



**International  
Standard**

**ISO 25184**

**Molecular biomarker analysis —  
Nucleotide sequencing — Verified  
next generation sequences (VNGS)**

*Analyse des biomarqueurs moléculaires — Séquençage  
nucléotidique — Séquences vérifiées de nouvelle génération  
(VNGS)*

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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 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](http://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](http://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](http://www.iso.org/iso/foreword.html).

This document was prepared by Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 16, *Horizontal methods for molecular biomarker analysis*, in collaboration with AOAC INTERNATIONAL. It is being published by ISO and separately by AOAC INTERNATIONAL. The requirements described in this document are equivalent to the Official Methods of Analysis of AOAC INTERNATIONAL, Appendix T, *Standard Requirements for Nucleotide Sequences used in Biothreat Agent Detection, Identification, and Quantification: Verified New Generation Sequences (VNGS)*.

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](http://www.iso.org/members.html).

## Introduction

Nucleotide sequencing is cross-cutting across biomolecular methods in agriculture, food production and the life sciences. Bioinformatic analytical methods and pipelines require reliable reference sequences for comparison with unknown sample sequences. The verified next generation sequence (VNGS) concept was developed by the AOAC INTERNATIONAL. It is designed to provide the requirements necessary for a reliable and accurately determined next generation reference sequence. This document provides the requirements for a VNGS.

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# Molecular biomarker analysis — Nucleotide sequencing — Verified next generation sequences (VNGS)

## 1 Scope

This document specifies requirements for reference next generation nucleotide sequences.<sup>[1][2]</sup>

This document is applicable to all verified next generation (VNGS) nucleotide sequences determined by next generation sequence (NGS) technology that are accessible on the semantic web and included in a database (public or private).<sup>[3][4][5][6]</sup>

## 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 16577, *Molecular biomarker analysis — Vocabulary for molecular biomarker analytical methods in agriculture and food production*

## 3 Terms and definitions

For the purposes of this document, the terms and definitions given in ISO 16577 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 alignment

sequence alignment

arrangement of *nucleotide sequences* (3.27) so that regions of similarity versus dissimilarity, polymorphic regions and haplotype are shown

### 3.2 ASCII character set

**American Standard Code for Information Interchange character set**  
character encoding standard for electronic communication

### 3.3 assemblies

set of DNA segments or sequences that overlap in a way that provides a contiguous representation of a genomic region

### 3.4 base calling

computational process in massively parallel sequencing for translating raw optical and electrical signals to *nucleotide sequence* (3.27)