Neth. J. Pl. Path. 86 (1980) 45–53

Conidiogenesis of Fusarium nivale and Rhynchosporium oryzae and its taxonomic implications

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Accepted 18 September 1979

Abstract

Scanning electron microscopy has shown that *Fusarium nivale* Ces. ex Sacc., the anamorph of *Monographella nivalis* (Schaffnit) E. Müll., has annellate conidiogenous cells and this character together with its amphisphaeriaceous teleomorph distinguishes it from other fusaria. *F. nivale* and its variety *majus* are assigned to a new genus, *Gerlachia*, as *G. nivalis* var. *nivalis* and *G. nivalis* var. *major*. For the teleomorph of the latter the new combination *Monographella nivalis* var. *neglecta* (Krampe) W. Gams & E. Müll. is proposed. The causal agent of rice leaf scald, *Rhynchosporium oryzae* Hashioka & Yokogi, has the same conidiogenesis and is transferred to *Gerlachia* as *G. oryzae* (Hashioka & Yokogi) W. Gams and distinguished from *G. nivalis* by conidial shape and higher cardinal temperatures.

Additional keywords: Gerlachia gen. nov., *Gerlachia nivalis*, *Gerlachia oryzae*, *Monographella nivalis* var. *neglecta* comb. nov., morphology, taxonomy, scanning electron microscopy.

Introduction

Fusarium nivale Ces. ex Sacc. is an outsider in the genus *Fusarium* since the conidia lack any sign of foot-cell differentiation; it is entirely light-dependent for sporulation; it also shows some deviating reactions to fungicides, being unusually sensitive to benomyl (ED_{50} = 0.1 µg/ml, Van Tuyl, 1978). This fungus is responsible for damages on grasses and cereals, especially winter cereals (snow mould) (e.g. Årsvoll, 1975; Duben, 1978). Additionally, recent investigations (Petrini et al., 1979) suggest that it normally lives as a harmless leaf endophyte and only turns to be a pathogen under a snow cover lasting for several weeks.

The teleomorph[3], *Monographella nivalis* (Schaffnit) E. Müll., stands outside the Hypocreaceae because of the amyloid ascus tips, the presence of paraphyses and brownish ascomata immersed in the host tissue (Müller, 1977) and a development of the ascomata deviating from the *Nectria* type (Subramanian and Bhat, 1978). Müller (1977) accomodated it, like the genus *Griphosphaeria* in which this fungus was previously classified, in the Amphisphaeriaceae; he also suggested (in Kendrick, 1979, p. 181–182) that the conidia might be formed on annellidic conidiogenous cells like in

[3] The terms teleomorph and anamorph are used here as suggested by Hennebert and Weresub (1977) to indicate perfect and imperfect states, respectively.

other genera with amphisphaeriaceous teleomorphs. Therefore in this study the conidiogenous cells and conidia of *F. nivale* and the morphologically similar *Rhynchosporium oryzae* Hashioka & Yokogi were examined in detail for annellidic and other characteristics so that their affinities could be more accurately assigned.

Material and methods

The sporulating isolates of *F. nivale* var. *nivale* and var. *majus* preserved at CBS were compared by light microscopy. Three isolates of var. *nivale*, CBS 162.77, 319.78 and 320.78, and one of var. *majus*, CBS 111.78, were examined by scanning electron microscopy (SEM). Three isolates of *Rhynchosporium oryzae*, CBS 289.79, 290.79 and 291.79, were obtained from Dr V. O. Parkinson, Orsay, France, who had isolated them from rice scald in Sierra Leone and Ivory Coast, respectively; CBS 291.79 was studied by SEM.

Colonies of both varieties of *F. nivale* were grown on oatmeal agar at 16–18 °C under near-UV light. Those of *R. oryzae* at 25 °C in darkness (Parkinson, 1979). As the conidiogenous cells tend to collapse very soon after producing their conidia, repeated trials were necessary in order to obtain suitable material. The specimens were fixed and prepared for SEM according to Samson et al. (1979). Light-microscopic studies were

Fig. 1. *Gerlachia nivalis* var. *nivalis*, annellate conidiogenous cells with attached conidia. CBS 162.77, SEM; bar represents 2 µm.

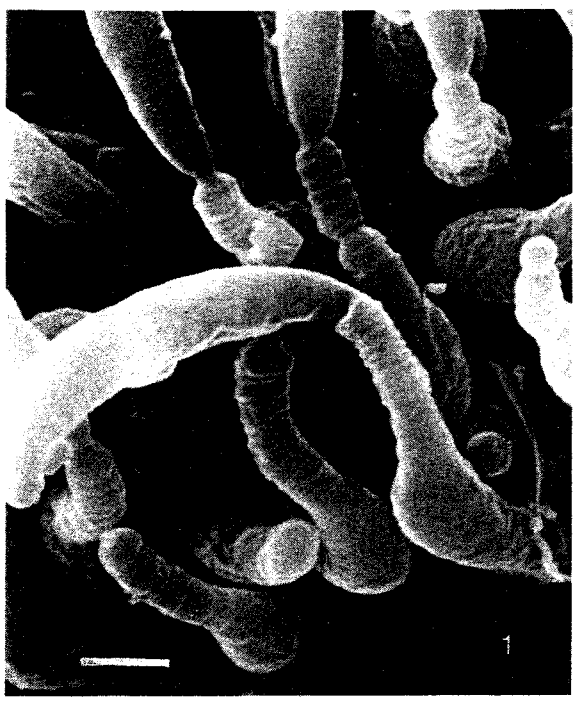

Fig. 1. Gerlachia nivalis var. nivalis, geringde conidiogene cellen met nog vastzittende conidiën, CBS 162.77, scanning elektronenmicroscopie; vergrotingsstaaf geeft 2 µm weer.

Fig. 2. *Gerlachia nivalis* var. *nivalis*, two annellate conidiogenous cells. CBS 319.78, SEM; bar represents 2 µm.

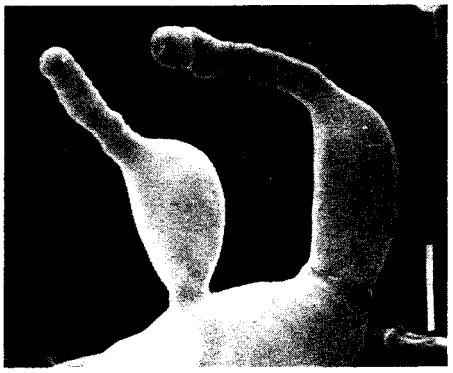

Fig. 2. Gerlachia nivalis var. nivalis, twee geringde conidiogene cellen, CBS 319.78; vergrotingsstaaf geeft 2µm weer.

done with water and lactic acid with aniline blue as mounting media, the latter giving narrower spore measurements.

Results and discussion

Fusarium nivale var. nivale

Representative SEM pictures are shown in Fig. 1 and 2. They leave no doubt about the presence of conspicuous annellations. However, the differentiation and therefore the sharpness of the annellations may vary between isolates, CBS 162.77 being more distinctly annellate than 319.78. Under the light microscope the annellations as such could not be seen, but an elongation of a narrowed tip of the conidiogenous cell is clearly visible (see also the drawings by Subramanian and Bhat, 1978) whilst an interior apical wall thickening characteristic of other *Fusarium* species is absent.

The presence of percurrently elongating conidiogenous cells in *F. nivale* distinguishes it from the rest of the genus. Comparable structures are found in several genera of the Melanconiales like *Seimatosporium*, *Sporocadus* and *Pestalotia* with segregates which, however, have mostly pigmented conidia.

In recent years it has become a generally recognized principle that different kinds of conidiogenesis are not to be admitted in a single genus of Hyphomycetes, since different conidiogenesis is often correlated with different teleomorph affinities (Kendrick, 1979). For example, the distinction between *Penicillium* and *Scopulariopsis* is based on the occurrence of phialides and annellides which is also reflected by differences in the correlated teleomorphs in the Trichocomaceae and Microascaceae. It seems that the distinction of slimy conidia formed on phialides in the Hypocreaceae and on annellides in the Amphisphaeriaceae is a comparable case where there is a good correlation of teleomorphic and anamorphic distinctions.

The original concept of annellophores by Hughes (1953) has been shown to comprise different modes of conidiogenesis (Hammill, 1973, 1974), viz. (a) phialide-like con-

Fig. 3a. *Gerlachia nivalis* var. *nivalis*, light-microscopic view. CBS 112.78, top row, CBS 113.78, bottom row of conidia. 3b. *Gerlachia nivalis* var. *major*. CBS 177.29, top row, CBS 111.78, bottom row of conidia with conidiogenous cells.

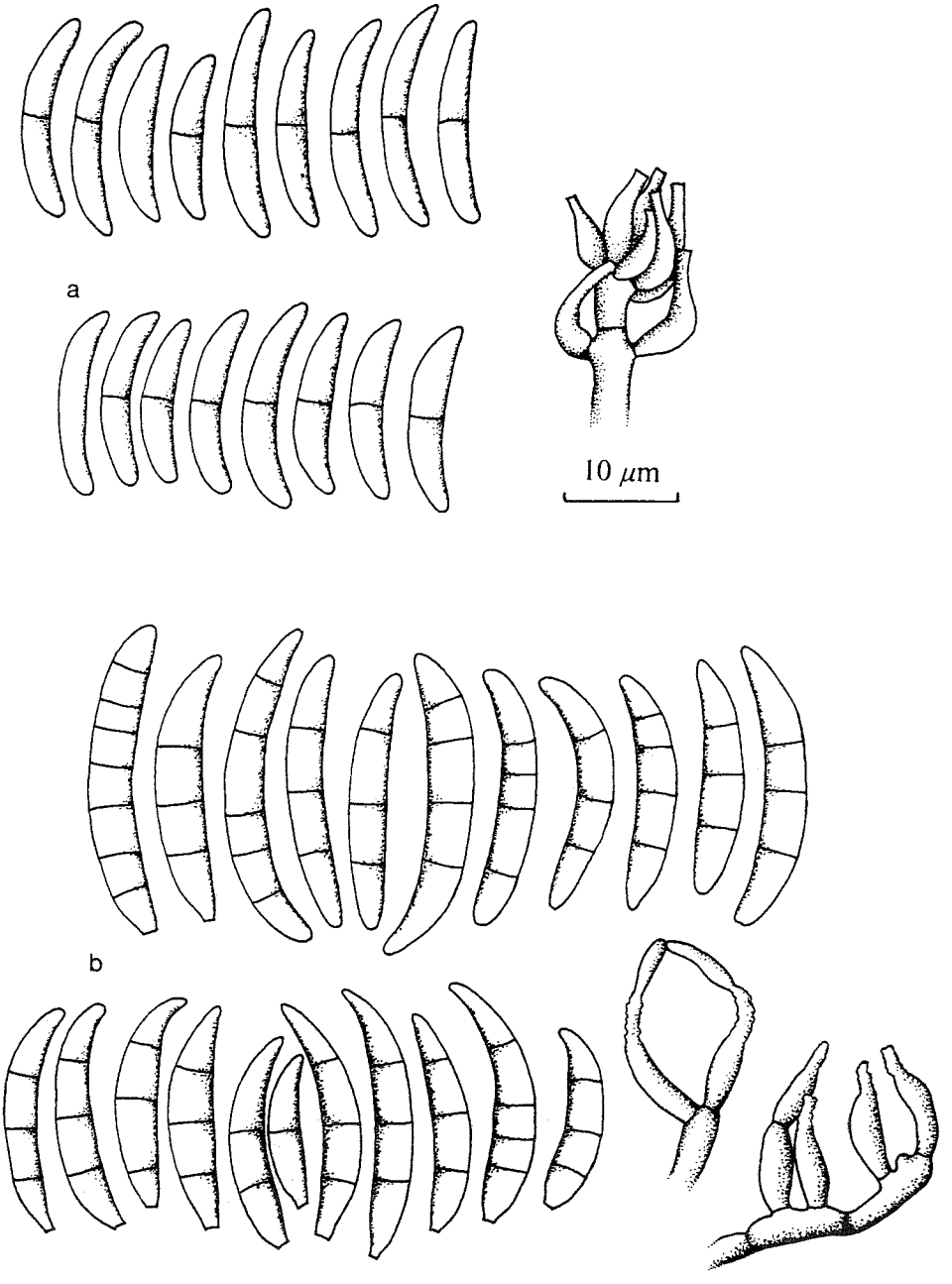

Fig. 3a. Gerlachia nivalis var. nivalis, lichtoptisch beeld, CBS 112.78 boven, CBS 113.78 onder. 3b. Gerlachia nivalis var. major, CBS 177.29 boven, CBS 111.78 onder.

idiogenous cells in which the wall material apposed internally near the opening gradually elongates outwards, and (b) blastic conidiogenous cells proliferating percurrently by the apposition of new wall layers clothing the entire apical cell with each newly formed conidium. On the other hand, ultrastructural studies have shown that a continuum exists between certain groups of phialide- and annellide-forming species determined solely by the fact whether the conidiogenous locus remains constant or moves gradually upwards (Hammill, 1974; Cole and Samson, 1979). Nevertheless, in other cases this criterion allows a reliable distinction of genera.

In view of the above-mentioned correlation of annellides in the anamorph with the amyloid ascus tip in the amphisphaeriaceous teleomorph, we regard it as justified to segregate *F. nivale* from *Fusarium* and accomodate it in a genus of its own.

Gerlachia W. Gams & E. Müll. gen. nov.

Genus Hyphomycetum *Fusarii* simile. Coloniae hyalinae, sporodochiis roseae ad aurantiae. Cellulae conidiogenae in sporodochiis acervatae, lageniformes, sursum percurrenter elongascentes, annellatae. Conidia fusiformia, curvata, plerumque 1–3(–7) septata, hyalina, levia, mucida. Anamorphosis *Monographellae* Petrak.

Species typica: *Gerlachia nivalis* (Ces. ex Sacc.) W. Gams & E. Müll.

Etymology: The genus is named after W. Gerlach, Biologische Bundesanstalt, Berlin, in recognition of his contributions to *Fusarium* taxonomy.

Gerlachia is a genus of the Hyphomycetes similar to *Fusarium* but with percurrently elongating annellate conidiogenous cells (annellides) with sometimes pronounced tendency to sporodochium formation. The conidia are slimy, falcate, without any basal differentiation ('foot cells'), one- to many-septate.

Gerlachia nivalis (Ces. ex Sacc.) W. Gams & E. Müll. comb. nov. var. **nivalis** – Fig. 1, 2, 3a.

basionym: *Fusarium nivale* Ces. ex Sacc., Syll. Fung., Suppl. 1–4: 390. 1886.
(syn.: *F. nivale* (Fr.) Sorauer; for author's citation see Boerema and Verhoeven, 1977).

teolemorph: Monographella nivalis (Schaffnit) E. Müll. 1977 var. *nivalis*.

Further representative isolates: CBS 146.68, 112.78, 113.78, all from wheat.

Other taxa in Gerlachia

Gerlachia nivalis var. **major** (Wollenw.) W. Gams & E. Müll. comb. nov. – Fig. 3b.

basionym: *Fusarium nivale* var. *majus* Wollenw., Fusaria autogr. delin. No. 882. 1930.

teleomorph: **Monographella nivalis** var. **neglecta** (Krampe) W. Gams & E. Müll. comb. nov.

basionym: *Calonectria graminicola* var. *neglecta* Krampe, Angew. Bot. 8: 252. 1926.

The most conspicuous difference from *G. nivalis* var. *nivalis* are the multiseptate conidia (Fig. 3). Ecology and temperature relationships do not show conspicuous differences (Duben, 1978, p. 12).

Representative isolates: CBS 177.29, 256.61, 111.78, all from wheat.

Fig. 4. *Gerlachia oryzae*, annellate conidiogenous cells and conidia. CBS 219.79, SEM; bar represents 2 μm.

Fig. 4. Gerlachia oryzae, geringde conidiogene cellen en conidiën, CBS 219.79, SEM; vergrotingsstaaf geeft 2 μm weer.

Gerlachia oryzae (Hashioka & Yokogi) W. Gams comb. nov. – Fig. 4, 5.

basionym: *Rhynchosporium oryzae* Hashioka & Yokogi in Hashioka & Ikegami, Contr. Lab. Pl. Dis. Sci. Gifu Univ. 6: 51. 1955.

teleomorph: *Monographella* spec. (V. O. Parkinson pers. commun., see also Naito and Koshimizu, 1974).

The rather meagre original description of *R. oryzae* in Hashioka and Ikegami (1955) was repeated by Ou (1972) and thus became generally accessible. There is no doubt in the correct identification of the causal agent of rice leaf scald with this species by Parkinson (1979) and we take her isolates as representative of this species. The presence of annellides may already be assumed from the illustrations by Naito and Koshimizu (1974) and has further been corroborated by our SEM study (Fig. 4).

At first sight this species is closer to *G. nivalis* var. *nivalis* than its var. *major*, and if it were not for the different ecology and cardinal temperatures (Table 1) we could regard it as a variety of the latter species. The only rather distinctive feature is the mostly recognizable swelling in the lower part of the less curved conidia which renders them somewhat *Rhynchosporium*-like. For the rest the fungus has no affinities to *R. secalis* (Oudem.) J. J. Davis, the blastic conidiogenesis of which has been described and illustrated by Owen (1973). The cultural behaviour of *G. oryzae* has been described and illustrated in detail by Parkinson (1979).

Fig. 5. *Gerlachia oryzae*, light-microscopic view. CBS 289.79, top row, CBS 290.79, bottom row.

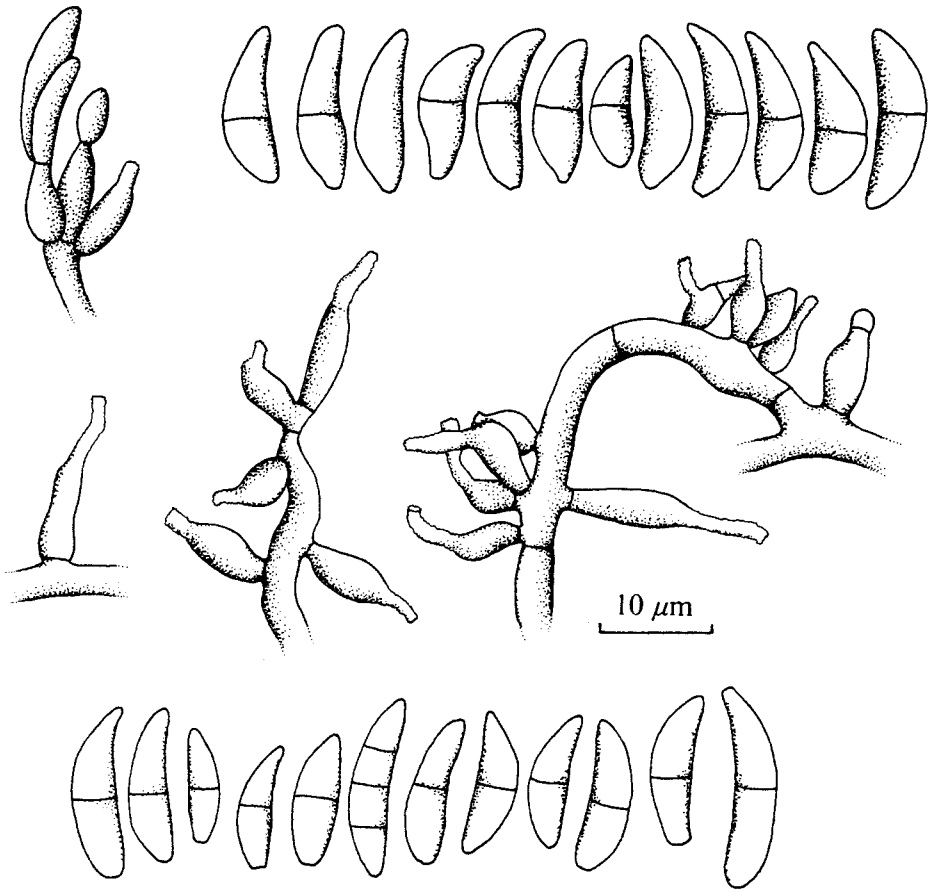

Fig. 5. Gerlachia oryzae, lichtoptisch beeld, CBS 289.79 boven, 290.79 onder.

Table 1. Some differential characteristics of the three taxa of *Gerlachia*.

	G. nivalis var. *nivalis*	*G. nivalis* var. *major*	*G. oryzae*
Number of septa	1(–3)	(1–)3(–7)	(0–)1(–3)
Conidial size (in water)	13–20 × 2.5–3.3 μm	19–30(–37) × 3.5–4.5 (–6)μm	11–16 × 3.5–4.5 μm
Temperature (°C)			
minimum	–5[1]		0[2]
optimum	20–21		24–28
maximum	28		36

[1] After Årsvoll, 1975; Jamalainen, 1974; Colhoun, 1970.
[2] After Parkinson, 1979.

Tabel 1. Onderscheidende kenmerken van de drie schimmels in Gerlachia.

Another fungus of interest in this connection is *Fusarium nivale* var. *oryzae* Zambettakis 1950 (nomen invalidum, Art. 36) which was described and illustrated as having (2–)3–5(–8) septate conidia, 12–38(–47) × 3–5(–6) µm, without any swelling of the basal cell and thus most similar to *G. nivalis* var. *major*, but the conidia are longer. No representative isolate is now available and its status is doubtful. More work needs to be done on the question whether *G. nivalis* or further taxa also occur on rice. That some further taxonomic problems in *G. nivalis* occurring either on cereals or on grasses are to be solved is suggested by Smith (1978).

Acknowledgments

It is a pleasure to acknowledge helpful suggestions received from Dr J. A. von Arx and prof. W. Gerlach, the contribution of cultures of *Rhynchosporium oryzae* by Dr V. O. Parkinson, the correction of the English text by Dr P. R. Merriman, assistance with the SEM work by Dr R. A. Samson and Miss M. Nieuwstad and inking of the drawings by Miss I. ten Hoedt.

Samenvatting

Conidiënvorming van Fusarium nivale en Rhynchosporium oryzae en taxonomische consequenties

Door middel van scanning elektronenmicroscopie werd aangetoond dat *Fusarium nivale* Ces. ex Sacc., de imperfecte vorm van *Monographella nivalis* (Schaffnit) E. Müll., geringde conidiogene cellen bezit. Door dit kenmerk en de tot de Amphisphaeriaceae horende perfecte vorm wijkt de soort van de overige *Fusarium*-soorten af. Deze soort en de variëteit *majus* worden in een nieuw geslacht, *Gerlachia*, geplaatst als *G. nivalis* var. *nivalis* and *G. nivalis* var. *major*. Voor de perfecte vorm van deze laatste wordt de nieuwe combinatie *Monographella nivalis* var. *neglecta* (Krampe) W. Gams & E. Müll. geïntroduceerd. De verwekker van een bladschroeiziekte bij rijst, *Rhynchosporium oryzae* Hashioka & Yokogi, heeft dezelfde soort van conidiënvorming en wordt naar *Gerlachia* overgebracht als *G. oryzae* (Hashioka & Yokogi) W. Gams. Deze soort is te onderscheiden van *G. nivalis* door de conidiënvorm en temperatuureis voor groei.

References

Årsvoll, K., 1975. Fungi causing winter damage on cultivated grasses in Norway. Meld. Norg. LandbrHøgsk. 54(9): 49 pp.

Boerema, G. H. & Verhoeven, A. A., 1977. Check-list for scientific names of common parasitic fungi. Series 2b: Fungi on field crops; cereals and grasses. Neth. J. Pl. Path. 83: 165–204.

Cole, G. T. & Samson, R. A., 1979. Patterns of development in conidial fungi. Pitman, London: 190 pp.

Colhoun, J., 1970. Epidemiology of seed-borne *Fusarium* diseases of cereals. Annls Acad. Sci. fenn., Ser. A, IV, 168: 31–36.

Duben, J., 1978. Untersuchungen zum Fusskrankheitskomplex an Winterweizen unter besonderer Berücksichtigung von Arten der Gattung *Fusarium* Lk. Diss. Univ. Göttingen: 149 pp.

Hammill, T. M., 1973. Fine structure of annellophores. IV. *Spilocaea pomi*. Trans. Br. mycol. Soc. 60: 65–68.

Hammill, T. M., 1974. Electron microscopy of phialides and conidiogenesis in *Trichoderma saturnisporum*. Am. J. Bot. 61: 15–24.
Hashioka, Y. & Ikegami, H., 1955. The leaf scald of rice. Jubilee publication in commemoration of the sixtieth birthdays of Prof. Yoshihito Tochinai and Prof. Teikichi Fukushi. Contr. Lab. Pl. Dis. Sci., Fac. Agric., Gifu Univ. 6: 45–51.
Hennebert, G. L. & Weresub, L. K., 1977. Terms for states and forms of fungi, their names and types. Mycotaxon 6: 207–211.
Hughes, S. J., 1953. Conidiophores, conidia, and classification. Can. J. Bot. 31: 577–659.
Jamalainen, E. A., 1974. Resistance of winter cereals and grasses to low-temperature parasitic fungi. A Rev. Phytopath. 12: 281–302.
Kendrick, B. (Ed.), 1979. The whole fungus. National Museum of Natural Sciences, Ottawa. 2 vols, 793 pp.
Müller, E., 1977. Die systematische Stellung des 'Schneeschimmels'. Revue Mycol. 41: 129–134.
Naito, H. & Koshimizu, Y., 1974. (Morphology and hyphal growth temperature of brown leaf spot fungus of rice (Rice scald fungus)). Ann. phytopath. Soc. Japan 40: 319–328.
Ou, S. H., 1972. Rice diseases. Commonwealth Mycological Institute, Kew: 368 pp.
Owen, H., 1973. *Rhynchosporium secalis*. CMI Descr. pathog. Fungi Bact. No. 387.
Parkinson, V. O., 1979. Cultural characteristics of the rice leaf scald fungus. *Rhynchosporium oryzae*. Trans. Br. mycol. Soc. (in press).
Petrini, O., Müller, E. & Luginbühl, M., 1979. Pilze als Endophyten von grünen Pflanzen. Naturwissenschaften 66: 262.
Samson, R. A., Stalpers, J. A. & Verkerke, W., 1979. A simplified technique to prepare fungal specimens for scanning electron microscopy. Cytobios 24: 7–12.
Smith, J. D., 1978. *Fusarium nivale* from cereals and grasses: Is it the same fungus? Preprint from Fusarium Newsletter.
Subramanian, C. V. & Bhat, D. J., 1978. Developmental morphology of Ascomycetes. III. *Monographella nivalis*. Revue Mycol. 42: 293–304 and 43: 218 (1979).
Tuyl, J. M. van, 1978. Genetics of fungal resistance to systemic fungicides. Meded. LandbHogesch. Wageningen 77(2): 136 pp.
Zambettakis, C., 1950. Une fusariose du panicle du riz en Oubangui. Revue Mycol. 15, Suppl. colon. 2: 106–111.

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