

Loughborough University
Institutional Repository

*The influence of natural flow
regimes on
macroinvertebrate
assemblages in a semiarid
Mediterranean basin*

This item was submitted to Loughborough University's Institutional Repository by the/an author.

Citation: BELMAR, O. ... et al, 2013. The influence of natural flow regimes on macroinvertebrate assemblages in a semiarid Mediterranean basin. *Ecology*, 6 (3), pp. 363 - 379

Additional Information:

- This is the peer reviewed version of the following article: BELMAR, O. ... et al, 2013. The influence of natural flow regimes on macroinvertebrate assemblages in a semiarid Mediterranean basin. *Ecology*, 6 (3), pp. 363 - 379, which has been published in final form at <http://dx.doi.org/10.1002/eco.1274>. This article may be used for non-commercial purposes in accordance With Wiley Terms and Conditions for self-archiving.

Metadata Record: <https://dspace.lboro.ac.uk/2134/17358>

Version: Accepted for publication

Publisher: © John Wiley & Sons, Ltd.

Rights: This work is made available according to the conditions of the Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International (CC BY-NC-ND 4.0) licence. Full details of this licence are available at: <https://creativecommons.org/licenses/by-nc-nd/4.0/>

Please cite the published version.



The influence of natural flow regimes on macroinvertebrate assemblages in a semiarid Mediterranean basin.

Journal:	<i>Ecohydrology</i>
Manuscript ID:	ECO-11-0181.R1
Wiley - Manuscript type:	Research Article
Date Submitted by the Author:	10-Apr-2012
Complete List of Authors:	Belmar, Oscar; University of Murcia, Ecology and Hydrology Velasco, Josefa; University of Murcia, Ecology and Hydrology Gutierrez-Canovas, Cayetano; University of Murcia, Ecology and Hydrology Mellado, Andres; Centro de Estudios Hidrograficos CEDEX, Wood, Paul; Loughborough University, Geography;
Keywords:	natural flow regime, flow stability, minimum flows, macroinvertebrate composition, richness, Segura River Basin, semiarid Mediterranean streams

SCHOLARONE™
Manuscripts

View

1
2
3 **The influence of natural flow regimes on macroinvertebrate assemblages in a**
4 **semiarid Mediterranean basin.**
5
6

7
8 **O. Belmar*, J. Velasco*, C. Gutiérrez-Cánovas*, A. Mellado-Díaz**, A. Millán***
9 **P.J. Wood***.**
10

11 *** Department of Ecology and Hydrology. University of Murcia. Espinardo**
12 **Campus. 30100. Murcia (Spain).**
13

14 **** Centro de Estudios Hidrográficos CEDEX. Paseo Bajo de la Virgen del Puerto**
15 **3. 28005. Madrid (Spain).**
16

17 ***** Department of Geography. Loughborough University, Leicestershire, UK.**
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

1 Abstract

2 The investigation of flow-ecology relationships constitutes the basis for the
3 development of environmental flow criteria. The need to understand hydrology-ecology
4 linkages in natural systems has increased due to the prospect of climate change and flow
5 regime management, especially in water-scarce areas such as Mediterranean basins. Our
6 research quantified the macroinvertebrate community response at family, genus and
7 species level to natural flow regime dynamics in freshwater streams of a Mediterranean
8 semiarid basin (Segura River, SE Spain), and identified the flow components that
9 influence the composition and richness of biotic assemblages. Flow stability and
10 minimum flows were the principal hydrological drivers of macroinvertebrate
11 assemblages, whereas the magnitude of average and maximum flows had a limited
12 effect. Perennial stable streams were characterised by flow sensitive lotic taxa
13 (Ephemeroptera, Plecoptera, Tricoptera) and intermittent streams by predominately
14 lentic taxa (Odonata, Coleoptera, Heteroptera and Diptera). Relatively minor biological
15 changes were recorded for intermediate flow regime classes along a gradient of flow
16 stability. Seasonal variation and minimum flows are key hydrological components that
17 need to be considered for river management and environmental flows in the Segura
18 River Basin and other Mediterranean basins. The anthropogenic modification of these
19 parameters, due to both human activities and climate change, would probably lead to
20 significant changes in the structure and composition of communities in perennial stable
21 streams. This would be characterised by a reduction of flow sensitive EPT taxa and an
22 increase in more resilient OCHD taxa.

23
24 **Key words: natural flow regime, flow stability, minimum flows, macroinvertebrate**
25 **composition, richness, Segura River Basin, semiarid Mediterranean streams**

27 Introduction

28
29 The search for links between instream ecology and hydrology has become one of the
30 fundamental issues in contemporary river science (Vaughan *et al.*, 2009). Empirical
31 investigation of regional flow-ecology relationships constitutes the basis for the
32 development of environmental flow (e-flow) criteria (Arthington *et al.*, 2006; Poff *et*
33 *al.*, 2010). In addition, the need to understand ecology-hydrology linkages in natural
34 systems has been highlighted by the need to define reference conditions against which

1
2
3 1 modified dynamics can be compared (Tockner *et al.*, 2003). These needs are
4 2 particularly pressing in the light of predicted climate change (European Environment
5 3 Agency, 2008) and anthropogenic modification of natural flow regimes, especially in
6 4 water-scarce areas such as Mediterranean basins.
7
8
9

10 5
11 6 Instream hydrological variability, encapsulating elements of the entire flow regime such
12 7 as the daily, seasonal and annual patterns of discharge, the frequency, timing,
13 8 predictability and duration of extreme flows (high and low), rates of change in
14 9 discharge, and the magnitude of flows, are widely recognised as key ecological
15 10 organizers in fluvial ecosystems (Richter *et al.*, 1996; Poff *et al.*, 1997; Hart and Finelli,
16 11 1999; Bunn and Arthington, 2002). Spatial variation of these characteristics is
17 12 determined by variations in climate and mediated by basin geology, topography and
18 13 vegetation (Winter, 2001). These hydrological and environmental factors influence the
19 14 physical habitat for aquatic and riparian biota determining the conditions for
20 15 reproduction and recruitment and affecting the availability of trophic resources, refuges
21 16 during adverse situations and opportunities for dispersal (Naiman *et al.*, 2008).
22 17 Consequently, flow variability has strong ecological implications which shape the
23 18 structure and function of riverine ecosystems from the local to regional scales, and from
24 19 days (ecological effects) to millennia (evolutionary effects) (Lytle and Poff, 2004). It
25 20 has been hypothesised that sites with similar hydrological characteristics should share
26 21 similar faunal community composition, traits and ecosystem functioning (Poff and
27 22 Ward, 1989). Therefore, as Arthington *et al.* (2006) and Poff *et al.* (2010) suggested,
28 23 ecological responses of flow regimes to a given anthropogenic change should be
29 24 broadly similar in rivers with similar natural flow regimes.
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44

45 26 This hypothesis provides a powerful foundation to predict ecological responses to future
46 27 flow regime changes, constituting the key element of a new holistic framework for
47 28 developing scientifically-credible regional environmental flows: the ‘Ecological Limits
48 29 of Hydrologic Alteration’ (ELOHA) (Arthington *et al.*, 2006; Kennard *et al.*, 2010;
49 30 Poff *et al.*, 2010). Therefore, identifying and quantifying specific relationships between
50 31 flow regimes and biological communities in undisturbed river ecosystems are essential
51 32 steps to ensure sustainable river management (Arthington *et al.*, 2006; Jowett and
52 33 Biggs, 2009). Such relationships have been studied in general at the regional scale,
53 34 using macroinvertebrates (e.g. Monk *et al.*, 2006; Konrad *et al.*, 2008; Kennen *et al.*,
54
55
56
57
58
59
60

1
2
3 1 2010, Armanini *et al.*, 2011), fisheries (e.g Poff and Allan, 1995; Pegg and Pierce,
4 2002; Kennard *et al.*, 2007; Snelder *et al.*, 2009) or multiple taxonomic groups (e. g.
5 Jowett and Duncan, 1990; Clausen and Biggs, 1997). However, the strength and nature
6 of relationships between the flow regime and the biological assemblage vary depending
7 on the geographical region, the floral or faunal group considered and the taxonomic
8 resolution analysed.
9
10
11
12

13
14 7
15 8 In some areas, such as Mediterranean-climate regions, organisms have to withstand high
16 9 intra and interannual hydrological variability, together with frequent natural flow
17 10 extremes (floods and droughts) (Gasith and Resh, 1999). Species may respond over
18 11 evolutionary time scales by developing morphological, physiological and/or life-history
19 12 traits to bear such stresses (Poff *et al.*, 1997; Bonada *et al.*, 2007a; Bonada *et al.*,
20 13 2007b). Previous studies of Mediterranean streams (e. g. Bonada *et al.*, 2002; Jáimez-
21 14 Cuéllar *et al.*, 2002; Vivas *et al.*, 2002; Bonada *et al.*, 2004; Mellado, 2005; Sanchez-
22 15 Montoya *et al.*, 2007; Argyroudi *et al.*, 2009) as well as other semiarid areas (e. g.
23 16 Boulton and Lake, 2008) have highlighted the importance of flow permanence on the
24 17 composition and structure of macroinvertebrate communities. A progressive
25 18 replacement of Ephemeroptera, Plecoptera and Trichoptera (EPT) taxa by Odonata,
26 19 Coleoptera and Heteroptera (OCH) taxa has been reported as flow permanence
27 20 decreases (Sánchez-Montoya *et al.*, 2007; Argyroudi *et al.*, 2009) or hydrological
28 21 connectivity is reduced (Bonada *et al.*, 2006); although Diptera have also been
29 22 associated with river sections with low or no flows and dominate lentic habitats in
30 23 Southeast Spain (Vivas *et al.* 2002). Consequently, flow stability and hydrological
31 24 extremes (especially low flows) are expected to be the most important components of
32 25 Mediterranean flow regimes shaping instream assemblages, although its relative
33 26 importance is still unclear.
34
35
36
37
38
39
40
41
42
43
44
45
46
47

48 28 The aim of this study was to quantify the effect of different flow regimes on
49 29 macroinvertebrate communities. We utilised a dataset containing stream
50 30 macroinvertebrate records at family, genus and species level across a semiarid
51 31 Mediterranean region that encompasses a wide gradient of hydrological regimes
52 32 (Belmar et al 2011) to test these predictions: (1) Flow stability and minimum flows
53 33 should be the principal hydrological drivers of macroinvertebrate assemblage
54 34 composition and richness; (2) an increase in the explanatory power of hydrology should
55
56
57
58
59
60

1 occur as taxonomic resolution increases; and (3) a replacement of taxa should take place
2 along a hydrological gradient from permanent streams with stable discharges to streams
3 with high flow intermittence and flow variability. In general, a decrease in the
4 percentage of flow sensitive Ephemeroptera, Plecoptera and Trichoptera families should
5 occur as an increase in the percentage of more resilient Odonata, Coleoptera,
6 Heteroptera and Diptera families takes place.

7 8 **Methods**

9 *Study area*

10 Located in the Southeast of Spain, the Segura River Basin drainage network, including
11 coastal watercourses draining to the Mediterranean Sea, was selected as the study area.
12 The management area of the Segura River Basin, one of the most arid zones of the
13 Mediterranean region, includes watercourses with highly heterogeneous flow regimes.
14 These water-bodies range from perennial rivers, with low seasonal and interannual flow
15 variability, to highly seasonal ephemeral streams (Belmar *et al.*, 2011). This variability
16 is due to a strong climatic and altitudinal gradient from NW to SE, despite its relatively
17 small size (18 870 km²). Climate ranges from wet (>1 000 mm mean annual
18 precipitation) and cold in the high elevation mountains of the NW (>1 000 m.a.s.l.) to
19 semiarid and hot in the SE lowlands (< 350 mm mean annual precipitation). Mean
20 annual temperatures range between 10 and 18 °C (CHS, 2007). The lithology of the
21 plains is characterised by limestone (karst) and Miocene and Triassic marls, with some
22 small influences of volcanic strata. In contrast, calcites and dolomites dominate the
23 mountainous headwaters. The vegetation is varied and ranges from Mediterranean
24 conifer forests in the NW mountains to arid and semi-arid shrublands in the SE
25 lowlands. This gradient in altitude and climate is coupled with an anthropogenic
26 population density gradient. The river network has low population densities in the
27 forested headwaters, intermediate densities in the agricultural midlands (with major
28 flow regulation) and highly populated cities in the lowlands (Mellado, 2005).
29 Agricultural (52.1%), forest and semi natural (45.2%), and artificial (2.1%) are the
30 dominant landuses in the Segura basin (estimated from Corine Land Cover 2000),
31 making the Segura River Basin one of the most regulated in Europe (Ministerio de
32 Medio Ambiente MMA, 2004). Water resource demands exceed 224% of that available
33 and only 4% of runoff reaches the mouth of the river (Zimmer, 2010). This has resulted
34 in over exploitation of the surface waters, an inter-basin transfer from the Tagus River

1 (a mean of $325 \text{ hm}^3 \text{ yr}^{-1}$), a mean groundwater extraction of around $478 \text{ hm}^3/\text{year}$ (over
2 80% of natural recharge) and a high regulatory capacity of 770 hm^3 (over 90% of the
3 natural input) due to 24 dams over 10 m in height (Grindlay *et al.*, 2009; Grindlay *et al.*,
4 2011).

5 6 *Hydrological data*

7 A drainage network was derived from a 25 m digital elevation model (DEM) developed
8 by the *Instituto Geológico Nacional* (IGN) and layers available from the website of the
9 Spanish Ministry of Environment, using the ArcGIS software (v 9.2) and the ArcHydro
10 extension (v 1.2) (ESRI, Redlands, California, U.S.A.). The network comprises sections
11 that link each network junction or node, and each node was associated with its
12 corresponding watershed (derived from the DEM). The minimum watershed area to
13 define a river section was 10 km^2 , resulting a hydrological network with 390 river
14 sections.

15
16 The hydrological classification developed for the Segura River Basin in Belmar *et al.*
17 (2011) was used to define distinct natural hydrological regimes. This classification was
18 developed using 73 indices based on the “Indicators of Hydrologic Alteration” (IHA)
19 (Mathews and Richter, 2007). These flow indices represent a wide range of
20 ecologically-relevant flow statistics (Richter *et al.*, 1996; Olden and Poff, 2003; Monk
21 *et al.*, 2006; Mathews and Richter, 2007; Monk *et al.*, 2007) and comprise monthly and
22 annual flow statistics including measures of duration of droughts as well as the central
23 tendency and dispersion of flow magnitude (average, low and high flow conditions).
24 Indices related to the frequency, duration and rate of change of high flow events were
25 not used by Belmar *et al.* (2011) due to the absence of daily flow data. Natural flows
26 were derived from a monthly rainfall-runoff model developed by the Centre for
27 Hydrographic Studies (CEDEX, Ministry of Environment and Public Works, Spain), for
28 the period 1980/81 – 2005/06. The classification of the flow regimes recorded
29 comprised eight flow-regime classes (names are provided throughout to aid
30 interpretation) principally characterised by the magnitude of mean annual flow, the
31 duration of droughts and the interannual variation of flow (Table I). The resulting flow
32 regimes can be placed into four broad hydrological groups: (1) mainstem rivers, with
33 perennial flow thorough the year, low interannual variation and an average annual
34 discharge greater than $10 \text{ m}^3/\text{s}$ (class 1, *large rivers*) or between 2 and $10 \text{ m}^3/\text{s}$ (class 2,

1 *medium rivers*); (2) perennial stable streams, which only difference respect to mainstem
2 rivers is their reduced average discharge, between 0.3 and 2 m³/s (class 3, *creeks*) or
3 lower than 0.3 m³/s (class 4, *headwater streams*); (3) perennial seasonal streams, which
4 eventually cease flowing (although perennial surface water persists) and with peak
5 discharges in winter (class 5, *winter peak flow seasonal streams*) or spring (class 6,
6 *spring peak flow seasonal streams*); and (4) temporary streams, including *intermittent*
7 *streams* (class 7), which do not flow for between 20% and 50% of the time, and
8 *ephemeral streams*, that do not experience flow for more than 50% of the time (class 8).
9 Indices and classes were assigned to their corresponding river section.

11 *Macroinvertebrate data*

12 Macroinvertebrate abundance data at family, genus and species level were compiled
13 from the *Biodiversidad* database (*Ecología Acuática* research group, Department of
14 Ecology and Hydrology, University of Murcia, Spain). Species data were available for
15 beetles (Coleoptera), which have been recorded in all kinds of water bodies in the
16 region and have been shown to be good indicators of aquatic biodiversity (Bilton *et al.*,
17 2006; Sanchez-Fernandez *et al.*, 2006). Samples had been taken along 100 m stream
18 transects using a kick-net (500 – 1000 µm) and following the multi-habitat protocol
19 (Jáimez-Cuéllar *et al.*, 2002). Baseline macroinvertebrate samples were collected
20 between 1980 and 2006.

22 A minimum of 5 samples per hydrological class were selected, ensuring that they had
23 been collected in freshwater streams (conductivity < 5 000 µS cm⁻¹), above water
24 regulation infrastructures (e.g., dams or weirs) and abstraction areas and in absence of
25 significant evidences of anthropogenic alteration. However, using the criteria above two
26 classes did not have any biological data: *large rivers* (class 1), due to the absence of
27 reference conditions, and *ephemeral streams* (class 8), where no sampling had been
28 undertaken due to their frequent dry status.

30 Every sample was collected during the spring or early summer from a different
31 sampling site (Figure 1). This time-period is considered the most representative of the
32 annual macroinvertebrate community composition in Mediterranean streams (Bonada *et*
33 *al.*, 2009). Each site was paired with the closest downstream node in the drainage
34 network. In order to avoid pseudoreplication, when there was more than one site (and

1 sample) available for the same node, only the closest to the hydrological node was
2 selected. The final dataset consisted of 35 samples associated with 83 macroinvertebrate
3 families, and 133 genera, and 43 samples associated with 110 Coleoptera species
4 (Appendix A).

6 *Environmental data*

7 Climatic, topographic and geologic variables that were assumed to control hydrological
8 processes (Snelder *et al.*, 2005) were derived from different GIS layers available for the
9 watershed. Average annual precipitation and air temperature were derived from 1 km
10 grid maps created by the Spanish Ministry of Environment by means of interpolation
11 using data from the Spanish weather stations network (Estrela *et al.*, 1999). Drainage
12 area, mean altitude and slope were calculated using the IGN's digital elevation model
13 (DEM). Geology was characterised by the percentage of karst area in each watershed
14 and derived from the "Spain's Map of Karst" 1:1 000 000 developed by the *Instituto*
15 *Geológico y Minero de España* (IGME) and, indirectly, through water conductivity
16 (recorded for every biological sample). We hypothesised that the karstic surface would
17 control groundwater storage and baseflow (Snelder and Biggs, 2002) and that higher
18 conductivities would reflect the predominance of sedimentary marls that result in flashy
19 hydrographs that reflect precipitation patterns (Bracken *et al.*, 2008).

21 *Data analysis*

22 A Principal Component Factor Analysis (PCFA) (i.e. a Principal Components Analysis
23 (PCA) combined with a Varimax rotation) was used to examine dominant patterns of
24 intercorrelation among the hydrological indices (Belmar *et al.* 2011) and to identify
25 subsets of indices that describe the major sources of variation while minimizing
26 redundancy (i.e. multicollinearity). The Varimax rotation allows obtaining a clearer
27 pattern of loadings (indices clearly marked by high loadings for some axes and low
28 loadings for others) and, therefore, a better interpretation of the meaning of each axis.
29 The hydrological characteristics of each stream in the network were defined through the
30 corresponding PCFA scores (hydrological components) and hydrological class.

31
32 Rare taxa (those collected at fewer than 5% of sampling sites) were removed for
33 multivariate analyses. Abundance data were transformed by means of the Beals
34 smoothing function (Beals, 1984; McCune, 1994) to reduce noise by enhancing the

1
2
3 1 pattern of joint occurrences. This function is appropriate in the current investigation
4 because the data consist of a large number of small sample units (Peck *et al.*, 1995) and
5 fulfill the requirements established by De Cáceres and Legendre (2008).
6
7
8

9
10 5 For each taxonomic level analysed, we performed a non-metric multidimensional
11 scaling (NMDS) ordination based on Bray-Curtis distances among the sampling sites.
12 The strength of the correlation between the NMDS axes and the environmental
13 variables, as well as the hydrological components, was plotted as vectors. In addition,
14 the individual variables and components were analyzed using Pearson coefficients.
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

15 Distance based linear models (DistLM) were developed to assess the importance of
16 hydrological components driving taxonomical differences among sites. DistLM
17 calculates a multivariate multiple regression analysis between any symmetric distance
18 matrices, including a permutation test, as described by McArdle and Anderson (2001).
19 The final models were selected following a forward-stepwise procedure. For each
20 taxonomic level, marginal tests determined the variance explained by each flow
21 component and the sequential procedure discarded the variance shared by more than one
22 thereby avoiding the overestimation of their effect on the community.
23

24 Similarly, generalised linear models (GLM) were employed to determine how
25 hydrological components (independent variables) affected faunal richness patterns.
26 Models were constructed using log-transformed data following a forward-stepwise
27 procedure, assuming a Gaussian error distribution for the dependent variables. These
28 variables were the richness of Coleoptera species, number of macroinvertebrate genera,
29 number of macroinvertebrate families and the ratio EPT/EPTOCHD (defined by the
30 richness of Ephemeroptera, Plecoptera, Trichoptera, Odonata, Coleoptera, Heteroptera
31 and Diptera families). The latter is based on the EPT/EPTOCH ratio, which is used to
32 characterise temporary and lotic-lentic conditions in Mediterranean-climate regions
33 (Bonada *et al.*, 2006).
34

1 A non-metric single-factor Analysis of Similarity (ANOSIM) was used to test whether
2 assemblage composition differed among hydrological classes and, therefore, if natural
3 regimes can be used to differentiate distinct groups of invertebrate communities. Global
4 R indicates if assemblages are randomly grouped (i.e., $R=0$) or not (usually $0 < R \leq 1$,
5 although negative values are possible *sensu* Clarke (1993)). R pairwise values were also
6 obtained for each pair of classes, indicating whether intra-class similarities were greater
7 than inter-class similarities (R value close to 1).

8
9 Indicator taxa were defined for each hydrological class using the Indicator Species
10 analysis (IndVal) of Dufrene & Legendre (1997). This analysis generates an indicator
11 value index (IV) for each taxon and class, calculated on the basis of the specificity
12 (maximum when a taxon only occurs in one class) and fidelity (maximum when all sites
13 in a class have the taxon) of each taxon to each class.

14
15 All permutation tests (DistLM, ANOSIM and IndVal) were undertaken using 999
16 permutations. PCFA was undertaken in STATISTICA v 6 (Statsoft, 2001). NMDS and
17 IndVal were conducted using PC-ORD software v 4.42 (McCune and Grace, 2002).
18 ANOSIM and DistLM were undertaken in PRIMER v6 (Clarke and Gorley, 2006).
19 GLM were performed using the R statistical software v 2.12.2 (R Development Core
20 Team, 2008).

21 **Results**

22 *Hydrological components*

23 The three first PCFA axes were selected to represent the set of hydrological indices
24 since all of them explained greater than 10% of the variance (46, 28 and 12%,
25 respectively) and the forth axis only explained an additional 4%. The first axis was
26 positively correlated with mean and maximum monthly flows (Table IIa), representing
27 the flow magnitude component of the IHA. The second axis was negatively correlated
28 with the inter-annual coefficients of variation in monthly flows, the intra-annual
29 coefficient of variation in maximum monthly flows and the percentage of time without
30 flows. These variables characterise the inter- and intra-annual variability of the flow
31 regime and as a result this axis was defined as the flow stability component (Table IIb).
32 The third axis, magnitude of minimum flows, was correlated with all the minimum
33 monthly flows and their average value (Table IIc).
34

1
2
3 1
4 2 These three hydrological components (PCFA axes) displayed significant positive
5 3 correlations with mean altitude and precipitation in the watershed, and negative
6 4 correlations with mean temperature (Table III). In addition, karst surface and slope were
7 5 positively correlated with flow stability and minimum flows, while drainage area was
8 6 associated with the magnitude of flow. As anticipated, conductivity displayed a negative
9 7 association with flow magnitude and stability.
10 8

16 9 *Hydrological components determining assemblage composition*

17 10 The macroinvertebrate NMDS ordinations for different taxonomic resolutions identified
18 11 similar patterns (Figure 2). Sites were structured along a flow stability gradient from
19 12 perennial headwater streams (left side, class 4) to intermittent streams (right side, class
20 13 7), although some classes were widely dispersed (particularly class 6 - spring peak flow
21 14 seasonal streams). This gradient was associated with several environmental variables
22 15 and hydrological components (PCFA axes). Perennial stable streams (classes 3 and 4)
23 16 were predominately located on karstic rocks and sites in higher altitude areas with
24 17 steeper slopes, higher flow stability and relatively high minimum flows. In contrast,
25 18 intermittent streams were associated to low slopes, reduced flow stability and low
26 19 minimum flows, but higher conductivity and air temperature.
27 20

28 21 DistLM models indicated that hydrological components accounted for a significant
29 22 proportion of the variance in the macroinvertebrate community that increased with
30 23 taxonomic resolution (Table IV): 28% for families, 30% for genus and 38% for
31 24 Coleoptera species. In all cases, flow stability and minimum flows were the dominant
32 25 hydrological drivers of taxonomical differences among sites.
33 26

36 27 *Response of taxonomic richness to hydrological components*

37 28 GLM results showed a moderate effect of hydrological variables on the richness of
38 29 macroinvertebrate families, genera and species (Table V). However, the model obtained
39 30 for the EPT/EPTOCHD ratio explained 36 % of the variance using flow magnitude and
40 31 flow stability as independent variables. Gradual changes to the relative richness of EPT
41 32 families were observed from perennial to intermittent hydrological classes, decreasing
42 33 along the flow magnitude gradient, whilst the OCHD families displayed the opposite
43 34 pattern (Figure 3).
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47 48 49 50 51 52 53 54 55 56 57 58 59 60

1
2 *Differences in assemblage composition among hydrological classes*

3 The hydrological classes identified supported significantly different invertebrate
4 assemblages at the family (ANOSIM, $R = 0.39$; P -value < 0.05), genus (ANOSIM, $R =$
5 0.34 ; P -value < 0.05) and species taxonomic level (ANOSIM, $R = 0.40$; P -value < 0.05)
6 (Table VI). Pair-wise comparisons revealed significant assemblage differences at all
7 taxonomic resolutions between the extremes of the hydrological gradient, perennial
8 stable streams (creeks and headwaters, classes 3 and 4 respectively) and intermittent
9 streams (class 7). Differences between creek and medium river communities (class 2) as
10 well as between creeks and perennial seasonal streams with peak flows during the
11 winter (class 5) increased with the taxonomic resolution, except for the genus level.
12 However, intermittent streams and perennial seasonal streams, both with winter (class
13 5) and spring peak flows (class 6), differed at the genus or at the genus and species
14 levels, respectively. No significant differences were found both between creeks and
15 headwater streams or within seasonal streams (winter and spring peak flows) (Table
16 VI).

17
18 The IndVal analyses determined indicator families for medium rivers (class 2),
19 headwater streams (class 4), spring peak flow seasonal streams (class 6) and intermittent
20 streams (class 7) (Table VII). Medium rivers were characterised by Polycentropodidae
21 (Trichoptera) and Potamanthidae (Ephemeroptera). Headwater streams were defined by
22 one Ephemeroptera (Leptophlebiidae), five families of Trichoptera (Limnephilinae and
23 Beraeidae showed slightly higher Indicator Values) and one Crustacea (Astacidae).
24 Spring peak flow seasonal streams were characterised by Syrphidae (Diptera), which
25 presented the highest Indicator Value in the Segura Basin. Intermittent streams were
26 defined by the presence of Coenagrionidae and Libellulidae (Odonata), Pleidae
27 (Heteroptera) and Noteridae and Hydrophilidae (Coleoptera).

28
29 Indicator genera were found for all classes except creeks (class 3) and winter peak flow
30 seasonal streams (class 5). Medium rivers (class 2) and headwater streams (class 4)
31 were characterised by Ephemeroptera: *Habrophlebia* and *Potamanthus* for the former
32 and *Epeorus* and *Rhithrogena* for the latter. Headwaters were also characterised by
33 seven Coleoptera genera (*Oreodytes*, *Graptodytes*, *Esolus*, *Limnebius*, *Normandia*,
34 *Hydrocyphon* and *Oulimnius*), two Trichoptera (*Rhyacophila* and *Sericostoma*), one

1 Crustacea (*Austropotamobius*) and two Plecoptera (*Perla* and *Isoperla*). Spring peak
2 flow seasonal streams (Class 6) were characterised by one genus of Coleoptera
3 (*Dytiscus*), Hirudinea (*Helobdella*), Mollusca (*Pseudamnicola*) and Odonata
4 (*Platynemis*), with identical indicator values. Intermittent streams (class 7) highlighted
5 the highest number of indicator genera, with the highest Indicator Values for two
6 Diptera (*Dasyhelea* and *Anopheles*), two Heteroptera (*Heliocorisa* and *Anisops*), two
7 Odonata (*Anax* and *Sympetrum*) and two Coleoptera (*Enochrus* and *Berosus*).

8
9 Coleoptera indicator species were detected for all classes except spring peak flow
10 seasonal streams (class 6) (Table VII). Medium rivers (class 2) were primarily
11 characterised by *Hydraena manfredjaechi* and *Normandia nitens*; creeks (class 3) by
12 *Hydraena exasperata*; headwater streams (class 4) by *Helophorus alternans*; winter
13 peak flow seasonal streams (class 5) by *Eretes griseus* and *Ranthus suturalis*; and
14 intermittent streams (class 7) by *Ochthebius delgadoi*.

15 16 **Discussion**

17 *The importance of hydrological components on macroinvertebrate assemblages*

18 The research presented herein supports the general hypothesis that streams with similar
19 flow regimes express greater than random similarity in macroinvertebrate assemblages
20 composition (Resh *et al.*, 1988; Poff, 1996). Our results demonstrate relatively strong
21 relationships between community composition and the flow regimes at different
22 taxonomic levels. The strength of these relationships increased with taxonomic
23 resolution suggesting that the species level data yields the strongest relationships and
24 that, where it is available, it should be used in ecohydrological investigations (Monk *et al.*,
25 2012). Flow stability and minimum flows were shown to be the principal
26 hydrological drivers/descriptors of the macroinvertebrate community assemblages in the
27 Segura River Basin. Similar results were reported by Chinnayakanehalli *et al.* (2011) in
28 western USA, where baseflows and seasonality were the main predictors of invertebrate
29 composition. However, these results contrast with studies performed in temperate-
30 maritime regions where the magnitudes of mean flows or high flows were reported to be
31 the best predictors of macroinvertebrate assemblages (Clausen and Biggs, 1997; Monk
32 *et al.*, 2006; Monk *et al.*, 2008).

1
2
3 1 Flow stability and minimum flows are major determinants of habitat availability and
4 2 connectivity that affect aquatic macroinvertebrate assemblages. Flow stability reflects
5 3 seasonal and interannual patterns of variation, associated with the predictability of flows
6 4 (Poff, 1996) and the stability of habitat conditions in terms of depth, flow velocity and
7 5 hydraulic forces (Suen and Herricks, 2009). The variation of stream flow velocity
8 6 configures stream morphology, water temperature, bed stability and consequently the
9 7 availability of aquatic habitats for instream organisms (Jowett and Duncan, 1990).
10 8 Minimum flows represent an extreme of the flow, particularly in the dry season, and
11 9 reflect the magnitude of seasonal droughts (Smakhtin, 2001). Habitat heterogeneity is
12 10 reduced under low flow conditions because wetted width, water depth and flow velocity
13 11 also diminishes (Walters and Post, 2011). In addition, extreme low flows can reduce
14 12 longitudinal connectivity and increase physical stresses transforming streams into series
15 13 of isolated pools with higher water temperature and elevated conductivity (Stanley *et al.*,
16 14 1997). Consequently, droughts have been recognised as an important part of the
17 15 natural flow regime in intermittent streams (Boulton, 2003; Lake, 2003; Sheldon and
18 16 Thoms, 2006, Chase, 2007). Species inhabiting intermittent streams must have
19 17 physiological, behavioural or life-history adaptations to cope with higher conductivities,
20 18 predation pressures and habitat isolation, such as short life-histories, generalist feeding,
21 19 aerial respiration or active aerial dispersal (e.g. Bonada *et al.*, 2007b). Under these
22 20 conditions, dispersal abilities and distances between or along water bodies have been
23 21 found to be primary determinants of community composition (McAbendroth *et al.*,
24 22 2005), because active movement when the riverbed is dry is limited to a small number
25 23 of taxa such as dytiscid and hydrophilid beetles (Boulton *et al.*, 2006; Larned *et al.*,
26 24 2010).

25
26 26 Our results indicate a moderately strong relationship between flow regime and faunal
27 27 richness at the different taxonomic resolutions, weaker than that between flow regime
28 28 and community composition (especially at species level). Other studies have also
29 29 reported a moderate effect of minimum flows (Walters and Post, 2011), flow seasonality
30 30 or the number of days with zero flow (Chinnayakanahalli *et al.*, 2011).

31
32 32 In Mediterranean regions, ephemeral and intermittent streams are recognised to be
33 33 significantly less diverse than perennial streams (Bonada *et al.*, 2007b) and to differ in
34 34 community composition (e.g. Bonada *et al.*, 2006; Argyroudi *et al.*, 2009). Our results

1 found a strong relationship between flow magnitude, and stability, and the ratio of
2 EPT/EPTOCHD. This supports the findings of Bonada *et al.* (2006) and Sánchez-
3 Montoya *et al.* (2007), who reported a decrease in EPT richness as hydrological
4 isolation and the length of the dry period (temporality) increased. EPT taxa in particular
5 tend to occur in riffles, whereas pools support the majority of OCHD taxa (Vivas *et al.*,
6 2002; Oscoz *et al.*, 2011). Therefore, riffle permanence has a strong effect on the
7 structure of benthic assemblages in streams (Feminella, 1996).

8 9 *Biological significance of hydrological classes*

10 The six hydrological classes examined in this study indicate distinct macroinvertebrate
11 assemblages at all of the taxonomic resolutions considered. Taxonomic differences were
12 greatest between the classes at both extremes of the flow stability gradient, and are
13 similar to results reported by other studies in the Iberian Peninsula (Sanchez-Montoya *et al.*
14 *et al.*, 2007) and in the Segura Basin (Millan *et al.*, 2006; Diaz *et al.*, 2008; Carbonell *et al.*
15 *et al.*, 2011). However, when the other classes were considered, only minor and gradual
16 biological changes along the gradient were detected. Consequently, a simpler
17 classification with four broad hydrological types (Belmar *et al.* 2011) is more
18 appropriate for management purposes in the Segura River Basin and other semi-arid
19 Mediterranean basins: (1) mainstream rivers (classes 1 and 2), (2) perennial stable
20 streams (classes 3 and 4), (3) perennial seasonal streams (classes 5 and 6) and (4)
21 temporary streams (classes 7 and 8).

22
23 We found a clear agreement between the selection of indicator taxa in this study and
24 those from other studies in the Mediterranean region in Spain (e.g., Bonada *et al.*, 2004;
25 Mellado, 2005; Sanchez-Montoya *et al.*, 2007). Headwater streams were characterised
26 by taxa that inhabit the upper reaches of rivers with colder and oxygen-rich waters, in
27 areas of cobbles and small boulders. These sites supported the greatest presence of
28 Ephemeroptera (Leptophlebiidae) and Trichoptera (e.g. Limnephilinae and Beraidae)
29 families and were also characterised by the presence of typically reophilic
30 Ephemeroptera (*Epeorus* and *Rhitrogena*) and Plecoptera (*Perla* and *Isoperla*) genera.
31 In general, these taxa are considered to have high oxygen requirements and their
32 presence is associated with good water quality (Jacobsen *et al.*, 2003). Medium rivers
33 were characterised by Ephemeroptera genera, such as *Potamanthus* and *Habrophlebia*,
34 typical of reaches of large rivers where low to moderate flow velocities, associated with

1 gravel and sand substrates, predominate (Puig *et al.*, 1984). Intermittent streams were
2 associated with taxa from shallow standing waters or those with reduced velocities, such
3 as numerous Coleoptera (e.g., *Enochrus*, *Berosus* and *Noterus*), Odonata (e.g., *Anax*,
4 *Sympetrum* and *Ischnura*) and Heteroptera (e.g., *Heliocoris*, *Anisops* and *Sigara*),
5 with highly mobile adults (Bilton *et al.*, 2001) and short life-history development times
6 (Velasco *et al.*, 1990; Barahona *et al.*, 2005). The importance of Coleoptera in
7 temporary streams highlighted in this study has also been demonstrated in several
8 previous studies (Picazo *et al.*, 2012).

10 *Implications to river restoration and conservation*

11 Based on the results presented, the magnitude of monthly minimum flows and the inter-
12 and intra-annual natural variation of flows are two key flow components for the
13 definition of environmental flows in Mediterranean basins. Currently, many historically
14 perennial streams have already become intermittent due to excessive abstraction and
15 impoundment, while others exhibit an inverse seasonal pattern due to water release from
16 reservoirs during the summer months (Belmar *et al.*, 2010). Such hydrological
17 modifications could become more intense in the future as a result of climate change
18 (European Environment Agency, 2008), which is expected to intensify supra-seasonal
19 droughts and lead to more anthropogenic water withdrawals. This may lead to the
20 depletion of groundwater in local aquifers and, therefore, flow intermittency in
21 previously perennial streams. Such intermittency could result in significant changes to
22 the faunal community, increasing the risk of local extinctions of drought-sensitive taxa.
23 This effect has already been documented in desert streams (Bogan and Lytle, 2011),
24 where simplified pools composed of the most tolerant and resilient species have been
25 described (*sensu* Cote and Darling, 2010). Therefore, the conservation and, where
26 appropriate, restoration of natural hydrological variability is crucial for the maintenance
27 of riverine ecosystem integrity (i.e., ecosystem structure and function) (Thoms, 2006;
28 Vaughan *et al.*, 2009).

30 Future research should focus on how the degree of hydrological alteration affects
31 aquatic communities and ecosystem functioning. Aquatic macroinvertebrates are ideal
32 candidates for the development of hydro-ecological models to quantify the effects of
33 flow reduction (Castella *et al.*, 1995; Niu and Dudgeon, 2011a; Niu and Dudgeon,
34 2011b). Using the four broad hydrological types stated we will be able to provide a

1
2
3 1 reference framework in the near future to achieve a more sustainable management of
4 2 ecohydrological resources in the Segura River Basin and other Mediterranean basins,
5 3 fulfilling the objectives of ELOHA and EU Water Framework Directive criteria.
6
7
8 4

9
10 5 Acknowledgements

11 6 We wish to thank the University of Murcia and the *Fundación Séneca* for their financial
12 7 support to Óscar Belmar and Cayetano Gutiérrez-Cánovas, respectively, by means of
13 8 pre-doctoral grants; the Euromediterranean Institute of Water for its support to the
14 9 project “Hydrological classification of the rivers and streams in the Segura basin and
15 10 associated macroinvertebrate communities”; the *Confederación Hidrográfica del*
16 11 *Segura* for providing the climatic data and the SIMPA model; and the *Instituto*
17 12 *Geográfico Nacional* as well as the *Instituto Geológico y Minero de España* for the GIS
18 13 data.
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

Table I. Mean and standard deviation of the mean annual flow (MADIS), time with zero flow (D_L) and coefficient of variation in annual flows (CV_{INTER}) for the natural flow regime classes defined in the Segura River Basin (Belmar *et al.* 2011).

Hydrological class	Number of stream sections	MADIS (m^3/s)	D_L (%)	CV_{INTER}
Class 1: Perennial large size rivers	17	11.30 (± 0.74)	0.00 (± 0.00)	0.52 (± 0.01)
Class 2: Perennial medium size rivers	31	3.76 (± 2.26)	0.00 (± 0.00)	0.50 (± 0.13)
Class 3: Perennial stable creeks	21	1.00 (± 0.45)	0.00 (± 0.00)	0.32 (± 0.09)
Class 4: Perennial stable headwater streams	43	0.18 (± 0.17)	0.00 (± 0.00)	0.26 (± 0.13)
Class 5: Perennial winter peak flow seasonal streams	26	0.37 (± 0.09)	2.31 (± 2.06)	1.39 (± 0.29)
Class 6: Perennial spring peak flow seasonal streams	110	0.06 (± 0.06)	4.46 (± 6.32)	0.81 (± 0.30)
Class 7: Temporary intermittent streams	101	0.04 (± 0.04)	24.88 (± 13.15)	1.71 (± 0.38)
Class 8: Temporary ephemeral streams	41	0.01 (± 0.01)	61.90 (± 20.21)	3.43 (± 0.84)

Table II. Pearson correlation coefficients between the three rotated PCFA axes and the 73 hydrological indices. Coefficients higher than |0.70| are in bold letter. Horizontal lines separate indices associated to the three flow components represented by the axes: (a) magnitude (average and maximum flows), 1st axis (46% of variance); (b) flow stability, 2nd axis (28% of variance); and (c) minimum flows, 3rd axis (12% of variance).

Variable	Description	PCFA axis			
		1 st	2 nd	3 rd	
(a)	M_A 1	Mean monthly flow (October)	0.98	0.13	0.02
	M_A 2	Mean monthly flow (November)	0.98	0.13	0.02
	M_A 3	Mean monthly flow (December)	0.99	0.12	0.05
	M_A 4	Mean monthly flow (January)	0.98	0.14	0.04
	M_A 5	Mean monthly flow (February)	0.98	0.14	0.04
	M_A 6	Mean monthly flow (March)	0.99	0.12	0.03
	M_A 7	Mean monthly flow (April)	0.98	0.14	0.02
	M_A 8	Mean monthly flow (May)	0.98	0.15	0.04
	M_A 9	Mean monthly flow (June)	0.98	0.15	0.03
	M_A 10	Mean monthly flow (July)	0.97	0.16	0.04
	M_A 11	Mean monthly flow (August)	0.97	0.16	0.05
	M_A 12	Mean monthly flow (September)	0.98	0.13	0.02
	M_A 16	Mean annual flow divided by catchment area	0.18	0.50	0.35
	MEDDIS/A	Median annual discharge divided by catchment area	0.22	0.52	0.35
	M_H 1	Mean of the maximum monthly flows (October)	0.96	0.08	0.01
	M_H 2	Mean of the maximum monthly flows (November)	0.96	0.06	0.07
	M_H 3	Mean of the maximum monthly flows (December)	0.91	0.00	0.05
	M_H 4	Mean of the maximum monthly flows (January)	0.97	0.14	0.08
	M_H 5	Mean of the maximum monthly flows (February)	0.97	0.15	0.11
	M_H 6	Mean of the maximum monthly flows (March)	0.94	0.03	0.02
	M_H 7	Mean of the maximum monthly flows (April)	0.98	0.10	0.04
	M_H 8	Mean of the maximum monthly flows (May)	0.98	0.15	0.08
	M_H 9	Mean of the maximum monthly flows (June)	0.98	0.13	0.00
	M_H 10	Mean of the maximum monthly flows (July)	0.98	0.13	-0.03
	M_H 11	Mean of the maximum monthly flows (August)	0.98	0.13	-0.03
	M_H 12	Mean of the maximum monthly flows (September)	0.95	0.05	-0.04
	M_H 13	Mean of the mean maximum flows for all months	0.98	0.08	0.04
	MADIS	Mean annual flow for all years	0.98	0.14	0.03
	RANGE	Maximum annual discharge minus minimum annual discharge	0.98	0.06	-0.05
	Q1	Percentile flow with the annual discharge exceeded 1% of time	0.99	0.09	0.01
	Q50	Median annual flow for all years	0.97	0.14	0.03
(b)	CV_A 1	Coefficient of variation (October)	-0.08	-0.83	-0.30
	CV_A 2	Coefficient of variation (November)	-0.12	-0.86	-0.15
	CV_A 3	Coefficient of variation (December)	-0.09	-0.84	-0.19
	CV_A 4	Coefficient of variation (January)	-0.19	-0.88	-0.21
	CV_A 5	Coefficient of variation (February)	-0.21	-0.89	-0.17
	CV_A 6	Coefficient of variation (March)	-0.19	-0.81	-0.25
	CV_A 7	Coefficient of variation (April)	-0.26	-0.90	-0.20
	CV_A 8	Coefficient of variation (May)	-0.02	-0.91	-0.19
	CV_A 9	Coefficient of variation (June)	0.02	-0.83	-0.35
	CV_A 10	Coefficient of variation (July)	0.09	-0.82	-0.37
	CV_A 11	Coefficient of variation (August)	0.09	-0.84	-0.36
	CV_A 12	Coefficient of variation (September)	-0.03	-0.81	-0.34
	M_A 13	Range divided by median monthly flow	-0.06	-0.90	-0.03
	M_A 14	Interquartile divided by median monthly flow	0.09	-0.80	0.05
	CV_{INTRA}	Coefficient of variation in mean monthly flows	0.02	-0.90	-0.03
	M_A 15	Mean minus median monthly flow divided by median monthly flow	-0.15	-0.73	0.06
	M_A 17	Range divided by median annual flow	-0.22	-0.93	-0.10
	M_A 18	Interquartile divided by median annual flow	-0.17	-0.83	-0.05
	M_A 19	Mean minus median annual flow divided by median annual flow	-0.17	-0.84	0.03
	CV_H	Coefficient of variation in mean maximum monthly flows	-0.27	-0.79	-0.08
	D_L	Percentage of months with zero flow	-0.38	-0.75	-0.24
	CV_{INTER}	Coefficient of variation in annual flows for all years	-0.21	-0.92	-0.25
	Q5/Q50	Q5 divided median monthly flow	-0.23	-0.88	-0.08
	Q10/Q50	Q10 divided median monthly flow	-0.21	-0.87	-0.06
	STDEV	Standard deviation of annual discharge	0.99	0.07	-0.09
	AMAX/Q50	Maximum annual discharge divided by Q50	-0.23	-0.92	-0.08
	AMIN/Q50	Minimum annual discharge divided by Q50	-0.25	0.63	0.42
	I_H	Q5 divided mean monthly flow	0.08	-0.04	-0.27
	I_L	Q95 divided mean monthly flow	-0.26	0.60	0.48
(c)	M_L 1	Mean minimum monthly flow (October)	0.02	0.19	0.92
	M_L 2	Mean minimum monthly flow (November)	0.04	0.19	0.92
	M_L 3	Mean minimum monthly flow (December)	0.03	0.19	0.92
	M_L 4	Mean minimum monthly flow (January)	0.11	0.20	0.77
	M_L 5	Mean minimum monthly flow (February)	0.08	0.18	0.88
	M_L 6	Mean minimum monthly flow (March)	0.04	0.18	0.93
	M_L 7	Mean minimum monthly flow (April)	0.10	0.23	0.78
	M_L 8	Mean minimum monthly flow (May)	0.03	0.17	0.93
	M_L 9	Mean minimum monthly flow (June)	0.00	0.17	0.90
	M_L 10	Mean minimum monthly flow (July)	0.01	0.17	0.90
	M_L 11	Mean minimum monthly flow (August)	0.04	0.16	0.89
	M_L 12	Mean minimum monthly flow (September)	0.05	0.16	0.88
	M_L 13	Mean of the mean minimum flows for all months	0.06	0.20	0.96

Table III. Pearson correlation coefficients between environmental variables and hydrological components (PCFA axes). Significant correlations ($p < 0.05$) are in bold letter.

Environmental variable	Flow magnitude	Flow stability	Minimum flows
Mean precipitation (mm)	0.26	0.64	0.39
Conductivity ($\mu\text{S}/\text{cm}^2$)	-0.28	-0.54	-0.21
Mean altitude (m)	0.34	0.64	0.34
Mean slope ($^\circ$)	0.24	0.37	0.27
Karst surface (%)	0.21	0.36	0.37
Mean temperature ($^\circ\text{C}$)	-0.37	-0.57	-0.27
Drainage area (km^2)	0.83	-0.16	-0.14

For Peer Review

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49

Table IV. Results of the DistLM analyses for each taxonomic level. Significance levels are indicated with asterisks (*: p≤0.05; **: p≤0.01; ***: p≤0.001).

Hydrological component	Macroinvertebrate families		Macroinvertebrate genera		Coleoptera species	
	Marginal (%)	Sequential (%)	Marginal (%)	Sequential (%)	Marginal (%)	Sequential (%)
Flow magnitude	7	6*	6	5	4	3*
Flow stability	12***	9**	24***	24***	27***	27***
Minimum flows	13**	13***	11**	6*	16***	8***
Total (%)		28		30		38

For Peer Review

Table V. GLM analyses for the different dependent variables, based on richness. Significance levels are indicated with asterisks (*: $p \leq 0.05$; **: $p \leq 0.01$; ***: $p \leq 0.001$).

Dependent variable	Variance explained (%)	Explanatory hydrological components
EPT/EPTOCHD	36	Flow magnitude**, flow stability*
Macroinvertebrate families	21	Minimum flows**
Macroinvertebrate genera	24	Minimum flows**
Coleoptera species	17	Minimum flows**

For Peer Review

Table VI. Results of ANOSIM analyses. Significance levels are indicated with asterisks (*: $p \leq 0.05$; **: $p \leq 0.01$; ***: $p \leq 0.001$).

Classes	Macroinv. families	Macroinv. genera	Coleop. species
2, 5	0.22	0.15	0.50**
2, 7	0.59*	0.62*	0.49**
3, 2	0.26*	0.06	0.56**
3, 5	0.25*	0.20	0.76***
3, 6	0.49**	0.30*	0.05
3, 7	0.50**	0.53**	0.86***
4, 2	0.85**	0.67**	0.37**
4, 3	0.00	-0.02	-0.09
4, 5	0.81**	0.80**	0.66***
4, 6	0.53**	0.77**	0.09
4, 7	0.88**	0.86**	0.77***
5, 7	0.29	0.42*	0.17
6, 2	0.27**	0.33**	-0.01
6, 5	0.12	0.02	0.16
6, 7	0.09	0.44*	0.38**
Global R	0.39***	0.34***	0.40***

Class 2: Perennial medium rivers
 Class 3: Perennial stable creeks
 Class 4: Perennial stable headwater streams
 Class 5: Perennial winter peak flow seasonal streams
 Class 6: Perennial spring peak flow seasonal streams
 Class 7: Temporary intermittent streams

Table VII. Indicator taxa ($IV \geq 25$ & $p \leq 0.05$) for each hydrological class and taxonomic level.

Hydrological class	Macroinvertebrate families	IV (%)	Macroinvertebrate genera	IV	Coleoptera species	IV (%)
2. Perennial medium rivers	Polycentropodidae	31	<i>Habrophlebia</i>	28	<i>Hydraena manfredjaechi</i>	47
	Potamanthidae	27	<i>Potamanthus</i>	27	<i>Normandia nitens</i>	47
					<i>Limnius intermedius</i>	44
					<i>Ochthebius difficilis</i>	34
					<i>Limnius opacus</i>	28
3. Perennial stable creeks					<i>Pomatinus substriatus</i>	25
					<i>Hydraena exasperata</i>	55
					<i>Ilybius meridionalis</i>	50
					<i>Ochthebius bellieri</i>	46
					<i>Limnius volckmari</i>	34
					<i>Agabus brunneus</i>	32
					<i>Hydroporus marginatus</i>	30
					<i>Ochthebius bonnairei</i>	30
					<i>Anacaena bipustulata</i>	29
					<i>Deronectes moestus</i>	29
					<i>Hydraena carbonaria</i>	29
					<i>Hydraena capta</i>	27
					<i>Hydraena rufipennis</i>	26
4. Perennial stable headwater streams					<i>Stictionectes epipleuricus</i>	26
					<i>Agabus didymus</i>	25
	Leptophlebiidae	41	<i>Oreodytes</i>	45	<i>Helophorus alternans</i>	29
	Limnephilinae	29	<i>Epeorus</i>	35	<i>Helophorus brevipalpis</i>	28
	Beraeidae	29	<i>Rhyacophila</i>	31	<i>Laccobius obscuratus</i>	28
	Brachycentridae	28	<i>Graptodytes</i>	30	<i>Hydroporus tessellatus</i>	26
	Rhyacophilidae	27	<i>Austropotamobius</i>	30	<i>Limnebius cordobanus</i>	26
	Sericostomatidae	26	<i>Esolus</i>	29		
	Astacidae	26	<i>Sericostoma</i>	29		
			<i>Limnebius</i>	28		
			<i>Normandia</i>	27		
			<i>Hydrocyphon</i>	27		
			<i>Rhithrogena</i>	27		
			<i>Oulimnius</i>	25		
	5. Perennial winter peak flow seasonal streams			<i>Perla</i>	25	
			<i>Isoperla</i>	25		
					<i>Eretes griseus</i>	76
					<i>Rhantus suturalis</i>	76
					<i>Hydrochus nooreinus</i>	52
					<i>Stictotarsus duodecimpustulatus</i>	52
					<i>Berosus hispanicus</i>	34
6. Perennial spring peak flow seasonal streams					<i>Hydrophilus pistaceus</i>	32
					<i>Laccobius moraguesi</i>	31
					<i>Agabus ramblae</i>	29
	Syrphidae	85	<i>Dytiscus</i>	35		
			<i>Helobdella</i>	35		
7. Temporary intermittent streams			<i>Pseudamnicola</i>	35		
			<i>Platycnemis</i>	35		
	Noteridae	35	<i>Dasyhelea</i>	63	<i>Ochthebius delgadoi</i>	42
	Pleidae	35	<i>Anopheles</i>	63	<i>Enochrus politus</i>	38
	Coenagrionidae	30	<i>Helocoris</i>	63	<i>Helophorus fulgidicollis</i>	38
	Libellulidae	28	<i>Anisops</i>	63	<i>Laccophilus minutus</i>	38
	Hydrophilidae	26	<i>Anax</i>	52	<i>Ochthebius auropallens</i>	38
			<i>Enochrus</i>	48	<i>Ochthebius grandipennis</i>	38
			<i>Sympetrum</i>	48	<i>Ochthebius viridis fallaciosus</i>	38
			<i>Berosus</i>	45	<i>Ochthebius jamei</i>	35
			<i>Sigara</i>	45	<i>Helochaers lividus</i>	27
			<i>Plea</i>	45		
			<i>Ischnura</i>	45		
			<i>Noterus</i>	42		
			<i>Potamopyrgus</i>	42		
			<i>Cercion</i>	42		
			<i>Libellula</i>	42		
			<i>Helochaers</i>	41		
			<i>Bidessus</i>	40		
			<i>Procambarus</i>	40		
			<i>Limnophora</i>	40		
			<i>Tipula</i>	40		
			<i>Microvelia</i>	40		
			<i>Agabus</i>	36		
			<i>Dryops</i>	32		
			<i>Laccobius</i>	32		
			<i>Orthetrum</i>	32		
		<i>Gerris</i>	30			
		<i>Nebrioporus</i>	27			
		<i>Cloeon</i>	27			
		<i>Micronecta</i>	25			

Appendix A. Taxa collected in the Segura Basin grouped by taxonomic order.

Hirudinea	Oreodytes
Erpobdellidae	Stictonectes
Dina	<i>Stictonectes epiplericus</i> (Seidlitz, 1887)
Glossiphoniidae	<i>Stictonectes optatus</i> (Seidlitz, 1887)
Helobdella	Yola
Mollusca	<i>Yola bicarinata</i> (Latreille, 1804)
Ancylidae	Elmidae
Ancylus	Elmis
Ferrissia	<i>Elmis aenea</i> (Müller, 1806)
Hydrobiidae	<i>Elmis maugetii maugetii</i> Latreille, 1798
Mercuria	<i>Elmis rioloides</i> (Kuwert, 1890)
Potamopyrgus	Esolus
Pseudamnicola	<i>Esolus parallelepipedus</i> (Müller, 1806)
Lymnaeidae	Limnius
Lymnaea	<i>Limnius intermedius</i> Fairmaire, 1881
Melanopsidae	<i>Limnius opacus</i> Müller, 1806
Melanopsis	<i>Limnius volckmari</i> (Panzer, 1793)
Physidae	Normandia
Physella	<i>Normandia nitens</i> (Müller, 1817)
Planorbidae	<i>Normandia sodalis</i> (Erichson, 1847)
Gyraulus	Oulimnius
Planorbarius	<i>Oulimnius troglodytes</i> (Gyllenhal, 1827)
Sphaeriidae	<i>Oulimnius tuberculatus perezii</i> Sharp, 1872
Pisidium	Potamophilus
Crustacea	Riolus
Astacidae	<i>Riolus cupreus</i> (Müller, 1806)
Austropotamobius	<i>Riolus illiesi</i> Steffan, 1958
Atyidae	Gyrinidae
Atyaephyra	Aulonogyrus
Cambaridae	<i>Aulonogyrus striatus</i> (Fabricius, 1792)
Procambarus	Gyrinus
Gammaridae	<i>Gyrinus dejeani</i> Brullé, 1832
Echinogammarus	Orectochilus
Coleoptera	<i>Orectochilus villosus</i> (Müller, 1776)
Dryopidae	Haliplidae
Dryops	<i>Peltodytes rotundatus</i> (Aubé, 1836)
<i>Dryops gracilis</i> (Karsch, 1881)	Haliplus
<i>Dryops sulcipennis</i> (Costa, 1883)	<i>Haliplus lineatocollis</i> (Marshall, 1802)
Pomatius	<i>Haliplus mucronatus</i> Stephens, 1832
<i>Pomatius substriatus</i> (Müller, 1806)	Helophoridae
Dytiscidae	Helophorus
<i>Eretes griseus</i> Motschulsky 1849	<i>Helophorus alternans</i> Gené, 1836
<i>Hygrotus confluens</i> (Fabricius, 1767)	<i>Helophorus brevipalpis</i> Bedel, 1881
<i>Hyphydus aubei</i> Ganglbauer, 1892	<i>Helophorus fulgidicollis</i> Motschulsky, 1860
<i>Ilybius meridionalis</i> Aubé, 1836	<i>Helophorus occidentalis</i> Angus, 1983
<i>Meladema coriacea</i> Castelnau, 1834	<i>Helophorus nubilis</i> Fabricius, 1776
<i>Rhantus suturalis</i> (McLeay, 1825)	<i>Helophorus seidlitzii</i> Kuwert, 1885
<i>Stictotarsus duodecimpustulatus</i> (Fabricius, 1792)	Hydraenidae
Agabus	Hydraena
<i>Agabus biguttatus</i> (Olivier, 1795)	<i>Hydraena capta</i> Orchymont, 1936
<i>Agabus bipustulatus</i> (Linnaeus, 1767)	<i>Hydraena carbonaria</i> Kiesenwetter, 1849
<i>Agabus brunneus</i> (Fabricius, 1798)	<i>Hydraena exasperata</i> Orchymont, 1935
<i>Agabus didymus</i> (Olivier, 1795)	<i>Hydraena hernandoi</i> Fresneda & Lagar, 1990
<i>Agabus nebulosus</i> (Forster, 1771)	<i>Hydraena mantredjaechi</i> Delgado & Soler, 1991
<i>Agabus nitidus</i> (Fabricius, 1801)	<i>Hydraena pygmaea</i> Waterhouse, 1833
<i>Agabus paludosus</i> (Fabricius, 1801)	<i>Hydraena quillisi</i> Lagar, Fresneda & Hernando, 1987
<i>Agabus rambiae</i> Millán & Ribera, 2001	<i>Hydraena rufipennis</i> Boscá Berga, 1932
Bidessus	<i>Hydraena servilla</i> Orchymont, 1936
<i>Bidessus minutissimus</i> (Germar, 1824)	Limnebius
Deronectes	<i>Limnebius cordobanus</i> Orchymont, 1938
<i>Deronectes depressicollis</i> (Rosenhauer, 1856)	<i>Limnebius maurus</i> Balfour-Browne, 1978
<i>Deronectes fairmairei</i> (Leprieur, 1876)	<i>Limnebius oblongus</i> Rey, 1883
<i>Deronectes hispanicus</i> (Rosenhauer, 1856)	Ochthebius
<i>Deronectes moestus</i> Leprieur, 1876	<i>Ochthebius auropallens</i> Fairmaire, 1879
Dytiscus	<i>Ochthebius bellieri</i> Kuwert, 1887
Graptodytes	<i>Ochthebius bonnairei</i> Guillebau, 1896
<i>Graptodytes fractus</i> (Sharp, 1880-82)	<i>Ochthebius delgadoi</i> Jäch, 1994
<i>Graptodytes ignotus</i> (Mulsant, 1861)	<i>Ochthebius difficilis</i> Mulsant, 1844
<i>Graptodytes varius</i> (Aubé, 1836)	<i>Ochthebius dilatatus</i> Stephens, 1829
Hydroglyphus	<i>Ochthebius (Enicocerus) exsculptus</i> Germar, 1824
<i>Hydroglyphus geminus</i> (Fabricius, 1792)	<i>Ochthebius grandipennis</i> Fairmaire, 1879
<i>Hydroglyphus signatellus</i> (Klug, 1834)	<i>Ochthebius jaimiei</i> Delgado & Jäch, 2007
Hydroporus	<i>Ochthebius quadrifoveolatus</i> Wollaston, 1854
<i>Hydroporus discretus</i> Fairmaire, 1859	<i>Ochthebius tudmirensis</i> Jäch, 1997
<i>Hydroporus lucasi</i> Reiche, 1866	<i>Ochthebius viridis fallaciosus</i> Ganglbauer, 1901
<i>Hydroporus marginatus</i> (Dufschmid, 1805)	Hydrochilidae
<i>Hydroporus nigrita</i> (Fabricius, 1792)	Hydrochus
<i>Hydroporus pubescens</i> (Gyllenhal, 1808)	<i>Hydrochus grandicollis</i> Kiesenwetter, 1870
<i>Hydroporus tessellatus</i> Drapiez, 1819	<i>Hydrochus noereinus</i> Henegouven & Sáinz-Cantero, 1992
Laccophilus	Hydrophilidae
<i>Laccophilus hyalinus</i> (De Geer, 1774)	<i>Anacaena bipustulata</i> (Marshall, 1802)
<i>Laccophilus minutus</i> (Linnaeus, 1758)	<i>Anacaena globulus</i> (Paykull, 1798)
Nebrioporus	<i>Anacaena lutescens</i> (Stephens, 1829)
<i>Nebrioporus bucheti cazorlensis</i> (Lagar, Fresneda & Hernando, 1987)	<i>Coelostoma hispanicum</i> (Küster, 1848)
<i>Nebrioporus clarki</i> (Wollaston, 1862)	<i>Hydrophilus pistaceus</i> (Castelnau, 1840)

Appendix A (cont.).

1		
2		
3		
4		
5	Berosus	<u>Corixidae</u>
6	<i>Berosus hispanicus</i> Küster, 1847	Helioecoris
7	Enochrus	Micronecta
8	<i>Enochrus ater</i> (Kuwert, 1888)	Sigara
9	<i>Enochrus politus</i> Küster, 1849	<u>Gerridae</u>
10	Helochares	Aquarius
11	<i>Helochares lividus</i> (Forster, 1771)	Gerris
12	Laccobius	<u>Hydrometridae</u>
13	<i>Laccobius bipunctatus</i> (Fabricius, 1775)	Hydrometra
14	<i>Laccobius hispanicus</i> Gentili, 1974	<u>Naucoridae</u>
15	<i>Laccobius gracillis gracillis</i> Motschulsky, 1849	Naucoris
16	<i>Laccobius moraguesi</i> Régimbart, 1898	<u>Nepidae</u>
17	<i>Laccobius neapolitanus</i> Rottenberg, 1874	Nepa
18	<i>Laccobius obscuratus</i> Rottenberg, 1874	<u>Notonectidae</u>
19	<i>Laccobius sinuatus</i> Motschulsky, 1849	Anisops
20	<i>Laccobius ytenensis</i> Sharp, 1910	Notonecta
21	<u>Noteridae</u>	<u>Pleididae</u>
22	Noterus	Plea
23	<i>Noterus laevis</i> Sturm, 1834	<u>Veliidae</u>
24	<u>Scirtidae</u>	Microvelia
25	Cyphon	Velia
26	Elodes	Odonata
27	Hydrocyphon	<u>Aeshnidae</u>
28	Diptera	Anax
29	<u>Anthomyiidae</u>	Boyeria
30	Limnophora	<u>Calopterygidae</u>
31	<u>Athericidae</u>	Calopteryx
32	Atrichops	<u>Coenagrionidae</u>
33	Ibisia	Cercion
34	<u>Ceratopogonidae</u>	Ischnura
35	Dasyhelea	Pyrrhosoma
36	<u>Chironomidae</u>	<u>Cordulegastridae</u>
37	Chironomini	Cordulegaster
38	Corynoneura	<u>Gomphidae</u>
39	Tanytarsini	Gomphus
40	<u>Culicidae</u>	Onychogomphus
41	Anopheles	<u>Libellulidae</u>
42	<u>Diamesinae</u>	Libellula
43	<u>Dixidae</u>	Orthetrum
44	<u>Empididae</u>	Sympetrum
45	<u>Ephydriidae</u>	<u>Platycnemididae</u>
46	<u>Hemerodromiinae</u>	Platycnemis
47	<u>Limoniidae</u>	Plecoptera
48	Eloeophyla	<u>Leuctridae</u>
49	Pseudolimnophila	Leuctra
50	<u>Orthocladiinae</u>	<u>Nemouridae</u>
51	<u>Simuliidae</u>	Nemoura
52	<u>Stratiomyidae</u>	Protonemura
53	Oxycera	<u>Perlidae</u>
54	<u>Syrphidae</u>	Dinocras
55	<u>Tabanidae</u>	Eoperla
56	Tabanus	Perla
57	<u>Tanypodinae</u>	<u>Perlodidae</u>
58	<u>Tipulidae</u>	Isoperla
59	Tipula	Trichoptera
60	Ephemeroptera	<u>Beraeidae</u>
	<u>Baetidae</u>	<u>Brachycentridae</u>
	Baetis	Micrasema
	Centroptilum	<u>Drusinae</u>
	Cloeon	<u>Hydropsychidae</u>
	Procloeon	Cheumatopsyche
	<u>Caenidae</u>	Hydropsyche
	Caenis	<u>Hydroptilidae</u>
	<u>Ephemerellidae</u>	Agraylea
	Ephemerella	Hydroptila
	Serratella	<u>Lepidostomatidae</u>
	Torleya	Lasiocephala
	<u>Ephemeridae</u>	<u>Leptoceridae</u>
	Ephemera	Athripsodes
	<u>Heptageniidae</u>	<u>Limnephiliidae</u>
	Ecdyonurus	Allogamus
	Epeorus	Halesus
	Rhithrogena	Stenophylax
	<u>Leptophlebiidae</u>	<u>Limnephiliinae</u>
	Habroleptoides	<u>Polycentropodidae</u>
	Habrophlebia	<u>Psychomyiidae</u>
	Paraleptophlebia	Metalype
	<u>Polymitacidae</u>	Tinodes
	Ephoron	<u>Rhyacophiliidae</u>
	<u>Potamantidae</u>	Rhyacophila
	Potamanthus	<u>Sericostomatidae</u>
	Hemiptera	Sericostoma
	<u>Aphelocheiridae</u>	
	Aphelocheirus	

References

- 1
2
3
4
5 Argyroudi A, Chatzinikolaou Y, Poirazidis K, Lazaridou M. 2009. Do intermittent and
6 ephemeral Mediterranean rivers belong to the same river type? *Aquatic Ecology* **43**:465-
7 476.
8
- 9
10 Armanini DG, Monk WA, Tenenbaum DE, Peters DL, Baird DJ. 2011. Influence of
11 runoff regime type on a macroinvertebrate-based flow index in rivers of British
12 Columbia (Canada). *Ecohydrology*. DOI: 10.1002/eco.234.
13
- 14 Arthington AH, Bunn SE, Poff NL, Naiman RJ. 2006. The challenge of providing
15 environmental flow rules to sustain river ecosystems. *Ecological Applications* **16**:1311-
16 1318.
17
- 18 Barahona J, Millan A, Velasco J. 2005. Population dynamics, growth and production of
19 *Sigara selecta* (Fieber, 1848) (Hemiptera, Corixidae) in a Mediterranean hypersaline
20 stream. *Freshwater Biology* **50**:2101-2113.
21
- 22 Beals EW. 1984. Bray-Curtis ordination: an effective strategy for analysis of
23 multivariate ecological data. *Advances in Ecological Research* **14**:1-55.
24
- 25 Belmar O, Velasco J, Martinez-Capel F. 2011. Hydrological classification of natural
26 flow regimes to support environmental flow assessments in intensively regulated
27 Mediterranean rivers, Segura River basin (Spain). *Environmental Management* **47**:992-
28 1004.
29
- 30 Belmar O, Velasco J, Martinez-Capel F, Marin AA. 2010. Natural flow regime, degree of
31 alteration and environmental flows in the Mula stream (Segura River basin, SE Spain).
32 *Limnetica* **29**:353-368.
33
- 34 Bilton DT, Freeland JR, Okamura B. 2001. Dispersal in freshwater invertebrates.
35 *Annual Review of Ecology and Systematics* **32**:159-181.
36
- 37 Bilton DT, McAbendroth L, Bedford A, Ramsay PM. 2006. How wide to cast the net?
38 Cross-taxon congruence of species richness, community similarity and indicator taxa in
39 ponds. *Freshwater Biology* **51**:578-590.
40
- 41 Bogan MT, Lytle DA. 2011. Severe drought drives novel community trajectories in
42 desert stream pools. *Freshwater Biology* **56**:2070-2081.
43
- 44 Bonada N, Doledec S, Statzner B. 2007a. Taxonomic and biological trait differences of
45 stream macroinvertebrate communities between mediterranean and temperate regions:
46 implications for future climatic scenarios. *Global Change Biology* **13**:1658-1671.
47
- 48 Bonada N, Murria C, Zamora-Munoz C, El Alami M, Poquet JM, Punti T, Moreno JL,
49 Bennis N, Alba-Tercedor J, Ribera C, Prat N. 2009. Using community and population
50 approaches to understand how contemporary and historical factors have shaped species
51 distribution in river ecosystems. *Global Ecology and Biogeography* **18**:202-213.
52
- 53 Bonada N, Prat N, Munne A, Plans M, Sola C, Alvarez-Cabria M, Pardo I, Moya G,
54 Ramon G, Toro M, Robles S, Aviles J, Suarez ML, Vidal-Abarca MR, Mellado A,
55 Moreno JL, Guerrero C, Vivas S, Ortega M, Casas J, Sanchez-Ortega A, Jaimez-Cuellar
56
57
58
59
60

1
2
3 P, Alba-Tercedor J. 2002. Intercalibración de la metodología GUADALMED. Selección
4 de un protocolo de muestreo para la determinación del estado ecológico de los ríos
5 mediterráneos. *Limnetica* **21**:13-33.
6

7 Bonada N, Rieradevall M, Prat N. 2007b. Macroinvertebrate community structure and
8 biological traits related to flow permanence in a Mediterranean river network.
9 *Hydrobiologia* **589**:91-106.
10

11 Bonada N, Rieradevall M, Prat N, Resh VH. 2006. Benthic macroinvertebrate
12 assemblages and macrohabitat connectivity in Mediterranean-climate streams of
13 northern California. *Journal of the North American Benthological Society* **25**:32-43.
14

15 Bonada N, Zamora-Munoz C, Rieradevall M, Prat N. 2004. Ecological profiles of
16 caddisfly larvae in Mediterranean streams: implications for bioassessment methods.
17 *Environmental Pollution* **132**:509-521.
18

19 Boulton AJ. 2003. Parallels and contrasts in the effects of drought on stream
20 macroinvertebrate assemblages. *Freshwater Biology* **48**:1173-1185.
21

22 Boulton AJ, Lake PS. 2008. Effects of drought on stream insects and its ecological
23 consequences. In *Aquatic Insects: Challenges to Populations*, 81-102. Lancaster J,
24 Briers RA (eds). CAB International. Wallingford.
25

26 Boulton AJ, Sheldon F, Jenkins KM. 2006. Natural disturbance and aquatic
27 invertebrates in desert rivers. In *Ecology of Desert Rivers*, 103-153. Kingsford RT(ed).
28 Academic Press. New York.
29

30 Bracken LJ, Coxi NJ, Shannon J. 2008. The relationship between rainfall inputs and
31 flood generation in south-east Spain. *Hydrological Processes* **22**:683-696.
32

33 Bunn SE, Arthington AH. 2002. Basic principles and ecological consequences of
34 altered flow regimes for aquatic biodiversity. *Environmental Management* **30**:492-507.
35

36 Carbonell JA, Gutierrez-Canovas C, Bruno D, Abellan P, Velasco J, Millan A. 2011.
37 Ecological factors determining the distribution and assemblages of the aquatic
38 hemiptera (Gerromorpha & Nepomorpha) in the Segura River Basin (Spain). *Limnetica*
39 **30**:59-70.
40

41 Castella E, Bickerton M, Armitage PD, Petts GE. 1995. The effects of water
42 abstractions on invertebrate communities in UK streams. *Hydrobiologia* **308**:167-182.
43

44 Chase JM. 2007. Drought mediates the importance of stochastic community assembly.
45 *Proceedings of the National Academy of Sciences of the United States of America*
46 **104**:17430-17434.
47

48 Chinnayakanahalli KJ, Hawkins CP, Tarboton DG, Hill RA. 2011. Natural flow regime,
49 temperature and the composition and richness of invertebrate assemblages in streams of
50 the western United States. *Freshwater Biology* **56**:1248-1265.
51

52 CHS. 2007. *Estudio general sobre la Demarcación Hidrográfica del Segura*.
53 Confederación Hidrográfica del Segura.
54
55
56
57
58
59
60

- 1
2
3 Clarke KR. 1993. Nonparametric multivariate analyses of changes in community
4 structure. *Australian Journal of Ecology* **18**:117-143.
5
6 Clarke KR, Gorley RN. 2006. *PRIMER v6: User Manual/Tutorial*. PRIMER-E,
7 Plymouth, United Kingdom.
8
9 Clausen B, Biggs BJF. 1997. Relationships between benthic biota and hydrological
10 indices in New Zealand streams. *Freshwater Biology* **38**:327-342.
11
12 Cote IM, Darling ES. 2010. Rethinking ecosystem resilience in the face of climate
13 change. *Plos Biology* **8**:e1000438.
14
15 De Caceres M, Legendre P. 2008. Beals smoothing revisited. *Oecologia* **156**:657-669.
16
17 Diaz AM, Alonso MLS, Gutierrez M. 2008. Biological traits of stream
18 macroinvertebrates from a semi-arid catchment: patterns along complex environmental
19 gradients. *Freshwater Biology* **53**:1-21.
20
21 Dufrene M, Legendre P. 1997. Species assemblages and indicator species: the need for a
22 flexible asymmetrical approach. *Ecological Monographs* **67**:345-366.
23
24 Estrela T, Cabezas F, Estrada F. 1999. La evaluación de los recursos hídricos en el
25 Libro Blanco del Agua en España. *Ingeniería del Agua* **6**:125-138.
26
27 European Environment Agency. 2008. *Energy and environment report 2008*.
28 Copenhagen, Denmark.
29
30 Feminella JW. 1996. Comparison of benthic macroinvertebrate assemblages in small
31 streams along a gradient of flow permanence. *Journal of the North American*
32 *Benthological Society* **15**:651-669.
33
34 Gasith A, Resh VH. 1999. Streams in Mediterranean climate regions: abiotic influences
35 and biotic responses to predictable seasonal events. *Annual Review of Ecology and*
36 *Systematics* **30**:51-81.
37
38 Grindlay A, Zamorano M, Rodriguez MI, Molero E, Urrea MA. 2009. Territorial
39 transformation and water utilization: hydrological planning scenarios in the Segura river
40 basin. In *Sustainable Development and Planning IV, Vols 1 and 2*. Wit Press.
41 Southampton, 975-+.
42
43 Grindlay AL, Zamorano M, Rodriguez MI, Molero E, Urrea MA. 2011. Implementation
44 of the European Water Framework Directive: integration of hydrological and regional
45 planning at the Segura River Basin, southeast Spain. *Land Use Policy* **28**:242-256.
46
47 Hart DD, Finelli CM. 1999. Physical-biological coupling in streams: the pervasive
48 effects of flow on benthic organisms. *Annual Review of Ecology and Systematics*
49 **30**:363-395.
50
51 Jacobsen D, Rostgaard S, Vasconez JJ. 2003. Are macroinvertebrates in high altitude
52 streams affected by oxygen deficiency? *Freshwater Biology* **48**:2025-2032.
53
54
55
56
57
58
59
60

1
2
3 Jáimez-Cuéllar P, Vivas S, Bonada N, Robles S, Mellado A, Álvarez M, Avilés J, Casas
4 J, Ortega M, Pardo I, Prat N, Rieradevall M, Sáinz-Cantero CE, Sánchez-Ortega A,
5 Suárez ML, Toro M, Vidal-Abarca MR, Zamora-Munoz C, Alba-Tercedor J. 2002.
6 Protocolo GUADALMED (PRECE). *Limnetica* **21**:187-204.
7

8 Jowett IG, Biggs BJF. 2009. Application of the natural flow paradigm in a New Zealand
9 context. *River Research and Applications* **25**:1126-1135.
10

11 Jowett IG, Duncan MJ. 1990. Flow variability in New Zealand rivers and its
12 relationship to in-stream habitat and biota. *New Zealand Journal of Marine and*
13 *Freshwater Research* **24**:305-317.
14

15 Kennard MJ, Mackay SJ, Pusey BJ, Olden JD, Marsh N. 2010. Quantifying uncertainty
16 in estimation of hydrologic metrics for ecohydrological studies. *River Research and*
17 *Applications* **26**:137-156.
18

19 Kennard MJ, Olden JD, Arthington AH, Pusey BJ, Poff NL. 2007. Multiscale effects of
20 flow regime and habitat and their interaction on fish assemblage structure in eastern
21 Australia. *Canadian Journal of Fisheries and Aquatic Sciences* **64**:1346-1359.
22

23 Kennen JG, Riva-Murray K, Beaulieu KM. 2010. Determining hydrologic factors that
24 influence stream macroinvertebrate assemblages in the northeastern US. *Ecohydrology*
25 **3**:88-106.
26

27 Konrad CP, Brasher AMD, May JT. 2008. Assessing streamflow characteristics as
28 limiting factors on benthic invertebrate assemblages in streams across the western
29 United States. *Freshwater Biology* **53**:1983-1998.
30

31 Lake PS. 2003. Ecological effects of perturbation by drought in flowing waters.
32 *Freshwater Biology* **48**:1161-1172.
33

34 Larned ST, Detry T, Arscott DB, Tockner K. 2010. Emerging concepts in temporary-
35 river ecology. *Freshwater Biology* **55**:717-738.
36

37 Lytle DA, Poff NL. 2004. Adaptation to natural flow regimes. *Trends in Ecology &*
38 *Evolution* **19**:94-100.
39

40 Mathews R, Richter BD. 2007. Application of the indicators of hydrologic alteration
41 software in environmental flow setting. *Journal of the American Water Resources*
42 *Association* **43**:1400-1413.
43

44 McAbendroth L, Foggo A, Rundle SD, Bilton DT. 2005. Unravelling nestedness and
45 spatial pattern in pond assemblages. *Journal of Animal Ecology* **74**:41-49.
46

47 McArdle BH, Anderson MJ. 2001. Fitting multivariate models to community data: A
48 comment on distance-based redundancy analysis. *Ecology* **82**:290-297.
49

50 McCune B. 1994. Improving community analysis with the Beals smoothing function.
51 *Ecoscience* **1**:82-86.
52

53 McCune B, Grace JB. 2002. *Analysis of ecological communities*. MjM. Gleneden
54 Beach, OR, USA.
55
56
57
58
59
60

- 1
2
3 Mellado A. 2005. *Ecología de las comunidades de macroinvertebrados de la cuenca del*
4 *Río Segura (SE de España)*, Thesis/Dissertation, University of Murcia (Spain).
5
6 Millan A, Abellan P, Ribera I, Sanchez-Fernandez D, Velasco J. 2006. The
7 Hydradephaga of the Segura Basin (SE Spain): twenty years studying water beetles
8 (Coleoptera). *Memorie della Società entomologica italiana* **85**:137-158.
9
10 Ministerio de Medio Ambiente (MMA). 2004. *Water in Spain*. Secretaría de Estado de
11 Aguas y Costas.
12
13 Monk WA, Wood PJ, Hannah DM, Extence CA, Chadd RP, Dunbar MJ. 2012. How
14 does macroinvertebrate taxonomic resolution influence ecohydrological relationships in
15 riverine ecosystems. *Ecohydrology* **5**:36-45.
16
17 Monk WA, Wood PJ, Hannah DM, Wilson DA. 2007. Selection of river flow indices
18 for the assessment of hydroecological change. *River Research and Applications* **23**:113-
19 122.
20
21 Monk WA, Wood PJ, Hannah DM, Wilson DA. 2008. Macroinvertebrate community
22 response to inter-annual and regional river flow regime dynamics. *River Research and*
23 *Applications* **24**:988-1001.
24
25 Monk WA, Wood PJ, Hannah DM, Wilson DA, Extence CA, Chadd RP. 2006. Flow
26 variability and macroinvertebrate community response within riverine systems. *River*
27 *Research and Applications* **22**:595-615.
28
29 Naiman RJ, Latterell JJ, Pettit NE, Olden JD. 2008. Flow variability and the biophysical
30 vitality of river systems. *Comptes Rendus Geoscience* **340**:629-643.
31
32 Niu SQ, Dudgeon D. 2011a. Environmental flow allocations in monsoonal Hong Kong.
33 *Freshwater Biology* **56**:1209-1230.
34
35 Niu SQ, Dudgeon D. 2011b. The influence of flow and season upon leaf-litter
36 breakdown in monsoonal Hong Kong streams. *Hydrobiologia* **663**:205-215.
37
38 Olden JD, Poff NL. 2003. Redundancy and the choice of hydrologic indices for
39 characterizing streamflow regimes. *River Research and Applications* **19**:101-121.
40
41 Oscoz J, Galicia D, Miranda R. 2011. *Identification guide of freshwater*
42 *macroinvertebrates of Spain*. Springer. Dordrecht, Netherlands.
43
44 Peck JE, Hong WS, McCune B. 1995. Diversity of epiphytic bryophytes on 3 host tree
45 species, thermal meadow, hotspots island, Queen-Charlotte-Islands, Canada.
46 *Bryologist* **98**:123-128.
47
48 Pegg MA, Pierce CL. 2002. Fish community structure in the Missouri and lower
49 Yellowstone rivers in relation to flow characteristics. *Hydrobiologia* **479**:155-167.
50
51 Picazo F, Bilton DT, Moreno JL, Sanchez-Fernandez D, Millan A. 2012. Water beetle
52 biodiversity in Mediterranean standing waters: assemblage composition, environmental
53 drivers and nestedness patterns. *Insect Conservation and Diversity* **5**:146-158.
54
55
56
57
58
59
60

1
2
3 Poff NL. 1996. A hydrogeography of unregulated streams in the United States and an
4 examination of scale-dependence in some hydrological descriptors. *Freshwater Biology*
5 **36**:71-91.

6
7 Poff NL, Allan JD. 1995. Functional organization of stream fish assemblages in relation
8 to hydrological variability. *Ecology* **76**:606-627.

9
10 Poff NL, Allan JD, Bain MB, Karr JR, Prestegard KL, Richter BD, Sparks RE,
11 Stromberg JC. 1997. The natural flow regime. *Bioscience* **47**:769-784.

12
13 Poff NL, Richter BD, Arthington AH, Bunn SE, Naiman RJ, Kendy E, Acreman M,
14 Apse C, Bledsoe BP, Freeman MC, Henriksen J, Jacobson RB, Kennen JG, Merritt DM,
15 O'Keefe JH, Olden JD, Rogers K, Tharme RE, Warner A. 2010. The ecological limits
16 of hydrologic alteration (ELOHA): a new framework for developing regional
17 environmental flow standards. *Freshwater Biology* **55**:147-170.

18
19 Poff NL, Ward JV. 1989. Implications of streamflow variability and predictability for
20 lotic community structure: a regional analysis of streamflow patterns. *Canadian Journal*
21 *of Fisheries and Aquatic Sciences* **46**:1805-1818.

22
23 Puig MA, Gonzalez G, Soriano O. 1984. Introducción al estudio de las comunidades
24 macrobentónicas de los ríos asturianos: efemerópteros, plecópteros, tricópteros,
25 simúlidos y quironómidos. *Limnetica* **1**:187-196.

26
27 R Development Core Team. 2008. *R: a language and environment for statistical*
28 *computing*. Vienna, Austria.

29
30 Resh VH, Brown AV, Covich AP, Gurtz ME, Li HW, Minshall GW, Reice SR, Sheldon
31 AL, Wallace JB, Wissmar RC. 1988. The role of disturbance in stream ecology. *Journal*
32 *of the North American Benthological Society* **7**:433-455.

33
34 Richter BD, Baumgartner JV, Powell J, Braun DP. 1996. A method for assessing
35 hydrologic alteration within ecosystems. *Conservation Biology* **10**:1163-1174.

36
37 Sanchez-Fernandez D, Abellan P, Mellado A, Velasco J, Millan A. 2006. Are water
38 beetles good indicators of biodiversity in Mediterranean aquatic ecosystems? The case
39 of the segura river basin (SE Spain). *Biodiversity and Conservation* **15**:4507-4520.

40
41 Sanchez-Montoya MD, Punti T, Suarez ML, Vidal-Abarca MD, Rieradevall M, Poquet
42 JM, Zamora-Munoz C, Robles S, Alvarez M, Alba-Tercedor J, Toro M, Pujante AM,
43 Munne A, Prat N. 2007. Concordance between ecotypes and macroinvertebrate
44 assemblages in Mediterranean streams. *Freshwater Biology* **52**:2240-2255.

45
46 Sheldon F, Thoms MC. 2006. Relationships between flow variability and
47 macroinvertebrate assemblage composition: data from four Australian dryland rivers.
48 *River Research and Applications* **22**:219-238.

49
50 Smakhtin VU. 2001. Low flow hydrology: a review. *Journal of Hydrology* **240**:147-
51 186.

- 1
2
3 Snelder TH, Biggs BJF. 2002. Multiscale River Environment Classification for water
4 resources management. *Journal of the American Water Resources Association* **38**:1225-
5 1239.
6
- 7 Snelder TH, Biggs BJF, Woods RA. 2005. Improved eco-hydrological classification of
8 rivers. *River Research and Applications* **21**:609-628.
9
- 10 Snelder TH, Lamouroux N, Leathwick JR, Pella H, Sauquet E, Shankar U. 2009.
11 Predictive mapping of the natural flow regimes of France. *Journal of Hydrology*
12 **373**:57-67.
13
- 14 Stanley EH, Fisher SG, Grimm NB. 1997. Ecosystem expansion and contraction in
15 streams. *Bioscience* **47**:427-435.
16
- 17 StatSoft, Inc. 2001. *STATISTICA (data analysis software system), version 6.*
18 www.statsoft.com.
19
- 20 Suen JP, Herricks EE. 2009. Developing fish community based ecohydrological
21 indicators for water resources management in Taiwan. *Hydrobiologia* **625**:223-234.
22
- 23 Thoms MC. 2006. Variability in riverine ecosystems. *River Research and Applications*
24 **22**:115-121.
25
- 26 Tockner K, Ward JV, Arscott DB, Edwards PJ, Kollmann J, Gurnell AM, Petts GE,
27 Maiolini B. 2003. The Tagliamento River: a model ecosystem of European importance.
28 *Aquatic Sciences* **65**:239-253.
29
- 30 Vaughan IP, Diamond M, Gurnell AM, Hall KA, Jenkins A, Milner NJ, Naylor LA,
31 Sear DA, Woodward G, Ormerod SJ. 2009. Integrating ecology with hydromorphology:
32 a priority for river science and management. *Aquatic Conservation-Marine and*
33 *Freshwater Ecosystems* **19**:113-125.
34
- 35 Velasco J, Millan A, Nieser N. 1990. Observaciones sobre la colonización y el ciclo de
36 vida de *Heliocoris vermiculata* (Puton, 1874) (Heteroptera, Corixidae) en pequeños
37 estanques del SE español. *Limnetica* **6**:101-108.
38
- 39 Vivas S, Casas J, Pardo I, Robles S, Bonada N, Mellado A, Prat N, Alba-Tercedor J,
40 Alvarez-Cabria M, Bayo MM, Jaimez-Cuellar P, Suárez ML, Toro M, Vidal-Abarca
41 MR, Zamora-Munoz C, Moyá G. 2002. Aproximación multivariante en la exploración
42 de la tolerancia ambiental de las familias de macroinvertebrados de los ríos
43 mediterráneos del proyecto GUADALMED. *Limnetica* **21**:149-173.
44
- 45 Walters AW, Post DM. 2011. How low can you go? Impacts of a low-flow disturbance
46 on aquatic insect communities. *Ecological Applications* **21**:163-174.
47
- 48 Winter TC. 2001. The concept of hydrologic landscapes. *Journal of the American Water*
49 *Resources Association* **37**:335-349.
50
- 51 Zimmer A. 2010. New water uses in the Segura Basin: conflicts around gated
52 communities in Murcia. *Water International* **35**:34-48.
53
54
55
56
57
58
59
60

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

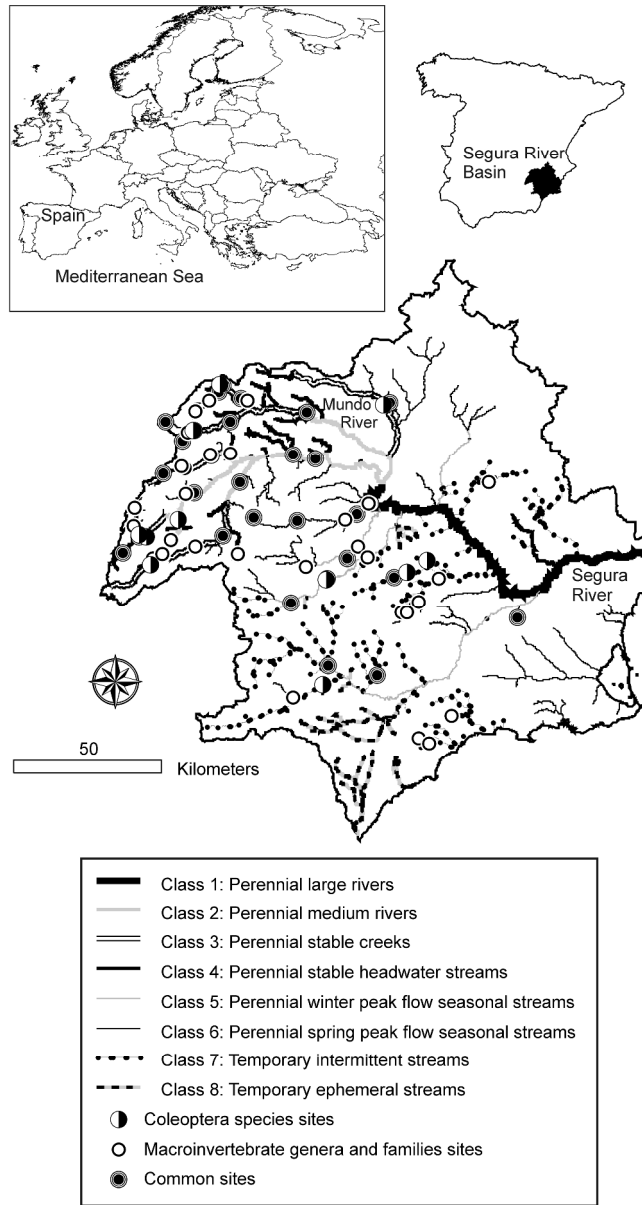


Figure 1. Location of the study area, hydrological classes in the river network and sampling sites.
290x533mm (300 x 300 DPI)

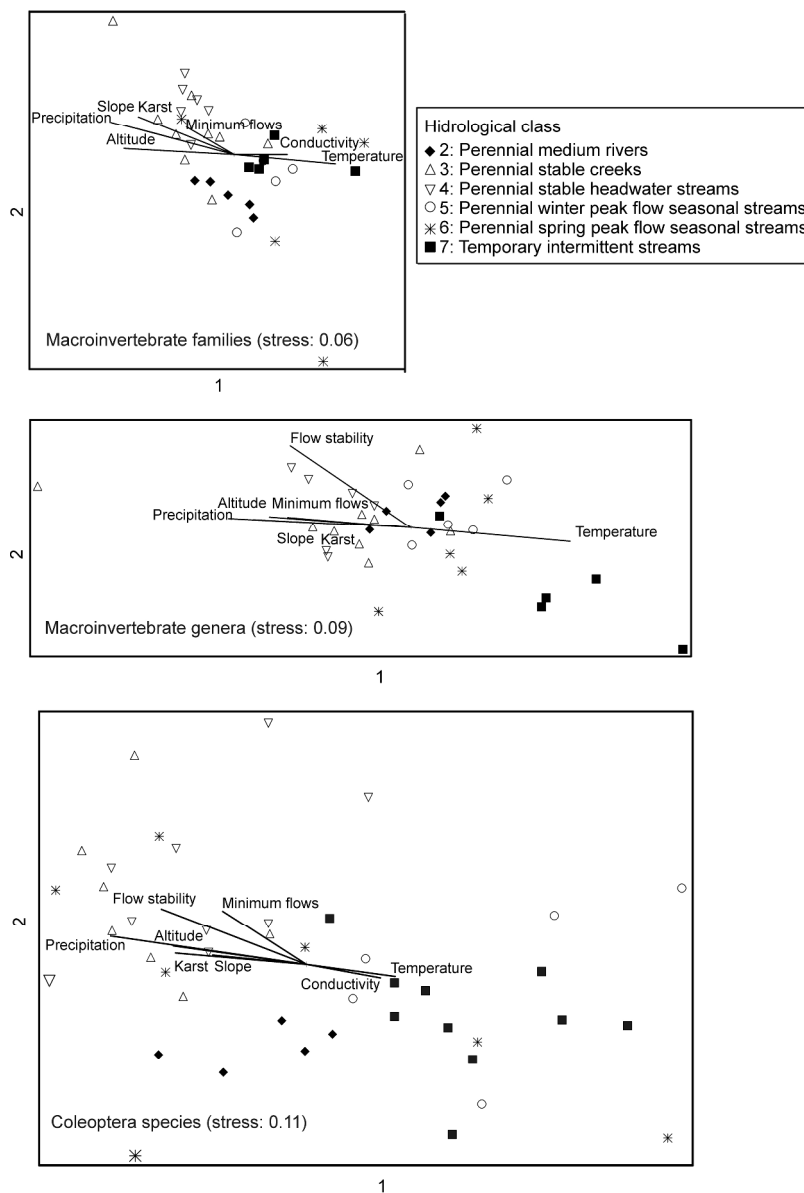


Figure 2. NMDS plots of sites for each taxonomic level. The magnitudes of the correlations between the NDMS axes and the hydrological components as well as the environmental variables are shown as vectors.
289x431mm (300 x 300 DPI)

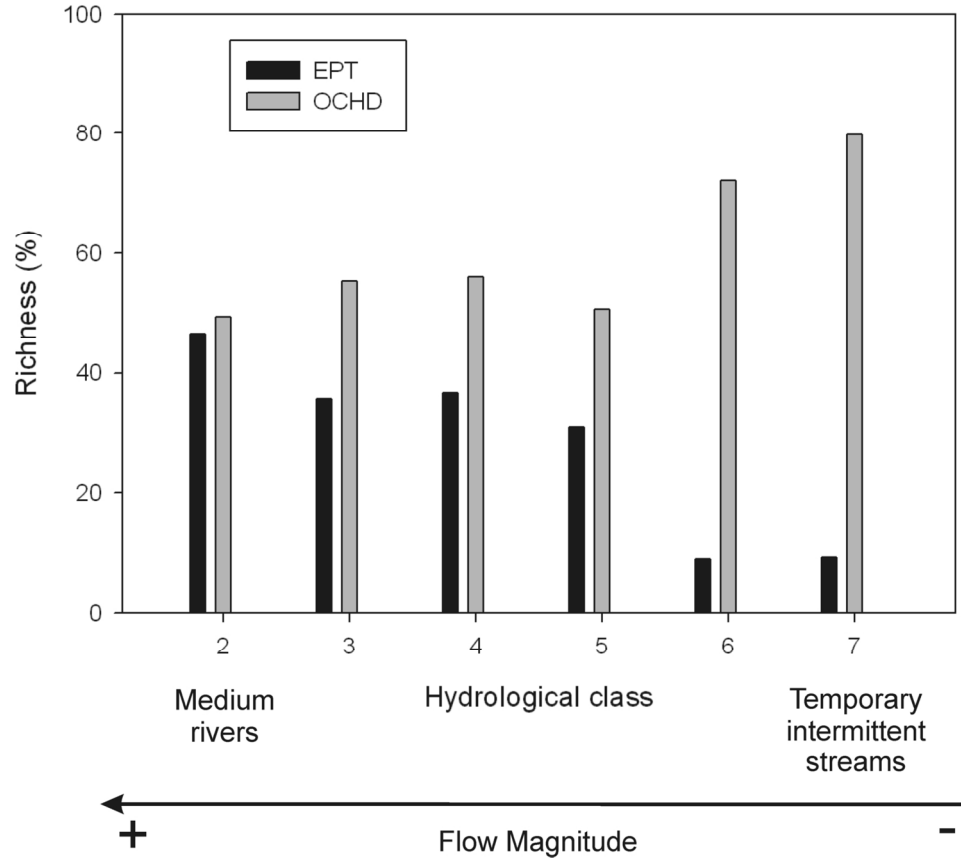


Figure 3. Variation of the percentage of families of the EPT and OCDH groups in the different hydrological classes along the flow magnitude gradient.
140x130mm (300 x 300 DPI)