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PRESENCE OF THE EXOTIC WEEVIL *STENOPELMUS RUFINASUS* GYLLENHAL, 1836 (COLEOPTERA: ERIRHINIDAE) IN TER RIVER (NE IBERIAN PENINSULA)

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Abstract: *Stenopelmus rufinasus* Gyllenhal, 1836 has been found in the medium-low stretch of Ter River (NE Iberian Peninsula). This allochthonous species has been related to the presence of the American water fern *Azolla filiculoides* Lamarck. This note contains the localities where this weevil has been detected and its estimated population sizes, in the context of different studies conducted in Ter River between 2008 and 2009.
Key words: *Stenopelmus rufinasus*, *Azolla filiculoides*, exotic species, Iberian Peninsula, Ter River.

Presencia del curculiónido *Stenopelmus rufinasus* Gyllenhal, 1836 en la ribera del río Ter (NE Península Ibérica)
Resumen: Se cita el curculiónido *Stenopelmus rufinasus* Gyllenhal, 1836 en el tramo medio y bajo del río Ter (nordeste de la Península Ibérica). Esta especie alóctona está relacionada con la presencia de *Azolla filiculoides* Lamarck, helecho acuático de origen americano. La siguiente nota da a conocer las nuevas localidades de este gorgojo y una estimación de los tamaños poblacionales encontrados en el marco de varios estudios realizados en el río Ter entre 2008 y 2009.
Palabras clave: *Stenopelmus rufinasus*, *Azolla filiculoides*, especies exóticas, Península Ibérica, río Ter.

Introduction

Are some alien species the way of entrance for other ecologically-related exotic species? Many examples have proved this. For instance, the importation of *Phoenix canariensis* Chabaud from other countries to Iberian Peninsula originated the entrance of the red palm weevil *Rhynchophorus ferrugineus* Olivier, 1790, a wood-boring curculionid from tropical Asia that affects different species of cultivated palms (Barranco *et al.,* 1996). This is also the case of the introduction of the water weevil *Stenopelmus rufinasus* Gyllenhal, 1836, in the Ter River (NE Iberian Peninsula) by means of the water fern *Azolla filiculoides* Lamarck.

Azolla filiculoides is a floating aquatic fern, native from America that has invaded aquatic habitats around the world (Wagner, 1997; García-Murillo *et al.*, 2007). *A. filiculoides* establishes symbiosis with *Anabaena azollae* Strasburger, a blue-green alga (cianobacteria) capable of fixing atmospheric nitrogen, making it more competitive in nitrogen-deficient waters (Wagner, 1997; Pereira *et al.*, 2001). This biological feature has allowed humans to use *A. filiculoides* as a fertilizer in rice fields (Shuying, 1985; Zhuang-Ta *et al.*, 1985) and this has been the main cause of the spread of the water fern out of its natural areas. In spite of this, many other causes such as preventing mosquitoes breeding in stagnant water (Moore, 1969), the direct commerce of *Azolla* (Guillarmod, 1979) and accidental transport through commerce of other aquatic plants, should also be taken into account. Chevalier (1926) stated the presence of *Azolla filiculoides* in natural sites that might have its origin in botanical gardens. Apart from human activities and the dispersion of the fern through water flows, aquatic birds may also constitute a possible dispersal vector (Smith, 1938).

In the Iberian Peninsula, *Azolla filiculoides* is spread all over its western part (García-Murillo *et al.*, 2007). However, in the north eastern part, there are known populations of the water fern in the mouths of Ebre (Conesa & Sanz Elorza, 1998) and Llobregat Rivers (García Murillo *et al.*, 2007). In Ter River *Azolla filiculoides* was found in the middle stretch, in Vilanna (near Bescanó), in 2003 (J. Girbal, *pers. comm.*), without becoming a social concern until 2005 when it spread all along the middle-low stretch (published in local newspapers).

The weevil *Stenopelmus rufinasus*, a 2 mm long beetle originary from south and west USA, bases its diet on species of the genus *Azolla* (Pemberton & Bodle, 2009). It has been imported to South Africa as a biocontroler of *Azolla filiculoides* (Hill, 1999; McConnachie *et al.*, 2003) whereas in Europe *Stenopelmus rufinasus* has been probably introduced through water birds migration (Janson, 1921) and it is associated with the different introduction ways of *Azolla filiculoides*. In the Iberian Peninsula the weevil was found for first time in Guadiana River between May and June 2002 (Fernández Carrillo *et al.*, 2005). One year later, in May 2003, *S. rufinasus* was reported in the National Park of Doñana (Dana & Viva, 2006).

Life history of *S. rufinasus* has been described by Richerson & Grigarick (1967) and Hill (1998). Females make holes in the tip of the fronds of *Azolla* spp. where one egg is laid, and is covered with frass for incubation from 4 to 5 days. Larvae period lasts up to 6 to 9 days, showing a very voracious behaviour during the last days (Hill, 1999). Larvae construct a chamber around themselves using anal secretions in which will become an adult after a 5 to 7 day pupation process. The adult period lasts between 55 and 60 days and

Table I. Date and localities where *Stenopelmus rufinasus* has been found in the Ter River.
The numbers of the localities correspond to the numbers in Fig. 2.

Locality	Municipality	Date	UTM (x, y) (ED50)
1. Pont Major	Girona	04 Apr 2008	31T 485793, 4651626
2. Sobrànigues	Sant Jordi Desvalls	06 May 2008	31T 496748, 4656617
3. Girona	Girona	07 Apr 2009	31T 484528, 4648707
4. Girona	Girona	07 Jun 2009	31T 485407, 4649612

the females produce a mean of 325 offsprings each one. *Stenopelmus rufinasus* is not a truly aquatic insect but adults are prepared for survival in case of accidental immersions. This faculty is possible due to proofing structures that allow the insect to create a ventral air bubble (Thorpe & Crisp, 1949). Larvae, however, do not present this faculty (Baars & Caffery, 2008).

Methods

The Ter River is situated in the NE Iberian Peninsula. It has its headwaters in the Eastern Pyrenees (2400 m altitude) and flows into the Mediterranean Sea after 208 km. This river is characterised by a reservoirs system (Sau-Susqueda-Pasteral) placed in its medium stretch. More than 75% of the total water volume retained (in the period 1996-2007) was derived for human water supply to the urban area of Barcelona (Pou-Rovira *et al*., 2009b). In addition to this, the great number of small reservoirs related to agricultural activity can contribute to drought periods in its medium-low stretch, and the consequent "lentification" of the river (Sabater, 2008), creating favourable conditions for the establishment of *Azolla* populations (Willby *et al*., 2000).

In the framework of a study of *A. filiculoides* management, *Stenopelmus rufinasus* was detected in 2008 in the medium-low stretch of Ter River (between Sau-Susqueda reservoir system and its mouth). In order to quantify its abundance, monthly inspections were carried out between February and June 2009 in 27 localities along the medium-low stretch of the Ter River. In case *A. filiculoides* was found, 6 replicates of 0,04 m^2 were extracted from each mat. Samples were processed in laboratory, were the water fern was separated from *Lemna spp*. and density of the different developmental stages of *S. rufinasus* (larva, pupa and adult) was estimated. In order to determine the biomass of *A. filiculoides*, fresh samples were dried at 60 ºC during 48 hours.

Results

The first findings of *S. rufinasus* specimens (Fig. 1) in the Ter River took place in 2008 near Pont Major (Girona) and Sobrànigues (Sant Jordi Desvalls) (localities 1 and 2 in Table I and Fig. 2). In April 2009 *S. rufinasus* was found in two *A. filiculoides* mats located in the Ter River near Girona (locality 3 in Table I, Fig. 2 and 3).

Although *A. filiculoides* was already detected in February, significant growths of *A. filiculoides* biomass were recorded during April and May (Fig. 4), coinciding with the presence of sporocarps (sexual structures). During this period, changes in the morphology of the fronds were observed (increase of surface and volume of the fronds). In May, 100% of the fronds presented sexual structures.

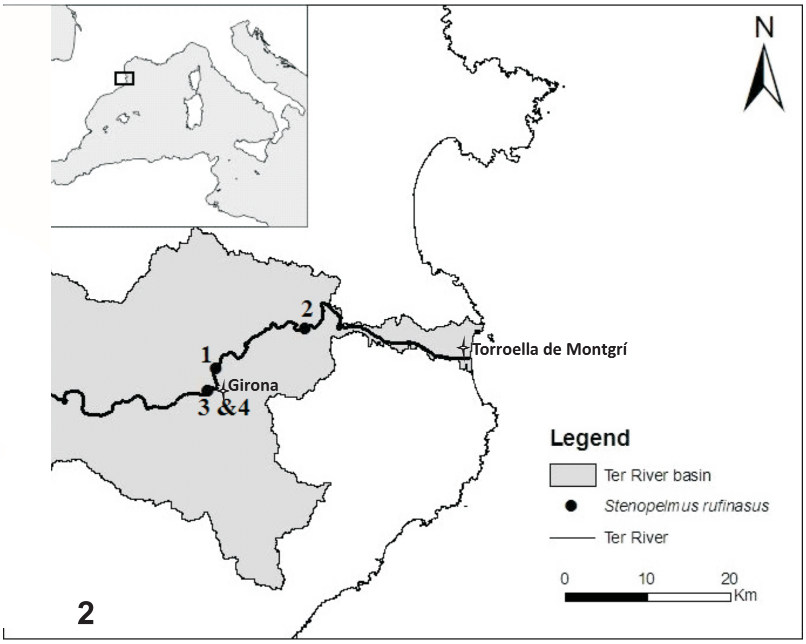
A density increase of *Stenopelmus rufinasus* was observed coinciding with the growth of *Azolla* mats (R^2= 0,294, p<0,01) (Fig. 5 and 6). In May the highest densities of the weevil were recorded ($\bar{x}$= 156,25 ± 96,56 adults·m^{-2}). Later, in June, *A. filiculoides* mats decayed and *Lemna minor* Linnaeus, 1753 and *L. gibba* Linnaeus, 1753 replaced the water fern. Small densities of the weevil ($\bar{x}$= 16,67 ± 20,41 adults·m^{-2}) were found associated with *Lemna* spp. mats. During this month, fronds of water fern were observed being transported downstream and adults of *S. rufinasus* were detected on them (locality 4 in Table I and Fig. 2).

Discussion

The finding of *S. rufinasus* increases the number of exotic species present in the Ter river basin downstream the reservoirs system, where a large number of exotic species are currently known (Table II). Specifically, 59 exotic species are known: 30 animal and 29 plant species strictly related to aquatic or riparian habitats. The number of exotic vertebrates is especially high, and fish is the most problematic group (16 out of 19 exotic vertebrates are fish). This situation is even worse if we take into account the entire basin, since more exotic species are known in the reservoirs (Carol *et al*., 2003) and upstream the reservoir system (e.g. presence of the exotic fish *Barbatula barbatula* (Linnaeus, 1758); Boix *et al*., 2010). Although invertebrates are more difficult to detect than vertebrates, 11 species have been reported. It is important to note that several fish species and one invertebrate are known only from Banyoles Lake, where species introduction became frequent along XX century (Zamora & Pou-Rovira, 2003).

Although the effects of *S. rufinasus* on autochthonous populations in the Ter River is at present unknown, some studies performed in South Africa revealed that the specificity of this weevil for *Azolla* spp. was very high (Hill, 1998). This study showed that for a group of 31 plant species, *S. rufinasus* only fed or laid eggs in the four species of *Azolla*. This high specificity agrees with the observations made in Ter River, where densities of *S. rufinasus* increased with the increases of biomass of *A. filiculoides* in April and May. Relationships between *S. rufinasus* and other aquatic plants, however, have not been considered in this study.

In June, when a decrease of the biomass of *A. filiculoides* took place, the densities of *S. rufinasus* also decreased. The decrease of *A. filiculoides* was probably due to a maximum daily temperature higher than 25ºC. Janes (1997) established that over this temperature an increase of respiration process could not be compensated by photosynthesis. A faster dehydration of fronds with sporocarps compared to other without these structures was also observed. This, combined with high temperatures, may have accelerated the death of *A. filiculoides*. The substitution of *A. filiculoides* by *Lemna* spp. mats during June did not prevent the decrease of *S. rufinasus* density, as it seems that the weevil can not survive in *Lemna* spp. (Hill, 1998).



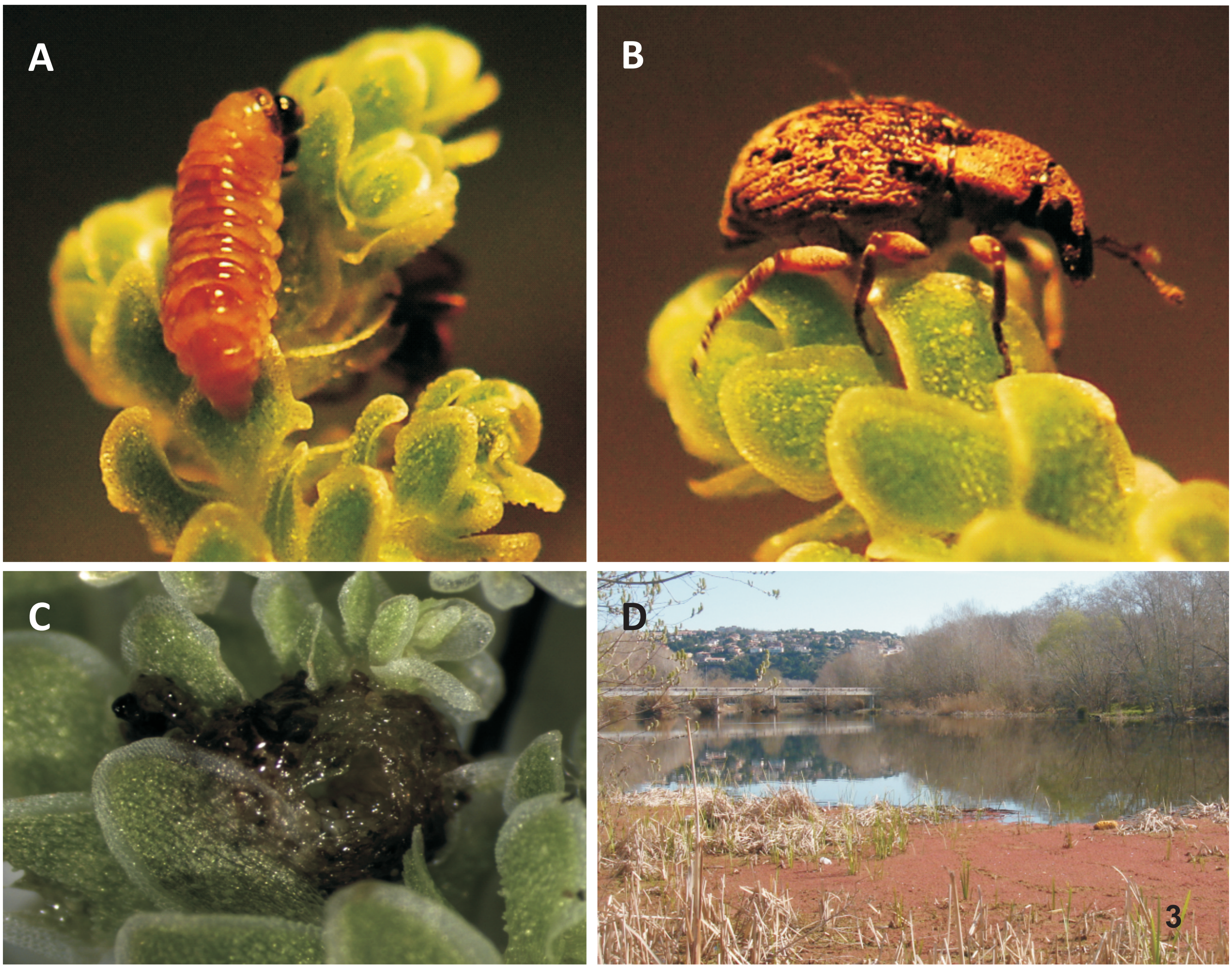


Fig. 1. Habitus of *Stenopelmus rufinasus*. (Author: Jordi Sala). **Fig. 2.** Localities of *Stenopelmus rufinasus* in medium-low stretch of the Ter River. **Fig. 3.** Alive larva (**A**) and adult (**B**) of *Stenopelmus rufinasus* (Author: Albert Ruhí). Larval chamber (**C**) among *A. filiculoides* fronds (Author: Jordi Sala). In the foreground, *Azolla filiculoides* mat in Ter River (**D**). (Author: Jordi-René Mor).

Table II. Exotic species list found in the medium-low stretch of the river Ter basin (downstream reservoirs system).
HGI is the international code identification of the herbarium of University of Girona in the Index Herbariorum (http://sciweb.nybg.org/science2/IndexHerbariorum.asp).

		Reference
PTERIDOPHYTA	*Azolla filiculoides* Lam.	J. Girbal *(pers. comm.)* [17.IV.2003]
	Salvinia natans (L.) All.	L. Polo in HGI [19.IX.1969]
MAGNOLIOPHYTA	*Acer negundo* L.	L. Polo in HGI [15.V.1976]
	Artemisia verlotiorum Lamotte	Bolòs (1947)
	Arundo donax L.	Codina (1908)
	Bidens frondosa L.	Sierra (1979)
	Buddleja davidii Franchet	Girbal (1984)
	Catalpa bignonioides Walter	L. Vilar in HGI [03.XI.2006]
	Cenchrus incertus M.A. Curtis	E. Bisbe in HGI [19.IX.1997]
	Cuscuta campestris Yuncker	Casasayas (1984)
	Cyperus eragrostis Lam.	J. Vicens in HGI [15.IX.1982]
	Euphorbia lathyris L.	Codina (1908)
	Fallopia baldschuanica (Regl) Holub	Casasayas (1989)
	Fraxinus pennsylvanica Marshall	L. Vilar in HGI [03.XI.2006]
	Gleditschia triacanthos L.	Casasayas (1989)
	Helianthus tuberosus L.	Girbal (1984)
	Hemerocallis fulva (L.) L.	Girbal (1984)
	Lepidium virginicum L. subsp. *virginicum*	Girbal (1984)
	Ludvigia grandiflora (Michx.) Greut. et Burdet	Margalef-Mir (1981)
	Oenothera biennis L. subsp. *biennis*	Costa (1877)
	Oenothera glazioviana Micheli	Codina (1908)
	Parthenocissus quinquefolia (L.) Planchon	Girbal (1984)
	Paspalum distichum L. (= *P. paspalodes* (Michx.) Scribner)	Codina (1908)
	Paspalum dilatatum Poiret in Lam.	Codina (1908)
	Sicyos angulatus L.	E. Fabregas in HGI [22.X.1995]
	Solidago altissima L.	Girbal (1984)
	Tritonia x *crocosmiflora* (Lemoine) Nicholson	Girbal (1984)
	Xanthium echinatum subsp. *italicum* (Moretti) O. Bolòs et Vigo	Vayreda (1879)
	Xanthium strumarium L. subsp. *strumarium*	Vayreda in Cadevall et col. (1919-1931)
CNIDARIA	*Craspedacusta sowerbyi* Lankester, 1880(1)	Abellà [25.IX.1977] in Prat (1979)
ANNELIDA	*Branchiura sowerbyi* Beddard, 1892 (1)	Prat *et al*. (1992)
	Ficopomatus enigmaticus (Fauvel, 1923)(2)	Martinoy *et al*. (2004)
MOLLUSCA	*Anodonta woodiana* (Lea, 1834)	Pou *et al*. (2009a)
	Corbicula fluminea (Müller, 1774)	Pou *et al*. (2009a)
	Physella acuta (Draparnaud, 1805)	Martinoy *et al*. (2004)
	Potamopyrgus antipodarum (Gray, 1843) J. Sala *leg*.	
CRUSTACEA	*Ankylocythere tiphophila* (Crawford, 1959)	J. Sala *leg*.
	Isocypris beauchampi (Paris, 1920)	J. Sala *leg*.
	Procambarus clarkii (Girard, 1852)	Boix *et al*. (2005)
INSECTA	*Stenopelmus rufinasus* Gyllenhal, 1836	this study
OSTEICHTHYES	*Ameiurus melas* (Rafinesque, 1820)(1,3)	Zamora & Pou-Rovira (2003)
	Carassius auratus (Linnaeus, 1758)	Pou-Rovira *et al*. (2007)
	Cobitis bilineata Canestrini, 1866	I. Doadrio (*pers. comm.*)
	Cyprinus carpio (Linnaeus, 1758)	Pou-Rovira *et al*. (2007)
	Esox lucius Linnaeus, 1758 (1,3)	Zamora & Pou-Rovira (2003)
	Gambusia holbrooki (Agassiz, 1859)	Pou-Rovira *et al*. (2007)
	Lepomis gibbosus (Linnaeus, 1758)	Pou-Rovira *et al*. (2007)
	Luciobarbus graellsii (Steindachner, 1866)(4)	Pou-Rovira *et al*. (2007)
	Micropterus salmoides (Lacépède, 1802)	Pou-Rovira *et al*. (2007)
	Mysgurnus anguillicaudatus (Cantor, 1842)	Franch *et al*. (2008)
	Perca fluviatilis Linnaeus, 1758 (1)	Zamora & Pou-Rovira (2003)
	Pseudorasbora parva (Temminck & Schlegel, 1846)	Pou-Rovira *et al*. (2007)
	Rutilus rutilus Linnaeus, 1758 (1)	Zamora & Pou-Rovira (2003)
	Sander lucioperca (Linnaeus, 1758)(1,3)	Zamora & Pou-Rovira (2003)
	Scardinius erythrophthalmus (Linnaeus, 1758)	Pou-Rovira *et al*. (2007)
	Tinca tinca (Linnaeus, 1758)	Pou-Rovira *et al*. (2007)
AMPHIBIA	*Discoglossus pictus* (Otth, 1837)	Boix *et al*. (2004)
REPTILIA	*Trachemys scripta* Schoepff, 1792	Boix *et al*. (2004)
MAMMALIA	*Mustela vison* Schreber, 1777	Pou-Rovira *et al*. (2007)

(1) Exotic species with presence only documented in Banyoles lake.
(2) Exotic species with presence only documented in Baix Ter coastal lagoons.
(3) Exotic species nowadays disappeared.
(4) Translocated species from Ebro basin.

Stenopelmus rufinasus is considered the only feasible long-term way to control the water fern *Azolla filiculoides* (Hill, 1999). Its effectivity to control *Azolla* has been widely verified in South Africa (McConnachie *et al.*, 2003), but the results achieved in the UK are contradictory (Gassmann *et al.*, 2006). In Ter River, sexual reproduction of *A. filiculoides* occurred before the increase of the weevil populations, and most likely they could not prevent the formation and liberation of sporocarps.

During 2009, the extension of *A. filiculoides* in the Ter River was lower than in previous years. This fact could be explained as the result of a large river flow during the studied period, which limited its growth and reduced its distribution to the localities near Girona. This could be determining for the distribution of *S. rufinasus*. The weevil was not detected between January and March 2009, probably as a consequence of low density of *A. filiculoides* mats or even its absence. Therefore, it is difficult to determine the effect of *S. rufinasus* as an *Azolla* biocontroller in the Ter River.

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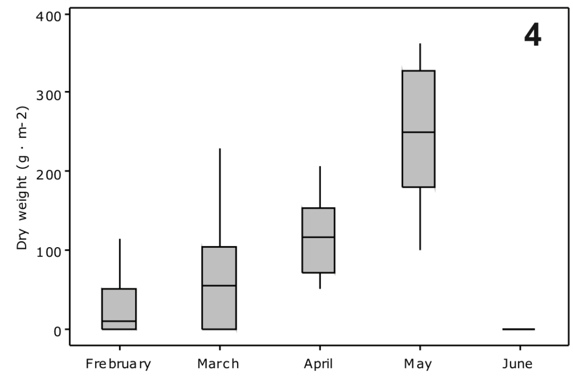

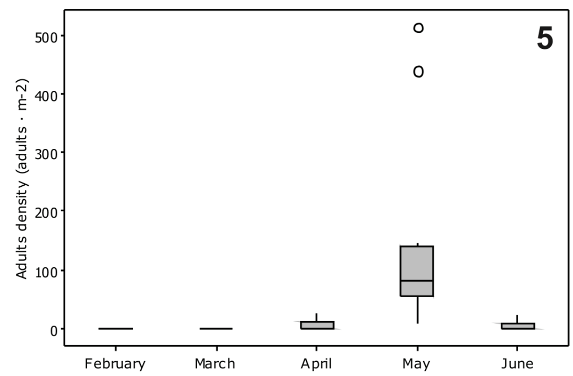

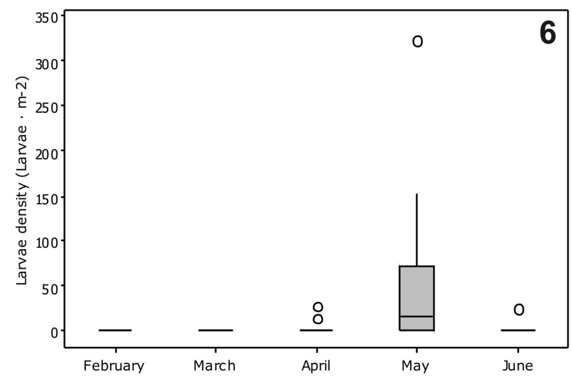

Fig. 4. Temporal variation of *Azolla filiculoides* density in the Ter River. **Fig. 5.** Temporal variation of *Stenopelmus rufinasus* adult density (individuals · m^{-2}) in the Ter River. **Fig. 6.** Temporal variation of *Stenopelmus rufinasus* larva density (individuals · m^{-2}) in the Ter River.

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