

# Detection and monitoring of invertebrate non-indigenous species through DNA metabarcoding in a recreational marina of the Northwest of Portugal

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## Abstract

In marine and coastal ecosystems, the proliferation of non-indigenous species (NIS) is among the top causes of biodiversity loss. Thus, it is essential to prevent the entrance and proliferation of NIS by detecting their presence as early as possible at the most susceptible locations, such as ports and marinas. Molecular techniques, like DNA metabarcoding, may be faster and more effective in the early detection of NIS. Its high sensitivity allows the detection of species at any stage of their life cycle and when present at very low densities. However, the detection power can be affected by the methodologies employed through the DNA metabarcoding analytical chain. This study aimed to assess the seasonal effects, sampling method (hard and artificial substrates, zooplankton and eDNA) and genetic marker (COI and 18S) in the recovery of NIS through DNA metabarcoding. Sampling was conducted in 3 seasons (spring, autumn, and winter of 2020/2021) in the “Porto Atlântico” marina, which is located near a major commercial port of the North of Portugal (Leixões). Overall, 626 species were identified through DNA metabarcoding, with the dominance of Crustacea, Ascidiacea and Mollusca. The sampling method and season significantly affected the marine invertebrate species recorded, for both molecular markers, with a high proportion of species detected exclusively using one method (7 to 20%). Overall, 31 NIS were detected, 7 of which are considered invasive and 6 are presumed new records in mainland Portugal. Zooplankton sampling and winter were, respectively, the method and the season where the highest number of NIS was detected, but the maximum number was only attained when pooling the results from all sampling methods and seasons. Our study highlights that different sampling methods and genetic markers, over different seasons, are needed to detect NIS in marine habitats, since the number of species shared by these factors is low.

## **Keywords**

DNA metabarcoding, Marine and coastal ecosystems, Marine invertebrates, non-indigenous species (NIS), Northwest Portugal

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## **Conflicts of interest**