Arthroderma silverae sp. nov. and Chrysosporium vallenarense, keratinophilic fungi from arctic and montane habitats

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Arthroderma silverae sp. nov. is described from canine dung collected in lowland high arctic areas in Svalbard and from high elevation montane woodlands in Jasper National Park, Alberta, Canada. Arthroderma silverae is similar to other species of Arthroderma in possessing small, smooth-walled, oblate ascospores, anastomosed hyphae forming a mesh-like peridium, the ability to degrade keratin, and rhexolytically dehiscing conidia assignable to the form-genus Chrysosporium. It is distinguishable by confluent rather than discrete ascocarps, peridial hyphal cells which are often curved, irregularly swollen and dichotomously branched, and by lack of ascocarp appendages. An isolate of Chrysosporium vallenarense obtained from arctic fox (Alopex lagopus) dung from high arctic regions near Ny Ålesund, Svalbard, is described and compared to the type strain.

During the Third International Symposium on Arctic and Alpine Mycology, held in Svalbard during August 1988, the senior author collected a number of samples of dung of Alopex lagopus (arctic fox) as a probable substrate for keratinophilic fungi endemic to the high arctic region. A distinctive fungus formed large clusters of ascomata on the surface of several scat samples which had been dehydrated and subsequently incubated in moist chambers. Axenic cultures were obtained from the ascomata. The same fungus was observed again in June 1989 (SPA) fruiting on hair-laden scat of Canis lupus (grey wolf) collected in upper montane woodlands in Jasper National Park, Alberta, Canada. These isolates represent an undescribed ascomycete species belonging in the Arthrodermataceae of the Onygenales, as defined by Currah (1985, 1988). Because of the unique combination of morphological characteristics of the teleomorph and anamorph, placement in either Nannizzia or Arthroderma sensu stricto (Currah, 1985) is unsatisfactory, but the broader generic concept of Arthroderma sensu lato (Weitzman et al., 1986) is appropriate. The new species is described as Arthroderma silverae.

A second keratinophilic fungus recovered from scat of the arctic fox collected in Svalbard was a strain of *Chrysosporium vallenarense* Oorschot & Piont., a hyphomycete known only from a single isolate described from keratin-rich substrate in Chile (van Oorschot & Piontelli, 1985). A description of morphological and cultural features is provided for the Svalbard isolate along with comparisons to the type strain.

Methods of culturing and recording taxonomic data follow Currah (1985) and Sigler & Carmichael (1976). Additional media used to encourage ascocarp production included cornmeal agar (McGinnis, 1980) and Takashio agar (Takashio, 1972). Cultures are deposited in the University of Alberta Microfungus Collection and Herbarium (UAMH).

DESCRIPTIONS OF TAXA

Arthroderma silverae sp. nov.

(Figs 1-7)

Entym.: after E. Silver Keeping, Canadian mycologist

Ascomata alba vel pallido brunnea, globosa, 50–150 µm diam., solitaria vel aggregata ad 4 mm diam. Asci octospori, globosi, $6\cdot5-10$ µm diam., evanescentes. Ascosporae hyalinae, oblatae, laeves, $1\cdot8-2\cdot5 \times 3\cdot5-4\cdot2$ µm. Hyphae peridiorum asperulatae, hyalinae, anastomosantes et ramosae, ad septa constrictae sunt. Status anamorphosis Chrysosporium. Typus: cultura dessicata, UAMH 6517, ex fimo Alopex lagopus.

Ascomata white to cream or buff, globose or subglobose, 50–150 µm diam., rarely discrete, often forming large clusters up to 4 mm diam. (Fig. 1). Asci eight-spored, hyaline, globose to subglobose, 6·5–10 µm diam., evanescent (Figs 2, 3). Ascospores hyaline to buff in mass, oblate, smooth, 1·8–2·5 × 3·5–4·2 µm. Peridial hyphae hyaline to buff, septate, dichotomously branched and anastomosed, moderately thick-walled, densely asperulate, often curved, often irregularly swollen (Fig. 4). Anamorph *Chrysosporium*; aleurioconidia singlecelled, rarely with one septum, pyriform and short-stalked, lateral or terminal, 4–9 × 2–4 µm (Fig. 5); intercalary arthroconidia occasionally present, 7–12 × 2·5–4 µm. When grown with hair on OAT or water agar, marked digestion occurs after 21–60 d, with the formation of penetrating bodies (Fig. 6). In culture at 22 °C after 28 d on OAT, colonies white or



Figs 1–7. Arthroderma silverae. **Fig. 1.** Globose to subglobose ascomata in clusters on OAT after 16 wk. UAMH 7304. Bar, 1 mm. **Fig. 2.** Scanning electron micrograph of a single ascus of smooth-walled, oblate ascospores. UAMH 7304. Bar, 1 μm. **Fig. 3.** Asci and ascospores from a mature ascoma. UAMH 7304. Bar, 20 μm. **Fig. 4.** Thick-walled, densely asperulate and curved peridial hypha with a constriction at septum (arrowhead). (SEM) UAMH. Bar, 5 μm. **Fig. 5.** *Chrysosporium* state of UAMH 6517. Bar, 10 μm. **Fig. 6.** Hair shafts showing penetrating bodies after 20 d incubation of UAMH 7304. Bar, 100 μm. **Fig. 7.** Colonial morphology of UAMH 6517 on OAT after 28 wk. Actual size. **Figs 8–10.** *Chrysosporium vallenarense* UAMH 6914. **Fig. 8.** Colonial morphology on OAT after 28 wk. Actual size. **Fig. 9.** Conidiophore and ellipsoid to ovoid conidia arising from short lateral branches. Bar, 10 μm. **Fig. 10.** Scanning electron micrograph showing vertucose to tuberculate structure of the conidial walls. Bar, 5 μm.

yellowish-white to buff, patchy, with dense raised clusters of ascomata (Fig. 7). On PYE after 14 d, colonies 19–23 mm diam., white to yellowish-white, cottony, convex, margin finely fimbriate; by 28 d, yellowish-white to pale buff, reverse yellow-brown beneath oldest zone, thick, felted cottony, sometimes concentrically sulcate, slightly umbonate, lacking ascocarp production. On CER after 14 d, colonies 15–17 mm, yellowish-white, cottony to felty. Optimum temperature between 15 and 20°; active growth occurs between 4 and 30°. One strain fruited vigorously on a variety of media, but the others produced ascocarps only after incubation for 3–8 wk on OAT, cornneal agar or Takashio agar.

Type material: Holotype a dried culture on Takashio agar, UAMH 6517 from dung of *Alopex lagopus*, Ny Ålesund, Svalbard.

Cultures examined: Svalbard: Currah 12 Aug. 1988 (Sv 32i), ex dung arctic fox (*Alopex lagopus*), Glundnest area, Ny-Ålesund, UAMH 6517 (ex type strain); Currah 14 Aug. 1988 (Sv 43d), ex dung arctic fox (*Alopex lagopus*), Ny Ålesund, UAMH 6518; Canada: Alberta: Abbott 28 Jun. 1989 (SA 79), ex dung of grey wolf (*Canis lupus*), Jasper National Park, Wilcox Pass, in upper montane woodland (2100 m elev.) under *Picea engelmanii, Abies lasiocarpa*, and *Pinus contorta* Douglas ex Loudon, UAMH 7304.

Chrysosporium vallenarense Oorschot & Piont., 1985. Persoonia 12: 487 (Figs 8–10)

In culture (Fig. 8) after 28 d at 22°, colonies on PYE and CER white to pale yellow or pallid brown, reverse brown on PYE with brown diffusing pigment, convex, cottony to somewhat velutinous, margin finely fimbriate. Colonies slow-growing with an optimum temperature between 15 and 18°; growth from 4 to 30°; after 14 d on CER at 25° colonies 5–10 mm diam.; on PYE colonies 9–13 mm diam. Cycloheximide resistant. Keratinolytic as demonstrated by *in vitro* hair digestion with penetrating bodies after 60 d. Conidia 5–10 × 4–6 µm, ellipsoid to ovoid, broadly truncate at base, hyaline (Fig. 9), verrucose to tuberculate (Fig. 10), rhexolytically dehiscing from short lateral branches of the hyaline, septate, vegetative hyphae, 2–4 µm diam.

Cultures examined: Svalbard: Currah 12 Aug. 1988 (Sv 26b), Ny Ålesund, ex dung of arctic fox (*Alopex lagopus*) collected at base of bird cliff near Loven Glacier, UAMH 6914; Chile: near Vallenar, Piontelli 1983, ex keratinic substrates, UAMH 5713 (ex type strain = CBS 627.83).

DISCUSSION

The Arthrodermataceae was established by Currah (1985) to include keratinophilic fungi with smooth, oblate ascospores and anamorphs with rhexolytically dehiscing conidia. The family as originally described contained the genera Arthroderma, Nannizzia, and Ctenomyces. Arthroderma silverae is somewhat intermediate between Arthroderma and Nannizzia, which supports the broad generic concept of Arthroderma proposed by Weitzman et al. (1986) and supported by recent phylogenetic analysis using ribosomal RNA sequence comparisons (LeClerc, Philippe & Gueho, 1994).

A. silverae is similar to other species of Arthroderma in having a mesh-like peridium of relatively thick-walled, asperulate to tuberculate hyphae (Fig. 4) and in having smooth oblate ascospores (Figs 2, 3). Rather than being discrete and globose as in all other species, the ascocarps of A. silverae are confluent and occur in dense clusters. Some characteristics of the new taxon suggest a closer affinity to species formerly treated in Nannizzia. Peridial cells are long and constricted at the septa, but they are irregularly swollen and often curved rather than ossiform. Peridium branching is dichotomous in A. silverae and in species of Arthroderma sensu stricto while it is verticillate in Nannizzia, but this feature is not diagnostic in all cases. A. silverae lacks peridial appendages in contrast to other Arthroderma species which typically produce helical appendages from terminal cells of the peridial hyphae. The microconidial anamorphs of some Arthroderma species are similar to the Chrysosporium state of A. silverae. Unlike most species of Arthroderma, macroconidia are absent in A. silverae.

Ctenomyces, the only other genus in the family, is easily separated from Arthroderma by its thick-walled, ctenoid appendages and oblate-convex ascospores. In addition, the Ctenomyces peridium consists of an inner membranous layer and an outer reticulum of thick-walled hyphae bearing ctenoid or comb-like appendages. The verrucose conidia of the Chrysosporium state of Ctenomyces serratus Eidam are larger $(10-17 \times 7-9 \ \mu\text{m})$ and are produced on swollen conidiogenous cells (Currah, 1985).

The peridial hyphae of *A. silverae* bear a strong resemblance to those of *Nannizziopsis vriesii* (Apinis) Currah, a representative of the Onygenaceae. The mesh-like peridium of tuberculate hyphae constricted at the septa is very similar, but the branching pattern of peridial hyphae is more irregular in *N. vriesii* than the regularly dichotomous branching and anastomosis seen in *A. silverae. N. vriesii* has globose, pitted ascospores (Currah, 1985; Guarro, Cano & de Vroey, 1991), a feature that separates this genus from those in the Arthrodermataceae. *Nannizziopsis vriesii* also differs in having a well-developed arthroconidial anamorph.

One *Chrysosporium* isolate examined (UAMH 3867, provenance unknown) forms dense masses of asperulate, dichotomously branched hyphae constricted at the septa. These are similar to the peridial hyphae of *A. silverae*, but no ascospores were formed by this strain in culture. The conidia are slightly smaller $(4-7.7 \times 2-3.5 \ \mu\text{m})$ and tend to be navicular in shape.

The isolate of *Chrysosporium vallenarense* obtained from Svalbard differs from the type culture in a number of respects: (1) less yellow pigmentation of colonies and weaker brown reverse colours and diffusing pigmentation, (2) slightly faster growth rate, especially at 5°, (3) more markedly tuberculate conidia, and (4) larger and more elongate conidia. The Svalbard isolate may be a unique taxon but is placed in *C. vallenarense* until more isolates are available. *C. vallenarense* may be compared with *C. merdarium* which also has yellowish to yellowish-green colonies and warty conidia but *C. merdarium* grows more rapidly and fails to digest hair with penetrating bodies. A newly described species, *C. pilosum* Gené, Guarro & Ulfig (Gene *et al.*, 1994) is distinguished from *C. vallenarense* by its subglobose to globose conidia and formation of pigmented sterile hyphae.

The occurrence of *Arthroderma silverae* and *Chrysosporium* vallenarense in widely separated geographic areas suggests a restricted habitat preference for arctic and montane regions. These species utilize the keratin-rich substrata which carnivore dung provides, and are moderately psychrophilic, capable of extensive growth at low temperatures $(4-6^{\circ})$.

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