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**Water quality — Determination of  
nitrate in water using small-scale  
sealed tubes —**

**Part 2:  
Chromotropic acid colour reaction**

*Qualité de l'eau — Détermination du nitrate dans l'eau par la  
méthode à petite échelle en tubes fermés —*

*Partie 2: Réaction colorimétrique à l'acide chromotropique*



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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 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 147, *Water quality*, Subcommittee SC 2, *Physical, chemical and biochemical methods*.

A list of all parts in the ISO 23696 series can be found on the ISO website.

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

Nitrate plays an important role in nature and are naturally present in ecosystems. The main anthropic sources are agriculture (fertilizers and manure) and industrial sources such as food, chemical, paper companies, etc. and as a result of natural nitrification processes of reduced forms of nitrogen. In turn, nitrate can be converted to nitrogen gas by denitrifying bacteria. This process takes on strategic importance within biological water purification plants.

The presence of significant concentrations of nitrate in surface and bathing water, drinking water and wastewater can pose a risk to human health and/or environment.



# Water quality — Determination of nitrate in water using small-scale sealed tubes —

## Part 2: Chromotropic acid colour reaction

**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.

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

### 1 Scope

This document specifies a method for the determination of nitrate as  $\text{NO}_3\text{-N}$  in water of various origin such as natural water (including groundwater, surface water and bathing water), drinking water and wastewater, in a measuring range of concentration between 0,20 mg/l and 30 mg/l of  $\text{NO}_3\text{-N}$  using the small-scale sealed tube method. Different measuring ranges of small-scale sealed tube methods can be required.

The measuring ranges can vary depending on the type of the small-scale sealed tube method of different manufacturers.

It is up to the user to choose the small-scale sealed tube test with the appropriate application range or to adapt samples with concentrations exceeding the measuring range of a test by preliminary dilution.

**NOTE 1** The results of a small-scale sealed tube test are most precise in the middle of the application range of the test.

Manufacturers' small-scale sealed tube methods are based on chromotropic colour reaction, depending on the typical operating procedure of the small-scale sealed tube used, see [Clause 9](#).

**NOTE 2** Laws, regulations or standards can require that the data is expressed as  $\text{NO}_3$  after conversion with the stoichiometric conversion factor 4,426 81 in [Clause 11](#).

**NOTE 3** In the habitual language, use of sewage treatment and on the displays of automated sealed-tube test devices,  $\text{NO}_3$  without indication of the negative charge has become the common notation for the parameter nitrate and especially for the parameter nitrate-N. This notation is adopted in this document even though not being quite correct chemical nomenclature.

### 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 5667-1, *Water quality — Sampling — Part 1: Guidance on the design of sampling programmes and sampling techniques*

ISO 5667-3, *Water quality — Sampling — Part 3: Preservation and handling of water samples*

ISO 5667-10, *Water quality — Sampling — Part 10: Guidance on sampling of waste water*

ISO 8466-1, *Water quality — Calibration and evaluation of analytical methods — Part 1: Linear calibration function*

ISO 8655-2, *Piston-operated volumetric apparatus — Part 2: Pipettes*

ISO/IEC 17025, *General requirements for the competence of testing and calibration laboratories*

### 3 Terms and definitions

For the purposes of this document, the following terms and definitions 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**  
**small-scale sealed tube**  
 glass tube commercially available on the market, prefilled by the manufacturer with reagent(s), to develop a colour to be read by a photometer or a spectrophotometer

### 4 Principle

The nitrate content of the sample to be analysed reacts in acidic solution with chromotropic acid, which is read at maximum absorption by a photometer or a spectrophotometer.

The method serves to obtain the concentration of nitrate present in the sample.

### 5 Interferences

Typical interfering substances are metals, chloride and organic load [chemical oxygen demand (COD)]. Examples of maximum compatible concentrations are listed in [Table 1](#).

Every manufacturer of small-scale sealed tube shall provide information about interference levels above which the ion interferes. The concentration of interfering substances can depend on the ratio of sampled volume and pre-dosed reagents in the small-scale sealed tube.

**Table 1 — Examples of interfering ions and organic load**

Substances	Maximum tolerable concentrations
	mg/l
K <sup>+</sup>	500
Na <sup>+</sup>	500
Cl <sup>−</sup>	500
COD	200
Ag <sup>+</sup>	100
Ca <sup>2+</sup>	50
Fe <sup>2+</sup>	10
Cr(VI)	5

High concentrations of chloride require precipitation in form of silver chloride with silver sulfate or, if applicable, by dilution as described below in this clause.

When calcium concentrations are greater than those indicated as examples in [Table 1](#), the reaction can produce turbidity.