Drechslera longirostrata (Subramanian) and other Drechslera species pathogenic to humans and animals

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Abstract. Four species of Drechslera usually soil saprophytes or plant parasites were recognized over the last few years as agents of deep mycoses with subcutaneous, osteolytic, endocardic and pulmonary localizations as well as superficial mycoses of cornea and skin. The key of the potentially pathogenic fungi Drechslera longirostrata (agent of an endocarditis and spondylodiscitis), Drechslera hawaiensis (agent of a pulmonary mycosis) D. rostrata and D. spicifera (agents of several cases of phaeohyphomycoses) is described. The sensitivity of these species to actual systemic antifungal agents (amphotericin B, ketoconazole, itraconazole) is reported.

Keywords. Drechslera species; mycoses; antifungal agents; amphotericin B; ketoconazole; itraconazole.

1. Introduction

A great variety of fungi, soil saprophytes or plant parasites usually considered as non-pathogenic to man may become pathogenic in humans and animals when the host conditions are favourable; these fungi are called opportunistic (Drouhet 1972) and may produce superficial or deep mycoses, sometimes very severe as we observe with Drechslera longirostrata (Subramanian) comb. nova; this fungus usually isolated from dead leaves (Subramanian 1956; Subramanian and Jain 1966) rice and sorghum (Nath et al. 1970) was recognized recently (Drouhet et al. 1982) for the first time in literature as responsible of an endocarditis and spondylodiscitis in a young woman operated in Martinica for a cardiac valve prosthesis.

Three other species of Drechslera chiefly Graminae pathogens or soil saprophytes were recognized during the last few years as responsible for various human and animal mycoses called phaeohyphomycoses (Ajello et al. 1980) due to the dematiaceous characters of the filaments invading the deep tissues: Drechslera hawaiensis (Fuste et al. 1973; Koenig et al. 1984; Krachmer et al. 1978; McAleet et al. 1981; Young et al. 1978), D. rostrata (Ajello et al. 1980; Forster et al. 1975; McKenzie and Connole 1977) and D. spicifera (Bridger et al. 1960; Estes et al. 1977; Kaplan et al. 1975; Patton 1977; Sobol et al. 1984; Zapater et al. 1975).

In honour of Prof. C V Subramanian, whose work consecrated to the Hyphomycetes (Subramanian 1956; Subramanian and Jain 1966; Subramanian 1983) is internationally unanimously appreciated*, we relate in this contribution the principal mycological and pathological characters of the Drechslera opportunistic species, pathogenic for

* Post-doctoral fellow under Indo-French Scientific Cooperation

* Prof. C V Subramanian was elected President of the International Mycological Association and of the 3rd International Congress of Mycology held in Tokyo from 28th August – 3rd September 1983.
man and animals, as well their sensitivity to antifungal agents in view of an effective chemotherapy.

2. Mycological aspects


2.1 General characters

Taxonomic studies by Ito (1930) and Shoemaker (1957), showed that the genus Helminthosporium was a heterogeneous assemblage of species. This realization led to the creation of 2 new genera—Drechslera Ito (1930) and Bipolaris Shoemaker (1959) to accommodate the anomalous species.

The genus contains 40 to 45 species (Ellis 1971, 1976). Many of the species are cosmopolitan but are more frequent in tropical or subtropical countries. Most of the known host plants belong to the family Graminaceae. Drechslera may cause leaf spot, seedling blights or root rots. Several species are opportunistic and cause diseases in man and animals.

On malt agar the colonies are fast to mild growing, velvety to cottony, grey, brown or blackish. Reverse is dark brown to black, hyphae are hyalin or pigmented. Conidiophores are single or in small groups, generally unbranched, and geniculated due to sympodial elongation; spores are produced through a conspicuous pore (poroconidia) fusiform to ellipsoidal, pale to dark brown, smooth walled with several pseudoseptates (phragmospores) sometimes with end cells pale coloured.

The relationship of Drechslera with other allied genera and connexious anamorph-teleomorph are discussed by Lutrell (1977). On the basis of the spore germination some authors maintain Bipolaris Shoemaker (1959) as a separate genus, the conidia of the Drechslera sensu stricto germinate from any cell and those of Bipolaris by the end cells. Some species showing a protuberant hilum in conidia, like Drechslera rostrata, are placed in the new genus Exserohilum Leonard and Suggs (1974) but we do not follow this opinion.

2.2 Key to the potentially pathogenic species

All these species grow at 37° C, which is an obligatory condition for human and animal pathogenicity.

Drechslera longirostrata Subramanian
Conidiophores thick-walled, single or in groups of 2–5 bearing 2–6 poroconidia at their tip. Poroconidia have numerous septa (up to 24). The basal and rarely apical septum are darker. Conidia with prominent hilum are mostly stout, sometimes abruptly bent.

According to the medium the maximum length varies (as very often with other species) from 310 to 434 μm and the width from 12 to 20 μm. This species is partly distinguished from the closely related Drechslera rostrata on the basis of the length and the arrangement of conidia and top darker septum in Drechslera longirostrata. Groups of conidia, provided with a dark basal septum, give a palm tree like appearance. On the contrary in Drechslera rostrata, conidia provided with a dark basal and top septum are
Figure 1. Morphology of *Drechslera longirostrata* strain P1229-80 from human endocarditis. Malt extract agar (a,b). Sabouraud glucose agar (c,d). a. Multiseptated macroconidia (17 septa) (obj. x 100 oc x 10). b. Macroconidia up to 20 septa (obj. x 40 oc x 10). c,d. Macroconidia (obj. x 40 oc x 10).
arranged in an alternate fashion. Figure 1 illustrates the microscopic morphology of the strain isolated from mycotic endocarditis and osteoarthritis.


Conidiophores single or in small clusters, generally flexuous or geniculate, dark brown, up to 200 μm long and 6–8 μm thick. Poroconidia stout, slightly curved to obclavate with 6 to 10 dark septa, end cells less pigmented, 40 to 180 μm long and 14 to 22 μm thick, with a prominent hilum.

Figure 2 illustrates the microscopic morphology of some of the strains studied:

- IMI 76563—ex soil, India, coll. R. Y. Roy, 1959 (in the catalogue of the Commonwealth Mycological Institute under *D. halodes* which is now considered conspecific with *D. rostrata*).
- IMI 136668—ex leaf of *Sorghum* sp., Cairo, UAR, Coll. O. Abdel-Azim—CMI 1968.
- IMI 197560—ex Hay 3 x IMI 76563, USA, K. Leonard (S, A, A—mating type A).

*Drechslera hawaiensis* (Bugnicourt) Subramanian and Jain ex Ellis 1966. Perfect state: *Cochliobolus hawaiensis*. In 1966 Subramanian and Jain emended the genus *Drechslera* and transferred the species *Helminthosporium hawaiensis* to the genus *Drechslera*.

Conidiophores solitary flexuous to geniculate with several septa, brown coloured up to 100 to 120 μm long and 2–7 μm thick. Poroconidia ellipsoidal, oblong to cylindrical with rounded ends, pale to dark brown, mostly 5 septate, but varying from 3 to 7 septate. In some strain length varies from 12 to 37 μm. Thickness varies from 5 to 11 μm; 5 septate poroconidia are mostly 9 x 25 μ. Figures 3a, b, c illustrate the microscopic morphology of the strain isolated from the case reported by Koenig et al (1984).

*Drechslera spicifera* Nelson. Teleomorph: *Cochliobolus spicifer* Nelson 1964. Conidiophores as in other species arise singly or in small clusters, repeatedly geniculate, with conspicuous scars giving a typical appearance, mid to dark brown, up to 300 μm long and 4 to 9 μm thick. Poroconidia are oblong or cylindrical with rounded ends generally golden brown in colour, with 3 septa length varies from 20 to 40 μm and thickness from 9 to 14 μm. Figure 3d illustrates the microscopic morphology of a human strain of *D. spicifera*.

3. Parasitic and saprophytic life of *Drechslera* species potentially pathogens for man and animals

3.1 *Drechslera longirostrata*

*D. longirostrata* was isolated by Subramanian (1956) from dead leaves of *Borassus flabellifer* and by Nath et al (1970) as seed-borne in rice and sorghum, as well as other seeds (wheat, pearl, millet). Experimental infections carried out at 2 stages of growth of rice and sorghum proved their plant pathogenicity but their role in growing crops remains to be established.

The first human case of mycosis due to *D. longirostrata* was reported by Drouhet et
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Figure 2. Macroconidia of *Drechslera rostrata*. a. strain TP 1260 CMI 197560 b, c. strain TP 1263 CMI 76563 (obj. x 40 oc x 10).
al (1982); the patient a young woman, 20 year old, operated at Fort de France (Martinica) with valvular prosthesis for a congenital heart defect, developed in the following 3 months a mycotic endocarditis diagnosed when she was reoperated in Paris. At surgery there was an aortic valvular vegetation in which hyphal elements were seen microscopically and from which \textit{D. longirostrata} was isolated and identified. In spite of amphotericin B treatment followed by ketoconazole treatment, vertebral arthritis due to invasion of the fungus localized first at the intervertebral disc of 2 lumbar vertebral was extended to 3 lumbar segments. Only combination of the 2 antifungal agents, amphotericin B and ketoconazole, permitted the complete cure of this severe case.

3.2 \textit{Drechslera rostrata}

This species is cosmopolitan and common on Graminae and irregularly isolated from soil (Ellis 1971).

Jones (1975) incriminated \textit{D. rostrata} as an agent of oculomycosis.

Ajello \textit{et al} (1980) described this species as an agent of phaeohyphomycosis in a 12 year-old black boy from Bakersfield (California); the severe chronic and progressive osteolytic infection in the foot of this boy was cured after six months of amphotericin B treatment. An experimental infection was obtained in some mice inoculated by intraperitoneally: a lesion noted in the liver of a mouse was found to contain dematiaceous mycelium. \textit{D. rostrata} was observed as an agent of mycoses in Australian bovines; Pritchard and Chick (1977) described multiple mycetoma in a cow with multiple lesions of the skin, nasal mucosa and lymph nodes and mycelial elements organized into granules; McKenzie and Connole (1977) observed also mycotic nasal granuloma due to \textit{D. rostrata} in cattle.

3.3 \textit{Drechslera hawaiensis}

This species described by the French mycologist Bugnicourt in 1955 was first isolated from rice in Hawai and later from several other plants in many countries but particularly in the tropical and subtropical areas.

\textit{D. hawaiensis} was incriminated as the etiologic agent of a fatal case of meningocerebralitis in Memphis, Tennessee (Fuste \textit{et al} 1973); the victim was a 31 year old woman with an underlying lymphocytic lymphosarcoma. Numerous hyphae, (2–4–2 μm dia.) bizarrely shaped and swollen fungal cells were observed during autopsy in the brain. Experimental infection in guinea pigs and mice were obtained with the human isolate as well as with plant isolates of \textit{D. hawaiensis}.

In France, \textit{D. hawaiensis} has been isolated by Koenig \textit{et al} (1984) from a pulmonary allergic bronchocele in a 12 year old boy who lived in Tahiti. The septate, dematiaceous hyphae were observed in the pulmonary excised lesion. Eosinophilic infiltrative lesions accompanied the pulmonary granuloma; the child is still under ketoconazole treatment. A nasal obstruction with bone erosion caused by \textit{D. hawaiensis} in a 15 year old boy was reported by Young \textit{et al} (1978). Several corneal ulcers due to a \textit{Helminthosporium} sp. presumed to be \textit{D. hawaiensis} according to de Hoog (1983) were reported by Krachmer \textit{et al} (1978). In animals granulomatous proliferations of the nasal mucosa were described in cattle by Roberts and co-workers as due to
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*Helminthosporium anomatum*. Gilman and Abbott; according to de Hoog the fungus of this case is also recognized as *D. hawaiensis*.

3.4 *Drechslera spicifera* (Bainer)

Plant parasites, isolates from textiles, soil and air in all parts of the world, especially in the tropical and subtropical areas were identified as *D. spicifera* (Ellis 1971).

In man, Zapater *et al* (1975) and Forster *et al* (1975) reported the first cases of corneal ulcers due to *D. spicifera*.

A primary cutaneous phaeohyphomycosis in a 5 year old leukemic boy was reported by Estes *et al* (1977). Hyphae and swollen hyphal cells resembling chlamydoecpes were observed in the body specimens. Resolution of the infection occurred following excisional biopsy and systemic amphotericin B therapy. Two cases of chronic pneumonia caused by a *Helminthosporium* sp that retrospectively seems to be close to *D. spicifera* (de Hoog 1983) were reported by Dolan *et al* (1970).

In animals, phaeohyphomycoses due to *D. spicifera* was reported by Kaplan *et al* (1975) in a young horse from Virginia, and by Muller *et al* (1975) in a cat. Clinically the horse had multiple black, pustulous and papular cutaneous plaques, 1–3 cm in diameter; within the tissues septate hyphae measuring 5 to 35 μm in diameter were observed. The cat also showed chronic granulomatous skin lesions as well as the cat mycetoma reported by Bridges and Beasley (1960). A maduromycotic type mycetoma was also reported in an aged mare due to a fungus close to *D. spicifera* (Schauffer 1972), in a dog (Hall 1965) and in a cow (Patton 1977). Recently Sobol *et al* (1984) reported a new case of phaeohyphomycosis of the maxilloethmoid sinus caused by *Drechslera spicifera*.

3.5 Sensitivity of Drechslera pathogenic strains to systemic antifungal agents

The four species of *Drechslera*, potentially pathogenic for humans and animals *D. longirostrata, D. rostrata, D. hawaiensis* and *D. spicifera* are agents of deep mycoses with subcutaneous, osteoarticular, pulmonar or endocardic localisations; their severity of infection and chronicity necessitate systemic chemotherapy by intravenous amphotericin B and oral ketoconazole combined with surgical excision; for superficial cutaneous or ocular localisations, topical treatment (iodide, amphotericin B, pimaricin) showed some limited effects. The infection severity of the first reported case of *D. longirostrata* infection in man (Drouhet *et al* 1982), where a spondylodiscitis extended to 3 lumbar vertebrae as a complication of endocarditis from a valve prosthesis infection, was cured only after combined administration of i.v. amphotericin B and oral ketoconazole; both drugs have been proved unsuccessful when given alone. The minimal inhibitory concentration (MIC) of each individual tested antifungal agent in liquid casitone medium was 1 μg/ml, while complete inhibition of the growth was recorded at 0-12 μg amphotericin B and 0-25 μg/ml ketoconazole, showing synergism which explains the excellent therapeutic effect when the two antifungal agents were used concomitantly. The strains of *D. hawaiensis, D. spicifera* and *D. rostrata* reported from different cases by Young *et al* (1970), Estes *et al* (1977) and Ajello *et al* (1980) were found responsible for severe subcutaneous and osteolytic phaeohyphomycoses and showed their sensitivity to amphotericin B. Their prolonged treatment with this drug associated with surgery permitted complete cure but with sequelae. Among the new
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Table 1. MIC of ketoconazole and itraconazole against *Drechslera* sp after 24 and 48 hr using casitone and YNB liquid media, employing microtiter technique.

<table>
<thead>
<tr>
<th>Strain (Nber)</th>
<th>Ketoconazole (µg/ml)</th>
<th>Itraconazole (µg/ml)</th>
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<tbody>
<tr>
<td></td>
<td>Casitone 24 hr</td>
<td>48 hr</td>
</tr>
<tr>
<td>1</td>
<td>0.39</td>
<td>0.39</td>
</tr>
<tr>
<td>2</td>
<td>0.39</td>
<td>1.56</td>
</tr>
<tr>
<td>3</td>
<td>0.39</td>
<td>0.78</td>
</tr>
<tr>
<td>4</td>
<td>1.56</td>
<td>3.125</td>
</tr>
<tr>
<td>5</td>
<td>1.56</td>
<td>1.56</td>
</tr>
</tbody>
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Concentrations tried: 100, 50, 25, 12.5, 6.25, 3.125, 1.56, 0.78, 0.39 and 0.18 µg/ml.

Media: Casitone complex medium (Drouhet and Dupont 1983) YNB (Difco) + asparagine 1.5 g/l + glucose 10 g/l


imidazole derivatives two systemic antifungal agents, the ketoconazole (Drouhet and Dupont 1983) and itraconazole (R 5121 1) (Van Cutsem et al 1983) are promising drugs in systemic mycoses. The sensitivity of several strains of *Drechslera* species studied in our laboratory showed that these drugs are effective in vitro against these potentially pathogenic fungi for human and animals and may be used in vivo for the treatment of their mycotic infection.

4. Conclusions

Among the great variety of fungi, soil saprophytes or plant parasites considered usually as non-pathogenic for human and animals, four species of *Drechslera* were recognized in recent years as agents of deep mycoses with subcutaneous, osteolytic, endocardic and pulmonary localizations as well as superficial mycoses of cornea and skin. The potential pathogenicity of species of *Drechslera longirostrata* (Subramanian) Nath, *D. rostrata* (Drechsler) Richardson and Fraser, *D. hawaiensis* (Bugnicourt) Subramanian and Jain and *D. spicifera* Nelson, is described as well the pathological manifestations in humans and animals have also been reviewed. *D. longirostrata* reported for the first time as an agent of endocarditis followed by vertebral localizations in a young woman submitted to heart surgery. A pulmonary mycosis was also observed in France in a boy arriving from Tahiti. The other cases reported in literature are particularly from tropical and subtropical countries.

The above mentioned four species are sensitive to systemic antifungal agents (amphotericin B, ketoconazole and itraconazole) but the chemotherapy necessitates in some severe cases a prolonged treatment with combined surgery.

Acknowledgements

The authors express their thanks to Dr Phyllis Stockdale and Dr A Sivanesan (Commonwealth Mycological Institute Kew, UK) for help in identification of the strain.
of *D. longirostrata* and the cultures of various *Drechslera* species sent for comparison. They acknowledge Luce Improvisi, O Ronin, A Piet froid and Mafie-Claire Thibovt for their excellent technical assistance and Dr J Van Cutsem (Jansen Pharmaceutica, Beerse, Belgium) for Ketoconazole and itraconazole samples.

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