

Long-term effects of climatic–hydrological drivers on macroinvertebrate richness and composition in two Mediterranean streams

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SUMMARY

1. The unique information provided by multiyear data sets can aid in detecting major long-term trends and making predictions with respect to environmental and biological variables. These trends are expected to differ among climatic regions but little information exists concerning long-term variability within a particular climate region.
2. We analysed the long-term (14-year time span) patterns in species richness and composition of aquatic insects (EPT: Ephemeroptera, Plecoptera and Trichoptera) in relation to changes in temperature, precipitation and discharge in two reference streams in Catalonia (north-east Spain). Our study sites were located in the Mediterranean macroclimate region but in different mesoclimatic areas, one in the Wet Mediterranean climate area and the other in the Dry Mediterranean climate area (WM and DM, respectively).
3. At the macroclimate scale, precipitation decreased and the frequency of drought events increased during the 14-year period. Although the mean number of taxa in each of the two sites remained similar, there was a replacement of species characteristic of riffle-like conditions by species characteristic of pool-like conditions over time in both sites.
4. EPT composition differed between sites at the mesoclimate scale, demonstrating the importance of regional factors. In particular, changes in EPT composition were less variable in WM than in DM both in the short term and in the long term, indicating that the WM site was less constrained by climate than the DM site. Such differences between mesoclimates are related to differences in flood magnitude (higher in WM) and drought frequency (more frequent in DM) as well as local hydromorphological differences.
5. Our findings indicate that the streams responded similarly to long-term patterns in large-scale variables (i.e. those affecting the overall macroclimatic region), but differed with respect to the effect of local-scale variables (i.e. those differentially affecting each mesoclimatic area). Predictive bio-assessment models that use reference conditions must incorporate long-term variability to accurately assess the ecological status of Mediterranean climate streams.

Keywords: droughts, floods, mesoclimates, reference conditions, spatial scale

Introduction

Long-term variability and trends in the world's climate, especially global warming, have been identified as serious threats to the biosphere (Rosenzweig *et al.*, 2008). Global surface temperatures have increased by 0.6 °C during the 20th century, and climate models predict a

rise of 1.4–5.8 °C in the next 100 years, depending primarily on the quantity of carbon dioxide emissions from anthropogenic sources (IPCC, 2001). The consequences of global warming are reflected at global and regional scales in terms of changes in key climatic variables such as precipitation and atmospheric moisture, snow cover, extent of land and sea ice, sea level and atmospheric

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and oceanic circulation (Palmer & Raisanen, 2002; Milly, Dunne & Vecchia, 2005). Models forecast that water stress may become particularly acute in the south-west USA, Mexico, the Mediterranean Basin and the Middle East, where rainfall may decrease from 10 to 25% regionally and up to 40% locally (Shindell, 2007). In addition, global change has been shown to significantly affect biodiversity at global, regional and local scales (Heino, 2009; Heino, Virkkala & Toivonen, 2009), and future predictions are not optimistic (Sala *et al.*, 2000). This problem is particularly acute for freshwater ecosystems with increasing evidence suggesting that streams and rivers will be among the most sensitive of ecosystems to the effects of global change (Ormerod & Durance, 2009).

Multiyear data sets (e.g. with more than 10 years) provide vital information concerning long-term variability in ecosystem characteristics and dynamics. In addition, such data sets allow us to detect major trends and to make future predictions with respect to the environmental and biological variables (Legendre & Legendre, 1998) relevant to investigations of global change. However, the period of observation must be sufficiently long to allow the determination of statistical significance of findings and to support predictions (Müller *et al.*, 2010). Only a small number of freshwater studies comprise more than a few years of data (i.e. >10 years; Jackson & Füreder, 2006). These have mainly focused on the effects of long-term fluctuations in hydrology, temperature, extreme events or climatic cycles (e.g. hydrology: Daufresne, Lengfellner & Sommer, 2009; drought: Mouthon & Daufresne, 2006; freezing: Mulholland *et al.*, 2009; NAO: Bradley & Ormerod, 2001; or ENSO: Bêche & Resh, 2007b). Such studies have demonstrated that long-term trends in these environmental variables induce changes in life history characteristics and community structure (Harper & Peckarsky, 2006; Burgmer, Hillebrand & Pfenninger, 2007; Durance & Ormerod, 2007; Richter *et al.*, 2008). For example, studies of the mayfly *Baetis bicaudatus* in high-altitude streams of western Colorado (USA) have shown that the amount of run-off from accumulated winter precipitation is correlated with the timing of metamorphosis to the adult stage, with mayflies emerging earlier in drought years and later in wetter years (Peckarsky, Taylor & Caudill, 2000). Furthermore, studies in central France confirmed that climatic warming, particularly the effects of the heat wave of 2003, affected Mollusca community structure, with a rapid decrease in densities and species richness (Mouthon & Daufresne, 2006).

Most long-term research in streams has focused on temperate climate ecosystems, particularly those of central Europe (Rosenzweig *et al.*, 2008). However, long-term changes in temperate streams might not be representative of other climate regions in Europe where precipitation and temperature patterns vary widely among years, such as the Mediterranean region (Gasith & Resh, 1999). For example, changes in the North Atlantic Oscillation index (NAO index) lead to changes in temperature, precipitation and run-off across Europe (Arnell, 1999), with patterns in the northern temperate regions being positively correlated with the NAO index and those in the southern Mediterranean regions exhibiting a negative correlation (Shorthouse & Arnell, 1997). In addition, Mediterranean rivers are naturally subject to stress, with high seasonal variability in flow and interruption of superficial water for several months during the dry period. Apart from this seasonal variability, the few existing long-term studies of these systems have also revealed strong interannual variability that has significant consequences for stream biota (e.g. in California: Bêche *et al.*, 2006; Bêche & Resh, 2007a,b; Lawrence *et al.*, 2010).

In addition to the variability observed among macroclimatic regions (i.e. temperate and Mediterranean), the climate within a region can be modified by orography, altitude, orientation relative to coastline and continental and oceanic influences (Aschmann, 1973; Conacher & Sala, 1998). These local-scale variables can produce particular mesoclimatic and microclimatic patterns (Nahal, 1981; Daget, Ahdali & David, 1988). For example, it is generally accepted that altitude is the main factor governing spatial variability in precipitation and temperature, with drier and warmer mesoclimates at low altitudes than at high altitudes (Sevruk, 1997). However, less information exists concerning long-term variability within a particular climatic region, and long-term trends may not be uniform in highly heterogeneous regions. The need to analyse long-term changes at different spatial scales has been of great interest in bioclimatic modelling (Wiens, 2002; Araújo & Guisan, 2006). As different processes dominate at different scales (Heuvelink, 1998), and because spatial scale affects model projections (Trivedi *et al.*, 2008), large-scale models should be compared with local-scale models to improve prediction accuracy. However, although local-scale models may be more appropriate for projecting climate change, they have the disadvantage of requiring fine-resolution climate and species data, which are not always easily obtained.

The main objectives of this study were (i) to describe the long-term changes (i.e. over a 14-year

time span) in the taxonomic composition of aquatic macroinvertebrates in two undisturbed headwater sites located in a common macroclimatic region (the Mediterranean climate region) but in different mesoclimatic areas and (ii) to relate these biological changes to long-term climatic and hydrological variability. The two mesoclimatic areas were the Wet Mediterranean climate area and the Dry Mediterranean climate area: WM and DM, respectively. Our general hypothesis was that the two streams should respond similarly with respect to long-term patterns of large-scale variables (i.e. those affecting the overall macroclimatic region) but differently with respect to local-scale variables (i.e. those differentially affecting each mesoclimatic area). Thus, for example, the observed decrease in precipitation in Southern Europe (IPCC, 2001; Shindell, 2007) would result in a decrease in river flow regimes and an increase in low pulse duration with time in both streams. Consequently, the number of taxa adapted to such high-stress conditions would

increase in both mesoclimatic areas over time. However, given that the two sites differed with respect to local-scale variables, such as mesoclimate variables (e.g. small differences in temperature or rainfall related to differences in altitude), long-term biological changes were likely to differ. In particular, we hypothesised that changes in community structure in WM will be less variable than those in DM, where seasonal and interannual variability is expected to be higher.

Methods

Study area

The two study sites were located in the Llobregat River catchment in NE Spain (Table 1). The WM site (Castellar de n'Hug: 42°15'31.65"N; 1°58'34.19"E) was located at higher altitude (1360 m.a.s.l.) in the headwaters of the Llobregat River. The DM site (Santa Maria de Merlès:

Table 1 Stream type and geographical, hydromorphological and physicochemical characteristics of the studied streams

Sub-basin Site	Stream sites	
	Llobregat Castellar de n'Hug	Merles Santa Maria de Merlès
Stream type characterisation		
Hydrology	Perennial	Perennial
Geology	Calcareous (Karstic)	Calcareous (Karstic)
Ecoregion (Illies, 1978)	Mediterranean	Mediterranean
River type (Med-GIG, WFD 2000/60/CE)	RM-4	RM-4
Subtypes of river management (Munné & Prat, 2004)	Calcareous Wet mountain rivers (1b)	Calcareous Mediterranean mountain rivers (2b)
Microclimate	Wet Mediterranean	Dry Mediterranean
Code	WM	DM
Geographical and hydromorphological features		
Latitude	42°15'31.65"N	42°1'25.31"N
Longitude	1°58'34.19"E	1°59'38.31"E
Altitude (m.a.s.l.)	1360	550
Distance from source (km)	3.5	25.4
Slope of the waterbody	16	1.45
Drainage basin (km ²)	9.62	107.08
Average channel width (m)	8.75	5.54
Average max depth (m)	0.44	0.33
Physicochemical characteristics		
P-PO ₄ ³⁻ (mg L ⁻¹) (mean ± SD)	0.03 ± 0.08	0.02 ± 0.02
N-NO ₃ ⁻ (mg L ⁻¹) (mean ± SD)	0.62 ± 0.46	0.42 ± 0.86
N-NO ₂ ⁻ (mg L ⁻¹) (mean ± SD)	0.02 ± 0.07	0.02 ± 0.06
Conductivity 25 °C (µS cm ⁻²) (mean ± SD)	256.9 ± 51.48	480.51 ± 98.31
pH (mean ± SD)	8.30 ± 0.32	8.27 ± 0.35
D.O. (mg L ⁻¹) (mean ± SD)	10.46 ± 1.53	9.32 ± 1.52
Ecological quality state (<i>sensu</i> WFD)	Reference condition	Reference condition

42°1'25.31"N; 1°59'38.31"E) was situated in the Merlès Stream, one of the main tributaries of the Llobregat River, in a mid-altitude mountainous area (550 m a.s.l.). Site characteristics can be found in the ECOSTRIMED public database (www.diba.cat/parcsn/qualitatrius). The two sites were subjected to similar macroclimatic conditions, shared similar geological characteristics (calcareous) and had karstic permanent sources. Riparian vegetation was dominated by *Pinus sylvestris*, *Ulmus minor*, *Alnus glutinosa* and *Fraxinus angustifolia* in WM, whereas *Pinus halepensis*, *Populus nigra* and *Salix alba* were dominant in DM. Substrates mainly comprised boulders and cobbles in both sites. Filamentous green algae, diatoms and some macrophytes occurred in some years, mainly in DM.

Following Munné & Prat (2004), both sites were classified under the same river type (Mountain Calcareous) but into two different river subtypes: 1b-Calcareous Wet mountain for WM and 2b-Calcareous Mediterranean mountain for DM. The two sites were therefore located in the same macroclimatic (regional) area but in different mesoclimatic areas, and they differed with respect to several local-scale variables (Table 1). In addition, both sites fulfilled the criteria for reference conditions proposed by Sanchez-Montoya *et al.* (2009), which allowed us to study long-term changes in aquatic communities under nearly undisturbed environmental conditions.

Environmental data

We built an ombrothermic diagram for each site to describe its general macroclimatic characteristics using monthly temperature and precipitation data recorded at nearby sites for the period 1941–2008 (data from the Meteorological Catalan Institute). We also calculated several climatic and hydrological variables using gaug-

ing station data recorded at nearby sites for the period 1995–2008 (data from the Catalan Water Agency). To calculate the hydrological variables that reflect the different components of the flow regime during the study period, we used the Indicators of Hydrological Alteration software (IHA ver 7.1, The Nature Conservancy, 2009). From the available IHA parameters, we selected four surrogates of flooding and drought: the magnitude of monthly condition (Q mean), the magnitude of extreme condition (Q Max), the frequency of minimum discharge (low pulse) and the constancy of flow (Base Flow Index, BFI) (Table 2). These four variables were considered to be optimal for our purposes, and many other variables calculated with IHA were strongly correlated with the selected variables.

Invertebrate data

We used macroinvertebrate data collected during the ECOBILL project (<http://ecobill.diba.cat/index.php?page=portada>), a long-term monitoring programme conducted by the Freshwater Ecology and Management (F.E.M.) research group at the University of Barcelona (<http://www.ub.edu/fem>) since 1994. This programme includes data from 42 sites in the Barcelona region (NE Spain; Prat & Rieradevall, 2006). To produce comparable results, sampling followed the same protocol for all sites and years. Each site was visited twice each year (spring and summer). The protocol consisted of an initial 3-min kick sample from all of the available habitats with a net of 250- μ m mesh size. Samples were first examined in the field; to capture all site biodiversity, successive samples were taken until no additional families were found (GUADALMED protocol; Jáimez-Cuellar *et al.*, 2002). The first kick sample was taken and complemented with one individual of each new family found in the succes-

Table 2 Summary of IHA parameters selected in our study (The Nature Conservancy, 2009)

Code	Type	Descriptions	Ecosystem influences
Q mean	Magnitude of monthly water conditions	Mean value for each calendar month	Habitat availability for aquatic organisms. Influences water temperature, oxygen levels, photosynthesis in water column
Q Max	Magnitude and duration of annual extreme water conditions	Annual maxima calculated on 3-day means	Structuring of aquatic ecosystems by abiotic versus biotic factors. Structuring of river channel morphology and physical habitat conditions. Influences bed-load transport, channel sediment textures, and duration of substrate disturbance
Base Flow Index (BFI)	Annual water conditions	Calculated as: 7-day minimum flow / mean flow for year	Structuring of river channel morphology and physical habitat conditions
Low pulse	Frequency and duration of extreme water conditions	Number of low pulses within each water year	Duration of stressful conditions such as low oxygen and concentrated chemicals in aquatic environments

sive samples. Samples were sorted in the laboratory and preserved in 70% alcohol. For the purposes of this study, we identified all Ephemeroptera, Plecoptera and Trichoptera (EPT) collected from 1995 until 2008 to the highest possible taxonomic resolution (generally species, or operational unit, and genera when the highest level of resolution was not possible). We used several existing taxonomic keys for Spanish and European faunas (Belfiore, 1983; Buffagni, 1997; Viera-Lanero, 2000; Tierno de Figueroa *et al.*, 2003; Bonada *et al.*, 2004), and for the most difficult genera, we consulted Spanish and European specialists. For each identified taxon, we categorised abundance into one of four ranks: 1 for 1–3 individuals/sample, 2 for 4–10, 3 for 11–100 and 4 for more than 100 individuals/sample.

Data analysis

We used mean annual and seasonal values of precipitation, temperature and discharge data from 1995 to 2008 (Table 3) and applied a Wilcoxon test (Hollander & Wolfe, 1973) using log-transformed data to identify gen-

Table 3 Mean annual climatic data and discharge attributes for the Wet Mediterranean (WM) and Dry Mediterranean (DM) sites during the cold (October–April) and the hot (April–July) periods

Variables	Mesoclimates		Wilcoxon	
	WM	DM	z	P
Climatic data				
Annual mean temperature (°C)	11.9	12.6	3.29	<0.01
Annual mean precipitation (mm)	862	731	2.48	0.01
Cold period				
Mean temperature (°C)	7.4	8.1	3.29	<0.01
Mean precipitation (mm)	383 (45.6%)	356 (49.2%)	1.91	0.05
Hot period				
Mean temperature (°C)	15.9	16.8	2.73	<0.01
Mean precipitation (mm)	321 (37%)	257 (37%)	2.47	0.01
Discharge data				
Mean annual discharge (m ³ s ⁻¹)	0.77	0.42	3.17	<0.01
Minimum (m ³ s ⁻¹) discharge (year)	0.02 (2005)	0 (2005)		
Maximum (m ³ s ⁻¹) discharge (year)	67.9 (1997)	32.03 (1997)		
Cold period				
Mean discharge (m ³ s ⁻¹)	0.79	0.36	3.17	<0.01
Hot period				
Mean discharge (m ³ s ⁻¹)	1.01	0.25	3.29	<0.01

eral environmental differences between mesoclimates (WM versus DM sites).

In addition, a total of 5114 daily data series for temperature, precipitation and discharge (from 1 October 1994 to 31 December 2008) were used to calculate 12 climatic and IHA hydrological variables. These environmental parameters were calculated separately for two periods, namely cold and hot (Tables 4 and 5, respectively). For the cold period, we used data collected from the previous autumn (October) until the day of biological sampling the following spring (April); for the hot period, we used data from the days between the two biological sampling periods (April–July). Therefore, the end of the cold period included the spring sample of macroinvertebrates, while the hot period included the summer sample. This method allowed us to capture hydrological variability preceding the sampling period that was likely to be of relevance to macroinvertebrates (Durance & Ormerod, 2007). We used these environmental variables to analyse long-term climatic and hydrological variability using an independent Kendal-Tau trend test for each mesoclimate and for each period (cold and hot). This test is widely used in trend analysis and is preferable over the parametric alternative (e.g. regression) because it requires fewer assumptions and has a higher power when data are both normally and non-normally distributed (Esterby,

Table 4 Kendal-Tau values (τ) and trend direction for precipitation, temperature and flow attributes for the Wet Mediterranean (WM) and Dry Mediterranean (DM) sites during the cold period (October–April)

Cold period	WM			DM		
	Tau (τ)	P-value	Trend	Tau (τ)	P-value	Trend
Precipitation						
P max	-0.22	0.29	ns	-0.37	0.07	(-)
Total P	-0.40	0.05	(-)	-0.18	0.37	ns
Total P/rainy days	-0.64	<0.01	(-)	-0.45	0.02	(-)
Annual mean P	-0.30	0.09	ns	-0.28	0.18	ns
Temperature						
T mean	-0.22	0.30	ns	-0.31	0.14	ns
T Max	0.08	0.73	ns	-0.04	0.86	ns
T min	0.08	0.73	ns	-0.31	0.14	ns
T (annual mean)	0.31	0.15	ns	-0.02	0.95	ns
Flow attributes						
Q mean	-0.38	0.05	(-)	-0.42	0.04	(-)
Q Max	-0.47	0.02	(-)	-0.35	0.08	ns
BFI	0.17	0.40	ns	0.01	0.90	ns
Low pulse	0.49	0.02	(+)	0.43	0.04	(+)

(+), positive trend; (-), negative trend; ns, non-significant trend.

Table 5 Kendall-Tau values and trend direction for precipitation, temperature and flow attributes for the Wet Mediterranean (WM) and Dry Mediterranean (DM) sites during the hot period (April–July)

Hot period	WM			DM		
	Tau (τ)	P-value	Trend	Tau (τ)	P-value	Trend
Precipitation						
P Max	-0.07	0.74	ns	-0.05	0.82	ns
Total P	-0.12	0.58	ns	-0.12	0.58	ns
Total	-0.158	0.47	ns	-0.14	0.50	ns
P/rainy days						
Annual mean P	-0.30	0.09	ns	-0.28	0.18	ns
Temperature						
T mean	0.32	0.13	ns	0.01	0.98	ns
T Max	-0.04	0.86	ns	-0.19	0.39	ns
T min	0.16	0.46	ns	-0.21	0.32	ns
T (annual mean)	0.31	0.15	ns	-0.02	0.95	ns
Flow attributes						
Q mean	0.04	0.86	ns	0.07	0.74	ns
Q Max	0.25	0.22	ns	0.04	0.86	ns
BFI	-0.53	<0.01	(-)	-0.54	<0.01	(-)
Low pulse	0.01	0.99	ns	0.16	0.46	ns

(+), positive trend; (-), negative trend; ns, non-significant trend.

1996). A significant positive Kendall-Tau (τ) indicates that the test variable increases with time, while a negative value indicates the contrary.

To assess whether the mesoclimates differed in macro-invertebrate composition, we first investigated differences in EPT fauna with principal component analysis (PCA) using the mean annual rank abundance matrix of each taxon. We tested for significant differences between mesoclimates with a between-class analysis tested with permutations (1000 runs) (Romesburg, 1985). In addition, we used similarity percentages analysis (SIMPER; Past ver 1.94b) to determine the % dissimilarity between the two sites and to calculate the contribution of individual taxa to the observed differences. SIMPER uses a Bray–Curtis similarity matrix to compute the average dissimilarity between all pairs of intergroup samples (Clarke, 1993).

Long-term biological variability was analysed within and between mesoclimates. To assess the long-term variability within each mesoclimate and the relationship between EPT composition and the environmental variables, we used canonical correspondence analysis (CCA) on the 'site x taxon seasonal rank abundance matrix' (Ter Braak, 1986). The obtained CCA scores were analysed with a Kendall-Tau trend test to investigate the presence and significance of trends.

To assess the long-term variability in each mesoclimate, we used a Bray–Curtis similarity matrix calculated separately for each site. We compared the coefficient of similarity obtained for the last year (year 14) with that of each of the preceding years (from 1 to 13 in the series); high values of similarity between the last year and the first years in the series indicate low variability with time. In addition, we assessed short-term variability between mesoclimates using the same Bray–Curtis similarity matrix by comparing the coefficients among successive years (1 with 2, 2 with 3, etc.); high values indicated that assemblages were more similar and hence more stable from year to year. All statistical analyses were performed with PAST ver.1.94b (Hammer, Harper & Ryan, 2001) and the 'Kendall' and 'ade4' libraries (Hipel & McLeod, 2005; Dray & Dufour, 2007; McLeod, 2011) within the R statistical package (R version. 2.13.1).

Results

Long-term environmental variability

Ombrothermic diagrams for the two mesoclimates showed that maximum and minimum temperatures occurred in July–August and January, respectively (Fig. 1a). Mean daily temperatures in February and March were relatively mild. Rainfall was usually concentrated in late summer, autumn and spring, and the lowest precipitation levels occurred during the winter season. Despite the high similarity of the two diagrams (macroclimate level), reflecting a sub-humid Mediterranean climate (*sensu* Gallart *et al.*, 2002), significant differences between mesoclimates were detected for both climatic and discharge variables (Table 3).

With respect to long-term annual trends in climatic variables, mean annual precipitation decreased progressively over the studied years at both sites, with the period from 1995 to 1997 being wetter and the period from 2004 to 2007 being drier than the mean (Fig. 1c). Mean annual discharge decreased between 1995 and 2007 in both mesoclimates (Fig. 1d). The 1996–1997 period was characterised by floods, with a peak discharge value recorded on 18 December 1997 in both sites (WM: $Q = 67.91 \text{ m}^3 \text{ s}^{-1}$; DM: $Q = 32.03 \text{ m}^3 \text{ s}^{-1}$). In contrast, there were two periods of low flow: the year 2000 and the period 2005–2007. The lowest discharge was recorded in 2005 when DM was completely dry for several days in the summer, something that occurs only rarely.

When analysing long-term seasonal (cold versus hot period) trends, Kendall-Tau tests showed no significant

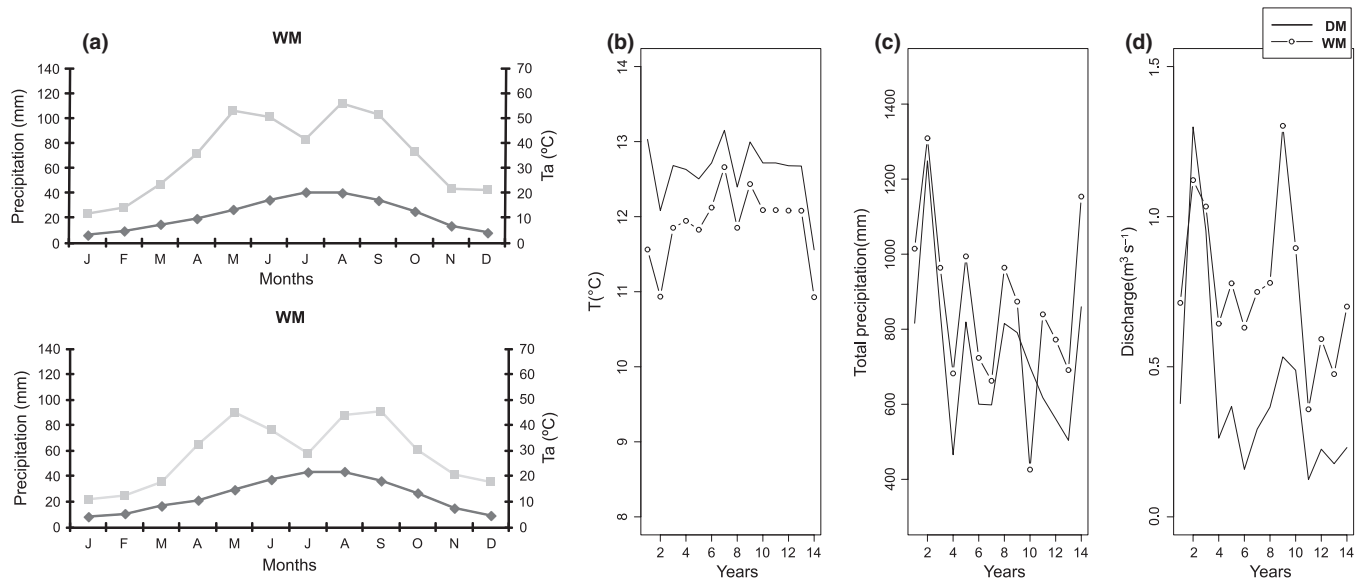


Fig. 1 (a) Ombrothermic diagrams for both studied sites (from 1941 to 2007) and trends in several parameters over a 14-year period (from 1995 to 2008). (b) Mean annual temperature, (c) Total precipitation (annual mean) and (d) mean annual discharge. Codes for years are progressive: 1 = 1995 to 14 = 2008.

temporal trends in temperature regardless of the mesoclimate and the time period considered (Fig. 1b; Tables 4 and 5). In particular, for the cold period, negative trends in Q mean were observed in both mesoclimates ($\tau_{WM} = -0.38$, $P = 0.05$; $\tau_{DM} = -0.42$, $P = 0.04$) and total precipitation accumulated for rainy days (Total P/rainy days: $\tau_{WM} = -0.64$, $P < 0.01$; $\tau_{DM} = -0.45$, $P = 0.02$), whereas there was a positive trend in the number of low-flow days observed (low pulse: $\tau_{WM} = -0.49$, $P = 0.02$; $\tau_{DM} = -0.43$, $P = 0.04$). In the trend analysis, differences between mesoclimates were detected for maximum discharge (Q Max) and total precipitation accumulated (Total P); these differences were significant for WM (Q Max: $\tau_{WM} = -0.47$, $P = 0.02$; Total P: $\tau_{WM} = -0.4$, $P = 0.05$) but not for DM. For the hot period (Table 5), negative trends were found in both mesoclimates for BFI ($\tau_{WM} = -0.53$, $P < 0.01$; $\tau_{DM} = -0.54$, $P < 0.01$) only. Therefore, the hot period was more irregular interannually, with higher between-year variability.

Macroinvertebrate composition and mesoclimates

A total of 61 EPT taxa (from 24 families) were collected in the two mesoclimates during the 14-year study period. Only 15 taxa, up to 25% of the total, were present in both streams (Appendix 1). Significant differences in EPT composition between the two streams were found (Obs = 0.549, $P < 0.01$), with 62.5% of the variance in

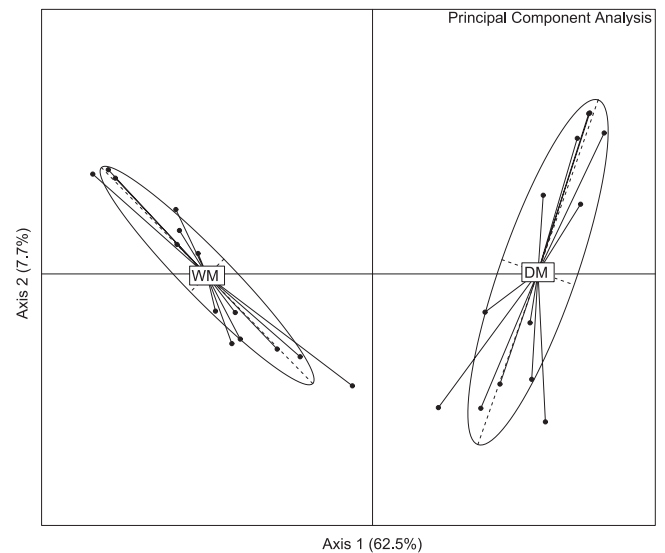


Fig. 2 Principal component analysis (PCA) based on annual means of EPT abundance ranks during the period 1995–2008. PCA axis 1 (62.5% of explained variability) and PCA axis 2 (7.7% of explained variability). Ellipses represent 95% of the inertia of each group of sites: Wet Mediterranean (WM) and Dry Mediterranean (DM).

composition explained by the first PCA axis (Fig. 2). Taxa that contributed most to the dissimilarities between streams (Table 6, SIMPER, 80.35%) were, for DM, *Hydropsyche pellucidula*, *Baetis pavidus*, *Chimarra marginata*, *Caenis macrura* and *Leuctra geniculata* and for WM, *Serratella ignita*, *Dinocras cephalotes*, *Protonemoura beatensis*, *Baetis nicolae*, *Rhitrogena semicolorata*, *Hydropsyche instabilis*,

Table 6 Annual mean abundance rank of taxa that contributed up to 60% of Bray–Curtis similarity (SIMPER analysis) between samples from the Wet Mediterranean (WM) and Dry Mediterranean (DM) sites

Taxon	Cont.	Cum.per	WM	DM
			Abb	Abb
<i>Hydropsyche pellucidula</i>	4.489	5.587	0	2.71
<i>Serratella ignita</i>	4.485	11.17	2.84	0.03
<i>Baetis pavidus</i>	3.707	15.78	0	2.21
<i>Chimarra marginata</i>	3.481	20.12	0	2.13
<i>Dinocras cephalotes</i>	3.242	24.15	2.05	0
<i>Protonemura beatensis</i>	3	27.88	1.95	0
<i>Caenis beskidensis</i>	2.9	31.49	1.89	0
<i>Baetis nicolae</i> (gr. <i>alpinus</i>)	2.81	34.99	2	0.2
<i>Caenis</i> gr. <i>macrura</i>	2.752	38.42	0.07	1.77
<i>Rhitrogena</i> gr. <i>semicolorata</i>	2.627	41.69	1.7	0
<i>Baetis rhodani</i>	2.58	44.9	2.96	1.41
<i>Hydropsyche instabilis</i>	2.355	47.83	1.4	0
<i>Leuctra</i> sp.	2.321	50.72	3	1.91
<i>Epeorus silvicola</i>	1.901	53.08	1.23	0
<i>Ecdyonurus</i> gr. <i>venosus</i>	1.888	55.43	2.71	1.86
<i>Leuctra geniculata</i>	1.748	57.61	0	1.07
<i>Rhyacophila meridionalis</i>	1.713	59.74	1.02	0
<i>Odontocaerum albicorne</i>	1.711	61.87	1.13	0

Cont, contribution; Cum. per, cumulative percentage; Abb, mean abundance rank, values between 1 and 4, see text for details.

Epeorus silvicola and *Rhyacophila meridionalis*. *Baetis* and *Hydropsyche* genera had different species in the two mesoclimate areas. However, despite the fact that the two EPT communities differed significantly, the mean annual EPT richness did not vary greatly between streams (20.5 taxa in DM and 25 in WM).

Long-term biological variability within mesoclimates

Given that trends in environmental variables that are common to both streams were only observed during the cold period, the relationships between EPT species composition and climatic and hydrological variables were only investigated for this period. We used hydrological variables, rather than precipitation or temperature, because these variables are more directly associated with aquatic organisms than is precipitation and because temperature did not exhibit significant long-term trends (Table 4). In addition, we found a high correlation between some hydrological and precipitation variables (WM: Q Max and P Max $r = 0.77$; DM: Q Max and P Max $r = 0.85$, Q mean and P (O-A) $r = 0.92$; all $P < 0.01$). The CCA analysis (Table 7, Figs 3 and 4) produced an ordination in which the overall analysis and the first axes were highly significant for both streams (Monte Carlo test with 1000 permutations, $P < 0.001$).

Table 7 Canonical correspondence analysis (CCA) among selected hydrological variables and EPT abundance data for the cold period (October–April) for the Wet Mediterranean (WM) and Dry Mediterranean (DM) sites. The fraction of total variation explained by the first three axes and the scores for each constrained variable (hydrological variables) were reported

Results CCA	WM			DM		
	Axis1	Axis2	Axis3	Axis1	Axis2	Axis3
% Variance	47.57	34.04	18.39	54.62	31.04	14.23
Eigen value	0.09	0.07	0.04	0.19	0.11	0.05
P-value	<0.01	<0.01	0.02	0.04	0.04	Ns
Biplot scores for constraining variables (sc)						
Q mean	-0.55	-0.41	0.74	-0.70	-0.32	0.07
Q max	-0.45	-0.83	0.12	-0.55	0.34	-0.06
BFI	-0.04	0.85	-0.38	0.16	-0.20	-0.62
Low pulse	0.84	-0.16	0.14	0.71	-0.29	0.16
Kendall-Tau test results						
Tau	0.48**	ns	ns	0.73**	ns	ns
Trend	(+)	None	None	(+)	None	None

Values of the Kendall-Tau and trend direction for each CCA axis are also presented: + = positive; none = no significant trend.

** $P < 0.01$; ns = not significant.

The study years from 1 to 14 were oriented from left to right along the first axis, and Kendall-Tau tests revealed a significant temporal trend in EPT composition along the first axis in both streams (Table 7) that matched trends in hydrological variables. In particular, variation in species composition along the first axis in both streams was positively constrained by low pulse (WM: sc = 0.84; DM: sc = 0.71) and negatively constrained by Q mean (WM: sc = -0.55; DM: sc = -0.7). However, the species that responded to these two variables differed by site. In WM, we observed a shift over time (Fig. 3) from species such as *Siphonoperla torrentis*, *Rhyacophila meridionalis*, *Rhyacophila tristis*, *Habrophlebia modesta* and *Amphinemoura sulcicollis*, which dominated during the first years in the series, to species such as *Glossosoma* sp., *Lasiocephala basalis*, *Polycentropus* sp., *Isoperla acicularis*, and *Baetis muticus*, which were mainly present during the last years. In DM, *Mesophylax aspersus*, *Mystacides azurea*, *Cloeon dipterum*, *Tinodes dives*, *Hydropsyche bulbifera* and *Serratella ignita* were the most dominant species during the last years in the series (Fig. 4); in contrast, *Hydropsyche exocellata*, *Hydropsyche brevis* and *Ecdyonurus venosus* were mainly present during the first years. No temporal trends along the second axis were detected in EPT structure of either stream. In particular, for WM, the second axis was positively constrained by Base Flow Index (sc = 0.85) and negatively constrained by Q max (sc = -0.83). In contrast, for DM, no significant relationships between the second axis and the hydrological variables were observed.

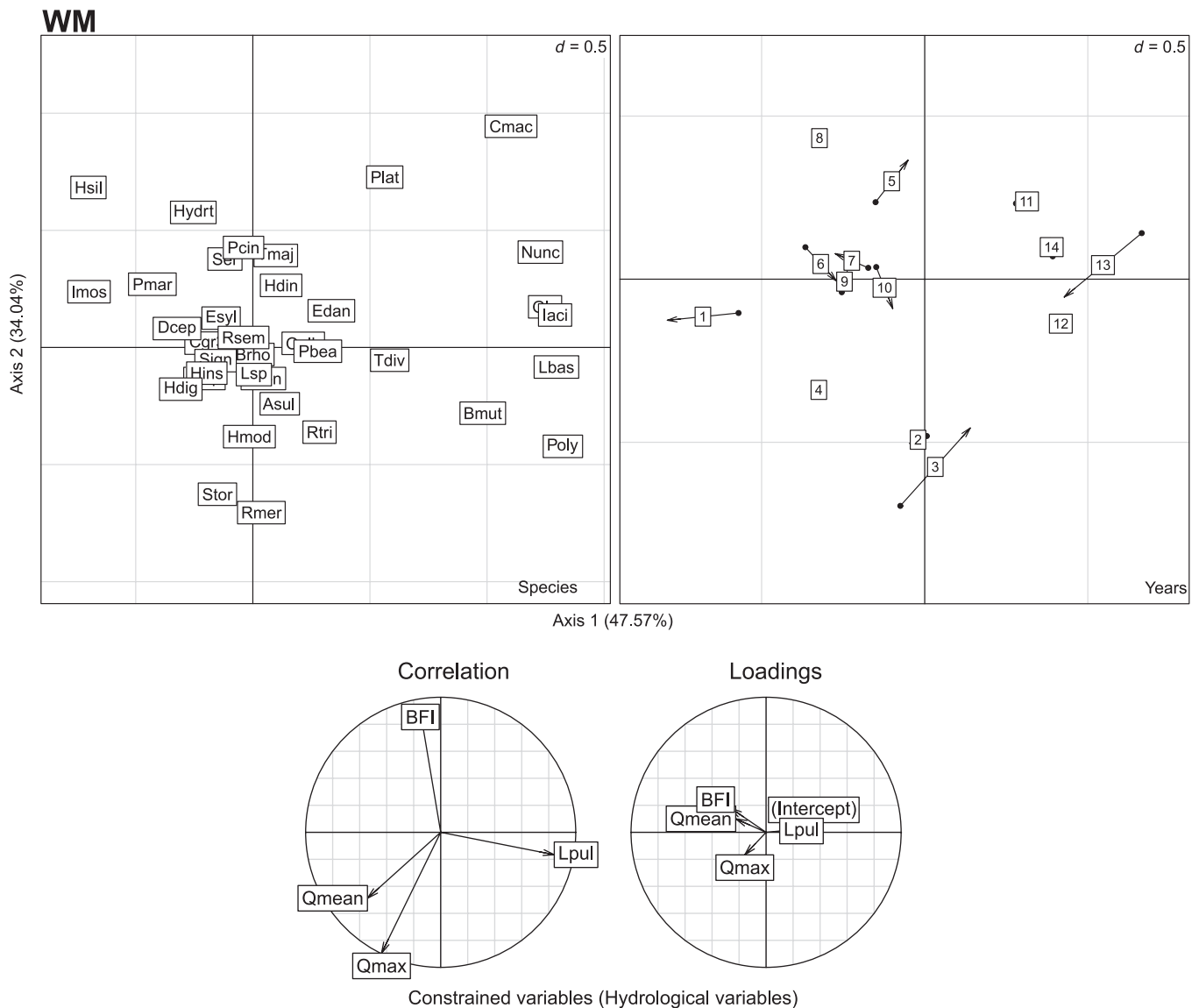


Fig. 3 Canonical correspondence analysis (CCA) among selected hydrological variables and EPT abundance data in the Wet Mediterranean site (WM) during the cold period. The plot shows the ordination of species (left) and years (right) along the first (47.57% of explained variability) and the second (34.04% of explained variability) axes. Arrows indicate distances between the position by regression on environmental variables and the position by regression on biological data. Correlation and loadings for constrained (hydrological) variables are also displayed to allow interpretation of the ordination drivers. (Codes for species are listed in Appendix 1; Codes for years are progressive: 1 = 1995 to 14 = 2008; BFI = Base Flow Index; Lpul = number of low pulse; Qmax = maximum of discharge; Qmean = mean seasonal discharge).

Long-term biological variability between mesoclimates

Although significant temporal trends in EPT composition were detected in both streams with a corresponding increase in EPT richness over the study period at both sites (Fig. 5a), different results were found when we analysed the long-term and short-term biological variability in WM and DM. With respect to long-term variability (i.e. similarity of each year with the final study

year; Fig. 5b), our results indicated that similarity values of EPT fauna increased with time in both sites, but these values were always higher in WM than in DM. Regarding short-term variability (Fig. 5c), WM exhibited a steady percentage of similarity greater than 70% from the 4–5th year of the series to the 14th; during the first 4 years, the similarity values decreased. In contrast, the percentage of similarity in DM was always more variable and always below 70% during the series.

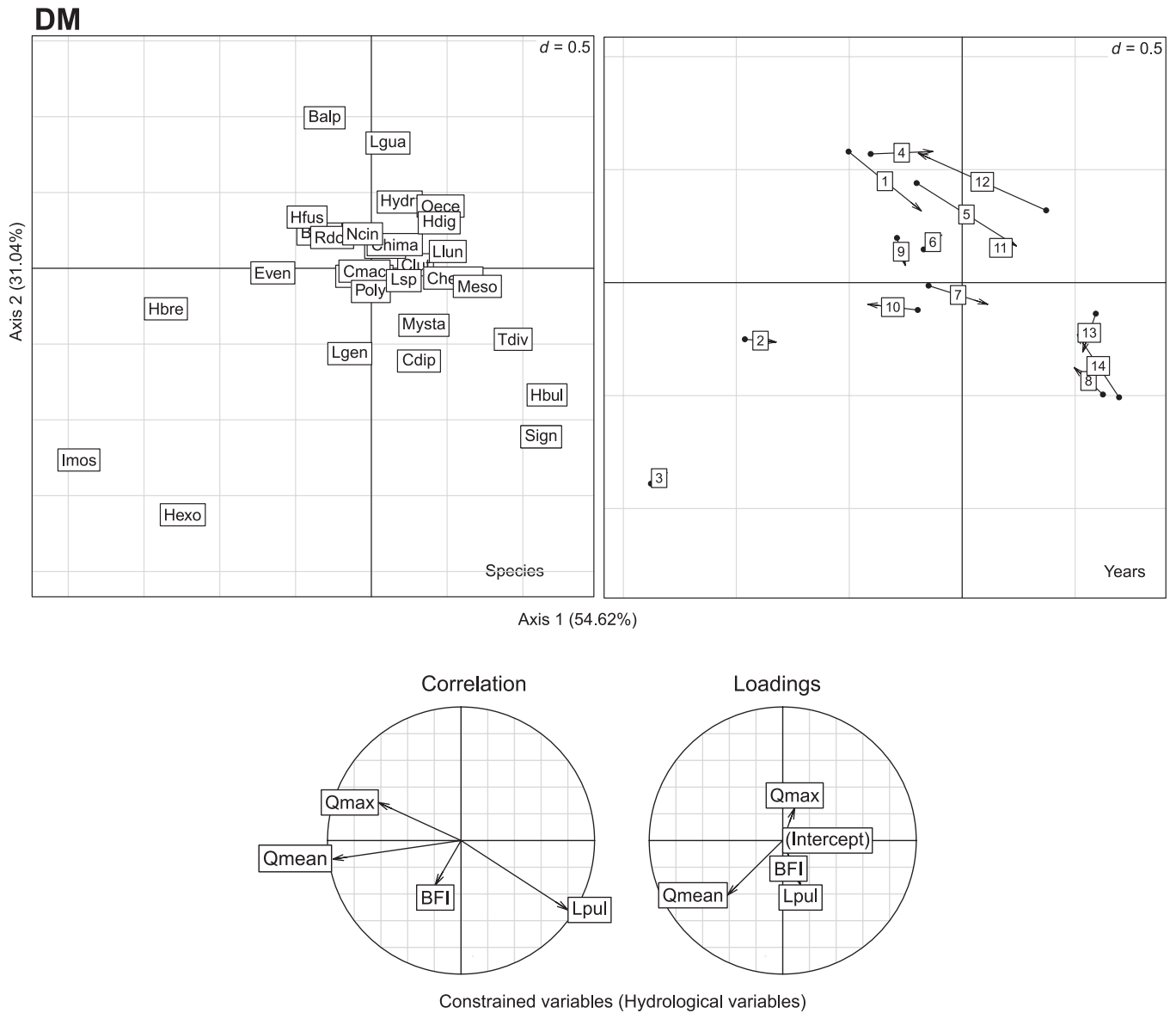


Fig. 4 Canonical correspondence analysis (CCA) among selected hydrological variables and EPT abundance data in the Dry Mediterranean site (DM) during the cold period. The plot shows the ordination of species (left) and years (right) along the first (54.62% of explained variability) and the second (31.04% of explained variability) axes. Arrows indicate distances between the position by regression on environmental variables and the position by regression on biological data. Correlation and loadings for constrained (hydrological) variables are also displayed to allow in interpretation of the ordination drivers. (Codes for species are listed in Appendix 1; codes for years are progressive: 1 = 1995 to 14 = 2008; BFI = Base Flow Index; Lpul = number of low pulse; Qmax = maximum of discharge; Qmean = mean seasonal discharge).

Discussion

Our work has provided information regarding long-term environmental and biological responses in a Mediterranean climate region. At the macroclimate scale, long-term trends indicated that precipitation has decreased over the last 14 years and that the frequency of drought events has increased, which is consistent with climate-change predictions made for Southern Europe (IPCC,

2001; Shindell, 2007). The extreme flood and drought periods observed in our study also correspond with what is known for the region: floods were more common at the beginning of the series and droughts more common at the end. For example, Amengual *et al.* (2009) found that two of the most catastrophic extreme floods during the last 50 years in the Llobregat catchment occurred in November 1996 and December 1997. In contrast, in 2005, Spain, Portugal and France experienced an

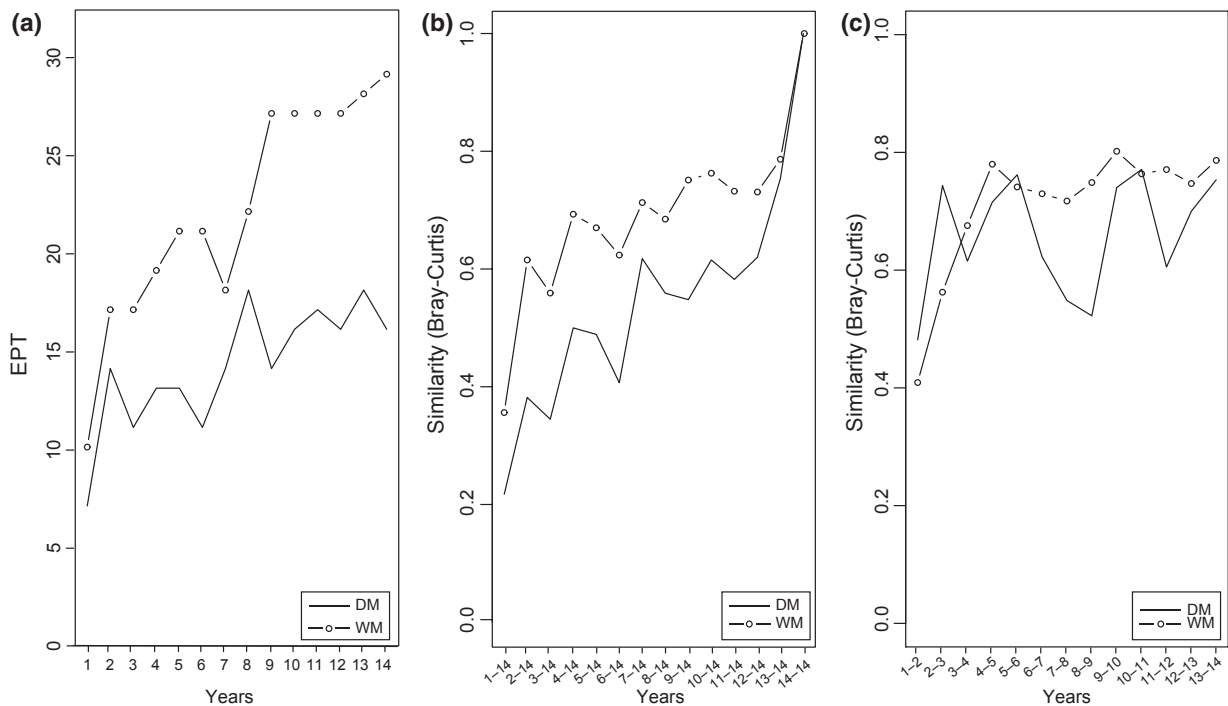


Fig. 5 Variability in spring between the two sites in richness and taxonomic composition. (a) EPT richness variability; (b) Long-term composition structure variability (Bray–Curtis index): the coefficient of similarity obtained for the last year (year 14) was compared with that of each of the preceding years (from 1 to 13); (c) Short-term composition structure variability (Bray–Curtis index): the coefficients of similarity obtained were compared among successive years (1 with 2, 2 with 3, etc.). Codes for years are progressive: 1 = 1995 to 14 = 2008.

extreme drought with the lowest annual precipitation recorded since 1947. The drought impacted drinking water availability, hydropower potential, water quality and fire risk (European commission, 2007). Consistent with the shift from floods to droughts, a significant dominance of EPT species specific to riffle-like conditions was observed during the first period of the time series in both sites, while EPT species characteristic of pool-like conditions dominated in the most recent years (see families' characteristics of riffles and pools in Bonada *et al.*, 2008).

Several studies have analysed the effects of floods and droughts on macroinvertebrate assemblages, recognising them as major categories of natural disturbance in running waters (Hildrew & Giller, 1994; Lake, 2000) those studies conducted in high and low flows in the Mediterranean region described a shift from taxa with riffle-like strategies to those with pool-like strategies (for details, see Bonada *et al.*, 2008). Similarly, for California, Bêche & Resh (2007a,b) showed that much of the temporal variation in composition and abundance of benthic macroinvertebrates was explained by short-term climatic patterns (i.e. annual differences in precipitation). In Australian streams (New South Wales), Chessman (2009) found that families of invertebrates that live in faster-

flowing habitats and in cold waters were most likely to have declined during a 13-years period of decreasing rainfall and river flow. Interestingly, although the detected trend was consistent between study sites at the macroclimate scale, EPT fauna was less variable in the stream in the Wet Mediterranean climate area (WM) than in the stream in the Dry Mediterranean climate area (DM) with respect to both short-term and long-term changes, which confirms our general hypothesis.

Different trends were detected between WM and DM during the low-flow and drought events that characterise the second part of the series (the EPT communities appeared stable during the low flows in WM, but were highly variable in DM). Droughts are considered to be a 'ramp disturbance' (Lake, 2000, 2003), and the intensity of its effects on macroinvertebrates is influenced by factors such as timing, duration, intensity and the presence of refuges (Boulton, 2003). As flows decrease, habitat space is generally reduced; organisms respond to these conditions by continuous colonisation–extinction processes that often lead to a reduction in invertebrate density (Suren & Jowett, 2006). The higher interannual variability in DM might reflect typical Mediterranean climate conditions, including some years with normal conditions and others with drought conditions in the

spring and summer (the lowest discharge was recorded in 2005 and during this year, DM was dry for several summer days). Therefore, according to Bond, Lake & Arthington (2008), our DM site could be characterised by a greater number of taxa that possess adaptations that allow them either to survive the drought by 'sitting it out' (resistance traits) or to recolonise and recruit after the drought ceases (resilience traits), which explains the higher interannual variability observed in this site.

However, during the first part of the study period, which was characterised by flood events, WM had very different EPT communities between consecutive years (Fig. 5c). Floods are considered to be a 'pulse disturbance' (Lake, 2000). During these events, invertebrates are affected by a combination of water velocity (Holmuzki & Biggs, 2000) and physical scouring due to bed movement (Biggs *et al.*, 2001). General responses of invertebrate communities to floods include a reduction in density and taxonomic richness (Gjerlov, Hildrew & Jones, 2003). Therefore, the higher biannual variability in WM might be related to the higher intensity (magnitude) of the floods detected in WM during the first period studied (e.g. WM: $67 \text{ m}^3 \text{ s}^{-1}$; DM: $32 \text{ m}^3 \text{ s}^{-1}$ during the extreme flood of 1997) and might be influenced by the increase in taxon richness during the post-flood recovery period. Our study also showed that differences between mesoclimates (faunal composition and biological variation) were maintained throughout the study period, indicating that differences between mesoclimates (spatial scale) were greater than differences among years (temporal scale). For example, species typically found in the Mediterranean Basin, such as *Baetis pavidus*, *Cloeon* sp., *Centroptilum luteolum*, *Chimarra marginata*, *Mystacides azurea*, *Mesophylax* sp. and *Leuctra geniculata*, were only present in DM (Appendix 1 and SIMPER analysis); in contrast, *Perla marginata*, *Siphonoperla torrentis*, *Dinocras cephalotes* (and other species in the order Plecoptera), whose nymphs generally live in mostly cold, well-oxygenated running waters (Fochetti & Tierno de Figueroa, 2008), were found only in WM. Such differences persisted throughout the 14-year study period without regard to the climatic trend, indicating that natural spatial variability between the two streams influences abiotic characteristics. This finding may be due to patterns in species occurrence and community composition at regional spatial scales, which are primarily associated with the dispersal of organisms across the landscape (Shurin, 2000). Once an organism has arrived in a habitat, local-scale factors, such as the suitability of the abiotic environment and interactions with established species, determine community composition (Ricklefs & Schluter, 1993).

Therefore, our results suggest that within-region variability can be especially important in highly heterogeneous regions such as the Mediterranean, thus supporting the general view that complex local processes that emerge at local scales can have variable impacts on streams (Heino, 2011).

Future challenges

One of the key challenges facing freshwater ecologists is to develop tools for detecting the impacts of climate change in natural systems that are as general and realistic as possible (Hampe, 2004). However, decision-making requires much more precise information such as a quantitative estimate of the changes, a more precise regional distribution of their effects and an associated evaluation of their variability. Under the possible scenario of increased frequency of extreme events, management will require measures to adapt societies or ecosystems to climate change. Our results showed that EPT species in two reference sites that belonged to two different river sub-types varied differentially with time. Because reference conditions used to assess the ecological status of rivers worldwide (e.g. in Europe, European Commission, 2003; Nijboer *et al.*, 2004; Noges *et al.*, 2009) can be highly dynamic, interannual variability (natural or driven by climate change) should be considered when the reference conditions are established and used in bioassessment (e.g. Buffagni, Armanini & Erba, 2009; Poquet *et al.*, 2009; Munné & Prat, 2011).

The presence of other stressors in addition to interannual variability can affect organisms in ways that are even more unpredictable. For example, summer droughts will not only lead to elevated temperatures and habitat fragmentation but may also exacerbate the impacts of eutrophication and toxins (Lindh, 1992; Avila, Neal & Terradas, 1996; Bonada *et al.*, 2007). Therefore, according to other studies (Feio *et al.*, 2010; Woodward, Perkins & Brown, 2010), the ways in which we currently assess 'ecological status' could become increasingly obsolete over time as the baseline drifts away from earlier (cooler or wetter) reference conditions. In conclusion, for biomonitoring purposes, regular assessment of reference sites will be needed to adjust classification systems for climate-induced changes. Given the limited number of monitoring programmes addressing interannual variability at multiple freshwater sites, new programmes should be launched to facilitate direct examination of the effects of climate change and other major anthropogenic stressors on biodiversity (e.g. Daufresne, Bady & Fruget, 2007; Durance & Ormerod,

2007). Such programmes should also help to determine how reference conditions for freshwater bio-assessment could be altered by natural environmental change.

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References

- Amengual A., Romero R., Vich M. & Alonso S. (2009) Inclusion of potential vorticity uncertainties into a hydrometeorological forecasting chain: application to a medium size basin of Mediterranean Spain. *Hydrology and Earth System Sciences Discussions*, **6**, 535–587.
- Araújo M.B. & Guisan A. (2006) Five (or so) challenges for species distribution modelling. *Journal of Biogeography*, **33**, 1677–1688.
- Arnell N.W. (1999) The effect of climate change on hydrological regimes in Europe: a continental perspective. *Global Environmental Change*, **9**, 5–23.
- Aschmann H. (1973) Distribution and peculiarity of Mediterranean ecosystems. In: *Mediterranean Type Ecosystems: Origin and Structure* (Eds F. Di Castri & H.A. Mooney), pp. 405. Springer-Verlag, New York.
- Avila A., Neal C. & Terradas J. (1996) Climate change and implications for streamflow and streamwater chemistry in a Mediterranean catchment. *Journal of Hydrology*, **177**, 99–116.
- Bêche L.A., McElravy E.P. & Resh V.H. (2006) Long-term seasonal variation in the biological traits of benthic macroinvertebrates in two Mediterranean-climate streams. *Freshwater Biology*, **51**, 56–75.
- Bêche L.A. & Resh V.H. (2007a) Biological traits of benthic macroinvertebrates in California Mediterranean-climate streams: long-term annual variability and trait diversity patterns. *Fundamental and Applied Limnology*, **169**, 1–23.
- Bêche L.A. & Resh V.H. (2007b) Short-term climatic trends affect the temporal variability of macroinvertebrates in California 'Mediterranean' streams. *Freshwater Biology*, **52**, 2317–2339.
- Belfiore C. (1983) 24. Efemerotteri (Ephemeroptera). In: *Guide per il riconoscimento delle specie animali delle acque interne italiane* (Ed. S. Ruffo), pp. 113. C.N.R., Museo Civico di Storia Naturale di Verona, Verona.
- Biggs B.J.F., Duncan M.J., Suren A.M. & Holomuzki J.R. (2001) The importance of bed stability to benthic ecosystems. In: *Gravel-Bed Rivers V* (Ed. M.P. Mosley), pp. 423–449. New Zealand Hydrological Society, Christchurch.
- 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.
- Bonada N., Rieradevall M., Dallas H., Davis J., Day J., Figueroa R. *et al.* (2008) Multi-scale assessment of macroinvertebrate richness and composition in Mediterranean-climate rivers. *Freshwater Biology*, **53**, 772–788.
- Bonada N., Zamora-Muoz C., Rieradevall M. & Prat N. (2004) Trichoptera (Insecta) collected in Mediterranean river basins in Spain: taxonomic remarks and notes on ecology. *Graellsia*, **60**, 41–69.
- Bond N.R., Lake P.S. & Arthington A.H. (2008) The impacts of drought on freshwater ecosystems: an Australian perspective. *Hydrobiologia*, **600**, 3–16.
- Boulton A.J. (2003) Parallels and contrasts in the effects of drought on stream macroinvertebrate assemblages. *Freshwater Biology*, **48**, 1173–1185.
- Bradley D.C. & Ormerod S.J. (2001) Community persistence among upland stream invertebrates tracks the North Atlantic Oscillation. *Journal of Animal Ecology*, **70**, 987–996.
- Buffagni A. (1997) Mayfly community composition and the biological quality of streams. In: *Ephemeroptera & Plecoptera. Biology-Ecology-Systematics* (Eds P. Landolt & M. Sartori), pp. 235–246. Mauron + Tinguely & Lachat SA, Fribourg.
- Buffagni A., Armanini D.G. & Erba S. (2009) Does the lentic-lotic character of rivers affect invertebrate metrics used in the assessment of ecological quality? *Journal of Limnology*, **68**, 92–105.
- Burgmer T., Hillebrand H. & Pfenninger M. (2007) Effects of climate driven temperature changes on the diversity of freshwater macroinvertebrates. *Oecologia*, **151**, 93–103.
- Chessman B.C. (2009) Climatic changes and 13-year trends in stream macroinvertebrate assemblages in New South Wales, Australia. *Global Change Biology*, **15**, 2791–2802.
- Clarke K.R. (1993) Non-parametric multivariate analyses of changes in community structure. *Australian Journal of Ecology*, **18**, 117–143.
- Conacher A.J. & Sala M. (1998) *Land Degradation in Mediterranean Environments of the World: Nature and Extent*

- Cause and Solutions*. John Wiley & Sons, New York, pp. 491.
- Daget P.H., Ahdali L. & David P. (1988) Mediterranean bioclimate and its variation in the Palearctic region. In: *Mediterranean-Type Ecosystems: A Data Source Book* (Ed. R.L. Specht), pp. 139–148. Kluwer Academic Publishers, Dordrecht.
- Daufresne M., Bady P. & Fruget J.F. (2007) Impacts of global changes and extreme hydroclimatic events on macroinvertebrate community structures in the French Rhône River. *Oecologia*, **151**, 544–559.
- Daufresne M., Lengfellner K. & Sommer U. (2009) Global warming benefits the small in aquatic ecosystems. *Proceedings of the National Academy of Sciences of the United States of America*, **106**, 12788–12793.
- Dray S. & Dufour A.B. (2007) The ade4 package: implementing the duality diagram for ecologists. *Journal of Statistical Software*, **22**, 1–20.
- Durance I. & Ormerod S.J. (2007) Climate change effects on upland stream macroinvertebrates over a 25-year period. *Global Change Biology*, **13**, 942–957.
- Esterby S.R. (1996) Review of methods for the detection and estimation of trends with emphasis on water quality applications. *Hydrological Processes*, **10**, 127–149.
- European Commission (2003) *Common Implementation Strategy for the Water Framework Directive (2000/60/EC) Guidance document no. 10. River and Lakes—typology, reference conditions and classification systems*. Office for Official Publications of the European Communities, Luxembourg. pp 116.
- European commission (2007) *Mediterranean Water Scarcity and Drought Report*. Mediterranean Water Scarcity & Drought Working Group (MED W.S. & D.W.G.) Technical Report. Office for Official Publications of the European Communities, Luxembourg.
- Feio M.J., Coimbra C.N., Garça M.A.S., Nicholas S.J. & Norris R.H. (2010) The Influence of extreme climatic events and human disturbance on macroinvertebrate community patterns of a Mediterranean stream over 15 y. *Journal of North American Benthological Society*, **29**, 1397–1409.
- Fochetti R. & Tierno de Figueroa J.M. (2008) Global diversity of stoneflies (Plecoptera; Insecta) in freshwater. *Hydrobiologia*, **595**, 365–377.
- Gallart F., Llorens P., Latron J. & Regúés D. (2002) Hydrological processes and their seasonal controls in a small Mediterranean mountain catchment in the Pyrenees. *Hydrology and Earth System Sciences*, **6**, 527–537.
- Gasith A. & Resh V.H. (1999) Streams in Mediterranean climate regions: abiotic influences and biotic responses to predictable seasonal events. *Annual Review of Ecology and Systematics*, **30**, 51–58.
- Gjerlov C., Hildrew A.G. & Jones J.I. (2003) Mobility of stream invertebrates in relation to disturbance and refugia: a test of habitat templet theory. *Journal of the North American Benthological Society*, **22**, 207–223.
- Hammer Ø., Harper D.A.T. & Ryan P.D. (2001) PAST: paleontological statistics software package for education and data analysis. *Palaeontologia Electronica*, **4**, 9. Available at: http://palaeo-electronica.org/2001_1/past/issue1_01.htm. Last access date: 08/08/2012.
- Hampe A. (2004) Bioclimate envelope models: what they detect and what they hide. *Global Ecology and Biogeography*, **13**, 469–476.
- Harper M.P. & Peckarsky B.L. (2006) Emergence cues of a mayfly in a high-altitude stream ecosystem: potential response to climate change. *Ecological Applications*, **16**, 612–621.
- Heino J. (2009) Biodiversity of aquatic insects: spatial gradients and environmental correlates of assemblage level measures at large scales. *Freshwater Reviews*, **2**, 1–29.
- Heino J. (2011) A macroecological perspective of diversity patterns in the freshwater realm. *Freshwater Biology*, **56**, 1703–1722.
- Heino J., Virkkala R. & Toivonen H. (2009) Climate change and freshwater biodiversity: detected patterns, future trends and adaptations in northern regions. *Biological Reviews*, **84**, 39–54.
- Heuvelink G.B.M. (1998) Uncertainty analysis in environmental modelling under a change of spatial scale. *Nutrient Cycling in Agroecosystems*, **50**, 255–264.
- Hildrew A.G. & Giller P.S. (1994) Patchiness, species interactions and disturbance in the stream benthos. In: *Aquatic Ecology: Scale, Pattern and Process* (Eds P.S. Giller, A.G. Hildrew & D.G. Raffaelli), pp. 21–62. Blackwell Scientific Publications, London, UK.
- Hipel K.W. & McLeod A.I. (2005) Time series modelling of water resources and environmental systems. Electronic reprint of our book originally published in 1994. <http://www.stats.uwo.ca/faculty/aim/1994Book/>.
- Hollander M. & Wolfe D.A. (1973) *Nonparametric Statistical Methods*. John Wiley & Sons, New York, pp. 787.
- Holomuzki J.R. & Biggs J.F. (2000) Taxon-specific response to high flow disturbance in stream: implication for population persistence. *Journal of the North American Benthological Society*, **19**, 670–679.
- Illies J. (1978) *Limnofauna Europaea*. (Ed J. Illies), pp. 532. Gustav Fischer Verlag, Stuttgart, New York.
- IPCC (2001) *Climate Change 2001: Impacts, Adaptation and Vulnerability*. Intergovernmental Panel on Climate Change: Working Group II.
- Jackson J.K. & Füreder L. (2006) Long-term studies of freshwater macroinvertebrates: a review of the frequency, duration and ecological significance. *Freshwater Biology*, **51**, 591–603.
- Jáimez-Cuellar P., Vivas S., Bonada N., Robles S., Mellado A., Álvarez M. et al. (2002) Protocolo GUADALMED (PRECE). *Limnetica*, **21**, 187–204.

- Lake P.S. (2000) Disturbance, patchiness, and diversity in streams. *Journal of the North American Benthological Society*, **19**, 573–592.
- Lake P.S. (2003) Ecological effects of perturbation by drought in flowing waters. *Freshwater Biology*, **48**, 1161–1172.
- Lawrence J.E., Lunde K.B., Mazor R.D., Bêche L.A., McElravy E. & Resh V.H. (2010) Long-term macroinvertebrate responses to climate change: implications for biological assessment in mediterranean-climate streams. *Journal of the North American Benthological Society*, **29**, 1424–1440.
- Legendre P. & Legendre L. (1998) *Numerical Ecology*. 2nd English edn. Elsevier, Amsterdam, pp. 870.
- Lindh G. (1992). Hydrological and water resources impact of climate change. In: *Climatic Change and the Mediterranean: Environmental and Societal Impacts of Climatic Changes and Sea-Level Rise in the Mediterranean Region* (Eds L. Jeftic, J.D. Milliman & G. Sestini), pp. 58–93. Edward Arnold, London.
- McLeod A.I. (2011) Kendall: Kendall rank correlation and Mann-Kendall trend test. R package version 2.2. <http://CRAN.R-project.org/package=Kendall>.
- Milly P.C.D., Dunne K.A. & Vecchia A.V. (2005) Global pattern of trends in streamflow and water availability in a changing climate. *Nature*, **438**, 347–350.
- Mouthon J. & Daufresne M. (2006) Effects of the 2003 heat-wave and climatic warming on mollusc communities of the Saone: a large lowland river and of its two main tributaries (France). *Global Change Biology*, **12**, 441–449.
- Mulholland P.J., Roberts B.J., Hill W.R. & Smith J.G. (2009) Stream ecosystem responses to the 2007 spring freeze in the southeastern United States: unexpected effects of climate change. *Global Change Biology*, **15**, 1767–1776.
- Müller F., Baessler C., Frenzel M., Klotz S. & Schubert H. (2010) Long-term ecosystem research between theory and application – an introduction. In: *Long-Term Ecological Research between Theory and Application* (Eds F. Müller, C. Baessler, H. Schubert & S. Klotz), pp. 3–9. Springer, Dordrecht, Heidelberg, London, New York.
- Munné A. & Prat N. (2004) Defining river types in a Mediterranean area: a methodology for the implementation of the EU water framework directive. *Environmental Management*, **34**, 711–729.
- Munné A. & Prat N. (2011) Effects of Mediterranean climate annual variability on stream biological quality assessment using macroinvertebrate communities. *Ecological Indicators*, **11**, 651–662.
- Nahal I. (1981) The Mediterranean climate from a biological viewpoint. In: *Ecosystems of the World 11. Mediterranean-Type Shrublands*. (Eds F. di Castri, D.W. Goodall & R.L. Specht), pp. 63–86. Elsevier, Amsterdam.
- Nijboer R.C., Johnson R.K., Verdonschot P.F.M., Sommerhäuser M. & Buffagni A. (2004) Establishing reference conditions for European streams. *Hydrobiologia*, **516**, 91–105.
- Noges P., van de Bund W., Cardoso A.C., Solimini A.G. & Heiskanen A.S. (2009) Assessment of the ecological status of European surface. *Hydrobiologia*, **633**, 197–211.
- Ormerod S.J. & Durance I. (2009) Restoration and recovery from acidification in upland Welsh streams over 25 years. *Journal of Applied Ecology*, **46**, 164–174.
- Palmer T.N. & Raisanen J. (2002) Quantifying the risk of extreme seasonal precipitation events in a changing climate. *Nature*, **415**, 512–514.
- Peckarsky B.L., Taylor B.W. & Caudill C.C. (2000) Hydrologic and behavioral constraints on oviposition of stream insects: implications for adult dispersal. *Oecologia*, **125**, 186–200.
- Poquet J.M., Alba-Tercedor J., Punti T., Sánchez-Montoya M.D.M., Robles S., Álvarez M. *et al.* (2009) The MEDiterranean prediction and classification system (MEDPACS): an implementation of the RIVPACS/AUSRIVAS predictive approach for assessing Mediterranean aquatic macroinvertebrate communities. *Hydrobiologia*, **623**, 153–171.
- Prat N. & Rieradevall M. (2006) 25-years of biomonitoring in two mediterranean streams (Llobregat and Besòs basins, NE Spain). *Limnetica*, **25**, 541–550.
- R version. 2.13.1. The R Project for Statistical Computing. <http://www.r-project.org>. Last access date: 29/09/2012.
- Richter O., Suhling F., Muller O. & Kern D. (2008) A model for predicting the emergence of dragonflies in a changing climate. *Freshwater Biology*, **53**, 1868–1880.
- Ricklefs R.E. & Schluter D. (1993) *Species Diversity in Ecological Communities: Historical and Geographical Perspectives*. The University of Chicago Press, Chicago & London, pp. 414.
- Romesburg H.C. (1985) Exploring, confirming and randomization tests. *Computers and Geosciences*, **11**, 19–37.
- Rosenzweig C., Karoly D., Vicarelli M., Neofotis P., Wu Q., Casassa G. *et al.* (2008) Attributing physical and biological impacts to anthropogenic climate change. *Nature*, **453**, 353–357.
- Sala O.E., Chapin F.S., Armesto J.J., Berlow E., Bloomeld J., Dirzo R. *et al.* (2000) Global biodiversity scenarios for the year 2100. *Science*, **287**, 1770.
- Sánchez-Montoya M.M., Vidal-Abarca M.R., Punt T., Poquet J.M., Prat N., Rieradevall M. *et al.* (2009) Defining criteria to select reference sites in Mediterranean streams. *Hydrobiologia*, **619**, 39–54.
- Sevruk B. (1997) Regional dependency of precipitation-altitude relationship in the Swiss Alps. *Climatic Change*, **36**, 355–369.
- Shindell D. (2007) Estimating the potential for twenty-first century sudden climate change. *Philosophical Transactions of the Royal Society A*, **365**, 2675–2694.
- Shorthouse C. & Arnell N.W. (1997) Spatial and temporal variability in European river flows and the North Atlantic oscillation. In: *FRIEND 97- Regional Hydrology: Concepts and Models for Sustainable Water Resource Management* (Eds A. Gustard, S. Blazkova, M. Brilly, S. Demuth, J. Dixon, H. van Lanen, C. Llasat, S. Mkhani & E. Servat), pp.

- 77–85. IAHS Publication, Institute of Hydrology, Wallingford, Oxfordshire, UK.
- Suren A.M. & Jowett I.G. (2006) Effects of floods versus low flows on invertebrates in a New Zealand gravel-bed river. *Freshwater Biology*, **51**, 2207–2227.
- Ter Braak C.J.F. (1986) Canonical correspondence analysis: a new eigenvector technique for multivariate direct gradient analysis. *Ecology*, **67**, 1167–1179.
- The Nature Conservancy (2009) Indicators of Hydrologic Alteration. Version 7.1 User's Manual. <http://www.nature.org>. Last access date: 29/09/2012.
- Tierno de Figueroa J.M., Sanchez-Ortega A., Membiela Iglesias P. & Luzon-Ortega J.M. (2003) Plecoptera. In: *Fauna Ibérica*, vol. 22 (Ed. M.A. Ramos). pp. 404. Museo Nacional de Ciencias Naturales. CSIC. Madrid.
- Trivedi M.R., Berry P.M., Morecroft M.D. & Dawson T.P. (2008) Spatial scale affects bioclimate model projections of climate change impacts on mountain plants. *Global Change Biology*, **14**, 1089–1103.
- Viera-Lanero R. (2000) Las larvas de los Trichopteros de Galicia (Insecta: Trichoptera). PhD Thesis. Universidade de Santiago de Compostela. Facultade de Bioloxia. Departamento de Bioloxia Animal. Laboratorio de Hidrobioloxia.
- Wiens J.A. (2002) Predicting species occurrences: progress, problems, and prospects. In: *Predicting Species Occurrences: Issues of Accuracy and Scale* (Eds J.M. Scott, P.J. Heglund, M.L. Morrison, J.B. Haufler, M.G. Raphael, W.A. Wall & F.B. Samson), pp. 739–749. Island Press, Covelo, CA.
- Woodward G., Perkins D.M. & Brown L.E. (2010) Climate change and freshwater ecosystems: impacts across multiple levels of organization. *Philosophical Transactions of the Royal Society B*, **365**, 2093–2106.

Supporting Information

Additional Supporting Information may be found in the online version of this article:

Appendix 1 Mean abundance rank and occurrence of EPT (Ephemeroptera, Plecoptera and Trichoptera) collected during the study period of 14 years in the two streams and ANOVA results (P-value) for taxa collected between spring and summer.

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