

FULL PAPER

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Nervostroma*, gen. nov. in the Sclerotiniaceae, the teleomorph of *Cristulariella*, and *Hinomyces* anam. gen. nov. to accommodate the anamorph of *Grovesinia*: reassessment of the genus *Cristulariella

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Abstract Taxonomy of the genus *Cristulariella* is revised, retaining *Cristulariella* (*Crist.*) *depraedans* as the type. Two new species, *Crist. cercidiphylli* and *Crist. corni*, are additionally described under the genus. The new anamorphic genus *Hinomyces* is erected to accommodate *Botrytis* (*Cristulariella*) *moricola* and *Cristulariella pruni*. A new genus and species, *Nervostroma depraedans*, is erected in the Sclerotiniaceae to accommodate the teleomorph of *Crist. depraedans*, with an additional species, *Nervostroma cercidiphylli*.

Key words *Cristulariella* · *Hinomyces* · *Nervostroma* · Sclerotiniaceae · Taxonomy

Introduction

Species of the genus *Cristulariella* (*Crist.*) Höhn. emend. Redhead (Redhead 1975) have been known to mycologists and plant pathologists as zonate leaf spot fungi on herbaceous and woody plants. They are members of anamorphic fungi with hyaline many-celled conidia on the spots and then black sclerotia on diseased fallen leaves. The sclerotia may germinate to produce apothecia after they overwinter and when they encounter proper environmental conditions.

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The genus at present involves *Crist. depraedans* (Cooke) Höhn., the cause of *Acer* leaf spot disease (Fig. 1), *Crist. moricola* (I. Hino) Redhead, the cause of common zonate leaf spot disease, and *Crist. pruni* Y. Harada et Noro, the cause of *Prunus* zonate leaf spot disease (Höhnel 1916; Bowen 1930; Sawada 1933; Waterman et al. 1947; Redhead 1975, 1979; Harada and Noro 1988). Teleomorphs of *Crist. moricola* and *Crist. pruni* were found and named *Grovesinia pyramidalis* M.N. Cline, J.L. Crane, et S.D. Cline (see Fig. 28) and *G. pruni* Y. Harada et Noro, respectively, in the Sclerotiniaceae (Cline et al. 1983; Harada and Noro 1988). In this article, two new *Cristulariella* species, i.e., one on *Cercidiphyllum japonicum* Siebold et Zucc. (Fig. 2) and another on *Cornus controversa* Hemsl., are described (Fig. 3). Further, teleomorphs of two *Cristulariella* species, i.e., *Crist. depraedans* and *Cristulariella* on *C. japonicum*, were first found on overwintered, diseased leaves of their respective hosts. Consequently, we realized that the teleomorph of *Crist. depraedans*, the type species of the genus *Cristulariella*, cannot properly be accommodated in the genus *Grovesinia*, in view of morphological and cultural characteristics. These findings led us to a new taxonomic treatment of the fungus, as described next.

Material and methods

Fungus species studied

Cristulariella depraedans on *Acer japonicum* Thunb. and two newly found *Cristulariella* species, occurring on *Cercidiphyllum japonicum* and *Cornus controversa*, respectively, were studied for their pathogenicity, life cycle, and morphology. Cultural experiments were also made of these fungi as compared with *Crist. moricola* (teleomorph: *Grovesinia pyramidalis*) and *Crist. pruni* (teleomorph: *G. pruni*).

The fungus isolates used in cultural experiments are shown in Table 1. They were isolated from ascospores, apothecial tissues, or conidia, as follows.

Figs. 1–11. Disease symptoms, conidial morphology, and colony appearance of *Cristulariella* (*Crist.*) and *Hinomyces*. **1** *Acer japonicum* leaves with spots caused by *Crist. depraedans*. **2** *Cercidiphyllum japonicum* leaves with spots caused by *Crist. cercidiphylli*. **3** *Cornus controversa* leaves with spots caused by *Crist. corni*. **4–6** Photomicrographs (**a**) and line drawings (**b**) of conidia of *Crist. depraedans* (**4**), *Crist. cercidiphylli* (**5**), and *Crist. corni* (**6**). **7–11** Colonies of *Crist. depraedans* (**7**), *Crist. cercidiphylli* (**8**), *Crist. corni* (**9**), *Hinomyces moricola* (**10**), and *H. pruni* (**11**), grown for 45 days at 15°C under 12:12 L:D. Bars **4b**, **5b**, **6b** 50µm

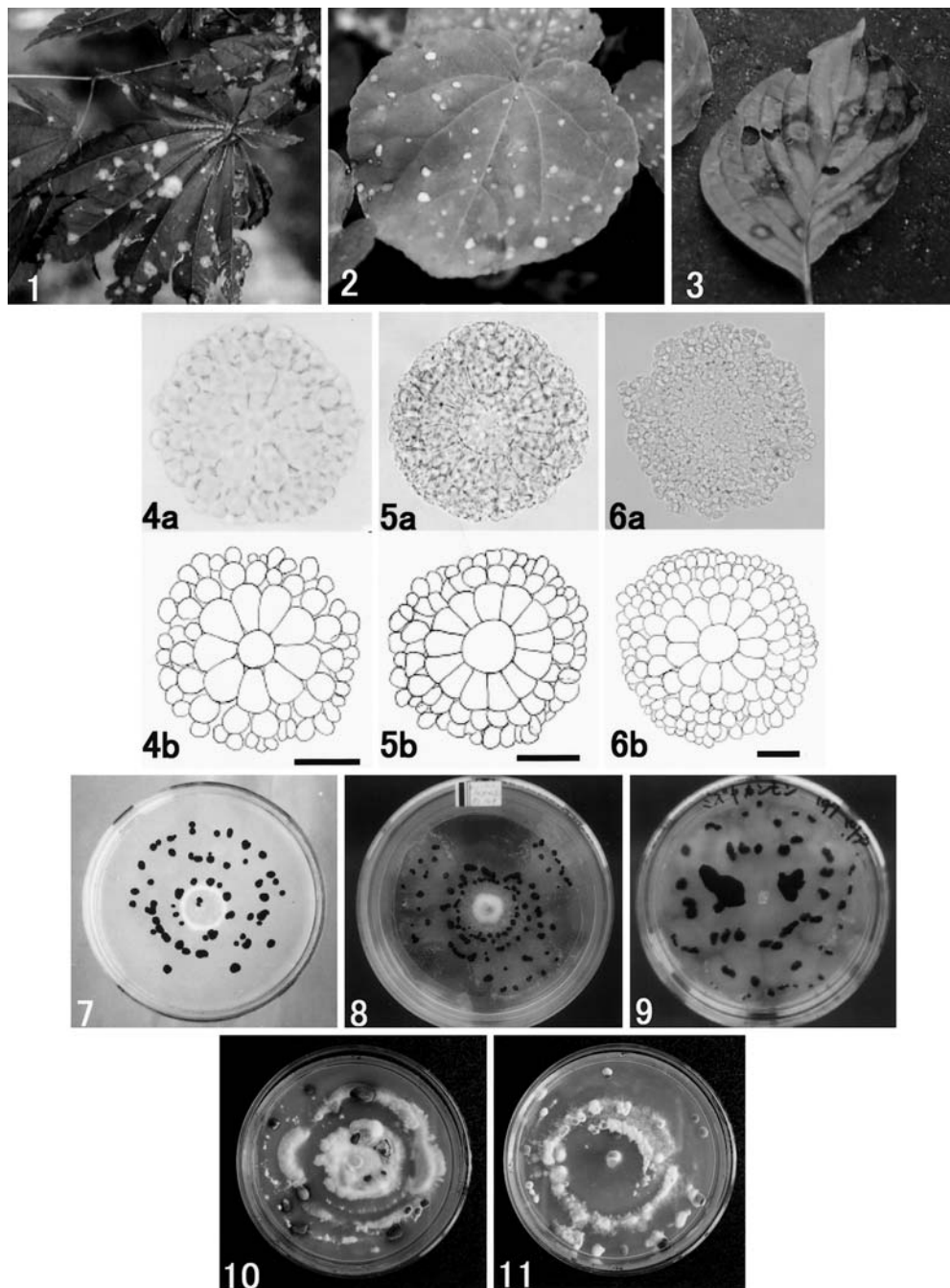


Table 1. Fungus isolates used in inoculation and culture experiments

Isolate no. (JCM)*	Species	Original host	Isolation source	Collection site
14252	<i>Cristulariella depraedans</i>	<i>Acer japonicum</i>	Ascospores	Hakkoda, Aomori Prefecture
14248	<i>Cristulariella</i> sp.	<i>Cercidiphyllum japonicum</i>	Tissue of apothecium	Hirosaki, Aomori Prefecture
14249	<i>Cristulariella</i> sp.	<i>Cornus controversa</i>	Conidia	Hakkoda, Aomori Prefecture
14250	<i>Cristulariella moricola</i>	<i>Acer negundo</i>	Conidia	Tomakomai, Hokkaido
14251	<i>Cristulariella pruni</i>	<i>Prunus mume</i>	Conidia	Gonohe, Aomori Prefecture

*JCM: Japan Collection of Microorganisms, Riken Bioresource Center, Wako, Japan

Isolation from ascospore: Part of apothecial discs were set with hymenium downward on the inner surface of the lid of a Petri dish, which contained plain agar plus 100ppm streptomycin sulfate. The lid was turned a little every 2h to obtain fallen ascospores on different parts of the agar surface. Under a light microscope, germinating ascospores were isolated with a fine needle and transferred to potato sucrose agar (PSA) (potato 200g, sucrose 20g, agar 20g, distilled water 1000ml) slants and grown under alternating dark and light (12:12 L:D) (15W fluorescent lamp; 1500lux) at 15°C for subsequent use.

Isolation from apothecial tissues: Part of the tissue was cut from an apothecial stipe and placed on the PSA plate under 12:12 L:D at 15°C. Several days later, agar pieces with mycelia were transferred to PSA slants for subsequent use.

Isolation from conidia: Conidia naturally occurring on infected leaves were singly transferred with a fine needle onto plain agar plus 100ppm streptomycin sulfate in a Petri dish and grown under 12:12 L:D at 15°C. Several days later, agar pieces with mycelia were transferred to PSA slants for subsequent use.

Inoculation experiments

A few conidia of each *Cristulariella* species were taken with a fine needle from diseased plants and placed on marked areas (four parts per leaf) of detached leaves of *Acer japonicum*, *Cercidiphyllum japonicum*, and *Cornus controversa* in large Petri dishes (18–21 cm in diameter) with moist filter paper, which were then kept for observation under 12:12 L:D at 15°C.

Cultural experiments

Observation on structure of sclerotia. Sclerotia of each *Cristulariella* species produced on PSA plates were sectioned with a freezing microtome (MICROM HM 400R) at 8µm thick, and the sections were mounted in lactophenol for observation under a light microscope.

Temperature response for mycelial growth. Agar pieces with mycelia of each *Cristulariella* species were placed on PSA plates (9cm) and grown under 12:12 L:D at various temperatures ranging from 5° to 30°C for 24 days.

Production of apothecia

Leaves infected with each *Cristulariella* species were collected from the field and brought to the experimental plot at the Hirosaki University campus. They were each put in a wire-netting box, placed on the ground in the shade, and occasionally sprayed with water to prevent excess drying. In December, when sclerotia had been formed on the veins, the leaves were put on moist quartz sand in large Petri dishes (24 cm in diameter), which were then kept at 0°C in the dark for cold treatment. Sclerotia were observed every month for germination. When sclerotia began to germinate, the dishes were transferred to 5°C under 12:12 L:D for maturation of apothecia. The low-temperature treatment was adopted from Harada (1977) and Noro (1988).

Morphological observation on apothecia and conidia

Apothecia were cut into sections (8–10µm thick) on a freezing microtome, the sections being mounted in lactophenol, cotton blue lactophenol, and Melzer reagent. Conidia of each fungus species were taken from infected host leaves using a fine needle, put into drops of FAA(II) solution for a moment to remove the air, and then mounted in lactophenol. Preparations of apothecia and conidia were observed under a light microscope at magnifications of 150, 600, and 1500.

Results

Pathogenicity

Results of inoculation experiments are shown in Table 2. In 2 days after inoculation, *Crist. depraedans* 14252 from *Acer japonicum* incited small brown spots on *A. japonicum* leaves at all the inoculated parts. The spots gradually enlarged to cause entire leaf blight within 3 weeks, producing conidia on blade and late sclerotia on the vein. In contrast, on *Cercidiphyllum japonicum* and *Cornus controversa* leaves, spots remained very small (less than 3mm in diameter) in 3 weeks without forming conidia or sclerotia.

Cristulariella sp. 14248 from *Cercidiphyllum* (*Cer.*) *japonicum* incited small spots on *Cer. japonicum* leaves at

Table 2. Results of cross-inoculation of host plants with *Cristulariella* species

Fungus species (isolate no.)	Original host	Plant species inoculated		
		<i>Acer japonicum</i>	<i>Cercidiphyllum japonicum</i>	<i>Cornus controversa</i>
<i>Cristulariella depraedans</i> (14252)	<i>Acer japonicum</i>	+++ ^a , C ^b , S ^c	+	++
<i>Cristulariella</i> sp. (14248)	<i>Cercidiphyllum japonicum</i>	— ^d	+++ S	+
<i>Cristulariella</i> sp. (14249)	<i>Cornus controversa</i>	+	++	+++ S

^a+++ , large brown spot; ++, small spot; +, very small spot incited, respectively

^bC, conidia formed

^cS, sclerotia

^d—, no symptoms produced

about half of the inoculated parts in 2 days after inoculation. The spots enlarged to cause blight extending over half of the leaf in 3 weeks with sclerotia formed on the vein. However, the spots remained very small (less than 2 mm in diameter) on *Cornus controversa* leaves without forming conidia and sclerotia.

No spots occurred on *A. japonicum*.

Cristulariella sp. 14249 from *Cornus controversa* incited spots 1–3 mm in diameter in 5 days, the spots gradually enlarged to form lesions of medium size (5–10 mm in diameter) in 3 weeks with sclerotia formed on the vein. The spots on *Cercidiphyllum japonicum* and *Acer japonicum* remained small (1–2 mm in diameter) in 3 weeks, with no conidia or sclerotia.

Cultural characteristics

Sclerotium formation. *Cristulariella depraedans* 14252 from *A. japonicum*: Colonies on PSA plates grown for 30–45 days at 15°C were smooth and whitish with well-developed submerged hyphae, frequently colored pale yellow in the center. Black, irregularly shaped, flatly submerged sclerotia, 1.5–4 × 1–4 mm in diameter, were produced in the agar (see Fig. 7). No conidia were produced. Microconidia were produced on the agar surface near the sclerotia.

Cristulariella sp. 14248 from *Cer. japonicum*: Colonies on PSA plates grown for 30–45 days at 15°C were smooth and white with well-developed submerged hyphae, frequently colored pale brown in the center. Black, irregularly shaped, flatly submerged sclerotia, 0.5–4 × 0.5–3 mm in diameter, were produced in the agar (see Fig. 8). No conidia were produced. Microconidia were produced on the agar surface near the sclerotia.

Cristulariella sp. 14249 from *Cornus (Cor.) controversa*: Colonies on PSA plates grown for 30–45 days at 15°C were smooth and white with well-developed submerged hyphae. Black, irregularly shaped, flatly submerged sclerotia, (2–)4–13(–18) × (1–)3–7(–11) mm in diameter, were produced in the agar (see Fig. 9). No conidia were produced. Microconidia were produced on the agar surface near the sclerotia.

Temperature responses for mycelial growth of each fungus are shown in Fig. 12. Optimum temperature for mycelial growth was 15°C in all fungi. The growth of *Cristulariella* sp. 14249 from *Cornus (Cor.) controversa* was fastest, followed in turn by *Crist. depraedans* 14252 from *A. japonicum* and *Cristulariella* sp. 14248 from *Cer. japonicum*.

Apothecium production. *Cristulariella depraedans* on *A. japonicum*: Naturally infected leaves of *A. japonicum* were collected on Sept. 11, 1998, at Jogakura, Aomori city, and placed on the ground in Hirosaki until Dec. 24 of that year, when sclerotia of the fungus were formed on veins of the leaves. Then, the leaves were transferred to being kept at 0°C in the dark. Some of the sclerotia germinated to produce apothecial initials in 6 months, and the apothecial initials developed into mature apothecia after 3–4 weeks incubation under 12:12 L:D at 5°C (see Fig. 13).

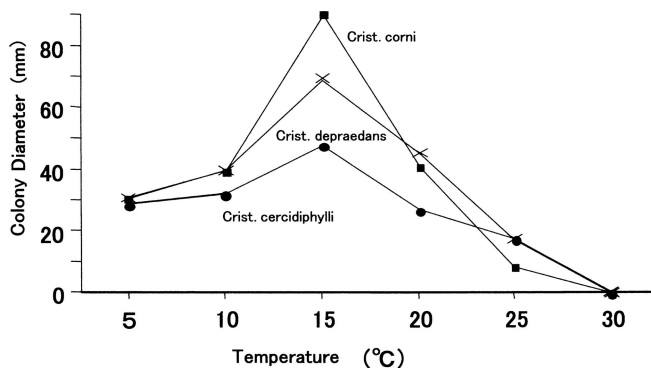


Fig. 12. Temperature relationships for colony growth of three *Cristulariella (Crist.)* spp. on potato sucrose agar (PSA) plates at different temperatures under 12:12 L:D for 24 days

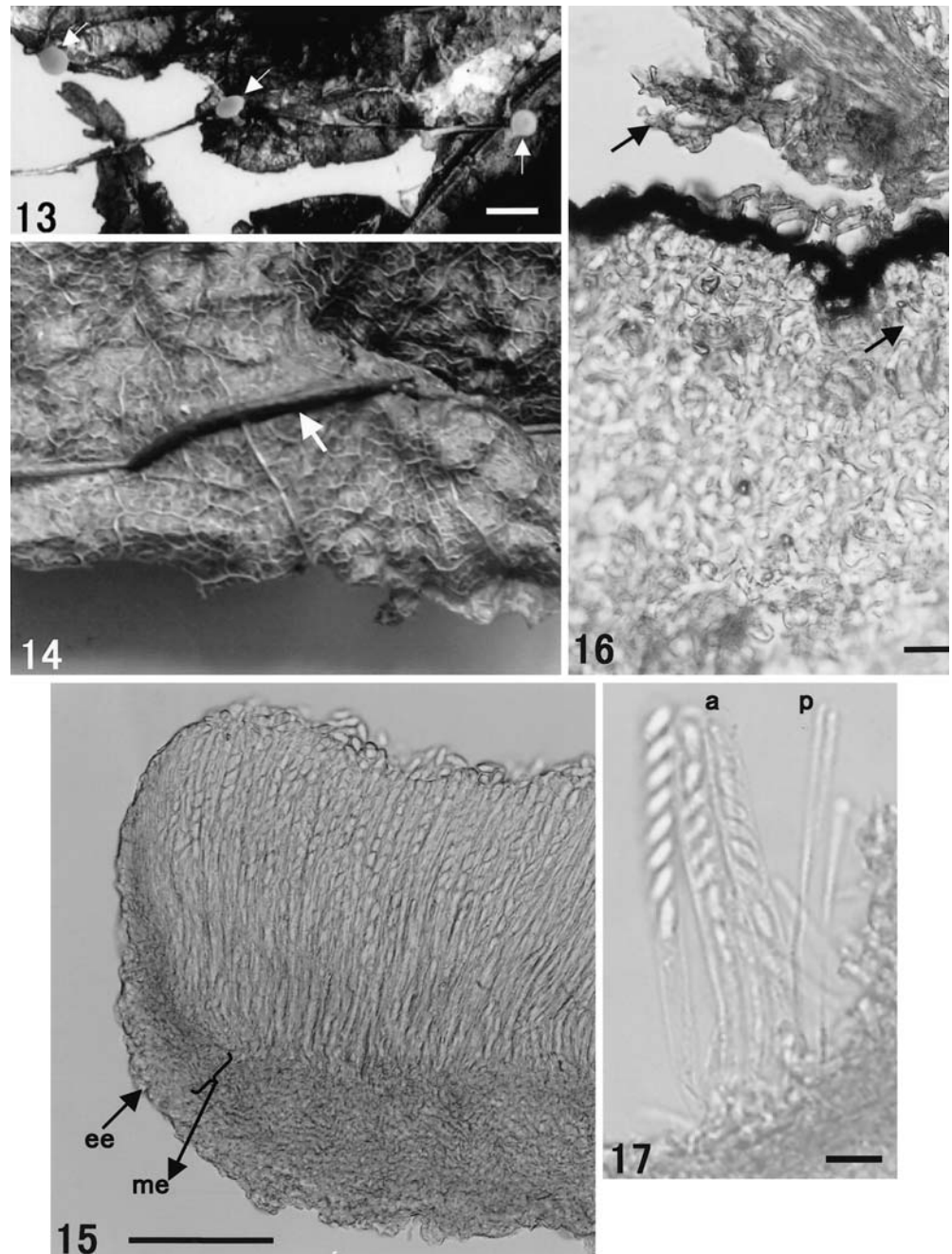
Cristulariella sp. on *Cer. japonicum*: Naturally infected leaves of *Cer. japonicum* were collected on Aug. 25, 1998, at Tsuta, Towada City, and placed on the ground in Hirosaki until Dec. 1 of that year, when sclerotia of the fungus were formed on veins of the leaves. Then, the leaves were kept at 0°C in the dark. Some of the sclerotia germinated to produce apothecial initials in 5 months, and the apothecial initials developed into mature apothecia after 3–4 weeks incubation under 12:12 L:D at 5°C (see Fig. 18).

Cristulariella sp. on *Cor. controversa*: The infected leaves of *Cor. controversa* were collected on Sept. 17, 1996, at Hokkaido University Experiment Forest in Tomakomai and placed on the ground in Hirosaki until late December. Sclerotia were formed on the leaves; then the leaves were kept at 0°C in the dark. No germination from the sclerotia was observed, even in 1 year of incubation at 0°C.

Morphology

Cristulariella depraedans on *A. japonicum*: Conidiophores cylindrical, septate. Conidia solitary on a conidiophore, globose, slightly depressed, 120–180 µm in diameter (Fig. 4a,b), consisting of a globose central cell 35–50 µm in diameter, whorl cells 30–50 × 25–35 µm, and secondary and tertiary cells 15–20 µm in diameter. Sclerotia black, fusiform to linear on leaf vein, 2–5.5(–10) × 0.5–1 mm, composed of a black rind with 2–3 layers of globose or subglobose cells 3.8–5.8 × 4.2–5.6 µm wide and white medulla consisting of interwoven mycelia 3.8–5.2 µm wide, intermixed with host tissues (see Figs. 14, 16). Teleomorph: Apothecia arising from substratal sclerotia formed in vein of infected leaves, carnose, brown, cup-shaped, 1–2.5 mm in diameter, stipitate, 2–4 mm high. Ectal excipulum 17.3–28.3 µm wide and consisting of subglobose to angular cells 3.5–8.7 × 4.7–7.2 µm. Medullary excipulum 44.9–51.8 µm wide and consisting of interwoven hyphae 2.5–4.8 µm wide. Asci clavate, 85–115 × 5–8 µm, apical pores J+. Paraphyses numerous, filiform, septate, 80–120 × 2.5–2 µm. Ascospores oblong to ellipsoid, continuous, hyaline, 9.5–11.5 × 2.5–5.5 µm, often germinating in the ascus to produce microconidia. Microconidia globose, 2.5–3 µm in diameter (see Figs. 13, 15, 17, 25, 26).

Figs. 13–17. Apothecial and sclerotial morphology of *Nervostroma depraedans*. **13** Apothecia (arrows) on infected leaves of *Acer japonicum*. **14** Sclerotia (arrow) on leaf vein of *A. japonicum*. **15** Section of apothecium. Arrows indicate ectal excipulum (*ee*) and medullary excipulum (*me*). **16** Section of the sclerotium. Arrows indicate host tissues. **17** Asci with ascospores (*a*) and paraphyses (*p*). Bars **13** 5 mm; **15** 50 μ m; **16** 10 μ m; **17** 10 μ m

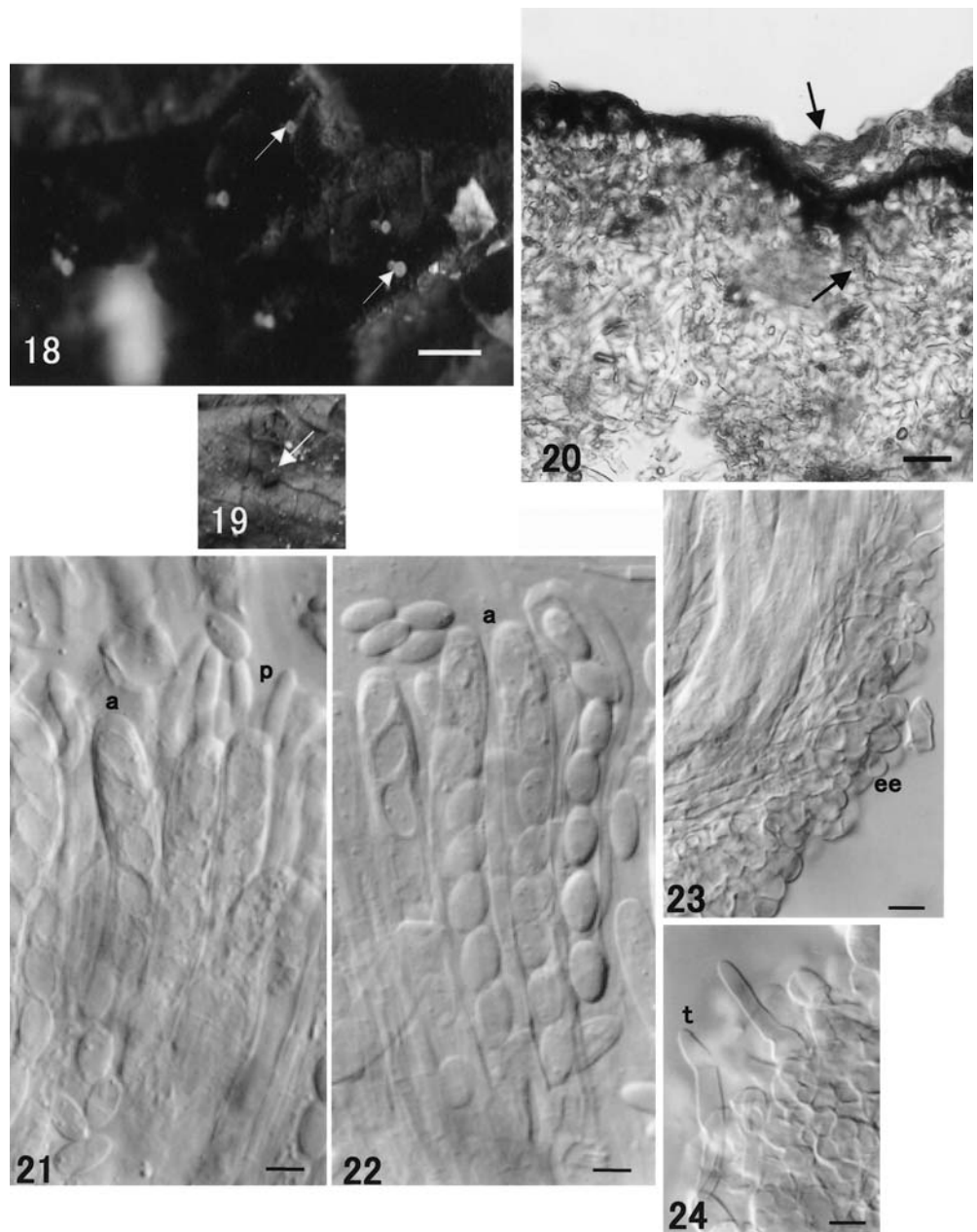


Cristulariella sp. on *Cer. japonicum*: Conidiophores cylindrical, septate. Conidia solitary on a conidiophore, globose, slightly depressed, (110–)120–130(–140) μ m in diameter (Fig. 5a,b), consisting of a globose central cell 35–50 μ m in diameter, whorl cells 25–40 \times 17–30 μ m, and secondary and tertiary cells 10–16 μ m in diameter. Sclerotia black, fusiform to linear on leaf vein, 0.6–3.6 \times 0.3–0.8 mm, composed of black rind with 2–3 layers of globose or subglobose cells, 3.8–6.1 \times 3.4–4.2 μ m wide, and white medulla consisting of interwoven mycelia 3.5–4.2 μ m wide, intermixed with host tissues (see Figs. 19, 20). Teleomorph: Apothecia arising from substratal sclerotia formed in vein of infected leaves, carnose, brown, cup-shaped, 0.2–0.8(–2) mm in diameter, 0.9–1.5 mm high, stipitate. Ectal

excipulum of 2 layers, outer layer consisting of subglobose to angular cells, 10–15 \times 5–10 μ m, sometimes with tomentum hyphae up to 30 μ m long, inner layer consisting of slightly granulate dark-colored hyphae 3–5 μ m wide. Asci clavate, 50–110 \times 5–7.5 μ m, apical pores weakly J+. Paraphyses numerous, filiform, septate, 45–72 \times 2.5–3 μ m. Ascospores oblong ellipsoid, continuous, hyaline, 7.5–10 \times 2.5–2.8 μ m, often germinating in asci to produce microconidia globose, 2.5–3 μ m in diameter (see Figs. 18, 21–24, 27).

Cristulariella sp. on *Cor. controversa*: Conidiophores cylindrical, septate. Conidia solitary on a conidiophore, globose, slightly depressed, 225–325 μ m in diameter (Fig. 6a,b), consisting of a globose central cell 32–48 μ m in diameter,

Figs. 18–24. Apothecial and sclerotial morphology of *Nervostroma cercidiphylli*. **18** Apothecia (arrows) on leaves of *Cercidiphyllum* (*Cerc.*) *japonicum*. **19** Sclerotia (arrow) on leaf vein of *Cerc. japonicum*. **20** Section of the sclerotium. Arrows indicate host tissues. **21** Asci (*a*) and paraphyses (*p*). **22** Asci (*a*) with ascospores. **23** Ectal excipulum (*ee*). **24** Tomentum hyphae (*t*). Bars **18** 5 mm; **20** 10 μm; **21, 22** 5 μm; **23, 24** 10 μm



whorl cells $40\text{--}60 \times 25\text{--}35\ \mu\text{m}$, secondary globose cells $22\text{--}38 \times 20\text{--}30\text{--}(33)\ \mu\text{m}$ in diameter, tertiary cells $12\text{--}25\ \mu\text{m}$ in diameter, and fourth cells $10\text{--}16\ \mu\text{m}$ in diameter.

Taxonomy

Cristulariella cercidiphylli Narumi-Saito et Y. Harada, sp. nov.

Conidiophora cylindrica, simplicia, septata. Conidiae ad apicem conidiophorum nascentes, solitariae, globosae, leniter depressae, $(110\text{--})120\text{--}130\text{--}(140)\ \mu\text{m}$ diametro, ex cellula centrali globosa $35\text{--}50\ \mu\text{m}$ diametro, cellis verticillatis $25\text{--}40 \times 17\text{--}30\ \mu\text{m}$ et cellulis secundariae tertiariaeque $10\text{--}16\ \mu\text{m}$ diametro compositae. Coloniae in

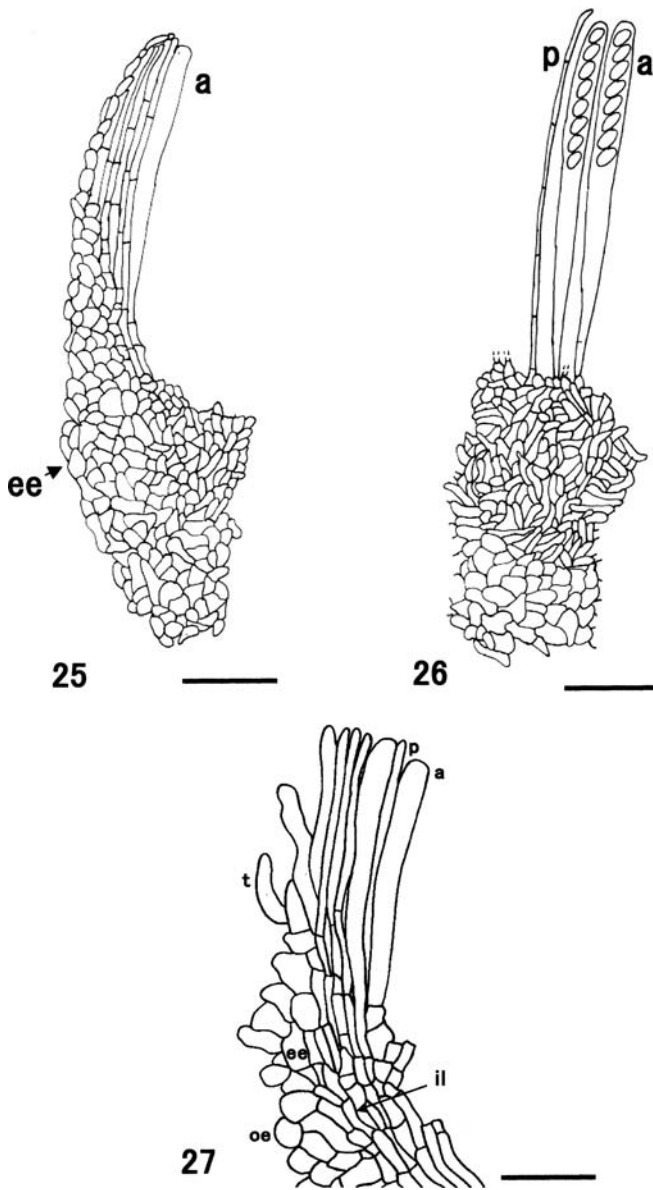
PSA ad 15°C laeves, hyalinae, sclerotiiis melanis globosis vel subglobosis irregularibus, $0.5\text{--}4 \times 0.5\text{--}3\ \text{mm}$ submersis producentes. Microconidia globosa, $2.5\text{--}3\ \mu\text{m}$ diametro.

Holotypus. Japan. Honshu: Oirase, Towada, Aomori Prefecture, on leaves of *Cercidiphyllum japonicum* Siebold & Zucc., kept in the Herbarium of Hirosaki University, Fungi (HHUF24768).

Etymology: *cercidiphylli*, from the generic name of the host plant.

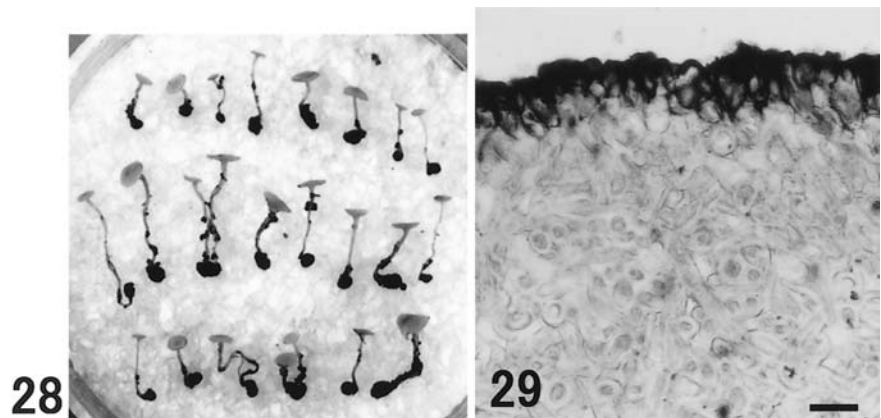
Cristulariella corni Narumi-Saito et Y. Harada, sp. nov.

Conidiophora cylindrica, simplicia, septata. Conidiae ad apicem conidiophorum nascentes, solitariae, globosae, leniter depressae, $225\text{--}325\ \mu\text{m}$ diametro, ex cellula centrali globosa $32\text{--}48\ \mu\text{m}$ diametro, cellis verticillatis $40\text{--}60 \times 25\text{--}$



Figs. 25–27. Line drawings of apothecia of *Nervostroma depraedans* and *N. cercidiphyllum*. **25** Ectal excipulum (*ee*) of apothecia of *N. depraedans*. **26** Middle part of apothecia of *N. depraedans*, showing asci (*a*) and paraphyses (*p*). **27** Ectal excipulum (*ee*), outer layer (*oe*), inner layer (*il*) and tomentum hyphae (*t*) of apothecia of *N. cercidiphylli*. Bars 25 μ m

Figs. 28, 29. Apothecial and sclerotial morphology of *Grovesinia pyramidalis*. **28** Apothecia of *G. pyramidalis*. **29** Section of a sclerotium. Bar **29** 25 μ m



35 μ m, cellulis secundariis globosis 22–38 \times 20–30(–33) μ m diametro, tertiariae 12–25 μ m diametro et quartiis 10–16 μ m compositae. Coloniae in PSA ad 15°C laeves, hyalinae, sclerotiiis melanis globosis vel subglobosis (2–)4–13(–18) \times (1–)3–7(–11)mm submersis producentes. Microconidia globosa, 2.5–3 μ m diametro.

Holotypus. Japan. Hokkaido: Tomakomai Experiment Forest, Hokkaido University, on leaves of *Cornus controversa* Hemsl., kept in the Herbarium of Hirosaki University, Fungi (HHUF23729).

Etymology: *corni*, from the generic name of the host plant.

Hinomyces Narumi-Saito et Y. Harada, anam. gen. nov.
Misapplied name: *Cristulariella* sensu Redhead (1975) non-Höhn., pro parte.

Conidia conoidea in folia, ex cellulis globosis circum conidiophorum copiose composita. Sclerotia nigra, globosa vel subglobosa.

Etymology: In honor of the late Iwao Hino, who first described the type species of the genus under the name of *Botrytis moricola*.

The type species: *Hinomyces moricola* (I. Hino) Narumi-Saito et Y. Harada.

Hinomyces moricola (I. Hino) Narumi-Saito et Y. Harada, comb. nov.

Basionym: *Botrytis moricola* I. Hino, Bull. Miyazaki College Agric. For. 1:80, 1929.

\equiv *Cristulariella moricola* (I. Hino) Redhead, Mycologia 71: 1249, 1974.

Hinomyces pruni (Y. Harada et Noro) Narumi-Saito et Y. Harada, comb. nov.

Basionym: *Cristulariella pruni* Y. Harada et Noro, Trans. Mycol. Soc. Jpn 29:86, 1988.

Nervostroma Narumi-Saito et Y. Harada, gen. nov.

Sclerotia in nervis folii hospitalis formata, fusiformia vel subglobosa, nigra. Apothecia a sclerotia surgentia, ad maturitate brunnea, carnea, calathino-disciformia, stipitata. Asci cylindrici, stipitati, octospori, J+. Paraphysis filiformes, septatae. Ascosporae oblongae vel ovoideae, unicellulares, hyalinae.

Etymology: *nervostroma*, *nervus* (vein) + *stroma*, thus meaning stroma on leaf vein, from the sclerotial morphology.

The type species: *Nervostroma depraedans* Narumi-Saito et Y. Harada

Nervostroma depraedans Narumi-Saito et Y. Harada, sp. nov.

Sclerotia in nervi folii formata, nigra, fusiformia, 2–5.5(–10) × 0.5–1 mm. Apothecia a sclerotia surgentia, ad maturitate brunnea, carnea, calathino-disciformia, stipitata, 1–2.5 mm diametro. Excipulum ectale 17.3–28.3 μm crassum, ex cellulis globosis compositam. Asci cylindrici, stipitati, octospori, 85–115 × 5–8 μm, J+. Paraphysis filiformes, septatae, 80–120 × 2.5–2.8 μm. Ascospores unistichae, 9.5–11.5 × 2.5–5.5 μm, oblongo-ovoideae, unicellulares, hyalinae, saepe intra ascum germinantes et microconidia formantes. Microconidia globosa, 2.5–3 μm diametro.

Holotypus. Japan. Honshu: Produced in the laboratory on infected leaves of *Acer japonicum* Thunb. collected at Jogakura, Hakkoda, Aomori Prefecture; kept in the Herbarium of Hirosaki University, Fungi (HHUF25294).

Anamorphic state: *Cristulariella depraedans* (Cooke) Höhn.

Nervostroma cercidiphylli Narumi-Saito et Y. Harada, sp. nov.

Sclerotia in nervi folii formantia, nigra, fusiformia vel subglobosa, 0.6–3.6 × 0.3–0.8 mm. Apothecia a sclerotia surgentia, ad maturitate brunnea, carnea, calathino-disciformia, stipitata, 0.2–0.8(–2) mm diametro. Asci cylindrici, stipitati, octospori, 50–110 × 5–7.5 μm, dilute J+ (saturate J+ ad adjunctionem KOH). Paraphysis fusiformes, septatae, 45–72 × 2.5–3 μm. Ascospores unistichae, 7.5–10 × 2.5–2.8 μm, oblongo-ovoideae, unicellulares, hyalina, intra ascum germinantes et microconidia formantes. Microconidia globosa, 2.5–3 μm diametro.

Holotypus. Japan. Honshu: Produced in the laboratory on infected leaves of *Cercidiphyllum japonica* Siebold & Zucc. collected at Tsuta spa, Hakkoda, Aomori Prefecture; kept in the Herbarium of Hirosaki University, Fungi (HHUF 25310).

Etymology: *cercidiphylli*, from the generic name of the host plant.

Anamorphic state: *Cristulariella cercidiphylli* Narumi-Saito et Y. Harada

Discussion

Species of *Cristulariella* Höhn. emend. Redhead are anamorphic fungi causing zonate leaf spot diseases of herbaceous and woody plants with hyaline multicellular propagules, either globose or conoid in whole shape, on the spots. So far, three species have been described in the genus: *Crist. depraedans*, *Crist. moricola*, and *Crist. pruni*. It was proved that the species do not belong to a natural taxonomic group because *Crist. depraedans* produces glo-

Table 3. Comparisons in teleomorphs of *Nervostroma* and *Grovesinia*

Fungus species	Type of sclerotia		Apothecia		Ascospore (μm)		Conidia		References
	Substratal	True	Disc (mm in diameter)	Ascus (μm)	Ascospore (μm)	Whole shape	Size (μm)		
<i>Nervostroma depraedans</i> anam. <i>Cristulariella depraedans</i>	Substratal		1–2.5	85–115 × 5–8	9.5–11.5 × 2.5–5.5	Globose (slightly depressed)	120–180	This study	
<i>Nervostroma cercidiphylli</i> anam. <i>Cristulariella cercidiphylli</i>	Substratal		0.2–2	50–110 × 5–7.5	5–8 × 5–5.5	Globose (slightly depressed)	(100–)120–130(–140)	This study	
anam. <i>Cristulariella corni</i> ^a	Substratal		–	–	–	Globose (slightly depressed)	225–325	This study	
<i>Grovesinia pyramidalis</i> anam. <i>Hinomyces moricola</i>	True		(2–)3–5(–8)	(115–)113–150(–176) × 6–8	10–12(–15) × 4–5	Conoid	250–450 × 80–120	Cline et al. (1983) Waterman and Marshall (1947)	
<i>Grovesinia pruni</i> anam. <i>Hinomyces pruni</i>	True		(1–)2–4(–5)	140–180 × 9–13	(9.5–)11–15 × (6–)7–7.5	Conoid	150–190(–210) × 80–150	Harada and Noro (1988)	

^a Apothecial state is unknown

bose conidia, substratal sclerotia firmly attached to the host tissue, comprising fungus mycelia and host tissue (tentatively referred to group 1), while *Crist. moricola* and *Crist. pruni* produce conoid conidia, true sclerotia that comprise fungus mycelia only, freed from host tissues (group 2). In this study, two additional species, i.e., *Crist. cercidiphylli* on *Cercidiphyllum japonicum* and *Crist. corni* on *Cornus controversa*, both belonging to group 1, were newly described. Then, new taxonomic treatment of the genus *Cristulariella* was proposed; *Cristulariella* should be retained for *Crist. depraedans*, the type species, and two additional new species. A new genus, *Hinomyces*, was established to accommodate *Crist. moricola* and *Crist. pruni*.

Teleomorphs of *Crist. depraedans* and *Cristulariella* sp. on *Cer. japonicum* were newly found, which should properly be placed in the Sclerotiniaceae, on the basis of apothecia arising from substratal sclerotia formed in leaf veins of infected plants. However, the teleomorphs of both fungi could not be placed in the genus *Grovesinia*, to which the anamorphs of *Crist. moricola* and *Crist. pruni* belong (see Table 3). They appeared to be rather close to *Valdensinia* Pyronel, *Septotinia* Whetzel ex J.W. Grows et M.E. Elliott, *Seaverinia* Whetzel, and other related genera in Korf's key (1973), all of which, however, have different anamorphs. Apparently, apothecia of known *Grovesinia* species are much larger than those of *Crist. depraedans* and *Crist. cercidiphylli* (Table 3). Based on these findings, a new genus *Nervostroma* was erected to accommodate the teleomorphs of *Crist. depraedans* and *Crist. cercidiphylli*.

On PSA plates, species of *Nervostroma* produced colonies with submerged mycelia with scant aerial hyphae and substratal sclerotia that could not easily be removed from the agar. In contrast, species of *Grovesinia* produced colonies with aerial hyphae, and then with true sclerotia, which freely come off the agar when mature (see Figs. 7–11, 28, 29).

In inoculation experiments, species of *Cristulariella* were shown to cause spots with conidia and sclerotia on their respective host plants. The results suggested that each species may complete their life cycles on their host plants only, as they could overwinter in the form of sclerotia, which in turn do form apothecia in spring to discharge ascospores as the primary inocula.

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