Mycorrhizal fungi associated with three species of turfgrass

R.E. Koske, J.N. Gemma, and N. Jackson

Abstract: Small plots of highly maintained turfs of creeping bentgrass (*Agrostis palustris* cv. Penncross) and velvet bentgrass (*Agrostis canina* cv. Kingstown) and a marginally maintained stand of annual bluegrass (*Poa annua*) were sampled intensively over a 15-month period to measure the populations of spores of arbuscular mycorrhizal fungi (AMF) associated with their root systems. Direct isolation of spores and trap cultures were used to assess the AMF communities. Spores of more than 18 species of AMF were isolated. The six dominant species (as measured by the abundance and frequency of occurrence of spores) were *Acaulospora mellea*, an undescribed species of *Acaulospora*, *Scutellospora calospora*, *Glomus occultum*, *Glomus etunicatum*, and *Entrophospora infrequens*. Spores of 17 species of AMF were recovered from the root zones of velvet bentgrass, 15 species from creeping bentgrass, and 14 from annual bluegrass. Soil fertility differed among the three sites, and it was not possible to ascribe differences in the AMF communities in each plot to any particular variable (e.g., host, pH, soil P). Average spore abundance was greatest in the creeping bentgrass plot (191.0 spores/100 mL), next in the velvet bentgrass plot (82.4 spores/100 mL), and least in the bluegrass plot (28.4 spores/100 mL). Spores were recovered from a significantly greater percentage of the samples from the bentgrass plots (88.5–96.8%) than from the bluegrass plot (76.6%). Spores of an average of 4.5 species of AMF were isolated monthly from creeping bentgrass, 3.3 from velvet bentgrass and 2.0 from bluegrass. Average species richness and spore abundance were positively correlated in the creeping bentgrass and bluegrass plots ($r = 0.77$, $p = 0.001$, and $r = 0.68$, $p = 0.006$), but not in the velvet bentgrass plot. Spore abundance showed strong seasonal trends in all three plots ($p = 0.03–0.001$), with numbers increasing from spring until November. Richness and abundance declined from December until the following spring. In the bluegrass area, which experienced summer drought, spore populations and richness also showed a precipitous decline in July and August in the 1st year of the study (1990), but not in the 2nd year (1991). No such summer decline occurred in the bentgrass plots that received irrigation. The AMF community that was circumscribed by direct spore counts from the field usually was highly dissimilar to the community that was estimated by trap cultures initiated using soil from the turf areas.

Key words: annual bluegrass, arbuscular mycorrhizal fungi, creeping bentgrass, putting greens, turfgrass, velvet bentgrass.

Résumé : Au cours d'une période de 15 mois, les auteurs ont échantillonné intensivement les sols de petites parcelles de gazon fortement entretenues comportant de l'*Agrostis palustris* cv. Penncross et de l'*Agrostis canina* cv. Kingstown, ainsi qu'une parcelle faiblement entretenue de *Poa annua*, afin de mesurer les populations de spores de champignons mycorhiziens arbusculaires (AMF) associés aux systèmes racinaires de ces plantes. Pour connaître les communautés des AMF, ils ont utilisé l'isolement direct et le trappage. Ils ont isolé les spores de plus de 18 espèces des AMF. Les six espèces dominantes (telles que mesurées par l'abondance et la fréquence de présence des spores) sont l'*Acaulospora mellea*, une espèce non-décrite d'*Acaulospora*, le *Scutellospora calospora*, le *Glomus occultum*, le *Glomus etunicatum* et l'*Entrophospora infrequens*. Ils ont obtenu les spores de 17 espèces des AMF au voisinage des racines de l'*A. canina*, 15 espèces avec l'*A. palustris* et 14 espèces avec le *P. annua*. La fertilité du sol variait selon les trois sites, et il n'a pas été possible d'attribuer les différences observées dans les communautés AMF à aucune des variables en particulier (e.g., hôte, pH, P du sol). La plus grande abondance relative des spores a été retrouvée en présence de l'*A. canina* (191,0 spores/100 mL), suivie de l'*A. palustris* (82,4 spores/100 mL) et la plus faible chez le *P. annua* (28,4 spores/100 mL). Ils ont obtenu des spores d'un pourcentage significativement plus grand d'échantillons provenant de l'*A. canina* (88,5–96,8%) que de ceux provenant de l'*A. palustris* (76,6%). Les spores d'une moyenne de 4,5 espèces d'AMF ont été isolées mensuellement de l'*A. palustris*, de 3,3 pour l'*A. canina* et de 2,0 pour le *P. annua*. La richesse moyenne en espèces et l'abondance des spores montrent entre elles une corrélation positive dans les parcelles de l'*A. palustris* et du *P. annua* ($r = 0,77$, $p = 0,001$ et $r = 0,58$, $p = 0,006$), mais pas dans le cas de l'*A. canina*. L'abondance des spores montre de fortes tendances saisonnières dans les trois parcelles ($p = 0,03–0,001$) les nombres augmentant du printemps jusqu'en novembre. La richesse et l'abondance diminuent de décembre jusqu'au printemps suivant. Dans la surface de *P. annua*, qui a subi une sécheresse estivale, les populations de spores et la richesse

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en espèces ont également montré des chûtes drastiques en juillet et août de la première année (1990), mais pas de la deuxième (1991). Il n'y a pas eu de telle diminution estivale dans les parcelles de l'*A. palustris* qui ont reçu de l'irrigation. La communauté AMF qui s'est révélée par les numérations directes de spores provenant du terrain est très différente de celle qui a été perçue par les cultures pièges mises en marche à partir de sols provenant des surface engazonnées.

Mots clés : *Poa annua*, champignons mycorhiziens arbusculaires, *Agrostis palustris*, verts de golf, gazon, *Agrostis canina*.
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Introduction

The species of arbuscular mycorrhizal fungi (AMF) associated with amenity turfgrasses have received almost no attention despite the knowledge that the fungi may confer a variety of benefits (e.g., improved mineral nutrition and drought tolerance) to host plants. Although an estimated 84% of grass species form arbuscular mycorrhizae (AM) (Newman and Reddel 1987; Trappe 1987), it is generally believed that AMF are of relatively little importance to plants with fine roots and abundant root hairs such as turfgrasses or to plants grown under conditions of high fertility and maintenance (e.g., Baylis 1975; Harley and Smith 1983; Miller et al. 1987; Smiley et al. 1992). Warm-season grasses with coarse root systems are significantly more dependent on mycorrhizae (as measured by promotion of growth) than are cool-season, finer-rooted species (Hetrick et al. 1988, 1990, 1991), but some fine-rooted grasses and turf species are known to respond with increased growth to inoculation with AMF (Hayman 1983; Petrovic 1984; Charest, personal communication). The perception of the minor importance of AMF to turfgrasses seems to have discouraged researchers from sampling highly maintained turfs such as those growing on golf courses and putting greens to assess the AMF populations, and there are few published surveys (Herskowitz and Estey 1978; Rhodes and Larsen 1981).

The present study was designed to identify the species of AMF associated with two perennial turf species used in golf greens, creeping bentgrass (*Agrostis palustris* L. cv. Penncross) and velvet bentgrass (*Agrostis canina* Huds. cv. Kingstown), and a marginally maintained stand of annual bluegrass (*Poa annua* L.), a weedy species that grows as a short-lived perennial when cultivated. All three species are cool-season grasses with C_3 photosynthesis. Our hypothesis was that soil samples from the root zones of the highly maintained bentgrasses would have fewer species of AMF and lower population of spores than would samples from the annual bluegrass plot. The three plots were sampled intensively and frequently over a 15-month period to determine which species of fungi were dominant and the seasonality of spore populations.

Materials and methods

Study site

All soil samples were collected from three sites in the experimental plots of the Turf Research Farm at the University of Rhode Island (U.R.I.) (41°29'N, 71°32'W). One plot each of creeping bentgrass, velvet bentgrass, and annual bluegrass were sampled throughout the study. The bentgrass plots had been in place and maintained as part of long-term field trials for the previous 20 years. Each plot measured 1.5 × 3.0 m and was located in the midst of 72 similar-sized plots of the same species. The two bentgrass areas were separated by approximately 300 m. The bluegrass plot measured approximately 1 × 4 m and was 10 m away from the creeping bentgrass plot. Unlike the bentgrass plots, it was not a pure, dense stand. Tussocks of annual bluegrass were interspersed with plants of Kentucky bluegrass (*Poa pratensis* L.) and creeping bentgrass, and total cover in this plot was approximately 60%.

The soil in which the bentgrasses were growing was a fine sandy loam, while the bluegrass was in a silt loam. Physical characteristics are given in Table 1.

The bentgrass turf was maintained as required (water, lime, fertilizer, and herbicides) and mown weekly throughout the growing season to maintain a height of approximately 1.5 cm. Plots were fertilized during the growing season three times in 1990 (April, June, and September) and twice in 1991 (April and June) with several different low-P formulations (e.g., 15:0:30, 18:3:12, 22:0:16). The bluegrass plot was mown to a height of 5 cm, but it did not receive lime, fertilizer, herbicides, or regular irrigation during the experiment.

Daily rainfall and air temperature measurements were made in a standard U.S. Weather Service station at the site. These are summarized in Fig. 1. Spring warming in 1991 occurred earlier than in 1990. April 1991 was 2.5°C warmer and May 1991 was 5.4°C warmer than the previous year. The average maximum temperature for June 1990 (25°C) was nearly equaled by May (24°C) of the following year. Rainfall patterns over the two years were similar, although spring in 1990 (May–June) was 28% wetter than in 1991.

Collection of samples

Soil samples were collected monthly from May 1990 to July 1991 (except in January and March 1991) from the root zones of the three host species. At each sampling date, five samples were collected from each plot using a 75 × 12 mm soil corer inserted to a depth of 15 cm (volume = 135 mL). Sampling points within each plot were selected randomly, and no point was sampled more than once. In the bluegrass area, care was taken to sample plants that were at least 15 cm from other species, but it is likely that roots of other species sometimes were included in the samples.

In the lab, the upper 3 cm of each soil core containing the leaves, tillers, and thatch was cut away and discarded. The remaining piece was cut vertically into two portions, each with a volume of ca. 40 mL. One portion was processed to recover spores of AMF, and the other was used to set up trap cultures.

Recovery and identification of spores

Spores were extracted from each field sample and from the trap cultures by wet-sieving and sucrose centrifugation (Walker et al. 1982). Spores were mounted in a polyvinyl alcohol solution (Koske and Tessier 1983) on microslides and were identified by comparison with authenticated specimens. Only healthy appearing spores were counted. Voucher specimens are maintained in the authors' collection at U.R.I.

Trap cultures

Trap cultures were initiated monthly from May 1990 to May 1991, except for three months (January–March, 1991). The portions of five soil samples from each monthly collection of each grass were mixed together (total volume = 200 mL) and combined with 250 mL of a 1:1 mix of a calcined clay (Terra-Green®, Oil Dri Corp., Chicago, Ill.) and a coarse gravel from a local quarry. The pH of

Table 1. Some soil characteristics of the study site.

Plot	pH	Nitrate[a]	Phosphorus	Potassium
Creeping bentgrass	5.5*a*	303*a*	159*b*	72*a*
Velvet bentgrass	6.1*a*	245*a*	282*a*	80*a*
Annual bluegrass	4.7*b*	375*a*	51*c*	59*a*

Note: Values are means of five samples. Values in columns followed by the same letter did not differ significantly (Duncan's Multiple Range Test, $p < 0.05$).

[a]Nitrate, phosphate, and potassium are expressed as mg/kg soil.

the granitic gravel was 5.3. Particle size distribution of the gravel was 2–4 mm (15.7%), 1–2 mm (21.6%), 0.50–1.0 mm (26.4%), 0.25–0.50 mm (22.3%), 0.11–0.25 mm (11.0%), and <0.11 mm (3.0%). The gravel was sterilized by steam (2 h each of 2 successive days) and was set aside for 2 weeks before being combined with the Terra-Green and used for plants. After mixing, the gravel–Terra-Green medium contained 3.5 mg/kg nitrate, 55 mg/kg phosphorus, and 53 mg/kg potassium. The pH of the gravel–Terra-Green mix was adjusted to 6.5 with lime and checked with a pH meter using a 2:1 soil–water slurry.

For samples collected from May to Deeember 1990, three trap cultures were set up in Cone-tainers® (tapered plastic tubes measuring 20.5 × 4.0 cm, Steuwe and Sons, Corvallis, Oreg.) for each species each month. Thus, nine trap cultures were established monthly. Pots were seeded with Sudan grass (*Sorghum sudanense* Piper var. *niger*) and maintained in a heated greenhouse for 5–7 months. Pots were fertilized every 2 weeks with 20 mL of a half-strength Hoagland's solution (Hoagland and Arnon 1950) with quarter-strength P and micronutrients and full strength Fe (Epstein 1972). Trap cultures from April and May 1991, were set up in larger plastic containers (Deepots® measuring 6.5 × 25 cm; Steuwe and Sons, Corvallis, Oreg.). A single Deepot was established for each turf species monthly. After December 1990, supplemental light was supplied by high pressure sodium vapor lamps for 16 h/day giving an intensity at the leaf surface of 350–1375 $\mu mol \cdot m^{-2} \cdot sec^{-1}$. At the end of the growth period, 50 mL of soil from the middle of each container was removed, and spores were extracted as described above, identified and counted.

Assessment of AMF communities and statistical tests

Frequency of occurrence of each species was calculated by determining the percentage of samples from which spores of each species were isolated. Differences in frequency of occurrence were analyzed using the chi-squared test. A significance level of $p < 0.05$ was used for all tests.

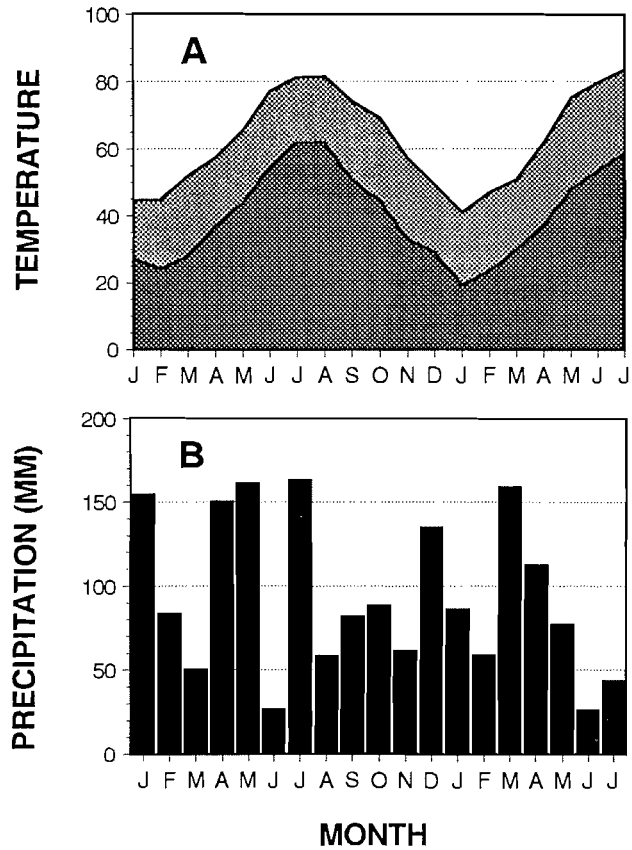
Spore abundance was calculated as spores per 100 mL soil. Spore count data underwent log transformation ($\log(x + 1)$) (St. John and Koske 1988) prior to ANOVA. Means were separated using Duncan's multiple range test ($p < 0.05$).

A spore frequency and abundance (SFA) index was calculated for each species by summing relative frequency and relative abundance of the spores for the entire sampling period. The index is similar to the importance value, an index calculated for higher plants by summing relative density and relative frequency (Mueller-Dombois and Ellenberg 1974), but it differs in being based upon spores rather than upon vegetative individuals.

Monthly species richness of the AMF community associated with each of the turfgrass species was determined by counting the number of AMF species whose spores were recovered from the five collections. An average monthly richness was calculated from the individual monthly values. Differences were compared using one-way ANOVA and Duncan's multiple range test.

A modified Sorensons' Coefficient of Similarity (Bray and Curtis 1957; Southwood 1978) was calculated to compare the AMF populations isolated throughout the year from different plots. Similarity (S) was calculated by the formula:

Fig. 1. Weather data at the study site from January 1990 through July 1991. A. Average monthy temperature maxima and minima. B. Average monthly precipitation.



$$[1] \quad S = \frac{2cN}{an + bN}$$

where aN is the average number of spores (all AMF species combined) per 100 mL isolated from plot a over the 15-month sampling period, bN is the average number of spores per 100 mL recovered with plot b, and cN is the sum of the lesser values of spore abundance of species common to both plots. The maximum value for this index, 1.00, indicates exact similarity.

Similarity between populations of spores isolated from field collections and their corresponding trap cultures was assessed by applying a modified version of the above formula. Because of the large difference in the abundance of spores in culture versus the field, spore counts were recalculated as relative abundance prior to the analysis. To avoid the problem of spores from the original inoculum being counted in the trap culture, only those species whose spores were present at an absolute abundance of greater than 5 per 100 mL were included in the calculation. For consistency, species from the field collections also had to exceed 5 spores/100 mL for inclusion in this comparison. Similarity (S) was calculated by the formula:

$$[2] \quad S = \frac{c}{100}$$

where c is the sum of the lesser values of relative abundance of species common to both field and trap collections. The maximum value of the coefficient is 1.00.

Results

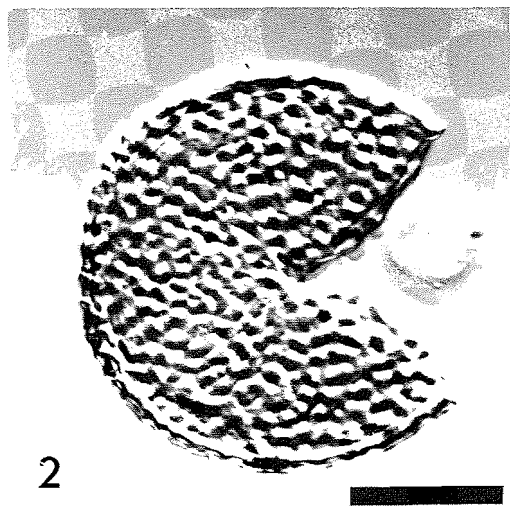
Field collections

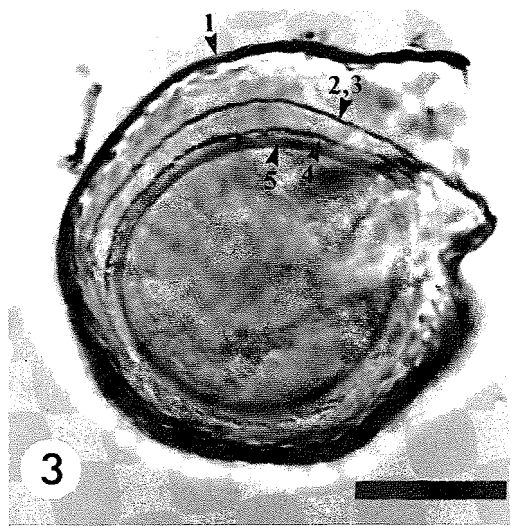
Spores of more than 18 species of AMF were isolated from the three sites over the study period. Spores of 18 of these species occurred with sufficient frequency or abundance or were morphologically distinct enough to be identified and followed throughout the study. In addition, there were several *Glomus* species whose spores were not distinctive and occurred only occasionally and in low abundance. These species were not identified, although their presence and abundance were recorded. They were combined as a single entry, *Glomus* spp., in the tables but were excluded from the calculations of richness and similarity (see below).

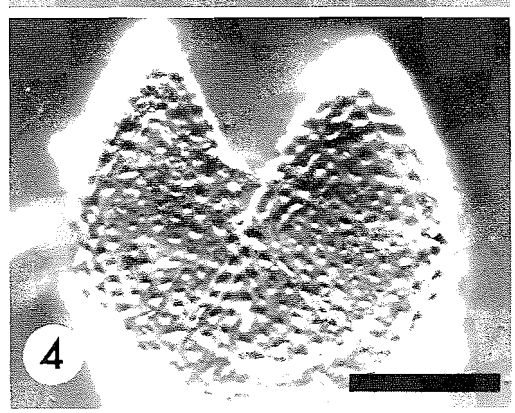
Of the 18 species, 4 are undescribed and are given informal descriptions below. Terminology follows that recommended by Walker (1983, 1986) and Morton (1986). In the following listing, comments on the extent of the population of spores of individual AMF species are based on the calculated SFA index. Species were assigned to one of four importance categories on the basis of their contribution to the total spore population: none (SFA = 0), minor ($0 < \text{SFA} \leq 10$), moderate ($10 < \text{SFA} \leq 30$), and major (SFA > 30).

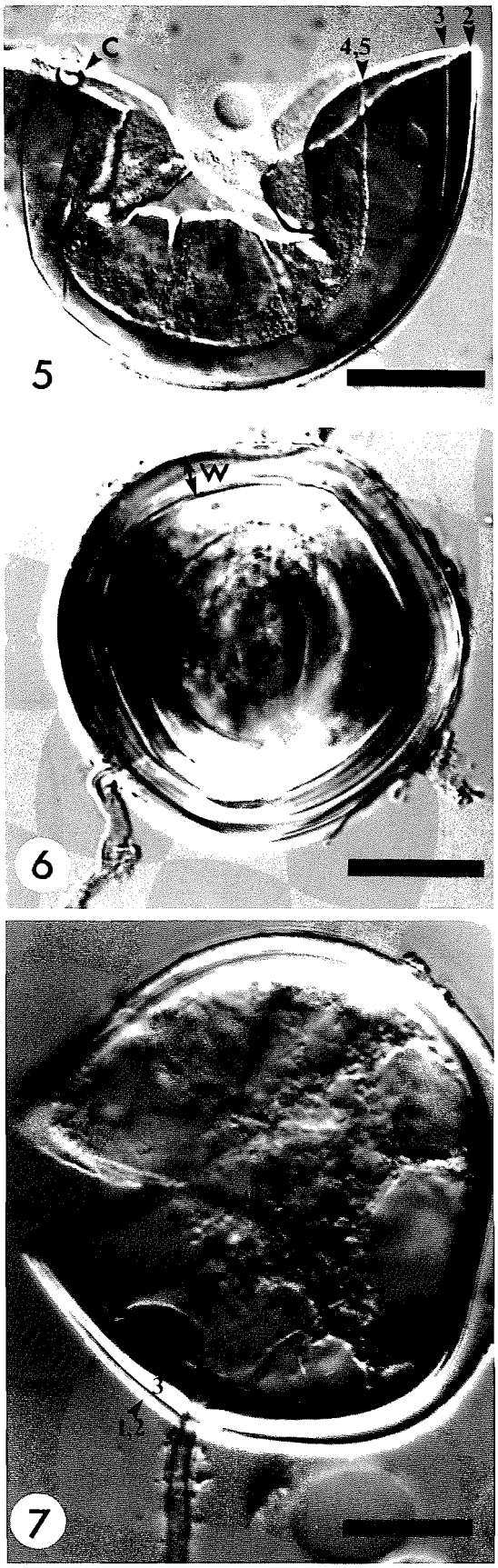
1. *Acaulospora gerdemanni* Schenck and Nicol.
   Importance: none (bentgrasses) to moderate (bluegrass)
   Spores were isolated only from the bluegrass plot. The other spore-state of this species, *Glomus leptotichum* Schenck and Smith, was isolated from all three plots.
2. *Acaulospora mellea* Spain and Schenck
   Importance: minor (bluegrass) to moderate (bentgrasses)
   Spores of *A. mellea*, like those of *A.* 3106, were more frequent and abundant in the bentgrass plots than in the *Poa annua* plot.
   Acaulospora mellea is a member of a group of *Acaulospora* species whose spores closely resemble each other in size and wall structure (i.e., wall 1 membranous, wall 2 laminated, walls 3 and 4 membranous and occurring as a close pair, wall 5 a beaded, membranous wall adherent to the amorphous wall 6). The original description of the species does not include 6 walls, but these have been reported by Morton (1994).
3. *Acaulospora paulinae* Blaszk. (Fig. 2)
   Importance: none (bluegrass and creeping bentgrass) to minor (velvet bentgrass)
   This is another member of the group of *Acaulospora* spp. that includes *A. mellea* (see above). Spores were globose, golden yellow, and 80–95 μm in diameter. Spores of *A. paulinae* were found only in the velvet bentgrass plot and were present in low abundance. This species has been reported previously from Poland (Blaszkowski 1994).
4. *Acaulospora* 3106 (Figs. 3, 4)
   Importance: minor (bluegrass) to major (bentgrasses)
   Spores smokey yellow–gray, translucent, globose to irregular, 60–100 × 60–100 μm. Spore wall structure of 5 walls. Wall 1 a laminated wall 2–3 μm thick, ornamented with scattered, elongated depressions (1.0–)2.5–5.0 × (1–)1.5–2.5 μm × 1.2 μm deep. Walls 2 and 3 adherent membranous walls, each <0.5 μm thick. Wall 4 a beaded membranous wall 0.5 μm thick, adherent to wall 5. Wall 5 amorphous, staining purplish red in Melzer's reagent, 2–3 μm thick in lightly crushed

Figs. 2–4. Spores of *Acaulospora* spp. Fig. 2. *Acaulospora paulinae*. Ornamentation of the laminated wall is shown. Scale bar = 40 μm. Fig. 3. Wall structure of spore of *A.* 3106. Walls 1–5 are indicated. The ornamentation of wall 1 is apparent on the edges of the spore. Walls 2 and 3 are tightly paired and are not distinctly separated in this figure. Note beaded membranous wall (wall 4) adherent to the amorphous wall 5. Scale bar = 30 μm. Fig. 4. Pitted surface of spore of *A.* 3106. Scale bar = 30 μm.









Figs. 5–7. Spores of AMF from turf. Fig. 5. Wall structure of spore of *A.* 3566. Outer membranous wall (wall 1) is not evident. Note beaded membranous wall (wall 4) adjacent to the innermost wall 5, a coriaceous wall. The circatrix is indicated (*c*). Scale bar = 40 μm. Fig. 6. Spore of *Glomus* 3633. Note thick laminated wall (*w*) and narrow attachment hypha. Scale bar = 40 μm. Fig. 7. Spore of *Glomus* 3638. Walls 1 and 2 have separated from the thicker, laminated wall (wall 3). Note relatively narrow attachment hypha. Scale bar = 20 μm.

spores. Circatrix 5–6 μm diam. The ornamentation of spores of *A.* 3106 is similar to that of *A. scrobiculata* Trappe (Trappe 1977), to WUM 18 (Hall 1984), and to *A. paulinae* Blaszk. (Blaszkowski 1994).

Acaulospora 3106 had the highest SFA indices in both of the bentgrass plots, but it was rarely isolated from the bluegrass plot. In association with creeping bentgrass, it had the highest SFA (71.1) recorded in the study. Spores of *A.* 3106 occurred in 32.3% of all the samples, and its average spore abundance (47.2/100 mL) was the greatest of any AMF species.

5. *Acaulospora* 3556 (Fig. 5)

   Importance: none (bluegrass) to minor (bentgrasses)

   Spores globose, pale yellow, 70–82 μm in diameter. Spore wall structure of 5 walls. Wall 1 hyaline, membranous, <0.5 μm thick, often missing, frequently with adherent debris when present. Wall 2 smooth, brittle, laminated, pale yellow, 0.5–2.2 μm thick. Wall 3 membranous, <1.0 μm thick. Walls 4 and 5 closely adherent. Wall 4 beaded, membranous, 0.5–0.8 μm thick. Wall 5 coriaceous, 0.8–1.5 μm thick. Circatrix 7–8 μm in diameter.

   Spores of *A.* 3556 are similar to those of *A. mellea* in size and color, but differ in having a thinner laminated wall, a smaller circatrix, a single membranous wall between the laminated wall and the beaded wall, and an innermost coriaceous wall instead of an amorphous wall.

   Spores of this species occurred only in the bentgrass plots.

6. *Entrophospora infrequens* (Hall) Ames and Schneider

   Importance: minor (bluegrass) to moderate (bentgrasses)

   Spores of *E. infrequens* were isolated frequently (30–44%) from the bentgrass plots.

7. *Gigaspora gigantea* (Nicol. and Gerd.) Gerd. and Trappe

   Importance: none (bluegrass) to minor (bentgrasses)

   Spores occurred only in the two bentgrass plots where they were infrequent and in very low abundance.

8. *Glomus etunicatum* Becker and Gerd.

   Importance: minor (velvet bentgrass) to moderate (bluegrass and creeping bentgrass).

   Spores of this species occurred in 28.6% of all samples. *Glomus etunicatum* had its highest SFA index (16.3) in association with creeping bentgrass.

9. *Glomus leptotichum* Schenck and Smith

   Importance: minor

   Spores of *Glomus leptotichum* occurred in less than 11% of the samples. Its SFA index was greatest (5.9) in the bluegrass plot. See *A. gerdemanni* (above).

10. *Glomus microaggregatum* Koske, Gemma, and Olexia

    Importance: minor

Spores occurred in all plots and were most frequently recovered from the bluegrass area (15.6% of the samples). The small spores of this species typically were found inside dead spores of other AMF (Koske et al. 1986).

11. *Glomus mosseae* (Nicol. and Gerd.) Gerd. and Trappe
    Importance: minor
    Spores were infrequently recovered in association with all three host species.
12. *Glomus occultum* Walker
    Importance: minor (creeping bentgrass) to major (velvet bentgrass and bluegrass)
    This species sporulated abundantly in the velvet bentgrass and bluegrass plots. Its spores were found in 40.0% of all samples examined.
13. *Glomus* 3633 (Fig. 6)
    Importance: minor (bentgrasses) to major (bluegrass)
    Spores globose to subglobose, glistening yellow, 75–130 μm in diameter. Spore wall structure of 2 walls. Wall 1 evanescent, hyaline-gray, 1–5 μm thick, adherent to wall 2. Wall 2 laminated, yellow, very refractive, 5–9 (–18) μm thick. Attachment hyphae hyaline to pale yellow, often broken off at spore base, 8–9 μm wide, walls up to 1 μm thick.
    Spores of *Glomus* 3633 typically are very thick walled and have a glistening appearance in the compound microscope. The laminate nature of wall 2 is evident in most spores, and the laminations separate readily in crushed specimens. The laminate wall is similar in refractivity to the laminated wall of *Glomus nanolumen* Koske and Gemma.
    Spores were significantly more abundant in the bluegrass samples, and the highest SFA index for this species (30.6) occurred in association with bluegrass.
14. *Glomus* 3638 (Fig. 7)
    Importance: minor (bentgrasses) to moderate (bluegrass)
    Spores hyaline, glistening, globose to subglobose, 90–125 × 90–140 μm, produced singly in the soil. Spore wall structure of 3 walls. Wall 1 evanescent, hyaline, 1.0–1.2 μm thick, appressed to wall 2. Wall 2 a unit wall, hyaline, 1.0–1.4 μm thick. In young spores, walls 1 and 2 are present as a distinct pair of walls, but in age they sequentially become granular and are lost, leaving the persistent wall 3. Wall 3 laminated, glistening, 3–4(–5) μm thick, and with easily visible laminations. Attachment hypha concolorous with spore, 6–8 μm wide; walls of attachment hyphae formed from a continuation of the laminated spore wall, up to 2 μm thick. When walls 1 and 2 have been lost, spores are very smooth. The loss of walls and the easily visible laminations of wall 3 that continue into the attachment are reminiscent of the spores of *Glomus intraradices* Schenck and Smith. *Glomus laccatum* Blaszk. produces spores similar in color, size, and smoothness, but the two outermost walls of *Glomus* 3638 are lacking and the dimensions of the attachment hypha are greater.
    Glomus 3638 had its highest SFA value (25.3) in the bluegrass area.
15. *Scutellospora calospora* (Nicol. and Gerd.) Walker and Sanders
    Importance: moderate (bluegrass) to major (bentgrasses)
    Spores of this species were especially numerous in the bentgrass plots. Spores occurred in 72.1% of the velvet bentgrass samples and in 85.7% of the creeping bentgrass samples. Overall, its spores were the second most abundant in the three plots, and it began sporulating earlier in the year than did other species.
16. *Scutellospora erythropa* (Koske and Walker) Walker and Sanders
    Importance: minor
    Spores of *S. erythropa* were isolated in low abundance and frequency from all three study areas.
17. *Scutellospora pellucida* (Nicol. and Schenck) Walker and Sanders
    Importance: minor (bentgrasses) to moderate (bluegrass)
    Spores of *S. pellucida* were more numerous and frequent in the bluegrass plot than in the bentgrass plots, but the spores of this species were of low abundance in all plots.
18. *Scutellospora reticulata* (Koske, Miller and Walker) Walker and Sanders
    Importance: none (bluegrass and creeping bentgrass) to minor (velvet bentgrass)
    The characteristic spores of *S. reticulata* were isolated only three times from the velvet bentgrass site.

Spores of 17 recognizable species (i.e., unidentified *Glomus* spp. not included) of AMF were isolated from the velvet bentgrass plot, 15 species from the creeping bentgrass plot, and 14 from the annual bluegrass plot (Table 2). Average spore abundance was greatest in the creeping bentgrass samples (191.0 spores/100 mL), next in the velvet bentgrass samples (82.4 spores/100 mL), and least in the bluegrass samples (28.4 spores/100 mL). Differences were statistically significant.

Spores were recovered from a significantly greater percentage of the bentgrass samples (88.5–96.8%) than from the bluegrass samples (76.6%) (Table 2).

Average monthly richness (unidentified *Glomus* spp. not included) was significantly higher in the creeping bentgrass samples (4.5 species) than in the bluegrass samples (2.0), but the average richness of velvet bentgrass samples (3.3) did not differ significantly from either of the other grasses. There was a strong seasonality to richness (see seasonal variation section below).

Each plot differed in the SFA indices of AMF species, and three unique AMF communities were distinguishable (Fig. 8). The AMF spore population from velvet bentgrass plot was dominated by *A.* 3106, *Glomus occultum*, and *S. calospora*, whereas samples from the creeping bentgrass plot were characterized by spores of *A.* 3106, *S. calospora*, and *A. mellea*. The bluegrass samples showed an AMF spore community dominated by *Glomus occultum*, *Glomus* 3633, *S. calospora*, and *Glomus* 3638. The dominant species on the basis of SFA index calculated for all three turf species were *A.* 3106 (SFA = 57.3), *S. calospora* (36.9), *Glomus occultum* (24.0), and *A. mellea* (18.2).

Similarity of the populations was greatest for the velvet – creeping bentgrass comparison (S = 0.45), less for velvet bentgrass – bluegrass (S = 0.23), and least for creeping bentgrass – bluegrass (S = 0.12).

Table 2. Frequency of occurrence and abundance of spores of AMF isolated from field plots of three turf species sampled between May 1990 and July 1991.

Fungi	Frequency of occurrence[a]				Spores/100 mL[b]			
	Creeping bentgrass	Velvet bentgrass	Annual bluegrass	All plots	Creeping bentgrass	Velvet bentgrass	Annual bluegrass	All plots
Acaulospora gerdemanni	0*a*, *z*	0*a*, *z*	10.9*b*, *wxy*	3.7*xyz*	0*b*, *z*	0*b*, *z*	3.8*a*, *x*	1.3*xyz*
A. mellea	55.6*c*, *u*	34.4*a*, *uv*	1.6*b*, *yz*	30.2*tu*	30.0*a*, *u*	5.4*ab*, *y*	0.1*b*, *z*	8.8*v*
A. paulinae	0*a*, *z*	3.3*a*, *yz*	0*a*, *z*	1.1*z*	0*a*, *z*	0.2*a*, *z*	0*a*, *z*	0.1*z*
A. 3106	60.3*c*, *u*	32.8*a*, *uv*	4.7*b*, *wxyz*	32.3*tu*	109.7*a*, *s*	32.8*b*, *x*	0.2*c*, *yz*	47.2*u*
A. 3556	9.5*b*, *yz*	4.9*a*, *yz*	0*ab*, *z*	4.8*xyz*	2.6*a*, *xyz*	0.6*ab*, *z*	0*b*, *z*	1.0*yz*
Entrophospora infrequens	44.4*a*, *uv*	29.5*a*, *uvw*	1.6*b*, *yz*	24.9*uvw*	4.0*a*, *vw*	1.9*a*, *yz*	0.1*b*, *z*	2.0*wxy*
Gigaspora gigantea	3.1*a*, *z*	3.3*a*, *yz*	0*a*, *z*	2.1*z*	0.2*a*, *z*	0.7*a*, *yz*	0*a*, *z*	0.3*yz*
Glomus etunicatum	57.1*c*, *u*	14.8*a*, *wxy*	14.1*b*, *vwx*	28.6*tuv*	8.1*a*, *uv*	0.6*b*, *yz*	1.5*b*, *xy*	3.4*vw*
Glomus leptotichum	9.5*a*, *yz*	3.3*a*, *yz*	7.8*a*, *wxyz*	6.9*yz*	0.8*a*, *xyz*	0.1*a*, *z*	0.6*a*, *yz*	0.5*yz*
Glomus microaggregatum	9.5*a*, *yz*	6.6*a*, *yz*	15.6*a*, *vw*	10.6*xyz*	0.3*a*, *yz*	0.2*a*, *z*	0.5*a*, *yz*	0.3*yz*
Glomus mosseae	1.6*a*, *z*	3.3*a*, *yz*	3.1*a*, *xyz*	2.6*yz*	0.1*a*, *z*	0.1*a*, *z*	0.1*a*, *z*	0.1*z*
Glomus occultum	34.9*a*, *vw*	45.9*a*, *u*	28.1*a*, *uv*	40.0*t*	3.2*b*, *wx*	23.3*a*, *x*	8.5*b*, *x*	11.5*v*
Glomus 3633	17.5*a*, *xy*	14.8*a*, *wxy*	28.1*a*, *uv*	20.1*vw*	1.1*b*, *xyz*	0.5*b*, *z*	4.8*a*, *x*	2.2*vwx*
Glomus 3638	19.0*ab*, *wxy*	9.8*a*, *xyz*	26.7*b*, *uv*	18.5*w*	1.7*ab*, *xyz*	0.6*b*, *z*	3.5*a*, *x*	2.0*wxy*
Glomus spp.	30.2*a*, *vwx*	21.3*a*, *vwx*	7.8*a*, *wxyz*	19.6*vw*	3.0*a*, *wxy*	2.2*a*, *yz*	0.7*a*, *yz*	1.9*wxy*
Scutellospora calospora	85.7*a*, *t*	72.1*a*, *t*	37.5*b*, *u*	64.6*s*	32.6*a*, *t*	12.6*b*, *x*	2.7*c*, *x*	16.7*u*
S. erythropa	3.1*a*, *z*	3.3*a*, *yz*	1.6*a*, *z*	2.6*yz*	0.2*a*, *z*	0.2*a*, *z*	0.1*a*, *z*	0.1*yz*
S. pellucida	3.1*b*, *z*	8.2*a*, *xyz*	15.6*ab*, *vw*	9.0*x*	0.1*b*, *z*	0.4*b*, *z*	1.1*a*, *yz*	0.5*yz*
S. reticulata	0*a*, *z*	4.9*a*, *yz*	0*a*, *z*	1.9*z*	0*a*, *z*	0.1*a*, *z*	0*a*, *z*	0.1*z*
All species of AMF	96.8*a*[c]	88.5*a*	76.6*b*	86.8	191.0*a*	82.4*b*	28.4*c*	99.9

[a]Percentage of samples collected over the 15-month sampling period in which spores of a species were present. Total number of samples collected: creeping bentgrass, 63; velvet bentgrass, 61; annual bluegrass, 64. Values in rows followed by a different letter (*a*, *b*, *c*, etc.) differ significantly ($p < 0.05$). Letters at the end of the alphabet (e.g., *z*, *y*, *x*, *w*, etc.) are used to indicate significant differences between values in each column ($p < 0.05$).

[b]Average number of spores/100 mL recovered during the duration of the study.

[c]Percentage of samples in which spores of any species of AMF were recovered.

Seasonal variation

Seasonal spore abundance in each plot (all AMF species combined) displayed significant polynomial trends, and two different patterns were evident (Fig. 9). In the bentgrass plots, abundance increased from spring through November and then declined. The pattern with bluegrass was generally similar, but showed a marked decline in July and August 1990, with a recovery in September through December. No live spores of any species of AMF were recovered from the August samples of bluegrass.

The seasonal variation in AMF species richness (average number of species per monthly sample) of the three plots (Fig. 9) paralleled the trends in spore abundance. Average AMF species richness in the creeping bentgrass and bluegrass plots displayed significant seasonal polynomial trends ($p = 0.03$ and $p = 0.05$, respectively), but not for AMF from the velvet bentgrass plot. Average richness and spore abundance were positively and significantly correlated for creeping bentgrass ($r = 0.77$, $p = 0.001$) and for bluegrass ($r = 0.68$, $p = 0.006$), but not for velvet bentgrass ($r = 0.45$, $p = 0.07$).

Spores of only seven species were recovered at a great enough frequency and abundance from at least one plot for patterns of seasonality of individual species to be assessed with confidence over the study period, and these data are illustrated in Fig. 10. Monthly spore abundance for most species followed the pattern illustrated in Fig. 9 for combined species abundance.

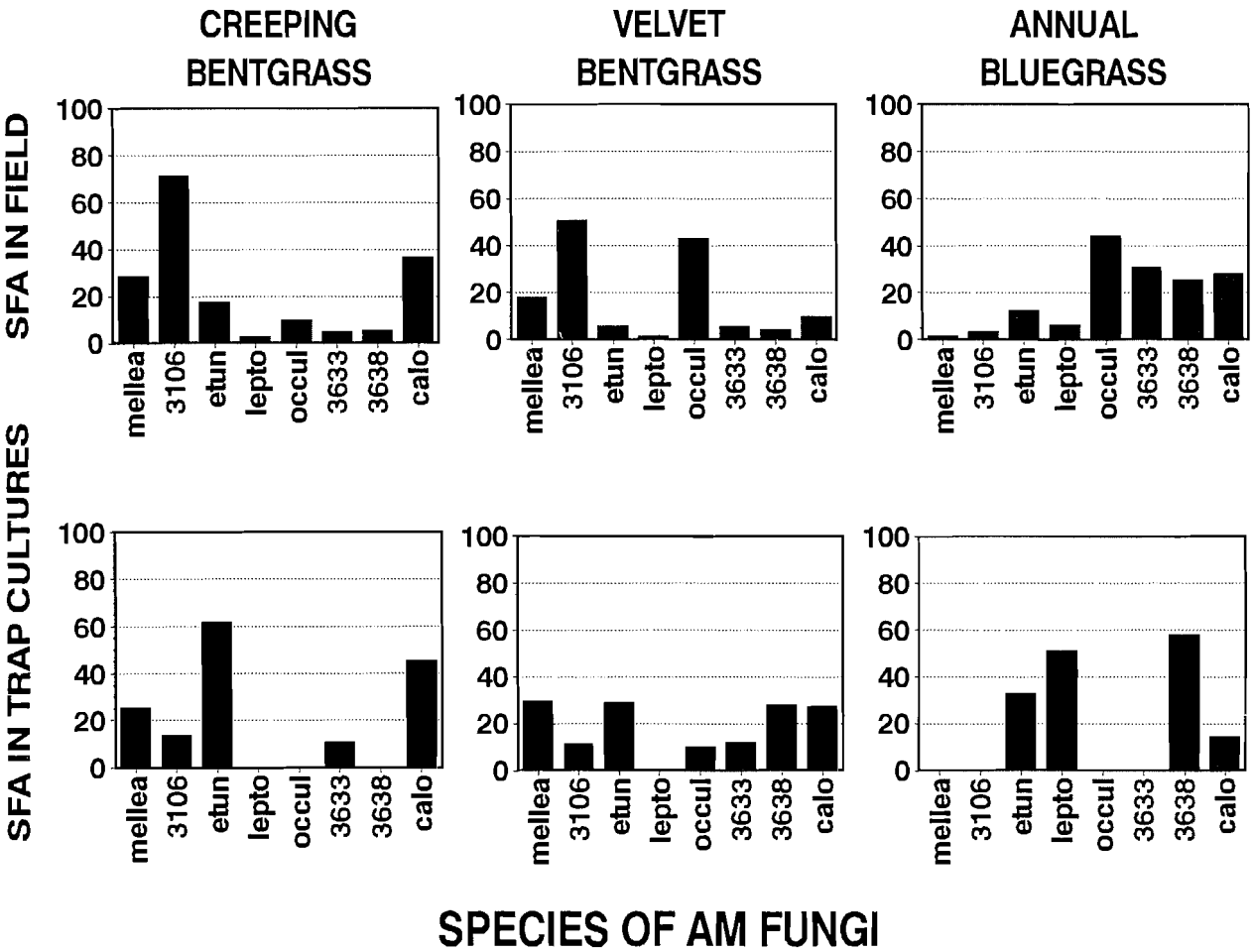
Spores of some species displayed apparent interruptions in their presence in the turf sites that did not appear to be part of any seasonal trend (Fig. 10). For example, spores of *Glomus* 3633 were not recovered in the February 1991 sampling from the bluegrass plot, although they were present in the previous and following sampling times. These gaps in recovery typically were noted for those species whose spores were not present in abundance.

Trap cultures

Spores of 10 species of AMF were recovered from trap cultures at an abundance of more than 5 spores/100 mL (Table 3). Species sporulating well in the trap cultures included *Glomus etunicatum*, *Glomus leptotichum*, *Glomus* 3638, *S. calospora*, and *A. mellea*. Different SFA indices were evident for AMF isolated from the three plots (Fig. 8).

The monthly variation in spore abundance in the trap cultures was very large, and standard deviation values frequently exceeded means. Only a single species (*Glomus leptotichum*) produced significantly more spores in trap cultures started from a particular site (the bluegrass plot) (Table 3). The monthly average abundance of spores of *Glomus etunicatum* present in trap cultures from the bluegrass plot illustrates the amount of variation that was routinely encountered. Spore

Fig. 8. Spore frequency and abundance (SFA) indices of the major species of AMF isolated from the field and recovered from trap cultures over the sampling period. Only species with a SFA greater than 20 in association with at least one host or treatment (field or trap culture) were included. Note similar profiles in the creeping and velvet bentgrass field samples and great differences between field samples and trap samples. mellea, *A. mellea*; 3106, *A.* 3106; etun, *Glomus etunicatum*; lepto, *Glomus leptotichum*; occul, *Glomus occultum*; 3633, *G.* 3633; 3638, *G.* 3638; calo, *S. calospora*.



abundance for the 10 months of samples (May 1990 – May 1991) were 560.0, 0.0, 0.0, 0.0, 0.0, 286.0, 0.0, 0.0, 0.0, and 0.0 spores/100 mL.

Species richness in the trap cultures also was very low. Monthly richness ranged from 0–4 and averaged 1.3 (creeping bentgrass), 1.2 (velvet bentgrass), and 1.6 (bluegrass).

Comparison of trap cultures and field collections

Over the 13-month trapping period, the spore communities of AMF species developing in the trap cultures differed greatly from the communities recorded from the field collections (Fig. 8; Tables 2, 3). For example, *A.* 3106 and *Glomus occultum*, whose spores were numerous and frequently isolated from field collections, sporulated poorly in the trap cultures, occurring there only in December 1990. The opposite situation also occurred. In the bluegrass trap cultures, *Glomus leptotichum* ranked second in SFA index, but it ranked ninth in the field collections.

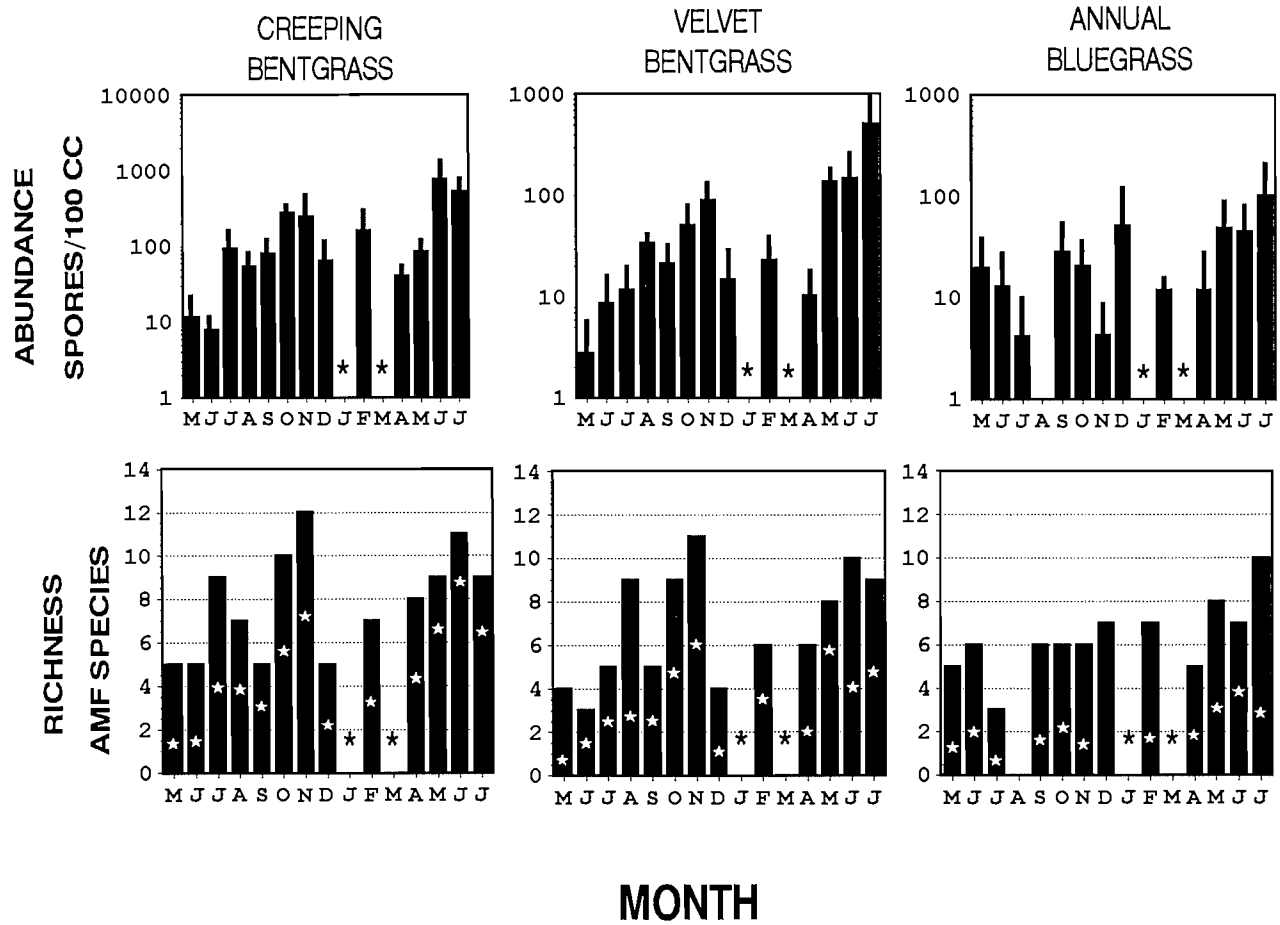
Direct monthly comparisons of the relative abundance of spores of AMF isolated from the field with those recovered from trap cultures generally revealed little similarity (Table 4). For one date and host species (December 1990 for creeping bentgrass), nearly identical results ($S = 0.87$) were obtained, but, estimates of similarity were typically very low, and the average similarity for all comparisons was 0.14.

Discussion

Spores of a variety of AMF were abundant in the root zones of the three turfgrasses. The average species richness and spore abundance of the study plots were comparable to values reported from grasses growing in natural habitats (e.g., Molina et al. 1978; Sylvia 1986; Koske 1987; Brundrett 1991). The management practices in the bentgrass plots appeared to encourage sporulation by a diverse and extensive AMF community, as the unmaintained bluegrass area had fewest spores and lowest richness. The relatively high similarity between the AMF communities of the bentgrasses and the low similarity between the bentgrasses and the bluegrass samples probably is more a reflection of similarities or differences of soil conditions (pH, nutrients, and watering) than of host effects on sporulation and AMF diversity (Koske 1981). The

Fig. 10. Monthly abundance of spores per 100 mL of the major species of AMF whose spores were isolated from field plots of creeping bentgrass, velvet bentgrass and annual bluegrass from May 1990 until July 1991. Note expanded *Y*-axis for *A. mellea*, *A.* 3106, and *Glomus occultum*. Asterisks indicate months when no samples were collected (January and March 1991). Significant polynomial trends were demonstrated for the following combinations: *A. mellea* – creeping bentgrass, $\ln(Y + 1) = -9.06 + 2.517X - 0.131X^2 + 0.002X^3$, $p = 0.04$; *A.* 3106 – creeping bentgrass, $\ln(Y + 1) = -16.685 + 4.835X - 0.379X^2 + 0.010X^3$, $p = 0.03$); *A.* 3106 – velvet bentgrass, $\ln(Y + 1) = 15.520 - 7.841X + 1.344X^2 - 0.090X^3 + 0.002X^4$, $p = 0.0008$; *Glomus occultum* – creeping bentgrass, $\ln(Y + 1) = -1.675 + 0.335X - 0.045X^2 - 0.002X^3$, $p = 0.04$; *Glomus occultum* – velvet bentgrass, $\ln(Y + 1) = -5.529 + 1.008X - 0.167X^2 + 0.009X^3$, $p = 0.03$; *S. calospora* – creeping bentgrass, $\ln(Y + 1) = -4.343 + 1.191X - 0.200X^2 + 0.009X^3$, $p = 0.01$; *S. calospora* – velvet bentgrass, $\ln(Y + 1) = 0.676 - 0.161X + 0.298X^2 - 0.048X^3 + 0.002X^4$, $p = 0.0008$. See caption for Fig. 9 for explanation.

Fig. 9. Average yearly spore abundance (spores/100 mL) and total monthly species richness (bars) and average monthly richness (white stars) of AMF isolated from field plots of creeping bentgrass, velvet bentgrass and annual bluegrass from May 1990 until July 1991. Spore abundance and average richness were significantly correlated in the creeping bentgrass and bluegrass plots. Asterisks indicates months when no samples were collected (January and March 1991). Spore abundance showed a significant polynomial trend with each host species: creeping bentgrass ($\ln(Y + 1) = -1469.700 + 467.501X - 43.333X^2 + 1.278X^3$, $p = 0.03$); velvet bentgrass ($\ln(Y + 1) = 1404 - 641.269X + 102.692X^2 - 6.781X^3 + 0.158X^4$, $p = 0.0001$); bluegrass ($\ln(Y + 1) = 532.238 - 217.663X + 33.738X^2 - 1.925X^3 + 0.042X^4$, $p = 0.007$), where Y is spores/100 mL and X is month (for May 1990, $X = 5$, for May 1991, $X = 17$). Average AMF species richness displayed significant seasonal trends with the bentgrasses, but total monthly richness did not (see text).



reduced community of AMF spores in the bluegrass plot apparently resulted more from these edaphic factors (e.g., lower pH and soil P, differences in other nutrients, lack of summer irrigation) than from a host effect, but the relative importance of each factor could not be determined by this study. The P levels in the bentgrass plots had been kept relatively low for over 20 years in comparison to levels in more highly maintained greens (Turgeon 1980; Hind et al. 1995), but they were three to five times greater than in the bluegrass area. At much higher levels of soil P, an attenuation of AMF spore populations would be expected (e.g., Mårtensson and Calgren 1994).

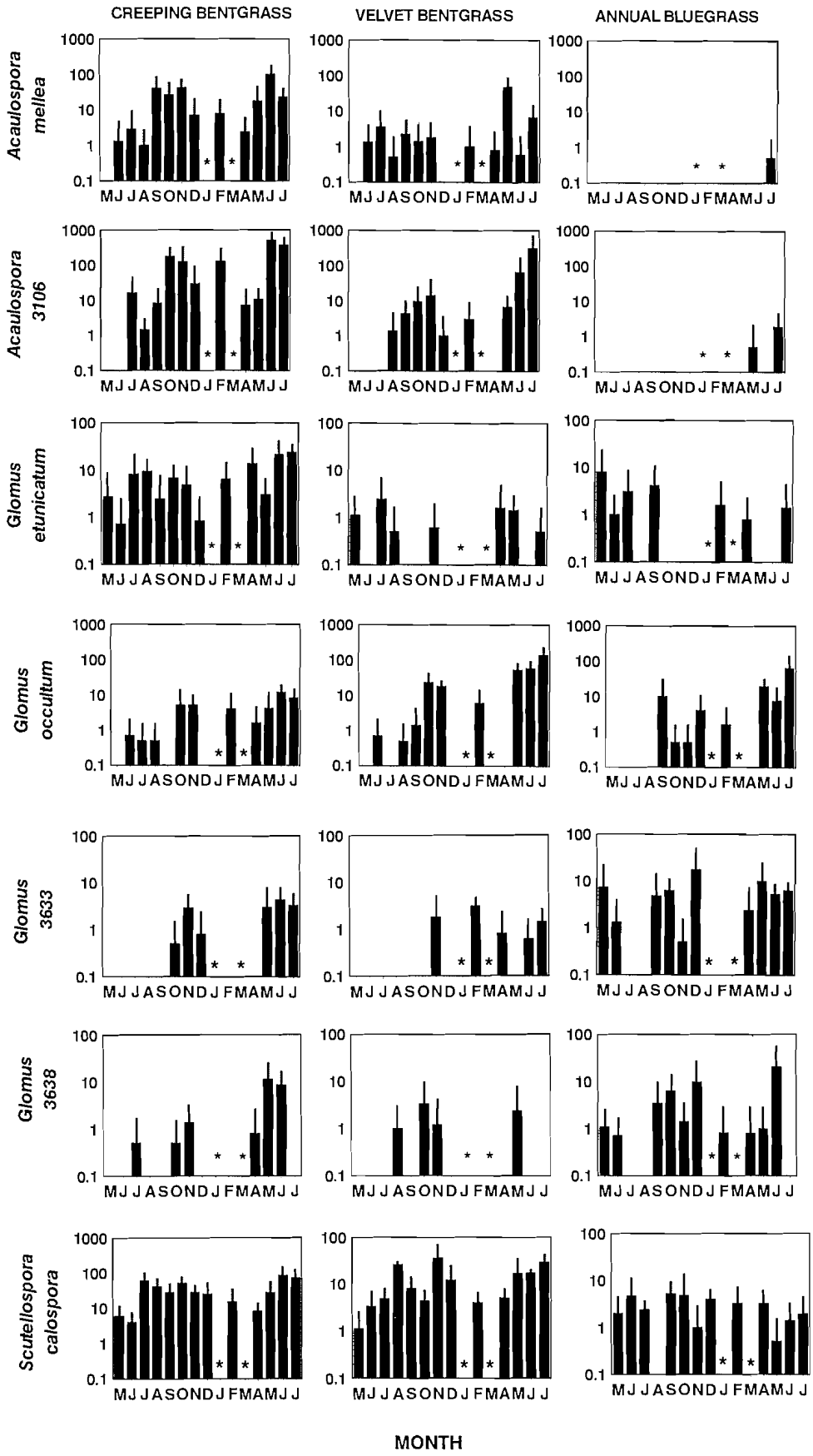


Table. 3. Frequency and abundance of spores of AMF isolated from trap cultures using soil samples from three turf species sampled for 10 months between May 1990 and May 1991.

AMF species	Frequency of occurrence[a]			Spores/100 mL[b]		
	Creeping bentgrass	Velvet bentgrass	Annual bluegrass	Creeping bentgrass	Velvet bentgrass	Annual bluegrass
Acaulospora mellea	20*a*	20*a*	0*a*	55.6*x* ±171.6	50.0*x* ±149.9	0.0*x* ±0.0
Acaulospora 3106	10*a*	10*a*	0*a*	10.8*x* ±34.2	2.9*x* ±9.3	0.0*x* ±0.0
Glomus etunicatum	40*a*	20*a*	20*a*	15.5*x* ±26.6	22.4*x* ±47.4	84.6*x* ±190.0
Glomus leptotichum	0*a*	0*a*	40*a*	0.0*y* ±0.0	0.0*y* ±0.0	81.4*x* ±155.8
Glomus occultum	0*a*	10*a*	0*a*	0.0*x* ±0.0	2.5*x* ±6.2	0.0*x* ±0.0
Glomus 3633	10*a*	10*a*	0*a*	1.1*x* ±3.5	6.8*x* ±21.5	0.0*x* ±0.0
Glomus 3638	0*a*	20*a*	50*a*	0.0*x* ±0.0	726.2*x* ±2254.9	289.5*x* ±618.5
Entrophospora infrequens	0*a*	10*a*	10*a*	0.0*x* ±0.0	2.7*x* ±8.6	2.0*x* ±6.3
Scutellospora calospora	50*a*	20*a*	20*a*	10.8*x* ±18.1	8.8*x* ±22.8	16.1*x* ±35.3
Scutellospora pellucida	0*a*	0*a*	10*a*	0.0*x* ±0.0	0.0*x* ±0.0	80.0*x* ±253.1

[a]Percentage of months over the 10-month sampling period (May 1990 – May 1991) in which spores of this species were recovered from trap cultures. Values in rows followed by a different letter were significantly different ($p < 0.05$).

[b]Average number of spores/100 mL ± SD recovered from trap cultures during the duration of the study. Values in rows followed by a different letter were significantly different ($p < 0.05$).

Table 4. Comparison of AMF populations estimated by direct counts of spores from the field and counts from trap cultures. A modified Sorenson's coefficient of similarity was used to compare results from 10 months of sampling. Relative abundance was used for the calculations.

Collection date	Coefficient of similarity		
	Velvet bentgrass	Creeping bentgrass	Annual bluegrass
5/90	0.00	0.00	0.00
6/90	0.00	0.00	0.00
7/90	0.00	0.39	0.00
8/90	0.00	0.27	0.00
9/90	0.00	0.00	0.29
10/90	0.00	0.03	0.07
11/90	0.40	0.22	0.00
12/90	0.71	0.87	0.53
4/91	0.00	0.00	0.00
5/91	0.39	0.00	0.00
Mean	0.15	0.18	0.11

The only other survey of AMF associated with bentgrass that we found concerned creeping bentgrass in a putting green in Ohio. Spores of three species of fungi (*Glomus macrocarpum* Tul. and Tul., *Glomus microcarpum* Tul. and Tul., and *Glomus tenuis* (Greenhall) Hall) were isolated (Rhodes and Larsen 1981). None of these species was recovered from the Rhode Island plots. Earlier, Herskowitz and Estey (1978) found spores of *Glomus geosporum* (Nicol. and Gerd.) Walker and *Glomus mosseae* (Nicol. and Gerd.) Gerd. and Trappe in an unidentified turf in Quebec.

Seasonal patterns in AMF spore abundance result from increases in populations due to sporulation and decreases caused by parasitism, predation, and germination (e.g., Gemma et al. 1989; Lee and Koske 1994*a*). Climatic conditions may affect abundance by their influence directly on the AMF (sporulation, germination, and aging), and indirectly via the host plants (export of photosynthate to roots, root growth, and physiology), or through the activity of soil organisms associated with loss of spore viability (Harley and Smith 1983; Lee and Koske 1994*a*, 1994*b*).

Most species of AMF in our study showed a steady increase in spore abundance (sporulation) throughout the growing season (ca. May–November). Spore numbers declined from December until the beginning of the next growing season. The warmer, drier spring of 1991 correlated with an earlier onset of sporulation by most species than in the previous year. All three grasses are cool-season species, showing active growth of roots and shoots in spring and fall, and may become dormant in the summer in response to high temperatures, especially when not watered (Turgeon 1980).

In the bluegrass plot, spore abundance and species richness displayed precipitous declines in July and August 1990,

but not in July 1991. The bluegrass plot was close enough to the creeping bentgrass plot that it may have been watered accidentally more in 1991 than in 1990, but this was not measured. As noted above, however, decreases in spore abundance can result from a variety of factors, and the reason for the decline in the summer of 1990 is unknown.

Seasonal trends in AMF spore populations in agricultural and natural areas are well described in the literature (e.g., Sutton and Barron 1972; Giovannetti 1985; Gemma and Koske 1988; Gemma et al. 1989; An et al. 1993; Guo et al. 1993), and the results from our study generally match those of other studies. A study of AMF associated with the cool-season grass *Festuca arundinacea* Schreb. in Kentucky, U.S.A., identified 15 species (An et al. 1993), two of which (*Gigaspora gigantea* and *Glomus mosseae*) occurred in our turf sites. Most of the species displayed a strong seasonality, with minimal spore populations between spring and mid-summer and increasing abundance from August to October. This pattern was more similar to that occurring in the unmaintained bluegrass plot in our study than to the AMF populations in the irrigated and fertilized bentgrass plots.

In the present study, the occasional absence of spores of some species of AMF from the monthly collections (e.g., *Glomus occultum*, *Glomus* 3633) probably was the result of sampling small populations with aggregated distributions (Southwood 1978). Such variation is common in AMF spore count data, especially for species that are not dominants (Tews and Koske 1986). Collection of more samples per month probably would have smoothed out the monthly abundance charts.

Comparison of data from the trap cultures with field collections confirms observations by other authors of the different results that can be given by the two techniques (e.g., Liberta et al. 1983; Bever et al. 1996). Spores of fewer species were recovered from the trap cultures than from the field, and estimates of species richness were 1.25–3.46 times less when trap culture data were used. The relatively uniform conditions of trap culturing are more likely to favor sporulation by fewer species than does the variation in climatic and edaphic conditions that occur in the field in the course of one year. The near absence in trap cultures of species that were abundant in the field confirms the selective nature of pot culturing. However, neither method has been shown to accurately assess the biological importance of particular AMF species, and the calculation of SFA indices based on spore abundance and frequency can be only an estimate of activity.

The demonstration of a diverse AMF community in bentgrass turf and the results from recent experiments showing increased growth and improved drought tolerance in turf inoculated with AMF (C. Charest and Y. Dalpé, personal communication; J.N. Gemma et al., in preparation) suggest that highly maintained turf may benefit more from AMF than previously has been thought.

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References

An, Z.Q., Guo, B.Z., and Hendrix, J.W. 1993. Populations of spores and propagules of mycorrhizal fungi in relation to the life cycles of tall fescue and tobacco. Soil Biol. Biochem. **25**: 813–817.

Baylis, G.T.S. 1975. The magnolioid mycorrhiza and mycotrophy in root systems derived from it. *In* Endomycorrhizas. *Edited by* F.E. Sanders, B. Mosse, and P.B. Tinker. Academic Press, London, U.K. pp. 373–389.

Bever, J.D., Morton, J.B., Antonovics, J., and Schultz, P.A. 1996. Host-dependent sporulation and species diversity of arbuscular mycorrhizal fungi in a mown grassland. J. Ecol. **84**: 71–82.

Blaszkowski, J. 1994. Arbuscular fungi and mycorrhizae (Glomales) of the Hel Peninsula, Poland. Mycorrhiza, **5**: 71–88.

Bray, J.R., and Curtis, J.T. 1957. An ordination of the upland forest communities of southern Wisconsin. Ecol. Monogr. **27**: 325–349.

Brundrett, M. 1991. Mycorrhizas in natural ecosystems. Adv. Ecol. Res. **21**: 171–313.

Epstein, E. 1972. Mineral nutrition of plants: principles and perspectives. John Wiley & Sons Inc., New York.

Gemma, J.N., and Koske, R.E. 1988. Seasonal variation in spore abundance and dormancy of *Gigaspora gigantea* and in mycorrhizal inoculum potential of a dune soil. Mycologia, **80**: 211–216.

Gemma, J.N., Koske, R.E., and Carreiro, M.M. 1989. Seasonal dynamics of five species of VA fungi in a sand dune. Mycol. Res. **92**: 27–32.

Giovannetti, M. 1985. Seasonal variation of vesicular–arbuscular mycorrhizas and endogonaceous spores in a maritime sand dune. Trans. Brit. Mycol. Soc. **84**: 679–684.

Guo, B.Z., An, Z.-Q., Hendrix, J.W., and Dougherty, C.T. 1993. Influence of a change from tall fescue to pearl millet or crabgrass on the mycorrhizal fungal community. Soil Sci. **155**: 398–405.

Hall, I.R. 1984. Some Endogonaceae from south western Australia. Trans. Brit. Mycol. Soc. **83**: 203–208.

Harley, J.L., and Smith, S.E. 1983. Mycorrhizal symbiosis. Academic Press, London.

Hayman, D.S. 1983. The physiology of vesicular–arbuscular endomycorrhizal symbiosis. Can. J. Bot. **61**: 944–963.

Herskowitz, J.M., and Estey, R.H. 1978. Endogonaceae from Quebec soils. Can. J. Bot. **56**: 1095–1098.

Hetrick, B.A.D., Kitt, D.G., and Wilson, G.W.T. 1988. Mycorrhizal dependency and growth habit of warm-season and cool-season tallgrass prairie plants. Can. J. Bot. **66**: 1376–1380.

Hetrick, B.A.D., Wilson, G.W.T., and Todd, T.C. 1990. Differential response of C_3 and C_4 grasses to mycorrhizal symbiosis, P fertilization, and soil microorganisms. Can. J. Bot. **68**: 461–467.

Hetrick, B.A.D., Wilson, G.W.T., and Leslie, J.F. 1991. Root architecture of warm- and cool-season grasses: relationship to mycorrhizal dependence. Can. J. Bot. **69**: 112–118.

Hind, P.D., Baker, S.W., Lodge, T.A., Hunt, J.A., and Binns, D.J. 1995. A survey of golf greens in Great Britain. I. Soil properties. J. Sports Turf Res. Inst. **71**: 9–22.

Hoagland, D.R., and Arnon, D.I. 1950. The water culture method of growing plants without soil. Calif. Agric. Exp. Stn. Circ. No. 347.

Koske, R.E. 1981. A preliminary study of interactions between species of vesicular–arbuscular mycorrhizal fungi in a sand dune. Trans. Brit. Mycol. Soc. **76**: 411–416.

Koske, R.E. 1987. Distribution of VA mycorrhizal fungi along a latitudinal temperature gradient. Mycologia, **79**: 55–68.

Koske, R.E., and Tessier, B. 1983. A convenient, permanent slide mounting medium. Mycol. Soc. Am. Newsl. **34**(2): 59.

Koske, R.E., Gemma, J.N., and Olexia, P.D. 1986. *Glomus microaggregatum*, a new species in the Endogonaceae. Mycotaxon, **26**: 125–132.

Lee, P.-J., and Koske, R.E. 1994*a*. *Gigaspora gigantea*: seasonal abundance and aging of spores in a sand dune. Mycol. Res. **98**: 453–457.

Lee, P.-J., and Koske, R.E. 1994*b*. Biology of *Gigaspora gigantea*: parasitism of spores by fungi and actinomycetes. Mycol. Res. **98**: 458–466.

Liberta, A.E., Anderson, R.C., and Dickman, L.A. 1983. Vesicular–arbuscular mycorrhiza fragments as a means of endophyte identification at hydrophytic sites. Mycologia, **75**: 169–171.

Mårtensson, A.M., and Calgren, K. 1994. Impact of phosphorus fertilization on VAM diaspores in two Swedish long-term field experiment. Agric. Ecosys. Environ. **47**: 27–334.

Miller, R.M., Jarstfer, A.G., and Pillai, J.K. 1987. Biomass allocation in an *Agropyron smithii–Glomus* symbiosis. Am. J. Bot. **74**: 114–122.

Molina, R.J., Trappe, J.M., and Strickler, G.S. 1978. Mycorrhizal fungi associated with *Festuca* in the western United States and Canada. Can. J. Bot. **56**: 1691–1695.

Morton, J.B. 1986. Three new species of *Acaulospora* (Endogonaceae) from high-aluminum, low-pH soils in West Virginia. Mycologia, **78**: 641–648.

Morton, J.B. 1994. A problem group of five species in *Acaulospora*. International Collection of VA Mycorrhizal Fungi Newsl. **4**(2): 8.

Mueller-Dombois, D., and Ellenberg, H. 1974. Aims and methods of vegetation ecology. John Wiley & Sons Inc., New York.

Newman, E.I., and Reddel, P. 1987. The distribution of mycorrhizas among families of vascular plants. New Phytol. **106**: 745–751.

Petrovic, A.M. 1984. Endomycorrhizal fungi: friend or foe. Proceedings of the 53rd Annual Northeastern Turfgrass Conference, March 5–6, 1984, Springfield, Mass. University of Massachusetts, Amherst, Mass. pp. 18–20.

Rhodes, L.H., and Larsen, P.O. 1981. Effects of fungicides on mycorrhizal development of creeping bentgrass. Plant Dis. **65**: 145–147.

Smiley, R.W., Dernoeden, P.H., and Clarke, B.B. 1992. Compendium of turfgrass diseases. 2nd ed. APS Press, St. Paul, Minn.

Southwood, T.R.E. 1978. Ecological methods with particular reference to the study of insect populations. Chapman and Hall, London, U.K.

St. John, T.V., and Koske, R.E. 1988. Statistical treatment of endogonaceous spore counts. Trans. Brit. Mycol. Soc. **91**: 117–121.

Sutton, J.C., and Barron, G.L. 1972. Population dynamics of *Endogone* spores in soil. Can. J. Bot. **50**: 1909–1914.

Sylvia, D.M. 1986. Spatial and temporal distribution of vesicular–arbuscular mycorrhizal fungi associated with *Uniola paniculata* in Florida foredunes. Mycologia, **78**: 728–734.

Tews, L.L., and Koske, R.E. 1986. Toward a sampling strategy for vesicular–arbuscular mycorrhizas. Trans. Brit. Mycol. Soc. **87**: 353–358.

Trappe, J.M. 1977. Three new Endogonaceae: *Glomus constrictum*, *Sclerocystis clavispora*, and *Acaulospora scrobiculata*. Mycotaxon, **6**: 359–366.

Trappe, J.M. 1987. Phylogenetic and ecologic aspects of mycotrophy in the angiosperms from an evolutionary standpoint. *In* Ecophysiology of VA mycorrhizal plants. *Edited by* G.R. Safir. CRC Press, Boca Raton, Fla. pp. 5–26.

Turgeon, A.J. 1980. Turfgrass management. Reston Publishing Co., Reston, Va.

Walker, C. 1983. Taxonomic concepts in the Endogonaceae. I. Spore wall characteristics in species descriptions. Mycotaxon, **18**: 443–455.

Walker, C. 1986. Taxonomic concepts in the Endogonaceae. II. A fifth morphological wall type in endogonaceous spores. Mycotaxon, **25**: 95–99.

Walker, C., Mize, C.W., and McNabb, H.S. 1982. Populations of endogonaceous fungi at two locations in central Iowa. Can. J. Bot. **60**: 2518–2529.