# Assessing the seasonal dynamics of zooplankton in a recreational marina of the northwest of Portugal through multi-marker DNA metabarcoding

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#### Abstract

The monitoring of larvae in plankton samples in recreational marinas, ports, or the close vicinities, may provide key information about non-indigenous species (NIS) introduction status or detect their presence at an earlier stage. DNA metabarcoding is a powerful method to assess biodiversity but its efficiency is dependent on the methodologies employed along the analytical chain, namely the targeted genetic markers. Therefore, we aim to analyze the ability of two genetic markers - the mitochondrial cytochrome C oxidase subunit I gene (COI) and the variable region V4 of the nuclear small subunit ribosomal gene (18S) - to assess the seasonal dynamics of zooplankton communities, including NIS, through DNA metabarcoding, and further to compare it with a compiled list of zooplanktonic species identified via morphology that occur in the Lima Estuary. To this end, we sampled zooplankton communities, spanning three consecutive seasons (spring, autumn, and winter 2020/2021) in three sampling points in the recreational marina of Viana do Castelo, located in the Lima Estuary, North of Portugal. Globally, metabarcoding recovered 157 species belonging to 19 phyla, with a dominance of Annelida, Crustacea, and Mollusca that represent 64.3% of all species. Even though our sampling area poorly represents the Lima Estuary, this approach allowed the detection of 6 NIS, which were not yet reported by traditional methods of identification. The composition of the zooplankton communities recovered was greatly affected by the markers employed, and to a lesser extent by the season. Overall 18S detected more species and fewer species were recovered in winter. NIS were predominantly detected during spring and autumn, but only 5.1% of all species were detected in all seasons. This study shows the DNA metabarcoding efficiency in assessing the dynamics of zooplankton communities and highlights the interest in using a multi-marker and seasonal approach to species detection, particularly to target NIS.

## Keywords

DNA metabarcoding, multi-marker approach, zooplankton, non-indigenous species, inter-seasonal dynamics

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# **Conflicts of interest**