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1	Effects of two offshore gas platforms on soft-bottom benthic communities (northwestern
2	Adriatic Sea, Italy)
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11	
12	Abstract
13	The macrozoobenthos living around two offshore gas platforms, Barbara NW (pB) and Calipso
14	(pC) located in the northwestern Adriatic Sea were investigated for three years after their
15	construction to detect eventual effects due to the platforms. The sampling stations were spaced at
16	increasing distance from the platforms up to 1000 m. Both multivariate and univariate analysis
17	showed an initial defaunation (short-term effect) at pB and within a 120 m radius at pC. A general
18	recovery in terms of abundance, species richness and diversity was observed for the benthic
19	communities after one year. During the third monitoring year a mussel mound developed at both the

platforms (longer-term effect) extending up to 30 m from pC, whereas similar soft-bottom communities were found at all the other distances. The geographical position play an important role in the "timing" of the above effects. Moreover, despite the different environmental features, a 3year monitoring plan can be effective to evaluate the potential impact on benthic communities of offshore gas platforms in the north-central Adriatic Sea.

25

- 1 Keywords: Offshore structures, Environmental impact, Zoobenthos, Community composition,
- 2 Adriatic Sea, Mussel mound.

1 1. Introduction

2 Over the last 50 years, non-renewable resources exploitation has strongly increased to meet the global growing demand for energy. The offshore oil and gas industry plays a leading role in the 3 4 marine field extraction activity exceeding 14 billion tonnes of oil equivalent (btoe; Ghisel, 1997; 5 Pinder, 2001). In the Mediterranean basin, the Italian production of natural gas was 11.4 million 6 tonnes of oil equivalent (mtoe) in 2004, rising to 0.2 bloe in 2005, the fourth-largest in the EU. 7 About 80% of the total energy comes from this type of activity (Brighenti et al., 2003), and almost 8 half of it is provided from offshore reserves in the Adriatic continental shelf (OGP, 2005). More 9 than 110 offshore gas platforms have been deployed since the 1960s in the northern and central 10 parts of this basin (Maggi et al., 2007), representing the highest concentration of fossil fuel 11 extraction platforms in the Mediterranean area.

12 It is well known that drilling operations generate waste fluids and cuttings that could cause 13 strong environmental impacts related to drilling mud discharges or hydrocarbons associated waters 14 (Terlizzi et al., 2008). Moreover, the physical structure of the platform may modify local water flow 15 and alter sediment type, affecting the benthic community composition (Wilson-Ormond et al., 16 2000). Generally, during the installation phase and the drilling operations, benthic communities 17 closer to the platforms show a decrease in diversity and a change in species' abundance, while 18 further away, assemblages may appear more similar to those in the surrounding unaffected area (Grant and Briggs, 2002). 19

The radius of impact can be different depending on the number of platforms placed in a restricted area, the structures' dimensions and biogeographic/climatic factors. For example, in the North Sea a few studies found effects up to 3,000 m, with more severe impacts within a 500 m radius (Gray et al., 1999; Kingston, 1987; Olsgard et al., 1997). In the Gulf of Mexico the long-term effects of oil-related activities on the benthos were evident up to 200 m from the source and up to 800 m from produced water discharge points (Hernández Arana et al., 2005; Montagna and Harper, 1996; Peterson et al., 1996; Rabalais et al., 1992).

1 The water-based drill-cuttings, commonly used in the Adriatic Sea, are less toxic than oil-2 based drilling muds (Cameron et al., 2004; Olsgard and Gray, 1995). Moreover, in most of cases 3 neither drill-cuttings nor formation water are discharged at sea during the drilling operations.

However, structures' installation, drilling operations as well as the associated great
occurrence of ships can cause an increase of metals, hydrocarbons and other chemical agents in the
environment causing potential impacts to the surrounding physical and biotic compartments.

Some studies highlighted the effects of platforms on benthic communities over the time
(Fabi et al., 2007; Frascari et al., 1991, 1992; Spagnolo et al., 2002, 2006; Trabucco et al., 2006). In
most of these cases, the more relevant ecological alteration due to the presence of a gas platform
was a mussel mound development affecting the surrounding seabed communities within a radius of
a few meters.

However, there is still much research to be undertaken to fully understand the gas platforms' potential impacts on the surrounding zoobenthos in the Adriatic Sea. Nevertheless, these platforms are installed in a wide variety of environments, with different types of sediments (from mud to sand) that may be under the influence of rivers such as the Po River representing the main source of fresh water into the northern Adriatic Sea (Cattaneo et al., 2003; Grilli et al., 2005; Marini et al., 2008). This situation, associated with the high drilling activity in the Adriatic basin, makes it difficult to develop general models for prediction of offshore platforms' impact.

19 The present work compares the possible effects on the surrounding zoobenthic communities 20 of two 4-leg gas platforms (Barbara NW and Calipso) in the northwestern Adriatic Sea installed at 21 similar depth but at different distances from the coastline and different types of seabed. It represents 22 an approach to detect possible common impact patterns both in temporal and spatial terms.

Data were collected in the framework of a wider monitoring programme aimed to evaluate the impacts of the platforms' installation on the different components of the marine environment (i.e. water column, sediments, benthic communities, fish assemblage, ecotoxicological response).

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4

1 **2. Materials and Methods**

2 2.1 Study area

3 The Adriatic Sea is a small continental shelf about 800 km long in NW-SE direction and 4 about 200 km wide. The northern part of the basin is very shallow, rarely exceeding 46 m depth, 5 while in the central part the depth can reach 80 m. The continental slope located around 600 km 6 from the northern limit separates the central Adriatic from the southern one where the depth reaches 7 1,200 m. The northern basin receives a high nutrient input from Po River and other minor streams 8 which enhances primary production near the surface and oxygen demand near the bottom (Grilli et 9 al., 2005; Marini et al., 2008). The Po watershed is very large and includes densely populated 10 regions (Crema et al., 1991).

11 The study was conducted in the northwestern Adriatic Sea. Two 4-leg offshore gas 12 platforms, Barbara NW (hereafter indicated as pB) and Calipso (hereafter indicated as pC), were 13 monitored for three years.

pB is located on a muddy sand seabed at a mean depth of 68.5 m, 56 km off Ancona (De 14 15 Biasi, unpublished results; Fig. 1). The submerged part of pB was installed in February 1999. The 16 drilling operations finished in June of the same year and the production started soon after. Six field 17 cruises were carried out from 2000 to 2002 (two per year). The main bottom current direction measured during the study period fluctuated between Northwest and Southeast and had a speed 18 sporadically reaching 10 cm s⁻¹ (Fabi et al., 2005). Grain size was homogeneous at the different 19 20 distances from the platform. Heavy metal content in the sediments did not show significant changes 21 at increasing distance from the platform, except for some peaks of cadmium, zinc, mercury, and 22 lead recorded in the surroundings of the structure during the first monitoring year after installation. 23 These anomalies gradually disappeared in the subsequent years (De Biasi et al., 2008; Fabi et al., 24 2005). Polycyclic Aromatic Hydrocarbon (PAH) levels increased in the second monitoring year 25 (2001) and strongly declined at the end of 2002. However, with only a few exceptions, the values of 26 the above contaminants usually agreed with those reported in other areas of the Adriatic Sea

(Guerzoni et al., 1984; Magi et al., 2002). Organic matter always showed the highest
 concentrations close to the platform and gradually decreased with increasing distances from it,
 indicating an intense biological activity around the structure (unpublished results).

- 4 pC is located on a muddy seabed approximately 35 km off Ancona in 75 m of water (De 5 Biasi et al., 2007; Fig. 1). The structure was installed in May 2002 and the drilling operations 6 started in the following month and finished in October 2002. Production began immediately 7 thereafter. Again, six field cruises were carried out from 2003 to 2005 (two per year). During the 8 study period the main current at the sea bottom flowed from Northwest towards Southeast, following the basin topography with a speed of 5-10 cm s⁻¹ sporadically reaching 20 cm s⁻¹ 9 10 (Paschini, unpublished results). The sediment texture was homogeneous at the different distances from the platform in the overall study period. A gradual decrease of heavy metal content was 11 observed from the first to the last monitoring year. Nevertheless, some anomalies for copper, lead, 12 13 barium and zinc were recorded during the overall monitoring period. During the first two years after installation PAH concentrations were higher around the structure compared with those recorded at 14 15 the farther distances, but they decreased in 2005 (De Biasi et al., 2006, 2007). Also in this case the 16 recorded contaminant levels were comparable with those reported in other Adriatic areas (Guerzoni 17 et al., 1984; Magi et al., 2002). Organic matter content was homogeneous in the overall area, 18 showing a consistent increase in the second monitoring year which persisted until the end of 2005 19 (De Biasi et al., 2007).
- At both platforms water-based muds were used for the drilling operations and neither cuttings nor formation waters were discharged at sea. Moreover, no water is discharged during the production phase.
- 23 2.2 Sampling design and analyses

The sampling design was planned according to the 'gradient design' approach that is particularly useful when a stressor or disturbance attenuates with the distance from the point source of impact (Ellis and Schneider, 1997).

1 For each platform, four sampling sites (about 20 m apart) were randomly selected at 5 m, 30 2 m, 60 m, 120 m, and 1000 m from the structure. At each site three samples were collected using a Van-Veen grab (capacity = 13 L and width = 0.105 m^2), for a total of 12 samples for each selected 3 4 distance.

5 The overall 720 biological samples (60 samples x 2 platforms x 6 surveys) were sieved in 6 situ through a 0.5 mm mesh and all organisms retained were preserved in 5% buffered formalin. In 7 the laboratory, macrofauna was sorted through a stereomicroscope and a binocular microscope, 8 identified and classified to the species level when possible using standard nomenclature, quantified, 9 and weighted.

10 2.3 Data analysis

Multivariate analyses were performed to identify spatial and temporal changes in the 11 12 composition and abundance of macrofauna communities using PRIMER[™] ecological software 13 package developed by the Plymouth Marine Laboratory (Clarke, 1993; Clarke and Warwick, 2001). 14 Prior to any analysis, the species abundance data were fourth-root transformed to reduce the 15 contribution of prevalent taxa and therefore increase the importance of less abundance species. 16 Afterwards, species contributing at least 10% of the total community abundances were selected and 17 the Gower exc 0-0 similarity matrix was calculated. Gower coefficient is well-suited for 18 quantitative abundance data excluding double-zeros from comparison (Legendre and Legendre, 19 1998). The differences among macrofauna assemblages at increasing distance from platforms 20 during the three monitoring years were tested by means of a permutation analysis of variance. This 21 method allowed testing of the general multivariate hypothesis of differences in the composition 22 and/or relative abundances of organisms of different species in samples from different groups 23 (Anderson, 2001; McArdle and Anderson, 2001). In the 3-way PERMANOVA, Distance (five 24 levels fixed factor), Year (three levels fixed factor) and Platform (two levels random factor orthogonal to the other two factors) were considered. Since the test results were significant for the 25 26 factor Platform, pB and pC were investigated separately and independently from each other.

7

For both pB and pC, multivariate patterns of variation among distances and years were tested by PERMANOVA and the significant terms were investigated using pair-wise comparisons. Successively, similarity percentage breakdown procedure (SIMPER; Clarke and Warwick, 2001) was used to determine the contribution of individual taxa towards the dissimilarity between and similarity within the two distance and year groups.

6 Univariate measures such as species abundance (N), species richness (S_m) and Simpson index (λ ; Simpson, 1949) were calculated on benthic abundance data for each sampling site at each 7 8 distance for the three sampling years. Changes in these biological indices were examined using the 9 same statistical design previously described for PERMANOVA. The random factor Platform and/or 10 the interactions between Platform and the other terms showed again significant differences. 11 Therefore a 2-way ANOVA was conducted separately for each platform considering Distance and 12 Year as fixed factors. In the case of significant interactions, a one-way ANOVA was employed for 13 each temporal factor level to compare the distance. Prior to performing statistical analyses, normal 14 distribution and heterogeneity of variances were evaluated through the Kolmogorov-Smirnov and 15 Bartlett tests, respectively (Lindman, 1992). When the latter was significant, the relationship between means and the respective standard deviations was analysed to check whether the ANOVA 16 17 assumptions were effective at any rate. Based on the tests, total abundance data (N) were log-18 transformed. The Tukey HSD test for balanced samples was used to make comparisons across all 19 pairs of group means when corresponding ANOVA tests were significant. Levels of significance 20 were set at p ≤ 0.01 (highly significant) and p ≤ 0.05 (significant).

21

22 **3. Results**

23 3.1 Overview

A total of 206 taxa were recorded at pB. This group included 92 polychaetes, 52 crustaceans, 41 molluscs, 8 echinoderms, 5 cnidarians, 3 sipunculids, 1 ascidian, 1 nemertean, 1

nematod, 1 platelmint and 1 tunicate, for a total of 1,369 ind 0.105 m⁻². The community mostly
 consisted of both limicoulus and sandy organisms.

On the other hand, a total of 139 taxa were found at pC. Of these, polychaete worms made up again the greatest proportion of the macroinvertebrate population with 67 taxa, followed by 32 crustaceans, 26 molluscs, 6 echinoderms, 4 cnidarians, 2 sipunculids, 1 nemertean and 1 platelmint, for a total of 1,481 ind 0.105 m⁻². Limicoulus species dominated the community.

Although at both pB and pC sites most taxa were typical of soft-bottom communities, close to the platforms, hard-bottom species were also found, especially crustaceans (e.g., *Pilumnus hirtellus*, *Galathea intermedia*) and molluscs (e.g., *Mytilus galloprovincialis*, *Neopycnodonte* cochlear, *Hiatella arctica*). Although the number of these species was initially very low in all the sampling sites, it increased by the end of the monitoring (during the third year) at both pB and pC.

12 3.2 Multivariate patterns

The 3-way PERMANOVA highlighted a significant difference in the abundance of 13 macrobenthos between pB and pC (Table 1). Moreover, significant interactions among Distance, 14 15 Year and Platform were shown by PERMANOVA (Table 1). Analyzing separately the two 16 platforms, significant differences among distances and years were found for both pB and pC (Table 17 2), indicating a potential effect of the platforms on the spatial and temporal variation of zoobenthic 18 communities. Pair-wise test revealed that 5 m sites for pB were different from all the other distances 19 during the overall sampling period (Table 2), while the 1000 m sites were different from all the 20 others only during the first sampling year. A spatial gradient was determined by the presence of the 21 platform from sites nearest to the platform to those furthest away. The macrofauna assemblages 22 evolved in the second and third year. The communities became similar at all distances with the 23 exception of 5 m sites. On the other hand, as regards pC, pair-wise tests pointed out a temporal 24 evolution of 5 m sites which became completely different from all the other sites in the last sampling year (Table 2). 25

SIMPER analysis showed the highest dissimilarities between 5 m sites and all the other 2 distances for both pB and pC (Table 3). The crustacean *Callianassa subterranea*, together with the polychaetes Nephtys hystricis, Capitella capitata, and Paralacydonia paradoxa were not collected 3 4 in 5 m sites for pB and were the major contributors to these dissimilarities.

5 On the other hand, the decrease of abundant polychaete species such as Caulleriella 6 caputesocis, Sternaspis scutata, and Prionospio cirrifera was associated with a greater contribution 7 of the polychaetes Marphysa bellii, Lumbrineris impatiens, of the gastropod Hyala vitrea, and of C. 8 subterranea at increasing distance from the platform. These species were the major ones 9 responsible for the differences at pC.

10 On temporal scale (year groups, Table 3) the dissimilarity was slightly higher between the 11 second and third years rather than between the first and the second ones for pB, while highly 12 decreased for pC.

13 3.3 Univariate patterns

As in the multivariate analysis, also the 3-way ANOVA highlighted highly significant 14 15 interactions among the three considered factors (Distance, Year, and Platform; Table 4). Therefore, the analysis was again computed separately for pB and pC. 16

17 As regards pB, S_m , N, and λ showed different spatial trends in each sampling year causing a significant interaction between Distance and Year (Table 5). During the first year the benthic 18 19 community was represented everywhere by a low number of both species and individuals. S_m (Fig. 2) changed from 4.63 \pm 1.03 (5 m) to 14.50 \pm 3.37 (1000 m; one-way ANOVA, *p*<0.01). A significant 20 21 increment (p < 0.05) was observed from 5 m to 30 m, which showed a value of 13.38±1.64 similar to all the other distances. No differences among distances were recorded for N, varying from 22 12.00 ± 1.55 ind 0.105 m⁻² (5 m) and 27.38±5.16 ind 0.105 m⁻² (60 m; Fig. 2). In contrast, the λ value 23 24 at 5 m (0.38±0.0.30) was significantly higher than the others (p < 0.01) due to the dominance of the polychaete Prionospio malmgreni. In the second year, S_m and N were characterized by similar 25 26 trends (Fig. 2). The lowest S_m and the highest λ (11.63±7.13 and 0.16±0.10 respectively) were still

1 recorded at 5 m. The difference was significant only for S_m in respect to 30 m (19.13±1.55; 2 p<0.05). At the end of the second monitoring year, a mussel mound started to develop close to the 3 platform hosting a varied macrobenthic community composed by both soft- and hard-bottom 4 species. As a consequence, during the third year, S_m and N reached the highest values (23.25±6.74 5 and 104.00±31.25 ind 0.105 m⁻² respectively) at 5 m, showing significant differences with the other 6 distances. λ was similar everywhere due to the high number of species with low dominance.

7 S_m , N, and λ showed different spatial trends in each year also for pC, making significant the 8 interaction between Distance and Year (Table 5). In the first sampling year the benthic community 9 living just close to the platform up to 1000 m was very poor in terms of number of both individuals 10 and species. The highest value of λ was recorded at 5 m (0.55±0.39) and the lowest at 1000 m $(0.16\pm0.05;$ Fig. 2). The only significant differences (p<0.05) were observed between 5 m and 1000 11 m for both S_m (3.38±0.98 and 11.50±2.95 respectively) and N (7.50±1.65 ind 0.105 $m^{\text{-2}}$ and 12 36.88±15.44 ind 0.105 m⁻² respectively). During the second year all the three indices did not show 13 any spatial trend and all the values were similar to each other (Fig. 2). Also in this case a mussel 14 15 mound developed at the end of the second monitoring year but differently from pB, it extended up 16 to 30 m from the platform. As a consequence, S_m and N increased within this area in the last 17 sampling period. Some hard-bottom species (e.g., N. cochlear, Ostrea edulis, Modiolarca subpicta), 18 not observed at the other distances, in fact, were recorded in addition to the typical soft-bottom taxa. The values of S_m obtained within a 30 m radius were similar to each other (5 m: 23.00±3.32; 30 m: 19 20 22.13 \pm 2.10) and significantly higher than those observed from 120 up to 1000 m (p<0.05; Fig. 2). The greater occurrence of *M. galloprovincialis* at 5 m caused a very high N value close to the 21 platform (243.50 \pm 33.15 ind 0.105 m⁻²), which was statistically different from the other distances 22 (p < 0.01). Also the 30 m value $(87.00 \pm 9.87 \text{ ind } 0.105 \text{ m}^{-2})$ was significantly higher than those 23 24 recorded at the further distances (p < 0.01). Finally, λ index was significantly higher at 5 m (0.20 ± 0.06) in comparison to 30 m and 60 m (p<0.01 and p<0.05 respectively). 25

26

1 **4. Discussion**

2 The Adriatic Sea is largely affected by many different human activities (e.g., tourism, fishing, open-sea aquaculture), and the extraction of natural gas represents one of the most 3 4 important. The quantification and interpretation of the environmental effects of offshore gas 5 platforms can be difficult and hard to generalize. Although the physical presence of the structure is 6 a common characteristic to all the platforms, several local abiotic and biotic variables, especially in the Adriatic Sea, influence the effects of each platform in a unique and different way. In fact, the 7 8 peculiar physiography of this long, narrow, shallow water basin with many fresh water inputs, 9 enhances the effects of nutrients and oxygen concentration, salinity, temperature, turbidity, currents, 10 primary and secondary production (Grilli et al., 2005; Marini et al., 2008).

11 The natural sea bottom in most areas proposed for offshore gas platforms in the Adriatic 12 shelf consists of soft sediments ranging from coastal mud to offshore coarse sand inhabited by an 13 adapted fauna. Many benthic organisms are sessile, and can hardly escape natural and 14 anthropogenic disturbances. In addition, several species are known to be susceptible to disturbances 15 or environmental changes. Due to their sessile character and ability to "integrate" environmental 16 influences over longer time scales, these species are commonly regarded as good indicators for 17 environmental impacts (Underwood, 1996), as well as for long-term changes in the ecosystem 18 (Kröncke, 1995).

19 This study assesses and compares the possible effects of two gas platforms located in the 20 north-western Adriatic Sea on the surrounding benthic communities. The aim is to find a common 21 pattern in the populations' response to the installation of Adriatic offshore structures and identify 22 the ecological features mostly leading the macrofauna changes. The two platforms are quite similar 23 in shape and dimensions, but located at different distances from the coast. Consequently, according 24 to grain size distribution maps (George et al., 2007; Harris et al., 2008) and water circulation pattern in the northwestern Adriatic basin, they are situated on different sediment types (De Biasi, 25 unpublished results; De Biasi et al., 2007) and subjected to different environmental variables. 26

Bottom currents, river plumes, and turbidity, for example, tend to decrease from the coast towards
 offshore (Boldrin et al., 2009).

3 Some differences in the composition of the benthic communities recorded in the 4 surroundings of the two platforms may be associated with the diverse sediment texture. Limicoulus 5 organisms, indeed, were found around pC, whereas a mix of sandy and limicoulus species 6 characterized pB. These differences in the benthic communities living in the natural habitats as 7 well as the distance from the coast and other abiotic features (i.e. currents, river plumes, and 8 turbidity) could explain the different response to the impacts caused by the platforms' installation 9 and drilling operations. Distance from the coast was found to be one of the main factors affecting 10 metal distribution around offshore platforms in the western Adriatic Sea (De Biasi et al., 2008). In 11 addition, a strong relationship between changes in benthic communities and decreasing gradients of 12 contaminants' concentration at increasing distance from the platforms was reported by Olsgard and 13 Gray (1995). As regards pC and pB, the results of sediment analysis did not show significant levels of contamination, suggesting a minor role of chemical pollution in determining the observed 14 15 gradients of variation in benthic assemblages (De Biasi et al., 2006, 2007, 2008). Similar findings 16 were also reported by Terlizzi et al. (2008) for inshore and offshore platforms located in the Ionian 17 Sea.

18 The zoobenthos close to pB was different from that living at the various distances from the 19 platform in the overall survey period. During the first year, indeed, just after the end of the drilling 20 operations, the community at 5 m was very poor especially in terms species number and strongly 21 dominated by the opportunistic polychaete P. malmgreni, typical of defaunated, organic matter 22 enriched areas, and also living in polluted habitats (Lu and Wu, 2000; Pearson and Rosemberg, 23 1978). The great occurrence of this species agreed with the high level of organic matter observed at 24 that time close to the platform (unpublished results). From 30 m to 120 m apart some soft-bottom species not observed at 1000 m were found. Some of these taxa were suspension- or deposit feeders 25 26 (e.g., Onuphis conchilega), likely attracted to this area by detritus and microparticulate matter made

available by the bottom mixing due to the drilling operations. Other taxa were carnivorous (e.g., *Alpheus glaber*) foraging on the above mentioned species. This scenario indicates a more severe
effect just close to the platform and a slight influence on the surrounding seabed restricted to 120 m,
likely due to the weak bottom currents characterizing the area (Fabi et al., 2005).

A different situation was observed for pC where an initial defaunation in terms of number of species and individuals was observed up to 120 m. This wider spreading out of the impact may be attributed to the more intensive bottom currents compared to pB (Paschni, unpublished results). Some opportunistic species were found within 120 m of radius (e.g., *C. caputesocis*, *P. cirrifera*) acting as pioneers in defaunated bottoms and preparing the substrate for other organisms' colonization which have more elaborate settlement needs (Pearson and Rosemberg, 1978).

In the second sampling year the benthic communities living close to the platforms showed a recovery from the previous defaunation. The appearance and/or the abundance increase of some common soft-bottom species (e.g., *O. conchilega* for pB and *H. vitrea*, *M. bellii* for pC) were recorded at all the distances. Moreover, in the case of pB a reduction of *P. malmgreni* was observed. However, this recovery process seemed to be faster at pC compared with pB, where the number of species close to the platform was still lower than those recorded at the further distances.

17 At the end of the second monitoring year a mussel mound developed close to both the platforms, extending up to 30 m in case of pC. Again, this spatial difference between the two 18 19 platforms may be related to the higher hydrodynamics characterizing pC. Specimens of M. galloprovincialis fell down from the submerged parts of the platforms providing natural hard 20 21 substrates and forming new habitat for marine epifaunal organisms which rarely occur in soft-22 bottom communities. For example, hard-bottom molluscs such as N. cochlear, Anomia ephippium, 23 and H. arctica as well as crustaceans such as P. hirtellus and G. intermedia were observed only at 5 24 m for pB and up to 30 m for pC. This process made 5 m for both platforms and 30 m for pC again different from the other distances in terms of either number of species, number of organisms and 25 26 community structure. Some mobile species use the living mussels as a food source, and the shells of

1 dead specimens as shelter (Kneib and Weeks, 1990; Warburg and Schwartz, 1993; Zariquiey 2 Alvarez, 1968). Some sessile species, instead, settle on the mussel shells utilizing them as substrate. 3 As a consequence of above, hard bottom organisms, attracted by the mussels, enrich the community (Bomkamp et al., 2004; Page et al., 1999; Thayer et al., 1997; Wolfson et al., 1979). This scenario 4 5 is common in the northern and central Adriatic Sea and it was already observed at other offshore 6 platforms located at different depths and distances from the coastline (Fabi et al., 2007; Spagnolo et 7 al., 2002, 2006; Trabucco et al., 2006, 2008). Formation of mussel mounds, increased substrate 8 heterogeneity, and development of hard bottom communities close to the offshore platforms have 9 been also reported in southern California (Love et al., 1999; Page et al., 1999; Wolfson et al., 1979), 10 in the southern Arabian Gulf (Stachowitsch et al., 2002), and in the south-eastern Australia (Currie 11 and Isaacs, 2005). An enrichment of the benthic macrofauna up to 100 m from the platform, mainly 12 due to the increase of deposit-feeding polychaetes, was also observed by Montagna and Harper 13 (1996) in the Gulf of Mexico.

In conclusion, two main typologies of effects on soft-bottom benthic communities were observed for both pB and pC. An initial short-term (~ one year), localized effect, consisting in the defaunation, may be attributed to the installation of the platforms and drilling operations. A following longer-term effect bound to the physical presence of the structures started at the end of the second year after installation and consisted of the development of mussel mounds and the subsequent macrofauna enrichment/diversification close to the platforms.

Geographical position likely plays an important role in the "timing" of the above effects. In the case of platforms placed in shallower water close to the western Adriatic coast (less than 20 km), all the potential effects appeared accelerated. In fact, the recovery of natural soft-bottom communities from the initial defaunation and the mussel mound's development started 7-8 months and about 1.5 years after the drilling operations, respectively (Fabi et al., 2005, 2007; Spagnolo et al., 2002, 2006). The higher variability of the environmental parameters and the frequent sediment mixing due to sea storms and currents typical of these near shore areas seem to be the main factors

that induced a major population dynamic reducing all the effects' duration. As a matter of fact, a
slight difference in the time of recovery was also highlighted between pC and pB, located at similar
depth but 35 km and 56 km offshore respectively.

The homogeneity observed from 30 m up to the farther distances from the platforms during the third sampling year after installation agrees with what was previously reported for other offshore structures located in the same area (Fabi et al., 2005, 2007; Spagnolo et al., 2002, 2006) and indicates that, despite the different environmental features, a 3-year monitoring planning can be effective to evaluate the potential impact on benthic communities of offshore gas platforms in the north-central Adriatic Sea.

10 From the ecological point of view, the increased seabed heterogeneity due to shells, 11 fragments and dead organisms falling for natural dislodgement or structure cleaning, associated to 12 the physical extension along the water column, make the platforms a unique habitat. Many aquatic 13 organisms, indeed, can settle on these substrates or gather around them either for orientation, 14 protection and/or greater food availability (e.g., Bomkamp et al., 2004; Fabi et al., 2004; Neira, 15 2005; Page et al., 1999, 2007; Stanley and Wilson, 1997; Wolfson et al., 1979). As a consequence, 16 after the first impact caused by the installation and drilling operations, the offshore platforms can 17 play an important ecological role in areas characterized by flat soft bottoms and relatively small 18 amount of natural reef habitat like the Adriatic Sea, inducing a diversification and an enrichment of 19 both the natural benthic and finfish communities.

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6

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1 Figure captions

- Fig. 1. Study area and position of the two investigated gas platforms, Barbara NW (pB) and Calipso
 (pC).
- 4 Fig. 2. Univariate indices of macrofaunal communities at each distance (5, 30, 60, 120, and 1000 m)
- 5 for both pB and pC during the three monitoring years. S_m = mean species richness (number of
- 6 species \pm SD); N = mean abundance (number of specimens 0.105 m⁻² \pm SD); λ = mean Simpson
- 7 index (\pm SD).

PERMANOVA analyzing differences among assemblages at increasing distance from platforms during the three monitoring years based on Gower exc 0-0 similarity

Source of variability	d.f.	SS	MS	F	р
Distance = Di	4	15937.00	3984.20	0.94852	0.5665
Year = Ye	2	22416.00	11208.00	1.05590	0.5356
Platform = Pl	1	44032.00	44032.00	30.77900	0.0001
Di x Ye	8	15807.00	1975.90	1.00670	0.4865
Di x Pl	4	16802.00	4200.40	2.93610	0.0001
Ye x Pl	2	21230.00	10615.00	7.41980	0.0001
Di x Ye x Pl	8	15702.00	1962.70	1.37190	0.0008
Residual	210	3.0043E5	1430.60		
not significant p>0.05; significant 0.05≥	p>0.01; highly significant p	≤0.01			

PERMANOVA analyzing separately differences among assemblages at increasing distance from each platform during the three monitoring years

Source of variability	d.f.	SS	MS	F	р
pB	u.i.	66	NIS .	1	P
Distance = Di	4	26528.00	6632.00	3.25000	0.0001
Year = Ye	2	17769.00	8884.40	4.35380	0.0001
Di x Ye	8	25227.00	3153.40	1.54530	0.0001
Residual	105	2.1426E5	2040.60		
Pair-wise tests for term Di x Ye					
Ye 1	Ye 2		Ye 3		
$5 \text{ m} \neq 30 \text{ m} = 60 \text{ m} = 120 \text{ m}$	$5 \text{ m} \neq 30 \text{ m} = 6$	0 m = 120 m	$5 \text{ m} \neq 30$	m = 60 m = 120 m	n = 1000 m
$5 \text{ m} \neq 1000 \text{ m} \neq 30 \text{ m} = 60 \text{ m} = 120 \text{ m}$	5 m ≠ 1000 m ≠	± 30 m			
	60 m = 120 m =	= 1000 m			
Source of variability	d.f.	SS	MS	F	р
pC	4	0140 50	2595.10	1 40010	0.0010
Distance = Di	4	9140.50	2585.10	1.49010	0.0010
Year = Ye	2	26919.00	13459.00	8.78270	0.0001
Di x Ye	8	14601.00	1825.10	1.19100	0.0203
Residual	105	1.6091E5	1532.50		
Pair-wise tests for term Di x Ye					
Ye 1	Ye 2		Ye 3		
		0 m = 120 m = 1		0 m = 60 m = 120	m = 1000 m
5 m = 30 m = 60 m = 120 m = 1000 m not significant p>0.05; significant 0.05 \ge p>0.01; hig	ghly significant p≤0.01			0 111 00 111 120	1000 11

Summary of SIMPER analysis for pB and pC

Species	Avg. ab.	Contr. %	Cum. %	Avg. dis. %		Species	Avg. ab.	Contr. %	Cum. %	Avg. dis. %	
Distances across years						Distances across years					
pB						pC					
5 m						5 m					
Prionospio malmgreni	0.51	12.15	12.15	5 m vs 30 m	88.13	Caulleriella caputesocis	1.25	11.25	11.25	5 m vs 30 m	65.98
Naticarius hebraeus	0.52	9.50	21.65	5 m vs 60 m	88.52	Marphysa bellii	0.81	9.52	20.77	5 m vs 60 m	69.49
Solemya togata	0.57	8.15	29.80	5 m vs 120 m	89.57	Sternaspis scutata	0.80	8.38	29.15	5 m vs 120 m	70.44
Myrtea spinifera	0.72	6.83	36.63	5 m vs 1000 m	89.74	Prionospio cirrifera	0.67	7.43	36.58	5 m vs 1000 m	71.06
Galathea intermedia	0.56	5.88	42.51	30 m vs 60 m	61.20	Hyala vitrea	0.70	6.78	43.36	30 m vs 60 m	57.64
Onuphis conchilega	0.53	5.72	48.23	30 m vs 120 m	64.21	Cirratulus filiformis	0.73	6.64	50.00	30 m vs 120 m	58.67
30 m				30 m vs 1000 m	65.47	30 m				30 m vs 1000 m	58.42
Callianassa subterranea	0.88	10.28	10.28	60 m vs 120 m	59.86	Marphysa bellii	1.33	23.49	23.49	60 m vs 120 m	57.85
Capitella capitata	0.89	9.75	20.73	60 m vs 1000 m	60.00	Hyala vitrea	1.07	10.62	34.11	60 m vs 1000 m	58.09
Onuphis conchilega	0.91	8.92	28.95	120 m vs 1000 m	62.38	Lumbrineris impatiens	0.68	8.37	42.48	120 m vs 1000 m	56.49
Paralacydonia paradoxa	0.79	7.68	36.63			Tharyx marioni	0.81	6.80	49.29		
Nephtys hystricis	0.79	6.74	43.37			·					
Alpheus glaber	0.66	6.64	50.01								
60 m						60 m					
Callianassa subterranea	1.12	13.46	13.46			Marphysa bellii	1.27	27.18	27.18		
Nephtys hystricis	1.13	13.39	26.85			Hyala vitrea	1.00	13.51	40.69		
Capitella capitata	0.94	8.38	35.23			Callianassa subterranea	0.71	8.57	49.26		
Paralacydonia paradoxa	0.94	8.09	43.32								
Onuphis conchilega	0.85	7.62	50.95								
120 m						120 m					
Callianassa subterranea	1.12	13.46	13.46			Marphysa bellii	1.13	21.25	21.25		
Nephtys hystricis	1.13	13.39	26.85			Hyala vitrea	1.10	16.21	37.45		
Onuphis conchilega	0.94	8.38	35.23			Lumbrineris impatiens	0.61	9.31	46.76		
Paralacydonia paradoxa		8.09	43.32			Callianassa subterranea	0.66	8.55	55.31		
Capitella capitata	0.85	7.62	50.95								
1000 m						1000 m					
Nephtys hystricis	1.39	23.54	23.54			Marphysa bellii	1.10	15.58	15.58		
Callianassa subterranea	1.31	19.88	43.42			Hyala vitrea	1.20	11.23	26.81		
Capitella capitata	0.83	10.13	53.54			Lumbrineris impatiens	0.73	10.57	37.38		
						Callianassa subterranea	0.93	10.14	47.52		
						Cirratulus filiformis	0.67	7.37	54.89		

Continue

Species	Avgab	Contr.	Cum.	Avg. dis.		Species	Avg. ab.	Contr.	Cum.	Ava die	
species	Avg. ab.	%	%	Avg. uis. %	•	Species	Avg. ab.	%	%	Avg. dis. %	
Years across distances		70	/0	/0		Years across distances		70	70	/0	
pB						рС					
1 st year						1 st year					
Callianassa subterranea	0.83	12.78	12.78	1 st vs 2 nd year	69.90	Marphysa bellii	0.86	30.44	30.44	1 st vs 2 nd year	78.41
Nephtys hystricis	0.75	12.47	25.25	1 st vs 3 rd year	76.05	Labidoplax digitata	0.53	13.05	43.49	1 st vs 3 rd year	79.03
Onuphis conchilega	0.87	11.04	36.29	2 nd vs 3 rd year	71.24	Hyala vitrea	0.62	11.45	54.94	2 nd vs 3 rd year	60.37
Alpheus glaber	0.52	6.44	42.74	-						-	
Paralacydonia paradoxa	0.60	6.38	49.12								
Sternaspis scutata	0.48	6.12	55.30								
2 nd year						2 nd year					
Nephtys hystricis	1.08	12.62	12.62			Marphysa bellii	1.19	24.58	24.58		
Callianassa subterranea	1.09	11.99	24.61			Lumbrineris impatiens	0.90	15.40	39.98		
Onuphis conchilega	1.16	11.33	35.95			Callianassa subterranea	0.80	11.01	50.99		
Capitella capitata	0.89	9.06	45.01			Hyala vitrea	0.69	6.75	57.74		
Paralacydonia paradoxa	0.72	5.69	50.70								
3 rd year						3 rd year					
Callianassa subterranea	0.89	16.10	16.10			Hyala vitrea	1.73	15.48	15.48		
Nephtys hystricis	0.83	13.35	29.45			Cirratulus filiformis	1.55	13.13	28.61		
Capitella capitata	0.84	11.38	40.83			Marphysa bellii	1.33	11.76	40.38		
Onuphis eremita	0.74	8.24	49.07			Caulleriella caputesocis	1.55	10.82	51.20		
Paralacydonia paradoxa	0.72	7.51	56.58								

Average abundances (Avg. ab.), % contribution (Contr.), and % cumulative contribution (Cum.) to the average similarity are given for each species within distance groups and year groups. Only species

reaching the cumulative contribution of about 50% are reported. % average dissimilarities (Avg. dis.) between distance and year groups are also pointed out.

R CR

Summary of 3-way ANOVA applied to mean values of mean species richness (S_m), mean abundance (N), and mean Simpson index (λ) per each distance during the three monitoring years

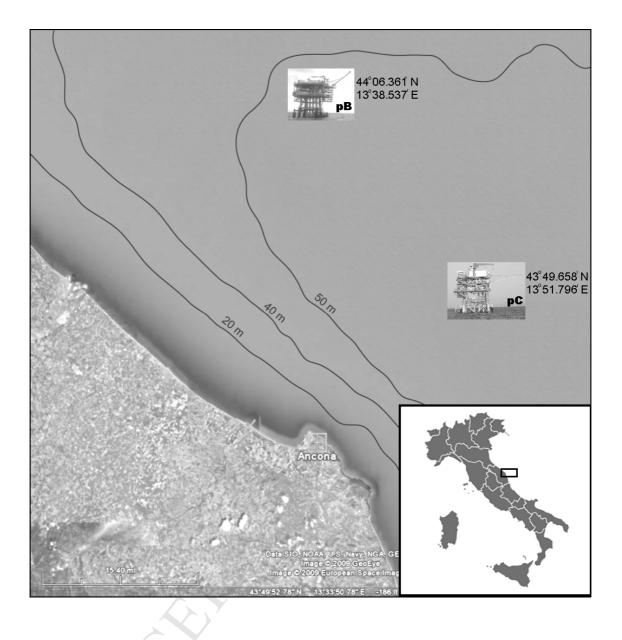
Source of	d.f.	Sm			Ν			λ		
variability		MS	F	р	MS	F	р	MS	F	р
Distance = Di	4	56.16	1.66968	0.3158	0.33	2.72861	0.1772	0.09	4.5624	0.0015
Year = Ye	2	1005.65	2.74148	0.2673	4.14	1.87247	0.3481	0.34	16.4272	0.0001
Platform = Pl	1	250.10	6.86101	0.0095	0.07	1.15466	0.2838	0.47	22.4301	0.0001
Di x Ye	8	219.69	3.93937	0.0348	0.61	7.12767	0.0059	0.07	3.4380	0.0010
Di x Pl	4	33.64	0.92271	0.4516	0.12	1.97517	0.0995	0.01	0.5387	0.7074
Ye x Pl	2	366.83	10.06308	0.0001	2.21	36.66851	0.0000	0.11	5.4710	0.0048
Di x Ye x Pl	8	55.77	1.52983	0.1484	0.09	1.40891	0.1942	0.02	0.8098	0.5946
not significant p>0.0)5; signific:	ant 0.05>p>0.0	l; highly signifi	cant p<0.01						

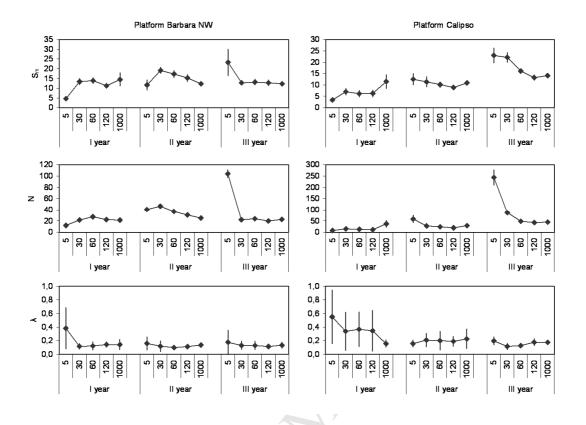
Summary of 2-way ANOVAs for pB and pC applied to mean values of mean species richness (S_m), mean abundance (N), and mean Simpson index (λ) per each distance during the three monitoring years

Source of	d.f.		Sm			Ν			λ	
variability		MS	F	Р	MS	F	Р	MS	F	Р
pB										
Distance = Di	4	24.62	0.53140	0.7130	0.07	1.25200	0.2936	0.064	5.8516	0.0003
Year = Ye	2	158.31	3.41730	0.0365	0.54	9.49700	0.0002	0.036	3.2701	0.0419
Di x Ye	8	183.33	3.95740	0.0004	0.26	4.56700	0.0001	0.023	2.1459	0.0377
pC										
Distance = Di	4	65.18	2.45220	0.0505	0.37	5.80900	0.0003	0.04	1.3678	0.250
Year = Ye	2	1214.18	45.68040	0.0000	5.82	90.44400	0.0000	0.42	13.7004	0.001
Di x Ye	8	95.12	3.4659	0.0014	0.43	6.73900	0.0000	0.06	2.1160	0.041
	8									

not significant p>0.05; significant 0.05>p>0.01; highly significant p<0.01

>> highly significant; > significant





R