Contribution to the taxonomy of the Central European species of *Rhithrogena loyolaea* species-group (Ephemeroptera: Heptageniidae)

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Abstract

The subimago male of *Rhithrogena gorganica* is described, and a complementary description of the imago female of this species is presented. The imago female and subimago female of *Rhithrogena loyolaea* are described and illustrated. Morphological characters of larvae and winged stages of *R. loyolaea* are considered and the synonymy of this species with *Rhithrogena tatrica* is confirmed. Critical characteristics for distinguishing *Rhithrogena zelinkai* from other species of *R. loyolaea* species-group are discussed. Complementary descriptions of eggs of *R. zelinkai* and *R. gorganica* are presented. General remarks on the distribution and biology of the above species are given. Keys to identification of larvae and imago males of *R. loyolaea* species-group are presented.

Keywords: *Rhithrogena loyolaea* species-group, synonymy, distinguishing characters, distribution, Alps, Carpathians.

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Introduction

Modern taxonomic knowledge of the European species from the genus *Rhithrogena* allows us to classify *Rhithrogena gorganica* KLAPÁLEK, 1907, *Rhithrogena loyolaea* NAVÁS, 1922 and *Rhithrogena zelinkai* SOWA & SOLDÁN, 1984 to the *R. loyolaea* species-group as defined by Sowa (1984). *R. gorganica* from the Eastern Carpathians was described by Klapálek (1907). The analysis of the material from the Polish Carpathians allowed Sowa (1971) to redescribe imago male and describe imago female, subimago female and larvae. Godunko (2000) presented a complementary description of larva of this species. Godunko and Soldán (2001) described the type material and defined lectotype, paralectotype and critical distinguishing characteristics of the imago male.

R. loyolaea is known from the mountain systems of Central and Southern Europe. Zelinka (1953) described *R. tatrica* ZELINKA, 1953 as a new species from the High Tatra (Slovakia). Moreover, Thomas (1970) proposed to synonimize *R. loyolaea* with *R. tatrica.* Although the synonymy proposed by Thomas was accepted in some papers (Sowa, 1984; Sowa and Soldán, 1984; Soldán and Landa, 1999), some authors stated *R. tatrica* as valid binomen (Zelinka, 1980; Tomka and Rasch, 1993; Novikmec and Krno, 1998). Hitherto only larval stages of *R. zelinkai* from Czech Republic, Slovakia and Austria are known (Sowa and Soldán, 1984; Deván, 1991; Soldán and Landa, 1999).

The aim of this paper is to redescribe the imago female of *R. gorganica* and *R. loyolaea*, the egg of *R. zelinkai*, describe the unknown subimago male and egg of *R. gorganica,* and to confirm the synonymy of *R. loyolaea* with *R. tatrica*. The critical distinguishing characteristics of winged stages and larvae have been defined in order to separate species within the *R. loyolaea* species-group. Finally, the data on distribution of species of this group have been analyzed.

Material (m: male; f: female; l: larvae)

Rhithrogena gorganica: UKRAINE. Ukrainian Carpathians: River Bystrets' 900 m a.s.l., 15.VII.1997, 2 m. imagines, Kłonowska-Olejnik and Godunko leg.; Natural Reserve "Gorgany", River Dovzhynets', "Ozirnyï" locality, 900 m, 22.VII.1997, 9 m. subimagines, 8 f. subimagines, 20 l., ibid, 24.VII.1997, 3 m. imagines, 4 f. imagines, Kłonowska-Olejnik leg.; Natural Reserve "Gorgany", S. slope Doboshanka Mt.,

River Zubra, 23.VII.1997, 4 m. imagines, 1 m. subimago, Kłonowska-Olejnik and Godunko leg.; Pozhyzhevs'ka Mt., Stream Breskulets', 1100 m, 27.VI.1998, 9 m. imagines, 3 f. imagines; S. slopes Khomyak Mt., Stream Bogdan, 850 m, 17.VII.1998, Godunko leg., 11 m. imagines, 5 f. imagines, 71 l; Natural Reserve "Gorgany", River Dovzhynets', "Ozirnyï" locality, 900 m, 20.VI.1999, 7 f. imagines, 1 m. subimago, 1 f. subimago, Godunko leg. POLAND. Polish Carpathians: Bieszczady Mts., Stream Wołosatka (upper part), 21.VI.1969, 1 m. imago, 1 f. imago, 2 l, Sowa leg.; Stream Wołosatka, 1050 m, 4.VI.1991, 6 l., Kłonowska-Olejnik leg.

Rhithrogena loyolaea: UKRAINE. Ukrainian Carpathians: River Yasenytsya, 850 m, 6.VI.1998, 16 l.; Verkhnya Yablon'ka vil., River Yablon'ka, 700 m, 12.VI.1998, 14 m. imagines, 6 f. imagines, 1 f. subimago, Godunko leg. POLAND. Polish Carpathians: Tatra Mts., Stream Sucha Woda, 17.VII.1986, 2 l; Stream Olczyski, 15.VIII.1986, 2 l, Kłonowska-Olejnik leg.; Stream Sucha Woda, 16.VIII.1986, 22 m. imagines, Kłonowska-Olejnik leg. SLOVAKIA. Slovakian Carpathians: River Mlynička, 1180 m, 4.VII.1973, 4 l; Popradské pleso, 8.VII.1973, 6 l.; Mlynica, Srbské pleso, 10.VII. 1973, 11 l.; Mlynica, waterfall Skok, 10.VII.1973, 10 l., Soldán leg. CZECH REPUBLIC. Šumava Mts., Horské Kuilova, 18.VIII.1974, 1 m. imago, 1 f. imago; Studený, Stream Žuberec, 19.VII.1975, 1 m. imago, 1 f. imago, Soldán leg. AUSTRIA. Alps: Kühtai, 2400 m, 13.VIII.1980, 1 l., Margreiter-Kownacka leg. FRANCE. Alps: tributaries left of Vorz, 19.VIII.1955, 3 l.; tributaries of de la Muzelle, 31.VIII. 1956, 4 larval skins; tributaries of Lake Mort, 4.VII.1959, 2 m. subimagines; Jsère, "st. 5", 1.IX.1970, 2 m. subimagines, 2 l; La Bonne, "st. 7", 8.IX. 1971, Degrange leg., 8 l., Sowa leg. Pyrénées: Estaragne, 19-21.VII.1983, 1 m. imago, Thomas leg. *Specimens labelled* "*Rhithrogena tatrica*": POLAND. Polish Carpathians: Babia Góra Mt., Markowe Szczawiny, 15-20.VIII.1962, 1 f. subimago, 1 subimaginal skin; Tatra Mts., Hala Ornak, Stream Kościeliski, 9.VIII.1965, 6 f. imagines, Sowa leg.; Stream Sucha Voda, 28.VIII.1967, 5 m. genitalia on slides Kownacki leg.; SLOVAKIA: Slovakian Carpathians: Valley Temnomerčanská, 5.VIII.1947, 6 l.; Stream Koprovský, 7.VIII.1947, 9 l. Landa leg.

Rhithrogena zelinkai (paratypes): AUSTRIA. Inferior Austria, Miesau Graben, 2 km of Moos Lassing (near Hochkarstrasse), 2.VIII.1975, 5 l. (part on slides), Puthz leg.

All materials are preserved in 75% alcohol and partially mounted on slides.

Results

Rhithrogena gorganica KLAPÁLEK, 1907

Rhitrogena gorganica; Klapálek, 1907, Čas. Čes. spol. entomol. 4: 32.
Rhithrogena gorganica; Zelinka, 1953, Spisy Přír. fak. Masar. Univ. 348: 161.
Rhithrogena gorganica; Sowa, 1971, Acta Hydrobiol. 13: 29.
Rhithrogena gorganica; Sowa, 1984, Proc. IV Internat. Confer. Ephemeroptera: 40.
Rhithrogena gorganica; Tomka and Rasch, 1993, Mitt. Schweiz. entomol. Ges. 66: 271.
Rhithrogena gorganica; Soldán and Landa, 1999, Klapalekiana 35: 28.
Rhithrogena gorganica; Godunko, 2000, Vestnik Zool. Suppl. 14: 61.

Description

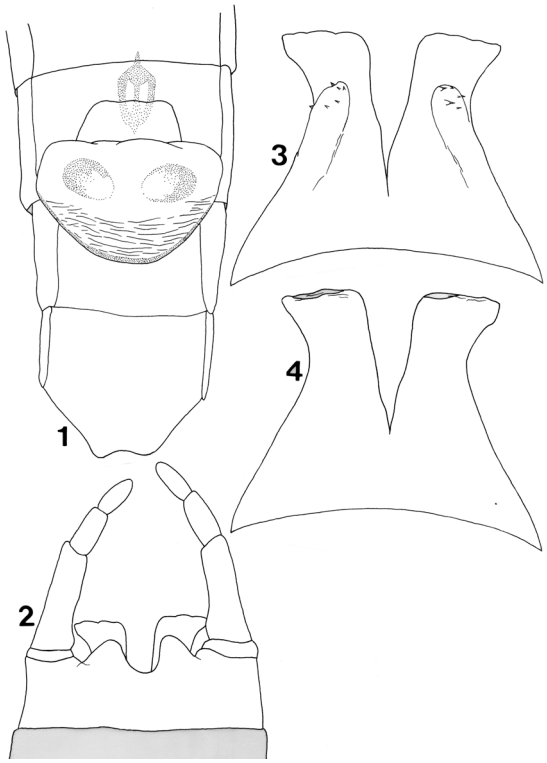
Female imago. Body length: 12.5-14.2 mm, fore wings: 13.0-15.0 mm, cerci: 21.1-23.6 mm. General coloration of body brown to dark brown. Head yellowish-brown to brown with paler spots at the facial keel. Eyes darker, slightly greyish towards the apex. Ocelli darker. Antennae light brown. General coloration of thorax yellowish-brown: protorax light brown, mesothorax yellowish brown, metathorax brown. Lateral part of thorax uniformly yellowish-grey. Ventral side of thorax brown, with clear visible darker violet nerve ganglia. Fore wings transparent, unicolorous, light brownish with light brown venation, darker in basal and medial parts. Pterostigmatic area milky. Hind wings of the same coloration as fore wings. Forelegs yellowish-brown. Femora with well visible dark elongated spot on each. Abdomen yellowish-brown with light reddish spots. Lateral part of abdominal segments with markings similar to those in subimago male. Ventral part of abdomen yellowish-brown with well visible darker violet nerve ganglia (Fig. 1). Cerci light brown, slightly darker in basal part.

Genitalia. Posterior abdominal segments in ventral view, as in Fig. 1. Subgenital plate relatively wide with slightly rounded posterior margin. Subanal plate rather short, with a slight incision.

Male subimago. Body length: 11.5-12.3 mm, fore wings: 11.8-12.7 mm, cerci: 11.3-12.0 mm. General coloration of body yellowish-brown. Head paler, yellowish-grey with paler spots at the

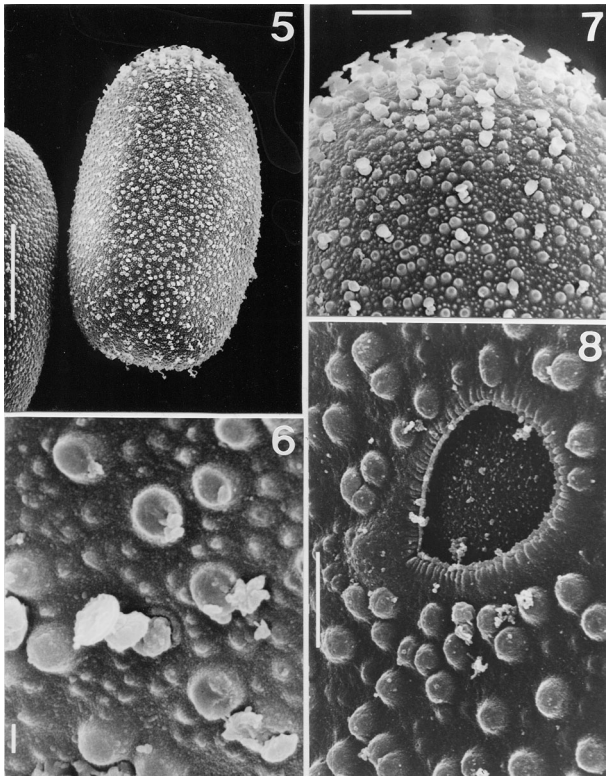
facial keel. Eyes darker, slightly grayish towards the apex, each bordered with a narrow yellowish stripe well visible in lateral view. Ocelli with paler spots at the apex. Thorax yellowish, paler, with well visible brown stripe on pronotum, prosternum and mesosternum. Wings uniformly grayish with grayish venation. Legs light, yellow to yellow-brownish. Femora with elongated dark spot at the central part. Forelegs with hardly visible irregular spot. Coxae yellowish-brown, trochanters darker. Segments of thorax in slight contrast with darker abdominal yellowish-brown to brown segments. Lateral part of abdominal segments from 2 to 8 with typical light spots. Nerve ganglia well visible, darker violet. Cerci light brown, slightly darker in basal part.

Genitalia. Styliger uniformly yellowish-brown. General coloration of forceps similar to styliger. Posterior margin of styliger with two well visible lateral protuberances, slightly pointed at the apex (Fig. 2). Penis lobe divergent (Figs. 3, 4) with bluntly pointed projection, well apparent in ventral and dorsal view. Titilators relatively large and rounded at the apex, with slightly divergent outline towards the apex. Titilators generally with 4 to 6 asymmetric teeth.



Figs. 1-4 - *Rhithrogena gorganica*: 1-female imago: genitalia, ventral view; 2-4 male subimago: 2-genitalia, ventral view; 3-penis, ventral view; 4-penis, dorsal view.

Egg. The egg has been first described by Sowa (1971) in light microscopy. The egg is oval (Fig. 5). Dimensions: 172 µm in length and 98 µm in width. The surface of the egg is rugose. Many tubercles (macrogranules, 2.0-3.0 µm in diameter) are scattered all over the surface of the chorion. Between tubercles, small protuberances are also visible (Fig. 6). Knob-terminated coiled threads (KCT) attachment structures are of two kinds: large and small. There is a concentration of large KCT attachment structures at one pole (Fig. 7). Small KCT attachment structures are densely distributed all over the chorion surface. One or two oval micropyles are located in the equatorial or subequatorial area. Sperm guide ovoidal 8.1 µm in length and 6.2 µm in width. The macropylar rim is relatively wide and denticulate (Fig. 8).



Figs. 5-8 - *Rhithrogena gorganica,* egg: 5-general view, scale bar=50µm; 6-chorionic surface with tubercles, small protuberances and small KCT attachment structures, scale bar=1 µm; 7-egg pole with large KCT attachment structures, scale bar=10 µm; 8-micropyle with the sperm guide, scale bar= 5 µm.

***Rhithrogena loyolaea* NAVÁS, 1922**

Rhithrogena loyolaea; Navás, 1922, Bol. Soc. entomol. Esp. Zaragoza 5: 62.

Rhithrogena tatrica; Zelinka, 1953, Spisy Přír. fak. Masaryk Univ. 348: 159.

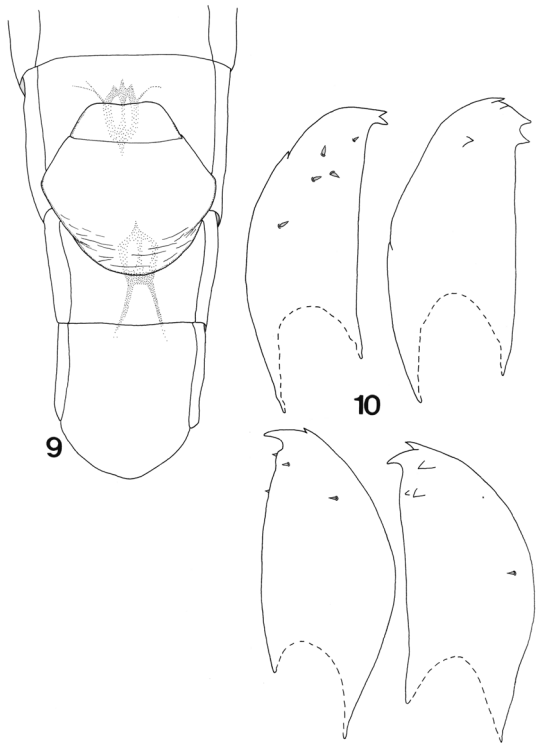
Rhithrogena nivata; Grandi, 1960, Fauna d'Italia 3: 84.

Rhithrogena loyolaea; Thomas, 1970, Annls Limnol. 6: 305.

Rhithrogena loyolaea; Sowa, 1984, Proc. IV Internat. Confer. Ephemeroptera: 40.
Rhithrogena loyolaea; Tomka and Rasch, 1993, Mitt. Schweiz. entomol. Ges. 66: 271.
Rhithrogena tatrica; Tomka and Rasch, 1993, Mitt. Schweiz. entomol. Ges. 66: 271.
Rhithrogena loyolaea; Soldán and Landa, 1999, Klapalekiana 35: 28.
Rhithrogena loyolaea; Godunko, 2000, Vestnik Zool. Suppl. 14: 63.

Description

Female imago. Body length: 11.5-13.0 mm, fore wings: 12.0-14.0 mm, cerci: 16.0-18.0 mm. General coloration of body yellowish-brown. Head yellowish-brown. Eyes darker, each bordered with a narrow yellowish stripe well apparent in lateral view. Ocelli darker. Antennae brown. General coloration of thorax yellowish-brown to brown: prothorax and mesothorax yellowish-brown to brown, mesothorax darker brown. Lateral part of thorax yellowish-brown. Ventral side of thorax brown, nerve ganglia darker violet, well visible. Fore wings transparent, unicolorous, light brownish with light brown venation. Hind wings of the same coloration as fore wings. Pterostigmatic area milky. Legs yellowish-brown. Femora with a more or less visible elongated darker spot on each.



Figs. 9-10 - *Rhithrogena loyolaea*: 9-female imago: genitalia, ventral view; 10-*Rhithrogena loyolaea*: male imago, titilators (the Slovakian Carpathians, specimen labelled as "*R. tatrica*").

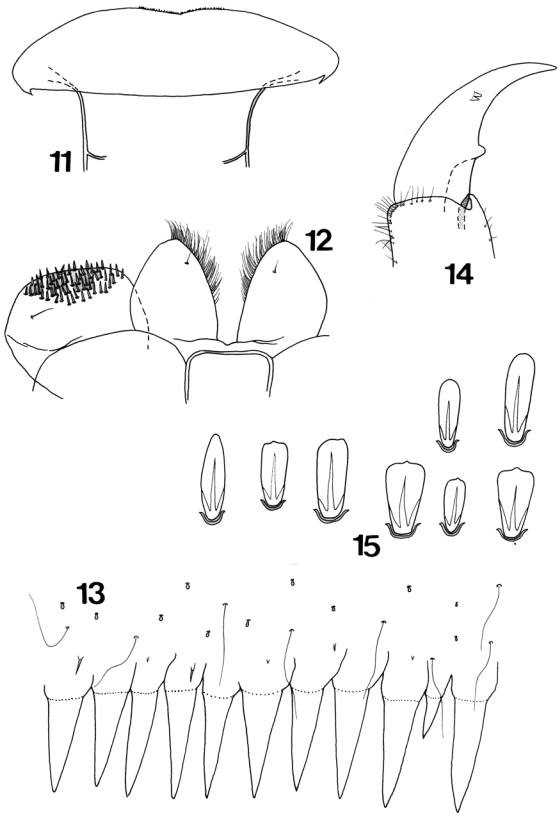
Abdomen yellowish-brown, unicolorous, without markings. Ventral part of abdomen yellowish-brown with well visible darker violet nerve ganglia (Fig. 9). Cerci brown.

Genitalia. Posterior abdominal segments in ventral view as on Fig. 9. Subgenital plate relatively wide with rounded posterior margin. Subanal plate rounded without incision.

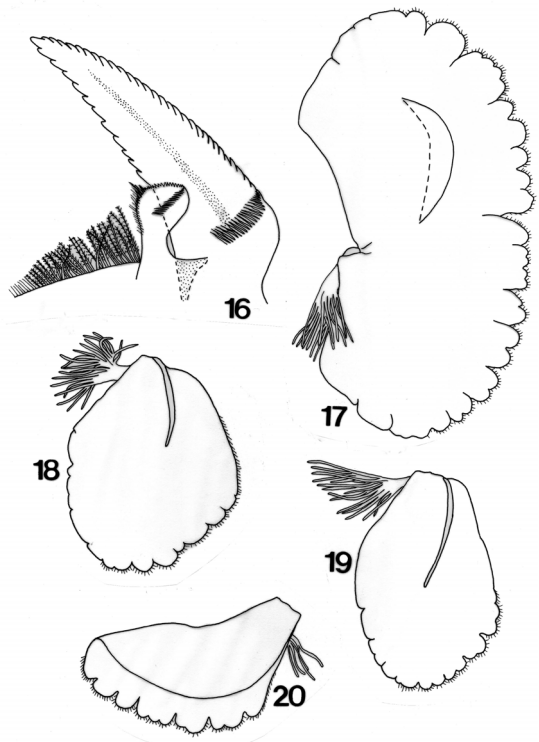
Female subimago. Body length: 11.0 mm, fore wings: 12.0 mm, cerci: 11.0 mm. General coloration of body similar to that in female imago. Wings uniformly greyish with distinct greyish venation. Ventral part of abdomen with well visible darker violet nerve ganglia. General view of posterior part of abdomen similar to that in female imago.

Rhithrogena tatrica, from the High Tatra, was described by Zelinka (1953). Thomas (1970) indicated *Rhithrogena loyolaea* as the senior synonym of *R. tatrica.* Unfortunately, our attempts to find the type material on *R. tatrica* in Masaryk University in Brno have failed and this material was probably lost (S. Zahrádková, T. Soldán, personal communications). However, we were able to critically analyze the available material on larvae and imago males from the collection of V. Landa and R. Sowa labelled "*R. tatrica*", and study the larvae of *R. loyolaea* collected by T. Soldán in the Mlynica region (the High Tatra, Slovakia) which is in immediate proximity to Zelinka's localities of *R. tatrica*. Examination of these materials has identified *R. tatrica* with *R. loyolaea*. The general appearance and coloration of the styliger and forceps, the shape of the penis lobes and the arrangement and orientation of the outer and inner teeth in specimens labelled "*R. tatrica*" are similar to those of the *R. loyolaea* researched by us and illustrated by Thomas (1970). The titilators of *R. tatrica* (Fig. 10) have a shape typical of *R. loyolaea*, large only at the base, curved, with a convergent outline towards the apex, and with numerous teeth on the titilator's surface. The structure of the larvae examined, labelled "*R. tatrica*", is in complete uniform with the diagnostic characteristics of *R. loyolaea* (Figs. 11-20).

A comparative study of *R. loyolaea* from some regions of the Carpathians, Czech Republic, Austrian and French Alps, has testified to differences in structure between two groups of larvae (cited below as "Carpathians" form and "Alpine" form).

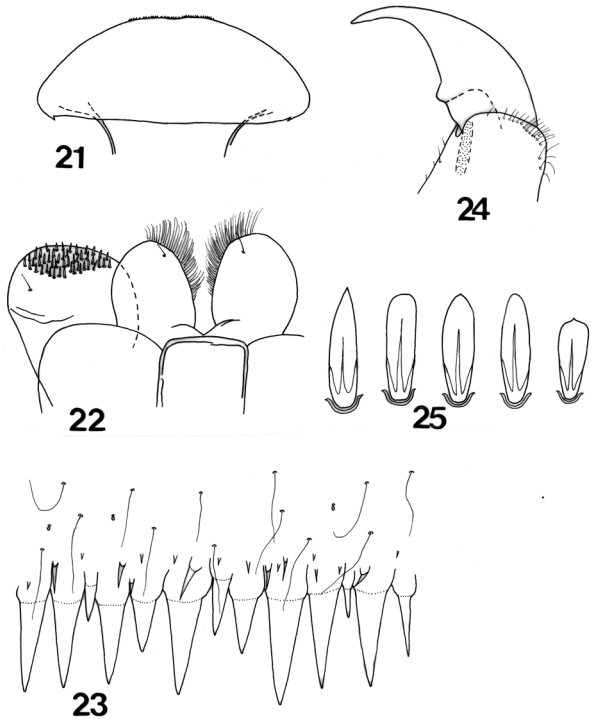


Figs. 11-15 - *Rhithrogena loyolaea*, larva (the Slovakian Carpathians, specimens labelled as "*R. tatrica*"): 11-labrum; 12-glossae and paraglossa; 13-posterior margin of abdominal tergites; 14-tarsal claw; 15-femoral spatulas.



Figs. 16-20 - *Rhithrogena loyolaea*, larva (the Slovakian Carpathians, specimens labelled as "*R. tatrica*"): 16-inner incisor of mandible; 17-first gill; 18-second gill; 19-sixth gill; 20-seventh gill.

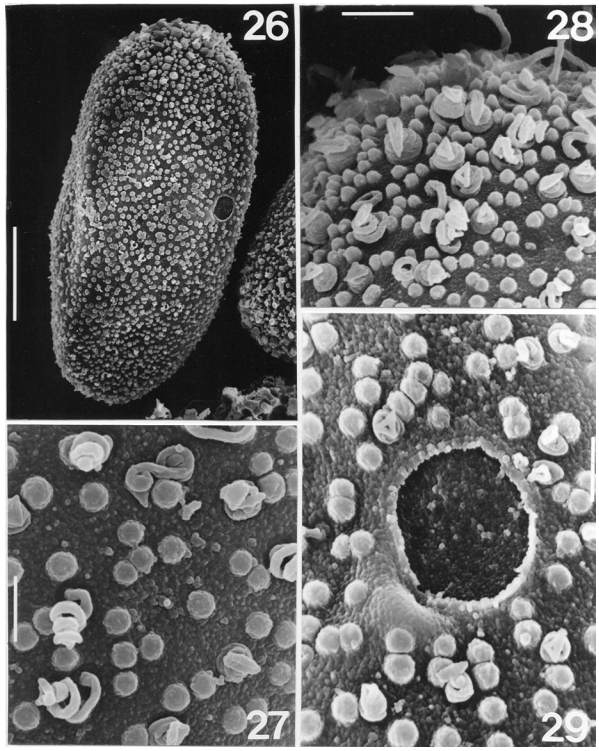
The "Carpathians" form can be distinguished by a combination of the following characteristics: (1) labrum relatively elongated and narrow (Fig. 11) with width / length ratio mean 0.26 (σ = 0.162; n = 20); glossae non-massive, slightly bluntly pointed at the apex (Fig. 12); (3) posterior part of abdominal terga with non-numerous subapical small pointed spines (Fig. 13); (4) tarsal claw with 0-2 teeth (mainly with 1-2) (Fig. 14); (5) femoral spatulas numerous on dorsal surface, mainly widened towards apex and slightly pointed at the apex (Fig. 15). To distinguish the *R. loyolaea* larvae of the "Alpine" form, we propose the combination of the following characteristics: (1) labrum relatively short and wide (Fig. 21) with width / length ratio mean 0.37 (σ = 0.195; n = 18); (2) glossae oval, massive, not pointed at the apex (Fig. 22); (3) posterior part of abdominal terga with numerous subapical small pointed spines (Fig. 23); (4) tarsal claw without teeth (sometimes with 1-2 small teeth) (Fig. 24); (5) spatulas on dorsal surface of femora not numerous, mainly narrow or tapered and rounded at the apex (Fig. 25).



Figs. 21-25 - *Rhithrogena loyolaea*, larva (the French Alps): 21-labrum; 22-glossae and paraglossa; 23-posterior margin of abdominal tergites; 24-tarsal claw; 25-femoral spatulas.

However, in the morphological structure of winged stages in both species forms, no differences have yet been revealed.

***Rhithrogena zelinkai* SOWA & SOLDÁN, 1984**
Rhithrogena zelinkai; Sowa, 1984, Proc. IV Internat. Confer. Ephemeroptera: 40.
Rhithrogena zelinkai; Sowa and Soldán, 1984, Proc. IV Internat. Confer. Ephemeroptera: 81.
Rhithrogena zelinkai; Tomka and Rasch, 1993, Mitt. Schweiz. entomol. Ges. 66: 271.
Rhithrogena zelinkai; Soldán and Landa, 1999, Klapalekiana 35: 28.



Figs. 26-29 - *R. zelinkai*, egg: 26-general view, scale bar=50 µm; 27-chorionic surface with tubercles and small KCT attachment structures, scale bar=5 µm; 28-egg pole with large KCT attachment structures, scale bar=10 µm; 29-micropyle with the sperm guide, scale bar= 5 µm.

Egg. The egg has been described and illustrated by Sowa and Soldán (1984) in light microscopy. The egg is oval (Fig. 26). Dimensions: 210 µm in length and 102 µm in width. The surface of the egg is rugose with a clear granular structure, composing of numerous randomly scattered tubercles (1.9-2.5 µm in diameter). Granular ground matrix covers both the chorion surface and tubercles. (Fig. 27). The attachment structure is characterized by knob-terminated coiled threads (KCT) of different size and arrangement. Large KCT attachment structures are concentrated at one egg pole (Fig. 28). Small KCT attachment structures are scattered all over the chorion surface. There are one or two micropyles in the equatorial or subequatorial area. Sperm guide ovoidal measures 11.4 µm in length and 9.7 µm in width. The micropylar rim is narrow, with no specific structure (Fig. 29).

Discussion and Conclusions

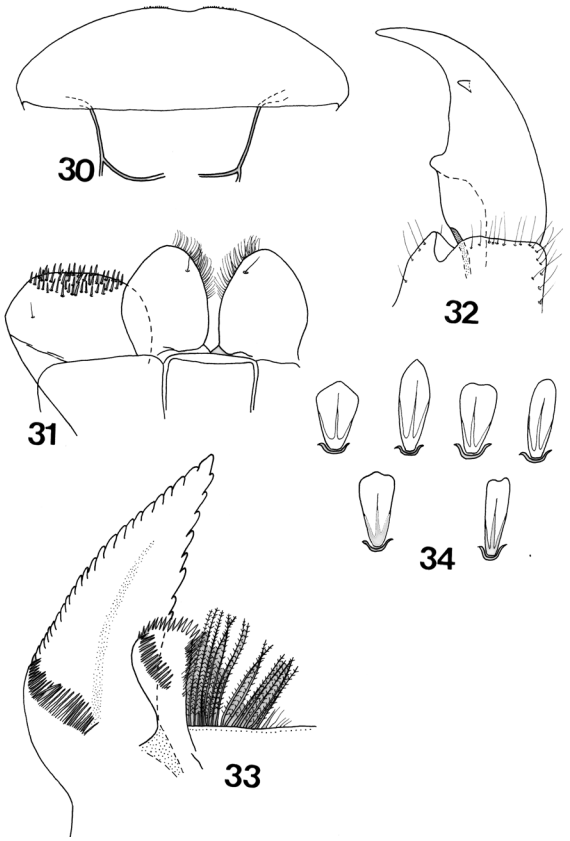
While describing the species *Rhithrogena tatrica*, Zelinka (1953) defined its position within the *semicolorata* species-group. Sowa (1971) indicated the intermediate position of the *Rhithrogena gorganica* between *semicolorata* and *nivata* species-groups. According to modern knowledge on taxonomy of the European species of the genus *Rhithrogena*, *R. gorganica*, *R. loyolaea* and *R. zelinkai* are ranged to the *R. loyolaea* species-group, as was defined by Sowa (1984). Larva of all species of *R. loyolaea* species-group are relatively well known and can be identified by existing keys (Tomka and Rasch, 1993; Soldán and Landa, 1999). The description of two forms of *R. loyolaea*, as well as, additional characteristics of species from *R. loyolaea* species-group induce critical analysis of the distinguishing characteristics formerly used for separating these species (Table 1).

The winged stages of *R. gorganica* and *R. loyolaea* are relatively well known and are described in literature (Thomas, 1970; Godunko and Soldán, 2001). Imago males of *R. gorganica* and *R. loyolaea* can be distinguished by the structure of penis lobes and titilators. In *R. gorganica,* penis lobes are slightly divergent, relatively short, with posterolateral projection bluntly pointed, well visible in ventral view; titilators are large, oblong-shaped, with parallel lateral margins. In *R. loyolaea,* penis lobes are clearly divergent, relatively elongated, well visible in ventral view; titilators are large only at the base, curved, with a convergent outline towards the apex. Subimago male of *R. gorganica* can be separated by the appearance of penis lobes, which are massive in ventral view, with clear bluntly pointed posterolateral projection, and by the shape of the titilators, which are massive and slightly rounded at apex (Figs. 3, 4). The peculiarities in structure of genitalia in male larva of *R. zelinkai* draw this species close to *R. gorganica* (Sowa and Soldán, 1984). Imago and subimago females of *R. gorganica* and *R. loyolaea* can be distinguished by the structure of the subanal and subgenital plate (Figs. 1, 9).

Larvae of *R. gorganica* can be clearly separated from those of other species of *R. loyolaea* species-group by the shape of gills II-VII

Table 1 - Main morphological characteristics for separating the species of the *R. loyolaea* species-group. 1 – Sowa, 1971; 2 – Sowa and Soldán, 1984; 3 – Kłonowska-Olejnik, 1997; 4 - Soldán and Landa, 1999; 5 – Godunko, 2000; 6 – Godunko and Soldán 2001: * – only male nymph characteristics.

Characteristics	***R. gorganica***	***R. loyolaea*** "Carpathians" form	***R. loyolaea*** "Alpine" form	***R. zelinkai***
Larva:				
Head				
labrum	robust[4, 5]	relatively elongated and narrow	relatively short and wide	relatively elongated and narrow
inner incisors of mandibles	elongated and oval[1]	triangular[4]	triangular[4]	elongated and oval[4]
glossa	bluntly pointed[4]	slightly pointed	not pointed, oval, massive	bluntly pointed[2]
Legs				
femoral coloration	diffused central spots in pale field	lighter field in darker frame[4]	lighter field in darker frame[4]	diffused central spots in pale field[4]
femoral spatulas	circular, pointed distally[4, 5]	elongated, mainly pointed distally	elongated, mainly rounded distally	elongated, slightly or bluntly pointed distally
number of teeth of the tarsal claw	generally 2[1]	generally 1-2	mainly without teeth	generally 2[2]
Abdomen				
gills II-VII	with 1-3 hard visible shallow incisions	with 6-15 well visible incisions	with 6-15 well visible incisions	with 4-6 well visible incisions
subapical spines on posterior margin of abdominal terga	small, not numerous	small, not numerous	massive, numerous	small, not numerous
Male imago:				
Body coloration	brown to dark brown[1, 6]	yellowish-brown	yellowish-brown	-
Genitalia				
penis lobes	slightly divergent, relatively short[6]	clearly divergent, relatively elongated	clearly divergent, relatively elongated	with rounded apex[2, *]
titilators	large, oblong-shaped[6]	large only at base, curved	large only at base, curved	large and wide[2, *]
Female imago:				
Body coloration	brown to dark brown	yellowish- brown relatively wide	yellowish- brown	-
Abdomen				
subgenital plate	relatively wide, with slightly rounded posterior margin	relatively wide, with rounded posterior margin	relatively wide, with rounded posterior margin	-
subanal plate	rather short, with slight incision	rounded, without incision	rounded, without incision	-
Egg	oval, tubercles and small protuberances of the chorion, micropylar rim wide and denticulate	oval or spindle-shaped, only tubercles of the chorion, micropylar rim thin, slightly denticulate[3]	-	oval, tubercles of the chorion with granular ground matrix, micropylar rim narrow



Figs. 30-34 - *Rhithrogena zelinkai*, larva (paratypes, Austria): 30-labrum; 31-glossae and paraglossa; 32-tarsal claw; 33- femoral spatulas; 34-inner incisor of mandible.

(especially of gills VII), which are slightly crenulated, nearly smooth with 1-3 (mainly 1-2) hardly visible shallow incisions (Soldán and Landa, 1999). Other distinguishing characteristics of *R. gorganica* - robust labrum, bluntly pointed glossae with straight or concave anterolateral margin, inner tooth of mandible almost straight, tarsal claw with 2 teeth and circular femoral spatulas (Soldán and Landa, 1999) - can't be accepted in full at present. Thus, the general appearance of labrum and its width / length ratio in *R. gorganica* are closer to that in the "Carpathians" form of *R. loyolaea* and in *R. zelinkai* (Fig. 30), which also possess bluntly pointed glossae (Fig. 31). In general, the availability of 0-2 teeth on tarsal claws (some examined paratypes of *R. zelinkai* lacked teeth on tarsal claws) and slightly or bluntly pointed femoral spatulas are typical of all species of *R. loyolaea* species-group (Figs. 32, 33). The straight oval inner tooth on the mandible (Fig. 34) and a well visible central spot on the femur in *R. gorganica* are also present in a part of *R. zelinkai* specimens. While separating *R. loyolaea* from *R. zelinkai*, Bauernfeind (1994) used the peculiarities of structure of femoral spatulas and tarsal claws. Taking into account the existence of two forms of *R. loyolaea*, the characteristics proposed by this

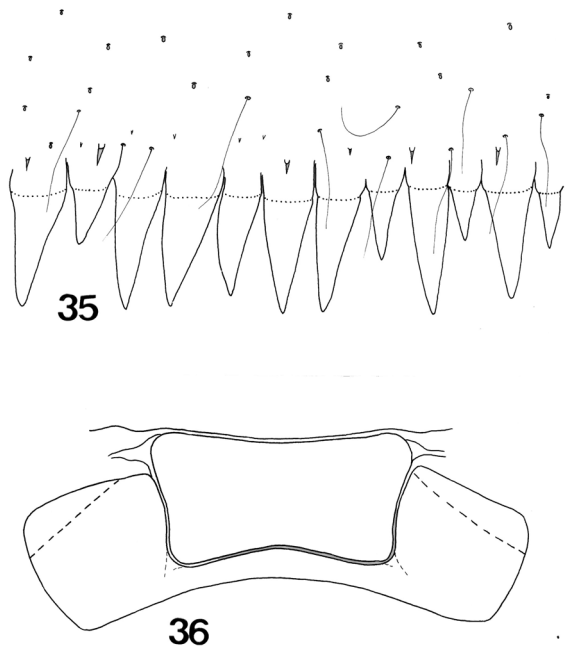
author are good only for distinguishing *R. zelinkai* from the "Alpine" form of *R. loyolaea*, but not from the "Carpathians" form with certainty.

Sowa and Soldán (1984), Soldán and Landa (1999) noted the shape and number of spatulas on the femora surface, the structure of the segment surface of caudal filament, the shape and crenulations of gills, the structural peculiarities of the inner incisor of the mandible and the availability of spots on femora, as distinguishing characteristics of *R. zelinkai*. As to the possibility of using such characteristics as the shape and crenulations of the gills, the shape of the inner incisor of the mandible, and the femora coloration for separating *R. zelinkai* from *R. loyolaea*, we completely agree with these authors. Thus, the number of incisions on gills II-VII of *R. loyolaea* vary from 6 to 15 (Figs. 17-20). In *R. zelinkai* gills II-VII bear only 4-6 well visible deep incisions, which mainly separate large lobes (Sowa and Soldán, 1984). The structure of the posterior margin of abdominal terga in *R. zelinkai* (Fig. 35) is similar to that of the "Carpathians" form of *R. loyolaea* (Fig. 13), and the shape of the first abdominal sternite (Fig. 36) is similar to that of *R. gorganica* and *R. loyolaea* s. l. (Sowa, 1971; Soldán and Landa, 1999) and in general is typical of the whole *R. loyolaea* species-group.

R. gorganica is Eastern Carpathians endemic (Godunko and Soldán, 2001). The distribution of *R. gorganica* in Europe are central mountain ridges along the main watershed range of the Ukrainian Carpathians (Godunko and Soldán, 2001); the Polish Carpathians: Bieszczady Mts. (Sowa, 1971; Kukuła 1991, 1995; Kłonowska-Olejnik, 1997, 2000); the Slovakian Carpathians: the Biosphere Reserve of the Eastern Carpathians (Bitušík and Novikmec, 1997; Novikmec and Krno, 1998) (Fig. 37).

R. loyolaea can be found in Europe in the Carpathian Mts. (Poland, Slovakia, Ukraine) (Sowa, 1975; Soldán and Landa, 1999; Godunko, 2000); the Sudete Mts. and the Šumava – Bohemian Forest Mts. (Czech Republic) (Soldán and Landa, 1999); the Adriatic and the Danube basin (Slovenia) (Zabric and Sartori, 1997); the Rhodopes Mts. (Bulgaria) (Braasch *et al.*, 1985); the Alps (Austria, Germany, Italy, France, Switzerland) (Soldán and Landa, 1999); the Pyreneyes Mts. (France and Spain) (Thomas, 1970; Soldán and Landa, 1999).

Hitherto, *R. zelinkai* is known from a few localities in the Elbe river-basin, the Krkonoše-Giant Mts. (Czech Republic); the High Tatra Mts. (Slovakia) and the Alps (Austria) (Sowa and Soldán, 1984; Deván, 1991; Bauernfeind, 1994; Soldán and Landa, 1999).



Figs. 35, 36 - *Rhithrogena zelinkai*, larva (paratypes, Austria): 35-posterior margin of abdominal tergite; 36-first abdominal sternite.

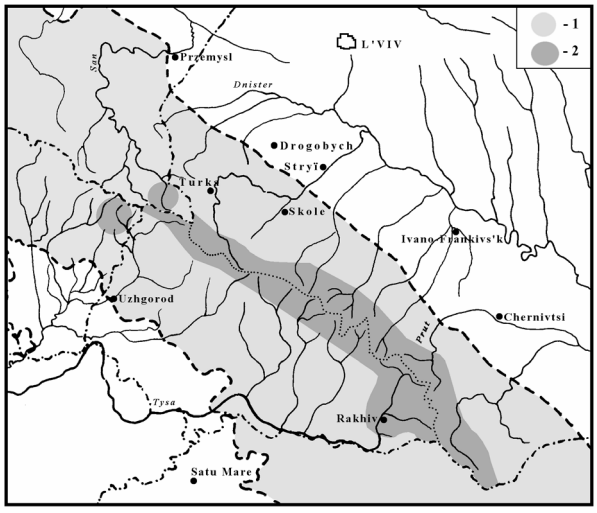


Fig. 37 - The general distribution of *Rhithrogena gorganica* KLAPÁLEK, 1907 in the Carpathian Mts.: 1– the Carpathian Mts.; 2– distribution of *R. gorganica*.

This study makes the description of two forms of *R. loyolaea* possible: the "Carpathians" and "Alpine" forms. Of the whole material on *R. loyolaea* investigated from the Carpathians and Czech Republic, not more than 20% larvae satisfied the diagnosis of "Alpine" form by some characteristics (for instance by proportions of the labrum and the structure of the tarsal claw); the greater being attributed to the "Carpathians" form.

At the same time, the material on this species investigated from the Alps was clearly attributed to the "Alpine" form of larvae. The analysis of specimens labelled "*R. tatrica*" has made it possible to attribute them with certainty to the "Carpathians" form of *R. loyolaea,* and thus confirm the synonymy of these species.

The final conclusion about the taxonomic status of *R. zelinkai* and both forms of *R. loyolaea* could only be drawn after a description of winged stages, and an investigation of much more larvae in the former species followed by the precise analysis of the variability of characteristics in larvae and imagines of the latter species in its populations from the Carpathians, the Alps and the Pyreneyes.

The key to identification of species of *Rhithrogena loyolaea* species-group:

Imago male (except *R. zelinkai*)

1. Penis lobe slightly divergent, relatively short, posterolateral projection bluntly pointed, well visible in ventral view. Titilators large, oblong-shaped with parallel lateral margins..... *R. gorganica*

− Penis lobe clear divergent, relatively elongated, posterolateral projection sharply pointed, well visible in ventral view. Titilators large only at basis, curved, with convergent outline towards the apex (Fig. 10)…..... *R. loyolaea*

No differences of male imago of *R. loyolaea* "Alpine" form and *R. loyolaea* "Carpathians" form have been observed.

Mature larvae

1. Lamellae of gill VII slightly crenulated, nearly smooth with 1-3 (mainly 1-2) hardly visible shallow incisions ..... *R. gorganica*

− Lamellae of gill VII apparently crenulated, at least with 4-5 deeper, well visible incisions (Fig. 20)……..... 2

2. Lamellae of gills II-VII with 4-6 well visible incisions. In general, inner incisor elongated, oval, reaching at least a half of the length of outer one (Fig. 34). Femora with diffused central spots in pale field..... *R. zelinkai*

− Lamellae of gills II-VII with 6-15 well visible incisions (Fig. 17-20). In general, inner incisor triangular, slightly stretched laterally, reaching about one third of the length of the outer one (Fig. 16). Femora without distinct spots, only with a lighter field in darker frame.....…….. 3

3. Labrum relatively short and wide (Fig. 21). Glossae oval, massive, not pointed at the apex (Fig. 22). Subapical part of posterior margin of abdominal terga with numerous strong spines (Fig. 23). Tarsal claw without teeth (sometimes with 1 hardly visible small tooth) (Fig. 24). Spatulas on the dorsal surface of femora mainly rounded at the apex (Fig. 25)..... *R. loyolaea* "Alpine" form

− Labrum relatively elongated and narrow (Fig. 11). Glossae slightly pointed at the apex (Fig. 12). Subapical part of posterior margin of abdominal terga with not numerous small spines (Fig. 13). Tarsal claw with 1-2 teeth (Fig. 14). Spatulas on the dorsal surface of femora mainly slightly pointed at the apex (Fig. 15) .....……....……..... *R. loyolaea* "Carpathians" form

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References

Bauernfeind E., 1994. Bestimmungsschlüssel für die österreichischen Eintagsfliegen. Insecta, Ephemeroptera. Wasser und Abwasser Suppl. 4: 3-96.

Bitušík P., Novikmec M., 1997. Štruktúra makrozoobentosu Zbojského Potoka (BR CHKO Východné Karpaty). Ochrana prírody 15: 127-139. (in Slovakian).

Braasch D., Soldán T., Sowa R., 1985. *Rhithrogena bulgarica* n. sp. und zwei für die Fauna Bulgariens neue Eintagsfliegenarten (Ephemeroptera, Heptageniidae). Faun. Abhand. Staat. Mus. Tierk. Dresden 12: 125-127.

Deván P., 1991. Nové a vzácne druhy podeniek (Ephemeroptera) fauny Slovenska. Biológia, Bratislava 46: 509-511. (in Slovakian).

Godunko R. J., 2000. Little Known Species of the Genera *Rhithrogena* and *Ecdyonurus* (Ephemeroptera, Heptageniidae) from Ukraine. Vestnik Zool. Suppl. 14: 60-66.

Godunko R. J., Soldán T., 2001. Lectotype fixation and male imago distinguishing characters of *Rhithrogena gorganica* (Ephemeroptera: Heptageniidae). Klapalekiana 37: 453-460.

Grandi M., 1960. Ephemeroidea. Fauna d'Italia. Bologna: Edizioni Calderini. pp. 472.

Klapálek F., 1907. Příspěvek k znalosti chrostíků a jepic východních Karpat. Čas. Čes. spol. entomol. 4: 24-36. (in Czech).

Kłonowska-Olejnik M., 1997. Monitoring jętek (Ephemeroptera) w potokach Bieszczadzkiego Parku Narodowego. Rocz. Bieszcz. 6: 313-318. (in Polish).

Kłonowska-Olejnik M., 1997. The use of egg morphology in the taxonomy of some species of the genus *Rhithrogena* (Ephemeroptera, Heptageniidae). In: Landolt P., Sartori M., (eds). Ephemeroptera & Plecoptera: Biology-Ecology-Systematics. Fribourg: MTL. pp. 372-381.

Kłonowska-Olejnik M., 2000. Jętki (Ephemeroptera) Bieszczadów Zachodnich (Mayflies *Ephemeroptera* in the Western Bieszczady Mountains). Monogr. Bieszcz. 7: 145-155. (in Polish).

Kukuła K., 1991. Mayflies (*Ephemeroptera*) of the Wołosatka stream and its main tributaries (The Bieszczady National Park, Southeastern Poland). Acta Hydrobiol. 33: 31-45.

Kukuła K., 1995. Life cycles of selected species of mayflies (*Ephemeroptera*) of the Wołosatka and Terebowiec streams (The Bieszczady National Park, Southeastern Poland). Acta Hydrobiol. 37: 213-224.

Navás L., 1922. Efemerópteros nuevos o poco conocidos. Bol. Soc. entomol. Esp., Zaragoza 5: 54-63.

Novikmec C., Krno I., 1998. First record of *Rhithrogena gorganica* (Ephemeroptera) from Slovakia. Biológia, Bratislava 53: 188

Soldán T., Landa V., 1999. A key to the Central European species of the genus *Rhithrogena* (Ephemeroptera: Heptageniidae). Klapalekiana 35: 25-37.

Sowa R., 1971. Note sur les deux espèces de la famille (Ephemeroptera) des Carpathes polonaises Heptageniidae. Acta Hydrobiol. 13: 29-41.

Sowa R., 1975. Ecology and biogeography of mayflies of running waters in the Polish part of the Carpathians. 1. Distribution and quantitative analysis. Acta Hydrobiol. 17: 223-297.

Sowa R. 1984. Contribution a la connaissance des espèces européennes de *Rhithrogena* Eaton (Ephemeroptera, Heptageniidae) avec le rapport particules aux espèces des Alpes, et des Carpates. In: Landa V., Soldán T., Tonner M., (eds). Proc. IVth Internat. Confer. Ephemeroptera. České Budějovice: Institute of Entomology, Czechoslovak Academy of Science. pp. 37-52.

Sowa R., Soldán T., 1984. Two new species of *Rhithrogena* Eaton (Ephemeroptera, Heptageniidae) from Central Europe. In: In: Landa V., Soldán T., Tonner M., (eds). Proc. IV Internat. Confer. Ephemeroptera. České Budějovice: Institute of Entomology, Czechoslovak Academy of Science. pp. 75-84.

Thomas A., 1970. Sur la taxonomie de deux *Rhithrogena* des Pyrénées (Ephemeroptera, Heptageniidae). Annls Limnol. 6: 305-315.

Tomka I., Rasch P., 1993. Beitrag zur Kenntnis der europäischen *Rhithrogena*-Arten (Ephemeroptera, Heptageniidae): *R. intermedia* Metzler, Tomka & Zurwerra, 1987 eine Art der *alpestris*-Gruppe sowie ergänzende Beschreibungen zu fünf weiteren *Rhithrogena*-Arten. Mitt. Schweiz. entomol. Ges. 66: 255-281.

Zabric D., Sartori M., 1997. First contribution to the mayfly fauna from Slovenia (Ephemeroptera). In: Landolt P., Sartori M., (eds). Ephemeroptera & Plecoptera: Biology – Ecology - Systematics. Fribourg: MTL. pp. 147-151.

Zelinka M., 1953. K poznání jepic (Ephemeroptera) Vysokých Tater. Spisy Přír. fak. Masaryk Univ. 348: 157-167. (in Czech).

Zelinka M., 1980. Řád Jepice – Ephemeroptera. Klič vodních larev hmyzu. Praha: Československá akademie věd. pp. 39-67. (in Czech).