



## Aquatic bryophytes as ecological indicators of the water quality status in the Tiber River basin (Italy)

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### ARTICLE INFO

#### Article history:

Received 29 March 2011

Received in revised form 31 July 2011

Accepted 12 August 2011

#### Keywords:

Aquatic bryophytes

Bioindicators

Ecological responses

Fuzzy set

Watercourses

Water quality

### ABSTRACT

A survey of 18 watercourses of the Tiber River basin was carried out to define the ecological niche breadth of some aquatic bryophyte species in relation to environmental factors. Aquatic bryophytes were sampled and water environmental parameters were measured at 99 stations distributed along the catchment (from the headwater regions to the downstream reaches). The datasets of the collected species and environmental data were analyzed by using a multivariate statistical analysis (PCA biplot). Ecological responses of the recorded aquatic bryophytes were obtained using a fuzzy set approach, and were compared with data from literature. The results show that the presence of the aquatic bryophytes in watercourses is affected negatively by the reduction of water velocity, clearness, substratum size and the worsening quality of the water physico-chemical status. In fact, aquatic bryophytes show a general preference for stations characterized by medium-large granulometry, and fast-flowing, clear, oxygenated (mean value 9.2 mg/l), cool waters (mean value 15.0 °C), with low loads of nutrients, particularly ammonia (mean value 0.10 mg/l) and phosphates (mean value 0.09 mg/l). However, ecological responses reveal different patterns in the distribution of aquatic bryophyte species mainly in relation to water physico-chemical parameters (e.g. temperature, conductivity, ammonia, phosphates). E.g. *Palustriella commutata* var. *commutata*, *Cratoneuron filicinum*, *Fissidens viridulus* and *Cinclidotus aquaticus* show high preference for clear, turbulent and fast-flowing waters, with temperature below 12 °C, conductivity below 300 μS/cm, and concentrations about 0.01 mg/l for phosphates, not exceeding 0.10 mg/l for ammonium ions and 0.90 mg/l for nitrates. *Leptodictyum riparium* and *Riccia fluitans* are for their part more linked to turbid and slow waters affected by eutrophication, showing optimum values for about 0.30 mg/l for ammonia concentration, 0.90 mg/l for nitrates and 0.11 and 0.22 mg/l for phosphates respectively. Conversely, *Fontinalis antipyretica* is not closely related to specific conditions, showing wide ecological ranges for most of the analyzed environmental factors. This paper has evaluated and discussed the possible use of sampled species as bioindicators for biomonitoring of the water quality.

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### 1. Introduction

The Water Framework Directive (WFD) (2000/60/EC) gives great importance to biological indicators since they are more reliable than physico-chemical analysis for defining the ecological and quality status of the aquatic ecosystems. Therefore, the application of the Directive suggests the use of new bioindicators for the assessment of water quality, in addition to the already widely used benthic macroinvertebrate communities. Thus, the WFD has enlarged the use of possible bioindicators to fishes, diatoms and

macrophyte communities (Mancini, 2003; Mancini and Andreani, 2008). Specifically, the study of macrophytes consists of an analysis of all aquatic plants visible to the naked eye, including phanerogams, pteridophytes, macroalgae and bryophytes growing in water. Although there are different macrophyte components, plant research on the assessment of water quality is mainly carried out through the analysis of phanerogamic macrophytes.

In the biomonitoring of water quality, aquatic bryophytes can be used as bioindicators for their bioaccumulation ability or for the dimension of their ecological niches. The majority of studies in Europe deal with the use of aquatic bryophytes as bioaccumulators. Some aquatic species [e.g. *Fontinalis antipyretica* Hedw., *Platyhypnidium riparioides* (Hedw.) Dixon] can efficiently bioaccumulate contaminants (e.g. heavy metals and radionuclides) and their concentrations in bryophyte tissues can be then used as biomarkers of water pollution (e.g. Claveri et al., 1995; Allegrini et al., 1998; Cenci,

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2000; Nimis et al., 2002; Van Hullebusch et al., 2003). On the other hand, the body of literature analyzing the ecology of bryophytes for biomonitoring is underdeveloped.

The composition of bryophyte communities and the presence/absence of pollution-sensitive or tolerant species can be used and analyzed to assess the water quality, especially to estimate the trophic water pollution (Vanderpoorten and Palm, 1998, 2001; Tremp, 1999; Vanderpoorten and Klein, 2000). This approach requires thorough knowledge of the bryophyte ecology in order to correlate the presence and abundance of species to the various environmental factors' conditions. The main environmental factors affecting the assemblage and distribution of aquatic bryophytes are substratum morphology, current velocity, level and transparency of water (Muotka and Virtanen, 1995; Suren, 1996; Englund et al., 1997; Vanderpoorten and Klein, 1999; Scarlett and O'Hare, 2006).

Some physico-chemical parameters of water are also important, such as pH, temperature, conductivity and nutrient load (Empain, 1978; Vrhosek et al., 1984; Arts, 1990; Düll, 1991; Thiebaut et al., 1998; Tremp, 1999; Vanderpoorten, 1999a,b; Vanderpoorten and Durwael, 1999; Vanderpoorten et al., 1999, 2000; Allegrini, 2000).

The majority of these ecological studies are based on a qualitative approach or separate statistical analyses of physico-chemical data and bryophyte diversity data; thus they cannot be used to quantify the relationship between bryophyte species and environmental factors. Only few studies assess quantitatively the correlation between aquatic bryophytes and the trophic status of water (Haury et al., 1996; Thiebaut et al., 1998; Vanderpoorten and Palm, 1998, 2001; Vanderpoorten and Durwael, 1999; Vanderpoorten et al., 2000; AFNOR, 2003).

Thus, in this paper, the correlation between aquatic bryophyte species and main environmental factors were statistically analyzed in order to:

- (i) Increase data on the ecology of aquatic bryophytes;
- (ii) define quantitatively the ecological responses of each bryophyte species to the analyzed environmental factors (temperature, conductivity, dissolved oxygen, pH, concentrations of ammonia, nitrates, phosphates, turbidity, current velocity and substratum morphology), estimating their optimum ecological values and ranges of compatibility to these factors;
- (iii) identify a set of bryophyte species that can be used as reliable bioindicators to evaluate water quality.

This work is part of a more comprehensive research program aimed at analyzing the relations between the macrophyte component and water quality within the Tiber River basin, which has been identified by governmental institutions as a pilot basin for ecological studies (Bagnini et al., 2005).

## 2. Materials and methods

### 2.1. Study area

This research was conducted in the hydrographical basin of the Tiber including both the Tiber River and its main tributaries (Fig. 1). The Tiber basin extends over an area of more than 17,000 km<sup>2</sup> and it is the widest in the Italian Peninsula. This catchment covers an extensive part of Central Italy (mainly Umbria and Latium regions) and flows through important cities (Rome, Perugia, Rieti and Terni).

Biogeographically, the Tiber basin area is part of the *Mediterranean district of the middle-Tyrrhenian sector* (Zunino and Zullini, 2004). From a hydromorphological perspective, it is possible to define three distinct hydro-morphological sectors in the main watercourses of the basin (Tiber, Aniene and Nera). The

upper sector starts in the mountainous belt, where watercourses are generally characterized by steep slopes, rocky substratum and a torrential character. A middle sector is characterized by pebbly-gravelly substratum and variable slopes that affect the behavior of water flow from fast flowing, turbulent shallow waters to medium laminar flowing water (usually deeper tracts). Finally, there is a lower sector with typical fluvial regime, slow-flowing waters, and calcareous sandy-muddy substrata (IRSA, 1978).

The main lithotype of the basin is calcareous. The bioclimate of the upper and middle sectors is Temperate (*sensu Rivas Martinez, 1993*) (Biondi and Baldoni, 1994) and Mediterranean in the lower sector (Blasi, 1994).

The natural vegetation of the basin is generally preserved along the rivers in the mountainous and sub-mountainous zones. On the other hand, the bottom of valleys and plains, with some exceptions, have lost the main part of their original fluvial ecosystem characteristics, due principally to agricultural activities and urbanization. These land uses are the main cause of the general mineralization and eutrophication of waters in the lower sector (Casini and Giussani, 2006).

### 2.2. Biological sampling

The aquatic macrophytes, including bryophytes, were sampled at 99 stations. The sampling stations were selected along 18 permanent watercourses of the Tiber River basin, where the water level does not go below the minimum flow status.

The sampling activity was conducted in June and October 2007–2008. At each sampling station all aquatic macrophyte species were sampled and their relative abundances were estimated (as percentage cover within approximately a 50 m × 2 m stretch of watercourse). Specifically, bryophyte samples were taken mainly from boulders, cobbles, or tree stumps that were in submerged conditions for most of the year.

Aquatic bryophytes were identified using Cortini Pedrotti (2001, 2005) for the mosses and Paton (1999) for the liverworts, while the nomenclature was updated using Aleffi et al. (2008). For the taxonomical determination, the morphological features were analyzed using both optical microscope (Leica DM RB) and stereoscope (Olympus SZX16). Voucher specimens are deposited at the *Herbarium* of the University Roma Tre (URT) (Thiers, continuously updated).

### 2.3. Environmental factors sampling

The physico-chemical parameters of water were registered at each sample stations, concurrently with biological sampling. Specifically, data on water temperature (*T*), conductivity (*C*), dissolved oxygen (*O*<sub>2</sub>), pH, were recorded *in situ* by probes for immersion (WTW Multi340i/SET); while ammonia (NH<sub>4</sub><sup>+</sup>), nitrates (NO<sub>3</sub><sup>-</sup>), and phosphates concentrations, were recorded in laboratory by a spectrophotometer (WTW Photometer MPM 3000).

Moreover, data on water turbidity, current velocity combined with flow water type, and substratum morphology were estimated by direct field observations. These data were converted into numerical data, according to a scale ranging from 0 to 2 for turbidity (0 = clear, 1 = quite clear, and 2 = turbid), from 0 to 8 for the current velocity (0 = very slow, 2 = slow, 4 = mean and laminar, 6 = mean and turbulent, and 8 = fast and turbulent) and from 1 to 5 for the substratum morphology, based on prevalent type of riverbed substratum (1 = silt, 2 = sand, 3 = gravel, 4 = cobbles, and 5 = boulders). Therefore, values closer to zero indicate fine substratum, clearer and slower-flowing conditions, whereas higher values represent progressively larger granulometry, more turbid and faster-turbulent flowing waters.

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