

Redescriptions of Three Marine Peritrichous Ciliates, *Zoothamnium alternans* Claparède et Lachmann, 1859, *Z. sinense* Song, 1991 and *Z. commune* Kahl, 1933 (Ciliophora, Peritrichia), from North China

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Summary. Three marine peritrichous ciliates, *Zoothamnium alternans* Claparède et Lachmann, 1859, *Z. sinense* Song, 1991 and *Z. commune* Kahl, 1933, were isolated from coastal waters near Qingdao, China. The living morphology, infraciliature and silverline system were studied in both living and silver-impregnated specimens. Based on the Qingdao populations, improved diagnoses and redescrptions are provided for each species: *Z. alternans* is distinguished from its congeners by the alternately branched stalk, the macrozooids on the main stalk trunk, the J-shaped macronucleus and in having 40-55 silverlines between the oral area and the trochal band and 20-30 between the trochal band and the scopula; *Z. sinense* can be recognized by the small zooid size, the alternately branched stalk, and in having 55-70 silverlines between the oral area and the trochal band and 20-35 between the trochal band and the scopula; *Z. commune* can be distinguished from other species of similar zooid shape and size by the form of peniculus 3 and the number of silverlines between the oral area and the trochal band (59-70) and between the trochal band and the scopula (38-43).

Key words: marine ciliate, morphology, Peritrichida, *Zoothamnium*.

INTRODUCTION

Zoothamnium Bory de St. Vincent, 1826 is a well-known genus with more than 70 species, many of which are abundant in marine biotopes (Kent 1880-1882, Kahl 1935, Stiller 1971, Küsters 1974, Corliss, 1979, Jankowski

1985). In common with most other genera of sessile peritrichs, however, few *Zoothamnium* species have been studied using modern methods (e.g. silver impregnation) as recommended by Foissner and Schiffmann (1974). Furthermore, there has been no revision of this genus since that of Kahl (1935). Consequently species separation and identification among *Zoothamnium* spp. is often very difficult (Kahl 1933; Precht 1935; Sommer 1951; Stiller 1953a, b; Stiller and Stevčić 1967).

In the present study we report on three marine *Zoothamnium* species collected from the littoral zone of coastal waters near Qingdao, China, following observations on both living and silver-impregnated specimens.

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All three species are known forms for which detailed information concerning the infraciliature and silverline system are lacking. Therefore, based on the Qingdao populations, an improved diagnosis and redescription of each species is presented here.

MATERIALS AND METHODS

Sample collection. Ciliates were collected from coastal waters at three sites in the Qingdao region, P.R. China, (36°08'N; 120°43'E) from both natural (*Zoothamnium alternans*) and artificial (*Zoothamnium sinense*, *Z. commune*) substrates. The natural substrate was the shell of the scallop *Chlamys* sp. The artificial substrates were glass slides fixed to a frame that was immersed in water and left for 10 days to allow colonization to occur. After this time the slides were retrieved and transported to the laboratory for examination.

Zoothamnium alternans was collected on 19 April 2004 from a scallop (*Chlamys* sp.) farming pond, water temperature 12°C, salinity about 30‰.

Zoothamnium sinense was collected on 10 July 2002 from the littoral zone of coastal waters off Qingdao, water temperature 26°C, salinity 30‰.

Zoothamnium commune was collected on 22 May 2002 from a shrimp-farming pond, water temperature 20°C, salinity 28‰.

Observations: Ciliates were observed *in vivo* using bright field and differential interference contrast microscopy. The infraciliature was revealed with protargol impregnation according to Wilbert (1975). The "dry" silver nitrate method (Foissner 1976) was used to demonstrate the silverline system.

Drawings and terminology: Drawings of impregnated specimens were made with the help of a camera lucida at $\times 1250$ magnification. Terminology is mainly according to Foissner and Schiffmann (1974) and Warren (1986).

Deposition of slides: Two slides of *Zoothamnium alternans* (No. 0404190101, 0404190102) with protargol-impregnated specimens are deposited at the Laboratory of Protozoology, OUC, China. Another slide (No. 2005:3:24:7) is deposited in the collections of the Natural History Museum, London, U.K.

One slide of *Zoothamnium sinense* (No. 0207100101) with protargol-impregnated specimens is deposited in the collection of the Laboratory of Protozoology, OUC, China.

One slide of *Zoothamnium commune* (No. 0205220401) with protargol-impregnated specimens is deposited at the Laboratory of Protozoology, OUC, China. Another slide (No. 2005:3:24:8) is deposited in the collections of the Natural History Museum, London, U.K.

RESULTS

Class: Oligohymenophora de Puytorac *et al.*, 1974
 Subclass: Peritrichia Stein, 1859
 Order: Sessilida Kahl, 1933
 Family: Vorticellidae Ehrenberg, 1838
 Genus: *Zoothamnium* Bory de St. Vincent, 1826

Redescription of *Zoothamnium alternans* Claparède et Lachmann, 1859 (Figs 1, 2; Table 1)

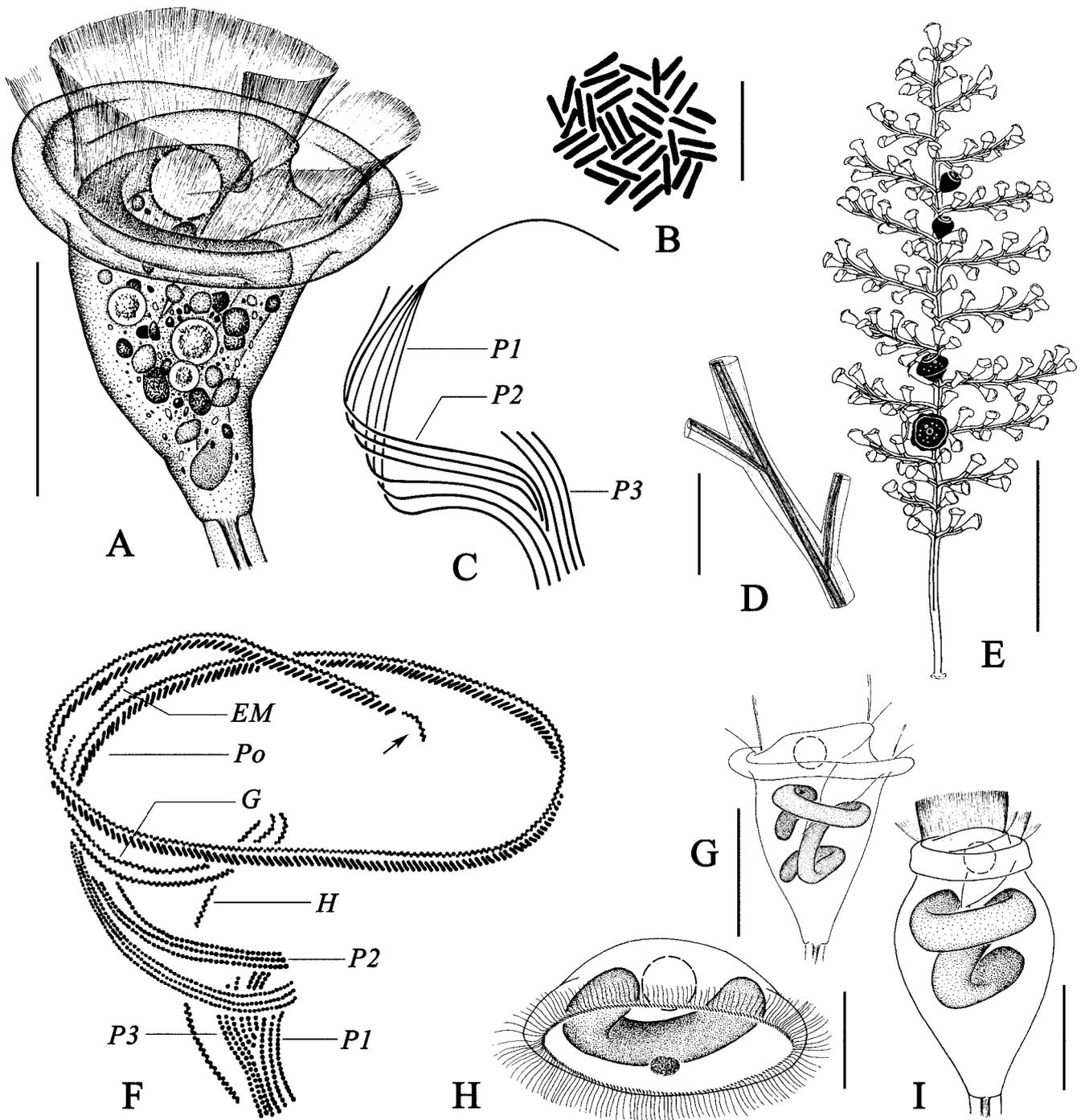
Synonym: *Zoothamnium chlamydis* Hu et Song, 2001 (p216)

Although *Zoothamnium alternans* has been known for a long time (Claparède and Lachmann 1859; Kent 1880-1882; Kahl 1933, 1935), it has never been investigated using silver impregnation methods. Therefore its infraciliature and silverline system remain unknown. Based on the population found in Qingdao, a detailed redescription and an improved diagnosis are presented here.

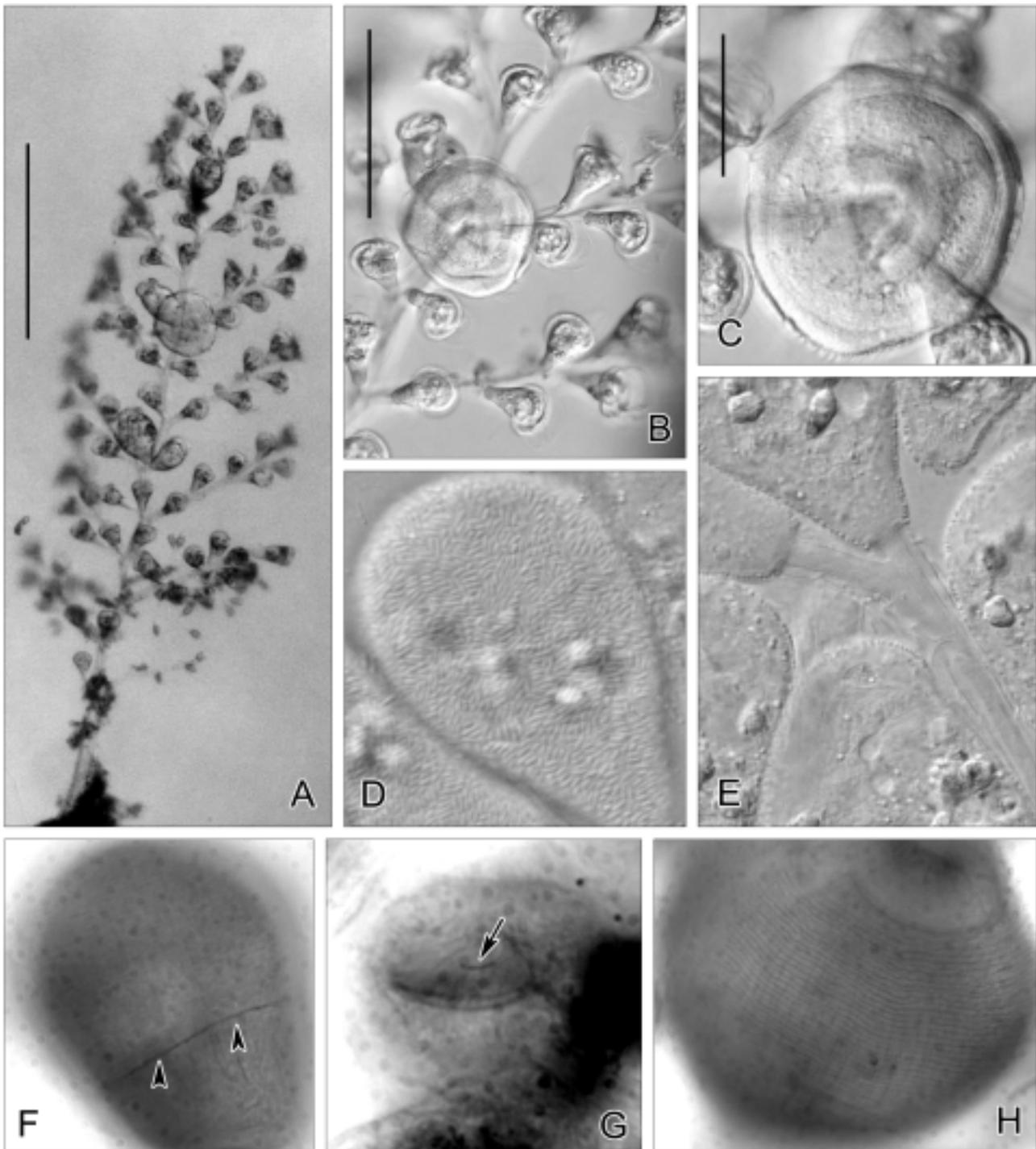
Amended diagnosis: Marine *Zoothamnium* with alternately branched stalk. Cells differentiated into micro- and macrozooids. Macrozooids ovoid, *ca* 70-90 \times 45-55 μm *in vivo*, located only on main stalk trunk; microzooids inverted bell-shaped, 40-56 \times 26-32 μm in size. Both micro- and macrozooids have a J-shaped macronucleus and an apically located contractile vacuole. Number of silverlines from oral area to aboral trochal band *ca* 40-55, from aboral trochal band to scopula *ca* 20-30.

Amended description: Colonies with micro- and macrozooids. Microzooid inverted bell-shaped, 40-56 \times 26-32 μm *in vivo*, with thin, distinctly everted peristomial lip and moderately elevated peristomial disc (Figs 1A, G). Cytoplasm colourless to slightly grey, usually containing granules and several vacuoles (Fig. 1A). Macronucleus J-shaped in lateral view. Oral section of macronucleus elongate, cylindroid in cross-section, semicircular, transversely oriented. End of oral section sometimes recurved aborally (Fig. 1G). Aboral continuation of macronucleus cylindroid, straight, oriented diagonally to longitudinal axis. Aboral extremity of macronucleus usually recurved orally. Single contractile vacuole located slightly above peristomial lip and pulses slowly (3-5 min intervals). Pellicle appears smooth, usually covered with a layer of rod-shaped bacteria (Figs 1B, 2D). Fine pellicular striations can be observed occasionally under $\times 1000$ magnification (Fig. 2H). Number of silverlines from aboral trochal band to scopula *ca* 20-30, from peristome to aboral trochal band *ca* 40-55.

Macrozooid oval-shaped, 70-90 \times 45-55 μm *in vivo*, widest at mid-body, with thick peristomial lip, J-shaped macronucleus and single, apically located contractile vacuole (Figs 1I, 2A). Macrozooids located only on main trunk of stalk, one to several per colony, but absent at newly formed colonies. Telotroch acetabuliform (sau-



Figs 1A-I. *Zoothamnium alternans* from life (A, B, D, E, G-I) and after protargol impregnation (C, F). **A** - a microzooid; **B** - bacteria on pellicle; **C** - detail of infundibular polykineties; **D** - stalk structure; **E** - colony form, from Grell (1968); **F** - oral infraciliature, arrow marks the distal kinety fragment; **G** - lateral view of a microzooid; **H** - a telotroch; **I** - a macrozooid. EM - epistomial membrane; G - germinal kinety, H - haplokinety, P1-3 - infundibular polykinety 1-3, Po - polykinety. Scale bars: 30 μ m (1A, G-I); 10 μ m (1B); 20 μ m (1D); 300 μ m (1E).



Figs 2A-H. Photomicrographs of *Zoothamnium alternans* from life (A-E) and after protargol impregnation (F-H). **A**- colony form; **B** - showing a telotroch, a macrozooid and several microzooids; **C** - a telotroch; **D** - to show bacteria covering the pellicle; **E** - to show structure of stalk; **F** - lateral view, arrowheads mark the aboral trochal band; **G** - to show the epistomial membrane (arrow); **H** - silverline system. Scale bars: 300 μ m (2A); 200 μ m (2B); 50 μ m (2C).

Table 1. Morphometric characters of *Zoothamnium alternans* (first line, data for microzooids), *Z. sinense* (second line) and *Z. commune* (third line). Min - minimum, Max - maximum, Mean - arithmetic mean, SD - standard deviation, n - sample number.

	Min	Max	Mean	SD	n
Body length <i>in vivo</i> in μm	40	56	48.4	4.91	16
	36	48	40.7	3.46	9
	60	104	84.4	12.9	10
Body width <i>in vivo</i> in μm	26	32	29.3	1.65	16
	30	38	33.3	2.65	9
	48	56	50.0	2.83	9
No. of silverlines from oral area to aboral trochal band	40	55	-	-	4
	55*	70*	-	-	4
	59	70	65.0	3.80	10
No. of silverlines from aboral trochal band to scopula	20	30	-	-	3
	20*	35*	-	-	4
	38	43	40.6	1.67	9

* Detected from living cells.

cer-shaped), 100-120 μm in diameter and 40-50 μm in height, with large C-shaped macronucleus (Figs 1H; 2B, C). Telotrochs only formed by macrozooids.

Colonies up to 1.2 mm high, alternately branched, and containing up to 120 zooids (Figs 1E, 2A). Stalk with smooth surface, main stalk trunk 18 μm in diameter, 8 μm at distal end. Stalk spasmoneme 3-6 μm in width, comprising a dark fibrous bundle within a transparent thecoplasmic sheath, sometimes with a few mitochondria (Figs 1D, 2E).

Buccal apparatus of similar structure to most congeners. Haplokinety and polykinety describing one and one-third turns around peristomial disc before entering infundibulum, where they make another turn (Fig. 1F). One short kinety fragment is always visible at the peristomial end of the haplokinety (Fig. 1F, arrow).

In infundibulum, polykinety (P1) is accompanied by two other polykineties, P2 and P3, each of which consists of three ciliary rows (Fig. 1C). P1 and P2 much longer than P3, and originate from mid-infundibulum. P1 extends posteriorly to end of infundibulum, where it converges with P3 and the two terminate at the same level. Three rows of P2 terminate at different levels and significantly above the posterior ends of P1 and P3 (Fig. 1C). Germinal kinety parallel to haplokinety within abostomal half of infundibulum (Fig. 1F). Epistomial membrane short, located at opening of oral cavity (Figs 1F; 2G, arrow).

Aboral trochal band composed of a row of dikinetids which encircle the cell in aboral third of zooid (Fig. 2F).

Remarks: Given its highly distinctive morphology, i.e. alternately branched stalk, zooid size and appearance, marine habitat and the presence of macrozooids, the identity of the Qingdao population as *Z. alternans* is beyond reasonable doubt (Claparède and Lachmann 1859, Grell 1968).

Zoothamnium alternans is similar to *Z. niveum* Ehrenberg, 1838, which also has an alternately branched stalk, macrozooids on the main trunk of the stalk and a marine habitat, but the latter forms considerably larger colonies (> 1 cm high vs. 1.2 mm high in *Z. alternans*) and has a relatively shorter P2 (about equal to P3 in length vs. more than twice as long as P3 in *Z. alternans*), thus the two can be clearly separated (Bauer-Nebelsick *et al.* 1996).

Another marine species, *Zoothamnium plumula* Kahl, 1933, also has enlarged zooids and an alternately branching stalk, and so is somewhat similar as the current organism. However, these two species can be separated by the number of silverlines between the aboral trochal band and the peristomial border (*ca* 70 in *Z. plumula* vs. 40-55 in *Z. alternans*) and the location of the macrozooids which are located at the distal ends of the accessory branches in *Z. plumula* vs. on the main trunk in *Z. alternans* (Song *et al.* 2002).

Additionally, two other forms of *Z. alternans* were reported by Greeff (1870) and Kent (1880-1882) respectively, both of which are considered to be misidentifications. The population described by Kent has many enlarged zooids on the accessory branches but

Table 2. Morphological comparison between three *Zoothamnium* spp. and morphologically similar congeners. Measurements in μm .

Species	Body length <i>in vivo</i>	Body width <i>in vivo</i>	Number of silverlines from peristome to aboral trochal band	Number of silverlines from aboral trochal band to scopula	Shape of macronucleus	Habitat	Data source
<i>Z. alternans</i>	40-56	26-32	40-55	20-30	J-shaped	marine	present study
<i>Z. chlamydis</i>	50-60	-	27-47	19-29	vertically C-shaped	marine	Hu and Song (2001)
<i>Z. commune</i>	60-104	48-56	59-70	38-43	C-shaped	marine	present study
<i>Z. hentscheli</i>	63-84	35-42	-	-	C-shaped	freshwater	Kahl (1935)
<i>Z. niveum</i>	54-66	16-22	-	-	C-shaped	marine	Bauer-Nebelsick <i>et al.</i> (1996)
<i>Z. plumula</i>	50-100	30-50	70	24-28	C-shaped	marine	Song <i>et al.</i> (2002)
<i>Z. sinense</i>	36-48	30-38	55-70	20-35	C-shaped	marine	present study
<i>Z. sinense*</i>	33-61	29-53	-	-	C-shaped	marine	Song (1991)
<i>Z. thiophilum</i>	90-95	-	-	-	C-shaped	freshwater	Stiller (1946)

*Called *Z. commune* sensu Song, 1991

lacks typical macrozooids on the main stalk trunk while Greeff's population possesses long cylindrical microzooids of a type that are not found in *Z. alternans*.

Zoothamnium chlamydis Hu *et al.* Song, 2001 has a strong similarity to *Z. alternans* in all key characters (Table 2). Therefore *Z. chlamydis* is considered to be a junior synonym of *Z. alternans* (Hu and Song 2001).

Redescription of *Zoothamnium sinense* Song, 1991 (Figs 3, 4; Table 1)

Synonyms: *Zoothamnium commune* sensu Song, 1991 (p47); *Z. truncatum* Song, 1986 (primary homonym) (p229)

Amended diagnosis: Marine *Zoothamnium* with alternately branched stalk; zooids inverted bell-shaped, measuring about $36-48 \times 30-38 \mu\text{m}$ *in vivo*; peristomial disc flat, occasionally very slightly elevated above distinctly everted peristomial lip; macronucleus C-shaped and transversely oriented; single contractile vacuole apically located; pellicle finely striated, with *ca* 20-35 striations from scopula to aboral trochal band and *ca* 55-70 striations from aboral trochal band to oral area.

Amended description: Zooid inverted bell-shaped, usually measuring $36-48 \times 30-38 \mu\text{m}$ *in vivo*, with maximum width just below everted, single-layered peristomial lip (Figs 3A, I; 4B). Peristomial disc flat, occasionally slightly elevated above peristomial lip (Figs 3I,

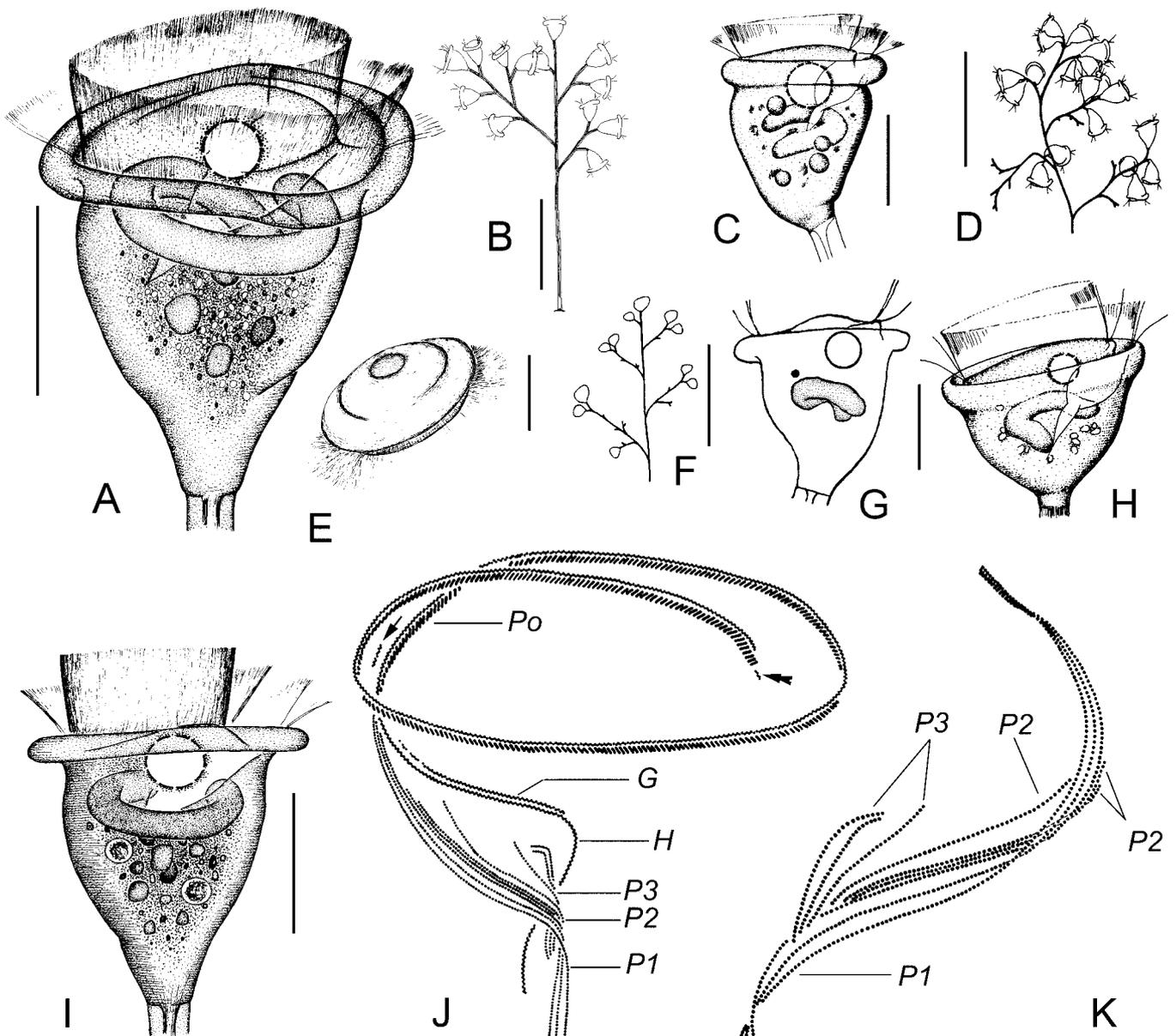
4B). Pellicle appears smooth at low magnification, fine pellicle striations only visible at $\times 1000$ magnification or higher.

Cytoplasm colourless, usually containing granules and a few food vacuoles (Figs 3A, I). Single contractile vacuole apically positioned (Figs 3A, I; 4C, arrow). Macronucleus generally C-shaped, transversely oriented, located in oral 2/3 of body and surrounds the aboral portion of infundibulum (Figs 3A, I).

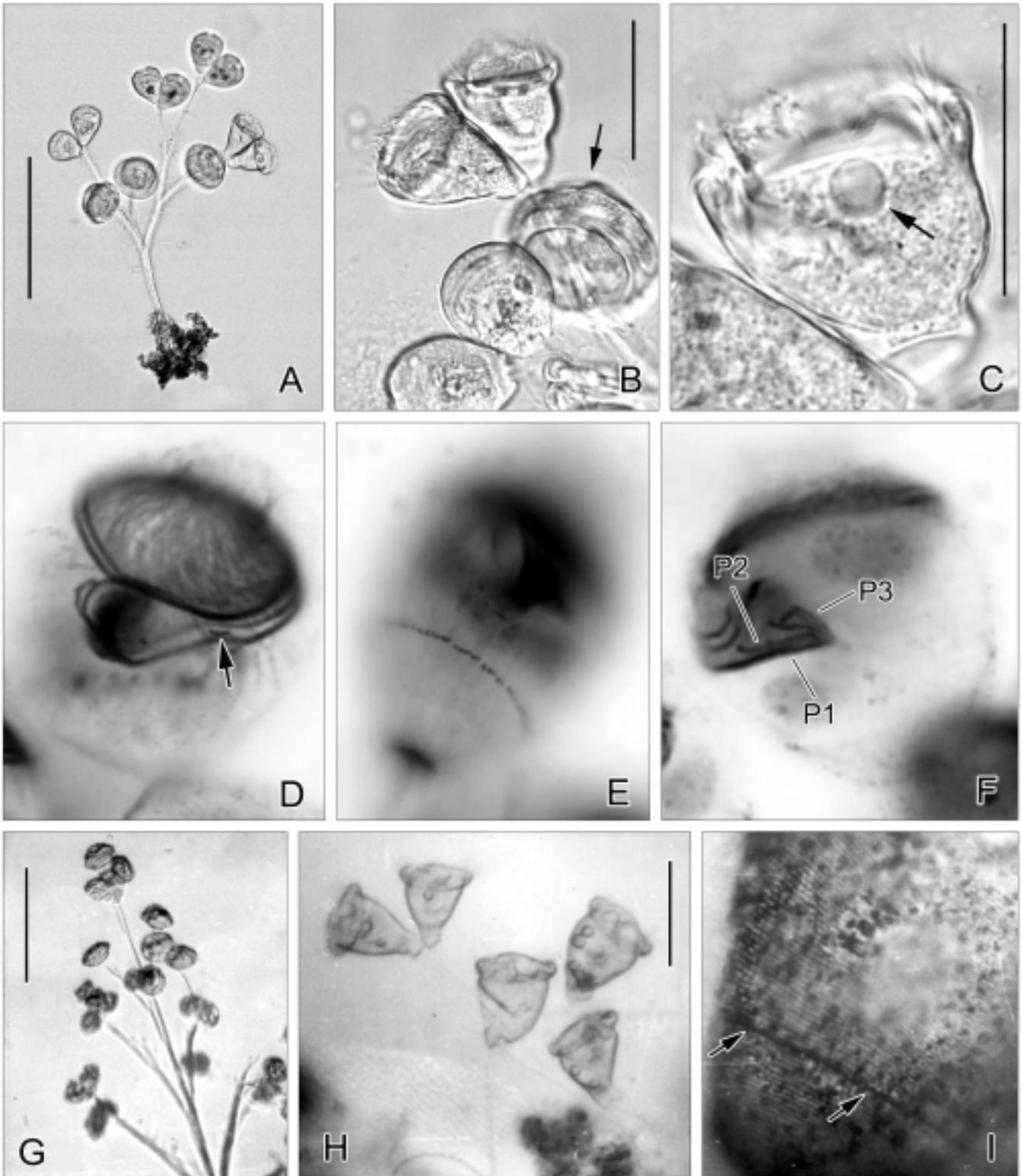
Cells highly sensitive to stimuli and contract readily. Also zooids readily convert to telotrochs, which are acetabuliform, *ca* $25 \mu\text{m}$ high and $45 \mu\text{m}$ in diameter (Figs 3E; 4B, arrow).

Colony *ca* $400 \mu\text{m}$ high, containing up to 40 zooids (Figs 3B, 4A); macrozooid formation was not observed. Stalk smooth, typically alternately branched, $6-9 \mu\text{m}$ in diameter. Spasmoneme $1.5-3 \mu\text{m}$ in diameter, mitochondria not observed.

Oral infraciliature as shown in Figs 3J, K and 4D, F. Haplo- and polykinety both make one and one-third turns of peristome before entering infundibulum, where they make a further turn. At adoral end of polykinety, a short kinety fragment is always recognizable (Fig. 3J, double arrowhead). Each of three infundibular polykineties consisting of three ciliary rows. Rows of P1 about equal in length, terminating at end of infundibulum. P2 about half length of P1. Inner two rows of P2 lie close to P1 while outer one is separated from the other two; all three



Figs 3A-K. *Zoothamnium sinense* Song, 1991 from life (A-I) and after protargol impregnation (J, K). **A** - a typical zooid; **B** - colony form; **C, D** - zooid (C) and colony (D), after Song (1991) (called *Z. commune*); **E** - telotroch; **F-H** - colony (F) and zooid (H), after Song (1986) (called *Z. truncatum*); **I** - zooid, lateral view; **J** - general oral infraciliature, arrow depicts the epistomial membrane, double arrowhead marks the distal kinety fragment; **K** - detail of infundibular polykineties. G - germinal kinety, H - haplokinety, P1-3 - infundibular polykinety 1-3, Po - polykinety. Scale bars: 20 μ m (3A, C, E, G-I); 150 μ m (3B, D, F).



Figs 4A-I. Photomicrographs of *Zoothamnium sinense* from life (A-C, G, H) and after protargol (D-F) and silver nitrate impregnation (I). **A** - a small colony; **B** - to show zooid shape and a developing telotroch (arrow); **C** - zooid at high magnification, to show the contractile vacuole (arrow); **D** - oral infraciliature, arrow marks the epistomial membrane; **E** - to show aboral trochal band; **F** - to show infundibular polykineties. **G** - photomicrograph of holotype of *Z. sinense* as described in Song (1991); **H** - photomicrograph of specimen from population described by Song (1991) (called *Z. commune*); **I** - silverline system of specimen from population described by Song (1991), arrows mark the aboral trochal band. P1-3 - infundibular polykinety 1-3. Scale bars: 150 μ m (4A, G); 50 μ m (4B, C, H).

rows of P2 end conspicuously above the posterior end of P1 (Figs 3J, K). P3 about 2/3 length of P2, anterior end considerably below those of P1 and P2 and terminates posteriorly above P1 but below P2. Abdomal half of inner row of P3 separated from the other two ciliary rows but converges with them adstomally (Figs 3J, K; 4F). Haplokinety passes around infundibulum on wall opposite to peniculi; germinal kinety runs parallel to haplokinety in abdomal half of infundibulum before terminating in mid-region of infundibulum (Fig. 3J). Epistomial membrane located near opening of infundibulum as commonly seen in other peritrichs (Figs 3J arrow; 4D, arrow).

Aboral trochal band appears as a row of loosely arranged dikinetids that encircle the body about 1/4 of the body length from the scopula (Fig. 4E).

Silverline system typical of genus, consisting of many parallel, transverse rows about 0.5 μm apart, *ca* 55-70 between peristomial area and aboral trochal band, *ca* 20-35 between aboral trochal band and scopula, and with sparsely distributed pellicular pores (Fig. 4I). Aboral trochal band stains heavily with silver, probably composed of two rows of closely arranged silverlines (Fig. 4I, arrows).

Remarks: *Zoothamnium sinense* was originally reported by Song (1986) under the name *Z. truncatum* (Figs 3F-H, 4G), and later renamed *Z. sinense* (Song 1991). According to the original description, *Z. sinense* has small body (33-59 \times 35-64 μm), an alternately branched stalk, a single-layered peristomial lip, a C-shaped macronucleus and a marine habitat, all of which correspond well with the present population (Song 1986).

Considering general living appearance, the following congeners should be compared with *Z. sinense*: *Z. alternans* Claparède et Lachmann, 1859, *Z. commune* Kahl, 1933 and *Z. plumula* Kahl, 1935 (Table 2). According to our present study, *Zoothamnium alternans* is similar to *Z. sinense* in zooid size, stalk branching style and habitat, but can be easily recognized from the latter by the presence of several large macrozooids (*vs.* absent in *Z. sinense*), its J-shaped macronucleus (*vs.* C-shaped in *Z. sinense*) and in having 40-55 silverlines between the peristome and the aboral trochal band (*vs.* 55-70 in *Z. sinense*).

Zoothamnium commune and *Z. plumula*, like *Z. sinense*, also possess alternately branching stalks, inverted bell-shaped zooids and marine habitats. However, both can be separated from *Z. sinense* by their distinctly larger body size (>50 μm *vs.* 36-48 μm long *in*

vivo in *Z. sinense*) and differences in the arrangement of the kineties of P3 (all three parallel in *Z. commune* and *Z. plumula* *vs.* innermost kinety separated and with a different orientation in *Z. sinense*) (Song *et al.* 2002). Furthermore, *Z. commune* has 38-43 silverlines between the aboral trochal band and the scopula (*vs.* 20-35 in *Z. sinense*).

Another marine *Zoothamnium*, which also has a small body size (33-61 μm long) and an alternately branched stalk, was reported by Song (1991) under the name *Z. commune* (Figs 3C, D; 4H). Considering the apparent difference from the original description of *Z. commune* by Kahl (1933) and the similarity with the current form, we consider that the population described by Song (1991) was misidentified and should be *Z. sinense*.

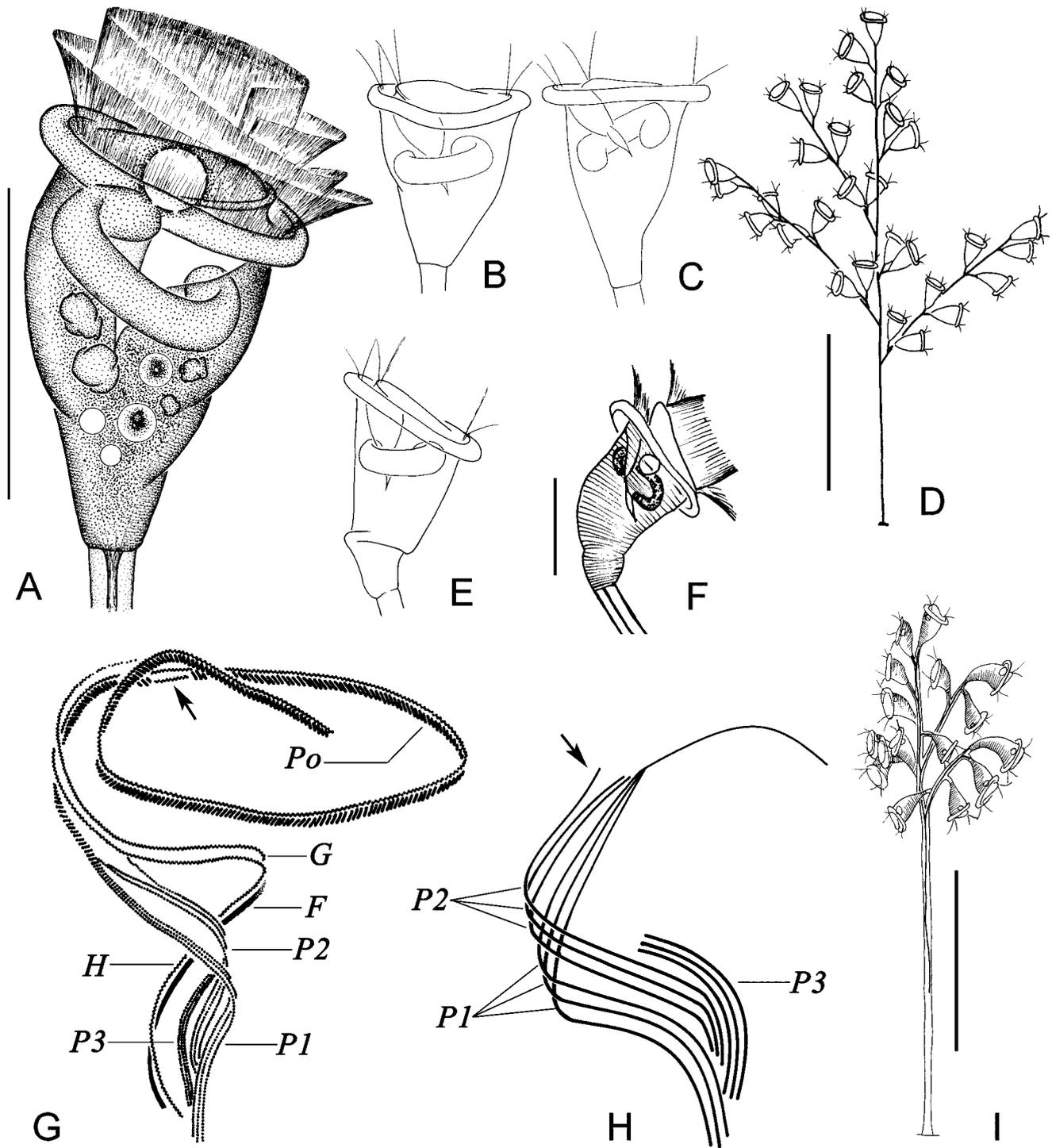
Redescription of *Zoothamnium commune* Kahl, 1933 (Figs 5, 6; Table 1)

Amended diagnosis: Marine *Zoothamnium* with alternately branched stalk. Zooids elongate, measuring about 60-104 \times 48-56 μm *in vivo*. Single contractile vacuole apically located; macronucleus C-shaped and transversely to obliquely oriented. About 59-70 silverlines between aboral trochal band and peristomial lip, and 38-43 between aboral trochal band and scopula. Three ciliary rows in the third infundibular polykinety (P3) lie parallel to each other; P3 about half-length of P2 and terminates posteriorly conspicuously above P1.

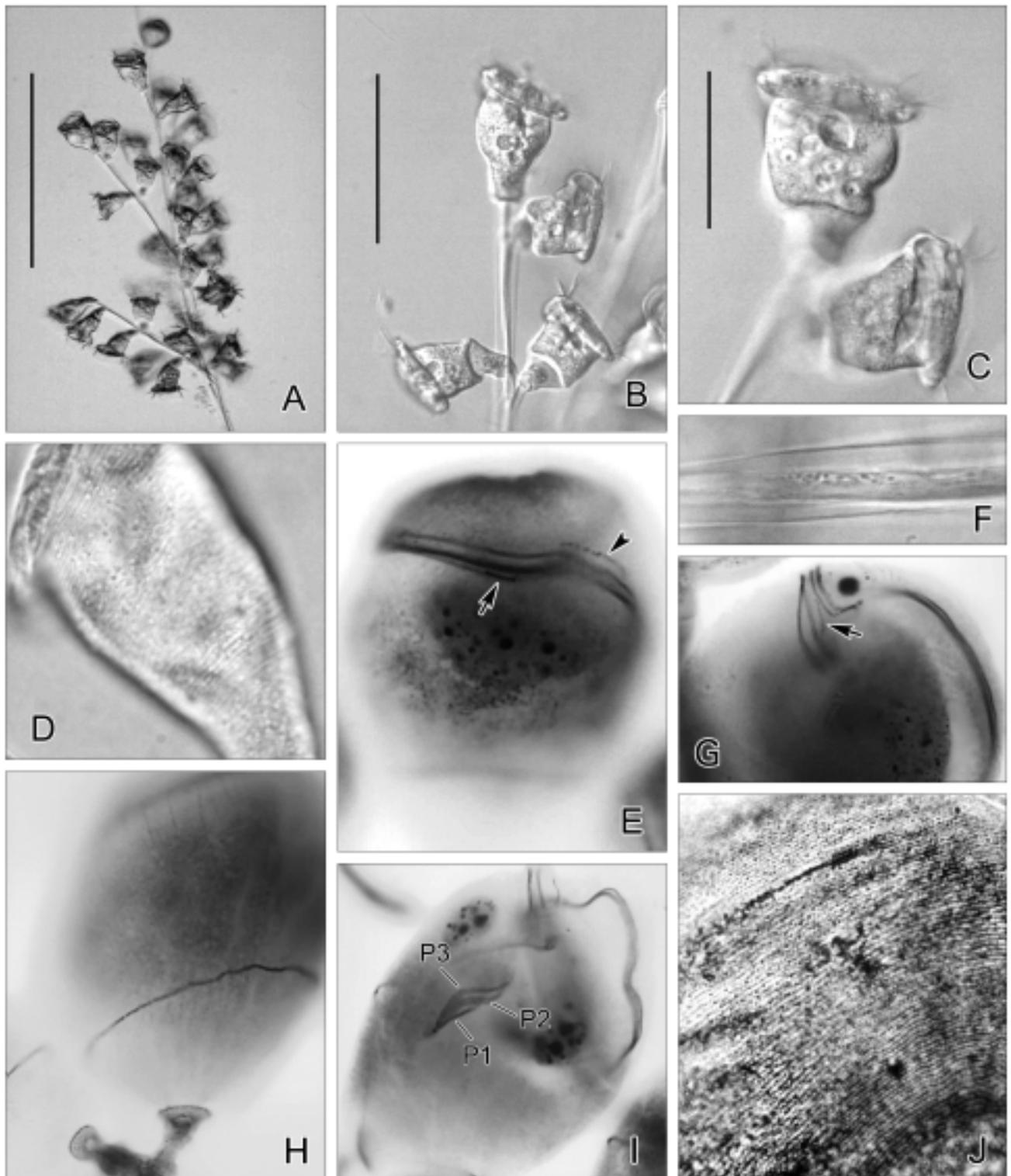
Amended description: Zooids elongate, inverted bell-shaped to conical; 60-104 \times 48-56 μm *in vivo* (Figs 5A-C, E, F; 6A-C). Body widest just below peristomial lip. Peristomial disc slightly elevated above peristomial lip and contains the single contractile vacuole. Macronucleus C-shaped, more or less transversely oriented in third of body length to oral area and surrounding adstomal portion of infundibulum. Cytoplasm colourless and transparent, usually with granules and several food vacuoles (Figs 5A; 6B, C).

Pellicle appears smooth at low magnification; fine pellicular striations observed only above $\times 400$ magnification (Fig. 6D). After staining with silver nitrate, many tiny pellicular pores visible along pellicular striations (Fig. 6J). Aboral trochal band not detectable in living zooids and only faintly stained with silver nitrate. About 59-70 pellicular striations between aboral trochal band and peristomial lip, and 38-43 between aboral trochal band and scopula.

Colony alternately branched, up to 1mm or more in height and contains up to 50 zooids (Figs 5D, I; 6A).



Figs 5A-I. *Zoothamnium commune* from life (A-F, I) and after protargol impregnation (G, H). **A** - a typical zooid at high magnification; **B**, **C**, **E** - shape variability; **D** - colony; **F** - zooid (after Kahl, 1933); **G** - oral infraciliature, arrow marks the epistomial membrane; **H** - detail of infundibular polykineties, arrow marks the separated anterior end of outer ciliary row in P2. **I** - colony (after Kahl, 1933). **F** - filamentous reticulum; **G** - germinal kinety, **H** - haplokinety, **P1-3** - infundibular polykinety 1-3, **Po** - polykinety. Scale bars: 50 μ m (5A); 300 μ m (5D, I); 30 μ m (5F).



Figs 6A-J. Photomicrographs of *Zoothamnium commune* from life (A-D, F), after protargol (E, G-I) and silver nitrate impregnation (J). **A** - colony form; **B**, **C** - zooids at low magnification; **D** - zooid at high magnification, to show pellicular striations; **E** - to show epistomial membrane (arrow) and germinal kinety (arrowhead); **F** - detail of stalk, to show mitochondria in spasmoneme; **G** - to show the separated anterior end of outer kinety in peniculus 2 (arrow); **H** - lateral view, to show the aboral trochal band; **I** - to show infundibular polykineties; **J** - silverline system. P1-3 - infundibular polykinety 1-3. Scale bars: 300 μm (A); 100 μm (B); 50 μm (C).

Stalk with smooth surface, 16 μm in diameter in main trunk and 10 μm in diameter at distal ends of branches. Spasmoneme 7 μm in diameter in main trunk and 4 μm in diameter at distal ends of branches, and with numerous dark mitochondria (Fig. 6F).

Oral infraciliature of *Z. commune* formed of peristomial and infundibular parts separated by the epistomial membrane (Figs 5G, H; 6E arrow, G., I). Peristomial part consisting of haplo- and polykinety, which are parallel to one another throughout their length and make one and one-quarter turns of the peristome before entering the infundibulum. Within infundibulum haplokinety and polykinety spiral on opposite walls and end at border of cytostome. Infundibular part of haplokinety accompanied by germinal kinety in adoral half (Figs 5G; 6E, arrowhead) and a filamentous reticulum in aboral half (Fig. 5G). Infundibular polykinety transforms into three polykineties at adstomal half, designated P1-3 (Figs 5G, H; 6I). P1 consists of three parallel rows of kinetosomes that extend to the edge of the cytostome. The three rows of P2 originate in mid-infundibulum, adjacent to point where the polykinety differentiates into P1, the outermost row being slightly separated from the other two at this point (Figs 5H arrow; 6G, arrow); P2 terminates between and above adstomal ends of P1 and P3 (Fig. 5H). P3 is about half the length of P2. It originates in aboral half of infundibulum and comprises three parallel ciliary rows that terminate slightly above those of P1 (Fig. 5H).

Aboral trochal band composed of a row of loosely arranged double kinetosomes that encircle the aboral quarter of the cell (Fig. 6H).

Remarks: The current population is identified as *Zoothamnium commune* based on the following: (1) the stalk is alternately branched; (2) accessory branches are unequal in length giving the colony an asymmetrical shape; (3) zooids are similar in shape; (4) marine habitat (Kahl 1933).

In terms of the stalk branching pattern, body shape and size, *Zoothamnium hentscheli* Kahl, 1935 and *Z. thiophilum* Stiller, 1946 are similar to *Z. commune* (Table 2), although their infraciliature and silverline system remain unknown. However, both of these taxa are freshwater forms unlike the Qingdao population which is marine (Kahl 1935, Stiller 1946).

Zoothamnium sinense also has an alternately branched stalk and marine habitat, but differs from *Z. commune* in having distinctly smaller size zooids (*ca* 36-48 \times 30-38 μm *vs.* 60-104 \times 48-56 μm).

Zoothamnium plumula Kahl, 1933 also possesses an alternately branched stalk and is marine, but differs from the Qingdao population in having enlarged zooids at the distal ends of the stalk branches (*vs.* absent) in *Z. commune* and fewer silverlines between scopula and aboral trochal band (24-28 *vs.* 38-43 in *Z. commune*) (Song *et al.* 2002).

In general, the present study contains three alternatively branched species, which have similar living appearance and can be easily confused with each other as well as related congeners. Their previous misidentifications indicate that the criterion for identification on *Zoothamnium* species should be adequately improved.

Based on our study, we suggest that the following characters should be considered for species identification/separation: The branching pattern, the length of the main trunk below the first ramification, zooid differentiation (micro-/macrozooid), zooid size, appearance of oral border (single/double-layered), shape of macronucleus, location of contractile vacuole, arrangement of the third oral polykinety, number of silverline and habitat.

Acknowledgements. This work was supported by the "Natural Science Foundation of China" (Project number: 30430090) and by the Darwin Initiative Programme (Project number: 14-015). The authors would like to thank Ms. Xiaofeng Lin, OUC, for her help with sampling.

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Received on 21st September, 2005; revised version on 3rd January, 2006; accepted on January 16th, 2006