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<td>Single</td>
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<tr>
<td>Double-Twin Beds</td>
<td>$14.95</td>
</tr>
<tr>
<td>2 Double Beds—2 persons</td>
<td>$18.95</td>
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</tr>
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ADDRESS __________________________________________

Date of Arrival ___________________________________

Date of Departure _________________________________

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Reserve Double-Twin Beds Rate __________

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

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EFFECTS OF CARBON DIOXIDE AND VACUUM PACKAGING ON COLOR AND BACTERIAL COUNT OF MEAT

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(Received for publication September 26, 1969)

ABSTRACT

Fresh hamburger was packaged under vacuum or in air in films having different oxygen permeability and stored in a display case at 5°C for 30 days. Bacon was packaged with a laminated material, under vacuum, in air or carbon dioxide, and similarly stored. Aerobic and anaerobic bacterial counts were made at various intervals. In addition, bacon was examined for Clostridium perfringens and lactobacilli. Color changes also were evaluated.

Bacterial numbers increased on hamburger with either packaging method. However, growth of aerobes on vacuum-packaged meat was slower than on meat packaged in air. Anaerobes increased in numbers in hamburgers in evacuated packages after 3 days in storage and after 6 days in meat in unevacuated packages.

Packaging bacon in carbon dioxide resulted in reduction of total numbers of aerobes and lactobacilli. Few C. perfringens were recovered. Color retention was improved for as long as 30 days with either vacuum or CO₂ packaging, as compared with air controls.

In general, growth of anaerobes occurred earlier on fresh meat packaged under vacuum than in air; the converse was true for aerobic bacteria. Carbon dioxide inhibited bacterial growth on packaged bacon and provided good color retention during storage.

In 1889, Frankel (as reviewed by Ogilvy and Ayres, 22) showed that microorganisms varied widely in their susceptibility to carbon dioxide; proliferation of some was completely suppressed, but others were less affected. There have been many reports that growth of bacteria, molds, and yeasts was inhibited by carbon dioxide (3, 6, 8, 15, 17, 20, 21, 23). King and Nagel (16) stated that, at 70% carbon dioxide (v/v), the generation time was nearly doubled for Pseudomonas aeruginosa. Ogilvy and Ayres (24) and Hays et al. (12) reported that lactic acid bacteria were not inhibited by carbon dioxide. The number of viable spores of Clostridium botulinum and Clostridium butyricum was reduced by carbon dioxide after 42 days of incubation (12).

Preservation of color of bacon by carbon dioxide was reported by Callow (6). Compared with storage in air, bacon held in an atmosphere of pure carbon dioxide had significantly longer shelf life. Ulrich and Halvorson (30) stated that vacuum or gas packaging inhibited rancidity but did not hinder growth or activity of microorganisms normally present on bacon or introduced during processing. Concentrations of carbon dioxide up to 50% were observed to improve the storage life of frankfurters (23). The increased keeping time of frankfurters was attributed by Ogilvy and Ayres (24) to the control of the total microbial population and to the restriction of the types of microorganisms that most rapidly cause deterioration.

Since Jensen (14) discussed vacuum packaging of cured meats, many workers have reported preservation of color and flavor in cured meats when oxygen was removed by vacuum packaging (1, 5, 18, 25, 27, 28, 29, 31).

Some doubt remains as to the effect of vacuum packaging on microorganisms on cured meats. Several investigators have reported that various microorganisms were eliminated or inhibited when cured meats were packaged under vacuum (7, 13, 29, 31).

On the other hand, in Ulrich's studies (29), microorganisms normally present on bacon were not inhibited by vacuum packaging. He found that all the microorganisms present were bacteria—90% micrococci, 7% sporeformers, and the remaining 3% lactobacilli. Hankins et al. (10) also reported that the genus Micrococcus was the predominant organism in bacon. Lactobacilli have been commonly reported to occur on vacuum-packaged meats (1, 7, 11, 24).

Whether or not vacuum packaging inhibits the growth of microorganisms is still questioned. Ingram (13) suggested that it is wrong to generalize for a variety of meat products. The effect of packaging may depend not only on permeability of the wrapper to oxygen, carbon dioxide, and perhaps water, but equally on the exact kind of meat and on the nature and number of bacteria present.

Obviously, further work is needed to help clarify the influence of packaging materials, atmosphere of packages, and type of product on the quality factors mentioned. Our study was undertaken to compare effects of various packaging materials when meats were packaged in vacuum, in carbon dioxide, and in air. Bacon and hamburger were analyzed for color retention, shelf-life, and microbial counts. Consideration was given to growth of anaerobes, particularly Clostridium perfringens.

¹Present address: Department of Health, Education, and Welfare, Atlanta, Georgia 30323.
Materials and Methods

For studies in which bacon was inoculated, *C. perfringens* ATCC strain 3624 was used. Vegetative cells were grown in cooked meat medium (Difco Labs., Detroit, Mich.), incubated for 24 hr at 37 C, and then used to inoculate bacon samples. One milliliter of inoculum (approximately \(3 \times 10^7\) cells) was spread over the bacon strips.

The recovery of *C. perfringens* from food samples was demonstrated by the use of sulfite-polynymxin-sulfadiazine (SPS) agar (2) with the pouch method developed by Bladel and Greenberg (4). The SPS agar was placed into plastic film pouches made of a laminated film (Miltonprint cubate) for 24 hr at 37 C. The outer layer, Saran (polyvinylidene chloride), 0.1 ml, was the middle film, and an inside layer of polyethylene, 2 ml. This material is designated by the manufacturer as "Nealam" 60601. Black colonies suspected of being *C. perfringens* were confirmed in motility-nitrate medium (2).

Total aerobic counts were made by sampling 40 g of bacon from each package, homogenizing the meat in 360 ml of 0.1% peptone water, and enumerating on plate count agar (Difco). Plates were incubated at 30 C for 72 hr. Lactobacilli MRS broth (Difco), with the addition of 15 g per liter of Bacto agar (Difco), was used for pour plates for enumeration of lactobacilli. These plates were incubated at 30 C for 48 hr. Colonies were later tested with 10% hydrogen peroxide solution to determine the catalase reaction.

Fresh ground hamburger was purchased from a local food store and packaged the same day. Hamburger was the fresh meat chosen because *C. perfringens* were present naturally. The meat was formed into 50 g patties and packaged with different materials under vacuum and in air. The meat was stored for 30 days at about 5 C in a display case. At periods of 0, 1, 3, 6, 10, 15, 20, and 25 days of storage, the packages were opened and sampled by adding 40 g of meat to 360 ml of sterile water. The suspension was homogenized with an Osterizer (John Oster Mfg. Co., Milwaukee, Wisconsin) and appropriate dilutions were made in 99 ml of sterile 0.1% peptone solution. Total aerobic counts were determined from pour plates of plate count agar (Difco) incubated at 37 for 48 hr.

Counts of *C. perfringens* were made for each meat sample by using the pouch method described previously; pouches were incubated at 37 C for 24 hr.

Bacon was obtained from a packing plant in Iowa and selected on the basis of uniformity of color, amount of lean, and general appearance. Similar strips were chosen for all treatments. Bacon was packaged in a laminated film known as "Curpolene"; this material is described in a later section. Packages contained about 12 slices of bacon per bag, giving little free gas space. After packaging, all samples were stored in the dark at 5-7 C for 48 hr before being placed in a display case at 5 C under fluorescent light. Color changes were determined by a panel of judges. Bacterial counts were made from duplicate samples at 0, 3, 7, 17, 21, and 30 days of storage.

Samples of bacon were packaged in air, in commercial CO\(_2\), and under a vacuum of 28.5 inches of Hg. Vacuum packaging was accomplished by using a chamber equipped with a sealing bar heated to 300 F (149 C). The chamber was evacuated to 28.5 inches of Hg by means of a vacuum pump (Precision Scientific Co., Model 75), the package sealed, and the vacuum released. Barometric and manometric measurements also were made to determine adequacy of evacuation.

Carbon dioxide packaging was accomplished by using the same chamber. After removal of air by the vacuum pump, the chamber was flushed with commercial carbon dioxide to 5 mm of Hg. The chamber was again evacuated, and a second flushing with carbon dioxide was performed. Bags were then sealed and stored as described. During dark storage for 48 hr, carbon dioxide was slowly absorbed by the meat to form a vacuum-type package.

Initially, color was compared with color plates in the Maerz and Paul "Dictionary of Color" (19) and later with color prints made of photographs of packaged bacon. Color was evaluated by a panel of four judges who were previously trained in examining bacon for differences in color. The four judges were selected on the basis of their ability to detect color differences.

Six different types of packaging materials were used for packaging fresh meat (Table 1). These films gave a rather wide range of various oxygen and water vapor transmission rates; these variables were considered to have a possible effect on numbers of aerobic and anaerobic bacteria in or on the meat.

With bacon, only one type of packaging material, "Curpolene 200", was used. "Curpolene 200" (Curwood, Inc., New London, Wisc.) is a four-ply lamination consisting of 50 gauge oriented polypropylene, .0005 inch polyethylene, cellophone coated on two sides with 250 gauge Saran, and .002 inch low-density polyethylene (high melt index). The oxygen permeability is 0.5 cc per 100 in.\(^2\) per 24 hr at 72 F and 30% relative humidity, and the water vapor transmission rate is 0.25 per 100 in.\(^2\) in 24 hr at 90-95% relative humidity and 100 F. The bags measured 9 x 5 inches and were already sealed when received in the laboratory. They were then cut open, the bacon placed inside, and the packages sealed.

Oxidative rancidity of bacon was determined by the 2-thiobarbituric acid (TBA) test described by Tarladgis et al. (28). The optical density of the sample was read in a Beckman DU spectrophotometer against the blank at a wavelength of 532 m. The reading was multiplied by the factor 7.8 to convert to milligrams of malonaldehyde per 1000 g of meat. It is recognized that the curing process (sugars, nitrates, etc.) may affect the TBA test. The bacon used in this work was commercially cured under controlled conditions and selected for uniformity. All samples were considered to be comparable. Initial TBA values for three sample packages of bacon were 0.187, 0.187, and 0.203, with a mean value of 0.192.

Results and Discussion

Fresh hamburger.

When fresh meat was packaged with various materials, there was a tendency for films having high oxygen permeability to support aerobic growth and to limit growth of anaerobic organisms, as compared with less-permeable materials. The number of aerobic bacteria per gram of fresh ground hamburger stored at 5 C in vacuum and nonvacuum conditions is shown in Fig. 1. Counts were averaged for the various types of films employed. For both packaging treatments, aerobic bacteria increased in numbers throughout the 30-day period. Vacuum packaging of hamburger resulted in bacterial growth attaining a stationary phase after six days of storage, whereas hamburger packaged in air showed continued development of aerobes until 21 days of storage, after
Effects of Carbon Dioxide

Figure 1. Aerobic bacteria recovered from fresh hamburger packaged under vacuum or in air.

which these organisms began to decline. Vacuum packaging appeared to have little effect on aerobic counts in fresh hamburger. This may have resulted from the specific product; hamburger has air incorporated in the meat and also is known to be contaminated with various types of organisms, resulting in high initial counts.

Halleck et al. (9) indicated that vacuum packaging in a cellophane-pliesfilm laminate resulted in greater growth inhibition than with other packaging materials used in his studies. In our work, “Nealam”, Maraflex 4-8, and Clysar were used for vacuum and nonvacuum packaging. The higher the oxygen permeability of the film, the greater the difference in aerobic counts between vacuum-and air-packaged meat. In Clysar (oxygen permeability of 5 cc/100 in²/24 hr), there was a stationary phase after 6 days of storage for aerobes recovered from vacuum-packaged hamburger. In “Nealam” and Maraflex 4-8 (both with very low oxygen permeabilities), little or no difference was observed for vacuum-and air-packaged samples. A possible explanation for these results is that with the more permeable film, aerobic bacteria have oxygen more readily available to be utilized before anaerobic conditions exist.

Clostridium perfringens grew in fresh hamburger packaged under vacuum or in air with the same film, but the maximum rate of growth was faster in vacuum conditions. Figure 2 shows the number of C. perfringens per gram of fresh ground hamburger stored at 5°C in evacuated and nonevacuated packages, with counts averaged for all materials tested. Anaerobic bacteria were able to grow in meat packaged with all types of films used regardless of oxygen permeability. The vacuum packages allowed initial growth of C. perfringens up to 3 days of storage followed by a decline in number of cells. When air was permitted to remain in packages, a lag phase occurred until the third day, followed by a brief increase in population of C. perfringens and then a decline in growth. It is possible that anaerobic conditions in air packages favorable to growth of C. perfringens were reached only after the aerobic bacteria had utilized the oxygen present in the package.

Cured bacon

One objective of studies with bacon was to determine differences in color retention with vacuum, air, and carbon dioxide packaging. Table 2 shows re-
TABLE 1. TYPE AND PROPERTIES OF PACKAGING MATERIALS USED FOR FRESH MEAT

<table>
<thead>
<tr>
<th>Packaging material</th>
<th>Type of film</th>
<th>Water vapor transmission g/100 in.2/24 hr at 1 atm</th>
<th>Oxygen permeability rate ml/100 in.2/24 hr at 1 atm</th>
<th>% relative humidity and temperature of test</th>
</tr>
</thead>
<tbody>
<tr>
<td>195-MSAD-80</td>
<td>One coated side cellophane</td>
<td>60 g</td>
<td>100 ml</td>
<td>80-100% RH 77 F (25 C)</td>
</tr>
<tr>
<td>Nealam 60601</td>
<td>Polyester/Saran/Polyethylene</td>
<td>0.45-0.65 g</td>
<td>0.4-0.7 ml</td>
<td>90% RH 100 F (38 C)</td>
</tr>
<tr>
<td>Maraflex 4-8</td>
<td>Polyester/Polyvinyl alcohol/Polyethylene</td>
<td>0.23 g</td>
<td>0.7 ml</td>
<td>90% RH 100 F (38 C)</td>
</tr>
<tr>
<td>Maraflex 4-8</td>
<td>Polyester/Polyvinyl alcohol/Polyethylene</td>
<td>0.03 g</td>
<td>0.03 ml</td>
<td>90% RH 100 F (38 C)</td>
</tr>
<tr>
<td>Maraflex 7f</td>
<td>Polyester/aluminum foil/Polyethylene</td>
<td>25 g</td>
<td>5 ml</td>
<td>80-100% RH 77 F (25 C)</td>
</tr>
<tr>
<td>Clyser 200 pp-2A</td>
<td>Polyolefin Polypropylene coated</td>
<td>1.8 g</td>
<td>45 ml</td>
<td>95% RH 100 F (38 C)</td>
</tr>
<tr>
<td>Pliofilm 75 B.f.</td>
<td></td>
<td></td>
<td></td>
<td>Dow cell 73 F (23 C)</td>
</tr>
</tbody>
</table>

Effects of Carbon Dioxide

TABLE 2. COLOR EVALUATIONS OF BACON WITH AND WITHOUT 48 HOURS OF STORAGE IN THE DARK

<table>
<thead>
<tr>
<th>Without 48 hr dark storage</th>
<th>Days stored at 5 C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Packaging treatments</td>
<td>0</td>
</tr>
<tr>
<td>Vacuum</td>
<td>1</td>
</tr>
<tr>
<td>CO₂</td>
<td>2</td>
</tr>
<tr>
<td>0.5% Residual O₂</td>
<td>3</td>
</tr>
<tr>
<td>3.5% Residual O₂</td>
<td>4</td>
</tr>
<tr>
<td>Air</td>
<td>5</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>With 48 hr dark storage</th>
<th>Days stored at 5 C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Packaging treatments</td>
<td>0</td>
</tr>
<tr>
<td>CO₂</td>
<td>1</td>
</tr>
<tr>
<td>Vacuum</td>
<td>1</td>
</tr>
<tr>
<td>Air</td>
<td>1</td>
</tr>
</tbody>
</table>

1Lowest score = most desirable product
**TABLE 3.** *Clostridium perfringens* recovered from inoculated bacon

<table>
<thead>
<tr>
<th>Packaging treatment</th>
<th>Days stored at 5°C</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
</tr>
<tr>
<td>Control, vacuum packaged</td>
<td>&lt;10⁰</td>
</tr>
<tr>
<td>Control, air packaged</td>
<td>&lt;10⁰</td>
</tr>
<tr>
<td>Inoculated, air⁡</td>
<td>41</td>
</tr>
<tr>
<td>Inoculated, vacuum⁡</td>
<td>20</td>
</tr>
</tbody>
</table>

⁡Inoculum = 3.3 x 10⁷ cells per ml
⁡No. of cells x 10⁷ per gram

**TABLE 4.** TBA tests for packaged bacon

<table>
<thead>
<tr>
<th>Atmosphere</th>
<th>Days at 5°C</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>mg of malonaldehyde per 1000 g of bacon</td>
</tr>
<tr>
<td>Vacuum</td>
<td>0.192</td>
</tr>
<tr>
<td>Air</td>
<td>0.192</td>
</tr>
<tr>
<td>CO₂</td>
<td>0.192</td>
</tr>
</tbody>
</table>

¹Mean of two samples per treatment.

Counts of lactobacilli. Lactobacilli on the vacuum-packaged meats were inhibited until 14 days and then the bacterial counts increased in about the same manner as those for air-packaged bacon. The product in the air package had the lowest initial count, but lactobacilli increased steadily until 21 days of storage.

No *C. perfringens* were detected on control samples throughout the storage period of 30 days at 5°C (Table 3). This correlates with work of Ulrich (29), Cavett (7), and Ingram (13). Examinations of bacon samples inoculated with *C. perfringens* showed no signs of growth of the organism even after 18 days. However, *C. perfringens* persisted on inoculated samples during the storage period.

Since packaged bacon may not always be consumed at one time in the home, it was of interest to determine effects of various packaging treatments when packages were opened and then closed and stored. Packages were stored for 48 hr in the dark and then opened and resealed. They were placed in the bottom shelf of the refrigerator at 5°C. Carbon dioxide, after three days, inhibited bacterial growth in opened packages, whereas the bacon in the vacuum and air packages showed increases in bacterial counts throughout most of the storage period. Packages of meat treated with CO₂, even after being opened, still possessed sufficient absorbed carbon dioxide in the tissue to inhibit bacterial growth. The gas therefore exerted a residual effect.

Another trial was performed in which after initial storage for 0, 3, 7, 14, 21, or 30 days, packages were opened, folded closed, and then stored in the refrigerator for three days. Both vacuum and CO₂ treatments produced a lag in bacterial counts for the first 7 days of storage. Counts on bacon packaged in air increased steadily until 21 days of storage.

Table 4 shows the average of two tests for oxidative rancidity by the TBA method. Thiobarbituric acid values generally were lower for bacon packaged in CO₂ than for samples packaged in air or under vacuum. Obviously, CO₂-packaged bacon appeared to develop less rancidity than vacuum-packaged bacon during holding for as long as 30 days at 5°C.

From the results of this study, it may be concluded that vacuum-or CO₂-packaging plays an important role in bacterial growth and color retention in meat products. Gas permeability of various films influences effectiveness of packaging treatments. Vacuum packaging of fresh hamburger had little effect on growth of aerobic bacteria but did influence rate of growth of *C. perfringens*. The higher the oxygen permeability of the packaging film, the larger was the difference in aerobic counts between vacuum and non-vacuum packaging.
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REFERENCES


THE FATE OF RADIO-LABELED BOTULINAL TOXIN WITHIN THE BODY OF THE RAT

B. L. VERMILYEA and H. W. WALKER

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(Received for publication October 29, 1969)

ABSTRACT

Botulinal toxin type A, bovine serum albumin, and rat serum albumin were labeled with iodine-131 and injected intraperitoneally into white rats. Examination of tissues at various time intervals up to 48 hr showed many similarities in the distribution patterns in the rat for these radiolabeled proteins. Relatively large amounts of radioactivity were found in the blood, diaphragm, stomach, spleen, intestine, and tail of most rats. Passive immunity for botulinal toxin reduced the amount of localization in the stomach, spleen, intestine, and thyroid and increased the rate of excretion of botulinal toxin.

The neurotoxins produced by the six types of Clostridium botulinum paralyze certain motor nerves in the animal by suppressing release of acetylcholine at the cholinergic synapses (3, 4, 6, 8). The mechanism by which these toxins reach the synapses has not been resolved; ferritin-labeled toxin, however, has been observed by electron microscopy (13) in neuromuscular junctions. Botulinal toxin radiolabeled with sulfur-35 (9) and tetanal toxin (5) labeled with iodine-131 have been traced through the bodies of mice. The purpose of this study was to compare the fate of botulinal toxin and of nontoxic proteins in the rat by using materials labeled with iodine-131.

MATERIALS AND METHODS

Radion labeling of proteins

Purified type A botulinal toxin was obtained from Dr. Edward Schantz, U. S. Army Biological Laboratory, Fort Detrick, Md. Type A equine antitoxin was obtained from the U. S. Public Health Service, Communicable Disease Center (CDC), Atlanta, Georgia. The antiserum was rehydrated in a 1:1 solution of sterile glycerin and distilled water and contained a final concentration of 10 International Units (IU) antisera per ml (1).

Crystalized and lyophilized bovine serum albumin (BSA) (Sigma Chemical Co., St. Louis, Mo.), fraction V rat serum albumin (BSA) (Pentex, Inc., Kankakee, III.) and purified type A botulinal toxin were radiolabeled by a method similar to that described by Kirelenko et al. (5). A small crystal of elemental resublimed iodine was added to 0.1 ml of sodium iodine 131 (Nuclear Chicago, Des Plaines, III.) and allowed to react for 20 min at 37 C. Fifty microliters of this preparation was added to 1 ml volumes of the previously mentioned proteins which were dissolved in Bacto-FA buffer, pH 7.2 (Difco Laboratories, Detroit, Michigan) and allowed to react for 4 hr at 4 C. Unbound iodine was separated by filtration of the protein solutions through a 30 cm column packed with Sephadex G50, fine (Pharmacia, Uppsala, Sweden), which was swelled in the same phosphate buffer used to dissolve the proteins. The protein material was eluted with phosphate buffer, pH 7.2, flowing at a rate of 0.5 ml per min. Two-ml fractions were collected. One ml of each fraction was measured for radioactivity; the second ml was mixed with cold 3% trichloroacetic acid; the precipitate and the supernatant were assayed for radioactivity.

Radioactivity was measured on a Picker Autowell II gamma radiation counter. The upper and lower level discriminators were set in such a way as to count the 131I photo peak without counting background noise. The counter was recalibrated before each counting period with 137Cs and 125I standards.

Loss of toxicity by the botulinal toxin during the radiolabeling procedure was measured by LD50 bioassays. A toxic solution was divided into two equal parts; one part was radiolabeled with iodine-131, the other was not. The unlabeled toxin was filtered through a 30-cm column packed with Sephadex G50. The buffer system and flow rate were identical to those described previously except 5-ml fractions were collected. The fraction starting with the 15th ml and stopping with the 20th ml contained most of the proteinaceous material and was used for LD50 determinations. The column was washed for several hours with phosphate buffer to insure removal of all proteinaceous material. The radiolabeled sample was treated similarly. Both samples were diluted with physiological saline in tenfold dilutions from 104 to 10-4. Five albino mice per dilution were inoculated intraperitoneally with 0.5 ml of toxic material. Nonradiolabeled and radiolabeled toxins were inoculated into 50 animals for each of the two toxin groups. The animals were observed for 96 hr and LD50 values were calculated using the method of Reed and Muench (10).

Detection of radioactive material in animal tissues

Distribution patterns in rat tissues were established for bovine serum albumin, rat serum albumin, purified type A botulinal toxin, and purified type A botulinal toxin introduced into rats that had received 0.6 IU of type A antitoxin 24 hr earlier. The antiserum was introduced by intraperitoneal injection into the rat's hind leg muscle.

Female albino rats, weighing 140 to 160g, were obtained from Simonsen Laboratories, White Bear Lake, Minn. Two rats were used for each of the sampling periods, which were 1, 2, 4, 8, and 24, and 48 hr from the time of introduction of the radiolabeled protein into the animals. In each determination the rats were anesthetized with ether, and the radiolabeled protein was introduced by intraperitoneal inoculation. Sampling began 60 min after the last pair of rats was inoculated. Only one-half the lethal dose of toxin was given the rats to insure survival through the 48th hour of sampling.

At each sampling period two rats were anesthetized with chloroform and exsanguinated by cardiac puncture. After death, animals were stretched on a small animal operating...
board and opened ventrally from chin to tail and the following tissues were removed: brain, lymph node in the neck, thyroid gland, blood, heart, lung, liver, spleen, kidney, stomach, small intestine, a portion of the spinal column containing muscle tissue, bone and central nervous cord, and the last 3-4 cm of the tail. Each tissue was placed in a disposable 5 ml microbeaker and weighed. The tissue samples in the microbeakers were folded and put into glass counter tubes and counted twice for periods of 60 sec to assay for radioactivity. The count, corrected for decay factor, was divided by the tissue weight to obtain count per min per g of tissue, or the specific count of the tissue. The specific counts for each of the tissues from the duplicate animals in each sampling period were averaged to obtain the average specific count for the pair of rats.

A volume of radiolabeled protein material equal to the volume used for injection of each of the animals was counted along with the tissue samples to determine the total number of counts received by each animal in each distribution pattern determination. This value was constant within treatment groups since all animals received equal volumes of material, but varied among treatment groups. Radiological dosages per animal varied from 0.6 x 10^6 counts per minute (CPM) to 1.6 x 10^6 CPM. The total radioactive dose per animal was used to correct all specific counts statistically to a common base to enable a direct comparison of the values from different experiments. Statistical adjustments were calculated using analysis of covariance (II).

**Elimination rate determination**

Sublethal doses of radiolabeled toxin were placed on pieces of dry bread, which were then coated with a thin layer of peanut butter. The toxic peanut butter sandwiches were fed to rats that had fasted for 24 hr. Rats were kept in metabolism cages, and samples of urine and feces were collected at 12 hr intervals for 48 hr. The excretory samples as well as a reference sample of toxin equal in volume to that consumed by each rat were assayed for radioactivity. The total count was divided by the count obtained for the reference sample to yield the percentage of radioactive material lost by excretion. The effect of passive immunity on the rate of elimination of toxin was determined by injecting 0.5 IU of type A antiserum into the hind leg muscles of two rats 24 hr before feeding them toxic peanut butter sandwiches.

Differences in excretion rates between rats that had been fed radiolabeled protein and that had been inoculated intraperitoneally with the same material were also determined. Two rats were fed peanut butter sandwiches on which radiolabeled bovine serum albumin had been placed. A second pair of rats was inoculated intraperitoneally with an equivalent dose of radiolabeled bovine serum albumin. The counts from the excretory products collected in metabolism cages were divided by the weight of the excreta to obtain the specific count.

**Recovery of toxin from tissues**

A lethal dose of radiolabeled type A botulinic toxin was inoculated intraperitoneally into two white mice. Following death, the tissues listed previously were assayed for radioactivity. Tissues were homogenized in 0.85% sterile saline using a Sorvall Omni-Mixer Homogenizer. Homogenization was generally complete in 3 to 4 min at maximal speed in the microhomogenizer attachment. Samples were centrifuged at 12,350 x g for 30 min at 2 C in the Sorvall RC2-B centrifuge and 0.2 ml of the supernatant material from each tissue was inoculated intraperitoneally into a 25 g female albino mouse. A second 0.2 ml of the supernatant material was mixed with an equal volume of type A antiserum and inoculated intraperitoneally into a white mouse.

**RESULTS AND DISCUSSION**

The radiolabeling procedure produced toxin and serum albumins with high radioactive counts. Filtration through Sephadex G50 was satisfactory for removal of the unbound iodine from the toxin solution. In the final preparations, bovine serum albumin precipitated by trichloroacetic acid (3%) contained approximately 99.5% of the radioactivity, and precipitated botulinic toxin contained 95.0% of the total radioactivity.

The biological activity of botulinic toxin type A was reduced by radiolabeling with iodine-131 from 315,000 mouse units of toxin per ml to 315 mouse units of toxin per ml. Addition of iodine to the toxic solution resulted in the formation of a precipitate after incubation for several hours at 4 C. Precipitate did not form in the toxic solution to which no iodine was added. Brazis et al. (2) reported that iodine in concentrations of 7.7 mg/l of water (pH 6.25, 25 C) inactivated 99.99% of partially purified type A botulinic toxin within 60 sec. The actual amount of iodine on a weight per volume basis was not known since the radioactive iodine sample was calibrated only in terms of total radioactivity in millicuries; however, it was much less than the amount of iodine used by Brazis et al. (2). Despite the loss of biological activity, a toxic solution with moderate biological activity and high radioactivity after radiolabeling was obtained. The biological activity of the radiolabeled toxin sample used in the tissue distribution patterns was approximately 1000 mouse units per ml of solution.

The biological activity of the radiolabeled toxin possibly resulted from the presence of unlabeled toxin. Toxic activity for mice and high levels of radioactivity in the blood of rats inoculated with sublethal quantities of toxin would suggest that a portion of the radiolabeled toxin was biologically active. It would be expected that unlabeled and labeled toxin would be diluted in the body of the rat and that labeled molecules contributed to the lethal effect in mice. No direct evidence is available to substantiate this assumption, however.

Toxin used for elimination studies was placed in a food sample that would be readily eaten by the rats and that would provide protection for the toxin. Non-immune rats eliminated only 50 to 55% of total radioactivity in 24 hr and 63 to 70% in 48 hr. Passively immune rats excreted radiolabeled toxin more rapidly than nonimmune animals; after 24 hr, 70 to 73% of the total activity was excreted by immune rats, and after 48 hr, they had eliminated 83 to 88%. Thus, immunized rats eliminated nearly as
much activity in 24 hr as was eliminated in 48 hr from nonimmunized rats. Elimination values of paired rats seldom differed by more than 5%.

Treatment of urine samples with 3% trichloroacetic acid indicated less than 1.0% of the total radioactivity was precipitable. Radioactive iodine was probably not attached to protein, but rather to breakdown products. No attempt was made to determine if urine and feces contained biologically active toxin in quantities lethal to mice.

The specific counts for urine and feces from rats fed radiolabeled BSA were much higher than the specific counts for excretory products from intraperitoneally inoculated rats. Animals fed radiolabeled protein apparently degraded and excreted the material in a shorter time than did animals that received protein by intraperitoneal inoculation. If the assumption were made that radiolabeled toxin behaved in the same manner as BSA, it would explain why intraperitoneal doses of toxin were more potent than oral doses; that is, rats digested and excreted a greater amount of the oral dose than of the intraperitoneal dose of type A botulinal toxin.

Blood was the only tissue that produced death in mice when inoculated intraperitoneally. Mice protected with specific antitoxin survived; thus, it was concluded that toxin was present in quantities lethal to mice. Rogers et al. (12) reported the presence of toxin in the blood of humans during clinical botulism. Pak and Bulatova (9), on the other hand, found accumulation of toxin to be least in the blood and most in the lungs of white mice injected intravenously with type B toxin labeled with sulfur-35. Perhaps other methods of inoculation of the animal, such as intravenous, intramuscular, or oral, produce absorption patterns differing from those when the intraperitoneal route is used. In addition, Lamanna and Hart (7) have suggested that the mode of inoculation may influence the amount of toxin required for

Figure 1. Distribution patterns of radioactivity for the blood, heart, lung, and diaphragm of rats receiving radiolabeled bovine serum albumin, rat serum albumin, toxin, and toxin plus antitoxin.
Figure 2. Distribution patterns of radioactivity for the liver, spleen, kidney, and stomach of rats receiving radio-
labeled bovine serum albumin, rat serum albumin, toxin, and toxin plus antitoxin.

d lethality because of the different barriers encountered during systemic absorption.

In most of the tissues sampled, the largest portion of radioactivity was found either in the particulate or trichloroacetic acid precipitated material. The small intestine appeared to be an exception to that observation. Most of the radioactivity for this particular tissue was found in the supernatant. The thyroid gland, in which one would expect to find free iodine, revealed very little radioactivity in non-
precipitated material. The tail was too tough to be broken up by the homogenizer, so the entire structure was counted and recorded as particulate count.

Radioactive material found in the nonprecipitable samples might have been amino acids and short peptides to which iodine was bound. These materials probably resulted from the degradation of the botulinal toxin in the rat's body. Ability of the rat to excrete large quantities of radioactive material in urine indicated that the toxin was being broken down into excretable products.
Distribution patterns were determined with proteins that were inoculated into rats by the intraperitoneal route; the normal route of entry of toxins and proteins into an animal's body is by the oral route. This method of inoculation was used because of ease of introducing the radiolabeled material into a number of animals within a short time and of assurance that equal volumes of protein were received by each animal. Feeding radiolabeled proteins to rats was unsatisfactory because of inability to control the amount of food consumed and the time in which it was consumed.

The amount of radioactivity localized per gram of tissue during the seven sampling periods is presented in Table 1. Radioactivity from the nontoxic proteins localized to a high degree in the thyroid, blood, and diaphragm. Although localization was less pronounced for the group of rats receiving bovine serum albumin, all groups accumulated relatively high total specific counts in the stomach, spleen, intestine, and tail. Very little localization occurred in the lymph node or brain with any of the groups.

Figures 1, 2, and 3 show the relative concentrations of radioactivity in different tissues at the various sampling times. A comparison of these distribution patterns shows more similarities than differences between the toxic and nontoxic proteins. Bovine and rat serum albumins, although the former is foreign and the latter is native to the rat, displayed almost identical distribution patterns. These, in turn, were similar to the patterns obtained for sublethal doses of toxins. The highest specific counts for diaphragm, spleen, and intestines of rats inoculated with rat serum albumin were observed at the 1 hr sampling period, and the counts decreased with each successive sampling. Radioactive counts for all proteins generally increased to a maximal value at either 2 or 4 hr and then declined during successive sampling periods in blood, heart, lung, liver, stomach, and tail. This pattern was duplicated for the kidney and spinal column in all but the group of rats receiving antitoxin and toxin.

The main effect of passive immunity was the markedly reduced magnitude of the count in tissues...
such as the stomach, spleen, intestine, diaphragm, and thyroid over that observed for the nonimmune rats that received only toxin. The more rapid excretion of radioactive material by immune than by nonimmune animals may partially explain this observation. The values reported by Kirilenko et al. (5) for radiolabeled tetanin toxin in tissues were similarly reduced when animals were passively immunized before injection of toxin.

ACKNOWLEDGMENTS

We wish to express our sincere appreciation to Dr. M. L. Kaebler, Department of Microbiology and Preventive Medicine, College of Veterinary Medicine, Iowa State University, for his assistance with radiological counting.

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REFERENCES


ANNOUNCEMENT OF CERTIFICATION AUTHORIZATIONS, BAKING INDUSTRY SANITATION STANDARDS COMMITTEE

The Office of (Equipment) Certification of the Baking Industry Sanitation Standards Committee announces the Authorization of equipment in compliance with BISSC Standards for the following companies. For clarity, the company for which certification has been approved is listed under the specified standard number and subject. This Certification Authorization list has been prepared and officially released as of February 8, 1970.

Standard No. 1—Flour Handling Equipment
Champion Machinery Company, Joliet, Illinois
Fred D. Pleney Company, Columbus, Ohio
G. A. Sewer Rondo Sales, Inc., Wood-Ridge, New Jersey
Yarway Corporation, Blue Bell, Pennsylvania

Standard No. 2—Dough Troughs
Union Steel Products Company, Albion, Michigan

Standard No. 3—Mechanical Intermediate Proofer
G. J. Benier, Den Haag, Holland

Champion Machinery Company, Joliet, Illinois
N. V. Machinefabriek De Ridder, The Hague, Holland

Standard No. 5—Cake Depositors, Fillers and Icing Machines
Food Equipment Development Corp., Mentor, Ohio

Standard No. 6—Horizontal Mixers and Vertical Mixers
Baker Perkins, Inc., Saginaw, Michigan
Champion Machinery Company, Joliet, Illinois
Hobart Manufacturing Company, Troy, Ohio
E. T. Oakes Corporation, Islip, Long Island, New York
Triumph Manufacturing Company, Cincinnati, Ohio

Standard No. 7—Conveyors
Alto Corporation, York, Pennsylvania
Baker Perkins, Inc., Saginaw, Michigan
Champion Machinery Company, Joliet, Illinois
Lanham Machinery Company, Atlanta, Georgia
G. A. Sewer Rondo Sales, Inc., Wood-Ridge, New Jersey

Continued on Page 108
E-3-A SANITARY STANDARDS FOR THERMOMETER FITTINGS AND CONNECTIONS USED ON EGG PRODUCTS EQUIPMENT

Serial #E-0900

Formulated by
International Association of Milk, Food and Environmental Sanitarians
United States Public Health Service
United States Department of Agriculture
Institute of American Poultry Industries
Dairy and Food Industries Supply Association

It is the purpose of the IAMFES, USPHS, USDA, IAPI and DFISA in connection with the development of the E-3-A Sanitary Standards program, to allow and encourage full freedom for inventive genius or new developments. Thermometer Fittings and Connections which are developed and which so differ in design, material construction, or otherwise, so as not to conform with the following standards, but which in the opinion of the manufacturer or fabricator are equivalent or better, may be submitted at any time for the joint consideration of IAMFES, USPHS, USDA, IAPI and DFISA.

E-3-A STANDARDS FOR THERMOMETER FITTINGS AND CONNECTIONS

The following fittings and connections, when manufactured to the specification of dimension and construction listed below, shall be classified as E-3-A Standard Thermometer Fittings and Connections:

<table>
<thead>
<tr>
<th>Part Name</th>
<th>Page No.</th>
<th>Drawing No.</th>
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<tbody>
<tr>
<td>3A1 Indicating Thermo. Fitting (4&quot; side wall Conn.)</td>
<td>2</td>
<td>3A-101-01</td>
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<tr>
<td>3A1 Recording Thermo. Fitting (4&quot; side wall Conn.)</td>
<td>3</td>
<td>3A-101-02</td>
</tr>
<tr>
<td>3A2 Indicating Thermo. Fitting (5-11/16 wall Conn.)</td>
<td>2</td>
<td>3A-101-01</td>
</tr>
<tr>
<td>3A2 Recording Thermo Fitting (5-11/16 wall Conn.)</td>
<td>3</td>
<td>3A-101-02</td>
</tr>
<tr>
<td>3A3 Indicating &amp; Recording Thermo. Fitting (Cover Insertion)</td>
<td>4</td>
<td>3A-101-03</td>
</tr>
<tr>
<td>Adapters for 3A3 Fitting</td>
<td>4</td>
<td>3A-101-03</td>
</tr>
<tr>
<td>3A4 Indicating Thermometer Fitting (For Pipe Lines)</td>
<td>5</td>
<td>3A-101-04</td>
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<td>3A4 Recording Thermometer Fitting (For Pipe Lines)</td>
<td>6</td>
<td>3A-101-05</td>
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<tr>
<td>3A4 3 in 1 (Split type) Recording Thermometer Fitting (For Pipe Lines)</td>
<td>7</td>
<td>3A-101-06</td>
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<td>Removable Nut for 3A1, 3A2, and 3A4 Fittings</td>
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<td>3A7 Thermometer Well (Short) for indicating and recording thermometers on egg storage tanks</td>
<td>8</td>
<td>3A-101-09</td>
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<tr>
<td>3A8 Thermometer Well (Long) for indicating and recording thermometers on egg storage tanks</td>
<td>9</td>
<td>3A-101-10</td>
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</tbody>
</table>

Additional Thermometer fittings and connections may be added to the E-3-A approved list as the demand arises. In each case the new thermometer fittings shall be assigned 3A drawing numbers and page numbers, and shall thereby be considered to be incorporated into this standard, after acceptance by IAMFES, USPHS, USDA, IAPI and DFISA.

A. Material:

All E-3-A standard thermometer fittings and connections shall be constructed throughout of stainless steel, nickel alloy, or equally corrosion resistant material, that is non-toxic and non-absorbent.

a. All product contact surfaces shall be finished to an equivalent of not less than 120 grit finish properly applied.

b. All outside surfaces must be smooth.

B. Construction:

These fittings shall conform, in general, to the construction illustrated in the applicable 3A standard thermometer fitting and connection drawing. Those parts, such as ferrules, which have a similar counterpart in the 3A Standard for Sanitary Fittings, shall conform dimensionally to that standard.

C. Gaskets:

Single service gaskets having product contact surfaces, if used, shall be of the sanitary type. The material used for multiple service gaskets of the rubber or rubber-like type shall conform to the applicable provisions of the "3-A Sanitary Standards for Multiple-Use Rubber and Rubber-Like Materials Used as Product Contact Surfaces in Dairy Equipment, Serial #1800."

These standards shall become effective June 23, 1970.
3A1 & 3A2 (Type RN) Indicating Thermometer For Tanks and Vats (Side Wall Connection)

3A1 for 4" size
3A2 for 5 11/16
3A1 & 3A2 3 in 1 Fitting for Recording Thermometers and Controllers
(For Jacketed Tanks & Vats)

3A STANDARD THERMOMETER FITTINGS & CONNECTIONS
3A-101-02
3A3 Fitting

Flange Type Adapter

Sleeve Type Adapter

(Adapters shown to illustrate acceptable methods of adjusting a thermometer stem length to fit a specific tank)

3A3 Umbrella-Flange Fitting For Cover Insertion Of Indicating Or Recording Thermometer Bulbs

3A Standard Thermometer Fittings & Connections

3A-101-03
3A4 (Type RN) 
INDICATING THERMOMETER 
FOR PIPE LINES

3A STANDARD 
THERMOMETER 
FITTINGS & CONNECTIONS

3A-101-04
REMOVABLE UNION NUT

This nut can be removed over the ferrule. Its use is optional for instruments that would not otherwise permit removal. Design as illustrated or equivalent is acceptable.

INTEGRAL 3A4 FERRULE
SANITARY FITTING

13H NUT

3A4 (Type RN)
RECORDING THERMOMETER BULB FOR PIPE LINES
FLANGE FOR SPLIT FERRULE

BEVEL SEAT FOR SPLIT FERRULE

1/2" OR LARGER HEX UNION NUT .(13H)

1/2" OR LARGER SANITARY FITTING

3A4 3 IN 1 FITTING FOR
RECORDING THERMOMETERS AND CONTROLLERS
(PIPE LINE FORM)

3A STANDARD
THERMOMETER
FITTINGS & CONNECTIONS
3A-101-06
3A7 THERMOMETER WELL (SHORT)
FOR INDICATING & RECORDING THERMOMETERS ON MILK STORAGE TANKS

DAIRY RECORDER

GROUND & POLISHED SURFACE
FREE FROM PITS

0.180" TAPER PER FT. 25\% FIT (SURFACE ENGAGEMENT) WITH TAPER GAUGE.

3A STANDARD THERMOMETER FITTINGS & CONNECTIONS

3A-101-09
3A8 THERMOMETER WELL (LONG)
FOR INDICATING & RECORDING THERMOMETERS
ON MILK STORAGE TANKS

3A STANDARD
THERMOMETER
FITTINGS & CONNECTIONS
3A-101-10
E-3-A SANITARY STANDARDS FOR PUMPS FOR LIQUID EGG PRODUCTS

Serial #E-0200

Formulated by
International Association of Milk, Food and Environmental Sanitarians
United States Public Health Service
United States Department of Agriculture
Institute of American Poultry Industries
Dairy and Food Industries Supply Association

It is the purpose of the IAMFES, USPHS, USDA, IAPI, and DFISA in connection with the development of the E-3-A Sanitary Standards program, to allow and encourage full freedom for inventive genius or new developments. Specifications for pumps which are developed and which so differ in design, material, and construction, or otherwise, as not to conform to the following standards, but which, in the fabricator's opinion, are equivalent or better, may be submitted for the joint consideration of the IAMFES, USPHS, USDA, IAPI, and DFISA at any time.

E-3-A STANDARDS FOR CENTRIFUGAL AND POSITIVE ROTARY TYPE PUMPS

A. MATERIAL:

1. All metal pump parts having any surface in contact with the product shall be constructed of stainless steel, nickel alloy, or equally corrosion resistant material that is nontoxic and nonabsorbent.

   a. All egg product contact surfaces shall be finished to an equivalent of not less than 120 grit finish properly applied.

   b. All outside surfaces shall be smooth and easily cleanable.

2. Exteriors of structural parts not in contact with the product shall be of corrosion resistant material with a smooth finish; or shall be rendered corrosion resistant or painted, and shall be constructed as to be easily cleanable.

3. Pump impellers or rotors, and cases or stators which operate in conjunction with a metallic counterpart, may be made of, or covered with, rubber or rubber-like materials. Rubber or rubber-like materials used for pump impellers or rotors, and cases or stators, shall be of such composition as to retain their surface and conformation characteristics under conditions encountered in normal use and cleaning operations.

4. All rubber and rubber-like materials when used for specified applications shall meet the applicable provisions of the "3-A Sanitary Standards for Multi-Use Plastic Materials Used as Product Contact Surfaces for Dairy Equipment, Serial #2000."

B. CONSTRUCTION:

1. All egg product contact surfaces shall be readily removable or accessible for cleaning and inspection. All exterior surfaces shall be self-draining.

2. The parts forming the space between the motor and the pump body shall be constructed in such a way that they are easily accessible for cleaning, and drain freely.

3. If legs are used, they shall be smooth with rounded ends and no exposed threads. Legs made of hollow stock shall be sealed. On pumps with legs designed to be fixed to the floor the minimum clearance between the lowest part of the base and the floor shall be four inches.

   a. Readily portable pumps not permanently attached may have leg heights of 2 inches. (Readily portable pumps are defined as those having a base area of not more than one square foot, or, in the case of motor mounted pumps, an area encompassed by the legs that does not exceed one square foot.)

   b. Bases when used shall be constructed without ribs or flanges and shall have a smooth top and bottom surface.

4. Pumps which because of their size and type
cannot be mounted on legs, shall be mounted on a base designed for grouting and sealing.

5. The driving means between the impeller or rotor and the pump shaft shall be so arranged as not to form a pocket or crevice that is not readily cleanable.

6. There shall be no threads in the product zone, except where necessary for attaching the impeller to the shaft. In such case(s) the thread shall conform to the following drawing known as the "brass valve stem" thread. The threaded angles shall be not less than sixty degrees and with not more than eight threads to the inch, nor less than five-eights inch major basic diameter. The length of the nut shall not exceed three-quarters of the thread basic major diameter and the nut shall be of the open type.

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<tr>
<td>S.D. SINGLE DEPTH</td>
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<td>T.F. TOP FLAT</td>
<td>T.F. x 280 x P</td>
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<td>B.F. BOTTOM FLAT</td>
<td>B.F. x 280 x P</td>
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<tr>
<td>T.P.L. TH'RS PER INCH</td>
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7. All surfaces in contact with the product shall have smooth, rounded corners and shall be readily accessible for cleaning.

8. The rubber or rubber-like coating of pump impellers or rotors, and cases or stators (if covered) shall be bonded in such manner that the bond is continuous and mechanically sound, and so that in normal service the rubber or rubber-like material does not separate from the base metal. The final bond shall conform in all respects to the criteria established in paragraph A (4).

9. The surface of rubber or rubber-like covering of pump impellers or rotors, and cases or stators shall be equal in cleanability to stainless steel with 120 grit finish properly applied.

10. The surface of plastic pump impellers or rotors, and cases or stators, shall comply with the applicable provisions of the "3-A Sanitary Standards for Multiple-Use Plastic Materials Used as Product Contact Surfaces for Dairy Equipment, Serial #2000."

11. The plastic coating of pump impellers or rotors and cases or stators (if covered) shall be bonded in such a manner that the bond is continuous and mechanically sound so that in normal service the plastic material does not separate from the base metal. The final bond shall conform in all respects to the criteria established in paragraph A (6).

12. The finish of the product contact surface at the interface juncture of the plastic and metal shall conform with the requirements of paragraphs A (1) a. and B. (10).

C. OPENINGS:

Inlet and outlets shall conform with the 3-A "Sanitary Standards for Fittings Used on Milk and Milk Products Equipment and Used on Sanitary Lines Conducting Milk and Milk Products," Serial #0800, and Supplements thereto, as amended.

D. SHAFT SEAL:

Shaft seal shall be of the sanitary type easily removable for inspection and cleaning, and shall be constructed of material not injurious to eggs.

E. GASKETS:

Single service gaskets of the sanitary type, or removable rubber type gaskets that can be easily cleaned, shall be used.

F. MOUNTINGS:

Mountings of motor, pump, and drive shall be of sanitary construction and shall be either sealed to the base or mounted to permit easy cleaning with minimum clearance of not less than one inch.

G. SEALING:

A pump used as the timing pump of a high-temperature short-time pasteurizer shall be provided with easily accessible or externally visible means of sealing that will prevent its operation at greater capacity than that which gives legal holding time without breaking the seal.

These standards shall become effective June 23, 1970.

*Pending development of a standard procedure for measuring the cleanability of surfaces, conformance with this item may be judged by comparing the removal of standard soil from the rubber or rubber-like material and from the stainless steel having a 120 grit finish, when standardized cleaning procedures are used. A technique for such comparisons has been developed by Dr. O. W. Kaufman, Michigan State University.
E-3-A SANITARY STANDARDS FOR SIFTERS
FOR DRY EGG PRODUCTS
Serial #E-2600

Formulated by
International Association of Milk, Food and Environmental Sanitarians
United States Public Health Service
United States Department of Agriculture
Institute of American Poultry Industries
Dairy and Food Industries Supply Association

It is the purpose of the IAMFES, USPHS, USDA, IAPI, and DFISA in connection with the development of the E-3-A Sanitary Standards program, to allow and encourage full freedom for inventive genius or new developments. Dry Egg Products Sifters specifications heretofore or hereafter developed and which so differ in design, material, construction or otherwise so as not to conform with the following standards, but which in the opinion of the manufacturer or fabricator are equivalent to or better, may be submitted at any time for the joint consideration of IAMFES, USPHS, USDA, IAPI, and DFISA.

A. SCOPE:
This standard covers the sanitary aspects of sifters used for processing dry egg products. In order to conform with these E-3-A Sanitary Standards, sifting equipment for dry egg products shall comply with the following in design, material, and fabrication criteria.

B. DEFINITIONS:
(1) Product: Shall mean dry egg products.
(2) Product Contact Surfaces: Shall mean all surfaces with which the product may come in contact.
(3) Non-Product Contact Surfaces: Shall mean all other exposed surfaces.

C. MATERIAL:
(1) All product contact surfaces shall be of 18-8 stainless steel with a carbon content of not more than 0.08 percent, or other equally corrosion resistant metal, that is non-toxic and non-absorbent, except that:
   (a) Plastic materials may be used for screening media, screen frame assemblies, balls, gaskets, flexible connectors, and inspection port covers. These materials shall comply with the applicable provisions of the “3-A Sanitary Standards for Multiple-Use Plastic Materials Used as Product Contact Surfaces for Dairy Equipment, Serial #2000.”
   (b) Rubber and rubber-like materials may be used for balls, gaskets, flexible connectors, and inspection port covers. These materials shall comply with the applicable provisions of the “3-A Sanitary Standards for Rubber and Rubber-Like Materials Used as Product Contact Surfaces in Dairy Equipment, Serial #1800.”
   (c) Cotton, linen, silk, or synthetic fibers may be used for screening surfaces. These materials shall be non-toxic, relatively insoluble, easily cleanable, and shall not impart a flavor to the product.
   (d) Welded areas and the deposited weld material shall be substantially as corrosion resistant as the parent material.
   (e) Solder, when used, shall have a tin content not less than 50%, and the remainder shall contain no more lead than is necessary under good manufacturing practices, and shall be corrosion resistant, cadmium free, non-absorbent, and shall not impart any toxic substance to the product under conditions of intended use.
(2) All non-product contact surfaces shall be of corrosion-resistant material or material that is rendered corrosion-resistant. If painted, the paint used shall adhere. All non-product contact surfaces shall be relatively non-absorbent, durable and cleanable. Parts removable for cleaning having both product contact and non-product contact surfaces shall not be painted.

D. FABRICATION:
(1) All product contact surfaces except the screen surfaces shall be equivalent to a number 4 mill finish or 120 grit properly applied, or a number 2B mill finish on stainless steel sheets, free of imperfections such as chips, flakes, or pits. All joints shall be smooth and flush. All permanent joints in metallic product contact
surfaces shall be welded. All welded areas of product contact surfaces shall be at least as smooth as the adjoining surfaces. Solder may be used to smoothly fill the joints where the screen is attached to the frame.

(2) All appurtenances having product contact surfaces shall be easily removable for cleaning or shall be readily cleanable in place.

(3) All product contact surfaces shall be easily accessible and readily cleanable, either when in an assembled position or when removed. Removable parts shall be readily demountable.

(4) All internal angles of 135° or less on product contact surfaces shall have minimum radii of 1/8 inch, except as provided in D. (5).

(5) Gaskets shall be removable or continuously bonded so as to be smooth and easily cleanable. Gasket retaining grooves for removable gaskets shall be no deeper than 1/8 inch, except that a 3/32 inch radius is permissible where a standard 1/4 inch O-Ring is to be used. Grooves in gaskets shall be no deeper than their width and the minimum radius of any internal angle shall be not less than 1/8 inch unless the gasket is readily reversible for cleaning.

(6) All openings in the cover shall have raised rims and flanges of at least 3/8 inch.

(7) All openings in the cover not continuously in use, shall be provided with removable covers designed to prevent foreign material from entering the product zone.

(8) The sifter shall be constructed so as to provide for prompt continuous removal of rejected material.

(9) Woven stainless steel wire may be used for screening surfaces.

(10) Non-product contact surfaces to be painted shall be effectively prepared for painting.

(11) There shall be no exposed threads in product contact areas.

(12) All outside welded seams shall be smooth and waterproof. All mechanical joints shall be dust tight and splash proof.

(13) Legs, if used, shall be smooth with no exposed threads, and shall be of sufficient length to provide a clearance between the lowest fixed point of the machine and the floor of no less than 6 inches. If legs are hollow tube stock, they shall be effectively sealed. When legs are not used the base shall be designed to permit sealing to the mounting surface.

APPENDIX A

For the general guidance of sifter manufacturers and the egg drying industry, the following screen size openings may be considered as recommended openings to result in satisfactory screening of the listed dry egg products:

<table>
<thead>
<tr>
<th>Product</th>
<th>Sieve Designation (From ASTM 223.1)</th>
<th>Maximum mm</th>
<th>Sieve Opening (Inches approx.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Egg white spray</td>
<td>#40</td>
<td>0.42</td>
<td>0.0165</td>
</tr>
<tr>
<td>Dry whole egg &amp; yolk</td>
<td>#12</td>
<td>1.68</td>
<td>0.0661</td>
</tr>
<tr>
<td>Blends</td>
<td>#12</td>
<td>1.68</td>
<td>0.0661</td>
</tr>
</tbody>
</table>

It is recognized that larger screen size openings may be necessary for sifting certain special dry egg products (such as "instant" products) and for desired classification of products into different particle sizes. Openings referred to above are based on general experience as to what constitutes satisfactory screening to remove product lumps or potential product contaminants, and also on ability of most currently used sifters to successfully sift dry egg products through such size openings without excessive loss of fine powder into the "reject material" outlet. (Other factors also affect such loss, such as percent of "open area" in screen used, uneven flow rates to the sifter, ratio of screening surface area to drier capacity, amount and kind of mechanical energy applied to the screening surface, sifter design and construction, and nature of dry egg product being sifted.)

The desired screen opening dimensions may be obtained by a number of combinations of wire thickness and number of wires per inch. Various combinations allow a choice to obtain desired balance between screen strength and percent open area.

REFERENCES


APPENDIX B

RECOMMENDATIONS FOR CLEANING DRY EGG SIFTERS

I. DAILY CLEANING PROGRAM—The procedures set forth below should be followed as a daily cleaning program.

1. Completely dismantle and thoroughly vacuum or dry brush clean all product contact surfaces of the dry egg sifter. Reassemble as soon as
finished and make every effort to keep all parts dry.

2. Check sifter screen(s) for broken or displaced wires (threads) and for other openings around the frame of the sifter, which might permit the passage of unsifted product. Other parts of the sifter, including ball trays and balls, if used, should also be inspected for condition. Any necessary repair or replacement should be made as soon as possible.

3. Flexible rubber or cloth socks at the inlet and outlets of the sifter should be thoroughly cleaned daily, following the procedures as recommended for the sifter. At this time socks should be closely examined for holes, cracks or other damage. (To facilitate removal for cleaning, use of easily removable fastening devices are recommended.)

4. Thoroughly vacuum or dry brush clean all external parts of the sifter, including the sifter frame and drive mechanism.

II. WEEKLY CLEANING PROGRAM—The procedures set forth below should be followed at weekly intervals.

1. Completely dismantle as in I. above, remove all loose dry egg, then rinse all parts with clear water and follow by a thorough hand brushing of all parts using a general purpose cleaner. Rinse thoroughly to remove all evidence of cleaning solution or soil. It is recommended that hot water (170° F. or above) be used for rinsing in order to sanitize the equipment and to aid the subsequent drying. Allow all parts to air dry completely prior to reassembly. The wet wash should be done more frequently if necessary and should be done after each use if the sifter is not being used on a daily basis. After cleaning, drying and reassembly the powder outlet should be protected against recontamination.

III. GENERAL RECOMMENDATIONS

1. Vacuum cleaning is preferred to brush cleaning or cleaning with air under pressure as it decreases the dust drift problem to other areas of the plant.

2. Brushes or vacuum cleaner fittings used for cleaning product contact surfaces should not be used for cleaning non-product contact surfaces or for other uses which might result in contamination. Such brushes and special fittings should be stored in an enclosed cabinet when not in use. (For protection and housekeeping considerations, such cabinets preferably should be of non-wood construction and should have open mesh metal shelving.)

These Standards shall become effective June 23, 1970.
E-3-A SANITARY STANDARDS COVERING
HOMOGENIZERS AND HIGH PRESSURE PUMPS OF THE PLUNGER
TYPE FOR LIQUID EGG PRODUCTS

Serial #E-0400

Formulated by
International Association of Milk, Food and Environmental Sanitarians
United States Public Health Service
United States Department of Agriculture
Institute of American Poultry Industries
Dairy and Food Industries Supply Association

It is the purpose of the IAMFES, USPHS, USDA, IAPI and DFISA in connection with the development of the E-3-A Sanitary Standards program, to allow and encourage full freedom for inventive genius or new developments. Specifications for homogenizers and high pressure pumps of the plunger type which are developed and which so differ in design, material and construction, or otherwise, as not to conform to the following standards but which, in the fabricator's opinion, are equivalent or better, may be submitted for the joint consideration of the IAMFES, USPHS, USDA, IAPI and DFISA at any time.

E-3-A STANDARDS

Homogenizers and High Pressure Pumps of the Plunger Type to be used for the processing of liquid egg products to conform to E-3-A Standards, shall comply with the following standards.

A. MATERIAL:
1. Power Frames: Exteriors shall be rust-proofed or shall be rendered corrosion-resistant or painted.
2. Cylinder Block and Fittings: All metal pump parts having any surface in contact with the product shall be constructed of stainless steel, nickel alloy, or equally corrosion-resistant material that is non-toxic and non-absorbent.
3. Homogenizing and/or Relief Valve, including the pressure regulating spring, shall be corrosion resistant and be easily removable for cleaning and inspection.

B. CONSTRUCTION:
1. Power Frame and Frame Covers:
   (a) Exteriors of structural parts not in contact with the product shall have a smooth finish, and shall be so constructed as to be easily cleanable.
   (b) The bottom of the frame or base shall be entirely closed. Any skirt extending below the bottom wall of the base shall be limited to a distance of 1-1/2" below the bottom wall.
   (c) All exterior surfaces shall be self draining. The minimum clearance between the lowest point on the base and the floor shall be not less than 4 inches.
2. Legs or Feet:
   (a) Legs or Feet shall be smooth with rounded ends and have no exposed threads.
   (b) Legs made of hollow stock shall be sealed.
3. Cylinder Block and Fittings:
   (a) All product contact surfaces shall be readily removable or accessible for cleaning and inspection.
   (b) All surfaces shall be machined or polished to not less than 120 grit finish properly applied. Sharp corners and edges shall be avoided.
   (c) There shall be no threads in contact with the product.
   (d) The cylinder block shall be constructed so that it will be possible to brush through all openings and passages in at least one direction. There shall be no dead-ended passages.
   (e) The space between the cylinder block and drive housing shall be readily accessible for cleaning, self-draining, and protected so that liquids will not enter the drive housing. This space shall be provided with a cover.
1. The cover over the plungers may, however, be designed to permit observation of packing leakage and of water applied to plungers without removing cover from the machines.
(f) All cylinder block fittings shall be of the flanged type.

(g) Inlet and outlet openings shall conform with the 3-A Sanitary Standard for Fittings. In the case of high pressure pumps they shall be of the flange type construction.

(h) Suction and discharge valve springs, when used, shall be smooth, have open ends, and be readily cleanable. Pitch of springs shall be such that open space between coils is not less than 3/32". Openings at ends of springs shall be not less than 3/32".

(i) Homogenizing pressure regulating springs and/or relief valve springs shall not be in contact with the product.

C. RECORDING OR INDICATING GAUGES:

1. The gauge shall be of the sanitary diaphragm or pressure bulb type.

2. The gauge connection to the gauge well shall be of the flange type.

3. The gauge parts having contact with the product shall be smooth, readily cleanable, and constructed of stainless steel, nickel alloy, or equally corrosion-resistant material that is non-toxic and non-absorbent.

D. GASKETS:

1. Gaskets shall be easily removable and cleanable or of the single service type. They shall be of rubber or rubber-like material, or of a plastic material, or of other suitable sanitary material.

   (a) If of rubber or rubber-like materials they shall conform to the applicable provisions of “3-A Sanitary Standards for Multiple-Use Rubber and Rubber-Like Materials Used as Product Contact Surfaces in Dairy Equipment, Serial #1800.”

   (b) If of plastic material, they shall conform to the applicable provisions of “3-A Sanitary Standards for Multiple-Use Plastic Materials Used as Product Contact Surfaces for Dairy Equipment, Serial #2000.”

E. PLUNGER AND VALVE ROD PACKING:

1. Packing shall be easily removable for inspection and cleaning.

2. Packing shall be of a reasonably impervious material.

F. NAMEPLATE:

1. A nameplate shall be permanently attached to the machine indicating:
   (a) Name of the Manufacturer
   (b) Model and Serial Number
   (c) Capacity

G. SEALING:

1. Homogenizers used as the timing pump of a high-temperature short-time pasteurizer shall be provided with easily accessible or externally visible means of sealing that will prevent its operation at greater capacity than that which gives legal holding time without breaking the seal.

These Standards shall become effective June 23, 1970.
INSULATED SERVER PACKS FOR TRANSPORTING FOOD FROM COMMISSARY TO THE TABLE

W. L. MALLMANN

Department of Microbiology and Public Health
Michigan State University, East Lansing 48823

(Received for publication November 14, 1969)

ABSTRACT

The insulated food server pack makes possible the placing of 9 oz portions in single service plastic containers of both hot and cold foods in a compartmentalized food tray at the commissary. When foods have initial temperatures of 180 °F or above for hot food and 32-35 °F for cold food, the food after 3 hr storage will have temperatures of 140 °F or above for hot foods and 45 °F or below for cold food at time of service.

At 180 °F, all disease-producing bacteria are destroyed in the foods and no contamination is possible during handling. The close seal between trays and compartments seals against dust contamination and heat transfer between compartments.

The search is continuous to find more economical and efficient methods of transporting food from the commissary to the table of the consumer. The public health aspects of a method, which appears to be economical and effective with built-in protection from contamination will be presented.

The equipment consists of compartmentalized plastic trays insulated on the edges and between compartments. Food is placed in separate throw-away plastic inserts that fit into the compartments. After food is heated or cooled in the inserts, both hot and cold food are placed in the same tray. The trays are then stacked on a special insulated base tray and covered with an insulated plastic cover (Fig. 1). To lessen temperature changes in the food compartments and to seal against dust and microbial contaminants, overlapping match joints seal each compartment and tray to the trays above and below. After the trays are strapped together, the stack can be placed in a plastic bag ready for transport. After each food service, the trays are washed in a regular dishwashing machine.

When foods are in transit from the commissary to the point of consumption, it is necessary to maintain temperatures in the foods either high enough or low enough to prevent growth of disease-producing microorganisms. The U.S.P.H.S. Food Service Sanitation Manual-1962 (8) states that perishable food must be held at temperatures above 140 °F or below 45 °F except for the period necessary for serving. In addition, any cooked foods should have been heated at temperatures lethal for disease-producing microorganisms.

It is interesting to note that Sternberg (6), in 1887, reported that such disease-producing organisms as the typhoid bacillus and staphylococcus were killed at 132 to 136 °F in 10 min. The method that he devised, called the thermal death point procedure, is still used. In 1899, Marshall (4) reported the tubercle bacilli in ground tubercles suspended in milk were killed at 155 °F in 20 min. He did not determine the minimum lethal temperature or exposure.

These references are cited to emphasize that the lethality of moist heat ranging from 130 to 150 °F for pathogenic bacteria has been known for 82 years.

Angelotti et al. (2), in 1960, determined lethal temperatures for salmonellae and staphylococci in menstrua of custard, chicken ala king, and ham salad. All organisms were killed at 150 °F in exposures ranging from 4 to 8 min.

After cooking, foods may be recontaminated and stored temporarily at temperatures favoring microbial multiplication. Most food poisoning outbreaks can be traced to improper storage. Hot food storage should be above the maximum temperature for growth of disease-producing bacteria. In 1912, Marshall (5) stated that the maximum temperature for growth of pathogenic bacteria is 10 to 15 °C higher than the optimum temperature. Pathogenic bacteria have an optimum growth temperature of 95 to 98 °F so their maximum growth temperature would be 113 to 116 °F. These figures were based on growth in nutrient media.

Maximum growth temperatures were determined by Angelotti et al. (2) for salmonellae and staphylococci in custard, chicken ala king, and ham salad. They found that no growth occurred during storage at 116 °F for all menstrua.

Angelotti et al. (1) also conducted studies on the growth of staphylococci and salmonellae in the same three menstrua at 40, 42, 44, 46, 48, and 50 °F over a period of 5 days. A very slight growth (0.014 generations) of salmonellae occurred at 40 °F in chicken ala king. At 46 °F, growth was limited to less than a generation except for salmonellae in chicken ala

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1Professor Emeritus
king where 3.1 generations occurred in 5 days.

METHODS AND MATERIALS

Fifteen stacks consisting of six insulated server trays each were used. Nine ounces of custard were placed in each entree insert, heated in an oven until the desired temperatures were attained, and individually transferred to the trays. As soon as one insert of custard was placed in a tray, another tray was placed on the stack. Each stack consisted of six trays. In the second tray from the top, two vials of each of two cultures and a thermocouple were immersed in the custard. The culture vials were warmed to approximately 110 F before being placed in the custard. All trays were held at room temperature for a minimum of 4 hr. Temperatures were recorded with a 12 point recorder.

Staphylococcus aureus (toxigenic strain) and Salmonella senftenberg 775W were used. The latter strain has a heat resistance greater than that of other salmonellae, shigellae, and coliform types. Fresh cultures were placed in 3 ml screw-capped vials in 2 ml portions of heart infusion broth and refrigerated until being warmed to insert in the custard.

Three experiments were performed. In the first, the custard in the five stacks of trays had initial temperatures of 189, 185, 183, 161, and 144 F when the vials were inserted in the custard. In the second, the custard in the five stacks of trays had initial temperatures of 140, 135, 130, 125, and 120 F when the vials were inserted. In the third, the custard in the five stacks of trays had initial temperatures of 50, 45, 40, 35, and 32 F.

At the end of 4 hr storage at room temperature, the vials were removed and immediately placed in cold storage until population counts were made. The vials used in the first experiment contained per milliliter 1.8 x 10^7 S. senftenberg 775W and 1 x 10^7 S. aureus. In the second and third experiment, the vials contained S. senftenberg 775W, 1 x 10^6 and S. aureus, 1 x 10^6.

RESULTS

When initial temperatures of the custard were 189, 185, 183, and 161 F, all bacterial cells were destroyed (Table 1). When the initial temperature was 144 F, only a few organisms from both cultures survived. The percentage reduction of S. senftenberg was 99.995.

When lower temperatures, 120-140 F (Table 2) were used on low populations, complete kill was obtained at 130, 135, and 140 F. There was no reduction of S. senftenberg at 120 F but no increase in population occurred. Apparently many of the cells of S. aureus died during the refrigeration period as evidenced by a population of 4 at 125 F. It will be noted, however, that staphylococcus populations were very low as evidenced by counts obtained in the third series ranging from zero to 90. Obviously, kill did occur at temperatures of 130, 135, and 140 F.

The temperature curves for initial storage temperatures of 189, 185, and 155 for a period of four hours are recorded in Fig. 2. When the initial temperature of the custard was 189 F, the temperature

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1 Aladdin Industries of Nashville, Tenn. supplied the Temp-Rite Servers.

**Table 1. The effect of heat in destruction of salmonellae and staphylococci in high populations in insulated servers.**

<table>
<thead>
<tr>
<th>Initial Temperature</th>
<th>Population after 4 hr storage</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S. senftenberg 775W</td>
</tr>
<tr>
<td>189 F</td>
<td>0</td>
</tr>
<tr>
<td>185 F</td>
<td>0</td>
</tr>
<tr>
<td>183 F</td>
<td>0</td>
</tr>
<tr>
<td>161 F</td>
<td>0</td>
</tr>
<tr>
<td>144 F</td>
<td>850</td>
</tr>
</tbody>
</table>

1 Initial population of S. senftenberg - 1.8 x 10^7
2 Initial population of S. aureus - 1 x 10^7

**Table 2. The effect of heat in destruction of salmonellae and staphylococci in low populations in insulated servers.**

<table>
<thead>
<tr>
<th>Initial Temperature</th>
<th>Population after 4 hr storage</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S. senftenberg 775W</td>
</tr>
<tr>
<td>140 F</td>
<td>0</td>
</tr>
<tr>
<td>135 F</td>
<td>0</td>
</tr>
<tr>
<td>130 F</td>
<td>0</td>
</tr>
<tr>
<td>125 F</td>
<td>225</td>
</tr>
<tr>
<td>120 F</td>
<td>22,250</td>
</tr>
</tbody>
</table>

1 Initial population of S. senftenberg - 1 x 10^6
2 Initial population of S. aureus - 1 x 10^6

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Figure 1. A stack of insulated trays with special insulated cover. (Photograph by courtesy of Aladdin Industries, Nashville, Tennessee.)
S. senftenberg \((1 \times 10^5)\) and S. aureus \((1 \times 10^6)\) were inserted into custard, complete kill occurred with initial temperatures of 140, 135, and 130 F (Table 2). These results indicate that although temperatures of the food might fall below 140 F, the temperature was sufficient during the 3 hr storage in the trays to continue to kill until the food fell to a temperature of 130 F.

The temperature curves for initial temperatures of 35, 45, and 48 F are presented in Fig. 4. Custards at initial temperatures of 32 and 35 F were below 40 F at 3 hr and below 45 F at 4 hr. Custard at an initial temperature of 45 was slightly below 50 F at 3 hr. Bacterial populations were relatively static at all refrigeration temperatures. Foods should be cooled to at least 35 F before being introduced into the trays.

**DISCUSSION**

An examination of the temperature curves show that in the entree compartment of the insulated trays, 9 oz of heated custard in each of the six trays will hold to a temperature above 140 F and the chilled custard at temperatures not exceeding 45 F for 3 hr if the food was introduced into the trays at temperatures in excess of 180 F for hot food and the cold foods at temperatures of 32 to 35 F.

For short holding times, 0.5 to 1 hr, lower initial temperatures could be used and still stay within the limits 140 F or above and 45 F or below. However, in practice, the highest possible temperature for hot foods should be used.

If foods were left undisturbed in the insulated packs for 4 hr, the temperature would still be above 130 F for hot foods and below 45 F for cold foods. However, the hot foods would be below the optimum palatability temperatures of 140-150 F as recommended by Blaker et al. (3).

Thermal death points, as previously mentioned,

**Table 3. Effect of Cold in Preventing the Multiplication of Salmonellae and Staphylococci in Low Population in Insulated Servers.**

<table>
<thead>
<tr>
<th>Initial Temperature</th>
<th>Population after 4 hr storage</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. senftenberg</td>
<td>S. aureus</td>
</tr>
<tr>
<td>32 F</td>
<td>114,300</td>
</tr>
<tr>
<td>35 F</td>
<td>412,000</td>
</tr>
<tr>
<td>40 F</td>
<td>158,000</td>
</tr>
<tr>
<td>40 F</td>
<td>116,800</td>
</tr>
<tr>
<td>50 F</td>
<td>161,000</td>
</tr>
</tbody>
</table>

\(^1\)Initial population of S. senftenberg - \(1 \times 10^5\)

\(^2\)Initial population of S. aureus - \(1 \times 10^6\)
range from 132 to 136 F for 10 min. When a vial of *S. senftenberg* was introduced into custard at 130 F, sterility occurred in 4 hr and at 125 F, 99.98 percent were destroyed in 4 hr (Table 2). The organisms at 130 F were undoubtedly destroyed in less than 10 min exposure.

If salmonellae infected foods were placed in insulated servers at 140 F, even after 2 hr holding, lethal temperatures (125 F) would still be in effect.

From a public health standpoint, food at 120 F would still be safe because there would be no multiplication of pathogens even if recontamination occurred.

The mechanically sealed server trays are comparable to thermos bottles as far as external contamination is concerned. When servers are transported by truck, plastic bag covers give added protection against dust contamination.

This type of food service makes possible the use of a central commissary that can be utilized for service to airlines, railroads, school lunch service, hospital, and industrial plant feeding.

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**BAKING INDUSTRY STANDARDS**

**Continued From Page 88**

**Standard No. 8—Dividers, Rounders and Bun Machines**

G. J. Benier, Den Haag, Holland

Champion Machinery Company, Joliet, Illinois

N. V. Machinefabriek De Ridder, The Hague, Holland

Dutchess Bakers’ Machinery Co., Superior, Wisconsin

G. A. Sewer Rondo Sales, Inc., Wood-Ridge, New Jersey

**Standard No. 9—Bread Moulders**

Champion Machinery Company, Joliet, Illinois

N. V. Machinefabriek De Ridder, The Hague, Holland

G. A. Sewer Rondo Sales, Inc., Wood-Ridge, New Jersey

**Standard No. 10—Prefabricated Enclosure and Air Conditioning Equipment for Fermentation, Proofing, Cooling and Retarding**

Baker Perkins, Inc., Saginaw, Michigan

Industrial Air Conditioning Systems, Inc., Chicago, Illinois

Fred D. Pfenning Company, Columbus, Ohio

Union Steel Products Company, Albion, Michigan

**Standard No. 13—Bread, Cake and Roll Slicing, Wrapping and Bagging Machines**

Alto Corporation, York, Pennsylvania

Maine Machine Works, New York, New York


**Standard No. 14—Mechanical Ovens**

Baker Perkins, Inc., Saginaw, Michigan


Universal Oven Company, Westbury, New York

**Standard No. 15—Caster Assemblies and Wheels**

G. A. Sewer Rondo Sales, Inc., Wood-Ridge, New Jersey

**Standard No. 16—Doughnut Equipment**

DCA Food Industries, Inc., New York, New York

Joe Lowe Company, Englewood, New Jersey

**Standard No. 18—Emulsifiers and Homogenizers**

E. T. Oakes Corporation, Islip, Long Island, New York

**Standard No. 20—Liquid Ferment and Continuous Mix Processing Equipment**

Waukesha Foundry Company, Waukesha, Wisconsin

**Standard No. 21—Dough Chutes, Dough Hoppers, Dough Trough Hoists and Automatic Dough Trough Dumps**

Baker Perkins, Inc., Saginaw, Michigan

**Standard No. 22—Depanners and Delidders for Bakery Products**

Alto Corporation, York, Pennsylvania

Baker Perkins, Inc., Saginaw, Michigan

**Standard No. 25—Kettles and Kettle Agitators**

Hamilton Kettles Division, Cincinnati, Ohio

**Standard No. 27—Facilities for Handling and Storing Refined Liquid and Dry Sweetening Products**

Jamesbury Corporation, Worcester, Massachusetts

Victaulic Company of America, South Plainfield, New Jersey

Yarway Corporation, Blue Bell, Pennsylvania

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**References**


A Research Note

ROAD DUST AS A SOURCE OF SEDIMENT IN MILK AND CREAM

J. G. ARMSTRONG

Alberta Dairymen's Association Research Unit,
Department of Food Science, University of Alberta, Edmonton

(Received for publication October 13, 1969)

Abstract

Four tests have been conducted with two types of cans to determine the amount of dust that may enter milk and cream cans during transport over dusty roads. The least amount of dust was found in cans when they were filled with individual plastic covers. The greatest amounts of dust were found in cans of both types when waxed paper was placed under the lids. In these tests a tarpaulin over the truck, and the type of can, made no difference in the amount of dust entering the cans. Relatively large differences were observed, however, between individual cans.

Attempts to reduce amounts of extraneous matter in milk and cream received at dairy plants has led to questions respecting sources of contamination. In milk and cream shipped in cans, road dust is usually considered to be one of these sources (1). It is thought to enter cans prior to pick-up from road-side stands and during transit between farm and dairy plant. And, if cans are not carefully rinsed on the farm before filling, some may have entered while the empty can was in transit from plant to farm. Various suggestions have been made locally respecting ways of reducing or preventing this form of contamination. This paper reports results of tests conducted under late summer conditions to obtain information respecting the possible extent of contamination from road dust and the effectiveness of some control methods.

Methods

Twenty-four cans of eight Imperial gallon capacity were obtained for the tests. Twelve had "mushroom" or "umbrelli" tops and 12 had inset tops. In both groups the age of the cans varied from new to well-used, however, the proportion of new to old was not the same in the two groups and there were more relatively new cans among those with the "mushroom" top.

Two types of supplementary coverings were tested. One was a plastic cover that fitted over the top of the can and had an elastic band to provide a close fit about the neck of the can. The other consisted of waxed paper which was applied by placing the paper over the top of the open can and then pushing down the lid so that the paper formed a "filler" between the lid and the neck of the can.

The vehicle used for the road tests was a half-ton truck which had the box built up with boards to a height of approximately 6-inches above the tops of the cans. In order to carry all of the cans in each test and not overload the vehicle, each can was "filled" with only two Imperial gallons of water. Tap water was used for this purpose after tests showed it to contain negligible amounts of sediment. In each test, the truck with the filled cans was driven over a route of 30 miles on a recently-gravelled road at a speed of approximately 50 miles per hour. Each complete trip included, in addition, 15 miles of hard-surfaced road and city streets when leaving and returning to the laboratory. To ensure suitable dust conditions for the tests, another vehicle was driven ahead of the truck on the gravelled road.

Before each test the cans were carefully rinsed using tap water and a vigorous swirling action. In each test, after returning to the laboratory, the lid was removed from each can and the can was rotated so the water in the can washed the whole of the interior surface including the neck of the can. A 16-fluid ounce "mixed sample" of the water was then taken and filtered through a circular area 0.40 inches in diameter (3). The amount of sediment was estimated by comparing the density of the disc so obtained with the Research Unit's "Guide for Sediment in Milk" (2). The amount of sediment observed was recorded for each can.

Results and Discussion

Difference between tests

The nature of the four tests is indicated below and the average amount of sediment observed in each test is shown in parentheses (mg/16 fl oz). Because the results indicate the amounts observed and as the cans were only one-quarter full, the concentrations of sediment recorded were approximately four times that which would have been expected if full cans had been used in the tests. (a) Test 1. Cans in the open truck (0.02). (b) Test 2. Cans in truck covered by a tarpaulin (0.02). (c) Test 3. Individual plastic covers on each can and truck top open (0.01). (d) Test 4. Wax paper placed under lids of cans and truck top open (0.04).

These results indicate that under the conditions of the tests: (a) a tarpaulin cover over the truck had no effect on the amount of dust entering the cans; (b) placing waxed paper under the lids of the cans increased the amount of dust entering the cans; and (c) of the treatments tested, only the individual plastic covers on each can reduced the amount of dust found in the cans.

Difference between types of cans

Averages of the results for the two types of cans on the four tests are presented in Table 1. It is apparent that in these tests the type of can made little
or no difference in the amount of dust entering the cans.

Differences between individual cans

From the records for each can on the four tests, averages were calculated to provide a measure of the differences between cans. The results presented in Table 2 show that the differences were considerable (five-fold in the extremes).

In relation to the amounts of extraneous matter observed previously in milk and cream samples (2, 3, 4), even the maximum contamination found (0.05 mg/16 fl oz) must be considered light—especially since the dust entering the can was distributed through only 2 gallons and not 8 gallons as would have been true had the cans been full. Furthermore, in this test the cans probably received a greater than normal exposure to dust. The exposure would appear to be much greater than that likely to be encountered when cans are transported by trucks with closed all-metal bodies. However, at many Alberta dairy plants proportions of the cream and milk received in cans are delivered by producers and in these instances, exposure to dust may be as great, or perhaps even greater, than that provided in these tests. In either event, the results suggest that road dust is not the most important source of contamination in milk and cream delivered in cans to manufacturing plants, but if protection from dust is desired, individual plastic covers for the cans are effective.

Acknowledgements

The author wishes to thank the National Research Council of Canada and the Alberta Agricultural Research Trust for their financial support and Mr. Leonard Ewanyk for technical assistance in this investigation.

References


TEXAS WATER POLLUTION CONTROL CONFERENCE

The Texas Water Pollution Control Association and the University of Houston are co-sponsoring the 9th Texas Water Pollution Control Association Conference at the University of Houston on July 9 and 10, 1970.

The conference program will consist of technical research, development, and operations papers of industrial water pollution control processes and systems, chemical analysis of water and wastewaters, biological aspects of water pollution, domestic and industrial wastewater system planning and management, wastewater collection systems design, Galveston Bay water quality program reports, and oil pollution control technology. Abstracts will be welcomed until May 1, 1970.

For complete information write or call Dr. H. Nugent Myrick, University of Houston, Cullen College of Engineering, Department of Civil Engineering, Environmental Science and Engineering Program, 3801 Cullen Boulevard, Houston, Texas 77004, phone 713-748-6600, Extensions 562-563, or 564, or Ronald D. Sadow, Monsanto Company, P. O. Box 1311, Texas City, Texas 77590, phone 713-945-4431, Extension 2771.
EXAMINATION OF FROZEN VEGETABLES FOR SALMONELLAE

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ABSTRACT

Salmonellae detection methods developed for other foods were not entirely satisfactory for frozen vegetables because of the presence of slow lactose-fermenting organisms that resembled the pathogens on various differential media. The problem was solved by raising the incubation temperature of brilliant green agar to 42 C. Examination of a number of frozen vegetables, many collected in processing line surveys, failed to uncover salmonellae. Data were obtained that suggested the organisms were inhibited by the normal microflora of frozen vegetables.

Although negative epidemiological evidence indicates that salmonellae are not a problem in frozen vegetables, their presence in this class of food cannot be ruled out completely. Closely related organisms, the coliforms, are numerous (5) and it is possible that salmonellae at times also are present. The fact that most vegetables are cooked prior to consumption could explain their excellent public health record.

The objectives of this study were to evaluate established methods to determine whether they would permit the detection of low numbers of salmonellae in vegetables and to determine whether the organisms were present in these foods.

MATERIALS AND METHODS

The samples of frozen vegetables were obtained both from the lines of area processors and from retail markets. Those collected in line surveys were transported to the laboratory over ice and then held at -23 C until cultured.

Methods recommended by Galton et al. (2) served as a guide for the enumeration procedures that were examined. In general, the method consisted of aseptically weighing 30 g of frozen vegetables into 500 ml erlenmeyer flasks containing 100 ml of a selective enrichment broth. After an incubation of 24-48 hr at 37 C, the broth was streaked on plates containing a selective agar. Colonies resembling salmonellae were transferred to triple sugar iron agar (TSI) slants that were incubated 24 hr at 37 C. The cultures resembling salmonellae on TSI were tested further for lactose fermentation, indole, methyl red, Voges-Proskauer and citrate reactions, production of urea, and the ability to grow in the presence of 0.0075% potassium cyanide (1). Commercial dehydrated media were used for preparation of selective broths and agars. The other media and tests were made according to procedures of Edwards and Ewing (1).

The effectiveness of various procedures for recovering salmonellae was evaluated by inoculating vegetables with one of two serotypes: Salmonella anatum VPHL 2383 or S. senftenberg 775W. In these trials, at least five replica 30 g samples of vegetable were cultured in a given selective enrichment broth so that most probable number (MPN) recovery figures could be related to the number of cells added.

RESULTS AND DISCUSSION

The initial studies revealed that frozen vegetables were commonly contaminated with organisms that resembled salmonellae when grown on TSI slants as well as on the following selective agars: brilliant green, brilliant green-sulfadiazine, SS agar, desoxycholate, bismuth sulfite, and MacConkey. They differed from salmonellae in that they were urease positive and grew in the KCN medium. Other tests showed they did not produce H2S on TSI slants, were indole and methyl red negative, and were positive for the Voges-Proskauer and citrate tests. When inoculated into lactose broth containing brom-cresol-purple indicator, it was observed that this sugar was weakly fermented after 72 hr at 37 C. It was concluded from these results that the organisms responsible for the false-positive reactions were slow lactose-fermenting members of the Klebsiella-Aerobacter group.

The type of selective enrichment broth influenced the number of “false-positive” organisms recovered (Table 1). Although as illustrated here, the selenite broths usually yielded fewer false-positives than the tetrathionate broth, all three media gave counts that would seriously interfere with attempts to detect low numbers of salmonellae in frozen vegetables.

Pure culture studies on some of the slow lactose-fermenting isolates and the two salmonella serotypes suggested that raising the incubation temperature to 42 C would inhibit many of the salmonella-like organisms. A similar observation has been reported for meat (3). This was studied further by culturing frozen vegetables that had been inoculated with low numbers of salmonellae. In these studies, the elevated incubation temperature was applied to the second step, the selective agar, because it was assumed that at this point salmonellae, if present, would be more numerous and thus better able to initiate growth. Results of numerous recovery experiments, as illustrated in Table 2, confirmed that the higher incubation...
temperature permitted detection of low numbers of S. anatum and S. senftenberg and eliminated the problem of slow lactose fermenting organisms. On the basis of this work, the method adopted for examination of frozen vegetables was to incubate 30 g of vegetable in 100 ml of tetrathionate broth for 24 hr at 37 C. The broth was streaked on a brilliant green agar plate which was incubated 24 hr at 42 C. Pink colonies on this medium were transferred to TSI slants. Isolates showing typical growth on TSI were inoculated into aqueous vegetable extracts. The extracts, prepared by homogenizing one part by weight of vegetable with nine parts of water, simulate the soluble materials found on the surfaces of processing equipment. When non-sterile extracts were inoculated, only limited growth occurred (Table 4). It can be seen that most growth occurred in the bean extract which had the lowest initial total count

and lima beans were examined. The results were that no salmonellae were isolated and the slow-lactose fermenting organisms presented few difficulties. Although some brilliant green agar plates exhibited pink growth, usually in areas of confluent colonies, none of these cultures resembled salmonellae when transferred to TSI agar slants.

Most of the samples collected in the line surveys (Table 3) were cultured within a month after freezing and, therefore, death of salmonellae during prolonged frozen storage does not appear responsible for the negative results. A high degree of sanitation during processing also is not a likely explanation since a number of the samples yielded total counts of over 10^6/g (Table 3).

Evidence that vegetable processing lines simply do not provide a favorable environment for salmonellae was obtained when low numbers of S. anatum were inoculated into aqueous vegetable extracts. The extracts, prepared by homogenizing one part by weight of vegetable with nine parts of water, simulate the soluble materials found on the surfaces of processing equipment. When non-sterile extracts were inoculated, only limited growth occurred (Table 4). It can be seen that most growth occurred in the bean extract which had the lowest initial total count.

### Table 1. Incidence of Organisms That Might Be Confused with Salmonellae

<table>
<thead>
<tr>
<th>Vegetable</th>
<th>Enrichment broth</th>
<th>Salmonella-like organisms, MPN/g^b</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peas</td>
<td>Selenite-F</td>
<td>1.8</td>
</tr>
<tr>
<td></td>
<td>Selenite-cystine</td>
<td>3.6</td>
</tr>
<tr>
<td></td>
<td>Tetrathionate</td>
<td>17</td>
</tr>
<tr>
<td>Green beans</td>
<td>Selenite-F</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>Selenite-cystine</td>
<td>6.1</td>
</tr>
<tr>
<td></td>
<td>Tetrathionate</td>
<td>430</td>
</tr>
<tr>
<td>Corn</td>
<td>Selenite-F</td>
<td>9.3</td>
</tr>
<tr>
<td></td>
<td>Selenite-cystine</td>
<td>9.3</td>
</tr>
<tr>
<td></td>
<td>Tetrathionate</td>
<td>52</td>
</tr>
</tbody>
</table>

^aThe samples, from line surveys, yielded the following "total" counts per g X 10^3: peas, 340; green beans, 370; corn, 410. ^bBased on the number of brilliant green agar plates exhibiting pink colonies.

### Table 2. Recovery of Salmonella anatum from Vegetables Using Tetrathionate Enrichment Followed by streaking on Brilliant Green Agar Incubated at 42 C.

<table>
<thead>
<tr>
<th>Trial</th>
<th>Vegetable</th>
<th>&quot;Total&quot; count per g (X 10^3)</th>
<th>Salmonellae/30 g vegetable</th>
<th>MPN recovered^b</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>Peas</td>
<td>23</td>
<td>4</td>
<td>2.2</td>
</tr>
<tr>
<td></td>
<td>Green beans</td>
<td>26</td>
<td>4</td>
<td>9.2</td>
</tr>
<tr>
<td></td>
<td>Corn</td>
<td>110</td>
<td>4</td>
<td>9.2</td>
</tr>
<tr>
<td>B</td>
<td>Peas</td>
<td>76</td>
<td>1.5</td>
<td>0.22</td>
</tr>
<tr>
<td></td>
<td>Green beans</td>
<td>400</td>
<td>3.5</td>
<td>0.51</td>
</tr>
<tr>
<td></td>
<td>Corn</td>
<td>240</td>
<td>3.5</td>
<td>1.6</td>
</tr>
</tbody>
</table>

^aCalculated from plate counts made on the inoculum. ^bBased on the number of cultures giving typical reactions on TSI, KCN, and urea media. Uninoculated controls were negative on TSI slants.

### Table 3. Vegetable Samples from Line Surveys Cultured for Salmonellae.

<table>
<thead>
<tr>
<th>Vegetable</th>
<th>Source</th>
<th>No. of samples</th>
<th>&quot;Total&quot; count range (X 10^3)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peas</td>
<td>Airlift to freezer</td>
<td>5</td>
<td>140-900</td>
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<tr>
<td></td>
<td>Flume to freezer</td>
<td>4</td>
<td>8-59</td>
</tr>
<tr>
<td>Beans</td>
<td>Inspection belt</td>
<td>3</td>
<td>140-480</td>
</tr>
<tr>
<td></td>
<td>Flume to freezer</td>
<td>6</td>
<td>30-440</td>
</tr>
<tr>
<td></td>
<td>Airlift to freezer</td>
<td>3</td>
<td>22-200</td>
</tr>
<tr>
<td>Corn</td>
<td>Airlift to freezer</td>
<td>4</td>
<td>500-1000</td>
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<tr>
<td></td>
<td>Flume to freezer</td>
<td>3</td>
<td>3000-3000</td>
</tr>
</tbody>
</table>

### Table 4. Multiplication of S. anatum in Non-sterile Vegetable Extracts.

<table>
<thead>
<tr>
<th>Extract</th>
<th>Initial &quot;total&quot; count</th>
<th>S. anatum inoculum</th>
<th>Extract after 24 hr at 25 C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pea</td>
<td>20 x 10^6</td>
<td>750</td>
<td>10^6</td>
</tr>
<tr>
<td>Bean</td>
<td>87 x 10^6</td>
<td>44</td>
<td>10^6</td>
</tr>
<tr>
<td>Corn</td>
<td>11 x 10^6</td>
<td>44</td>
<td>negative</td>
</tr>
</tbody>
</table>

^a1 part vegetable homogenized with 9 parts water ^bhighest dilution yielding salmonellae on brilliant green agar
and that salmonellae were not recovered from the corn extract which was most heavily contaminated. In other trials in which extracts were filter-sterilized prior to inoculation, populations of over \(10^4\) salmonellae per ml were obtained. These results suggest that the microflora intrinsic to vegetable processing lines, largely acid formers (4), may suppress the growth of a *Salmonella* cell that, by chance, is introduced onto a given piece of equipment. This would explain why salmonellae have not been found in frozen vegetables even though these foods are processed under very open, exposed conditions.

**Acknowledgement**

This investigation was supported in part by Public Health Service grant FD 00214 from the Food and Drug Administration.

**References**


**Holders of 3-A Symbol Council**

**Authorization on February 20, 1970**

"Questions or statements concerning any of the holders of authorizations listed below, or the equipment fabricated, should be addressed to C. A. Abele, Secretary-Treasurer, 2617 Hartzell St., Evanston, Ill. 60201."

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<td>Cherry-Burrell Corporation</td>
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**0204 Pumps for Milk and Milk Products Revised, as Amended**

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<td>G &amp; H Products Corporation</td>
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<td>145R</td>
<td>ITT Jabsco, Incorporated</td>
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<td>Waukesha Foundry Company</td>
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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

7 CP Division, St. Regis (10/19/56)
1243 W. Washington Blvd., Chicago, Illinois 60607

75 Manton-Gaulin Mfg. Co., Inc. (9/26/57)
44 Carden Street, Everett, Massachusetts 02149

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

131 Almont Welding Works, Inc. (9/3/60)
4091 Van Dyke Road, Almont, Michigan 48003

98 Beseler Steel Products, Inc. (3/24/58)
417 East 29th, Marshfield, Wisconsin 54449

70 Jacob Brener 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

190 Eastern Industries, Limited (11/18/66)
830 Blvd., Lemire, Drummondville, Quebec, Canada

121 The J. A. Gosselin Co., Ltd. (12/9/59)
P. O. Box 280, Drummondville, Quebec, 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)
Holdingford, Minn. 56340

144 Portersville Stainless Equipment Div., Gibson Industries, Inc.
Portersville (Butler County), 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)
10th & Howell Streets, 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 and Supplements 2, 3, 4, 5, 6, and 7, As Amended

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

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

82 Cherry-Burrell Corporation (12/11/57)
105 W. Adams St., Chicago, Ill. 60603

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

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

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

190 Gray Company, Inc. (12/8/67)
60 Eleventh Ave., N.E., Minneapolis, Minn. 55413

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

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

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

200 Paul Mueller Co. (3/5/68)
1016 Phelps St., Springfield, Mo. 65601

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

89 Sta-Rite Industries, Inc. (12/23/68)
343 Wright Street, Delavan, Wis. 53115

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

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

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

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

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)
Neposet 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. (9/4/58)
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. (8/15/58)
5th & Tilgham Streets, Chester, Pennsylvania 19013

38 CP Division, St. Regis (10/19/56)
1243 W. Washington Blvd., Chicago, Illinois 60607

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

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

The DeLaval Separator Co. (11/18/69)
350 Duchess Turnpike, Poughkeepsie, N. Y. 12602
holders of 3-a symbol

1303 farm milk cooling and holding tanks — 
revised, as amended

11r cp division, st. regis (7/25/56)
1243 w. washington street, chicago, illinois 60607
11r dairy craft, inc. (10/28/59)
st. cloud industrial park, st. cloud, minn. 56301
4r dairy equipment company (6/15/56)
1919 s. stoughton road, madison, wisconsin 53716
92r de laval company, ltd. (12/27/57)
113 park street, south peterborough, ontario, canada
49r the de laval separator company (12/5/56)
duchess turnpike, poughkeepsie, n. y. 12602
10r garton 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)
635 laurel st., preston, ont., canada
12r paul mueller company (7/31/56)
1616 w. phelps street, 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
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

122 cherry-burrell corporation (12/11/59)
105 w. adams st., chicago, ill. 60603
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. thomson & sons, inc. (11/20/57)
1303 43rd street, kenosha, wisconsin 53140

1603 evaporators and vacuum pans for milk and
milk products, as amended

132 a.p.v. company, inc. (10/26/60)
137 arthur street, buffalo, new york 14207
111 blaw-knox company (2/12/59)
dairy equipment division
750 e. perry, buffalo, n. y. 14210
110 arthur harris & company (11/10/58)
210-218 north aberdeen street, chicago, illinois 60607
164 mura industries, inc. (4/25/65)
112 south park street, mora, minnesota 55051
107 c. e. rogers company (8/1/58)
8731 witt street, detroit, michigan 48209
185 marriott walker corporation (9/6/66)
925 east maple road, birmingham, mich. 48008

1702 fillers and sealers of single service containers,
for milk and milk products, as amended

192 cherry-burrell corporation (1/3/67)
2400 sixth st., s. w., cedarm Rapids, iowa 52404
139 exact weight sale company (4/15/68)
538 east town street, columbus, ohio 43215
137 ex-cell-o corporation (10/17/62)
p. o. box 386, detroit, michigan 48232
140 general films, inc. (4/23/63)
covington, ohio 55318
142 polygal company (4/15/63)
div. of inland container corp.
p. o. box 68074, indianapolis, indiana 46268
210 twintap, ltd. (2/4/70)
270 st. joseph blvd., lachine, quebec
211 twintap, inc. (2/4/70)
1133 avenue of the americas, new york, n. y. 10010

1901 batch and continuous freezers, for ice cream,
ices and similarly frozen dairy foods, as amended

168 cherry-burrell corporation (6/10/65)
575 e. mill st., little falls, n. y. 13365
154 cp division, st. regis (2/10/65)
1243 w. washington blvd., chicago, illinois 60607
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)
1616 w. phelps street, springfield, missouri 65801
195 paul mueller (canada) ltd. (7/6/67)
84 wellington st., sa., st. mary's 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 sing'e service containers

174 anderson bros. mfg. co. (9/28/65)
1303 samuelson road, rockford, illinois 61009
209 doughboy industries, inc. (7/23/69)
machine division
899 so. main ave., new richmond, wisc. 54017
193 triangle packaging machinery co. (1/31/67)
6555 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)
1243 w. washington blvd., chicago, illinois 60607
187 dairy craft, inc. (9/26/66)
st. cloud industrial park
st. cloud, minn. 56301
## ASSOCIATION AFFAIRS

### NOMINATIONS FOR OFFICES OF IAMFES, INC.—1970-1971

#### FOR SECOND VICE-PRESIDENT AND SECRETARY-TREASURER

WALTER WILSON

Walter Wilson was born April 29, 1922, on a dairy farm in Adair, Illinois.

Family moved to Southern California in 1926 where public schools were attended.

College education at University of California, Davis, was interrupted by three years service in the U.S.M.C.R., as an Amphibian Tractor Officer. He returned to Davis to receive an Associates of Science Degree in Dairy Manufacturing. During his working years he has also earned a B.S. in Biologic Science with a major in Public Health from Los Angeles State College.

Graduate work in Public Administration at University of Southern California has also been pursued at night school classes.

In 1947, Mr. Wilson was employed by Carnation Company in their Fresh Milk and Ice Cream operation in Los Angeles. He left Carnation for a similar job with Creameries of America which also had operational responsibilities of that Company's Los Angeles area plant.

In 1952, he was employed by Los Angeles County Health Department as a Dairy Sanitarian. In 1958, he was promoted to Chief of Section for Los Angeles County. In 1964, consolidation of Los Angeles City and County Health Department resulted in his being appointed as Chief of Milk Inspection Services both within Los Angeles County and a four county milk-shed located in Central California.

He is a past president of California Association of Dairy Sanitarians, member of California Association of Sanitarians (N.A.S.), past president of the California Dairy Industry Association, past president of Los Angeles County Health Sanitarians Association and member of the Conference of Local Health Administrations Inc. He is co-chairman host af-
The Wilson family lives in Whittier, California, a suburb of Los Angeles known best for being the hometown of President Nixon. The two younger children are high school students and the oldest is presently residing in Washington, D.C., where her husband is stationed at the Walter Reed Hospital.

A. P. Bell

A native of Colorado, A. P. Bell graduated from Colorado University with a B.S. in Civil Engineering and an option in Sanitary Science. He entered the field of Public Health as a Junior Engineer in the District of Columbia Health Department where he spent eight (8) years before moving to Louisville, Kentucky.

A. P. has headed the Division of Environmental Health for the Louisville and Jefferson County Department of Public Health since 1948 and has been active in a large number of health oriented associations and worked on many committees. His activities in the International date back to the formation of the Kentucky Chapter of which he proudly boasts Charter membership.

In addition to being an active participant in State and National affairs of the International Association, A. P. is well known in the fields of Food, Drugs and Sanitary Engineering. He has served as President of the Kentucky Public Health Association, Ohio Valley Food and Drug officials, and the Kentucky Affiliate. He has been a Director of the Conference of Local Environmental Health Administrators, and has served on many committees of the Engineering Section of the American Public Health Association.

He and his wife, Betty, live in suburban Louisville and are proud of their two (2) children, Ann and Mike, especially proud of their two (2) grandchildren.

Richard P. March is a professor in the Department of Food Science at the New York State College of Agriculture, Cornell University, Ithaca, New York. Until 1965, he devoted 75% of his time to extension work as a specialist in milk quality and fluid milk handling and processing, and the balance of his time in research and teaching courses in fluid milk processing and quality control. At present, extension accounts for 90% of his time with 10% for research activities.

He was raised in Massachusetts, majored in dairy industry at the University of Massachusetts, receiving a B. S. degree in 1944. After a tour in the U. S. Marine Corps, he entered the Graduate School at Cornell University to major in dairy industry, receiving an M.S. degree in 1948.

Professor March taught a one-year program in dairy manufacturing until its termination in 1951, at which time he was promoted from instructor to assistant professor. He became an associate professor in 1955, and full professor in 1965. In 1965 he also became department extension leader and is still serving in this capacity.

He is active in the New York State Association of Milk and Food Sanitarians, serving as secretary-treasurer from 1957 and executive secretary since 1967, secretary of the Dairy Industry Equipment Committee from 1952-57, secretary of the Farm

In both the State and International Associations he has served as chairman of a number of subcommittees including the Uniform Milkhouse Plans for the Northeast, Milk Transfer Systems, Sediment Testing, and Training Programs for Bulk Tank Truck Operators, and co-chairman of the Northeast Committee on Uniform Guidelines for Loose Housing Systems. In 1963 he was the recipient of the New York State Association's Dr. Paul B. Brooks Memorial Award for outstanding contributions to the organization.

Ivan E. Parkin was born in Connecticut and received his degree from the University of Connecticut. After working in the dairy business for awhile he accepted a position at Pennsylvania State University as a member of the Dairy Extension Staff where he remained until retired as a professor emeritus.

He has been an active member of IAMFES for many years and was president in 1954-1955. The Honorary Life Award was presented to Ivan in 1965. He is at present serving as acting-Secretary-Treasurer during the remainder of Roy Fairbanks term due to Roy's illness.

Ivan and Phyllis have three children and seven grandchildren and live at Grove Beach Point, Westbrook, Connecticut.

(Notice to membership—ballots can only be mailed to paid up members as of April 15, 1969)

MERLE P. BAKER


He was born in Kozta, January 17, 1899, and came to Ames from Toledo to attend Iowa State University. He graduated from Iowa State in dairy industry in 1921 and also received his Masters (1923) and Ph.D. degrees (1931) there.

He taught at ISU for many years in the dairy industry department with major emphasis in bacteriology, retiring in 1962.

Dr. Baker had also worked for the Wisconsin Alumni Research Assn. He was a veteran of World War I.

He was given the citation award of International Association of Milk, Food and Environmental Sanitarians and also a life membership award in 1963.

An M. P. Baker Award is given each year to the Iowa Outstanding Sanitarian.

Survivors include his wife, Gilberte; and two sisters, Mrs. F. D. Harlan, Miami, Fla., and Mrs. Lew Buescher, Kennowick, Wash.

There is a memorial set up in Dr. Baker's name through the ISU Alumni Achievement Fund and if anyone wishes, he can send gifts to the Alumni Achievement Fund designated for the M. P. Baker Memorial, 242 Memorial Union, ISU, Ames, Iowa 50010.

Merle was loved and respected by all who knew him. A great teacher, a great friend who will be missed by many, many people whose lives were influenced and enriched by association with him.

ONTARIO MILK AND FOOD SANITARIANS ANNUAL AWARDS

Mr. Herman Cauthers was the recipient of the Sanitarian of the Year Award in 1969. He was presented the Award by Mr. J. L. Baker at the Annual Meeting of the Association at the Holiday Inn, Eto-bicoke, on January 28, 1970.

Herm was born at Stayner, Ontario on December 23, 1905 and was educated in the schools of that
Herm commenced his dairy career with Besse's Dairy Products of Stayner in 1929.

In 1940 Herm attended the Dairy School at the Ontario Agricultural College and from there went to work for the late Mackenzie Robertson of Belleville Creameries Ltd., most of his time with the Robertson organization was spent in the Napanee plant after which he went to Trenton Dairies Ltd., Trenton, Ont.

In the spring of 1942 Herm joined the staff of Coburg City Dairy Ltd., Coburg, Ont., as buttermaker. It was there that he became interested in the fluid milk branch of the industry and acquired some of his practical knowledge in this field.

In 1945 he went to Lakeview Dairy Ltd., in Barrie as superintendent and fieldman. Through these positions he contributed much to this progressive dairy.

There are few people in the Canadian Dairy Industry with a keener interest in the technical problems of the industry and Herm has shared his knowledge and enthusiasm through wide participation in association affairs. Herm was the founder and first president of the Central Ontario Dairy Club. He has been a director and president of the Ontario Milk & Food Sanitarians Association. He is a long time member of the International Association of Milk, Food & Environmental Sanitarians Inc., The National Association of Dairy Plant Fieldmen and the Vermont Dairy Industry and Managers Association. Herm has been a member of the Quality Committee of O.M.D.A.

At the present time Herm represents the Ontario Milk & Food Sanitarians Association as an ex-officio member of the Board of Directors and Ambassador at large, representing our Association before all similar groups. He was honored by the Association in 1968 by receiving a life-membership.

Herm has always been keenly interested in music, has been President of the Barrie City Band, Past-President of the Huronia Federation of Bands. Herm was a member of the Orillia Kiltie Band.

Herm graduated from the Dale Carnegie Institute in 1951 and has served as an instructor in public speaking. He is a member of the First Baptist Church, Barrie.

Herm's enthusiasm for the production of quality food carries him far beyond the call of duty and he is recognized as one of our finest authorities in solving problems in the field in all areas of milk production and distribution.

In January, 1963, he joined the staff of the Lazurus Laboratories Division of the West Chemical Company. This move has been good for him, but it has also been good for the Dairy Industry and his fellow members of O.M.F.S.A. as his dedication to quality has been given full scope, this coupled with his friendly good nature, capacity for hard work, thorough knowledge of the industry has made Herm Cauthers a most deserving recipient for the 1969 Sanitarian of the Year Award.
COURSE ANNOUNCEMENT BY NATIONAL COMMUNICABLE DISEASE CENTER

The National Communicable Disease Center announces Course 3230-G, "Communicable Disease Control in the Community," to be presented at the Center in Atlanta, Georgia, May 18-22, 1970.

This course will review the elements of public health administration, establish their relationship to communicable disease control, and provide epidemiological information about certain communicable diseases. This content, combined with the selected methods of instruction, will enhance the participant's ability to discern health indicators in data from his own community. It will also improve the participant's understanding and application of the administrative and epidemiological procedures that have been demonstrated to be effective in controlling communicable diseases.

This course is intended for all public health personnel administratively responsible for, or directly engaged in, activities for the control of communicable diseases.

For further information about the course, write: National Communicable Disease Center, Attention: Director, Training Program, Atlanta, Georgia 30333.

NEW FARM METHODS COMMITTEE FOR NEW YORK STATE AND NEW NORTHEAST DAIRY PRACTICES COMMITTEE

New York State has a new Farm Methods Committee. This committee is the result of the New York State Association of Milk and Food Sanitarians' Executive Board decision to split the Farm Practices Committee into two groups, namely the Farm Methods Committee to deal with in-state problems and the Northeast Farm Practices Committee to deal with northeastern problems. The former New York State Farm Practices Committee functioned primarily as a northeastern organization. Most of the time this group met outside of New York State and dealt primarily with northeastern dairy farm sanitation problems.

The Northeast Farm Practices Committee has been relinquished by the New York Association and has now become an independent committee which will have its own constitution and bylaws and will broaden its scope from farm topics to all dairy industry sanitation and related topics. Its new title is the Northeast Dairy Practices Committee and its next and hopefully final organizational meeting will be held April 21 and 22 at the Old Newgate Coon Club, Norfolk, Conn. All are welcome to attend. The new Farm Methods Committee met early this year and immediately formed four subcommittees to deal with the following topics: (1) Abnormal Milk, (2) Bulk Tanks, (3) Milk Sampling and Testing, and (4) Uniform Interpretations and Enforcement of Codes.

TEXAS A&M FOOD PLANT MANAGEMENT CLASS SHOWS INCREASE IN ENROLLMENT

The Food Plant Management Class is composed of junior and senior students majoring in either Dairy Manufacturing or Food Technology. This year represents the largest enrollment to date of this class and shows the progress which is being made at Texas A&M to improve the quality and quantity of personnel entering the dairy and food industry.

The course is based upon a seminar approach towards industry management problems. Top management personnel from many Dairy and Food oriented companies come to A&M to present their views and company policies to the class. Through this approach, the students receive interesting and applicable facts concerning the industry which will be beneficial to them in the future. Dr. H. E. Randolph is in charge of this course.

TEXAS A&M FOOD PLANT MANAGEMENT CLASS SHOWS INCREASE IN ENROLLMENT

This year's Food Management class at Texas A&M University is shown in the above photograph.

Identified from left to right: Seated: Konnie Hall, David Klavens, Al Diorio, Robert Aragona, Rafael Vargas, Michael Worontzoff, Dan Lewis, and Jack Burns. Standing: John Robinson, James Becklet, Jim Roffan, Collier Watson, Harold Luedecke, Larry Lane, Wilfredo Moscoso, Philip Tybor, Delagado Francisco, Henry Nichols, Adam A. Mendez, Ronnie Shaw, James Head.
ATTENTION STUDENTS

JOURNAL AVAILABLE TO YOU AT SPECIAL RATE

Effective with volume 33 (begins with January, 1970 issue), the Journal of Milk and Food Technology will be available to full-time undergraduate and graduate students at a special rate of $4.00 per year.

To qualify you must:

- Be enrolled as a full-time undergraduate or graduate student at a junior college, college, or university. Full-time students enrolled in non-degree programs at colleges or technical institutes also are eligible.
- Pay $4.00 per year.
- Include with your payment a statement from your advisor, major-professor, or departmental chairman certifying that you are a full-time student.
- Payment and statement should be sent before March 15, 1970 to:
  
  MR. H. L. THOMASSON  
  Executive Secretary  
  International Association of Milk, Food, and Environmental Sanitarians, Inc.  
  Box 437  
  Shelbyville, Indiana 46176

(Faculty, please note: You can help expedite this program by: (a) Bringing it to the attention of students and (b) Designating one person in each department who will collect all monies, certify all students, and submit form and funds to the above address.)

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REVISED 1966 EDITION

Procedure for
The Investigation
of
Foodborne Disease Outbreaks

Recommended By

INTERNATIONAL ASSOCIATION OF MILK, FOOD AND ENVIRONMENTAL SANITARIANS, INC.

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International Association of Milk, Food and Environmental Sanitarians, Inc.  
Box 437, Shelbyville, Indiana

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25-100 copies, 75 cents each. Please do not send stamps.
Cleanliness is a must on a dairy farm. For this the dairyman needs an adequate supply of good, clean water. He can’t take water for granted if he wants to produce milk that will meet the quality standards of the dairy industry.

Statistics show that almost all home or farm water has some deficiencies. For example, hard water containing calcium and magnesium affects 85% of the U.S. and Canadian water supply. In addition to these minerals, there are thousands of other water problems in the country today.

The minerals calcium and magnesium in water cause it to be hard. The greater the concentration of these minerals the harder the water. They combine with soaps to form scum and with milk residue in milking machines and bulk tanks to form milkstone deposits. These deposits in the pipeline offer a breeding ground for bacteria. Also, hard water can cloud and deposit mineral film on equipment surfaces, causing stains which are difficult to remove. Hard water greatly affects the cleaning efficiency of detergents, causing increased usage and higher costs.

Dairymen know that iron in the water causes rust and results in a metallic taste. Manganese is another metallic element responsible for similar water problems. Both of these elements must be held in suspension or eliminated to obtain proper balance of water and detergents for efficient cleaning.

Many dairymen do not know the condition of their water. It may look clean, but unless the water supply is analyzed, the problems it causes in daily clean-up cannot be properly dealt with.

The Surge dealer is equipped with a water analyzer kit that enables him to make most of the primary checks on water quality. He can give on-the-spot answers on hardness, iron content, chloride content, sulfate content, and information on Calcium, Manganese, Chlorine and PH. He can detect mineral, protein and fat deposits which are not apparent to the naked eye, with special “black-light” detection equipment.

Bacteriological readings on water require a more complete laboratory testing. Knowing specific water problems is necessary before the dairyman can properly deal with them. One prime solution to consider is the use of a Water Conditioner. It will filter, refine and soften the water supply in one operation.

As a sanitarian, you know the value of clean water. Surge has equipment especially designed to assist dairymen with this problem. Let’s work together to serve dairy farmers better.

SURGE...the accent is on YOU

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