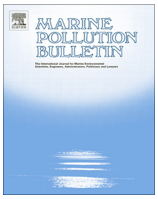
Marine Pollution Bulletin 87 (2014) 338–344

Contents lists available at ScienceDirect

Marine Pollution Bulletin

journal homepage: www.elsevier.com/locate/marpolbul



Baseline

Monitoring polycyclic aromatic hydrocarbons in the Northeast Aegean Sea using *Posidonia oceanica* seagrass and synthetic passive samplers

Maria-Venetia Apostolopoulou [a,*], Els Monteyne [b], Konstantinos Krikonis [c], Kosmas Pavlopoulos [d], Patrick Roose [b], Frank Dehairs [a]

[a] *Analytical, Environmental and Geo-Chemistry and Earth System Sciences Research Group, Vrije Universiteit Brussel, Pleinlaan 2, B-1050 Brussels, Belgium*
[b] *Royal Belgian Institute of Natural Sciences, Management Unit of the North Sea Mathematical Models, MARCHEM, 3e & 23e Linieregimentsplein, B-8400 Ostend, Belgium*
[c] *Department of Mathematics, Aristotle University of Thessaloniki, 541 24 University Campus, Thessaloniki, Greece*
[d] *Department of Geography and Planning, Paris Sorbonne University, P.O. Box 38044, Al Reem Island, Abu Dhabi, UAE*

ARTICLE INFO

Article history:
Available online 7 August 2014

Keywords:
PAHs
Posidonia oceanica
Passive sampling
Mediterranean Sea
Aegean Sea
Alexandroupolis Gulf

ABSTRACT

The concentrations of 22 polyaromatic hydrocarbons (PAH) in *Posidonia oceanica* seagrass, sediments, and seawater from the Alexandroupolis Gulf in the Aegean Sea, were investigated from 2007 to 2011. Temporal trends of total PAH contents in *P. oceanica* and sediments were similar. PAH levels in seawater, sediments, and seagrasses generally decreased with increasing distance from Alexandroupolis Port. Leaves and sheaths of *P. oceanica* had higher PAH levels than roots and rhizomes. *P. oceanica* accumulates PAHs and has good potential as a bioindicator of spatiotemporal pollution trends. PAH concentrations were also examined using in situ passive seawater sampling and were compared to results of passive sampling in the laboratory using local sediments and seawater. Levels of high molecular weight PAHs assessed using passive samplers confirmed the decreasing gradient of pollution away from Alexandroupolis Port. Passive sampling also proved useful for investigating sources of PAHs in *P. oceanica* meadows.

Seagrass meadows are considered to be the most productive ecosystems in coastal waters (Hemminga and Duarte, 2000). *Posidonia oceanica* is the dominant endemic seagrass in the Mediterranean Sea, occurring until a depth of approximately 40 m.

Although many studies have highlighted the usefulness of *P. oceanica* as a recorder of ambient trace metal pollution (Maserti et al., 1988; Catsiki and Panayotidis, 1993; Malea, 1993; Malea et al., 1994; Warnau et al., 1995; Pergent-Martini, 1998; Delgado et al., 1999; Ferrat et al., 2002; Ancora et al., 2004; Balestri et al., 2004; Gosselin et al., 2006), much less information is available regarding the content of organic contaminants in this seagrass. However, the few studies that have reported on bioaccumulation of organic contaminants in *P. oceanica* tissues (e.g., Viso et al., 1993; Lewis and Devereux, 2009; Pergent et al., 2011) highlight the potential of this plant to serve as an indicator of organic pollutants in the Mediterranean Sea. For instance, Pergent et al. (2011) report increased PAH concentrations (Σ16 PAHs) in *P. oceanica* leaves from a site next to Toulon's military and industrial harbor (France) compared to a pristine site located in Calvi (Corsica). Organic contaminants, including PAHs, are lipophilic and can penetrate lipid membranes and thus cells (Clark et al., 1990; Cerniglia, 1993; Sikkema et al., 1995). In aquatic plants, these compounds accumulate in the thylakoid membranes of chloroplasts (Ren et al., 1994). Among organic pollutants, polycyclic aromatic hydrocarbons (PAHs) are of major concern because they are persistent and potentially genotoxic and carcinogenic. Anthropogenic PAHs can be pyrogenic or petrogenic in origin. Pyrogenic PAHs are formed from the incomplete combustion of organic matter (e.g., coal, petroleum, and wood), whereas petrogenic PAHs are derived mainly from crude oil and petroleum products such as kerosene, gasoline, diesel, lubricating oil, and asphalt. PAHs tend to accumulate in sediments and in biological tissues to concentrations that may have adverse effects on resident biota, and may accumulate along the food chain (Irwin et al., 1997; Gogou et al., 2000; Zadeh et al., 2010).

Passive sampling techniques have been used to analyze PAHs in sediments, water, and biota (Vrana et al., 2005; Smedes et al., 2010; Monteyne et al., 2013). Passive sampling involves the transfer of analyte molecules from the solution of interest to a receiving phase in the sampling device until equilibrium is reached. This technique relies on the difference in chemical potential between the sampled medium and the receiving phase. Silicone rubber

---

* Corresponding author. Tel.: +32 2 629 1265; fax: +32 2 629 3274.
E-mail addresses: mapostol@vub.ac.be (M.-V. Apostolopoulou), E.Monteyne@mumm.ac.be (E. Monteyne), krikonis.math@gmail.com (K. Krikonis), kpavlop@hua.gr (K. Pavlopoulos), P.Roose@mumm.ac.be (P. Roose), fdehairs@vub.ac.be (F. Dehairs).

http://dx.doi.org/10.1016/j.marpolbul.2014.07.051
0025-326X/

strips have been used as passive samplers because of their large affinity for PAHs and their ability to detect very low concentrations of pollutants (Vrana et al., 2005; Monteyne et al., 2013). Results for PAHs obtained via passive sampling compare well with those obtained from biomonitoring using organisms such as mussels (*Mytilus edulis*, Smedes, 2007, 2010; Smedes et al., 2010) and ragworms (*Nereis virens*, Yates et al., 2011).

The Alexandroupolis Gulf, located in the Northeast Aegean Sea, is subject to significant anthropogenic stress caused by inputs of organic pollutants (Kanellopoulos et al., 2007). Marine traffic in Alexandroupolis port increased between 2007 and 2011 (Hellenic Statistical Authority, 2013) and the number of vessels passing through the northern Aegean Sea doubled between November 2009 and October 2011 (Miliou et al., 2012). The United States Energy Information Administration (EIA, 2012) reported that transportation of crude oil and petroleum products through the Turkish straits between 2007 and 2010 reached 2.9 million barrels per day. In addition, according to the Turkish Marine Research Foundation (Oral and Öztürk, 2006), the number of oil tankers passing yearly through the Bosporus and Dardanelles straits has increased from 4200 in 1996 to 10,000 in 2010 and the quantity of oil transported through this area is estimated to be 145 million tons per year. Finally, the European Maritime Safety Agency reported that the number of shipping accidents in the Aegean Sea between Greece and Turkey has increased (EMSA, 2010).

In the present study, we assess the levels and sources of PAHs in coastal waters, sediments, and *P. oceanica* seagrass from the NE Aegean Sea and Evros River. For direct analysis of the different substrates for PAH content we also used passive samplers consisting of silicone rubber sheets to determine PAH levels. Our main objectives were to examine the potential of *P. oceanica* as an indicator of spatial and temporal trends of PAH pollution and to assess the utility of passive sampling as a monitoring tool.

Three study sites were selected in coastal waters and three sites in Evros River (Fig. 1, Table 1). Sites A, B, and C are located in Alexandroupolis Gulf, whereas site D stations were located at the downstream end of Evros River, outside (D1) and inside (D2 and D3) Evros Delta National Park. Details for each sampling site are listed in Supplementary Material 1. In total, 178 samples (80 marine and 45 river surface sediments, 41 *P. oceanica* tissues, and 12 water samples) were collected for PAH and bulk analyses. Marine sediments and *P. oceanica* seagrasses were collected by diving in 2007, 2009, 2010, and 2011 at sites A, B, and C. Note that seagrasses were absent at site C. The river stations (D1, D2, and D3) were sampled only in 2010 and 2011 (Table 1). At sites C and D the fine sediment fraction (<63 μm) represents >60% of total weight, while at sites A and B the fine sediment fraction is less (about 40%). Passive and water sampling was only performed in 2011. Details for the sampling procedures of sediments, *P. oceanica*, and water samples are presented in Supplementary Material 1.

Passive sampling at marine and riverine stations took place only in 2011. Silicone rubber strips composed of polydimethylsiloxane (PDMS, AlteSil Laboratory Sheet, Altec Products Ltd., Bude, UK) were used as passive samplers. These were suspended inside stainless-steel cages (40 × 25 × 35-cm commercial bird cages from which all plastic parts were removed) to protect the silicone rubber strips from mechanical damage after deployment. Ten cages were prepared, each containing three strips. The passive samplers were pretreated in the laboratory at the Management Unit of the North Sea Mathematical Models and the Scheldt Estuary (MUMM) following Booij et al. (2002) and Monteyne et al. (2013). Detailed procedures of the in situ and in vitro passive sampling are presented in Supplementary Material 2. PAHs were Soxhlet-extracted in the MUMM laboratory, which was accredited according to the ISO 17025 standard, and details are presented in Supplementary Material 3. Known concentrations of deuterated PAHs (naphthalene, acenaphthene, anthracene, pyrene, benz[a]anthracene, benzo[a]-pyrene, and indeno[1,2,3-cd]pyrene) were added as internal standards before extraction. The efficacy of the Soxhlet extraction procedure was verified by extracting duplicate samples with a pressurized liquid extraction (Dionex Thermo Scientific ASE 200). Chrysene was added as a recovery standard at the end of the procedure.

We investigated 22 PAHs, 16 of which are included in the United States Environmental Protection Agency priority list (EPA 610): naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benz[a]anthracene, chrysene, benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[a]pyrene, dibenz[a,h]anthracene, benzo[ghi]perylene, indeno[1,2,3-cd]pyrene (EPA, 2009); methylnaphthalene, biphenyl, 2,6-dimethylnaphthalene, 1-methylphenanthrene, benzo(e)pyrene, and perylene were measured in the passive samplers and in the different natural substrates sampled. Capillary gas chromatography (GC) coupled to a quadrupole mass spectrometer (MS) was used for method development and validation. The analytical method of GC is presented in in Supplementary Material 4.

Our recovery rates for PAH's from *P. oceanica* and sediments were between 65% and 124%, fitting the generally accepted range (i.e., between 40% and 120%; see Kimbrough et al., 2006). The recovery of the reference compounds by the passive samplers ranged between 81% and 112% for field samples and between 63% and 115% for the in vitro passive sampling experiments. The reported results were corrected based on the recovery rates of internal standards. The limits of quantification were 3.34 and 1.65 ng ml^{-1} for the in-situ and in vitro passive sampling experiments, respectively, and 0.49 ng g^{-1} (dry-weight basis [DW]) for sediments and seagrasses. Quality assurance data and validation parameters of the analytical methods are described in Supplementary Materials 5 and 6. For the various data conversions and calculations required to estimate the freely dissolved aqueous-phase concentrations from the passive samplers, we refer to Supplementary Material 7. Statistical analyses were performed using SPSS for Windows v.20 software (IBM, New York, USA) (see Supplementary Material 8).

To the best of our knowledge this is the first study to report on temporal trends of PAH concentrations for the Alexandroupolis Gulf. The total PAH concentrations ($\sum$22 PAHs) in sediments ranged from 5 to 49 ng g^{-1} DW for site A, with the highest median values observed in 2009 and 2010 (21 and 20 ng g^{-1} DW, respectively, Fig. 2a). At site B, PAH concentrations ranged from 5 to 81 ng g^{-1} DW with the highest median values also occurring in 2009 and 2010 (22 and 25 ng g^{-1} DW, respectively, Fig. 2b). At site C, PAH concentrations ranged from 25 to 223 ng g^{-1} DW, and the highest median values were recorded in 2010 (116 ng g^{-1} DW, Fig. 2c). Overall, the minimum $\sum$22PAH value was 5 ng g^{-1} DW (site A) and the maximum value was 223 ng g^{-1} DW (site C). The $\sum$22 PAH concentrations in marine sediments reported here overlap with the lower end of ranges reported by other researchers for the northern Aegean Sea: 88–807 ng g^{-1} DW in the Strymonian Gulf (Hatzianestis and Sklivagou, 2001), 44–166 ng g^{-1} DW in the Gulf of Kavala (Papadopoulou and Samara, 2002), and 23–746 ng g^{-1} DW in the Thermaic Gulf (Panagiotopoulos et al., 2009; Kapsimalis et al., 2010). PAH concentrations for the Evros River sites ranged from 6 to 727 ng g^{-1} DW with the highest values at the most downstream station D3 (Fig. 2d). Additionally, concentrations differed significantly among the 3 river stations ($p < 0.01$; Fig. 2d). The highest values were in agreement with published values for the Evros River Delta (628–980 ng g^{-1} DW; Kanellopoulos et al., 2007) and with other studies conducted in rivers in northern Greece, such as in the Strymon River (404–747 ng g^{-1} DW; Pavlidou et al., 2002).

Whole plant *P. oceanica* from site B had a similar temporal trend for $\sum$22 PAH concentrations and sediments (Figs. 2b and 3b); concentrations increased in 2009 and 2010 (the maximum median

Fig. 1. Study area: A: pristine site (extensive *P. oceanica* meadows); B: flood-prone site (extensive *P. oceanica* meadows); C: Alexandroupolis Port; and D: Evros River (*P. oceanica* meadows are absent here).

Table 1
Year, location and sample types.

Year	Sampling site	Sample type
2007	A, B	Sediment, *P.oceanica*
	C	Sediment
2009	A, B	Sediment, *P.oceanica*
	C	Sediment
2010	A, B	Sediment, *P.oceanica*
	C	Sediment
	D1, D2, D3	Sediment
2011	A, B	Sediment, water, *P.oceanica*, passive sampler
	C	Sediment, water, passive sampler, water and sediments for in vitro passive sampling experiment
	D1, D2, D3	Sediment, water, passive samplers, water and sediments for in vitro passive sampling experiment

value for *P. oceanica* was 1770 ng g^{-1} DW) and then decreased slightly in 2011. For site A, we note that for sediments (Fig. 2a) and whole plant *P. oceanica* (Fig 3a) alike, the lowest PAH concentrations were recorded in 2007 (Supplementary Material 9). Overall, the temporal trend of PAHs in *P. oceanica* tissues reflects is similar to that for sediments (Figs. 2–4), which highlights the usefulness of seagrasses as pollution indicators, because the PAH levels expressed per dry weight of substrate are higher for seagrasses than for sediments, and thus easier to determine.

PAH concentrations in sediments from sites A and B did not differ significantly and had median values of approximately 20 ng g^{-1} DW. Additionally, PAH contents in *P. oceanica* from sites A and B were not significantly different ($p > 0.05$; Supplementary Material 9), which would initially suggest a low level of pollution in those locations. However, seawater in 2011 showed increasing concentrations of $\sum$21 PAHs from site A to C (note that naphthalene is not considered in the analysis of seawater; Fig. 5; also see below), revealing a clear spatial gradient in the degree of contamination.

We also expressed the PAH contents in the sediments and seagrasses relative to the organic carbon (POC) contents (Fig. 4). The methods used and the data set for analysis of C and N in bulk sediments and *P. oceanica* tissues are presented in the Supplementary Material 10. The sediment POC content ranged from 0.7 mg g^{-1} DW (site A) to 7.2 mg g^{-1} DW (site C), whereas the average *P. oceanica* POC content was 371 ± 174 mg g^{-1} DW. It appears from Fig. 4 that PAH concentrations in sediments and seagrasses, when expressed relative to POC content, are similar, but sediments generally have higher PAH/POC ratios than seagrasses.

Because several studies have reported that seagrass roots and rhizomes exhibit lower levels of pollutants than leaves and sheaths, which are the most exposed parts of the plant (Lacerda and Resende, 1986; Ward et al., 1986; Lewis and Devereux, 2009), we also analyzed the PAH contents of different *P. oceanica* tissues (leaves, sheets, roots, rhizomes). The concentrations of Σ22PAHs ranged from 364 to 4072 ng g^{-1} DW, 769 to 2153 ng g^{-1} D, 135 to 1818 ng g^{-1} DW, and 54 to 825 ng g^{-1} DW in the leaves, sheaths, roots, and rhizomes, respectively (Fig. 6). There were significant differences in total PAH content between the leaves and rhizomes ($p = 0.001$), leaves and sheaths ($p = 0.013$), roots and sheaths ($p = 0.043$), roots and rhizomes ($p = 0.01$), and sheaths and rhizomes ($p = 0.001$) Supplementary Material 11). Therefore, it appear that uptake of PAHs into *P. oceanica* indeed occurs mainly via the water-to-leaf (epiphyte) pathway.

Naphthalene was not measured in passive samplers and seawater because it is a very volatile and fugacious compound and may be easily contaminated during sample handling. Most blank samples

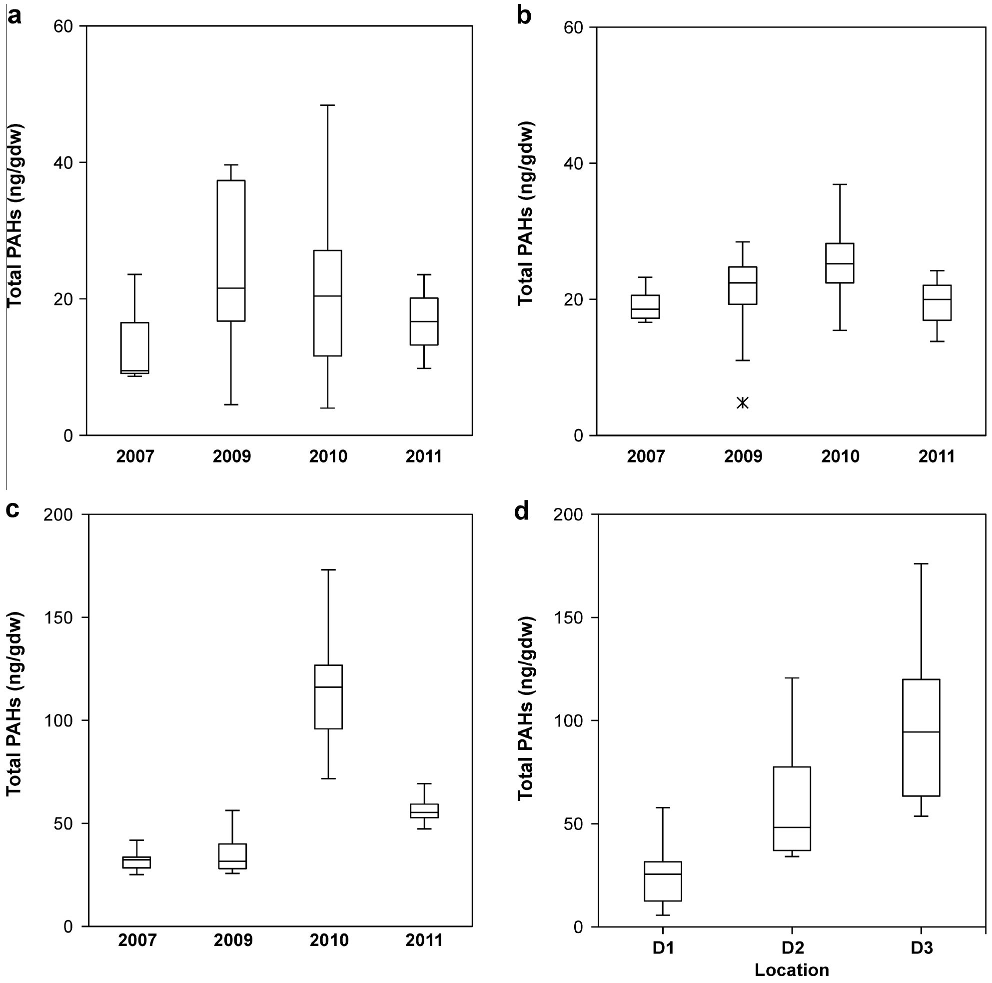


Fig. 2. Temporal trends in PAH concentrations (ng g^{-1} dw) in sediments from coastal sites A (a), B (b), and C (c); Spatial trends in Evros River sediments from sites D1 to D3 (d) (note that for Evros River data are only available for 2010 and 2011). The box and whisker diagram illustrates the spread of a set of the data, including the median and upper and lower quartiles. Twenty-two PAHs were measured: naphthalene (Na), acenaphthylene (Ac), acenaphthene (Acn), fluorine (Fl), phenanthrene (Phe), anthracene (An), fluoranthene (Fa), pyrene (Py), benz[a]anthracene (B(a)A), chrysene (Chr), benzo[b]fluoranthene (B(b)F), benzo[k]fluoranthene (B(k)F), benzo[a]pyrene (B(a)P), dibenz[a,h]anthracene (D(a,h)A), benzo[ghi]perylene (B(ghi)P), indeno[1,2,3-cd]pyrene (IP), methylnaphthalene (1-Mna), biphenyl (Bph), 2,6-dimethylnaphthalene (2,6-dMna), 1-methylphenanthrene (1-Mph), benzo(e)pyrene (B(e)P), and perylene (Pyl).

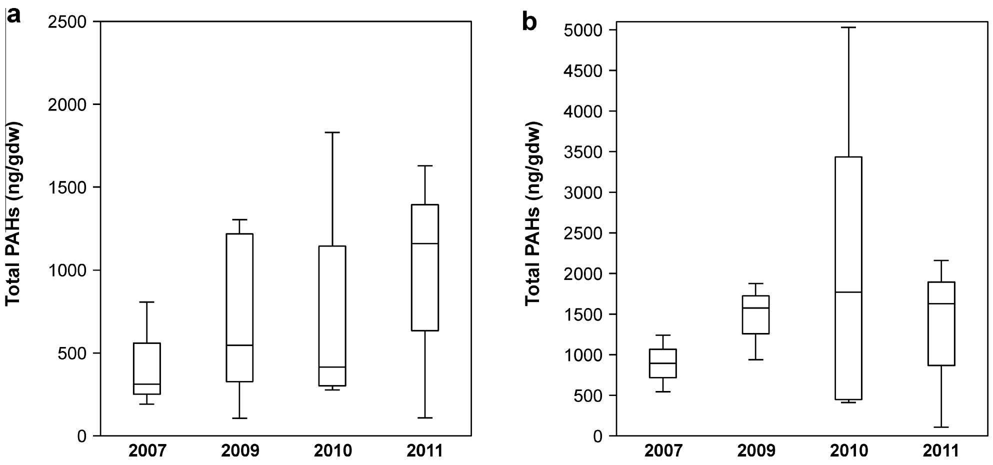


Fig. 3. Temporal trends of PAH concentrations (ng g^{-1} dw) in *P. oceanica* leaves at sites A (a) and site B (b).

exhibited higher concentrations and standard deviations for naphthalene than the deployed samplers. However, because naphthalene is moderately hydrophobic and has a tendency to adsorb onto particulate matter and accumulate in biota (Bates et al. 1997), higher levels do occur in the latter substrates, making high blank values less of a problem there. Freely dissolved concentrations of $\sum 21$ PAHs (i.e., without naphthalene) obtained from passive samplers ranged from 55.1 ± 7.6 to 61.5 ± 4.2 ng L^{-1} ($p > 0.05$,

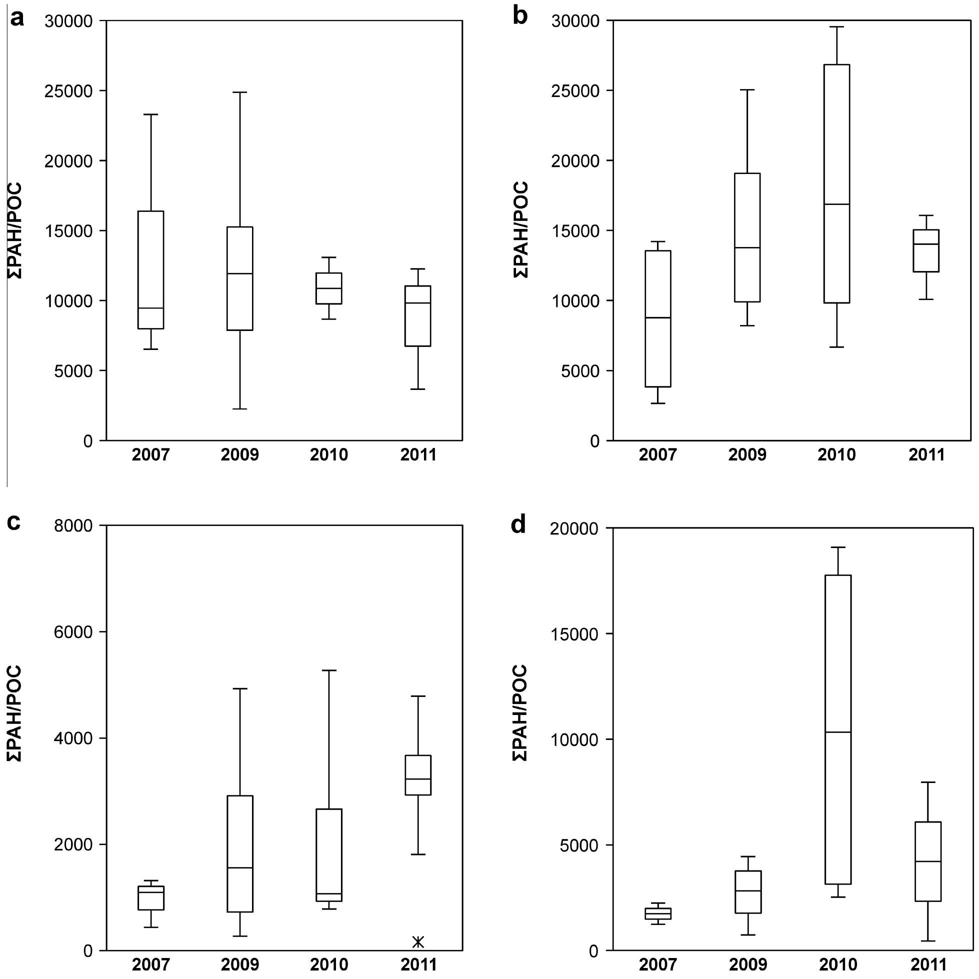


Fig. 4. Temporal trends of PAH concentrations relative to POC content (ng g^{-1} POC) in sediments from sites A (a) and B (b), and in bulk *P. oceanica* tissue from sites A (c) and B (d).

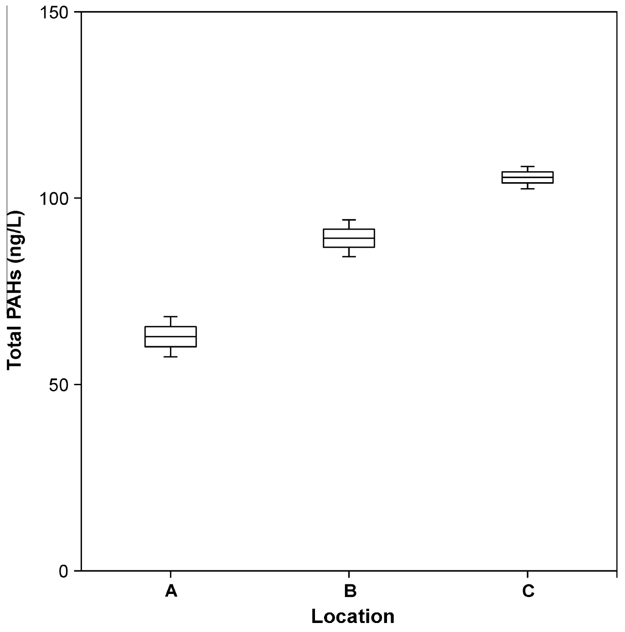


Fig. 5. Spatial trends in PAH concentrations (ng L^{-1}) in seawater in 2011.

Fig. 7). The highest levels were observed at station C near Alexandroupolis Port for both passive sampling and direct surface water analysis (note that seawater was not filtered before analysis). PAHs are hydrophobic and bind readily to suspended material, with the highest suspended matter POC concentrations (up to 10.3 μmol l^{-1}; see Supplementary Material 10) observed at site C. The POC concentrations of suspended matter at site C approached the values for the Evros River, which is a major supplier of particulate material to the Alexandroupolis Gulf (e.g., Pehlivanoglou 1989; Kanellopoulos et al., 2007). In addition, at site C, the sediment granulometry is relatively fine (Pehlivanoglou, 1989; Karditsa and Poulos, 2007) and thus more easily prone to resuspension, thereby offering further substrates for PAH accumulation. Because the $\sum$21 PAHs were measured in unfiltered surface water, any compounds associated with suspended particulate matter would have contributed to the total analysis, which may explain why PAH concentrations from bottom-tethered passive samplers were generally lower than those measured directly in surface waters. In addition, surface waters may have acquired higher concentrations from hydrocarbon pollution resulting from recent boat traffic. Because the passive samplers were deployed 10 m below the surface, they may not have detected recent surface water pollution. In contrast, *P. oceanica* and passive samplers at sites A and B are expected to be similarly exposed to pollutants because they are both in contact with bottom waters and also because the water column there is generally well mixed because of high turbulence (Xeidakis et al., 2007; Athanasoulis and Skarsoulis, 1992; Karditsa and Poulos, 2007).

Inspecting the results of individual PAHs the high-molecular-weight (HMW) PAHs using two-way multivariate analysis of variance, reflected the spatial differences better in passive samplers

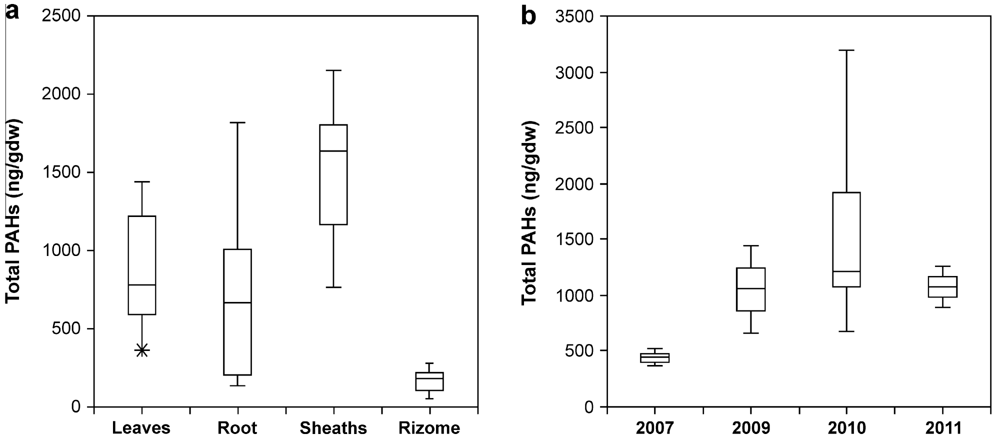


Fig. 6. Total concentration of 22 PAHs (ng g^{-1} dw) in *P. oceanica* leaves, roots, sheaths, and rhizomes (a); temporal evolution of 22 PAHs in *P. oceanica* leaves (b).

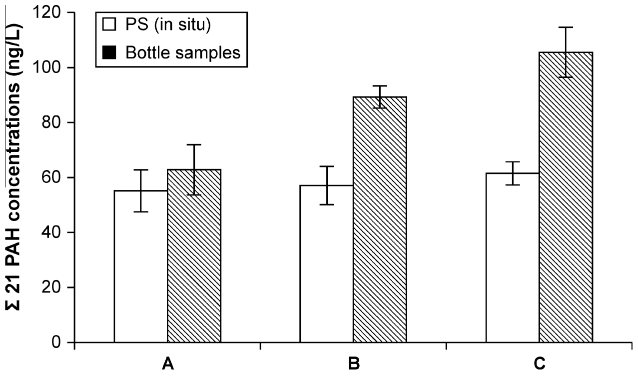


Fig. 7. Average concentrations (ng L^{-1}) of 22 dissolved PAHs measured using bottom-tethered in-situ passive samplers and measured directly in surface seawater.

than in sediments, *P. oceanica* tissues, and seawater. Specifically, in-situ exposure to seawater via passive samplers indicated that concentrations of the following HMW PAHs differed significantly between sites: benz[a]anthracene ($p = 0.025$), benzo[b]fluoranthene ($p = 0.017$), benzo[k]fluoranthene ($p < 0.001$), perylene ($p < 0.001$), dibenz[a,h]anthracene ($p = 0.049$), and benzo[ghi]perylene ($p < 0.001$). For benzo[b]fluoranthene, benzo[k]fluoranthene, and perylene, this spatial variability was confirmed from the in vitro experiments with exposure of the passive samplers to freely dissolved concentrations of PAHs in sediment–seawater suspensions ($p = 0.005$, $p = 0.040$, and $p < 0.001$, respectively). Because HMW PAHs are less volatile and less water soluble than low-molecular-weight compounds, they may have a high affinity for exposed surface structures such as the silicone rubber sheets of the passive samplers, thus yielding a better indication of the concentrations of freely dissolved HMW PAHs in ambient water.

In contrast, few HMW PAHs in sediments, *P. oceanica* plants, and seawater showed significant differences between sites. Only benzo[b]fluoranthene ($p = 0.039$) and perylene ($p < 0.001$) were present at higher levels in sediments from pristine areas (A and B) than in sediments from polluted areas (C and D) (Supplementary Material 12). Similarly, the benzo[b]fluoranthene and benzo[k]fluoranthene content in *P. oceanica* tissues at site B was higher than that at site A ($p < 0.001$ and $p = 0.06$, respectively). The fluoranthene and benz[a]anthracene concentrations in seawater were significantly different among the four sites ($p = 0.011$ and $p = 0.035$, respectively).

This study examined the potential use of *P. oceanica* as a bioindicator for spatiotemporal monitoring of PAH pollutants in the Mediterranean Sea. PAH concentrations in bulk *P. oceanica* tissues and sediments showed a similar temporal trend. The leaves of *P. oceanica* had the highest PAH concentrations (when compared to the sheaths, roots and rhizomes) likely reflecting the fact that PAH contaminants are adsorbed directly from the ambient water, rather than from the sediment pore water. We thus conclude that PAH levels in seagrass leaves best represent the pollution status of seawater.

The different passive sampling approaches (in situ vs. in vitro sampling) revealed spatial differences in the absorption of HMW PAHs when compared to sediments, *P. oceanica*, and seawater samples. Although passive sampling cannot replace direct analysis of PAHs in seagrasses, this approach provides a useful tool for investigating sources of PAHs in *P. oceanica* meadows by enabling the assessment of the freely dissolved pollutants to which these plants are exposed.

Acknowledgments

We would like to express our gratitude to Gijs Coulier, Eveline De Meulemeester, Marijke Neyts, Daniël Saudemont, Marc Knockaert, and Edwige Devreker, technical personnel at MUMM institute in Ostend, for assistance during laboratory work. The authors acknowledge also the Management Body of Evros Estuary for helping us to collect samples from Evros River and the Greek Ministry of Education and Religious Affairs for offering study leave and financial support to Maria-Venetia Apostolopoulou.

Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.marpolbul.2014.07.051.

References

Ancora, S., Bianchi, N., Butini, A., Buia, M.C., Gambi, C., Leonzio, C., 2004. *Posidonia oceanica* as a biomonitor of trace elements in the Gulf of Naples: Temporal trends by lepidochronology. Environ. Toxicol. Chem. 23, 1093–1099.

Athanasoulis, G.A., Skarsoulis, E.K., 1992. Wind and Wave Atlas of North-East Mediterranean Sea. Laboratory of Nautical and Marine Hydrodynamics, NTUA, Athens.

Balestri, E., Benedetti-Cecchi, L., Lardicci, C., 2004. Variability in patterns of growth and morphology of *Posidonia oceanica* exposed to urban and industrial wastes: contrasts with two reference locations. J. Exp. Mar. Biol. Ecol. 308, 1–21.

Bates, K., Young, W., Sutton, A., 1997. Proposed Environmental Quality Standards for Naphthalene in Water. Environment Agency R&D Technical Report, P. 54.

Booij, K., Smedes, F., van Weerlee, E.M., 2002. Spiking of performance reference compounds in low density polyethylene and silicone passive water samplers. Chemosphere 46, 1157–1161.

Catsiki, V.A., Panayotidis, P., 1993. Copper, chromium, and nickel in tissues of the Mediterranean seagrasses *Posidonia oceanica* and *Cymodocea nodosa* from Greek coastal areas. Chemosphere 26, 963–978.

Cerniglia, C.E., 1993. Biodegradation of polycyclic aromatic hydrocarbons. Curr. Opin. Biotechnol. 4, 331–338.

Clark, K.E., Gobas, F.A.P.C., Mackay, D., 1990. Model of chemical uptake and clearance by fish from food and water. Environ. Sci. Technol. 24, 1203–1213.

Delgado, O., Ruiz, J., Perez, M., Romero, J., Ballesteros, E., 1999. Effects of fish farming on seagrass (*Posidonia oceanica*) in a Mediterranean bay: seagrass decline after organic loading cessation. Oceanol. Acta 22, 109–117.

EPA, 2009. List of Contaminants and their Maximum Contaminant Level (EPA 816-F-09-0004). United States Environmental Protection Agency, 6 pp.

European Maritime Safety Agency (EMSA), 2010. Maritime Accident Review 2010. <http://www.emsa.europa.eu>.

Ferrat, L., Bingert, A., Romeo, M., Gnassia-Barelli, M., Pergent-Martini, C., 2002. Mercury uptake and enzymatic response to *Posidonia oceanica* after an experimental exposure to organic and inorganic forms. Environ. Toxicol. Chem. 21, 2365–2371.

Gogou, A., Bouloubassi, I., Stephanou, G., 2000. Marine organic geochemistry of the Eastern Mediterranean: 1. Aliphatic and polyaromatic hydrocarbons in Cretan Sea surficial sediments. Mar. Chem. 68, 265–282.

Gosselin, M., Bouquegneau, J.M., Lefebuie, F., Gilles, L., Pergent, G., Pergent-Martini, C., Sylvie, G., 2006. Trace metal concentrations in *Posidonia oceanica* of North Corsica (northwestern Mediterranean Sea): use as a biological monitor? BMC Ecol. 6, 1–19.

Hatzianestis, I., Sklivagou, E., 2001. Distribution and origin of hydrocarbons and chlorinated compounds in sediments from Strymonikos gulf in northern Aegean Sea. Greece. Rapp. Comm. Int. Mer. Medit. 36, 134.

Hellenic Statistical Authority (El.Stat.), 2013. Laid-up Greek and Foreign Merchant Ships of 100 GRT and Over in Greek Ports. <http://www.statistics.gr>.

Hemminga, M.A., Duarte, C.M., 2000. Seagrass Ecology. Cambridge University Press, Cambridge.

Irwin, R.J., VanMouwerik, M., Stevens, L., Seese, M.D., Basham, W., 1997. Environmental Contaminants Encyclopedia. National Park Service, Water Resources Division, Fort Collins, Colorado. <http://www.nature.nps.gov/water/ecencyclopedia/index.cfm>.

Kanellopoulos, T.D., Hatzianestis, I., Angelidis, M.O., Kapsimalis, V., 2007. Aliphatic and polycyclic aromatic hydrocarbons in recent prodelta sediments of the Evros River, NE Aegean Sea. Freshw. Environ. Bull. 16, 776–783.

Kapsimalis, V., Panagiotopoulos, I., Kanellopoulos, T.D., Hatzianestis, I., Antoniou, P., Anagnostou, C., 2010. A multi-criteria approach for the dumping of dredged material in the Thermaikos Gulf, Northern Greece. J. Environ. Manag. 91, 2455–2465.

Karditsa, A., Poulos, S., 2007. Bathymetric changes of the bottom relief in the gulf of Alexandroupolis (NE Aegean Sea) between 1960–2004, in relation to its harbor expansion. Rapport du Commision Internationale Mer Mediterranee CIESM 38, 99.

Lacerda, L.D., Resende, C.E., 1986. Effect of metals on the seagrass *Halodule wrightii* Aschers during one growing season. Rev. Bras. Bot. 9, 87–90.

Lewis, M.A., Devereux, R., 2009. Review non-nutrient anthropogenic chemicals in seagrass ecosystems: fate and effects. Environ. Toxicol. Chem. 2, 644–661.

Malea, P., 1993. Bioaccumulation of aluminum in the seagrasses *Cymodocea nodosa* (Ucria) Aschers and *Posidonia oceanica* (L.) Delile and in macroalgae of the Gulf of Antikyra (Greece). Bot. Ma. 36, 423–431.

Malea, P., Haritonidis, S., Kevrekidis, T., 1994. Seasonal and local variations of metal concentrations in the seagrass *Posidonia oceanica* (L.) Delile in the Antikyra Gulf. Greece. Sci. Total. Environ. 153, 225–235.

Maserti, B.E., Ferrara, R., Paterno, P., 1988. *Posidonia* as an indicator of mercury contamination. Mar. Pollut. Bull. 19, 381–382.

Miliou, A., Acasuso-Rivero, C., Shepperson, J., Lindholm, A., Caprio C., Demetriou, M., 2012. Evidence of high risk maritime traffic conditions in the Aegean Sea. In: Proceedings of 3rd International Conference on Industrial and Hazardous Waste Management, 12–14 September 2012, Chania, Crete, Greece.

Monteyne, E., Roose, P., Janssen, C.R., 2013. Application of a silicone rubber passive sampling technique for monitoring PAHs and PCBs at three Belgian coastal harbors. Chemosphere 91, 390–398.

Oral, N., Öztürk, B., 2006. The Turkish Straits, maritime safety, legal and environmental aspects. Turkish Marine Research Foundation, İstanbul. Publication no. 25. In: Abdulla, Ameer, Linden, Olof (Eds.), Maritime Traffic Effects on Biodiversity in the Mediterranean Sea: Review of Impacts, Priority Areas and Mitigation Measures. IUCN Centre for Mediterranean Cooperation, Malaga, Spain, 184 pp.

Panagiotopoulos, I., Kapsimalis, V., Kanellopoulos, T.H.D., Hatzianestis, I., 2009. Identification of inorganic pollutants in the sediments of the coastal area near to "Makedonia" airport in the Gulf of Thessaloniki (Thermaikos Gulf). In: Proceedings of the 9th Pan Hellenic Symposium of Oceanography and Fisheries 13–15/5/09, I, pp. 217–222.

Papadopoulou, D., Samara, C., 2002. Polycyclic aromatic hydrocarbon contamination and LUMIStox solvent extract toxicity of marine sediments in the North Aegean Sea. Greece. Environ. Toxicol. 17, 556–566.

Pehlivanoglou, K., 1989. Evros Delta, evolution of continental self sediments. Mar. Geol. 87, 27–29.

Pavlidou, A., Hatzianestis, I., Sklivagou, E., Papadopoulos, V., Zervakis, V., 2002. Hydrology and pollution assessment in a coastal estuarine system: The case of Strymonikos Gulf (North Aegean Sea). Mar. Mediterr. Sci. 3 (1), 65–78.

Pergent, G., Céline Labbe, C., Lafabrie, C., Kantin, R., Pergent-Martini, C., 2011. Organic and inorganic human-induced contamination of *Posidonia oceanica* meadows. Ecol. Eng. 37, 999–1002.

Pergent-Martini, C., 1998. *Posidonia oceanica*: A biological indicator of past and present mercury contamination in the Mediterranean Sea. Mar. Environ. Res. 45, 101–111.

Ren, L., Huang, X.D., McConkey, B.J., Dixon, D.G., Greenberg, B.M., 1994. Photoinduced toxicity of three polycyclic aromatic hydrocarbons (fluoranthene, pyrene, and naphthalene) to the duckweed *Lemna gibba* L. G-3. Ecotoxicol. Environ. Saf. 28, 160–171.

Sikkema, J., de Bon, J.A., Poolman, B., 1995. Mechanisms of membrane toxicity of hydrocarbons. Microbiol. Mol. Biol. Rev. 59, 201–222.

Smedes, F., 2007. Monitoring of chlorinated biphenyls and polycyclic aromatic hydrocarbons by passive sampling in concert with deployed mussels. In: Greenwood, R., Mills, G., Vrana, B. (Eds.), Comprehensive Analytical Chemistry, vol. 48. Passive Sampling Techniques in Environmental Monitoring, pp. 407–448.

Smedes, F., 2010. Passive sampling in biomonitoring. Deltares Report, 1202337-004.

Smedes, F., Bakker, D., de Weert, J., 2010. The Use of Passive Sampling in WFD Monitoring. The Possibilities of Silicon Rubber as a Passive Sampler. Deltares Report 1202337-004-BGS-0027.

US Energy Information Administration (EIA), 2012. Worlds Transit Chokepoints. Report, August 2012.

Viso, A.C., Pesando, D., Bernard, P., Marty, J.C., 1993. Lipid components in the Mediterranean seagrass *Posidonia oceanica*. Phytochemistry 34, 381–387.

Vrana, B., Mills, G.A., Allan, I.J., Dominiak, E., Svensson, K., Knutsson, J., Morrison, G., Greenwood, R., 2005. Passive sampling techniques for monitoring pollutants in water. Trends Anal. Chem. 24, 845–868.

Ward, T.W., Correll, R.L., Anderson, R.B., 1986. Distribution of cadmium, lead and zinc among the marine sediments, sea grasses and fauna and the selection of sentinel accumulators near a lead smelter in South Australia. Aust. J. Mar. Freshw. Res. 37, 567–586.

Warnau, M., Ledent, G., Temara, A., Bouquegneau, J.M., Jangoux, M., Dubois, P., 1995. Heavy metals in *Posidonia oceanica* and *Paracentrotus lividus* from seagrass beds of the northwestern Mediterranean. Sci. Total Environ. 171, 95–99.

Xeidakis, G.S., Delimani, Z.P., Skias, Z.S., 2007. Erosion problems in Alexandroupolis coastline, Northeastern Greece. Environ. Geol. l53, 835–848.

Yates, K., Pollard, P., Davies, I.M., Webster, L., Colin, F., Moffat, C.F., 2011. Application of silicone rubber passive samplers to investigate the bioaccumulation of PAHs by *Nereis virens* from marine sediments. Environ. Pollut. 159, 3351–3356.

Zadeh, C.M., Saify, A., Shalikar, H., 2010. Polycyclic aromatic hydrocarbons (PAHs) along the eastern Caspian Sea coast. Global Environ. Res. 4, 59–63.