
**Water quality — Requirements for
the performance testing of membrane
filters used for direct enumeration of
microorganisms by culture methods**

*Qualité de l'eau — Exigences relatives aux essais de performance
des membranes filtrantes utilisées pour le dénombrement direct des
micro-organismes par des méthodes de culture*

This document is a preview generated by ELS



COPYRIGHT PROTECTED DOCUMENT

© ISO 2023

All rights reserved. Unless otherwise specified, or required in the context of its implementation, no part of this publication may be reproduced or utilized otherwise in any form or by any means, electronic or mechanical, including photocopying, or posting on the internet or an intranet, without prior written permission. Permission can be requested from either ISO at the address below or ISO's member body in the country of the requester.

ISO copyright office
CP 401 • Ch. de Blandonnet 8
CH-1214 Vernier, Geneva
Phone: +41 22 749 01 11
Email: copyright@iso.org
Website: www.iso.org

Published in Switzerland

Contents

	Page
Foreword	v
Introduction	vii
1 Scope	1
2 Normative references	1
3 Terms and definitions	1
3.1 General terminology.....	2
3.2 Terminology of performance testing.....	3
3.3 Terminology for test microorganisms.....	4
4 Principle	5
4.1 General.....	5
4.1.1 Introduction.....	5
4.1.2 Batch testing.....	6
4.1.3 Supplementary testing.....	6
4.2 Performance testing.....	7
4.2.1 Modules for batch and supplementary testing.....	7
4.2.2 Absence of microbial contamination.....	7
5 Apparatus and glassware	8
6 Culture media and diluents	8
7 Preparation of microorganisms for performance testing	8
7.1 General.....	8
7.2 Reference count.....	8
7.2.1 Quantitative productivity testing.....	8
7.2.2 Qualitative selectivity testing.....	9
7.2.3 Qualitative specificity testing.....	9
7.3 Preparation of a standardized test suspension using a working culture.....	9
7.3.1 General.....	9
7.3.2 Preparation of the working culture.....	9
7.3.3 Preparation of a standardized test suspension (inoculum) for the test.....	10
7.4 Preparation of a test suspension using reference material.....	10
8 Sampling of membrane filters for testing	10
9 Procedure	11
9.1 General.....	11
9.2 Inoculation by spread plate technique.....	11
9.2.1 General.....	11
9.2.2 Inoculation.....	12
9.3 Inoculation by membrane filtration technique.....	12
9.3.1 General.....	12
9.3.2 Inoculation.....	12
9.4 Incubation and counting.....	13
9.5 Test for absence of microbial contamination.....	13
10 Calculation, expression and interpretation of results	13
10.1 General.....	13
10.2 Productivity testing.....	13
10.3 Selectivity testing.....	14
10.4 Specificity testing.....	14
11 Documentation of test results	14
11.1 Test report.....	14
11.2 Information provided by the manufacturer.....	15
11.3 Traceability.....	15

Annex A (normative) Diagram of the procedure for batch testing	16
Annex B (informative) Example of a card for recording test results from batch testing	17
Annex C (informative) Quantitative supplementary testing of membrane filters	18
Annex D (informative) Qualitative supplementary testing of membrane filters	21
Annex E (informative) Practical example of quantitative batch testing and quantitative supplementary testing by the end user	25
Bibliography	35

document is a preview generated by EVS

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.

The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO documents should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

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. Details of any patent rights identified during the development of the document will be in the Introduction and/or on the ISO list of patent declarations received (see www.iso.org/patents).

Any trade name used in this document is information given for the convenience of users and does not constitute an endorsement.

For an explanation of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT), see www.iso.org/iso/foreword.html.

This document was prepared by Technical Committee ISO/TC 147, *Water quality*, Subcommittee SC 4, *Microbiological methods*, in collaboration with the European Committee for Standardization (CEN) Technical Committee CEN/TC 230, *Water analysis*, in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement).

This second edition cancels and replaces the first edition (ISO 7704:1985), which has been technically revised.

The main changes are as follows:

- the scope has been changed to cover the requirements for the performance testing of membrane filters used for retention and direct enumeration;
- clauses have been added for terms and definitions, microorganisms, sampling and replicates, procedure, inoculation and incubation, counting, calculation and documentation;
- the clauses referencing to culture media and diluents, test strain preparation, performance testing and procedure have been revised to align with ISO 8199 and ISO 11133;
- [Annex A](#) has been added with a diagram of the batch testing;
- [Annex B](#) has been added to give an example of a card to record the test results from batch testing and supplementary testing of membrane filters;
- [Annex C](#) has been added to describe the quantitative additional testing of membrane filters including a diagram of the procedure;
- [Annex D](#) has been added to describe the qualitative supplementary testing of membrane filters;
- [Annex E](#) has been added to give a practical example of batch testing and quantitative additional testing by the end user including a diagram of the procedure;
- the Bibliography has been added.

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.

This document is a preview generated by EVS

Introduction

In laboratories carrying out microbiological examinations, the main objectives are to either capture, resuscitate, grow, detect or enumerate, or all, a wide variety of microorganisms. Membrane filters are used in many traditional microbiological culture techniques and are commercially available in various brands and types. Many comparison studies of membrane filters which have been reported in the literature show differences in their ability to recover bacteria from water samples, see References [22], [23], [28], [30], [31], [32], [33] and [34]. The complex manufacturing process means that the chemical composition, pore size and pore structure can vary, depending on the brands, and even on the lot of material. Furthermore, the manufacturing process can also release leachables that can potentially interfere with the recovery of microorganisms.

Thus, it is very important to standardize the performance testing of membrane filters as much as possible, not only to provide consistent results, but also to enable the development of standardized procedures for enumerating specific microorganisms.

Water quality — Requirements for the performance testing of membrane filters used for direct enumeration of microorganisms by culture methods

1 Scope

This document specifies the requirements for the performance testing of membrane filters used for the retention followed by direct enumeration of microorganisms by culture methods.

This document is applicable to membrane filters which are used for retention followed by direct enumeration of specific microorganisms on solid media or on other devices containing media, like absorbent pads^[19].

This document is not applicable for membrane filters used for concentration and elution or for qualitative methods.

These tests are applicable to the membrane filters intended for the microbiological analysis of different types of water, such as:

- drinking water, bottled water and other types of water with expected low numbers of microorganisms;
- water with expected higher numbers of microorganisms, for example, surface water and process water.

These tests are intended to demonstrate the suitability of the whole system (membrane filter together with the culture medium including the filtration step) required for the specific tests described in References [3], [6], [8], [10], [12] and [13].

This document applies to:

- manufacturers producing membrane filters;
- microbiological laboratories using membrane filters for their own testing or providing these to other end users.

2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements 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 8199:2018, *Water quality — General requirements and guidance for microbiological examinations by culture*

ISO 11133:2014, *Microbiology of food, animal feed and water — Preparation, production, storage and performance testing of culture media*

ISO 11133:2014/Amd1:2018, *Microbiology of food, animal feed and water — Preparation, production, storage and performance testing of culture media*

3 Terms and definitions

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