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FULL PAPER

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Pleosporales in Japan (4). The genus *Massariosphaeria*

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Abstract Six species of the genus *Massariosphaeria* collected from northern Japan are described and illustrated. *Massariosphaeria moricola* (on dead twigs of *Morus australis*) and *M. megaspora* (on submerged dead twigs of an unknown woody plant) are reported as new species. A new combination of *M. clavispora* (basionym: *Hysterium clavispora*) is proposed. Three species, *M. typhicola*, *M. roumeguerei*, and *M. grandispora*, are newly added to the mycoflora of Japan.

Key words *Chaetomastia* · Lophiostomataceae · Massarinaceae · *Massariosphaeria* · Pleosporales

Introduction

This is the fourth in a series dealing with taxonomy of Pleosporales fungi in Japan. We report six species of the genus *Massariosphaeria* (E. Müll.) Crivelli, of which two are new species, one is a new combination, and three are new to Japan.

The name *Massariosphaeria* was used for the first time by Müller (1950) as a section of *Leptosphaeria* Ces. & De Not. This group was characterized by its transversely septate ascospores, which are relatively large, thick walled, and surrounded by a prominent gelatinous sheath. Crivelli (1983) elevated the group to the generic rank and expanded it to include dictyosporous species. He emphasized cultural characteristics, such as purple-red pigments and type of anamorph produced, as well as the ascospore morphology noted by Müller (Huhndorf et al. 1990). This generic concept was accepted by Leuchtmann (1984), but was not by Barr (1989, 1990a,b).

Barr (1990a) transferred some dictyosporous species treated as *Massariosphaeria* by Crivelli to the genus *Karstenula* Speg., based on their melanommataceous centrum. Moreover, Barr (1989) proposed the genus *Chaetomastia* (Sacc.) Berl. as an earlier name for *Massariosphaeria*, and transferred some species of *Massariosphaeria* having a phaeophragmospore to *Chaetomastia*. However, Barr (2002) changed the concept of the genus, accepting only two species in *Chaetomastia* among the seven species previously recognized as *Chaetomastia* by Barr (1989). Since *M. phaeospora*, the type of the *Massariosphaeria*, was excluded from *Chaetomastia*, the genus *Massariosphaeria* is valid. At present, this genus is placed in the Lophiostomataceae (Kirk et al. 2001) or Massarinaceae (Barr 2002).

At first, we supposed that the hyalophragmosporous species and phaeophragmosporous species would belong to different genera, respectively. However, we could not find differences between these two taxa, based on the specimens so far studied by us. In this study, we treated phragmosporous species as *Massariosphaeria* without considering the ascospore coloration. We also did not accept the dictyosporous species that Crivelli (1983) disposed under the genus for the same reason as Barr (1990a).

There are at least eight phragmosporous species in this genus: 19 species are described in the monograph of Leuchtmann (1984), five additional species are proposed by Shoemaker and Babcock (1989), and six other species are reported by Ahn and Shearer (1998), Holm and Holm (1988), Huhndorf et al. (1990), Kohlmeyer et al. (1996), Leuchtmann (1987), and Scheuer (1988). Among them, *Massariosphaeria scirpina* (G. Winter) Leuchtm. has been transferred to *Massarina* Sacc. (Tanaka and Harada 2003b). In Japan, the genus *Massariosphaeria* is a taxon to which little attention has been paid, and no species has been described.

All specimens examined were maintained at the Herbarium of Hirosaki University, Fungi (HHUF), and all isolates were deposited at the culture collection of MAFF.

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Taxonomy

Massariosphaeria (E. Müll.) Crivelli, Dissert. ETH Nr. 7318: 141, 1983.

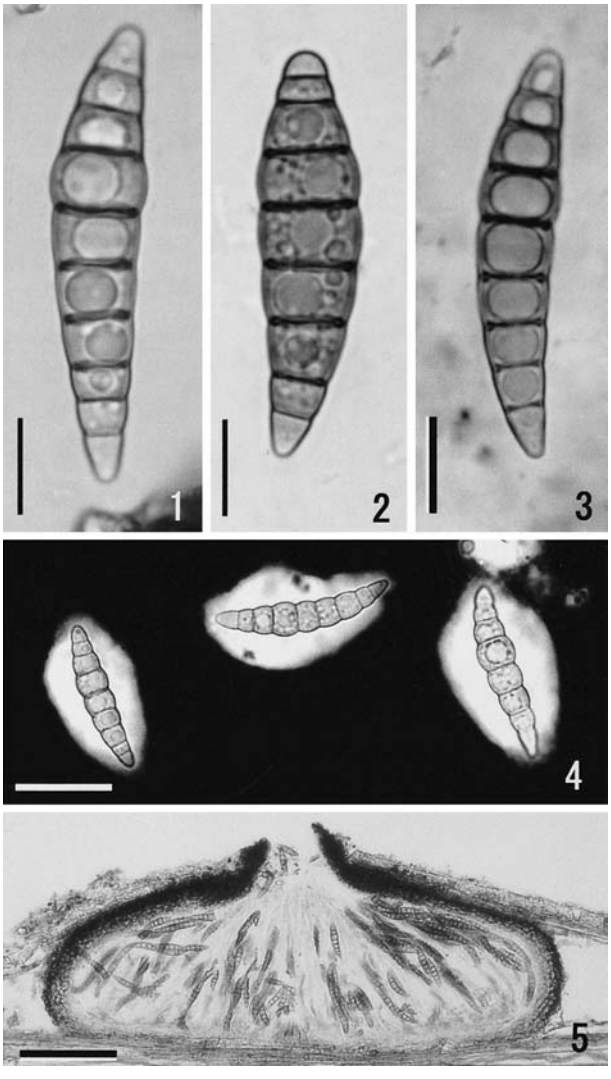
Basionym. *Leptosphaeria* sect. *Massariosphaeria* E. Müll., Sydowia 4: 206, 1950.

Type species. *Massariosphaeria phaeospora* (E. Müll.) Crivelli.

Anamorphs. *Aposphaeria* Sacc.-like (Leuchtmann 1984), *Phoma* Sacc.-like (Sivanesan 1984).

(1) ***Massariosphaeria typhicola*** (P. Karst.) Leuchtm., Sydowia 37: 168, 1985 ('1984'). Figs. 1–5, 46

≡*Leptosphaeria typhicola* P. Karst., Mycol. Fenn. 2: 100, 1873. ≡*Phaeosphaeria typhicola* (P. Karst.) Hedjar., Sydowia 22: 86, 1968. ≡*Chaetomastia typhicola* (P. Karst.) M.E. Barr, Mycotaxon 34: 514, 1989.



Figs. 1–5. *Massariosphaeria typhicola*. **1–4** Ascospores (**4** in india ink); **5** Ascoma. (**1** from HHUF 27785; **2** from HHUF 27779; **3** from MAFF 239220; **4** from HHUF 27786; **5** from HHUF 27781.) *Bars* **1–3** 10 μm; **4** 25 μm; **5** 100 μm

=*Leptosphaeria occidentalis* Ellis & Everh., Erythea 2: 20, 1894.

=*Leptosphaeria baldingerae* Fautrey & Lambotte, Rev. Mycol. (Toulouse) 19: 3, 1897. ≡*Phaeosphaeria baldingerae* (Fautrey & Lambotte) Hedjar., Sydowia 22: 87, 1968.

=*Leptosphaeria cladii* Cruchet, Bull. Soc. Vaud. Sci. Nat. 55: 161, 1923.

Ascomata 180–305 μm high, 215–570 μm diameter. Beak 45–85 μm high, 60–105 μm diameter. Ascomal wall 15–43 μm thick at sides, 5–23 μm thick at the base. Pseudoparaphyses cellular, 1.5–3 μm thick. Asci (80–) 82.5–115 (–157) × 14–19 (–21) μm (mean = 98.7 × 15.9 μm, n = 50). Ascospores (30.5–) 32–43 (–49) × 6.5–10.5 (–12.5) μm (mean = 38.8 × 8.5 μm, n = 115), L/W 3.8–5.5 (mean = 4.6, n = 103), (6–) 7–8 (–10)-septate (septa of upper hemisphere + the primary septum + septa of lower hemisphere = mostly 3 + 1 + 4), with the supramedian 0.38–0.49 (mean = 0.44, n = 103) primary septum, brown, verruculose, with a sheath when young. Ascospores germinating usually from both end cells, sometimes from the other cells.

Cultural characteristics. Colonies on potato dextrose agar (PDA; Difco, Detroit, MI, USA) 49 mm in diameter after incubation for 4 weeks at 20°C in the dark, Dull-Green (Kornerup and Wanscher 1978; 30E3); reverse Olive (3E3) or Dark-Green (25F3); no pigment produced. On rice straw agar (RSA; Tanaka and Harada 2003a), ascomata formed on the surface of rice straws within 2 months. Ascospores similar in shape and size to those found in nature.

Materials examined. On dead culms of *Phragmites australis* (Cav.) Trin. ex Steud.: Toyohira River, riverbank, Sapporo, Hokkaido, 141°21.49′ E, 43°02.23′ N, June 23, 2000, KT. 306-2 (HHUF 27768); Yokohamamachi, Kamikita, Aomori, 141°13′ E, 40°58′ N, Oct. 26, 2002, S. Hatakeyama 925 (HHUF 27769); Mohei Pond, Aoki, Hirosaki, Aomori, 140°26.25′ E, 40°34.12′ N, Dec. 7, 2002, N. Asama and KT. 963 (HHUF 27771). On culms of *Phragmites japonica* Steud.: Magarikawa, riverbank, Sakekawa, Mogami, Yamagata, 140°11′ E, 38°48′ N, Oct. 13, 2002, Y. Ooki and YH. 914 (HHUF 27772). On dead culms of *Dactylis glomerata* L.: Campus of Hirosaki University, Hirosaki, Aomori, 140°28′ E, 40°35′ N, June 21, 2001, KT. 548 (HHUF 27775). On dead stems of unknown herbaceous plants: Sanpinai, Hirosaki, Aomori, 140°30.13′ E, 40°34.51′ N, Aug. 12, 2001, KT. 667-3 (HHUF 27779, culture MAFF 239218); Oowasawa River, riverbank, Chitose Bridge, Kadoke, Hirosaki, Aomori, Aug. 25, 2001, KT. 708 (HHUF 27781). On dead twigs of unknown woody plants: Shibayachi Moor, Oodate, Akita, 140°34.5′ E, 40°19.1′ N, Oct. 5, 2001, Y. Ooki and KT. 797 (HHUF 27785, culture MAFF 239219); Oct. 5, 2002, KT. 906 (HHUF 27786); Oowasawa River, riverbank, Nakachitose Bridge, Shimizumori, Hirosaki, Aomori, 140°30.16′ E, 40°34.18′ N, Oct. 28, 2001, KT. 818 (HHUF 27787, culture MAFF 239220).

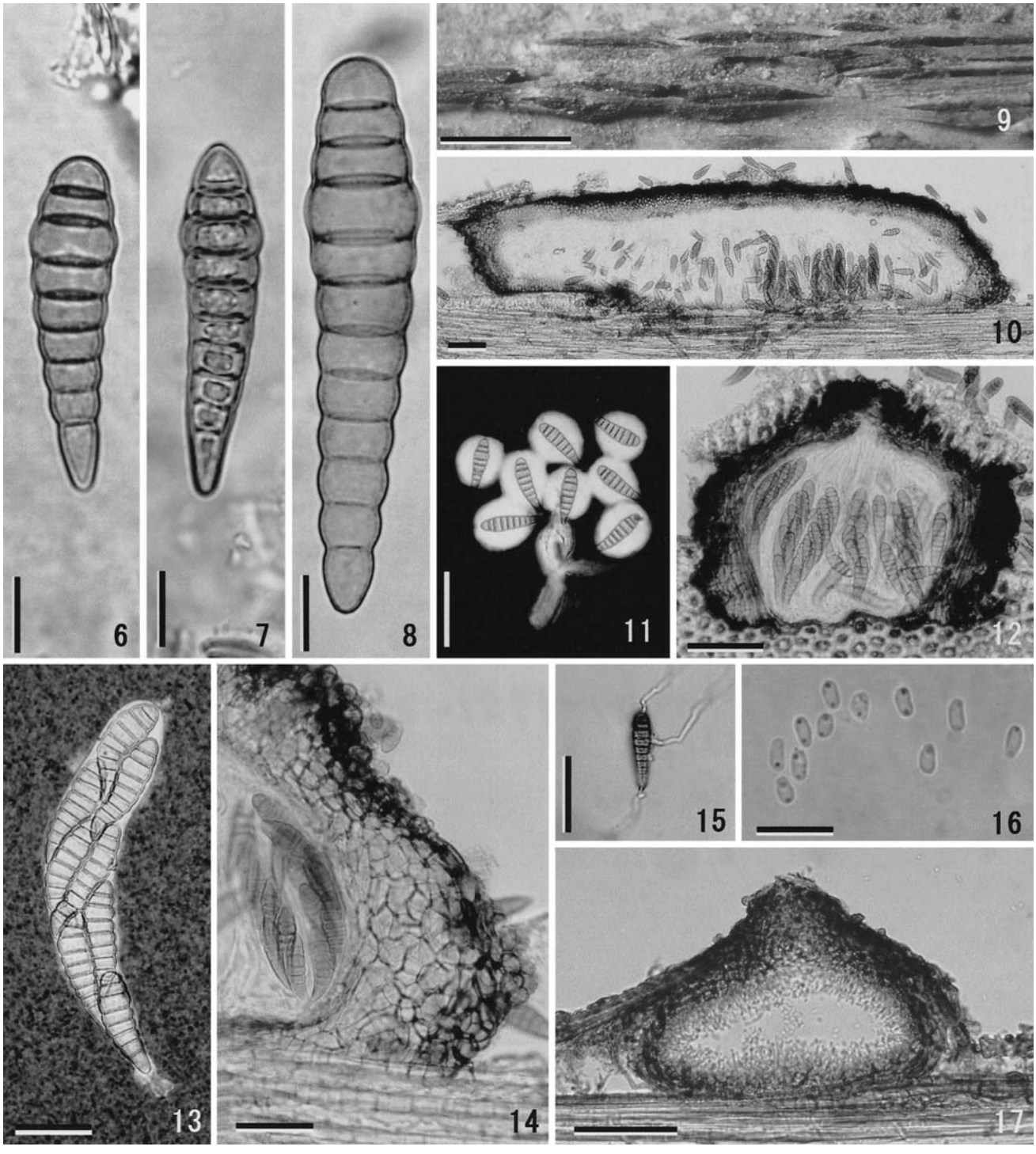
Notes. *Massariosphaeria typhicola* is a common species and occurs on old culms of various monocots (Barr 1989). In addition to land habitats, it is reported from both freshwater (Fallah and Shearer 2001) and the marine (Kohlmeyer and Volkmann-Kohlmeyer 1991) environment. Lucas and Webster (1967) reported formation of an anamorph of this

fungus from cultural studies, and Sivanesan (1984) treated it as *Phoma*-like. Leuchtmann (1984) also found both anamorph and teleomorph from different isolates, and described the conidial state as *Aposphaeria*-like. However, all our isolates formed only ascomata in culture, as was noted by Fallah and Shearer (2001).

(2) ***Massariosphaeria clavispora*** (Cooke & Peck) Kaz. Tanaka & Y. Harada, comb. nov. Figs. 6–17, 47, 52

Basionym: *Hysterium clavisporum* Cooke & Peck, Bull. Buffalo Soc. Nat. Sci. 3: 34, 1875; Ann. Rep. New York State Museum 28: 69, 1876 ("1874").

≡*Dothidea clavispora* (Cooke & Peck) Peck, Ann. Rep.



Figs. 6–17. *Massariosphaeria clavispora.* **6–8** Ascospores (**8** abnormal-type ascospore); **9** Ascomata on host surface; **10** Ascoma in longitudinal section; **11** Ascospores (in india ink); **12** Ascoma in cross section; **13** Ascus (in india ink); **14** Ascomal wall in longitudinal section; **15** Germinating ascospore; **16** Conidia; **17** Conidioma in longitudinal section. (**6**, **10–15** from HHUF 28082; **7**, **8** from HHUF 28083; **9** from HHUF 28085; **16**, **17** from MAFF 239221.) *Bars* **6–8**, **16** 10 μm; **9** 1 mm; **10–12**, **15**, **17** 50 μm; **13**, **14** 25 μm

New York State Museum 29: 63, 1878 ("1875"). ≡*Rhopographus clavisporum* (Cooke & Peck) Sacc., Syll. Fung. 2: 648, 1883. ≡*Bruneaudia clavispora* (Cooke & Peck) Kuntze, Rev. Gen. Pl., 3: 447, 1898. ≡*Calospora clavispora* (Cooke & Peck) Theiss. & Syd., Ann. Mycol. 13: 428, 1915. ≡*Phaeosphaeria clavispora* (Cooke & Peck) M.E. Barr in Barr et al., Bull. New York State Museum 459: 15, 1986. ≡*Chaetomastia clavispora* (Cooke & Peck) M.E. Barr, Mycotaxon 34: 509, 1989.

=*Leptosphaeria clavicarpa* Ellis & Everh., J. Mycol. 1: 43, 1885. ≡*Heptameria clavicarpa* (Ellis & Everh.) Cooke, Grevillea, 18: 32, 1889.

Ascomata elongate, 150–260 μm high, 600–1100 μm long, 230–380 μm wide, immersed in host sheath, later superficial on bare culm, glabrous. Beak composed of globose to polygonal slightly thick-walled cells of 4–10 μm diameter. Ascomal wall textura anguralis, in cross section 32–88 μm thick and composed of parallel rows (6–8 layers) of rectangular to polygonal pale brown cells of 7.5–18 × 3.5–8.5 μm at sides, poorly developed at the base; in longitudinal section 40–65 μm thick at sides, composed of irregular row cells. Pseudoparaphyses cellular, 1.5–2.5 μm thick, septate, branched, and anastomosed. Asci (88–) 95–130 (–136.5) × (17–) 18.5–27 μm (mean = 115.2 × 22.0, n = 50), L/W 4.0–6.1 (mean = 5.3, n = 50), clavate, with an ocular chamber, short stalked (5–15 μm long), with 8 biseriate ascospores. Ascospores (31–) 34–48 (–53) × 8–13 μm (mean = 39.8 × 10.2 μm, n = 100), L/W 3.6–4.4 (mean = 3.9, n = 100), clavate, slightly curved, thick walled, (7–) 8 (–10)-septate (2 + 1 + 4, 2 + 1 + 5, 2 + 1 + 6, 3 + 1 + 5, 3 + 1 + 6; mostly 2 + 1 + 5), with the primary septum supramedian 0.29–0.36 (mean = 0.33, n = 100), the third (or fourth) cell from the apex enlarged, yellow to dark brown, without guttules, slightly echinulate. Ascospore sheath conspicuous, obovoid, 3–12 μm thick at sides.

Cultural characteristics. Colonies on PDA attaining 80 mm in diameter within 2 weeks at 20°C in the dark, Greenish-Grey (27D2); reverse Brownish-Grey (6F2); no pigment produced. On RSA, a *Aposphaeria*-like anamorph (sensu Leuchtmann 1984) formed on the surface of rice straws within 2 months. Conidiomata 75–145 μm high, 125–160 μm diameter, depressed globose. Beak 40–80 μm long, 50–85 μm wide, central, papillate. Conidiomatal wall at sides composed of 4–5 layers of polygonal to subglobose cells of 3–8.5 μm diameter. Conidiophores 4–7 (–9) × 2–3 μm, cylindrical to lageniform, simple, formed from all round the locular cavity. Conidiogenous cells phialidic. Conidia 3–5 × 1.5–2 μm, L/W 2.0, ellipsoid, aseptate, hyaline, smooth. Sometimes teleomorph produced but not mature within 4 months.

Materials examined. On dead culms of *Phragmites australis*: Shizukuishi River (Oota Bridge), Jyousei, Morioka, Iwate, 141°07.4′ E, 39°41.6′ N, Feb. 18, 2003, YH. 1013 (HHUF 28082, culture MAFF 239221); 1016 (HHUF 28083); 1017 (HHUF 28084); Maruyama Pond, Kizukuri, Nishitsugaru, Aomori, 140°18.3′ E, 40°50.3′ N, Apr. 26, 2003, KT. and N. Asama 1080 (HHUF 28085); 1081 (HHUF 28086); Kansuke Pond, Dougizaka, Kizukuri, Nishitsugaru, Aomori, 140°20.1′ E, 40°53.4′ N, Apr. 26, 2003, KT. and N. Asama 1085 (HHUF 28087).

Notes. This fungus was originally described as a species of *Hysterium* Pers., and the placement was accepted by Shoemaker and Babcock (1989). However, it does not fit within the generic concept of *Hysterium* in having ascomata of pseudoparenchymatous pale brown cells and asymmetrically septate ascospores. *Hysterium pulicare* Pers.:Fr., the type of the genus, is characterized by black, carbonaceous, elongate ascomata (referred to as hysterothecia), and ellipsoid symmetrically septate ascospores with the end cells usually paler than the two middle cells (Eriksson 1981). Although the elongate fruit bodies with a poorly defined slit are unusual among *Massariosphaeria*, we propose a new combination based on the ascospore morphology.

The specimens we examined are a good match to the descriptions by Barr (1989) and Shoemaker and Babcock (1989), but sometimes abnomal type ascospores (see Fig. 8) were observed; they were larger, (55–) 57–73 × 13–15 μm (mean = 64.3 × 14.3 μm, n = 30), with 9–12 septa (2 + 1 + 6, 3 + 1 + 5, 3 + 1 + 6, 3 + 1 + 7, 4 + 1 + 6, 4 + 1 + 7; mostly 3 + 1 + 6), and might be released from the 4-spored ascus. In the foregoing description, we excluded the measurements of this type of ascospore.

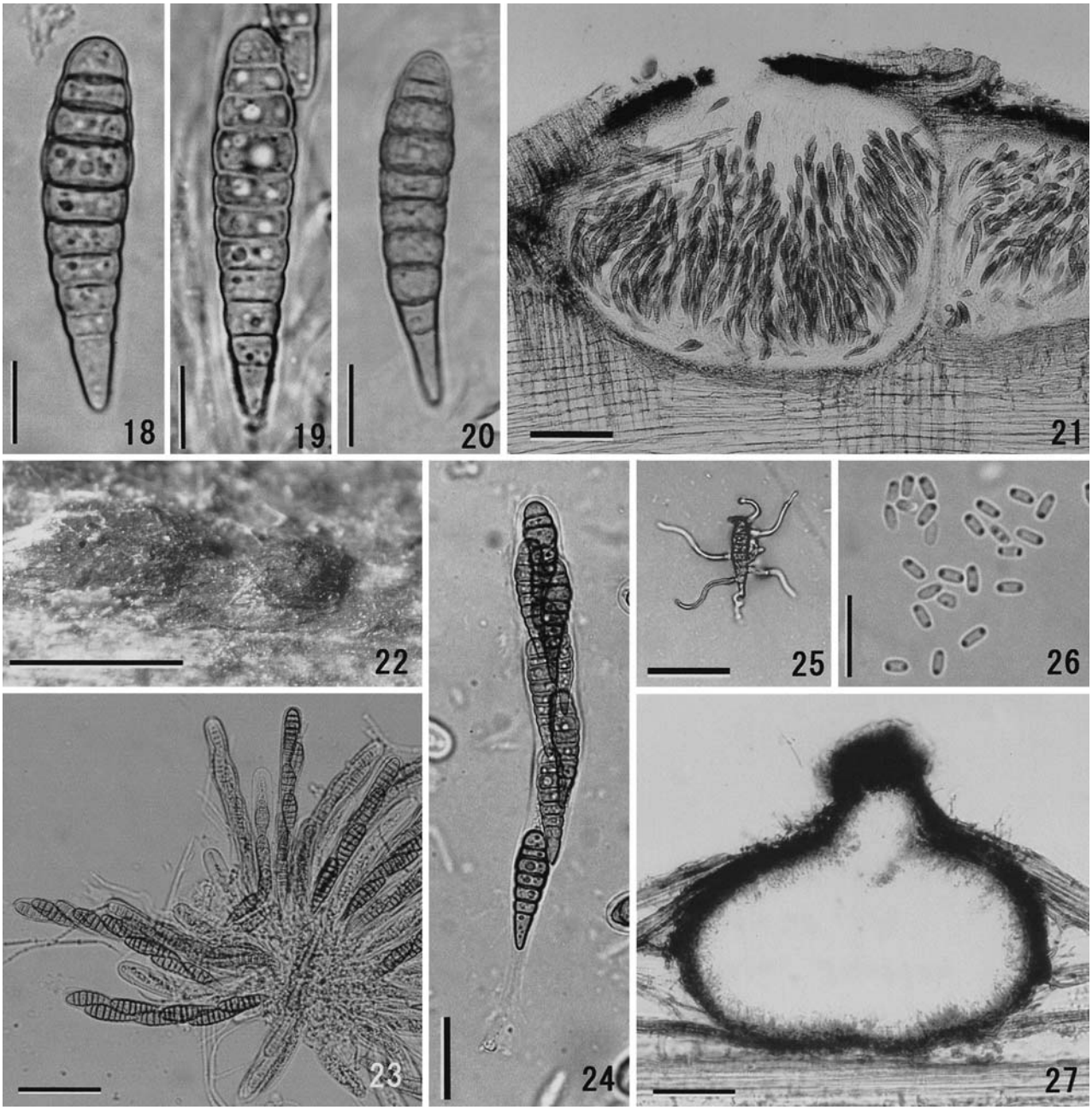
(3) ***Massariosphaeria moricola*** Kaz. Tanaka & Y. Harada, sp. nov. Figs. 18–27, 48, 53

Ascomata 320–450 μm alta, 400–600 μm diametro, dispersa vel congregata, immersa vel erumpentia, subglobosa, lanata. Rostrum nullum vel papillatum, centrale, ex cellulis polygonis vel subglobosis 2.5–7.5 μm compositum, sine periphysibus. Paries ascomatis "textura angularis" vel "textura prismatica," 8–15 μm crassus, ad latus ex cellulis hyalinis vel pallide brunneis 4.5–12.5 × 2.5–6.5 μm compositus, ad basim ex cellulis polygonis vel subglobosis 2.5–8 × 2.5–5 μm compositus. Pseudoparaphyses anguste cellulosae, copiosae, 1.5–3 μm latae, ramificantes et anastomosantes. Asci (100–) 115–170 (–174) × 13–18 (–19.5) μm, bitunicati, copiosi, basales et leviter laterales, cylindrici vel clavati, apice rotundati, stipitati, (5–) 10–40 (–50) μm longi, (4–) 8-spori. Ascosporae 31–46 (–51.5) × 8–11.5 μm, uni- vel biseriatae, fusiformes vel clavatae, rectae vel leviter curvatae, crassitunicatae, (7–) 8–10-septatae, cum septo primum supramedio formantes, ocharaceae, guttulatae, leviter verruculosae, sine tunica gelatinosa.

Holotypus. HHUF 27791.

Etymology. Latin "moricola," referring to the host genus *Morus*, and the Latin suffix *cola* meaning "dweller."

Ascomata 320–450 μm high, 400–600 μm diameter, scattered to clustered, immersed to erumpent, subglobose, unilocular, with brown sparse hyphae around the ostiole and sides. Beak none or forming low papillae, central, heavily pigmented black in upper part, composed of polygonal to subglobose slightly thick-walled cells of 2.5–7.5 μm diameter, without periphyses. Ascomal wall textura angularis, or textura prismatica, 8–15 μm thick, at sides composed of 3–5 layers of hyaline to pale brown polygonal thin-walled cells of 4.5–12.5 × 2.5–6.5 μm, at the base composed of polygonal to subglobose cells of 2.5–8 × 2.5–5 μm.



Figs. 18–27. *Massariosphaeria moricola*. **18–20** Ascospores (**19** in ascus); **21** Ascoma in longitudinal section; **22** Ascomata on host surface; **23** Asci and pseudoparaphyses; **24** Ascus; **25** Germinating ascospore; **26** Conidia; **27** Conidioma in longitudinal section. (**18**, **19**, **25** from HHUF 27790; **20**, **24** from HHUF 27789; **21**, **22** from HHUF 27791; **23** from HHUF 27789; **26**, **27** from MAFF 239222.) *Bars* **18–20**, **26** 10 μm; **21** 100 μm; **22** 1 mm; **23**, **25**, **27** 50 μm; **24** 25 μm

Pseudoparaphyses narrowly cellular, numerous, 1.5–3 μm thick, septate, branched, and anastomosed. Asci (100–) 115–170 (–174) × 13–18 (–19.5) μm (mean = 148.5 × 15.6 μm, n = 58), bitunicate, numerous, basal and somewhat lateral, cylindrical to clavate, rounded at the apex, with an apical chamber, with a stipe (5–) 10–40 (–50) μm long, (4–) 8-spored. Ascospores 31–46 (–51.5) × 8–11.5 μm (mean = 38.3 × 9.4 μm, n = 132), L/W 3.6–4.7 (mean = 4.1, n = 118), uniseriate to biseriate, fusiform to clavate with rounded upper part and tapered lower part, straight or slightly curved, thick-walled, (7–) 8–10-septate (2 + 1 + 4, 2 + 1 + 5, 3 + 1 + 4, 3 + 1 + 5, 3 + 1 + 6; mostly 3 + 1 + 5), with the primary septum supramedian 0.34–0.41 (mean = 0.37, n = 118), yellowish-brown, with or without guttules, slightly verruculose, without sheath. Ascospores germinating from each cell.

Cultural characteristics. Colonies on PDA 20 mm in diameter after 4 weeks at 20°C in the dark, mainly Reddish-White (12A2), in some parts Greenish-Grey (29D2); reverse Greyish-Ruby (12C7); Greyish-Rose (11B3) pig-

ment produced in the agar. On RSA, a *Aposphaeria*-like anamorph formed within 5 weeks on the surface of rice straws. Conidiomata 80–180 (–275) μm high, 145–275 (–350) μm diameter, associated with a purple coloration, scattered to clustered, erumpent, subglobose, with brown sparse hyphae around the beak. Beak 50–75 μm long, 50–70 μm diameter, central, sometimes eccentric, papillate. Conidiomatal wall 7–12.5 μm wide at sides and base, composed of brown pseudoparenchymatous cells. Conidiophores (7–) 15–20 (–25) × 2–3.5 μm, cylindrical to lageniform, branched, formed from all around the locular cavity. Conidiogenous cells phialidic. Conidia 3–4 × 1–2 μm, L/W mostly 2.4, ellipsoid, aseptate, hyaline, smooth.

Materials examined. On dead twigs of *Morus australis* Poir.: Campus of Hirosaki University, Hirosaki, Aomori, 140°28′ E, 40°35′ N, Apr. 13, 2001, KT. 481 (HHUF 27788); Apr. 20, 2001, KT. 506 (HHUF 27789); Mar. 14, 2002, KT. 848 (HHUF 27790, culture MAFF 239222); Nishigaoka, Hirosaki, Aomori, Apr. 6, 2002, YH. 851 (HHUF 27791 holotype). Dried culture specimen: grown on culms of *Oryza sativa* L.: from MAFF 239222 (HHUF 27792, anamorph state).

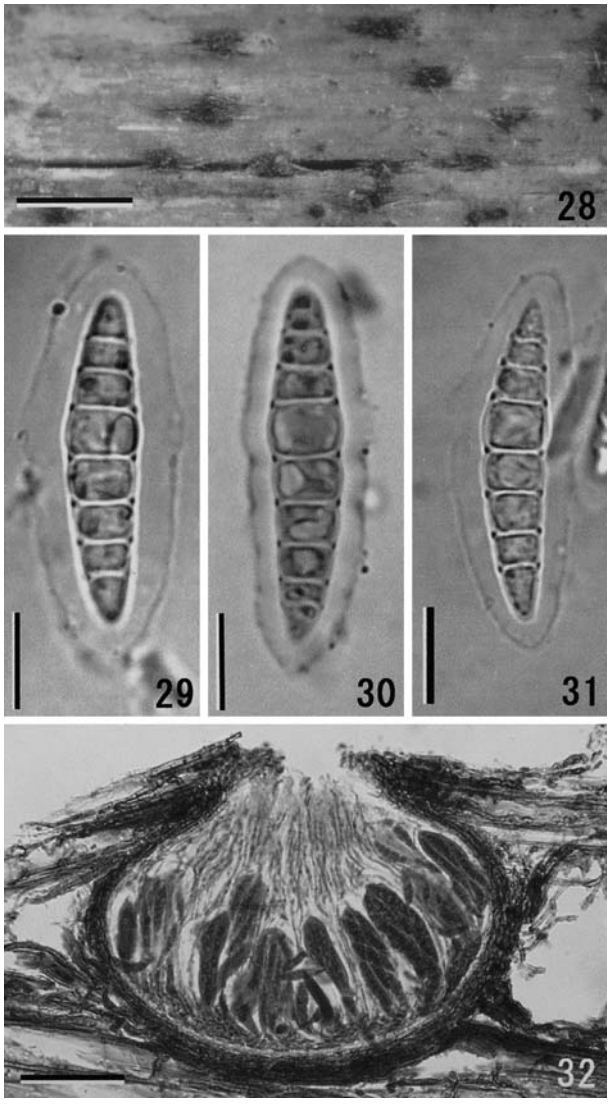
Notes. This species is highly similar to *M. clavispora*, which also has clavate, 8- to 11-septate, and similar sized ascospores. In *M. moricola* however, the fruit bodies are smaller (up to 600 μm), not elongate, without slitlike opening, and composed of thin peridium at lower sides (8–15 μm vs. 32–88 μm thick). The asci of *M. moricola* are longer and slender (148.5 × 15.6 μm on average) and do not fall within the range of those found in *M. clavispora* [(88–) 95–130 (–136.5) × (17–) 18.5–27 μm]. There are also small differences in the ascospore features. In *M. moricola*, the position of the primary septum is 0.34–0.41, and without a sheath even in the premature conditions, whereas in the latter species, the ascospores have a more supramedian primary septum (0.29–0.36) and with an obovoid conspicuous sheath. Also, colony pigmentation and conidiophore morphology in cultural conditions are quite different in both species.

(4) ***Massariosphaeria roumeguerei*** (Sacc.) Leuchtm., Sydowia 37: 173, 1985 ("1984"). Figs. 28–32, 49

≡*Leptosphaeria roumeguerei* Sacc., Michelia 2: 62, 1880 (*ut* "*roumegueri*"). ≡*Metasphaeria roumeguerei* (Sacc.) Sacc., Syll. Fung. 2: 181, 1883 (*ut* "*roumegueri*").

Ascomata 150–200 μm high, 170–235 μm diameter. Beak 20–55 μm high, 45–60 μm diameter. Ascomal wall 8.5–12.5 μm thick at side and base. Pseudoparaphyses cellular, 2–3 μm thick. Asci (72–) 76.5–97 (–103.5) × 17–20 μm (mean = 84.5 × 17.9 μm, *n* = 50). Ascospores 30–36 (–38) × 6.5–8 μm (mean = 33.5 × 7.3 μm, *n* = 50), L/W 4.2–5.2 (mean = 4.6, *n* = 44), with a supramedian 0.45–0.49 (mean = 0.47, *n* = 43) primary septum, 7- to 8-septate (3 + 1 + 3 or 3 + 1 + 4), hyaline, smooth, with a sheath 2–7 μm wide. Senescent ascospores pale brown, echinulate.

Cultural characteristics. Not examined. According to Leuchtmann (1984), this species produces *Aposphaeria*-like anamorph with 3.5–4.5 × 1–1.5 μm conidia in culture.



Figs. 28–32. *Massariosphaeria roumeguerei*. **28** Ascomata on host surface; **29–31** Ascospores (**30** in blue ink); **32** Ascoma in longitudinal section. (All from HHUF 27795.) *Bars* **28** 500 μm; **29–31** 10 μm; **32** 50 μm

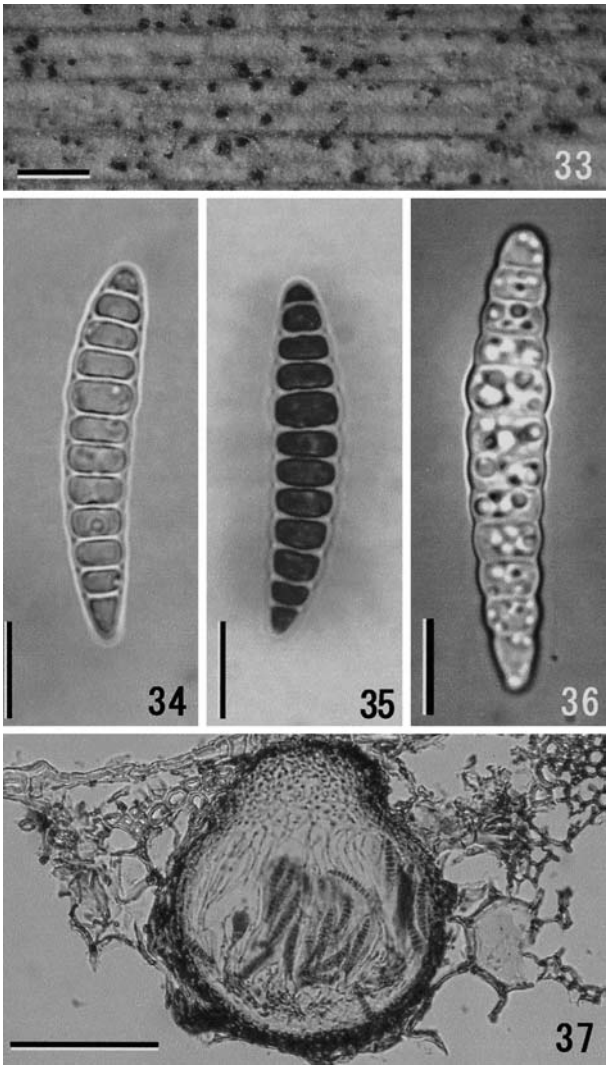
Material examined. On dead culms of *Phragmites australis*: Sanpinai, Hirosaki, Aomori, 140°30.13′ E, 40°34.51′ N, May 27, 2001, KT. 849 (HHUF 27795).

Notes. This material matches well with the description of *M. roumeguerei* provided by Eriksson (1967, as *Metasphaeria roumegueri*). The species has been reported from Gramineae (*Cynosurus* and *Elymus*) and Agavaceae (*Phormium*) in Europe (Crane and Shearer 1991; Leuchtmann 1984).

(5) ***Massariosphaeria grandispora*** (Sacc.) Leuchtm., Sydowia 37: 172, 1985 ("1984"). Figs. 33–37, 50

≡*Leptosphaeria grandispora* Sacc., Michelia 1: 341, 1878. ≡*Metasphaeria grandispora* (Sacc.) Sacc., Syll. Fung. 2: 181, 1883. ≡*Lophiotrema grandisporum* (Sacc.) Shoemaker & C.E. Babc., Can. J. Bot. 67: 1580, 1989 (*ut* "*grandispora*").

=*Lophiotrema microthecum* Vestergr., Bot. Not. p. 158, 1899.



Figs. 33–37. *Massariosphaeria grandispora*. **33** Ascomata on host surface; **34–36** Ascospores (**35** in blue ink); **37** Ascoma in longitudinal section. (**33**, **34**, **37** from HHUF 27793; **35**, **36** from MAFF 239223.) *Bars* **33** 500 μm; **34–36** 10 μm; **37** 100 μm

Ascomata 140–200 μm high, 135–215 μm diameter. Beak 25–45 μm high, 32–75 μm diameter. Ascomal wall 7.5–10 μm thick at side and base. Pseudoparaphyses narrowly cellular, 1–1.5 μm thick. Asci (98–) 100–130 (–141.5) × 19–24.5 (–27) μm (mean = 114.7 × 22.0 μm, n = 20). Ascospores 35–41 (–44) × 6.5–8 μm (mean = 37.8 × 7.3 μm, n = 55), L/W 4.7–5.6 (mean = 5.2, n = 55), 9- to 12-septate (3 + 1 + 5, 3 + 1 + 6, 4 + 1 + 5, 4 + 1 + 6, 4 + 1 + 7, 5 + 1 + 5, 5 + 1 + 6; mostly 4 + 1 + 6), with a supramedian 0.39–0.46 (mean = 0.41, n = 55) primary septum, hyaline, smooth, with a sheath up to 20 μm wide. Ascospores become brown at germination.

Cultural characteristics. Colonies on PDA 10 mm in diameter after incubation for 4 weeks at 20°C in the dark, Greenish-Grey (29D2); reverse Purplish-Grey (13F2); Greyish-Rose (11B3) pigment produced in the agar. On RSA, numerous ascomata found on the surface of rice straws and agar medium within 2 months. Ascospores slightly larger than those found in nature, measuring 38–48.5 × 8–9 μm (mean = 42.9 × 8.5 μm, n = 32).

Materials examined. On dead culms of *Phragmites australis*: Shibayachi Moor, Akita, Oodate, 140°34.5′ E, 40°19.1′ N, Oct. 5, 2001, Y. Ooki and KT. 798 (HHUF 27793, culture MAFF 239223); 799 (HHUF 27794).

Notes. Shoemaker and Babcock (1989) assigned this fungus to *Lophiotrema* Sacc. based on the slitlike ostiole of ascomata. We consider, however, it belongs to *Massariosphaeria* rather than *Lophiotrema*, because the fungus has ascomata composed of thin-walled pale brown cells around the ostiole, and has asymmetrically multitransseptate ascospores unlike *Lophiotrema nucula* (Fr.: Fr.) Sacc., the type species of *Lophiotrema*.

It has been recorded on many monocots (*Agrostis*, *Ammophila*, *Arrhenatherum*, *Carex*, *Festuca*, *Molinia*, *Phragmites*, and *Typha*) and dicots (*Aconitum*) (Holm and Holm 1988; Shoemaker and Babcock 1989), and so far is known chiefly from Europe (Barr 1992).

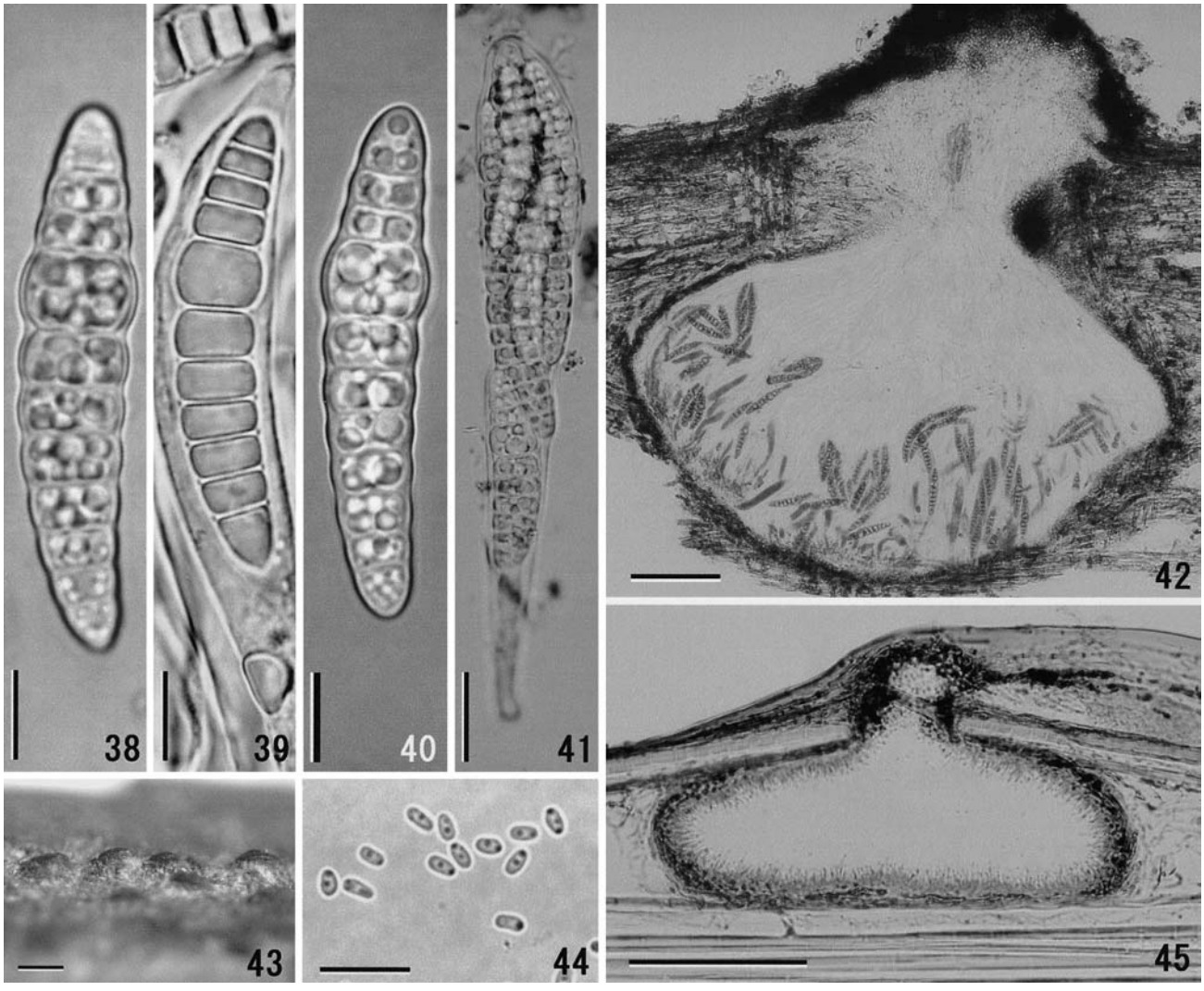
(6) ***Massariosphaeria megaspora*** Kaz. Tanaka & Y. Harada, sp. nov. Figs. 38–45, 51, 54

Ascomata 600–780 μm alta, 600–900 μm diametro, dispersa, fere immersa, ad apicem erumpentia, globosa, lanata. Rostrum 200–280 μm longum, 380–500 μm latum, centrale, cristatum, ex cellulis globosis vel polygonis 2–5 μm compositum. Paries ascomatis "textura angularis," 10–15 μm crassus, ad latus ex cellulis brunneis polygonis vel subglobosis 2.5–16 × 2.5–6 μm compositus, ad basim ex cellulis globosis vel polygonis 2.5–10 μm compositus. Pseudoparaphyses copiosae, circa 1.5–2 μm, latae, septatae, ramificantes, et anastomosantes. Asci 154–218 × 21–30 μm, bitunicati, copiosi, basales et laterales, cylindrici vel clavati, apice rotundati, stipitati, 15–43 μm longi, octospori. Ascosporae (46–) 48–61.5 (–65) × 9.5–14 μm, superiore biseriatae et inferiore uniseriales, fusiformes, leviter curvatae, asymmetricae, 9- vel 11-septatae, cum septo primum supramedio, hyalinae, guttulatae, laeves, strato mucoso 1 μm lato circumdatae.

Holotypus. HHUF 27796.

Etymology. "megaspora," referring to large ascospores of this species in the genus.

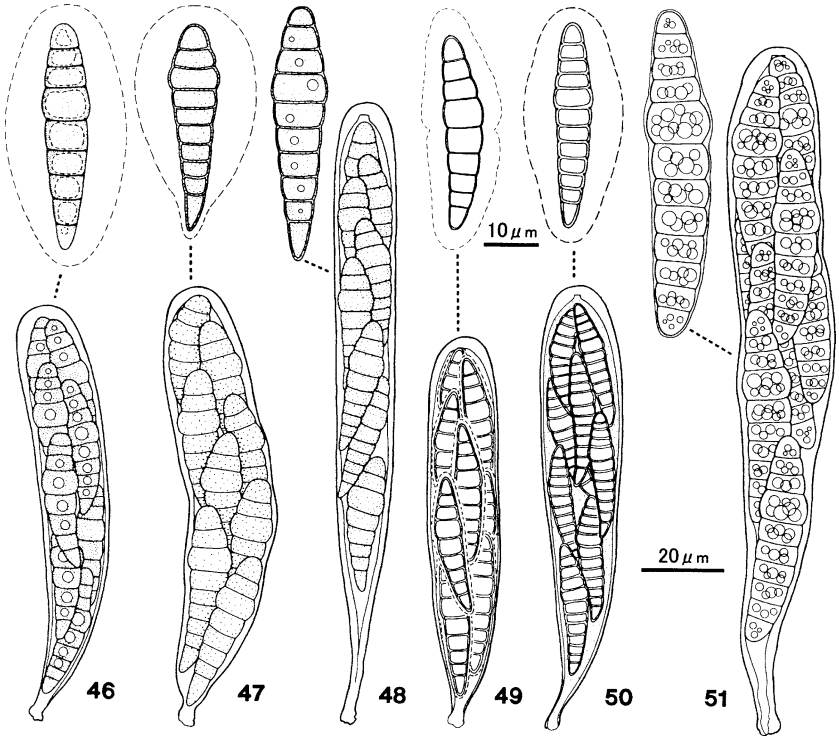
Ascomata 600–780 μm high, 600–900 μm diameter, scattered, mainly immersed, erumpent at the beak, globose, covered with brown sparse hyphae at side, with a slitlike ostiole. Beak 200–280 μm long, 380–500 μm wide (in frontal section), central, cristate, composed of globose to polygonal thick-walled cells of 2–5 μm diameter, heavily pigmented black in upper part, filled with the tips of branched pseudoparaphyses; periphyses not seen. Ascomal wall textura angularis, 10–15 μm thick, at side composed of 3–5 layers of polygonal to subglobose brown thin-walled cells of 2.5–16 × 2.5–6 μm, at the base composed of globose to polygonal brown cells of 2.5–10 μm diameter. Pseudoparaphyses numerous, 1.5–2 μm thick, with thin septa at 10- to 20-μm intervals, branched and anastomosed. Asci 154–218 × 21–30 μm (mean = 183.8 × 25.1 μm, n = 34), bitunicate, numerous, basal and lateral, cylindric-clavate,



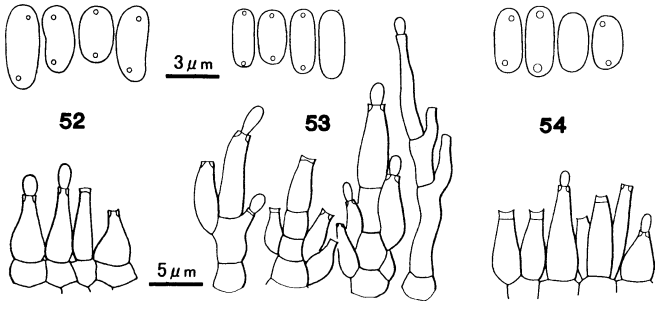
Figs. 38–45. *Massariosphaeria megaspora*. **38–40** Ascospores (**39** in ascus); **41** Ascus; **42** Ascoma in longitudinal section; **43** Ascomata on host surface; **44** Conidia; **45** Conidioma in longitudinal section. (**38**, **39**, **41–43** from HHUF 27796; **40**, **44**, **45** from MAFF 239224.) *Bars* **38–40**, **44** 10 μm; **41** 25 μm; **42**, **45** 100 μm; **43** 250 μm

rounded at the apex, with an apical chamber, short stalked (15–43 μm long), 8 spored. Ascospores (46–) 48–61.5 (–65) × 9.5–14 μm (mean = 55.2 × 12.4 μm, n = 57), L/W 4.1–4.8 (mean = 4.5, n = 57), biseriate above and uniseriate below, fusiform, slightly curved, asymmetric, 9- to 11-septate (3 + 1 + 5, 3 + 1 + 6, 4 + 1 + 5, 4 + 1 + 6), with a supramedian 0.41–0.45 (mean = 0.43, n = 57) primary septum and where strongly constricted, slightly constricted at other septa, hyaline, with small guttules, smooth, surrounded by a 1-μm-wide sheath appearing thickend outer wall; sheath slightly swelling and with irregular margin after a long soak in water. At germination, ascospores become brown and produce germ tubes from each cell.

Cultural characteristics. Colonies on PDA 18 mm in diameter after 4 weeks at 20°C in the dark, Olive-Brown (4E3) in the center, Brownish-Grey (6F2) to black in other parts; reverse similar; no pigments formed. On RSA, a *Aposphaeria*-like anamorph (sensu Leuchtmann 1984) formed on the surface of rice straws or in agar near straws within 4 weeks, and white masses of conidia exuded from the ostiole. Conidiomata 100–150 μm high, (140–) 245–330 μm diameter, scattered to sometimes clustered, erumpent or superficial, depressed globose to globose. Beak 45–50 μm long, 42–58 μm diameter, central, short papillate, composed of brown subglobose to polygonal cells of 4.5–9.5 μm diameter. Conidiomatal wall at sides composed of 3–4 layers of pale brown polygonal to subglobose cells of 2.5–11 × 2.5–4 μm, at the base poorly developed. Conidiophores 5–10 (–16) × 2–3.5 μm, cylindrical to lageniform, simple, formed from the whole round of the locular cavity. Conidiogenous cells phialidic. Conidia 3–4.5 × 1.5–2 μm, L/W 2.0, ellipsoid, aseptate, hyaline, smooth. After conidial formation, the teleomorph produced on the surface of rice straws within 3 months. Ascospores similar in shape and size to those found in nature, (50.5–) 52–61 (–66) × 11–14 μm (mean = 56.0 × 12.4 μm, n = 50), L/W 4.2–4.8 (mean = 4.5, n = 44), and with a supramedian primary septum at 0.41–0.45 (mean = 0.42, n = 44).



Figs. 46–51. Ascospores (*upper*) and asci (*lower*) of *Massariosphaeria* spp. **46** *M. typhicola*; **47** *M. clavispora*; **48** *M. moricola*; **49** *M. roumeguerei*; **50** *M. grandispora*; **51** *M. megaspora*



Figs. 52–54. Conidia (*upper*) and conidiogenous cells (*lower*) of *Massariosphaeria* spp. **52** *M. clavispora*; **53** *M. moricola*; **54** *M. megaspora*

Materials examined. On submerged dead twigs of an unknown woody plant: Shibayachi Moor, Oodate, Akita, 140°34.5′ E, 40°19.1′ N, Oct. 5, 2002, KT. 905 (HHUF 27796 holotype, culture MAFF 239224). Dried culture specimens: grown on culms of *Oryza sativa* L.: from MAFF 239224, anamorph state (HHUF 27797); teleomorph state (HHUF 27804, 27805, 28038).

Notes. The large ascospores of *M. megaspora* (55.2 × 12.4 μm on average) superficially resemble those of *M. erucacea* Kohlm., Volkm.-Kohlm. & O.E. Erikss. (1996; 60.3 × 10.4 μm on average) on *Juncus roemerianus* reported from North America. *Massariosphaeria megaspora*, however, is distinguished from the latter by number of septa in its ascospores [9–11 vs. (10–) 11–14], by longer asci (154–218 μm vs. 110–150 μm), and by larger ascomata having a slitlike ostiole [600–900 μm vs. 320–420 (–520) μm in diameter].

In its 9- to 11-septate ascospores, *M. megaspora* is also similar to *M. alpigena* (Fuckel) L. Holm & K. Holm, but it can be separated from the latter by its ascospore length [(46–) 48–61.5 (–65) μm vs. 40–45 μm in *M. alpigena*; Holm and Holm 1988].

Key to the species of *Massariosphaeria* treated

1. Ascospores yellow to brown 2
1. Ascospores hyaline 4

2. Ascospores fusiform, with a primary septum at 0.38–0.49 (mean = 0.44) *M. typhicola*
2. Ascospores fusiform to clavate, with more supramedian primary septum 3

3. Ascospore primary septum supramedian about 0.33; asci mostly more than 18 μm wide; on *Phragmites* *M. clavispora*
3. Ascospore primary septum supramedian about 0.37; asci mostly less than 18 μm wide; on *Morus* *M. moricola*

4. Ascospores 7- to 8-septate *M. roumeguerei*
4. Ascospores more than 8-septate 5

5. Ascospores 35–41 (–44) × 6.5–8 μm *M. grandispora*
5. Ascospores (46–) 48–61.5 (–65) × 9.5–14 μm *M. megaspora*

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