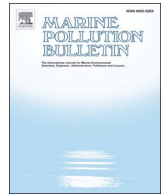




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## Note

## Assessing the ecological status of Italian lagoons using a biomass-based index

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## ABSTRACT

We compared the performance of abundance- and biomass-based M-AMBI in the 13 major Italian lagoons, using a benthic dataset constituted by 208 sampling sites. The relative importance of ecological groups changed when using abundance or biomass, sometimes leading to an improved ecological status classification. Being biomass more ecologically relevant than abundance, the adoption of a biomass-based index may better describe the ecological status of lagoons, where the community is naturally disturbed and dominated by tolerant and opportunistic species.

Ecological indicators are an effective way to characterize marine ecosystem health, and their number is rapidly increasing (Borja et al., 2015). Among the biological quality elements highlighted by the European Water Framework Directive 2000/60/EC, benthic macrofauna is known to be probably the best effective indicator of pollution stress, as it shows predictive responses to different levels of anthropogenic impact. Based on the Pearson and Rosenberg (1978) paradigm several biotic indices have been proposed in recent years (Borja et al., 2015). The AZTI's marine biotic index (AMBI; Borja et al., 2000) and its multivariate version (M-AMBI; Muxika et al., 2007) are probably the most widely used benthic indices all over the world (Borja et al., 2015). In Europe many countries have officially adopted the index for the description of ecological quality of coastal waters (Bulgaria, France, Germany, Italy, Romania, Slovenia and Spain; Borja et al., 2009; Birk et al., 2012). AMBI relies on the calculation of the biotic coefficient, which is based in turn on the proportion of disturbance-sensitive taxa and is expressed on a continuous scale ranging from 0 (best status) to 7 (worst status: azoic). The AMBI approach follows a model (Grall and Glemarec, 1997) which categorizes benthic invertebrates into five ecological groups (from EG-I, sensitive, to EG-V, first order opportunists), depending on their dominance along a gradient of organic enrichment. Recently, Warwick et al. (2010) suggested to estimate AMBI using biomass (bAMBI) and production (pAMBI). This because in an assemblage the abundance of a species can be relatively a poor measure of its functional importance, particularly in stressed situations when the insensitive species tend to be small bodied opportunists (Warwick et al.,

2010). Muxika et al. (2012) successfully assessed the proposed modification to AMBI along the Basque coast (northern Spain), showing that those AMBI modifications were highly correlated and thus useful to assess the benthic quality status, if boundaries between quality classes were re-determined. More recently, Mistri and Munari (2015) tested the performance of biomass-based AMBI (bAMBI and pAMBI) in transitional ecosystems, finding good agreement between the response of all biomass-based indices and disturbance expressed by the severity of pressures. The use of a biomass-based index for the assessment of the ecological quality status in transitional systems is not trivial, since several studies (Magni et al., 2009; Sigovini et al., 2013; Prato et al., 2014) suggested that the use of indices based on species tolerance/sensitivity need to be adapted where the community is naturally disturbed and dominated by small-sized opportunists. The use of biomass in calculating M-AMBI (M-bAMBI) was tested by Cai et al. (2014, 2015) in assessing the benthic status of Bohai Bay (north of China), a shallow water basin receiving industrial and municipal wastewater from coastal cities. Those authors found that M-bAMBI seemed more effective than M-AMBI in indicating human pressures of the Bay (Cai et al., 2015).

Since lagoons are often characterized by high benthic biomass (McLusky, 1989), and biomass is a measure of ecosystem functions (Warwick et al., 2010), in this note we explore the performance of M-bAMBI in the most important Italian lagoons, at which the disturbance status was known. We assembled a data set of macrofaunal counts from 13 large Italian lagoons (Fig. 1) occurring along a cline of 7° of latitude (between 45°44'N and 39°56'N). Along Italian coasts there are almost

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Fig. 1. Location of the studied lagoons in the Italian coast.

Table 1

Major geographical and ecological features of the 13 studied lagoons (mt: microtidal; nt: non-tidal; S: richness; H': diversity; SD: standard deviation).

Lagoon	Latitude	Longitude	Area (km <sup>2</sup> )	Mean depth (m)	Salinity	Typology	Sampling sites	Tot n samples	S ( ± SD)	H' ( ± SD)
Grado Marano	45°42'N	13°20'E	160	1.5	Poly/euhaline	mt	21	21	25.6 ± 14.3	2.9 ± 1.0
Venice	45°24'N	12°19'E	500	2.5	Poly/euhaline	mt	20	43	18.4 ± 12.4	2.1 ± 0.6
Caleri	45°05'N	12°18'E	11	2.0	Meso/polyhaline	mt	4	8	14.0 ± 6.8	1.8 ± 0.6
Marinetta	45°03'N	12°21'E	10	0.8	Meso/polyhaline	mt	6	12	14.6 ± 4.1	1.3 ± 0.8
Barbamarco	45°00'N	12°27'E	8	0.8	Meso/polyhaline	mt	2	4	15.5 ± 8.7	1.6 ± 0.5
Canarin	44°55'N	12°29'E	10	0.8	Meso/polyhaline	mt	3	6	12.7 ± 2.3	2.0 ± 0.9
Scardovari	44°51'N	12°24'E	32	1.5	Meso/polyhaline	mt	5	10	22.3 ± 4.4	2.3 ± 0.5
Goro	44°49'N	12°18'E	37	2.0	Meso/polyhaline	mt	16	16	26.2 ± 6.3	1.9 ± 0.9
Comacchio	44°36'N	12°10'E	117	0.8	Euhaline	nt	4	22	9.6 ± 4.7	1.7 ± 0.7
Baiona	44°30'N	12°14'E	12	1.0	Polyhaline	mt	3	6	32.5 ± 3.5	3.0 ± 0.2
Lesina	41°52'N	15°26'E	51	0.8	Meso/polyhaline	nt	4	12	14.4 ± 4.3	2.3 ± 0.5
Orbetello	42°26'N	11°11'E	27	1.5	Polyhaline	nt	9	36	13.1 ± 6.6	2.0 ± 0.7
Tortoli	39°56'N	09°40'E	3	1.0	Poly/euhaline	nt	6	12	25.3 ± 8.5	3.1 ± 0.8

Table 2

Reference conditions for various lagoon typologies used in M-AMBI calculations. H': diversity, S: richness.

Tidal range	Salinity	AMBI	H'	S
Not tidal	–	1.85	3.3	25
Microtidal	Oligo-meso-poly	2.14	3.4	28
Microtidal	Eu-iper	0.63	4.23	46

170 lagoons, but 140 of them have a surface area < 10 km<sup>2</sup>. With the exclusion of Orbetello Lagoon and Stagno di Tortoli (Tyrrhenian Sea), all lagoons with area > 10 km<sup>2</sup> (e.g. Grado-Marano, Venice, Po Delta, Comacchio, Lesina) are located along the Western Adriatic coasts. Our data set comprises all main Adriatic and Tyrrhenian Italian lagoons (Caleri, Marinetta, Barbamarco, Canarin, Scardovari and Goro are those in the present Po Delta). In Table 1, major geographical and ecological features of the 13 lagoons are shown. A total of 103 sites, representative of the different habitats found within each lagoon, were sampled

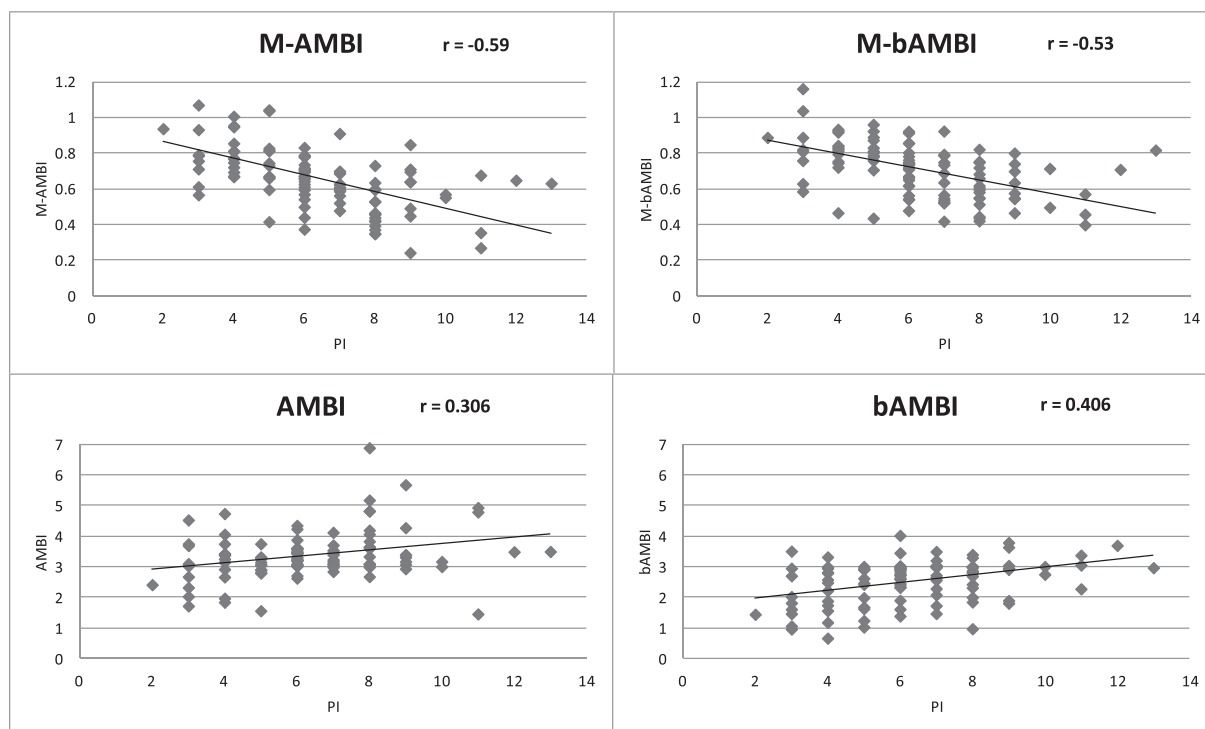


Fig. 2. Correlation between Pressure Index (PI) and abundance-based (AMBI and M-AMBI) and biomass-based (bAMBI and M-bAMBI) indices.

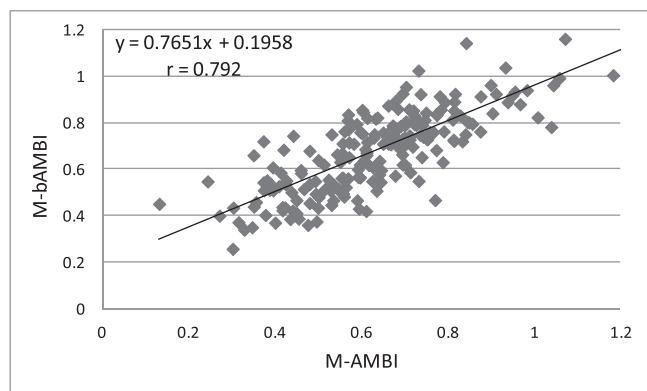


Fig. 3. Regression between M-AMBI and M-bAMBI.

**Table 3**  
Quality class boundaries for M-AMBI and M-bAMBI.

	M-AMBI	M-bAMBI
High/good	0.96	0.930
Good/moderate	0.71	0.739
Moderate/poor	0.57	0.632
Poor/bad	0.46	0.548

repeatedly over time, for a total of 208 sampling points. In Table 1 the main benthic community parameters at each of the 13 lagoons (mean number of species,  $S$ , and mean diversity,  $H'$ ) are shown. Over 400 macrobenthic taxa were gathered at the 208 sampling points, with annelids displaying the highest number of taxa, followed by crustaceans and molluscs. Most species found in the 13 lagoons are cosmopolitan (i.e. with a wide geographical distribution, such as *Capitella capitata*, *Polydora ciliata*, *Streblospio shrubsolii*, and *Hediste diversicolor*). Several endemic species were also found (such as *Corophium orientale*, *Microdeutopus algicola*, *Pectinaria koreni* and *Ampithoe riedli*), together with many non-indigenous species (such as *Anadara inaequalis*, *A. demiri*,

*Arcuatula senhousia*, *Ruditapes philippinarum*, *Rhithropanopeus harrisi*, *Dyspanopeus sayii* and *Grandidiellerella japonica*), which were most common in the Po Delta lagoons.

Pressures (Supplementary material, Table S1) were quantified (1: low, 2: medium and 3: high) for each location and sampling station, as partial pressure, total pressure and as a pressure index (PI), following an approach close to that proposed by Aubry and Elliott (2006), based upon best professional judgment. According to Borja et al. (2011) the total pressure was the sum of partial pressures, and the pressure index was calculated as an average value of the pressures. Abundance (M-AMBI) and biomass (M-bAMBI) based indices were calculated using AMBI 5.0 software (freely available at <http://ambi.azti.es>). Reference conditions were those reported by the Italian Act 260/10, which considers three lagoon typologies, with different reference conditions, as a function of tidal range and salinity (Table 2). Regression between AMBI-based indices and Pressure Index (PI) was performed to analyze the agreement in the pollution classification, and significance was assessed through regression ANOVA. In Fig. 2 the relationship between PI and abundance-based indices (AMBI and M-AMBI) and biomass-based indices (bAMBI and M-bAMBI) at the 103 lagoonal stations is shown (M-AMBI:  $F = 53.6$ ;  $P < 0.001$ ; AMBI:  $F = 10.4$ ;  $P < 0.01$ ; M-bAMBI:  $F = 39.4$ ;  $P < 0.001$ ; bAMBI:  $F = 19.7$ ;  $P < 0.001$ ). The relationship between M-AMBI and M-bAMBI was fitted using a trend line (Fig. 3;  $F = 116.9$ ;  $P < 0.001$ ). The regression equation was then used to calculate the quality class boundaries of M-bAMBI corresponding to those for M-AMBI. Once the sampling sites were classified according to these boundaries (Table 3), a Kappa analysis (Landis and Koch, 1977) was carried out to detect the agreement in the quality classification between the indices, as done during the European intercalibration exercises (Borja et al., 2007). The level of agreement between the indices was established, based upon the equivalence table from Monserud and Leemans (1992). The results showed a good agreement (kappa coefficient = 0.694) between the classifications obtained from M-AMBI and M-bAMBI.

Compared to M-AMBI, there were more sites assigned to an undisturbed status in M-bAMBI. In Fig. 4, the ecological status (ES) gathered through M-AMBI and M-bAMBI is shown. At certain lagoons,

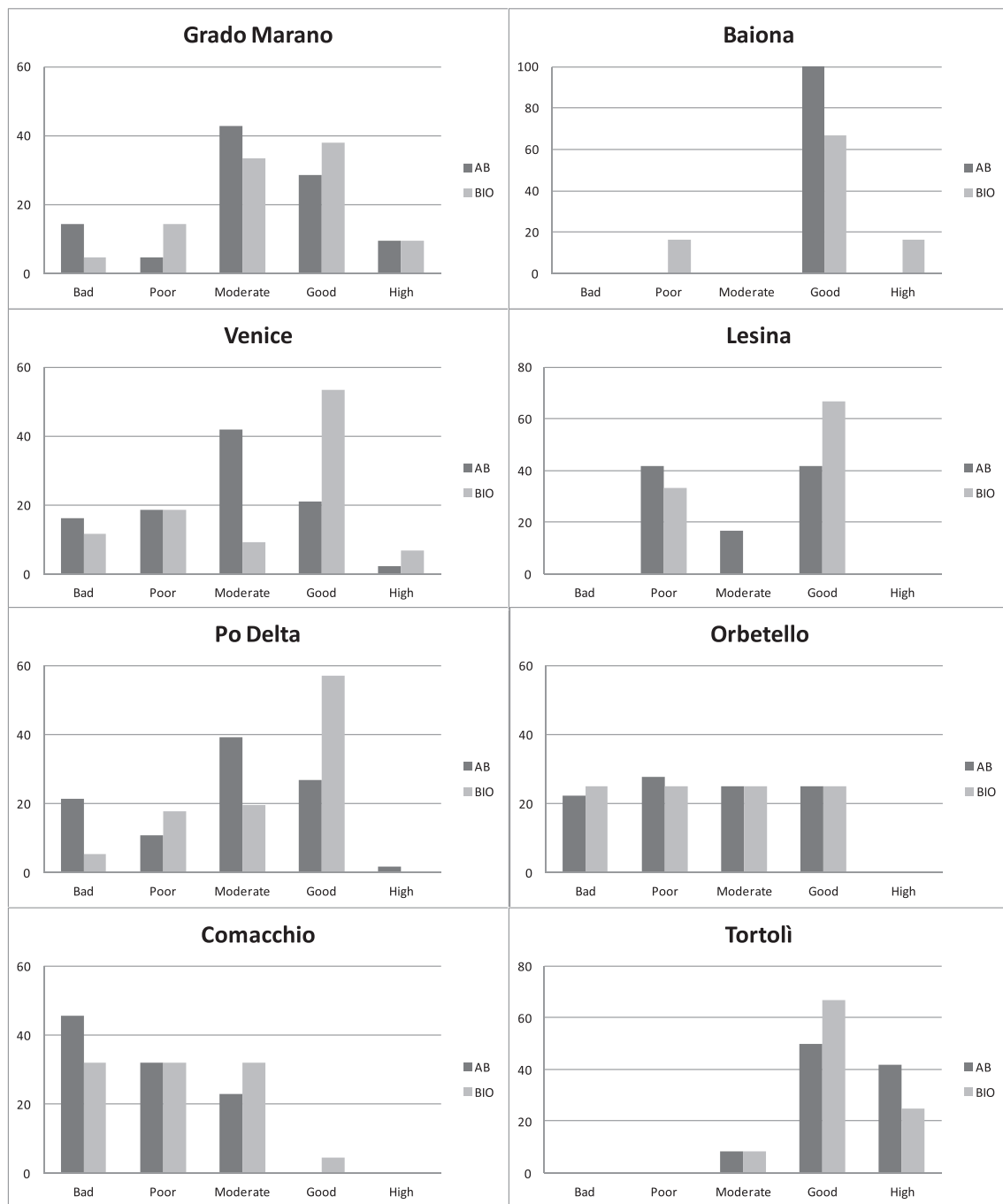
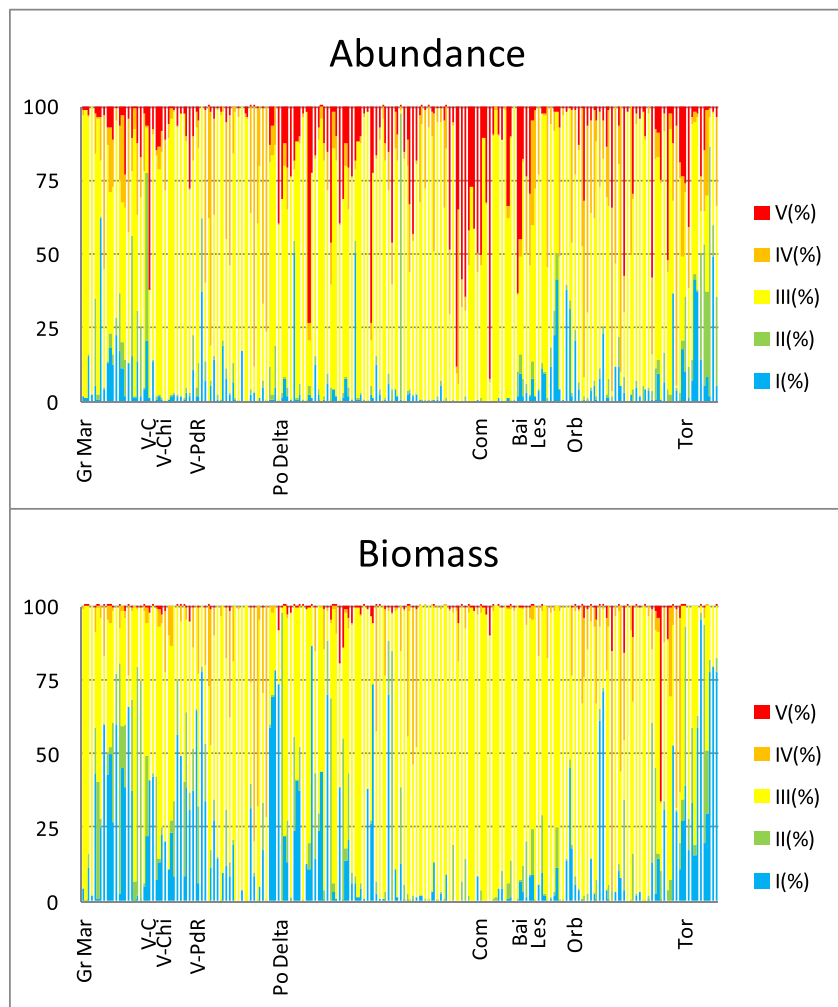


Fig. 4. Ecological status at the various lagoons through M-AMBI (dark grey) and M-bAMBI (light grey). AB: abundance, BIO: biomass.

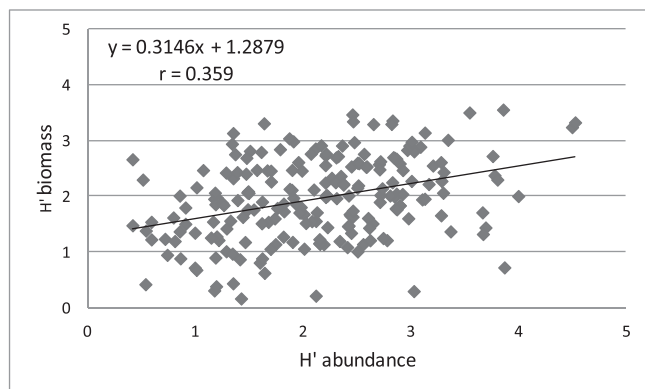
differences in ES were minimal, e.g. at Comacchio, where the benthic community is so degraded (Munari and Mistri, 2014) that the ES was disturbed, either by using abundance or biomass. At other lagoons (e.g. Grado Marano, Venice and Po Delta), instead, a slight improvement in the ES can be noticed, with some sites changing their ES from Moderate to Good.

AMBI bases its functioning on dividing benthic species into previously defined ecological groups (EG-I to EG-V), and then determining the respective proportion of the different groups in the benthic community. Its calculation relies on the relative decrease of sensitive species (EG-I) confronted with increasing disturbance in the sediment or, conversely, the increase of species that are resistant or indifferent to disturbance (EG-II and EG-III), or that are even encouraged by such conditions like the opportunist species (EG-IV and EG-V) that

proliferate when the sediment is rich in organic matter or pollutants. Different level of disturbance was present at our 103 sampling stations, and this was reflected by the relative abundance of ecological groups with, e.g., EG-I dominant at some stations at Grado Marano, Baiona, Lesina and Tortoli lagoons, and EG-V dominant, e.g., at Comacchio, and some sites in the Po Delta and Orbetello (Fig. 5). The different distribution of EG-I to EG-V taxa in our 208 sites was obviously reflected by M-AMBI scores. We are however aware that AMBI/M-AMBI, considering the abundances of stress-tolerant species to detect anthropogenic impacts, does not consider the fact that tolerant species may also be tolerant of natural stressors (the “Estuarine Quality Paradox”, Dauvin and Ruellet, 2009). Warwick et al. (2010) suggested to use production data instead of abundance because production is the most common measure of ecosystem function. However, given the difficulty



**Fig. 5.** Concentration of Ecological Groups (EG) I- to V at each sampling site considering abundance or biomass (GrMar: Grado Marano; V-C: Venice Central; V-Chi: Venice Chioggia; V-PdR: Venice Palude della Rosa; Po Delta: Caleri, Marinetta, Barbamarco, Canarin, Scardovari, Goro; Com: Comacchio; Bai: Baiona; Les: Lesina; Orb: Orbetello; Tor: Tortoli).



**Fig. 6.** Regression between abundance-based and biomass-based diversity ( $H'$ ).

of production assessment, biomass is often used as a proxy measure (terHorst and Munguia, 2008). Since biomass (and thus production) is more ecologically relevant than abundance, its use to derive AMBI-based indices is intriguing, especially in lagoons where the community is naturally disturbed and dominated by tolerant and opportunist species.

Assessing the ES of Bohai Bay (Yellow Sea, China), Cai et al. (2014) found M-AMBI and M-bAMBI to produce quite similar results, with slight lower M-bAMBI values. Those authors justified lower M-bAMBI values with the lower diversity values calculated with biomass

compared with those calculated using density. We also found  $H'$  calculated with biomass to be generally lower than  $H'$  calculated with abundance (Fig. 6), however, differently from Cai et al. (2014) we found slight lower M-AMBI values. In their study on the Basque coast, Muxika et al. (2012) found that the distributions of ecological groups' dominances were very similar when biomass was used instead of abundance. Conversely, in our lagoonal data set, the proportion of ecological groups into the community varied greatly if we considered abundance or biomass-based data (Fig. 5). In certain cases, the use of biomass instead of abundance resulted in a better ES classification, despite the “Moderate/Good” boundary for M-bAMBI was higher than that for M-AMBI (0.739 vs 0.710). For example, the site 4.09 in the Caleri lagoon (Po Delta) scored the ES “Moderate” (EQR = 0.694) by M-AMBI, but “Good” (EQR = 0.926) by M-bAMBI. At this site, considering abundance, the benthic community was numerically dominated by EG-III (60.5%), followed by and EG-V (20.8), and EG-IV (14.2%). Numerically dominant species were *Streblospio shrubsolii* (48.7% of the whole community abundance), *Capitella capitata* (15.9%), *Polydora ciliata* (14.2%), and *Hydroides dianthus* (7.9%). These figures changed when we considered biomass, since *S. shrubsolii* constituted 11.3% of the total biomass, *C. capitata* (3.7%), *Polydora ciliata* (3.2%), and *Hydroides dianthus* (0.7%). Conversely, *Palaemon serratus* constituted 19.8% of community biomass (but only 0.3% of community abundance), and *Neanthes succinea* constituted 14.5% of total biomass (1.3% of total abundance). Lagoonal macrobenthos is mainly composed by species tolerant to the naturally disturbed conditions, which include highly variable temperature, salinity and oxygen concentration (Elliott



and Quintino, 2007). Because of the dominance in terms of abundance of tolerant species, the use of abundance-based indices does not always guarantee a correct assessment of ES, as it occurs in the marine environment.

Italian lagoons exhibit different and peculiar characteristics depending on their geographical, hydrodynamic and ecological features: these variations generate composite gradients that involve salinity, marine water renewal (e.g. residence time), nutrients, turbidity and sediment structure. (Tagliapietra et al., 2009). Superimposed to this huge natural variability there is a century-old human activity producing pressures. Coastal lagoons represent important and fragile ecosystems in the coastal landscape, providing key ecosystems services such as water quality improvement, fisheries resources, habitat and food for migratory and resident animals. For the evaluation of their ecological status, the metric adopted (abundance or biomass) can lead to changes in results. Because of lagoonal inherent variability that leads to the coexistence of many species spanning all ecological groups, the relative contribution of the different macrobenthic taxa can vary greatly depending on the adopted metric. The concept of ecological status must consider the structure and function of the lagoon ecosystem, and results from this study suggest that a biomass-based index may better describe the ecological status of Italian lagoons.

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