

Guidelines for monitoring of phytobenthic plant and animal communities in the Baltic Sea

Annex for HELCOM COMBINE programme

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1. Introduction

These guidelines are giving advice on monitoring the changes of phytobenthic communities on hard, soft and mixed substrate bottoms in the Baltic Sea. The phytobenthic communities include the plant and animal communities of the photic zone. Soft bottom animals are covered in HELCOM's macrozoobenthos monitoring.

There is a need for a comprehensive assessment of the state of the phytobenthos and a demand to forecast anthropogenic impacts and its consequences on phytobenthos of the Baltic Sea. Monitoring of the main abiotic and biotic characters of the Baltic Sea phytobenthic zone is especially needed because this zone is productive, sensitive to human impact and thus affected in many areas.

The guideline for monitoring phytobenthic communities in the Baltic Sea has been compiled under the EC MON project CMP. Workshops have been organised and phytobenthos specialists from the Baltic Sea countries have given valuable corrections. The developments in phytobenthos monitoring guidelines of the OSPARCOM and the national phytobenthos monitoring programmes of the Baltic Sea Countries have been followed. The Nordic Council of Ministers financed project (PhytoBios) dealt with the phytobenthos biodiversity monitoring programme for the Baltic Sea and fractions of the PhytoBios programme are implemented in these HELCOM phytobenthos monitoring guidelines.

2. Aims

The strategy is designed to identify environmental changes, mainly the impacts of eutrophication. The programme thus aims to:

- monitor temporal trends (in time scale of years) in floristic and faunistic variables within the maritime area in relation to changes in environmental conditions
- describe the status of floristic and faunistic variables in relation to spatial differences in environmental conditions.

3. Monitoring variables

Core variables form the minimum level of monitoring which every country is encouraged to follow up. Main variables form the second level on which the participating countries should follow. Supporting studies form the third level of monitoring and can be used when the state of the phytobenthos zone is assessed.

The transect method is the most widely used and recommended as a principal method along the Baltic Sea countries. Alternative methods like remote sensing, e.g. aerial photography, video techniques and echo sounders can be used as complimentary methods.

Monitoring along the transects is recommended once a year in a period of June-September, preferably August-September. Within the certain area should be carried out within same month each year.

List of biotic variables for the Baltic phytobenthos monitoring with reference to Core, Main parameters and Supporting studies.

Monitored variables

Site position	Core
transect depth profile	Core
Substrate	Core
Siltation	Core
Non covered substrate	Core
Depth distrib. of important plant species	Core
Composition of plant species	Core
Coverage% of plant species	Core
Algae belt depth distribution	Main
Loose plants	Core
Cover. & depth of <i>Mytilus edulis</i>	Main
Monitoring of important species	
<i>Fucus vesiculosus</i>	Supporting studies
<i>Zostera marina</i>	Supporting studies
<i>Phragmites australis</i>	Supporting studies
Depth distribution of animals	Supporting studies
Coverage% of animal species	Supporting studies
Animal species composition	Supporting studies
Biomass of vegetation	Supporting studies
Biomass of bottom fauna	Supporting studies
Area distr. of animal species	Supporting studies
*Temperature	Core
*Water transparency (Secchi depth or light meter data)	Core
*Salinity	Core
*Nutrients in water	Main
*Particular organic matter (POM)	Supporting studies
*Dissolved organic matter (DOM)	Supporting studies
*Dissolved oxygen	Supporting studies
Water level from nearest mareograph	
Cartographic exposition value	

*Water hydrography and chemical variables preferably are measured in the framework of other parts of the

coastal monitoring programme.

4. Data storage, analyse and reporting

The database and electronic reporting formats for phytobenthos monitoring will be developed by HELCOM consult ICES in cooperation with CMP. In the present situation the national databases are kept in the countries.

These methods are based on the expertise from the national programmes. In monitoring design three transects are located on each sampling site. This should cover the spatial heterogeneity. The higher number of transect within an area might strengthen the statistical significance. As in HELCOM fish monitoring, higher number of transects should be located and monitored within the area and monitored over the years and reduce the number after statistical analysis. However, three transects should allow us to use the data for analysing the inter-year variation in monitored variables. The data can be used in trend analysis and analysis of variance. To study differences between the years Mann-Whitney U-test or Kruskal-Wallis test can be used. Differences in species composition can be analysed by calculating β -diversity index. However, the monitoring programme should be evaluated after few years to check the statistical validity.

5. Quality assurance (QA)

These guidelines for monitoring the phytobenthos of the Baltic Sea are one of the first attempts to standardize overall phytobenthic sampling by focussing on QA with definition of objectives, study design, field sampling sample processing, data analyses and reporting. The quality assurance should be integrated into the monitoring programme from the beginning and it should be presented at different levels of the programme. The implementation of quality control programmes must include sampling and analytical procedures, the staff conducting the field work, the interpretation of results as well as the development and maintenance of data bases.

The common practice is that QA procedures are developed by expert groups and then evaluated at SGQAB. The QA procedures form a part of the monitoring activities and have to be established before the actual monitoring can start. These guidelines need to be revised in future years.

6. *In situ* methods

The method of *in situ* diving transects has been widely applied in phytobenthos monitoring mainly for vegetation analysis and somewhat less for faunal studies. Also monitoring the transect by using either a diver operating or ship-towed underwater-video systems are acceptable. SCUBA diving has been used for vegetation analysis, for species composition, percentage cover and for determining algal zonation. The species composition of the associated animals has been studied from quantitative samples taken by a diver.

6.1 Location of the transects

The monitoring transects have to be representative for the larger area according to the bottom morphology and benthic communities as well as environmental factors influencing the development of the phytobenthic communities.

Monitoring activities are based on the transects covering the depth interval from the water edge to the lower limit of distribution of a phytobenthic zone. In large areas without depth gradient the detailed investigations can be carried out in sampling points (shorter interception) along the transects.

The location of transects could be studied on nautical charts or geological maps to evaluate their distribution. A set of transects (minimum 3) are recommended to be located on the representative habitat in the monitoring area. Habitat could be described as soft and sandy shores, rocky shores on exposed or sheltered shores. A set of transects should be placed on the shores with similar exposure and substrate type. The substrate of the transect does not have to be homogenous all the way. It could start with hard substrate or have patches of hard substrate and end with mixed, sandy and soft substrates. The transects are normally oriented at a right angle to the coastline.

6.2 Establishment and documentation of the transect

The water level during the period of the monitoring is documented from the nearest mareography.

The transects are marked and on the map (scale 1:50 000). To locate the same transects from year to year preferably use differential dGPS for end and start positions. This is relevant especially in case if performing an interrupted (point) transects. The permanent marking can be done, e.g. by drawing a 50 cm white strip on the rock or by drilling a hole. The marking must be situated high enough above the highest water level to avoid ice scraping. Also below the water surface the transects can be marked e.g. bottom anchored lines or marked on brick stones and given intervals.

The location of the transect on the shore, as well as on the sea bottom should be documented by photographing. The length of the transect depends on the morphology of the bottom and the depth penetration of vegetation. If possible and needed a shrink proof line, marked at every metre, is first attached to the starting point, then laid down on the sea bottom following the chosen compass course.

Before monitoring, time, date, exact dGPS location, depth and name of investigator and other responsible person are recorded in the field form. The field form (protocol) used, are copied on water resistant plastic and attached to a writing plate.

The necessary field information for the individual transects/point from each sampling date should be noted in a field form as follows

Transect number and Date:
Method applied (SCUBA/Video)
Name of investigator
Nearest coastal monitoring station
Type of sampling 1)
Start distance from land
Start position 2)
End position 3)
Location type 4)
Description 5)

- 1) Type of sampling: transect/point
- 2) Start positions: longitude/latitude
- 3) End position: longitude/latitude
- 4) Location type: fjord/bay/sound/enclosed shallow area/open shallow areas/open area/rocky coast/reef
- 5) Description: description of the transect/point or vegetation and weather information, turbidity.

6.3 Depth profile of the transect

Along a short rocky transect where a line is laid out, the water depth is measured for every linemeter of using a calibrated diver's depth gauge.

In large shallow areas, the use of marked line is not possible due to the length of the transects. The transect can be divided into sections of 30 - 40 m long, located along the transect after certain depth interval (1-2 m). The depth profile of the transect may be recorded by echo sounder beforehand using the boat operated echo sounder and dGPS. If dGPS is not available, the depth profile could be generated by recording the depth values from an echo sounder in certain time interval (e.g. 10 sec.) while moving along the transect with constant speed (1-2 knots). The depth profile of the transect has to be established at the first time a transect is visited and set up and later on it could be used each time the transect is visited.

6.4 Substrate composition in depth intervals

The substrate is categorised based on their physical properties that enable ratings of substrate firmness (hard - soft). A priori set assumptions of substrate and type of habitat could be made with the aid of cartographic work or by using previous mapping studies.

The percentage cover of the individual substrate types should be estimated for each depth interval. The percentage of a hard substrate suitable for colonization by perennial macro algae should be estimated for each depth interval. Similarly the overall percentage of soft/sandy bottom should be estimated for each depth interval.

Categories used for description of sediment composition.

1. Rock
2. Boulders >30 cm
3. Stones >10 cm
4. Gravel
5. Sand
6. Sand (firm bottom)
7. Soft substrate
8. Shells
9. Common mussel (<i>Mytilus edulis</i>)
10. Others (peat, boulder clay, mud, silt)

Then the character of dominating substrate is described with proportion of a different substrate in relation to a depth interval.

Example:

Depth one m: rock bottom 70% and stones 30%, 1/70, 3/30
Depth two m: rock bottom 70% and stones 20% and sand 10%, 1/70, 3/20, 5/10
Depth three m: rock bottom 50% and sand 50%, 1/50, 5/50
Depth four m: Sandy bottom 95% and stones 5%, 5/95, 5/5

A sediment composition should be expressed as percentage of the category in relation to the depth interval. In the field the actual percentage numbers could be expressed and later transformed in the same categories as used in vegetation coverage estimation. In the field form, a substrate composition of 70% rock bottom and 30% stones can be written as 1/70, 3/30

6.4 Siltation

The amount of loose sediment on the bottom and on the vegetation should be estimated by using the following scale: (1) none, (2) moderately that quickly settles back, (3) much, (4) very much, e.g. on rocky bottom algae cannot grow epilithically.

6.5 Surveys along transects

Monitoring of benthic vegetation along transects is based on visual observations or videoing by Scuba diver or ship-towed video system. This is carried out from the coastline to the maximum depth with benthic vegetation if possible. The measurements are related to depth intervals, e.g. 0-1 m, 1-2 m, 2-4 m, 4-6 m etc. with a possibility of subdivision of the intervals. The lower limit of the scale can be extended by two m intervals to cover the maximum depth of the vegetation. The lower limit is when macrovegetation disappears and on hard substrates when *Mytilus edulis* disappears.

Information on the substrate is also registered by the diver or ship-towed video system in each depth interval simultaneously with the detailed investigation of the vegetation. The process of vegetation surveys along transects or in points includes various steps which will be specified in the following paragraphs. The paragraph also specifies the necessary field information to be collected and noted in a field form.

6.6 Vegetation coverage in the depth intervals

For each depth interval, the coverage percentage of the overall vegetation is estimated for soft/sandy and hard bottom, respectively. The coverage of the vegetation is estimated by projecting the outline of the shoots perpendicularly to the seabed.

Coverage of individual species is also estimated by projecting the outline of the shoots perpendicularly to the seabed. Coverage of soft bottom species is estimated relative to the soft bottom while coverage of attached macro algae should be estimated relative to the suitable hard substrate. As species occur in layers, epiphytes, large algae and phanerogams, bottom layer crustose algae, the total coverage may well exceed 100%.

Coverage of the vegetation and species is estimated percentage number which can be converted to the scale 1-6 afterwards.

Description of the total vegetation coverage

% vegetation coverage/ suitable hard substrate
% vegetation coverage / soft-sandy substrate

Species coverage in individual depth intervals

	Coverage %					
Species	0-1.0 m	1-2 m	2-4 m	4-6 m	6-8 m	8-10m...
Species a						
Species b						
Species c						
-						
-						

Coverage scale

- 1 Species registered without coverage
- 2 <2%
- 3 2 -<10%
- 4 10-<25%
- 5 25-<50 %
- 6 50-<75%
- 7 75-<100%

Estimation of the coverage of the vegetation could be carried out by various methods:

a) *Average estimations of coverage in a depth interval*

This is a subjective, average estimate of the coverage of the vegetation in the depth interval. The observations should be done in a corridor of about five m width at both sides of the transect. If the transect is extremely long, the coverage of the vegetation can be estimated in various points along each depth interval.

Within each depth interval coverage of the vegetation could be analysed within a frame. The squares are located randomly. Preferably in each depth interval one square is located. The frame should be located next to the line on the left side, viewed from the shore.

b) *Continuous estimations of coverage*

The starting distance from shore and the depth read from calibrated depth gauges, and the substrate need to be noted, then the estimations of the coverages are done towards the shore. Coverage estimations are made along the section of transect by estimating the coverage in the corridor of 3-5 m width on both sides of the transect line. As soon as any new substrate or species occur in the corridor or changes of cover degree, depth and the new cover degree of the species are noted. The process is repeated until the shore is reached. The occurrence of first and last individual occurring along the line and at which depths the species has its maximum cover degree. During the process of estimating the cover degree, photographs of the communities are taken preferably as "view-pictures" more horizontally.

6.7 Loose plants

They are fast-growing annual algae like, *Cladophora* spp., *Enteromorpha* spp., *Ulva lactuca*, *Ectocarpus* spp., *Pilayella littoralis*, *Ceramium* spp and an occurrence of such mat should always be estimated for each depth interval. These algae can accumulate as dense layers on the seabed. The total % coverage of this group should be noted in the field form. Important variables to record are, e.g. extension and thickness of the mats, dominating species and depth range.

6.8 Algal belt depth distribution

The upper and lower limits of the dominant vegetation belts (e.g. green algae, *Fucus vesiculosus*, red algae or belt below *Fucus vesiculosus*) are noted. If separate algal belts can be distinguished, it is advisable to analyse the coverage values within the frame.

6.9 Depth distribution

The lower depth limit can be divided into a main depth limit and a maximum depth limit. The main depth limit is defined as the depth where the number of shoots per area is reduced and it can normally easily be defined within 0.2 m depth. The maximum depth limit is the absolute depth limit of individual shoots. For eelgrass both the main and the maximum depth limit should be determined. Maximum depth of the area should also be noted in the field form for comparison with the depth limits of the vegetation. In addition it should be noted whether the depth limit is set by lack of a suitable substrate. The parameters to be collected in the field are listed.

Main depth limit of species
Maximum depth limit of species

Depth limit set by substrate (yes/no)

Preliminary list of species which depth range should be recorded accurately.

Fucus vesiculosus, *Fucus serratus*, *Delesseria*, *Laminaria*, *Cladophora* ssp., *Pilayella/Ectocarpus*, *Furcellaria lumbricalis*, *Sphacelaria arctica*, *Phyllophora* ssp., *Rhodomela confervoides*, *Chara* ssp., *Tolypella nidifica*, *Zostera marina*, *Potamogeton* ssp., *Zannichellia palustris*

6.10 Species composition

The identification of the species is sometimes difficult under the water thus quantitative samples can be collected to obtain algae species composition of a given depth. To obtain a precise species list along the depth interval the use of small frames, e.g. 0.2m x 0.2m of size can be used for sampling. For species composition and species which are difficult to identify under water, samples can be collected into individually numbered bags and then species can be identified later.

7. Quantitative samples

The quantitative samples are collected to obtain species composition and biomass of a given depth. Within each identified belt established through the line transect estimates, at least three frames are located. Location of sampling frames is fixed in advantage. This can be a certain distance from the shore. Samples are collected along with the estimates of cover degree of species along the line transect. Data collected is as follows:

- Distance from shore of the sampling site
- Depth of the sampling site
- Type of frame (size)
- Type of substrate within the frame
- Dominating species composition within the frame
- Cover degree of dominant species within the frame
- Species biomass of each species separately

Determination of sampling sites. The site of sampling is determined using either information of previous years, distance from shore and depth or the notes done during the line transect. If samples have to be placed for the first time, an experienced diver must observe of the position (e.g. distance from the shore) chosen and any bias in her or his choice of direction she or he drops the frame.

The intercept of the belt is extracted from the line transect protocol. Within each vegetation belt, frames are placed at random, e.g. if the *Fucus*-belt of 100 to 50% cover extends from 5 m to 30 m of the line, any random number within 5 and 30 is accepted.

Choice of frame size. For sampling, frames of 0.2 to 0.5 m can be used.

Protocol of a frame sample. The frame is placed on the sampling site which is determined on exact distance from shore. The substrate and the cover degree of the species within the frame are noted on the protocol. Each frame has its unique number which is noted.

Handling of quantitative samples. All plants and animals within each frame are collected in a net bag. The bag and frame are brought to the boat. Into each net bag an information sheet with the date, names of the station, distance from shore, depth and contents (short) and frame number is included. The sample is

transferred into a plastic bag for deep freezing for preservation in fixative. In the case of abundant stones and/or fine sand and silt, the sample can be sieved before it is transferred into the plastic bag. A sieve of mesh size not larger than 0.5 mm can be used.

Sorting, handling and protocol of quantitative samples. When necessary, the sample is sieved, sorted to species of plants and animals separately. Each species is counted (animals) and dried on aluminium foil separately. The aluminium foil has been weighted in accuracy of .001 g. After drying in 60°C to constant weight (about 2-3 weeks), samples are weighted and the dry weight per m² is calculated.

8. Photo documentation

The photographic picture of the plant and animal communities has a major information value if each photo is properly documented. The vertical picture is used as a tool for the estimation of species composition and cover degree within a given frame (the picture size). However, vertical pictures only show a minor part of the substrate.

Another approach would be to document the environment as seen by the diver, i.e. more horizontally taken pictures. These pictures give a good overview of the area. The pictures have to be documented by stating distance and depth in the protocol. If any conspicuous object is included in the picture, the same spot can be repeatedly photographed and comparisons between years can be done. Conspicuous items could be a crack in the rock wall, a large boulder in the background, or deliberately placed objects, e.g. brick stones, on the sea floor at the picture point.

9. Video method

The ship-towed video system is especially useful for transects that are too long for an examination by SCUBA divers. The method provides records of the complete transect, so that a detailed investigation of vegetation and substrate can be carried out. Normally, the system consists of a video-camera, mounted in a sledge e.g. tripod with a fin, and connected to the ship with a cable that transfers the pictures to a video-recorder. The camera is towed by the ship at a constant speed of 1-2 knots and at a constant distance to the ground over the entire length of the transect to film the sea bottom. The ship is following the transect, beginning at the shallowest point in a direction away from the coastline towards the greater depths. Thus the handling of the system on small vessels without crane is easier because it has not to be lifted to keep the defined distance to the ground.

The camera system should be connected to the nautical system on board in a way that dGPS-position and depth values together with date, time, name of the transect and additional remarks are faded in into the recording (corresponding systems are produced e.g. by MARISCOPE at Kiel/Germany). By this method, the exact geographical position of every recorded picture can be determined at any time. This system facilitates the evaluation of the tapes in combination with the recording of data on a personal computer. If necessary, hydrographical probes, mounted on the sledge, can be connected to the system in the same way as the nautical system.

As an alternative, nautical and other data can be noted in a protocol. In this case it is very important to note the tape counter and/or elapsed time together with the nautical and other data. Evaluation of the tapes is not possible without the protocols. The pictures should be recorded by video recorders with the specification S-VHS, Hi8 or digitally, if the tapes are to be copied, to avoid losses of quality. If the evaluation is carried out with the original tapes, VHS is sufficient.

For the evaluation a depth profile in correlation to the covered distance can be displayed. On the depth profile

the evaluated data from the tapes can be placed. The tapes are displayed in slow motion, changes in covering rates of macrophyte species as well as substrate type and quality are noted according to the coverage scales as explained in chapter 6.6 resp. 6.4.

Depending on the quality of the used camera system, the following parameters can be obtained:

a) coverage of:

-*Zostera marina*, *Laminaria saccharina*, *Fucus* spp., Red algae (determination of some species is possible), other brown algae, green algae, loose algae/plants, *Mytilus edulis*
-sediment composition.

In the cases that the connection of video-tapes on the ship is impossible a SCUBA diver can operate a camera under water. Nautical and other data from the navigation-system, composition of species and substrate with regard to depth, location and other data is only possible with difficulties. For the purpose of documentation, however, this method can be very useful, because in contrast to photographs it provides a continuous overview on the conditions that occur on the transect.

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