



# Benthic freshwater cyanobacteria as indicators of anthropogenic pressures



Laura Monteagudo\*, José Luis Moreno

University of Castilla-La Mancha, Regional Center of Water Research (CREA), Crta. de Las Peñas km. 3, Albacete 02071, Spain

## ARTICLE INFO

### Article history:

Received 23 October 2015

Received in revised form 15 March 2016

Accepted 17 March 2016

Available online 25 April 2016

### Keywords:

Water quality

Biological indicators

Cyanobacteria

Algae

Periphyton

Eutrophication

## ABSTRACT

Freshwater cyanobacteria are hardly used as biological indicators of anthropogenic pressures, possibly for two main reasons: (a) their response to anthropogenic pressures is often poorly known; (b) reliably identifying cyanobacteria species is a challenge for technicians and researchers. We assessed the usefulness of cyanobacteria species as biological indicators of two human stressors: the high orthophosphate input from urban wastewaters; the high nitrate concentration produced by agricultural land use. We analysed variation in benthic cyanobacterial assemblages at 85 sites in South-Central Spain as a response to eight environmental variables: pH, conductivity, temperature, altitude, nitrate, orthophosphate, irrigation land use and non-irrigation land use.

Results revealed that conductivity was the main environmental factor that contributed to differences between assemblages. Orthophosphate was more influential for community composition than nitrate. Changes in species composition related to human pressures suggested that some cyanobacteria species (e.g. *Nostoc verrucosum*, *Phormidium autumnale*, *Plectonema tomasinianum*, *Rivularia haematites*, *Tolythrix distorta*) could be useful tools for the bioindication of anthropogenic pressures; while others species provide more information about natural physicochemical reference conditions (*Nostoc caeruleum*, *Phormidium fonticola*). Further research into cyanobacteria and macroalgae assemblages in different impacted scenarios could help improve macrophyte indices.

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## 1. Introduction

Since the Water Framework Directive (WFD) came into force, EU Member States are required to achieve a 'good status' in all surface and groundwater bodies, which implies achieving a 'good chemical' and 'good ecological' status. Regarding ecological status, Member States shall establish monitoring systems to estimate the values of the biological quality elements specified per surface water category (WFD – Annexe V – 1.4.1.). This requirement increased the use of metrics based on different groups of organisms (e.g. invertebrates, diatoms or macrophytes) to assess ecological status. In general, indices based on macrophytes are less popular than those based on macroinvertebrates or diatoms (Ferreira et al., 2005). Demars et al. (2012) reviewed several macrophyte indices and highlighted the poor accuracy observed in this metrics when applied to different regions. Some criticisms about macrophyte indices indicate that they are based on riverine plants prior to exclusive aquatic taxa and the usual exclusion of benthic macroalgae,

despite these organisms being closely linked to water properties. Regarding cyanobacteria, such discrimination could be due to a poor understanding of their response to anthropogenic pressures (Thacker and Paul, 2001), and technicians and researchers' difficulty in reliably identifying species (Marquardt and Palinska, 2007). Despite these difficulties, studies worldwide have related cyanobacteria to several human pressures responsible for eutrophication in freshwater ecosystems, such as agricultural loadings, urban pollution or industrial discharges (e.g. Johansson, 1982; Jafari and Gunale, 2006; Parikh et al., 2006). Although cyanobacteria have been usually included in studies that have related algal communities to environmental factors (e.g. Potapova et al., 2005; Dell'Uomo and Torrisi, 2009), some physiological features have established remarkable differences in the way cyanobacteria are influenced by nutrients. Unlike other algae, cyanobacteria are able to fix atmospheric nitrogen, which allows them to grow at low rates of dissolved nitrogen compounds. Some authors have suggested that this characteristic may diminish their dependence on nitrogen availability in the water column (e.g. Larkum et al., 1988). In addition, heterocysts allow some cyanobacteria to fix atmospheric nitrogen (N<sub>2</sub>) under aerobic conditions, while N<sub>2</sub> fixation is limited to anaerobic and (or) dark conditions in the

\* Corresponding author. Tel.: +34 967599200 Ext. 2576.

E-mail address: [laura.monteagudo@gmail.com](mailto:laura.monteagudo@gmail.com) (L. Monteagudo).

case of non-heterocystous cyanobacteria (Potts, 1979; Lee, 2008). Loza et al. (2014) suggested that this ecophysiological advantage could explain not only the dominance of heterocystous species at low levels of combined nitrogen, but also the preference of non-heterocystous species for high levels of these compounds. As for phosphorus, certain cyanobacteria, such as *Rivularia* sp., are able to live at phosphorus limitation because of phosphatase activity, which is a good indicator of oligotrophic conditions (Mateo et al., 2010).

All these facts evidence that cyanobacteria are a large diverse group of organisms which includes freshwater species that grow at unimpaired and impaired sites. Thus some of these species are expected to be suitable indicators to account for the development of biological indices of water quality. In order to identify them, it is essential to examine the relationship between the cyanobacteria community and the surrounding environmental conditions, and define the environmental range of species indicative of good ecological conditions. Hence the main objectives of this study were to: (1) analyse which variables determine the benthic cyanobacteria community in the study area; (2) examine the differences between heterocystous and non-heterocystous species; (3) identify which species of cyanobacteria in the study area are suitable as indicators of human pressures.

## 2. Material and methods

### 2.1. Study area and sampling design

This study was carried out in Castilla-La Mancha (South-Central Spain), a region chiefly used for agriculture. As semi-arid climatic conditions predominate in the study area, irrigation is needed for high-yielding crops. Thus 6.26% of agriculture is conducted under irrigation (Spanish Survey of Surfaces and Crop Yields (ESYRCE), 2013), usually located on the banks of streams and rivers.

Sites were selected to cover the most complete gradient of environmental conditions, such as lithology and altitude (Fig. 1). Three main lithological types were defined: calcareous (Cretacic and Jurassic limestones), mixed (Quaternary sedimentary valleys) and siliceous (granites, slates and shales). Reaches were waded in an upstream zigzag pattern and sampling was conducted in all habitat types. Macroscopic cyanobacteria thalli that grew submerged or in the splash zone were collected by hand and fixed with 4% formalin. Species determination was carried out in the laboratory under a Leica M165C stereoscope and a light microscope Olympus BX50. Glycerine-gelatine was used to make permanent slides. In this study, we considered only those species that predominated in macroscopic colonies.

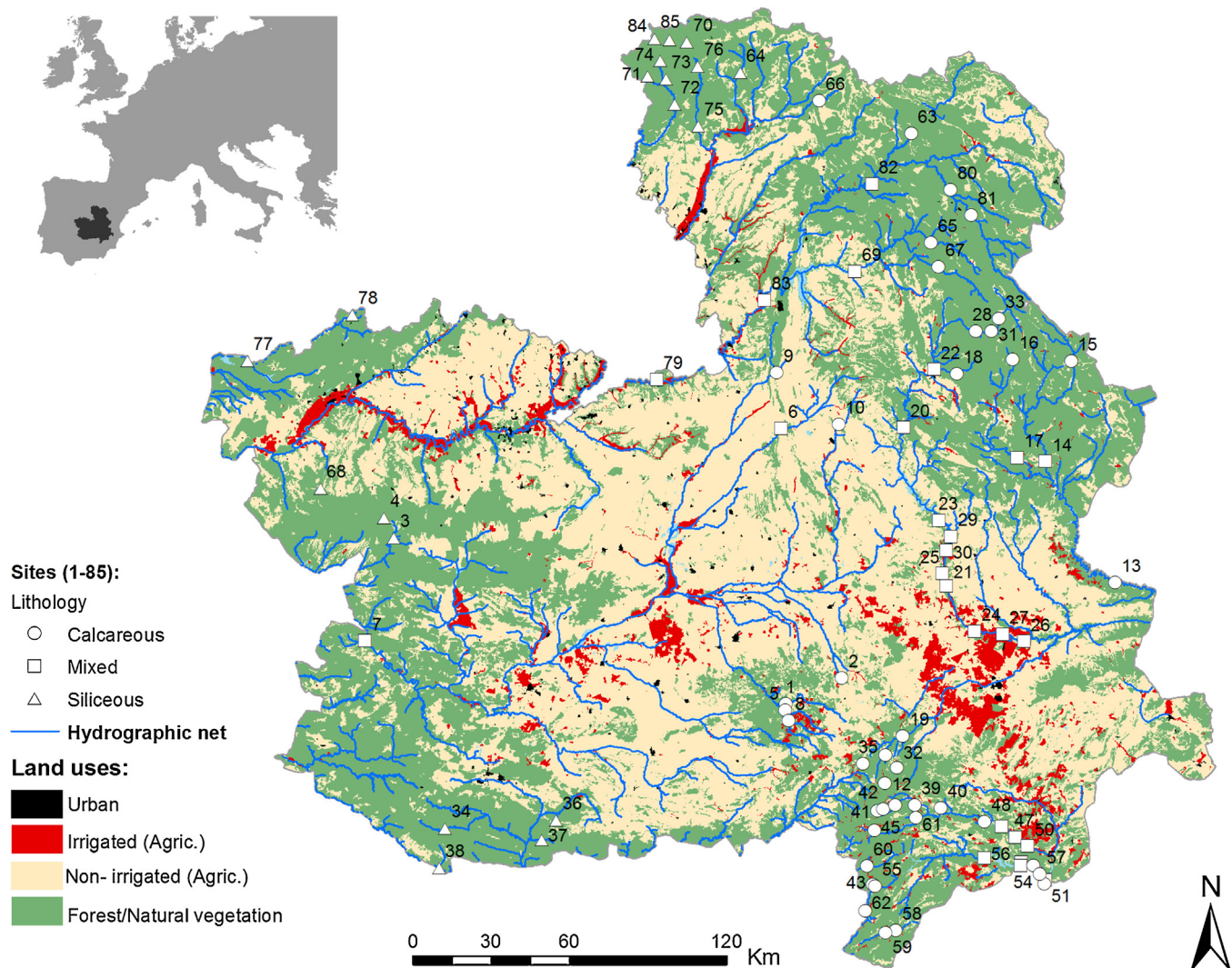


Fig. 1. Study area showing land uses and location of sites. Site symbols correspond to three lithology categories: calcareous (circles), mixed (squares) and siliceous (triangles).

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