

Description of some West-Palaearctic species of *Eukiefferiella*
Thienemann, *Tvetenia* Kieffer and *Tokunagaia* Sæther
(Diptera, Chironomidae)

Описание некоторых западнопалеарктических видов
Eukiefferiella Thienemann, *Tvetenia* Kieffer и *Tokunagaia* Sæther
(Diptera, Chironomidae)

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Keywords: Diptera, Chironomidae, Orthocladiinae, taxonomy, redescription, key, autoecology.

Ключевые слова: Diptera, Chironomidae, Orthocladiinae, таксономия, переописание, ключи, аутоэкология.

Abstract. West Palaearctic larvae of the genera *Eukiefferiella* Thienemann, 1926, *Tokunagaia* Sæther, 1973 and *Tvetenia* Kieffer, 1922 are redescribed and a key to species is provided. The three genera are recognized by the following characters of larva: head and body colour, head capsule shape, number and length of segments of antenna, labrum with simple or branched setae anteriores S_p , simple or bifid setae anteriores S_{III} , mandible with mola provided with spines or bristles, presence of long setae on body segments, developed procerci with a long subapical seta. The shape of mentum is a useful character with a single or a bifid, large or narrow median tooth and 4 or 5 lateral teeth, but often the teeth are worn out and their shape must be used with caution. Number and length of antennal segments, antennal blade and distal setal mark position are other diagnostic characters; the third antennal segment may be reduced to completely disappeared. Species live in different types of running waters, from glacial streams to lowland large rivers, their different ecology can aid in species identification.

Резюме. Приведены переописания западнопалеарктических личинок родов *Eukiefferiella* Thienemann, 1926, *Tokunagaia* Sæther, 1973, *Tvetenia* Kieffer, 1922 и определительные таблицы для видов. Для личинок трех родов характерны следующие признаки: цвет головы и тела, форма головной капсулы, количество и длина члеников антенны, лабрум с простыми или разветвленными передними щетинками S_p , простыми или раздвоенными передними щетинками S_{III} , внутренний край мандибулы (mola) с шипами или щетинками, сегменты тела с длинными щетинками, подставка преанальной кисточки с длинной субапикальной щетинкой. Ментум с одним или раздвоенным, большим или узким срединным зубцом или двумя зубцами и 4 или 5 боковыми зубами, но часто зубцы стерты и этот признак следует использовать с осторожностью. Другими диагностическими признаками являются число и длина члеников антенны, положение лаутерборновых и кольцевых органов; третий членик антенны может быть редуцирован вплоть до полного исчезновения. Виды обитают в разных типах проточных вод, от ледниковых ручьев до крупных рек в низинах; их различные экологические характеристики могут помочь в идентификации видов.

Introduction

The aim of the present paper is a revision of some of the known larvae belonging to the genus *Eukiefferiella* Thienemann, 1926 and related genera *Tokunagaia* Sæther, 1973 and *Tvetenia* Kieffer, 1922. The descriptions are based on samples collected in Italy, the species found are widespread in West Palaearctic region. Descriptions and keys to larvae are in Thienemann [1936], Zavřel [1939], Chernovskii [1949], Rossaro [1982], Bode [1983], Moller Pillot [1984] and Schmid [1993], but are updated and consider only some species. The genus *Tvetenia* was revised [Sæther, Halvorsen, 1981] considering species formerly included in *Eukiefferiella*, the genus *Tokunagaia* was revised [Halvorsen, Sæther, 1987] including species formerly included in the *Eukiefferiella rectangularis* group, as *E. tonollii* [Rossaro, 1983], which were moved to *Tokunagaia*. The genus *Eukiefferiella sensu lato* including the two other genera was revised by Lehmann [1972], but only adult males and pupal exuviae were considered; the genus was divided into groups, no information about larvae was included, so the groups formation did not consider larval characters. Cranston et al. [1983], Epler [2001] and Andersen et al. [2013] gave a key to larvae of all genera of Orthocladiinae, including the three genera. A key to species groups within *Eukiefferiella* was given in Andersen et al. [2013], considering the following groups: *devonica*, *gracei*, *brehmi*, *coerulescens*, *claripennis*, *cyanea*; the *rectangularis* and *verralli* groups [Lehmann, 1972] were included in *Tokunagaia* and *Tvetenia* respectively. Many other species were more recently described especially from areas outside West-Palaearctic region, some species were described including also larval descriptions [Imada, 2020; Makarchenko, Makarchenko, 2007; 2009; 2010; 2012; 2017; Makarchenko et al., 2019; Qi et al., 2012a–b; Moubayed-Breil, 2012, 2015]. Emphasis is here given only to characters relevant in species identification as

number of antennal segments, length of antennal blade, position of ring organ and distal setal mark, shape of teeth of mentum, setae anteriores of labrum, head capsule colour, development of setae on abdominal segments.

After a short description of single species, a key to species is given. The genus is particularly important in freshwater ecology; it is rich in species preferring fast flowing waters. Longitudinal zonation of different species is observed within the genus *Eukiefferiella* and will be briefly discussed in the ecological section.

Material and methods

The specimens included in this study were collected between 1974 and 2019 in different streams (Table 1). Larvae were sampled with a Surber net, pupal exuviae and pupae with a Brundin net [Brundin, 1966], adults with a hand net [Langton, Pinder, 2007]. Larvae and mature pupae were transported alive to the laboratory using a portable refrigerator. Individuals were reared to adults within Petri dishes or within glass tubes in a controlled-temperature chamber at a temperature ranging from 6 to 15 °C. Moreover, mass rearing was carried out within small tanks at the same temperature range, with aeration guaranteeing dissolved oxygen saturation. A photoperiod of 14 h of light and 10 h of dark was maintained using a fluorescent lamp OSRAM LUMILUX COMBI-N/P, 18W. Sampled and reared individuals were conserved in 75 % ethanol. For microscopic slides preparation, specimens were transferred in acetic acid and mounted in Faure, alternatively some specimens were transferred from acetic acid to butanol and to a phenol: xylene mixture 3:1, then mounted in balsam [Sæther, 1969; Wirth, Marston, 1968]. In the case of successful rearing, adult, larval and pupal exuviae were mounted on the same slide. To examine the mouth parts (antennae, labrum, mentum, mandible) it was often necessary to dissect the head capsule in separate parts.

Descriptions provided in this study are based on reared adult males with associated pupal exuviae and larvae, when available. Body parts measurements were made at different magnifications (10–1000×) using a LEICA LS B2 optic microscope connected to a LEICA DFC320 camera and analysed with LEICA LAS software V4.8. Some measures as the length and width of head capsule were generally taken from literature [Schmid, 1993] because it was impossible to have correct values, when only slides with dissected heads were available. Measurements were given in µm or in mm. Photos of characters of taxonomic interest were obtained with the LEICA DFC320 camera. Some characters were drawn on paper with a drawing tube and scanned with an EPSON Perfection V370 scanner. Adopted terminology follows Schmid [1993], Sæther [1980], Bode [1983], Epler [2001], [Andersen et al., 2013].

The samples examined at present are deposited in the personal collection of the author.

Table 1. Longitude and latitude in decimal degrees of the most important sampled localities and prevalent habitat type

Таблица 1. Долгота и широта в десятичных градусах наиболее важных мест отбора материала и основных типов местообитаний

Locality	Habitat	Longitude	Latitude
Acqualba	krenal	8.37	45.84
Adda	epipotamal	9.53	45.56
Agogna	rhithral	8.48	45.85
Aso	rhithral	13.75	43.06
Brembo	rhithral	9.82	46.04
Brenta	rhithral	12.30	45.36
Caré Alto	kryal	10.60	46.10
Chiareggio	rhithral	9.77	46.31
Curone	krenal	9.22	45.46
Dora Veny	kryal	6.88	45.79
Elvo	rhithral	7.96	45.55
Forni	kryal	10.22	46.54
Lambro	rhithral	9.26	45.91
Lys	kryal	7.80	45.86
Mignone	rhithral	11.93	42.22
Ofanto	rhithral	15.37	41.40
P.N. Abruzzi	rhithral	13.89	41.87
Po	potamal	9.96	45.12
Potenza	rhithral	13.67	43.42
Quiliano	rhithral	8.38	44.30
rio Mannu	rhithral	9.15	39.53
Sangro	rhithral	13.75	41.86
Sarca di Genova	kryal	10.96	46.45
Tagliamento	rhithral	12.95	46.17
Taro	rhithral	10.19	44.80
Ticino	epipotamal	8.72	45.50
Toce	rhithral	8.28	46.05
Velino	rhithral	12.73	42.52
Vetoio	krenal	13.34	42.36
Veza	rhithral	10.41	46.31
Dirillo	rhithral	14.68	37.14

Description of genera

Tvetenia Kieffer, 1922

Detailed descriptions of the generic characters of larva are in Zavřel [1939], Schmid [1993], Epler [2001], Andersen et al. [2013].

The characters useful to separate species are the yellow head capsule, branched setae anteriores S_p , the length of antennal segments, the distance of seta submenti from posterior margin of ventromental plates, developed setae on abdominal segments and proceri with a long subapical seta.

Tokunagaia Sæther, 1973

Descriptions of the generic characters of larva are in Halvorsen and Sæther [1987], Epler [2001], Andersen et al. [2013].

The larvae are similar to the ones of *E. claripennis* group, and are separated by the presence of an antenna with 5 seg-

ments; a mentum with a paired median narrow tooth, 5 lateral teeth, distal setal mark near to ring organ also characterize the genus.

Eukiefferiella Thienemann, 1926

Descriptions of the generic characters of larva are in Zavřel [1939], Moller Pillot [1984], Schmid [1993], Epler [2001], Andersen et al. [2013]. Here some additional information is given.

Small or middle sized larvae up to 7 µm long, generally well characterized by a strong basal subapical seta on a developed procerus (Figs 1–2). Head capsule yellow, yellow brown, dark-brown, when yellow tip of mentum and mandible brown, head capsule colour has taxonomic importance, but may vary in different populations. Cephalic index (head length/wide, abbr. = C.I.) about 1.25–1.66, note that Schmid [1993] as cephalic index (abbr. CI) gives the reciprocal of C.I. Antenna with 4 or 5 segments (Figs 3, 7, 10, 14, 16, 20, 22, 80, 83, 86, 89, 92, 94, 96, 99, abbr. A_{1-4} , A_{1-5}), when antenna has 4 segments the 3rd and 4th ones are probably homologous to the 4th and 5th ones of species with 5 segments; when 5 segments are present, the 3rd is often the shortest, sometime very short and difficult to see, in species with 4 segments it can be assumed that the 3rd segment was reduced to disappear. On the basal antennal segment, a ring organ (abbr. R.O.) and an accessory organ or sense pit [Bode, 1983] or distal setal mark (abbr. D.S.M.) [Epler, 2001] are visible. D.S.M. generally is in distal position, far from R.O., sometime as in *E. coerulescens* is more proximal, near to R.O. (Figs 48, 80). Setae anteriores S_{I-III} are simple in *Eukiefferiella* and *Tokunagaia*, S_{III} is bifid in *E. ilkleyensis*, S_I is branched in *Tvetenia*, fringed in *T. calvescens* gr., palmate in *T. discoloripes-verralli* gr. (Figs 6, 9, 13, 27, 31). Mandible with inner margin (mola) with spines (Figs 5, 18, 42, 46, 101) (e.g. *E. claripennis* gr.) or long bristle-like appendages (*E. minor-gracei* gr. Figs 66, 70, 78), rarely smooth (*T. discoloripes-verralli* gr.) (Figs 12, 29). Mentum with simple or paired median tooth, sometime very large, with 4–5 lateral teeth (Figs 15, 53, 69, 77, 87, 95, 97). The length of mentum (abbr. M.L.) is measured between the apex of median tooth and a line joining the setae submenti; the distance between setae submenti (abbr. SM.S.D.) can be considered a measure of the width of mentum (Fig. 4). The wide of median tooth of mentum can be bound to total size of the larva, so the ratio between median tooth width and width of the nearest lateral teeth of mentum may be of interest. Abdominal segments with strong setae in *E. gracei* and *E. minor* (Fig. 1). Procerus very developed, with a strong basal subapical seta (Figs 2, 104) and a tuft of 7 long apical setae or anal macrosetae [Bode, 1983; Andersen et al., 2013], short in *E. cyanea* (Fig. 105). 10th abdominal segment with a dorsal, lateral and ventral seta; the dorsal one is called supraanal seta [Bode, 1983; Andersen et al., 2013], the ventral one ventral seta [Bode, 1983] or subbasal seta [Epler, 2001] (Fig. 103). Posterior parapods about 300 µm (Figs 1, 103), much more long in *E. cyanea* (Fig. 105).

Description of species

Tvetenia bavarica (Goetghebuer, 1934)

Small species 3–4.6 mm, head yellow with brown tips of mentum and mandible. Thorax green, abdomen often violet. C.I. 1.47. Antenna with 5 segments, 4th antennal segment 4–6 times longer than 3rd [Schmid, 1993], in our samples larvae associated to pupal exuviae of *T. bavarica* have the 4th segment

only about 3–4 times longer than 3rd (Fig. 24). Length of antennal segments 51.2–65.3, 15–23, 2.2–2.6, 7.4–7.9, 3.5–6.0 µm, A_{2-5} 39.8, AR 1.16–1.32, antennal blade 19.4–40 µm, R.O. 7.8 µm, D.S.M. 29.7 µm distant from base. M.L. 78.6–80 µm, SM.S.D. 51.0 µm, median tooth 19.7–20.9 µm wide, distance between seta submenti and base of ventromental plates 18.6 µm. Body with strong setae 130–180 µm. Procerus 29.3–38 x 22–27 µm, subapical seta 113–115 µm, anal tubuli 102–106 µm, supraanal seta 147–205 µm, lateral seta 20 µm, subbasal seta 120 µm, posterior parapods 220–248 µm. Sampling locality: Northern Italy, Lombardia, stream tributary of Oglio river, Vezza, 25.III.78, 19.VIII.79, Dosegù glacial stream, tributary of Adda River, Trentino, Vermigliano stream, 22.IV.78, Sarca di Genova, 12.IX.90.

Tvetenia calvescens (Edwards, 1929)

Figs 7–9, 24–27.

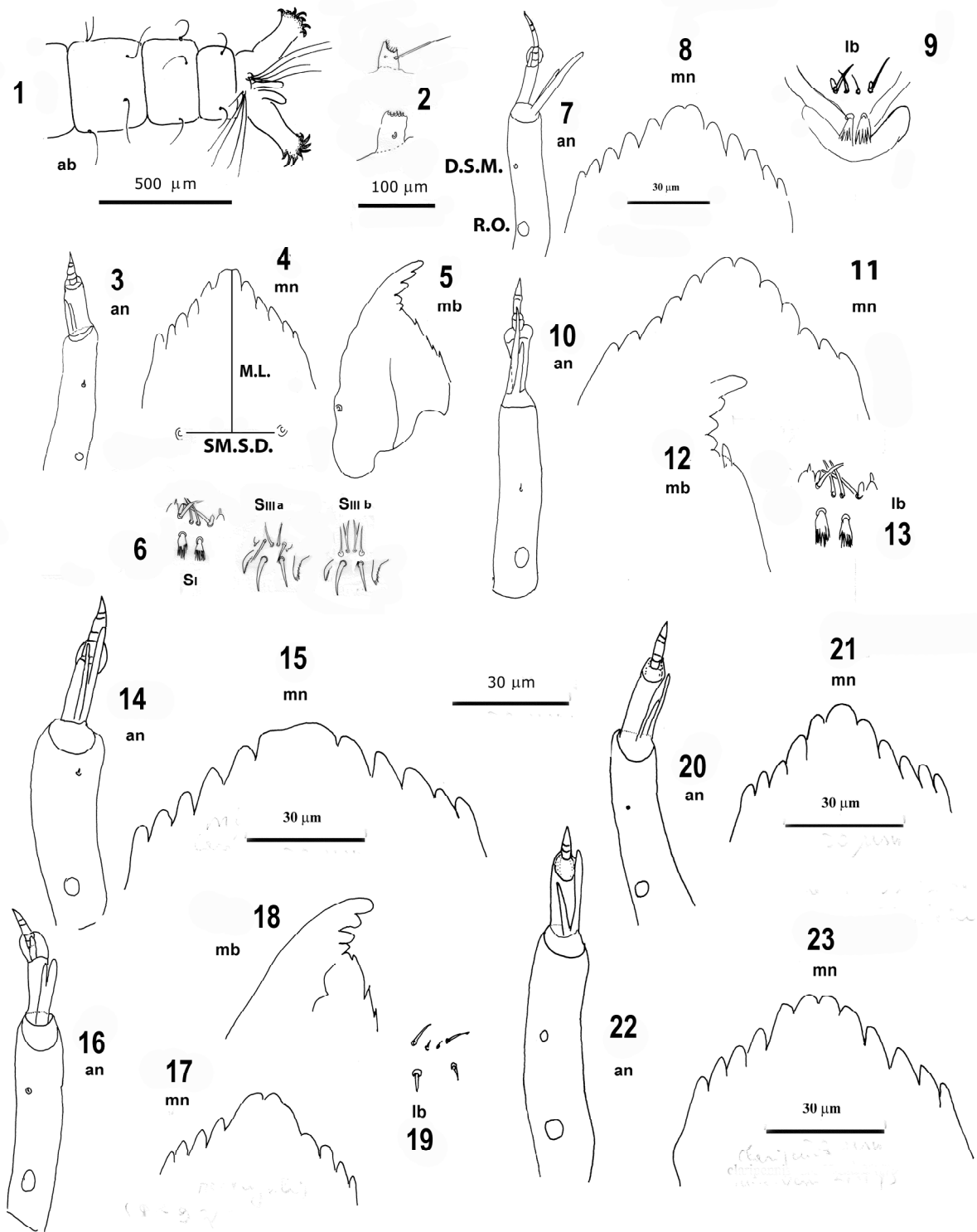
Small species 3–4.6 mm, head capsule yellow with brown tips of mentum and mandible, thorax green, abdominal segments often yellow, sometime violet. C.I. 1.49. Antenna with 5 segments, 1st long, 2nd < 1/2 than 1st, 3rd very short, 4th 2–3 times longer than 3rd [Schmid, 1993], at most 2 times longer in our samples, length of antennal segments: 43.8–55.3, 17.0–20, 1.9–3.2, 4.4–5.2, 1.8–6.5 µm. A_{2-5} 35.5–36.4 µm, AR 1.26–1.49, antennal blade 26.7 µm extending to the 4th segment, R.O. 6.9–8.7 µm, D.S.M. 32.1 µm distant from base. Setae anteriores SI fringed, other setae simple. Mentum with a paired median tooth 19.0–19.8 µm and 5 lateral teeth. M.L. 30.7 µm, SM.S.D. 27.6 µm, distance between seta submenti and base of ventromental plates 11.9–20 µm. Mandible mola with small spines. Body with strong setae. Procerus very high 39.4 x 25.5 µm bearing a strong subapical seta 95.5–97.5 µm, often with a dark median area. Anal tubuli slender more than 1/2 long as posterior parapods. Supraanal seta 206 µm, long as posterior parapods, two short lateral setae 12.8–25.7 µm, subbasal seta 117 µm. Posterior parapods 147–249 µm with yellow claws. Sampling locality: Northern Italy Piemonte, Acqualba stream, 27.XII.78, Lombardia, Lambro stream st. 2, 27.XII.77, Trentino, Adamello glacier, Caré Alto st. 4, 1.VIII.96.

Tvetenia discoloripes

(Goetghebuer et Thienemann, 1936)

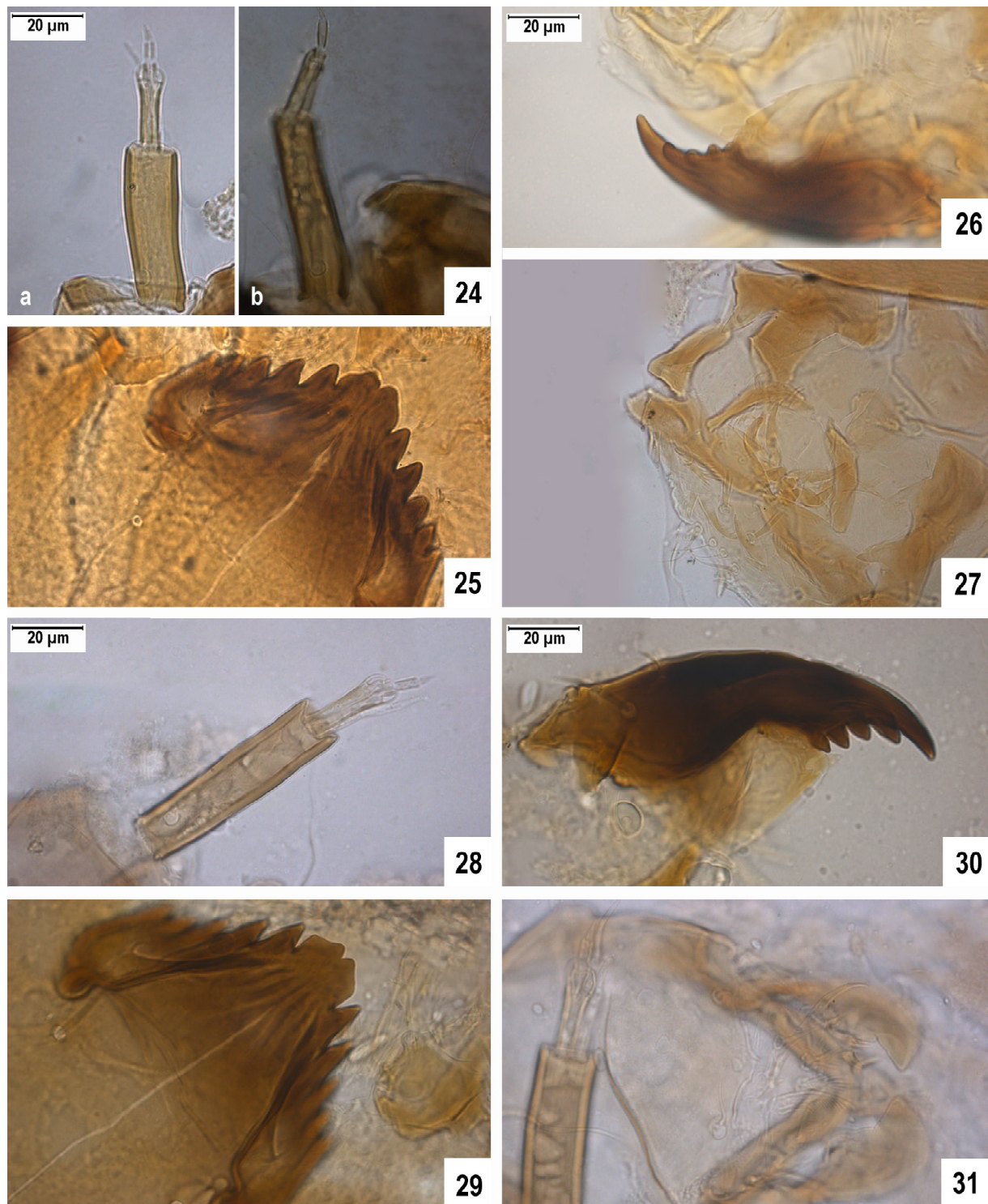
Figs 10–13, 28–31.

Large species, 5–6 mm, head capsule yellow or yellow brown. Body colour green or whitish. C.I. 1.56. Antenna with 5 segments, 1st segment longer than in *T. calvescens*, 3rd short, antennal blade reaching 5th segment. Antennal segments length 67.4, 19.7, 5.6, 6.0, 6.4 µm, AR 1.70, R.O. 10 µm, D.S.M. 42 µm from base, antennal blade 24.3 µm. Setae anteriores SI palmate, other setae simple. Large Lauterborn organs. Mentum with a paired median tooth 18.9 µm wide, and 5 lateral teeth. M.L. 85.5 µm, SM.S.D. 70.6 µm, distance between seta submenti and base of ventromental plates 6.4 µm. Mandible with inner margin smooth. Procerus dark well sclerotized, high about 1.5–2 higher than wide 41 x 31 µm, with a spur bearing a strong subapical seta 150 µm. Anal papillae large, less than 1/2 posterior parapods. Very long setae on the body, supraanal seta 248 µm, long as posterior parapods, subbasal seta 119–161 µm, about 1/2 long as parapods. Anal tubuli blistered. Posterior parapods 264 µm, claws of posterior parapods brown. The species cannot be separated by *T. verralli* in the larval stage. Sampling localities: Northern Italy, Piemonte, Acqualba stream, 2.IX.76.



Figs 1–23. Some total features of studied larvae (1–6) and larvae of *Tvetenia calvescens* (7–9), *T. discoloripes* (10–13), *Eukiefferiella minor* (14–15), *Tokunagaia rectangularis* (16–19), *E. brevicar* (20–21) and *E. claripennis* (22–23). 1 – abdomen; 2 – procerci; 3, 7, 10, 14, 16, 20, 22 – antenna; 4, 8, 11, 15, 17, 21, 23 – mentum, 5, 12, 18 – mandible; 6 – S setae anteriores S_p , $S_{III a}$ – simple setae anteriores S_{III} , $S_{III b}$ – bifid setae anteriores S_{III} ; 9, 13, 19 – labrum. ab – abdomen, p – procerci, an – antenna, mn – mentum, mb – mandible, lb – labrum. See text for other designations.

Рис. 1–23. Некоторые общие структуры изученных личинок (1–6) и личинок *Tvetenia calvescens* (7–9), *T. discoloripes* (10–13), *Eukiefferiella minor* (14–15), *Tokunagaia rectangularis* (16–19), *E. brevicar* (20–21) и *E. claripennis* (22–23). 1 – abdomen; 2 – подставка преанальной кисточки; 3, 7, 10, 14, 16, 20, 22 – антенна; 4, 8, 11, 15, 17, 21, 23 – ментум, 5, 12, 18 – мандибула; 6 – передние центральные щетинки S_p , $S_{III a}$ – простые щетинки S_{III} , $S_{III b}$ – двойные щетинки S_{III} ; 9, 13, 19 – лабрум. ab – abdomen, p – подставка преанальной кисточки, an – антенна, mn – ментум, mb – мандибула, lb – лабрум. Другие обозначения см. в тексте.



Figs 24–31. Larvae of *Tvetenia calvescens* (24a–27), *T. bavarica* (24b) and *T. discoloripes* (28–31). 24, 28 — antenna; 25, 29 — mentum; 26, 30 — mandible; 27, 31 — labrum.

Рис. 24–31. Личинки *Tvetenia calvescens* (24a–27), *T. bavarica* (24b) и *T. discoloripes* (28–31). 24, 28 — антенна; 25, 29 — ментум; 26, 30 — мандибула; 27, 31 — лабрум.

Tvetenia verralli (Edwards, 1929)

The species is here described from larvae associated with pupal exuviae and adults. It does not seem separable from *T. discoloripes* (see above). Antennal segments length 59.2–69.7, 17.8–22.8, 2.4–3.6, 4.8–6.2, 3.6–5.5 μm , AR 1.52–1.58, R.O. 10.4–13 μm , D.S.M. 37.3–41.2 μm from base, antennal blade 28.3–31.1. Setae anteriores S_{I} palmate, other setae simple. Large Lauterborn organs. Mentum with a paired median tooth 20.5–21.6 μm wide, and 5 lateral teeth, M.L. 84.5–88.5, SM.S.D. 60.1–69.6, distance between seta submenti and base of ventromental plates 11.7 μm . Mandible with inner margin smooth. Body with strong setae on abdominal segments, Procercus 38 x 30 μm , subapical seta 117 μm , supraanal seta 279 μm much longer than anal tubuli 100.1 μm long, subbasal seta 145.3 μm , anal tubuli 2/5 of posterior parapods, these 249.6–278.5 μm long. The species cannot be separated by *T. discoloripes* in the larval stage. Sampling locality: Northern Italy, Veneto, Brenta river, 30.VII.77; Emilia Romagna, Taro river, Compiano 1.VII.2002.

Tokunagaia rectangularis (Goetghebuer, 1940)

Figs 16–19, 32–35.

The larva of the genus was described in detail by Halvorsen and Sæther [1987]. Small species up to 5.5 mm. Head capsule 287 x 175 μm , yellow, light brown or dark brown, 287 x 230 μm , thorax yellow, abdomen violet. Antenna with 5 antennal segments, 37.9–48.9, 13.4, 1.9, 2.8–3.1, 1.8–3.6 μm , 1st and 2nd segment long, A_{2-5} 31.3 μm , AR 1.6–1.8, antennal blade shorter than 2nd not reaching 3rd segment, 3rd antennal segment very short, R.O. near the basis of antenna, 5.7 μm from base, well separated from D.S.M. 24.9 μm from base. Labrum with all setae anteriores simple. Mentum with a paired median tooth, 8.5–11.3 μm wide, when worn may appear simple, 5 lateral teeth. 1st lateral tooth poorly developed, M.L. 45–50.1 μm , SM.S.D. 28.4–34 μm . Mandible mola with small spines on inner margin. Abdominal setae long, about half long as abdominal segments. Procercus 22 x 12 μm , supraanal seta 43 μm , shorter than anal tubuli, subbasal seta 32 μm , anal tubuli 78.3–127 μm long, posterior parapods 226–246 μm , Sampling locality: Northern Italy Trentino, Caré Alto st. 8, 24.VI.96.

Tokunagaia tonolli (Rossaro, 1980)

Larva not separable from *T. rectangularis*. Head capsule yellow brown, 232 x 140 μm . Antenna with 5 antennal segments, 47.6, 10.8, 2.1, 3.8, 3.7 μm , A_{2-5} 26.6 μm , AR 1.7, antennal blade shorter than 2nd not reaching 3rd segment, 3rd antennal segment very short, R.O. near the basis of antenna, 5.7 μm from base, well separated from D.S.M., D.S.M. 24.9 μm from base. Mentum with a paired median tooth, 14.5 μm wide, 5 lateral teeth, 1st lateral tooth poorly developed, M.L. 54.8 μm , SM.S.D. 28.8 μm . Mandible mola with small spines on inner margin. Anal tubuli 106 μm , subbasal seta 55 μm , posterior parapods 258–285 μm . Sampling locality: Northern Italy Lombardia, Chiareggio, 6.XI.98.

Eukiefferiella brehmi Gowin, 1943

Andersen et al. [2013] includes in the key to *Eukiefferiella* groups the *brehmi* group, with S_{III} simple, antennal blade reaching the apex of the 4th segment. Mentum with paired median tooth and 5 laterals. No larvae assigned to this species were sampled.

Eukiefferiella brevicar (Kieffer, 1911)

Figs 20–21, 36–39.

Small species 3–4 mm, but larger than the similar *E. tirolensis*. Head capsule dark brown, length 260–290 μm ,

C.I. 1.53 [Schmid, 1993]. thorax yellow, abdomen violet. Antenna with 5 segments, 1st and 2nd segment long, 3rd very short. Length of antennal segments: 44–46.9, 13.8–17.0, 1.1–2.3, 2.9–3.3, 3.3–4.4 μm , antennal blade 14–16 μm long, shorter than 2nd segment, or reaching the 3rd segment [Zavřel, 1939], AR 1.47–1.60, R.O. 7.2–10.6 μm from base, D.S.M. 18.4 μm from base. Setae anteriores simple. Mentum with a single narrow median tooth 6.8–8.2 μm and 5 lateral teeth, M.L. 59.4 μm , SM.S.D. 34.6 μm . Mandible mola with 4 strong short spines. Body without strong setae. Procercus 29.4 x 18.9 μm , basal subapical seta 84.3 μm . Anal tubuli 124 μm . Segment 10th with a strong subbasal seta 70 μm long, supraanal and lateral setae very reduced. Posterior parapods 267 μm long. Sampling locality: Northern Italy, val D'Aosta, Dora di Veny, La Les Blanche st. 4–5, 7.IX.96, Northern Italy Trentino, Caré Alto, st. 3–5, 24.VI.96.

Eukiefferiella claripennis (Lund., 1898)

Figs 22–23, 40–43.

Small species 3–4 mm, head capsule dark brown, length 241 μm , C.I. 1.35 [Schmid, 1993], thorax yellow, abdomen light brown. Antenna with 4 segments, 3rd very short, length of antennal segments 50.6–61.3, 12.9–15.8, 1.6–2.3, 4.3–5.1 μm , A_{2-4} 27.9. AR 1.9–2.2, antennal blade 15–21.4 μm reaching 3rd segment. Lauterborn organ small surrounding 3rd segment, R.O. 8.8–11.25 μm , D.S.M. 33.5–35–37.3 μm distant from base. Setae anteriores simple. Mentum with a narrow paired median tooth 10.4–11.7 μm wide, each tooth about 5 μm large, 5 lateral teeth, M.L. 68.9 μm , SM.S.D. 45.1 μm . Mandible mola with 4 short spines. Body without strong setae at most 60 μm long. Procercus 20.7–31.9 x 20.5–22.5 μm heavily sclerotized, apical setae on procercus 480 μm . Anal tubuli 75 μm , 10th abdominal segment with a strong supraanal seta 95 μm and a strong subbasal seta 45–68 μm . Posterior parapods, 174–390 μm . Sampling locality: Northern Italy, Lombardia, Oglio river, Vezza, 12 8 79, brook above Vezza, 2.I.79, Central Italy, Lazio, Mignone stream 29.V.80.

Eukiefferiella clypeata (Thienemann, 1919)

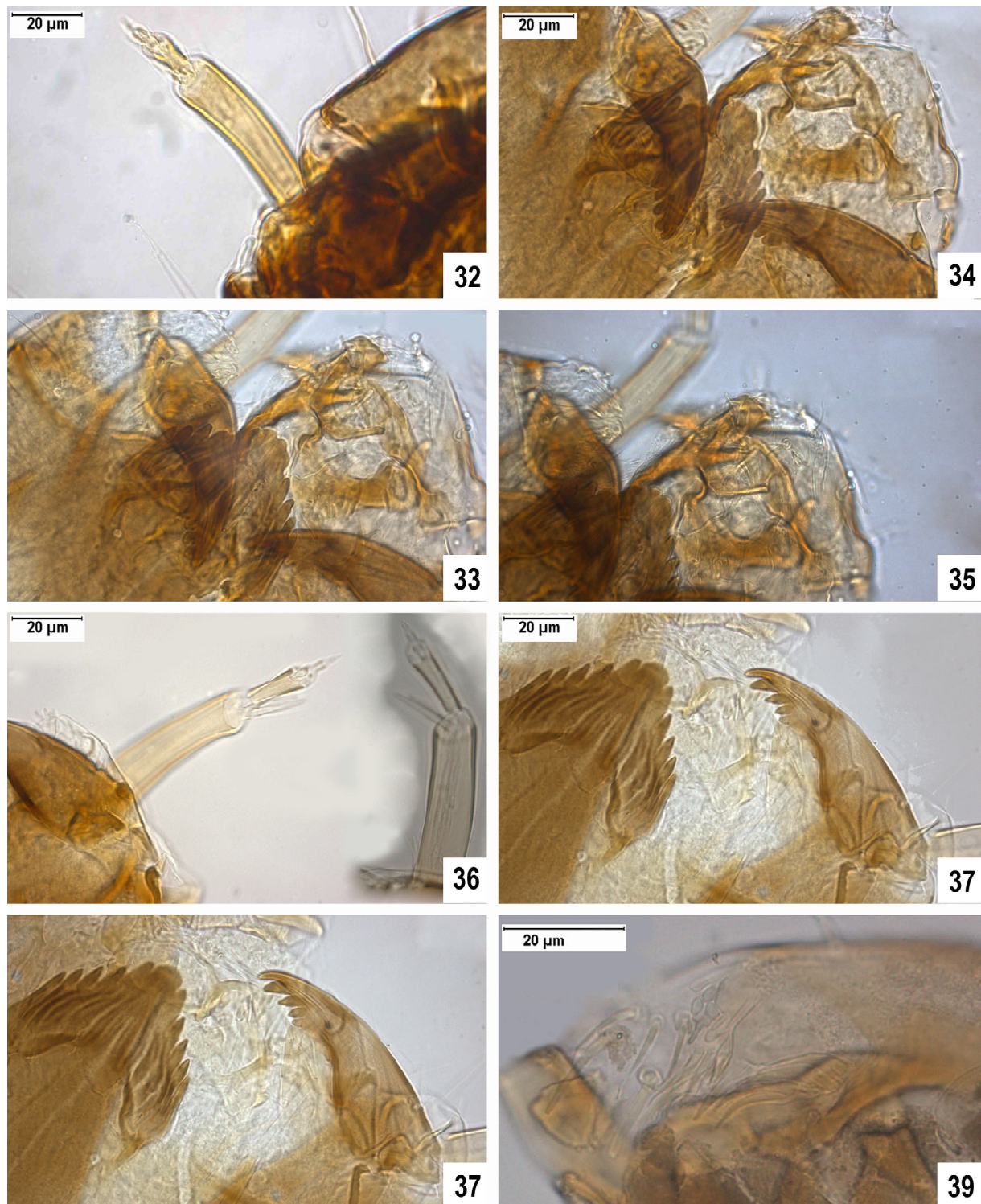
Figs 44–47, 83–85.

Large species 4–5 mm, head capsule light brown, body colour yellow. C.I. 1.64 [Schmid, 1993]. Antenna with 4 segments, 2nd and 3rd segment long, length of antennal segments: 45.6–54.8, 15.6–18.8, 2.7–3.1, 4.4–4.7 μm , A_{2-4} 33.8 μm , AR 1.6–1.8, antennal blade 18.8–20 μm not overreaching the 2nd segment. R.O. 7.7–9 μm , D.S.M. 29.4–37.3 μm from base. Setae anteriores simple. Mentum with a large paired median tooth 19.4–20.9 μm and 5 lateral teeth, first lateral tooth 3.4–4.5 μm . M.L. 69.7 μm , SM.S.D. 49.5 μm . Mandible mola with spines. Body without strong setae. Procercus 19 x 18 μm , basal subapical seta 38–46 μm , 10th abdominal segment with a strong subbasal seta 30–33 μm long. Anal tubuli 85–93 μm long, posterior parapods 228–232 μm . Sampling locality: Northern Italy Lombardia, Ticino river, Boffalora, 2.IX.79.

Eukiefferiella coerulescens (Kieffer, 1926)

Figs 48–51, 80–82.

Small species 3–4 mm. Setae anteriores S_{III} simple. C.I. 1.32 [Schmid, 1993]. Antenna with 5 antennal segments, but with 4 segments according to Schmid [1993] and Epler [2001], with 2nd antennal segment long, 3rd and 4th short, subequal. In our samples antennal segments length: 46.6–48.1, 19.8, 2.2, 2.6, 1.9, A_{2-5} 33.8 μm , AR 1.4–1.5 μm , antennal blade 21.8 μm beyond the apex of the 2nd segment. R.O. 8.5 μm ,



Figs 32–39. Larvae of *Tokunagaia rectangularis* (32–35) and *Eukiefferiella brevicealcar* (36–39). 32, 36 — antenna; 33, 37 — mentum; 34, 38 — mandible, 35, 39 — labrum.

Рис. 32–39. Личинки *Tokunagaia rectangularis* (32–35) и *Eukiefferiella brevicealcar* (36–39). 32, 36 — антенна; 33, 37 — ментум; 34, 38 — мандибула, 35, 39 — лабрум.

D.S.M. 18.4–19.5 μm from basis of 1st antennal segment, distal setal mark near to R.O. [Schmid, 1993]. Mentum with a paired very prominent median tooth 7.9 μm and 5 lateral teeth, the 1st lateral tooth appears as an appendix of the median tooth [Andersen et al., 2013, Fig. 9.27C], M.L. 48.2 μm , S.M.S.D. 30.9 μm . Procercus light-brown 28 x 17 μm , with a small spur on median margin. Anal tubuli 88–94 μm . Supraanal seta small, strong subbasal seta 63 μm long. Posterior parapods very long, 327 μm . Claws of posterior parapods yellow brown. Sampling locality: Northern Italy Trentino, Caré Alto st. 7, 1.VIII.96

Eukiefferiella cyanea Thienemann, 1936

Figs 52–55, 86–88.

Antenna with 5 segments, 2nd segment short, 3rd and 4th subequal, antennal blade reaching segment 4th. Antennal segments: 43.5–50.1, 9.5–12.7, 5.0–6.3, 3.3–5.7, 3.1–6.8 μm , AR 1.4–1.6, antennal blade 26.9 μm , R.O. 10.0 μm distant from base, D.S.M. 30.1 μm from base. Mentum with very large median tooth 23–28.7 μm and 5 laterals, M.L. 66.3 μm , S.M.S.D. 77.0 μm . Procercus small 9.0 x 9.9 μm , apical setae short 80–118 μm (Zavřel 1939). 10th segment without strong setae. Anal tubuli 117–189 μm . Posterior parapods very long 439–711 μm with 3 short claws and about 8 long ones. Sampling locality: Northern Italy Lombardia, Valtellina, Viola stream, 27.VI.85, South Italy Puglia, Celone stream, 15.IX.90.

Eukiefferiella devonica (Edwards, 1929)

Figs 56–59, 89–91.

According to Schmid [1993] not separable from *E. ilkleyensis* except for the smaller size of head capsule (310–360, mean = 341 μm). In the examined larvae (Scoffera pass, 29.II.80) associated to pupal exuviae, we observed simple S_{III} , antenna with 4 antennal segments, mentum with 5 lateral teeth. C.I. 1.35 [Schmid, 1993] Antennal segments 40.0–53.4, 13.0–14.5, 2.5–4.5, 2.4–5.2, A_{2-4} 26.7 μm , AR 1.58–1.66, Lauterborn organ well developed, antennal blade 19.4–22.2 μm long, reaching the half or the apex of 3rd segment, R.O. 5.3–11.9 μm from base, D.S.M. 28.8–38.4 μm . Mentum has 5 lateral teeth, the median tooth is 10.9–20.2 μm wide, but it can be fused with the 1st lateral tooth, resulting in an apparent much larger tooth, with only 4 lateral teeth. M.L. 49.8–62.6 μm , S.M.S.D. 32–44.4 μm . Mandible mola with short spines, seta interna not divided at base [Epler, 2001]. Procercus 15–23 x 12–22 μm , supraanal seta small about 15 μm , subbasal seta 29–64 μm , anal tubuli 63 μm long. Posterior parapods 228–286 μm . Sampling locality: Northern Italy Emilia, Taro river 12.XI.2002, Liguria, stream near Scoffera pass, 29.II.1980, Central Italy Abruzzo, Raiale, 11.VIII.1995.

Eukiefferiella fittkaui Lehmann, 1972

Not separable from *E. minor*. C.I. 1.49 [Schmid, 1993]. Larvae associated to pupal exuviae: antennal segments 53.1, 14.8, 5.1, 5.2, 5.0 μm , AR 1.44, antennal blade 32.9 μm , R.O. 5.1 μm D.S.M. 34.7 μm . Mentum with a large median tooth 23.7 μm and 5 laterals, M.L. 68.2 μm , S.M.S.D. 71.8 μm . Abdominal segments with strong setae 125–148 μm . Procercus 30 x 27 μm , apical setae on procercus 239–312 μm , subapical seta 119–131, supraanal seta 164–181 μm , lateral seta 34–57 μm , subbasal seta 93–106 μm , anal tubuli 124–155 μm . Posterior parapods 212–228 μm . Sampling locality: Northern Italy, Lombardia, brook in Bighera valley, tributary of Oglio river, 8.X.79.

Eukiefferiella fuldensis Lehmann, 1972

Figs 60–63, 92–93.

Small species 3–4 mm, body colour light, head capsule light brown, slender. C.I. 1.55. Antenna with 4 segments, length of antennal segments: 36.6–43.9, 10–11.4–12, 4–6, 3.8–5.4 μm , A_{2-4} 25.3–28.8 μm , AR 1.3–1.45. Antennal blade reaching 3rd segment, 15.5–20.5 μm long, 3rd antennal segment long. R.O. 7–8.8 μm from base, D.S.M. 26–29.4 μm from the base of 1st antennal segment. Distal setal mark is far from R.O. at least 6 μm [Schmid, 1993]. Mentum with a narrow paired median tooth 11.6 μm , and 5 laterals, but the median tooth can be worn out appearing as a single tooth. M.L. 50.8 μm , S.M.S.D. 30.1 μm . Mandible mola with fine spines on inner margin. Body without strong setae. 10th segment without strong anal setae, supraanal seta 14–17 μm , strong subbasal seta 42 μm long. Anal tubuli about 87–100 μm long. Posterior parapods 263–295 μm long. Sampling locality: Northern Italy Trentino Caré Alto st. 4, 24.VI.96.

Eukiefferiella gracei (Edwards, 1929)

Figs 64–67, 94–95.

Head yellow, only distal mentum and mandible dark brown, Anterior eye kidney shaped. C.I. 1.67 [Schmid, 1993]. Antennal segments 47–54, 11.1–12.4, 5.4–6.2, 5–6, 3.4–3.9 μm , A_{2-5} 30.9 μm , AR 1.51–1.75, antennal blade 25.7, R.O. 4.8–10.6 μm and D.S.M. 34.7–36.2 μm (ratio D.S.M./ A_1 0.64–0.67) far from base, about from base. M.L. 93.3 μm , S.M.S.D. 83.0 μm , with median tooth 22.0–24.9 μm wide, 5 lateral teeth 5.4–5.7, 5.2–6.2, 5.5–6.2, 5–5, 3.6–4 μm . Mandible mola with three inner teeth and three long setae on mola. Supraanal seta 37 μm , subbasal seta 41 μm , anal tubuli 82 μm . Posterior parapods 190 μm . Sampling locality: Northern Italy Piemonte, Lombardia, Ticino river 26.I.2001, 30.IV.2001, Central Italy Toscana, I.Elba San Francesco rio Calanche, 1.VIII.80.

Eukiefferiella ilkleyensis (Edwards, 1929)

Figs 68–71, 96–98.

Large species 4–5 mm, abdomen yellow light or green, thorax yellow, head capsule dark brown. Head capsule length about 370–430, m = 406 μm , C.I. 1.35 [Schmid, 1993]. Antenna with 5 segments, 4th segment (7.8–7.9 μm) much longer than 3rd (4.2–4.3 μm). Antennal blade 26.2–28.1–29.6 μm reaching the 4th segment. Length of antennal segments: 44.7–52.4, 11.5–14.4, 4.2–6.9, 7.6–7.9, 5.8–7.3 μm , A_{2-5} 37.5 μm , AR 1.2–1.3. R.O. 5–6.4–6.8–7.6 μm , D.S.M. 29.8–29.9–33–34.9 μm distant from the basis. Setae anteriores S_{I-II} simple, S_{III} bifid. Mentum with black margin, darker than basal part, it has a large median tooth (27.0–35.5 μm) and 4 laterals. M.L. 73.0 μm , S.M.S.D. 43.2 μm . Mandible mola with 3 inner teeth and mola with spines, seta interna not divided at base [Epler, 2001]. Abdomen with strong setae 100–130 μm long. Procercus 19.3 x 28 μm with a tuft of 7 apical setae up to 300 μm long. Anal tubuli 90–170 μm , shorter than posterior parapods. 10th abdominal segment with a 101–150 μm supraanal seta, 80 μm lateral seta and a 68–100 μm subbasal seta. Posterior parapods 170–240 μm long. Sampling locality: Northern Italy Piemonte, Agogna stream st. 1, 28.VII.81, Central Italy, Marche, Aso stream, 28.III.79.

Eukiefferiella lobifera Goetghebuer 1934

Figs 72–75, 99–102.

Head capsule dark. Torax yellow Abdomen bleu. C.I. 1.43 [Schmid, 1993]. Antenna with 5 segments: 49.8–61.8,

12–14.7, 1.5–1.8, 3.4–4.1, 2.5–5 μm , AR 1.86–1.92, R.O. 7.4 μm from base, D.S.M. 32.4–35, μm from base. Antennal blade 18.2 μm , longer than segment 2, reaching the 3rd–4th segment [Zavřel, 1939]. Mentum with a paired median tooth [Schmid, 1993], but in the examined specimens a single median tooth is visible [Zavřel, 1939], 8.4 μm wide with five lateral teeth. M.L. 60.7–65 μm , S.M.S.D. 38.4–38.5 μm . Mandible mola with four spines. Body with few strong setae at most 40 μm . Claws of anterior parapods yellow. Procercus with a median sclerotized area 21–28 x 18 μm , subapical seta of procercus 69 μm , tuft of apical setae 274 μm . Supraanal seta 60 μm , lateral anal seta 22 μm , ventral subbasal seta 30–37 μm . Anal tubuli 100–120 μm . Posterior parapods 260–330 μm long. Sampling locality: Northern Italy, Friuli, Tagliamento river, S. Daniele, 15.VI.97, Liguria, Quiliano 19.XII.98.

Eukiefferiella minor (Edwards, 1929)

Figs 14–15, 76–79.

Large species 4–6 mm, head capsule dark brown, abdomen yellow light or green, thorax yellow. Body with strong setae about 110 μm long. Anterior eye kidney shaped, on concave side trilobed. C.I. 1.45. Antenna with 5 segments. Antennal blade 25.6 μm reaching the 4th segment. Length of antennal segments 47.8–59.4, 12–18.4, 2.5–5, 5.8–6.5, 3.3–5, A₂₋₅ 38.4, AR 1.46–1.69. R.O. 6.7–7.4 μm , D.S.M. 22.2–37.8 μm from base, about 0.62–0.66 from base. Setae anteriores simple. M.L. 77.7–88.3 μm , S.M.S.D. 79–85.4 μm with a large median tooth 24.9–31.7 μm , larger than the first 3 lateral teeth together and 5 laterals. Mandible mola with long setae instead of spines. Abdominal segments with strong setae 120–130 μm long. Procercus 30–31 x 14–21 μm with a sclerotized area bearing a strong basal subapical seta about 80 μm . 10th abdominal segment on anal margin with a strong supraanal seta 150–170 μm long, a shorter lateral seta 35–60 μm and subbasal seta 80–93 μm . Anal tubuli 90–140 μm . Posterior parapods 203–220 μm . Sampling locality: Northern Italy, Aosta Valley, Dora Veny, 13.IX.97, Lombardia, Oglio river, Vezza, 28.IV.78, Trentino, Caré Alto st. 7, 24.IX.96, Southern Italy, Sicilia, Dirillo stream, 4.X.83.

Eukiefferiella similis Goetghebuer, 1939

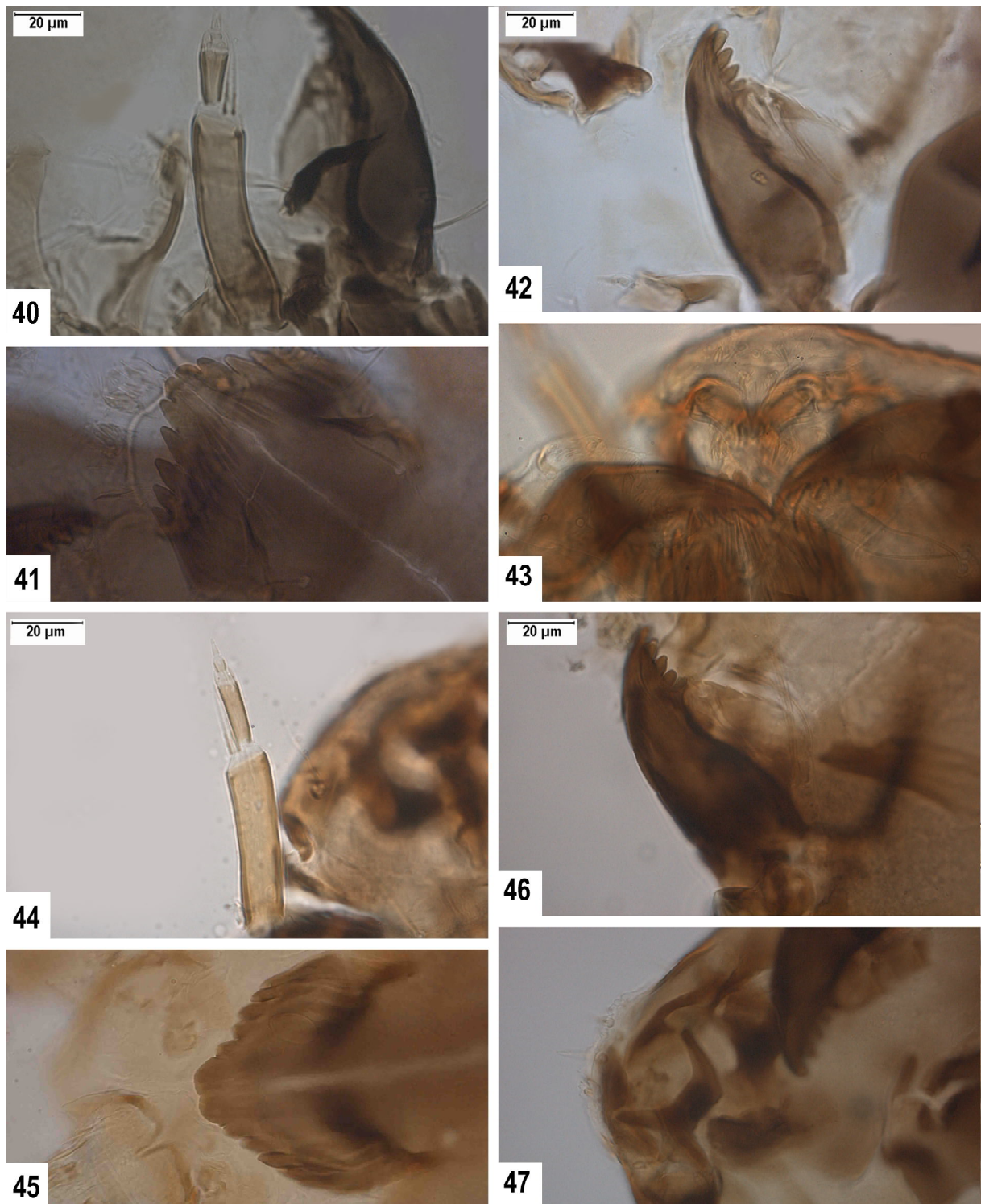
The species was described as separable from the similar *E. minor* because of a distinct hump in the middle of median tooth of mentum [Schmid, 1993], eyespot shape, distance between R.O.–D.S.M. and presence of a sclerotization on procercus [Zavřel, 1939]. Large species 4–6 mm, head capsule dark brown, mentum entirely black, thorax yellow, abdomen yellow light or green. Antenna with 5 segments, length of antennal segments 48.2–50.2, 11.9–13.4, 3.0–3.1, 3.7–4.2, 4.4–5.0 μm , A₂₋₅ 33.2–37.6 μm , AR 1.7–1.8, antennal blade 26.7–27.7 μm reaching the 4th segment, R.O. 6.5–9.6 μm , D.S.M. 24.9–34 μm from base. Setae anteriores simple. M.L. 70–74.2 μm , S.M.S.D. 70–80 μm with a large median tooth 24–25.9 μm , wider than the first 3 lateral teeth together and 5 laterals, hump not visible in the examined specimens. Mandible mola with very long setae instead of spines. Body with strong setae about 85–110 μm long. Procercus 35 x 18 μm , 10th abdominal segment on anal margin with a strong supraanal seta 84–153 μm , a shorter lateral 39 μm and subbasal seta 51–53 μm . Anal tubuli 35.9. Posterior parapods 127 μm . Sampling locality: Northern Italy Lombardia, Chiareggio 14.V.99.

Eukiefferiella tirolensis Goetghebuer, 1938

Larva not separable from *E. minor*, except for smaller size, 2–3 mm. C.I. 1.27 [Schmid, 1993]. Labrum with Simple S_{III}, mentum with narrow median tooth and 5 lateral teeth, antennal blade reaching the 4th segment, head capsule light-brown, 200–240 μm [Schmid, 1993].

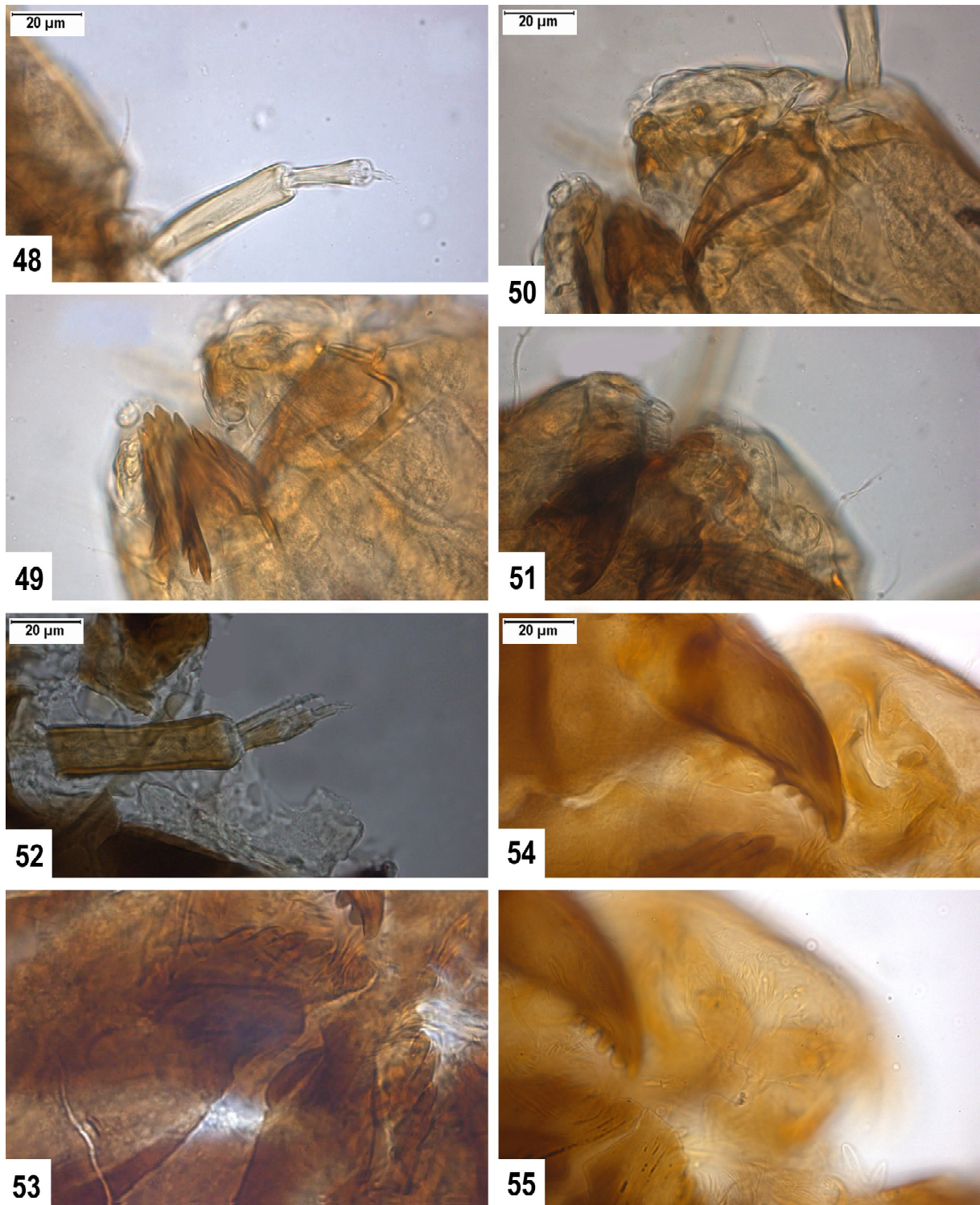
KEY TO SPECIES

1. Setae S_I branched, fringed or palmate (Figs 6, 9), head capsule yellow light, procercus very developed, abdomen with very strong setae (Figs 1–2) ... *Tvetenia* 2
- Setae S_I simple (Figs 6, 19), head capsule generally dark, rarely yellow with dark areas, apical setae on procercus and abdominal setae generally developed, sometime reduced 4
2. Mola of mandible with smooth inner margin (Fig. 12), seta submenti near to posterior margin of ventromental plates (Fig. 30), S_I palmate (Figs 13, 31), body violet *discoloripes* (Goetghebuer in Thienemann., 1936), *verralli* (Edw., 1929)
- Mola of mandible with toothed inner margin (Fig. 5), seta submenti far from posterior margin of ventromental plate (Fig. 25), S_I divided into short fine branches only at apex (Fig. 27), body yellow-green 3
3. Ratio between 4th and 3rd antennal segments high (> 3) .
..... *bavarica* (Goethebuer, 1934)
- Ratio between 4th and 3rd antennal segments lower (< 3) (Fig. 24) *calvescens* (Edwards, 1929)
4. Posterior parapods very long > 400 μm (Fig. 105), apical setae of procercus short (about 100 μm), very large median tooth (Fig. 87) *cyanea* Thienemann, 1936
- Posterior parapods shorter (Fig. 103), apical setae of procercus much longer 5
5. Mentum with 4 lateral teeth (Fig. 69), setae anteriores S_{III} bifid (Figs. 6, 71), antenna with 5 segment (Fig. 68), antennal blade reaching the 4th segment, mentum with very dark distal margin *ilkleyensis* (Edwards, 1929)
- Mentum with 5 lateral teeth, but the mentum can be worn out, so a very large median tooth and 4 laterals are simulated 6
6. Antenna with 4 segments (Figs 22, 40, 56, 60, 92) 7
- Antenna with 5 segments (Figs 7, 10, 14, 16, 20, 32, 36, 44, 48, 64, 76, 72) 9
7. 3rd antennal segment long (Figs 89, 92) 8
- 3rd antennal segment very short, with rounded Lauterborn organ, head dark (Figs 22–23, 40–43) *claripennis* (Lundbeck, 1898)
8. Median tooth simple (Figs 56–59, 89–91) *devonica* (Edwards, 1929)
- Median tooth bifid, head capsule dark, but lighter than in *E. claripennis*, small larva, sometime the median tooth is worn out and appears as simple (Figs 60–63, 92–93) ... *fuldensis* Lehmann, 1972
9. Mentum with simple median tooth, sometime a small notch may simulate a bifid one (Figs 95, 100) 10
- Mentum with clearly paired median teeth, even if, when worn, can be misinterpreted as simple (Figs 81, 84) 13
10. Median tooth very large (Figs 65, 77), large as the first 3 lateral teeth; long abdominal setae, antennal blade well beyond the 2nd segment (Figs 4, 12, 14, 16) 11
- Median tooth narrow (Figs 73, 37), 3rd–5th antennal segments very short (Figs 5–7, 15–16) 12



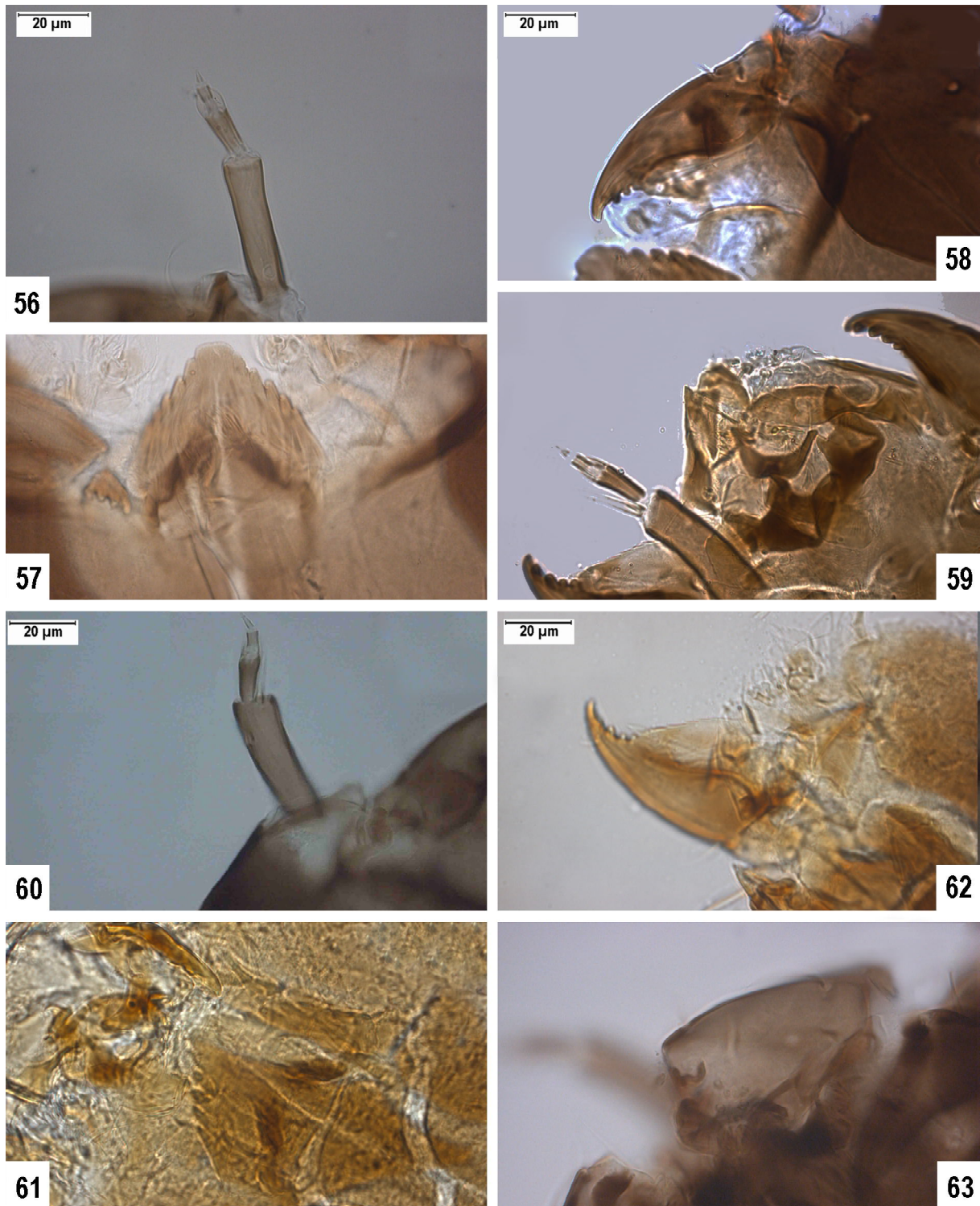
Figs 40–47. Larvae of *Eukiefferiella claripennis* (40–43) and *E. clypeata* (44–47). 40, 44 — antenna; 41, 45 — mentum; 42, 46 — mandible; 43, 47 — labrum.

Рис. 40–47. Личинки *Eukiefferiella claripennis* (40–43) и *E. clypeata* (44–47). 40, 44 — антенна; 41, 45 — ментум; 42, 46 — мандибула; 43, 47 — лабрум.



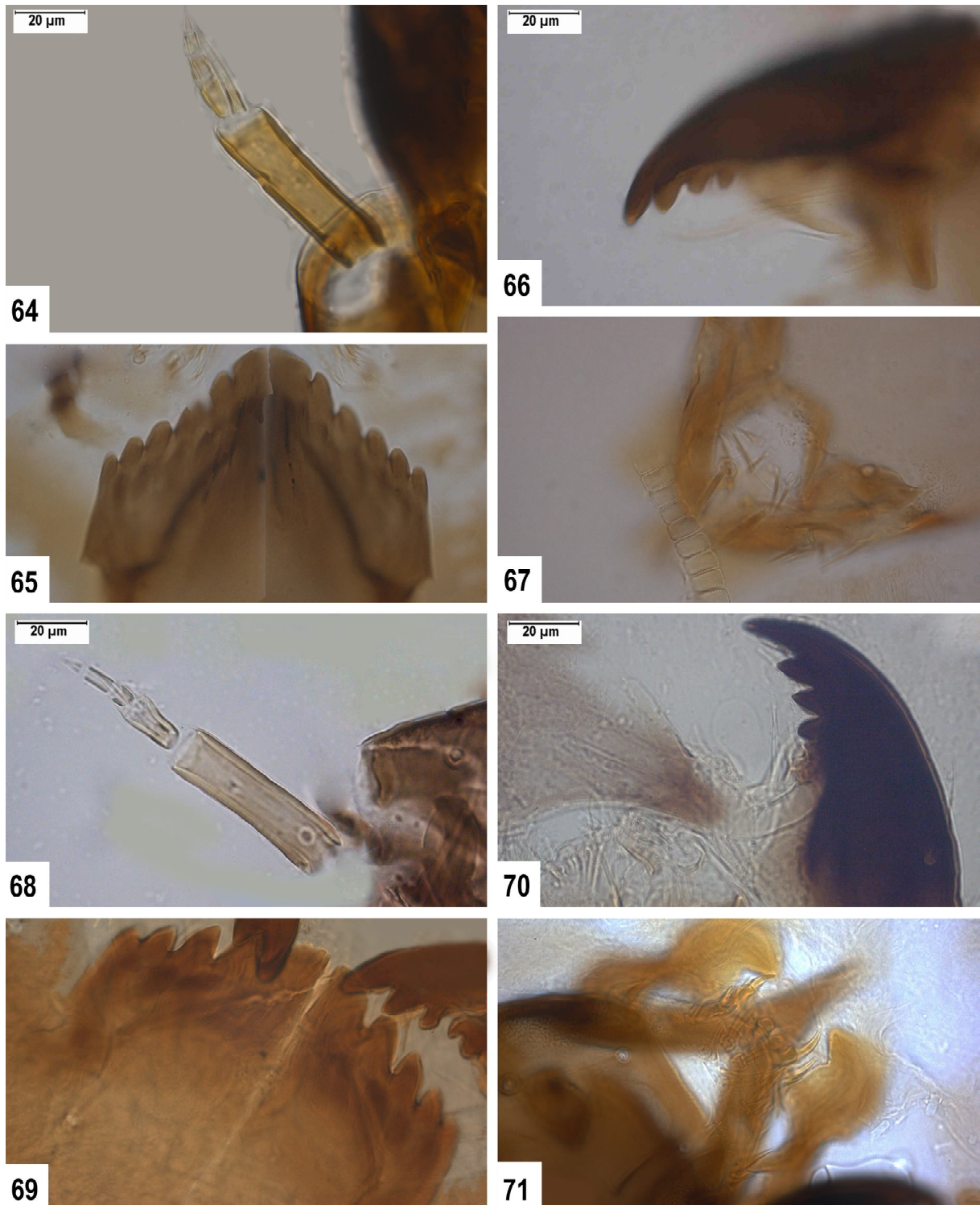
Figs 48–55. Larvae of *Eukiefferiella coerulea* (48–51) and *E. cyanea* (52–55). 48, 52 — antenna; 49, 53 — mentum; 50, 54 — mandible; 51, 55 — labrum.

Рис. 48–55. Личинки *Eukiefferiella coerulea* (48–51) и *E. cyanea* (52–55). 48, 52 — антенна; 49, 53 — ментум; 50, 54 — мандибула; 51, 55 — лабрум.



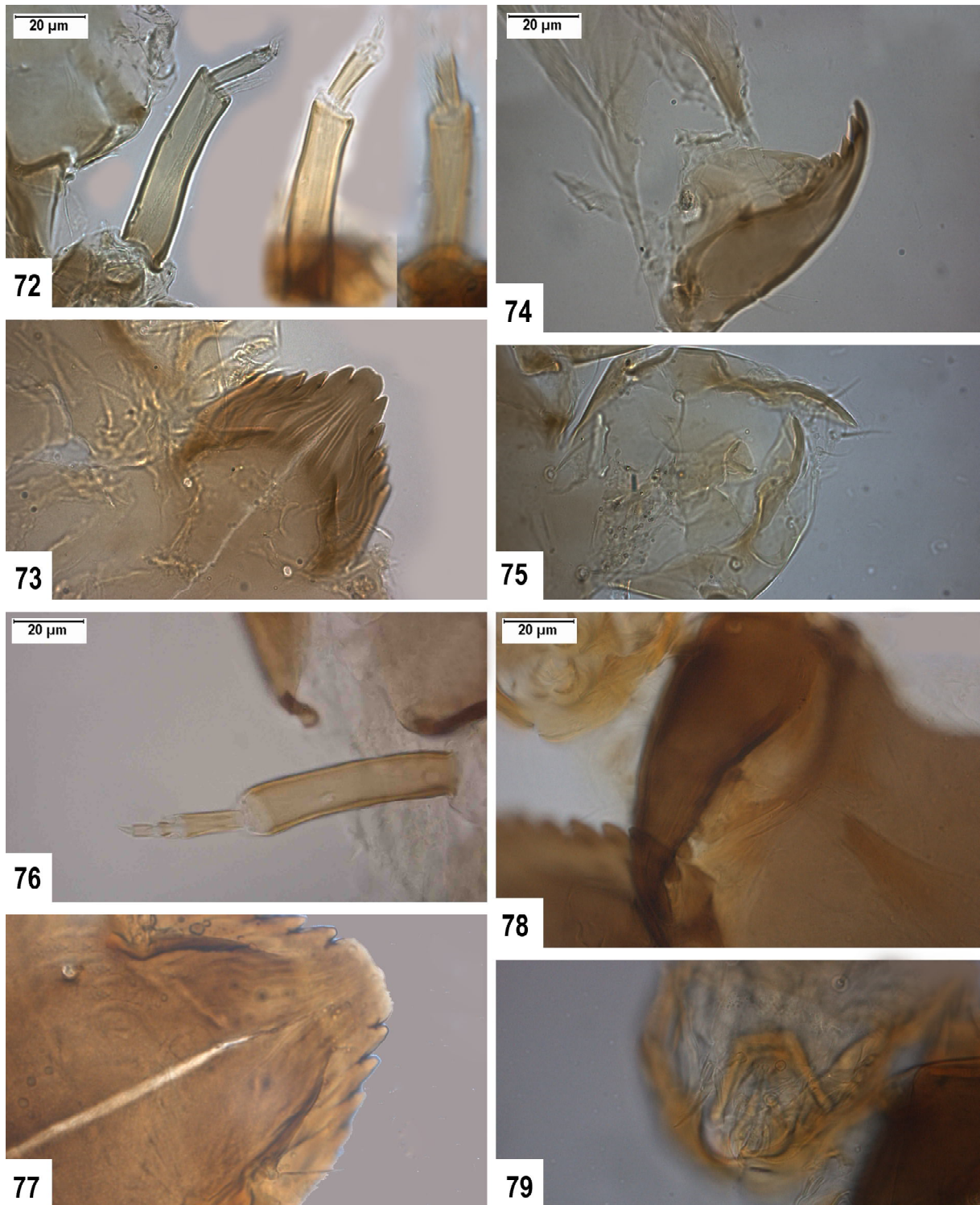
Figs 56–63. Larvae of *Eukiefferiella devonica* (56–59) and *E. fuldensis* (60–63). 56, 60 — antenna; 57, 61 — mentum; 58, 62 — mandible; 59, 63 — labrum.

Рис. 56–63. Личинки *Eukiefferiella devonica* (56–59) и *E. fuldensis* (60–63). 56, 60 — антенна; 57, 61 — ментум; 58, 62 — мандибула; 59, 63 — лабрум.



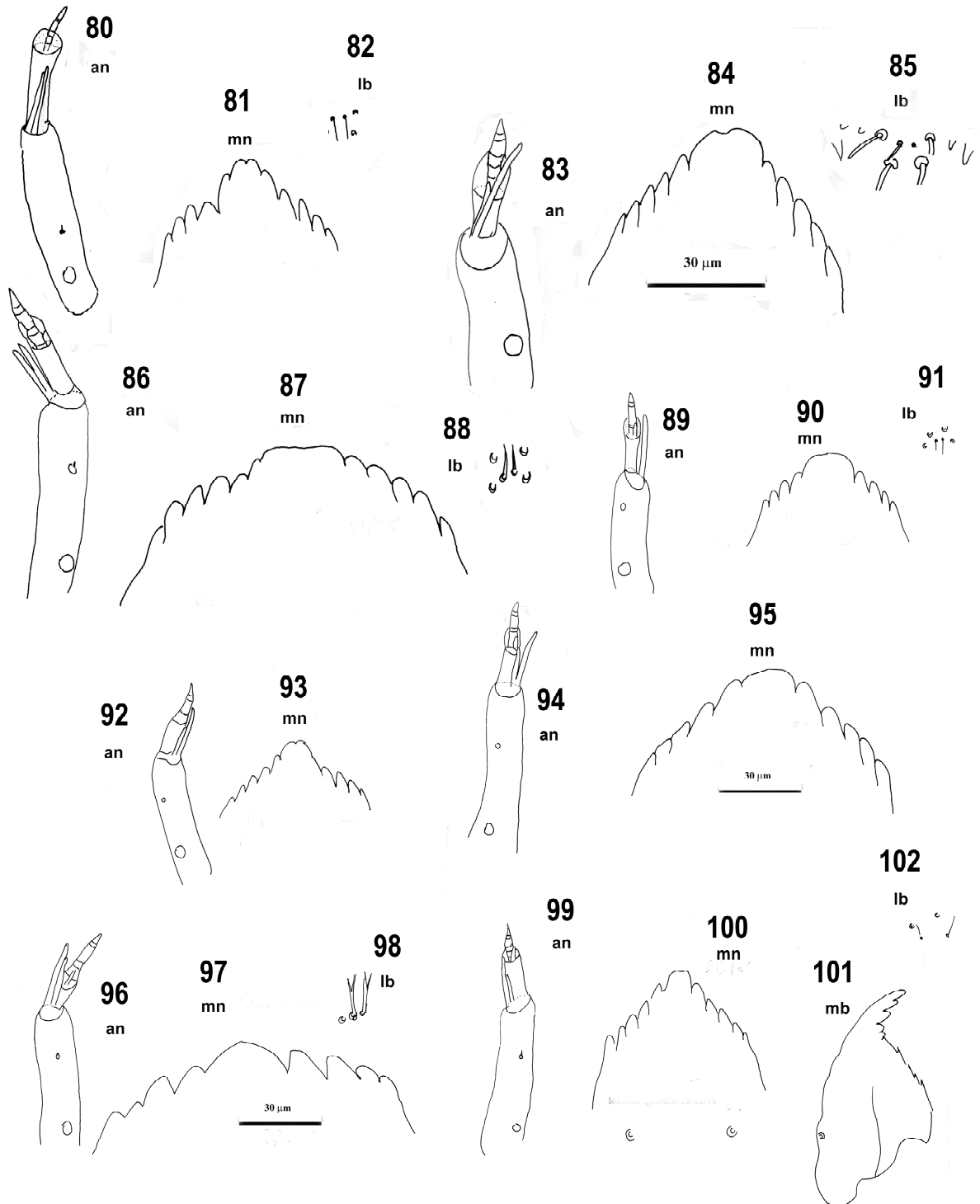
Figs 64–71. Larvae of *Eukiefferiella gracei* (64–67) and *E. ilkleyensis* (68–71). 64, 68 — antenna; 65, 69 — mentum; 66, 70 — mandible, 67, 71 — labrum.

Рис. 64–71. Личинки *Eukiefferiella gracei* (64–67) и *E. ilkleyensis* (68–71). 64, 68 — антенна; 65, 69 — ментум; 66, 70 — мандибула, 67, 71 — лабрум.



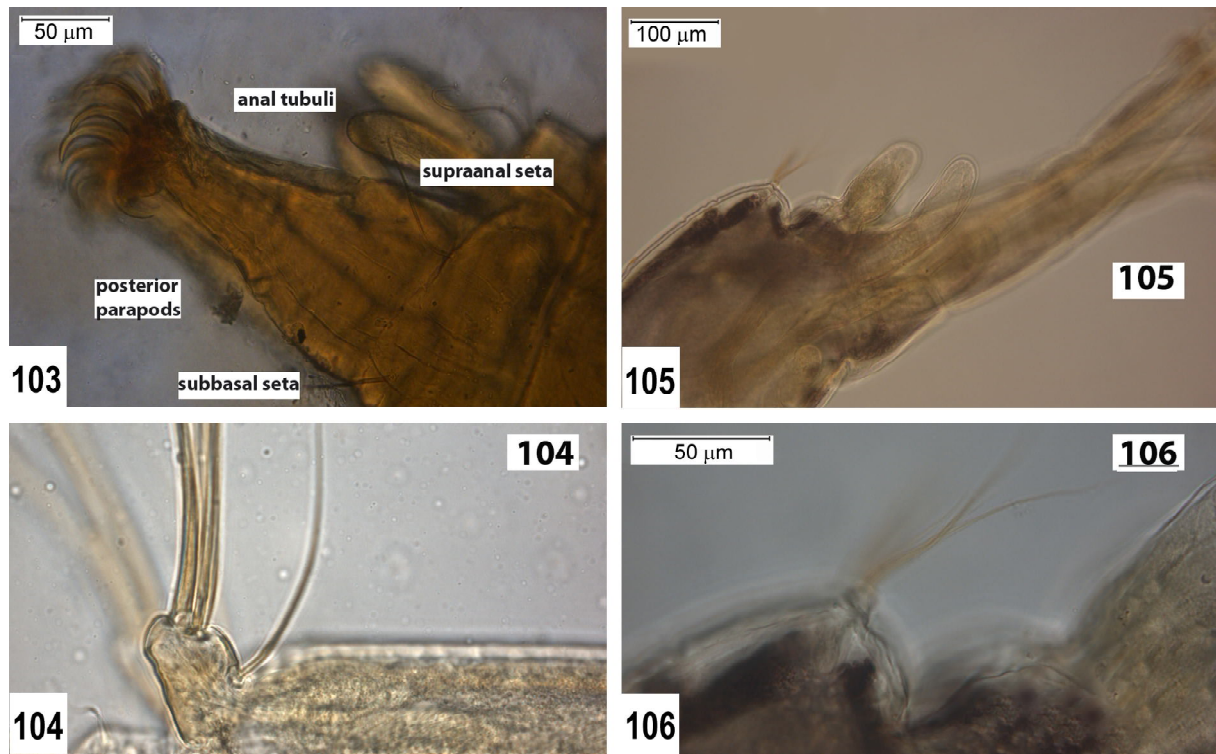
Figs 72–79. Larvae of *Eukiefferiella lobifera* (72–75) and *E. minor* (76–79). 72, 76 — antenna; 73, 77 — mentum; 74, 78 — mandible; 75, 79 — labrum.

Рис. 72–79. Личинки *Eukiefferiella lobifera* (72–75) и *E. minor* (76–79). 72, 76 — антенна; 73, 77 — ментум; 74, 78 — мандибула; 75, 79 — лабрум.



Figs 80–102. Larvae of *Eukiefferiella coerulescens* (80–82), *E. clypeata* (83–85), *E. cyanea* (86–88), *E. devonica* (89–91), *E. fuldensis* (92–93), *E. gracei* (94–95), *E. ilkeleyensis* (96–98) and *E. lobifera* (99–102). 80, 83, 86, 89, 92, 94, 96, 99 — antenna; 81, 84, 87, 90, 93, 95, 97, 100 — mentum; 82, 85, 88, 91, 98, 102 — labrum; 101 — mandible. Abbreviations are the same as in Figs 1–23.

Рис. 80–102. Личинки *Eukiefferiella coerulescens* (80–82), *E. clypeata* (83–85), *E. cyanea* (86–88), *E. devonica* (89–91), *E. fuldensis* (92–93), *E. gracei* (94–95), *E. ilkeleyensis* (96–98) и *E. lobifera* (99–102). 80, 83, 86, 89, 92, 94, 96, 99 — антенна; 81, 84, 87, 90, 93, 95, 97, 100 — ментум; 82, 85, 88, 91, 98, 102 — лабрум; 101 — мандибула. Обозначения те же, что на рисунках 1–23.



Figs 103–106. Larvae of *Eukiefferiella minor* (103), *E. brevicealcar* (104) and *E. cyanea* (105–106). 103 — caudal end; 104 — procerci; 105 — posterior parapods; 106 — procercal setae.

Рис. 103–106. Личинки *Eukiefferiella minor* (103), *E. brevicealcar* (104) и *E. cyanea* (105–106). 103 — задний конец тела; 104 — подставки преанальной кисточки; 105 — задние подталкиватели; 106 — щетинки преанальной кисточки.

11. Head yellow (Figs 64–67, 94–95) *gracei* (Edwards, 1929)
— Head dark brown (Figs 14–15, 76–79)
..... *minor* (Edwards, 1929), *fittkaii* Lehmann, 1972,
similis Goetghebuer, 1939
12. Antennal blade beyond 2nd segment (Figs 72–75, 99–102)
..... *lobifera* Goetghebuer, 1934
— Antennal blade shorter than 2nd segment (Figs 20–21, 36–
39)
..... *brevicalcar* (Kieffer, 1911), *tirolensis* Goetghebuer, 1938
13. Paired median tooth large, about 20 µm, 1st lateral well
separated from median tooth, head capsule light with small
dark areas (Figs 44–47, 83–85)
..... *clypeata* (Thienemann, 1919)
— Paired median teeth narrower (Figs 17, 81) ... 14
14. D.S.M. far from R.O., 5 antennal segments, 3, 4, 5th
segments very short (Figs 16–19, 32–35)
..... *Tokunagaia* Sæther, 1973
— D.S.M. near to R.O. (Figs 48–51, 80–82)
..... *coerulescens* (Kieffer, 1926)

A dendrogram summarizing the key to species is in Fig. 107.

Discussion

In the present work larvae belonging to *Eukiefferiella* and related genera *Tvetenia* and *Tokunagaia* are described and figured. Species whose larvae are not well known are not considered in this work. The key to species groups given by Cranston et al [1983], Anders-

en et al. [2013] and the species descriptions given in Zavřel [1939], Chernovskii [1949], Schmid [1993] and Epler [2001] are emended. Some characters, as the length of the antennal blade, the length of antennal segments, the presence of 1 or 2 median teeth of mentum, are sometime difficult to use, because require that the antenna be mounted in a quite horizontal plane and that the mentum be not worn out, often the examined specimens do not meet these requests. The length of the antennal blade was used in the present key to separate *E. lobifera* from *E. brevicealcar* [Zavřel, 1939], instead of the presence of a mentum with 2 median teeth in *E. lobifera* and 1 median tooth in *E. brevicealcar* [Schmid, 1993], having observed, as figured in Zavřel [1939, Fig. 3D], that *E. lobifera* has 1 median tooth; unfortunately, the antennal blade is very transparent and in some specimens is hidden near the second antennal segment, so the separation of these 2 species is questionable; indeed differences between these two species are minimal also in pupal and adult stage [Lehmann, 1972]. The large or narrow median tooth of mentum is used to separate *E. minor*, *gracei*, *similis* from *E. brevicealcar*, *lobifera*; the presence of a hump in the median tooth separates *E. similis* from *E. minor*, but a hump was observed also in *E. bedmari* Vilchez-Quero, Laville, 1987 [Moubayed-Breil, Mary, 2019]; often the hump is not visible because the mentum is worn, so the separation of these species require further study. The most

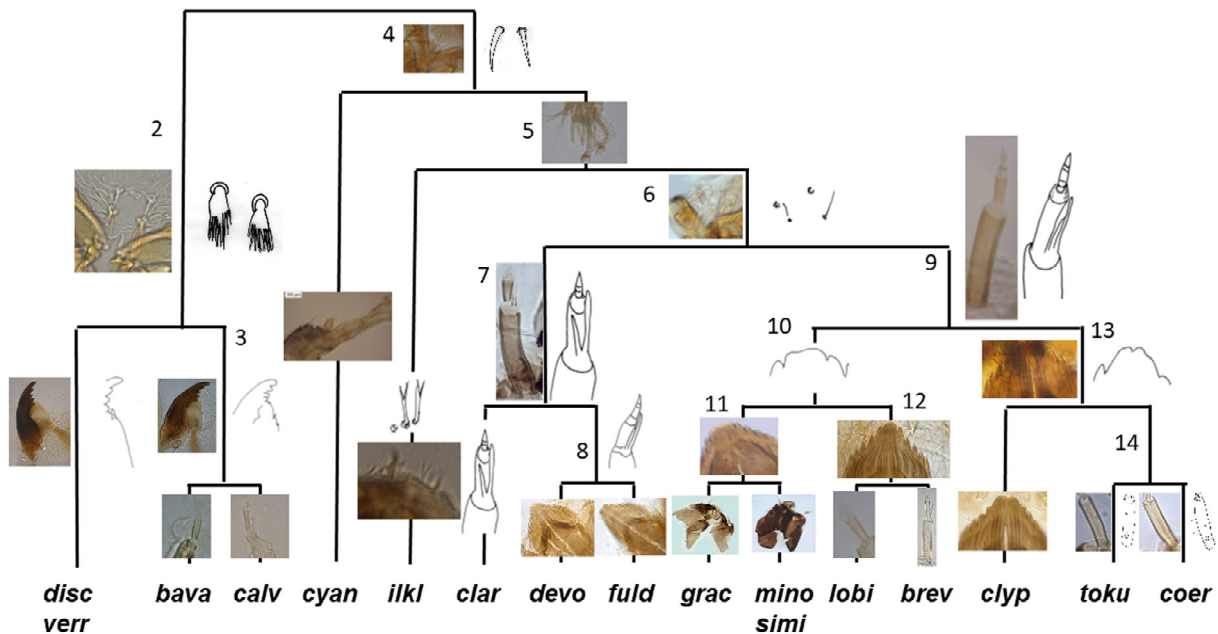


Fig. 107. Dendrogram summarizing the key to species. Abbreviations: *disc* — *T. discoloripes*, *verr* — *T. verralli*, *bava* — *T. bavarica*, *calv* — *T. calvescens*, *cyan* — *E. cyanea*, *ilkl* — *E. ilkleyensis*, *clar* — *E. claripennis*, *devo* — *E. devonica*, *fuld* — *E. fuldensis*, *grac* — *E. gracei*, *mino* — *E. minor*, *simi* — *E. similis*, *lobi* — *E. lobifera*, *brev* — *E. brevicarcar*, *clyp* — *E. clypeata*, *toku* — *Tokunagaia*, *coer* — *E. coerulescens*.

Рис. 107. Дендрограмма, обобщающая ключ к видам. Обозначения: *disc* — *T. discoloripes*, *verr* — *T. verralli*, *bava* — *T. bavarica*, *calv* — *T. calvescens*, *cyan* — *E. cyanea*, *ilkl* — *E. ilkleyensis*, *clar* — *E. claripennis*, *devo* — *E. devonica*, *fuld* — *E. fuldensis*, *grac* — *E. gracei*, *mino* — *E. minor*, *simi* — *E. similis*, *lobi* — *E. lobifera*, *brev* — *E. brevicarcar*, *clyp* — *E. clypeata*, *toku* — *Tokunagaia*, *coer* — *E. coerulescens*.

important characters useful in separating genera and species groups are the shape of setae anteriores S_1 and S_{III} . S_1 are branched in *Tvetenia*, simple in *Eukiefferiella* and in *Tokunagaia*, S_{III} are simple in all genera, except in *E. ilkleyensis* where S_{III} are bifid. *E. devonica* was included in *ilkleyensis* group on the basis of adult males and pupal exuviae [Lehmann, 1993], and in defining larval groups *E. devonica* and *E. ilkleyensis* were again joined [Epler, 2001; Andersen et al, 2013], but we were not able to see bifid S_{III} in *E. devonica*, S_{III} appear simple in the examined specimens; unfortunately the S_{III} is very small and not easily examined when the labrum is not mounted in a perfect dorsal-horizontal position. We also separated *E. devonica* and *E. ilkleyensis* on the basis of 4 or 5 antennal segments respectively, availability of more material will confirm or refute this conclusion.

The number of antennal segments is useful in separating species, but in some species the very reduced 3rd segment may be difficult to see. *E. coerulescens* was described with 4 antennal segments [Schmid, 1993; Epler, 2001], but in our samples a very short 3rd antennal segment is evident.

Zavřel [1939] describes «*atrofasciata*» Goetgh. and includes it in «*discoloripes* s. str.», describing the group with yellow head capsule, thorax greenish, abdomen bluish, labrum with a simple S_p , eyespot narrow, elongated [Zavřel, 1939: Fig. 2a], mandible mola with small

spines, anal tubuli cylindrical; what «*atrofasciata*» really is remains questionable, it is not cited in Ashe, O'Connor [2012]; simple S_1 conflicts with its inclusion in *Tvetenia*, as *discoloripes* s.str. should suggest.

The presence of 4 or 5 lateral teeth of mentum is also a valid character, but worn teeth sometime simulate a large median tooth, with the median tooth fused with the first lateral, and only 4 lateral teeth visible; it is here interesting to note that *E. quadridentata* Chernovskii, 1949, considered a *nomen dubium* [Ashe O'Connor, 2012], may be interpreted as a specimen with the median tooth fused with the first lateral. Similarly, *E. devonica* was described with 4 lateral teeth and a large median tooth [Schmid, 1993], in our samples indeed a separation between the median tooth and the first lateral is evident, but in other samples 4 lateral teeth seem associated to a large median tooth.

DNA barcode will aid to solve some morphological uncertainty; DNA sequences stored in GenBank® (<https://www.ncbi.nlm.nih.gov/nucleotide/>) could be analysed, but it is recommended that a study of DNA sequences be accompanied by morphological analysis, to check the correct association with morphological species. Rearing single larvae with association to the emerging pupal exuviae and adults, accompanied by the inspection of DNA sequences, even if laborious, is surely the definitive tool to solve these taxonomic problems [Gadawski et al., 2022; Rossaro et al., 2022].

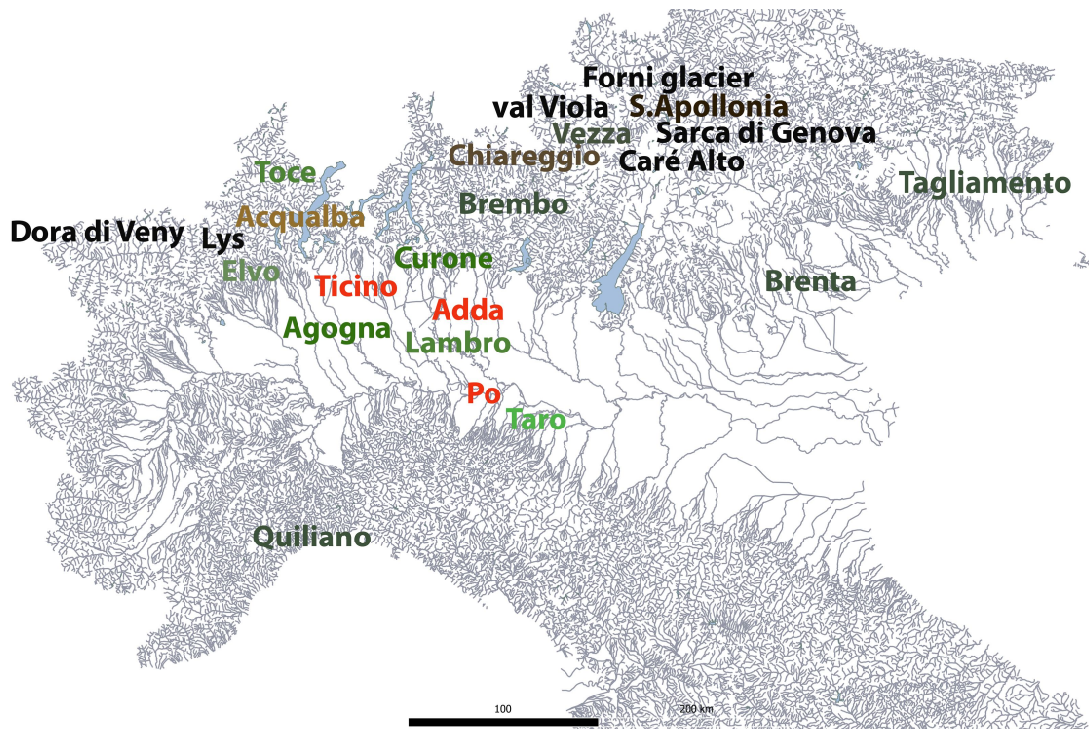


Fig. 108. Map of sampled localities in North Italy. Station names are coloured according to habitat type: black — kryal, brown — krenal, green — rithral, red — potamal.

Рис. 108. Карта местонахождений отобранного материала в Северной Италии. Названия станций окрашены в соответствии с типом местообитания: черный — криаль, коричневый — креналь, зеленый — ритраль, красный — потамаль.

ECOLOGY

The species treated in the present study are of high interest in freshwater ecology, because they colonize different habitats and have different tolerance to pollution. The species are restricted to running waters and cold springs, most species are found in different running water habitats from krenal to potamal, they seem to have wide habitat range and no species seems to be strictly restricted to only one habitat. Notes on the ecology of species are in many hydrobiological works [Thienemann, 1954; Lehmann, 1971], detailed information of single species are in Moller Pillot [2013]. No species seems restricted to glacial streams, but many species live in glacial streams, even if can be found in other habitats, some are strict cold stenothermal, others are more eurithermal. The following notes on the ecology of species [Moller Pillot, 2013] are based also on the author's direct experience. The localities where the samples were collected are in Tab. 1 and in Fig. 108 (North Italy) and Fig. 109 (Centre — South Italy).

T. bavarica is cold stenothermal, widespread but restricted to running waters in the Alps, it was found in small streams at high altitude as in the upper reach of Oglio river (Vezza), in rivers at lower altitude, but restricted in winter season as in Curone stream.

T. calvescens is widespread in running waters in different substrates, mosses, gravel, it is the most common species of the genus, it is also present in lowland rivers.



Fig. 109. Map of sampled localities in Centre South Italy. Station names are coloured according to habitat type.

Рис. 109. Карта местонахождений отобранного материала в центре Южной Италии. Названия станций окрашены в соответствии с типом местообитания.

T. discoloripes is rare in our samples, found in cold springs, in northern (Acqualba) and central Italy (Vetioio), but pupal exuviae were also collected in Po and Velino river.

T. verralli more frequent than *T. discoloripes* both in aquatic vegetation and in stones, both in large river (Toce, Brenta, Tagliamento) and small streams (Curone, Quiliano).

T. rectangularis characteristic of cold springs especially in glacial areas (Dora di Veny, Lys, Chiareggio).

T. tonollii is presumably a krenophilous species only found in a cold spring (Chiareggio).

E. ancyla: larvae unknown.

E. brevicealcar lives preferentially in cold streams, including glacial areas at high altitude (Dora di Veny), but is also found in Mediterranean streams (Sangro, Dirillo).

E. claripennis is very common everywhere, but with a preference for rhithral, it is tolerant to pollution.

E. clypeata found in stones substrates, frequent in epipotamon (Ticino, Adda).

E. coeruleascens in springs (S. Apollonia) or in small streams in the upper reaches (Vezza), it is probably indicator of waters of higher quality (PN Abruzzi)

E. cyanea is rheophilous, with morphological adaptation to fast flowing waters, considered indicator of high water quality (PN Abruzzi), not very frequent, present also in Mediterranean streams (Ofanto).

E. devonica is present in kryal, krenal, small streams (tributaries of Aviolo lake, Vezza) and more frequently in rhithral (Brembo, Lambro, Tagliamento).

E. dittmari larva unknown, pupal exuviae collected in different streams/rivers (Toce, Ticino, Brembo, Lambro, Adda, Po, Tagliamento, Brenta, Taro, Mignone, Aso, Sangro, rio Mannu)

E. fittkai restricted to mountain areas, larva not separable from *E. minor*.

E. fuldensis characteristic of glacial streams, not found in lower reaches (Dora di Veny, Viola, Forni, Caré Alto).

E. gracei seems to prefer reaches of lowland streams or rivers (Ticino, Lambro, Po, Taro)

E. ilkleyensis is widespread, frequent in rhithron, also in presence of aquatic vegetation (Agogna, Oglio)

E. lobifera is present also in cold streams, but prefers riffles zones in large lowland rivers and Mediterranean springs or streams. It seems more temperature tolerant (Ticino, Lambro, Po, Taro, Tagliamento, Quiliano, Potenza, Aso, PN Abruzzi)

E. minor is widespread in mountain areas in stones and mosses, it is considered a tolerant species, among cold-stenothermal species, present in almost all stations especially in winter

E. pseudomontana larvae are not known.

E. similis possibly more temperature tolerant respect to *E. minor*, its presence in Alps (Chiareggio) should be confirmed.

E. tirolensis is abundant in cold fast flowing streams near glaciers, but at some distance from the glacier

mouth (Dora di Veny), its similarity with *E. brevicealcar* larva makes the map of distribution difficult to prepare.

It can be concluded that at present the information about the ecology of species is rather generic and require the collection of more data, where correct species identification will be associated to environmental information.

Acknowledgements

This work has been partially supported by a grant of the CEE project: DG X_{II}-D; ENV4-CT95-0164 Arctic and Alpine Stream Ecosystem Research (A.A.S.E. R.) and Italian Ministero dell'Università (M.U.R.S.T.).

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Поступила в редакцию 27.5.2022