

Methods for

Microbiological examination of food and animal feeding stuffs —

Part 9: Enumeration of *Clostridium
perfringens* —

[ISO title: Microbiology — General
guidance for enumeration of
Clostridium perfringens — Colony
count technique]

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- Agricultural and Food Research Council: Food Research Institute
- British Food Manufacturing Industries' Research Association
- British Poultry Federation Limited
- Campden Food Preservation Research Association
- Department of Agriculture, Northern Ireland
- Department of Health and Social Security
- Department of Trade and Industry (Laboratory of the Government Chemist)
- Flour Milling and Baking Research Association
- Food Manufacturers' Federation Incorporated
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- Joint Committee of the Milk Marketing Board and the Dairy Trade Federation
- Ministry of Agriculture, Fisheries and Food
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- Pet Food Manufacturers' Association
- Public Health Laboratory Service
- Society for Applied Bacteriology
- UK Association of Frozen Food Producers
- United Kingdom Dairy Association
- University of Reading

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National foreword

This Part of BS 5763 has been prepared under the direction of the Food and Agriculture Standards Committee. It is identical with ISO 7937-1985 “*Microbiology — General guidance for enumeration of Clostridium perfringens — Colony count technique*” published by the International Organization for Standardization (ISO) and in the preparation of which the United Kingdom played a full part.

Terminology and conventions. The text of the international standard has been approved as suitable for publication as a British Standard without deviation. Some terminology and certain conventions are not identical with those used in British Standards; attention is drawn especially to the following.

The comma has been used as a decimal marker. In British Standards it is current practice to use a full point on the baseline as the decimal marker. It is also current practice in British Standards to use the symbol “L” for litre (and its submultiples) instead of “l” and to use the spelling “sulphur” etc. instead of “sulfur” etc.

Wherever the words “International Standard” appear, referring to this standard, they should be read as “British Standard”.

Cross-reference

International standard	Corresponding British Standard
ISO 6887-1983	BS 5763 Methods of microbiological examination of food and animal feeding stuffs Part 6:1983 Preparation of dilutions (Identical)

A British Standard does not purport to include all the necessary provisions of a contract. Users of British Standards are responsible for their correct application.

Compliance with a British Standard does not of itself confer immunity from legal obligations.

Summary of pages

This document comprises a front cover, an inside front cover, page i and ii, pages 1 to 8, an inside back cover and a back cover.

This standard has been updated (see copyright date) and may have had amendments incorporated. This will be indicated in the amendment table on the inside front cover.

0 Introduction

0.1 This International Standard is intended to provide general guidance for the examination of products not dealt with by existing International Standards and for the reference of bodies preparing microbiological methods of test for application to food products or to animal feeding stuffs. In view of the large variety of products within this field of application, these guidelines may not be appropriate for some products in every detail, and for some other products it may be necessary to use different methods. Nevertheless, it is hoped that, in all cases, every attempt will be made to apply these guidelines as far as possible and that deviations from them will only be made if absolutely necessary for technical reasons.

When this International Standard is next reviewed, account will be taken of all information then available regarding the extent to which the guidelines have been followed and the reasons which necessitated deviation from them in the case of particular products.

The harmonization of test methods cannot be immediate, and for certain groups of products, International Standards and/or national standards that do not comply with the guidelines may already exist. In cases where International Standards already exist for the product to be tested, they should be followed, but it is hoped that, when they are reviewed, they will be aligned with this International Standard so that, eventually, the only remaining departures from these guidelines will be those necessary for well established technical reasons.

0.2 For practical reasons, the definition of *Clostridium perfringens* given in clause 3 and used as the basis for the procedure does not exclusively describe strains of *Clostridium perfringens*. In particular, the confirmatory tests are inadequate to distinguish between *Clostridium perfringens* and other closely related but less commonly encountered *Clostridium* species such as *C. paraperfringens* and *C. absonum*.

0.3 For statistical reasons, the lowest limit for the number of colonies counted per dish has been set at 15, but, for practical purposes, a count of lower numbers of *Clostridium perfringens* is often required. The confidence limits of such determinations (estimated counts) are given in the annex.

1 Scope and field of application

This International Standard gives general guidance for the enumeration of viable *Clostridium perfringens* in products intended for human consumption or feeding of animals.

2 Reference

ISO 6887, *Microbiology — General guidance for the preparation of dilutions for microbiological examination*.

3 Definitions

For the purpose of this International Standard, the following definitions apply.

3.1

clostridium perfringens

bacteria that form black colonies in the specified selective medium and which give positive confirmatory reactions when the test is carried out by the method specified in this International Standard

3.2

enumeration of C. perfringens

determination of the number of viable and confirmed *C. perfringens* bacteria per millilitre or per gram of sample, when the test is carried out by the method specified in this International Standard

4 Principle

4.1 Inoculation of Petri dishes with a specified quantity of the test sample if the initial product is liquid, or a specified quantity of the initial suspension in the case of other products.

Inoculation, under the same conditions, using decimal dilutions of the test sample or of the initial suspension.

Mixing with a selective medium (poured plate technique) and adding an overlayer of the same medium.

4.2 Anaerobic incubation of the plates at 35 °C or 37 °C¹⁾ for 20 h.

4.3 Calculation, from the number of black colonies appearing on the plates, of the number of characteristic colonies.

4.4 Subjection of characteristic colonies to confirmatory procedures and calculation of the number of *C. perfringens* per millilitre or per gram of sample.

¹⁾ The temperature should be agreed between the parties concerned and recorded in the test report.