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## A multidisciplinary approach to evaluate the environmental impact of offshore gas platforms in the western Adriatic Sea

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## A multidisciplinary approach to evaluate the environmental impact of offshore gas platforms in the western Adriatic Sea

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Detecting the anthropogenic impacts of offshore gas platforms requires reliable tools, because the traditional evaluation based only on chemical analyses is neither appropriate nor sufficiently sensitive. Thus, a 3-year monitoring project was carried out to evaluate the impact of a platform based on a chemical–biological approach. Benthic communities are investigated as they are widely used to monitor the effects of marine impacts because the organisms are mostly sessile and integrate the effects of pollutants over time. Changes in benthic infauna, sediments and water quality, as well as biota bioaccumulation, caused by drilling and platform operations were evaluated experimentally. Furthermore, mussels (*Mytilus galloprovincialis*) were collected seasonally from the platform legs, both close to and far from the sacrificial anodes and at a control site. Responses of biomarkers of exposure and effect were related to the average levels of polycyclic and aliphatic hydrocarbons, organic matter content and heavy metals in bivalve tissues. Our data suggested that a slight perturbation can be detected only by integrating the results of chemical analyses on water and sediments and those obtained from benthic biological surveys.

Keywords: gas platforms; benthic community; sediments; biomarkers; body burden accumulation

#### 1. Introduction

The Adriatic Sea is a semi-enclosed sea subjected to intensive anthropogenic pressure characterised by heavy maritime traffic, intense fishing pressure and increasing inputs of environmental contaminants. The main sources of pollutants are related to industrial and agricultural land-based activities. Nevertheless, the rising contribution from offshore gas exploitation and production activity has become of great concern due to the potential ecological risks posed by the chemicals released during extraction tasks and platform maintenance. Since the 1960s, more than 110 offshore gas platforms have been constructed in the central and northern Adriatic Sea [1] where there is currently the highest concentration of fossil fuel extraction platforms in the Mediterranean area.

Several environmental issues are associated with the exploitation of offshore reservoirs. Both oil and gas extracted from the seabed contain some water (production water; PW), which must

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be eliminated before transporting the gas to the refinery. The water is generally extracted by physical separators and then discharged into the sea. PW may contain varying concentrations of production chemicals (e.g. complexing agents) or biocides, as well as natural components such as polyaromatic hydrocarbons (PAHs), alkylphenols and metals [2]. Although PW is rapidly diluted in the surrounding seawater and pollutants are present at low concentrations, the total volume of the final effluent is remarkable. Thus marine organisms are exposed to low but chronic concentrations of PW components. Moreover, additional environmental issues have to be evaluated when assessing adverse ecological impacts from oil and gas extraction activities. For example, muds which are used as coolant and lubricant materials during drilling activity can be wateror oil based according to the nature of the geological strata. Although there are some attempts to separate the oil-based muds from the cuttings before dumping them onto the seabed, they remain heavily contaminated. This represents one of the greatest potential hazards to the marine environment. In the worst affected areas, fauna can experience a loss of diversity and severe changes in benthic composition [3-5]. Furthermore, these platforms are often situated under a wide range of environmental conditions from shallow to deep waters, with different types of sediments (from mud to sand). In addition, in the Adriatic Sea, they may be under the influence of the Po River, it being the main source of fresh water into the northern Adriatic Sea [6]. Hence, the large variability in natural conditions affects sediment and seawater characteristics differently, making it difficult to provide a reliable ecological risk assessment.

To overcome this problem, a multidisciplinary, chemical-biological approach, focused on changes in benthic communities and sediment chemical accumulation was developed to assess the environmental impact of an offshore gas exploitation and production activity in the Adriatic Sea. Furthermore, organic and inorganic pollutant body burden accumulation, as well as biological markers of stress, were quantified in sentinel animals, *Mytilus galloprovincialis*, as early warning index of water column pollution [7]. Measurement of the biological effects of pollutants has become crucial for a correct quality assessment of the marine environment. The responses at a cellular level can provide information about long-term effects at higher biological levels (i.e. population and ecosystem) which have much higher economic and ecological relevance [8]. As recognised in recent years by international organisations and environmental agencies, the environmental risk assessment cannot be based solely on chemical analyses of environmental samples because this approach, *per se*, does not provide indications of actual pollutant bioavailability and hence of their deleterious effects on aquatic organisms. This study work explores the results of a medium-term investigation of the impact of a four-leg platform located in the Adriatic Sea.

#### 2. Materials and methods

#### 2.1. Study area and sampling strategy

The platform is located at  $\sim$ 37 km off Ancona (north-western Adriatic Sea) on a muddy seabed at 75 m depth (Figure 1). The main current at the sea bottom flows from the north-west towards the south-east, following the basin topography and has a speed of 5–10 cm·s<sup>-1</sup>, sporadically reaching 20 cm·s<sup>-1</sup>.

The platform was installed in May 2002 and the drilling operations started in the following month and finished in October 2002. Production began immediately after. The first sampling activity involving only sediment analyses was performed 1 month (November 2002) after the end of drilling operations (hereafter ADO), whereas the following six field cruises were carried out from 2003 to 2005, i.e. 5, 11, 18, 24, 30 and 36 ADO, and included the macrozoobenthic investigation. The sampling design was planned according to the 'gradient design' approach, which is particularly useful when a stressor or disturbance attenuates with the distance from the



Figure 1. Study area and position of the investigated gas platform. The sampling scheme of both sediments and the benthic community is also reported.

point source of impact. This design is sensitive to change for point source data, enabling the scale of the effects of a disturbance to be readily identified [9].

Four sampling sites ( $\sim 20$  m apart) were randomly selected at 5, 30, 60, 120 and 1000 m from the platform (Figure 1). Three samples were collected using a Van-Veen grab (capacity, 13 L; surface, 0.105 m<sup>2</sup>) for benthic community investigation. Superficial sediments (0–2 cm) for physical-chemical analyses were collected by box-corer.

#### 2.2. Data analyses

#### 2.2.1. Macrofauna

The overall biological samples were sieved *in situ* through a 0.5-mm mesh and preserved in 5% buffered formalin. Macrofauna was sorted under magnification, classified to the species level when possible and counted. To analyse spatial and temporal changes in species composition, we used the permutational multivariate analysis of variance (PERMANOVA). The model was a three-way design with distance (five levels) and period (ADO; six levels) as fixed orthogonal factors, and sites (four levels) as random factor nested in distance/period. In each sampling period, pairwise comparisons among distances were made. To perform the PERMANOVA, the PRIMER<sup>TM</sup>

ecological software package developed by the Plymouth Marine Laboratory was used [10]. Each term in the model was put through permutation tests based on 9999 permutations of residuals under a reduced model to determine *p*-values. Analyses were made on Gower exc 0-0 similarity matrix. The Gower coefficient is well-suited for quantitative abundance data excluding double-zeros from comparison [11,12]. Before calculation of the matrix, species abundance data were fourth-root transformed to reduce the contribution of prevalent taxa. Afterwards, species contributing at least 10% of the total community abundances were selected.

#### 2.2.2. Sediments

Textural properties of sediments were analysed investigated. In addition the concentration of aluminium (Al), cadmium (Cd), chromium (Cr), barium (Ba), cuprum (Cu), mercury (Hg), nickel (Ni), lead (Pb), zinc (Zn) and PAH were measured.

Sediment samples were analysed for their particle size according to the Udden–Wentworth Phi classification [13]. Each sample was washed in 16% hydrogen peroxide for 24 h and then wetsieved on a 63-µm mesh to sort out the fine fraction. The sand fraction was sieved through a stack of geological test-sieves ranging from 0 Phi to +4 Phi. The fine fraction was analysed by sedigraph. Trace elements analysis was carried out digesting 0.5 g of fresh sediment in Teflon vessels with a mixture of concentrated HNO<sub>3</sub>, HCl and HF (suprapur). The digestion was performed using a microwave digester. The analyses were performed by graphite furnace absorption spectrometry (Varian SpectrAA 240Z) for Cd and Pb analysis, by atomic emission spectrometry (ICP AES) for Cu, Cr, Al, Zn and Ni quantification, and by cold vapour atomic absorption spectrometry after reduction by stannous chloride technique for Hg estimation. The accuracy of the analytical methods was checked by evaluating the recoveries of processed PACS-2 (harbour sediments) and MESS-3 (estuarine sediments, NRC, Canada) certified reference materials. Recoveries ranged from 95 to 103%.

For PAH analysis, 20 g of wet weight subsample was extracted using methanol/benzene (1:1 v/v). Two other extractions were sequentially carried out using 50 mL of benzene. The extracts were then pooled and dried.

The differences between sites were tested by the non-parametric Kruskal–Wallis test, the variance homogeneity assumption for the ANOVA test being violated. The level of significance was set at 0.05.

#### 2.2.3. Biomarkers and bioaccumulation levels

Native mussels (*M. galloprovincialis*) were collected seasonally, in summer and winter (18, 24, 30 and 36 months ADO), from two submerged sites of the platform, close to and far from the platform's anodes (CA and FA sites respectively), and at a reference site located at Portonovo Bay (Ancona, Italy). Biomarkers of physiological stress (stress on stress, condition index), cellular damage (latency of N-acetyl-β-hesosaminidase assay and Neutral Red retention time), DNA damage, such as micronuclei frequency (MN), oxidative stress (catalase) and exposure, such as metallothionein (MT) and mixed function oxigenase (MFO), were applied to evaluate the potential toxic effects of aliphatic hydrocarbons (AHs), PAHs and trace elements [7,8,14–16].

Heavy metal concentrations in whole body (4 g, wet wt) were detected by graphite furnace absorption spectrometry (Cd and Pb), ICP AES (Cu, Cr, Al, Zn and Ni) and cold vapour after reduction by NaBH<sub>4</sub> (Hg) according to EPA 3050B method. Quality assurance and quality control were assessed by processing blank samples and reference standard material (Mussel Tissue Standard Reference Material SRM 2977, NIST). Concentrations obtained for standard reference

materials were always within the 95% confidence interval of certified values aliphatic hydrocarbons levels ( $C_5-C_{40}$ ) and were analysed according to Carro et al. [17]. PAHs were extracted with *n*-pentane and dried over sodium sulfate. PAHs were identified by comparing their retention times with those from the aromatic analytical standards by Supelco 48743, according to the priority PAHs from method EPA 610. The extracts were then analysed using Hewlett-Packard 1090A liquid chromatography equipped with a reverse-phase column (200 × 2.1 mm ODS Hypersil 5 mm), and a Hewlett-Packard 1046A fluorescence detector. Analyte concentrations in the standard solutions were in the range 5–5000 ng· $\mu$ L<sup>-1</sup>.

### 3. Results and discussion

#### 3.1. Macrofauna

In total, 139 taxa were collected, including polychaete worms that made up the greatest proportion of the macroinvertebrate population (67 taxa: 49 at species level, 9 at genus level and 9 to family level), 32 crustaceans (24 at species level, 5 at genus level and 3 to family level), 26 molluscs (25 at species level and 1 at genus level), 6 echinoderms, 4 cnidarians, 2 sipunculids, 1 nemertean and 1 platelmint, for a total of 1481 in  $0.105 \text{ m}^{-2}$ . The benthic community was dominated by few soft-bottom species (*Marphysa bellii*, *Aphelochaeta marioni* and Cirratulidae).

Significant differences in the interaction distance  $\times$  period (ADO) were found (Table 1), indicating a potential effect of the platform on the spatial and temporal variation in zoobenthic communities. The consecutive pair-wise tests highlighted homogeneity from 5 to 120 m sites during all sampling periods. In the first survey (5 ADO) only, sites located from 5 to 120 m from the platform showed a significant difference with 1000 m sites. In fact, an initial defaunation in terms of both number of species and individuals was observed within 120 m radius, where some opportunistic species were found (e.g. *Caulleriella caputesocis, Minuspio cirrifera*). These species act as pioneers in defaunated bottoms and prepare the substrate for other organisms colonisation that have more elaborate settlement needs [18].

Defaunation was followed by recolonisation of the area in the second and third years (from 18 to 36 ADO), when the appearance/increment and/or a larger distribution of some common soft-bottom species (e.g. *Hyala vitrea*, *M. bellii*) was recorded at all distances. Moreover, starting from 24 months ADO, the settlement of a mussel mound within 30 m of the platform induced a

Source of variability	df SS		MS		F	p
Time (T)	5	59772.00	1195	4.00	4.9038	0.0001
Distance (Di)	4	16303.00	407	5.00	1.6719	0.0001
T × Di	20	54449.00	272	2.50	1.1168	0.0354
Site $(T \times Di)$	90	2.19E + 05	243	7.80	1.3630	0.0001
Pair-wise tests for term T						
5 ADO	11 ADO			18 ADO		
$5 \text{ m} = 30 \text{ m} = 60 \text{ m} = 120 \text{ m} \neq 1000 \text{ m}$	5  m = 30  m = 60  m = 120  m = 1000  m			5  m = 30  m = 60  m = 120  m = 1000  m		
24 ADO 5 m = 30 my = 60 m = 120 m = 1000 m	30 ADO 5  m = 30  m = 60  m = 120  m $5 \text{ m}, 30 \text{ m} \neq 1000 \text{ m}$ 60  m, 120  m = 1000  my			36 ADO 5 m = 30 m = 60 m = 120 m 5 m, 30 m $\neq$ 1000 m 60 m, 120 m = 1000 m		

Table 1. PERMANOVA analyzing differences among assemblages at increasing distances from the platform during the six sampling periods. Time: 6 fixed levels; Distance: 5 fixed levels; Site: 20 random levels.

Note: SS, sum of squares; MS, mean of squares; ADO, time in months after the end of the drilling operations. Not significant p > 0.05; significant  $0.05 \ge p > 0.01$ ; highly significant  $p \le 0.01$ .

proliferation of some hard-bottom species (e.g. *Ostrea edulis, Hydroides pseudouncinata, Pomatoceros triqueter*), with a consequent increase in community diversity. This enrichment caused a significant difference between 5–30 m sites and 1000 m sites in the third year.

Hence, the presence of the platform seems to induce diversification and enrichment of the natural benthic community [19–22] as a consequence of increased seabed heterogeneity due to shells, fragments and dead organisms falling from the platform due to natural dislodgement or platform cleaning. In fact, specimens of *M. galloprovincialis* that have fallen from the submerged parts of the platform provide natural hard substrates that create a new habitat for marine epifaunal organisms that do not or only rarely occur in soft-bottoms. The settlement of hard-bottom species, otherwise unable to flourish, determined an increased complexity in the original community if compared with typical soft-bottom infauna in the surrounding area. These effects, however, are spatially confined and time limited, as pointeded out in previous studies regarding benthic communities [23–25].

#### 3.2. Sediments

Most of the sediments showed no black anoxic trails along the sedimentary profile. The oxidised superficial stratum varied between 1.5 and 2 cm. Signs of bioturbation, commonly observed, were mainly due to polychaete activity. Similar features were observed in sediments collected at sites located 1000 m from the platform. Bentonite clays and black anoxic trails were observed during the first two cruises (1 and 5 ADO) in samples collected under the platform and at ites located at 30 and 60 m from it. Grain size analysis revealed that most of the sediments consisted of mud (silt + clay) with a low percentage of sand. At all sites, the percentage of sand was slightly higher during the cold periods (5, 18, 30 months ADO) than during the warm periods (11, 24, 30 months ADO). However, the opposite trend was observed for the fine fractions. These observations were consistent over time; however the differences were never statistically significant. Conversely, the percentage of clay increased significantly only during the two cruises carried out after 5 and 11 months ADO.

PAH total content determined in the sediments varied throughout the study period between 0.09 and  $3.19 \text{ mg} \cdot \text{kg}^{-1}$ . Despite the large variability in these concentrations (Figure 2), most values were in good agreement with those previously reported in the central Adriatic Sea [1].

The highest value was observed 1 month ADO, the total sum of PAH being  $10\times$  that reported by Maggi et al. [1] in the same area. However, average levels decreased gradually during the following 3 years of monitoring activity because a homogeneous distribution was observed in the last survey. A single abnormality was detected at 30 m from the platform (1.34 mg·kg<sup>-1</sup>) 24 months ADO. Similar to PAH, most heavy metals showed some anomalies during the first survey after platform installation, but their concentrations followed a decreasing pattern up to the end of the study when no evidence of impact was observed.

By contrast, Ni and Cr increased slightly according to a common pattern among sites near the platform and those located 1000 m from it. Hence, a possible effect related to the platform could be excluded, even if some signs of disturbance were assessed for Ba and Zn (Figure 2) throughout the study.

In particular, the Ba concentration close to the platform  $(1370.2 \text{ mg} \cdot \text{kg}^{-1})$  and at 30-60 m away was  $\sim 3-5 \times$  the average concentration observed at sites 1000 m from the platform  $(300.7 \pm 43.8 \text{ mg} \cdot \text{kg}^{-1})$ . Zn showed significantly higher concentrations near the platform where values  $(174.8 \text{ mg} \cdot \text{kg}^{-1})$  were 1.6-fold higher than the average observed at the sites farthest from the platform  $(107.3 \pm 16.8 \text{ mg} \cdot \text{kg}^{-1})$ .

Likewise, the clearest signs of impact on sediment properties were related to the installation phase and to drilling operations because the highest concentrations of pollutants on sediments



Figure 2. Conditional box plots of polyaromatic hydrocarbon (PAH), Ba and Zn concentrations  $(mg \cdot kg^{-1})$ . F, farthest sites (1000 m from the platform); O, other sites; M ADO, months after drilling operations.

were detected 1 month after these activities ceased, corroborating previous studied carried out in the Adriatic Sea [26,27]. From the first survey onward, they decreased and only occasional signs of disturbance persisted near the platform. Similar to the results of other surveys carried on in the Adriatic Sea, only Ba and Zn showed significant increases, baryte-enriched muds (BEM) [28,29] being the most probable source of both metals.

Baryte, a soft dense natural mineral, is commonly used as a weighting agent in drilling muds in all types of oil and gas exploration [29]. Several metals are present in most BEM, but As, Ba, Cr, Cd, Cu, Fe, Pb, Hg, Ni and Zn are of greatest concern for their potential toxicity and/or abundance in drilling fluids [30,31]. In addition, the potential biological effects of discharged muds and cuttings near an offshore platform require great attention, especially if related to the bioavailability of BEM ingredients. Environmental bioavailability depends on interactions between an organism and its environment. It is controlled by the relative amount of permeable epithelia in contact with the different environmental media, by the duration of the contact, and by the physical form of the chemicals. Dissolved, free ionic forms, some metal-organic colloid complexes and low molecular mass organo-metal compounds are the most bioavailable forms of most metals [32]. The most bioavailable fraction of metals in both BEM and cuttingassociated sediments is that dissolved in the pore water or loosely complexed with particles. Deuel and Holliday [33] found that almost all investigated metals (with one exception) were in organic/sulfide complexes or in the residual fraction, both considered inaccessible for bioaccumulation by marine organisms. Nearly 50% of the Pb and more than 20% of the Cd, Cr and Zn in the BEM tested samples were in the Fe–Mn oxide fraction, which dissolves under anoxic conditions in sediments, releasing adsorbed metals. However, the solubility of Ba in seawater and marine sediments is controlled by the concentration of reactive sulfate, which is high in seawater [34]. In the presence of normal concentrations of inorganic sulfate, Burton et al. [34] estimated a barite seawater solubility of  $81 \,\mu g \cdot L^{-1}$ , with  $48 \,\mu g \cdot L^{-1}$  as Ba at 20 °C. Sulfatereducing bacteria in suboxic layers of the cutting pile use dissolved sulfate as an electron acceptor for organic matter biodegradation and, in the process, convert sulfate to sulfide. As the sulfate concentration in cuttings pile pore water decreases, barite becomes more soluble, releasing small amounts of Ba into solution. This reaction is probably self-limiting because dissolution of barite releases sulfate into solution. However, if the barite concentration in sediments is high, it can serve as a source of reducible sulfate for sulfate-reducing bacteria [35], releasing dissolved Ba into the pore water [36]. Furthermore, solid metals and metal salts associated with barite, clay and cutting particles are not readily bioaccumulated by animals living in close association with the cuttings pile; the metals are not passed efficiently through marine food chains [37]. When accumulated, such heavy metals are often not assimilated into the tissues, but remain as insoluble, inert concretions [37]. Many authors evaluated the bioavailability to several species of bottomliving marine animals of several metals in different purities of drilling mud barite in sediments [38], and it is probable that some of the metals apparently bioaccumulated by the marine animals were actually still associated with fine particulate barite or other sediment particles in both the gut and the gills. Marine invertebrates can take up fine particles into epithelial cells by pynocytosis. The metals associated with the particles remain linked to them and are only partially assimilated by the organisms, with low or absent effects on benthic community distribution and association.

Field studies confirm that metals present in drilling muds and cuttings are bioaccumulated scarcely if at all by benthic animals. Only a small increase in Ba concentrations in tissues as been demonstrated, whereas the bioaccumulation of other metals was rarely observed [3,39]. Cripps et al. [26] reported that mussels (*M. edulis*) collected from the surface of cutting piles in the Norwegian Sector of the North Sea contained higher concentrations of several metals in their soft tissues than controls. Possible sources of these metals were the metal platform where the mussels lived, the PW or other waste discharges, and the cutting piles.

#### 3.3. Biomarkers responses and body burden accumulation

Among all biological responses, MN ranged from 2.7 to 4.8% in the haemolymph of mussels sampled near the platform. Reference mussels scored significantly lower frequencies (2.0-2.8%). underlying the effects of a slight, transient increase in genetic stress (Figure 3). However, sampled bivalves evidenced an increment of heavy metals stress-related syndrome as shown by the significant increase in cytosolic levels of MTs. Among all sampling campaigns, levels ranged from 89 to  $325 \,\mu g \,\mathrm{MT} \cdot g \,\mathrm{tissue}^{-1}$  in bivalves collected near the platform, while reference organisms presented significantly lower MT inductions (18-105 µg·g tissue<sup>-1</sup>; Figure 3). Such biological responses were related to the significant bioaccumulation levels of two nonessential, suspected MT inducers, trace elements like Cd and Ni and the essential trace element Zn [27]. Cd ranged from 0.6 to 0.8 mg  $kg^{-1}$  dry weight in reference bivalves (Figure 4), while mussels collected at platform sites exhibited average higher accumulation levels  $(0.7-1.2 \text{ mg} \cdot \text{kg}^{-1} \text{ dry weight})$ . Likewise, Ni presented average low bioaccumulation levels in tissues of mussels collected within the reference site  $(1.9-2.5 \text{ mg} \cdot \text{kg}^{-1} \text{ dry wt.}, \text{ Fig. 4})$ , while filter feeders collected both at CA and FA sites showed significantly higher concentrations (2.0-4.2 mg·kg<sup>-1</sup> dry wt.). Zn levels were within the range  $57.0-153.0 \text{ mg} \cdot \text{kg}^{-1}$  dry wt. in the control mussels, whereas organisms collected near the offshore platform revealed significantly higher rates of accumulation  $(121-189 \text{ mg} \cdot \text{kg}^{-1})$ dry wt., Figure 4). These results agree with other studies involving mussels as sentinel species



Figure 3. Biomarkers recorded in mussels collected near the *Calipso* platform. Micronuclei frequencies (MN) and metallothionein (MT) content. C, control site; CA, close to platform anode; FA, far from platform anode. Data are expressed as mean, standard error (box) and standard deviation (outer line).  $\cdot$  denotes significant differences from the control site (G-test, Mann–Whitney, p < 0.05).



Figure 4. Conditional box plots on Cd, Ni, Zn, polycyclic aromatic hydrocarbon (PAH) and aliphatic hydrocarbons (AH). C, control sites; CA, close to platform anode, FA, far from platform anode. Data are expressed as mean, standard error (box) and standard deviation (outer line).  $\cdot$  denotes significant differences from the control site (Mann–Whitney, p < 0.05).

for biomonitoring activities along coastal and marine sites in the Adriatic Sea [27,40–43]. The pattern of metal distribution, characterised by the constant increase in some metals (Zn and Cd), suggests anode corrosion as a possible source of bioavailable trace elements. Besides, PAHs presented low concentrations in all sites (Figure 4), often below detection limits (LOD), except for benzo(*a*)anthracene (from <LOD to  $38.8 \text{ ng} \cdot \text{g}^{-1}$  dry wt., data not presented), the most aromatic compound, the accumulation of which may be related to motor boat traffic [27]. By contrast, the total AH content in bivalves collected at the platform ranged from 1824 to 9126 ng  $\cdot \text{g}^{-1}$  dry wt. (Figure 4) mostly within normal levels for coastal areas subjected to moderate pollution like the Galician coast, the coast of the Canary Islands, the Gulf of Naples and the north-west coast of Portugal [17,19,43,44].

In contrast with the results of sediments chemical analysis, mussels sampled during our survey showed significant increases in other heavy metals like Cd, Ni and Zn. This supports the hypothesis of a distinct source of pollution for water column biota. Similarly, concentrations of Ba and Ch, but not other metals, were slightly elevated in sediments near exploratory drilling operations on Georges Bank after drilling [37]. However, metals concentrations in sea clams, *Artica islandica*, collected from surface sediments near the drilling platforms were in the normal range for bivalve

molluscs [45]. However, the significant induction of cytosolic free MTs in mussels collected near the platform provides early warning signals of stress correlated to a bioavailable heavy metals fraction in the seawater near the platform. Both Cd and Ni are considered strong inducers of MT synthesis [46]. Nevertheless, if considering the results of similar biomonitoring programmes carried out in coastal areas [47], such MT inductions appear moderate but continuous over time, pointing out, in particular, the technical function of the galvanic anodes as a possible source of metals in the aquatic environment [27,42]. The distribution of Pb, Cu and Cr in sediments indicated a probable origin from drilling mud and cuttings discharges. Zn and Al in sediments were primarily from sacrificial anodes on platform platforms. Bivalves from platform legs and nearby sediments contained higher concentrations of Pb, Cu and Zn than bivalves distant from the platform, suggesting metal bioaccumulation from platform sources. Thus, laboratory and field studies are consistent in showing that there is a restricted bioavailability of metals from sediments to marine animals. However, biological responses in sentinel organisms showed a transient increase in genetic stress, which was correlated to the temporal trend of organic pollutants. PAH showed low levels around the platform. Petroleum hydrocarbon levels were mostly within normal levels reported for mildly polluted zones such as the North Sea [48], Nova Scotia [49] and the north-west coast of Portugal [50]. All have not been regularly monitored in the Adriatic Sea, and it is not possible to establish baseline levels and temporal trends for these contaminants. Nevertheless, the levels of aliphatic compounds quantified in *M. galloprovincialis* tissues during this study were lower than those reported for coastal sites along the Mediterranean Sea and Atlantic Ocean [17,43,44,49,50].

#### 4. Conclusions

The results of this survey regarding an offshore production platform showed the importance of a multidisciplinary approach including both chemical and biological analyses. Platform-related inputs did not cause serious environmental impacts, even though temporary alterations were detected in both benthic composition and sediment pollutant distribution, as well as in biological responses and pollutant-accumulation profiles of sentinel organisms. Interestingly, the outcomes indicated that after the first impact, caused by the installation and drilling operations, the offshore platform can play an important ecological role in areas characterised by flat soft-bottoms and a relatively small amount of natural reef habitat, as seen in the Adriatic Sea.

As a whole, the results indicated that in the north–central Adriatic Sea a 3-year period can be considered a sufficient time lag to restore environmental conditions similar to those observed before the drilling activities. Finally, a 3-year monitoring programme appears to be effective to follow the time-dependent potential impact of such industrial structures.

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