HUMAN IMPACT, FISHERY MANAGEMENT AND AQUACULTURE

Spedicato, M.T.; Lembo, G.; Marmulla, G. (eds.)
Aquatic telemetry: advances and applications. Proceedings of the Fifth Conference on Fish Telemetry held in Europe. Ustica, Italy, 9-13 June 2003.
Rome, FAO/COISPA. 2005. 295p.

Using electromyogram telemetry to study the spawning migration of sea lamprey (*Petromyzon marinus* L.)

P. R. Almeida*[1, 2], B. R. Quintella[2], A. Koed[3] & N. O. Andrade[2]

[1] *Unidade de Biologia da Conservação, Departamento de Biologia, Universidade de Évora, 7002-554 Évora, Portugal. Tel: + 351 266 760 800, Fax: + 351 266 711 231, *Corresponding Author; e-mail: pmraposo@mail.telepac.pt*
[2] *Instituto de Oceanografia, Faculdade de Ciências da Universidade de Lisboa, Campo Grande, 1749-016 Lisboa,Portugal.*
[3] *Danish Institute for Fisheries Research, Department of Inland Fisheries, Silkeborg, Denmark.*

Key words: EMG telemetry, swimming performance, activity effort, obstacle negotiation, River Mondego, Portugal.

Abstract

Laboratory and field experiments were conducted to assess activity patterns in adult sea lamprey (*Petromyzon marinus* L.) using electromyogram (EMG) telemetry. The good correlation between EMG values and sea lamprey activity levels observed during laboratory trials, allowed us to study *in situ* the behaviour of these animals during their spawning migration. Special attention was given to the sea lamprey activity patterns observed during the upstream migration when negotiating obstacles in specific reaches of the River Mondego, Portugal. High activity levels were usually associated with subsequent periods of low activity or rest periods. When swimming through stretches free from obstacles, the EMG values showed a constant pattern of activity. Intense activity peaks were frequently observed when lamprey negotiated difficult passage areas, less common during negotiation of smaller obstacles, and seldom recorded for stretches free from obstructions.

Introduction

During the course of evolution, anadromous fish populations' evolved genetically determined strategies that maximize fitness, and presumably become adjusted to the natural obstructions that they encounter in their upstream spawning migration (Gross, 1987; Clay, 1995). Usually these species stop feeding prior to migration, so the allocation of energy reserves must guarantee migration and spawning success. Such a delicate balance is often disrupted as a result of the modifications made in watersheds by human actions. Hinch *et al*. (1996) stated that river conditions that delay migration or require great energy expenditure to negotiate, are responsible for a post spawning mortality increase in sockeye salmon *Oncorhynchus nerka* (Walbaum).

Recent works using electromyogram (EMG) telemetry on free-swimming specimens have been successfully applied to different species, e.g., lake trout *Salvelinus namaycush* (Walbaum) (Weatherley *et al*., 1996), largemouth bass *Micropterus salmoides* (Lacépède) and smallmouth bass *M. dolomieui* Lacépède (Demers *et al*., 1996), sokeye salmon (Hinch *et al*., 1996) and Atlantic salmon *Salmo salar* L. (Booth *et al*., 1997; Økland *et al*., 2002). Telemetry techniques have been developed for the detection and transmission of electromyograms produced during muscle activity of free swimming fish (e. g. Ross *et al*., 1981; Rogers *et al*., 1984; Kaseloo *et al*., 1992; Demers *et al*., 1996; Hinch *et al*., 1996; Weatherley *et al*., 1996; Booth *et al*., 1997; Thorstad *et al*., 2000). Electromyograms are bioelectrical voltage changes that are strongly related with strength and duration of muscle contractions (Kaseloo *et al*., 1992)

In previous studies developed in the River Mondego, Almeida *et al*. (2000, 2002) described the migratory behaviour of sea lamprey *Petromyzon marinus* L., and identified the main obstacles and difficult passage reaches present in the migratory route of these animals. In the present study the exercise and activity patterns of sea lamprey during obstacle negotiation and movement through difficult passages areas was investigated using EMG telemetry. The results presented here are to the authors' knowledge the first that document the potential of using the electromyogram telemetry technology in lampreys.

Materials and methods

Five migrating adult sea lampreys (Table 1) were purchased from professional fishermen at the Mondego estuary (Portugal) and tagged with EMG radio transmitters manufactured by Lotek Engineering Inc. Newmarket, Ontario, Canada. Transmitters were cylindrical, weighed 18.3 g in the air, 8.0 g in water, and were 53.0 mm long by 16.2 mm diameter. The two Teflon-coated stainless steel electrodes were hooked and held in position in the swimming muscle using 18 carat gold tips (7 x 1 mm), as described by Hinch *et al.* (1996) and Thorstad *et al.* (2000). The transmitters detected signals over 1-2 μV in amplitude, and a radio pulse was transmitted whenever the 150 μV factory-determined threshold was exceeded (Kaseloo *et al.*, 1992). Consequently, the intervals between pulses correlated with the frequency of muscle contraction. A combined receiver and data logger model SRX-400 (Lotek Engineering Inc.) recorded the pulses as intervals (ms) between successive radio signals transmitted on a frequency (142.076 – 142.357 MHz), unique to each transmitter.

Prior to the implantation of the EMG transmitters, the surgical procedure and the animals' adaptation to the experimental conditions were tested in two lampreys, using dummy transmitters with dimensions similar to the EMG transmitters.

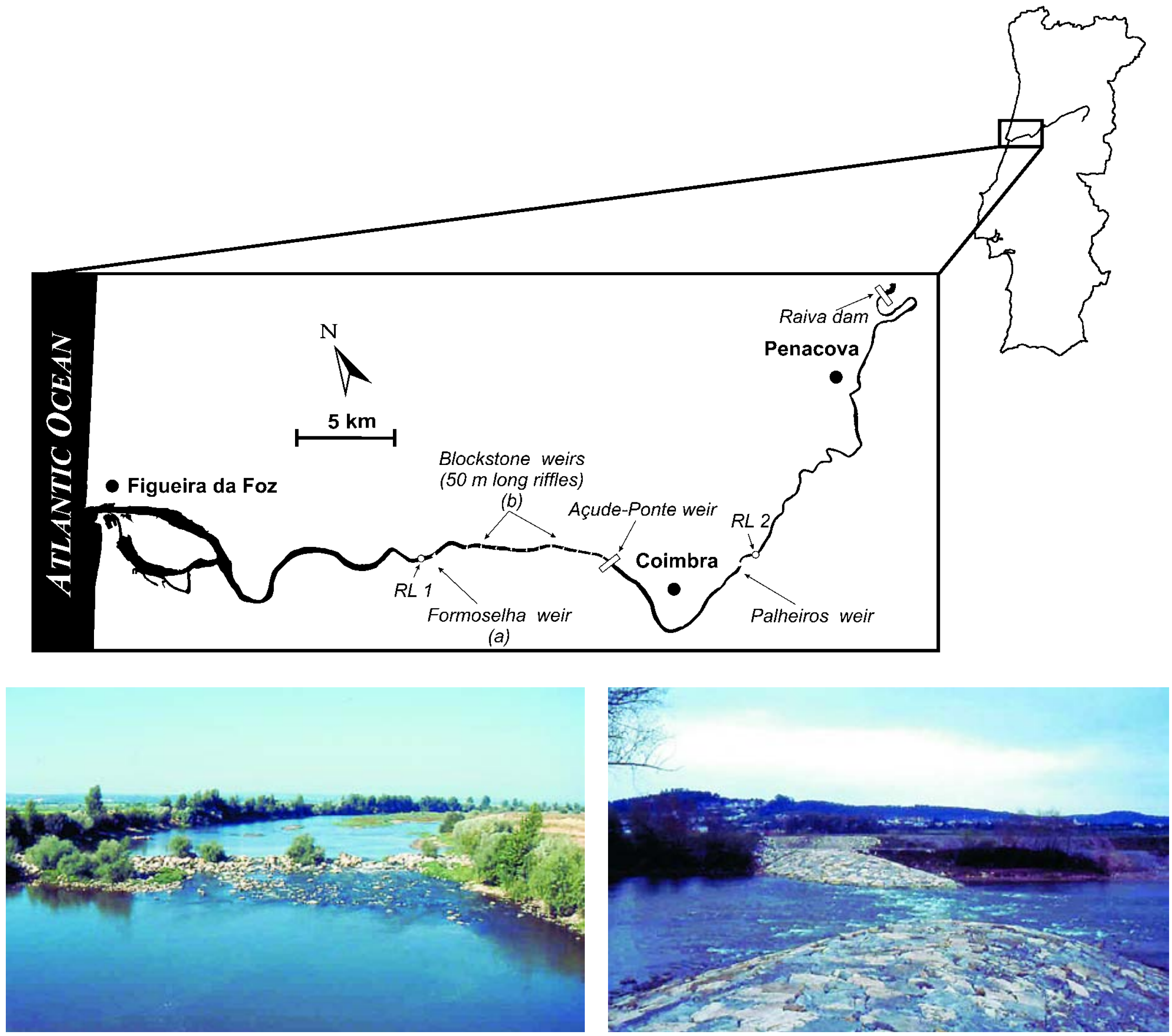
The lampreys were anaesthetised in a 0.5 ml 2-phenoxyethanol *per* liter water and a tag was surgically implanted in the body cavity of each fish. Surgery was initiated when the lamprey released its sucking disk from the container wall and stopped reacting to manipulation. The abdominal region was disinfected with an iodine solution (Betadine®) and a 3 cm incision was made in the midventral line, 8 cm from a point corresponding to the projection of the anterior insertion of the dorsal fin. The whip antenna was run through a hole in the abdominal wall, pierced with a blunt needle, leading the antenna to exit posterior.

The two gold-tipped electrodes (5-10 mm apart) were placed into the red lateral musculature, under the skin at the mid position, via an incision in the ventral region, using plunger devices (McKinley and Power, 1992). The incision was closed with 4-6 independent non-absorbable polyamide monofilament sutures (2/0 Dafilon DS24) and disinfected with Betadine. The complete procedure took 10-12 minutes. All lampreys were successfully revived in a 2-m^3 holding tank, and were left to recover for a period of 8 days under a light cycle of 10 h light: 14 h darkness. They were then carried to the River Mondego and released at two locations, one downstream from the Formoselha weir (RL1), and the other upstream from the Palheiros weir (RL2) (Fig. 1).

Table 1 – General information about the EMGs tracking sessions.

ID	TL (mm)	TW (g)	SD	RD	RL	TTT (h)	TDM U/D (m)	(ºC)
Lp 3	945	1445	03/03/02	18/03/02	*RL1*	78	7014/0	12.4
Lp 4	990	1800	25/03/02	01/04/02	*RL1*	161	5867/0	17.5
Lp 5	990	1900	09/04/02	15/04/02	*RL1*	163	3944/0	15.7
Lp 6	960	1700	23/04/02	06/05/02	*RL2*	158	7926/2892	16.1
Lp 7	930	1431	05/05/02	15/05/02	*RL2*	161	14267/10017	18.8

ID-animal identification, TL-tagged sea lampreys total length (mm), TW-tagged sea lampreys total weight (g), SD-surgery date, RD-release date, RL-release location, TTT-total time tracked, TDM U/D-total distance moved upstream/downstream (m), mean water temperature (ºC).



(a) (b)

Fig. 1 – Map of the River Mondego showing details of the study area. Photographs illustrate a view of the Formoselha weir (a) and of one riffle zone caused by a submerged blockstone weir (b). RL–releasing points.

During the recovery time, we tested the transmitter response to forced lamprey movements. For each animal, an EMG interval was associated with one of four different behaviour categories: i) *resting*, corresponding to periods when the lamprey used the sucking disk to remain motionless on the bottom or on the wall of the holding tank; ii) *low activity*, slow swimming movements typically made in the same direction, avoiding sudden changes of depth; iii) *medium activity*, fast swimming with regular changes of direction; iv) *high activity*, fast swimming with some burst accelerations, attempts to escape from the holding tank raising the head out of the water, swimming with one third of the body out of water and occasional jumps out of water.

Following release of the lampreys, a three-element hand-held (Yagi) antenna was used to track the location of lampreys and to monitor the EMGs. The tagged lampreys were tracked continuously, following the procedure described by Almeida *et al*. (2000; 2002). Data were downloaded to a laptop computer via an RS-232 serial communication port.

Results

The two animals that were tagged with the dummy transmitters survived the experimental procedure and indicated that the EMG transmitters could be successfully implanted into adult sea lamprey. After eight weeks the animals were euthanized and a *post mortem* observation revealed no internal damage and that gonad continued its maturation process.

Laboratory observations of the five EMG tagged lampreys indicated a clear difference between EMGs recorded when all lampreys were resting motionless and attached via the sucking disk, and periods of active swimming (Fig. 2). Resting periods corresponded to a steady high EMG pulse interval, and movement intensity lowered EMG pulse intervals. Pulse intervals during activity were highly dependent on the swimming speed, giving an accurate report of the animals' behaviour (Fig. 3). Laboratory records showed that lamprey were particularly active during the dark hours (Fig. 2).

The three animals that were released downstream from the Formoselha weir spent 21 hours, 27 hours and 93 hours passing this obstacle (Table 1). Two of them halted their upstream migration halfway between the Formoselha weir and the Açude-Ponte dam (Fig. 1). The lamprey that took longer to negotiate the Formoselha weir moved 3.4 km upstream in the next two nights, died and was recovered 1.3 km downstream. A closer look at the incision of the deceased specimen revealed that it was open, probably as a result of the intense exercise made by the animal while passing the Formoselha weir, although the electrodes maintained the position following surgery.

The two lampreys that were released upstream from the Palheiros weir (Fig. 1) presented two different types of behaviour. One moved 7.9 km upstream during the first night, and on the second night moved 2.9 km downstream and halted near the confluence of a small tributary for the rest of the tracking period (Table 1). The other animal made a short downstream movement just after being released, and passed over the Palheiros weir. During the first night the lamprey continued to move downstream for another 10 km, ending in the reservoir of the Açude-Ponte dam. On the next night the lamprey moved upstream and reached the Palheiros weir, where it stopped for the next two nights. Finally, on the fourth night the lamprey passed the weir and halted after moving 5 km upstream. No other movement was observed for the rest of the tracking period.

All five lampreys showed a preference for moving during the night. They spent 93% of the time resting during the day and only 44% of the time resting during the night. The cruising ground speed (*GS*) attained by the three lampreys released downstream from the Formoselha weir on the stretches free from obstacles was 11.1 BL (Body Length) min^{1}. For an animal with a total length of 85 cm, this value represents a GS of 0.6 km h^{-1}. The two sea lampreys released upstream the Açude-Ponte dam in the upper reaches of River Mondego attained a cruising GS of 23.9 BL min^{1}. A lamprey with 85 cm of total length would attain a GS of 1.2 km h^{-1}.

There were small variations in the EMG pulse intervals among individual lampreys with the resting EMG records, which corresponded to a steady high EMG baseline, with different values for each tagged animal. When comparing the percentage of time spent by the lampreys at each activity level throughout the different river habitats it was noticed that the lampreys showed similar behaviours when passing through river stretches morphologically identical. The lamprey *Lp 4* demonstrated a considerably more energetic swimming than the rest of the released animals, with a higher percentage of time corresponding to a more active behaviour (Table 2).

The EMGs records showed high activity when the lampreys were negotiating the Formoselha weir. This indicates that this obstacle is difficult to pass and demands intense exercise for a substantial period of time (Fig. 4). On average 28 percent of the total time was spent to passing the weir.

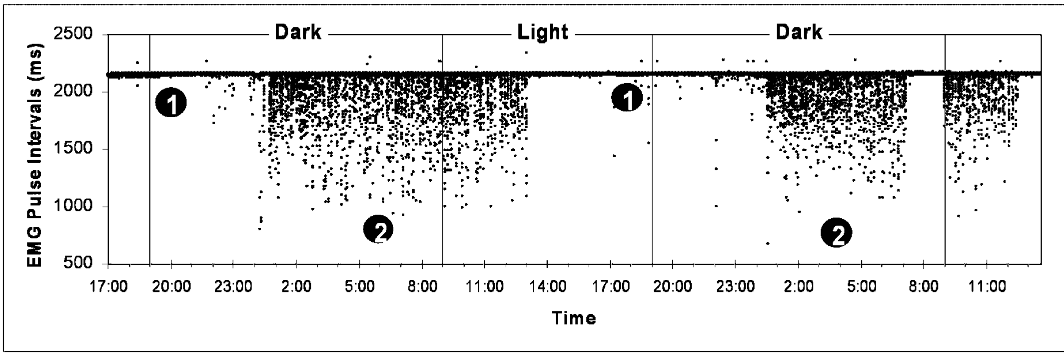


Fig. 2 – Records of radio transmitted EMG pulse intervals from axial swimming muscle of a representative sea lamprey (*Lp 7*) during laboratory trials. 1-Resting motionless; 2–Active swimming.

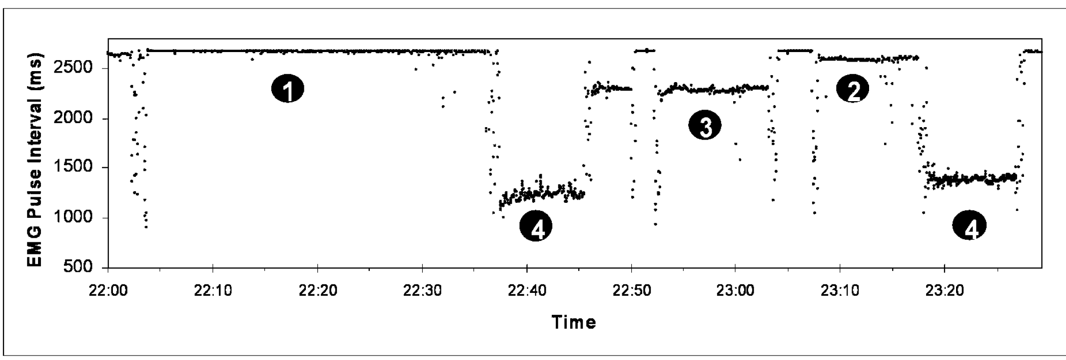


Fig. 3 – Records of radio transmitted EMG pulse intervals from axial swimming muscle of a representative sea lamprey (*Lp 6*) during laboratory trials. Activity levels: 1–Resting; 2–Low; 3–Medium; 4–High.

Table 2 – Percent of time spent by tagged lampreys at each activity level in the distinct river habitats.

River habitat		Activity level	*Lp 3*	*Lp 4*	*Lp 5*	*Lp 6*	*Lp 7*
Free stretches	% time spent each activity level	*Rest*	4.6	9.9	2.9	14.0	17.5
		L_act	5.8	28.0	42.7	5.5	8.3
		M_act	86.5	41.3	52.1	72.2	72.0
		H_act	3.1	20.9	2.3	8.2	2.1
Blockstone weir		*Rest*	7.7	0.8	1.0	—	—
		L_act	29.3	31.8	31.3	—	—
		M_act	49.2	40.3	60.3	—	—
		H_act	13.8	27.1	7.4	—	—
Formoselha weir		*Rest*	12.6	9.6	5.9	—	—
		L_act	18.4	9.5	26.5	—	—
		M_act	53.5	33.6	46.4	—	—
		H_act	15.5	47.2	21.2	—	—
Palheiros weir		*Rest*	—	—	—	29.3	—
		L_act	—	—	—	29.3	—
		M_act	—	—	—	33.3	—
		H_act	—	—	—	8.0	—

Rest – resting, L_act – low activity, M_act – medium activity, H_act – high activity.

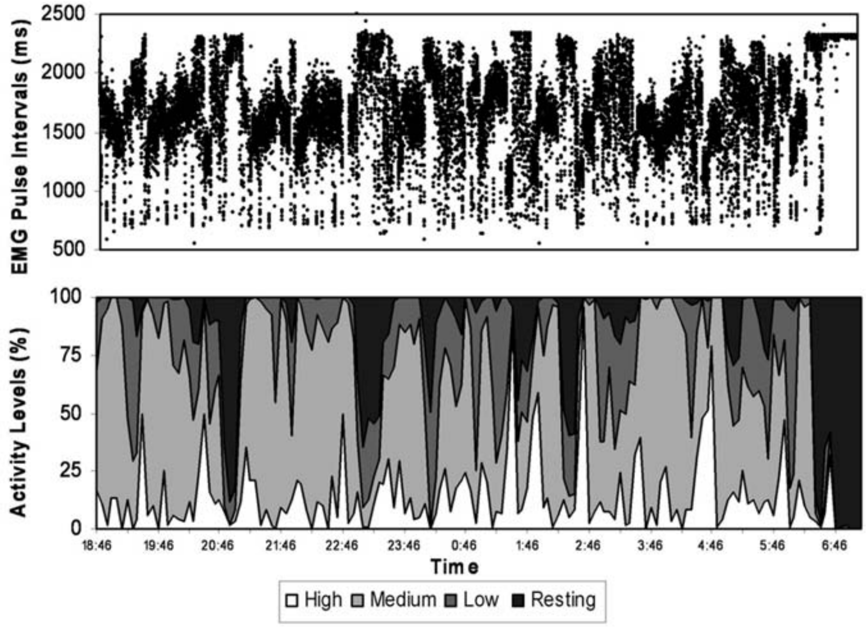


Fig. 4 – EMG records and proportion of time spent in each of the behaviour categories corresponding to sea lamprey *Lp3* during the Formoselha weir negotiation.

Submerged blockstone weirs created riffle zones where the lamprey changed their behaviour. The uniform pattern of EMGs observed in river stretches that separated the riffle zones changed to a sequence that shows peaks of high intensity movements, usually associated with low activity and resting moments (Fig. 5).

Movements through river stretches free from obstacles, like the river section upstream from the Palheiros weir (Fig. 1), exhibited a regular pattern of EMGs pulses, occasionally interrupted by high activity episodes and resting moments (Fig. 6). These probably resulted from changes in the morphology of the river bed.

Discussion

Lampreys tagged with EMG transmitters exhibited the same circadian activity patterns as lamprey bearing smaller radio or acoustic transmitters (Almeida *et al*., 2000; 2002). EMG transmitters registered efficiently all the animals' movements, even slight oscillations of the body when sea lampreys were attached to the wall, or to the bottom of the holding tank. During the laboratory trials it was observed that the EMG pulse rate varied depending on the position adopted by the lamprey. For instance, if lampreys were resting on the wall of the tank, the EMGs values were lower than when lampreys were resting in the bottom.

Constant swimming in the river stretches free from obstacles also gave indications of variations in EMGs. Because the electrodes were located in the left axial swimming muscle, it is possible that contractions of the right side muscle were not registered, which would produce a high EMG pulse interval. Økland *et al*. (2002) described a similar situation in an EMG telemetry study conducted with Atlantic salmon.

Lampreys take advantage of their sucking disk to rest motionless in areas of the riverbed where the current is high. This behaviour explains the resting moments that usually were associated with fast swimming periods and burst swimming episodes. This sequence of events was normally observed when the animals were migrating through difficult

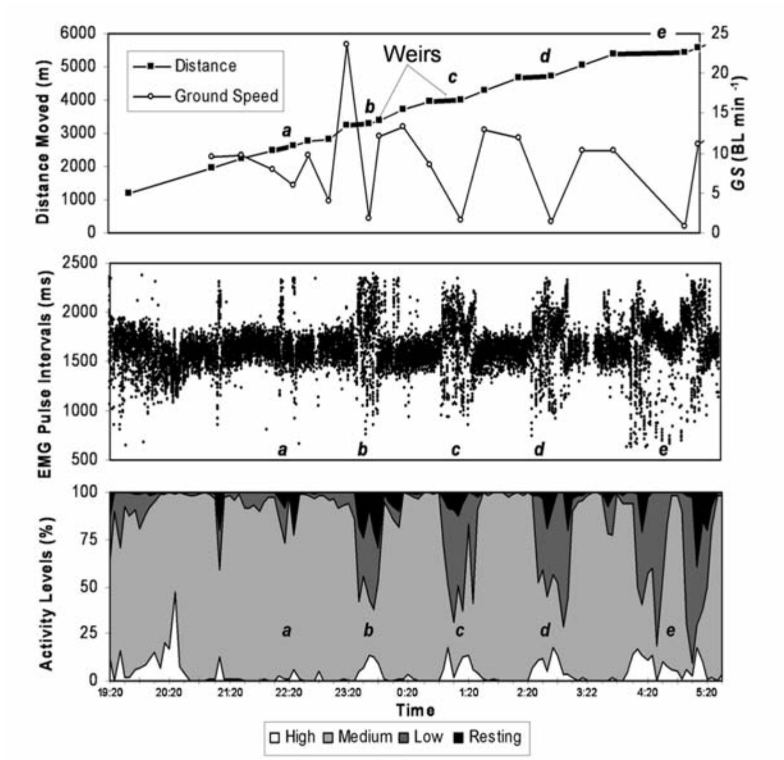


Fig. 5 – Distance moved from the Formoselha weir, ground speed, EMG records and proportion of time spent in each of the behaviour categories corresponding to sea lamprey *Lp3* tracked in the river stretch between the Formoselha weir and the Açude-Ponte dam. Also identified the location of the submerged blockstone weirs (a, b, c, d and e) throughout.

passage zones (*e. g.* riffles), or during negotiation of obstacles (*e. g.* Formoselha weir).

The results demonstrated that the ground speed estimates made in the past using conventional radio and acoustic telemetry gave a rough idea about the behaviour of the animals in difficult passage zones. In fact, Almeida *et al.* (2000; 2002) assumed that sea lampreys were moving continuously through the riffle areas, when the animals probably were alternating between resting moments and burst swimming, as a strategy to overcome these fast current stretches as observed in the present study.

Comparing the results obtained by Almeida *et al.* (2000; 2002) studies, which used externally attached radio or acoustic transmitters, with the cruising GS of sea lampreys tagged with surgically implanted EMG transmitters it was possible to notice that, apparently, lampreys with EMG transmitters migrate more slowly than with external tags. It is possible that internal implantation might affect the swimming performance of the sea lampreys not only because it is a more intrusive technique, which may provoke additional stress, but also due to the necessary recovering period in laboratory after the tagging procedures which may alter to some extent the normal behaviour of the migrating sea lamprey. Mesa *et al.*, (2003) work which evaluated the effect of surgically implanted telemetry transmitters on the swimming performance of adult Pacific lampreys (*Lampetra tridentata*, Richardson) showed significant but minor effects.

Lucas *et al.* (1991) and Demers *et al.* (1996) concluded that using conventional telemetry tracking-based swimming speeds as an index of fish activity could be misleading. This is particularly the case when one wants to estimate the metabolic cost

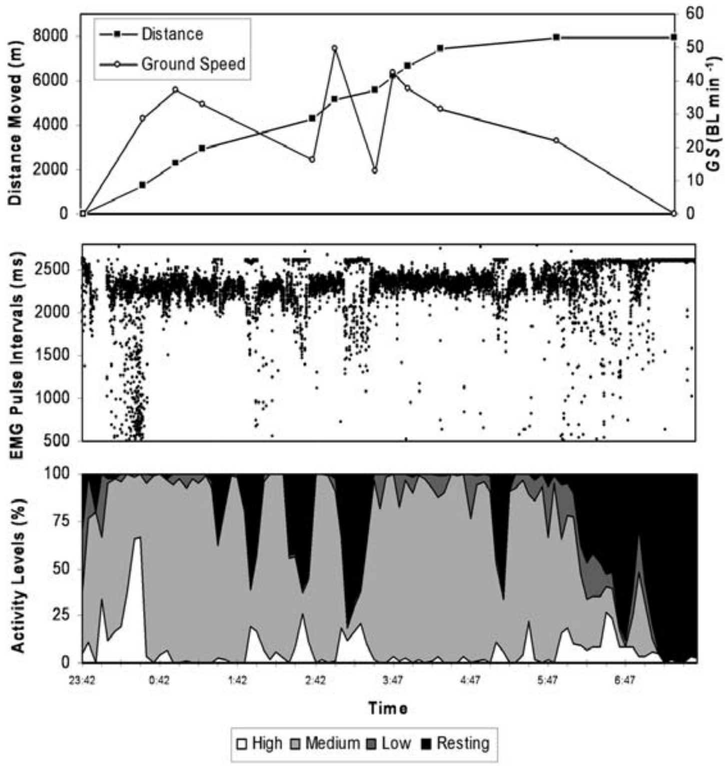


Fig. 6 – Distance moved from the Palheiros weir, ground speed, EMG records and proportion of time spent in each of the behaviour categories corresponding to sea lamprey *Lp6* tracked in the upstream stretch of River Mondego.

associated with an apparent swimming speed. The present study supports this conclusion, because movement through difficult passage areas (*e. g.* riffles zones) produced an increase in EMG pulse rate, although the ground speed decreased through these river stretches.

Sea lampreys recover rapidly from post-exercise metabolic and respiratory acidosis (Wilkie *et al.*, 1998), which means that they are able to continue their migration shortly after an exhaustive exercise episode. However, the energy spent negotiating obstacles and migrating through a sequence of difficult passage zones, like the riffle areas in the River Mondego, may increase the amount of energy allocated to migration and, consequently, limit spawning success. Further investigations are necessary to establish whether the excessive energy costs of migration through anthropogenic or natural obstacles affects the overall spawning success of sea lamprey.

Acknowledgements

This research was financially supported by the Water Institute (INAG) of the Ministry of Environment and by FCT (Fundação para a Ciência e Tecnologia-Project PNAT/1999/BIA/15019/P) and PhD grant to B.R. Quintella with the following reference SFRH/BD/9125/2002.

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