Trans. Br. mycol. Soc. **56** (2), 261–266 (1971)
Printed in Great Britain

PYRENOPHORA TETRARRHENAE SP.NOV., *DRECHSLERA TETRARRHENAE* SP.NOV., AND *D. BISEPTATA* ON EHRHARTEAE IN AUSTRALIA

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(With Plate 31 and 1 Text-figure)

Drechslera tetrarrhenae sp.nov. (perfect state *Pyrenophora tetrarrhenae* sp.nov.) and *D. biseptata* (Sacc. & Roum.) Richardson & Fraser are described causing leaf spots on *Tetrarrhena juncea* R.Br. and *Microlaena stipoides* (Labill.) R.Br.

The Victorian members of the Gramineae, tribe Ehrharteae, number three genera, two of which are endemic, *Microlaena* and *Tetrarrhena*, and the other, *Ehrharta*, an introduction from South Africa.

M. stipoides (Labill.) R.Br. is a common and widespread perennial grass in Victoria with a wide range of habitats, being best developed in forest, woodland and heath communities. Of the three species of *Tetrarrhena*, *T. juncea* R.Br. is most common, being a frequent understory component of the vegetation in all damp forests of the state. It is known as forest wire grass or tangle grass and forms dense masses round bases of forest trees and dense thickets, covering some of the smaller shrubby understory with a tangle of branching stems.

Leafspots on *T. juncea* and *M. stipoides* from several localities have been collected and two small-spored species of *Drechslera* Ito have been consistently isolated. These were sent to the Commonwealth Mycological Institute for examination. The larger-spored and more common of the two is a previously undescribed form-species, *Drechslera tetrarrhenae*. The second fungus was identified as *Drechslera biseptata* (Sacc. & Roum.) Richardson & Fraser, a species with a wide host range.

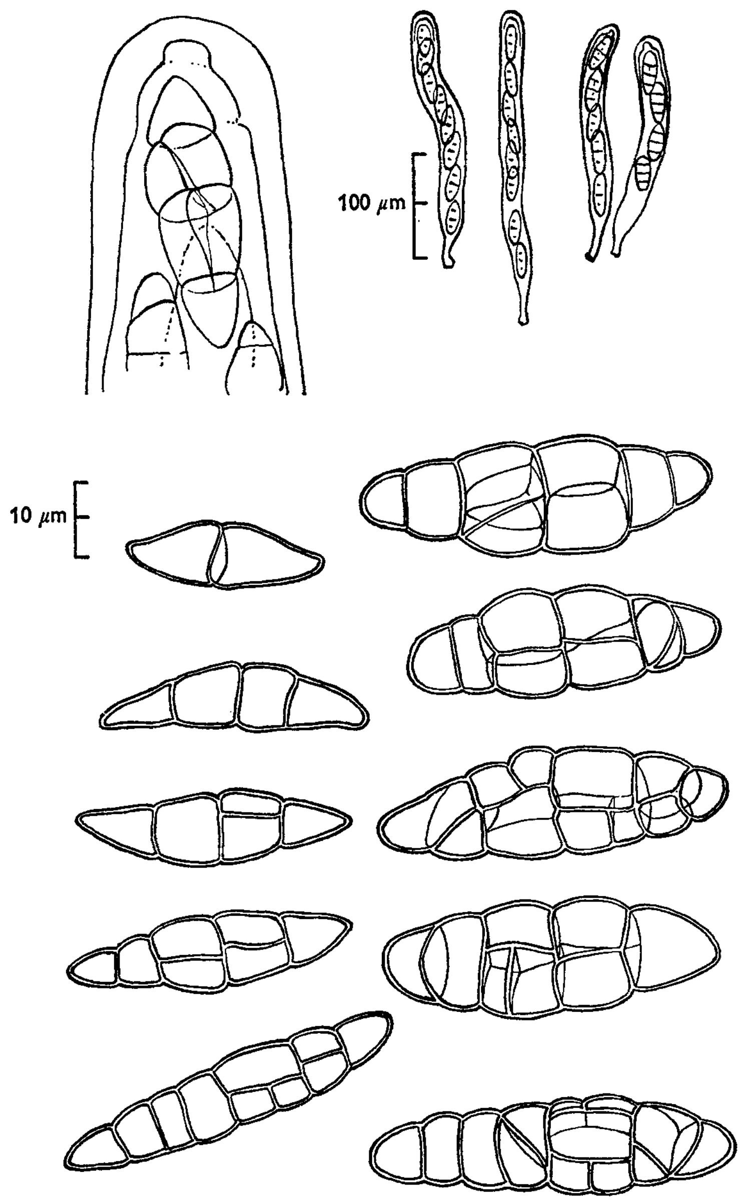
Pyrenophora tetrarrhenae sp.nov. (Text-fig. 1)

Perithecia 650–800 × 380–450 μm, superficiale, solitaria, atrobrunnea, ellipsoidea, basi breviter stipitata, setis (usque ad 440 × 3–10 μm) atrobrunneis, septatis, copiose obtectis. Asci maturi 170–280 × 22–32 μm, 4- ad 8-spori, cylindrici, bitunicati, leviter curvi, quoque ad apicem late rotundo et ad basin breviter stipitato; ascosporae 36–55 × 15–20 μm, uniseriales, pallide flavobrunneae, fusiformes usque ad ellipsoideae, muriformes, earum septis transversis ad octo.

Holotypus: **MELU**, F 7362, cultura sicca e conidiis commixtis *Dreschslerae tetrarrhenae*, Victoria, Australia, July 1969, A. R. Paul.

Ascocarps superficial, solitary, dark brown, ellipsoidal, with a shortly stipitate base, covered with numerous dark brown septate setae up to 440 μm in length and 3–10 μm in width; ascocarps 650–800 × 380–450 μm.

Asci bitunicate, cylindrical, slightly curved, rounded at the apex and stipitate at the base. Mature asci 170–280 × 22–32 μm containing 4–8 spores. Ascospores uniseriate, pale yellow-brown, fusoid-ellipsoid, muriform, with up to 8 transverse septa, 36–55 × 15–20 μm. Young spores develop a primary transverse septum followed by two secondary transverse divisions forming a fusoid 4-celled phragmospore. The spore expands



Text-fig. 1. *Pyrenophora tetrarrhenae*. Asci and ascospores showing stages in development of septa.

followed by the formation of one or two longitudinal septa in the median cells. Further development is variable. Usually at least a further two transverse septa form and these may develop in one half of the spore only (Text-fig. 1). Mature spores are constricted at the septa.

The development of sclerotia in most isolates indicated that the production of the perfect state in culture was possible. Several isolates were placed together on tap-water agar to which various host materials had been added. This tissue consisted of small pieces of leaves, stems and spikelets of *Microlaena stipoides* and *Tetrarrhena juncea*. As the cultures developed, small amounts of sterile soil water were added to keep the medium moist. Cultures were maintained at room temperature during autumn and winter (March–July) under prevailing diurnal conditions. Many sclerotia developed after about 4 weeks but soon became covered with dense setae and conidia. On plates where little host material had been included, a few dark, setose, globular bodies formed on the surface of some of the grass fragments after 3–4 months. Mature asci and ascospores formed within these bodies during the fifth month. A technique similar to this was used by Paul (1969) to produce the perfect state of *Drechslera verticillata* (O'Gara) Shoemaker in culture.

Drechslera tetrarrhenae sp.nov. (Pl. 31, fig. 3)

Status conidicus *Pyrenophora tetrarrhenae* Paul

Conidiophorae erectae, simplices, super basin bulbosam (10–12 µm diam) aliquando ramosae; color badius, ad apicem (non inflatum) pallidior; stirps 6–8·5 µm lata, septata spatiis 15–32 µm metientibus; longitudovaria, maxime 1100 µm; genicula prominentia, usque ad 200 µm. Separata, propius apicem congesta et indistincta.

Conidia 30–72 × 9–16 µm, 3- ad 6-septata (usitate 4–5), ut videtur granulata, subcylindrica, prope medium latissima, in extremitates (late rotundas) leviter contracta; color fulvidus, cellulis terminalibus semiellipsoideis subhyalinis usque luteis, cellulis centralibus lineis vel maculis nubilibus in septis et muris sporae praeditis; hilum nigrum, prominens, 3–5 µm latum, in lineamento basali muri sporae inclusum. Germinatio sporae promiscua, sed usitate a cellulis apicalibus et basalibus. Conidia secondaria rara.

Holotypus: **MELU** F 7363, in foliis *Tetrarrhenae junceae*. Belgrave, Victoria, Australia, March 1969, A. R. Paul.

Conidiophores arise individually or in groups of two or three from the substrate, usually simple and erect although sometimes branched close to the base which is usually enlarged and bulbous, being between 10 and 12 µm diam. Conidiophores are brown, of constant width of 6–8·5 µm until the first geniculation, apex of each conidiophore not inflated, pale yellow-brown in colour and generally 4·5–8·5 µm wide, multiseptate at intervals of 15–32 µm. Length is extremely variable, depending on the substrate and the length of time for development prior to measurement. On leaves where occasional conidiophores arise, conidiophores may exceed 1000 µm bearing conidia on geniculations up to 200 µm apart. More usually when a large group of conidiophores arises from and around a leafspot they rarely exceed 500 µm and bear numerous conidia crowded closely together near the apex.

In culture with added host material in the substrate, numerous protothecia develop bearing large numbers of long conidiophores. These often grow to 300 μm in length before producing the initial conidium. Numerous conidia are then formed close together and geniculations of the conidiophores are imperceptible.

Conidia are 30–72 × 9–16 μm, 3- to 6-septate but most commonly 4- or 5-septate, subcylindric, widest near the centre and tapering slightly to each broadly rounded end, of granular appearance, pale yellow-brown to orange-brown with tiny black dots at the junction of septa with the outer wall or short black lines in the endospore running parallel to the spore margin; septa straight, including dark lines in central septa; apical and basal cells subhyaline to pale yellow and semi-ellipsoid. Central cells of conidia are sometimes heavily pigmented compared with the apical cells. However they are not enlarged like those of *Curvularia* spp. Hila are black, prominent and included in the contour of the base, 3–5 μm in width.

Germination is variable but usually more than one germ-tube arises from each polar cell and occasionally from other cells. Secondary conidiophores and conidia sometimes develop from existing conidia but are rare.

In rich culture media, isolates of *D. tetrarrhenae* produce a flat grey-green mycelium with little aerial growth. Hyphae submerged in the agar develop numerous small chlamydospores and small sclerotia. The fungus does not usually sporulate on rich media unless some host tissue is added. On sucrose–proline agar, a medium used by Shoemaker (1962) to compare cultural characters of *Drechslera* species, *D. tetrarrhenae* produced a flat grey mycelium with few protothecia. Few isolates sporulated abundantly on this medium. There was no pigment production by the fungus into the surrounding medium. In media with added host material, numerous large sclerotia developed. Usually these developed conidiophores which bore conidia after several weeks growth.

Conidia of *Drechslera tetrarrhenae* are smaller than in most other *Drechslera* spp. producing *Pyrenophora* perfect states. It resembles *D. dactylidis* Shoemaker (1962) in some features but differs from it in several ways. Conidia of *D. dactylidis* are usually longer, more highly septate, have a greater length-to-width ratio and taper in markedly at the basal septum. Central cells of this species are compressed, a feature not apparent in *D. tetrarrhenae*. The ascal state of *D. dactylidis* has ascospores with five transverse septa. Ascospores of *Pyrenophora tetrarrhenae* show greater development of transverse septa and the shape of each spore is more fusoid than those of other *Pyrenophora* species.

The only other *Drechslera* with cylindrical conidia having asexual spores of similar dimensions to *D. tetrarrhenae* is *D. erythrospila* (Drechs.) Shoemaker (1962). This form-species has light yellow conidia with a dark basal septum. Isolates of *D. erythrospila* produce a dark chocolate coloured pigment in the agar medium. The sexual state of this fungus is not known.

Collections of Drechslera tetrarrhenae. On *Microlaena stipoides* from Amphitheatre, Avoca, Harrow, Fern Tree Gully, Langi Ghiran, Lexton and Waubra; on *Tetrarrhena juncea* from Belgrave, Daylesford and Mt Macedon.

On leaves of *T. juncea* lesions caused by *D. tetrarrhenae* are discrete, elliptical, up to 15 mm long by 3 mm wide, the long axis being parallel to the leaf veins. Colour is uniformly dark brown to purple, sometimes with a yellow-green margin (Pl. 31, fig. 1). Pathogenicity was tested by spraying a high concentration of conidia on to runners of *T. juncea* which were maintained under moist conditions for 5 days. Large areas of the leaf became infected and irregular chocolate-brown blotches developed over much of the leaf surface. Runners of *T. juncea* sprayed with water and maintained under similar conditions remained unaffected.

On *M. stipoides*, symptoms were variable but usually recognizable by the presence of several short chocolate-coloured leaf stripes. The leaf size and shape of *M. stipoides* varied considerably according to location. On the long narrow leaves common in moist forest areas, the leaf stripe symptom was predominant. In woodland and heath areas where leaves were relatively wide and short, irregular chocolate-brown blotches covered up to half the leaf area.

DRECHSLERA BISEPTATA (Sacc. & Roum.) Richardson & Fraser (Pl. 31, fig. 4).

D. biseptata was originally described as *Helminthosporium biseptatum* Saccardo & Roumeguere in Saccardo (1886). Recently, Richardson & Fraser (1968), following the proposal by Subramanian & Jain (1966) that all graminicolous Helminthosporia be transferred to the genus *Drechslera*, created the new binomial *Drechslera biseptata* (Sacc. & Roum.) Richardson & Fraser. As this form-species is not commonly encountered, a full description of the collections from *Microlaena* and *Tetrarrhena* will be given here.

Conidiophores arise in groups from the substrate and often branch near the base or are simple, erect, straight or nearly so until the first conidium is produced. At the point of emergence from the host, the basal cell of the conidiophore is slightly swollen. Conidiophores are brown and slightly darker in the fertile region than in the stalk. A number of conidia are produced on a series of close geniculations, and the whole conidiophore apex curving irregularly in a hook-like manner. Conidiophore width is 4·5–6 μm in the fertile region. The straight stalk may be up to 200 μm long, but the total length of the fertile region rarely exceeds 40 μm unless conidiophores remain undisturbed for several weeks on natural substrates or culture media in a humid environment.

Conidia are 16–38 μm long by 9–16 μm wide and 2- or 3-septate. Spores are obovate to clavate, with the widest point at or about the distal septum, yellow-brown to orange-brown with the basal cell being generally subhyaline to yellow. The apex of each spore is broadly rounded, the apical compartment semicircular in shape, and the base is rounded, the basal cell being cone-shaped. Spores are not constricted at the septa. The conidial scar is black, prominent and included in the basal contour of the spore wall.

Spores germinate by producing one to three germ-tubes from the basal cell. Germination from central or apical cells occurs rarely. Secondary conidiophores and conidia do not develop from existing conidia.

Collections of Dechslera biseptata. On *Microlaena stipoides*, from Belgrave, Emerald, Fern Tree Gully and Wattle Park; on *Tetrarrhena juncea*, from Belgrave, Kalorama and Mt Macedon.

The naming of *D. biseptata* is unfortunate as most conidia are thrice rather than twice septate as the name implies. This fungus should not be confused with *Helminthosporium triseptatum* Drechsler (1923), a parasite of *Agrostis*, *Dactylis* and *Holcus* spp. which has dark olivaceous, thick-walled, ellipsoidal conidia slightly longer and consistently wider than those of *D. biseptata*.

The most similar form-species to *D. biseptata* is *D. dematioidea* (Drechs.) Subramanian & Jain (≡ *H. dematioideum* Drechsler, 1923), a weak parasite of *Agrostis* and *Anthoxanthum* spp. This species has similar conidial dimensions to *D. biseptata* but spores are characteristically 2- to 6-septate. In addition, conidiophores are usually shorter than 60 μm. The only other species of comparable size is *H. spiciferum* (Bainer) Nicot (1953). However, this species has true bipolar germination and conidia are widest near the centre of the spore.

D. biseptata produced a number of small orange-brown spots up to 3 × 1 mm on mature leaves of *T. juncea*. Where spots were elongated the long axis followed the parallel venation of the leaf. Spots were often surrounded by a halo of yellow green leaf tissue (Pl. 31, fig. 2). *D. biseptata* is abundant on senescing leaves of *T. juncea* and is probably only a weak parasite. It has been isolated from dead leaves of four collections of *M. stipoides*. Under experimental conditions leafspots were produced on leaves of *T. juncea* when sprayed with spore suspensions of *D. biseptata* but did not develop on leaves of *M. stipoides*.

The author wishes to thank Dr H. J. Swart, who prepared the drawings, and Dr M. B. Ellis, who examined cultures sent to the Commonwealth Mycological Institute.

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EXPLANATION OF PLATE 31

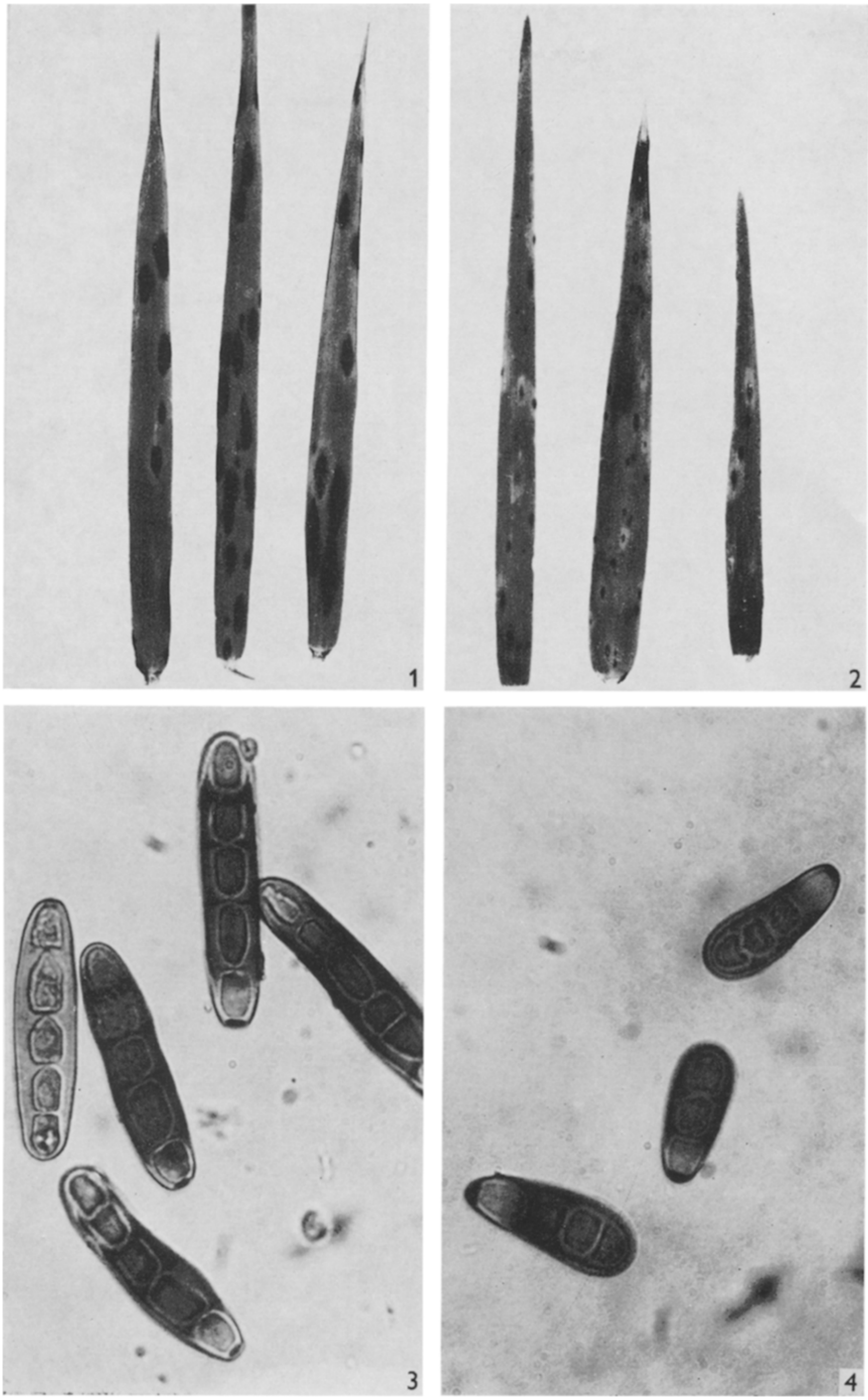
Fig. 1. *Drechslera tetrarrhenae* leafspot on *Tetrarrhena juncea* (natural size).

Fig. 2. *Drechslera biseptata* leafspot on *T. juncea* (natural size).

Fig. 3. Conidia of *D. tetrarrhenae* (× 750).

Fig. 4. Conidia of *D. biseptata* (× 750).

(*Accepted for publication* 29 *October* 1970)



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