

RAMICHLORIDIUM, VERONAEA AND STENELLA: GENERIC DELIMITATION, NEW COMBINATIONS AND TWO NEW SPECIES

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The causative agent of dieback of *Pinus contorta* in Scotland is described as *Ramichloridium pini* sp. nov. The taxonomic relationships of the genera *Veronaea*, *Ramichloridium* and *Stenella* are discussed. Two new combinations are proposed in *Ramichloridium*, and in *Stenella* one new combination and a new species, *S. anomocanis*. The recognized *Ramichloridium* species and related taxa are keyed out.

In the course of 1982, the second author completed a study on a fungal dieback disease of *Pinus contorta* in Scotland. The causative agent was found to be a hyphomycete with erect conidiophores, producing one-celled conidia in sympodial order in the apical region. It was thought to represent a species of *Ramichloridium* Stahel ex de Hoog, close to *R. apiculatum* (Miller *et al.*) de Hoog, but deviating sufficiently to warrant its description as a new species.

***Ramichloridium pini* de Hoog & Rahman, sp. nov. (Fig. 1)**

Coloniae in agar maltoso 20-22 °C post 14 dies ad 8.5 mm diam, velutinae, compactae, ad 2 mm altae, olivaceo-virides; reversum olivaceo-nigrum. Hyphae bifformes, seu hyalinae ad dilute olivaceae, 0.5-2.8 µm latae, seu olivaceae ad fuscae, fere crassitunicatae, 1.8-3.8 µm latae, plerumque verrucosae, saepe substantia capsulari aggregata dilute olivacea circumdatae. Conidiophora simplicia, orthotropica ex hyphis prostratis oriunda, crassitunicata et levia, obscure olivaceobrunnea, ad 60 µm longa, 2.0-3.2 µm lata, continua vel 1-3(-5) septis tenuibus divisa, conidiophora breviora saepe septo basilari carentia; pars distalis conidiophorum cicatricibus planis vel modice prominentibus, subhyalinis vel brunneis, ad 1 µm latis sparsa. Conidia dilute olivacea, fere tenuitunicata, plerumque levia, obovata ad obconica, 3-8 × 2.2-2.8 µm, basi truncata, hilo obscuriora.

Typus CBS 461.82, vivus et exsiccatus, isolatus a M. A. Rahman e ramis *Pini radiatae* morbo affectae, Aberdeen in Scotia.

Colonies on 2% malt agar at 20-22 °C attaining 8.5 mm diam in 14 days, velvety, compact, up to 2 mm high, olivaceous green, later becoming greenish grey, with straight and sharp margin. Reverse olivaceous black. Hyaline, later yellow-brown, gelatinous exudate produced in abundance on Czapek-Dox agar. Hyphae of two types: (a) hyaline to pale olivaceous, 0.5-2.8 µm wide, and (b) olivaceous to dark brown, rather thick-walled, 1.8-3.8 µm wide, mostly verrucose, often with irregular clumps of pale olivaceous, capsular material; hyphae with thin septa at intervals of 10-30 µm. Conidiophores unbranched, arising orthotropically from prostrate hyphae, thick- and smooth-walled, dark olivaceous brown, up to 60 µm long, 2.0-3.2 µm wide, slightly tapering towards the rounded apex, continuous or with 1-3(-5) thin septa, shorter conidiophores often without basal septum; apical portion of conidiophores with scattered, flat or slightly protuberant, subhyaline or brown scars up to 1 µm wide, conidia often directed upwards. Conidia pale olivaceous, rather thin- and mostly smooth-walled, obovate to obconical, 3-8 × 2.2-2.8 µm, base truncate, scars slightly darkened; sporulation scanty. In fresh cultures hyphal clumps with numerous conidiophores were seen.

The species differs from *R. apiculatum* in having restricted colonies, with darker, shorter and stout conidiophores on which relatively few, scattered, usually smooth-walled conidia are formed on rather inconspicuous scars. In addition, the new species

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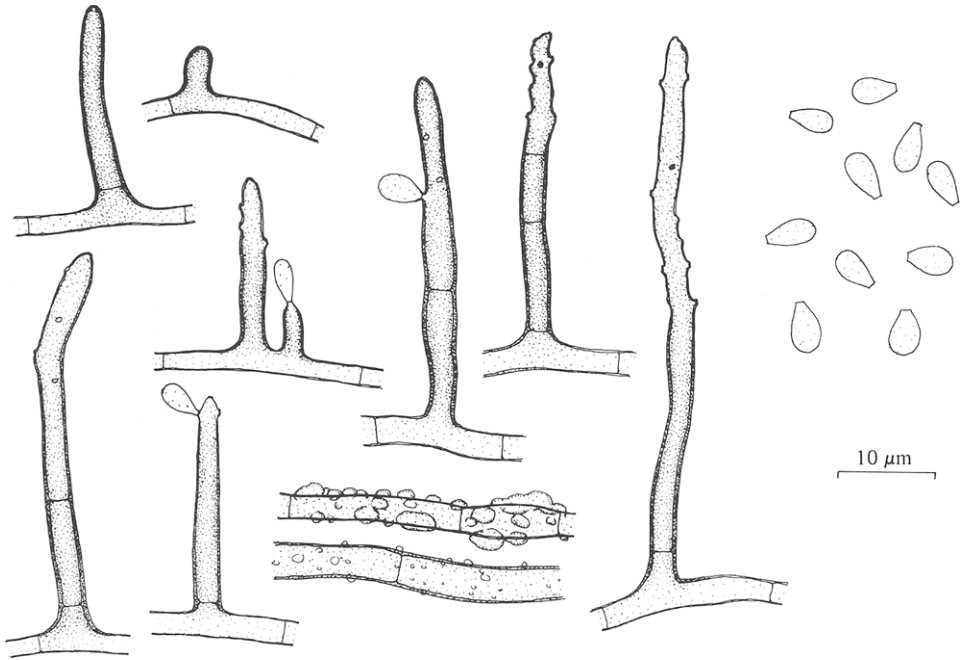


Fig. 1. *Ramichloridium pimi*, CBS 461.82, 2-week-old culture on 2% MEA.

proved to be pathogenic to *Pinus contorta* (Rahman, 1982), while *R. apiculatum* is a common saprophyte (de Hoog, 1977). Both species produce unbranched, typical *Ramichloridium* conidiophores in that they grow out orthotropically from prostrate, intercalary hyphal cells.

Ramichloridium pimi also bears some resemblance to *R. anceps* (Sacc.) de Hoog, though this species has smaller, guttuliform conidia and occurs as a saprophyte on rotten wood, often associated with or overgrowing other fungi. An older synonym of *R. anceps* may be *Psilonia deflexa* Preuss (in Sturm, 1848), but since the type material is not available at herb. Berlin (B) and is probably lost, this species is regarded as doubtful.

Veronaea elliptica Sharma & Munjal was described as having long, erect conidiophores, and one-celled, obovoidal, verruculose conidia. It would be very similar to *Ramichloridium apiculatum*, differing mainly by the much longer conidiophores. Unfortunately the type specimen preserved at HCIO was not available for study, but from the description we believe that it is preferable to reclassify the species as follows: ***Ramichloridium ellipticum*** (Sharma & Munjal) de Hoog, comb.nov. (Basionym: *Veronaea elliptica* Sharma & Munjal, *Kavaka* 7: 75, 1979). With the restriction

of *Veronaea* to species with flat scars and truncate conidial bases (de Hoog, 1977), the following new combination should also be made: ***Ramichloridium bambusae*** (Morgan-Jones) de Hoog, comb.nov. (Basionym: *Veronaea bambusae* Morgan-Jones, *Mycotaxon* 8: 149, 1979).

Several bitunicate Ascomycetes have been described with anamorphs similar to *R. anceps* and related taxa. One of these is *Wentomyces javanicus* Koorders (Joly, 1965). Joly's specimen (preserved at PC) is in poor condition, but some conidiophores present on young perithecia do indeed show much resemblance to *R. anceps*. The conidia should, however, be larger and ellipsoidal to ovoidal in shape. The anamorph of the ascomycete *Berlesella fungicola* Samuels & E. Müller (1978) has small conidia, local annellations and budding cells, and is probably indistinguishable from *R. anceps*.

A remarkable anamorph, with large, thick-walled, pale olivaceous conidiophores and scattered, upward-directed, cylindrical denticles, is that of the ascomycete *Parodiopsis lophiorae* Deighton (Moreau & Moreau, 1959; PC). The pale olivaceous conidia are obovoidal, with slightly protruding scars. A very similar but slightly smaller state is known in *Chevalieropsis ctenotricha* (Pat. & Har.)

Arn. (Arnaud, 1921; Moreau & Moreau, 1955; PC). The anamorph of *Apiosporina morbosa* (Schw.) v. Arx (Ellis, 1976) also belongs to this group. The form-genus *Fusicladium* may be suitable for all these fungi.

There is some disagreement in the literature on the delimitation of *Ramichloridium* and *Veronaea* Cif. & Montemartini. Ellis (1976) defined *Veronaea* as having continuous or few-celled conidia arising from prominent, pigmented scars; Morgan-Jones (1979) stressed the presence of denticles with light scars. The type species of *Veronaea*, *V. botryosa* Cif. & Montemartini, however, is a saprophyte with dark conidiophores and flat, broad, pigmented conidial scars; the conidia have truncate bases. *Veronaea* should therefore be regarded as a close relative of *Pseudospiropes* M. B. Ellis, a genus which comprises species inhabiting putrid wood with multiseptate instead of 1-septate conidia. *Veronaea filicina* Dingley (1972) and *V. compacta* Papendorf (1976) are good species of *Veronaea*. The taxonomic entity conceived by Ellis (1976) and Morgan-Jones (1979) is closer to *Ramichloridium* in that the scars are (sub-)hyaline and often protruding, and the conidial bases are mostly narrow. These characters are often somewhat variable, and dependent on, for example, the age of the conidiophore; this is also the case in *R. pini*. The differences between such species may then be so subtle that whether or not they are recorded can be dependent on the style of drawing of the taxonomist. The strict taxonomic application of these characters, which are evident in some species but unclear or variable in others, is one of the major drawbacks of Ellis's otherwise excellent reviews of dematiacean hyphomycetes. De Hoog (1977) chose to centre *Ramichloridium* on species with conidiophores arising orthotropically from prostrate, mature hyphal cells and with continuous conidia. In this sense the genus comprises mainly weak parasites or saprophytes on living or dead phanerogam leaves; transitions to other anamorph genera are acknowledged.

Ramichloridium musae Stahel ex de Hoog is close to *Periconiella andina* (Syd.) v. Arx (von Arx, 1981). The affinity of *Ramichloridium* and *Rhinochlaediella* Nannf. was discussed by de Hoog (1977) and was further demonstrated by the description of *Rhinochlaediella phaeophora* Veerkamp & W. Gams (1983). *Ramichloridium* is also close to *Cladosporium* Link, a relationship which is exemplified by *R. cerophilum* (Borelli) de Hoog (1977). Recently *Spadicesporium* Borissova & Dvoinos (1982) was described as a genus intermediate between *Ramichloridium* and *Cladosporium*. The relationship of *Ramichloridium* to *Fusicladium* is demonstrated by *Veronaea harunganae* (Hansf.) M. B. Ellis, which is very similar to e.g. *F. viticis* M. B. Ellis. It can be

concluded that a clear-cut taxonomic arrangement of these fungi is impossible. Several species described in *Veronaea*, such as *V. caricis* M. B. Ellis and *V. carlinae* M. B. Ellis, are difficult to place in any genus.

Another line of relationship is with *Stenella* Syd. and *Zasmidium* Fr., both genera comprising species with verrucose, superficial hyphae. *Zasmidium* has catenulate conidia. *Stenella* has been expanded considerably in recent years; it contains mainly tropical species which have been keyed out largely via the host plant (Ellis, 1976). The conidia are usually multiseptate, but several species are known to have a low incidence of continuous conidia. This is seen in *Stenella cynanchi* Yen et al. (1982) in particular. In other species of *Stenella* the one-celled, smooth-walled conidia are a transitional stage in development, mature conidia being brownish, multiseptate and verrucose. *Veronaea gorakhpurensis* also shows such development, the mature conidia all being multiseptate. The species should therefore be renamed as follows: ***Stenella gorakhpurensis*** (Kamal & Kumar) de Hoog, comb. nov. (Basionym: *Veronaea gorakhpurensis* Kamal & Kumar, *Indian Phytopath.* 33: 265, 1980). *Veronaea tectonae* Kamal & Kumar (1980) should probably also be renamed, but this decision awaits examination of the type collection from the CMI herbarium. *Veronaea smilacis* Singh et al. (1981), which is another *Stenella* species, is probably identical to *Stenella smilacis* Kumar et al. (1980), but more material is needed to decide.

A peculiar species collected by the third author showed an abundance of one-celled, apparently mature conidia, intermingled with much larger, multicellular conidia. The species is described as follows:

***Stenella anomoconis* de Hoog & Boekhout, sp. nov. (Fig. 2)**

Coloniae hypophyllae, irregulariter extensae, nigrobrunneae, velutinae. Mycelium reticulum superficiale hypharum dilute flavobrunnearum constanter 1.8–2.2 µm laturum fere tenuitunicatarum verrucosarum distanter septatarum formans. Conidiophora orthotropica ex hyphis prostratis oriunda, plerumque 125–200 µm longa, ad basim 3.0–3.8 µm lata, sursum modice angustata, crassitunicata, 3–12 septis regulariter distantibus tenuibus divisa, levia, primum flavobrunnea, deinde obscurius rubrobrunnea, rhachide sympodiali cicatricibus planis vel modice prominentibus, subhyalinis ad dilute brunneis sparsa. Conidia dilute brunnea, verrucosa, 1–5-cellularia, minora cylindrica, continua, majora obclavata, septis tenuibus divisa, hilo basilari brunneo 1 µm diam; conidia continua 7.5–12.5 µm, septata ad 50 µm longa, amba 2.8–4.0 µm lata. Corpora sclerotialia late ellipsoidea, rubrobrunnea ad nigrobrunnea, ad 50 × 40 µm.

Holotypus CBS H-2723, in foliis putridis *Pruni* sp.,

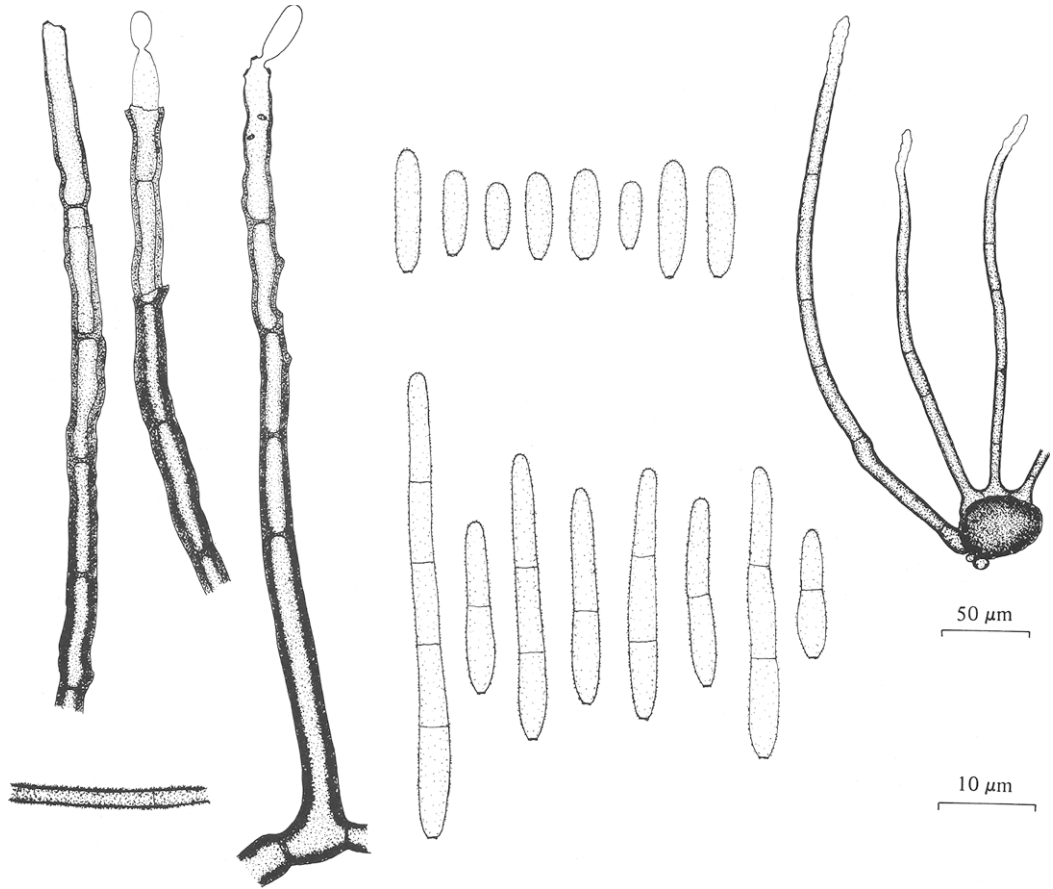


Fig. 2. *Stenella anomoconis*, CBS H-2723 on the natural substrate.

Valle del Rio Totarito, Tolima in Columbia, 3315 m altitudinis, leg. T. Boekhout, 13 Feb. 1980.

Colonies hypophyllous, irregularly spreading, blackish-brown, velvety. *Mycelium* forming a superficial network, hyphae light golden brown, regularly 1.8–2.2 μm wide, walls rather thin, verrucose, with thin septa at large intervals. *Conidiophores* arising orthotropically from prostrate hyphae, mostly 125–200 μm long, 3.0–3.8 μm wide at the base, slightly tapering towards the apex, often with somewhat bulbous base, thick-walled, with 3–12 regularly spaced, thin septa; walls smooth, at first golden, later dark reddish brown, apical portion somewhat lighter, with an up to 200 μm

long rhachis with scattered, flat or slightly prominent, subhyaline to pale brown scars which are directed upwards; conidiophores occasionally percurrently elongating. *Conidia* pale brown, verrucose, 1- to 5-celled, smaller conidia cylindrical, continuous, longer ones obclavate, septate, septa thin, basal scar 1 μm diam, brown; one-celled conidia 7.5–12.5 μm long, septate conidia up to 50 μm long, both 2.8–4.0 μm wide. Sclerotial bodies broadly ellipsoidal, reddish to blackish brown, up to 50 × 40 μm.

The species of *Ramichloridium* and some of their close relatives are keyed out below:

- 1 Conidia usually septate 2
- 1 Conidia continuous, septate conidia rare 6
- 1 Conidia of two types: continuous, and multiseptate. *Stenella anomoconis*
- 2 Conidia mostly 1-septate 3
- 2 Conidia 2- to 3-septate *Veronaea carlinae*

3	Conidia ellipsoidal or clavate	4
3	Conidia obpyriform	<i>Veronaea caricis</i>
4	Conidia clavate	<i>Veronaea flicina</i>
4	Conidia broadly ellipsoidal	5
5	Conidiophores erect, dark brown	<i>Veronaea botryosa</i>
5	Conidiophores inconspicuous, pale	<i>Veronaea compacta</i>
6	Conidial scars markedly darkened, flat	7
6	Conidial scars hyaline to pale brown, often protruding	8
7	Conidia with pointed apex	<i>Fusicladium viticis</i>
7	Conidia with rounded apex	<i>Veronaea harunganae</i>
8	Conidiophore walls over 1 µm thick, pale olivaceous; denticles large, cylindrical; conidia calabash-shaped	9
8	Not combining above characters	10
9	Conidial width 3–5 µm	anamorph <i>Apiosporina morbosa</i>
9	Conidial width 5–7 µm	anamorph <i>Chevalieropsis ctenotricha</i>
9	Conidial width 6–9 µm	anamorph <i>Parodiopsis lophiorae</i>
10	Conidiophores with verticillate or penicillate branching	11
10	Conidiophores unbranched or irregularly branched	12
11	Conidiophore stipe stout, profusely branched at the apex	<i>Periconiella andina</i>
11	Conidiophores with remote verticillate branching	<i>Ramichloridium musae</i>
12	Conidiophores discrete, usually erect	13
12	Conidiophores integrated in a plagiotropically branched system	32
13	Conidiophores with hyaline, sharp, tapering denticles; conidial base usually distinctly acuminate	14
13	Conidiophores with (sub-)hyaline, flat or cylindrical denticles; conidial bases otherwise	16
14	Conidiophores straight, erect	15
14	Conidiophores flexuose	<i>Ramichloridium schulzeri</i> var. <i>flexuosum</i>
15	One-septate conidia present	<i>Ramichloridium schulzeri</i> var. <i>tritici</i>
15	All conidia continuous	<i>Ramichloridium schulzeri</i> var. <i>schulzeri</i>
16	Conidia verruculose	17
16	Conidia smooth-walled	22
17	Conidia (sub-)globose	18
17	Conidia obovoidal, clavate to cylindrical	19
18	Conidia 5–7 µm diam, continuous	<i>Ramichloridium indicum</i>
18	Conidia 6–10 µm diam, frequently 1-septate	<i>Ramichloridium verrucosum</i>
19	Hyperparasitic species	<i>Ramichloridium epichloes</i>
19	On other substrates	20
20	Conidia obovoidal to clavate, continuous	21
20	Conidia cylindrical, occasionally septate	<i>Stenella cynanchi</i>
21	Conidiophores usually less than 60 µm; conidia 3–8 × 2.2–2.8 µm; colonies restricted	<i>Ramichloridium pini</i>
21	Conidiophores usually 40–100 µm; conidia 4–7.5 × 2.5–3.2 µm; colonies spreading	<i>Ramichloridium apiculatum</i>
21	Conidiophores usually over 200 µm; conidia 5–10 × 3–5 µm	<i>Ramichloridium ellipticum</i>
22	Conidia catenulate	<i>Ramichloridium cerophilum</i>
22	Conidia single	23
23	Conidiogenous cells slightly tapering	24
23	Conidiogenous cells cylindrical	25
24	Hyperparasitic species	<i>Ramichloridium meliolae</i>
24	Saprophytic species	<i>Ramichloridium subulatum</i>
25	Conidial scars scattered	<i>Ramichloridium torvi</i>
25	Conidial scars aggregated in a rhachis	26
26	Conidia guttuliform to broadly clavate	27
26	Conidia elongate	29
27	Conidiophores stout	<i>Ramichloridium pini</i>
27	Conidiophores slender	28
28	Conidia 2.5–3.8 × 1.8–2.5 µm	<i>Ramichloridium anceps</i> ; anamorph <i>Berlesiella fungicola</i>
28	Conidia 4–6 × 2–3 µm	anamorph <i>Wentomyces javanicus</i>
29	Conidia ellipsoidal to cylindrical, hyaline	30
29	Not combining above characters	31
30	Conidia 5.5–8.5 × 2–2.5 µm	<i>Ramichloridium musae</i>
30	Conidia 4–5 × 1.5–2 µm	<i>Ramichloridium bambusae</i>
31	Conidia ellipsoidal, pale brown	<i>Ramichloridium obovoideum</i>
31	Conidia fusiform, hyaline	<i>Ramichloridium matsushimae</i>
32	Conidia catenulate	33
32	Conidia single	34

- 33 Conidia verrucose *Zasmidium cellare*
 33 Conidia smooth-walled *Rhinocladiella pedrosoi*
 34 Conidiophores pale olivaceous; conidia continuous. *Rhinocladiella atrovirens*
 34 Conidiophores brown; conidia frequently 1-septate *Rhinocladiella phaeophora*

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