Habitat Enhancement Solutions for Iberian Cyprinids Affected by Hydropeaking: Insights from Flume Research

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Abstract: Due to peak electricity demand, hydropeaking introduces rapid and artificial flow fluctuations in the receiving river, which alters the river hydromorphology, while affecting the downstream ecological integrity. The impacts of hydropeaking have been addressed in flumes and in rivers. However, few studies propose mitigation solutions based on fish responses. The objective of this communication was to assemble the methods and outputs of flume research focused on Iberian cyprinids and to present recommendations to be used by freshwater scientists and hydropower producers. Emphasis was given to the critical role of integrating ecology and hydraulics to find the causal pathway between a flow change and a measurable fish response. The use of diverse behaviour quantification methods, flow sensing technologies, and statistical tools were decisive to strengthen the validity of the findings and to identify fish-fluid relationships, according to flow events. This communication encourages further research to identify flow thresholds for key life-cycle stages and complementary river studies to design and assess mitigation solutions for hydropeaking. Although the research focused on an Iberian cyprinid, the methods suggested have the potential to be extended to other fish species affected by hydropeaking.

Keywords: hydropeaking; Iberian cyprinids; flow refuges; hydropower; flow variability; fish physiology; fish behaviour; ecohydraulics

1. Introduction

In response to daily peaks of electricity demand, the hydropower plants (HPP) control the flow through the turbines very rapidly, which results in downstream hydropeaking [1]. These rapid flow fluctuations result from the distinct stages of hydropeaking: base-flow discharge (no electricity production), increasing discharge or up-ramping (powering-on of the turbines), continuous high discharge (peak electricity demand), and decreasing discharge or down-ramping (powering-off of the turbines) [2,3]. This continuous flow variability alters the downstream river morphological and hydrological processes [4–9], with consequences to the ecological integrity of the river system [10]. Notwithstanding, new hydropower plants are planned or under construction [11,12]. In addition, the estimates of global hydropower potential are convenient for producers [13], therefore, hydropower production continues to expand [14]. Given these and the predicted impacts of climate change, it is expected that the artificial flow fluctuations will be particularly severe for Iberian rivers affected by a Mediterranean climate [15], especially in the summer when water is scarce and the flow downstream is low, even if an ecological flow is provided by the HPP.

Fish responses to hydropeaking range from organism-level adjustments (neuroendocrine and metabolic) [16,17], to changes in diel activities (e.g., foraging, finding a suitable habitat, or avoiding...
predators), and alterations in key life-cycle events (e.g., reproductive migration, recruitment, and survival) [18]. This range of responses is mostly related to the spatiotemporal scale of the effects. The study of population or community dynamics is to larger spatiotemporal scale effects, whereas organism-level responses are to finer spatial scales and short-term effects. Different experimental approaches have been adopted to address those biological responses, namely studies conducted in riverine conditions [19], in artificial flumes [20–22], and numerical modelling studies [23,24]. In rivers, changes in fish assemblages along the longitudinal gradient [25,26], in diel activity [27], and in life-history traits (growth and longevity) [28] have been associated with hydropoaking. In addition, smaller [19,29–32] to wider spatial-scale movements [33–37] have been reported. This high variability has been mainly attributed to the difficulty in isolating external factors, to the presence and use of natural velocity refuges, or to inter-individual and intra-individual variability [19,30]. It is clearly challenging to find a causal pathway between flow variability and a measurable fish response [38], explained by the overlapping effects of other physical and biological factors that are difficult to isolate in rivers [19,31].

Inter-individual and intra-individual changes associated with flow fluctuations are mostly addressed in flume conditions because it is possible to control the flow events and to isolate potential confounding factors. Physiological responses, such as elevations in corticosteroid levels and changes in glucose, have been associated with simulated rapid flow fluctuations in experimental flumes [16]. A high swimming effort [20], no changes in social interactions or growth rates associated with down-ramping [39], and diverse preference patterns according to substrate and refuges [16,22,25,40,41] have been reported in experimental flumes. Although some of these findings clearly illustrate that rapid flow fluctuations affected fish performance, others are inconsistent and even contradictory. It is still challenging to extrapolate the findings regarding specific changes in a flow component (e.g., magnitude or frequency) to population or community-level responses because these flow changes occur at timescales much shorter than population responses [31]. This is justified by the difficulty to isolate each flow component in river conditions. Although the impacts of hydropoaking in population and community dynamics are recognized, it is still complex to attribute a higher effect to magnitude or frequency. This aspect is crucial to implement operational or morphological mitigation strategies. Either in rivers or in flumes, it is possible to characterize the hydraulic conditions by using diverse flow sensing technologies. The measurements are then incorporated in hydraulic numerical models to either characterize hydropoaking and propose habitat enhancement measures [23,42,43], or to propose adaptations in the operational schemes [44,45] to predict potential scenarios, and to develop conceptual frameworks to serve as grounds for hydropoaking studies [2,8,18].

Even recognizing the research effort that has been directed to address the hydropoaking problematic, few studies have proposed mitigation strategies. Specifically referring to habitat enhancement solutions, lateral refuges (deflectors) were effectively used for flow-refuging by Iberian barbels in simulated hydropoaking conditions [21]. T-shaped structures [46] and lateral refuges were suggested as potential flow-refuges for brown trout [22] and young grayling [47]. However, the proposed artificial refuges were not tested in natural conditions. Most of the projected structures were designed to offer flow refuge for salmonids based on changes in the swimming activity [22,46], or the interaction with conspecifics in flume conditions [48,49]. However, considering flow as the abiotic factor that determines fish movement, it is expected that it has considerably different effects on the movement behaviour of fish species, especially considering the morphological adaptations and the diverse swimming modes resulting from millions of years of evolution [50]. For this research, we considered the existing knowledge about habitat enhancement strategies to mitigate the effects of rapid flow fluctuations for salmonids and to bridge the knowledge gap for cyprinids.

Prior to the work that resulted in this communication, the effects of hydropoaking on cyprinids had been scarcely addressed [26,28]. Moreover, the application of habitat mitigation measures for cyprinids was inexistent. Remarkably, this group is the most abundant and dispersed in rivers worldwide [51]. Grounded on these knowledge gaps and on the previous findings for salmonid species, the effects of
hydropeaking events were assessed for an Iberian cyprinid and the potential of artificial flow-refuges were evaluated at an indoor flume. The main objective of this communication is to assemble the major findings of those previous experiments by presenting the adopted multidisciplinary approach and a set of practical guidelines for the design of flow-refuges to freshwater scientists and hydropower producers. Particular emphasis is given to the critical role of integrating ecology and hydraulics to find the causal pathway between a specific flow change and a measurable fish response. Lastly, the limitations of this research are enumerated, and future research opportunities are discussed.

2. Research Approach

2.1. Problem Identification

The first step to implement flow-refuges as a mitigation measure for hydropeaking is the clear identification of the changes in magnitude, duration, and frequency of a flow event likely to occur in rivers affected by hydropower production. Thus, the simulated flow events were based on the operational scheme of Portuguese hydropower plants operating in hydropeaking regime [45,52]. Recognizing the impracticability of dam removal, the potential constraints inherent to the hydropower facility, and the difficulty of convincing hydropower producers to adapt the operational scheme, the research focused on the design of potential habitat enhancement solutions, given as flow-refuges. The methods adopted incorporated a multidisciplinary approach conducted at an indoor flume where physiological and behavioural responses were measured. The resultant hydraulic conditions were characterized (Figure 1).

![Flow Chart: Steps to conduct flume research for hydropeaking assessment](image)

**Figure 1.** Circular flow chart displaying a potential sequence of steps to study the impacts of hydropeaking in flume conditions and to propose mitigation measures accordingly. HPP-Hydropower Plant.

With this approach, it was possible to relate physiological adjustments, changes in motion, and structure use, with the hydraulic conditions created by flow fluctuations and the presence of structures. Although it was adopted to assess the hydropeaking problematic for an Iberian cyprinid, this research approach has the potential to be adapted to other flume-based research and fish species.
2.2. Experimental Design

The experiments were conducted at an indoor flume with a rectangular cross-section, constructed on a steel frame with glass panels on both sides (Figure 2). The flume length was shortened to 6.5 m using two perforated metallic panels. The discharge and the water level were controlled by a sluice gate upstream and by a flap gate downstream and the maximum discharge was set to 60 L·s\(^{-1}\). This flume is equipped with a false bottom, which means it is highly versatile when testing a wide range of structures.

![Figure 2. Top (a) and lateral (b) view of the indoor experimental flume, respective dimensions (m). C1-C5—Behaviour observation areas.](image)

A diverse set of flow events was tested, where magnitude, peak duration, peak frequency, and event duration were changed (Figure 3). For all experiments, the base-flow conditions consisted of a continuous 7 L·s\(^{-1}\) flow event (Figure 3, blue line). The peak discharges tested were 20, 40, and 60 L·s\(^{-1}\) and were adapted during the progress of the experiments based on the findings of each setup (Figure 3).

The peak events were initially studied in the presence of lateral deflectors (Figure 3, Setups 1 and 2) [21,53]. For these structures, two configurations were tested-meandered, and single-sided-in two experimental setups termed setup 1 and 2, respectively (Figure 3). The reasoning behind the selection of lateral structures was: (i) their broad use in restoration actions [54], (ii) velocity preferences of the selected cyprinid species [55,56], (iii) velocity refuges proposed for other species [22], (iv) habitat modelling studies based on suitability curves [23], and (v) in studies aiming at designing effective fishways for this species [57].

Instream structures, namely v-shaped structures and solid triangular pyramids, were tested in the third experimental setup (Figure 3, Setup 3) [58]. The instream structures were designed according to: (i) the results from the previous experiments with lateral deflectors, (ii) fishway experiments with this species [59], and (iii) the potential effect of these structures on the hydrodynamic conditions (e.g., formation of vortices and shaded areas) and, consequently, on specific fish motion patterns.
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Figure 3. Flow events and flume configurations for each experimental setup. The tested structures were: for Setup 1, lateral meandered deflectors, for Setup 2, lateral single-sided deflectors, and, for Setup 3, v-shaped structures and solid triangular pyramids, respectively.

2.3. Hydraulic Characterization

To characterize the hydraulic conditions and the hydrodynamic environment, two measuring technologies were adopted: (i) velocity measurements using acoustic Doppler velocimetry (ADV), followed by a calibration to a theoretical 3D numerical model, and (ii) a novel technology using an artificial lateral line probe (LLP), which measures the pressure gradients over the probe body, which enables a characterization of fluid-body interactions [58]. For both, we selected a measurement grid that covered the areas that would be most affected by the structures and by the peak event. Acoustic Doppler Velocimetry (ADV) technology (Nortek-AS Vectrino 10 MHz, 100 Hz sampling rate) was used to characterize the velocity field in the flume when equipped with meandered deflectors for 7, 20, and 60 L.s⁻¹. The ADV output was used to calibrate a numerical model, extending its measurements to the single-sided deflector configuration, and to predict the flow field, according to the imposed changes [21]. One disadvantage of ADV technology is that it only performs point flow measurements, which neglects the fluid-body interactions that fish use to process flow. This and other conventional techniques often measure at a sampling rate that is below (1–50 Hz) the sensory range of lateral lines (10s–100s Hz) [60]. Artificial lateral lines have emerged to provide a closer representation of the hydrodynamic changes that are actually perceived by fish [61,62]. The artificial lateral line used for this research consisted of a 0.22 m length standard NACA 0025 airfoil shape, which is most similar to a cross section of a fish. This sensor measures pressure gradients over its body, mimicking the natural analogue, by using six differential pressure sensors, two in the front (snout) and two at each side of the probe body [58]. The pressure measurements were performed for two depths, using a 200 Hz sampling rate. The selected flow metrics derived from the LLP (i.e., mean pressure, mean front fluctuations, and mean pressure asymmetry) result from previous measurements conducted in an experimental vertical slot fishway where those and other pressure variables were compared with fish behaviour [63]. Those pressure metrics are quadratically related with flow velocity, turbulence, and cyclic flow patterns [63].

2.4. Assessment of Fish Responses

The selected cyprinid species was the Iberian barbel, *Luciobarbus bocagei* Steindachner, 1864 (hereafter, *L. bocagei*). This potamodromous cyprinid is endemic to the Iberian Peninsula, and it has specific habitat requirements and velocity preferences during its ontogeny [51]. Thus, it is possible to study the effects of hydropeaking for different life-cycle stages of this species. Furthermore, the
swimming mode of *L. bocagei* differs from the widely studied salmonids. Although often identified as slower swimmers, by using the posterior half of the body for propulsion, these fish can be more efficient in turning and accelerating abilities [50].

Two types of fish responses were quantified: organism-level and whole-animal performance. With this approach, it was possible to capture immediate homeostatic adjustments and behavioural patterns, according to the imposed flow changes. To find whether they presented a stressor for *L. bocagei*, the selected organism-level responses were blood glucose and lactate. The reasons to select glucose were: (i) it generally increases after the cortisol response through the cortisol-mediated gluconeogenesis [64,65], which means there is a time latency between cortisol and the glucose response, and (ii) it has been broadly used in flow variability studies with reported increments comparatively to pre-stress levels [19,66]. Following the scientific recommendation that more than one response should be quantified [16], the other physiological response selected was lactate. Lactate is a metabolite of muscle activity, and is expected to increase during continual swimming [67]. Glucose and lactate level adjustments have been widely used as secondary physiological indicators of stress to flow variability [38]. Hence, both physiological responses may represent reliable surrogates of a stress response to flow variability.

Whole animal performance was analysed by quantifying changes in motion related to the structures tested and the occurrence of the peak event. The behavioural metrics were divided into two categories: (1) structure use and (2) swimming activity in the flume. Successful structural use was considered when a single fish or a group of two to five fish were observed in the immediate downstream area of the structure. The swimming activity metrics were separated into two types: fish sprints, defined as a swimming activity lasting a few seconds and characterized by several tail beats, and fish drifts, defined as downstream fish displacements driven by passive advection of the body in the flow direction. The frequency of behaviour was defined as the number of occurrences, in absolute frequency, over the duration of the flow event.

The biological results were analysed and interpreted according to the characterization of the hydraulic and hydrodynamic conditions. Thus, it was possible to relate inter-individual and intra-individual responses with the hydraulic conditions created in the flume and to find the most advantageous hydraulic conditions given by the flow event and the flow-refuge for *L. bocagei*.

3. Discussion of the Major Findings

3.1. Is There an Effect of the Simulated Peak Events?

After initially testing two peak discharges, i.e., 20 and 60 L·s\(^{-1}\), under the highest discharge in the presence of meandered deflectors, (i) the onset of a glucose response occurred sooner, (ii) the use of deflectors as flow refuges was inhibited, and (iii) the sprinting and drifting activity was higher [21]. Additionally, event duration and peak frequency dictated a sustained glucose response in the 60 L·s\(^{-1}\) flow events, and the unpredictability of the two-steps event (i.e., 20 L·s\(^{-1}\) followed by 60 L·s\(^{-1}\)) resulted in elevated glucose levels [21]. From these findings, it would be tempting to suggest that there was a clear adverse effect of the hydropoeaking events. However, signs of exhaustion were undetected and corroborated by the absence of a lactate response. Fish were able to cope with water velocities > 60 cm·s\(^{-1}\) by hiding in the downstream area of the deflectors [21]. Still, the ability of *L. bocagei* to cope with the hydraulic conditions created under 60 L·s\(^{-1}\) was lower and the swimming activity decreased especially after peak repetition.

The unpredictability of the events when more than one of the flow components (e.g., magnitude and frequency) was altered resulted in the most visible responses. Longer peak flows, with higher magnitudes and repeated peaks, resulted in glucose increments, increased swimming effort, and difficulty to find the available refuges [21]. Similar findings occurred in the presence of the single-sided deflectors [53]. This cumulative effect is recognized in the literature [18,68]. On the other hand, when a
hydropeaking event was tested in the absence of these structures, the lowest physiological responses were obtained [53]. This was explained by the highly predictable and homogenous flow conditions.

Regarding the instream structures tested in the third experimental setup, the effects of the same flow event differed significantly between the tested configurations [58]. After testing the instream structures, it was clear that the combined effect of the flow event and structure created local hydrodynamic changes that dictated the range of responses shown by *L. bocagei*, and not solely by the peak event itself [58].

### 3.2. Which Flow Event Structure Is the Most Ecologically Conclusive?

The most ecologically relevant findings regarding the hydropeaking problematic for this species are not solely those that identify the 60 L·s⁻¹ flow events as the most adverse, but mostly the exceptions to this trend. In one hand, the presence of deflectors created a more heterogeneous flow environment, yet *L. bocagei* were still able to use them for flow-refuging. Nonetheless, the scarcer search for deflectors under 60 L·s⁻¹ in comparison with the 20 L·s⁻¹ flow events and the increased overall swimming activity suggest that the critical hydraulic conditions created in the vicinity of the deflectors reduced *L. bocagei* ability to find the low flow areas downstream of the deflectors [21]. On the other hand, the combination of a flow event with the presence of instream structures altered the distribution of velocity, turbulent fields, and pressure fluctuations, which generates distinct behavioural responses [58].

The movement behaviour of *L. bocagei* was not solely related to the unpredictable flow conditions resulting from the peak discharge (i.e., 60 L·s⁻¹). Specifically referring to the instream structures, under the single-step flow event, the individual and group use were higher for the solid triangular pyramids [58]. The lower velocities expected inside the v-shaped structures, which would favour *L. bocagei* for flow-refuging, were masked by the complex flow conditions created by these structures. Even if the swimming activity was more pronounced under 60 L·s⁻¹, the physiological responses were not always indicative of stress or exhaustion.

From all experiments, it was possible to demonstrate that the most conclusive responses occurred in the events where more than one flow component (given as magnitude, frequency, or duration) changed in the presence of the structures that created the most complex flow conditions. These were: (i) the single-step and two-steps events for 60 L·s⁻¹ for meandered deflectors [21], (ii) the two-steps and the repeated single-step events for one-sided deflectors [53], and (iii) the single-step event for v-shaped structures [58] (see Figure 3).

### 3.3. Which Hydraulic Conditions Are the Most Beneficial?

From the analysis of the three experimental setups, the most favourable hydraulic conditions were those where flow magnitude, peak frequency, and peak duration were lower. These events were the base-flow events in the presence of deflectors and v-shaped structures [53,58], and the single-step events where the lowest peak discharge, i.e., 20 L·s⁻¹, was tested [21]. For these events, there were no physiological adjustments, the swimming effort was low demonstrated by the lower frequency of individual sprints and the balanced frequency of individual and group behaviour), and fish could easily access the refuges.

The movement behaviour results indicated that *L. bocagei* were using the deflectors more frequently than the instream structures, and that, under moderate peaks, their use was enhanced [21,53,58]. However, the physiological responses were not so clear. Glucose increments occurred in the presence of both deflector configurations [21,53], whereas lactate increments occurred only in the presence of v-shaped structures [58]. With these findings, a question emerged: does the higher use of deflectors imply that these structures are the most adequate for *L. bocagei* under hydropeaking conditions? By combining ecological and hydraulic perspectives, it was possible to assert that the hydraulic conditions created by the presence of deflectors motivated their use. It was clear that *L. bocagei* were able to use the meandered and the single-sided deflectors as flow refuges more effectively than the two
triangular-shaped structures. Under these conditions, the swimming activity (frequency of sprints and drifts) was enhanced.

Analysing the numerical models for deflectors [21,53], and the pressure distribution maps of the Lateral Line Probe (LLP) for the instream structures [58], it was evident that, rather than just velocity, it was the combination of local scale hydrodynamic features, given by mean front pressure fluctuations and mean pressure asymmetry that determined the movement patterns. These explained the difficulty for *L. bocagei* to use the triangular structures as velocity refuges, but also their ability to use those changes in hydrodynamic features to find other areas for flow-refuging [69], move upstream (favouring rheotactic behaviour) [70,71], or to benefit from group behaviour [72].

3.4. Flow Sensing Technologies: Acoustic Doppler Velocimetry and Lateral Line Probe

From the Acoustic Doppler Velocimetry (ADV) measurements and the hydraulic models, it was possible to define a flow threshold that represented the resting state for *L. bocagei*. For lateral structures (i.e., deflectors), velocity magnitudes ranging from near 0 to 0.41 m·s$^{-1}$ allowed *L. bocagei* to use the available flume area. This range enhanced *L. bocagei* swimming performance, without any physiological response [21,58]. The hydraulic characterization given by the FLOW-3D® models was relevant to explain deflector use and swimming activity patterns. However, the information that the velocity ranges provided was not sufficient to explain the diversity of *L. bocagei* organism level responses.

The results from the Lateral Line Probe (LLP) provided unique findings on the role of the hydrodynamic conditions as primary triggers of movement patterns by *L. bocagei* [58]. After analysing the fish responses together with the derived pressure variables, it was possible to define pressure thresholds referring to the mean front pressure, mean pressure fluctuations, and mean pressure asymmetry that provided the most and the least favourable hydrodynamic conditions for *L. bocagei* [58]. Asymmetry was the pressure variable that was the most related with *L. bocagei* responses, with a favourable asymmetry window being observed for the base-flow event with v-shaped structures [58]. On the other hand, the highest-pressure asymmetry and the high mean front pressure and pressure fluctuations measured in the single-peak event for the same structures hindered the fish’s ability to cope with the hydrodynamic conditions and resulted in lactate adjustments [58]. These pressure metrics were identified for the first time as potential surrogates of local hydrodynamic stimuli, according to the observed fish responses. Rather than just velocity, it is the combination of local hydrodynamic changes (i.e., mean front pressure, mean fluctuations, and asymmetry) that lead fish to select the flow conditions that ensure the lowest energetic costs. These regions are not only represented by the low flow areas inside the refuge, but also represented by regions that favour the formation of vortices and turbulent areas that fish can take advantage of [58]. Considering the range of pressure readings by the lateral line system [72], the LLP represents a potential tool to assess the distributed sensing capacity of fish. Thus, in comparison with the ADV, the LLP provided a better representation of how specific hydrodynamic changes affect the movement behaviour of fish. That study was the first to use this technology to understand smaller-scale movement behavioural patterns of fish associated with flow variability. The results are promising considering the role that local-scale hydrodynamic changes have on the swimming performance of fish, and represent a step forward to understand the ecological significance of the pressure field around the fish and its movement patterns.

4. Recommendations for Hydropower Producers

The research effort to find mitigation solutions to hydropeaking consequences is expanding. However, it is urgent to bridge the gap between this scientific knowledge and hydropower producers. With this flume-based research, it was possible to propose potential design guidelines for flow-refuges and recommendations to adapt the operational schemes in a hydropower plant. Specifically referring to the physiological findings, with this research, there was no unequivocal evidence that supported the existence of a flow threshold representing the resting state for *L. bocagei*. However, the lowest physiological responses were observed when *L. bocagei* was subjected to 7 and 20 L·s$^{-1}$ in the presence
of lateral deflectors, and in the absence of instream structures. For lateral deflectors, this absence of a physiological response, corresponding to a velocity threshold of \(0-0.41 \text{ m s}^{-1}\), which is in accordance with velocity preferences for this species [73–75], represents a further step regarding the definition of the resting state with respect to flow variability. Nonetheless, contrary to what would be expected, the physiological responses were not consistently lower under base flow conditions (continuous 7 L s\(^{-1}\)), but fluctuated, according to the flow event and structural configuration. With the exception of the hydropoaking event with v-shaped structures, the levels of blood lactate did not differ significantly between events, and the movement patterns of \(L. bocagei\) were not indicative that fish were exhausted [21,53]. These results emphasize the importance of habitat heterogeneity as a decisive factor for the definition of the most adequate conditions for this species. The concomitant analysis of physiological and movement behavioural responses was essential to find the most suitable flow event-structure configuration for \(L. bocagei\). For any study aiming to assess the potential of mitigation measures to hydropoaking based on quantified fish responses, we recommend selecting more than one level of an ecological organization (e.g., individual and population), and then to quantify different levels of organism-level responses (e.g., metabolic changes and whole-animal performance metrics likely to be associated with rapid flow fluctuations). Considering the absence of a universal threshold for a physiological response that represents the resting state for flow variability, the validity of a physiological result is significant only if compared with a base-flow or control condition. Lastly, in riverine conditions, there is a myriad of environmental variables that are difficult to isolate. Therefore, it is critical to isolate confounding external factors that may mask the effect of the flow changes. This can be partially outweighed by conducting experiments in outdoor and indoor flumes where it is possible to control flow variability and isolate external variables, and to replicate the conditions.

The metrics to describe movement behaviour were selected for their likelihood to be expressed under the simulated peak events. Remarkably, the fish responses were not always indicative that the proposed lateral or instream structures would be effective for flow-refuging. In rivers affected by hydropoaking, it is substantially more challenging to relate specific motion patterns and organism-level responses with the hydrodynamic changes. To implement lateral refuges in natural conditions, the most decisive factors for their design are the hydraulic and hydrodynamic changes that result from hydropoaking, which, in turn, determine the choice of the type, number, and positioning of structures. First, it is necessary to characterize the rapid flow fluctuations and the changes in water depth, velocity, and wetted profile before the design and implementation of a lateral or instream refuge [6]. Afterward, the proposed structures can be tested through numerical modelling to understand whether the added habitat heterogeneity provides velocity refuging areas, or creates unstable hydraulic conditions for fish [43,76]. According to our findings, both classical velocity measurements and novel biomimetic technologies provided sound results regarding the characterization of the flow patterns. Feasibility, cost, and post-processing are aspects to consider regarding the choice of a flow sensing technology. Yet, to assess the potential of habitat enhancement measures, it is more important to consider the fluid changes and their impact on fish organism responses and the motion patterns of fish. Thus, ADV technology provides a solid representation of the flow field, whereas the LLP illustrates fluid-body interactions, which provides a better representation of the pressure field that fish would perceive.

Specifically referring to recommendations to implement the artificial flow-refuges in natural conditions, the mitigation structures to be proposed should assure velocity refuges during up-ramping and lateral connectivity with the main channel during the down-ramping stage. To implement lateral refuges (deflectors), their length, angle, and height are crucial design factors [77,78]. Thus, the opening angle should be at least in the same order of magnitude as the fish body length [79], favour group behaviour, guide the flow, prevent clogging [22], and create or maintain the flow dynamics of the pool-riffle (sediment deposition-transport) systems while avoiding bank erosion [78]. The opening angle, excessive wide angles, in relation to the river bank, are discouraged. To avoid fish stranding, especially during down-ramping, the area behind the deflectors should avoid potential stranding zones, or assure a minimum water depth of 0.5 m [22,76]. Although a heterogeneous habitat is recommended,
adding refuge areas is not always the most effective solution [43,79]. There must be a trade-off between existing river habitat conditions and the effectiveness of adding new structures, while remembering that, in natural conditions, each case is unique and generalizations have to be made with caution.

One final recommendation refers to an unexpected finding regarding the fish responses to the two-step flow event (i.e., 20 followed by 60 L·s⁻¹). The high glucose levels, the inability to find the deflectors particularly during the transition between discharges, and the increased swimming activity in the second step, demonstrated that a stepped flow event is not favourable for _L. bocagei_ [21]. The explanation for these results refers to the difficulty of _L. bocagei_ to cope with the unpredictable flow conditions. This finding may be convenient for hydropower producers, since the economic benefit of a two-step event, would likely be lower than, for example, a continuous high peak single-step event. We recommend that the duration of the base-flow events between two high peaks is extended to enable fish to recover from the effort required to cope with the continuous high peak. Nevertheless, prior to the implementation of any operational scheme, it is crucial to quantify the impacts in river conditions.

### 5. Identify Limitations to Find Research Opportunities

In flumes, it is difficult to simulate rapid flow fluctuations of the same order of magnitude and rate of change as in some regulated rivers due to pumping capacity or to limitations inherent with the flow control [68]. Although it was possible to simulate rapid flow fluctuations, the maximum discharge of our flume was limited to 60 L·s⁻¹. This construction limitation was partially overcome by reducing the flume cross-section with two parallel deflectors that were installed in the upstream flume area for the meandered deflector’s setup. With this solution, it was possible to get higher velocity ranges during the hydropeaking events [21]. The deflectors were clearly used for flow-refuging, but, at the same time, the more complex flow environment reduced the success for _L. bocagei_ to use these areas in comparison with the 20 L·s⁻¹ flow events [21]. Thus, the effect of the rapid flow fluctuations was still evident. To achieve the most comprehensive results, the construction limitations inherent in the facilities must be recognized and alternatives to surpass potential limitations have to be considered.

Experimental flume research has been suggested as a valuable tool to understand the effects of rapid flow fluctuations in downstream fish communities [18,68]. A wide diversity of studies conducted in laboratory conditions discovered the behavioural diversity that is found in nature [80]. Although experimental flume research is reliable to study behavioural patterns, extrapolations to natural conditions are discouraged and its transferability should be further substantiated. Nonetheless, the behaviour patterns observed (e.g., use of available refuges, longitudinal movements, take advantage of flow changes to minimize energetic costs) have been observed in studies conducted in rivers [17,19], in nature-like channels [20], and in artificial streams [66]. Ideally, complementary river studies should be conducted to assess the ecological consequences of those patterns, and, particularly, in the context of hydropeaking.

One of the biggest problems of ecological studies conducted in controlled conditions is the difficulty to replicate the experiments and their findings. These studies are usually constrained by the number of organisms that is possible to obtain from the natural conditions, which reduces the robustness of the findings. To outweigh this limitation, the recommendations are to use a diverse set of behavioural quantification tools (e.g., physiology and movement behaviour) complemented with a detailed characterization of the hydraulic conditions, and the use of statistical tools that are adequate for small-sized samples. With the analysis of distinct levels of fish responses, it is possible to identify trends and to establish relationships, according to flow variability.

Lastly, although the present recommendations are mostly directed for young adults of _L. bocagei_, they can be extended to other Iberian cyprinid species. Nonetheless, single-species studies are difficult to extrapolate to communities, or the river ecosystem. Although the single-species limitation was recognized, this work represents a step forward to understand and mitigate the impacts of hydropeaking for cyprinids.
6. Future Hydropeaking Research: Recommendations for Freshwater Scientists

The findings that motivated this communication present the initial steps to propose effective mitigation measures for hydropeaking consequences for cyprinids inhabiting rivers affected by the Mediterranean climate. Further research opportunities are described afterward.

Long duration and repeated flow peaks produced physiological adjustments as well as a reduction of the swimming activity during the second peak with the same magnitude. Thus, we hypothesized whether the swimming effort was higher in the second peak, which resulted in a reduction of sprinting, drifting, and in an inability to find the velocity refuges, or if *L. bocagei* were getting adapted to the peak. To address these questions, the simulation of more than one peak repetition is recommended.

There was not enough evidence supporting that a specific flow change resulted in an actual stress response. Thus, the identification of a flow threshold that represented the resting state for *L. bocagei* was not straightforward and deserves further investigation. Fish have evolved adaptive mechanisms to cope with natural extremes. However, under hydropeaking conditions, the flow changes are significantly more severe and frequent than in a free-flowing river. The energy-cost that is associated with those rapid and artificial changes is likely proportional. Although fish are able to adapt to novel conditions, the associated energy expenditure has considerable drawbacks. It will certainly reduce the energy available for diel activities or to successfully complete life-cycle stages. If a biomarker for flow variability is established, it will be possible to infer whether the flow fluctuations in natural conditions represent a potential stressor, or, otherwise, initiate behavioural strategies to compensate for the adverse flow fluctuations and maintain homeostasis.

Given the particular role of event predictability for *L. bocagei*, the study of alternative up-ramping and down-ramping rates is strongly advised. It is predicted that the slower onset of the up-ramping stage will enable fish to gradually adapt not only to the severity of a peak flow, but to the water level change. In the same way, slower down-ramping rates have the potential to reduce the stranding probability since the gradual flow decrease may function as a cue for fish to escape from potential isolated pools. A particular focus should be addressed to the critical down-ramping stage since this is usually associated with reduced chances of survival and high mortality rates.

The water velocity preferences of *L. bocagei* vary during its ontogeny. Given that most literature focuses on salmonids, the design of flow refuges according to velocity and habitat preferences for *L. bocagei*, and other cyprinid species, will likely differ. Thus, more studies focusing on other fish species (with distinct swimming modes) and habitat guilds are encouraged. In addition, it is necessary to further the investigation to specific life-cycle stages, namely larvae and juvenile survival, foraging, reproductive migrations, or spawning. Specific aspects of the life-cycle of salmonids have been recently proposed to be included during the development of mitigation solutions for salmonids affected by hydropeaking [81]. However, such guidelines are inexistent for cyprinids. In addition, flow regulations specifically addressed to hydropower plants operating in hydropeaking exist for a few regions in European countries, however, they focus exclusively on salmonid species [82]. Thus, it is critical to study the flow requirements for other fish families, cyprinids in particular, and for critical bottlenecks to ensure the sustainability of fish communities affected by hydropeaking, and to increase the awareness of policymakers to include flow recommendations in national legal instruments.

Local hydrodynamic changes were related to specific movement behaviour and physiological responses [58]. These findings represent the first step of a novel perspective to study behavioural patterns related to rapid flow fluctuations. Thus, it is still premature to generalize. To strengthen the findings regarding fluid-body interactions under rapid flow fluctuations and habitat heterogeneity, it is necessary to establish a quantitative relationship. With it, it would be possible to predict the magnitude of specific movement patterns, relate them with local hydrodynamic changes, and interpret them in terms of swimming effort.

Although it was possible to define guidelines for the design of deflectors and to propose alternative operational measures, it was not possible to upscale them in the river system. Thus, it should be reinforced that, prior to the implementation of any mitigation measure based on flume research, it is
necessary to perform experimental tests in riverine conditions. In addition, it is strongly recommended that future research combines flume experiments with research conducted in rivers.

7. Conclusions

The motivation to write this communication emerged from the need to assemble the methods and the main findings obtained from flume research, with the purpose that this knowledge could be used by freshwater scientists and hydropower producers. Although the scientific findings were obtained for an Iberian cyprinid inhabiting river affected by the Mediterranean climate, the methods that were adopted have the potential to be extended to other river systems affected by hydropoeaking, fish species, and particular bottlenecks. Despite the limitations inherent in indoor flumes, the possibility to control the flow changes and to quantify fish responses provided unique clues regarding fluid-body interactions, given by pressure thresholds, which are difficult to observe in natural conditions. The refined scale of the research not only identified the potential negative aspects of hydropoeaking, but also the opportunities that can emerge from them, and the ability of fish to adapt and compensate under adverse flow conditions. The use of diverse behavioural quantification methods (e.g., different levels of fish responses), flow sensing technologies and statistical tools were decisive to strengthen the validity of the findings and to identify fish-fluid relationships, according to flow variability. Emphasis is given to habitat heterogeneity as a decisive factor to propose solutions to mitigate hydropoeaking. This communication encourages further research to identify flow thresholds for key life-cycle stages and complementary river studies to design and assess mitigation solutions to hydropoeaking.


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