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PRIMARY RESEARCH PAPER

Ecological interplay between diatoms and plants in Mediterranean temporary ponds habitat

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Abstract In the Mediterranean region, temporary ponds (MTPs) are priority habitats hosting rare and endemic species significantly threatened globally. This research represents the first characterization of diatom communities of MTPs in Central Italy, coupling phytosociological surveys and physical and chemical characterization of water and sediments. We selected 3 areas and 13 ponds with different levels of protection. A total of 89 diatom species and 52 plant species were recorded. No significant differences in taxonomic and functional α diversity were observed among sites. Water conductivity was significantly associated with taxonomic richness. Functional α diversity showed significant correlation with several environmental variables, including conductivity, pH, sediment density, dissolved oxygen. Both taxonomic and functional total β diversity and turnover differed significantly among sites, in particular between Foglino and Castelporziano sites. RDA analysis based on vegetation as variable showed that the measured environmental variables explained 51.45% of the diatom community variation, providing the first evidence of ecological correspondence between diatom and plant communities in MTPs. Our results underpin the potential role of diatoms as bioindicators in MTPs.

Keywords Ephemeral waters · Habitat directive · Aquatic plants · Macrophyte ecology · Natura 2000 network

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Introduction

Ponds are small (< 5 ha) and shallow (< 5 m) water bodies, with < 30% emergent vegetation (Bolpagni et al., 2019; Olmo et al., 2022; Richardson et al., 2022). According to the European Pond Conservation Network's "Pond Manifesto", ponds are recognized as distinct, valuable freshwater systems whose conservation requires specific attention (E.P.C.N., 2008). They represent the most abundant freshwater ecosystem on Earth (Meerhoff and Beklioğlu, 2024), recognized as important habitats in landscapes throughout the world (Fehlinger et al., 2021) and playing a significant role in the provision of ecosystem services (Fu et al., 2018; Krivstov et al., 2022). Temporary ponds differ from permanent ponds due to hydroperiod: a wet or inundate phase is followed by a prolonged dry period whose duration can change every year (Florencio et al., 2020).

In the Mediterranean region, temporary ponds (hereafter MTPs) are threatened ecosystems, and currently show an "unfavorable-bad" conservation status (EEA 2020), despite receiving protection through the EU Habitats Directive (92/43/CEE) (Coccia et al., 2024). However, while the Habitats Directive recognize Mediterranean temporary waters as priority habitats, it still provides limited protection for small water bodies as a whole (Della Bella et al., 2005). As reported by Bagella (2023), recognizing MTPs as priority habitats (Code 3170*) was crucial for the extremely rare and isolated organisms they host, including several plant communities belonging to the *Isoëto-Nanojuncetea* class. MTPs also often contain rare, endemic and/or Red Data List species and are therefore likely to disappear, when these ponds are degraded, due to the prevailing threats (Zacharias & Zamparas, 2010). In the last decades climate change has been a particular challenge for ponds (Segura & Palomar, 2023). Droughts may represent a strong environmental constraint that selects species with traits related to drought tolerance (Cunillera-Montcusí et al., 2020). Consequently, MTPs have recently displayed strong interannual variability that affects biodiversity (e.g., Florencio et al. 2020; Caria et al. 2021).

As described above, MTPs can host a large number of species, in particular ephemeral plant communities linked to temporarily submerged soils (Carta, 2016). Based on the environmental conditions, mainly related to the duration of flooding and pond morphology, different types of plant communities can be found, which are seasonally differentiated Bagella & Caria, 2012; Tomaselli et al., 2020; Caria et al., 2021;). In deeper ponds, where water persists for longer periods, communities and species well adapted to aquatic environments can still be observed in spring, and macrophytes may occur in the deepest zones. During the desiccation phase, two main groups of plant communities are typically present. The first group includes associations mostly characterized by small plants (also known as dwarf species, or microphytes) with early flowering; The second group consists of plant communities dominated by therophytes, generally exhibiting a prostrate or prostrate-ascending growth form and a later (summer) phenology. These communities are associated with habitats that remain submerged for longer periods and are characterized by a very rapid desiccation dynamics (Tomaselli et al., 2020).

In addition to plants, numerous algal species, such as diatoms, also inhabit MTPs. Diatoms are ubiquitous unicellular eukaryotic brown algae (Soininen & Teittinen, 2019), widely recognized as important primary producers at the base of food webs and source of gen (Letáková et al., 2018; B-Béres et al., 2023). Diatoms are one of the most used biological quality elements (BQEs) in freshwater monitoring, adopted by the European Water Framework Directive 2000/60/EC (WFD), focusing on their advantages as bioindicators (Masouras et al., 2021). The analysis of the diatom assemblage structure, species' autecology, and biological traits allows to characterize a specific water body from an ecological and biological point of view (Taurozzi et al., 2023). However, while pollution and water stress can have pronounced and well-documented effects on many bioindicators, such as macroinvertebrates (Della Bella et al., 2005; Wheeler et al., 2020; Strungaru et al., 2021), only few studies have addressed this issue using diatoms as the primary tool for assessing the ecological status of MTPs (Taurozzi et al., 2024a). While diatoms are commonly used as bioindicators in rivers and lakes, research on benthic diatom assemblages in wetland environments is surprisingly scarce, especially in Italy, where only a few studies, such as that of Della Bella et al. (2007), have examined these ecosystems.

Moreover, their β diversity patterns are not well documented especially at intermediate spatial scales, such as within and between MTPs (Jyrkänkallio-Mikkola et al., 2016). This diversity level has been shown to be strictly related with environmental heterogeneity, productivity and niche-based mechanisms, in many ecosystems (e.g., Tuomisto, 2010) as well as in freshwaters (e.g., Ziorza-Almeida et al., 2017). Significant β diversity patterns may have important implications for biomonitoring. For instance, Jyrkänkallio-Mikkola et al. (2016) showed that variation in diatom communities was significantly related to the general physical habitat structure and conservation status.

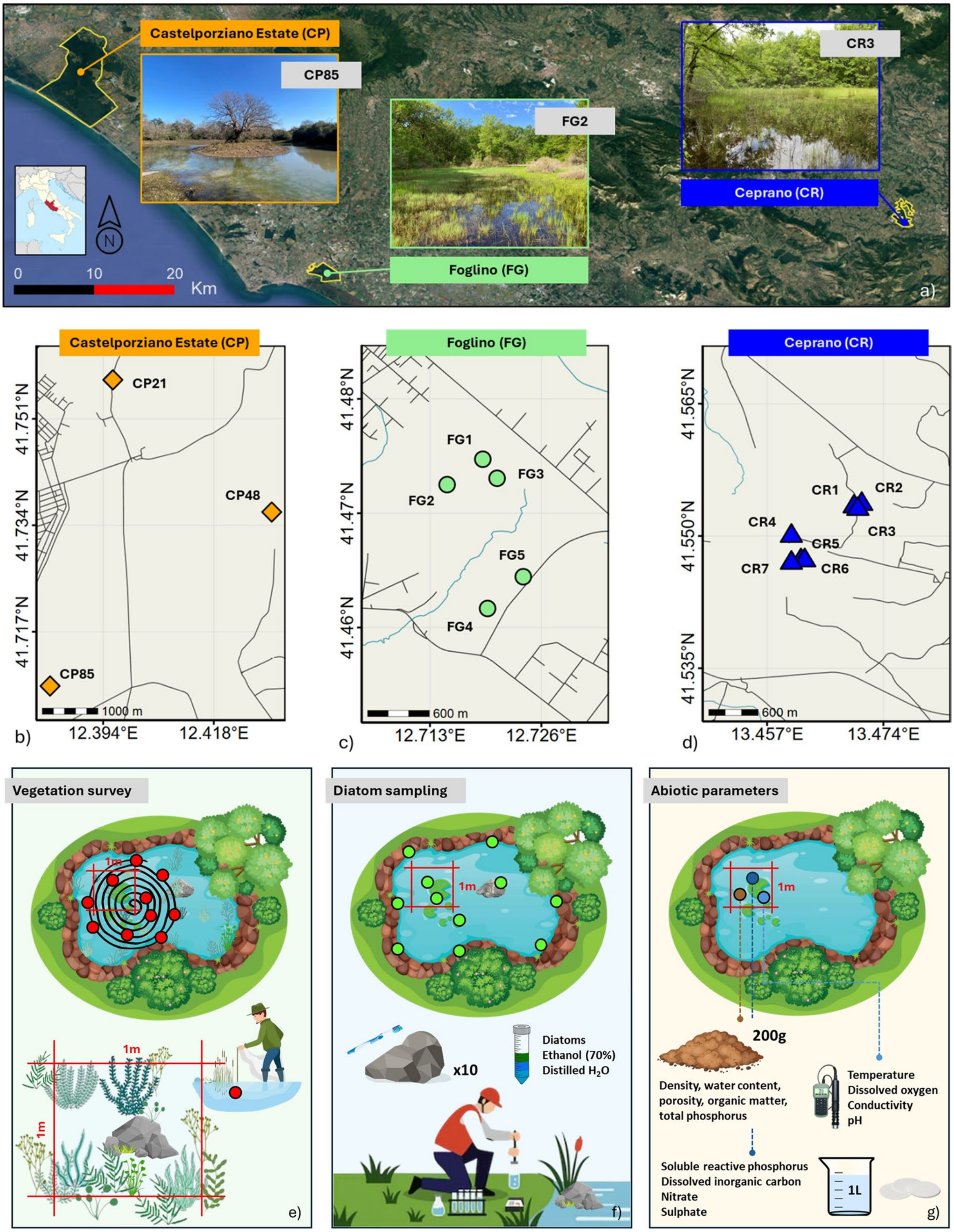
Plants are excellent biological indicators for assessing the conservation status of MTPs. The monitoring of plant communities belonging to MTPs requires samplings at least twice a year (early spring and summer, e.g., Bagella et al., 2016) and, based on different approaches, the monitoring programmes require a strong effort in terms of time, field activities and laboratory identifications. In contrast, diatoms can be sampled whenever ponds are full of water, using simpler sampling and identification methods (Taurozzi et al., 2024a). Despite their frequent use as bioindicators in freshwater ecosystems, the ecological patterns of diatom communities in lowland Mediterranean ponds remain poorly known (Della Bella & Mancini, 2009). Understanding the ecological relationship between plants and diatoms in MTPs could improve biomonitoring strategies (Jyrkänkallio-Mikkola et al., 2016), providing new insights for more effective and efficient conservation plans.

In this context, the main aim of this study is to evaluate the effectiveness of diatoms as bioindicators in MTPs, with a specific focus on the cross-taxon correspondence with plant communities characterizing Habitat 3170*. In particular, this research provides, to the best of our knowledge, the first characterization of diatom communities in MTPs, using a combined approach integrating physical and chemical parameters with phytosociological surveys to describe their taxonomic and functional diversity. Considering the high variability of MTPs in Lazio (Filippino, 2018; Bazzanti et al., 2000), we also explore how environmental conditions and protection measures influence diatom and plant communities across different sites. In particular, we hypothesize the following: (a) Diatom diversity is expected to reflect environmental gradients in MTPs; (b) There is expected to be significant correspondence between diatom and plant community composition, supporting the use of diatoms as complementary bioindicators; (c) Integrating diatom and plant data could provide a more complete and efficient tool for biomonitoring MTPs under the EU Habitats Directive.

Materials and methods

Study area

There are some published studies that have carried out floristic surveys in the sites where are the MTPs we investigated (Lucchese, 1990; Anzalone et al., 1997; Filibeck & Lattanzi, 2008; Minutillo et al., 2014; Lattanzi et al., 2021). However, although it has long been known that temporary ponds are a typical community in lowland forests dominated by *Quercus cerris* L. and *Quercus frainetto* Ten. belonging to the *Mespilo gemanicae-Quercetum frainetto* association (Blasi et al. 2002), few vegetation assessments are available for Lazio region (e.g., Stanisci et al., 2001), and MTPs were studied in other Italian regions (Umbria—Gigante et al., 2007; Sardinia—Bagella & Caria, 2012; Apulia—Tomaselli et al., 2020). In order to improve this knowledge, phytosociological and diatomic investigations were carried out in three sites and 15 MTPs in Lazio, mainly in the southern part (Fig. 1a). In all the sites, only 3170* MTPs were sampled. The classification of these ponds as Habitat 3170* was based on a preliminary survey conducted in 2022. This survey verified the coherence of the ecological conditions (temporary ponds in oligotrophic conditions), and the presence of the characteristic species reported in the European interpretation manual (EUR 28) and the Italian interpretation manual. The classification has been confirmed by the presence of diagnostic plant species, including *Isoëtes histrix* Bory, *Isoëtes longissima* Greuter, and *Damasonium alisma* (Mill.) A. Braun. Other species recorded represent typical associated flora of temporary ponds (e.g., *Callitriche spp., Illecebrum verticillatum* L.). The habitat typology was validated according to the criteria outlined in the EU Habitats Directive (92/43/CEE) and the relevant literature on Mediterranean Temporary Ponds in Italy (Della Bella


Castelporziano Estate (CP)
CP85
FG2
Foglino (FG)
CR3
Ceprano (CR)
N
0
10
20
Km
a)
Castelporziano Estate (CP)
CP21
CP48
CP85
41.751°N
41.734°N
41.717°N
1000 m
12.394°E
12.418°E
b)
Foglino (FG)
FG1
FG2
FG3
FG4
FG5
41.48°N
41.47°N
41.46°N
600 m
12.713°E
12.726°E
c)
Ceprano (CR)
CR1
CR2
CR3
CR4
CR5
CR6
CR7
41.565°N
41.550°N
41.535°N
600 m
13.457°E
13.474°E
d)
Vegetation survey
1m
1m
1m
e)
Diatom sampling
1m
x10
Diatoms
Ethanol (70%)
Distilled H_2O
f)
Abiotic parameters
1m
200g
Density, water content, porosity, organic matter, total phosphorus
Temperature
Dissolved oxygen
Conductivity
pH
Soluble reactive phosphorus
Dissolved inorganic carbon
Nitrate
Sulphate
1L
g)

◂**Fig. 1** Geographical and spatial distribution of the study sites considered (**a**): The Castelporziano Presidential Estate (**b**); Foglino wood (**c**); Ceprano wood (**d**). **e** Schematic representation of sampling activities, including vegetation hybrid sampling approach, with one-square-meter plots and then expanded to the whole community, **f** diatom sapling with at least 10 stones scraped and 3 replicates for each site and **g** abiotic parameters recording

et al., 2008; Bagella, 2023). The selected ponds represent a type of Mediterranean temporary ponds. In fact, these habitats are difficult to identify (Bagella et al. 2007). While the European interpretation manual describes these habitats as "very shallow temporary ponds (a few centimetres deep)", there are significant differences in interpretation at national level (e.g., Grillas et al. 2004) In Italy, as in Spain and France, ponds with an average depth of a few tens of centimeters are also considered to be 3170.

The first site is the "Castelporziano Presidential Estate" (hereafter, CP) (41°43′47"N 12°24′13"E), a natural reserve on the Tyrrhenian coast near Rome. It covers an area of about 6000 hectares (Fig. 1b), where only a limited number of people are allowed to visit it (Castracani et al., 2010). The area in issue is mostly flat, riseing up to 80 m a.s.l. at a distance of about 4 km from the coast (De Nicola et al., 2014). This protected area includes the last remnants of the original Mediterranean plain forest that oce covered the majority of the Latium coast, which is now surrounded by an urban and agricultural landscape (Della Bella et al., 2008). Three sites were selected: CP21 (41°45′26"N 12°23′46"E); CP48 (41°44′10"N 12°25′50"E); CP85 (41°42′30"N 12°22′57"E).

The second site is the Foglino wood (hereafter, FG) (41°28′22"N 12°43′01"E), a large thermophilus deciduous oak forest mixed with Mediterranean scrub communities, covering an area of 550 ha and ranging in altitude between 19 and 33 m a.s.l. (above sea level) (Fig. 1c). It is located in the administrative province of Latina (50 km from Rome) and included in the Natura 2000 network (Lattanzi et al., 2004). The Foglino forest is characterized by the dominance of *Q.frainetto* and *Q. cerris*, while the shrub layer is mainly composed by *Cornus sanguinea* L., *Mespilus germanica* L., *Crataegus monogyna* Jacq. Access to the area by visitors is always free. Five sites were selected: FG1 (41°28′29"N 12°43′09"E); FG2 (41°28′21"N 12°42′54"E); FG3 (41°28′23"N 12°43′15"E); FG4 (41°27′42"N 12°43′11"E); FG5 (41°27′52"N 12°43′26"E).

The third site is the Ceprano wood (hereafter, CR) (41°33′27"N 13°28′6"E), an area of 400 hectares and ranging in altitude between 88 and 107 m a.s.l., full of high ecological-naturalistic value with wooded areas, paths and wetlands (Minutillo et al. 2014), out of the Natura 2000 network (Fig. 1d). In the forest patches, *Q. frainetto* and *Q. cerris* are the dominant tree species. Access to the area by visitors is always free. Seven sites were selected: CR1 (41°33′12"N 13°28′11"E); CR2 (41°33′13"N 13°28′15"E); CR3 (41°33′11"N 13°28′13"E); CR4 (41°33′00"N 13°27′38"E); CR5 (41°32′50"N 13°27′43"E); CR6 (41°32′50"N 13°27′45"E); CR7 (41°32′49"N 13°27′38"E).

Sampling activities and data analysis

Samplings were performed during Spring 2023. For the analysis and monitoring of MTPs, it is suggested in Italy to analyse vegetation using one-square-meter plots (Bagella et al., 2016). However, more squares are often needed to characterize the vegetation of a large MTP. Therefore, a hybrid sampling approach was used. Starting with a one-square-meter plot randomly arranged within the pond, a survey was carried out first in the square and then to the whole community, following the phytosociological approach (Biondi, 2009; Géhu and Rivas-Martinez, 1981) (Fig. 1e). The coverage of species, both in the square and in the relevé, was assigned in percentages and not using the Braun-Blanquet scale. The list of species, with relative coverage estimates, used for the analyses is based on phytosociological sampling plot. Given the aim of analysing the diatom community as a function of ecological and vegetation parameters, summer vegetation sampling was omitted. Species nomenclature follows Bartolucci et al. (2018), while for the syntax names reference was made to Mucina et al. (2016). The deviation from the classical phytosociological method lies in identifying the elementary population—the starting point of the classical phytosociological survey—and in using a percentage scale for estimating species cover.

Epilithic diatom samplings were performed following ISPRA (2014) (Fig. 1f): for each sample, at least 10 stones (dimensions between 64 and 256 mm) were scraped using a commercial toothbrush; then, the toothbrush was immersed in a 50 ml Falcon containing 70% diluted ethanol and distilled water. Stones were collected throughout the pond, covering a range of depths up to approximately 100 cm to capture variability in epilithic diatom communities. Once in the laboratory, diatom samples were oxidized following the hydrogen peroxide method (Taurozzi et al. 2023). Briefly, 5 ml of sample were centrifuged at 1500 rpm for 10 min, the supernatant discarded, and the sediment was mixed with 20 ml of H_2O_2 and heated for 3 h to degrade organic matter. After complete oxidation, diatoms were centrifuged at 1500 rpm for 10 min (the procedure was repeated for three times), mounted on permanent slides using Naphrax® (high refractive index resin) to fix the coverslip. For each site, three different replicates were performed as suggested by Maitland et al. (2020). For each replicate, at least 10 stones were sampled per site to ensure representativeness, following the ISPRA (2014) guidelines. Separate commercial toothbrushes and Falcon tubes were used for each replicate to avoid cross-contamination. The morphological identification of diatom species was carried out using many taxonomic references (Krammer & Lange-Bertalot, 1986; Krammer & Lange-Bertalot, 1988; Krammer & Lange-Bertalot, 1991a; Krammer & Lange-Bertalot, 1991b; Krammer et al., 2002; Taylor et al., 2007; Ector & Hlúbiková, 2010). In the laboratory, 400 valves were counted per slide (three slides per pond), and the counts from the three slides were combined, resulting in a total of 1200 valves per pond. The survey was combined with an assessment of ecological parameters. Temperature (T), dissolved gen (DO, expressed in terms of % saturation), conductivity (Cond.), and pH were measured in situ with a multi-parameter probe (MANTA + 30, EasyProbe) (Fig. 1g). Simultaneously, an aliquot of water was collected (1 L) and immediately filtered through glass microfiber filters (GF/F Whatman®; 0.7 µm) for subsequent laboratory analysis of soluble reactive phosphorus (SRP), dissolved inorganic carbon (DIC), nitrate (NO_3^-) and sulfate (SO_4^{2-}) using standard methods (A.P.H.A., 2017). At the maximum depth of the water column, a sediment sample (200 g) was collected using a manually infixed Plexiglas core (4 cm circumference). The sediment, corresponding to the 0–5 cm surface layer, was subsequently characterized for density, water content, porosity, organic matter (OM, %) and total phosphorus (TP) content. For the determination of OM content, a portion of dry sediment (0.5 g) was weighed inside ceramic crucibles and incinerated in a muffle furnace at 450 °C for a time of 4 h (Buchanan et al., 1984). TP in the sediments was determined by the potassium persulfate oxidation method (Aspila et al., 1976).

Statistical analysis

Physical and chemical parameters

Pairwise correlations were computed between environmental variables using the base function *cor()* from *corrplot* package (Wei & Simko, 2024). A principal component analysis (PCA) was performed with *princomp()* (base R), and the contribution of each variable to the first two principal components was examined through loadings.

Diatom overview and water quality

Since no diatoms were detected in sites CR4 and CR5 ($n < 10$), all diatom-related analyses were restricted to the remaining sites. Diatom Red List species was taken from https://www.rote-liste-zentrum.de/. The ecological quality of each site was then assessed using the EPI-D index, which is a diatom-based metric suitable for standing waters, including temporary ponds. Unlike Italian indices designed for rivers (ICMi) or permanent lakes (EPI-L), the EPI-D index effectively accounts for the variable hydrology and ecological characteristics of temporary pond ecosystems. The index was calculated with the *diaThorAll()* function from the *diaThor* R package (Nicolisi and Sathicq, 2025).

Diatom taxonomic α and β diversity

Taxonomic α diversity of diatoms was calculated for each sample using species richness (*specnumber()* function from *vegan* package), and Pielou's evenness (computed as Shannon divided by log species richness) (Oksanen et al., 2025). Correlations between diversity indices were assessed using Pearson correlation (*cor()*) and associated p-values were obtained with *cor.test()*). Differences in richness and Pielou's evenness among sites were tested with the Kruskal–Wallis test (*kruskal.test()* from base R). To evaluate the influence of environmental parameters on richness and Pielou's evenness, generalized linear models were fitted (*glm()* from base R) with Poisson distribution for richness and Gaussian distribution for Pielou. Predictor selection was performed with stepwise forward selection (*step()* from base R), comparing a null model to a full model including all measured environmental variables.

Considering taxonomic β diversity of diatoms, presence-absence data were obtained using *decostand (method="pa")* (*vegan*), and Bray–Curtis dissimilarities were calculated with *vegdist()* function (*vegan*). Within-site β diversity components (total, turnover, and nestedness) were computed using *beta.multi()* (*betapart* package) (Baselga et al., 2025), while pairwise Sorensen, turnover, and nestedness dissimilarities were obtained using *beta.pair()* (*betapart*). Differences in β diversity among sites were tested using permutational multivariate analysis of variance (PERMANOVA, *adonis2()* function from *vegan* package) on the pairwise dissimilarity matrices. Pairwise comparisons between sites were performed by subsetting the dissimilarity matrices and applying *adonis2()* for each site pair. To evaluate the effect of environmental variables on β diversity, pairwise differences in environmental parameters were calculated using a custom R function (*pairwise_env()*) on the environmental data matrix. Generalized linear models (*glm()* from base R) were then fitted for total β diversity, turnover, and nestedness, with Gaussian errors. Stepwise forward model selection (*step()* from base R) was used to identify the most relevant environmental predictors.

Diatom functional α and β diversity

18 Functional traits data were taken from Rimet & Bouchez (2011) and Rimet & Bouchez (2012), including ecological guilds, life-forms and size classes (Table S1). Functional α diversity indices, including richness (FRic), evenness (FEve), dispersion (FDis) were calculated with *dbFD()* from the *FD* package (Laliberté et al., 2014). Traits were standardized (stand.x = TRUE), and negative eigenvalues were corrected with the Cailliez method (corr = "cailliez"). Pearson correlations among functional diversity indices were computed using *cor()* (base R), and significance tested with a custom function applying *cor.test()* (base R). Differences among sites were tested using Kruskal–Wallis tests (*kruskal.test()*, base R) for each index. To investigate the effect of environmental variables on functional α diversity, generalized linear models (*glm()*, base R) with Gaussian errors were fitted for FRic, FEve, and FDis. Stepwise forward selection (*step()*, base R) was applied to identify the most relevant predictors.

Functional β diversity indices were computed using *dbFD()* and *functcomp()* from the *FD* package. Negative values were corrected with the Cailliez method (*corr = "cailliez"*). Pairwise functional dissimilarities were calculated using Bray–Curtis distances (*vegdist()*, *vegan*) on the standardized trait matrices. Global and pairwise differences among sites were tested using permutational ANOVA (*adonis2()*, *vegan*) on total β diversity, turnover, and nestedness matrices. Finally, to identify the traits most represented in each site, we calculated the Community Weighted Mean (CWM) for each functional trait. The CWM is defined as the abundance-weighted average of trait values in a community. The differences between CWM were tested using Kruskal–Wallis tests (*kruskal.test()*, base R) for each trait. All calculations were performed using the *dplyr* package (Wickham et al., 2025) for data manipulation and summarization.

Plant taxonomic α and β diversity

For vegetation communities, the same taxonomic analyses used for diatoms were applied.

Influence of abiotic factors on diatom and plant communities

To investigate the joint response of diatom and vegetation communities to environmental gradients, redundancy analysis (RDA) was performed using the *vegan* package. The 20 most abundant diatom taxa and the 20 most abundant vegetation taxa were selected and their abundance transformed with Hellinger standardization (*decostand(…, method = "hellinger")*, *vegan*). Environmental variables were standardized (*scale()*, base R) prior to analysis. Significance of the overall model, canonical axes, and individual predictors was tested by permutation tests (*anova.cca()*, *vegan*) with 999 permutations. Variance inflation factors were calculated using *vif.cca()* (*vegan*) to assess multicollinearity among predictors. The proportion of explained variance by the first two canonical axes was computed from the eigenvalues of the constrained ordination (*eig()*, vegan).

Similarly, RDA was performed to assess the influence of vegetation composition on diatom communities. A full RDA model was computed (*rda()*, *vegan*) with all vegetation variables as predictors. Forward selection of explanatory variables was performed using *ordiR2step()* (*vegan*) with 1000 permutations, starting from a null model and considering the adjusted R^2 as a selection criterion. The explanatory power of the final model was evaluated using adjusted R^2 (*RsquareAdj()*, *vegan*). The significance of the model, the individual axes, and each explanatory variable was assessed by permutation tests (*anova.cca()*, *vegan*) with 1000 permutations. The proportion of variance explained by the first two canonical axes was calculated from the eigenvalues of the constrained ordination.

All statistical analyses were performed using R software (R Core Team, 2024; version 4.4.2), and significance was assessed at $P < 0.05$.

Results

Physical and chemical parameters

The mean temperature across sites was 12.9 °C, ranging from 10.0 °C (CR) to 15.3 °C (CP). pH values varied between 5.9 and 7.8, with slightly more acidic conditions in FG and CR (mean 6.8) compared to CP (7.5). Dissolved oxygen ranged from 58.1 to 110.9%, with lower values at CR (71%) than at FG and CP (83% and 78%, respectively). Cond. varied widely (47–552 µS cm^{-1}), with CR showing the highest mean (197 µS cm^{-1}). DIC concentrations were generally low (0.37–4.41 mEq L^{-1}), with higher averages at CR (1.7). Nitrate values were low overall (0.02–2.70 mg L^{-1}), with CP reaching the highest mean (0.8). Sulfate concentrations showed spatial differences, from 0.61 mg L^{-1} at CR to 2.22 mg L^{-1} at CP. Organic matter ranged from 15% (FG) to 24% (CP), while total phosphorus was highest at CP (512 µg g^{-1}) and lowest at CR (341 µg g^{-1}) (Table S2).

All the physical and chemical parameters analysed showed positive correlation with all the variables considered, except for sediment density, O_2 and SO_4^{2-} (Fig. 2a). Standard two-dimension PCA showed that the distribution of the component concentrations was highly concentrated in the upper right quadrant. NO_3^-, sediment water content, Conductivity, DIC were the top four variables with the highest cos2, hence contributing the most to PC1 and PC2 (Fig. 2b). From the biplot, NO_3^-, sediment water content, Conductivity and OM have higher magnitude compared to T, pH, O_2 and SO_4^{2-}.

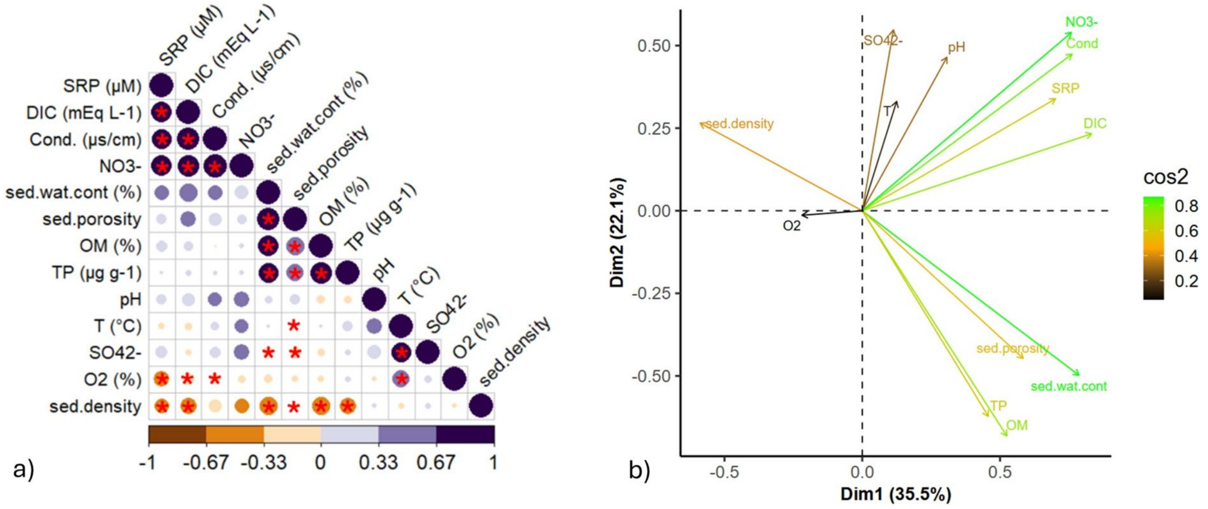


Fig. 2 **a** correlation plot for the physical and chemical parameters showing the statistically significative values (*$P < 0.05$) and, below, the r Pearson value; **b** Principal component analysis (PCA) for physical and chemical parameters (cos2 = the square cosine value for each variable with respect to the first two principal components). Variables that are negatively correlated are displayed to the opposite sides of the biplot's origin

Diatom overview and water quality

=An investigation of the epilithic diatoms resulted in the description of 89 diatom species (Table S3) in the 13 ponds analysed. Despite sampling epilithic diatoms from rocks at sites CR4 and CR5, no diatom cells were observed on the slides. This absence is likely due to the very short inundation periods, which may have prevented the establishment of epilithic communities. Other possible factors include limited availability of colonizing diatom cells due to site isolation, a predominance of planktonic diatoms over benthic species, physical disturbance by fauna, and stochastic variability typical of temporary ponds. The highest number of species was found in CR (54), followed by FG (44) and CP (42). *Eunotia bilunaris* (Ehrenberg) Schaarschmidt showed the highest records with 1760 individuals (34% of the total population), followed by *Encyonema silesiacum* (Bleisch) D.G.Mann with 729 individuals (14%) and *Eunotia minor* (Kützing) Grunow with 433 individuals (8%). Considering the Red List of threatened diatoms, most of species ($n=61$) were classified as "Least concern", $n=7$ were classified as "Vulnerable" and $n=3$ as "Endangered"; the remaining ones were classified as "Not Evaluated" ($n=6$) and "Data Deficient" ($n=12$) (Table S4). Water quality, assessed using the EPI-D index, varied slightly among the three pond groups (Table S5). CP showed the highest EPI-D value (EPI-D = 1.37; EPI-D20 = 13.52), indicating slightly lower water quality compared to the other sites. CR presented the lowest EPI-D (EPI-D = 1.07; EPI-D20 = 14.91), suggesting the best water quality among the ponds. FG showed intermediate values (EPI-D = 1.15; EPI-D20 = 14.53).

Diatom taxonomic α and β diversity

Taxonomic species richness (N) ranged from a minimum of 10 species in CR2 to a maximum of 25 species in CR3. The evenness index (Pielou's J) showed the highest values in CP85 ($J=0.79$) and CR7 ($J=0.77$), suggesting relatively balanced communities, while the lowest values were found in FG2 ($J=0.44$) and CR2 ($J=0.47$), indicating more uneven species distributions (Table S6). No significant differences in taxonomic α diversity were observed among the three pond groups (FG, CR, CP). Species richness did not differ significantly between groups (Fig. 3a; $\chi^2=1.26$, $P=0.53$). Similarly, for evenness no significant differences were found between the three site groups (Fig. 3b; $\chi^2=2.86$, $P=0.24$). Water conductivity (Cond) (AIC = 68.71) was the only environmental variable significantly associated with variation in richness (Fig. 3c; $z=2.37$, $P=0.018$). Regarding evenness, forward selection (AIC = − 18.26) identified sediment water content (sed.wat.cont) and pH as the best predictors. However, neither variable reached statistical significance (sed.wat.cont, $\mathrm{t}=1.81$, $P=0.101$; pH $\mathrm{t}=1.69$, $P=0.122$).

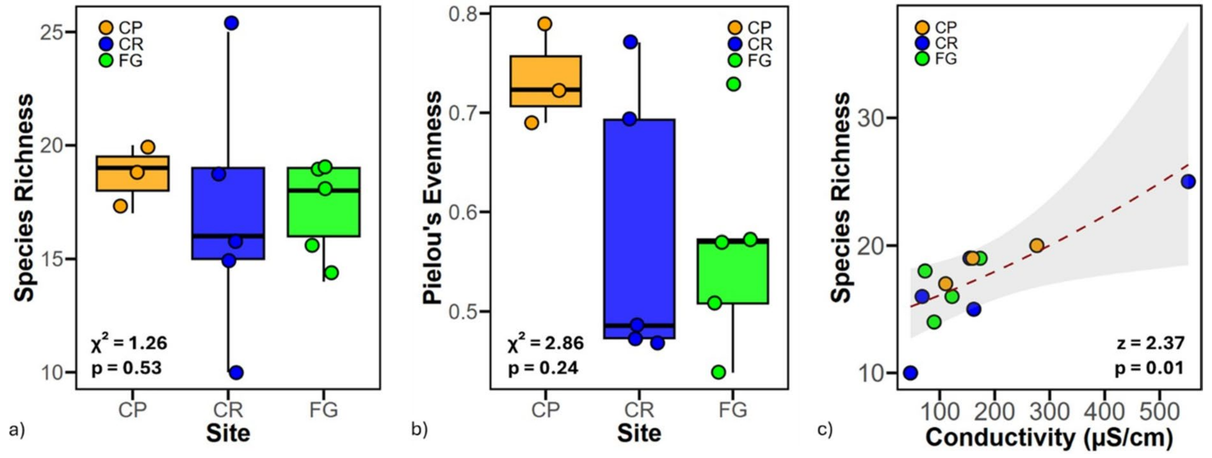


Fig. 3 Boxplots showing the distribution of diatom α diversity across sampling sites. **a** Species richness, expressed as the number of observed taxa. **b** Pielou's evenness, indicating the evenness of species abundances within each site. **c** Relationship between species richness and water conductivity (µS/cm) across sampling sites

Total β diversity (β SOR) was highest at CR (0.79), followed by CP (0.71) and FG (0.69). Turnover (β SIM) was the main contributor at all sites, ranging from 0.66 at FG to 0.73 at CR. In contrast, the nestedness component (β SNE) was consistently low across sites, from 0.018 at CP to 0.062 at CR, indicating that differences in community composition were primarily driven by species replacement rather than by loss or gain of species without replacement.

PERMANOVA showed that total β diversity (β SOR) differed significantly among sites ($F=1.96$, $R^2=0.28$, $P=0.006$), indicating that the composition of diatom communities varied across CP, CR, and FG (Fig. 4a). Turnover component (β SIM) also differed significantly among sites ($F=2.18$, $R^2=0.30$, $P=0.002$), suggesting that species replacement was the main driver of community differences. In contrast, the nestedness component (β SNE) did not differ among sites ($F=0.26$, $R^2=0.05$, $P=0.746$). Pairwise PERMANOVA for total β diversity showed significant differences between FG and CP (Fig. 4b; $F=3.02$, $R^2=0.33$, $P=0.018$), while the difference between CR and CP was marginally significant ($F=1.76$, $R^2=0.23$, $P=0.053$), and no significant difference was found between FG and CR ($F=1.39$, $R^2=0.15$, $p=0.118$). Similarly, for the turnover component, the most pronounced differences were between FG and CP (Fig. 4c; $F=3.31$, $R^2=0.36$, $P=0.015$) and CR and CP ($F=1.93$, $R^2=0.24$, $P=0.029$), whereas FG and CR were not significantly different ($F=1.53$, $R^2=0.16$, $P=0.106$). These results indicate that CP communities were taxonomically the most distinct, exhibiting higher turnover and overall functional differentiation compared to the other sites. In contrast, FG and CR displayed more similar functional compositions, suggesting less variation in taxonomic composition between these sites.

The GLM forward selection analyses revealed that total β diversity was entirely explained by the two other components, turnover and nestedness ($P<0.001$ for both), with no direct effect of environmental parameters retained in the final model (AIC = − 5382). For turnover, sed.wat.cont was marginally significant ($P=0.053$), while all other environmental variables were not statistically significant. For nestedness, the environmental variables were not significant (all $p>0.05$).

Diatom functional α and β diversity

FRic values ranged widely across samples (0.15–111.77), FEve showed a relatively narrow range (0.37–0.52), while FDis varied between 1.96 and 4.47 (Table S7). Kruskal–Wallis tests revealed no significant differences among the three sites for FRic ($\chi^2=1.93$, $P=0.38$) and FEve ($\chi^2=2.20$, $P=0.33$). FDis showed a marginal tendency to differ between sites ($\chi^2=4.83$, $P=0.09$), indicating slightly higher functional dispersion in some sites.

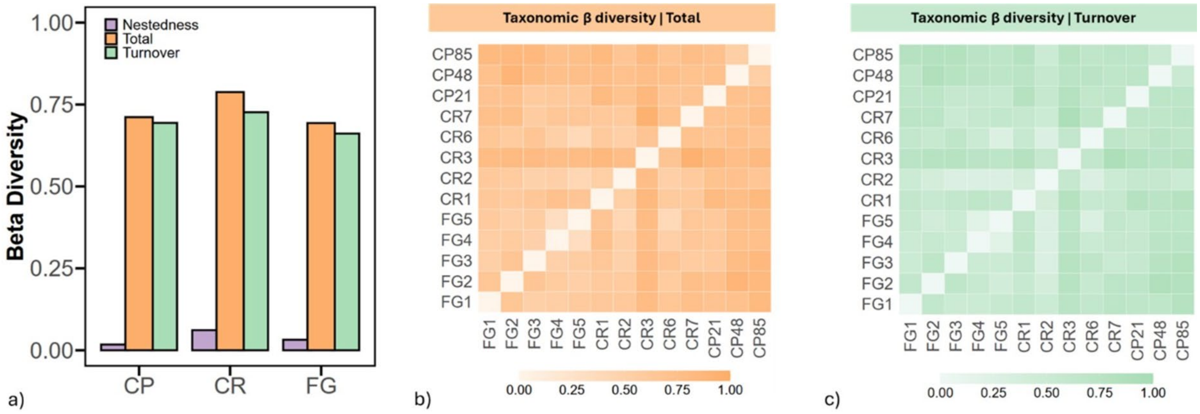


Fig. 4 **a** Partitioning of β diversity into turnover and nestedness components across sampling sites. Bars represent the contribution of each component to total β diversity, with values ranging from 0 to 1. **b** Heatmap of total taxonomic β diversity (βSOR) among sample pairs. Values range from 0 (identical communities) to 1 (completely dissimilar communities). **c** Heatmap of the turnover component of taxonomic β diversity (βSIM) among sample pairs. Values range from 0 (no species replacement) to 1 (complete species turnover)

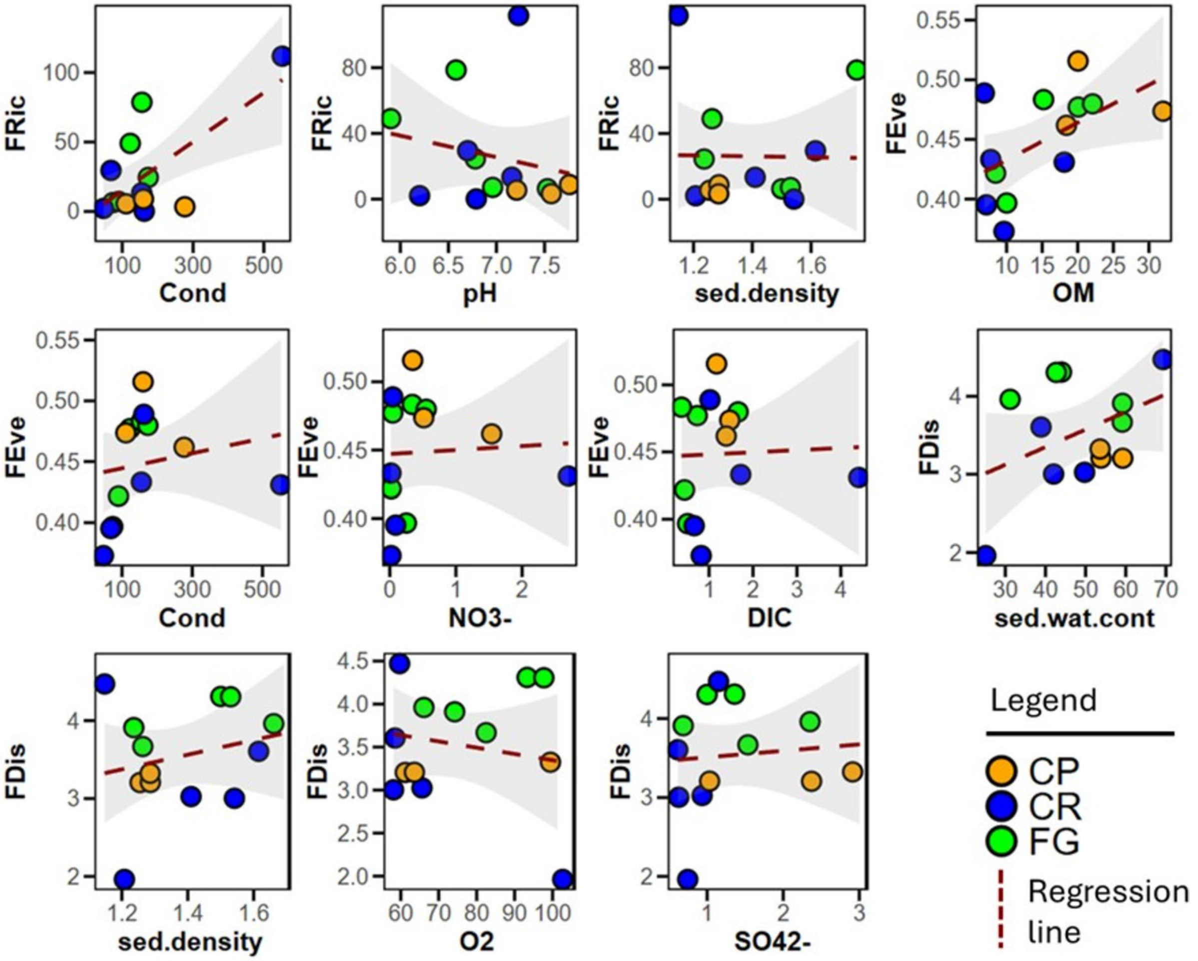


Fig. 5 Relationships between environmental variables and functional α diversity indices of diatom communities. Generalized linear models (GLMs) with Gaussian distribution were fitted to assess the effects of environmental gradients on three functional diversity metrics: functional richness (FRic), functional evenness (FEve), and functional dispersion (FDis). Each point represents a sample, colored by site, while dashed lines indicate the fitted GLM with 95% confidence intervals. CP = Castelporziano, FG = Foglino, CR = Ceprano. O2 = Dissolved oxygen; Cond = Conductivity; T = Temperature; OM = Organic matter; DIC = Dissolved inorganic carbon; NO3- = Nitrate; SO42- = Sulfate; Sed. density = Sediment density; Sed. wat. cont. = Sediment Water Content

Functional α diversity showed significant correlation with environmental variables (Fig. 5). For FRic, the best-fitting model included conductivity (Cond.), pH, sediment density, and SRP as predictors. FRic increased significantly with higher conductivity ($z=0.20$, $P=0.01$) and sediment density ($z=73.05$, $P=0.058$, marginally significant), and decreased with higher pH ($z=-\ 29.73$, $P=0.024$). FEve was positively influenced by organic matter ($z=0.0059$, $P=0.0013$) and conductivity ($z=0.00084$, $P=0.0038$), and negatively by nitrate (NO_3^-, $z=-\ 0.082$, $P=0.029$) and dissolved inorganic carbon (DIC, $z=-\ 0.045$, $P=0.022$). Finally, Functional Dispersion (FDis) was strongly affected by sediment characteristics and water chemistry. Sediment water content ($z=0.0598$, $P=0.0024$), sediment density ($z=6.77$, $P<0.001$), and oxygen ($z=0.0353$, $p=0.0032$) were positively related to FDis, while sulfate (SO_4^{2-}, $z=-\ 0.753$, $P=0.0079$) had a negative effect.

Considering functional β diversity, on average, the CR and CP sites exhibited higher total functional

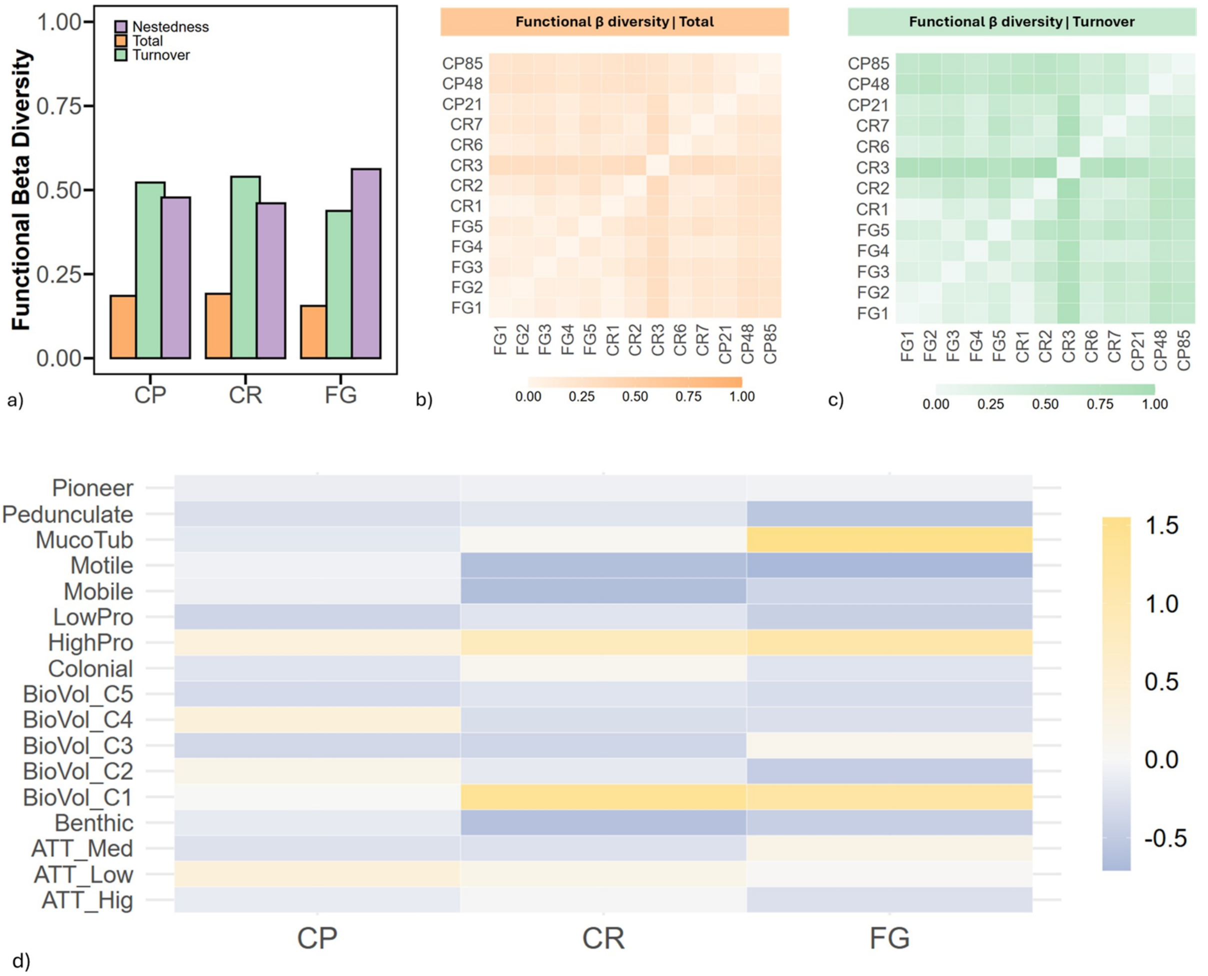


Fig. 6 **a** Partitioning of functional β diversity into total, turnover, and nestedness components across sampling sites. Bars represent the contribution of each component to total functional β diversity (values range from 0 to 1); **b** Heatmap of total functional β diversity among sample pairs. Values range from 0 (identical functional composition) to 1 (completely dissimilar functional composition); **c** Heatmap of the turnover component of functional β diversity among sample pairs. Values range from 0 (no functional replacement) to 1 (complete functional turnover). **d** Heatmap of community-weighted mean (CWM) values for functional traits across sampling sites. CP = Castelporziano, FG = Foglino, CR = Ceprano

β diversity (0.191 and 0.185, respectively) compared to FG (0.155) (Fig. 6a). This pattern was primarily driven by turnover, which accounted for a larger proportion of the total dissimilarity in CR (0.539) and CP (0.522) than in FG (0.438), whereas nestedness contributed more to FG (0.562) than in CR (0.461) or CP (0.478). PERMANOVA analyses confirmed that differences in total functional β diversity ($F = 3.592$, $P = 0.008$) and turnover ($F = 3.592$, $P = 0.008$) among sites were statistically significant, indicating that species replacement between ponds was a major driver of functional dissimilarity. In contrast, nestedness did not differ among sites ($F = -0.872$, $P = 0.999$). Pairwise PERMANOVA showed that total functional β diversity between FG and CP was significant ($F = 14.96$, $R^2 = 0.714$, $P = 0.021$), while FG and CR were marginally different ($F = 2.44$, $R^2 = 0.234$, $P = 0.059$) and CR and CP were not significantly different ($F = 1.90$, $R^2 = 0.241$, $P = 0.159$) (Fig. 6b). Similar patterns were observed for the turnover component, with FG and CP differing significantly ($F = 14.96$, $R^2 = 0.714$, $P = 0.023$), FG and CR showing marginal differences ($F = 2.44$, $R^2 = 0.234$, $P = 0.061$), and CR and CP not differing significantly ($F = 1.90$, $R^2 = 0.241$, $P = 0.169$) (Fig. 6c).

Community-weighted mean (CWM) analyses revealed that the traits showing statistically significant variation included MucoTub ($P = 0.014$), BioVol_C3 ($P = 0.020$), BioVol_C1 ($P = 0.027$), BioVol_C4 ($P = 0.040$), and ATT_Med ($P = 0.043$) (Fig. 6d, Table S8). The FG site was characterized by higher values of MucoTub (1.55) and BioVol_C1 (1.17), indicating a dominance of mucilaginous and larger-sized species, whereas CR and CP ponds had lower MucoTub values (0.09 and − 0.17) and varied BioVol_C1 values (1.41 in CR and 0.01 in CP). BioVol_C4 and ATT_Low were more abundant in CP (0.41 and 0.42, respectively) compared to FG and CR, suggesting a prevalence of smaller or later-successional traits in CP ponds. Medium-sized species (ATT_Med) were relatively enriched in FG (0.23) but less represented in CR and CP (− 0.24). Overall, these results indicate that FG ponds tend to host larger, mucilaginous species with moderate size traits, CR ponds show intermediate trait values, and CP ponds are characterized by smaller-bodied or pioneer-associated traits.

Total functional β diversity was not directly explained by any measured environmental variable. Similarly, turnover showed no significant relationship with organic matter (OM), despite being included in the model. Nestedness was also independent of dissolved oxygen (O_2) and soluble reactive phosphorus (SRP), as neither variable had a significant effect ($P > 0.4$ in all cases). These results indicate that, within our dataset, the three components of functional β diversity did not exhibit clear correlations with the measured abiotic parameters.

Plant taxonomic α and β diversity

A total of 52 plant species were recorded (Table S9). Species richness varied among samples, ranging from 3 to 19 species per site, with the highest richness observed in FG1 (19 species) and the lowest in CR7 (3 species). Pielou's evenness ranged from 0.67 to 0.96, indicating generally moderate to high evenness across samples, with the most even community in CR6 (0.96) and the least even in FG5 (0.67) (Table S10).

Kruskal–Wallis tests showed no significant differences in species richness among the three sites (FG, CR, CP; $\chi^2 = 1.96$, $P = 0.37$). Evenness differed slightly among sites, with a marginally non-significant trend ($\chi^2 = 5.41$, $P = 0.067$), suggesting that some sites may host more evenly distributed species than others. Forward selection of generalized linear models identified SRP, sediment–water content, and DIC as the best predictors of species richness. Among these, sediment–water content showed a significant positive effect (estimate = 0.022, $P = 0.038$), while DIC had a marginal negative effect (estimate = − 0.311, $P = 0.087$). For Pielou's evenness, organic matter (OM) was the only variable retained in the forward selection, showing a marginal negative effect on evenness (estimate = − 0.0055, $P = 0.079$).

β diversity analysis showed that turnover was the dominant process across sites, with values ranging from 0.50 (CP) to 0.62 (FG), while nestedness contributed less (0.11–0.18). Overall Sørensen dissimilarity (β SOR) ranged from 0.68 (CP) to 0.76 (CR). Permutational ANOVA confirmed that total β diversity differed significantly among sites (β SOR: $R^2 = 0.30$, $F = 2.11$, $P = 0.008$), driven primarily by turnover (β SIM: $R^2 = 0.40$, $F = 3.33$, $P = 0.007$), whereas nestedness was not significantly different (β SNE: $P = 0.87$). Pairwise comparisons indicated significant differences in total β diversity and turnover between FG–CP (β SOR $p = 0.015$; β SIM $P = 0.025$) and CR–CP (β SOR $P = 0.020$; β SIM $P = 0.019$), while FG–CR did not differ significantly.

Forward selection of generalized linear models indicated that environmental variables had a limited influence on β diversity patterns. For total β diversity, temperature (T) and DIC were included in the best model but were not significant (T, $P = 0.516$; DIC: $P = 0.790$). Similarly, turnover and nestedness were not significantly affected by any measured environmental parameter, with sediment density and pH showing only marginal effects ($P = 0.090$ and $P = 0.896$, respectively).

Influence of abiotic factors on diatom and plant communities

The RDA of diatoms and vegetation against standardized environmental variables was not significant (ANOVA, $F = 0.99$, $P = 0.615$), indicating that the measured parameters explained little of the community variation. Individual axes were also

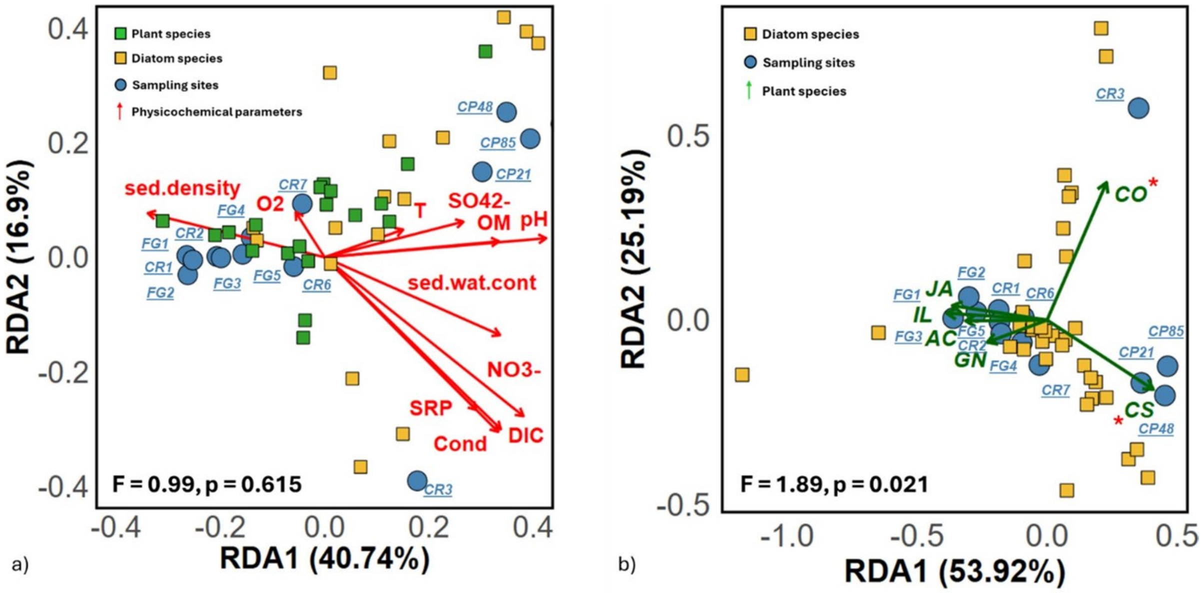


Fig. 7 **a** Canonical redundancy analysis (RDA) ordination of the top 20 diatom and vegetation taxa in relation to standardized environmental parameters. Arrows indicate the direction and strength of environmental gradients, while squares represent species (yellow = diatoms, green = plants) and circles represent sampling sites. Percentages on axes show the variance explained by the first two RDA axes. **b** RDA ordination of diatom communities constrained by vegetation variables treated as environmental predictors. Arrows represent significant vegetation variables selected by forward selection, squares represent diatom species, and circles indicate sampling sites. Percentages on the axes show the variance explained by the first two RDA axes. AC = *Agrostis canina*; CO = *Carex otrubae*; CS = *Callitriche stagnalis*; GN = *Glyceria notata*; IL = *Isoëtes longissima*; JA = *Juncus articulatus*. O2 = Dissolved oxygen; Cond = Conductivity; T = Temperature; OM = Organic matter; SRP = Soluble reactive phosphorus; DIC = Dissolved inorganic carbon; NO3- = Nitrate; SO42- = Sulfate; Sed. density = Sediment density; Sed. wat. cont. = Sediment Water Content; *$P < 0.05$a

non-significant, with RDA1 and RDA2 explaining 40.7% and 16.9% of the variance, respectively (RDA1: $F = 4.42$, $P = 0.582$; RDA2: $F = 1.83$, $P = 0.892$) (Fig. 7a). None of the environmental variables showed a significant effect.

Considering vegetation as environmental variables, the RDA model explained 65.4% of the total variance in species composition (Fig. 7b). The first two constrained axes captured 53.9% and 25.2% of the constrained variance, respectively, while the residual (unconstrained) variance accounted for 19.0% of the total variance. Forward selection identified six plant species, *Callitriche stagnalis* L. (CS), *Carex otrubae* Podp. (CO), *Isoëtes longissima* Durieu (IL), *Juncus articulatus* L. (JA), *Agrostis canina* L. (AC), and *Glyceria notata* Chevall. (GN), as significant contributors to community structure. The reduced RDA model including these variables had an adjusted R^2 of 0.309 and was significant overall ($F = 1.893$, $P = 0.021$). Sequential permutation tests by term indicated that CS ($F = 3.673$, $P = 0.009$) and CO ($F = 3.575$, $P = 0.002$) were highly significant, while IL ($F = 1.262$, $P = 0.255$), JA ($F = 0.746$, $P = 0.612$), AC ($F = 0.499$, $P = 0.800$), and GN ($F = 1.604$, $P = 0.152$) were not individually significant. Axis-wise permutation tests revealed that the first RDA axis (RDA1) explained a substantial portion of variance and was significant ($F = 6.124$, $P = 0.037$), whereas subsequent axes (RDA2–RDA6) did not reach significance.

Discussions

Studies indicating the influence of physical and chemical and biotic factors governed by natural and anthropogenic processes on plant and diatom community structure and species richness are still scarce in MTPs. The present study assessed the influence of physical and chemical and biotic variations in influencing the composition and structure of plant and diatom in MTPs of Lazio. Our results provide the first contribution to the knowledge of plant and diatom coupled biodiversity associated with MTPs and their ecological relationship, confirming that both plants and diatoms represent excellent bioindicators for MTPs (Lumbreras et al., 2016; Ansari et al., 2017; Taurozzi et al., 2024a). Moreover, considering the ecological relevance of diatoms for biomonitoring strategies, our research provides the first insight about the characterization of typical diatom species inhabiting 3170* Habitats.

Diatom taxonomic α and β diversity characterization

Our results show that diatom communities in MTPs respond similarly to abiotic conditions typical of the Mediterranean climate zone. Despite the sites being subject to different levels of anthropogenic pressures and management, the number of species recorded at the three sites was comparable. This is consistent with the findings from Della Bella & Mancini (2009), who studied benthic diatoms and macroinvertebrates in permanent freshwater ponds along a human impact gradient in central Italy. They found that, while macroinvertebrate richness decreased with increasing anthropogenic pressure, diatom species richness alone did not reflect this relationship. This suggests that these fragile environments, despite their strong seasonal variability, support diverse communities that are shaped primarily by large-scale phenomena (Fehlinger et al., 2023), rather than the intrinsic properties of the individual water bodies. Likewise, Red List diatom species were evenly distributed across taxonomic groups at the different sites. Most of these species are not currently at risk, indicating that MTPs provide a suitable environment for the development and survival of many taxa (Zacharias et al., 2007). Despite their short hydroperiod, these ecosystems do not appear to pose particular risks of extinction for most species. However, the presence of vulnerable and endangered species highlights the importance of MTPs in safeguarding fragile species (Beja & Alcazar, 2003; Zacharias & Zamparas, 2010). At the same time, community composition revealed that, although the CP site is the most protected, being fenced and inaccessible, the water quality, as expressed by the EPI-D index, was the lowest. Several hypotheses can be proposed: First, water quality may be influenced by indirect atmospheric pollution (Taurozzi et al., 2024a, b) or by the abiotic characteristics of the site itself (Akhtar et al., 2021; Ji et al., 2021). Second, as highlighted by Dey et al. (2021) and Ouyang et al. (2006), water quality can also undergo marked seasonal fluctuations. This means that the values recorded at the sampling sites may represent only a snapshot of the conditions at the moment of sampling. Finally, it is important to consider the potential impact of ungulates inhabiting the estate, particularly wild boar. As demonstrated by Burrascano et al. (2015) in the nearby Circeo forest, current vegetation patterns partly reflect the legacy effect of past rooting disturbance. In addition, their droppings can affect nutrient levels in the water, thereby influencing overall water quality (Hubbard et al., 2004).

When considering α diversity, the sites did not show significant differences, confirming the preliminary evidence that Habitat 3170 plays a key ecological role in supporting diverse communities, regardless of their geographical location. Our results are consistent with previous studies: for example, Hill et al. (2019) reported comparable α diversity levels in macroinvertebrate communities of temporary ponds, identifying pond surface area, dry-phase duration, and macrophyte cover as consistent predictors of different facets of α diversity. Similarly, although focusing on invertebrates, Epele et al. (2019) demonstrated that α diversity in temporary ponds was primarily driven by local features and only secondarily by climatic descriptors, emphasizing the importance of intrinsic site characteristics and supporting the observation that MTPs host similar communities.

Our study also highlights the relationship between diatom richness and water conductivity in MTPs. These results are in line with previous findings (Urrea and Sabater, 2009; Mangadze et al., 2017): conductivity is frequently associated with changes in diatom composition in both lentic and lotic environments and also in temporary ponds (Della Bella et al., 2007). For instance, Cantonati et al. (2012) demonstrated in alpine ponds that conductivity represents a fundamental parameter for diatom communities. Comparable patterns were also observed in zooplankton (Coccia et al., 2024) and in lake ecosystems (Pestryakova et al., 2018), confirming conductivity as a key driver of aquatic communities. Higher conductivity may enhance diatom diversity by creating a wider range of nutrients and chemical microhabitats that promote coexistence, whereas very low conductivity can limit resource availability and reduce settlement opportunities for more demanding species (Chen et al., 2016; Mangadze et al., 2017).

Conversely, the analysis of taxonomic β diversity revealed a marked differences among the sites, particularly between Castelporziano (CP) and Foglino (FG). This is a noteworthy finding, given that the two sites share several ecological features, such as their coastal proximity and the presence of fences that limit direct anthropogenic pressures (although FG remains accessible to the public). In addition, there are no major pollution sources associated with vehicular traffic. Instead, the observed divergence may reflect more subtle differences related to the ponds' hydrological and geomorphological history, the duration and depth of the hydroperiod, or the specific characteristics of the substrate and surrounding vegetation. These factors influence the availability of diatom microhabitats (Nemes-Kokai et al., 2024). In addition, FG's greater ecological connectivity with neighboring habitats, coupled with potential indirect anthropogenic pressures (e.g., recreational or agricultural activities in the surrounding area), could have contributed to the differentiation of its communities compared to CP's.

This outcome highlights the fact that even similar environments are not necessarily colonized by the same species. This suggests that each site acts as an "environmental filter" that selects distinct communities, as demonstrated by Lozada-Gobilard et al. (2019) for plants. One possible explanation is that, as a closed and relatively inaccessible reserve, CP represents an isolated environment that is less exposed to anthropogenic disturbance. This isolation may favor the persistence of distinct communities that are less influenced by dispersal and colonization processes from adjacent habitats, thereby increasing differentiation from other sites (Orsini et al., 2013; Burns, 2019). In contrast, CR is located near agricultural areas and infrastructure (e.g., the highway), which likely enhance ecological connectivity with the surrounding environment and intensify the impact of anthropogenic pressures (Rudnick et al., 2012; Baguette et al., 2013; Mullu, 2016). These conditions could promote diatom communities that are more similar to those of FG, where there are unrestricted access and greater exposure to indirect disturbances.

Finally, the absence of direct effects of the considered factors on taxonomic β diversity supports the view, as also suggested by Teittinen et al. (2016), that environmental variables, particularly chemical ones, play only a limited role in structuring diatom β diversity, whereas physical factors may exert a stronger influence. As Michel & Knouft (2014) observed, chemical and physical parameter; s may primarily affect differences between distinct environments rather than within the same environment. Although diatoms clearly respond to abiotic variation, our results suggest that these parameters do not directly determine species' composition and relative diversity.

Diatom functional α and β diversity characterization

The results of the functional trait analysis partially mirror those observed for taxonomic diversity. Specifically, functional α diversity did not differ significantly among sites, suggesting that FRic remained relatively constant across the MTPs examined. As emphasized by Rimet & Bouchez (2012), diatom life-forms have undergone strong selective pressures, reflecting adaptations related to attachment, light and nutrient acquisition, sinking dynamics, and habitat selection. The temporary nature of the ponds studied may explain the absence of pronounced differences in guild distribution across MTPs: species belonging to the motile guild, thanks to their ability to actively move, can escape localized disturbances (Stenger-Kovács and B-Béres, 2024) and are particularly well adapted to colonize habitats subject to strong seasonal hydrological and chemical fluctuations, such as MTPs, regardless of anthropogenic influences (Taurozzi and Scalici, 2024). Furthermore, our findings confirm the high sensitivity of functional diversity to abiotic drivers, showing that physical and chemical variables modulated by temporal dynamics play a key role in shaping the composition of diatom functional communities (Riato et al., 2017; Guo et al., 2020).

The correlation between FRic and environmental variables indicates that ion availability and sediment–water physical and chemical conditions are major drivers of diatom functional diversification (Rusanov et al., 2024). FRic increased with conductivity, suggesting that higher ionic concentrations provide more resources and heterogeneous conditions, supporting a wider range of traits (Diaz et al., 1998; Edwards et al., 2013; Batriu et al., 2015). A similar, though weaker, effect was observed with sediment density, likely reflecting greater substrate stability (Beauchard et al., 2022). In contrast, pH showed a negative effect, probably because alkaline conditions restrict community composition to tolerant taxa. FEve was enhanced by organic matter and conductivity, consistent with studies showing that resource availability promotes functional heterogeneity (Villacorte et al., 2015; Blume et al., 2015; Passy et al., 2018; Gutierrez et al., 2020; Yan et al., 2025). Conversely, nitrate and DIC reduced FEve, likely by favoring opportunistic taxa with efficient resource-use strategies (Fasching et al., 2020). FDis increased with sediment water content, density, and oxygen, but declined under high sulfate levels. This suggests that stable, oxygenated sediments promote trait diversification, whereas sulfate selects for tolerant taxa, reducing functional variability (Pacheco et al., 2011).

Functional β diversity showed a pattern broadly consistent with that observed for the taxonomic component, with CR and CP displaying higher mean values than FG. In both cases, turnover was the main driver of the differences, while the contribution of nestedness was greater in FG. This suggests that in the CR and CP ponds, functional dissimilarity is primarily determined by the substitution of ecological traits between species, reflecting communities characterized by heterogeneous functional strategies that vary across sites. Previous studies confirm that in temporary and fragmented systems, turnover typically represents the dominant mechanism of community differentiation (Baselga, 2010; Heino et al., 2015), as fluctuating hydrological and microenvironmental conditions select for different ecological traits rather than simply causing diversity loss. Moreover, the pronounced distinction between FG and CP, already observed at the taxonomic level, confirms that these two sites host functionally divergent communities, likely due to differences in anthropogenic pressures and ecological connectivity with surrounding environments.

Community-weighted mean (CWM) analyses have strong ecological foundations. For instance, Ács et al. (2019), working on epiphytic diatoms in temporary saline ponds, demonstrated that stressful environmental gradients tend to favor functionally more compact communities, whereas less stressful conditions promote diversification in size and species composition. Broader trait-based approaches, as discussed by Weithoff & Beisner (2019), also highlight that cell size and mucilage are frequently used traits to distinguish ecological strategies related to survival, colonization, and resilience in aquatic systems subjected to environmental fluctuations. In this context, the predominance of mucilaginous and larger species in FG may reflect more stable conditions, favoring organisms with defensive and resistance strategies. By contrast, the CP environment, potentially more isolated or subject to specific stresses such as hydrological variability or nutrient limitation, appears to promote species with rapid life cycles and smaller cell sizes, traits typical of ecological colonizers (Tchabovsky, et al., 2024).

The selected physical and chemical parameters did not significantly influence the distribution or functional structure of diatom communities. Our results do not align with those of Jyrkänkallio-Mikkola et al. (2016), who reported that diatom β diversity was significantly related to the general physical habitat structure and heterogeneity. We also found that, regardless of spatial scale, β diversity was independent of nutrient levels and that higher enrichment did not affect β diversity, in contrast with the findings of Leboucher et al. (2019), who reported that diatom β diversity in streams decreases with nutrient enrichment.

Ecological interplay between plants and diatoms

The results obtained from the analysis of plant communities are consistent with the literature on temporary pond vegetation, in which water availability and sediment characteristics are key determinants of diversity. These factors influence both the establishment and survival of species under hydrological stress (Batzer & Baldwin, 2012). As reported by Della Bella et al. (2008), temporary ponds have fewer plant species than permanent ones during the flooded period, but the species found in these two pond types are highly different In particular, temporary ponds host more annual fast-growing species, such as *Callitriche spp*. and *Ranunculus spp*. The main environmental factors influencing plant richness in these ponds are generally maximum depth, surface area, dissolved oxygen, and nitrogen concentration in the water. With respect to β diversity, turnover represented the predominant component of dissimilarity between sites, while nestedness played only a minor role. Such a pattern is in line with our findings on diatom communities and with studies on temporary ecosystems, which emphasize how spatial and temporal variability in hydrological conditions promotes high turnover, reflecting differences in colonization and survival among neighboring ponds (Mašková & Poschlod, 2022; Church, 2008). Overall, plants from temporary ponds in Lazio exhibit a diversity pattern primarily driven by turnover, consistent with the ecological dynamics of these highly variable and fragmented ecosystems. Since β diversity generally results from the interplay of biogeographic, environmental, and spatial gradients, the correspondence observed between diatom and plant β diversity in this study supports our hypothesis regarding the effectiveness of diatoms as bioindicators for 3170* ponds.

The fact that the combined RDA of diatoms and vegetation with respect to standardized environmental variables was not significant indicates that the measured parameters (conductivity, pH, nutrients, sediment characteristics, etc.) explained only a small proportion of the variation in community composition. Ecologically, this may reflect several processes. First, communities in temporary ponds are often shaped by stochastic and historical dynamics, such as colonization sequence, random propagule dispersal, and flooding or drying events, that may override the influence of physical and chemical conditions (Sahuquillo & Miracle, 2013; Jeffries, 2011). Second, unquantified factors such as pond connectivity, trophic pressures (e.g., herbicides, grazing by fauna), may play a more significant role in structuring communities (Vanschoenwinkel et al., 2010; Montaña et al., 2022).

Differently, The RDA results showed that the included plants (considered as environmental variables) explained 65.4% of the diatom community variation. We demonstrated the significant contribution of *Isoëtes longissima, Callitriche stagnalis, Glyceria notata* and other dominant plant species in structuring diatom communities. CP's sites were dominated by *Callitriche stagnalis*, that is a driving force for diatom distribution. *Eunotia pectinalis* Ehrenb., *Gomphonema italicum* (Rabenh.) Kütz., *G. olivaceum* (Hornem.) C. Agardh, *G. zellense* Rabenh., *Navicula cryptocephala* Kütz., *N. phyllepta* Kütz., *Nitzschia filiformis* (Kütz.) Grunow, *N. frustulum* (Kütz.) Grunow are considered as typical species inhabiting generally unpolluted, oligotrophic and β-mesosaprobic habitats, characterized by low conductivity values, reflecting the ecological optimum of *Callitriche stagnalis*. Based on the RDA grouping of sites, the C3 site hosted few diatom species, such as *Achnanthidium eutrophilum* Hustedt, *Cymbella excisa* (Ehrenb.) Cleve, *Diatoma vulgaris* Bory, *Gomphonema minutum* (Kütz.) Grunow, *G. parvulum* (Kütz.) Grunow, well adapted to mesotrophic waters with a medium electrolyte content.

In addition, we reported a preliminary and partial assessment of the characteristic diatom species inhabiting 3170* Habitat, in relation to the typical vegetation present. As some plant species are commonly found in this Habitat, such as *Damasonium alisma* and *Isoëtes longissima*, we could hypothesize that the associated diatom speciesmay also be considered as common species in these Habitats. For instance, in our study, *Isoëtes longissima*, is often associated with *Encyonema silesiacum* (Grunow) Krammer, *Gomphonema exilissimum* Kütz., *Navicula capitatoradiata* Hustedt and *Pinnularia viridiformis* W. Smith. We suggest that this approach could be replicated at European level to identify the characteristic/diagnostic diatom species of 3170*, thus filling a significant ecological knowledge gap. Further research should therefore apply this sampling method to different 3170* ponds to identify diatom species typical of 3170* Habitat.

Finally, the study of 13 sites of the habitat 3170*MTPs in the same geographical area (Lazio) allowed us to demonstrate that Foglino wood and Ceprano wood host similar plant and diatom species. While the two sites differ significantly in terms of protection measures (Foglino wood is a Natura 2000 site, Ceprano wood is not included in any protected area), both sites exhibit a similar number of plant species and Pielou's index values. Protected areas, such as those involved in the Nature 2000 network, have been shown to be valuable instruments for species conservation and habitat management, thereby enhancing the overall quality of target ecosystems (Chape et al., 2005; Trochet & Schmeller, 2013). Nevertheless, the lack of routine and targeted management in the Foglino wood means that it only functions as a Natura 2000 site only in legal terms, failing to effectively ensure species conservation or habitat quality. The numerous plant and diatom communities shared by the two sites highlight their similar conservation status and ecological conditions. On the other hand, the Castelporziano site turned out to be well-preserved ponds, reflecting the high level of protection within the Reserve. In particular, the diatom and plant assemblages testify to the undisturbed nature of the Reserve, and, according to the β diversity analysis, show dissimilar communities in terms of species richness compared to the other two sites. These differences also reflect varying levels of protection among the sites, which were used as a proxy for human impact. Although a quantitative assessment of anthropogenic pressure was beyond the scope of this study, we acknowledge that including such a metric would be valuable for future research. Our results emphasize the significant impact that human activities can have on the biotic communities of MTPs (Taurozzi and Scalici, 2024). The increasing competition from more commonplace plant species (Zacharias & Zamparas, 2010), the replacement of rare and sensitive diatom species that are characteristic of MTPs (Blanco et al., 2020) and the introduction of alien species (Van den Broeck et al., 2015) are the main threats to the survival of MTPs plant communities. Otherwise, Red List diatom species were equally represented across all three sites.

However, insights on effects of climate change of 3170* Habitat began only few years ago. From the effects of hydrological regimes on diatom and plant communities, to the abiotic variations and their influences on biota inhabiting MTPs, the extent to which MPs' bioindicators are predictably impacted requires knowledge of how the relevant physical and physiological features of 3170* act in terms of intensity and frequency. In this sense, here we provide the first evidence that diatoms can be considered ecologically relevant bioindicators for complementary or alternative studies on 3170* communities using vegetation analysis.

Limitations of the study

Although our study was conducted using rigorous sampling and analytical procedures, certain limitations must be acknowledged, primarily concerning site selection and seasonal constraints. The selection of a heterogeneous number of MTPs in the three sampling sites was driven by practical needs: Only MTPs showing a constant water supply during the winter were selected. We excluded very shallow ponds (maximum depth lower than 10 cm) and temporary flooded meadows with extremely reduced hydroperiods, where water levels are too low for reliable diatom sampling and communities cannot properly develop despite the presence of typical 3170* vegetation. Many CP ponds fall into this category, and many others were excluded due to the absence of typical 3170* vegetation; for this reason, only three pond sites met the minimum depth and typical vegetation requirements to be investigated. We also acknowledge that the number of ponds included in this study is limited, and that further research should extend sampling to additional sites. Logistical and economic constraints meant that a broader geographic coverage was not possible in the present study. Furthermore, it must be emphasised that the plant community studied is not representative of all the variability of species that are present in these environments. The floristic contingent of therophytes and dwarf plants did not develop in the spring. Thus, diatoms represent a possible bioindicators in a specific typology of MTPs.

Conclusions

Studies of MTPs have revealed significant knowledge gaps among different taxonomic groups. Aquatic and non-aquatic vascular plants are among the most widely studied organisms and are frequently used as MTPs indicators. However, data from the scientific literature reveal that, although the assessment of diatom contribution to evaluating the ecological status of MTPs but highly sensitive and specific, providing exhaustive data about environmental changes, it is poorly exploited. Our research is the first to present and define the ecological relevance of diatoms and vascular plants in MTPs in a similar way. The proposed methodology could facilitate the identification of typical diatom species inhabiting 3170* Habitats, making conservation activities more effective and straightforward in practice. Diatoms can be considered an early warning system as they develop rapidly and respond rapidly to stress. This research could lay the groundwork for the future use of diatoms as indicators and proxies for functional monitoring. It was also observed that physical and chemical parameters varied significantly between ponds under different ecological scenarios. Our findings suggest that the protection measures are crucial for determining the plant diversity and diatom autecology when using diatoms as biomonitoring agents in water quality assessments in the Mediterranean Region of Lazio. This would have implications for the development of tools for water resources included in the Natura 2000 network. However, although some of the sites included were part of the Natura 2000 network, the institutional negligence, legal loopholes and anthropogenic impacts rendered this type of protection ineffective: plant and diatom communities showed comparable ecological responses to human threats, highlighting the similarities among communities in unprotected sites. In contrast, the plant and diatom communities included in the Protected Reserve significantly differed from those in unprotected sites. The proposed approach could be used to generate a comprehensive dataset of diatoms typical of 3170* Habitats which would be useful for monitoring spatial–temporal changes, as well as the anthropogenic impacts in ponds both inside and outside the Natura 2000 network. Given the novelty of this research, further investigations into the ecological relevance of plants and diatoms in MTPs are certainly needed. The ecological correspondence between these two biological indicators could provide important conservation insights for stakeholders and contribute to long-term socio-environmental monitoring plans for protected areas.

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Author contributions Davide Taurozzi: Conceptualization, Validation, Investigation, Visualization, Formal Analysis, Data curation, Writing—original draft, Writing—review & editing. Massimiliano Scalici: Conceptualization, Resources, Supervision, Validation, Funding acquisition, Project administration, Writing—review & editing. Romeo Di Pietro: Writing—original draft, Supervision, Validation, Writing – review & editing. Rossano Bolpagni: Conceptualization, Investigation, Visualization, Writing—original draft, Supervision, Writing—review & editing. Mattia Martin Azzella: Conceptualization, Investigation, Visualization, Funding acquisition, Writing —original draft, Project administration, Supervision, Writing—review & editing.

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Data availability The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Conflict of interest The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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References

Ács, É., A. Földi, C. F. Vad, Z. Trábert, K. T. Kiss, M. Duleba & Z. Botta-Dukát, 2019. Trait-based community assembly of epiphytic diatoms in saline astatic ponds: a test of the stress-dominance hypothesis. Scientific Reports 9(1): 15749. https://doi.org/10.1038/s41598-019-52304-4.

Akhtar, N., Syakir Ishak, M. I., Bhawani, S. A., & Umar, K., 2021. Various natural and anthropogenic factors responsible for water quality degradation: a review. Water, 13(19), 2660. https://www.mdpi.com/2073-4441/13/19/2660#.

Ansari, A. A., Shalini Saggu, S. S., Al-Ghanim, S. M., Abbas, Z. K., Gill, S. S., Khan, F. A., ... & Khan, A. A., 2017. Aquatic plant biodiversity: a biological indicator for the monitoring and assessment of water quality. In Plant biodiversity: monitoring, assessment and conservation (pp. 218–227). Wallingford UK: CABI. https://doi.org/10.1079/9781780646947.0218.

Anzalone, B., E. Lattanzi, F. Lucchese & M. Padula, 1997. Flora vascolare del parco nazionale del circeo (Lazio). Webbia 51(2): 251–341.

Aspila, K. I., H. Agemian & A. S. Y. Chau, 1976. A semiautomated method for the determination of inorganic, organic and total phosphate in sediments. Analyst 101: 187–197. https://doi.org/10.1039/AN9760100187.

Bagella, S., 2023. Which perspectives for Mediterranean temporary ponds in the European Union in the third millennium? Biodivers. Conserv. 32(3): 839–849. https://doi.org/10.1007/s10531-023-02549-3.

Bagella, S. & M. C. Caria, 2012. Diversity and ecological characteristics of vascular flora in Mediterranean temporary pools. Comptes Rendus Biologies 335(1): 69–76. https://doi.org/10.1016/j.crvi.2011.10.005.

Bagella, S., M. C. Caria, E. Farris & R. S. Filigheddu, 2007. Issues related to the classification of Mediterranean temporary wet habitats according with the European union habitats directive. Fitosociologia 44(2-suppl. 1): 245–249.

Bagella, S., Aleffi, M., Azzella, M.M., Bolpagni, R., Gigante, D., Maneli, F., Poponessi, S., Puglisi M., 2016. 3170 * Stagni temporanei mediterranei. In: Angelini P., Casella L., Grignetti A., Genovesi P. (ed.), 2016. Manuali per il monitoraggio di specie e habitat di interesse comunitario (Direttiva 92/43/CEE) in Italia: habitat. ISPRA, Serie Manuali e linee guida, 142/2016

Baguette, M., S. Blanchet, D. Legrand, V. M. Stevens & C. Turlure, 2013. Individual dispersal, landscape connectivity and ecological networks. Biological Reviews 88(2): 310–326. https://doi.org/10.1111/brv.12000.

Bartolucci, F., L. Peruzzi, G. Galasso, A. Albano, A. Alessandrini, N. M. G. Ardenghi, et al., 2018. An updated checklist of the vascular flora native to Italy. Plant Biosystems 152(2): 179–303. https://doi.org/10.1080/11263504.2017.1419996.

Baselga, A., 2010. Partitioning the turnover and nestedness components of beta diversity. Global Ecol. Biogeogr. 19(1): 134–143.

Baselga, A., Orme, D., Villeger, S., De Bortoli, J., Leprieur, F., Logez, M., Martinez-Santalla, S., Martin-Devasa, R., Gomez-Rodriguez, C., Crujeiras, R., 2025. Betapart: partitioning beta diversity into turnover and nestedness components. R package version 1.6.1, https://CRAN.R-project.org/package=betapart.

Batriu, E., J. M. Ninot & J. Pino, 2015. Filtering of plant functional traits is determined by environmental gradients and by past land use in a M editerranean coastal marsh. J. Veg. Sci. 26(3): 492–500. https://doi.org/10.1111/jvs.12251.

Batzer, D. P., & Baldwin, A. H. (Eds), 2012. Wetland habitats of North America: ecology and conservation concerns. Univ of California Press.

Bazzanti, M., M. Seminara, S. Baldoni & A. Stella, 2000. Macroinvertebrates and environmental factors of some temporary and permanent ponds in Italy. Internationale Vereinigung Für Theoretische und Angewandte Limnologie: Verhandlungen 27(2): 936–941. https://doi.org/10.1080/03680770.1998.11901377.

B-Béres, V., C. Stenger-Kovács, K. Buczkó, J. Padisák, G. B. Selmeczy, E. Lengyel & K. Tapolczai, 2023. Ecosystem services provided by freshwater and marine diatoms. Hydrobiologia 850(12): 2707–2733. https://doi.org/10.1007/s10750-022-04984-9.

Beauchard, O., S. Mestdagh, L. Koop, T. Ysebaert & P. M. Herman, 2022. Benthic synecology in a soft sediment shelf: habitat contrasts and assembly rules of life strategies. Marine Ecology Progress Series 682: 31–50. https://doi.org/10.3354/meps13928.

Beja, P. & R. Alcazar, 2003. Conservation of Mediterranean temporary ponds under agricultural intensification: an evaluation using amphibians. Biological Conservation 114(3): 317–326. https://doi.org/10.1016/S0006-3207(03)00051-X.

Biondi, E., Blasi, C., Burrascano, S., Casavecchia, S., Copiz, R., Del Vico, E., Galdenzi, D., Gigante, D., Lasen, C., Spampinato, G., Venanzoni, R., & L. Zivkovic, 2009. Manuale Italiano di Interpretazione degli Habitat della Direttiva 92/43/CEE. http://vnr.unipg.it/habitat/index.jsp

Blanco, S., A. Olenici, F. Ortega, F. Jiménez-Gómez & F. Guerrero, 2020. Identifying environmental drivers of benthic diatom diversity: the case of Mediterranean mountain ponds. PeerJ 8: e8825. https://doi.org/10.7717/peerj.8825.

Blasi, C., Stanisci, A., Filesi, L., Milanese, A., Perinelli, E., & Riggio, L., 2002. Syndinamics of lowland Quercus frainetto & Q. cerris forests in Lazio (central Italy). Fitosociologia, 39(1): 23–43.

Blume, H. P., G. W. Brümmer, H. Fleige, R. Horn, E. Kandeler, I. Kögel-Knabner & B. M. Wilke, 2015. Soil organisms and their habitat, Scheffer/schachtschabel soil science. Berlin, Heidelberg, Springer, Berlin Heidelberg. https://doi.org/10.1007/978-3-642-30942-7_4.

Bolpagni, R., S. Poikane, A. Laini, S. Bagella, M. Bartoli & M. Cantonati, 2019. Ecological and conservation value of small standing-water ecosystems: a systematic review of current knowledge and future challenges. Water 11(3): 402. https://doi.org/10.3390/w11030402.

Buchanan, J. B., Holme, N. A., & McIntyre, A. D. (1984). Methods for the study of marine benthos, p. 41–65. In IBP Handbook. Blackwell Scientific Publications.

Burns, K. C., 2019. Evolution in isolation: the search for an island syndrome in plants, Cambridge University Press:

Burrascano, S., R. Copiz, E. Del Vico, S. Fagiani, E. Giarrizzo, M. Mei & C. Blasi, 2015. Wild boar rooting intensity determines shifts in understorey composition and functional traits. Community Ecology 16(2): 244–253. https://doi.org/10.1556/168.2015.16.2.12.

Cantonati, M., L. Füreder, R. Gerecke, I. Jüttner & E. J. Cox, 2012. Crenic habitats, hotspots for freshwater biodiversity conservation: toward an understanding of their ecology. Freshwater Sci. 31(2): 463–480. https://doi.org/10.1899/11-065.1.

Caria, M. C., P. Grillas, G. Rivieccio & S. Bagella, 2021. Effect of inter-annual meteorological fluctuations and wild boar disturbance on the vegetation of Mediterranean temporary ponds. Appl. Veg. Sci. 24(4): e12624. https://doi.org/10.1111/avsc.12624.

Carta, A., 2016. Seed regeneration in Mediterranean temporary ponds: germination ecophysiology and vegetation processes. Hydrobiologia 782: 23–35. https://doi.org/10.1007/s10750-016-2808-5.

Castracani, C., D. A. Grasso, A. Fanfani, et al., 2010. The ant fauna of Castelporziano presidential reserve (Rome, Italy) as a model for the analysis of ant community structure in relation to environmental variation in Mediterranean ecosystems. J. Insect Conserv. 14: 585–594. https://doi.org/10.1007/s10841-010-9285-3.

Chape, S., J. Harrison, M. Spalding & I. Lysenko, 2005. Measuring the extent and effectiveness of protected areas as an indicator for meeting global biodiversity targets. Philos. Trans. Royal Soc. B: Biol. Sci. 360(1454): 443–455. https://doi.org/10.1098/rstb.2004.1592.

Chen, X., Z. Bu, M. A. Stevenson, Y. Cao, L. Zeng & B. Qin, 2016. Variations in diatom communities at genus and species levels in peatlands (central China) linked to microhabitats and environmental factors. Sci. Total Environ. 568: 137–146. https://doi.org/10.1016/j.scitotenv.2016.06.015.

Church, D. R., 2008. Role of current versus historical hydrology in amphibian species turnover within local pond communities. Copeia 2008(1): 115–125.

Coccia, C., B. A. Almeida, A. Badosa, L. P. Diniz, L. Brendonck, D. Frisch & A. J. Green, 2024. Hydroperiod length, not pond age, determines zooplankton taxonomic and functional diversity in temporary ponds. Ecol. Indic. 159: 111632. https://doi.org/10.1016/j.ecolind.2024.111632.

Cunillera-Montcusí, D., M. Arim, S. Gascón, I. Tornero, J. Sala, D. Boix & A. I. Borthagaray, 2020. Addressing trait selection patterns in temporary ponds in response to wildfire disturbance and seasonal succession. J. Animal Ecol. 89(9): 2134–2144. https://doi.org/10.1111/1365-2656.13265.

De Nicola, C., A. Zanella, A. Testi, G. Fanelli & S. Pignatti, 2014. Humus forms in a Mediterranean area (Castelporziano Reserve, Rome, Italy): classification, functioning and organic carbon storage. Geoderma 235: 90–99. https://doi.org/10.1016/j.geoderma.2014.06.033.

Della Bella, V. & L. Mancini, 2009. Freshwater diatom and macroinvertebrate diversity of coastal permanent ponds along a gradient of human impact in a Mediterranean eco-region. Hydrobiologia 634(1): 25–41. https://doi.org/10.1007/s10750-009-9890-x.

Della Bella, V., M. Bazzanti & F. Chiarotti, 2005. Macroinvertebrate diversity and conservation status of Mediterranean ponds in Italy: water permanence and mesohabitat influence. Aquatic Conserv.: Marine Freshwater Ecosyst. 15(6): 583–600.

Della Bella, V., M. Bazzanti, M. G. Dowgiallo & M. Iberite, 2008. Macrophyte diversity and physico-chemical characteristics of Tyrrhenian coast ponds in central Italy: implications for conservation. Hydrobiologia 597(1): 85–95.

Della Bella, V., Puccinelli, C., Marcheggiani, S., & Mancini, L., 2007. Benthic diatom communities and their relationship to water chemistry in wetlands of central Italy. In Annales de Limnologie-International Journal of Limnology (Vol. 43, No. 2, pp. 89–99). EDP Sciences.

Dey, S., S. Botta, R. Kallam, R. Angadala & J. Andugala, 2021. Seasonal variation in water quality parameters of Gudlavalleru engineering college pond. Curr. Res. Green Sustain. Chem. 4: 100058. https://doi.org/10.1016/j.crgsc.2021.100058.

Diaz, S., M. Cabido & F. Casanoves, 1998. Plant functional traits and environmental filters at a regional scale. J. Veg. Sci. 9(1): 113–122. https://doi.org/10.2307/3237229.

E.P.C.N., 2008. The Pond Manifesto. Stagni e zone umide minori: un manifesto per la loro conoscenza e conser vazione. Provincia di Roma, Assessorato alle Politiche dell'Agricoltura. Gangemi editore, Roma. https://www.europeanponds.org/wp-content/uploads/2014/12/EPCN-manifesto_italian.pdf.

Ector, L., Hlúbiková, D., 2010. Atlas des diatomées des Alpes-Maritimes et de la Région Provence-Alpes-Côte d'Azur.

Edwards, K. F., E. Litchman & C. A. Klausmeier, 2013. Functional traits explain phytoplankton responses to environmental gradients across lakes of the United States. Ecology 94(7): 1626–1635. https://doi.org/10.1890/12-1459.1.

Epele, L. B., C. Brand & M. L. Miserendino, 2019. Ecological drivers of alpha and beta diversity of freshwater invertebrates in arid and semiarid Patagonia (Argentina). Sci. Total Environ. 678: 62–73. https://doi.org/10.1007/s10750-025-06080-0.

European Environment Agency (EEA), 2020. State of nature in the EU–results from reporting under the nature directives 2013–2018. Pub. Office European Union. https://doi.org/10.2800/088178.

Fasching, C., C. Akotoye, M. Bižić, J. Fonvielle, D. Ionescu, S. Mathavarajah & M. A. Xenopoulos, 2020. Linking stream microbial community functional genes to dissolved organic matter and inorganic nutrients. Limnol. Oceanography 65: S71–S87. https://doi.org/10.1002/lno.11356.

Fehlinger, L., B. Misteli, D. Morant, N. Juvigny-Khenafou, D. Cunillera-Montcusí, F. Chaguaceda & B. Rimcheska, 2023. The ecological role of permanent ponds in Europe: a review of dietary linkages to terrestrial ecosystems via emerging insects. Inland Waters 13(1): 30–46. https://doi.org/10.1080/20442041.2022.2111180.

Fehlinger, L., Rimcheska, B., Rubio-Ríos, J., Fenoy, E., Mathieu-Resuge, M., Sarkezi, M., ... & Fekete, J. (2021). 3rd European FreshProject "EUROPONDS". In Libro de Resumenes: IX Simposio de Invetigación en Ciencias Experimentales, 13 November 2020, Almería, Spain.

Filibeck, G., & Lattanzi, E., 2008. Il Poligono Militare di Nettuno (prov. Roma), testimonianza di un paesaggio scomparso: contributo alla conoscenza floristica e proposte per la tutela.

Filippino, G., 2018. Distribution, ecology and conservation of bryoflora in the Mediterranean temporary ponds.

Florencio, M., R. Fernández-Zamudio, M. Lozano & C. Díaz-Paniagua, 2020. Interannual variation in filling season affects zooplankton diversity in Mediterranean temporary ponds. Hydrobiologia 847(5): 1195–1205. https://doi.org/10.1007/s10750-019-04163-3.

Fu, B., P. Xu, Y. Wang, K. Yan & S. Chaudhary, 2018. Assessment of the ecosystem services provided by ponds in hilly areas. Science of the Total Environment 642: 979–987. https://doi.org/10.1016/j.scitotenv.2018.06.138.

Gigante, D., Maneli, F., & Venanzoni, R. (2007). Aspetti connessi all'interpretazione e alla gestione degli Habitat della Dir. 92/43/EEC in Umbria. Fitosociologia, 44(2 (S1)), 141–146.

Grillas, P., Gauthier, P., Yavercovski, N., & Perennou, C. (2004). Les mares temporaires méditerranéennes: Volume 1–Enjeux de conservation, fonctionnement et gestion. Station biologique de la Tour du Valat. 122 pp.

Guo, K., N. Wu, P. Manolaki, A. Baattrup-Pedersen & T. Riis, 2020. Short-period hydrological regimes override physico-chemical variables in shaping stream diatom traits, biomass and biofilm community functions. Sci. Total Environ. 743: 140720. https://doi.org/10.1016/j.scitotenv.2020.140720.

Gutierrez, M. F., N. R. Simões, D. Frau, M. Saigo & M. Licursi, 2020. Responses of stream zooplankton diversity metrics to eutrophication and temporal environmental variability in agricultural catchments. Environ. Monit. Assess. 192(12): 792. https://doi.org/10.1007/s10661-020-08766-5.

Heino, J., J. Soininen, J. Alahuhta, J. Lappalainen & R. Virtanen, 2015. A comparative analysis of metacommunity types in the freshwater realm. Ecol. Evolut. 5(7): 1525–1537. https://doi.org/10.1002/ece3.1460.

Hill, M. J., J. Heino, J. C. White, D. B. Ryves & P. J. Wood, 2019. Environmental factors are primary determinants of different facets of pond macroinvertebrate alpha and beta diversity in a human-modified landscape. Biol. Conserv. 237: 348–357. https://doi.org/10.1016/j.biocon.2019.07.015.

Hubbard, R. K., G. L. Newton & G. M. Hill, 2004. Water quality and the grazing animal. J. Animal Sci. 82(suppl_13): E255–E263. https://doi.org/10.2527/2004.8213_supplE255x.

ISPRA, 2014. Metodi biologici per le acque superficiali interne. In: Manuali e Linee Guida, 111th edn. ISPRA, Roma, p 234.

Jeffries, M. J., 2011. The temporal dynamics of temporary pond macroinvertebrate communities over a 10-year period. Hydrobiologia 661(1): 391–405. https://doi.org/10.1007/s10750-010-0551-x.

Ji, L., Y. Li, G. Zhang & Y. Bi, 2021. Anthropogenic disturbances have contributed to degradation of river water quality in arid areas. Water 13(22): 3305. https://doi.org/10.3390/w13223305.

Jyrkänkallio-Mikkola, J., J. Heino & J. Soininen, 2016. Beta diversity of stream diatoms at two hierarchical spatial scales: implications for biomonitoring. Freshwater Biol. 61(2): 239–250. https://doi.org/10.1111/fwb.12697.

Krammer, K., Lange-bertalot, H., 1986. Bacillariophyceae, 1.Teil : Naviculaceae, Band 2/1, Subwasserflora Von Mitteleuropa, Veb Gustav Fischer Verlag, pp 876.

Krammer, K., Lange-bertalot, H., 1988. Bacillariophyceae, 2.Teil : Bacillariophyceae, Epithemiaceae, Surirellaceae, Band 2/2, Subwasserflora Von Mitteleuropa, Veb Gustav Fischer Verlag, pp. 595.

Krammer, K., Lange-bertalot, H., 1991a. Bacillariophyceae, 3.Teil : Centrales, Fragilariaceae, Eunotiaceaeceae, Band 2/3, Subwasserflora Von Mitteleuropa, Veb Gustav Fischer Verlag, pp. 576

Krammer, K., Lange-bertalot, H., 1991b. Bacillariophyceae, 4.Teil : Achnanthaceae, Kritische Ergänzungen zu Navicula (Lineolatae) und Gomphonema , Band 2/4, Subwasserflora Von Mitteleuropa, Veb Gustav Fischer Verlag, Jena.

Krammer, K., 2002. Diatoms of the European inland waters and comparable habitats. In: Lange-Bertalot (ed) Cymbella, vol 3. ISBN 978–3–904144–84–1

Krivtsov, V., H. Forbes, S. Birkinshaw, V. Olive, D. Chamberlain, J. Buckman & C. Diekonigin, 2022. Ecosystem services provided by urban ponds and green spaces: a detailed study of a semi-natural site with global importance for research. Blue-Green Syst. 4(1): 1–23. https://doi.org/10.2166/bgs.2022.021.

Laliberté, E., Legendre, P., Shipley, B., & Laliberté, M. E., 2014. Package ‘fd’. Measuring functional diversity from multiple traits, and other tools for functional ecology, 1, 0–12.

Lattanzi, E., E. Perinelli & L. Riggio, 2004. Flora vascolare del bosco di Foglino (Nettuno-Roma). Inform. Bot. Ital 36(2): 337–361.

Lattanzi, E., Tilia, A., Bonacquisti, S., Capotorti, G., Del Vico, E., Facioni, L., ... & Blasi, C., 2021. La flora della Tenuta Presidenziale di Castelporziano. Censimento, pressioni e presenza di specie alloctone. In: Il sistema ambientale della tenuta presidenziale di Castelporziano. Ricerche sulla complessità di un ecosistema forestale costiero mediterraneo (pp. 437–469). Accademia Nazionale delle Scienze detta dei Quaranta

Leboucher, T., W. R. Budnick, S. I. Passy, S. Boutry, A. Jamoneau, J. Soininen & J. Tison-Rosebery, 2019. Diatom β-diversity in streams increases with spatial scale and decreases with nutrient enrichment across regional to sub-continental scales. J. Biogeogr. 46(4): 734–744. https://doi.org/10.1111/jbi.13517.

Letáková, M., M. Fránková & A. Poulíčková, 2018. Ecology and applications of freshwater epiphytic diatoms—review. Cryptogamie, Algologie 39(1): 3–22. https://doi.org/10.7872/crya/v39.iss1.2018.3.

Lozada-Gobilard, S., S. Stang, K. Pirhofer-Walzl, T. Kalettka, T. Heinken, B. Schröder & J. Joshi, 2019. Environmental filtering predicts plant-community trait distribution and diversity: kettle holes as models of meta-community systems. Ecol. Evolut. 9(4): 1898–1910. https://doi.org/10.1002/ece3.4883.

Lucchese, F., 1990. La flora della riserva naturale di Palo Laziale (Roma). *Annali di Botanica*, *48*.

Lumbreras, A., J. T. Marques, A. F. Belo, M. Cristo, M. Fernandes, D. Galioto & C. Pinto-Cruz, 2016. Assessing the conservation status of Mediterranean temporary ponds using biodiversity: a new tool for practitioners. Hydrobiologia 782: 187–199. https://doi.org/10.1007/s10750-016-2697-7.

Maitland, V. C., C. V. Robinson, T. M. Porter & M. Hajibabaei, 2020. Freshwater diatom biomonitoring through benthic kick-net metabarcoding. PLoS One 15(11): e0242143. https://doi.org/10.1371/journal.pone.0242143.

Mangadze, T., R. J. Wasserman & T. Dalu, 2017. Use of diatom communities as indicators of conductivity and ionic composition in a small austral temperate river system. Water, Air, Soil Pollution 228(11): 428. https://doi.org/10.1007/s11270-017-3610-3.

Mašková, T. & P. Poschlod, 2022. Hydrology regime and hydrology recruitment niche predict changes in plant community composition across a pond habitat. J. Veg. Sci. 33(6): e13164. https://doi.org/10.1111/jvs.13164.

Masouras, A., I. Karaouzas, E. Dimitriou, G. Tsirtsis & E. Smeti, 2021. Benthic diatoms in river biomonitoring—present and future perspectives within the water framework directive. Water 13(4): 478. https://doi.org/10.3390/w13040478.

Meerhoff, M., & Beklioğlu, M., 2024. Shallow lakes and ponds. In Wetzel's Limnology (pp. 859–892). Academic Press. https://doi.org/10.1016/B978-0-12-822701-5.00026-4.

Michel, M. J. & J. H. Knouft, 2014. The effects of environmental change on the spatial and environmental determinants of community-level traits. Landscape Ecol. 29(3): 467–477. https://doi.org/10.1007/s10980-013-9977-7.

Minutillo, F., G. Tondi & R. Di Pietro, 2014. Contributo alla conoscenza della flora della Media Valle Latina (Appennino Centrale). Informatore Botanico Italiano 46(1): 47–52.

Montaña, C. G., F. W. Keppeler, C. P. Laughrey & C. M. Schalk, 2022. Community assembly within ponds: the roles of space, time, and environmental gradients. Aquatic Ecol. 56(1): 123–142. https://doi.org/10.1007/s10452-021-09902-0.

Mucina, L., H. Bültmann, K. Dierßen, J. P. Theurillat, T. Raus, A. Čarni & L. Tichý, 2016. Vegetation of Europe: hierarchical floristic classification system of vascular plant, bryophyte, lichen, and algal communities. Appl. Veg. Sci. 19: 3–264. https://doi.org/10.1111/avsc.12257.

Mullu, D., 2016. A review on the effect of habitat fragmentation on ecosystem. J. Nat. Sci. Res. 6(15): 1–15.

Nemes-Kokai, Z., G. Borics, E. Csépes, A. Lukacs, P. Török, E. T-Krasznai & V. B-Béres, 2024. Role of microhabitats in shaping diversity of periphytic diatom assemblages. Hydrobiologia 851(4): 959–972. https://doi.org/10.1007/s10750-023-05336-x.

Oksanen, J., Simpson, G., Blanchet, F., Kindt, R., Legendre, P., Minchin, P., O'Hara, R., et al., 2025. vegan: Community Ecology Package. R package version 2.8–0, https://github.com/vegandevs/vegan.

Olmo, C., Á. Gálvez, M. Bisquert-Ribes, F. Bonilla, C. Vega, A. Castillo-Escrivà & F. Mesquita-Joanes, 2022. The environmental framework of temporary ponds: A tropical-Mediterranean comparison. Catena 210: 105845. https://doi.org/10.1016/j.catena.2021.105845.

Orsini, L., J. Vanoverbeke, I. Swillen, J. Mergeay & L. De Meester, 2013. Drivers of population genetic differentiation in the wild: isolation by dispersal limitation, isolation by adaptation and isolation by colonization. Mol. Ecol. 22(24): 5983–5999. https://doi.org/10.1111/mec.12561.

Ouyang, Y., P. Nkedi-Kizza, Q. T. Wu, D. Shinde & C. H. Huang, 2006. Assessment of seasonal variations in surface water quality. Water Res. 40(20): 3800–3810. https://doi.org/10.1016/j.watres.2006.08.030.

Pacheco, A. S., M. T. González, J. Bremner, M. Oliva, O. Heilmayer, J. Laudien & J. M. Riascos, 2011. Functional diversity of marine macrobenthic communities from sublittoral soft-sediment habitats off northern Chile. Helgoland Marine Res. 65(3): 413–424. https://doi.org/10.1007/s10152-010-0238-8.

Passy, S. I., C. A. Larson, A. Jamoneau, W. Budnick, J. Heino, T. Leboucher & J. Soininen, 2018. Biogeographical patterns of species richness and abundance distribution in stream diatoms are driven by climate and water chemistry. The Am. Nat. 192(5): 605–617.

Pestryakova, L. A., U. Herzschuh, R. Gorodnichev & S. Wetterich, 2018. The sensitivity of diatom taxa from Yakutian lakes (north-eastern Siberia) to electrical conductivity and other environmental variables. Polar Res. 37(1): 1485625. https://doi.org/10.1080/17518369.2018.1485625.

R Core Team, 2024. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org/.

Riato, L., V. Della Bella, M. Leira, J. C. Taylor & P. J. Oberholster, 2017. A diatom functional-based approach to assess changing environmental conditions in temporary depressional wetlands. Ecol. Indic. 78: 205–213. https://doi.org/10.1016/j.ecolind.2017.03.018.

Richardson, D. C., M. A. Holgerson, M. J. Farragher, K. K. Hoffman, K. B. King, M. B. Alfonso & J. N. Sweetman, 2022. A functional definition to distinguish ponds from lakes and wetlands. Scientific Reports 12(1): 10472. https://doi.org/10.1038/s41598-022-14569-0.

Rimet, F. & A. Bouchez, 2011. Use of diatom life-forms and ecological guilds to assess pesticide contamination in rivers: lotic mesocosm approaches. Ecol. Indic. 11(2): 489–499. https://doi.org/10.1016/j.ecolind.2010.07.004.

Rimet, F. & A. Bouchez, 2012. Life-forms, cell-sizes and ecological guilds of diatoms in European rivers. Knowl. Manag. Aquatic Ecosyst. 406: 01. https://doi.org/10.1051/kmae/2012018.

Rudnick, D., Ryan, S. J., Beier, P., Cushman, S. A., Dieffenbach, F., Epps, C., ... & Trombulack, S. C., 2012. The role of landscape connectivity in planning and implementing conservation and restoration priorities. Issues in Ecology.

Rusanov, A. G., E. A. Kurashov, A. M. Rasulova, T. Bíró, E. Vadkerti & É. Ács, 2024. Diatom metacommunity structuring in a large lake: geomorphic, water chemistry and dispersal effects on diatom guilds in Lake Ladoga (northwestern Russia). Aquat. Sci. 86(2): 39. https://doi.org/10.1007/s00027-024-01055-0.

Sahuquillo, M. & M. R. Miracle, 2013. The role of historic and climatic factors in the distribution of crustacean communities in I berian M editerranean ponds. Freshwater Biol. 58(6): 1251–1266. https://doi.org/10.1111/fwb.12124.

Segura, A., & Palomar, G., 2023. Hydroperiod of temporary ponds threats amphibian recruitment in Mediterranean environments. Amphibia-Reptilia, 1(aop), 1–10. https://doi.org/10.1163/15685381-bja10146.

Soininen, J. & A. Teittinen, 2019. Fifteen important questions in the spatial ecology of diatoms. Freshwater Biol. 64(11): 2071–2083. https://doi.org/10.1111/fwb.13384.

Stanisci, A., A. Acosta, V. Gargini, F. Fiore & C. Blasi, 2001. Zonazione della vegetazione nelle "piscine" dei querceti planiziari del Lazio. Inform. Bot. Ital. 33(2): 379–389.

Stenger-Kovács, C., & B-Béres, V., 2024. Guilds for diatoms: history and future prospects. Diatom Ecology: From

Molecules to Metacommunities, 233–279. https://doi.org/10.1002/9781394174898.ch8.

Strungaru, S. A., C. M. Pohontiu, M. Nicoara, C. Teodosiu, E. S. Baltag, R. Jijie & C. Faggio, 2021. Response of aquatic macroinvertebrates communities to multiple anthropogenic stressors in a lowland tributary river. Environ. Toxicol. Pharmacol. 87: 103687. https://doi.org/10.1016/j.etap.2021.103687.

Taurozzi, D., G. Cesarini & M. Scalici, 2023. New ecological frontiers in the plastisphere: diatoms and macroinvertebrates turnover assessment by a traits-based approach. Sci. Total Environ. 887: 164186. https://doi.org/10.1016/j.scitotenv.2023.164186.

Taurozzi, D., G. Cesarini & M. Scalici, 2024a. Diatoms as bioindicators for health assessments of ephemeral freshwater ecosystems: a comprehensive review. Ecol. Indic. 166: 112309. https://doi.org/10.1016/j.ecolind.2024.112309.

Taurozzi, D., L. Gallitelli, G. Cesarini, S. Romano, M. Orsini & M. Scalici, 2024b. Passive biomonitoring of airborne microplastics using lichens: a comparison between urban, natural and protected environments. Environ. Int. 187: 108707. https://doi.org/10.1016/j.envint.2024.108707.

Taylor, J.C., Harding, W.R., Archibald, C.G.M., 2007. An illustrated guide to some common diatom species from South Africa. Water Research Commission.

Tchabovsky, A. V., E. N. Surkova & O. N. Batova, 2024. Colonization: individual traits of colonists and population processes. Biol. Bulletin 51(7): 2221–2239. https://doi.org/10.1134/S1062359024700699.

Trochet, A. & D. S. Schmeller, 2013. Effectiveness of the Natura 2000 network to cover threatened species. Nat. Conserv. 4: 35–53.

Tuomisto, H., 2010. A diversity of beta diversities: straightening up a concept gone awry. Part 2 Quantifying beta diversity and related phenomena. Ecography. 33(1): 23–45. https://doi.org/10.1111/j.1600-0587.2009.06148.x.

Urrea, G. & S. Sabater, 2009. Epilithic diatom assemblages and their relationship to environmental characteristics in an agricultural watershed (Guadiana River, SW Spain). Ecol. Indic. 9(4): 693–703. https://doi.org/10.1016/j.ecolind.2008.09.002.

Van den Broeck, M., A. Waterkeyn, L. Rhazi, P. Grillas & L. Brendonck, 2015. Assessing the ecological integrity of endorheic wetlands, with focus on Mediterranean temporary ponds. Ecol. Indic. 54: 1–11. https://doi.org/10.1016/j.ecolind.2015.02.016.

Vanschoenwinkel, B., A. Waterkeyn, M. Jocqué, L. Boven, M. Seaman & L. Brendonck, 2010. Species sorting in space and time—the impact of disturbance regime on community assembly in a temporary pool metacommunity. J. North Am. Benthol. Soc. 29(4): 1267–1278.

Villacorte, L. O., Y. Ekowati, T. R. Neu, J. M. Kleijn, H. Winters, G. Amy & M. D. Kennedy, 2015. Characterisation of algal organic matter produced by bloom-forming marine and freshwater algae. Water Res. 73: 216–230. https://doi.org/10.1016/j.watres.2015.01.028.

Wei, T., Simko, V., 2024. R package 'corrplot': Visualization of a Correlation Matrix. (Version 0.95), https://github.com/taiyun/corrplot.

Weithoff, G. & B. E. Beisner, 2019. Measures and approaches in trait-based phytoplankton community ecology–from freshwater to marine ecosystems. Front. Marine Sci. 6: 40. https://doi.org/10.3389/fmars.2019.00040.

Wheeler, C. R., C. R. Gervais, M. S. Johnson, S. Vance, R. Rosa, J. W. Mandelman & J. L. Rummer, 2020. Anthropogenic stressors influence reproduction and development in elasmobranch fishes. Rev. Fish Biol. Fish. 30(2): 373–386. https://doi.org/10.1007/s11160-020-09604-0.

Wickham, H., François, R., Henry, L., Müller, K., Vaughan, D., 2025. Dplyr: a grammar of data manipulation. R package version 1.1.4, https://dplyr.tidyverse.org.

Yan, H., H. F. Guo, J. X. Li, Z. Chen, S. Q. Yin, M. Y. Shen & H. Y. Hu, 2025. Deciphering the contribution of algae-rich microbial communities to dissolved organic matter in a eutrophic lake with distinct rainy and dry seasons. Algal Res. 88: 104037. https://doi.org/10.1016/j.algal.2025.104037.

Zacharias, I. & M. Zamparas, 2010. Mediterranean temporary ponds a disappearing ecosystem. Biodivers. Conserv. 19: 3827–3834. https://doi.org/10.1007/s10531-010-9933-7.

Zacharias, I., E. Dimitriou, A. Dekker & E. Dorsman, 2007. Overview of temporary ponds in the Mediterranean region: threats, management and conservation issues. J. Environ. Biol. 28(1): 1–9.

Zorzal-Almeida, S., L. M. Bini & D. C. Bicudo, 2017. Beta diversity of diatoms is driven by environmental heterogeneity, spatial extent and productivity. Hydrobiologia 800: 7–16. https://doi.org/10.1007/s10750-017-3117-3.