





## Uninvited dinner guests: the effect of invasive fish and temperature on the foraging efficiency of Southern Iberian Chub

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Biological invasions have multiple ecological effects on native biota, mainly through trophic interactions. Competition for food between invasive and species may be particularly important, with cascading effects potentially disrupting ecosystem function and food webs (Baxter *et al.*, 2004). The outcome of trophic interactions is mediated by abiotic factors, such as temperature, and may thus be significantly affected under future climates (Ficke *et al.*, 2007).

The Iberian Peninsula holds a rich and highly endemic freshwater fish fauna, which is highly threatened by invasive species, habitat degradation, and climate change (Clavero, 2011). Climate change is expected to strongly mediate biological invasions, with higher temperature and more severe droughts leading to the expansion in range and population size of warm-water invaders (Rahel & Olden, 2008). Therefore, clarifying how invasive and native fish interact under different temperature regimes may be valuable for the conservation of Iberian freshwater biodiversity in future climates.

### Extended abstract SIBIC2016 2016.011: 3p

In this study, we experimentally assessed the effects of temperature on the foraging success of the endangered native Southern Iberian chub *Squalius pyrenaicus* (Günther, 1868) in relation to two ecologically damaging invaders, the pumpkinseed sunfish *Lepomis gibbosus* (Linnaeus, 1758), native to the North American Great Lakes, and the chameleon cichlid *Australoheros facetus* (Jenyns, 1842), native to warm waters of South America.

We collected fish measuring 45 to 75 mm total length in the Sado catchment, between October 2015 and May 2016, using electrofishing. For acclimation to the laboratory, individuals of each species were held for two weeks in separate aquaria (25 to 90 L), with gravel and air filters, maintained at room temperature (15-21°C) and 12:12h photoperiod. Afterwards, fish were acclimated to experimental temperatures in smaller aquaria (10 to 25 L), at a rate of 1 °C per day, and kept at experimental temperatures for one week. Throughout this period, fish were fed red chironomid larva *ad libitum* once daily, but prior to experiments individuals were held for a 48h without food.

Our design included feeding trials for single and pairs of individuals. These included single trials for each species, intraspecific trials with two chubs, and interspecific trials with one chub and either one pumpkinseed or one cichlid, and trials with the two invasives. Each combination was tested at three temperatures (19, 24 and 29°C), thus totalizing 21 treatment combinations, replicated at least five times. Feeding trails conducted so far and presented here involved single chub and cichlid and pairs with both species. This resulted in 56 feeding trials, involving 73 individuals, each used only once.

Feeding trials were conducted in 40 L aquaria, divided in two equal compartments by a removable plastic mesh barrier, and surrounded with black plastic to the back and sides. Fish were placed in different sides of the barrier, and allowed 15 minutes to recover from handling. During each trial fish were videotaped, with observations beginning immediately after the plastic barrier was gently raised to the surface. A trial consisted of 10 releases of individual prey, at approximately two-minute intervals, through one of five randomly selected tubes placed at the corners and back of the aquaria. After the experiments, invasive fish were sacrificed using MS222, and chubs were moved to holding aquaria, and slowly acclimated to room temperature, before being returned to the field.

Foraging success was derived from variation in prey capture rate and prey capture speed for each individual. Capture rate was determined from the number of preys captured per trial, and capture speed from the time to capture each prey. A capture represented a fish grasping a prey independently of whether or not the prey was consumed. Variation among temperatures in capture rate and speed was tested using Kruskal-Wallis tests, and post-hoc Dunn tests.

In single trials (Fig. 1), chub showed no significant variation in capture rate and speed with temperature. The rate of prey capture also remained constant for single cichlids, but the time to capture was significantly reduced at the higher temperatures (H: 52.2; p<0.001). Overall, the cichlid generally captured all the prey released.

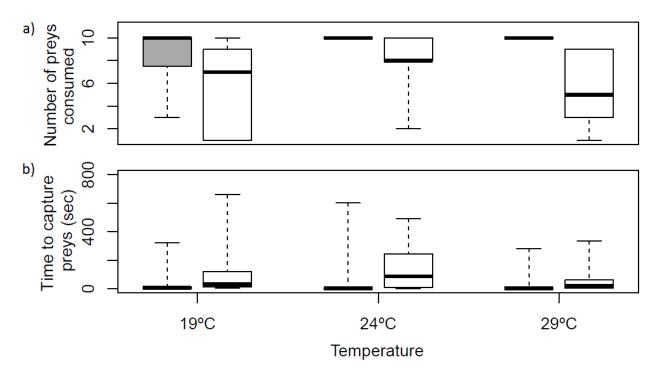
In interspecific trials, the capture rate and speed for chub were not significantly altered by temperature, with the chub always capturing only a few of the preys released (0-3). Conversely, capture rate for cichlid was always very high (7-10), but the capture time was significantly reduced at  $29^{\circ}$ C (H: 29.9; p<0.001).

The preliminary results of this study indicate that the foraging success of chub is not affected by temperature, though it is generally lower than that of the cichlid. This tends to be accentuated as temperature increased and the cichlid tends to capture preys faster.

Taken together our results suggest that temperature may play an important role in mediating trophic relationships between chub and the cichlid, with the latter tending to perform better at increased temperatures (Baduy *et al.*, 2016). In these circumstances, determining the extent to which warming waters in mediated interactions between other native and invasive

### Extended abstract SIBIC2016 2016.011: 3p

species is of critical importance in developing conservation efforts for Iberian freshwater fishes.



**FIGURE 1**. Variation in capture rate (a) and capture time (b) of prey for single individuals of *A. facetus* (grey), and *S. pyrenaicus* (white) in relation to temperature. Medians (bold line), quartiles ( $Q_1$ ;  $Q_3$ ) (box edges) and minimum and maximum values (whiskers) are shown.

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