Immature Stages of the Cleptoparasitic Bee *Dioxys cincta* (Apoidea: Megachilidae: Megachilinae: Dioxyini)

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ABSTRACT

The fourth larval instar and pupa of *Dioxys cincta* are described and illustrated, and anatomical details of the first and last larval instar are interpreted from cast exuviae. The cocoon of this species is also described. The discovery of a hatched egg containing the first-instar exuviae indicates that the first instar remains mostly surrounded by the egg chorion. The second instar emerges through a hole in the thick dorsal surface of the chorion, presumably by chewing its way out. This discovery and the interpretation of the larval head anatomy of this species and that of *D. pomonae* Cockerell indicate that the genus has five larval instars. The second, third, and fourth instars are adapted to destroying the host egg or larva and any competing cleptoparasites. The fifth instar is not so adapted.

As with *Dioxys*, other cleptoparasitic Megachilidae tend to have a sequential series of instars modified for attacking immatures of hosts and competing cleptoparasites, although which instars are so adapted varies. This contrasts with the cleptoparasitic lineages of the Apidae in which only a single instar, usually the first, is hospicidal.

ÖZET

Kleptoparazitik yaşama sahip olan *Dioxys cincta* (Jurine)'nın dördüncü larva ve pupa evreleri tanımlanmış ve çizimleri yapılmış, birinci ve son larva evrelerinin anatomik yapıları ayrıntılı bir şekilde deri parçacıklarının değerlendirilmesi ile ortaya konmuştur. Benzer şekilde,

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aynı türün kokonu da tanımlanmıştır. Birinci evre larvanın deri parçalarını içeren açılmış yumurta incelendiğinde; birinci evrenin çoğunlukla yumurta korionu ile kuşatıldığı,ikinci evrenin korionun kalın dorsal yüzeyinde muhtemelen çiğneyerek oluşturduğu bir delikten çıkış yaptığı anlaşılmaktadır. Bu tespite ek olarak, bu türün ve *Dioxys pomonae* Cockerell'in larvalarının baş anatomik yapılarının incelenmesinden *Dioxys* cinsinin beş larva evresinin olduğu, ikinci, üçüncü ve dördüncü evrelerinin konukçunun veya rekabet durumundaki diğer kleptoparazitin yumurta ve larvasını parçalama özelliğine sahip olduğu anlaşılmıştır. Beşinci evrede ise böyle bir adaptasyon mevcut değildir.

Dioxys'de olduğu gibi, kleptoparazitik yaşam sürdüren diğer Megachilidae türleri, hangi evre olduğu değişmekle birlikte, konukçu ve rekabet durumundaki diğer kleptoparazitin genç dönemlerine saldırabilme yeteneğine sahip evreleri bulunmakadır. Bu durum, tek evresi, çoğunlukla da birinci evresi konukçuya saldırma özelliğine sahip olan Apidae'nın kleptoparazitik yaşama sahip olan grupları ile zıtlık oluşturmaktadır.

INTRODUCTION

We present information about the immature stages and biology of the cleptoparasitic leafcutter bee *Dioxys cincta* (Jurine) resulting from two field trips that visited the same nesting site in northeastern Turkey in separate years. Discovery of some of the immature stages permits us to reevaluate and refine Micheli's (1936) and Rozen's (1967) interpretations of larval anatomy of the genus and to make comparisons with the immatures of the North American *D. pomonae* Cockerell (Rozen, 1967; Rozen and Favreau, 1967). *Dioxys* is Holarctic in distribution; all other genera of the Dioxyini are from the Old World (Michener, 2000).

The specimens for this study were excavated from a large, complex nesting site occupying a vertical bank at 22 km WSW of Oltu, Erzurum Province, Turkey, in June 2001 and again in July 2003 (fig. 1). The bank was part of an arroyo, which was nearly

Figs. 1, 2.   Vertical nesting site in bank at 22 km WSW of Oltu, Erzurum, Turkey, July 2003. **1.** From a distance. **2.** Close-up of area identified by rectangle in fig. 1, showing numerous burrow openings of various diameters.

waterless at the time of our field studies. Numerous burrows of bees and wasps penetrated the face of the bank (fig. 2). The complexity of anastomosing fresh and old tunnels suggested that the embankment had served the nesting requirements of these animals over many years. Nonparasitic bee genera nesting there included, among others: *Hylaeus* (Colletidae); *Osmia*, *Protosmia*, *Megachile* (Megachilidae); and *Xylocopa* (*Proxylocopa*) and *Anthophora* (Apidae). Parasitic genera collected from the site included *Coelioxys*, *Dioxys*, and *Stelis* (Megachilidae: Megachilinae) and *Melecta* and *Thyreus* (Apidae: Melectini).

When Rozen (1967) described the immatures of *Dioxys pomonae*, he concluded that the species had four larval instars. He knew that an earlier cryptic stage was known for some bees and was termed the ''first instar'' by Hackwell and Stephen (1966). He looked for but did not detect such a stage in *D. pomonae*. Since then he has become aware that the first instar of many (but apparently not all) bees is inactive relative to subsequent instars and remains mostly surrounded by (i.e., pharate within) the egg chorion; in such cases the second instar is the first to become active (e.g., Hackwell and Stephen, 1966; Baker, 1971; Torchio et al., 1988: Torchio, 1989; Garófalo and Rozen, 2001). On reviewing pictures of the *D. pomonae* study, Rozen was convinced even before the present study that the apparent egg ''just before'' hatching in figure 5 of Rozen and Favreau (1967) was actually the first instar surrounded by the chorion and that the instars described by Rozen (1967) were actually the second, third, fourth, and fifth and not the first, second, third, and fourth. The original picture of the true first instar has been digitized, cropped, and reproduced here (fig. 4) to provide more details of the appearance of a pharate first instar. The picture of the egg from the former study (Rozen, 1967: fig. 4) is also shown (fig. 3) so that visual comparisons can be made between the two stages. In the following passages, we assume that all species of *Dioxys* have five instars, and we identify them accordingly.

EGG AND FIRST LARVAL INSTAR

Presumably entangled in the outer surface of the cocoon, a shed chorion of *Dioxys cincta* with its nodular dorsal chorion[3] intact (see Rozen and Özbek [2003] for a description of the unusual egg/mature oocyte of this species) was preserved with a female pupa that had been accidentally killed during our excavation. When we examined the chorion in the laboratory, we discovered the cast skin of the first instar within it. The skin, though colorless and nearly featureless, could be identified by the crossed mandibles (fig. 5), which were slender, slightly curved, and tapered gradually to a single point, unlike the bidentate mandibular apices of the presumed first instars of *Stelis chlorocyanea* Cockerell (Rust and Thorp, 1973) and *S. elongativentris* Parker (Rozen, 1987). This discovery confirms the conviction (expressed above) that *Dioxys* has a pharate first instar within the egg chorion and that the second instar is the one that first becomes active. Other identifiable features of the exuviae, seen through a compound microscope, were the salivary opening below the mandibles and the spiracles. A linear string of granules (spicules?) ran between the spiracles on each side of the body, as was noted by Alves dos Santos et al. (2002) on the first instar of *Tetrapedia diversipes* Klug (Apidae: Apinae: Tetrapediini). Because such linear series of granules are found in separate families, they are probably a widespread phenomenon among first instars of bees. Their function is unknown but may be associated with the rupturing of the chorion during the early stages of eclosion, as the chorion of some bees is known to split along the spiracular line on each side (Rozen, 1964: fig. 5; Alves dos Santos et al., 2002).

The hatched egg (fig. 6) was 1.63 mm long and 0.88 mm in maximum width. The eclosion hole was 0.38 mm from the anterior end, with the latter being identified by the micropyle occurring on a shred of the thin ventral chorion. The hole (fig. 6) was off-center,

[3] Rozen and Özbek (2003) concluded that the thick nodular surface of the chorion was the dorsal surface because it seemed likely that the egg is placed against the cell wall with this surface exposed (rather than the thin-shelled opposite surface) to thwart discovery and attack by the returning host female. The emergence hole in the shed chorion supports this assumption, but the discovery of the egg placement of this species has yet to be made.
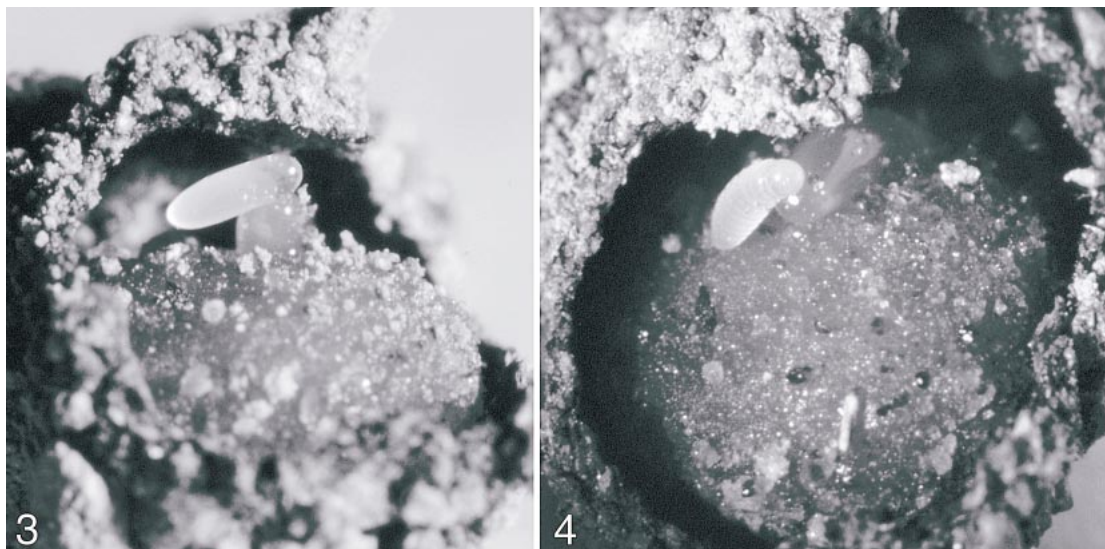
Figs. 3, 4. Early stages of *Dioxys pomonae* resting on the egg of *Osmia nigrobarbata* Cockerell. **3.** Egg before hatching. **4.** Pharate true first instar that developed from same egg. For further explanation see text.

more-or-less transverse, and slightly askew. Its maximum width and height were 0.25 and 0.11 mm, respectively. Its borders were somewhat irregular and the chorion around it was milky (not visible in fig. 6 because of backlighting), contrasting with the clearer chorion elsewhere. Possibly the milky area was created by an eclosion fluid that modified the texture of the chorion. How the second instar penetrates the thick chorion is unknown, but the irregular nature of the opening suggests that the second instar might chew its way out. If this is true, then this is an unusual form of eclosion for a cleptoparasitic bee larva; most have chorions that split

along defined lines (e.g., Rozen et al., 1978; Rozen, 1994, 2003).

By dividing the length (1.63 mm) of the egg by the maximum distance (3.60 mm) between the outer rims of the pupal tegulae, we calculated an egg index of 0.45. The index is a system of defining the size of a bee's egg relative to the size of the female's body, as developed by Iwata and Sakagami (1966). The egg index in this case is somewhat greater than 0.39, which Rozen and Özbek (2003) calculated for the same species using mature oocytes, but both values fall within the "dwarf" category defined by Iwata and Sakagami (1966: table 2).

MATERIAL STUDIED: One exuviae of first larva instar enclosed in egg chorion, Turkey: Erzurum, 22 km WSW Oltu, VII-28–2003 (J.G. Rozen, H. Özbek).
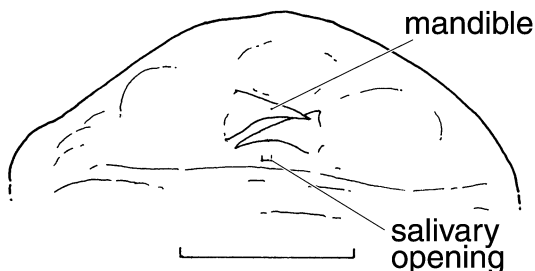
FOURTH LARVAL INSTAR

The recovery of a fourth larval instar of *Dioxys cincta* makes possible its description and a comparison with the cast skin of the last (fifth) larval instar. Its assignment to the fourth larval stage is based on its large size and on the fact that the relatively short antenna of the last instar and its bidentate man-

Fig. 5. Anterior part of cast exoskeleton of first instar of *Dioxys cincta* taken from hatched egg. Scale line = 0.1 mm.

mandible

salivary opening

Fig. 6.   Hatched egg of *Dioxys cincta* showing presumed eclosion opening through which second instar emerged, dorsal view.

dibles were visible within the cleared head capsule.

The single specimen was first studied and drawn before the head capsule was cleared in an aqueous solution of sodium hydroxide. The capsule and the sclerotized portions of the mouthparts were darkly pigmented in certain areas. After clearing, much of the pigmentation was lost, suggesting that dark pigments are subcuticular. Although sclerotized areas tend to be faintly darker than nonsclerotized areas on the cleared head, labral and maxillary sclerites, which were darkly pigmented on uncleared specimens, were rendered nearly colorless on cleared preparations. Cast larval skins are shed with the dark pigments in place, as evidenced by exami-

nation of two cast fifth-instar exuviae of this species and a single cast skin of a third instar of *Dioxys pomonae* from an earlier study by Rozen (1967). Thus, in the following description of the head, descriptions of dark pigmentation refer to the uncleared specimen. The dark pigment, while presumably noncuticular, indicates areas of sclerotization.

The specimen described was nearing ecdysis when preserved. The unexpected loss of pigmentation during the clearing treatment, the obfuscation caused by the developing fifth-instar cuticle within the cleared head, and our lack of making detailed sketches of pigmentation and sclerites before treatment combine to make certain features in the following description and accompanying illustrations uncertain, as indicated.

LENGTH: 8.0 mm.

HEAD (figs. 9–11): Integument of head capsule and mouthparts with scattered, conspicuously elongate, setiform sensilla. Much of sclerotized integument tan (internal ridges darker) becoming mottled toward top of head capsule, all as seen on uncleared head; extensive broad colorless ecdysial lines visible on uncleared head (fig. 10) but completely obliterated after clearing.[4]

Head more or less hypognathous, distinctly not prognathous; parietals swollen so head appearing broad, with vertex at midline lower than parietals in frontal view (fig. 10); head capsule sharply constricted behind both dorsally and laterally, so that diameter of foramen magnum much smaller than outline of head in frontal or rear view (fig. 10). Tentorium probably complete and well developed but specimen approaching ecdysis, so that internal skeletal structures missing. Anterior and posterior tentorial pits in normal position (figs. 8, 9); postoccipital ridge well developed; hypostomal ridge well developed, dark on uncleared specimen; pleurostomal ridge dark on uncleared specimen but not easily identifiable on cleared specimen, probably because of thick integument of head capsule; epistomal ridge not evident on either cleared or uncleared head capsule, probably because of thick integument of capsule (an-

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[4] We observed similar ecdysial lines on the uncleared cast skin of a third instar of *Dioxys pomonae* and thus deduced their function.
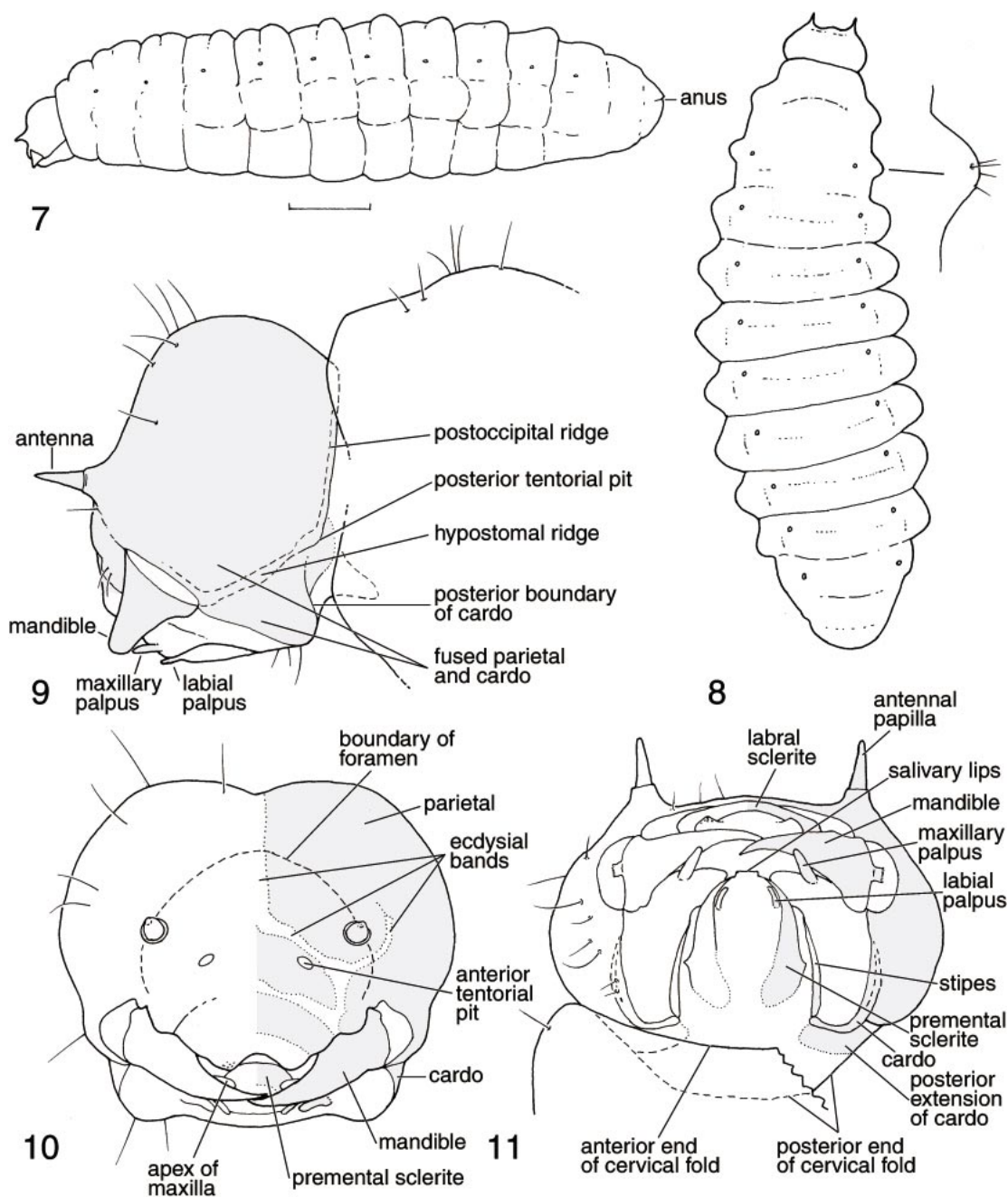
Figs. 7–11. Fourth larval instar of *Dioxys cincta*. **7.** Entire larva, lateral view, setae not shown. **8.** Same, dorsal view, with setae indicated on enlarged lateral swelling. **9.** Head, lateral view, with sclerotized areas diagrammatically depicted by uniform gray overtone (in actuality, depth of pigmentation varies on sclerotized areas on uncleared specimen, as indicated in description); pigmentation of front of face and labium not certainly known in this view. **10.** Head, frontal view, with setae approximately represented on left and pigmentation represented diagrammatically on right by uniform gray tone; broad unpigmented ecdysial bands only roughly bilaterally symmetrical on actual specimen. **11.** Head, ventral view, with setae approximately represented on left and pigmentation represented diagrammatically on right. Scale line (= 1.0 mm) refers to figs. 7 and 8.

terior tentorial pits appear as conspicuous invaginations of front of head and not on ridge) (faint internal epistomal ridge detected on cleared head capsule of fourth instar of *Dioxys pomonae* laterad of anterior tentorial pit). Antenna arising abruptly from front of head (figs. 9, 10); papilla elongate, tapering, apically pointed, as least three times longer than basal diameter, and with approximately three sensilla tightly clustered at apex. Labrum with apical median emargination resulting in lower clypeal edge having somewhat pointed, sensilla-bearing projection on each side of emarginaton; labral sclerite darkly pigmented, distinct (fig. 10) (but not evident on cleared head capsule).

Mandible (figs. 9–11) large, elongate, curving to single apical tooth; dorsal and ventral edges somewhat uneven but without teeth; cuspal area not developed. Labiomaxillary region recessed in lateral view (fig. 9). Cardo (fig. 9) a large triangular sclerite fused at base with parietal along entire hypostomal ridge, this sclerotization extending backward behind junction of postoccipital ridge with hypostomal ridge at posterior tentorial pit; stipes (fig. 11) a narrow, darkly pigmented sclerite on uncleared head (scarcely noticeable on cleared head); maxillary palpus large, much longer than basal diameter; galea not evident. Labium not divided into prementum and postmentum; premental sclerite evident as elongate, inverted U shape, with posterior ends not meeting, as seen in ventral view of head (fig. 11) on uncleared specimen; labial palpus long, distinctly smaller than maxillary palpus. Salivary lips (fig. 11) slightly projecting, narrowly transverse (hidden by mandibles in frontal view, fig. 10).

BODY (figs. 7, 8): Integument with often elongate, setiform sensilla; these sensilla not abundant (but conspicuous because of length), primarily on posterior part of pronotum, caudal annulets of other body segments, apices of lateral body tubercles, and anal area of abdominal segment 10; spicule pattern not examined. Intersegmental lines moderately incised except those of abdominal segments 8–10, which may have been altered by approaching ecdysis or postmortem changes; most body segments dorsally divided into cephalic and caudal annulets, with caudal annulets higher than cephalic annu-

lets; dorsolateral body tubercles absent; conspicuous lateral body tubercles present below spiracular level on following segments: meso- and metathorax, abdominal segments 1–8; thoracic body tubercles less pronounced than most abdominal ones as seen from above (fig. 8); abdominal segment 10 oriented centrally on 9 and anus centrally positioned on segment 10 as seen in lateral view (fig. 7). Spiracles not studied.

MATERIAL STUDIED: One larva, Turkey: Erzurum, 22 km WSW Oltu, VI-23–2001 (J.G. Rozen, H. Özbek).
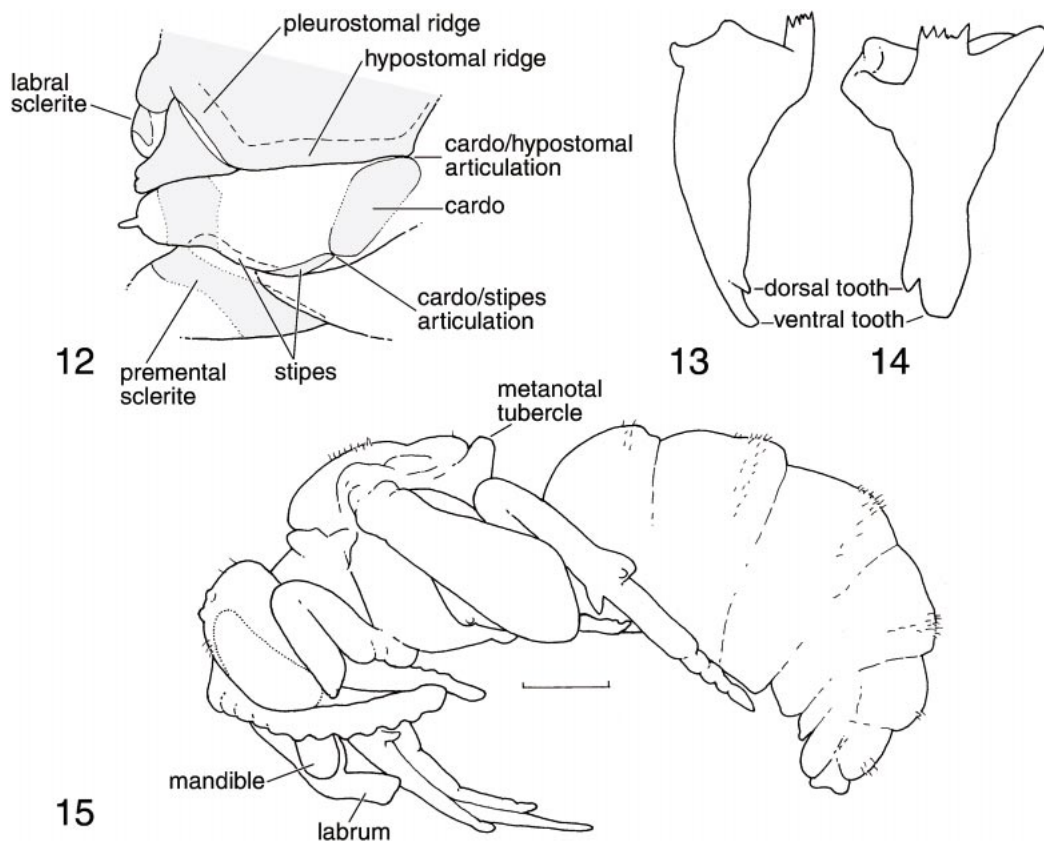
FIFTH LARVAL INSTAR

The last larval instar of *Dioxys cincta* was described and illustrated by Micheli (1936). Recovery of the cast larval skins associated with the pupae allows us to (1) add details to augment his description of the head, (2) compare it with the fourth instar, described above, and (3) compare it with the fifth instar of other species of the genus. Although description of the pigmentation is based on that of the untreated cast skin, one of the cast skins was boiled in an aqueous solution of sodium hydroxide. Unpredictably, no diminution of pigmentation was detected in contrast to the treatment of the head capsule of the fourth instar. The labral sclerite, maxillary sclerites, and internal head ridge remained dark.

DIAGNOSIS: The last instar of this species can be distinguished from those of *Dioxys pomonae* and another species tentatively identified as *D. productus* (Cresson)? by its somewhat shorter antennal papilla, as previously suspected by Rozen (1967).

HEAD (fig. 12): Integument of head capsule and mouthparts with scattered, conspicuously elongate, setiform sensilla, much as described for fourth instar. Integument of head capsule pale tan except following darkly pigmented: postoccipital ridge, hypostomal and pleurostomal ridges (but not epistomal ridge).

Head hypognathous according to Micheli (1936: fig. 1); parietals not swollen. Anterior tentorial pit not identified; posterior tentorial pit in normal position; postoccipital ridge well developed; hypostomal and pleurostomal ridges well developed; epistomal ridge

Figs. 12–15.   *Dioxys cincta*. **12.** Lower part of head of cast exuviae of fifth larval instar, lateral view, setae omitted and nonsclerotized cuticle partly reconstructed, drawn to same scale as head of fourth instar (fig. 9). **13, 14.** Right mandible of cast exuviae of fifth larval instar, dorsal and inner views, respectively. **15.** Pupa, lateral view. Scale line = 1.0 mm.

not evident. Antennal papilla moderately long, its length somewhat less than two times basal diameter, tapering apically, and with approximately three sensilla (Micheli, 1936, counted four). Labrum with apical median emargination; labral sclerite broadly transverse, darkly pigmented.

Mandible, as seen in inner view (fig. 14), broad at base, tapering first abruptly toward apex, then tapering gradually so as to become almost parallel-sided, and finally broadening to form two apical teeth; dorsal tooth subapical, smaller, more sharply pointed than ventral tooth; ventral tooth wide, subtruncate; mandible as seen in dorsal view (fig. 13) tapering gradually, curved; cuspal area not produced; adoral surface slightly concave apically; setae and tubercles not evident on outer surface. Labiomaxillary region

probably projecting in lateral view. Cardo a moderately large sclerite but much more restricted than that of fourth instar, articulating at one point with rear of hypostomal ridge as in most bee larvae that spin cocoons, not fused with parietal as in fourth instar; stipes a narrow, darkly pigmented sclerite extending forward from the cardo/stipes articulation; at point where maxillary apex becomes distinct from labial apex, stipital sclerite widening and circumscribing maxilla before ending; maxillary apex not sclerotized; articulating arms of stipes long, darkly pigmented, conspicuous; maxillary palpus long, about as long as antennal papilla, darkly pigmented; galea not evident. Labium almost certainly divided into prementum and postmentum; premental sclerite conspicuous, surrounding prementum (and not interrupted ventrally as

in fourth instar); labial palpus long but smaller than maxillary palpus, pigmented. Salivary lips strongly projecting, transverse, wide (as illustrated by Micheli, 1936: fig. 2–1).

BODY: The cast larval exoskeleton provides no information about body contour, but Micheli's (1936: fig. 1–1) illustration shows that most body segments are divided dorsally into cephalic and caudal annulets. The integument has setiform sensilla, some of which are moderately long and pigmented. Spiracles on the cast skin are large, as so stated by Micheli; other spiracular features seem to agree with the illustration and description of those of *Dioxys productus*? given by Rozen (1967: fig. 10): atrial wall with ridges; atrium projects beyond body wall, with rim; peritreme narrow; subatrium short. In addition, the atrial wall has denticles, as also mentioned by Micheli (1936).

MATERIAL STUDIED: Two cast fifth-instar exuviae, Turkey: Erzurum, 22 km WSW Oltu, VII-28–2003 (J.G. Rozen, H. Özbek) from separate cells.

COCOON

We retrieved two cocoons of this species from different nests. In neither case was the host identified. One cocoon contained a pupa, which was accidentally killed during our excavation; the other held a postdefecating larva that pupated the day after discovery. This pupa was studied and illustrated while alive, and the species identification was verified after the adult emerged. The cast larval skins associated with both pupae are described above; the pupa is described below.

The two cocoons were of pale cream color, contrasting with the dark brown fecal pellets that adhered loosely to their fuzzy exterior surface. The cocoons did not cling closely to the cell walls, as they could easily be removed from the substrate. No fecal material was incorporated in the cocoon fabric itself, and no feces occurred in the interior of the cocoon, indicating that defecation takes place prior to spinning. The cocoon fabric was semitransparent and soft (nonrigid) with numerous silk threads visible on the outer surface, which therefore appeared dull. The inner surface was somewhat shinier because the silk strands fused with one another there. One cocoon measured 11 mm long and 8.5 mm in maximum diameter. The front end was somewhat more pointed than the rear but was not as acutely pointed as the cocoon pictured by Micheli (1936: fig. 6). The fabric was thicker and denser at the pointed end than elsewhere, an indication that the pointed end was a weak nipple, a specialized feature seen on other megachilid cocoons. The variation in the distinctiveness of the nipple may depend on the size of the cell in which the cocoon is spun.

The cocoon of the North American *Dioxys pomonae* described by Rozen and Favreau (1967) contrasted with that of *D. cincta* described here. Not only did it have a more distinct nipple anteriorly, but it also was dark in tone, rigid, and consisted of three layers: an outer more fibrous one, a middle layer of dark feces (accounting for its overall dark appearance and rigidity), and an inner one that was ''loose and light brown'', forming ''a cellophane-like coating even though some individual silk strands were detected.'' Thus the differences in the cocoons of the two species can be explained by the behavioral trait of whether the larva defecates before cocoon spinning (*D. cincta*) or whether feces are sandwiched between two layers of silk (*D. pomonae*).

PUPA

The pupa of *Dioxys cincta* (fig. 15) is nearly identical to that of *D. pomonae* (Rozen, 1967) except for size. The median metanotal tubercle on pupae of these two species, accommodating the development of the same feature on adults, permits easy identification of the pupae to genus. The pupa of *D. cincta* will readily key correctly to genus in Rozen (2000). A formal description is unnecessary here because the pupal description of *D. pomonae* given by Rozen (1967) applies equally to that of *D. cincta* except that setae are found on the vertex of the latter (fig. 15).

MATERIAL STUDIED: One pupa, Turkey: Erzurum, 22 km WSW Oltu, collected as larva VII-28–2003, pupated VII-29–2003, adult emerged ~VIII-22–203, preserved VIII-27–2003 (J.G. Rozen, H. Özbek).

DISCUSSION

We interpret both the swollen parietals and the strong constriction of the posterior part of the head capsule of the fourth larval instar to be adaptations providing a stable platform for the strong mandibular musculature, with the latter being necessary for attacking host immatures and those of other cleptoparasites. The enlargement and fusion of the clearly defined cardines with the parietals are similarly interpreted as a ventromesal extension that adds firmness to the head capsule. This combined enlargement of the cardines and their fusion to the head capsule is an unusual feature in bee larvae. It is quite visible in the cast head capsule of a third instar of *D. pomonae* that was attached to the fourth instar described (as a third instar) by Rozen (1967). The fourth instar from that study was cleared with an aqueous solution of sodium hydroxide so that its pigmentation is no longer visible, but the cast skin of the third instar remained untreated in the collection of the American Museum of Natural History. Its large fused cardo is similar to that described here for the fourth instar of *D. cincta*, an indication that third instars of at least these two species are equipped to attack.

It seems likely that in all species of *Dioxys*, the second, third, and fourth instars will exhibit this fusion since mandibles of these instars of *D. pomonae* in Rozen (1967: figs. 21–23, 27, 28) and the fourth instar of *D. cincta* pictured here (figs. 10, 11) are massive, elongate, strongly curved, and sharply pointed, and are obviously weapons of attack against host or competing cleptoparasite immatures. The fact that the cardo of the fifth instar of *D. cincta*, as indicated above, articulates with the rear of the hypostomal ridge suggests that the last larval instar is not hospicidal, as do its apically bidentate mandibles, with the upper tooth only slightly shorter than the rather blunt ventral one.

It is instructive to compare these larval adaptations for cleptoparasitism with those of other cleptoparasitic Megachilidae. Larvae of other Dioxyini, all of which are parasitic, are unknown. Distantly related parasitic clades in the Megachilinae whose larvae have been described show certain similar (but nonhomologous) adaptations. Thus, the second and third instars of *Coelioxys* have strongly sclerotized head capsules that are bridged ventrally by a nearly complete sclerotization of all but the apex of the labiomaxillary region. This construction provides a firm platform for the massive mandibular musculature required for the huge mandibles (Baker, 1971). The ventral sclerotization of the labiomaxillary region can also be interpreted as armament, shielding this part of the head from attack. The foramina magna of *Coelioxys* are constricted, further strengthening the head. Baker pointed out that the third instar is the stage that usually kills the host egg or young larvae. Subsequent instars have a more normal anatomy and are presumably nonhospicidal.

Torchio (1989) published an excellent account of the development of *Stelis montana* Cresson that clearly showed that the species has five larval instars, with the first instar being pharate within the egg chorion and with the fifth instar alone being hospicidal. Many earlier published observations regarding *Stelis* must be cautiously interpreted because it has not been generally recognized that first instars of many bees are inactive and pharate within their egg chorions. None of these earlier accounts recorded the total number of instars, and thus the true first instars may have been overlooked.

Michener (1955, and references therein) concluded several species of *Stelis* may battle immatures of host or other cleptoparasites when the *Stelis* are partly grown. He observed the mature larva of *S. lateralis* Cresson killing the host. He reported that the mandibles of the species are "acutely pointed . . . throughout larval life", but it is unclear that he actually observed the true first instar. Rust and Thorp (1973) concluded that the supposed first instar of *S. chlorocyanea* Cockerell "usually does not destroy the host, but later forms do." Although they term the mandible of this instar as bidentate, their illustrations (1973: figs. 10, 11) depicted it as sharply pointed with one tooth much shorter than the other, and that they observed one "first instar" feeding on the egg of the host (p. 554). They reported that the "second instar" has the upper tooth reduced to an obtuse angle and subsequent instars have elongate, curved mandibles with acute apices. We

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think that they may have actually seen the true first-instar as they watched late embryogenesis and may have overlooked the simultaneous shedding of the chorion and first-instar cuticle, just as Rozen and Favreau (1967) had overlooked this phenomenon in *Dioxys pomonae*.

Rozen (1987) found that the supposed first instar of *Stelis elongativentris* Parker, ''still partly in its chorion'', had bidentate mandibles that probably would be incapable of killing the host egg. Although this larva may have been a true first instar, it was also reported to have consumed some pollen, which has not been an activity of known pharate first instars. The mandible of the last instar is also bidentate, but the dorsal tooth is small and far removed from the sharply pointed apex of the robust ventral tooth. This feature and the enlarged, heavily sclerotized parietals strongly suggest that the last instar is equipped to battle host larvae and perhaps competing larval cleptoparasites.

In contrast to these megachilid cleptoparasites, the last instar (as well as the supposed first instar) of *Hoplostelis bilineolata* (Spinola) has bidentate, apically broad mandibles, and the parietals are not swollen and heavily sclerotized (Rozen, 1966). Also in contrast to the other megachilid cleptoparasites, the adult female *H. bilineolata* is known to kill the host egg (or young larva) (Bennett, 1966). As implied by Michener (2000), the differences in the mode of parasitism between *Hoplostelis* and *Stelis* suggest that these genera did not have a common parasitic ancestor.

An interesting feature of many cleptoparasitic megachilids whose larvae are hospicidal is that a sequential series of instars (not just a single instar) may be capable of dealing with host immatures or competing parasitic larvae, although *Stelis montana* (Torchio, 1989) and perhaps other *Stelis* species are exceptions. On the basis of limited data, we venture the following preliminary conclusions: the second, third, and fourth instars of *Dioxys* exhibit hospicidal adaptations; the second and third instars of *Coelioxys* are hospicidal, but with *Stelis*, depending on the species, all larval instars may be hospicidal (although we strongly suspect that the first instar needs reexamination) or only one, the

last instar, is hospicidal. These observations contrast with what we know about the cleptoparasitic lineages of the Apidae. Only one instar, the first, is hospicidal in the Nomadinae, Protepeolini, Isepeolini, Melectini, Tetrapediini (*Coelioxoides*), Rhathymini, and Ericrocidini (Rozen, 1991; Alves dos Santos et al., 2002). In the case of *Exaerete*, only one instar, the second, may be hospicidal, although the modifications of this instar may have been driven by competition with other cleptoparasite immatures since the cleptoparasite mother attempts to eliminate the host eggs when she oviposits (Garófalo and Rozen, 2001, and references therein). It will be interesting to see if future studies substantiate this difference between cleptoparasitic Megachilidae and Apidae.

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