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Non-ortho polychlorinated biphenyls (PCBs) in Baltic fish in the 1999-2003 period

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Abstract. The results of determinations of non-ortho PCB contents in Baltic fish and a few fish products are presented. Samples of fillets from herring, sprat, and salmon and from cod liver were examined. A method for determining non-ortho PCB was developed at the SFI Testing Laboratory. It involves purifying samples by dialysis through a semi-permeable polyethylene membrane (SPM) and high-performance liquid chromatography (HPLC). The final determinations were performed with gas capillary chromatography /electron capture detector (GC/ECD).

The contents of four congeners – 77, 81, 126, and 169, which were classified in 1998 by the World Health Organization as dioxin-like compounds, were determined. Based on the results obtained from each sample, the TEQ of non-ortho PCBs was calculated. The highest TEQ (even above 70 pg TEQ-WHO/g sample wet weight) were confirmed in samples of cod liver. In contrast, in other fish samples these values were significantly lower. The TEQ in salmon ranged from 3.5 to 8.4, while those in most of the sprat samples were about 3.0 pg TEQ-WHO/g sample wet weight. The lowest levels of TEQ, ranging from 0.41 to 4.73 pgTEQ-WHO/g sample wet weight, were detected in the herring samples.

Key words: non-ortho polychlorinated biphenyls, persistent organic pollutants, dioxin-like compounds, Baltic fish, method validation

INTRODUCTION

Polychlorinated biphenyls (PCBs) are a group of 209 individual chlorinated congeners that differ in the location and number of chlorine atoms in the molecule. These differences affect their degree of toxicity. PCBs, along with polychlorinated dibenzo-*p*-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs), comprise a large group of persistent organic pollutants (POPs) that poses a serious threat to the environment (Falandysz 1999, Falandysz 1996, Anon. 2000A, Hites *et al.* 2004). As a result of their lipophilic nature, they accumulate in the subsequent links of the food chain creating a potential route of human exposure (Tam 1999, Obiedziński *et al.* 2000, Dudzińska *et al*. 2001, Anon. 2000b). Taking into account the toxicity of these compounds, the monitoring of their concentrations in food seems to be of primary importance.

During a meeting of the International Council for the Exploration of the Sea (ICES), seven congeners (PCB: 28, 52, 101, 118, 138, 153, 180) were designated for obligatory determination in environmental monitoring studies (Anon. 1988, Anon. 1997, Falandysz 1999).

However, in the 1980s and 1990s intense analytic and toxicological research was conducted on other special groups of PCBs which have not yet been included on this list. This research led to the designation in 1998 by the World Health Organization (WHO) of a group of dioxin-like PCB compounds that should be analyzed together with dioxins PCDDs and furans PCDFs in order to describe the total toxicity of samples, the so-called Toxic Equivalency (TEQ) (Atuma *et al.* 1998, Birnbaum *et al.* 1995, Ahlborg *et al.* 1994, Maged Younes 2000). The dioxin-like PCBs included (Ahlborg *et al.* 1994) four coplanar, non-ortho-PCBs (77, 81, 126 169), and eight mono-ortho-PCBs (123, 118, 114, 105, 167, 156, 157, 189) with nearly planar molecular structures. The experimental toxic equivalent factor (TEF) of each one relative to the most toxic 2,3,7,8-tetrachlorodibenzodioxin (2,3,7,8-TCDD) was determined. These coefficients are used to calculate the TEQ of samples (Birnbaum *et al.* 1995) which are indicators of the degree of samples contamination.

In Council Regulation (EC) No. 2375/2001(Anon. 2001) not only were the permissible levels of PCDDFs for various food groups stipulated, it was also announced that limits for dioxin-like PCBs would be set (Anon. 2004a). This requires European Union countries to monitor these substances, particularly in foodstuffs since the current database is too small and incomplete. Although there are relatively large amounts of data regarding the levels of PCDDs/Fs in environmental samples, research on the levels of non-ortho PCBs is still rare in Poland as well as in other European Union countries. To some extent this is due to the fact that non-ortho PCBs occur in food samples at several orders of magnitude lower concentrations than ICES indicator PCBs, for example, and therefore they are complicated to analyze. Since Poland does not have any institutions with laboratories accredited to determine dioxin-like PCBs in foodstuffs, the accredited Testing Laboratory of the SFI undertook a project to develop a method of determining non-ortho PCB in fish and fish products.

The aim of our work was:

– to adapt, based on the available literature, and validate a simple and inexpensive method for determining non-ortho PCBs in fish and fish products that would be possible to perform at the SFI Testing Laboratory;

– to determine levels of non-ortho-PCBs in selected fish and fish products, to calculate the TEQ of the tested samples and to compare these results with existing literature data from other regions.

MATERIALS AND METHODS

The following materials were used for method quality control and validation:

– homogenized cod liver oil from fish caught in the Gdańsk Deep. The livers (from 3 specimens) came from cod measuring from 38 to 40 cm. This oil was used to determine repeatability and reproducibility of the method;

– laboratory control standard for the recovery determination was prepared by spiking 7 g of PCBs free corn oil (Mazola) with 1 ml of PCBs standard solution in which the concentration of each congener was 0.001 μg/ml ,

– IAEA 140 certified reference material (Halogenated Hydrocarbons in Sea Plant Homogenate from the International Atomic Energy Agency, Marine Environmental Laboratory, Analytical Quality Control Services).

The following materials were used to determine non-ortho PCBs:

– the individual crystalline pure non-ortho-PCBs used to prepare the solution were obtained from Promochem; standard solutions of individual non-ortho PCBs were prepared by dissolving 20 mg of each solid compound in 100 ml of hexane in a volumetric flask; subsequently these solutions were used to prepare the standard mixture;

– the samples of Baltic herring, sprat, and cod used to determine coplanar PCBs were collected during cruises of the r/v Baltica during the 1999-2003 period. Salmon samples were purchased from fishermen. The following fish species were tested: herring *(Clupea harengus*); sprats (*Sprattus sprattus*), Baltic salmon (*Salmo salar*), and cod (*Gadus morhua*) liver oil. The catches were conducted in Baltic fishing grounds (Fig. 1); the characteristics of the samples are presented in Tables 1 and 2.

Each pooled sample of herring was comprised of 15-20 specimens and those of sprat of approximately 30 specimens. The salmon sample for testing was comprised of one specimen weighing from 3.5-6.0 kg. Each cod liver oil sample was obtained from the livers of two to three cod specimens. The herring and salmon samples analyzed were skinned fillets, while those of sprat were with skin.

The samples caught during cruises were frozen and stored at a temperature of -18°C until the analyses were conducted. The samples of Baltic fish from 1999 and 2000 were analyzed in the SFI Testing Laboratory according to the adapted method. The final determinations were performed with gas capillary chromatography/electron capture detector (GC/ECD).

To confirm that the method proposed by the SFI is reliable, samples from the 2002-2003 period were analyzed by a joint Polish-Norwegian effort.

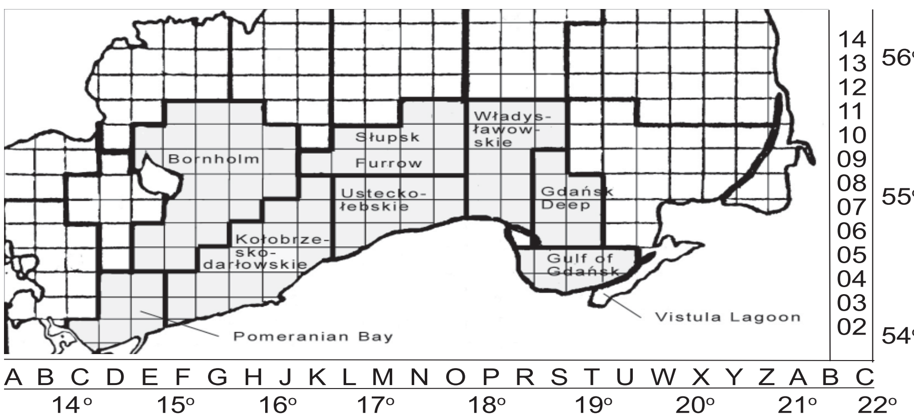

Fig. 1. Baltic fishing grounds where the samples were caught

Table 1. Characteristics of the samples analyzed in the SFI Testing Laboratory in 1999-2000

Sample code	Sample type	Date caught	Fishing ground	Length range of specimens [cm]	Dry weight [%]	Fat [%]
8S	herring	9.10.1999	Słupsk Furrow	21.5 – 22.0	27.84	9.01
5S		10.10.1999	Słupsk Furrow	20.5 – 21.0	26.53	7.29
17S		17.10.1999	Słupsk Furrow	19.0 - 019.5	27.18	7.69
44S		6.11.1999	Słupsk Furrow	20.5 – 21.0	27.55	8.35
103S		12.06.2000	Gdańsk Deep	19.0 – 20.0	23.63	5.42
104S		12.06.2000	Gdańsk Deep	25.0 – 26.0	25.07	6.65
180S		17.10.2000	Słupsk Furrow	23.0 – 24.0	26.34	7.38
130Sdw	cod liver	05.03.2000	Gulf of Gdańsk	70.0	n.d *	44.09
135dw		05.03.2000	Gulf of Gdańsk	110	n.d.	22.72
133Sdw		11.03.2000	Gdańsk Deep	86	n.d.	31.05
160Sdw		04.10.2000	Gdańsk Deep	41.0 – 42.0	n.d.	33.12
120PM	fish livers à la Caucasienne				n.d.	38.52
123PM	canned fish				n.d.	49.26
129PM	smoked				36.25	31.57

n.d. * – not determined

Homogenized samples of fish muscle tissue were prepared at the SFI. Then they were frozen and freeze-dried. Each such freeze-dried sample was divided into two parts. One was sent to NIFES for determinations of non-ortho PCBs according to the accredited methods of that laboratory. The remaining part of the sample was processed at the SFI. Extracts obtained at the SFI were also sent to NIFES so that final determinations could be performed with high-resolution gas chromatography with a high resolution mass detection (HRGC/HRMS) according to current stipulations of the Commission Directive 2002/69/EC of 26.07.2002 (Anon.2002). The comparison of the results obtained for samples processed at the NIFES and at the SFI would show if the method developed at the SFI is reliable.

Method applied at the SFI

The analytical protocol developed in our laboratory (Barska *et al.* 2002) is presented in brief below:

Sample preparation
The fish samples were filleted and homogenized. Homogenates were placed in Petri dishes and frozen. The samples were freeze-dried in order to dehydrate them. They were then granulated in an electric grinder.

Extraction
About 20 g of freeze-dried sample was extracted with hexane in a Soxtec-Avanti 2050 apparatus at 2.5 hours of extraction in boiling solvent and 4.5 hours in cold solvent.

Table 2. Characteristics of samples of sprat, herring, salmon, and cod livers analyzed in the 2002-2003 period as part of Polish-Norwegian cooperation

Sample code	Sample type	Date caught	Fishing ground	Length range of specimens [cm]	Dry weight [%]	Fat [%]
72S	sprat	18.02.02	Gulf of Gdańsk	11.0 – 12.0	28.86	12.15
89S		22.02.02	Gulf of Gdańsk	11.0 – 12.0	28.42	11.28
112S		23.02.02	Gdańsk Deep	12.0 – 12.5	30.70	11.53
109S		24.02.02	Gdańsk Deep	12.5 – 13.0	28.59	11.26
116S		01.03.02	Gdańsk Deep	12.0 – 13.0	28.61	11.13
95S		02.03.02	Słupsk Furrow	12.0 – 13.0	27.50	10.86
113S		02.03.02	Słupsk Furrow	12.0 – 13.0	27.68	7.69
98S		02.03.02	Słupsk Furrow	11.0 – 12.0	27.69	8.35
94S		03.03.02	Władysławowskie	13.5 – 14.0	27.79	9.17
107S		21.03.02	Gdańsk Deep	12.0 – 13.0	25.80	8.46
66S		26.03.02	Gulf of Gdańsk	8.0 – 12.0	26.58	10.20
67S		04.04.02	Gulf of Gdańsk	9.0 – 12.0	24.07	7.23
92S		13.04.02	Koł.-darłowskie	12.0 – 13.0	23.71	6.97
80S		16.04.02	Ustecko-łebskie	13.5 – 14.0	24.64	7.49
81S		16.04.02	Ustecko-łebskie	10.0 – 11.0	24.07	7.27
114S		08.05.02	Władysławowskie	13.0 – 13.5	22.99	5.31
102S		20.10.02	Bornholm	13.0 – 14.5	31.67	13.40
101S		21.10.02	Koł.-darłowskie	13.0 – 14.0	32.64	14.22
108S		23.10.02	Słupsk Furrow	13.0 – 14.5	31.57	13.32
971S		12.03.03	Gdańsk Deep	12.0 – 13.0	26.59	9.47
96S		04.06.03	Gulf of Gdańsk	12.0 – 13.0	21.74	3.78
99S		04.06.03	Koł.-darłowskie	13.0 – 14.0	24.77	6.15
100S	herring	19.02.02	Gulf of Gdańsk	22.0 – 23.0	23.36	4.75
97S		20.02.02	Gulf of Gdańsk	22.0 – 23.0	23.22	3.69
117S		20.02.02	Gulf of Gdańsk	21.0 – 22.0	24.09	4.48
71S		22.02.02	Gulf of Gdańsk	21.0 – 22.0	23.63	3.15
91S		22.02.02	Gulf of Gdańsk	22.0 – 23.0	22.60	3.54
111S		23.02.02	Gdańsk Deep	21.0 – 22.0	22.75	3.52
118S		01.03.02	Gdańsk Deep	19.0 – 20.0	23.66	2.71
70S		02.03.02	Słupsk Furrow	21.0 – 22.0	23.28	3.68
991S		02.03.02	Słupsk Furrow	23.0 – 29.0	24.26	4.72
961S		02.03.02	Słupsk Furrow	21.0 – 22.0	23.14	4.45
69S		03.03.02	Władysławowskie	23.0 – 24.0	21.91	2.54
110S		03.03.02	Władysławowskie	21.0 – 22.0	21.68	2.51
119S		21.03.02	Gdańsk Deep	20.0 – 21.0	20.85	2.28
65S		26.03.02	Gulf of Gdańsk	22.0 – 24.0	21.32	2.86
123S		26.03.02	Gulf of Pomerania	20.0 – 21.0	22.57	2.64
124S		26.03.02	Gulf of Pomerania	20.0 – 21.0	22.57	2.64
122S		25.05.02	Gulf of Pomerania	20.0 – 21.0	23.13	3.25
98S		11.03.03	Gdańsk Deep	18.0 – 19.0	21.85	2.52
95S		24.04.03	Gulf of Gdańsk	19.0 – 20.0	19.84	1.26
1001S		14.06.03	Władysławowskie	20.0 – 21.0	19.57	3.64
104S		01.07.03	Władysławowskie	18.0 – 19.0	21.80	3.89
103S		23.10.02	Słupsk Furrow	18.0 – 19.0	25.37	5.44
130S	salmon	29.09.03	Władysławowskie	59.0	26.45	6.40
129S		29.09.03		76.0	24.91	2.08
133S		06.10.03		83.0	29.18	8.99
134S		06.10.03		68.0	32.35	11.67
135S		06.10.03		57.0	29.45	7.87
136S		14.10.03		54.0	26.10	5.43
137S		14.10.03		57.0	26.85	5.66
143S		20.10.03		78.0	24.89	2.88
144S		20.10.03		71.0	22.56	1.70
122Sdw	cod liver	19.02.03	Ustecko-łebskie	56.0	n.d. *	30.86
112Sdw		22.02.03	Koł.-darłowskie	46.0	n.d.	48.20
116Sdw		26.02.03	Gdańsk Deep	55.0	n.d.	74.80

n.d.* not determined

Following extraction, the solvent was removed with a vacuum evaporator. After hexane evaporation the samples were weighed and the fat content of each sample was determined. Prior to the extraction of samples from which extracts were to be analysed with HRGC/HRMS, the samples were spiked with 1 ml isotope labeled mixture of PCDDs/Fs and dioxin-like PCBs standards, according to EPA-1613 and EPA-1668 standards (Anon. 1994, Anon. 1998).

Dialysis

The separation of lipids was achieved by dialysis with semipermeable polyethylene membranes (length – 30 cm, width – 26 mm, pore size – 10A, Exposmeter AB, Sweden), which, prior to the process were kept in hexane for approximately two weeks with solvent changes every two days. 2-5 g of fat were dissolved in hexane (1 ml of hexane for each gram of fat) and transferred into the interior of the membrane. The membrane with the fat solution was placed in 250 ml glass cylinder containing approximately 100-130 ml of solvent mixture (hexane : methyl chloride at a ratio of 6 : 4, Strandberg *et al.* 1998, Grochowalski 2000). Dialysis was conducted in darkness (the determined compounds are photolabile) for four days with changes of extraction solvents every 24 hours. The combined dialysis extracts were evaporated using a vacuum evaporator to a volume of 10 ml and the remaining fat was removed by treatment with a mixture of concentrated sulfuric acid and 30% oleum at a ratio of 1 : 1 (Grochowalski *et al.* 2000, Muccio 1993, Falandysz *et. al.* 1999).

Purification of extracts with high performance liquid chromatography (HPLC)

HPLC system consisted of a precolumn and a 25 cm Cosmosil 5 PYE column, which is filled with 2-(1-pyrenyl)ethyl-dimethylsilylated silica gel of a granule size of 5 µm, pump (L-7100), an autosampler (L-7200, Merck-Hitachi), and an injector fitted with a 100 µl loop. The fractions were monitored with Lachrome L-7485 detector at 254 nm. The mobile phase was hexane at a flow rate of 1ml/min. The working temperature in the column was 10°C.

The volume of the sample was up to 500 µl, and it was injected automatically 5-6 times at 60 µl into the HPLC system. Prior to each fractionation the system was equilibrated within 30 minutes. The appropriate fraction was collected by hand. The retention time windows were established on the basis of the analysis of the standard mixture of non-ortho PCBs and were from 9 to 20 minutes. A blank sample was analyzed for each series of five samples.

Final determinations with gas chromatography/electron capture detector (GC/ECD)

Conditions of chromatographic determination:

Rtx-5 column 60 m in length, precolumn 1.5m in length, internal diameter 0.25 mm, thickness of the stationary phase film 0.1 µm.

Carrier gas – helium at a constant pressure of 160 kPa, make up gas for the ECD – nitrogen.

Program temperature: 55°C hold for 0.5 min, temperature increase of 25°C/min to 180°C, increase of 3°C/min to 280°C and 280°C hold for 10 minutes.

The injector and the detector were set at 250°C and 310°C, respectively.

Volume of the injected samples – 2 µl; splitless injection.

The identification of PCBs was based on the agreement between the retention times of peaks in the sample chromatogram with the retention time windows established through the analysis of the standard solutions. Quantification was carried out on the basis of area of standard peaks.

Measurements were taken within the linear range of the detector. The determination limits were approximately1.5 pg/g.

Each series of chromatographic determinations was preceded by analyzing a non-ortho PCB standard solutions in a concentration range of 0.0025-0.01 µg/ml.

Method applied at NIFES

The method applied at the NIFES laboratory, which is accredited for this type of analysis, included the following stages:

– prior to extraction freeze-dried samples were spiked according to EPA-1613 and EPA-1668 standards (Anon. 1994, Anon. 1998) with 1 ml of isotope labeled standard mixture of PCDDs/Fs and dioxin-like PCBs and mixed with Hydromatrix for further drying and to increase the effectivity of the extraction;

– the samples were extracted under high pressure in an ASE 300™ extractor (Dionex), the extraction parameters were 125°C and 1500 PSI;

– the solvent was evaporated in a stream of nitrogen in a Turbovap II™ (Zymark);

– the solution was defatted (max. 3 g fat) and clean-up was done with the automated Power-Prep System™ (Fluid Management system). This device has four columns with the following sequence of adsorbents: I – sulfuric acid set on silicon oxide; II – three layers of silicon oxide – basic, neutral, and acidic; III – basic aluminum oxide; IV – activated carbon;

– the addition of a syringe standard solution prior to the final analysis;

– the final determinations were done with high resolution gas chromatography with mass-spectrometry detection in a MAT 95XL HRGC/HRMS (Thermo Finnigan) tuned to a resolution of 10,000 (max. 10% valley), SIM acquisition. DB-5MS column – 30m length, 0.25 mm inner diameter, 0.25 µm phase thickness.

RESULTS

Validation of the developed method

The method proposed by the SFI Testing Laboratory was subjected to validation stages selected from the literature: recovery, accuracy, repeatability, reproducibility.

Method recovery was determined by five parallel analyses of corn oil spiked with standard mixtures of non-ortho-, mono-ortho-, and seven ICES indicator PCBs. The con-

Table 3. Content of some PCBs in IAEA 140 certified reference material – accuracy

PCB congener	Certified content	Confidence intervals	Determined content *
	[ng/g wet weight]		
PCB 28	1.7	1.0 – 2.5	1.4
PCB 52	3.8	2.6 – 4.9	2.3
PCB 101	2.4	1.9 – 3.2	1.9
PCB 118	1.0	0.97 – 1.05	0.9
PCB 153	1.7	1.2 – 2.6	1.0
PCB 138	1.7	1.25 – 3.3	0.9
PCB 77	0.19	0.035 – 6.86	0.12

*average value from three simultaneous determination

Table 4. Content of non-ortho PCBs in cod liver oil determined with the method developed at the SFI Testing Laboratory (GC/ECD) – method repeatability

PCB	Content of non-ortho PCBs in cod liver oil [ng/g oil]							
	1	2	3	4	5	X	Sd	RSD [%]
77	0.30	0.42	0.44	0.47	0.40	0.41	0.0063	16
126	0.32	0.36	0.43	0.45	0.40	0.39	0.053	14
169	0.087	0.13	0.073	0.029	0.041	0.072	0.041	56

Table 5. Comparison of SFI (GC/ECD) and Cracow University of Technology (GC/MS) results

Congener	SFI	Cracow Technical University	Average X	Standard deviation Sd	RSD
	[ng/g oil]				[%]
77	0.41	0.96	0.69	0.389	56.8
126	0.39	0.34	0.37	0.035	9.69
169	0.072	0.07	0.071	0.0014	7.1

centration of particular compounds in the spiked oil were about 0.000143 μg/ml, which are concentration comparable with those in natural samples.

The recovery of PCB 77 was approximately 80%, PCB 126 – 87%, PCB 169 – 78%. The remaining PCBs were in the range of 63-90%.

The accuracy of the method was evaluated on the basis of three parallel analyses of IAEA 140 certified reference material. The results of determinations are presented in Table 3.

The repeatability of the method was assessed on the basis of five simultaneous determinations of non-ortho PCBs (PCB: 77, 126, 169) in homogenized cod liver oil. The results are presented in Table 4. The related standard deviation of PCB 77 and 126 was 16 and 14%, respectively, while it was higher for PCB 169 at 56%. All of these values are acceptable. The relatively high standard deviation for PCB 169 can be due to the very low concentrations of this compound.

The reproducibility of the method was confirmed by comparing the results obtained for the same sample of homogenized cod liver oil at the SFI Testing Laboratory and the laboratory of the Institute of Inorganic Chemistry and Technology, Cracow University of Technology. The results of the comparison are presented in Table 5. The value of 56%

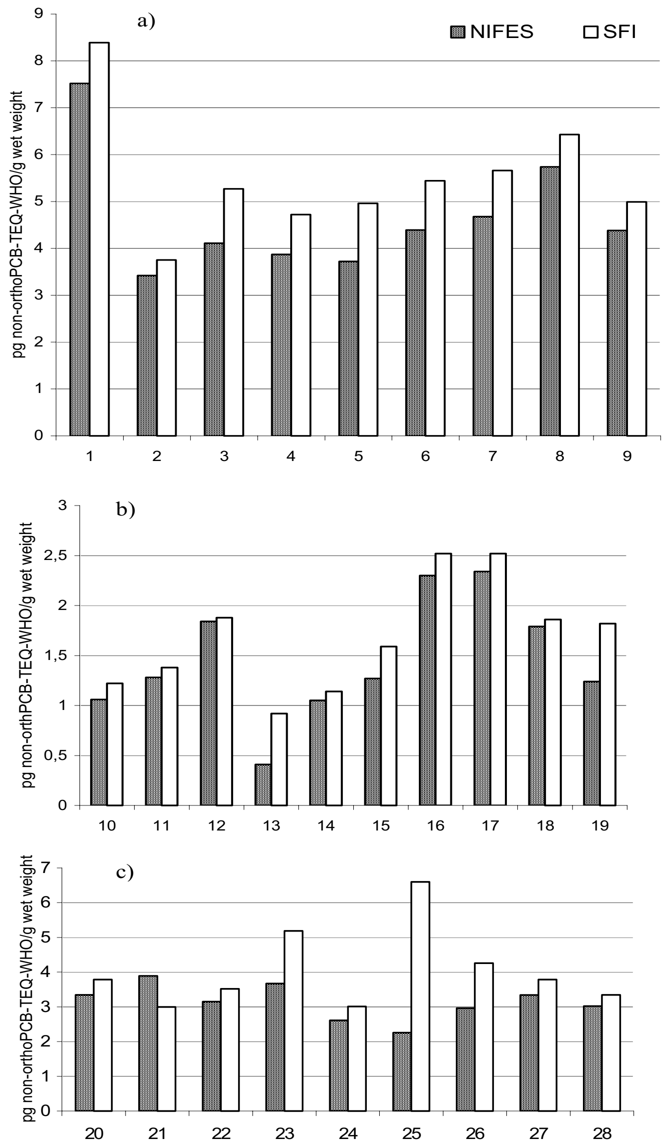

Fig. 2. TEQ calculated in extracts prepared at the SFI and NIFES; final determination performed with HRGC/HRMS in Bergen; a) salmon samples b) herring samples c) sprat samples

obtained for PCB 77 is acceptable since, according to the Horwitz equation (Wood 1996), RSD up to 64% is permitted for analyt concentrations of 10^{-12}. For further confirmation that the sample preparation method for non-ortho PCB determination proposed by the SFI is suitable, the laboratory at NIFES conducted simultaneous analyses of the extracts prepared at the SFI and those prepared in NIFES. The TEQ values presented in Figure 2, calculated on the basis of the contents of non-ortho PCBs determined in extracts prepared at the SFI and NIFES; 90% of the samples were consistent. This indicates that the two methods of preparing samples lead to a similar result in the assessment of the health quality of the tested samples.

Results of determinations of non-ortho PCB content in Baltic fish

The TEQ value was calculated for each sample. Only congeners 77, 126, and 169 were used for TEQ calculations. Congener 81 occurred only in very small concentrations, and since its TEF was 0.0001, its contribution to TEQ is so low that it can be disregarded. Due to this, it is not considered in many publications on non-ortho PCB. The data from Tables 6 and 7 indicate that herring samples had the lowest values of TEQ from non-ortho PCB, expressed as pg TEQ-WHO/g sample wet weight. The range of this value in the 29

Table. 6. Contents of non-ortho PCBs in samples of raw fish and fish products determined at the SFI Testing Laboratory – final determinations were performed with (GC/ECD)

Sample code	Contents of non-ortho PCB [pg/g wet weight]			Total TEQ toxicity content in samples [pgWHO-non-ortho PCB-TEQ/g wet weight]
	CB 77	CB 126	CB 169	
Herring				
8S	11 (0.0011)	20 (2.0)	5 (0.05)	2.05
5S	28 (0.0028)	35 (3.5)	9 (0.09)	3.59
17S	15 (0.0015)	47 (4.7)	2 (0.02)	4.72
44S	49 (0.0049)	32 (3.2)	7 (0.07)	3.27
103S	19 (0.0019)	22 (2.2)	not detected	2.20
104S	48 (0.0048)	16 (1.6)	35 (0.35	1.95
180S	33 (0.0033)	30 (3.0)	3 (0.03)	3.03
Cod liver				
130Sdw	114 (0.0114)	358 (35.8)	58 (0.58)	36.39
135Sdw	57 (0.0057)	239 (23.9)	25 (0.25)	24.16
133Sdw	234 (0.0234)	562 (56.2)	133 (1.33)	57.55
160Sdw	103 (0.0103)	163 (16.3)	13 (0.13)	16.44
Canned fish livers				
120PM	326 (0.0326)	400 (40.0)	16 (0.16)	40.19
123PM	168 (0.0168)	315 (31.5)	12 (0.12)	31.63
Smoked mackerel				
129PM	15 (0.0015)	14 (1.4)	4 m(0.04)	1.44

Figures in brackets stand for the TEQ values

TEF values for CB 77 – 0.0001; CB 126 – 0.1; CB 169 – 0.01

Table 7. Contents of non-ortho PCB in sprat, herring, salmon, and cod liver oil determined at NIFES according to the accredited method of that laboratory

Sample code	Content of non-ortho PCB [pg/g wet weight]				Total TEQ toxicity content in samples [pgWHO-non-ortho PCB-TEQ/g wet]
	CB 77	CB 81	CB 126	CB 169	
1	2	3	4	5	6
Sprat					
72S	86.93 (0.0087)	3.44 (0.0003)	25.93 (2.5932)	3.93 (0.0393)	2.64
89S	68.35 (0.0068)	1.55 (0.0002)	19.59 (1.9597)	2.90 (0.0290)	1.99
112S	78.52 (0.0079)	1.77 0.0002)	23.00 (2.2995)	3.31 (0.0331)	2.34
109S	71.02 (0.0071)	1.63 (0.0002)	21.41 (2.1406)	3.02 0.0302)	2.18
116S	72.78 (0.0073)	1.64 (0.0002)	23.04 (2.3042)	4.03 (0.0403)	2.35
95S	66.48 (0.0066)	1.34 (0.0001)	21.26 (2.1261)	3.09 (0.0309)	2.16
113S	57.50 (0.0058)	1.10 (0.0001)	22.34 (2.2340)	3.89 (0.0389)	2.28
98S	80.49 (0.0080)	1.71 (0.0002)	30.77 (3.0767)	6.04 (0.0604)	3.15
94S	81.76 (0.0082)	1.96 (0.0002)	27.76 (2.7761)	4.01 0.0401)	2.82
107S	102.56 (0.0103)	2.15 (0.0002)	35.96 (3.5964)	6.71 (0.0671)	3.67
66S	71.08 (0.0071)	1.74 (0.0002)	23.19 (2.3187)	3.60 (0.0360)	2.36
67S	75.96 (0.0076)	1.86 (0.0002)	23.65 (2.3648)	15.26 0.1526	2.53
92S	80.52 (0.0081)	1.72 (0.0002)	28.81 (2.8812)	4.77 (0.0477)	2.94
80S	111.61 (0.0112)	3.06 (0.0003)	38.08 (3.8080)	7.01 (0.0701)	3.89
81S	94.22 (0.0094)	1.78 (0.0002)	32.74 (3.2735)	6.32 (0.0632)	3.35
114S	64.33 (0.0064)	1.14 (0.0001)	23.76 (2.3762)	4.03 (0.0403)	2.42
102S	71.44 (0.0071)	1.46 (0.0001)	24.34 (2.4339)	4.99 0.0499)	2.49
101S	69.69 (0.0070)	1.35 (0.0001)	22.08 (2.2079)	4.35 (0.0435)	2.26
108S	74.99 (0.0075)	1.45 (0.0001)	25.49 (2.5490)	5.16 (0.0516)	2.61
971S	113.41 (0.0113)	2.49 (0.0002)	32.77 (3.2767)	5.20 (0.0520)	3.34
96S	81.33 (0.0081)	1.24 (0.0001)	29.56 (2.9559)	5.50 (0.0550)	3.02
99S	84.40 (0.0084)	1.68 (0.0002)	28.90 (2.8902)	5.90 (0.0590)	2.96

table 7, continued

1	2	3	4	5	6
Herring					
100S	43.07 (0.0043)	1.10 (0.0001)	22.70 (2.2696)	6.61 (0.0661)	2.34
97S	32.95 (0.0033)	1.01 (0.0001)	18.37 (1.8367)	4.66 (0.0466)	1.89
117S	26.57 (0.0027)	0.67 (0.0001)	14.20 (1.4203)	3.20 (0.0320)	1.46
71S	27.83 (0.0028)	2.19 (0.0002)	16.00 (1.5995)	3.32 (0.0332)	1.64
91S	28.08 (0.0028)	0.97 (0.0001)	15.84 (1.5840)	4.14 (0.0414)	1.63
111S	28.63 (0.0029)	0.98 (0.0001)	17.32 (1.7322)	4.18 (0.0418)	1.78
118S	26.85 (0.0027)	0.83 (0.0001)	17.30 (1.7303)	5.37 (0.0537)	1.79
70S	28.75 (0.0029)	2.26 (0.0002)	20.92 (2.0917)	5.98 (0.0598)	2.15
991S	32.38 (0.0032)	1.00 (0.0001)	22.03 (2.2027)	9.30 0.0930)	2.30
961S	33.77 (0.0034)	0.96 (0.0001)	21.49 (2.1487)	6.12 (0.0312)	2.21
69S	23.53 (0.0024)	2.00 (0.0002)	13.33 (1.3334)	3.53 (0.0353)	1.37
110S	21.57 (0.0022)	0.72 (0.0001)	13.38 (1.3380)	4.03 (0.0403)	1.38
119S	21.53 (0.0022)	0.70 (0.0001)	14.25 (1.4249)	4.29 (0.0429)	1.47
65S	35.79 (0.0036)	2.04 (2.04)	21.96 (2.1963)	4.93 (0.0493)	2.25
123S	25.87 (0.0026)	0.70 (0.0001)	12.13 (1.2131)	2.87 (0.0287)	1.24
124S	24.81 (0.0025)	0.68 (0.0001)	12.08 (1.2083)	3.35 (0.0335)	1.24
122S	14.79 (0.0015)	0.45 (0.0000)	10.01 (1.0006)	2.33 (0.0233)	1.03
98S	30.86 (0.0031)	0.75 (0.0001)	12.53 (1.2530)	2.30 (0.0230)	1.28
95S	24.14 (0.0024)	0.56 (0.0001)	10.45 (1.0450)	1.67 (0.0167)	1.06
1001S	40.10 (0.0040)	1.13 (0.0001)	18.05 (1.8050)	3.44 (0.0344)	1.84
104S	22.03 (0.0022)	0.64 (0.0001)	10.25 (1.0250)	2.35 (0.0235)	1.05
103S	10.41 (0.0010)	0.32 (0.0000)	3.98 (0.3981)	0.81 (0.0081)	0.41
Salmon					
130S	157.63 (0.0158)	3.68 (0.0004)	73.62 (7.3623)	13.88 (0.1388)	7.52
129S	66.38 (0.0066)	1.75 (0.0002)	33.38 (3.3384)	7.66 (0.0766)	3.42

table 7, continued

1	2	3	4	5	6
Salmon					
133S	97.54 (0.0098)	1.86 (0.0002)	40.16 (4.0161)	8.26 (0.0826	4.11
134S	104.76 (0.0105)	1.80 (0.0002)	37.77 (3.7773)	8.06 (0.0806)	3.87
135S	85.12 (0.0085)	2.39 (0.0002)	36.50 (3.6496)	6.21 (0.0621)	3.72
136S	101.40 (0.0101)	2.11 (0.0002)	42.95 (4.2954)	8.72 (0.0872)	4.39
137S	104.20 (0.0104)	2.12 (0.0002)	45.84 (4.5841)	8.74 (0.0874)	4.68
143S	117.65 (0.0118)	3.04 (0.0003)	56.09 (5.6093)	11.37 (0.1137)	5.74
144S	93.13 (0.0093)	1.80 (0.0002)	42.87 (4.2872)	8.40 (0.0840)	4.38
Cod liver					
122Sdw	691.88 (0.0692)	18.14 (0.0012)	690.33 (69.0326)	168.50 (1.6850)	70.79
112Sdw	280.82 (0.0280)	12.97 (0.0013)	206.05 (20.605)	37.85 (3.7847)	24.42
116Sdw	853.33 (0.0853)	26.98 (0.0027)	692.83 (69.2828)	182.56 (1.8256)	71.20

Figures in brackets stand for the TEQ values

TEF values for CB 81 - 0.0001
CB 77 - 0.0001
CB 126 - 0.1
CB 169 - 0.01

analyzed herring samples was from 0.41 to 4.73. Of these, 18 samples had values lower than 2 and in only 4 samples was it higher than 3.

The TEQ in 22 sprat samples ranged from 1.99 to 3.89, of these 15 were from 2 to 3. Slightly higher TEQ values from 3.42 to 7.52 were determined in salmon samples.

The highest TEQ values, that sometimes even exceeded 70 pg TEQ-WHO/g sample wet weight, were noted in samples of cod liver.

The analysis of the data from the samples (Tables 6 and 7) indicates that the lowest TEQ are not connected with any particular fishing ground, but are rather related to fish species and age.

In most cases, each of the tested species is characterized by a similar non-ortho PCB profile (Table 7). Congener 77 in herring pg/g sample wet weight ranged from 10.41 to 43.07, PCB 81 from 0.32 to 2.26, PCB 126 from 3.98 to 47.0, PCB 169 from 0.81 to 6.61. In sprat PCB 77 occurred in the range of 57.50-113.41, PCB 81 from 1.10 to 3.44, PCB 126 from 19.59 to 38.0, PCB 169 from 2.90 to 15.26. In salmon PCB 77 ranged from 66.38 to 157.6, PCB 81 from 1 75 to 3.68, PCB 126 from 33.38-73.62, PCB 169 from 6.21-13.88.

The majority of tested fish had the highest levels of congener 77; however, congener 126 had the greatest impact on TEQ as determinations indicated that this one had the highest TEF (= 0.1) of this group of PCBs. Its contribution to total TEQ ranged from 96 to 98 % in the studied samples.

DISCUSSION

Due to the few available data on the non-ortho PCBs content in Baltic fish and the increasing interest in this group of compounds as well as the toxic impact they have on the environment (Anon. 2004a), SFI researchers developed a determination method that can be performed in their laboratory. Co-operation with the NIFES in Bergen permitted confirming the reliability of the method proposed by the SFI. It also permitted obtaining results of non-ortho PCB content in 72 samples of Baltic fish.

The analysis of coplanar PCBs in foodstuffs (as well as in other environmental elements) is not an easy task for several reasons:

– above all, they occur in samples at very low concentrations (expressed as pg/g);

– there are 209 congeners with similar physical and chemical properties, which makes differentiation extremely difficult;

– there are many compounds in the tested matrix that interfere with determining non-ortho PCBs, and these compounds have to be successfully separated (Jaouen-Madoulet *et al.* 2000, Hess *et al.* 1995, Megginson *et al.* 1995).

In consequence, the preparation of samples for analyses requires several steps of purification and separation, which usually results the losses of analyts. Therefore significant effort was devoted to improving the efficiency of the consecutive steps of the analysis. Different methods have been developed to extract, purify, and detect non-ortho PCBs as well as other chloroorganic compounds. These methods involved such techniques as:

– supercritical fluid extraction (Saito and Yamauchi 1990, Atuma *et al.* 1998);

– solid phase extraction (Grochowalski *et al.* 1993, Mattaleb and Abedin 1999);

– accelerated solvent extraction (Pihlstrom *et al*. 2002);

– gel permeation chromatography (Kuechi and Leonard 1978, Chamberlain 1990, Fisher *et al.* 1993);

– column adsorption chromatography with different, highly varied adsorbents (Wells *et al.* 1985, Voogt *et al.* 1986, Loos *et al.* 1997);

– dialysis with semipermeable polyethylene membranes (Berquist *et al*. 1993, Strandberg *et al*. 1998 , Grochowalski 2000, Grochowalski *et al.* 2000, De la Torre *et al.* 1995, Meadows *et al.* 1993, Hofelt and Shea 1997);

– high performance liquid chromatography using various columns (Wells *et al.* 1995, Molina *et al.* 2000, Atuma *et al*. 1998, Martinez-Cored *et al.* 1999).

The relatively simple method proposed by the SFI is based on the critical evaluation of several reports in the literature. It involves extracting fat with hexane in a Soxtec-Avanti device, defatting with dialysis through a semipermeable polyethylene membrane followed by treatment with concentrated sulfuric acid with oleum, isolating non-ortho PCBs with high performance liquid chromatography and performing the final determinations with capillary gas chromatography / electron capture detection.

Dialysis is recommended by increasing numbers of researchers as an efficient approach to the separation of hydrophobic organic compounds such as PCBs from large amounts of lipids. The recoveries of analyts with dialysis were satisfactory in the range of 50 to 90%. It is a simple, non-labor-intensive technique that does not require complicated apparatuses or special materials, in contrast to gel chromatography or the useful, but extremely labor-intensive, adsorption column chromatography. This technique appeared to be very useful in our laboratory.

Applying the HPLC technique to purify samples from matrix compounds permitted determining levels of coplanar PCBs with the GC/ECD. It is commonly thought that when this method is used to analyze food samples the complexity of the matrix often can lead to false negative or positive results caused by the presence of co-extractable compounds that can interfere with non-ortho PCB analysis. Therefore, GC/MS or preferably HRGC/HRMS are recommended for this type of determination. However, our tests conducted on reference material and the comparison of the our results with those obtained at the Cracow University of Technology (GC/MS) proved that separating non-ortho PCBs from extracts of fish samples with the HPLC technique was effective enough to apply cheaper GC/ECD to the final analysis. Although it was not possible to separate chloroorganic pesticides such as hexachlorobenzene (HCB) or p,p'-DDD, these compounds do not interfere with the identification and determination of non-ortho PCBs during GC/ECD since their retention times differ enough from those of the determined compounds. The co-operation with the laboratory in Bergen also confirmed that the SFI method of sample preparation for non-ortho PCB determination is appropriate.

To summarize, the results obtained during validation indicate that the method proposed by the SFI produces reliable results with acceptable repeatability and reproducibility and can be applied in determinations of non-ortho PCBs.

The results of TEQ from non-ortho PCBs expressed as pg TEQ-WHO/g sample wet weight in the tested Baltic fish depending on species ranged from 0.43 to 8.4. Only in the case of cod liver oil were these results higher, exceeding 70 pg TEQ-WHO/g sample wet weight. The comparison of the results of our study with data from the fish of other regions such as the Mediterranean, the North Sea or the Atlantic ranging from 0.23 to 1.49; (Anon. 2000b, Horst *et al*. 2002), indicated that Baltic fish contain more non-ortho PCBs. However, the comparison of our data with that in a Swedish report (Anon. 2004b) or with that from monitoring conducted in Estonia (Otsa *et al.* 2003) indicates that the data for Baltic fish are within the ranges determined by both Swedish and Estonian researchers.

It is essential, however, to emphasize that, to date, the available data base regarding non-ortho PCBs in Baltic fish is relatively small. Thus, it is currently difficult to draw any unequivocal conclusions regarding the health quality of Baltic fish with regard to non-ortho PCBs, especially since permissible limits for these compounds have yet to be set. Additionally, many authors stress that levels of dioxin and dioxin-like PCBs depend on many factors including specimen size, length, age, sex, fat content, and the location and time of catch. Thus, in order to identify regions that are definitively clean and decreasing or increasing tendencies, a huge amount of data is necessary. Therefore it is very important that the PCDD/F/PCB contents of Baltic fish are monitored.

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The status of the turbot *Psetta maxima* (L.) stock supporting the Baltic fishery

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Abstract. Turbot catches in the Baltic Sea reached 1,200 tons in 1996, whereas before the 1990s the reported annual turbot catches seldom exceeded 200 tons. Half of the total annual turbot catch was taken from sub-divisions 25 and 26. In recent years, turbot catches have exhibited a downward trend, and the average catch rate index recorded in Lithuania fishery fell from 5 kg to 1.6 kg per standard net. The results of the analysis of 67,062 turbot length measurements and 5,187 fish age determinations from materials collected in 1995-2004 were used to describe the turbot population.

Length ranged from 16 to 63 cm and fish age ranged from the age groups 2 to 14. The bulk of the catch was comprised of fish measuring from 32-35 cm. Of the turbot caught with gill nets used in flounder fishery (65-70 mm mesh size), 72% of fish were undersized (< 30 cm). This paper presents and discusses the effects of the mesh size used in gill net fishing on the length composition of the turbot caught. Gill nets with 110 mm mesh size used by fishermen specializing in turbot fishery proved to be the most selective. The growth rate of turbot males is slower in comparison with that of females and their maximum sizes recorded in the sampled materials were 37 cm and 63 cm, respectively. The values of the von Bertalanffy's growth equation parameters given in the literature were reviewed and those of $L_\infty = 55$ cm, $K = 0.122$, $t_0 = -2.569$ (for females) estimated from the data collected in 2004 were used to evaluate the optimum minimum length size (t_c) at the range of natural mortality coefficient 0.1-0.25.

The regulatory measures for turbot fishing currently enforced in Lithuania, Poland, and Russia are reviewed and discussed in light of the authors' findings.

Key words: Baltic turbot, annual catch, growth rate, mortality rate, age structure, gillnet selectivity, minimum landing size, closed season

INTRODUCTION

The turbot (*Psetta maxima* [L.]) is a sinistral (with the eyes on the left side) fish that is differentiated from other flatfish by its almost circular body form. The scales have transformed into bony tubercles and the long base of the pelvic fins is devoid of a spine (Nielsen 1986). According to Nelson (1984), the turbot belongs to the family Bothidae (subfamily Scophthalminae). Nielsen (1986) gives the subfamily the status of a family, similarly to Chanet and Wagemans (2001). The virtual absence of genetic diversity in turbot is, according to Blanquer *et al.* (1992), an effect of a very low evolutionary rate. Results obtained by Estoup *et al.* (1998) and Bouza *et al.* (2002) seem to suggest high gene flow in turbot, even between geographically distant populations. This fish species is common in European costal waters, including the Mediterranean and Black seas (Nelson 1984, Anon. 1999) and is highly fecund producing from eight to ten million (Benguria and Camińa, 1975) and even as many as thirteen million eggs (Popova 1966). Despite this and the fact that it has few potential predators, it is not abundant.

Turbot is frequently reported in Baltic catches taken in the waters extending from the western boundary near Øresund to the Åland Sea. This species reaches larger sizes than other fishes of this family in the Baltic Sea, and among the fish spawning there, with the exception of salmon and cod, turbot attains the largest sizes. Results of experiments with turbot tagging by Aneer and Westin (1990) suggest this species is sedentary.

Turbot contributes 1% to the global flatfish catch (9,200 tons in 2000 and 2001). Turbot has been recorded in Baltic fisheries statistics since the early twentieth century (Anon. 1910), when it did not exceed 200 tons annually. Since 1987, catches of turbot had grown to 1,200 tons by the 1993–1996 period (Anon. 2004). Catches are limited practically to southern Baltic waters (ICES sub-divisions 22, 24, 25, and 26).

Due to a lack of demand for turbot in Poland after World War II, this species was only caught as by-catch in targeted cod and flounder fisheries. Larger individuals were valued only in coastal localities where there was a long-standing tradition of marine fish consumption. Smaller individuals were considered less valuable than flounder. According to information collected during interviews with older fishermen, prior to World War II some boat fishermen specialized in turbot fishing with large mesh size gill nets in the Gdansk Bay. In Lithuania and Russia (formerly the USSR), coastal boat fishery was banned and the turbot caught by cutters operating in Baltic offshore waters was not sorted from flounder and was sold as “flatfish”.

The fishermen’s attitude towards turbot changed at the beginning of the 1990s when it became possible to export turbot to western European countries. The prices paid to Lithuanian, Polish, and Russian fishermen for turbot suddenly rocketed in relation to other fish species, and this prompted them to direct more fishing effort towards it. The primary threat to this species at the moment is that it is caught frequently with flounder that is fished with small meshed gears. This results in the catch of undersized turbot, which might mean that this species may soon cease to be of importance to the fisheries of the southern Baltic. The importance of the Baltic turbot should not be considered in light of economic criteria only. This fish, which begins to prey on small fish when it reaches a length of 60 mm (Iglesias *et al*. 2003), plays an important role as top predator in the Baltic environment.

Extensive descriptions of the turbot inhabiting the southern Baltic was published by Kändler (1944) and Stankus (2003), while the characteristics of the Baltic turbot fished at the Polish and Lithuanian coasts in the 1990s were described by Draganik *et al*. (1996).

The aim of the current work was to describe the state of the population of exploited turbot in the exclusive economic zones (EEZ) of Lithuania, Poland and the Russian Federation in the 1995-2004 period. It also reviews and discusses the respective fishery regulatory measures enforced by the International Baltic Sea Fisheries Commission that are binding in these three countries, and the impact they have on turbot stocks.

MATERIALS AND METHODS

The material was collected in the waters of the Lithuanian, Polish, and Russian fisheries exclusive economic zones **(**Fig. 1**)**. Sample collection for research, specimen measurement, the evaluation of biological features, and the measurement and description of the applied fishing gears was performed by personnel from the Fishery Research Laboratory in Klaipeda, Lithuania, the Sea Fisheries Institute in Gdynia, Poland, and AtlantNIRO in Russia. The materials from the Polish and Russian EEZs were supplied by the turbot sample analysis results collected from commercial fisheries in the 1995-2004 period. The numbers of fish measured and aged in Lithuania, Poland, and Russia annually are presented in Table 1. The total length, body mass, and maturity stage of the turbot sampled were recorded, and otoliths were removed for age determinations.

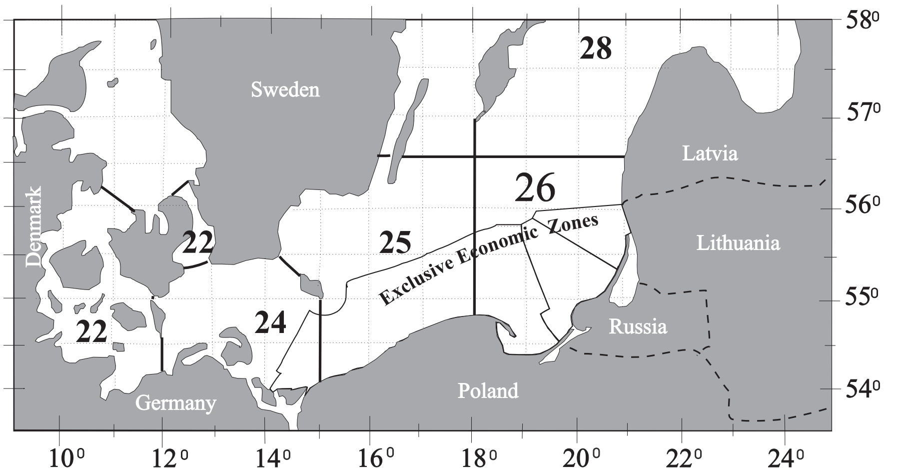


Fig. 1. Statistical sub-divisions and respective turbot catch distribution in the southern Baltic.

Table 1. Number of measured and aged turbot considered in the study

Year	Lithuania[1]		Poland		Russia	
	measured	aged	measured	aged	measured	aged
1995	520				1,943	335
1996	95		1,605	390	766	257
1997	636		1,784	147		
1998	248		595	124	1,027	332
1999	184	720	895	274	4,506	300
2000	102		136	38	13,618	671
2001	290		768	302	11,483	510
2002	98		397	74	20,908	668
2003	87		436		1,506	159
2004	39		637	157	1,753	449 226[2]

[1] Experimental catches only
[2] Fish sampled from the trawl catches in 1995-2004

Descriptions of catch location, timing, and the characteristics of the applied fishing gear were recorded for each sample. The fish sampling was conducted according to relevant procedures described in ICES documents (Anon. 1994, 2002a). The turbot gonad maturity stage was determined according to the BITS five-stage code (Anon. 1994, 2002a).

The ages of the sampled turbot were determined by counting the number of annual rings on the otoliths. It was assumed that the pair of concentric rings visible on the otoliths in alternating zones of opaque and hyaline constitutes separate annual growth rings.

The materials regarding gill net selectivity were obtained from experimental catches performed with gill nets of different mesh sizes ranging from 45 to 120 mm bar length. Catches were made in five fishing grounds located within the Lithuanian EEZ (Fig. 2) in the 1995-2003 period. A set of gill nets consisting of twelve nets, each one with a different mesh bar length (45, 50, 55, 60, 65, 70, 75, 80, 90, 100, 110, and 120 mm), was set in each fishing ground at least five times monthly throughout the year. The nets with mesh sizes 45, 50, 55, and 60 mm measured 1.6 m x 30 m; while nets with mesh sizes over 60 mm bar length measured 3.2 m x 60 m. The depths where experimental fishing was conducted ranged from 6 m to 14 m, and soaking time was 24 hours.

The survey on the abundance and distribution of juvenile turbot (fish in age groups 0 and I) in shallow coastal waters of up to 2 m was conducted according to the procedure in Sandström *et al*. (1994) using a fry drag with a mesh size of 6 mm at the wings and 4 mm in the codend. The catches were conducted from August 21 to September 10 at twenty-two selected Lithuanian shallow coastal water stations (Fig. 2). The exact geographical position of the trawling places was determined with GPS.

The Beverton and Holt model (1957) was used to evaluate the dependence of turbot stock productivity in the southern Baltic on the intensity of exploitation. Model efficiency was limited by the necessity of having information on the numbers of fish (*R*) which were recruited to the exploited stock annually. Since this information is rarely available, this requirement is bypassed by evaluating the yield index in yield per recruitment units assuming that recruitment level (*R*) oscillates over a series of years without a trend, which justifies the acceptance of a relative yield index. Thus, this model (equation 1) facilitates identifying changes that will affect catches in relation to the magnitude of the fish mortality generated by fishing effort and the age at which fish become vulnerable to fishing gear.

$$Y/R = FW_\infty \exp(-M(t_c - t_r)) \sum_{n=0} \frac{3Q_n \exp(-nK(t_c - t_0)}{Z + nK} [1 - \exp(-(Z + nK)(t_m - t_c)] \quad [1]$$

where:
Y – yield in weight;
Z – instantaneous rate of total mortality ($F + M$);
F – instantaneous rate of fishing mortality;
M – instantaneous rate of natural mortality;
t_m – maximum age (in calculations it was assumed that $t_{max} = 10$);
t_c – age at entry to exploited phase (≈ mean selection age);
t_r – age at recruitment (in this paper it was assumed that $t_c = t_r$);
W_∞, K, t_0 parameters of the Brody-Bertalanffy growth equation (Ricker 1975)
$Q_0 = 1$, $Q_1 = -3$, $Q_2 = 3$, $Q_3 = -1$;

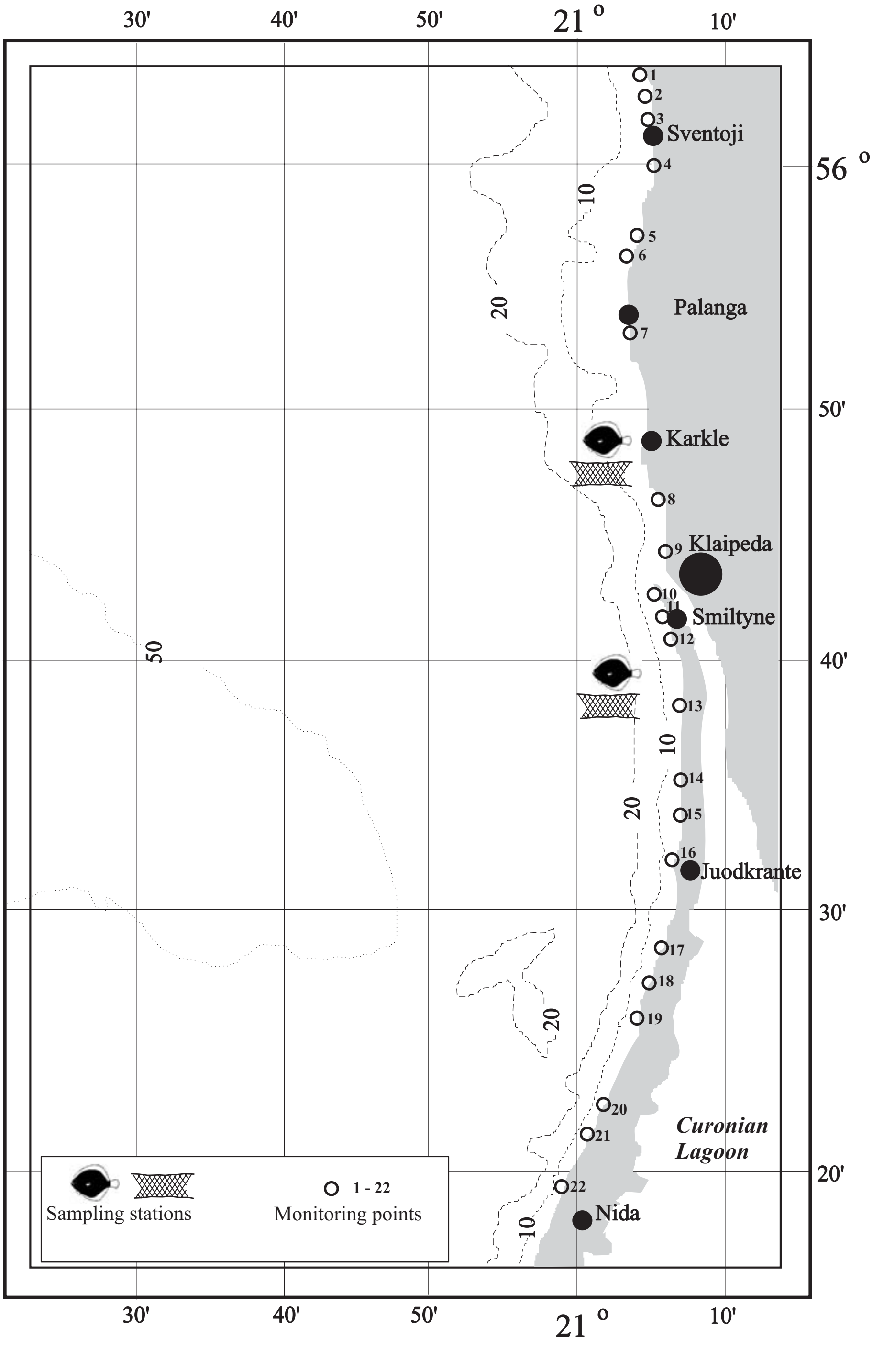


Fig. 2. Localities of monitoring stations and points in the Lithuanian coastal zone

Equation (1) was modified by Jones (1957) (cited in Ricker 1975) to:

$$Y = \frac{F \cdot N_0 \cdot e^{F \cdot r} \cdot W_\infty}{K} \left(\beta\,[X, P, Q] - \beta\,[X_1, P, Q]\right) \qquad [2]$$

Slipke and Maceina (2000) modified equation [2] to:

$$Y = \frac{F \cdot N_t \cdot e^{Z \cdot r} \cdot W_\infty}{K} \left(\beta\,[X, P, Q] - \beta\,[X_1, P, Q]\right) \qquad [3]$$

where:
$X = e^{-Kr}$;
$X_1 = e^{-\mathbf{K}(t_\lambda - t_0)}$;
$t_\lambda = t_m$ = maximum age of fish in the population;
$P = Z/K$;
Q = slope of the weight-length relation + 1;
β = incomplete Beta function;
Z = instantaneous rate of total mortality ($F + M$);
r = time (in years) to recruit to fishery ($t_r - t_0$), $t_c = t_r$;

The value of the natural mortality coefficient (M) for studied turbot was estimated based on the Jensen (1996) equation:

$$M = 1.50 \cdot K \qquad [4]$$

and Chen and Watanabe (1989) (cited in Slipke and Maceina 2000):

$$M = (1/t_m - t_c)\ \ln(e^{K \cdot t_m} - e^{K \cdot t_0})/(e^{K \cdot t_c} - e^{K \cdot t_0}) \qquad [5]$$

In addition to the easier calculation procedure, equation (2) can also be applied to fish populations for which the equation coefficient between weight and length (b) is different than 3.

Formula (3) was used in this paper to generate the contours of the diagram of turbot stock efficiency as functions of the variable exploitation rate ($u = [F/Z \cdot (1 - e^{-Z})]$ and length (l_c) that corresponds to the age (t_c) when fish become susceptible to fishing gear.

$$u = (1 - e^{-(M+F)}) \cdot F/(F + M)\text{, Ricker (1975)} \qquad [6]$$

Fish length growth is described as follows:

$$l_t = L_\infty\,(1 - \exp(-K(t - t_0)) \qquad [7]$$

Two sets of data on the relation between mean turbot length at age groups were considered, namely derived from the length-age keys for the 1998-2000 period and for 2004. In the last case, only the data for females were considered and the values for $L\infty$, K, t_0 (Table 5) were calculated under the assumption that L_∞ is constant (Slipke and Maceina,

2000) and those of W_∞ were estimated using estimated parameter values *a* and *b,* which determine the relationship between length (*L*) and fish weight (*W*) (Ricker 1975).

$$W = a\,L^b \qquad [8]$$

In the entire monitored age data set only two fish were assigned to age groups older than 12; in the calculations the maximum age ($t_{max.}$) attained by turbot in the fishable population was assumed to be 12 years. The program FAST (Slipke and Maceina, 2000) was used to evaluate changes in l_c versus u values at *M* ranging from 0.1 to 0.25.

The relationship between average total turbot length (L_{av}) and mesh size (m_s) was calculated using the linear relationship:

$$L_{av} = a\,m_s + b \qquad [9]$$

The selectivity curves were fitted following the Millar and Holst (1997) model with the open source code (GNU license) for *R* environment written by Russel Millar (Department of Statistics, University of Auckland, New Zealand).

Turbot catch distribution within the limits of the Polish EEZ according to the defined standard fishing grounds (Długosz *et al.* 1993) was evaluated using daily catch records in the fishing logs reported to the Sea Fisheries Institute in Gdynia in the 1993-2003 period.

RESULTS

Reproduction

Observations of maturity stages in both sexes indicated that in the Russian and Polish zones of southern Baltic coastal waters males reach sexual maturity at a younger age (per age group) in comparison with females. This means that in the spawning population observed there were males belonging to age groups 2 and 3 (total length 17 to 27 cm). Individuals categorized in age group 4 dominated (22-29 cm Tl) while the females noted belonged to age group 4 and older. Males exceeding 31 cm Tl were seldom noted in the catches.

Analyses of materials collected in the waters of the Polish EEZ indicate that females reached sexual maturity in age group 4 and most of them in the spawning population belonged to age groups 4 to 6 (in the Russian zone – 4 to 5) and that these fish measured from 31 to 40 cm Tl.

The sampling results of Russian EEZ waters in the Kaliningrad region and the analyses of daily water temperatures and state records of turbot gonad development indicated that the gonad development rate and, in consequence, turbot spawning time are correlated with the increasing temperature of coastal surface waters. Observations made in the 2000-2004 period confirmed that the abundance of spawning females over the last ten days of May was dependent on the water temperatures of the first ten-day period of May (Table 2). The strictest relationship (correlation factor of $r = 0.92$) was noted between these two variables and was described by the following:

Table 2. Relationship between average surface water temperature in the Russian EEZ in May and the proportion of turbot females with ripe* gonads

Year	2000	2001	2002	2003	2004	R^2
Average temperature – May 1-10	13.4	10.5	14.7	9.2	12.2	0.84
% turbot females with ripe* gonads – May 21-31		4.7	12.6	1.7	4.1	

*Maturity stage "spawning " - BITS code 3

$$y = 0.0001\ x^{4.2743}. \qquad [10]$$

where:
y – proportion of turbot females with ripe gonads in the last ten day period of May,
x – average surface water temperature in the first decade of May.

Table 3. Ratio of turbot males to females in turbot catches made in the Kaliningrad region in May – June 2000-2004

Decade	Month	
	May	June
I	0.07	0.22
II	0.10	0.20

Data collected in the 2000-2004 period indicated that turbot females with gonads in the spawning stage assembled in spawning grounds when water temperatures reached 9.2ºC and that the quantities of females increased as the water temperature rose. As the spawning intensity increased (measured by the proportion of fish with gonads in spawning stages), the proportion of males in catches increased (Table 3). It could not be precluded that the observed domination of females in the spawning grounds might have been an effect of the selectivity of the applied gear (mesh size 110-120 mm), which retained more females since they had faster growth rates (Stankus 2001), or they had gathered earlier at the spawning grounds.

Young turbot distribution

Testing the results of young turbot from Lithuanian coastal waters provided a basis for the indirect assessment of changes in abundance over a series of years.

The annual indices of the abundance and biomass of young turbot (per 100 m^2) were assessed based on the analysis of the collected materials (Piščikas 2004). The average annual index of young turbot abundance was calculated from the material collected at points situated along a 90-kilometer stretch of shallow water of up to 2 m deep. The results indicate there were abundant generations in 2000 and 2001 **(**Fig. 3), and the average turbot abundance index for twenty-two sampling sites in these years was 7.16 and 5.9 specimens per 100 m^2 of trawled bottom surface. These figures were 4.6 times higher in comparison with the average index from the 1997-1999 period. This relation for biomass was 7.4.

This provides the foundation for forecasting an increase in the turbot population at spawning grounds located along the Lithuanian coast in 2005-2006 as a result of the spawning success in 2000-2001.

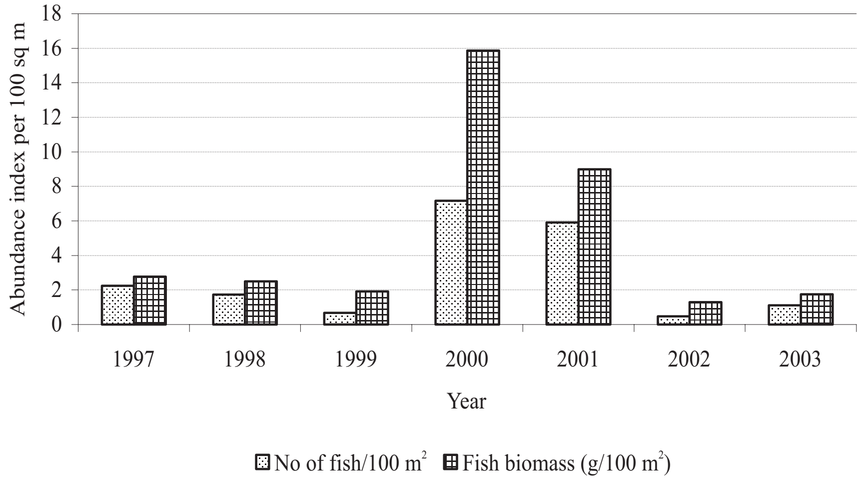


Fig. 3. Indices of young turbot abundance in Lithuanian coastal waters.

Length, age, and growth of turbot in the exploited population of the southern Baltic

The total length (TL) of the turbot registered in samples taken from catches made by Polish and Russian fishermen ranged from 15 to 53 cm and 16 to 63 cm, respectively. A significant quantity of the turbot measured came from samples of flounder landings that had been caught using gill nets with 60-65 mm mesh bar length in which turbot was by-catch. If it is accepted that 5% the quantity of the sample is the minimum criteria of fish catch contribution for assessment, then the scope of turbot size range in Polish catches was from 22 to 36 cm and that in the Russian catch was from 25 to 41 cm (Figs. 4A and 4B).

The length-age key constructed using Russian (1995-2004) and Polish (1998-2004) materials indicates that fish assigned to age groups 2 to 14 were present in the exploited Baltic turbot population (Annex 1 and Annex 2).

The analysis of the age of the exploited turbot population in the Polish and Russian zones indicated that the catches in the southern Baltic in 1995-2004 were sustained by fish assigned to age groups 4-7 (Tables 4A and 4B). A similar age scheme in the experimental catch of turbot in the Lithuanian zone in 1990-1999 was noted by Stankus (2002).

Data on the turbot length at age available in the literature and those from the Polish turbot aged samples in 2004 (females only) were used to evaluate the von Bertalanffy's growth equation parameters compiled in Table 5. The growth curves constructed on the basis of these parameters emphasized the differences between the male and female growth rate regardless of the time or the ageing techniques of the author. The parameter values of the female turbot growth rate from Polish data from 2004 were used to apply the model of fishing influence on the efficiency of the Baltic turbot stock. These values were close

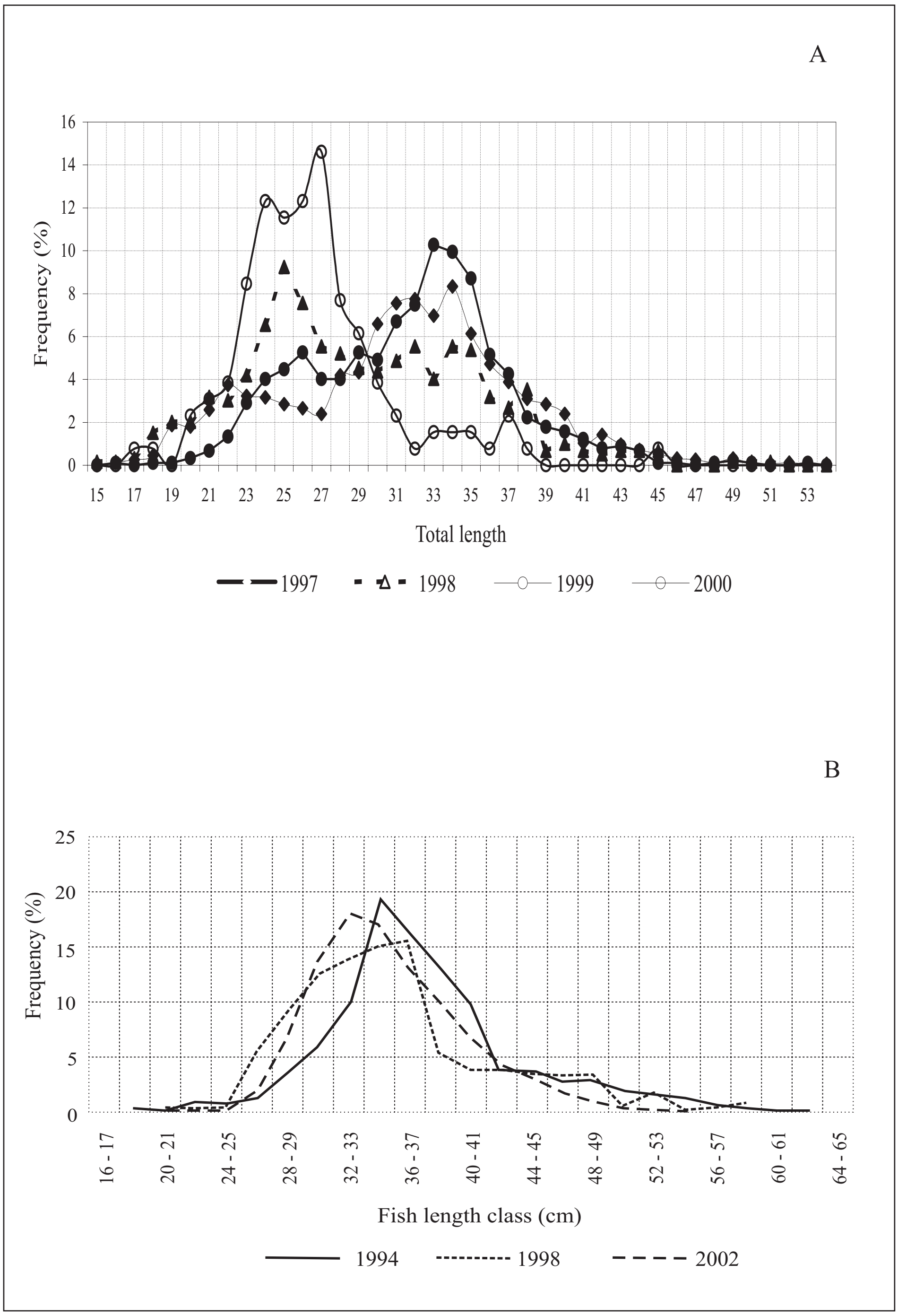


Fig. 4. Length structure of the turbot catches sampled in Poland (A) and Russia (B).

Table 4A. Age composition of the turbot caught by the Polish fishery (%)

Year	Age group										
	2	3	4	5	6	7	8	9	10	11	12
1998	8.1	21.0	12.9	16.1	19.4	8.1	6.5	1.6	0.0	3.2	3.2
1999		8.0	15.3	16.1	19.0	22.6	7.3	5.8	5.8		
2002	6.4	17.5	15.7	9.5	17.4	18.6	10.0	3.1	0.8	0.8	0.3
2003		0.0	2.7	6.8	24.0	34.0	18.5	8.5	2.6	1.6	1.2
2004		10.3	39.4	21.3	14.8	9.7	3.2	1.3			

Table 4B. Age composition of the turbot caught by the Russian fishery (%)

Year	Age group								
	2	3	4	5	6	7	8	9	10+
1995	0.2	2.6	20.1	32.2	28.5	7.9	3.6	2.6	2.3
1996	0.5	3.8	16.5	20.5	30.1	15.9	5.5	2.7	4.5
1998	0.2	10.0	25.3	24.1	12.2	6.7	6.5	5.4	9.6
1999		4.2	19.4	25	22	16	8.3	3	3
2000	0.1	8.8	19.2	17	20	15	12.8	3.5	4
2001	0.2	3.9	30.5	15	16	14	14.2	2.8	4
2002		12	26.3	23	12	13	9.3	2.2	2
2003	0.1	2.9	28.4	26.5	16.5	9.6	10.0	1.2	4.8

Table 5. Values of von Bertalanffy's growth equation parameters for turbot from the Baltic, North,
and Black seas

Source	Parameter values					
	L_∞ (cm)		K		t_0	
	males	females	males	females	males	females
Baltic Sea						
Kändler (1944)	34.75	53.5	0.21	0.152	0.035	0.301
Cięglewicz *et al*. (1969)	32.9	51.3	0.381	0.215	0.45	0.35
Stankus (2002	35.0	53.5	0.301	0.186	0.35	0.28
Polish data (1998-2000)	40.4	60.6	0.123	0.091	-3.66	-2.603
Estimated from Polish data (2004)*		55.0		0.122		-2.569
North Sea						
Derived from the data of Leeuwen and Rijnsdorp (1986) from 1984		47.3 67.8		0.505 0.291		0.466 0.353
Black Sea						
Avşar (1999)	82.6		0.17		-0.93	

* R^2 – 0.9145; Prob. > F 0.0007

to those used to describe a longer period by other authors (Stankus 2002; Kändler 1944). Correspondingly, the same growth data was used in equations (4) and (5) and produced an M value close to 0.18.

Effects of gill net mesh size on the length distribution of turbot in catches

The range of flatfish length retained in gill nets is relatively wider in comparison with other fish species. This is due to the flatfish body shape, with its high ratio of height to length. This, in turn, confirms the assumption that "knife-edge" selection for flatfish caught with gill nets is hypothetical. This complicates fitting the minimum mesh size to the fixed minimum landing length. This issue entails more problems as Baltic turbot and flounder coastal fishing grounds overlap, and the specific growth rates and ages of first maturity of these two species require different regulatory measures.

Figure 5 presents the different curve shapes that represent the length composition of the turbot catch made with three types of gear. The share of undersized fish was smallest in specialized turbot fishing with gill nets (110 mm mesh bar length). In contrast, fish measuring less than 30 cm Tl constituted the bulk of the turbot catch taken with trawl and flounder gill nets (65 mm mesh bar length).

The data collected in 1995-2004 allowed calculating the relationship between the average length (L_{av}) of turbot caught and mesh size (m_s) of the gill net. The curves reflecting length frequency of turbot retained by gill nets with twelve different mesh sizes did not present a clear picture as they overlapped, particularly with regard to smaller fish lengths.

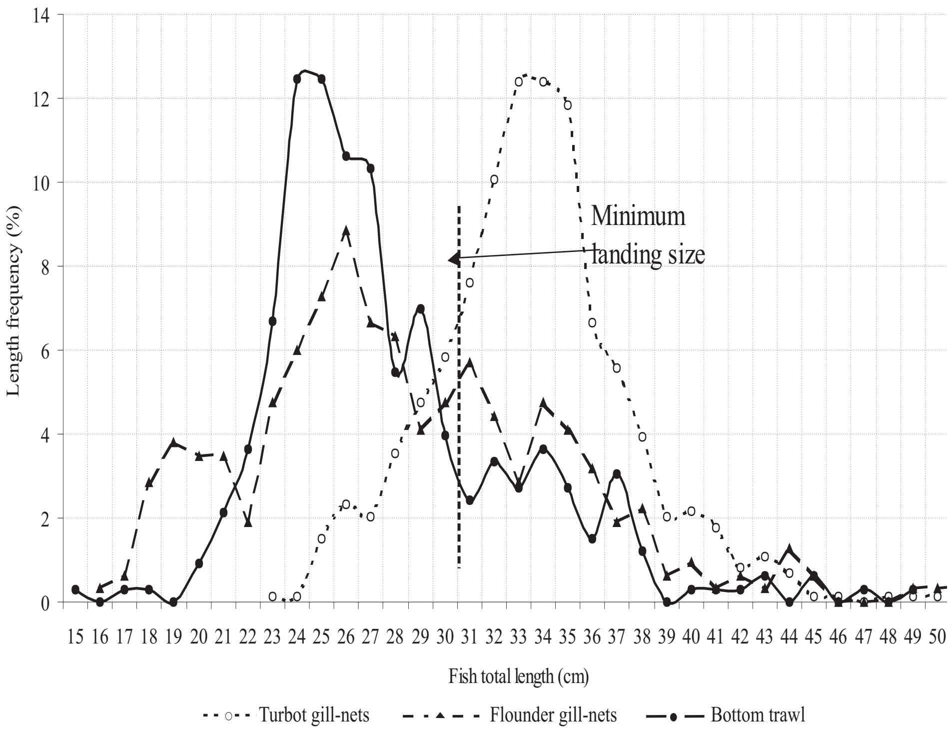


Fig. 5. Length structure of the turbot caught with various gear in the Polish EEZ.

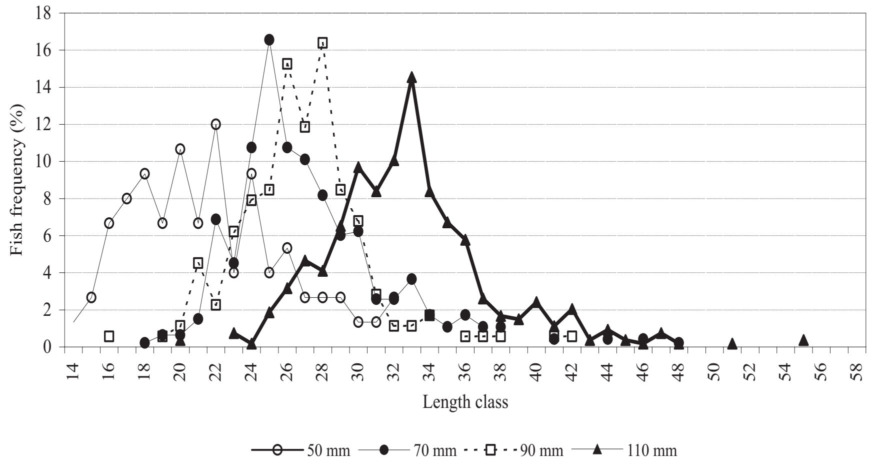


Fig. 6. Length frequency curves for the turbot fished with gill nets in experimental catches (Lithuania, for selected mesh sizes).

Figure 6 presents selected length catch curves derived for mesh bar lengths of 50, 70, 90, and 110 mm. Of the gill nets tested in turbot fishing, the one with 110 mm mesh bar length proved to be the most selective in protecting undersized fish measuring less than 30 cm Tl. However, a considerable amount undersized fish (22%) was retained.

Parameter values for the selectivity factor ($k = 0.2760$; $\sigma = 6.8516$) were derived. The deviation value of the model (664.6) at 122 degrees of freedom confirmed good model fit (Fig. 7). The results indicated that the probability that a fish below a 30 cm Tl would

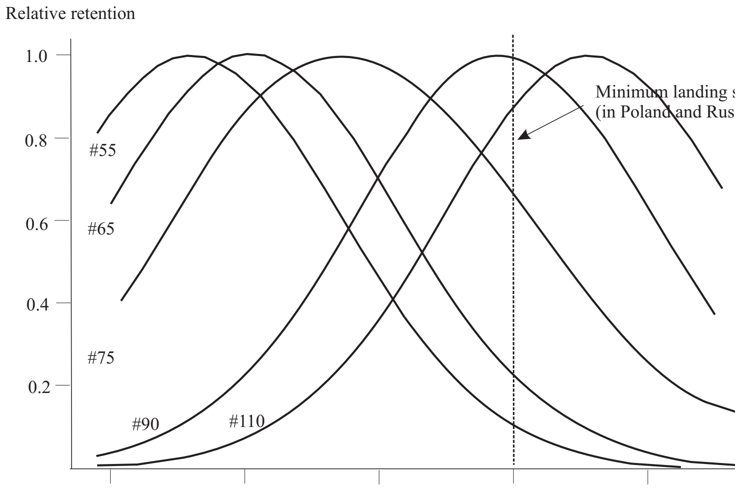


Fig. 7. Turbot selectivity curves for gill nets of different mesh size.

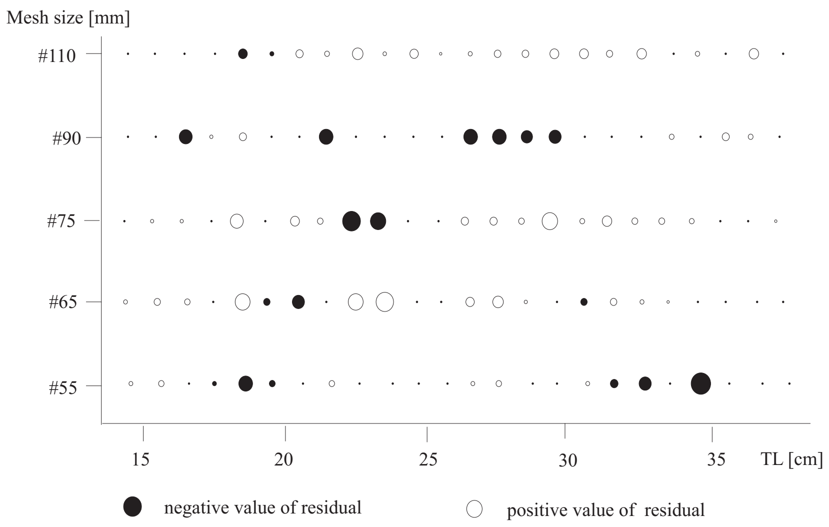


Fig. 8. Distribution of residuals reflecting the fitness of the data evaluated with the selectivity model to empirical data.

be entangled in a gill net with 110 mm mesh bar length was 0.8. The current binding minimum mesh size for gill nets used in turbot fishing in the Baltic Sea is 120 mm (Anon. 1997). Fishermen that deploy gill nets with 110-120 mm meshes are not only obeying the minimum landing length, they are also responding to consumer demand for larger fish. The residual distribution (McCullagh and Nedler 1989) presented in Figure 8 confirms there is good fit between the estimated model values and the empirical data.

Baltic turbot fishery and its impact on the stock

Hensen (1875) is a good source of historical information regarding Baltic Sea turbot fishery. He reported that turbot was an important component of the fisheries of the southern Baltic waters stretching from the Curonian Spit to the Pomeranian Bay, excluding the strip of coast between Łeba and Dziwnów. Kändler (1944), Nielsen (1986), and Virbickas (2000), all authors who describe turbot ecology, emphasize the preference of this species for sandy-stony bottoms. Interestingly, the German, Lithuanian, and Latvian words for turbot are all related to the respective words for 'stone'. The analyses of multi-year bottom trawl catch data for turbot in the Polish waters during the 1993-2002 period (Długosz *et al.* 1994, 1995, 1996, 1997, 1998, 1999, 2000, Miłosz *et al.* 2001, 2002, 2003) confirm these regularities. The highest quantity of turbot were caught in the fishing grounds of the Słupsk Furrow region where the bottom is sandy or stony**.** Following a review of detailed, long-term statistics series on the Polish fishing fleet, the authors concluded that the impact of the fishing effort on turbot can be omitted.

Turbot was recorded in fishery statistics published by the ICES in the first decade of the twentieth century, and the total catch reported by Sweden and Germany in 1910 was 67 tons Anon. (1910). In the 1920s, this figure oscillated from 243 to 444 tons (Anon. 1908-1944). In the 1930s, when Poland joined the countries which were recording and reporting turbot catches to the ICES, the average, annual Polish catch in the 1929-1938 period was 37 tons. The quantities of turbot caught by Lithuanian fishermen in the years preceding World War II were not recorded in ICES statistics, but at the end of the 1920s this figure was 28 tons (Table 6). Polish turbot catches in the 1920s were included in total flounder catches (Anon. 1928, 1931). Hryniewiecki (1925) estimated that Polish turbot catches comprised as much as 5% of the total flatfish catch. Taking into consideration that landings of flatfish by Polish fishermen operating in the Baltic in 1922-1927 oscillated between 417 and 909 tons (Hryniewiecki 1925, Anon. 1928), it can be assumed that the range of the annual turbot catch was within 20-45 tons. Although minimal legal lengths for flounder and plaice were obligatory in the Polish Baltic fishery in the 1920s, these regulations did not encompass turbot (Hryniewiecki 1925, Anon. 1928). The species was also omitted from one of the earliest international treaties regulating fish catches in the Baltic (Anon 1929).

Table 6.Turbot catches made by Lithuanian and Polish fisheries in 1926-1938 (kg)

Year	Lithuania[1]	Poland
1926	11,200	
1927	6,500	
1928	14,400	44,000[2]
1929	28,700	46,000
1930	21,300	30,000
1931	3,400	43,000
1932	18,000	43,000
1933		31,000
1934		33,000
1935		40,500
1936	26,200	
1937	28,700	
1938	28,600	

[1] Anon. 1939
[2] Anon. 1931

According to the ICES Bulletin Statistique, the annual turbot catch from Baltic Sea fishing grounds in the 1965-1982 period (except in 1977) did not exceed 200 tons (Anon. 2004), and until 1972 the data on turbot catches from statistical sub-divisions 25 and 26 were not available. This resulted from reporting combined catches of flounder, plaice, and turbot under the heading “Baltic flatfish”.

Baltic Sea turbot catches grew slowly from 1986 and reached their maximum of over 1,000 tons in 1993-1996. Following this period, there was a decline in catches (Fig. 9), which was especially notable in Polish catches and in other countries that intensified their turbot fishing efforts in the last decade of the twentieth century. This resulted from increased demand for turbot caused by the possibility of exporting it to western European markets where turbot is more highly valued than in central Europe. The quantity of the annual Polish turbot catch is estimated based on official catch statistics and the amounts of turbot reported as flounder by-catch. The amount of turbot by-catch is estimated using samples collected at flounder landings. The Polish turbot catch data in 1995 reported in statistics compiled by the ICES Baltic Working Group (Anon. 2004) do not include estimates that were reflected by the drastic decrease in the Polish turbot catch that year. In analyzing these turbot catch statistics, the authors assumed that the Polish turbot catch in that year was the average of the two closest years. The Polish data included the estimated turbot catches taken as by-catch in flounder fishery.

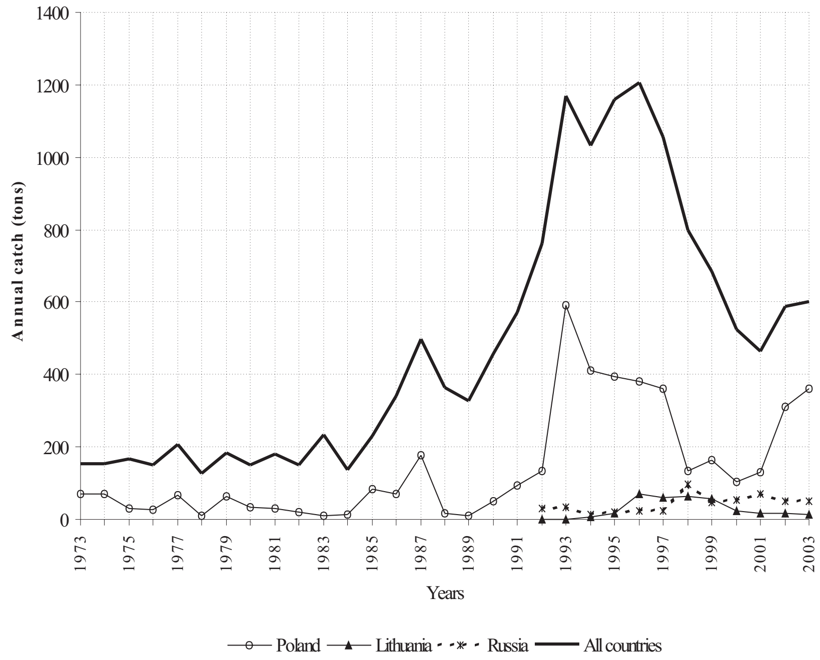


Fig. 9. Turbot catches made by Polish, Lithuanian, and Russian fishery in 1973-2003.

The mechanism of fishery influence on the Baltic turbot stock is presented by the authors based on the example of Lithuanian and Polish fisheries. The following four periods were selected in Lithuanian turbot fishery:

– the period prior to the war (1926-1938) characterized by catches of 7-27 tons;

– the 1939-1945 period – lack of fishery statistics;

– the 1945-1992 period – no coastal fisheries, unsorted cutter fisheries of Baltic flatfish;

– the period from 1993 to the present – specialized turbot fishery and an increase in catch statistics accuracy.

The Polish turbot fishery had a similar history, but in the 1945-1990 period flatfish was a substantial component of the catches taken in Polish coastal waters by boat fishery. Flatfish were important in the eastern region of the Gulf of Gdańsk where they constituted 54% of the total weight of fish caught in coastal fishery (Romański 1968). According to this author, in the 1962-1967 period turbot contributed from 3 to 8.5% to the flatfish catch in coastal fishery. The total length of the fish caught in the Gulf of Gdańsk ranged from 15 to 56 cm, while 22.4% of the turbot caught measured from 21 to 23 cm. The lack of demand from internal markets did not prompt fishermen to specialize in turbot fishery, and this species was seldom sorted out from flatfish catches. A similar Baltic turbot population exploitation model functioned in other central European countries.

The influence of increasing exploitation is reflected in the results of catch efficiency (catch per fishing effort unit – kg/75 m net/ fishing day) in commercial turbot catches for the 1997-2003 period in Lithuanian waters (Zolubas 2003; Fig. 10**).**

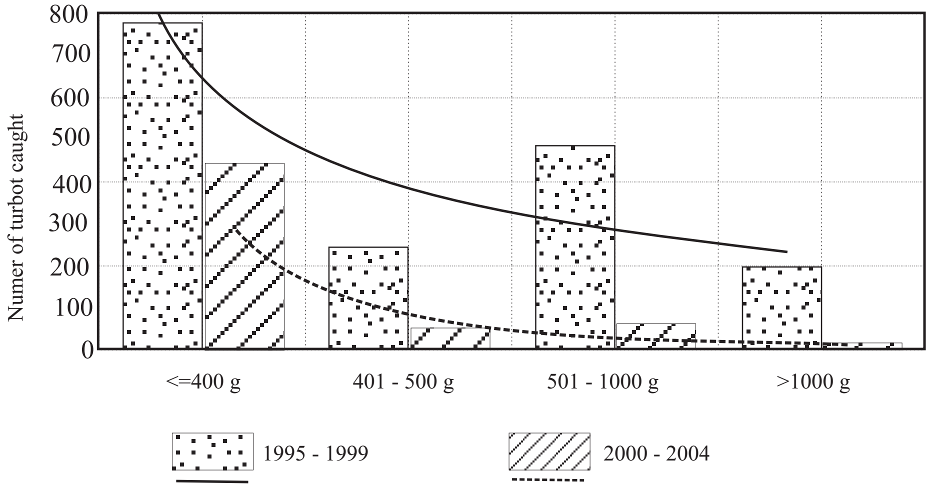


Fig. 10. CPUE indices for turbot fished with gill nets in Lithuanian coastal waters in 1997-2003 (Zolubas 2003).

The results of abundance and length analyses of turbot caught in Lithuanian waters proved that fish measuring from 32 to 51 cm contributed a considerable amount to the total catch in the 1995-1999 period. In the 2000-2004 period, the contribution of fish belonging to this section decreased such that their abundance was restricted to several individuals only **(**Fig. 11**).**

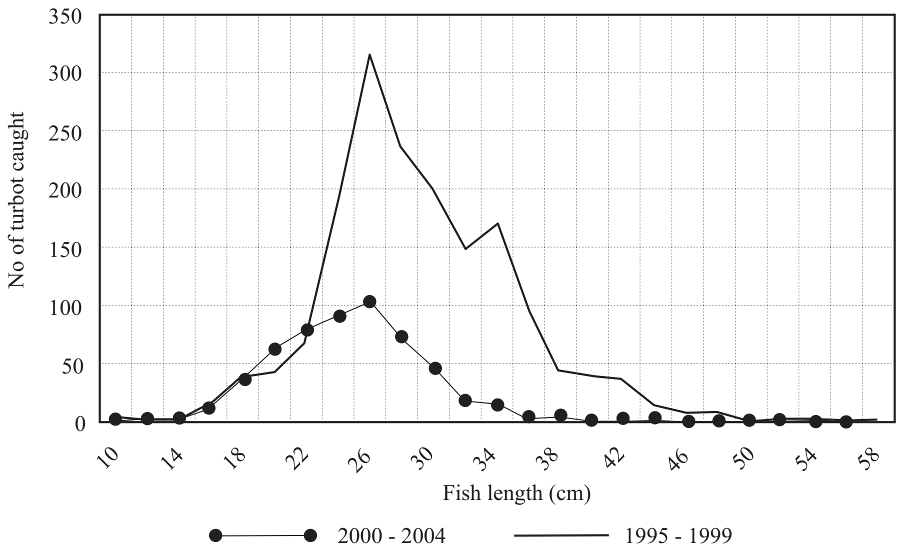


Fig. 11. Number of turbot caught with gill nets in experimental catches in 1995-2004.

In Polish fishery, the minimum turbot length of 18 cm was implemented by the Directive of the Ministry of Industry and Trade in 1930 (Anon. 1930). The regulatory measures in force in 2003 for turbot fisheries in waters under the jurisdiction of Lithuania, Poland, and Russia are compared in Table 7.

Table 7. Binding regulatory measures in specialized fishing for Baltic turbot in Lithuania, Poland, and Russia in 2003

Regulatory measures	Country		
	Lithuania	Poland	Russia***
Minimum mesh size (mm)	110*	130**	110****
Closed season	May 16-July 31	June 1-July 31	June 1-July 31
Minimum landing size (cm)	30	30	30

* bar length;
** mesh opening, referred to "flatfish" Anon. (2002b);
*** Anon. 2003;
**** no special minimum mesh size was provided for turbot fishing with gill nets; fishermen employed nets with 220 mm mesh opening length in specialized turbot fishing.

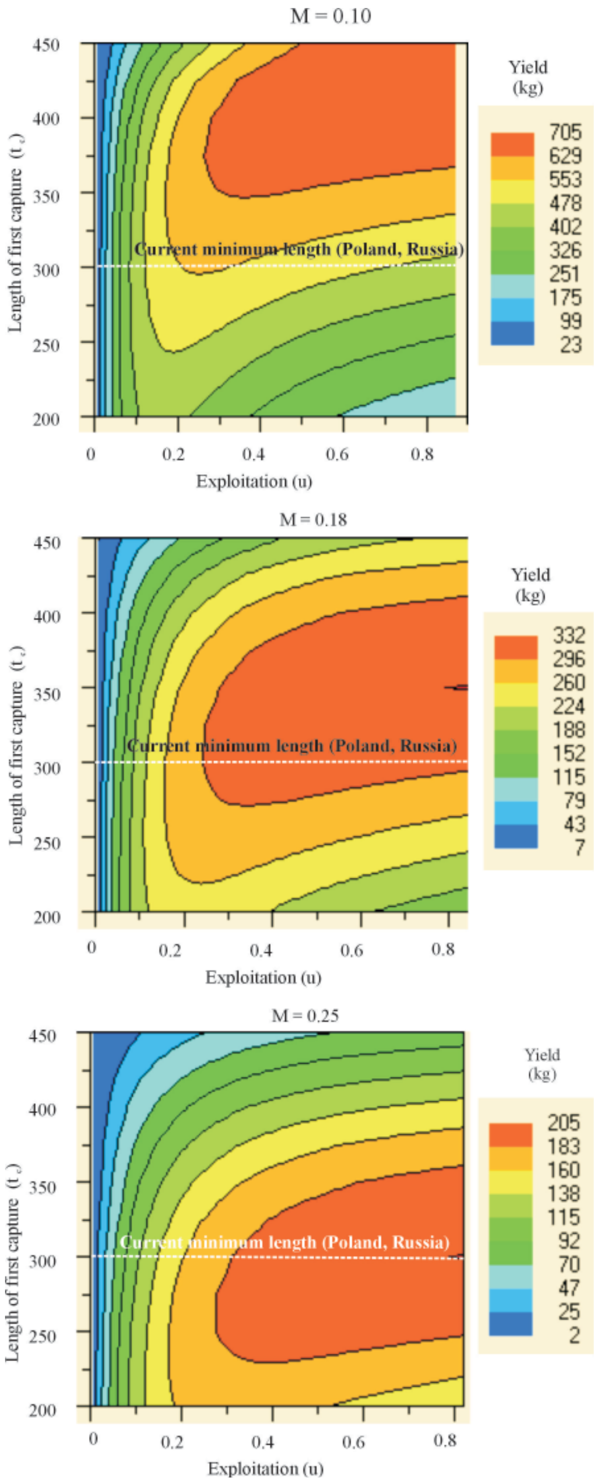
According to current EC regulations, the minimum 120 mm mesh size (mesh bar length of the long diagonal) is binding in flatfish fisheries (Anon. 1997). In the first version of the proposal for a council regulation of fishery resources in the Baltic and Sound (Anon. 2005), the minimum mesh size in gill nets to be used in fishing for turbot ranges from 90 to 157 mm.

In the 1995-1999 period, experimental catches indicated that turbot weighing from 501 to 1000 g occurred in a quantity of 500 specimens, while in the next quinquennium the share of this fish group decreased to one tenth of the former volume and equaled less than 50 specimens. Similar results of fishery intensity were reported by Stankus (2002, 2003). According to his analyses of results since 1995, when specialized turbot fishery was developed, the average length of turbot caught in 1995 exceeded by 2 to 6 cm the average length observed in 2000 (Stankus 2003).

According to the Malkin (1997) theory, the age at which female and male fish reach first maturity is a determinant factor for estimating the biomass volume that can be removed from an exploited stock. Consequently, from 20.4 to 20.7% of the Baltic turbot biomass can be removed annually.

FAST, a computer program created by Slipke and Maceina (2000), was used to evaluate the productivity of the Baltic turbot stock as a function of fish length when they become fully vulnerable to fishing mortality and applied fishing mortality. The results showed that, at a natural mortality coefficient value above 0.15 (annual rate), changes in fishing intensity do not have a significant impact on stock productivity (Fig. 12). Furthermore, the currently binding allowable minimum landing length satisfies the requirements for maintaining the balance between fishing effort and minimum landing length if M is > 0.15.

Fig. 12. Yields of the Baltic turbot in relation to the length of first capture and rate of exploitation.



DISCUSSION

Since the publication of the earliest papers devoted to Baltic fishery (Hensen 1875, Kändler 1944), turbot has been described as one of the species which is being overfished. Interesting as it seems, this species was not mentioned by authors who described Baltic ichthyofauna based on archeological materials (Chełkowski1960, 1965, Rulewicz 1994). It appears in the first decade of the twentieth century in Baltic fishery statistics. The annual amounts of Baltic turbot caught until the 1990s was several hundred tons and did not surpass 400 tons. These reported values, which, in the case of a fish that frequently occurs as by-catch in the fishery of other species, should be acknowledged as minimal. In other words, it has been underreported. Until the 1990s, the fisheries of Poland and other Baltic countries of the former USSR were not interested in catching turbot. In the mid 1990s, annual turbot catches reached 1,200 tons, only to fall to 600 tons in the following years. There has been a decreasing trend in Baltic turbot fishery in recent years, which has been noted mostly in the Lithuanian fishery. Based on the analyses of the available data, the authors acknowledge that this has resulted from the following factors:

– increasing demand for turbot;
– intensive turbot targeted fishery;
– the increased catchability of fishing gears according to Andreev (1998); the shift from gill nets made of polyamide to monofilament fiber increases the respective gear catchability from five to seven times;
– insufficient control of compliance with turbot fishing regulatory measures.

Bearing in mind the characteristics of this species described earlier, it appears that the Baltic turbot stock will not only be unable to sustain the fishery but will be depleted eventually. Based on the review of currently binding regulatory measures for the turbot fishery, it can be surmised that these measures should be sufficient for conserving the exploitable population at least in comparison to the conservation measures obligatory for other Baltic species. Nevertheless, since gill nets catch 90% of the turbot, there is little chance of choosing a mesh size for this gear that would serve as a tool for management of turbot fishing mortality. In view of the current model of Baltic fish species exploitation, an allowable minimum landing length and a closed season should be considered as effective regulatory measures in fishing for turbot. These would not interfere with fishing for other species that contribute a larger catch biomass.

According to the authors, the significance of this species to Baltic fisheries as well as for sustaining the biodiversity of the marine ecosystem requires a separate approach to efforts to sustain populations inhabiting open waters. Perhaps restocking in the manner done for salmon and sea trout should be considered as a one of the most promising methods.

The elimination of the largest top predator from the ichthyocenosis might be of imperceptible significance in an ecosystem untouched by man. However, when stress is caused by the combined effects of coastal water eutrophication, contamination, global climate change, and anthropogenic pressure, this will influence considerably the ability of an ecosystem to maintain equilibrium that oscillates around a multi-year average (Constanza *et al.* 1993).

Kandler (1944) and Stankus (2003) described the biological features of the Baltic turbot as well as the ecology of this species. The turbot inhabiting the Baltic (*Psetta maxima maxima*) and those from the Black Sea (*Psetta maxima maeotica)* are regarded as sub-species (Nielsen 1986). Taking into account the fact that turbot prices are high in comparison with those for salmon or tuna, the scope of knowledge regarding this naturally-occurring species in European water basins cannot be regarded as sufficient for the effective equalization of the exploitation level or stock productivity. Many indicators affect this; the most significant of which is the insufficient precision of turbot catch statistics. In the case of Baltic turbot, this resulted from low consumption that, on the other hand, caused restricted trade with other European countries and meant a lack of fishery interest in this fish. The opening of western European markets spurred an increase in turbot catches in Lithuania, Poland, and the Kaliningrad region of Russia and the specialization of the turbot fishery through the deployment of gill nets with 110 mm mesh bar lengths and above.

The available fishery statistics on Baltic turbot catch are poor and are not a very useful measure for estimating the size of the stock. The comparison of the length composition of turbot caught by Polish coastal fishery in the 1960s and between 1995 and 2003 indicates that the differences in the catch curves are not significant. This is a faulty interpretation since in the 1960s turbot was fished exclusively by nets with bar lengths of 50-60 mm used in flounder fishery and, moreover, trawls were the most frequently used gear and gill nets were used in coastal fishery. The increase in fish under 30 cm Tl in catches partly taken by specialized turbot fishery applying gill nets with a bar length of 110 mm or more demonstrates a shift in fishery pressure towards younger specimens.

The growth rate of Baltic turbot seems to be much slower than that of this species from the North Sea. A four-year-old turbot female from the North Sea reaches an average length of 36 cm. In order to reach this length, a female turbot from the Baltic Sea requires no less than six years, while this figure is an average of eight years according to Kändler (1944). According to Leeuwen and Rijnsdorp (1986), a turbot female from the southern North Sea that has reached an age of four is an average of 45.1 cm Tl. Some turbot researchers maintained that "…the Baltic turbot is a dwarfed form of the North Sea species" (Johanssen 1915, cited in Kändler 1944). The results of analysis indicate that turbot attain 5-9 cm Tl at an average of 7 cm Tl in their first year of life until winter and then attain 9-16 cm Tl at an average of 14.2 cm in the second year (Kändler 1944, Stankus 2003). These results correspond with those reported by Cięglewicz *et al*. (1969) based on back calculations.

Many researchers stress the existence of substantial differences in Baltic turbot growth depending on fish sex and age (Kändler 1944, Stankus 2003). For example, Stankus (2003) stated that the differences in growth between the sexes in the first years of life are insignificant at 0.2-0.3 cm. They become more apparent when the fish reach the third age group, and following this they become more and more apparent. The results of the current analysis confirm these findings.

Analysis of Baltic turbot stock productivity was performed using the Beverton and Holt (1957) formula modified by Slipke and Maceina (2000), which provided an index of yield per recruitment unit as a function of age at first capture, growth, and natural mortality (that which reduced the year-class number independently of fishing operations). The formula makes it possible to fit the age (length) of fish retained by gear to fishing effort in a manner that identifies the maximum biomass that might be removed from the stock at a given level of fishing effort. The results of analysis indicate that the currently binding allowable minimum landing length of 30 cm Tl (Anon. 1997) ensures the optimal utilization of turbot resources.

The authors are aware that the results presented in this paper are based on the acceptance of assumptions that can hardly be confirmed under real conditions. Thus, there might be reservations regarding the acknowledged constant natural mortality indicator value of $M = 0.18$, regardless of fish age or constant level of recruitment. In this context, ignoring the difference of sex growth rate seems to be justified. The use of growth equation parameter 'average' values would signify the acceptance of fishing a considerable quantity of female year-class prior to them having reached maximum biomass production.

The problem remains of adjusting gill net bar mesh size so that a minimum of undersized fish are caught. The trawl selectivity effect, considering the insignificant amount of turbot caught using this gear and the relatively low mortality of discarded fish, may be considered of little importance. Observations of the length of turbot caught in gill nets indicated that with the 110 mm bar mesh size fish that are 30 cm Tl had an 80% chance of being caught and that about 20% of the catch was comprised of undersized fish. In the light of the current findings, it is clear that the current EC regulation of a 120 mm minimum mesh size (Anon. 1997) is far from what might be considered satisfactory. In other words, the minimum mesh size should be at least over 200 mm in order to ensure the protection of Baltic turbot resources. Acknowledging the fact that over a certain time turbot constitutes a considerable part of the gill net catch targeted at flounder, very strict compliance with measures forbidding the retention of undersized turbot in any fishing is required. Perhaps an extension of the closed season should be considered.

Although setting annual quotas of total allowable catch (TAC) might be the solution, this requires detailed information on the numbers of fish caught each year from the exploited stock and the generation to which they belong. The lack of precise statistics on turbot annual catch, which is fragmented by the various fishing gears applied, makes it impossible to produce reliable, scientifically sound resource assessments. Consequently, it is impossible to elaborate TAC as a binding regulation for the turbot fishery in the Baltic Sea.

Just like in other European seas, one of the relevant questions regarding Baltic turbot remains unanswered: why is this abundance of this species still insignificant despite the considerable absolute fecundity of its females? Based on the analyses of results concerning 700 series of relations between spawning males and the breeding population supplement, Myers *et al*. (1999) contended that the maximum fish reproduction rate is relatively constant within species and that there is relatively little variation among species. Froese and Luna (2004) confirm that there is no basis for the assumption that high fish fecundity confers high resilience to exploitation. The acceptance of such an approach is hazardous to fish that are highly fecund (Sadowy 2001), and this should be stressed regarding issues related to turbot resource management.

The results of maturation and age distribution observed in Lithuanian coastal waters were reported by Stankus (2002) and Repečka *et al.* (1998). The existence of eggs differing in developmental phase inside single female ovaries were observed among spawning turbot females in Gulf of Gdańsk waters in 2004. This may indicate batch spawning. Analogical observations were made by Stankus (2001), who reported that turbot spawns in three portions. According to Stankus (2001), the maximum absolute female fecundity of Baltic turbot exceeds seven million eggs. Observations made in the Kaliningrad region in the 2000-2004 period provided evidence of the dependence of turbot female maturation on the water temperature in May. Kändler (1944) emphasized that water temperature determined the place and period of Baltic turbot spawning as well as later larval development. This same author reported that in June of 1936, a year he acknowledged as 'warm', the Baltic Sea water surface layer in the Kołobrzeg region on the slope leading to the Bornholm Basin was heated to a depth of 10 meters. The surface temperature reached 18.1ºC while at a depth of 10 m the temperature was 13.6ºC. This was reflected in the spawning success expressed in the number the turbot larvae present in the water layer and subsequent relative year-class abundance.

The high mortality rate of turbot larvae, one of possible causes of low population levels in comparison with the large number of eggs produced by females, was observed by several researchers. The percentage of dead turbot eggs in samples collected in the Black Sea was 80-90% (Popowa 1972). The mortality of turbot belonging to age group 0 from September to May in the Celtic Sea was 25% per month (Jones 1973). According to Iles and Beverton (1991), the daily indicator of natural mortality in this region was $M = 0.025$.

Zuev and Melnikova (2002), who compared the proposed coefficient by Derzavin of commercial return defined as the number of individuals (in %) generated by a spawning stock which reached first sexual maturity versus the mean absolute fecundity of the species, concluded that the Black Sea turbot occupies the extreme position of the lowest commercial return coefficient value at the highest fecundity index.

The comparison of values of lipids, glycogen, and polyunsaturated fatty acids in gonads, embryos and larvae of turbot, horse mackerel, round goby, and bunt-snouted mullet inhabiting the Black Sea revealed that turbot spawn is the least abundant in lipids and glycogen (Chepurnov and Tkachenko 1982). According to these authors, the low level of energetic material reserves are compensated for by the large number of eggs spawned.

When considering turbot adjustment to changing environmental conditions, it should be remembered that this species inhabits seas of different salinity. The optimal salinity conditions for egg fertilization in the Black Sea was 16-20 PSU and for hatching – 17-18 PSU (Bityukova and Tkachenko 1998) at an egg incubation temperature of 11-15ºC (Zaika and Makarova 1983).

Turbot larvae switch to exogenous food on day four post hatch, and the lack of food during a 24-36-hour period causes irreversible changes in the digestive system (Chepurnov *et al*. 1986). Having finished their metamorphosis, juvenile stage turbot migrate to sedentary water layers. According to Kändler (1944), turbot specimens that had attained the typical body shape for the species (15-23 mm) resided in coastal bottom waters.

The turbot features described above indicate just how dependent recruitment success is on abiotic environmental factors that affect embryonic and larval development. It remains unknown what impact predators which feed on turbot eggs and larvae have on this species. These facts confirm the validity of the hypothesis put forth by Froese and

Luna (2004) that the high fecundity of the teleost fish developed to counterbalance high mortality in the larval period.

The analyses of the results of the fisheries impact on the Baltic Sea turbot stock indicate that this species exhibits a downward trend despite fisherman compliance with compulsory preservation measures. The consequences of not taking action to sustain turbot resources may be that in the next five to eight years this species will have lost its significant in Baltic fisheries. It should be kept in mind that, in addition to its economic importance, the turbot is a predatory fish (Wyche and Shackley 1986, Aarnio *et al*. 1996, Virbickas 2000), and as such plays a crucial role in the energy flow of the Baltic Sea ecosystem. This indicates the necessity of developing substantial measures to protect and sustain Baltic turbot resources at a level that permits maintaining specialized turbot coastal fishery. This requires prompt action that results in:

– improving the current system of collecting fish catch statistics so that the by-catch of all species is recorded;

– raising the minimum mesh size in gill nets used in turbot fishery to at least 200 mm;

– identifying turbot spawning grounds and banning fisheries in some of them in order to implement a pilot program for Baltic turbot protection;

– creating a turbot stocking material cultivation center with the intention of stabilizing the Baltic turbot resource level.

The authors would like to emphasize that, in addition to 'traditional' measures aimed at sustaining the turbot fishery, the key to ensuring the stability of this population is stocking with reared hatch.

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Annex 1. Polish length-age key for the Baltic turbot (combined data for 1998 - 2003)

Length (cm)	Age group																												Total
	II		III		IV		V		VI		VII		VIII		IX		X		XI		XII		XIII		XIV		
	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	
16	1	1																									2
17	1		3																								4
18	1		1																								2
19			3		2																						5
20			9		2																						11
21	2		4	1	7																						14
22	2		8	1	5		3																				19
23			9	3	3	2	5		2																		24
24			10	1	15	4	5																				35
25			7		13	2	8																				30
26				2	6	7	8	1	3																		27
27			1		5	8	10	1	7		1																33
28					4	6	2	2	4	1	2																21
29					2	2	8	6	6	5																	29
30						3		11	1	13	4	1															33
31						5	3	7	2	10	2	6		1													36
32						2		4	5	11	1	11		1													35
33							1	3	2	13		16	2	1	1												39
34							1	3	1	10	1	15	2	5		1											39
35								1		8		17		4	1	2											33
36										2		16		4		1											23
37									2	5		14		10		2											33
38										4		6		11		3	1										25
39										1				12		2		1									16
40												2		4		4		3									13
41												2		6		4		4		1							17
42												2		3		4		2		1							12
43														1		2		3									6
44														1		2		4									7
45												1				1		1		2							5
46																1		1									2
47																		2									2
48																1		2									3
49																		2				1					3
50																		1				2					3
51																		1		1						1	3
52																							1				1
53																			1								1
Total	8		63		105		93		118		120		68		32		28		6		3		1		1		646
Females	1		8		41		39		83		109		64		30		27		6		3		1		1		413
Males	7		55		64		54		35		11		4		2		1										233

Annex 2. Russian length-age key for the Baltic turbot, data from 1995-2004

Length class (cm)	Age group																		Total
	II		III		IV		V		VI		VII		VIII		IX		X+		
	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	
18	1	1		1															3
19	1	2																	3
20		4		1															5
21		3				3		1											7
22		4	1	2		1													8
23		1	2	9		1													13
24		1		15		1		1		1									19
25			8	17		10		2											37
26			9	33		25		1											68
27			31	6	5	57		8		1									108
28			31	2	5	50		15		3									106
29			44		15	34	2	25		8									128
30			61		31	10	5	30		24		5							166
31			39		79	7	6	29		17	1	4		2					184
32			28	1	117		28	17	1	16		3							211
33			9		139	2	53	6	8	14		3		2				1	237
34			2		101	1	100	6	21	4	1	3							239
35			2		52		115	1	66	2	8	1	2	1		2			252
36					22		139	3	81	2	20	2			1	3		1	274
37					6		52	1	120	2	44	2	5	2				1	235
38					7		31		82	3	89		10	2		1		1	226
39					2		12		40	1	106		32						193
40					1		4		23		91		50		1				170
41							4		8		70		62		3				147
42							3		22		38		65		7				135
43							1		1		22		66		10		2		102
44							1		7		8		72		14		2		104
45							2		3				47		25		9		86
46							2		3		1		30		37		13		86
47											1		7		41		32		81
48											1		3		23		24		51
49													1		9		47		57
50													1		3		32		36
51							1				1						23		25
52																	35		35
53													1				22		23
54																	19		19
55													1				12		13
56																	4		4
57																	3		3
58																	3		3
59																	3		3
60																	1		1
61																	1		1
Total	2	16	267	87	582	202	561	146	486	98	502	23	455	9	174	6	287	4	3907
	18		354		784		707		584		525		464		180		291		

Long-term trends in the macrozoobenthos of the Vistula Lagoon, southeastern Baltic Sea. Species composition and biomass distribution

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Abstract. Drastic changes of the macrobenthos species composition was noted in the Vistula Lagoon during the past century. Some species of bottom invertebrates, previously widely distributed in the lagoon, are either now rare or have disappeared. The biomass of zoobenthos increased several-fold this period. Until the end of the 1980s, the zoobenthos of the open waters of the lagoon did not exceed 20-36 g · m^{-2}, but it increased to 81-103 g · m^{-2} in the 1990s. Simultaneously, the thickness of the inhabited sediment layers increased from 10 to 20-25 cm. The spatial patterns of benthos biomass, both of vertical distribution in the sediment layer and horizontal distribution, changed significantly, mainly due to the introduction of the North American spionid polychaete *Marenzelleria* cf. *viridis*. The relations of these trends to hydrological changes, eutrophication, and bioinvasion are discussed.

Key words: Vistula Lagoon, zoobenthos, species composition, biomass distribution, eutrophication, *Marenzelleria* cf. *viridis.*

INTRODUCTION

The Vistula Lagoon is a shallow, brackish-water basin divided between Poland and Russia. The economic and environmental value of it is rather significant for both countries as it is fished intensively and many resorts are located on its shores, and both activities provide employment for local residents. The ecological sustainability of this shallow lagoon is important for the health of the Baltic Sea ecosystem because it partially protects the open Baltic from nutrient loading and pollution.

The Vistula Lagoon macrobenthos has been well studied since the nineteenth century. Prior to the man-made changes to lagoon hydrology which were carried out in 1914-1916, the zoobenthos was described by Mendthal (1889), Seligo (1895), and Vanhöffen (1911). Immediately following this important hydrological change, detailed papers were published by the German authors Vanhöffen (1917), Willer (1925), Riech (1926), and Lundbek (1935). These studies concentrated primarily on faunistic or ecological problems and did not include quantitative data. Since the mid twentieth century, the macrozoobenthos of the eastern part of the lagoon has been studied by many authors (Aristova 1965a, b, 1973, Krylova and Ten 1992, Rudinskaja 1999, Ezhova 2000, 2002, Ezhova and Pavlenko 2001, Ezhova *et al.*

2004). Although these authors analyzed abundance and biomass distribution and dynamics, in comparison with the works of the German authors, they paid little attention to faunistic problems. Investigations of zoobenthos (Żmudziński 1957, Cywińska and Różańska 1978, Różańska and Cywińska 1983, Żmudziński 1995, 1996, 2000, Żmudziński *et al.* 1996) and some taxonomic groups (Klimowicz 1958, Jażdżewski *et al.* 2004) were also conducted in the western part of the lagoon. However, to date, the numerous zoobenthos data have never been considered jointly for the Polish and Russian parts of the lagoon.

During the past century, the Vistula Lagoon has been exposed to strong and diverse anthropogenic pressure. The aim of the current paper is to trace the development of macrozoobenthos in these changing conditions throughout the Vistula Lagoon during the twentieth century.

STUDY AREA

The Vistula Lagoon is a semi-enclosed shallow coastal basin that is separated from the Gulf of Gdansk by the Vistula Spit (Fig. 1) and is connected with the Baltic Sea by the Baltijsk Strait. The length of the lagoon is 90.7 km, its average depth is 3.1 m, the maximal depth is 5.2 m, and the depth of the dredged ship channel in the northeastern part of the lagoon is 11 m. The area of the lagoon is 861 km^2, of which 473 km^2 belongs to the Russian Federation and 388 km^2 to Poland (Lazarenko and Majewski 1975).

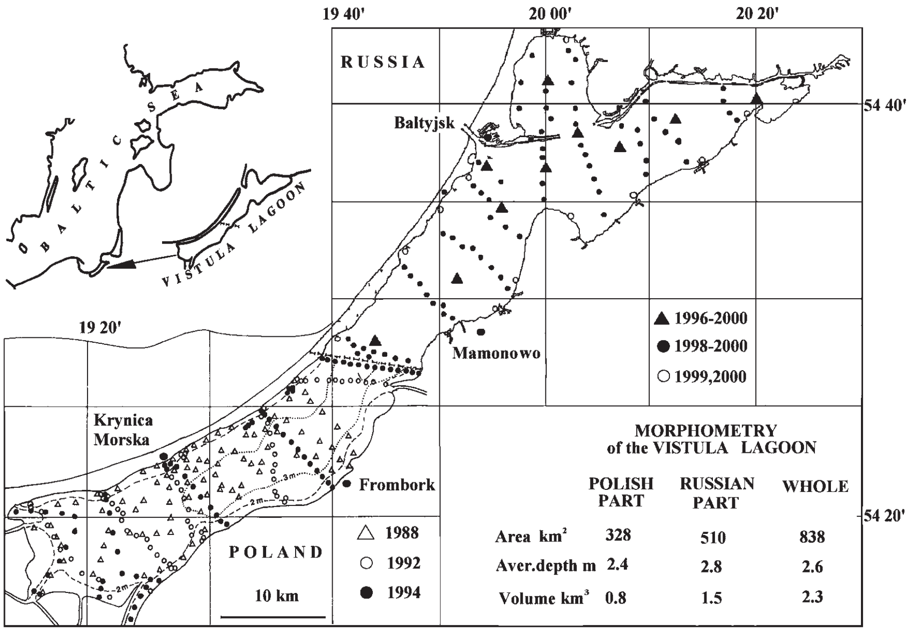

Fig. 1. Location of sampling sites in the Vistula Lagoon.

The hydrological status of the lagoon is defined mainly by the inflow of fresh water from twenty rivers and water exchange with the Baltic Sea. The water exchange through the Baltijsk Strait (37.5 $km^3 \cdot a^{-1}$) constitutes about 88.5% of the lagoon water balance, whereas the combined river runoff (3.6 $km^3 \cdot a^{-1}$) accounts for only 8.5%; thus, the Vistula Lagoon is an estuarine basin with a prevalence of marine factors. The Pregel River discharges into the lagoon about 41% of the total freshwater runoff, while the Nogat and Pasłęka rivers, the largest on the Polish side, contribute 18% and 14%, respectively (Lazarenko and Majewski 1975).

Water salinity is rather low and varies from 0.1 (min) to 10 (max) PSU at an average of 4-5.5 PSU. The spatial distribution of salinity in the lagoon depends on hydrometeorological conditions, winds, and, particularly, on sea level fluctuation. The usual average salinity of the various parts of the lagoon is as follows: southern part – about 1.5 PSU (Renk *et al.* 2001); central part – 2.9 PSU; eastern part – 3.8 PSU; in the vicinity of the Baltijsk Strait – 4.0-5.0 PSU (Lazarenko and Majewski 1975). There is a seasonal increase in salinity in the lagoon from the spring to late fall.

The average concentration of suspended matter (30.7 $mg \cdot l^{-1}$) (Chechko 2002) is approximately ten-times higher than in the open Baltic Sea. Due to the shallow depth, the high concentration of suspended matter, and frequent wave and wind re-suspension events, water transparency is low and varies from 1.0 to 0.2 m. It is distinctly lower in the western part of the lagoon where it does not exceed 0.5 m during summer.

The lagoon bottom is soft, with the prevalence of silt, and sand is distributed mostly along the shores. The organic matter content of the sediments can reach 10% in silt and mud bottoms (Blazchishin 1998). This basin is a eutrophic water body, and strong phytoplankton blooms are typical during the summer when biomass can reach 26 $g \cdot m^{-2}$ (Aleksandrov 2003). Primary production is at least twice as high as that in the southern Baltic Sea with average annual primary production estimated at 303.8 $gC \cdot m^{-2} \cdot year^{-1}$ (Renk *et al.* 2001).

MATERIALS AND METHODS

Samples from the western (Polish) part of the lagoon were collected from May to December 1952-1954 and in mid summer in 1988, 1992, and 1994. Benthic macrofauna was sampled at from 73 to 92 stations, the majority of which were located along several profiles across the lagoon (Fig. 1). In 1988, bottom samples were collected within a depth range of 1.0-3.2 m, while in 1992 and 1994 it was widened to include the coastal zone and the deepest areas of the Polish part of the lagoon (Tab. 1).

Table 1. Characteristics of macrobenthic data for the western part of the Vistula Lagoon

Sampling period	Number of stations	Depth range (m)	Sampler type
30 VII - 31 VIII 1988	92	1.0 - 3.2	Ekman-Birge grab
3 - 23 VIII 1992	76	0.4 - 4.7	Core sampler
20 - 26 VII 1994	73	0.5 - 5.0	Core sampler

During the 1950s and in 1988, sampling in Poland was done with an Ekman-Birge sampler (230 cm^2). In the 1990s, a square sampler (50 cm^2) designed by Żmudziński was used (Żmudziński 1996). The device was pressed into the bottom using a rotating aluminum tube made of several sections. This enabled collecting bottom cores measuring 20-35 cm and 10-15 cm from muddy and sandy-muddy substrates, respectively. The Ekman-Birge sampler only penetrated to depths of 10-15 cm and 5 cm, respectively.

The field data from the eastern (Russian) part of the lagoon were collected by Aristova (Aristova 1965a, b, 1973) in the 1955-1967 period and by the Group for Hydrobiology AB IORAS in the 1996-2000 period. Sampling was conducted at the same ten stations during monthly or seasonal monitoring cruises. In the 1998-2000 period, sampling was also conducted twice during summer at from 40 to 79 stations in order to study the biomass distribution and bottom assemblages. Additionally, zoobenthos was collected at twelve coastal stations in the summers of 1999 and 2000. The locations of sampling sites are presented in Fig. 1.

In the 1996-2000 period, samples from the eastern part of the lagoon were collected with a Petersen grab (1/40 m^2, 10-15 cm penetration depth; three replicate samples per station). Since 1998, a DAK-100 box corer (Bakanov 1979) (1/100 m^2, 10-20 cm penetration depth, from three to five replicate samples at each station) and a geological tube (Ø 62 mm, penetration depth – 55 cm, five replicates at each station) have been used. Just as the Żmudziński corer, both of these samplers permit collecting long sediment cores. The use of the benthic corers or geological tube instead of grabs became necessary after the introduction of the burrowing American polychaete *Marenzelleria* cf. *viridis* since, under the new conditions, the Ekman-Birge and Petersen grabs both underestimated benthic biomass several-fold.

Samples were sieved through 0.4 mm mesh and preserved in 4% neutral formalin. Biomass is presented as the so-called formalin wet weight. The animals were identified, counted, and weighed in the laboratory. Generally, identification was conducted to the species level, excluding Oligochaeta and Chironomidae. Chironomids from the Żmudziński and Aristova samples were identified to the species level, while in other materials the species composition of this group was not determined. Due to this, the current authors were not able to trace the biodiversity of chironomids in the lagoon the during study period. However, they decided to include the Żmudziński and Aristova data on chironomids in the list of species (Table 2) as this information might be useful for other researchers.

The positive identification of the polychaete *M.* cf. *viridis* is usually problematic due to the indistinct taxonomy of the genus and the few external characters suitable for determination. Since this polychaete is a dominant benthic species in the lagoon, particular attention was paid to identifying it correctly. Specimens of Marenzelleria were identified and later verified using the key to species in Sikorski and Buzhinskaya (1998) and Sikorski and Bick (2004). In samples from the Vistula Lagoon, only *Marenzelleria* cf. *viridis* is present. Species *M.* cf. *wireni* and *M. neglecta* sp. nov., which are both very similar to *M.* cf. *viridis*, were not found.

The specific names of animals were taken from the five-volume *Wykaz zwierząt Polski* (Anon. 1990-1997).

Table 2. Benthic macrofauna species composition in the Vistula Lagoon during the twentieth century. Data for 1917-1926 from Vanhoffen (1917), Willer (1925), and Riech (1926); 1950s-1960s from Zmudzinski (1957), Klimowicz (1957), and Aristova (1973); 1988-2000 – authors' own data

Taxa	Region/Years				
	Whole lagoon	Russian part	Polish part	Russian part	Polish part
	1917-1926	1958-1967	1952-1954	1996-2000	1988-1994
1	2	3	4	5	6
Hydrozoa					
Cordylophora caspia (Pallas)	xx	xx	xx	xx	xx
Turbellaria indet.	xx	xx	-	xx	-
Nemertini indet.	xx	xx	xx	xx	-
Oligochaeta indet.	xxx	xxx	xxx	xxx	xxx
Hirudinea					
Helobdella stagnalis (L.)	x	x	x	-	-
Erpobdella octoculata (L.)	-	x	x	-	-
Glossiphonia complanata (L.)	x	x	x	-	-
Glossiphonia heteroclita (L.)	-	x	x	-	-
Piscicola geometra (L.)	x	x	x	x	-
Total (varia)	**7**	**9**	**8**	**5**	**2**
Polychaeta					
Marenzelleria viridis (Verrill)	-	-	-	xxx	xxx
Nereis diversicolor O.F.Muller	xx	xx	-*	xx	-
Manayunkia aestuarina (Bourne)	-	-	-	x	-
Alkmaria romijni Horst	-	-	-	x	-
Streblospio benedicti (Webster)	-	-	-	x	-
Total (Polychaeta)	**1**	**1**	**0**	**5**	**1**
Mollusca, Bivalvia					
Anodonta anatina (L.)	x	-	x	-	-
Anodonta complanata Rossm.	x	x	-	-	-
Anodonta cygnea (L.)	x	-	x	-	x
Dreissena polymorpha (Pallas)	x	x	xx	x*	xx
Unio crassus Philipsson	x	-	x	-	-
Unio pictorum (L.)	x	-	x	-	x
Unio tumidus Philipsson	x	-	-	-	-
Sphaerium rivicola (Lamarck)	x	-	-**	-	-
Sphaerium corneum (L.)	x	x	x**	-	-
Sphaerium solidum (Normand)	x	x	-**	-	-
Pisidium amnicum (O.F.Muller)	x	x	-	-	-
Cerastoderma glaucum (Poiret)	x	x	-	x	-
Macoma balthica (L.)	x	x	-	x	-
Mya arenaria L.	x	x	-	x	-
Mytilus edulis trossulus (L.)	x	x	x*	x	-
Total (Bivalvia)	**15**	**9**	**7**	**5**	**3**
Mollusca, Gastropoda					
Acroloxus lacustris (L.)	x	-	x**	-	-
Bithynia leachi (Sheppard)	x	-	x**	-	-
Bithynia tentaculata (L.)	x	x	x**	x	x
Lymnaea auricularia (L.)	x	x	x**	-	-
Lymnaea ovata (Drap.)	x	x	x**	x	-
Lymnaea palustris (O.F.Muller)	x	x	x**	-	-
Lymnaea peregra (O.F.Muller)	-	x	-	-	-
Lymnaea stagnalis (L.)	x	x	x**	-	-
Litoglyphus naticoides (Pfeiffer)	x	-	x	-	-
Physa fontinalis (L.)	x	x	x**	-	-

Table 2 continued

1	2	3	4	5	6
Planorbarius corneus (L.)	x	x	x**	-	x
Planorbis planorbis (L.)	x	-	x**	-	-
Anisus vortex (L.)	x	-	x**	-	-
Anisus vorticulus (Troschel)	x	-	-**	-	-
Anisus spirorbis (L.)	x	-	-**	-	-
Anisus leucustomus (Millet)	x	-	-**	-	-
Anisus septemgyratus (Rossm.)	x	-	-**	-	-
Anisus contortus (L.)	-	-	x**	-	-
Gyraulus albus (O.F.Muller)	x	-	-	-	-
Theodoxus fluviatilis (L.)	x	x	x**	x	-
Valvata piscinalis (O.F.Muller)	x	x	x**	x	x
Viviparus viviparus (L.)	x	-	x	-	-
Viviparus contectus (Millet)	x	-	x**	-	-
Hydrobia ventrosa (Montagu)	x	x	-	xxx	-
Potamopyrgus antipodarum (Gray)	x	x	x	xxx	-
Eubranchus pallidus (Alder and Hancock)	x	x	-	x	-
Total (Gastropoda)	**24**	**13**	**18**	**7**	**3**
Crustacea					
*Asellus aquaticus (*L.)	x	x	x	-	-
Balanus improvisus Darwin	x	x	x	x	-
Jaera albifrons Leach	x	-	x	x	-
Sphaeroma rugicauda (Leach)	x	-	-	x	-
Gammarus zaddachi Sexton	x	x	x	x	x
Gammarus locusta (L.)	x	x	-	x	-
Gammarus pulex (L.)	x	-	-	-	-
Gammarus lacustris G.O.Sars	-	-	-	x	-
Gammarus salinus Spooner	-	x	-	x	x
Gammarus duebeni Lilljeborg	-	-	x***	xx	x***
Gammarus oceanicus Segerstråle	-	-	-	xx	-
Gammarus tigrinus Sexton	-	-	-	x	x***
Pontogammarus robustoides (G.O.Sars)	-	-	-	xx	x***
Dikerogammarus haemobaphes (Eichwald)	-	-	-	x	x***
Obesogammarus crassus (G.O.Sars)	-	-	-	-	x***
Orchestia cavimana Heller	-	x	x	-	x
Apocorophium lacustre (Vanhöffen)	x	x	x	x	-
Chelicorophium curvispinum (*G.O.*Sars)	x	x	-	x	-
Corophium volutator (Pallas)	-	x	x	-	-
*Crangon crangon (*L.)	-	x	x	x	-
Rhithropanopeus harrisii (Gould)	-	xx	xx	xx	xx
Neomysis integer (Leach)	x	x	xx	x	xx
Total (Crustacea)	**10**	**12**	**11**	**17**	**10**
Insecta, larvae					
Procladius Skuse		xx	xxx	xxx	xxx
Tanypus Kratzi		x	x	-	x
Chironomus plumosus (L.)		xxx	xxx	xxx	xxx
Dicrotendipes gr. nervosus (Staeg.)		x	xx	-	x
Dicrotendipes tritomus (Kieffer)		x	-	-	x
Polypedilum gr. nubeculosum (Meig.)		x	xx	-	x
*Glyptotendipes gr. gripekoveni (*Kieffer)		x	x	-	xx
Cryptochironomus gr. defectus Kieffer		x	xx	-	xx
Microchironomus gr. tener (Kieffer)		x	xx	x	xx
Cladotanytarsus gr. mancus (Walk.)		x	xx	-	xx
Cricotopus gr. sylvestris (Fabricius)		x	x	-	x
Chironomidae indet.	xxx			x	
Total (without Chironomidae)	**57**	**44**	**44**	**39**	**19**

*occasional, single findings, **data from Klimowicz (1957), ***recorded in 1998-2000, data from Jażdżewski *et al.,* 2002

RESULTS

Species composition and frequency of occurrence

The current study did not confirm the wide distribution or sometimes even the presence in the lagoon of many of the species reported previously by Vanhöffen (1911, 1917), Willer (1925), Riech (1926), Żmudziński (1957), or Aristova (1973). This included twenty-nine mollusc species, several oligochaetes, several crustaceans, leeches, and others, while many species that had not been recorded earlier, appeared in the lagoon.

Only twelve mollusc species were recorded in the eastern part of the lagoon and seven in the western part during the 1990s, which was in marked contrast with Riech (1926) (37 species), Aristova (1973) (22), Żmudziński (1957), and Klimowicz (1958) (both 26 species) (Table 2). All of the molluscs, which were fairly abundant until the 1950s and 1960s, are now rare or absent and are only represented by freshwater species. There are only three *bivalves* and three gastropods, mostly marine or brackish-water. *Macoma balthica*, *Mya arenaria, Mytilus edulis, Potamopyrgus antipodarum, Hydrobia ventrosa*, and *Theodoxus fluviatilis* are the species that are constantly present now in the lagoon.

In the last decade, only five freshwater mollusc species were noted in the eastern part of the lagoon usually in the vicinity of river mouths. These rare findings were always of single specimens and can be considered as unrepresentative of the lagoon. In the western part of the lagoon where salinity is generally lower, several freshwater molluscs have been found to date (*Bithynia tentaculata, Planorbarius corneus, Dreissena polymorpha, Unio pictorum, Anodonta cygnea*), but their distribution is very local. All of the species, except *D. polymorpha*, were found at single stations usually on the coast. Both the abundance and frequency of mollusks are much lower than in the first half of the century and the 1950s.

Two brackish-water gastropods, *P. antipodarum* and *H. ventrosa*, became very abundant in the 1996-2000 period with a frequency of occurrence of almost 100%. H. ventrosa has never been noted in the western part of the lagoon due to unfavorable salinity conditions, while *P. antipodarum* was rather frequent in the 1950s, but was not recorded in the following years in the Polish part of the lagoon (Cywińska and Różańska 1978; Różańska and Cywińska 1983; current authors' data from 1988-1994). Although historical data for the Polish part of the lagoon are not available, it would seem very likely that *P. antipodarum* could have appeared after 1994 in areas where it was recorded formerly due to the previously mentioned progressive development of hydrobiid populations that began in the eastern part of the lagoon in 1992 (Rudinskaya 1999; Ezhova *et al*. 2004).

The species composition of crustaceans also has changed significantly since the early twentieth century, but the number of species in this group increased in contrast to molluscs (Table 2), and, in the 1990s, fourteen species occurred in the lagoon in comparison to eight in the first decades of the century. The amphipods *Apocorophium lacustre* and *Corophium volutator* disappeared from the bottom communities in the western part of the lagoon. In the eastern part, *A. lacustre* and *Chelicorophium curvispinum* were noted rarely the 1996--2000 period, and *C. volutator* was not recorded at all.

In contrast to corofiids, the gammarid species are currently represented much more diversely than earlier. Ten species were recorded in the 1990s, while Vanhöffen (1917) and Rieh (1926) noted only two (Table 2). Among them are four non-native species and the common Baltic gammarids *Gammarus duebeni*, *G. locusta*, *G. zaddachi*, *G. salinus*, *G. oceanicus*, and *G. lacustris*. While *G. duebeni*, *G. zaddachi*, and *G. salinus* occur throughout the lagoon, the remaining species were found only in the eastern part. These three species were firstly recorded in the Vistula Lagoon in the current authors' samples in the 1996-2000 period.

Three alien gammarid species were noted in Russian samples in 1999-2000: these were the Ponto-Caspian species *Dikerogammarus haemobaphes* and *Pontogammarus robustoides*, and *Gammarus tigrinus* of North American origin. These species were absent in the authors' 1992-1994 samples from the Polish part of the lagoon. These three alien species and another Ponto-Caspian gammarid, *Obesogammarus crassus*, were first recorded in the Polish waters of the Vistula Lagoon somewhat later in 1998-2000 (Jażdżewski and Konopacka 2000; Jażdżewski *et al.* 2002). In the most freshwater southwestern areas of the lagoon near the deltaic region of the Vistula River, invasive species predominate, especially *G. tigrinus* and *O. crassus.* The share of native *G. duebeni* in the gammarid communities increases northeastwards along the Vistula Spit. In the eastern (Russian) part of the lagoon, *G. duebeni* is one of the most frequent species at 70%, together with native *G. oceanicus*, and the invasive *P. robustoides*, while *G. locusta, G. zaddachi,* and *D. haemobaphes* can be regarded as rather common. The native *G. salinus* and *G. lacustris* along with the alien *G. tigrinus* were rare. The authors' own data and that from the literature (Jażdżewski *et al.* 2004) led to the conclusion that populations of non-native gammarids had established themselves successfully by the end of the twentieth century and that some now dominate native gammarids in some locations.

The number of polychaete species increased during the century. Until the end of the 1980s, the only polychaete recorded in the Vistula Lagoon was Nereis diversicolor. During the late 1990s, five polychaete species were constantly recorded in the lagoon. One of these newcomers, the North American Atlantic species *Marenzelleria* cf. *viridis*, appeared in 1988 (Żmudziński *et al*. 1996) and now occurs throughout the lagoon, including the more freshwater western areas. Another three species, *Alkmaria romijni*, *Streblospio benedicti,* and *Manayunkia aestuarina* (according to determinations by Khlebovich), have been registered since 1996. The distribution of all polychaete species, excluding *M.* cf. *viridis*, is restricted to the eastern part of the lagoon mostly in areas where the influence of saline waters is key and the near-bottom salinity is usually not less 3-4 PSU. *Streblospio benedicti* and *Manayunkia aestuarina* inhabit coastal marine areas near the lagoon entrance, but *Alkmaria romijni* was not recorded in the Russian marine coastal zone. All these species of polychaetes were certainly absent from the lagoon at least in the first three decades of the twentieth century and until the 1950s and 1960s, when very detailed taxonomic studies of zoobenthos were conducted (Vanhöffen 1917, Rieh 1928, Aristova 1973).

The current observations from the 1998-2000 period indicate that the list of oligochaetes is now only comprised of eleven species (determinations by Finogenova), as opposed to the eighteen noted previously. However, there is no comparable data for the whole basin that would serve for an analysis of the actual changes in the species composition of this group.

In general, the species diversity of macrobenthos has decreased in comparison with that of the first half of the century (Fig. 2), even though the number of alien species in-

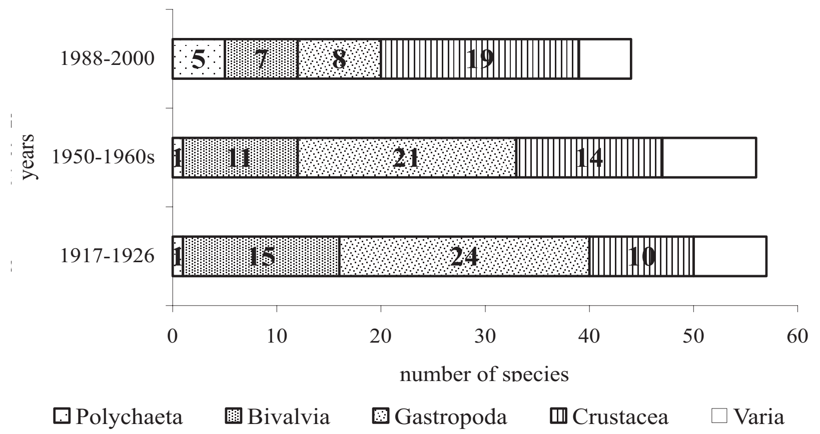

Fig. 2. Species diversity of macrobenthos in the Vistula Lagoon throughout the twentieth century.

creased remarkably during this period. This phenomenon was noted in both the western part of the lagoon where the water is less saline as well as in the eastern brackish-water part, but this decrease was especially sharp in the former. The most evident changes were observed in the taxonomic groups of molluscs, crustaceans, and polychaetes.

Biomass spatial and temporal variability

Patterns of biomass spatial distribution

The biomass of the macrobenthos in the lagoon is presently distributed in a manner which is generally consistent with the pattern described by previous investigators (Aristova 1965a, Żmudziński 1957, Cywińska and Różańska 1978; Krylova and Ten 1992). The lowest biomass is found in the northeastern part of the lagoon near the mouth of the Pregel River (0.36-15.6 g · m^{-2}), while the most productive zone is located in close proximity to the Baltijsk Strait and to the southwest of it (70-452 g · m^{-2}) as well as in the southwestern sector of the lagoon (170-1137 g · m^{-2}) (Fig. 3). The rather large areas beyond these regions are currently characterized by a benthic biomass of approximately 100 g · m^{-2}, while until the end of the 1980s these values did not exceed 20 g · m^{-2}. Maximal and minimal values of benthos biomass were observed in these same regions from the 1950s to the 1980s (Aristova 1965a, Żmudziński 1957, Różańska and Cywińska 1983, Krylova and Ten 1992). The bivalve *M. balthica* dominated the benthic biomass in the most saline area near the strait and *Dreissena polymorpha* was the dominant in the southwestern freshwater region. *M. balthica* is now almost absent in the lagoon, occurring only very locally in the immediate proximity of the Baltijsk Strait. *D. polymorpha* is still abundant at several locations in the southwestern region (especially near Kąty Rybackie) where it forms dense communities and dominates in terms of both biomass (Fig. 3) and abundance. However, in the region adjacent to the Baltijsk Strait, the invading polychaete

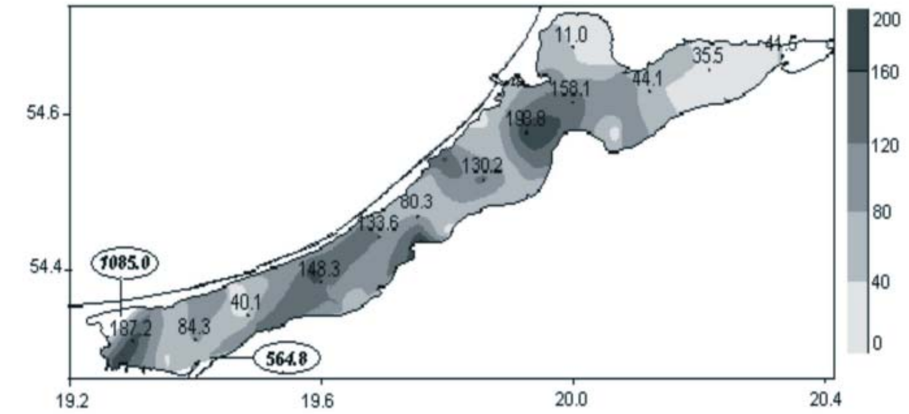

Fig.3. Distribution of the average summer biomass of macrobenthos in the Vistula Lagoon after the introduction *Marenzelleria* cf. *viridis* (1990s). Numbers on the map indicate biomass values at the stations along the longitudinal transect of the lagoon. Numbers in circles indicate biomass in the local areas of *Dreissena polymorpha* dominance.

M. cf. *viridis* is now the primary contributor to the high benthic biomass. It also contributes approximately 50% of the biomass in the southwestern, freshwater near-shore areas. The share of *M.* cf. *viridis* in the benthos biomass can reach 25-30% even in *D. polymorpha* communities.

Vertical distribution of biomass in sediments

An important aspect of benthos distribution during the 1990s was the increase in the thickness of the inhabited sediment layers. Ezhova and Chechko (2003) have shown that more than a half of the eastern part of the lagoon is covered by sediments which are bioturbated to a depth up to 20-25 cm and more. On average, 54% of the total biomass is confined to the upper 10-cm layer, while the deeper horizons are responsible for 46%. This pattern of benthos vertical distribution in the sediments is absolutely different from those observed during the 1950s and 1960s (Aristova, 1965b) when 83% of the biomass was restricted to the 0-10 cm layer and only 17% to the deeper horizons (Fig. 4). This phenomenon is strongly connected with the appearance of the alien species M. cf. viridis (Żmudziński 1996, 2000, Ezhova 2000). This polychaete can penetrate into sediments rather deeply. In the area of interest, the maximal depth of its penetration was 32 cm. The authors recorded

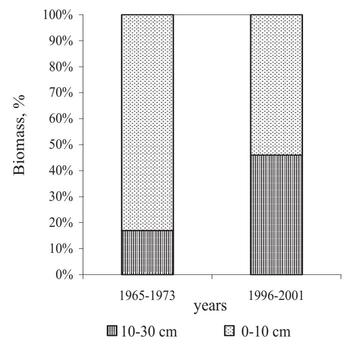

Fig. 4. Vertical distributions of benthos biomass in the bottom sediments before and after the introduction of *Marenzelleria* cf. *viridis* in the Vistula Lagoon.

the maximum of M. cf. viridis biomass, as well as the maximum of biomass of the entire benthos, between 10 and 15 cm in the sediments. This contrasts with the 1960s when the maximum benthic biomass was never noted deeper than the 0-10 cm horizon (Aristova 1965b).

Long-term changes of benthic biomass

Benthic macrofauna biomass values increased several-fold in the 1990s in comparison with that in the 1950s. Until the 1980s, the average macrozoobenthos biomass in the open lagoon was from 10 to 37 g · m^{-2}. In the 1950s, the average benthos biomass in the open lagoon was calculated at 36.7 g · m^{-2} for the western part (Żmudziński, 1957) and approximately 20 g · m^{-2} for the eastern part based on the Aristova data (1965a, b). Towards the end of the 1980s it was 22 and 31 g·m^{-2} for these parts of the lagoon, respectively. Thus, the macrobenthos biomass varied only slightly until 1988. Following the introduction of the North American spionid polychaete *M.* cf. *viridis* in the 1990s and its wide distribution in the lagoon, the benthic biomass increased up to 81-103 g · m^{-2} with a tendency to increase over the 1992-1994 period.

The lack of quantitative data from the early 1990s for the eastern part of the lagoon and the end of this decade for the western part did not permit calculating precise average values for these periods on the scale of the whole lagoon. However, the trend presented in Fig. 5 is well supported by existing qualitative and quantitative data and should closely reflect reality in the open waters of the Vistula Lagoon.

The ratio between the components of biomass has also been rather constant over the decades. Until the end of the 1980s, *Chironomidae* larvae dominated (especially *Chironomus plumosus* L.) with *Oligochaeta* as the co-dominant. In more saline eastern areas, chironomids constituted 80% and oligochaetes approximately 11% of the benthic biomass (Krylova and Ten 1992), while in the western part the contribution of these groups was 48-86% and 9-47%, correspondingly. Only in the southwestern coastal areas did Mollusca (especially *D. polymorpha*) predominate, although *Chironomidae* larvae were co-dominant (41%). The increase in biomass in the 1990s was accompanied by a fundamental change

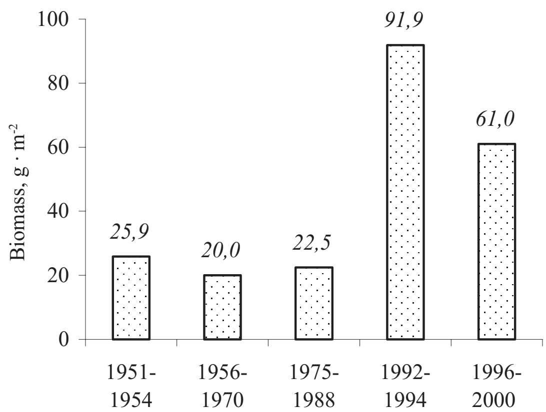

Fig. 5. Average annual biomass of macrobenthos in the open waters of the Vistula Lagoon in 1951-2000. Biomass was calculated as follows: 1951-1954 according to Murina (1951), Żmudziński (1957); 1956-1970 according to Aristova (1973); 1975-1988 according to Krylova and Ten (1992), Cywińska and Różańska (1978), Różańska and Cywińska (1983);1992-1994 and 1996-2000 according to the authors' own data.

in the biomass components. In the early 1990s, the share of chironomids and oligochaetes fell several times and *Polychaeta* comprised over 95-97% of the biomass in the open waters. However, an increase of *Oligochaeta* and *Chironomidae* biomass was noted in subsequent years (Rudinskaya 1999, the current authors' data). In 1996-2000, it varied from 1 to 19 g · m^{-2} at an average of 7 g · m^{-2}, depending on biotope and season. Although this value is somewhat less than the average annual of 11.6 g · m^{-2}, it is within the range of fluctuation. Thus, the current data do not indicate that there was a sharp decrease in *Chironomidae* biomass.

The share of hydrobiid gastropods, mainly *P. antipodarum*„ increased. Previously, it was observed in the northeastern part of the Vistula Lagoon and its average biomass was 1.3-3.2 g · m^{-2} (Aristova 1973). According to data from the 1996-2000 period, this group is very abundant in the lagoon. It is found throughout the central lagoon, and the biomass of this group varies from 0.04 g · m^{-2} in the parts of the lagoon with the lowest salinity and up to 29 g · m^{-2} in areas of maximum abundance.

DISCUSSION

The decrease of species diversity in the Vistula Lagoon during the last century was most apparent in the mollusc group. The decline in species richness was not as sharp in the middle of the century, but by its close the total number of mollusc species occurring in the lagoon had decreased five-fold. The impoverishment of the benthic fauna began in 1914-1916, when the inflows of Vistula and Nogat rivers waters into the Vistula Lagoon were regulated and cut off, which resulted in an average salinity increase to 3-5 PSU. A fundamental change of species composition in the bottom assemblages, distribution, and quantitative development of species populations occurred due to an insignificant rise in salinity

Since 1914 and the 1920s, the mass death or reduction of populations of many mollusk species occurred as a consequence of the influence of higher salinity. In approximately 70% of the lagoon, the range of salinity fluctuation and the average value of it could no longer support the survival of freshwater stenohaline and partially freshwater euryhaline animals. Thick layers of shells from *Anodont*, *Unio*, *Dreissena*, *Lymnaea*, *Viviparu*, *Planorbis*, *Valvata*, *Bythinia*, and others formed mostly in the eastern part of the Vistula Lagoon, (Lundbeck 1928, 1935). At present, these shell layers are deposited under several dozen centimeters of silt and are of varied thickness and represent different species assemblages. Thick horizons of unionid shells were noted in silt sediments deeper than 30 cm along the Vistula Spit (present authors' data), while layers of *Dreissena* and gastropod shell beds were noted in the northeast of the lagoon. The formation of these shell horizons began in 1914-1916 (Blazchishin 1998).

The comparison of Polish and Russian data with the records of German authors (Table 2) revealed the dynamics of species composition change in the malacofauna. The stenohaline species of freshwater origin disappeared simultaneously throughout the lagoon; these included *Unio tumidus*, *Sphaerium rivicola*, *Anisus spirorbis*, *A. vorticulus*, *A. leukostomus*, *A. septemgyratus*, and *Gyraulus albus*. Many stenohaline species disappeared first in the eastern more saline region, but were recorded as late as the 1950s in the western part of the lagoon, and these included *A. anatina*, *A. cygnea*, *U. crassus*, *U. pictorum*, *Acroloxus lacustris*, *Bithynia leachi*, *Litoglyphus naticoides*, *Planorbis planorbis*, *Anisus fortex*, *Anisus contortus*, *Viviparus viviparus*, and *V. contectus*. Thus, while some species disappeared, others sharply constricted their area of occurrence to the limits where salinity oscillations did not exceed the specific range of salinity tolerance. This mollusc fauna impoverishment occurred in the first half of the twentieth century. During this period manifestations of eutrophication were insignificant and the only evident reason for the marked decrease in species richness could have been anthropogenic changes in lagoon hydrology.

It is significant that the disappearance or decrease of mollusk populations did not occur at once. All of the freshwater species discussed above were recorded some years after river run-off was halted (Vanhöffen 1917; Willer 1925; Riech 1926). Salinity increased and the fluctuation range often exceeded the limits of the optimum salinity tolerance range for the most of the freshwater stenohaline and euryhaline species. Inhabiting environmental conditions where factor values are usually far from the physiological optimum does not always result in the immediate death of animals, but it can gradually lead to the decrease of vital parameters for a population and inhibit its reproduction (Khlebovich 1974). This situation initiated the disappearance of the freshwater species complex, while the rate and scale of this process depended on the degree of the euryhalinity of a given species.

Other species, including four leeches, twenty-one mollusks and two crustaceans, disappeared or became very rare from the 1970s to the 1990s. This further impoverishment of macrobernthic fauna is connected mostly with anthropogenic eutrophication. After the 1950s, constantly increasing nutrient loading had a primary impact on the Vistula Lagoon ecosystem. Both Russian and Polish data (Różańska and Więcławski 1978, 1981; Senin 1990, Khlopnikov 1994, Renk *et al.* 2001) confirm hard nutrient loading from the 1960s to the 1980s with the maximum in the late 1980s. Decreases have been noted since the 1990s. The concentration of inorganic nitrogen and phosphorus, the content of organic matter, water oxidation, chlorophyll concentrations, and primary production were at levels that corresponded to highly eutrophicated basins as early as 1974-1975 (Różańska and Wiktor 1978). Water bodies with high trophic status are characterized by increased amounts of organic matter in accessible forms both in the water column and on the bottom surface, as well as by bottom silting and a decrease in water transparency. This set of environmental conditions facilitates the development of a few highly tolerant dominant species and leads to the disappearance of many other species. Usually, species with long life cycles, such as *Unionidae* and *Dreissena* and other mollusks, are very vulnerable under such conditions. On the contrary, animals with short life cycles that produce several generations per year overcome this in eutrophic water bodies. This corresponds well with the marked qualitative and quantitative changes of macrobenthos in the lagoon during the period from the 1960s to the 1980s. While the chironomids and oligochaetes totally predominated the macrobenthos, the large mollusks with a perennial life cycle almost disappeared and only small molluscs, such as *P. antipodarum* and *H. ventrosa*, which breed throughout during the year, could reproduce successfully.

The appearance and sustained development of three small polychaete (*Alkmaria romijni*, *Streblospio benedicti*, *Manayunkia aestuarina*) populations over the last nine years are also probably connected to the increase of the trophic level of the lagoon ecosystem. The successful development of small-sized detritophagous organisms with short life cycles is supported by the extensive pool of organic matter available in easily accessible forms in eutrophic ecosystems (Kondratiev and Koplan-Diks 1988). Some authors consider the appearance of these polychaete species to be an indicator of developing eutrophication. Noji and Noji (1991) emphasized that the mass development of organisms with short generation times that are tolerant of wide ranges in salinity and substrates and with the ability to switch feeding modes, all of which are characteristics of the polychaetes mentioned above, correlates with rising habitat instability, which is normal in disturbed, eutrophic ecosystems.

The impoverishment of lagoon macrofauna and the changing of some environmental characteristics due to eutrophication results in the formation of unsaturated ecological niches. The unintentional introduction of alien species was very successful under such conditions, and currently the share of alien species comprises nearly 27% of the total number of macrobenthos species. The most important alien benthic species in the ecosystem are *M.* cf. *viridis, P. antipodarum*, and *Rhithropanopeus harrisii.* Their contribution to productivity, matter transformation, and habitat modification in the Vistula Lagoon is critical. Some of them, such as *M.* cf. *viridis*, due to its burrowing activity, can effectively modify the bottom environment and influence different sediment transport processes, organic matter transformation, and nutrient fluxes.

In the 1950s, communities of the alien snail *Potamopyrgus antipodarum*, accompanied by *Apocorophium lacustre* and *Corophium volutator*, were a characteristic element of the lagoon benthos (Żmudziński 1957). This community was not found in the following years in the western part of the lagoon (Cywińska and Różańska 1978, Różańska and Cywińska 1983; the authors' own data). The absence of these species in the 1970s (Cywińska and Różańska 1978, Różańska and Cywińska 1983) could have resulted from the low number of stations sampled during investigations. It could also have been related to low salinities in the western part of the lagoon. Brackish-water and marine organisms inhabit this area where conditions are close to the limits of their adaptive capabilities, and any negative impact could have reduced their abundance. Increased nutrients, high organic matter content, and low oxygen content, all of which were typical of this eutrophic area in the 1970s and 1980s, could have been such negative factors (Różańska and Wiktor 1978; Różańska and Więcławski 1981).

In the saline eastern part of the Vistula Lagoon, where salinity conditions are more complimentary for *P. antipodarum,* this gastropod still occurs at present; moreover, it has become extremely abundant (Ezhova and Polunina 1999, Ezhova and Pavlenko 2001, Ezhova 2002). It was noted that *P. antipodarum* and *H. ventrosa* comprised from 20 to 24% of the benthos biomass in the 1992-1996 period (Ezhova *et al.* 2004). Corophiid frequency is also very low in the eastern area and *C. volutator* does not occur there.

The authors maintain that the progressive development of the *Potamopyrgus* population and the decrease in corophiid abundance are not only consequences of eutrophication, but that the distribution of the alien polychaete *M.* cf. *viridis* throughout the lagoon is also a factor. It was demonstrated that significant negative correlation exists between this spionid and *C. volutator* (Zettler 1996). *M.* cf. *viridis* negatively impacts corophiids through bioturbation as the polychaete provides spatial portioning on the sediment surface and below it. At the same time, the borrowing activity of *M.* cf. *viridis* can encourage substrate settling (Zettler 1996) by loosening it and facilitating its colonization by small organisms like hydrobiids.

A significant phenomenon was the increased species diversity of gammarids during the 1990s. Six gammarid species that were new to the lagoon were recorded: four alien species of Ponto-Caspian and North American origin and two autochthonous Baltic species that had not previously been noted in the lagoon. The appearance of native *G. oceanicus* and *G. lacustris* demonstrates that there is some similarity with the three Baltic polychaetes which extended their distribution area from adjacent marine coastal waters to the Vistula Lagoon. Evidently, the lagoon environment currently provides these newcomers with more favorable conditions as regards biotic and abiotic factors. However, the cause of the nearly simultaneous invasion of three Ponto-Caspian species is likely one and the same. Among the hypotheses put forth by Jażdżewski *et al.* (2004) regarding such a trigger, the present authors' consider the climate warming trend to be the most probable facilitator of the widening of the distribution range of Ponto-Caspian invaders. This concurs with recent introductions and the active secondary dispersal of Ponto-Caspian planktonic crustaceans in the Baltic Sea basin and correlates with the increasing ratio of thermophilic species in the coastal Baltic zooplankton in recent years (Polunina, personal communication).

The consideration of the data of the current authors and Jażdżewski *et al.* (2002, 2004) regarding the distribution and occurrence of these four invaders in the Vistula Lagoon allows for the supposition that the appearance of any of them from the east, *i.e.*, through the Baltijsk Strait or from the Curonian Lagoon via the Deima-Pregel route, is less probable. *G. tigrinus* can disperse easily along the sea coast thanks to the high level of euryhalinity. In the event that it had arrived in the lagoon via the Baltijsk Strait, the species abundance would be high in the brackish eastern part. On the contrary, this species is dominant in the Polish part of the lagoon and rare in the Russian part. The second invasive species, *O. crassus*, is abundant in the western part of the lagoon while it is not noted in the Russian part of the basin. As Jażdżewski *et al.* (2004) demonstrated, *D. haemobaphes* invaded the Vistula Lagoon via the Pripet-Bug canal. The route taken by *P. robustoides*, which is rather frequent in the eastern part of the lagoon, is very likely to have been that through the Pregel river. In general, however, all non-native gammarids in the Russian part of the lagoon are more frequent and more abundant along the coast of the Vistula Spit towards the southwest. Northward, the dominance of native gammarid species increases. Thus, in all likelihood, all four gammarid species came to the eastern part of the lagoon from the Polish basin.

In summary, it is evident that clear trends in the state of the macrobenthos of the Vistula Lagoon occurred during the twentieth century. These included:

– a fundamental change in species composition through a reduction in the number of freshwater species and the introduction of alien species;

– an increase in the benthos biomass;

– an increase in sediment depth penetration by zoobenthos due to the invasion of the North American polychaete *M.* cf. *viridis*.

These phenomena are the result of the joint effect of three forcing factors, namely, the artificial change in the lagoon salinity regime in the 1914-1916 period, strong anthropogenic eutrophication, and the invasion of new species. Anthropogenic eutrophication redoubled the fauna impoverishment caused by the salinity increase at the beginning of the twentieth century and provided conditions for the successful naturalization of new species.

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5. Tekst. Objętość maszynopisu prac kategorii 1 nie powinna przekraczać 40 stron, a kategorii 2 – 15 stron. W pracach kategorii 1 i 2 stosuje się tradycyjny podział: 1) wstęp, 2) materiał i metoda badań, 3) wyniki badań, 4) dyskusja, 5) bibliografia. Wyniki pomiarów należy podawać w jednostkach miar przyjętych w systemie metrycznym, a ich skróty – zgodnie z Międzynarodowym Układem Jednostek Miar (SI).
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