

Morphological Studies on Five Trachelocercids from the Yellow Sea Coast of China, with a Description of *Tracheloraphis huangi* spec. nov. (Ciliophora, Karyorelictea)

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Summary. The morphology and infraciliature of five trachelocercid ciliates: *Tracheloraphis huangi* spec. nov., *T. colubis* (Kahl, 1933) comb. nov., *T. phoenicopterus* (Cohn, 1866) Dragesco, 1960, *T. oligostriata* (Raikov, 1962) Foissner and Dragesco, 1996 and *Trachelocerca incaudata* Kahl, 1933, isolated from the intertidal zone of a beach at Qingdao, China, were studied in live and protargol impregnated specimens. *Tracheloraphis huangi* spec. nov. was distinguished from its congeners mainly by its single nuclear group composed of 25–30 round macronuclei and 29–37 somatic kineties. The poorly known *T. colubis* is redescribed including for the first time information on its infraciliature. An improved diagnosis is also provided. In light of its infraciliature, *T. colubis* is transferred to *Tracheloraphis* from the genus *Trachelocerca*. Additional data on other three species is supplied based on the Qingdao populations.

Key words: Infraciliature, Karyorelictea, marine ciliates, *Trachelocerca*, *Tracheloraphis*.

INTRODUCTION

The Trachelocercidae Kent, 1881 is the largest family within the class Karyorelictea Corliss, 1974 and is very commonly represented in marine littoral sands

(Al-Rasheid 1996, 1997, 1998, 2001; Al-Rasheid and Foissner 1999; Foissner and Dragesco 1996a). Since the first species was described over 200 years ago, about 70 species have been reported (Carey 1992). Until the latter part of the last century species descriptions were based only on live observation (Sauerbrey 1928; Dragesco 1954a, b; Kahl 1933). During the 1960s and 1970s some fundamental studies were published that provided superficial information on the infraciliature mainly based on Feulgen stained specimens (Aga-

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maliev 1966; Borror 1963; Dragesco 1960, 1963, 1965; Dragesco and Raikov 1966; Kovaleva 1966; Kovaleva and Golemansky 1979; Raikov 1957, 1962, 1963; Raikov and Kovaleva 1968). Subsequently, the generic classification of trachelocercids became available. But this classification was still limited as it was based mainly on characters observed *in vivo* such as the absence or presence of the glabrous stripe (Dragesco 1960). Dragesco and Dragesco-Kernéis (1986) and Wilbert (1986) revealed for the first time using silver impregnation techniques the infraciliature of trachelocercids, especially the oral ciliary pattern. Subsequent studies addressed many of the confusing issues associated with trachelocercid systematics with some species being meticulously redescribed using modern techniques. Furthermore, new standards for genus- and species-level diagnoses and classification were established based mainly on the infraciliature, and in particular the oral ciliature (Foissner 1996, 1997a, 1997b, 1998; Foissner and Al-Rasheid 1999a, b; Foissner and Dragesco 1996a, b). However, to date the infraciliature has been described for only 20 out of the ~ 70 trachelocercid species. Thus, the remaining ~ 50 species are all questionable in terms of their validity and generic placement (Dragesco and Dragesco-Kernéis 1986; Foissner 1996, 1997b; Foissner and Al-Rasheid 1999a; Foissner and Dragesco 1996a, b; Xu *et al.* 2011).

In recent years several reports on the morphology and phylogeny of karyorelicteans have been published (Aleksperov *et al.* 2007, Andreoli *et al.* 2009, Gao *et al.* 2010, Mazei *et al.* 2009, Xu *et al.* 2011). In the present study, five trachelocercids isolated from the intertidal zone of a beach at Qingdao, China, were investigated both *in vivo* and following protargol impregnation.

MATERIALS AND METHODS

Ciliates were sampled from the intertidal zone of the No. 1 sandy beach at Qingdao (36°06'N; 120°34'E), China.

Tracheloraphis huangi spec. nov. was collected in November 2005, when the water temperature was 18°C and salinity was 29‰. *Tracheloraphis oligostriata* was collected in April 2010, with water temperature 11°C, salinity 20‰. *Tracheloraphis colubis* (Kahl, 1933) comb. nov., *T. phoenicopterus* (Cohn, 1866) Dragesco, 1960 and *Trachelocerca incaudata* (Kahl, 1933) Dragesco, 1960 were collected in May 2010, when the water temperature was about 20°C, and salinity was 30‰. Sampling methods were mainly according to Fan *et al.* (2010). Living cells were studied by bright field and differential interference microscopy (100 × to 1000 × magnifications). The infraciliature was revealed by the protargol impreg-

nation method (Wilbert 1975) using the following fixative: 10 ml saturated, aqueous mercuric chloride and 3 ml Bouin's solution, mixed just before use (Xu *et al.* 2011). Counts and measurements of stained specimens were performed at a magnification of 1000 ×. Drawings were made with the help of a camera lucida. Terminology is mainly according to Foissner (1996).

RESULTS AND DISCUSSION

Tracheloraphis huangi spec. nov. (Figs 1, 2; Table 1)

Diagnosis: Extended cells *in vivo* about 500–1200 × 60–100 µm in size. 29–37 somatic kineties. Glabrous stripe up to one third of cell width. Single nuclear group composed of ca. 25–30 macronuclei. Cortical granules minute and colorless.

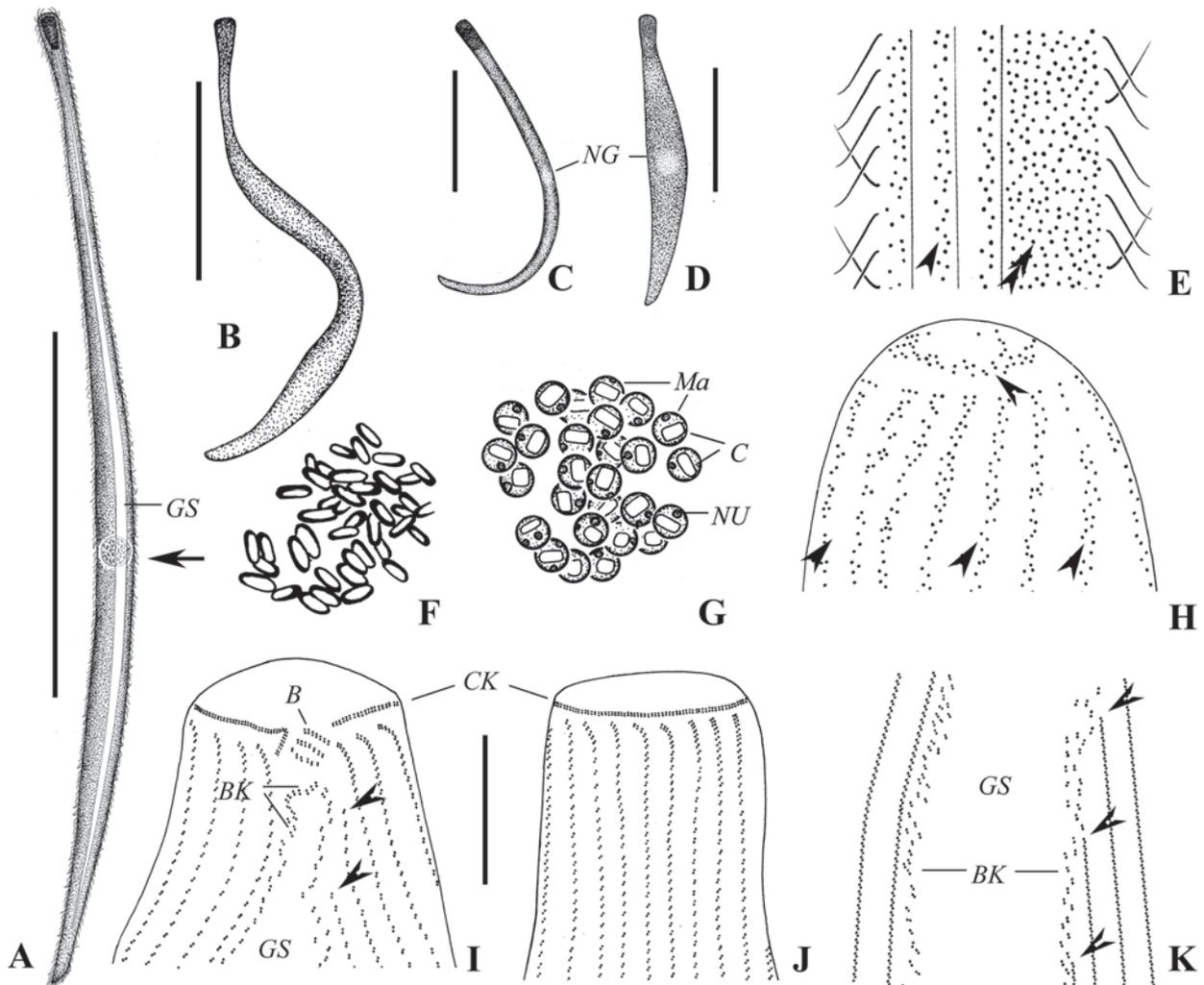
Type locality: The intertidal zone of the No. 1 sandy beach at Qingdao (36°03'N; 120°20'E), China. Yellow Sea coast.

Type specimens: A protargol slide containing the holotype specimen marked with ink circle is deposited in the Natural History Museum, London, UK, with registration number NHMUK 2011.4.27.1. One paratype slide with protargol-impregnated specimens is deposited in the Laboratory of Protozoology, OUC, China (No. WYG2005111701).

Dedication: The species is named in honor of Prof. Zongguo Huang, The Third Institute of Oceanography State Oceanic Administration, China, in recognition of his contributions in the field of marine biodiversity.

Description: Fully extended cells about 500–1200 × 60–100 µm *in vivo*, filiform in shape; flexible and contractile (Figs 1A–D, 2A–D); cell distinctly tripartite, with neck, tail and trunk regions (Figs 1A, 2A, B); trunk flattened ca. 2–3 : 1 (Fig. 2C); head conspicuous, claviform (Figs 1A, 2G). Endoplasm grayish and opaque due to multiple refractile (crystalline?) inclusions about 2 × 4 µm in size (Figs 1F, 2A–E). Nuclear apparatus (capsule) in centre of trunk, containing about 25–30 globular macronuclei which form tight cluster ca. 30–40 µm in diameter (Figs 1G, 2H, I, O). Micronuclei difficult to detect. Contractile vacuole not observed. Cortical granules globular, minute (about 0.5 µm in diameter), and colorless, forming narrow stripes between ciliary rows and densely distributed in glabrous stripe (Figs 1E, H, 2F). Locomotion by gliding, winding between sand grains and organic debris.

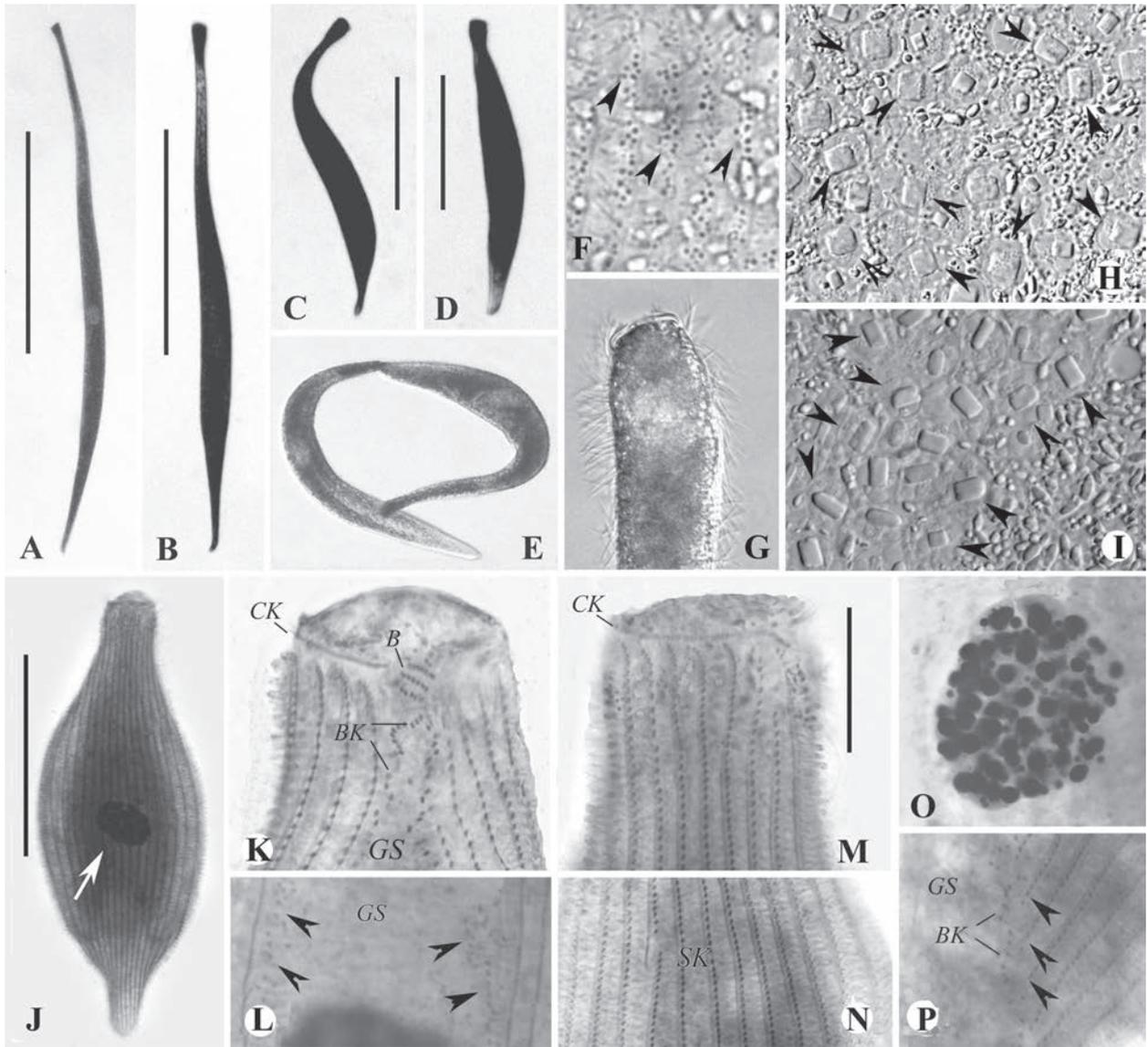
Cell surface densely ciliated but with an unciliated zone, the glabrous stripe, which extends the whole



Figs 1A–K. *Tracheloraphis huangi* spec. nov. from life (A–H) and after protargol impregnation (I–K). **A** – typical individual, noting the glabrous stripe occupying about one quarter of the body width; arrow shows the single nuclear group; **B**, **C** – shape variants; **D** – contracted cell; **E**, **H** – distribution of cortical granules between ciliary rows (arrowheads), around buccal edge (arrowheads) and in the glabrous stripe (double arrowheads) in the mid-body (**E**) and at the anterior end (**H**); **F** – ellipsoidal (crystalline?) inclusions; **G** – to show about thirty macronuclei inside the nuclear group, noting the (protein) crystals and the nucleoli; **I**, **J** – anterior end, indicating the circumoral kinety, brosse and glabrous stripe bordered by the bristle kinety; arrowheads in (**I**) show anterior secant system; **K** – mid-body region, marking the glabrous stripe, bristle kinety and anterior secant system (arrowheads). B – brosse, BK – bristle kinety, C – (protein) crystal, CK – circumoral kinety, GS – glabrous stripe, Ma – macronuclei, NG – nuclear group, NU – nucleoli. Scale bars: 400 μm (A–C), 200 μm (D), 30 μm (I, J).

body length in the midline of the left side; maximum width in trunk region approximately one third of body width (Fig. 1A). Entire infraciliature consisting of dikinetids. Somatic cilia about 10 μm long *in vivo* and arranged in longitudinal rows. Anterior ends of ciliary rows curved to right and composed of densely spaced dikinetids (Figs 1I, J, 2K, M). Anterior and posterior secant system formed on left side of glabrous stripe where some kineties abut to the bristle kinety (Figs 1I,

K, 2P). Ciliary rows neighboring the right branch of the bristle kinety unshortened anteriorly and thus extend along the glabrous stripe. Glabrous stripe bordered by the bristle kinety, and the kinetids of which are more widely spaced and irregularly arranged than those of the somatic ciliary rows (Figs 1I, K, 2K, L, P). Oral infraciliature consisting of a circumoral kinety, which is interrupted by the inserted brosse kineties (Figs 1I, J, 2K, M).



Figs 2A–P. Photomicrographs of *Tracheloraphis huangi* spec. nov. from life (A–I) and after protargol impregnation (J–P). **A, B** – extended individuals gliding; **C, D** – contracted individuals; **E** – a dividing cell; **F** – to show the distribution of cortical granules between the ciliary rows (arrowheads); **G** – anterior end of the body; **H, I** – nuclear group comprising about thirty macronuclei (arrowheads); **J** – infraciliature and nuclear group (arrow); **K, M** – anterior infraciliature of left and right sides, circumoral kinety, brosse, glabrous stripe and bristle kinety; **L, N** – mid-body of right and left sides respectively, showing the glabrous stripe bordered by the bristle kinety (arrowheads in L) and somatic kineties (N); **O** – nuclear group, noting the nuclei forming a tight cluster; **P** – left side of the glabrous stripe, marking the posterior secant system (arrowheads). B – brosse, BK – bristle kinety, CK – circumoral kinety, GS – glabrous stripe, SK – somatic kineties. Scale bars: 400 μ m (A–C), 150 μ m (D, J), 30 μ m (M).

Comparison: As most species of the family Trachelocercidae have only been studied based on live observation, their generic classifications remain questionable based on the new generic definitions (Foissner 1996, 1997b; Foissner and Al-Rasheid 1999a; Foissner and

Dragesco 1996a, b). Therefore, the comparison between *Tracheloraphis huangi* spec. nov. and its most closely related species should not be limited to those within the genus *Tracheloraphis*. Of the 70 trachelocercid species, 34 have their macronuclei arranged in a single group

Table 1. Morphometric data of *Tracheloraphis huangi* spec. nov. (first line), *Tracheloraphis colubis* (second line), *Tracheloraphis phoenicopterus* (third line), *Tracheloraphis oligostriata* (forth line) and *Trachelocerca incaudata* (fifth line). All data based on protargol-impregnated specimens. CV – coefficient of variation in %, n – number of specimens investigated, SD – standard deviation of the mean.

Characters	Minimum	Maximum	Mean	SD	CV	n
Body length in μm	206	360	286.0	45.0	15.7	15
	148	302	210.4	51.2	24.4	20
	230	512	328.8	86.9	26.4	25
	108	385	174.6	53.8	30.8	24
	120	270	190.3	45.3	23.8	20
Body width in μm	77	123	98.4	14.6	14.9	15
	42	59	49.9	5.2	10.5	20
	19	60	40.8	13.1	32.0	25
	27	51	38.1	5.2	13.6	24
	26	60	36.4	7.5	20.6	20
Glabrous stripe width in μm	14	34	24.9	6.8	27.4	15
	–	–	–	–	–	–
	11	37	20.8	6.7	32.0	25
	–	–	–	–	–	–
	7	16	11.1	2.9	26.7	20
Somatic kineties, number	29	37	33.8	2.3	6.7	15
	23	31	27.9	1.7	5.9	20
	23	32	25.7	1.9	7.3	25
	6	6	6.0	0	0	24
	23	26	24.3	1.0	4.2	20
Somatic kineties on head, number	19	25	21.6	2.0	9.0	15
	12	17	13.9	1.4	10.1	20
	12	16	14.2	1.1	8.1	15
	6	6	6.0	0	0	24
	13	15	13.9	0.8	5.9	20

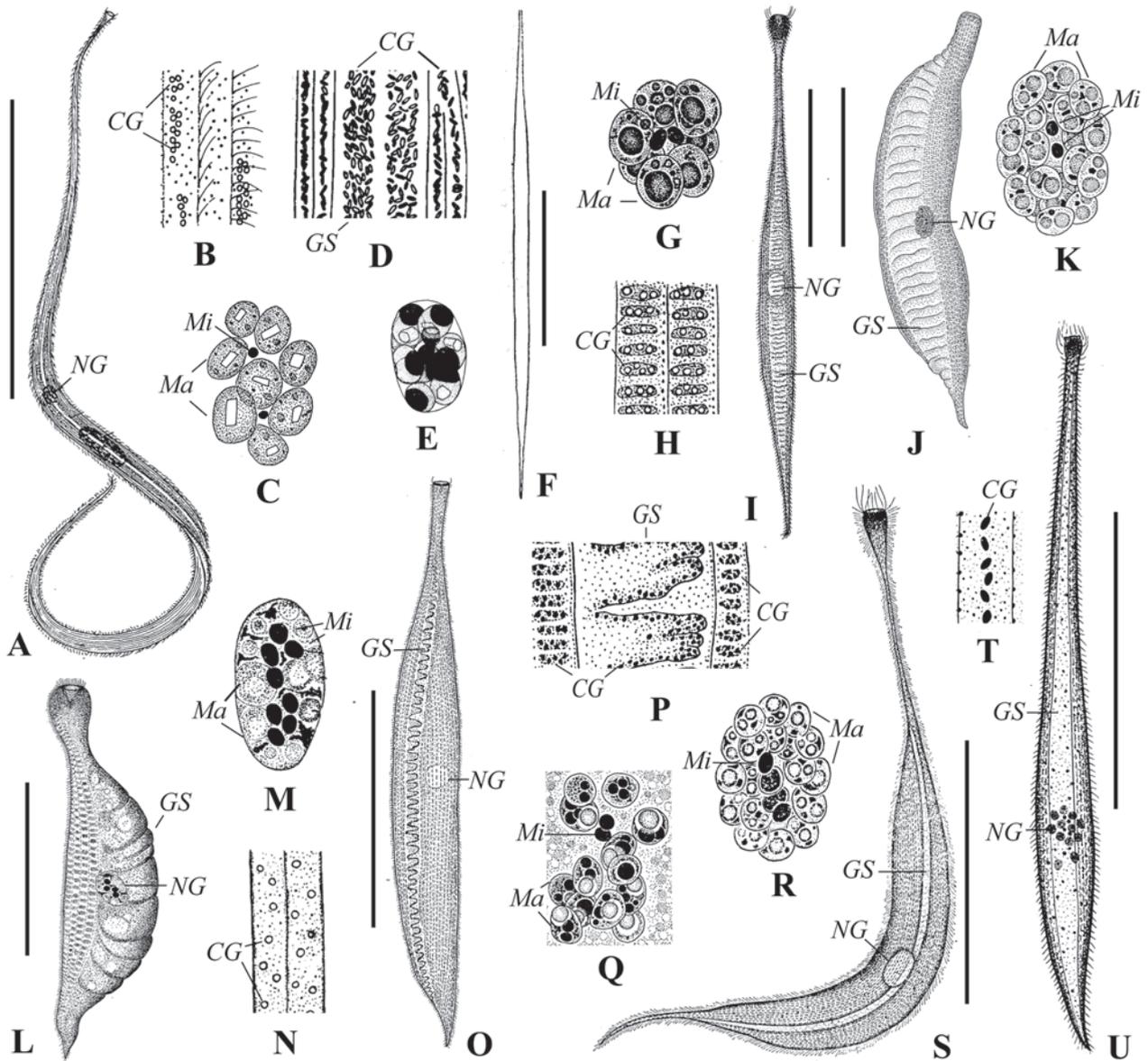
– Data not available.

or capsule. Based on the number of macronuclei and the general cell morphology, there are nine species that should be compared with *T. huangi* spec. nov. (Figs 3A–U; Table 2).

Tracheloraphis multicineta (Raikov and Kovaleva, 1968) Carey, 1992, which was originally described as a morphotype of *T. prenanti*, can be separated from the new species by having fewer macronuclei (16–20 vs. 25–30) and somatic kineties (20–26 vs. 29–37) (Table 2; Raikov and Kovaleva 1968). However, there is no description of its morphology *in vivo*, e.g. body shape, size, cortical granules etc, and the only figure of this species is from a fixed and contracted cell (Figs 3J,

K). This species therefore needs to be reinvestigated. A second morphotype originally described as morphotype of *T. prenanti*, namely *T. oligocineta* (Raikov and Kovaleva, 1968) Carey, 1992 (Figs 3G–I), is also distinct from the new form in having fewer macronuclei (6–8 vs. 25–30) and somatic kineties (14–18 vs. 29–37) (Table 2; Raikov and Kovaleva 1968).

Tracheloraphis totevi Kovaleva and Golemansky, 1979 (Figs 3R, S) differs from the new form by its larger size *in vivo* (2000–3000 μm vs. 500–1200 μm in length), more somatic kineties (42–60 vs. 29–37) and fewer macronuclei (16–22 vs. 25–30) (Table 2; Kovaleva and Golemansky 1979).



Figs 3A–U. Morphology of some closely-related congeners of *Tracheloraphis huangi* spec. nov. **A–C** – *Tracheloraphis aragoi*, general view (A), cortical granules (B) and the nuclear group (C) (from Foissner and Dragesco 1996b); **D–F** – *Tracheloraphis dracontoides*, general view (F), cortical granules (D) and the nuclear group (E) (from Borror 1963); **G–I** – *Tracheloraphis oligocineta*, general view (I), nuclear group (G) and cortical granules (H) (from Raikov and Kovaleva 1968); **J, K** – *Tracheloraphis multicineta*, general view (J) and nuclear group (K) (from Raikov and Kovaleva 1968); **L–N** – *Tracheloraphis sarmatica*, general view (L), nuclear group (M) and cortical granules (N) (from Agamaliev 1966); **O–Q** – *Tracheloraphis serrata*, general view (O), nuclear group (P) and cortical granules (Q) (from Raikov and Kovaleva 1968); **R, S** – *Tracheloraphis totevi*, general view (S) and nuclear group (R) (from Kovaleva and Golemansky 1979); **T, U** – *Tracheloraphis drachi*, general view (U) and cortical granules (T) (from Dragesco 1960). CG – cortical granules, GS – glabrous stripe, Ma – macronuclei, Mi – micronuclei, NG – nuclear group. Scale bars: 600 µm (A, J, O, U), 300 µm (F, I, L), 800 µm (S).

Tracheloraphis phoenicopterus (Figs 6A–S) has a similar body size to the new species. However, it can be distinguished by having fewer macronuclei (6–10 vs. 25–30) and somatic kineties (23–32 vs. 29–37) (Table 2).

Tracheloraphis sarmatica Agamaliev and Kovaleva in Agamaliev, 1966 (Figs 3L–N) can be clearly separated from the new species by having a smaller body size *in vivo* (400–600 µm vs. 500–1200 µm in length)

Table 2. Comparison of *Tracheloraphis huangi* spec. nov. with some closely related congeners. SK – somatic kineties, Ma – macronuclei.

Species	Body length <i>in vivo</i> , μm	SK, number	Ma, number	Data source
<i>Tracheloraphis huangi</i> spec. nov.	500–1200	29–37	25–30	Present work
<i>T. multicineta</i>	800–1600	20–26	16–20	Raikov and Kovaleva (1968)
<i>T. oligocineta</i>	500–1000	14–18	6–8	Raikov and Kovaleva (1968)
<i>T. phoenicopterus</i>	600–1300	23–32	6–10	Present work
<i>T. totevi</i>	2000–3000	42–60	16–22	Kovaleva and Golemansky (1979)
<i>T. sarmatica</i>	400–600	13–17	*	Agamaliev (1966)
<i>T. serrata</i>	up to 1500	40–45	8–12	Raikov and Kovaleva (1968)
<i>T. aragoi</i>	1100–2300	36–42	5–12	Foissner and Dragesco (1996b)
<i>T. dracontoides</i>	950–1500	31–37	7–11	Borror (1963)
<i>T. drachi</i>	1300–2000	28–30	12	Dragesco (1960)

*Macronuclei forming a tight cluster so the number is difficult to determine.

and fewer somatic kineties (13–17 vs. 29–37) (Table 2; Agamaliev 1966).

Tracheloraphis serrata Raikov and Kovaleva, 1968 (Figs 3O–Q) and *T. aragoi* (Dragesco, 1953) Dragesco, 1960 (Figs 3A–C) can be separated from the new species by having more somatic kineties (40–45, 36–42 vs. 29–37) and fewer macronuclei (8–12, 5–12 vs. 25–30) (Table 2; Foissner and Dragesco 1996b, Raikov and Kovaleva 1968). Moreover, the distribution of cortical granules of *T. aragoi* is conspicuously different from that of the new species (clustered in piles vs. scattered) (Fig. 3B; Foissner and Dragesco 1996b).

Tracheloraphis dracontoides (Bullington, 1940) Borror, 1963 and *T. drachi* (Dragesco, 1953) Dragesco, 1960 both resemble the new species with respect to the number of somatic kineties. However, they differ from *T. huangi* spec. nov. in having fewer macronuclei (7–11, 12 vs. 25–30) and distinctly different cortical granules (ovoid and conspicuous vs. globular and inconspicuous) (Figs 3D–F, T, U; Table 2; Borror 1963, Dragesco 1960).

***Tracheloraphis colubis* (Kahl, 1933) comb. nov. (Figs 4, 5; Table 1)**

Basionym: *Trachelocerca coluber* Kahl, 1933. “*Tracheloraphis*” has feminine gender, thus the species name should be changed to concord with the gender of the genus.

Kahl (1933) described this species for the first time based on live observations. However, he neither provided a figure nor fixed a type specimen hence this species is a nomen nudum according to the ICZN

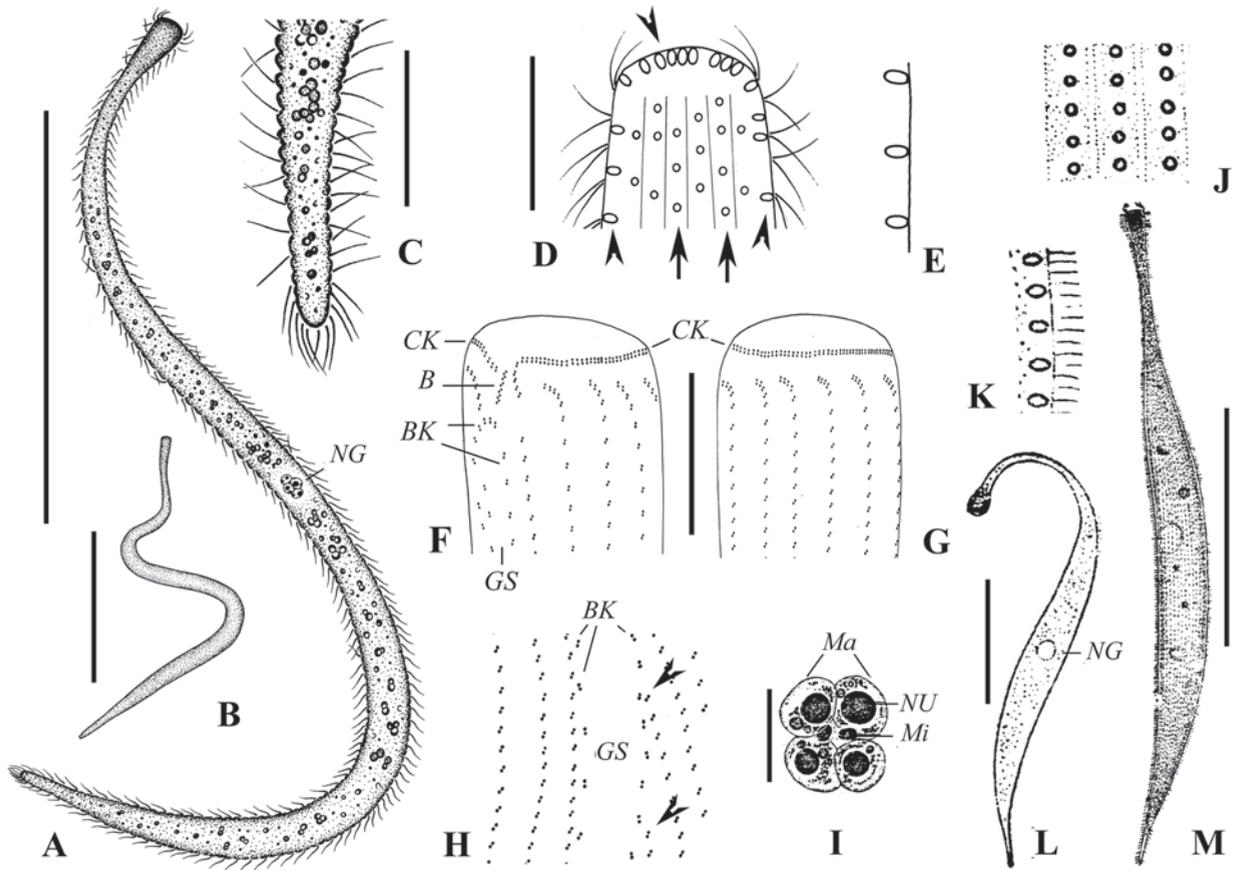
(1999). Raikov (1963) made a detailed redescription of its living morphology and supplied an illustration. However, information about its infraciliature remained unavailable. This species was therefore reinvestigated following observations of both live and silver-stained specimens from the Qingdao population. A redescription and an improved diagnosis are here supplied including details of the infraciliature.

Improved diagnosis of *Tracheloraphis colubis*:

Extended cells about $500\text{--}1000 \times 20\text{--}40 \mu\text{m}$ *in vivo*; claviform tail. 23–31 somatic kineties. Glabrous stripe narrow, about the width of one kinety. 4 macronuclei in a single group. Cortical granules ellipsoid, ca. $1.5\text{--}2 \times 2.5 \mu\text{m}$, colorless.

Deposition of voucher materials: A voucher slide with protargol-impregnated specimens has been deposited in the laboratory of Protozoology, Ocean University of China (registration number: XY2010051102). A second voucher slide is deposited in the Natural History Museum, London, UK, with registration number NHMUK 2011.4.27.2.

Redescription: Extended cell about $500\text{--}1000 \times 20\text{--}40 \mu\text{m}$ *in vivo*; body flattened about 3 : 1, flexible and contractile (Figs 4A, B, 5A, B); neck and tail indistinctly separated from trunk with triangular head and conspicuous claviform tail (Figs 4C, 5C). Cortical granules ellipsoid, ca. $1.5\text{--}2 \times 2.5 \mu\text{m}$, colorless in bright field at high magnification; circular in outline when viewed from above, elliptical in lateral view; arranged in rows between somatic kineties and sparsely distributed in glabrous stripe (Figs 4D, E, 5E–I). Cytoplasm colorless and transparent, packed with cytoplasmic



Figs 4A–M. *Tracheloraphis colubis* from life (A–E, J–M), after protargol impregnation (F–I). **A** – typical individual, noting the nuclear group; **B** – showing the flexibility of the cell; **C** – posterior end, showing the rounded, claviform tail; **D** – anterior part, marking the arrangement of the cortical granules, the outlines of which are round in top view (arrows) and elliptical in lateral view (arrowheads); **E** – lateral view, showing the elliptical cortical granules at the cell margin; **F, G** – infraciliature of anterior end to mark circumoral kinety, brosse, glabrous stripe and bristle kineties; **H** – infraciliature of mid-body, showing the glabrous stripe bordered by bristle kinety and anterior secant system (arrowheads); **I** – nuclear group composed of macronuclei and micronuclei (from Raikov 1963); **J, K** – to show the distribution of cortical granules (from Raikov 1963); **L, M** – general view (from Raikov 1963). B – brosse, BK – bristle kinety, CK – circumoral kinety, GS – glabrous stripe, Ma – macronuclei, Mi – micronuclei, NG – nuclear group, NU – nucleoli. Scale bars: 200 μm (A, B, L, M), 50 μm (C, D, F, G), 10 μm (I).

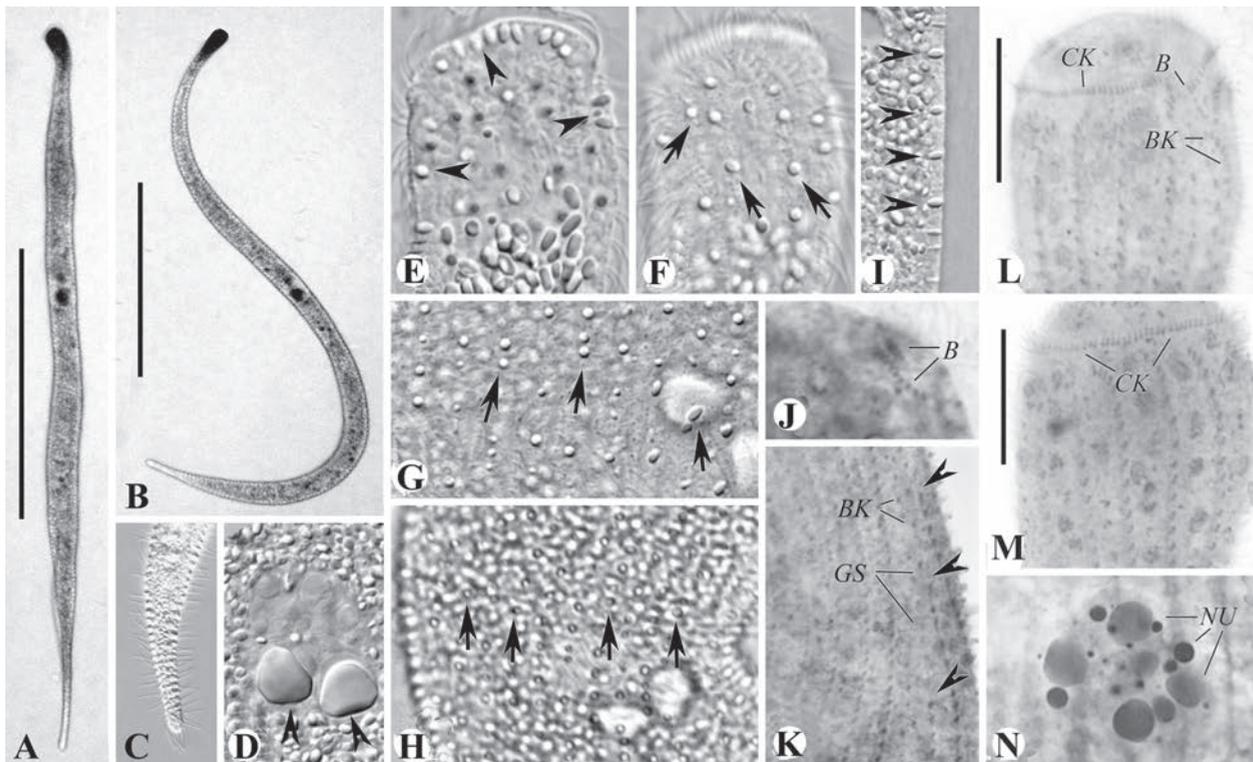
granules that are ellipsoid, about 1–3 μm long and colorless. Contractile vacuole not observed.

Locomotion by gliding, winding between sand grains and organic debris.

Entire infraciliature consisting of dikinetids. Cilia about 10 μm long *in vivo* and arranged in longitudinal rows. Usually one or two brosse kineties (Figs 4F, 5J, L). Glabrous stripe very narrow, bordered by irregularly spaced bristle kinety (Figs 4H, 5K). Anterior and posterior secant system formed on left side of glabrous stripe where some kineties abut to the bristle kinety (Figs 4H, 5K). Four macronuclei in a single group (Figs 5D, N); micronuclei difficult to detect.

Discussion: Kahl (1933) assigned this species to the genus *Trachelocerca* based on its curved posterior end. Later, Raikov (1963) redescribed it and retained it in the genus *Trachelocerca* because of its narrow glabrous stripe. However, these characters are of very limited value for the generic classification of trachelocercids (Foissner and Dragesco 1996b). According to present study, this species has the circumoral kinety interrupted by a brosse and thus should be transferred to the genus *Tracheloraphis* (Foissner and Dragesco 1996b).

The Qingdao population matches the population described by Raikov (1963) in most characters, particularly the claviform tail, the large ellipsoid cortical



Figs 5A–N. Photomicrographs of *Tracheloraphis colubis* from life (A–I) and after protargol impregnation (J–N). **A, B** – two typical individuals; **C** – claviform tail; **D** – nuclear group; arrowheads point to the (protein) crystals (?); **E, F, I** – anterior end, noting cortical granules in lateral view (arrowheads in **E** and **I**) and top view (arrows in **F**); **G, H** – arrows indicate the cortical granules between the ciliary rows; **J, L, M** – anterior end, to show the circumoral kinety, brosse and bristle kineties; **K** – mid-body, noting the narrow glabrous stripe bordered by bristle kinety and anterior secant system (arrowheads); **N** – tightly clustered nuclei. B – brosse, BK – bristle kinety, CK – circumoral kinety, GS – glabrous stripe, NU – nucleoli. Scale bars: 200 μm (A, B), 50 μm (L, M).

granules and the single nuclear group. The main difference between these two populations is the range of the number of somatic kineties, i.e. 23–31 in the Qingdao population vs. 28–30 in the population described by Raikov (1963). Nevertheless, the two values overlap so we have no doubt that these populations are conspecific.

***Tracheloraphis phoenicopterus* (Cohn, 1886) Dragesco, 1960 (Fig. 6; Table 1)**

This species was redescribed in detail by Foissner and Dragesco (1996b). Therefore, only a brief description based on the Qingdao population is documented here.

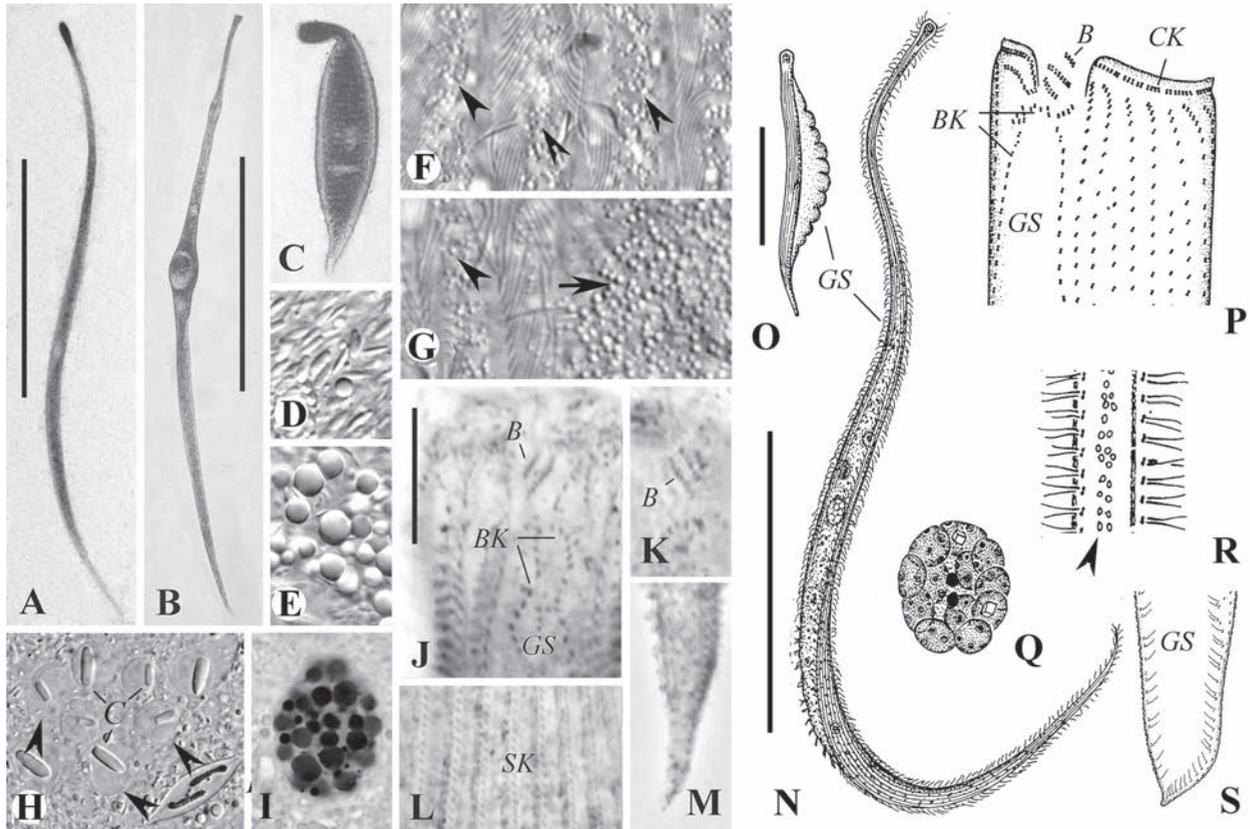
Deposition of voucher materials: A voucher slide with protargol-impregnated specimens has been deposited in the laboratory of Protozoology, Ocean University of China (registration number: XY2010051401). A second voucher slide is deposited in the Natural

History Museum, London, UK, with registration number NHMUK 2011.4.27.3.

Description: Extended cell about 600–1300 \times 20–40 μm *in vivo*; body laterally flattened and contractile (Figs 6A–C); triangular head and conspicuously pointed tail. Cytoplasm colorless packed with cytoplasmic granules, that are either ellipsoid or round and colorless (Figs 6D, E). Cortical granules globular, 0.5–1 μm in diameter, colorless, located between ciliary rows and in glabrous stripe (Figs 6F, G). Glabrous stripe almost half of the body width. Contractile vacuole not observed.

Locomotion by gliding, winding between sand grains and organic debris.

Entire infraciliature consisting of dikinetids. Cilia about 8–9 μm long *in vivo* and arranged in longitudinal rows. 23–32 somatic kineties. Usually two or three brosse kineties (Figs 6J, K). Glabrous stripe bordered by irregularly spaced bristle kinety (Fig. 6J). One nuclear



Figs 6A–S. *Tracheloraphis phoenicopterus* from life (A–H, N, O, Q, R) and after protargol impregnation (I–M, P, S). **A, B** – typical individuals; **C** – a contracted cell; **D, E** – ellipsoidal (D) and spherical (E) cytoplasmic granules; **F, G** – cortical granules between ciliary rows (arrowheads in F and G) and in glabrous stripe (arrow in G); **H** – nuclear group, noting macronuclei (arrowheads) and (protein) crystals; **I** – nuclear group, to show the nuclei forming a tight cluster; **J, K** – anterior end, noting the brosse, glabrous stripe and bristle kinety; **L** – somatic kineties; **M** – posterior end of cell showing the conspicuously pointed tail; **N** – typical individual (from Foissner and Dragesco 1996b); **O** – contracted individual (from Foissner and Dragesco 1996b); **P, S** – anterior (P) and posterior (S) ends, noting circumoral kinety, brosse and glabrous stripe bordered by bristle kinety (from Foissner and Dragesco 1996b); **Q** – nuclear group (from Foissner and Dragesco 1996b); **R** – surface view of cortex (from Foissner and Dragesco 1996b). B – brosse, BK – bristle kinety, C – (protein) crystal, CK – circumoral kinety, GS – glabrous stripe, SK – somatic kineties. Scale bars: 300 μm (A, B), 15 μm (J), 200 μm (N), 100 μm (O).

group ca. 15–20 μm in diameter located in middle of body, containing about 6–10 macronuclei (Figs 6H, I).

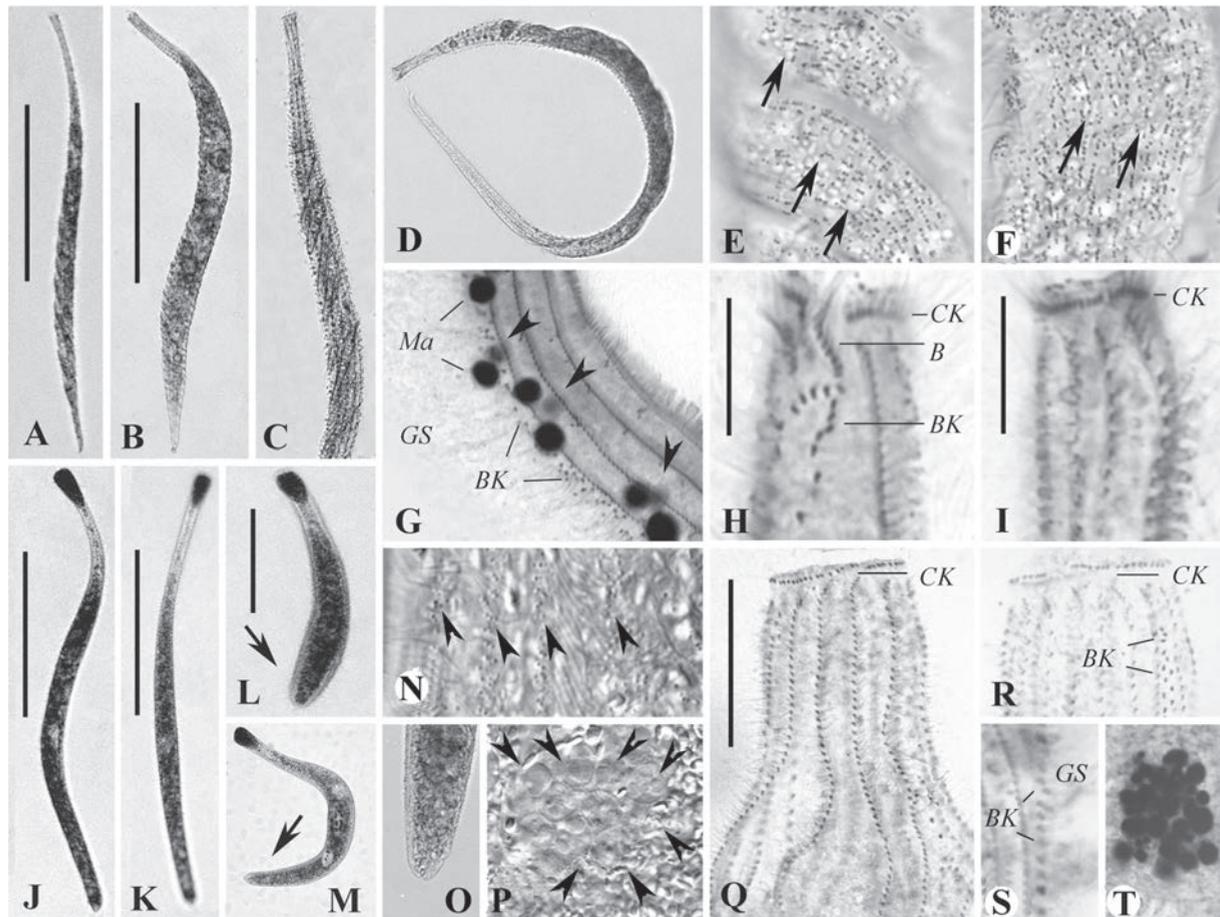
Discussion: There have been several redescrptions of *Tracheloraphis phoenicopterus* since it was first reported by Cohn (1866). The Qingdao population corresponds well with the Roscoff population described by Foissner and Dragesco (1996b) in terms of body size and shape and the number of macronuclei. The main difference between these two populations is the range of the number of somatic kineties (23–32 in Qingdao population vs. 23–27 in Roscoff population), although the two values overlap so the difference is not significant. Furthermore, the cortical granules of the Roscoff population of *T. phoenicopterus* are ellipsoidal

and about $0.6 \times 1.2 \mu\text{m}$ in size (Fig. 6R; Foissner and Dragesco 1996b), whereas in the Qingdao population they are globular and 0.5–1 μm in diameter (Figs 6F, G). Again, this difference is not considered significant and we have no doubt that the two populations are conspecific.

***Tracheloraphis oligostriata* (Raikov, 1962) Foissner & Dragesco, 1996 (Figs 7A–I; Table 1)**

This species was redescrbed in detail by Foissner and Dragesco (1996b). Therefore, only a brief description of the Qingdao population is documented here.

Deposition of voucher materials: A voucher slide with protargol-impregnated specimens has been depos-



Figs 7A–T. *Tracheloraphis oligostriata* from life (A–F) and after protargol impregnation (G–I); *Trachelocerca incaudata* from life (J–P) and after protargol impregnation (Q–T). A–D – typical individuals; E, F – cortical granules (arrows) in the glabrous stripe of a contracted (E) and extended (F) cell; G – mid-body, showing the glabrous stripe, bristle kinety, macronuclei and micronuclei (arrowheads); H, I – anterior end, noting the circumoral kinety, brosse and bristle kinety; J–M – different individuals, arrows in (L) and (M) point to the rounded posterior end; N – to show the distribution of cortical granules between ciliary rows (arrowheads); O – rounded posterior end; P – nuclear group, arrowheads indicate the macronuclei forming a tight cluster; Q, R – anterior end, noting the circumoral and bristle kineties (R); S – mid-body, marking the glabrous stripe bordered by the bristle kinety; T – nuclear group, to show the nuclei forming a tight cluster. B – brosse, BK – bristle kinety, CK – circumoral kinety, GS – glabrous stripe, Ma – macronuclei. Scale bars: 200 μm (A, B, J–L), 20 μm (H, I, Q).

ited in the laboratory of Protozoology, Ocean University of China (registration number: XY2010042901). A second voucher slide is deposited in the Natural History Museum, London, UK, with registration number NHMUK 2011.4.27.4.

Description: Extended cell about 300–600 \times 20–40 μm *in vivo*; body flattened, flexible and contractile (Figs 7A–D). Body yellowish to colorless; anterior part transparent; posterior end pointed and slightly curved (Figs 7A–C). Cortical granules rounded, ca. 1 μm in diameter, yellow to brown, arranged between ciliary rows and densely distributed in glabrous stripe (Figs

7E, F). Glabrous stripe as wide as the body width. Contracted cells show many transverse and oblique folds with left side protruding and tuberculate (Figs 7C–E). Contractile vacuole not observed.

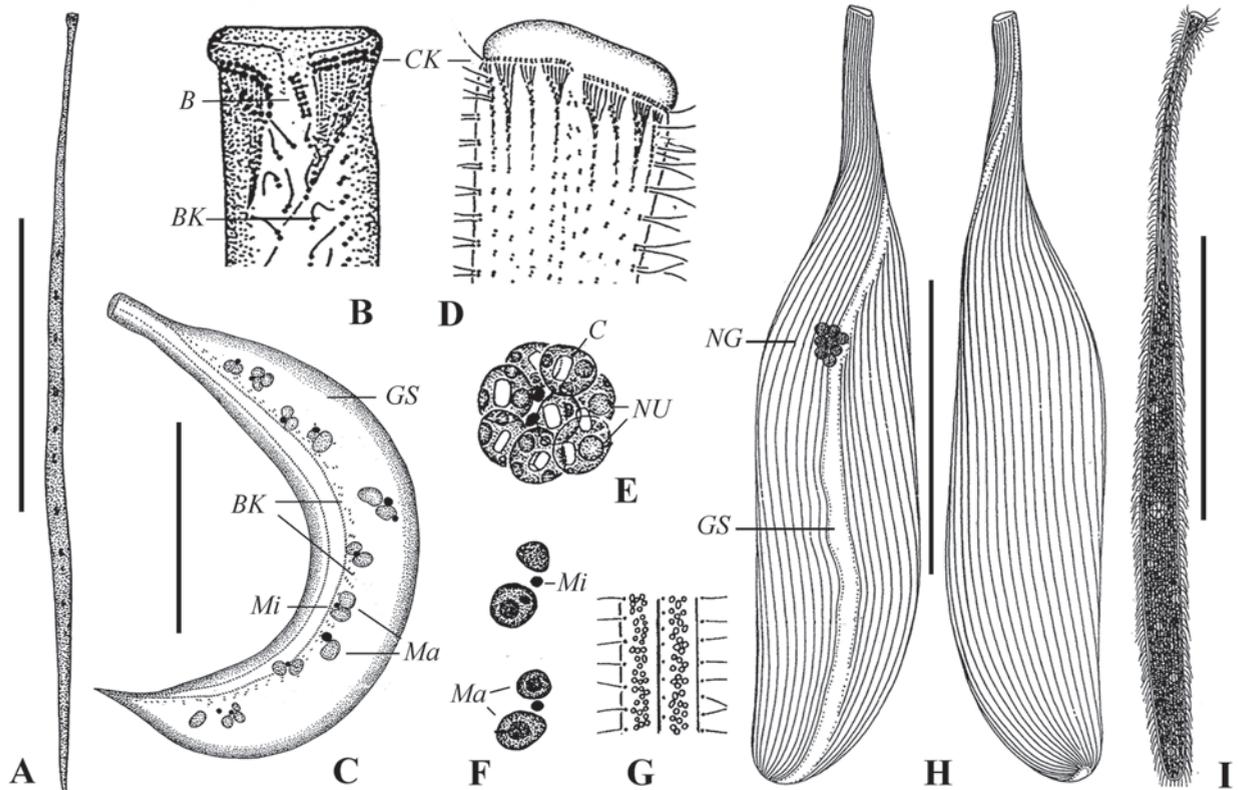
Invariably 6 somatic kineties; no anterior or posterior secant system on either side of the glabrous stripe (Fig. 7G). One brosse kinety (Fig. 7H). Glabrous stripe bordered by irregularly spaced bristle kinety (Figs 7G, H). 4–16 macronuclei and 3–8 micronuclei forming a strand in the cell midline comprising several small nuclear groups, usually with 2 or 3 macronuclei and 1 or 2 micronuclei in each group (Fig. 7G).

Remarks: This species was originally assigned to the genus *Trachelonema* (Raikov 1962). However, when the classification of trachelocercid karyorelictids was revised, it was transferred to the genus *Tracheloraphis* (Foissner and Dragesco 1996b). The Qingdao population corresponds closely both with the original and the Roscoff populations (Raikov 1962, Foissner and Dragesco 1996b) in terms of body size, shape and the number of somatic kineties (Figs 8A–C, F), the only difference being a slight variation in the number of macronuclei. However, we believe this is probably population-dependent so the identity of the Qingdao population is beyond doubt.

***Trachelocerca incaudata* Kahl, 1933 (Figs 7J–T; Table 1)**

This species was redescribed in detail by Foissner (1997b). Therefore, only a brief description based on Qingdao population is documented here.

Deposition of voucher materials: A voucher slide with protargol-impregnated specimens has been deposited in the laboratory of Protozoology, Ocean University of China (registration number: XY2010051101). A second voucher slide is deposited in the Natural History Museum, London, UK, with registration number NHMUK 2011.4.27.5.



Figs 8A–I. *Tracheloraphis oligostriata* from Foissner and Dragesco (1996b) (A–C, F), and *Trachelocerca incaudata* from Foissner (1997b) (D, E, G–I). **A** – typical individual; **B** – infraciliature of anterior end, marking circumoral kinety, brosse and bristle kinety; **C** – lateral view to show the general infraciliature, glabrous stripe, bristle kinety and nuclear groups distributed in a strand along the cell midline; **D** – infraciliature of anterior end, to indicate the circumoral and bristle kineties; **E** – nuclear group, noting the (protein) crystals and nucleoli; **F** – nuclear groups, marking the macronuclei and micronuclei; **G** – to show the distribution of cortical granules between the ciliary rows; **H** – showing the general infraciliature, single nuclear group and glabrous stripe; **I** – typical individual. B – brosse, BK – bristle kinety, C – (protein) crystal, CK – circumoral kinety, GS – glabrous stripe, Ma – macronuclei, Mi – micronuclei, NG – nuclear group, NU – nucleoli. Scale bars: 200 μ m (A, I), 70 μ m (C, H).

Description: Extended cell about 300–600 × 20–40 µm *in vivo*; body claviform and contractile; head and neck areas distinct, posterior end rounded (Figs 7J–M, O). Body grey to blackish in color due to numerous inclusions (Figs 7J–L); cortical granules rounded, ca. 0.5 µm in diameter, colorless, located between ciliary rows and in glabrous stripe (Fig. 7N). Glabrous stripe narrow, about the width of 2 or 3 ciliary rows. Contractile vacuole not observed.

Twenty-three to 26 somatic kineties with cilia about 10 µm long *in vivo*; with anterior and posterior secant system on the left side of the glabrous stripe (Fig. 7Q). Glabrous stripe bordered by irregularly spaced bristle kinety (Figs 7R, S). Single nuclear group, ca. 15 µm in diameter, composed of 6–8 macronuclei (Fig. 7T).

Remarks: The original description of *Trachelocerca incaudata* by Kahl (1933) was rather brief and detailed data were not available until it was redescribed by Foissner (1997b). The Qingdao population corresponds well with Foissner's (1997b) Roscoff population (Figs 8D, E, G–I) in terms of body size and shape, the width of the glabrous stripe and the number of macronuclei. The only minor difference is the range of the number of somatic kineties, i.e. 23–26 vs. 25–40 in the Roscoff population (Foissner 1997b). Nevertheless, since these values overlap we do not consider this to be a significant difference. Therefore, we have no doubt that the two populations are conspecific.

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