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# Water quality — Enumeration of *Clostridium perfringens* — Method using membrane filtration

**WARNING** — Persons using this document should be familiar with normal laboratory practice. This document does not purport to address all of the safety problems, if any, associated with its use. It is the responsibility of the user to establish appropriate safety and health practices and to ensure compliance with any national regulatory conditions.

**IMPORTANT** — It is absolutely essential that tests conducted in accordance with this document be carried out by suitably qualified staff.

## 1 Scope

This International Standard specifies a method for the enumeration of vegetative cells and spores of *Clostridium perfringens* by the membrane filtration method in samples of water intended for human consumption. However, the method can be applied to all types of water samples provided they do not contain particulate or colloidal matter that interferes with filtration.

## 2 Normative references

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

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

ISO/TS 11133-1, *Microbiology of food and animal feeding stuffs — Guidelines on preparation and production of culture media — Part 1: General guidelines on quality assurance for the preparation of culture media in the laboratory*

ISO 19458, *Water quality — Sampling for microbiological analysis*

ISO/IEC Guide 2:2004, *Standardization and related activities — General vocabulary*

## 3 Terms and definitions

For the purposes of this document, the terms and definitions given in ISO/IEC Guide 2 and the following apply:

### 3.1

#### **presumptive *Clostridium perfringens***

bacteria which produce all shades of black or grey to yellow brown colonies on tryptose-sulfite-cycloserine agar, even if the colour is faint, after anaerobic incubation at  $(44 \pm 1)^\circ\text{C}$  for  $(21 \pm 3)$  h

Note 1 to entry: Unlike colonies growing directly on the agar medium, colonies on the membrane do not always display a distinct blackening, so faint colonies are included in the count.

### 3.2

#### **confirmed *Clostridium perfringens***

bacteria that produce characteristic colonies on tryptose-sulfite-cycloserine agar and possess the enzyme acid phosphatase