

# New Records and an Annotated Key for the Identification of *Graphis* Adans. in South Korea

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**Abstract** The following new species for the lichen genus *Graphis* in Korea are reported: *G. chlorotica*, *G. nanodes* and *G. tenuirima*. A brief description of these species, together with their distribution, ecology, and illustrations are provided. A key to all known species of this genus from Korea is also presented.

**Keywords** Graphidaceae, Lichen, Lirellate, Taxonomy

*Graphis*, the largest genus of the lichen family Graphidaceae (Ascomycota: *Ostropales*) was recently revised and redefined [1, 2]. The worldwide status of the genus has been exposed by modern taxonomic and revisionary studies, with about 40% of the species extensively belonging to the eastern Palaetropical region of the world [1-3]. As it is mainly found in the tropics, the diversity of *Graphis* in Korea was scarcely noticed and studied. In Korea, the genus was represented by thirteen species: *Graphis anfractuosa* (Eschw.) Eschw., *G. cervina* Müll. Arg., *G. cognata* Müll. Arg., *G. dupaxana* Vain., *G. handelii* Zahlbr., *G. intricata* Eschw., *G. proserpens* Vain., *G. intermediella* Stirt. [= *G. rikuzensis* (Vain.) M. Nakan.], *G. scripta* (L.) Ach., *G. aperiens* Müll. Arg., *G. flavopalmiticola* Y. Joshi, Lücking & Hur, *G. jejuensis* K. H. Moon, and *G. tsunodae* Zahlbr. [4-6]. This is undoubtedly a rather low estimate for the number of potential species compared to geographically similar countries such as Japan and Taiwan [7, 8]. Nevertheless, the scope of the genus in Korea is reflected by the amount of new reports and species described in the country [5, 6].

The present study broadens our knowledge of the lirellate

species of Graphidaceae in South Korea. This inventory reveals three new members of *Graphis* (*G. chlorotica*, *G. nanodes*, and *G. tenuirima*) and facilitates the identification of all known *Graphis* species of the country through an identification key.

## MATERIALS AND METHODS

Under the Korean National Research Resource Center Program and the Korean Forest Service Program, students of lichen taxonomy visited several places in the country (including islands) and collected several micro and macrolichen mycota. The members of Graphidaceae recently collected and preserved in the herbarium of Korean Lichen Research Center (KoLRI) from 2009 to 2012 were sorted and identified using standard microscopic techniques. The morphological and anatomical characteristics of the specimens were examined under a SMZ-168 dissecting microscope (Motic, Hong Kong, China) and an OLYMPUS BX 50 compound microscope (Olympus, Tokyo, Japan), respectively. The spot color reaction tests and thin layer chromatography (TLC) were performed using the methods described by Orange *et al.* [9]. For TLC, solvent system C (toluene:acetic acid = 85:15) was used. The range of measurements for anatomical characteristics of the species are the minimum and maximum values recorded on thin, hand-cut thallus and ascomatal sections mounted on water. The photographs were taken using an AxioCamERc 5c (Carl Zeiss, Jena, Germany) and plates were prepared in Coral Draw (version 12).

## RESULTS AND DISCUSSION

### Artificial key to *Graphis* species from South Korea

1. Labia striate, no chemical substances present ..... 2
- 1a. Labia entire, chemical substances present or absent ... 5

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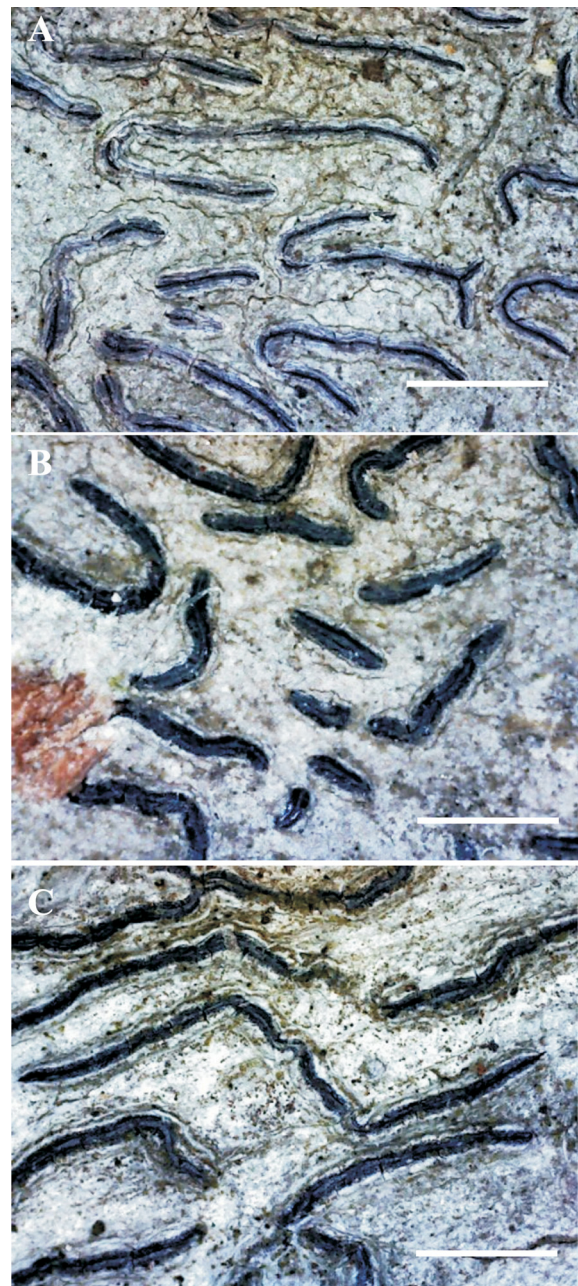
2. Proper exciple completely carbonized, lirellae erumpent to prominent, with basal or lacking thalline margin, ascospores 20~40  $\mu\text{m}$  long ..... *G. dupaxana*  
 2a. Proper exciple apically carbonized, ascospores 20~50  $\mu\text{m}$  long ..... 3  
 3. Lirellae prominent,  $\pm$  lacking thalline margin ..... *G. tsunodae*  
 3a. Lirellae erumpent ..... 4  
 4. Lirellae with lateral thalline margin, short to elongate, and sparsely to irregularly branched ..... *G. chlorotica*  
 4a. Lirellae with basal or lacking thalline margin, elongate and irregularly branched ..... *G. proserpens*  
 5. Proper exciple completely carbonized ..... 6  
 5a. Proper exciple apically or laterally carbonized ..... 11  
 6. Hymenium inspersed, ascospores 20~45  $\mu\text{m}$  long ..... 7  
 6a. Hymenium clear ..... 8  
 7. Norstictic acid present, lirellae erumpent with lateral thalline margin ..... *G. aperiens*  
 7a. Chemical substances absent, lirellae prominent, lacking thalline margin ..... *G. anfractuosa*  
 8. Chemical substances absent, lirellae prominent to sessile, with basal to lacking thalline margin, ascospores 40~70  $\mu\text{m}$  long ..... *G. cognata*  
 8a. Chemical substances present, lirellae immersed to erumpent with lateral thalline margin ..... 9  
 9. Lichexanthones present, thallus UV+ yellow, ascospores 19~27  $\mu\text{m}$  long ..... *G. flavopalmicola*  
 9a. Norstictic and stictic acids present, thallus UV- ..... 10  
 10. Norstictic and stictic acids present, lirellae short and sparsely branched, ascospores 15~25  $\mu\text{m}$  long, thallus often saxicolous ..... *G. cervina*  
 10a. Norstictic acid present, lirellae elongate to very long and irregularly to radiately branched, ascospores 15~30  $\mu\text{m}$  long, thallus corticolous ..... *G. intricata*  
 11. Hymenium inspersed, lirellae erumpent, apothecial disc exposed and epruinose ..... 12  
 11a. Hymenium clear ..... 13  
 12. Proper exciple  $\pm$  apically carbonized, no chemical substances present, ascospores 20~25  $\mu\text{m}$  long ..... *G. jejuensis*  
 12a. Proper exciple laterally carbonized, norstictic acid present, ascospores up to 45  $\mu\text{m}$  long ..... *G. handelii*  
 13. Proper exciple apically carbonized, lirellae erumpent, very long and radially branched, apothecial disc concealed, ascospores 20~45  $\mu\text{m}$  long ..... *G. intermediella*  
 13a. Proper exciple laterally carbonized, lirellae erumpent, apothecial disc exposed ..... 14  
 14. Ascospores transversely septate, usually more than 45  $\mu\text{m}$  long, apothecial disc distinctly white pruinose ..... *G. scripta*  
 14a. Ascospores muriform ..... 15  
 15. Ascospores 20~25  $\mu\text{m}$  long, apothecial disc thinly white pruinose ..... *G. tenuirima*  
 15a. Ascospores 25~30  $\mu\text{m}$  long, apothecial disc concealed ..... *G. nanodes*

### Taxonomic description of the species.

#### *Graphis chlorotica* A. Massal. (Fig. 1A)

Verh. Zool.-bot. Ges. Wien 21: 865 (1871).

**Description:** Thallus corticolous, epiperidermal, crustose, continuous, smooth, dull, whitish-grey to greenish-grey or dark grey, up to 100  $\mu\text{m}$  thick in cross section. Cortex layer indistinct to 20  $\mu\text{m}$ . Algal layer 60~70  $\mu\text{m}$ . Medulla indistinct endoperidermal. Prothallus whitish. Ascomata lirelliform, immersed to erumpent. Lirellae dispersed, 5~10 mm in length, black, covered by lateral thalline margin, short to elongate and irregularly branched (*tenella-*



**Fig. 1.** Habit of new records of lichens. A, *Graphis chlorotica* A. Massal.; B, *Graphis nanodes* Vain., C, *Graphis tenuirima* (Shirley) A. W. Archer (scale bars: A~C = 1 mm).

morph). Labia striate, epruinose. Disc closed, slit-like. Proper exciple apically carbonized, convergent, 70~160  $\mu\text{m}$  wide. Epihymenium 10~15  $\mu\text{m}$  high. Hymenium clear, up to 100  $\mu\text{m}$  high. Paraphyses 2~3  $\mu\text{m}$  thick. Hypothecium hyaline up to 25  $\mu\text{m}$  high. Ascus clavate 8-spored. Ascospores hyaline, fusiform, transversely septate, 9~13-loculate, 25~45  $\times$  7~10  $\mu\text{m}$ , halonate, halo 3~4  $\mu\text{m}$  thick.

**Chemistry:** Thallus K-, P-, C-, KC-; no lichen substances detected in TLC.

**Distribution and ecology:** Pantropical [2]. In South Korea, the species was collected in sub-temperate forests of Halla Mountain (at an altitude between 500~1,200 m) where it was growing among different graphidoid taxa in patches on the smooth trunk of trees.

**Remarks:** *Graphis chlorotica* closely resembles *G. tsumodae* and *G. proserpens* in most of its morpho-anatomical and chemical characteristics. Lücking *et al.* [2] treated all three species separately based on lirellae morphology, which in the latter two cases is  $\pm$  elongate and irregularly branched (*striatula*-morph), with basal to lacking thalline margin. The examined material clearly showed the immersed to erumpent lirellate apothecia surrounded by lateral thalline margin that marginally spread over striated black labia.

**Specimens examined:** Jeju-si, Jeju-do, Mt. Halla, Seongpanak Trail, alt. c. 833 m, on tree, 6 July 2012, Hur *et al.*, 121520 (KoLRI); alt. c. 1,164 m, on tree, 6 July 2012, Hur *et al.*, 121526 (KoLRI); Gwanumsa Trail, alt. c. 577 m, on tree, 1 June 2012, Hur *et al.*, 120919 (KoLRI).

#### ***Graphis nanodes* Vain. (Fig. 1B).**

Ann. Acad. Sci. Fenn. Sér. A 15: 209 (1921).

**Description:** Thallus corticolous, epi- to endo-peridermal, crustose, continuous, smooth, dull, whitish-grey to dark grey, 50~100  $\mu\text{m}$  thick in cross section. Cortex layer 20~25  $\mu\text{m}$ . Algal layer 40~70  $\mu\text{m}$ . Medulla indistinct, endoperidermal. Prothallus indistinct to whitish. Ascomata lirelliform, prominent. Lirellae scattered, 1~5 mm in length, black. Labia entire, epruinose. Disc closed, surrounded by lateral thalline margin (*lineola*-morph). Proper exciple laterally carbonized, convergent, 20~70  $\mu\text{m}$  wide. Epihymenium 15~20  $\mu\text{m}$  high. Hymenium clear, up to 100  $\mu\text{m}$  high. Paraphyses 1~2  $\mu\text{m}$  thick. Hypothecium hyaline, indistinct. Ascus clavate, 8-spored. Ascospores hyaline, ellipsoidal, muriform, 7~8  $\times$  1~4-loculate, 25~30  $\times$  8~10  $\mu\text{m}$ , halonate, halo 4~5  $\mu\text{m}$  thick.

**Chemistry:** Thallus K-, P-, C-, KC-; no lichen substances detected in TLC.

**Distribution and ecology:** Pantropical [2]. In South Korea, the species was collected at an altitude between 700~1,000 m from the northeastern and southern part of the country, where it was growing in irregular patches on the thick bark of trees.

**Remarks:** The specimens of *Graphis nanodes* examined have an ascospore size related and slightly smaller to *G. consimilis* Vain., but the latter species differs in the basal thalline margin (*lineola*-morph) of the lirellae. Another

species, *Graphis tenuirima*, with similar apothecial anatomical characteristics to that of *G. nanodes*, differs in having erumpent lirellae and an exposed, slightly white, pruinose apothecial disc.

**Specimens examined:** Gangwon-do, Yangyang-gun, Mt. Seorak, Heulringol valley, alt. c. 798 m, on bark, 25 May 2009, Joshi & Wang, 090906, 090911 (KoLRI); Jeju-do, Mt. Halla, Seongpanak Trail, alt. c. 812 m, on tree, 6 July 2012, Hur *et al.*, 121511 (KoLRI).

#### ***Graphis tenuirima* (Shirley) A. W. Archer, (Fig. 1C)**

Telopea 11: 74 (2005).

**Description:** Thallus corticolous, epidermal, crustose, continuous, smooth, dull, whitish-grey to grey, 100~120  $\mu\text{m}$  thick in cross section. Cortex layer indistinct to 15  $\mu\text{m}$ . Algal layer 50~70  $\mu\text{m}$ . Medulla indistinct, endoperidermal. Prothallus indistinct to whitish. Ascomata lirelliform, erumpent. Lirellae scattered, 5~10 mm in length, black; labia entire, over mature lirellae appears to be striated in the middle, epruinose. Disc open, thinly white pruinose (*scripta*-morph). Proper exciple laterally carbonized, convergent, 45~75  $\mu\text{m}$  wide. Epihymenium 10~15  $\mu\text{m}$  high. Hymenium clear, up to 100  $\mu\text{m}$  high. Paraphyses 1~2  $\mu\text{m}$  thick. Hypothecium hyaline, indistinct to 25  $\mu\text{m}$  high. Ascus clavate, 8-spored. Ascospores hyaline, ellipsoidal, muriform, 7~8  $\times$  1~2-loculate, 20~25  $\times$  10~13  $\mu\text{m}$ , initially halonate, halo 4~5  $\mu\text{m}$  thick.

**Chemistry:** Thallus K-, P-, C-, KC-; no lichen substances detected in TLC.

**Distribution and ecology:** Eastern Palaeotropics [2]. In South Korea the species was growing on thin and smooth tree bark in close association with other crustose, particularly those of the Lecanoroid group, and collected from the sub-temperate region of Jeju Island at an altitude above 1000 m.

**Remarks:** *Graphis sophisticascens* Nyl. resembles *G. tenuirima* in having erumpent lirellae, an exposed apothecial disc with white pruina, and a lack of secondary substances. In addition it has characteristics like an entire labia, laterally carbonized proper exciple, clear hymenium and muriform ascospores. However, *Graphis sophisticascens* Nyl. has comparatively smaller ascospores 15~20  $\mu\text{m}$  long, a thickly pruinose apothecial disc, and an entirely different geographical distribution in Eastern North America [2]. *Graphis consimilis* Vain. and *G. nanodes* also lack lichen compounds and have an ascomatal anatomy similar to *G. tenuirima*, but differ in having erumpent to prominent lirellae with a concealed disc, and slightly larger ascospores.

**Specimens examined:** Jeju-si, Jeju-do, Mt. Halla, Yeongsil Trail, alt. c. 1,184 m, on tree, 4 June 2012, Hur *et al.*, 121484 (KoLRI).

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