Final report on field studies on marine and beach litter

Prepared by Estonian Marine Institute, University of Tartu

Project	GES-REG
Work package	Work Package 3 – Advance knowledge base to support assessment of GES
Preparation date	22 July 2013
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Marine and coastal litter case studies

Report

Henn Ojaveer, Estonian Marine Institute, University of Tartu

The current report summarises results of the three case studies performed by the GES-REG project partner institutes. These were carried out in different parts of the NE Baltic Sea and on different topics related to the marine and coastal litter.

Our results indicate that microplastic particles may be introduced to the Baltic Sea planktonic food web through different mesozooplankton taxa.

We can also conclude, based on experimental studies, that plastic products can cause a negative impact on planktonic crustaceans and influence their sustainable development, evidenced by the fact that 60 % of all investigated plastic products caused a negative impact on the test organisms *Daphnia magna* and *Artemia salina*.

Coastal macrolitter surveys performed in three sites in the Gulf of Riga and Gulf of Finland indicate that the strongly dominating type of litter across all three study sites was plastic/polystyrene making up to 86% of the total litter abundance.

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Finnish case study on microlitter

Outi Setälä, Finnish Environment Institute (SYKE)

Studies carried out in the last decade have pointed out the commonness of plastic microparticles in the marine environment. In the Baltic Sea, according to the study of Magnusson & Noren (2011) the concentration of microlitter in the size range of 10 - 220 µm in the coastal Baltic Sea was up to 4 fibres L^{-1} and 32 other anthropogenic litter particles L^{-1}. Microlitter, and microplastics in general are of concern especially because they can be ingested by a variety of marine organism, and possibly also transfer along the food web. In years 2012-2013 we carried out a case study both with micro-sized debris distribution (I) and the ability of zooplankton to ingest and transfer microplastics (II).

(I) Field studies

To estimate microlitter distribution, we carried out field studies in the open Gulf of Finland in 2012 and 2013 onboard R/V Aranda. In 2012 the material for the detection of plastics was collected by filtering 500 L surface water from approx. 4m depth, using a submerged pump. The samples were fractioned through 300 µm net (fraction >300 µm) and 1-2 L through 10 µm net (fraction 10 - 300 µm). Altogether 8 stations were sampled with a submerged pump (pumping speed approx. 15 L/min). Control samples were made by filtering tap water in controlled conditions with the same pumping equipment in the Kumpula facilities of SYKE Marine Research Centre. Prior to each filtration event the filters used were investigated under stereomicroscope to exclude the possibility that plastic particles or other litter were attached to them. In year 2013 the uses of a Manta trawl for the collection of >330 µm microplastic particles was tested. This method was compared with the use of a submerged pump, and the idea is to compare the results obtained from these two methods. The preliminary idea behind this study was to estimate the pros and flaws of using different monitoring methods for surface microplastics.

(II)Laboratory experiments

In this study we experimentally tested these hypotheses; the potential of different zooplankton taxa to ingest microplastics and food web transfer, in order to find information on possible of negative impacts in the food web. The study was carried out by using 10 µm fluorescent latex microspheres to trace the ingestion and transfer of microspheres in zooplankton.

Results

(I) Field studies onboard R/V *Aranda*

Microscopic analysis of particles from the 300 µm net filtrations was easy to perform and litter particles easily detected from organic material. In contrast to that, the 10µm net filtrations were much more difficult to study, and the results may be over- as well as underestimates of the amount of debris. Both fractions contained litter particles and were preliminary sorted as fibers or other foreign particles (including black anthropogenic particles with unknown status). The average amount of debris in the >300 µm fraction was 7.4 fibers m^{-3} and 2.7 other particles m^{-3}, whereas in the 10-300 mm fraction 1.8 fibers L^{-1} and 6.0 others particles L^{-1} were found. To be sure of contents of the smaller fraction, the samples should be re-analyzed with some other method designed for material analysis. From this dataset differences between stations were observed. The highest numbers of marine debris were found at the two easternmost stations (Figure 3.) at the XV3 and XV1 where altogether 27 and 28 particles m^{-3} were found. High numbers were also found at the station LL6 in the middle of the Gulf of Finland between Tallinn and Helsinki.

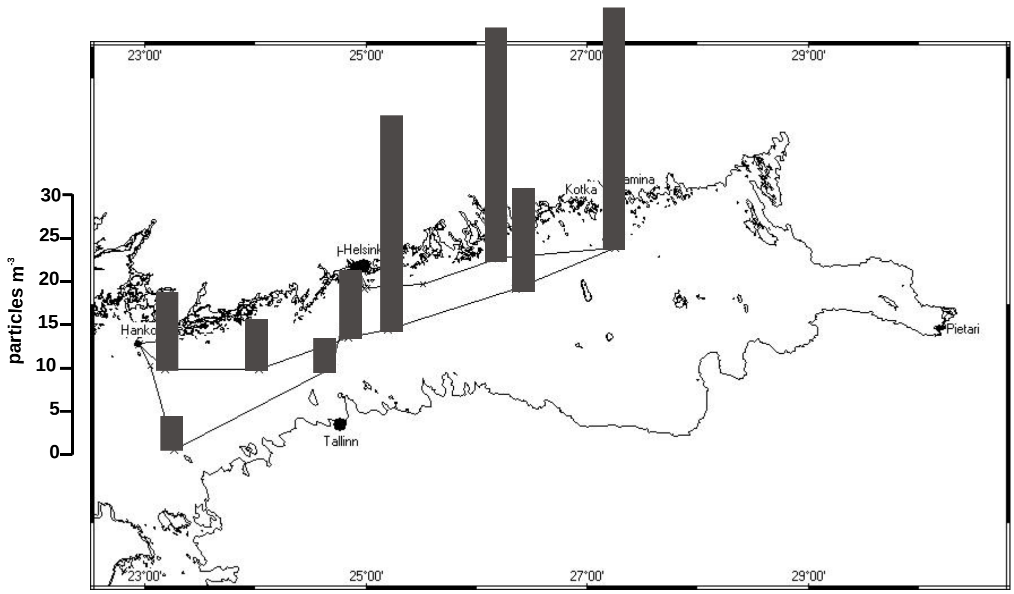

Figure 3. Total numbers of >300 µm litter particles (particles m^{-3}) at different stations in the Gulf of Finland.

(I) Laboratory studies with plastic microspheres and zooplankton

The study confirmed both of these hypotheses, how several zooplankton taxa were ingesting plastic microspheres (Figure 2.), and for the first time the potential of plastic microparticle transfer via planktonic organisms from one trophic level (mesozooplankton) to a higher level (macrozooplankton). The work is to be published in February 2014 in the journal *Environmental Pollution:* http://www.sciencedirect.com/science/journal/02697491

Abstract

Experiments were carried out with different Baltic Sea zooplankton taxa to scan their potential to ingest plastics. Mysid shrimps, copepods, cladocerans, rotifers, polychaete larvae and ciliates were exposed to 10 mm fluorescent polystyrene microspheres. These experiments showed ingestion of microspheres in all taxa studied. The highest percentage of individuals with ingested spheres was found in pelagic polychaete larvae, *Marenzelleria* spp. Experiments with the copepod *Eurytemora affinis* and the mysid shrimp *Neomysis* integer showed egestion of microspheres within 12 h. Food web transfer experiments were done by offering zooplankton labelled with ingested microspheres to mysid shrimps. Microscopy observations of mysid intestine showed the presence of zooplankton prey and microspheres after 3 h incubation. This study shows for the first time the potential of plastic microparticle transfer via planktonic organisms from one trophic level (mesozooplankton) to a higher level (macrozooplankton). The impacts of plastic transfer and possible accumulation in the food web need further investigations.

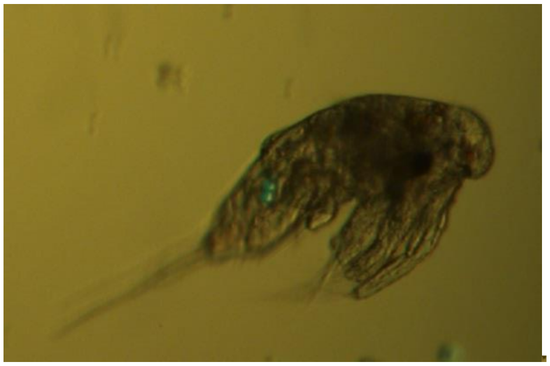

Figure 2. *Limnocalanus macrurus* nauplii with ingested microspheres.

References

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Latvian pilot study on potential toxicity of microlitter

Maija Balode, Vija Jurkovska, Liene Muzikante. Latvian Institute of Aquatic Ecology (LHEI)

Introduction

Characteristics of plastic litter in the marine and coastal environment. Source and amount trends, analysis of its composition and spatial distribution.

Global production of plastics has increased from 1,5 million metric tonnes in 1950 to 230 million metric tonnes by 2008 [12] and according to PlasticsEurope [15] 25% is produced in Europe. World's plastic production and plastic debris goes hand in hand. Due to the increasing plastic production, increases of plastic debris have been also observed.

Plastic litter (also known as plastic debris) is found in each part of the marine environment: 1.debris on shorelines; 2. floating debris on the sea and ocean surface; 3. debris throughout the water column; 4. debris on the seafloor [4;8]. Plastic debris of all sizes and shapes nowdays is a serious transboundary pollution problem.

According Derraik J. [4] plastic's constitute the major part of marine litter worldwide. Proportion of plastic among marine debris (persentage): beach - 32-90%; surface water -86%; sea floor - 47-85%. The methods used in the studies of amount trends, composition and spatial distribution of plastic litter are not standardized hence can not be compared. The most distributed plastic types and plastic products are shown in Table 1.

Table 1. Plastic types and plastic products

Plastic types	Products	Characteristic

Polyethylene terephthalate (PET or PETE)	Used in soft drink, juice, water, beer, mouthwash, peanut butter, salad dressing, detergent and cleaner containers	Leaches antimony trioxide and (2ethylhexyl) phthalate (DEHP) DEHP has been strongly linked to asthma and allergies in children. It may cause certain types of cancer and it has been linked to negative effects on the liver, kidney, spleen, bone formation, and body weight. In Europe, DEHP has been banned since 1999 from use in plastic toys for children under the age of three
High-density polyethylene (HDPE)	Used in opaque milk, water and juice containers, bleach, detergent and shampoo bottles, garbage bags, yogurt and margarine tubs and cereal box liners	Considered a **safer plastic**. Research on risks associated with this type of plastic is ongoing
Polyvinyl chloride (V or Vinyl or PVC)	Used in toys, clear food and non-food packaging (e.g., cling wrap), some squeeze bottles, shampoo bottles, cooking oil and peanut butter jars, detergent and window cleaner bottles, shower curtains, medical tubing, and numerous construction products (e.g., pipes, siding).	PVC has been described as **one of the most hazardous consumer** products ever created. Leaches di (2-ethylhexyl) phthalate (DEHP) or butyl benzyl phthalate (BBzP), depending on which is used as the plasticizer or softener (usually DEHP). In Europe, DEHP, BBzP, and other dangerous phthalates have been banned from use in plastic toys for children under three since 1999.

Low-density polyethylene (LDPE)	Used in grocery store, dry cleaning, bread and frozen food bags, most plastic wraps, and squeezable bottles (honey, mustard).	Considered a **safer plastic**. Research on risks associated with this type of plastic is ongoing.
Polypropylene (PP)	Used in ketchup bottles, yogurt and margarine tubs, medicine and syrup bottles, straws, and Rubbermaid and other opaque plastic containers, including baby bottles.	Considered a **safer plastic.** Research on risks associated with this type of plastic is ongoing.
Polystyrene (PS)	Used in Styrofoam containers, egg cartons, disposable cups and bowls, take-out food containers, plastic cutlery, and compact disc cases	Leaches styrene is an **endocrine disruptor** mimicking the female hormone estrogen, and thus has the potential to **cause reproductive and developmental problems**. Long-term exposure by workers has shown brain and nervous system effects and adverse effects on red blood cells, liver, kidneys, and stomach in animal studies. Styrene migrates significantly from polystyrene containers into the container's contents when oily foods are heated in such containers

Specific gravity of sea water is ~1,025. As seen from Table 2. only a few of the plastics used in marine environment have a specific gravity lower than that of seawater.

Table 2. Types of plastics commonly encountered in the marine environment [1]

Plastic class		Specific gravity	Percentage production*	Products and typical origin
Low-density polyethylene	LDPE LLDPE	0,91-0,93	21%	Plastic bags, six-pack rings, bootles netting, drinking straws
High- density polyethylene	HDPE	0,94	17%	Milk and juice jugs
Polypropylene	PP	0,85-0,83	24%	Rope, bottle caps, netting
Polystyrene	PS	1,05	6%	Plastic utensils, food containers
Foamed Polystyrene				Floats, bait boxes, foam cups
Nylon	PA		<3%	Netting and traps
Thermoplastic polyester	PET	1,37	7%	Plastic beverage bottles
Polyvinylchloride	PVC	1,38	19%	Plastic film, bottles, cups
Celuloze acetate	CA			Cigarette filters

*Fraction of the global plastics production in 2007 after (Brien 2007,[1])

<u>Hazardous impact of microplastics to human health and environment.</u>

The high molecular mass polymers are inert and not hazardous from a toxicity point of view [7]. Their large size limits transport across biological membranes. Instead, it is the presence of additives (e.g. antioksidants, stabilisers, plasticisers, flame retardants, catalysts), low molecular mass polymers and unpolymerised residual monomers, mainly determine the migration potency of chemical substances.

Several plastic additives are hazardous to human health and environment:

- toxic for reproduction - *di(2-ethylhexyl)phthalate (DEHP), bisphenol A;*
- carcinogenic - *vinyl chloride, acrylonitrile, benzene, 1,3-butadiene*;
- allergenic - *formaldehyde, acrylonitrile, toluene diisocyanate (TDI)*
- mutagenic - *benzene, phenol, 1,3-butadiene*;
- high chronic toxicity - *benzene;*

- very high acute toxicity - *phosgene, toluene diisocyanate (TDI);*
- environmentally hazardous with long term effects – *pentabromodiphenyl (PeBDE), acrylonitrile*, *toluene diisocyanate (TDI*) [7].

Direct environmental impacts of plastic litter:

Ingestion

Ingesting marine debris can seriously harm marine life. Seabirds, sea turtles, fish, and marine mammals often ingest marine debris that they mistake for food. The impact of the plastic debris (>5mm) are known at least 267 species worldwide, including 44% of all seabirds, 43% of all marine mammals , 86% of all turtles, as well as fish species (Laist 1997, after [6]).

In some instances the debris may pass through the gut without harming the animal. In other cases it can become lodged in their throats or digestive tracts. This can lead to starvation or malnutrition if the digestive tract is blocked (US EPA 1992a). Debris can accumulate in the gut and give a false sense of fulness.

Some plastics contain toxic substances that can cause death or reproductive failure in any marine life. Plastic particles have even been determined to contain certain chemicals up to one million times higher amounts then found in the water alone [10].

Entanglement

Today, many fishing gear items are made of plastic, including nets, pots, and traps. Because of this, they last a long time when lost or discarded in the marine environment. These derelict fishing gear items pose an entanglement risk to marine species of all types (Laist 1997) Entanglement can lead to suffocation, starvation, drowning, increased vulnerability to predators, or other injury [11].

Indirect environmental impacts of plastic litter:

Pollutants

Plastic debris accumulates persistent organic pollutants (POPs)such as PCBs (polychlorinated biphenyls), DDT, PAHs, and aliphatic hydrocarbons [13]. All chemicals have many non-point sources to the marine environment. This makes it difficult to determine the contributions of plastic debris pollutants to concentrations in marine species, the potential transfer of chemicals throughout the food chain and the implications for the bioaccumulation [14].

Invasive species

Marine debris can contribute to the transfer and movement of invasive species. Floating marine debris can carry invasive species from one location to another.

Microplastics. Amount trends, distribution and composition of micro-particles.

Only recently the International Research Workshop on the Occurence, Effects and Fate of Microplastic Marine Debris in September, 2008 defined term „microplastics" as „plastic particles smaller than 5 mm (recognising 333 µm as a practical lower limit when neuston nets are used for sampling) [9].

Sources of microplastic particles:

First source – microplastics originates from industrial and domestic products include:

- toilet, hand, body and facial cleaners
- cosmetics, tiny bead scrubers used in washing products
- powders and resin pellets used as the basic thermostatic industry feedstocks
- abrasive plastic beads used to clean ships (after [9]).

Those products are found to contain small (less than 1mm in diameter) polyethylene and polystyrene particles.

Plastic micrograins (>0,63 µm diameter) are found in hand cleaning and cosmetic products from 0,19-6,91 g in 100 g product (Gregory,1996, after [6]).

Secondary source – microplastics formed from the breakdown and degradation of larger plastic material. It is happened with or without assistance from UV radition and mechanical forces in the seas (e.g. wave, sand action, and oxidation) [12]. The photo-degradation process continues down to the molecular level, yet photo-degraded plastic remains a polymer. Estimates for plastic degradation at sea has been ranged from 450 to 1,000 years [8]

Impacts of litter on marine life

Several recent studies have identified potential effects of plastic particles, including:

- desorption of persistent, bioaccumulating and toxic (PBT) substances from plastics
- leaching of additive from the plastics
- physical harm [12]

Lithner D. et al. [7] has studied leachates from 32 different plastic products, with 15 different plastic types. Chemical substances leaching from plastic products to water caused acute toxic effects for *Daphnia magna* in 9 out of 32 products.

According Zitko (after Derraik [4]) low molecuar weight compounds from polystyrene particles are leached by seawater, and the fate and effects of such compounds on aquatic biota are not known.

The aim of the Pilot Project was to detect potential toxicity of microplastics on aquatic organisms of different trophic level.

Materials and Methods

Series of laboratory experiments were carried out to study the impact of plastic products on different test organisms. Screening for acute toxicity was made on 6 different newly bought plastic products, with 4 different plastic types and various applications represented (Table 1). New plastic products were cut with stainless steel scissors in 10 x 10 mm pieces and 10 g was placed in a 200 ml glass bottle (Figure 1). No pre-washing of the products was made, so the first leaching water was being tested. 100 ml of deionised water was added, giving a concentration of 100 g plastic material L^{-1}, equivalent to a liquid to solid ratio (L/S) of 10 L kg^{-1} (Lithner 2009). All bottles were places in 20 ± 2 ºC temperature for 7 days. After a week plastic product leachates were strained through a filter paper or GF/C filter to remove plastic pieces and the water phase was tested for acute toxicity.

Table 1. Plastic products and plastic types tested in this study

Nr.	Plastic product	Plastic type	Symbol	Plastic code
1.	Plastic dishes	polypropylene	PP	5
2.	Compact disc	polyvinyle chloride	PVC	3
3.	Garbage bag	low density polyethylene	LDPE	4
4.	Dish sponge	polyurithene	PU	7
5.	Phytoplankton bottle	polyvinyle chloride	PVC	3
6.	Binding covers	polyvinyle chloride	PVC	3
7.	Big water bottle	polycarbonate	PS	7
8.	Plastic egg container	polystyrene	Other	6

Figure 1. Plastic product leachates.

Figure 2. All tested plastic products.

Acute toxicity tests

One species of microalgae, 4 species of benthic amphipods and 2 zooplankton species were used as testobjects in eco-toxicological tests (Table 2). Acute toxicity tests were performed on green microalgae - *Desmodesmus communis* (Figure 6) according to the ISO standart test (ISO 8692:2005), 4 species of benthic amphipods - *Hyalella azteca* (Figure 2), *Gammarus pulex* (Figure 3), *Monoporeia affinis* (Figure 4) and *Corophium volutator* (Figure 5) according to the ISO standart test (ISO 16712:2005), 2 zooplankton species *Daphnia magna* (Figure 7) according to the ISO standard test (ISO 6341:1996) and *Artemia salina* (Figure 8) according to Artoxkit protocol (Artoxkit 1990).

Table 2. Testobjects and test conditions

Nr.	Testobjects	Test medium	Temperature
1.	*Desmodesmus communis*	Brackish	24ºC
2.	*Corophium volutator*	Brackish	15ºC
3.	*Monoporeia affinis*	Brackish	4ºC
4.	*Gammarus pulex*	Freshwater	4ºC
5.	*Hyalella azteca*	Freshwater	24ºC
6.	*Daphnia magna*	Freshwater	20ºC
7.	*Artemia salina*	Marine	20ºC

Figure 2. *Hyalella azteca* [17]

Figure 3. *Gammarus pulex* [18]

Figure 4. *Monoporeia affinis* [19].

Figure 5. *Corophium volutator* [20].

The acute toxicity (48 h and 96 h) of plastic products was detected using juveniles of 4 different amphipod species as test objects. Sampling of amphipods was performed in NW part of the Gulf of Riga, in the coastal zone of the Open Baltic Sea and in the Lake Liepaja. *Hyalella azteca* was obtained from the Culture Collection of Hayes, VA (North America Chesapeake). Before bioassays test objects were acclimatized under natural conditions. For each product leachate 3 replicates were made. Bioassays were performed in 250 ml beakers. For each replicate 8 zooplankton or zoobenthos individuals were used. Beakers were placed under different temperatures depending on species. After 48 h and 96 h live and dead individuals were counted and mortality of test objects was calculated.

Figure 6. *Desmodesmus communis* [16].

Green microalgae *Desmodesmus communis* (Figure 6) was obtained from the culture collection of Latvian Institute of Aquatic Ecology. Monoculture strain DCGR - 3 was isolated from the coastal zone of the Gulf of Riga (Eastern part of the Baltic Sea) and cultivated in BG - 11 medium. Three replicates were made for each plastic product and leaching samples were placed in 30 ml Nalgene centrifuge tubes. Duration of experiments was 72 hours. Intensity of algal growth was measured every 24 h at 640 nm by the Turner Fluorometer10-AU and specific growth rate was calculated.

Figure 7. *Daphnia magna*

Figure 8. *Artemia salina*

The acute toxicity (24 h, 48 h and 72 h) of 8 different plastic product leachates was detected using juveniles of 2 zooplankton species. *Daphnia magna* culture *was* obtained from the culture collection of Latvian Institute of Aquatic Ecology, but *Artemia salina* - from the resting eggs. Before bioassays test objects were acclimatized under natural conditions (Table 2).

There were made 3 replicates (*Daphnia magna*) and 5 replicates (*Artemia salina*) for each plastic product leachate and placed in 250 ml beaker. For each replicate 7 (*Daphnia magna*) and 10 (*Artemia salina*) individuals were used. Beakers were placed under different temperature depending on species. After 24 h, 72 h and 96 h live and dead individuals were counted and mortality of testobects was calculated.

Results and Discussion

Effects on green algae

The impact of 6 different plastic product leachates were tested on green microalgae *Desmodesmus communis* after 72 hours exposure (Figure 9). Results of acute standarttest (ISO 8692:2005) showed, that five of six tested product leachates have no toxic effects on cell development or growth rate of test culture, but dish sponges caused significant decrease in specific algal growth rate, indicating hazardous impact of this plastic product made from polyurethane (PU). In polyurethanes manufacture hazardous chemicals are used. PU is formed by reacting a polyether or polyester with an isocyanate usually in the presence of catalysts. Polyether besides is made from ethylene oxide or propylene oxide, which are both classified as carcinogenic and mutagenic, they may be very toxic, allergenic and may cause long term effects in the aquatic environment.

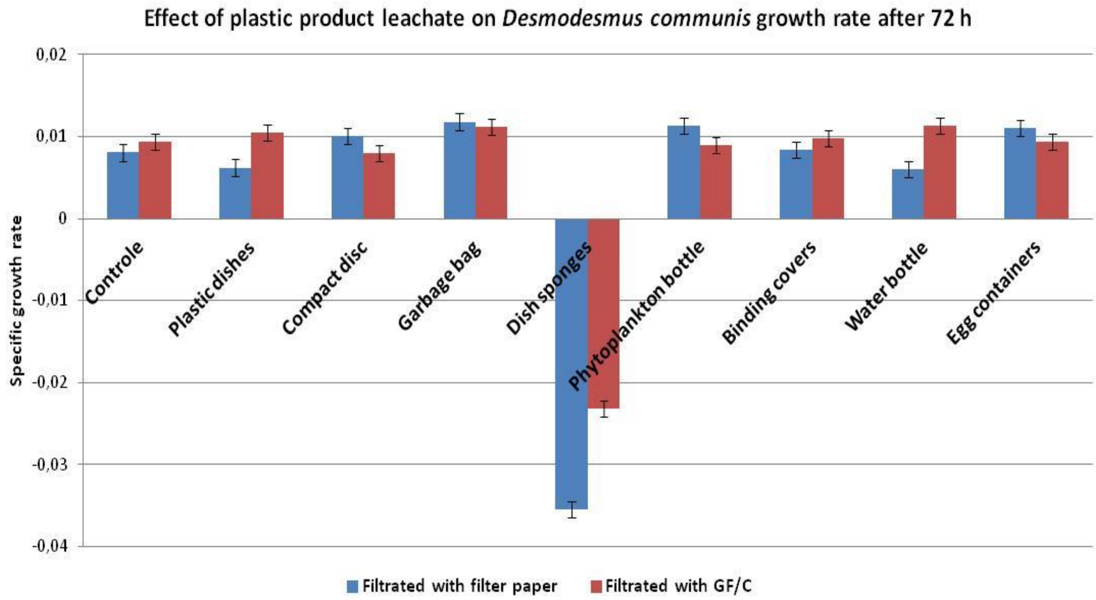

Figure 9. Effect of 8 different plastic product leachates on green algae *Desmodesmus communis* after 72 hours exposure.

Effect on zooplankton

60 % of all tested plastic products caused a negative impact on pelagic crustaceans *Daphnia magna* and *Artemia salina*. The most toxic from all tested plastic products were dish sponges (made from polyurithene PU). Freshwater amphipods *Daphnia magna* and marine water amphipods *Artemia salina* were extremly sensitive to the presence of "Dish sponges", evoking 70 – 100 % mortality after 24 – 72 hours exposure in dish sponge's leachates.

Inhibition of *Daphnia magna* and *Artemia salina* was observed also after the impact of drinking water bottles (made from polycarbonates PC), causing 30 – 68 % mortality.

Artemia salina was sensitive to the presence of compact discs (made from polyvinyle chloride - PVC).

The lowest negative impact was caused by leachates from plastic dishes (made from polypropylene - PP), from binding covers (polyvinyle chloride - PVC) and Plastic egg conteiners (polystyrene - PS).

Diverse results using different filtration methods reveal about the role of injection interpretation of experimental results (Fig10 and 11).

In general we can conclude that plastic products can cause a negative impact on frehwater and marine crustaceans and influence their sustainable development.

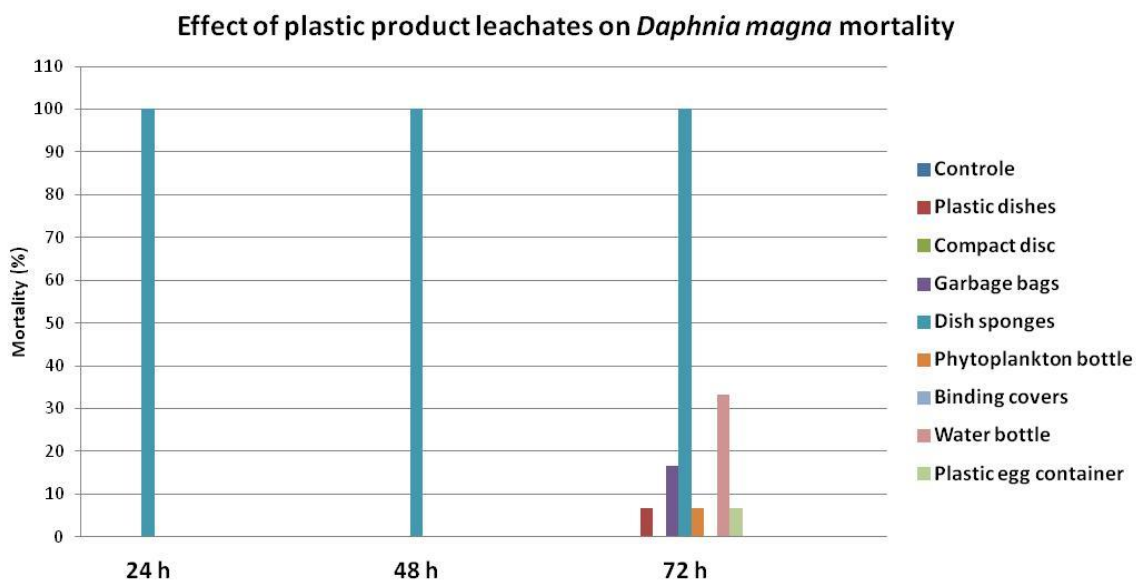

Figure 10. Effect of 8 different plastic product leachates on *Daphnia magna* mortality after 72 hours exposure (filtered with GFC/C).

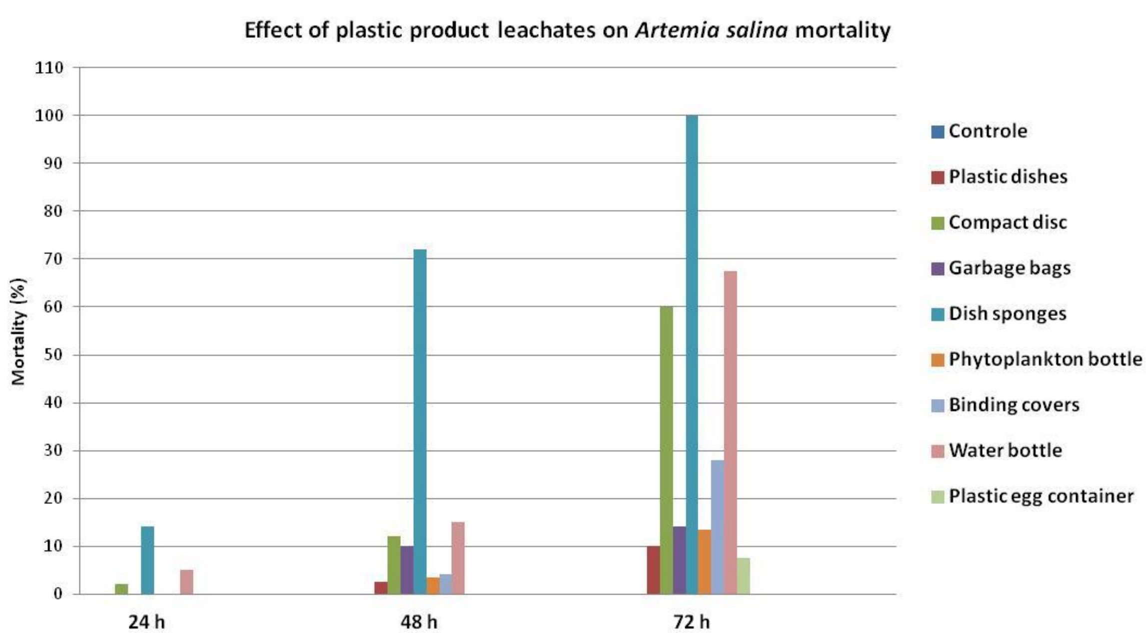

Figure 11. Effect of 8 different plastic product leachates on *Artemia salina* mortality after 72 hours exposure (filtered with GF/C).

Effects on benthic crustaceans

Dish sponge (PU) leachates caused 100% mortality of amphipods – *Gammarus pulex*, *Monoporeia affinis*, *Corophium volutator* and 50% mortality of *Hyalella azteca* after 96 hours of plastic product leachates impact (Figure 12).

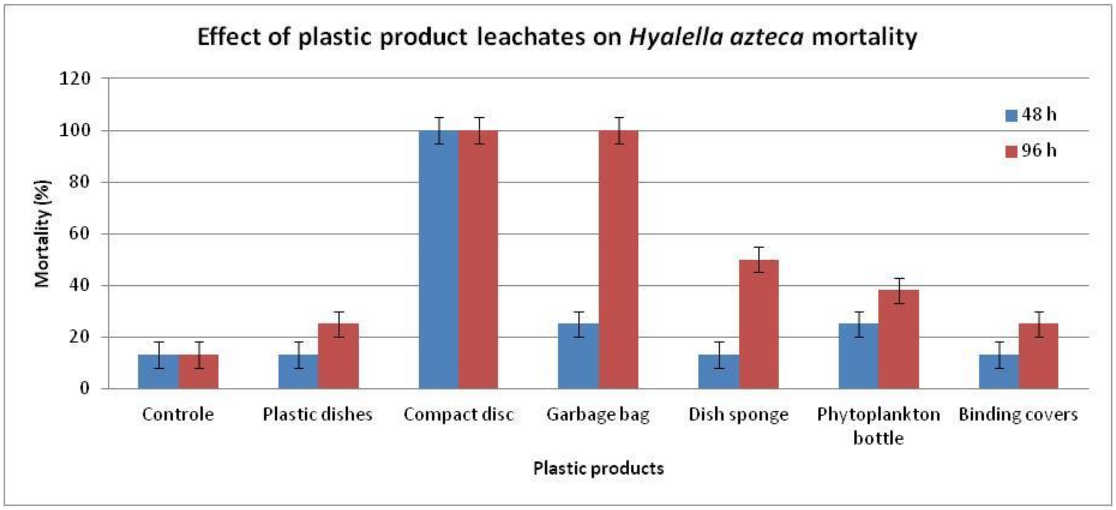

Figure 12. Effect of six different plastic product leachates on *Hyalella azteca* mortality after 48 and 96 hours exposure (filtered with filter paper).

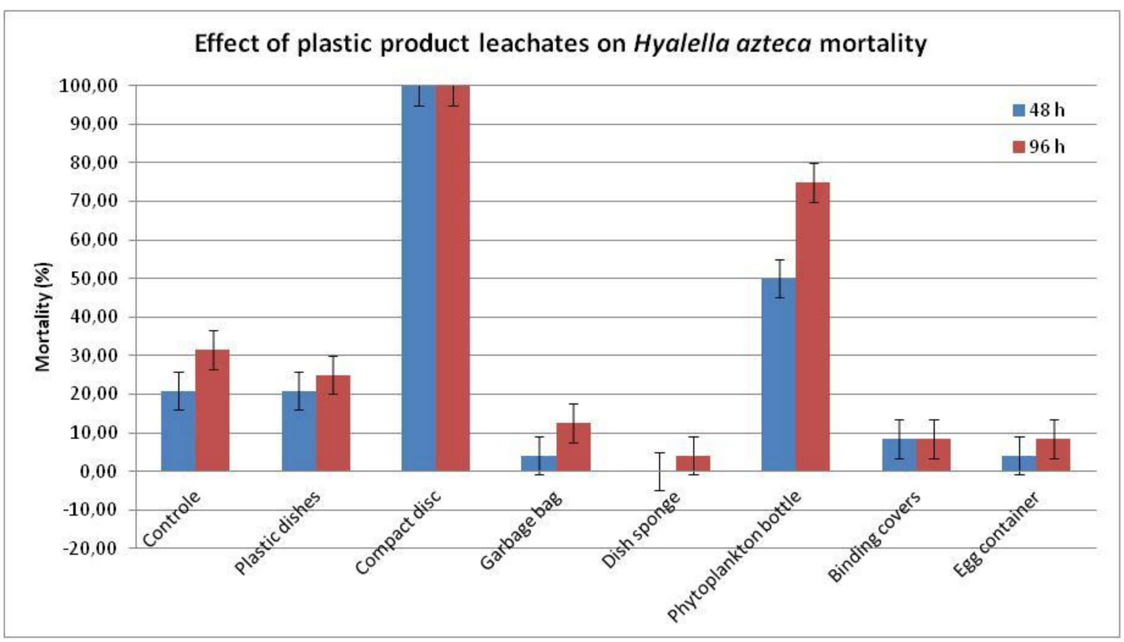

Figure 13. Effect of six different plastic product leachates on *Hyalella azteca* mortality after 48 and 96 hours exposure (filtered with GF/C).

As shown in Figure 12, after 48 hour exposure, garbage bag leachate caused 25 % mortality of *Hyalella azteca*, but after 96 hour exposure - 100 % mortality of individuals was observed. Garbage bags are made of low-density polyethylene (LDPE). Until now it was considered as one of a safer plastic, not toxic for aquatic environment. Research on risks associated with this tipe of plastic is ongoing. Probably the chemicals covering bag's surface causes negative effect to mentioned amphipod species. 60 % mortality after 96 h exposure in garbage bag leachate showed also *Monoporeia affinis*, but two other amphipod species didn't react to the presence of mentioned plastic product.

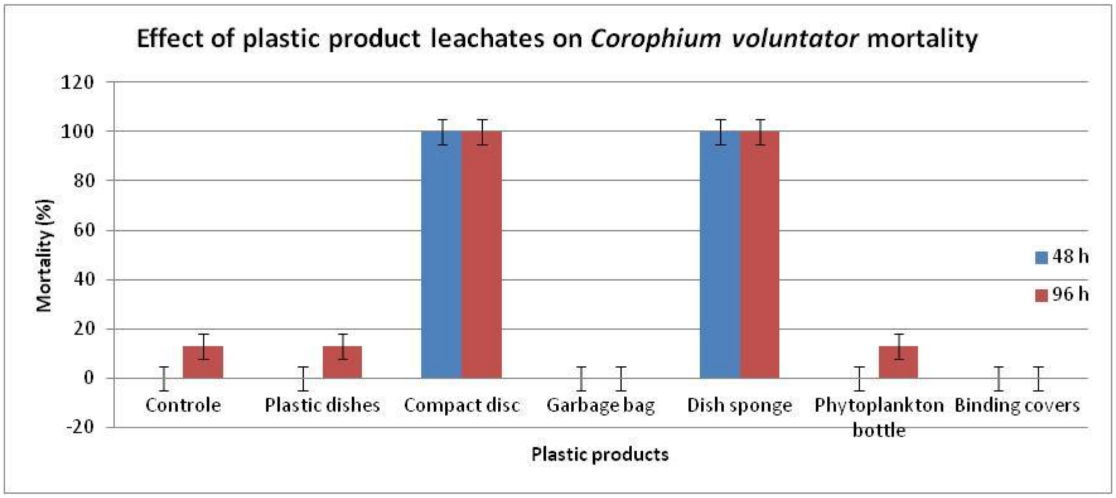

Figure 14. Effect of six different plastic product leachates on *Corophium volutator* mortality after 48 and 96 hours exposure (Filtered with filter paper).

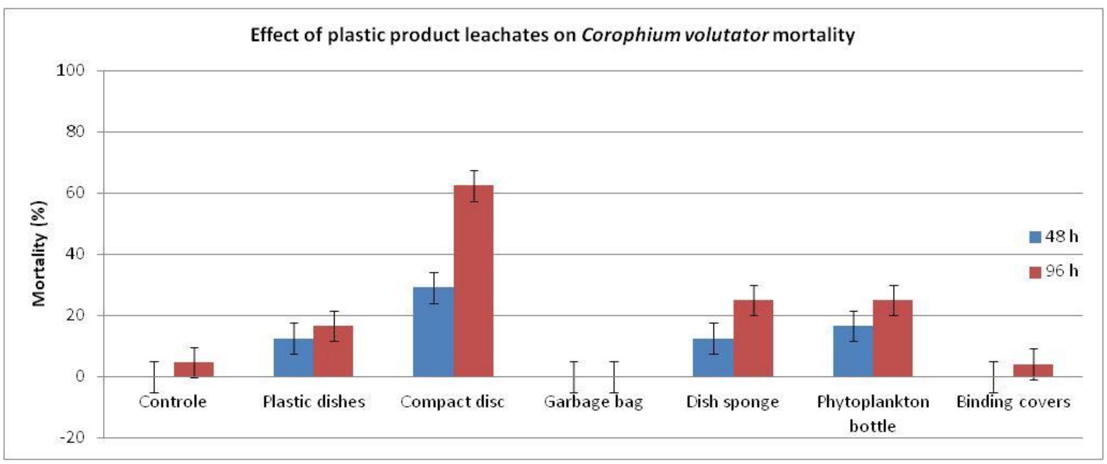

Figure 15. Effect of 6 different plastic product leachates on *Corophium volutator* mortality after 48 and 96 hours exposure (Filtered with GF/C).

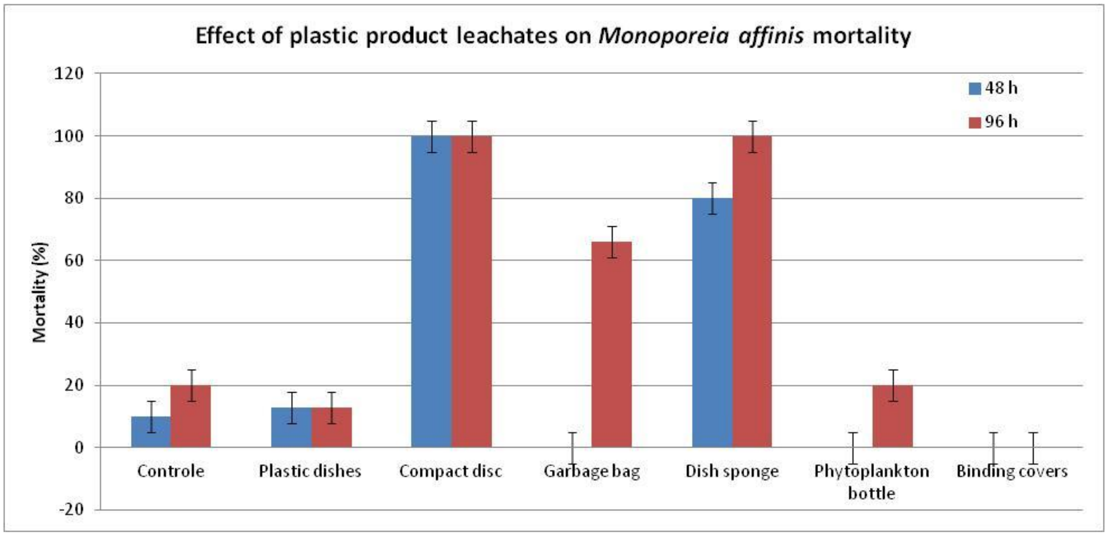

Figure 16. Effect of 6 different plastic product leachates on *Monoporeia affinis* mortality after 48 and 96 hours exposure (filtered with filter paper).

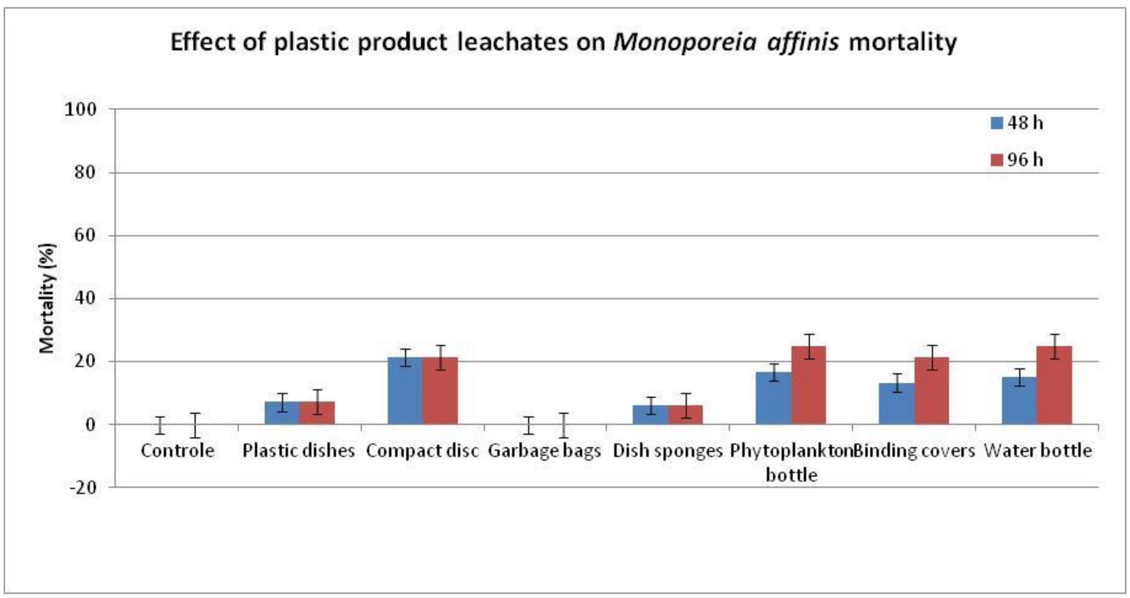

Figure 17. Effect of 7 different plastic product leachates on *Monoporeia affinis* mortality after 48 and 96 hours exposure (filtered with GF/C).

High toxicity showed also leachates of compact discs, made of polycarbonate plastic which on the one side is covered with four different layers. First there is thin layer of organic dye, consisting of an azo dye with metals and additives, on which there is a layer of pure silver, which is protected by a UV-cured lacquer and a top later of crystal lacquer (Lithner 2009). As test results show, compact disc leachates have toxic effect on all amphipod species, but no negative impact was observed on specific growth rate of green algae *Desmodesmus communis*. As Lithner (2009) explains, compact discs are made of polycarbonate, which does not cause toxic effect, but inhibition could be caused from the crystal lacquer and metals. Acute tests with amphipods, showed expressed toxicity of compact discs already after 48 hours of exposure.

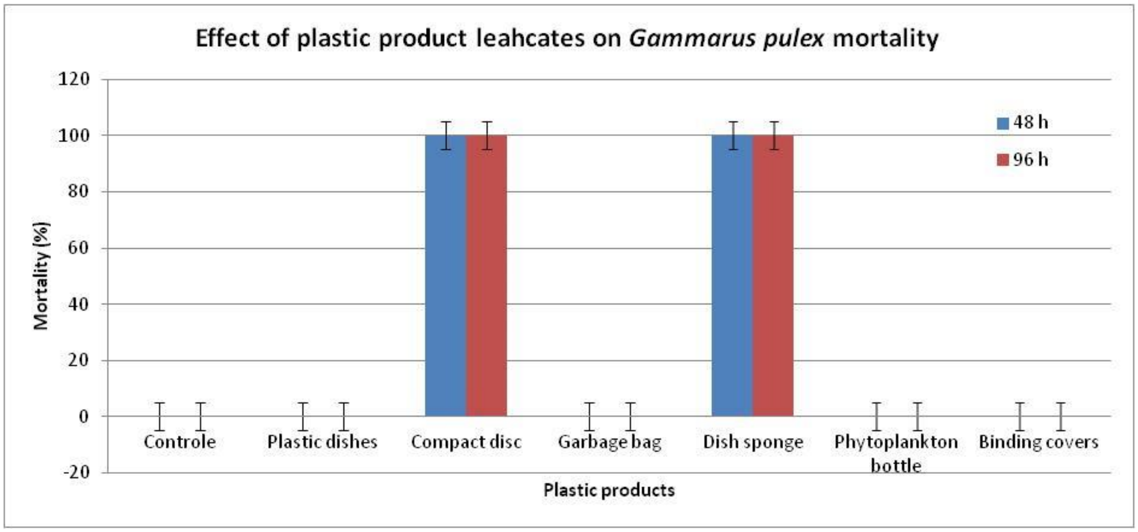

Figure 18. Effect of 6 different plastic product leachates on *Gammarus pulex* mortality after 48 and 96 hours (filtered with filter paper).

No toxic effect or low toxicity (*Hyalella azteca*) was observed at the presence of leachates from plastic dishes - made from polypropylene (PP), as well as from phytoplankton bottles and binding covers (polyvinyle chloride; PVC).

Diverse results using different filtration methods reveal about the role of injection interpretation of experimental results (Fig12 and 18).

CONCLUSIONS

- In general above 50% of tested plastic products caused a negative impact on majority of used testobjects
- The most toxic from all tested plastic product leachates were dish sponges made of polyurithene (PU or PUR) and compact discs made of polyvinyle chloride (PVC), causing 50 - 100% mortality of all testobjects
- Compact discs were toxic to amphipods, but didn't influence the growth of algae.
- Toxic were also leachates from garbage bags - made of low-density polyethylene (LDPE) and

showing even 100% morality of some amphipod's test species - *Monoporeia affinis* and *Hyalella azteca*.

- The lowest negative impact was caused by leachates from plastic dishes – (polypropylene; PP), as well as from phytoplankton bottles and binding covers (polyvinyle chloride; PVC).
- In comparisson to amphipods, algae are less sensitive to the presence of potentially toxic plastics.
- *Hyalella azteca* is one of the most sensitive amphipod's species against the impact of plastic product's leachates showing negative impact of all tested plastics
- In general we can conclude that plastic products can cause a negative impact on freshwater and marine organisms and influence their sustainable development.

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Estonian case study on coastal litter

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Description of field work

Investigations on coastal litter were performed in three sites at the Estonian coast with the following details (for study sites location, please see Figure 1). During the fieldwork planning and performing field works, HELCOM (2008) and EC (EC 2011) recommendations were followed.

Table 1. Detailed information on the three coastal litter sampling sites.

County	Location name	Baltic Sea basin	Coordinates	Coast width/length	Date	Site characteristics
Pärnu county	Häädemeeste	Gulf of Riga	Start: N57°54,265\` E024°22,246' End: N57º54,582\` E024º22,346\`	30-35/350 m	17.07.2012	Sand, exposed to the open sea
Pärnu County	Varbla	Gulf of Riga	Start: N58º20,715\` E023º44,644\` End: N58º20,529\` E023º44,789\`	35-40/390 m	19.07.2012	Gravel, rocks, exposed to the open sea; seaweed.
Lääne county	Nõva	Gulf of Finland	Start N59º14,337\` E023º 38,655\` End: N59º14,521\` E023º38,777\`	30-35/350 m	24.07.2012	Sand, exposed to the open sea.

Figure 1. Locations of sampling sites (red dots)

Sample processing and data interpretation

The collected material was analysed by the following major categories - plastic/polystyrene, paper/cardboard, rubber, glass, wood, rubber, cloth and metal – in terms of weight and numbers.

Results and Discussion

It appeared that the quantity of litter was much more stable across different sampling sites (variation 24.6-35.4 pieces per 100 m coastline) than the litter weight with the latter varying about three times (1.2 – 3.7 kg). The strongly dominating type of litter across all three study sites was plastic/polystyrene with 20.3-24.9 pieces per site (making between 70 and 86% of the total litter). The key results of this case study can be summarised in the Table 2.

Table 2. Results of the coastal litter sampling in three sites at Estonian coast. Litter weight is given in grams either in wet weight (w/w) and/or dry weight (d/w) per 100 m coastline. In case wet and dry weights are similar, no unit is given. Litter number (per 100m coastline) is indicated in pieces.

Item	Häädemeeste	Varbla	Nõva
20 01 39 PLASTIC/ POLYSTYRENE	2 558.1 (w/w) /20.3 pieces 1 339 (d/w)	709,2 / 20.3 pieces	1 546 / 24.9 pieces
20 01 01 PAPER/ CARDBOARD	19.5 / 1.1 pieces	23.3 / 1.0 pieces	6.4 (w/w) / 2.9 pieces 4.1 (d/w)
20 01 02 GLASS		140.0 / 0.8 pieces	64.9 / 0.6 pieces
20 01 38 WOOD(MACHINED)		305.1 / 0.3 pieces	364.3 / 3.4 pieces
20 01 11 RUBBER	2.3 / 0.9 pieces	0.3 / 0.3 piece	58.3 / 1.4 pieces
20 01 11 CLOTH	375.7 / 1.4 pieces	52.1 / 0.5 pieces	1 648.6 (w/w) / 0.6 pieces 1 154.3 (d/w)
20 01 40 METAL	23.1 / 0.9 pieces	4.6 / 0.5 pieces	23.1 / 1.7 pieces
	2 978.8 g (w/w) **1 759.6 g (d/w)** **24.6 pieces**	**1 234.7 g** **23.6 pieces**	**3 711.6 g (w/w)** **3 215.1 g (d/w)** **35.4 pieces**

The Baltic Marine Litter project (MARLIN, 2011-2013), funded by the Central Baltic Interreg IVA programme) focuses on activities to raise awareness on marine litter as well as increased knowledge on amounts, sources, types of litter and how to mitigate the negative effects of marine litter. One of the main activities in MARLIN is to try out and evaluate the UNEP method and protocols. MARLIN has planned to carry out beach litter assessments in 20 key areas in Sweden, Finland, Estonia and Latvia (in total 120 beach litter assessments) with local landowners, organisations and municipalities to be trained in performing the assessments (http://www.projectmarlin.eu/sa/node.asp?node=3005). Far the most common litter item found within the MARLIN project was similar to the results of our study – plastic (by ca. 56%). However, this is lower than found in our study (70-86%). The number of litter per 100 m coastaline was substantially lower in our study than that found in the MARLIN project (23.6 vs. 136.7, respectively) http://www.projectmarlin.eu/documents/MARLIN/LitterReport_BalticSea2012_10_04.pdf.

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