INVASION NOTE 10 pag. max. (9 ms + 1 table)

Inter-population variability in dietary traits of invasive bleak *Alburnus alburnus* (L., 1758) across the Iberian Peninsula

D. Latorre¹, G. Masó^{1,2}, A. Hinckley^{3,4}, D. Verdiell-Cubedo¹, G. Castillo-García⁵, A. Vila-Gispert¹, G. H. Copp^{6,7}, J. Cucherousset⁸, E. da Silva⁹, C. Fernández-Delgado¹⁰, E. García-Berthou¹, R. Miranda¹¹, F. J. Oliva-Paterna¹², A. Ruiz-Navarro¹², R. Merino-Aguirre³, J. M. Serrano³, D. Almeida^{5,*}

¹GRECO, Institute of Aquatic Ecology, University of Girona, E-17071 Girona, Spain; ²Department of Biodiversity and Ecological Restoration, Instituto Pirenaico de Ecología (IPE-CSIC), E-22700 Jaca, Spain; ³Department of Ecology, Complutense University of Madrid, E-28040 Madrid, Spain; ⁴Conservation and Evolutionary Genetics Group, Estación Biológica de Doñana-CSIC, E-41092 Sevilla, Spain; ⁵Departamento de Ciencias Médicas Básicas, Facultad de Medicina, Universidad San Pablo-CEU, CEU Universities, Campus de Montepríncipe, E-28925 Alcorcón, Madrid, Spain; ⁶Salmon & Freshwater Team, Cefas, Lowestoft, Suffolk NR33 OHT, U.K.; ⁷Department of Life & Environmental Sciences, Bournemouth University, Poole, BH12 5BB, U.K.; ⁸CNRS, Université Toulouse III Paul Sabatier, ENFA, UMR 5174 EDB (Laboratoire Évolution & Diversité Biologique), Toulouse F-31062, France; ⁹Department of Anatomy, Cell Biology & Zoology, University of Extremadura, E-06006 Badajoz, Spain; ¹⁰Department of Zoology, University of Córdoba, E-14071 Córdoba, Spain; ¹¹Department of Environmental Biology, University of Navarra, E-31080 Pamplona, Spain; ¹²Department of Zoology and Physical Anthropology, University of Murcia, E-30100 Murcia, Spain.

*Correspondence to: David Almeida, E-mail: david.almeidareal@ceu.es

Abstract To be added in the end (150-250 words).

Keywords gut content analysis, Mediterranean rivers, non-native fish, prey richness, trophic niche breadth

Running title Diet of invasive bleak across Iberia

Introduction

Biological invasions have resulted in serious impacts on native communities at the global scale (Mooney and Hobbs, 2000). Non-native species are regarded as a major driver of biodiversity loss, especially in freshwater ecosystems, where fish are the most common invaders (e.g. Copp et al. 2005). The disruptive effect of fish invasions is of particular conservation concern in the Iberian Peninsula, where endemism is high in freshwater ecosystems. Indeed, >50% of native fish species are unique to this region (Reyjol et al. 2007; Almeida et al., 2013), but the proportion of the ichthyofauna that non-native fishes represent (>30%) continues to increase (Leunda 2010; Almeida et al., 2013).

The bleak *Alburnus alburnus* (L., 1758) is a cyprinid species native to most of Europe, from the Pyrenees to the Urals. In the native area, bleak inhabit lakes or still-waters in medium/large rivers and feed chiefly on zooplankton (Vinni et al. 2000; Vašek and Kubečka 2004; Keith et al. 2011). In the Iberian Peninsula, this species was mainly introduced in reservoirs during the 1990s as a ‘forage fish’ for non-native piscivorous fishes, such as northern pike *Esox lucius* (L., 1758), largemouth bass *Micropterus salmoides* (Lacépède, 1802) or pikeperch *Sander lucioperca* (L., 1758) (Vinyoles et al. 2007). Since its introduction, the bleak has displayed a strong invasive character throughout Spain and Portugal, taking advantage of anthropogenic disturbances on Mediterranean freshwater ecosystems (Masó et al. 2016; Matono et al. 2018). Moreover, this species threatens several Iberian fishes, mainly through trophic and interference (i.e. aggression) competition (Leunda 2010). This is because Iberian fish communities are least diverse and are poorly adapted to a such invasion level of non-native fishes (Reyjol et al. 2007), which usually displace natives from available resources. As an example, direct observations by snorkelling have clearly shown that foraging behaviour of endemic Ebro nase *Parachondrostoma miegii* (Steindachner, 1866) is disturbed by bleak (Almeida and Grossman 2012). However and despite the high ecological risk posed by this species (Almeida et al. 2013), the available information on bleak dietary traits in the Iberian Peninsula is only provided from a few particular sites (Latorre et al. 2016; Almeida et al. 2017), with no study comparing data at the scale of this entire eco-region. Indeed, dietary traits of non-native fish can show wide variability under contrasting ecological conditions (Copp et al. 2005; Almeida et al. 2012, 2017) and this has implications from a conservation perspective. Thus, a better understanding of dietary data can be used by policy-makers and environmental managers to assist monitoring programmes in identifying which other areas are likely to be colonised by this invasive species (Almeida et al., 2013). Therefore, information in the present paper is highly relevant to understand invasion features of this fish species throughout Iberian fresh waters.

Consequently, the aim of the present study was to assess the inter-population variability in dietary traits of invasive bleak across the Iberian Peninsula. For this purpose, bleak populations were compared among the main rivers within this region, along with a native bleak population from France also being assessed. Specifically, diet composition was analysed by means of two overall indices: percentages of occurrence and ingested mass; and four dietary parameters were examined: ingested mass, prey richness, trophic diversity and trophic niche breadth.

Materials and methods

Study areas

Five main Iberian rivers were sampled for bleak along a latitudinal gradient (from North to South): Ebro (41°47'30''N–1°05'24''W and 41°26'39''N–0°28'26''W), Tagus (39°49'07''N–4°20'25''W and 39°58'12''N–4°42'17''W), Guadiana (38°59'58''N–5°51'52''W and 38°50'18''N–6°13'53''W), Segura (38°06'05''N–1°17'50''W and 38°04'54''N–0°53'37''W) and Guadalquivir (37°37'01''N–5°35'45''W and 37°30'46''N–5°56'38''W). These geographic coordinates correspond to two sampling sites per river (first coordinates are for sites located upstream). For comparative purposes, we surveyed a main river within the bleak's native range close to the Iberian Peninsula, i.e. historically a potential 'donor region' for non-native fish introductions to this region along the so-called 'Perpignan-Barcelona corridor' (see Clavero and García-Berthou 2006 for details on invasion routes). Thus, the River Saône (eastern France) was selected as a 'reference' population, although only one site (47°02'13''N–5°06'52''E) was finally surveyed. In any case, a comparable number of bleak specimens (200 individuals, see below) were collected from the River Saône and data from the two sampling sites in Iberian rivers were pooled for statistical analyses (i.e. no 'site effect' was found, see Data Analyses below). All the study rivers were selected because they represent large water courses (i.e. >500 km river length), where bleak are widely distributed and reach relatively high abundances within the local fish assemblages. A final key point for selection of these particular Iberian rivers was that bleak were sequentially introduced into the different catchments and thus, their populations will potentially reflect particular 'invasion stages' on the examined dietary traits. Specifically, bleak were introduced into the study rivers in the following years (first mention) (Vinyoles et al. 2007): Ebro in 1992, Guadiana in 1999, Segura in 2004, Tagus in 2005 and Guadalquivir in 2006. The climate in the study area of the River Saône is Temperate Oceanic (800–1000 mm of mean annual rainfall, 10–13 °C mean annual temperature) (Ministry of Environment France 2019), the most common climatic conditions for the bleak's native range in Europe (Keith et al. 2011). The climate regime for the study Iberian rivers is typical Mediterranean, with rainfall concentrated in autumn–winter (≈500 mm) and intense summer drought (<100 mm). The mean annual temperature ranges between 15–18 °C. The lowest temperatures occur in winter (down to –5 °C) and the highest in summer (>35 °C) (Ministry of Environment Spain 2019).

Field sampling and laboratory procedures

Fish were collected from May to June 2012, just before the spawning period of bleak for each study area (authors, pers. observ.), thereby avoiding any effect of the 'reproductive status' on foraging habits. Moreover, the year 2012 is considered to have been hydrologically 'average' in the study areas (Ministry of Environment France 2019; Ministry of Environment Spain 2019). As a result, the effects of particular dry or wet years on dietary traits are avoided within our study, which allows the data to be considered representative for this species in the invaded Mediterranean region of Europe. Sampling sites ($n = 11$, 2 per Iberian river plus 1 in the River Saône) were selected to encompass similar environmental conditions. Specifically, sites were located in well-regulated middle reaches of the main channel, where water level fluctuations are controlled throughout the year, avoiding the effects of strong increases/decreases in river discharge. These habitat conditions are representative of large rivers in both the native and Iberian ranges.

Also, sampling sites were located in the vicinity of similar surroundings (e.g. land use for agricultural exploitation) and far from influences of main tributaries and towns. Finally, sites were separated >50 km within each Iberian river to ensure the data were more representative from the study areas and also to minimise data dependence among sampling sites per river. A variety of consistent sampling protocols were followed by wading and from boats according to the European legislation (CEN/ISO Standards, EC Directive 2014/101/UE, European Commission 2014). This allowed obtaining a representative sample of bleak across the broadest possible body size range from each river. Catch methods consisted of following a zigzagging and upstream direction in both banks at each site (100 m river length) by electrofishing (2000 W pulse DC generator at 200–250 V, 2–3 A, 30 min per bank), dip nets (1.5 m long pole, 30 cm diameter net, 10 mm mesh size), seine nets (20 × 2 m, 10 mm mesh size) and gill nets (20 × 1 m, 10 mm mesh size, 50% hanging ratio, 1.5 m deep). All surveys followed the same sampling protocols (e.g. proportional effort in terms of people and time) to ensure comparability among the study rivers. Additionally, a professional fisherman sampled for bleak in France. To encompass the existing environmental variability, fish were collected from all meso-habitats present in the study rivers (e.g. runs, pools, shallows).

After each survey was concluded, bleak were immediately euthanized by immersion in an overdose solution of anaesthetic (MS-222) for 15 min, followed by severance of the spinal cord. Fish were stored on ice during transport to the laboratory (<2 h within the same sampling date). Individuals of the remaining fish species were identified and counted. Native species were kept in a tank with supplied oxygen (two battery operated aerators with portable pumps) until fully recovered before being released. Non-native species were euthanized (see details above). All field procedures complied with animal use and care regulations of Europe (specific Licenses were granted for Spanish and French Scientific Field Research). Fishes were collected by trained personnel (i.e. the holders of the Licenses, six researchers in the author list). Thus, no adverse effects were caused on the wildlife/habitat in the sampling sites and all native fish were fully recovered.

On arrival at the laboratory, bleak individuals ($n = 1200$, 100 ind. × 2 sites × 5 Iberian rivers + 200 ind. × 1 site from the River Saône) were measured for standard length (SL, ±1 mm). This particular fish length was selected because it avoids ‘noise’ given by variation of caudal fin length not related to body size (e.g. wounds and cuts in the fish skin and rays). Specifically, bleak size ranged between 40–195 mm SL. Fish were dissected to examine the sex and annotate the eviscerated weight (eW, ±0.1 g), which avoids the effect of gonad and gut masses. Only the anterior one-third of the intestinal tract was preserved in 4% formalin for subsequent examination to avoid food remains severely digested (see a similar procedure in Latorre et al. 2016). Food items were identified to the lowest possible taxonomic level (e.g. Tachet et al. 2003) using a dissecting microscope (40×) and weighed using an electronic balance (±0.1 mg).

Data analyses

Preliminary analyses did not find any difference between sexes. Consequently, this categorical factor was not included in subsequent data analyses to simplify the analytical models and thus, increase the statistical power of the remaining sources of variation, which would otherwise be seriously compromised (see a

similar procedure in Alcaraz and García-Berthou 2007). Also, data were pooled per Iberian river because the effect of ‘sampling site’ within each water course was not significant, after performing Generalised Linear Mixed Models (GLMMs) with ‘site’ as the random factor (see a comprehensive review of this statistical technique in Johnson et al. 2015). This analytical approach (i.e. pooled data) considerably improved the clarity of results and also eased biological interpretation.

Two overall dietary indices were calculated (omitting empty guts) and expressed as a percentage for each food category (e.g. Latorre et al. 2016): ‘occurrence’ (frequency of fish guts in which a particular food category occurred relative to the total number of fish individuals) and ‘ingested mass’ (frequency of the mass of a particular food category relative to the total ingested mass in all the examined intestinal tracts). Both of these percentages indicate whether a given food item is commonly eaten within the population and whether this food category is energetically important to the population. Furthermore, four dietary parameters were also calculated for each fish (e.g. see Almeida et al. 2012 for details on the particular formulae): ingested mass (mg), prey richness (S), trophic diversity (Shannon index, H') and trophic niche breadth (standardised Levin index, B).

Analysis of covariance (ANCOVA) was used to reveal significant differences between the study populations for the four dietary parameters. The effect of body size was controlled by using SL as the covariate. ANCOVAs were followed by a *post hoc* Tukey-Kramer honestly significant difference (HSD) test. Data were transformed by using $\ln(x + 1)$. Assumptions of normality of distributions and homogeneity of variances were verified through Shapiro-Wilks and Levene’s tests, respectively. Statistical analyses were performed with R version 3.1.3 (R Core Team 2018). The significance level was set at $\alpha = 0.05$. Sequential Bonferroni corrections were performed for every set of multiple tests.

Results

Diptera larvae (e.g. taxonomic Families Chironomidae and Simuliidae), zooplankton (e.g. water fleas) and detritus were the most frequent food categories for the reference native population (i.e. the River Saône), both in terms of occurrence and ingested mass (Table 1). The Ebro population showed a similar diet composition as per native bleak, although Ephemeroptera and Plecoptera nymphs were also important prey items. For the rest of study rivers, percentages for other categories were highly variable (Table 1). More in detail to be highlighted per river, vegetation and other benthic invertebrates (e.g. freshwater snails) were frequent food categories in the River Tagus. Flying insects (e.g. wasps, mosquitoes or butterflies) and Trichoptera larvae were very common in the Guadiana and Segura populations. The highest ingested mass (19%) was for nektonic insects (e.g. water boatmen or predatory beetles) in the River Guadalquivir. Detritus was an important food category for all bleak populations: 21–42% in occurrence and 13–27% as ingested mass (Table 1).

Regarding dietary parameters, significant differences were found between bleak populations for ingested mass ($F_{5,503} = 32.94$, $P < 0.001$). Four distinct groups were found (from low to high adjusted mean): Saône/Segura (≈ 12 mg), Guadalquivir (≈ 25 mg), Guadiana (≈ 40 mg) and Tagus (almost 60 mg) (Fig. 1a). Differences were also found for prey richness ($F_{5,503} = 18.68$, $P < 0.001$). Two river groups were found: Ebro/Saône (≈ 3 –4 prey items) and Guadiana/Guadalquivir/Tagus (≈ 5 prey) (Fig. 1b). Significant differences were found between populations for trophic diversity ($F_{5,503} = 19.76$, $P < 0.001$). Two river

groups were found: Saône/Ebro/Guadalquivir ($H' \approx 1$) and Guadiana/Tagus ($H' > 1.5$) (Fig. 1c). Differences were found between rivers for trophic niche breadth ($F_{5,503} = 20.49$, $P < 0.001$). Two distinct groups were found: Saône ($B \approx 1.4$) and Tagus/Guadiana ($B > 2$). The remaining three rivers showed intermediate B values (1.6–1.7) (Fig. 1d).

Discussion

Given that bleak mainly inhabits still-waters in its native area, this species is well-adapted to play an ecological role as an open-water feeder, with diet being chiefly based on zooplankton (Vinni et al. 2000; Vašek and Kubečka 2004). Thus, planktonic Crustacea were very important for bleak in the River Saône, although benthic Diptera larvae and detritus were more consumed. This difference between scientific literature and the present results was probably because of most surveys for bleak have been carried out in lakes and reservoirs (e.g. Vinni et al. 2000; Vašek and Kubečka 2004), whereas bleak diet has received less attention in flowing-waters. But even in lentic environments, bleak can modulate this zooplanktivorous strategy at each particular habitat patch to increase resource partitioning and consequently, reduce competition with co-existing fish species, such as roach *Rutilus rutilus* (L., 1758) or freshwater bream *Abramis brama* (L., 1758) (Vinni et al. 2000; Vašek and Kubečka 2004). Dietary indices were more similar between the River Saône and the River Ebro, the closest (geographically) study area to the reference population. This was also observed by Latorre et al. (2018) for other population responses (i.e. growth and reproduction), which was explained in terms of climatic ‘proximity’. In the main Iberian rivers, bleak showed a high consumption of detritus, which was also found elsewhere within this eco-region, from streams to reservoirs (Latorre et al. 2016; Almeida et al. 2017). More specifically, other food items were prominent among the study populations, including a variety of contrasting ecological features for each trophic resource: vegetation, neuston, nekton and benthos. Thus, consumption of algae and plant material was higher in the River Tagus. In this respect, Vinni et al. (2000) demonstrated that intra- and mainly inter-specific competition provided a potential explanation to understand this result (i.e. high use of vegetation) in the native area. However, Latorre et al. (2018) did not find any effect of this ‘low nutritious’ feeding type (detritus and vegetation) or competition on the growth rate in the River Tagus, with this population showing the highest values. In the Guadiana and Segura populations, bleak took advantage of prey from the water surface, such as fallen arthropods, which results in a low effort in terms of energy investment. Bleak also caught prey from the water column in the River Guadalquivir, but this population displayed a ‘costlier’ foraging alternative, as nektonic insects (e.g. taxonomic Family Dytiscidae) require higher pursuit and capture efforts. But more importantly, bleak fed on a great variety of benthic prey, from nymphs and larvae to molluscs and annelids, in all river populations. This finding indicates an elevated capacity of this fish to use food resources apparently less suitable to its morphological adaptations (e.g. conspicuous superior mouth). In overall, these results suggest that bleak could deeply change the foraging strategy, from a ‘pelagic’ to a ‘benthic’ feeder, which may aid bleak to better thrive at each Iberian river, where zooplankton availability may be a more limited trophic resource (Almeida et al. 2017). Such a significant dietary shift has been also observed in the Iberian Peninsula for other invasive fishes adapted to a more stable hydrological regime in their native

ranges, although they are phylogenetically and geographically very ‘distant’ (e.g. taxonomic Family Centrarchidae from North America, see Almeida et al. 2012).

Regarding dietary parameters, variation limits for the four examined predictors were similar as per other study areas in the Iberian Peninsula (Latorre et al. 2016 in the NE; Almeida et al. 2017 in the SW). The present data were obtained from large rivers and the just mentioned studies included streams and reservoirs. Thus, these overall results indicate that all Iberian bleak populations possess the same capacity of adapting to the particular habitat conditions where this invasive fish inhabits. No clear pattern was found for any dietary parameter in relation to the year of introduction, which was in accordance with Latorre et al. (2018) for growth and reproduction traits. This lack of relationship is probably because the establishment stage has been reached in a few years after bleak introductions (e.g. Bøhn et al. 2004). However, foraging strategy appeared to follow a unimodal response in relation to the latitudinal gradient (Oksanen and Minchin 2002), with Guadiana and mainly Tagus showing the maximum values. A potential explanation is that bleak may be more favoured by moderate Mediterranean climate in central Iberia than Temperate conditions in the native area, whereas bleak are affected by slightly more ‘severe’ ecological conditions in southern Iberia: stronger summer drought with the consequent decrease in discharge or lower oxygen level because of high temperature (Gasith and Resh 1999). Specifically for ingested mass, Tagus River showed the highest value of this dietary parameter, resulting in only one ‘statistical group’. This could compensate a higher proportion of vegetation and detritus in the diet (see above). Indeed, Tagus population showed the highest back-calculated lengths at all ages and growth index among the same study rivers (Latorre et al. 2018). Additionally, more ‘benign’ conditions in central Iberia may improve the physiological status of bleak, which facilitates this fish species to use alternative prey and widen its diet, with the corresponding reflection in prey richness and chiefly, trophic diversity and breadth. In contrast to this ‘generalist’ strategy, more northerly and southerly populations (i.e. distant from the optimum, see Oksanen and Minchin 2002) displayed a ‘selective’ or ‘specialist’ strategy, reducing the complexity of diet composition.

Ecological responses, such as dietary traits, usually show wide variability in non-native species when invading new habitats, with this being particularly clear in freshwater fishes (Copp et al. 2005; Almeida et al. 2012; Latorre et al. 2016). In the Iberian Peninsula, invasive fishes usually display wide trophic variability under contrasting environmental conditions, which contributes to their invasion process (Leunda 2010; Latorre et al. 2016; Almeida et al. 2017). Accordingly, dietary traits were clearly variable among the study rivers, indicating wide plasticity in bleak foraging strategies dependent on particular river conditions (i.e. habitat heterogeneity, food supply). Similarly as per other biological attributes (e.g. growth and reproduction), this high inter-population variability is considered as a mechanism for bleak to successfully invade novel Mediterranean freshwater ecosystems, from streams to large rivers (Masó et al. 2016; Latorre et al. 2018). Indeed, Iberian endemic species are specialized to narrow trophic niches (Reyjol et al. 2007) and this low level of co-evolutionary competition is the reason why Iberian fish fauna is not adapted to strong invasive competitors, such as bleak. This is one of the main reasons to consider this fish species as a bio-invader of high risk in the Iberian Peninsula (Almeida et al. 2013). Consequently, environmental managers should apply urgent conservation measures to control bleak populations across Iberian fresh waters. Firstly, these measures should focus on the target species (i.e. fish

culling), but also include actions on aquatic habitats to restore the ‘Mediterranean conditions’ that benefit native fish communities and hamper invasive species (e.g. recovery of natural flow regimes), specifically the bleak (Matono et al. 2018).

Acknowledgements A. Hinckley held a ‘collaboration’ grant from the Complutense University of Madrid. This study was also partly supported by the Spanish Ministry of Education (FPU 13/03291), the Spanish Ministry of Economy and Competitiveness (projects CGL2015-69311-REDT and CGL2016-80820-R) and the Government of Catalonia (ref. 2014 SGR 484). Participation by G. H. Copp was supported by the Centre for Environment, Food & Rural Affairs.

References you can change some references, but 30 max.

- Alcaraz, C., and García-Berthou, E. (2007). Food of an endangered cyprinodont (*Aphanius iberus*): ontogenetic diet shift and prey electivity. *Environmental Biology of Fishes* 78, 193–207.
- Almeida D, Grossman GD (2012) Utility of direct observational methods for assessing competitive interactions between non-native and native freshwater fishes. *Fisheries Manag. Ecol.* **19**: 157–166
- Almeida D, Almodóvar A, Nicola GG, Elvira B, Grossman G D (2012) Trophic plasticity of invasive juvenile largemouth bass *Micropterus salmoides* in Iberian streams. *Fish Res* **113**: 153–158
- Almeida D, Ribeiro F, Leunda P M, Vilizzi L, Copp G H (2013) Effectiveness of FISK, an Invasiveness Screening Tool for Non-Native Freshwater Fishes, to Perform Risk Identification Assessments in the Iberian Peninsula. *Risk Anal.* **33**: 1404–1413
- Almeida, D., Fletcher, D. H., Rangel, C., García-Berthou, E., da Silva, E. (2017). Dietary traits of invasive bleak *Alburnus alburnus* (Actinopterygii, Cyprinidae) between contrasting habitats in Iberian fresh waters. *Hydrobiologia*, 795: 23-33.
- Bøhn, T., Sandlund, O. T., Amundsen, P. A., and Primicerio, R. (2004). Rapidly changing life history during invasion. *Oikos* 106, 138–150.
- Clavero, M., and García-Berthou, E. (2006). Homogenization dynamics and introduction routes of invasive freshwater fish in the Iberian Peninsula. *Ecological Applications* 16, 2313–2324.
- Copp GH et al. 2005. To be, or not to be, a non-native freshwater fish? *Journal of Applied Ichthyology* 21:242–262.
- European Commission (2014). Commission Directive 2014/101/EU of 30 October 2014 amending Directive 2000/60/EC of the European Parliament and of the Council establishing a framework for Community action in the field of water policy. *Official Journal of the European Union – Legislation* 311, 32–35.
- Gasith, A. & V. H. Resh, 1999. Streams in Mediterranean climate regions: abiotic influences and biotic responses to predictable seasonal events. *Annual Review of Ecology and Systematics* 30: 51–81.
- Johnson, P. C. D., Barry, S. J. E., Ferguson, H. M., and Müller, P. (2015). Power analysis for generalized linear mixed models in ecology and evolution. *Methods in Ecology and Evolution* 6, 133–142.
- Keith P, Persat H, Feunteun E, Allardi J (2011) Les poissons d'eau douce de France. Muséum National d'Histoire Naturelle, Paris

- Latorre, D., Masó, G., Hinckley, A., Rubio-Gracia, F., Vila-Gispert, A., Almeida, D. (2016). Inter-population plasticity in dietary traits of invasive bleak *Alburnus alburnus* (Linnaeus, 1758) in Iberian fresh waters. *Journal of Applied Ichthyology*, 32: 1252-1255.
- Latorre D et al. 2018. Inter-population variability in growth and reproduction of invasive bleak *Alburnus alburnus* (Linnaeus, 1758) across the Iberian Peninsula. *Marine and Freshwater Research* 69:1326–1332.
- Leunda PM (2010) Impacts of non-native fishes on Iberian freshwater ichthyofauna: current knowledge and gaps. *Aquatic Invasions* 5: 239–262
- Masó G, Latorre D, Almeida D, Tarkan A S, Vila-Gispert A (2016) Inter-population plasticity in growth and reproduction of invasive bleak, *Alburnus alburnus* (Cyprinidae, Actinopterygii), in northeastern Iberian Peninsula *Folia Zool* 65: 10–14
- Matono, P., da Silva, J., Ilhéu, M. (2018). How does an invasive cyprinid benefit from the hydrological disturbance of Mediterranean temporary streams? *Diversity*, 10: 47.
- Ministry of Environment France (2019) <http://www.meteofrance.fr/climat-passe-etfutur>. Accessed 19 January 2019
- Ministry of Environment Spain (2019) <http://www.aemet.es/es/serviciosclimaticos>. Accessed 19 January 2019
- Mooney H A, Hobs R J (2000) *Invasive Species in a Changing World*. Washington, DC: Island Press, 456 pp
- Oksanen J, Minchin PR (2002) Continuum theory revisited: what shape are species responses along ecological gradients? *Ecological Modelling* 157: 119–129.
- R Core Team. R: A Language and Environment for Statistical Computing; R Foundation for Statistical Computing: Vienna, Austria, 2017. Available online: <http://www.r-project.org> (accessed on 1 October 2018).
- Reyjol Y, Hugueny B, Pont D et al (2007) Patterns in species richness and endemism of European freshwater fish. *Global Ecol Biogeogr* 16: 65–75
- Tachet, H., P. Richoux, M. Bournaud & P. Usseglio-Polatera, 2003. *Invertébrés d'eau Douce. Systématique, Biologie, Écologie*. CNRS Éditions, Paris.
- Vašek M, Kubečka J (2004) In situ diel patterns of zooplankton consumption by subadult/adult roach *Rutilus rutilus*, bream *Abramis brama*, and bleak *Alburnus alburnus*. *Folia Zool* 53: 203–214
- Vinni M, Horppila J, Olin M, Ruuhijärvi J, Nyberg K (2000) The food, growth and abundance of five co-existing cyprinids in lake basins of different morphometry and water quality *Aquat Ecol* 34: 421–431
- Vinyoles D, Robalo JI, de Sostoa A et al (2007) Spread of the alien bleak *Alburnus alburnus* (Linnaeus, 1758) (Actinopterygii cyprinidae) in the Iberian Peninsula: the role of reservoirs. *Graellsia* 63:101–110

Figure caption

Fig. 1 Comparison of four dietary parameters for bleak *Alburnus alburnus* among the study rivers. Results are adjusted means $\pm$ SE (ANCOVA, covariate: SL). Letters above columns indicate significant differences in bleak populations (Tukey HSD tests, $P < \text{critical } P\text{-value from Bonferroni correction}$)