

TECHNICAL
SPECIFICATION

ISO/TS
22964

IDF/RM
210

First edition
2006-02-01

**Milk and milk products — Detection
of *Enterobacter sakazakii***

Lait et produits laitiers — Détection de l'Enterobacter sakazakii



Reference numbers
ISO/TS 22964:2006(E)
IDF/RM 210:2006(E)

© ISO and IDF 2006

PDF disclaimer

This PDF file may contain embedded typefaces. In accordance with Adobe's licensing policy, this file may be printed or viewed but shall not be edited unless the typefaces which are embedded are licensed to and installed on the computer performing the editing. In downloading this file, parties accept therein the responsibility of not infringing Adobe's licensing policy. Neither the ISO Central Secretariat nor the IDF accepts any liability in this area.

Adobe is a trademark of Adobe Systems Incorporated.

Details of the software products used to create this PDF file can be found in the General Info relative to the file; the PDF-creation parameters were optimized for printing. Every care has been taken to ensure that the file is suitable for use by ISO member bodies and IDF national committees. In the unlikely event that a problem relating to it is found, please inform the ISO Central Secretariat at the address given below.

This document is a preview generated by EVS

© ISO and IDF 2006

All rights reserved. Unless otherwise specified, no part of this publication may be reproduced or utilized in any form or by any means, electronic or mechanical, including photocopying and microfilm, without permission in writing from either ISO or IDF at the respective address below.

ISO copyright office
Case postale 56 • CH-1211 Geneva 20
Tel. + 41 22 749 01 11
Fax + 41 22 749 09 47
E-mail copyright@iso.org
Web www.iso.org

International Dairy Federation
Diamant Building • Boulevard Auguste Reyers 80 • B-1030 Brussels
Tel. + 32 2 733 98 88
Fax + 32 2 733 04 13
E-mail info@fil-idf.org
Web www.fil-idf.org

Published in Switzerland

Contents

Page

Foreword.....	iv
1 Scope	1
2 Normative references	1
3 Terms and definitions	1
4 Principle (see also annex A).....	1
4.1 Pre-enrichment in non-selective liquid medium	1
4.2 Enrichment in selective liquid medium	1
4.3 Plating out and identification	2
4.4 Confirmation	2
5 Culture media and reagents	2
5.1 General	2
5.2 Culture media	2
6 Apparatus and glassware	7
7 Sampling	8
8 Preparation of test sample	8
9 Procedure (see the scheme in Annex A).....	8
9.1 Test portion	8
9.2 Pre-enrichment	8
9.3 Selective enrichment	8
9.4 Isolation of presumptive <i>Enterobacter sakazakii</i>	8
9.5 Confirmation	9
9.6 Interpretation of the results of the confirmation tests	10
10 Control cultures	11
11 Expression of results	11
12 Test report	11
Annex A (informative) Method flow scheme	12
Bibliography	13

Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

International Standards are drafted in accordance with the rules given in the ISO/IEC Directives, Part 2.

The main task of technical committees is to prepare International Standards. Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

In other circumstances, particularly when there is an urgent market requirement for such documents, a technical committee may decide to publish other types of normative document:

- an ISO Publicly Available Specification (ISO/PAS) represents an agreement between technical experts in an ISO working group and is accepted for publication if it is approved by more than 50 % of the members of the parent committee casting a vote;
- an ISO Technical Specification (ISO/TS) represents an agreement between the members of a technical committee and is accepted for publication if it is approved by 2/3 of the members of the committee casting a vote.

An ISO/PAS or ISO/TS is reviewed after three years in order to decide whether it will be confirmed for a further three years, revised to become an International Standard, or withdrawn. If the ISO/PAS or ISO/TS is confirmed, it is reviewed again after a further three years, at which time it must either be transformed into an International Standard or be withdrawn.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights.

ISO/TS 22964|IDF/RM 210 was prepared by Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 5, *Milk and milk products*, and the International Dairy Federation (IDF). It is being published jointly by ISO and IDF.

Foreword

IDF (the International Dairy Federation) is a worldwide federation of the dairy sector with a National Committee in every member country. Every National Committee has the right to be represented on the IDF Standing Committees carrying out the technical work. IDF collaborates with ISO in the development of standard methods of analysis and sampling for milk and milk products.

The main task of technical committees is to prepare International Standards. Draft International Standards adopted by the Action Teams and Standing Committees are circulated to the National Committees for voting. Publication as an International Standard requires approval by at least 50 % of IDF National Committees casting a vote.

In other circumstances, particularly when there is an urgent market requirement for such documents, a Standing Committee may decide to publish another type of normative document which is called by IDF: *Reviewed method*. Such a method represents an agreement between the members of a Standing Committee and is accepted for publication if it is approved by at least 50 % of the committee members casting a vote. A *Reviewed method* is equal to an ISO/PAS or ISO/TS and will, therefore, also be published jointly under ISO conditions.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. IDF shall not be held responsible for identifying any or all such patent rights.

ISO/TS 22964|IDF/RM 210 was prepared by the International Dairy Federation (IDF) and Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 5, *Milk and milk products*. It is being published jointly by IDF and ISO.

All work was carried out by the Joint ISO-IDF Action Team on *Harmonization*, of the Standing Committee on *Microbiological methods of analysis*, under the aegis of its project leaders, Mr D.J.C. van den Berg (NL) and Mr H. Joosten (CH).

This document is a preview generated by EVS

Milk and milk products — Detection of *Enterobacter sakazakii*

1 Scope

This Technical Specification specifies a method for the detection of *Enterobacter sakazakii* in milk powder and powdered infant formula.

The method is also applicable to environmental samples collected from milk powder or infant formula factories.

2 Normative references

The following referenced documents are indispensable for the application of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO 8261|IDF 122, *Milk and milk products — General guidance for the preparation of test samples, initial suspensions and decimal dilutions for microbiological examination*

ISO 7218, *Microbiology of food and animal feeding stuffs — General requirements and guidance for microbiological examinations*

3 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

3.1

presumptive *Enterobacter sakazakii*

microorganisms which form typical colonies on a chromogenic isolation agar, when tests are carried out in accordance with this Technical Specification

3.2

Enterobacter sakazakii

microorganisms which form typical colonies on a chromogenic isolation agar, form yellow colonies on tryptone soya agar and display biochemical characteristics as described, when tests are carried out in accordance with this Technical Specification

4 Principle (see also annex A)

4.1 Pre-enrichment in non-selective liquid medium

The pre-enrichment medium is inoculated with the test portion and incubated at $37\text{ °C} \pm 1\text{ °C}$ for 16 h to 20 h.

4.2 Enrichment in selective liquid medium

The selective enrichment medium is inoculated with the culture obtained in 4.1 and incubated at $44\text{ °C} \pm 0,5\text{ °C}$ for 22 h to 26 h.