

Nutritional physiology and taxonomy of human-pathogenic *Cladosporium-Xylohypha* species

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Physiological profiles of type, authentic and some additional isolates of *Cladosporium-Xylohypha* species of purported herpotrichiellaceous relationship are established. This group comprises melanized catenate hyphomycetes which are prevalently found on the human host. The species are excluded from the genus *Cladosporium* and are classified in the genus *Cladophialophora*. *Taeniolella boppii* is also transferred to this genus. *Cladosporium bantianum* (= *Xylohypha emmonsii*) and *C. trichoides* are considered conspecific and are now referred to as *Cladophialophora bantiana*.

Meso-erythritol, L-arabinitol, ethanol and growth at 40 °C are found to be the most useful criteria for species distinction. The species *Cladosporium carrionii* is found to be heterogeneous. The anamorph of the saprophytic ascomycete *Capronia pilosella* is morphologically similar to an authentic strain of *Cladosporium carrionii*, but physiologically distinct. A diagnostic key for the recognized *Cladophialophora* species and to morphologically similar taxa is provided.

Introduction

The hyphomycete genus *Cladosporium* Link:Fr., characterized by melanized, dry conidia in chains, is phylogenetically heterogeneous, since teleomorphs, where known, belong to different Ascomycete families. The teleomorph of the generic type species, *C. herbarum* (Pers.:Fr.) Link, is *Mycosphaerella tassiana* (De Not.) Johanson, a member of the family Mycosphaerellaceae [1]. This family mainly comprises saprophytic and plant-pathogenic species. Another *Cladosporium* species, *C. carrionii* Trejos, one of the agents of human chromoblastomycosis, has recently been brought in connection with the ascomycete genus *Capronia* Sacc., which is a member of the family Herpotrichiellaceae. The supposed affinity to *Capronia* was based on close morphological similarity to the anamorph of *Capronia pilosella* (Karst.) E. Müller *et al.* [2] and on partial large subunit (LS) rRNA sequencing studies [3]. The latter study showed

that *Cladosporium carrionii* and two *Capronia* species, *viz.* *C. pilosella* and *C. mansonii* (Schol-Schwarz) E. Müller *et al.*, were all found in a single, monophyletic branch, which was taken to represent the (anamorphs of) Herpotrichiellaceae. The cluster also contained *Cladosporium trichoides* Emmons [= *Xylohypha bantiana* (Sacc.) McGinnis *et al.*] and *C. devriesii* Padhye & Ajello, as well as human-pathogenic species of *Exophiala* Carmichael, *Fonsecaea* Negroni, *Phialophora* Medlar, *Ramichloridium* Stahel ex de Hoog and *Taeniolella* Hughes. *Cladosporium herbarum* and other saprophytic *Cladosporium* anamorphs of Mycosphaerellaceae were found at a large phylogenetic distance.

The LS rRNA sequencing study [3] strongly favoured a separation of the melanized catenate hyphomycetes in a group of frequently human-pathogenic species around *Cladosporium carrionii*, probably belonging to the Herpotrichiellaceae, from a group of saprophytic, ubiquitous species around *C. herbarum*, all anamorphs of Mycosphaerellaceae. The two groups can be distinguished morphologically: *Cladosporium carrionii* and related species lack differentiated conidiophores, have pale,

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Table 1 List of strains studied

CBS no.	Status	Corresponding no.	Original name	Depositor	Source	Country
173.52	T	ATCC 10958	<i>Cladosporium trichoides</i>	C. W. Emmons	human brain	USA
364.80		CDC B-2283b	<i>Cladosporium trichoides</i>	L. Ajello	cat brain	USA
328.65			<i>Cladosporium trichoides</i>	G. A. de Vries	dog liver	Curaçao
564.82	T	ATCC 46715	<i>C. trichoides</i> var. <i>chlamydosporum</i>	K. J. Kwon-Chung	human brain	USA
678.79	AUT	CDC B-3658	<i>Xylohypha emmonsii</i>	K. J. Kwon-Chung	cat skin	USA
147.84	T	CDC B-3897	<i>Cladosporium devriesii</i>	A. A. Padhye	human disseminated	Grand Cayman Island
260.83	T	CDC B-1352	<i>Cladophialophora ajelloi</i>	D. Borelli	chromoblastomycosis	Uganda
306.94	T		<i>Cladophialophora arxii</i>	K. Tintelnot	human trachea	Germany
160.54	AUT	ATCC 16264	<i>Cladosporium carrionii</i>	A. Trejos	chromoblastomycosis	Australia
362.70			<i>Cladosporium carrionii</i>	M. J. Campos	chromoblastomycosis	Mozambique
164.54			<i>Cladosporium carrionii</i>	A. Trejos	chromoblastomycosis	Venezuela
454.82			<i>Cladosporium carrionii</i>	G. S. de Hoog	culture contaminant	Netherlands
125.88		ETH ZT-9189	<i>Capronia pilosella</i>	E. Müller	decaying wood	Switzerland
126.86	T		<i>Taeniolella boppii</i>	D. Borelli	human skin	Brazil
316.56			<i>Taeniolella boppii</i>	O. Tappeiner	human skin	Austria
683.92			<i>Taeniolella boppii</i>	E. Döllefeld	human nail	Germany
136.90			<i>Taeniolella boppii</i>	G. Kraepelin	pine needle	Germany
146.33		ATCC 11280	<i>Cladosporium elatum</i>	E. Melin	wood pulp	Sweden

T = type strain, AUT = authentic strain. ATCC = American Type Culture Collection, Rockville; CBS = Centraalbureau voor Schimmelcultures, Baarn; CDC = Centers for Disease Control and Prevention, Atlanta, GA; ETH = Eidgenössische Technische Hochschule, Zürich.

non-protuberant conidial scars, and their conidial chains disarticulate more reluctantly than those of *C. herbarum* and its mycosphaerellaceous relatives, which have dark conidiophore stalks and blackish-brown conidial scars. Espinel-Ingroff *et al.* [4], Kwon-Chung *et al.* [5] and Malkina & Sevzyuk [6] further distinguished the two groups by the thermotolerance and inability to liquefy gelatin shown by the human-pathogens, although some variation was noted within the saprophytes.

The above data require reclassification of the herpochytriacean species of *Cladosporium* in a separate anamorph genus. McGinnis *et al.* [7] noted a morphological similarity to the genus *Xylohypha* (Fr.) Mason. However, Kwon-Chung *et al.* [5] showed that conidial scar ultrastructure is fundamentally different in *Xylohypha*. Concomitantly, Masclaux [3], using LS rRNA partial sequencing, proved *X. nigrescens* (Fr.) Mason to be phylogenetically unrelated to either group of *Cladosporium*.

The generic type species of *Cladophialophora* Borelli, *C. ajelloi* Borelli, is identical to *Cladosporium carrionii* [8]. *Cladosporium carrionii* and related taxa can thus be placed in *Cladophialophora*. The present article classifies the fungi concerned in accordance with supposed teleomorph connections and along lines of phylogenetic relationship indicated by Masclaux *et al.* [3]. In addition, the set of parameters for the identification of the taxa recognized is expanded by using nutritional physiology and tolerance tests.

Materials and methods

Strains studied are listed in Table 1. Stock cultures were maintained on 2% malt–1% yeast extract agar (MYA) slants. Inocula were prepared from fresh subcultures transferred with cells suspended in sterile water. Growth and fermentative abilities were tested in duplicate in liquid medium, essentially using the classical method of yeast physiology described by Van der Walt & Yarrow [9]. For assimilation, cells were grown in test tubes at 25 °C and were aerated by rocking in a nearly horizontal position at 50 rev. min⁻¹. Results were read weekly and compared with positive and negative controls. Final reading was carried out two more weeks after the exhaustion of C-source in the glucose assimilation test tube, tested with Diabur 5000 strips (Boehringer, Mannheim, Germany); some strains with little biomass production were maintained for 60 days in total. Fermentation was tested over a 2 week period in vertical test tubes kept stationary apart from brief manual shaking every 2 days prior to reading. Urease activity [9] was tested on Christensen's agar. Gelatin (12%; Gist-Brocades, Delft, the Netherlands) liquefaction was tested on agar plates, using HgCl₂ in 7·6% HCl as a reagent. Production of extracellular, starch-like compounds was verified in glucose assimilation test tubes after 3 weeks incubation by microscopic examination in Melzer's and Lugol reagents. Acid production was tested with chalk agar in Petri dishes [9]; a positive

result was read as a clear zone around the colony. For extracellular DNase, a commercially available medium (Difco, Detroit, MI) was used. Thermotolerance was tested on 4% malt extract agar (MEA4%) slants, inoculated with fresh cell suspensions and incubated for 2 weeks at 24, 30, 37, 40 and 42 °C. Halotolerance was tested by comparison of biomass production at three salt concentrations (2.5, 5 and 10%, w:v) in shaken liquid medium. Cycloheximide tolerance was tested both in liquid medium at 0.01, 0.05 and 0.1% [9] and on commercial Mycosel agar (0.04%; Becton Dickinson, Cockeysville, MD). Positive controls for all tolerance tests in liquid medium were grown in glucose with yeast nitrogen base (Difco, Detroit, MI). Production of synanamorphs with percurrent conidiogenesis and of isodiametrically expanding cells was checked after 3 weeks' incubation in assimilation test tubes with D-glucose, D-glucosamine, succinic acid and ethylamine; the final pH values of these media were tested with Acilit 0-6 test strips (Merck, Darmstadt, Germany).

Cladophialophora Borelli

Colonies olivaceous, powdery to hairy. Conidia produced in sessile or ascending, branched or unbranched, often firmly adhering, dry, acropetal chains; differentiated conidiophores absent. Conidia one-celled, subspherical, limoniform or fusiform, with smooth, rather firm, melanized walls and with pale scars at the narrow ends. Additional flask-shaped phialides (anamorph genus *Phialophora*) with deep, funnel-shaped, darkly pigmented collarettes occasionally present; phialoconidia hyaline, subspherical, one-celled, adhering in slimy heads. Thick-walled chlamydospores or muriform cells occasionally present. Lactose, nitrate, creatine and creatinine are assimilated. Fermentation is absent. Urease is present. Cycloheximide is tolerated. Gelatin liquefaction is absent. Extracellular starch is not produced. No growth at 10% NaCl.

Type species: Cladophialophora ajelloi Borelli (= *Cladosporium carrionii* Trejos).

Generic discussion

In 1980 Borelli [10] described the genus *Cladophialophora* for an agent of chromoblastomycosis, *C. ajelloi* Borelli. The species was characterized by the presence of phialides in addition to acropetal conidial chains. The catenate synanamorph was later found to be identical to *Cladosporium carrionii* [3,8], as this species was also found to be able to produce phialides on nutritionally poor media (Fig. 1). Phialidic synanamorphs are also known in

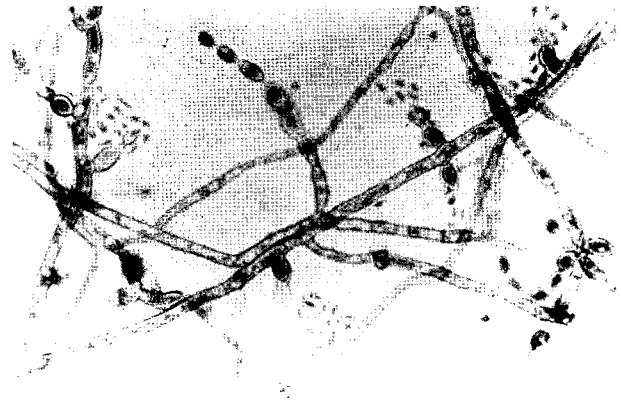


Fig. 1 *Cladophialophora carrionii*, CBS 454.82, catenate conidia and phialoconidia.

related species of *Exophiala* [11] and *Fonsecaea* [12,13]. Generally the phialides are referred to as the *Phialophora*-synanamorph of these species, without attributing a separate binomial. In accordance with this common practice, the name *Cladophialophora* may be used for the prevailing catenate synanamorph of *C. carrionii* and is thus also applicable to congeneric anamorphs without additional *Phialophora* synanamorphs.

All species of *Cladophialophora* have dry conidia arranged in chains which elongate acropetally. Characteristically, differentiated conidiophores are absent and the conidial scars are not or hardly darker than the conidial bodies. *Cladosporium elatum* (Harz) Nannf. has a very similar morphology. It is a very rare species on decaying wood [14] and has once been described from a case of subcutaneous phaeohyphomycosis [15]. Masclaux *et al.* [3] found it to be phylogenetically distant from the mycosphaerellaceous anamorphs, but also from herpotrichiellaceous species of *Cladosporium*. Its sensitivity to cycloheximide and inability to assimilate glucono- δ -lactone, D-glucuronate, creatine and creatinine discriminate it from *Cladophialophora* (Table 2). Since it cannot be maintained in *Cladosporium*, but also seems unrelated to *Cladophialophora*, its taxonomic position remains to be determined.

Conversion to isodiametric growth [16] was noted in several of the test tubes for assimilation, probably due to acidification of the culture medium [17]. Single, pale brown and thick-walled chlamydospore-like cells were produced, transforming to muriform cells in *C. carrionii* [18,19]. In *Cladosporium carrionii* and *Taeniolella boppii* such cells were most common at a final pH of 2.5–3.0. In *C. bantiana*, *C. devriesii* and *C. arxii* optimal production of non-septate swollen cells was found at a final pH of 4–5. Similar structures have been reported in *Exophiala (Wangiella) dermatitidis* (Kano) de Hoog [11] and *Phialophora verrucosa* Medlar [16]. Mendoza *et al.* [16]

Table 2 Physiological pattern of accepted species of *Cladophialophora*, of *Cladosporium elatum* and *Capronia pilosella*

	173.52 <i>banitana</i>	364.80 <i>banitana</i>	328.65 <i>banitana</i>	564.82 <i>banitana</i>	678.79 <i>banitana</i>	147.84 <i>devriesii</i>	306.94 <i>arxii</i>	260.83 <i>carrionii</i>	160.54 <i>carrionii</i>	362.70 <i>carrionii</i>	454.82 <i>carrionii</i>	125.88 <i>Capronia pilosella</i>	126.88 <i>boppii</i>	316.56 <i>boppii</i>	683.92 <i>boppii</i>	136.90 <i>boppii</i>	146.33 <i>Cladosporium elatum</i>
D-Glucose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-Galactose	+	+	+	+	+	+	+	+	W	+	+	+	+	+	+	+	+
L-Sorbose	+	+	+	+	+	+	+	+	W	W	+	+	+	W	+	+	+
D-Glucosamine	-	W	W	W	-	-	-	-	-	-	+	W	W	-	-	W	W
D-Ribose	W	W	+	+	W	W	+	+	W	W	W	+	+	W	W	+	+
D-Xylose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
L-Arabinose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-Arabinose	+	W	+	W	W	+	+	+	+	+	+	+	+	+	W	+	+
L-Rhamnose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Sucrose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Maltose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
α,α-Trehalose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
methyl-α-D-Glucoside	+	+	+	+	+	+	W	+	+	+	-	+	-	-	-	+	W
Cellobiose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Salicin	+	+	+	+	+	W	+	+	W	+	W	+	+	+	+	+	W
Melibiose	+	+	+	W	+	+	+	W	W	+	-	+	+	+	+	+	+
Lactose	+	+	+	+	W	+	+	W	W	W	W	-	-	+	+	+	W
Raffinose	+	+	+	+	+	+	+	W	W	+	+	+	+	+	+	+	+
Melezitose	+	+	+	+	+	+	+	+	W	+	+	+	+	+	+	+	+
Inulin	W	-	-	-	-	-	-	-	W	W	-	-	-	+	+	-	-
Sol. starch	+	+	+	+	W	+	W	W	+	W	+	W	W	W	W	-	W
Glycerol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
meso-Erythritol	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	+
Ribitol	+	+	+	+	W	+	W	W	+	+	+	+	+	+	W	+	+
Xylitol	+	+	+	+	W	+	+	W	+	+	+	+	+	+	+	W	+
L-Arabinitol	-	W	-	W	-	+	W	W	+	+	+	+	W	-	-	-	W
D-Glucitol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	W
D-Mannitol	+	+	+	+	+	+	+	+	W	+	+	+	+	+	+	+	+
Galactitol	+	+	W	+	W	+	+	-	-	-	+	+	+	W	+	W	-
myo-Inositol	+	+	+	+	+	+	+	W	W	+	+	+	W	W	-	+	-
Glucono-δ-lactone	+	+	+	+	+	+	+	W	W	W	+	+	+	+	+	+	+
5-keto-D-Gluconate	+	+	+	W	W	+	+	+	+	+	+	+	+	+	+	-	-
D-Gluconate	+	W	W	+	W	W	+	+	+	+	+	W	+	+	+	-	-
D-Glucuronate	+	+	+	+	+	+	+	?	+	W	+	+	W	W	+	+	-
D-Galacturonate	+	+	+	+	+	+	+	?	+	+	+	+	+	-	+	+	+
DL-Lactate	W	-	-	W	-	W	-	W	-	-	-	-	-	-	-	-	-
Succinate	W	W	W	W	W	-	W	W	W	+	+	W	W	W	-	-	-
Citrate	-	+	+	W	W	W	+	-	-	-	W	-	-	-	-	-	-
Methanol	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Ethanol	+	+	+	+	+	-	+	+	+	+	W	-	+	W	-	-	-
Nitrate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Nitrite	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+
Ethylamine	+	+	+	+	+	+	+	+	W	+	+	+	+	+	+	+	W
L-Lysine	+	+	+	+	+	+	+	+	+	+	+	+	W	+	+	+	+
Cadaverine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Creatine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-
Creatinine	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	W	-
10% MgCl ₂	-	W	-	W	-	-	W	-	W	+	W	W	W	-	W	-	+
10% NaCl	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	W
0.01% Cycloheximide	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
0.1% Cycloheximide	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	-
Mycosel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Urease	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
30C	+	+	+	+	+	+	+	+	+	+	+	W	+	+	+	-	+
37C	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-
40C	+	+	+	+	+	-	+	-	-	-	-	-	-	-	-	-	-
Fermentation	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Gelatin	-	-	-	-	-	-	W	-	-	-	-	-	-	-	-	-	-
Acid production	-	-	-	-	-	-	?	-	-	-	-	-	-	-	-	-	-

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ascribed this conversion to pH-dependent Ca^{2+} -ion exchange. Szanislo *et al.* [20] linked this growth form to the muriform cells found *in vivo* in the tissue of human patients with chromoblastomycosis [21]. The potential presence of isodiametric propagules may be characteristic for purported anamorphs of Herpotrichiellaceae, but the optimal pH for production seems to differ with the species.

All strains reclassified below in *Cladophialophora* were unable to grow with 10% NaCl, did not produce amyloid components, showed no gelatin liquefaction and were unable to ferment sugars with gas production. A relatively large number of carbon and nitrogen sources were assimilated (Table 2) in comparison to strains of *Exophiala* [22]. D-Glucosamine, inulin and DL-lactate were mostly not or only weakly assimilated. In contrast to most *Capronia* species [W. A. Untereiner & G. S. de Hoog, unpublished data], species of *Cladophialophora* are tolerant to cycloheximide and assimilate lactose, creatine and creatinine. Meso-erythritol, L-arabinitol and ethanol are assimilated by the majority of black yeasts [22] but are variable within *Cladophialophora* and can be used for distinction of species.

The species of *Cladophialophora* listed below are frequent aetiologic agents of human mycoses, either cerebral [14,23–25], disseminated [26,27], subcutaneous [28], or of chromoblastomycosis [12,19,29]. Where deep mycoses are concerned, fatality rates are high. *Cladosporium carrionii* has also been obtained from mammals other than humans [30] and from cold-blooded animals [31]. Most species, except for those based on single strains from humans, have been isolated from soil and wood, but proved virulent in experimental infection [32].

Key to accepted species of *Cladophialophora* and morphologically similar taxa

- 1a No growth with 0.1% cycloheximide 2
 1b Growth with 0.1% cycloheximide 3
 2a Growth with galactitol and D-glucuronate
 Cladosporium elatum
 2b No growth with galactitol and D-glucuronate
 Cladophialophora anam. of *Capronia pilosella*
 3a Conidia subspherical; conidial chains sessile on creeping hyphae, without ramo-conidia; no or weak growth with ethanol
 Cladophialophora boppii
 3b Conidia ellipsoidal to fusiform; conidial chains gradually differentiated from hyphae and supported by ramo-conidia; growth or no growth with ethanol 4
 4a No growth with ethanol; conidial chains often borne on denticles 5
 4b Growth with ethanol; conidial chains sessile on cylindrical ramo-conidia 6

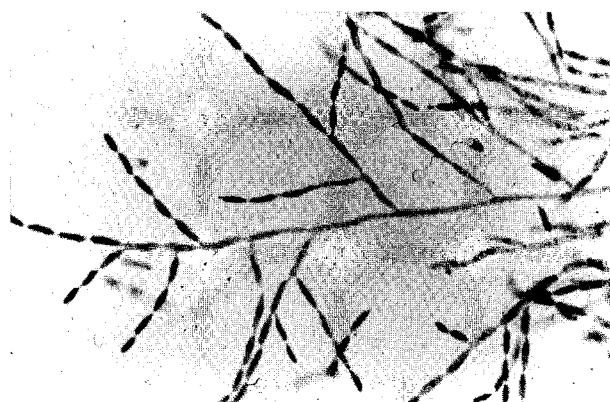


Fig. 2 *Cladophialophora bantiana*, CBS 173.52 ($\times 400$).

- 5a Conidial chains short, branched at most cells; maximum temperature for growth 37 °C
 Cladophialophora devriesii
 5b Conidial chains long, profusely branched; maximum temperature for growth 40 °C *Cladophialophora arxii*
 6a No assimilation of nitrite
 Cladophialophora synanam. of *Exophiala dermatitidis*
 6b Assimilation of nitrite 7
 7a Conidial apparatus with tree-like branching; conidia limoniform, shorter than 6 μm , narrower than 2.5 μm
 Cladophialophora carrionii
 7b Conidial apparatus poorly branched; conidia ellipsoidal, limoniform or fusiform, longer than 6 μm , wider than 2.5 μm
 Cladophialophora bantiana

Cladophialophora bantiana (Sacc.) de Hoog, Kwon-Chung & McGinnis, comb. nov. (Fig. 2)

Synonyms

- Torula bantiana* Sacc.—*Ann Mycol* 1912; 10: 320, (basionym).
Cladosporium bantianum (Sacc.) Borelli—*Riv Anat Patol Oncol* 1960; 17: 620.
Xylohypha bantiana (Sacc.) McGinnis, Padhye, Borelli & Ajello—*J Clin Microbiol* 1986; 23: 1150.
Cladosporium trichoides Emmons in Binford *et al.*—*Am J Clin Pathol* 1952; 22: 535.
Cladosporium trichoides var. *chlamydosporum* Kwon-Chung—*Mycologia* 1983; 75: 320.
Xylohypha emmonsii Padhye, McGinnis & Ajello—*J Clin Microbiol* 1988; 26: 704.

Key features. Conidia olivaceous-brown, limoniform to fusiform, in long, poorly branched chains, smooth- and finally thick-walled. Chlamydospores *in vitro* absent or present. Maximum temperature for growth 42 °C. Methyl- α -D-glucoside, meso-erythritol and ethanol

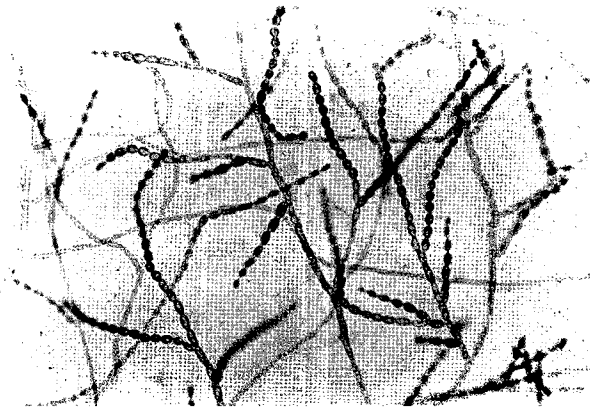


Fig. 3 *Cladophialophora bantiana*, CBS 678.79 ($\times 400$).

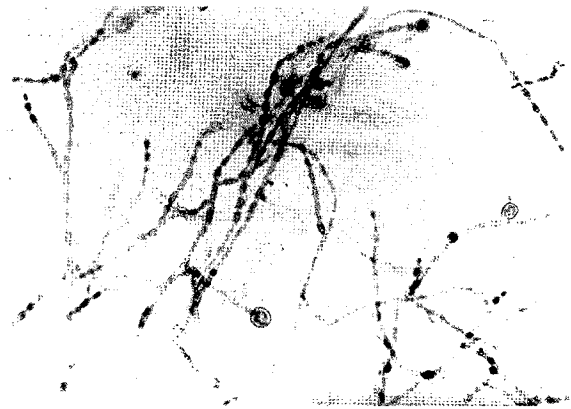


Fig. 4 *Cladophialophora bantiana*, CBS 564.82 ($\times 400$).

assimilated; no or poor growth with L-arabinitol; good or weak growth with ribitol and galactitol. No phialidic synanamorph known. Mole%GC of DNA: 51.4–51.7% (Tm [3]).

Discussion. *Torula bantiana* was originally erected by Saccardo [33] for an agent of cerebral mycosis in a cat, a case reported by Banti [34]. Since then, numerous cases have been reported, mostly of cerebral abscesses in otherwise healthy humans. A recent review is that of Dixon *et al.* [14]; subsequent cases reported include Heney *et al.* [24] and Sekhon *et al.* [25]. McGinnis *et al.* [7] and Padhye *et al.* [35] distinguished *Xylohypha emmonsii* as a taxonomic entity mainly involved in cutaneous phaeohyphomycosis. However, the two species have been found to be conspecific, based on partial LS rRNA sequences and nDNA/DNA reassociation [3]. One of the criteria for distinction, *viz.* absence of growth at 40 °C in *X. emmonsii*, seems unreliable since CBS 678.79, one of the authentic strains of *X. emmonsii* showing characteristic histopathological features of the species in experimental inoculation [35,36], was found to be able to grow at this temperature. Strains from cutaneous and cerebral mycoses show different branching patterns and were distinct in conidial shape and pigmentation, but these characteristics can be influenced by culture conditions [37]. Intraspecific physiological variation in our strains listed as *Cladophialophora bantiana* (Table 2), noted with the assimilation of DL-lactate and citrate, did not coincide with the previous species distinction [7,35]. The variety *chlamydsoporum* was described by Kwon-Chung [38] for an aberrant strain of *C. trichoides*, but the condition-dependent occurrence of chlamydsopore-like cells is fairly common. For this reason, McGinnis *et al.* [39] regarded the two taxa as identical, which was confirmed by Masclaux *et al.* [3] by genomic comparison.

Cladophialophora devriesii (Padhye & Ajello) de Hoog, Kwon-Chung & McGinnis, comb. nov. (Fig. 3)

Synonym

Cladosporium devriesii Padhye & Ajello in Gonzalez *et al.*—*Sabouraudia* 1984; 22: 430 (basionym).

Key features. Conidia pale olivaceous, limoniform to saturn-shaped, formed on distinct denticles in short, branched chains, smooth-walled. Chlamydsopores *in vitro* absent or present. Maximum temperature for growth 42 °C. Assimilation of *methyl- α -D-glucoside*, *meso-erythritol*, ribitol, L-arabinitol and galactitol; no growth with ethanol. No phialidic synanamorph known. Mole%GC of DNA: 52.4% (Tm [3]).

Discussion. The species was described from a single, fatal case of disseminated phaeohyphomycosis [26,40]. Its small conidia in tree-like arrangement are reminiscent of *C. carrionii*. However, conidial chains are shorter, often borne on peg-like outgrowths [41] and conidia are larger. Physiological differences are found with galactitol and ethanol.

Cladophialophora arxii Tintelnot (Fig. 4)

Cladophialophora arxii Tintelnot in Tintelnot *et al.*—*J Med Vet Mycol* 1995; 33: 349–54.

Key features. Conidia pale olivaceous, limoniform to fusiform, in long, profusely branched chains, smooth-walled. Chlamydsopores *in vitro* absent or present. Maximum temperature for growth 42 °C. *Meso-erythritol* and galactitol assimilated; *methyl- α -D-glucoside*, ribitol and L-arabinitol weakly assimilated; no growth with ethanol. No phialidic synanamorph known. Mole%GC of DNA: 52.0% (Tm [3]).

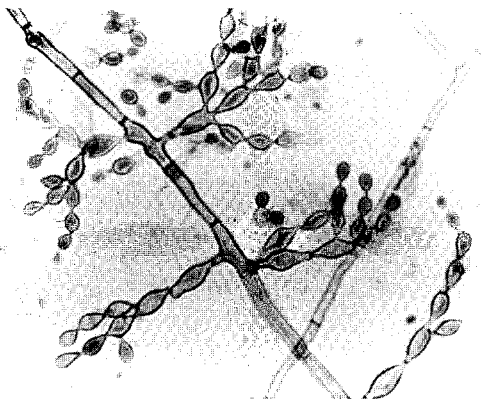


Fig. 5 *Cladophialophora devriesii*, CBS 147.84.

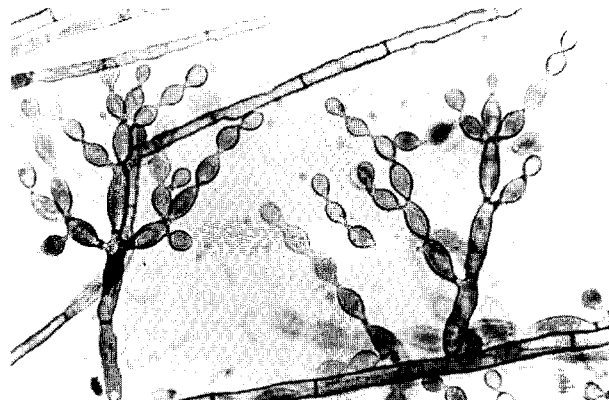


Fig. 6 *Cladophialophora arxii*, CBS 306.94 ($\times 1000$).

Discussion. The species differs from *C. bantiana* in profuse branching of conidial chains and the presence of conidium-bearing denticles [41]; furthermore it is unable to assimilate ethanol. *Cladophialophora devriesii* is very similar, also having denticles, but has shorter conidial chains. The physiological profiles of the two species are nearly identical, except for *C. devriesii* being slightly more temperature sensitive. The two species *in vivo* share a systemic localization outside the central nervous tissue, while *C. bantiana*, when systemic, shows unambiguous neurotropism. Masclaux *et al.* [3] confirmed that *C. bantiana*, *C. devriesii* and *C. arxii* are separate taxa based on low nDNA/DNA homology data and a large number of base changes in partial LS rRNA sequencing.

***Cladophialophora carrionii* (Trejos) de Hoog, Kwon-Chung & McGinnis, comb. nov. (Fig. 5)**

Synonyms

Cladosporium carrionii Trejos—*Revta Biol Trop* 1954; **2**: 106 (basonym).

Cladophialophora ajelloi Borelli—*Pan Am Health Organ Sci Publ* 1980; **396**: 335.

Key features. Acropetal conidia pale olivaceous, limoni-form, arising in long, profusely branched chains, smooth-walled or slightly verrucose. Chlamyospores or muriform cells *in vitro* absent or present. Maximum growth temperature 37 °C. *Methyl- α -D-glucoside*, *meso-erythritol*, *ribitol*, *L-arabinitol* and ethanol can be assimilated; no growth with *galactitol*. Phialidic synanamorph may be present. Mole%GC of DNA: 54.0–54.4% (Tm [3]).

Discussion. The species is nearly exclusively known from cases of human chromoblastomycosis in warm, arid areas [42]. The above set of key features is based on CBS 160.54 and CBS 362.70 from cases of human chromoblasto-

mycosis. CBS 160.54 is the first strain mentioned in the original description [19, p. 106] and is therefore taken as the most representative strain of the species. A poorly sporulating strain from a patient in Venezuela sent by A. Trejos, CBS 164.54, differed by being unable to assimilate *methyl- α -D-glucoside*, by larger conidia and by the occasional occurrence of an *Exophiala* synanamorph. It was found to be identical to *Exophiala jeanselmei* (Langer.) McGinnis & Padhye var. *lecanii-corni* (Benedek & Specht) de Hoog, which occasionally produces chains of barrel-shaped cells [41]. This identity was confirmed by nDNA/DNA reassociation and partial LS rRNA sequencing [3]. Strain CBS 454.82, isolated as a culture contaminant in a strain of *Scytalidium lignicola* Pesante from Northern Europe, was morphologically identical to *C. carrionii* but was *methyl- α -D-glucoside* and *melibiose* negative but assimilated *D-glucosamine* and *galactitol*. The anamorph of *Capronia pilosella*, isolated from wood in Switzerland, resembles *C. carrionii* in morphology, but is weakly sensitive to cycloheximide and unable to grow with *lactose* and ethanol. The two strains were found next to each other in a phylogenetic tree based on partial LS rRNA [3], though at a relatively large distance. The possibility cannot be excluded that *Cladophialophora carrionii*/*Capronia pilosella* are members of a species complex, the strains from temperate climates representing other taxa.

***Cladophialophora boppii* (Borelli) de Hoog, Kwon-Chung & McGinnis, comb. nov. (Fig. 6)**

Synonym

Taeniolella boppii Borelli—*Med Cutan Ibero Lat Am Venez* 1983; **11**: 232 (basonym).

Key features. Conidial chains sessile, very long, unbranched. Conidia olivaceous, subspherical, smooth-walled. Chlamyospores *in vitro* absent or present.

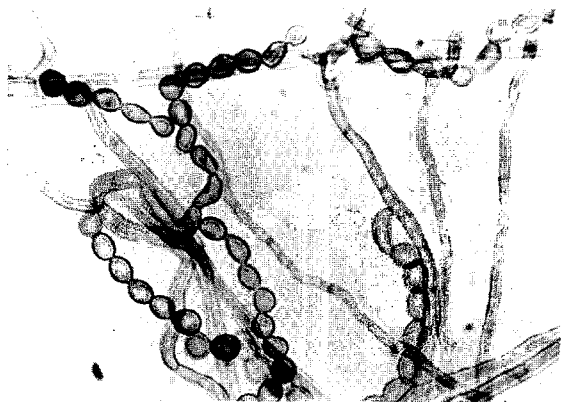


Fig. 7 *Cladophialophora boppii*, CBS 126.86 ($\times 1000$).

Maximum temperature for growth 37 °C. Ribitol assimilated; no or poor growth with *methyl- α -D-glucoside*, *meso-erythritol*, *L-arabinitol* and ethanol; good or weak growth with galactitol. No phialidic synanamorph known. Mole%GC of DNA: 54.6% (Tm[3]).

Discussion. The three strains from skin (CBS 126.86, 316.56 and 683.92) are identical or similar in their growth reactions to *meso-erythritol*, *L-arabinitol* and ethanol, but variable in assimilation of lactose, inulin and *D-galacturonate*. The strain from a pine needle, CBS 136.90, differs conspicuously in assimilation of *methyl- α -D-glucoside*, *5-keto-D-gluconate*, *D-gluconate* and nitrite. A species complex rather than a single species may be concerned.

Acknowledgements

We are indebted to N. Yurlova and J. Boon-Long for their contributions in carrying out the experiments.

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