

# Impact of hydromorphological pressures on the macrophytes bioindicators of the ecological water quality in Mediterranean rivers

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## Abstract

One of the important tools to evaluate the ecological quality of surface water is the Macrophytes indices based on the bioindication capacity of aquatic plants. In Mediterranean rivers (France, Spain, and Portugal), the development of some macrophytes indices like l'Indice Biologique Macrophytes Rivières (IBMR), the biological metric score (BMS), as well as the Fluvial Macrophyte Index (IMF) are founded on the determination of the indicator values of the floristic reference lists.

The aim of this study was to test the impact of the eco-Mediterranean differences (from one country to another) on the indicator taxa by comparing the indicator values of the Euro- Mediterranean macrophyte indices. With this in mind, we explore the possibility of the introduction of the Euro-Mediterranean macrophytes-based indices in Morocco (i.e. the hydrological basin of Sebou (HBS)) as a part of a preliminary attempt to develop the first Afro-Mediterranean macrophyte index.

We confirm that the ecological amplitude and species optima vary between Mediterranean ecoregions, and indicator taxa differ between countries: There are medium to small correlations between Mediterranean indices: IBMR/BMS ( $p = 0.000$ ,  $R^2 = 0.57$ ), IMF/BMS ( $p = 0.000$ ,  $R^2 = 0.34$ ), and IBMR/IMF ( $p = 0.000$ ,  $R^2 = 0.30$ ). Five species exhibit major differences in indicator values: *Zannichellia palustris* and *Potamogeton pectinatus* have more eutrophic indicator values in France (IBMR) than in Spain (IMF). *Potamogeton nodosus*, *Amblystegium riparium* and *Lycopus europaeus* have broader ecological amplitudes in Portugal (BMS) than in France (IBMR) and in Spain (IMF), where it is restricted to eutrophic conditions. Furthermore, the three indicator systems include different indicator-taxon numbers.

The comparison of the HBS elaborated list with the Euro-Mediterranean indices revealed the low level of common taxa approximately 6.76% of all indicator species used in the French index (IBMR), 10.48% in the Portuguese index (IMF) and 12.38% in the Spanish index (BMS).

These results show the inadequacy of the trophic indices approach with the HBS conditions and thus the need for the development of an index based on biotic indices approach.

### Keywords

Ecological water quality, Macrophytes, reference list, bioindication, hydromorphology, Mediterranean rivers

## Introduction

Due to their high sensibility to different environmental stresses and their ability to assess the dynamic and the cumulative effects of different stressing factors, macrophytes species are considered good bioindicators. This bioindication power of macrophytes has generated a proliferation in the number of macrophyte-based indices in the last decades.

At the present time, the approaches for estimating macrophyte communities' quality in the Mediterranean rivers are:

- The approach based on the assumption that environments that have not been impacted have a greater diversity of species than degraded environments (community structure approaches): Indice di Biodiversita` Riparia (IBR) (Maggioni et al. 2009) in Italy is based on biodiversity of macrophytes on the banks. (Patrick 1977) proved that assemblages with similar diversity scores could represent streams with significantly different chemical conditions.
- The Biotic indices approach based on the assumption that biological assemblages in impaired sites should be different from those in reference sites:
  - The Iberian multimetric plant index (IMPI) (Ferreira et al. 2005), in the Iberian Peninsula (Portugal, Spain).
  - The Riparian Vegetation index (RVI) (Aguiar et al. 2009) in Portugal.
  - River Macrophyte Index (RMI) (Kuhar et al. 2011) in Slovenia, based on the relative abundance of sensitive and/or tolerant taxa.
- The approach based on indicator values calculated for an elevated number of aquatic species, according to the species' relative sensitivity and tolerance to nutrients and/or to other abiotic stress factors. The Indices designed to respond to nutrient enrichment using indicator species in Mediterranean rivers are:
  - The Indice Biologique Macrophytique en Rivières (IBMR): developed in France by (Haury et al. 2006) for assessing water trophy and organic pollution and calculated using the following formula:

$$IBMR = \frac{\sum_i^n Ei \times Ki \times Csi}{\sum_i^n Ei \times Ki}$$

where  $C_{si}$  is the specific rate of trophic level– ranged from 0 (heavy organic pollution and heterotrophic taxa) to 20 (oligotrophy);  $E_i$  represents the coefficient of ecological amplitude: Coefficient 1, representing wide amplitude, covered three classes of trophy, and coefficient 3, representing a very limited amplitude, was restricted to just one class;  $K_i$  is the scale of cover, going from 1 to 5 (1: <0,1%; 2: 0,1 – <1%; 3: 1– <10%; 4: 10 – <50%; 5: ≥50%).

- The biological metric scores (BMS): developed by (Dodkins et al. 2012) in Portugal. This index is the mean of the species scores that occur at that site, weighted by their cover, i.e. the Weighted Averaging (WA) equation (Braak and Looman 1986):

$$S = \frac{\sum_i^n C_i \times Q_i}{\sum_i^n C_i}$$

where  $S$  = site score,  $n$  = number of species;  $C_i$  = cover scale value of species  $i$ ; and  $Q_i$  = score of species  $i$ . The cover scale values used to weight the mean were: 0 (for 0% macrophyte cover relative to the channel area), 1 (≤1% cover), 2 (≤5% cover), 5 (≤33% cover) and 6 (>33 cover).

- The index of macrophytes (IM), the Macroscopic Aquatic Vegetation Index (IVAM) and The Fluvial Macrophyte Index (IMF) (Alcaraz et al. 2006; Flor-Arnau et al. 2015; Suárez et al. 2005) in Spain. The Fluvial Macrophyte Index (IMF) is calculated using the following formula:

$$I.M.F = \frac{\sum_i^n E_i \times K_i \times C_{si}}{\sum_i^n E_i \times K_i}$$

where  $K_i$  is the coating of the taxa at the station -range: 1-5; 1 (<0.1%), 2 (0.1–1%), 3 (1–10%), 4 (10–50%), 5 (> 50%);  $C_{si}$  is the sensitivity value for eutrophy (range: 1–20);  $E_i$  is the value of stenoicity or ecological amplitude (range: 1–3). The IMF score is obtained from the formula of Zelinka and Marvan (1961).

Taking into consideration that the development of macrophytes assemblages strongly depends on a variety of abiotic and biotic factors and it is assumed that the most important of them are nutrient concentrations (Dodkins et al. 2012; Robach et al. 1996; Schneider et al.2000; Szoszkiewicz et al. 2006; Thiebaut et al. 2002; Whitton 1975), and hydromorphological characteristics, such as altitude, flow velocity, water depth, width of river bed and type of substrate (Baláži and Hrivnák 2017), the overall purpose of this paper is to investigate the influence of localized hydromorphological differentiation for the bioindication of macrophytes in Mediterranean countries. In

particular, we focus on the following question: Is there evidence of a role of hydromorphological differentiation in the diversity of macrophyte taxa included in Mediterranean indices? Is there any evidence for the impact of ecoregion differentiation on the macrophytes indicator values? In other words, are the macrophytes more impacted by trophic status or by the hydromorphological characteristics of each Mediterranean country? Is there any possibility to adopt and/or adapt any Euro-Mediterranean macrophytes-based indices in Morocco (HBS)?

## Methods

All currently used and published Mediterranean macrophyte indices based on species indicator values for assessment of river trophic status are included in this study. We didn't take into consideration indices with low taxonomic rank resolution (family and order): Macroscopic Aquatic Vegetation Index (IVAM) and the index of macrophytes (IM). Three macrophyte indices meet the above-indicated criteria: The Fluvial Macrophyte Index (IMF), the Biological metric scores (S), and l'Indice Biologique Macrophytes Rivières (IBMR).

Comparison of species indicator values between different Mediterranean indices was performed using correlation analysis.

An extensive field survey of macrophytes communities (aquatic and riparian species) in HBS and its tributaries (39 stations) has been carried out. Identification of the macrophytes was taken using field identification guides (Ahayoun et al. 2007; Coudreuse et al. 2005; Fennane et al. 1999; Fennane et al. 2007; Valdés 2002).

In order to ensure comparability of species, taxa names were screened for synonyms and harmonized if necessary.

## Results

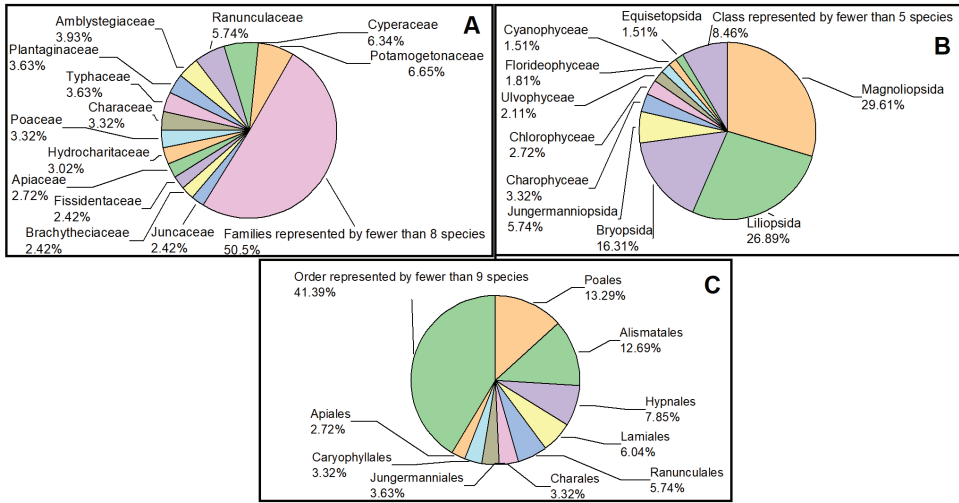
### Mediterranean indices comparison

The most striking results to emerge from Mediterranean indices comparison are:

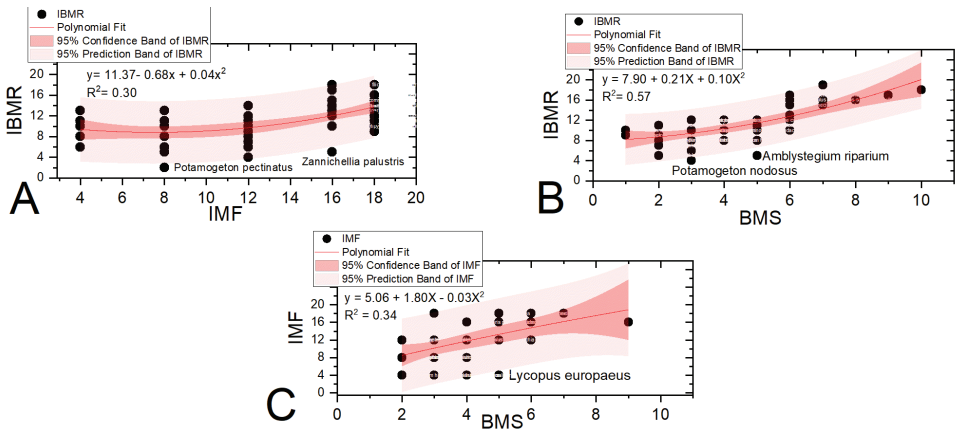
### IBMR compared to IMF

A total of 68 species are included in both IBMR and IMF. Half of these species have an IMF value between 16–18 (Figure 3). The indicator values are significantly correlated ( $p = 0.000$ ,  $R^2 = 0.30$ ) (Figure 2). Two species differ from the regression curve. In the two cases the IBMR value is lower than the IMF (*Zannichellia palustris*, *Potamogeton pectinatus*).

A total of 158 taxa have only an IBMR, but not an IMF indicator value, while 56 taxa have only IMF indicator value but not an IBMR.



**Figure 1.** **A** families **B** classes, and **C** orders of macrophytes species included in the Mediterranean trophic indices: IBMR, IMF, and BMS.

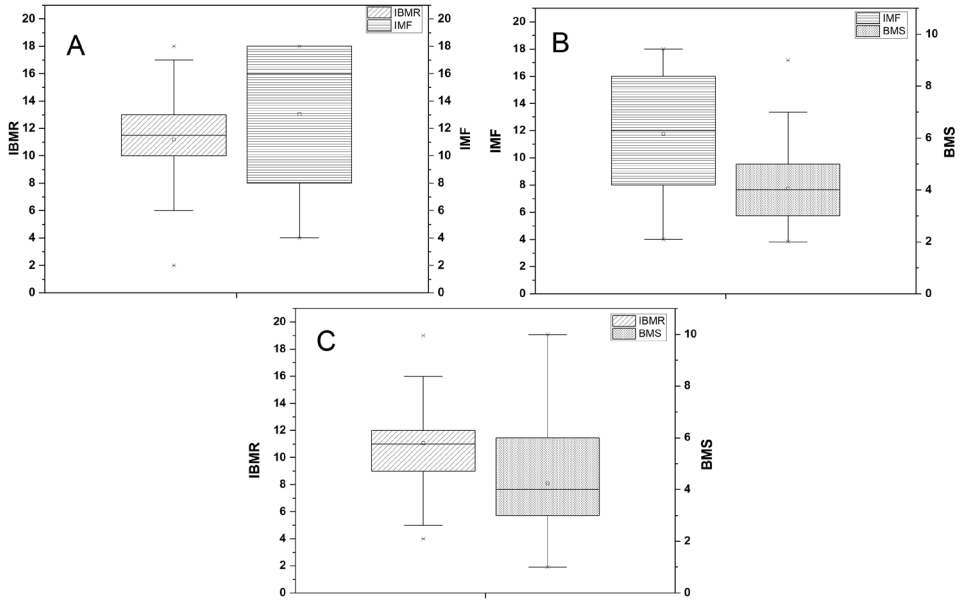


**Figure 2.** Polynomial regression of **A** IBMR and IMF **B** IBMR and BMS **C** IMF and BMS.

**IBMR compared to BMS**

A total of 47 species are included in both IBMR and BMS. The indicator values are significantly correlated ( $p = 0.000$ ,  $R^2 = 0.57$ ). Two species differ from the regression curve. In the two cases the IBMR value is lower than the IMF (*Amblystegium riparium*, *Potamogeton nodosus*).

A total of 179 taxa have only an IBMR, but not a BMS indicator value, while 58 taxa have only an IMF indicator value but not an IBMR.



**Figure 3.** boxplots: indicator values of species that are included in **A** (IBMR) and (IMF) **B** (IMF) and (BMS) **C** (IMF) and (BMS).

### IMF compared to BMS

A total of 35 species are included in both IMF and BMS. The indicator values are significantly correlated ( $p=0.000$ ,  $R^2=0.34$ ). One species differs from the regression curve. In this case the IMF value is lower than the BMS (*Lycopus europaeus*).

A total of 89 taxa have only an IMF, but not BMS indicator values, while 70 taxa have only BMS indicator values but not IBMR.

### HBS macrophytes compared to European trophic indices

Our field work and analysis revealed that a limited number (23 indicator species) of macrophytes recorded in HBS are utilized as bioindicators in biological monitoring for the ecological status assessment in rivers in Euro-Mediterranean countries (Table 1). Fourteen species are used in IBMR, thirteen species in BMS and IMF. This limited number of indicator species represents only 6.76% of all indicator species used in the French index (IBMR), 10.48% in the Portuguese index (IMF) and 12.38% in the Spanish index (BMS).

If we extend our analysis to other European indices i.e.:

- The British index: The Mean Trophic Rank (MTR), there are only ten species of HBS that have MTR indicator value: *Berula erecta*, *Elodea canadensis*,

**Table 1.** The list of aquatic taxa of HBS that are included in IBMR, BMS, IMF.

Species	IBMR		BMS	IMF	
	Csi	Ei	Qi	Csi	Ei
<i>Agrostis stolonifera</i>	10	1		12	2
<i>Arundo donax</i>			1		
<i>Berula erecta</i>	14	2			
<i>Elodea canadensis</i>	10	2	1		
<i>Epilobium hirsutum</i>			2	4	1
<i>Equisetum ramosissimum</i>				18	3
<i>Helosciadium nodiflorum</i>	10	1	3	4	1
<i>Hygrohypnum luridum</i>	19	3			
<i>Lemna gibba</i>	5	3	2	8	2
<i>Ludwigia palustris</i>			5		
<i>Mentha aquatica</i>	12	1	3	12	2
<i>Mentha longifolia</i>				18	3
<i>Mentha pulegium</i>			4		
<i>Nasturtium officinale</i>	11	1	2	8	2
<i>Phragmites australis</i>	9	2	1		
<i>Potamogeton nodosus</i>	4	3	3		
<i>Potamogeton pectinatus</i>	2	2		8	3
<i>Ranunculus bulbosus</i>			4		
<i>Rumex conglomeratus</i>				8	2
<i>Scrophularia auriculata</i>				4	1
<i>Typha angustifolia</i>	6	2			
<i>Veronica beccabunga</i>	10	1	3	12	3
<i>Zannichellia palustris</i>	5	1		16	3

*Helosciadium nodiflorum*, *Hygrohypnum luridum*, *Lemna gibba*, *Nasturtium officinale*, *Phragmites australis*, *Potamogeton pectinatus*, *Typha angustifolia*, *Zannichellia palustris*.

- The German index: Trophic Index of Macrophytes (TIM), there are only eight species of HBS that have TIM indicator value: *Berula erecta*, *Elodea canadensis*, *Mentha aquatica*, *Nasturtium officinale*, *Potamogeton nodosus*, *Potamogeton pectinatus*, *Veronica beccabunga*, *Zannichellia palustris*.

All these species are included in the Euro Mediterranean indices, especially in the French index.

One of the most common species used in European countries' indices (MTR, TIM, IBMR, IMF and BMS) and taking place in HBS is *Nasturtium officinale*.

Based on IBMR index we have in HBS some species representing wide amplitude ( $Ei = 1$ ): *Mentha aquatica*, *Nasturtium officinale*, *Agrostis stolonifera*, *Helosciadium nodiflorum*, *Veronica beccabunga*, *Zannichellia palustris*. And some species representing a very limited amplitude ( $Ei = 3$ ): *Hygrohypnum luridum*, *Lemna gibba*, *Potamogeton nodosus*. Furthermore, some species indicating hypertrophic conditions (e.g. *Potamogeton pectinatus*, *Potamogeton nodosus*,  $Csi = 2-4$ ) and others indicating oligotrophic conditions (e.g. *Hygrohypnum luridum*,  $Csi = 19$ ).

Based on BMS index, species associated with high conductivity and nutrient enrichment ( $Qi = 1$ ) are: *Elodea canadensis*, *Phragmites australis*, *Arundo donax*.

IMF index reveals some species representing wide amplitude ( $E_i = 1$ ): *Epilobium hirsutum*, *Helosciadium nodiflorum*, *Scrophularia auriculata*. Species representing a very limited amplitude ( $E_i = 3$ ): *Equisetum ramosissimum*, *Mentha longifolia*, *Veronica beccabunga*, *Potamogeton pectinatus*, *Zannichellia palustris*. Some species indicating hypertrophic conditions (e.g. *Epilobium hirsutum*, *Helosciadium nodiflorum*, *Scrophularia auriculata*,  $C_{si} = 4$ ) and others indicating oligotrophic conditions (e.g. *Equisetum ramosissimum*, *Mentha longifolia*,  $C_{si} = 18$ ).

## Discussion

The most obvious difference between the three indicator systems is the number of included indicator taxa: IBMR (226), IMF (124), BMS (105), and TIM (49).

The IMF and the BMS have the fewest species in common (35 common taxa compared to 47 between IBMR and BMS and 68 between IBMR and IMF).

The allocation of the trophic values was based on empirical studies (correlation between species occurrence and impact parameters), literature data and expert opinion in TIM and IBMR. In BMS and IMF, the trophic values were determined only by empirical studies.

IBMR and BMS are moderately correlated ( $R^2=0.57$ ). The worst correlation occurs between IBMR and IMF ( $R^2=0.30$ ).

In France (IBMR), *Zannichellia palustris* and *Potamogeton pectinatus* have more eutrophic indicator values than in Spain (IMF) (Figure 2). *Zannichellia palustris* is commonly associated with nutrient-rich conditions (Vukov et al. 2018) as well as *Potamogeton pectinatus*. For instance, in Germany (Trophic Index of Macrophytes (TIM) (Schneider and Melzer 2003)) and Poland (Macrophyte Index for Rivers (MIR)) *Zannichellia palustris* and *Potamogeton pectinatus* are used as indicator of eutrophic conditions. However, in the UK (Mean Trophic Rank (MTR) (Dawson et al. 1999)), those species are seen to be tolerant of eutrophication, or cosmopolitan in their requirements (Table 2).

*Potamogeton nodosus*, *Amblystegium riparium* and *Lycopus europaeus* have more oligotrophic indicator values in Portugal (BMS) than in France (IBMR) and in Spain (IMF) (Figure 2).

In Poland (MIR), *Potamogeton nodosus* tends to be used to refer to eutrophic conditions. In Germany (TIM), it is used as an indicator of eutrophic to polytrophic conditions, which is consistent with the eutrophic BMS, IBMR and IMF indicator values. It is therefore likely that *Potamogeton nodosus* has a broader ecological amplitude. For instance, in Zambia (The Zambian Macrophyte Trophic Ranking scheme (ZMTR) (Kennedy et al. 2016)), this species is considered as ubiquitous species, occurring across from oligotrophic to eutrophic conditions (Table 3).

*Amblystegium riparium* is described as tolerant of eutrophication or cosmopolitan in its requirements. So, it is therefore likely that this species has a broader ecological amplitude.



**Table 2.** *Zannichellia palustris* and *Potamogeton pectinatus* indicator values in MTR, TIM, and MIR.

	<i>Zannichellia palustris</i>	<i>Potamogeton pectinatus</i>
Mean Trophic Rank (MTR) UK	STR = 2 tolerant of eutrophication or are cosmopolitan in their requirements.	STR = 1 tolerant of eutrophication or are cosmopolitan in their requirements.
Trophic Index of Macrophytes (TIM) Germany	IV = 2.93 meso-eutrophic (m-eu) – eutrophic (eu)	IV = 2.88 meso-eutrophic (m-eu) – eutrophic (eu)
Macrophyte Index for Rivers (MIR) Poland	L = 2 eutrophic	L=1 eutrophic

**Table 3.** *Potamogeton nodosus* and *Amblystegium riparium* indicator values in TIM, ZMTR, MTR, and MIR.

	<i>Potamogeton nodosus</i>	<i>Amblystegium riparium</i>
Trophic Index of Macrophytes (TIM) Germany	IV=3.1 eutrophic (eu) – eu-polytrophic (eu-p)	
The Zambian Macrophyte Trophic Ranking scheme (ZMTR) Zambia	ZTRSp=(3 U) ubiquitous species, occurring across trophic categories from oligotrophic to eutrophic	
Mean Trophic Rank (MTR) UK		STR = 1 tolerant of eutrophication or are cosmopolitan in their requirements
Macrophyte Index for Rivers (MIR) Poland	L = 3 eutrophic	

The apparent weak and moderate correlation and the difference of the included taxa and their indicator values from one index to another can be attributed to the hydromorphological characteristics of the Mediterranean rivers.

331 species are included in the Euro Mediterranean indices (IBMR, BMS and IMF) belonging to 98 families, 66 orders and 24 classes. The most diversified families are: Potamogetonaceae, Cyperaceae, Ranunculaceae, Amblystegiaceae, Typhaceae, Plantaginaceae, Characeae, Poaceae, Hydrocharitaceae, Apiaceae, Juncaceae (Figure 1). The most used genera are: Potamogeton (19 species), Ranunculus (19), Sparganium (9), Fissidens (8), Juncus (8), Carex (7), Callitriche (7), Chara (6), Equisetum (5), Montia (5) and Najas(5). These indices include some species of Chromista, Bacteria and Fungi (Table 4).

The comparison of the HBS elaborated list with the Euro-Mediterranean indices revealed the low level of similarity between HBS community species and the floristic reference of the French index (IBMR), the Portuguese index (IMF) and the Spanish index (BMS).

Furthermore, there is a limited number of HBS aquatic species (31 species), which is in agreement with previous research (Benamar and Maissour 2014).

The high level of aquatic species in France and the low-level of aquatic species in HBS compared to the Euro-Mediterranean countries can be ascribed to the climate transition from the temperate climate of central Europe to the arid climate of northern Africa (Giorgi et al. 2008). These Afro-Mediterranean conditions deeply affect stream flows (mixture of perennial and intermittent rivers) and the occurrence of aquatic species.

**Table 4.** List of Chromista, Bacteria and Fungi taxa used in Euro-Mediterranean indices.

kingdom	species	IMF	IBMR
Bacteria	Nostoc	+	+
	Oscillatoria	+	+
	Phormidium	+	+
	Sphaerotilus		+
Chromista	Cymbella	+	
	Leptomitius		+
	Melosira	+	+
	Tribonema	+	+
Fungi	Vaucheria	+	+
	Collema dichotomum		+
	Dermatocarpon luridum		+

These results demonstrate the inadequacy of the trophic indices approach especially with the HBS conditions and in general in the Afro-Mediterranean region, and thus the need for the development of an index based on biotic indices approach taking into consideration also the riparian species.

The Biotic indices approach, which is originally developed by Karr and Dudley (1981), is a widely used method for evaluating anthropogenic pressures on aquatic and wetland ecosystems: Floristic Quality Assessment Index (FQAI) (Lopez and Siobhan Fennessy 2002), Integrity Biotic Index (IBI) (Miller et al. 2006), Iberian Multi metric Plant Index (IMPI) (Ferreira et al. 2005), Index of Plant Community Integrity (IPCI) (DeKeyser et al. 2003), Index of biotic integrity in Itanhaém (MIBI-ITA) (Umetsu et al. 2018), Plant Index of Biotic Integrity (PIBI) (Simon et al. 2001), Plant-based index of biotic integrity (PIBI) (PIBI(M)) (Moges et al. 2016), Riparian Forest Quality index (QBR) (Munné et al. 2003), Riparian Quality Index (RQI) (Del Tanago et al. 2006; González del Tánago and García de Jalón 2006), Vegetation Index of Biotic Integrity (VIBI) (Mack 2007), and Vegetation-based index of biotic integrity (VIBI(Y)) (Yang et al. 2018).

Among the potential characteristics of the aquatic vegetation (candidate metrics) that can be responsive to disturbance in HBS are: diversity, species habitat, life cycle, life form, nutritional resources, riparian structure, and species tolerance (Table 5).

Future work will involve the selection of the reference sites. This is because the reference sites provide the baseline information to detect the deviation of a metric from a natural or least-disturbed condition. And the selection of suitable metrics in our context. So, we need to evaluate the ability of every potential candidate metric in terms of its ability to distinguish reference (undisturbed or least-disturbed) from impaired (moderately or heavily disturbed) sites. Only the metrics showing significant difference between reference and impaired sites will be selected as the IBI-HBS metrics (Yang et al. 2018). The next step is to score the selected core metrics.

**Table 5.** Potential candidate and core metrics in IBI- HBS.

Candidate metrics	Expected response to decreasing quality	River and wetland indices
<b>Diversity</b>		
Species richness	Decrease	FQAI, PIBI, IPCI, MIBI-ITA
<b>Species habitat</b>		
% Endemic species	Decrease	FQAI
% Native species	Increase	PIBI(M)
% Exotic species	Increase	PIBI(M)
<b>Life cycle</b>		
% Annual species	Decrease	IMPI, IBI, VIBI,
% Perennial species	Increase	VIBI, PIBI, IPCI, VIBI(Y)
<b>Life form</b>		
% Terrestrial species		
% Hygrophyte species		RQI, PIBI
% Helophyte (emergent species)+ hydrophyte species (floating-leaved, free-floating, and submerged species)	Decrease	IMPI, VIBI, PIBI; MIBI-ITA
<b>Nutritional resources</b>		
% Ruderal species	Increase	IMPI
% Nitrophyllous species	Increase	IMPI, RQI
<b>Riparian structure</b>		
% Woody species richness (trees, shrubs, woody climbers)	Variable	IMPI, IBI, RQI, PIBI(M)
<b>Species tolerance</b>		
Tolerant species richness	Increase	PIBI(M), VIBI(Y)
Sensitive species richness	Decrease	PIBI(M)

## Conclusion

We have confirmed that the ecological amplitude and species optima vary between Mediterranean ecoregions, and that indicator taxa differ between countries.

It was found that the trophic indices of the Euro Mediterranean rivers can't be applied easily to the Afro- Mediterranean rivers, particularly in Morocco (HBS), and we don't have a good opportunity to enrich the list of indicative species due to the limited number of species recognized as bioindicators (23 species) and the limited number of aquatic species. So, it seems more appropriate to develop an index based on a biotic-integrity approach.

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## Supplementary material I

### Checklist of hydrological basin of Sebou macrophytes

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Data type: occurrences

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