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Interim Australian Standard®

Food microbiology

Method 2.15: Examination for specific organisms—*Listeria monocytogenes* in dairy products

STANDARDS AUSTRALIA

This Australian Standard was prepared by Committee FT/4, Food Microbiology. It was approved on behalf of the Council of Standards Australia on 6 March 1991 and published on 9 August 1991.

The following interests are represented on Committee FT/4:

Australian Government Analytical Laboratories
Australian Institute of Food Science and Technology
Australian Poultry Industries Association
Confederation of Australian Industry
Council of Australian Food Technology Associations
CSIRO Division of Food Processing
Dairy Industry Association of Australia
Department of Agriculture and Rural Affairs, Vic.
Department of Defence
Department of Health, N.S.W.
Department of Health, Qld
Department of Health, Tas.
Department of Primary Industries and Energy
Department of Primary Industries, Qld
National Association of Testing Authorities, Australia
National Health and Medical Research Council
New South Wales Dairy Corporation
Queensland Agricultural College
Royal Melbourne Institute of Technology
University of Melbourne

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PREFACE

This Interim Standard was prepared by the Standards Australia Committee on Food Microbiology in response to a request from the Department of Primary Industries and Energy for a standard method for determining whether *Listeria monocytogenes* is present in dairy products. It is technically identical with and has the same format as ISO document TC 34/SC5 N307 *Milk and milk products—Detection of Listeria monocytogenes*, which has also been adopted as a Provisional IDF Standard.

The method set out in ISO document TC 34/SC5 N307 was developed by the Joint IDF/ISO/AOAC Group of Experts E64—Detection and enumeration of *Listeria monocytogenes*—(Chairman Prof. G. Terplan, Germany, F.R.). The method is based on the FDA and USDA methods. An earlier version of the method had already been circulated as IDF Questionnaire 2389/E to IDF National Committees before ISO representatives were nominated to the joint group. (The National Committees of the following countries made comments: Australia, Austria, Belgium, Bulgaria, Canada, Czechoslovakia, Denmark, Finland, France, Germany, F.R., Ireland, Japan, Netherlands, Norway, South Africa, Spain, Sweden, UK, USA). Two collaborative studies have been carried out with the method, the second being the definitive one. The results of the second collaborative study will be published in the scientific literature. On the basis of the second collaborative study the method in IDF Questionnaire 2389/E was modified and issued in document ISO TC 34/SC5 N307.

Because methodology for examining dairy products for *Listeria* spp. is a fairly recent development and is subject to ongoing work at the international level, the Standards Australia Committee on Food Microbiology decided to adopt the method being developed by Joint Group of Experts E64 as an Interim Australian Standard. The method is used in the laboratories of some members of the Standards Australia Committee and although there has been discussion on ways to improve the method, adoption of the IDF/ISO/AOAC method has the advantage that, as the method is subjected to further trials and critical appraisal at the international level, the feedback from this work will apply directly to the Interim Australian Standard.

For the purposes of this Australian Standard, the ISO text should be modified as follows:

Reference to ISO or IDF Standard		*Australian Standard*	
ISO		AS	
7218	Microbiology—General instructions for microbiological examinations	—	
IDF			
50B	Milk and milk products—Methods of sampling	1166	Methods for sampling milk and milk products
122A	Milk and milk products—Preparation of test samples and dilutions for microbiological examination	1095	Microbiological methods for the dairy industry
		or	
		1766	Food microbiology

Standards Australia invites comment on this Interim Australian Standard from persons and organizations concerned with this subject. The date of expiry for comment is 1 August 1993, at which time this Interim Australian Standard will either be withdrawn or revised in the light of public comment, with the view to the preparation of an Australian Standard.

During the life of this document the Committee will monitor all comment or field data as it is received.

Attention is drawn to the fact that this document is an Interim Australian Standard and should be regarded as a developmental Standard and liable to future alteration.

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1 SCOPE AND FIELD OF APPLICATION This Standard specifies procedures recommended for the detection of *Listeria monocytogenes* in milk and milk products.

2 REFERENCES

ISO 7218 Microbiology—General instructions for microbiological examinations

IDF Standard 50B:1985—Milk and milk products—Methods of sampling
IDF Standard 122A:1988—Milk and milk products—Preparation of test samples and dilutions for microbiological examination.

3 DEFINITIONS For the purpose of this recommended method the following definitions apply.

3.1 *Listeria* spp. Microorganisms which form typical colonies on a solid selective medium and which display the morphological, physiological and biochemical characteristics described when tests are carried out in accordance with this Standard.

3.1.1 *Listeria monocytogenes* A *Listeria* species which is considered as pathogenic and which can be differentiated from other non-pathogenic species occurring in milk and milk products by specific biochemical characteristics.

3.2 Detection of *Listeria monocytogenes* Determination of the presence or absence of this micro-organism in a specified mass or volume, when tests are carried out in accordance with this Standard.

4 PRINCIPLE In general, the detection of *Listeria* spp. necessitates at least three successive stages as in 4.1 to 4.3. See also the diagram of procedure in Annex I.

4.1 Enrichment in selective liquid medium The selective medium is inoculated with the test portion of the sample and incubated at 30°C for 48 h.

4.2 Isolation and presumptive identification The diagnostic isolation medium is inoculated from the culture obtained in the enrichment medium (4.1), incubated at 37°C and examined after 48 h to check for the presence of colonies which, from their appearance, are considered to be presumptive *Listeria* spp.

4.3 Confirmation of identity Colonies of presumptive *Listeria* spp. (4.2) are sub-cultured onto a non-selective solid medium for confirmation of identity by means of appropriate morphological, physiological and biochemical tests.

5 CULTURE MEDIA AND REAGENTS

5.1 Basic materials In order to improve the reproducibility of the results, it is recommended that, for the preparation of the culture media, dehydrated basic components or complete dehydrated media are used. The manufacturer's instructions shall be rigorously followed.

The chemical products used for the preparation of the culture media and reagents shall be of recognized analytical quality.

The water used shall be distilled or deionized water, free from substances that might inhibit the growth of microorganisms under the test conditions.

When agar is specified, the amount used should be varied according to the manufacturer's instructions to give media of suitable firmness.

Measurements of pH shall be made using a pH meter, measurements being referred to a temperature of 25°C. Adjustments, which may not always be necessary, are made by adding either 1 mol/L hydrochloric acid or 1 mol/L sodium hydroxide solution.

If the prepared culture media and reagents are not used immediately, they shall, unless otherwise stated, be stored in the dark at a temperature between 2°C and 5°C for no longer than 1 month, conditions which do not produce any change in their composition.