



No increase in marine microplastic concentration over the last three decades – A case study from the Baltic Sea

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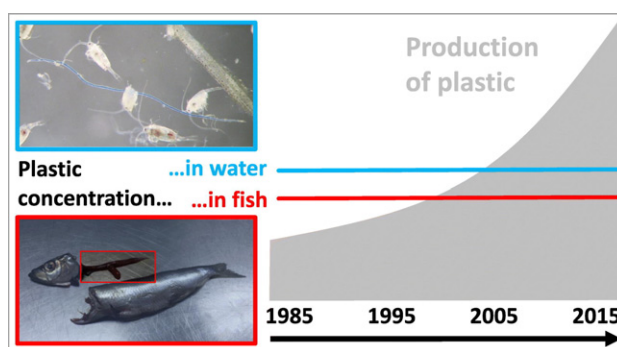
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HIGHLIGHTS

- First long-term study on microplastic in the marine environment
- Case study based on a unique sample set from the highly human impacted Baltic Sea
- Water column microplastic concentration constant over past three decades
- Microplastic concentration in forage fish constant over past three decades
- We hypothesise that household waste is the dominant source of Baltic marine plastics.

GRAPHICAL ABSTRACT



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ABSTRACT

Microplastic is considered a potential threat to marine life as it is ingested by a wide variety of species. Most studies on microplastic ingestion are short-term investigations and little is currently known about how this potential threat has developed over the last decades where global plastic production has increased exponentially. Here we present the first long-term study on microplastic in the marine environment, covering three decades from 1987 to 2015, based on a unique sample set originally collected and conserved for food web studies. We investigated the microplastic concentration in plankton samples and in digestive tracts of two economically and ecologically important planktivorous forage fish species, Atlantic herring (*Clupea harengus*) and European sprat (*Sprattus sprattus*), in the Baltic Sea, an ecosystem which is under high anthropogenic pressure and has undergone considerable changes over the past decades. Surprisingly, neither the concentration of microplastic in the plankton samples nor in the digestive tracts changed significantly over the investigated time period. Average microplastic concentration in the plankton samples was 0.21 ± 0.15 particles m^{-3} . Of 814 fish examined, 20% contained plastic particles, of which 95% were characterized as microplastic (<5 mm) and of these 93% were fibres. There were no significant differences in the plastic content between species, locations, or time of day the fish were caught. However, fish size and microplastic in the digestive tracts were positively correlated, and the fish contained more plastic during summer than during spring, which may be explained by increased food uptake with size and seasonal differences in feeding activity. This study highlights that even though microplastic has been present in the Baltic environment and the digestive tracts of fishes for decades, the levels have not changed in this period. This underscores the need for greater understanding of how plastic is cycled through marine ecosystems. The stability of plastic concentration and contamination over time observed here indicates that the type and level of

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microplastic pollution may be more closely correlated to specific human activities in a region than to global plastic production and utilization as such.

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1. Introduction

The United Nations, as part of sustainable development goal 14 has called for the prevention and significant reduction of marine pollution of all kinds, in particular from land-based activities (United Nations, 2017). One of the main indexes measuring progress toward this goal is the amount of floating plastic debris. There is a rapidly growing awareness of marine litter in general and plastics in particular. The global use and production of plastic has steadily increased since mass production started in the 1940s, with annual global production now exceeding 300 million tons (Suaria et al., 2016). Plastics in the form of small particles, so called 'microplastics' (i.e. <5 mm) have been observed in the environment worldwide (Auta et al., 2017), and are now considered a major component of plastic pollution in the marine environment. These microplastics mostly originate from the breakdown of larger plastic litter, but also include micro-particles already manufactured in such small sizes, e.g. for utilization in cosmetic products (Andrady, 2011). UV-radiation, physical fragmentation and weathering gradually degrade plastics into smaller and smaller fragments which can persist for a long period of time in marine habitats (Andrady, 2011; Ivar do sul and Costa, 2014). Much of the concern with respect to plastic debris involves their introduction into the marine food web, because microplastic particles may harm biota directly or indirectly by blocking the digestive tract (Derraik, 2002; Foekema et al., 2013; Lusher et al., 2013), by transporting persistent, bioaccumulated and toxic substances (Rochman et al., 2013; Teuten et al., 2009), and by leaking toxic plastic additives (Browne et al., 2013; Nobre et al., 2015).

The uptake of microplastics in the marine food web depends on the size, shape and density of the particles, as these parameters determine their position in the water column and thus their availability to potential consumers (Browne et al., 2007). Additionally, wind-driven mixing and currents play a major role for the distribution and fate of plastic particles (Lattin et al., 2004; Yamashita and Tanimura, 2007; Kukulka et al., 2012), hence the highest concentrations are observed in coastal waters, enclosed seas, and oceanic gyres (Eriksen et al., 2013; Eriksen et al., 2014; Goldstein et al., 2012; Ryan et al., 2009; Zarfl and Matthies, 2010).

Microplastics are taken up by marine organisms through ingestion and in some cases microplastic particles may cross the gills or the intestine walls and enter the tissue (Sussarellu et al., 2016). The kinetics of uptake of plastic particles by organisms in the marine food web is governed by a combination of their feeding biology and the concentration and size of the particles. As microplastics are suspected to transfer harmful substances to body tissues, particular concern has been allocated to microplastic ingestion by commercially important marine fish species intended for human consumption (Rummel et al., 2016).

Documentation of plastic in the digestive system of fish is indeed common (Lusher, 2015; Rochman et al., 2015; Carpenter et al., 1972), and plastic has been found in fish species from coastal waters and open oceans down to depths of 850 m (Anastasopoulou et al., 2013; Rochman et al., 2015). Small pelagic forage fish that prey mainly on zooplankton (Bernreuther et al., 2013; Casini et al., 2004) can mistake plastic for prey (Schuyler et al., 2014), ingest particles accidentally while feeding on zooplankton (Rummel et al., 2016), or via prey containing microplastics (Cole et al., 2013; Lusher et al., 2016). In marine ecosystems, small pelagic forage fish are key species, both ecologically and economically, as they are a major food resource for a variety of predators, channeling energy from their plankton prey to higher trophic levels (Smith et al., 2011), and contribute substantially to global food security (Alder et al., 2008). Thus, these forage fish also act as potential

vectors of microplastics from the planktonic environment to top predators and may in fact potentially even transfer microplastics to other livestock bred for human consumption, as the majority of forage fish catches are nowadays used for the production of fishmeal which is then used as fodder in aquaculture and terrestrial livestock industries (Alder et al., 2008), e.g. as chicken feed. In the generally species poor Baltic Sea, the two clupeid species herring (*Clupea harengus*) and sprat (*Sprattus sprattus*) are by far the two dominating pelagic fish species in terms of their abundance, biomass, their ecological relevance as consumers and as key prey for top predators (Ojaveer et al., 2010; Eero et al., 2012), including e.g. cod, salmon, sea birds, marine mammals and humans. This importance is also reflected in their economic value for the local fisheries (Ojaveer et al., 2010; Eero et al., 2012).

However, despite its importance for both commercial and recreational fisheries (Sparrevoorn and Storr-Paulsen, 2012), there are few studies that investigate the long-term fluctuation of microplastics in the Baltic Sea, an ecosystem already under heavy anthropogenic pressure which has resulted in regime shifts and changes in ecosystem health and functioning over the past decades (Andersen et al., 2015; BACC II Author Team, 2015). What has been shown recently, is that 5–16% of Baltic Sea fish do contain plastics (Rummel et al., 2016; Lenz et al., 2016a, 2016b). There has generally been a rapid, world-wide increase in the number of investigations on microplastics in marine biota during recent years, but these studies are temporally restricted snap-shots only. Thus, while there is increasing awareness about the global extent of microplastic contamination and its potentially detrimental effects, data on long-term changes in microplastic concentrations, which are urgently needed to assess and forecast potential impacts, are presently lacking. In the present study these challenges are addressed utilizing a unique and extensive sample collection of Baltic plankton samples as well as sprat and herring samples covering a period of approximately three decades that was originally collected and conserved for food web studies. To our knowledge, this is the first study on microplastics in marine organisms and their ambient environment covering such a long period, and we aim at providing strongly needed information on baseline levels and long-term trends of marine microplastic concentrations. Our objectives were to investigate if the increasing global plastic production over the last three decades is reflected in an increasing concentration of microplastics in (1) plankton samples and (2) the digestive tracts of the dominating planktivorous forage fish herring and sprat.

2. Methods

2.1. Sample collection

The study area is located in the Bornholm Basin, one of several deep basins in the Baltic Sea with a maximum depth of 95 m, which is located in the south-central Baltic between Sweden in the north, Poland in the south and the Danish island Bornholm in the west (Fig. 1). Samples of plankton and the fish species Atlantic herring (*Clupea harengus*) and European sprat (*Sprattus sprattus*) were collected between 1987 and 2015, covering 245 stations, of which 98 were plankton stations and 147 were trawling stations (Fig. 1). This long-term sample series was originally initiated for the purpose of food web studies, but provided a unique opportunity to address long-term microplastic trends in the context of the present study. Plankton samples were collected on a 24 h basis on a regularly spaced station grid using a Baby-Bongo net (Ø 20 cm, mesh size 150 µm) equipped with a flowmeter (General Oceanics). The net was towed in a double oblique haul integrating the entire water column,

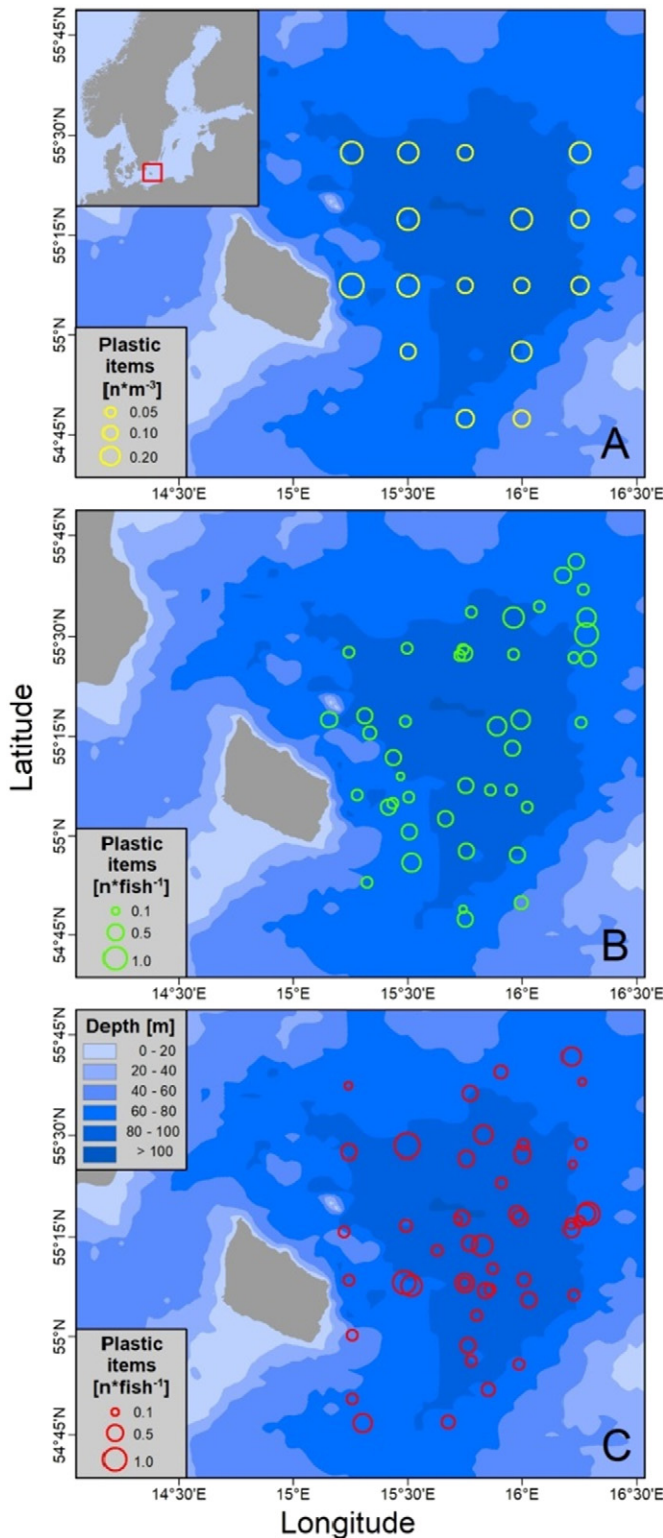


Fig. 1. Distribution of sampling stations and average microplastic concentration in A) plankton samples and digestive tracts of sprat and herring in B) spring (April–June) and C) summer (July–September). Samples covered the period 1987–2015. Size of circles is proportional to the concentration/amount of microplastics in the samples.

from 5 m above the bottom to the surface, at a towing speed of approximately 3 knots. The wire was paid out at 0.7 m s^{-1} and retrieved at 0.5 m s^{-1} . Samples were stored in 5% formalin. A sub-set of these samples was selected for the microplastic analyses in the present study covering the area with a temporal resolution of 3–5 years and a spatial

resolution of approx. 15 to 20 km (Fig. 1). Most samples were taken in the spring (April to early June) but for three years (1999, 2011, 2015) additional summer samples (July to September) were included.

Herring and sprat were sampled by pelagic trawling and stored at -20°C . Sub-samples were selected for the analysis of microplastic in the fish digestive tracts, again with a temporal resolution of 3 to 5 years as for the plankton samples but with a somewhat higher spatial resolution (Fig. 1). Samples from both spring and summer were selected, except for 1987 and 2006 when only spring samples were available. A total of 299 herring and 515 sprat were selected (Table 1).

2.2. Sample preparation

Plankton samples were filtered onto a $100 \mu\text{m}$ sediment sieve and after being rinsed with 25 mL filtered ($20 \mu\text{m}$) demineralized water to remove formalin, the sample was transferred to a glass beaker and immediately covered with a glass lid. The samples were dissolved in 30% solution of potassium hydroxide (KOH) and sodium hypochlorite (NaClO) adapted by Strand and Tairova (2016), i.e. 150 mL saturated KOH solution (1120 g L^{-1}) and 150 mL NaClO solution (14% active chlorine) to 700 mL MilliQ water. Previous tests confirmed digestion of organic tissue without causing extensive damage to the plastics (Enders et al., 2017). Two milliliter digestion solution was added per mL of plankton sample. First samples were subjected to a 10-minute ultrasonic treatment followed by 1 h of thorough shaking, on a standard shaking board, which decomposed the dominant fractions of natural organic matter. The digest was filtered through metal sediment sieves with mesh sizes of first $300 \mu\text{m}$ and then $100 \mu\text{m}$, rinsed into a petri dish and analysed under an Olympus dissection microscope at $\times 50$ magnification.

Fish samples were thawed at room temperature before examination in the laboratory. Total length (mm) and body weight (g) were measured, after which individuals were dissected and digestive tracts weighed separately. Digestive tracts were rinsed with 25 mL filtered ($20 \mu\text{m}$) demineralized water and dissolved in the same digestion solution as plankton samples. For optimal digestion 5 mL solution was used per gram of tissue. After ultrasonic treatment and thorough shaking, the digest was filtered through metal sediment sieves (1 mm and $300 \mu\text{m}$) stacked on each other. The remaining filtrate was filtered on to a $100 \mu\text{m}$ plankton net and rinsed with filtered ($20 \mu\text{m}$) demineralized water. The net was transferred to a closed glass petri dish for transport and subsequent analysis under an Olympus dissection microscope at $\times 50$ magnification.

2.3. Microplastic identification

Particles retained on the sieve mesh and the plankton net were visually inspected under a light microscope and photographs of all potential microplastics were taken. Potential microplastics were verified using established criteria for visual characterization (Enders et al., 2015) and in part confirmed with the hot needle test, which involved the application of a heated needle tip to each plastic to confirm that it would melt (Karlsson et al., 2017). All observed microplastic particles were size measured and classified by color and type (fibres or fragments).

2.4. Contamination avoidance

All laboratory equipment was rinsed with acetone before use, and rigorous precautions were taken throughout the entire procedure to avoid contamination. Direct contact with samples and filters was avoided, as was the use of plastic wash bottles. All actions, prior to microscopic observations, took place in a fume hood which was kept closed as much as possible. Controls were conducted for every five samples analysed; blank samples were processed as above by using pre-filtered ($20 \mu\text{m}$) water. Only 3 cellulosic and/or semi-synthetic particles were found in 162 control samples, and contamination was considered

Table 1
Fish collected during cruises in the Baltic Sea between 1987 and 2015.

Year	Season	Species	N	Average fish length (mm) (±SD)	Average fish weight (g) (±SD)	Percentage ingestion (%)	Average plastic per fish	Average plastic per fish with plastic
1987	Spring	Herring	35	202 ± 32	59 ± 28	20	0.26	1.3
	Spring	Sprat	25	131 ± 13	14 ± 3	20	0.20	1.0
1991	Spring	Herring	15	218 ± 18	62 ± 16	27	0.27	1.0
	Autumn	Herring	30	215 ± 29	73 ± 18	27	0.27	1.3
	Spring	Sprat	25	130 ± 9	14 ± 3	16	0.16	1.3
	Autumn	Sprat	35	131 ± 9	17 ± 4	40	0.40	1.3
1996	Spring	Herring	60	194 ± 23	45 ± 15	22	0.22	1.2
	Autumn	Herring	59	170 ± 29	31 ± 16	20	0.2	1.0
	Autumn	Sprat	60	110 ± 9	8 ± 1	23	0.23	1.0
1999	Spring	Herring	15	196 ± 24	55 ± 20	20	0.27	1.3
	Spring	Sprat	60	107 ± 16	8 ± 4	17	0.17	1.0
	Autumn	Sprat	60	121 ± 32	14 ± 16	52	0.23	1.2
2002	Spring	Sprat	50	119 ± 9	11 ± 3	18	0.18	1.1
	Autumn	Sprat	50	127 ± 8	13 ± 2	20	0.2	1.1
2006	Spring	Sprat	65	117 ± 17	12 ± 5	6	0.06	1.0
2011	Autumn	Herring	10	142 ± 40	23 ± 14	20	0.20	1.0
	Spring	Sprat	25	114 ± 16	10 ± 4	8	0.08	1.0
	Autumn	Sprat	25	119 ± 15	12 ± 4	12	0.12	1.0
2015	Spring	Herring	35	220 ± 13	64 ± 11	20	0.20	1.3
	Autumn	Herring	40	210 ± 19	59 ± 15	18	0.18	1.3
	Spring	Sprat	35	112 ± 16	10 ± 4	11	0.11	1.0

to be negligible. To investigate potential loss of plastics during the filtration of the plankton and stomach samples, 50 standard samples were filtered (100 µm) and the water that passed the filter was visually inspected under a light microscope (×50 magnification). No plastic particles were found in this control.

2.5. Statistical analysis

To assess differences in the concentration of microplastic particles (1) in plankton samples (number of particles m³ of filtered sea water) and (2) in herring and sprat individuals (number of particles/digestive tract) between sampling years and sampling seasons within the years, we applied two-way (nested) ANOVAs. Since data for both seasons were not available for all years, we conducted additional linear regression analysis to test for temporal changes in microplastic concentrations in plankton and fish over the entire time span, and two-sided unpaired *t*-tests to test for differences between seasons. Results from both approaches were consistent. To test for interspecies differences in the amount and size of ingested plastic particles, two-sided unpaired *t*-tests were applied. Since there were no significant differences, the species were pooled for subsequent analysis. To test for differences between locations within the Bornholm Basin, one-way ANOVAs followed by Tukey-Kramer post hoc tests were used. This analysis was not incorporated in the nested ANOVAs, as sample sites differed between years. Triangular distance to nearest shore (Bornholm, Sweden or Polen) was calculated for all sampling stations for both plankton and fish using the program ArcGIS (Version 10.4) and the correlation between microplastic concentration in the plankton samples and distance from nearest shore was tested by calculating Pearson's correlation coefficient *r*.

The association of the concentration in plankton and in fish samples from the same locations was assessed with linear regression analysis. Differences in concentrations between fish caught during different times of the day (daytime versus nighttime) were tested with an unpaired two-sided *t*-test. For each species the association of the concentration with fish size was tested by Pearson's correlation coefficient *r*.

Data were normally distributed and were thus not transformed. All statistical tests were considered significant at a critical *p* value of 0.05. BioStat Pro 6 was used for the ANOVAs and *t*-tests and GraphPad PRISM V7 was used for the linear regressions and Pearson's correlation tests.

3. Results

3.1. Microplastic concentration in the environment

The average microplastic concentration in the plankton samples was 0.21 ± 0.15 particles m⁻³, *n* = 97 (mean ± SD) between 1991 and 2015 (Fig. 1A). No significant change was found over time (one way ANOVA, $F_{8,88} = 1.49$, *p* = 0.17, Fig. 2A). The highest concentration was found in summer 2011 (0.28 ± 0.23 m⁻³, *n* = 12) and the lowest in spring 2006 (0.11 ± 0.07 m⁻³, *n* = 11). Also, no significant difference was found when comparing the different seasons (unpaired two-sided *t*-test with equal variance, *p* = 0.63), with a microplastic concentration of 0.22 ± 0.14 particles m⁻³, *n* = 32 in the spring samples and 0.24 ± 0.17 m⁻³, *n* = 35 in the summer samples (Fig. 2A). For the three years where both spring and summer samples were available, a two-way ANOVA was used to test the combined effect of year and season and this supported that there are no significant differences ($F_{\text{season}} = 0.23$, *p* = 0.63; $F_{\text{year}} = 0.87$, *p* = 0.43; $F_{\text{combined}} = 1.3$, *p* = 0.27). Finally, the microplastic concentration did not differ throughout the Bornholm Basin (one-way ANOVA, $F_{12,84} = 1.35$, *p* = 0.21) and no correlation was found between the microplastic concentration and the distance to the coast (Pearson's correlation coefficient *r*, *r* = -0.45, *p* = 0.09, Fig. 3).

3.2. Plastic content in the fish samples

Overall, microplastic particles were found in 160 (63 herring and 97 sprat) of the 814 examined fish (20%). Sprat contained 0.21 ± 0.47 (mean ± SD) plastic particles fish⁻¹ (total number of examined fish = 515) and the herrings contained 0.25 ± 0.52 (mean ± SD) plastic particles fish⁻¹ (total number of examined fish = 299) (Table 1). The 160 fish that contained plastic had between one to three pieces of plastic in their digestive tract, with a mean of 1.15 ± 0.13 particles fish⁻¹, *n* = 160. There was no significant difference between the two species in the amount of plastic in the digestive tracts (unpaired two-sided *t*-test with equal variance, *p* = 0.27) or in the particle size ingested (unpaired two-sided *t*-test with unequal variance, *p* = 0.06), and thus in all subsequent analyses except the test for correlation with fish size we pooled the data from the two species. There was no significant difference in the plastic content of the fish over the period of 28 years (one-way ANOVA, $F_{5,683} = 2.13$, *p* = 0.06, Fig 2B), but they contained a significantly higher amount of plastic particles during the summer months (0.28 ± 0.54 pieces fish⁻¹, *n* = 369) than during spring ($0.20 \pm$

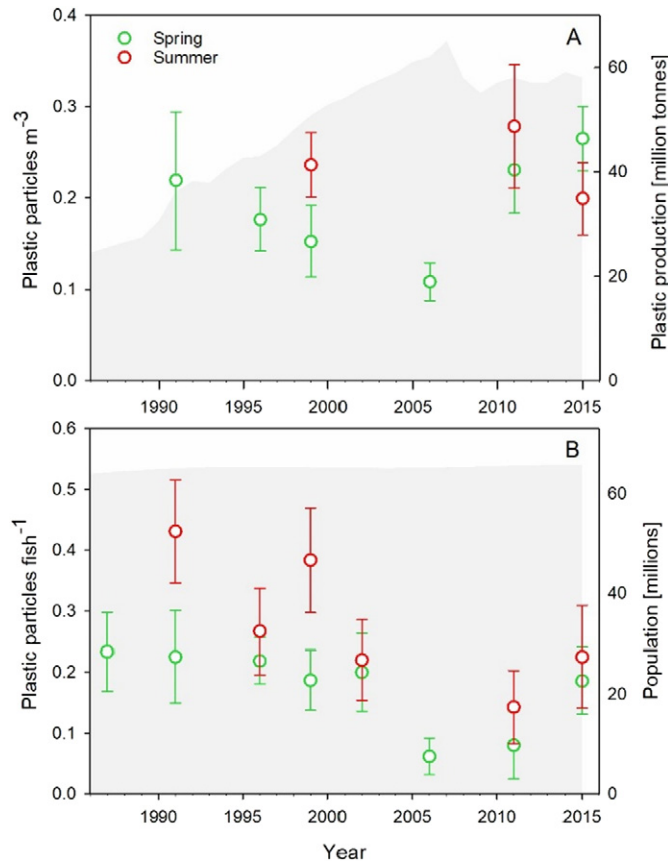


Fig. 2. Microplastic concentration from 1987 to 2015 in (A) plankton samples and (B) fish digestive tracts. Each point represents the average of all samples taken a given year and season (spring = green circles, summer = red circles) and error bars indicate SD. Grey area in (A) refers to the development of European plastic production (Plastics Europe, 2015), in (B) to the development of total population in the countries with the majority of their land area located within the Baltic Sea catchment area, i.e. Denmark, Sweden, Finland, Estonia, Lithuania, Latvia, Poland (United Nations, 2015).

0.46 pieces fish⁻¹, $n = 320$) (unpaired two-sided *t*-test with equal variance, $p = 0.04$, Fig. 2B). Again, a two-way ANOVA was used to test the combined effect of year and season. It confirmed that the content did

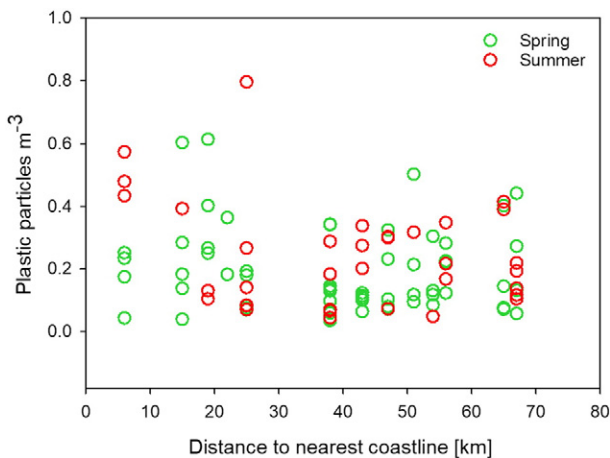


Fig. 3. Distribution of microplastic in the sampling area. The microplastic concentration in the water column did not correlate with the distance to the nearest coast line and the microplastic was thus homogeneously distributed in the Bornholm Basin. Each circle represents one individual plankton sample.

not change significantly over the 28 years and that the content was significantly higher during summer than during spring ($F_{\text{year}} = 2.1$, $p = 0.06$; $F_{\text{season}} = 4.0$, $p = 0.045$; $F_{\text{combined}} = 1.2$, $p = 0.29$). For both herring and sprat the size of the fish was positively correlated with the number of plastic particles in the digestive tract (Pearson's correlation coefficient r , sprat = 0.80, $p = 0.01$; herring = 0.64, $p = 0.005$).

During most of the cruises trawling was only conducted during daytime but in summer 1991, 7 hauls were conducted at daytime (between 03:00 and 19:00 UTC) and 6 during nighttime (between 19:00 and 03:00 UTC), defined as the period from 30 min before sunset to 30 min after sunrise. No significant difference was observed between daytime (0.38 ± 0.65 particles fish⁻¹, $n = 34$) and nighttime (0.48 ± 0.72 particles fish⁻¹, $n = 31$) (unpaired two-sided *t*-test with equal variance, $p = 0.50$).

3.3. Microplastic characterization

The size of plastic particles found in the plankton samples ranged from 0.1 to 11.5 mm (mean: 1.6 ± 1.7 mm, $n = 356$) and microplastics (i.e. <5 mm) constituted the majority (94%) (Fig. 4). The plastic was dominated by fibres (93%, $n = 330$) compared to fragments (7%). A total of 184 plastic particles ranging in size from 0.12 to 27.5 mm (mean: 1.2 ± 2.4 mm) were identified in the digestive tracts of the fish (Fig. 4). Of these, 175 particles were <5 mm in length, and thus nearly 95% of the detected particles were microplastics. Just as for the plankton samples, the digestive tracts contained far more fibres than fragments (93% and 7% respectively). Also, a similar size frequency distribution was found in fish and in the plankton samples (Fig. 4), despite the slight difference in mesh size between the plankton net (150 μm) and the sieves (100 μm) used to collect the plastic particles from the dissolved digestive tracts. Fibres from both the plankton samples and from the digestive tracts were between 10 and 40 μm in diameter. The size frequency also shows that the smaller particles are the most abundant (Fig. 4); 76% were smaller than 2 mm. The plastics represented a wide variety of colors with black being the most prevalent in both plankton samples (70%) and fish (79%) (Supplementary Table 1).

4. Discussion

Here we present the first long-term study of microplastics in the marine environment as well as in the digestive tracts of pelagic planktivorous forage fish. In contrast, previous studies on marine

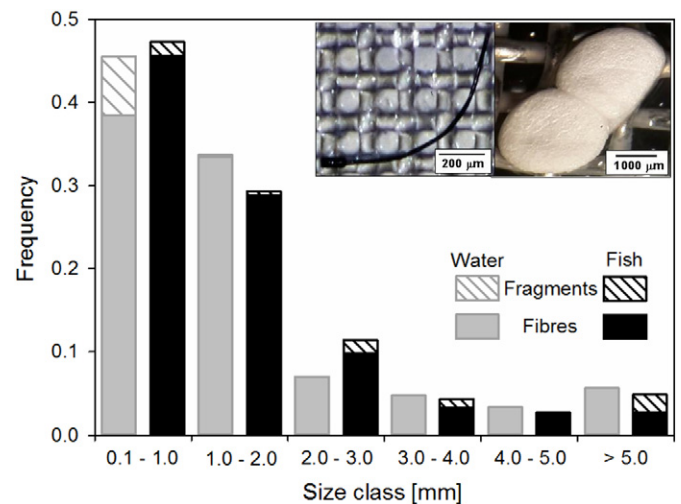


Fig. 4. Size and type of microplastic particles in the plankton (grey bars) and in the fish digestive tracts (black bars). Pictures show a piece of a plastic fiber and fragment found in samples.

microplastics are only short-term presenting a snapshot of the contamination levels at a given point in time. Surprisingly our extensive dataset shows that despite the gradually increasing global production of plastics, the microplastic concentrations in both the plankton and the digestive tracts of herring and sprat have been constant in the Baltic Sea over the last three decades.

4.1. Unchanged microplastic concentration during the last 25 years

Prior to our study, the only long-term studies of marine plastic were showing an increase from the 1960s and 1970s to the 1980s and 1990s (Thompson et al., 2004), levelling off over the last three decades (Law et al., 2010; Law et al., 2014). However, these studies were focusing on surface macroplastics and microplastics in sediments, and thus nothing was known about long-term changes in microplastic concentration in the water column and in marine organisms. Our results show that microplastics in the Baltic Sea are ubiquitous and homogeneously distributed in space, but also that concentrations have remained unchanged through time. The concentrations in the plankton samples remained unchanged from 1991 to 2015 despite a steadily rising global plastic production during the last 50 years. Likewise, European annual plastic production has almost tripled from approximately 22 to about 60 million tons during the period of our investigation, i.e. from the mid 1980s until the 2000s (see Fig. 2A, Plastics Europe, 2015).

Considering the increase in plastic production, we expected also an increase in plastic concentration in the plankton samples and in fish over time. A possible explanation for our finding of unchanged microplastic concentration is that different types of plastics have different probabilities of ending up as marine microplastics. The vast majority of plastics we recovered from the plankton samples were fibres (93%). It is known that the most likely source of such micro fibres is waste water from washing clothes and other synthetic textiles (Browne et al., 2011; Murray and Cowie, 2011). The monofilaments from textile fibres are typically between 10 and 50 μm in diameter (Chattopadhyay, 2010; Sinclair, 2014; Tanaka and Takada, 2016), which matched the fibres from the present study, ranging between 10 and 40 μm . Following this, the abundance of the plastic fibres should be closer related to the textile production and especially the amount of clothes washed in the countries around the Baltic Sea than to the total European or global production of plastic (DHI, 2015; C  zar et al., 2017). This again should at least to some degree correlate with the population size around our study site, and interestingly, the total population size of the main countries of the Baltic Sea catchment area has also been constant over the past 30 years (see Fig. 2B). In more remote locations, like Antarctica, not only much lower concentrations but also different types of microplastics are found. Here fragments are the predominant microplastics while only very few fibres are found (Cincinelli et al., 2017). Taken together these results suggest that the type and degree of microplastic pollution in a given marine ecosystem is more likely to be correlated to the level of specific human activities, like washing of

clothes, in the region than to total plastic production and utilization as such (Jambeck et al., 2015).

The observed microplastic concentration of 0.21 particles m^{-3} in the Baltic Sea was similar to concentrations reported from the English Channel (0.27 m^{-3} , Cole et al., 2014) and in Mediterranean waters (0.15 m^{-3} , de Lucia et al., 2014). Other studies have found up to 10 times higher concentrations (e.g. Lusher et al., 2014, see Table 2 for list of worldwide concentrations). What should be kept in mind is that these absolute concentrations are highly influenced by the mesh size and type of the sampling gear. Most studies have used a mesh size of $\sim 300 \mu\text{m}$ (Hidalgo-Ruz et al., 2012), but in order to get a larger fraction of the total microplastic we applied a 150 μm mesh size. Still, when looking at the size distributions of the particles (Fig. 4), it is clear that abundance increases with a decrease in size probably due to fragmentation processes (Eriksen et al., 2014). Thus, the real concentrations are likely somewhat higher than reported here and elsewhere in the literature.

4.2. The microplastics in fish reflect the concentration in plankton samples

Our results also demonstrate that microplastics were present in the digestive tracts of two key fish species in the Baltic Sea, herring (*Clupea harengus*) and sprat (*Sprattus sprattus*) and have been so during the last three decades. Yet again, we found no increase in the microplastic content over the last three decades. Both microplastic content and composition in the digestive tracts directly mirrored what we found in the water column. The similar ratio between fibres and fragments as well as the similar size distributions between the plankton samples and the fish digestive tracts indicated that at least these two fish species were non-selective in their plastic ingestion.

We found that 20% of the examined fish had ingested microplastic, which is remarkably similar to what has recently been reported for cod and herring from the North and Baltic Sea, where 23% of the fish had ingested plastic (Lenz et al., 2016a, 2016b). Both lower and higher values have been reported for other fish in the same region (Foekema et al., 2013; Rummel et al., 2016; Lusher et al., 2016; Grellier and Hammond, 2006, and Table 2), and even though these different results may reflect differences in the sampling methods and processing procedures, it may also be related to seasonal and/or species-specific differences in feeding biology. As an example of this, the present study is to our knowledge the first to document a seasonal influence on microplastic ingestion, with more particles present in the digestive tracts during summer than during spring. This corresponds well with the feeding ecology of the two species, which both show increasing feeding rates from spring to summer (Bernreuther, 2007).

Comparing six fish species of different size ranges, Boerger et al. (2010) documented higher microplastic abundance in the digestive tracts of larger fish, implying that by increasing food uptake, larger fish may encounter more plastic particles. This pattern was confirmed here by the significant correlation of fish size and the number of

Table 2
Mean plastic abundance in surface waters (plastic m^{-3}) and in fish (items fish $^{-1}$) around the world.

	North Pacific Ocean	North Atlantic Ocean	Indian Ocean	South Atlantic Ocean	South Pacific Ocean
Water column	0.12 (Goldstein et al., 2012) 0.17 (Zhao et al., 2014) 7.25 (Moore et al., 2001) 2.23 (Moore et al., 2002) 3.92 (Lattin et al., 2004) 0.004–0.19 (Doyle et al., 2011)	0.27 (Cole et al., 2014) 0.15 (de Lucia et al., 2014) 2.46 (Lusher et al., 2014) 15–501 (Enders et al., 2015)	0.0008 (Reisser et al., 2013)	1.15 (La Daana et al., 2017)	0.17 (Jensen et al., 2017)
Fish	2.1 (Boerger et al., 2010)	0.03 (Foekema et al., 2013) 0.70 (Lusher et al., 2013) 0.13 (Lusher et al., 2016) 0.27 (Neves et al., 2015) 1.56 (Bellas et al., 2016)	2.1 (Naidoo et al., 2016)		4.1 (Jensen et al., 2017)

ingested microplastic particles in both herring and sprat. Furthermore, the lack of correlation between fish size and the size of ingested microplastics in our study matched an earlier study comparing fish ranging from 10 cm (herring) to almost 1 m (cod) (Foeckema et al., 2013).

4.3. Potential implications of ingested microplastics

In those fish which contained plastic only few pieces were found, which strongly suggests that microplastics do not accumulate in the digestive tract. Gut evacuation times of herring and sprat vary according to temperature and feeding intensity (Bernreuther et al., 2008; Bernreuther, 2007). Depending on ambient temperatures the gut can be considered emptied after 12 to 24 h, after which plastics are likely to be evacuated along with the faeces (Lenz et al., 2016a, 2016b). Constipation is therefore not likely to be a problem for the fish eating microplastics. The main concern of ingesting microplastics seems to be the potentially detrimental effects of hazardous chemicals present as additives in the polymers or compounds adhering to the surface (Teuten et al., 2009; Mato et al., 2001). Unfortunately, no solid experiments have been published which test such effects under natural conditions and concentrations.

5. Conclusion

The mounting body of literature documenting microplastic occurrence across our planet reflects an increasing awareness and concern regarding microplastics in our ecosystems and the possible implications of this pollution. Previous studies from marine ecosystems have been snapshots in time, and there has been a lack of quantitative long-term data essential to define baseline levels and evaluate the development of microplastic contamination over time. In the present study we provide such a baseline and document the long-term development in a marine environment with a high anthropogenic impact. Over a period of three decades microplastic was present in both the water and in two key forage fish species consistently, but concentrations did not increase over time. While the stable situation may to some extent be encouraging, as increasing trends in plastic production are not reflected in the Baltic environment, it is of vital importance to obtain more data on the plastic retention times and potential releases of chemicals from the plastic particles in the gut to better understand the impact of the observed microplastic levels. We also need to learn how plastic gets circulated and breaks down in the marine environment, and its role in the ecosystem. Such studies have to be conducted with particles containing environmentally relevant compounds and in naturally occurring concentrations (Lenz et al., 2016a, 2016b). Not until such data are available will it be possible to quantify the role and impact of plastic in the food web.

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.scitotenv.2017.10.101>.

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