



BSI Standards Publication

Water quality — Spectrophotometric determination of chlorophyll-a content by ethanol extraction for the routine monitoring of water quality

National foreword

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English Version

Water quality - Spectrophotometric determination of chlorophyll-a content by ethanol extraction for the routine monitoring of water quality

Qualité de l'eau - Détermination spectrophotométrique
de la teneur en chlorophylle a par extraction à l'éthanol
pour la surveillance de routine de la qualité de l'eau

Wasserbeschaffenheit - Bestimmung des Chlorophyll-
a-Gehalts durch Ethanolextraktion für die
routinemäßige Überwachung der Wasserqualität

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European foreword

This document (EN 17899:2024) has been prepared by the Technical Committee CEN/TC 230 “Water analysis”, the secretariat of which is held by DIN.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by January 2025, and conflicting national standards shall be withdrawn at the latest by January 2025.

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Introduction

Chlorophyll-a is the most common essential photosynthetic pigment present in photoautotrophic plankton organisms. It is the main component of the dynamically regulated photosystem of these organisms, in which other accessory pigments are involved, some of which are chemically very similar to chlorophyll-a.

The chlorophyll-a content depends on the species composition of the phytoplankton, the time of day and season, the place and the depth of sampling. It is also suitable for quantifying the change in the algal biomass (cell proliferation) in biological tests to check the toxicity of substances dissolved in water.

The chlorophyll concentration of a water sample can provide information about the trophic state of a water body. It is used as an easily determinable measure of the phytoplankton biomass and is a key variable in many trophy scoring systems. Even if this value cannot be used as an absolute measure for the phytoplankton biomass, the determination of the chlorophyll-a concentration together with other biomass and bioactivity parameters provides information about the quantitative occurrence and the potential metabolic performance of the phytoplankton in waters.

Due to the sensitivity of chlorophyll to light, acids and enzymes, there is currently no universally applicable routine analytical method that enables an accurate, artefact-free and at the same time simple determination of the chlorophyll-a content in water samples containing phytoplankton. The extractive, spectrophotometric method described in this document therefore provides an operationally defined value. As extracting agent hot ethanol is used. Various other extractants (e.g. acetone or methanol) are described in the literature, but these may have lower extraction efficiency or are toxicologically problematic.

WARNING — Persons using this document should be familiar with usual 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.

Annexes A, B, C and D of this document are for information only.

1 Scope

This document describes a spectrophotometric method for determining the chlorophyll-a content corrected for phaeopigments as a measure of the amount of phytoplankton for all types of surface water including marine water. The determination limit is usually 2 µg/l to 5 µg/l and is calculated by each laboratory individually. It can be as low as 0,5 µg/l using 2 l of sample (or even more) and a 50 mm cuvette.

NOTE In some measurement programs like marine studies on time series data and ecological status/classification no correction for phaeopigments is used and acidification is omitted, e.g. as recommended by OSPAR.

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.

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

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 chlorophyll-a

natural plant pigment that is present in phytoplankton, most important photosynthetic pigment of photoautotrophic organisms

Note 1 to entry: For the purposes of this document, the pigment whose concentration can be determined by measuring the absorbance at 665 nm in an ethanolic solution corrected for phaeopigments.

3.2 phaeopigments

collective term for chlorophyll breakdown products, which do not contain any magnesium

EXAMPLE Phaeophytin is a phaeopigment.

3.3 phytoplankton

community of free-living, suspended, mainly photosynthetic organisms in aquatic systems comprising cyanobacteria and algae

3.4 accessory pigments

auxiliary pigments that support photosynthesis

EXAMPLE Carotinoids, phycobilins, chlorophyll-b and chlorophyll-c.