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Mycorrhiza

Multiple mycorrhizal associations of individual calcicole host plants in the alpine grass-heath zone

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Summary. Mycorrhizal colonization of the fibrous roots of alpine grasses and perennial herbs in microhabitats on rendzina soil were examined. Various host plants were associated simultaneously with more than one species of vesicular-arbuscular mycorrhizal fungi. All dominant graminoids had a high degree of endomycorrhizal infection. Septate endophytes (*Phialophora* sp., *Rhizoctonia* sp.) often occurred together with *Acaulospora* sp., *Glomus tenue*, *G. tortuosum* and *Scutelispora calospora* on individual hosts.

Key words: Vesicular-arbuscular mycorrhizae – VAM fungi – Endophytes – Alpine vegetation

Introduction

Patterns of plant colonization are largely determined by the availability of the propagules of mycorrhizal fungi. These fungi are thought to be essential for the survival of many plants in the natural competitive situations and contrasting habitats found in alpine environments (Trappe 1988). Field studies in alpine plant communities demonstrate variable levels of mycorrhizal colonization and the presence of endomycorrhiza- and ectomycorrhiza-forming fungi with various host plants (Haselwandter 1987; Read and Haselwandter 1981). However, a survey of the literature revealed very little systematic work on the range of species of endomycorrhizal fungi and their symbiotic interactions with individual host plants in alpine habitats (Haselwandter and Read 1980).

The present communication on the association of vesicular-arbuscular mycorrhizal (VAM) fungi with dominant calcicole alpine plants describes part of a field study on the disturbance and revegetation of grassland above the timberline, including aspects of plant establishment and populations of indigenous mycorrhizal fungi.

Materials and methods

Constituent floral elements (graminoids, sedges and perennial herbs) together with the surrounding soil were collected in June 1988 and September 1989 from the study site Köllebachtal (a forest district of Füssen on the northern calcareous alps of Bavaria, elevation 1910–1990 m) located in a Seslerio-Semperviretum (Braun-Blanquet and Jenny 1926). Terminal fine roots and lower-order laterals (traced back to individual parent plants) were washed carefully and cut into 2-cm-long segments. Subsamples of both categories of roots (diameter size class 100–400 μm) were clarified in 10% KOH and stained with trypan blue following the methods described by Koske and Gemma (1989). Root segments were then mounted on slides and examined under a compound microscope to determine mycorrhizal colonization and VAM infection levels relative to root length (Kormanick and McGraw 1982), and the intensity of infection using a rating scale from 0 (no infection) to 5 (most intense colonization), as proposed by Trouvelot et al. (1986). Spores of VAM fungi were isolated from rhizosphere soil by wet sieving and decanting (Gerdemann and Nicolson 1963). VAM species were identified using the keys of Trappe (1982) and Schenck and Pérez (1988).

Results

An inventory of the populations of arbuscular mycorrhizal fungi showed a high percentage of root infection of calcicole host plants growing above the timber-line. Colonization ranged from 66% to 100% of root length for mature plants and from 57% to 97% for established seedlings and juvenile plants, respectively, at the end of the growing season. The intensity of colonization by VAM fungi was highest with graminoid species and most of their fibrous roots had a high degree of infection. The intensity of colonization was 2.1–2.4 in *Alchemilla alpina*, 2.3–2.5 in *Festuca pumila*, 2.4–3.4 in *Deschampsia caespitosa*, 2.6–3.0 in *Poa alpina* and 3.1–3.9 in *Sesleria varia*.

All dominant grasses in closed communities (Seslerio-Caricetum sempervirentis) were frequently associated with several species of VAM fungi. The same kind of endomycorrhizal infection was recorded in the Dryadeto-Firmetum with host plants such as *Festuca*

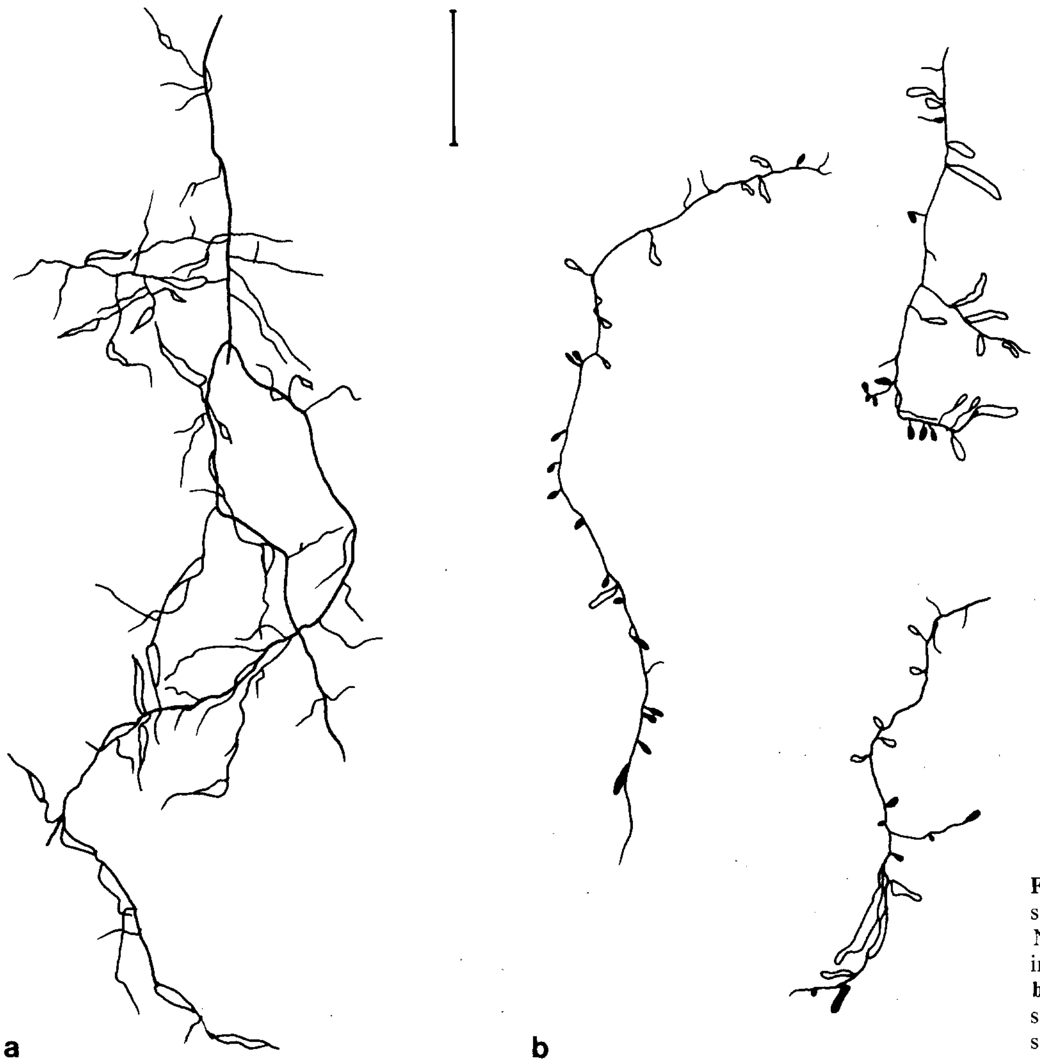

Fig. 1. a Part of the fibrous root system of *Carex sempervirens*. Note the swollen terminal and intercalary carrot-like roots. **b** *Polygonum viviparum* with simple monopodial mycorrhizal short roots. *Scale bar* = 5 mm

pumila and *Carex firma*. Individual swollen, carrot-like roots of sedges (Fig. 1a) occasionally contained branched hyphae and scattered vesicles of an endomycorrhizal fungus (Fig. 2). This has not been reported previously from *Carex* sp. in alpine settings (Haselwandter and Read 1982).

Endomycorrhizal infections with both hyphae and vesicles were observed together with ectomycorrhiza-forming fungi in the same root segments of *Polygonum viviparum* growing in open initial stages on gravelly patches (Figs. 1b, 3, 4).

Hyphal networks of *Glomus tenue* (Fig. 5) and infections by miscellaneous septate (MS) hyphae of secondary endophytes (MS fungi with dark septate hyphae like *Phialophora* sp., *Rhizoctonia* sp.) frequently occurred together on five other constituent hosts, including pioneer plants established on eroded soil. *Scutelispora calospora* and *Phialophora* sp. were found in association with *Deschampsia caespitosa*, *Festuca pumila* and *Sesleria varia*, the latter two host species also being associated with *Glomus tortuosum* (Figs. 6, 7). Coarse extramatrical hyphae of VAM fungi emanated from infested root segments into the soil-root interface and acted as contact points with soil particles by forming stable aggregates. For example, 2.4–2.7 g of soil (oven dry weight) were found to adhere to 1-cm fibrous root segments of *Festuca* tussocks.

Poa alpina ssp. *viviparum* was found in association with *Acaulospora* sp., *Acaulospora trappei* and *Glomus tenue*; 73–94% of the root length was colonized by these VAM species and secondary MS endophytes.

Simultaneous infection of individual host plants by more than one species of VAM fungi was common not only on graminoid host species in closed communities having high root densities but also on constituent pioneer plants found in patches on partly denuded sites above the timberline. A total of eight species of indigenous VAM fungi was recorded in association with dominant host plants on study sites at 1910–1990 m. Again, *Glomus tenue* was recorded most frequently with individual pioneer plants scattered on screes. Fibrous roots of *Silene vulgaris*, which were well represented in gravelly limestone sites, were colonized by MS fungi. Sausage-shaped vesicles of an endomycorrhizal fungus were observed in coarse roots penetrating more than 40 cm into the soil.

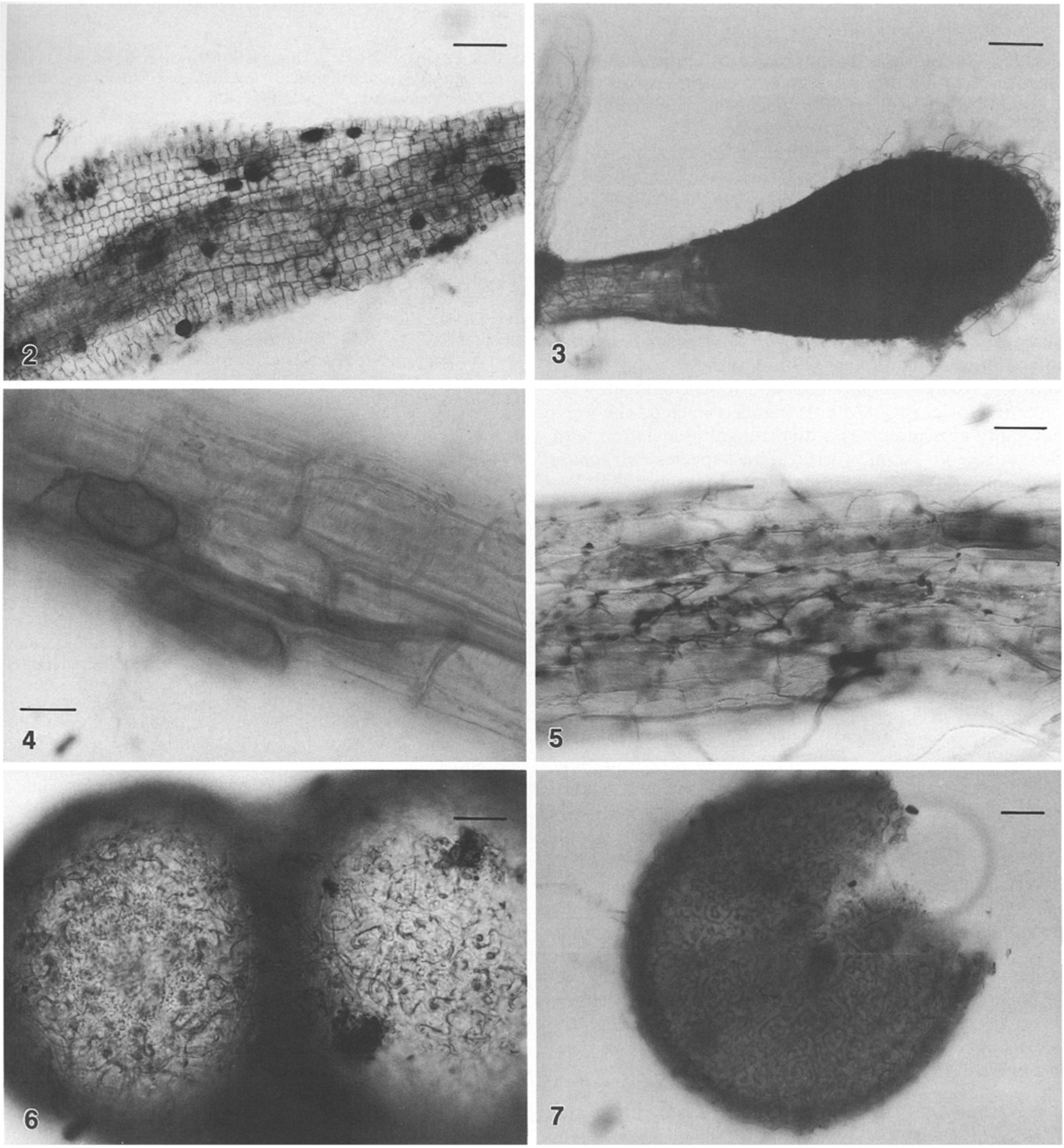

Fig. 2. Vesicles of an endomycorrhizal fungus in a carrot-like root of *Carex sempervirens*. *Scale bar* = 100 μm

Fig. 3. Typical ectomycorrhizal short root of *Polygonum viviparum*. *Scale bar* = 100 μm

Fig. 4. Fibrous root of *Polygonum viviparum*. Note the polymorphic vesicles of an endomycorrhizal fungus. *Scale bar* = 20 μm

Fig. 5. Fibrous root of *Festuca pumila* showing hyphal network of *Glomus tenue*. *Scale bar* = 100 μm

Fig. 6. Two chlamydospores of *Glomus tortuosum* adhering to each other by layers of hyphae. *Scale bar* = 20 μm

Fig. 7. Crushed chlamydospore of *Glomus tortuosum* showing mantle of sinuous hyphae on the spore surface. *Scale bar* = 25 μm

Discussion

All calcicole grasses had mycorrhizal associations and fibrous roots had a high degree of endomycorrhizal infection. Differences in the intensity of colonization of roots appeared to be related to size classes of root diameter, the presence of root hairs and the age of the root (Reinhardt and Miller 1990), even though all fibrous and coarse roots (diameter range 100–1400 μm) were colonized by VAM fungi.

The co-occurrence of different VAM fungi and MS fungal species on a single host seems to be a common and widespread phenomenon (Molina et al. 1978; Read and Haselwandter 1981). Indeed in the grass-heath communities examined, dual and multiple mycorrhizal infection was a consistent feature of the root systems of various hosts. *Phialophora* sp. and *Rhizoctonia* sp. were recorded for most VAM host plants collected. The present study also indicates a dual fungal association with the herbaceous ectomycorrhizal host species *Polygonum viviparum* (Fontana 1977; Harley and Harley 1987). Besides ectomycorrhizal short roots a second endomycorrhizal fungus forming vesicles was noted in the same root segments.

These results suggest that infection of individual roots by more than one species of VAM fungi is typical for host plants in grass-heath communities above the timberline. The pattern of colonization and intraradical development of endomycorrhizal fungi seems to be largely dependent on the species, the morphology and distribution of roots, root growth dynamics and rhizosphere soil conditions (Reinhardt and Miller 1990). As a consequence, on sites examined above the sub-alpine zone, mycorrhizal colonization was highest where guilds were formed, which also suggests contact between root systems and the facilitation of infection due to extramatrical hyphae of already established VAM fungi on pioneer plants (Allen et al. 1987; Powell 1980). Aboveground interactions are probably related to connections between plants via mycorrhizae at both interspecific and intraspecific levels (Miller 1987). Revegetation studies are at present the underway (Blaschke et al. 1990) in order to examine relationships between growth response, stress alleviation and mycorrhizal dependency of some calcicole floral elements on contrasting microsites above the alpine timberline.

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