

Water quality — Guidance standard on the enumeration of phytoplankton using inverted microscopy (Utermöhl technique)

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National foreword

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A list of organizations represented on EH/3/5 can be obtained on request to its secretary.

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Water quality - Guidance standard on the enumeration of phytoplankton using inverted microscopy (Utermöhl technique)

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l'abondance et de la composition du phytoplancton par
microscopie inversée (méthode d'Utermöhl)

Wasserbeschaffenheit - Anleitung für die Zählung von
Phytoplankton mittels der Umkehrmikroskopie (Utermöhl-
Technik)

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Foreword

This document (EN 15204:2006) has been prepared by 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of an identical text or by endorsement, at the latest by February 2007, and conflicting national standards shall be withdrawn at the latest by February 2007.

According to the CEN/CENELEC Internal Regulations, the national standards organizations of the following countries are bound to implement this European Standard : Austria, Belgium, Cyprus, Czech Republic, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Iceland, Ireland, Italy, Latvia, Lithuania, Luxembourg, Malta, Netherlands, Norway, Poland, Portugal, Romania, Slovakia, Slovenia, Spain, Sweden, Switzerland and United Kingdom.

Introduction

The European Water Framework Directive (2000/60/EC) has created a need for a uniform procedure to assess ecological quality of surface waters using phytoplankton abundance and composition. This European Standard will meet this need and will help laboratories improve the quality of their analytical results.

A single standard procedure for the assessment of phytoplankton composition and abundance cannot be given as the questions which drive monitoring programmes are diverse in character and therefore require specific protocols. This European Standard, therefore, aims to provide guidance on basic aspects of microscopic algal analyses and to provide statistical procedures for the design, optimization and validation of methods and protocols. Though mentioned in Annex C, a method for the estimation of biovolume is not included.

WARNING — Persons using this European Standard should be familiar with normal laboratory practice. Long periods of microscopic phytoplankton analysis can cause physical fatigue and affect eyesight. Attention should be given to the ergonomics of the microscope and advice from a health and safety practitioner should be sought to ensure that risks are minimized. The use of chemical products mentioned in this European Standard can be hazardous and users should follow guidelines provided by the manufacturers and take necessary specialist advice.

This European Standard 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 health and safety practices and to ensure compliance with any national regulatory guidelines.

1 Scope

The procedure described in this European Standard is based on the standard settling technique as defined by Utermöhl in 1958 [31]. It describes a general procedure for the estimation of abundance and taxonomic composition of marine and freshwater phytoplankton by using inverted light microscopy and sedimentation chambers, including the preceding steps of preservation and storage. Emphasis is placed on optimizing the procedure for the preparation of the microscopic sample. Many of the general principles of the approach described may also be applied to other techniques of enumerating algae (or other entities) using a (conventional) microscope, some of which are described in Annex E. This guidance standard does not cover field collection of samples or the analysis of picoplankton, quantitative analysis of free-floating mats of Cyanobacteria or specific preparation techniques for diatoms.

2 Normative references

Not applicable.

3 Terms and definitions

For the purpose of this document, the following terms and definitions apply.

3.1

accuracy

closeness of agreement between a test result or measurement result and the true value

3.2

algal object

unit/cluster of one or more algal cells encountered during the phytoplankton analysis that is discrete from (liable to settle independently of) other particles in the sample

3.3

detection limit

minimum number and/or size of a specific taxon or group of organisms in a sample at which its presence can be detected with a specified probability

NOTE This definition is analogous to the definition used in chemistry (smallest true value of the measurand which is detectable by the measuring method).

3.4

error

difference between an individual result and the true value

3.5

fixation

protection from disintegration of the morphological structure of organisms

3.6

microscope counting field

delimited area (e.g. a square or grid) in the microscope field of view, used for enumeration

3.7

nanoplankton

small algae between 2 µm and 20 µm in size

3.8
numeric aperture (NA)
difference in refraction index of the medium between objective and object multiplied by the sine of half the angle of incident light

3.9
performance characteristic
characteristics of a specific analysis protocol which encompass qualitative and quantitative aspects for data precision, bias, method sensitivity and range of conditions over which a method yields satisfactory data

3.10
phytoplankton
community of free-living, suspended, mainly photosynthetic organisms in aquatic systems comprising Cyanobacteria and algae

3.11
picoplankton
very small algae between 0,2 µm and 2 µm in size

3.12
precision
closeness of agreement between independent test/measurement results obtained under stipulated conditions

3.13
preservation
process that protects organic substances from decay

3.14
(analysis) protocol
specific analytical procedure concerning (sub)sample volume, magnification, number of cells to count, taxonomic level of identification etc.

3.15
repeatability
precision under repeatability conditions

3.16
repeatability conditions
conditions where independent test/measurement results are obtained with the same method on identical test/measurement items in the same test or measuring facility by the same operator using the same equipment within short intervals of time

NOTE This definition should be interpreted as the error occurring between replicate sub-samples from the same sample, counted using the same counting chamber, performed by one analyst using one microscope in a continuous run on one day.

3.17
reproducibility
precision under reproducibility conditions

3.18
reproducibility conditions
conditions where independent test/measurement results are obtained with the same method on identical test/measurement items in different test or measurement facilities with different operators using different equipment