

The effect of algal blooms on fish at their inshore nursery grounds in the Gulf of Gdańsk

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ABSTRACT

*Studies of cyanobacterial bloom dynamics show that the highest biomass accumulation of *Nodularia spumigena* is observed in the shallowest area of the Gulf of Gdańsk in summer. In the same region and time, the highest fish abundance is observed. Mostly young individuals of gobies, small sandeel, flounder, three-spine stickleback and young herring occur. In this work we compare how toxic bloom of cyanobacteria influence on number and structure of fish community in coastal zone. The results obtained in our study were rather unexpected. More fish species were caught and the biomass of fish was higher during a bloom than in a month following the sampling (no bloom).*

KEY WORDS

shallow water area, Baltic Sea, fish biomass, toxic blooms, *Nodularia spumigena*

INTRODUCTION

The shallowest waters of the Gulf of Gdańsk play an important role in the ontogeny of many fish species. They are places of spawning, feeding and growing (Skóra, 1993). In the same region, blooms of cyanobacteria are observed every summer. The blooms are mainly composed of *Aphanizomenon flos-aquae* (L.) Ralfs ex Bornet et Flahault, *Nodularia spumigena* Mertens ex Bornet et Flahault and less abundant *Dolichospermum* (*Anabaena*) species. As N₂-fixing microorganisms, they can form large biomass even when dissolved inorganic nitrogen is depleted. Additionally, gas vesicles (aerotopes) make them buoyant and facilitate formation of surface accumulates when weather is calm. Summer blooms of *N. spumigena* are regular over the whole Baltic Sea. Based on satellite measurements, it has been possible to determine that cyanobacteria can cover over 100 000 km² of the Baltic Proper (Kahru *et al.*, 2007). In general, cyanobacterial blooms begin to develop in the open water, but the waves and wind can push the scums towards the coastal zone.

Large-scale development of cyanobacteria in the Baltic Sea has a negative impact on the ecosystem. Blue-green algae may affect the Baltic Sea ecosystem directly or indirectly. High-biomass blooms can prevent sunlight penetration into the water column, which contributes to the loss of water clarity and consequently suppresses aquatic macrophytes, and negatively affects invertebrate and fish habitats. Furthermore, effects of cyanobacterial blooms include visual and mechanical interference in feeding and changes in the pelagic environment, from transparent to turbid conditions offering a refuge for smaller prey (Karjalainen *et al.*, 2007). Bacterial decomposition of dying blooms may lead to oxygen depletion (hypoxia and anoxia), and subsequent fish kills (Paerl & Otten, 2013). These organisms can produce a number of metabolites through their secondary metabolism, some of which can be extremely harmful. The blooms can be particularly harmful when the nodularin (NOD)-producing species, *N. spumigena*, dominates. NOD is a cyclic pentapeptide with hepatotoxicity associated with liver

accumulation, where it inhibits hepatocellular serine: threonine protein phosphatase-1 and -2A (PP1 and PP2A, respectively). The effect and NOD accumulation in aquatic animals from different trophic levels were documented, both in field studies and in laboratory experiments (Landsberg, 2002; Karjalainen, 2005; Karjalainen *et al.*, 2007; Pääkkönen *et al.* 2008; Persson *et al.*, 2011; El-Shehawy & Gorokhova, 2013; Mazur-Marzec *et al.*, 2015).

In consequence, cyanobacteria-induced fish mortalities are caused by either poisoning due to toxin release, anoxia or a combination of both. However, not every cyanobacterial bloom causes a fish kill. The released toxins should normally dissolve rapidly within a water body and therefore, the concentration of toxins usually remains below the lethal level. Moreover, fish may migrate within a given water body and thus avoid an exposure to a toxic cyanobacterial bloom (Ernst, 2008 and references therein).

It is commonly considered that fish, as mobile organisms, leave a region contaminated with harmful substances.

Therefore, it can be presumed that algal blooms reduce the number of fish in the study area.

The objective of our study was to compare the species composition of ichthyofauna in a shallow zone of the Gulf of Gdańsk during *N. spumigena* bloom and in the period with no algal bloom.

MATERIAL AND METHODS

The study was carried out in the Gulf of Gdańsk (the Southern Baltic Sea) on 29/30 June and 29/30 July 2011 in the Gdynia-Redłowo area (54°27,979' N, 18°34,219' E) (Figure 1).

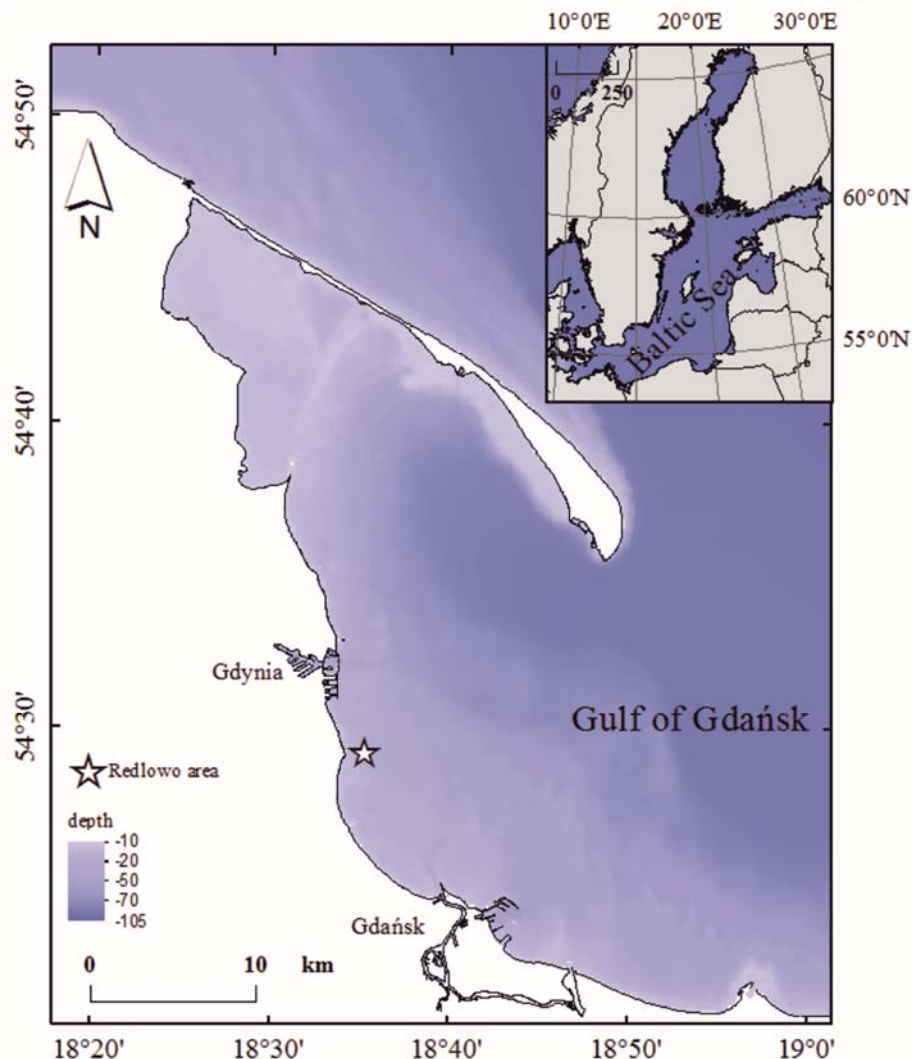


Fig. 1. Study area in the Redłowo territory in the western part of the Gulf of Gdańsk.

Every year, this area is affected by summer blooms of the toxic cyanobacterium *N. spumigena*. Gillnetting was carried out to study fish composition and abundance. In addition, the cyanobacteria composition was investigated. Samples were collected during a cruise of Oceanograf II. Each experimental set up consisted on 3 gillnets (Nordic Nets 64) multi-mesh and each gillnet composed of 7 different mesh sizes, ranging from 10 mm to 50 mm, knot to knot (Table 1). Each time, experimental setups were deployed parallel to the shoreline, at a depth of 4 m and 6 m and were left for 24 hours.

Taxonomy, total length and wet weight were determined for each fish. The abundance and biomass of each fish species were determined in each sampling and net set separately.

Basic physicochemical water parameters (temperature, salinity, oxygen, pH) were measured in situ with a WTW Multi 340i meter (measure each 1 m from surface to bottom). Surface water samples for qualitative and quantitative analyses of phytoplankton were collected into 200 ml bottles during deployment and collection of gillnets. Water samples were preserved with Lugol's solution (1%) and stored under cool and dark conditions. The biomass of cyanobacteria was determined according to the Utermöhl method (Edler, 1979) using an inverted microscope (Nikon TMS, Tokyo, Japan) with 200× and 400× magnification. The size of the counting chambers (2 or 10 ml) and the sedimentation time (8 or 24 h) depended on the abundance of phytoplankton. The bloom samples (30 June 2016) were diluted 10 times. The biomass of cyanobacteria was calculated from cell size and shape using appropriate geometric formulas proposed by Olenina *et al.* (2006).

Table 1. Gillnet (Nordic Nets 64) parameters (CEN. 2005; Psuty 2011)

No.	Panel order in the net	Mesh size [mm]	Line thickness [mm]	Panel length [m]
1	1	30	0.15-0.16	30
2	2	17	0.14-0.16	30
3	3	40	0.16-0.20	30
4	4	10	0.12-0.14	10
5	5	22	0.14-0.16	30
6	6	50	0.20	30
7	7	25	0.14-0.16	30
Additional information:				
	Total net length	[m]	190	
	Total net high	[m]	1.8	
	Net colour	green		

Statistical analyses

Differences between sampling sets were analysed with Factor Analysis (PCA, without rotation) and Cluster Analysis (Ward's, 1-Pearson's r).

Cluster Analysis (Ward's, 1-Pearson's r) was chosen as Ward's method is generally known as effective nevertheless produces relatively small groups and we want to group items according to variance present in all investigated material. Factor Analysis (PCA, without rotation) was chosen to check if it's possible to basing on reduction of number of variables find factors explaining generally observed changes in fish community. As a rule factors different when presented in data.

All analyses were completed with Statsoft Statistica v. 12 software.

RESULTS

In 2011, only a short episode of the toxic *N. spumigena* bloom (6.27×10^7 100 μm filaments dm^{-3} , 1,531 $\mu\text{g dm}^{-3}$) was recorded in week 26 (29/30 June) (Table 2). One month later, i.e. at the end of July, *N. spumigena* and nodularin were not detected in the phytoplankton samples.

Table 2. Water temperature [$^{\circ}\text{C}$], salinity, abundance [100 μm filaments dm^{-3}] and biomass [$\mu\text{g dm}^{-3}$] of *N. spumigena* during fish sampling. Concentration of nodularin (NOD) after Mazur-Marzec *et al.* 2013 and unpublished data.

Date	Nets	Cyanobacteria bloom	Salinity	Water temperature	<i>N. spumigena</i>		NOD [$\mu\text{g dm}^{-3}$]
					Abundance	Biomass	
29 June	exposure	+	6.9	18.2	6.27×10^7	19,345	1.1
30 June	collect	+	7.0	21.4	5.32×10^9	1,531 488	1 236
29 July	exposure	-	6.5	19.3	n.d.	n.d.	n.d.
30 July	collect	-	6.5	19.1	n.d.	n.d.	n.d.

No significant differences were determined for any of the measured environmental parameters (salinity, water temperature, oxygen concentration, pH). Water temperature variability between June and July sampling was less than 1°C , salinity less than 0.5, and pH - 0.4 (Table 2). Oxygen concentration during sampling was close to 100 %, in the inshore area of the Gulf of Gdańsk, it is typical situation almost during all year.

A total of 458 fish belonging to 11 species, were captured (Table 3).

Table 3. Abundance of fish species in samplings (p-predator, n-not predator)

Species		Bloom		No bloom	
		depth 4 m	depth 6 m	depth 4 m	depth 6 m
<i>Platycthis flesus</i>	n	x	x		x
<i>Scophthalmus maximus</i>	n	x	x		
<i>Hyperoplus lanceolatus</i>	n	x	x	x	x
<i>Clupea harengus</i>	n	x	x	x	x
<i>Sprattus sprattus</i>	n			x	x
<i>Osmerus eperlanus</i>	n		x		x
<i>Neogobius melanostomus</i>	n	x	x	x	x
<i>Perca fluviatilis</i>	p	x	x	x	x
<i>Gadus morhua</i>	p	x	x		
<i>Myoxocephalus scorpius</i>	p	x	x		
<i>Sander lucioperca</i>	p			x	x

During June sampling, 49 fishes with a total weight of 4,823.6 g were caught at a depth of 4 m. They were represented by 8 fish species (Figure 2), the most abundant of which were perch (41%) and round goby (27%), and they contributed (in terms of weight) 17% and 0.3% to the total fish biomass, respectively. Smaller fish were more common in this sampling. Cod represented 54% of the fish biomass and 4% of the total fish count in the sampling.

In the sampling at 6 m depth, 148 fishes with a total weight of 12,048.8 g, represented by 9 fish species, were caught (Figure 2). Perch (43%) and round goby (35%) were most abundant.

Flounder (52%) dominated in terms of the biomass (Figure 2). Three predator fish species, i.e. cod, perch and sculpin, were observed in these samplings.

Altogether 261 fishes with a total weight of 1,395.1 g, represented by 8 fish species, were caught in July samplings. During 4 m depth sampling, 37 fishes with a total weight of 1,530.4 g, represented by 6 fish species, were collected (Table 1). Round goby was represented by the largest number of specimens and contributed 57% to the total catch (in terms of the total number of individuals) and 41% to the total fish biomass (Figure 2).

During the 6 m depth sampling (the total number of 224 fishes and total weight of 12,423.7 g), sprat and pikeperch were caught in addition to fish species present in the 4 m sampling (Figure 2).

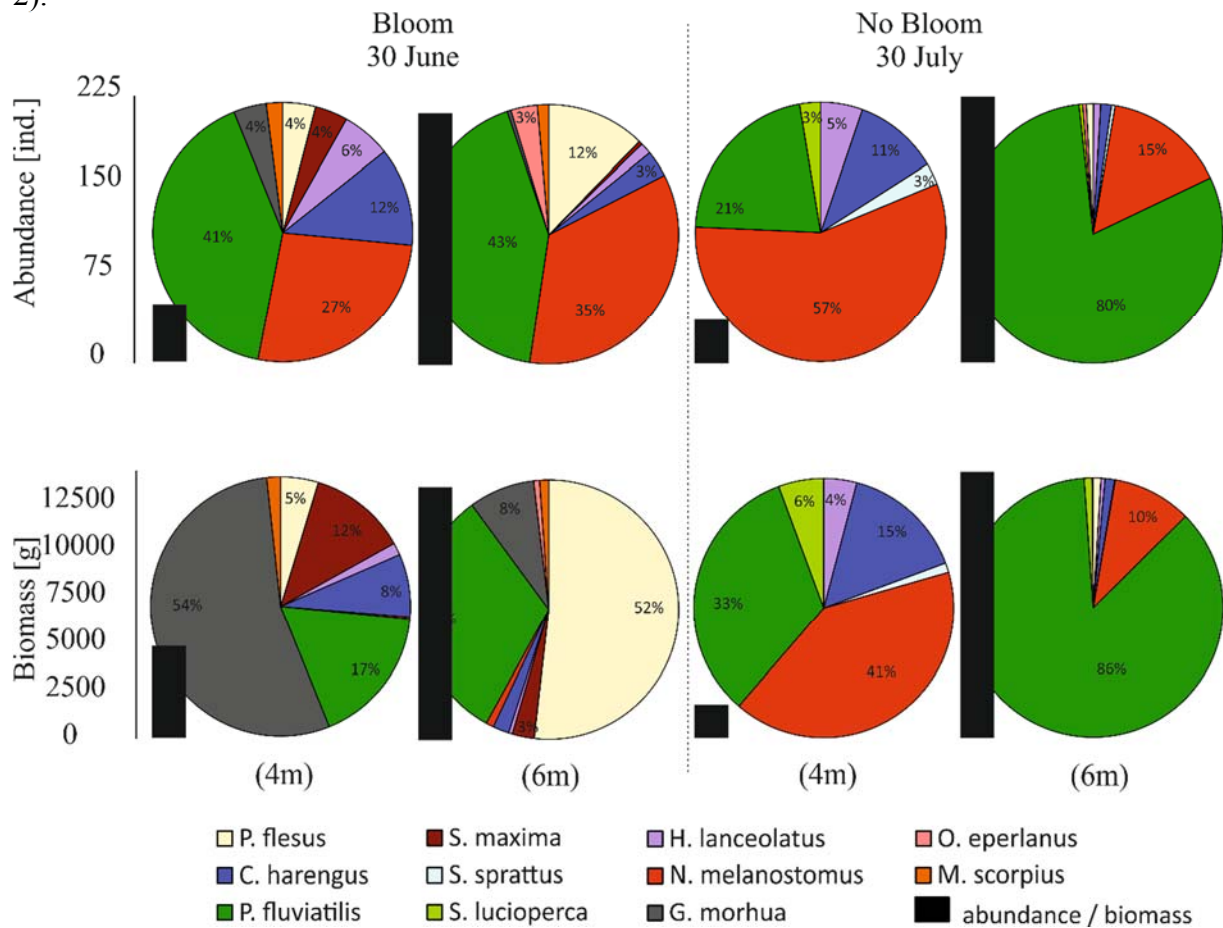


Fig. 2. Fish abundance, biomass and species composition in samplings during and after cyanobacterial bloom.

Two predator species were observed in July samplings – pikeperch and perch (Table 3).

The similarity between the samples was checked by cluster analysis. The resulting dendrogram separated the sampled species into two groups (Figure 3). There are large differences in the fish structure in June (bloom) and July (no bloom) samplings (Figure 3). The date of sampling was more significant than the depth of sampling set exposure.

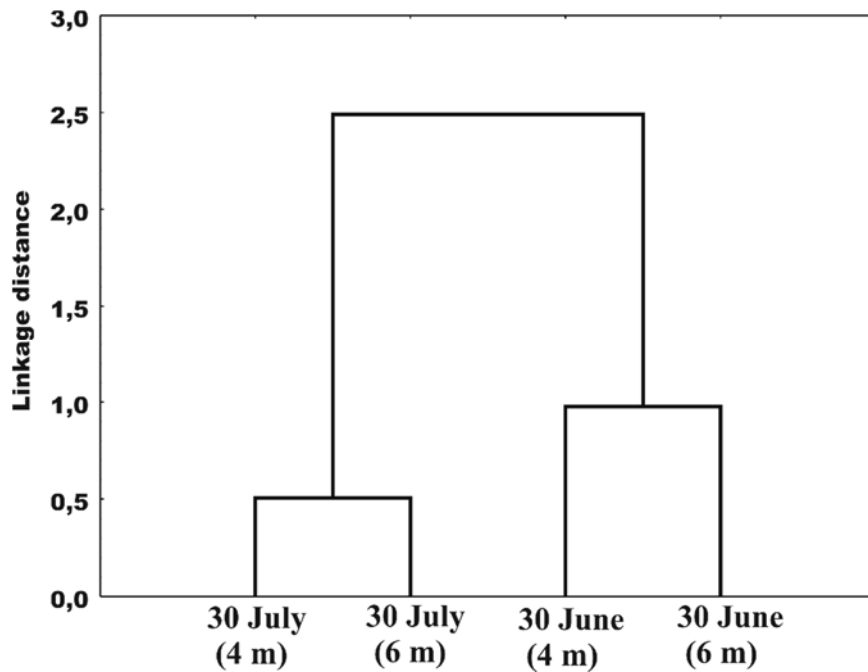
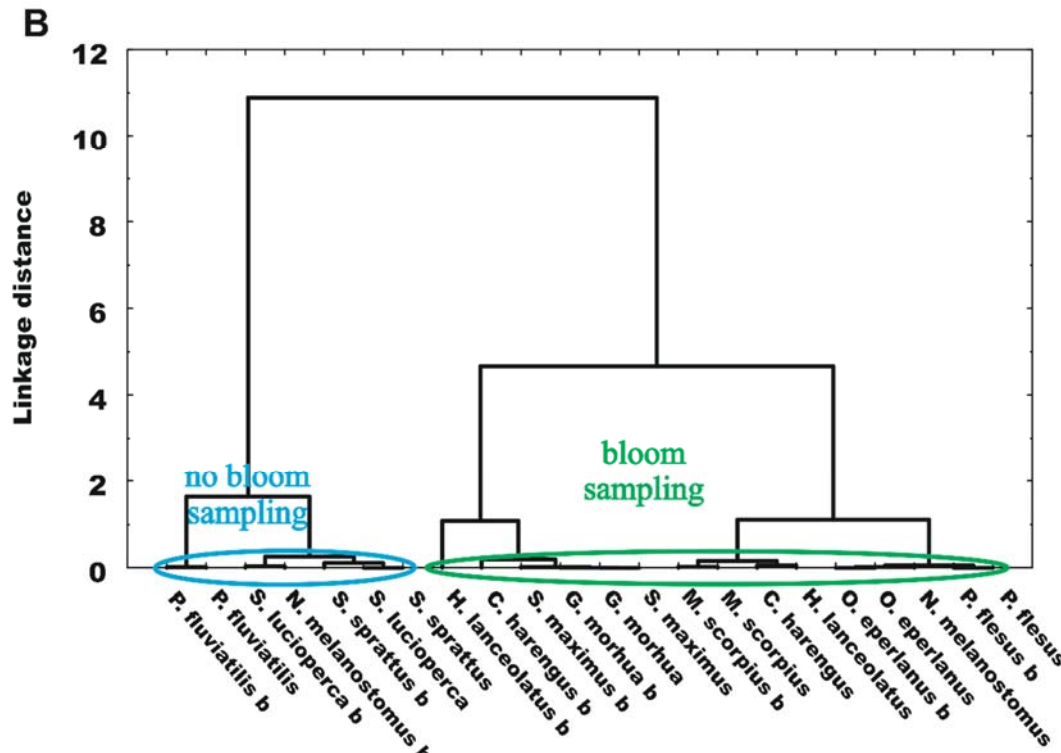
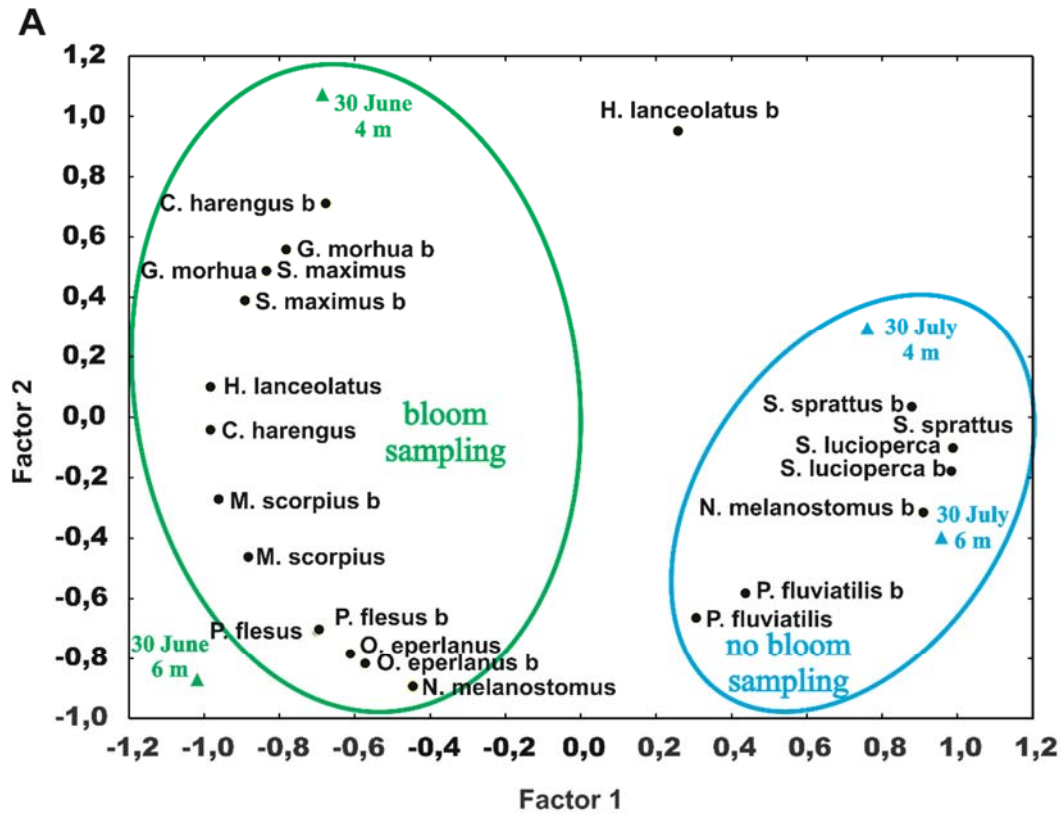


Fig. 3. Similarity between gillnet samplings (Ward's, 1-Pearson's r).

In the factor analysis, two groups of fish were recognized (Figure 4). Factor 1 divides the fish into two groups according to time of sampling. Factor 2 divides the fish mostly according to the main depth of occurrence.

Group 1 includes fish with higher abundance and biomass during bloom samplings, Group 2 includes fish (sprat, pikeperch, perch) correlated with no bloom samplings.



▲ sampling time and depth ● scientific name - fish number ● scientific name + b - fish biomass

Fig. 4. Statistical similarities between fish assemblages present during sampling (the number and biomass), A – PCA, without rotation: factor 1 – 62.10%, factor 2 – 32.10%; B – Ward's, 1-Pearson's r

DISCUSSION

In 2011 a short episode of the toxic *N. spumigena* bloom was observed. One month later *N. spumigena* and nodularin were not detected in the phytoplankton samples. No significant differences were determined for any of the measured environmental parameters (salinity, water temperature, oxygen concentration, pH) during samplings. We have investigated, in shallow water areas of the Gulf of Gdańsk, fish communities since 1993 and algal blooms since 1997. Due to heterogenic characteristic of blooms in this area it was first time we were able to check fish structure, composition, biomass and abundance during *N. spumigena* bloom. In our studies, we were able to check how the occurrence of *N. spumigena* blooms affects the fish structure and abundance in the coastal area of the Gulf of Gdańsk. This is the first study related to the species composition and abundance of fish occurring in the initial phase of cyanobacteria blooms in the inshore zone.

Fish distribution in natural ecosystems during toxic cyanobacterial blooms has been studied in the last decade (Ernst *et al.*, 2007; Sotton *et al.*, 2011). The response of fish to toxic cyanobacteria may vary depending on fish and blue-green algae species, as well as the level of toxicity (Baganz *et al.*, 2004; Ernst *et al.*, 2007; Godlewska *et al.*, 2015). Changes in the behaviour of fish in response to contact with toxic blue-green algae are probably complex and may depend both on the fish species and cyanobacteria species composition, as well as the intensity of blooms and their toxicity (Baganz *et al.*, 2004; Ernst *et al.*, 2007). Toxic blooms may affect swimming activity of fish (Baganz *et al.*, 2004), their feeding, reproduction and survival (Karjalainen *et al.*, 2007).

The first research in Poland on the number of fish during cyanobacterial blooms was conducted by Godlewska *et al.* (2015) in the shallow Sulejów reservoir. The number of fish was determined using hydroacoustic methods. Changes in the dominant fish species were observed due to the bloom intensity in the study area. Fish were less abundant in the area of the bloom, although not totally absent, when a low cyanobacteria biomass was detected, the fish biomass was unpredictable and could have both very small and very high values (Godlewska *et al.* 2015).

Shallow inshore waters of the Gulf of Gdańsk are mostly inhabited by marine fish species (Skóra, 2000; unpublished data from coastal fish monitoring). In one-year studies of fish composition in the Redłowo area, thirteen marine, three freshwater and one anadromous species were identified. The most common fish species in summer include fresh water (i.e. perch *P. fluviatilis*, pike-perch *S. lucioperca*, three-spined stickleback *G. aculeatus*) and marine (i.e. flounder *P. flesus*, round goby *N. melanostomus* and sandeel *A. tobianus*) organisms (Koszarowski unpublished; unpublished data from coastal fish monitoring). In samplings conducted in 2011, altogether 11 fish species were identified, including eight marine and three freshwater species. According to previous investigation fish species composition, during the bloom sampling, was typical for the coastal area of the Gulf of Gdańsk in summer.

In 2011, only a short episode of toxic *N. spumigena* was recorded on 29-30 June in coastal waters in the Gulf of Gdańsk. But large-scale blooms of this species were observed in July and August in the central and northern Baltic (ICES, 2012). The peak of *N. spumigena* biomass and the highest concentration of nodularin (1,236 $\mu\text{g dm}^3$) was determined in the surface bloom samples collected on the 30th of June (Mazur-Marzec *et al.*, 2013).

During the conducted study, we were able to correlate the time of bloom occurrence with the net setting. It enabled us to observe differences in the species composition, abundance and biomass of fish during June (bloom) and July (no bloom in the water column) samplings. Fish, as mobile organisms, are expected to escape from the region where toxic algal blooms occur. According to Sotton *et al.* (2011), however, whitefish (*Coregonus lavaretus*) was found in the same water layer of perialpine Lake Bourget as the bloom of *Planktothrix rubescens*. Based on statistical analysis, we were able to distinguish two groups of fish species, i.e. species not

affected by bloom and fishes avoiding the bloom region. In both groups, pelagic and benthic fish species were present.

It is likely that fish could respond to environmental conditions altered by algal blooms. Data on pH during algal blooms from coastal areas are sparse, but pH may increase above 9 and become as high as 9.25 and 9.75 in enclosed water bodies. In the Baltic Sea, pH values above 9.3 were measured during such blooms (Møgelhøj *et al.*, 2006 and references therein). In the Gulf of Gdańsk, pH values above 9.1 were measured during *N. spumigena* blooms in 2008-2010 (unpublished data). During our investigations pH was from 8.7 to 9.1 during bloom sampling and 8.2 month later.

Environmental conditions (e.g. oxygen concentration, pH), during samplings, within the bloom and a month later were typical for shallow waters of the Gulf of Gdansk during summer and suitable for the fish. We observed that the total number of fish and their biomass were higher, as well as predators were more abundant within the bloom sampling. In June, we observed the occurrence of large individuals of cod, sculpin, flounder and turbot, usually present in a shallow water area during the night (Lizińska, 2002). Similar regularity was observed during studies of diel shifts conducted by Yeoh *et al.* (2016) in nearshore estuaries where small pelagic fishes were more abundant during the day, while larger pelagic, benthic-pelagic and demersal fishes prevail at night.

We assume that those differences were probably caused by a decrease in visibility. The increase in organic matter raises the level of eutrophication and encourages algal blooms, which leads to further increase in turbidity (Utne-Palm, 2002). Several studies reveal that relatively small increases in turbidity may change the structure of fish communities affecting, for example, their migratory activity, increase the use of open water by young fish and reduce the predation risk (Utne-Palm, 2002 and references therein). Turbidity may also affect prey-searching behaviour (Utne-Palm, 2002). In the laboratory study, foraging of cod on evasive prey in highly turbid water was attributed to the use of both visual and non-visual senses (Meager *et al.*, 2005).

In addition to fish predation, we can expect bird predation, especially by cormorants and herons which are abundant in this region and pose a direct threat to fish in shallow water areas during the day (Bzoma, 2004; Jakubas & Mioduszewska, 2005). This type of predation occurs mostly during the day (Terorde, 2008) and its range is small within turbid macrotidal estuaries (Baker & Sheaves, 2007; Tweedley *et al.*, 2016).

There are various methods used to monitor cyanobacterial blooms. Most frequently, the biomass of potentially toxic species has been determined by spectrophotometric measurements of Chl-a concentration and by microscopic counts and biomass calculated of phytoplankton. There is lack of literature describing fish abundance and phytoplankton abundance and biomass or chlorophyll a concentration in the same time in the shallow water areas. There are the first observation about fish reaction in coastal zone during cyanobacterial bloom in the Gulf of Gdańsk. Mastin *et al.* (2002) conducted laboratory and environmental study where influence of toxic algal blooms (with blue algae *Lyngbya* domination) on fish (*Pimephales promelas*) ontogenesis was examined. Fish survival rate was 76.7 % even if chlorophyll-a (Chl-a) concentration was over 2,600 $\mu\text{g dm}^{-3}$. Same investigations revealed that *P. promelas* significantly ($p < 0.05$) avoided *Lyngbya* - dominated cyanobacteria, collected in the spring (175 $\mu\text{g Chl-a dm}^{-3}$). In summer and winter time not detected statistical significant avoid-reaction when the maximum concentration of Chl-a were 200 and 140 $\mu\text{g dm}^{-3}$, respectively.

During our study the Chl-a concentration were no measured, because in the Baltic Sea the correlation between Chl-a and nodularin concentration may vary in wide range, from $r = 0.683$ (Henriksen 2005) to $r = 0.943$ (Schlüter *et al.* 2004). Mean correlation between Chl-a and nodularin concentration carried out in the coastal waters of the Gulf of Gdańsk in 2004 and 2005 was $r = 0.8$ (Mazur-Marzec *et al.*, 2006). Much better correlation is determined between

biomass of *N. spumigena* and NOD concentrations. Therefore the blooms were determined by microscopic methods. In surface water the abundance, biomass of *N. spumigena* and nodularin concentration were 6.27×10^7 100 μm filaments dm^{-3} , 1,531 mg dm^{-3} (Table 2) and 1,236 $\mu\text{g dm}^{-3}$ (Mazur-Marzec *et al.*, 2013), respectively. The concentration of NOD with the biomass of *N. spumigena* in seston measured in 2008-2011 by Mazur-Marzec *et al.* (2013) was well correlated ($P < 0.05$, $r = 0.946$). This results were in agreement with earlier studies by Henriksen (2005).

It is well known, that nodularin can accumulate in various organs of fish that are exposed to *Nodularia* blooms (Sipiä *et al.*, 2001a; Sipiä *et al.*, 2001b; Sipiä *et al.*, 2008; Karlsson *et al.*, 2003; Mazur-Marzec *et al.*, 2007; Sotton *et al.*, 2014). NOD produce some cellular effects, such as oxidative stress, leading to apoptotic processes and, at a larger scale, causing physiological disturbances. However, fish can detoxify NOD when exposed to the toxins and thus to counteract the negative effects of NOD (Sotton *et al.*, 2014). It should not be forgotten that *Nodularia* can also produce other metabolites that may adversely affect fish. To better understand the impact of toxic bloom of *Nodularia* on fish, the NOD concentration was investigated in fish by Mazur-Marzec *et al.* (2013). According to studies conducted in parallel with ours, NOD accumulation in round goby (*N. melanostomus*), caught during the most intensive *N. spumigena* bloom (30 June), reached 257.7 ± 151.2 ng NOD g^{-1} in muscles and 23.6 ± 7.0 ng NOD g^{-1} in liver. A month later, on 29 July, round goby muscles and liver contained 347.3 ± 564.2 ng NOD g^{-1} and $920.9 \pm 1,769.1$ ng NOD g^{-1} , respectively. Nodularin concentration in flounder (*P. flesus*) muscles and liver on 29 July was 13.6 ng g^{-1} and 191.8 ng g^{-1} , respectively. In that fish muscles, the toxin content exceeded the tolerable daily intake value (TDI) for humans (Mazur-Marzec *et al.*, 2013). This study shown, round goby accumulated higher amounts of the toxin than flounder. These differences may be due to the type of food being eaten and to the presence of these fish in different habitats. Round goby prefers shallow water and mainly feed on mussel (Sapota, 2004; Karlsson *et al.*, 2003), which is contaminated throughout the year by NOD (Sipiä *et al.*, 2001a; Sipiä *et al.*, 2001b; Sipiä *et al.*, 2008; Karlsson *et al.*, 2003; Mazur-Marzec *et al.*, 2007). On the other hand, the flounder migrates between the coastal zone and the open sea and feeds primarily on the *Macoma baltica* and, to a lesser degree, on the mussel. This composition of florist foods reduces the risk of NOD intake in comparison with round goby. Those investigations were conducted on fish from both samplings described in this publication. Our investigations shown that fish do not avoid *N. spumigena* blooms in initial phase. One of protection ways may be increased of biosynthesis of enzymes involved in biotransformation of xenobiotics as redox couple GSH/GSSG and related enzymes such as GS, GST and GSPXx. But the responses of these enzymes in different fish exposed to different toxins (also cyanotoxin) are inconsistent for the different type of fish species studied (Mazur-Marzec *et al.*, 2013; Srikanth *et al.*, 2014). Future studies are needed regarding the detoxification capacities and the effects of NOD on the reproduction processes of fish that are exposed to toxic *Nodularia* blooms both for short and long durations to better understand the potential risk of NOD contamination on fish populations.

CONCLUSION

Given the observed level of nodularin concentration, the fish tendency/behaviour to encroach on areas of *N. spumigena* blooms may increase accumulation of toxic substances in fish tissue and then the risk of contaminated fish consumption by humans. For a better assessment of the actual risk (not only for fish but also for humans), it is important to determine to what extent algal blooms tend to attract the fish and what is the residence time of nodularin in the fish body.

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