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

*Microbiologie des aliments — Exigences générales et
recommandations*



Reference number
ISO 7218:2007(E)

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. The ISO Central Secretariat accepts no 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. In the unlikely event that a problem relating to it is found, please inform the Central Secretariat at the address given below.



COPYRIGHT PROTECTED DOCUMENT

© ISO 2007

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 at the address below or ISO's member body in the country of the requester.

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

Published in Switzerland

Contents

Page

Foreword.....	v
Introduction	vi
1 Scope	1
2 Normative references	1
3 Premises	2
3.1 General.....	2
3.2 Safety considerations.....	2
3.3 Laboratory design.....	2
3.4 Laboratory areas.....	3
3.5 Layout and fittings of the premises	4
3.6 Cleaning and disinfection	5
4 Staff	5
4.1 General.....	5
4.2 Competence	6
4.3 Verification of on-going staff competence.....	6
4.4 Hygiene	6
5 Apparatus and equipment	6
6 Preparation of glassware and other laboratory materials	28
6.1 Preparation	28
6.2 Sterilization/decontamination.....	28
6.3 Disposable equipment and materials	28
6.4 Storage of clean glassware and materials	28
6.5 Management of sterile glassware and materials	29
6.6 Use of decontamination and disinfection	29
6.7 Waste management	29
6.8 Washing	30
7 Preparation and sterilization of culture media	30
8 Laboratory samples.....	30
8.1 Sampling.....	30
8.2 Transport	31
8.3 Receipt	31
8.4 Storage.....	32
8.5 Test portion	32
9 Examination.....	32
9.1 Hygienic precautions during analysis	32
9.2 Preparation of initial suspension and dilutions	34
10 Enumeration	34
10.1 General.....	34
10.2 Enumeration using a solid medium	35
10.3 Calculation and expression of results obtained with solid media	37
10.4 Enumeration of yeasts and moulds	43
10.5 Enumeration using a liquid medium.....	44
11 Detection method (qualitative method)	49
11.1 General.....	49
11.2 Principle	50
11.3 Measurement of uncertainty.....	50

12	Confirmatory methods.....	50
12.1	General.....	50
12.2	Preparation of a pure culture.....	50
12.3	Gram's stain (modified Hucker technique).....	50
12.4	Use of biochemical galleries for identification.....	52
12.5	Use of nucleic probes for identification.....	52
12.6	Serological methods.....	53
13	Test report.....	53
14	Validation of microbiological methods.....	54
14.1	Validation of reference methods.....	54
14.2	Validation of alternative methods.....	54
14.3	Validation of in-house methods.....	54
15	Quality assurance of results/quality control of performance.....	54
15.1	Internal quality control.....	54
15.2	Reference strains.....	54
15.3	External quality assessment (proficiency testing).....	55
Annex A	(informative) Properties of some disinfectants.....	56
Annex B	(normative) Determination of most probable number (MPN).....	57
Bibliography	64

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.

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 7218 was prepared by Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 9, *Microbiology*, in collaboration with CEN Technical Committee CEN/TC 275, *Food analysis — Horizontal methods*.

This third edition cancels and replaces the second edition (ISO 7218:1996), which has been technically revised. It also incorporates the Amendment ISO 7218:1996/Amd.1:2001.

Introduction

When conducting microbiological examinations, it is especially important that

- only those microorganisms which are present in the samples are isolated and enumerated;
- the microorganisms do not contaminate the environment.

In order to achieve this, it is necessary to pay attention to personal hygiene and to use working techniques which ensure, as far as possible, exclusion of extraneous contamination.

Since, in this International Standard, it is possible to give only a few examples of the precautions to be taken during microbiological examinations, a thorough knowledge of the microbiological techniques and of the microorganisms involved is essential. It is important that the examinations are conducted as accurately as possible, including monitoring and recording aspects that may affect results and calculation of the number of microorganisms and the uncertainty of the results.

Ultimately, it is the responsibility of the head of the laboratory to judge whether the manipulations are safe and can be considered to be good laboratory practice.

A large number of manipulations can, for example, unintentionally lead to cross-contamination, and the analyst should always verify the accuracy of the results given by his or her technique.

In order to conduct the examinations correctly, it is necessary to take certain precautions when constructing and equipping the laboratory.

Certain precautions must be taken, not only for reasons of hygiene, but also to ensure good reproducibility of the results. It is not possible to specify all the precautions to be taken in all circumstances, but this International Standard at least provides the main measures to be taken when preparing, sterilizing, storing the media, and using the equipment.

If the guidance given in this International Standard is followed, this will also contribute towards maintaining the health and safety of personnel. Additional information on this subject is to be found in the literature listed in the Bibliography.

In order to distinguish the guidance in this International Standard, it has been printed in a different typeface (Times New Roman).

Microbiology of food and animal feeding stuffs — General requirements and guidance for microbiological examinations

1 Scope

This International Standard gives general requirements and guidance/options intended for three main uses:

- implementation of ISO/TC 34/SC 9 or ISO/TC 34/SC 5 standards for detection or enumeration of microorganisms, named hereafter “specific standards”;
- good laboratory practice for food microbiological laboratories (the purpose is not to detail them in this International Standard, manuals are available for that purpose);
- guidance for accreditation of food microbiological laboratories (this International Standard describes the technical requirements according to Annex B of ISO/IEC 17025:2005 for the accreditation of a microbiological laboratory by national organizations).

The requirements of this International Standard supersede the corresponding ones of existing specific standards.

Additional instructions in the field of molecular biology examinations are specified in ISO 22174.

This International Standard covers examination for bacteria, yeasts and moulds and can be used if supplemented with specific guidance for prions, parasites and viruses. It does not cover the examination for toxins or other metabolites (e.g. amines) from microorganisms.

This International Standard applies to the microbiology of food, animal feeding stuffs, the food production environment and the primary production environment.

The purpose of this International Standard is to help to ensure the validity of food microbiology examinations, to assist in ensuring that the general techniques used for conducting these examinations are the same in all laboratories, to help achieve homogeneous results in different laboratories, and to contribute towards the safety of the laboratory personnel by preventing risks of infection.

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 835 (all parts), *Laboratory glassware — Graduated pipettes*

ISO 6887 (all parts), *Microbiology of food and animal feeding stuffs — Preparation of test samples, initial suspension and decimal dilutions for microbiological examination*

ISO 8199, *Water quality — General guidance on the enumeration of micro-organisms by culture*

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

ISO 8655-1, *Piston-operated volumetric apparatus — Part 1: Terminology, general requirements and user recommendations*

ISO/TS 11133 (all parts), *Microbiology of food and animal feeding stuffs — Guidelines on preparation and production of culture media*

ISO 16140, *Microbiology of food and animal feeding stuffs — Protocol for the validation of alternative methods*

ISO/TS 19036, *Microbiology of food and animal feeding stuffs — Guidelines for the estimation of measurement uncertainty for quantitative determinations*

ISO 22174, *Microbiology of food and animal feeding stuffs — Polymerase chain reaction (PCR) for the detection of food-borne pathogens — General requirements and definitions*

3 Premises

3.1 General

This clause gives general requirements, e.g. the principles of design and organization, for the layout of a microbiological laboratory.

Examination of primary production stage samples (especially for sample reception and sample preparation) shall be separated from examination of other samples to reduce the risks of cross-contamination.

3.2 Safety considerations

The laboratory design shall comply with safety requirements which will depend on the type of microorganism. To this end, microorganisms are classified in four risk categories:

- **Risk category 1** (no or very low risk to the individual and to the community).

A microorganism that is unlikely to cause human or animal disease.

- **Risk category 2** (moderate risk to the individual, low risk to the community).

A pathogen that can cause human or animal disease but is unlikely to be a serious hazard to laboratory workers, the community or the environment. Laboratory exposures may cause serious human infection, but effective treatment and preventive measures are available and the risk of spread of infection is limited.

- **Risk category 3** (high risk to the individual, low risk to the community).

A pathogen that usually causes serious human or animal disease but does not ordinarily spread from one infected individual to another. Effective treatment and preventive measures are available.

- **Risk category 4** (high risk to the individual and to the community).

A pathogen that usually causes serious human or animal disease and that can be readily transmitted from one individual to another, directly or indirectly. Effective treatment and preventive measures are not usually available.

WARNING — Refer to national regulations which will define, in particular, the risk category of the microorganisms encountered within the boundaries of the country concerned.

3.3 Laboratory design

The guidelines for laboratory layout described below cover examinations for the detection of microorganisms belonging to risk category 1, 2 and 3 for food microbiology.