# New larvae of Caeninae from Madagascar (Ephemeroptera: Caenidae)

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#### Abstract

Larvae of four different species of *Caenis* and one of *Callistinella* from Madagascar are described. Furthermore, a description of a new Malagasy genus and species, *Trichocaenis inexperta* **n. gen.**, **n. sp.**, is provided, based on larval characters. Characters of subimaginal genitalia of larvae of two species of *Caenis* allow a preliminary assignment to *C. spinosa* and *C. johannae*. The studied material of *Callistinella* is very similar to the *Caenis rutila*group.

K e y w o r d s : Madagascar, Caenid larvae, new genus, new species, Caenis, Trichocaenis, Callistinella.

#### Zusammenfassung

Vier verschiedene Larvenformen der Gattung *Caenis* sowie eine der Gattung *Callistinella* von Madagaskar werden beschrieben. Außerdem wird aufgrund von Larvenmerkmalen eine für die Wissenschaft neue madagassische Gattung und Art, *Trichocaenis inexperta* **n. gen.**, **n. sp.**, beschrieben. Merkmale der subimaginalen Genitalien von zwei Arten von *Caenis* ermöglichen eine vorläufige Zuordnung zu *C. spinosa* und *C. johannae*. Das studierte Material von *Callistinella* ist der *Caenis rutila*-Gruppe sehr ähnlich.

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# **1** Introduction

DEMOULIN (1973) was the first who mentioned a Caenidae larva from Madagascar, determined by him as Austrocaenis sp. MALZACHER (1995) described five species of the genus Caenis and one of the new genus Madecocercus from Madagascar, but all were based on imaginal material only. While the larvae of Madecocercus were further treated by McCafferty & Wang (1995), ELOUARD & SAR-TORI (2001) and MALZACHER & STANICZEK (2006), the larvae of the Malagasy Caenis remained unknown. When SUN & McCafferty (2002) described an additional genus. Callistina (because of homonymy renamed to Callistinella by SUN & McCAFFERTY 2004), based on larval material only, the authors conceded the possibility that the new larva could prove to be the unknown aquatic stage of one of the Malagasy Caenis species already described. This becomes very likely by the results of this investigation.

Four different larval forms of *Caenis* and one of *Callistinella* are described in the present paper, and are – in part – preliminary assigned to taxa already known from described males (*Caenis spinosa* Malzacher, 1995 and *C*.

*johannae* Malzacher, 1995). Furthermore a new genus and species, *Trichocaenis inexperta* n. gen., n. sp., is described, based on larval characters. However, a correct assignment of the treated larval forms to known species can only be confirmed when males are reared from larvae.

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#### 2 Material and methods

The extensive material of Malagasy Caenidae adults collected in the 1990s by the ORSTOM-team of Dr. JEAN-MARC ELOUARD, which I received from Dr. MICHEL SARTORI (Lausanne) also contained 35 samples with a total of 61 larvae stored in 75 % ethanol. Besides numerous larvae of *Caenis*, the material consists of two specimens of the new genus *Trichocaenis*, one specimen of *Callistinella*, two samples with *Clypeocaenis afrosetosa* Soldán, 1978 from Guinea and Côte d'Ivoire and one larva of *Barnardara demoori* from the Democratic Republic of Congo. Additionally two larvae of *Clypeocaenis oligosetosa* Soldán, 1983 (coll. SOLDÁN) were examined, as well as four samples with *Callistinella* larvae from the Montagne d'Ambre in the North of Madagascar, collected by REINHARD GERECKE and TOM GOLD-SCHMIDT, two specialists of water mites.

The studied material is hosted in the Museum of Zoology Lausanne, except of a few duplicate specimens of the ORSTOMmaterial and the two specimens of *Clypeocaenis oligosetosa* which are stored in my collection.

Specimens used for SEM were dehydrated through a stepwise immersion in ethanol and then dried by critical point drying. The mounted material was coated with a 20 nm Au/Pd layer, and examined with an ISI-SS40 scanning electron microscope at 10 kV, and digital photographs were subsequently aquired by using DISS 5 (point electronic).

#### **3** Systematic account

# 3.1 Genus Caenis Stephens, 1835

Larvae of the genus *Caenis* can be characterised and distinguished from all other genera of Caenidae by the following combination of characters:

Thorax not broadened. Head neither with ocellar tubercles nor with ridges and microscopic pits. Clypeus not protruding anteriorly. Maxillar and labial palps three-segmented. Basal segment of labial palp longer than broad. Fore tibia and fore tarsus without very long bristles. Femora distinctly longer than broad and distinctly broader than tibiae, without extended plates. Operculate gill ventrally with a very regular row of scale-shaped microtrichia reaching hind margin of the gill. A great number of filaments of gills III–V(VI) with 3–4(5) branches. Lateral spines of abdomen not bent dorsally. Posterior part of sternum IX with shagreen field dorsally (Fig. 2c). Abdominal terga VII–IX without mediolongitudinal ridge. Hind margin of sternum IX without a bi-pointed process with concave margin between the points.

# Caenis sp. spinosa-group (larvae) (Fig. 1)

#### Material

1 larva: P0047, Madagascar, Betsiboka, Andranolave, Andakana, 18.IV.1991. – 1 larva: P0299, Madagascar, Sahankazo, Antsahamaiky, Joffreville, 12°30'00"S, 49°24'00"E, 28.III.1995. – 4 larvae (1 ♂ larva with SI-genitalia): P0652, Madagascar, Namorona, Tsaratango, 21°16'20"S, 47°31'35"E, 13.XI.1996. – 1 larva: P0868, Madagascar, PK 13 route Anosibe an'ala, 19°03'00"S, 48°13'57"E, 11.IV.1999. – 1 larva (♂ larva with SIgenitalia): P1008, Madagascar, Lokoho, Manantenina, Marojejy camp 1, 14°24'43"S, 49°46'48"E, 25.XI.1999.

#### Description

Measurements:  $\bigcirc$  larva of last instar: Body length 3.7-4.0 mm; length of cerci 1.7 mm; length of

paracercus 2.0 mm. -  $\bigcirc$  larva of last instar: Body length 4.5–4.7 mm; length of cerci 3.0 mm; length of paracercus 3.5 mm.

Colouration of chitinous layers: Redishyellowish-brown. Genae, lateral margins of pronotum, operculate gills, and posterolateral parts of abdominal terga lightened, also a round spot anterior to the frontal ocellus, a median longitudinal dash on vertex, a round spot on each side of pronotum and 4–5 spots or dashes on each mesonotum half. Pale spots are also present on second abdominal tergum in front of the insertion of operculate gills. Antennae, legs and terminal filaments pale yellow.

E pider mal pigmentation: Weak and diffuse on pronotum, metanotum and abdominal terga.

Head: Genae slightly bulged. Proximal comb of epipharynx of medium length, consisting of stout, slightly curved, apically blunt setae, the proximal ones pinnate. Mandibles laterally with bristles distally shortened and broadened. Dorsal surface densely tuberculate. Aboral side of postmentum laterodorsally without setae. Third segment of labial palp very short, second segment about 2.7 times as long as third segment (measured along the longitudinal centre line).

T h o r a x : Sides of pronotum broadly flattened, anteriorly diverging and bowed, margins notched. Side of mesonotum in its anterior part with a more or less hooked process (Fig. 1k). Coxal processes semicircular or tongueshaped, with strong denticles (Fig. 11, 1m). Transverse row of bristles on fore femur dense and straight (Fig. 1i); bristles of the row short and very broad, apically not pinnate (Fig. 1j). Similar bristles on dorsal surface and hind margins of mid and hind femora. Fore tarsus with a row of 5-6long simple bristles. Mid tarsus with a row of 7-8 strong simple bristles, the two apical ones pinnate. Hind tarsus with an inner row of 9(-10) strong, simple bristles and an outer row of about 6 bipinnate bristles. Fore claws slightly curved (Fig. 1n). Mid and hind claws apically strongly bent, mid claws with about 8 denticles (Fig. 1o). Hind claws heterodont, with 3 basal denticles and a serrated band consisting of a great number of microdenticles apically (Fig. 1p).

A b d o m e n : Outline of abdomen as in Fig. 1a. Lateral bristles on abdominal segments posteriorly decreasing in length, becoming stout and spatulate, very short on segments VIII and IX (Fig. 1c, 1d). Posteromedian process of tergum II triangular and of medium length. Operculate gill with short bristles on lateral and medial margins, posterior part of lateral margin with short spatulate bristles contrasting strongly with the long and thin bristles of hind margin (Fig. 1f, 1g). Row of microtrichia and lateral margin of operculate gill posteriorly diverging. The row runs more medially as in most other *Caenis* species, not reaching hind margin and posteromedial corner of the gill



**Fig. 1.** *Caenis* sp. *spinosa*-group, larva. – **a**. Outline of abdomen, sterna IV–IX,  $\bigcirc$ . **a'**. Hind margin of sternum IX. **b**. Sternum IX,  $\bigcirc$ . **c**. Marginal setation of segment V. **d**. Marginal setation of segment VII. **e**. Marginal setation of segment IX. **f**. Operculate gill, general view. **g**. Bristles from hind margin of operculate gill, transition from short spatulate bristles to very long and thin ones. **h**. Microtrichia from ventral side of operculate gill. **i**. Fore femur with transverse row of bristles on dorsal side. **j**. Bristles from the transverse row on fore femur. **k**. Left half of pronotum and anterior part of mesonotum. **l**. Coxal process of mid leg. **m**. Coxal process of hind leg.

(Fig. 1f), microtrichia more or less circular (Fig. 1h). Hind margins of terga VIII–X with denticles, VII with strong bristles of different length. Hind margin of sternum IX with a deep indentation and short spatulate bristles (Fig. 1a, 1a', 1b). Shagreen field on the base of the indentation large and triangular. Dorsal surface of abdomen densely covered with small denticles.

# Remarks

Subimaginal male genitalia from an adult larva show forcipes with tufts of long and strong spines. These characters, in combination with a dilated base of antennal flagellum (not dilated in *Caenis rugosa*) and sternum IX without colouration of sclerites (coloured in *C. johannae* and *C. rugosa*) lead to the *C. spinosa*-group.

# Caenis sp. cf. spinosa (larvae) (Fig. 2)

#### Material

3 larvae: Madagascar, Betuboha, 8.XI.1991. – 4 larvae: P0205, Madagascar, Ambre 2, 4.IV.1994. – 1 larva: P0207, Madagascar, Sahankazo Ambany, Sakaramy, 5.IV.1994. – 2 larvae: P0307, Madagascar, Mangoky, Menamaty, Ranohira, 22.IV.1994. – 1 larva: P0336, Madagascar, Mandrare, Mananara, Ihazofotsy, 27.V.1994. – 1 larva: P0411, Madagascar, Mandrare, Antalimanga, Besomosoy, 24°05′45″S, 46°27′59″E, 25.IV.1995. – 5 larvae: P0416, Madagascar, Mandrare, Aff. Abetolo, Esira, 28.IV.1995. – 1 larva (♂ larva with SI-genitalia): P0417, Madagascar, Mandrare, Anatranatra, Esira, 24°17′37″S, 46°39′04″E, 28.IV.1995. – 1 larva: P0526, Madagascar, Manampanihy, Enosiari, 24°40′37″S, 46°49′19″E, 21.XI.1995. – 2 larvae: P0834, Madagascar, Antongombato, between Diégo et Namakia, 30.III.1999.

#### Description

M e a s u r e m e n t s :  $3^{\circ}$  larva of last instar: Body length 2.8–3.0 mm; length of cerci 2.0 mm; length of paracercus 2.2 mm. –  $2^{\circ}$  larva of last instar: Body length 4.0–4.6 mm; length of cerci 2.6 mm; length of paracercus 2.8 mm.

Colouration of chitinous layers: Yellowish-brown.

E p i d e r m a l p i g m e n t a t i o n : In most specimens no pigmentation visible, sometimes with a very inconspicuous greyish band on vertex and some spots on pronotum.

H e a d : Genae not bulged. Proximal comb of epipharynx short, consisting of long, stout, apically curved setae. Mandibles with elongated field of long bristles laterally. Dorsal surface without denticles. Aboral side of postmentum laterodorsally with numerous stout setae. Third segment of labial palp very short, second segment about 3 times as long as third segment (along the centre line).

Thorax: Sides of pronotum slightly convex, diverging posteriorly, with broadly rounded fore corners. Few strong bristles between coxal cavities of mid and hind legs. Coxal processes semi-elliptical or semicircular, margin with small denticles (Fig. 2k). Fore femur dorsally with a dense transverse row of sturdy, pseudobifid, not or very shortly pinnate, apically rounded bristles (Fig. 2j). Mid and hind femur dorsally with similar bristles of different length. Hind margins of all femora with long and pointed bristles. Fore tarsus ventrally with inner row of about 7 slightly unipinnate bristles. Mid tarsus with inner row of about 8 and outer row of 4 unipinnate bristles. Hind tarsus with inner row of 8-9 and outer row of 7-8 unipinnate bristles. Pinnae of bristles always directed medially. Fore claws with slender tip, basally with few very small denticles (Fig. 2f). Mid claws more compact and also with few basal denticles (Fig. 2g). Denticles of fore and mid claws sometimes hardly visible or lacking. Hind claws with a little stronger curved tip, few basal denticles and a row of micro-denticles (heterodont) (Fig. 2h).

A b d o m e n : Outline of abdomen as in Fig. 2a. Lateral margin densely covered with long bristles which are shorter on segments VII-IX (Fig. 2c, 2e). Posteromedian process of tergum II very flat and short triangular. Operculate gill dorsally with only few short bristles; ventral row of microtrichia originating near the gill base running parallel to the lateral margin and very close to the hind margin (pinnation of microtrichia sometimes overlapping the hind margin), ending close to the inner corner of the gill (Fig. 2d). Microtrichia semicircular, slightly pinnate. Hind margins of terga VII and VIII with long bristles. Hind margins of terga IX and X without bristles, but with denticles. Hind margin of sternum IX with a deep indentation and very thin and short bristles (Fig. 2a-c). Dorsal side of sternum IX caudally with a circular field of shagreen consisting of regularly arranged denticles (Fig. 2c).

Subimaginal genitalia from last instar larva (Fig. 2c): Forceps relatively long and slightly bowed, with tuft of short spines. Penis broad. Styliger sclerite with short apophyses. Sternum IX with long posterolateral spines and sublateral pigment dashes.

# Remarks

Subimaginal characters from an adult male larva (Esira, 28.IV.1995) are: Forceps with tuft of short spines (Fig. 2c); antenna with dilated base of flagellum; prosternal triangle narrow, pointed and basally pigmented (Fig. 2i); two transverse pigment-bands on vertex; pronotum at hind margin with two submedian spots; abdominal terga with sublateral marks. All these characters lead to *Caenis spinosa*.



**Fig. 2.** *Caenis* sp. cf. *spinosa*, larva. – **a**. Outline of abdomen, sterna VI–IX,  $\bigcirc$ . **b**. Sternum IX,  $\bigcirc$ , another shape. **c**. Sternum IX,  $\bigcirc$ , with subimaginal genitalia. **d**. Operculate gill, general view. **e**. Marginal setation of segment VII. **f**. Claw of fore leg. **g**. Claw of mid leg. **h**. Claw of hind leg. **i**. Subimaginal prosternum from an adult  $\bigcirc$  larva. **j**. Bristles from the transverse row on fore femur. **k**. Coxal processes of mid leg (left) and hind leg (right).

# Caenis sp. cf. johannae (larvae) (Fig. 3a–h)

# Material

10 larvae (1 3 larva with SI-genitalia): P0602, Madagascar, Maharivo, Befasy, 20°34'18"S, 44°20'45"E, 27.V.1996.

#### Description

M e a s u r e m e n t s :  $\eth$  larva of last instar: Body length 2.8 mm. – [ $\heartsuit$  larva of last instar not available].

Colouration of chitinous layers: Yellowish-brown.

E p i d e r m a l p i g m e n t a t i o n : Except a band between the lateral ocelli, the region around the frontal ocellus, a spot at the base of the wing buds, and dashes on the fore corners of pro- and metanotum, no further pigmentation can be observed (in contrary to the subimago).

Head: Genae slightly bulged. Proximal comb of epipharynx of medium length, consisting of strong, slightly curved, apically denticulated setae, the proximal ones pinnate. Mandibles laterally without long bristles, densely tuberculate on dorsal surface. Aboral side of postmentum laterodorsally without strong setae. Third segment of labial palp short, second segment about twice as long as third segment (along the centre line).

Thorax: Sides of pronotum more or less parallel, slightly curved. Few fine hairs between coxal cavities of fore and mid legs and between mid and hind legs. Coxal processes narrow sickle-shaped, margins smooth or with few very small denticles, 2-3 long setae (Fig. 3c, 3d). Fore femur dorsally with an irregular transverse row or band of long and sturdy, pseudobifid bristles which are not or very shortly pinnate (Fig. 3h). Mid and hind femur dorsally with similar bristles of same length or longer. The same type of bristles on hind margins of all femora. Tibiae with sturdy pointed bristles. Fore tarsus ventrally with inner row of 5-9 simple bristles. Mid tarsus with inner row of 6-8 simple bristles and outer row of 2-5 unipinnate bristles. Hind tarsus with inner row of 6-10 simple bristles and outer row of 4-7 unipinnate bristles. Pinnae of bristles always directed medially. All claws long and slender, with 4-6 basal denticles (Fig. 3e-g). Hind claws additionally with a row of micro-denticles (heterodont) (Fig. 3g).

A b d o m e n : Posterolateral processes short. Lateral margin densely covered with bristles of medium length, on segments VIII–IX shorter. Posteromedian process of tergum II very flat and short triangular, often not visible in dorsal view. Operculate gill dorsally with numerous long bristles (Fig. 3b). Ventral row of microtrichia originating near the gill base. Row of microtrichia and lateral margin of operculate gill parallel. The row ends near the inner hind corner of the gill, running very close to the hind margin (pinnation of microtrichia sometimes overlapping the hind margin) (Fig. 3b). Microtrichia short, with

straight lateral spines, trapezoid or semicircular. Hind margins of terga VII and VIII with long bristles. Hind margins of terga IX and X without bristles, but with denticles. Hind margin of sternum IX with very thin bristles and a deep indentation (Fig. 3a). Dorsal side of sternum IX at the base of the indentation with a circular field of shagreen consisting of regularly arranged denticles.

Subimaginal genitalia from last instar larva (Fig. 3a): Forceps short and straight, with tuft of long spines. Penis relatively narrow. Strong pigment dashes, marking lateral and basolateral sclerites, are situated relatively close to styliger sclerite. Lateral parts of sternum IX strongly pigmented, with long sublateral pigment dashes.

#### Remarks

Subimaginal characters from an adult male larva are: Forceps relatively short, with tuft of long spines (Fig. 3a); antenna with dilated base of flagellum; the strong and differentiated pigmentation of epidermis nearly identical with that of males of the type series (MALZACHER 1995), particularly the strong brown colour of head, abdomen and lateral parts of sternum IX (the latter marks the position of the lateral sclerite) (Fig. 3a). All these characters lead to *Caenis johannae*.

# Caenis sp. A (larvae) (Figs. 3i–k, 7)

#### Material

1 larva: P0177, Madagascar, Mangoky, Zomandao, Andringitra nord camp 6, 28.XI.1993. – 1 larva: P0207, Madagascar, Sahankazo Ambany, Sakaramy, 12°26'50"S, 49°17'21"E, 5.IV.1994. – 1 larva: P0226, Madagascar, Namorona, Namorona, route Ifanadiana–Tolongoina, 22.IV.1994. – 1 larva: P0339, Madagascar, Mandrare, Mananara, Hazofotsy, 2.VI.1994. – 1 larva: P0476, Madagascar, Tsiribihina, Aff. de Sakay, Sakay Babetville, 18°59'57"S, 46°31'47"E, 10.X.1995. – 1 larva: P0495, Madagascar, Betsiboka, Mananara, Mangamila, 18°34'40"S, 47°51'42"E, 30.X.1995. – 1 larva: P0595, Madagascar, Tsiribihina, Sahaomby, Manatoloza, 19°25'50"S, 46°57'53"E, 21.V.1996. – 1 larva: P0725, Madagascar Rianila, Ambodiaviavy, Sandrakatrana, 18°57'31"S, 48°39'45"E, 30.IV.1998. – 3 larvae: P0727, Madagascar, Sandrakatrana, Ambodiaviavy, 18°57'45"S, 48°40'16"E, 30.IV.1998.

#### Description

M e a s u r e m e n t s : [ $\eth$  larva of last instar not available].  $- \Im$  larva of last instar: Body length 5.5–5.7 mm; length of cerci 2.8 mm; length of paracercus 3.0 mm.

Colouration of chitinous layers: Yellowish-brown to yellowish-white, no lightenings visible.

Epidermal pigmentation: Very sparsely and diffuse on vertex and operculate gills, often totally lack-



**Fig. 3.** *Caenis* sp. cf. *johannae*, larva (a–h). – a. Sternum IX,  $\mathcal{J}$ , with subimaginal genitalia. b. Operculate gill, general view. c. Coxal process of hind leg. d. Coxal process of mid leg. e. Claw of fore leg. f. Claw of mid leg. g. Claw of hind leg. h. Bristles from the transverse row on fore femur. – *Caenis* sp. A, larva (i–k). – i. Coxal process of hind leg. j. Outline of abdominal sterna VIII and IX. k. Operculate gill, general view.

ing. Only one specimen shows a stronger blackish-brown pigmentation: a band between the lateral ocelli, numerous spots and dashes on pro- and mesonotum, femora with subapical spots, and sublateral marks on abdominal terga. Intensity of pigmentation seems to be highly variable (see Remarks below).

Head: Genae slightly bulged. Proximal comb of epipharynx consisting of strong, slightly curved, apically denticulated setae. Mandibles with elongated field of long bristles laterally. Dorsal surface without denticles. Second segment of labial palp about twice as long as third segment (along the centre line).

Thorax: Sides of pronotum nearly parallel, straight or slightly bowed. Coxal processes narrow band-shaped, not bulging out from the outline of coxa (Fig. 3i). Transverse row of bristles on fore femur dense and straight, consisting of 8-14 long, blunt and pseudobifid bristles which are apically not or only slightly pinnate. Dorsal surface and margins of mid and hind femora with long, slender and pointed bristles which are sometimes very long and hair-like. Fore tarsus with an irregular row of 15-18 very long, simple bristles. Mid tarsus with an inner row of about 13 and an outer row of about 10 long and slender, more or less pinnate bristles. Hind tarsus with an inner row of about 15 and an outer row of about 13 bristles of the same shape. Fore claws very long and slender, slightly curved. Mid and hind claws also long and slender, apically more or less bent, all claws with 6–8 denticles. Hind claws without row of microdenticles (homodont).

A b d o m e n : Lateral bristles on abdominal segments long or very long. Posteromedian process of tergum II very short, band-shaped, nearly not visible in dorsal view. Operculate gill with long to very long and hair-like marginal bristles, same bristles on the dorsal side (Fig. 3k). Row of microtrichia and lateral margin of operculate gill posteriorly diverging. The row ends at the inner posterior corner of the gill cover (Fig. 3k), microtrichia more or less circular or elliptical. Hind margins of terga VII and VIII with long hair-like bristles, IX with bristles and median denticles, X with denticles. Sternum IX broader than in other species; hind margin with a broad v-shaped incision and long pointed bristles. Shagreen field on the base of the incision elliptical or elongated triangular (Fig. 3j).

Structure of surface: Epicuticula extensively smooth. Dorsal and ventral body surface covered with hair-like bristles of different length. Larva looks mouldy in lower magnification.

#### Remarks

Intensity of setation, especially of the operculate gills, and intensity of pigmentation can be variable as well as other characters: mid and hind claws are sometimes strongly bent apically and the outer rows of bristles on mid and hind tarsi can be reduced to few bristles. So it seems to be possible that there are two or more *Caenis* species with body setation.

In case the herein treated species turns out to be undescribed, "trichocaenoides" would be a suitable species name for it, referring to the numerous long bristles on the surface of the body (similar as in the new genus Trichocaenis which is described below). It can, however, not be ruled out that the larvae described above might belong to one or more already known species.

# 3.2 Genus Trichocaenis n. gen. (larvae)

Type species: Trichocaenis inexperta n. sp.

# Etymology

The first part of the genus name refers to the great number of hair-like bristles covering the body surface.

# Diagnosis

Larvae of this genus can be characterised and distinguished from all other genera of Caenidae by the following combination of characters:

Body surface covered with hair-like bristles. Head with ridges (Fig. 8), but neither with ocellar tubercles nor with microscopic pits. Clypeus not protruding anteriorly. Maxillar and labial palps three-segmented. Basal segment of labial palp nearly as broad as long. Thorax broadened (Fig. 4). Sides of pronotum converging anteriorly. Fore tibia and fore tarsus without very long bristles. Femora distinctly longer than broad and distinctly broader than tibiae, without extended plates. Operculate gill ventrally with lateral band of spines, clusters of spines and preliminary stages of scales (Fig. 5b, 5c). Filaments of gills III-VI simple or with 2 branches only (one or two apical filaments with 3 or 4 branches). Lateral spines of abdomen not bent dorsally. Abdominal terga VII-IX without mediolongitudinal ridge. Shagreen field on dorsal side of posterior part of sternum IX lacking. Hind margin of sternum IX without a bi-pointed process with concave margin between the points.

*Trichocaenis* n. gen. has ridges on head and the broad thorax with converging sides of pronotum (Figs. 4, 8). The new genus can be distinguished from closer related genera as follows: Long hair-like bristles are lacking on dorsal and ventral surface of *Clypeocaenis*, *Barnardara* and *Callistinella*. All *Caenis* species have the ventral side of the operculate gill with a very regularly arranged row or band of very differentiated scale-shaped microtrichia (such a row is absent in *Trichocaenis*).

# Trichocaenis inexperta **n. sp.** (larvae) (Figs. 4, 5, 8, 12)

# Material

H o l o t y p e (larva): P0636, Madagascar, Namorona, Tsaratango, 21°16'33"S, 47°31'50"E, 8.XI.1996.

P a r a t y p e : 1 larva: without data.

Holotype and paratype are stored in the Museum of Zoology Lausanne.

#### Etymology

The species name *inexperta* (Latin: inexpertus = still unknown) refers to the still unknown imaginal stages.

#### Description

Measurements:  $\bigcirc$  larva subadult: Body length 4.8 mm.

Colouration of chitinous layers: Very pale, brownish-yellow to white. Diffuse lightenings medially to wing buds.

E pider mal pigmentation: Metanotum and abdominal terga I–VIII with brown pigments, IX and X as well as lateral marginal region pale.

H e a d: Genae flat. Proximal comb of epipharynx short, consisting of strong slightly curved and apically denticulated setae, a field of long and thin setae laterad to the comb overlapping it partially. Right distal field of thick setae only slightly smaller than the left one. Mandibles laterally with numerous hair-like bristles. Maxillar palp relatively short, not overlapping the galea-lacinia. First segment of labial palp nearly as broad as long, third segment relatively long, second segment about 1.4 times as long as the third segment (along the centre line) (Fig. 5d). Ventral side of labium, particularly postmentum, praementum, and first and second segment of labial palp with numerous long hair-like bristles.

T h o r a x : Sides of pronotum nearly without flattened border, straight and smooth, anteriorly converging (Fig. 4). Coxal processes narrow band-shaped, not bulging out from the outline of coxa, not denticulated (Fig. 5h, 5i). Fore femur without transverse row of bristles, but with numerous hair-like bristles of different length all over the surface, as in mid and hind femora (Fig. 4). All tarsi with only one row of 10–15 simple bristles. Claws slender and slightly curved, with 6–8 denticles. Hind claws homodont (Fig. 5e–g).

A b d o m e n : Posterolateral spines on abdominal segments rather short with narrow bases (Fig. 5a). Posteromedian process of tergum II lacking. Operculate gill dorsally with numerous long bristles along the y-shaped ridge; three long bristles in a sublateral row (Figs. 5b). Ventral side with a sublateral band of spines, clusters of spines and preliminary stages of scales (Figs. 5b, 12). Gills III–VI relatively small, delicate and translucent, marginal filaments simple or bifurcate, only 1–2 apical filaments with more than two branches. Hind margins of terga VII and VIII with bristles of different length, IX with bristles and denticles, X with denticles only. Hind margin of sternum IX rounded with long sturdy bristles bent together medially. No shagreen field on dorsal side. Caudal filaments with short and strong spines (Fig. 5j, 5k).

Structure of surface: Epicuticula extensively smooth. Body surface covered with hair-like bristles of different length, evenly distributed on ventral side and concentrated in groups dorsally (Fig. 4). Operculate gill dorsally with numerous long bristles. Larva looks mouldy in lower magnification.

# **Differential Diagnosis**

See diagnosis of Trichocaenis n. gen.

# 3.3 Genus Callistinella Sun & McCafferty, 2004

Callistina Sun & McCafferty, 2002 [homonym of Callistina Jukes-Browne, 1908 (Mollusca)]

Larvae of this genus can be characterised and distinguished from all other genera of Caenidae by the following combination of characters:

Long bristles on body surface extensively lacking. Thorax broadened. Head without ocellar tubercles. Vertex, frons and thoracic nota with ridges and pits with branched setae (Figs. 9, 10). Clypeus not protruding anteriorly. Maxillar and labial palps three-segmented. Basal segment of labial palp as long as broad, square or circular. Sides of pronotum converging anteriorly. Prosternum anteriorly with rounded process covered with short and stout bristles (Fig. 6g). Fore tibia and fore tarsus without very long bristles. Femora distinctly longer than broad and distinctly broader than tibiae, without extended plates. Operculate gill ventrally with sublateral band of spines, clusters of spines and preliminary stages of scales not reaching the hind corner (Fig. 6e). Filaments of gills III-VI simple or with 2 branches only (one or two apical filaments with 3 or 4 branches). Lateral spines of abdomen not bent dorsally. Abdominal terga VII-IX with mediolongitudinal ridge anteriorly more or less protruding (Fig. 6a). Shagreen field on dorsal side of posterior part of sternum IX lacking. Hind margin of sternum IX without a bi-pointed process with concave margin between the points.

# Callistinella sp. (larvae) (Figs. 6, 9–11)

# Material

l larva (d' larva with SI-genitalia): Madagascar, riv. Zomandao sur le plateau (Mangoky-bassin), 29.XI.1993. – 1 larva: 159, Madagascar, R. Antomboka, Joffreville (M. D'Ambre, An-



Fig. 4. Trichocaenis inexperta n. sp., larva. Habitus; right half with setae on body surface and outline, left half with setae on legs and second gill. – Scale: 1 mm.



**Fig. 5.** *Trichocaenis inexperta* n. sp., larva. – **a**. Outline of abdomen, sterna III–IX,  $\mathcal{Q}$ . **b**. Operculate gill, general view. **c**. Sector of sublateral band of microtrichia on ventral side of operculate gill. **d**. Labial palp. **e**. Claw of fore leg. **f**. Claw of mid leg. **g**. Claw of hind leg. **h**. Coxal process of mid leg. **i**. Coxal process of hind leg. **j**. Basal sector of terminal filament. **k**. Medial sector of terminal filament.



**Fig. 6.** *Callistinella* sp., larva. – **a**. Abdomen, lateral view of dorsal part. **b**. Sternum IX,  $\mathcal{J}$ , with subimaginal genitalia. **c**. Claw of fore leg. **d**. Claw of hind leg. **e**. Operculate gill, general view. **f**. Operculate gill, epicuticular structure of dorsal surface. **g**. Anterior process of sternum. **h**. Bristles on anterior process of sternum.

tsiranana), 950 m, 19.XI.2001. – 2 larvae: 165, Madagascar, R. Antomboka, Joffreville (M. D'Ambre, Antsiranana), 850 m, 21.XI.2001. – 1 larva: Same data as before, but 166.

#### Description

Measurements:  $\Im$  larva of last instar: Body length 3.0–3.7 mm; length of cerci 1.6 mm; length of paracercus 2.3 mm. – [ $\Im$  larva of last instar not available].

Colouration of chitinous layers: Brownish-yellow to orange.

E p i d e r m a l p i g m e n t a t i o n : Central parts of clypeus and frons, and posterior margin of vertex brown, rest of vertex light, sometimes with few brown dashes. Thoracic nota and pleura dark-brown to blackish-brown, sometimes with a pattern of a little lighter spots and areas, abdominal terga I, II, and VII–X brown to dark-brown, terga III–VI with light-brown pigments visible through the pale and translucent gills. Ventral side more or less lightbrown, abdominal sterna III–VI light. Femora and tibiae sometimes with longitudinal light-brown bands.

H e a d : Vertex and frons with cephalic ridges and elevations (Fig. 9). Genae bulged, semi-elliptical. Labrum with long bifurcate, pinnate bristles (Fig. 11). Proximal comb of epipharynx long, consisting of long, apically blunt setae, the proximal ones strongly curved. Mandibles without long bristles. Dorsal surface strongly tuberculate. Aboral side of postmentum laterodorsally with thin and short bristles. Third segment of labial palp nearly as long as the second one, slightly bent medially.

T h o r a x : Sides of pronotum nearly without flattened border, straight and strongly notched, anteriorly converging. Prosternum anteriorly with a rounded process covered with numerous short and stout, apically frayed or bifurcate bristles (Fig. 6g, 6h). Coxal processes semicircular or tongue-shaped, strongly tuberculate, with few thin and short bristles at the margin. Fore femur dorsally with an interrupted transverse row of short and sturdy pseudobifid bristles which are apically rounded but not pinnate. Mid and hind femur dorsally without bristles. Margins of all femora with short, stout pseudobifid bristles. Fore tarsus ventrally with inner row of 7–9 simple bristles, outer row with about 4 simple bristles which are sometimes slightly unipinnate. Mid tarsus with inner row of about 7 and outer row of about 5 unipinnate bristles. Hind tarsus with inner row of 7-9 and outer row of 5-7 unipinnate bristles. Pinnae of bristles always directed medially. All claws very similar, somewhat stout, with 5-7 sturdy denticles. Hind claws homodont (Fig. 6c, 6d).

A b d o m e n : Posterolateral processes short, posteriorly directed; process II conspicuously stronger and longer. Lateral margins with few short, spine-shaped bristles. Posteromedian process of tergum II triangular, pointed. Margin of operculate gill only with few short, sturdy, pseudobifid and apically rounded bristles; 3–5 sturdy pointed bristles at base of inner ridge (Fig. 6e). An irregular row or band of microtrichia with different preliminary stages of scales on the ventral side ending medially to the outer corner of the gill, not reaching the hind margin (Fig. 6e). Gills III–VI relatively small, delicate and translucent, marginal filaments simple or bifurcate, only one apical filament with 3 branches. The longest filament does not reach the length of the gill plate. Gill VI only with simple filaments. Hind margins of all visible terga strongly denticulated, without bristles. Abdominal terga VII–IX with mediolongitudinal ridge anteriorly more or less protruding (Fig. 6a). Hind margin of sternum IX slightly indented (Fig. 6b). No shagreen field on dorsal side.

Structure of surface: Head and thoracic nota with ridges and pits with branching setae, but not as conspicuous as in *Callistinella panda* (Figs. 9, 10). Other parts of body surface, femora, tibiae and operculate gills densely covered with tubercles or denticles, sometimes forming overlapping, strongly pointed epicuticular scales (Fig. 6f). Long bristles or setae lacking.

Subimaginal genitalia from last instar larva (Fig. 6b): Forceps relatively short, without spines, basally narrowed. Penis with strongly marked ventral fold and rounded lobes. Sternum IX with very long and slender posterolateral spines.

#### Remarks

Callistinella sp. is very similar to Callistinella panda (Sun & McCafferty, 2002) but there are some differences. The most conspicuous ones concern the colouring: The clear contrast between light parts (head and operculate gill area) and blackish-brown parts (rest of the body) in Callistinella panda is slightly diminished in the herein described species. Its head is partly coloured (see above) and terga III–VI are more or less with brownish pigments shining through operculate gills, causing a pale brownish colour, whereas in Callistinella panda these parts are yellowish-white to cream-coloured. Ridges on head and thorax are comparatively weaker and pits not as deep and regular as in Callistinella panda, the enclosed setae more irregular and with very thin branches (only visible in SEMs). Circum-antennal depressions do not reach the lateral ocelli, i. e. parts between ocelli and antennal bases are elevated (they are deeply depressed in Callistinella panda). Dorsal side of labrum densely covered with bifurcate and strongly pinnate bristles (labrum of Callistinella panda dorsally only with sparce setae) (compare Figs. 9-11 with figs. 3, 9 and 10 in SUN & McCAFFERTY 2002). Further remarks see chapter Discussion.



Figs. 7–8. Caeninae spp., larvae, head, dorsal view. – 7. Caenis sp. A. 8. Trichocaenis inexperta n. sp.



Figs. 9–10. *Callistinella* sp., larva. – 9. Head, dorsal view. 10. Vertex, epicuticula with pits and branching setae.



**Figs. 11–12.** Caeninae spp., larvae. – **11**. *Callistinella* sp., labrum with bifurcate and pinnate bristles, dorsal side. **12.** *Trichocaenis inexperta* n. sp., spines, clusters of spines and preliminary stages of scales on ventral side of operculate gill.

#### 4 Preliminary key to Caeninae larvae of Madagascar

- Pro- and mesonotum without longitudinal ridges. Abdominal terga VII–IX without mediolongitudinal ridge. Prosternum without a conspicuous process. Lateral margin of abdomen with longer bristles.
- Long hair-like bristles on dorsal and ventral body surface lacking. Hind claw heterodont, with 2–5 basal denticles, and a band of microdenticles more or less fused together (Figs. 1p, 2h, 3g).
- **3** Lateral margins of pronotum converging anteriorly. Longitudinal band or cluster of long bristles on dorsal side of fore femur, no transverse row (Fig. 4). Microtrichia on ventral side of operculate gill forming a sublateral band with spines, clusters of spines and primitive scales (Figs. 5b, 5c, 12). ... *Trichocaenis* **n. gen.**
- Lateral margins of pronotum nearly parallel, straight or slightly bowed. Transverse row of 10–14 bristles on dorsal side of fore femur. A row of well-developed scale-shaped microtrichia on ventral side of operculate gill.

- Dorsal surface of operculate gill with long or very long bristles (Fig. 3b). Coxal processes narrow sickle-shaped (Fig. 3c, 3d). Mid and hind claws very elongated and slender; 4–5 basal denticles (Fig. 3f, 3g). . . . . . *Caenis* sp. cf. *johannae*

#### 5 Discussion

The assignment of the herein described larvae to *Caenis spinosa* and *C. johannae* is likely because of the subimaginal genitalia, but at present it cannot be proved with absolute certainty.

Four genera of the subfamily Caeninae from Africa and Madagascar have been described from striking larval characters, without knowledge of the adults: Clypeocaenis Soldán, 1978, Barnardara Provonsha & McCafferty, 1995, Callistinella Sun & McCafferty, 2004 and the herein newly described Trichocaenis. The latter three genera are known from the Afrotropical region only, whereas *Clypeocaenis* was described first from Iran and India. Adults and a new species from Africa, Clypeocaenis afrosetosa, were described later by SOLDÁN (1983). SOLDÁN gives a couple of imaginal characters for separating Clypeocaenis, but most of them are within the variability of Caenis. The only character that does not occur in species of *Caenis*, the protrusion of the clypeus, is inconspicuous and often hardly visible. So in *Clypeocaenis* the adults are actually known but not distinguishable from adults of Caenis.

The subimaginal male genitalia visible through the cuticutle of last instar larvae of *Callistinella* are very similar to the genitalia of *Caenis rutila* Malzacher, 1995 (compare Fig. 6b with fig. 6a of MALZACHER 1995). Therefore there is a high probability that the larvae of *Callistinella* actually represent species of the *Caenis rutila*-group. However, until all doubts are dispelled by rearing adults from larvae, *Caenis rutila* should not be renamed to *Callistinella rutila*.

It also cannot be completely ruled out that the larvae of *Trichocaenis* n. gen. might belong to an already known species of *Caenis* (e. g. *C. rugosa* Malzacher, 1995), but this is not very likely.

As already mentioned above in this chapter, *Trichocaenis* belongs to the Caeninae genera which show striking apomorphic larval characters (in *Trichocaenis*: ridges on head and the broad thorax with converging sides of pronotum).

Recently the genera *Clypeocaenis*, *Barnardara*, *Callistinella* and *Trichocaenis* (the latter described herein) were united in the *Clypeocaenis*-group, based on the shape and arrangement of the microtrichia on the ventral side of the operculate gills (MALZACHER in press). An assessment of the phylogenetic position of this group, including all differential diagnostic characters, is in preparation.

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