

REPORT OF PROGRESS¹—

STANDARD METHODS FOR THE EXAMINATION OF DAIRY PRODUCTS, 12TH EDITION

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In 1905 Professor S. C. Prescott of the Massachusetts Institute of Technology reported on "The Need for Uniform Methods in the Sanitary Examination of Milk" at a meeting of the laboratory section of the American Public Health Association in Boston. In his presentation he mentioned that differences in composition of the culture medium employed, variations in methods, amount of dilution, temperature and duration of incubation, as well as other minor differences, all tended to produce results which were valueless for comparison. At his suggestion a committee was appointed to study the various methods used for the bacteriological examination of milk and to recommend a uniform procedure. The first report of the Committee on Standard Methods of Bacterial Milk Analysis was published by the American Public Health Association in 1910.

In subsequent editions of "Standard Methods" a committee sought and obtained the cooperation of committees of other associations interested in the sanitary control of milk. Cooperating agencies included the American Dairy Science Association, The International Association of Dairy and Milk Inspectors, The Society of American Bacteriologists and the American Association of Medical Milk Commissions.

The 7th edition, published in 1939 with Robert S. Breed as Chairman, was called *Standard Methods for the Examination of Dairy Products*. In recent years the preparation of "Standard Methods" has been under the general supervision of the Coordinating Committee on Laboratory Methods which in turn is under the Committee on Evaluation and Standards of the American Public Health Association.

The 12th edition of *Standard Methods for the Examination of Dairy Products* which hopefully will appear in 1966 has been the work of many individuals representing numerous organizations. In the early planning stages definite attempts were made to have geographical representation as well as quality control people from industry, representatives of regulatory agencies and research-oriented university personnel on each sub-committee. This was not always possible, unfortunately, because of interest and com-

mitments of some who were requested to serve. The majority of people have been most conscientious in the detailed and time-consuming requirements of committee membership. A large number of the sub-committee chairmen as well as the membership of the committees are also members of the International Association of Milk, Food and Environmental Sanitarians. Those who have had the main responsibility for editing the manuscripts and compiling the various materials involved are most appreciative of the cooperation and aid of members of this association.

BASIC PHILOSOPHY

The basic philosophy regarding the preparation of this new edition was outlined at the start along these lines: "No new method or modification of an old method should be introduced unless it has undergone careful comparative testing in several laboratories, with the data available to the committee and to any other interested parties, preferably by publication in a recognized scientific journal. Notice of intention to include or modify should appear in print in several places with enough time to present evidence for or against to be submitted by any interested party with recommendations." Several outstanding examples in which these policies were followed will be found in the case of approving the use of plastic pipettes for the agar plate count, the selection of 32 C incubation temperature for the agar plate method, the shortening of the incubation period for standard plate counts on dry milk from 72 to 48 hours, and the inclusion of the dialysis phosphatase test.

On the basis of numerous recommendations a definite attempt was made to prepare chapters in a more concise manner than previously with the anticipation that the book would be more useful for the laboratory worker. In addition much of the interpretive material has been eliminated since it was felt that this is not the responsibility of "Standard Methods" but of regulatory agencies. In most instances references at the end of chapters have been carefully scrutinized with many being deleted and other more pertinent and up-to-date citations added.

It is likely that many laboratory workers interested in "Standard Methods" are more aware of changes and have had more to say regarding changes than

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in any previous edition. In the last three years special sessions have been held at annual meetings of the American Public Health Association, American Dairy Science Association, American Society for Microbiology and The International Association of Milk, Food and Environmental Sanitarians. In addition there have been several publications in different journals relating to proposed changes as in the case of the 32 C versus 35 C incubation for the agar plate method and the 48 versus 72 hr incubation for dry milk plates.

SPECIFIC CHANGES IN THE 12TH EDITION

Chapter 1, Quality Tests, presents some of the guiding principles followed in this edition. In an attempt to recognize a standard or reference test as a basis for official control actions, some procedures have been placed in an appendix. These latter methods often have considerable merit for control of dairy products because of simplicity, speed, cost or other advantages over the reference procedure. The appendix serves in some instances for "phasing in or out" a technic from the standard method or reference category.

In past editions many chemical procedures have been reprinted from publications of the *Association of Official Analytical Chemists (AOAC)*, but in the 12th edition many of these chemical tests are cited by reference. In other cases, chemical procedures which have not already been approved by AOAC are incorporated in "Standard Methods" because of their particular applicability to a situation or product.

In the 11th edition, Chapter 8 was entitled Detection of Pathogens and numerous methods were given for isolating and identifying specific pathogens found in milk and dairy products. In the present edition this material is covered in Chapter 2 entitled Significant Pathogens in Dairy Products. No methods for the detection of pathogens are included but a broad presentation of those organisms which may have milk as a vehicle is given. Although all will not agree, this has considerable merit because in many instances there is no standard method of detection of a specific pathogen. Numerous references are presented which will enable the laboratory worker to find suggested technics that may be of assistance.

Chapter 3, Collection of Milk and Cream Samples, has been completely rewritten and condensed to give greater emphasis in procedures involved in bulk tank samples and plant line samples. General instructions are included which has eliminated repetition of some of this material in subsequent chapters.

The authors of Chapter 4, Agar Plate Method, have prepared this material in a very useable form

for the laboratory analyst. Plastic pipettes meeting certain specifications are permitted as well as plastic petri dishes. Incubation of plates is specified at $32\text{ C} \pm 1\text{ C}$ rather than 32 or 35 C. Certain sections relating to preparation of media, and tests for growth inhibition or stimulation have been placed in an appendix. These changes make for greater facility in performing the various steps in the agar plate method.

In so far as possible attempts have been made to designate one method as the standard method. This was not possible in all cases as illustrated in Chapter 5 on Coliform Bacteria. Use of both solid and liquid media have been included with incubation at 32 C.

Chapter 6 concerns Thermoduric, Thermophilic and Psychrophilic Bacteria. In determining numbers of psychrophilic bacteria a temperature of $7\text{ C} \pm 1\text{ C}$ for ten days has been adopted rather than the previous 5-7 C for 7-10 days. The Oval Tube or Bottle Culture Method previously included in this chapter has been transferred to Chapter 19 and the Storage Quality Test to the appendix.

Chapter 7 is new and entitled Detection of Inhibitor Substances in Milk. This chapter outlines the procedure for the Disk Assay Method to detect inhibitory substances in milk.

Chapter 8 concerns Microbiological Methods for Concentrated Milk and Dry Milk but excludes cultured milks which are now relegated to another chapter. Only the Levowitz-Weber stain is acceptable for direct microscopic counts of dry milk and only clump counts are to be reported. A standard plate count of dry milk is to be made at 32 C for 48 hr ± 3 hr instead of the 72 hr as required in the previous edition.

Chapter 9 concerns Microbiological Methods for Butter and in this edition the phosphatase test, Burri slant method and mold mycelia determinations are deleted. The incubation of plates for yeast and mold counts now is $23\text{ C} \pm 2\text{ C}$ instead of 21 or 25 C as in the previous edition. Likewise the incubation of plates for proteolytic counts has been changed from 21 to $23\text{ C} \pm 2\text{ C}$. Also such plates for proteolytic counts on butter are to be flooded with 1% HCl or 10% acetic acid before counting.

Chapter 10 on the Microbiological Methods for Cheese now includes other cultured products. The membrane filter technic has been deleted for these products.

Chapter 11 now combines material on ingredients of ice cream and related products and ice cream and related frozen products into the one chapter entitled Microbiological Methods for Ice Cream and Related Frozen Products. This arrangement eliminates much duplication of sampling equipment, care of and preparation of samples, plating, incubating, and count-

ing of plates. Chemical tests involved are cross-referenced to another chapter. Methods for sweetening agents are referenced to AOAC and egg and egg products procedures are referenced to the APHA, *Recommended Methods for the Microbiological Examination of Foods*, 2nd edition.

The next section considers chapters not directly related to microbiological plating methods. Chapter 12 on the Direct Microscopic Method permits only one stain (Levowitz-Weber). The use of the loop for making smears has now been relegated to the appendix as a screening method. Plates I and II concerning microscopic appearance of raw and pasteurized milk have been deleted since they are no longer as useful as in earlier editions. Limitations have been included relative to acceptability of certain types of microscope lamps. The use of microscopes with factors of less than 500,000 and with eyepieces having less than 10 X magnification are no longer acceptable.

In the Reduction Methods described in Chapter 13 the incubation temperature is now stipulated as 36 ± 1 C. Also both the "triple reading" and "one hour" resazurin reduction tests are included.

Chapter 14 includes Microbiological Tests for Equipment, Water and Air. This chapter combines methods previously included in two different chapters, but omits procedures for surface agar counts of bottle caps, hoods and gaskets. The disintegration method for paper materials and standards for these have been omitted. The basic container rinse method is expanded to include flexible wall containers, and the basic large equipment rinse methods include CIP equipment.

The "swab contact method" now permits only insoluble cotton swabs and the use of soluble swabs is indicated in an appendix.

General methods for the microbiological examination of water supplies and of membrane filter procedures are omitted from the present edition and are referenced to the 1965 edition of *Standard Methods for the Examination of Water and Wastewater*.

Chapter 15 concerns Sediment in Fluid Milk. The preparation of sediment standard disks is referenced rather than giving detailed directions (11th edition of APHA "Standard Methods" and 10th edition of AOAC).

New photographic grading charts prepared by USDA in cooperation with FDA and APHA are recommended.

Provision is made for two methods of grading (*a*) to the nearest standard disk of those previously available, (*b*) above or below a particular standard disk of the new USDA charts. An increase in temperature for filtering of mixed milk samples to 32.2-37.8 C is required.

In Chapter 16 relating to Phosphatase Methods to Determine Pasteurization, it was originally hoped that this could be limited to one method. The Sanders-Sager, Gilcreas-Davis, and Scharer lab methods have been deleted. The Scharer rapid method was retained from the previous edition but the modified Scharer, the Cornell (1 hr test) and the dialysis test (Kosikowski) methods were also added. This chapter has been rewritten and reorganized to provide for methods for each product. Also included is the method for detection of raw admixed with heated milk.

Chapter 17 is entitled Miscellaneous Chemical Methods. Much of the material from the previous edition has been retained and full recognition of the chemical methods published in *Official Methods of Analysis* of the AOAC is acknowledged and referenced.

Chapter 18 on Radionuclides in Milk is a new chapter and includes a modified method suitable for routine monitoring for determination of four radionuclides: total radiostrontium, strontium-90, barium-140, and cesium-137 in ash from one liter of milk. Also a simplified method is given for the determination of iodine-131.

Chapter 19 concerns Simplified Technics for Viable Counts in Raw Milk. This is a new chapter and includes (*a*) the oval tube or bottle culture method, (*b*) the plate loop method, and (*c*) the roll tube method. These are considered standard methods because of their rather widespread use but are grouped at this point because of their specific use for raw milk.

APPENDICES

Extensive use has been made in this edition of appendices.

Appendix A includes material taken from various chapters of the previous edition and combined under the heading Culture Media and Preparation. Because of the increased interest both by manufacturers and users of laboratory media two different methods for productivity tests for standard methods agar are given. It is anticipated that in the future additional media will be checked for productivity by these or modified technics.

Appendix B has various Miscellaneous Microbiological Control Methods which previously were included in different chapters. For example, several screening tests for TTC reduction, reverse-phase disc assay, large equipment, air sedimentation, and psychrophilic microorganisms in water supplies are discussed. Also appearing in this appendix are suggestions for cleaning glassware and the microbiological testing for growth inhibition, preparation of buf-

ferred distilled water, the microbiological testing for toxicity and the testing for distilled water suitability.

Appendix C describes Chemical Auxiliary or Screening Methods such as the Gerber method for fat and frozen dessert and the Babcock method for fat in homogenized milk. A screening test is also given for pesticide residues in milk.

Appendix D describes Screening Technics for the Detection of Abnormal Milk. These include the modified Whiteside test, the California mastitis test, the catalase test, the Wisconsin mastitis test, and the Feulgen-DNA measurements of total somatic cells. Possibly others should have been included but the editorial decision was to limit the number of tests since this was the first edition in which such technics have appeared.

ACKNOWLEDGMENTS

Although it may seem that many changes have been made in the 12th edition actually a great deal of material from the previous edition has been re-

tained. Since the 7th edition in 1939, outstanding chairmen of this publication such as Drs. Breed, Robertson, and Black and their committees have made many contributions to the progress of standard procedures for the examination of dairy products. Each group has made changes and deletions in line with laboratory findings and the demands of the dairy field. Public health as well as industrial and regulatory interests have been considered and decisions made, not always popular with everyone, for the good of the public. It is hoped that the vast amount of time and effort expended by the 48 subcommittee members as well as many other interested individuals has been as thorough and will prove to be as worthwhile as the contributions of our predecessors. To those of you of this Association who have cooperated in many ways please accept my personal thanks as well as those of the American Public Health Association and those who will use the 12th edition of *Standard Methods for the Examination of Dairy Products*.

SALMONELLOSIS CONTROL URGED BY AVMA COUNCIL

Because salmonellosis, a form of food poisoning, now affects more people—an estimated 2 million yearly in the United States—than any other disease, there is need for concerted action leading to its control and prevention by officials of industry, agriculture, and public health agencies.

This is the principal point made in two reports prepared by veterinarians in the U. S. Departments of Agriculture and Health, Education and Welfare for the Council on Public Health and Regulatory Medicine of the American Veterinary Medical Association. The reports state that "industry and government need to give increasing, continuous emphasis to the prevention of contamination and recontamination of feed and feed ingredients in rendering plants, feed mills, and on-the-farm utilization and storage of feeds."

Salmonellosis varies in severity from a mild infection to a serious ailment which may even be fatal to the very young or elderly. It has been estimated that one percent of the population of the United States becomes infected each year. Studies of fresh poultry in retail markets have revealed that 42 percent of the samples examined were contaminated with salmonellae.

Though all warm-blooded animals, including man, and many cold-blooded ones are potential harborers of salmonella organisms, the most common "home-sites" of the infection are dogs, cats, chicks, ducklings, parakeets, canaries, and most recently, turtles. Not to be overlooked, however, according to the reports are contaminated farms, vehicles, animal concentration points, and slaughter and processing plants. Also, infected persons transmit the disease to livestock, poultry,

and other persons. The infection sometimes is spread further by contaminated animal feed and protein supplements made from animals and animal parts.

Authorities agree that the heat of rendering at rendering plants kills salmonellae but, they warn, recontamination is possible after processing especially during storage or in transport. Therefore, rigid sanitation in the production and handling of processed products is essential.

Since modern agricultural methods make for a greater number of animals being produced on fewer, highly specialized farms, the reports strongly recommend increased attention by producers to strict sanitation practices. The centralization of food processing and the speed and ease of widespread food distribution are other factors aiding in the increased dissemination of salmonella-contaminated food and food products.

To combat the problem, the veterinarians recommend that "control activities be directed toward eliminating salmonella contamination of animal feedstuffs, applying terminal pasteurization or other bactericidal treatment to human foods and food ingredients, developing food manufacturing and distribution methods to prevent salmonella contamination and bacterial growth, and training food handlers and food processors in the principles of strict sanitary measures, including personal hygiene."

The veterinary practitioner especially is advised to stress the importance of sound disease prevention and sanitation practices in the management of livestock and poultry and in the housing and care of pets.