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**Microbiology of the food chain —  
Horizontal methods for surface  
sampling**

*Microbiologie de la chaîne alimentaire — Méthodes horizontales pour  
les prélèvements de surface*

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## 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](http://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](http://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 on 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 the following URL: [www.iso.org/iso/foreword.html](http://www.iso.org/iso/foreword.html). (standards.iteh.ai)

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This second edition cancels and replaces the first edition (ISO 18593:2004), which has been technically revised. The main changes are as follows:

- recommendations on sampling procedure have been added, including information on sampling location, area and sampling time;
- examples of neutralizers have been added as [Annex A](#).

## Introduction

It can be important to determine the presence of, or the number of microbes on, the surfaces of utensils, work surfaces and other equipment in the food chain environment in order to estimate the level of contamination in the food chain environment.

This document describes horizontal methods for surface sampling.

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# Microbiology of the food chain — Horizontal methods for surface sampling

## 1 Scope

This document specifies horizontal methods for sampling techniques using contact plates, stick swabs, sponges and cloths on surfaces in the food chain environment in order to detect and enumerate culturable microorganisms such as pathogenic or non-pathogenic bacteria or yeasts and moulds.

NOTE The term “environment” means any item in contact with the food product or likely to represent a contamination or recontamination source; for example, material, premises or operators.

This document does not apply to the validation of cleaning and disinfection procedures.

This document does not apply to sampling techniques for primary production samples, which are covered by ISO 13307. Sampling techniques for carcasses are covered by ISO 17604. Sampling techniques for analysis of noroviruses and hepatitis A viruses are covered by ISO 15216-1.

This document does not give advice on sampling frequency, the number of sampling points, or the need to rotate sampling points, as these are chosen on a case-by-case basis.

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## 2 Normative references (standards.iteh.ai)

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 6887-1, *Microbiology of the food chain — Preparation of test samples, initial suspension and decimal dilutions for microbiological examination — Part 1: General rules for the preparation of the initial suspension and decimal dilutions*

ISO 6887-5, *Microbiology of food and animal feeding stuffs — Preparation of test samples, initial suspension and decimal dilutions for microbiological examination — Part 5: Specific rules for the preparation of milk and milk products*

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

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

ISO 16140-2, *Microbiology of the food chain — Method validation — Part 2: Protocol for the validation of alternative (proprietary) methods against a reference method*

## 3 Terms and definitions

No terms and definitions are listed in this document.

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

- ISO Online browsing platform: available at <https://www.iso.org/obp>
- IEC Electropedia: available at <http://www.electropedia.org/>

## 4 Principle

Sampling schemes aim to evaluate microbial contamination levels of surfaces from the food chain environment in order to implement corrective actions to avoid food contamination by microorganisms. Ineffective sampling programmes or techniques may result in the non-detection of microorganisms when they are present.

This document describes surface sampling methods to detect or enumerate microorganisms from surfaces in the food chain environment. Different sampling techniques are described, including contact plates, stick swabs, cloths and sponges.

This document also gives recommendations on the locations and areas to be sampled and the most appropriate sampling time.

According to the device and the microorganism to be detected or enumerated, determination of the microbial contamination of the surface can be performed by

- sampling of the surface, and
- analysis according to specific standards.

## 5 Culture media and reagents

Follow current laboratory practice as specified in ISO 7218. Follow performance testing of culture media as specified in ISO 11133.

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### 5.1 Diluent.

In general, the diluent is sterile buffered peptone water, peptone salt as specified in ISO 6887-1, peptone solution at 1 g/l or quarter-strength Ringer's solution as specified in ISO 6887-5, with neutralizer(s) (5.3) added if necessary. <https://standards.iteh.ai/catalog/standards/sist/a761e123-92c6-4707-bba3-9ff7ad8bc53a/iso-18593-2018>

NOTE To extend transport time, an appropriate transport diluent can be added, if properly validated.

### 5.2 Medium for contact plate.

Plates (6.1) may vary in diameter or area, according to the type of surface to be sampled. The medium is chosen according to the ISO method for the microorganism(s) of concern, with neutralizer(s) (5.3) added if necessary. The medium shall form a convex meniscus with the contact plate.

Alternative media formulations shall be supported by a validation study as specified in ISO 16140-2.

### 5.3 Neutralizer.

In cases where residues of disinfectants are expected, appropriate neutralizer(s) should be added to the diluent (5.1) or media (5.2) before sampling, to prevent any inhibitory effect of the disinfectants on the growth of microorganisms.

Do not add neutralizer to the diluent when no residual disinfectant is expected<sup>[9]</sup>. A neutralizer used to quench residual disinfectant can have a slight deleterious impact on bacterial cells and it is likely that such an impact would be greater when cells are stressed.

An appropriate neutralizer for all situations (a “universal neutralizer”) cannot be prescribed<sup>[9]</sup>. A number of neutralizers are recommended in EN 1276, EN 1650, EN 13697 and EN 13704.

See [Annex A](#) for examples of neutralizers.



## 6 Equipment and consumables

Disposable apparatus is an acceptable alternative to reusable glassware if it has similar specifications.

Usual microbiological laboratory equipment in accordance with ISO 7218 and, in particular, the following.

### 6.1 Contact plate, plastic dish of variable diameter.

NOTE It is also possible to use any other flexible or rigid container which enables contact with the sampled surface.

### 6.2 Sterile stick swab, stick with cotton or synthetic material (such as alginate or rayon) contained in a tube or envelope. The material used shall be documented to be free of inhibitory substances.

NOTE For some types of surface, the cotton residues can contaminate the internal parts of these surfaces after sampling.

### 6.3 Sterile cloth (or wipe), free from inhibitory substances.

### 6.4 Sterile sponge, with or without stick/handle, free from inhibitory substances.

### 6.5 Containers, such as bottles, tubes or flasks, suitable for the sterilization and storage of culture media.

### 6.6 Cool box, cooler, insulated box containing ice packs, capable of maintaining the samples at low temperature during transportation to the laboratory.

### 6.7 Mixer, for mixing liquids in culture tubes, e.g. a vortex mixer.

### 6.8 Peristaltic blender, with sterile plastic bags to prepare initial suspensions by peristaltic movement.

### 6.9 Petri dishes, made of plastic or glass.

### 6.10 Sterile disposable or reusable template, enclosing a specified area.

## 7 Sampling procedure

### 7.1 General

Sampling locations and areas, sampling times and sampling techniques should be selected according to risk-based principles and should relate to the higher probability of detecting contaminated surfaces during food processing, when measuring the hygiene of specific production steps or the entire process as appropriate. Always keep the same sampling procedure for a specific routine to allow trending of data.

### 7.2 Sampling location

Microorganisms can be found on visually clean surfaces but are most frequently found on wet and soiled places where the bacteria are able to grow and persist. Hard to reach places such as holes or crevices in fibrous, porous, difficult-to-clean equipment, rusting and hollow materials, are potential harbourage sites that should be sampled. It can be difficult to sample unreachable areas where food debris can collect. Dismantling may be necessary to sample unreachable locations.

The choice of sampling location shall be defined according to historical data linked to each site and after step-by-step examination of the process.