THE AMPHIPODA AND CUMACEA ASSEMBLAGE RESPONSE TO LNG TERMINAL INSTALLATION IN ADRIATIC SEA (MEDITERRANEAN SEA)

Veronica Marusso, Benedetta Trabucco

Abstract: The Italian government have installed the first offshore liquefied natural gas (LNG) terminal that stores and re-gasifies liquefied natural gas due to the increasing of energy demand. The LGN terminal was positioned in the North Adriatic Sea and ISPRA, had monitored the installation and exercise activities for about 10 years; the monitoring program was executed in three phases: *ante operam*, during terminal installation and during terminal exercise. The monitoring process concerned different matrices: water, sediments, and biota These communities are recognized as an important tool to evaluate the environmental conditions, since they live in close contact with the seabed, etc. Our study was on two groups of macrozoobenthic crustacea: Amphipoda and Cumacea, providing information about the effect due to the first LGN terminal on this portion of benthic ecosystem. In the last years other LNG terminals were installed in the Italian Sea, the results of this study will be used to improve the planning sampling survey to understand the building effect on benthic communities.

Keywords: macrozoobenthic community, Amphipoda, Cumacea, LNG Terminal, North Adriatic Sea

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Introduction

The Italian government have installed the first offshore liquefied natural gas (LNG) terminal that stores and re-gasifies liquefied natural gas due to the increasing of energy demand. The LGN terminal was positioned in the North Adriatic Sea, nearby the city of Chioggia, and ISPRA, the Institute for Environmental Protection and Research, had monitored the installation and exercise activities for about 10 years. The monitoring program was executed in three phases: *ante operam*, during terminal installation and during terminal exercise. The monitoring process concerned different matrices: water, sediments, and biota; in this study, the results carried out by the macrozoobenthic community analysis, are shown. These communities are recognized as an important tool to evaluate the environmental conditions, because the animals that make up these communities live in close contact with the seabed, have a long-life cycle and can indicate a seabed perturbation, moreover these animals play an important role in the marine food chain, as they constitute the food for many benthic fishes. [1, 2, 3].

This is the first offshore LNG terminal in Italy and the first in the world Gravity Based Structure (GBS) for unloading, storing and re-gasifying [4, 5, 6]. The realization of such structures could generate different impacts both on the water column and on the sea bottom. Up to now several studies have proved that offshore activities can induce changes in the characteristics of sediment. In particular, the presence and the activity of these structures might have some sort of impact on benthic communities inhabiting the surrounding seabed. The variations in sediment physical features (e.g. sediment grain-size, sedimentation rates) might determine qualitative and quantitative changes in the structure of soft-bottom benthic communities living immediately around the installations. [7, 8, 9, 10, 11, 12, 13, 14]. Regarding GBS LNG terminal being the first in the world, very few studies on benthic communities were available, nevertheless on potential impacts on marine ecosystem due to this kind of structure [15, 16].

To verifying possible impacts on marine environment associated to the project, on behalf of Adriatic LNG, acting under the vigilance and policy guidance of the Italian Ministry for the Environment, in 2005 we ISPRA elaborated and then carried out a multidisciplinary monitoring plan. The monitoring plan provides three phases: 1) before and 2) during the construction of the structures, and 3) during terminal operation. With the purpose of monitoring disturbance degree on surrounding environment data on sediment grain size and macrozoobenthic community around the Terminal were analysed. These assemblages are commonly used as environmental indicators. They have such particular dynamics they permit an integrated valuation of the space – temporal alteration of the ecosystem. [17, 18, 19].

In this paper we report results regarding the potential effects of terminal installation and storage activities on two groups of crustacea belonging to macrozoobenthic community: Amphipoda and Cumacea, for a period ranging from 2006 to 2015. Benthic assemblage structure was examined focusing the attention on compositional characteristics of the macrofauna at increasing distances from the LNG structure. This study provided information about the effect due to the first LGN terminal in our Sea on two benthic *taxa* and allowed to collect information about temporal change of the Amphipoda and Cumacea communities. In the last years other LNG terminals were installed in the Italian Sea, the results of this study then could be used to improve the planning sampling survey to understand the building effect on benthic communities.

Material and methods

The terminal is located approximately 12 km away from the nearest Adriatic Sea coast (Porto Levante, Rovigo, Italy). A radial sampling design of 13 stations (3 stations at 100 m, 6 stations at 200 m, 4 stations at 500 m), arranged at increasing distances from the terminal site and 3 control site 4 km apart from terminal, was developed [20, 8, 21] were chosen (Figure 1). Sampling surveys took place throughout the period 2006 - 2015. In June 2006 a preliminary survey was carried out before the construction activities of the terminal (survey 1) in October 2008 a survey was carried out during yard activities of the terminal construction (survey 2); then during the following years more surveys were carried out after terminal installation (respectively 3, 4, 5, 6, 7, 8). Sediment was collected around the terminal with a Van Veen grab (0.1 m^2), taking two samples for each station. The samples were then processed through a sieve (1mm mesh-size) and the retained fraction was fixed in 4 % formaldehyde buffered with $CaCO_3$. In laboratory samples were then sorted with the use of microscopes into main taxonomic groups (Crustacea Peracardia: Amphipoda and Cumacea) and identified at the lowest possible taxonomic level (i.e. species), in addiction ecology of all species was collected and grouped into nine different bottom types: mixed, silty, soft, organic matter, hard, sandy, detritical, vegetable and gravel." [22, 23, 24, 25, 26].

Total macrofauna abundance (N), total species richness (S), Shannon index (H') and equitability (J'), Margalef specific Richness (d) were calculated in order to explore quantitative and qualitative changes in assemblage structure among stations and surveys. Similarity matrices were calculated using the Bray-Curtis similarity index [24] and data were graphically represented using non-metric

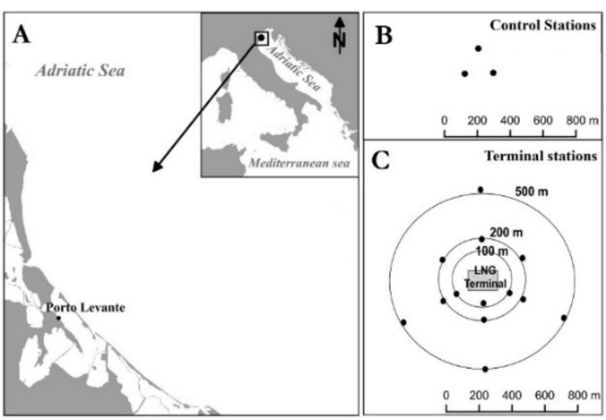

Figure 1 – Study Area (A); Control stations (B) sampling stations around the Terminal structure (C).

Multi-Dimensional Scaling (nMDS) ordinations. The grain size percentage of gravel, sand, silt and clay were provided for each station and survey, and similarity matrices were calculated using the Euclidean distance and data were graphically represented using non-metric Multi-Dimensional Scaling (nMDS) ordinations.

Results

A total of 4218 individuals belonging to 96 species of Peracarida belonging to the *taxa* of Amphipoda and Cumacea were collected. The table 1 shows the species collected during the 8 surveys.

Table 1 – List of species belong to Amphipoda and Cumacea collected during the 8 surveys.

Amphipoda	
Ampeliscidae	*Ampelisca diadema* (A. Costa, 1853)
	Ampelisca ledoyeri Bellan-Santini & Kaim-Malka, 1977
	Ampelisca massiliensis Bellan-Santini & Kaim-Malka, 1977
	Ampelisca pseudospinimana Bellan-Santini & Kaim-Malka, 1977
	Ampelisca rubella A. Costa, 1864
	Ampelisca ruffoi Bellan-Santini & Kaim-Malka, 1977
	Ampelisca sarsi Chevreux, 1888
	Ampelisca spinifer Reid, 1951
	Ampelisca spinipes Boeck, 1861
	Ampelisca tenuicornis Liljeborg, 1856
	Ampelisca truncata Bellan-Santini & Kaim-Malka, 1977
	Ampelisca typica (Spence Bate, 1857)
Aoridae	*Aora spinicornis* Afonso, 1976
	Autonoe spiniventris Della Valle, 1893
Argissidae	*Argissa hamatipes* (Norman, 1869)
Atylidae	*Nototropis guttatus* (A. Costa in Hope, 1851)
	Nototropis massiliensis (Bellan-Santini, 1975)
	Nototropis vedlomensis (Spence Bate & Westwood, 1862)
Bathyporeiidae	*Bathyporeia lindstromi* Stebbing, 1906
Calliopiidae	*Apherusa vexatrix* Krapp-Schickel, 1979
Caprellidae	*Liropus elongatus* Mayer, 1890
	Phtisica marina Slabber, 1769
	Pseudolirius kroyeri (Haller, 1879)
Cheirocratidae	*Cheirocratus assimilis* (Lilljeborg, 1852)
	Cheirocratus sundevallii (Rathke, 1843)
Corophiidae	*Apocorophium acutum* (Chevreux, 1908)
	Corophium orientale Schellenberg, 1928
	Leptocheirus bispinosus Norman, 1908
	Leptocheirus guttatus (Grube, 1864)
	Leptocheirus longimanus Ledoyer, 1973
	Leptocheirus mariae Karaman, 1973
	Leptocheirus pectinatus (Norman, 1869)
	Medicorophium aculeatum (Chevreux, 1908)
	Medicorophium rotundirostre (Stephensen, 1915)
	Medicorophium runcicorne (Della Valle, 1893)
Dexaminidae	*Dexamine spiniventris* (A. Costa, 1853)
	Dexamine spinosa (Montagu, 1813)
Eusiridae	*Eusirus longipes* Boeck, 1861
	Rhachotropis ind. S.I. Smith, 1883
Gammaridae	*Gammarus aequicauda* (Martynov, 1931)
	Gammarus insensibilis Stock, 1966
Iphimediidae	*Iphimedia gibbula* Ruffo & Schiecke, 1979
Ischyroceridae	*Centraloecetes dellavallei* (Stebbing, 1899)
	Ericthonius brasiliensis (Dana, 1853)
	Ericthonius punctatus (Spence Bate, 1857)
	Siphonoecetes ind. Krøyer, 1845
Kamakidae	*Cerapopsis longipes* Della Valle, 1893
Leucothoidae	*Leucothoe incisa* Robertson, 1892
	Leucothoe oboa Karaman, 1971
	Leucothoe occulta Krapp-Schickel, 1975
	Leucothoe serraticarpa Della Valle, 1893
	Leucothoe spinicarpa (Abildgaard, 1789)
Liljeborgiidae	*Liljeborgia dellavallei* Stebbing, 1906
	Liljeborgia psaltrica Krapp-Schickel, 1975

Lysianassidae	*Lysianassa caesarea* Ruffo, 1987
	Lysianassa costae H. Milne Edwards, 1830
Maeridae	*Ceradocus (Ceradocus) orchestiipes* A. Costa, 1853
	Hamimaera hamigera (Haswell, 1879)
	Maera grossimana (Montagu, 1808)
	Maera sodalis Karaman & Ruffo, 1971
	Othomaera schmidtii (Stephensen, 1915)
Megaluropidae	*Megaluropus* ind. Hoek, 1889
Melitidae	*Abludomelita gladiosa* (Spence Bate & Westwood, 1862)
Nannastacidae	*Campylaspis glabra* Sars, 1878
	Procampylaspis bonnieri Calman, 1906
Oedicerotidae	*Bathymedon* ind. G.O. Sars, 1892
	Deflexilodes acutipes (Ledoyer, 1983)
	Deflexilodes griseus (Della Valle, 1893)
	Deflexilodes subnudus (Norman, 1889)
	Kroyera carinata Spence Bate, 1857
	Perioculodes longimanus (Spence Bate & Westwood, 1868)
	Westwoodilla rectirostris (Della Valle, 1893)
Photidae	*Gammaropsis dentata* Chevreux, 1900
	Gammaropsis maculata (Johnston, 1828)
	Gammaropsis ulrici Krapp-Schickel & Myers, 1979
	Latigammaropsis togoensis (Schellenberg, 1925)
	Photis longicaudata (Spence Bate & Westwood, 1862)
Phoxocephalidae	*Harpinia dellavallei* Chevreux, 1911
	Metaphoxus ind. Bonnier, 1896
Stenothoidae	*Stenothoe* ind. Dana, 1852
Tryphosidae	*Orchomene humilis* (A. Costa, 1853)
	Orchomene massiliensis Ledoyer, 1977
	Orchomene similis Chevreux, 1912
	Tryphosa nana (Krøyer, 1846)
Cumacea	
Bodotriidae	*Bodotria scorpioides* (Montagu, 1804)
	Iphinoe rhodaniensis Ledoyer, 1965
	Iphinoe serrata Norman, 1867
	Iphinoe tenella Sars, 1878
Leuconidae	*Eudorella nana* Sars, 1879
	Eudorella truncatula (Bate, 1856)
	Leucon (Leucon) mediterraneus Sars, 1878
Diastylidae	*Diastylis cornuta* (Boeck, 1864)
	Diastylis neapolitana Sars, 1879
	Diastylis richardi Fage, 1929
	Diastylis rugosa Sars, 1865
	Diastylis tumida (Liljeborg, 1855)

The highest value of abundance N (806 individuals) was observed at the first survey (1) while the lowest value was recorded at the survey 4 (Fig 2a). The highest value of number species (S) was observed at survey 7, while the lowest at survey 5 (Fig 2b).

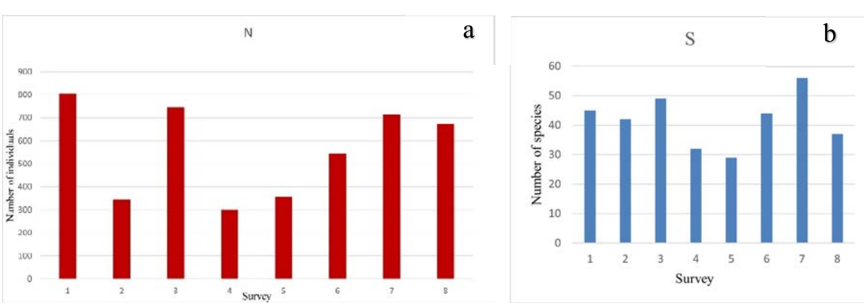

Figure 2 – The average value of Abundance N (a) and Number of species S (b) calculated for each survey.

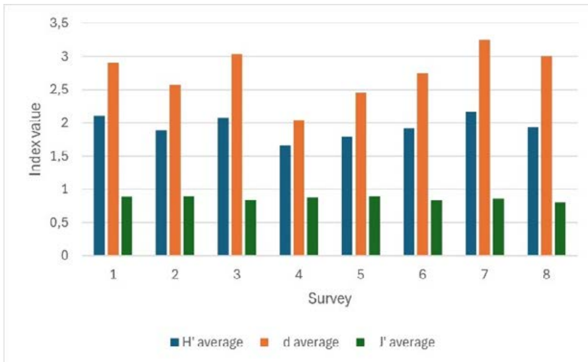

Figure 3 – The average value of H’, d and J’ index calculated for each survey.

The average value of index H’, d and J’ was calculated for each survey (Fig. 3), the highest value of H’ was observed in the survey 7, while the lowest in the survey 4, the highest value observed for the index d was in the survey 7 while the lowest in the survey 4, at the end the J’ index showed the highest value in the survey 2 and the lowest in the survey 8.

All stations collected during the 8 surveys were showed at the MDS plot. A weak difference among the stations near and far from the terminal installation was observed, indeed it is possible to note that the stations near the terminal were generally separated from the others. It is possible observe a low difference among the eight surveys analyzed especially in the surveys 4 and 5 (Fig 4a). Also, the centroid analysis showed the difference among the surveys: the Peracarida community observed during the survey 2 (during the terminal installation) showed differences from that observed in survey 1, while in the survey 3 the community seem to be more like survey 1then survey 2 (Fig 4b). The assemblage observed in the surveys 4, 5, 6 and 7 showed weakly mutual different each other, while the community observed in the survey 8 seemed to be like the survey 3.

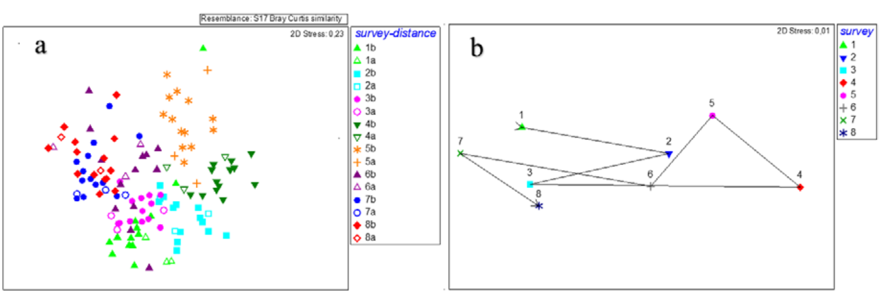

Figure 4 – a) The MDS plot shows the similarity among the stations and the surveys calculated on the Peracarid community. Number from 1 to 8 indicated the surveys, the empty symbol represents the stations near to the terminal (call a), the full symbols represent the stations far from the terminal at a distance at greater than 100 meters, (call b). b) The plot shows the centroid distance among the surveys.

The MDS plot (Fig 5) showed the stations similarity carried out on the sediment features: two different groups of stations were plotted: one at the lower part of the graphic characterized by the gravel presence, and another in the higher part of the plot the stations with the sand, clay and silt where weakly difference among the stations near and far from the terminal installation and among the surveys was observed. Analysis of the centroids showed that the granulometric conditions produced by the installation of the terminal seemed to return in a condition more similar to the initial conditions one (survey 1).

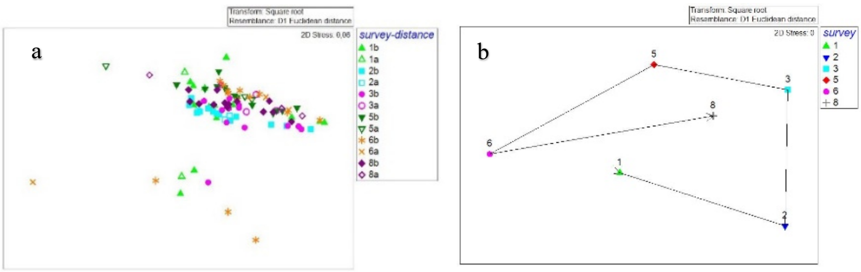

Figure 5 – a) The MDS plot shows the similarity among stations and survey calculated on grain size. Number from 1 to 8 indicated the surveys, the empty symbol represents the stations near to the terminal (call a), the full symbols represent the stations far from the terminal at a distance at greater than 100 meters, (call b). b) The plot shows the centroid distance among the surveys calculated on grain size.

Information about ecology of each species collected was gathered and grouped in nine different bottom types: mixed, silty, soft, organic matter, hard, sandy, detritical, vegetable and gravel. Figure 6 showed the abundance of species and their respectively ecology only for the community belong to the station near the terminal (T4, T16 and T27) for each survey.

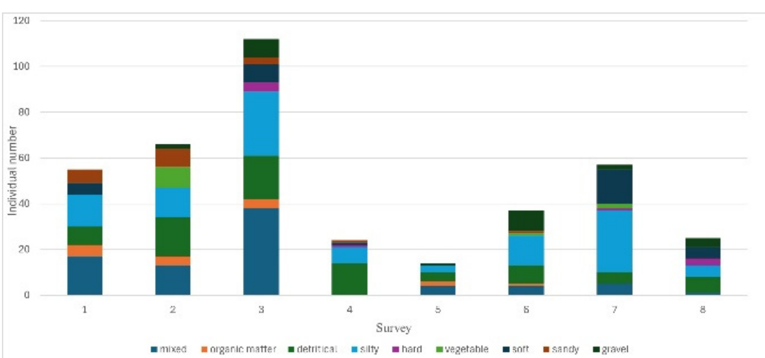

Figure 6 – The ecology of specie observed at the three stations close to the terminal installation (T7, T17 and T27) in each survey.

Species with mixed and detritical ecology showed an increase of their abundance after construction of the terminal structure (survey 3) and a decrease on the following surveys. The species related to organic matter showed a continuous decrease from the first to the last survey. Species living in silty, soft and gravel bottom showed a fluctuating trend, while species linked to the hard bottom showed a peak in the survey 3 followed by a decrease in the surveys 4, 5 and 6, and a recovery in the survey 7. For the species linked to vegetable bottom a peak was observed during the survey 2, during the terminal installation, and finally, sandy species showed a peak in the survey 2 and then a decrease at the following surveys.

Figure 7 –The most abundant specie at each survey.

The most abundant species collected were Amphipods belonged to Ampelisca genera, while the Cumaceans were represented generally with few individuals that belonged to few species (Fig. 7). *Ampelisca diadema*, *Ampelisca typica* e *Ampelisca tenuicornis* were respectively the most abundant species of surveys 1 and 4, while in the survey 2 *Ampelisca sarsi*, *A. typica* and *Leucothoe oboa*. *A. tenuicornis*, *A. diadema* and *A. typica* were the most abundant in the survey 3. *A. tenuicornis*, *A. typica* and *Ampelisca ledoyeri* were the most abundant in the surveys 5. The Caprellidae *Phtisica marina* and *A. tenuicornis* and *A. ledoyeri* showed the hight number of individuals in the survey 6, while *A. typica*, *P. marina* and *Photis longicaudata* were the most abundant in the survey 7. At the end *A. typica*, *A. diadema* and *Ericthonius punctatus* were the species with the hight number of individuals in the survey 8.

Discussion

Cumaceans and Amphipods are benthic organisms, generally 1–10 mm in size, that are strongly linked to the seabed where they can burrow tunnels. These organisms are characterized by morphological features that differentiate them from other peracarid crustaceans. Cumaceans together with Amphipods live in seawater from intertidal shelves to great depths [28] but can also be found in brackish water and rivers [29]. They can be influenced by the type and nature of the sediment, as well as its organic matter content, which can generate changes in their abundance [30, 31]. These taxa represent an important link in marine trophic webs because these animals are common food for many species of fish living near the bottom. [32]. They are also known as indicators of organic enrichment [30] and the eutrophication of soft bottoms [34] and are therefore often used together with other benthic organisms to monitor environmental quality [35, 36], as also requested by the major European directives (WFD/2000/60/EC; MSFD/2008/56/EC).

The study showed that the Terminal installation generated a change on the Peracarid benthic community, principally focused on the period during the installation and at the stations near the structure. The lowest N value observed in the survey 2, is caused by sediment movement resulting from the installation activities, nevertheless the high value of the number of specie observed in this survey indicate that the assemblage has preserved the biodiversity as showed by the indexes (H' d and J). Nevertheless, after the terminal installation effect, the community on average recovered the initial abundance. In fact, our studies seemed to reveal that changes in benthic assemblages were mostly linked to the construction phase of the LNG plant, and, to some extent, are limited to the first period of activity of the terminal [13]. During the following surveys just some oscillations were observed, in terms of abundance and species number. These oscillations are most likely due to natural events, which could determine the sediment grain size modification, and then change in the benthic community structure [37, 38, 39, 40].

From the 1 to the 8 surveys, following the sediment modifications, it was observed a change of the dominant species at each survey, the most abundant taxon at most of the surveys was the taxon of Ampelisca. The most abundant species in the survey 1 were linked to mixt and silty sediment (*A. diadema* and *A. typica*), while in the survey 2 the most abundant species prefer organic matter and mixed sediment (*A. sarsi* and *A. typica*). The presence of this species is compatible with the effects product by the installation activity that can caused sediment movement and consequent resuspension of organic matter.

In the survey 3 the most numerous species collected were detritic and mixed respectively (*A. tenuicornis* and *A. diadema*), so after human activity it was possible observed the effects of Terminal construction. In the assemblage collected during the survey 4 it was possible to observe came back the most numerous species *A. typica* that was link to silty sediment with *A. diadema*, instead linked to mixed bottom. The most abundant species link to detritic bottom was observed in the survey 5 and 6 and at the end in the survey 7 and 8 the most abundant species *A. typica* was linked to silty bottom and mixed sediment *A. diadema*.

The Peracarida community appeared to return to similar conditions observed during the survey 1 after 4 years.

Conclusions

Environmental quality assessment, according to the guidelines, involves the use of a species list that associates each species with an ecological class based on the group's ability to indicate a disturbed or undisturbed environment. The collection of data allows species lists to be updated with new records, therefore representing an important aspect for the conservation of marine ecosystems and for peracarids taxonomy insiders.

The scarcity of amphipods and in particular cumacean taxonomists and experts is another predicament: in fact, the taxonomist's profession requires many years of study and practice, and unfortunately the existing reference bibliography used as a credential tool for identification purposes is limited and often outdated. We hope more studies on minor taxa, such as peracarids in general, will be published in order to address this knowledge gap.

The data obtained by the sample analysed confirm findings from similar studies conducted in other coastal areas and highlight the challenges in predicting the processes and timing of colonization by macrozoobenthic organisms. Monitoring activities remain highly relevant because the evolution of the benthic population needs to be studied and evaluated over time. The collection of a longtime data series is expected to bring more light on the effects of the presence of the LNG structure on the surrounding marine seabed. First analyses seem to indicate that the effects on benthic macrofauna, if any, were limited to the period of the construction yard, and some signal of alterations in diversity measures were found for the subsequent 4 years. This research allowed us to gain a high amount of data that could provide a reference for this type of studies in the future and will let us to optimize field and laboratory work, addressing monitoring actions at the best.

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