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The application of the Weight-Of-Evidence approach for an integrated ecological risk assessment of marine protected sites

Manuela Piccardo [a,b,*], Verdiana Vellani [a,b], Serena Anselmi [c], Eleonora Grazioli [c], Monia Renzi [a,b], Antonio Terlizzi [a,e], Lucia Pittura [d], Giuseppe D'Errico [d], Francesco Regoli [d], Stanislao Bevilacqua [a,b]

[a] Dipartimento di Scienze della Vita, Università di Trieste, 34127 Trieste, Italy
[b] CoNISMa, Consorzio Interuniversitario per le Scienze del Mare, Piazzale Flaminio 4, 00196 Roma, Italy
[c] Bioscience Research Center, Via Aurelia Vecchia, 32, 58015 Orbetello, GR, Italy
[d] Department of Life and Environmental Science, Polytechnic University of Marche, Via Brecce Bianche, 60131, Italy
[e] Stazione Zoologica Anton Dohrn, Naples, Italy

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ABSTRACT

The effective management of marine ecosystems in the face of growing anthropogenic pressures requires the integration of data from different ecological components. Holistic approaches to evaluate the ecological status of marine ecosystems are still scarce, likely due to the challenge of integrating the complex information from a variety of indicators. In this study, we provided an application of a quantitative Weight-Of-Evidence (WOE) model based on the Sediqualsoft® software, combining environmental and biological data to assess ecological risk in soft-bottom habitats within Natura sites 2000 in the Northern Adriatic Sea (Mediterranean Sea). Here, the WOE approach combined three lines of evidence (LOE): chemical characterization (LOE1), ecotoxicological properties (LOE4), and benthic community status (LOE5). A separate hazard quotient was derived for each LOE prior to a weighted integration into a synthetic WOE assessment. The chemical analysis of the sediments revealed concentrations of pollutants far lower the reference limits, except for As and Hg and for polycyclic aromatic hydrocarbons which determined a 'Slight' to 'Severe' chemical hazard in coastal sites. Ecotoxicological hazard was rated as 'Absent' at all sampling stations, and the analysis of benthic communities indicated 'undisturbed' conditions for most sites. The WOE approach classified the overall ecological risk to be 'Absent' for offshore sites and 'Slight' in nearshore sites. Although results suggested a general low ecological risk, the potential for future risks is recognized, especially in coastal areas, due to well-known sediment pollution in the region. The application of the WOE approach may represent a valuable tool for managing marine protected sites, and to characterize the overall ecological status of these areas and improve conservation strategies in highly anthropized environmental contexts.

1. Introduction

The marine environment is experiencing an increasing level of human pressure at a global scale due to overfishing, contamination by hazardous substances, nutrient inputs, physical alteration of habitats, and climate change (Bryhn et al., 2020; Lotze et al., 2006). Cumulative human impacts are causing a widespread decline of marine biodiversity (Korpinen and Andersen, 2016), with potentially profound consequences to the functioning of marine ecosystems and the provision of goods and services that yield socio-economic benefits (Barbier, 2017). Efforts in environmental impact assessment have thus moved from single-impact perspectives towards more comprehensive approaches investigating the combined ecological response of marine ecosystems to multiple interacting human disturbances (Bevilacqua et al., 2018; Halpern et al., 2019; Gissi et al., 2021; Korpinen et al., 2021). The integration of data from different ecosystem components in relation to the complexity of anthropogenic pressures, however, remains one of the most challenging issues for environmental management (Rosenberg & McLeod, 2005). Indeed, progress in the implementation of new methods and indicators to assess the ecological status of several marine ecosystem

* Corresponding author at: Department of Life Science, University of Trieste, Via L. Giorgieri 10, 34127 Trieste, Italy.
E-mail address: manuela.piccardo@units.it (M. Piccardo).

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components flourished throughout the last two decades (e.g., Vollenweider et al., 1998; Borja et al., 2000; Gobert et al., 2009; Teixeira et al., 2016), although inclusive analytical frameworks integrating the response of a range of abiotic and biotic variables to human pressures have been rare, despite the emphasis on holistic approaches to environmental issues that characterizes policies and regulations at international level (Leslie & McLeod, 2007; Borja et al., 2020).

In the European Union, the Marine Strategy Framework Directive (MSFD; EC, 2008) represents one of the most comprehensive sets of legislation on the marine environment. The MSFD requires monitoring marine ecosystems considering 11 descriptors, ranging from biodiversity to underwater noise, for an adaptive management that aims to achieve and maintain the 'Good Environmental Status' over the European seas and oceans. Attempts to put into practice an integrated assessment combining the different descriptors have been scant in the European Member States (Borja et al., 2016), probably due to the limited availability of suitable tools allowing the aggregation of information at different scales (spatial and temporal) and the integration of multiple environmental and biological aspects. One example in this direction is the implementation of the NEAT tool (Nested Environmental status Assessment Tool), which incorporates several biological (e.g., population density of target species) and abiotic (e.g., concentration of pollutants) variables, though the latter have generally received little consideration (Kazanidis et al., 2016; Fraschetti et al., 2022; but see Borja et al., 2021). The second pillar of management of European marine and coastal waters is the Water Framework Directive (WFD; EC, 2000), which focuses on preventing deterioration, protecting and enhancing the status of aquatic ecosystems. The WFD involves a dual assessment of the health of water bodies accounting for both biological and chemical quality elements and it is based on the "one-out, all-out" principle, so that the worst status recorded among the considered quality elements determines the final status of the water body, irrespective of the conditions of all the other quality elements (Prato et al., 2014). Hence, the two assessments run in parallel in the absence of a truly integrative framework, which makes it difficult to achieve a weighted evaluation of the system under study.

A critical aspect of monitoring and assessment in the framework of both WFD and MSFD concerns the ancillary role of sediments in the evaluation of the environmental status. The WFD, for example, does not clearly provide homogeneous procedures to assess the status of aquatic sediments (Carere et al., 2012). This is mostly due to the fact that WFD sets ecological quality standards with no legally binding ecological quality standards for sediments (Vasilakopoulos et al., 2022). Since thresholds and criteria for priority substances under the MSFD refers to the ecological quality standards defined by the WFD, the lack of references also extends to the assessment of pollutants in sediments under the MSFD (descriptor D8 – Hazardous substances). The quantification of thresholds through the cooperation among Member States, which should compensate this lack, is still far from being set for most regions and sub-regions (Vasilakopoulos et al., 2022), representing a critical regulatory gap for a comprehensive assessment of the environmental status, as marine sediments act as sinks for a variety of organic and inorganic pollutants (Perelo, 2010).

Among the EU Member States, the Italian government provided one of the most complete sets of legislation on the environmental management of marine sediments with the national regulation enshrined in the Decree of the Italian Ministry of the Environment no. 173/2016 (hereafter referred to as DM 173/2016), concerning the technical procedures, monitoring and assessments of all activities related to the movement, dislodgement, discharge, use and treatment of marine sediments, also defining baseline threshold concentrations of pollutants (all priority substances and metals) for national marine waters. Given its completeness and since the Italian marine waters span over different Mediterranean sub-basins (i.e., NW Mediterranean Sea, Ionian Sea and Adriatic Sea), being therefore representative of different marine ecoregions, thresholds set in DM 173/2016 also found applications for assessments in neighboring coastal EU countries (e.g., Borja et al., 2021).

In the framework of DM 173/2016, the Sediqualsoft® software was implemented as an operational tool to integrate chemical and ecotoxicological features of marine sediments, though the software is actually structured to encompass several additional aspects. Specifically, Sediqualsoft® is a quantitative Weight-Of-Evidence (WOE) model developed to elaborate data from different lines of evidence (LOEs; i.e., sediment chemistry – LOE1, bioaccumulation – LOE2, biomarkers – LOE3, ecotoxicological bioassays – LOE4, and benthic communities – LOE5) in ecological risk assessment studies of contaminated or potentially contaminated sites. A synthetic and quantitative hazard index is provided for each of the considered LOEs before their overall integration in the final WOE assessment (Regoli et al., 2019), and the independent elaborations for each LOE are based on specific criteria that take into account the different data types. These criteria have been validated through national and international case studies for ecological risk assessments associated with polluted sediments, harbor areas, or complex natural and human disturbances on the marine environment (Benedetti et al., 2012, 2014; Broccoli et al., 2021; d'Errico et al., 2021; Li et al., 2023; Manfra et al., 2021; Maradonna et al., 2020; Morroni et al., 2020; Piccardo et al., 2021; Pittura et al., 2018; Piva et al., 2011). The software assigns an ecological risk class to sediments samples based on a multidisciplinary approach that integrates the evidence from chemical analyses on a wide range of priority and hazardous contaminants, ecotoxicological bioassays and assessments on other biological components (e.g., macrobenthic community structure).

The software-based WOE approach relying on Sediqualsoft® has found its preferential field of application in assessing the ecological risk in heavily polluted areas (e.g., Piva et al., 2011; Benedetti et al., 2014; Regoli et al., 2019), while its potential application to understand whether conservation measures are effective in mitigating the ecological risk within protected areas has been underexplored. Moreover, though the approach has benefited of the integration of biological data (e.g., biomarkers), case studies integrating information on macrobenthic assemblage structure are limited. Since sediments and associated invertebrate assemblages play a central role for the functioning of marine ecosystems, assessing their ecological quality status and pollution levels is essential for environmental management and marine biodiversity conservation (Snelgrove et al., 1997).

In this study, we applied the WOE approach of Sediqualsoft® by integrating both environmental and biological community data in order to assess the ecological risk in soft bottom habitats within Natura 2000 sites, established based on the European Council Directive 92/43/EEC on the conservation of natural habitats and of wild fauna and flora (hereafter EU Habitat Directive) in the Northern Adriatic Sea (Mediterranean Sea). This approach can represent a powerful tool that, while assessing the ecological status of marine habitats based on a multidisciplinary perspective, may also allow to adapt management actions to ecological risk in areas under protection regimes.

2. Material and methods

2.1. Study area and sampling design

The study area was located in the Gulf of Trieste (North eastern Adriatic Sea, Fig. 1), a shallow (average depth ~19 m), semi-enclosed basin characterized by large river runoff and wide seasonal variability in temperature and salinity (Trobec et al., 2018). It is one of the most anthropized areas of the Adriatic Sea, where maritime transport, coastal industries, fisheries, and tourism activities densely concentrate (Giani et al., 2012; Furlan et al., 2019), and well represent the typical Mediterranean marine biodiversity and priority coastal habitats.

Four sites (hereafter referred to as C, I, S, and B) were selected to be representative of the range of soft bottom habitats at a regional scale (Fig. 1). Two of them, namely C and I, are close to the coast (within 4 km), while the other two sites (S and B) are far from the shoreline (>8

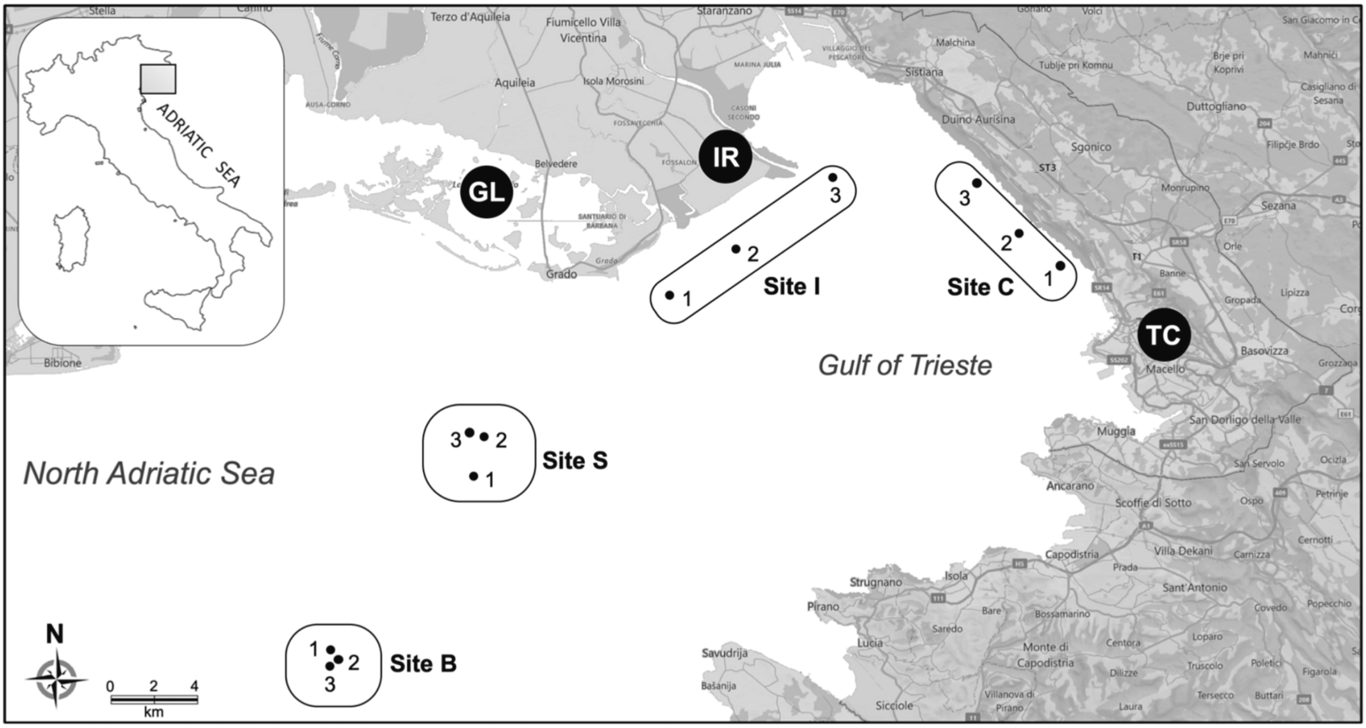

Fig. 1. Study area and sites. C = Marine Protected Area of Miramare – City of Trieste (TC), I = Isonzo/Soča River (IR) – Grado lagoon (GL), S-B = offshore areas with biogenic outcrops. For each site, sampling stations (1, 2, 3) were also showed.

km up to 20 km offshore) (Table A1, Appendix A in supplementary material). Site C embraced sandy bottoms nearby the coast from the Marine Protected Area of Miramare until the urban area of Trieste, one of the largest Italian harbors. Site I comprised a mosaic of coastal muddy bottoms and seagrass (*Cymodocea nodosa* (Ucria) Ascherson) beds from the mouth of the Isonzo/Soča River to the coastal lagoon of Grado. Finally, sites S and B were in offshore areas characterized by the presence of alternate patches of sandy/detritic bottoms and biogenic outcrops.

All sites fell entirely (I, S, B) or partially (C, station 2) within the Natura 2000 network implemented under the EU Habitat Directive as Special Protection Areas (SPAs), Special Areas of Conservation (SACs) or Sites of Community Importance (SCIs), due to the presence of priority marine habitats and their ecological importance as reproductive, sheltering and feeding grounds for several marine species, including seabirds and a number of invertebrates and fishes of commercial interest that sustain the local and regional tourism and fishery economy. Specifically, site C included the Marine Protected Area (MPA) of Miramare (SCI IT3340007), site I is embedded within three SPAs/SACs (IT3330005, IT3330006, IT3331001), whereas sites S and B were within a Site of Community Importance (SCI IT3330009). At each site, three sampling stations were randomly selected and soft sediments were sampled (November 2021-February 2022) using a standard stainless steel Van Veen grab (14 L). For chemical and ecotoxicological analyses, a single sediment sample per station was collected with a steel spatula from a grab sample and divided into separate aliquots. Aliquots were kept in plastic bottles and stored at −20 °C and +4 °C, for chemical and ecotoxicological bioassays respectively, until processing. For the analysis of benthic assemblages, three replicate grab samples were taken at each station. Sediments were sieved onboard on 1 mm mesh and immediately fixed in 70 % solution of ethanol and seawater.

2.2. The Weight Of Evidence (WOE) approach

The Weight-Of-Evidence (WOE) approach proposed by Regoli et al. (2019) was used to obtain a multidisciplinary characterization by combining three different lines of evidence (LOE): (i) the traditional lines of chemical characterization (LOE1), including compounds classified as 'non-priority' to 'priority and hazardous' according to the European Directive 2013/39 (EC, 2013) on priority substances in water policy; (ii) the investigation of ecotoxicological effects of sediments (LOE4) through a set of bioassays following standardized protocols (involving algae, sea urchins, and amphipods); and (iii) the assessment of the ecological status of benthic communities (LOE5) based on the AMBI index (Borja et al., 2000).

2.2.1. LOE1 – Chemical analysis

Concentrations in sediment samples of a total of 58 contaminants were analytically determined, including trace metals (i.e., Al, As, Cd, Cr, Cu, Fe, Hg, Ni, Pb, V, Zn), organotin compounds (i.e., TBT), 13 congeners of dioxin-like and non-dioxin-like polychlorinated biphenyls (PCBs), 16 types of pesticides, total heavy hydrocarbons (C > 12), and 16 polycyclic aromatic hydrocarbons (PAHs). Details on analytical procedures are given in supplementary material (Table A2, Appendix A in supplementary material).

The conversion of the chemical data into the corresponding hazard quotient (HQ_C) is described in Piva et al. (2011). For each compound with normative reference values, a ratio to reference (*RTR*) was calculated as the ratio between the measured concentration and the limit given in the sediment quality guidelines. This study applied the limits of concentration from the DM 173/2016, which are summarized in Table A2 (Appendix A in supplementary material).

To highlight the importance of the most hazardous chemicals according to the EC (2013), the *RTR* value was multiplied by a correction factor (weighting) assigned to each pollutant. A value of 1.1 or 1.3 was applied to pollutants included in the list of 'priority' or 'priority and hazardous' substances respectively, while the weighting for non-priority substances was set to 1.0 (Piva et al., 2011). The weighted *RTR* values were used to calculate the hazard quotient for chemicals (HQ_C) as follows (see Piva et al., 2011 for further details):

$$HQ_C = \frac{1}{n}\sum_{i=1}^{n} RTR_W + \sum_{j=1}^{m} RTR'_W$$

where RTR_W referred to weighted RTR values of the n chemicals which had $RTR \leq 1$ (i.e., concentration below the legal limit), and RTR'_W to the weighted RTR values of the m chemicals which had $RTR > 1$.

Finally, the values of HQ_C were assigned to six classes of chemical hazards (*Absent* [HQ_C: 0 - <0.7], *Negligible* [HQ_C: 0.7 - <1.3], *Slight* [HQ_C: 1.3 - <2.6], *Moderate* [HQ_C: 2.6 - <6.5], *Major* [HQ_C: 6.5 - <13], *Severe* [HQ_C: ≥13]), which were defined based on expert judgment, depending on the number, typology, and magnitude of the exceeding chemicals (Piva et al., 2011; Regoli et al., 2019).

2.2.2. LOE4 – ecotoxicological bioassays

A battery of three ecotoxicological bioassays based on three different species were performed on sediment samples from each station according to standardized procedures, for a total of 36 bioassays. Selected species for bioassays were the diatom *Phaeodactylum tricornutum* (testing acute effects of elutriates on growth in six replicates; ISO 10253:2017), the sea urchin *Paracentrotus lividus* (testing chronic effects of elutriates on larval development in four replicates; ISPRA-SNPA, 2017) and the amphipod *Monocorophium insidiosum* (testing acute effects of the solid phase on mortality in three replicates; ISO 16712:2005).

For the conversion of the ecotoxicological data into the corresponding HQ, a specific threshold and weighting were given to the different bioassays, depending on the biological endpoints, the tested matrix, the exposure time, and the possibility of hormetic responses (see Regoli et al., 2019 for further details). For each bioassay a, the effect (E) was calculated as the percentage of variation compared to the control conditions. The effect E was corrected for the statistical significance according to function z and divided by the limit threshold of the assay to obtain a corrected and standardized effect (E_w). The cumulative hazard quotient for bioassays ($HQ_{Battery}$) is then obtained by summing the different E_w of the assay battery after a further weighting based on a factor that account for the biological significance of the endpoint and the exposure conditions (w_2).

The weighting factor w_2 was set to 1.0 for behavioral changes, 1.2 for growth, 1.5 for mutagenicity/genotoxicity, 1.9 for reproduction and development, 2.4 for bioluminescence, and 3.0 for mortality (Piva et al., 2011). In this study, therefore, w_2 was set to 1.2 for the effect on *P. tricornutum*, 1.9 for effect on *P. lividus*, and 3.0 for the effect on *M. insidiosum*, and $HQ_{Battery}$ calculated as follows:

$$HQ_{Battery} = \sum_{k=1}^{a} E_w \bullet w_2$$

Finally, the cumulative $HQ_{Battery}$ is normalized to range between 0 and 10, by dividing its value to the value obtained if all assays have $E = 1$. Values of $HQ_{Battery} < 1$ indicate that the effect for all bioassays is below their specific threshold, $HQ_{Battery} = 1$ means that all measured bioassays have an effect equal to their specific threshold, and $HQ_{Battery} = 10$ is obtained when all assays have a 100 % effect. Then, based on the normalized value of $HQ_{Battery}$, one of five hazard classes from *Absent* to *Severe* were assigned to the sample as for LOE4. Values of $HQ_{Battery}$ for class assignment were: <1.0 (*Absent*), 1.0 - <1.5 (*Slight*), 1.5 - <3.0 (*Moderate*), 3.0 - <6.0 (*Major*), 6.0 – 10.0 (*Severe*).

2.2.3. LOE5 – Status of benthic assemblages

Samples were sorted under magnification and macrobenthic organisms counted and identified to the lowest possible taxonomic level by expert taxonomists. For the analysis of data on benthic assemblages (LOE5), a specific procedure implemented by Regoli et al. (2019) and based on the AZTI' Marine Biotic Index (AMBI; Borja et al., 2000) was employed to integrate the information on the assemblage structure into the classification of the ecological risk. AMBI takes into account the faunal composition, ascribing each species to five ecological groups based on their tolerance to pollution (Group I: species sensitive to disturbance; EG II: species indifferent to disturbance; Group III: species tolerant to disturbance; Group IV: second order opportunistic species; Group V: first order opportunistic species), summarizing a considerable amount of ecological information into a single representative value (Sigamani et al., 2015). This index was selected because (i) it explores the response of soft-bottom communities to man-induced changes in water and sediment quality by integrating long-term environmental effects, (ii) it is commonly used for environmental assessments, and (iii) it has been formally introduced into the environmental legislation of many European countries for the determination of the ecological quality status within the context of the WFD (Borja & Tunberg, 2011; Forchino et al., 2011; Muxika et al., 2005).

Following the procedure reported in Regoli et al. (2019), the value of AMBI index for each sample was firstly calculated using the AMBI 6.0 software (https://ambi.azti.es/download). Values of the AMBI index (which can vary between 0 and 6) were then rescaled to vary between 0 and 100 % to obtain the hazard quotient for benthic communities (HQ_{BC}) and used for the integration with other LOEs in the final WOE elaboration of ecological risk. Limit for the five risk classes for HQ_{BC} were fixed as follows: < 20 % (*Absent*), 20 % - < 40 % (*Slight*), 40 % - <60 % (*Moderate*), 60 % - <80 % (*Major*), ≥80 % (*Severe*).

2.2.4. WOE ecological risk assessment through the Sediqualsoft® software

The Sediqualsoft® software was used to perform an integrated evaluation of the datasets containing the results of the three LOEs and to obtain a weighted risk. The conceptual elaborations of the WOE model were first described in detail by Piva et al. (2011) and then further expanded by Regoli et al. (2019). The quantitative hazard quotients determined for the individual LOEs were normalized to a common scale (i.e., to range between 0 and 100 %) and weighted differently depending on their ecological relevance.

The following weighting factors were assigned to the different LOEs: 1.0 for LOE1 (chemical analysis); 1.2 for LOE4 (ecotoxicological bioassay); 1.3 for LOE5 (benthic community structure) following Piva et al. (2011) and Regoli et al. (2019). The logic underlying this weighting is to give a relatively higher importance to biological responses, which indicate a potential effect of sediment pollution, with respect to chemical assessments, which in turn only reflect the presence and concentration of pollutants.

The overall hazard quotient integrating the different LOEs (HQ_{WOE}), scaled to range between 0 and 100 %, can be calculated as follows:

$$HQ_{WOE} = \frac{HQ_C + 1.2 \bullet HQ_{Battery} + 1.3 \cdot HQ_{BC}}{HQ_{WOE}^{Max}}$$

where HQ^{Max}_{WOE} is the overall cumulative quotient value obtained when all hazard quotients are equal to 100 %. HQ_{WOE} values were then assigned to one of five risk classes from '*Absent*' to '*Severe*' based on the following ranges: < 20 % (*Absent*), 20 % - < 40 % (*Slight*), 40 % - <60 % (*Moderate*), 60 % - <80 % (*Major*), ≥80 % (*Severe*).

3. Results

3.1. LOE1 – Chemical analysis

Chemical analysis of sediments at the four sites included 58 analytes with a total of 696 analytical results for interpretation and comparison with normative thresholds. The concentration of chemicals fell far below the reference limits, except for some metals (i.e., As and Hg) and 13 out of 16 PAHs (especially in site C), which showed critical values exceeding their respective normative thresholds (Table A3, Appendix A in supplementary material).

The quotient of chemical hazard (HQ_C, Table 1) was classified between '*Absent*' and ''*Negligible*' in the two offshore sites (S and B), while it was more variable at the nearshore sites C and I, ranging between

Table 1
Hazard quotient for LOE1 – chemical characterization (HQ_C) and associated class of hazard integrating the results of chemical analysis (58 substances). Details on concentrations of single substances in sediments from each sampling station were reported in Table A2 (Appendix A in supplementary material).

Site	Station	HQ_C	Class of hazard
C	ST1	57.98	*Severe*
	ST2	6.97	*Major*
	ST3	4.41	*Moderate*
I	ST1	1.7	*Slight*
	ST2	3.43	*Moderate*
	ST3	9.43	*Major*
S	ST1	0.04	*Absent*
	ST2	0.04	*Absent*
	ST3	0.04	*Absent*
B	ST1	0.06	*Absent*
	ST2	0.05	*Absent*
	ST3	1.14	*Negligible*

Slight and *Severe*.

A more critical condition was found at coastal sites. Site C had the highest HQ_C, recording a total of 14 chemicals exceeding the reference limits, with benzo-[a]-pyrene and Hg being the most important pollutants. The severity of chemical hazard increased from station ST3 to ST1, the latter being the station closest to the city of Trieste (Table 1, see also Fig. 1). A gradient of HQ_C, with Hg being the most important pollutant, was also found at site I with severity decreasing at increasing distance from the mouth of the Isonzo/Soča River, ranging from *Slight* at ST1 to *Major* at ST3 (Table 1, see also Fig. 1).

3.2. LOE4 – Ecotoxicological bioassays

A few significant responses for the embryotoxicity bioassays were found, with samples from a single station (i.e., ST1 of site I) showing effects above limits and only for the bioassay on *P. lividus* (% of abnormal larvae) (Table 2). Values of $HQ_{Battery}$ were always lower than 1 (Table 2) and classified the ecotoxicological hazard as *Absent* for all sampling stations.

3.3. LOE5 – Status of benthic assemblages

A total of 205 taxa were found (Table A4, Appendix A in supplementary material), accounting for 2,091 individuals. Most of taxa (74 %) were identified at species level, while in fewer cases at genus (16 %) or higher taxonomic levels (10 %). The AMBI index highlighted 'undisturbed' conditions for most of sampling stations, except for ST1 of site C, ST3 of site I, and ST2-3 of site S, which were classified as 'slightly disturbed' (Table 3). The corresponding hazard quotient (HB_{BC}) of all stations was rated as *Absent* for site B, and *Absent* to *Slight* for stations in sites C, I and S (Table 3).

3.4. Ecological risk from the WOE approach

The level of ecological risk for each station in each site obtained from the WOE integration was summarized in Fig. 2. WOE rated ecological risk as *Slight* for all sampling stations at the nearshore sites C and I, and *Absent* at the offshore sites S and B. Exact values of cumulative HQ_{WOE} for each sampling station were reported in Table A5 (Appendix A in supplementary material).

4. Discussion

We assessed the status of sediments focusing on three different lines of evidence including chemical (LOE1), ecotoxicological (LOE4) and benthic community (LOE5) conditions, which were combined through a Weight-of-Evidence (WOE) approach for an integrated evaluation the ecological risk in soft bottom habitats within protected sites of the EU Natura 2000 network. Overall, a low ecological risk characterized the investigated sites, ranging from *Absent* in offshore sites to *Slight* in sites close to the coast. Such findings are aligned with results of independent assessments of the vulnerability of the seabed to a comprehensive set of anthropogenic threats acting in the study area (e.g., Pagano et al., 2023), and seems to suggest the effectiveness of environmental management strategies implemented in the region. Future applications of the WOE approach to a range of environmental contexts across the Adriatic Sea

Table 3
Hazard quotient for LOE5 – status of benthic assemblages (HQ_{BC}) and associated class of hazard of each sampling station in each site. Values of the AMBI index were also reported.

Site	Station	Value of AMBI	HQ_{BC}	Class of hazard
C	ST1	1.43	0.24	*Slight*
	ST2	1.12	0.19	*Absent*
	ST3	0.65	0.11	*Absent*
I	ST1	0.81	0.14	*Absent*
	ST2	1.19	0.19	*Absent*
	ST3	1.29	0.21	*Slight*
S	ST1	0.97	0.16	*Absent*
	ST2	1.39	0.23	*Slight*
	ST3	1.22	0.20	*Slight*
B	ST1	0.63	0.10	*Absent*
	ST2	0.77	0.13	*Absent*
	ST3	1.13	0.19	*Absent*

Table 2
Hazard quotient for LOE4 – ecotoxicological bioassays ($HQ_{Battery}$) and associated class of hazard integrating the results of ecotoxicological bioassays on sediments per each sampling station in each site. Results of bioassays on *P. tricornutum* (growth inhibition), *P. lividus* (% of abnormal larvae), and *M. insidiosum* (mortality) were also reported. Bioassays recording ecotoxicological response above legal limits were given in bold.

		P. tricornutum Growth inhibition(%) 72 h		*P. lividus* Abnormal larvae(%) 72 h			*M. insidiosum* Mortality(%) 10 days			$HQ_{Battery}$	Class of hazard
Site	Station	Mean	Standard	Mean	Stand. Dev.	Adj. mean	Mean	Stand. Dev.ard	Adj. mean		
C	ST1	−11.66	7.64	8.00	2.00	0.00	8.33	7.64	6.78	0.58	*Absent*
	ST2	−8.80	1.71	7.67	0.58	0.00	10.00	5.00	8.47	0.65	*Absent*
	ST3	2.18	3.84	9.67	3.51	0.00	3.33	5.77	1.69	0.28	*Absent*
I	ST1	−12.18	3.71	25.33	1.53	**17.04**	11.67	2.89	10.17	0.72	*Absent*
	ST2	−7.66	5.34	7.33	0.58	0.00	8.33	7.64	6.78	0.27	*Absent*
	ST3	−2.74	3.46	9.33	2.08	0.00	1.67	2.89	0.00	0.00	*Absent*
S	ST1	−5.85	1.28	11.67	2.08	1.85	6.67	2.89	1.75	0.39	*Absent*
	ST2	−4.37	0.00	14.00	4.58	4.44	10.00	5.00	5.26	0.54	*Absent*
	ST3	−2.69	3.09	11.67	2.08	1.85	10.00	0.00	5.26	0.53	*Absent*
B	ST1	−3.58	0.68	13.67	4.04	4.07	6.67	2.89	1.75	0.09	*Absent*
	ST2	−1.46	3.00	15.67	2.31	6.30	10.00	0.00	5.26	0.29	*Absent*
	ST3	−6.21	1.24	14.33	3.51	4.81	6.67	2.89	1.75	0.10	*Absent*

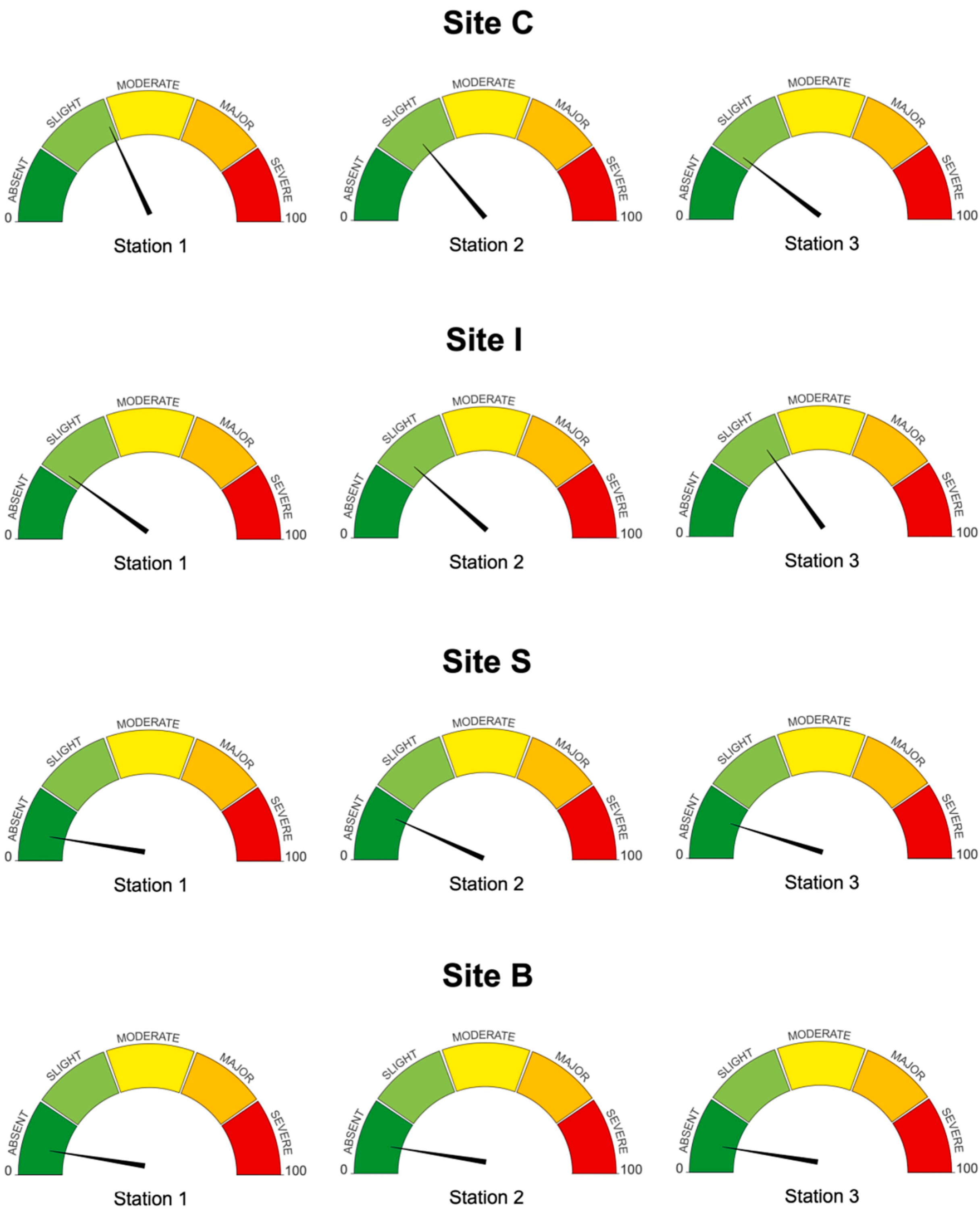

Fig. 2. Cumulative ecological risk (HQ_{WOE}) for each station at the 4 investigated sites. The HQ_{WOE} was the result of the WOE integration of chemical hazard, ecotoxicological effects, and the status of benthic assemblages. The hands of risk-meters are indicative of the true levels of risk on a 0–100% scale.

and in other basins may help elucidate the sensitiveness of this tool in assessing the ecological risk within protected sites at varying cumulative human pressure and management scenarios.

The Gulf of Trieste, nonetheless, remains one of the most impacted areas of the Adriatic Sea (Covelli et al., 2001; Cibic et al., 2017; Pagano et al., 2023) and the level of sediment pollution recommends a careful consideration of the overall ecological risk estimated by the use of integrative approaches like WOE with respect to specific issues in monitoring and managing protected sites. For example, in coastal sites, some station (e.g., ST1 of site C) exhibited impressive level of contamination for single priority substances (e.g., benzo[a]pyrene), raising concerns on chemical hazard and stressing the need of close monitoring notwithstanding the overall risk from WOE was *Slight* in these stations. A further note of caution is also required as our assessment is constrained to a single campaign, thus limiting the analysis of potential temporal variations in environmental and biological conditions and the associated ecological risk. However, the WOE approach allowed to identify specific hazards and critical issues while providing a synoptic indication of the overall ecological risk in marine protected sites, highlighting the potential of this approach as a promising tool for their management.

4.1. Chemical hazard and historical sources of pollution in the study area

Site C was characterized by the highest levels of chemical hazard, spatially distributed along a gradient of decreasing hazard from ST1, the station closest to the City of Trieste, towards ST3. The high chemical hazard at ST1 and ST2 was mostly due to a high contamination of benzo [a]pyrene (BaP). Actually, the mean BaP concentrations in these two stations were 72 (ST1) and 10 (ST2) times higher than the reference limit set by DM 173/2016 for marine sediments (i.e., 30 μg kg^{-1} dry weight). BaP is a five-ring compound formed by incomplete combustion at temperatures between 300 °C and 600 °C, and generally found with other polycyclic aromatic hydrocarbons (PAHs) as a by-product of a wide range of processes, from food cooking (Park et al., 2017) to industrial production (Appel et al., 1990; Khesina, 1994). In the marine environment, PAHs mainly derive from oil spills, atmospheric deposition, and land-based pollution from terrestrial runoff (especially in urbanized coastal areas where high PAHs concentrations may occur; Stout et al., 2004; Shi et al., 2022). PAHs are lipophilic substances with low water solubility and, once deposited in water or sediments, they are strongly adsorbed to sediments and organic particles and slowly degraded over many years (US EPA, 2017). Since high molecular weight PAHs (4 rings or more) are mainly produced by fossil fuel combustion at high temperatures (Shi et al., 2022), the observed gradient of BaP in marine sediments of site C was probably caused by the combustion of fossil fuels at high temperatures associated with several industrial activities, including production of coking coal and cast iron, and several steel furnaces, which operated in the harbor area of Trieste from 1896 until 2020.

A second gradient of chemical hazard mostly due to Hg contamination was detected at site I, ranging from a *Slight* risk for ST1 to a *Major* risk for ST3, respectively the farthest and the closest sampling station to the mouth of the Isonzo/Soča River. In this case, a long history of inland cinnabar mining was mostly responsible of Hg contamination in coastal sediments (Covelli et al., 2001; Pavoni et al., 2020). Since the 16th century, more than 5×10^6 metric tons of Hg were extracted before mining activities definitively ceased in 1995. After smelting, almost ¼ of the total amount of extracted Hg has been dispersed in the environment, reaching the Isonzo/Soča River basin and ultimately ending up in the Gulf of Trieste (Covelli et al., 2001). A complex interplay among current circulation, meteomarine and riverine hydrological conditions acting in the Gulf forces river plumes and sediment accumulation to concentrate along the coast and especially near the river mouth (Malacic, 1991; Covelli et al., 2007), thus explaining the observed gradient of Hg contamination. Regional-scale sedimentary dynamics also prevent the accretion of thick sediment layers in the central portion of the Gulf (Trobec et al., 2018). This reduced transport of sediments from coastal areas, which are more exposed to land-based contamination, towards offshore areas could underlay the low contamination levels recorded at sites S and B.

4.2. Insights from ecotoxicological bioassays

The battery of ecotoxicological bioassays showed good agreement among tests on distinct target organisms that differ in ecology, sensitivity, and measured endpoints. The weighted ecotoxicological hazard was *Absent* for all sediment samples from the four sites, in contrast to results of chemical analysis which highlighted a range of chemical hazard levels up to *Severe* risk in coastal sites (i.e., sites C and I), and mostly related to BaP and Hg contamination.

BaP, and more generally all PAHs, can be heavily adsorbed onto and absorbed by particulate matter and transferred to benthic habitats, thereby increasing the concentration of these substances in surface sediments, which may represent the preferential destination for accumulation of PAHs (Shi et al., 2022). The hydrophobic nature of PAHs confers to these substances a higher affinity for the solid phase of sediments rather than for the aqueous medium. It was not surprising, therefore, that the ecotoxicological bioassays based on elutriates (i.e., tests on *P. lividus* and *P. tricornutum*), testing the aqueous extracts of sediments for toxicity of the water-soluble fraction of pollutants, did not detected significant effects in sampled sediments, even for those from coastal sites where chemical analysis recorded very high concentrations of BaP. However, significant effects were also absent for all stations when testing for toxicity of the solid phase of sediments on the mortality of the amphipod *M. insidiosum*, which instead focused on the potential toxicity of the fraction of pollutants that tends to remain bound to sediment particles due to their chemical properties, solubility, adsorption, and degree of complexation with the organic matter (ISPRA, 2011). Ecotoxicological effects of pollutants largely depends on their bioavailability, which reflects the amount of pollutants present in the environment that can actually interact with organisms and cause biological responses. Larger (high molecular weight) PAHs with high K_{ow} coefficient (i.e., the n-octanol–water partitioning coefficient, which indicate the partitioning behavior of chemicals from aqueous media to organic matrices) such as BaP ($K_{ow} = 6.04$), would less easily partition into seawater from sediments and be less bioavailable with respect to smaller PAHs, like for example phenanthrene ($K_{ow} = 4.57$), resulting in reduced toxicity (Hellou et al., 2014). In addition, large PAHs may be preferentially associated to specific types of particles (in term of composition and grain) that may not represent the preferential food of the test species (e.g., *M. insidiosum*, *Corophium volutator*, and other amphipods), leading to decreased ingestion rates of specific PAHs and, consequently, to limited biological responses (Hellou et al., 2014).

Bioavailability is also critical to understand the potential effects of Hg, one of the most important pollutants in the study area. An important aspect affecting the bioavailability and mobility of Hg in sediments is its speciation (Acquavita et al., 2021; Cinnirella et al., 2019). In the aquatic environment, the main form of Hg is the inorganic Hg(II) though, from an ecotoxicological point of view, the most important species is MeHg, as it is highly neurotoxic and nephrotoxic and bioaccumulates and biomagnifies throughout the food web (Alava et al., 2017; Hsu-Kim et al., 2013; Zheng et al., 2019). MeHg is formed in sediments only under particular abiotic (anoxia, presence of methyl donors such as methylcobalamin, humic substances, methyltin) and biotic (presence of sulphate-reducing bacteria) conditions (Acquavita et al., 2021; Hsu-Kim et al., 2013). In our assessment, the total Hg was measured irrespective of its chemical speciation, making difficult an exhaustive explanation of the lack of toxic effects observed in ecotoxicological bioassays, and especially for sediments from coastal sites (i.e., site I and C) where the highest concentration of total Hg was recorded. Concentration of MeHg in coastal sediments of the study area can largely vary both among and within seasons, with the largest intra-seasonal variation occurring in autumn–winter (Bratkič et al., 2017). As sampling was carried out in these seasons, and only once in each station, it could have incidentally occurred during a period of low concentrations of MeHg in surface sediments. Also, the Hg demethylation rates in the Gulf generally increase from offshore towards shallow areas (Hines et al., 2017), and may have counterbalanced the availability of bioactive Hg species in nearshore sediments despite the overall high concentrations of total Hg.

4.3. The importance of integrating the response of benthic assemblages in risk assessment

A complex, and not yet completely understood, interplay among physical–chemical, biological and anthropogenic factors may influence the amount of pollutants in marine sediments, their bioavailability, and their effects on benthic organisms (Heggleton and Thomas, 2004; Hellou et al., 2014; Lindgren et al., 2014; Wang et al., 2021). Hence, the low ecological hazard reported by LOE4 does not preclude the possibility that this condition could worsen in the future, or could have been worse in the past, given the high chemical hazard highlighted by LOE1 in some sites. Pollutants can be released from sediments to the water column through pore diffusion (Heggleton and Thomas, 2004; Ramalhosa et al., 2006), anthropogenic physical remobilization of sediments (e.g., from dredging, trawling or turbo-charging) (Palanques et al., 2022), natural erosion amplified during intense flood events and alterations of currents and sedimentary regimes as a consequence of climate change (Alava et al., 2017), or merely increase in concentration due to increasing contamination from human activities. In this view, if monitoring LOE1 may inform on chemical hazard arising from changes in sediment contamination, the role of monitoring LOE4 is crucial to understand whether such changes may turn into a varying risk of harmful biological effects on the marine biota.

The level of biological hazard detectable through ecotoxicological bioassays, nevertheless, may respond to transient episodes of increased or decreased contamination or bioavailability of pollutants, their small-scale patchiness, or may be affected by the frequency of sampling (Heggleton and Thomas, 2004). In this respect, evidence from LOE4 in our assessment is constrained to single snapshot of ecotoxicological hazard, as sampling occurred only once per station during the study period. The additional evidence from LOE5, through the assessment of the ecological status of macrobenthic assemblages, could help compensate these limits. Marine macroinvertebrate assemblages from soft sediments are mostly composed by sedentary species, with varying sensitivity to disturbance and different ecological traits, so that their overall structure reflects the combined response to multiple drivers of change at a range of spatial and temporal scales (Pearson and Rosenberg, 1978; Borja et al., 2000; Bessa et al., 2014, Dauvin, 2018). Monitoring macrobenthos could, therefore, complement evidence from LOE1 and LOE4 by providing an integrated biological response of benthic assemblages to long-term regimes of environmental conditions in sediments and enlightening the potential for community-wide effects of sediment contamination. Moreover, changes in macrobenthic assemblage structure may underline the effects of human-driven pressures other than pollution (e.g., physical disturbance, alteration of sedimentation rates), which can remain unnoticed when assessments just rely on chemical and ecotoxicological analyses.

We found a general low hazard based on the status of benthic assemblages classified though the AMBI index. Only at a single station of site C (ST1) and site I (ST3), and at two stations of site S (ST2-3), the hazard quotient from LOE5 rated the hazard as *Slight*, whereas the hazard was *Absent* for all the remaining stations. The slightly disturbed condition of benthic assemblages recorded at ST1 of site C and ST3 of site I reflected the evidence from LOE1 on chemical hazard, underlining the sensitiveness of benthic assemblages to historical contamination. Beyond potential mechanisms of adaptation to chronic pollution (Klerks and Weis, 1987; Medina et al., 2007), the high concentration of some pollutants (e.g., Hg and PAHs) found in sediments at these sites has probably led to the selection more tolerant species which was detected by the AMBI index, despite no significant ecotoxicological effects arose from ecotoxicological bioassays (LOE4). Interestingly, the *Slight* level of hazard assigned by LOE5 to stations ST2-ST3 of site S was not associated with similar or higher hazard levels in the other LOEs. However, if soft bottoms in this offshore site were less influenced by land-based pollution, which explain the low levels of chemical and ecotoxicological hazard, they could be affected by other sources of environmental stress. For example, the central part of the Gulf of Trieste, were site S is located, is subjected to medium–high trawling pressure exerted especially by small otter and beam trawls (Eigaard et al., 2017; Russo et al., 2020). Although fishing activities are banned within a large portion of the site, their effects (e.g., increased sedimentation, resuspension, abrasion) could have propagated to close neighboring protected zones causing the slight, but detectable, changes in the structure of macrobenthic assemblages highlighted by LOE5.

4.4. The WOE approach for environmental management of marine protected sites

Assessing the effectiveness of MPAs has so far focused on quantifying the outcomes of protection in enhancing the diversity, size of individuals and abundance of populations of specific groups of organisms, mostly fish and some invertebrates of commercial interest (Sciberras et al., 2013), often overlooking the physical–chemical conditions of benthic habitats and the associated assemblages (Abessa et al., 2018; Chen, 2021). Yet, a more comprehensive evaluation of the status of the benthic compartment as a whole, including its physical, chemical and biological integrity, may be crucial to improve conservation strategies (Moreira et al., 2021). This could be even more important for other area-based marine conservation measures, such as marine SPAs/SACs, which are specifically tailored to protect not only target species but also their habitats. Indeed, for most of marine SCIs of the Natura 2000 network, the information on conservation status of protected habitats and species is fragmented or based on expert opinion, if not completely lacking (Mazaris et al., 2018; Gianni et al., 2022). In such contexts, the WOE approach could help capitalize the existing information and the integration of environmental data from independent sources (e.g., environmental agencies, management bodies) for a more holistic approach to the characterization of the ecological status of protected sites.

A major strength of the WOE procedure is that it allows a synoptic classification of the ecological status, overcoming the difficulty of interpreting the individual LOEs. At the same time, the approach informs on the contribution of each single LOE to the composition of the overall ecological risk, providing indications of specific threats. Integrating the level of risk from the different LOEs into an overall weighted value ranked into five simple classes, moreover, makes easily understandable the outcomes of complex environmental analyses improving the flow of information from practitioners/researchers to stakeholders and environmental managers.

A further advantage of the WOE approach is its modularity and adaptability to different environmental and/or legislative settings. Currently, the Sediqualsoft® software encompasses several thresholds, indicators, methodological standards, and substances formally recognized by the EU environmental regulations (e.g., WFD, MSFD), whereas in some cases limits for pollutants are fixed based on the Italian law; all parameters can be nonetheless updated or changed to adapt to different jurisdictions. Also, additional LOEs can be integrated depending on data availability and/or study-specific needs, or can be developed to integrate new data type (e.g., transcriptomics; Cecchetto et al., 2023a,b; Maradonna et al., 2020) or other environmental compartments (e.g., water column; Li et al., 2023).

The WOE approach based on Sediqualsoft® was initially proposed as a tool to assess the environmental and biological risk associated to the management (e.g., movement, treatment, or destination) of marine sediments in highly polluted areas (Piva et al., 2011). Analogously, we provided an application of the approach to soft bottom habitats, but virtually applicable to other benthic habitats, within marine protected sites to put its logic into a conservation perspective. Effective conservation strategies require a careful consideration of potential hazards associated to a range of increasing anthropogenic pressures on marine ecosystems that could easily extend within the boundaries of protected sites and risk to vanish conservation efforts. This can be particularly the case of coastal ecosystems in densely populated geographic areas, where conservation priorities may often collide with the increasing exploitation of resources and uses of marine space (Guarnieri et al., 2016). In this view, environmental monitoring through the WOE approach can be used as a tool to assess the risk of undesirable changes in protected ecosystems given the status of their different abiotic and biotic components, to outline trade-offs among human threats and conservation needs, to identify early-warning signals of hazard, or even be used for preliminary assessments of the risk of failure of conservation initiatives in relation to the cumulative level of ecological hazard (Game et al., 2008), providing guide for managing the existing protected sites and supporting decision-making for the implementation of conservation networks.

5. Ethics statement

Ecotoxicological assays were carried out on target species according to international standardized protocols and do not require authorization or approval by ethic committees.

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CRediT authorship contribution statement

Manuela Piccardo: Writing – review & editing, Writing – original draft, Investigation, Formal analysis, Conceptualization. **Verdiana Vellani:** Writing – review & editing, Investigation. **Serena Anselmi:** Writing – review & editing, Investigation. **Eleonora Grazioli:** Writing – review & editing, Investigation. **Monia Renzi:** Writing – review & editing, Resources, Conceptualization. **Antonio Terlizzi:** Conceptualization, Resources, Writing – review & editing. **Lucia Pittura:** Formal analysis. **Giuseppe D'Errico:** Formal analysis. **Francesco Regoli:** Formal analysis. **Stanislao Bevilacqua:** Writing – review & editing, Writing – original draft, Supervision, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

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References

Abessa, D. M. S., Albuquerque, H. C., Morais, L. G., Araújo, G. S., Fonseca, T. G., Cruz, A. C. F., Campos, B. G., Camargo, J. B. D. A., Gusso-Choueri, P. K., Perina, F. C., Choueri, R. B., Moreira, B. L., 2018. Pollution status of marine protected areas worldwide and the consequent toxic effects are unknown. Environ. Pollut. 243, Part B, 1450–1459. https://doi.org/10.1016/j.envpol.2018.09.129.

Acquavita, A., Floreani, F., Covelli, S., 2021. Occurrence and speciation of arsenic and mercury in alluvial and coastal sediments. Curr. Opin. Environ. Sci. Health 22, 100272. https://doi.org/10.1016/j.coesh.2021.100272.

Alava, J. J., Cheung, W. W. L., Ross, P. S., Sumaila, U. R., 2017. Climate change–contaminant interactions in marine food webs: toward a conceptual framework. Glob. Change Biol. 23, 3984–4001. https://doi.org/ 0.1111/gcb.13667.

Appel, B.R., Guirguis, G., Kim, I.-S., Garbin, O., Fracchia, M., Flessel, P., Kizer, K.W., Book, S.A., Warriner, T.E., 1990. Benzene, benzo(a)pyrene, and lead in smoke from tobacco products other than cigarettes. Am. J. Public Health 80, 560–564. https://doi.org/10.2105/ajph.80.5.560.

Barbier, E. B., 2017. The Economics of Aquatic Ecosystems: An Introduction to the Special Issue. Water Econ. Policy 3, 1702002. https://doi.org/10.1142/S2382624X17020027.

Benedetti, M., Ciaprini, F., Piva, F., Onorati, F., Fattorini, D., Notti, A., Ausili, A., Regoli, F., 2012. A multidisciplinary weight of evidence approach for classifying polluted sediments: Integrating sediment chemistry, bioavailability, biomarkers responses and bioassays. Environ. Int. 38, 17–28. https://doi.org/10.1016/j.envint.2011.08.003.

Benedetti, M., Gorbi, S., Fattorini, D., d'Errico, G., Piva, F., Pacitti, D., Regoli, F., 2014. Environmental hazards from natural hydrocarbons seepage: Integrated classification of risk from sediment chemistry, bioavailability and biomarkers responses in sentinel species. Environ. Pollut. 185, 116–126. https://doi.org/10.1016/j.envpol.2013.10.023.

Bevilacqua, S., Guarnieri, G., Farella, G., Terlizzi, A., Fraschetti, F., 2018. A regional assessment of cumulative impact mapping on Mediterranean coralligenous outcrops. Sci. Rep. 8, 1757. https://doi.org/10.1038/s41598-018-20297-1.

Borja, A., Elliott, M., Snelgrove, P. V. R., Wilson, C., 2016. Bridging the Gap between Policy and Science in Assessing the Health Status of Marine Ecosystems. Front. Mar. Sci. 3, 175. https://doi.org/doi: 10.3389/fmars.2016.00175.

Borja, A., Menchaca, I., Garmendia, J.M., Franco, J., Larreta, J., Sagarminaga, Y., Schembri, Y., González, R., Antón, R., Micallef, T., Camilleri, S., Solaun, O., Uriarte, A., Uyarra, M.C., 2021. Big insights from a small country: the added value of integrated assessment in the marine environmental status evaluation of malta. Front. Mar. Sci. 8, 638232 https://doi.org/10.3389/fmars.2021.638232.

Borja, A., Tunberg, B.G., 2011. Assessing benthic health in stressed subtropical estuaries, eastern Florida, USA using AMBI and M-AMBI. Ecol. Indic. 11, 295–303. https://doi.org/10.1016/j.ecolind.2010.05.007.

Borja, A., Franco, J., Perez, V., 2000. A marine biotic index to establish the ecological quality of soft-bottom benthos within european estuarine and coastal environments. Mar. Pollut. Bull. 40, 1100–1114. https://doi.org/10.1016/S0025-326X(00)00061-8.

Borja, A., White, M.P., Berdalet, E., Bock, N., Eatock, C., Kristensen, P., Leonard, A., Lloret, J., Pahl, S., Parga, M., Prieto, J.V., Wuijts, S., Fleming, L.E., 2020. Moving toward an agenda on ocean health and human health in Europe. Front. Mar. Sci. 7, 37. https://doi.org/10.3389/fmars.2020.00037.

Bratkič, A., Koron, N., Ribeiro Guevara, S., Faganeli, J., Horvat, M., 2017. Seasonal variation of mercury methylation potential in pristine coastal marine sediment from the gulf of trieste (Northern Adriatic Sea). Geomicrobiol. J. 34, 587–595. https://doi.org/10.1080/01490451.2016.1247482.

Broccoli, A., Morroni, L., Valentini, A., Vitiello, V., Renzi, M., Nuccio, C., Pellegrini, D., 2021. Comparison of different ecotoxicological batteries with WOE approach for the environmental quality evaluation of harbour sediments. Aquat. Toxicol. 237, 105905 https://doi.org/10.1016/j.aquatox.2021.105905.

Bryhn, A., Kraufvelin, P., Bergstrom, U., Vretborn, M., Bergstrom, L., 2020. A model for disentangling dependencies and impacts among human activities and marine ecosystem services. Environ. Manage. 65, 575–586. https://doi.org/10.1007/s00267-020-01260-1.

Carere, M., Dulio, V., Hanke, G., Polesello, S., 2012. Guidance for sediment and biota monitoring under the Common Implementation Strategy for the Water Framework Directive. TrAC 36, 15–24. https://doi.org/10.1016/j.trac.2012.03.005.

Cecchetto, M., Giubilato, E., Bernardini, I., Bettiol, C., Asnicar, D., Bertolini, C., Fabrello, J., Bonetto, A., Peruzza, L., Ciscato, M., Matozzo, V., Marin, M.G., Bargelloni, L., Patarnello, T., Marcomini, A., Milan, M., Semenzin, E., 2023a. A Weight of Evidence approach to support the assessment of the quality of Manila clam farming sites in a coastal lagoon. Mar. Pollut. Bull. 197, 115668 https://doi.org/10.1016/j.marpolbul.2023.115668.

Cecchetto, M., Peruzza, L., Giubilato, E., Bernardini, I., Dalla Rovere, G., Marcomini, A., Regoli, F., Bargelloni, L., Patarnello, T., Semenzin, E., Milan, M., 2023b. An innovative index to incorporate transcriptomic data into weight of evidence approaches for environmental risk assessment. Environ. Res. 227, 115745 https://doi.org/10.1016/j.envres.2023.115745.

Chen, E.-Y.-S., 2021. Often overlooked: understanding and meeting the current challenges of marine invertebrate conservation. Front. Mar. Sci. 8, 690704 https://doi.org/10.3389/fmars.2021.690704.

Cibic, T., Franzo, A., Nasi, F., Auriemma, R., Del Negro, P., 2017. The Port of Trieste (Northern Adriatic Sea) – a case study of the "ecosystem approach to management". Front. Mar. Sci. 4, 336. https://doi.org/10.3389/fmars.2017.00336.

Cinnirella, S., Bruno, D.E., Pirrone, N., Horvat, M., Živković, I., Evers, D.C., Johnson, S., Sunderland, E.M., 2019. Mercury concentrations in biota in the Mediterranean Sea, a compilation of 40 years of surveys. Sci. Data 6, 205. https://doi.org/10.1038/s41597-019-0219-y.

Covelli, S., Faganelli, J., Horvat, M., Brambati, A., 2001. Mercury contamination of coastal sediments as the result of long-term cinnabar mining activity (Gulf of Trieste, northern Adriatic Sea). J. Appl. Geochem. 16, 541–558. https://doi.org/10.1016/S0883-2927(00)00042-1.

Covelli, S., Piani, R., Acquaviva, A., Predonzani, S., Faganelli, J., 2007. Transport and dispersion of particulate Hg associated with a river plume in coastal Northern Adriatic environments. Mar. Pollut. Bull. 55, 436–450. https://doi.org/10.1016/j.marpolbul.2007.09.006.

d'Errico, G., Nardi, A., Benedetti, M., Mezzelani, M., Fattorini, D., Di Carlo, M., Pittura, L., Giuliani, M.E., Macchia, S., Vitiello, V., Sartori, D., Scuderi, A., Morroni, L., Chiaretti, G., Gorbi, S., Pellegrini, D., Regoli, F., 2021. Application of a multidisciplinary weight of evidence approach as a tool for monitoring the ecological risk of dredging activities. Front. Mar. Sci. 8, 765256 https://doi.org/10.3389/fmars.2021.765256.

Dauvin, J.-C., 2018. Twenty years of application of Polychaete/Amphipod ratios to assess diverse human pressures in estuarine and coastal marine environments: A review. Ecol. Indic. 95, 427–435. https://doi.org/10.1016/j.ecolind.2018.07.049.

DM 173/2016. Regolamento recante modalità e criteri tecnici per l'autorizzazione all'immersione in mare dei materiali di escavo di fondali marini. Decreto del Ministero dell'Ambiente e della Tutela del Territorio e del Mare no. 173, July 2016.

EC, 2000. Directive 2000/60/EC of the European Parliament and of the Council establishing a framework for community action in the field of water policy.

EC, 2008. Directive 2008/56/EC of the European Parliament and of the Council Establishing a Framework for Community Action in the Field of Marine Environmental Policy.

EC, 2013. Directive 2013/39/EU of the European Parliament and of the Council amending Directives 2000/60/EC and 2008/105/EC as regards priority substances in the field of water policy.

Eigaard, O.R., Bastardie, F., Hintzen, N.T., Buhl-Mortensen, L., Buhl-Mortensen, P., Catarino, R., Dinesen, G.E., Egekvist, J., Fock, H.O., Geitner, K., Gerritsen, H.D., González, M.M., Jonsson, P., Kavadas, S., Laffargue, P., Lundy, M., Gonzalez-Mirelis, G., Nielsen, J.R., Papadopoulou, N., Posen, P.E., Pulcinella, J., Russo, T., Sala, A., Silva, C., Smith, C.J., Vanelslander, B., Rijnsdorp, A.D., 2017. The footprint of bottom trawling in European waters: distribution, intensity, and seabed integrity. ICES J. Mar. Sci. 74, 847–865. https://doi.org/10.1093/icesjms/fsw194.

Forchino, A., Borja, A., Brambilla, F., Rodriguez, J.G., Muxika, I., Terova, G., Saroglia, M., 2011. Evaluating the influence of off-shore cage aquaculture on the benthic ecosystem in Alghero Bay (Sardinia, Italy) using AMBI and M-AMBI. Ecol. Indic. 11, 1112–1122. https://doi.org/10.1016/j.ecolind.2010.12.011.

Fraschetti, S., Fabbrizzi, E., Tamburello, L., Uyarra, M.C., Micheli, F., Sala, E., Pipitone, C., Badalamenti, F., Bevilacqua, S., Boada, J., Cebrian, E., Ceccherelli, G., Chiantore, M., D'Anna, G., Di Franco, A., Farina, S., Giakoumi, S., Gissi, E., Guala, I., Guidetti, P., Katsanevakis, S., Manea, E., Montefalcone, M., Sini, M., Asnaghi, V., Calò, A., Di Lorenzo, M., Garrabou, J., Musco, L., Oprandi, A., Rilov, G., Borja, A., 2022. An integrated assessment of the Good Environmental Status of Mediterranean Marine Protected Areas. J. Environ. Manage. 305, 114370 https://doi.org/10.1016/j.jenvman.2021.114370.

Furlan, E., Torresan, S., Critto, A., Lovato, T., Solidoro, C., Lazzari, P., Marcomini, A., 2019. Cumulative impact index for the Adriatic Sea: accounting for interactions among climate and anthropogenic pressures. Sci. Total Environ. 670, 379–397. https://doi.org/10.1016/j.scitotenv.2019.03.021.

Game, E.T., Mcdonald-Madden, E., Puotinen, M.L., Possingham, H.P., 2008. Should we protect the strong or the weak? risk, resilience, and the selection of marine protected areas. Conserv. Biol. 22, 1619–1629. https://doi.org/10.1111/j.1523-1739.2008.01037.x.

Giani, M., Djakovac, T., Degobbis, D., Cozzi, S., Solidoro, C., Fonda Umani, S., 2012. Recent changes in the marine ecosystems of the northern Adriatic Sea. Estuar. Coast. Shelf Sci. 115, 1–13. https://doi.org/10.1016/j.ecss.2012.08.023.

Gianni, F., Manea, E., Cataletto, B., Pugnetti, A., Bergami, C., Bongiorni, L., Pleslić, G., Vilibić, I., Bandelj, V., 2022. Are we overlooking Natura 2000 sites? Lessons learned from a transnational project in the Adriatic Sea. Front. Mar. Sci. 9, 1070373. https://doi.org/10.3389/fmars.2022.1070373.

Gissi, E., Manea, E., Mazaris, A.D., Fraschetti, S., Almpanidou, V., Bevilacqua, S., Coll, M., Guarnieri, G., Lloret-Lloret, E., Pascual, M., Petza, D., Rilov, G., Schonwald, M., Stelzenmüller, V., Katsanevakis, S., 2021. A review of the combined effects of climate change and other human stressors on the marine environment. Sci. Tot. Environ. 755, 142564 https://doi.org/10.1016/j.scitotenv.2020.142564.

Gobert, S., Sartoretto, S., Rico-Raimondino, V., Andral, B., Chery, A., Lejeune, P., Boissery, P., 2009. Assessment of the ecological status of Mediterranean French coastal waters as required by the Water framework directive using the *Posidonia oceanica* rapid easy index: PREI. Mar. Pollut. Bull. 58, 1727–1733. https://doi.org/10.1016/j.marpolbul.2009.06.012.

Guarnieri, G., Bevilacqua, S., De Leo, F., Farella, G., Maffia, A., Terlizzi, A., Fraschetti, S., 2016. The challenge of planning conservation strategies in threatened seascapes: understanding the role of fine scale assessments of community response to cumulative human pressures. PLoS One 11, e0149253.

Halpern, B.S., Frazier, M., Afflerbach, J., Lowndes, J.S., Micheli, F., O'Hara, C., Scarborough, C., Selkoe, K.A., 2019. Recent pace of change in human impact on the world's ocean. Sci. Rep. 9, 11609. https://doi.org/10.1038/s41598-019-47201-9.

Heggleton, J., Thomas, K.V., 2004. A review of factors affecting the release and bioavailability of contaminants during sediment disturbance events. Environ. Int. 30, 973–980. https://doi.org/10.1016/j.envint.2004.03.001.

Hellou, J., Johnston, D., Cheeseman, K., Gronlund, A., Desnoyers, E., Leonard, J., Robertson, S., 2014. Bioavailability and bioaccumulation of PAHs in amphipods exposed to reference and harbor sediments. Pacs 29, 12–27. https://doi.org/10.1080/10406630802686322.

Hines, M.E., Covelli, S., Faganeli, J., Horvat, M., 2017. Controls on microbial mercury transformations in contaminated sediments downstream of the Idrija mercury mine (West Slovenia) to the Gulf of Trieste (northern Adriatic). J. Soils Sediments 17, 1961–1971. https://doi.org/10.1007/s11368-016-1616-x.

Hsu-Kim, H., Kucharzyk, K.H., Zhang, T., Deshusses, M.A., 2013. Mechanisms regulating mercury bioavailability for methylating microorganisms in the aquatic environment: a critical review. Environ. Sci. Technol. 47, 2441–2456. https://doi.org/10.1021/es304370g.

ISPRA, 2011. Batterie di saggi ecotossicologic per sedimenti di acquesalate e salmastre [Manuali e Linee Guida 67/2011].

ISPRA-SNPA, 2017. Saggio di fecondazione e saggio di sviluppo embrionale con il riccio di mare Paracentrotus lividus (Lamarck) (echinodermata: Echinoidea). Quaderni Di Ecotossicologia 11.

Kazanidis, G., Orejas, C., Borja, A., Kenchington, E., Henry, L.-A., Callery, O., Carreiro-Silva, M., Egilsdottir, H., Giacomello, E., Grehan, A., Menot, L., Morato, T., Ragnarsson, S.A., Rueda, J.L., Stirling, D., Stratmann, T., van Oevelen, D., Palialexis, A., Johnson, D., Roberts, J.M., 2016. Assessing the environmental status of selected North Atlantic deep-sea ecosystems. Ecol. Indic. 119, 106624 https://doi.org/10.1016/j.ecolind.2020.106624.

Khesina, A., 1994. Urban air pollution by carcinogenic and genotoxic polyaromatic hydrocarbons in the former USSR. EHP 102, 49–53. https://doi.org/10.2307/3431930.

Klerks, P.L., Weis, J.S., 1987. Genetic adaptation to heavy metals in aquatic organisms: A review. Environ. Pollut. 45, 173–205. https://doi.org/10.1016/0269-7491(87)90057-1.

Korpinen, S., Andersen, J. H., 2016. A global review of cumulative pressure and impact assessments in marine environments. Front. Mar. Sci. 3, 153. https://doi.org/doi:10.3389/fmars.2016.00153.

Korpinen, S., Laamanen, L., Bergström, L., Nurmi, M., Andersen, J.H., Haapaniemi, J., Harvey, E.T., Murray, C.J., Peterlin, M., Kallenbach, E., Klančnik, K., Stein, U., Tunesi, L., Vaughan, D., Reker, J., 2021. Combined effects of human pressures on Europe's marine ecosystems. Ambio. Glob Chang Biol. 50, 1325–1336. https://doi.org/10.1111/gcb.16312.

Leslie, H.M., McLeod, K.L., 2007. Confronting the challenges of implementing marine ecosystem-based management. Front. Ecol. Environ. 5, 540–548. https://doi.org/10.1890/1540-9295(2007)5[540:CTCOIM]2.0.CO;2.

Li, Z., Qi, R., Miao, J., Li, Y., Wang, Q., Lei, F., Wu, M., Chen, Y., Pan, L., 2023. A comprehensive assessment framework for the risk and source of polycyclic aromatic hydrocarbons (PAHs) using two weight of evidence (WOE) approaches in bays of Shandong Province, China. *J. Clean. Prod.* 429, 139572 https://doi.org/10.1016/j.jclepro.2023.139572.

Lindgren, J.F., Hassellöv, I.-M., Dahllöf, I., 2014. PAH effects on meio- and microbial benthic communities strongly depend on bioavailability. Aquat. Toxicol. 146, 230–238. https://doi.org/10.1016/j.aquatox.2013.11.013.

Lotze, H.K., Lenihan, H.S., Bourque, B.J., Bradbury, R.H., Cooke, R.G., Kay, M.C., Kidwell, S.M., Kirby, M.X., Peterson, C.H., Jackson, J.B., 2006. Depletion, degradation, and recovery potential of estuaries and coastal seas. Science 312, 1806–1809. https://doi.org/10.1126/science.1128035.

Malacic, V., 1991. Estimation of the vertical eddy diffusion coefficient of heat in the Gulf of Trieste (Northern Adriatic). Oceanol. Acta 14, 23–32.

Manfra, L., Maggi, C., d'Errico, G., Rotini, A., Catalano, B., Maltese, S., Moltedo, G., Romanelli, G., Sesta, G., Granato, G., Lanera, P., Amici, M., Martuccio, G., Onorati, F., Di Mento, R., Berducci, M.T., Chiaretti, G., Faraponova, O., Regoli, F., Tornambè, A., 2021. A Weight of Evidence (WOE) approach to assess environmental hazard of marine sediments from adriatic offshore platform area. Water 13, 1691. https://doi.org/10.3390/w13121691.

Maradonna, F., Ancillai, D., Notarstefano, V., Valenti, A., Leoni, T., Carnevali, O., 2020. An integrated approach to evaluate port sediment quality: From chemical characterization to multispecies bioassays. Sci. Tot. Environ. 746, 141204 https://doi.org/10.1016/j.scitotenv.2020.141204.

Mazaris, A.D., Almpanidou, V., Giakoumi, S., Katsanevakis, S., 2018. Gaps and challenges of the European network of protected sites in the marine realm. ICES J. Mar. Sci. 75, 190–198. https://doi.org/10.1093/icesjms/fsx125.

Medina, M.H., Correa, J.A., Barata, C., 2007. Micro-evolution due to pollution: Possible consequences for ecosystem responses to toxic stress. Chemosphere 67, 2105–2114. https://doi.org/10.1016/j.chemosphere.2006.12.024.

Moreira, L.B., Fernandes, G.M., Figueira, R.C.L., do Nascimento, R.F., Cavalcante, R.M., Valle Machado, W.T., Abessa, D.M.S., 2021. Sediment contamination and toxicity of Marine Protected Areas in southeastern Brazilian coast: A baseline for management planning. Mar. Pollut. Bull. 170, 112613 https://doi.org/10.1016/j.marpolbul.2021.112613.

Morroni, L., d'Errico, G., Sacchi, M., Molisso, F., Armiento, A., Chiavarini, S., Rimauro, J., Guida, M., Siciliano, A., Ceparano, M., Aliberti, F., Tosti, E., Gallo, A., Libralato, G., Patti, F.P., Gorbi, S., Fattorini, D., Nardi, A., Di Carlo, M., Regoli, F., 2020. Integrated characterization and risk management of marine sediments: The case study of the industrialized Bagnoli area (Naples, Italy). Mar. Environ. Res. 160, 104984 https://doi.org/10.1016/j.marenvres.2020.104984.

Muxika, I., Borja, A., Bonne, W., 2005. The suitability of the marine biotic index (AMBI) to new impact sources along European coasts. Ecol. Indic. 5, 19–31. https://doi.org/10.1016/j.ecolind.2004.08.004.

Pagano, M., Fernetti, M., Busetti, M., Ghribi, M., Camerlenghi, A., 2023. Multicriteria GIS-based analysis for the evaluation of the vulnerability of the marine environment in the Gulf of Trieste (north-eastern Adriatic Sea) for sustainable blue economy and maritime spatial planning. People Nat., in press. https://doi.org/10.1002/pan3.10537.

Palanques, A., Paradis, S., Puig, P., Masqué, P., Lo Iacono, C., 2022. Effects of bottom trawling on trace metal contamination of sediments along the submarine canyons of the Gulf of Palermo (southwestern Mediterranean). Sci. Tot. Environ. 814, 152658 https://doi.org/10.1016/j.scitotenv.2021.152658.

Park, K.-C., Pyo, H., Kim, W., Yoo, K.S., 2017. Effects of cooking methods and tea marinades on the formation of benzo[a]pyrene in grilled pork belly (Samgyeopsal). Meat Sci. 129, 1–8. https://doi.org/10.1016/j.meatsci.2017.02.012 0309-1740/.

Pavoni, E., Crosera, M., Petranich, E., Adami, G., Faganelli, J., Covelli, S., 2020. Partitioning and mixing behaviour of trace elements at the Isonzo/Soča River mouth (Gulf of Trieste, northern Adriatic Sea). Mar. Chem. 223, 103800 https://doi.org/10.1016/j.marchem.2020.103800.

Pearson, T.H., Rosenberg, R., 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. Oceanogr. Mar. Biol. Annu. Rev. 16, 229–311.

Perelo, L. W., 2010. Review: In situ and bioremediation of organic pollutants in aquatic sediments. 177, 81–89. https://doi.org/10.1016/j.jhazmat.2009.12.090.
Piccardo, M., Provenza, F., Anselmi, S., Broccoli, A., Terlizzi, A., Renzi, M., 2021. Use of Sediqualsoft® to determine the toxicity of cigarette butts to marine species: a weather simulation. TEST 9, 734. https://doi.org/10.3390/jmse9070734.
Pittura, L., Avio, C.G., Giuliani, M.E., d'Errico, G., Keiter, S.H., Cormier, B., Gorbi, S., Regoli, F., 2018. Microplastics as vehicles of environmental PAHs to marine organisms: combined chemical and physical hazards to the mediterranean mussels, Mytilus Galloprovincialis. Front. Mar. Sci. 5, 103. https://doi.org/10.3389/fmars.2018.00103.
Piva, F., Ciaprini, F., Onorati, F., Benedetti, M., Fattorini, D., Ausili, A., Regoli, F., 2011. Assessing sediment hazard through a weight of evidence approach with bioindicator organisms: A practical model to elaborate data from sediment chemistry, bioavailability, biomarkers and ecotoxicological bioassays. Chemosphere 83, 475–485. https://doi.org/10.1016/j.chemosphere.2010.12.064.
Prato, S., La Valle, P., De Luca, E., Lattanzi, L., Migliore, G., Morgana, J.G., Munari, C., Nicoletti, L., Izzo, G., Mistri, M., 2014. The "one-out, all-out" principle entails the risk of imposing unnecessary restoration costs: a study case in two Mediterranean coastal lakes. Mar. Pollut. Bull. 80, 30–40. https://doi.org/10.1016/j.marpolbul.2014.01.054.
Ramalhosa, E., Río Segade, S., Pereira, E., Vale, C., Duarte, A., 2006. Mercury cycling between the water column and surface sediments in a contaminated area. Water Res. 40, 2893–2900. https://doi.org/10.1016/j.watres.2006.05.023.
Regoli, F., d'Errico, G., Nardi, A., Mezzelani, M., Fattorini, D., Benedetti, M., Di Carlo, M., Pellegrini, D., Gorbi, S., 2019. Application of a weight of evidence approach for monitoring complex environmental scenarios: the case-study of off-shore platforms. Front. Mar. Sci. 6, 377. https://doi.org/10.3389/fmars.2019.00377.
Rosenberg, A.A., McLeod, K.L., 2005. Implementing ecosystem-based approaches to management for the conservation of ecosystem services. Mar. Ecol. Prog. Ser 300, 270–274. https://doi.org/10.3354/meps300270.
Russo, E., Anelli Monti, M., Mangano, M.C., Raffaetà, A., Sarà, G., Silvestri, C., Pranovi, F., 2020. Temporal and spatial patterns of trawl fishing activities in the Adriatic Sea (Central Mediterranean Sea, GSA17). Ocean Coast. Manage. 192, 105231 https://doi.org/10.1016/j.ocecoaman.2020.105231.
Sciberras, M., Jenkins, S.R., Kaiser, M.J., Hawkins, S.J., Pullin, A.S., 2013. Evaluating the biological effectiveness of fully and partially protected marine areas. Environ. Evidence 2, 4. https://doi.org/10.1186/2047-2382-2-4.
Shi, W., Xu, M., Liu, Q., Xie, S., 2022. Polycyclic aromatic hydrocarbons in seawater, surface sediment, and marine organisms of Haizhou Bay in Yellow Sea, China: Distribution, source apportionment, and health risk assessment. Mar. Pollut. Bull. 174, 113280 https://doi.org/10.1016/j.marpolbul.2021.113280.
Sigamani, S., Perumal, M., Arumugam, S., Jose, H.M.P.M., Veeraiyan, B., 2015. AMBI indices and multivariate approach to assess the ecological health of Vellar-Coleroon estuarine system undergoing various human activities. Mar. Pollut. Bull. 100, 334–343. https://doi.org/10.1016/j.marpolbul.2015.08.028.
Teixeira, H., Berg, T., Uusitalo, L., Fürhaupter, K., Heiskanen, A.-S., Mazik, K., Lynam, C. P., Neville, S., Rodriguez, J.G., Papadopoulou, N., Moncheva, S., Churilova, T., Kryvenko, O., Krause-Jensen, D., Zaiko, A., Veríssimo, H., Pantazi, M., Carvalho, S., Patrício, J., Uyarra, M.C., Borja, À., 2016. A catalogue of marine biodiversity indicators. Front. Mar. Sci. 3, 207. https://doi.org/10.3389/fmars.2016.00207.
Trobec, A., Busetti, M., Zgur, F., Baradello, L., Babich, A., Cova, A., Gordini, E., Romeo, R., Tomini, I., Poglajen, S., Diviacco, P., Vrabec, M., 2018. Thickness of marine Holocene sediment in the Gulf of Trieste (northern Adriatic Sea). Earth Syst. Sci. Data 10, 1077–1092. https://doi.org/10.5194/essd-2017-135.
US EPA., 2017. Toxicological Review of Benzo[a]pyrene. EPA/635/R-17/003Fa.
Vasilakopoulos, P., Palialexis, A., Boschetti, S.T., Cardoso, A.C., Druon, J.-N., Konrad, C., Kotta, M., Magliozzi, C., Palma, M., Piroddi, C., Ruiz-Orejón, L.F., Salas-Herrero, F., Stips, A., Tornero, V., Hanke, G., 2022. Marine Strategy Framework Directive, Thresholds for MSFD Criteria: state of play and next steps, EUR 31131 EN, Publications Office of the European Union, Luxembourg, 2022, ISBN 978-92- 76-53689-5, doi:10.2760/640026, JRC128344.
Vollenweider, R.A., Giovanardi, F., Montanari, G., Rinaldi, A., 1998. Characterization of the trophic conditions of marine coastal waters with special reference to the NW Adriatic Sea: proposal for a trophic scale, turbidity and generalized water quality index. Environmetrics 9, 329–357. https://doi.org/10.1002/(SICI)1099-095X(199805/06)9:3<329::AID-ENV308>3.0.CO;2-9.
Wang, W., Lin, C., Wang, L., Jiang, R., Liu, Y., Lin, H., Chen, J., 2021. Effects of human activities on the spatial distribution, ecological risk and sources of PTEs in coastal sediments. Int. J. Environ. Res. Public Health 18, 12476. https://doi.org/10.3390/ijerph182312476.
Zheng, N., Wang, S., Dong, W., Hua, X., Li, Y., Song, X., Chu, Q., Hou, S., Li, Y., 2019. The toxicological effects of mercury exposure in marine fish. Bull Environ Contam Toxicol 102, 714–720. https://doi.org/10.1007/s00128-019-02593-2.

Further reading

Guidetti, P., Milazzo, M., Bussotti, S., Molinari, A., Murenu, M., Pais, A., Spanò, N., Balzano, R., Agardy, T., Boero, F., Carrada, G., Cattaneo-Vietti, R., Cau, A., Chemello, R., Greco, S., Manganaro, A., Notarbartolo di Sciara, G., Russo, G.F., Tunesi, L., 2008. Italian marine reserve effectiveness: does enforcement matter? Biol. Conserv. 141, 699–709. https://doi.org/10.1016/j.biocon.2007.12.013.
Habitat Directive, 1992. Directive 92/43/EEC of the Council on the Conservation of natural habitats and of wild fauna and flora.
Neff, J.M., Stout, S.A., Gunster, D.S., 2004. Ecological risk assessment of polycyclic aromatic hydrocarbons in sediments: identifying sources and ecological hazard. Integr. Environ. Assess. Manage. 1, 22–33. https://doi.org/10.1897/ieam_2004a-016.1.