The previously unknown larva of *Elpiscladius capicola* Harrison & Cranston (Diptera: Chironomidae) is described. It was predicted by postulated phylogeny to be associated with aquatic immersed leaves or wood, and has been found. The larva is xylophagous, occurring in a limited area of the Western Cape, in the Eerste River and Leopard’s Kloof stream in the Hottentots Holland mountains. Association with *E. capicola* is assured by a distinctive (pharate) pupa visible within one mature larva, and the co-occurrence of larvae and diagnostic pupal exuviae in soft immersed wood. The characteristic mentum of *Elpiscladius* resembles that of *Xylotopus* Oliver, a Nearctic wood-mining genus, but other features including the dorsal head sclerites and the simple SI seta differ. Although phylogeny predicted the larval biology, addition of 17 larval features for *Elpiscladius* to that data matrix destroyed the relationship with *Austrobrillia* Freeman, reducing the Brillia group essentially to a polytomy.

Larvae of *Elpiscladius* co-occur with the immature stages of an undescribed species of *Stenochironomus* (s.s.) Kieffer. Both these rare xylophagous taxa indicate clean flowing waters with intact native riparian vegetation, and thus can be regarded as biodiversity and environmental conservation indicators.

Key words: immersed wood, phylogenetic prediction, mining, aquatic conservation.

INTRODUCTION

The immature stages in the Diptera family Chironomidae can dominate, in terms of species diversity, the macroinvertebrate biota of running water: there is substantial representation also in standing waters. The Afrotropical chironomid fauna was catalogued by Freeman & Cranston (1980), who brought many disparate published names into a modern framework based increasingly on immature stage morphology. With revisionary study has come increased recognition especially of pupal morphology in both biodiversity estimation and in systematics. Cast pupal skins (exuviae) left on the water surface as the adult emerges, can provide accurate estimates of community structure without intensive sampling of either adult or larvae. Such sampling has revealed cryptic diversity, in the sense of taxa uncollected by standard methods, assuming that adults are ephemeral and larvae may occupy unsampled aquatic microhabitats. Just such an example involves a rarely encountered pupal taxon, including one pharate (a male retained within its pupal exuviae), found in two streams of the Western Cape in the past decade. With no candidate larvae known despite extensive sampling in the area, the species was described in the subfamily Orthocladiinae, as a new genus *Elpiscladius* with species *E. capicola* (Harrison & Cranston 2007). The results of a morphological phylogenetic analysis including data from all available life history stages, indicated that the species belonged in a clade unrecognized previously in southern Africa, namely the Brillia group. The immediate relationships of *Elpiscladius* within this group predicted that the unknown larva could live in either aquatic leaf packs, or in immersed wood (Harrison & Cranston 2007).

In mid-summer 2007 opportunity arose to survey streams from within which pupal exuviae had been found, in the Hottentots-Holland Mountains of the Western Cape. Intensive sampling of in-stream leaf packs derived from both native and alien riparian vegetation revealed no candidate larvae; shredders present were freshwater river crabs (*Potamonautes* sp.), amphipods or chironomid larvae belonging to *Polypedilum*, a genus of Chironominae. Submerged wood of terrestrial origin was uncommon, although many once-immersed branches and sticks were stranded above the water level, representing post-flood deposition from April 2005 when some 200 mm rain fell in 24 hours in the catchment. However, seeking immersed wood firmly wedged into the
rocky stream bed revealed some sticks soft enough
to be fragmented by hand, certain of which con-
tained pale-bodied chironomid larvae with dark,
sclerotized head capsules. Furthermore, some
pupal exuviae found in the same wood could be
identified as belonging to *Elpiscladius*. With verifi-
cation that the larvae were indeed wood-mining,
subsequent survey revealed the larvae occurred
somewhat more widely, and in a greater range of
wood types, within the upper Eerste River drain-
age, in Jonkershoek Nature Reserve, east of
Stellenbosch.

Here the larva is described and illustrated, some
additional comments provided on the evolution-
ary relationships and remarks made on under-
sampled aquatic refugial habitats and their conser-
vation.

**MATERIAL AND METHODS**

Immersed wood was sampled initially by pulling
apart soft ‘punky’ pieces in a dish under a dissec-
tion microscope. Larvae were encountered rarely,
and harder wood could not be prized apart for
inspection. By chance, a plastic 25 litre bucket
containing recovered wood, filled to immersion
with in-stream water was left in a cold room,
temperature 6°C, for three days. This simple treat-
ment encouraged departure of live larvae from
their residence. Individual larvae were isolated
into 10 mm vials with a little fragmented wood and
native water in an unsuccessful attempt to rear to
adults. Larvae were treated in one of three ways:
some were preserved whole into 100 % isopropanol
for subsequent DNA extraction and sequencing;
some were mounted on microscope slides using
Hoyer’s mountant, with slides ringed subsequently
with clear nail varnish, others were cleared and
dehydrated in isopropanol and slide-mounted in
Euparal. Pupal exuviae also were mounted in
Euparal.

Morphological terms used in the description are
standard and together with abbreviations follow
Sæther (1980) and Cranston (1994). Material is
housed in the Albany Museum, Grahamstown,
South Africa (AMGS), the South African National
Collection of Insects (SANC), the Natural History
Museum, London, United Kingdom (BMNH), and
the Australian National Insect Collection, CSIRO,
Canberra City, Australia (ANIC).

**Material examined (all collected P.S. Cranston)**

SOUTH AFRICA: 10 larvae (6 slides, 2 each to
AMGS, BMNH, ANIC), 3 pupal exuviae (2 slides,
SANC), Western Cape, Betty’s Bay, Harold Porter
National Botanic Garden, Leopard’s Kloof stream,
34°21’S, 18°56’E, .i–.ii.2007; 1 larva, 1 pharate pupa
(within larva) (BMNH), Jonkershoek Nature
Reserve, Upper Eerste River, White Bridge,
33°59.38’S, 18°58.30’E, .i–.ii.2007; 11 larvae (4 slides,
SANC). ‘2nd waterfall’, 34°00.21’S, 18°59.40’E,
.i–.ii.2007; 2 larvae (2 slides, AMGS), Lang Rivier,
33°59.16’S, 18°58.14’E, .ii.2007.

**RESULTS**

*Description of Elpiscladius capicola 4th instar Larva (n = 10)*

All measurements are in µm unless stated
otherwise.

Body length 8.1–8.5 mm; body white, with
distinctive wood fibre-filled gut (Fig. 10). Head
capsule 640–740 long, light brown-yellow with
slightly darkened postoccipital margin, black
mandibles, and dark brown-black mentum. Dorsal
surface of head (Fig. 1) comprising frontal
apotome, a single sclerite bearing S2 and S3
comprising clypeus + labral sclerite 1, separated
from anteriormost, elongate, narrowed sclerite 2
(bearing SI). Sclerites 3, 4 and 5 not differentiated.
Antenna (Figs 2, 8) five-segmented, 75–84, 20–22,
5–7, 8–10, 6–7, AR 1.7–2.0, blade annulate, 57–70
long extending well beyond antennal apex;
style/peg sensillum placed subapically on second
segment, extending to mid-fourth segment, 13–16
long. Lauterborn organs absent. Ring organ in
basal quarter of segment 1, with two adjacent pits.

Labrum (Fig. 3) with SI simple, lanceolate,
arising from stout adjacent bases; SII and SIII
simple, bases adjacent to each other, posterior to SI
bases; short SIa & b posterolateral to SIII base. Pair
of spinulae arise close to base of SII, simple. Labral
lamellae absent, or perhaps represented by cluster
of few minute (c. 1 µm) tubercles in mid-labrum;
labrum strongly extended anterior to SI setae in
smooth plate with scalloped anterior margin.
Epipharynx (Fig. 3) with three rounded lobes
forming pecten epipharyngis; three pairs of
robust chaetulae, posterolateral pectinate, antero-
medial simple; with two small chaetulae basales.
Premandible dark, broad and blunt, with one apical
tooth, without brush. U-sclerite strongly
sclerotized.

Mandible (Figs 4, 7, 9) heavily sclerotized, with
outer and inner margins smooth, with long apical
tooth subequal to width of three inner teeth (no
evidence of any outer, dorsal tooth); three inner teeth of which innermost is smaller, and partly fused to darkened mola from which narrow, terminally trifid seta subdentalis arises and extends to second inner tooth. Seta interna with 4–5 narrowly serrate branches.

Mentum (Figs 5, 7) 320–380 wide, with elongate anterior-projecting, paired median teeth separate by U-shaped cleft in which two small, blunt teeth are evident; with four pairs of lateral teeth, third and fourth subequal in size; without evidence of ventromental plates or beard.

Maxilla (Figs 6, 9) lying dorsal to sclerotized, anteriorly-projecting cardo (Fig. 9, arrowed); palpiger with well-developed maxillary palp, numerous fine, hyaline, simple galear lamella, basally (underlying cardo) with sclerotized transverse ridges; without pecten galearis; lacinial
chaetae all serrate, numerous and dense, without differentiated antaxial, anterior lacinial or appendix on mesal margin (the posteriormost chaeta may represent an undifferentiated appendix).


**Differential diagnosis**

The larva of *Elpiscladius* shares a characteristic anterior-projecting, medially deeply-incised mentum (Fig. 5) with *Xylotopus* Oliver, and resembles that genus in other features including the mandible shape and the five-segmented antenna lacking any hyaline division of the second seg-

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**Figs 7–10. Elpiscladius capicola.** 7, Ventral view mandibles, mentum and labrum; 8, antenna; 9, antenna, mandible, maxilla; 10, wood fibres in gut.
ment. However, the absence of labral lamellae, the simple (not plumose) labral S1 setae, and the structure of the dorsal head sclerites differ, as does the absence of the marginal setal tufts on the body that characterize *Xylotopus*. The location of cephalic setae S2 and S3 on the same sclerite, considered to represent fusion of the clypeus with labral sclerite 2, is unusual but, given the range of conditions observed in the Brilla group, is unlikely to be diagnostic.

**DISCUSSION**

The verification of the predicted xylophagous larval habitat for *Elpiscladius* adds another taxon to the Brilla group that appear to have adopted this substrate early in the evolution of the subfamily Orthocladiinae, and to have radiated in this and similar habitats. That a wood-mining member of the Brilla group has been found in South Africa, the earliest departing (or remnant core) continent from Gondwana (Sanmartin & Ronquist 2004), may indicate an ancient association, extending the clade and the habit back to an early period when a united Gondwana included Africa. With the loss of the phylogenetic (and therefore biogeographic) signal within the Brilla group when the larva of *Elpiscladius* is added to the matrix, an alternative interpretation remains possible: closer relationships of *Elpiscladius* to northern hemisphere Brilla group taxa must be re-considered.

The immature stages of the previously east Palearctic genus *Tokyobrillia* from the Eastern Arc Mountains of Tanzania (Sæther & Ekrem, 2003) now may be considered to provide, with *Elpiscladius*, evidence of a continuous track for northern Brilla group taxa down the African continent joined (or remnant core) continent from the Miocene creating semi-continuous suitable habitat for chironomids to move southward (Harrison 1992; Willansen & Cranston 1986). Differentiating these scenarios may require evidence from molecular analyses, which are under way but constrained by the need for adequate representation from some extremely uncommon, globally-distributed taxa. Never-the-less, ongoing molecular studies (Morse & Cranston in preparation) postulate *Elpiscladius* as sister taxon to the Australian-Neotropical *Austrobatilla*, with northern hemisphere, including Asian, Brilla group taxa more distant. Furthermore, evidence that the evolution of the subfamily Chironominae shows a similar ‘austral’ pattern (e.g. *Xylochironomus* Cranston 2006; the Stenochironomus group and *Imparipsecten* Cranston) independently may substantiate the idea of survivorship of deteriorating palaeo-environmental conditions in immersed wood (Cranston & McKie 2006).

Monitoring the health of aquatic ecosystems with aquatic invertebrates can reveal disturbances by changes in both presence and abundances of taxa. Collecting techniques are standardized to particular substrates e.g. riffles, in which the biota of immersed stones can be washed into a downstream net. In South Africa, the Scoring System (SASS) developed from Chutter’s (1994, 1998) work is a prominent example of the use of macroinvertebrates in regional water quality assessment. Although the gain in our understanding of aquatic biodiversity from such monitoring programmes has been invaluable, gaps remain concerning unsampled or under-sampled in-stream microhabitats. Foremost amongst these is immersed wood, which is a refractory and often patchily distributed resource, especially in southern African streams, many of which have modest riparian cover. Woody substrates in southern African streams do support aquatic invertebrates, amongst which are *Elpiscladius* and another chironomid genus, namely *Stenochironomus* Kieffer. The globally distributed chironomine subgenus *Stenochironomus s. str.* is the most diverse wood-miner: its sister-subgenus *Petalopholeus* Borkent mines in submerged leaves. Six of the 10 described Afrotropical species are recorded from South Africa (Freeman 1957). The immature stages of the *Stenochironomus* species that shares habitat with *Elpiscladius* conform with, but cannot be distinguished from the generic diagnoses of, larvae and pupae (e.g. Borkent 1984; Pinder & Reiss 1983, 1986). However, the adult differs from any described species in the all-dark thorax, with an evenly ‘smoky’ wing and lacking pale areas on the legs, and with a mostly dark female abdomen with paler transverse vittae and pleurae (the teneral/pharate male colour cannot be determined). Pupal exuvial evidence for *Stenochironomus* suggests a wider distribution in southern African streams compared to *Elpiscladius*, which seems to be very restricted geographically to the Hottentots Holland area. The maximum distance between sites for *Elpiscladius* is 40 km in a straight line and exuvial sampling on a much wider scale has revealed no wider distribution.
Xylophagous taxa are never common in aquatic immersed wood in southern Africa: *Elpiscladius* and *Stenochironomus* represent rare taxa associated with clean streams with intact natural riparian vegetation. Neither is found in altered systems, including those in which no riparian vegetation remains to support xylophilous species. As such, these previously unrecognized elements of southern African biodiversity essentially act as habitat indicators and reflect survival from deeper history of an ancient lineage. Such elements ought to be protected and their conservation will be enhanced by activities such as those of Working for Water (WFW 2007) whose removal of aliens and enhanced native riparian development can only help in their conservation.

**REFERENCES**


ACKNOWLEDGEMENTS

I thank B. Carolus, horticulturist with South African National Biodiversity Institute, for permission to collect aquatic insects in Harold Porter National Botanic Garden; CapeNature for permit AAA004-0071-0035 to hunt with a prohibited hunting method of wild animals for Chironomidae in Jonkershoek Nature Reserve, although I used only drift and kick netting; M. Samways for logistical support from the University of Stellenbosch; the many staff and postgraduate students there, especially E. Brendenhand, for discussions on aquatic biology and monitoring with reference to Jonkershoek and, as ever, A. Harrison provided helpful comments on several issues raised in this study.

Accepted 31 October 2007