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**Microbiology of the food chain —
Horizontal method for the
determination of *Vibrio* spp. —**

**Part 2:
Enumeration of total and
potentially enteropathogenic *Vibrio*
parahaemolyticus in seafood using
nucleic acid hybridization**

*Microbiologie de la chaîne alimentaire — Méthode horizontale pour
la détermination des Vibrio spp. —*

*Partie 2: Dénombrement de Vibrio parahaemolyticus total et
potentiellement entéropathogène dans les fruits de mer, par
hybridation des acides nucléiques*

Reference number
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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

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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).

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 34, *Food products*, Subcommittee SC 9, *Microbiology*.

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

Potentially enteropathogenic strains of *Vibrio parahaemolyticus* possess thermostable direct haemolysin (TDH) and/or thermostable direct hemolysin-related hemolysin (TRH). TDH positive strains manifest Kanagawa phenomenon (KP)^[1]. This characteristic is traditionally utilized in the identification of enterotoxigenic strains of *V. parahaemolyticus*. Strains possessing TRH do not share the haemolytic characteristics of TDH positive isolates and no conventional identification assay has been reported for TRH identification. Pathogenic strains in the environment are a minority^[2] and differentiation between enteropathogenic and total *V. parahaemolyticus* presence is therefore useful.

This document enables the enumeration of potentially enteropathogenic *V. parahaemolyticus* and/or of total *V. parahaemolyticus*.

Microbiology of the food chain — Horizontal method for the determination of *Vibrio* spp. —

Part 2:

Enumeration of total and potentially enteropathogenic *Vibrio parahaemolyticus* in seafood using nucleic acid hybridization

1 Scope

This document specifies a method for the direct enumeration of potentially enteropathogenic *V. parahaemolyticus* (*tdh* and/or *trh* positive) and/or the enumeration of total *V. parahaemolyticus* in seafood.

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

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 6887-3, *Microbiology of the food chain — Preparation of test samples, initial suspension and decimal dilutions for microbiological examination — Part 3: Specific rules for the preparation of fish and fishery products*

ISO 11133, *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.

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/>

3.1

oligoprobe

labelled, artificially produced segment of DNA with a defined sequence used to detect target DNA by hybridization