

**Biology and morphology of immature stages
of *Coniocleonus nigrosuturatus*
(Coleoptera: Curculionidae: Lixinae)**

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Abstract. Mature larvae and pupae of *Coniocleonus (Plagiographus) nigrosuturatus* (Goeze, 1777) (Curculionidae: Lixinae: Cleonini) are described and compared with three other cleonine taxa with known larvae. The biology of the species was studied in Romania, Hungary and Slovakia. Common Stork's-bill (*Erodium cicutarium*) (Geraniaceae) is identified as a host plant of both larvae and adults of this weevil. The weevil is very likely monophagous, and previous records of thyme (*Thymus* sp., Lamiaceae) as the host plant hence appear incorrect. *Coniocleonus nigrosuturatus* prefers dry, sunny places in grassland habitats, with sparse vegetation, bare ground and patchily growing host plants. Overwintering beetles emerge in early spring (March), feed and mate on the host plants. The highest activity of adults was observed from mid-April to mid-May. Larvae live in the soil in a vertical tunnel near the host plant and feed externally on the base of the stem. Usually, a single larva was observed at an attacked plant, however, sometimes up to 3 larvae were found. At the beginning of July, the larvae pupate in a closed earthen cell. A reared adult emerged in the laboratory at the end of July. We suppose that part of the population overwinters in pupation cells, although some adults leave the cells at the end of summer. They then most likely spend some time feeding on the host plants and look for suitable shelters in which to overwinter later.

Key words. Coleoptera, Curculionidae, Lixinae, *Coniocleonus nigrosuturatus*, mature larva, pupa, morphology, host plant, *Erodium cicutarium*, Geraniaceae, larval development, life history, central Europe, Palearctic Region

Introduction

Weevils of the subfamily Lixinae (Coleoptera: Curculionidae) are medium to large-sized beetles distributed mainly in Palaearctic and Afrotropic Regions. The subfamily includes approximately 1,200 species (TER-MINASIAN 1967). Although generally accepted as a separate subfamily (e.g., LÖBL & SMETANA 2013), the group is occasionally treated as a part of the Molytinae (OBERPRIELER et al. 2007) or Curculioninae (LAWRENCE & NEWTON 1995). Lixinae is considered a rather derived weevil group standing close to the Hyperinae Marseul, 1863 and Mesoptiliinae Lacordaire, 1863, and is divided into two tribes: Cleonini Schönherr, 1826 and Lixini Schönherr, 1823 (LÖBL & SMETANA 2013). Although a preliminary phylogenetic analysis is only available for Mediterranean and Afrotropical taxa, a general analysis of the whole tribe is now in progress (MEREGALLI & SILVESTRO 2008, unpubl. data).

Until recently, the immature stages of the Lixinae were largely unknown and unstudied (ANDERSON 1987). The situation has slightly changed within the last 10 years, but this mainly concerns the tribe Lixini, for which several studies describing the morphology of immature stages have been published (NIKULINA 2001, 2007; NIKULINA & GÜLTEKIN 2011; GOSIK & SKUHROVEC 2011; GOSIK & WANAT 2014). The knowledge of the immature stages of the Cleonini remains very limited, as immature stages of only three species have been described by SCHERF (1964).

The genus *Conioleonus* Motschulsky, 1860, belongs to the tribe Cleonini. It comprises 27 known species classified in five subgenera (MEREGALLI & FREMUTH 2013). All species are known from the Palaearctic Region, two of them also reach the Nearctic Region. Seventeen species are known from Europe, occurring mainly in the Mediterranean area (MEREGALLI & FREMUTH 2013).

Data on the biology of the species of *Conioleonus* are very scarce. They prefer warm and dry habitats in steppe or semidesert regions (TER-MINASIAN 1967, 1988; DIECKMANN 1983; KOCH 1992). Host plants are unknown for the majority of species. For a few species, only data on refuge plants are available, which are used as shelter for adults and in some cases may be food sources. *Conioleonus cicatricosus* (Hoppe, 1795) and *C. nebulosus* (Linnaeus, 1758) were collected under stands of *Calluna vulgaris* or *Erica* spp. (Ericaceae) (KOCH 1992). *Conioleonus hollbergi* (Fåhraeus, 1842) and *C. turbatus* (Fåhraeus, 1842), both of which occasionally feed on some woody plants (e.g., *Pinus sylvestris*, *Quercus* spp.), are supposed to develop on *Rumex acetosella* (Polygonaceae) (DIECKMANN 1983, BAYER & WINKELMANN 2005, RHEINHEIMER & HASSLER 2010).

Larval development has been observed only for two species: *Conioleonus astragali* Ter-Minasian & Korotyaev, 1977, which developed on the roots of *Astragalus* sp. and *Oxytropis* sp. (both Fabaceae) with larvae and pupae hidden in chambers made of plant debris and excrements (TER-MINASIAN 1988). Ectophagous larvae are also known for *Conioleonus excoriatus* (Gyllenhal, 1834), which is associated with *Emex spinosa* (Polygonaceae) (JULIEN 1980, SCOTT & YEOH 1996). Nevertheless, descriptions of the immature stages of these weevils have not been published.

One of the species with very poorly known biology is *Conioleonus nigrosuturatus*. The species belongs to the subgenus *Plagiographus* Chevrolat, 1869 (MEREGALLI & FREMUTH 2013)

which is regarded as a separate genus by some authors (e.g., ARZANOV 2006). The species' range includes North Africa, a large part of Asia to western Siberia and most of Europe except the northern part and British Isles (MEREGALLI & FREMUTH 2013). In central Europe, the species is regarded as a xerothermic stenotopic species inhabiting sandy habitats, heathlands, dry grasslands and pastures (KOCH 1992). In several central European countries, *C. nigrosuturatus* is an endangered species (e.g., GEISER 1998, BENEDIKT & STREJČEK 2005, STEJSKAL & TRNKA 2013), and this species is only known from historical records in many regions (LOHSE 1983). Thus far, only thyme (*Thymus* sp.) was reported as host plant of adults, without mentioning other details of the biology of the species, likely first reported by KLEINE (1910). Later authors either only repeated his data (FLEISCHER 1927–1930, FREMUTH 1982, TER-MINASIAN 1988, ARZANOV 2006) or regarded *C. nigrosuturatus* as a species with an unknown host plant (e.g., DIECKMANN 1983, LOHSE 1983). THÉRON (1976) listed the findings of *C. nigrosuturatus* on *Limonium* sp. (Plumbaginaceae) in France. The immature stages of *C. nigrosuturatus* have never been observed. In this paper, we describe immature stages of the species and provide details on its life history based on our observations obtained during field work in Romania, Hungary and Slovakia.

Material and methods

The material used to describe immature stages was collected, and field observations were conducted, in the following localities:

ROMANIA: CARAŞ-SEVERIN COUNTY: Sfânta Elena env., 44°40'42"N, 21°43'14"E. Visit dates: 26.iv.–1.v.2010, 28.iv.–1.v.2011, 24.–28.vii.2011, 4–9.vi.2012, 18.–24.v.2013, 4.vii.2013. Habitats: pastures (cow, sheep, goats), road margins and dry grasslands. Bedrock: limestone. Altitude: 400 m a. s. l. (see Figs 5D–E).

HUNGARY: BÁCS-KISKUN COUNTY: Petőfiszállás env., 46°36'33"N, 19°50'19"E. Visit dates: 18.v.2013. Habitat: closely cut lawn around a petrol station. Bedrock: sand. Altitude: 90 m a. s. l.

SLOVAKIA: TRNAVA REGION: Dolné Zelenice env., 48°22'6"N, 17°45'2"E. Visit dates: 9.v.2013 and 16.vi.2013. Habitats: dry grasslands and road margins. Bedrock: alluvial sediments (sandy gravel). Altitude: 130 m a. s. l. **NITRA REGION:** Koliňany env., 48°20'35"N 18°11'15"E. Visit dates: 26.v.2012. Habitats: military area with disturbed grasslands and free sand. Bedrock: tertiary deposits (loess), sand. Altitude: 300 m a. s. l. (see Fig. 5F).

Rearing and life cycle observations were conducted during the vegetation seasons of 2012–2013. Laboratory observations were conducted in Sfânta Elena, Romania, in Znojmo, Czech Republic (48°51'31"N, 16°2'40"E) and in Olomouc, Czech Republic (49°35'36"N, 17°15'3"E) using larvae from Sfânta Elena, Romania, and Dolné Zelenice, Slovakia. Host plant associations were studied by direct observation in the field and by testing selected plants in the laboratory.

Larvae were collected by R. Stejskal and F. Trnka in the soil within the vicinity of the host plant (*Erodium cicutarium*) and maintained in the laboratory using the following two rearing techniques: (1) Some larvae were excavated using a garden trowel along with the complete host plant including its root ball and soil. The plant was then cultivated in a common flowerpot. (2) Some larvae were first removed from their soil tunnels and then placed in a prepared flowerpot with the host plant. In these cases, the host plants were cultivated in soil sifted in advance to remove unwanted materials and organisms (e.g., carnivorous beetle larvae).

The flowerpots containing the host plants and larvae were placed in a sunny place at room temperature and moistened approximately once every 3–5 days. Moulding or dead plants were replaced with new plants.

Part of the material of the larvae and pupae was preserved in Pampel fixation liquid (4 parts glacial acetic acid, 6 parts 4% formaldehyde, 15 parts 95% ethylalcohol and 30 parts distilled water) and used for the morphological descriptions. These specimens are now deposited in the Group Function of invertebrate and plant biodiversity in agro-ecosystems of the Crop Research Institute (Prague, Czech Republic). Plants were identified by the collectors and the identification was reviewed by a botanist.

Slides were prepared as follows (for details see MAY 1994): a larva was decapitated, and its head was rinsed in distilled water and cleaned in a 10% solution of potassium hydroxide (KOH). Then, the mouthparts were separated from the head capsule. All body parts were then mounted on temporary microscope slides in 10% glycerin.

The specimens were examined and measurements were made using a light microscope with ocular scale (Olympus SZ60 and SZX16). The following characteristics were measured for each larva (head width; length of the body [larvae fixed in C-shape were measured by segment], width of the body in the widest place [abdominal segments]) and pupa (length and width). The thorax and abdomen are not sclerotised and their proportions may be affected by the fixation process, thus measurements of these parts are only given for comparison purposes.

Drawings were made using a drawing tube attached to the light microscope and processed by computer (GIMP 2). The thoracic spiracle is placed on the boundary of the prothorax and mesothorax, as shown in the drawings (see Fig. 2B); however, it is in fact of mesothoracic origin (MARVALDI et al. 2002, MARVALDI 2003). Drawings of the thoracic and abdominal spiracles are schematic (see Figs. 2B–D). Terms and abbreviations regarding the setae of the mature larva and pupa follow SCHERF (1964), MAY (1977, 1994) and MARVALDI (1998).

Morphology of mature larva and pupa

Coniocleonus nigrosuturatus (Goeze, 1777)

Material examined. ROMANIA: Sfânta Elena: 24.v.2013 (1 larva), 4.vii.2013 (1 larva, 1 pupa) (all F. Trnka leg.), 7.vi.2012 (1 larva), 10.vii.2012 (1 pupa reared, partly damaged), 19.v.2013 (2 larvae), 7.vii.2013 (1 pupa reared from larva from v.2013 (partly dried-up)) (all R. Stejskal leg.). SLOVAKIA: Dolné Zelenice, 16.vi.2013 (2 larvae), R. Stejskal leg.

Description of mature larva. *Colouration.* Head light brown or brown. All thoracic and abdominal segments white (Fig. 4A). Cuticle finely spiculate.

Head capsule and mouth parts. Head width: 1.5–1.7 mm (mean 1.6 mm), suboval, flattened laterally, endocarinal line absent. Frontal sutures on head distinct, Y-shaped, extended to stemmata. Single stemma (st), in the form of a dark pigmented spot, located on each side anterolaterally. *Des1* near to frontal suture, *des2* and *des3* located in the central part of epicranium, *des4* located anteriorly near to frontal suture, *des5* located anterolaterally; *des1–2* and *des4–5* long, equal in length, *des3* distinctly shorter (Fig. 1A). *Fs1* and *fs2* placed medially, *fs3* located anteriomedially, *fs4* located anteriolaterally, and *fs5* located laterally, close to the epistoma; all setae relatively long, *fs3* and *fs4* shorter than the remaining three setae (Fig. 1A). *Les1–2* as long as *des1*; and *ves1–2* as long as *des3*. Epicranial area without pores. Antennae

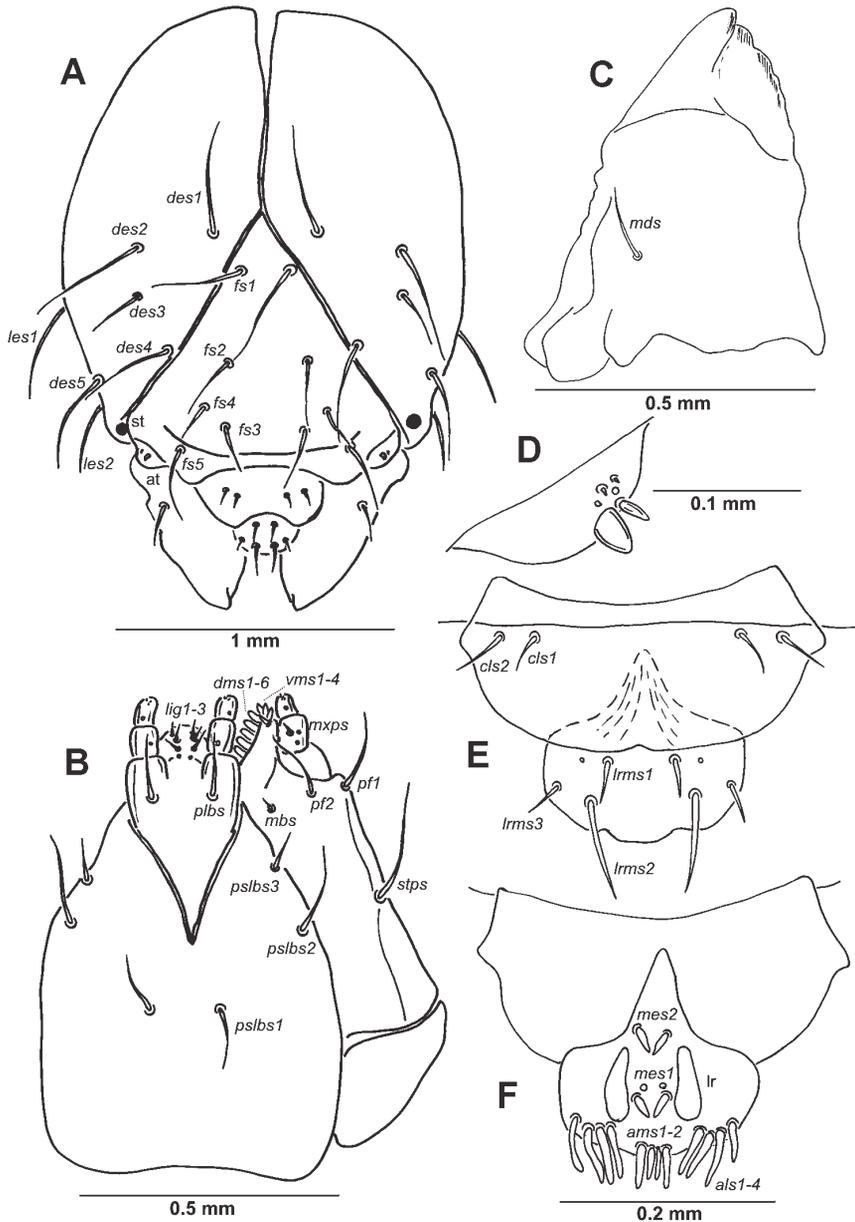


Fig. 1. *Conioleoneus nigrosuturatus* (Goeze, 1777), mature larva head, antenna and mouth parts: A – dorsal view (*des* – dorsal epicranial seta(e), *les* – lateral epicranial s., *st* – stemmata, *at* – antenna); B – right maxilla and labium (both ventral view) (*dms* – dorsal malae s., *vms* – ventral malae s., *mxps* – maxillary palps s., *mbs* – basiventral s., *pf*s – palpiferal s., *stps* – stipal s., *plbs* – prelabial s., *pslbs* – postlabial s., *lig*s – ligular s.); C – right mandible (*mds* – mandible dorsal s.); D – antenna; E – labrum and clypeus (*cls* – clypeal s., *lrms* – labral s.); F – epipharynx (*ams* – anteromedial s., *als* – anteriolateral s., *mes* – median s., *lr* – labral rods).

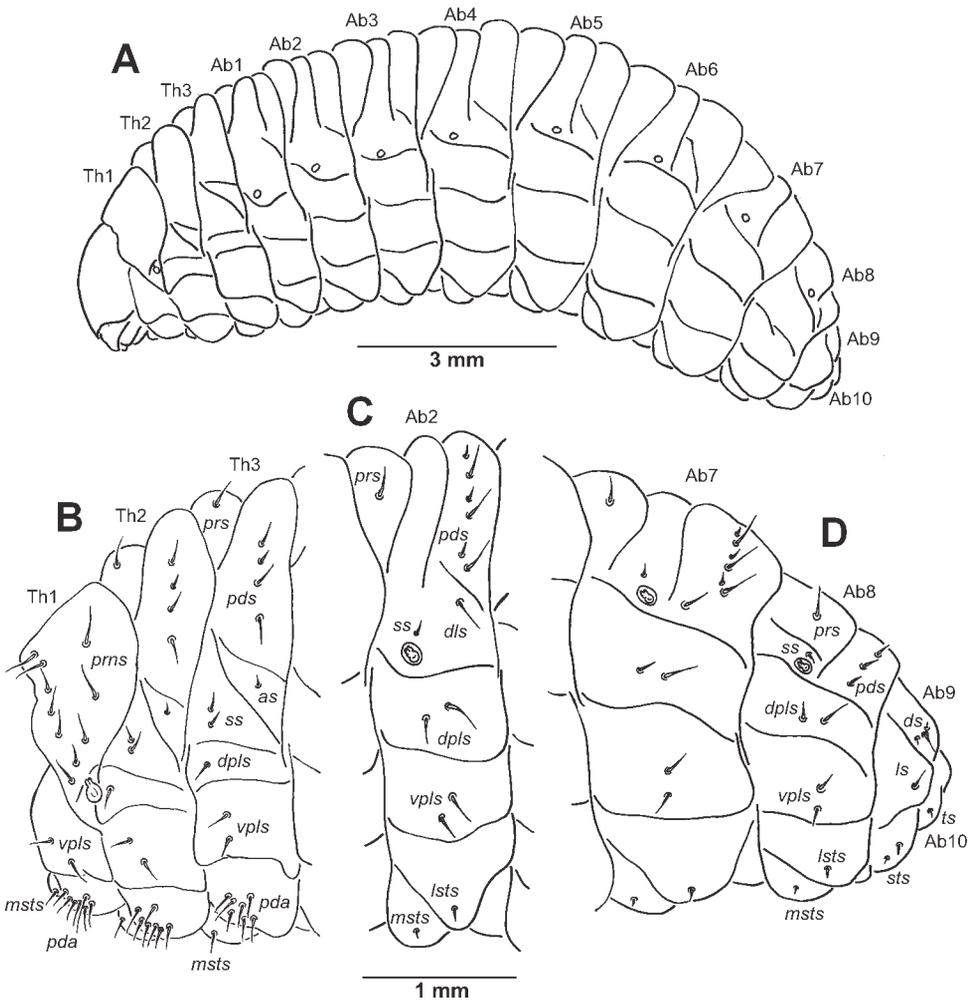


Fig. 2. *Conioleonus nigrosuturatus* (Goeze, 1777), mature larva habitus: A – shape of the body (lateral view), B – lateral view of thoracic segments, C – lateral view of abdominal segment II, D – lateral view of abdominal segments VII–X (*prns* – pronotal seta(e), *prs* – prodorsal s., *pds* – postdorsal s., *as* – alar s., *ss* – spiracular s., *dpls* – dorsopleural s., *vpls* – ventropleural s., *pda* – pedal s., *dls* – dorsolateral s., *lst*s – laterosternal s., *msts* – mesosternal s., *ds* – dorsal s., *ls* – lateral s., *sts* – sternal s., *ts* – terminal s.; Th1–3, Ab1–10 – number of thoracic or abdominal segment).

located at the end of the frontal suture on each side, membranous and slightly convex basal article bearing conical triangular sensorium, relatively long; basal membranous article with 3 sensillae different in both shape and length (Fig. 1D).

Labrum (Fig. 1E) approximately 2 times as wide as long, with 3 pairs of hairform *lrms*, of different length; *lrms3* and *lrms1* distinctly shorter than *lrms2*; *lrms1* placed close to

the margin with clypeus, *lrms2* located anteriomedially and *lrms3* located anteriolaterally; anterior margin double sinuate. Clypeus (Fig. 1E) approx. 2.5 times as wide as long with 2 relatively short *cls*, unequal in length, localized posteriolaterally; anterior margin rounded to the inside; median part covered by thorn-shaped cuticular processes. Epipharynx (Fig. 1F) with 4 pairs of blunt, finger-like *als*, of almost equal length; 2 pairs of *ams*, *ams1* distinctly shorter than *ams2*; 2 pairs of short, blunt *mes*; labral rods (lr) elongated, converging anteriorly. Mandibles (Fig. 1C) relatively broad, bifid, tooth of unequal height; slightly truncate; *mds* relatively long, hairform. Maxilla (Fig. 1B) stipes with 1 *stps*, 2 *pfs* and 1 *mbs*, *stps* and *pfs1-2* long, equal in length, *mbs* very short; mala with 6 bacilliform *dms* of different length; 4 *vms* short, almost equal in length; *vms* distinctly shorter than *dms*. Maxillary palpi with two palpomeres; basal palpomere with 1 *mxps* and two pores; length ratio of basal and distal palpomeres: 1:0.7; distal palpomere with one pore and a group of conical, cuticular apical processes. Praelabium (Fig. 1B) heart-shaped and distinctly elongated, with 1 pair of *plbs*; ligula with sinuate margin and 3 pairs of hairform micro *ligs*, equal in length; premental sclerite well visible. Labial palpi with two palpomeres; length ratio of basal and distal palpomeres: 1:0.6; distal palpomere with one pore and short, cuticular apical processes; basal palpomere with 1 dorsal pore. Postlabium (Fig. 1B) with 3 *pslbs*, *pslbs1* located anteriorly, remaining two pairs laterally; different in length, *pslbs3* distinctly shorter than *pslbs1* and *pslbs2*.

Thorax and abdomen. Body length: 11.0–13.5 mm (mean 12.0 mm) stocky, slightly curved, rounded in cross section (Fig. 2A). The widest place in the body (abdominal segments V–VI) measuring up to 4.5 mm. Prothorax distinctly smaller than meso- and metathorax. Metathorax and abdominal segments I–III of almost equal length, next abdominal segments increasing gradually to abdominal segments V–VI (the largest) and then decreasing to the terminal parts of the body. Abdominal segment X reduced to four anal lobes of unequal size, the dorsal being distinctly the largest, the lateral pair equal in size, and the ventral lobe very small. Anus located terminally. Spiracles (9 pairs) bicameral, the first placed between the pro- and mesothorax (see Material and methods), the abdominal spiracles located laterally, close to the anterior margin of abdominal segments I–VIII.

Chaetotaxy of mature larva. Slightly reduced, but in some parts with more than the most frequent number of setae in weevils (e.g., meso- and metathorax with 2(3) *pds*, and abdominal segments I–VII with 5 *pds*; the state in *C. nigrosuturatus* see below). Setae thin, long, light yellow or orange. **Thorax.** Prothorax (Fig. 2B) with 10 *prns* unequal in length, 2 *vpls* and 1 *msts*. Mesothorax (Fig. 2B) with 1 *prs*, 4 *pds* (*pds2* distinctly shorter than the three remaining setae), 1 very short *as*, 2 short *ss*, 1 *dpls*, 2 *vpls* of different length and 1 short *msts*. Chaetotaxy of metathorax (Fig. 2B) identical to mesothorax. Each pedal area of thoracic segments well separated, with 6–8 *pda* unequal in length. **Abdomen.** Abdominal segments I–VII (Figs 2C–D) with 1 *prs*, 6 *pds* (*pds1*, *pds3* and *pds5* very short), 1 *dls*, 1 very short *ss*, 2 *dpls* of different length, 2 *vpls*, 1 very short *lsts* and 1 micro *msts*. Abdominal segment VIII (Fig. 2D) with 1 *prs*, 3 very short *pds*, 1 very short *ss*, 2 *dpls* of different length, 2 short *vpls*, 1 very short *lsts* and 1 micro *msts*. Abdominal segment IX (Fig. 2D) with 3 *ds* (*ds2* relatively long, *ds1* and *ds3* microsetae); 1 short *ls* and 2 *sts* of different length (*sts1* very short, *sts2* microsetae). Each anal lobe on abdominal segment X (Fig. 2D) with 1 microseta (*ts*).

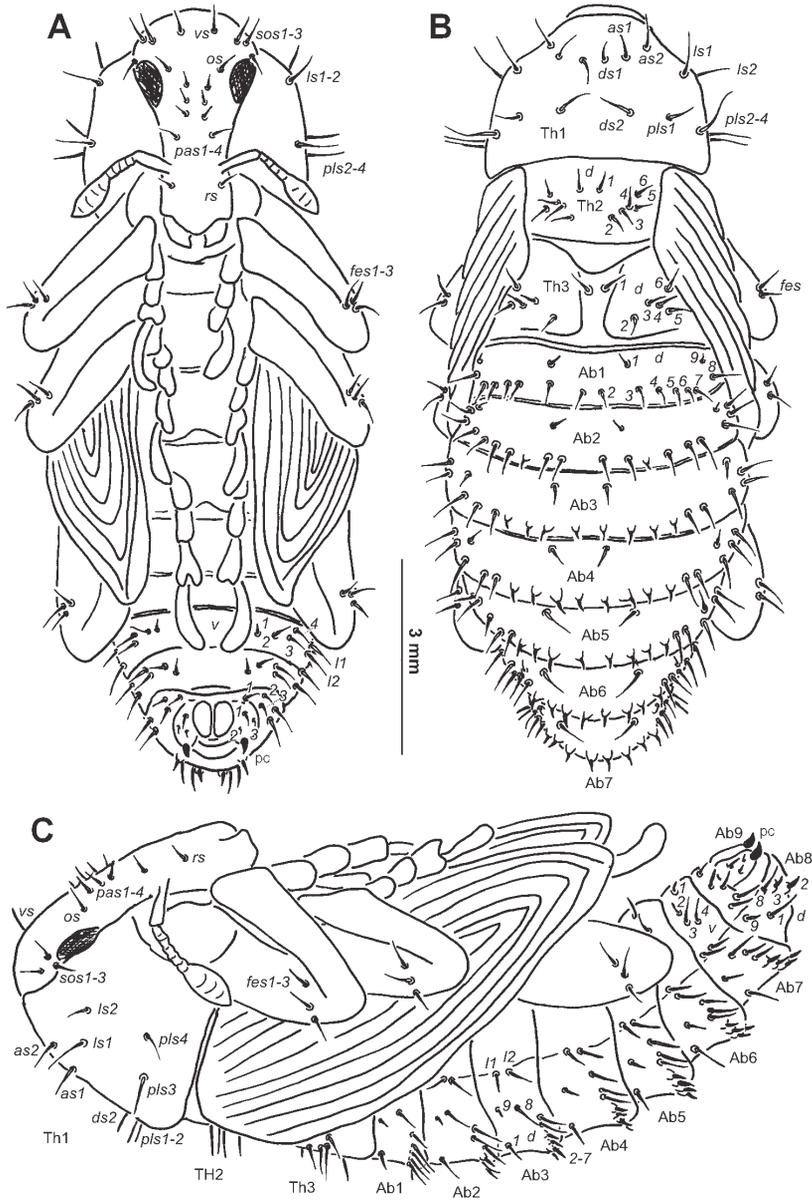


Fig. 3. *Coniocleonus nigrosuturatus* (Goeze, 1777), pupa habitus: A – ventral view, B – dorsal view, C – lateral view (vs – vertical seta(e), sos – superorbital s., os – orbital s., pas – postantennal s., rs – rostral s., as – apical s., ls – lateral s., ds – discal s., pls – posterolateral s., fes – femoral s., d – dorsal s., l – lateral s., v – ventral s.; Th1-3, Ab1-10 – number of thotacic and abdominal segment; pc – pseudocerci).

Description of pupa. Morphology (Figs 3A–C, 4C). Body length: 11.0–11.6 mm, at the widest region: 4.5–5.3 mm. The widest place in the body is commonly between the apex of the meso- or metafemora. Body stocky, elongated, white or yellowish. Cuticle smooth. Rostrum relatively short, approximately 1.8 times as long as wide, extended beyond procoxae. Antennae relatively long and stout. Pronotum almost 1.7 times as wide as long. Mesonotum and metanotum of almost equal length. Abdominal segments I–V of almost equal length; abdominal segment VI semicircular, next abdominal segments diminish gradually to the end of the body. Abdominal segments VII–IX distinctly smaller than other abdominal segments. Gonotheca (abdominal segment IX) of all three specimens divided. Sexual dimorphism in weevils is visible mainly in the length of rostrum and in the structure of abdominal segment IX: gonotheca of ♂ undivided, of ♀ divided (GOSIK & SPRICK 2012a,b, 2013; GOSIK & WANAT 2014).

Chaetotaxy (Figs 3A–C). Setae relatively long, unequal in length, light yellow or orange, some setae on abdominal segments II–VIII distinctly get stronger and located on protuberances. Setae well visible. Head capsule includes 1 *vs*, 3 *sos*, 1 *os* and 4 *pas*. Rostrum with 1 *rs*, located on the anterior margin. Setae on head capsule and rostrum straight, *rs* and *pas* 1–3 distinctly shorter than the remaining setae on head, thoracic and abdominal segments.

Pronotum with 2 *as*, 2 *ds*, 2 *ls* and 4 *pls*. Dorsal parts of mesothorax with 1 pair of setae located posteromedially and 5 pairs located along its anterior margin. Chaetotaxy of metathorax identical to mesothorax. Each apex of femora with groups of 3 *fes*. Dorsal parts of abdominal segments I–VIII each with 2 pairs of setae located posteriorly (*d1*, *d9*) and 7 pairs (*d2*–8) located along their anterior margins. Seta *d4* (on abdominal segment II) and setae *d2*–5 (on abdominal segments III–VII) short, thorn-like, located on protuberances. Remaining setae long, hair-like. Abdominal segments I–VII with groups of 2 lateral setae and 4 pairs of ventral setae. Dorsal part of abdominal segment VIII with 2 pairs of setae located posteriorly (*d1*, *d9*) and 3 pairs (*d2*, *d3* and *d8*) located along its anterior margin; *d2*–3 thorn-like, located on protuberances; remaining setae elongated. Abdominal segment VIII with groups of 2 lateral setae and 3 pairs of short ventral setae. Abdominal segment IX with 2 pairs of ventral microsetae and 1 pair of short, thin setae. Pseudocerci short, triangular.

Comparison with larvae of other Cleonini. Larvae of three cleonine taxa have been described so far (SCHERF 1964): *Cleonis pigra* (Scopoli, 1763), *Cyphocleonus dealbatus* (Gmelin, 1790) (as *Cyphocleonus tigrinus* (Panzer, 1789)), and ?*Pachycerus segnis* (Germar, 1824) (as *Pachycerus scabrosus* Brullé, 1832, but identification is probably incorrect, see below). The comparison of the larva of *Coniocleonus nigrosuturatus* with those described by SCHERF (1964) is somewhat problematic due to the use of differing terminology for morphology and chaetotaxy and/or an absence of good quality drawings. Despite these problems, we were able to compare the morphology of all four taxa (Table 1). However, the identity of the larva described as *Pachycerus segnis* by SCHERF (1964) is unclear and it likely represents a genus other than *Pachycerus*. According to extensive surveys by BRUN et al. (1993) conducted in France, Greece and Turkey, larvae of *P. segnis* were always found within earthen cells attached to the taproot, but not in gall-like swellings as reported by SCHERF (1964). Descriptions by SCHERF (1964) perfectly fit the larval development of some species of the cleonine genus

Table 1. Differential diagnosis of mature larvae and pupae of 4 species from the tribe Cleonini.

	<i>Coniocleonus nigrosuturatus</i>	<i>Cleonis nigra</i>	<i>Cyphocleonus dealbatus</i>	' <i>Pachycerus scabrosus</i> ' sensu SCHERF (1964)*
Larva				
Endocarina	absent	present	absent	absent
Number of stemmata	1	2	1	1
Number of <i>des</i>	5	4	5	5
Number of <i>fs</i>	5	4	5	5
Number of <i>les</i>	2	not presented	3	3
Number of <i>ves</i>	2	not presented	not presented	not presented
Number of <i>cls</i>	2	2	2	3
Position of <i>lrms1–3</i>	in a triangle	in a triangle	in a triangle	in a line
Number of <i>als</i>	4	4	6	3
Number of <i>mds</i>	1	2	2	2
Number of <i>ligns</i>	3	3	2	3
Number of <i>mbs</i>	1	0	1	1
Number of <i>pds</i> on abdominal segments I–VII	6	7	not presented	5
Pupa				
Number of setae on mesonotum	6	4	unknown	4
Number of <i>fes</i>	3	2	unknown	not presented
Number of setae on dorsum of abdominal segments I–VII	9	7	unknown	5–7

* This larva was probably misidentified by SCHERF (1964) and more likely belongs to the genus *Rhabdorrhynchus* (see the text for details).

Rhabdorrhynchus, which takes place in a root gall-like swelling (DIECKMANN 1983, STEJSKAL & TRNKA, unpubl. data).

MAY (1993) considered the increased number of *pds* on meso- and metathorax and abdominal segments I–VII and the increased number of epipharyngeal lining setae (*als*) (i.e. higher than the most frequent number of setae in weevils [for details see Chaetotaxy of mature larva of *C. nigrosuturatus*]) as diagnostic for the mature larva of the subfamily Lixinae. Descriptions of mature larvae from the tribe Lixini (*Larinus* species: GOSIK & SKUHROVEC 2011; *Lixus* species: SCHERF 1964, MAY 1994, NIKULINA 2001, 2007, NIKULINA & GÜLTEKIN 2011, GOSIK & WANAT 2014; *Rhinocyllus conicus*: MAY 1994) fit this diagnosis, as do all known species from the tribe Cleonini (see Table 1). The comparison of both tribes is recently not possible because our knowledge of immature stages in Cleonini is very scarce.

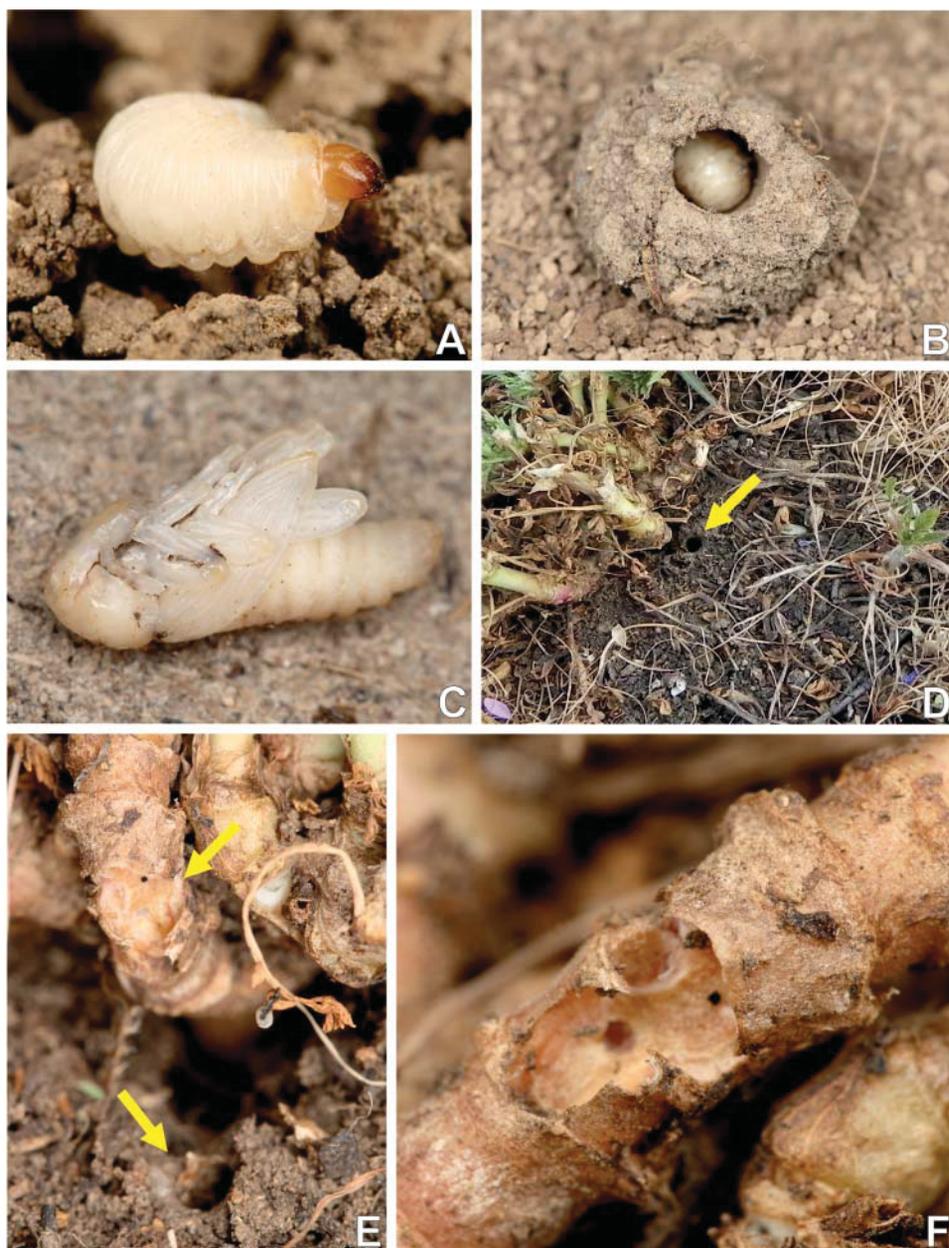


Fig. 4. Immature stages and feeding marks of *Conioleonus nigrosuturatus* (Goeze, 1777). A – mature larva; B – earthen cell with mature larva; C – pupa (removed from the earthen cell); D – opening of larval tunnel in the soil; E – opening of the larval tunnel and feeding marks; F – detail of larval feeding marks.



Fig. 5. Adult, host plant and habitats of *Coniocleonus nigrosuturatus* (Goeze, 1777). A – adult beetle; B – host plant *Erodium cicutarium* (L.) L'Hér.; C – an excavated host plant with larval feeding marks; D–E: habitats in Romania (Sfânta Elena); F – habitat in Slovakia (Koliňany).

Biology of *Coniocleonus nigrosuturatus*

Habitat. Based on our observations, *C. nigrosuturatus* prefers dry, sunny places in grassland habitats with sparse vegetation, bare ground and patchily growing host plants. The weevil often inhabits road margins, pastures and dykes along rivers and requires a special habitat structure originating from a proper disturbance regime. Heterogeneous patches of properly

disturbed sites occur for instance due to vehicle traffic (Figs 5E–F) or the grazing of domestic animals (Fig. 5D). The adults avoid places with compact and overgrown vegetation, where we suppose the soil is not sufficiently warm for larval development. Regarding bedrock, the weevil is quite flexible, occurring on a wide range of substrata, such as limestone and various sediments (sand, sandy gravel and loess).

Adult behaviour. The weevils are diurnal and can be very agile during sunny weather, however, during cold weather, they remain motionless. We usually observed adults (Fig. 5A) on the ground or on host plants, where they spend most of their time sitting on the top or underside of the host plant rosette (Fig. 5B). An attacked plant was usually occupied by 1–4 beetles. The adults were not observed in flight but are fully winged (ANGELOV 1963). Specimens examined by us for this character (in total 3 specimens from Hungary, Romania and Slovakia) were all macropterous. Adults can be observed from March onwards, with peak activity from mid-April to mid-May. Single adults can be observed at the end of summer or during autumn (August/October). We also observed beetles mating on the host plants. A mating male tightly embraces the female and repeatedly stimulates her with his mid-legs by knocking very rapidly on her metathorax.

Host plant. Both adults and larvae were observed feeding exclusively on Common Stork's-bill (*Erodium cicutarium* (L.) L'Hér.) (Geraniaceae, Figs 5B–C). Adults feed on leaves, whereas larval development occurs underground on the stem base. We also tested feeding on thyme (*Thymus* sp.), which was mentioned as a host plant by several authors (KLEINE 1910, FLEISCHER 1927–1930, FREMUTH 1982, TER-MINASIAN 1988, ARZANOV 2006). The adults completely avoided feeding on thyme, both in the laboratory and in the field, and we thus consider thyme solely as an occasional refuge plant. Other plant species were not tested. Based on our observations and experiments, *Coniocleonus nigrosuturatus* hence appears to be most likely monophagous, feeding exclusively on *Erodium cicutarium*.

Life cycle. *Coniocleonus nigrosuturatus* is a univoltine species. We did not find the site of egg deposition, but we suppose that females lay eggs in the soil near the root neck. Mature larvae live ectophagously near the host plant root and construct a vertical tunnel in the soil up to several centimeters long. The larval tunnel, which has a circular to oval shape in cross section with diameter of 4–5 mm, opens onto the soil surface, where the larvae feed externally on the stem base (Figs. 14D–F). We have not observed younger instar larvae, but we suppose that they most likely also live ectophagously, like the mature larvae. Their development inside roots/stems could be excluded because we did not find any elongated tunnels inside the plants despite exhaustive searching. No feeding marks were found on roots. At an attacked plant, a single larva was usually observed; however, sometimes up to 3 larvae were found. The host plant infestation rate may reach 50–75% according to our estimation. We observed higher larval feeding activity during the morning hours (before 10 a.m.), in the evening (after 7 p.m.) and in cloudy weather. During midday under warm and sunny conditions, larvae hid deeper in the tunnels. From the beginning of July, larvae pupate in oval, closed earthen cells, 25–30 mm long and 10–15 mm wide (Fig. 4B), located several centimeters below the soil surface under the host plant. The inner diameter of the earthen cell is approximately 5 mm, and its inner length is approximately 20 mm. The only reared adult emerged in the laboratory at the end of July. Further details of the life cycle were not observed. However, we suppose

that a part of the population overwinters in the pupation cells, and some adults leave the cells at the end of summer, resulting in the records of adults in August to October (DIECKMANN 1983, Stejskal & Trnka, unpubl.). Most likely, the weevils then spend some time feeding on the host plants and look for suitable shelters to overwinter in later.

Rearing of the larvae. In general, rearing of the larvae proved very difficult. We observed high larval mortality particularly due to the drying or decay of host plants. Only three larvae (of 25) pupated in the laboratory, of which one dried out, and another one was destroyed accidentally while examined. A fresh, not fully coloured adult was found in the earthen cell on July 22nd. After emergence, it remained in its pupation cell for at least a few days. The reared beetle was kept alive for observation but died on August 13th. The rearing of larvae on specially prepared host plants (technique 2 as described in Material and methods) was more successful because it prevented predation of the weevil larvae by carabid larvae taken accidentally from the field with the host plants.

Discussion

Erodium cicutarium, i.e. the exclusive host plant of *Coniocleonus nigrosuturatus* recorded in our study, is a very variable herbaceous annual or biennial plant, often regarded as a complex of taxa (SLAVÍK 1997). This plant is supposed to be native to the Mediterranean, partially to western and northern Europe, and reaching central Asia (SLAVÍK 1997). In central Europe, *E. cicutarium* is an archeophyte species. Recently, it has an almost worldwide distribution (SLAVÍK 1997). It is frequently an important weed of cereal and other crops, particularly in semi-arid ranges (FRANCIS et al. 2012). Using the weevil *Coniocleonus nigrosuturatus* as a potential biological control agent would be less effective, in our opinion, compared to some other Cleonini, such as *Cyphocleonus achates* (Fåhræus, 1842) which feeds on invasive spotted knapweed (*Centaurea maculosa* Lam.) and diffuse knapweed (*Centaurea diffusa* Lam.) (STINSON et al. 1994). The larvae of *Cyphocleonus achates* mine the roots, causing extensive root damage, reduced shoot biomass or even death of attacked plants (STEINGER & MÜLLER-SCHÄRER 1992, CORN et al. 2006). However, the larvae of *C. nigrosuturatus* live ectophagously on the underparts of stems and cause only negligible damage to the host plants, which appear perfectly adapted to disturbances, such as grazing, trampling or the feeding of larvae/adults. *Erodium cicutarium* also hosts other weevils in Europe, such as the root borer *Lixus vilis* (Rossi, 1790), the leaf-eating *Brachypera dauci* (Olivier, 1807), and *Limobius borealis* (Paykull, 1792) whose larvae feed on flower buds (KOCH 1992). In studied localities, we often found *Coniocleonus nigrosuturatus*, *Lixus vilis* and *Brachypera dauci* sharing the same habitat and plant.

External larval feeding of *Coniocleonus nigrosuturatus* on the base of the stem is quite unique within Cleonini. A similar feeding place is known only for the larvae of *C. excoriat* that feed at the junction of the stem and the upper taproot of *Emex spinosa* (Polygonaceae) (YEOH et al. 2012). Cleonine larvae more typically feed on the exterior of roots (known for the genera *Asproparthenis*, *Pachycerus* and *Mecaspis*) or are endophagous inside the root/root neck (reported for *Cleonis*, *Cyphocleonus*, *Pseudocleonus* and *Rhabdorrhynchus*) (SCHERF 1964; KOCH 1992; Stejskal & Trnka, unpubl. data).

C. nigrosuturatus appears to be a characteristic species of grazed or even overgrazed habitats (FADDA et al. 2008). For example, this weevil was one of the dominant species of beetle communities in the plain of La Crau (southern France), which has had steppe-like vegetation managed by extensive sheep grazing since the Neolithic period with a high coverage of stones (FADDA et al. 2007, 2008). The weevil occurred only in dry grassland of the original steppe and was absent on formerly cultivated fields, which were used for the cultivation of melons and cereals until the 1970s. The abundance of *C. nigrosuturatus* was positively correlated with very short vegetation (< 5 cm), bare ground percentage cover and plant species richness. This is consistent with our data from Romania, where *C. nigrosuturatus* preferred heavily grazed and trampled places. However, *C. nigrosuturatus* is very sensitive to grazing abandonment, as reported by FADDA et al. (2008), based on long-term experiments on the effects of grazing abandonment on beetle communities in southern France. The authors documented that the abundance of *C. nigrosuturatus* was drastically decreased after only four years of grazing abandonment, and was very low after 23 years of abandonment. Although it is not mentioned in the paper, in our opinion, this may be connected with a change in the abundance of *Erodium*, which also tends to decrease after grazing abandonment (FADDA et al. 2008). We guess that grazing abandonment in warm areas with xerothermophilous steppe grasslands is the main cause of the sharp decline of *C. nigrosuturatus* in the Czech Republic, where this weevil is considered a critically endangered species (BENEDIKT & STREJČEK 2005, STEJSKAL & TRNKA 2013). Nevertheless, our findings in Slovakia (a grassy parking place on a river bank) and Hungary (a closely cut lawn around a petrol station) demonstrate that this weevil can also survive, at least for some time, in ungrazed habitats, both semi-natural and secondary ones. We guess that a proper regime of disturbances that retain sufficient bare ground in the habitat is essential for both *C. nigrosuturatus* and its host plant.

Knowledge of immature stages and life histories can help more effectively protect endangered species (including the species presented here). The detailed description of the larva and pupa and its comparison with known descriptions reported here, demonstrates the possibility of identifying immatures in these species, as has been done in other groups (Hyperini: SKUHROVEC 2006, 2007; *Bagous*: GOSIK 2008; Ceutorhynchinae: GOSIK 2010; Lixinae: NIKULINA 2001, 2007, NIKULINA & GÜLTEKIN 2011, GOSIK & SKUHROVEC 2011, GOSIK & WANAT 2014; Entiminae: GOSIK & SPRICK 2012a,b, 2013, Tychiini: SKUHROVEC et al. in press). In practice, species identification of larvae with chaetotaxy is relatively easy, and is generally much less expensive than identification using molecular methods (HIRSCH et al. 2010). Unfortunately, the relatively low number of available descriptions in comparison to the great number of known adult weevil species represents a current problem in the taxonomic use of the immature stages.

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