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# The lichen genus Fissurina (Graphidaceae) in Vietnam

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ABSTRACT — Nine species of *Fissurina* from Vietnam are briefly commented on. *Fissurina dumastii*, *F. dumastioides*, *F. instabilis*, *F. rubiginosa*, and *F. undulata* are recorded as new for the Vietnam lichen biota. Characteristic morpho-anatomical and chemical features are described and summarized in an artificial key to all known taxa of *Fissurina* from Vietnam.

KEY WORDS — Chu Yang Sin National Park, corticolous, graphidoid, taxonomy

#### Introduction

Fissurina Fée (Ascomycota: Ostropales) accommodates species with mostly slit-like lirellae (Staiger 2002; Archer 2009). The diagnostic characters of the genus includes a pale to yellow-brown to olive green (rarely whitish), mostly smooth and glossy thallus, fissurine, simple to branched, immersed to prominent lirellae, uncarbonized or rarely carbonized proper exciple, clear hyaline nonamyloid hymenium, 1–8-spored asci, and hyaline, oval or narrowly to broadly ellipsoid, trans-septate to muriform, amyloid to non-amyloid, thick-walled, mostly halonate ascospores (Archer 2009; Sharma et al. 2012). Fissurina occurs in the tropics and presently comprises more than 100 species worldwide. Many species have recently been added to the genus as a consequence of a molecular and phylogenetic revision of the *Graphidaceae* (Rivas Plata et al. 2012; Sharma et al. 2012).

Aptroot & Sparrius (2006) recorded four *Fissurina* species — *F. marginata* (= *F. elaiocarpa*), *F. egena*, *F. incrustans*, and *F. radiata* — from different Botanical Gardens and National Parks of Vietnam. During a recent field excursion to Chu Yang Sin National Park (Vietnam), several interesting graphidoid taxa were collected, including five new records of *Fissurina* for Vietnam: *F. dumastii*, *F. dumastioides*, *F. instabilis*, *F. rubiginosa*, and *F. undulata*. Among the *Fissurina* 

species in the country, *F. dumastioides* and *F. rubiginosa* produce stictic acid, whereas the others lack secondary metabolites.

This report is a preliminary attempt to assess the taxonomy of fissurine lichen species known so far from Vietnam. The synopsis clarifies the current status of the genus within the country and confirms the high diversity of graphidoid taxa in tropical rain forests. Diagnostic characters are studied from the available material. Unfortunately, many of the samples were either immature or collected in low frequency and many others that could increase the diversity considerably were sterile. In the following account nine reports of *Fissurina* are briefly described and summarized in an artificial key.

#### Materials & methods

Material deposited in the herbarium of the Korean Lichen Research Institute (KoLRI) was examined morphologically, anatomically, and chemically. The morphology of thallus and the ascomata were observed using a SMZ-168 dissecting microscope. Thin hand-cut sections mounted in tap water, in lactophenol cotton blue, in 5% KOH, or in Lugol's solution were observed under a Olympus BX50 compound microscope. Chemical spot tests and TLC (using solvent system C & A) were performed according to Orange et al. (2010).

### **Taxonomic descriptions**

*Fissurina dumastii* Fée, Essai Crypt. Écorc.: xc, 1825.

PL. 1

Thallus corticolous, epiperidermal, corticate, to ca. 250  $\mu m$  thick, glossy, smooth, continuous to uneven, greenish grey, pale green or olivaceous to dull green; cortex 15–30  $\mu m$  thick; algal layer well developed, continuous, 20–35  $\mu m$  thick; medulla indistinct, mostly endophloeodal, studded with crystals; calcium oxalate crystals small to moderately large, clustered or scattered; prothallus distinct, white. Vegetative propagules not seen.

Ascocarps numerous, lirelliform,  $\pm$  conspicuous, immersed to erumpent, indicated by a thin line between two lips, sometimes slightly open, slightly raised and paler than the thallus, straight, curved or sinuous, often branched,  $0.6-2 \times 0.2-0.25$  mm, *dumastii*-type (Staiger 2002); disc slit-like, narrow to sometimes open in mature apothecia, flesh-colored. Thalline margin concolorous with the thallus or slightly paler, thin to thick,  $\pm$  incurved or straight, 10-40 µm thick. Proper exciple brownish to dark brown, ca. 15-50 µm thick. Epihymenium hyaline, indistinct, with greyish or brownish granules,  $\le 10$  µm high. Hymenium hyaline, clear, to ca. 50-95 µm high, I–. Paraphyses straight to bent,  $\pm$  interwoven, unbranched, with slightly thickened tips, moderately to distinctly conglutinated, 1-2 µm thick. Subhymenium indistinct to hyaline, ca. 20 µm high. Asci clavate, 8-spored, ca. 65-80 µm, I–. Ascospores uniseriate, hyaline, becoming mottled or brown in late maturity, transversely

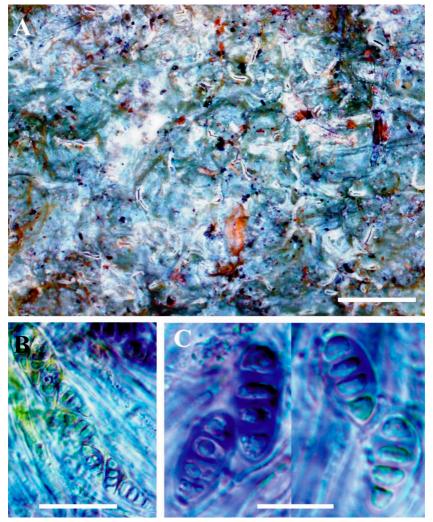


PLATE 1. Fissurina dumastii. A. Habit. B. Ascus with ascospores. C. Ascospores. Scale bars: A = 2 mm; B = 25  $\mu$ m; C = 10  $\mu$ m.

septate, oblong to ellipsoidal, with  $\pm$  rounded to narrowly rounded to subacute ends, thick walled, halonate, 15–18  $\times$  4–9  $\mu m$ , with 4 lenticular to  $\pm$  round locules, I+ blue violet.

CHEMISTRY: K-, PD-, C-; no lichen compound detected in TLC.

DISTRIBUTION & ECOLOGY: This species has a pantropical distribution and was previously reported from Queensland, New South Wales, Indonesia,

Solomon Islands, and Brazil (Staiger 2002; Archer 2006, 2009). In Vietnam it was collected from tree trunks of an evergreen forest in the Chu Yang Sin National Park at an altitude of ca. 600 m.

SPECIMEN EXAMINED: VIETNAM. DAK LAK PROVINCE: Chu Yang Sin National Park, 12°28′04.4″N 108°20′39.0″E, alt. ca. 622 m, on tree trunk, 20 April 2012, Hur, Oh & Nguyen 120282 (KoLRI).

REMARKS: Fissurina dumastii seems close to F. insidiosa C. Knight & Mitt. in having trans-septate ascospores and thallus lacking chemical substances, but differs in thallus and apothecial morphology. Fissurina insidiosa has distinct verrucose-bullate thallus and labiate lirellae. Moreover, the ascospores of F. dumastii show distinct amyloidity, absent in F. insidiosa. Another species belonging to the dumastii species complex is F. analphabetica Common & Lücking, but it has very small delicate lirellae (0.3–0.5 mm) and comparatively smaller ascospores of  $15 \times 7 \, \mu m$  (Lücking et al. 2011). Fissurina egena also has roof-like apothecial margins but differs in its muriform ascospores.

Fissurina dumastioides (Fink) Staiger, Biblioth. Lichenol. 85: 161, 2002. Pl. 2

Thallus corticolous, epiperidermal, corticate, to ca. 250  $\mu m$  thick, glossy, smooth, continuous to uneven, green to olive-green; cortex 15–25  $\mu m$  thick; algal layer well developed, continuous, 20–30  $\mu m$  thick; medulla indistinct, mostly endophloeodal, studded with crystals; calcium oxalate crystals small to moderately large, clustered or scattered; prothallus distinct, white. Vegetative propagules not seen.

Ascocarps numerous, lirelliform, ± conspicuous, immersed to erumpent, indicated by a thin line between two lips, sometimes broadly open, slightly raised and paler than the thallus, straight, curved or sinuous, often branched, ≤5 mm long and 0.2–0.3 mm wide with recurved margins, dumastii-type (Staiger 2002); disc narrow, open in mature apothecia, greyish to flesh-colored. Thalline margin concolorous with the thallus or slightly paler, thin to thick, ± incurved or straight, 10-40 µm thick. Proper exciple brownish to dark brown, ca. 15–50 µm thick. Epihymenium hyaline, indistinct, with greyish or brownish granules, ≤10 µm high. Hymenium hyaline, clear, to ca. 50–90 µm high, I–. Paraphyses straight to bent, ± interwoven, unbranched, with slightly thickened tips, moderately to distinctly conglutinated, 1-2 µm thick. Subhymenium indistinct to hyaline, ca. 20 μm high. Asci clavate, 8-spored, ca. 60-80 μm, I-. Ascospores uniseriate, hyaline, becoming mottled or brown in late maturity, transversely septate, oblong to ellipsoidal, with  $\pm$  rounded to narrowly rounded to subacute ends, thick walled, halonate,  $15-20 \times 4-9 \mu m$ , with 4 lenticular to ± round locules, I+ blue violet.

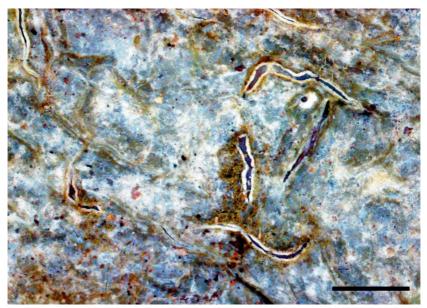


PLATE.2. Fissurina dumastioides. Habit. Scale bar: 2 mm

CHEMISTRY: K+ yellow, PD+ yellow-orange, C-; stictic acid detected in TLC.

DISTRIBUTION & ECOLOGY: The species is known to occur in Puerto Rico and India (Fink 1927, Sharma et al. 2012), and has now been identified from Vietnam. It was collected from trees in an evergreen tropical forest of the Chu Yang Sin National Park at an altitude between 700–800 m.

Specimen examined: VIETNAM. Dak Lak province: Chu Yang Sin National Park,  $12^{\circ}27'57.0''N$   $108^{\circ}20'34.9''E$ , alt. ca. 780 m, on tree trunk, 21 April 2012, Hur, Oh & Nguyen 120267-1 (KoLRI).

Remarks: *Fissurina dumastioides* resembles *F. dumastii* morphologically and anatomically, but differs in containing stictic acid as thallus compound.

## Fissurina egena (Nyl.) Nyl., Sert. Lich. Trop.: 13, 1891

This taxon is morphologically similar to *F. cingalina* (Nyl.) Staiger, with a ochre, yellow-grey, yellowish green, slightly rough or warty thallus (Staiger 2002). However, *F. egena* can be distinguished from *F. cingalina* in its immersed lirellae with roof-like margins and concealed to slightly exposed disc, its 8-spored asci, its muriform, amyloid,  $15-23\times7-10~\mu m$  ascospores and the lack of secondary metabolites.

It has been reported from Singapore, North and South America, and Vietnam (Staiger 2002; Aptroot & Sparrius 2006).

# Fissurina elaiocarpa (A.W. Archer) A.W. Archer, Telopea 11: 71, 2005.

= Fissurina marginata Staiger, Bibliotheca Lichenologica 85: 144, 2002.

This species is characterized by a olive green, slightly subtuberculate, glossy thallus, conspicuous, sessile, straight, curved or sinuous, unbranched, ca. 1–8  $\times$  4 mm ascomata, scarcely open lip, reddish brown proper exciple, 150–200  $\mu m$  high hymenium, 4–5  $\times$  3–4 locular, 21–28  $\times$  13–18  $\mu m$  large, amyloid ascospores and absence of secondary metabolites (Archer 2009).

Aptroot & Sparrius (2006) reported this species from Vietnam as *F. marginata* and collected it from a secondary mountain forest of Ba Vi National Park (Hanoi Province), at a 1200–1300 m altitude. The species has a worldwide distribution and is found in Africa, Australia, South and Western America, and Western Indian Ocean (Staiger 2002).

### Fissurina incrustans Fée, Essai Crypt. Écorc.: 60, 1825.

This species is characterized by a beige green, smooth to warty thallus, elongate to short ( $1-4 \times 0.3-0.4$  mm), sinuous, branched ascomata, erumpent labia, gaping disc, poorly developed proper exciple, usually intermingled with bark substratum, 8-spored asci, muriform, amyloid ascospores of  $15-28 \times 7-13$  µm and absence of secondary compounds. It closely resembles *F. egena* in having similar-sized muriform amyloid ascospores and a thallus lacking chemical compounds, but *F. incrustans* has erumpent lirellae more similar to the ones found in *Platythecium* than the ones found in *Fissurina* (Staiger 2002; Lücking et al. 2011).

The species has been reported in Vietnam from a secondary mountain forest in Ba Vi National Park by Aptroot & Sparrius (2006). It is also found in South America (Staiger 2002).

# Fissurina instabilis (Nyl.) Nyl., Sert. Lich. Trop.: 36, 1891. PL. 3

Thallus corticolous, epiphloeodal, corticate,  $\leq$ 300 µm thick, glossy, smooth, continuous to  $\pm$  verrucose (due to bark texture), non-rimose, green, olive green to pale green; cortex 35–45 µm thick; algal layer well developed, continuous, densely integrated with crystals,  $\leq$ 215 µm thick; medulla indistinct, mostly endophloeodal, integrated with crystals; calcium oxalate crystals small to moderately large, clustered or scattered; prothallus indistinctly black. Vegetative propagules not seen.

Ascocarps numerous, lirelliform, inconspicuous, immersed to  $\pm$  erumpent, indicated by a thin line between two lips (labia), sometimes slightly open, slightly raised, curved or sinuous, often branched,  $\leq 10 \times 0.6$  mm, *globulifica*-type (Staiger 2002); labia entire, paler than the thallus; disc slit-like, narrow, scarcely open in mature apothecia, flesh-colored. Thalline margin concolorous with the thallus, thick,  $\pm$ incurved or straight, 180-215 µm thick. Proper

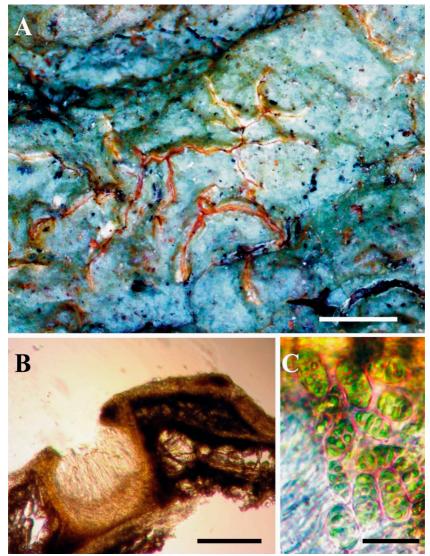


Plate 3. Fissurina instabilis A. Habit. B. Cross section of apothecia. C. Ascospores. Scale bars: A = 2 mm; B =  $100~\mu m$ ; C =  $20~\mu m$ .

exciple well developed, brownish to reddish brown, superficially exposed as a red-brown lining, ca.  $30–55~\mu m$  thick. Epihymenium hyaline, indistinct, with greyish or brownish granules,  $3–6~\mu m$  high. Hymenium hyaline, clear, to ca.  $75–100~\mu m$  high, I–. Paraphyses straight to bent,  $\pm interwoven$ , unbranched,

with slightly thickened tips, moderately to distinctly conglutinated, 1–2  $\mu m$  thick. Subhymenium distinct, hyaline, ca. 40–50  $\mu m$  high. Asci clavate, 8-spored, ca. 65–80  $\mu m$ , I–. Ascospores uni- or biseriate, hyaline, muriform, oblong to ellipsoidal, with  $\pm$  rounded ends, thick walled, 13–20  $\times$  7–11  $\mu m$ , with 3–4(–5)  $\times$  1–2  $\pm round$  locules, I+ blue violet.

CHEMISTRY: K-, PD-, C-; no lichen compound detected in TLC.

DISTRIBUTION & ECOLOGY: This species has a Pacific distribution (Staiger 2002). In Vietnam it was collected from tree trunks in an evergreen forest of the Chu Yang Sin National Park at an altitude between 700–800 m.

Specimens examined: VIETNAM. Dak Lak province: Chu Yang Sin National Park,  $12^\circ27'57.0''N$   $108^\circ20'34.9''E$ , alt. ca. 780 m, on tree trunk, 21 April 2012, Hur, Oh & Nguyen 120132 (KoLRI);  $12^\circ28'12.3''N$   $108^\circ20'59.9''E$ , alt. ca. 763 m, on tree trunk, 20 April 2012, Hur, Oh & Nguyen 120087 (KoLRI).

REMARKS: Fissurina instabilis resembles F. confusa Common & Lücking and F. globulifica (Nyl.) Staiger in having the same thallus morphology, globulificatype lirellae, and muriform amyloid ascospores but F. confusa and F. globulifica produce psoromic acid and F. globulifica has smaller globose ( $10 \times 10 \mu m$ ) ascospores (Lücking et al. 2011).

## Fissurina radiata Mont., Annls Sci. Nat., Bot., sér. 2, 18: 280, 1842.

This species differs from other *Fissurina* species in having aggregated, mostly radiate ascomata forming a pseudostroma, inconspicuous and gaping labia, amyloid, 4-locular ascospores of  $15 \times 7~\mu m$ , and a thallus lacking secondary metabolites (Lücking et al. 2011). It is comparable to material described from Florida as *Fissurina aggregatula* Common & Lücking (Lücking et al. 2011), which differs in having conspicuous labia and a concealed disc, sparsely branched aggregated lirellae, and non-amyloid ascospores.

*Fissurina radiata* has been reported from the Botanical Garden (Bach Thao Park) in Hanoi City in Vietnam at an altitude between 50–60 m (Aptroot & Sparrius 2006). It has a distribution in tropical regions of America (Lücking et al. 2011), Malaysia, Singapore, and Taiwan.

# Fissurina rubiginosa (Fée) Staiger, Biblioth. Lichenol. 85: 148, 2002. Pl. 4

Thallus corticolous, epiphloeodal, corticate,  $\leq\!350~\mu m$  thick, glossy, smooth, continuous, non-rimose, green, olive green to pale green; cortex 30–50  $\mu m$  thick; algal layer well developed, continuous, densely integrated with crystals,  $\leq\!200~\mu m$  thick; medulla indistinct, mostly endophloeodal, studded with crystals; calcium oxalate crystals small to moderately large, clustered or scattered; prothallus indistinctly black. Vegetative propagules not seen.

Ascocarps numerous, scattered, lirelliform, inconspicuous, immersed, indicated by a thin line, sometimes slightly open, slightly raised and paler than

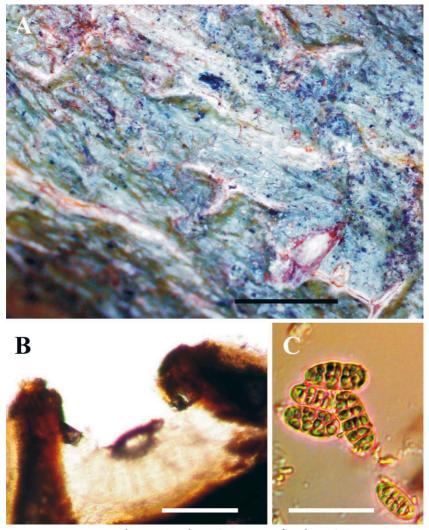


Plate 4. Fissurina rubiginosa. A. Habit. B. Cross section of apothecia. C. Ascospores. Scale bars: A = 2 mm; B =  $100~\mu m$ ; C =  $30~\mu m$ .

the thallus, straight, curved or sinuous, often branched,  $\leq 2 \times 0.25$  mm, *dumastii*-type; disc slit-like, narrow, scarcely open in mature apothecia, flesh-colored. Thalline margin concolorous with thallus, thick, ±incurved or straight, 75–150  $\mu$ m thick. Proper exciple well developed, pale to reddish brown, intermingled with bark cells,  $\leq 75~\mu$ m thick. Epihymenium hyaline, indistinct, with greyish

or brownish granules, 10–15 µm high. Hymenium hyaline, clear,  $\leq 120-150$  µm high, I–. Paraphyses straight to bent, ±interwoven, unbranched, with slightly thickened tips, moderately to distinctly conglutinated, 1–2 µm thick. Subhymenium indistinct, hyaline, ca. 25–30 µm high. Asci clavate, 8-spored, ca. 65–80 µm, I–. Ascospores uni- or biseriate, hyaline, muriform, oblong to ellipsoidal, with ±rounded ends, thick walled, 20–27  $\times$  10–12 µm, with 6–8  $\times$  2–3  $\pm$  round locules, I–.

CHEMISTRY: K-, PD-, C-; no lichen compound detected in TLC.

DISTRIBUTION & ECOLOGY: The species is found in French Guiana, USA and India (Sharma et al. 2012), but is now also reported from Vietnam where it was and collected from tree trunks in an evergreen forest of the Chu Yang Sin National Park at an altitude between 700–800 m.

SPECIMEN EXAMINED: VIETNAM. DAK LAK PROVINCE: Chu Yang Sin National Park, 12°28′12.3″N 108°20′59.9″E, alt. ca. 763 m, on tree trunk, 20 April 2012, Hur, Oh & Nguyen 120246 (KoLRI).

Remarks: This species is close to F. elaiocarpa. Their thallus morphology is similar and they both have muriform ascospores, although the spores are comparatively larger ( $18-32\times13-19~\mu m$ ) and I+ blue in F. elaiocarpa. Another closely related taxon, Fissurina aff. elaiocarpa, resembles F. rubiginosa in having muriform, I- ascospores of similar size ( $25\times15~\mu m$ ), but differs in labiate ascomata and verrucose thallus. The description above does not agree with that of F. rubiginosa provided by Sharma et al. (2012), which describes a brown, cracked and rough thallus and much smaller lirellae (0.4-0.6~m m). However, the investigated sample shares some main characteristics with F. rubiginosa, such as dumastii-type lirellae and non-amyloid ascospores. Because of insufficient material, we provisionally identify our specimen from Vietnam as F. rubiginosa.

Fissurina undulata (Müll. Arg.) M. Nakan. & Kashiw., Bull. natn. Sci. Mus., Tokyo, B 29: 87, 2003. Pl. 5

Thallus corticolous, epiphloeodal, corticate, to ca. 400  $\mu m$  thick, glossy, verrucose to smooth, continuous, non-rimose or slightly cracked due to bark texture, green, olive green to pale green or dull green; cortex 25–45  $\mu m$  thick; algal layer well developed, continuous, densely integrated with crystals,  $\leq \! 100$   $\mu m$  thick; calcium oxalate crystals small to moderately large, clustered or scattered; medulla indistinct, mostly endophloeodal, studded with crystals; prothallus not seen. Vegetative propagules not seen.

Ascocarps numerous, scattered, lirelliform, inconspicuous, immersed, indicated by a thin line, sometimes slightly open, slightly raised and paler than the thallus, straight, curved or sinuous, often branched,  $\leq 2 \times 0.3$  mm, dumastii-type; disc slit-like, narrow, scarcely open in mature apothecia, flesh-

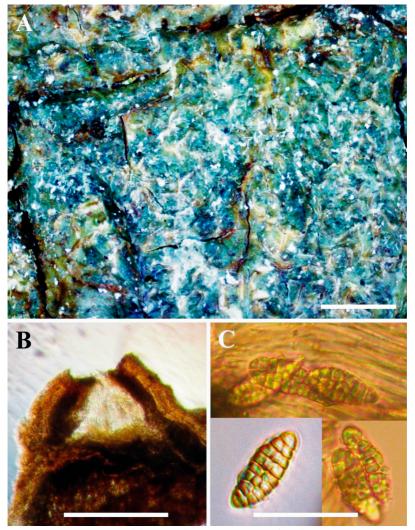


Plate 5. Fissurina undulata. A. Habit. B. Cross section of apothecia. C. Ascospores. Scale bars: A = 2 mm; B = 200  $\mu$ m; C = 30  $\mu$ m.

colored. Thalline margin concolorous with thallus, thick,  $\pm$ incurved or straight, 50–100  $\mu$ m thick. Proper exciple poorly to well developed in mature fruits, hyaline to pale, reddish brown when intermingled with bark cells,  $\leq$ 30  $\mu$ m thick. Epihymenium hyaline, indistinct, with greyish or brownish granules. Hymenium hyaline, clear, to ca. 60–100  $\mu$ m high, I–. Paraphyses straight to bent,  $\pm$ interwoven, unbranched, with slightly thickened tips, moderately to

distinctly conglutinated, 1–2 µm thick. Subhymenium indistinct, hyaline, ca. 20–30 µm high. Asci clavate, 8-spored, ca. 70–95  $\times$  12–15 µm, I–. Ascospores uni- or biseriate, hyaline, muriform, oblong to ellipsoidal, with ±rounded ends, thick walled, gelatinous sheath present, 22–32(–35)  $\times$  10–13 µm, with 6–12  $\times$  2–4 locules, I–; locules ±round to angular.

CHEMISTRY: K-, PD-, C-; no lichen compounds detected in TLC.

DISTRIBUTION & ECOLOGY: The species is found in Japan (Nakanishi 1966), but is now also reported from Vietnam where it was collected from tree trunks in an evergreen forest in Chu Yang Sin National Park at an altitude between 700–800 m.

SPECIMEN EXAMINED: VIETNAM. DAK LAK PROVINCE: Chu Yang Sin National Park, 12°28′12.3″N 108°20′59.9″E, alt. ca. 763 m, on tree trunk, 20 April 2012, Hur, Oh & Nguyen 120247 (KoLRI).

Remarks: The above description is based on a specimen with mostly immature fruits, which appear more similar to F. undulata than to F. inabensis (Vain.) M. Nakan. & Kashiw. (erroneously cited as F. fujisanensis (Kashiw. & M. Nakan.) M. Nakan. & Kashiw. in Nakanishi et al. 2003). Both F. undulata and F. inabensis produce stictic acid and have similar-sized ascospores (21–30  $\times$ 10-14 μm for F. inabensis). However, a thin thallus, a well developed proper exciple, crenate labia, a medulla with gonidia and 2-3-celled ascospores clearly separate F. inabensis from F. undulata (Nakanishi 1966). Although the descriptions of F. undulata by Nakanishi (1966), Nakanishi et al. (2003), and Sharma et al. (2012) differ in the number of ascospores per ascus (8-spored asci sensu Nakanishi vs. 1-spored asci sensu Sharma), our material agrees well with a species with 8-spored asci. Moreover, the ascospore size  $(30-38 \times 15-18 \mu m)$ of *F. undulata* is in conflict with the ascospore size of species with 1-spored asci. A further revision of F. undulata would be necessary, as it seems closely related to F. inabensis, particularly in immature stages (Nakanishi 1966). Pending some more collections, the Vietnamese material is placed in *F. undulata*.

### Key to Fissurina species recorded from Vietnam

1a. Thallus containing stictic acid	
1b. Thallus lacking secondary metabolites	
2a. Ascospores transversely septate         2b. Ascospores muriform	
3a. Ascospores transversely septate         3b. Ascospores muriform	
4a. Lirellae mostly aggregated, radiate, forming a pseudostroma 4b. Lirellae disperse, solitary, branched, non-radiate	
5a. Ascospores non-amyloid, 22–32(–35) × 10–13 μm	

6a. Lirellae immersed, fissurine, roof like margins,
as cospores $15-23 \times 7-10~\mu m$ F. egena
6b. Lirellae immersed to erumpent or prominent, labiate
7a. Lirellae immersed to $\pm$ erumpent, ascospores 13–20 $\times$ 7–11 $\mu m$
7b. Lirellae erumpent to prominent
8a. Thallus ± smooth, proper exciple poorly developed,
ascospores $15-28 \times 7-13 \ \mu m$ F. incrustans
8b. Thallus verrucose, proper exciple well developed,
ascospores $21-28 \times 13-18 \ \mu m$ F. elaiocarpa

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