Determination of legacy pollutants and emerging contaminants in the marine environment of Black Sea

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Abstract

The objectives of this project was to describe and document the degree of contamination and the effects of pollution by hazardous substances in the Black Sea, mainly its Eastern (Georgian area) and Western (Ukrainian area) sides. An in-depth contamination survey was carried out based on the profiling of WFD priority substances (2013/39/EC) and the screening of potential Black Sea Specific Pollutants in different environmental compartments (seawater and sediments) and marine organisms (mussels and fish). Target, suspect and non-target screening approaches were followed for the detection of priority pollutants and emerging contaminants, while advanced software and sophisticated tools were used for results' extraction and toxicity prediction. Analysis results indicated that the Ukrainian samples and especially those that were withdrawn closest to the Danube and Rioni estuaries were by far the most polluted ones. Moreover, risk assessment revealed an alarming chemical status concerning the presence of PAHs and OCPs in specific pollution "hot spots" in the Black Sea. Toxicity prediction results of emerging contaminants emphasize the need for monitoring of transformation products (TPs) with wide-scope screening techniques and that TPs need to be included in risk assessment studies.

1. Introduction

Monitoring activities over several decades have revealed the ubiquitous presence of organic micro-contaminants and trace metals in all compartments of the marine environment (water, sediment and biota).

The Water Framework Directive (WFD) (2000/60/EC) sets a comprehensive management planning system to help protect and improve the ecological and chemical status of European water bodies. This is underpinned by the use of environmental quality standards (2013/39/EU) to help assess risks to the ecological quality of water environment and to identify the scale of improvements that would be needed to bring waters under pressure back into a good condition. The chemical pollutants that are regulated under international legislation represent a very small fraction of the universe of chemicals that occur in the environment as a result of human activities [Daughton (2004)]. According to NORMAN (Network of reference laboratories, research centers and related organizations for monitoring emerging environmental substances), emerging contaminants (ECs) are compounds that are not included in routine environmental monitoring programs and may be candidates for future legislation due to their adverse effects and/or persistency (http://www.norman-network.net/). ECs encompass a diverse group of compounds, including *i.a.* pharmaceuticals and personal-care products (PCPs), psychotropic drugs, drugs of abuse, steroids and hormones, endocrine disrupting compounds, surfactants, perfluorinated compounds, pesticides, phosphoric ester flame retardants, industrial additives and agents (e.g., benzotriazoles and benzothiazoles), siloxanes, artificial sweeteners, gasoline additives and the transformation products of the aforementioned groups of compounds [Bletsou (2015)].

There is a concern that these compounds may have an impact on aquatic life and thus they may be candidates for future legislation due to their adverse effects and/ or persistency. Although ECs include a wide range of compounds, only a small proportion of the chemical compounds have been sufficiently monitored in the water bodies [Loos (2013)]. The development of high resolving power mass analyzers (HRMS) has contributed a lot towards the wide-scope screening of emerging contaminants. HRMS full scan acquisition offers the possibility of retrieving all the information concerning the analytes in post-acquisition approaches. Moreover, for evaluating their probable negative effects in human life and ecosystem, toxicity assessment performance is needed. Novel prediction models are used for extracting the harmful levels of pollution for every tested contaminant.

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Specific Pollutants in different environmental compartments (seawater and sediments) and marine organisms (mussels and fish). Several analytical methods and novel techniques were applied for the analysis of the samples. Target, suspect and non-target screening approaches were followed for the detection of priority pollutants and emerging contaminants, while advanced software and sophisticated tools were used for results' extraction and toxicity prediction. Moreover, the risk of the identified emerging contaminants on the aquatic environment was evaluated following the EU risk assessment guidelines (Directive 93/67/EEC, 1488/94/EC, Directive 98/8/EC).

2. Sampling and Methods

The study area for contamination state was covered by three transects of Black Sea; the western side close to Ukraine, the eastern Black Sea close to Georgia and the central side including (open sea) sampling points across the length of Black Sea and out of reach of any coastal city. Seawater and sediment samples close to Georgia (GE) and Ukraine (UA) were obtained during the National Pilot Monitoring Studies (NPMS), while the Open Sea samples during Joint Open Sea Surveys (JOSS). Thirty three seawater samples (ten JOSS, nine NPMS-UA and fourteen NPMS-GE) and nineteen sediment samples (five JOSS, eight NPMS-UA and six NPMS-GE) in total were collected and analyzed.

Priority pollutants and emerging contaminants may be present at ng or even pg/L in seawater samples. Thus, the most common procedures used to carry out the determination of organic compounds in aquatic environmental matrices applied sample pre-concentration steps, such as solid-phase extraction (SPE) or liquid-liquid extraction (LLE) followed by separation and determination using liquid (LC) or gas chromatography (GC), coupled with mass spectrometry (LC–MS or GC–MS). Tandem mass spectrometry (MS/MS) offers higher performance than single-quadrupole instruments, in terms of sensitivity and selectivity. Moreover, the screening of known and suspect emerging contaminants is performed by LC- and/or GC- high resolution MS and post-acquisition data treatment workflows. ICP-MS was used for the determination of metals and arsenic after acidification (for seawater) or microwave digestion and dilution (for sediments) and on-line addition of internal standards.

Apart from the 45 (groups of) priority substances that are included in the Environmental Quality Standard Directive (EQS Directive 2008/105/EC, updated by Directive 2013/39/EU), 2041 additional emerging contaminants were screened by an in-house LC-QToFMS screening method, developed at the Laboratory of Analytical Chemistry of the National and Kapodistrian University of Athens. Organic compounds of several classes, like pharmaceuticals, veterinary drugs, industrial chemicals, drugs of abuse, naturally occurring compounds, pesticides, stimulants etc., as well as their transformation products (TPs) were included in the screening list for applying a holistic target screening approach on environmental basis.

Acute toxicity data were estimated by using two different prediction models. The predictive ECOSAR software [USEPA (2012)] was used for three different trophic levels (daphnids, fishes and green algae) to evaluate the potential risk of the identified contaminants with no EQS, individually, in the aquatic environment. The second model used was ToxTrAMS [Aalizadeh *et al* (2017)]. ToxTrAMS is an in-house robust quantitative structure–toxicity relationships model that accurately estimates the acute toxicity in *daphnia magna*, *pimephales promelas* (fish) and *pseudokirchneriella subcapitata* (algae) with a wide and defined applicability domain.

3. Results and Discussion

A. Target screening

Seawater samples

The Ukrainian samples closest to the Danube estuary (UA 5 & 7) were by far the most polluted ones. The maximum number of pollutants (129 in total) were detected in UA 7 sample. Both the number of detected compounds and the % pollution seem to gradually decrease from the Danube estuary to the Open Sea. The seawater samples close to Anaklia (GE 13-15) presented also a high level of pollution and an elevated number of total detected compounds. The less polluted sample was UA 13 (both total number of pollutants and total pollution), taken close to Dnieper estuary.

Sediments

Many pollutants, including a large number of hazardous compounds, are hydrophobic and their environmental behavior varies markedly between sorbed and dissolved states. Sediments are known to effectively sequester hydrophobic chemical pollutants entering water bodies such as estuaries.
The Ukrainian samples were by far the most polluted. The maximum number of pollutants (40 in total) and the maximum % pollution were noticed in UA 6 and 5, respectively. There is a clear specific trend of pollution. Coastal and shelf sediment samples are highly polluted, whereas Open Sea sediment samples are significantly less polluted. UA 7 & 5 were the most polluted sampling points, close to the Danube estuary, taking into consideration the results of analysis of both seawater and sediment samples.

Biota

The extent of contamination in the marine organisms of Black Sea was also investigated. Fish and shellfish tissue monitoring serves as an important indicator of contaminated sediments and water, and many states routinely conduct tissue analyses as a component of their comprehensive environmental monitoring programmes. Two different marine organisms, mussels and fish of different species were collected and analyzed. Mussels samples were collected during the NPMS/JOSS GE-UA, fish samples were collected by ONU in the Odessa bay and Zmiinyi Island in Ukraine and by NEA in Batumi - Gonio in Georgia. The Guidance document on chemical monitoring of sediment and biota under the WFD suggests that analyses should be done in the whole fish, however, several EU Member States propose to analyse the tissue. This latter also seem to match better the requirements of the MSFD. To be on the safe side, both fish tissue and whole fish samples were analysed.

A trend of higher pollution in the whole fish matrix compared to the fish muscle tissue, was noticed in three of the four biota samples where both matrices were analyzed.

The Georgian fish sample from Batumi (whole body matrix) is by far the most polluted one, while the maximum number of detected priority substances and emerging contaminants was observed in the mussels sample from Batumi.

B. Non-target screening

Successful non-target identifications are summarized in 4 groups; surfactants, phosphates, compounds with oil origin and industrial chemicals.

The “most polluted” sampling sites seem to be UA 1 (near Odessa), GE 11 (near the city of Poti and Rioni River) and JOSS 12 (the sampling site between Turkey and Crimea). These findings led to the conclusion that Rioni River (although small comparing to other rivers, such as Danube) may contribute significantly to the pollution of the Black Sea. UA 1 site is affected by input from land (city of Odessa), shipping activity and may also be affected by the three major rivers (Dniester, Dnieper and Danube). JOSS 12 as the third “probably most polluted” seawater sample was an unexpected result. This station may be influenced by shipping activity and probable currents that may circulate the pollution. This is also supported by the fact that rest of JOSS samples do not look “less contaminated” in terms of number of compounds and total intensity when compared with the other sampling sites.

The initial hypothesis that the Danube River is one of the major pollution hotspots of the Black Sea is supplemented with new findings through this analysis which unequivocally excludes contribution from additional significant pollution sources. A pollution link between the Danube and the Black Sea was studied in more detail using non-target screening data from the Joint Danube Survey 3 organized by the ICPDR in 2013 [Liška (2015]).

Components with continuously decreasing response in space were investigated. Spatial generalized additive model (GAMs) was used to predict the response near the sampling stations. Out of the 20,000 components, 633 were found to comply with the prioritization rule of decreasing response in space. 30 out of these 633 compounds (26 at level 1 from target screening, 5 at level 3 [Schymanski (2015]) were identified (or tentatively identified for level 3).

C. Risk assessment

The results are alarming for the chemical status regarding the presence of PAHs and OCPs in specific pollution “hot spots” in the Black Sea. EQS are not yet established for ECs. However, some ECs (e.g. Terbuthylazine, Terbuthylazine desethyl, Valsartan) showed RQs higher than one, implying the need for more systematic monitoring. Additional experimental data regarding the environmentally relevant concentrations of these ECs seems to be urgent, as it affects the calculation of RQs. The detection of desethyl-terbuthylazine, a TP of terbuthylazine, with RQ >1, emphasizes the need for monitoring TPs with wide-scope screening techniques. TPs need to be included in risk assessment studies, as the transformation of the parent compound does not necessarily imply attenuation of their ecotoxicological risks.

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