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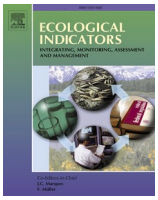

Beta-diversity and stressor specific index reveal patterns of macroinvertebrate community response to sediment flushing

Alberto Doretto [a,b,*,1], Elena Piano [b,c], Stefano Fenoglio [b,c], Francesca Bona [b,c], Giuseppe Crosa [d], Paolo Espa [e], Silvia Quadroni [d,*,2]

[a] Department of Sciences and Technological Innovation, University of Piemonte Orientale, Viale Teresa Michel 11, 15121 Alessandria, Italy
[b] ALPSTREAM – Alpine Stream Research Center, 12030 Ostana, Italy
[c] Department of Life Sciences and Systems Biology, University of Turin, Via Accademia Albertina 13, 10123 Turin, Italy
[d] Department of Theoretical and Applied Sciences, Via JH Dunant 3, 21100 Varese, Italy
[e] Department of Science and High Technology, Via GB Vico 46, 21100 Varese, Italy

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ABSTRACT

Anthropogenic increase of fine sediment loading is one of the main pressures for rivers worldwide. Particularly, Alpine streams are increasingly facing this issue due to sediment flushing operations from hydropower reservoirs, aimed at recovering storage for preserving electricity generation. Although Controlled Sediment Flushing Operations (CSFOs) are becoming increasingly frequent, ecological indicators to adequately assess and monitor their impact on the stream ecosystem have been poorly developed. In this work, we aimed to perform a screening of currently available biomonitoring tools to evaluate the CSFO effects on the riverine biota and adequately assess its recovery, starting from the recognition of the main ecological mechanisms triggered by the mentioned activities on benthic macroinvertebrate communities. We used two independent datasets concerning two reservoirs in the central Italian Alps to investigate the temporal effects of CSFOs repeated for four consecutive years (case-study I), and the impact of a single CSFO at a seasonal scale through a before/after-control/impact approach (case-study II). Initially, we quantified the CSFO impact on the richness and beta-diversity of macroinvertebrate communities by combining multivariate and univariate statistical techniques. Then, we compared the performance of the Siltation Index for LoTic EcoSystems (SILTES), recently developed for detecting siltation impact in Alpine streams, with that of the generic index currently adopted to assess the ecological status (*sensu* Water Framework Directive) of the Italian rivers, and of another sediment-specific index, but developed for a different bio-geographical area. The analysis of the two case-studies demonstrated that the nestedness (i.e. taxa loss) is the primary source of biological impairment caused by CSFOs. Moreover, we found that SILTES was more effective than the other indices because of its strong correlation with the nestedness, and since it properly discriminated impaired and pristine conditions, at both multi-annual and seasonal scale. In the first case-study, a threshold in the temporal trend of this index was detected, indicating a recovery within three months. In the second one, SILTES showed a recovery to pre-event seasonal values after nine months from the CSFO, due to larger and more persistent sediment deposition. This study demonstrates that SILTES could be adopted as a benchmark to improve the management of CSFOs from an ecological viewpoint. Our findings can be extended to the management of other sediment-related activities affecting mountainous streams worldwide, and, more generally, the adopted approach can be replicated for developing new ecological tools to manage other disturbances to river environments.

* Corresponding authors at: Department of Sciences and Technological Innovation, University of Piemonte Orientale, Viale Teresa Michel 11, 15121 Alessandria, Italy (A. Doretto).
E-mail addresses: alberto.doretto@uniupo.it (A. Doretto), silvia.quadroni@uninsubria.it (S. Quadroni).
[1] ORCID ID: 0000-0002-4105-473X.
[2] ORCID ID: 0000-0003-1295-2108.

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1. Introduction

Increased fine sediment loading by anthropogenic activity is recognized as a global threat to lotic ecosystems (Turley et al., 2014). In the European Alps, as well as in several mountain areas worldwide, the natural dynamics of sediment has been deeply altered by several human manipulations of the landscape, including channelization, deforestation, mining (Doretto et al., 2016; Wohl, 2006), and, probably to a major extent, by massive hydropower development (Espa et al., 2019; Parish et al., 2019). In this context, sediment deposition into hydropower reservoirs generally determines degradation in sediment-starved stream sections below dams (Comiti, 2012). Moreover, the need to recover reservoirs capacity to preserve hydropower production typically results in different sediment management strategies being undertaken, including sediment flushing from reservoirs (Morris, 2020). Although sediment flushing can partly restore the natural sediment flux, the sediment pulse during the operation and possible persistent sediment deposition after the operation can severely affect the aquatic ecosystem, both in the short and long term (Espa et al., 2019). The current tendency to mitigate the downstream impact of sediment flushing comprises thresholds of suspended sediment concentration (SSC) and duration of the flushing operation, i.e. controlled sediment flushing operations, CSFOs (Espa et al., 2019; Tritthart et al., 2019). These thresholds are usually estimated to limit fish mortality, according to the dose–response model by Newcombe and Jensen (1996) that, in some cases, proved to underestimate the measured impact (Espa et al., 2019). Only rarely these operations are integrated in a comprehensive sediment management strategy (Gabbud and Lane, 2016; Wohl et al., 2015) aimed to sustain long-term utilization of reservoir storage and improving at the same time the environmental quality of the dammed rivers. Moreover, specific and reliable biomonitoring indices or biological metrics to properly assessing the ecological effects of this activity are still scarce (Espa et al., 2019).

In the European Union, several metrics were developed by the member states to evaluate the ecological status (i.e. overall ecosystem health) of lotic ecosystems in the context of the Water Framework Directive (WFD, 2000/60/EC). An example is the Standardization of River Classifications_Intercalibration Common Metric index (STAR_ICMi – Buffagni and Erba, 2007), i.e. the index officially used to determine the ecological status of Italian watercourses through benthic macroinvertebrates. However, concerns arise when indices of this kind are adopted to assess the effects of specific stressors like increased sediment loading, as it currently occurs within the monitoring programs aimed to assessing the impact of CSFOs (Doretto et al., 2019; Espa et al., 2015). In contrast, during the last decade, some indices, mostly adopting benthic macroinvertebrates as bioindicators (Extence et al., 2013; Hubler et al., 2016; Murphy et al., 2015; Relyea et al., 2012), have been specifically developed to detect the impact of increased fine sediment loading. However, these indices are usually bio-geographically limited. For instance, the Proportion of Sediment-sensitive Invertebrates (PSI) index, scoring each benthic invertebrate taxon according to its sensitivity or tolerance to fine sediment, was developed in the United Kingdom and, as a consequence, referred only to that pool of taxa (Extence et al., 2013). For the Alpine context, only the multi-metric index recently proposed by Doretto et al. (2018a) has been developed to assess the impact of siltation (hereafter: SILTES – Siltation Index for LoTic EcoSystems), but its application is so far restricted to few case studies (i.e. a pulse sediment flushing event – Doretto et al., 2019; an extraordinary sedimentation event characterized by high sediment input and long-term deposition – Salmaso et al., 2020). In accordance to studies evidencing changes in taxonomy-based and trait-based metrics after sediment disturbance (e.g. Bona et al., 2016; Buendia et al., 2013; Descloux et al., 2014; Mathers and Wood, 2016), the mentioned index includes both compositional and functional metrics. Specifically, it accounts for taxon richness, richness in EPT (Ephemeroptera, Plecoptera and Trichoptera) taxa, and abundance of rheophilous taxa preferring coarse substrata, typical of oligotrophic, alpine habitats (i.e. ecological group A *sensu* Usseglio-Polatera et al., 2000).

Bonada et al. (2006) pointed out that an ideal biomonitoring index should be based on sound theoretical concepts in ecology and discriminate specific human impacts, along with other criteria. The sediment pulse during the CSFO and its associated riverbed siltation alter the physical environment, potentially influencing how biodiversity is organized in space and time. In this perspective, beta-diversity, i.e. the variation in taxonomic composition between sampling units, can represent an important indicator to support the assessment of the ecological integrity of rivers (Ward and Tockner, 2001) and possible shifts from undisturbed conditions due to CSFO disturbance. Although being mostly neglected in the field of biomonitoring (Hawkins et al., 2015), beta-diversity is a key metric in ecological studies (Chase and Myers, 2011) and it has been recently proposed as a conservation and monitoring tool (Hillebrand et al., 2018). It represents a powerful tool to evaluate the similarity (i.e. stability) of a community over time, with the rationale that stronger disturbances determine lower stability of communities (Doretto et al., 2018b; Stubbington et al., 2019). Moreover, decomposing the total beta-diversity into its nestedness (i.e. species losses/gains) and turnover (i.e. species replacement) components provides a quantitative evaluation of the specific response of biological communities along a disturbance gradient (Baselga, 2010; Cardoso et al., 2015).

Previous research (Buendia et al., 2013; Doretto et al., 2017; Espa et al., 2019) demonstrated that increased fine-sediment loading results in the depletion of biodiversity through the loss of the most sensitive taxa (i.e. nestedness effect) rather than in species replacement, especially in relation to anthropogenic activities. However, the link between these mechanistic changes in community composition and stressor-specific biomonitoring indices requires in our opinion a more thorough investigation.

In this study, we aimed to perform a screening of currently available biomonitoring tools to evaluate the CSFO impact on the riverine biota and adequately assess its recovery. For this purpose, we investigated the temporal response of macroinvertebrate communities to the CSFO disturbance by analysing two independent datasets of benthic macroinvertebrate communities sampled before/after CSFOs from hydropower reservoirs located in the central Italian Alps (Espa et al., 2019). The first dataset (hereafter case-study I) concerns one sampling site, where similar CSFOs were performed once per year for four consecutive years, and was used to investigate the recovery of macroinvertebrate communities in terms of days since sediment flushing along a pluriannual time frame. The second dataset (hereafter case-study II) concerns a control and an impacted site affected by a single, short CSFO and allowed us to assess its impact at a seasonal scale, through a before/after-control/impact (BACI) approach. As riverine communities can be shaped by the species phenology and habitat heterogeneity across seasons, especially in Alpine streams, case-study II allowed us to focus on the seasonal comparisons, providing a complementary approach to assess the temporal recovery alternative to drawing trajectory over time since the CSFO.

The effects of CSFOs on the richness and beta-diversity of macroinvertebrate communities were first quantified by combining multivariate and univariate statistical techniques. Then, different biomonitoring indices, including generic (i.e. the STAR_ICMi) and siltation-specific (i.e. the PSI and SILTES index) ones, were tested to evaluate their performance in detecting macroinvertebrate response to CSFO. In particular, we hypothesized that: i) CSFOs determine a nestedness effect (i.e. taxa loss); and ii) siltation-specific indices are more effective than generic indices in detecting the impact of CSFOs, since the formers are more tightly linked to the hypothesized nestedness effect. Finally, among the siltation-specific indices, after verifying the adequacy of the SILTES index proposed by Doretto et al. (2018a) to identify the potential impact of CSFOs on the Alpine zoobenthic assemblages, we proposed a threshold distinguishing reference from impacted conditions, which

enables to assess the temporal recovery at different timescale.

2. Material and methods

2.1. Study context

The analysed CSFOs occurred in the Valgrosina and Madesimo hydropower reservoirs, respectively located in the catchments of the Adda and Mera rivers, i.e. the main tributaries of Lake Como (Northern Italy, Fig. 1). Essential information concerning the mentioned CSFOs is summarized in the following sub-sections, referring to specific bibliography for additional detail.

2.1.1. Case-study I: Effects of multi-annual CSFOs

CSFOs at Valgrosina Reservoir (1.3 Mm^3 storage capacity) were operated following the same procedure for four consecutive years (2006–2009) (Crosa et al., 2010; Espa et al., 2013). Sediment flushing took place between August and September, for 12–13 consecutive days. Monitoring campaigns were carried out at the sampling site R1, ca. 6 km downstream from Valgrosina Reservoir, along the Roasco Stream (Fig. 1). At R1, during the CSFOs, the average water discharge was around 3–4 m^3/s, i.e. quite close to the mean annual natural flow. According to the CSFO schedule, SSC increased up to about 10 g/l during daytime, and decreased below 1 g/l overnight, when the dislodging works by mechanical equipment were interrupted. SSC over the whole operations averaged between 3 and 5 g/l, resulting in a flux of fine sediment at R1 amounting to 17,000 tons approximately per CSFO. Most of the sediment (predominantly silt) was transported in suspension, while only a very low fraction settled on the riverbed (estimated in the R1 area to 1.0–2.5 kg/m^2).

2.1.2. Case-study II: Seasonal effects of a CSFO

CSFO at Madesimo Reservoir (0.13 Mm^3 storage capacity) was performed during three consecutive days in October 2010 (Brignoli et al., 2017; Quadroni et al., 2016). The monitoring campaign concerned two sites located along the Liro Stream, above (ca. 0.1 km, control site L0) and below (ca. 3.5 km, impacted site L1) the junction with the stream impounded by the Madesimo Reservoir (Fig. 1). Clear water was released into the Liro Stream from an upstream reservoir as mitigation measure. At L0, streamflow varied between 10–14 m^3/s during daytime and 4 m^3/s overnight, with an additional contribution of 1.4 m^3/s at L1. In analogy to case-study I, SSC at L1 displayed diurnal peaks of 10–15 g/l and nocturnal values below 1 g/l, averaging 2.9 g/l over the entire operation (6 g/l when considering sediment transported by bedload). The evacuated mass of sediment (mainly sand) was ca. 22,000 tons, giving deposition per unit area of about 30 kg/m^2 in the L1 area. Since our previous analysis excluded a relevant impact of flow increases alone during the CSFO on benthic macroinvertebrate community composition (Quadroni et al., 2016), we considered L0 as a good control site.

2.2. Benthic macroinvertebrate monitoring

Biomonitoring was carried out in step-pool or riffle reaches characterized by rather coarse substrate, varying from boulders to pebbles. In the study area, natural runoff is essentially driven by snowmelt in spring and early summer, and by rainfall, occasionally intense, in summer and autumn. In the investigated streams, due to the massive hydropower exploitation, the streamflow is highly regulated and it is generally close to the mandatory minimum flow (i.e. 5–10% of the mean annual flow, established since 2009) in absence of significant contribution by the residual unexploited basin. Hydropower has a long history in the study area: Roasco Stream was initially impounded in 1925, by a small dam located short distance below the present Valgrosina Dam, which was closed in 1960. Madesimo Dam was completed in 1964. The first CSFOs performed in the study area are the documented ones (i.e. 2006 and 2010 for Roasco and Liro streams, respectively). SSC at baseflow is usually lower than 10 mg/l and no relevant chemical alteration related to anthropogenic activity is documented at the investigated sites (Quadroni et al., 2017).

In case study I, benthic macroinvertebrate sampling was performed three times before the CSFOs (i.e. from 284 to 20 days before the first CSFO), and on three to ten further occasions per year (i.e. the first post-CSFO sampling from 11 to 26 days after the event, the last post-CSFO sampling from 187 to 343 days after the event) to depict the recolonization process, for a total of 26 samples. In case study II, biomonitoring was performed seasonally, in the year before (i.e. from 305 to 48 days

Fig. 1. Location of the two hydropower reservoirs (black triangles) subjected to CSFOs and of the biomonitoring sites (white circles).

before the event) and after (i.e. from 27 to 326 days after the event) the CSFO, for a total of 16 samples. Each sample was obtained by pooling together 10 replicates that were collected with a Surber sampler (0.1 m^2 area and 500 μm mesh), following consolidated quantitative and multi-habitat methods (Doretto et al., 2020; Espa et al., 2013; Quadroni et al., 2016). Replicates were allocated based on the visual estimation of the percentage occurrence (i.e. minimal threshold = 10%) of mineral microhabitats. The benthos was preserved in formalin (4%), identified to the lowest possible taxonomic level: family or genus (Plecoptera, Ephemeroptera, Turbellaria, and Hirudinea), and counted. Such a systematic resolution represents a commonly used trade-off between the need of taxonomic accuracy and practical issues. Moreover, this mixed taxonomic level was used for all data analysis, except for the calculation of the STAR_ICMi because this must be computed at family level according to the current Italian normative (see below).

2.3. Data analysis

2.3.1. Case-study I: Effects of multi-annual CSFOs

To test our first hypothesis, differences in the community composition between sampling periods (i.e. pre-CSFO and post-CSFO, from the first - F1 - to the fourth - F4 - CSFO) were visually and statistically examined by non-metric multidimensional scaling (NMDS) and permutational multivariate analysis of variance (PERMANOVA), respectively. The Bray-Curtis dissimilarity was applied to macroinvertebrate abundances of each sampling occasion.

In a second step, we calculated the total beta-diversity and decomposed it into nestedness and turnover components to evidence the mechanisms underlying the differences in the taxonomic composition between sampling occasions. Total pair-wise dissimilarities among samples (total beta-diversity) and their replacement (i.e. turnover) and richness (i.e. nestedness) components were calculated following Cardoso et al. (2015). In particular, pre-CSFO samples were averaged and used as a reference community. Then, the total beta-diversity and the relative contribution of nestedness and turnover were calculated for each post-CSFO sampling occasion using the Sorensen index of dissimilarity. In this way, we obtained an average measure of the distance of the samples perturbed by CSFO from the reference community.

To test our second hypothesis, we performed Generalized Additive Mixed Models (GAMMs) after visually inspecting for the non-linear response of community metrics and biomonitoring indices to CSFOs (i.e. days since flushing event). We adopted mixed effect models to take into account the temporal dependency of our data: hence, the sampling period (i.e. post-CSFO, from F1 to F4) was included as random factor, while the variable time, expressed as days since flushing event, was included as fixed factor in the models. We ran individual GAMMs for taxon richness, beta-diversity, percentage of nestedness and three biomonitoring indices: STAR_ICMi (Buffagni and Erba, 2007), PSI (Extence et al., 2013) and SILTES (Doretto et al., 2018a). As mentioned in the Introduction, the former is the normative biomonitoring index for evaluating the ecological status of Italian rivers according to WFD requirements and it is also the tool currently used to monitor the CSFO impact. The STAR_ICMi is a multimetric index composed of six community metrics, belonging to three categories (i.e. diversity, abundance, and sensitivity/tolerance), and normalized by reference values. It is ranked into five quality classes (bad, poor, moderate, good, and high), respectively bounded at 0.24, 0.48, 0.71, and 0.95 for the study sites (Buffagni and Erba, 2007). Both reference values and thresholds of quality classes are specific for the Italian hydro-geographic region and river typology (e.g. substrate geology, distance from the source, source typology) and are referred to pristine or poorly disturbed (by anthropogenic pressures) conditions. Differently from the STAR_ICMi, both PSI and SILTES were developed to detect the extent of fine sediment deposits on the riverbeds and the related ecological impact, but in different geographic context (UK lowland rivers and Alpine streams respectively). The PSI value is calculated as the proportion of the most sensitive taxa to fine sediment at the sampling site, adjusted to their range of abundance, and ranges between 0 and 100. Based on its value, also this index is ranked into five quality classes: 0–20 heavily sedimented, 21–40 sedimented, 41–60 moderately sedimented, 61–80 slightly sedimented, 81–100 minimally sedimented/unsedimented. The SILTES index was calculated as the average of the values of the three mentioned metrics (i.e. taxon and EPT richness, and ecological group A) scaled accounting for the whole dataset, i.e. by subtracting the value of the metric of the considered sample for the minimum value of this metric observed in the dataset and dividing the obtained value by the range min–max of the metric. The index value ranges from 0 (worst condition) to 1 (best condition) but quality classes have been not yet developed.

When considering beta-diversity, we focused our attention only on the nestedness component because it proved to explain the greatest part of the total beta-diversity for the study dataset. The Poisson and binomial distribution were specified in the models for count and percentage data, respectively, while a Thin Plate Regression Spline was used as smoothing method in the models.

Finally, to assess the performance and specificity of the selected indices to CSFO disturbance, their correlation with the contribution of nestedness to total beta-diversity, expressed as percentage, during the post-CSFO recovery, was statistically tested with a Pearson correlation test.

As the GAMM performed on the SILTES index was significant and displayed a high correlation with the nestedness component, we used this dataset to identify a threshold, which would allow us to discriminate pristine from disturbed conditions. We therefore transformed the index values referred to the post-CSFOs into an Ecological Quality Ratio (EQR SILTES) (Hering et al., 2006a) by calculating the ratio between the index value observed in every post-CSFOs sampling occasion and the mean index value observed in the pre-CSFOs samples. The threshold was then calculated as the average EQR value observed in the temporal interval where the slopes of the relationships between response and predictor reached its peak before decreasing again (Aspin et al., 2019; Yin et al., 2017). Finally, the suitability of this threshold was compared to that of the good quality class determined through the STAR_ICMi.

2.3.2. Case-study II: Seasonal effects of a CSFO

To test our first hypothesis, beta-diversity and its two components were calculated considering the seasonality of benthic assemblages. We first compared post-CSFO samples to the pre-CSFO ones collected in the same season within each site (L0 and L1). Then, we compared samples collected at the impacted site (L1) to the corresponding ones collected at the control site (L0).

Moreover, regarding the second hypothesis, the STAR_ICMi, the PSI and the SILTES were computed for both the sites and compared using the BACI approach.

Statistical analyses were performed in R (R Core Team, 2019), using basic functions and the following packages: vegan (Oksanen et al., 2015) for NMDS and PERMANOVA, BAT (Cardoso et al., 2020) for beta-diversity decomposition and mgcv (Wood and Wood, 2015) for GAMMs.

3. Results

3.1. Case-study I: Effects of multi-annual CSFOs

A total of 62,881 macroinvertebrates were collected, belonging to 35 different taxa. *Baetis* sp., *Leuctra* sp., Chironomidae, Simuliidae and Limnephilidae were the most abundant taxa, accounting for 85% of the whole macroinvertebrate community. The average number of taxa per sample was 14, while the average density was 2,418 individuals/m^2.

NMDS ordination and PERMANOVA did not depict significant differences in the taxonomic composition of benthic communities between sampling periods ($F_{4,25}$ = 0.758; p = 0.803). However, multivariate analysis clearly showed that macroinvertebrate communities collected on the first sampling date after each CSFO were oriented toward the

right side of the plot and separated from the other samples (Fig. 2).

When the differences in the community composition between the pre- and post-CSFO samples were analysed by decomposing the total beta-diversity, we generally found a prevailing contribution of nestedness rather than turnover (Fig. 2). In particular, the percentage contribution of nestedness was highest on the first sampling occasions after CSFOs, whereas the relative contribution of turnover increased over time (Fig. 2).

Due to the high comparability of the CSFOs, we focused on the post-CSFO response of benthic communities. When looking at the total taxon richness (i.e. alpha diversity), we found that the number of macroinvertebrate taxa significantly increased over time and peaked around 200 days since flushing event (Table 1, Fig. 3a). Beta-diversity was instead highest on the first days after CSFOs, progressively decreased within 100 days, and then increased again, in particular between 240 and 270 days since flushing event (F3), when we observed the highest percentage contribution of turnover (Fig. 2b and 3b). We found the same significant pattern (Table 1) for the percentage contribution of nestedness over the first 100 days, but then the nestedness remained almost constant (Fig. 3c).

When the post-CSFO response of macroinvertebrate communities was investigated by applying biomonitoring indices, we found significant patterns (Table 1) for STAR_ICMi (Fig. 4a) and SILTES (Fig. 4b), while PSI did not vary significantly and values fell always in the classes slightly sedimented and minimally sedimented/unsedimented (Fig. 4c). STAR_ICMi was lowest on the first days after CSFOs and then progressively increased over time, achieving a plateau around 200 days since flushing event (Fig. 4a). By contrast, the SILTES index showed a pronounced increment during the initial stage of post-CSFO recovery and peaked around 200 days since flushing event. Then it deceased again over time (Fig. 4b). Interestingly, these two biomonitoring indices were strongly correlated (Fig. 4d) and both were negatively and significantly correlated to the percentage contribution of nestedness during the post-

Table 1
Statistics of the Generalize Additive Mixed Models testing the effect of time (i.e. days since flushing events) on macroinvertebrate metrics and indices: Int = intercept, SE = standard error, t = t-value, F = F-value, p = p-value. Significant values are in bold.

Metric	Int	SE	t	F	p
Taxon richness	2.535	0.071	35.66	9.873	**0.001**
Beta-diversity	0.702	0.014	50.91	17.75	**<0.001**
Nestedness	0.667	0.029	22.57	11.61	**<0.001**
STAR_ICMi	0.826	0.022	38.26	11.99	**<0.001**
PSI	2.043	0.669	3.054	0.001	0.978
SILTES	0.305	0.034	8.852	17.43	**<0.001**

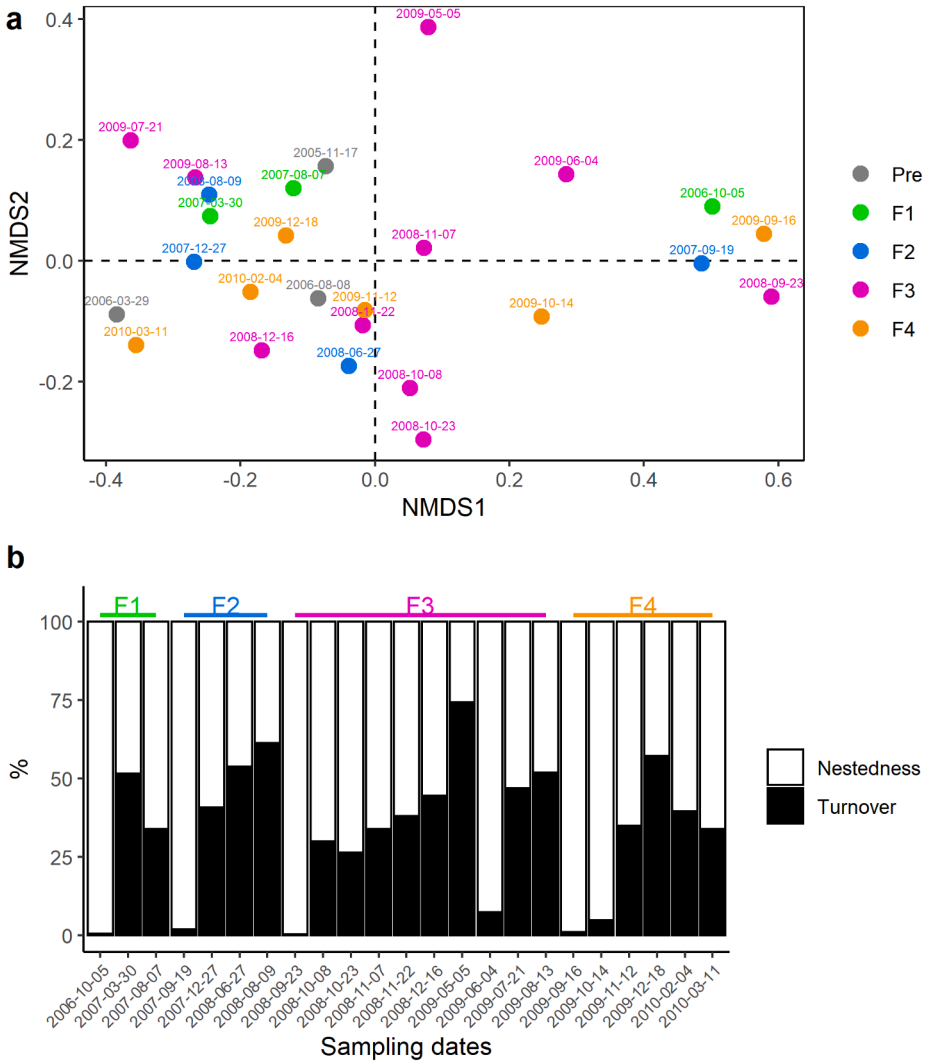

Fig. 2. NMDS ordination plot (a): dots represent sampling dates, colours indicate the sampling period (i.e. pre-CSFO and post-CSFO, from F1 to F4). Labels above dots indicate sampling dates (year-month-day). Stacked bars (b) represent the percentage contribution of nestedness and turnover to the overall beta-diversity between sampling dates. Pre-flushing samples were used as reference communities and labels above bars indicate the four consecutive CSFOs (F1-F4).

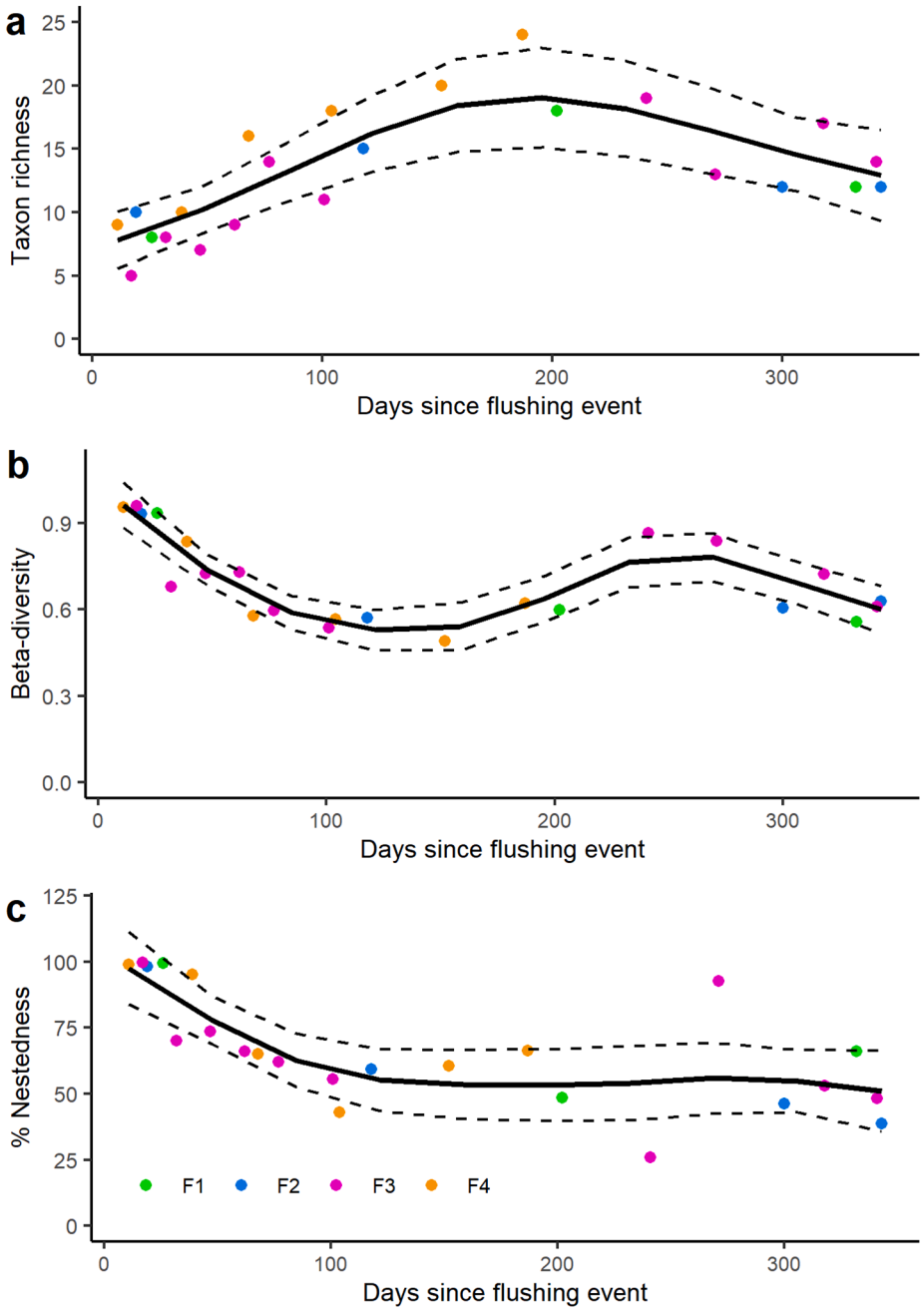

Fig. 3. Generalize Additive Mixed Models for post-CSFO variation of: (a) taxon richness, (b) beta-diversity, and (c) nestedness percentage contribution. Black line represents the predicted values, while the dotted lines represent the 95% confidence interval. Dots indicate sampling occasions, while colours indicate the sampling period (i.e. post-CSFO, from F1 to F4).

CSFO recovery (Fig. 4e and 4f).

As the GAMM of SILTES showed a clear change in slope between 100 and 200 days since the CSFOs, we calculated the average value of the EQR SILTES within this interval, and we obtained the threshold value of 0.75. When analysing the trend of the SILTES index along the sampling dates after CSFOs, we could observe how its values were always lower than our threshold of 0.75 in samples collected before 100 days after each CSFO. In particular, this threshold was reached after the 4th CSFO, when multiple samples were collected between 100 and 200 days after the CSFO. Conversely, the STAR_ICMi indicated good quality class before 100 days after each CSFO (Table 2).

3.2. Case-study II: Seasonal effects of a CSFO

A total of 69,321 macroinvertebrates were collected, belonging to 52 different taxa. *Amphinemura* sp., *Leuctra* sp., *Baetis* sp., Chironomidae, Simuliidae and Limnephilidae were the most abundant taxa, accounting for 84% of the whole community. The average taxon richness and density (individuals/m^2) per sample were 22 and 2,888, respectively.

When the two sampling sites were analysed individually by using the pre-CSFO samples as reference communities, total beta-diversity remained almost constant during the post-CSFO period at the control site (L0). The percentages of nestedness and turnover equalled in autumn and winter, while an increase of nestedness and turnover were recorded in spring and summer, respectively (Fig. 5a). Differently, at the impacted site (L1), beta-diversity decreased during the post-CSFO

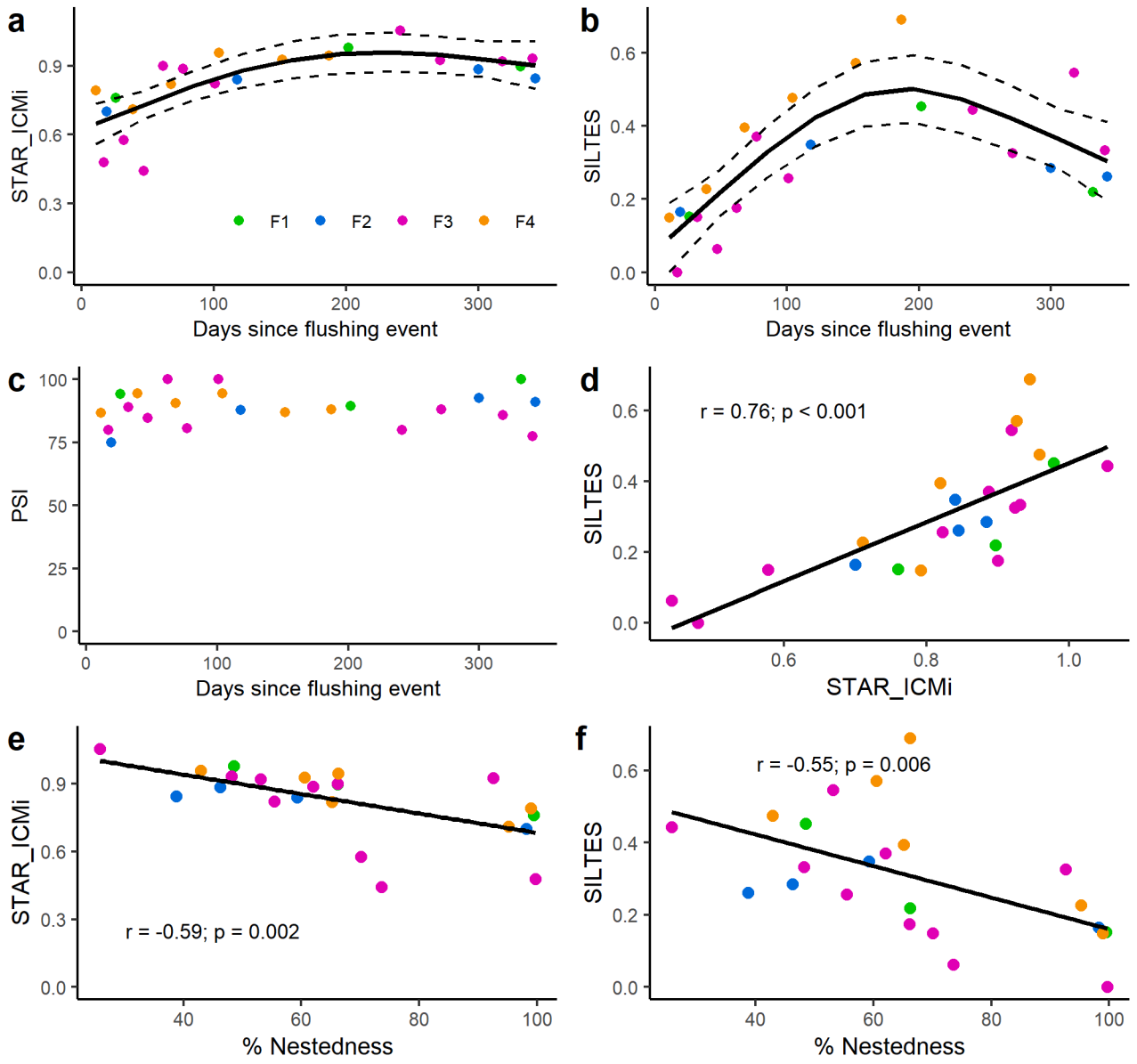

Fig. 4. Generalize Additive Mixed Models for post-CSFO variation of the three considered indexes: (a) STAR_ICMi, (b) SILTES, and (c) PSI. Black line represents the predicted values, while the dotted lines represent the 95% confidence interval. Correlations (r and p values) between the former two indexes (d), and between these indexes and the nestedness percentage contribution (e and f) are also shown. Dots indicate sampling occasions, while colours indicate the sampling period (i.e. post-CSFO, from F1 to F4).

period, showing the highest dissimilarity from the pre-CSFO community in the first post-CSFO sample. Correspondingly, the percentage contribution of nestedness to the total beta-diversity was 100% immediately after the CSFO (i.e. in autumn), then it decreased in winter and increased again in spring and summer (Fig. 5b).

When we compared the impacted site (L1) to the control site (L0) assumed as a reference, we noticed that pre- and post-CSFO seasonal values of beta-diversity were basically similar, except for spring. In this season, a higher dissimilarity from the control site was observed after the CSFO. Also in this case, an increase of nestedness percentage contribution was detected in the first post-CSFO sample (Fig. 5c). However, in winter this contribution decreased to a value similar to that recorded in the same season during the pre-CSFO period (Fig. 5c). In spring, nestedness was higher in the post- than in the pre-CSFO period, while in summer it was lower (Fig. 5c).

The pattern of nestedness and turnover detected during the pre-CSFO period at L1 was similar to that recorded during the post-CSFO period at L0, using in both cases pre-CSFO samples at L0 as reference (Fig. 5a and 5c). Except for spring, both the patterns detected during the post-CSFO period at L1, i.e. using as reference in one case the corresponding pre-CSFO samples at L1 (Fig. 5b) and in the other case the corresponding post-CSFO samples at L0 (Fig. 5c), were similar.

The STAR_ICMi values of samples collected after the CSFO showed variations smaller than that of the pre-CSFO samples at both control (Fig. 6a) and impacted site (Fig. 6b), and were always above the good quality threshold. The SILTES index, instead, presented larger variations before and after the CSFO at both sites. Particularly, it increased at L0 while decreased at L1 in the first post-CSFO sample (Fig. 6c and 6d). During the post-CSFO period this index had values always lower than the corresponding pre-CSFO ones at the impacted site (L1), except for the last sample (i.e. summer, Fig. 6d), collected approximately one year after the CSFO. The PSI did not vary significantly neither between control and impacted site, nor between before/after CSFO at the impacted site (Fig. 6e and 6f). Moreover, the values at the impacted site

Table 2
Values of the EQR SILTES and STAR_ICMi after each CSFO. EQR SILTES values greater than the threshold of 0.75, and STAR_ICMi values greater than 0.71 (limit value of the good quality class) are in bold.

CSFO	Days after CSFO	EQR SILTES	STAR_ICMi
1st	26	0.24	**0.76**
	202	0.72	**0.98**
	332	0.35	**0.90**
2nd	19	0.26	0.70
	118	0.56	**0.84**
	300	0.45	**0.88**
	343	0.42	**0.85**
3rd	17	0.00	0.48
	32	0.24	0.58
	47	0.10	0.44
	62	0.28	**0.90**
	77	0.59	**0.89**
	101	0.41	**0.82**
	241	0.71	**1.00**
	271	0.52	**0.92**
	318	**0.87**	**0.92**
	341	0.53	**0.93**
4th	11	0.24	**0.79**
	39	0.36	**0.71**
	68	0.63	**0.82**
	104	**0.76**	**0.96**
	152	**0.91**	**0.93**
	187	**1.10**	**0.95**

after the CSFO were above the threshold for the minimally sedimented/unsedimented class.

4. Discussion and conclusions

Current environmental policies at a global scale are mainly addressed towards the setting or recovery of a sustainable use of natural resources, i.e. a balance between ecosystem health and human needs (Reyjol et al., 2014). To reach this aim, the knowledge of cause-effect relationships between human activities and ecosystem impact is fundamental to develop adequate and reliable monitoring tools (Bonada et al., 2006). In the European Union, the WFD (2000/60/EC) committed each member state to develop monitoring tools and plans to evaluate the achievement of an overall good ecological status of watercourses. This led to the development of generic biological indexes, as the Italian STAR_ICMi (Buffagni and Furse, 2006), revealing not fully adequate when applied to specific pressures, including siltation (Doretto et al., 2019; Espa et al., 2015) and flow-related alterations (Larsen et al., 2019).

To fill this gap, in this work we tried to test the effectiveness of a biomonitoring tool for the management of CSFOs (or similar human activities causing sediment pressure to river environment), starting from the evidence of their impact on an important component of the stream ecosystem, i.e. benthic macroinvertebrates (Buss et al., 2015).

Gutiérrez-Cánovas et al. (2013) pointed out that anthropogenic stressors that have limited occurrence in nature, like CSFOs, result in the

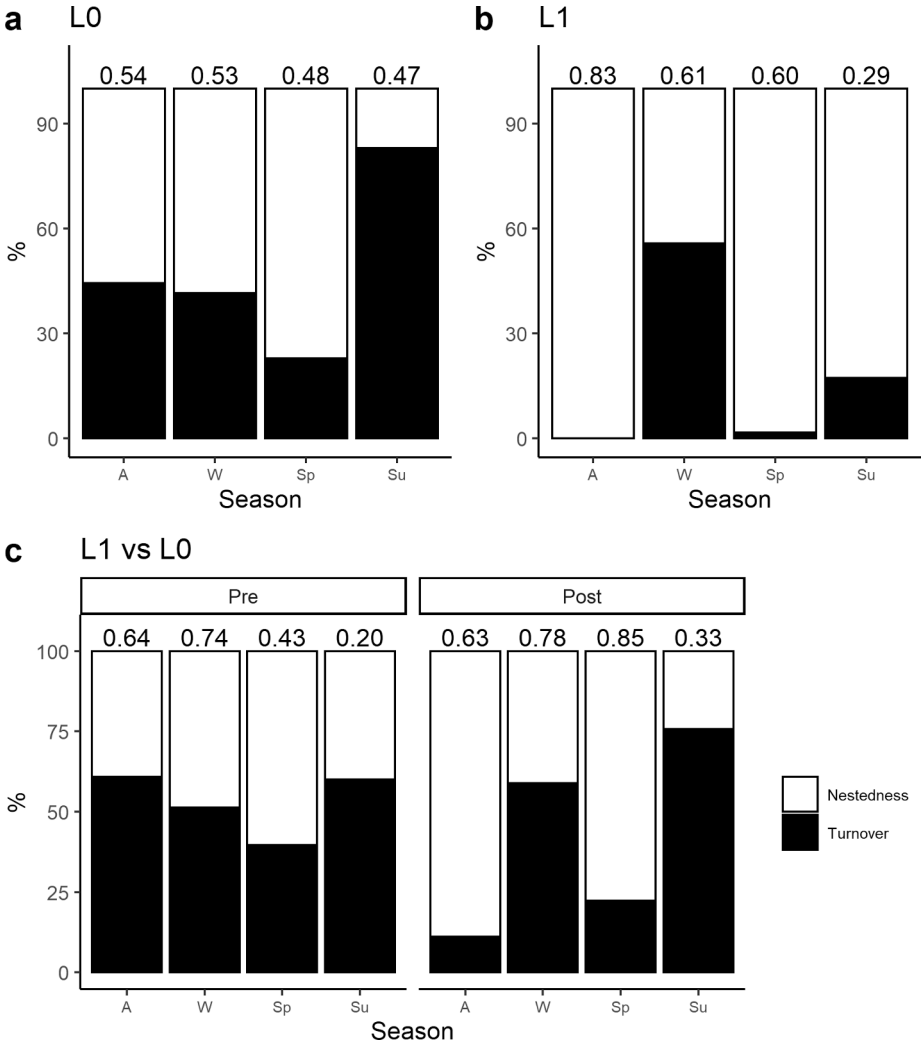

Fig. 5. Stacked bars represent the percentage contribution of nestedness and turnover to the overall beta-diversity (i.e. numbers above bars) between seasons. Pre-CSFO samples within each site, L0 (a) and L1 (b), and season (A = autumn, W = winter, Sp = spring, Su = summer) separately, were used as reference communities. In the case of the impacted site L1, nestedness and turnover contribution was also calculated using the corresponding samples of L0 (control site), and both pre- and post-CSFO periods were considered (c).

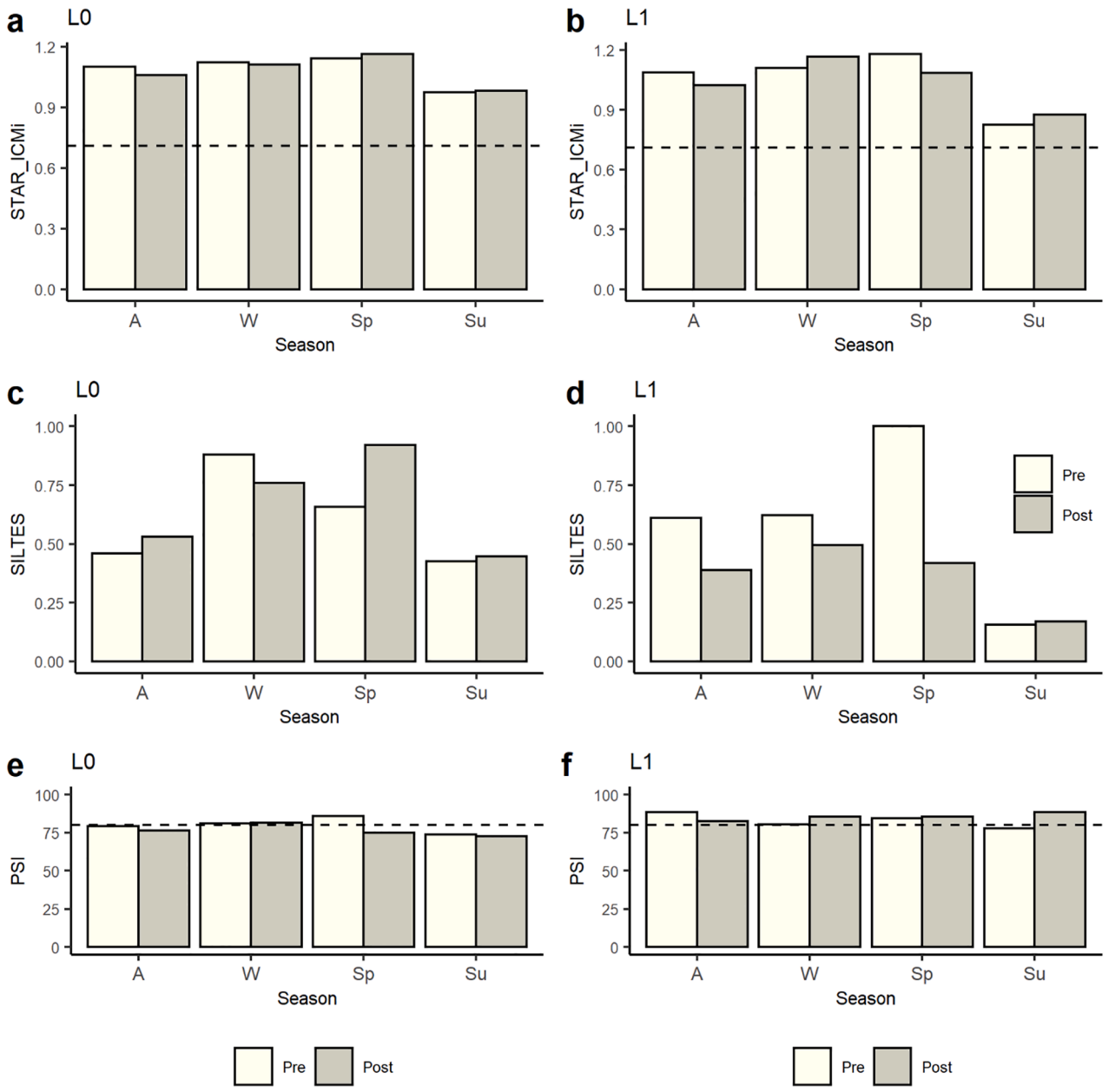

Fig. 6. Before/after-CSFO comparison of the values of STAR_ICMi (a and b), SILTES (c and d), and PSI (e and f) for the control (L0) and impacted (L1) site respectively. Horizontal dashed lines in figures a and b represent the good quality threshold of STAR_ICMi, in figures e and f the threshold of the PSI between minimally sedimented/unsedimented and higher sedimentation extent.

loss of specialist taxa, thus giving rise to nestedness effect. Our results corroborate this evidence, because we found that CSFOs significantly reduced the total macroinvertebrate richness, generating nestedness-driven changes in beta-diversity of impacted communities. This was particularly evident in the first case-study, where nestedness was always highest on the first sample after each CSFO within a month from the end of the activities, and then a recovery was observed within approximately three months. When considering the effects of CSFO at seasonal scale with case-study II, we found similar results. Nestedness mostly explained changes in the communities of the impacted site for the majority of the seasonal comparisons with pre-impact communities of the same site and with post-impact communities of the control site.

Overall, these findings indicate that the sediment load associated with CSFOs acts as an environmental filter by selecting the most tolerant taxa, so that macroinvertebrate communities affected by this disturbance are usually a subset of the more diverse pre-impact or reference communities, as shown by previous research (Doretto et al., 2019; Espa et al., 2016). Moreover, our study showed that the analysis of beta-diversity represents a sound tool to detect mechanistic changes in taxonomic composition of benthic communities related to sediment pressure, as highlighted by other authors (Buendia et al., 2014; Doretto et al., 2017).

Although nestedness resulted the dominant mechanism in both case studies, the BACI experimental design of the second case study showed a more pronounced role of turnover at seasonal scale. This phenomenon is likely associated to the high-flow period typical of Alpine streams, species phenology and inter-annual variability due to different climatic conditions, along with a non-negligible sediment deposition that characterized the investigated CSFO (Quadroni et al., 2016; 2017). This is consistent with the field results by Gabbud et al. (2019) that emphasize the role of spatio-temporal complexity for a correct ecological assessment of Alpine streams. The second case-study, hence, allowed us to

demonstrate the importance of seasonal monitoring of benthic macroinvertebrates and the higher suitability of before samples collected at the impacted site than samples collected at the control site to determine reference condition. In fact, possible differences between pre-CSFO communities sampled at the control and impacted sites could be induced by the different distance of the two sites from dams and intakes in the regulated catchment, and thus to different streamflow alteration (Quadroni et al., 2017).

These results were confirmed and even strengthened by the application of biomonitoring indices. Between the two sediment-specific indexes, only the index developed by Doretto et al. (2018a) (SILTES – Siltation Index for LoTic EcoSystems) showed a significant and negative correlation with the percentage contribution of nestedness and, in turn, revealed appropriate to detect the CSFO impact. We acknowledge that two out of three metrics composing this multi-metric index are richness metrics, i.e. total taxon richness and EPT richness. This aspect explains the high correlation between SILTES and nestedness, and confirms that the adoption of richness-based metrics is effective when disturbances inducing taxa loss at community level are considered. By contrast, the PSI was not correlated to nestedness and did not respond to the investigated pressure. Reasons justifying this poor performance may include the bio-geographical taxon-specific scores assigned to macroinvertebrates, precluding PSI extension to Alpine streams. Moreover, Chironomidae are not considered in this biomonitoring tool, but this midge family is often abundant, and specifically associated to deposition of fine sediment (Kochersberger et al., 2012). Both these aspects probably explain the lack of correlation between the PSI and the percentage of nestedness observed in this study. Furthermore, the similar performance of PSI in both case-studies excluded a relation to the specific sediment input (mainly high suspended sediment load during the CSFO in case-study I, but also relevant sediment deposition after the CSFO in case-study II). In contrast, as also documented by previous studies of sedimentation events characterized by different duration, sediment load, and persistency of deposition (Doretto et al., 2019; Salmaso et al., 2020), SILTES revealed effective in both the analyzed cases.

The STAR_ICMi also showed a strong correlation with the nestedness, which is probably explained by the metrics composing this biomonitoring tool. In fact, similarly to SILTES, the STAR_ICMi is a multimetric index based on different metrics, and three of them are richness or diversity metrics (i.e. total family richness, EPT richness and Shannon-Wiener index). Nevertheless, a bias between the extent of community impairment expressed by the nestedness and the stream quality class expressed by the index was detected. For instance, in case-study I, STAR_ICMi indicated good ecological quality class on most of the sampling occasions, while the threshold proposed for SILTES showed that recovery was achieved only in some sampling occasions, especially after 100–200 days since CSFO. This poor sensitivity was even more evident for case-study II, where stream quality classes expressed by STAR_ICMi were the same for any season at control and impacted sites. According to previous observations and to the recognized sensitivity of the index to organic pollution (Azzellino et al., 2015; Bo et al., 2017; Larsen et al., 2019; Quadroni et al., 2017), this result was expected. Our study pointed out how a stressor-specific index is necessary to correctly assess streams subjected to CSFOs. Both the threshold value we proposed for the SILTES index in the first case-study and the seasonal before/after comparison approach used in the second case-study reflect indeed the recovery of the community from the nestedness effect caused by the CSFO (Espa et al., 2013; Quadroni et al., 2016).

To conclude, in both case-studies the SILTES index proposed by Doretto et al. (2018a) could properly summarize the perturbation of the macroinvertebrate assemblages associated with CSFOs, with useful information on their temporal trajectories in terms of both post-disturbance recovery and seasonal comparison. This is in accordance with the idea that multi-metric indices are the best biomonitoring tools with respect to stressor-specific impacts because of the selection of the best suitable and related metrics (Birk et al., 2012; Bonada et al., 2006; Hering et al., 2006b). Moreover, such a specificity in relation to the ecological consequences of CSFOs is probably enhanced by the effectiveness of SILTES in describing the mechanism by which CSFOs impact on the macroinvertebrate communities (i.e. strong correlation with nestedness effect). To our knowledge, this is the first study that evaluates the performance of a fine-sediment stressor-specific index by correlating it with the main mechanism of biological impairment due to the sediment pressures. Finally, when considering quality classes, we provided two approaches that could be used to verify the achievement of the ecological target (i.e. the recovery to pristine conditions), which gave better results than all the other candidate indices. We thus suggest that the SILTES index could become a suitable and straightforward tool to manage CSFOs and other sediment-related activities in mountainous contexts.

CRediT authorship contribution statement

Alberto Doretto: Conceptualization, Formal analysis, Writing - original draft, Writing - review & editing. **Elena Piano:** Formal analysis, Writing - review & editing. **Stefano Fenoglio:** Writing - review & editing. **Francesca Bona:** Writing - review & editing. **Giuseppe Crosa:** Writing - review & editing. **Paolo Espa:** Writing - review & editing. **Silvia Quadroni:** Conceptualization, Formal analysis, Writing - original draft, Writing - review & editing.

Declaration of Competing 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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