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ORIGINAL PAPER

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# Ectomycorrhizas associated with a relict population of *Dryas octopetala* in the Burren, western Ireland II. Composition, structure and temporal variation in the ectomycorrhizal community

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**Abstract** The composition, structure and temporal variation of ectomycorrhizal (EM) communities associated with mountain avens (*Dryas octopetala*) in grass heaths of the Burren, western Ireland were assessed by using soil core sampling in two permanent plots and 30 other sites (196 cores in total). Of the 34 different EM types observed, 11 were common and constituted over 80% of the EM biomass. Four EM types, *Craterellus lutescens*, *Tomentella* sp., *Dryadirhiza fulgens* and *Cenococcum geophilum* were the most abundant as measured by EM length and frequency of occurrence in cores. The species profile and relative abundances were very similar in cores from the permanent plots and different sites in the Burren, indicating that they were all representative of the same EM community. The below-ground EM community in both plots was compared with production of basidiomes, and the latter was found to be an unreliable indicator of EM community structure. Temporal variation in the EM community was assessed by repeated core sampling of the two permanent plots over a 14-month period (between March 1998 and May 1999). No statistically significant shifts in EM abundance were found between sampling dates, probably as a consequence of the large variation in EM abundance between core samples over the sampling period. No significant relationship was found between rainfall, soil moisture or soil temperature and fluctuations in EM abundance. Patterns of total EM abundance and fluctuations in EM diversity were strongly correlated between the two permanent plots over the sampling period. Temporal fluctuations in the dominant EM type, *Craterellus lutescens*, were similar in both plots with respect to mycorrhizal length, biomass and relative abundance, and the patterns between both plots were positively correlated. EM diversity was negatively correlated with biomass of ectomycorrhizas of *Craterellus lutescens* in both plots, but it was significant only in plot 1.

**Keywords** Ectomycorrhizal community · Basidiomes · Temporal variation · *Craterellus*

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## Introduction

Relict populations of the arctic-alpine mountain avens, *Dryas octopetala* L., occur sporadically in lowland areas of western Europe, where they have survived postglacial climate warming. The largest of these is found in western Ireland in the 450 km$^2$ karst region known as the Burren. Much of the Burren is covered by treeless grass heaths on shallow soils over limestone. The vegetation is phytogeographically unusual as it includes arctic-alpine, temperate, and Mediterranean elements growing together in a mild oceanic climate. *Dryas octopetala* is the most conspicuous of the arctic-alpines, and dominates large areas of the grass heaths, especially on higher ground.

Twenty-one ectomycorrhizal (EM) types, including the most common types, have been described on *Dryas* roots in the Burren, and some of these produce abundant basidiomes, which have facilitated their identification by molecular and other methods (Harrington and Mitchell 2002). Thirty-nine species of putative EM fungi have been recorded as basidiomes in the Burren grass heaths (Harrington 1996, 2003). *Cortinarius* was the most diverse EM genus (17 species) and *Craterellus lutescens* was by far the most abundant in terms of biomass and frequency. On *Dryas* roots however, ectomycorrhizas of *Craterellus lutescens* and an unidentified *Tomentella* species appear to be the most abundant in terms of biomass and frequency [Harrington and Mitchell 2005 (this volume)].

Most investigations of EM community composition and structure have been conducted in forest ecosystems (see

Dahlberg 2001; Horton and Bruns 2001 for reviews). Only a few studies of ectomycorrhizas have been carried out in shrub communities, including *Arctostaphylos uva-ursi* (Mejstrik and Hadac 1975; Largent et al. 1980; Acsai and Largent 1983), *Salix repens* (van der Heijden et al. 1999; van der Heijden and Vosatka 1999), *Salix herbacea* (Graf and Brunner 1996), arctic shrubs (Bledsoe et al. 1990), and dwarf shrub communities at the treeline zone in the Canadian Rockies (Kernaghan and Currah 1998; Kernaghan 2001; Kernaghan and Harper 2001).

Temporal variation in EM populations has been less frequently examined because of the sampling effort required to obtain valid estimates of population changes over time. A number of studies have examined long-term successional changes in EM assemblages on roots that are related to stand age (Mason et al. 1982; Last et al. 1983; Danielson 1991; Deacon and Fleming 1992; Visser 1995) or disturbance (Jonsson et al. 1999; Mahmood et al. 1999), but few have examined short-term temporal variation in mature stable habitats. Seasonal changes in host physiology or the influence of the weather could cause short term changes in the density and composition of EM communities.

The objectives of this study were (1) to establish the composition of the EM community on *D. octopetala* and to obtain a quantitative estimate of the relative contributions of each EM type to community structure; (2) to examine temporal variation in the EM community of *Dryas* and to relate this variation with soil moisture, soil temperature and rainfall patterns; (3) to compare the below-ground structure of the EM community with production of EM basidiomes.

## Materials and methods

### Study area

The composition, structure and temporal variation in the EM community in the grass heaths of the Burren, western Ireland, was assessed by analysis of 196 soil cores collected from *Dryas*-dominated areas between March 1998 and May 1999. Three sets of cores were collected; 70 cores were taken at time intervals from each of two permanent plots (plots 1 and 2) and 56 cores were taken once from 30 other sites in October 1999. The permanent plots (1 and 2) were chosen because they were likely to have ectomycorrhizas in common based on the results of a previous mycocoenological study (Harrington 2003). The repeated coring of the permanent plots was used to assess temporal variation in the EM community, and the total sample of 196 cores was used to assess the community structure and composition of the EM assemblage over a wider area than that represented in the permanent plots alone.

The two permanent plots and 30 other sites were located at altitudes of between 20 and 300 m a.s.l. in the Burren over an area of approximately 100 km$^2$ (Table 1). Plots 1 and 2 were each 10×10 m and located on the eastern (National Grid reference: M349042) and western (M102032) margins of the Burren, respectively, and approximately 25 km distant from each other. The soils in the plots were very shallow and highly organic, while the soils of the 30 sites were more variable in depth and organic matter (Table 1). *Dryas octopetala* was abundant and frequently dominant at all of the areas sampled and was usually accompanied by *Sesleria albicans*, *Calluna vulgaris* and *Carex flacca*. The vegetation of both permanent plots belongs to the Hyperico-Dryadetum association, the principal *Dryas*-dominated plant community in the Burren. The vegetation of the 30 other sites consisted of the Hyperico-Dryadetum (13 sites), the Asperulo-Seslieretum (9 sites), and the Arctostaphylo-Dryadetum (6 sites) [Harrington and Mitchell 2005 (this volume)]. Soil temperature and volumetric soil moisture ($m^3$ $m^{-3}$) were measured at a depth of 10 cm, using a 2-K thermistor-type soil temperature probe and soil moisture sensor (ThetaProbe-type ML1) connected to a Delta-T data logger (Delta-T Devices, Cambridge, UK) located at 200 m a.s.l. altitude on Gleninagh Mt (M175100), about equidistant from plots 1 and 2. Rainfall readings were supplied by the Meteorological Service for Carran (R285992), in the centre of the Burren. Total rainfall, mean daily soil moisture and mean daily soil temperature were calculated for the 2-week period prior to each sampling date.

**Table 1** Characteristics of the two permanent plots and the 30 other sites. Values in parenthesis represent SD

| | Plot 1 | Plot 2 | Sites 1–30 |
|---|---|---|---|
| Altitude (m a.s.l.) | 260 | 50 | 20–300 |
| Soil depth (cm) | 4.8 (3.1) | 3.8 (3.3) | 5.5 (2.2) |
| Organic matter % | 84.6 (4.1) | 85.8 (4.4) | 8.0–92.7 |
| *Dryas* cover % | 79 | 54 | 20–79 |

### EM sampling

Soil cores were collected using a 53-mm internal diameter cylindrical stainless steel corer, which was inserted as far as the bedrock. The samples were collected from the edge of *Dryas* mounds where adventitious roots from the creeping stems could be easily distinguished. Care was taken to ensure that each core encompassed a *Dryas* adventitious root. Assessment of population changes in the EM assemblage in plots 1 and 2 was carried out by soil sampling on seven dates over a 14-month period from March 1998 to May 1999 at 8- to 10-week intervals. A stratified sampling protocol was adopted. For sampling purposes, plots 1 and 2 were divided into nine segments (approx. 3.3×3.3 m) following the cardinal points of the compass, and one central segment. One sample was taken from each of the peripheral segments and two from the central segment; i.e. ten samples were collected on each sampling date. Sampling was nonrandom in the sense that random coordinates were not assigned to each core. The vicinity of previous coring was avoided by at least 0.5 m. The 30 other sites were sampled once in late October 1999 and a soil core was taken from the margins of each of one to two *Dryas* plants (1–3 m apart) at each site. Cores were stored in plastic bags

at 4°C for no longer than 2 weeks before examination of ectomycorrhizas.

### EM enumeration

The volume of each core sample was determined after soaking overnight in tap water [see Harrington and Mitchell 2005 (this volume) for procedures]. The sample was then washed free of soil material over a 400-μm sieve in running tap water. *Dryas* roots and attached and detached mycorrhizas were picked manually from the washed sample. Moribund and dead EM tips were distinguished from live tips by lack of turgidity or shrunken appearance, and dark discolouration of the mantle and cortex. Live tips were then sorted into morphotypes based on general appearance. Representative examples were selected for a more detailed examination that followed the protocols described by Agerer (1986). Identification of some of the main EM types was made by PCR-RFLP and sequence analysis of the ITS region of the rDNA gene (see Harrington and Mitchell 2002 for primers and protocols).

The abundance of individual EM types was determined by four different measures: tip density (number of EM tips per 100 $cm^3$ soil); EM length (centimetres per 100 $cm^3$ soil); EM biomass (milligrams dry weight, per 100 $cm^3$ soil); frequency (percentage of soil cores in which the EM type occurred). The numbers of live mycorrhizal tips of each morphotype were distinguished and counted. The total lengths of live mycorrhizal tips of each morphotype were then determined by the grid-line-intersect method (Tennant 1975) using a 10- or 5-mm grid (depending on the numbers of tips in the sample) in 8.5-cm diameter plastic Petri dishes. Samples were counted three times and were redistributed between counts. Lengths were calculated from the average of three counts. Large samples were subdivided and the lengths determined for each subdivision were pooled to give the total length. The accuracy of the method was checked several times before sampling began, using different grid sizes and roots of predetermined total length. The biomass (dry weight) of EM tips and the total biomass of the nonmycorrhizal roots were determined after drying the material in a fan oven at 75°C for 48 h. Estimates were also made of moribund and dead mycorrhizal tips. The density, length and biomass of moribund tips were determined as described above and the length and biomass of nonmycorrhizal roots were also determined. The average EM biomass (standing crop, kg 100 $m^{-2}$, dry weight) in each plot was estimated as: EMb × $C$ × $D$, where EMb = mean EM dry biomass (kg) per core; $C$ = number of core areas per plot; $D$ = proportion of plot area covered by *Dryas*.

### Basidiome sampling

Basidiomes were collected from each of the permanent plots over a 3-year period: 1997–1999. Each of the plots was sampled at 2- to 3-week intervals during the main fruiting season of each year, i.e. between the last week of August and the first week of November. Each plot was sampled six times in 1997, nine times in 1998 and ten times in 1999, giving a total of 50 samples for the two plots. All visible basidiomes were collected from the plots and were identified, counted and weighed on the same day. Basidiome biomasses were determined by oven-drying at 75°C for 48 h.

### Data analyses

In plots 1 and 2, differences in the abundance of ectomycorrhizas between sampling dates were tested using one-way ANOVA on data (EM density, length, biomass) that were $\sqrt{x}$-transformed to equalise the variance (Zar 1999). Correlation of the mycorrhizal data between plots 1 and 2 were calculated using Spearman's rank correlation coefficient ($r_s$) (Zar 1999). EM diversity at each sampling date was assessed using the Shannon-Wiener diversity index ($H'$) (Shannon 1948) and Simpson's diversity index ($1-D_S$, the probability of picking two organisms that are different species; Simpson 1949; Krebs 1989). The similarities of the EM assemblages in plot 1, plot 2 and the 30 other sites were quantified using the modified form (Horn 1966) of Morisita's modified index of similarity ($I_M$) (Morisita 1959). Similarities in EM composition between sampling dates were compared between plots 1 and 2 by averaging $\sqrt{x}$-transformed $I_M$ values obtained on the seven sampling dates for each plot and by testing for significant differences between the averages using a Student's $t$-test.

The distribution patterns (uniform, random or clumped) of ectomycorrhizas in each plot on each sampling date were quantified and compared using the standardised form (Smith-Gill 1975) of Morisita's index of dispersion ($I_P$) (Morisita 1962). $I_P$ values <0 indicate uniform distribution, $I_P$=0.0 indicates random distribution, $I_P$>0 indicates clumped distribution. $I_P$ values of >0.5 indicate clumping at the 95% confidence level (Krebs 1989). Dispersion of the principal EM types was compared between plots by averaging $\sqrt{x}$-transformed $I_P$ values obtained on the seven sampling dates for each type in each plot and by testing for significant differences between the averages using Student's $t$-tests.

## Results

### Structure of the EM assemblage on *Dryas* roots

In the 196 soil cores sampled, a total of 142,745 EM tips were counted from the three sets of cores (plot 1, plot 2 and the 30 other sites). Thirty-four different EM morphotypes were distinguished, 29 from plot 1, 16 from plot 2 and 24 from the 30 other sites (Table 2). Eleven EM types were identified to species or genus using either RFLP comparisons or sequence data from the ITS-rDNA region (*Craterellus lutescens*, *Tomentella* sp. *Boletus* sp. *Tricholomamyomyces*), rhizomorph tracing to basidiomes (the

**Table 2** Frequency and length (percentage of total) of ectomycorrhizas (EM) on roots of *Dryas octopetala,* and biomass (grams) of EM basidiomes (sampled in plots 1 and 2 only), in *1* plot 1, *2* plot 2, and *3* 56 soil cores from 30 other sites in the Burren, western Ireland

| | EM frequency | | | EM Length | | | Basidiome biomass | |
|---|---|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 1 | 2 | 3 | 1 | 2 |
| *Craterellus lutescens* | 85.7 | 52.9 | 48.3 | 52.40 | 33.70 | 40.70 | 76.7 | 6.4 |
| *Tomentella* sp. | 95.7 | 98.6 | 85.7 | 10.20 | 22.61 | 22.40 | – | – |
| *Dryadirhiza fulgens* | 68.6 | 45.7 | 30.3 | 11.31 | 6.06 | 6.04 | – | – |
| *Cenococcum geophilum* | 78.6 | 8.6 | 34.0 | 9.00 | 0.03 | 12.30 | – | – |
| *Cortinarius* 3 | –[a] | 8.6 | – | – | 11.60 | – | – | – |
| *Boletus* sp. | 17.1 | 32.9 | 17.8 | 2.76 | 4.52 | 0.98 | – | – |
| *Cortinarius* 1 | 12.9 | 17.1 | 5.4 | 2.46 | 5.7 | 0.15 | – | – |
| Brown type | 10.0 | – | 7.1 | 0.81 | – | 5.04 | – | – |
| *Cortinarius mussivus* | 7.1 | 11.4 | 11 | 2.09 | 1.16 | 1.44 | – | 0.3 |
| *Hebeloma* sp.? | 11.4 | 7.1 | 12.5 | 0.43 | 1.03 | 3.05 | | 0.8 |
| *Cortinarius atrovirens* | 7.1 | 25.7 | 1.8 | 0.28 | 3.71 | 0.02 | 0.5 | 5.8 |
| *Cortinarius infractus* | 2.9 | – | 1.8 | 0.13 | – | 3.63 | – | 2.6 |
| *Cortinarius* 6 | 8.6 | 4.3 | 8.9 | 0.15 | 3.04 | 0.41 | – | – |
| *Cortinarius calochrous* | 7.1 | 4.3 | 1.8 | 0.54 | 3.04 | 0.11 | 0.4 | 29.3 |
| *Cortinarius* 4 | 2.9 | 8.6 | – | 0.05 | 2.19 | – | – | – |
| *Cortinarius odorifer* | 1.4 | – | 10.9 | 0.86 | – | 1.0 | 4.1 | 7.0 |
| *Cortinarius* 7 | 7.1 | – | 5.4 | 1.26 | – | 0.12 | – | – |
| *Cortinarius* 2 | 5.7 | 4.3 | – | 1.02 | 0.42 | – | – | – |
| *Cortinarius brunneus* | 5.7 | – | – | 1.48 | – | – | 5.9 | – |
| *Tricholoma myomyces* | 27 | – | 5.4 | 1.04 | – | 0.24 | 0.6 | – |
| Orange type 5 | 10.0 | 1.4 | 5.4 | 0.68 | 0.09 | 0.49 | – | – |
| *Dryadirhiza cerina* | 5.7 | – | 8.9 | 0.13 | – | 0.97 | – | – |
| Orange type 1 | 12.9 | – | 7.1 | 0.27 | – | 0.37 | – | – |
| *Dryadirhiza rugosa* | 5.7 | – | 3.6 | 0.53 | – | 0.14 | – | – |
| *Dryadirhiza truncata* | – | 2.9 | 1.8 | – | 0.57 | 0.02 | – | – |
| *Cortinarius* 5 | 2.9 | – | – | 0.47 | – | – | – | – |
| Orange type 3 | – | – | 1.8 | – | – | 0.17 | – | – |
| Orange type 2 | – | – | 1.8 | – | – | 0.17 | – | – |
| Russet type | 2.9 | – | – | 0.05 | – | – | – | – |
| Orange type 4 | – | – | 1.8 | – | – | 0.11 | – | – |
| Brown hispid type | 1.4 | – | – | 0.06 | – | – | – | – |
| Grey coralloid type | 1.4 | – | – | 0.03 | – | – | – | – |
| Golden type | 1.4 | – | – | 0.02 | – | – | – | – |
| *Hebeloma* type | 1.4 | – | – | 0.01 | – | – | – | – |
| *Boletus erythropus* | – | – | – | – | – | – | – | 6.8 |
| *B. luridus* | – | – | – | – | – | – | 6.5 | – |
| *Cortinarius anomalus* | – | – | – | – | – | – | 4.3 | – |
| *C. caesiocanescens* | – | – | – | – | – | – | – | 36.7 |
| *C. atrovirens* | – | – | – | – | – | – | – | 5.8 |
| *C. croceocaeruleus* | – | – | – | – | – | – | 0.1 | – |
| *C. uraceus* | – | – | – | – | – | – | 0.1 | – |
| *C. venetus* | – | – | – | – | – | – | 0.0 | – |
| *Cortinarius* sp. | – | – | – | – | – | – | 0.01 | – |
| *Cortinarius* sp. | – | – | – | – | – | – | 0.01 | 0.1 |
| *Hebeloma* sp. | – | – | – | – | – | – | – | 0.8 |
| *Inocybe bongardii* | – | – | – | – | – | – | 0.01 | – |
| *Ramaria* sp. | – | – | – | – | – | – | – | 3.8 |
| *Tricholoma scalpturatum* | – | – | – | – | – | – | 0.2 | – |
| *Telamonia* sp. 1 | – | – | – | – | – | – | 0.5 | – |
| *Telamonia* sp. 2 | – | – | – | – | – | – | 0.2 | 0.3 |
| Number of morphotypes/species | 29 | 16 | 24 | | | | 18 | 14 |

[a]Not present

**Table 3** Mean EM density, biomass and EM basidiome production in plots 1 and 2. Values in parenthesis represent SD

| | Plot 1 | Plot 2 |
|---|---|---|
| EM tips ($m^{-2}\times10^5$) | 5.86 (4.4) | 2.50 (2.3) |
| EM biomass (kg $ha^{-1}$) | 220.6 (223.7) | 70 (110) |
| Basidiome production (kg $ha^{-1}$ $year^{-1}$) | 10.9 (6.6) | 1.9 (1.6) |

six identified *Cortinarius* spp.), and anatomical features (*Cenococcum geophilum*). Four types that could not be identified to species were characterized by morphotyping (denoted by the generic epithet "*Dryadirhiza*" in Table 2). This group of 15 EM types together constituted 88% of the total EM length measured in the 196 soil cores (see Harrington and Mitchell 2002, for detailed descriptions of these types).

The remaining 19 types were for the most part uncommon or rare types that could not be identified reliably to recognized EM species or morphotypes. Seven of these have been provisionally identified as *Cortinarius*-type ectomycorrhizas because of their gross morphology and plectenchymatous mantle structure. Six EM types are designated 'Orange' types based on their appearance. All of these are short-branched ectomycorrhizas with mantle structures ranging from plectenchymatous to pseudoparenchymatous. Two brown types (brown and brown hispid types) are possibly species of *Tomentella* on account of their pigmentation and mantle structure.

Three EM morphotypes, *Craterellus lutescens*, *Tomentella* sp. and *D. fulgens* dominated the EM assemblage. *Cenococcum geophilum* was uncommon in plot 2 but was abundant elsewhere. Ectomycorrhizas of *Craterellus lutescens* were by far the most abundant in terms of tip density, biomass and total length (42%), while those of *Tomentella* sp. were the most common in terms of frequency. Some morphotypes that were poorly represented in terms of biomass occurred relatively frequently in the soil cores, for example *Boletus* sp. and *Cortinarius atrovirens*. Combining the three datasets, frequency and EM length were significantly correlated using Spearman's correlation coefficient ($r_s$=0.736, $P$<0.0001). Morisita's similarity index ($I_M$) was

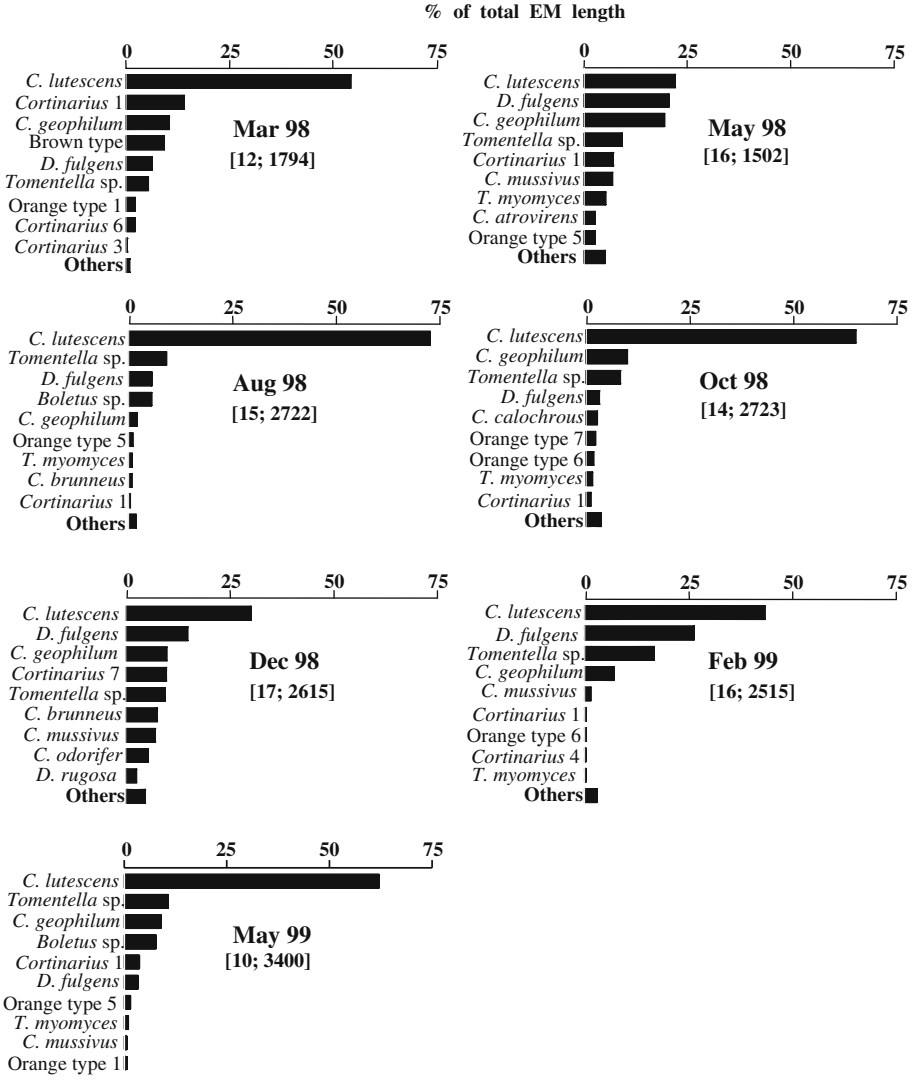


**Fig. 1** Variation in occurrence of different ectomycorrhizal (EM) morphotypes on roots of *Dryas octopetala* in plot 1 (eastern margin of the Burren, western Ireland) over the sampling period, expressed as a percentage of total mycorrhizal length on each sampling date. Numbers in *square brackets* for each month refer to the number of EM types recorded and total EM length (cm)

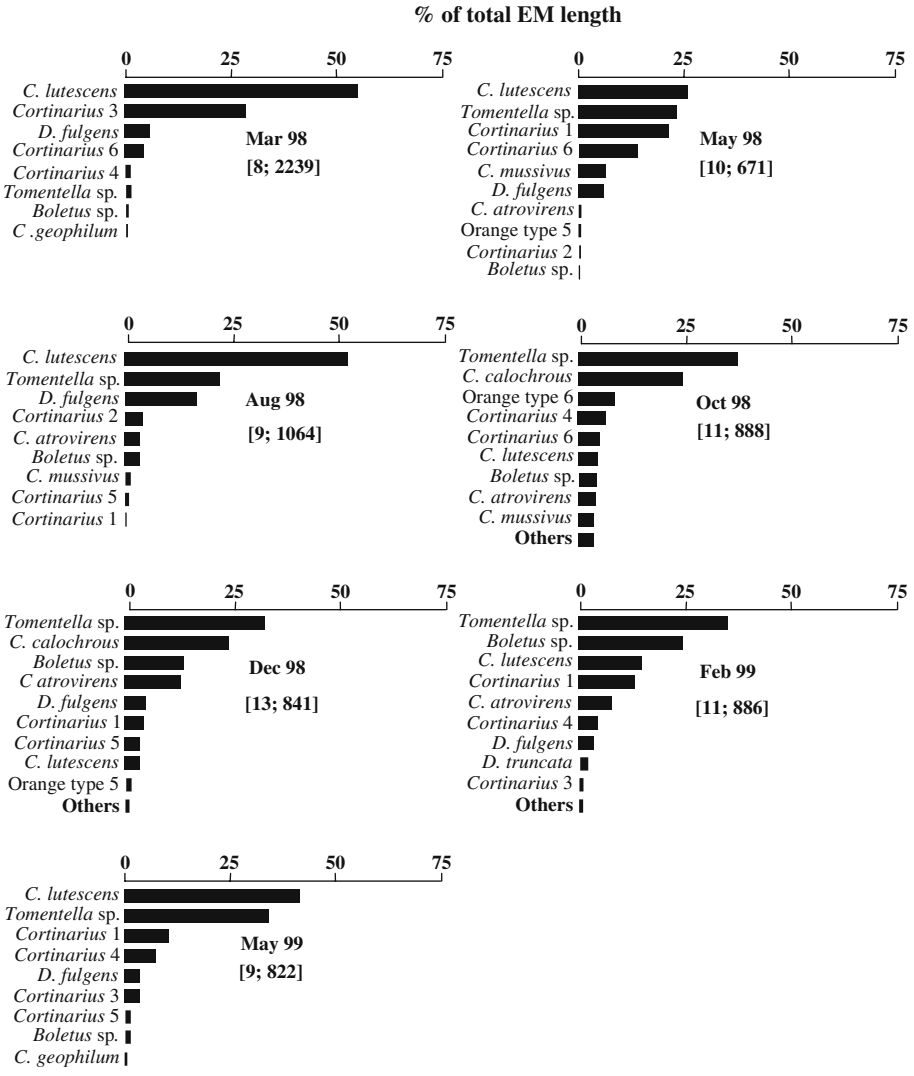


**Fig. 2** Variation in occurrence of different mycorrhizal morphotypes on roots of *D. octopetala* in plot 2 (western margin of the Burren, western Ireland) over the sampling period, expressed as a percentage of total mycorrhizal length on each sampling date. Numbers in *square brackets* for each month refer to number of EM types recorded and total EM length (cm)

calculated to assess the similarity between the three sets of cores. The index values for each pairwise comparison of the three sets were very high (i.e. $I_M$ of 0.84 between plots 1 and 2, 0.93 between plot 1 and the 30 other sites, and 0.89 between plot 2 and the 30 other sites), indicating that all three datasets are derived from the same EM community.

Mean EM density and biomass were estimated on an area basis for plot 1 and plot 2 from 70 sample cores each (Table 3). Mean density of EM tips, mean EM biomass and average annual production of basidiomes of EM species were significantly greater in plot 1 than in plot 2. In both plots, EM morphotype richness exceeded EM 'basidiome species' richness (Table 2). In plot 1, six species were identified as common to both basidiome and ectomycorrhizal assemblages (*Craterellus lutescens*, *Cortinarius atrovirens*, *Cortinarius brunneus*, *Cortinarius calochrous*, *Cortinarius odorifer* and *Tricholoma myomyces*), and five were identified in plot 2 (*Craterellus lutescens*, *Cortinarius atrovirens*, *Cortinarius calochrous*, *Cortinarius mussivus* and *Hebeloma* sp.). *Craterellus lutescens* was the most abundant EM species below-ground in both plots in terms of EM length (Table 2) and biomass, and was also the most prolific fruiting species in plot 1. In plot 2, however, *Cortinarius caesiocanescens*

**Table 4** Mean of standardised values of Morisita's dispersion index ($I_P$) for the five most abundant EM types, and for all EM types combined, in plots 1 and 2. Means are the average of $I_P$ values calculated for each of the seven sampling dates. SD in parentheses

| | Mean $I_P$ | |
|---|---|---|
| | Plot 1 | Plot 2 |
| *Cenococcum geophilum* | 0.67 (0.09) | 0.93 (0.14)* |
| *Craterellus lutescens* | 0.57 (0.04) | 0.66 (0.09)* |
| *Dryadirhiza aerea* | 0.83 (0.16) | 0.80 (0.13) |
| *Dryadirhiza fulgens* | 0.63 (0.05) | 0.78 (0.12)* |
| *Dryadirhiza nigra* | 0.54 (0.02) | 0.57 (0.06) |
| All EM types | 0.53 (0.01) | 0.53 (0.01) |

*Differences in mean $I_P$ values between plots are significantly different ($P<0.05$) using *t*-test

and *Cortinarius calochrous* produced the greatest basidiome biomass (Table 2), even though *Cortinarius* ectomycorrhizas as a group constituted only 30% of the total EM length.

## Temporal variation in the EM community

*Craterellus lutescens* dominated the EM community in plot 1 on all of the sampling occasions (Fig. 1), and frequently exceeded the biomass of all other EM types. In August 1998, for example, in plot 1, this ectomycorrhiza comprised 73% of the EM length (Fig. 1), 59% of the EM tips, and 80% of the EM biomass. In plot 2, *Craterellus lutescens* dominated the EM community on four of the seven sampling dates (Fig. 2). Mycorrhizas of *Tomentella* sp., *Boletus* sp. and *Cortinarius* were more abundant in relative terms, and sometimes in absolute terms, in plot 2 than in plot 1 for each abundance measure. Similarity values calculated between all pairs of sampling dates were significantly higher ($P<0.001$) in plot 1 (mean $I_M=0.81$; SD=±0.13) than in plot 2 (mean $I_M=0.59$; SD=±0.21), indicating a greater degree of homogeneity in EM community composition in plot 1 than in plot 2 over the sampling period. Standardised values of Morisita's index of dispersion ($I_P$) were in excess

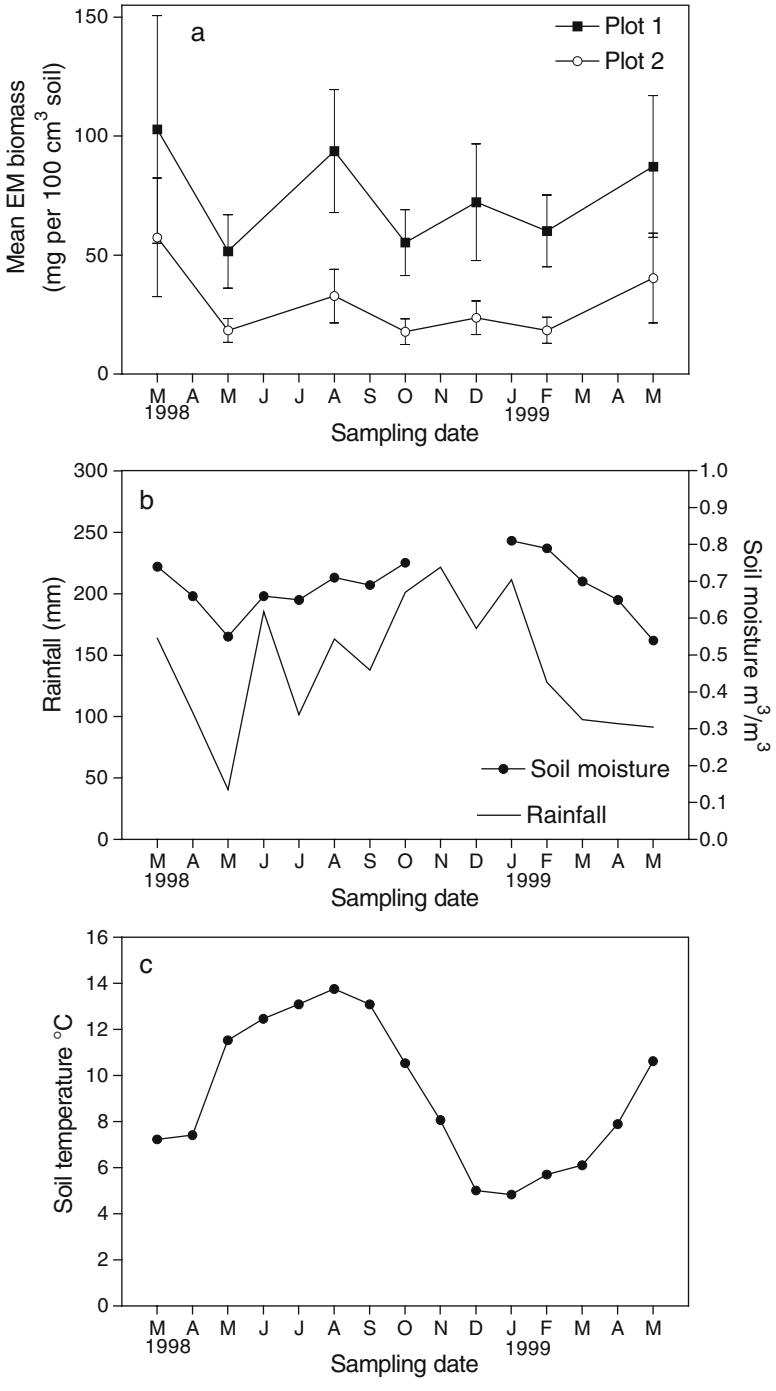


**Fig. 3** **a** Variation in mean live EM biomass (mg dry mass) on roots of *D. octopetala*, per 100 $cm^3$ soil in plots 1 and 2 (Burren, western Ireland), over the sampling period March 1998–May 1999. *Error bars* 1 SE. **b** Mean monthly soil moisture content on Gleninagh Mt. and rainfall at Carron. **c** Mean monthly soil temperature at Gleninagh

**Table 5** Correlations (Spearman's $r_s$) and probabilities ($P$) between plots 1 and 2 in respect of variation in EM abundance and diversity over the sampling period

| | $r_s$ | $P$ |
|---|---|---|
| EM biomass | 0.86 | 0.01 |
| EM length | 0.13 | 0.78 |
| EM density | 0.13 | 0.78 |
| Live/moribund ratio | 0.75 | 0.05 |
| Number of EM types | 0.69 | 0.86 |
| Shannon–Wiener index ($H'$) | 0.93 | 0.003 |
| Simpson's index ($1-D_S$) | 0.86 | 0.01 |
| *Craterellus* EM biomass | 0.75 | 0.05 |
| *Craterellus* EM length | 0.43 | 0.34 |
| *Craterellus* EM density | 0.83 | 0.02 |

of +0.5 for all EM morphotypes on all sampling dates, indicating statistically ($P$>0.95) clumped distribution patterns (Table 4). The average $I_P$ values over the seven sampling dates for *Craterellus lutescens*, *Cenococcum geophilum* and *D. fulgens* were significantly greater in plot 2 than in plot 1, indicating a greater degree of clumping in the distribution of these ectomycorrhizas in plot 2.

The maximum numbers of EM types in each plot were recorded in the same month (i.e. December 1998) (Figs. 1, 2) and were approximately 70% and 62% greater than the minima in plots 1 and 2, respectively. However, there was no evidence of a marked seasonal pattern in EM abundance in plots 1 or 2. One-way ANOVA showed that, for each measure of EM abundance, the variation between sampling dates was not significantly different ($P$>0.05) because of statistical error between samples, which was generally very large on most sampling dates, reflecting clumping in the distribution of mycorrhizas. The variation in EM biomass was not related to soil temperature and rainfall data over the 14-month period of study. There was a poor, insignificant ($P$>0.05) correlation between tip density, length or biomass of all EM types combined or individual EM types, and either soil moisture, soil temperature or rainfall.

However, comparison of abundance patterns between the plots suggests that time-related trends were present. EM biomass in plot 1 greatly exceeded that in plot 2 on all sampling dates (Fig. 3a) but was strongly correlated between both plots (Table 5). However, EM density and length were

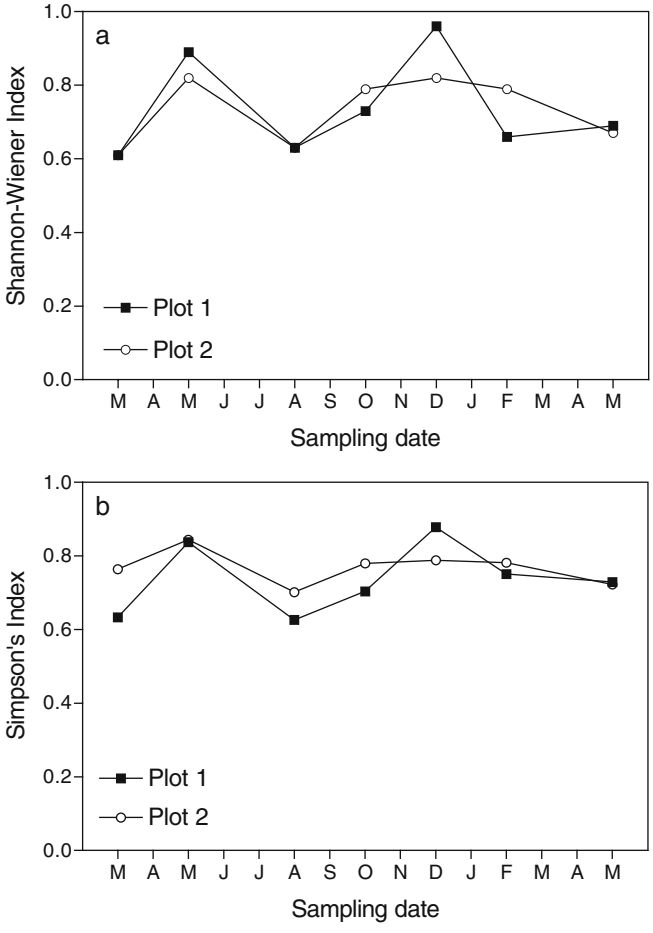


**Fig. 4** **a** Variation in Shannon-Wiener diversity values ($H'$); **b** variation in Simpson's diversity values ($1-D_S$), in plots 1 and 2 (Burren, western Ireland) at each sampling date from March 1998 to May 1999

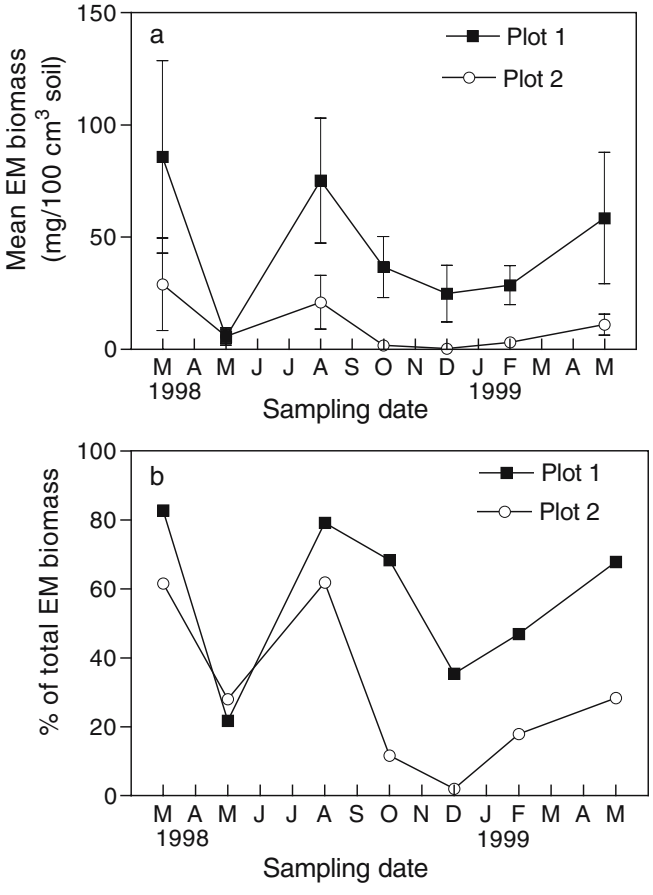


**Fig. 5** Variation in EM biomass of *Craterellus lutescens* ectomycorrhizas on roots of *D. octopetala*, in plots 1 and 2 in the Burren, western Ireland, from March 1998 to May 1999 expressed as **a** mean biomass (mg) per 100 cm$^3$ soil (*error bar* 1 SE), **b** percentage of total EM biomass

**Table 6** Correlations (Spearman's $r_s$) of diversity indices and number of EM types, with tip density, length and biomass of *Craterellus lutescens* mycorrhizas in plots 1 and 2 over the sampling period

| | Density | | Length | | Biomass | |
|---|---|---|---|---|---|---|
| | Plot 1 | Plot 2 | Plot 1 | Plot 2 | Plot 1 | Plot 2 |
| Shannon-Wiener index ($H'$) | −0.86** | −0.91** | −0.50 | −0.84* | −0.86** | −0.84* |
| Simpson's index ($1-D_S$) | −0.89** | −0.71 | −0.71 | −0.64 | −0.89** | −0.64 |
| Number of EM types | −0.76* | −0.95** | −0.79* | −0.99** | −0.76* | −0.99** |

*Significant correlation at $P \leq 0.05$
**Significant correlation at $P \leq 0.01$

not significantly correlated between both plots. Changes in the relative proportions of live and moribund EM tips in the two plots also showed correspondence and synchronicity. Shannon-Wiener diversity indices and Simpson's diversity indices calculated for each sampling date were similar in both plots (Fig. 4) and changes in both indices were strongly correlated between both plots over the sampling period (Table 5). Shannon-Wiener values were significantly inversely related to EM length and biomass in plot 2 ($r_s$= −0.93, $P$<0.01 and $r_s$=−0.80, $P$<0.05, respectively) and Simpson's index values were significantly inversely related to EM length and tip density in plot 2 ($r_s$=−0.83, $P$<0.05 and $r_s$=−0.82, $P$<0.05, respectively). Plots 1 and 2 showed similar abundance patterns of *Craterellus lutescens* mycorrhizas throughout the period of study. Biomass (Fig. 5) and density of *Craterellus* mycorrhizas were significantly correlated between both plots (Table 5). *Craterellus* EM biomass declined in absolute and relative terms in May and December 1998 in cores from both plots, the decline in May coinciding with the lowest levels of soil moisture recorded in 1998 (Fig. 3b). Number of EM types recorded on sampling dates were negatively correlated with tip density, length and biomass of *Craterellus* mycorrhizas in both plots (Table 6). Shannon-Wiener values showed a significant negative correlation with density and biomass of *Craterellus* mycorrhizas in both plots and Simpson's index values were significantly negatively correlated with tip density and biomass in plot 1 (Table 6). In both plots, therefore, peaks in EM biomass, largely contributed by *Craterellus*, (e.g. March, August 1998; May 1999) were accompanied by declines in diversity (see Figs. 1, 2).

## Discussion

This is the first intensive study to investigate the composition and temporal variation in the EM assemblage on *D. octopetala* in a natural community. The number of EM types found on *Dryas* in the Burren is greater than that reported in previous studies of *Dryas* ectomycorrhizas in arctic-alpine habitats (Harrington and Mitchell 2002) but this may be a consequence of greater sampling effort in the present study. Numbers of EM types on *Dryas* in the Burren were similar to the numbers recorded on *Arctostaphylos uva-ursi* in California (Horton et al. 1999) but were over twice as numerous as the number of types in *Salix repens* stands in the Netherlands (van der Heijden et al. 1999).

The four EM types (*Craterellus lutescens*, *Tomentella* sp., *Dryadirhiza fulgens* and *Cenococcum geophilum*) that dominated the EM assemblage on *D. octopetala* together comprised over 70% of the EM biomass in each permanent plot and in the 30 other sites. Most other morphotypes were infrequently encountered and were low in abundance. This pattern of species abundance has been described in most studies of EM diversity in forest habitats (Visser 1995; Gehring et al. 1998; Fransson et al. 2000; Dahlberg 2001; Horton and Bruns 2001), even though the number of EM morphotypes recorded in EM assemblages in forests is generally greater than here. The average biomass of *Dryas* ectomycorrhizas per sampling date (220 and 77 kg ha$^{-1}$ in plots 1 and 2, respectively) was, not surprisingly, less than the comparable figures for forests. Horton and Bruns (2001), in a review of studies of EM communities in forests, have concluded that even the most abundant EM species are confined to a minority of soil cores (<10%) because they have extremely clumped distributions. The *Dryas* mycorrhizas also show clumped distributions (Table 4), but the highly significant correlation between EM frequency and biomass indicates that EM distribution patterns, at least for the dominant EM types, show less clumping here than is found in EM forest communities, where EM frequency and abundance are generally unrelated. This may be due to the shallow Burren soils lacking horizons and spatial heterogeneity, the presence of a single abundant host (*Dryas*), and the relatively low number of EM types compared to forests.

Of the four dominant EM types, *Craterellus lutescens* is regarded as an EM associate of forest trees. Cantharelloid or craterelloid EM fungi have not previously been reported as EM associates of *D. octopetala*. Tomentelloid fungi are among the primary dominants in several North American forest ecosystems (Danielson et al. 1984; Gardes and Bruns 1996; Horton and Bruns 2001; Visser 1995) and are also very common in some European forests (Brand 1991; Fransson et al. 2000; Erland and Taylor 1999). *Cenococcum geophilum* is a very common EM coloniser of tree roots and can frequently be the dominant type, or among the generally small number of dominating species (Baxter et al. 1999; Vogt et al. 1981). It is likely that the number of EM types may have been overestimated in that some may be morphological variants of a single species. The un-

identified *Cortinarius* types 1–7, for example, may have included identified *Cortinarius* spp. but were not distinguished because of variation caused by age. Egli et al. (1993) came to a similar conclusion in relation to Norway spruce ectomycorrhizas. On the other hand, as has been found in many other studies (Kårén and Nylund 1997; Erland et al. 1999), it is also likely that some EM types (e.g. *Cortinarius* 1) contain more variation than is apparent from morphological and anatomical criteria alone. This problem can be solved only by further molecular analysis.

The relative abundance of basidiomes of EM species reflected the relative abundance of ectomycorrhizas below-ground in plot 1, but not in plot 2. Here particularly, *Cortinarius* ectomycorrhizas were less abundant below-ground than was indicated by their basidiomes, a phenomenon frequently observed in forests. Basidiome diversity and abundance in *Dryas* vegetation in the Burren is, therefore, an unreliable indicator of the composition of the below-ground EM community. A similar conclusion was drawn from a study on *Salix repens* stands (van der Heijden et al. 1999) and from those of many forest ecosystems (Dahlberg 2001).

Population variables such as numbers of EM tips, length and biomass showed considerable variation between sampling dates but these differences were not significant because of the large variation in EM abundance between core samples collected within plots on individual sampling dates. The numbers of EM types also varied between sampling dates. However, when the dominant types are considered, community composition remained very similar in individual plots over the sampling period, particularly in plot 1. It appears, therefore, that there were no significant shifts in population structure evident in either plot at the end of the sampling period compared to the start. Peter et al. (2001) also found very little difference in EM composition and population structure over 3 years in a Norway spruce stand in Switzerland using ITS-RFLP analysis.

Comparison between plots 1 and 2 indicate that changes in EM populations over the sampling period were synchronised and correlated, at least with respect to some measures such as biomass of ectomycorrhizas and diversity. Synchronous declines in EM abundance in May 1998 and increases in August 1998 were conspicuous in both plots, and in both cases are due primarily to changes in the population of the dominant *Craterellus lutescens*. Peaks of *Craterellus* EM biomass in August in both plots occurred approximately 1 month before the period of basidiome production. In both plots, the total number of EM types and diversity were negatively correlated with abundance of *Craterellus* mycorrhizas over the sampling period. It is unlikely that such correlations would be found between the two plots over the sampling period if the observed differences in EM abundance between dates were due solely to variation in the spatial distribution of ectomycorrhizas in the plots. Van der Heijden et al. (1999) also observed shifts in abundance of certain EM types between spring and autumn in *S. repens* sites, which they concluded could not be due to sampling errors because they occurred in most sites.

The causes of these fluctuations are unclear. They did not show a seasonal trend, and such trends are unlikely because of the climatic regime. Rainfall patterns in the Burren may have contributed to some of the variation. Burren soils appear to have a very limited water storage capacity (Jeffrey 2003), and the drying of these shallow, free-draining soils could have a negative impact on some of the EM associates of *Dryas* that are normally found in forests. For example, mycorrhizas of drought-tolerant *Cenococcum geophilum* were most abundant, as a proportion of the total EM population, in May 1998 in plot 1, when soil moisture levels were low. Correspondingly, mycorrhizas of *Craterellus lutescens* declined in this month in both plots. Worley and Hacskaylo (1959) have shown that such reductions in soil moisture can change EM relationships. Taylor and Alexander (1989) also found that fluctuations in the absolute numbers of ectomycorrhizas of the dominant morphotypes on Sitka spruce in Scotland were related specifically to rainfall rather than seasonal changes.

In conclusion, this study has provided further evidence that four EM types dominate the EM community on *D. octopetala* in the Burren. The species profiles and relative abundance from both the permanent plots and other sites suggest that they represent the same EM community. There were no significant short-term temporal variations in total EM biomass but changes did occur in abundance of *Craterellus lutescens*, a dominant EM type, during the 14-month study period. However, there was no evidence that these changes were related to weather patterns.

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