

ECDYONURUS NIGRESCENS (KLAPÁLEK, 1908) (EPHEMEROPTERA: HEPTAGENIIDAE) – NEOTYPE DESIGNATION, TAXONOMICAL AND NOMENCLATURE NOTES

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Abstract.— The neotype of *Ecdyonurus nigrescens* (Klapálek, 1908) has been designated, basing on male imago reared from larva, collected in the Chornohora Range (the Ukrainian Carpathians). The male imago of this species is redescribed; male subimago, female imago and subimago, larvae and the structure of egg chorion are described and illustrated for the first time. Critical diagnostic characters distinguishing this species from other representatives of the *Ecdyonurus helveticus* species-group are discussed. The type locality is defined in accordance with the Article 76.3 and Recommendation 76A of ICZN as follows Ukraine: Zakarpattia Region, Carpathian Biosphere Reserve, the Chornohora Range, Polonyna Brebeneska district, mountain valley between SW slope of Gutyn Tomnatek Mt. and NW slope of Brebeneskul Mt., upper section of the Brebeneskul stream (left-bank tributary of the Hoverla stream, Tysa river-basin), 1450 m. a.s.l. The data on the original syntype series and type locality are discussed in detail. All known specimens of *E. nigrescens* collected by J. Dziędziewic between 1908 and 1910 are observed and reidentified. The data on distribution, ecology and life cycle of *E. nigrescens* are presented.



Key words.— Ephemeroptera, Heptageniidae, *Ecdyonurus*, *E. nigrescens*, neotype, type locality, synonymy, description, distribution, ecology, Ukrainian Carpathians.

INTRODUCTION

The description of the species *Heptagenia nigrescens* Klapálek, 1908 was published by František Klapálek in co-authorship with Józef Dziędziewic in two papers. The first one was published at the beginning of 1908 in the volume 5 of the “Časopis České Společnosti Entomologické” (Journal of the Czech

Entomological Society), and included a brief description of the species in Latin, description in detail in Czech and outlined drawing of male genitalia (styliger, forceps base and penes) (Dziędziewic and Klapálek 1908a: 24, Fig. 3). The second paper, which was published at the end of 1908 in “Kosmos” (Journal of the Polish Naturalists Society), included the same picture, the Latin and Polish versions of the previous Czech

text with reference to the first paper (Dziędziewic and Klapálek 1908b: 255–256, Fig. 3).

In those two papers *E. nigrescens* was originally described by F. Klapálek from the Chornohora Range of the Eastern Carpathians (“Czarnohora” in author’s spelling), basing on the adult male collected by J. Dziędziewic in September 14, 1907. In 1908 and 1910 J. Dziędziewic found this species in other localities within the Chornohora Range. In his last summarized paper, he gave the previous description in Polish and the picture of *E. nigrescens* without any changes or supplements, stating more detailed information on its distribution in the Eastern Carpathians (the area of present Ukrainian territory) and peculiarities of ecology of male imagoes (Dziędziewic 1919: 120–121, Fig. 4a).

Mikulski (1936: 86–87) for the first time attributed *E. nigrescens* into the genus *Ecdyonurus* Eaton, 1868. A brief characteristics of male imago included in his paper wasn’t accomplished by any drawings and was grounded on the previous descriptions of F. Klapálek and J. Dziędziewic. Besides, in the key to identification of male imagoes, Mikulski (1936: 84) has ascertained the details of styliger structure evidently based on the material studied by J. Dziędziewic from the collection of the Museum of Physiographical Commission of the Academy of Sciences and Letters, Kraków, in 1927 (see Kłowska-Olejnik *et al.* 2005).

Puthz (1975: 323) has studied 5 specimens of male imagoes, labeled as “Czarnohora. Dancerz. 16.IX.1910” from the collection of Georg Ulmer (Zoologisches Museum, Universität Hamburg), which were passed to latter by J. Dziędziewic and were designated as “*Heptagenia nigrescens* Klap.” V. Puthz has considered them as those, belonging to the species *E. picteti* (Meyer-Dür, 1864), and on this basis has indicated *E. nigrescens* as a junior synonym to *E. picteti*. This author has also noted, that in spite of subimagoes *E. nigrescens* being unknown (in his publication in 1975 the females of *E. picteti* were designated as lectotype and paralectotype), the available knowledge on the matter had allowed to consider this taxon as a synonym. In further papers dealt with the taxonomy of *E. helveticus* species-group, this point of view was either accepted (Jacob and Braasch 1984: 53) or *E. nigrescens* wasn’t mentioned at all (Hefti and Tomka 1986, Hefti *et al.* 1989, Belfiore and Buffagni 1994).

The objectives of the present paper are as follows: (1) to make a revision of previously published data on the species *E. nigrescens* and to get the information on the number of specimens collected by J. Dziędziewic and their housing; (2) to re-determine J. Dziędziewic’s specimens designated as „*Heptagenia nigrescens*”; (3) to designate neotype and to define a type locality of *E. nigrescens*; (4) to describe eggs, larvae and reared subimagoes and imagoes of both

sexes, originated from different localities of the Chornohora Range, including those, where the syntypes of this species have been collected; (5) to determine a combination of critical distinguishing characters of the male imagoes, larvae and eggs with a view to separate this species from other representatives of the *E. helveticus* species-group; (6) to present the data on distribution and ecology of *E. nigrescens* in the Chornohora Range (the Eastern Carpathians, Ukraine).

Type series

F. Klapálek (Dziędziewic and Klapálek 1908a, 1908b) has not designated the holotype specimen of *E. nigrescens*. Moreover, in both published papers any information about the number of specimens collected, unfortunately, is absent. Thus, the size of type series, that is the number of syntypes of *E. nigrescens* (these specimens present syntypes according to the ICZN Article 73.2), is unknown.

The description of F. Klapálek was based on study of male imago, although in the original text the definition “males” was not precise. This conclusion is confirmed by morphometric proportions of fore tarsal segments given by the author, who is typical of male imago of the genus *Ecdyonurus*, and also the description of body and wing colors.

The information on collecting localities of male imago syntypes somewhat differs in two papers (Dziędziewic and Klapálek 1908a, 1908b). In the first one (in Časopis České Společnosti Entomologické) F. Klapálek gave only brief information: “Czarnohora, potoki z Dancerza, 14 IX. 1907, leg. Dziędziewic” [the Chornohora Range, the streams running from Dantsezh Mt.], which is the same as in original labels by J. Dziędziewic in Polish (Dziędziewic and Klapálek 1908a: 24). In the Polish text of the second paper (in “Kosmos”) the data on the collecting locality of syntypes are specified by J. Dziędziewic himself: “Spotkane dnia 14. września 1907, pojedynczo i w małej ilości osobniki unosiły się nad potokiem, w wąwozie między Dancerzem a Pożyżewską na Czarnohorze w wysokości po nad 1.300 m. n.p.m. wśród kosodrzewiny” [Was found on the 14th of August, 1907, in a few number of specimens flying over the stream in a glen between Dantsezh Mt. and Pozhyzhevs’ka Mt. of the Chornohora Range at the height of 1300 m. a.s.l. among the brushwood of mountain pine] (Dziędziewic and Klapálek 1908b: 256). Beyond doubt, the question is about the Orendar stream (“Arendarski”, see Dziędziewic 1919: 121), a right-bank tributary of the Upper Prut river, running in a glen between Dantsezh Mt. and the Pozhyzhevs’ka Mt. (“Dancerz” and “Pożyżewska”, in old polish papers and maps) of the Chornohora

Range. Thus, according to the ICZN Article 76.1, Recommendations 76A.1.2 and 76A.1.3 all male imagoes collected by J. Dziędziewicz on the 14th of August, 1907 over the Orendar stream in a glen, mentioned above, at the altitude of 1300 m. a.s.l. (the Chornohora Range, the Eastern Carpathians, Ukraine) can be regarded as syntypes of the species *E. nigrescens*.

Dziędziewicz (1919: 121) noted the distribution of *E. nigrescens* within height ranges of 1300 m. a.s.l. and 1400 m. a.s.l. (a question is about altitudes, where male imagoes were collected). Besides the type locality, this species was found "... na górnym biegu potoków: Siklawy pod Breskulem ..." [in upper current of the Siklava stream at the foot of Breskul Mt.] (see also Godunko 1999). However, a stream named "Siklawka" has been absent from both modern and old Polish and Hungarian maps of the Chornohora Range. Evidently, the author by the name "Siklawka" (= waterfall) has erroneously indicated the Prutchyk Zarosliatskyi stream, which has the largest waterfall of glacial origin in the Chornohora Range.

The study of F. Klapálek's collection at the National Museum, Praha in September 2000, and its re-study in November 2006, unfortunately has failed in revealing any other specimen, which could be regarded as syntype of *E. nigrescens*. Moreover, none specimen of this species collected by J. Dziędziewicz in the Eastern Carpathians has been found in Praha.

The information on availability of specimens designated as "*Heptagenia nigrescens*" in collections of different scientific institutions can be generalized as follows. Puthz (1975) has reported the availability of five specimens of male imagoes in the collection of G. Ulmer at the Zoologisches Museum der Universität Hamburg (ZMUH), which were collected three years later than the syntype series in the same locality. At our request in October 2006, Prof. Hans Strümpel (Zoologisches Museum der Universität Hamburg) has looked through that collection for the second time and found only three male imagoes (in alcohol) from those mentioned in the Puthz's paper (1975). In other parts of G. Ulmer's collection, housed at the Museum and Institute of Zoology, Polish Academy of Sciences, Warszawa (looked through by us in April, 2002) and at Institut für Systematische Zoologie bei dem Museums für Naturkunde der Humboldt-Universität Berlin (looked through by us in November 2004) none specimen of this species is present.

Godunko (1999) has found seven male imagoes designated as "*Heptagenia nigrescens*" and collected by J. Dziędziewicz in 1908 and 1910 in different localities of the Chornohora Range (including the collecting place of syntypes), in the collection of the State Museum of Natural History, National Academy of Sciences, Lviv (SMNH). Two of them are labeled as those

from the G. Ulmer's collection, and one specimen was collected in 1910 in Vorokhta village [Worochta] in a distance of 16 km from Dantsezh Mt.

Any specimens labeled as "*Heptagenia nigrescens*" are lacking in the collection of the Museum of Natural History of the Institute of Systematics and Evolution of Animals, Polish Academy of Sciences, Kraków (formerly the Museum of Physiographical Commission of the Academy of Sciences and Letters, Kraków) where a part of J. Dziędziewicz's collection has been housed. However, Kłonowska-Olejnik *et al.* (2005) have reported of availability of a damaged specimen of male imago labeled as "Czarnohora. Dancerz 12.IX.1909." and "6/24", which was identified by them as *E. picteti* (see below).

Grounding on the detailed analysis of the collection materials available, we can state that all syntypes of *E. nigrescens* have been lost.

Synonymy of *E. nigrescens* and *E. picteti*

The original description of *E. nigrescens* by F. Klapálek is brief enough and doesn't contain sufficient information on the critical morphological characters of male imago. Moreover, an outlined picture of *E. nigrescens* male genitalia made by this author was presented in all three papers (Dziędziewicz and Klapálek 1908a, 1908b, Dziędziewicz 1919), hitherto has been the only known depiction of these structures.

The synonymy of species *E. nigrescens* and *E. picteti* was stated by Puthz (1975) on the basis of the analysis of dried pinned specimens not belonging to the syntype series. Additionally, this author has regarded the species *E. picteti* and *E. austriacus austriacus* Kimmins, 1958 as conspecific ones and proposed to synonymize them under the name *E. picteti*. However, Jacob and Braasch (1984) have stated that a question is about two taxa, which clearly differ both in adults and larvae. Later on, this statement has been confirmed by the data on morphological and biochemical study, carried out by Hefti and Tomka (1968) and Hefti *et al.* (1989), who analysed the specimens of both species, originated from the type locality of *E. austriacus austriacus* in the Bodingbach stream near Lunz, Austria. In spite of this, such a material collected in Czech Republic during last 30 years, was treated according to V. Puthz's opinion as *E. picteti*. On the other hand, during a long time in regional and local references the species *E. silvaegabretae* Soldán et Godunko, 2006, recently described from the Šumava Mountains in the Czech Republic, has been currently confused with *E. austriacus austriacus* and *E. picteti* (Soldán and Godunko 2006).

The recent research in *E. helveticus* species-group has resulted in the description of new species (adults and larvae being examined) from the Ukrainian Carpathians, namely *E. austriacus nataliae* Godunko et Klonowska-Olejnik, 2004 and *E. rizuni* Godunko, Klonowska-Olejnik et Soldán, 2004 (Godunko and Klonowska-Olejnik 2004, Godunko *et al.* 2004). However, because of insufficient knowledge on the taxonomy of *E. nigrescens* and lack of information on its critical distinguishing characters (both in adults and larvae), it is not excluded, that some new preconceived assumption may arise about possible synonymy of this and other representatives of *E. helveticus* species-group, recently described from the Eastern Carpathians. Besides, some faunistic papers, dealing with mayfly fauna of the Ukrainian Carpathians and adjacent area (Gáldean 1999: 424, Cser and Andrikovics 2001: 347, Derka 2003: 25), include information on taxa belonging to the *E. helveticus* species-group (e.g., “*E. cf. helveticus*”, “*E. cf. austriacus*”, “*E. cf. picteti*”, “*E. parahelveticus*” and “*E. cf. zelleri*”) which taxonomic status needs more accurate definition.

The precise analysis of all available specimens of male imagoes *E. nigrescens*, collected by J. Dziędzielewicz between 1908 and 1910, and study of new rich material on larvae and imagoes from the Chornohora Range, collected by us between 2002 and 2006, has shown clear difference of this species not only from *E. picteti*, but also from other species of the *E. helveticus* species-group. Beyond all question, *E. nigrescens* is a valid species name of a taxon belonging to the genus *Ecdyonurus* s. str.

The taxonomic problems within the *E. helveticus* species-group have not been entirely settled, especially in the case of species occurring in mountains and foot-hills of the Eastern and South-Eastern Europe. That is why, the determination of a proper taxonomic status of *E. nigrescens* and its possible synonymy with other species of the group and defining of critical distinguishing characters of its adults and larvae with respect to other close species will essentially contribute to the knowledge on diversity and biogeography of all taxa of the *E. helveticus* species-group.

Designation of neotype and type locality

There is obvious necessity to designate neotype of nominal taxon *E. nigrescens* and it is stipulated by following: (1) loss of syntypes; (2) availability of previous conclusions about its status as junior synonym to *E. picteti*; (3) shortage of existing descriptions and requirement in objective definition of critical distinguishing characters.

Neotype: male imago (reared from larva with preserved larval and subimaginal skins; genitalia on slide), Ukraine, Zakarpattia Region, Carpathian Biosphere Reserve, the Chornohora Range, Polonyna Brebeneska district, mountain valley between SW slope of Gutyn Tomnatek Mt. and NW slope of Brebeneskul Mt., upper section of the Brebeneskul stream (left-bank tributary of the Hoverla stream, Tysa river-basin), 1450 m. a.s.l. (48°4'37"E, 24°33'16"N) 23.07.2002, R. J. Godunko and M. Klonowska-Olejnik leg.

The neotype has been designated as such by R. J. Godunko and M. Klonowska-Olejnik in March, 2008 and housed at the State Museum of Natural History, National Academy of Sciences of Ukraine, Lviv, Ukraine, under the No E1.05.04.08/01 in SMNH Catalog.

We define the **type locality** of *E. nigrescens* (Klapálek, 1908) according to the Article 76.3 and Recommendation 76A of ICZN as follows: Ukraine, Zakarpattia Region, Carpathian Biosphere Reserve, the Chornohora Range, Polonyna Brebeneska district, mountain valley between SW slope of Gutyn Tomnatek Mt. and NW slope of Brebeneskul Mt., upper section of the Brebeneskul stream (left-bank tributary of the Hoverla stream, Tysa river-basin), 1450 m. a.s.l. (48°4'37"E, 24°33'16"N).

We consider it impossible to designate one of the specimens, collected by J. Dziędzielewicz between 1908 and 1910, as neotype, because of its substantial damage during keeping on pin for almost 100 years. Using of this specimen as nomenclature type of nominal taxon *E. nigrescens* could in future complicate determination of taxonomic identity of the species and bring to incorrect conclusions concerning its taxonomic status.

The specimen of male imago is designated as the neotype, being reared from larva in the Brebeneskul stream at the altitude of 1450 m. a.s.l. (48°4'37"E, 24°33'16"N). The type locality is situated in Transcarpathian section of the Chornohora Range at a distance 7.5–8.0 km from the original type locality in the slopes of the Dantsezh Mt. (ICZN, Article 75.3.6).

MATERIAL AND METHODS

E. nigrescens. The collection of the State Museum of Natural History, National Academy of Sciences of Ukraine, Lviv, Ukraine (SMNH): 2 male imagoes labeled as “Czarnohora. Pożyżewska – Breskul. 11. – 9. – 1908.”, “nigrescens Kl.”, No E1.05.04.08/02-03; male imago “Czarnohora. Potoki z Dancerza. 29/9. – 1908.”, No E1.05.04.08/04; male imago “Czarnohora Breskul pot. Siklawka. 14.IX.1910.”, “H. nigrescens Kl.” No E1.05.04.08/05; male imago “Worochta. 20.IX.1910.”, No E1.05.04.08/06; 2 male imagoes “Czarnohora. Dancerz. 16.IX.1910.”, No E1.05.04.08/07-08.

The neotype: male imago (reared from larva with preserved larval and subimaginal skins; genitalia on slide), Ukraine, Zakarpattia Region, Carpathian Biosphere Reserve, the Chornohora Range, Polonyna Brebeneska district, mountain valley between SW slope of Gutyn Tomnatek Mt. and NW slope of Brebeneskul Mt., upper section of the Brebeneskul stream (left-bank tributary of the Hoverla stream, Tysa river-basin), 1450 m. a.s.l. (48°4'37"E, 24°33'16"N), 23.07.2002, R. J. Godunko and M. Kłonowska-Olejnik leg., No E1.05.04.08/01.

3 male imagoes, male subimago, female imago, female subimago (reared from larvae with larval and subimaginal skins; genitalia on slides), ibidem, 1400 m. a.s.l. (48°4'32"E, 24°32'54"N), 27.07.2002, R. J. Godunko and M. Kłonowska-Olejnik leg., No E1.05.04.08/09-14; 35 mature larvae, ibidem, 1400–1450 m. a.s.l., 27.07.2002, R. J. Godunko and M. Kłonowska-Olejnik leg., No E1.05.04.08/15-19; 27 mature larvae, Ukraine, Zakarpattia Region, Rakhiv district, N vicinity of Kvasy village, Keveliv stream (left-bank tributary of the Tysa river), 800 m. a.s.l. (48°10'39"E, 24°20'51"N), 09.08.2006, R. J. Godunko leg., No E1.05.04.08/20-23; 12 larvae, Ukraine, Zakarpattia Region, Rakhiv district, Bilyn village, W slope of Shtev'era Mt., small unnamed stream (left-bank tributary of the Tysa river), 720 m. a.s.l. (48°30'36"E, 24°14'53"N), 09.08.2006, R. J. Godunko leg., No E1.05.04.08/24-25.

The collection of the Department of Hydrobiology, Institute of Environmental Sciences, Jagiellonian University, Kraków, Poland (DH IES): male imago (reared from larva with larval and subimaginal skins; genitalia on slides), the same locality and date as neotype, R. J. Godunko and M. Kłonowska-Olejnik leg.; 2 male imagoes, 2 male subimagoes, 2 female subimagoes (reared from larvae with larval and subimaginal skins; genitalia on slides), ibidem, 1400 m. a.s.l., 27.07.2002, R. J. Godunko and M. Kłonowska-Olejnik leg., 59 mature larvae, ibidem, 1400–1450 m. a.s.l., 27.07.2002, R. J. Godunko and M. Kłonowska-Olejnik leg.

The collection of the Museum of Natural History of the Institute of Systematics and Evolution of Animals, Polish Academy of Sciences, Kraków, Poland (MNH ISEA): male imago labeled as "Czarnohora. Dancerz. 12 IX.1909.", "6/24" (genitalia and right fore wing on slide No 119).

The collection of the Zoologisches Museum der Universität Hamburg (ZMUH): 3 male imagoes labeled as "Czarnohora. Dancerz. 16.IX.1910."

E. austriacus austriacus. The collection of the Naturhistorisches Museum Wien (NMW): male imago, 3 female imagoes labeled as "Rotmoosgraben, 1100 m, 19/9/44, F 159", "Ecdyonurus "austriacus" F 159 det. Pleskot 1956."; female subimago, 12 larvae, 4 larval skins "F 158", "Ecd. helveticus 1 ♀ 5 im det. Pleskot

1956", "Rotmoosbach 19/9/44"; male imago, larva "Ecd. "austriacus" F 161 det. Pleskot 1956", "Oberseeznfließ 19/9/44"; 2 female imagoes, female subimago "Ecd. helveticus (nür ♀ ♀) det. Pleskot 1956", "Jbbs, Ois. Klaise, 26.8.55 Zücht"; 5 larvae "Pomeils", "Ecd. austriacus Hernalus, 28. Juli 57"; 3 female imagoes "Ecd. helveticus X 440 det. Pleskot 1956" "X 440"; male imago, 4 female imagoes "X 238", "X 517b", "Ecdyonurus "austriacus" 1 ♂ Rest? det. Pleskot 1956", "X 238+X 517b"; male imago "Ecd. "austriacus" K 179 det. Pleskot 1956", "K 179"; 2 male imagoes, female imago "X 289, 1 ♀", "X 444b" "Ecdyonurus helveticus ♂ X 533"; male imago "Ecd. helveticus X 537 det. Pleskot 1956", "X 537"; 3 male imagoes "E. helveticus Etn. det. Pleskot 1952", "T 232"; male imago "T 228-229"; male imago "41/88a, Karner Alm, sbg. 2000 m. 6.8.88"; larva "41/88c, 6.8.88, Karner, Alm."; 3 mature female larvae "22./89, Landach, Onellbach 1.9.89" (from these specimens the eggs for SEM study are obtained); male imago "Lunz/see Kotberg 18.6.94, leg Kust"; male imago (genitalia only preserved) "Rothwald, Urwald 21.7.98 Ressler leg. N°2"; male imago (genitalia only preserved) "Gaming, Nö Urwald Rothwald 8.8.98. Ressler leg. N°3"; male imago "Molln, Breitenau Hausbach NP Kalkalpen 6.8.2001"; male imago „Molln, Breitenau 6.8.2001 OÖ, Wimmer leg."; male imago (genitalia only preserved) "Steinbach Lunz/see, Nö oktober 2001, W. Graf leg."; male imago (genitalia and fore wing only preserved) "Tschalgraben Nö 14.6.2004, Graf leg."

All specimens have been preserved in 70–80% alcohol (except one dried pinned specimen from the collection of MNH ISEA). The larval and imaginal material of *E. nigrescens* from Zakarpattia Region was collected between 2002 and 2006 and has been housed in SMNH and DH IES.

Drawings were made by means of binocular Olympus SZX7 with camera Lucida (Olympus SZX-DA) and Olympus GHG 12345/X700. Photographs were made using a Leica MZ FL III microscope with photo camera Leica DC 200.

Permanent slides were prepared to transfer the objects directly into Liquid de Faure. For the scanning electronic microscopy examination, the eggs and larval structures were dehydrated in the ethyl alcohol series, and critical-point dried using CO₂ in a Bomar apparatus, mounted with double-sided tape on SEM stubs and coated with carbon-gold. They were observed in a JEOL JSM 7401F and HITACHI S-4700 scanning electronic microscopes. The SEM photographs of the eggs and larval structures were taken in the Department of Electron microscopy of Biological Faculty of the University of South Bohemia (Ěeské Budějovice, Czech Republic) and in the Department of Cytology and Histology of the Jagiellonian University (Kraków, Poland).

DESCRIPTION

Heptageniidae Needham, 1901

Ecdyonurus Eaton, 1868 s. str.*Ecdyonurus nigrescens* (Klapálek, 1908)
(Figs 1–34)

Heptagenia nigrescens Klapálek, 1908: *Dziędzielewicz*, Klapálek, 1908a, *Časopis České Společnosti Entomologické*, 5(1): 24, Fig. 3.

Heptagenia nigrescens: *Dziędzielewicz*, Klapálek, 1908b, *Kosmos*, 33(4–5): 255–256, Fig. 3; *Dziędzielewicz*, 1919, *Rozprawy i Wiadomości Muzeum Dzieduszyckich*, 3(3–4): 120–121, Fig. 4a.

Ecdyonurus nigrescens: Mikulski, 1936, *Fauna słodkowodna Polski*, 15: 84, 86–87.

Ecdyonurus picteti: partim, nec Meyer-Dür, 1864; Puthz, 1975, *Revue Suisse de Zoologie*, 82(2): 321–324; Jacob, Braasch, 1984, *Entomologische Abhandlungen und Berichte Staatliches Museum für Tierkunde Dresden*, 48(5): 53; Godunko, 1998, *Proceedings of the State Natural History Museum*, 14: 99; Godunko, 1999, *Roczniki Bieszczadzkie*, 8: 396; Godunko, 2000, *Proceedings of the State Natural History Museum*, 15: 161; Kłonowska-Olejnik, Godunko, Pawłowski, 2005, *Acta zoologica cracoviensia*, 48B (1–2): 186.

Male imago. Size. Body length: 11.3–14.5 mm; fore wings length: 12.5–14.0 mm; cerci length: 22.8–28.2 mm.

General coloration of body dark, brown to dark brown with some light brown spots on the lateral sides of body, and distinct black spots dorsally.

Head brown to dark brown. Facial keel with unclear smudges, generally dark brown. Antennae brown, scapus and pedicels darker than flagellum. Ocelli white apically, brown to dark brown basally. Apical part of eyes milky-greyish, basal part greyish-black.

Thorax brown to dark brown, occasionally dorsal side dark brown to black. Pronotum unicolorous brown. Mesonotum of the specimens reared from larvae in 2002 and later preserved in 75% alcohol with light transversal spots on anteronotal protuberance and scutoscuteellar impression, longitudinal spots around the lateroparapsidal sutures and narrow light area around the medioparapsidal suture (morphological terminology and nomenclature of thoracic structures used here follows that by Kluge 1994, 2004). In dried pinned specimens (examined in MNH ISEA, 2006 and SMNH, 1999) mesonotum almost unicolorous dark brown to black, only with unclear light area around medioparapsidal suture. Thoraxal sterna brown to dark brown and have the structure typical of the genus *Ecdyonurus*.

Fore wings hyaline and transparent, light brown to dark brown, darker at base. Pterostigmatic area opaque, yellowish to yellowish-brown. Hind wings of the same color as fore ones.

Fore legs distinctly darker than middle and hind ones. Fore femora and tibiae unicolorous brown to dark brown. Fore tarsi unicolorous brown. Middle and hind legs with light brown femora and tibiae; tarsi brown, occasionally dark brown to black. Surface of femora with unclear garish maculation.

Abdomen brown. Lateral sides of terga II–VIII with roundish or triangular spots surrounded with an L-shape drawing typical of species of the *Ecdyonurus helveticus* species-group. Terga I, IX and X unicolorous brown with some reddish maculation. Terga II–VIII with pair of longitudinal triangular spots near anterior part of segment. Sterna slightly paler than terga, brown to reddish-brown. Sterna I–VIII with two pairs of light spots centrally. Nerve ganglia dark, well visible. Cerci

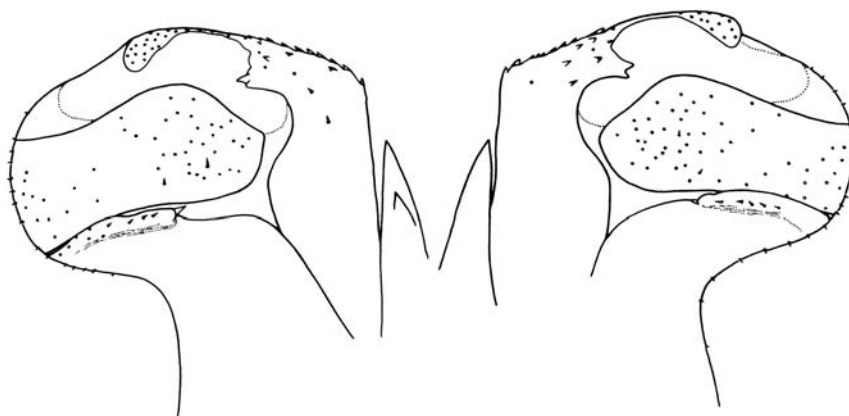


Figure 1. *Ecdyonurus nigrescens* (Klapálek, 1908), male imago, Brebeneskul stream, Chornohora Range, Ukrainian Carpathians: penis lobes dorsally.

brown to dark brown. Basal portion of cerci (approximately $\frac{1}{4}$ of their length) darker than distal portion.

Styliger dark, yellowish-brown to dark brown. Forceps base with two slightly asymmetrical protuberances (Fig. 7). Forceps light brown to dark brown, darker at the base. Penis lobes pale, yellowish-brown to light brown, not elongate, slightly stretched laterally (Figs 1–6, 8–9). External part of the penis lobes relatively roundish. Apical sclerite not massive, roundish apically. Straight lines of apical sclerite forming distinctly obtuse angle mean 110° (min. = 107° ; max. = 113.5°). Inner denticulation well developed, with 1–3 strong teeth and 1–2 small subapical spines directed approximately towards the middle of the lobe, perpendicular to the axis of their symmetry or sub-parallel to margins of apical sclerite (Fig. 1). Apex of apical sclerite distinctly projecting. Lateral sclerite well developed, large, without deep narrowing, tapered proximally. The wideness part of lateral sclerite situated approximately in $\frac{2}{3}$ of its length; the tip of lateral sclerite relatively broad. Basal sclerite relatively massive with 1–2 strong teeth directed perpendicularly to axis of symmetry of the penis lobe. Lobes' surface covered with numerous small spines. Penis stem without hump. Titilators light brown, pointed at apex (Figs 1–6, 8–9).

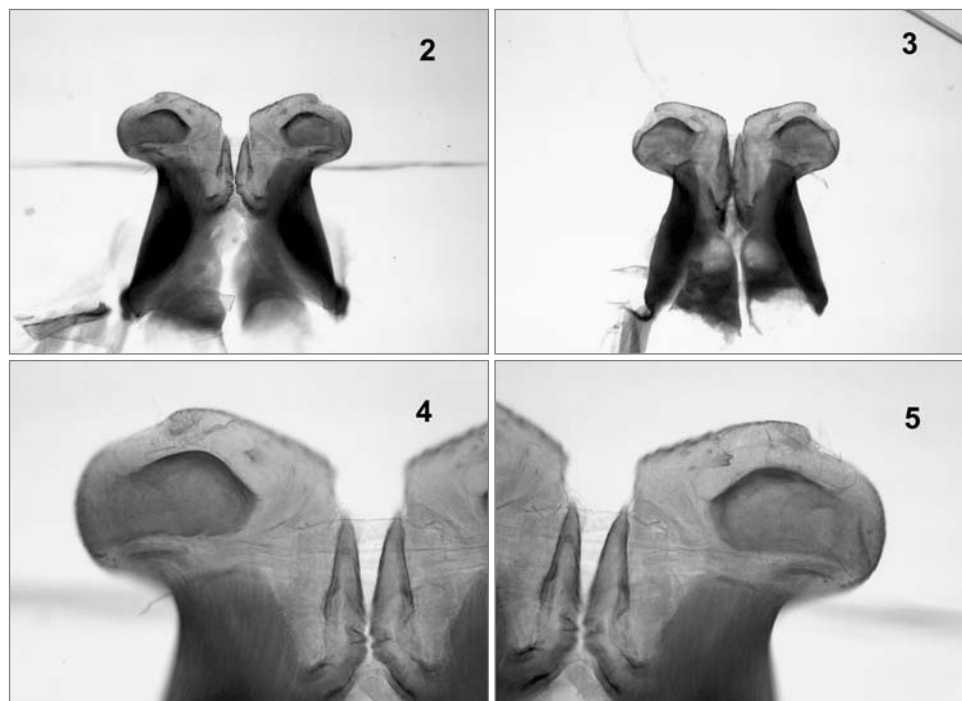
Male subimago. Size. Body length: 10.0–11.4 mm; fore wings length: 11.6–13.0 mm; cerci length: 11.4–12.0 mm.

General color of body pale, yellowish-brown to brown, with yellowish smudges and reflections. The specimens at once after transformation from larva yellow to yellowish-brown, occasionally yellowish-white.

Head and facial keel pale, yellowish, with some greyish smudges. Antennae unicolorous light brown. Basal part of ocelli greyish-black, apical part white.

Thorax yellowish-brown to light brown, with some brown to dark brown bands. Pronotum light brown with greyish reflections. Medioscutum pale, yellowish-brown, with two longitudinal brown bands between medioparapsidal and lateroparapsidal sutures. Anterior part of mesonotum darker than posterior part, light brown to brown, with brown to dark brown transversal spots around mesonotal suture. Lateral sides of thorax yellowish-white to light brown with brownish spots around the thoracic sutures. Sterna yellowish-white to yellowish-brown. The margins of basisternum and furcasternal protuberances with narrow brown band. Basisternum of prothorax brown. Metanotum unicolorous brown.

Fore wings opaque, yellowish-grey to light brown. Distinct zigzag-shaped drawing formed by pointed transversal light brown bands is present. Pterostigmatic area yellowish-brown. Hind wings with central transversal yellowish-grey band, other part light brown. Wing venation brownish.



Figures 2–4. *Ecdyonurus nigrescens* (Klapálek, 1908), male imagoes, Brebeneskul stream, Chornohora Range, Ukrainian Carpathians: (2–3) penis dorsally; (4) right penis lobe dorsally; (5) left penis lobe dorsally.

Fore legs darker than middle and hind ones, unicolorous brown, occasionally fore femora with dark brown smudges. Middle and hind legs unicolorous light brown.

Abdominal terga light brown to brown. Terga VI–X darker than other ones, intensively brown. Two well visible longitudinal light spots situated in anterior part of terga II–X. Lateral sides of terga with *L*-shaped drawing around unclear reddish-brown smudge. Sterna light brown with two pairs of spots and well visible nerve ganglia. Cerci brown, slightly paler at the tip.

Styler and forceps segments light brown to brown, last segment of forceps slightly paler. Forceps bases with two asymmetrical lateral protuberances (Fig. 11). Penis lobes light brown, slightly darker at the bases. Lobes not stretched laterally, apparently spherical, with nearly rounded external margin (Figs 10, 12, 13). Apical sclerite blunt at the tip, with 1–2 strong inner teeth. External margin with numerous spines. Lateral sclerite the same structure as in male imago. Basal sclerite relatively massive, with 1–2 strong teeth. Penis stem without hump, titilators small, bluntly pointed apically (Figs 12, 13).

Female imago. Size. Body length: 12.3–15.7 mm; fore wings length: 14.5–16.0 mm; cerci length: 13.6–16.5 mm.

General color of body paler than in male imago, yellowish-brown to light brown, with distinct brownish-red colored abdominal terga.

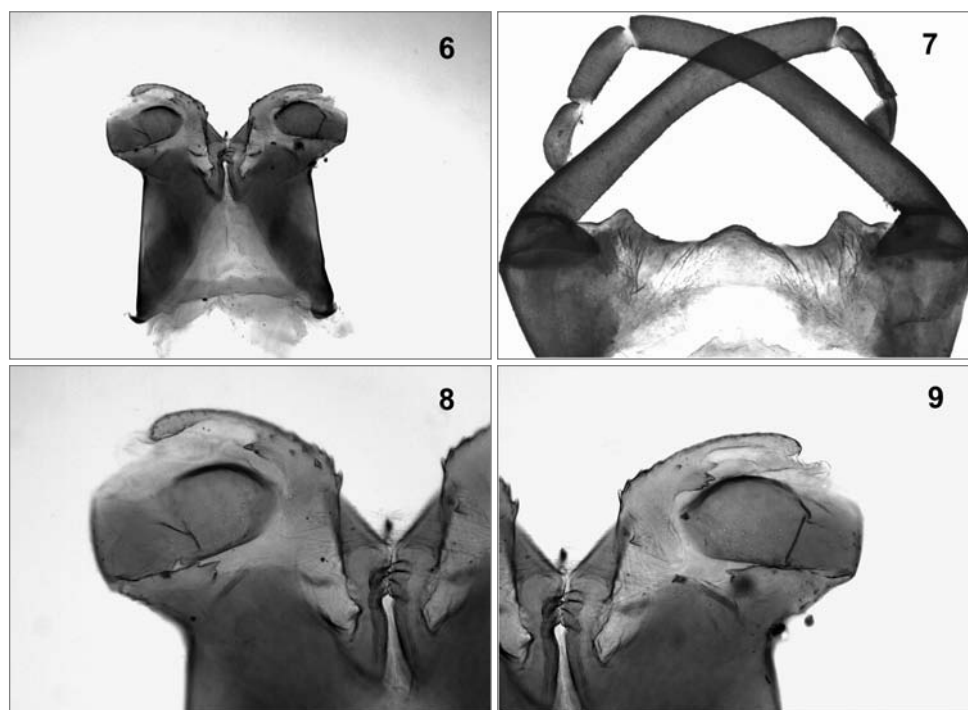
Head yellowish-brown to yellowish-grey, with unicolorous greyish smudges on facial keel. Antennae unicolorous brown. Eyes greyish-black. Basal part of ocelli black, apical part whitish-grey.

Thorax pale, yellowish-brown to light brown: anteronotal protuberance and area around the mesonotal suture are brown; metascutum generally yellowish-brown with darker area between medioparapsidal sutures and median longitudinal suture; posterior scutal protuberances distinctly brown. Metanotum intensively brown with unclear dark smudges. Lateral sides of thorax yellowish-white to yellowish-brown. Ventrally thorax yellowish-brown, with brown furcasternal protuberances.

Fore legs darker than middle and hind ones, distinctly brown with unclear greyish and reddish smudges on distal part of fore femora. Middle and hind legs with yellowish-white to yellowish-brown femora and tibiae, and slightly darker tarsi.

Fore and hind wings hyaline, transparent, with yellowish-brown to brown venation. Pterostigmatic area of fore wings opaque, yellowish brown to milky.

Abdomen pale, light brown, with distinct reddish spots on each segment. Terga with three longitudinal whitish spots dorsally, well visible on the segments II–VII. Lateral sides of terga with *L*-shaped drawing surrounded by a reddish spot. Sterna yellowish-brown. Nerve ganglia dark. Subgenital plate relatively wide, its



Figures 6–9. *Ecdyonurus nigrescens* (Klapálek, 1908), male imago from collection of the MNH ISEA labeled as “Czarnohora. Dancierz. 12 IX.1909.”, “6/24”: (6) penis dorsally; (7) forceps structure ventrally; (8) right penis lobe dorsally; (9) left penis lobe dorsally.

posterior margin with smooth rectangular contour. Subanal plate pointed apically. Cerci light brown to brown, distinctly darker at the base.

Female subimago. Size. Body length: 10.0–12.5 mm; fore wings length: 12.0–13.2 mm; cerci length: 12.7–13.8 mm.

General coloration similar to that in male imago, especially the peculiarities of thorax coloration. Shape of subgenital and subanal plate similar to that in female imago.

Mature larva. Size. Body length: 12.3–14.0 mm; cerci length: 8.7–10.3 mm.

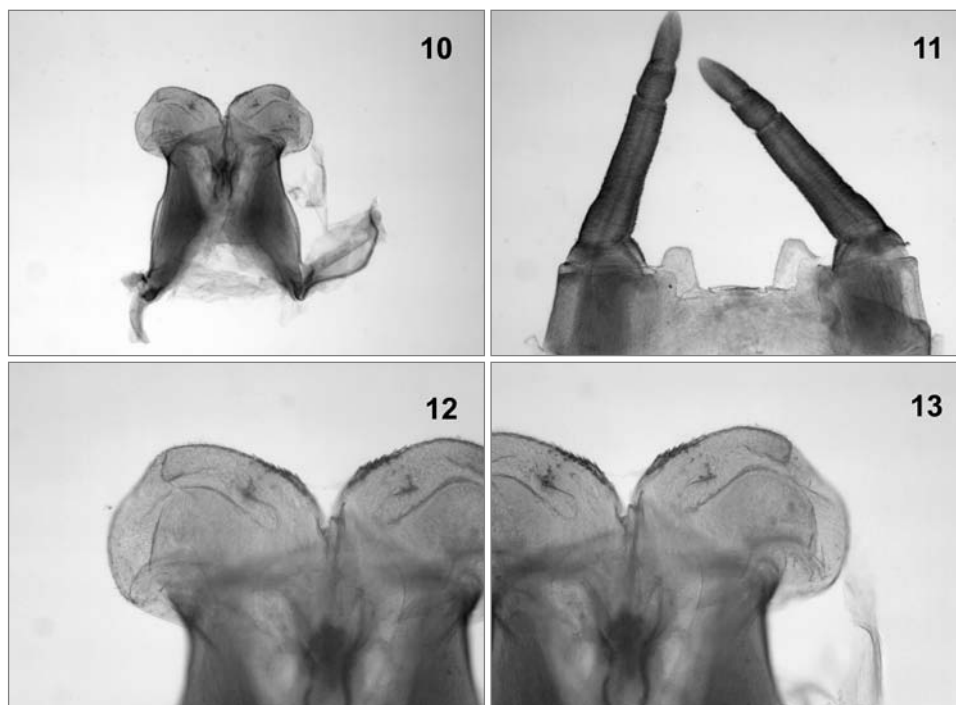
General coloration of body pale, yellowish-white to brown.

Head generally yellowish-brown to brown. Head relatively wide, with rounded anterior margin. Antennae with brownish scape and pedicel; flagellum yellowish-white. Eyes and ocelli black. Labrum relatively narrow with slightly curved lateral lobes (Fig. 16). 14–19 bristles arranged in two rows are present on ventral surface of labrum; the first 2–5 bristles more robust than the others (Fig. 17). First segment of maxillary palps with 7–20 tiny long bristles at the anterior margin; more than 25 long bristles along the outer margin; 20–50 long bristles in dorsal part of galea-lacinia anterolateral area. Glossae and paraglossae as in Fig. 15. Glossae not stretched laterally, paraglossae relatively

massive. Superlinguae of hypopharynx massive, distinctly stretched laterally, with a pilosity typical of *Ecdyonurus helveticus* species-group (Fig. 14).

Thorax yellowish-brown to brown, with distinct whitish bands. Posterolateral pronotal projection asymmetrical, tapered distally and rounded at the tip (Figs 18, 27–29). Distal part of pronotal projection directed towards the body. Legs yellowish-brown, with slightly darker tibiae and tarsi. Femora yellowish-white to yellowish-brown, with unclear cross-like drawing dorsally (Fig. 33). Femora relatively long and slender. Length/width ratio of metafemora mean 2.67 ($\sigma = 0.173$; $n = 10$). Spatulas on dorsal surface of femora in their central area as in the figures 19a and 31; proximally and laterally as in the figures 19b and 32. External margin of femora with long setae and short spines. Metafemora with more than 12 external short spines. Tibiae with pale spot distally, trochanters with distinctly darker tip.

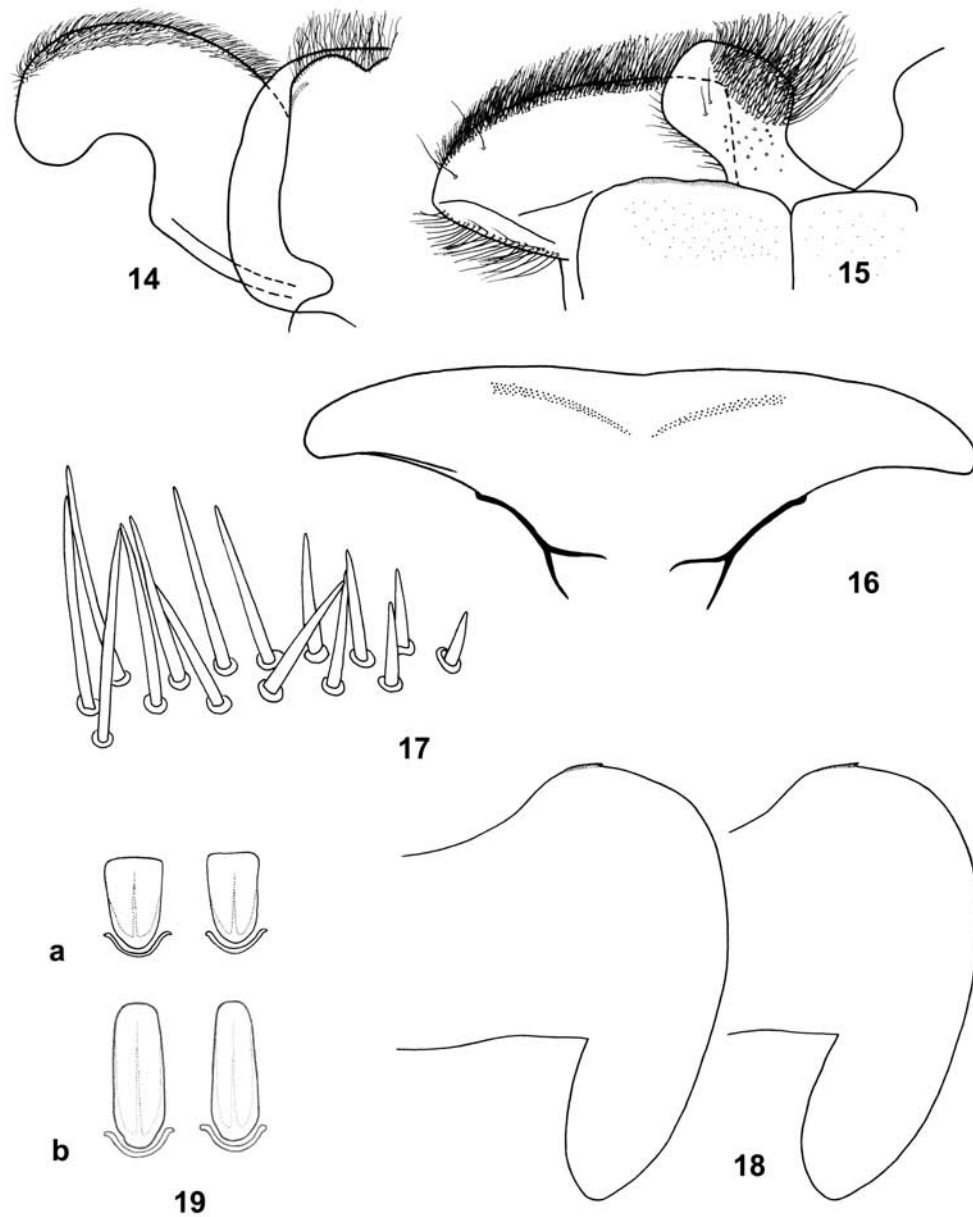
Abdomen generally pale, yellowish-brown to light brown. Surface of terga with well visible drawing (Fig. 34): segment I with central smudge near posterior margin; segment II with a pair of pale oval transversal spots centrally and two pale spots laterally; segments III–IV with a pair of pale small spots on the dark background centrally, pale spot near posterior part of tergum, and two pale spots laterally; segments V–VII



Figures 10–13. *Ecdyonurus nigrescens* (Klapálek, 1908), male subimago, Brebeneskul stream, Chornohora Range, Ukrainian Carpathians: (10) penis dorsally; (11) forceps structure ventrally; (12) right penis lobe dorsally; (13) left penis lobe dorsally.

uniformly colored with a three pale spots forming V-shaped drawing centrally and two pairs of pale spots laterally; VIII–IX unicolorous, with V-shaped dark spot around pale area centrally and two dark spots laterally; segment X unicolorous, yellowish-brown to brown, occasionally with unclear two smudges centrally. Posterior margin of terga with two dominant kinds of spines' arrangement (Fig. 22): (1) the dense row of large spines tapered distally and

bluntly pointed at the tip, occasionally chipped apically; these spines alternating with smaller pointed spines, widened at the base; (2) the dense row of large rectangular-like spines, widened centrally and slightly tapered apically, blunt or rounded at the tip; these spines alternating with smaller pointed spines, widened at the base. Occasionally posterior margin of terga laterally with spines of the first kind alternating with those of the second kind. Submarginal part and

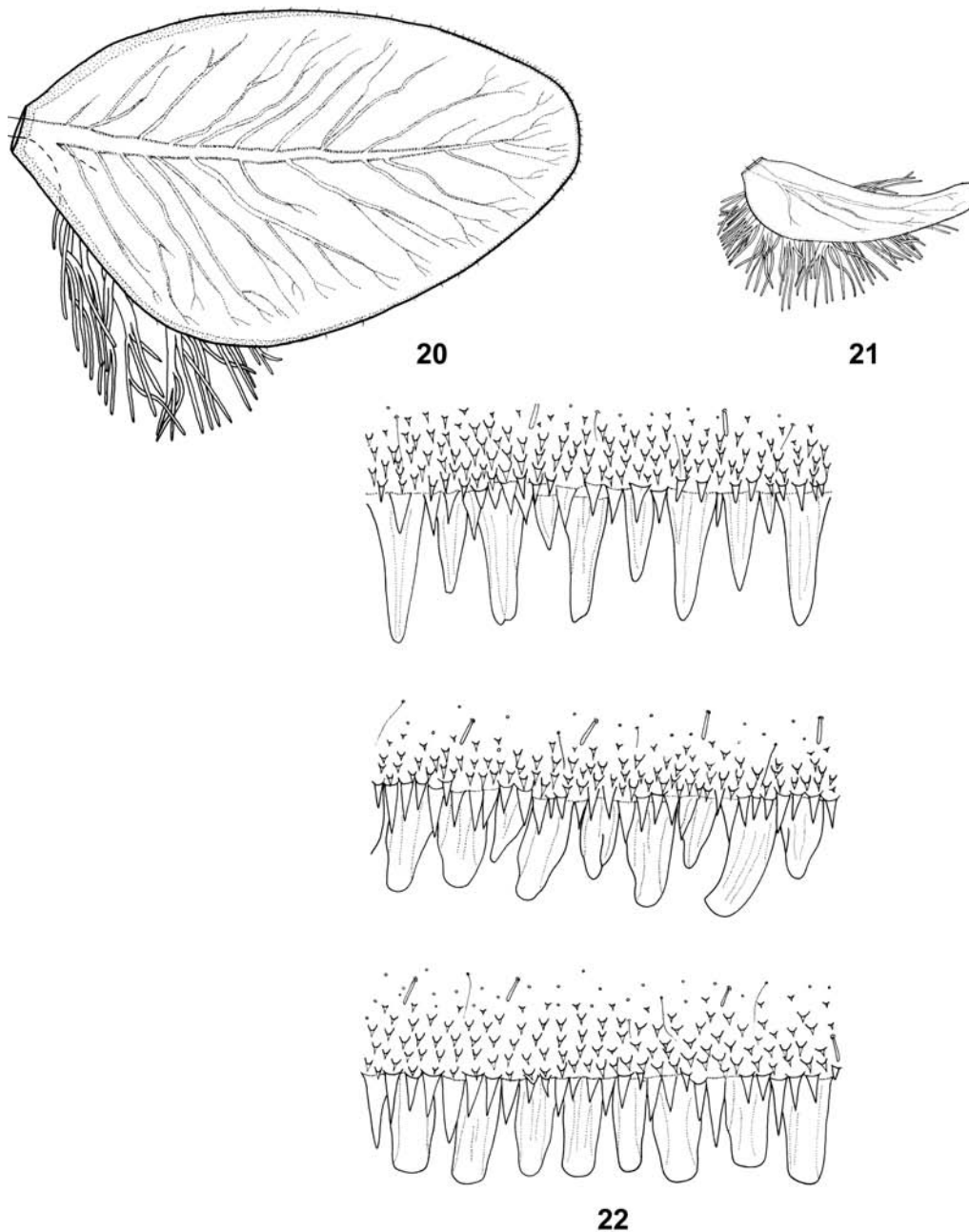


Figures 14–19. *Ecdyonurus nigrescens* (Klapálek, 1908), mature larva, Brebeneskul stream, Chornohora Range, Ukrainian Carpathians: (14) lingua and superlingua of hypopharynx; (15) glossa and paraglossa of labium; (16) labrum; (17) submedial bristles of labrum arranged into a two rows; (18) posterolateral pronotal projection; (19) femoral spatulas situated in centrally (19a), proximally and laterally (19b).

surface of terga with numerous small pointed spines, long spatulas and hairs. Tracheal gills yellowish-brown to brownish-grey. Gill 1 relatively long and slender, distinctly tapered distally (Fig. 21). Gill 4 distinctly asymmetric and wide, distal margin blunt (Figs 20, 30). Length/width ratio of the gill 4 mean 1.75 ($\sigma = 0.146$; $n = 10$). Sterna yellowish-white to yellowish-brown, with a pair of brownish central spots on segments I-IX.

Sternum X with central pale spot, occasionally unicolorous, yellowish-brown. Nerve ganglia well visible, dark pigmented. Caudal filaments unicolorous, yellowish-brown.

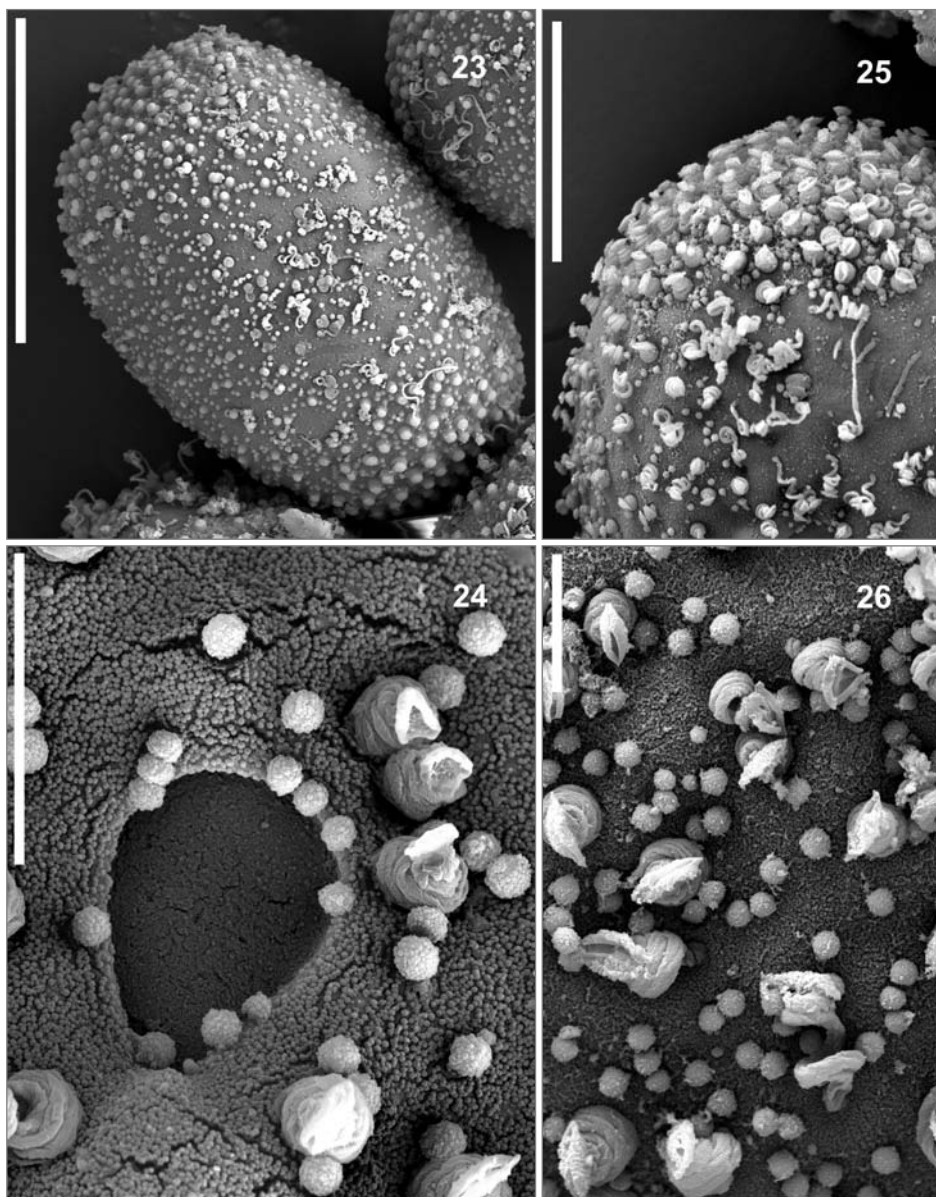
Egg. Measurements: length 150–164 μm ; width 96–115 μm . Egg oval (Figs 23–26). Chorionic surface typical of the *Ecdyonurus* eggs, with attachment structures, tubercles and small granules (Fig. 23).



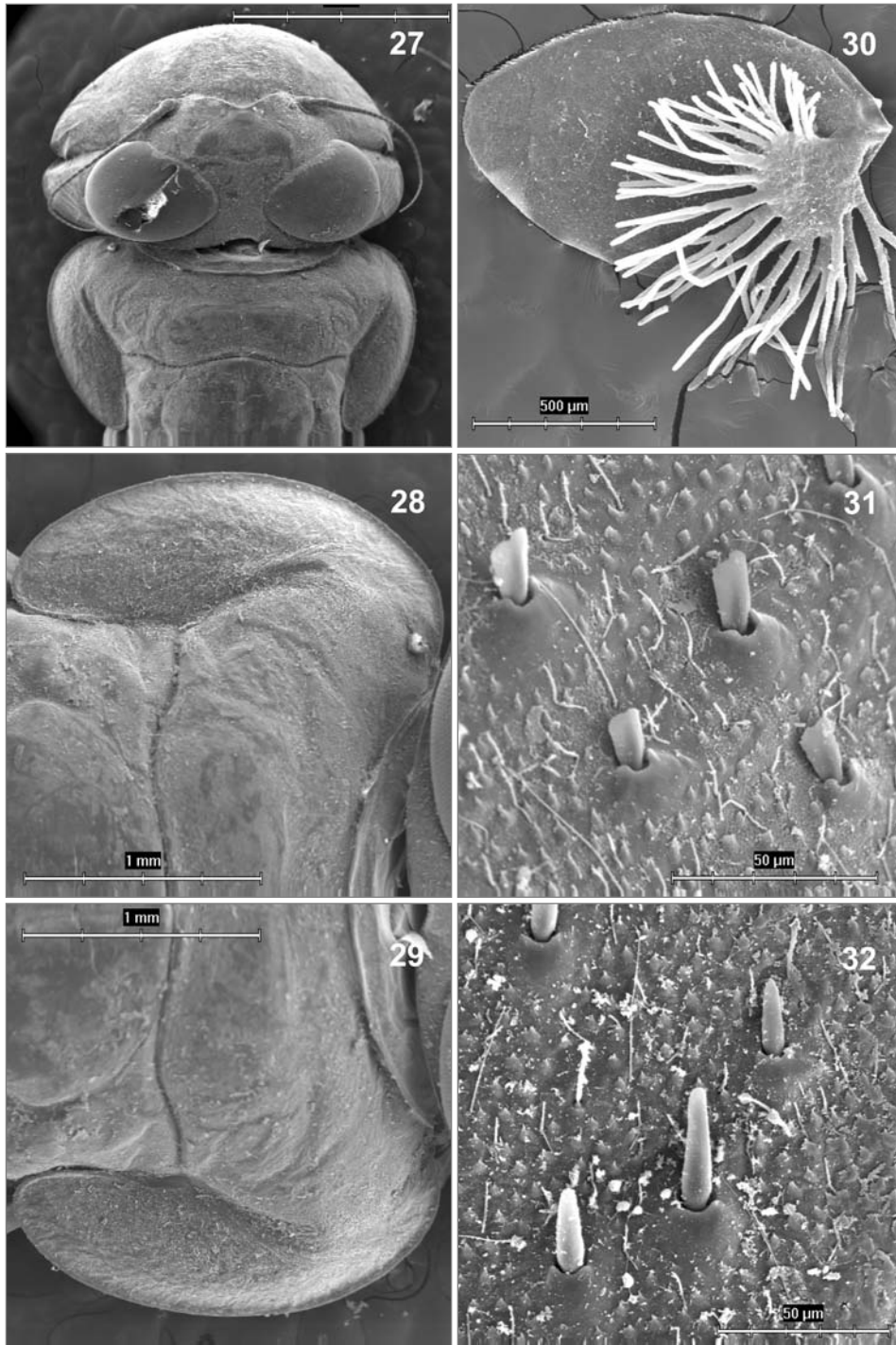
Figures 20–22. *Ecdyonurus nigrescens* (Klapálek, 1908), mature larva, Brebeneskul stream, Chornohora Range, Ukrainian Carpathians: (20) fourth gill; (21) first gill; (22) variability of central part of posterior margin of fifth abdominal tergum.

Attachment structures are characterized by knob-terminated coiled threads (KCTs), like in other species of *Ecdyonurus* genus (Koss and Edmunds 1974, Gaino and Rebra 2003) and are of two kinds. The larger ones (diameter 3–3.8 μm) are concentrated at two egg poles rather densely (distance between them 0.8–3.0 μm) (Figs 23, 25). The smaller KCTs attachment structures (diameter 2.2–2.8 μm) are scattered over the whole chorionic surface (distance between them 9.7–2.6 μm). Small rounded tubercles (0.9–1.6 μm in diameter) are

rather densely and irregularly distributed over the chorionic surface (0.3–6.5 μm distance between them) (Figs 24, 26). Tubercles and the whole chorionic surface are covered by very small rounded granules (0.15–0.19 μm in diameter) (Fig. 26). In equatorial area 2–3 micropyles are situated. Sperm guide ovoidal, 11.1–9.6 μm in length and 7.3–6.8 μm in width. Micropylar rim thin, with a few sparsely distributed tubercles (some micropylar rims with one tubercle, or without tubercles (Fig. 24)).



Figures 23–26. *Ecdyonurus nigrescens* (Klapálek, 1908), egg (from female imago): (23) general outline of the egg, scale bar = 100 μm ; (24) micropyle and detail of chorionic surfaces (tubercles and small granules), scale bar = 10 μm ; (25) egg pole with large KCTs attachment structures, scale bar = 50 μm ; (26) chorionic surface with small KCTs attachment structures, tubercles and small granules, scale bar = 10 μm , (HITACHI S-4700 scanning electronic microscope).



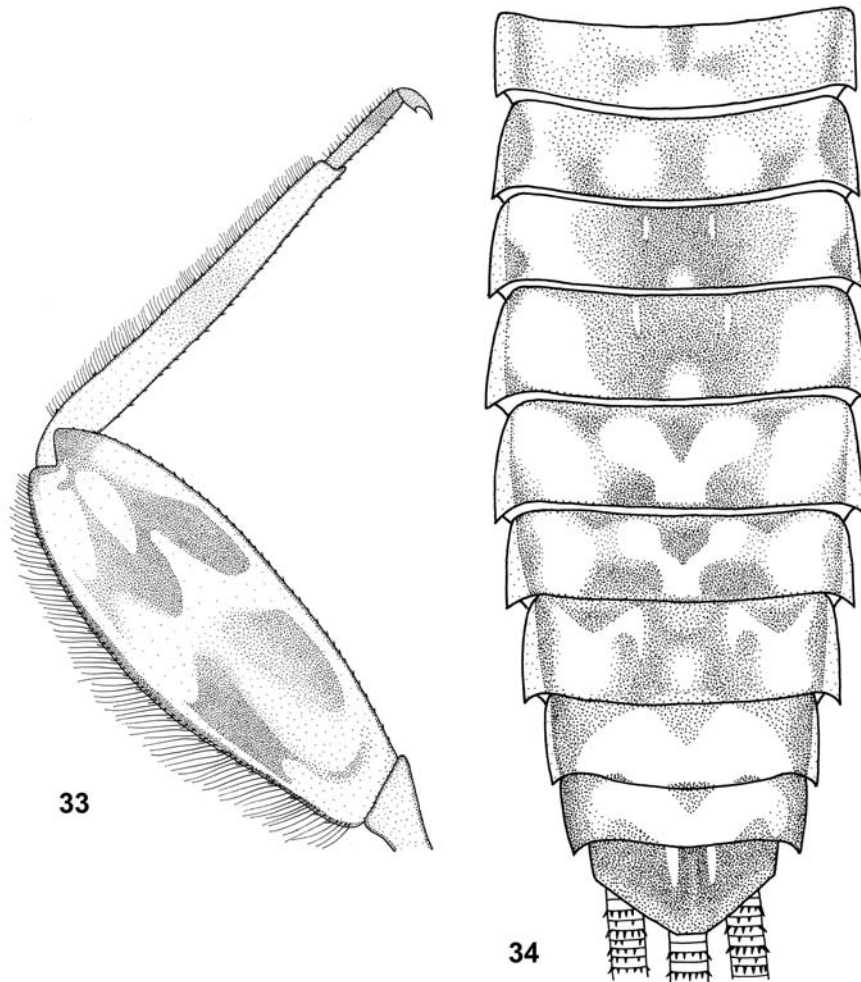
Figures 27–32. *Ecdyonurus nigrescens* (Klapálek, 1908), mature larva, Brebeneskul stream, Chornohora Range, Ukrainian Carpathians: (27) head and pronotum; (28–29) posterolateral pronotal projections; (30) fourth gill; (31) femoral spatulas situated in centrally; (32) femoral spatulas situated proximally and laterally (JEOL JSM 7401F scanning electronic microscope).

DISCUSSION

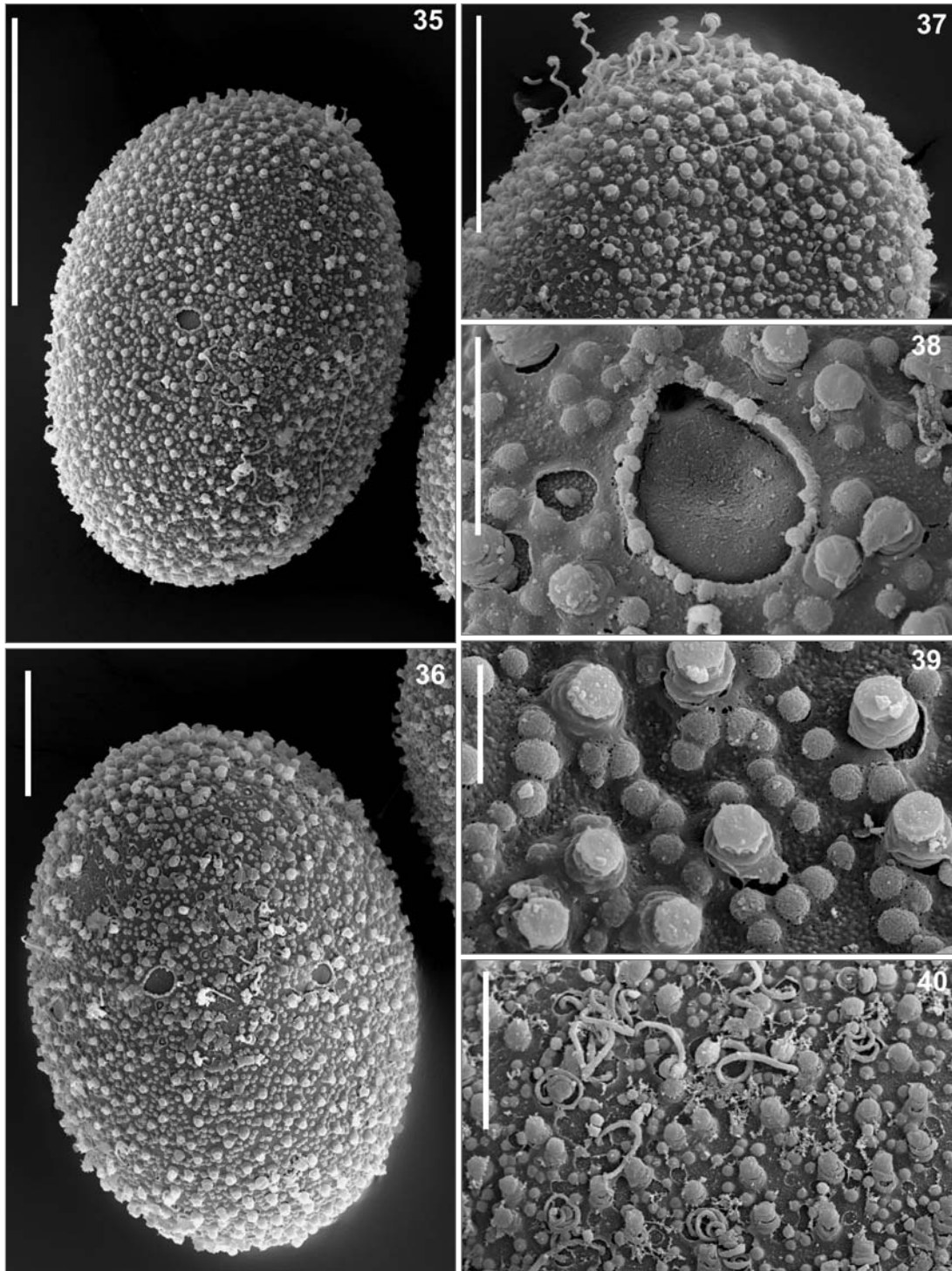
E. nigrescens belongs to the *E. helveticus* species-group, which can be characterized by the combination of following features (see Hefti *et al.* 1989, Belfiore and Buffagni 1994): larval hypopharynx without long and dense setae distally; posterolateral extensions of larval abdominal segments short and directed parallelly to the body axis; apical sclerite of penis lobes of imago elongated laterally.

This species can be distinguished from other representatives of the *E. helveticus* species-group by the following combination of characters: **in male imago:** (1) penis lobes not elongate, slightly stretched laterally; (2) straight lines of apical sclerite form obtuse angle mean 110°; (3) lateral sclerite is large, with wideness

part situated approximately in 2/3 its length; (4) the tip of lateral sclerite relatively broad; (5) basal sclerite relatively massive with 1–2 strong teeth directed perpendicularly to axis of symmetry of the penis lobe; (6) penis stem without hump; (7) coloration of body, legs and wings; **in larva:** (8) position and shape of bristles on ventral side of labrum; (9) position and number of setae and bristles on the first segment of maxillary palps and dorsal part of anterolateral area of galealacinia; (10) shape of posterolateral pronotal projection (asymmetrical, tapered distally and rounded at the tip with distal part directed towards the body); (11) different kinds and shape of spines' arrangement in posterior margin of abdominal terga; (12) structure and proportions of the gills 1 and 4, and proportions of the metafemora; (13) general coloration of body, and



Figures 33–34. *Ecdyonurus nigrescens* (Klapálek, 1908), mature larva, Brebeneskul stream, Chornohora Range, Ukrainian Carpathians. (33) hind leg dorsally; (34) abdominal color patterns dorsally.



Figures 35–40. *Ecdyonurus austriacus austriacus* Kimmins, 1958, egg (from mature larvae): (35) general outline of the egg – type A, scale bar = 100 μm ; (36) general outline of the egg – type B, scale bar = 50 μm ; (37) egg pole with KCTs attachment structures, scale bar = 50 μm ; (38) micropyle, scale bar = 10 μm ; (39) detail of chorionic surfaces (KCTs, tubercles and small granules), scale bar = 10 μm ; (40) chorionic surface with KCTs attachment structures, tubercles and small granules, scale bar = 20 μm (HITACHI S-4700 scanning electronic microscope).

especially of the abdominal terga and legs; (14) general shape of body (especially shape of head); (15) structure of the eggs.

The combination of characters 1–3, 5, 8–11, 13 and 15, distinctly distinguishes *E. nigrescens* from close related species *E. picteti*.

The larvae of *E. nigrescens* can be easily separated from *E. austriacus nataliae* by features 8, 9 and 12–14 (Godunko and Klonowska-Olejnik 2004). In comparison with *E. nigrescens*, the larvae of the subspecies *E. austriacus austriacus* (observed material from Austria) can be characterized by: (1) the presence of slender and longer bristles on the ventral surface of labrum, arranged in two dense rows (up to 18–20 bristles on each sides); (2) more robust superlinguae of hypopharynx with basal part being clearly wider; (3) the different position and number of setae and bristles on the first segment of maxillary palps and dorsal part of anterolateral area of galea-lacinia (e.g., 4–10 tiny long bristles at the anterior margin; 30–50 long bristles along the outer margin; 20–35 long bristles in dorsal part of galea-lacinia anterolateral area); (4) the different coloration of abdominal terga (e.g., segments III–IV mainly without pale spot near posterior margin of tergum; V-shape drawing of segment V is unclear, and enclose three pale spots arranged in pale smudge; coloration of segments VI–VII is close to that of segment III–IV); (5) not slender, relatively massive gill 1, slowly tapered distally; (6) the different proportions and shape of gill 4 (e.g., length/width ratio of gill 4 mean 1.72); (7) body length and proportions (see also Hefti and Tomka 1986, Hefti *et al.* 1989, Bauernfeind and Humpesch 2001).

The eggs of *E. nigrescens*, can be clearly distinguished from the eggs of *E. rizuni* and *E. austriacus nataliae* by concentration of KCTs attachment structures at both egg poles (Godunko and Klonowska-Olejnik 2004, Godunko *et al.* 2004). In species *E. picteti* (Hefti and Tomka 1986), *E. subalpinus* (Godunko and Soldán 2003) and *E. silvaegabretae* (Soldán and Godunko 2006), concentration of KCTs attachment structures is present at one egg pole only.

A brief description and schematic depiction of eggs of *E. austriacus austriacus* was published by Hefti and Tomka (1986), without any details on surface structure of chorion. We have studied the egg structure of *E. austriacus austriacus* in scanning electronic microscope with a purpose to find some additional characters to distinguish this species from close related *E. nigrescens* (see below).

E. austriacus austriacus (egg description).

Measurements: length 168–173 μm ; width 110–122 μm . Egg oval (Figs 35–36). Chorionic surface with KCTs attachment structures, tubercles and small granules, typical of the *Ecdyonurus* eggs (Koss and Edmunds 1974, Gaino and Reborá 2003). Chorionic surface with

many KCTs attachment structure (diameter 2.1–2.6 μm , distance between them 2.2–5.7 μm , generally 3.3–4 μm) (Figs 39–40). At one or two poles of eggs the KCTs attachment structures are slightly larger (diameter 2.9–3.6 μm), but not more concentrated than those on the chorionic surface (distance between them 2.1–5.7 μm , generally 3.6 μm) (Figs 35–37). The whole chorionic surface with many rounded tubercles (diameter 1.1–2 μm), densely covered the egg (distance between them 0.4–1.8 μm) (Figs 39–40). Tubercles and chorionic surface with very fine granules (diameter 0.14 μm) (Figs 38–39). Three to five micropyles are visible in subequatorial area. Sperm guide ovoidal, 10.5–9.6 μm in length and 8.2–6.4 μm in width. The micropylar opening situated to the side. Micropylar rim narrow, with rather sparsely distributed small tubercles (Fig. 38).

The eggs of *E. austriacus austriacus* can be clearly distinguished from the eggs of *E. austriacus nataliae*, *E. rizuni* and *E. nigrescens* (Godunko and Klonowska-Olejnik 2004, Godunko *et al.* 2004). The size of KCTs attachment structures on the chorion surface and at the egg poles in *E. austriacus austriacus* is like that in *E. nigrescens*. However, the concentration of KCTs attachment structures at both egg poles in *E. nigrescens* is more dense (e.g. the number of KCTs attachment structures is higher, distance between them being clearly smaller in comparison with *E. austriacus austriacus*). The chorion surface in *E. austriacus austriacus*, as concerns the density of KCTs attachment structures and tubercles arrangement, is similar to that in *E. austriacus nataliae*. However, in contrast to *E. austriacus nataliae*, the KCTs attachment structures in *E. austriacus austriacus* are clearly larger at the egg poles than on the surface of the chorion. The number of tubercles on the chorion surface of eggs in *E. austriacus austriacus* is clearly higher than in *E. nigrescens*, and their rounded shape is essentially different from that in *E. rizuni*. The structure of micropyle in *E. austriacus austriacus* more resembles that in *E. austriacus nataliae* and *E. rizuni*, and is characterized by the presence of a narrow micropylar rim with rather sparsely distributed small tubercles. The micropyle in *E. nigrescens* with thin, hardly visible micropylar rim, and isolated tubercles (some micropylar rims with one tubercle, or without tubercles). Some differences in the general egg shape and measurements are present as well (Figs 23, 35–36).

The chorionic structures of eggs in other representatives of *E. helveticus* species-group (e.g., *E. alpinus* Hefti, Tomka & Zurwerra, 1987, *E. helveticus* (Eaton, 1885), *E. parahelveticus* Hefti, Tomka & Zurwerra, 1986, *E. zelleri* (Eaton, 1885)) haven't been described sufficiently. Basing on the available descriptions and very sketchy depictions (Hefti and Tomka 1986, Hefti *et al.* 1986, Hefti *et al.* 1987), we can just assume that in

species mentioned the KCTs attachment structures are also present at two egg poles, as in *E. nigrescens*, the precise study in electronic microscope being necessary.

The structure of imago male genitalia in *E. nigrescens* partly bears resemblance to that in both subspecies of the *E. austriacus*. The imago male of *E. nigrescens* can be distinguished from *E. austriacus nataliae* by the characters 2–4 (especially by the shape of proximal part of lateral sclerite) and 6–7. At the same time, penis lobes of male imagoes in *E. nigrescens* and *E. austriacus austriacus* are very similar, especially in structure of apical and lateral sclerites, general shape and proportions. However, male imago of *E. nigrescens* can be distinguished from *E. austriacus austriacus* by the structure of styliger and forceps, viz.: (1) lateral protuberances of styliger only slightly asymmetrical with slightly incised inner margin (in contrast to distinctly asymmetrical and incised inner margin protuberances in *E. austriacus austriacus*); (2) segment 4 of forceps shorter than segment 3 (in contrast to *E. austriacus austriacus*, in which segment 4 is mainly longer than segment 3 or equal to it). The general coloration of body in imago and subimago of *E. nigrescens* and *E. austriacus austriacus* is greatly similar. However, the adult specimens of these taxa differ from each other by body and wing sizes.

The coloration of body, legs and wings in male imagoes of *E. nigrescens* described in the papers of J. Dziędzielewicz and F. Klapálek (1908a, b) in general meets the coloration of our material reared from larvae and subimagoes. It should be noted, that male imagoes, just after moulting from subimago, have somewhat lighter coloration with slight greyish tint, disappearing in few hours, and body color becoming clearly brown or dark brown (occasionally black) with reddish tint on lateral sides of thorax and abdominal terga. The picture of genitalia of *E. nigrescens* in the papers of J. Dziędzielewicz and F. Klapálek (1908a, 1908b), although being schematic, displays the general view of penis lobes, which slightly stretch laterally. Straight lines of apical sclerite of illustrated specimen form obtuse angle of approximately 100–105°. This feature is close to values obtained in our study of imaginal material on *E. nigrescens* (Figs 1–6, 8, 9). In contrast, the apical sclerite in *E. picteti* is almost rectangular-like bent (Hefti and Tomka 1986, Hefti *et al.* 1989), and in both subspecies of *E. austriacus* it forms obtuse angle approximating between 110–120° (Hefti and Tomka 1986, Godunko and Kłonowska-Olejnik 2004, and observed material from Austria). The comparison of body, wing and leg colors and of genitalia structure of male specimens reared from larvae in 2002 (Figs 1–5) with those in specimens collected by J. Dziędzielewicz in 1908–1910 (Figs 6, 8, 9) completely confirms the

attribution of the whole material to a single species, *E. nigrescens*. As mentioned above, these characters meet the original description by F. Klapálek. Moreover, the comparison of genitalia of three male imagoes from the collection of ZMUH (on the basis of which V. Puthz has brought *E. nigrescens* and *E. picteti* to the synonymy) with reared material from the Brebeneskul stream, kindly made by Prof. Hans Strümpel in October 2006, has shown the identity in their structure.

BIOLOGY AND DISTRIBUTION

The larvae of *E. nigrescens* occur in small rapid and cool-water streams and rivers of the Chornohora Range of the Eastern Carpathians. The species has been found from crenalic to epirhithralic sections of streams along the main chain of the Chornohora Range between 1100–1450 m. a.s.l. (northern and southern slopes), and from epirhithralic to metarhithralic sections of small brooks in subalpine forest and in rivers of the north-western slopes of this Range between Vorokhta village, Yasinia village, Kvasy village and Rakhiv town, within 700–1200 m. a.s.l. Possibly, this species also occurs in streams and rivers of adjoining mountain-regions of the Ukrainian Carpathians, e.g. Svydivets, Marmarosh and Chyvchyny. Thus, *E. nigrescens* seems to be the endemic species of the Eastern Carpathians.

The larvae of this species evidently prefer stony sections of streams and rivers with pebbles, 0.5–4.0 m wide and 0.3–1.0 m deep; current velocity higher than 1 m/s. The water temperature in summer months is between 7 and 13°C; pH 7.0–7.5. In these habitats *E. nigrescens* occurs with *Ameletus inopinatus* Eaton, 1887, *Baetis alpinus* (Pictet, 1843), *B. rhodani* (Pictet, 1843), *B. vernus* Curtis, 1834 and *Rhithrogena iridina* (Kolenati, 1859).

Most probably, *E. nigrescens* is a univoltine winter species with one generation a year, belonging to the Uw category following Clifford (1982). The larvae occur from late autumn and growth during the winter. The flying period extends from July to August–September (probably also to the first half of October at low altitudes).

Up to 40% of larvae of *E. nigrescens* collected in the Brebeneskul stream within 1400–1450 m. a.s.l. bear the larvae and pupae of parasitic species of the genus *Symbiocladius* Kieffer, 1925 (Diptera: Chironomidae) (for the more details see Gilka *et al.* 2007).

ACKNOWLEDGEMENTS

Our sincere thanks are due to Prof. Tomáš Soldán (Institute of Entomology, Academy of Sciences of the

Czech Republic), to Dr. Ernst Bauernfeind (Naturhistorisches Museum Wien) and to Dr. Pavel Chvojka (Department of Entomology, National Museum Praha) for the great support during our stay at these institutions. We are very obliged to Prof. Hans Strümpel (Zoologisches Museum der Universität Hamburg) for the great consultations.

This research was financially supported by the INTAS Fellowship Grant for Young Scientists (INTAS Ref. Nr 05-109-4162) and by a Grant Agency of the Academy of Sciences of the Czech Republic No. QS500070505 for the first author, and by the grants of Institute of Environmental Sciences BW/V/INOS/4/06 and DS/WBiNoZ/INoS/756/06 for the second author. The stay in Kraków supported by Kasa J. Mianowski for the first author.

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Received: April 28, 2008

Accepted: October 20, 2008