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# Habitat quality affects the condition of *Barbus sclateri* in Mediterranean semi-arid streams

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# **Synopsis**

We investigated the site level relationships between fish condition and environmental variables in Sclater's barbel, *Barbus sclateri*, from semi-arid streams in the south-eastern Iberian Peninsula. These freshwater ecosystems are characterized by strong seasonal fluctuations in flow levels (droughts and floods) as a consequence of their irregular hydrological regime on both an annual and pluriannual scale. We analysed the relationships between 11 ecosystem variables [conductivity, oxygen concentration, water temperature, pH, seasonal water flow, dominant substrate, submerged vegetation, QBR (Riparian Ecosystems Quality Index), BMWP' (Spanish version of the Biological Monitoring Working Party), fish refuge index and fish density] and fish condition by comparing mass–length relationships of nine populations of Sclater's barbel located in five sub-basins with different environmental conditions. Fish condition differed between the populations studied and was mainly dependent on the ecological variables related with water flow and, consequently, the physical structure of the streams, which is directly related to substrate and fish refuge.

# Introduction

Mediterranean streams are unstable ecological systems subject to high water temperatures and droughts in summer, and unpredictable winter floods (Giudicelli et al. 1985). In the south-east of the Iberian Peninsula, such ecological conditions are drastic and these semiarid streams are exposed to natural disturbances as a consequence of their irregular hydrological regime (Vidal-Abarca et al. 1992). The dominant fish species in such streams is the cyprinid *Barbus sclateri* Günther, 1868 (Sclater's barbel), an endemic fish of the mid-south of the Iberian Peninsula (Doadrio 2001).

Several authors have pointed to the fact that fishes inhabiting variable ecosystems are able to adjust some of their life history traits in response to different ecological conditions (Hutchings & Myers 1994). Many studies of Iberian fish species have shown how environmental fluctuations greatly influence the condition of barbels, and how most of barbel populations exhibit a seasonal dynamic of condition (Herrera et al. 1988, Herrera & Fernández-Delgado 1992, Encina & Granado-Lorencio 1997a, Torralva et al. 1997, Aparicio & Sostoa 1998, Miñano et al. 2000, Soriger et al. 2000). However, there have been few attempts to investigate how the relative influence of these factors varies in Mediterranean streams (Vila-Gispert et al. 2000, Vila-Gispert & Moreno-Amich 2001) and only one study has referred to semi-arid streams (Torralva et al. 1997).

We analysed the relationships between 11 environmental variables and fish condition by comparing several populations of Sclater's barbel located in the most arid zone of its distribution range. As well as contributing scientific data, the information gained in this study may be important for future recovery programmes which, it is believed, will soon be necessary for many Mediterranean cyprinid species because of the high rate of destruction of their habitats (Crivelli & Maitland 1995). 14

## Material and methods

Sclater's barbel is one of the most abundant endemic fishes of the mid-south of the Iberian Peninsula including the Guadiana, Guadalquivir and Segura rivers basins (Elvira 1995, Doadrio 2001). This benthic species reproduces between May and July (Herrera & Fernández-Delgado 1992, Rodríguez-Ruiz & Granado-Lorencio 1992, Torralva et al. 1997, Soriguer et al. 2000). The populations studied inhabited five streams with different hydrological regimes located in Sector III (Mas 1986) of the Segura river basin, which covers a drainage area of about 14,432 km<sup>2</sup> in the south-east of the Iberian Peninsula (Figure 1). Except for the Segura stream, the singularity of sampling streams is due to hydrological regimes, which are very variable on both spatial and temporal scales, so we sampled in channels with continuous flow but with seasonal water level fluctuations and channels with discontinuous flow in the summer and reduced to isolated pools, all of them less than 3.5 m wide (annual mean). The Segura stream was the main channel (6 m wide) and presented less drastic flow fluctuations.

The term semi-arid stream is applied to streams in regions where the water balance is negative, creating an environmental stress which, unlike those occurring in arid lands or deserts, is neither permanent nor predictible (Vidal-Abarca et al. 1992). These streams are subject to natural disturbances (droughts and floods), as a consequence of their irregular hydrological regime on both annual and pluriannual scale. In Spain, only two regions exist with semi-arid climate characteristics, one in the central region of the Ebro depression and the other in the south-east, where the analysed populations are located.

A total of 927 Sclater's barbels from nine total sampling sites of the Luchena, Mula, Quípar, Benamor and Segura streams were analysed (Figure 1). Whenever it was possible, sampling sites were selected to represent the main flows of each tributary. Individuals were captured by electrofishing at nine sites in November 1998. Limiting sampling to this period avoided the capture of prespawning and spawning fish in this basin (Torralva et al. 1997), and ensured that variations in body condition were not affected by gonad development (Herrera & Fernández-Delgado 1994, Encina & Granado-Lorencio 1997a,b).

We collected fish with standard AC electrofishing equipment (1800 W generator, working voltage between 200 and 350 V, 2–3 A). Two fishermen with electric dip-nets supported by two assistants each with a non-electric dip-net, removed fish from the lower to the upper part of each sampling stretch (100 m long), which was blocked off with barrier nets. Immediately after catching, we placed the fish in nets outside the sampling stretch and away from any electrical influence. Triplicate fishing was carried out to keep the effort constant, and to satisfy the assumptions of catch-effort methods (Seber 1982).

After capture, we anaesthetized fish with benzocaine and measured standard length ( $\pm 1 \text{ mm}$ ) and total mass ( $\pm 0.1 \text{ g}$ ) for each individual. Fish smaller than 75 mm [<2+ age class, in García de Jalón et al. (1999)] were excluded from the analysis to avoid possible differences in body shape between juveniles and adults (Murphy et al. 1990) and to minimize measurement errors associated with weighing small fish in the field (Vila-Gispert & Moreno-Amich 2001). We estimated the number of fish by the Zippin method (Zippin 1958), which is particularly suited for triplicate fishing.

We characterized each sampling site by 11 environmental variables and indices relating to water quality (fortnightly mean values), the physical state of stretches, possible resources exploited by fish populations and possible intraspecific interactions: conductivity, oxygen concentration, water temperature, pH, seasonal water flow, dominant substrate, amount of submerged vegetation, QBR [Riparian Ecosystems Quality Index *sensu* Munné et al. (1998)], BMWP' [Spanish version of the Biological Monitoring Working Party *sensu* Hellawell (1978)] (Alba-Tercedor & Sánchez-Ortega 1988), fish refuge index [*sensu* García de Jalón & Schmidt (1995)] and fish density (Table 1).

We classified seasonal water flow and dominant substrate sensu Vila-Gispert et al. (2000). Seasonal variation in water flow was considered high when flow ceased in the summer and the stream was reduced to small isolated pools, moderate if the flow was continuous but with water level fluctuations in accordance with the wet-and-dry cycle, and low if the flow remained relatively constant throughout the year. We classified dominant substrate according to the size of different particles: sand (100% sand, 2-5 mm), muddy-sandystony (equal percentage of mud, 1-2 mm, sand and stones, 25–100 mm), sandy-stony (more than 50% sand, the rest stones), stony-sandy (more than 50% stones, the rest sand) and stony (100% stones). We classified submerged vegetation as absent (no aquatic vegetation), little (covering less than 10% of the stretch), poor (covering more than 10% and less than 30% of the stretch), disturbed (covering more than 30% and less than 60% of the stretch), and developed



*Figure 1*. Sampling sites from the Segura river basin, south-eastern Iberian Peninsula. Luchena-1: 1; Luchena-2: 2; Mula-1: 3; Mula-2: 4; Quípar: 5; Benamor: 6; Segura-1: 7; Segura-2: 8 and Segura-3: 9.

(it covered more than 60% of the stretch). We obtained QBR data of Suárez & Vidal-Abarca (2000).

The statistical analysis used to compare fish condition followed that used in two studies of *Barbus meridionalis* Risso, 1826 (Vila-Gispert

et al. 2000, Vila-Gispert & Moreno-Amich 2001) and proposed by García-Berthou & Moreno-Amich (1993), which is based on the application of univariate analysis of covariance (ANCOVA) using total mass as the dependent variable and standard length

1 (moderate (absent), 2 (1	), 2 (contin little), 3 (po	nous flow); Don or), 4 (disturbed)	ninant substrate: ), 5 (developed).	1 (sand), 2 (mu	ddy-san	ıdy-stony), 3	3 (sandy-ston)	y), 4 (stony-sa	ndy), 5 (	(stony); Sub	merged veg	getation: 1
Sampling site	Sample size	Conductivity $(\mu s  cm^{-1})$	Oxygen concentration (ppm)	Water temperature (°C)	Hq	Seasonal water flow	Dominant substrate	Submerged vegetation	QBR	BMWP'	Refuge index	Fish density (ind m <sup>-2</sup> )
Luchena 1	118	2,620	6.44	23.5	8.48	1	2	1	50	84	3.00	0.90
Luchena 2	118	2,900	11.75	11.8	9.18	0	1	2	50	76	1.00	0.69
Mula 1	23	700	8.14	16.1	6.75	1	1	1	45	76	4.25	1.53
Mula 2	39	930	8.38	13.9	6.16	0	1	1		6	2.50	1.94
Quípar	87	1,310	7.66	6.1	9.14	0	1	5	85	51	4.50	5.42
Benamor	39	50	11.70	6.4	9.45	1	4	4	80	95	3.40	0.87
Segura 1	212	560	9.94	13.4	8.12	2	3	2	50	85	3.75	0.70
Segura 2	42	660	11.60	17.2	8.27	2	ю	1		90	3.75	0.79
Segura 3	249	700	12.04	17.0	7.51	2	3	2	34	83	4.00	0.03

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as the covariate. The relationship between total mass and length was clearly non-linear, but was linear after log-transformation (ln). Therefore, we used the ln-transformation of total mass as dependent variable and ln-transformation of standard length as the covariate. We tested the homogeneity of the regression coefficients (slopes) of the dependent–covariate relationship with an ANCOVA design that analysed the pooled covariate-by-factor interaction. If the covariateby-factor interaction (homogeneity of slopes) was not significant (p > 0.05), we developed a standard ANCOVA to test significant differences in parameter a (the y-intercept) between populations.

We performed a stepwise multiple-regression analysis to determine the amount of variation in parameter a (the y-intercept) associated with environmental variables. We also analysed bivariate relationships among environmental variables using Pearson's correlations for categorical variables (seasonal water flow, dominant substrate and submerged vegetation). We performed the statistical analyses with the SPSS<sup>®</sup> software package and accepted a significant level of 0.05.

# Results

Sclater's barbel was the only fish species living in four sampling sites studied and the dominant fish species (80–100%) in the rest, coexisting with *Chondrostoma polylepis* Steindachner, *Gobio* gobio (Linnaeus), *Squalius pyrenaicus* (Günther) (= *Leuciscus pyrenaicus*) and *Oncorhynchus mykiss* (Walbaum), depending on the stream in question. This species has been shown to be dominant in the communities of the Segura river basin (García de Jalón et al. 1999).

Parameters of the mass–length relationship in each population studied are presented in Table 2 and the results of the ANCOVA are shown in Table 3. There was a significant degree of homogeneity (p = 0.209) between sampling sites on slope (b) of the relationships between total mass and standard length (Preliminary design, Table 3), although the y-intercept (a) varied significantly (p < 0.0005) between sampling sites (Final design, Table 3). Benamor and Segura streams showed the highest fish condition, while Luchena-2 sampling site showed the lowest value (y-intercept higher and lower respectively, Table 2). As a result, sampling sites could be differentiated according to differences in parameter a of the mass–length relationship.

Correlations between parameter a of the masslength relationship and environmental variables and intercorrelations between these variables are presented in Table 4. Conductivity, seasonal water flow, dominant substrate, BMWP' and refuge index present significant correlations (Pearson's coefficient or Spearman's coefficient, according to the particular case) with the parameter a.

The first multiple-regression model showed that seasonal water flow accounted for most of the variation (64.1%) of parameter a in the mass–length relationship (Table 5). However, due to the great weight of this variable in the model (no other variables can enter into the model) and because seasonal water flow showed a significant correlation with dominant substrate (Spearman coefficient = 0.76), we performed a second multipleregression model not involving seasonal water flow. This second model indicated that dominant substrate and refuge index are the most important variables

Sampling site	n	b (slope)	a (the y-intercept)	r	Mean ± CL of standard length (mm)
Luchena 1	118	2.95	-11.11	0.994	$128.3 \pm 10.3$
Luchena 2	118	3.11	-11.90	0.986	$114.6 \pm 4.0$
Mula 1	23	3.00	-11.22	0.998	$110.3 \pm 24.3$
Mula 2	39	2.95	-11.38	0.998	$139.6 \pm 16.9$
Quípar	87	2.99	-11.21	0.994	$143.2 \pm 10.5$
Benamor	39	2.92	-10.79	0.981	$93.3 \pm 4.4$
Segura 1	212	2.98	-10.83	0.990	$171.7\pm5.2$
Segura 2	42	2.94	-10.93	0.971	$124.5 \pm 4.5$
Segura 3	249	2.99	-10.51	0.992	$128.1\pm2.9$

*Table 2.* Regression (a, b) and correlation coefficients (r) of the ln-transformed mass–length relationship in each population studied.

*Table 3.* ANCOVA analyses of the mass-length relationship in *Barbus sclateri*: F-statistics, degrees of freedom (df) and p values. All variables (dependent and covariate) were ln-transformed. Standard length is the covariate.

Source of variation	F	df	р
Preliminary design (test for i	interaction)		
Length	18969.87	1,926	< 0.0005
Sampling site	1.31	8,926	0.234
Length × sampling site	1.36	8,926	0.209
Final design (no interaction)			
Length	62029.87	1,926	< 0.0005
Sampling site	35.38	8,926	< 0.0005

for explaining the variation in parameter a between sampling sites (89.0%) (Table 5).

#### Discussion

To investigate inter-population variations in fish condition, mass-length relationship analysis have proved to be a good alternative to relative mass indices (ratio-related techniques), assuming that the genetically determined mass-length slope does not vary between these populations and that the slope is homogeneous for the populations on a local level (Sutton et al. 2000). Relative mass indices do not normally fulfil these underlying assumptions, and have been criticized on statistical grounds (Bolger & Connolly 1989, Jakob et al. 1996). In fact, the adjustment of size variation in the data by regression-related techniques has recently been used with ANCOVA approach providing valid results (Vila-Gispert et al. 2000, Vila-Gispert & Moreno-Amich 2001).

Our results showed that the condition of Sclater's barbel differed between the populations studied. Any differences in parameter a of the mass–length relationship were probably caused by differences in habitat conditions. Although the condition of Sclater's barbel in the south of the Iberian Peninsula varies seasonally with reproductive cycles, water temperatures and temporal ecosystem productivity (Herrera & Fernández-Delgado 1992, Rodríguez-Ruiz & Granado-Lorencio 1992, Torralva et al. 1997), the fact that the sampling period was short and in November prevented capture of prespawning and spawning fish and thus ensured that differences in parameter a between sampling sites were not the result of changes in these variables.

Direct or indirect interaction effects at stream level are often ignored in multiple-regression models when relating site level habitat variables to fish population characteristics (Dunham & Vinyard 1997). It should be noted that when the analyses proposed by Dunham and Vinyard (1997) are applied, stream effects may be too weak to be detected if no more than three sites per stream are sampled. Nevertheless, because any variability in fish-condition data (standard residuals of mass–length relationship) was shown to be lower between sampling streams than among sampling sites (ANOVA test; sampling site as a factor: F(8,926) =55.97; sampling stream as a factor: F(4,926) = 35.55), it is clear that our site level model explains more variation than a hypothetical stream model.

In such site level analysis of habitat–fish condition relationships, seasonal water flow, dominant substrate and fish refuge index (significant variables in the multiple-regression analyses) were the ecological variables that best correlated with Sclater's barbel condition. Sampling sites with a continuous water flow, the greatest diameter of its dominant substrate and the highest fish refuge values provided the highest values for parameter a, demonstrating the better fish condition.

In similar previous studies with *B. meridionalis* (Vila-Gispert et al. 2000, Vila-Gispert & Moreno-Amich 2001), oxygen concentration and riparian cover were found to be the ecological variables that accounted for most of the variation in fish condition between populations, the authors describing periodic scouring and low flows as the most important factors affecting fish condition. These findings and the results of the present study, point to seasonal water flow as an important factor in the condition in both species.

Certainly, flowing water is one of the fundamental attributes of lotic ecosystems, and has profound effects on physical, chemical and biological characteristics (Schlosser 1995). Sampling sites with drastic intermittent flow regimes (high or moderate seasonal water flow: Luchena, Mula, Quípar and Benamor), contained small pool refugia and exhibited substantial temporal variations in physical and chemical characteristics. Except in the Benamor stream, where the effects were less drastic, flows ceased in the summer and the streams became small isolated pools. In such pools, physical, chemical and biotic fluctuations are common and often extreme. Environmental stress is greater due to water pollution and water extraction for agricultural purposes. During this period, most of the fish living in these streams become concentrated into these habitats. Hence, fish density increases and competition for

correlation coeffi	cient in brackets	s) for sampling sit	tes. (*)Significanc	e level p < 0.05							
Environmental variables	a (y-intercept)	Conductivity	Oxygen concentration	Water temperature	Hq	Seasonal water flow	Dominant substrate	Submerged vegetation	QBR	BMWP'	Refuge index
Conductivity Oxygen	$-0.69^{*}$ 0.22	-0.29									
Water	0.12	0.32	-0.26								
temperature pH	-0.05	0.28	0.27	-0.43							
Seasonal	$(0.79)^{*}$	(-0.66)	(0.32)	(0.53)	(-0.21)						
water flow Dominant	*(U0 U)	*(22,0)	(0.45)	(0.15)	(10.07)	*(0) 76)*					
substrate				(21.0)	(12:0)	(0.10)					
Submerged vegetation	(0.30)	(-0.13)	(0.28)	$(-0.82)^{*}$	(0.62)	(-0.17)	(0.21)				
QBR	-0.09	-0.19	-0.15	$-0.78^{*}$	0.72	(-0.59)	(-0.02)	$0.88^{*}$			
BMWP'	$(0.72)^{*}$	$(-0.68)^{*}$	(0.31)	(0.21)	(0.42)	$(0.71)^{*}$	$(0.91)^{*}$	(0.08)	(0.00)		
Refuge index	$0.71^{*}$	-0.65	-0.21	-0.06	-0.13	(0.32)	(0.04)	(0.27)	0.18	0.18	
Fish density	-0.16	-0.03	-0.47	0.49	0.12	(-0.44)	(-0.45)	(-0.10)	0.66	-0.47	0.43

Table 4. Correlation matrix of parameter a (y-intercept) of the mass-length relationship and environmental variables (Pearson's correlation coefficient; Spearman's

Environmental variables	Partial correlation	Regression equations	Adjusted r <sup>2</sup>	F	df	p values
Model 1						
Seasonal water flow		a = -11.468 + 0.373 (seasonal water flow)	0.641	12.504	(1,7)	0.010
Model 2 (without seas	onal water flow	w)				
Dominant substrate		a = -11.671 + 0.273 (dominant substrate)	0.623	11.562	(1,7)	0.011
Refuge index	0.842	a = -12.241 + 0.223 (dominant substrate) + 0.202 (Refuge index)	0.890	24.303	(2,6)	0.001

*Table 5.* Stepwise multiple-regression models used to predict parameter a of the mass–length relationships from environmental variables in the streams studied.

space and food may become very important (Spranza & Stanley 2000). Seasonal and diel variability in isolated pools can lead to the death of some fish, particularly in small pools with high fish densities (Mundahl 1990). Associated with these changes in environmental conditions, basic changes may occur in life history attributes (Schlosser 1990), as has been demonstrated in the case of several cyprinids in similar streams in the south of the Iberian Peninsula (Herrera & Fernández-Delgado 1992, 1994, Fernández-Delgado & Herrera, 1995a,b, Rodríguez-Ruiz et al. 1998, Soriguer et al. 2000, among others). Moreover, in seasonal streams from the Iberian Peninsula it has been observed that the autumnal condition values are affected by the minima reached in summer (Herrera & Fernández-Delgado 1994, Fernández-Delgado & Herrera 1995b, Oliva-Paterna et al. 2002). In the present study, the autumn condition values in the intermittent streams, affected by their summer values, were clearly lower than in the Segura sampling sites (Table 1).

Because they were in the main channel of the basin and especially because of water regulation for agricultural purposes, the Segura sampling sites present a modified flow dynamic with a continuous water flow, large well-developed pool refugia and lower temporal variations. Torralva et al. (1997) reported that females of the same species from a regulated sampling site with a continuous water flow showed higher condition values than females from non-regulated sampling sites with highly variable water levels. Our results agree, and in the present study the fish condition was higher in the Segura sampling sites. Although oxygen concentration was not found to be a good predictor of fish condition in the present study, in agreement with Vila-Gispert et al. (2000) and Vila-Gispert & Moreno-Amich (2001), it is possible that this variable, which normally takes on higher values in constantly running waters, had an important effect.

It should be noted that the sampling area is one of the largest irrigated areas of the Iberian Peninsula, consequently the anthropogenic influences on stream flows are drastic. In this way, it is interesting to show how two sorts of anthropogenic influence (i.e., water diversion that results in isolated pools or increases environmental stress in habitats during summer versus stream flow regulation that results in continuous flow) for the same purpose (i.e., agricultural production) result in different effects on the fish condition. From a conservation biology standpoint, it is very interesting that B. sclateri showed higher condition values in our regulated sampling sites, although the lack of historical data prevents us from knowing whether stream flow regulation has really improved fish conditions in this portion of the species range.

Moreover, although Benamor stream showed a moderate seasonal water flow variation (Table 1), it produced high fish condition values. This sampling site had a high submerged vegetation value and a high index of quality based on aquatic macroinvertebrates (BMWP'), suggesting greater food availability and nutrient availability, two factors normally related to fish condition (Goede & Barton 1990). Consequently, this could be related with the high condition values found in this locality.

The interrelationships between intraspecific abundance (promoting an increase in intraspecific competition and social stress) and condition factor have been characterized in a number of studies (Sloman et al. 2001, among others). However, we found no relationship between fish density and condition, probably due to the number of sampling sites or perhaps because the relationship was clouded by the very complexity of the ecological interactions (e.g., a non-linear relationship between these variables). To a certain extent, this demonstrates the need for more investigation into the relationships between habitat characteristics, environmental variation and fish condition in the Mediterranean area.

In conclusion, the condition of Sclater's barbel in the semi-arid streams studied was affected by several ecological variables, which are related mainly to water flow and consequently to the physical structure of the streams, the latter variable being directly related to substrate and fish refuge. Streams with a continuous water flow produced fish with a better condition because they provide a great variety of habitats and substantially increase food availability, two factors which would reduce intraspecific competition for space and/or food. In the streams with an intermittent water flow (mainly in summer) fish are confined to small isolated pools where, particularly when the pools are small and contain high densities of fish, biological interactions could become important reducing fish condition.

It is known that artificial river regulation, among other factors, has a strong influence on fish recruitment (Jurajda 1995), fish growth (Torralva et al. 1997), fish production (Almodóvar & Nicola 1999), fish hybridization (Balon 1992) and on the whole fish community (Meador & Matthews 1991). In this study it was also observed that fish population condition is also affected. In particular, the modification of stream segments into isolated pools during summer could have a negative effect on fish with low condition values. These effects should be considered when such populations are subjected to sports fishing regulations, recovery programmes or any other management activity.

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