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Assessing the impact of artificial summer drainage on the benthic macroinvertebrates in a freshwater wetland in northeast Italy

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Abstract

Valle Mandriole is one of the two last remaining freshwater wetlands in the coastal area of Ravenna (NE Italy). In 2011 a management technique that involves the complete drainage of the southern portion of Valle Mandriole during summer has been undertaken. In the present study, the effects of this artificial drying on the benthic macroinvertebrate fauna were assessed using a beyond before-after-control-impact (beyond BACI) sampling design. Macroinvetebrates dwelling on macrophytes and in bare sediments were sampled in the impact location and in four control locations, two times before and two times after the drying period. Simultaneously, water samples were collected to monitor chemical properties potentially affecting the studied organisms. Biological and chemical data were analysed by multivariate statistical methods.

The statistical analysis did not detect any significant effect of the management action on the benthic macroinvertebrates or on the water chemical and physical properties. This contrasts with some previous results, suggesting that the effects of a management strategy based on draining completely dry and then reflooding a wetland area are site specific. However, it is necessary to consider that the present study does have some limitations, in particular the differences between impact and control locations and the timing of the sampling. The highest biodiversity was observed in one small and isolated control location; this highlights how maintaining, protecting, restoring and even creating small ponds may play an important role in biodiversity conservation.

Keywords: impact assessment; beyond BACI; benthic macroinvertebrates; water quality; water level management; managed wetlands

Declarations

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Conflicts of interest/Competing interests

The Authors have no conflict of interest or competing interest to declare.

Ethics approval

Not applicable

Consent to participate

All the participants voluntarily agreed to participate in study on which the present manuscript is based.

Consent for publication

All the Authors gave their consent to publish the present manuscript in Hydrobiologia.

Availability of data and material

Research data are provided as electronic supplementary material to the present manuscript.

Code availability

Not applicable.

Introduction

Wetlands are extremely diverse in terms of habitats and biological communities (Moss, 2000; Dudgeon et al., 2006) and are among the most productive environments all over the world (Ramsar Convention Secretariat, 2013). The value of wetlands, related to the number of ecosystem services they provide, has been repeatedly highlighted during the last forty years (Tiner, 1984; Postel & Carpenter, 1997; Mitsch & Gosselink, 2000; Barbier, 2011; Maltby & Acreman, 2011; Sievers et al., 2018).

Despite their importance, over the centuries the extent of wetlands on our planet has been considerably reduced as a result of human activities, *in primis*, land reclamation (Denny, 1994; Gordon et al., 2010, Antonellini et al., 2015).

In natural alluvial landscapes, the extension of wetlands is influenced by many factors, such as the hydrological cycle, the landscape morphology and the riverine regime, which can lead wetlands to evolve into terrestrial environments (Ward, 1997). Under these conditions, the natural burying processes can be counterbalanced by flooding of new areas. However, in inhabited regions river basin management prevents these dynamics. Thus, maintaining the water level and bathymetry of wetlands present in these areas often requires an active human intervention.

In the coastal areas of the Po Valley (northeast Italy), the alluvial wetlands generated by the Po River have been reduced considerably to make room for crops (Cencini, 1998; Buscaroli et al., 2011; Antonellini et al., 2015). After the reclamation process, excluding minor and hunting ponds, in the coastal area surrounding the city of Ravenna only two freshwater coastal wetlands have survived to nowadays: Valle Mandriole and Punte Alberete. Recognition of their important role as unique habitats that survived the reclamation process has triggered actions aimed at preventing progressive burying and onset of anoxic conditions, such as hydraulic intervention and vegetation clearing (Buscaroli et al., 2011).

A particularly drastic measure has been taken in Valle Mandriole, due to the recurrence of anoxia and deterioration of vegetation. Starting from year 2011, the southern portion of Valle Mandriole has been completely drained every July to allow for mineralization of sediment organic matter and to facilitate vegetation clearing, then flooded again at the end of September to maintain wintering habitat for waterbirds.

Information on the benthic macroinvertebrates is highly relevant to the whole wetland ecosystem, since they participate in the transformation of organic matter and they contribute to the cycle of energy and nutrients of the ecosystem. In particular they are a primary food source for organisms at the top of the food chain, i.e., fish, birds, mammals and reptiles (Covich et al., 1999, Cooper et al., 2009). Benthic macroinvertebrates are often used in biomonitoring to obtain information to implement management plans and improvement of environmental quality (Rosenberg & Resh, 1993). In particular, changes in the hydrological regime can significantly influence the composition of the benthic macroinvertebrate assemblages (Kaster & Jacobi, 1978; Furey et al., 2006; White et al., 2008; McEwen & Butler, 2010).

The aim of the present study was to assess if the dry period after complete drainage of the wetland impacted on the benthic macroinvertebrate assemblages, by comparison of the drained location with other reference locations in the same area, using a beyond before-after-control-impact (beyond BACI) sampling design (Underwood, 1992, 1994). Along with the benthic macroinvertebrate assemblages, the main water chemical and physical parameters have been evaluated pre and post the draining event.

Although it could be trivial to state that the benthos is affected in the period when the basin is completely dry, the point was to assess how the imposed dry period affected the benthic assemblages when the basin returned to be flooded. The drainage is supposed to improve the oxygenation conditions, of the wetland, historically affected by anoxia. This improvement is hypothesized to increase the diversity of the benthic assemblages and the abundance of taxa that are more sensitive to the oxygenation conditions. On the other hand, this intervention annihilates the benthos for a few months, and this could result in lower abundance and diversity for several months after the basin returns to be flooded (Lindegarth & Chapman, 2001; Bedford & Powell, 2005).

Although this study focuses on a specific wetland of limited extension, its scope is beyond the local interest, as it can contribute to understand the implications of a management practice of simple applicability and wide exportability.

Methods

Study area

The freshwater wetlands complex of Valle Mandriole (VM) and Punte Alberete (PA) is located in the Po Plain, northeast Italy, 4 km from the coast of the north Adriatic Sea, 3.5 km south of the brackish Comacchio lagoons, 10 km north of the city of Ravenna, covering an area of 420 hectares (Fig. 1). The two wetlands are separated by the River Lamone and are the remnants of a larger complex (about 8000 hectares), mostly reclaimed during the 1960s. The extension of the whole complex, as it appears today, was finally established in 1972 (Lazzari, 1994; Buscaroli et al., 2011).

Fig. 1

The entire study area lies in an anthropized landscape characterized by intensive farming and by the presence of a highway running along the eastern edge of both PA and VM. Moreover, the international port and the industrial area of Ravenna are located just a few kilometres to the South (Fig.1).

The area is part of the Po Delta Regional Park, established in 1988. In 1977, both wetlands were designated Ramsar sites, according to Ramsar Convention Secretariat (2013). In 1994 they were included in the Natura 2000 network as *site of community importance* (Council Directive 92/43/EEC) and as *special protected areas* (European Parliament and Council Directive 2009/147/EC).

Nevertheless, strong pressures on these habitats driven by alien organisms such as *Procambarus clarkii* (red swamp crayfish or Louisiana crawfish) and *Myocastor coypus* (coypu), saltwater intrusion and water turbidity, have compromised over time their ecological status (Antonellini et al., 2010; Studio Silva, 2012; Mollema et al., 2013).

Valle Mandriole, extending over 240 ha, lies in the north of the complex and is mostly made up of marshes with populations of *Phragmites australis*. One single sluice, located northeast, takes water in and out of the River Reno through an artificial channel.

Until 2011, to offset the significant lowering of the water level in the summer, VM was loaded to +0.80 m above mean sea level (AMSL) in September/October. This was implemented because Valle Mandriole was used as a reservoir for the water supply of Ravenna. However, this practice has led to a deterioration of emergent vegetation. Thus, starting from 2011 a management plan aimed at restoring the functionality of the biotope has been implemented. A complete drainage of the southern half of the wetland has been carried out every year during summer to foster mineralization of sediment organic matter. This intervention was accompanied in 2013 by the dredging of submerged channels to increase water circulation and guarantee refuge habitats for fishes.

Punte Alberete, extending over 186 ha, occupies the southern portion of the complex and consists mostly in alluvial forests alternating with low-lying perennially submerged marshes that are connected by ditches. The water level is controlled by two sluices. The first one, located in the southeast, is used to intake water from River Lamone through an artificial channel. The second one, in the northeast, is used to drain water to Adriatic Sea, through the Pialassa Baiona lagoon.

The water level of Punte Alberete is usually not actively managed. However, occasionally it is set by acting on the sluices to allow for vegetation clearing. The average water level calculated on three years (2010-2012) observed in the northeast sluice was +0.64 m AMSL in March and −0.18 m AMSL in August. A small interdunal wetland (0.1 ha), here referred to as SV, was also included in this study (see below for the sampling design). The pond is located within the San Vitale Pinewood, established on the sand dune system that extends immediately east of PA and VM (Fig.1). This habitat differs from the other sampling locations due to the vegetation, that includes *Thypa latifolia*, and to the higher water salinity.

Field and laboratory methods

A beyond before-after-control-impact (beyond BACI) sampling design was adopted here to assess the effect on the benthic macroinvertebrate assemblages. In particular, we studied the effect of the drainage of the southern part of VM, that occurred during summer 2013. In the beyond BACI design a single putatively impacted location is compared with multiple control locations All the locations are sampled at multiple times before and after the impacting event has occurred.

Five sampling locations (one impact, four controls) were identified in the study area. VM was divided into two portions (Fig. 1). The southern portion (VMS), extending approximately 120 ha, was identified as the impact location, since it was completely drained and kept dry from July to September. The control locations were the northern portion of Valle Mandriole (VMN), two marshes within Punte Alberete, one in the northern part (PAN), one in the southern part (PAS), and the pond within the San Vitale Pinewood (SV). Control locations maintained their own water level throughout the year, although minimal during summer. Within each location, two sites were identified, labelled as 1 and 2 (e.g., VMS1 and VMS2). All sites were located at the border between bare sediments and thickets of *Phragmites australis*, or *Typha latifolia* in the case of the SV pond where *P. australis* was not present. None of the sampled sites, neither in the impact location nor in the control locations, was subject to vegetation clearing during 2013. All ten sites were sampled twice before summer drainage (May and June), and twice after (October and November).

At each site, the macroinvertebrates were sampled separately from bare sediments and from emergent vegetation. For bare sediments, one sample for each site was taken using a PONAR grab with opening

155x175 mm (sampling surface: 0.027 m^2). On vegetation, a squared-frame kick net with opening 210x210 mm and a 0.5 mm mesh was used by scraping the immersed portion of the stems from the base upward for two minutes. Both types of samples were sieved on site with a 0.5 mm mesh conical net and then immediately preserved with 10% formaldehyde. In the laboratory, all sampled specimens were identified to family level and counted using a stereo microscope.

At each site, temperature (°C), electrical conductivity (µS/cm), pH, redox potential (mV) and dissolved oxygen concentration (mg/l) of the water were measured in the field, both on the surface and at the bottom, just above the sediment, using OX22 Aqualytic probe for dissolved oxygen and PCD 650 (EUTHECH Instruments) for the other parameters. Water alkalinity (meq/l) was also measured in the field by titration using alkalinity titration test (Merck Mcolortest^{TM}). Depth was determined using a graduated pole. In addition, a 2 l sample of surface water was taken and brought to the lab where dissolved sulphate (SO_4^{2-}-S, mg/l), ammonium (NH_4^+-N, mg/l), nitrite (NO_2^+-N, mg/l), nitrate (NO_3^+-N, mg/l) and phosphate (PO_4^{3-}-P, mg/l) were measured using a Hach DR/2010 spectrophotometer, and total and volatile suspended solids (g/l) were determined gravimetrically using GF/F Whatman® glass microfiber filters (Eaton & Franson, 2005).

Data analysis

Three different multivariate data sets were developed and then analysed separately: i) abundance of taxa collected from vegetation, expressed as number of individuals per minute of sampling; ii) abundance of taxa sampled from bare sediments, expressed as number of individuals per square meter; iii) physical and chemical properties of the water. In addition, number of taxa, Shannon index of diversity and Pielou's index of evenness were calculated for invertebrates from both vegetation and bare sediments.

To assess the impact of the drainage of VMN and the effects of the other factors, each data set was analysed by PERMANOVA (Anderson, 2001; Anderson et al., 2008). PERMANOVA is a non-parametric analysis analogue to the analysis of variance (ANOVA) that allows for multivariate (and univariate) data to be analysed and tested based on any resemblance measure. Analysis of complex designs, involving several orthogonal and nested factors is possible. All tests of hypothesis, including those regarding interactions between orthogonal factors, are performed using permutation techniques; 9999 permutations were used for all the analyses presented here.

The Bray-Curtis distance was used as the resemblance measure for taxa abundances, after square root transformation of the raw data. The Euclidean distance was used as the resemblance measure for chemical and physical properties, after standardization of the raw data. The Euclidean distance was used also for the diversity indices.

PERMANOVA was applied in accordance with the principles of Beyond BACI, following the indications of Anderson et al. (2008) for the analysis of asymmetrical designs.

The factors included in the analysis were:

- control vs. impact (I, fixed, two levels: control and impact);
- location (L, random, nested in the factor control vs. impact);
- site (S, random, nested in the factor location, two sites for each location);
- period (P, fixed, two levels: before and after the drainage of VMS);
- time (T, random, nested in the factor period, two times before and two times after the drainage of VMS).

The design is asymmetrical, since only one impact location was available (VMS), as opposed to four control locations (VMN, PAN, PAS and SV). For each combination of site and time, only one sample was collected for the benthos on vegetation, one for the benthos in bare sediments and one for the water. Replicated sites were sampled within each location, but there were no replicates within each site; as a consequence, the lowest-level interaction, i.e., site×time, had to be used as the residual source of variation and could not be tested.

According to the beyond BACI rationale, an effect of the drainage of VMS would cause the temporal pattern of benthic abundances or of chemical properties in the impacted location to differ from the temporal patterns in the control locations. Thus, an impact would be detected if the I×P interaction or the I×T interaction were significant.

To complement PERMANOVA, two additional multivariate methods, based on resemblance measures, were applied. Non-metric multidimensional scaling (MDS) was used to graphically represent the relationships among samples. Similarity percentage (SIMPER) was used to quantify the contribution of each taxon to the similarity within and to the dissimilarity between groups of samples (Clarke, and Warwick, 2001). Ordination of samples based on water chemistry variables was performed by discriminant function analysis, using the location as the grouping criterion (Legendre & Legendre, 2012).

The software StatSoft Statistica was used to carry out the discriminant function analysis. All the other calculations were performed using the software PRIMER 6 with the PERMANOVA+ add-on.

Results

Macroinvertebrates on vegetation

It was not possible to demonstrate any impact of the drainage of VMS on the abundances of benthic macroinvertebrate taxa sampled from vegetation, since the PERMANOVA analysis did not detect a significant I×P or I×T interaction (Table 1).

Table 1 Results of the PERMANOVA test for the benthic macroinvertebrates sampled from vegetation. *: significant (P < 0.05); **: highly significant (P < 0.01)

Source of variation	Degrees of freedom	P value			
		Abundances	N of taxa	H'	J'
I: impact vs. control	1	0.771	0.934	0.980	0.522
P: period, before vs. after	1	0.522	0.262	0.948	0.988
L: location (nested in I)	3	0.005**	0.003**	0.006**	0.001**
T: time (nested in P)	2	0.025*	0.880	0.034*	0.013**
I×P	1	0.636	0.516	0.272	0.254
S: site (nested in L)	5	<0.001**	0.192	0.236	0.834
I×T	2	0.181	0.140	0.411	0.567
L×P	3	0.433	0.008**	0.206	0.334
L×T	6	<0.001**	0.445	0.781	0.776
S×P	5	0.042*	0.960	0.584	0.580
Residual	10	–	–	–	–
Total	39	–	–	–	–

The L×T interaction was significant, denoting that the temporal patterns of benthic abundances differed among locations; however, these differences could not be ascribed to an effect of the drainage of VMS, since they were present also among control locations.

The factor location was significant also as a main effect, indicating that the abundances of benthic organisms differed among locations not only for their temporal pattern of variation but also for their mean values over the considered time span.

In fact, The MDS plot (Fig. 2) shows that samples from different locations are clearly separated on the ordination plane; more precisely the samples from the pond in San Vitale Pinewood (SV) are grouped on the higher part of the diagram, the samples from both locations of Punte Alberete (PA) are grouped on the lower right and the samples from both locations in Valle Mandriole (VM) are grouped on the lower left side of the plane.

Fig. 2

According to the SIMPER analysis, the taxa that most contributed to the distance between the samples from SV and those from PA are Physidae, Baetidae, Chironomidae, which were more abundant in SV samples, and Naididae, more abundant in PA samples (Tables S1, S2 in the Supplementary Data). The same taxa gave the highest contribution to the distance between SV and VM, all being more abundant in SV samples (Tables S1, S3 in the Supplementary Data). The pinewood pond was characterized by the highest abundance of all taxa, excluding Naididae and Cambaridae, and by the presence of several families of Diptera, Colepotera, Hetroptera and Odonata, absent in the other areas. Naididae is the taxon that by far gave the highest contribution to the distance between PA and VM, due to its high abundances in PA (Tables S1, S4 in the Supplementary Data). In general, VM was characterized by the highest abundances of Cambaridae and low abundances of other taxa. However, Baetidae were more abundant than in PA.

The horizontal sorting of the points in the MDS plot is mostly determined by the abundance of Naididae, which increases from left to right (Fig. S1a in the Supplementary Data). The vertical sorting is determined by the abundances of several taxa (notably Chironomidae, Baetidae, Physidae) that increase from bottom to top (Fig. S1b–d).

As for the temporal variation, in SV, Chironomidae, Baetidae and Physidae decreased in abundance over time, while Naididae increased in abundance. Coenagrionidae were sampled only in June (second time of the before period). In both locations of PA the abundance of Naididae increased from May to June and then decreased to the lowest values in the after period. Corixidae were relatively abundant in May and June and almost absent in October and November. One difference between PAS and PAN is the relatively high abundance of Gammaridae in the former in May and June. It is difficult to identify clear temporal patterns for VM due to the general low abundance of macrobenthic organisms. However, Cambaridae were generally more abundant in the after period, particularly in October. The highest abundance of Baetidae was recorded in June for VMS and in October for VMN.

The I×P and I×T interactions were not significant also for the diversity indices, while the main factor location was highly significant for all of them (Table 1). SV had the highest richness (14-20 taxa), VM constantly had a low richness (4-6 taxa) and PA had a decreasing richness over time from 9-11 taxa to 1-4 taxa (Fig. S2a in the Supplementary Data). The highest values of Shannon's index of diversity H' were recorded in SV, the lowest in PA, were they decreased over time alongside with the reduction of the number of taxa (Fig. S2b). Despite the low number of taxa, H' values in VM were higher than in PA, since all taxa had low abundance, and none were dominant. In fact, the highest values of Pielou's index of evenness J' were recorded in VM, the lowest in PA, where Naididae were highly dominant (Fig. S2c).

Macroinvertebrates in bare sediments

It was not possible to demonstrate any impact of the drainage of VMS on the abundances of benthic macroinvertebrate taxa sampled from bare sediments, since the PERMANOVA analysis did not detect a significant I×P or I×T interaction (Table 2).

Table 2 Results of the PERMANOVA test for the benthic macroinvertebrates sampled from bare sediments. *: significant (P < 0.05); **: highly significant (P < 0.01)

Source of variation	Degrees of freedom	P value			
		Abundances	N of taxa	H'	J'
I: impact vs. control	1	0.962	0.848	0.979	0.994
P: period, before vs. after	1	0.021*	0.993	0.736	0.384
L: location (nested in I)	3	0.028*	0.009**	0.003**	0.013*
T: time (nested in P)	2	0.893	0.735	0.488	0.393
I×P	1	0.513	0.949	0.984	0.980
S: site (nested in L)	5	0.041*	0.409	0.330	0.310
I×T	2	0.878	0.599	0.158	0.091
L×P	3	0.507	0.022*	<0.001**	0.079
L×T	6	0.016*	0.410	0.208	0.223
S×P	5	0.156	0.108	0.191	0.399
Residual	10	–	–	–	–
Total	39	–	–	–	–

Similarly to the vegetation samples, both the L×T interaction and the location main factor were significant, even if at higher P values, indicating that the mean values and the temporal patterns of the abundances of benthic taxa were different among locations, but that these differences could not be ascribed to an impact of the drainage.

The MDS plot (Fig. 3) shows that the points that represent the samples from VM and PA are clustered in the lower part of the diagram and largely overlap, indicating that the benthic assemblages in the two areas were rather similar in bare sediments, while on vegetation they were quite distinct. The samples from SV are scattered in the upper part of the plot, fairly spaced from each other and mostly clearly separated from those from VM and PA, indicating that the benthic assemblages of bare sediments of SV were not only different from those of the other areas but were also more heterogeneous.

Fig. 3

According to the SIMPER analysis, the samples from SV were characterized by the lowest mean abundance of Naididae and by the significant abundance of other taxa, in particular Chironomidae, Cerataopognidae and Chaoboridae (Tables S5-S7 in the Supplementary Data). On the other hand, the macrobenthos of bare sediments in VM and PA consisted almost exclusively of Naididae and the main difference between the two locations was that these organisms were on average more abundant in PA (Tables S5, S8 in the Supplementary Data).

The sorting of the points in the MDS plot is mostly determined by the abundance of Naididae, which increases moving to the lower right corner of the plot (Fig. S3a in the Supplementary Data) and by the abundance of Chironomidae, which increases moving in the opposite direction (Fig. S3b).

The abundance of Naididae decreased over time in all the locations, although to a different extent in each of them.

Again, the I×P and I×T interactions were not significant for the diversity indices, while the location factor was significant as main effect for all the indices and the L×P interaction was significant for H' (Table 2). The highest values were observed in SV, where they further increased in the after period (Fig. S4 in the Supplementary Data).

In summary, the temporal pattern of variation of the benthic assemblages at the impact location was within the range defined by the other wetlands in the area, both on vegetation and in bare sediments. The structure of the benthic assemblages and their temporal patterns were related to the wetland where each location was situated (SV, PA or VM) and not to the classification of the location as impact or control. Consequently, it was not possible to detect any statistically significant impact of the drainage of VMS on the macrobenthic fauna.

Water chemistry

Fig 4. shows the range of variation of chemical and physical properties at the five locations and at three sites on the main rivers and channels in the surrounding area. Dissolved oxygen below 1 mg/l was measured only once (site PAS1, October); PAN was the location with lower values on average. The dissolved oxygen recorded at VMS and VMN was always relatively high, with no indications of anoxia; however, the months from July to September in which anoxic events are more frequent were excluded from the sampling.

Fig. 4

It was not possible to demonstrate any impact of the drainage of VMS on the water chemistry variables, since the PERMANOVA analysis did not detect a significant I×P or I×T interaction (Table 3).

Table 3 Results of the PERMANOVA test for the chemical properties of the water. *: significant ($P < 0.05$); **: highly significant ($P < 0.01$)

Source of variation	Degrees of freedom	P value
I: impact vs. control	1	0.957
P: period, before vs. after	1	0.369
L: location (nested in I)	3	0.007**
T: time (nested in P)	2	0.057
I×P	1	0.643
S: site (nested in L)	5	0.010*
I×T	2	0.690
L×P	3	0.079
L×T	6	<0.001**
S×P	5	0.289
Residual	10	−
Total	39	−

The L×T interaction and the location main factor were significant, as for the macrobenthos on vegetation and in bare sediments, indicating that the mean values and the temporal patterns of the considered chemical variables were different among locations, but that these differences could not be ascribed to an impact of the draining.

Fig. 5 shows the ordination plot based on the discriminant function analysis, using the location as the grouping criterion. The analysis identified electrical conductivity, sulphates, volatile suspended solids and phosphates as the variables giving a significant contribution to the separation between locations. The first discriminant axis has a strong negative correlation with electrical conductivity and clearly separates SV (average conductivity around 4 mS/cm) from the other locations, characterized by conductivity below 1 mS/cm. The second discriminant axis is positively correlated with volatile suspended solids, on average higher in the PA locations, and is negatively correlated with sulphates, higher in the VM locations. The samples from the two locations within Valle Mandriole, the impact location (VMS) and the control location (VMN), largely overlap. The two locations from PA, that are actually two separate ponds, are close to each other on the ordination plane, but still clearly distinct.

Fig. 5

In summary, similarly to what observed for the benthic assemblages, the values of the water chemistry and their temporal patterns were related to the area where the location was situated and not to the classification of the location as impact or control.

Discussion

The preservation and protection of wetlands is a goal that has been repeatedly emphasized in recent years (e.g., Mitsch & Gosselink, 2000; Sievers et al., 2018). In many cases this goal is pursued with specific management actions, of which the summer drainage of the southern part of Valle Mandriole is an example. Assessing the consequences of these actions is fundamental to identify the most effective practices.

The present study did not detect any adverse effect of the drainage on the benthic macroinvertebrates, suggesting that this practice is acceptable, at least as regards the investigated assemblages. On the other hand, the results also suggested that the drainage did not produce any benefit on the macrobenthic fauna, at least in the short term.

To adequately evaluate this result, it is necessary to take into consideration the characteristics of the studied wetlands and of their benthic assemblages. In addition, it is important to be aware of the limitations of the present study and, in general, of the problems involved in assessing the environmental impact of a specific event.

The studied wetlands and their benthic macroinvertebrate assemblages

In the present study benthic macroinvertebrates were identified at family level. We acknowledge that a more detailed taxonomical resolution would have been preferable and that, working at family level, relevant information is lost, in particular regarding the assessment of biodiversity. This was considered an acceptable compromise based on Bowman & Bailey (1997) and Mueller et al. (2013). These studies analysed several independent benthic macroinvertebrate datasets collected in freshwater ecosystems, both lotic and lentic. Both studies acknowledge the importance of identification to species or genus level in some circumstances. However, both studies concluded that multivariate characterizations of community composition do not seem to be sensitive to taxonomic resolution, at least to family level, if quantitative data (abundances) are considered. In fact, in many ecological studies on the benthic macroinvertebrates of shallow lentic freshwater habitats organisms are identified at the family level, including those cited in the next section.

Even if based only on identification at family level, the present study evidenced that the macroinvertebrate assemblages dwelling on vegetation and in bare sediments of Valle Mandriole (VM) and Punte Alberete (PA) had low abundance and diversity in comparison to the pond in the San Vitale Pinewood (SV). In particular, the macrobenthic fauna of bare sediments was almost entirely composed by Naididae.

A comparison with the results of Zanni (1998) shows that the taxa richness radically decreased over the last decades. In particular, several families of Coleoptera (Dytiscidae, Haliplidae, Dryopidae), Odonata (Lestidae, Libellulidae and Aeshnidae), and Heteroptera (Pleidae, Mesovelidae) once present in both VM and PA were not sampled during the present study.

The low water quality in the two wetlands and in the rivers that supply them water could have played a role in the decreased diversity of the benthic fauna. In this regard, both the Lamone River, which supplies Punte Alberete, and the Reno River, which supplies Valle Mandriole, failed to achieve good chemical status, under the European Water Framework directive in the period 2010–2012 (ARPAE, 2015).

In addition, the annual mean concentrations of ammoniacal nitrogen in the Reno River in the period 2010–2013 were in the range 0.43–0.64 mg/l, placing the river at the fifth level (worst quality class) for this indicator, under the Italian regulations (Fig. 4). Even higher concentrations of ammoniacal nitrogen were recorded in the Destra Reno channel. This artificial channel does not supply water to VM or PA; however, since it drains the farmland area surrounding VM and PA, its water properties give some indications on the surface runoff entering the two wetlands. Indeed, high concentrations of ammoniacal nitrogen (up to 1.1 mg/l) were measured in several water samples collected from PA.

A feature of PA is the recurrence of suspended mucilage, often observed in the water also during the present study, consistently with the high values of volatile suspended solids, generating strong water turbidity (Fig. 4).

A second possible reason for the impoverishment of the benthic assemblages is the presence the invasive alien crayfish *Procambarus clarkii*. This species was not recorded by Zanni (1998); on the contrary, it was frequently sampled during the present study. In Europe, the first introductions of *P. clarkii* from North America occurred in Spain in 1973 for aquaculture. The species was introduced in Italy in 1989, for the same purpose. After escaping into freshwater bodies, it has since steadily spread across Europe due to its ecological plasticity, high fecundity, rapid life cycle and dispersal capacities. *P.clarkii* has been documented to heavily affect abundance and diversity of benthic invertebrate assemblages both by direct predation and by modifying habitat composition, through consumption of living macrophytes and litter (Souty-Grosset et al., 2016).

Thirty-one families were identified in the macrobenthic fauna of the pond in the San Vitale Pinewood (SV), including several recorded by Zanni (1998) in PA and VM and not sampled in these wetlands during the present study (Culicidae, Tipulidae, Aeshnidae, Lestidae, Libellulidae, Mesovelidae, Dytiscidae, Haliplidae). Among the features of SV that might concur in determining the higher taxonomic diversity is the origin of its water, which comes mainly from the local underneath aquifer. The San Vitale Pinewood is established on a paleodune system constituted by 20 m thick coarse sand deposits (Amorosi et al., 1999 and Greggio et al., 2018) where the coastal aquifer is phreatic and rainwater infiltrate and freshen the underneath saline groundwater (Antonellini et al., 2008). If the budget between precipitation and evapotranspiration is positive, freshwater lenses settle down on top of saline groundwater; where the shallow water table meets low topographic surface, interdunal wetlands appear (Cozzolino et al., 2017). So, SV is not affected by the sources of pollution occurring to PA and VM where water is supplied by the final stretch of Lamone and Reno rivers running through a heavily anthropized lowland area. Accordingly, most of the chemical properties of the water of SV such as nutrients and suspended solids, are indicative of a good state (Fig. 4). On the other hand, the electrical conductivity of the water is rather high (about 4 mS/cm) compared to VM and PA (less than 1 mS/cm), even if it can be considered relatively fresh for Ravenna coastal aquifer (Mollema et al., 2013; Greggio et al., 2020). However, this does not seem to hinder the development of a well differentiated, typically freshwater, biological community. Based on the comparison with SV, it can be inferred that the richness of the benthic fauna in VM and PA was clearly below the potential of the area.

The effect of drainage on benthic macroinvertebrates

The impoverished fauna of VM could be a reason why no effects of the drainage were detected in the present study: there was little potential for change due to the presence of few taxa and low abundances. The benthic assemblages of both VM and PA testify the low habitat quality of the whole wetland system. The draining of VMS was apparently inadequate to increase abundance and diversity of the benthic macroinvertebrate assemblages, possibly because the starting point was so disadvantaged. In addition, even if the environmental conditions had improved in VMS, there would have been little possibility to develop a richer fauna by colonization from nearby areas. On the other hand, it could be argued that it was impossible to further worsen an already compromised situation.

However, the absence of lasting effects on the benthic assemblages in cases of drying events has been documented also in previous studies. Vander Vorste et al. (2016) applied a multisite before-after-control-impact (BACI) design to quantify the effects of drying events of different durations on benthic invertebrates in gravel-bed, braided rivers in southeast France and concluded that taxonomic composition, functional diversity, trait richness, and trait composition showed no effects of drying events.

On the other hand, effects of dry periods on the macrobenthic fauna, lasting long after the water level had recovered, were documented, both in cases of management actions and in cases of drought. Lindegarth & Chapman (2001) tested the effect on benthic invertebrates of controlled drainage and subsequent refilling in a small managed marsh in Australia. The goal of the water level manipulation was to improve the habitat for water birds and an increase in abundances of benthic invertebrates was expected. Contrary to the expectations, a decrease in abundance and taxa richness of benthic invertebrates, chironomids in particular, was evidenced.

Bedford & Powell (2005) monitored the invertebrate fauna associated with the litter of *Phragmites australi*s in a managed reedbed in northwest England by bi-monthly sampling over 6 years. Management involved an annual cycle of summer draw-down and winter reflooding. Based on comparisons with data from previous studies on permanently flooded reedbeds, these Authors concluded that the manipulation of water levels caused low invertebrate abundances and prevented the development of diverse communities.

Bertoncin et al. (2019) assessed the effect of a prolonged drought on the benthic invertebrates of a small (60×15 m) pond located within an island of the upper Paraná River, Brazil. The benthic assemblages became more homogeneous after the drought, in the sense that the differences between the three sites within the pond decreased. However, both diversity and abundance increased within each site, in contrast to the results of Lindegarth & Chapman (2001).

Unambiguously measuring the impact of a specific event in a natural ecosystem is challenging (see next section) and the above cited studies, including ours, have limitations. However, taken together, they indicate that the effects of a management strategy based on completely drying and then reflooding a wetland area are site specific. As for benthic invertebrates both an increase and a decrease in abundance and diversity is possible, as well as it is possible that no substantial changes occur. In our view, this depends on the balance between the actual improvement of the oxygenation conditions and the disturbance caused by the temporary cancellation of the aquatic habitat. The relative importance of these two drivers is influenced by climate, morphology, hydrology and water quality, along with the ecological community present at the site and the availability of refuges or nearby habitats from which recolonization can occur.

Impact assessment

Evaluating the outcomes of management actions is an essential but often neglected aspect of environmental management (Walsh et al., 2012). The effect of a specific action can only be evaluated within an experimental framework that should be planned before the action takes place (Lindegarth & Chapman, 2001). Even when this condition is met, achieving a quantitative and unambiguous assessment is challenging.

The major problem in assessing the impact of a specific event in a natural ecosystem is that there is usually only one potentially affected location (Stewart-Oaten et al., 1986; Underwood, 1992). This lack of replication makes it difficult to separate the anthropogenic effects from natural variability in space and time.

In an attempt to overcome this problem, the before-after-control-impact (BACI) sampling design was proposed. The BACI approach dictates that a single impact location, putatively affected by the event of interest, is compared with a single control location, surely unaffected by the same event. Both the impact and the control locations are sampled at multiple times both before and after the event has occurred (Bernstein & Zalinsky, 1983; Stewart-Oaten et al., 1986).

The shortcomings of this approach were highlighted by Underwood (1992, 1994), that proposed the beyond before-after-control-impact (beyond BACI) sampling design, where the single impact location is compared with multiple control locations, the same design we adopted in the present study. While the approach was originally devised to deal with human interventions expected to cause adverse effects on the ecosystems, it is equally suited to assess the effectiveness of actions aimed at some improvement.

Some authors (notably Stewart-Oaten & Bence, 2001; Stewart-Oaten, 2008; Paul, 2011), strongly criticized the Beyond BACI and other “design based” approaches, which are founded on sampling design and ANOVA or related statistical methods. In open contrast with Underwood (1992, 1994), they maintain that multiple control locations are not indispensable to properly measure the ecological effects of an event.

These authors advocate “model based” approaches, i.e., the use of predictive models, parametrized using data collected from the putatively impacted location before the event of interest occurs. The ecological impact should then be measured as the difference between the predictions of the model and the actual observations made after the event has occurred.

One of the fundamental tenets stated by Underwood (1992, 1996) is that impact assessment studies should be treated as experiments. This is the most criticized aspect of the approach. As Stewart-Oaten & Bence (2001) and Stewart-Oaten (2008) pointed out, statistical inferences in experiment are based on the assumption that the experimental units are randomly chosen from the same large population or, at least, randomly assigned to the treatments. Conversely, in an impact assessment study, the impact location is not chosen at random, either from a population or from the sites used in the study.

We acknowledge that the theoretical framework of the beyond BACI approach may have weaknesses, if considered from a formal statistical standpoint. However, we adopted this approach in the present study, because it is our opinion that its fundamental rationale is sound: an ecological effect is detected if the temporal pattern of variation observed at the impact location is outside the range defined by a set of control locations, which are unaffected by the event of interest but otherwise comparable with the impact location. The use of more than one control location takes into account that different unaffected locations may exhibit different temporal patterns. If only one control location is sampled, differences that could exist even if the impact location were actually unaffected could be interpreted as an ecological effect of the event of interest.

On the other hand, a model based impact assessment requires a validated model that can predict how the value of the response variables would change over time at the impact location if this were unaffected by the planned intervention. Complete confidence in the model is required since any deviation from its predictions would be interpreted as an actual impact. A long data series, collected before the planned disturbance, is essential to develop and validate the model. We did not follow this approach as it appeared unrealistic to develop a sufficiently reliable model for the abundances of benthic invertebrates and the values of the chemical properties of Valle Mandriole.

While we think that, under the circumstances, there were no better alternatives to evaluate the effect of the summer drainage of VM on the benthic invertebrate fauna, the specific application of the Beyond BACI approach carried out in the present study does have some important limitations.

The real situation hardly fitted to the reference model of a population of locations from which the impact locations and the control locations were independently and randomly selected. While all the sampled locations are freshwater wetlands, the benthic assemblages and the chemical properties of Valle Mandriole (impact location and one control location), of Punte Alberete, (two control locations) and of the pinewood pond were quite distinct.

According to Underwood (1994) in the application of the beyond BACI approach, there is no need to attempt to choose places with identical characteristics or abundances of the investigated populations. The set of locations chosen to serve as controls must simply represent the range of habitats like the one that might be affected (the Impact location). Obviously, the control locations must be a representative sample of places of the same general habitat as the impact location.

However, what can be considered the same general habitat, is rather subjective. Our strategy was to sample the highest possible number of locations and to represent the whole range of freshwater wetlands in the area. Since the control locations define the norm against which the temporal pattern observed in the impact location is evaluated, our idea was that the norm should have some degree of generality. In our opinion this is consistent with the rationale of the Beyond BACI approach.

The opposite strategy would have been to favour the homogeneity between the impact and the control locations. It could be argued that the differences between SV and the other locations are too large to represent the same general habitat. Admittedly, the benthic assemblages of SV are clearly distinct from those of VM and PA. However, regarding the temporal patterns (how the structure of the benthic assemblages changes over time) the difference between SV and the other locations do not seem to be larger than the differences among the other locations, at least for the fauna sampled from vegetation (Fig 2, Fig. S2 in the Supplementary Data). In any case, omitting the SV from the statistical analyses does not cause major changes in the results and the interaction terms relevant to the detection of the impact are still not significant (data not shown).

In fact, while PA and VM both had an impoverished benthic fauna, their benthic assemblages and the properties of their water were still clearly distinct. Adopting homogeneity with the impact location as the sole criterion to select the control locations, would imply that VMN is the only legitimate control location for VMS, and would make it impossible to apply a beyond BACI approach.

Arguably, in the present study, the range of temporal patterns observed at the control locations, which defined the norm, was so wide that possibly prevented the detection of any deviation of the impact location from the norm itself, except for extreme deviations.

Indeed, the most important shortcoming of the present application of the Beyond BACI approach is probably that all the samples were collected during one single year (2013) and "before" and "after" are referred to a single drainage of VMN. Since the drainage was performed on summer every year, starting from 2011, sampling over a longer time span, including a period before the first occurrence of the summer drainage would have been more relevant to the problem considered. Unfortunately, the local authority that planned and funded the intervention, apparently did not deem important to plan a study to properly assess its effectiveness.

Conclusions

The present study, even taking account of its limitations, did not detect any statistically significant effect on the benthic macroinvertebrates, suggesting that this practice is acceptable, at least in regard to the investigated assemblages. On the other hand, the results also suggest that the drainage does not produce any benefit on the macrozoobenthic fauna, at least in the short term. The effects of a management strategy based on draining completely dry and then reflooding a wetland area appear to be site specific.

Our study, by comparison to previous data, evidenced a severe impoverishment of the benthic fauna of Valle Mandriole and Punte Alberete, as can be inferred by comparison to the San Vitale pinewood pond which, despite being of lesser extension, sustains a much more diverse fauna. This also highlights how small water bodies can preserve the diversity of aquatic species of an area, even when larger water bodies are degraded, and that they are potentially important pools of species that could recolonize larger habitats, once the environmental quality of the latter is restored. Maintaining, protecting, restoring and even creating small ponds may play an important role in nature conservation.

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Captions for figures

Fig.1 Study area and sampling sites

Fig. 2 Non-metric multidimensional scaling (MDS) plot of 10 sites sampled four times, two times before (May, June) and two after (October, November) the drainage of the southern part of Valle Mandriole (VMS). The MDS is based on square root transformed abundances of families of benthic macroinvertebrates sampled from vegetation and Bray-Curtis distance

Fig. 3 Non-metric multidimensional scaling (MDS) plot of 10 sites sampled in four times, two times before (May, June) and two after (October, November) the drainage of the southern part of Valle Mandriole (VMS). The MDS is based on square root transformed abundances of families of benthic macroinvertebrates sampled from bare sediments and Bray-Curtis distance

Fig. 4 Range of variation of water column properties in the five wetland locations and three related riverine locations in year 2013. The wetland data are from the present study, the riverine data were collected by the regional environmental agency (available at: https://www.arpae.it/dettaglio_documento.asp?id=6312&idlivello=2020). VMS: Valle Mandriole South (impact location); VMN: VMN: Valle Mandriole North; PAN, PAS: Punte Alberete north and south; SV: San Vitale. Riverine locations: Lamone, Destra Reno, Reno. Boxes: 2nd and 3rd quartile; the whiskers represent the whole range without outliers. Outliers are defined as value outside 1.5 times the interquartile range above the 3rd quartile and below the 2nd quartile

Fig. 5 Discriminant function analysis ordination plot using the location as the grouping criterion and the water chemistry variables as predictors

Figure

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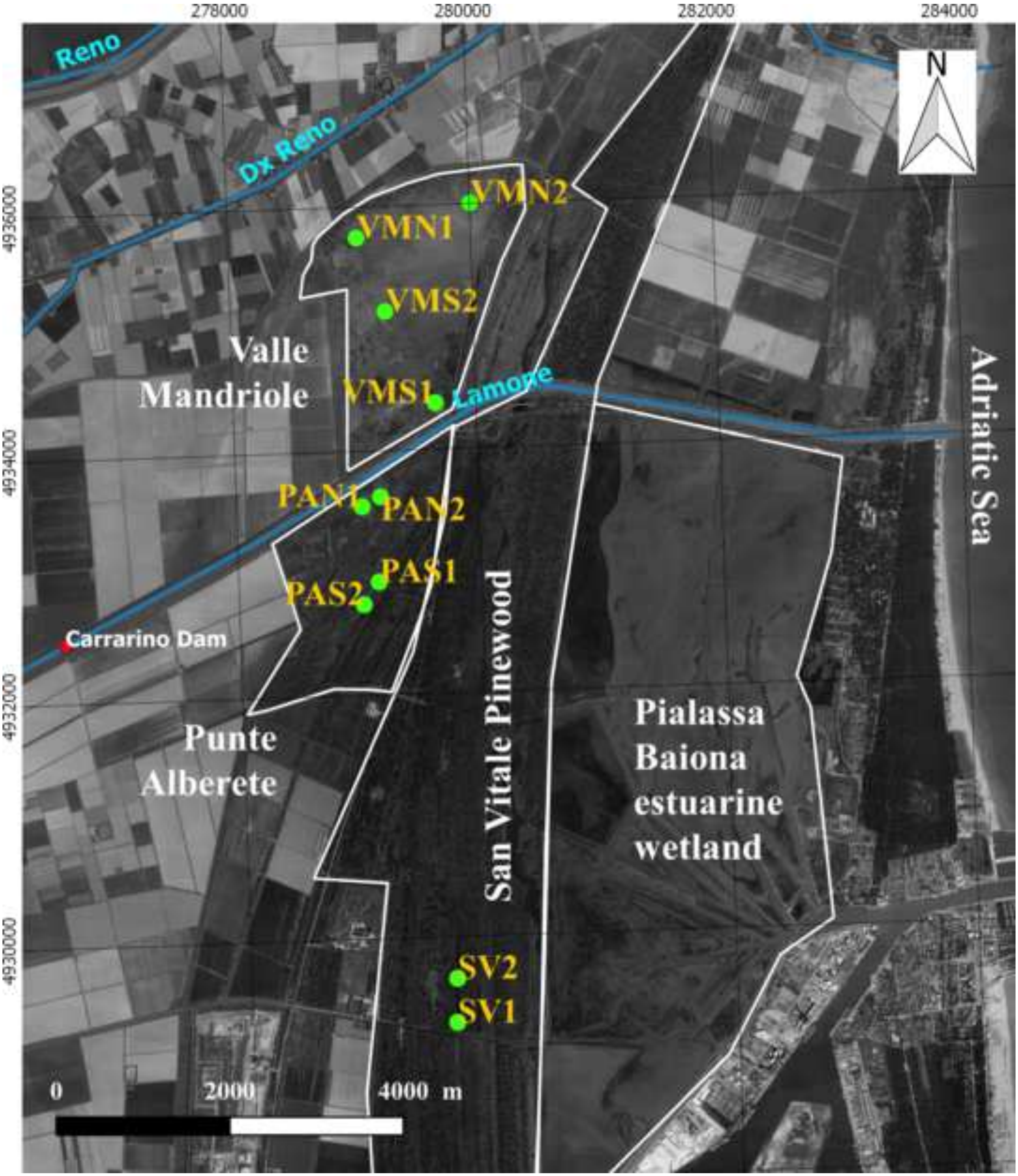
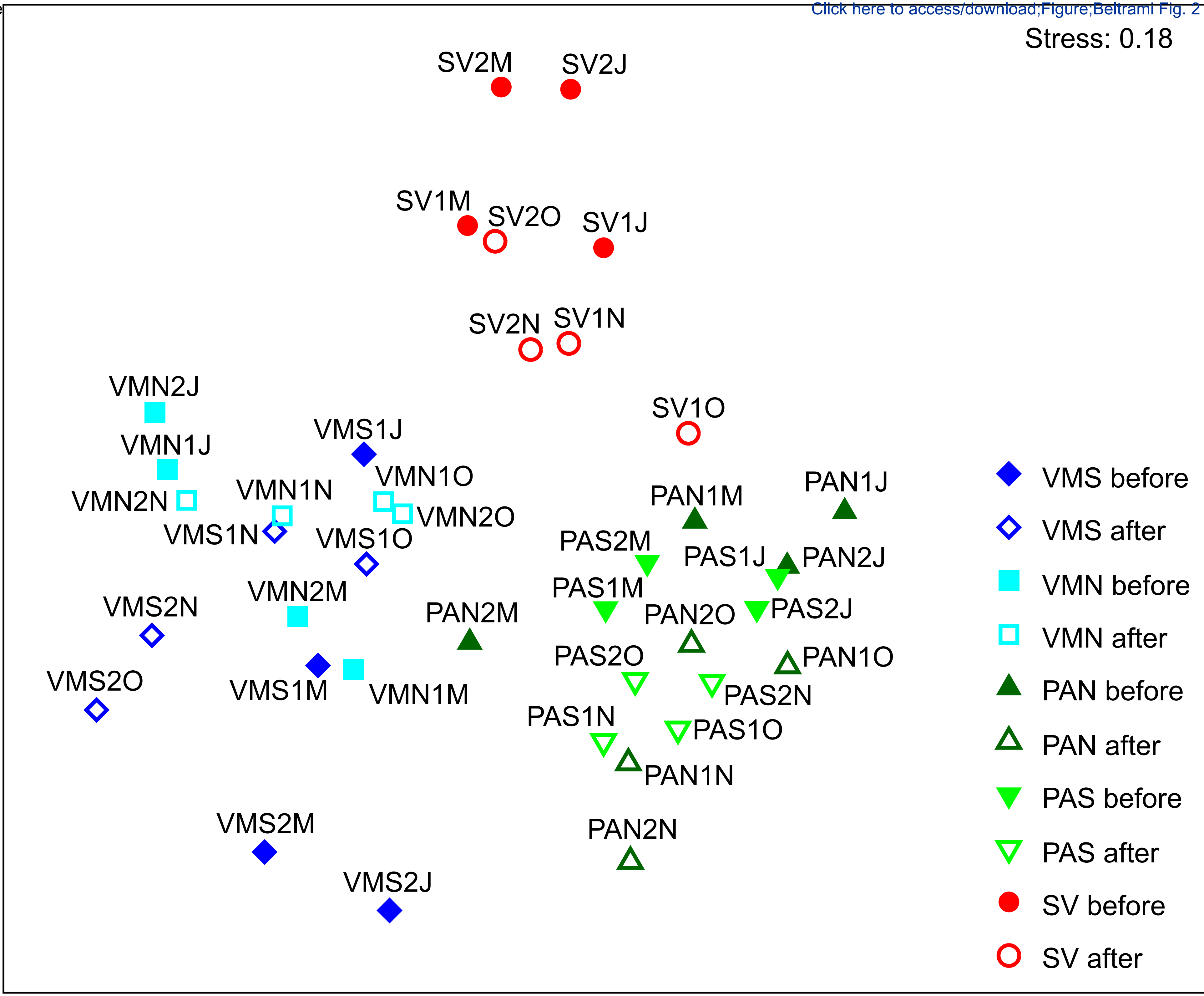
Fig. 1 .tif



Figure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VMN1N
VMN2O
VMS1N
VMS1O
PAN1M
PAN1J
PAS2M
PAS1J
PAN2J
PAS1M
PAS2J
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VMS2N
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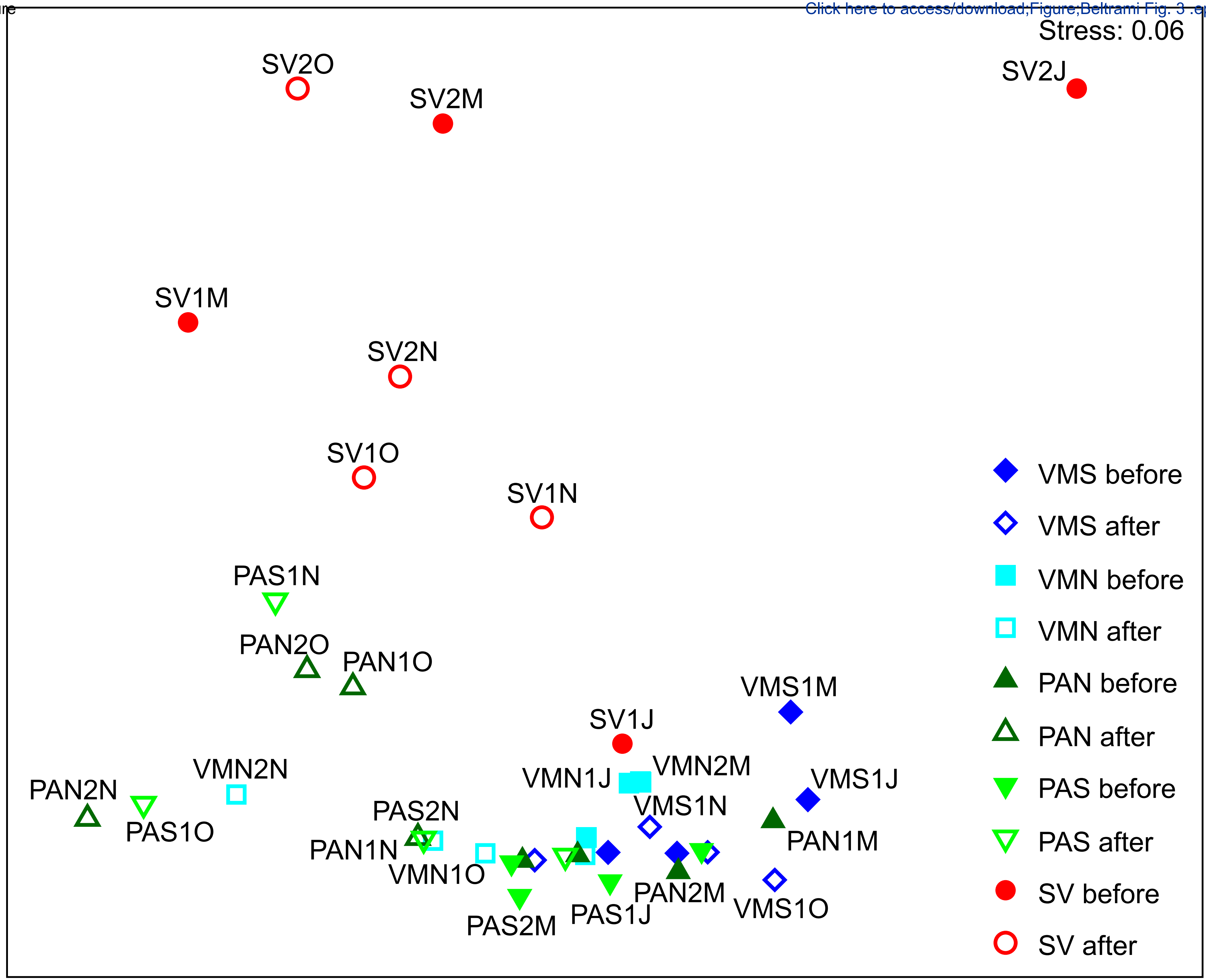
VMS2O
VMS1M
VMN1M
PAS2N
PAS1N
PAS1O
PAN1N
PAN2N
VMS2M
VMS2J
VMS before
VMS after
VMN before
VMN after
PAN before
PAN after
PAS before
PAS after
SV before
SV after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VMS1N
PAS2N
PAN1M
PAN1N
VMN1O
PAN2M
PAS1J
VMS1O
PAS2M
VMS before
VMS after
VMN before
VMN after
PAN before
PAN after
PAS before
PAS after
SV before
SV after

Figure

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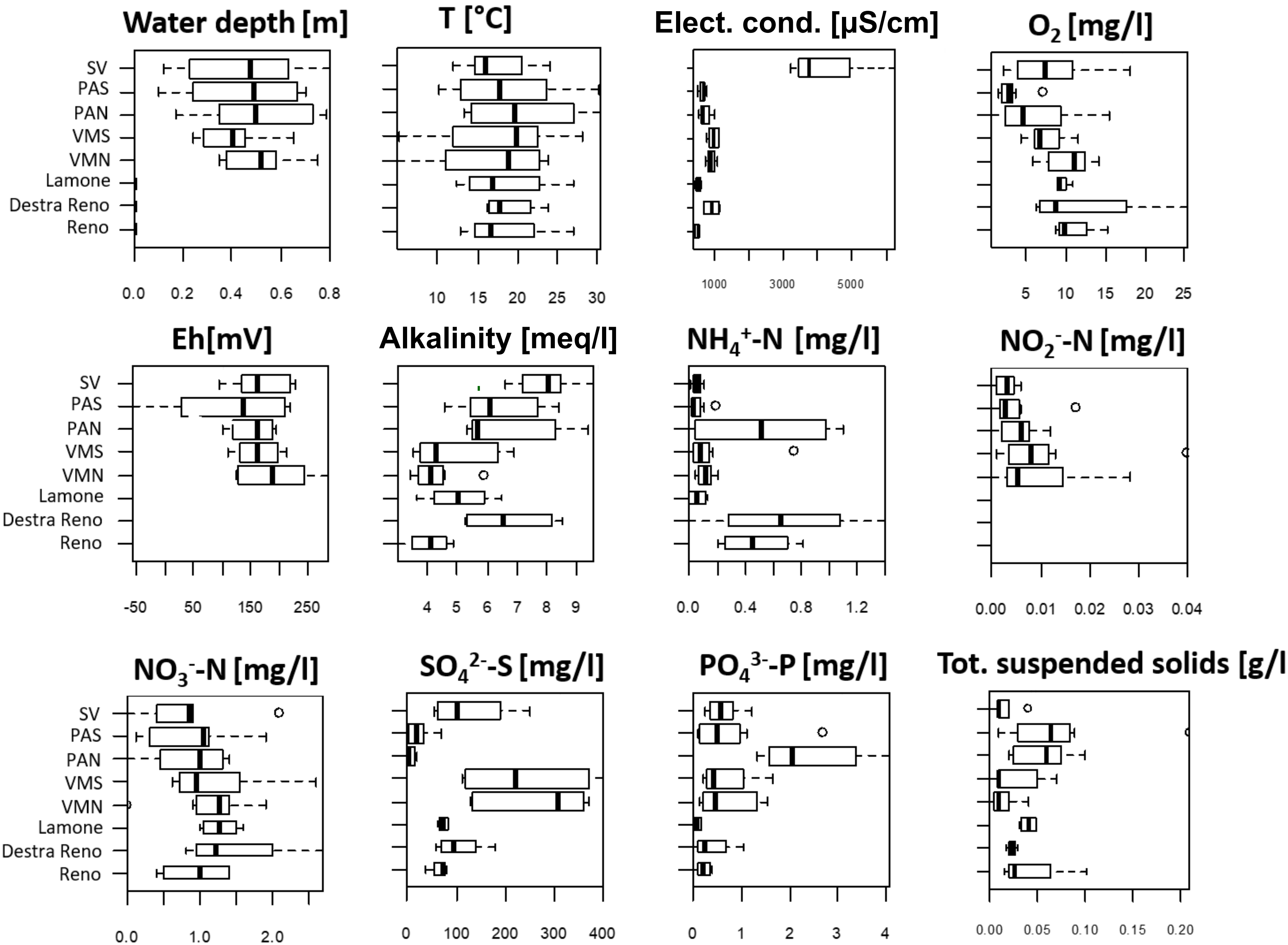

Figure

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