



A study of eight newly reported species of Chlorophyte and Eustigmatophyte, Korea

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Abstract

In this study, aquatic and aerial algae were collected in various environments in Hongcheon-river of Gangwan-do between December 2011 and June 2012, with the aim of adding newly described genera and species to the Korean flora. As a result, five genera and eight species were recorded for the first time in Korea. These newly recorded genera and species were *Cylindrocapsa geminella*, *Leptosira mediciana*, *Pseudendozonium basiliense* var. *brandii*, *Stichococcus minor*, *S. deasonii*, *Eustigmatos polyphem*, *Nephrodiella lunaris*, and *Xanthonema exile*. The eight taxa identified in this study mostly corresponded to their reported morphological characteristics. However, some differences from previous published descriptions were found; *N. lunaris*, reported to be an aquatic species in a previous study, was found to be an aerial algae inhabiting on rocks and mosses. *Cylindrocapsa geminella* was found to transform into attached or planktonic algae depending on the environmental condition, and the cell wall was found to be changed. Likewise, *E. polyphem* was seen to change cell-shape or chloroplast color according to the environment.

Key words: aerial algae, freshwater, Hongcheon-river, newly recorded species

INTRODUCTION

Today, the ecosystem of the earth is facing the problem of reducing species diversity due to various factors (Jaenike 2007, Butchart et al. 2010). Advanced countries have been conducting continual research on about species diversity in order collect useful genes and construct gene banks for various organisms (Na et al. 2012). The South Korean government, for example, has been studying unexplored species and attempting to secure living resources and native Korean specimens since 2006. Microalgae are a useful species due to their physiological characteristics, which make them a widely used tool for genomics and proteomics (Shay 1993, Minowa et al. 1995, Lee et al. 2010). However, studies on the environments that algae

inhabit in Korea are less advanced than those in other countries. Furthermore, Korean native species have not been precisely collected, as investigations have lacked the professional manpower to perform sufficiently thorough work (Na et al. 2012). Also, few studies on microalgae have been conducted and are largely limited to species of industrial importance (Um and Kim 2009, Yoo et al. 2010, Lee et al. 2010). Studies on algae in Korea have focused especially on aquatic algae, while those on aerial algae in Korea are less advanced than in other countries (Škaloud 2009, Khaybullina et al. 2010). In Hongcheon-river, specifically, research on algae diversity has only looked at the unrecorded order Chlorococcales (Shin et al. 2013). Thus,

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Fig. 1. The map showing the sampling sites of Hongcheon-river in Gangwon-do from December 2011 to June 2012. Site numbers (H1 to H5) correspond to Table 1.

in this study, aquatic and aerial algae were collected in various environments within Hongcheon-river, with the aim of adding newly described genera and species to the known Korean flora.

MATERIALS AND METHODS

Freshwater and aerial algae were sampled in Hongcheon-river at Gangwon-do, Korea, from December 2011 to June 2012 (Fig. 1 and Table 1). Freshwater algae samples were collected from planktonic, periphytic, or subaerial habitats by using a phytoplankton net and a soft brush. Aerial algae samples were collected from stones, soil, and mosses by using a soft brush and a spatula. Each sample was sealed and refrigerated in a light-tight container with sterilized distilled water and transferred to the laboratory (Crispim et al. 2004). Some of the samples were fixed and stored in 1% formalin. Enriched cultures of aerial algae were made in Bold’s basal media (Stein 1973) and maintained in the algal culture collection of Kyonggi University (ACKU). The taxonomic classification system used was based on John et al. (2002, 2011) and Algaebase (Guiry and Guiry 2014), and taxa were identified based on the work of

Prescott et al. (1972, 1977, 1981, 1982), Prescott (1973), Hirose et al. (1977), Komárek and Fott (1983), Chung (1993), and Wehr and Sheath (2003). The samples were examined at $\times 400$ – $1,000$ magnification under a light microscope (BX41; Olympus, Tokyo, Japan) equipped with Nomarski differential interference optics. Species were illustrated by using a drawing attachment together with light microscope photographs.

RESULTS AND DISCUSSION

In this study, five genera were first recorded in Korea. In addition, eight species belonging to these five genera were added to the known Korean flora. The newly recorded genera for Korea are *Leptosira*, *Pseudendoclonium*, *Eustigmatos*, *Nephrodiella*, and *Xanthonema*. Three of these species are aquatic algae and five species are aerial algae. Information on the habitats of these species is shown in Table 2. The morphological and ecological characteristics of the eight, newly recorded, Korean species from Hongcheon-river from newly recorded genera listed above are as follows.

Table 1. Sampling sites in Hongcheon-river of Gangwon-do from December 2011 to June 2012

| Sampling sites | Local name | Latitude | Longitude |
|----------------|---|--------------|---------------|
| H1 | Jaeun-ri, Duchon-myeon, Hongcheon-gun | 37°52'24.16" | 128°01'27.36" |
| H2 | Yeongnae-ri, Duchon-myeon, Hongcheon-gun | 37°50'16.98" | 128°01'03.67" |
| H3 | Gurun-ri, Hwachon-myeon, Hongcheon-gun | 37°44'19.01" | 127°56'30.22" |
| H4 | Hahwagye-ri, Bukbang-myeon, Hongcheon-gun | 37°40'46.51" | 127°51'30.82" |
| H5 | Gwancheon-ri, Nam-myeon, Chuncheon-si | 37°44'25.17" | 127°30'42.93" |

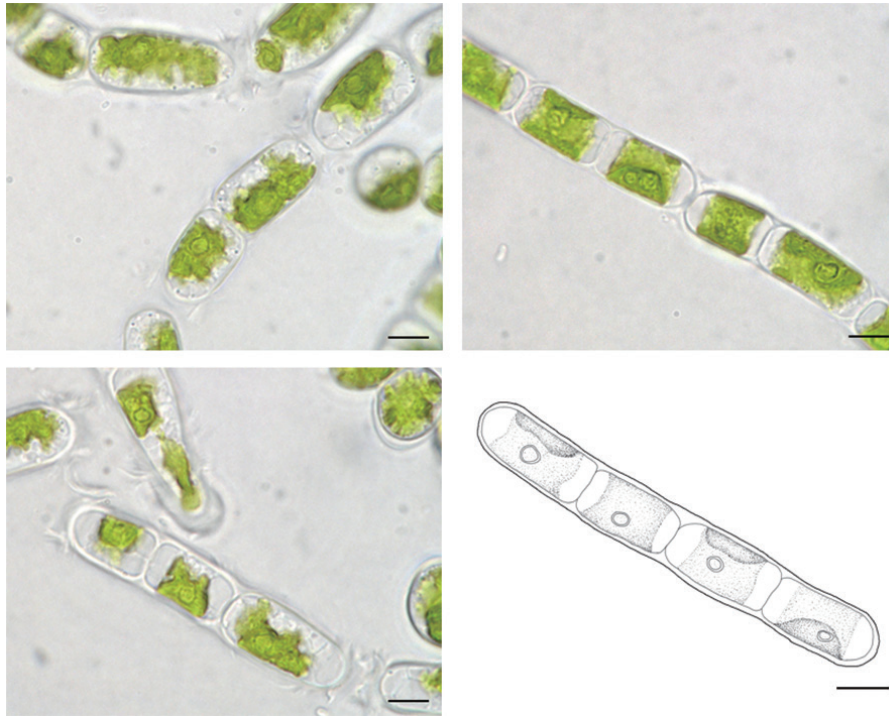


Fig. 2. Microscopic photographs and illustration of *Cylandrocapsa geminella* Wolle found in Hongcheon-river of Gangwon-do from December 2011 to June 2012. Scale bars, 10 µm.

Freshwater algae

Order Chlamydomonadales
Family Cylandrocapsaceae
Genus Cylandrocapsa

***Cylandrocapsa geminella* Wolle 1887 (Fig. 2)**

Cells 14-20 µm wide, usually 2-3 times as long as the width. Cells filamentous, cylindrical, with both sides rounded, one end slightly narrowed. Cell wall is composed of several layers, with a parietal star-shaped chloroplast. Cell walls are generally thin, but old cell walls are sometimes thick. This species is abundantly distributed

in freshwater (John et al. 2011). In this study, we expected this species to inhabit places with attached stone samples on the waterside and planktonic samples (Table 2). John et al. (2011) reported that *Cylandrocapsa* filaments grow by attaching and then subsequently detach and float. Thus, we hypothesized that the *Cylandrocapsa* in this study would be detached from the substrate.

Ecology and distribution: in Europe, Britain (John et al. 2002) and Romania (Carauş 2002); in North America, Arkansas (Smith 2010); in South-west Asia, Pakistan (Mehwish and Aliya 2005); in Australia and New Zealand, New Zealand (Broady et al. 2012), Queensland (Day et al. 1995), and South Australia (Day et al. 1995).

Table 2. Habitats of eight newly reported species in Hongcheon-river of Gangwon-do from December 2011 to June 2012

| Date | Sites | Species | Habitats |
|------------|--------|---|--|
| 2012-03-28 | H4, H5 | <i>Cylandrocapsa geminella</i> | Freshwater: attached stones, waterside, planktonic |
| 2012-03-28 | H3 | * <i>Leptosira mediciana</i> | Freshwater: planktonic |
| 2012-03-28 | H3 | * <i>Pseudendozonium basiliense</i> var. <i>brandii</i> | Freshwater: attached stones, waterside, inflow water |
| 2011-12-22 | H1 | <i>Stichococcus minor</i> | Aerial: attached stones, mosses |
| 2011-12-22 | H1 | <i>Stichococcus deasonii</i> | Aerial: attached stones, mosses |
| 2012-06-20 | H2 | * <i>Eustigmatos polyphem</i> | Aerial: moist soil |
| 2011-12-22 | H1 | * <i>Nephrوديella lunaris</i> | Aerial: attached stones, mosses |
| 2012-06-20 | H2 | * <i>Xanthonema exile</i> | Aerial: moist soil |

*newly reported genera; site numbers correspond to Table 1.

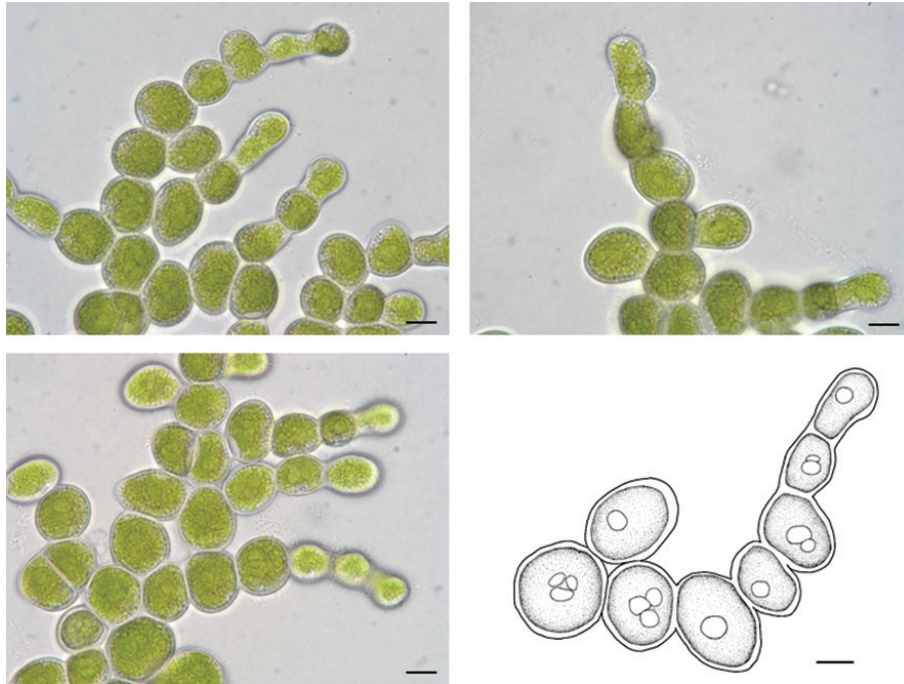


Fig. 3. Microscopic photographs and illustration of *Leptosira mediciana* Borzi found in Hongcheon-river of Gangwon-do from December 2011 to June 2012. Scale bars, 10 μ m.

Sites of collection: H4, H5; hereafter see Table 1 for site information.

Specimen locality: NIBRCL0000104602, ACKU HC2-146, 147, 155, 225.

Order Chaetophytales
Family Chaetophoraceae
Genus *Leptosira* Borzi 1883

This genus was first named by Borzi (1883) and is newly recorded in Korea by this study. Genus *Leptosira* has a solitary cell or irregularly branched cell, and it has no pyrenoid. Genus *Leptosira* is similar to genus *Pleurastrum*, but the latter is classified according to the presence of a pyrenoid (John et al. 2011, Guiry and Guiry 2014).

***Leptosira mediciana* Borzi 1883 (Fig. 3)**

Cells irregular, spherical, cylindrical, 11-25 μ m in diameter with irregularly branched cell. Last cell fragment is the smallest. Chloroplast parietal, with starch in the center of cells. Cell wall usually thick, lacking pyrenoid. In a study by John et al. (2011), this species was found in aquatic environments such as bogs and pools, as well as in aerial environment, including mosses and plants. In this study, we found this species in freshwater (Table 2).

Ecology and distribution : in Europe, Britain (John et

al. 2002, 2011).

Sites of collection: H3.

Specimen locality: ACKU HC2-74, 77.

Order Ulvales
Family Kornmanniaceae
Genus *Pseudendozonium* Wille 1901

This genus was named by Wille (1901) and is newly recorded in Korea by this study. *Pseudendozonium* contains bundle of filamentous cells.

***Pseudendozonium basilense* var. *brandii* Vischer 1933 (Fig. 4)**

Cells irregular, spherical, cylindrical, 5-13 μ m in diameter, with irregular branching. Last fragment cell is the smallest. Chloroplast parietal, with pyrenoid in the center of the cell. This species has been found on both artificial and natural matrices in water (John et al. 2011). In this study, we found this species on stones lying on the shore (Table 2).

Ecology and distribution: in Europe, Britain (John et al. 2002).

Sites of collection: H3.

Specimen locality: NIBRCL0000107670, ACKU HC2-120, 105.

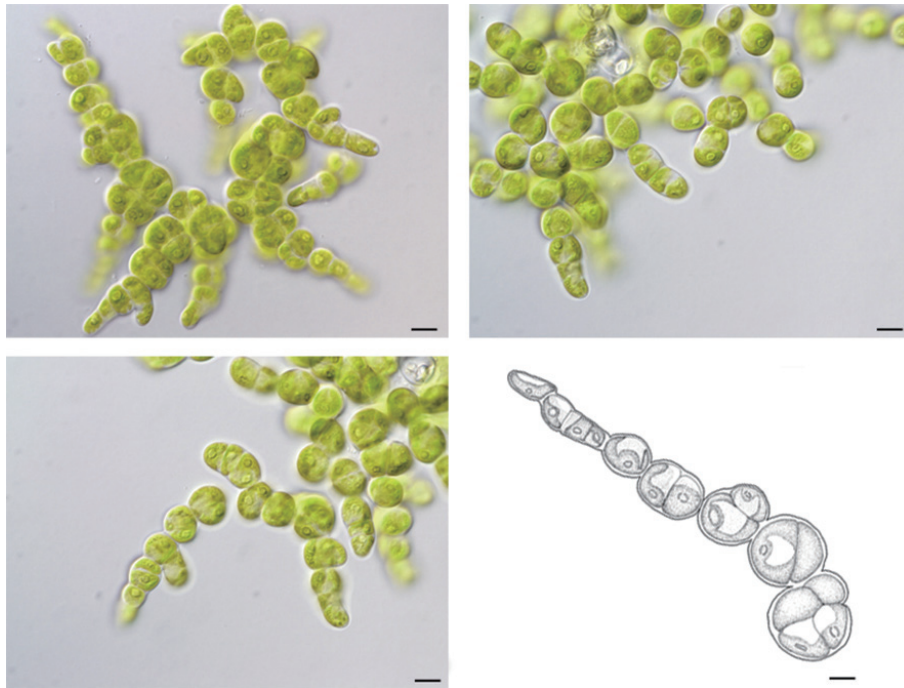


Fig. 4. Microscopic photographs and illustration of *Pseudoclonium basiliense* var. *brandii* Vischer found in Hongcheon-river of Gangwon-do from December 2011 to June 2012. Scale bars, 10 μ m.

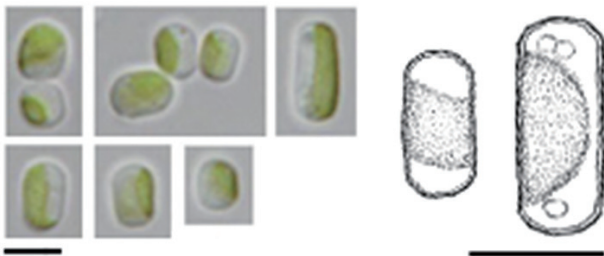


Fig. 5. Microscopic photographs and illustrations of *Stichococcus minor* Nägeli found in Hongcheon-river of Gangwon-do from December 2011 to June 2012. Scale bars, 5 μ m.

Aerial algae

Order Prasiolales

Family Prasiolaceae

Genus *Stichococcus* Nägeli 1849

Stichococcus minor Nägeli 1849 (Fig. 5)

Cells cylindrical, ellipsoidal or spherical, 3-9 μ m long, 2-3 μ m wide. Smaller than *Stichococcus bacillaris*. Chloroplast parietal, with starch, lacking pyrenoid. Globally,

this species is known to be an aerial alga (Guiry and Guiry 2014). In this study, this species was found in aerial environments, such as stones and mosses.

Ecology and distribution: in Europe, Baltic Sea (Hällfors 2004), Romania (Carauș 2002), Spain (Alvarez-Cobelas and Gallardo-Garca 1986).

Sites of collection: H1.

Specimen locality: NIBRCL0000104611, ACKU HC1-83.

Stichococcus deasonii Neustupa, Elias & Sejnova 2007 (Fig. 6)

Cells cylindrical with both sides rounded, 8-17 μ m long, 3-5 μ m wide. Cells solitary, with thin cell wall. Chloroplast parietal. The morphological characteristic of this species is that it has a starch envelope pyrenoid while other *Stichococcus* species do not. In a study by Neustupa et al. (2007), two species of genus *Stichococcus*, having the pyrenoid, were first reported. The study found the species in aerial environments such soil and tree bark. In the current study, the species was isolated from the aerial environment, including rocks and mosses.

Ecology and distribution: in Caribbean Islands, Alabama (Ettl and Gartner 1995).

Sites of collection: H1.

Specimen locality: NIBRCL0000107671, ACKU HC1-82.

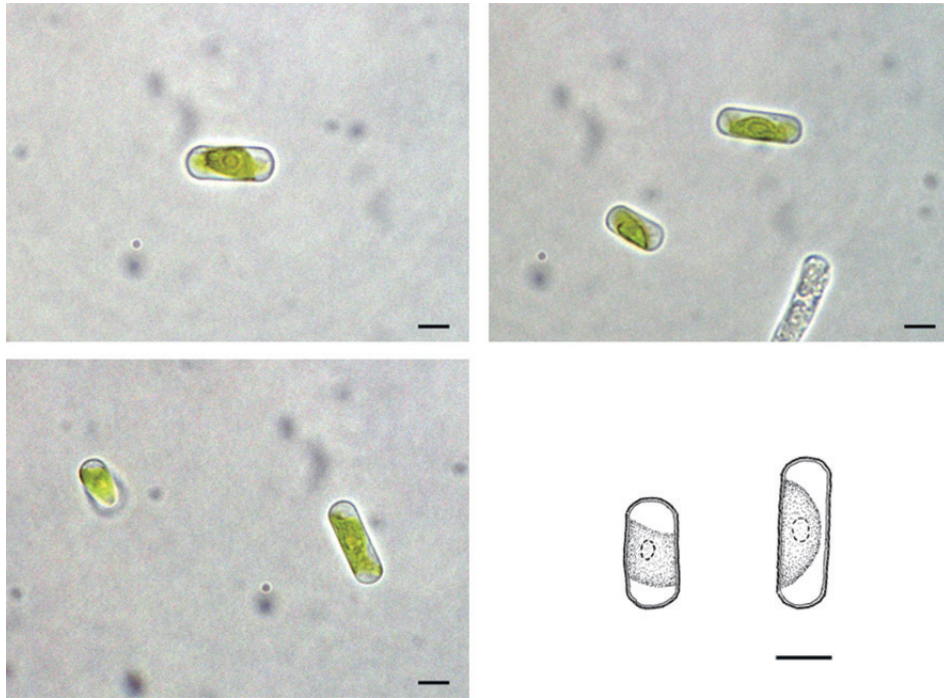


Fig. 6. Microscopic photographs and illustrations of *Stichococcus deasonii* Neustupa, Eliás & Sejnohová found in Hongcheon-river of Gangwon-do from December 2011 to June 2012. Scale bars, 5 μ m.

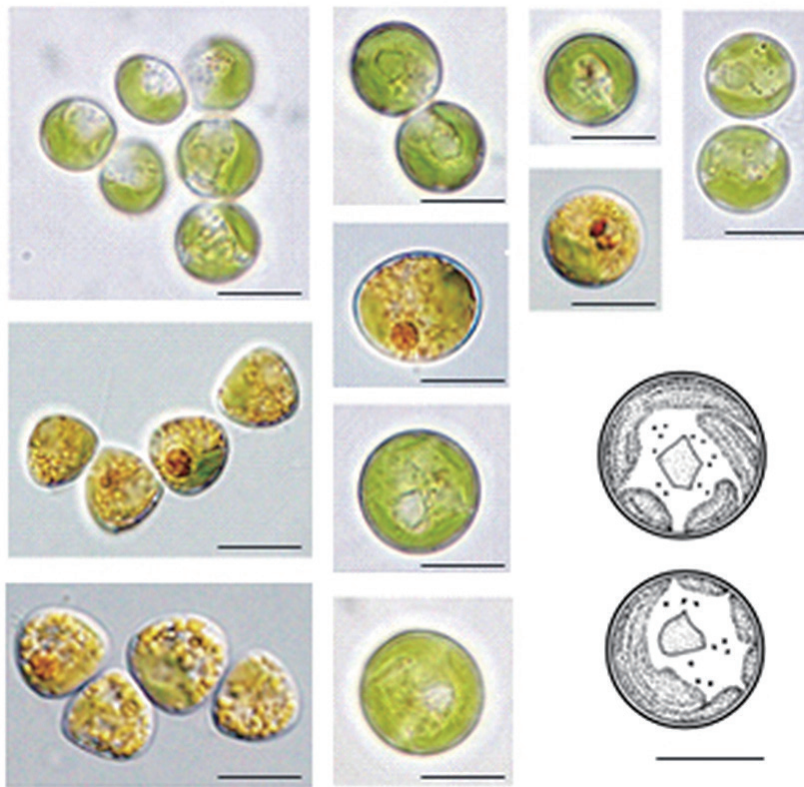


Fig. 7. Microscopic photographs and illustrations of *Eustigmator polyphem* (Pitschmann) Hibberd found in Hongcheon-river of Gangwon-do from December 2011 to June 2012. Scale bars, 10 μ m.

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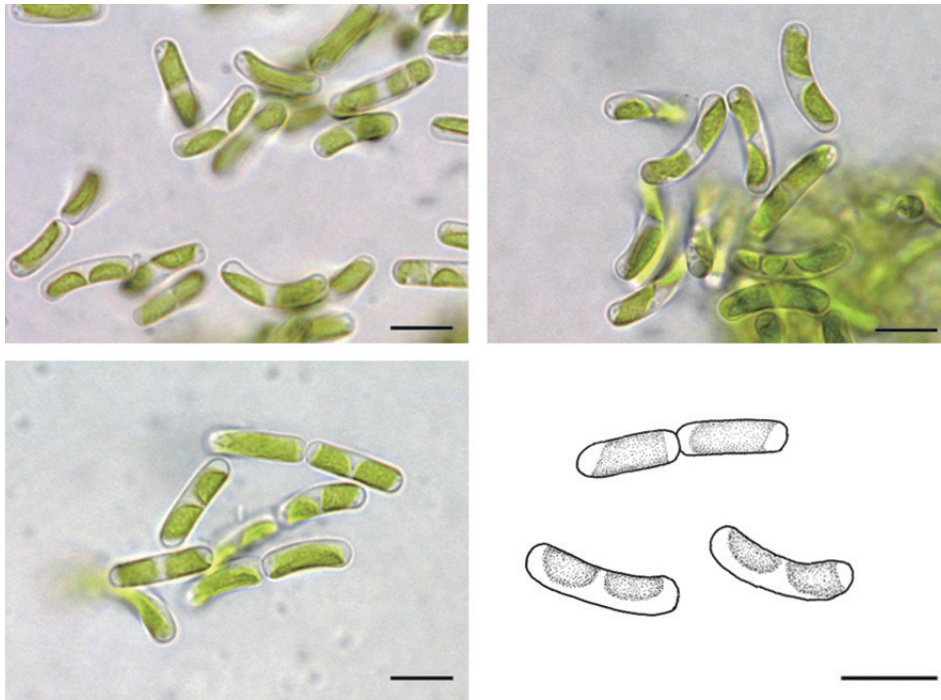


Fig. 8. Microscopic photographs and illustrations of *Nephrodiella lunaria* Pascher found in Hongcheon-river of Gangwon-do from December 2011 to June 2012. Scale bars, 10 μm .

Order Eustigmatales
Family Eustigmataceae
Genus *Eustigmatos* Hibberd 1981

This genus was named by Hibberd (1981) and is newly recorded in Korea by this study. This genus has morphological characteristics of a yellow-green chloroplast, a polyhedral pyrenoid and a red eyespot.

***Eustigmatos polyphem* (Pitschmann) Hibberd 1981 (Fig. 7)**

Basionym: *Pleurochloris polyphem* Pitschmann 1969.

Cells usually solitary, spherical or ellipsoidal, 5-22 μm in diameter. Shape of cell changes from spherical to pyramidal over cell life cycle. Cell wall is thin. Chloroplast is parietal with green color, but old cells change from yellow-green to yellow color. The cells have a polyhedral pyrenoid and one red eyespot. Škaloud (2009) and Ettl and Gärtner (1995) reported this species to be aerial algae. In the current study, this species was sampled and isolated from humid substrate as moist soils.

Ecology and distribution: in Africa, Kenya (Ettl and Gärtner 1995)

Sites of collection: H2.

Specimen locality: NIBRCL0000104613, ACKU HC3-21.

Order Mischococcales
Family Pleurochloridaceae
Genus *Nephrodiella* Pascher 1939

This genus was named by Pascher (1939) and is newly recorded in Korea by this study. Genus *Nephrodiella* has a kidney-shaped, lunate or bended cylindrical form. Chloroplast is parietal with a thin cell wall.

***Nephrodiella lunaria* Pascher 1939 (Fig. 8)**

The cell is 7-22 μm long, 3-4 μm wide and usually exists in solitary; but occasionally, two cells are attached together. The cells are kidney-shaped, lunate, or cylindrical. The chloroplast is parietal, with 1-3 fragments, lacking a pyrenoid. John et al. (2002) reported that it is commonly found in acidic to weakly alkaline water, moorlands and bogs. However, we found it in the aerial environment, on mosses and rocks.

Ecology and distribution: in Europe, Britain (John et al. 2002, 2011), Poland (Ettl and Gärtner 1995), Romania (Ettl and Gärtner 1995, Caraus 2002), Russia (Black Sea) (Ettl and Gärtner 1995).

Sites of collection: H1.

Specimen locality : NIBRFL0000129459, ACKU HC1-57.

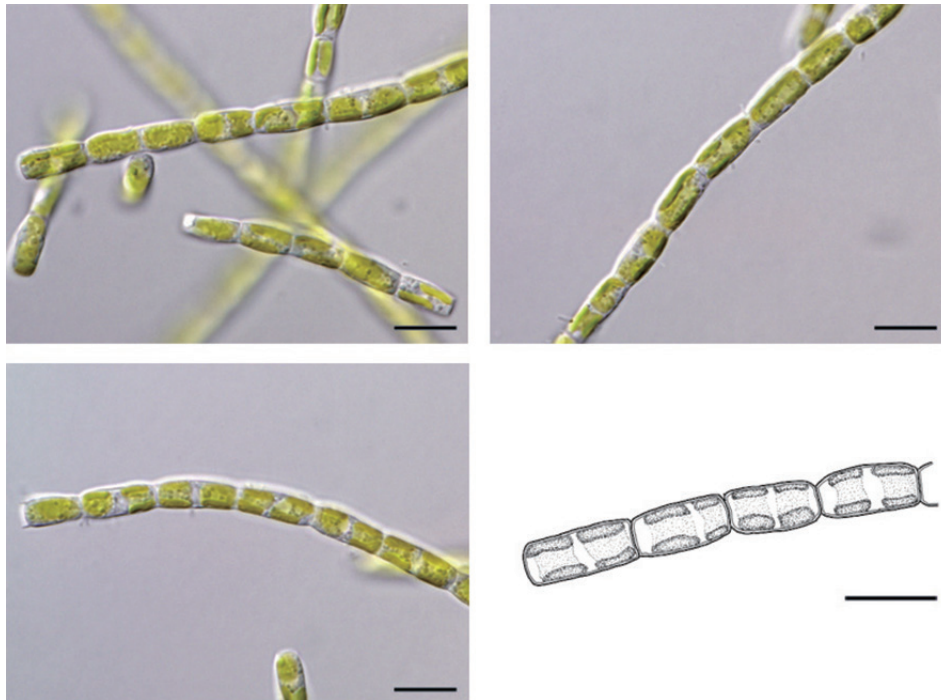


Fig. 9. Microscopic photographs and illustration of *Xanthonema exile* (Klebs) Silva found in Hongcheon-river of Gangwon-do from December 2011 to June 2012. Scale bars, 10 μm .

Order Tribonematales
Family Xanthonemataceae
Genus *Xanthonema* Silva 1979

This genus was named by Silva (1979) and is newly recorded in Korea by this study. Genus *Xanthonema*, with irregular branching, is known to be an aerial alga present in humid soils and weakly acidic water (Guiry and Guiry 2014). Species of genus *Xanthonema* are distinguished by a number of chloroplasts and shapes, cell size and shape of cell.

***Xanthonema exile* (Klebs) Silva 1979 (Fig. 9)**

Synonyms: *Bumilleria exilis* Klebs 1986; *Heterothrix exilis* (Klebs) Pascher 1932.

Cells are unbranched and filamentous, 6-15 μm long, 3-5 μm wide. Cells are cylindrical, and the cell-linked parts become narrow. Each cell has two chloroplasts, and those are parietal. This species is reported to be aerial algae (Guiry and Guiry 2014), and we also isolated this species in moist soil samples.

Ecology and distribution: in Europe, Austria (Ettl and Gärtner 1995), Britain (Ettl and Gärtner 1995, John et al. 2002, 2011), Russia (Black Sea) (Ettl and Gärtner 1995),

Switzerland (Ettl and Gärtner 1995); in Atlantic Islands, Iceland (Ettl and Gärtner 1995); in Asia, Japan (Ettl and Gärtner 1995); in New Zealand (Harper et al. 2012).

Sites of collection: H2.

Specimen locality: NIBRFL0000129460, ACKU HC3-32.

The morphological characteristics of the eight taxa identified in this study largely correspond to their reported characteristics. However, some differences from previous descriptions were found. *Nephrodiella lunaria* has been reported to be an aquatic species, but we found it living on rocks and mosses. *Cylindrocapsa geminella* takes the form of either attached or planktonic algae depending on the environmental condition, and the cell wall was changed. Likewise, *E. polyphem* was found to change cell-shape or chloroplast color according to its environment.

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