International Standard

ISO 15883-7

Second edition
2025-03

Washer-disinfectors —

Part 7: Requirements and tests for washer-disinfectors employing chemical disinfection for non-critical thermolabile medical devices and health care equipment

Laveurs désinfecteurs —

Partie 7: Exigences et essais pour les laveurs désinfecteurs destinés à la désinfection chimique des dispositifs médicaux thermosensibles non critiques et des équipements de soins de santé

Reference number
ISO 15883-7:2025(en)

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

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 document 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).

ISO draws attention to the possibility that the implementation of this document may involve the use of (a) patent(s). ISO takes no position concerning the evidence, validity or applicability of any claimed patent rights in respect thereof. As of the date of publication of this document, ISO had not received notice of (a) patent(s) which may be required to implement this document. However, implementers are cautioned that this may not represent the latest information, which may be obtained from the patent database available at www.iso.org/patents. ISO shall not be held responsible for identifying any or all such patent rights.

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 198, *Sterilization of health care products,* in collaboration with the European Committee for Standardization (CEN) Technical Committee CEN/TC 102, *Sterilizers and associated equipment for processing of medical devices,* in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement).

This second edition cancels and replaces the first edition (ISO 15883-7:2016), which has been technically revised.

The main changes are as follows:

— deletion of 'non-invasive' from the document title and within clauses;

— incorporation of requirements of and reference to ISO 15883-1:2024 and ISO 15883-5:2021;

— revision of cross-references to relevant clauses in ISO 15883-1:2024 and ISO 15883-5:2021;

— alignment with terms and definitions in ISO 11139:2018 and ISO 11139:2018/Amd1:2024;

— update of Introduction and addition of reference to ISO/TS 5111 on water quality;

— clarification on requirement for reused process chemicals (see 4.2.4 and 6.6.2);

— Annex A changed from normative to informative;

— updated Annex C method description to align with ISO 15883-1:2024 and ISO 15883-4:2018;

— revision of normative references and bibliographic references.

A list of all parts in the ISO 15883 series can be found on the ISO website.

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.

Introduction

This document is the seventh part of the ISO 15883 series of standards specifying the performance of washer-disinfectors (WD) and the general requirements for performance applicable to instrument WD. The requirements given in this document apply to WD used for cleaning and chemical disinfection of non-critical thermolabile medical devices and health care equipment without further treatment in health care settings. Such reusable equipment is cleaned and disinfected, but processing in a WD for surgical instruments (see ISO 15883-2), for human waste containers (see ISO 15883-3), for endoscopes (see ISO 15883-4), or for thermal disinfection of non-critical medical devices and health care equipment (see ISO 15883-6), is inappropriate and/or impractical. Examples of the equipment to which this document applies are beds and bedside furniture, trolleys and transport carts, operating tables, footwear, wheelchairs, or aids for people with disabilities.

Requirements for WD for other applications are specified in other parts of ISO 15883.

Safety requirements for WD are given in IEC 61010-2-040.

The quality of water to be used in a WD is covered in ISO/TS 5111.

NOTE Local or national regulations can apply in respect of the potential adverse effects on the quality of water intended for human consumption or environmental impacts caused by the WD and its intended use.

Washer-disinfectors —

Part 7: Requirements and tests for washer-disinfectors employing chemical disinfection for non-critical thermolabile medical devices and health care equipment

1 Scope

This document specifies the requirements for washer-disinfectors (WD) intended to be used for the cleaning and chemical disinfection, in a single operating cycle, of reusable items such as:

a) bed frames;

b) bedside tables;

c) transport carts;

d) containers;

e) surgical tables;

f) sterilization containers;

g) surgical clogs;

h) wheelchairs;

i) aids for persons with disabilities.

This document also specifies the performance requirements for the cleaning and disinfection of the WD and its components and accessories.

Devices identified within the scopes of ISO 15883-2, ISO 15883-3, ISO 15883-4, and ISO 15883-6 do not fall within the scope of this document.

In addition, this document specifies the methods for type testing, works testing, validation (installation, operation, and performance qualification on first installation), routine control, and monitoring, as well as requalifications to be carried out periodically and after essential repairs.

NOTE 1 WD covered by this document can also be used for cleaning and chemical disinfection of other thermolabile and reusable devices as recommended in the instructions for use (IFU) for those devices.

NOTE 2 The performance requirements specified in this document cannot ensure the inactivation or removal of the causative agent(s) (prion proteins) of transmissible spongiform encephalopathies.

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 11139:2018, *Sterilization of health care products — Vocabulary of terms used in sterilization and related equipment and process standards*

ISO 11139:2018/Amd1:2024, *Sterilization of health care products — Vocabulary of terms used in sterilization and related equipment and process standards — Amendment 1: Amended and additional terms and definitions*

ISO 15883-1:2024, *Washer-disinfectors — Part 1: General requirements, terms and definitions and tests*

ISO 15883-4, *Washer-disinfectors — Part 4: Requirements and tests for washer-disinfectors employing chemical disinfection for thermolabile endoscopes*

ISO 15883-5:2021, *Washer-disinfectors — Part 5: Performance requirements and test method criteria for demonstrating cleaning efficacy*

ISO 15883-6[1)], *Washer-disinfectors — Part 6: Requirements and tests for washer-disinfectors employing thermal disinfection for non-invasive, non-critical medical devices and health care equipment*

IEC 61010-2-040:2020, *Safety requirements for electrical equipment for measurement, control and laboratory use — Part 2-040: Particular requirements for sterilizers and washer-disinfectors used to treat medical materials*

EN 10088-1, *Stainless steels — Part 1: List of stainless steels*

EN 10088-2, *Stainless steels — Part 2: Technical delivery conditions for sheet/plate and strip of corrosion resisting steels for general purposes*

EN 12353:2021, *Chemical disinfectants and antiseptics — Preservation of test organisms used for the determination of bactericidal (including Legionella), mycobactericidal, sporicidal, fungicidal and virucidal (including bacteriophages) activity*

EN 13727:2012+A2:2015, *Chemical disinfectants and antiseptics — Quantitative suspension test for the evaluation of bactericidal activity in the medical area — Test method and requirements (phase 2, step 1)*

3 Terms and definitions

For the purposes of this document, the terms and definitions given in ISO 11139:2018, ISO 11139:2018/Amd1:2024, ISO 15883-1, ISO 15883-4, ISO 15883-5, ISO 15883-6 and the following apply.

ISO and IEC maintain terminology databases for use in standardization at the following addresses:

— ISO Online browsing platform: available at https://www.iso.org/obp

— IEC Electropedia: available at https://www.electropedia.org/

3.1
non-critical device
<washer-disinfector> item processed in a washer-disinfector, whose surface(s) are intended to contact intact skin of a body but do not penetrate it, or device not intended for direct patient contact

EXAMPLE Blood pressure cuffs, wheelchairs, trays, bowls, dishes, glassware, receivers, containers for transit.

Note 1 to entry: National regulations can use alternative wording for the definition of this term when applied to medical devices.

[SOURCE: ISO 11139:2018/Amd1:2024, 3.357]

1) Under revision with a modified title, *Washer-disinfectors — Part 6: Requirements and tests for washer-disinfectors employing thermal disinfection for non-critical medical devices and health care equipment*. Stage at the time of publication: ISO/DIS 15883-6:2024.

4 Performance requirements

4.1 General

4.1.1 The requirements of ISO 15883-1 apply, with the exception of:

— ISO 15883-1:2024, 4.3.1 (which refers to thermal disinfection);

— ISO 15883-1:2024, 5.9 [process temperature control limits, however, ISO 15883-1:2024 5.9 d) and f) do apply];

— ISO 15883-1:2024, 5.11 [process verification, however ISO 15883-1:2024 5.11.4 b) does apply].

4.1.2 The WD shall be designed to clean and chemically disinfect non-critical thermolabile medical devices and health care equipment.

4.1.3 When necessary, the WD shall be provided with means to facilitate the correct alignment of the load in the washing chamber.

4.1.4 The means to control the volume of the process chemical(s) admitted (see ISO 15883-1:2024, 5.7.4 and 5.7.5) shall be adjustable by means of an access device. The accuracy of the dosing system shall be at least ±10 % or as specified and tested for conformity (see 6.6).

4.1.5 The automatic controller shall verify that the final concentration of disinfectant(s) is within specified limits.

NOTE Confirmation of the concentration of disinfectant can include the measurement of the volume of disinfectant and water admitted together with a certificate of conformity from the disinfectant supplier for the concentration of the disinfectant, together with data to support the shelf life, expiry date, etc.

4.2 Cleaning

4.2.1 Cleaning shall be tested in accordance with the requirements of ISO 15883-1 using the methods described in ISO 15883-5:2021, Clause 5 that are relevant to the loads to be processed.

4.2.2 During the washing stage:

a) the washing stage starts when the temperature at the control sensor of the WD is not less than the minimum specified washing temperature;

b) the washing temperature band shall have the lower limit defined by the washing temperature and an upper limit not greater than that defined in ISO 15883-1:2024, 4.2.3.

4.2.3 Cleaning efficacy shall be determined in accordance with 6.7.

4.2.4 If the WD is designed to allow the reuse of the cleaning agent on two (2) or more consecutive operating cycles, then care shall be taken to ensure that the efficacy and safety (e.g. accumulation of foreign material, device compatibility) of the cleaning solution is not impaired. This shall include testing in accordance with 6.6.2 and at least the following considerations.

a) Specified methods shall be used to ensure that the cleaning agent has retained the required cleaning efficacy. These methods shall be based on validation studies relevant to the cleaning agent, to determine suitable parameter(s) or indicator(s)/marker(s) that can be monitored. Suitable parameters may include the concentration of the active ingredient and other ingredients that can also affect performance (e.g. pH).

NOTE Minor changes in formulation of the cleaning agent can have a significant effect on its stability, cleaning efficacy, and other aspects of performance.

b) Recommendations to the user for the maximum period or number of operating cycles for which the cleaning agent may be used. This shall be based on validated experimental data.

c) Where validated use conditions (maximum period or number of operating cycles) are exceeded, the automatic controller shall

— operate an audible and visible alarm and prevent the use of the operating cycle until the cleaning agent is changed, or

— effect an automatic change of the cleaning agent in the WD.

d) If final rinse water including processing chemicals is to be reused (recycled) for a cleaning stage, absence of negative effects using the defined reuse instructions shall be demonstrated as defined in ISO 15883-1:2024, 4.2 and 4.4.

4.3 Disinfection

4.3.1 The operating cycle shall include a chemical disinfection stage, which may be combined with the cleaning and shall be deemed to have been achieved when testing requirements in 6.8 are met.

4.3.2 The requirements and tests in this document are based on the use of aqueous disinfectant solutions.

NOTE 1 Other systems based on gaseous disinfectants are not excluded. There are equivalent tests for these systems.

The requirements and tests shall include the following considerations.

a) Disinfectant(s) to be used, for which in vitro efficacy has been demonstrated based on relevant published standards. For the purpose of efficacy testing, a validated disinfectant neutralization method shall be used at the end of the disinfection stage of the operating cycle.

NOTE 2 Demonstration that the disinfectant meets the above requirements can be made employing methods based on relevant published standards or other relevant publications, e.g. EN 13624,[5] EN 13727, EN 14476,[6] EN 14561,[7] EN 14562,[8] EN 14885,[9] AOAC Use dilution test,[10] ASTM E2197 virucidal test,[11] OECD Guideline.[12]

NOTE 3 The method(s) can be provided with the disinfectant.

b) When tested on surfaces for the minimum exposure time at the minimum concentration and the minimum temperature to be used in the WD, the disinfectant demonstrates the following:

1) at least a 5 $\log_{10}$ inactivation of vegetative bacteria;

2) at least a 4 $\log_{10}$ inactivation of yeast-like fungi;

3) at least a 4 $\log_{10}$ inactivation of enveloped viruses.

NOTE 4 National regulatory authorities can require higher inactivation values or efficacy against a wider range of microorganisms. In this case, the tests listed in Note 2 can be modified to demonstrate efficacy.

NOTE 5 Efficacy tests against vegetative bacteria can exclude mycobacteria. See also 8 g).

c) The compatibility of the cleaning and disinfection agents is indicated, including any effect on disinfection efficacy from carryover of cleaning agent.

d) The experimental conditions of tests intended to demonstrate the microbicidal efficacy of the disinfectant in vitro shall reflect the conditions of use of the disinfectant. Thus, when cleaning and disinfection are combined, the disinfectant shall be tested in the presence of applicable interfering substances that shall include soiling typically found in the loads to be processed.

4.3.3 The temperature of the process throughout the disinfection stage shall be monitored to verify that it remains within the specified limits of the disinfectant, and it shall be compatible with the temperature limits for the non-critical device(s) to be processed.

This shall be achieved either by controlling the temperature of the process, or, if the temperature in the WD is not controlled, then operation of the WD is prevented outside the specified disinfectant temperature range.

4.3.4 If the WD is designed to allow the reuse of the disinfectant on two or more consecutive operating cycles, then care shall be taken to ensure that the efficacy and safety (e.g. accumulation of foreign material, device compatibility) of the disinfectant solution is not impaired. This shall include testing in accordance with 6.6.2 and at least the following considerations.

a) The means that shall be used to ensure that the disinfectant has retained the required antimicrobial disinfection efficacy. These means shall be based on validation studies, which are normally available for the disinfectant, to determine suitable parameter(s) or indicator(s)/marker(s) that may be monitored to indicate the antimicrobial efficacy of the disinfectant. Suitable parameters include the concentration of the active ingredient and other ingredients that can also affect performance (e.g. pH, stability).

   NOTE 1 Minor changes in formulation of the disinfectant can have a significant effect on storage life, antimicrobial efficacy, and other aspects of performance.

b) Recommendations to the user for the maximum period or number of operating cycles for which the disinfectant may be used [see also Clause 8 f)]. This shall be based on validated experimental data.

c) When validated use conditions (maximum period or number of operating cycles) are exceeded, the automatic controller shall provide an audible indication, or visible indication, or both, and prevent the use of the operating cycle until the disinfectant solution is changed (manually or automatically).

d) Provide a method for the user to monitor the disinfectant using a chemical indicator or other method specific for the disinfectant to show that the disinfectant is at or above the minimum recommended concentration [see also Clause 8 h)].

NOTE 2 The minimum recommended concentration is the lowest concentration of active and other ingredients to meet the label claim of a reusable disinfectant.

4.4 Final rinsing

The water quality used for rinsing after the disinfection stage shall not impair the result of cleaning/disinfection when tested in accordance with 6.3 and ISO 15883-1:2024, 6.10.5.

NOTE 1 WHO definition for potable water, ISO/TS 5111[4] definition of potable water, or national regulatory authorities can be considered.

NOTE 2 A contamination level higher than 100 CFU/ml under the test requirements of ISO 15883-1 can represent a risk.

4.5 Self-disinfection

4.5.1 If ISO 15883-1:2024, 4.7.2 does not apply, a self-disinfection cycle shall be provided so that the WD does not become a focus for contamination of the load, and to provide a means of disinfecting the WD after interventions for maintenance, repairs, or testing (see ISO 15883-1:2024, 4.7.6).

NOTE 1 The self-disinfection process also deals with the situation where the WD has become contaminated. Biofilm can easily develop in the piping used to convey rinse water to the load and can contain microorganisms in a state in which they are highly resistant to disinfection.

Thermal disinfection shall attain a minimum A_0 of 60 and shall be capable to be set to give an A_0 value of 600.

When chemical self-disinfection is used, a disinfectant containing a disinfecting agent different from that used for disinfecting the load or the same disinfectant under different conditions (e.g. formulation, increased temperature, higher concentration, extended contact time) shall be used.

NOTE 2 The use of a disinfectant based on the same active ingredients can carry the risk of allowing organisms resistant to a particular disinfectant family to proliferate.

4.5.2 Details of the parts of the WD subjected to the self-disinfection cycle shall be provided and whether this cycle includes other components such as the water treatment equipment.

4.5.3 The WD self-disinfection cycle shall:

a) be operated under the control of the automatic controller;

b) be a user selectable cycle;

c) provide for disinfection of the chamber and all piping and tanks which come into contact with the water or solutions used for cleaning, disinfecting, and rinsing the load;

d) include means to warn the user that the WD shall be operated without any load in the chamber and, as far as practicable, include means to verify that no device is present in the WD before the cycle will operate;

e) in the case of thermal self-disinfection of the WD, verify that all the parts of the heating system and the associated pipework, via which the water or the steam reach the WD tank, attain an A_0 value of at least 60.

4.5.4 The self-disinfection cycle shall ensure that contamination through failure of the water treatment equipment can be effectively disinfected. Conformance shall be verified by testing in accordance with 6.8.3.

4.5.5 Thermal disinfection systems shall be evaluated by thermometric monitoring of the system with sensors placed at those parts of the system specified as representative of the lowest temperatures in the system. The entire system subjected to thermal disinfection shall attain the required disinfection temperature.

4.5.6 For chemical self-disinfection cycles, a microbiological test shall be performed. The capability of the WD to provide self-disinfection shall be tested in accordance with Annex B and shall conform with its test criteria.

4.6 Drying

4.6.1 The WD shall, unless otherwise specified, be provided with equipment to dry the load.

4.6.2 Drying of the load in the WD shall be deemed to have been achieved when plain surfaces of the items are visibly dry (see 6.4).

4.7 Water treatment equipment

4.7.1 General

Means shall be provided to ensure any water treatment equipment that is part of the WD (softeners, de-ionizers, filters, etc.) is operated within the limits (e.g. flow rates, supply pressures) specified for the water treatment equipment.

4.7.2 Disinfection of water treatment equipment

4.7.2.1 When the water treatment equipment is a part of the WD, and used for post-cleaning rinse, disinfection or final rinse stages, the former shall be designed and constructed to allow for periodic disinfection. Guidance on the minimum frequency for equipment disinfection shall be provided in accordance with the information supplied by the purchaser for the quality of the water supply and the water treatment equipment [see Clause 8 c) and j)].

NOTE The disinfection of the water treatment equipment can be carried out during a self-disinfection cycle.

The actual frequency should be decided by the user based upon known factors, e.g. seasonal variations in the quality of water supplied to the WD and the operational history of the water treatment equipment.

The disinfection method shall not cause any damage to, nor impair the efficacy of, the treatment equipment.

The efficacy of the water equipment disinfection procedure to provide self-disinfection shall be deemed to have been established if, when tested in accordance with 6.3, there are less than 10 colony forming units (CFU) recovered from each of two 100 ml samples and other control parameters (e.g. temperature, holding time) have been achieved.

4.7.2.2 If the water treatment equipment is not part of the WD, the requirements for water supplied to the WD shall be specified. This shall include specification of the permissible microbial contamination of the water supply [see Clause 8 j)].

NOTE 1 To meet the specification of the permissible microbial contamination of the water supply, it can be necessary for the user to make provision for disinfection of the external water treatment equipment.

Final rinse water shall have less than 10 CFU/100 ml sample (see 6.3). If required, means shall be provided to disinfect water used for the final rinse.

NOTE 2 The following methods can be suitable for control of the microbial contamination of rinse water:

— maintaining in a dedicated reservoir at a temperature not less than 65 °C for the time demonstrated to achieve disinfection of the incoming supply;

— disinfecting immediately prior to use;

— filtering to remove suspended particles of a size greater than 0,2 µm;

— sterilizing, in a closed container, with a connection to the WD designed and constructed to provide aseptic transfer.

4.7.2.3 The connection between the water supply, which has been treated to remove microbial contamination and the circulation system for rinsing, shall be designed and constructed to ensure microbiological quality.

Provision shall be made for disinfection of this connection to be made periodically. The frequency and method of carrying out this disinfection shall be specified.

4.7.2.4 When the water treatment equipment is a part of the WD, from the time before the rinsing stage until the end of the operating cycle, as appropriate, the final water treatment used to fulfil the requirements of 4.4 shall be monitored by the WD automatic controller to verify that the parameters affecting the efficacy of the equipment remain within specification.

4.7.3 Maintenance of piping

The planned preventive maintenance required for the piping that is part of the WD design and is used to convey final rinse water to the load shall be specified.

5 Mechanical requirements

5.1 Materials: design, manufacture, and assembly

5.1.1 The WD should be constructed so that loading and unloading of loads is possible without the need to enter the chamber.

5.1.2 The WD design should consider the ease of access for maintenance (IEC 61010-2-040:2020, 5.4.3 and 7.5.101).

5.2 Process verification

The WD shall be equipped with a level b) process verification system [see ISO 15883-1:2024, 5.11.4 b)].

6 Testing for conformity

6.1 General

The tests described in this clause are in addition to the tests described in ISO 15883-1 and are specific for WD intended to process non-critical thermolabile medical devices and health care equipment. The tests in this subclause are reference tests intended for use in demonstrating conformance with the specified requirements of this document. They may be used in type tests, works tests, and in validation and requalification tests, or in routine tests carried out by, or on behalf of, the user. Other tests and methods providing equivalent assurance may be used as the basis of claiming conformance with this document. In any case of dispute, the reference tests given in this document shall be used.

The summary programme of tests in addition to those given in ISO 15883-1 is shown in Annex A.

NOTE A number of the tests can be carried out simultaneously with each other, or with those required by applicable safety standards such as IEC 61010-2-040, or both.

6.2 Test load

6.2.1 Loading with standard goods

The test load shall be defined to represent typical loads to be processed.

6.2.2 Loading with special goods

If loads that were not included in the type test are to be processed, then a performance qualification shall be carried out with representative loads.

6.3 Final rinse water

Water used for final rinsing shall be collected from within the WD chamber and tested for microbial quality in accordance with Annex C.

Where the final rinse water is collected after contact with the load, a neutralisation method shall be used to eliminate any antimicrobial activity.

The test shall be carried out following installation. Testing shall be repeated weekly until it has been established that the final rinse water is consistently within specification for at least one month. Testing shall then be repeated annually thereafter.

NOTE Tests for other microorganisms that can be of clinical significance can be performed.

6.4 Load dryness

6.4.1 General

When the WD is intended to dry the load, testing shall be carried out as described below.

6.4.2 Procedure

The WD is loaded in accordance with instructions for use (see 6.2) and run through a complete operating cycle including the drying stage. After completion of the operating cycle, the door (unloading) is opened and remains open for 5 min before the load is removed from the chamber. The load dryness is inspected visually.

6.4.3 Results

Report whether or not visible surfaces of the load are free of water.

6.5 Thermometric tests

6.5.1 General

When thermometric monitoring of the process is defined, the tests shall be performed in accordance with ISO 15883-1:2024, 6.8.

6.5.2 Load temperature test

The load temperature test shall be carried out using reference loads made up of a full load of items that the WD is intended to process (see 6.2). The items chosen shall be those with the greatest mass, highest specific heat capacity, and lowest thermal conductivity.

6.6 Chemical dosing tests

6.6.1 General

The tests shall be performed in accordance with ISO 15883-1:2024, 6.9.

6.6.2 Reused process chemicals

If the WD is designed to reuse a cleaning or disinfecting agent, the agent shall be above the minimum recommended concentration before use for the last permitted operating cycle (see 4.2.4 and 4.3.4). In the case where it is intended that a disinfecting agent be reused, the efficacy of the solution shall be determined as a function of its minimum recommended concentration (MRC), and, if required, at the minimum effective concentration (MEC). The determination of chemical concentration is performed in accordance with a method given for the process chemical(s). Record whether the requirements are fulfilled.

6.7 Tests of cleaning efficacy

6.7.1 General

The use of cleaning agents within the WD shall be within the conditions validated and specified as acceptable.

Cleaning agents with demonstrated performance at the end (or simulated end) of the shelf life of the cleaning agent shall be used.

The test procedure and test loads are designed to demonstrate conformance with the requirements of cleaning efficacy in accordance with ISO 15883-1:2024, 6.10, but take into consideration the nature of the loads and, if applicable, combination of cleaning and disinfection in a single stage.

The determination of cleaning efficacy shall be made on the defined cleaning stage only to generate data to establish that particular load(s) can be effectively cleaned in the WD.

6.7.2 Materials

6.7.2.1 Load carrier(s)

The load carrier(s) chosen for the test load shall be of the type recommended for the loads to be processed (see 6.2).

6.7.2.2 Test loads

The type test shall be carried out using the loads described in 6.2.

6.7.2.3 Test soils

Devices to constitute a test load shall be tested in accordance with ISO 15883-5:2021 using the particular test soil.

NOTE 1 Local requirements can specify the use of particular test soils and methods.

NOTE 2 The user's choice of test soils(s) and method(s) for performance testing can indicate a need to carry out similar testing before the WD is supplied.

The test soils used for the load, chamber wall, and load carriers may be the same or different. Where different test soils are used, the rationale for the choice of test soil shall be documented.

6.7.2.4 Water quality

If the efficacy of the cleaning solution is liable to be impaired by dilution with water above a certain level of hardness, then testing shall be carried out with a cleaning solution prepared with water at the maximum permitted hardness.

6.7.3 Procedure

Place the test load contaminated with the test soil in the chamber. Start a normal operating cycle for the load type under test. Interrupt the operating cycle after the cleaning stage and, if applicable, just prior to the start of the disinfection stage. A rinse with water may be provided after cleaning if defined as part of the cleaning stage. If an operating cycle with a combined cleaning and disinfecting stage is designed, interrupt the operating cycle after that combined stage and any rinse. Examine the test load as specified in ISO 15883-5:2021 for the particular test soil used for the presence of residual soil.

6.7.4 Results

Record the adequacy of the cleaning process stage. For routine test of the WD, visual inspection of load cleanliness is acceptable.

6.8 Test of disinfection efficacy

6.8.1 General

The conditions of use of disinfectant(s) in the WD shall be within the validated and specified conditions.

Disinfectant agents with demonstrated performance at the end (or simulated end) of the shelf life of the disinfectant agent shall be used.

NOTE National regulatory requirements can specify approval procedures prior to the use of a disinfectant in WD for medical devices.

6.8.2 Preliminary tests on chemical disinfectants

6.8.2.1 General

An initial series of tests, as set out in 4.3, intended to verify the in vitro microbicidal efficacy of the disinfecting agent under conditions identical to those which will be applied at the time of the cycle's disinfection stage, shall be carried out unless adequate data under relevant conditions can be supplied for shorter contact time(s).

For this, the microbicidal efficacy of the disinfecting agent shall be verified.

During these tests, experimental conditions intended to simulate the conditions within the WD shall be used.

6.8.2.2 Concentration

The disinfectant shall be tested at the minimum concentration available during the disinfection stage of the operating cycle, based on the MRC.

Each disinfectant recommended for use in the WD shall be tested.

In the case where it is intended that a disinfectant solution be re-used, the efficacy of the solution shall be determined as a function of its MRC, and, if required, at the MEC [see 4.3.4 and Clause 8 h)].

6.8.2.3 Temperature

Two cases shall be considered:

a) if the disinfection stage is carried out under uncontrolled temperature conditions, the test temperature shall be the minimum and maximum temperature permitted during the operating cycle's disinfection stage;

b) if the disinfection stage is carried out under temperature-controlled conditions, the testing shall be within the lower and upper temperature limits.

NOTE The temperature cannot be below the minimum or above the maximum temperatures indicated for the disinfectant.

6.8.2.4 Contact time

The contact time observed during the tests shall be the minimum duration of the disinfection stage.

6.8.2.5 Water quality

If the efficacy of the disinfectant is liable to be impaired by dilution with water above a certain level of hardness, then testing shall be carried out with the disinfectant prepared with water at the maximum permitted hardness.

NOTE Disinfectant testing according to standard methods is commonly performed in water of standardized hardness. ISO 15883-4:2018, 6.4 provides guidance on hardness of water used during type testing.

6.8.2.6 Neutralization

Before an investigation of the efficacy of the disinfectant, a method of neutralizing the disinfectant at the end of the exposure period shall be demonstrated and documented. This shall include demonstration that, for any neutralizing agent used, neither the neutralizing agent nor any reaction products with the disinfectant are microbicidal or inhibit the growth of the test organism. When a secondary host such as a cell culture is used as the detection system for the survival of test organisms, the absence of carry over effects on the cell culture system and detection of low numbers of test organisms added as a challenge to the test system shall be demonstrated.

NOTE ISO 11737-1 provides guidance on challenge testing to check for validity.

6.8.2.7 Test organisms

Test organisms shall be selected on the basis of antimicrobial claims made for the WD, with consideration of 4.3.

NOTE Guidance on choice of organisms can be obtained from relevant published method standards or generally accepted national guidelines on surface disinfectant efficacy testing, e.g. EN 13624,[5] EN 13727, EN 14476,[6] Reference [10], Reference [13].

6.8.2.8 Presentation of test organisms

While initial potency tests may be carried out using a suspension of test organisms, the demonstration of efficacy on contaminated surfaces shall be performed. The surfaces of the test pieces to be inoculated with test organisms shall be representative of those found in the WD chamber and the devices to be processed.

6.8.2.9 Detection of test organisms

The culture method used to enumerate the number of surviving microorganisms after exposure to the disinfectant shall be validated. The culture method shall be capable of recovering a low number, (approximately 10 CFU) of the organisms for which it is intended to be used.

6.8.3 Self-disinfection tests

6.8.3.1 Type test

The type test is intended to verify that the WD self-disinfection mode will disinfect those parts of the WD which come into contact with fluids which are intended to, or can, contact the load.

The bacterial levels in the final rinse water at the end of a normal operating cycle directly following the self-disinfection cycle shall be determined by the test method specified in Annex C.

In addition, thermometric verification of A_0 attainment shall be performed for thermal self-disinfection cycles in accordance with ISO 15883-1:2024, 6.8.

6.8.3.2 Operational and routine test

The bacterial levels in the final rinse water at the end of a normal operating cycle directly following the self-disinfection cycle shall be determined by the test method specified in Annex C.

NOTE 1 The test on the final rinse water can be sufficient to verify the self-disinfection cycle. The sample can be taken from any suitable point that ensures the water is collected following circulation through the WD.

NOTE 2 To increase the sensitivity of the test, a delay of time between the completion of the self-disinfection and the sampling (normal operating cycle) can be useful.

6.8.4 Chemical disinfection of the load

6.8.4.1 Type test

The test shall be carried out in accordance with Annex D.

The test method shall use a load in accordance with 6.2. Some examples of specific test load locations are suggested in Annex E, Figures E.1 through E.6.

A range of other microorganisms can be used in combination with a soil relevant to the load from ISO 15883-5:2021.

NOTE 1 A particular test organism is suggested in Annex D, but others can be used at the request of the user.

NOTE 2 Chilled storage of the test solution can prevent the growth of residual microorganisms if the time between neutralization of the disinfectant solution and quantification by plating the test suspension on agar is too long.

6.8.4.2 Operational and performance qualification tests

Operational and performance tests shall establish that the levels of all controlled process variables that affect the disinfection of the load are within the limits established during type testing.

NOTE When required by national regulation, specified test loads can be used to simulate the range of load items. Direct inoculation of test organisms, biological indicators, or bioburden testing (see ISO 11737-1) can be used to monitor the efficacy of the disinfection stage. Microorganisms incorporated into test soils can be used instead of, or as well as, inoculated test objects.

6.8.4.3 Routine tests

Routine tests shall establish that the levels of all controlled variables that affect the disinfection of the load are within the limits established during type testing.

7 Documentation

Documentation shall be in accordance with ISO 15883-1:2024, Clause 7.

8 Information to be supplied

In addition to the information specified in ISO 15883-1:2024, Clause 8, the following information shall be provided:

a) range of load supports available and required;

b) details of which parts of the WD are subjected to disinfection during the disinfection stage and self-disinfection cycle and which parts are not;

c) instructions on the frequency at which any water treatment equipment that is part of the WD requires disinfection;

d) locations of temperature sensors being representative of the lowest temperature of the system;

e) the cleaning agent(s) and disinfectant(s) used in the type testing;

f) directions to the user for the maximum period or number of operating cycles for which the reusable cleaning agent(s), or disinfectant(s), or both may be used;

g) the spectrum of antimicrobial activity (e.g. bactericidal, virucidal, fungicidal, mycobactericidal);

h) a method for the user to monitor the disinfectant to show that the disinfectant is at or above the MRC and, if necessary, the MEC (see 6.8.2.2);

i) maximum time required for the WD to be ready for use on start-up at minimum utilities;

j) if the water treatment equipment is not part of the WD, the requirements for water supplied to the WD including the requirement to control the microbial contamination of the water supply (see 4.7).

9 Marking, labelling, and packaging

The WD shall be marked externally in accordance with the requirements of IEC 61010-2-040:2020, Clause 5.

10 Information to be requested from the purchaser by the WD supplier

The requirements of ISO 15883-1:2024, Clause 10 apply.

Annex A
(informative)

Summary of test programmes

Table A.1 — Summary of tests in addition to ISO 15883-1

Brief description of test	Requirement Clause	Test Clause	Type test	Works test	OQ	PQ	Routine test
Accuracy of the dosing system	4.1.4	6.6	X	X	X	B	X (Q)
Cleaning efficacy	4.2.3	6.7	X	B	B	X	X[a]
Cleaning agent reuse	4.2.4	6.6.2	X	B	B	X	X (D)
Disinfectant reuse	4.3.4	6.6.2	X	B	B	X	X (D)
Load disinfection efficacy	4.3	6.8.4	X	B	X	X	X (D)
Chemical disinfectant	4.3.2	6.8.2	X	B	B	B	B
Final rinsing	4.4	6.3	O	B	B	X	X (Q)
Self-disinfection	4.5.4	6.8.3	X	B	B	X	X (Y)
Chemical Self-disinfection	4.5.4, 4.5.6	6.8.3, Annex B	X	B	O	B	B
Drying	4.6.2	6.4	X	B	X	B	X (D)
Water treatment equipment	4.7	6.3	X	B	B	X	X (Y)
Chemical dosing							
Accuracy and repeatability	in accordance with ISO 15883-1:2024, 5.7.5	in accordance with ISO 15883-1:2024, 6.9.1	X	—	—	X	—
Low level indicator	in accordance with ISO 15883-1:2024, 5.7.6	in accordance with ISO 15883-1:2024, 6.9.2	X	X	X	X	—
Concentration determination	4.1.4	6.6	X	—	X	X	—
Instrumentation							
Legibility	in accordance with ISO 15883-1:2024, 5.12.3	in accordance with ISO 15883-1:2024, 6.6.2	X	—	—	—	—
Calibration	in accordance with ISO 15883-1:2024, 5.11, 5.12.4, 5.13, 5.14 and 5.15	in accordance with ISO 15883-1:2024, 6.6.1	V	—	—	V	—
Fault indication	in accordance with ISO 15883-1:2024, 5.22.1 and 5.4.1.5	in accordance with ISO 15883-1:2024, 6.3.5, 6.3.6, 6.3.7 and 6.3.8	X	—	X	—	—

X = recommended

B = not recommended

O = optional test which can be requested by the purchaser or user

V = verification of calibration at the value(s) of interest for the particular instrument, e.g. the disinfectant temperature

Q = quarterly test interval, Y = yearly test interval, D = daily test interval

[a] For routine test of the WD, visual inspection of load cleanliness is acceptable.

The tests included in Table A.1 assume that all necessary installation qualification checks and tests have been completed satisfactorily.

Optional tests may be carried out at the discretion of the purchaser/user or can be required by local regulation.

Test intervals suggested are given for guidance only. Individual programmes of routine tests should be defined on the basis of a risk analysis, taking into account the conditions and reliability of the WD, the extent of independent monitoring of each cycle, and the use to which the WD is put.

Annex B
(normative)

Methods for microbiological evaluation of disinfection of liquid transport system

B.1 General

The following two methods are intended to simulate various incidents that can arise during normal use of the WD and that can give rise to contamination of the WD (see 4.7.2 and 6.3).

Method 1 (specified in B.5.2.1) tests the self-disinfection cycle after a simulated malfunction of the internal water treatment equipment that, although repaired quickly (24 h later), has caused contamination of the WD by the microorganisms present in the supply water.

Method 2 (specified in B.5.2.2) also simulates the case of WD contamination by microorganisms present in the supply water following a malfunction of the internal water treatment equipment. However, in this case, the self-disinfection cycle is only applied one week after a water equipment malfunction, and during this week the WD has continued to be used (one load washing/disinfection cycle per day). This allows evaluation of the efficacy of the self-disinfection cycle of a potentially contaminated WD after one week of use.

Monitoring the internal level of contamination of the WD during the interval of time between the water treatment equipment failure and the execution of the self-disinfection cycle allows evaluation of whether the WD's design is effective in limiting the development of microorganisms in the pipes of the WD.

B.2 Material

B.2.1 Microorganisms

Pseudomonas aeruginosa CIP A22 (or equivalent) as microorganism.

Bacterial suspension, with 1×10^9 CFU/ml to 1×10^{10} CFU/ml in sterile distilled water.

B.2.2 Culture medium

Soybean casein digest (SCD) agar or tryptone soya agar (TSA) (in accordance with EN 12353:2021 and EN 13727:2012+A2:2015), as maintenance and counting medium.

B.3 Washer-disinfector

B.3.1 Cycles

The following cycles shall be available:

— WD cycle;

— self-disinfection cycle;

— sampling cycle;

— contamination cycle.

B.3.2 Sampling cycle

The sampling cycle shall correspond to a routine cleaning and disinfection cycle interrupted during the stage before disinfection, and for which the cleaning agent shall be replaced by sterile distilled water. Once the cycle has been interrupted, a sample from the bottom of the tank containing water having circulated in the WD's pipe work shall be taken.

NOTE This sampling programme only includes the cleaning and rinsing stage and circulates water throughout the WD's pipe work, without there being any addition of disinfectant or cleaning agent.

If the cycle cannot be interrupted immediately prior to the disinfection stage, then a complete cycle substituting sterile purified (e.g. reverse osmosis) water for all process chemical solutions shall be used.

B.3.3 Contamination cycle

This special programme corresponds to a routine cleaning and disinfection cycle for which:

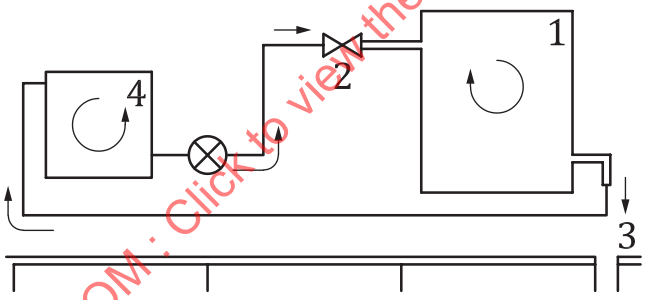
— the disinfectant solution heating system (if fitted) is deactivated;

— the cleaning agent and disinfectant are replaced by sterile distilled water.

During this contamination cycle, the WD is connected to the external tank containing the contamination solution (see Figure B.1), so that, during each stage of the contamination cycle, the WD is only fed with the contamination solution contained in the external tank.

B.4 Connection of the WD to the external tank

The connection of the WD to the external tank shall be as shown in Figure B.1.



Key

1 WD
2 water supply
3 drainage
4 external tank

NOTE As a function of the IFU for the WD, external peripherals can be inserted between the water supply network and the WD (water softener, etc.).

Figure B.1 — Connection of the WD to the external tank — Test configuration

B.5 Procedure

B.5.1 General

B.5.1.1 External tank disinfection

Before each test, subject the external tank in which the contamination solution is prepared to a thermal disinfection cycle with an A_0 of not less than 600.

B.5.1.2 Verification of absence of microbiocidal residue in the external tank after disinfection

During the last rinsing stage of the external tank, collect 9 ml of the water circulating in the external tank and associated pipework.

Incorporate 1 ml of a bacterial suspension of *Pseudomonas aeruginosa* at 1×10^3 CFU/ml in the previously sampled 9 ml of water.

After mixing thoroughly and 10 min of contact time, establish the number of viable bacteria present in the reaction mixture, T_N, by serial dilution and counting on a TSA agar plate.

The rinsing is only considered to be valid if Formula (B.1) is satisfied:

$$10 \times \frac{T_N}{T_t} \geq 0,8 \quad \text{(B.1)}$$

where

T_N is the number of viable bacteria present in the reaction mixture;

T_t is the exact concentration of bacteria in the bacterial suspension (control).

B.5.1.3 Preparation of the contamination solution

Fill the external tank with 30 l of tap water and 30 ml of a *Pseudomonas aeruginosa* suspension containing 10^9 CFU/ml. After thorough mixing, take a sample in order to establish, by serial dilution and counting on a TSA agar plate, the concentration of microorganisms in the contamination solution.

B.5.1.4 Contamination of the WD via the water supply network

After having prepared the contamination solution and deactivated the WD water treatment unit, connect the WD subjected to the tests to the external tank (Figure B.1). Then start the WD contamination cycle in order to ensure circulation of the contamination solution in all the internal piping of the WD.

B.5.1.5 Determination of the WD contamination level

During the different tests, determine the contamination level of the WD by running a sampling cycle and then establishing the concentration of microorganisms in the water having circulated in all the piping of the WD during this cycle. For this, during the sampling cycle collect 2 l of water in the tank of the WD. Filter 10 ml, 100 ml, and 1 000 ml of the 2 l of water through 0,2 µm membranes. Then rinse the membranes with 3 × 50 ml of sterile distilled water, place on counting medium and incubate at 37 °C for 24 h.

After incubation, count and identify the number of CFU and express the results as number of CFU per litre.

B.5.2 Establishment of the efficacy of the disinfection of the liquid transport system

B.5.2.1 Method 1

Proceed as follows:

a) install the WD;

b) run a self-disinfection cycle;

c) run a sampling cycle;

d) determine the WD contamination level;

e) deactivate the water treatment system (i.e. remove filter, deactivate heating system);

f) disinfect the external tank;

g) prepare the contamination solution;

h) contaminate the WD via the water supply network;

i) leave the WD to incubate at room temperature (not less than 20 °C) for 24 h;

j) connect the WD normally;

k) re-activate the water treatment system;

l) run a self-disinfection cycle;

m) run a sampling cycle;

n) determine the contamination level of the WD in accordance with B.5.1.5;

o) if the analysis of the results shows a total viable count of 10 CFU/100 ml or more or the presence of *Pseudomonas aeruginosa* in the sample taken during step n), repeat steps l), m) and n).

NOTE It is not necessary to determine the contamination level before the disinfection cycle [B.5.2.1, step l)], since the extent of contamination that will occur is specific to the design of the WD liquid transport system.

B.5.2.2 Method 2

Proceed as follows:

a) install the WD;

b) run a self-disinfection cycle;

c) run a sampling cycle;

d) determine the contamination level of the WD;

e) deactivate the water treatment system;

f) disinfect the external tank;

g) prepare the contamination solution;

h) contaminate the WD via the water supply network;

i) leave the WD to incubate at room temperature (not less than 20 °C) for 48 h;

j) connect the WD normally;

k) re-activate the water treatment system;

l) run a routine operating cycle;

m) run a sampling cycle;

n) determine the contamination level of the WD in accordance with B.5.1.5;

o) leave the WD to incubate at room temperature (not less than 20 °C) for 24 h;

p) repeat steps l) to o) four (4) times;

q) run a self-disinfection cycle;

r) run a sampling cycle;

s) determine the contamination level of the WD in accordance with B.5.1.5;

t) if the analysis of the results shows a total viable count of 10 CFU/100 ml or more or presence of *Pseudomonas aeruginosa* in the sample taken during step s), repeat steps q), r) and s).

B.6 Results and interpretation criteria

For both methods (Method 1 and Method 2), record the number of self-disinfection cycles needed to reduce the contamination to less than 10 CFU/100 ml with absence of *Pseudomonas aeruginosa*.

If multiple cycles are required to pass the test criteria, then this shall be included in the IFU for the WD.

Annex C
(normative)

Tests for microbiological contamination of final rinse water

C.1 Material

Sampling containers shall be 250 ml, or larger, and shall be sterile.

C.2 Procedure

Take samples from draw-off points adjacent to the WD and from the point of discharge into the WD chamber or load, and label them as “Supply Sample” and “WD Sample” respectively.

Test the samples within 4 h of collection or store at 2 °C to 5 °C and test within 48 h of collection.

Swab the discharge surfaces of the sampling points thoroughly with sterile or 0,2 µm filtered 70 % alcohol and allow to dry by evaporation immediately before the sample is taken.

Collect a sample of not less than 200 ml, or as specified, from each sampling point for each test to be carried out.

C.3 Test for aerobic mesophilic bacteria

Test post-disinfection rinse water for aerobic mesophilic bacteria in accordance with ISO 15883-1:2024, 6.4.2.4 and its Annex C.

C.4 Results

Report the number of aerobic mesophilic bacteria per 100 ml sample of final rinse water.

Report the number of *Pseudomonas aeruginosa* in 100 ml.

C.5 Interpretation criteria

There are fewer than 10 CFU/100 ml sample of final rinse water; and the water is free from *Pseudomonas aeruginosa* in 100 ml (see 6.3).

The presence of pseudomonads (e.g. *Burkholderia sp.)* or other bacteria in the rinse water can indicate the risk of biofilm development and this risk shall be further investigated.

Annex D
(normative)

Preparation and evaluation of indicators for microbiological testing of the efficacy of chemical disinfection of the load

D.1 Principles

Biological indicators shall contain *Enterococcus faecium* (for example, ATCC 6057[2)] or equivalent) as the test organism with at least 1×10^7 CFU for the quantitative method (see D.7.1) or 1×10^5 CFU to 1×10^6 CFU for the qualitative method (see D.7.2). For the carrier, use non-absorbent material in accordance with D.4.1. Suspend the test organisms in a test soil, apply them to the carrier surface, and allow them to dry.

The biological indicators shall be suitable to demonstrate the interaction of mechanical and chemical cleaning/disinfection. Mechanical removal alone (i.e. wash-off) shall be evaluated in the absence of the disinfectant.

D.2 Reagents

D.2.1 Suspension of the test organism in blood or bovine albumin and mucin (RAM), respectively in accordance with D.3.2.1 and D.3.2.2.

D.2.2 Tryptone soya agar (TSA) or tryptone soya broth (TSB) or in accordance with EN 13727:2012+A2:2015 or equivalent.

D.2.3 Phosphate buffer solution consisting of the following:

— solution A: dissolve 16 g NaCl, 0,4 g KCl, 0,4 g KH_2PO_4 in 1 600 ml of distilled water;

— solution B: dissolve 0,2 g $CaCl_2$ in 200 ml of distilled water;

— solution C: dissolve 0,2 g $MgSO_4$ in 200 ml of distilled water;

— pH 7,4.

Sterilize the solutions A, B, and C and, after completion of the cooling period, mix them under sterile conditions adding the inactivation substances at the same time, if required.

D.3 Test organisms

D.3.1 General

Enterococcus faecium (for example, ATCC 6057) is recommended for the test.

Enterococcus faecium cultures shall be inoculated on new tryptone soya agar (TSA) plates every 48 h. The incubation temperature shall be (36 ± 1) °C. To obtain the test organism suspension, wash the plates off using 10 ml of physiological NaCl solution. Wash the suspensions by centrifugation, followed by re-suspension in NaCl solution and another centrifugation. Subsequently, the test organisms are added to the test soil [blood or bovine albumin and mucin (RAM) in accordance with D.2] corresponding to the initial volume.

2) ATTC American Type Culture Collection, 10801 University Boulevard, Manassas, VA 20110-2209, USA, www.attc.org. The ATCC number is the collection number of strains supplied by these culture collections. This information is given for the convenience of users of this document and does not constitute an endorsement by ISO of the product named.

The minimum total of the test organism solution (blood or RAM) shall be 1×10^8 CFU/ml for the quantitative method or 1×10^6 CFU/ml to 1×10^7 CFU/ml for the qualitative method.

D.3.2 Test soils

D.3.2.1 For the blood test soil, use sterile defibrinated sheep blood.

D.3.2.2 For the RAM soil, dissolve 0,6 g bovine albumin and 1 g mucin in 100 ml NaCl peptone solution.

D.4 Test surfaces

D.4.1 General

Test surfaces should be identified as defined in 6.8.2.8. Stainless steel plates of type X5CrNi18-10 with finish 2B can be used, and if used shall be ground (2G) with grain size of approximately 80 in accordance with EN 10088-1 and EN 10088-2, with dimensions of 10 mm × 130 mm and an approximate thickness of 1,5 mm.

D.4.2 Cleaning

Test surfaces shall be cleaned in accordance with the following protocol:

a) immerse the test surfaces in acetone ACS while swirling the solution with a magnetic bar for 1 min to remove oil from the surface;

b) transfer test surfaces to 5 % solution of an appropriate cleaning agent and heat at 75 °C for 10 min;

c) decant cleaning agent solution;

d) rinse with deionized or distilled water followed by immersion in 70 % alcohol for 1 min to facilitate drying.

Remove the test surfaces with tweezers, cleaned as above, then place on clean, absorbent paper (e.g. filter paper) and allow to dry.

D.4.3 Contamination of the test surfaces

Use test soil samples of 0,1 ml each to distribute uniformly on an area within dimensions of 8 mm × 100 mm, taking care not to contaminate the sides. The surfaces thus soiled are allowed to dry for a duration of 24 h at a temperature of (22 ± 1) °C and a relative humidity of (50 ± 10) %.

The total per contaminated surface (at least 1×10^7 CFU) shall be high enough for a reduction factor (RF) > 5 to be representable considering the detection limit.

D.5 Storage

The biological indicators prepared in accordance with D.4 shall be stored at temperatures between 4 °C and 8 °C. They are suitable for testing as validated by the test laboratory to meet the microbial CFU counts specified in D.4.3.

D.6 Test for resistance

In order to determine the heat resistance of *Enterococcus faecium*, transfer 20 biological indicators into test tubes containing tryptone soya broth (TSB) previously heated to 70 °C in a water bath. The amount of TSB shall be chosen so that the biological indicators are completely immersed.

Immerse the test tubes with the carriers in a water bath of 70 °C for 10 min and cool in iced water.

Following the subsequent incubation at a temperature of (36 ± 1) °C for 24 h, 90 % of the carriers shall show growth of test organisms.

D.7 Evaluation and acceptance criteria

D.7.1 Evaluation of the biological indicators using the quantitative method

Evaluation of the biological indicators by means of the quantitative method is preferably used for type testing.

The biological indicators are placed into the WD and subjected to an operating cycle. Once the operating cycle is completed, remove the biological indicators under aseptic working conditions and subject them to a visual control for residues of the test soil. Any residues shall be stated in the accompanying report. Transfer each biological indicator individually into 10 ml phosphate buffer solution with a pH of 7,4 with neutralization media suitable for the disinfectants.

Without treatment in the WD, add evaluated three positive controls to 10 ml phosphate buffer solution in accordance with D.2.

Then shake the biological indicators transferred into phosphate buffer solution by means of an agitator (with a frequency of approximately $500 \cdot \text{min}^{-1}$) for at least 20 min in order to recover the test organisms. Subsequently, determine the CFU from the agitated liquid by culturing on TSA for (36 ± 1) °C for 72 h. The method used (including type of culture media and determination of CFU) shall be given in the test report.

The reduction factor (RF) is calculated by Formula (D.1):

$$RF = \log_{10}CFU_1 - \log_{10}CFU_2 \quad \text{(D.1)}$$

where

RF is the reduction factor;

$\log_{10}CFU_1$ is the mean of the logarithms of the number of CFU of transport/positive controls;

$\log_{10}CFU_2$ is the logarithm of the number of CFU of the treated test surface.

The performance is deemed to be sufficient when a reduction factor ≥ 5 has been achieved for all biological indicators.

The results from the testing of mechanical removal alone (i.e. wash-off in the absence of the disinfectant) shall be reported.

D.7.2 Evaluation of the biological indicators using the qualitative method

The detection of test organism is used preferably for the purposes of requalification.

For every examination, a level of 1×10^5 CFU to 1×10^6 CFU per 0,1 ml of the test soil shall be proven using a positive control.

Transfer the biological indicators individually into sterile tubes and send them to the laboratory.

Incubation is carried out in TSB at (36 ± 1) °C for 72 h; any nutrient solutions showing growth are sub-cultivated in order to detect the test organism. In case of a positive examination result, the test shall be repeated.

The performance is deemed to be sufficient when none of the exposed biological indicators shows growth.

The results from the testing of mechanical removal alone (i.e. wash-off in the absence of the disinfectant) shall be recorded.

D.7.3 Acceptance criteria

Incubate the sample volumes taken from the disinfectant solution at (36 ± 1) °C for a duration of 72 h.