Guidelines for sampling and determination of nitrate

1 Background

1.1 Introduction

Dissolved inorganic nitrogen is present in seawater both as nitrite, nitrate and ammonium. As a complement to the overall assessment of nutrient status, detailed information on the distribution of different species must be obtained.

1.2 Purpose and aims

Monitoring of nutrients in seawater is carried out to identify and quantify the amount of nutrients, which may cause eutrophication. The aim is to provide spatiotemporal information for detection of short-term status and long-term trends and to ensure that the data is comparable for the HELCOM core indicator 'Dissolved inorganic nitrogen'. The indicator description, including its monitoring requirements, is given in the HELCOM core indicator web site: http://helcom.fi/baltic-sea-trends/indicators/nitrogen-din.

2 Monitoring methods

2.1 Monitoring features

Water samples are collected from discrete depths and analysed. Well established wet chemistry methods are available.

The method most commonly used determines the sum of oxidized nitrogen species (nitrate + nitrite), sometimes referred to as TON or TOxN (Total Oxidixed Nitrogen).

2.2 Time and area

Monitoring of nitrate is carried out by all HELCOM contracting parties, and the monitored area covers the entire Baltic Sea area, both the open sea and coastal areas.

Winter pool of nutrients must be assessed in the surface layer; however, information about the annual cycle in the surface is also important. Furthermore, the vertical distribution has to be considered with respect to oxic/anoxic conditions.

2.3 Monitoring procedure

2.3.1 Monitoring strategy

Water samples are collected from discrete depths, and analysed. Samples need to be analysed or prepared for storage immediately after sampling.

For determination of nitrate, well-established wet chemistry methods are available (see section 2.3.3).

Samples are collected at depths of 1, 5, 10, 15, 20, 25 (Kattegat and the Belt Sea only), 30, 40, 50, 60, 70, 80, 90, 100, 125, 150, 175, 200, 225, 250, 300 and 400 metres; and as close to the bottom as possible.

Colorimetric methods described by Hansen and Koroleff (Grasshoff et al 1999) are considered sufficient.

2.3.2 Sampling method(s) and equipment

For general requirements for sampling, preservation, handling, transport and storage of water samples, see EN ISO 5667-3.

Samples are collected from sampling bottles attached to a CTD rosette, or clamped to a hydrographic wire.

2.3.3 Sample handling and analysis

For general requirements for sampling, preservation, handling, transport and storage of water samples see EN ISO 5667-3.

Subsamples should be collected without unnecessary exposure to air. Rinse bottles with sample water before filling them. Avoid trapping bubbles of air when filling and capping bottles.

Seawater contains microorganisms and other suspended particles, which may have to be removed prior to analysis, since turbidity caused by suspended matter interferes with colorimetric measurements.

Filters used should be free of contaminants, and have an appropriate pore size, e. g. 0.40 µm polycarbonate filters or Whatman GF/F filters. Glass fiber filters, if used, should be combusted at 450°C for at least 4 hours before use.

If samples are not filtered, a turbidity blank can be used to correct for interferences from turbidity or colour.

The procedure selected for removing interference from turbidity must be validated.

Unnecessary handling of samples should be avoided to prevent contamination.

Samples must be protected from airborne contamination from tobacco smoke or engine exhaust fumes.

Samples should be analyzed as soon as possible after sampling, preferably within a few hours. If samples must be stored for longer periods, storage in freezer is necessary to increase stability.

Samples stored in a freezer should first be filtered as described above, and frozen as rapidly as possible. In temperatures between -18 – -20°C, samples are stable for up to four months (temperature should be maintained within the specified interval, and recorded with a digital data logger, e.g. Testo 174, in particular if samples are also used to determine silicate). Before determination if nitrite, samples should be allowed to defrost slowly, preferably overnight, in darkness (experiences from the Laboratory of BSH, Germany).

The colorimetric method described by Hansen and Koroleff (Grasshoff et al 1999) is recommended.

2.4 Data analysis

Results from the method described above is expressed as the sum of nitrite and nitrate; nitrate is calculated as (nitrite + nitrate) – nitrite.

3 Data reporting and storage

Data is reported annually to the HELCOM COMBINE database, hosted by ICES.

4 Quality control

4.1 Quality control of methods

Laboratories carrying out analyses of nutrients should have established a quality management system according to EN ISO/IEC 17025.

Immediate analysis of samples is always preferable to preservation and prolonged storing. If samples are stored in freezer, temperature must be monitored.

Methods for preservation must be validated since results can be affected by biological activity, seasonal cycle, salinity or other matrix effects.

An internal reference material (IRM) should be analysed daily.

Certified reference materials (CRM) are available from VKI/Eurofins:
http://www.eurofins.dk/dk/milj0/vores-ydelser/reference-materialer.

It is strongly recommended that all laboratories participate in interlaboratory comparisons and proficiency testing programs, to provide external verification of laboratory performance. Proficiency testings for nutrients in seawater are provided by e.g. QUASIMEME or SYKE. More proficiency testing schemes are listed at www.eptis.bam.de.

Contracting parties should follow the HELCOM monitoring guideline but minor deviations from this are acceptable if the method achieves comparable results. Validation of the adopted method needs to be performed on the relevant matrix and concentration range e.g. by taking part in intercomparison studies or proficiency testing schemes.

4.2 Quality control of data and reporting

Measurement uncertainty should be estimated using ISO 11352. Estimation should be based on within-laboratory reproducibility, data from proficiency testings, IRM, and, when available, CRM.

Data must be flagged if normal QA routines or recommended storage conditions cannot be followed.

Collected data should be checked for consistency between sampled variables (e. g. dissolved inorganic nitrogen and total nitrogen).

5 Contacts and references

5.1 Contact persons

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5.2 References

Filtration and storage:
Kremling K and Brügman L, Chapter 2 p 27-40

Determination of nutrients:
Hansen H P and Koroleff F, Chapter 10 p 159-228
in
Grasshoff K, Kremling K and Erhardt M. Methods of Seawater Analysis 3rd ed. Wiley-VCH 1999. ISBN 3-527-29589-5

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

EN ISO 5667-9*: Water quality – Sampling – Part 9: Guidance on sampling from marine waters

EN ISO 11352*: Water quality – Estimation of measurement uncertainty based on validation and quality control data

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

*For undated references, the latest edition of the referenced document (including any amendments) applies.

5.3 Additional literature

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Chemical measurements in the Baltic Sea: Guidelines on quality assurance.
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Practical Guidelines for the Analysis of Seawater
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