

## Assessing effects of water abstraction on fish assemblages in Mediterranean streams

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### SUMMARY

1. Water abstraction strongly affects streams in arid and semiarid ecosystems, particularly where there is a Mediterranean climate. Excessive abstraction reduces the availability of water for human uses downstream and impairs the capacity of streams to support native biota.

2. We investigated the flow regime and related variables in six river basins of the Iberian Peninsula and show that they have been strongly altered, with declining flows (autoregressive models) and groundwater levels during the 20th century. These streams had lower flows and more frequent droughts than predicted by the official hydrological model used in this region. Three of these rivers were sometimes dry, whereas there were predicted by the model to be permanently flowing. Meanwhile, there has been no decrease in annual precipitation.

3. We also investigated the fish assemblage of a stream in one of these river basins (Tordera) for 6 years and show that sites more affected by water abstraction display significant differences in four fish metrics (catch per unit effort, number of benthic species, number of intolerant species and proportional abundance of intolerant individuals) commonly used to assess the biotic condition of streams.

4. We discuss the utility of these metrics in assessing impacts of water abstraction and point out the need for detailed characterisation of the natural flow regime (and hence drought events) prior to the application of biotic indices in streams severely affected by water abstraction. In particular, in cases of artificially dry streams, it is more appropriate for regulatory agencies to assign index scores that reflect biotic degradation than to assign 'missing' scores, as is presently customary in assessments of Iberian streams.

*Keywords:* drought, fish metrics, Mediterranean streams, natural flow regime, water abstraction

### Introduction

Freshwater ecosystems are essential for human society and have thus been widely altered by a suite of interacting human actions, including pollution, dam

building, habitat alteration and water abstraction (Prat & Munné, 2000; Baron *et al.*, 2002; Nilsson *et al.*, 2005; Xenopoulos & Lodge, 2006). These actions can profoundly alter the ecological structure and function of aquatic biota. Although humans exploit only 10–15% of current global runoff, water abstraction, mostly for irrigation, is very intensive in many arid or semiarid regions, where it can exceed the naturally available surface runoff (Vörösmarty & Sahagian, 2000). Streams in a Mediterranean climate

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(e.g. those along the Mediterranean itself or in coastal California) are particularly affected by water abstraction (either directly or via groundwater withdrawal) because they combine a deficit of surface water availability with high human population densities and agriculture development (Gasith & Resh, 1999). In Mediterranean regions, water abstraction may change a perennial stream to an intermittent one, increasing the duration and magnitude of droughts and limiting the stream's ability to support aquatic biota.

Droughts are a major selective force in structuring aquatic populations and communities. The drying process shrinks the water column, which reduces water quality, increases fish density, and alters food resources as well as the strength and structure of interspecific interactions (Magoulick & Kobza, 2003; Davey & Kelly, 2007). These changes commonly cause reductions in population size and species richness, especially of native and intolerant species (Magoulick, 2000; Lake, 2003; Matthews & Marsh-Matthews, 2003; Magalhães *et al.*, 2007). Negative effects on reproduction, condition and growth of individual fish have also been shown (Magoulick & Kobza, 2003; Keaton, Haney & Andersen, 2005).

Fish assemblages are excellent indicators of aquatic ecosystem health (Karr, 1981; Fausch *et al.*, 1990; Angermeier & Davideanu, 2004) and fish-based biotic indices have been widely developed, starting with the Index of Biotic Integrity (Karr, 1981; Karr *et al.*, 1986). These indices are sensitive to a wide range of anthropogenic impacts, including those due to agriculture, urbanisation and mining. In Europe, the Water Framework Directive [EC (European Community), 2000] requires the bioassessment of rivers using fish and the achievement of good 'ecological status' before 2015 and has stimulated the recent development of fish-based biotic indices (Pont *et al.*, 2006; Pont, Hugueny & Rogers, 2007). However, Mediterranean streams naturally support few fish species (but many endemics), which hampers the development of fish-based indices (Bramblett & Fausch, 1991; Ferreira *et al.*, 2007). Moreover, the ability of such indices to detect the effects of water abstraction, which are severe in southern Europe, has scarcely been tested. The development and calibration of metrics that accurately reflect effects of water abstraction on fish assemblages will be crucial to assessing the quality of Mediterranean streams.

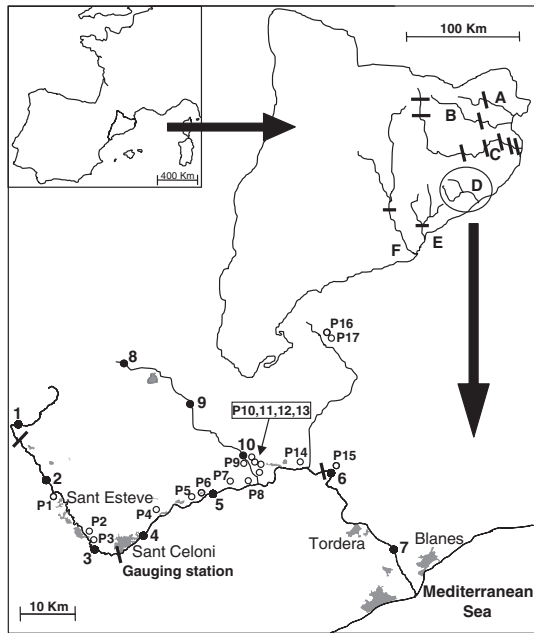
In this paper, we examine the effects of water abstraction on flow regimes and fish assemblages in Mediterranean streams. We analyse the trends for flow regime and related variables during the 20th century in six Mediterranean streams and the fish assemblage in one of them over 6 years. We aimed to assess: (i) to what extent the flow regime has been altered through water abstraction; and (ii) whether certain fish metrics may be useful in detecting effects of severe water abstraction. We also point out the need to incorporate detailed knowledge of natural flow regimes (including changes in drought frequency and severity) into fish-based assessments of streams affected by water abstraction.

## Methods

### *Study area and flow regime*

Variation in streamflow during the 20th century was studied in the six largest Mediterranean river basins in NE Catalonia, Spain: Muga, Fluvià, Ter, Tordera, Besòs and Llobregat (Fig. 1). These basins have drainage areas of 853–4948 km<sup>2</sup> and three (Muga, Ter, Llobregat) have large reservoirs (0.062–0.233 km<sup>3</sup> in volume); the other three basins have only small reservoirs (<0.001 km<sup>3</sup>) and weirs. The yearly mean daily flow of these streams is 1.1–17 m<sup>3</sup> s<sup>-1</sup>. Catalonia has a typical Mediterranean climate with dry, warm summers, mild winters and an average precipitation of 734 mm year<sup>-1</sup>. The region encompassing these basins is densely populated (5.8 million residents but 12 million summer inhabitants, living in 12 800 km<sup>2</sup>); the predominant land uses are agricultural, industrial and residential (Sala & Bar, 1992; Aparicio *et al.*, 2000).

Measures of daily streamflow for these six basins from 1940 to 2000 were obtained from the Catalan Water Agency (available at <http://www.gencat.cat/aca>). To compare the streamflow in different basins, we selected one gauging station in the middle reach, downstream of reservoirs; we also analysed data for all existing stations in the mainstem of the Tordera and Ter rivers (Fig. 1). For these sites, we also estimated the 'naturalised flow' (i.e. the flow expected if there was no direct human influence on the watercourse, e.g. from water abstraction) using the Sacramento Soil Moisture Accounting (SAC-SMA) model. The SAC-SMA model is a flexible, well-known model initially developed by Burnash, Ferral & Guire



**Fig. 1** The six Mediterranean streams where streamflow was studied: A, Muga; B, Fluvià; C, Ter; D, Tordera; E, Besòs; F, Llobregat. Bottom: Location of sampling sites (●) and piezometers (○) along Tordera stream. The main towns and gauging stations (■) are also shown.

(1973) and widely used by the US National Weather Service (e.g. Hartmann, Bales & Sorooshian, 2002), and also the Catalan Water Agency [ACA (Agència Catalana de l'Aigua), 2002, 2004]. The SAC-SMA model is a conceptually based rainfall-runoff model, with areal precipitation and areal potential evapotranspiration as inputs. The SAC-SMA model parameters have been previously calibrated by the Catalan Water Agency (ACA, 2002) to match historical observed discharge data and so to account for terrain characteristics, geographic location and the distribution of gauges.

#### *Field studies in the Tordera stream*

We also studied the effects of water abstraction on the fish assemblage in the Tordera stream, one of those thought to be strongly affected by abstraction. This stream rises in coastal mountains, drains an area of c. 895 km<sup>2</sup>, and has highly variable flow, with episodic floods. The highest flows occur in spring and autumn and low flows in summer. Mean annual rainfall in the basin ranges from 1000 mm near the summit to 600 mm on the coast; mean annual water yield is 170.4 million m<sup>3</sup> year<sup>-1</sup> and mean discharge is c.

4 m<sup>3</sup> s<sup>-1</sup> (ACA, 2002; Rovira & Batalla, 2006). Predominant land uses in this basin are arable (row-crop) agriculture, plantations of fast-growing trees in flood-plain areas, and residential areas. There are no large dams along the stream. Although there were problems with industrial and urban pollution until the early 1990s, wastewater treatment plants along the mainstem now ensure acceptable water quality (Prat *et al.*, 2002). The Catalan Water Agency monitors macroinvertebrates, diatoms and fish to assess water quality in the Tordera basin (Benejam *et al.*, 2008a, see <http://www.observatoriuriordera.org>).

From summer 2001 to autumn 2006, fish and abiotic variables were measured quarterly (except in winter) at ten sites along Tordera stream (Fig. 1). For each sampling site and date, several limnological and habitat variables were measured: altitude (m), wetted width (m), substratum composition (percentage of the reach with boulder, cobble, gravel, sand and silt), riparian vegetation structure (percentage of the reach covered by trees, bushes and hydrophytes), and percentage of refuges (e.g. large boulders, wood and tree roots), pools and riffles. Other variables, such as ammonium, nitrite, nitrate, phosphate, sulphate and chloride concentrations, streamflow, oxygen saturation, pH, water temperature and conductivity were simultaneously obtained by other monitoring teams (see Prat *et al.*, 2002; Boada *et al.*, 2006) and are largely available at <http://www.gencat.cat/aca>. The height of the water table was measured in 17 piezometers along the stream (Fig. 1) and rainfall data were obtained from the Catalan Water Agency and Meteorological Service of Catalonia respectively.

The sampling sites were 100 m long by 5 m mean width, with relatively intact riparian zones. Altitude ranged from 65 to 275 m and conductivity from 153 to 569 μS cm<sup>-1</sup>.

We collected fish samples to represent the species composition and proportional abundance of the assemblage at each site. Fish were sampled by electrofishing 100-m stretches (200–350 V, 2–3 A, fully rectified triphasic DC), following the CEN standard (CEN 2003). Our sampling protocol (four-pass removal with block nets and unequal species catchability, estimated with the Program MARK) in streams of this region with our equipment showed: (i) capture probabilities in the first pass of 43–52%, 39–63% and 31–67% for *Barbus meridionalis*, *Salmo trutta*, and *Anguilla anguilla*, respectively; (ii) that the species richness observed in the first

pass is 100% of the richness estimated with four-passes and a gamma richness estimator (second-order jack-knife) in the trout zone (i.e. headwaters, where only *Anguilla* has been historically detected) and 77–100% in downstream reaches. Similar efficiencies of 85–100% of richness estimations with a single electrofishing pass in small streams have been reported by others (e.g. Patton *et al.*, 2000; Kennard *et al.*, 2006). Therefore, given these data, the small size of this stream (median depth and wetted width of 0.42 and 4.9 m, respectively during the sampling period), and the low fish richness of the basin (11 species), we are confident that our data accurately reflect fish abundance and assemblage structure and doubt that the metrics analysed (mainly richness and proportional abundances) are strongly affected by sampling biases among sites or species. All fishes stunned were collected with nets, anaesthetised with tricaine methanesulfonate (MS-222), identified to species, counted, measured (fork length in mm) and then returned to the stream.

Variation in flow regime among sites enabled us to examine the effects of severe water abstraction on the fish assemblage. Of the 10 sampling sites, three were frequently dry, apparently due to nearby water abstractions (site 2 was dry on 29% of the times sampled; site 3, 33%; site 6, 21%). Other sampling sites were never dry. We analysed data from six sampling sites similar in habitat features and species composition; we identified sites 2 and 3 as impacted and sites 4, 5, 9 and 10 as unimpacted. We hereafter refer to the latter as 'unimpacted sites' because the effects of water abstraction (as reflected in drought frequency) are much less severe, although they are not pristine in terms of flow regime. Fish assemblages at all six sites were dominated by Mediterranean barbel (*Barbus meridionalis*) and chub (*Squalius laietanus*), the two most abundant species in northeastern Catalonia (Aparicio *et al.*, 2000). Other sites were not included in the analyses because they had different habitat features and fish species composition: sites 6 and 7 were the most downstream and sites 1 and 8 were at higher altitude and dominated by brown trout (*Salmo trutta*) (Benejam *et al.*, 2008a).

#### Fish metrics

We developed a list of candidate metrics that we judged to be potentially useful in assessing the effects of water abstraction on fish assemblages. Candidate

metrics were based on seven species traits: geographic origin, flow affinity, feeding habitat, trophic guild, reproductive guild, tolerance and age at maturity (Table 1). Species assignments follow Pont *et al.* (2006) and Aarts & Nienhuis (2003). The categories for each attribute are described below.

*Geographic origin.* Species are either native or introduced to the study area.

*Flow affinity.* Species are either rheophilic, eurytopic or limnophilic. Rheophilic species prefer strong flow and clear water, using this habitat both for breeding and feeding. Eurytopic species tolerate a wide range of flow conditions. Limnophilic species prefer to live, feed and reproduce in slow-flowing to stagnant water.

*Feeding habitat.* Species are either benthic or water-column. Benthic species live, feed, and breed on or near the bottom; they rarely feed at the surface. Water-column species live and feed in the water column but may occasionally feed near the bottom.

*Trophic guild.* Species are either omnivores, insectivores or piscivores. The adult diet of omnivores consists of more than 25% plant material and more than 25% animal material (Schlosser, 1982). The adult diet of insectivores and piscivores consists of more than 75% insects (Lyons, Wang & Simonson, 1996) and more than 75% fish (Lyons *et al.*, 1996; Goldstein & Simon, 1998) respectively.

*Reproductive guild.* Two categories were recognised: lithophils and phytophils. Lithophilic species spawn exclusively on silt-free mineral substratum (e.g. sand, gravel, bedrock). Phytophilic species mainly spawn on vegetation (e.g. live plants or dead vegetation).

*Tolerance.* Two categories were recognised: tolerant and intolerant. Tolerant species thrive under a wide range of perturbed conditions, whereas intolerant species exhibit substantial restrictions in distribution and/or reduced abundance in response to anthropogenic impacts. In cases where the literature was contradictory or ambiguous, species were assigned to neither category.

*Age at maturity.* Two categories for reproductive maturity of females were recognised: <3 years and ≥3 years.

**Table 1** Species assignments for seven attributes used to define candidate metrics. Attributes and criteria for assignments are described in the text (see *Fish metrics* in Methods)

Common name	Scientific name	Family	Native to						Maturity age (years)
			Tordera basin	Flow affinity	Feeding habitat	Trophic guild	Reproductive guild	Tolerance	
Eel	<i>Anguilla anguilla</i> (L.)	Anguillidae	Yes	EURY	B	INSV	–	TOL	6
Mediterranean barbel	<i>Barbus meridionalis</i> Risso	Cyprinidae	Yes	RH	B	INSV	LITH	INT	2
Chub	<i>Squalius laietanus</i> Doadrio, Kottelat & de Sostoa	Cyprinidae	Yes	RH	WC	OMNI	LITH	–	2
Common carp	<i>Cyprinus carpio</i> L.	Cyprinidae	No	EURY	B	OMNI	PHYT	TOL	2
Mosquitofish	<i>Gambusia holbrooki</i> Girard	Poeciliidae	No	LI	WC	INSV	–	TOL	0.2
Pumpkinseed	<i>Lepomis gibbosus</i> (L.)	Centrarchidae	No	EURY	WC	INSV	–	TOL	2
Largemouth bass	<i>Micropterus salmoides</i> (Lacepède)	Centrarchidae	No	LI	WC	PISC	PHYT	TOL	3
Mullet	<i>Liza</i> sp.	Mugilidae	Yes	EURY	–	–	–	TOL	–
Brown trout	<i>Salmo trutta</i> L.	Salmonidae	Yes	RH	WC	INSV	LITH	INT	3
Rainbow trout	<i>Oncorhynchus mykiss</i> (Walbaum)	Salmonidae	No	RH	WC	–	LITH	–	3
Minnow	<i>Phoxinus</i> sp.	Cyprinidae	No	RH	WC	OMNI	LITH	–	1

Blank entries (–) mean that the information is not well known or contradictory in the bibliography.

Codes for assignments are as follows. Flow affinity: RH, rheophilic; EURY, eurytopic; LI, limnophilic; Feeding habitat: B, benthic; WC, water-column; Trophic guild: OMNI, omnivore; INSV, insectivore; PISC, piscivore; Reproductive guild: LITH, lithophilic; PHYT, phytophilic; Tolerance: TOL, tolerant; INT, intolerant.

We evaluated the ability of 30 candidate metrics to detect the effect of severe water abstraction on fish assemblages. Three types of metrics were examined: species richness and composition, trophic composition, and abundance (Table 2). Candidate metrics (see predicted response in Table 2) were based on previous studies about the effects of drought on fish assemblages (Lake, 2003; Magoulick & Kobza, 2003; Matthews & Marsh-Matthews, 2003; Magalhães *et al.*, 2007) and references therein and on previous efforts to assess biotic integrity in European streams (e.g. Oberdorff *et al.*, 2002; Angermeier & Davideanu, 2004; Pont *et al.*, 2006). We developed all candidate metrics from our categorisation of fish natural history (Table 1).

### Statistical analyses

Time series of environmental data are often autocorrelated, dominated by low-frequency variability, thus violating the independence assumption of most statistical tests (Bence, 1995; Pyper & Peterman, 1998). To test for trends in streamflow and other time series while accounting for serial autocorrelation, we used autoregressive models (Bence, 1995) with the maximum-likelihood method (AREG procedure in SPSS). In these

models, the first autoregressive parameter was significant ( $P < 0.05$ ) and the positive autocorrelation can be seen in Fig. 2 as short-term trends in the residuals.

To compare the observed streamflow with the naturalised flow predicted by the SAC-SMA model we used conventional linear regression. To compare limnological and habitat features of sites affected or not by water abstraction, we used two-way ANOVA, with sampling date (five dates with comprehensive data) as an additional factor to control for seasonal variation. To test the ability of fish metrics to detect impacts of water abstraction, we used generalised linear models (GLMs), which provide more appropriate error structures for data such as counts and proportions. The GLMs also had two factors: water abstraction (yes/no) and sampling date (15 dates from summer 2001 to autumn 2006). For species richness metrics and abundance data we used GLMs with Poisson errors and log-link functions. For proportional abundances, we used binomial errors and logit link functions. For fish lengths, we used gamma errors and identity link functions.

Finally, linear correlation and principal components analysis (PCA) were used to examine relations among fish metrics. Kaiser–Meyer–Olkin's measure of sam-

**Table 2** Candidate metrics evaluated for their ability to detect effects of severe water abstraction on fish assemblages in Mediterranean streams

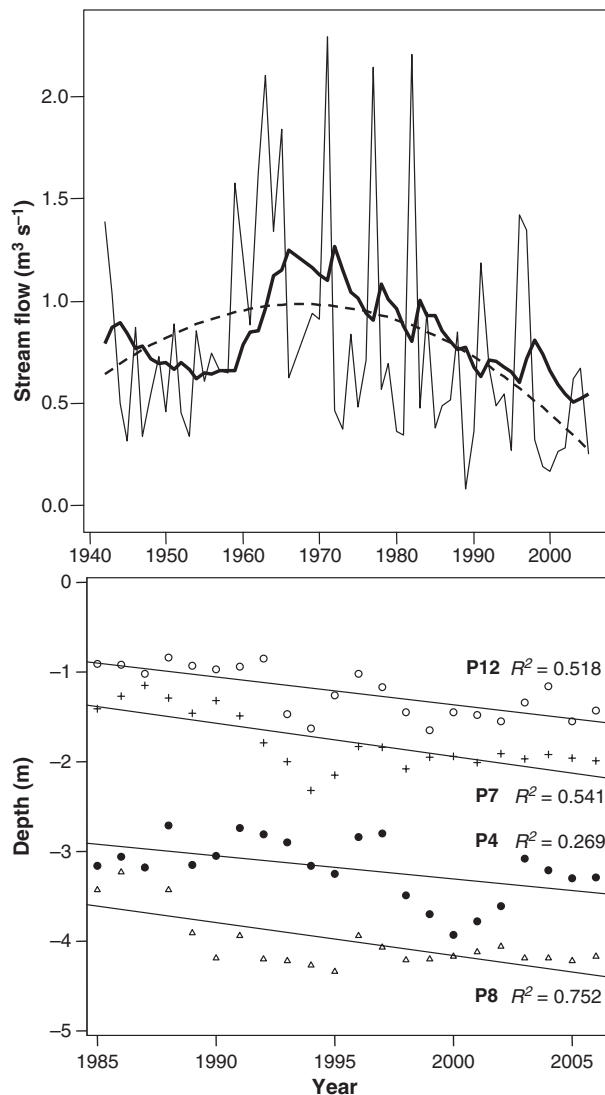
Fish metrics tested	Predicted response	<i>P</i> -value	Corrected <i>P</i> -value	Impacted sites	Unimpacted sites
<i>Species richness and composition</i>					
Number of families	–	0.067	0.286	1.27	2.09
Total number of species	–	0.118	0.444	2.44	3.35
Number of native species	–	0.050	0.251	1.82	2.76
Number of non-native species	+	0.275	0.688	0.00	0.00
Proportional abundance of native species	–	1.000	1.000	1.00	0.97
Proportional abundance of non-native species	+	1.000	1.000	0.00	0.03
Number of intolerant species	–	<b>&lt;0.001</b>	<b>&lt;0.001</b>	<b>1.00</b>	<b>1.23</b>
Number of tolerant species	+	0.381	0.816	0.11	1.11
Proportional abundance of intolerant species	–	<b>&lt;0.001</b>	<b>&lt;0.001</b>	<b>0.45</b>	<b>0.57</b>
Proportional abundance of tolerant species	+	1.000	1.000	0.03	0.07
Number of lithophil species	–	0.966	1.000	2.34	2.34
Proportional abundance of lithophil species	–	1.000	1.000	0.95	0.96
Number of rheophilic species	–	0.966	1.000	2.34	2.34
Number of eurytopic species	+	0.381	0.816	0.00	0.17
Proportional abundance of rheophilic species	–	1.000	1.000	0.95	0.96
Proportional abundance of eurytopic species	+	1.000	1.000	0.03	0.02
Average fork length of Mediterranean barbel	–	0.447	0.893	62.50	84.98
Average fork length of chub	–	0.183	0.524	66.73	109.46
Average fork length of minnow	–	0.784	1.000	48.88	51.19
Number of species maturing at <3 years	–	0.529	0.902	2.13	2.41
Number of species maturing at ≥3 years	–	0.541	0.902	0.00	0.02
<i>Trophic composition</i>					
Number of insectivore species	–	0.014	0.081	1.00	2.03
Number of omnivore species	+	0.870	1.000	1.39	1.35
Proportional abundance of insectivore species	–	0.192	0.524	0.38	0.24
Proportional abundance of omnivore species	+	0.185	0.524	0.62	0.76
Number of benthic species	–	<b>0.003</b>	<b>0.025</b>	<b>0.10</b>	<b>0.34</b>
Number of pelagic species	–/+	0.489	0.902	1.57	1.36
Proportional abundance of benthic species	–/+	0.623	0.934	0.37	0.22
Proportional abundance of pelagic species	–/+	0.623	0.934	0.63	0.78
<i>Abundance</i>					
Number of fish captured per site (CPUE)	–	<b>&lt;0.001</b>	<b>&lt;0.001</b>	<b>20.12</b>	<b>93.29</b>

The expected direction of response to water abstraction is given for each metric. ‘Proportional abundance’ refers to the number of individual fish of the named species’ category relative to the total fish in the sample. *P*-values are of generalised linear models comparing water abstraction group (sites impacted by strong water abstraction versus unimpacted sites), with sampling date as an additional factor. ‘Corrected’ *P*-values reflect application of the Benjamini & Hochberg (1995) procedure. Significant metrics ( $P < 0.05$ ) are highlighted in bold. Mean values for each metric for impacted and unimpacted sites are also given. All lengths are in mm.

pling adequacy (KMO) was used to assess the usefulness of a PCA. KMO ranges from 0 to 1 and should be >0.5 if variables are sufficiently interdependent for PCA to be useful.

Most measured variables (except conductivity and temperature) were log-transformed for linear models because homoscedasticity and linearity were clearly improved. To reduce problems with analysing proportional composition data, some categories (e.g. boulder + cobble) were pooled and redundant variables (adding to 100%) were excluded from the

analyses. In cases where multiple ANOVAs or GLMs were used, we corrected the *P*-values by using the procedure of Benjamini & Hochberg (1995), as implemented in the *R* package (R Development Core Team, 2008). The Benjamini & Hochberg (1995) procedure controls the false discovery rate (i.e. the proportion of erroneously rejected null hypotheses), balances type I and type II errors, and confers several other advantages (e.g. more statistical power and adaptation to the ‘amount of signal’ in the data) over more conventional procedures such as the Bonferroni correction



**Fig. 2** Upper panel: observed yearly average daily streamflow (—) versus year in Tordera stream. The best-fit time series model (—) and a quadratic model (---) are also shown. Lower panel: linear regressions of annual mean level of the aquifer, as measured by four piezometers, versus year (see Fig. 1 for locations).

(Garcia, 2004). All the other statistical analyses were performed with SPSS 15.0 (SPSS Inc., Chicago, IL, U.S.A.).

## Results

### *Streamflow: trends and natural regime*

The yearly mean daily streamflow of the Tordera stream (measured at Sant Celoni gauging station;

**Table 3** Linear regression models with first-order autoregressive errors of the yearly average aquifer level measured in 17 piezometers (see Fig. 1 for location) with year from 1976 to 2005

Piezometer	$t$	$n$ (years)
P1	-0.016	31
P2	-0.025	30
P3	-0.006	29
P4	-0.033*	22
P5	-0.051**	29
P6	-0.003	30
P7	-0.030**	22
P8	-0.038**	22
P9	-0.037**	22
P10	-0.038**	22
P11	-0.019*	31
P12	-0.023	27
P13	-0.023**	30
P14	-0.019**	30
P15	-0.007	31
P16	-0.034*	21
P17	-0.078**	21

The  $t$ -statistic and significance (\* $P \leq 0.05$ ; \*\* $P \leq 0.01$ ) of the regression coefficient of aquifer level as a function of year is given.

Fig. 1) has decreased (regression model with autoregressive errors;  $t_{58} = -2.264$ ,  $P = 0.027$ ) from  $1 \text{ m}^3 \text{ s}^{-1}$  in 1942 to  $0.5 \text{ m}^3 \text{ s}^{-1}$  in 2005 (Fig. 2). This decline has been more marked since 1960, as shown by the best-fit (Bayesian Information Criterion) time series model or a regular (linear regression) quadratic model (Fig. 2). The percentage of days that the stream was dry has also increased ( $t_{59} = 2.485$ ,  $P = 0.016$ ) and 11 of the 17 piezometers situated at mid-altitude (48–259 m above sea level) showed a significant decrease in the height of the aquifer over the last 30 years (1976–2005; Table 3; Fig. 2). By contrast, no significant decrease in precipitation has been observed during the same period (series at Sant Celoni town station from 1940 to 2006:  $t_{64} = -0.680$ ,  $P = 0.499$ ; series at Sant Esteve village station from 1985 to 2006:  $t_{19} = 0.351$ ,  $P = 0.730$ ; Fig. 3). The human population of the Tordera basin has increased over the last century ( $t_{22} = 5.691$ ,  $P = 0.000$ ; from 69 708 in 1940 to 371 387 in 2006), as has the number of industries ( $t_6 = 5.020$ ,  $P = 0.002$ ; from 3684 industries in 1994 to 4256 in 2002). According to the Catalan Water Agency (2002) the annual mean precipitation in the Tordera river basin is  $0.694 \text{ km}^3$ , the mean annual water yield of the stream is  $170.4 \text{ million m}^3$  and the quantity of abstracted water is around  $57.8 \text{ million m}^3$  per year. Therefore, about 34% of the water of Tordera stream

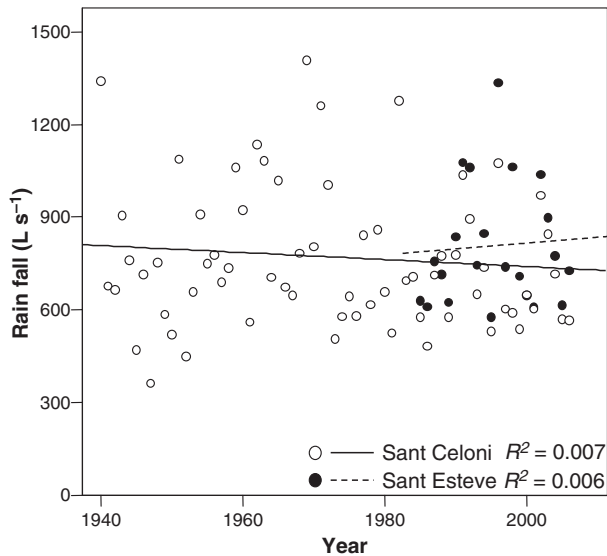


Fig. 3 Linear regressions of yearly average rainfall versus year at two sites (Sant Celoni, Sant Esteve) in the Tordera basin.

is abstracted for different uses (irrigation, industry and domestic).

Observed streamflow was weakly correlated with the naturalised flow predicted by the SAC-SMA model and observed flow was significantly less than predicted (slope significantly lower than 1; Table 4). On most of the occasions when the Tordera stream was dry (74 of 76 days), the model predicted that it should have been flowing, based on the precipitation and temperatures recorded. For example, except at the first sampling site, the observed yearly mean streamflow was lower than predicted, particularly at site 4

but also at site 6 (Fig. 4). Not surprisingly, differences were larger for medians and minima than for maxima. Notably, the model predicted a gradual increase in median and minimum flows along the river course, whereas at site 4 the median and minimum were lower than at site 1 (15 km upstream and 350 m higher in altitude), illustrating the strong impact of water abstraction in this area.

Results for the five other Mediterranean river basins were similar to those for Tordera stream. For all basins except Besòs, the comparison of observed and naturalised flow had slopes significantly lower than 1, i.e. flows were lower than expected (Table 4). In the largest river (Ter), which is also subject to heavy water abstraction, the annual mean streamflow has also decreased through time ( $t_{23} = -3.131$ ,  $P = 0.005$ ) and it was often dry when the SAC-SMA model predicted it to be flowing. The flow regime of the Ter river is strongly altered by an important withdrawal, with a maximum of  $8 \text{ m}^3 \text{ s}^{-1}$  diverted to the Barcelona region (Besòs and Llobregat river basins) since 1965. Downstream of the two large reservoirs from which the water is abstracted (c. 220 m of altitude), there is a strong decrease in the median (from c. 9 to  $5 \text{ m}^3 \text{ s}^{-1}$ ) and minimum flow of the river (but not the maximum). Flows continue to decline downstream, mostly due to abstraction for agriculture (Fig. 5).

#### Effects of water abstraction on fish metrics

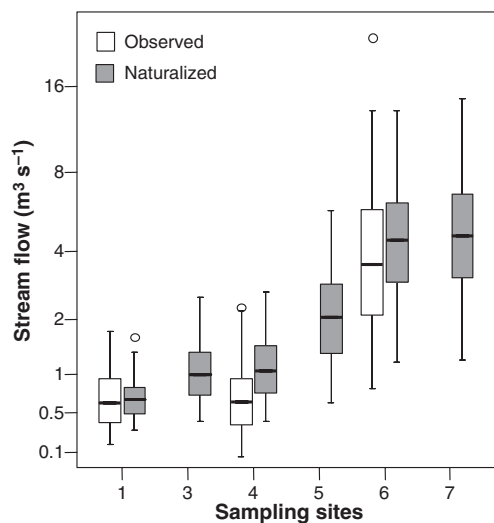
Similarities in physicochemical features between impacted and unimpacted sites in Tordera stream

Table 4 Comparison of the daily flow and number of dry days observed and predicted by the SAC-SMA model (see Methods) in six Mediterranean streams

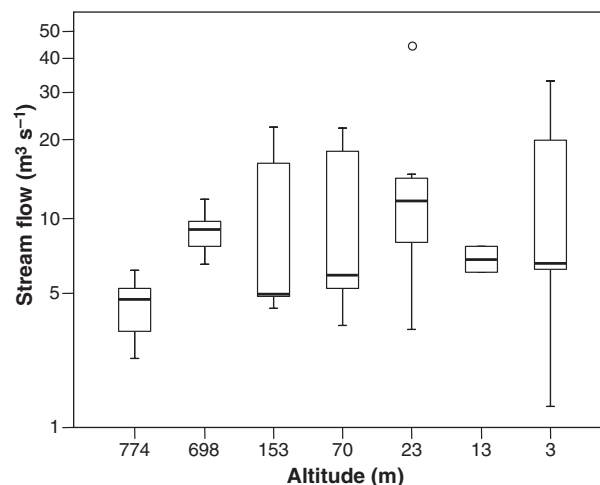
River basin	Year (range)	<i>n</i> (days)	Median daily flow ( $\text{m}^3 \text{ s}^{-1}$ )		Dry days		$r^2$	<i>P</i>	<i>b</i>	SE	<i>P</i>
			Observed	Predicted	Observed	Predicted					
Besòs	1968–98	9726	2.8	0.5	46	137	0.515	<0.001	1.026	0.010	0.009
Fluvià	1942–98	18 627	3.6	4.5	1	0	0.583	<0.001	0.757	0.005	<0.001
Llobregat	1942–99	19 066	10.9	11.3	0	0	0.630	<0.001	0.734	0.004	<0.001
Muga	1942–97	20 087	0.8	1.1	1	0	0.152	<0.001	0.395	0.007	<0.001
Ter	1967–92	8766	5.7	12.7	860	0	0.084	<0.001	0.410	0.014	<0.001
Tordera	1942–2000	19 500	0.3	0.4	76	2	0.501	<0.001	0.609	0.004	<0.001

The statistics of the linear regression between the observed and predicted daily streamflows are also shown. The first *P*-value tests whether there is correlation between both flows (but note the low  $r^2$ ), whereas the second *P*-value corresponds to the *t*-test of whether the regression coefficient or slope (*b*) is significantly different from 1. For five rivers the observed flow is significantly less than that predicted by the model. See Fig. 1 for the location of gauging stations (the second station corresponds to Tordera stream; the fourth station corresponds to Ter river).





**Fig. 4** Box-plot of observed and predicted natural annual mean daily streamflow along sampling sites in Tordera stream (see Fig. 1 for locations) from 1991 to 2000. Each box corresponds to 25th and 75th percentiles; the dark line inside each box represents the median; error bars show the minima and maxima except for outliers (open circles, corresponding to values >1.5 box-heights from the box).



**Fig. 5** Box-plot of observed yearly annual mean daily stream-flow at seven gauging stations along the Ter River. See Fig. 4 for box format.

allowed us to isolate the effects of water abstraction on fish metrics. Among the habitat features, unimpacted and impacted sites did not differ for most

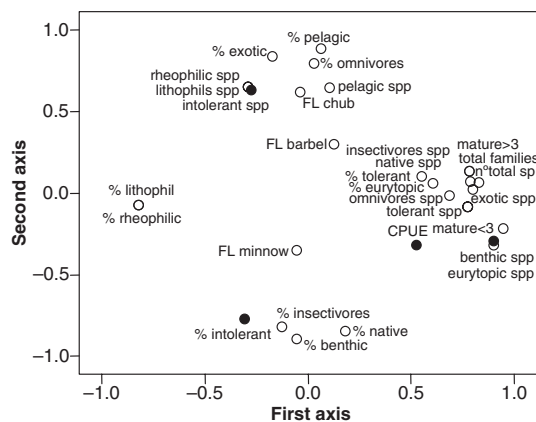
**Table 5** Results of the ANOVAS of limnological and habitat features, with date and water abstraction impact (yes/no) as factors

	<i>F</i>	d.f.	Corrected <i>P</i> -value	Impacted sites	Unimpacted sites
<i>Habitat features</i>					
Geomorphology: % pools	0.09	1, 37	0.771	18.57	13.84
Geomorphology: % runs	0.48	1, 37	0.631	51.43	66.98
Geomorphology: % riffles	0.17	1, 37	0.746	29.81	19.39
Substratum: % cobble + boulder	29.69	1, 37	<b>&lt;0.001</b>	88.71	46.47
Substratum: % gravel	12.33	1, 37	<b>0.007</b>	11.05	41.05
Substratum: % silt + detritus	22.03	1, 37	<b>&lt;0.001</b>	0.00	2.30
Refuges: % boulder	3.21	1, 37	0.162	37.90	28.49
Refuges: % hydrophytes	0.35	1, 37	0.644	11.29	11.05
Refuges: % total refuges	0.09	1, 37	0.771	71.10	80.07
Vegetation: % bushes + trees	10.07	1, 37	<b>0.013</b>	82.48	75.36
Vegetation: % hydrophytes	0.51	1, 37	0.631	6.10	4.02
<i>Limnological features</i>					
log ammonium (mg L <sup>-1</sup> )	1.64	1, 13	0.327	0.03	0.08
log nitrites (mg L <sup>-1</sup> )	10.23	1, 13	<b>0.022</b>	0.01	0.02
log nitrates (mg L <sup>-1</sup> )	0.45	1, 13	0.631	0.35	0.37
log phosphates (mg L <sup>-1</sup> )	22.45	1, 13	<b>&lt;0.001</b>	0.10	0.14
log sulphates (mg L <sup>-1</sup> )	12.11	1, 13	<b>0.015</b>	1.15	1.61
log chlorides (mg L <sup>-1</sup> )	8.72	1, 13	<b>0.027</b>	1.09	1.65
log oxygen (mg L <sup>-1</sup> )	2.56	1, 13	0.226	0.99	0.92
log suspended solids (mg L <sup>-1</sup> )	2.94	1, 13	0.202	0.93	1.14
Temperature (°C)	5.22	1, 13	0.087	16.88	18.93
Conductivity (µS cm <sup>-1</sup> )	9.72	1, 13	<b>0.022</b>	139.04	511.38
log pH	1.74	1, 13	0.327	0.92	0.93

Corrected *P*-values reflect application of the Benjamini & Hochberg (1995) procedure. Significant values ( $P < 0.05$ ) are highlighted in bold. Mean values of habitat features for impacted and unimpacted sites are also given. Log-transformation was  $\log_{10}(x + 1)$ .

structural variables, such as geomorphology and existence of refuges (Table 5). In contrast, substratum composition differed markedly because coarser substratum predominated at the impacted sites. Because the average altitude of unimpacted sites was slightly lower (113 m a.s.l.) than for impacted sites (205 m), conductivity (and marginally temperature) were significantly higher in the former (Table 5). This was also reflected in nutrient concentration (Table 5) but the overall differences were small. We expected these differences in water chemistry and temperature to have negligible effects on fish metrics. There was no significant date  $\times$  impact group interaction ( $P > 0.05$ ) for any of the habitat or limnological characteristics, suggesting that the overall differences between unimpacted and impacted sites did not vary strongly with time.

Of the 30 candidate metrics tested (Table 2) as potential indicators of water abstraction effects, four had significant effects after applying the false discovery rate correction: number of intolerant species, proportion of intolerant individuals, number of benthic species and catch per unit effort (CPUE). Three other metrics (number of families, number of native species, and number of insectivore species) showed significant or marginally significant effects prior to applying the correction. Although many of the metrics were significantly correlated, different groups of metrics could be identified and the four metrics



**Fig. 6** Principal components analysis of the 30 candidate fish metrics. The two axes explained 34.4% and 24.6% of the variance, respectively, in metrics among sites in Tordera stream. Fish metrics significantly different between unimpacted and impacted sites (see Table 2) by water abstraction are shown as filled circles.

discriminating between impacted and unimpacted sites were not completely redundant.

Most of the variables were significantly correlated and KMO (=0.57) indicated the usefulness of a PCA, which explained 59.1% of the variation among sites with two axes (Fig. 6). The first axis of the PCA highlighted the many metrics based on species richness (such as the number of benthic species) which were also related to different metrics such as CPUE and proportional abundance of rheophilic or lithophilic species. The second axis corresponded primarily to metrics based on species composition and included the two others related to abstraction (the number of intolerant species and the proportional abundance of intolerant species). Accordingly, the number of benthic species and CPUE were correlated with each other ( $r = 0.475$ ,  $n = 63$ ,  $P < 0.001$ ) but not with the number of intolerant species or proportional abundance of intolerant species ( $r < 0.12$ ), which were themselves significantly correlated ( $r = 0.499$ ,  $P < 0.001$ ). Therefore, these four metrics, which seem to discriminate between severe versus moderate impacts of water abstraction, are not redundant and correspond to different properties of the fish assemblage (fish abundance, number of species and species composition).

Moreover, the fish assemblage of Tordera stream continues to change in response to chronic water abstraction. There was a significant change in two fish metrics at the impacted sites during the 6 years of our sampling: a decrease in the proportion of intolerant individuals ( $t_9 = -2.706$ ,  $P = 0.024$ ) and the number of benthic species ( $t_9 = -2.533$ ,  $P = 0.032$ ), whereas no temporal trend in any other metric was observed at the unimpacted sites ( $P > 0.11$ ).

## Discussion

### *Water abstraction in Mediterranean streams*

Our analysis indicates that water abstraction has changed the flow regime of Tordera stream substantially. We have shown a significant decrease since the 1960s in the streamflow and aquifer height of Tordera stream, despite no observed decrease in precipitation. The number of days that the stream is dry has increased and the streamflow observed is well below that predicted by the SAC-SMA model. Meanwhile, the amount of water abstracted, and the numbers of

inhabitants and industries in the river basin has increased steadily. Part of this decline in streamflow may be due to climate variation, since a humid period around 1960–70 and a subsequent dry phase has been shown in several European rivers (Probst, 1989). However, two reasons suggest that this alone does not explain the patterns observed in Tordera stream: (i) we found no significant trend in precipitation, as is well known for this region (Lopez-Bustins, Martin-Vide & Sanchez-Lorenzo, 2008); and (ii) the SAC-SMA model predicts a naturalised flow much higher than that observed (Table 4). Moreover, the overexploitation of the water in the Tordera basin has been previously described. In particular, there is a water abstraction of 57.8 million  $\text{m}^3 \text{year}^{-1}$  (particularly in summer) for a mean annual water yield of 170.4 million  $\text{m}^3 \text{year}^{-1}$  (ACA, 2002), and seawater intrusion has been increasing in its delta in recent decades, as in other coastal regions in Catalonia (Mas-Pla, Montaner & Solà Martínez & Murillo, 2003).

Water abstraction is even stronger for another of the rivers studied (Ter) and similar but less severe for the other river basins: all but one (Besòs) have naturalised flows higher than those observed, with more dry days observed than predicted. River Ter has also shown a decrease in flow during the last century. The Ter is another Mediterranean stream with enormous human pressure for water abstraction. With a mean annual water yield of 845 million  $\text{m}^3 \text{year}^{-1}$  and a mean discharge of  $c. 10 \text{ m}^3 \text{ s}^{-1}$  (ACA, 2002), much water is abstracted from its middle reaches (a maximum of  $8 \text{ m}^3 \text{ s}^{-1}$ ; an average of  $6.1 \text{ m}^3 \text{ s}^{-1}$  and a total of  $193 \text{ hm}^3$  for 2006) to supply drinking water to the Barcelona region (in the Llobregat and Besòs basins), inhabited by 4 million people (Sabater *et al.*, 1992; Benejam *et al.*, 2008b). Notably, the only exception to the general pattern is the Besòs, a small basin near Barcelona that receives part of the water abstracted from the Ter, and thus now has more flow and fewer dry days than predicted by the SAC-SMA model (Table 4).

Many rivers in the Mediterranean basin are experiencing alterations to their natural flow regimes and drought frequencies similar to those reported here, including many Iberian rivers such as Júcar (Sánchez Navarro *et al.*, 2007), Guadiana (Pires, Cowx & Coelho, 1999; Collares-Pereira *et al.*, 2000), Ebro (Prat & Ibañez, 1995), and Duero and Tajo (Trigo *et al.*, 2004; Sabater *et al.*, 2009). Human population growth is

rapidly depleting available freshwater supply in many other areas of the world (Richter, Mathews & Wigington, 2003). During the 20th century, the global human population increased fourfold to more than six billion ( $6 \times 10^9$ ), whereas water withdrawn from natural freshwater ecosystems increased eightfold over the same period (Gleick, 1998; Richter *et al.*, 2003). The pressure on water availability is obviously higher in dry countries, such as those in the Mediterranean basin. For example, the average European *per capita* water consumption is  $207 \text{ m}^3 \text{ year}^{-1}$ , but in Mediterranean countries is higher (e.g.  $530 \text{ m}^3$  in Spain,  $523 \text{ m}^3$  in Italy and  $334 \text{ m}^3$  in Greece; Vergés, 2002).

Altered flow regimes due to water abstraction or other causes complicate assessments of river health. For example, one commonly overlooked implication of altered flow regimes in Mediterranean rivers is related to interpreting biotic indices of river health when a river is dry because of water withdrawals. In these cases, biotic indices (based on either macroinvertebrates, fish or other taxa) are invariably not computed. For instance, in Tordera stream (at site 3 in Fig. 4) from 2001 to 2006 macroinvertebrate and diatom indices were not computed on four occasions because the river was dry (Benejam *et al.*, 2008a), although it should have been flowing according to the SAC-SMA model. We suggest that a detailed characterisation of the natural flow regime should precede the application of biotic indices and that, in cases of rivers dry due to human impacts, the lowest possible score for biotic indices (but specifying that the river was artificially dry) be applied to reflect real impairment. Otherwise, reporting high-quality scores on other occasions will overestimate the average ecological status of the river. As far as we know, this problem has been neglected in the vast literature applying biotic indices in Mediterranean-type rivers (perhaps because this problem is much rarer in central and northern Europe, where water is more available). For instance, neither the recently developed European Fish Index (Pont *et al.*, 2006) nor Iberian studies (Ferreira *et al.*, 2007) mention this problem, despite including the same Catalan rivers that we studied. Although the choice of not computing biotic indices may be inevitable if no information on the flow regime is available, we suggest an assessment approach in which the natural flow regime is characterised *before* the application of biotic indices.

*Effects of severe water abstraction on fish assemblages*

We detected four fish metrics (catch per unit effort, number of benthic species, number of intolerant species and proportion of intolerant individuals) that distinguished between sites impacted by water abstraction and unimpacted sites and that showed significant temporal trends in response to abstraction. Although this analysis should be regarded as preliminary and needing further data and spatial validation, the relationships of these metrics to water abstraction are plausible and agree with previous studies (e.g. Lake, 2003; Magoulick & Kobza, 2003; Magalhães *et al.*, 2007). We would expect low fish abundance (catch per unit effort) in a site affected by water abstraction, given that drought reduces survival and reproduction and promotes emigration of many stream fishes (Matthews & Marsh-Matthews, 2003; Keaton *et al.*, 2005; Davey & Kelly, 2007). The lower number of benthic species observed at impacted sites was mediated by a decrease in the Mediterranean barbel (assigned as native) at these sites and its replacement by the minnow (a non-native, water-column species). Among the most frequent biological effects of drought are population declines and changes in community composition (Magoulick, 2000; Marchetti & Moyle, 2001; Matthews & Marsh-Matthews, 2003), including decrease in the number and proportional abundance of intolerant species (Danehy *et al.*, 1998; Lake, 2003; Magalhães *et al.*, 2007), as observed here.

These four significant fish metrics, and probably others that in our study were almost significant (number of insectivore species, number of native species, number of families), may be used to assess rivers suspected to have problems with abstraction. Some of these fish metrics are already used in existing European IBIs (Sostoa *et al.*, 2003; Angermeier & Davideanu, 2004; Pont *et al.*, 2006), so our results can help to understand their behaviour. In particular, low collective values of these fish metrics may warn of substantial hydrologic alteration.

Natural drought is a seasonally predictable event in Mediterranean ecosystems (Gasith & Resh, 1999), to which native species have adapted through evolutionary history (Moyle, 1995; Poff, 1997). However, population persistence is fragile in such harsh environments, and an increase in the frequency or severity of drought due to water abstraction may provoke serious problems for the conservation of

Mediterranean and semiarid native fish assemblages (Magalhães *et al.*, 2007). The IUCN (Smith & Darwall, 2006) has already identified water abstraction as one of the most important environmental issues in Mediterranean freshwaters, and the cause of four of the eight freshwater fish extinctions in the Mediterranean basin. As water abstraction increases the frequency, severity and duration of droughts in this region, we expect threats to fish to become increasingly widespread.

In conclusion, we have shown that some Mediterranean streams are profoundly altered by water abstraction, with a significant decrease in streamflow throughout the 20th century and an increase in the number and severity of droughts. We suggest that a detailed characterisation of the natural flow regime should precede the application of biotic indices and that, in cases of artificially dry rivers, the lowest possible score for biotic indices (but specifying that the river was artificially dry) be applied to reflect real impairment to ecological status. Our results show that the effects of altered flow regimes on stream-fish assemblages are clearly detectable, including reduced population densities, fewer benthic species, and reduced occurrence and abundance of intolerant species. These biotic responses, if properly calibrated, can be developed into management tools to assess the impacts of water abstraction and to inform stream conservation and restoration programmes.

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