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PERFORMANCE OF A VERTICAL-SLOT FISH PASS FOR THE SEA LAMPREY *PETROMYZON MARINUS* L. AND HABITAT RECOLONIZATION

E. PEREIRA[a], B. R. QUINTELLA[a,b]*, C. S. MATEUS[a], C. M. ALEXANDRE[a], A. F. BELO[a], A. TELHADO[c], M. F. QUADRADO[c] AND P. R. ALMEIDA[a,d]

[a] *MARE—Marine and Environmental Sciences Centre, Universidade de Évora, Évora, Portugal*
[b] *Departamento de Biologia Animal, Faculdade de Ciências, Universidade de Lisboa, Lisbon, Portugal*
[c] *Departamento de Recursos Hídricos, Agência Portuguesa do Ambiente, I.P., Amadora, Portugal*
[d] *Departamento de Biologia, Escola de Ciências e Tecnologia, Universidade de Évora, Évora, Portugal*

ABSTRACT

In 2011, a vertical-slot fish pass was built at the Coimbra Açude-Ponte dam (Mondego River, Portugal), approximately 45 km upstream from the river mouth. The performance of this infrastructure for sea lamprey passage was evaluated between 2011 and 2015 using several complementary methodologies, namely radio telemetry [conventional and electromyogram (EMG)], passive integrated transponder (PIT) telemetry and electrofishing surveys. During the study period, the electrofishing revealed a 29-fold increase in the abundance of larval sea lamprey upstream of the fish pass. Of the 20 radio-tagged individuals released downstream from the dam, 33% managed to find and successfully surpass the obstacle in less than 2 weeks, reaching the spawning areas located in the upstream stretch of the main river and in one important tributary. Fish pass efficiency was assessed with a PIT antenna installed in the last upstream pool and revealed a 31% efficiency, with differences between and within migratory seasons. Time of day and river flow significantly influenced the attraction efficiency of the fish pass, with lampreys negotiating it mainly during the night period and when discharge was below 50 $m^3 s^{-1}$. Sea lampreys tagged with EMG transmitters took 3 h to negotiate the fish pass, during which high muscular effort was only registered during passage, or passage attempts, of the vertical slots. The use of complementary methodologies provided a comprehensive passage evaluation for sea lamprey, a species for which there is a considerable paucity of valuable data concerning behavioural, physiological and environmental influences on obstacle negotiation. Copyright © 2016 John Wiley & Sons, Ltd.

KEY WORDS: fishway monitoring; upstream migration; efficiency; swimming behaviour; telemetry; electromyogram; PIT

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INTRODUCTION

Installation of fish passes has become a widespread measure to mitigate the impacts of anthropogenic barriers on fish migration and to improve the longitudinal connectivity of watercourses. The first facilities to be installed were usually designed as species specific, mainly for salmonids, and only recently fish passes were designed for a wider set of target species (Larinier and Marmulla, 2004). Even though it is generally accepted that adult anadromous lampreys must have access to upstream spawning areas, these species are rarely considered during the design or retrofitting of fish pass solutions (Kemp *et al*., 2011; Moser *et al*., 2011). Consequently, most fish pass solutions are not efficient for lampreys (Haro and Kynard, 1997; Laine *et al*., 1998; Moser *et al*., 2002, 2015), which is considered to be related with the poor swimming performance of these species (Beamish, 1978; Quintella *et al*., 2004).

*Correspondence to: B. R. Quintella, MARE—Marine and Environmental Sciences Centre, Universidade de Évora, Évora, Portugal.
E-mail: bsquintella@fc.ul.pt

Despite being a group of high conservation priority (Maitland *et al*., 2015) and economic interest, because of either commercial fishing activities (Almeida *et al*., 2002b) or control and eradication outside the natural distribution range (i.e. landlocked sea lamprey *Petromyzon marinus* L.; McLaughlin *et al*., 2007), so far, few studies have assessed the performance of distinct fish pass solutions for lampreys (Noonan *et al*., 2012). Fish pass monitoring is crucial to evaluate the suitability of the infrastructure for the target species, determine its efficiency and implement, if needed, the necessary adjustments (Naughton *et al*., 2007; Keefer *et al*., 2011). Among the few studies with lampreys, a significant number have focused on behaviour and fish pass performance through the use of telemetry techniques such as radio and/or passive integrated transponder (PIT) telemetry (e.g. Moser *et al*., 2002; Johnson *et al*., 2009; Keefer *et al*., 2010; Foulds and Lucas, 2013). Physiological telemetry [electromyogram (EMG)] has also been used by some authors to study the fine-scale behaviour of sea lamprey during spawning migration through difficult passage areas (Quintella *et al*., 2004; Almeida *et al*., 2005). However, in general, post-passage consequences on survival (e.g. Roscoe *et al*., 2010) and recruitment (e.g. Hogg *et al*., 2013), or the cumulative effects of barriers, are still poorly understood and should be included in programmes for fish pass assessment (Castro-Santos *et al*., 2009).

The sea lamprey is the largest extant petromyzontid species, attaining a maximum length of 1.2 m and weighing up to 2.3 kg (Hardisty, 1986). In Europe, it is strictly anadromous (Hardisty, 1986); thus, unimpeded migration corridors along rivers for downstream and upstream migrants are considered essential to maintain population levels in a favourable conservation status (Mateus *et al*., 2012; Maitland *et al*., 2015). Because of its decline across Europe, sea lamprey is listed under Annex II of the European Union Habitats Directive and Appendix III of the Bern Convention and is listed in the OSPAR convention list (Convention for the Protection of the Marine Environment of the North-East Atlantic) of threatened and/or declining species. Habitat fragmentation and reduction by construction of man-made barriers are considered two of the main threats to the sea lamprey entering Portuguese river basins (Mateus *et al*., 2012). One of the most important Portuguese river basins that represent a stronghold for diadromous species with a notorious conservation status and high socio-economic value is the river Mondego. However, until 2011, the installation year of a new fish pass designed for anadromous clupeids (*Alosa alosa* L. and *Alosa fallax* Lacepède, 1803) and sea lamprey, the available freshwater habitat for anadromous species was reduced to a 16-km stretch downstream of the first impassable dam (Almeida *et al*., 2000).

In the present study, complementary methodologies were used to evaluate the performance of the vertical-slot fish pass installed in Açude-Ponte dam, the first impassable obstacle in river Mondego. Specifically, this study was conducted to: (i) assess the behaviour of sea lamprey downstream and upstream of the fish pass through conventional radio telemetry; (ii) assess the fine-scale behaviour during the negotiation of the vertical-slot fish pass, using EMG telemetry; (iii) evaluate the overall passage efficiency of this infrastructure using PIT telemetry; and (iv) monitor the species recolonization of the upstream stretch of river Mondego (not available for 30 years), with electrofishing surveys.

MATERIAL AND METHODS

Study area

This study was conducted in the river Mondego, a highly regulated 234-km-long system in central Portugal (Figure 1). The Coimbra Açude-Ponte dam, located 45 km upstream the river mouth, is a gate dam with a height of 6.2 m, built in the early 1980s for discharge control, public and industry water supply and irrigation. This infrastructure was equipped, since the beginning, with an inadequate pool and weir fish pass with submerged orifices, which was not used by the target species. Consequently, anadromous species were forced, in regular hydrological conditions, to complete their life cycle downstream from this obstacle, in the lower 13-km stretch of available freshwater

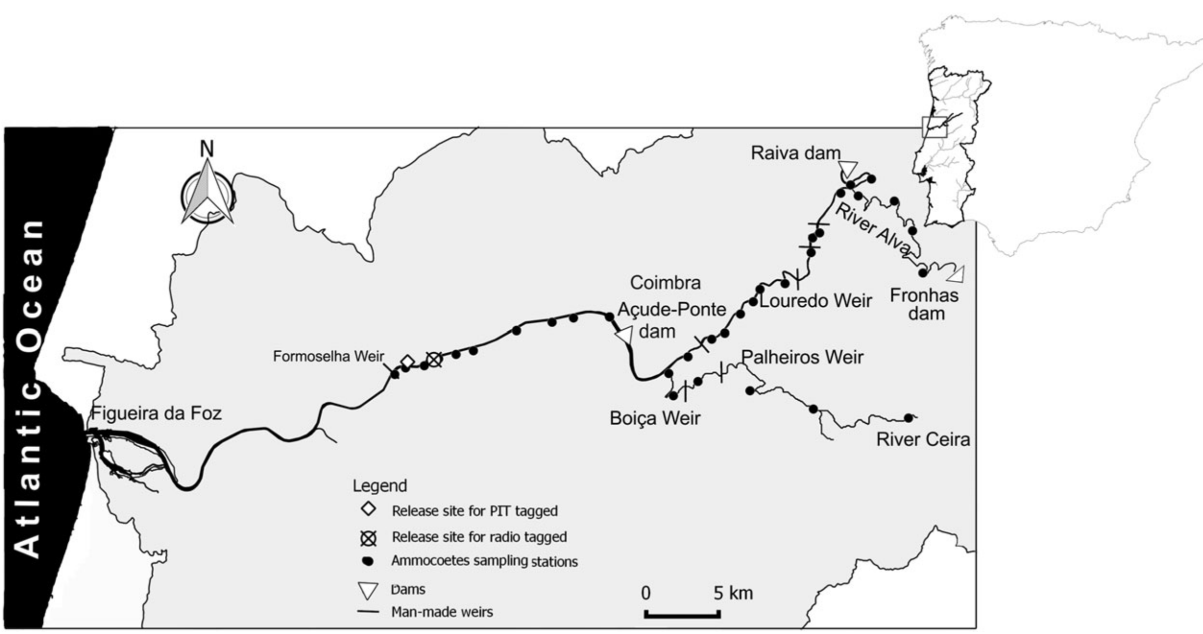

Figure 1. Map of the river Mondego showing details of the study area. Release sites for tagged lampreys and electrofishing sampling stations are also represented.

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habitat (Almeida *et al*., 2000). In 2011, a vertical-slot fish pass specifically designed to target anadromous clupeids and sea lamprey was constructed. The fish pass is a 125-m-long channel with 2-m water depth, divided into 23 uniform rectangular pools (4.5×3.0 m) connected by vertical slots (0.5-m width). The water level difference between adjacent pools is 0.25 m. The average velocity at the slots is ~1.1 $\mathrm{m\,s^{-1}}$, with dissipated powers in the resting pools below 150 $\mathrm{W\,m^{-3}}$. Fish pass flow is constantly maintained at 2.0 $\mathrm{m^3\,s^{-1}}$.

The upper Mondego basin is impounded by several dams, Raiva and Aguieira dams, in the main stem (located 80 and 86 km upstream from the river mouth, respectively), and Fronhas dam, in the tributary river Alva. Raiva dam presently represents the first impassable obstacle in the river Mondego main course, but within the 40 km of river between the Coimbra Açude-Ponte and Raiva dams, four low-head weirs partially block the spawning migration of sea lampreys during normal-flow periods (Figure 1).

Electrofishing surveys

To estimate the relative abundance [catch per unit effort (CPUE), ind $\mathrm{h^{-1}}$] of larval lamprey (i.e. ammocoetes), sampling was carried out at two distinct river stretches. In the lower stretch, downstream from the Coimbra Açude-Ponte dam (DM), 10 sampling stations were chosen. In the upstream river stretch (UM), which includes the tributaries Alva and Ceira, 23 sampling stations were selected (Figure 1). Each station was sampled once a year, between 2011 (fish pass pre-operational phase) and 2015. Sampling was conducted during August, when capture efficiency is higher because of low streamflow conditions, using a single-pass electrofishing method in wadeable areas (Steeves *et al*., 2003; Moser *et al*., 2007) with an electroshocker (Hans Grassl EL62, 600 V DC, 10A, Schönau am Königssee, Germany)). Sampling time per station ranged between 0.5 and 1.13 h, and fishing effort was directed towards preferred ammocoete habitats (cf. Almeida and Quintella, 2002). All captured ammocoetes were anaesthetized with a solution of 0.3 mL of 2-phenoxyethanol per litre of water and measured to the nearest millimetre (total body length, *Lt*).

A univariate two-way permutational multivariate analyses of variance (PERMANOVA) was used to statistically compare ammocoete abundance between sampling sites and years (factors: Site and Year). Captured ammocoetes were binned by 20-mm length classes, and a multivariate two-way PERMANOVA, followed by a SIMPER procedure, was applied to test for differences in length frequency distribution among sampling Sites and Years. PERMANOVA analyses were performed with the package PERMANOVA for PRIMER + v6.0 (Anderson *et al*., 2008).

Radio telemetry

Radio telemetry was used to study the migratory behaviour of adult sea lampreys. Between February and March 2013, a total of 20 sea lampreys caught with a fyke net when entering the river Mondego were tagged with external radio transmitters (ATS-F2020: 8 g in air, 12 mm in diameter and 46 mm in length), manufactured by Advanced Telemetry Systems, Isanti, MN, USA. The transmitters weighed less than 1% of the sea lamprey's body weight in the air. Fish were anaesthetized in a solution of 0.4 mL 2-phenoxyethanol per litre of water, measured and externally tagged following the method described by Almeida *et al*. (2000). After recovery, lampreys were released approximately 13 km downstream from the Coimbra Açude-Ponte dam (Figure 1). Tracking surveys were conducted weekly until July 2013 using a radio receiver (model R410, manufactured by Advanced Telemetry Systems) coupled to a Yagi antenna, and lamprey locations were geo-referenced with a Global Positioning System hand-held unit.

The total time between release and the first detection upstream from this obstacle was calculated for each lamprey. Data also allowed access to the proportion of lampreys that were able to negotiate the dam, to analyse their dispersal pattern upstream of the Açude-Ponte dam and to identify potential constraints to their movements upstream of this obstacle.

Electromyogram telemetry

Electromyogram radio telemetry, which measures the level of muscle activity as a proxy for the amount of physical effort performed, was used to study the fine-scale behaviour of adult sea lamprey during fish pass negotiation. During the 2014 spawning season, three lampreys caught upstream of the Formoselha weir were tagged with implantable coded EMG radio transmitters (cEMG-R11-25: 12 g in air, 12 mm in diameter and 56 mm in length), manufactured by Lotek Wireless, Newmarket, Ontario, Canada. The surgical procedure followed that described by Quintella *et al*. (2004). Briefly, after being anaesthetized and measured, a tag was surgically implanted in the intraperitoneal cavity, and a pair of gold-tipped electrodes was fixed in the axial red musculature, allowing registration of bioelectrical voltage changes that are strongly related to the strength and duration of muscle contraction (Kaseloo *et al*., 1992) and thus obtaining a relative index of muscle activity (EMG), ranging between 0 and 50 (Quintella *et al*., 2009).

After surgery, lampreys were left in an aerated recovery tank for a 24-h period during which the average EMG value associated with the motionless resting behaviour (EMG_{rest}) was recorded and defined as the threshold value for motionless behaviour. Lampreys were released in the fish pass entrance at dusk and were continuously monitored

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during negotiation. In case of a fallback from the fish pass, swimming activity was still continuously monitored for a period of 24 h after release.

Passive integrated transponder telemetry

Fish pass efficiency was assessed using a PIT antenna system installed at the fish pass (IS1001-MTS with thin-wall pass-through antenna; Biomark, Boise, ID, USA). The system consisted of a custom, thin-wall-shielded antenna that was approximately 0.7 m wide × 2.5 m tall. The antenna was placed between the last pool and the fish pass exit.

During the spawning season of 2014 and 2015, 225 and 103 sea lampreys, respectively, were captured with a fyke net in the lower tidal stretch of the river Mondego and PIT tagged. While in 2014, all individuals were captured and released during the migratory peak (April), in 2015, lampreys were captured and released throughout the spawning season, that is, January (10%), February (30%), March (30%) and April (30%), to assess potential differential behaviour of the species throughout the migratory season. Lampreys were anaesthetized (0.4 mL 2-phenoxyethanol per litre of water), measured (*Lt*) and weighted (*Wt*), and a PIT tag (High Performance Tag, 12.5 mm, 134.2 kHz; Biomark) was inserted ventrally in the abdominal cavity with an implant gun. In both years, individuals were released 15 km downstream from the fish pass (Figure 1).

Fish pass efficiency was defined as the proportion of tagged lampreys exiting the fish pass out of 90% of the total tagged lampreys. This percentage was based on the results from the radio telemetry conducted in this study that showed that 10% of the tracked animals never approach the fish pass. In 2015, the efficiency was also estimated for the migratory peak (March/April).

Generalized linear models were used to assess the influence of environmental predictors in the attraction efficiency of the fish pass. Data included the period between 06/04/2014 (the beginning of tagging) and 17/05/2014 (last individual detected). Two periods of day (night versus day) were defined according to the sunrise and sunset times, and mean temperature (°C), turbidity (Formazin Nephelometric Unit) and flow ($m^3 s^{-1}$) were obtained through a multiparameter probe and data provided by the Portuguese Environment Agency. The model was fitted using a negative binomial distribution, and goodness of fit to the expected distribution was tested with a chi-squared test. Modelling was performed using RSTUDIO v 0.99.491 (RStudio, Inc., Boston, MA, USA) and the stepAIC function (Venables and Ripley, 2002). Model fit was evaluated using the Akaike information criterion (Johnson and Omland, 2004). The significance of the variables included in the most parsimonious model was tested using a chi-squared test. A significance threshold of $p < 0.05$ was considered in all test procedures. To identify the flow conditions in which of these animals entered the fish pass, we identified the time of entrance (time of detection minus estimated passage time—3 h) and the respective flow discharge.

Median time between lamprey release and the first detection was estimated, and frequency distribution of the first detections recorded at each hour of the day was analysed. Size-related differences in passage were also analysed, considering three size classes (<800, 800–900 and >900 mm), with a G-test of independence.

RESULTS

Abundance and structure of larval sea lamprey population

PERMANOVA analyses of CPUE data identified significant effects of Year ($p < 0.002$), Site ($p = 0.001$) and their interaction ($p = 0.025$). Pairwise comparisons indicated that UM significantly differed from DM in every sampling year except 2015 ($p = 0.13$). In the first operational year (i.e. 2012), the CPUE was very similar to the pre-operational situation (i.e. 2011) (DM = 2.40 ind h^{-1} and UM = 0.26 ind h^{-1}) (Figure 2). Although the following years (2013 and 2014) revealed an overall increase, only in 2015 was a shift observed between the two sites. Subsequent pairwise analysis revealed no significant differences ($p = 0.19$) between sites (CPUE: DM = 11.34 ind h^{-1} and UM = 12.27 ind h^{-1}). From the group of ammocoetes captured in UM in 2015, 92% ($n = 200$) were found in the tributary river Ceira.

Length frequency (Figure 3) multivariate two-way PERMANOVA also showed significant effects of Year ($p = 0.001$), Site ($p = 0.001$) and their interaction ($p = 0.001$, Tables Ia and Ib). Additional pairwise comparisons indicated that, in UM, the year 2015 was significantly different from 2011 ($p = 0.013$) and 2012 ($p = 0.008$), with classes 100–120 and 80–100 mm being more abundant in 2015. In

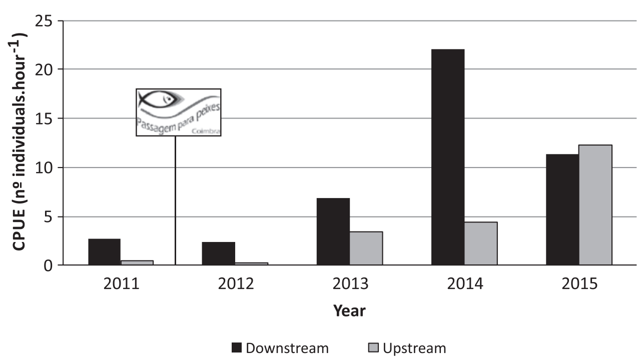

Figure 2. Catch per unit effort (CPUE; ind h^{-1}) of sea lamprey ammocoetes between 2011 and 2015 for downstream and upstream areas of the Coimbra Açude-Ponte dam. Fish pass operational year is indicated by the Portuguese fish pass logotype

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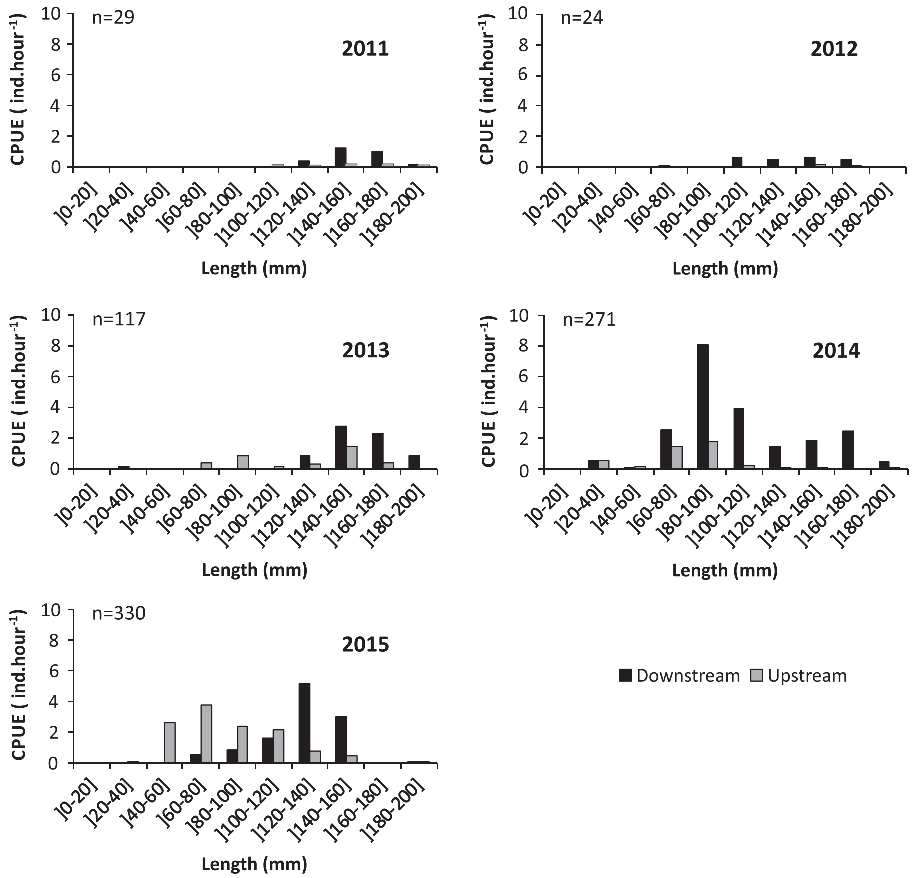

Figure 3. Length frequency distribution (catch per unit effort—CPUE; ind h^{-1}) of sea lamprey ammocoetes between 2011 (fish pass pre-operational monitoring) and 2012–2015 (post-operational monitoring) for downstream and upstream areas of the Coimbra Açude-Ponte dam

DM, 2014 was different from the remaining sampled years, and the remaining variation did not show a clear pattern (Tables Ia and Ib).

Migration pattern of radio-tagged adult sea lampreys

From the 20 radio-tagged lampreys, 90% ($n = 18$) reached the Coimbra Açude-Ponte dam by completing the 13-km river stretch from the release point up to the dam. The remaining individuals stopped in an area frequently used as spawning ground. From the group of individuals that reached the dam, six (33%) successfully passed this obstacle and reached the upstream areas with a median time of 11 days (10–22 days). When migrating in the upstream stretch, half of these animals ($n = 3$) continued through river Ceira, and only one individual was able to negotiate the first

Table Ia. Main test: results of the permutational multivariate analyses of variance applied on length frequency distribution among sampling Sites and Years

Factors	MS	Pseudo-*F*	*p*
Site	34 199	38	<0.01
Year	5893.2	6.54	<0.01
Site × year	3870	4.3	<0.01

MS, Mean squares.

Table Ib. Pairwise test: results of the permutational multivariate analyses of variance applied on length frequency distribution among sampling Sites and Years

	Downstream stretch		Upstream stretch	
	T	*p*	*T*	*p*
2011 × 2012	1.43	ns	0.48	ns
2011 × 2013	1.14	ns	1.40	ns
2011 × 2014	3.32	<0.01	1.04	ns
2011 × 2015	1.74	ns	1.94	0.01
2012 × 2013	2.24	0.01	1.83	ns
2012 × 2014	2.93	<0.01	1.35	ns
2012 × 2015	1.24	ns	2.33	<0.01
2013 × 2014	3.01	<0.01	0.63	ns
2013 × 2015	2.04	<0.01	0.93	ns
2014 × 2015	1.94	0.03	1.05	ns

ns, not significant.

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obstacle in this tributary (Boiça weir) located 4 km upstream the confluence with the river Mondego (Figure 4). Only three of the individuals that negotiated the fish pass continued their upstream migration in river Mondego main stem, one of which successfully surpassed the Palheiros weir and reached the Louredo weir (16 km from the Coimbra Açude-Ponte dam) (Figure 4).

From the three individuals tagged with EMG transmitters, two (Pm1 and Pm2) managed to successfully negotiate the fish pass. Both took approximately 3 h to complete the passage. A high muscular effort was only occasionally registered during the passage, or passage attempts, of the vertical slots that connect pools (Figure 5). In contrast, Pm3 fell back from the entrance and did not negotiate the fish pass. This individual remained downstream the spillway area (i.e., tailwater) and exhibited a high muscular effort when trying to overcome the high discharge released from the gates (Figure 5). When compared with the frequency distribution of EMG values from Pm1 and Pm2, this lamprey clearly displays higher frequency of high EMG values (Figure 6).

Fish pass efficiency—Passive Integrated Transponder telemetry study

During the peak of the spawning season of 2014, the overall efficiency was 31% and was not size related; that is, fish size and successful passage were statistically independent (G-test, d.f. = 2, $p > 0.05$). Lampreys took a median time of 14 days (range of 1.5–34 days) to first detection after release, and 88% of the animals were detected at the following instances: (i) the period between dusk and dawn (Figure 7) and (ii) flow discharge below 50 $m^3 s^{-1}$. The fitted generalized linear model explained 48% of the total variation in PIT detections, as estimated by the deviance of the final model (52.7) and that of the null model (55.6), with the variables time of the day and flow contributing 21.8% and 22.6%, respectively (Table II). T2

Sea lamprey behaviour varied during the spawning season of 2015, with lower proportions of tagged individuals negotiating the obstacle in the beginning of the migratory period (January 10% and February 11%) and maximum proportions during the peak of the spawning migration. During the migratory peak, pass efficiency was 21%. As in 2014, 75% of the animals were detected between dusk and dawn (Figure 7).

DISCUSSION

Before the vertical-slot fish pass construction, the Coimbra Açude-Ponte dam was clearly blocking the upstream migration of adult sea lampreys, as revealed by the 2011 results of near-zero ammocoete abundance. However, the presence of a few larvae belonging to higher length classes confirms that, in years of exceptionally large floods, some animals were able to negotiate this obstacle through the gates that open completely with discharge flows above 900 $m^3 s^{-1}$ (Almeida *et al*., 2002a), a situation that typically occurs four times per decade.

Both radio and PIT telemetry showed similar results in terms of overall passage efficiency (33% and 31%, respectively) between 2013 and 2014. These methods also documented a similar time required to find the fish pass entrance, negotiate it and reach the upstream stretch (11 and 14 days, respectively). These results apparently indicate that even relatively low samples (20 radio-tagged individuals) can be representative of the overall efficiency of a fish pass solution for sea lampreys.

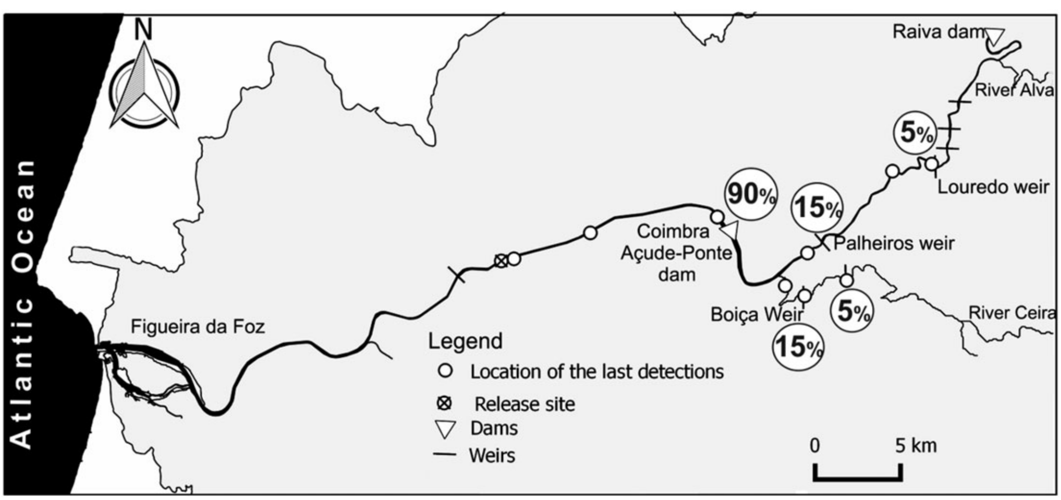

Figure 4. Dispersal pattern of the 20 radio-tagged adult sea lampreys released in the Mondego River. Open circles indicate final positions where lampreys were located. The proportion of animals that managed to negotiate each obstacle is also represented

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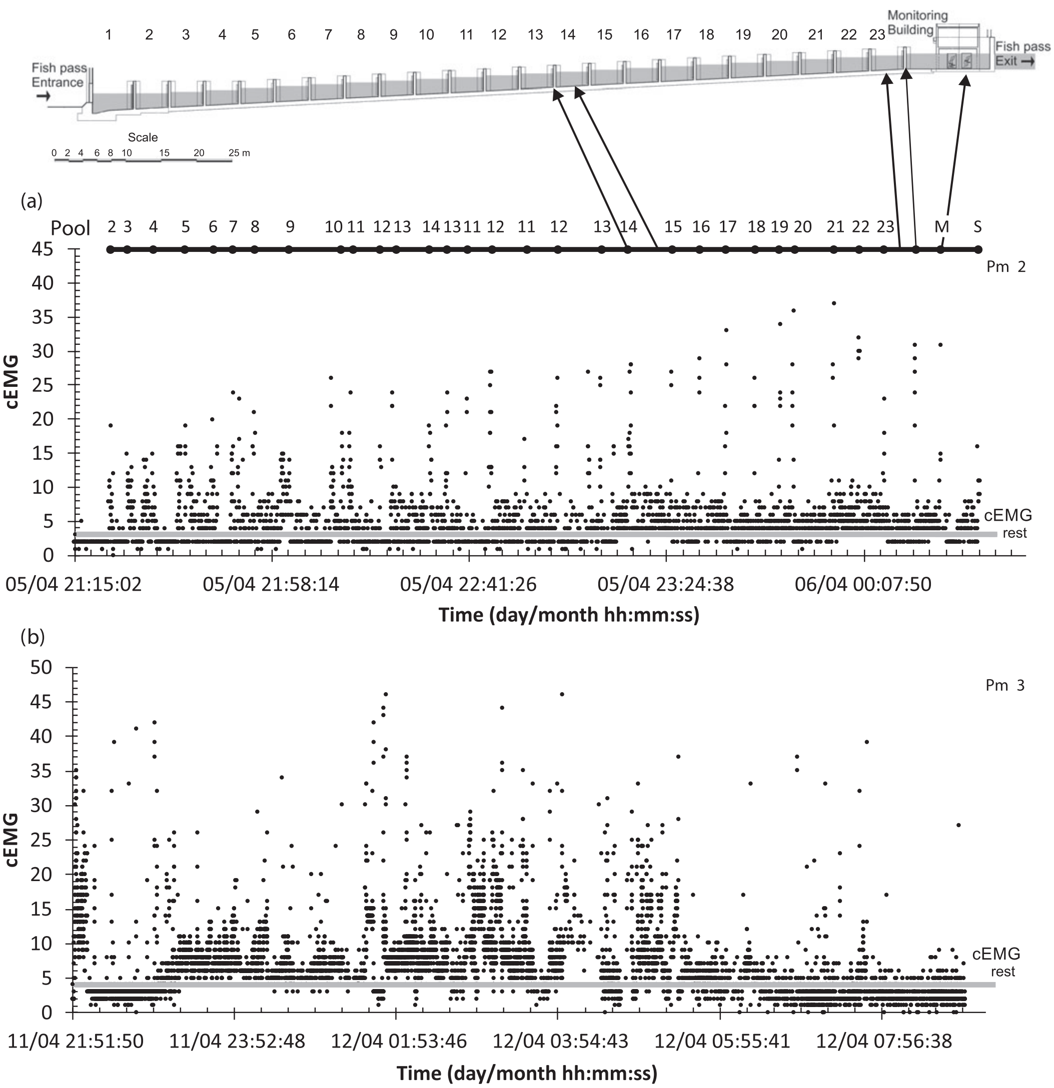

Figure 5. Behaviour [recorded as coded electromyogram (cEMG) values—i.e. muscle activity level] of lampreys: (a) Pm2 during fish pass negotiation and (b) Pm3 in the tailwater when attempting to overcome the dam through the spillway gates. In the *y*-axis, the grey line represents the average resting cEMG value ($cEMG_{rest}$). The specific location of Pm2 in each of the 23 pools that constitute the fish pass is represented on top of the chart; each dot represents the negotiation of the respective vertical slots that connect the pools. The longitudinal profile of the fish pass is also represented

Sea lampreys entering the Garonne and Dordogne Rivers in France are known to successfully use vertical slot fish passes facilities and fish elevators during their upstream migration, although the efficiency of each type of installation was never estimated (Travade *et al*., 1998). A study from Moser *et al*. (2002) with the Pacific lamprey (*Entosphenus tridentatus* Richardson, 1836), in the lower Columbia River (1997–2000), identified an overall passage efficiency of 38–47% at Bonneville dam and 50–82% at the Dalles dam. However, these are pool and weir fish passes with hydraulic conditions that are different from the Açude-Ponte dam vertical-slot fish pass. Additionally, Pacific lampreys have the ability to climb vertical surfaces (Reinhardt *et al*., 2008), which is quite distinct from the sea lamprey which possesses a higher swimming performance but without climbing capabilities.

From the analysis of the interaction between the abiotic factors and PIT tag detections, flow and circadian rhythms were identified as the factors that better explained the number of tagged lampreys moving through the fish pass. The majority of the movements happened with a peak frequency associated with low discharge (inferior to $50\,m^3\,s^{-1}$) and during dusk and darkness, confirming the typical nocturnal behaviour described for this species (Almeida *et al*., 2000; Moser *et al*., 2015).

Because of the strong rheophilic behaviour of sea lampreys, the high discharges released by the dam seem to prevent animals from finding the fish pass entrance, diverting them to the

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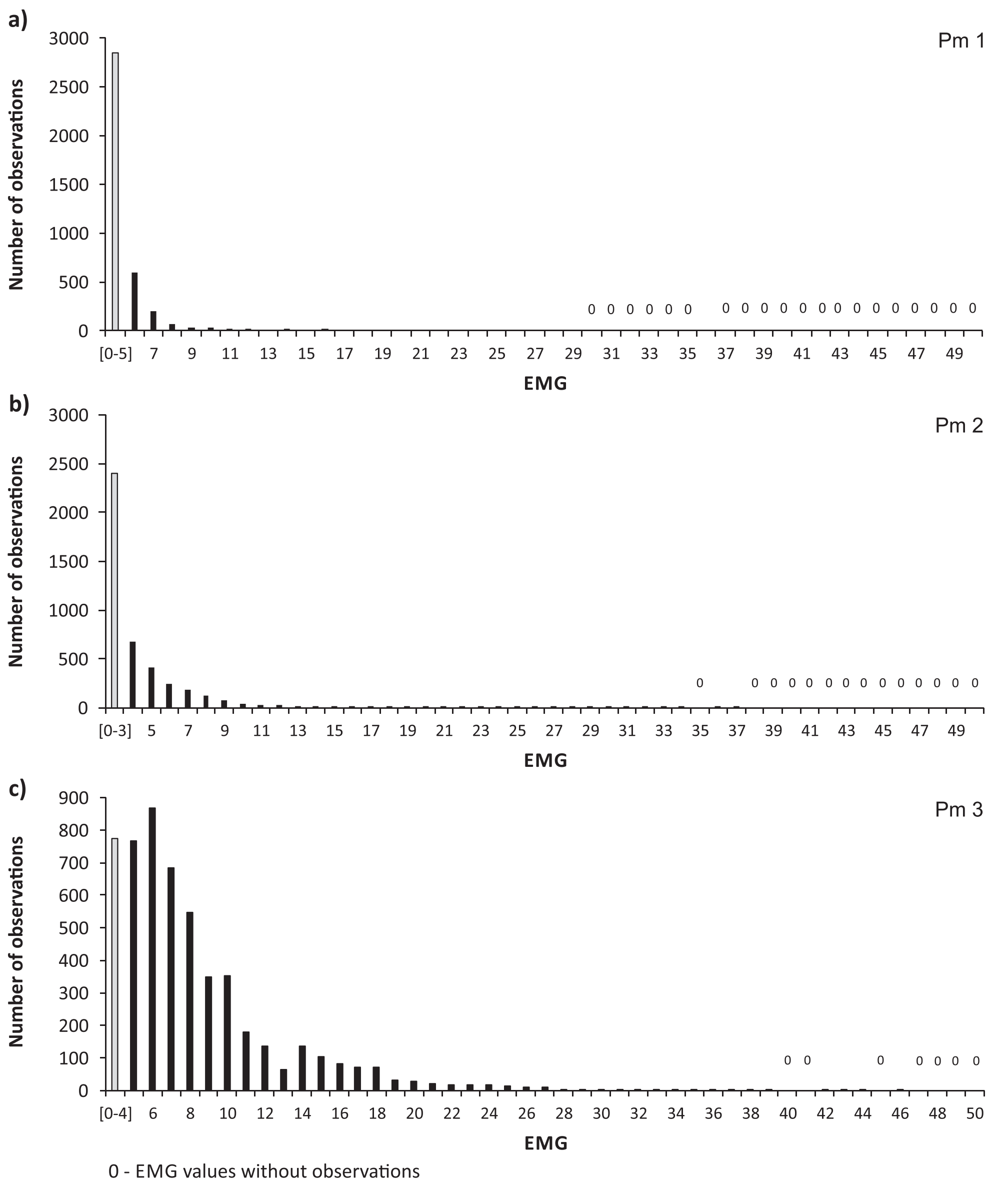

Figure 6. Frequency distribution of electromyogram (EMG) values from the three lampreys tagged with EMG transmitters: (a, b) data from lamprey Pm1/Pm2 that successfully negotiated the fish pass and (c) data from lamprey Pm3 that developed a high activity in front of the dam gates. Grey bars represent the resting EMG value

middle gate area where the strongest flow is released (Figure 8). This behaviour was assessed with the manual radio-tracking sessions. Additionally, as seen with the EMG data, the lamprey that did not enter the fish pass expended a considerable amount of energy trying to overcome the dam through the gates. EMG studies of free-swimming sea lamprey in the river Mondego identified multiple bursts of high-intensity exercise during difficult passage areas that seem to increase lampreys' fatigue (Quintella *et al*., 2004, 2009).

Once sea lampreys entered the fish pass, the negotiation took 3 h, and the higher muscular effort (EMG data) was registered only during the movement through or passage attempts at the vertical slots. Previous observations of burst swimming while negotiating submerged blockstone weirs (Quintella *et al*., 2004) suggest that energy use could be higher, confirming that fish pass negotiation at the Açude-Ponte dam was not energetically demanding. By holding their position with the oral disc (Quintella *et al*., 2004), the amount of energy can be reduced to a minimum, which can be a benefit when compared with other fish with continuous swimming behaviours. However, data concerning the behaviour of the sea lamprey during the fish pass negotiation need to be interpreted carefully because of the small sample size used.

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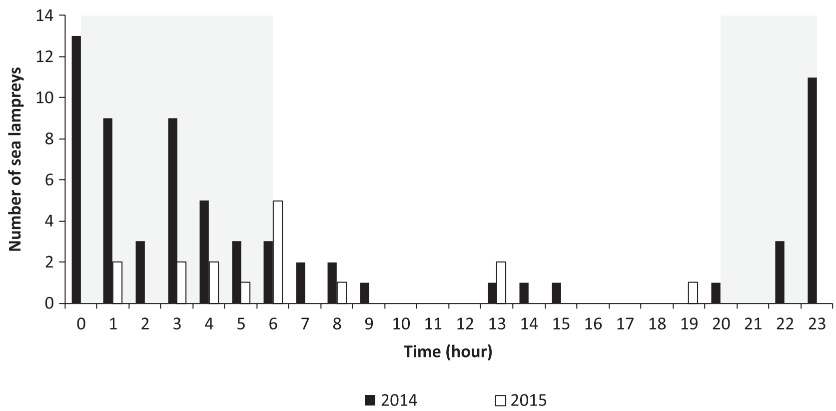

Figure 7. Circadian rhythm of passive integrated transponder-tagged sea lampreys detected exiting the Coimbra Açude-Ponte dam fish pass in 2014 and 2015. Grey bars indicate night periods according to local sunrise and sunset times

After fish pass negotiation, lampreys continued their upstream migration apparently with no behavioural changes, despite the presence of a ~4-km stretch with lentic characteristics that could have disoriented the adults and disrupted the migratory behaviour (Moser *et al*., 2015). When migrating in the lotic upstream stretch, migratory movement only stopped below relatively small weirs that, during periods of regular flow, are almost impassable for this species. Combining the ammocoete surveys with the fish passage outcomes allowed us to infer on the impact of the fish pass on the recolonization of the upstream stretch of river that was previously unavailable. Even with a relatively low passage efficiency when compared with values close to 90% that are commonly described in the literature for salmonids (e.g. Gowans *et al*., 1999; Burnett *et al*., 2014), a rapid recolonization of the upstream reaches was observed for sea lamprey. Larval abundances increased 29 times upstream the Coimbra Açude-Ponte dam compared with an increase of four times in the downstream stretch. Increases in younger classes (age groups below 2+ years) were particularly notable (Quintella *et al*., 2003), contributing to the river's 'pheromonal landscape' (Dawson *et al*., 2015). Nevertheless, abundances of ammocoetes belonging to the 0+ year age group were probably underestimated because of the low efficiency of electrofishing in sampling this length class (Lasne *et al*., 2010). The large increment of ammocoetes abundance in 2014 is considered an example of that, because it revealed an exceptional recruitment in the previous year that was not identified through captures of 0+ year age class during the 2013 electrofishing survey.

Table II. Goodness-of-fit statistics for the GLM fitted to assess the influence of environmental predictors in the attraction efficiency of the vertical-slot fish pass to PIT-tagged sea lampreys

Model	Coef.	Res. dev	Deviance	% exp.	*p*-value (χ^2)
Period of the day	2.27	176.23	49.184	21.8	<0.001
River flow	−0.03	125.27	50.957	22.6	<0.001
Total explained				**48%**	

The bold value indicates total deviance.
GLM, generalized linear model; PIT, passive integrated transponder; Coef., coefficient; Res. dev., residual deviance; % exp., percentage of the total deviance explained.

Even though river Ceira is a small system, representing 25–35% of the mean flow observed in the main stem, the river Mondego, half of the tagged spawners chose this tributary to reproduce. This is in agreement with the results attained with larval surveys, where a high abundance of ammocoetes was particularly detected in this tributary. This confirms the importance of smaller tributaries, like the river Ceira, for sea lamprey. The river Ceira is one of the most important tributaries of the Mondego basin with an average flow during the sea lamprey spawning migration of $4.4\,m^3\,s^{-1}$ and a drainage area of $736.6\,km^2$ (representing 11% of the Mondego River basin total drainage area). Together with the presence of adequate coarse substrate for the construction of nests for spawning activities (Hardisty, 1986), in Lower Ceira, there is also abundance of silt and sandy sediment, which is suitable for larval burrowing (Almeida and Quintella, 2002; Dawson *et al*., 2015). The fast recolonization capacity of sea lamprey after dam removals was also identified by Hogg *et al*. (2013) in a third-order tributary of river Penobscot (Maine, USA), where in 2 years after the dam removal, the abundance of nesting sites increased more than fourfold and their spatial distribution trended towards a more equitable longitudinal arrangement between downstream and upstream reaches of the former Mill dam.

The information gathered in this study clearly shows that the vertical-slot fish pass installed in Coimbra Açude-Ponte dam is contributing to the improvement of the longitudinal connectivity of the river Mondego and to the increase in the available habitat, not only for sea lamprey but also for other

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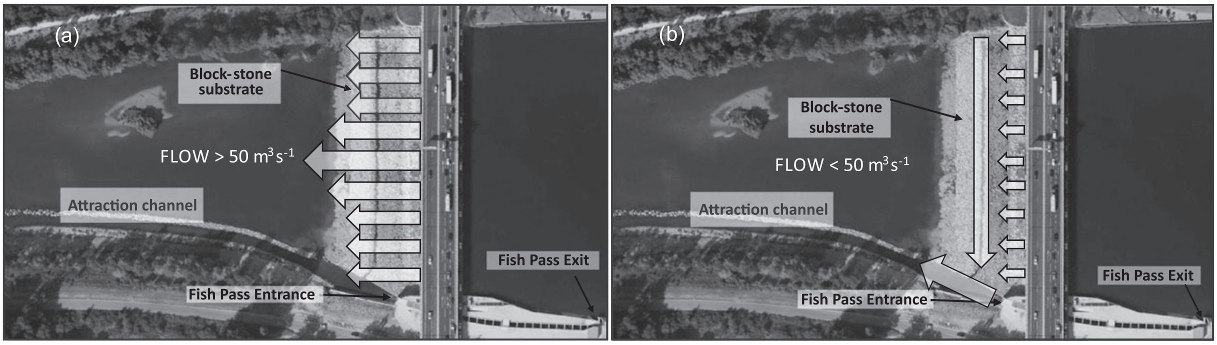

Figure 8. Schematic representation of the discharge flow pattern downstream Coimbra Açude-Ponte dam

diadromous species (Cardoso, 2014). The complementary sampling methods allowed a comprehensive evaluation of the fish pass performance and its impact on the abundance and habitat use of the sea lamprey in upstream reaches. Other obstacles were identified in the upper stretch of the river Mondego that, despite their relatively small height, may reduce substantially or completely interrupt the upstream movement of sea lamprey. Recently, the restoration of the longitudinal continuity in the river Mondego was complemented with the construction of five nature-like fish passes (one located downstream the Coimbra Açude-Ponte dam and four others located upstream) at smaller weirs identified as problematic for fish migration (cf. Figure 1). The performance of the new fish passes and their impact on habitat recolonization will continue to be assessed through the integrative approach reported here. This study is significant in that it presents the first data on the performance of a fish pass solution for sea lamprey, a highly valued species of both conservation and commercial importance.

For future fish pass monitoring programs targeting sea lamprey, we suggest that when assessing their efficiencies, animals should be preferably tagged during the peak of the spawning migration, as the bulk of individuals, that is, more than 90%, use the fish pass during this period. In this study, we obtained comparable results when assessing the overall efficiency of the fish pass with radio telemetry ($n = 20$) and PIT telemetry ($n = 225$) using sample sizes with different orders of magnitude. Thus, we consider that, for sea lamprey, a sample size of 25–50 individuals is acceptable to estimate the overall efficiency of a single fish pass. Being an anadromous and highly rheophilic species, tagged sea lampreys should be released downstream the dam, instead of the vicinity of the fish pass, to increase recovery time and allow more time to reinitiate a natural migratory behaviour. By doing so, the total number of animals that actually reach the dam should be assessed and accounted to correct the overall efficiencies obtained. Finally, fish pass performance should be assessed in more than one single spawning season, because interannual variability in fish pass efficiencies can occur in response to variation of environmental factors, such as discharged flow or water temperature, which can significantly condition the performance of the fish pass solution.

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REFERENCES

Almeida PR, Quintella BR. 2002. Larval habitat of the sea lamprey (*Petromyzon marinus* L.) in the River Mondego (Portugal). In Freshwater Fish Conservation: Options for the Future, Collares-Pereira MJ, Coelho MM, Cowx IG (eds). Fishing News Books, Blackwell: Oxford.

Almeida PR, Silva HT, Quintella BR. 2000. The migratory behaviour of the sea lamprey *Petromyzon marinus* L., observed by acoustic telemetry in River Mondego (Portugal). In Advances in Fish Telemetry, Moore A, Russel I (eds). CEFAS: Lowestoft, Suffolk; 99–108.

Almeida PR, Quintella BR, Dias NM. 2002a. Movement of radio-tagged anadromous sea lamprey during the spawning migration in the River Mondego (Portugal). *Hydrobiologia* **483**: 1–8.

Almeida PR, Quintella BR, Dias NM, Andrade N. 2002b. The anadromous sea lamprey in Portugal: biology and conservation perspectives. In Moser M, Bayer J, MacKinlay D (eds). The Biology of Lampreys,

DOI: 10.1002/rra

Symposium Proceedings. International Congress on the Biology of Fish. American Fisheries Society, 21–25 July, Vancouver; 49–58.

Almeida PR, Quintella BR, Koed A, Andrade NO .2005. Using electromyogram telemetry to study the spawning migration of sea lamprey (*Petromyzon marinus* L.). In Spedicato MT, Lembo G, Marmulla G (eds), Aquatic Telemetry, Advances and Applications. FAO-COISPA: Rome; 3–11.

Anderson MJ, Gorley RN, Clarke KR. 2008. PERMANOVA for PRIMER: Guide to Software and Statistical Methods. PRIMER-E Ltd.: Plymouth, United Kingdom; 214.

Beamish FWH. 1978. Swimming capacity. In Fish Physiology, Hoar WS, Randall DJ (eds), vol. **VII**. Academic Press: New York; 101–187.

Burnett NJ, Hinch SG, Donaldson MR, Furey NB, Patterson DA, Roscoe DW, Cooke SJ. 2014. Alterations to dam-spill discharge influence sex-specific activity, behavior and passage success of migrating adults sockeye salmon. *Ecohydrology* **7**: 1094–1104.

Cardoso G. 2014. Monitorização da passagem para peixes do Açude-Ponte em Coimbra: otimização da metodologia de contagem. Master's thesis, University of Évora, Évora.

Castro-Santos T, Cotel A, Webb P. 2009. Fishway evaluations for better bioengineering: an integrative approach. *American Fisheries Society Symposium* **69**: 557–575.

Dawson HA, Quintella BR, Almeida PR, Treble AJ, Jolley JC. 2015. The ecology of larval and metamorphosing lampreys. In Lampreys: Biology, Conservation, and Control, Docker MF (ed), vol. **1**. : Springer: Fish and Fisheries Series, New York; 75–137.

Foulds WL, Lucas MC. 2013. Extreme inefficiency of two conventional, technical fishways used by European river lamprey (*Lampetra fluviatilis*). *Ecological Engineering* **58**: 423–433.

Gowans ARD, Armstrong JD, Priede IG. 1999. Movements of adult Atlantic salmon in relation to a hydroelectric dam and a fish ladder. *Journal of Fish Biology* **54**: 713–726.

Hardisty MW. 1986. *Petromyzon marinus* (Linnaeus, 1758). In The Freshwater Fishes of Europe, Holčík J (ed), vol. **1**, Part 1, *Petromyzontiformes*. AULA: Wiesbaden; 94–116.

Haro A, Kynard B. 1997. Video evaluation of passage efficiency of American shad and sea lamprey in a modified Ice Harbour fishway. *North American Journal of Fisheries Management* **17**: 981–987.

Hogg R, Coghlan SM Jr, Joseph ZJ. 2013. Anadromous sea lampreys recolonize a Maine coastal river tributary after dam removal. *Transactions of the American Fisheries Society* **142**: 1381–1394.

Johnson JB, Omland KS. 2004. Model selection in ecology and evolution. *Trends in Ecology and Evolution* **19**: 101–108.

Johnson EL, Clabough TS, Keefer ML, Caudill CC, Peery CA, Moser ML. 2009. Effects of lowered nighttime velocities on fishway entrance success by Pacific lamprey at Bonneville Dam and fishway use summaries for lamprey at Bonneville and the Dalles dams, 2008. Technical Report 2009-10 of Idaho Cooperative Fish and Wildlife Research Unit to U.S. Army Corps of Engineers, Portland District.

Kaseloo PA, Weatherley AH, Lotimer J, Farina MD. 1992. A biotelemetry system recording fish activity. *Journal of Fish Biology* **40**: 165–179.

Keefer ML, Caudill CC, Johnson EL, Boggs CT, Ho B, Moser ML. 2010. Adult Pacific lamprey migration in the lower Columbia River: 2009 radiotelemetry and half-duplex PIT tag studies. Technical Report 2010-3 of Idaho Cooperative Fish and Wildlife Research Unit to U.S. Army Corps of Engineers, Portland District.

Keefer ML, Peery CA, Lee SR, Daigle WR, Johnson EL. 2011. Behaviour of adult Pacific lamprey in near-field flow and fishway design experiments. *Fisheries Management and Ecology* **18**: 177–189.

Kemp PS, Russon IJ, Vowles AS, Lucas MC. 2011. The influence of discharge and temperature on the ability of upstream migrant adult river lamprey (*Lampetra fluviatilis*) to pass experimental overshot and undershot weirs. *River Research and Applications* **27**: 488–498.

Laine A, Kamula R, Hooli J. 1998. Fish and lamprey passage in a combined Denil and vertical slot fishway. *Fisheries Management and Ecology* **5**: 31–44.

Larinier M, Marmulla G. 2004. Fish passes: types, principles and geographical distribution: an overview. In Proceedings of the Second International Symposium on the Management of Large Rivers for Fisheries, Welcomme R, Petr T (eds), vol. **II**, RAP Publication. FAO Regional Office for Asia and the Pacific: Bangkok, Thailand; 17.

Lasne E, Sabatie MR, Tremblay J, Beaulaton L, Roussel JM. 2010. A new sampling technique for larval lamprey population assessment in small river catchments. *Fisheries Research* **106**: 22–26.

Maitland PS, Renaud CB, Quintella BR, Close DA, Docker MF. 2015. Conservation of native lampreys. In Lampreys: Biology, Conservation, and Control, Docker MF (ed), vol. **1**. Springer: Fish and Fisheries Series, New York; 375–428.

Mateus CS, Rodríguez-Munoz R, Quintella BR, Alves MJ, Almeida PR. 2012. Lampreys of the Iberian Peninsula: distribution, population status and conservation. *Endangered Species Research* **16**: 183–198.

McLaughlin RL, Hallett A, Pratt TC, O'Connor LM, McDonald DG. 2007. Research to guide use of barriers, traps, and fishways to control sea lamprey. *Journal of Great Lakes Research* **33**(Special Issue 2): 7–19.

Moser ML, Matter AL, Stuehrenberg LC, Bjornn TC. 2002. Use of an extensive radio receiver network to document Pacific lamprey (*Lampetra tridentata*) entrance efficiency at fishways in the Lower Columbia River, USA. *Hydrobiologia* **483**: 45–53.

Moser ML, Butzerin JM, Dey DB. 2007. Capture and collection of lampreys: the state of the science. *Reviews in Fish Biology and Fisheries* **17**: 45–56.

Moser ML, Keefer ML, Pennington HT, Ogden DA, Simonson JE. 2011. Development of Pacific lamprey fishways at a hydropower dam. *Fisheries Management and Ecology* **18**(3): 190–200.

Moser ML, Almeida PR, Kemp PS, Sorensen PW. 2015. Lamprey spawning migration. In Lampreys: Biology, Conservation, and Control, Docker MF (ed), vol. **1**. Springer: Fish and Fisheries Series, New York; 215–263.

Naughton G, Caudill C, Peery C, Clabough T, Jepson M, Bjornn T, Stuehrenberg L. 2007. Experimental evaluation of fishway modifications on the passage behaviour of adult Chinook salmon and steelhead at Lower Granite Dam, Snake River, USA. *River and Research Applications* **23**: 99–111.

Noonan MJJ, Grant JWA, Christopher DJ. 2012. A quantitative assessment of fish passage efficiency. *Fish and Fisheries* **13**: 450–464.

Quintella BR, Andrade NO, Almeida PR. 2003. Distribution, larval stage duration and growth of the sea lamprey ammocoetes, *Petromyzon marinus* L., in a highly modified river basin. *Ecology of Freshwater Fish* **12**: 286–293.

Quintella BR, Andrade NO, Koed A, Almeida PR. 2004. Behavioural patterns of sea lamprey' spawning migration through difficult passage areas, studied by electromyogram telemetry. *Journal of Fish Biology* **65**: 961–972.

Quintella BR, Póvoa I, Almeida PR. 2009. Swimming behaviour of upriver migrating sea lamprey assessed by electromyogram telemetry. *Journal of Applied Ichthyology* **25**: 46–54.

Reinhardt UG, Eidietis L, Friedl SE, Moser ML. 2008. Pacific lamprey climbing behavior. *Canadian Journal of Zoology* **86**: 1264–1272.

Roscoe DW, Hinch SG, Cooke SJ, Patterson DA. 2010. Fishway passage and post-passage mortality of up-river migrating sockeye salmon in the Seton River, British Columbia. *River Research and Applications* **27**: 693–705.

Steeves TB, Slade JW, Fodale MF, Cuddy DW, Jones ML. 2003. Effectiveness of using backpack electroshocking gear for collecting sea lamprey (*Petromyzon marinus*) larvae in Great Lakes tributaries. *Journal Great Lakes Research* **29**(Suppl 1): 161–173.

Travade F, Larinier M, Boyer-Bernard S, Dartiguelongue J. 1998. Performance of four fish pass installations recently built in France. In Fish Migration and Fish Bypasses. Fishing News Books, Jungwirth M, Schmitz S, Weiss S (eds). Blackwell Science Ltd. Publisher: Oxford, UK; 146–170.

Venables W, Ripley BD. 2002. Modern Applied Statistics with S, 4th edn. Springer: New York.

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