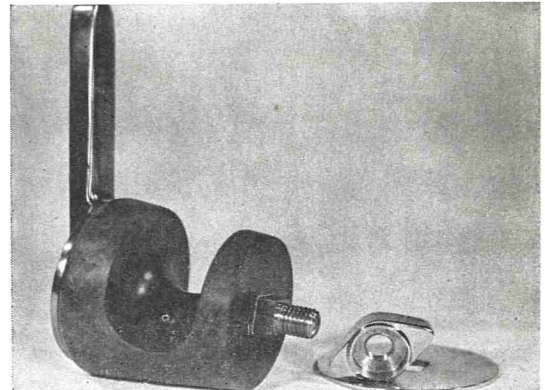
The course is scheduled for November 15-19, 1971 in Cincinnati, Ohio.

Applications should be sent to: Cincinnati Training Facility, Food and Drug Administration, 1090 Tusculum Avenue, Cincinnati, Ohio 45226.

Tanaco Plastic Plug Sanitary Valves

given Honors in Putman FOOD Awards

N
O
G
A
L
L



#316 Stainless Body and parts with the Industry Approved TANACO PLASTIC PLUG. Guaranteed Leakproof!

N
O
S
T
I
C
K



TANACO PRODUCTS

2825 Benedict St., Los Angeles, Cal. 90039



NEW!

LIQUID
SUD'N

Is Now Available

•
Phosphate Free

•
Biodegradable

•
Non Polluting

TODAY it's Free rinsing SUD'N for brush and ultra-sonic washing of all laboratory glassware, missile and other space components . . . used wherever the ultimate in cleaning is essential . . . where anything less would be too expensive.

CLEANS COMPLETELY . . .



RINSES freely without film or residue even in COLD WATER. TRY SUD'N. Contact your Sep-Ko dealer or write

SEP-KO CHEMICALS, INC.

3900 Jackson St., N.E. Minneapolis, Minn. 55421

INDEX TO ADVERTISERS

Babson Bros., Co.	Back Cover
IAMFES, Inc.	I, IV
Pennwalt Corporation	Inside Front Cover
Sep-Ko Chemicals, Inc.	466
Tanaco Products	466
The Haynes Mfg. Co.	Inside Back Cover

CLASSIFIED ADS

FOR SALE

Single Service milk sample tubes. For further information and a catalogue please write, Dairy Technology Inc., P. O. Box 101, Eugene, Oregon 97401.

Application for Membership
**INTERNATIONAL ASSOCIATION OF MILK, FOOD & ENVIRONMENTAL
 SANITARIANS, INC.**
 Box 437, Shelbyville, Indiana 46176

Name ----- Date -----
 Please Print

Address ----- Renewal
 ----- Zip Code ----- New
 Business Affiliation ----- Re-instatement

Direct Member Annual Dues \$14.00 Check Cash
 Membership Through An Affiliate—\$12.00 Plus Affiliate Dues
 Student Membership \$4.00
 (Membership Includes Subscription to Journal of Milk & Food Technology.)

Please Print

Recommended by -----
 Shelbyville, Ind. Subscription Order
 Box 437

JOURNAL OF MILK & FOOD TECHNOLOGY
 (Monthly Publication)

Name ----- Date -----
 Please Print

Address ----- New
 ----- Renewal

Educational Institution & Public Libraries (Annually) \$12.00 Check Cash Individual Non-Member Subscription (Annually) \$14.00
 Government Agencies, Commercial Organizations

I.A.M.F.E.S. & J.M.F.T.
 Box 437, Shelbyville, Ind. Change of Address
FROM

Name ----- Date -----
 Please Print

Address -----
TO

Name -----
 Please Print

Address -----
 I.A.M.F.E.S. & J.M.F.T.
 Box 437, Shelbyville, Ind. Order for 3A Standards

Name ----- Date -----
 Please Print

Address -----

- () Complete Set @ \$5.50 = ----- () Complete set bound (durable cover) @ \$7.00 = -----
 () Revised HTST Std.—without cover = \$1.50 F.O.B. Shelbyville, Ind.
 () Revised HTST Std.—25 or more = \$1.00 each F.O.B. Shelbyville, Ind.
 () E-3-A Sanitary Standards—Set Unbound @ \$1.25 = -----

3-A Accepted Practices For Milking Machines
 1-100 = 25c ea.; 100-1000 = 20c ea.; 1000 or more = 15c ea.
 5 Year Service on Standards as Published = \$6.00 additional

Order for Reprints of Articles

Amt. ----- Title -----

Schedule of letterpress prices for reprints within 30 days after publication, F.O.B. Shelbyville, Indiana

	1 Page	2 Pages	3 & 4 Pages	6 & 8 Pages	12 Pages	Cover
100 or less	\$19.50	\$23.40	\$32.80	\$46.80	\$78.00	\$33.60
Add'l 100's	2.40	3.00	5.00	6.60	10.80	5.50

After 30 Days Offset Reprint prices add \$2.00 per page

THE ONLY Approved
SANITARY METHOD OF APPLYING
A U. S. P. LUBRICANT
TO DAIRY & FOOD
PROCESSING EQUIPMENT

*Haynes
Spray*

U.S.P. LIQUID PETROLATUM SPRAY
U.S.P. UNITED STATES PHARMACEUTICAL STANDARDS

CONTAINS NO ANIMAL OR VEGETABLE FATS. ABSOLUTELY
NEUTRAL. WILL NOT TURN RANCID—CONTAMINATE OR
TAINT WHEN IN CONTACT WITH FOOD PRODUCTS.

SANITARY—PURE

ODORLESS—TASTELESS

NON-TOXIC

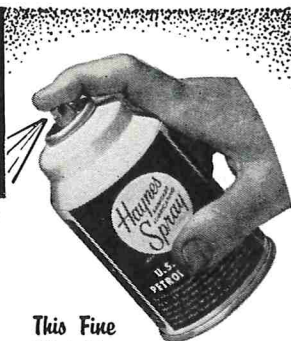
The Modern HAYNES-SPRAY Method of Lubrication
Conforms with the Milk Ordinance and Code
Recommended by the U. S. Public Health Service

The Haynes-Spray eliminates the danger of contamination which is
possible by old fashioned lubricating methods. Spreading lubricants
by the use of the finger method may entirely destroy previous
bactericidal treatment of equipment.

PACKED 6-12 oz. CANS PER CARTON

SHIPPING WEIGHT—7 LBS.

THE HAYNES MANUFACTURING COMPANY
4180 LORAIN AVENUE • CLEVELAND, OHIO 44113



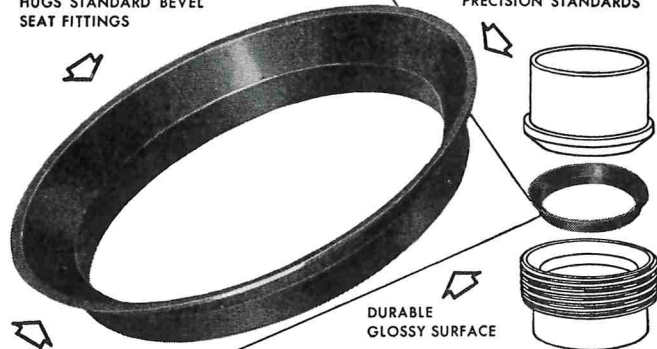
This Fine
Mist-like
HAYNES-SPRAY
should be used to lubricate:

SANITARY VALVES
HOMOGENIZER PISTONS — RINGS
SANITARY SEALS & PARTS
CAPPER SLIDES & PARTS
POSITIVE PUMP PARTS
GLASS & PAPER FILLING
MACHINE PARTS
and for ALL OTHER SANITARY
MACHINE PARTS which are
cleaned daily.

HAYNES SNAP-TITE GASKETS

"FORM-FIT" WIDE FLANGE
HUGS STANDARD BEVEL
SEAT FITTINGS

MOLDED TO
PRECISION STANDARDS



DESIGNED TO
SNAP INTO
FITTINGS

DURABLE
GLOSSY SURFACE

▶ **LOW COST...RE-USABLE**

▶ **LEAK-PREVENTING**

NEOPRENE GASKET for Sanitary Fittings

Check these **SNAP-TITE** Advantages

Tight joints, no leaks, no shrinkage

Time-saving, easy to assemble

Sanitary, unaffected by heat or fats

Self-centering

Non-porous, no seams or crevices

No sticking to fittings

Odorless, polished surfaces, easily cleaned

Eliminate line blocks

Withstand sterilization

Help overcome line vibrations

Long life, use over and over

Available for 1", 1½", 2", 2½" and 3" fittings.

Packed 100 to the box. Order through your dairy supply house.

THE HAYNES MANUFACTURING CO.
4180 Lorain Avenue • Cleveland 13, Ohio

HAYNES
SELF-CENTERING
SNAP-TITE
Gaskets

* MADE FROM
TEFLON

SIZES 1" - 1½"
2" - 2½" - 3"

"The Sophisticated Gasket"
THE IDEAL UNION SEAL FOR
BOTH VACUUM AND
PRESSURE LINES

Gasket Color...
slightly off-white

SNAP-TITE self-centering gaskets of TEFLON are designed for all
standard bevel seat sanitary fittings. They SNAP into place provid-
ing self-alignment and ease of assembly and disassembly.
HAYNES SNAP-TITES of TEFLON are unaffected by cleaning solu-
tions, steam and solvents. They will not embrittle at temperatures
as low as minus 200° F. and are impervious to heat up to 500° F.

FOR A FITTING GASKET THAT WILL OUT-PERFORM ALL OTHERS...

Specify . . . HAYNES SNAP-TITES of TEFLON

• TEFLON ACCEPTED SAFE FOR USE ON FOOD & PROCESSING
EQUIPMENT BY U. S. FOOD AND DRUG ADMINISTRATION

* Gaskets made of DuPont TEFLON® TFE-FLUOROCARBON RESINS

THE HAYNES MANUFACTURING COMPANY
4180 LORAIN AVENUE • CLEVELAND, OHIO 44113

A HEAVY DUTY SANITARY LUBRICANT



Available in both
SPRAY AND TUBE

All Lubri-Film ingredients are
approved additives and can be
safely utilized as a lubricant for
food processing equipment when
used in compliance with existing
food additive regulations.

ESPECIALLY DEVELOPED FOR LUBRICATION OF FOOD
PROCESSING AND PACKAGING EQUIPMENT

For Use in Dairies — Ice Cream Plants — Breweries —
Beverage Plants — Bakeries — Canneries — Packing Plants

SANITARY • NON TOXIC • ODORLESS • TASTELESS

SPRAY — PACKED 6 — 16 OZ. CANS PER CARTON
TUBES — PACKED 12 — 4 OZ. TUBES PER CARTON

THE HAYNES MANUFACTURING CO.
CLEVELAND, OHIO 44113



**In ten years
this pipeline joint will still be tight**

**No leaks — easily cleaned — maintenance free
permanent — welded by Surge**

Welded milk lines are available from your Surge Dealer, wherever you may dairy in the United States or Canada. A concept proven in milk processing plants and on the dairy in localized areas for more than 15 years is now offered by Surge Dealers everywhere.

Expansion and contraction of the milk line along with gasket aging in conventional couplings can cause cracks and leaks. Cracks rep-

resent contamination sources and leaks are detrimental to milk quality and efficient milking. Welded lines can eliminate these problems.

All welded lines installations are subject to the approval of local regulatory authorities.

Thorough, efficient cleaning, which results in a higher quality milk product, is of interest to everyone — dairymen, sanitation specialists, dairy plant fieldmen, and consumers.

↳ SURGE...the accent is on YOU

SURGE

**BABSON BROS. CO., OAK BROOK, ILLINOIS
BABSON BROS. CO., (Canada) LTD., PORT CREDIT, ONTARIO**