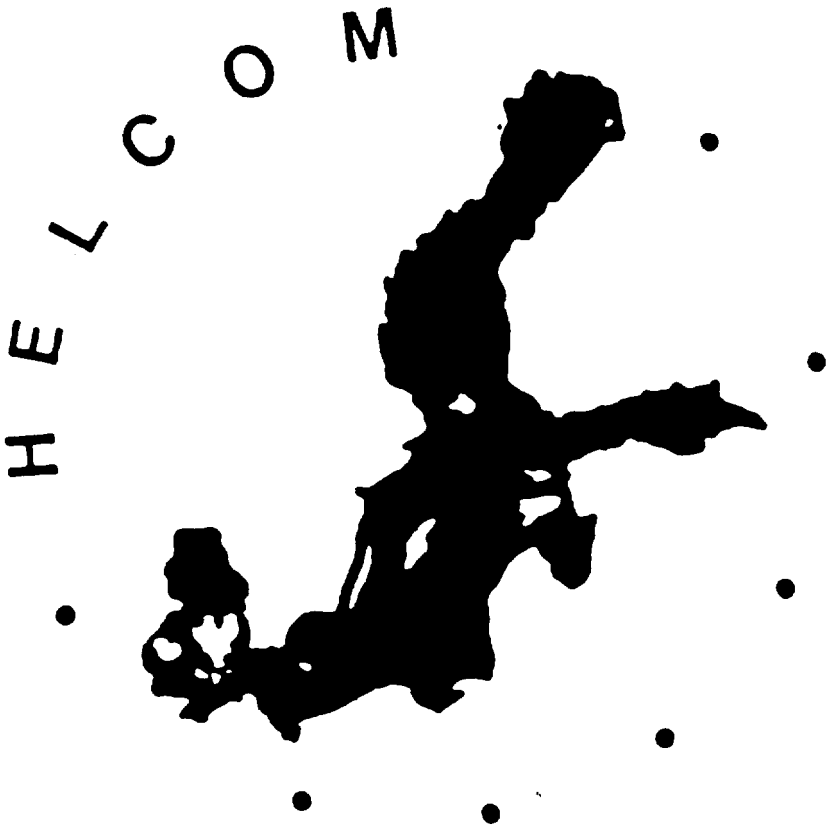
BALTIC SEA ENVIRONMENT PROCEEDINGS

No. 38

THIRD BIOLOGICAL INTERCALIBRATION WORKSHOP

27-3 1 August 1990
Visby, Sweden



BALTIC MARINE ENVIRONMENT PROTECTION COMMISSION
– HELSINKI COMMISSION –

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PREFACE

The Third Biological Intercalibration Workshop in the framework of the Helsinki Commission was held on 27-31 August 1990 in Visby, Sweden. Experts from all other Baltic Sea States except Poland attended the workshop. The International Council for Exploration of the Sea (ICES) coordinated the intercalibration on oxygen and hydrogen sulphide.

This publication contains the results of the intercalibration exercises on phytoplankton, primary production, zooplankton, chlorophyll-a and macrozoobenthos as well as on nutrients, oxygen and hydrogen sulphide.

The conveners of each group of determinands and the editors of the publication Ms. Lena Jacobsson and Mr. Sverker Evans are responsible for the text of this publication.

1. INTRODUCTION

The Swedish Environmental Protection Agency, in cooperation with the Swedish Meteorological and Hydrological Institute, arranged the Third Biological Intercalibration Workshop under the auspices of the Baltic Marine Environment Protection Commission - Helsinki Commission. The Workshop was held in Visby, Sweden, 28-31 August 1990.

The goals for the intercalibration workshop were to assess the degree of comparability of the results obtained by the institutes which produce results for the third five-year period (1989-1993) of the Baltic Monitoring Programme (BMP) of the Helsinki Commission.

The two previous workshops were held at Stralsund, German Democratic Republic, in 1979, and at Rønne, Denmark, in 1982 (HELCOM, 1983). In the 1982 intercalibration, improvements with regard to the comparability of the data were achieved. However, the results also showed a number of problems yet to be solved, and the need for regular intercalibration exercises between the various laboratories was strongly emphasized. The experiences and recommendations of the previous intercalibrations were taken as a starting point for the third intercalibration exercise in Visby. For coordination of the intercalibration, a Steering Group was established, which met in Uppsala 10 January 1990 to discuss the program for the workshop. Altogether six Working Groups were set up, and a convener was nominated for each group.

Steering Group Members

Mr Gunni drtebjerg, Denmark (DK)

Ms Ann-Britt Andersin, Finland (SF)

Mr Sigurd Schulz, German Democratic Republic (GDR)

Mr Stig Carlberg, Sweden (S)

Mr Sverker Evans, Sweden (S)

Ms Elisabet Fogelqvist, Sweden (S)

Mr Torbjörn Willén, Sweden (S)

Conveners

Nutrients, oxygen and hydrogen sulphide:
Mr Stig Carlberg, Ms Lotta Fyrberg, Mr Jorge Valderrama and Mr Bengt Yhlen (S).

Primary production: Mr Sigurd Schulz (GDR).

Chlorophyll: Mr Gunni Ærtebjerg (DX).

Phytoplankton: Ms Kaisa Kononen and Ms Maija Huttunen (SF).

Zooplankton: Ms Gerda Behrends (FRG).

Macrozoobenthos: Mr Hans Cederwall (S).

The Steering Group and the Conveners met at the SMHI Oceanographical Laboratory in Gothenburg 22-23 March 1990 in order to go through the requirements of each Working Group and to solve logistical problems.

Delegations from Denmark, Finland, German Democratic Republic, Federal Republic of Germany, Sweden, and the Union of Soviet Socialist Republics attended the workshop, as well as scientists from the Stockholm and Umeå Marine Sciences Centres. Observers from the Helsinki Commission, the Baltic Marine Biologists (BMB) and ICES were also present.

The following laboratories and research vessels participated in the intercalibration:

DK National Environmental Research Institute.
R/V GUNNAR THORSON

SF Finnish Institute of Marine Research.
R/V ARANDA

GDR Institute of Marine Research, Rostock/Warnemünde.
R/V PROFESSOR ALBRECHT PENCK

FRG Institute of Marine Research, Kiel.
R/V ALKOR

S Swedish Meteorological and Hydrological Institute.
Swedish Environmental Protection Agency.
R/V ARGOS

S	**Stockholm Marine Sciences Center.** Umeå Marine Sciences Center. KBV04 (Swedish Coast Guard)
USSR	**State Oceanographic Institute, Moscow.** **Hydrometeorological Observatory of Klaipeda, Lithuania.** **R/V LEV TITOV**

The timetable for the Third Biological Intercalibration Workshop is presented below in Section 1.1. The sampling stations are presented in Figure 1 (p. 149).

The Working Group reports have been drafted by the convener of each working group and the publication has been edited by Ms Lena Jacobsson and Mr Sverker Evans.

Acknowledgements

The generous and kind support by the County Council of Gotland, the Harbour Authority and the School Office of Visby greatly contributed to the success of the Workshop, and is gratefully acknowledged.

1.1 Time-table for the Third Biological Intercalibration Workshop

January 10	Meeting Steering Group in Uppsala.
March 22-23	Meeting Steering Group and Conveners in Gothenburg.
Monday, August 27	Research vessels meet in Visby. Meeting Steering Group and Conveners on board R/S ARGOS.
Tuesday, August 28	Opening of the Workshop. Working Group Meetings. Meetings Captains, Steering Group and Conveners.

Wednesday, August 29	Field sampling.
07.00	All ships leave Visby harbour. On the way to the sampling station, every ship determines with navigational aids the correct position of the buoy situated roughly at N 573777, E 1816.
08.15	At the sampling station (N 5740, E 1748) zooplankton sampling and secchi disc readings are performed from all ships. The ships then perform different tasks: nutrients by ARGOS; chlorophyll, primary production and secchi disc readings by GUNNAR THORSON. Subsequently, the distribution of samples starts at sea by means of rubber boats.
10.00	The ships split up. ARANDA and ARGOS go to the zoobenthos stations. GUNNAR THORSON collects oxygen samples. The remaining ships return to port.
19.00	ARANDA and ARGOS back in port. Oxygen and zoobenthos samples are distributed.
Thursday, August 30	Analyses on board in Visby.
Friday, August 31	Plenary Meeting. Closing of the Workshop.

2. REPORT OF THE WORKING GROUP ON PHYTOPLANKTON

2.1 Introduction

According to the conclusions of three phytoplankton inter-calibrations carried out within the Baltic Sea countries (Anon. 1979, HELCOM 1983, Leppänen et al. 1991) the critical point in the phytoplankton analysis is the species identification. The confidence limits of the actual counting procedure, as determined by counting culture samples or easily detectable and identifiable species, have generally been near the theoretical values of Lund et al. (1958).

The group therefore decided that the main effort during this intercalibration workshop will be laid on species identification problems.

2.2 Material and methods

2.2.1 Species identification intercalibration

During spring and summer 1990 altogether four water samples, preserved with Lugol+Aa solution were sent to the members of the phytoplankton group. Additionally the sample taken for the counting intercalibration was analyzed for species composition. The samples were as follows:

date	station name	depth	lat.	long.
19.4.1990	Stevens Klint (BMP K6)	O-10m	N55°16.3	E12°34.5
20.4.1990	LL19	5 m	N58°53.O	E20°18.8
04.7.1990	Kiel Lighthouse	5 m	N55°30.O	E10°10.O
31.7.1990	BMPJ1	l m	N57°19.2	E20°03.O
29.8.1990	Visby			

50 ml subsample was sedimented on a counting chamber. The species of the samples were identified and a rough estimation of their

abundance (1 = dominant....5 = scarce) was indicated.

In Visby the group mostly concentrated on microscopical work. The species composition of the samples was discussed during the workshop. Two high level taxonomy teachers - Dr. Bo Sundström and Dr. Karl Tangen gave lectures about the taxonomy of the diatoms, flagellates (Sundström) and dinoflagellates (Tangen) as well as examined samples together with the participants. Also other samples including living samples taken during the workshop and preserved samples from different sea areas were analyzed. Some dinoflagellate species were studied from living and preserved culture samples. A microscope equipped with video-recorder was found to be an excellent tool for all discussions.

2.2.2 Counting intercalibration

During the intercalibration workshop in Visby a water sample of 500 ml and a net sample, both preserved with Lugol+Aa solution were distributed to the group members for the counting intercalibration. For one laboratory, however, a sample of 15 1 was given because the method used by that laboratory requires larger sample volume. The samples were taken from the same water as samples for the primary production and chlorophyll-a measurements.

The species composition of the counting intercalibration sample was analyzed during the workshop.

Five most abundant taxa were counted in order to find out the variability in counting. The minimum number of units to be counted was 50 for all species. The species were selected so that they represented all size classes. The species counted were:

Taxon	Magnif.- of obj.	Counting unit
Aphanizomenon flos-aquae	***10x***	***loo un*** filament
Aphanizomenon flos-aquae filament	10x	whole filament

Chaetoceros danicus solitary cells	10x	cell
Chaetoceros danicus cells in chains	10x	cell
Pyramimonas sp.	40x	***cell***
Cryptomonadales sp.B	40x	cell
Katodinium rotundaturn	40x	***cell***

Five 50 ml subsamples were sedimented and counted by the participants in their own laboratories for checking out the variability between labs. For checking out the variability within laboratories one of the subsamples was counted five times. The countings were done according to the Guidelines for the Baltic Monitoring Programme for the Third Stage (HELCOM, 1988). However, one laboratory, number 9, used the reversed filtration technique (Sournia 1978) to concentrate the subsamples and then counted in a $0.02cm^3$ slide with standard (not inverted) light microscope.

2.3 Results

Results were received from following laboratories:

Denmark	Denmark Marine Identification Agency, Copenhagen. (Bo Sundström)
Finland	Finnish Institute of Marine Research, Helsinki (Maija Huttunen)
	National Board of Waters and Environment, Helsinki (Liisa Lepistö, Pirkko Kokkonen, Maija Niemelä)
FRG	Institut für Meereskunde, Kiel (Regina Hansen, Jaenette Göbel)
	Institut fiir Meereskunde, Warnemünde (Kate Kunert, Günter Breuel)
	University of Rostock (Eugen Kühner)
Sverige	Swedish Environmental Protection Agency, Uppsala (Susanna Hajdu)
USSR	Hydrometeorological Observatory of Klaipeda Klaipeda (Irina Olenina)

2.3.1 Species identification

Results were reported by 8 laboratories representing 12 phyto-plankton specialists. From one laboratory two separate lists were reported. Three participants reported species lists from all five samples, the rest only part of them (Table 1).

Tab.1 Number of species identification intercalibration lists received from the participants

Sample	Number of lists
Stevens Klint	a
LL-9	a
Kiel lighthouse	6
BMPJl	4
Visby	7

Taxa or groups (TU:s) in the original lists.
The number of taxa or groups of phytoplankton reported by the participants from a single sample varied by a factor of two to over a factor of three (Table 2.A).

Tab.2a Number of taxons or groups (TU:s) reported by the participants. The original lists. Total = total number of different TU:s reported by the participants.

Sample	Participant no									
	1	2	3	4	5	6	7	8	9	Total
Stevens Klint	42	33	12	17	30	30	43	22		146
LL-9	23	25	18	15	34	24	36	27		114
Kiel Lighthouse	27	17	20		31	25		33		95
BMPJ1		15			49	30	30			81
Visby		28		11	39	23	36	30	39	97

Tab.2b Number of taxons or groups (TU:s) registered in the revised lists. Total = total number of TU:s after revision

Sample	Participant no									
	1	2	3	4	5	6	7	8	9	Total
Stevens Klint	31	25	12	17	36	32	40	26		90
LL19	18	21	16	12	26	24	37	27		50
Kiel Lighthouse	27	17	20		31		25	33		54
BMPJl	15				49	30	29			57
Visby		24		12	35	28	34	36	39	71

In the sample BMPJl only 6% of all 81 reported units were reported by all participants. In the Stevens Klint sample not any of the 146 different taxa or groups was registered with the same name by all participants (Table 3).

Tab.3 Number of taxons or groups registered in common by (A) all participants and (B) half or more of the participants in original and revised lists.

Sample	A. Number of TU:s registered by all participants		B. Number of TU:s registered by 50% or more of the participants	
	Original	Revised	Original	Revised
Stevens Klint	0	2	a	22
LL19	3	5	311	18
Kiel Lighthouse	3	5	10	20
BMPJl	5	a	23	29
Visby	4	a	24	24

Taxa or groups (TU:s) in the revised lists.

During the Visby meeting it became evident that several of the TU:s reported could be grouped under common labels. Thus, in the one sample discussed by the participants during the meeting - Stevens Klint, 19.4, 1990 - the number of TU:s could be reduced

from 146 to 90 through the agreement that several of the original TU:s, in fact, denoted the same taxon or limited group of organisms (Table 2B. as compared to 2A.) Subsequent compilations made by Bo Sundström of the lists of the four other intercalibration samples also resulted in significant reductions of the original numbers of TU:s. The revised lists are presented in annex 1-5.

2.3.2 Counting

Counting intercalibration was participated by 7 laboratories.

Counting of 5 parallel samples

The cell number results obtained from the countings of 5 parallel chambers are represented in table 4. and Fig. 1. The highest total variability was found for the small flagellates, Pyramimomas spp. (cv% 90) and Cryptomonadales sp. B (cv% 82). This was mainly caused by exceptionally high cell numbers reported by one laboratory (lab. 4). If these values were omitted, the corresponding cv%:s would be 39% and 38% respectively.

Intermediate total variability values were resulted for C. danicus (cv% 56 and 61) and K.rotundatum (cv% 60). The high cv% of K.rotundatum was a result of the fact that generally less than the suggested 50 cells were found in the samples. For C. danicus results were reported only by 5 labs. The participants commented that there were a lot of empty cells even more than 50%, in the sample. One lab. counted empty and full cells separately, two only the cells with contents, one lab both together and one did not give information which kind of cells were counted. Thus the high variability was caused probably by the confusion whether only cells with cell contents should be counted or should also empty cells be included. Moreover the amount of cells counted was hardly reached the suggested 50.

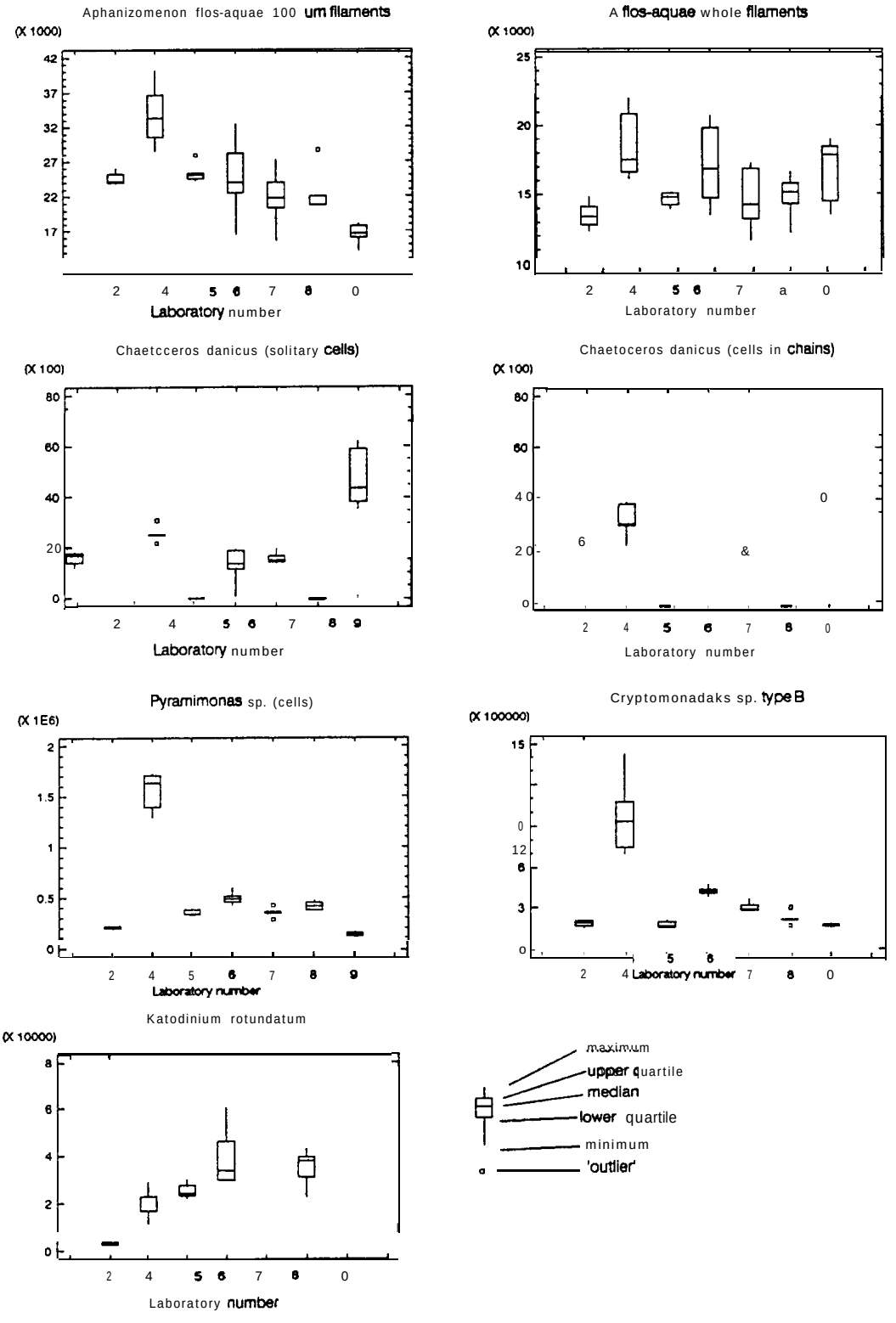
Lowest total variability was found for A. flos-aquae, for which counting of whole filaments resulted somewhat lower cv%:s (16) than counting of 100 um filaments (cv% 25). The reason for low cv% was that generally much more than 50 filaments, sometimes even over 1000 had been counted.

TABLE 4. Summary statistics of the countings of 5 parallel chambers. (cells or filaments x dm^{-3})

	Lab.no	n	min	max	mean	std	cv%
Aphanizomenon flos-aquae (100 um filaments)	2	5	23900	26000	24656	849	3
	4	5	28511	40129	33849	4659	14
	5	5	24320	27840	25440	1405	6
	6	5	16500	32400	24720	6002	24
	7	5	15600	27200	21800	4313	20
	8	5	17822	28516	22226	3912	18
	9	5	14220	18071	16531	1545	9
	total	35	14220	40129	24174	5953	25
Aphanizomenon flos-aquae (whole filaments)	2	5	12320	14840	13452	901	7
	4	5	16077	21980	18591	2656	14
	5	5	13920	17120	15016	1255	8
	6	5	13500	20700	17100	3125	18
	7	5	11600	17200	14600	2383	16
	8	5	12163	16566	14763	1681	11
	9	5	13509	18960	16614	2461	15
	total	35	11600	21980	15734	2594	16
Chaetoceros danicus (solitary cells)	2	5	1240	1920	1644	259	16
	4	5	2198	3077	2562	318	12
	5	-					
	6	5	1160	2100	1696	404	24
	7	5	1400	1960	1600	228	14
	8	-					
	9	5	3591	6237	4801	1210	25
	total	25	1160	6237	2461	1366	56
Chaetoceros danicus (cells in chains)	2	5	2220	2760	2516	193	8
	4	5	2198	3831	3153	673	21
	5	-					
	6	5	940	1120	1004	77	8
	7	5	1800	2000	1880	80	4
	8	-					
	9	5	3894	6715	5585	1431	26
	total	25	940	6715	2827	1712	61
Pyramimonas spp. (cells)	2	5	198531	227943	215034	12832	6
	4	5	1294298	1721542	1550644	194388	13
	5	5	330150	386595	352728	26831	8
	6	5	424000	592000	494400	63850	13
	7	5	281600	420800	353920	49566	14
	8	5	373758	470902	416238	44644	11
	9	5	109697	162641	135142	22432	17
	total	35	153596	1419958	352486	28847	90
Cryptomonas sp. type B (cells)	2	5	153596	200165	178433	18292	10
	4	5	691130	1419958	970095	293141	30
	5	5	177855	224715	196599	21808	11
	6	5	384000	464000	422400	29611	7
	7	5	278400	358400	306560	33055	11
	8	5	167944	302358	221620	49417	22
	9	5	155583	184860	171691	10944	6
	total	35	153596	1419958	352486	288471	82

	Lab.no	n	min	max	mean	sta	cv%
Katodinium rotundatum (cells)	2	5	2304	3968	3123	650	21
	4	5	11811	28900	19601	6561	33
	5	5	22365	29820	25560	3105	12
	6	5	30000	60000	40000	12961	32
	7	5	15680	17600	16512	737	4
	8	5	23051	42809	34906	7845	22
	9	5	13746	14813	14125	420	3
	total	35	2304	60000	21975	13196	60

Fig. 1: Results from the counting of 5 parallel samples



Individual variability (within laboratories) was found to be considerably lower. For all species the variability was below 35%.

Five **repeated countings of one chamber**

Results of cell numbers are presented in table 6. and Fig. 2. For all species the variabilities were within the same range as found for individual countings of 5 parallel samples.

2.3.3 **Biomass values**

The species mean plasma volumes used for biomass calculations by the laboratories are presented in table 6:

Tab.6 Mean plasma volumes of the species used for biomass calculations.

	Participant number						
Species	2	4	5	6	7	8	9
A. flos-aquae	1964	1818	1600	2400	1500	1318	1273
C.danicus	***1767***	1900		1000	1000	**-**	***4544***
Pyramimonas sp.	**28/153**	***227***	***24***	***800***	110	***38***	***230***
Cryptomonadales B	78	227	190	100	50	35	57
K. ***rotundaturn***	***42***	***318***	***580***	***600***	110	***231***	***135***

Great differences were found between the values used for biomass calculations especially in the group of small flagellates (Pyramimonas sp. range 24-800 um^3, Cryptomonadales sp. B 35-227 um^3, K. rotundaturn 42-600 um^3). The biomass values based on the average cell number obtained from 5 parallel samples and species mean plasma volumes are presented in Fig. 3. Remarkable differences between biomass values were resulted as a cumulative effect of differences in cell numbers and species volumes.

2.3.4 comparison of the importance of different variation sources

The sampling design of the experiment resulted information about three different hierarchical levels of variation sources:

Level 1 variability reflects differences caused by (1) subsampling the 500 ml aliquots which were delivered to the participants in Visby and (2) variation caused by differences in sample treatment (like storing, microscope equipment, knowledge of taxonomy etc.) between laboratories. The design used in this experiment did not allow to separate between these two variation sources.

Level 2 variability was caused by subsampling for sedimentation chamber.

Level 3 variability was resulted from the counting procedure. Based on the assumption of homogenous distribution of cells on the chamber it should follow the predictability according the Poisson-distribution.

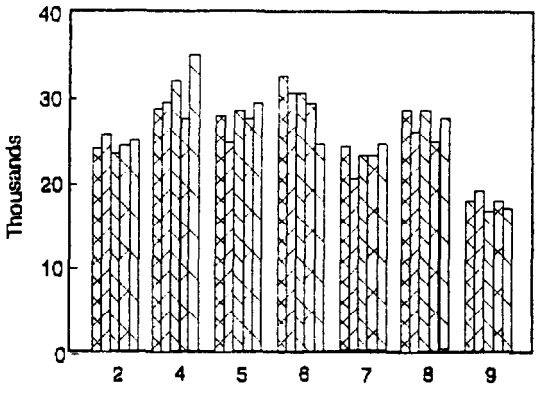
The importance of the three variability levels was tested by the nested variance analysis (SAS software, proc. NESTED). The results are presented in Table 7.

TABLE 5. Summary statistics of the 5 repeated countings of the same chamber (cells or filaments x dm^{-3})

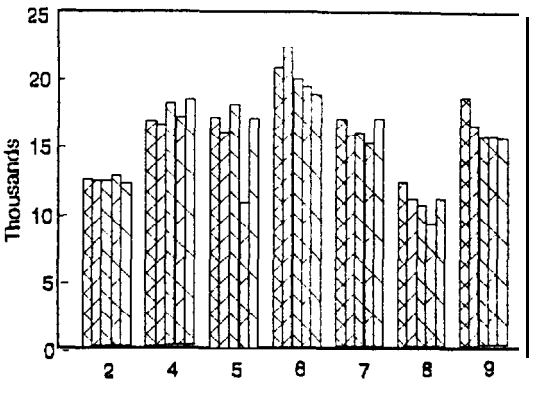
	Lab.no	n	min	max	mean	std	cv%
Aphanizcmenon flos-aquae (100 um filaments)	2	5	23180	25500	24280	880	4
	4	5	27417	34776	30355	2960	10
	5	5	24640	29200	27472	1723	6
	6	5	24300	32400	29280	3027	10
	7	5	20200	24200	22800	1594	7
	a	5	24742	28516	27006	1640	6
	9	5	16590	18960	17579	946	5
Aphanizcmenon flos-aquae (whole filaments)	2	5	12000	12580	12272	209	2
	4	5	16328	18086	17132	782	5
	5	5	10800	18000	15760	2869	la
	6	5	la600	22200	20100	1407	7
	7	5	15000	16800	16000	787	5
	a	5	9017	12163	10695	1129	11
	9	5	15326	18368	16168	1291	8
Chaetoceros danicus (solitary cells)	2	5	920	1240	1048	125	12
	4	5	2512	3769	3190	573	18
	5						
	6		1940	1940	1940	0	0
	7	5	1400	1520	1440	49	3
	a						
	9	5	4480	6237	5205	675	13
Chaetoceros danicus (cells in chains)	2	5	1880	2520	2152	286	13
	4	5	2135	4773	3228	1219	38
	5						
	6	5	1120	1120	1120	0	0
	7	5	1800	1960	1864	88	5
	a						
	9	5	4740	6715	5517	725	13
Pyramimcnas sp. (cells)	2	5	175655	222224	193139	17347	9
	4	5	1633580	1784372	1708976	61561	4
	5	5	309915	362100	338031	20604	6
	6	5	304000	544000	436800	90439	21
	7	5	358400	420800	399360	25913	6
	a	5	434679	500539	478805	27215	6
	9	5	94208	143977	116218	18294	16
Cryptomonades sp. type B (cells)	2	5	130720	153596	140034	a942	6
	4	5	741394	1068110	864541	137768	16
	5	5	208740	227910	221520	8146	4
	6	5	408000	584000	494400	72752	15
	7	5	312000	331200	320320	7552	2
	a	5	240977	329302	297806	33381	11
	9	5	154642	184860	172070	13340	a
Katodinium rotundatum (cells)	2	5	3072	3968	3558	367	10
	4	5	18345	23622	22165	2157	10
	5	5	21300	29820	25347	3401	13
	6	5	16000	34000	24400	6387	26
	7	5	15360	18240	16768	1167	7
	a	5	29637	39516	34906	4994	14
	9	5	13746	16590	14562	1156	a

Fig. 2: Results from 5 repeated countings of same chamber (cells or filaments * dm^3)

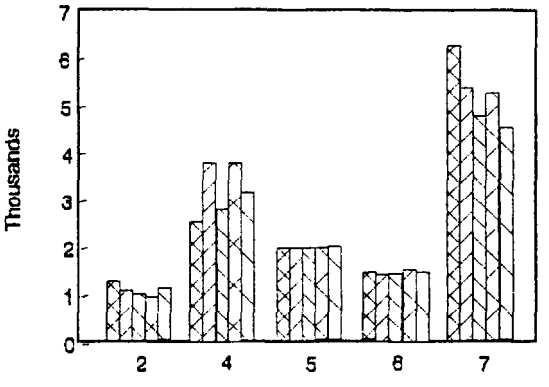
Aphanizomenon flos-aquae (100 um filaments)



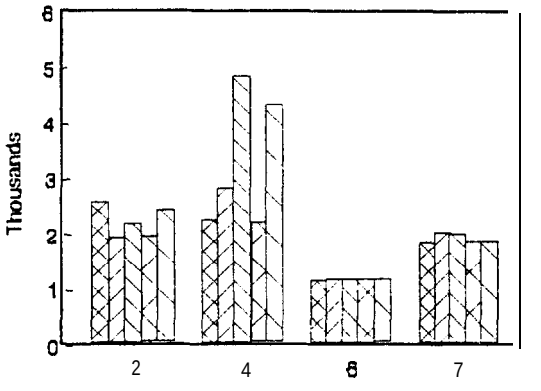
Aphanizomenon flos-aquae (whole fil)



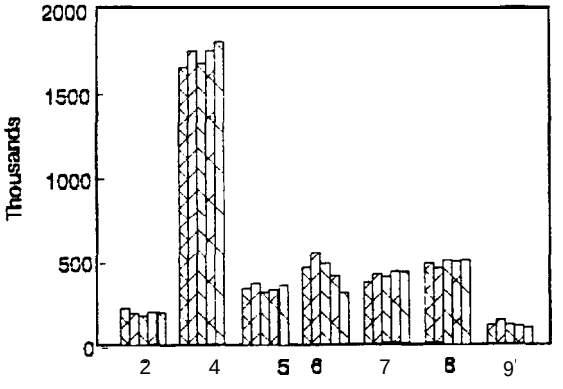
Chaetoceros danicus (solitary cells)



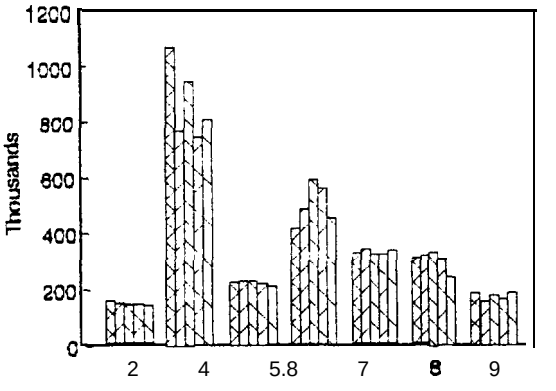
Chaetoceros danicus (cells in chains)



Pyramimonas sp.



Cryptomonadales sp. type B



Katodinium rotundatum

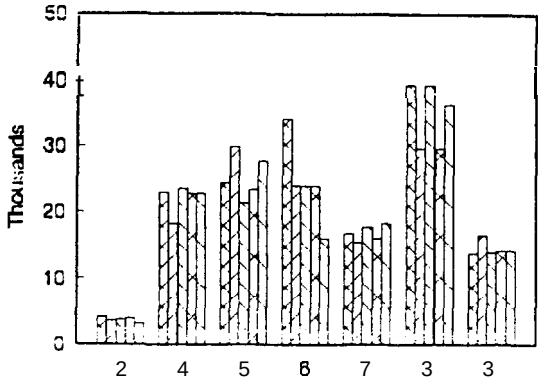
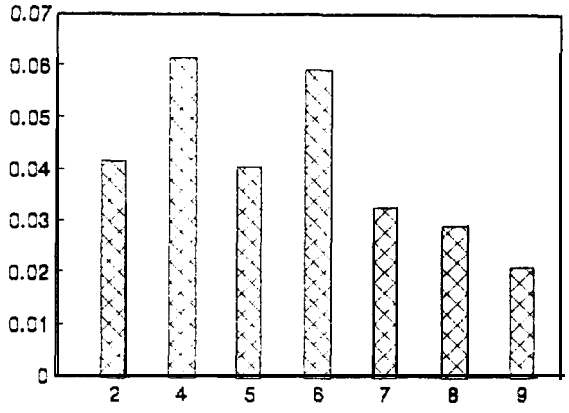
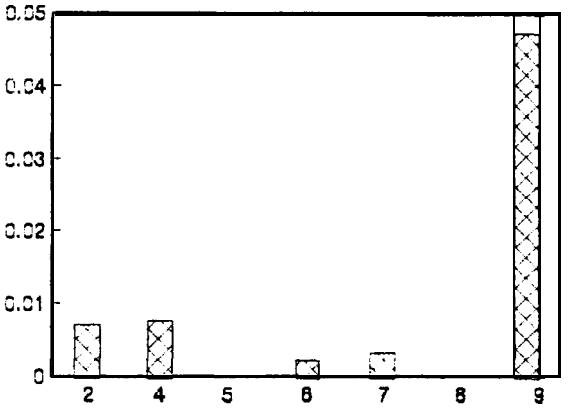


Fig. 3: Biomass values (mg dm^{-3}) of the species based on the average cell number of 5 parallel samples and species mean volumes used by the laboratories.

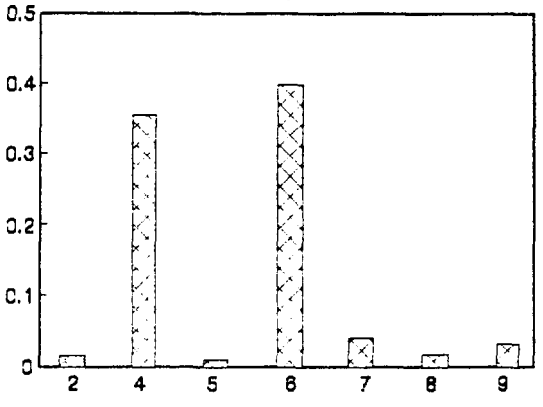
Aphanizomenon flos-aquae



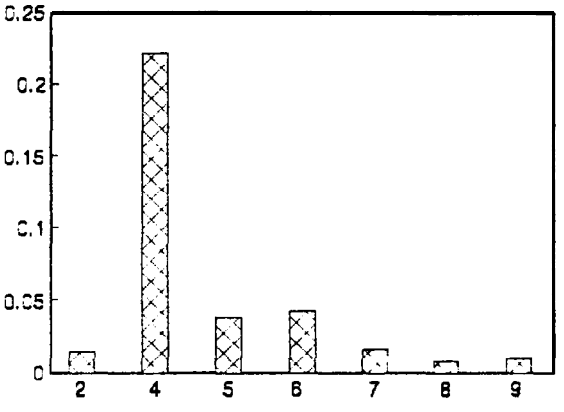
Chaetoceros danicus



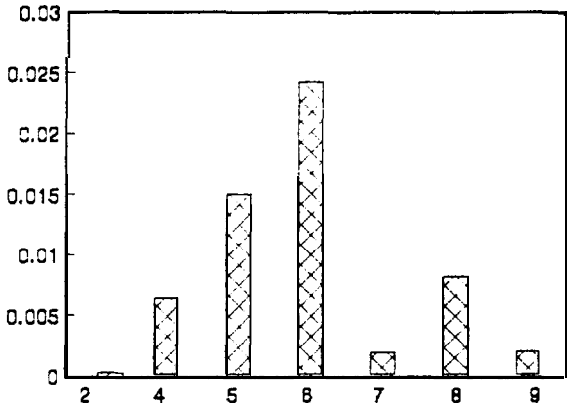
Pyramimonas sp.



Cryptomonas sp. B



Katodinium rotundaturn



Tab.7 Percentage of variance accounted for the three variance sources: laboratory (level 1), parallel chamber (level 2), repeated counting (level 3):

Species	Percentage of total variance accounted for Level 1	Level 2	Level 3
A. flos-aquae 100 um fil.	59	28	13
A. flos-aquae whole fil.	38	39	23
C.danicus, solitary cells	86	7	7
C. danicus	87	0	13
Pyramimonas sp.	97	2	1
Cryptomonadales	86	9	5
K. rotundaturn	68	23	8

It was found that for all species except A.flos-aquae (whole filaments) the most important variation source was that between laboratories and the least important was the counting procedure. For filaments of A. flos-aquae the variation caused by subsampling to chambers was equal to differences between laboratories. This is certainly due to the stick-like clump-formation of this species. As could be expected also the variability found in counting was highest for species occurring in chains or clumps (C. danicus, cells in chains, A. flosaquae, whole filaments).

2.4 Discussion

Phytoplankton as primary producers in the marine environment is with good reasons a fundamental object for monitoring programmes in the seas. Recent evidence and development in eutrofying marine environments points out that not only the amount of primary production, but changes in the qualitative composition of phytoplankton are of great importance in the eutrophication process. Blooms of harmful algal species have become a frequent phenomenon all over the world seas, and also the Baltic Sea.

The results of this intercalibration show that within the Baltic Monitoring Programme the identification and naming of taxa are the critical points in phytoplankton analysis and that, at the present level of coordination, it is difficult to compare phytoplankton data from laboratories on a scientifically meaningful level.

Although it can be assumed that all participants in the intercalibration experiment are well experienced in the identification of phytoplankton, it is obvious that the "schooling" varies. Thus, the cause of the "identification problem" is hardly to be found in varying abilities to recognize morphological particularities in phytoplankton, but rather in the fact that individual phytoplankton analysts have different "taxonomical" backgrounds. This is also reflected in the results of the counting experiment, in which the variation observed was in accordance with the methodological noise expected from values in Lund et al. (1958).

The Baltic monitoring samples are in most cases analyzed as preserved. During the Visby meeting according to the comparisons made between living and preserved samples it became clear that, especially concerning flagellates and dinophyceans, at the present stage it is often impossible to make identifications on the species level.

The counting itself was proved to be acceptable reflecting the variation caused by methodological noise according to estimations presented by Lund et al. (1958). Counting of 50 cells, as was decided to be done, should result maximum error of ± 50%, whereas the cv%:s obtained from 5 repeated countings of same chamber were in all cases except one below 30%. The main source of total variation was the differences between laboratories. For several species this could be accounted for confusions in identification or whether empty diatom frustules should be counted or not. At present all labs participating the BMP still do not use inverted microscope. The species mean volumes used for biomass calculation also differed remarkably. A good volume calculation is really necessary to obtain a reliable total volume biomass result.

The group also discussed the HELCOM data reporting formats for phytoplankton and it was realized that the present format does not allow additional information about phytoplankton taxa, e.g. cell dimensions, developmental stages, trophy etc. In order to facilitate the presentation and treatment of phytoplankton data it is, therefore, necessary that the data reporting format will be revised so that these types of information can be reported.

In summary this intercalibration experiment showed that there is a serious lack of coordination between laboratories especially concerning the naming of phytoplankton. At the present level of coordination, quantitative and qualitative phytoplankton data are, at the most, scientifically comparable only at the local level and, probably only when data originating from one analyst are compared. Species identification could be easily unified through the discussion of results between the participants and through the discussion of results between the participants and with the guidance of taxonomists.

2.5 Recommendations

To assure a scientifically acceptable level of phytoplankton data presentation it is necessary to:

1. Coordinate the analyzing procedures of all laboratories involved in the Baltic Sea Monitoring Programme. This should include
- an appointment of a phytoplankton coordinator – preferably a taxonomist for the Baltic Sea area
- regular meetings of the phytoplankton analysts on yearly basis
- agreement upon common nomenclature for certain groups of phytoplankton
2. Accelerate the publishing of the phytoplankton identification sheets.
3. Arrange courses on phytoplankton taxonomy and floristics for monitoring personnel.
4. Ensure that all phytoplankton analysts have facilities, including inverted microscopes, to analyze samples according to the Guidelines. Moreover, an opportunity to analyze living samples should be arranged, when possible.
5. Revise the HELCOM data reporting formats for phytoplankton.

Since some costs will be involved in the realization of the recommendations it is necessary that the countries involved in the Baltic Monitoring Programme agree to set new funds for these purposes.

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Sample: Stevens Klint (BMP K 6) 19.4.1990

Species	Laboratory number							
	1	2	3	4	5	6	7	8
CYANOPHYCEAE								
Aphanethece sp.			-	5	-	-	-	-
Gomphosphaeria sp.		+	4	3	+	+	+	5
M. reinboldii		+	-	+		+	+	2
Rhabdogloea clathrata			-	-	-	-	+	-
Aphanizomenon flos-aquae			+	3		+	+	-
CRYPTOPHYCEAE								
Cryptophyceae sp. type C			-		-	2	+	-
Cryptophyceae sp. type B	1	1	3		+	5	4	10
Cryptophyceae sp. type A	+	1			1	-	5	-
Cryptaulax sp.						+-		-
Katablepharis sp.						+-		-
K. ovalis						+-		-
DINOPHYCEAE								
Prorocentrum minimum		5			-	-	-	-
Dinophysis sp.	-	-		+	-	-	-	-
Dinophysis acuminata	5	5	+	5	+	+	+	+
Gymnodiniales sp. type A	4	4	+			+	+	-
A. crassum					+	-	+	-
Oxytoxum sp.					-	-	-	+
Gymnodiniales sp.	+	+	-		+	+	+	-
Gymnodinium sp. type V		4	5		-	3	+	+
G. cf. simplex		2			2	-	+	9
G. semidivisum					+_		-	-
G. cf. lohmannii					-	-	+	-
Gyrodinium sp.					-	-	-	6
G. spirale	-				+_		-	-
K. rotundatum	5				4	-	+	+
Peridiniales sp.	-		1		-	-	-	+
Peridiniales sp.	5	-			-	-	+	+
Peridinella catenata	-	5		4		+	+	-
Gonyaulax triacantha	5	5	+	3	+	+	+	+
He terocapsa triquetra	5			-		+	+	+
Oblea rotunda	-			-		+-		+
Protoperidinium brevipes	5			-	+	+	-	-
P. granii				-	+	-	-	-
P. pellucidum	-			-	-	-	+	-
Protoperidinium sp. 45 um	5			-	-	-	-	+
70 urn	5	-		-	-	-	-	-
Ebria tripartita	4	5	+	-	+	+	+	+
Scrippsiella cf. trochoidea	5			-	-	-	-	-
Dinophyceae (unident.)	+	+		1	-	-	-	-
PRYMNESIOPHYCEAE								
Chrysochromulina sp.	5	1			+	+	+	+
Prymnesiophyceae sp.		+			-	-	-	-
CRASPEDOPHYCEAE								
Monosiga sp.					-	-	+	-

PRASINOPHYCEAE								
Pyramimonas cf.virginica	5	1	-	-		-	-	-
Pyramimonas sp.	4	-	-	-		-	-	-
Tetraselmis sp. 6 um	4	1	-	-		-	-	-
CHRYSOPHYCEAE								
D. balticum	3	4	-	-	3	+	2	8
Pseudopedinella tricostata			-	-		+-		-
EUSTIGMATOPHYCEAE								
Nannochloris sp.			-	-		1	-	-
BACILLARIOPHYCEAE								
Eupodiscales sp.			-	-		+-		-
Actinocyclus octonarius			-	-	+	-	-	-
Chaetoceros sp.	-		-	-			+	-
Chaetoceros sp. 5x2.4 um	5		-	-		-	-	-
C. danicus	1		-	-	+	-	-	-
C. gracilis	-			5	+	-	-	-
C. ceratosporus	-		-	-		+-		-
C. subtilis	-		-	-		+	+	+
C. similis	-		-	-		-	-	+
2 x 9.6 um	5		-	-		-	-	-
Rhizosolenia fragilissima	-			5		-	-	-
R. minima	-		-	-	+	-	-	-
Thalassiosira sp.	-		-	-			+	-
T. angulata	-		-	-		-	-	+
T. baltica	-		+	4	+	-	-	-
T. punctigera	5		-	-		-	-	-
T. levanderi		-		4		-	-	-
Thalassiosira sp. 45 um		5	-	-		-	-	-
Skeletonema costatum		5		4	+	-	-	-
S. subsalsum		-		5		-	-	-
Gyrosigma sp.		-	-	-	+	-	-	-
EUGLENOPHYCEAE								
Eutreptiella sp.	4	1	-	-	+	+	+	+
Eutreptia Ianowii			+	-		-	-	-
Trachelomonas sp.			-	-	+	-	-	-
PRASINOPHYCEAE								
Pedinomonas sp.			-	-		+	+	-
Scourfieldia sp.			-	-		+	-	-
Micromonas pusilla	-		-	-	+		+	-
Halosphaera colonies 50 um	3		-	-		-	-	-
Mantoniella squamata	-		-	-	+	-	-	-
N. pyriformis	-		-	-	+		+	-
N. minuta	-		-	-	+	-	-	
Pyramimonas sp.	3	-	-	-	5	+	3	10
P. virginica	5	1	-	-	+	-	-	-
Ochromonas sp.	-	-	-	-			+	-
CHLOROPHYCEAE								
Oocystis sp.	5	+	2	5	+		+	+
Nannochloris coccoides			-	-		1	+	-
Planktonema lauterbomii			+	-	+		+	-
Polytoma sp.			-	-		+	-	-

Choanoflagellates spp.	-	-	-	-	-	4	+	4
Flagellates (unident)	+	-	-		-	1	-	-	+	+
Ultrapankton < 2 um	-	-	-	-	-	-	+	-
Mesodinium rubrum	5	+	-	-	+	-		+_

Sample: LL19 20.4 1990

Laboratory number	1	2	3	4	5	6	7	8
CYANOPHYCEAEA								
Ap hanothece sp.		-	-					7
Gomphosphaeria sp.			+	5	+	+	+	10
Microcystis reinboldii		+	-		+	-	+	-
Anabaena subcylindrica		-	-		+	-	-	-
Ap han izomenon flos-aquae	5	+	+	5	+	-	+	+
CRY PTOP HYCEAE								
Cryptomonadales type A		1	-	-	+	+	+	+
Cryptomonadales type B	1	1	+	-	+	3	4	8
Leucocryptos marina		-	-					5
DINOPHYCEAE								
Dinophysis acuminata	5	5	+	5	+	+	+	+
Dinophysis acuta		-	-				+	-
Di nophysis sp.			+	4		-	-	- -
Amphidinium sp.			+	-	+	+	+	-
Gonyaulax catenata	2	5	4	2	+	5	2	+
Gonyaulax triacantha		-	-	-	+	-		
Oblea rotundata		-	-			+	-	-
Naked dinos 6- 10 µm	3		2		-	-	- -	- -
Naked dinos 10- 15 µm		2		-	-	+	- -	- -
Naked dinos 15-20 µm		2		-	-	-	- 1	-
Gymnodinium sp.V		-	-	-	+	-	+	+
Naked dinos 20-40 µm	5		2	-	-	+	- -	- -
Gyrodinium spp.	4		5	-	-	+	- +	-
Katodinium rotundatum		-	-		+	-	+	-
Glenodinium sp.	5		-	2	-	-	- -	- -
Protoperidinium bipes	5	5	+	5	+	+	+	+
Protoperidinium spp.	5	5	-	5	+	-	+	6
Ebria tripartita	5	5	+		-	+ +	+	+
Dinophyceae sp. 14 µm	4		-	-	-	-	- -	- -
Dinophyceae sp. 25-35 µm	3		-	-	-	-	- -	- -
Unidentified dinoflag.		-	-	1	-	-	-	-
CHRYSOPHYCEAE								
Ochromonas sp.		-	-		+	-	+	
Pseudopedinella tricostata		-	-		+	+	+	-
EUSTIGMATOPHYCEAE								
Nannochloropsis sp.		-	-			1	-	-
BACILLARIOPHYCEAE								

Eupodiscales sp.		-	-			+	-	-
Chaetoceros wighamii		5	-	-	-	-	+	+
Chaetoceros ceratosporus		-	-	-	+	+	+	+
Chaetoceros subtilis		-	-			+	+	+
Skeletonema costatum	5	5	+	4	-	4	+	4
Thalassiosira cf. levanderii	5	5	5	5	+	+	3	3
Thalassiosira bal tica		5	+	4	+	+	+	+
Achnan tes taen iata			+	-	-	-		
Ni tzschia longissima		5	-	-	-	-	-	+
EUGLENOPHYCEAE								
Eutrep tiel la sp.	2	1	3	-	+	+	5	9
PRASI NOPHYCEAE								
Pyrami monas sp.		1	-	-	+	2	+	-
CHLOROPHYCEAE								
Monoraphidium contortum			+	-	+	+	+	+
Oocystis sp.		-	-			+		+
MISCELLANEOUS								
Choanoflagellates		-	-			+	+	+
Mesodinium rubrum	5	-	-	-	+	-	+	-
Miscellaneous 3-6 µm		-	-	-			+	1
Miscellaneous 6-10 µm	3	-	-	-	-	-	+	2
Unidentified flagellates		-	-	2	-	-	+	-

ANNEX 3.

Sample: Kiel-lighthouse 4.7 1990

Laboratory number	1	2	3	4	5	6	7	8
CYANOPHYCEAEA								
Gomphosphaeria sp.		-	+		+	+		+
Merismopedia punctata		+	+			+		+
Microcystis reinboldii		-			+	-		
Achroonema cf. len turn		-				-		+
Anabaena cf. inaequalis	1	+	+		+	4		4
Aphanizomenon flos-aquae	-							+
Nodularia spumigena	5		+					+
cf. Synechococcus	-							1
Chroococcaceae sp.	-							2
CRYPTOPHYCEAE								
Cryptomonadales type A	3				+	+		+
Cryptomonadales type B					+	+		8
Leucocryptos marina								5
cf. Rhodomonas minuta			1			5		
DINOPHYCEAE								
Prorocentrum micans	5	+	+		+	+		+
Dinophysis norvegica	5	+	+		+			+
Am phidinium sp.	-	-			+	+		
Gonyaulax spinifera	5	-						
Ceratium tripos	4	+	+		+	+		+
Cerati um fusus	4	+	+		+			+
Heterocapsa triquetra	4	+	+		+			+
Armoured dinos spp	4	-				-		+
Naked dinos spp.	5	+	2		+	+		+
Katodinium rotundatum	-	-			+	+		
Ebria tripartita	-	-	+					+
CHRYSOP HYCEAE								
Dinobryon petiolatum	-	-						9
Dinobryon balticum	-	-			+	+		+
Calycomonas spp.	5	-			+	+		+
Distephanus speculum		+	+					+
Monochrysis sp.						+		
Ochromonas sp.					+	+		
EUSTIGMATOPHYCEAE								
Nannochloropsis sp.						1		
BACILLARIOPHYCEAE								
Cerataulina pelagica			+					+
Chaetoceros danicus			+		+			+

Chaetoceros socialis	5	-	-			+
Coscinodiscus radiat us	-	+	-	-		-
Ditylum brightwellii	5	-	-			
Proboscia alata		-	-	+	-	+
Rhizosolenia fragi lissima	4	+	5	+	+	7
Rhizosolenia minima	-	-	-			+
Skeletonema costatum		-	-	+	-	+
Thalassiosira angulata			+			+
Thalassiosira baltica	-	-	-	+		
Thalassiosira leptopus			+			-
Nitzschia closterium	-	-	-			+
Nitzschia sp.	-	-	-	+	+	+
Thalassionema ni tzsch.	5	+	4			10
Eupodiscales sp. 3µm		-	-		+	+
Pennales SD.	-	-	-			5
PRASINOP HY CEAE						
Pyramimonas sp.	5	-	-		+	
CHLOROP HYCEAE						
Monoraphidi um contortum	-	-	-			+
Oocystis sp.	5	+	-	+	+	
MISCELLANEOUS						
Miscellaneous l-3 µm		-	-	-	-	3
Miscellaneous 6- 10 µm	4	-	-	-	-	-
Unidentified fl. 25*19µm	5	-	-	-	-	-

ANNEX 4.

Sample: BMBJ13 1.7 1990

Laboratory number	1	2	3	4	5	6	7	8
CYANOPHYCEAEA								
Aphanothece sp.						+	+	
Gomphosphaer ia sp.		+			2	5	+	
Microcystis reinboldii		+			+	3	+	
Anabena spp.		+			+	+	-	
Aphan izomenon flos-aquae		+			1	4	3	
Nodularia spumigena		+			3	+	+	
CRY PTOPHYCEAE								
Cryptomonadales type A		+			+	+	+	
Cryptomonadales type B		+			5	+	+	
Cryptomonadales type C		+			+	-	-	
Katablepharis spp.						+	-	
DINOPHYCEAE								
Prorocentrum balticum		-			+	-	+	
Dinophysis norvegica		+			+	-	+	
Dinophysis rotundata		-			+	-		
Amphidinium crassum		-			+	-		
Cerati um furca		-			+	-	-	
Ceratium tr ipos		-			+	-	-	
Gonyaulax catenata		-			+	-		
Gonyaulax grindleyi		-			+	-		
Naked dinos 6-10 µm		-					+	
Naked dinos 10-15 µm		+			+		+	
Naked dinos 15-20µm		-			+			
Naked dinos 20-40µm		-			+	+	+	
Gyrodinium glaucum		-			+			
Gyrodinium spp.		+					+	
Katodinium rotundaturn		-			+	-		
Ebria tripartita		-			+	-	+	
Dinophyceae sp. 8 µm		-				+		
CHRYSOPHYCEAE								
Dinobryon petiolatum		-			+	+	+	
Calycomonas wulffii		-			+	-	+	
Ochromonas sp.		-					+	
Pseudopedinella tricostata		-				+	+	
BACILLARIOPHYCEAE								
Actinocyclus octonarius		-			+	-	-	
Chaetoceros danicus		+			+	+	+	
Chaetoceros subtilis		-					+	
Cyclotella caspia		-					+	

Rhizosolenia minima		+	-	
Thalassiosira cf. pseudonana	+	+	+	
Nitzschia sp.		+	+	+
Tabellaria flocculosa			+	-
PRYMNESIOPHYCEAE				
Chrysochromulina sp. 3 µm	-	4	-	-
Chrysochromulina spp.	-	+	+	+
Prymnesium sp.	-	+	-	-
Pav lova sp.	-	+	-	-
EUGLENOPHYCEAE				
Eutrep tiel la sp.			+	-
PRASI NOPHYCEAE				
Mantoniel la squamata	-	+	-	-
Pyram i monas sp.	-	+	+	+
Pyram imonas vi rgi nica	-	+	-	
CHLOROPHYCEAE				
Chlamydomonas sp.			+	-
Polytoma sp.			+	-
Botryococcus brauni i				+
Dictyosphaerium pulchellum		+	-	-
Monoraphidium contortum		+	+	+
Oocystis borgei		+	+	
Oocystis lacustris			+	+
Planktonema lauterborni	+	+	+	+
MISCELLANEOUS				
Monad 3 µm	-		1	+
Monad spp			2	+

Sample: Visby 29.8 1990

Laboratory number	1	2	3	4	5	6	7	8	9
CYANOPHYCEAEA									
Achroonema sp.		-						+	-
Microcystis reinboldii		-				+	-	-	-
Anabaena baltica									+
Ap han izomenon flos-aquae		+		+	+	4	5	+	+
Nodularia spumigena		+		+	+	-	+	+	+
CRY PTOP HYCEAE									
Cryptomonadales type A		+			+	-	+	+	+
Cryptomonadales type B		+		+	+	+	2	+	+
Cryptomonadales type C					+	+	+	+	+
Chroomonas acuta		-							+
Katablepharis ovalis		-				+	-	-	-
Leucocryptos marina		-			+	-	+	5	-
DINOPHYCEAE									
Prorocentrum micans		+					+	-	+
Prorocentrum minimum		+			+	-	-	+	+
Dinophysis acuminata		+		+	+	+	+	+	-
Dinophysis cf. baltica		-						+	-
Dinophysis norvegica		+			+	-	+	+	+
Dinophysis rotundata		+			+	-	+	+	-
Amphidinium sp.		-			+	-			+
Ceratium lineatum		+			+	-		+	-
Ceratium tripos		+					+		
Gonyaulax spi nifera		-			+	-	-	-	-
Gonyaulax triacantha		-				+	-	-	-
Gonyaulax grindleyi		-					+	+	-
Gonyaulax verior		-						+	-
Oblea rotundata		-			+	-			
Naked dinos 10-15 µm		+		+	+	+	+	+	+
Naked dinos 15-20 µm		+		+	+	+	+	+	+
Naked dinos 20-40 µm		+			+	+	-		
Gymnodinium sp.V		-			+	-	+	+	+
Gymnodinium simplex		+			+	-	+	-	+
Gym nodinium cf. splendens		-				+	+	+	-
Katodinium rotundaturn		+		+	+	+	4	+	+
Katodinium glaucum		-			+	-	-	-	-
Diplopsalis group		-			+	-	-	-	-
Protoperidinium granii		-			+	-			
Protoperidinium ovatum		-			+	-	-	-	-
Protoperidinium pellucidum					+				
Scripsiel la trochoidea					+	-		-	
Peridiniales spp.				+	-	-			+

Helgolandinium subglobosum							+
Ebria tripar tita	+		+	+	+	+	+
CHRYSOPHYCEAE							
Ochromonas sp.	+			+	+	-	-
Pseudopedinella tricostata			+	+	+	+	-
Pseudopedinella pyriforme			+	+	+	+	-
Apedinella spinifera					+	-	-
Distephanus speculum	+						
PRYMNESIOPHYCEAE							
Chrysochromulina sp.	-		+	+	+	+	+
cf. Prymnesium parvum	-						+
BACILLARIOPHYCEAE							
Actinocyclus octonarius	-		+	+	+	+	+
Chaetoceros danicus	+	+	+	5	+	+	+
Chaetoceros cf. tenuissimus	-		+	-		+	
Melosira sp.	+			-			
Rhizosolenia fragilissima	-			-			+
Skeletonema costatum	-		-	-	-		+
Coscinodiscus radiatus	+		-	-	-		-
cf. Cyclotella sp.	-		-	-	-	+	+
Achnantes taeniata	-	-	-	-		-	+
Diatoma elongatum	-	-	-	+		-	-
Ni tzschia longissima	-	-	-	-	+	-	+
EUGLENOP HYCEAE							
Eutreptiel la sp.	-	+	-	+	+	+	+
Euglena sp.	+						+
PRASI NOPHYCEAE							
Pyramimonas cf. virginica				3	3	+	+
Pyrami monas sp.	+	+	-	1	1	+	+
Nephroselmis sp.					+		
CHLOROP HYCEAE							
Oocyst is borgei				+			+
Oocystis sp.	+	+	+	+	+	+	+
Monoraphidium contortum							+
Scenedesmus intermedius							+
MISCELLANEOUS							
Coenochloris cf. planoconv.					+	-	-
Mesodinium rubrum	+				+	+	-
Unidentified small flagellates			+	-	+	-	

3. REPORT OF THE WORKING GROUP ON PHYTOPLANTKON PRIMARY PRODUCTION

3.1 Participating countries/institutes/persons

Sweden	SMHI Göteborg	E.-G. Thelen A. Taglind J. Szaron
	UMF Umeå	L. Wennberg A. Hagström
	Inst. System Ecology, Askö	T. Lundberg
Finland	FIMR Helsinki	J.- M. Leppänen I. Kuparinen L. Grönlund
Denmark	Inst. Environmental Research	G. Aertebjerg I. Nielsen
USSR	State Oceanogr. Inst., Moscow	V. Andrjuschenko
FRG	Inst. Marine Research	A. Helms P. Fritsche
GDR	Inst. Marine Research	S. Schulz (Convener)

3.2 Primary Production Experiments

3.2.1 During the Workshop at 28 and 29 August two different experiments, A and C, were carried out.

In Experiment A, similar to the monitoring procedure every laboratory used its own 14-C-solution, filters as well as equipment and counted the activity of the filters with their own counter.

In Experiment C, all laboratories used the same 14-C-solution and type of filters supplied by the organizer. The activity of the filters was measured at an scintillation counter on board RV ARANDA.

In all experiments ten parallel light and three dark samples were used. The experimental water for the exclusively used mixed samples was kindly prepared by G. Aertebjerg and colleagues. The water was gathered at a site in the Sound (for experiments at 28 August) and at a site 12 nm off Visby at 29 August, where ships carried out field sampling.

3.2.2 Beside the incubator experiments three laboratories, supplied with the corresponding filter equipment, performed experiments for comparison of PI-curve estimation.

3.2.3 The participating Swedish labs from Umeå and Askö carried out in situ measurements at the experimental site off Visby at 29 August. Results of this exercise, hardly in any case to compare with the workshop procedure, have not been received.

3.3 Additional experiments and measurements

3.3.1 The irradiance of the incubators was measured. The results are compiled in Tab. 1.

3.3.2 At 29 August all ships observed the sight depth by using the Secchi disk. The results of the readings are compiled in Tab. 1.

3.3.3 The activity of the 14-C-solution used by the participating laboratories was checked. The activity is given in Tab. 2.

3.4 Results

3.4.1 Experiment A

The results of experiment Al are compiled in Fig.1.
The USSR did not take part in this experiment. The median values covered a range from 9.3 mg C m^{-3} h^{-1} (RV ALKOR) to 13.8 mg C m^{-3} h^{-1} (RV ARANDA). The overall average accounted for 11.73 mg C m^{-3} h^{-1} with a standard deviation and a coefficient of variation of 1.95 and 16.61% respectively. The 95% confidence intervals are displayed in Fig. 2. They show that at least two averages are significantly different from three others.
In Fig.3 the results of experiment A2 are given. The median values ranged from 9.2 mg C m^{-3} h^{-1} (RV ARGOS) to 13.3 mg C m^{-3} h^{-1} (RV GUNNAR THORSON). The overall average was 11.37 mg C m^{-3} h^{-1} with standard deviation and coefficient of variation of 1.87 respectively 16.42%.

In Fig.4 the 95% confidence limits of the averages are given. They show at least two groups with significantly different means.

3.4.2 Experiment C

In Experiment Cl RV ARANDA did not take part. The results are shown in Fig.5. The median values ranged from 7.4 mg m^{-3} h^{-1} (RV LEV TITOV) to 17.1 mg C m^{-3} h^{-1} (RV GUNNAR THORSON).

The overall average was 12.25 mg C m^{-3} h^{-1} with the corresponding standard deviation 4.29 and coefficient of variation 35.03 respectively.

The averages and 95% confidence intervals in Fig.6 exemplify the differences between the means and show that 5 averages are not significantly differing from each other.

The results of experiment C2 are compiled in Fig.7. The median values ranged from 7.5 mg C m^{-3} h^{-1} to 13.5 mg C m^{-3} h^{-1}. The overall average accounted for 10.54 mg C m^{-3} h^{-1} with the standard deviation of 2.96 and the coefficient of variation of 28.06 respectively.

In Fig. 8 the averages and the 95% confidence intervals are shown. Again only 4 respectively 3 means are not significantly different from the others.

3.4.3 PI-curve estimation

The results of the two experiments are compiled in Tab. 3.

3.5 Discussion

The results of the four experiments (Al, A2, Cl, C2) are rather confusing. First of all the averages of the different laboratories differ in some single cases and mostly also pooled as small groups (2-3 labs) from the others. The standard deviations are also considerable in the experiments and also different from experiment to experiment. The high variability within the groups might be caused by differences in filtration technique. At least one lab noted long and uneven filtration time. Generally the participants complained that the filtration of the material caused when by using the SARTORIUS filters.

Unexpected differences are also visible in the results of the corresponding experiments Al, Cl and A2, C2. The grand mean in the first experiment was higher for C in the second for experiment A (Tab.4). The means are however statistically not significantly different.
The averages at least of the four labs which took part in both Al and Cl differ considerably and changed also the pattern "high to low" or opposite for the means.
Opposite to the first attempt the pattern "high to low" in the means of the second experiment is comparable for the A and C parts.
If experiment A and C is compared, the experiment A showed a better agreement between the participating labs expressed as a lower coefficient of variation.

3.6 Conclusions

The variability in the results between the participating labs was surprisingly high. This is possibly caused by the physiological state of the phytoplankton which had to be used for preparing the samples.
The agreement between the labs was in experiment A (the normal in the BMP used procedure) better than in experiment C. This fact gives rise to the assumption that the data in the bank are better than the results of the intercalibration.

Tab.1 Secchi disk readings at the intercalibration site and Irradiances in the incubators. Mean values of measurements at top, bottom, right and left side

Lab	Secchi disk m	Irradiance uE m^{-2} s^{-1}	
ALK/FRG	8	420	
ARG/S	7	290	
ARA/SF	7	420	1.run
		410	2.run
GT/DK	8	420	l.run
		370	2.run (2 h later)
LT/Lithuania/USSR	7	270	
PAP/GDR	7.5	320	

ALK = RV "Alkor"
ARG = RV "Argos"
ARA = RV "Aranda"
GT = RV "Gunnar Thorson"
LT = RV "Lev Titov"
PAP = RV "Professor Albrecht Penck"

Tab.2 Activity of the C-14-solutions used by the participating labs (measured on board RV ARANDA)

Activity Lab	uCi ml^{-1}
ARA/SF	20.09
ALK/FRG	10.39
ARG/S	4.17
GT/DK	17.85
PAP/GDR	23.99

The Finnish colleagues noted that the batch E 70-l given by GT for the C-Experiment was certified with 19.48 uci ml^{-1}.

Tab.3 Irradiance in % of the normal irradiance in incubator

PP Lab.	5	10	15	25	50	100	175
ALK (Exp.1)	0.67	2.26	-	6.82	11.73	20.67	18.31
GT (Exp.1)	0.05	0.78	1.79	4.38	9.00	11.45	9.82
ARA (Exp.2)	0.16	1.55	-	24.13	9.22	10.36	10.83
ALK (Exp.2)	0.60	1.79	2.92	5.78	9.56	13.15	13.86
GT (Exp.2)	0.52	1.45	2.72	5.08	9.46	6.08	7.11

All data calculated as mg C m^{-3} h^{-1}

Tab.4 Grand means, standard deviation (mg C m^{-3} h^{-1}) and variation coefficient for the four experiments

	Exp. Al	Exp. Cl	Exp. A2	Exp. C2
Grand mean	11.73	12.25	11.37	10.54
Stand. dev.	1.95	4.29	1.87	2.96
Var. coeff.	16.61	35.03	16.42	28.06

Fig.1 Box-Whisker-plots of the results of Experiment AI.
The boxes represent the median as the horizontal line inside, the upper and lower quartil and the range of the samples as the vertical tine. L.T. did not take part in Experiment AI.

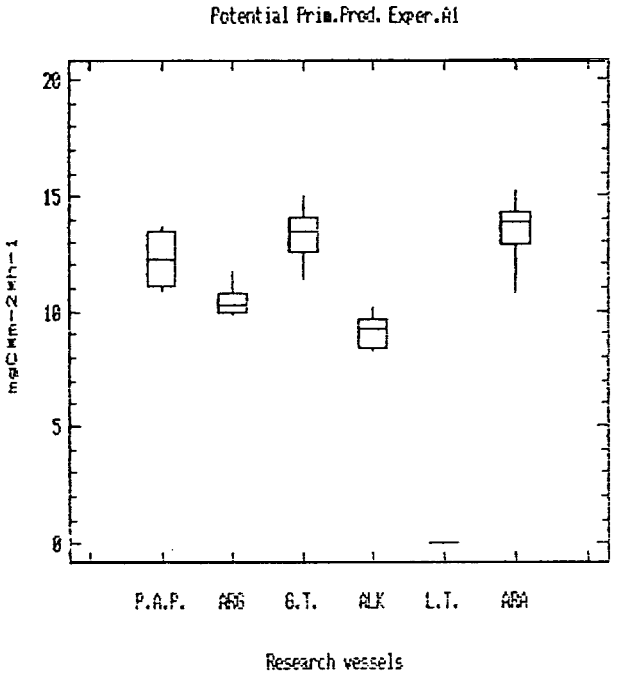


Fig.3 Box-Whisker-plots of the results of Experiment CI. Explanations as in Fig.1. RV ARA did not take part in the Experiment CI.

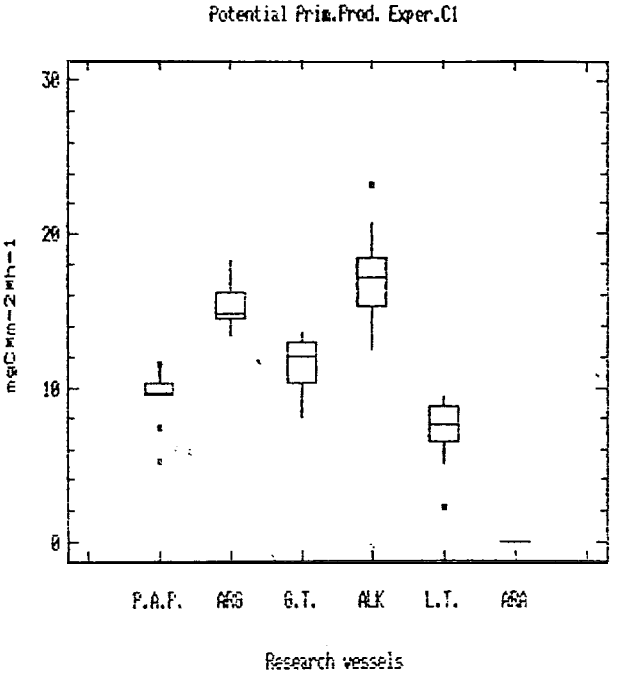


Fig.2 95% confidence intervals and mean values of Experiment AI for the participating labs.

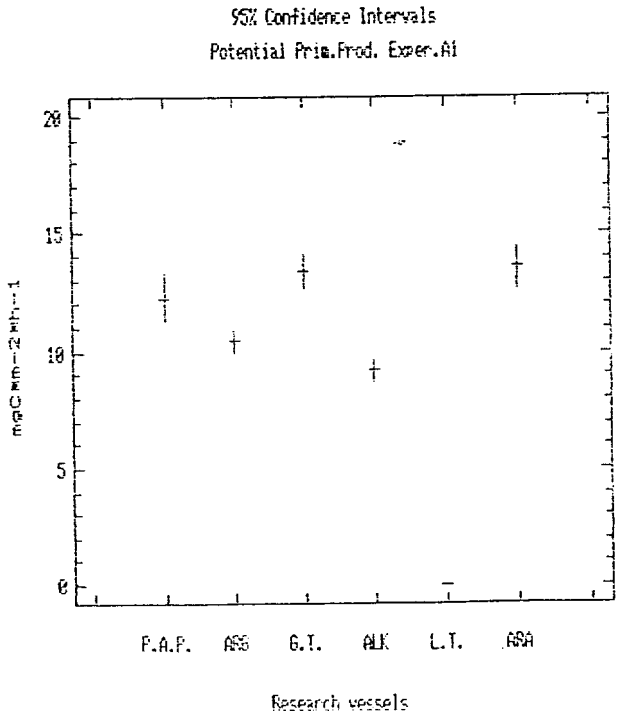


Fig.4 95% confidence intervals and means for Experiment CI.

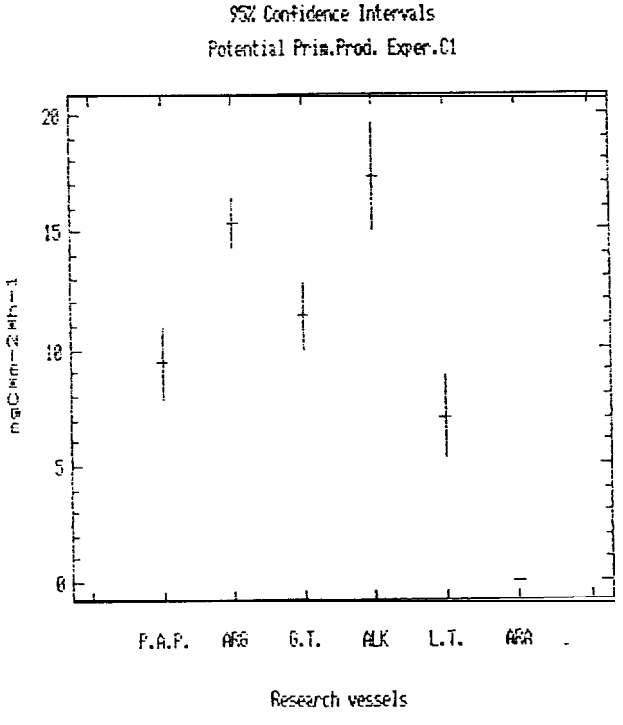


Fig.5 Box-Whisker-plots Of theresults Of Experiment A2. Explanations as in Fig.1.

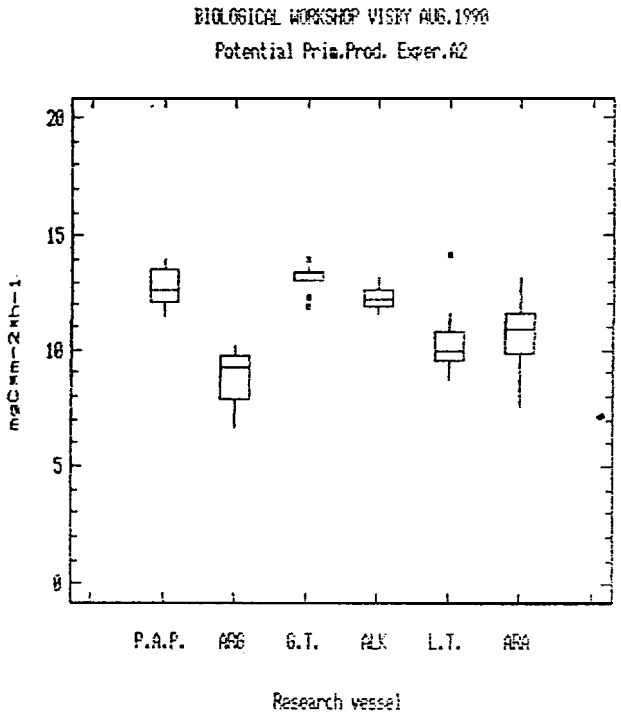


Fig.7 Box-Whisker-plots of the results of Experiment C2. Explanations as in Fig.1.

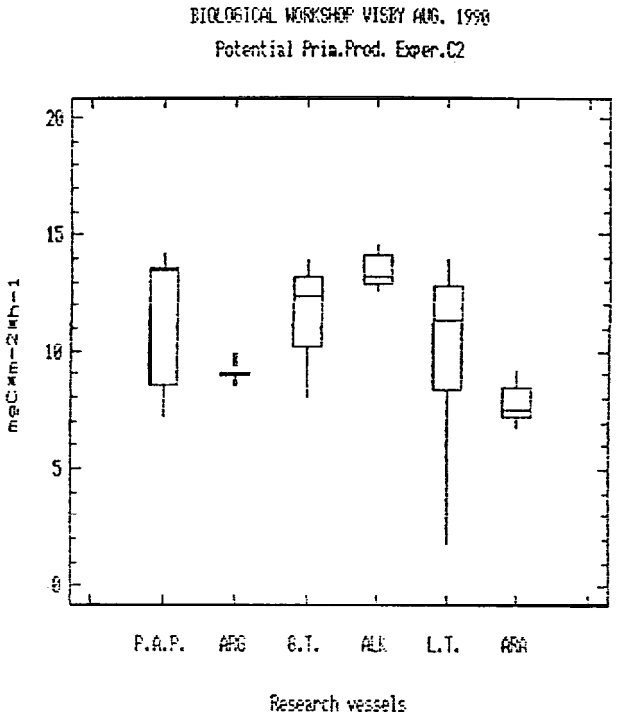


Fig.6 **95% confidence intervals and means for Experiment A2.**

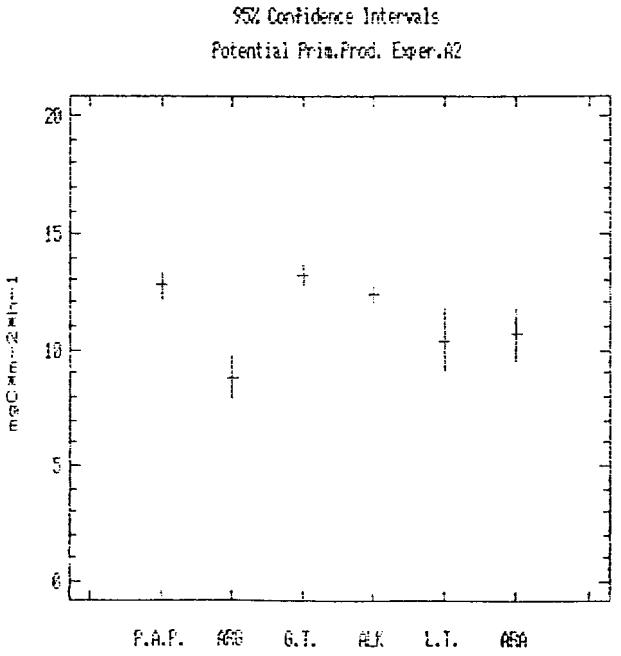
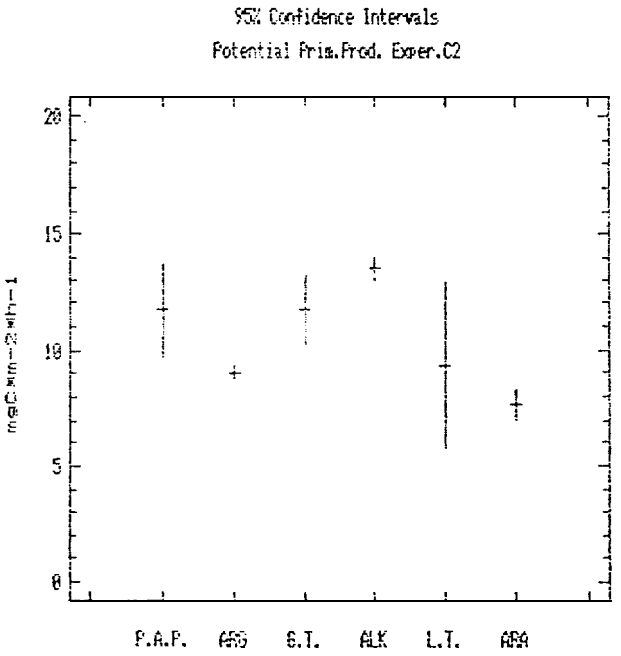


Fig.8 **95% confidence limits and means for Experiment C2.**



4. REPORT OF THE WORKING GROUP ON ZOOPLANKTON

4.1 Introduction

One of the factors influencing the results of a zooplankton sample counting is the type of the splitter used for producing the subsamples. In the BMP - Guidelines, the use of either Kott- or Folsom-splitters is recommended (HELCOM 1988), but an intercalibration of these instruments never took place. There was only a short remark in the report of the Intercalibration Workshop in Ronne (HELCOM 1983). On the other hand, it is known that splitters might influence the results quite well /(see for example GUELPHEN et al., 1982). So, it was decided to focus on this topic during the intercalibration workshop in Visby.

4.2 Members, Material and Methods

The following laboratories were represented in the zooplankton working group:

1. IfM Kiel, Diisternbrooker Weg 20, D - 2300 KIEL
   (Gerda Behrends) (in the text referred to as lab. 1)
2. IfM Warnemünde, Seestr. 15,
   D - 2350 ROSTOCK-WARNEMUNDE (Gunther Breuel) (lab. 6)
3. Finnish Institute of Marine Research, P.O.Box 33,
   SF- 00931 HELSINKI (Markku Viitasalo, Soili Saesmaa)
   (lab. 4)
4. Centre for Marine Research , University of Stockholm,
   S – 10691 STOCKHOLM (Sif Johansson, Helen Bjurulf) (lab. 2)
5. Swedish Environmental Protection Agency, P.O.Box 7050,
   S – 75007 UPPSALA (Cornelia Sellei) (lab. 5)
6. National Environmental Research Institute,
   Jaegersborg Allee 1B, DK - 2920 CHARLOTTENLUND
   (Louise Schlueter) (lab. 3)

Three steps and levels of analysis were performed:

1. Five parallel hauls were taken from the ships which were keeping position as close together as possible, and from each of the hauls one sample was treated like normally done within the monitoring work. One subsample counting of every haul was reported (Experiment A).

2. Another sample was divided to ten parts, one of which was distributed to be treated on like usually done in laboratories. Four subsample countings were reported (Experiment B).

3. A sample was divided into countable concentrations by one (Kott-)splitter and three of the subsamples were distributed to each of the members to be counted directly without any further treatment (Experiment C).

With this structure of the experiment, the influence of the different levels of error-sources (1. natural patchiness, ship equipment and sampling , 2. preservation, filtering and subsampling, and 3. counting) should be detectable.

The counting results were expressed as concentrations (Ind * m^-3). Most of the analyses were carried out with the "group-results", that is with sum of adult copepods and copepodides, copepcod nauplii, cladocerans, rotifers and meroplankters. Only the three directly distributed subsamples and 1 of the other sets were counted to species levels in order to get information about existing difficulties in determining.

4.3 Results

The results are shown in Fig. 1 - 3, and statistics of the experimental sets are summarized in Tables 1 and 2.

The means of total zooplankton abundance are varying most in the exp. A (range = 30 739) and are most equal in the exp. B (range =11 419). With a range of 15 080, the exp. C also shows unexpected high variability (Fig. 1). Within the laboratories, the coefficient of variation of exp. C exceeds even that of the

exp. A (6 - 27 % resp. 8 - 14 %). In the exp. B set, coefficients of variation are between 2 and 8 % (Table 1 and 2).

If the results are grouped to Kott- and Folsom-splitter- users, the analysis of variance shows no significant differences between them. Within the groups significant differences are found only in the Folsom-splitter-users group. They are caused by clearly lower numbers that were reported from two of the labs (fig. 1). As this phenomenon is not observable in the exp. C data, and it was more pronounced in exp. A than in exp. B, it has to be connected with the splitting procedure of the laboratories. It is, however, impossible to detect a graduate difference connected with the number of splitting steps. The lower values of the two laboratories are found in all taxonomic groups with exception of the copepod nauplii, which belonged to the rarest taxa. There is no clear difference in the species composition found by these labs, so an explanation of the differences in terms of under- or overestimation of certain taxa is impossible (see table 4).

Table 4: Percentage of taxonomic groups:
comparison of labs using Kott- vs. labs using Folsom-splitters

	Kott			Folsom		
	FRG	Askoe	Denmark	Finland	Sweden	GDR
Exp. A						
Copepods	18.86	-	16.21	13.14	13.27	19.42
Nauplii	3.38	-	5.20	5.34	8.62	3.10
Cladocera	69.04	-	73.36	77.06	72.64	73.20
Rotifers	8.44	-	5.15	4.32	4.92	4.28
Meroplank.	0.28	-	0.08	0.14	0.55	-
Exp. B						
Copepods	17.87	19.64	18.95	18.81	19.08	16.51
Nauplii	1.63	2.85	3.45	3.71	3.13	3.81
Cladocera	77.29	73.20	73.83	72.57	74.05	77.14
Rotifers	2.99	4.03	3.61	4.72	3.35	2.54
Meroplank.	0.18	0.28	0.16	0.19	0.39	-

	FRG	Kott Askoe	Danmark	Finland	Folsom Sweden	GDR
Exp. C						
Copepods	18.37	18.83	16.13	17.26	18.12	19.21
Nauplii	1.25	3.71	2.59	3.43	2.73	2.60
Cladocera	77.91	73.47	75.41	73.34	73.96	74.72
Rotifers	2.03	3.91	5.78	5.79	4.78	3.47
Meroplank.	0.18	0.08	0.09	0.18	0.41	-

The species determination is not a big problem at all. Table 3 shows the results of the exp. C for all of the laboratories. The coefficient of variation, when compared to those found in the respective taxa and labs in Ronne (HELCOM 1983), are often lower, mostly in the same order of magnitude and only with few exceptions higher than in 1982. These have always very low numbers. It can be stated, that if less than 20 animals are really counted in the subsamples, the coefficient of variation for these taxa starts to exceed 30 % and the data are not quantitative anymore.

4.4 Discussion

The highest variability was found in the set of data where the lowest were to be expected. The reasons for this must be looked for in the splitting procedure, as the splitter itself does not show such high deviations from subsample to subsample. Fig. 4 shows a combination of data sets: it is evident that the Kott splitter used to produce the subsamples is able to work much better, and on the other hand due to the fact that the variability in the Exp. B data is much lower, the counting error cannot be the reason for the high variability. Most probably the time of settling of the sample in the splitter wasn't long enough, as all subsampling had to be done at one afternoon. The better results of the lab 1, which is normally using that splitter, in experiment B also show that the splitter itself is working better. Though the results of exp. C are not satisfactory in this respect, they can give a good overview over the determination

problem. The species composition in all sample set results coincide quite good. Compared to the results of the BIW II in Ronne 1983 (HELCOM 1983), the coefficients of variation for most of the taxa and most of the laboratories are either lower or in the same order of magnitude with only few exceptions. These can be explained by the low numbers of specimen that have been counted.

As already stated in the Ronne intercalibration paper (HELCOM 1983), the coefficients of variation are increasing rapidly with decreasing numbers of specimen counted in the subsamples. If less than 20 animals are counted, the cv% lies over 30 % , and the results are not usable for quantitative analyses. On the other hand, especially the rare species can give valuable information about the state of the ecosystem. So, it should be recommended to report these data further on, but label them as qualitative ones. Quantitative analyses should be based exclusively on data with better comparability.

The clearly lower number (compared to the mean) obtained by two of the laboratories using Folsom-Splitter must be connected with some systematically error within the procedure. The third laboratory with Folsom-splitter did not get these low numbers, but the opposite was true: the results of this one were always slightly above the mean. So, it cannot be stated that Folsom-splitters lead to lower values, but the variability between the laboratories seems to be higher in the Folsom-splitter-user-group.

Regarding the accuracy of the results within the laboratories, all of them were in a quite good range. So, in interpreting data from the Baltic as a whole, the analyses should be regarded at as relative ones, being very significant when based on results of one laboratory, but in the case of stations which are sampled by different laboratories the results may not always be comparable. The idea that came up in the zooplankton working group to send all samples of certain stations to certain laboratories to be counted there with the same methods should be discussed very seriously, as it became supported by these results.

4.5 Recommendations

- Data based on countings of less than 20 animals per taxon in the subsamples should be labeled as being qualitative when reported to the HELCOM data bank. They should not be included in purely quantitative analyses. On the other hand, as they are of great importance for the assessment of the state of the ecosystem, they should be reported further on.

- Results obtained by different laboratories may have significant errors. Results from one laboratory are of a very good quality and comparability. Therefore it has to be discussed very seriously, whether the samples from certain stations which are sampled by different laboratories should be sent to certain laboratories to be treated there with always the same methods. The results can be regarded as relative ones with high significance.

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Table 1: Comparison of statistical values of the 3 experiments
SD = standard deviation, SE = standard error, CV % = coefficient of variation,
95 % CL = 95 % confidence limits (STUDENT)

	TOTAL ABUNDANCE			COPEPODS+COPEPODIDES			COPEPOD NAUPLII			CLADOCERA			ROTIPERA		
Experiment	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
Sample size	25	24	18	25	24	18	25	24	18	25	24	18	25	24	18
Range	43213	14905	33777	6301	4778	4800	2560	1661	1778	34702	10070	29778	3369	2560	3733
Average	38165	38368	48440	6073	7103	8687	1919	1182	1309	28035	28626	36262	2067	1377	2109
SD	13286	4546	8620	1982	1238	1039	899	430	464	10649	3247	7401	970	637	1096
SE	2657	928	2032	396	253	245	180	88	109	2130	663	1745	194	130	258
cv %	35	12	18	33	17	12	47	36	36	38	11	20	47	46	52
95 % CL	±5484	±1920	±4288	±817	±524	±517	±372	±182	±230	f4396	±1372	±3682	f400	±269	±544

Table 2: Comparison of statistical values of the laboratories in the 3 experiments (abbreviations see table 1)

	Experiment A					Experiment B					Experiment C				
Variable	Mean	SD	SE	cvx	95%CL	Mean	SD	SE	cvx	95%CL	Mean	SD	SE	CVX	95%CL
TOTAL ABUNDANCE															
lab. 1	37370	3676	1644	10	±4564	40911	1279	639	3	±2033	49630	3036	1753	6	±7549
lab. 2						39711	3196	1598	8	±5085	44683	3158	1824	7	±7851
lab. 3	50161	4417	1976	9	±5485	40000	2082	1041	5	±3313	58430	7719	4456	13	i19179
lab. 4	54395	4394	1965	8	±5455	43705	1304	652	3	±2075	50133	9612	5504	19	±24034
lab. 5	23656	3405	1523	14	±4228	32286	531	265	2	±843	43350	11567	6678	27	i28742
lab. 6	25236	3635	1626	14	±4514	33593	2825	1413	8	±4496	44415	9221	5324	21	i22915
COPEPODS (-NAUPLII)															
lab. 1	7049	171	77	2	±214	7311	983	491	14	i2994	9244	541	312	6	±1343
lab. 2						7000	694	347	9	±1104	8415	336	194	4	±835
lab. 3	8133	1199	536	15	±1488	7578	659	329	9	±1047	9422	1456	841	16	±3620
lab. 4	7145	970	434	14	±1205	8220	486	243	6	±773	8652	983	568	11	±2445
lab. 5	3138	198	89	6	±247	6160	134	67	2	±213	7853	1766	1020	23	±4390
lrb. 6	4902	840	375	17	±1041	5547	1593	797	29	±2536	8534	308	178	4	±766
COPEPOD NAUPLII															
lab. I	1262	448	200	36	±555	667	277	138	42	±439	633	178	103	29	±443
lab. 2						1133	380	190	34	±605	1659	185	107	11	±461
lab. 3	2608	645	289	25	±802	1378	401	200	29	±632	1511	615	355	41	±1528
lab. 4	2904	204	91	7	±253	1622	452	226	28	• 719	1719	185	107	11	±461
lab. 5	2038	370	166	18	±461	1010	256	128	25	±407	1185	185	107	16	±461
lab. 6	783	163	73	21	±203	1280	215	108	17	±344	1156	235	136	20	±585
CLADOCERA															
lab. 1	25804	3493	1562	14	±4336	31622	961	481	3	±1531	38667	3244	1873	8	±8061
lab. 2						29067	2225	1113	8	±3542	32830	3133	1809	10	±7786
lab. 3	36790	5497	2458	15	±6823	29533	1622	811	6	±2581	44059	6338	3659	14	±15748
lab. 4	41916	3606	1613	9	±4478	31716	1585	793	5	±2523	36770	8556	4940	23	±21262
lab. 5	17183	3162	1414	18	±3925	23908	707	254	3	±808	32060	10401	6005	32	±25845
lab. 6	18473	3237	1448	18	i4020	25913	1940	970	8	±3087	33185	8070	4659	24	±20052
ROTIPERA															
lab. 1	3156	505	226	16	±627	1223	432	216	35	±687	1007	312	180	31	±775
lab. 2						1600	654	327	41	f1041	1748	536	310	31	±1334
lab. 3	2585	888	397	34	±1102	1445	786	393	54	±1251	3378	759	438	23	±1885
lab. 4	2348	575	257	25	±713	2063	636	318	31	±1012	2904	1406	812	48	±3495
lab. 5	1165	182	81	16	±225	1081	191	96	18	±306	2074	951	549	46	±2363
lab. 6	1079	299	134	28	±372	853	476	238	56	±757	1541	713	412	46	±1773

Table 3: Counting results of the laboratories, Experiment C (n = 3)
(f = female, m = male, 1 = copepodid I - III, 2 = copepodid Iv - v, n = nauplii)

Land/Labor	IfM Kiel, FRG			Askoelab, S			Danmark			Finland			Sweden			IfM Warnemuende			all labs		
	lab. I			tab. 2			lab. 3			lab. 4			lab. 5			Lab. 6					
species	mean	std	cot	mean	std	cv%	mean	red	cv%	mean	std	cv%	mean	std	cv%	m e n	std	cv%	mean	std	co:
Acartia spp.1	919	312	34	415	136	33	356	154	43	533	308	58	296	223	75	1007	337	33	588	357	61
Acartia 9pp.2	1422	235	17	1007	103	10	1570	420	27	1422	356	25	1185	135	11	1452	51	4	1343	289	22
Acartia bifilosa f	1244	470	38	1244	320	26	1244	a9	7	1393	359	26	1096	667	61	1215	103	8	1240	340	27
Acartia bifilosa m	652	185	28	533	178	33	444	320	72	237	185	78	474	136	29	474	136	29	469	211	45
Acartia longiremis f	207	la5	a9	119	136	115				119	51	43	356	356	100				130	136	105
Acartia spp. n							533	320	60	711	178	25	503	103	20	356	89	25	526	356	68
Centropages hamatus 1	a9	a9	100	a9	a9	loo	a9	a9	100	30	51	173	178	a9	50	415	136	33	148	152	103
Centropages hamatus 2	326	51	16	178	235	132	178	154	a7	148	103	69	178	0	0	207	136	65	203	129	64
Centropages hamatus f	59	103	173	178	a9	50	30	51	173				89	89	100	89	89	100	72	86	118
Centropages hamatus m	59	51	a7	59	51	a7	59	51	87				148	103	69	30	51	173	59	68	115
Centropages hamatus n							30	51	173	59	51	87	89	89	100	30	51	173	44	46	103
Eurytemora affinis 1	30	51	173	59	51	87	30	51	173	474	185	39	178	154	a7				128	191	149
Eurytemora affinis 2	207	185	a9	207	103	49	563	286	51	385	103	27	267	259	97	237	136	57	314	212	68
Eurytemora affinis f	415	51	12	296	136	46	385	286	74	444	0	0	326	256	79	356	0	0	370	150	41
Eurytemora affinis m	593	136	23	711	356	50	593	185	31	267	0	0	356	267	7s	504	51	10	so4	232	46
Eurytemora affinis n							89	89	100	1215	490	40	178	a9	50				370	499	135
Pseudocalanus mi.el.1	978	470	48	1185	370	31	a30	205	25	385	224	58	978	178	18	444	235	53	800	392	49
Pseudocalanus mi.el.2	356	0	0	267	a9	33	178	154	87				296	206	96	148	103	69	207	170	82
Temora longicornis 1	385	312	81	415	185	45	474	51	11	504	103	20	415	185	45	533	235	44	434	175	38
Temora longicornis 2	681	224	33	652	136	21	948	370	39	a30	312	38	652	543	83	711	0	0	746	288	39
Temora longicornis f	148	51	35	296	286	96	563	136	24	237	205	a?	134	104	78	356	89	25	298	210	71
Temora longicornis m	474	136	29	504	224	4 4	622	89	14	237	224	94	652	103	lb	356	178	50	474	205	43
Temora longicornis n							859	272	32	741	286	39	so4	286	57	770	224	29	119	470	66
Copepod nauplii	622	178	29	1659	185	11	267	154	58										510	628	123
Bosmina cor.mar.	37422	2925	8	32119	3193	10	42430	6215	15	35526	8621	24	31111	10007	32	32030	7637	24	35106	7116	20
Evadne sp.	770	337	44	237	136	57	770	la5	24	681	51	a	652	591	91	415	337	81	588	340	58
Podoa sp.	474	103	22	474	51	11	859	185	22	563	51	9	296	135	46	741	337	45	568	240	42
Bivalve larvae	89	0	0	30	51	173	30	51	173	59	51	a7	148	51	35				59	61	103
Gastropod							30	51	173	30	51	173	89	a9	100				27	36	136
Keratella sp.	1007	312	31	1748	536	31	3289	902	27	2815	1494	53	2074	951	46	1481	667	45	2069	1096	53
Synchaeta							89	154	173	a9	89	100				59	51	87	79	86	109
Oikopleura dioica	30	51	173				30	51	173										12	29	243
SUMME	50074	3025	6	44681	3158	7	58459	7698	13	50133	9672	19	43350	11567	27	44415	9221	21	48519	0636	18

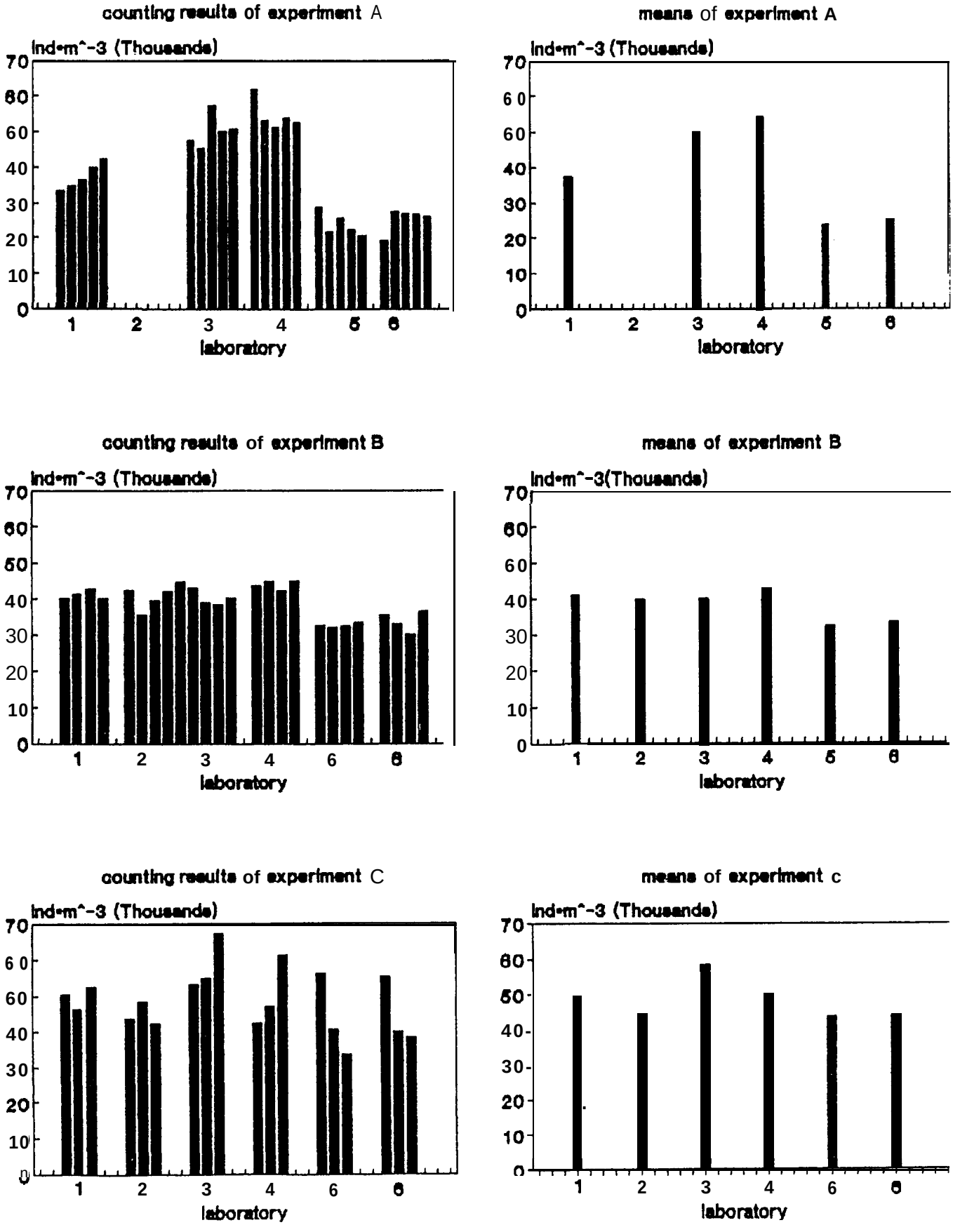
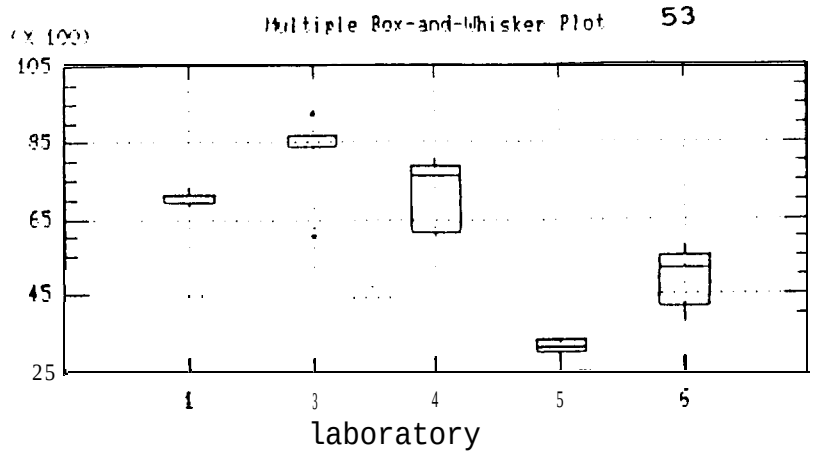
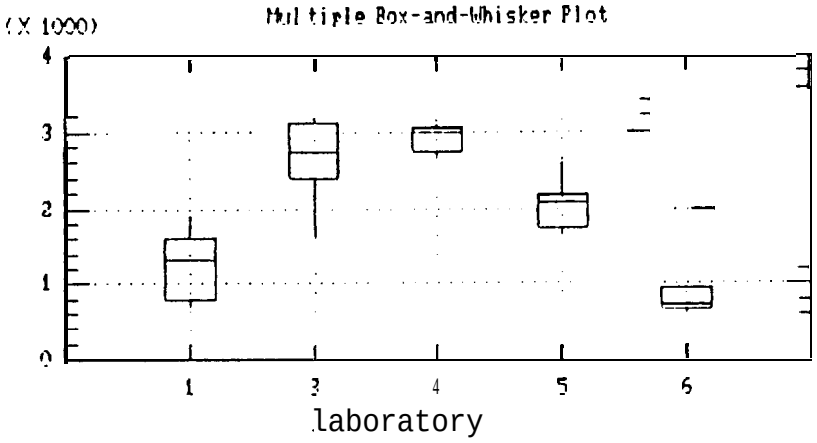


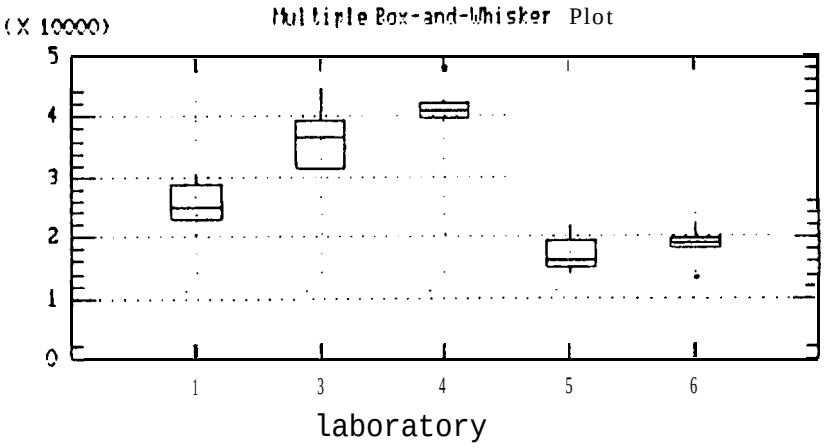
Fig. 1: comparison of the counting results and means of the 3 experiments, total abundances



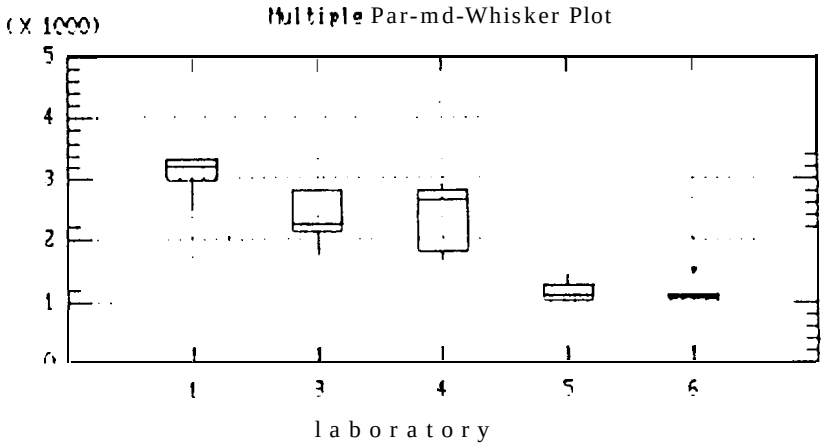
a) adult copepods and copepodides



b) copepod nauplii

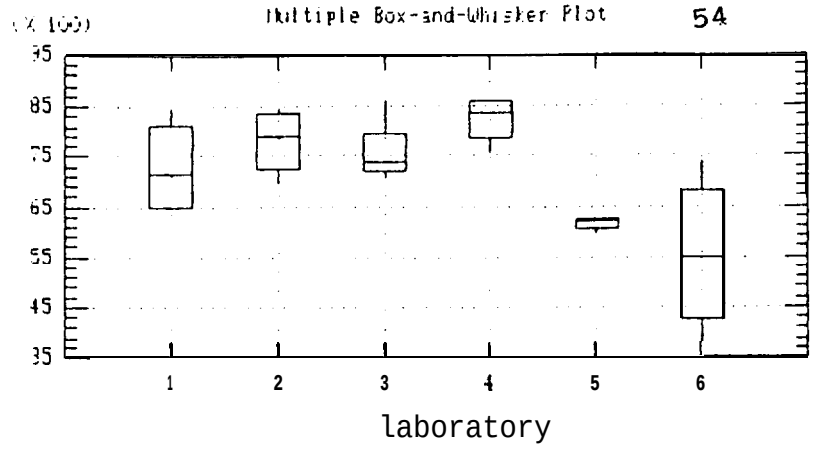


c) cladocera

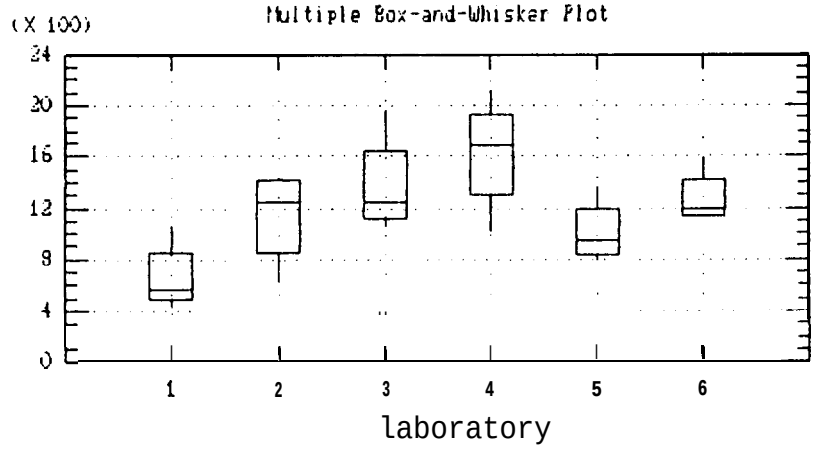


d) rotifera

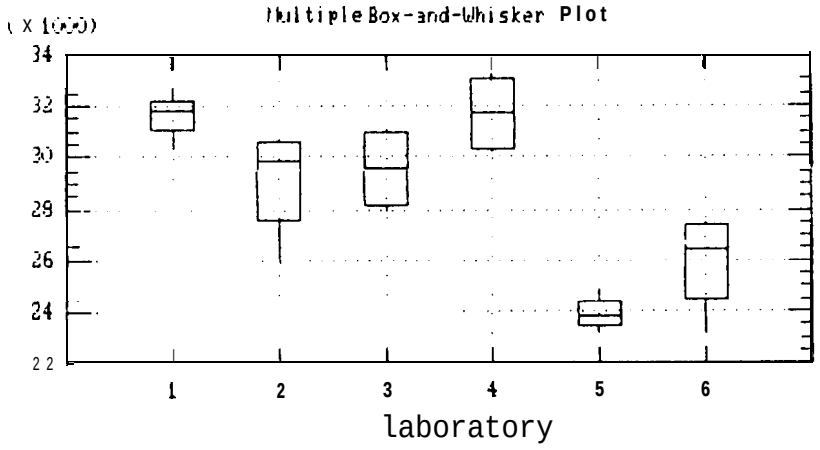
Fig. 2: Box- and Whisker-Plots of the "group results" of experiment A (Boxes indicate upper and lower quartile, horizontal lines mark the median, asterixes outliers, more than 2x standard deviation)



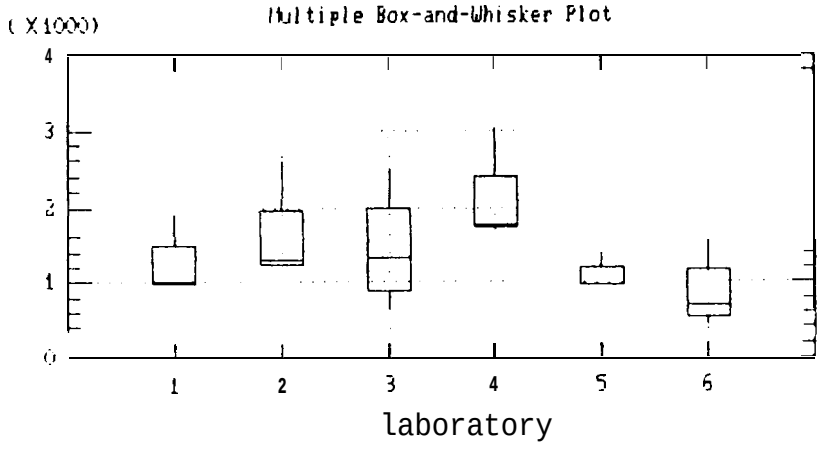
a) adult copepods and copepodides



b) copepod nauplii



c) cladocera



d) rotifera

Fig. 3: Box - and Whisker-Plots of the "group results" of experiment B (Boxes indicate upper and lower quartile, horizontal lines mark the median, asterixes outliers, more than 2x standard deviation)

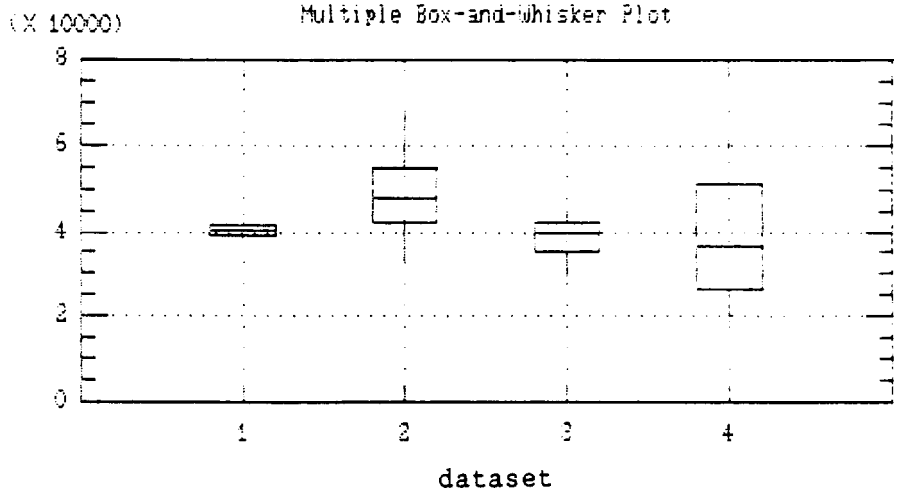


Fig. 4: Box- and Whisker- Plot of datasets.
set 1: calibration of the Kott-splitter used for the basic splitting (n = 10)
set 2: results of experiment C (n = 18)
set 3: results of experiment B (n = 24)
set 4: results of experiment A (n = 25)
Boxes indicate upper and lower quartile, horizontal line median, vertical line data range.

5. REPORT OF THE WORKING GROUP ON CHLOROPHYLL-A

5.1 Participating laboratories

STATE	SHIP	LABORATORY/PERSON
D	Alkor	Institute of Marine Research, Kiel, P. Fritsche
DDR	Prof. Albrecht Penck	Institute of Marine Research, Warnemünde, S. Schulz
DK	Gunnar Thorson	National Environmental Research Inst., Div. of Marine Ecology and Microbiology, G. Ærtebjerg
SF	Aranda	Finnish Institute of Marine Research, J.-M. Leppänen
S	Argos	Swedish Meteorological and Hydrological Institute, Oceanographical Laboratory, A. Taglind
S-Askö	Coast Guard 04	University of Stockholm, Askö laboratory, T. Lundeberg
S-Umeå	Coast Guard 04	University of Umeå, Marine Research Center, L. Wennberg
USSR	Lev Titov	Hydrometeorological Observatory of Klaipeda, J. Dubra

5.2 Introduction

The chlorophyll-a concentration is calculated from spectrophotometer measurements at the wavelength of the maximum absorption of chlorophyll-a and a given specific absorption coefficient for chlorophyll-a in the actual extraction solvent. Chlorophyll-a standards of known concentrations are not used in the routine measurements of chlorophyll-a concentrations, and fluorometer determinations are calibrated to spectrophotometer determinations.

In the routine methods for analysis of chlorophyll-a concentrations it is provided that the spectrophotometer measurements are

done with a narrow bandwidth and exactly at the peak of the chlorophyll-a absorption maximum. If the bandwidth is too broad or the used wavelength is not at the chlorophyll-a absorption peak, lower concentrations will be found. This means that the laboratories finding the highest mean values, when measuring the same sample, are closest to the true value.

5.3 Material and Methods

The design of the chlorophyll-a intercalibration was set up to analyze the comparability of the results from different laboratories, and the influences of different spectrophotometers, filtration procedures and of storage on the chlorophyll-a measurements. Ethanol was used for extraction by all laboratories except one (DDR), which used acetone. Another laboratory (Askö) used both solvents. This made it possible to compare the efficiency of ethanol and acetone as extraction solvents. One laboratory (SF) stored samples both as extracts and as dried filters, which made it possible to compare the way of storage. Spectrophotometers were used for analyses except by one laboratory (SF), which used a fluorometer. Four different samples were delivered to the participating laboratories during the Workshop.

5.3.1 Mixed samples

August 26 Gunnar Thorson made up a mixed sample of surface water from the southern Øresund. August 28 each laboratory filtered 20 subsamples. Ten of these were analyzed the next day, the other ten after one month storage in a freezer.

August 29 Gunnar Thorson made up another mixed sample of surface water from the Workshop Field Station, and subsamples were at once distributed to the other laboratories. The analyses were generally the same as mentioned above, but with the possibility to use own procedures, if different from the method in the Guidelines for the Baltic Monitoring Programme for the Third Stage.

5.3.2 Prepared extracts

DK delivered ten subsamples of a chlorophyll-a extract made by ethanol extraction from spinach to each laboratory, which analyzed the extract during the Workshop. However, the extract contained much water from the spinach, and the readings at 750 nm were very high. Therefore a new extract was prepared from a phytoplankton culture by SF, and five subsamples of this delivered to each laboratory.

5.3.3 Analysis of variance

For the evaluation of the intercalibration results, analysis of variance has been used to test the hypothesis, that all mean values in an experiment are equal, independent of laboratory and/or treatment. The basis in the analysis is that each single measurement of the chlorophyll-a concentration can be calculated as:

Conc. = (grand mean) +/- (between lab. variance) +/- (within lab. variance).

If all mean values are equal the F-ratio = (between lab. variance)/(within lab. variance) will be close to 1.00. The larger F-ratio, the larger is the difference between mean values. If the probability of obtaining an actual F-ratio by chance alone is less than 5 %, the hypothesis that the mean values are equal is rejected. (The statistical edp-programme SAS has been used for the analysis).

Three very deviating single values have been omitted from the analyses. These are: Øresund sample during Workshop Umeå 1.20 ug/l, Field Station during workshop S 1.63 ug/l and DK-extract S 3.05 ug/l.

5.4 Results and Discussion

The overall results of the chlorophyll-a intercalibration is given in Table 1 as mean, standard deviation and coefficient of variation for each laboratory and series of subsample. The

standard deviations and coefficients of variation are generally low. This shows that the precision (ability to reproduce the measurements) within the laboratories generally is high, but tells nothing about the accuracy (ability to get close to the true concentrations) of the measurements. In the mixed sample experiments the highest variations were found by USSR, followed by **Umeå** and **Askö.**

In Table 2 is shown the mean values found by the different laboratories in each experimental series. The highest mean value in each experiment is given the letter A, the next highest significantly different mean value is given the letter B and so on. If the same letter is given to two or more mean values in an experiment they are statistically equal. If a mean value is given two letters this means, it is not significantly different from other mean values in the experiment given at least one of the letters.

The good precision within the laboratories causes, that even small differences in mean values between laboratories become statistically highly significant. Therefore only very few mean values from different laboratories are statistically equal. This is true even for the prepared extract analyses. In the mixed sample experiments the highest and most comparable mean values are generally found by SF, DK, D and S, followed by DDR, while the lowest are found by USSR, **Askö** and **Umeå.** In the prepared extract analyses only **Askö** and partly **Umeå** are low, while the USSR is at the same level as SF, S, DK, DDR and D.

5.4.1 Spectrophotometers -- Filtration **procedure**

The standard deviations in the extract experiments are generally lower than in the mixed sample experiments. This means that the filtration and extraction procedures increases the variation compared to analyses on prepared extracts, but for most of the laboratories this increase in variation is low. Some of the variations within the laboratories in the mixed sample experiments may also be due to differences in the delivered subsamples (observed by DDR). The mixed samples might not have been fully

homogeneous. However, this has only little influence on the comparability between laboratories, as the subsamples were distributed randomly (except that DK got the last produced), and an increased variation within the laboratories will make the differences between laboratories less significant.

The low mean values from analyses of extracts found by Askö and partly Umeå suggests that the general low values found by these laboratories might be due to the spectrophotometer readings. However, Umeå has afterwards controlled their spectrophotometer, and found no errors. Instead they suggest, that high temperature (about 30 degrees) and bright sunlight, and/or the use of Millipore filters instead of GF/C filters, might be the reason for the low values found by Umeå and Askø, who worked in the same laboratory. With prepared extracts the USSR got high mean values and low standard deviations, but with mixed samples they got low mean values and high standard deviations. This suggests that the filtration and extraction procedures strongly influenced the results. The reason for this is not known, but exposure to acid or acid vapours can have the observed effects.

5.4.2 Storage of samples

The storage of samples for one month in a freezer generally increased the chlorophyll-a concentrations, but the increase was not statistically significant and not found by all laboratories or by the same laboratory in different experiments. Storage in the form of extracts instead of filters gave significantly higher mean values.

5.4.3 Ethanol -- Acetone

Pooling all data on ethanol and acetone in two groups within each experiment showed no significant differences between the efficiency of the two extraction solvents. Askö, which used both solvents, found in one experiment that ethanol gave a significantly higher mean value, but in another experiment acetone gave an insignificantly higher mean value.

DDR used only acetone in the analyses of the mixed samples. Comparing the mean values of DDR with the grand mean of the laboratories finding the highest mean values, and which deliver data to the HELCOM database (SF,DK,D,S), showed, that the acetone extraction by DDR always gave lower mean values (3 - 24 %). DDR has by comparing with data from the HELCOM database estimated, that the DDR acetone data on chlorophyll-a concentrations are about 10 % lower than ethanol data obtained by the other laboratories (Schulz, pers. comm.). The data from this inter-calibration can neither verify nor reject this. Analyzing the SF-extract the DDR got exactly the same mean value as the grand mean of the laboratories D, DK, S and SF, indicating that the use of acetone is the reason for the lower mean values generally obtained by DDR in the mixed sample experiments.

5.5 Conclusion

The chlorophyll-a intercalibration generally showed that the precision within the participating laboratories was high. However, the comparability, between the laboratories was rather low, as the low variances within the laboratories made even small differences between the laboratories statistically significant. The filtration and extraction procedures had only little influence on the variances within the laboratories.
Generally the storage of samples for one month in a freezer did not influence the results significantly. However, storage as extracts instead of filters gave significantly higher results.
Using acetone as extraction solvent generally resulted in lower mean values than ethanol, but it is not possible from this exercise to give an exact percentage difference.

The main reasons for the differences found between the laboratories seems to be the spectrophotometer measurements, and the laboratory conditions during filtration, extraction and measurement. To reduce the variance and increase the comparability between the laboratories it seems necessary to be more careful about measuring at the peak of the chlorophyll-a absorption with a bandwidth of not more than 2 nm, and to follow

the Guidelines for the BMP strictly, that is: to work in subdue light at about 20 degrees in a laboratory without acid vapours.

Most of the laboratories (D, DDR, DK, S and SF) participating in the Baltic Monitoring Programme and delivering chlorophyll-a data to the HELCOM database were also the laboratories -finding the highest and most comparable chlorophyll-a mean concentrations in the intercalibration exercise.

5.6 **Recommendations**

The Guidelines for the BMP shall be strictly followed.

The chlorophyll-a absorption peak should carefully be determined before measuring any series of chlorophyll samples, and the bandwidth shall be 2 nm.

Storage of chlorophyll-a samples in freezers should preferably be as extracts instead of filters.

Only 96 % pure ethanol should be used as extraction solvent.

After vigorous shaking of the extraction samples before centrifugation, the GF/C-filters should carefully be twisted against the inner wall of the glass above the solvent, in order to get as much ethanol and chlorophyll-a out of the filters as possible, before the filters are removed from the samples.

	Chlorophyll concentration																	
	Øresund, during worksho			Øresund, 1 month later			Field station, during workshop			Field station, 1 month later			extract from Denmark			extract from Aranda		
	MEAN	STD	CV	MEAN	STD	CV	MEAN	STD	CV	MEAN	STD	CV	MEAN	STD	CV	MEAN	STD	CV
Ship and method																		
D, alkor	3.89	0.08	2.04	4.06	0.12	3.01	2.45	0.06	2.34	2.60	0.05	2.07	3.19	0.05	1.59	1.80	0.04	2.38
DDR, Penck	3.64	0.05	1.46	3.44	0.23	6.66	2.00	0.08	4.24	1.95	0.10	4.89	3.89	0.03	0.68	1.86	0.01	0.30
DK, G. Thorsen	4.11	0.09	2.23	4.20	0.07	1.62	2.46	0.04	1.42			.	3.35	0.01	0.28	1.84	0.02	0.99
SF, Aranda, ext	3.55	0.07	1.87	4.13	0.05	1.25	2.59	0.04	1.72	2.52	0.05	2.16	.	.	.	1.91	0.01	0.28
SF, Aranda, dry f		.	.	3.94	0.08	1.93	.	.	.	2.39	0.10	4.16		.	.	.	.	.
S, Argos	3.50	0.16	4.50	3.90	0.09	2.27	2.33	0.09	3.96	2.61	0.03	1.30	3.38	0.10	2.87	1.88	0.03	1.86
S, Askø, ethanol	3.04	0.05	1.76	2.31	0.25	10.9	.		.	1.75	0.13	7.58	2.29	0.03	1.15	1.40	0.02	1.52
S, Askø, acetone	2.73	0.12	4.33		.		.	.		1.89	0.12	6.48	.	.			.	.
S, Umeå	2.38	0.25	10.3	2.5;	0.11	4.39	1.87	0.13	6.93	.	.	.	3.51	0.09	2.68	1.65	0.04	2.42
USSR, Lev Titov	2.13	0.55	25.7	2.02	0.12	5.80	1.54	0.22	14.5	.	.		3.51	0.10	2.77	1.84	0.03	1.64

Tabel 1. **Mean in** ug/l, **standard deviation and coefficient of variation for each laboratory and experiment type. Dry f = Stored as dried filters, Ext = Ethanol extraction evt. stored as extracts. Where** nothing **else is mentioned ethanol was used for extraction, except that Penck used acetone. The prepared extracts were made with ethanol.**

Laboratory and sample type	Sample from Øresund		Sample from field station		Prepared extracts	
	During Workshop	One month later	During Workshop	One month later	from DK	from SF
D	B 3.89	BC 4.06	B 2.45	AB 2.60	D 3.19	D 1.80
DDR Ace.	C 3.64	E 3.44	D 2.00	D 1.95	A 3.89	BC 1.86
DK	A 4.11	A 4.20	B 2.46		C 3.35	BC 1.84
SF Ext.	C 3.55	AB 4.13	A 2.59	B 2.52		A 1.92
SF Fil.		CD 3.94		C 2.39		
S	C 3.50	D 3.90	C 2.33	A 2.61	C 3.38	B 1.88
Askö Eth.	D 3.04	G 2.31		E 1.75	E 2.29	F 1.40
Askö Ace.	E 2.73			D 1.89		
Umeå	F 2.38	F 2.57	E 1.87		B 3.51	E 1.65
USSR	G 2.13	H 2.02	F 1.54		B 3.51	C 1.84
Grand mean	3.23	3.41	2.18	2.24	3.30	1.83

Table 2. Mean in ug/l for each laboratory and experiment type. The letter A denotes the highest mean value in each experiment, the letter B the second highest significantly different mean value and so on (see text). Sample type Ace. = Acetone, Eth. = Ethanol extraction evt. stored as extracts, Fil. = Stored as filters. Where nothing else is mentioned ethanol has been used for extraction. The prepared extracts were made with ethanol.

6. REPORT OF THE WORKING GROUP ON OXYGEN AND HYDROGEN SULPHIDE

S. Carlberg[1], K. Fyrberg, J. Valderrama and B. Yhlen
Swedish Meteorological and Hydrological Institute Oceanographical laboratory, Göteborg

6.1 Introduction

Following a discussion in the Scientific and Technological Committee of HELCOM, the Commission requested ICES in the autumn of 1988 to work out a proposal for an intercalibration of determinations of dissolved oxygen in sea water. A first proposal was drafted by ACMP in June 1989 and finalized by the Marine Chemistry Working Group in February 1990. The proposal was then discussed by the Steering Group for the BIW III and subsequently approved by ACMP in June 1990.

6.2 Organizational pre-arrangements

The proposal from ICES was slightly amended by the Steering Group and then sent to possible participants in the intercomparison as they could be identified by the Steering Group and the Conveners. Thus the proposal was distributed to institutes in all Baltic countries and comments by correspondence before 1 May 1990 were invited. The same information was also distributed by the Commission secretariat through its regular communication with national contact persons.

6.3 Participation

Six laboratories participated in the intercomparison of the determination of dissolved hydrogen sulphide and oxygen. They are identified below by number, vessel and institute. In the tables and graphs describing the results, participants are identified only by number.

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[1] The four authors were responsible for the first draft report. This was evaluated by the ICES Marine Chemistry Working Group. The final report was then prepared by Carlberg in his capacity as Chairman of the Chemical Oceanography subgroup of the Marine Chemistry Working Group.

	Vessel	Institute	Country
1	Argos	**Swedish Meteorological and Hydrological Institute**	**S**
2	Aranda	**Institute of Marine Research**	**SF**
3	Alkor	**Institute of Marine Research**	**FRG**
4	KBV04	**Umeå** Marine Research Center	**S**
5	G. **Thorson**	National Environment Research Institute	**DK**
6	Lev Titov	Hydrometeorological Observatory of Klaipeda	**USSR**
7	Argos	Swedish Meteorological and Hydrological Institute	**S**

NB! Participant No 7 is applicable to tests 1 and 2 only, as described below!
Regrettably, the relevant monitoring institutes in Poland, Hamburg and the (then) DDR did not find possibilities to participate.

6.4 **Programme plans**

6.4.1 The original programme

The original programme plan was outlined by ACMP, modified by the Marine Chemistry Working Group and finally endorsed by ACMP. Some little modifications were done at the spot as described below.
The entire work was to be carried out on one ship in order to minimize the influence of patchiness. The exercise should include two water masses with different oxygen saturations. The design of the sampling procedure implied that repeated sampling could be done from a homogeneous water body.
All analysts were supposed to use as part of test 3, the stock iodate and thiosulphate solutions of laboratory 1 (the conveners) and to compare it against their own preparations of these **reagents**.

1) Test of the variability caused by sampling equipment:
All different hydrocast bottles used by the participants should be operated by one person taking three water samples from each sampler. When all samples are taken, the first sampler is used to obtain three more samples in order to assess any possible variability in the water body during the sampling period. One person has to withdraw samples from all the hydrocast bottles and perform all subsequent steps of fixation, titration etc. Only mixed layer water is sampled for this exercise.

2) Test of the variability caused by sampling staff:
All steps according to test 1 with the exception that every participant operates his/her own hydrocast bottle to obtain the samples from the mixed layer water. From the water mass with lower concentrations of oxygen all the samples are taken simultaneously using the rosette sampler to eliminate the effects of the oxygen gradient.
Test 1 and 2 can be done simultaneously in the mixed layer water.

3) Test of the variability caused during the analysis:
Two bulk samples of water with different concentrations of dissolved oxygen is used. From each bulk sample every participant withdraws six subsamples which he/she subsequently takes through the entire procedure of fixation, titration etc. Three of these samples are analyzed using the participants' own reagents. For the remaining three samples, the reagents used will be provided by the conveners.

6.4.2 Modification of the programme

As a result of discussions in the Steering Group and with the participants at the planning meeting in the beginning of the meeting in Visby some modifications were made. The first change was that the Steering Group wanted to offer also an inter-comparison of dissolved hydrogen sulphide in water. As anoxic water would not be available in the vicinity of Visby, this part of the intercomparison would have to be based on pre-arranged natural samples stabilized with a preservative. This suggestion

together with the original programme from ICES was distributed by the HELCOM secretariat but also mailed by the conveners to identified institutes and colleagues and comments were invited. Other modifications or clarifications of the programme are described in the following section.

6.4.3 Execution of the programme

Dissolved hydrogen sulphide

A bulk sample of sea water containing hydrogen sulphide was sampled by the conveners on board R/V Argos before arriving at Visby. The water was distributed to ordinary oxygen sampling bottles (approximate volume 50 - 65 ml), three for each participating laboratory, and was preserved with cadmium chloride solution.

Dissolved oxygen

The entire work was carried out on one ship (R/V Gunnar Thorson) in order to minimize the influence of patchiness. The exercise included two water masses with different oxygen saturations. These water masses had been identified by R/V Argos on its way to Visby. At 10 m the temperature was 17,19°C and salinity 6,662 PSU and at 80 m 3,99°C and 8,264 PSU. Using the mean values of oxygen as determined by the participants this corresponds to about 97 per cent saturation in the surface water and 40 per cent in the deep water. Therefore, it could be suspected that significant exchange of oxygen might take place between the deep water samples and the atmosphere, but that the mechanism would not be a potential disturbance to the surface water samples. The sampling was carried out according to the original plan with the following modifications or clarifications as described below.

1) Test of the variability caused by sampling equipment:
All different hydrocast bottles used by the participants were handled at the wire by one person from Lab 1. Samples were obtained from 10 m depth. A second person from the same lab withdrew the samples from all the hydrocast bottles and performed all subsequent steps of fixation, titration etc. taking three

water samples from each sampler. When all samples were taken, the first sampler was used again to obtain three more samples in order to study variability in the water body during the sampling period.
All participants used hydrocast bottles either of Nansen type (TPN sampler from Hydro Bios in Kiel, FRG) or Niskin type (from General Oceanics, USA).

2) Test of the variability caused by sampling staff:
Samples were taken from 10 m and 80 m to obtain samples with high and low concentrations of oxygen. It was recognized that any variability would be caused in the withdrawal of the replicates and not by e.g. how the sampler was handled when attached to the wire etc. Therefore, for the mixed water layer, this test was combined with test 1 in such a way that when the person number 2 from Lab 1 had withdrawn three replicates for test 1, the participant (the "owner" of the hydrocast bottle) withdrew three replicates for test 2. The samples of water with low concentrations of dissolved oxygen were obtained using the rosette sampler. In that case all hydrocast bottles were fired sequentially in shortest possible time at 80 m depth. Subsampling started as soon as the rosette sampler was retrieved. In order to create reasonable working conditions only one participant at a time was let into the rosette lab. Each participant withdrew three replicates, from the particular Niskin hydrocast bottle assigned to him/her. Also in this test Lab. 1 sampled twice; as first and last lab in order to determine the effects of a possible oxygen gradient during the sampling or handling of the samplers. Again, all samples were precipitated and analyzed by the same person from Lab. 1 as in test 1. The total duration of sampling for test 1 and first part of test 2 (10 m) were 65 minutes.

3) Test of the variability caused during the analysis:
The two bulk samples of water with different concentrations of dissolved oxygen were obtained using a 30 L Niskin hydrocast bottle again at 10 m and 80 m. Subsampling started as soon as the sampler was retrieved, with the laboratories in the same order as before. From each bulk sample every participant withdrew six replicates. Three of these samples were precipitated using

the participants' *own* Winkler reagents. For the remaining three samples, the Winkler reagents were provided by the conveners. The participant subsequently took all replicates through the entire procedure of precipitation, titration etc. The replicates for Lab. 1 were withdrawn as three at the start and three at the end of the experiments in order to check any time dependence of the subsamplings. For this test aliquotes of iodate and thiosulphate solutions were distributed by the conveners. The idea was that every participant should use these two solutions to determine the normality of the thiosulphate used by the conveners and then use the iodide solution to determine the normality of his or her own thiosulphate solution. The normality of the conveners thiosulphate should be reported and should provide an estimation of the typical calibration error between laboratories.

6.5 Statistical analysis

The aim of statistical analysis is to extract useful information from the collected results, and to describe the performance of laboratories, both individually and as a community, summarized as follows.

a) Estimation of the consensus ("true") concentration of oxygen at each sampling occasion
b) Evaluation of the performance of each individual laboratory in terms of bias and precision
c) Evaluation of overall performance of laboratories in terms of reproducibility and identification of consistent laboratories.

The first approach was to apply a t-test to find outliers in the data material. However, it was realized that this was incorrect and would lead to inconclusive results since only three replicates had been used instead of at least five as required for the test.

The next step was to evaluate whether or not the extended sampling times had lead to heterogeneities between laboratories and samples in the different tests. This was done by applying linear regression (on tests 1 and 2). The results are presented below.

The calculation of the consensus concentrations is very important since the samples can be regarded as uncompromised reference materials. Therefore, the full data set was used for each test whatever is the representativity of each single value in relation to the whole population of laboratories. The calculated consensus values are used here as **representing the "true"** oxygen concentrations in the sampled water masses.

The general way of presenting the results for each experiment here is based on scatter plots (see figures 1 - 11) and simple descriptive statistics i.e. mean value (i.e. consensus value for **"true"** oxygen concentration), standard error, standard deviation, coefficient of variation and prediction values as calculated on the full dataset.

Also the data for the comparison of the thiosulphate solutions for test three are treated and presented simply as scatterplots and descriptive statistics.

6.6 Results and discussion

6.6.1 Dissolved hydrogen sulphide

This test failed completely for one single reason; all laboratories were unable to dissolve the precipitate of cadmium sulphide, probably because the samples had to be stored several days between sampling and analysis. Some of the participants had expressed doubts about cadmium as preservative and recommended zinc instead. The reason why the conveners choose cadmium was simply that it seemed to be advocated in the literature (Grasshoff: Methods of Seawater Analysis, 2nd edition, p 77).

6.6.2 Dissolved oxygen

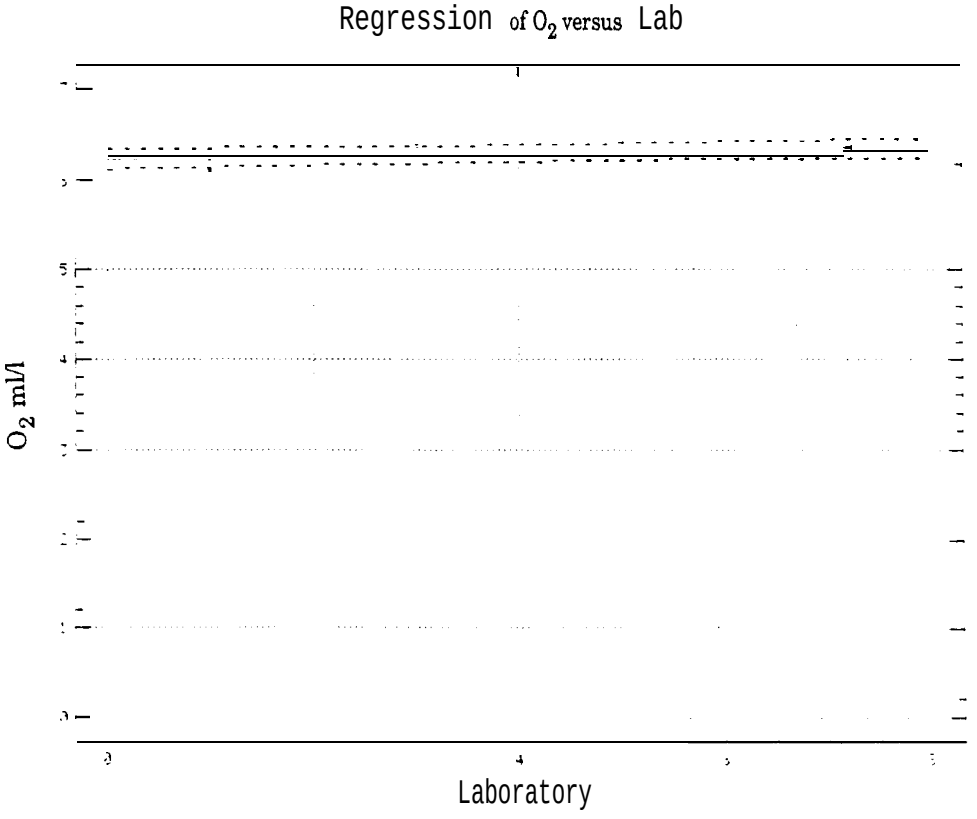
The results are presented below in Tables 1 - 2, and Figures 1 - 11 as Test 1, Test 2H and 2L (high and low oxygen content) and Test 3HC, 3HO, 3LC and 3LO, where C stands for Winkler reagents provided by the convener and 0 stands for the participants' own reagents.

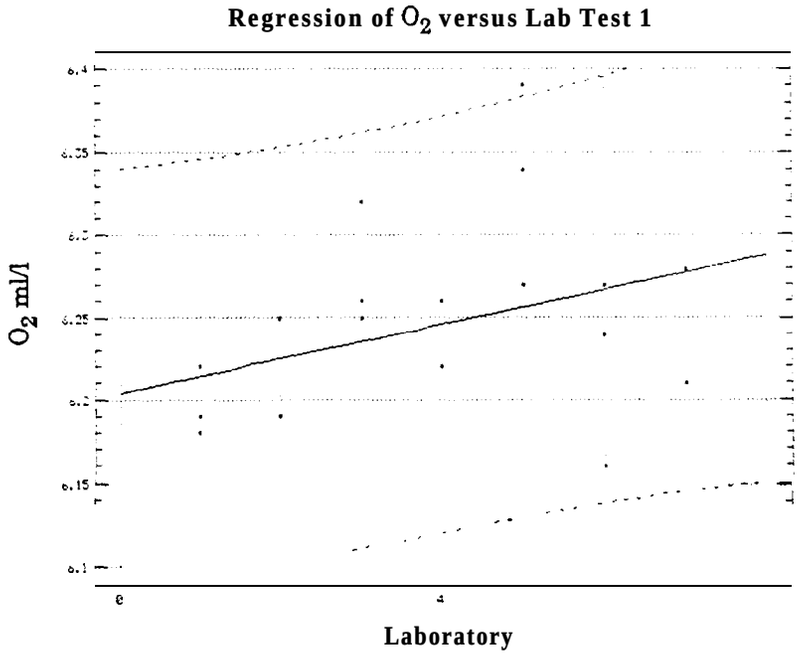
Test 1, The variability caused by sampling equipment

Linear regression calculation of the dataset showed that there was no significant difference between the first and the last sampling and the samples can therefore be regarded as homogeneous between laboratories (see Figure 1). The basic statistics calculated is presented below in the table. In this case the results from Lab. 7 were eliminated since actually they represent Lab. 1, which must not be over-represented in the calculation.

In Figure 2 the results are presented as a close-up scatterplot with the regression line as a slightly sloping solid line in the middle. Rather than introducing lines for standard deviations a different approach is used. The dotted lines represent the calculated prediction values at the 95 per cent confidence level. In simple terms this can be evaluated in the following way. If laboratory X would have produced yet another sampling at the same occasion the result would have occurred within these two dotted lines with a probability of 95 per cent. With this interpretation there is no difference between the various hydrocast bottles. It could be noted that Lab. 5 produced one of its three results as slightly higher than the predicted value. However, this does not change the conclusion above.

Fig.1





Data File: BIW III Oxygen test 1

Variable: Test 1 Oxygen, Observations: 1 7

Minimum: 6,140	Maximum: 6,390
Range: 0,250	Median: 6,250
Mean: 6,244	Standard Error: 0,0 16
Variance:	0,004
Standard Deviation:	0,065
Coefficient of Variation:	1,038
Skewness: 0,46 1	Kurtosis: -0,420

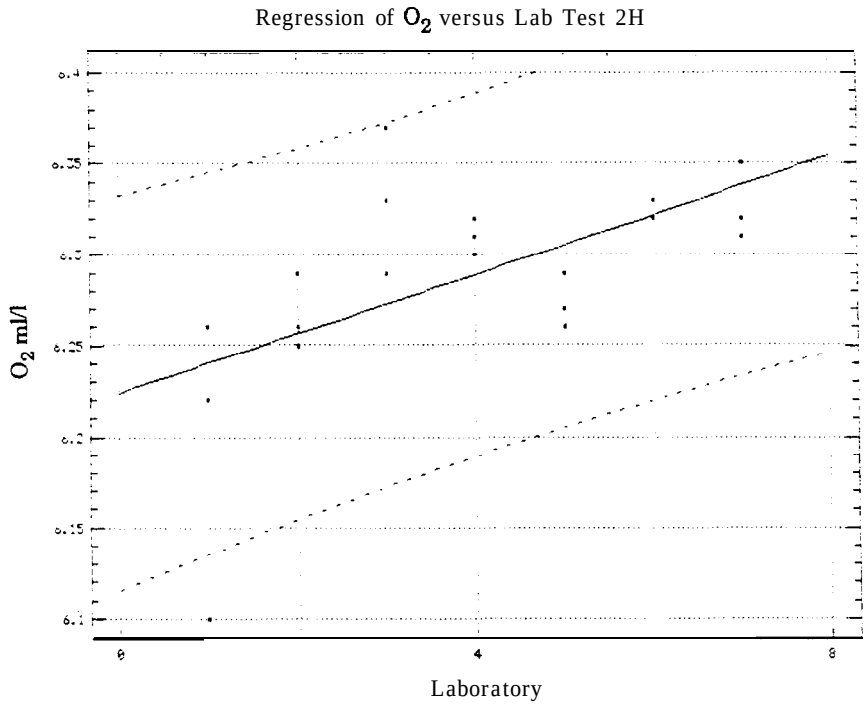
Fig.2

Test 2H, The variability caused by sampling staff.

Also in this experiment the linear regression calculation of the dataset showed that there was no significant difference between the first and the last sampling and the samples can therefore be regarded as homogeneous between laboratories (figure 3). The basic statistics calculated is presented below in the table. Also in this case the results from Lab. 7 were eliminated in the calculation of the basic statistics.

Although Lab. 1 produced one of its results below the lowest predicted value (and Lab. 5 one result almost on the borderline for the predicted highest value) the conclusion is that there is no significant difference between sampling staff of the participating institutes.

Fig.3



Data **File: BIW III Oxygen test 2**

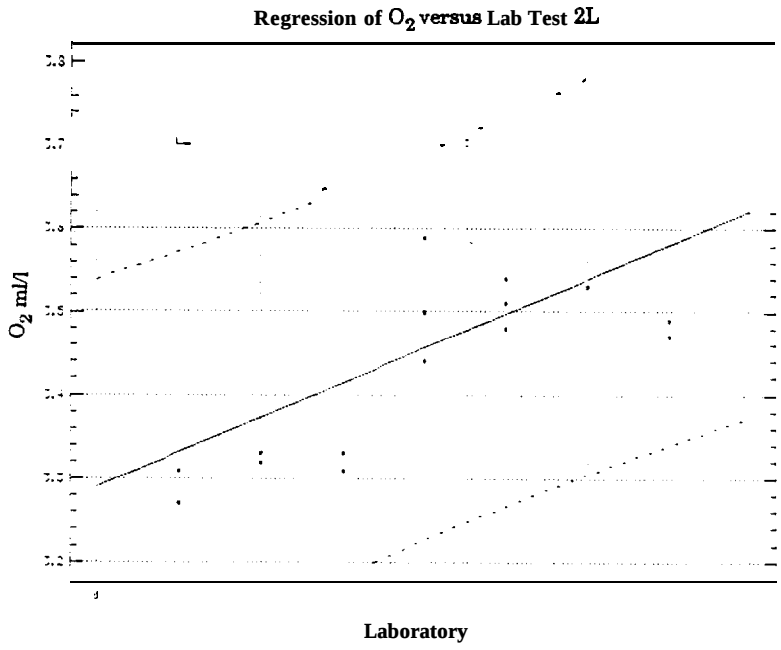
Variable: Test 2H, Oxyen Observations: 18

Minimum: 6,1 00	**Maximum: 6.370**
Range: 0,270	**Median: 6,290**
Mean: 6,263	**Standard Error: 0.014**
Variance:	**0,003**
Standard Deviation:	**0,056**
Coefficient of Variation:	**0,926**
Skewness: -1,5 14	**Kurtosis: 2.853**

Test 2L, The variability caused by sampling staff.

In this experiment the linear regression calculation on the dataset indicated that there was a slight difference between the first and the last sampling as can be seen in figure 4. Also, the results of the Labs 1, 2 and 3 (with the exception of one value) seem to belong to one group and the results of Laboratories 4 – 7 to a different group. The samples can therefore not be regarded as entirely homogeneous between laboratories. The basic statistics calculated is presented below in the table. Also in this case the results from Lab. 7 were eliminated in the calculations. From a statistical viewpoint it can be argued that the test is inconclusive because of the possible stratification of the results into two populations as identified above. From the viewpoint of calculated prediction values the conclusion would be that also at this low oxygen concentration there is no significant difference between participating sampling staff.

Fig.4



Data File: BIW III Oxygen test 2

Variable: Test 2L	Observations: 18
Minimum: 3,270	Maximum: 3,740
Range: 0,470	Median: 3.460
Mean: 3,453	Standard Error: 0,034
Variance:	0.021
Standard Deviation:	0,146
Coefficient of Variation:	4,218
Skewness: 0,388	Kurtosis: -1 ,273

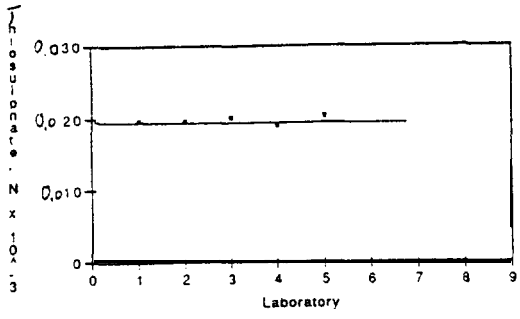
Test 3, The variability caused during the analysis. [2)]

The test was designed to reveal systematic and random **errors** between the analytical results from the different laboratories. Possible systematic errors could be caused by 1) the calibration of the thiosulphate solution, 2) the Winkler reagents and 3) the titration procedure. As described above in section @'Dissolved **oxygen"** arrangements were made to test 1) in a complementary exercise, the factor 2) by having all participating laboratories **analyze** duplicate sets of samples using Winkler reagents of their own as well as reagents distributed by the Conveners. Finally, 3) could be inferred by combining the results of 1) and 2).

Test of the comparability of the thiosulphate solutions.

The thiosulphate and iodate solutions of the convening laboratory were distributed to all participating laboratories. Their results in standardizing this thiosulphate solution is presented below in figure 5. Although the results seem to be close, the highest value is not less than 7 per cent higher than the lowest on this vital calibration procedure. As the standardization was made on one and the same iodate solution, the differences reflect titration errors.

Fig.5



Data File: BIXIII Thiosulphate

Variable: Thiosulohate. N Observations: 11

Minimum: 0.019000	Maximum: 0.020400
Range: 0.001400	Median: 0.020161
Mean: 0.019835	Standard Error: 0.000175
Variance:	0.000000
Standard Deviation:	0.000582
Coefficient of Variation:	2.934402
Skewness: -0.508170	Kurtosis: -1.576085

[2)] In order to reduce the possible influence by atmospheric oxygen *on* the aliquot samples, the glass bottles *were* stored under water in a container between sampling and analysis. However, unfortunately all but one of the bottles from Lab.3 lost their stoppers for some time in the water bath. This was discovered and the stoppers immediately replaced. As the precipitation in these bottles seemed to be undisturbed it was decided that all samples should by analyzed and the results evaluated. The corresponding values are put in brackets in Table 2.

Test of the comparability between Winkler reagents

The over-all results of high and low concentrations are presented as scatter plots below in figures 6 and 7. For each laboratory the first set of dots **represent the** analyses using the conveners Winkler reagents and the second set (slightly to the right of the first one) the use of the reagents from the participating laboratory. A first impression is that in both tests the oxygen content of the sampled water has been increasing with time. However, in both tests the convening Lab. 1 sampled as first and last participant and therefore the results show that the oxygen content can be regarded as constant during the duration of the experiment! There were no significant differences between the mean values for the whole population of results of each kind of reagent either at high or low oxygen concentration. See tables corresponding to figures 8 - 9 and 10 - 11 respectively.

The mean value for each data set (3HC, 3HO, 3LC and 3LO) of each laboratory were calculated and then the differences between the means (3HC - 3H0 and 3LC - 3LO) were studied for each lab. The calculations suffered from the fact that only three duplicates had been analyzed and therefore the results should be regarded as indicative rather than conclusive. For laboratories 1 - 4 no differences seem to be present within each laboratory for high or low oxygen concentrations. However, for the laboratories 5 and 6 there may be a difference in that their own reagents gave slightly lower results (0.04 - 0.10 ml/l). Calculations were made at 95 % confidence level.

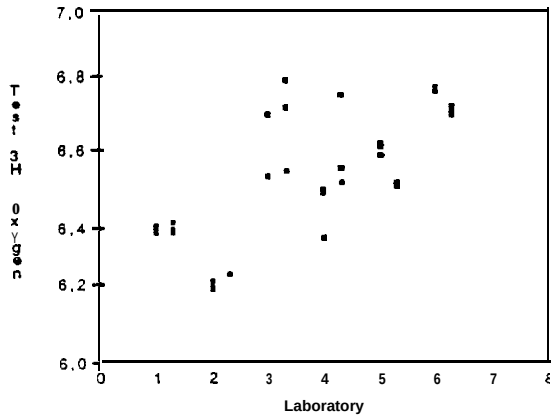


Fig. 6

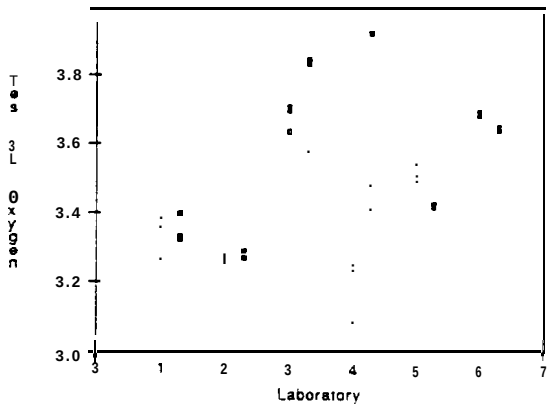


Fig.7

The comparability of titration procedures

The results for the surface water are presented as scatter plots and corresponding statistical tables below. Figure 8 and corresponding table represent the HC test and figure 9 and corresponding table the HO test. Concerning Lab. 3 the results may not be representative (see also note on page 9) and its results are therefore evaluated in general terms only. Furthermore, it should be noted that Lab. 2 was the only participant using automated titration; in this case with an electrochemical end-point detection.

Obviously there are systematic differences between laboratories although the differences are not drastic. All single results of HC fall within 95,6 - 103.9% of the consensus value and for HO between 96.0 - 103,5%, which is quite acceptable. For the low concentration the results were slightly worse, which is perhaps not surprising as the saturation was as low as only 40%. Results of 89.6 - 106.9% for LC are not really acceptable, but 93.2 - 103.9% for LO, which of course represents the normal procedure of the participants, is almost as good as the results for HC and HO.

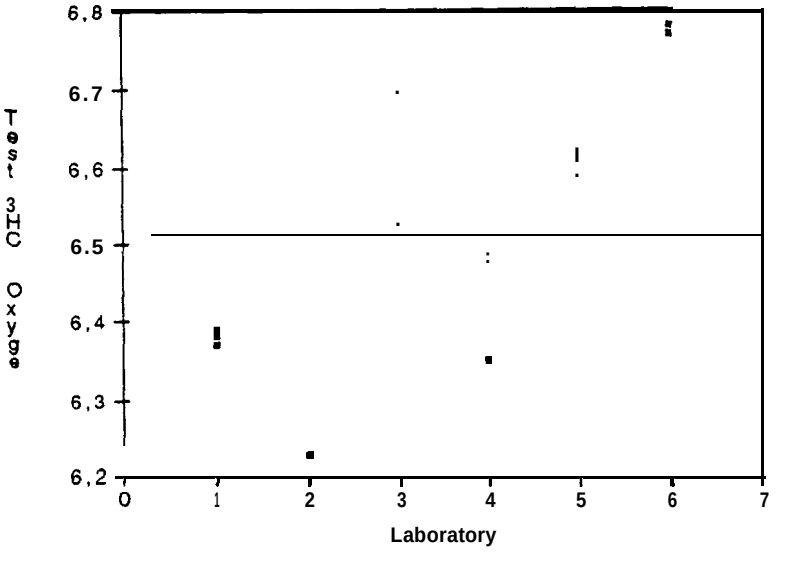
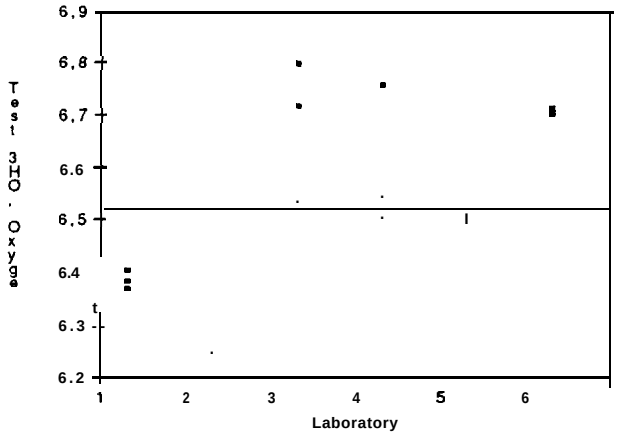


Fig.8

Data File: BIW III Oxygen test 3
Variable: Test 3HC, Oxyge Observations: 1 8

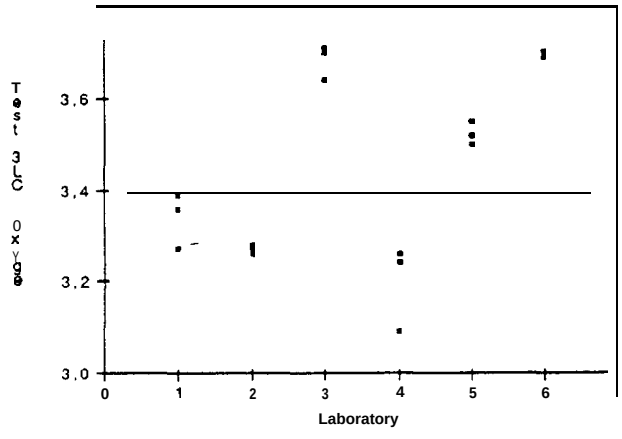
Minimum: 6,210	Maximum: 6,780
Range: 0,570	Median: 6,510
Mean: 6,511	Standard Error: 0,046
Variance:	0,038
Standard Deviation:	0,195
Coefficient of Variation:	2,991
Skewness: -0,096	Kurtosis: -1 ,417



Data File: BIW III Oxygen test 3
Variable: Test 3HO, Oxyge Observations: 17

Minimum: 6,250	Maximum: 6,800
Range: 0,550	Median: 6,510
Mean: 6.540	Standard Error: 0,042
Variance:	0,030
Standard Deviation:	0,174
Coefficient of Variation:	2,665
Skewness: -0.122	Kurtosis: -1,297

Fig.9

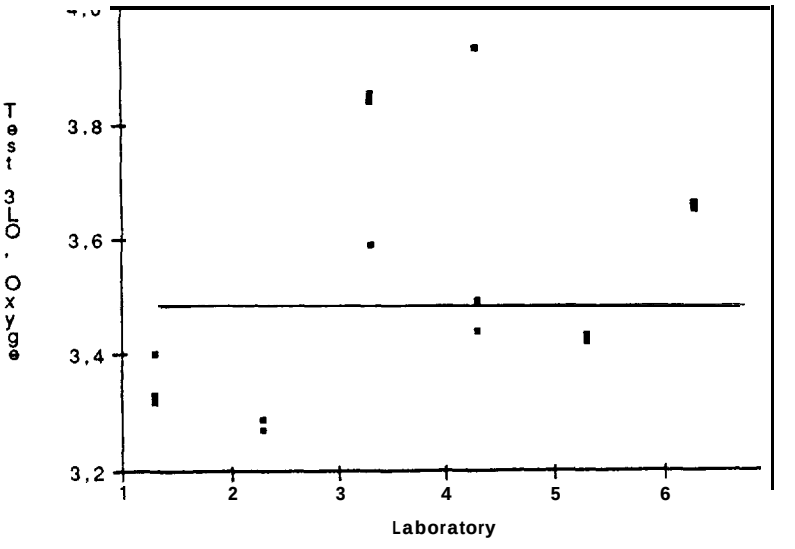


Data File: BIW III Oxygen test 3
Variable: Test 3LC, Oxyge Observations: 18

Minimum: 3.090	Maximum: 3.710
Range: 0.620	Median: 3,445
Mean: 3,452	Standard Error: 0,048
Variance:	0.042
Standard Deviation:	0,205
Coefficient of Variation:	5,952
Skewness: -0,027	Kurtosis: - 1,583

Fig.10

Fig. 11



Data File: BIW III Oxygen test 3
Variable: Test 3LO, Oxyge Observations: 18

Minimum:	3,270	Maximum:	3,930
Range:	0,660	Median:	3,435
Mean:	3,516	Standard Error:	0,049
Variance:		0,043	
Standard Deviation:		0,207	
Coefficient of Variation:		5,893	
Skewness:	0,595	Kurtosis:	-1,007

Concerning precision the picture is quite clear in that Lab. 4 has a problem to solve, whereas the results of Lab. 3 cannot be assessed for reasons described earlier.

Concerning accuracy it can be noted that in almost all cases of the four tests the laboratories produced consistent results in the sense that they are either higher or lower than the consensus value and the distance to this value does not differ much between the tests. If the systematic error could be explained as a pure calibration error one would expect that the Laboratories producing higher than average values in the standardization of the thiosulphate solution would also produce the higher than consensus value for the oxygen analyses. However, in comparing figure 5 with figures 8 - 11 it becomes obvious that also other systematic differences are involved. The present intercomparison does not yield material to clearly identify these factors. The most likely one is the end point detection in the titration.

6.7 Conclusions and recommendations

Conclusions

1. Too few replicates (3) were sampled and analyzed to provide a basis for good statistical evaluation of the results. The conclusions should therefore be regarded as indicative rather than fully conclusive.

2. The use of different hydrocast bottles does not yield results which differ significantly at the 95 per cent confidence level.

3. The use of different sampling persons does not yield results which differ significantly at the 95 per cent confidence level for the samples which were almost saturated with oxygen. Concerning the samples with low oxygen content there is a possibility that the samples were not homogeneous between laboratories and that the results, therefore, may be inconclusive.

4. There are systematic differences, most likely due to titration errors, between laboratories in standardizing their own thiosulphate solutions.

5. When using their own Winkler reagents and iodate solutions for calibration all laboratories produced acceptable results (96 - 104 % of the consensus value at the high oxygen concentration and 93 - 104 % at the low concentration).

Recommendations

1. It is recommended that the exercise (tests 2 and 3) is repeated at the next intercalibration arranged by HELCOM and that all laboratories reporting oxygen data to the Baltic Monitoring Programme participates.

2. It is further recommended that at the next occasion at least 5 replicates are analyzed in each test in order to allow for a proper statistical evaluation of the results.

Acknowledgements

The authors wish to thank Drs Shier Berman and Alain Aminot for their contributions to the statistical analysis of the data.

Table 1

OXYGEN EXPERIMENT all concentrations as ml/l	No 1	No 2A	No 2B
ARGOS	6.19 6.22 6.18 } Mean 6.20	6.26 (6.10) 6.22 } Mean (6.19) 6.24	3.27 3.31 3.31 } Mean 3.30
ARANDA	6.25 6.14 6.19	6.25 6.29 6.26	3.33 3.33 3.32
ALKOR	6.32 6.25 6.26	6.33 6.29 6.37	3.31 3.33 3.74
KBV 04	- 6.22 6.26	6.32 6.30 6.31	3.44 3.59 3.50
G. THORSON	6.34 6.27 6.39	6.26 6.27 6.29	3.48 3.54 3.51
LEV TITOV	6.24 6.27 6.16	6.33 6.32 6.32	3.65 3.66 3.53
ARGOS	6.28 6.28 6.21 } Mean 6.26	6.32 6.31 6.35 } Mean 6.33	3.47 3.49 3.47 } Mean 3.48

Oxygen Experiment No 3

All concentrations as ml/l

Table 2

Ship:	ARGOS	ARANDA		ALKOR		KBV 04		G. THORSON		LEV TITOV	
Reagents:	own reag.	Ar	Own	Ar	Own	Ar	Own	Ar	Own	Ar	Own
10 m	6.37	6.23	6.25	(6.53)	6.54	6.35	6.51	6.59	6.50	6.77	6.71
sample	6.39	6.22	-	(6.70)	(6.72)	6.49	6.55	6.62	6.50	6.78	6.72
	6.30	6.21	6.25	(6.70)	(6.80)	6.48	6.76	6.61	6.51	6.78	6.71
80 m	3.39	3.26	3.29	6.70)	(3.85)	3.09	3.93	3.55	3.42	3.70	3.65
sample	3.36	3.28	3.29	(3.71)	(3.59)	3.24	3.44	3.52	3.43	3.70	3.66
	3.27	3.27	3.27	(3.64)	(3.84)	3.26	3.49	3.50	3.43	3.69	3.65

Homogenity test by ARGOS

	first samples			last samples			difference of mean
10 m	6.37	6.39	6.38	6.40	6.30	6.37	± 0.00
80 m	3.39	3.36	3.27	3.40	3.32	3.33	- 0.01

7. REPORT OF THE WORKING GROUP ON MACROZOOBENTHOS

Hans Cederwall (convener)
Stockholm Centre for Marine Research, Stockholm University

7.1 Introduction

The intercalibration of soft bottom macrozoobenthos was carried out in Visby, with field sampling north of Visby, near the coast of Gotland. The meeting was held from 27 to 31 of August 1990. Laboratories from all contracting parties took part in the intercalibration, one from each state except Sweden who participated with two. Because of lack of ship and equipment Poland only took part in exercise two (prepared samples). A list of participants is given in Appendix 1.

In general the program set up ahead of the intercalibration meeting was followed. It contained four exercises:

1. To intercalibrate the positioning of the ships.
2. To intercalibrate the sorting of samples, species determination and weighing of species, by using "prepared samples".
3. To intercalibrate the sieving techniques (incl. sorting) at a station with clayey sediment. The station chosen was the Swedish national station no 4138 (Lat. 57°52,81, Long.18° 47,75), with a depth of 44 m.
4. To intercalibrate the sieving techniques (incl. sorting) at a station with sandy sediment. The station chosen was the Swedish national station 4002 (Lat. 57°51,39, Long. 18° 35,87), with a depth of 16 m.

During the intercalibration the group of participants also went round to the ships to inspect and videotape the different sampling devices and sieving equipment. Surprisingly several participants use other types of van Veen-grabs than the standard (Dybern et al. 1976) prescribed in the Guidelines (the Danish lab. is not using a van Veen at all in the HELCOM-monitoring),

and the weights of the grabs varied between 25 and 71 kg.

The group also met to discuss other things, e. g. the influence of patchiness, suggestions for future intercalibrations, data reporting and changestothe Guidelines. The recommendations from the group are listed in the end of the report.

7.2 Materials and methods

Positioning

When leaving Visby harbour in the morning of the 29th of August all ships went up to a navigational buoy, as close as possible, and read their position from their navigators (Decca and/or satellite).

Prepared samples

The prepared samples were taken at a station near the Askö Laboratory, in the Landsort area, on the 18th of May 1990. The samples were taken with a modified (Andersin & Sandler 1986) Olausson box-corer (Jonasson & Olausson 1966). They were sieved through a 1 mm net and preserved in 4 % formaldehyde solution, buffered with Hexamine and stained with Rose Bengal.

The samples were sorted in June. The animals were picked out, determined to species (in some cases to a highertaxonomic level) and counted. The animals were then kept in the preservation fluid until 3 months after sampling, when they were weighed. The weighing procedure was as follows: The animals were blotted on filter paper until they left no new wet stains on the paper and then transferred to preweighed Aluminium foil, formed to small "beakers". The "beakers" were then immediately closed and weighed on a 5 decimal balance.
Also the remaining sieving residue (after the animals had been picked out) was kept in 4 % buffered and stained formaldehyde during this time. After weighing, the animals were put back into

the sieving residue, and the samples were brought to Visby were they were distributed randomly to the participating laboratories. They brought the samples back home, where they treated them according to their own standard procedures.

For statistical analysis the abundance figures for the four most common species plus the total abundance and the number of taxa were used. For the analysis of biomass figures three different types of organisms were chosen: one softbodied (Harmothoe sarsi), one with exoskeleton (Pontoporeia affinis) and one with shells (Macoma balthica), plus the total biomass. Wilcoxon Signed-Rank test was performed on the data. With only 5 samples this test can however not give lower p-values than 0.059, so as a complement t-test was also performed.

Station 4130

The sampling at station 4138 was done with a standard van Veen grab (Dybern et al. 1976). The same grab was used for all 35 samples. The samples were distributed at random to the participating laboratories (5 to each lab.) who sieved them and preserved them on board, according to their own standard methods. They then brought the samples home to their institutes, where they were treated according to their standard procedures. Only abundance was determined for these samples.

For the statistical evaluation of the results, Kruskal-Wallis one-way analysis by ranks was used.

Station 4002

The sampling at station 4002 was taken with a shortarmed chainrigged van Veen-grab with buckets shaped according to Riddle (1989, Fig. 4), weighing 44 kg. The samples were treated in the same way as those from station 4138.
The statistical evaluation was the same as for station 4138.

7.3 Results

Positioning

The results from exercise 1 (positioning) are shown in Figs. 1 and 2. Except for the Finnish vessel Aranda (who gave figures about 0.6 nm from the others) the differences, when using the Decca-system, were small (max. 0.05 nm). The differences using satellite navigators were at maximum 0.4 nm, with the Soviet vessel Lev Titov excluded, 0.2 nm.

Prepared samples

The results from exercise 2 (prepared samples) are given in **Figs**. 3-4 and tables 1-2, primary data in appendix 2 to 11.

No significant differences were found between the number of **taxa** reported by the participating laboratories. There were however some small differences in species determination. FRG identified one priapulid specimen in sample 10 as Priapulus caudatus, while it **was** by Swedens lab 2 identified as Halicryptus spinulosus. The latter is most probably correct, since the former species has not been found anyway near the **Askö** area. Denmark identified Pygospio elesans in some samples, while no such species was found by Swedens lab 2.

The cases where significantly (t-test, $p<0.05$) lower abundance values, compared to Swedens lab 2, were reported are listed in table 1. Denmark reported significantly lower abundance figures for Harmothoe sarsi (Wilcoxon $p=0.10$), Pontoporeia affinis (Wilcoxon $p=0.059$), Pontonoreia femorata (Wilcoxon $p=0.059$) and total abundance (Wilcoxon $p=0.059$). Former GDR reported significantly less Harmothoe sarsi and total abundance than Sweden 2 and FRG found significantly less Pontoporeia femorata (Wilcoxon $p=0.10$). Poland reported lower total abundance (Wilcoxon $p=0.10$). No significant differences were found for Macoma balthica.

Table 1. Abundance figures reported, significantly lower (t-test, $p<0.05$) than those originally found by Swedish lab. 2 (Stockholm Univ.)

	Poland	GDR	FRG	Denmark
Total abundance	x	x		x
Harmothoe sarsi		x		x
Pontoporeia affinis				x
Pontoporeia femorata			x	x

Except for Macoma balthica Denmark reported markedly lower figures than other participants (See Fig. 1). For Harmothoe sarsi also former GDR and Finland reported fairly low figures. In all other cases the differences were not more than 10 %, compared to those found by Sweden 2.

The wet weight results differed much more than the abundance figures (Fig. 2), and most participating laboratories reported lower figures than those originally measured by Sweden 2. The results of the statistical analysis are given in table 2.

Table 2. Significance values (t-test and Wilcoxon signed rank test) for differences in wet weightcomparedto (=lower than) Swedens lab. 2 (Stockholm Univ.)

	SF	SU	PL	GDR	FRG	DK	SE1
Total biomass							
t-test	<.05	<.05	<.05		<.05	<.05	X.05
Wilcoxon	.059	.059	.059		.059	.059	.100
Harmothoe sarsi							
t-test	X.05			<.05		<.05	<.05
Wilcoxon	.100		.100			.100	.059
Pontoporeia aff							
t-test	<.05	<.05	<.05	<.05	<.05	<.05	
Wilcoxon	.059	.059	.059	.059	.059	.059	
Macoma balthica							
t-test	<.05	<.05	<.05		<.05	<.05	<.05
Wilcoxon	.059	.059	.059		.059	.059	.100

Finland and Denmark reported significantly lower values for all variables (3 species + total) tested, while former GDR reported lower values only for two, with the other participants placed inbetween. In most cases the differences reported were more than 10 %, often more than 25 % and as a maximum over 50 % (Denmark: Harmothoe).

Station 4138

Denmark reported they found 1 specimen of Leucon nasica on this station, but since that species, acc. to literature, has not been found in the Baltic proper they have most probably misdetermined a Diastylis rathkei. Finland reported they found 1 specimen of Aricidea suecica, which is doubtful since this species has not been found further into the Baltic proper than in the Gotland deep, and then much deeper.

The species Halicryptus spinulosus, normally occurring in each and other sample, was not found in the Danish samples. The mean number of taxa reported by the participating laboratories varied between 5.4 and 6.6. No significant differences were found.

Some abundance results are shown in Figs. 5-8 and basic statistics are for the whole material is listed in appendix 12.

Denmark found significantly (Kruskal-Wallis, $p<0.05$) lower abundance of Pontoporeia affinis and total abundance (Figs. 5 and 8), than the rest of the participants. FRG found significantly more Hydrobia, than the other participants.

The variability in the reported figures can be given as maximum deviation in % of overall mean values (calculated from the figures from all participating labs, the Danish excl.). For total abundance the maximum deviation was 17 %, while for Pontoporeia affinis it was 27 %, and for Pontoporeia femorata 24.5 %. For Macoma balthica (with Danish figures incl.) the maximum deviation was 15 %.

Station 4002

FRG reported they found 1 specimen of Ervthrops elesans on this station, but, according to literature, this species has not been found in the Baltic proper, so it is likely to be another mysid.

The number of taxa reported by the participating laboratories was somewhat higher than for the previous station. The mean number varied between 8.6 and 9.6 and no significant differences were found.

Some abundance results are given in Figs. 9-12 and basic statistics in appendix 13.

Several participating laboratories reported abundance values significantly (Kruskal-Wallis, $p<0.05$) deviating from the other participants (see table 3).

Denmark and Swedens lab 2 reported most of the deviating figures. The results from station 4002 are more variable (not so much the total abundance as the figures for separate species) than the results from station 4138. The variability, calculated as for station 4138, was for total abundance 26.7 %, for Pygospio elesans (excl. the figures from Swedens lab 2) 22.9 and for Hydrobia 54.7 %. For Cardium slaucum it was 45.5 while for Macoma balthica 22.4.

Table 3. Reported figures, significantly (Kruskal-Wallis, $p<0.05$) differing from the majority of reported figures. Higher values indicated by +, lower by -.

	SF	SU	GDR	FRG	DK	SE1	SE2
Pygospio elegans							-
Oligochaeta							
Bathyporeia pilosa					+	+	
Cardium glaucum				+	-		
Macoma balthica							+

The species Bathyporeiapilosa, earlier common at this locality, was now found only by three laboratories, and only in fairly low numbers.

In spite of the samples having been randomly distributed, the picture in the diagram for total abundance (Fig. 12) is very similar to that of station 4138 (Fig. 8) with Swedens lab 1 being an exception.

7.4 Discussion

Positioning

Since zoobenthos samples are taken from the sea floor and not from the free water, and the animals are not evenly distributed in the sediment, the precision in the positioning system is crucial for the obtained results. The results of the intercalibration of the navigators using the Decca system (with the outlying figures from Aranda excluded) showed much greater deviations than the "theoretical accuracy" of the system. Nevertheless the results are acceptable (Arandas values excl.) if you are sampling in areas with "homogeneous" macrofauna.

Surprisingly the results when using satellite navigation was less good than the Decca-navigator results, although satellite navigation is presumed to be better. Also here the results show a much lower precision than the one stated in prospects and manuals for satellite navigators, and the results are not acceptable. If the errors in positioning are of a systematic nature, satellite navigation can be used on stations where only one ship is doing the zoobenthos monitoring, but that is usually only the case for national stations.

Prepared samples

The samples contained only a few well known species and the results of species determination was almost identical.

The results of sorting and counting (Fig. 3) was acceptable for Macoma balthica. For total abundance and the dominating Pontoporeia species the results are acceptable except for the Danish figures. For Harmothoe sarsi the results are comparable for most of the participating laboratories, but not for the Danish, East German and Finnish ones. On the other hand Harmothoe only makes up about 3 % of the total abundance at this station.

The fact that the samples were stained with Bengal Rose, which some laboratories are not used to, may have influenced the results to some degree.

The results of weighing (Fig. 4) are much less comparable than the results of sorting and counting. The picture gets somewhat better if the Danish (for Harmothoe and Pontoporeia) and Eastgerman figures (for Harmothoe) are compensated for the lower numbers found by these laboratories (though this calculation is dubious since one can assume that small animals are missed to a greater extent). Then the differences for these two species becomes less than 20 %. Most probably the differences are bigger since, as mentioned, small animals are normally missed more often than big ones.

The wet weight results for Macoma balthica divides into two groups, within which the figures are comparable. The probable explanation to this is that some laboratories open the bivalves before weighing and other laboratories do not.

The wet weighing procedure opens for many sources of error e. g.:

1. How long time are the animals kept on filter paper?
2. Are they spread out on the filter paper or kept in a clump?
3. Are they weighed altogether in one weighing or in several portions?
4. Are they weighed immediately after blotting on filter paper or are several portions gathered before weighing (in which case longer time passes between blotting and weighing)?
5 Are the animals weighed in closed containers to prevent evaporation before and during weighing?

So it is only what could be expected that the differences in weight figures between laboratories are bigger than for the abundance determinations. On the whole the result of the intercalibration of weighing was not acceptable.

Station 4138

Of the two field sampling stations, this is the one with best resemblance of the BMP-stations in the Baltic proper, with regards to substrate and faunalcomposition. It is therefore good that there are very few significant differences between the results from the different laboratories. Nevertheless the variability is greater than could be accepted for trend monitoring.

As pointed out earlier there are two doubtful species determinations that should be checked, and the same holds for the fact that no Halicryptus were found in the Danish samples.

The Danish figures for total abundance and Pontoporeia affinis were significantly lower than the others. The reason for this is probably mostly an effect of the inadequate sorting efficiency found for the prepared samples (see above). If the Danish values are compensated for the differences in sorting efficiency they **are** more or less in level with the others. The statistically significant greater amount of Hydrobia found by FRG is insignificant,because Hydrobia makes up less than 0.003 % of the total abundance.

This means that there is in fact not possible to show, for this station, any statistically significant differences caused by sieving techniques. **Probably there** are such differences, **but with** only 5 samples per participant, they are overshadowed by the variability of the fauna. The differences are at least as big between each participants set of samples as they are between the sets. Differences caused by sieving might be detected if the number of samples is increased, if a bottom with a very homogeneous benthic community is sampled or if prepared samples,

containing both sediment and known number of animals, are used.

Station 4002

This station had both a sediment and a faunal composition untypical for BMP-stations. However, it harboured species that were supposed to be more sensible to differences in sieving technique, e. g. Pygospio elesans and oligochaets. The variability was much higher at this station than at 4138, and the results are not acceptable for trend monitoring.

The significantly lower numbers of Pygospio, found by the second Swedish lab, and oligochaets, found by the second Swedish lab and the Danish lab, may have been caused by different sieving techniques. On the other hand these species did not occur in the prepared samples, and consequently the sorting efficiency has not been tested for these species. Therefore the differences found can also have been caused when sorting. A species like Pygospio easily fragmentizes during the sieving and handling of samples. If not only the heads were counted as one specimen, it can have greatly influenced the results.

Also for species that should not be very sensible to differences in sieving technique, such as Cardium slaucum and Macoma balthica, significant differences were found. These differences should not have been caused during sorting, since no differences was found for Macoma in the results from the prepared samples, and Cardium, being another bivalve, ought to be found to the same extent.

7.5 Conclusions

At least in the central Baltic proper the Decca navigation system still seemed to be better (at least at the time of the intercalibration) than satellite navigation, and could be used with good results on bottoms with a homogeneous bottom fauna community. The Decca navigators should however be checked

frequently, since the values can, as shown in this intercalibration, be greatly erroneous.

Except for the Danish lab the errors in abundance figures, caused when sorting and counting, seem to be negligible. The errors in biomass figures, caused by differences in the weighing procedure, are much to big.

Some dubious species identifications reported, **indicates the** need for intercalibration exercises on species identification, recommended by the group (see below).

Although the mean abundance values from station 4138 differed much, no differences caused by sieving methods could be established. The lower values found by the Danish lab probably was caused by lower sorting efficiency.

Also for station 4002 it is hard to definitely rule out other sources of error than sieving methods, although there are many statistically significant differences between participants.

7.6 **Recommendations**

The text in the Guidelines, stating how the wet weights are to be determined, must be more precise and the laboratories must follow it strictly, if the wet weight is to be kept as a measure of biomass.

Another intercalibration exercise will have to be performed to establish if differences are caused by different sieving techniques.

Guidelines strictly prescribes the use of the standard van Veen-grab as modified by BMB (Dybern et al. 1976). In spite of this, some monitoring labs are using other equipment. If they continue doing so, an intercalibration exercise, to find out if differences are caused by grabs, is needed.

Since benthos patchiness can greatly reduce the possibilities to distinguish long term trends, the participants felt it necessary to carry out patchiness studies on many, if not all the BMP zoobenthos stations. In connection with this it must be stressed that ships must pay attention to their precision in positioning, and check their navigational equipment often.

The group of participants suggested two new intercalibration exercises. Firstly there should be regional intercalibrations, with participants only from those laboratories normally working in respective area. Secondly there is a need, at least for the labs working in the southern and southwestern part of the convention area, to meet and intercalibrate and discuss species determination.

The group noted that several laboratories are not reporting correctly to the HELCOM secretariat. E. g. the correct RUBIN-codes are not used, the codes are written in the wrong positions, old versions of the reporting formats are used. Many of these errors are caused by the contact addresses in some countries not distributing code-lists, new Guidelines, new reporting forms etc. This must be changed. Also the reporting forms has not been properly revised to fit the changes made in the new Guidelines.

The group did not recommend the development of special codes for all thinkable bottom type descriptions. Sediment descriptions should be reported the way it is now stated in the Guidelines. Codes for stating whether the sediment smells of H_2S or not should however be developed.

The group noted that outliers can still pass the datachecking of the consultant. Anyway all data must be thoroughly checked by the reporting laboratories before sending them to the HELCOM secretariat. Surprisingly this is not stated in the Guidelines.

The group meant it to be a good idea to distribute sampling responsibility for the BMP-stations among contracting parties. This would minimize errors caused by differences in positioning and methods, that would otherwise delay the discovery of trends.

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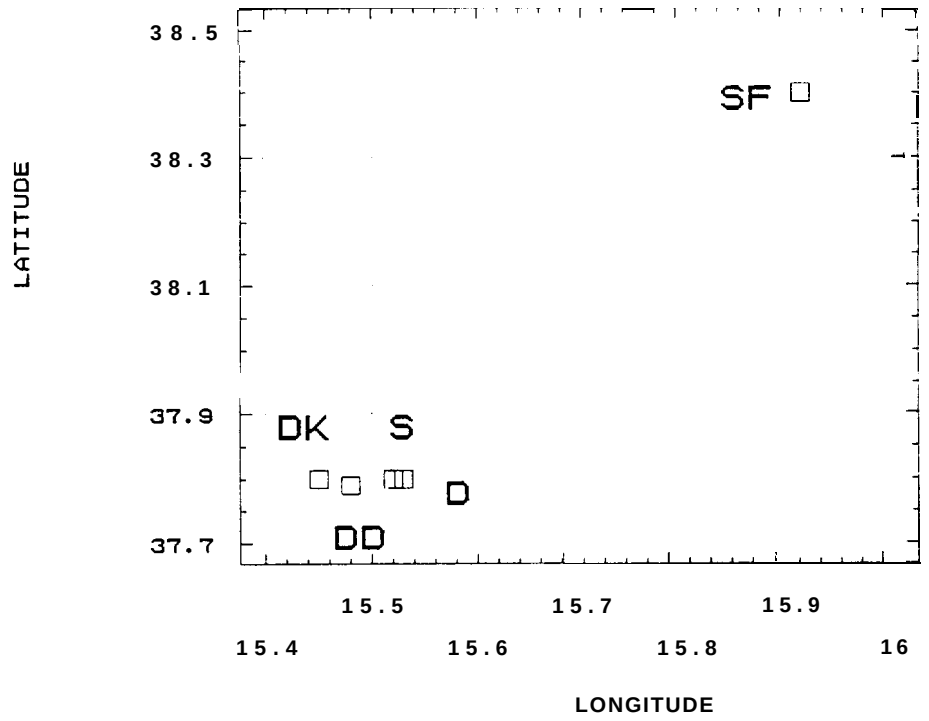


Fig. 1. Positions read fromnavigatorsusing the Decca system.

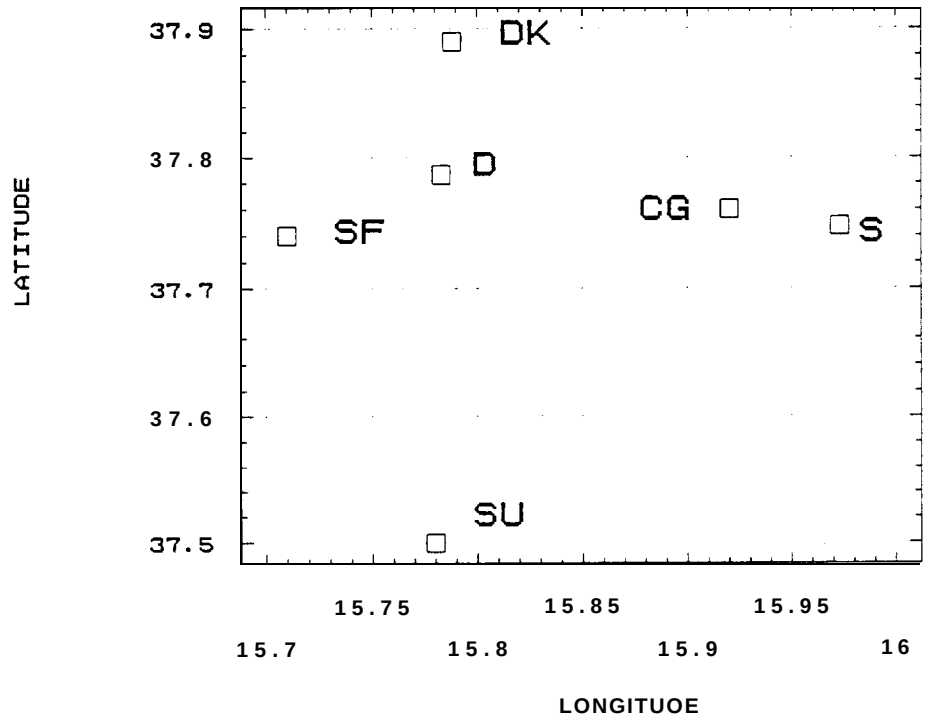


Fig. 2. Positions read from navigators using satellite systems.

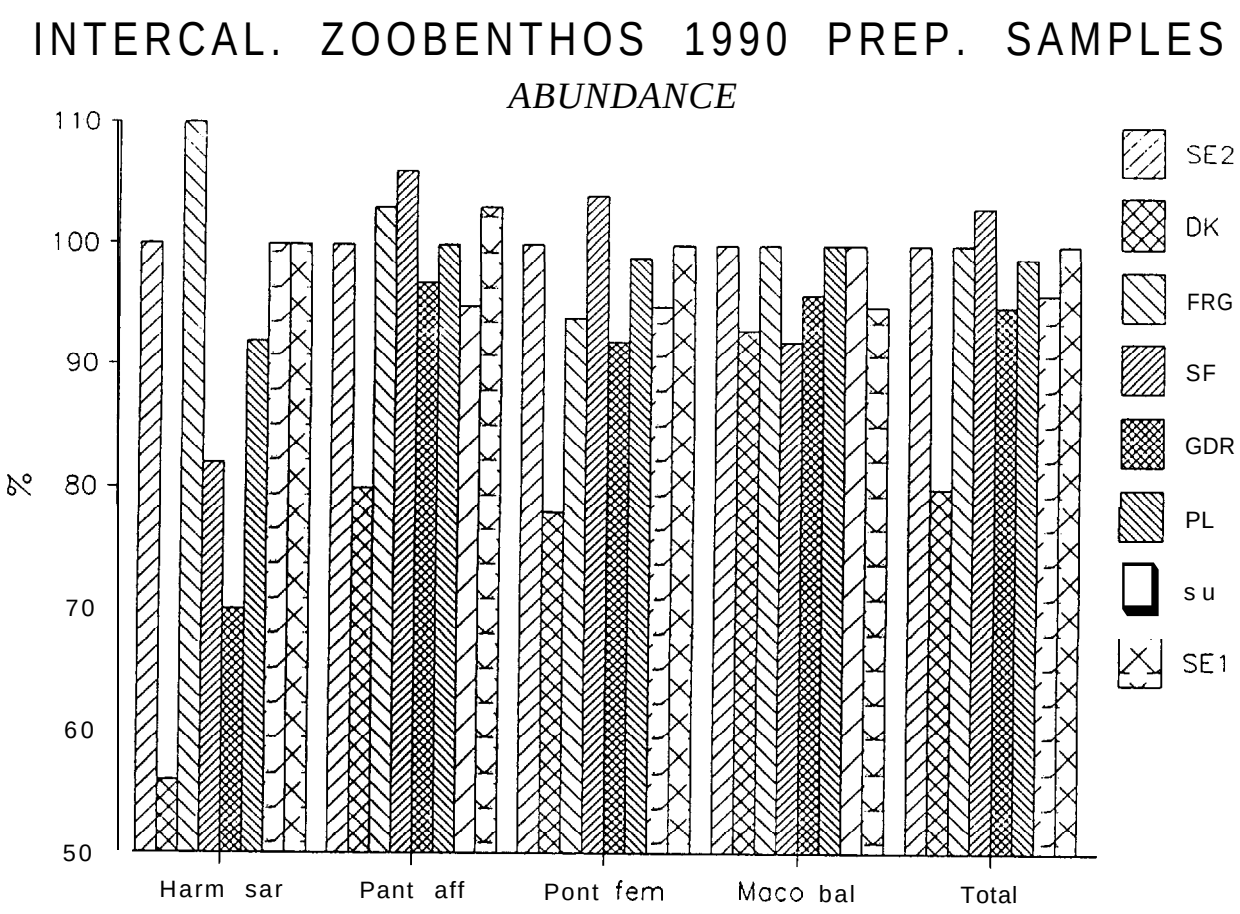


Fig. 3. Percentual relation between the number of individuals of most important species, found by the participating laboratories.

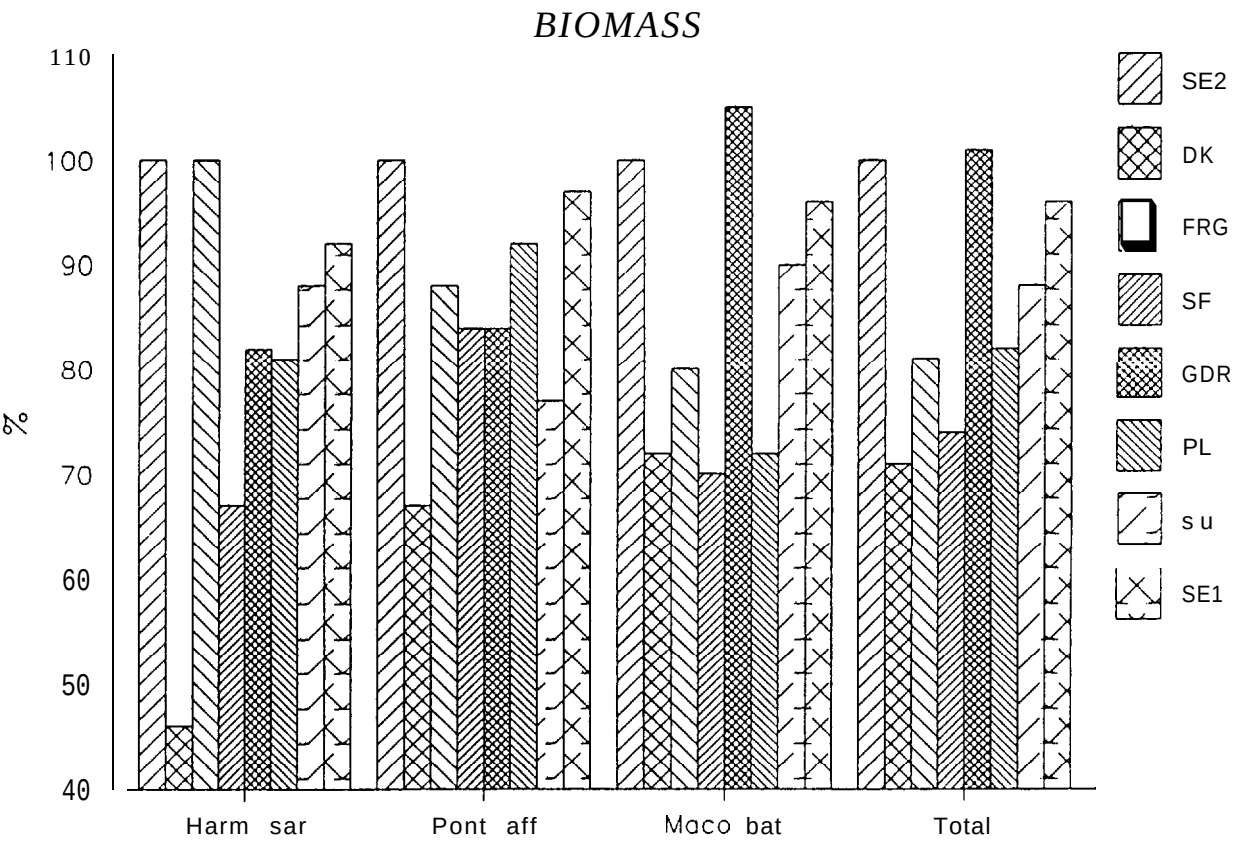


Fig. 4. Percentual relation between wet weights measured by the participating laboratories.

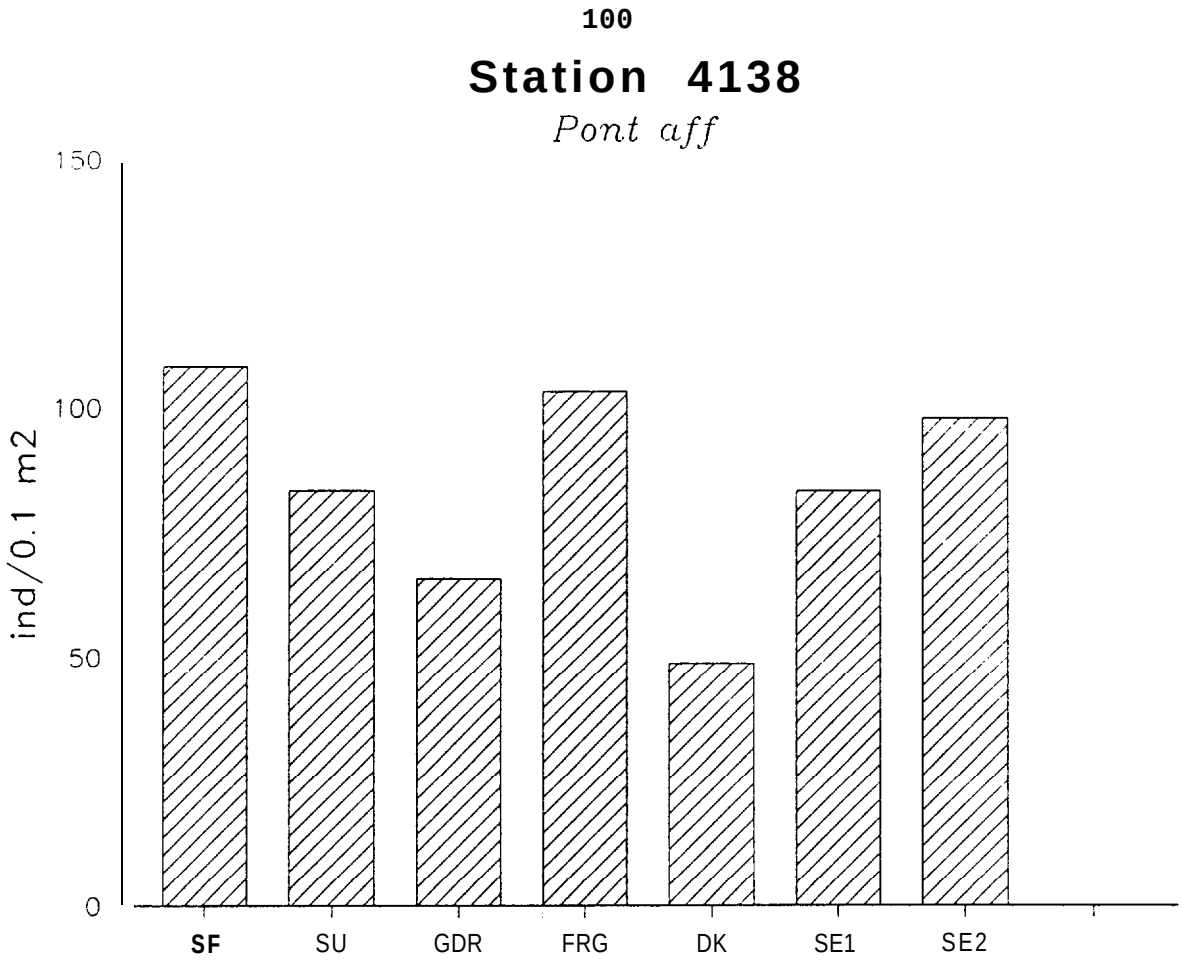


Fig. 5. Number of Pontoporeia affinis (mean per sample) found by the participating laboratories at the station with clayey sediment.

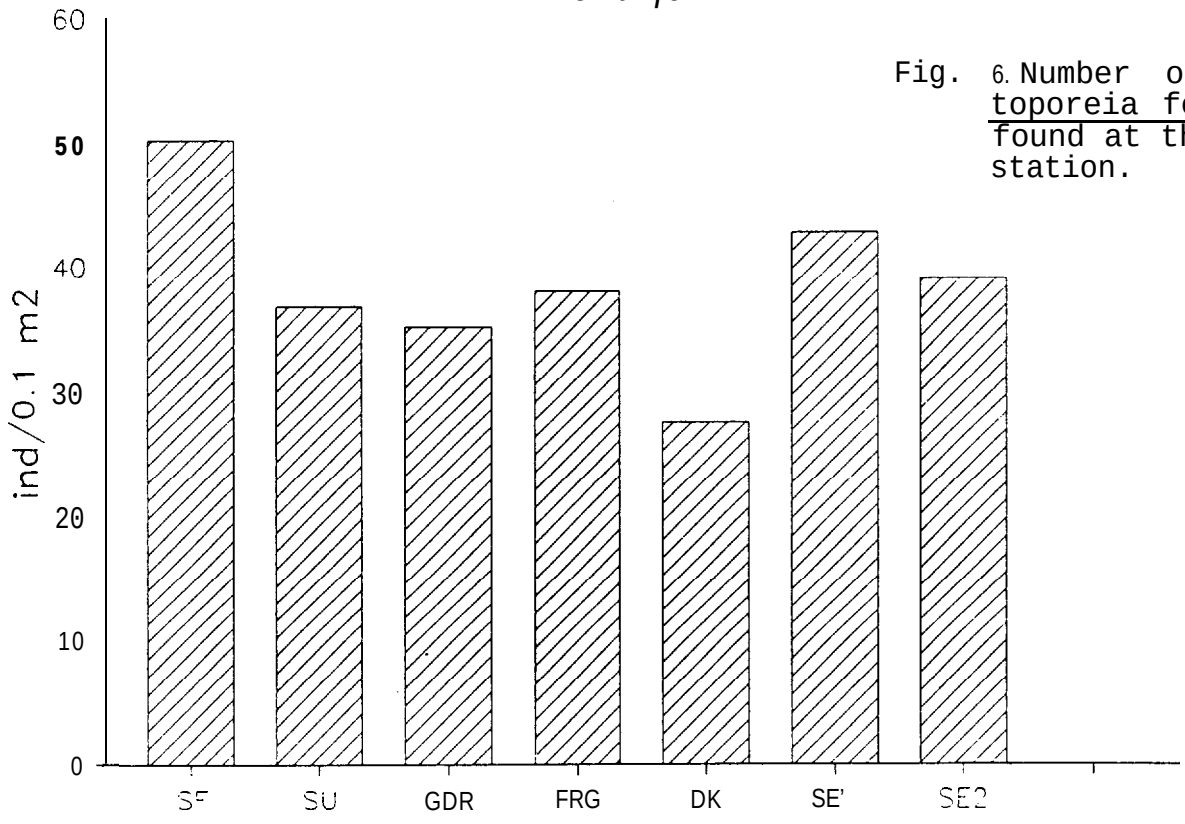


Fig. 6. Number of Pontoporeia femorata found at the clay station.

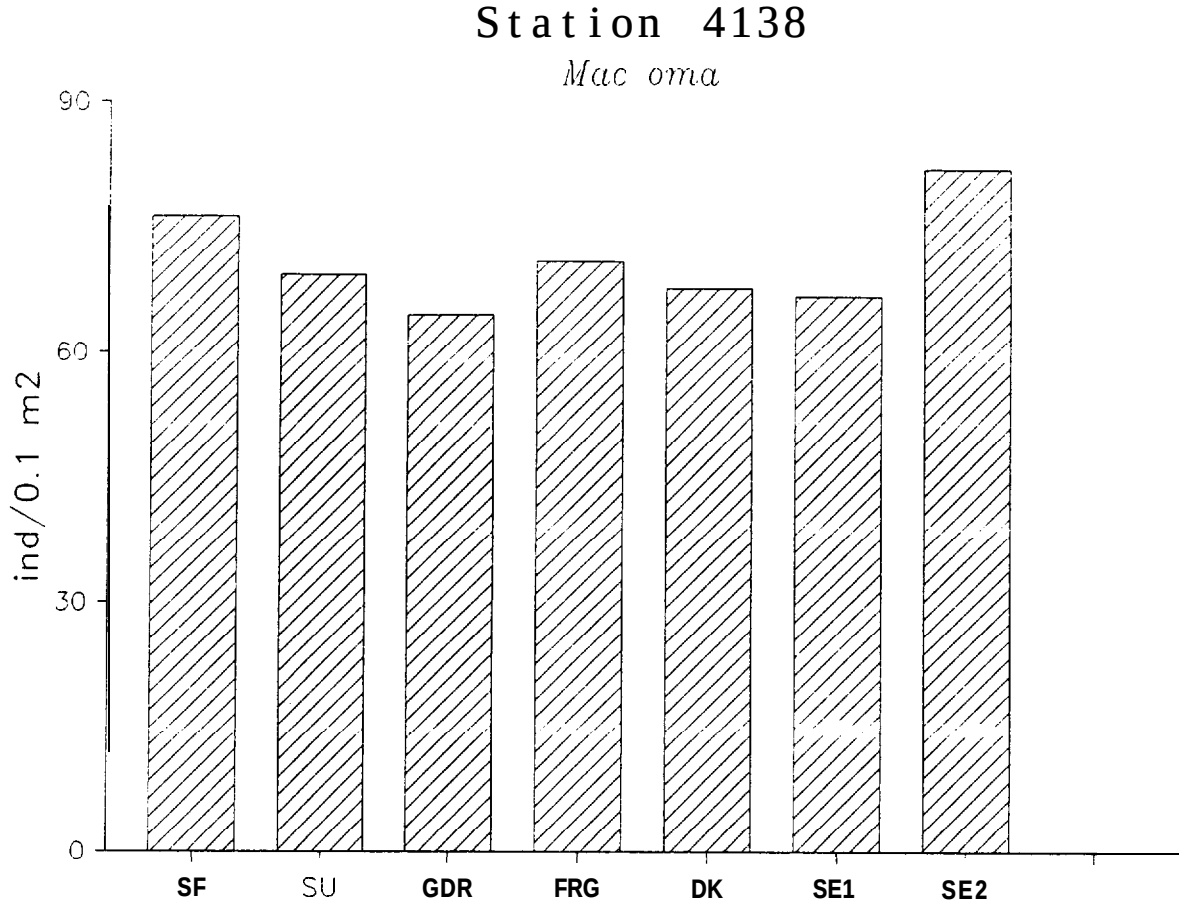


Fig. 7. Number of Macoma balthica (mean per sample) found'by the participating laboratories at the station with clayey sediment.

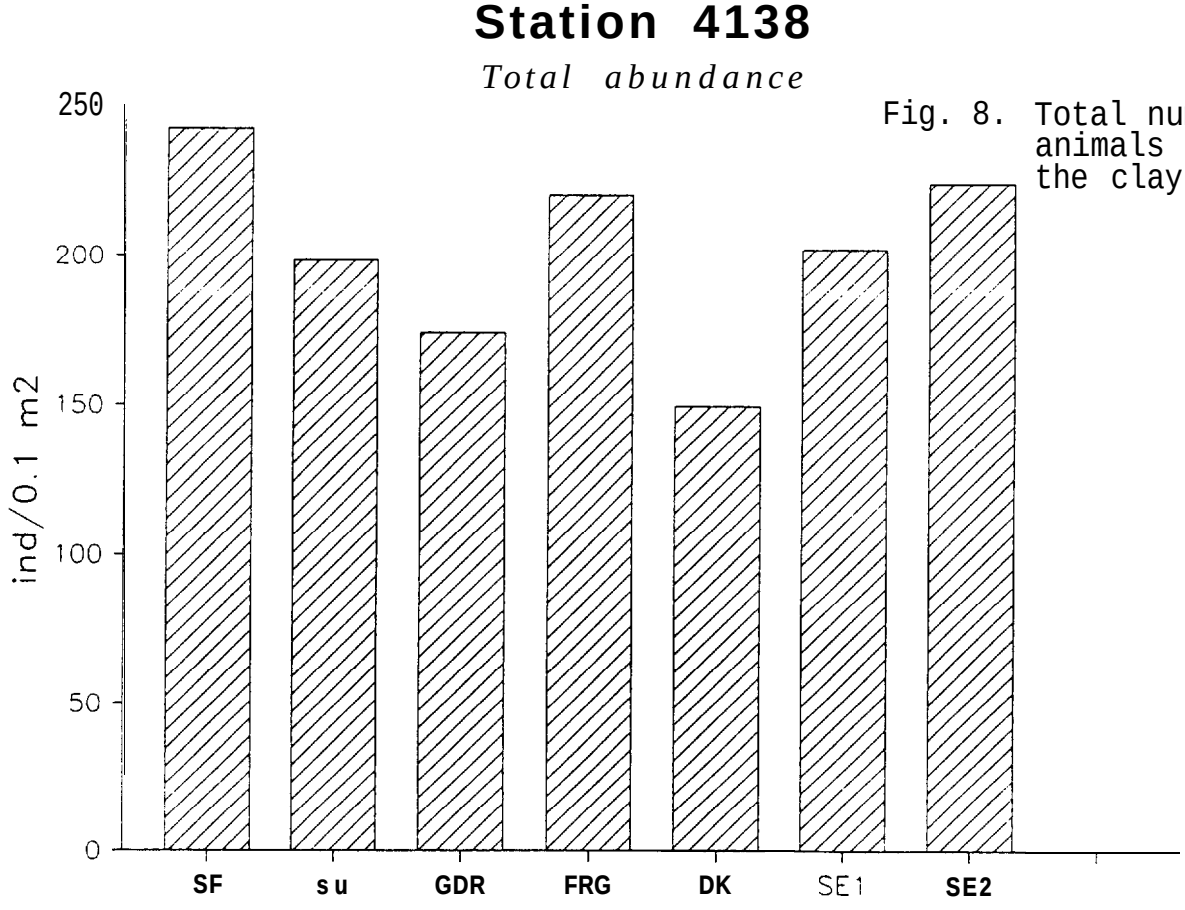


Fig. 8. Total number of animals found at the clay station.

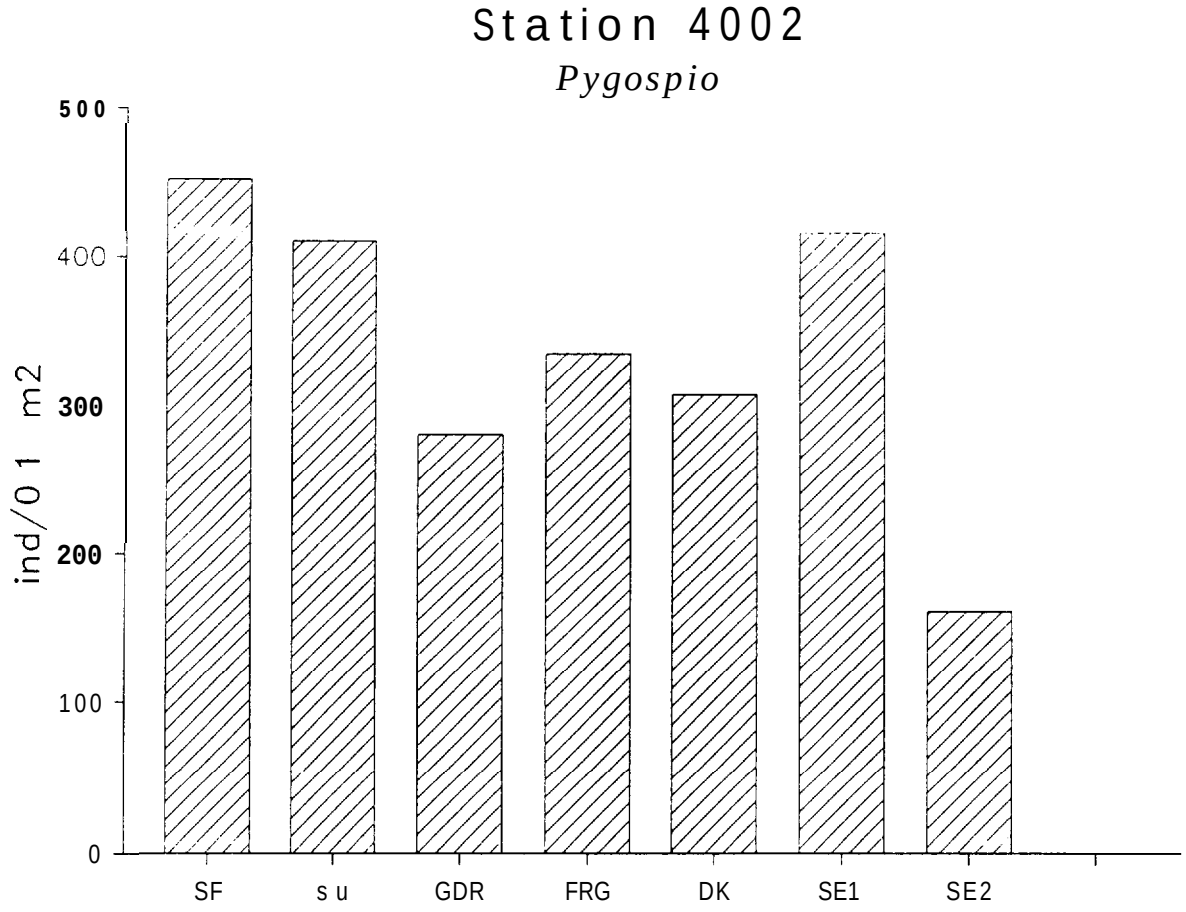


Fig. 9. Number of Pygospio elegans found by the participating laboratories (means per sample) at the station with sandy sediment.

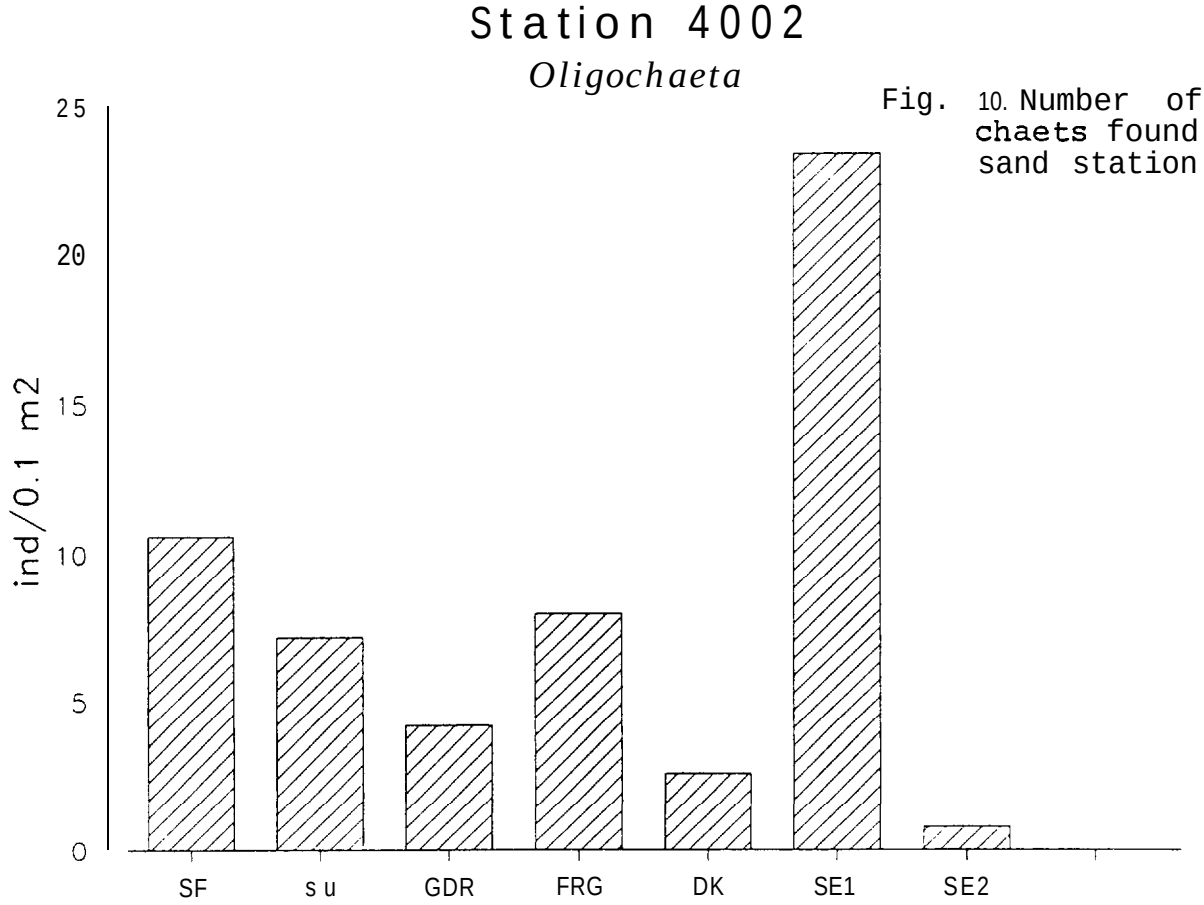


Fig. 10. Number of 01igochaets found at the sand station.

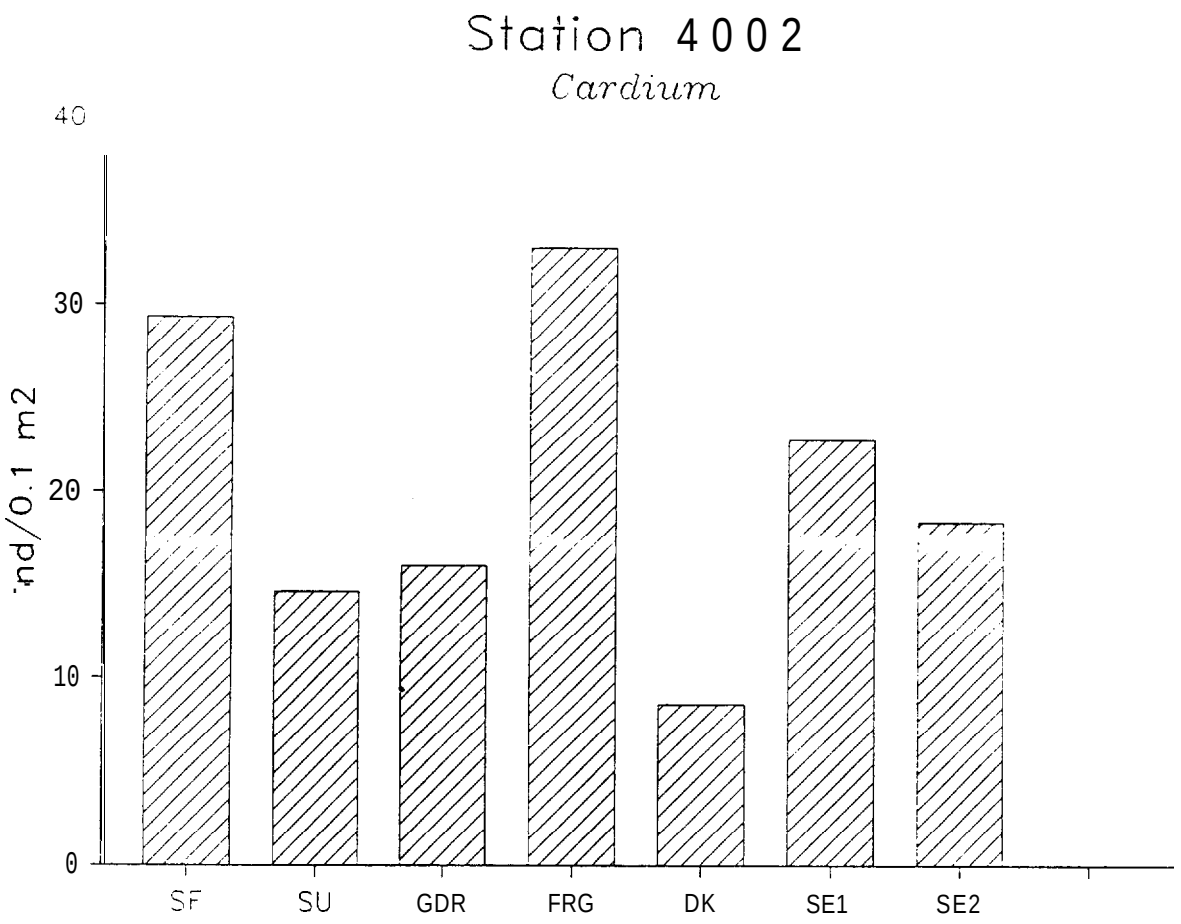


Fig 11. Number of Cardium glaucum (mean values per sample) found by the participating laboratories at the station with sandy sediment.

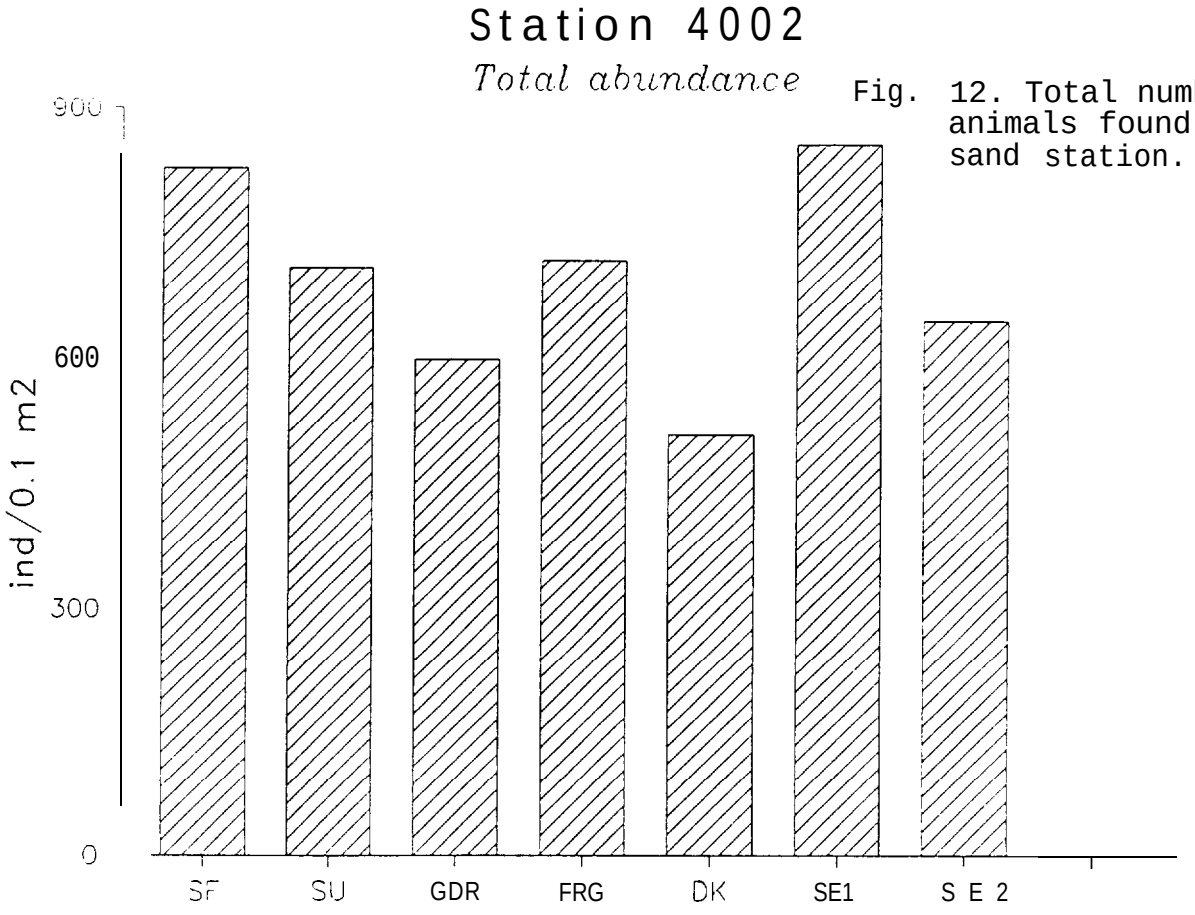


Fig. 12. Total number of animals found at the sand station.

Appendix 1

BIOLOGICAL INTERCALIBRATION WORKSHOP IN VISBY 1990.
LIST OF PARTICIPANTS.

Denmark

Jörgen Norrevang-Jensen	National Environmental Research Institute, Jaegersborg Allé 1 B DK-2920 Charlottenlund

Federal Republic of Germany

Renate Wrage H. Schomann	Institut fur Meereskunde an der Universität Kiel Dusternbrooker Weg 20 D-2300 Kiel

Finland

Ann-Britt Andersin Ari Laine	Finnish Institute of Marine Research P.O.Box 33 SF-00931 Helsinki

German Democratic Republic

Fritz Gosselck	University of Rostock Section Biologie Universitatsplatz 2 DDR-2500 Rostock

Poland

Andrzej Osowiecki	Institute of Environmental Protection ul. Slupska 25 PL-80-392 Gdansk

Sweden 1

Bengt Yhlen

SMHI
Oceanographical Laboratory
P.O.Box 2212
S-403 14 Gothenburg

Sweden 2

Hans Cederwall (convener)

Stockholm Center for Marine Research,
Stockholm University
S-106 91 Stockholm

Soviet Union

Sergei Olenin

Hydrometeorological Observatory of Klaipeda
Taikos
Pr./Avenue 26
SU-235 802 Klaipeda
Lithuania

Appendix 2

Number of Harmothoe sarsi found in the prepared samples.

row	sampleno	Sweden2	Sweden1	Finland	Sovietunio	Poland	GDR	FRG	Denmark
1	3	1	1						
2	8	3	3						
3	19	3	3						
4	25	1	1						
5	34	1	1						
6	18	0		0					
7	21	1		1					
8	23	2		0					
9	27	5		5					
10	31	3		3					
11	5	4			4				
12	12	0			0				
13	28	3			3				
14	32	2			2				
15	33	1			1				
16	1	4				4			
17	6	3							
18	7	4				3			
19	16	0				0			
20	17	1				1			
21	4	0					0		
22	9	0					0		
23	22	6					5		
24	24	2					1		
25	30	2					1		
26	10	2						2	
27	15	2						2	
28	20	2						2	
29	26	1						2	
30	29	3						3	
31	2	4							
32	11	1							
33	13	2							
34	14	2							
35	35	0							

Appendix 3

Numbers of Pontoporeia affinis found in prepared samples.

row	sampleno	Sweden2	Sweden1	Finland	Sovietunio	Poland	GDR	FRG	Denmark
1	3	38	38						
2	8	27	28						
3	19	29	29						
4	25	35	35						
5	34	38	41						
6	18	39		44					
7	21	48		47					
8	23	43		43					
9	27	4-J		50					
10	31	39		44					
11	5	38			33				
12	12	33			37				
13	28	38			37				
14	32	44			40				
15	33	39			36				
16	1	50				49			
17	6	39				39			
18	7	51				51			
19	16	39				40			
20	17	36				35			
21	4	47					47		
22	9	25					25		
23	22	46					45		
24	24	31					31		
25	30	46					42		
26	10	40						40	
27	15	44						47	
28	20	26						29	
29	26	33						34	
30	29	34						33	
31	2	49							39
32	11	28							21
33	13	49							38
34	14	37							32
35	35	38							31

Appendix 4

Numbers of Pontoporeia femorata found in prepared samples.

row	sampleno	Finland	Sovietunio	Poland	GDR	FRG	Denmark	Sweden2	Sweden1
1	3							16	16
2	8							8	8
3	19							18	18
4	25							25	25
5	34							6	6
6	18	16						16	
7	21	19						15	
8	23	17						17	
9	27	19						20	
10	31	9						9	
11	5		17					19	
12	12		23					25	
13	28		14					13	
14	32		17					18	
15	33		12					12	
16	1			24				24	
17	6			22				22	
18	7			22				22	
19	16			20				21	
20	17			13				13	
21	4				16			16	
22	9				20			20	
23	22				15			19	
24	24				5			6	
25	30				13			14	
26	10					15		17	
27	15					18		19	
28	20					21		22	
29	26					16		17	
30	29					15		15	
31	2						9	10	
32	11						12	20	
33	13						15	18	
34	14						9	12	
35	35						11	12	

Appendix 5

Number of Macoma balthica found in prepared samples.

row	sampleno	Sweden2	Finland	Sovietunio	Poland	GDR	FRG	Denmark	Sweden1
1	3	0							0
2	8	3							3
3	19	8							7
4	25	7							7
5	34	2							2
6	18	3	3						
7	21	3	3						
8	23	7	7						
9	27	7	5						
10	31	5	5						
11	5	4			4				
12	12	5			5				
13	28	4							
14	32	7							
15	33	6			6				
16	1	3				3			
17	6	4				4			
18	7	3				3			
19	16	1				1			
20	17	9				9			
21	4	3					3		
22	9	4					4		
23	22	13					12		
24	24	1					1		
25	30	6					6		
26	10	10						10	
27	15	5						5	
28	20	5						5	
29	26	3							
30	29	4						4	
31	2	6							5
32	11	5							5
33	13	5							5
34	14	4							3
35	35	7							7

Appendix 6

Total number of individuals found in prepared samples.

row	sampleno	Sweden2	Sweden1	Finland	Sovietunio	Poland	GDR	FRG	Denmark
1	3	55	55						
2	8	43	43						
3	19	60	59						
4	25	68	68						
5	34	49	50						
6	18	58		63					
7	21	68		71					
8	23	71		69					
9	27	82		82					
10	31	56		61					
11	5	65			58				
12	12	63			65				
13	28	59			58				
14	32	73			68				
15	33	58			55				
16	1	83				82			
17	6	70				69			
18	7	80				79			
19	16	61				61			
20	17	60				59			
21	4	67					67		
22	9	49					49		
23	22	84					77		
24	24	42					39		
25	30	68					62		
26	10	74						71	
27	15	71						73	
28	20	56						58	
29	26	61						61	
30	29	57						56	
31	2	70							57
32	11	56							39
33	13	76							60
34	14	56							47
35	35	57							49

Appendix 7

Number of taxa found in prepared samples.

row	sampleno	Sweden2	Sweden1	Finland	Sovietunio	Poland	GDR	FKG	Denmark
1	3	3	3						
2	8	5	5						
3	19	5	5						
4	25	4	4						
5	34	4	4						
6	18	3		3					
7	21	5		5					
8	23	5		4					
9	27	5		5					
10	31	4		4					
11	5	4			4				
12	12	3			3				
13	28	5			4				
14	32	6			6				
15	33	4			4				
16	1	5				5			
17	6	6				5			
18	7	4				4			
19	16	3				3			
20	17	5				5			
21	4	4					4		
22	9	3					3		
23	22	4					4		
24	24	6					5		
25	30	4					4		
26	10	6						7	
27	15	5						5	
28	20	5						5	
29	26	5						5	
30	29	5						5	
31	2	5							5
32	11	6							5
33	13	5							5
34	14	5							6
35	35	3							3

Appendix 8

Wet weights of Harmothoe sarsi found in the prepared samples.

sampleno	Sweden2	Sweden1	Finland	Sovietunio	Poland	GDR	FRG	Denmar
3	0.00184	1.75E-003						
8	0.00967	8.86E-003						
19	0.00651	6.03E-003						
25	0.00118	9.50E-004						
34	0.00196	1.90E-003						
18	0.00000		0.0000					
21	0.00187		0.0007					
23	0.00340		0.0000					
27	0.01709		0.0138					
31	0.00809		0.0058					
5	0.00482			4.1E-003				
12	0.00000			0.0E0000				
28	0.00560			4.73-003				
32	0.00113			1.3E-003				
33	0.00070			7.0E-004				
1	0.00696				4.E-003			
6	0.00519				5.E-003			
7	0.00658				6.E-003			
16	0.00000				0.E0000			
17	0.00216				2.E-003			
4	0.00000					0.0000		
9	0.00000					0.0000		
22	0.01924					0.0177		
24	0.00500					0.0029		
30	0.00233					0.0013		
10	0.00555						4.8E-003	
15	0.00719						7.0E-003	
20	0.00254						2.3E-003	
26	0.00154						2.8E-003	
29	0.00830						8.1E-003	
2	0.00886							4.7E-0
11	0.00122							0.0E00
13	0.00243							1.1E-0
14	0.00760							3.4E-0
35	0.00000							0.0E00

Appendix 9

Wet weights of Pontoporeia affinis found in prepared samples.

row	sampleno	Sweden2	Sweden1	Finland	Sovietunio	Poland	GDR	FRG	Denmar
	3	0.22118	0.225						
	8	0.16904	0.155						
	19	0.17469	0.171						
4	25	0.20614	0.191						
5	34	0.22976	0.224						
6	18	0.21341		0.1782					
	21	0.28101		0.2244					
	23	0.27312		0.2260					
9	27	0.30428		0.2646					
10	31	0.22919		0.1947					
11	5	0.17846			0.149				
12	12	0.19365			0.152				
13	28	0.20453			0.155				
14	32	0.24552			0.174				
15	33	0.25715			0.203				
16	1	0.29210				0.259			
17	6	0.19739				0.178			
18	7	0.29727				0.293			
19	16	0.22107				0.205			
20	17	0.20611				0.178			
21	4	0.25236					0.2276		
22	9	0.15801					0.1282		
23	22	0.27152					0.2269		
24	24	0.16213					0.1353		
25	30	0.27430					0.2173		
26	10	0.21307						0.1554	
27	15	0.28783						0.2869	
28	20	0.14137						0.1361	
29	26	0.18551						0.1640	
30	29	0.20035						0.1647	
31	2	0.29601							0.19
32	11	0.17129							0.10
33	13	0.26252							0.17
34	14	0.21137							0.14
35	35	0.21743							0.15

Appendix lo

weights of Macoma balthica found in prepared samples.

samoleno	Sweden2	Sweden1	Finland	Sovietunio	Poland	GDR	FRG	Denmar
3	0.00000	0.00						
8	0.91580	0.83						
19	3.30755	3.15						
25	4.45260	4.37						
34	0.80831	0.80						
18	1.30594		0.9905					
21	1.00911		0.6326					
23	3.77602		2.5942					
27	2.13511		1.5534					
31	2.95050		2.3507					
5	2.10221			1.62				
12	2.94603			2.65				
28	2.33921			2.27				
32	2.50551			2.32				
33	0.96531			0.90				
1	2.48953				1.567			
6	2.11508				1.316			
7	1.29760				0.957			
16	0.02111				0.015			
17	4.38985				3.543			
4	2.53560					2.4078		
9	2.01798					2.0277		
22	1.64647					1.7896		
24	0.03340					0.0029		
30	1.13176					1.5018		
10	3.98572						2.8885	
15	1.90743						1.7516	
20	2.83261						2.2324	
26	0.94418						0.7742	
29	2.60606						2.3435	
2	3.65219							2.3
11	3.24144							2.0
13	1.50360							1.4
14	2.88587							2.2
35	3.70994							2.6

Appendix 11

Wet weights of all animals found in the prepared samples.

row	sampleno	Sweden2	Sweden1	Finland	Sovietunio	Poland	GDR	FRG	Denmar
1	3	0.32106	0.32135						
2	8	1.30335	1.18926						
3	19	3.60910	3.43786						
4	25	4.81041	4.69595						
5	34	1.07912	1.05910						
6	18	1.59694		1.2285					
7	21	1.37755		0.9289					
8	23	4.15348		2.9074					
9	27	2.63332		1.9687					
10	31	3.23113		2.5844					
11	5	2.41713			1.858				
12	12	3.27811			2.911				
13	28	2.62408			2.494				
14	32	2.89161			2.595				
15	33	1.28549			1.147				
16	1	2.92374				2.837			
17	6	2.45066				1.622			
18	7	1.73961				1.381			
19	16	0.37529				0.329			
20	17	4.69124				3.809			
21	4	2.87467					2.7094		
22	9	2.29585					2.2511		
23	22	2.05113					2.1040		
24	24	0.23849					0.1898		
25	30	1.49752					1.7914		
26	10	4.44492						3.2266	
27	15	2.38175						2.1354	
28	20	3.09568						2.4788	
29	26	1.24548						1.0307	
30	29	2.97349						2.5860	
31	2	4.00806							2.596
32	11	3.50875							2.248
33	13	1.91869							1.699
34	14	3.19761							2.413
35	35	3.99521							2.866

Appendix 12

INTERCALIBRATION MACROZOOBENTHOS VISBY 1990, STATION 4138

		DENMARK	SWEDEN2	SOVIET U	SWEDEN1	GDR	FRG	FINLRNO
AVERAGE	Halicryptus rpinu	0	.4	.6	.6	.8	.6	.6
	Harmothoe sarsi	.8	1.4	3	2	1.8	2.4	2.8
	Hediste diversico	.2	0	0	0	.2	0	0
	SPIOMORPHA	.2	0	0	0	.2	0	.2
	OLIGOCHAETA	0	0	0	.2	0	.2	0
	Diastylis rathkei	.2	.2	0	0	0	0	0
	Sadurir entomon	3.2	3	4	4	4.8	3.5	3.6
	Idoteo spp.	.2	0	0	.2	0	0	0
	Gammarus spp.	0	0	0	0	.2	0	0
	Pontoporeia affin	48.6	97.8	83.6	83.4	65.8	103.2	108.2
	Pontoporeia femor	27.6	39	36.8	42.8	35.2	38	50.2
	Corophium volutat	0	0	.4	0	0	0	0
	Hydrobio spp.	0	0	0	.2	0	.6	0
	Mytilus edulis	.6	.2	.6	1.6	0	.6	.2
	Macoma balthica	67.8	82	69.4	66.8	64.6	71	76.4
	Total abundance	149.4	224	198.4	201.8	174	220.4	242.2
	No of taxa	5.8	5.4	6	6.4	6.2	6.6	6
MEDIAN	Halicryptus spinu	0	0	0	0	1	1	1
	Harmothoe sarsi	1	2	2	1	1	3	3
	Hediste diversico	0	0	0	0	0	0	0
	SPIOMORPHA	0	0	0	0	0	0	0
	OLIGOCHAETA	0	0	0	0	0	0	0
	Diastylis rathkei	0	0	0	0	0	0	0
	Saduria entomon	3	3	4	4	4	3	4
	Idotea spp.	0	0	0	0	0	0	0
	Gammarus spp.	0	0	0	0	0	0	0
	Pontoporeia affin	SO	92	88	82	53	109	102
	Pontoporeir femor	30	41	30	37	36	39	47
	Corophium volutat	0	0	0	0	0	0	0
	Hydrobia spp.	0	0	0	0	0	1	0
	Mytilus edulis	0	0	0	1	0	0	0
	Macoma balthica	69	84	77	73	66	70	79
	Total abundance	154	214	198	205	171	214	235
	No of taxa	6	5	5	7	7	7	6
VARIANCE	Halicryptus spinu	0	.3	.8	.8	.2	.3	.3
	Harmothoe sarsi	.7	1.8	4	4	2.7	2.3	1.7
	Hediste diversico	.2	0	0	0	.2	0	0
	SPIOMORPHA	.2	0	0	0	.2	0	.2
	OLIGOCHAETA	0	0	0	.2	0	.2	0
	Diastylis rathkei	.2	.2	0	0	0	0	0
	Sadurir entomon	3.2	2.5	2.5	1	2.7	6.7	.3
	Idotea spp.	.2	0	0	.2	0	0	0
	Gammarus spp.	0	0	0	0	.2	0	0
	Pontoporeia rffin	203.3	700.2	186.3	1782.8	654.7	941.7	281.2
	Pontoporeir femor	29.3	206	389.7	237.7	50.7	77.5	88.7
	Corophium volutat	0	0	.3	0	0	0	0
	Hydrobia spp.	0	0	0	.2	0	.3	0
	Mytilus edulis	.8	.2	1.8	6.3	0	.8	.2
	Macoma balthicr	110.7	443.5	314.3	507.7	121.3	133.5	81.8
	Total abundance	475.3	712.5	498.3	3557.2	677	1211.3	461.7
	No of taxa	.7	1.3	2	2.3	1.7	1.3	.5
STD DEV	Halicryptus spinu	0	.547723	.894427	.894427	.447214	.547723	.547723
	Harmothoe sarsi	.83666	1.34164	2	2	1.64317	1.51658	1.30384
	Hediste diversico	.447214	0	0	0	.447214	0	0
	SPIOMORPHA	.447214	0	0	0	.447214	0	.447214
	OLIGOCHAETA	0	0	0	.447214	0	.447214	0
	Diastylis rathkei	.447214	.447214	0	0	0	0	0
	Saduria entomon	1.78885	1.58114	1.58114	1	1.64317	2.58844	.547723
	Idotea spp.	.447214	0	0	.447214	0	0	0
	Gamma-us spp.	0	0	0	0	.447214	0	0
	Pontoporeia affin	14.2583	26.4613	13.6492	42.2232	25.5871	30.6871	16.769
	Pontoporeia femor	5.4129s	14.3527	19.7408	15.4175	7.12039	8.80341	9.41807
	Corophium volutat	0	0	.547723	0	0	0	0
	Hydrobia spp.	0	0	0	.447214	0	.547723	0
	Mytilus edulis	.894427	.447214	1.34164	2.50998	0	.894427	.447214
	Macoma balthica	10.5214	21.0594	17.7285	22.5322	11.0136	11.5542	9.04434
	Total abundance	21.8014	26.6927	22.3226	59.6423	26.0192	34.8037	21.4872
	No of taxa	.83666	1.14018	1.41421	1.51658	1.30384	1.14018	.707107
STD ERR	Halicryptus rpinu	0	.244949	.4	.4	.2	.244949	.244949
	Harmothoe sarsi	.374166	.6	.894427	.894427	.734847	.678233	.583095
	Hediste diversico	.2	0	0	0	.2	0	0
	SPIOMORPHA	.2	0	0	0	.2	0	.2
	OLIGOCHAETA	0	0	0	.2	0	.2	0
	Diostylis rrthkei	.2	.2	0	0	0	0	0
	Saduria entomon	.8	.707107	.707107	.447214	.734847	1.15758	.244949
	Idotea spp.	.2	0	0	.2	0	0	0
	Gammarus spp.	0	0	0	0	.2	0	0
	Pontoporeia affin	6.37652	11.8338	6.1041	18.8828	11.4429	13.7237	7.49933
	Pontoporeia femor	2.42074	6.41872	8.82836	6.89493	3.18434	3.937	4.21189
	Corophium volutat	0	0	.244949	0	0	0	0
	Hydrobia spp.	0	0	0	.2	0	.244949	0
	Mytilus edulis	.4	.2	.6	1.122s	0	.4	.2
	Macoma bolthicr	4.70532	9.41807	7.92843	10.0767	4.92544	5.1672	4.0447s
	Total abundance	9.74987	11.9373	9.98299	26.6728	11.6362	15.5647	9.60937
	No of taxa	.374166	.509902	.632456	.678233	.583095	.509902	.316228

INTERCALIBRATION MACROZOOBENTHOS VISBY *1990, STN 4002*

		DENMARK	FRG	FINLAND	GOR	SOVIET U	SWEDEN1	SWEDEN2
AVERAGE	Total abundance	508.6	717.4	825.8	597.8	708.2	853.8	643
	Harmothoe rrrsi	.2	.4	.2	.2	0	0	.2
	Hcdiste diverrico	21.8	31.6	23	23.8	24	28	19.4
	Pygospio elegans	307.8	335.2	451.2	281.2	410.6	415.4	160.6
	OLIGOCHAETA	2.6	8	10.6	4.2	7.2	23.4	.8
	Erythropr elegans	0	.2	0	0	0	0	0
	Crangon crangon	.6	0	0	0	.2	.2	.8
	Saduria entomon	0	.2	.4	.6	.2	0	.4
	Pontoporeia affin	4	3.4	2.6	1.2	2.6	2	1.6
	Pontoporcia femor	0	0	0	0	.2	0	0
	Bathyporeia pilor	1	0	0	0	.4	2.8	0
	Corophium volutat	.2	0	0	0	0	0	0
	Hydrobia rpp.	87.6	205.8	186.8	182	163.8	244.2	283.6
	Mytilus edulis	1.2	.6	1.4	.4	1	5.6	1.8
	Cardium glaucum	8.6	33	29.4	16	14.6	22.8	18.4
	Macoma balthica	69.4	91.6	113.4	81.8	76.2	100.4	143.8
	Mya arenaria	3.6	7.4	6.8	6.2	7	9	11.6
	No of taxa	9.6	9.4	9	8.6	9.4	9.4	8.8
MEDIAN	Total abundance	541	735	724	601	679	1041	646
	Harmothoe sarsi	0	0	0	0	0	0	0
	Hediste diversico	22	40	26	25	21	32	21
	Pygorpio elegans	370	352	420	265	462	498	161
	OLIGOCHAETA	3	7	8	4	7	23	0
	Erythrops elegans	0	0	0	0	0	0	0
	Crangon crangon	0	0	0	0	0	0	1
	Saduria entomon	0	0	0	0	0	0	0
	Pontoporeia affin	4	3	3	1	3	2	2
	Pontoporeia femor	0	0	0	0	0	0	0
	Bathyporeia pilos	1	0	0	0	0	3	0
	Corophium volutat	0	0	0	0	0	0	0
	Hydrobio spp.	73	173	197	187	148	308	308
	Mytilus edulis	2	1	2	0	1	2	2
	Cardium glaucum	7	33	32	17	12	31	19
	Macoma balthica	66	115	113	92	65	111	136
	Mya arenaria	4	8	8	6	6	8	11
	No of taxa	9	9	9	8	9	10	9
VARIANCE	Total abundance	19995.3	58138.3	31397.2	57930.7	46218.7	222897	8856
	Harmothoe sarsi	.2	.3	.2	.2	0	0	.2
	Hediste diverrico	13.7	144.3	180.5	109.7	99.5	197.5	34.3
	Pygospio elegans	14507.7	8045.7	8507.2	22292.7	14186.8	53108.8	1180.3
	OLIGOCHAETA	4.3	22.5	48.3	3.7	9.7	379.3	3.2
	Erythropr elegans	0	.2	0	0	0	0	0
	Crangon crangon	.8	0	0	0	.2	.2	.7
	Saduria entomon	0	.2	.3	.B	.2	0	.8
	Pontoporeia affin	6.5	.8	1.3	2.7	2.3	3.5	1.3
	Pontoporeia femor	0	0	0	0	.2	0	0
	Bathyporeia pilor	1.5	0	0	0	.8	4.7	0
	Corophium volutat	.2	0	0	0	0	0	0
	Hydrobia spp.	3758.3	14985.2	6186.2	4242	8579.2	22862.2	3989.3
	Mytilus edulis	1.2	.3	.8	.8	1	50.3	1.7
	Cardium glrucum	25.3	108.5	71.8	34	50.8	243.7	25.8
	Macoma balthica	289.8	1317.8	444.8	666.2	1030.7	2045.3	675.2
	Mya • renarir	7.3	5.8	42.7	4.2	16	52	8.3
	No of taxa	2.3	.8	2	1.8	1.3	3.8	.2
STD DEV	Total abundance	141.405	241.119	177.193	240.688	214.985	472.119	94.1063
	Harmothoe sarsi	.447214	.547723	.447214	.447214	0	0	.447214
	Hediste diversico	3.70135	12.0125	13.435	10.4738	9.97497	14.0535	5.85662
	Pygospio elegans	120.448	89.6978	92.2345	149.307	119.108	230.453	34.3555
	OLIGOCHAETA	2.07364	4.74342	6.94982	1.92354	3.11448	19.4756	1.78885
	Erythrops elegans	0	. 447214	0	0	0	0	0
	Crangon crangon	.894427	0	0	0	. 447214	.447214	.83666
	Saduria entomon	0	. 447214	.547723	.894427	. 447214	0	.894427
	Pontoporeia rffin	2.54951	.894427	1.14018	1.64317	1.51658	1.87083	1.14018
	Pontoporeia femor	0	0	0	0	.447214	0	0
	Bathyporeia pilos	1.22474	0	0	0	.894427	2.16795	0
	Corophium volutat	.447214	0	0	0	0	0	0
	Hydrobia spp.	61.305	122.414	78.6524	65.1306	92.624	151.203	63.1609
	Mytilus edulis	1.09545	.547723	.894427	.894427	1	7.09225	1.30384
	Cardium glaucum	5.02991	10.4163	8.47349	5.83095	7.12741	15.6109	5.07937
	Macoma balthica	17.0235	36.3015	21.0903	25.8109	32.1045	45.22s	25.9846
	Mya arenaria	2.70185	2.40832	6.53452	2.04939	4	7.2111	2.88097
	No of taxa	1.51658	.894427	1.41421	1.34164	1.14018	1.94936	.447214
STD ERR	Total abundance	63.2381	107.832	79.2429	107.639	96.1444	211.138	42.0856
	Harmothoe sarsi	.2	.244949	.2	.2	0	0	.2
	Hediste diversico	1.65529	5.37215	6.00833	4.68402	4.46094	6.2849	2.61916
	Pygospio elegans	53.8659	40.1141	41.2485	66.7723	53.2669	103.062	15.3642
	OLIGOCHAETA	.927362	2.12132	3.10805	.860233	1.39284	8.70976	.8
	Erythrops elegans	0	.2	0	0	0	0	0
	Crangon crangon	.4	0	0	0	.2	.2	.374166
	Saduria entomon	0	.2	.244949	.4	.2	0	.4
	Pontoporeia affin	1.14018	.4	.509902	.734847	.678233	.83666	.509902
	Pontoporeia femor	0	0	0	0	.2	0	0
	Bathyporeia pilor	.547723	0	0	0	.4	.969536	0
	Corophium volutat	.2	0	0	0	0	0	0
	Hydrobia spp.	27.4164	54.7452	35.1744	29.1273	41.4227	67.6198	28.2464
	Mytilus edulis	.489898	.244949	.4	.4	.447214	3.17175	.583095
	Cardium glaucum	2.24944	4.65833	3.78946	2.60768	3.18748	6.9014	2.27156
	Macoma balthica	7.61315	16.2345	9.43186	11.543	14.3576	20.2252	11.6207
	Mya arenaria	1.2083	1.07703	2.92233	.916515	1.78685	3.2249	1.28841
	No of taxa	.678233	.4	.632456	.6	.509902	.87178	.2

8. REPORT OF THE WORKING GROUP ON NUTRIENTS

8.1 Participating laboratories:

Denmark (DK)	National Environment Research Institute, Copenhagen H. Ferdinand
Germany (FRG)	Institute of Marine Research, Kiel J. Johanssen, H.P. Hansen
Soviet Union (USSR)	Hydrometeorological Observatory, Klaipeda J. Dubra, G. Grikshas
Finland (SF)	Institute of Marine Research, Helsinki K. Mäkelä
Sweden (S)	S-2 Marine Research Center, Umeå E. Lundberg s-3 Dept. of Systems Ecology and Marine Ecology, University of Stockholm A. Sjösten S-4 Meteorological and Hydrological Inst. (SMHI), Oceanographic Lab., Göteborg M. Larsson, D. Zagradkin S-5 Meteorological and Hydrological Inst. (SMHI), Oceanographic Lab., Göteborg J. Valderrama

0.2 Samples and sampling

In the evening of August 28, the Swedish lab 5 (S-5) delivered, to each laboratory, 5 different stock standards which should be analyzed as unknown samples. The stock standards had been prepared 10 days earlier and preserved with a few drops of chloroform.

The standards contained:

Standard No 1	NH_4-N :	NH_4Cl	104 µmol/l
" 2	NO_2-N :	NaN02	104 µmol/l
" 3	NO_3-N :	KN03	104 µmol/l
" 4	TNP :	glycin ($H_2NCHCOOH$)	6 x 10^4 µmol/l
		+ KH_2PO_4	104 µmol/l
" 5	SiO_4-Si:	Na_2SiF_6	104 µmol/l

Standard No 4 was used both for total-N, total P and PO_4-P.

In the morning of August 29 all ships cruised to a station in the vicinity of Visby. The sampling for nutrients was performed from R/V Argos.

Water from 10 and 55 metres were collected. 4 different samples per laboratory were prepared. The samples were distributed to the other ships by rubberboats and should be kept cool and dark until the analysis started the next day. At least 5 replicates from each sample should be analyzed for:

PO_4-P, total-P, NO_2-N, NO2 + NO_3-N, NH_4-N, total-N and SiO_4-Si.

The samples were:

1. Natural sample from 10 m
2. Natural sample from 55 m
3. Spiked sample from 10 m increased in concentration by the addition of:

PO_4-P	0.19 µmol/l
total-P	0.19 µmol/l
NO_2-N	0.39 µmol/l
NO2 + NO_3-N	1.35 µmol/l
NH_4-N	0.19 µmol/l
total-N (including NO2 + NO_3-N and NH_4-N)	2.7 µmol/l
SiO_4-Si	4,8 µmol/l

4. Spiked sample from 55 m increases in concentration with:

PO_4-P	1.47 µmol/l
total-P	1.47 µmol/l
NO_2-N	0.10 µmol/l
NO2 + NO_3-N	7.94 µmol/l
NH_4-N	0.98 µmol/l
total-N (including NO2 + NO_3-N and NH_4-N)	17.7 µmol/l
SiO_4-Si	24.5 µmol/l

0.3 Laboratory procedures.

Eight groups participated in all steps of the intercalibration. Three of them, USSR, S-3 and S-4 used manual procedures (SF also for ammonia-N), while the rest used automated methods with different analyzers.

The methods are described in "Methods of Seawater Analysis'* edited by Grasshoff K., Ehrhardt M. and Kremling K., Second, Revised and Extended Edition, Verlag Chemie, Weinheim 1983. (ISBN 3-527-25998-8). Smaller modifications are made especially when automated procedures are used. The methods are overviewed in table 1.

All samples were analyzed against own standards except for USSR and S-4 which used the stock-standards provided by S-5.

Working-standards were prepared in either synthetic sea water, sodium chloride-solution, deionized or distilled water. (Table 2).

No filtered samples were analyzed but some labs made corrections caused by turbidity and salt effects. (Table 2).

PO_4-P:

Reaction with acidified molybdate reagent followed by reduction with ascorbic acid. Small variations in the reduction agent can be notable i.e. ascorbic acid is acidified with sulphuric acid or not, as well as ascorbic acid solved in a mixture of water and acetone.

Total-P:

After oxidation of organic phosphorus the samples are treated as PO_4-P samples. All labs used wet digestion in autoclaves with potassium peroxodisulphate only, or in alkaline or acid medium.

NO_2-N:

Reaction of nitrite in acid medium with an aromatic amine (sulphanilamid) leading to the formation of a diazonium compound which is coupled to an other aromatic amine (N - (1-naphthyl) - ethylene diamine dihydrochloride) to give an azo dye. Only small variations in the concentration of the reagents were reported.

NO_3-N:

In buffered samples nitrate is reduced to nitrite and then treated as nitrite samples. Copperized or amalgamated cadmium reductors were used. The buffers used were either ammonium chloride or imidazole.

NH_4-N:

In weak alkaline solution, ammonia reacts with hypochlorite to monochloramine which in the presence of phenol, catalytic amounts of nitroprusside ions and excess of hypochlorite gives indophenol blue. Only small variations in the hypochlorite reagents are worth mentioning. Some labs used hypochlorite other trione (dichloro-s-triazins-2,4,6 (1H,3H,5H)-trione sodium salt) as chlorine donor.

Total-N:

Organic and inorganic nitrogen compounds are oxidized, by wet digestion in autoclaves with potassium peroxodisulphate in alkaline medium, to nitrate and then treated as nitrate samples. Only four labs participated (FRG, SF, S-2 and S-3). FRG and SF used the simultaneous method (for both tot-P and tot-N) while S-2 and S-3 used the method for tot-N only.

SiO_4-Si:

In acid samples dissolved silicon reacts with molybdate and forms silicomolybdic acid which is reduced to blue complexes. A complexing agent is added (mostly oxalic acid) to avoid reduction of the excess molybdate reagent and to eliminate the

influence of phosphate. All labs used oxalic acid as complexing agent except S-5 which used tartaric acid. All labs, except S-5 used ascorbic acid as reducing reagent. S-5 used stanuous chloride dissolved in hydrochloric acid.

8.4 Presentation of data.

All results and calculations are given in tables 3-8 and figures l-4.

Table 3. Results of the stock standards
Table 4. Results of natural sample from 10 m
Table 5. Results of natural sample from 55 m
Table 6. Results of spiked sample from 10 m
Table 7. Results of spiked sample from 55 m
Table 8. Overview of Balt. Intercal. Workshops in Kiel 1977, Rönne 1982 and Visby 1990.

Figure 1. Plots of phosphate and total phosphorus data from separate laboratories
Figure 2. Plots of nitrite and nitrate data from separate laboratories
Figure 3. Plots of ammonia and total nitrogen data from separate laboratories
Figure 4. Plots of silicate data from separate laboratories

8.5 Comments on the results

At the intercalibration exercise in Visby, eight different groups representing five countries participated. This is only a minor part of all the laboratories reporting to HELCOM. Therefore, any general conclusions drawn from this intercomparison are restricted to the limited number of laboratories participating in the exercise, and the fact that among them the well equipped laboratories with long experience may be over-represented.

It can be seen from the standard deviations, SD in Table 8, that for most of the parameters, PO_4, NO_2, NH4 and Tot-N, the SDs are virtually constant over time and concentration ranges. Other parameters are rather worse than better in the latest exercise. The conclusion would be that no improvement in analytical precision has occurred during the time elapsed since then, in spite of the fact that an increasing number of laboratories are using automated techniques. An explanation could be that with the introduction of autoanalyzers there has been a diversion of analytical methods, at least in details such as the composition and concentrations of reagents. This should not be a disadvantage as long as the laboratories strengthen their quality assurance programmes including regular participation in intercalibration exercises.

For the statistical treatment, data that deviate, within each laboratory from the mean more than three times the standard deviation, 3xSD, have been omitted. The same method for elimination of data was applied for the Kiel 1977 data in order to accomplish a comparison between the intercalibration exercises on equal terms.

In the following, the different parameters will be commented separately.

PO_4-P:

The recovery of the spiked amount in the 10 m sample varies from 80 to 125%, which is no cause for alarm, as the concentration level is low, and the range reflects an analytical variation (SD) of $\pm$ 0.04 µmol/l.

Total P:

The results of the intercalibration of inorganic phosphate were, generally speaking, acceptable. Decreased precision and accuracy in the measurements of total phosphorus as compared to inorganic phosphorus should therefore be due to the methods of digestion.

The phosphorus in the stock standard, as well as that added to the spiked samples, contains inorganic phosphorus only, a drawback in the evaluation of methods for digestion of the samples. For future exercises, it is recommended that organic phosphate, e.g. riboflavin 5'-phosphate, is included.

As the 10 m natural sample contains more than 90% organic phosphorus, a poor digestion is expected to have a more pronounced influence on the results as compared with the 55 m natural sample, where only about 30% is organic phosphorus.

NO_2-N:

The DK laboratory reports a concentration in the standard provided by the conveners, which is about 50% too high. However, this is not reflected in other measurements, and might therefore be a result of a miscalculation.

The recoveries of the spiked amount in 55 m spiked seawater are unexpectedly high, 134 - 177%, but with a reasonable agreement between the laboratories. A possible explanation is an error in the spiking procedure by the addition of a very small volume of nitrite solution, 0.5 ml diluted 100000 times with seawater.

NO_2+NO_3 :

The FRG laboratory reports double concentrations of stock standard solutions, presumably due to a second addition of the nitrite concentration.

The DK laboratory shows a clear downwards trend in stock standard concentration towards an end value (10900 µmol/l) that is **not too** far from the expected concentration (10000 µmol/l), which needs to be examined for possible carry-over problems in the analytical technique.

The S-5 laboratory reported all values in the 10 m natural sample less than the detection limit 0.2 µmol/l, where the detection limit was set as 1% of the full range of the recorder, in this

case determined by the large concentration range of the samples. A true detection limit for the method is an order of magnitude lower.

NH_4-N:

It is obvious that the determination of ammonia creates more problems than anything else in the intercomparison. Ammonia measurements are vulnerable to contamination from the ambient air, and it seems likely that the vessel, R/V Argos, on board which the samples were taken, spiked and bottled was contaminated. (The ship is used for fishing **at times.)** This might have had an effect on the different samples distributed, and especially on the results from the laboratory on board Argos, **S-**4, which are all higher than any other reported data.

The FRG laboratory, on the other hand, reported much lower concentrations. However, the determination of the stock standard was also too low. An error in the working standard could explain this deviation from the bulk of the other laboratories in the whole range of measurements.

Total-N:

Only a few laboratories reported any results on total nitrogen, which makes it difficult to draw any conclusions.

The SF laboratory, about 50% too high in the determination of the stock standard, reported the highest results, but not high enough to be explained by the deviation in stock standard measurements.

SiO_4-Si:

Although the measurements of the stock standard solution show a high degree of agreement and a reasonable variation, this is not obvious for the spiked seawater samples.

A consistent deviation from a **100%** recovery of the spiked amount in the 10 m sample could be explained the same way as in the case

of nitrite (see above), i.e. an error in the spiking procedure.

8.6 Recommendations

For future exercises the standards and spiking media of total phosphorus and total nitrogen should contain both organic and inorganic components in order to evaluate the digestion methods in relation to the analyses of nitrate and phosphate.

Special attention should be **directed towards** the ammonia methods, for which recommendations, e.g. on the prevention of contamination, would be an important accomplishment for the improvement of the quality of ammonia data.

TABLE 1.
Overview of analytical methods used

		DK-A (home made)	FRG-A (home made)	SF-A (Skalar) (NH_4 - manual)	USSR-M	S-2-A (Technicon TRAACS 8 8	S-3-M	S-4-M	S-5-A (Alpkem RFA 2)
molybdate - tartrate - H_2SO_4	PO_4-P	x	x	x	x	X	x	x	x
ascorbic acid - H_2SO_4					?		X	X	
ascorbic acid - H_2O		x	x	X					
ascorbic acid - H_2O - acetone						X			X
Digestion with $K_2S_2O_8$ in acid medium (Tot-P only)	Tot-P				?	X			
Digestion with $K_2S_2O_8$ only (Tot-P only)		X					X		-
Digestion with $K_2S_2O_8$ in alkaline medium (Tot-P and Tot-N together)			X	X				X	-
sulphanilamid	NO_2	X	x	x	x	x	x	x	x
naphtylethylendiamin		X	X	X	X	X	X	X	X
Cd/Cu - reductor	NO_3	x	X	X		x	x		X
Cd/Hg - reductor					?			X	
NH_4Cl - butter		x	x	X	?		x	x	
imidazol - buffer		x				X			X
phenol - nitroprussid - etanol	NH_4-N			X					-
phenol - nitroprussid - ethanol - aceton			X						-
phenol - nitroprussid - H_2O		x			?	X	x	x	-
hypochlorite					?	X			
trion		xx		X			x	x	-
citrate - NaOH				X		x	x		-
citrate - NaOH - EDTA		X							-
citrate - boric acid - citrate acid - NaOH					?			X	-
Digestion with $K_2S_2O_8$ in alkaline medium (Tot-N only)	Tot-N				-	X	X	-	-
Digestion with $K_2S_2O_8$ in alkaline medium (Tot-N and Tot-P togethe		-	X	X	-			-	-
molybdate - H_2SO_4	SiO_4-Si	-	X	X	X	-	X	X	X
oxalic acid		-	X	X	?	X		x	
tartaric acid		-				-			X
ascorbic acid		-	X	X	?	X		x	
$SnCl_2$ - HCl		-				-			X

TABLE 2.

Overview of corrections made and preparation of working standards

	turbidity blanks - one reagent omitted	turbidity blanks - by filters apart from absorbance peak	filtered sampler	absorbance of distilled or deionized water subtracted	absorbance of synthetic sea water or NaCl-solution subtracted	correction made for nutrient content in synthetic sea water or NaCl-solution	working standards prepared in destilled or deionized water	working standards prepared in synthetic sea water or NaCl-solution
DK-A (home made)	no	no	no	no	yes all	no	no	yes all
FRG-A (home made)	no	no	no	no	yes all	no	no	yes all
USSR-M	?	?	?	?	?	no	?	?
SF-A (Skalar) (NH_4-manual)	PO_4-P NO_2-N	yes all except NH_4-N	no	yes all	no	no	yes all	no
S-2-A (Technion TRAACS 800)	no	no	no	no	yes all	NH_4-N	no	yes all
S-3-M	yes all	no	no	no	no	no	yes all	no but SiO_4-Si corrected
S-4-M	PO_4-P NO_2-N	no	no	Tot-P NO_3-N NH_4-N SiO_4-Si	no	no	yes all except SiO_4-Si	SiO_4-Si
S-5-A (Alpkem RFA 2)	no	no	no	no	yes all	PO_4-P	no	yes all

TABLE 3.

Intercalibration of stock standards. Replicate analyses. Results in µmol/l. Letters after Lab.code indicates procedure: A = automated, M = Manual. Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	Mean	S_D	CV%	n	Expected µmol/l	%of expect.
PO_4-P	DK-A	(11200)	10060	10100	10267	10450	10520	10267.0	221.7	2.2	5	10000	102.7
	FRG-A	9480	9600	10480	10160	10160		9976.0	421.0	4.2	5		99.8
	USSR-M*	10005	10005	10005	10004	10005		10004.8	0.4	0.0	5		100.0
	SF-A	10212	10286	10434				10310.7	113.0	1.1	3		103.1
	S-2-A	9900						9900.0	-	-	1		99.0
	S-3-M	9960						9960.0					99.6
Tot.-P	DK-A											10000	
	FRG-A	11060	10480	9840	9640	9640		10120.0	600.7	5.9	5		101.2
	USSR-M*												
	SF-A	8100	8300	8600				8333.3	251.7	3.0	3		83.3
	S-2-A	9900						9900.0	-	-	1		99.0
	S-3-M	9980						9980.0	-	-	1		99.8
NO_2-N	DK-A	16006	15250	15250	15500	15375	15360	15445.8	267.4	1.9	6	10000	154.5
	FRG-A	9800	9600	9200	9200	9200		9400.0	262.8	3.0	5		94.0
	USSR-M*	10001	10062	10001	10001	10001		100012	0.4	0.0	5		100.0
	SF-A	10259	10330					10294.5	50.2	0.5	2		102.9
	S-2-A	10800						10800.0		- -	1		108.0
	S-3-M	10000						10600.0		- -	1		100.0
NO_3-N	DK-A	(17273)	13726	12274	11566	11542	10954	12011.2	1066.7	8,9	5	10000	120.1
	FRG-A**	19834	20049	20049	20224	20224		26076.0	161.1	0.8	5		200.8
	USSR-M*	10001	10002	10002	10002	10002		16001.8	0.4	0.0	5		100.0
	SF-A	9	6	3	0	9	9	9 6 9 8 9 0 9 9 9 8		9944.0	62.4	0.6	4		99.4
	S-2-A	9900						9900.0	-	-	1		99.0
	S-3-M	9880						9880.0	-	-	1		98.8

* not calibrated against own standard, but calibrated against delivered standard

** are the NO, - values added?

TABLE 3. (cont.)

Intercalibration of **stock** standards. Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = Manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	Mean	SD	CV%	n	Expected µmol/l	% of expect.
NH_4-N	DK-A	9400	(7200)	8700	8566	8825	8940	86862	319.3	3.6	5	1000o	88.9
	FRG-A	6640	5580	5740	5840	5760		5712.0	102.6	1.8	5		67.1
	USSR-M												
	SF-M	9900	9940	9920	10000	10100		9972.0	80.7	0.8	5		99.7
	S-2-A	11600						11600.0	-	-	1		116.0
	S-3-M	10100						10100.0	-	-	1		101.0
Tot.-N	DK-A											60000	
	FRG-A	59520	57040	60600	59800	60040		59440.0	1423.5	2.4	5		99.1
	USSR-M												
	SF-A	87000	89000	89000	87000			88000.0	1154.7	1.3	4		146.7
	S-2-A	62000						62000.0	-	-	1		103.3
	S-3-M	60000						60000.0	-	-	1		100.0
SiO_4-Si	DK-A											10000	
	FRG-A	9778	9374	10094	10146	10114		10001.2	164.5	1.6	5		100.0
	USSR-M*	10003	10003	10004	10003	10003		10003.0	0.4	0.0	5		100.0
	SF-A	10107	10107	10107	10164			10121.3	28.5	0.3	4		101.2
	S-2-A												
	S-3-M	9820						9820.0		- -	1		982

*** not calibrated against own standard, but calibrated against delivered standard**

TABLE 4.

Intercalibration of **natural sample from 10 m. Replicate analyses. Results in µmol/l. Letters after Lab.code indicates procedure: A =** automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	SD	CV%	n
PO_4-P	DK-A	0.02	0.01	0.00	0.01	0.03						0.01	0.01	81.4	5
	FRG-A	0.09	0.06	0.06	0.06	0.07						0.07	0.01	19.2	5
	USSR-M*	0.03	0.02									0.03	0.01	28.3	2
	SF-A	0.04	**0.04**	**0.05**	**0.05**	**0.05**						0.05	0.01	11.9	5
	S-2-A	0.04	0.04	0.04	0.03	0.03						0.04	0.01	15.2	5
	S-3-M	0.03	0.02	0.02	0.03	0.03						0.03	0.01	21.1	5
	S-4-M	0.05	0.07	0.03	0.05	0.04	0.04	0.04	0.04			0.05	0.01	26.6	8
	S-5-A	0.05	0.03	0.02	0.01	0.02	0.02	(0.21)	0.02	0.03	0.02	0.02	0.01	46.2	9
											Overall	0.04	0.02	50.7	44
Tot.-P	DK-A	0.55	0.49	0.63	(0.90)	0.67						0.59	0.08	13.8	4
	FRG-A	0.27	0.36	0.34	027	029						0.31	0.04	13.6	5
	USSR-M*	0.55	0.55									0.55	0.00	0.0	2
	SF-A	0.40	0.41	0.42	0.42	0.40						0.41	0.01	2.4	5
	S-2-A	0.35	0.34	0.33								0.34	0.01	2.9	3
	S-3-M	0.46	0.43	0.46	0.48	0.47						0.47	0.01	2.1	5
	S-4-M	0.57	0.59	0.52	0.56	0.52	0.52	0.52				0.54	0.03	5.5	7
	S-5-A														
											Overall	0.46	0.11	23.2	31
NO_2-N	DK-A	0.02	0.02	0.01	0.01	0.02						0.02	0.01	34.2	5
	FRG-A	0.06	0.06	0.03	0.02	0.03						0.04	0.02	46.8	5
	USSR-M*	0.01	0.01									0.01	0.00	0.0	2
	SF-A	0.01	0.01	0.01	0.01	0.01						0.01	0.00	0.0	5
	S-2-A	0.09	0.08	0.09	0.08	0.08						0.08	0.01	6.5	5
	S-3-M	0.00	0.00	0.01	0.01	0.00						0.00	0.01	136.9	5
	S-4-M	0.03	0.04	0.04	0.04	0.04	0.03	0.04	0.03			0.04	0.01	14.3	8
	S-5-A	0.03	0.02	0.02	0.02	0.04	0.02	0.02	0.02	0.02	0.03	0.02	0.01	29.1	10
											Overall	0.03	0.02	81.7	45

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5).

TABLE 4. (cont.)

Intercalibration of **natural sample from** 10 **m.** Replicate analyses. Results in µmol/l. Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	SD	CV%	n
NO_3-N	DK-A	0.08	0.08	0.07	0.06	0.07						0.07	0.01	11.6	5
	FRG-A	0.00	0.00	0.05	0.00	0.02						0.01	0.02	156.5	5
	USSR-M*	0.08	0.11									0.10	0.02	22.3	2
	SF-A	0.03	0.03	0.04	0.03	0.04						0.03	0.01	16.1	5
	S-2-A	0.06	0.06	0.06	0.07	0.06						0.06	0.00	7.2	5
	S-3-M	0.06	0.08	0.07	0.08	0.06						0.07	0.01	14.3	5
	S-4-M	0.12	0.12	0.10	0.11	0.10	0.09	0.09	0.09			0.10	0.01	12.5	8
	S-5-A	(<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	co.2			10)
											Overall	0.07	0.03	49.9	35
NH_4-N	DK-A	0.10	0.14	0.09	0.17	(0.30)						0.13	0.04	29.6	4
	FRG-A	0.06	0.00	0.02	0.01	0.04						0.03	0.02	92.6	5
	USSR-M*	0.26	0.24									025	0.01	5.7	2
	SF-M	0.45	0.42	0.41	0.41	0.39						0.42	0.02	5.3	5
	S-2-A	0.14	0.13	0.14								0.14	0.01	4.2	3
	S-3-M	0.04	0.04	0.04	0.04	0.04						0.04	0.00	0.0	5
	S-4-M	0.52	0.75	0.98	0.52	0.89	0.61	0.56				0.69	0.19	27.0	7
	S-5-A														
											Overall	0.28	0.27	98.3	31
Tot.-N	DK-A														
	FRG-A	19.02	19.88	18.32	18.36	18.54						18.82	0.65	3.5	5
	USSR-M*														
	SF-A	21.1	21.1	21.7	21.1	20.7						21.14	0.36	1.7	5
	S-2-A	17.64	18.68	18.21	18.39	18.32						18.25	0.38	2.1	5
	S-3-M	18.7	19.5	20.2	19.6	20.1						19.62	0.60	3.0	5
	S-4-M														
	S-5-A														
											Overall	19.46	1.21	6.2	20

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5).

TABLE 4. (cont.)

Intercalibration of natural sample **from 10 m.** Replicate analyses. Results in µmol/l.
Letters after Lab-code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	SD	CV%	n
SiO_4-Si	DK-A														
	FRG-A	8.44	8.60	8.79	8.93	9.02						8.76	0.24	2.7	5
	USSR-M*	10.45	10.64									10.60	0.06	0.6	2
	SF-A	8.4	8.3	8.3	8.6	8.6						8.44	0.15	1.8	5
	S-2-A														
	S-3-M	9.07	9.07	9.03	9.03	9.03						9.05	0.02	0.2	5
	S-4-M	10.3	(11.0)	8.6	8.6	9.1	8.8	9.1	9.1			9.09	0.58	6.4	7
	S-5-A	9.7	9.7	9.7	9.7	9.7	9.7	9.6	9.6	9.6	9.7	9.67	0.05	0.5	10
											Overall	9.19	0.60	6.6	34

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5).

TABLE 5.

Intercalibration of **natural sample from 55 m.** Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	S_D	CV%	n
PO_4-P	DK-A	0.65	0.65	0.65	0.67	0.66						0.66	0.01	1.4	5
	FRG-A	0.58	0.61	0.64	0.64	0.64						0.62	0.03	4.3	5
	USSR-M*	0.67	0.67									0.67	0.00	0.0	2
	SF-A	0.66	0.67	0.64	0.66	0.64						0.65	0.01	2.1	5
	S-2-A	0.69	0.69	0.68	0.68	0.68						0.68	0.01	0.8	5
	S-3-M	0.69	0.68	0.68	0.70	0.69						0.69	0.01	1.2	5
	S-4-M	0.72	0.72	0.72	0.73	0.75	0.76	0.76	0.74			0.74	0.02	2.4	8
	S-5-A	0.66	0.70	0.68	0.67	0.67	0.71	0.70	0.66	0.67	(0.79)	0.68	0.02	2.8	9
											Overall	0.68	0.04	5.5	44
To.-P	DK-A	1.14	1.02	1.13	1.19	0.99						1.09	0.09	7.8	5
	FRG-A	0.73	0.72	0.71	0.75	0.76						0.73	0.02	2.8	5
	USSR-M*	1.06	1.07									1.07	0.01	0.7	2
	SF-A	0.87	0.87	0.93	0.87	0.86						0.88	0.03	3.2	5
	S-2-A	0.84	0.85	0.83								0.84	0.01	1.2	3
	S-3-M	0.95	1.01	0.97	0.95	0.93						0.96	0.03	3.2	5
	S-4-M	1.02	1.05	1.05	1.07	1.05	1.05	1.06				1.05	0.02	1.5	7
	S-5-A														
											Overall	0.95	0.13	13.8	32
NO_2-N	DK-A	0.14	0.15	0.15	0.15	0.16						0.15	0.01	4.7	5
	FRG-A	0.18	0.16	0.14	0.15	0.13						0.15	0.02	12.7	5
	USSR-M*	0.17	0.17									0.17	0.00	0.0	2
	SF-A	0.19	0.18	0.18	0.18	0.18						0.18	0.00	2.5	5
	S-2-A	0.21	0.22	0.21	0.20	021						0.21	0.01	3.4	5
	S-3-M	0.17	0.16	0.17	0.17	0.17						0.17	0.00	2.7	5
	S-4-M	0.20	0.20	0.20	0.20	020	0.20	0.20	0.20			0.20	0.00	0.0	8
	S-5-A	0.18	0.20	0.18	0.18	0.17	0.18	0.18	0.19	0.17	020	0.18	0.01	5.8	10
											Overall	0.18	0.02	11.9	45

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5).

TABLE 5. (cont.)

Intercalibration of natural sample from 55 m. Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	S_D	CV%	n
NO_3+ NO_3-N	DK-A	2.89	2.83	2.89	2.95	2.85						2.88	0.05	1.6	5
	FRG-A	3.24	325	3.35	3.35	3.37						3.31	0.06	1.9	5
	USSR-M*	2.63	2.71									2.67	0.06	2.1	2
	SF-A	(3.36)	323	3.21	3.21	3.18						3.21	0.02	0.6	4
	S-2-A	3.37	3.35	3.35	3.29	3.30						3.33	0.04	1.0	5
	S-3-M	3.43	3.44	3.45	3.44	3.42						3.44	0.01	0.3	5
	S-4-M	3.23	3.28	3.40	3.51	325	3.46	3.47	3.44			3.38	0.11	3.3	8
	S-5-A	3.43	3.67	3.67	3.59	3.58	3.62	3.59	3.65	3.49	3.54	3.58	0.08	2.2	10
											Overall	3.32	0.26	7.7	44
NH_4-N	DK-A	0.26	0.20	0.17	0.31	025						0.24	0.05	22.9	5
	FRG-A	0.32	0.41	0.30	0.42	0.44						0.38	0.06	16.8	5
	USSR-M*	0.67	0.70									0.69	0.02	3.1	2
	SF-M	0.40	0.40	0.41	0.42	0.46						0.42	0.03	6.0	5
	S-2-A	0.55	0.54	0.52								0.54	0.02	2.8	3
	S-3-M	0.53	0.54	0.58	0.55	0.57						0.55	0.02	3.7	5
	S-4-M	1.09	(0.94)	1.18	1.13	122	1.11	1.06	1.18			1.14	0.06	5.0	7
	S-5-A														
											Overall	0.59	0.32	54.3	32
Tot.-N	DK-A														
	FRG-A	18.38	18.57	18.36	18.20	18.43						18.39	0.13	0.7	5
	USSR-M*														
	SF-A	21.7	21.5	22.3	21.7	21.5						21.74	0.33	1.5	5
	S-2-A	19.97	18.95	19.86	19.87	19.43						19.62	0.43	2.2	5
	S-3-M	18.7	18.6	18.8	19.3	19.3						18.94	0.34	1.8	5
	S-4-M														
	S-5-A														
											Overall	19.67	1.34	6.8	20

* not calibrated against own standard, but calibrated against delivered standard
(the same standards as were used by lab S-4 and S-5)

TABLE 5. (cont.)

Intercalibration of natural sample from 55 m. Replicate analyses. Results in µmol/l. Letters after Lab.code indicates procedure: A = automated, M = manual. Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	SD	CV%	n
SiO_4-Si	DK-A														
	FRG-A	13.04	13.12	13.06	13.13	13.29						13.13	0.10	0.7	5
	USSR-M*	19.47	19.65									19.66	0.13	0.7	2
	SF-A	12.6	(12.6)	12.7	12.7	12.6						(12.64)	0.06	0.4	5
	S-2-A														
	S-3-M	13.4	13.3	13.4	13.4	13.4						13.36	0.05	0.3	5
	S-4-M	13.0	(12.2)	13.3	(12.1)	12.1	(12.9)	11.6	(12.1)			(12.41)	0.58	4.7	8
	S-5-A	14.0	14.0	13.9	13.9	14.0	14.0	14.0	14.0	14.0	14.0	13.98	0.04	0.3	10
											Overall	13.54	1.65	12.2	35

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5).

TABLE 6.

Intercalibration of spiked sample from 10 m. Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	S_D	CV%	n	Added µmol/l	Recovery %
PO_4-P	DK-A	0.20	0.19	0.19	0.19	0.22						0.20	0.01	6.6	5	0.19	96.8
	FRG-A	0.24	0.24	(0.17)	0.22	0.22						0.23	0.01	5.0	4		85.3
	USSR-M*	0.22	0.22									0.22	0.00	0.0	2		102.6
	SF-A	0.20	0.20	0.21	0.20	0.20						0.20	0.00	2.2	5		82.1
	S-2-A	0.20	0.19	0.18	0.19	0.18						0.19	0.01	4.5	5		80.0
	S-3-M	0.18	0.21	0.19	0.19	0.19						0.19	0.01	5.7	5		87.4
	S-4-M	0.31	0.31	0.32	0.31	0.26	0.26	0.24	0.26			0.28	0.03	11.1	8		125.8
	S-5-A	0.26	0.26	0.26	0.27	0.26	0.26	0.25	0.26	0.25	0.25	0.26	0.01	2.5	10		123.2
											Overall	0.23	0.04	17.0	44		
Tot.-P	DK-A	0.84	0.78	0.81	1.06	1.14						0.93	0.16	17.6	5	0.19	179.5
	FRGA	0.44	0.48	0.46	0.57	0.53						0.50	0.05	10.7	5		100.0
	USSR-M*	0.71	0.71									0.71	0.00	0.0	2		84.2
	SF-A	0.68	0.70	0.70	0.69	0.67						0.69	0.01	1.9	5		146.3
	S-2-A	0.60	0.56	0.56								0.57	0.02	4.0	3		122.6
	S-3-M	0.66	0.66	0.63	0.65	0.67						0.65	0.02	2.3	5		94.7
	S-4-M	0.83	0.80	0.80	0.77	0.77	0.85	0.85	0.87			0.82	0.04	4.7	8		144.7
	S-5-A																
											Overall	0.71	0.15	21.6	33		

* not calibrated against own standard, but calibrated against delivered standard
(the same standards as were used by lab S-4 and S-5)

TABLE 6. (cont.)

Intercalibration of **spiked sample from 10 m**. Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	S_D	CV%	n	Added µmol/l	Recovery %
NO_3-N	DK-A	0.41	0.40	0.41	0.40	0.40						0.40	0.01	1.4	5	0.39	99.5
	FRG-A	0.44	0.45	0.39	0.40	0.40						0.42	0.09	6.5	5		96.4
	USSR-M*	0.39	0.39									0.39	0.00	0.0	2		97.4
	SF-A	0.39	0.40	0.39	0.40	0.39						0.39	0.01	1.4	5		98.5
	S-2-A	0.47	0.50	0.60	0.48	0.48						0.49	0.01	2.8	5		103.1
	S-3-M	0.33	0.34	0.34	0.34	0.34						0.34	0.00	1.3	5		87.2
	S-4-M	0.41	0.40	0.40	0.41	0.40	0.40	0.40	0.40			0.40	0.01	1.2	8		94.1
	S-5-A	0.44	0.45	0.45	0.44	0.45	0.44	0.44	0.43	0.44	0.45	0.44	0.01	1.5	10		107.4
											Overall	0.41	0.04	9.8	45		
NO_2 +	DK-A	1.20	1.31	1.33	1.22	1.24						1.26	0.06	4.5	5	1.35	88.0
NO_3-N	FRG-A	1.26	1.24	1.28	1.26	1.31						1.27	0.03	2.1	5		93.0
	USSR-M*	1.23	1.23									1.23	0.00	0.0	2		84.1
	SF-A	1.25	1.23	1.20	1.20	1.25						1.23	0.03	2.0	5		88.3
	S-2-A	1.38	1.36	1.37	1.37	1.34						1.36	0.02	1.0	5		96.4
	S-3-M	1.37	1.36	1.35	1.36	1.34						1.36	0.01	0.8	5		96.6
	S-4-M	1.26	1.29	1.32	1.26	1.24	1.28	1.24	1.33			1.28	0.03	2.7	8		87.0
	S-5-A	1.31	1.37	1.32	1.34	1.33	1.32	1.34	1.32	1.30	1.30	1.33	0.02	1.6	10		(98.1)
											Overall	1.30	0.05	4.2	45		

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5)

TABLE 6. (cont.)

Intercalibration of **spiked sample from 10 m.** Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean		S_D CV%	n	Added µmol/l	Recovery
NH,-N	DK-A	0.77	(0.57)	0.75	0.73	0.74						0.75	0.02	2.3	4	0.19	327.9
	FRGA	0.14	(0.20)	0.14	0.11	0.13						0.13	0.01	10.9	4		54.7
	USSR-M*	0.60	0.53									0.57	0.05	8.8	2		165.8
	SF-M	0.59	0.65	0.56	0.59	0.59						0.60	0.03	5.5	5		94.7
	S-2-A	0.40	0.42	0.40								0.41	0.01	2.8	3		142.1
	S-3-M	0.14	0.14	0.14	0.14	0.15						0.14	0.00	3.1	5		53.7
	S-4-M	0.88	0.80	0.88	1.01	(1.33)	0.90	0.92	0.98			0.91	0.07	7.6	7		115.8
	S-5-M																
											Overall	0.53	0.31	57.5	30		
Tot.-N	DK-A															2.7	
	FRGA	22.70	21.09	22.65	22.33	22.31						22.22	0.65	2.9	5		125.6
	USSR-M*																
	SF-A	26.1	25.9	26.3	26.1	25.3						25.94	0.39	1.5	5		177.8
	S-2-A	22.18	21.80	23.03	21.90	22.42						22.27	0.49	2.2	5		148.8
	S-3-M	24.5	23.6	22.4	23.6	24.0						23.62	0.78	3.3	5		148.0
	S-4-M																
	S-5-A																
											Overall	23.51	1.64	7.0	20		

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5)

TABLE 6. (cont.)

Intercalibration of **spiked sample from 10 m.** Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	S_D	CV%	n	Added µmol/l	Recovery %
SiO_4-Si	DK-A															4.8	
	FRGA	17.45	17.45	17.75	17.68	17.74						17.61	0.15	0.9	5		184.5
	USSR-M*	19.91	20.18									20.05	0.19	1.0	2		199.0
	SF-A	17.6	17.4	17.5	17.3	17.7						17.50	0.16	0.9	5		188.8
	S-2-A																
	S-3-M	17.9	17.9	17.9	17.9	17.8						17.88	0.05	0.3	5		184.0
	S-4-M	17.0	18.2	16.9	16.3	16.4	16.4	16.4	14.8			16.55	0.94	5.7	8		155.5
	S-5-A	19.4	19.3	19.2	19.2	19.2	19.2	19.2	19.2	19.3	19.2	19.24	0.07	0.4	10		199.4
											Overall	18.00	1.20	6.7	35		

* not calibrated against own standard, but calibrated against delivered standard
(the same standards as were used by lab S-4 and S-5)

TABLE 7.

Intercalibration of **spiked sample from 55 m.** Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean		S_D	CV%	n	Added µmol/l	Recovery %
PO_4-P	DK-A	2.12	2.15	2.19	2.15	2.15						2.15	0.03		1.2	5	1.47	101.8
	FRG-A	1.96	2.05	2.08	2.11	2.10						2.06	0.06		2.9	5		97.8
	USSR-M*	2.09	2.09									2.09	0.00		0.0	2		96.6
	SF-A	2.07	2.08	2.07	2.07	2.07						2.07	00.0		0.2	5		96.5
	S-2-A	2.12	2.08	2.10								2.10	0.02		1.0	3		96.3
	S-3-M	2.15	2.14	2.15	2.11	2.13						2.14	0.02		0.8	5		98.5
	S-4-M	2.21	2.19	2.21	2.21	2.22	2.22	2.22	2.21			2.21	0.01		0.4	8		100.2
	S-5-A	(2.11)	2.20	2.21	2.23	2.24	2.25	2.24	2.24	2.24	2.24	2.23	0.02		0.7	9		105.6
											Overall	2.15	0.07		3.2	42		
Tot.-P	DK-A	2.54	2.55	2.56	2.67	2.75						2.61	0.09		3.5	5	1.47	103.4
	FRG-A	2.11	2.19	2.09	2.27	2.03						2.14	0.09		4.4	5		95.5
	USSR-M*	2.42	2.46									2.44	0.03		1.2	2		93.5
	SF-A	2.23	2.25	2.19	2.12	2.06						2.17	0.08		3.6	5		87.8
	S-Z-A	2.43	2.37	2.36								2.39	0.04		1.6	3		105.2
	S-3-M	2.42	2.42	2.41	2.40	2.40						2.41	0.01		0.4	5		98.6
	S-4-M	2.68	2.65	2.70	2.63	2.68	2.68	2.70	2.65			2.67	0.03		0.9	8		110.3
	S-5-A																	
											Overall	2.43	0.22		9.0	33		

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5)

TABLE 7. (cont.)

Intercalibration of **spiked sample from 55 m.** Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	S_D	CV%	n	Added µmol/l	Recovery %
NO_2-N	DK-A	0.30	0.31	0.31	0.31	0.30						0.31	0.01	1.8	5	0.10	156.0
	FRGA	0.32	0.31	0.27	0.27	0.28						0.29	0.02	8.1	5		138.0
	USSR-M*	0.34	0.35									0.35	0.01	2.1	2		175.0
	SF-A	0.33	0.33	0.33	0.32	0.33						0.33	0.00	1.4	5		146.0
	S-2-A	0.37	0.38	0.37	0.38	0.37						0.37	0.01	1.5	5		164.0
	S-3-M	0.30	0.30	0.30	0.31	0.30						0.30	0.00	1.5	5		134.0
	S-4-M	0.33	0.34	0.35	0.35	0.35	0.35	0.34	0.34			0.34	0.01	2.2	8		144.0
	S-5-A	0.34	0.33	0.37	0.39	0.38	0.36	0.37	0.34	0.36	0.36	0.36	0.02	5.2	10		177.0
											Overall	0.33	0.03	9.1	45		
$NO_2 + NO_3$-N	DK-A	10.34	10.50	10.42	10.30	10.21						10.35	0.11	1.1	5	7.94	94.1
	FRG-A	11.39	11.53	11.48	11.57	11.62						11.52	0.09	0.8	5		103.4
	USSR-M*	10.28	10.19									10.24	0.06	0.6	2		95.3
	SF-A	10.36	10.30	10.30	10.28	10.28						10.30	0.03	0.3	5		89.4
	S-2-A	11.27	11.26	11.10	11.27	11.19						11.22	0.07	0.7	5		99.3
	S-3-M	11.49	11.46	11.46	11.42	11.39						11.44	0.04	0.3	5		100.8
	S-4-M	10.52	10.78	11.20	11.35	10.65	11.31	11.39	11.22			11.05	0.35	3.1	8		96.6
	S-5-A	(11.13)	11.46	11.89	12.00	11.99	11.75	11.85	11.73	11.75	11.88	11.81	0.17	1.4	9		103.6
											Overall	11.12	0.59	5.3	44		

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5)

TABLE 7. (cont.)

Intercalibration of **spiked sample from 55 m.** Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	SD	CV%	n	Added µmol/l	Recovery %
NH_4-N	DK-A	1.58	1.80	1.76	1.70	1.67						1.70	0.09	5.0	5	0.98	149.4
	FRG-A	1.71	1.67	1.71	1.74	1.60						1.69	0.05	3.2	5		133.5
	USSR-M*	1.76	1.64									1.70	0.09	5.0	2		103.6
	SF-M	1.56	1.66	1.62	1.55	1.53						1.58	0.05	3.4	5		119.0
	S-2-A	1.71	1.71	1.74								1.72	0.02	1.0	3		120.7
	S-3-M	1.63	1.63	1.67	1.62	1.58						1.63	0.03	2.0	5		109.4
	S-4-M	2.54	2.36	2.32	2.41	2.32	2.32	2.40	2.37			2.38	0.07	3.1	8		126.6
	S-5-A																
											Overall	1.84	0.32	17.5	33		
Tot.-N	DK-A															17.7	
	FRG-A	33.97	33.86	34.01	34.00	34.63						34.09	0.31	0.9	5		88.7
	USSR-M*																
	SF-A	37.7	38.7	38.5	38.3	38.1						38.26	0.39	1.0	5		93.3
	S-2-A	35.75	36.24	37.09	36.15	35.94						36.23	0.51	1.4	5		93.9
	S-3-M	35.6	36.2	35.8	36.4	35.5						35.90	0.39	1.1	5		95.8
	S-4-M																
	S-5-A																
											Overall	36.12	1.56	4.3	20		

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5)

TABLE 7. (cont.)

Intercalibration of **spiked sample from 55 m.** Replicate analyses. Results in µmol/l.
Letters after Lab.code indicates procedure: A = automated, M = manual.
Values between brackets not considered in the statistical evaluation.

		1	2	3	4	5	6	7	8	9	10	Mean	SD	CV%	n	Added µmol/l	Recovery %
SiO_4-Si	DK-A															24.5	
	FRG-A	41.69	41.55	42.32	42.37	42.21						42.03	0.38	0.9	5		118.0
	USSR-M*	44.29	44.56									44.43	0.19	0.4	2		101.5
	SF-A	41.0	41.0	41.3	41.2	41.5						41.20	0.21	0.5	5		116.6
	S-2-A																
	S-3-M	40.9	41.0	41.5	41.0	41.0						41.08	0.24	0.6	5		113.1
	S-4-M	36.4	35.6	36.0	36.6	36.3	36.9	36.8	(41.0)			36.37	0.46	1.3	7		97.8
	S-5-A	45.1	45.4	45.5	45.5	45.4	45.4	45.4	45.3	45.1	45.5	45.36	0.15	0.3	10		128.1
											Overall	41.72	3.27	7.8	34		

* not calibrated against own standard, but calibrated against delivered standard (the same standards as were used by lab S-4 and S-5)

Table 8 Overall means, standard deviation and coefficient of variation from the Baltic Intercalibration Workshops in Kiel1977, Rönne 1982 and Visby 1990.

	PO,-P			Tot-P			NO,-N			NO_2+NO_3-N*			NH_3-N			Tot-N**			SiO_4-Si		
	Mean	SD	CV%	Mean	SD	CV%	Mean	SD	CV%	Mean	SD	CV%	Mean	SD	CV%	Mean	SD	CV%	Mean	SD	CV%
Kiel 1977 5‰	0,06	0,03	46	0,15	0,04	26	0,04	0,02	41	0,19	0,11	58	0,38	0,26	68	2,5	1,7	68	0,93	0,21	22
Rönne 1982 Mixed sample	0,14	0,05	32	0,52	0,08	15	0,03	0,02	50	0,17	0,07	39	0,61	0,12	20	21,5	1,2	5	7,2	0,6	9
Visby 1990 Natural 10 m	0,04	0,02	51	0,46	0,11	23	0,03	0,02	82	0,07	0.03	50	0.28	0,27	98	19,5	1.2	6	9,2	0,6	7
Visby 1990 Spiked 10 m	0,23	0,04	17	0,71	0,15	22	0,41	0,04	10	1,30	0,05	4	0,53	0,31	58	23,5	1,6	7	18,0	0,6	7
Kiell977 Surface	0,47	0,03	7	0,99	0,10	10	0,70	0,04	5	6,37	0,22	3	1,01	0,34	34	31,8	3,2	10	9,2	0,6	7
Visby 1990 Natural 55 m	0,68	0,04	6	0,95	0,13	14	0,18	0,02	12	3,32	0,26	8	0,59	0,32	54	19,7	1,3	7	13,5	1,7	12
Visby 1990 Spiked 55 m	2,15	0,07	3	2,43	0,22	9	0,33	0,03	9	11,12	0,59	5	1,84	0,32	18	36,1	1,6	4	41,7	3,3	8

* Kiel and Rönne only NO3-N

** Rönne only two labs participate (n=10+1)

Visby only four labs participated (n=5+5+5+5)

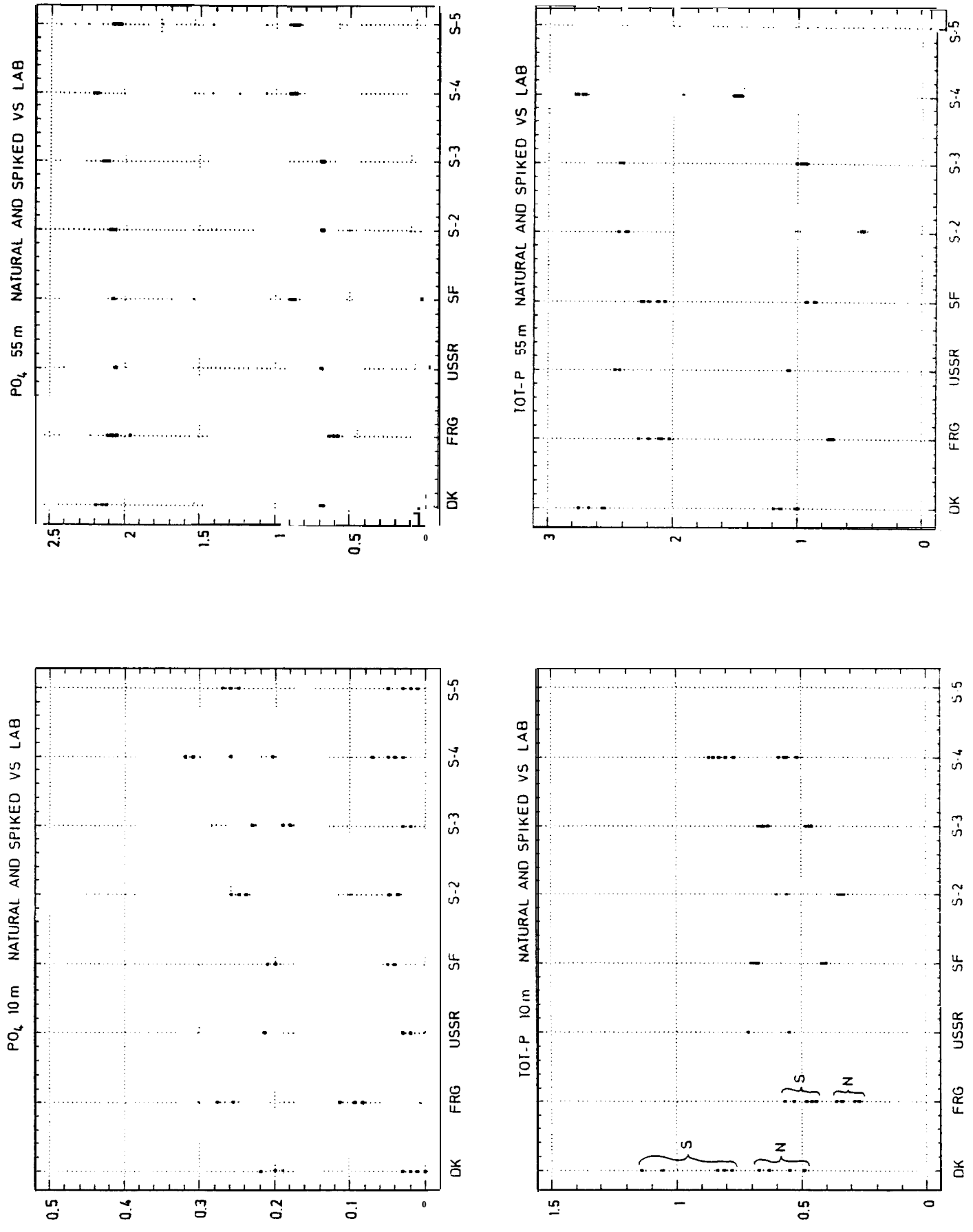

PO4 55 m NATURAL AND SPIKED VS LAB
TOT-P 55 m NATURAL AND SPIKED VS LAB
PO4 10 m NATURAL AND SPIKED VS LAB
TOT-P 10 m NATURAL AND SPIKED VS LAB
DK FRG USSR SF S-2 S-3 S-4 S-5
S
N
S
N

FIG. 1

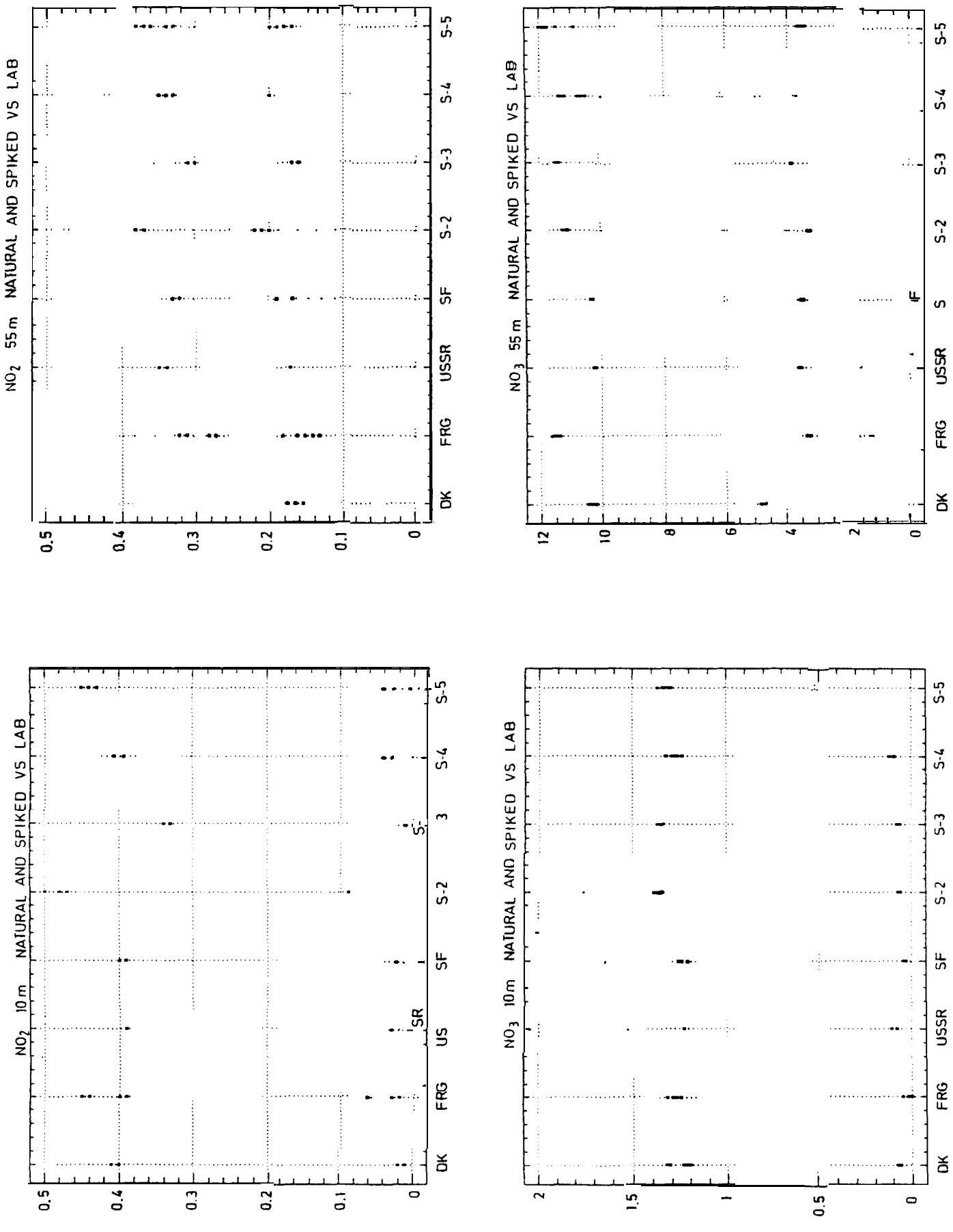

NO2 55m NATURAL AND SPIKED VS LAB
NO3 55m NATURAL AND SPIKED VS LAB
NO2 10m NATURAL AND SPIKED VS LAB
NO3 10m NATURAL AND SPIKED VS LAB
DK FRG USSR SF S-2 S-3 S-4 S-5
DK FRG USSR S S-2 S-3 S-4 S-5
DK FRG US SF S-2 3 S-4 S-5
DK FRG USSR SF S-2 S-3 S-4 S-5

FIG. 2

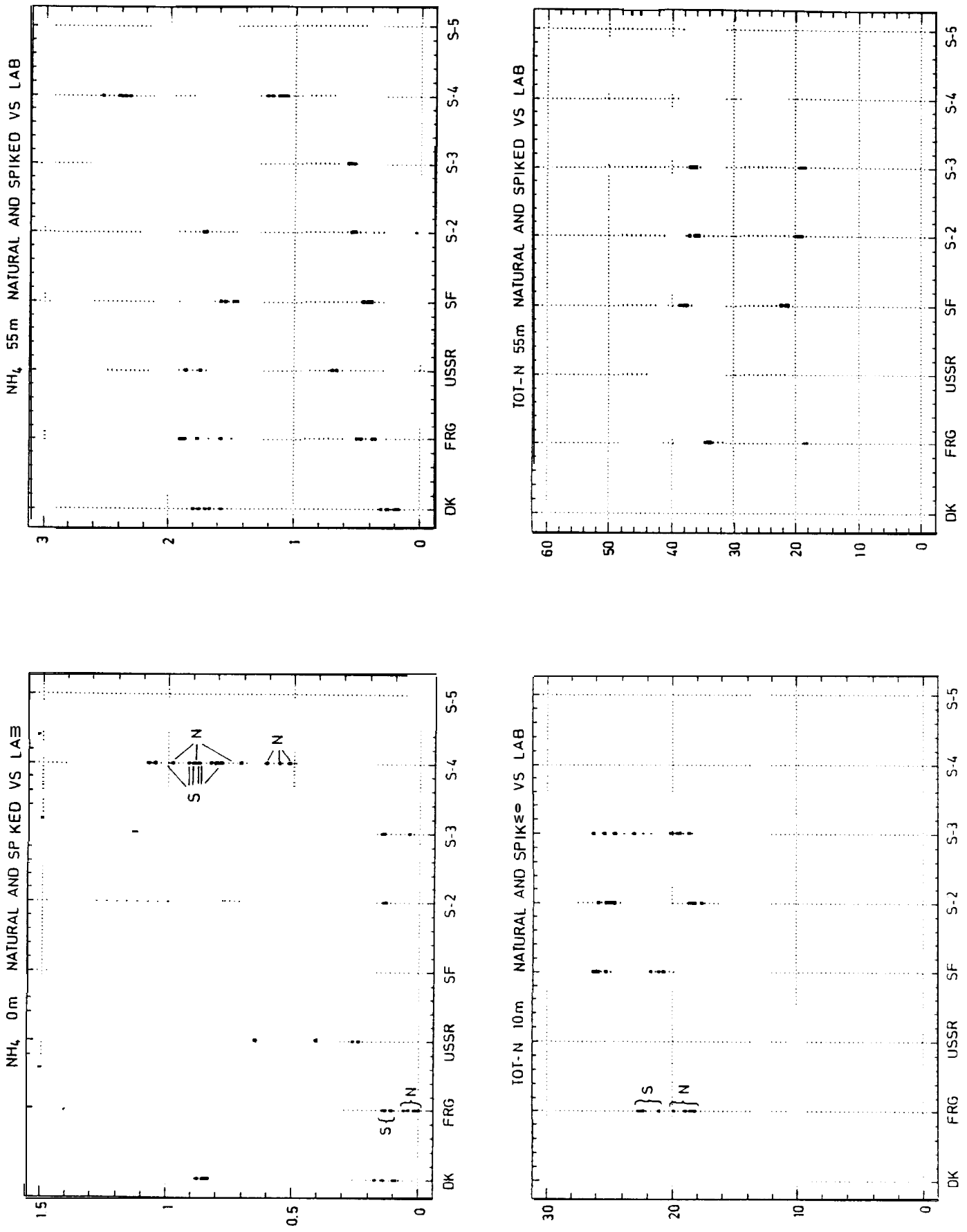
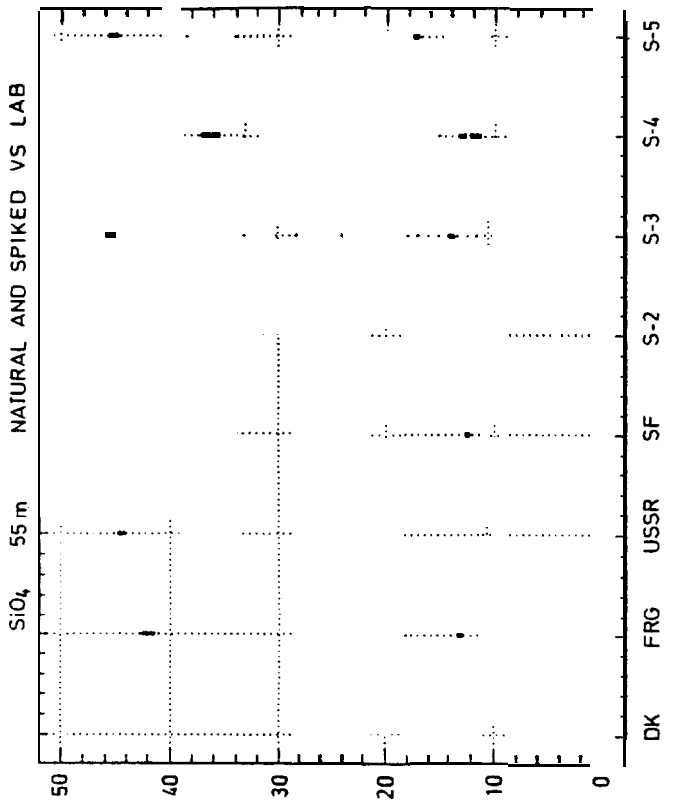

NH4 55m NATURAL AND SPIKED VS LAB
TOT-N 55m NATURAL AND SPIKED VS LAB
NH4 0m NATURAL AND SPIKED VS LAB
TOT-N 10m NATURAL AND SPIKED VS LAB
DK FRG USSR SF S-2 S-3 S-4 S-5
S
N

FIG. 3


SiO_4 55 m NATURAL AND SPIKED VS LAB
50
40
30
20
10
0
DK
FRG
USSR
SF
S-2
S-3
S-4
S-5

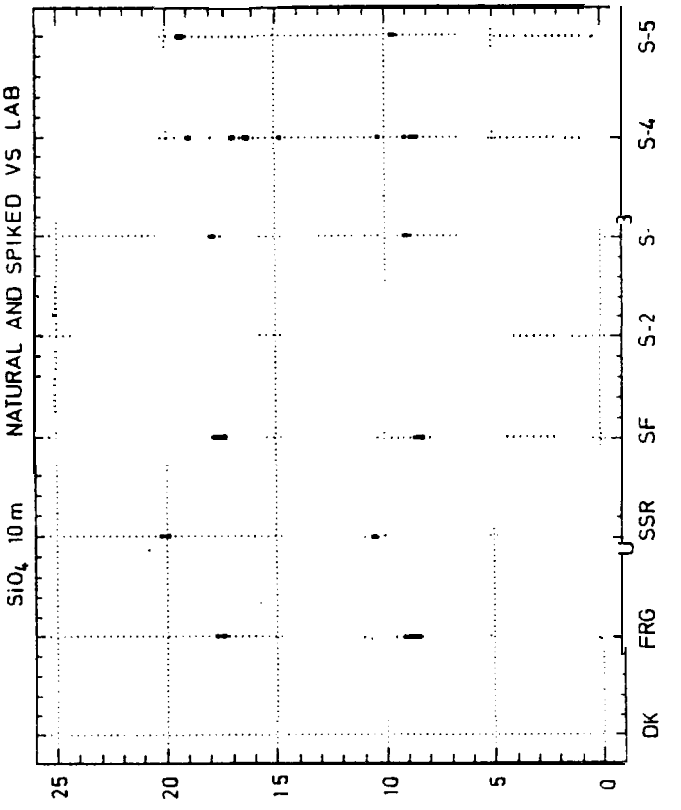

SiO_4 10 m NATURAL AND SPIKED VS LAB
25
20
15
10
5
0
DK
FRG
SSR
SF
S-2
S-4
S-5

FIG. 4

9. GENERAL CONCLUSIONS AND RECOMMENDATIONS

The previous experiences from the Second Intercalibration Workshop in Rønne 1982 guided the efforts of the third Workshop in Visby. It was concluded that the variations obtained by the different laboratories at earlier occasions were to a certain extent due to a patchy distribution of the measured determinands. Mixed samples, where applicable, were consequently distributed among the participating laboratories in order to reduce background disturbances.
It was revealed that for those determinants where the Guidelines prescribe a methodology , the HELCOM Guidelines (HELCOM 1988 A and B) are not followed as exactly as is required by the laboratories which participate in the routine monitoring activities of the BMP. Strict adherence to the Guidelines and participation in intercalibrations by all laboratories reporting data to the BMP is of vital importance in order to make joint assessments of the environmental status of the Baltic Sea. The pelagic and macrozoobenthos sampling stations are shown in Figure 1.

Oxygen

Neither the use of different types of hydrocast bottles nor the handling by different sampling staff influenced the results to any significant extent. The ability to reproduce the measurements within the different laboratories was generally high. However, it seemed to be systematic differences between the analytical procedures in the laboratories, and that much of this reflects differences in the standardization of their thiosulfate solutions. It is recommended that the tests of the variability caused by the sampling staff, and the variability due to the analysis, respectively, should be repeated at the next intercalibration exercise. Furthermore, the replicates in each test ought to be expanded in order to allow for a proper statistical evaluation.

Nutrients

Possible errors in the spiking procedure caused a consistent deviation in the recoveries of both nitrite and silicate.
The results from the intercalibration of inorganic phosphate were acceptable. The low precision and accuracy found for total phosphorous may be due to the digestion methods used. It is recommended that the standards and spiking media of total phosphorous and nitrogen should contain both organic and inorganic components in order to evaluate the digestion methods in relation to the analyses of nitrate and phosphate. It is also recommended to direct special attention towards the methods to measure ammonia, where contamination of the samples may affect the quality of the data considerably.

Chlorophyll-a

The precision within the laboratories was generally high. However, the comparability between the laboratories was relatively low. The best agreement was found between the laboratories delivering data to the HELCOM data bank. The spectrophotometer measurements and the laboratory conditions during filtration and extraction seemed to contribute most to the differences found between the laboratories. Besides, it is recommended to carefully determine the chlorophyll-a absorption peak at intervals, and to use a band-width not exceeding 2 nm.

Primary production

Neither the reproduction of data by the individual laboratories nor the comparability between the laboratories was acceptable. In repeated exercises the rank of the participating laboratories changed, too. The best result was achieved in the experiment which followed the procedure outlined by BMP. This gives hope that the BMP data delivered to the HELCOM data bank are better than those obtained at the intercalibration workshop.

Phytoplankton

The microscopical phytoplankton counting method was found to be acceptable - which is in accordance with results from previous intercalibrations. The critical point in the analysis is the species identification. Therefore the main effort of the Phytoplankton Group was focused on identification problems rather than the technical details in sampling or counting procedures. It was recommended to improve the comparability of the monitoring data by regular yearly meetings of the phytoplankton analysts. The HELCOM data reporting formats should also be revised so that all relevant information could be reported.

Zooplankton

The comparability of the zooplankton countings was very good within the laboratories but was less satisfactory between the laboratories. It should be considered whether all samples from stations visited by different laboratories should be treated by one specific laboratory. Although not significant, there seems to be a wider range between the laboratories using the Folsom-splitter than those using the Kott-splitter. It is recommended that data based on countings of less than 20 animals per taxon should be recorded, but not included in the quantitative analysis. The HELCOM data reporting format should also offer the possibility of including qualitative remarks.

Macrozoobenthos

Accurate determination of the position is of special importance in the sampling of benthos. The ships must pay attention to the repeatability in positioning and should check their Decca navigators frequently. The Guidelines strictly prescribes the use of the standard van Veen grab. In spite of this, some laboratories participating in the regular monitoring use other equipments. If they continue to use other types of devices also in the future, they must be intercalibrated. Generally, the errors in abundance figures caused by sorting and counting seem to be negligible. However, the errors in biomass figures, caused by differences in the weighing procedure, are unacceptably large. This is also a

reflection of the lack of exactness in wet weight determination methodology in the Guidelines, which must be improved. The participating laboratories are urged to report correctly to the HELCOM secretariat, i.e. to use the correct RUBIN codes, the new reporting formats, and to thoroughly check their data before sending them to the secretariat. It is recommended to include the presence of H_2S in the sediment description reporting format. Patchiness of macrozoobenthos can greatly reduce the possibility to distinguish long term trends. It is therefore recommended to carry out patchiness studies on a number of BMP stations. It is also suggested to distribute the sampling responsibility for the BMP stations among the contracting parties. This would minimize errors caused by differences in positioning and methods, that would delay the discovery of trends.

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HELCOM, 1988 B. Guidelines for the Baltic Monitoring Programme for the Third Stage; Part D. Biological Determinands. Balt. Sea Environ. Proc. No. 27 D.

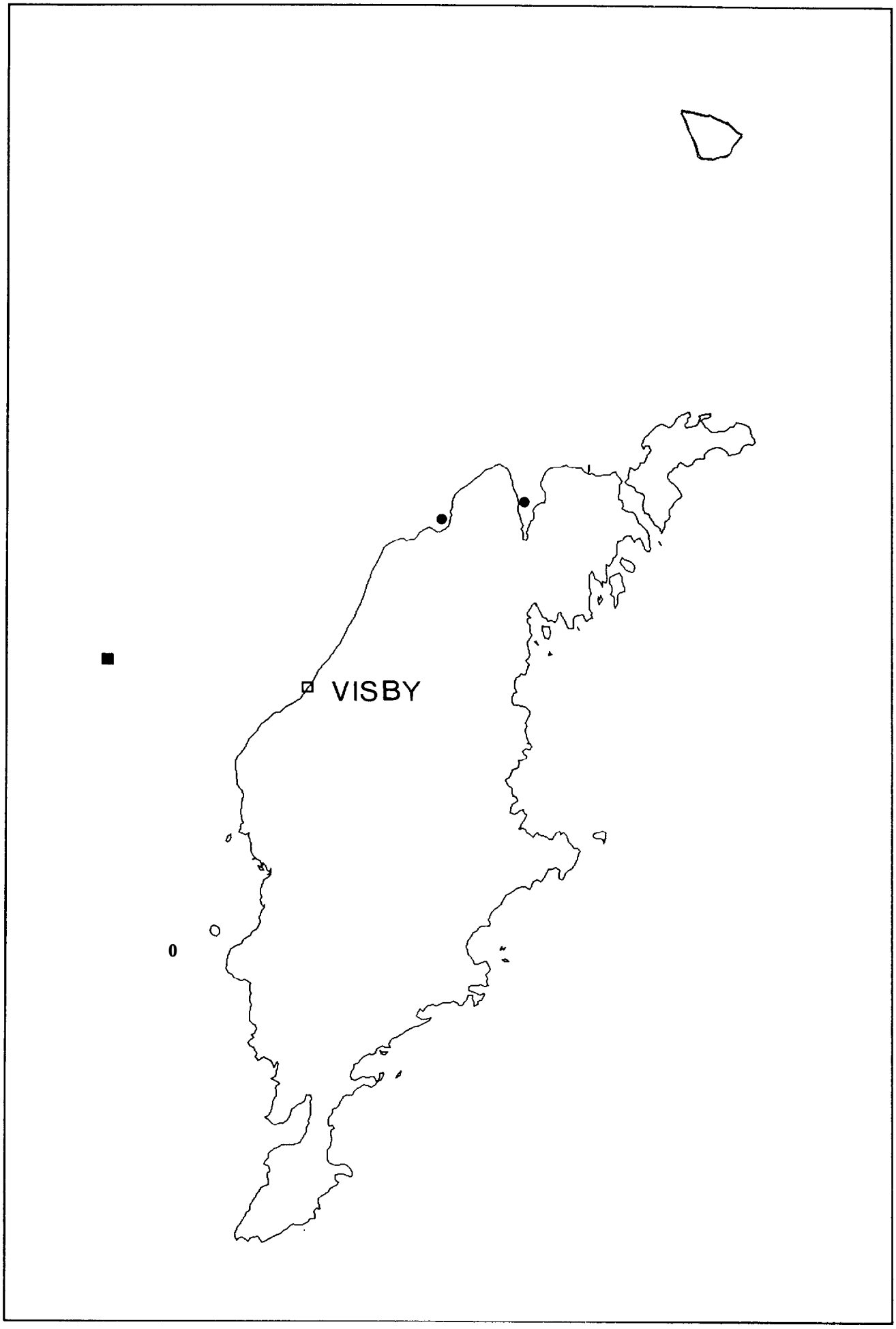

Figure 1. Map of Gotland with the pelagic (■) and macrozoobenthos (●) sampling stations.

BALTIC SEA ENVIRONMENT PROCEEDINGS

No. 1 JOINT ACTIVITIES OF THE BALTIC SEA STATES WITHIN THE FRAMEWORK OF THE CONVENTION ON THE PROTECTION OF THE MARINE ENVIRONMENT OF THE BALTIC SEA AREA 1974-1978
(1979)*)

No. 2 REPORT OF THE INTERIM COMMISSION (IC) TO THE BALTIC MARINE ENVIRONMENT PROTECTION COMMISSION
(1981)

No. 3 ACTIVITIES OF THE COMMISSION 1980
- Report on the activities of the Baltic Marine Environment Protection Commission during 1980
- HELCOM Recommendations passed during 1980
(1981)

No. 4 BALTIC MARINE ENVIRONMENT BIBLIOGRAPHY 1970-1979
(1981)

No. 5A ASSESSMENT OF THE EFFECTS OF POLLUTION ON THE NATURAL RESOURCES OF THE BALTIC SEA, 1980
PART A-l: OVERALL CONCLUSIONS
(1981)*)

No. 5B ASSESSMENT OF THE EFFECTS OF POLLUTION ON THE NATURAL RESOURCES OF THE BALTIC SEA, 1980
PART A-l: OVERALL CONCLUSIONS
PART A-2: SUMMARY OF RESULTS
PART B: SCIENTIFIC MATERIAL
(1981)

No. 6 WORKSHOP ON THE ANALYSIS OF HYDROCARBONS IN SEAWATER
Institut fiir Meereskunde an der Universität Kiel, Department of Marine Chemistry, March 23 - April 3, 1981
(1982)

No. 7 ACTIVITIES OF THE COMMISSION 1981
- Report of the activities of the Baltic Marine Environment Protection Commission during 1981 including the Third Meeting of the Commission held in Helsinki 16-19 February 1982
- HELCOM Recommendations passed during 1981 and 1982
(1982)

No. 8 ACTIVITIES OF THE COMMISSION 1982
- Report of the activities of the Baltic Marine Environment Protection Commission during 1982 including the Fourth Meeting of the Commission held in Helsinki 1-3 February 1983
- HELCOM Recommendations passed during 1982 and 1983
(1983)

No. 9 SECOND BIOLOGICAL INTERCALIBRATION WORKSHOP
Marine Pollution Laboratory and Marine Division of the National Agency of Environmental Protection, Denmark, August 17-20, 1982, Rønne, Denmark
(1983)

No. 10 TEN YEARS AFTER THE SIGNING OF THE HELSINKI CONVENTION
National Statements by the Contracting Parties on the Achievements in Implementing the Goals of the Convention on the Protection of the Marine Environment of the Baltic Sea Area
(1984)

No. 11 STUDIES ON SHIP CASUALTIES IN THE BALTIC SEA 1979-1981
Helsinki University of Technology, Ship Hydrodynamics Laboratory, Otaniemi, Finland
P. Tuovinen, V. Kostilainen and A. Hämäläinen
(1984)

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