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Life cycle and nymphal feeding of *Rhabdiopteryx christinae* Theischinger 1975 (Plecoptera: Taeniopterygidae)

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Abstract. In the present paper some aspects of the nymphal biology of *Rhabdiopteryx christinae*, an endemic species from the Iberian Peninsula, are studied in a temporary stream from Southern Spain. *R. christinae* has an univoltine life cycle, with very short, fast and constant nymphal development, only having four months (December-March), related with the seasonality of the stream. In order to resist the period in which the stream has no water, an egg diapause probably occurs. The study of nymphal gut contents showed that *R. christinae* acts mainly as collector (detritus) and scraper (epiphytic and epilitic diatoms), as it is generally accepted for the family Taeniopterygidae.

Résumé. Cycle vital et alimentation de la larve de *Rhabdiopteryx christinae* Theischinger 1975 (Plecoptera : Taeniopterygidae). Dans cet article certains aspects de la biologie larvaire de *Rhabdiopteryx christinae*, une espèce endémique de la Péninsule Ibérique, sont étudiés dans un cours d'eau temporaire du sud de l'Espagne. *R. christinae* présente un cycle vital univoltine, avec une période de développement larvaire très courte, rapide et constante, qui ne dure que quatre mois (décembre-mars), liée à la temporalité du ruisseau. Pour affronter la période dans laquelle le ruisseau est à sec, il se produit probablement une diapause chez l'œuf. L'étude des contenus digestifs des larves a montré que *R. christinae* se comporte notamment comme collecteur (détritus) et comme racleur (diatomées épiphytes et épilithiques), en accord avec ce qui a été habituellement accepté pour la famille Taeniopterygidae.

Keywords: Stonefly, *Rhabdiopteryx christinae*, growth, nymphal diet, temporary stream.

Rhabdiopteryx Klapálek 1902 is one of the four European genera of the Taeniopterygidae family. Apart from it, only two more are present in the Iberian Peninsula, i.e., *Taeniopteryx* Pictet 1842 and *Brachyptera* Newport 1849 (Tierno de Figueroa *et al.* 2003).

Rhabdiopteryx counts with ten species in Europe and Anatolia, three of them represented in the Iberian Peninsula, all with a restricted general distribution: *R. antoninoi* Vinçon y Ravizza 1999, an endemic species which has been collected in mountain streams of Asturias and Cantabria (Northern Spain), *R. thienemanni* Illies 1957, also endemic from Spain, Portugal and Southeastern France, and *R. christinae* Theischinger 1975, endemic of the Iberian Peninsula, although its distribution in this area is not well-known yet (Sánchez-Ortega *et al.* 2002, Tierno de Figueroa *et al.* 2003).

R. christinae has a flight period going from winter to spring (Theischinger 1975, Luzón-Ortega *et al.* 1998, Vinçon & Ravizza 1999), although in some places adults have been collected in June (Sánchez-Ortega *et al.* 2002, Tierno de Figueroa *et al.* 2003). This species was catalogued as vulnerable at a global scale by Sánchez-Ortega & Tierno (1996), although new collects in the last years make us be more optimistic and predict a wider distribution area (Tierno de Figueroa *et al.* 2003).

Rhabdiopteryx in general, and *R. christinae* in particular, are quite unknown. As shown before, there are only some data about its distribution and phenology. The purpose of this paper is to extend the knowledge of the nymphal biology of this species, studying some aspects such as the life cycle and the feeding habits. Thanks to this, we want to expand the biological knowledge of one of the less studied genera of the European stonefly fauna and to contribute to the general understanding of the life history of Plecoptera inhabiting temporary waters. Drought is a fundamental conditional factor in the growth and development of the aquatic insects that live in this type of rivers and streams (Giller & Malmqvist 1998, Tierno de Figueroa & Luzón-Ortega 2002), but only in a few Plecoptera species from temporary streams (and never from the *Rhabdiopteryx* genus) the nymphal biology has

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been studied. Moreover, we also want to remark the importance of studies on life cycles, feeding and some other aspects of stonefly biology at species level, given that it is not possible to infer this information from the general pattern obtained from some other species that, living in different conditions, may behave in a different way. Some traditionally, feeding categories at family level have been recognised but, as pointed by Stewart & Stark (1993), food habits of an unstudied species cannot be inferred or deduced from the placement of a genus or higher taxon in a generalized grouping based on studies of congeners.

Material and methods

Nymphs were collected in an Arroyo de las Perdices tributary, a temporary stream sited in Sierra de Huétor (Granada, Spain), UTM ED50: 30SVG574277, 1380 m, from December 2003 to March 2004. They were catched using a kick net of 300 µm of mesh size, and preserved in 70% ethanol. Samplings were carried out once per month from November, when the stream began to having water, but not to having flow, to July, when the stream became dry. No one nymph was collected in the last months. Those caught on March had already completely developed wing pads, representing the last instars of this species. Measures of water temperature were taken during each sampling.

For generating the life cycles, measures of both pronotum (from one side to the other) and right hind femur (on the outer side) were taken. Data were obtained with the micrometer of an Olympus binocular microscope at 40x.

In order to study the gut content, we proceeded as in other papers studying feeding (Tierno de Figueroa *et al.* 1998, Tierno de Figueroa & Sánchez-Ortega 1999, 2000, Tierno de Figueroa & Fochetti 2001, Derka *et al.* 2004), which are based on the methodology of Bello & Cabrera (1999) for the identification of aquatic insects, i.e., we introduced every individual in a vial with Herwitgs´ liquid (a variation of Hoyer´s liquid) for 20-24 hours and put them into an oven at 65ºC. After this time we put the specimens already transparented on a slide glass with a cover glass on, and brought them to an Olympus microscope, where we estimated the absolute content percentage (measured as percentage of occupied area) at 40x, and the relative percentage of each component present in the gut at 400x. In order to detect the variation of the nymphal diet along their development we established five size classes. We also studied the variation of each component in relation to the nymphal size.

When analyzing data for life cycle we used a normality test in order to see the distribution of our data set, and established a correlation (Spearman R) between femur length and pronotum width, for deciding which measure was more apropiate to represent the nymphal growth. For feeding we used measures of mean, standard deviation and range of each component in each size class, and a correlation analysis (Spearman R) for determining the variation of each component along the growth period.

Results and discussion

Life cycle

In order to infer the life cycle we employed one of the two measurements we took (pronotum), given the fact that the correlation between pronotum width and femur length was very straight (Shapiro-Wilk W= 0.93, p< 0.01, N= 268 for pronotum width; Shapiro-Wilk W= 0.88, p< 0.01, N= 268 for femur length; Spearman correlation R= 0.96, p< 0.05).

From the data we collected, we can say that *R. christinae* has an univoltine life cycle. As shown in figure 1, nymphal development is very short, with a fast growth, only having four months, related with the temporality of the stream, which only carries water a few months during the year. The nymphal growth during only four months each year (December-March) has been also noted for other temporary water stonefly, *Guadalgenus franzi* (Aubert 1963) in Guadalvacarejo stream (Southern Spain), despite to present a semivoltine life cycle (Agüero-Pelegrín & Ferreras-Romero 2002). In *R. christinae* nymphal growth is

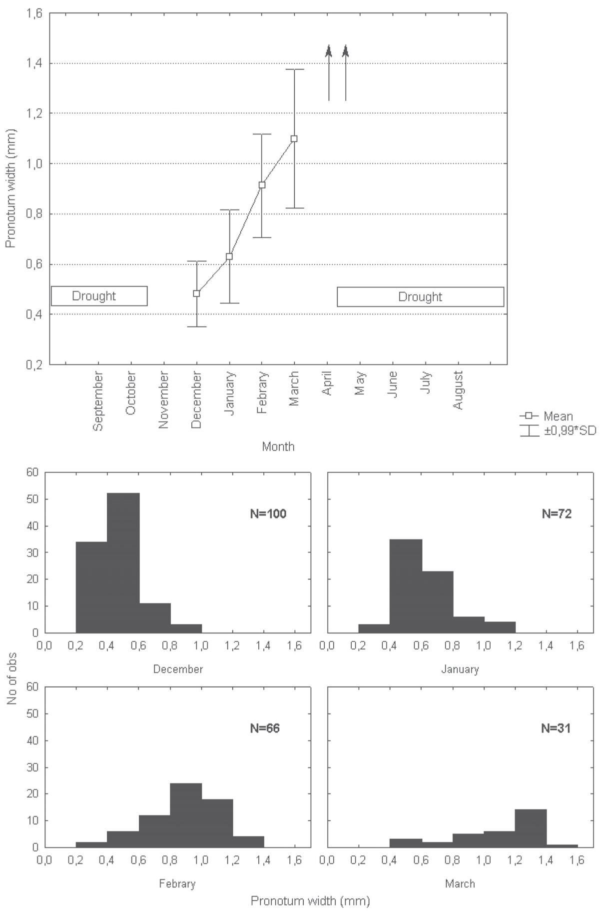

Figure 1
Life cycle of *Rhabdiopteryx christinae*; Vertical arrows indicating emergence. Histograms representing number of collected individuals of each size interval in the differents months.

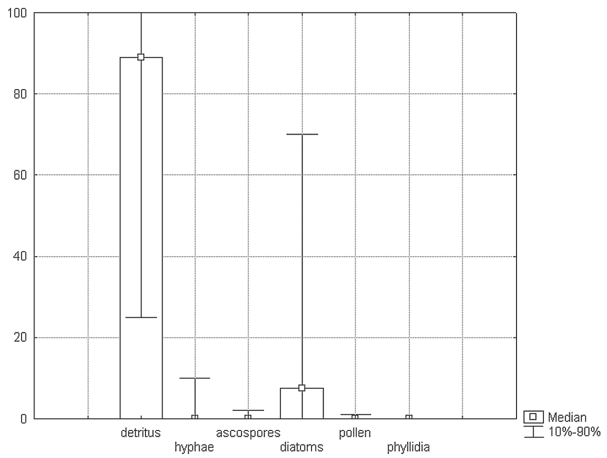

Figure 2
Diet composition in percentage of *Rhabdiopteryx christinae* nymphs.

Table 1. Absolute percentage of gut content by size class.

Size class	N	% absolute		
		Mean	SD	Range
[0.20, 0.50]	63	14.67	16.92	0-60
(0.50, 0.75]	83	31.93	29.34	0-100
(0.75, 1.00]	49	31.84	33.29	0-90
(1.00, 1.25]	35	44.71	37.22	0-100
(1.25, 1.50]	12	27.67	25.79	0-70

Table 3. Relationships between nymphal size (pronotum width) and percentage of the different gut components.

Correlation	R	p
Pronotum *vs.* detritus	- 0.38	< 0.05
Pronotum *vs.* hyphae	0.17	< 0.05
Pronotum *vs.* ascospores	0.12	> 0.05
Pronotum *vs.* diatoms	0.39	< 0.05
Pronotum *vs.* pollen	0.30	< 0.05
Pronotum *vs.* phyllidia	0.15	< 0.05

quite constant along the development, not having any period with a faster growth rate, as has been pointed out in other stonefly species (Harper & Hynes 1972, Benedetto 1973, Harper 1973, Barton 1980, Baekken 1981, Harper 1990, Sánchez-Ortega & Alba-Tercedor 1990, Harper *et al.* 1991, Derka *et al.* 2004). The growth of this species occurs in cold water (the data we recorded ranged between 3.5 to 9 ºC during the development).

Emergence occurs in late March-early April, and flight period seems to be like that pointed out in Luzón-Ortega *et al.* (1998) for the same stream.

In order to resist the period in which the stream has no water, an egg diapause probably occurs, given the fact that after emergence and before the estival drought no one nymph was found. Although nymphal diapause has been found in several species of the superfamily Nemouroidea, it has been reported in third to seventh instar, depending on the species (Zwick 1980, Stewart & Stark 1993), supporting our hypothesis of an egg diapause for this species.

Following the classification of life cycles made by Hynes (1970), *R. christinae* would be a fast seasonal species, due to it has a short period of development and spends a long time in egg diapause stage.

In other species of Taeniopterygidae, different types of life cycle have been found. In *Taeniopteryx nebulosa* (L. 1758) there is no diapause stage (Brittain 1977, Tierno de Figueroa *et al.* 2003, Zwick & Hohmann 2003). In other members of the same genera, such as *T. nivalis* (Fitch 1847) and *T. burksi* Ricker & Ross 1968, a nymphal diapause in the forth or fifth instar has been described (Harper & Hynes 1970, Harper & Hynes 1972, Stewart & Stark 1993), as also occurs in the members of the subfamily Brachypterainae, as *Strophopteryx fasciata* (Burmeister 1839) (Harper & Hynes 1970, Stewart & Stark 1993). In other members of this group, egg diapause has been described. In *Brachyptera seticornis* (Klapálek 1902), Neumann (1992) found an egg diapause of two or three months, after which the embryogenesis started (Tierno de Figueroa *et al.* 2003). Khoo (1964) pointed out a diapause of several months in the eggs of *B. risi*

Table 2. Percentages of the different gut components by size class.

Size class	N	% detritus			% hyphae			% ascospores			% diatoms			% pollen			% phyllidia		
		Mean	SD	Range	Mean	SD	Range	Mean	SD	Range	Mean	SD	Range	Mean	SD	Range	Mean	SD	Range
[0.20, 0.50]	61	92.98	17.78	20-100	0.87	2.56	0-15	1.02	6.42	0-50	5.10	15.82	0-80	0.03	0.26	0-2	0	0	0
(0.50, 0.75]	56	77.52	27.91	10-100	3.29	12.18	0-90	0.68	1.36	0-5	18.27	26.08	0-90	0.16	0.42	0-2	0.09	0.67	0-5
(0.75, 1.00]	29	80.72	25.12	15-100	0.86	2.15	0-10	0.28	0.96	0-5	17.97	24.56	0-85	0.17	0.47	0-2	0	0	0
(1.00, 1.25]	25	85.16	18.02	26-100	0.84	1.46	0-5	1.12	3.11	0-15	12.04	17.59	0-70	0.44	0.82	0-3	0.40	2.00	0-10
(1.25, 1.50]	9	74.11	15.19	47-100	1.89	2.15	0-5	0.22	0.44	0-1	21.11	16.16	0-50	0.44	0.53	0-1	2.22	6.67	0.20

(Morton 1896), and Lillehammer *et al.* (1989) noted in some Plecoptera species a prediapause stage in which the egg may remain for seven or eight months.

Feeding

The study of nymphal gut contents (tables 1 & 2) shows that detritus is the principal component of the species diet, followed by diatoms, while the others components (phyllidia, fungi -both hyphae and spores- and pollen) are usually present in a very low percentage (fig. 2). The diet composition indicates that *R. christinae* acts mainly as collector (detritus) and scraper (epiphytic and epilitic diatoms), coinciding with the usually accepted for the family Taeniopterygidae (Hynes 1976, Stewart & Stark 1993, Tierno de Figueroa *et al.* 2003).

When we studied the correlation between the different food categories and the nymphal size (table 3), we found a significant decrease ($p < 0.05$) of detritus percentage, supporting that pointed out for two Brachypterainae species from North Morocco by Azzouz & Sánchez-Ortega (2000). The other components significantly increase in relation with individual size, except for ascospores, that do not change significantly. From these results, we may conclude that the role as scraper for this species increases as it grows and that it also acts, although with an insignificant role, as shredder (phyllidia) when the nymphs become older.

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