Taxon-specific sensitivities to flow intermittence reveal macroinvertebrates as potential bioindicators of intermittent rivers and streams

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Abstract

As complex mosaics of lotic, lentic, and terrestrial habitats, intermittent rivers and ephemeral streams (IRES) support high biodiversity. Despite their ecological importance, IRES are poorly represented in routine monitoring programs, but recent recognition of their considerable—and increasing—spatiotemporal extent is motivating efforts to better represent IRES in ecological status assessments. We examine response patterns of aquatic macroinvertebrate communities and taxa to flow intermittence (FI) across three European climatic regions. We used self-organizing map (SOM) to ordinate and classify sampling sites based on community structure in regions with continental, mediterranean and oceanic climates. The SOM passively introduced FI, quantified as the mean annual % flow, and visualized its variability across classified communities, revealing a clear association between community structure and FI in all regions. Indicator species analysis identified taxa indicative of low, intermediate and high FI. In the continental region, the amphipod *Niphargus* was indicative of high FI and was associated with groundwater-fed IRES, whereas indicators of mediterranean IRES comprised Odonata, Coleoptera and Heteroptera taxa, which favor lentic conditions. In the oceanic region, taxa indicative of relatively high FI included leuctrid stoneflies and a limnephilid caddisfly, likely reflecting the colonization of IRES by aerial adults from nearby perennial reaches. The Diptera families Chironomidae and Simuliidae showed contrasting FI preferences among regions, reflecting environmental heterogeneity between regions and the coarse taxonomic resolution to which these organisms were identified. These region-specific community and taxon responses of aquatic biota to FI highlight the need to adapt standard biotic indices to enable effective ecological status assessments in IRES.

1. Introduction

Intermittent rivers and ephemeral streams (IRES) are defined as those that sometimes stop flowing, and many recede to isolated pools or dry completely (Leigh et al., 2016; Datry et al., 2017). IRES thus comprise complex mosaics of lotic, lentic and terrestrial habitats that vary in space and time (Larned et al., 2010; Datry et al., 2016a), enabling them to support high biodiversity that contributes to ecosystem processes which in turn deliver ecosystem services (Datry et al., 2018; Stubbington et al., 2020). Although they account for a substantial proportion of the world's waterways (Raymond et al., 2013), IRES have historically been neglected in monitoring, management and water resource assessment plans (AQEM Consortium, 2002; Hering et al., 2006; Stubbington et al., 2018). However, recent recognition of their prevalence and ecological importance is now motivating international efforts to develop effective, IRES-specific ecological status assessment methods (e.g., Gallart et al., 2017; Steward et al., 2018; Stubbington et al., 2018, 2019; Munné et al., 2021). Effective IRES management strategies are urgently needed as climate change causes these ecosystems to increase in spatiotemporal extent in many global regions (Döll and Schmied, 2012; Tramblay et al., 2021; Zipper et al., 2021).

IRES bioassessment is most developed in global regions in which climatic drivers make intermittence particularly widespread, namely dryland regions within the Mediterranean Basin, North America, South Africa and Australia (Dallas, 2013; Mazor et al., 2014; Prat et al., 2014; Munné et al., 2021). Within the Mediterranean Basin, region-specific multimetric indices have been developed for macroinvertebrates which inhabit streams including IRES (Munné and Prat, 2009; Prat et al., 2014). In contrast, in North America, South Africa and Australia, existing methods for assessing the biological integrity of perennial rivers have been adopted in IRES, but their ability to characterize ecological status may vary depending on the time of sampling and on their connectivity with perennial sites (Watson and Dallas, 2013; Mazor et al., 2014; Cid et al., 2020). In particular, status assessments can be compromised by the absence of species sensitive to flow intermittence (FI), which are eliminated from IRES by either flow cessation or streambed drying (Chadd et al., 2017; White et al., 2018; Straka et al., 2019). Such FI-sensitive species include many mayfly, stonefly and caddisfly juveniles, which are also sensitive to environmental degradation, and their absence can thus cause underestimation of ecological status in IRES (Chadd et al., 2017; Stubbington et al., 2018; Cid et al., 2020). Equally, species tolerant of FI could inform adaptation of bioassessment protocols (Cid et al., 2016), despite species richness typically decreasing with FI (Datry et al., 2014; Soria et al., 2017). IRES thus require adapted bioassessment protocols that recognize which taxa may be absent from high-status sites due to their sensitivity to FI (Buffagni et al., 2020; Theodoropoulos et al., 2021).

Aquatic macroinvertebrates are among the most widely used indicators of the ecological status of streams and rivers, due to their abundance, richness, and their diverse, well-known environmental tolerances (Hering et al., 2006; Birk et al., 2012). Many macroinvertebrate-based bioassessment programmes use univariate metrics to represent an assemblage, for example, BMWP-type indices (Armitage et al. 1983), richness metrics and diversity indices. In addition, multimetric indices summarize the biological data provided by multiple individual metrics and can outperform single structural metrics in ecological status assessments (Barbour et al., 1999; AQEM Consortium, 2002; Munné and Prat, 2009). Taxa indicative of specific habitats (including rivers with different FI) can represent informative contributors to multimetric indices, providing more information about stressor-specific responses than diversity metrics, which are sensitive to multiple factors (Carignan and Villard, 2002; Milošević et al., 2015). Identifying taxa indicative of different IRES flow patterns is thus necessary to inform the development of new multimetric indices for IRES bioassessment.

The aim of this study was to characterize macroinvertebrate community and taxon-level response patterns to FI across different climatic regions, and to identify associations of taxa with different FI patterns. Our ultimate goal is to inform adaptation of protocols for IRES bioassessment and to determine the extent to which adaptations could be common across Europe or should be region specific. To this end, we sourced datasets from European regions with contrasting climates, obtaining one continental, one mediterranean and one oceanic dataset. We then analyzed variability in the taxonomic structure of macroinvertebrate communities along an FI gradient in each region. Second, we quantified taxon-specific distributional patterns associated with different FI patterns and identified those associated with different FI regimes. These taxa represent potential bioindicators for exclusion from, or inclusion within, adapted protocols that enable effective IRES bioassessment.

2. Methods

2.1. Datasets and study area characteristics

Datasets were sourced from three countries in different European regions: continental (Albarine River; France), mediterranean (Rivers Llobregat, Ter, Besòs, Tordera and Pineda; Catalonia, NE Spain) and oceanic (Rivers Glen, Lathkill; UK) (Table 1). Data comprised macroinvertebrate samples collected from multiple sites within a river basin with FI gradient length of at least 20%. The sampling networks comprised 31, 17 and nine sites in continental, mediterranean, and oceanic regions respectively, including both perennial (12, 8, 2) and intermittent (19, 9, 7) sites with different FI gradients (Table 1).

The mediterranean study area has a mean annual air temperature of 15 ± 3ºC, a mean maximum temperature 31 ± 4ºC and mean annual rainfall of 560 ± 250 mm (Bonada and Resh, 2013; Cid et al., 2017). Precipitation is concentrated in spring and autumn, with very dry summers and high interannual variability (Munné and Prat, 2011; Bonada and Resh, 2013). These rainfall patterns cause high flow variability, including seasonal, predictable drying and rewetting events in IRES (Cid et al., 2017). IRES typically dry in summer for 2–5 months, with most sites maintaining isolated pools and a few drying completely, and floods then mark the resumption of flow in autumn (Sabater and Tockner, 2009). The mediterranean sites are distributed over four catchments in the province of Barcelona and discharge to the Mediterranean Sea. All are within protected areas and are thus exposed to low human impact levels (Fortuño et al., 2018). Thirteen sites are located in the headwaters on first or second order streams, three sites are third order and one is fourth order.

Within the continental and oceanic regions, precipitation is more evenly distributed throughout the year (Young, 2006). As a result, FI is reduced in spatial and temporal extent, but summer drying can nonetheless be predictable in IRES dominated by groundwater inputs (Sear et al., 1999; Stubbington et al., 2009). The Albarine River is a third-order river that flows through an area of continental eastern France with mean monthly rainfall of 450–606 mm, and a mean annual air temperature of 9.5°C. Seven Albarine study sites are distributed along a 45-km upstream perennial section and 11 are located within a 15-km downstream intermittent section. The intermittent section is underlain by a thick, permeable alluvium, and surface water infiltration into the aquifer causes summer drying for 4–6 months, with flow resumption generally occurring in late autumn/early winter (Datry, 2012).

Four oceanic study sites are in the upper reaches of the River Glen in eastern England (UK), which comprises two third-order tributaries that each flow for approx. 38 km through a predominantly agricultural landscape (Table 1). Mean monthly rainfall varies between 36–61 mm, and the area has a mean annual temperature of 10.5°C (Stubbington et al., 2011a and references therein). Both tributaries alternate between perennial and intermittent reaches, with changes in FI occurring in response to the underlying karst limestone bedrock (Maddock et al., 1995). At each intermittent site, complete drying typically begins in summer lasts up to approx. 3 months. Five UK study sites are on the River Lathkill (UK), a second-order headwater stream which flows for 8.5 km through a wooded valley underlain by karst limestone. Mean monthly rainfall is between 58 mm (August) and 136 mm (December), and the mean annual temperature is 8°C (Stubbington et al., 2011a and references therein). The studied stream section starts approx. 1 km downstream of the source, with two sites located within a <1 km upstream reach in which flow is typically perennial, although riffle crests are exposed during summer. The extent and duration of drying increases with progression downstream within a <2 km stretch, and intermittent sampling sites are located in a reach which dries for 2–3 months from July onwards.

2.2. Characterization of flow intermittence patterns

FI was quantified as the mean annual % flow (i.e., 100% = perennial; 50% = dry for six months per year) using data from the gauging station nearest to each site (i.e., <2 km for continental and <1 km for oceanic) and averaged over the two years before sampling. Due to the absence of flow gauging stations in the mediterranean IRES, their hydrological regimes were characterized using TREHS (Temporary Rivers Ecological and Hydrological Status) software (Gallart et al., 2017), as informed by alternative data sources, including interviews with experts, photos of sampling sites taken in the past 10 years and aerial photos from Google Earth. FI in the continental, mediterranean, and oceanic rivers ranged from 42–100%, 58–100%, and 80–100% flow, respectively.

2.3. Sampling design

The continental sampling campaign was conducted in each spring (prior to water loss) and autumn/early winter (two weeks after flow resumed) from 2008 to 2010. Macroinvertebrates were collected using a Hess sampler (0.125-m^2 area, 200-µm mesh), with two samples collected from each of two riffles and each pair of samples pooled. The mediterranean sampling campaign encompassed each spring and summer from 2012 to 2016. Sampling was conducted in all available habitats, using a kick net with a 250-µm mesh. In the oceanic region, four replicate Surber samples (0.09 m^2 area, 500-µm mesh) were collected from the dominant habitat type at each site at monthly intervals between May and September 2009. All samples were preserved and identified to genus-to-family, family and species-to-family level for continental, mediterranean and oceanic regions, respectively, except for Diptera, which were identified to family, and Oligochaeta and Hydrachnidia, which were identified as such.

2.4. Data analysis

2.4.1. Visualization of community structure

To visualize community structure and the relationship between taxa and % flow, we applied a self-organizing map (SOM), which is an unsupervised type of artificial neural network (ANN), meaning that output values were not provided to the neural network. ANN are mathematical models which use learning algorithms inspired by biological neural networks (Wäldchen and Mäder, 2018) and are built of layers which comprise interconnected neurons. In ecology, Kohonen ANN (Kohonen, 1982), also known as SOM, provide exploratory tools for ordination, clustering, classification and data mining, mostly to examine spatiotemporal patterns in community structure (Park et al., 2004; Chon, 2011; Milošević et al., 2013). Key benefits of SOM include their ability to process large datasets and to accommodate non-linear relationships between variables. In addition, SOM use non-linear transformations, which prevents outliers from biasing the results (Park et al., 2004; Milošević et al., 2015).

SOM comprise an input (Kohonen) and an output layer, which create a two-dimensional ordination space. The number of input neurons is determined by the number of input vectors (here, samples). The output layer is typically composed of hexagonal neurons (Kohonen, 2001), the number of which (i.e., the network resolution) is defined a priori, following established methods (Vesanto et al., 2000; Park et al., 2003). We built separate SOM models for each region, due to high regional variability in community structure as well as differences in sampling designs. Map resolutions of 7 × 6, 8 × 7 and 7 × 7 best fitted the data matrices constructed for the continental, mediterranean and oceanic regions, respectively.

Data matrices composed of samples (vectors), each comprising the abundance of macroinvertebrate taxa (parameters), were log(x + 1)-transformed and then normalized. During the training process, all input vectors went through the SOM and attached to a particular neuron in the output network. Vector positions collectively reflect variability in community structure and were determined by comparing vectors to each SOM neuron using Euclidean distances. Specifically, the neuron which carries the most similar community pattern to the input vector is termed the best-matching unit, and at the end of the training process the most similar vector is attached to it. All samples were ordinated on the network by the training process, with each constituted neuron including sampled communities with similar structures, and with increasing distance between networked neurons indicating increasing differences in community structure.

Output neurons were classified into subgroups with similar community structures using the non-hierarchical k-means method (Jain and Dubes, 1988), with the number of groups determined following the FI distributional pattern on the SOM and including sufficient clustered samples per group to enable univariate statistical analysis. Finally, structural differences between SOM groups, obtained by the k-means method, were tested using permutational multivariate analysis of variance (PERMANOVA; Anderson, 2001), with Bray–Curtis similarity matrices and with 9999 permutations used to estimate the p-value of the pseudo-F statistic.

2.4.2. Characterization of taxa responses to flow intermittence

To identify taxa that responded to FI, we visualized taxa abundances alongside FI (as the mean annual % flow) across the SOM. To define the spatial patterns of FI variability that influenced community structure, FI was passively introduced into the SOM, without influencing the previous ordination and classification. The SOM algorithm calculates the mean value of the introduced parameter for each neuron, here forming a distributional pattern of FI on the trained SOM. This output is termed a component plane, with the distributional patterns indicating whether the parameter influences variability in community structure. Differences in FI among SOM groups of output neurons were identified using non-parametric ANOVA (Kruskal–Wallis) tests with pairwise Mann–Whitney post-hoc tests, because data were not normally distributed. This statistical design identified differences among FI regimes for each SOM model.

2.4.3. Identification of flow intermittence indicator taxa

FI indicator taxa were defined as those whose distributions were associated with a particular range of FI values within each region. To quantify the association of each taxon with particular FI values, we used indicator species analysis (IndVal; Dufrêne and Legendre, 1997). Taxa which had an IndVal value ≥ 25, a p-value <0.05 and appeared in a SOM group characterized by a particular mean annual % flow were considered as indicative of that FI regime; the frequency and relative abundance of such taxa were both consistently $\geq 50\%$.

3. Results

3.1. Community structuring along flow intermittence gradients

The SOM ordinated and classified communities into four groups in all three regions (continental, oceanic and mediterranean; Fig. 1a, 2a and 3a). Community structure differed between SOM groups in the continental (PERMANOVA, pseudo-F=19.3, p<0.001) mediterranean, (pseudo-F=17.4, p<0.001) and oceanic (pseudo-F=39.8, p<0.001) regions. FI (as mean annual % flow) varied among SOM groups in the continental (Fig. 1b) and mediterranean (Fig. 2b) neural networks, while oceanic differences in FI were less pronounced, (Fig. 3b). Differences in % flow between SOM groups were confirmed by Kruskal–Wallis tests (p<0.05) and Mann-Whitney post-hoc tests (p<0.016; Table 2), indicating that FI drove spatial variability in community structure.

Continental SOM groups III and IV had the lowest FI, comprising communities from sites with 99.9% and 93.8% flow, respectively, whereas SOM groups I and II included communities from sites with substantially higher FI: 69.7% and 60.3% flow, respectively (Fig. 1a–b, Table 2). Mediterranean SOM group II included communities from sites with the highest FI (64.0% flow), whereas groups I, III and IV comprised communities at sites with substantially lower FI (90.4%, 95.5% and 94.5% flow, respectively; Fig. 2a–b). In the oceanic dataset, FI was comparable in groups I, III and IV ranging from 92% to 99% flow. Communities at these sites differed from those in SOM group II, which had higher FI (82.1% flow, Fig. 3a–b, Table 2).

3.2. Flow intermittence indicator taxa

IndVal identified taxa indicative of different SOM groups (i.e., different FI, quantified as mean annual % flow) in all regions (Figs. 1c, 2c, and 3c, Table 3). In the continental region, out of 28 FI indicator taxa, the 93.8% flow group (SOM group IV) included the most indicators: 20

taxa with IndVal scores of up to 89%, including several genera within the insect orders Trichoptera and Ephemeroptera (Table 3a). Four taxa were indicative of the highest % flow (99.9%; SOM group III), including one amphipod (*Gammarus*) and two Ephemeroptera. The lower % flow groups (69.7% and 60.3%, i.e. SOM groups I and II) were represented by only three Diptera families (Simuliidae, Ceratopogonidae and Chironomidae) and one amphipod (*Niphargus*), respectively (Table 3a).

The mediterranean dataset included 40 FI indicator taxa (Fig. 2c, Table 3b). Eleven taxa were indicative of sites with 90.4% flow (SOM group I; Table 3b), with families of the insect orders Coleoptera, Ephemeroptera and Trichoptera having IndVal scores >70%. For the highest FI group (64.0% flow; SOM group II), the strongest of five indicator taxa was the Coleoptera family Dytiscidae (IndVal score 71.9%; Table 3b). Thirteen taxa were indicative of the site with 95.5% flow (SOM group III), with the Plecoptera families Leuctridae and Nemouridae having IndVal scores >82%. In SOM group IV (94.5% flow), the amphipod family Gammaridae had extremely high IndVal score (95.7%), followed by the gastropod family Hydrobiidae (82.9%).

The largest number of FI indicator taxa (45) were identified in the oceanic dataset (Fig. 3c, Table 3c). Five taxa were indicative of sites with 82.1% flow (SOM group II), in particular the Plecoptera genus *Leuctra*, as well as the Trichoptera family Limnephilidae (Table 3c). Sites with substantially lower FI (92.6–99.7% flow; SOM groups I, III and IV) had the most indicator taxa, with IndVal scores >78% for the Diptera family Simuliidae, the Trichoptera genus *Hydroptila*, sphaeriid bivalves, and the Elmidae genus *Riolus* (Fig. 3c, Table 3c).

4. Discussion

In highly dynamic systems such as IRES, habitat conditions fluctuate in time and space, shaping the community structure of aquatic biota (Larned et al., 2010; Stubbington et al., 2017). In biotic groups including macroinvertebrate communities, different taxa are characteristic of perennial and intermittent flow regimes (Cañedo-Argüelles et al., 2016; Chadd et al., 2017; Straka et al., 2019). Accordingly, in systems across continental, mediterranean and oceanic climates, we identified family-to-species level taxonomic differences in macroinvertebrate assemblages at sites with contrasting flow intermittence (FI, as mean annual % flow), including a consistent group of taxa indicative of sites with high flow permanence. In contrast, and reflecting the typical decrease in taxonomic α-diversity that accompanies an increase in FI (Datry et al., 2014; Soria et al., 2017), fewer taxa were indicative of sites with higher FI across climatic regions, and all such taxa were region-specific (Table 3), which we suggest as driven by river type, not climate.

4.1. In the continental region, flow intermittence indicator taxa were associated with groundwater-fed IRES

In the continental region, the two SOM clusters comprising sites with relatively high FI (69.7% and 60.3% flow) supported communities with few indicator taxa. Of these few, only *Niphargus* amphipods were indicative of the highest FI, as also observed by White et al. (2018) in 'winterbourne' (i.e., intermittent) streams in oceanic England. A hypogean taxon, *Niphargus* may be transported into both perennial and intermittent reaches by upwelling groundwater (Larned et al., 2011), with its persistence in IRES then promoted by reduced densities of predatory and competitive taxa such as gammarid amphipods (Fišer et al., 2007). When surface water is lost, their relatively low requirements for energy and oxygen may enable *Niphargus* to survive within subsurface interstices in which epigean taxa die (Brunke and Gonser, 1999;

Hahn, 2006). However, the presence of such hypogean taxa may primarily be indicative of local groundwater influence (Datry, 2012), potentially compromising their utility as FI indicators.

Taxa indicative of the second continental group of intermittent (69.7% flow) sites comprised three Diptera families: Ceratopogonidae, Chironomidae and Simuliidae (Table 3a). Each family includes pioneer species which are among the first to colonize after flow resumes in IRES, and which then persist throughout flowing phases (Acuña et al., 2005; Hammock and Bogan, 2014; Cañedo-Argüelles et al., 2016). Morphological traits including small size and elongate shape can facilitate inhabitation of saturated and humid hyporheic interstices by these larvae following surface water loss in IRES, promoting their rapid recolonization (Datry, 2012; Stubbington, 2012; Vander Vorste et al., 2016). In addition, desiccation-tolerant resistance forms allow some chironomids, ceratopogonids (Stubbington and Datry, 2013) and simuliids (Bass, 1998) to persist in situ during dry phases, and their rapid emergence can support community recovery after flow resumes (Bogan et al., 2015, 2017). Some adapted species, such as the simuliid *Metacnephia amphora* in winterbourne streams in south England (Armitage and Bass, 2013) and several chironomids and simuliids in arid south-west USA (Bogan et al., 2013), are IRES specialists that rely on intermittence to complete their lifecycles; such taxa have locally high potential as FI indicators. However, the coarse taxonomic level to which these families are identified typically prevents this potential from being realized (e.g., Bunting et al., 2021).

4.2. In the mediterranean region, flow intermittence indicator taxa were associated with lentic conditions

In the mediterranean region, five families were indicative of samples from sites with the highest FI (64.0% flow), including two Odonata, one Coleoptera and one Heteroptera (Table 3b). All such OCH taxa are associated with lentic waters including IRES pools in mediterranean

(Bonada et al., 2006, 2007, 2020; Buffagni, 2021) and other regions (Hill and Milner, 2018; Valente-Neto et al., 2020), reflecting the capacity of flying adults to colonize aquatic habitats matching their environmental preferences after flow cessation (Papaček, 2001; Samraoui, 2009). For taxa such as the Dytiscidae (Coleoptera) and Notonectidae (Heteroptera), occurrence of adults in IRES pools may be fleeting if they emigrate after water quality or resource availability declines (Bogan et al., 2017). Alternatively, dytiscids can survive dry phases by migrating into subsurface interstices (Fenoglio et al., 2006) such as the true IRES specialists *Agabus brunneus* (Foster, 2010).

In those IRES channels where reproductive activity and larval development is possible, the aquatic juveniles of OCH present in flowing phases remain indicative of FI—and specifically, lentic waters, which in our mediterranean study region, typically comprise isolated pools (Bonada and Resh, 2013). The OCH families associated with high FI in our mediterranean study rivers may thus have high potential as indicators of such IRES in and beyond this region. In contrast, another apparent indicator of high FI, Sphaeriidae (Bivalvia, which had a marginal IndVal score), may occur in both lentic and lotic waters within both perennial and intermittent reaches (Stubbington et al., 2016; White et al., 2018), limiting its indicator value.

4.3. Oceanic indicators of flow intermittence included specialist insects

Within the oceanic region, five taxa (including leuctrid stoneflies identified to both genus and species) were indicative of sites with the highest FI—which was only 82% flow. Of these, *Leuctra* may burrow into saturated subsurface sediments as surface flow declines (Lopez-Rodriguez et al., 2009; Stubbington et al., 2011a). However, these facultative IRES inhabitants have rheophilic habitat preferences, often resulting in their association with perennial flow (Chadd et al., 2017; White et al., 2018) and thus limiting their indicator value. The occurrence of *Leuctra* at our IRES study sites may thus reflect its colonization from nearby upstream and downstream perennial waters during long flowing phases in this cool, wet region (Townsend and Hildrew, 1976; Datry et al., 2007). In contrast, the cased caddisfly family Limnephilidae include true IRES specialists, such as *Ironoquia dubia* (Wallace, 2016) as well as intermittence-associated species (Smith et al., 2003). The humid microhabitat within their cases may enable caddisfly juveniles to persist during dry phases (Larned et al., 2007; Stubbington et al., 2009), although our identification of *Chaetopteryx villosa* and *Drusus annulatus* as indicative of intermittence contrasts with reported association of these species with perennial waters in other UK (i.e., oceanic) limestone streams (Smith et al., 2003; White et al., 2018). The alternation of perennial and intermittent reaches over small spatial scales may enable aquatic dispersal to intermittent sites as flowing phases extend in duration, altering identification of IRES indicators. The snail Ampullaceana balthica was also indicative of the highest FI group, as observed for in previous oceanic studies (Armitage and Bass, 2013; White et al., 2018). However, we note its marginal IndVal (Table 3c; White et al., 2018), and the desiccation sensitivity (Collas et al., 2014) and its association with perennial waters (Datry, 2012) previously reported for the genus.

4.4. Taxa indicative of high flow intermittence were specific to regions—and river types

We identified contrasting FI preferences among regions for several taxa in our continental, mediterranean and oceanic rivers. Although drying is a primary determinant of macroinvertebrate community composition in IRES across regions (Datry et al., 2014; Leigh and Datry, 2017), these contrasts may reflect the considerable environmental heterogeneity within these dynamic ecosystems (Cid et al., 2017; Stubbington et al., 2017), including differences in the location and extent of intermittent sites within river networks (Datry et al., 2016b). For example, the Chironomidae and Simuliidae were indicative of the highest FI group

(69.7% flow) in the continental region, a low FI (90.4%) group in the mediterranean region, and essentially perennial sites (99.7%) in the oceanic region (Table 3). Higher occurrence of these Diptera in continental IRES may reflect drift from particularly extensive perennial upstream reaches in this river (Larned et al., 2011; Datry, 2012), with regional differences also reflecting variability among taxon-specific responses in these large, diverse families. Equally, our identification of Leuctridae as strong indicators of mediterranean sites with near-perennial (i.e., 95.5%) flow (Table 3b) is consistent with their typical habitat preferences (Pace et al., 2013), with dispersal from nearby perennial sites during long flowing phases explaining their occurrence in oceanic intermittent reaches Finally, our identification of *Niphargus* as an indicator taxon only in the continental region likely reflects the strength of upwelling groundwater in intermittent reaches of the Albarine (Larned et al., 2011), whereas intermittent oceanic sites are primarily downwelling, restricting the taxon's occurrence in hyporheic sediments (Stubbington et al., 2011b), and mediterranean sites are fed by surface water.

Contrasting identification of indicator taxa among regions may also reflect differences among regional species pools, including variability which we did not characterize due to the coarse resolution to which some taxa were identified. Our family-level identification of Diptera prevented characterization of the highly diverse habitat preferences of species within ubiquitous families such as Chironomidae (e.g., Armitage et al., 2012)—diversity which causes variability among river types within regions (Cañedo-Argüelles et al., 2016; Milošević et al., 2018) as well as among regions. Species-level identification may be necessary to distinguish indicator taxa, for example the mayfly genus *Paraleptophlebia* has three UK representatives, of which two are associated with perennial flow, whereas *P. werneri* is largely restricted to winterbourne chalk IRES (Macadam, 2016). Without species-level identification, taxa may thus be unreliable as FI indicators.

4.5. Taxa indicative of high flow permanence were consistent across regions

Whereas we identified a small number of region-specific indicators of sites with higher FI (i.e., 60.3–82% flow), those indicative of higher permanence (i.e., 90.4–99.9% flow) were more numerous, and dominated by representatives of comparable orders, including the Ephemeroptera, Plecoptera and Trichoptera (EPT). Of these indicators, taxa with IndVal scores >75% included the genera *Hydropsyche, Ecdyonurus, Habroleptoïdes* and *Torleya* in the continental region; the families Caenidae, Leuctridae, Nemouridae as well as Gammaridae (Amphipoda) and Hydrobiidae (Gastropoda) in the mediterranean region; and *Baetis* as well as *Riolus* (Coleoptera: Elmidae), Simuliidae and Sphaeriidae in the oceanic region. Except for sphaeriids, such taxa are dominated by typical rheophiles which require well-oxygenated waters and are thus common indicators of perennial flow across continental (Straka et al., 2019, 2021), oceanic (Chadd et al., 2017; White et al., 2018) and mediterranean (Bonada et al., 2006, 2007) regions. However, the hydrological connectivity of river networks facilitates aquatic dispersal, allowing rheophiles to colonize intermittent sites, and thus gradually homogenizing communities over time (Sarremejane et al., 2020) and potentially compromising indicator performance after long flowing phases (Bonada et al., 2007).

4.6. Identification of flow intermittence groups by self-organizing map (SOM)

Across regions, SOM classifications identified multiple groups of samples with similar FI, in particular mediterranean groups III–IV (95.5% and 94.5% flow) and oceanic groups I and IV (92.6% and 94.6% flow; Table 3b). These groups likely reflect non-hydrological habitat differences between reaches with comparably low FI. For example, oceanic group I included samples from sites on the Lathkill, a wooded headwater stream, and supported seven EPT indicator taxa which are also indicative of good ecological status; in contrast, oceanic group IV included sites on the Glen, which flows through arable farmland and supported three non-EPT indicator taxa, all with broad environmental tolerances (Paisley et al., 2014).

4.7. Study limitations and future research priorities

Spatiotemporal variability in IRES biota—including differences in aquatic community composition in river sections with different FI regimes—can confound the assessment of ecological status in IRES, prompting calls to better recognize the range of taxa expected in unimpacted rivers with intermittent flow (Stubbington et al., 2018). In response to such calls, our study complements previous region-specific studies by identifying taxa indicative of rivers with different FI across three European regions. As in previous studies (e.g., Chadd et al., 2017; Straka et al., 2019), we identified a consistent range of taxa indicative of high flow permanence—including those with high ecological status requirements (Munné and Prat, 2009; Paisley et al., 2014), which may be absent from unimpacted intermittent reaches (Hughes et al., 2009). We also identified FI indicator taxa that differed markedly among regions and, in some cases, contrasted with those identified by previous research (e.g. White et al. 2018). Such results may reflect limitations of our study, highlighting the need for future research that builds on our findings by consistently characterizing variability within and among regions.

Specifically, we only sourced datasets from European regions with temperate (or, in the case of the Albarine, continental but near-temperate; Datry, 2012) climates. Future research should thus prioritize representation of other climatic regions, including alpine, continental and semi-arid regions, all of which are experiencing increases in river drying (Beniston, 2012; Crabot et al., 2020; Tramblay et al., 2021). In addition, our representation of each region was limited to only 1–5 rivers. Further large-scale research is thus needed to compare and contrast variability in IRES ecosystems within regions, in particular to determine the conditions in which our identified taxa maintain their indicator value. Within each region, a key challenge is to represent the breadth of hydrological, geomorphological and physicochemical factors the characterize the range of river types classified as ‘IRES’ (Stubbington et al., 2017).

Within our study area, our findings may have been influenced by variability in sampling designs among region-specific datasets. First, the use of different sampling methods may have altered representation of habitats and thus taxa, with the comprehensive representation of all macrohabitat types by kick sampling in the mediterranean region being potentially more likely to capture taxa associated with lentic waters, even during flowing phases. Second, the collection of samples in different seasons—including isolated pools in summer in mediterranean IRES—may have promoted sampling of taxa associated with these habitats in this region. As such, in mediterranean IRES, our identified FI indicator taxa may not be indicative not of FI per se, but of the isolated pools that can persist in IRES during summer dry phases. As IRES increase in extent due to global change, we call for ambitious research that builds on our identification of regional FI indicators by characterizing the species and communities that represent the diversity of IRES types across and beyond Europe.

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References

Acuña, V., Muñoz, I., Giorgi, A., Omella, M., Sabater, F., Sabater, S., 2005. Drought and postdrought recovery cycles in an intermittent Mediterranean stream: structural and functional aspects. J. N. Am. Benthol. Soc. 24, 919–933. https://doi.org/10.1899/04-078.1.

Anderson, M.J., 2001. Permutation tests for univariate or multivariate analysis of variance and regression. Can. J. Fish. Aquat. Sci. 58, 626–639. https://doi.org/10.1139/f01-004.

AQEM Consortium, 2002. Manual for the application of the AQEM system. A comprehensive method to assess European streams using benthic macroinvertebrates, developed for the purpose of the Water Framework Directive. Version 1.0.

Armitage, P. D., Moss, D., Wright, J. F., Furse, M. T. 1983. The performance of a new biological water quality score system based on macroinvertebrates over a wide range of unpolluted running-water sites. Water Res. 17, 333–347.

Armitage, P.D., Pinder, L.C., Cranston, P.S. (Eds.), 2012. The Chironomidae: Biology and Ecology of Non-biting Midges. Springer Science & Business Media, Berlin/Heidelberg.

Armitage, P.D., Bass, J., 2013. Long-term resilience and short-term vulnerability of South Winterbourne macroinvertebrates. Proc. Dorset Nat. Hist. Archaeol. Soc. 134, 43–55.

Barbour, M.T., Gerritsen, J., Snyder, B.D., Stribling, J.B., 1999. Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates and Fish, second ed. U.S. Environmental Protection Agency, Office of Water, Washington, D.C.

Bass, J.A.B., 1998. Last-instar larvae and pupae of the Simuliidae of Britain and Ireland: a key with brief ecological notes. Scientific Publications of the Freshwater Biological Association (FBA) 55. FBA, Windermere, UK.

Beniston, M., 2012. Impacts of climatic change on water and associated economic activities in the Swiss Alps. J. Hydrol. 412, 291–296. https://doi.org/10.1016/j.jhydrol.2010.06.046.

Birk, S., Bonne, W., Borja, A., Brucet, S., Courrat, A., Poikane, S., Solimini, A.G., van der Bund, W., Zampoukas, N., Hering, D., 2012. Three hundred ways to assess Europe's surface waters: an almost complete overview of biological methods to implement the Water Framework Directive. Ecol. Ind. 18, 31–41. https://doi.org/10.1016/j.ecolind.2011.10.009.

Bogan, M.T., Boersma, K.S., Lytle, D.A., 2013. Flow intermittency alters longitudinal patterns of invertebrate diversity and assemblage composition in an arid-land stream network. Freshw. Biol., 58, 1016–1028. https://doi.org/10.1111/fwb.12105.

Bogan, M.T., Boersma, K.S., Lytle, D.A., 2015. Resistance and resilience of invertebrate communities to seasonal and supraseasonal drought in arid-land headwater streams. Freshw. Biol. 60, 2547–2558. https://doi.org/10.1111/fwb.12522.

Bogan, M.T., Chester, E.T., Datry, T., Murphy, A.L., Robson, B.J., Ruhi, A., Stubbington, R., Whitney, J.E., 2017. Resistance, resilience, and community recovery in intermittent rivers and ephemeral streams, in: Datry, T., Bonada, N., Boulton, A. (Eds.), Intermittent Rivers and Ephemeral Streams: Ecology and Management. Elsevier, Netherlands, pp. 349–376. https://doi.org/10.1016/B978-0-12-803835-2.00013-9.

Bonada, N., Rieradevall, M., Prat, N., Resh, V.H., 2006. Benthic macroinvertebrate assemblages and macrohabitat connectivity in Mediterranean-climate streams of northern California. J. N. Am. Benthol. Soc. 25, 32–43. https://doi.org/10.1899/0887-3593(2006)25[32:BMAAMC]2.0.CO;2.

Bonada, N., Rieradevall, M., Prat, N., 2007. Macroinvertebrate community structure and biological traits related to flow permanence in a Mediterranean river network. Hydrobiologia 589, 91–106. https://doi.org/10.1007/s10750-007-0723-5.

Bonada, N., Resh, V.H., 2013. Mediterranean-climate streams and rivers: Geographically separated but ecologically comparable freshwater systems. Hydrobiologia 719, 1–29. https://doi.org/10.1007/s10750-013-1634-2.

Bonada, N., Cañedo-Argüelles, M., Gallart, F., Von Schiller, D., Fortuño, P., Latron, J., Llorens, P., Múrria, C., Soria, M., Vinyoles, D., Cid, N., 2020. Conservation and management of isolated pools in temporary rivers. Water 12, 2870. https://doi.org/10.3390/w12102870.

Brunke, M., Gonser, T., 1999. Hyporheic invertebrates: the clinal nature of interstitial communities structured by hydrological exchange and environmental gradients. J. N. Am. Benthol. Soc 18, 344–362. https://doi.org/10.2307/1468448.

Buffagni, A., Erba, S., Cazzola, M., Barca, E., Belfiore, C., 2020. The ratio of lentic to lotic habitat features strongly affects macroinvertebrate metrics used in southern Europe for ecological status classification. Ecol. Ind. 117, 106563. https://doi.org/10.1016/j.ecolind.2020.106563.

Buffagni, A., 2021. The lentic and lotic characteristics of habitats determine the distribution of benthic macroinvertebrates in Mediterranean rivers. Freshw. Biol. 66, 13–34. https://doi.org/10.1111/fwb.13596.

Bunting, G., England, J., Gething, K., Sykes, T., Webb, J., Stubbington, R., 2021. Aquatic and terrestrial invertebrate community responses to drying in chalk streams. Water Environ. J. 35, 229–241. https://doi.org/10.1111/wej.12621.

Cañedo-Argüelles, M., Bogan, M.T., Lytle, D.A., Prat, N., 2016. Are Chironomidae (Diptera) good indicators of water scarcity? Dryland streams as a case study. Ecol. Ind. 71, 155–162. https://doi.org/10.1016/j.ecolind.2016.07.002.

Carignan, V., Villard, M. A., 2002. Selecting indicator species to monitor ecological integrity: a review. Environ. Monit. Assess. 78, 45–61. https://doi.org/10.1023/A:1016136723584.

Chadd, R.P., England, J.A., Constable, D., Dunbar, M.J., Murray-Bligh, J.A., Wood, P.J., Extence, C.A., Leeming, D.J., 2017. An index to track the ecological effects of drought development and recovery on riverine invertebrate communities. Ecol. Ind. 82, 344–356. https://doi.org/10.1016/j.ecolind.2017.06.058.

Chon, T.-S., 2011. Self-Organizing Maps applied to ecological sciences. Ecol. Inform. 6, 50–61. https://doi.org/10.1016/j.ecoinf.2010.11.002.

Cid, N., Verkaik, I., García-Roger, E.M., Rieradevall, M., Bonada, N., Sánchez-Montoya, M.M., Gómez, R., Suárez, M.L., Vidal-Abarca, M.R., Demartini, D., Buffagni, A., Erba, S., Karaouzas, I., Skoulikidis, N., Prat, N., 2016. A biological tool to assess flow connectivity in reference temporary streams from the Mediterranean Basin. Sci. Total Environ. 540, 178–190. https://doi.org/10.1016/j.scitotenv.2015.06.086.

Cid, N., Bonada, N., Carlson, S.M., Grantham, T.E., Gasith, A., Resh, V.H., 2017. High variability is a defining component of Mediterranean-climate rivers and their biota. Water 9, 1–24. https://doi.org/10.3390/w9010052.

Cid, N., Bonada, N., Heino, J., Cañedo-Argüelles, M., Crabot, J., Sarremejane, R., Soininen, J., Stubbington, R., Datry, T., 2020. A metacommunity approach to improve biological assessments in highly dynamic freshwater ecosystems. Bioscience 70, 427–438. https://doi.org/10.1093/biosci/biaa033.

Collas, F.P., Koopman, K.R., Hendriks, A.J., van der Velde, G., Verbrugge, L.N., Leuven, R.S., 2014. Effects of desiccation on native and non-native molluscs in rivers. Freshw. Biol. 59, 41–55. https://doi.org/10.1111/fwb.12244.

Crabot, J., Polášek, M., Launay, B., Pařil, P., Datry, T., 2021. Drying in newly intermittent rivers leads to higher variability of invertebrate communities. Freshw. Biol. 66, 730–744. https://doi.org/10.1111/fwb.13673.

Datry, T., Larned, S.T., Scarsbrook, M.R., 2007. Responses of hyporheic invertebrate assemblages to large-scale variation in flow permanence and surface–subsurface exchange. Freshw. Biol. 52, 1452–1462. https://doi.org/10.1111/j.1365-2427.2007.01775.x.

Dallas, H.F., 2013. Ecological status assessment in Mediterranean rivers: complexities and challenges in developing tools for assessing ecological status and defining reference conditions. Hydrobiologia 719, 483–507. https://doi.org/10.1007/s10750-012-1305-8.

Datry, T., 2012. Benthic and hyporheic invertebrate assemblages along a flow intermittence gradient: effects of duration of dry events. Freshw. Biol. 57, 563–574. https://doi.org/10.1111/j.1365-2427.2011.02725.x.

Datry, T., Larned, S.T., Fritz, K.M., Bogan, M.T., Wood, P.J., Meyer, E.I., Santos, A.N., 2014. Broad-scale patterns of invertebrate richness and community composition in temporary rivers: effects of flow intermittence. Ecography 37, 94–104. https://doi.org/10.1111/j.1600-0587.2013.00287.x.

Datry, T., Pella, H., Leigh, C., Bonada, N., Hugueny, B., 2016a. A landscape approach to advance intermittent river ecology. Freshw. Biol. 61, 1200–1213. https://doi.org/10.1111/fwb.12645.

Datry, T., Bonada, N., Heino, J., 2016b. Towards understanding the organisation of metacommunities in highly dynamic ecological systems. Oikos 125, 149–159. https://doi.org/10.1111/oik.02922.

Datry, T., Singer, G., Sauquet, E., Jorda-Capdevila, D., von Schiller, D., Stubbington, R., Magand, C., Pařil, C., Miliša, M., Acuña, V., Alves, M.H., Brunke, M., Cid, N., Csabai, Z., England, J., Froebrich, J., Koundouri, P., Lamouroux, N., Martí, E., Morais, M., Munné, A., Mutz, M., Pesic, V., Previšić, A., Renaud, A., Robinson, C., Sadler, J., Skoulikidis, N., Terrier, B., Tockner, K., Vesely, D., Zoppini, A., 2017. Science and management of intermittent rivers and ephemeral streams (SMIRES) Res. Ideas Outcomes 3, e21774. https://doi.org/10.3897/rio.3.e21774.

Datry, T., Boulton, A.J., Bonada, N., Fritz, K., Leigh, C., Sauquet, E., Tockner, K., Hugueny, B., Dahm, C.N., 2018. Flow intermittence and ecosystem services in rivers of the Anthropocene. J. Appl. Ecol. 55, 353–364. https://doi.org/10.1111/1365-2664.12941.

Döll, P., Schmied, H.M., 2012. How is the impact of climate change on river flow regimes related to the impact on mean annual runoff? A global-scale analysis. Environ. Res. Lett. 7, 14037. https://doi.org/10.1088/1748-9326/7/1/014037.

Dufrêne, M., Legendre, P., 1997. Species assemblages and indicator species: the need for a flexible asymmetrical approach. Ecol. Monograph. 67, 345–366. https://doi.org/10.2307/2963459.

Fenoglio, S., Bo, T., Bosi, G., 2006. Deep interstitial habitat as a refuge for *Agabus paludosus* (Fabricius) (Coleoptera: Dytiscidae) during summer droughts. Coleopt. Bull. 60, 37–41. https://doi.org/10.1649/842.1.

Fišer, C., Keber, R., Kereži, V., Moškrič, A., Palandančić, A., Petkovska, V., Potočnik, H., Sket, B., 2007. Coexistence of species of two amphipod genera: *Niphargus timavi* (Niphargidae) and *Gammarus fossarum* (Gammaridae). J. Nat. Hist. 41, 2641–2651. https://doi.org/10.1080/00222930701661225.

Fortuño, P., Bonada, N., Prat, N., Acosta, R., Cañedo-Argüelles, M., Castro, D., Cid, N., Múrria, C., Pineda, D., Rocha, K., Sória, M., Tarrats, P., Verkaik, I., 2018. Efectes del Canvi Ambiental en les comunitats d'organismes dels Rius MEDiterranis (CARIMED). Informe 2017 Diputació de Barcelona. Àrea d'Espais Naturals (Estudis de la Qualitat Ecològica dels Rius; 27). https://doi.org/10.13140/RG.2.2.21483.39204.

Foster, G.N., 2010. A review of the scarce and threatened Coleoptera of Great Britain. Part 3: Water beetles of Great Britain. Species status no. 1. Joint Nature Conservation Committee, Peterborough, UK.

Hahn, H.J., 2006. The GW-Fauna-Index: A first approach to a quantitative ecological assessment of groundwater habitats. Limnologica 36, 119–137. https://doi.org/10.1016/j.limno.2006.02.001.

Gallart, F., Cid, N., Latron, J., Llorens, P., Bonada, N., Jeuffroy, J., Jiménez-Argudo, S.-M., Vega, R.-M., Solà, C., Soria, M., Bardina, M., Hernández-Casahuga, A.-J., Fidalgo, A., Estrela, T., Munné, A., Prat, N., 2017. TREHS: An open-access software tool for investigating and evaluating temporary river regimes as a first step for their ecological status assessment. Sci. Total Environ. 607–608, 519–540. https://doi.org/10.1016/j.scitotenv.2017.06.209.

Hammock, B.G., Bogan, M.T., 2014. Black fly larvae facilitate community recovery in a mountain stream. Freshw. Biol. 59, 2162–2171. https://doi.org/10.1111/fwb.12419.

Hering, D., Johnson, R.K., Kramm, S., Schmutz, S., Szoszkiewicz, K., Verdonschot, P.F.M., 2006. Assessment of European rivers with diatoms, macrophytes, invertebrates and fish: a comparative metric-based analysis of organism response to stress. Freshw. Biol. 51, 1757–1785. https://doi.org/10.1111/j.1365-2427.2006.01610.x.

Hill, M.J., Milner, V.S., 2018. Ponding in intermittent streams: A refuge for lotic taxa and a habitat for newly colonising taxa? Sci. Total Environ. 628, 1308–1316. https://doi.org/10.1016/j.scitotenv.2018.02.162.

Hughes, S.J., Santos, J.M., Ferreira, M.T., Caraca, R., Mendes, A.M., 2009. Ecological assessment of an intermittent Mediterranean river using community structure and function: evaluating the role of different organism groups. Freshw. Biol. 54, 2383–2400. https://doi.org/10.1111/j.1365-2427.2009.02253.x.

Jain, A.K., Dubes, R.C., 1988. Algorithms for clustering data. Prentice-Hall, New Jersey.

Kohonen, T., 1982. Self-organised formation of topographically correct feature maps. Biol. Cybern. 43, 59-69. https://doi.org/10.1007/BF00337288.

Kohonen, T., Schroeder, M.R., Huang, T.S., 2001. Self-Organizing Maps, third ed. Springer-Verlag, Berlin, Heidelberg.

Larned, S.T., Datry, T., Robinson, C.T., 2007. Invertebrate and microbial responses to inundation in an ephemeral river reach in New Zealand: effects of preceding dry periods. Aq. Sci. 69, 554–567. https://doi.org/10.1007/s00027-007-0930-1.

Larned, S.T., Datry, T., Arscott, D.B., Tockner, K., 2010. Emerging concepts in temporary-river ecology. Freshw. Biol. 55, 717–738. https://doi.org/10.1111/j.1365-2427.2009.02322.x.

Larned, S.T., Schmidt, J., Datry, T., Konrad, C.P., Dumas, J.K., Diettrich, J.C., 2011. Longitudinal river ecohydrology: flow variation down the lengths of alluvial rivers. Ecohydrology 4, 532–548. https://doi.org/10.1002/eco.126.

Leigh, C., Boulton, A.J., Courtwright, J.L., Fritz, K., May, C.L., Walker, R.H., Datry, T., 2016. Ecological research and management of intermittent rivers: an historical review and future directions. Freshw. Biol. 61, 1181–1199. https://doi.org/10.1111/fwb.12646.

Leigh, C., Datry, T., 2017. Drying as a primary hydrological determinant of biodiversity in river systems: A broad-scale analysis. Ecography 40, 487–499. https://doi.org/10.1111/ecog.02230.

Lopez-Rodriguez, M.J., Tierno de Figueroa, J.M., Alba-Tercedor, J., 2009. Life history of two burrowing aquatic insects in southern Europe: *Leuctra geniculata* (Insecta: Plecoptera) and *Ephemera danica* (Insecta: Ephemeroptera). Aquat. Insect. 31, 99–110. https://doi.org/10.1080/01650420802620345.

Macadam, C.R., 2016. A review of the mayflies (Ephemeroptera) of Great Britain. Species status no. 28. Natural England Commissioned Report NECR193. Natural England, Peterborough, UK.

Maddock, I.P., Petts, G.E., Bickerton, M.A., 1995. River channel assessment: a method for defining channel sectors: the River Glen, Lincolnshire UK, in: Petts, G.E. (Ed.), Man's Influence on Freshwater Ecosystems and Water Use (Proceedings of a Boulder Symposium, July 1995). International Association of Hydrological Sciences, Wallingford, UK, pp. 219–226.

Mazor, R.D., Stein, E.D., Ode, P.R., Schiff, K., 2014. Integrating intermittent streams into watershed assessments: applicability of an index of biotic integrity. Freshw. Sci. 33, 459–474. https://doi.org/10.1086/675683.

Milošević, D., Simić, V., Stojković, M., Čerba, D., Mančev, D., Petrović, A., Paunović, M., 2013. Spatio-temporal pattern of the Chironomidae community: toward the use of non-biting midges in bioassessment programs. Aquatic Ecol. 47, 37–55. https://doi.org/10.1007/s10452-012-9423-y.

Milošević, D., Čerba, D., Szekeres, J., Csányi, B., Tubić, B., Simić, V., Paunović, M., 2015. Artificial neural networks as an indicator search engine: The visualization of natural and man-caused taxa variability. Ecol. Ind. 61, 777–789. https://doi.org/10.1016/j.ecolind.2015.10.029.

Milošević, D., Mančev, D., Čerba, D., Piperac, M.S., Popović, N., Atanacković, A., Đuknić, J., Simić, V., Paunović, M., 2018. The potential of chironomid larvae-based metrics in the bioassessment of non-wadeable rivers. Sci. Total Environ. 616, 472–479. https://doi.org/10.1016/j.scitotenv.2017.10.262.

Munné, A., Prat, N., 2009. Use of macroinvertebrate-based multimetric indices for water quality evaluation in Spanish Mediterranean rivers: an intercalibration approach with the IBMWP index. Hydrobiologia 628, 203–225. https://doi.org/10.1007/s10750-009-9757-1.

Munné, A., Prat, N., 2011. Effects of Mediterranean climate annual variability on stream biological quality assessment using macroinvertebrate communities. Ecol. Ind. 11, 651–662. https://doi.org/10.1016/j.ecolind.2010.09.004.

Munné, A., Bonada, N., Cid, N., Gallart, F., Solà, C., Bardina, M., Rovira, A., Sierra, C., Soria, M., Fortuño, P., Llorens, P., Latron, J., Estrela, T., Fidalgo, A., Serrano, I., Jiménez, S., Vega, R., Prat, N., 2021. A proposal to classify and assess ecological status in mediterranean temporary rivers: research insights to solve management needs. Water 13, 767. https://doi.org/10.3390/w13060767.

Pace, G., Bonada, N., Prat, N., 2013. Long-term effects of climatic–hydrological drivers on macroinvertebrate richness and composition in two Mediterranean streams. Freshw. Biol. 58, 1313–1328. https://doi.org/10.1111/fwb.12129.

Paisley, M.F., Trigg, D.J., Walley, W.J., 2014. Revision of the Biological Monitoring Working Party (BMWP) score system: derivation of present-only and abundance-related scores from field data. River Res. Appl. 30, 887–904. https://doi.org/10.1002/rra.2686.

Papáček, M., 2001. Small aquatic and ripicolous bugs (Heteroptera: Nepomorpha) as predators and prey: The question of economic importance. Eur. J. Entomol. 98, 1–12. https://doi.org/10.14411/eje.2001.001.

Park, Y.S., Céréghino, R., Compin, A., Lek, S., 2003. Applications of artificial neural networks for patterning and predicting aquatic insect species richness in running waters. Ecol. Model. 160, 265–280. https://doi.org/10.1016/S0304-3800(02)00258-2.

Park, Y.S., Chon, T.S., Kwak, I.S., Lek, S., 2004. Hierarchical community classification and assessment of aquatic ecosystems using artificial neural networks. Sci. Total Environ. 327, 105–122. https://doi.org/10.1016/j.scitotenv.2004.01.014.

Prat, N., Gallart, F., von Schiller, D., Polesello, S., Garcia-Roger, E.M., Latron, J., De Girolamo, A.M., 2014. The MIRAGE toolbox: an integrated assessment tool for temporary streams. River Res. Appl. 30, 1318–1334. https://doi.org/10.1002/rra.2757.

Raymond, P.A., Hartmann, J., Lauerwald, R., Sobek, S., McDonald, C., Hoover, M., Butman, D., Striegl, R., Mayorga, E., Humborg, C., Kortelainen, P., 2013. Global carbon dioxide emissions from inland waters. Nature 503, 355–359. https://doi.org/10.1038/nature12760.

Samraoui, B., 2009. Seasonal ecology of Algerian Lestidae (Odonata). Int. J. Odonatol. 12, 383–394. https://doi.org/10.1080/13887890.2009.9748352.

Sabater S., Tockner K., 2009. Effects of hydrologic alterations on the ecological quality of river ecosystems, in: Sabater S., Barceló D. (Eds.) Water Scarcity in the Mediterranean. The Handbook of Environmental Chemistry, vol 8. Springer, Berlin, Heidelberg. https://doi.org/10.1007/698_2009_24.

Sarremejane, R., England, J., Sefton, C.E., Parry, S., Eastman, M., Stubbington, R., 2020. Local and regional drivers influence how aquatic community diversity, resistance and resilience vary in response to drying. Oikos 129, 1877–1890. https://doi.org/10.1111/oik.07645.

Sear, D.A., Armitage, P.D., Dawson, F.H., 1999. Groundwater dominated rivers. Hydrol. Proc. 13, 255–276. https://doi.org/10.1002/(SICI)1099-1085(19990228)13:3%3C255::AID-HYP737%3E3.0.CO;2-Y.

Smith, H., Wood, P.J., Gunn, J., 2003. The influence of habitat structure and flow permanence on invertebrate communities in karst spring systems. Hydrobiologia 510, 53–66. https://doi.org/10.1023/B:HYDR.0000008501.55798.20.

Soria, M., Leigh, C., Datry, T., Bini, L.M., Bonada, N., 2017. Biodiversity in perennial and intermittent rivers: A meta-analysis. Oikos 126, 1078–1089. https://doi.org/10.1111/oik.04118.

Steward, A.L., Negus, P., Marshall, J.C., Clifford, S.E., Dent, C., 2018. Assessing the ecological health of rivers when they are dry. Ecol. Ind. 85, 537–547. https://doi.org/10.1016/j.ecolind.2017.10.053.

Straka, M., Polášek, M., Syrovátka, V., Stubbington, R., Zahrádková, S., Němejcová, D., Šikulová, L., Řezníčková, P., Opatřilová, L., Datry, T., Pařil, P., 2019. Recognition of stream drying based on benthic macroinvertebrates: A new tool in Central Europe. Ecol. Ind. 106, 105486. https://doi.org/10.1016/j.ecolind.2019.105486.

Straka, M., Polášek, M., Csabai, Z., Zweidick, O., Graf, W., Meyer, E.I., Elexová, E.M., Lešťáková, M., Pařil, P., 2021. Stream drying bioindication in Central Europe: A Biodrought Index accuracy assessment. Ecol. Ind. 130, 108045. https://doi.org/10.1016/j.ecolind.2021.108045.

Stubbington, R., Greenwood, A.M., Wood, P.J., Armitage, P.D., Gunn, J., Robertson, A.L., 2009. The response of perennial and temporary headwater stream invertebrate communities to hydrological extremes. Hydrobiologia 630, 299–312. https://doi.org/10.1007/s10750-009-9823-8.

Stubbington, R., Wood, P.J., Reid, I., Gunn, J., 2011a. Benthic and hyporheic invertebrate community responses to seasonal flow recession in a groundwater-dominated stream. Ecohydrology 4, 500–511. https://doi.org/10.1002/eco.168.

Stubbington, R., Wood, P.J., Reid, I., 2011b. Spatial variability in the hyporheic zone refugium of temporary streams. Aquat. Sci. 73, 499–511. https://doi.10.1007/s00027-011-0203-x.

Stubbington, R., 2012. The hyporheic zone as an invertebrate refuge: a review of variability in space, time, taxa and behaviour. Mar. Freshw. Res. 63, 293–311. https://doi.org/10.1071/MF11196.

Stubbington, R., Datry, T., 2013. The macroinvertebrate seedbank promotes community persistence in temporary rivers across climate zones. Freshw Biol, 58, 1202-1220. https://doi.org/10.1111/fwb.12121.

Stubbington, R., Gunn, J., Little, S., Worrall, T.P., Wood, P.J., 2016. Macroinvertebrate seedbank composition in relation to antecedent duration of drying and multiple wet-dry cycles in a temporary stream. Freshw. Biol. 61, 1293–1307. https://doi.org/10.1111/fwb.12770.

Stubbington, R., England, J., Wood, P.J., Sefton, C.E., 2017. Temporary streams in temperate zones: recognizing, monitoring and restoring transitional aquatic-terrestrial ecosystems. Wiley Interdiscip. Rev. Water 4, e1223. https://doi.org/10.1002/wat2.1223.

Stubbington, R., Chadd, R., Cid, N., Csabai, Z., Miliša, M., Morais, M., Munné, A., Pařil, P., Pešić, V., Tziortzis, I., Verdonschot, R.C.M., Datry, T., 2018. Biomonitoring of intermittent rivers and ephemeral streams in Europe: current practice and priorities to enhance ecological status assessments. Sci. Total Environ. 618, 1096–1113. https://doi.org/10.1016/j.scitotenv.2017.09.137.

Stubbington, R., Paillex, A., England, J., Barthès, A., Bouchez, A., Rimet, F., Mar Sánchez-Montoya, M., Westwood, C.G., Datry, T., 2019. A comparison of biotic groups as dry-phase indicators of ecological quality in intermittent rivers and ephemeral streams. Ecol. Ind. 97, 165-174. https://doi.org/10.1016/j.ecolind.2018.09.061.

Stubbington, R., Acreman M., Acuña, V., Boon, P. J., Boulton, A. J., England, J., Gilvear, D., Sykes, T., Wood, P. J., 2020. Ecosystem services of dry temporary streams differ among regions with contrasting climates and economies. People and Nature 2, 660–677. https://doi.org/10.1002/pan3.10113.

Theodoropoulos, C., Karaouzas, I., Stubbington, R., 2021. Biotic indices of hydrological variability as tools to inform dynamic ecological status assessments in river ecosystems. J. Environ. Manage. 295, 113124. https://doi.org/10.1016/j.jenvman.2021.113124.

Townsend, C.R., Hildrew, A.G., 1976. Field experiments on the drifting, colonization and continuous redistribution of stream benthos. J. Anim. Ecol. 45, 759–772. https://doi.org/10.2307/3579.

Tramblay, Y., Rutkowska, A., Sauquet, E., Sefton, C., Laaha, G., Osuch, M., Albuquerque, T., Alves, M.H., Banasik, K., Beaufort, A., Brocca, L., 2021. Trends in flow intermittence for European rivers. Hydrol. Sci. J. 66, 37–49. https://doi.org/10.1080/02626667.2020.1849708.

Valente-Neto, F., da Silva, F.H., Covich, A.P., de Oliveira Roque, F., 2020. Streams dry and ecological uniqueness rise: environmental selection drives aquatic insect patterns in a stream network prone to intermittence. Hydrobiologia, 847, 617–628. https://doi.org/10.1007/s10750-019-04125-9.

Vander Vorste, R., Malard, F., Datry, T., 2016. Is drift the primary process promoting the resilience of river invertebrate communities? A manipulative field experiment in an intermittent alluvial river. Freshw. Biol. 61, 1276–1292. https://doi.org/10.1111/fwb.12658.

Vesanto, J., Himberg, J., Alhoniemi, E., Parhankangas, J., 2000. SOM toolbox for Matlab 5. Helsinki University of Technology, Neural Networks Research Centre, Espoo, Finland.

Wäldchen, J, Mäder, P., 2018. Machine learning for image based species identification. Methods Ecol. Evol. 9, 2216–2225. https://doi.org/10.1111/2041-210X.13075.

Wallace, I., 2016. A review of the status of the caddisflies (Trichoptera) of Great Britain. Species status no. 27. Natural England Commissioned Report NECR191. Natural England, Peterborough, UK.

Watson, M., Dallas, H.F., 2013. Bioassessment in ephemeral rivers: constraints and challenges in applying macroinvertebrate sampling protocols. Afr. J. Aquat. Sci. 38, 35–51. https://doi.org/10.2989/16085914.2012.742419.

White, J.C., House, A., Punchard, N., Hannah, D.M., Wilding, N.A., Wood, P.J., 2018. Macroinvertebrate community responses to hydrological controls and groundwater abstraction effects across intermittent and perennial headwater streams. Sci. Total Environ. 610–611, 1514–1526. https://doi.org/10.1016/j.scitotenv.2017.06.081.

Young, A., 2006. Stream flow simulation within UK ungauged catchments using a daily rainfall-runoff model. J. Hydrol. 320, 155–172. https://doi.org/10.1016/j.jhydrol.2005.07.017.

Zipper, S.C., Hammond, J.C., Shanafield, M., Zimmer, M., Datry, T., Jones, C.N., Kaiser, K.E., Godsey, S.E., Burrows, R.M., Blaszczak, J.R., Busch, M.H., 2021., Pervasive changes in stream intermittency across the United States. Environ. Res. Lett. 16, 084033. https://doi.org/10.1088/1748-9326/ac14ec.

Table 1. Characteristics of rivers studied in regions with continental (France), Mediterranean (Catalonia, NE Spain) and oceanic (UK) climates.

Region	**River**	**Catchment area (km^2)**	**Altitude (m)**	**Sampling months**	**Sampling years**	**Number of sites**		**Samples per site**	**Sampling method**
						perennial	**intermittent**		
Continental	Albarine	311	653	Apr–Nov	2008–10	7	11	4	Hess
Mediterranean	Llobregat	4923	1259	Apr–Aug	2012–16	1	1	1	Kick
	Ter	3275	2400	Apr–Aug	2012–16	3	0	1	Kick
	Besòs	1038	72	Apr–Aug	2012–16	2	6	1	Kick
	Tordera	898	1500	Apr–Aug	2012–16	2	1	1	Kick
	Pineda	6	10	Apr–Aug	2012–16	0	1	1	Kick
Oceanic	Glen	349	80	May–Sept	2009	1	3	4	Surber
	Lathkill	54	200	May–Sept	2009	2	3	4	Surber

Table 2. Mean ± standard error % flow in each SOM group, in the continental, mediterranean and oceanic models. Superscript letters indicate statistically comparable groups; other values differ (non-parametric ANOVA [Kruskal-Wallis] tests with pairwise Mann-Whitney post-hoc tests, $p<0.016$)

SOM groups	**Continental**	**Mediterranean**	**Oceanic**
I	69.65 ± 4.71^{a}	90.37 ± 1.54^{b}	92.63 ± 1.28^{a}
II	60.32 ± 4.71^{a}	64.03 ± 1.77	82.05 ± 0.98
III	99.91 ± 0.08	95.54 ± 1.59^{a}	99.74 ± 0/25^{a}
IV	93.79 ± 2.42	94.47 ± 2.61^{ab}	94.58 ± 1.01^{a}

Table 3. Taxa identified by indicator species analysis (IndVal) as indicative of each SOM group in three regional datasets: a) continental, b) mediterranean and c) oceanic. Taxa are sorted by SOM groups (Gr, I–IV) and mean annual % flow (%f) intermittence and then by indicator value within the group (InV, as a %); asterisks indicate significance of: * p <0.05, ** p <0.01, *** p <0.001.

a)

Taxa	**Gr/%f**	**InV**	**p**	**Taxa**	**Gr/%f**	**InV**	**P**
Simuliidae	I/69.7	86.8	***	*Elmis*	IV/93.8	62.3	***
Ceratopogonidae		63.1	***	*Leuctra*		56.9	***
Chironomidae		38.2	**	*Odontocerum*		55.5	***
Niphargus	II/60.3	29.1	**	*Esolus*		54.1	***
Gammarus	III/99.9	59.8	***	Limoniidae		53.3	***
Rhithrogena		53.0	***	*Limnius*		53.1	***
Ephemerella		50.9	**	*Ampullaceana balthica*		49.6	***
Riolus		40.3	**	Dugesiidae		48.8	***
Hydropsyche	IV/93.8	89.2	***	*Ephemera*		48.2	***
Ecdyonurus		83.8	***	Hydrachnidia		47.9	***
Habroleptoïdes		83.7	***	Athericidae		44.5	***
Torleya		76.7	***	*Caenis*		44.3	*
Sericostoma		67.2	***	*Epeorus*		33.9	**
Rhyacophila		66.5	***	*Nemoura*		31.5	**

b)

Taxa	**Gr/%f**	**InV**	**p**	**Taxa**	**Gr/%f**	**InV**	**P**
Caenidae	I/90.4	81.7	***	Sericostomatidae	III/95.5	50.4	***
Hydrophilidae		71.7	***	Limoniidae		46.1	***
Hydroptilidae		70.4	***	Planariidae		42.6	***
Gerridae		48.0	***	Dixidae		36.4	**
Physidae		45.6	***	Rhyacophilidae		34.6	***
Simuliidae		42.8	**	Thaumaleidae		32.3	***
Anthomyiidae		42.5	***	Athericidae		31.7	*
Baetidae		41.8	**	Philopotamidae		27.6	*
Chironomidae		39.3	***	Glossosomatidae		25.6	**
Leptoceridae		37.6	**	Gammaridae	IV/94.5	95.7	***
Corixidae		36.1	***	Hydrobiidae		82.9	***
Dytiscidae	II/64.0	71.9	***	Calopterygidae		48.3	***
Lestidae		49.9	***	Scirtidae		46.7	***
Notonectidae		33.2	***	Heptageniidae		45.9	***
Sphaeriidae		25.9	***	Hydrochidae		45.2	**
Libellulidae		25.6	***	Elmidae		42.1	*
Leuctridae	III/95.5	85.0	***	Erpobdellidae		34.6	**
Nemouridae		82.4	***	Planorbidae		33.7	*
Chloroperlidae		68.1	***	Odontoceridae		30.1	*
Lepidostomatidae		55.8	***	Ephemeridae		26.5	**

c)

Taxa	**Gr/FI**	**InV**	**p**	**Taxa**	**Gr/FI**	**InVp**	**p**
Riolus	I/92.6	78.3	***	Oligochaeta	III/99.7	53.2	***
Polycelis felina		66.1	***	*Piscicola geometra*		47.9	***
Gammarus pulex		55.1	***	*Glossiphonia complanata*		40.9	***
Elmis aenea		50.2	***	*Oulimnius*		40.7	***
Dinocras cephalotes		49.7	***	*Sericostoma personatum*		39.8	***
Isoperla grammatica		45.5	***	*Asellus aquaticus*		38.5	***
Agapetus fuscipes		36.9	***	*Hydropsyche angustipennis*		36.9	***
Riolus subviolaceus		35.6	***	*Dicranota*		36.1	***
Silo nigricornis		29.8	***	*Potamopyrgus antipodarum*		36.1	***
Drusus annulatus		29.1	***	*Planorbis vortex*		36	***
Crenobia alpina		27.7	***	Ceratopogonidae		33.9	**
Rhyacophila dorsalis		26.8	***	Chironomidae		33.1	*
Serratella ignita		26	*	*Caenis luctuosa* group		32	***
Leuctra	II/82.1	60.7	***	*Helobdella stagnalis*		31.7	***
Chaetopteryx villosa		37.9	***	*Hydropsyche siltalai*		31.2	***
Leuctra geniculata		30.8	***	*Athripsodes bilineatus*		31.2	***
Drusus annulatus		27.6	**	*Goera pilosa*		29.5	***
Ampullaceana balthica		25.1	***	*Ancylus fluviatilis*		25.2	**
Simuliidae	III/99.7	88.8	***	*Lepidostoma hirtum*		25.0	***
Hydroptila		83.1	***	*Bithynia leachii*	IV/94.5	46.3	***
Sphaeriidae		81.6	***	*Polycelis tenuis/nigra*		30.0	***
Baetis		75.8	***	Haliplidae		25.8	***
Erpobdella octoculata		60.7	***				

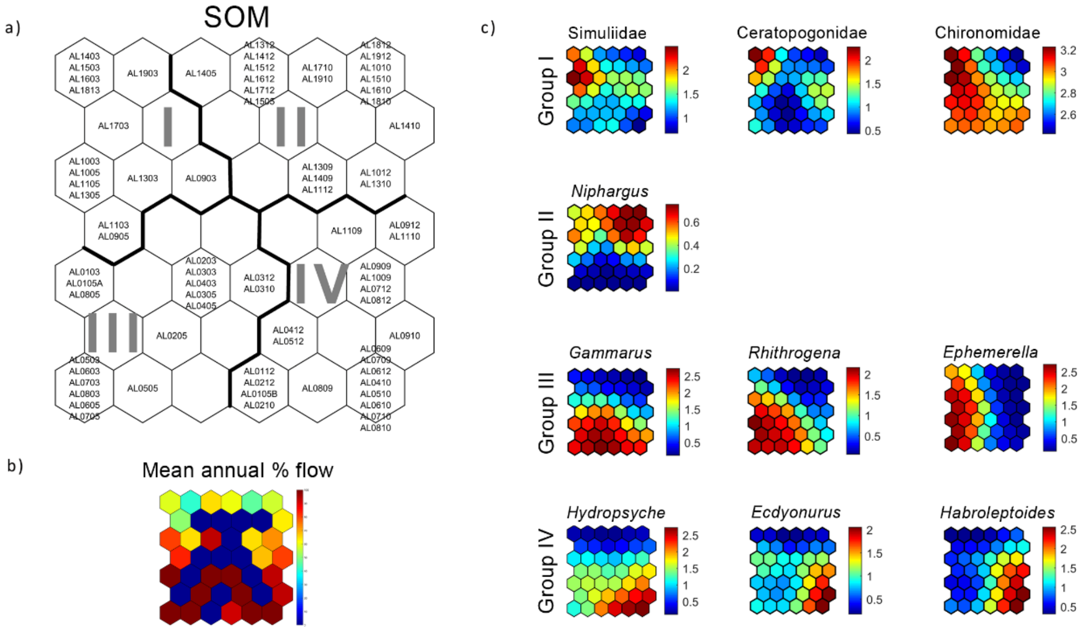

Fig. 1. The self-organizing map (SOM) of macroinvertebrate communities in different samples (a), the distribution of mean annual % flow (b) and abundance of indicator taxa with the three highest IndVal values (c) over the trained neural network for the continental region. The labels assigned to each SOM neuron indicate different samples and I–IV indicate groups of neurons. Color gradients indicate variability in the % flow and the abundance of indicator taxa. Neurons without assigned samples (empty neurons) for FI are colored in blue.

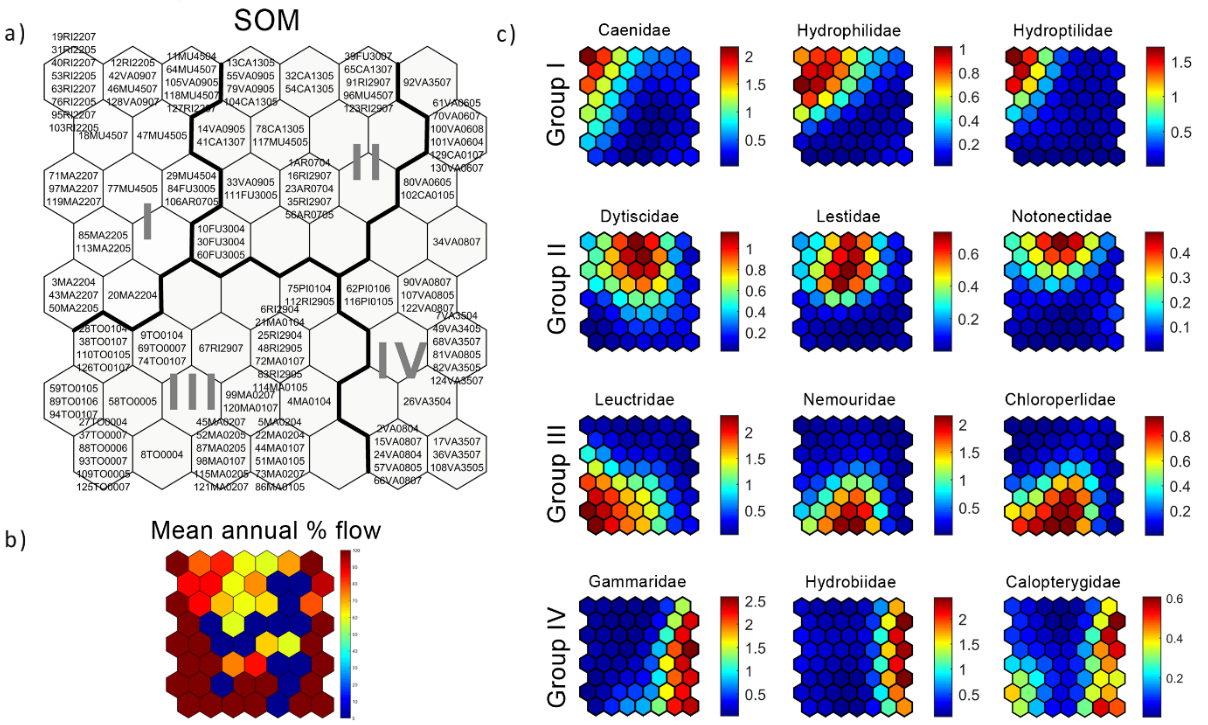

Fig. 2. The self-organizing map (SOM) of the macroinvertebrate community in different samples (a), the distribution of mean annual % flow (b) and abundance of indicator taxa with the highest IndVal values (c) over the trained neural network for the mediterranean region. See Fig. 1 for further details.

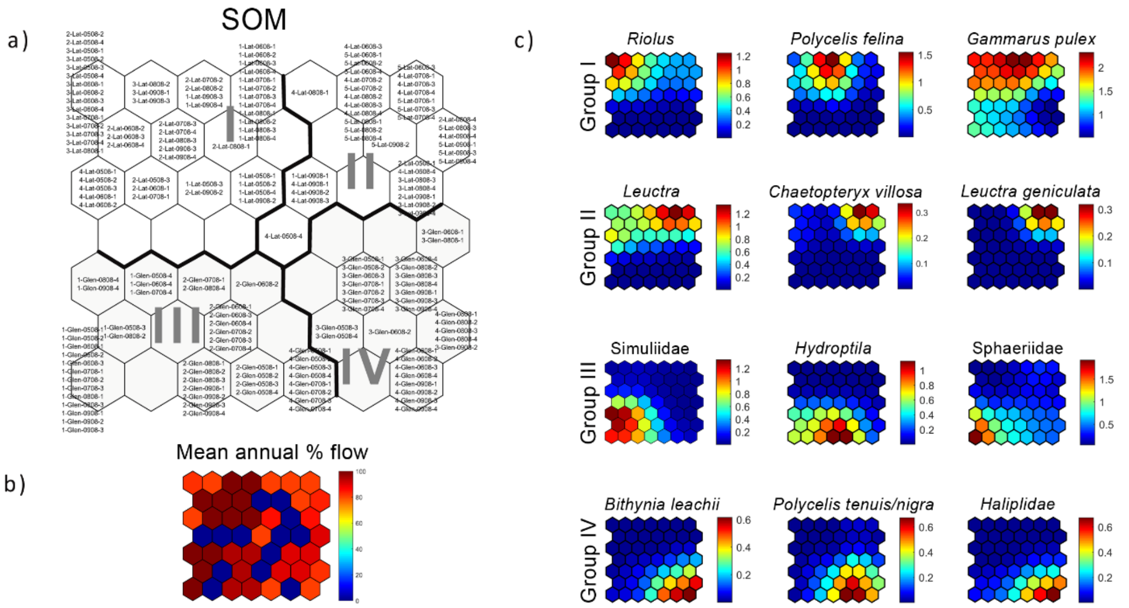

Fig. 3. The self-organizing map (SOM) of the macroinvertebrate community in different samples (a), the distribution of mean annual % flow (b) and abundance of indicator taxa with the highest IndVal values (c) over the trained neural network for the oceanic region. See Fig. 1 for further details.