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NOTES ON THE OCCURRENCE AND ECOLOGY OF *SADURIELLA LOSADAI* (ISOPODA: CHAETILIIDAE) IN THE MINHO RIVER ESTUARY (NW IBERIAN PENINSULA)

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ABSTRACT

Ecological aspects, habitat preferences, demographic structure, migration patterns and productivity of the isopod *Saduriella losadai* were analysed from monthly sediment samples and by-catch in glass eel fishery in the Minho River estuary. This species is typically distributed within the mesohaline-oligohaline boundary, so salinity plays a large role in the distribution of this species within the estuary, with sandy sediments having higher densities. High temperatures and saline intrusion have been shown to hinder the population growth. Adult populations were strongly female-biased. Two peaks in juvenile recruitment have been recorded with higher densities occurring in the spring-summer season. Tidal drift during flood tides is used as a mean of migration. Juvenile migrations to freshwater environments occur in late spring and summer seasons and are dependent on salinity increases. Juvenile migrations to the lower estuary occur in the autumn and mature adults migrate back in the winter before the reproductive season. There was a sharp decline in abundance compared with data collected 40 years ago for the same study area, but further studies are needed to confirm the trend.

Keywords. Isopoda; *Saduriella losadai*; estuaries; environmental factors; migration.

RESUMEN

Notas sobre la presencia y ecología de *Saduriella losadai* (Isopoda: Chaetiliidae) en el estuario del Río Miño (NO de la península Ibérica)

Se han analizado los aspectos ecológicos, la preferencia de hábitats, la estructura demográfica, los patrones de migración y productividad del isópodo *Saduriella losadai* en el estuario del Río Miño mediante muestreos mensuales de sedimentos y capturas incidentales en la pesca de angula de cristal. Esta especie está típicamente distribuida entre los límites mesohalinos y oligohalinos, por lo que la salinidad es un factor clave en su distribución en el estuario, siendo sus densidades mayores en sedimentos arenosos. Las altas temperaturas y la intrusión salina se han identificado como factores que limitan su crecimiento poblacional. Los machos componen una pequeña fracción de la población adulta. Se han registrado dos picos de reclutamiento de juveniles, con las mayores densidades en primavera y verano. La deriva mareal en mareas de inundación se utiliza como medio de migración. Las migraciones de juveniles a ambientes de agua dulce ocurren durante el final de la primavera y el verano, y están relacionadas con aumentos en la salinidad. Las migraciones de juveniles hacia el estuario inferior se producen en el otoño, mientras que las migraciones de adultos ocurren en invierno antes de la época de reproducción. Se ha registrado una disminución en la abundancia comparada con datos recopilados hace 40 años en la misma área de estudio, por lo que son necesarios más estudios para confirmar estas tendencias.

Palabras clave. Isopoda; *Saduriella losadai*; estuarios; factores ambientales; migración.

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Introduction

The isopod crustacean *Saduriella losadai* is a species endemic to the Iberian Peninsula, described by Holthuis (1964). It is a monotypic genus belonging to a mostly marine family, with very few species found in brackish environments (Bowman, 1955; King & Cawood, 2007). Since its description, records have been scarce. These records indicate that the distribution of this species is limited to estuaries located between the Rias Baixas (SW Galicia, Spain) and the Gulf of Cadiz (SW Andalusia, Spain). Its northernmost record is in the Ulla River (type locality) in south-western Galicia, Spain (Holthuis, 1964), followed by records at the estuary of the Minho River (Mazé *et al.*, 1993) on the Portuguese-Spanish north-western border, in the Portuguese estuaries of the Ria de Aveiro (Cunha *et al.*, 1999), Mondego River (Chainho *et al.*, 2006), Tagus River (Rodrigues *et al.*, 2006) and Guadiana River (Sánchez-Moyano & García-Asencio, 2011), and a southernmost record at the estuary of the Spanish Guadalquivir River (Baldó *et al.*, 2000). Apart from these records, very little is known about this isopod, except that it is an important prey of the fishes *Solea solea* (Linnaeus, 1758) and *Anguilla anguilla* (Linnaeus, 1758) (Costa *et al.*, 1992; Cabral, 2000). The few studies that have been carried out also indicate that its distribution is restricted to mesohaline-oligohaline boundaries (Mazé *et al.*, 1993; Cunha *et al.*, 1999; Chainho *et al.*, 2006). This study aims to describe the ecological aspects related to this species, focusing on habitat preference, demographic structure, migration patterns and biomass productivity for the Minho River estuary.

Material and methods

Study area and sampling procedures

The estuary of the Minho River, located on the north-western border between Portugal and Spain, is a mesotidal partially mixed system that tends to form a salt wedge during high tides (Sousa *et al.*, 2005). Samples were collected at the lower estuary in a polyhaline section, at the middle estuary in an oligohaline section and upstream in a tidal freshwater area (Fig. 1). Sampling was carried out in both subtidal and intertidal sediments. The intertidal sediment was further subdivided into contiguous areas consisting of sand, muddy sand and sediment associated with macrophytes. Subtidal and intertidal sediment sampling was carried out monthly from September 2021 to August 2022, at low tide on full moons. Five replicates of each intertidal sediment type were collected by delimiting quadrats with an area of 0.1 m^2. Five replicates of subtidal sediment were collected

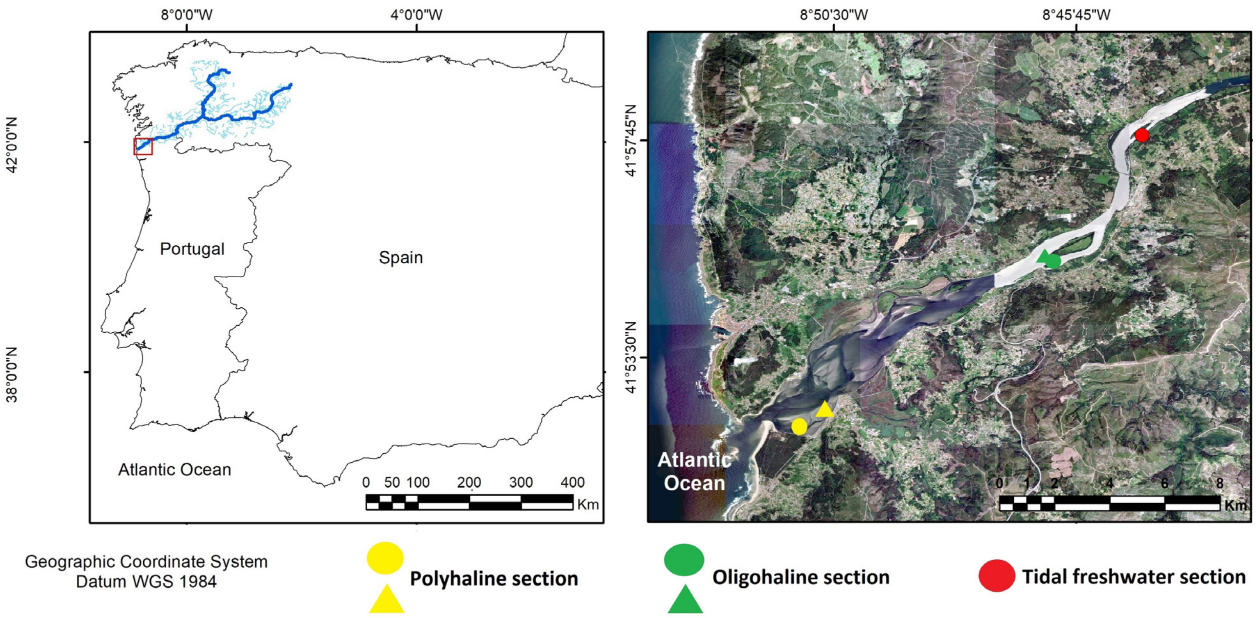

Fig. 1. Study area (circle represents sediment sampling area; triangle represents glass eel by-catch sampling area).

Fig. 1. Área de estudio (círculos representan los puntos de muestreo de sedimentos; triángulos representan los puntos muestreo de captura de anguila de cristal).

using a Van Veen grab sampler with an attack area of 500 cm^2. Sampling depths ranged from 3.3 to 3.9 m. The sediment was then sieved through a 500 µm mesh. An additional sample of each sediment type was collected for texture characterisation and quantification of organic matter content. Water samples used for suspended organic matter and chlorophyll a quantification were also collected at each site, from the margins adjacent to the intertidal sampling area and from the bottom of the river using a Nansen bottle.

Samples from the glass eel fishery by-catch were used to assess movement in the water column during high flood tides. Sampling was carried out monthly during the 2021–2022 fishing season (November to February), at two sites (polyhaline and oligohaline environments) (Fig. 1). Fishing was carried out by professional fishermen on new moon nights, during the rising tide. Fishing gear consisted of a stow net 8 m high, 20 m long float lines and 15 m long bottom-anchored lead lines (covering an area of 50 m^2), with a mesh size of 1–2 mm (Weber, 1986) and was set for three hours.

Laboratory procedures and data analysis

Collected individuals were classified into four categories, juveniles (without distinctive sexual characteristics), males (with genital apophyses), females (with oostegites) and ovigerous females (females carrying embryos). Individuals were then counted and the total body length measured from the anterior margin of the head to the tip of the telson. Specimens were dried for 48 hours at 60°C. Ash free dry mass (AFDM) was determined by burning the dried material in a muffle furnace at 550°C for 4 hours. One litre of sampled water was filtered through a 47 mm diameter GF/C filter. Dry filters and sediment were incinerated in a muffle at 550°C for 4 hours to quantify organic matter. Differences between female and male sizes were assessed through a Kruskal-Wallis analysis. An analysis of variance (ANOVA) was used to detect significant differences in abundance between sites. A two way permutational multivariate analyses of variance (PERMANOVA) was used to test the effects of the temporal scale (in months) and of the sediment characteristics on the abundance and biomass of the population. Correlations between environmental parameters and *Saduriella losadai* abundance were determined using the non-parametric Kendall's rank correlation coefficient. Productivity was calculated using the Morin-Bourassa method (Morin & Bourassa, 1992). Analyses were performed using Past ver. 4.10 software (Hammer *et al.*, 2001).

Results

Field observations indicate that this species is an efficient burrower. It burrows in the surface sediment layer with its head exposed, where it ambushes other crustaceans. When it leaves the sediment it is capable of swimming for short distances. Females with developed oostegites were observed when they reached 1 cm in total length, while males with developed apophyses were observed when they reached 0.9 cm. Females are significantly larger than males (Kruskal-Wallis, $p < 0.01$), each with a mean length of 1.16 ± 0.14 and 1.01 ± 0.09 cm (mean ± SD) respectively. Females preserved in ethanol tend to be darker in colour than males. The abundance of *Saduriella losadai* differs between the three sampled sites with no individuals being recorded in polyhaline benthic environments (Appendix 1). Very few individuals were present in freshwater environments (only small juveniles – 3 to 5 mm) and were only recorded from May to August (Appendix 1). The majority of the population was found in oligohaline environments (Appendix 1). In this section, *Saduriella losadai* was most abundant on subtidal and sandy intertidal sediment (Fig. 2) with the highest peak of abundance occurring in May (258 ± 152 and 214 ± 92 individuals/m^2 respectively). Abundances were significantly higher in subtidal sediments than in muddy intertidal sediments (ANOVA, $p = 0.002$) and in sandy intertidal sediments compared to muddy sediments (ANOVA, $p = 0.0002$). Peaks in biomass were detected from November to February (Fig. 3), coinciding with the highest abundance of adult individuals (Figs 4, 6). Smaller juveniles (2–3 mm) were observed from September to December and February to July (Fig. 6). Females were present from October to June, while ovigerous females were observed in December and February (Figs 4, 6). Furthermore, males were only present from December to April (Figs 4, 6). Juveniles were the most dominant demographic group in most months (85% of the total number of individuals collected), while females were the most dominant in terms of biomass contribution (65%). The spring-summer population accounted for 83% of the annual density, while the autumn-winter population accounted for 75% of the annual biomass. The abundance of *S. losadai* was found to be negatively correlated with pH and salinity (Table 1). In oligohaline environments, the abundance of *S. losadai* was found to be positively correlated with coarse sand, dissolved oxygen and oxidation-reduction potential and negatively correlated with silt, temperature, salinity and deposited organic matter (Table 1). In freshwater environments, abundance was found to be positively correlated with temperature and salinity (Table 1). The two way PERMANOVA analysis indicated that temporal scale had a higher importance on the variation of the abundance and biomass (Table 2). Data collected from the contents of glass eel by-catches

Table 1. Correlations between environmental variables with *Saduriella losadai* density (*p ≤ 0.05, **p ≤ 0.01).

Tabla 1. Correlaciones entre variables ambientales con densidades de *Saduriella losadai* (*p ≤ 0.05, **p ≤ 0.01).

Site	Environmental parameter	Kendal tau
Saduriella losadai abundance (all sites)	pH	-0.40**
	Salinity	-0.31**
Saduriella losadai abundance (Oligohaline section)	ORP	0.30**
	Salinity	-0.30*
	Temperature	-0.25*
	HDO	0.20*
	Deposited organic matter	-0.22*
	Coarse sand	0.22*
	Very fine sand	-0.23*
	Silt	-0.27**
Saduriella losadai abundance (Tidal freshwater section)	Salinity	0.26*
	Temperature	0.34**

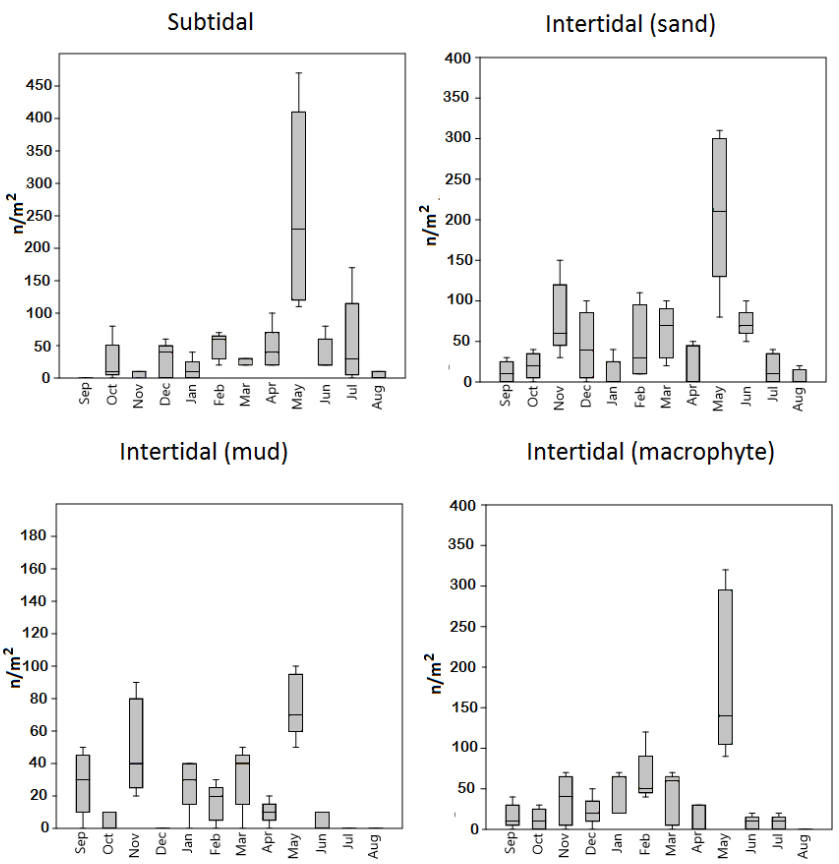

Fig. 2. Monthly *Saduriella losadai* abundances in oligohaline environments in different habitats.

Fig. 2. Abundancias mensuales de *Saduriella losadai* en ambientes oligohalinos en diferentes hábitats.

Table 2. Two-way PERMANOVA analysis (permutations = 9999) estimation the effects of the temporal scale (months) and sediment characteristics interaction on the abundance and biomass of the population.

Tabla 2. Análisis PERMANOVA de dos vías (permutaciones = 9999) sobre los efectos de la interacción de la escala temporal (meses) y las características del sedimento sobre la abundancia y biomasa de la población.

	Effect	R^2	df	F	p
Abundance	Month	0.21	11	6.78	0.0001
	Sediment	0.02	3	2.45	0.0296
	Interaction	0.22	33	2.41	0.0001
Biomass	Month	0.21	11	6.29	0.0001
	Sediment	0.02	3	1.92	0.0538
	Interaction	0,18	33	1.72	0.0006

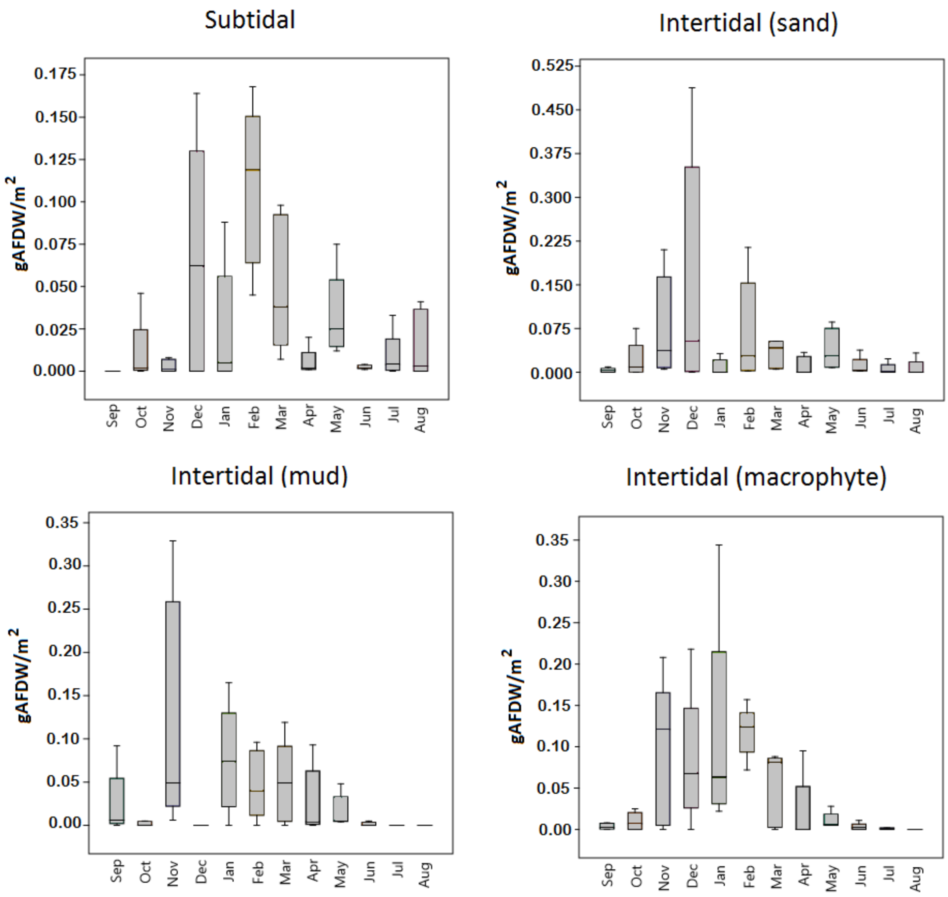

Fig. 3. Montlhy *Saduriella losadai* biomasses in oligohaline environments in different habitats.

Fig. 3. Biomasas mensuales de *Saduriella losadai* en ambientes oligohalinos en diferentes hábitats.

indicate that juveniles dominate the migrating population during the month of November on both sites (Fig. 5). In the following months females predominate in oligohaline environments, while in polyhaline environments juveniles were most frequently sampled (Fig. 5). Males are only recorded in February at both sites, with males accounting for approximately a third of the migrating population in the polyhaline section (Fig. 5). Annual productivity at the oligohaline benthic section is 0.26 gAFDM/m^2 in Ash Free Dry Weight and 0.42 g/m^2 in dry weight, while the P/B ratios were 7.35 and 6.02 respectively.

Discussion

The results obtained seem to confirm that the distribution of this species in benthic substrates is restricted to the mesohaline-oligohaline boundary and is mainly limited by salinity levels (Weber, 1985; Mazé *et al.*, 1993; Cunha *et al.*, 1999; Chainho *et al.*, 2006). The decline in abundance recorded during the late summer months may be due to the intermittent heat waves observed during this period. This is supported by the observed negative responses to increased temperatures and salinity intrusion, suggesting that this species is highly sensitive to environmental changes. Females appear to be larger than males, although the low number of collected males may have hindered the observed results. The abundance of *Saduriella losadai* is closely correlated with coarser sediments, with fewer individuals collected on intertidal muddy sands, rich in organic matter. Two peaks in the abundance of small juveniles were observed, one in the autumn/early winter and another in spring/early summer, suggesting that these periods are the main reproductive seasons. This pattern suggests that optimum reproductive conditions

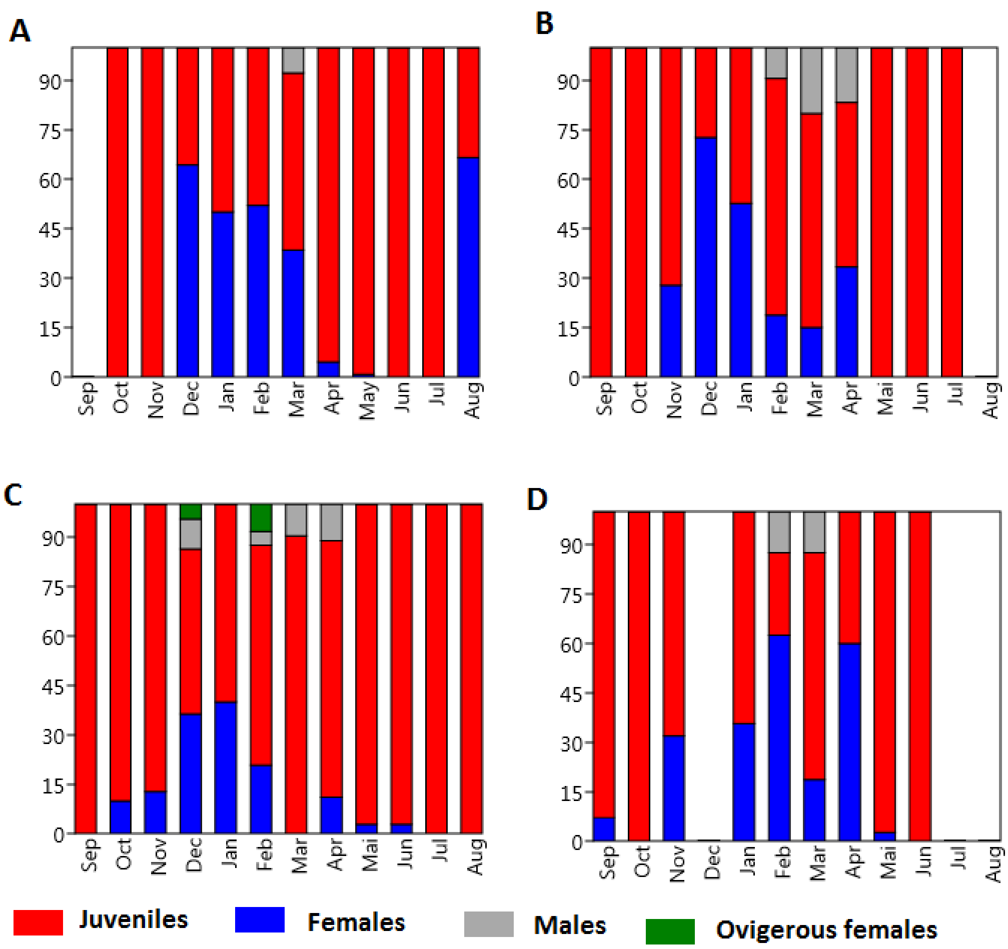

Fig. 4. Monthly *Saduriella losadai* class proportion in oligohaline environments. **A**. Subtidal. **B**. Intertidal sediment with *Juncus* spp. **C**. Intertidal sandy sediments. **D**. Intertidal muddy sediments.

Fig. 4. Proporción de clases mensuales de *Saduriella losadai* en ambientes oligohalinos. **A**. Submareal. **B**. Sedimentos intermareales con *Juncus* spp. **C**. Sedimentos intermareales arenosos. **D**. Sedimentos intermareales fangosos.

occur under moderate temperature and salinity conditions, favouring spring populations. A low representation of ovigerous females was also found in all sampled sites indicating that it must occur in mesohaline environments. Typical patterns of recruitment and subsequent growth (Kuris, 1991) were not observed. Therefore temporal changes in population structure appear to be strongly related to migration. Juveniles spawned in the spring-summer season migrate upstream during the summer months, due to the increase in salinity, whereas juveniles spawned in the autumn-winter migrate downstream where they mature. Adults migrating upstream constitute the majority of migrations in the middle estuary. The larger aperture of the glass eel fishing net may have also contributed to a lower percentage of drifting juveniles. Males appear to have a shorter life span as they only appear during a small window of time (December to July). In oligohaline benthic environments, males make up a small proportion of the population (2% of the total number of individuals collected). However, in February, a third of the population migrating from the lower estuary are males, suggesting that male development tends to occur downstream. Production and P/B ratios usually increase with higher water temperatures and higher percentages of smaller individuals (Morin & Bourassa, 1992). However water temperature rise coupled with decreased river flow was found to decrease benthic invertebrate biomass in assemblages from this estuary (Martins *et al.*, 2019). The intermittent heat waves observed during this period coupled with low river flow most likely contributed to the decrease of the abundance and biomass of *S. losadai* suggesting that P/B ratios were mostly influenced by the high percentage of juveniles. Mean annual abundances observed in the oligohaline intertidal sandy sediments are much lower compared with data collected at the same site 40 years earlier (Weber, 1985), with 170.5 individuals/m^2 recorded in 1981 and 50 individuals/m^2 recorded in this study. The causes of this decline are still largely unknown, but this section of the estuary has undergone significant changes following the colonisation by the exotic bivalve *Corbicula fluminea* (O.F.Müller, 1774) in the 1990s (Ilarri *et al.*, 2014). In conclusion, salinity plays a major role in the distribution of this species within the estuary, whereas sandy sediments contribute to higher densities. High temperatures seem to inhibit population growth. Males make up a small proportion of the population. Two peaks in juvenile recruitment have been recorded particularly in the spring and summer. Tidal drift is used as a means of migration. Upstream juvenile migrations, into the limnetic section, occur during the summer season and are dependent on saline intrusion. Juvenile migrations to the lower estuary occur in the autumn and mature adults migrate back in the winter before the reproductive season. The inclusion of sediment sampling in mesohaline environments and continuous sampling of migrating individuals during high and low tide may provide further insight into the dynamics of this species. There has been a sharp decline in abundance compared to data collected 40 years ago, but further annual sampling is needed to confirm the trend.

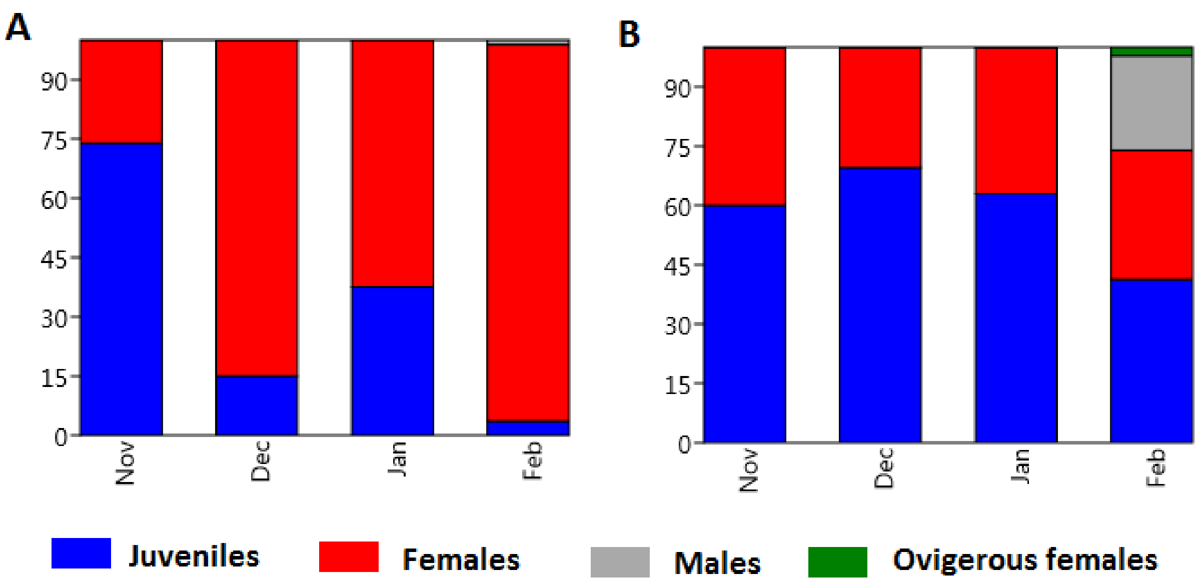

Fig. 5. *Saduriella losadai* drifting population class proportion. **A**. Middle estuary. **B**. Lower estuary.

Fig. 5. Proporción de clases de la población a la deriva de *Saduriella losadai*. **A**. Estuario medio. **B**. Estuario inferior.

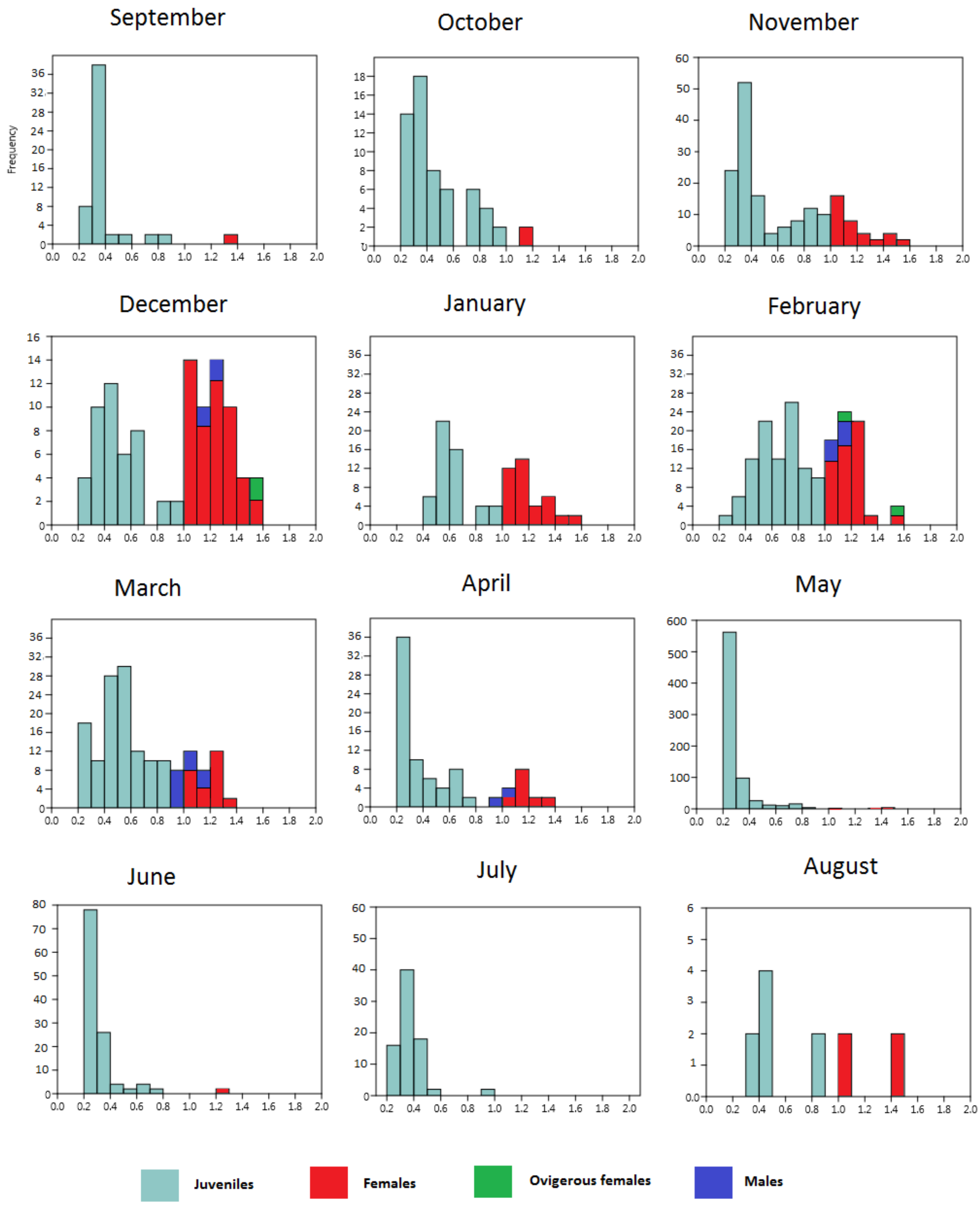

Fig. 6. Monthly *Saduriella losadai* length-frequency (length in cm; frequency in individuals/m²) for oligohaline environments.

Fig. 6. Frecuencia de tallas mensual de *Saduriella losadai* (talla en cm; frecuencia en individuos/m²) en ambientes oligohalinos.

Data availability

The datasets during and/or analysed during the current study available from the corresponding author on reasonable request.

Declaration of competing interest

The authors declare that they have no competing interests.

Authorship contribution statement

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Nuno Gomes. The first draft of the manuscript was written by Nuno Gomes and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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Appendix 1 (continued on next three pages). *Saduriella losadai* abundance and biomass (mean ± SD) per site.

Apéndice 1 (continúa en las tres páginas siguientes). Abundancias y biomasas de *Saduriella losadai* (media ± SD) por localidad de muestreo.

Sample	Month	Mean N (individuals/m^2)	Mean biomass (gAFDM/m^2)
Freshwater subtidal	September	0	0
	October	0	0
	November	0	0
	December	0	0
	January	0	0
	February	0	0
	March	0	0
	April	0	0
	May	0.6 ± 0.5	0.0002 ± 0.0002
	June	0.4 ± 0.5	0.0001 ± 0.0001
	July	0.2 ± 0.4	0.0001 ± 0.0001
	August	0.2 ± 0.4	0.0001 ± 0.0002
Freshwater intertidal sandflats	September	0	0
	October	0	0
	November	0	0
	December	0	0
	January	0	0
	February	0	0
	March	0	0
	April	0	0
	May	0	0
	June	0.2 ± 0.4	0.0001 ± 0.0001
	July	0	0
	August	0	0
Freshwater intertidal mudflats with *Egeria densa*	September	0	0
	October	0	0
	November	0	0
	December	0	0
	January	0	0
	February	0	0
	March	0	0
	April	0	0
	May	0	0
	June	0	0
	July	0	0
	August	0	0

Oligohaline subtidal	September	0	0
	October	2.4 ± 3.2	0.0009 ± 0.0002
	November	0.6 ± 0.5	0.0003 ± 0.0004
	December	2.8 ± 2.7	0.0064 ± 0.0069
	January	1.2 ± 1.6	0.0023 ± 0.0037
	February	5 ± 2	0.0109 ± 0.0047
	March	2.6 ± 0.5	0.0051 ± 0.0040
	April	4.4 ± 3.3	0.0005 ± 0.0008
	May	25.8 ± 15.2	0.0032 ± 0.0025
	June	3.6 ± 2.6	0.0002 ± 0.0001
	July	5.4 ± 6.8	0.0009 ± 0.0014
	August	0.6 ± 0.5	0.0015 ± 0.0019
Oligohaline intertidal sandflats	September	1.2 ± 1.3	0.0003 ± 0.0004
	October	2 ± 1.6	0.0020 ± 0.0031
	November	7.8 ± 4.5	0.0076 ± 0.0087
	December	4.4 ± 4.1	0.0152 ± 0.0207
	January	1 ± 1.7	0.0008 ± 0.0014
	February	4.8 ± 4.5	0.0068 ± 0.0089
	March	6.2 ± 3.2	0.0032 ± 0.0024
	April	1.8 ± 2.5	0.0011 ± 0.0015
	May	21.4 ± 9.2	0.0039 ± 0.0035
	June	7.2 ± 1.8	0.0010 ± 0.0015
	July	1.6 ± 1.8	0.0005 ± 0.0009
	August	0.6 ± 0.9	0.0007 ± 0.0014
Oligohaline intertidal mudflats	September	2.8 ± 1.9	0.0024 ± 0.0039
	October	0.4 ± 0.5	0.0002 ± 0.0002
	November	5 ± 2.9	0.0122 ± 0.0135
	December	0	0
	January	2.8 ± 1.6	0.0075 ± 0.0061
	February	1.6 ± 1.1	0.0047 ± 0.0039
	March	3.2 ± 1.9	0.0048 ± 0.0048
	April	1 ± 0.7	0.0026 ± 0.0039
	May	7.6 ± 1.9	0.0016 ± 0.0019
	June	0.4 ± 0.5	0.0001 ± 0.0002
	July	0	0
	August	0	0

Oligohaline intertidal sandflats with *Juncus* spp	September	1.6 ± 1.5	0.0004 ± 0.0004
	October	1.2 ± 1.3	0.0010 ± 0.0011
	November	3.6 ± 3.0	0.0092 ± 0.0087
	December	2.2 ± 1.8	0.0083 ± 0.0081
	January	3.8 ± 2.5	0.0111 ± 0.0132
	February	6.4 ± 3.2	0.0119 ± 0.0030
	March	4 ± 3.2	0.0052 ± 0.0045
	April	1.2 ± 1.6	0.0021 ± 0.0042
	May	18.8 ± 10.1	0.0053 ± 0.0010
	June	0.8 ± 0.8	0.0003 ± 0.0005
	July	0.8 ± 0.8	0.0001 ± 0.0001
	August	0	0
Polyhaline subtidal	September	0	0
	October	0	0
	November	0	0
	December	0	0
	January	0	0
	February	0	0
	March	0	0
	April	0	0
	May	0	0
	June	0	0
	July	0	0
	August	0	0
Polyhaline intertidal sandflats	September	0	0
	October	0	0
	November	0	0
	December	0	0
	January	0	0
	February	0	0
	March	0	0
	April	0	0
	May	0	0
	June	0	0
	July	0	0
	August	0	0

Polyhaline intertidal mudflats	September	0	0
	October	0	0
	November	0	0
	December	0	0
	January	0	0
	February	0	0
	March	0	0
	April	0	0
	May	0	0
	June	0	0
	July	0	0
	August	0	0
Polyhaline intertidal mudflats with *Zostera noltii*	September	0	0
	October	0	0
	November	0	0
	December	0	0
	January	0	0
	February	0	0
	March	0	0
	April	0	0
	May	0	0
	June	0	0
	July	0	0
	August	0	0