

First Record of Two Urostyloid Ciliates (Spirotrichea: Urostylida: Urostyloidea) from Brackish Water in Korea

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ABSTRACT

Two urostyloid ciliates, collected from brackish water in Korea, were identified as *Diaxonella pseudorubra pseudorubra* (Kaltenbach, 1960) Berger, 2006 and *Pseudokeronopsis flava* (Cohn, 1866) Wirnsberger, Larsen and Uhlig, 1987. The description was based on living, protargol impregnated specimens. These species are described as follows: *Diaxonella pseudorubra pseudorubra*: body size *in vivo* 145-230 × 40-60 μm, elongated ellipsoidal in shape. Cytoplasm reddish and flexible. Adoral zone of membranelles occupied 30-40% of the body; composed of 33-44 membranelles; 1-3 frontoterminal cirri, 1-4 frontal row cirri, 4-6 buccal cirri, 6-10 transverse cirri. Midventral rows composed of 14-24 cirri, four left marginal rows, one right marginal row. Two kinds of cortical granules; the larger one is yellowish and the smaller one is reddish. *Pseudokeronopsis flava*: body size *in vivo* 150-210 × 30-45 μm, elongated ellipsoidal shape. Cytoplasm yellowish and flexible. Adoral zone of membranelles occupied 25-30% of body; composed of 44-58 membranelles in number. Frontal cirri forming bicorona composed of 5-7 cirral pairs, 2-3 frontoterminal cirri, one buccal cirrus, and 2-3 transverse cirri. Midventral rows composed of 18-33 cirri, 34-53 left marginal cirri, and 40-58 right marginal cirri. Two kinds of cortical granules; the larger one is colorless and “blood-cell-shaped,” and the smaller one is yellowish. *Diaxonella pseudorubra pseudorubra* is different from the most similar subspecies, *D. pseudorubra pulchra*, in cytoplasmic color and number of midventral cirri. *Pseudokeronopsis flava* is different from its most similar congeners in pigment granular color, number of bicorona, number of midventral cirri, and position of the contractile vacuole.

Keywords: *Diaxonella*, *Pseudokeronopsis*, hypotrichs, redescription, estuary, brackish water, morphology

INTRODUCTION

The urostyloid ciliates are composed of a large group of hypotrichs, *sensu lato*, which belong to the superfamily Urostyloidea, and more than 2,000 species have been described (Berger, 2006; Lynn, 2008). Since the superfamily Urostyloidea was established by Bütschli (1889), its definition has been improved as usually having an ellipsoidal, middle to large sized body and a midventral complex between the marginal rows (Corliss, 1979; Lynn and Small, 2002; Berger, 2006; Lynn, 2008). The systematics of genus *Diaxonella* has a comparatively complex history, including many synonyms (Jankowski, 1979; Foissner, 1987; Oberschmidleitner and Aescht, 1996; Berger, 2006; Shao et al., 2007). *Diaxonella pseudorubra* (Kaltenbach, 1960) was a monotypic species of the genus *Diaxonella*, but three subspecies, *D. pseudoru-*

bra pseudorubra, *D. pseudorubra polystylata*, and *D. pseudorubra pulchra*, have been recognized recently by Berger (2006). The genus *Pseudokeronopsis* includes 10 species and is usually found in marine environments (Song et al., 2002, 2004, 2006). Most *Pseudokernopsis* species have cortical granules colored red, yellow, or orange-yellow, which can be used to distinguish the species. In this study, we provide morphological redescrptions and variations in two urostyloids from Korea.

MATERIALS AND METHODS

Sample collection and enrichment

Diaxonella pseudorubra pseudorubra and *Pseudokeronopsis flava* were collected from brackish waters in Korea. *Diaxo-*

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nella pseudorubra pseudorubra: (October, 2010; salinity 2‰) downstream of the sewage treatment plant (N34°55'23", E128°07' 07") located in Sadeung-dong, Sacheon-si, Gyeong-sangnam-do. The water samples including the ciliates were collected with twigs, leaves, wood, and mud. *P. flava*: (January, 2010; salinity 20‰) in the Soesokkak estuary (N33° 15'7.19", E126°37'24.59"), Seogwipo-si, Jeju-do. The specimens were transferred to a Petri dish (87 mm in diameter) or a cell culture dish (150 mm in diameter). These ciliates have been cultured at room temperature in the laboratory with collected water, enriched with dried wheat grain to supply fungal and bacterial nutrients

Morphological observation

The morphology of living specimens was observed under low (50-400×) and high (1,000×; immersion oil) magnifications using a light microscope with a DIC device (Axio Imager A1; Carl Zeiss, Oberkochen, Germany) and their images were captured using a CCD camera (Axio Cam MRC; Carl Zeiss). The infraciliatures were observed after impregnation using the protagol method (Wilbert, 1975; Foissner, 1992). Terminology and taxonomic classification are accord-

ing to Berger (2006) and Lynn (2008).

SYSTEMATIC ACCOUNTS

Order Urostyleida Jankowski, 1979
 Superfamily Urostyleoidea Bütschli, 1889
 Family Holostichidae Fauré-Fremiet, 1961
 Genus ^{1*}*Diaxonella* Jankowski, 1979

^{2*}*Diaxonella pseudorubra pseudorubra*
 (Kaltenbach, 1960) (Tables 1, 2, Figs. 1-3)

Keronopsis pseudorubra Kaltenbach, 1960 (cited from Berger, 2006: 470).

Keronopsis rubra Jerka-Dziadosz and Janus, 1972: 249.

Diaxonella trimarginata Jankowski, 1979: 83; Oberschmidleitner and Aescht, 1996: 21; Shao et al., 2007: 25.

Holosticha pseudorubra: Foissner, 1987: 225; Berger, 2001: 44.

Diaxonella pseudorubra pseudorubra: Berger, 2006: 468.

Description. General morphology and behavior: Body

Table 1. Morphometric data of *Diaxonella pseudorubra pseudorubra*

Characteristics	Method	Mean	Min	Max	Med	SD	SE	CV	n
Body length (μm)	L	171.5	145	230	167.5	22.0	4.9	12.8	20
	S	149.7	117	180	151.5	18.7	4.2	12.5	20
Body width (μm)	L	47.8	40	60	49.0	5.4	1.2	11.4	20
	S	55.6	42	67	56.0	7.4	1.7	13.4	20
Body length/Body width	L	3.6	3	4	3.7	0.4	0.1	10.1	20
	S	2.7	2	3	2.7	0.3	0.1	10.6	20
AZM, length (μm)	L	56.5	50	75	55.5	5.5	1.2	9.8	20
	S	61.9	54	71	61.5	5.0	1.1	8.1	20
Body length/length of AZM	L	3.0	2	4	3.0	0.3	0.1	11.4	20
	S	2.4	2	3	2.4	0.2	0.0	6.8	20
AMs, number	S	38.5	33	44	38.0	3.2	0.7	8.2	20
BC, number	S	5.3	4	6	5.0	0.6	0.1	10.8	20
FC, number	S	4.1	3	5	4.0	0.4	0.1	9.7	20
FTC, number	S	2.0	1	3	2.0	0.4	0.1	17.7	17
FRC, number	S	3.6	1	4	4.0	0.9	0.2	25.3	15
TC, number	S	7.9	6	10	8.0	1.0	0.2	13.2	20
PTC, number	S	2.0	2	2	2.0	0.0	0.0	0.0	20
MVC (pair), number	S	19.1	14	24	19.0	2.9	0.7	15.0	15
RMC, number	S	41.4	30	50	42.0	5.6	1.3	13.4	17
LMC ₁ , number	S	30.1	21	38	29.0	4.8	1.3	16.1	15
LMC ₂ , number	S	25.7	20	33	25.0	3.5	0.9	13.6	15
LMC ₃ , number	S	21.6	18	28	20.0	3.2	0.8	14.9	15
LMC ₄ , number	S	11.1	4	15	12.0	3.3	0.8	29.5	15
DKs, number	S	3.3	3	4	3.0	0.5	0.1	14.5	13

Min, minimum; Max, maximum; Med, median; CV, coefficient of variation in %; n, population size; L, live specimens; S, stained specimens; AZM, adoral zone of membranelles; BC, buccal cirrus; FC, frontal cirri; FTC, frontoterminal cirri; FRC, frontal row cirri; TC, transverse cirri; PTC, pretransverse cirri; MVC, midventral cirri; RMC, right marginal cirri; LMC, left marginal cirri; DK, dorsal kineties.

Korean name: ^{1*}쌍열충속, ^{2*}붉은쌍열충

Table 2. Comparisons of *Diaxonella pseudorubra pseudorubra* which has different names

Characters	<i>Diaxonella pseudorubra pseudorubra</i>	<i>D. trimarginata</i>	<i>D. trimarginata</i>	<i>Keronopsis rubra</i>
Body length <i>in vivo</i> (μm)	145-230	120-180	95-185	–
Body length (μm)	117-180	93-198	108-200	130-170
AMs, number	33-44	31-40	31-42	ca. 35
BC, number	4-6	5-7	5-8	ca. 5
FC, number	3-5	2-3	Constantly, 4	ca. 3
FTC, number	1-3	2-3	1-4	–
FRC, number	1-4	4-6	3-5	3-4
TC, number	6-10	6-9	7-10	ca. 10
MVC (pairs), number	14-24	18-27	16-29	ca. 34*
RMC, number	30-50	34-54	27-51	40-55
LMR, number	Constantly 4	3-4	Constantly, 4	ca. 2
PTC, number	Constantly 2	3-4	0-4	–
Mi, shape	Spherical	Ellipsoidal	Ovoid	–
Mi, size (diameter, μm)	ca. 2.5	ca. 2.5	ca. 5	–
UM, intersecting point	Middle	Middle	Posterior	Middle
Position of the last MVC	Posterior half	Posterior	Posterior half	Posterior half
Canals in the CV	Absent	Present	Absent	–
Data source	Present study	Oberschmidleitner and Aeschl, 1996	Shao et al., 2007	Jerka-Dziadosz and Janus, 1972

All data are based on protargol-impregnated specimens. AMs, adoral membranelles; BC, buccal cirri; FC, frontal cirri; FTC, frontoterminal cirri; FRC, frontal row cirri; TC, transverse cirri; MVC, midventral cirri; RMC, right marginal cirri; LMR, left marginal row; PTC, pretransverse ventral cirri; Mi, micronucleus; UM, undulating membranes; CV, contractile vacuole.

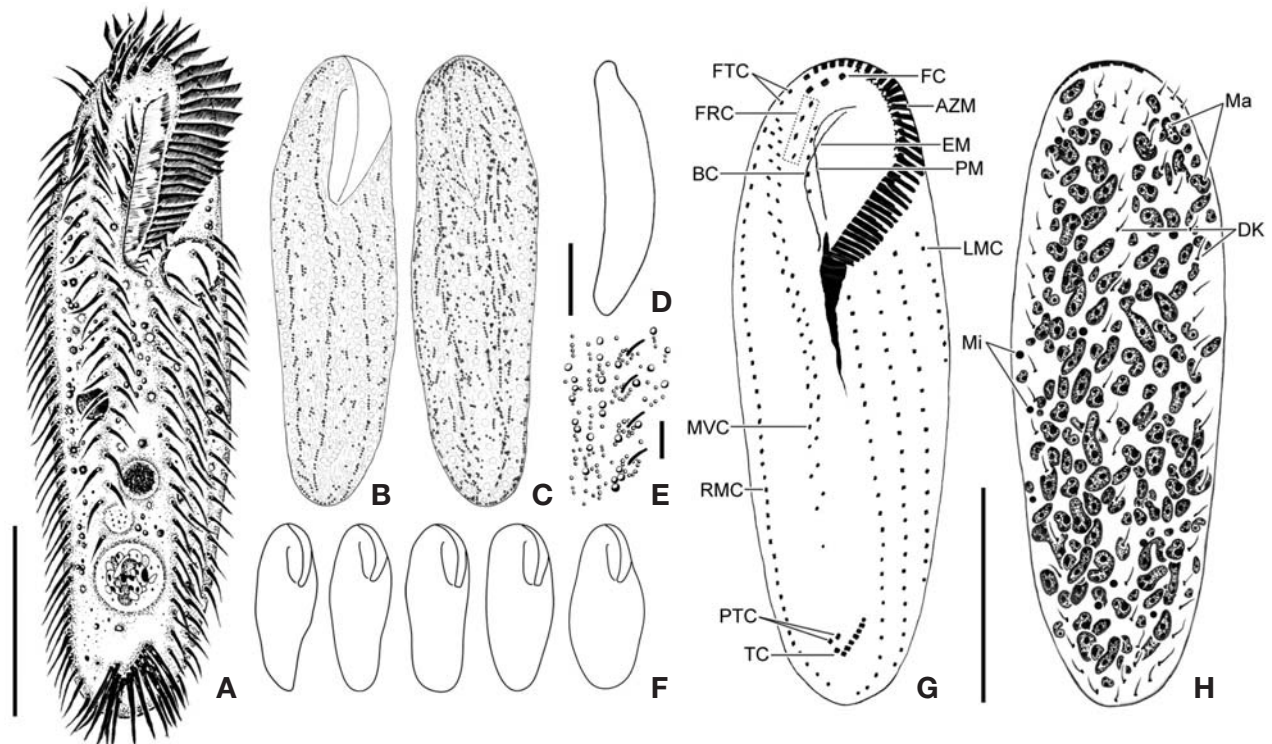


Fig. 1. *Diaxonella pseudorubra pseudorubra* from live (A-F) and impregnated specimens (G, H). A, Ventral view of a typical individual; B, C, Arrangement of cortical granules on ventral and dorsal sides; D, Flattened lateral view; E, Two kinds of cortical granules on dorsal side; F, Various body shapes; G, Somatic and oral infraciliature of ventral side; H, Dorsal kineties and nuclear apparatus. AZM, adoral zone of membranelles; BC, buccal cirrus; DK, dorsal kineties; EM, endoral membrane; FC, frontal cirri; FRC, frontal row cirri; FTC, frontoterminal cirri; LMC, left marginal cirri; Ma, macronuclear nodules; Mi, micronuclei; MVC, midventral cirri; PM, paroral membrane; PTC, pretransverse cirri; RMC, right marginal cirri; TC, transverse cirri. Scale bars: A, D, G=50 μm, E=5 μm.

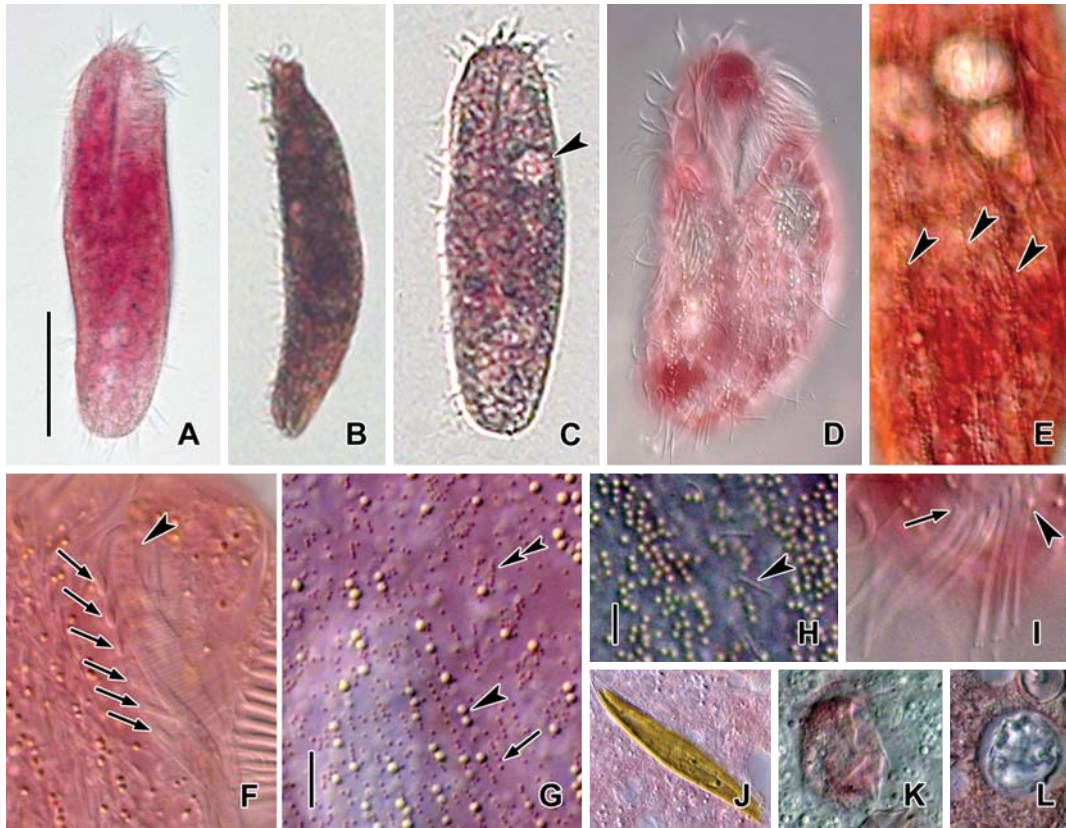


Fig. 2. Photomicrographs of *Diaxonella pseudorubra pseudorubra* from live specimens. A, Ventral view of a typical individual; B, Flattened lateral view; C, Contractile vacuole (arrowhead); D, Somatic ciliature of the ventral side; E, Arrangement of cortical granules on the ventral side (arrowheads); F, Buccal cirri (arrows) and undulating membranes (arrowhead); G, Patterns of cortical granules: larger (arrowhead) and smaller (arrow), arrangement around dorsal bristle of smaller cortical granules (double arrowhead); H, Dorsal bristles (arrowhead); I, Pretransverse cirri (arrow) and transverse cirri (arrowhead); J-L, Various food vacuoles; J, Diatom *Pleurosigma* sp.; K, *Aspidisca* sp.; L, Small particle of wheat grain. Scale bars: A=50 μ m, G, H=5 μ m.

size 145-230 \times 40-60 μ m, usually about 170 \times 40 μ m, length: width ratio about 4 : 1 on average in live specimens. Body shape elongated and ellipsoid with rounding at both ends (Figs. 1A, 2A, 3A), dorsoventrally flattened about 2 : 1, ventral side slightly concave, dorsal side convex (Figs. 1D, 2B). Cytoplasm reddish to wine color and flexible (Fig. 2D). Single contractile vacuole spherical and above the mid-body near the left margin (Figs. 1A, 2C). Locomotion usually crawling on substrate. Omnivorous feeding (Fig. 2J-L).

Buccal field and oral infraciliature: Adoral zone of membranelles occupies 30-40% of body length (Figs. 1G, 3A), distal to proximal continuously semicircular, consists of 33-44 adoral membranelles (Fig. 3B), widest membranelle about 10 μ m in length. Buccal area narrow and rather deep. Undulating membranes intersecting to half of the membranes, endoral membrane slightly curved and longer, paroral membrane anteriorly curved and shorter, distal end of paroral beyond endoral, about 45 μ m in total length (Figs. 1G, 3B).

Somatic infraciliature: Usually, four frontal cirri slightly enlarged, lying on front of the anterior region but one frontal cirrus near the distal end of the adoral zone of membranelles (Figs. 1G, 3C), about 17 μ m long. One to three frontoterminal cirri located on the right side of frontal cirri, about 12 μ m long (Fig. 3D). Four to six buccal cirri arranged along paroral membrane, about 8 μ m long (Figs. 2F, 3B). Inconspicuous, usually four frontal row cirri starting at the same level at the distal end of the paroral membrane and terminating at the same level at intersecting point of the undulating membranes (Figs. 1G, 3E). Midventral complex composed of 14-24 pairs of midventral cirri, continued frontoterminal cirri, arranged in a zigzag pattern, terminating at the posterior half but most cirri separated from others (Figs. 1G, 3G). Six to ten transverse cirri arranged in a J-shape, about 18 μ m long (Figs. 2I, 3H). Two pretransverse cirri located near right transverse cirrus (Fig. 3H). One right marginal row consisting of 30-50 cirri (Figs. 1G, 3G). Four left marginal rows that gradually

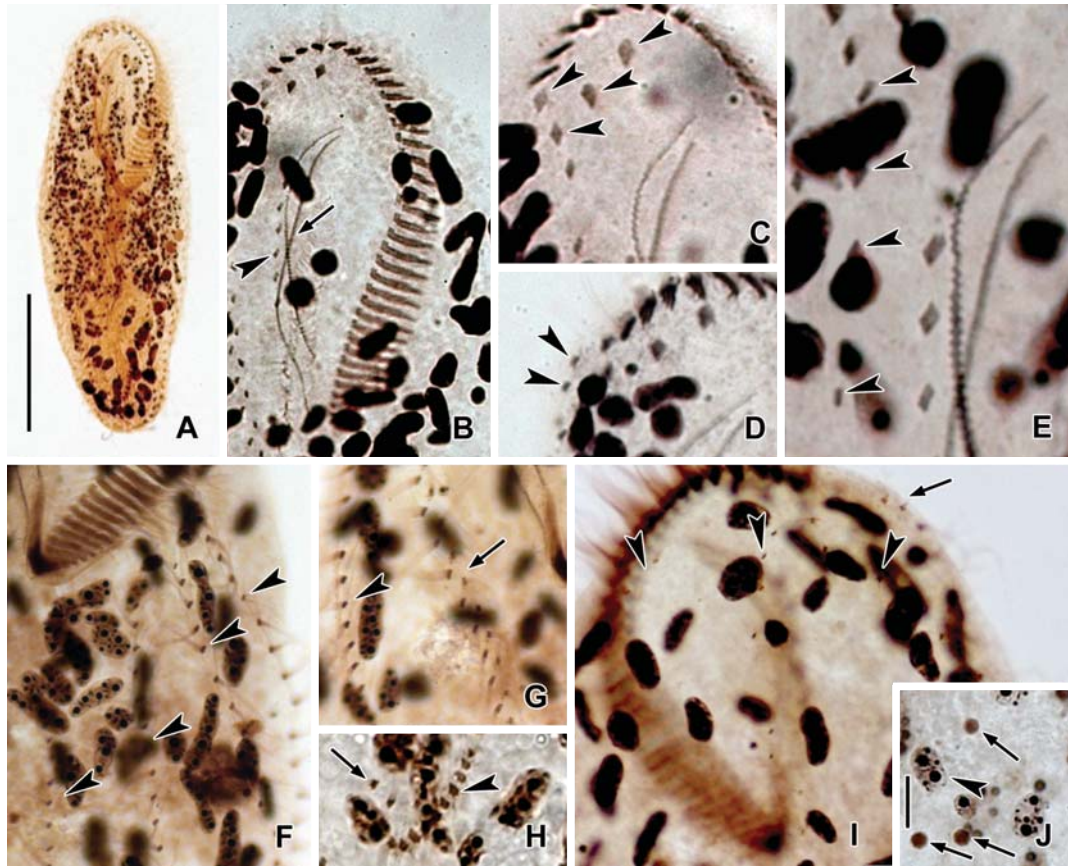


Fig. 3. Photomicrographs of *Diaxonella pseudorubra pseudorubra* from impregnated specimens. A, Ventral view of a typical individual; B, Buccal cirri (arrowhead) and undulating membranes (arrow); C, Four frontal cirri (arrowheads); D, Two frontoterminal cirri (arrowheads); E, Frontal row cirri (arrowheads); F, Four left marginal rows (arrowheads); G, Right marginal row (arrowhead) and midventral complex (arrow); H, Pretransverse cirri (arrow) and transverse cirri (arrowhead); I, Dorsal kineties (arrowheads) and extra dorsal bristles (arrow); J, Macronuclear (arrowhead) and micronuclei (arrows). Scale bars: A=50 μ m, J=10 μ m.

shorten from right to left composed of 21-38, 20-33, 18-28, and 4-15 cirri, respectively (Figs. 1G, 3F). Three dorsal kineties complete, but some cases of extra dorsal bristles present, bristle about 3-4 μ m in length (Figs. 1H, 2H, 3I).

Cortical granules: Two kinds of cortical granules present on both sides and pigmented: larger one greenish yellow about 1 μ m in diameter, arranged linearly in short groups beside the cirral rows on the ventral side and densely arranged on the dorsal side but loosely around each kineties (Fig. 1B, C). Smaller one red wine colored, about 0.3 μ m in diameter, scattered around the whole body and arranged linearly in short groups between larger granules or around each cirrus (Figs. 1E, 2G).

Nuclear apparatus: Various shaped which elongated ellipsoidal to ovoid macronuclear nodules throughout the whole body, about 120 in number with several nucleoli. Spherical shaped micronuclei scattered body and 2.5 μ m in diameter

(Figs. 1H, 3A, J).

Distribution. Europe (Austria, Germany, and Poland), Africa (Burundi) and Asia (China, Korea [present study]).

Remarks. The taxonomy of the genus *Diaxonella* is relatively complicated. According to a review by Berger (2006) on this subspecies, Kaltenbach (see Berger, 2006) described original population that had two frontal rows, a reddish body color, several buccal cirri, and one left marginal row. Berger (2006) considered *Keronopsis pseudorubra* sensu Kaltenbach in 1960 as a senior synonym of *Diaxonella trimarginata* sensu Jankowski (1979) which was renamed as a new species and new genus based on *Keronopsis rubra* sensu Jerka-Dziadosz and Janus (1972), because they misidentified it. Furthermore, he changed the name from *Keronopsis pseudorubra* to *Diaxonella pseudorubra*. Berger (2006) also suggested that the genus *Diaxonella* is monotypic and divided *D. pseudorubra* into three subspecies, which are distinguished by body color,

number of midventral pairs, and habitat. However, Shao et al. (2007) did not mention this suggestion of Berger (2006), because their publications overlapped in time.

Consequently, this Korean population of *Diaxonella pseudorubra pseudorubra* agrees with the original description except in the number of left marginal rows (4 vs. 1) and frontal cirri (3-5 vs. forming bicorona), which were intensively considered by Berger (2006). Additionally, our population agrees well with subsequent redescrptions of Austrian, Chinese, and Polish populations (Jerka-Dzidosz and Janus, 1972; Jankowski, 1979; Oberschmidleitner and Aescht, 1996; Shao et al., 2007). The Korean population differs slightly from the Austrian population of *D. pseudorubra pseudorubra* in shape of micronuclei (spherical vs. ellipsoidal), number of frontal cirri (3-5 vs. 2-3), canals in the contractile vacuole (absent vs. present), number of pretransverse cirri (2 vs. 3-4), and position of the last midventral cirrus (posterior half vs. posterior) (Oberschmidleitner and Aescht, 1996). Furthermore, the Korean population differs from the Chinese population at the intersecting point (middle vs. posterior) of the undulating membranes, the arrangement of reddish small cortical granules (linearly grouped vs. sparsely scattered), and the shape and diameter of micronuclei (spherical, about 2.5 μm vs. ovoid, about 5 μm) (Shao et al., 2007). Moreover, several populations (Europe, Asia, and Africa) of *D. pseudorubra pseudorubra* have been found in freshwater but this Korean population was recovered from brackish water of an estuarine

littoral zone (Berger, 2006).

Family Pseudokeronopsidae Borror & Wicklow, 1983

Genus *Pseudokeronopsis* Borror & Wicklow, 1983

¹**Pseudokeronopsis flava* (Cohn, 1866)

(Tables 3, 4, Figs. 4-6)

Oxytricha flava Cohn, 1866: 288.

Holosticha flava Kent, 1882: 769.

Keronopsis rubra Kahl, 1932: 571.

Pseudokeronopsis flava: Wirnsberger et al., 1987: 79; Berger, 2001: 55; Song et al., 2004: 1137; Sun and Song, 2005: 81; Berger, 2006: 940; Song et al., 2006: 272.

Description. General morphology and behavior: Body size 150-210 \times 30-45 μm , usually about 180 \times 40 μm , length : width ratio about 5 : 1 in live specimens. Body shape elongated elliptical, both ends narrowly rounded, anterior portion usually concave leftwards (Figs. 4A, 5A), dorsoventrally flattened about 2 : 1 (Fig. 5B). One contractile vacuole located below the mid-body near the left cell margin about 13 μm in diameter (Figs. 4A, 5E). Cytoplasm very flexible but not contractile (Fig. 5C), almost yellowish at low magnification. Locomotion usually crawling on substrate, wrapping to change direction (Fig. 5C). Omnivorous feeding (Fig. 5J, K).

Buccal field and oral infraciliature: Adoral zone of membranelles occupies 25-30% of body length, composed of 44-

Table 3. Morphometric data of *Pseudokeronopsis flava*

Characteristics	Method	Mean	Min	Max	Med	SD	SE	CV	n
Body length (μm)	L	182.2	150	208	184.7	17.3	3.9	9.5	20
	S	190.2	133	268	183.9	35.4	6.5	18.6	30
Body width (μm)	L	38.2	31	44	37.6	4.0	0.9	10.4	20
	S	43.9	23	70	44.0	15.2	2.8	34.6	30
Body length/Body width	L	4.8	4	5	4.8	0.4	0.1	8.8	20
	S	4.7	3	7	4.4	1.0	0.2	22.4	30
AZM, length (μm)	L	49.3	40	59	49.1	5.2	1.2	10.5	20
	S	56.0	43	77	57.1	8.3	1.5	14.9	30
Body length/length of AZM	L	3.7	3	5	3.6	0.5	0.1	12.9	20
	S	3.4	3	5	3.3	0.4	0.1	12.0	30
AMs, number	S	49.0	44	58	48.5	3.1	0.6	6.3	30
FC (Bicorona), number	S	5.8	5	7	6.0	0.6	0.1	10.0	27
BC, number	S	1.0	1	1	1.0	0.0	0.0	0.0	29
FTC, number	S	2.0	2	3	2.0	0.2	0.0	9.8	25
TC, number	S	3.1	2	4	3.0	0.4	0.1	11.9	30
MVC (pair), number	S	25.0	18	33	24.5	3.5	0.7	14.2	26
LMC, number	S	40.8	34	53	40.0	5.7	1.1	13.9	27
RMC, number	S	48.6	40	58	48.0	5.0	0.9	10.2	28
DKs, number	S	3.1	3	4	3.0	0.4	0.1	11.2	15

Min, minimum; Max, maximum; Med, median; CV, coefficient of variation in %; n, population size; L, live specimens; S, stained specimens; AZM, adoral zone of membranelles; FC, frontal cirri; Bicorona, FC arranged in arcs forming cirri; BC, buccal cirrus; FTC, frontoterminal cirri; TC, transverse cirri; MVC, midventral cirri; LMC, left marginal cirri; RMC, right marginal cirri; DK, dorsal kineties.

Korean name: ¹*노랑위각모충

Table 4. Comparisons of different populations of *Pseudokeronopsis flava*

Characters	<i>Pseudokeronopsis flava</i>	<i>P. flava</i>	<i>P. flava</i>
Body length <i>in vivo</i> (μm)	150-210	120-210	140-350
Body length (μm)	135-270	—	—
AMs, number	44-58	43-59	43-51
MVC (pairs), number	18-33	25-32	24-36
TC, number	2-4	1-3	3-4
UM, intersecting point	Mid part of membranes	Absent	Posterior end of membranes
EM, length	Similar to PM	Shorter than PM	Similar to PM
Data sources	Present study	Wirnsberger et al., 1987	Song et al., 2004

All data are based on protargol-impregnated specimens. AMs, adoral membranelles; MVC, midventral cirri; TC, transverse cirri; UM, undulating membranes; EM, endoral membrane; PM, paroral membrane.

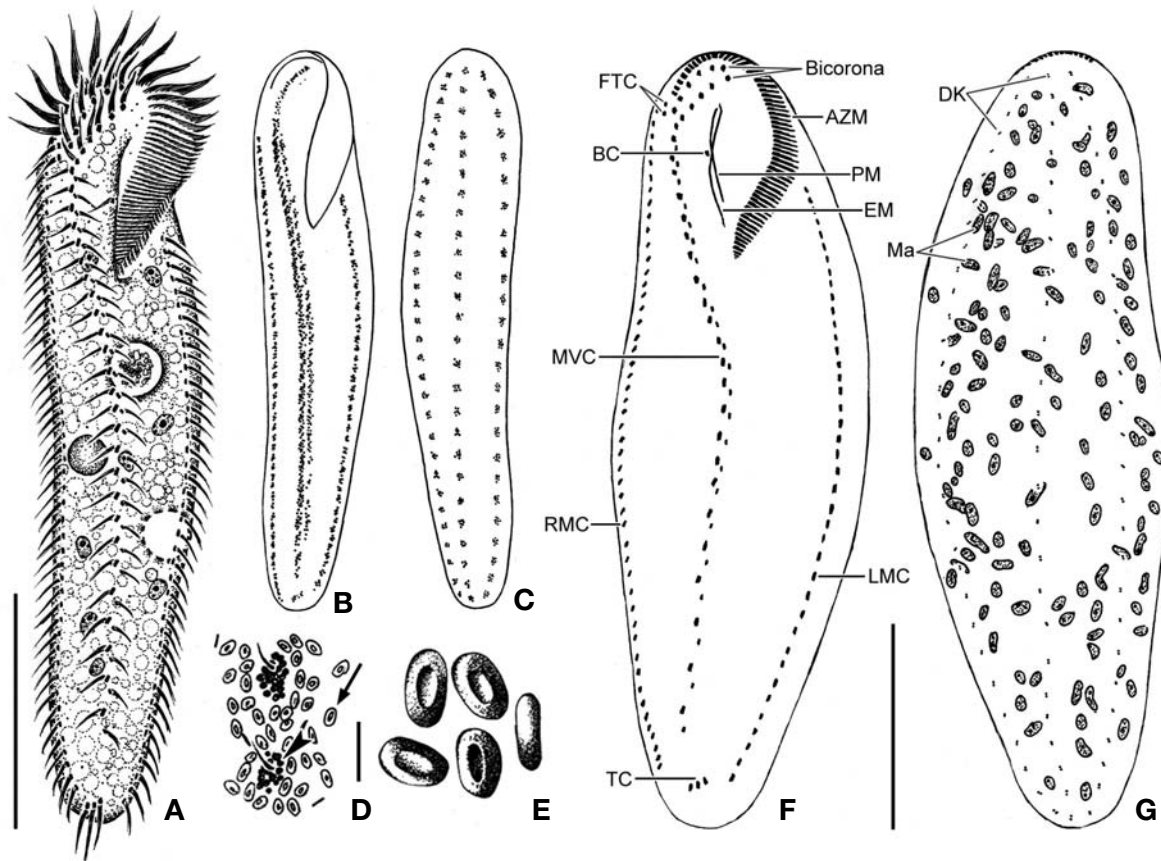


Fig. 4. *Pseudokeronopsis flava* from live (A-E) and impregnated specimens (F, G). A, Ventral view of a typical individual; B, Arrangement of cortical granules on the ventral side; C, Three cortical granular rows on the dorsal side; D, E, Cortical granule groups (arrowhead) and "blood-cell-shaped" granules (arrow) apparatus; F, Somatic and oral infraciliature of the ventral side; G, Three dorsal kineties and nuclear apparatus. AZM, adoral zone of membranelles; BC, buccal cirrus; Bicolorona, FC arranged in arcs forming cirri; DK, dorsal kineties; EM, endoral membrane; FTC, frontoterminal cirri; LMC, left marginal cirri; Ma, macronuclear nodules; MVC, midventral cirri; PM, paroral membrane; RMC, right marginal cirri; TC, transverse cirri. Scale bars: A, G=50 μm , E=2 μm .

58 adoral membranelles (Figs. 4F, 6A). Buccal area narrow and rather deep. Paroral and endoral membranes slightly curved, intersecting to half of the membranes, endoral longer than paroral, about 35 μm in length (Figs. 4F, 6A, D).

Somatic infraciliature: Frontal cirri arranged in two arcs

forming bicorona comprising 5-7 cirral pairs, bicorona connect to midventral complex (Fig. 6E). Midventral complex extended to subposterior and consisting of 18-33 cirri; distance between posterior most cirrus of midventral cirri and uppermost transverse cirrus about 20 μm (Figs. 4F, 6A). Two or



Fig. 5. Photomicrographs of *Pseudokeronopsis flava* from live specimens. A, Ventral view of a typical individual; B, Flattened lateral view; C, Showing flexible body; D, Dorsal kineties (arrowheads); E, Contractile vacuole (arrowhead); F, Arrangement of cortical granule on ventral side; G, H, Cortical granule groups (arrowheads) and "blood-cell-shaped" granules (arrow) apparatus on dorsal side; I, Three transverse cirri (arrowhead); J, K, Various food vacuoles; J, One kind of Chlorophyta; K, *Coleps* sp. Scale bar: A=50 μ m.

three frontoterminal cirri located at the right side of the right-most bicorona pair (Fig. 6C, E). Buccal cirrus located near the intersecting point of the undulating membranes (Fig. 6D). Two to four transverse cirri located posteriorly and distinctly

separated by marginal rows (Figs. 5I, 6F). Right marginal row commenced at level of the down most bicorona cirrus, terminated at the posterior part and comprising 40-58 cirri (Figs. 4F, 6A, E), left marginal row with 34-53 cirri; posterior

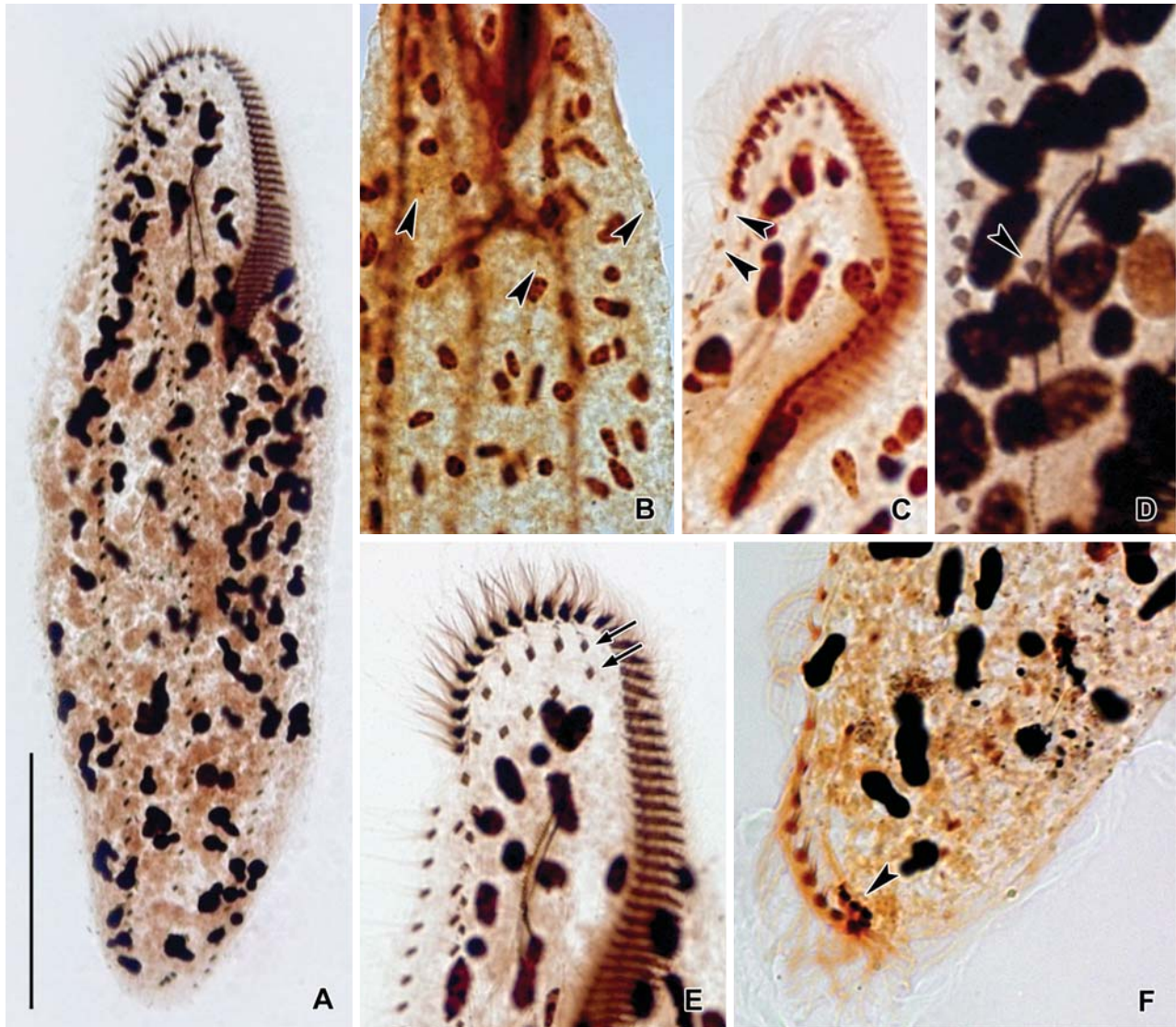


Fig. 6. Photomicrographs of *Pseudokeronopsis flava* from impregnated specimens. A, Ventral view of a typical individual; B, Dorsal kineties (arrowheads); C, Two frontoterminal cirri (arrowheads); D, Buccal cirrus (arrowhead) and undulating membranes; E, Infraciliature of bicorona (arrows); F, Three transverse cirri (arrowhead). Scale bar: A=50 μ m.

ends of marginal rows distinctly separated (Figs. 4F, 6A). Dorsal bristles length 5-10 μ m (Fig. 5D), three arranged kineties, dorsal kineties extending to entire dorsal surface (Figs. 4G, 6B).

Cortical granules: Two types of cortical granules present on both sides (Figs. 4D, 5H); yellowish cortical granules forming four rows with cirral rows on ventral side (Figs. 4B, 5F), small groups with dorsal bristles on dorsal side and 0.8-1 μ m in diameter (Figs. 4C, 5G, H), colorless cortical granules shaped ellipsoidal and “blood-cell-shaped” under both cortex and about 2 \times 1.5 μ m in size (Fig. 5G, H).

Nuclear apparatus: 70-100 ovoid to ellipsoidal macronuclear

nodules, size 5-10 μ m long in protargol-impregnated specimens. Micronucleus inconspicuous, about 3 μ m in diameter (Figs. 4G, 6A).

Distribution. Europe (England, France, Denmark, Germany, Italy, and Poland), North America (USA), and Asia (China and Korea [present study]).

Remarks. The Korean population of *Pseudokeronopsis flava* closely matched the original and subsequent redescriptions. However, the Korean population differs from the other populations in the pattern of undulating membranes. The undulating membranes of the Korean population usually have one intersecting point at the mid part, whereas those of the Chinese

population have no intersecting point at the mid part. The length of the endoral membrane is usually longer than that of the paroral in the Korean population, whereas that of both membranes is equal in the Chinese population. However, the lengths of the endoral and paroral membranes of the Danish population are similar to those of the Korean population (Cohn, 1866; Song et al., 2004, 2006; Sun and Song, 2005; Berger, 2006). The size of the pigment granules is slightly different in the Korean and Chinese populations (0.8-1 μm vs. 0.5 μm) (Song et al., 2004, 2006).

Closely-related species of *Pseudokeronopsis flava* are *P. carnea*, *P. flavicans*, and *P. rubra*, but they can be distinguished from each other as follows. *Pseudokeronopsis flava* is different from *P. carnea* in terms of pigment granule color (yellow vs. red), number of bicorona (5-7 vs. 9-11), number of midventral cirri (ca. 25 vs. ca. 40), number of transverse cirri (2-4 vs. 8-9), number of dorsal kineties (3 vs. 7-8), and number of adoral membranelles (ca. 50 vs. ca. 70). *Pseudokeronopsis flava* is different from *P. flavicans* in posterior body shape (slightly narrow vs. distinctly narrow), position of the contractile vacuole (posterior half vs. anterior half), number of dorsal kineties (3 vs. 5), and the gap between the posterior of the midventral row and transverse cirri (wide vs. narrow). *Pseudokeronopsis flava* is different from *P. rubra* in pigment granule color (yellow vs. red) and number of dorsal kineties (3 vs. usually 4-6) (Song et al., 2002, 2004, 2006, Berger, 2006).

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