

Manual for Marine Monitoring in the

COMBINE

Programme of HELCOM

Part C

**Programme for monitoring
of **eutrophication**
and its effects**



Last updated: 31.03.2006

PART C PROGRAMME FOR MONITORING OF EUTROPHICATION AND ITS EFFECTS

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C.1. INTRODUCTION

The programme on eutrophication and its effects considers short and long term variations in hydrographic conditions and in chemical and biological variable.

More specifically the aims of COMBINE mean for:

Hydrographic variations:

aim: to set the background for all other measurements related to the identification and quantification of the effects of anthropogenic discharges/activities, the variables providing an indication of natural fluctuations in the hydrographic regime of the Baltic Sea must be monitored on a continuous basis

Core variables:

- temperature, salinity, oxygen and hydrogen sulphide
- light attenuation

Main variables:

- current speed and direction

PROBLEMS RELATED TO EUTROPHICATION (CHEMICAL AND BIOLOGICAL VARIABLES):

aim: to determine the extent and the effects of anthropogenic inputs of nutrients and organic matter on marine biota, the following variables must be measured:

A) CONCENTRATIONS OF NUTRIENTS

Core variables:

phosphate, total phosphorus, ammonia, nitrite, nitrate, total nitrogen and silicate, to quantify the changes in the nutrient pool. In CMP nitrate and nitrite may be measured together.

Main variables (In CMP supporting studies):

- Particulate and dissolved matter (carbon, nitrogen and phosphorus). These parameters are all essential for budget calculations and the contracting parties are recommended to include these in their programmes in all areas.
- Humic matter is an important source of nutrients in the Baltic Sea, especially in the Gulf of Bothnia and in its estuaries and should be incorporated into the programme there.

B) THE RESPONSE OF THE DIFFERENT BIOLOGICAL COMPARTMENTS:

Core variables:

- chlorophyll- α , as an equivalent of the standing stock of phytoplankton;
- phytoplankton species composition abundance and biomass, to indicate a response in the biodiversity and a possible change in the food chain composition (e.g., introduction of alien species)

or increase in toxic species that are harmful to other organisms, and to indicate changes in the stock of primary producers;

- zoobenthos species composition, abundance and biomass (increase in biomass indicates eutrophication) and species composition (reduced species diversity). Excessive levels of eutrophication can result in low concentrations of oxygen in the bottom waters, resulting in damage to or death of zoobenthos.

Main variables (In CMP supporting studies, except zooplankton and phytobenthos):

- to measure the change in the rate of production, i.e. the first response of phytoplankton to the nutrient loading;
- zooplankton species composition, abundance and biomass, as changes can result, e.g. from changes in phytoplankton biomass and species composition. Especially in coastal waters zooplankton indicates different water masses, salinity fronts and other hydrological events.
- sinking rate of particulate matter;
- vertical profiles of chlorophyll a fluorescence, to give detailed information on vertical distribution of phytoplankton;
- phytobenthos, response to light climate and nutrient concentration results in depth distribution and species composition.

Supporting studies:

- Bacterial numbers and production are important in the cycling of nutrients in the Baltic Sea ecosystem. Especially in the Gulf of Bothnia, the role of bacteria is of major importance in the energy cycle, since the ratio of pelagic primary production to inputs of allochthonic organic matter is high. At least these bacteria should be a part of the high frequency sampling programme. However, bacteria are also of major importance in other areas of the Baltic Sea.
- Semi-quantitative analysis of phytoplankton can be used in addition to quantitative analysis to reveal temporal and spatial changes in phytoplankton communities.
- Microzooplankton plays a dominant role in certain shallow regions, and gives additional information on the functioning of the ecosystem.
- satellite imagery, as a tool for monitoring the spatial distribution of phytoplankton biomass in the surface layer, especially the accumulations of blue-green algae;
- annual primary production studies: important in assessing the changes in cycling of organic matter;
- fast repetition fluorometry, to record primary productivity with high resolution;
- flow cytometry, to describe the plankton community with an automatic method;

- HPLC pigment analysis, to get fast information of the phytoplankton pigment composition as indicator of the taxonomical composition;
- grain size distribution of sediment in relation to studies of macrozoobenthos;
- denitrification and nitrogen fixation, to describe the processes in the biological nitrogen cycle.

C) INTEGRATION AND EVALUATION OF RESULTS:

* Numerical and statistical models: It is essential that different kinds of models become part of the monitoring system, on equal terms with actual field measurements. The use of models also provides an opportunity to test the reliability of data. There are several uses of models;

- Real-time evaluations: if the monitoring should function as some kind of early-warning-system it is only with models in connection with measurements that we can assess the real time conditions.
- Budget calculations: models are necessary when interpolating/extrapolating measured data and are thus indispensable when making budget calculations.

An assessment of the results from the programme should be able to detect regional trends in hydrographical parameters, in nutrient concentrations, in phyto-, mesozooplankton, phytobenthos and macrozoobenthos abundance and species composition (where potentially toxic and/or alien species should be of particular concern) and in oxygen/hydrogen sulphide concentrations. For the assessment of the eutrophication status it is also important that the programme can resolve anthropogenic and climatological effects.

In order to meet the requirements of the strategy identified, the programme for the open sea, within each separate sub-basin, must be able to account for:

1. the winter pool of nutrients,
2. annual cycles of hydrographical parameters,
3. regional distribution and long-term changes in phyto- and zooplankton populations,
4. the spatial distribution of oxygen/hydrogen sulphide concentrations in the bottom water (in critical areas, especially during late summer/autumn),
5. spatial and long-term variability of macrozoobenthos,
6. occurrence of alien species which might have marked effects on the ecosystem,
7. events (e.g. toxic algal blooms) of importance for human health, recreational values or other economically important sectors, and
8. water exchange and nutrient fluxes between the Baltic Sea basins and between the Baltic Sea and the North Sea

C.2. SAMPLING STATIONS

To be able to fulfil these requirements, the programme should at least consist of:

- mapping of the winter pool of nutrients at least once per year before the onset of the phytoplankton growth period;
- mapping of oxygen/hydrogen sulphide and nutrient conditions in the near bottom waters a few times per year. It is important that this is carried out in late summer or autumn in certain critical areas.
- mapping of zoobenthos at least once a year;
- high frequency sampling which is needed especially for the pelagic variables and for monitoring water exchange between the various basins and between the Baltic Sea and the North Sea. This is obtained by visiting selected open sea or coastal stations frequently (preferably weekly measurements during the vegetative period), by using ships-of-opportunity sampling and automatic fixed stations. Automatic fixed stations are also needed for measurement of sinking rate of particulate matter.

Thus the COMBINE programmes comprises mapping stations and high-frequency stations. The BMP sampling stations presented by the Contracting Parties are shown in Figs. A.1-A.10 and Annex C-1.

MAPPING STATIONS

1. HYDROGRAPHY AND NUTRIENTS:

The choice of stations during mapping surveys should be governed by the objectives of the survey, except that the frequent stations in each region always should be included in a mapping. Consequently, a fixed network of mapping stations is not considered since the need will vary due to varying physical/biological/chemical conditions. However, the objectives with the different mapping surveys should be identified and clearly stated.

Sampling frequency:

A few times per year:

- mapping the winter pool of nutrients
- mapping the oxygen/H₂S conditions, particularly in critical areas and season (e.g. the late summer/autumn).

Core variables:

- temperature and salinity
- O₂ and H₂S

- PO₄ and Tot-P
- NO₂, NO₃, NH₄ and Tot-N
- SiO₂

2. MACROZOOBENTHOS

For studies of spatial and long term variations in macrozoobenthos, abundance biomass and species composition.

Sampling frequency:

Once or few times per year;

Core variables:

macrozoobenthos

Main variables:

- temperature and salinity,
- O₂ and H₂S in the near-bottom water
- weight-loss of ignition,
- smell (H₂S),
- depth of oxygenated layer in the sediment

Note: grain-size is listed on p. 2 of Part C

HIGH FREQUENCY SAMPLING

1. CRUISE STATIONS

Sampling frequency on sample stations should be >12 times per year (basically monthly sampling but weekly in the vegetative period)

Core variables:

- temperature and salinity
- O₂ and H₂S
- PO₄ and Tot-P
- NO₂, NO₃, NH₄ and Tot-N
- SiO₂

- Chlorophyll-*a*
- Phytoplankton

Main variables:

- Primary production
- pH and alkalinity
- Zooplankton

2. SHIP-OF-OPPORTUNITY SAMPLING

Unattended recording and sampling on ferries and other commercial ships with regular schedules gives a possibility to collect data with high temporal and spatial resolution in the surface layer of the sea with large spatial extent. These kinds of measurements supply information important especially for the real time monitoring, and early warning system of, e.g. toxic algal blooms, and can also serve as reference and calibration for satellite images.

Sampling frequency:

The sampling frequency should be about every 200 m and every 1-3 days for temperature, salinity and chlorophyll *a* fluorescence. For phytoplankton and nutrients about every 10 km and every 1 - 3 weeks.

Core variables:

- Temperature and salinity
- chlorophyll *a*
- PO₄ and Tot-P
- NO₂, NO₃, NH₄ and Tot-N
- SiO₂
- phytoplankton

3. AUTOMATIC FIXED STATIONS:

These stations make it possible to collect high frequency data on temperature, salinity, oxygen, light attenuation and current speed/direction. Data from such stations are essential in frontal areas as e.g. the Belt Sea for evaluation of the water exchange. These stations also give access to real time data as input to numerical models (dispersion models) and are thus an important part of a system giving on-line information on certain events (e.g. inflows of North Sea water, potentially toxic algal blooms, oil spill accidents). Automatic stations with high sampling frequency will also improve our understanding of the dynamics of the marine system. High-frequency sample stations should be located close to the fixed stations.

Sampling frequency:

Temporal sampling frequency range between minutes and hours (days and weeks for the sinking rate of particles)

Core variables:

Temperature and salinity

Main variables:

- current velocity and direction
- sinking rate of particles

C.3. SAMPLING PROGRAMME AS COMMITTED BY THE CONTRACTING PARTIES

DENMARK

The preliminary Danish marine monitoring programme consists of:

- 2 automatic stations to record current speed and direction as well as temperature and salinity;
- 7 high-frequency hydrography/hydrochemistry stations where measurements are made annually 30-47 times. Additionally 4 high frequent stations are temporarily established in the Sound area as part of the control monitoring programme for the construction of the link across the Sound;
- 3 high frequency pelagic biology stations (annual sampling 26 times). Plus 1 frequent station (BMP-K2) in the Bornholm Basin;
- 27 mapping stations: the existing BMP hydrography/hydrochemistry stations in the Kattegat, Sound and the Belt Sea already included in the Danish monitoring programme (15 st.), 2 BMP-stations in the Kiel and Mecklenburg bights, respectively. Plus some national stations (10 st.). Mapping of winter nutrients in February (1 cruise). Mapping of oxygen each month August-November (4 cruises). The cruises will be coordinated with Sweden and Germany. At all cruises and stations hydrography, hydrochemistry, oxygen and chlorophyll-*a* will be measured.

ESTONIA

- January (or February) - 30 stations covering whole area; measured variables: nutrient concentrations (PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si), temperature, salinity, Secchi depth, O₂ (or H₂S), chlorophyll-*a*;

- June - 20 stations, measured variables: macro-zoobenthos, temperature, salinity, Secchi depth, O₂, nutrient concentrations (PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si) and chlorophyll-*a*;
- October-April once a month, May-September every second week - 7 stations covering 2 high-frequent areas, measured variables: temperature, salinity, Secchi depth, O₂, nutrient concentrations (PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si), chlorophyll-*a*, primary production, phytoplankton (species composition and semi-quantitative abundance), zooplankton (biomass and species composition) and colony-forming bacterioplankton.
- August - phytobenthos observations at chosen transects in each high-frequent area and at additional reference areas.

FINLAND

The Finnish BMP programme includes:

- large number of stations with low sampling frequency (normally once a year) to map the winter pool of nutrients and the oxygen conditions in the near bottom water. The number and the positions of the mapping stations may vary slightly from year to year. The variables are temperature, salinity, Secchi depth, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N and SiO₂-S;
- large number of fixed stations for one annual (May-June) macrozoobenthos sampling including basic hydrography. The number of stations may vary slightly from year to year;
- high frequency sampling using ship-of-opportunity technique for temperature, salinity, chlorophyll-*a*, phytoplankton species composition and their semi-quantitative abundance as well as for PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, and SiO₂-Si. Additionally, phytoplankton is determined quantitatively with lower frequency.
- satellite imagery to monitor the extent of the blue green algal blooms;
- several fixed near coastal stations in each sub-basin with a sampling frequency of ca 20 times per year. The variables are temperature, salinity, turbidity, colour, pH, O₂, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si, alkalinity and chlorophyll-*a*;
- about 100 near coastal or coastal mapping stations where temperature, salinity, turbidity, colour, pH, O₂, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si, alkalinity are measured in March and July-August and chlorophyll-*a* in July-August.

GERMANY

The German programme for monitoring eutrophication and its effects includes:

A. fixed sampling stations in the open sea for measuring:

- nutrients and oxygen conditions. The variables are temperature, salinity, Secchi depth or light attenuation, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N and SiO₂-Si. PH and dissolved as well as particulate carbon and nitrogen are supplementary variables.
- the pelagic biology variables chlorophyll-*a*, phytoplankton species composition, abundance and biomass as well as mesozooplankton species composition and abundance.
- the macrozoobenthos variables species composition, abundance and biomass.
- the sinking rate of particulate matter with automated sediment traps.
- hydrographic variables temperature, salinity, O₂ and current speed and direction at autonomous mooring stations.

B. a larger number of fixed near coastal sampling stations for measuring:

- nutrients and oxygen. The variables are temperature, salinity, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N and SiO₂-Si.
- the pelagic biology variables chlorophyll-*a*, phytoplankton species composition, abundance and biomass.
- the macrozoobenthos variables species composition, abundance and biomass.

C. supporting studies to develop novel, efficient monitoring techniques at selected stations for:

- HPLC determination of pigments, particle counting by flow cytometry and shipborne bio-optical and video techniques for use in phytoplankton and benthos analyses in the open sea.
- autonomous nutrient measurements at different depths at one of the mooring stations (FB).
- phytobenthos investigations along the coastline on selected transects.

Detailed information about locations and sampling frequencies of investigations under A and B are given in Annex C-1.

LATVIA

The Latvian marine monitoring programme for monitoring the eutrophication and its effects includes:

The Gulf of Riga

- *mapping stations*
 1. winter pool of nutrients - 7 stations once in February.
 2. oxygen/hydrogen sulphide and nutrient conditions - 12 stations once in August. Measured variables are: temperature, salinity, Secchi depth, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si.

3. pelagic biology - 4 stations once in August. Variables are: chlorophyll-a, phytoplankton (species composition, abundance, biomass), mesozooplankton (species composition, abundance, biomass).
 4. macrozoobenthos species composition, abundance and biomass - 19 stations once in August.
- *frequent stations*
 1. hydrography and nutrients - 9 stations sampled 6-9 times per year (February - November). Measured variables are: temperature, salinity, Secchi depth, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si.
 2. pelagic biology - 7 stations sampled 7-8 times per year (February - November). Variables are: chlorophyll-a, phytoplankton (species composition, abundance, biomass), mesozooplankton (species composition, abundance, biomass), bacterioplankton.
 - *high-frequency stations*
 1. 2 stations sampled 20-21 times per year (February - December). Variables measured are: temperature, salinity, Secchi depth, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si; chlorophyll-a, phytoplankton (species composition, abundance, biomass), mesozooplankton (species composition, abundance, biomass), bacterioplankton (1 station).

Eastern Gotland Basin

- *mapping stations*
 1. hydrography, nutrients, oxygen/hydrogen sulphide - 7 stations sampled 3 times per year (February, May, August). Measured variables are: temperature, salinity, Secchi depth, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si.
 2. pelagic biology - 4 stations sampled 3 times per year. Variables are: chlorophyll-a, phytoplankton (species composition, abundance, biomass), mesozooplankton (species composition, abundance, biomass).
 3. macrozoobenthos species composition, abundance and biomass - 13 stations once in August.
- *frequent stations*
 1. hydrography, nutrients, pelagic biology - 6 stations sampled 5 times per year (May - September). Measured variables are: temperature, salinity, Secchi depth, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si; chlorophyll-a, bacterioplankton.

LITHUANIA

In the BMP Lithuania will investigate hydrography, hydrochemistry and hydrobiology as follows:

- 4 BMP stations (J1, J2, K1, L1) and 10 open sea (deep water) stations (46, 46a, 2c, 64a, 5b, 5c, 6b, 6c, D6, 43); sampling 4 times per year,

- 15 coastal zone stations (1, 1b, 2, 2b, 3, 4, 4c, 16, 64, 5, 6, 7, 20, 20a, 20b); sampling frequency 6 times per year,
- 3 "hot spot" stations (1K, 4K, 7K); sampling frequency 16 times per year

POLAND

The Polish monitoring programme comprises of the following measurements:

In the hydrological programme the variables are:

- water temperature and salinity, Secchi depth, O₂, H₂S, and sea currents

In the hydrochemical programme the variables are:

- PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si

The biological programme comprises microbiology (in the coastal zone), chlorophyll-*a*, primary production, phyto- and zooplankton species composition, abundance and biomass, zoobenthos species composition, abundance and biomass and fish species composition, size distribution and diseases in selected area of the coastal zone.

The Polish monitoring programme is to be carried out on the basis of the following number of the stations:

Open sea stations (sampling at least 6 times per year, except macrozoobenthos - once a year), including:

- - hydrology, hydrochemistry - 4 stations
- - biology (pelagic and benthic) - 3 stations

22 coastal stations including:

- hydrology and hydrochemistry - 21 stations
- microbiology - 10 stations, 2 times a year
- pelagic biology - 12 stations, 4 times per year
- macrozoobenthos - 5 stations, once a year
- macrophytobenthos - 4 stations, 2 times per year
- 4 high frequency stations including hydrology, hydrochemistry and pelagic biology - 12 times a year

In the Polish programme reference points for each sampling area have been defined. Three stations (SK, L7, R4) are located within the identified BSPA areas while two other (ZP 6, P 102) lay close to the BSPA.

RUSSIA

[Information is missing.]

SWEDEN

The Swedish proposal for the BMP comprises

- 52 stations with low sampling frequency (1-2 times per year) to map the winter pool of nutrients and the oxygen conditions in the near bottom water, especially in late summer or autumn in the Kattegat, the Arkona, Bornholm and Gotland basins. The variables are temperature, salinity, Secchi depth, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N and SiO₂-Si;
- 19 stations with a sampling frequency of at least 12 times per year. The variables are temperature, salinity, Secchi depth, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N and SiO₂-Si. At a subset of stations alkalinity, pH, chlorophyll, humic matter and phytoplankton abundance and biomass will be measured;
- 5 (2 coastal and 3 open sea) stations with high sampling frequency (20-30 times per year) with weekly sampling during the vegetative period. The variables are temperature, salinity, Secchi depth, O₂, H₂S, PO₄-P, tot-P, NO₂-N, NO₃-N, NH₄-N, tot-N, SiO₂-Si, alkalinity, pH, chlorophyll a, phytoplankton abundance and biomass, primary production. Zooplankton should be a supplementary variable at the high sampling frequency stations. In addition, supplementary variables are included depending of the local needs. These include particulate and dissolved carbon and nitrogen, bacteria, sedimentation and humic substances;
- one automated buoy station to monitor fluxes of water, salt, and nutrients between the Baltic Sea and the Skagerrak (North Sea);
- 139 soft bottom macrofauna stations are collected annually (May-June) from off-shore areas and in the coastal zone including basic hydrochemistry and sediment description;
- phytobenthos samples are collected annually once a year (August) from Baltic Proper (one area from coastal zone and one area from open sea which is further divided into 4 subareas). Totally 26 stations are visited. Variables to be measured are abiotic, plants and animals.

C.4. DIRECTIVES FOR SAMPLING AND ANALYSIS OF HYDROGRAPHIC, CHEMICAL AND BIOLOGICAL VARIABLES

Directive on the sampling methods and the procedure for analysis of eutrophication variables are given in Annexes C-1 to C-12:

[Annex C-1 Tables listing sample stations](#)

[Annex C-2 Hydrographic and hydrochemical variables](#)

[Annex C-3 Sediment traps](#)

[Annex C-4 Phytoplankton chlorophyll-*a*](#)

[Annex C-5 Phytoplankton primary production](#)

[Annex C-6 Phytoplankton species composition, abundance and biomass](#)

[Annex C-7 Mesozooplankton](#)

[Annex C-8 Soft bottom macrozoobenthos](#)

[Annex C-9 Guidelines for monitoring of phytobenthic plant and animal communities in the Baltic Sea](#)

[Annex C-10 Guidelines for coastal fish monitoring](#)

[Annex C-11 Guidelines concerning bacterioplankton growth determination](#)

[Annex C-12 Guidelines concerning bacterioplankton abundance determination](#)

In order to increase the number of plankton samples from open Baltic Sea areas and to improve the quality of plankton data from such pelagic biological station, the Contracting Parties have split the responsibility for working up plankton samples from different sub-areas among them. Whenever, a Contracting Party passes a pelagic biological station which is intended to be a high-frequent open sea station (see Annex C-1) quantitative phyto- and zooplankton samples should be collected and sent to the Contracting Party responsible for working up plankton samples from the specific sub-area as listed below. Each of the laboratories will have to decide the amount of resources it can allocate to the analysis. The sampling and working up of samples shall strictly follow the present manual concerning plankton samples.

TABLE C.1. Responsibilities of the Contracting Parties for working up plankton samples

Area	BMP area	Responsible Contracting Party	
		Phytoplankton	Zooplankton
Bothnian Bay	A	Sweden 10)	Finland 3)
The Quark	B	Sweden 10)	Finland 3)
Bothnian Sea	C	Sweden 10)	Finland 3)
Åland Sea	D	Finland 3)	Finland 3)
Archipelago Sea			
Gulf of Finland; eastern part	F	Russia 8)	Russia 8)
- central part		Estonia 2)	Estonia 2)
- western part		Finland 3)	Finland 3)
Gulf of Riga	G	Latvia 5)	Latvia 5)
Northern Baltic Proper	H	Sweden 11)	Finland 3)
Western Gotland Basin	I	Sweden 11)	
Eastern Gotland Basin	J	Sweden 11)	Germany 4)
		Lithuania 6)	Lithuania 6)

Area	BMP area	Responsible Contracting Party	
		Phytoplankton	Zooplankton
Southern Baltic Proper	K	Poland 7)	Poland 7)
		Sweden 11)	Germany 4)
Arkona Basin		Germany 4)	Germany 4)
Bay of Mecklenburg	M	Germany 4)	Germany 4)
Kiel Bight	N		
Little and Great Belt	P	Denmark 1)	Denmark 1)
Kattegat East	R	Sweden 9)	Denmark 1)
Kattegat West		Denmark 1)	Denmark 1)

Responsible laboratories:

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