

## Succession of ectomycorrhizal fungi in roadside verges planted with common oak (*Quercus robur* L.) in Drenthe, The Netherlands

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**Abstract.** In the period 1986–1988, sporocarps of ectomycorrhizal fungi were counted in 53 plots in roadside verges planted with common oak in Drenthe, The Netherlands, belonging to three vegetation types. Twenty five plots were *Hypochaeris radicata-Quercus* communities, comprising roadside verges in open landscapes on dry, acid, sandy soils, poor in nutrients. In each plot, the trees were even-aged but varied between 10 and 140 years. The relationship between tree age and numbers of ectomycorrhizal species and sporocarps was studied. The species composition of roadside verges was compared between young trees (10–20 years), medium-old trees (20–50 years) and old trees (50–140 years). The species composition in roadside verges in open landscape with old trees was also compared with the ectomycorrhizal fungi in two further different types of roadside verges with old oak trees, viz the *Anthriscus sylvestris-Quercus* type, comprising open to half-open plots on soil richer in nutrients, and the *Mnium hornum-Quercus* type, comprising shady plots surrounded by forest. The data reveal a distinct succession of ectomycorrhizal fungi with increasing tree age. The results are compared with data on succession of ectomycorrhizal fungi in forest stands. It is concluded that the succession cannot be modeled simply as suggested by some authors. Instead, changes in species composition and diversity showed much variation in relation to different environmental conditions. The factors relevant to the course of ectomycorrhizal succession are discussed. The concepts of early- and late-stage fungi are critically considered. It is concluded that this classification is not appropriate for the description of the ectomycorrhizal succession during stand development. A new, provisional classification of ectomycorrhizal fungi on the basis of their appearance during forest succession is proposed.

**Key words:** Ectomycorrhizal fungi – Forests – Roadside verges – Succession

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### Introduction

In the present paper, the temporal sequence of fungal communities in roadside verges is studied. According to Clements (1928), succession is the sequence of developmental stages of a (phytocoenological) formation, comparable with the life history of an individual plant. This concept was mainly based on the development of phytocoena from pioneer to climax stages under natural circumstances. In the present study, the concept of succession is applied to the sequence of fungal communities in a highly artificial habitat, where the natural climax vegetation will never develop. Therefore, in this paper succession is defined as “the non-seasonal, directional and continuous pattern of colonization and extinction on a site by species populations” (Begon et al. 1986).

Thus, the main aim was to describe the different fungal communities observed in roadside verges with trees of different ages but similar in other respects. The study was restricted to ectomycorrhizal fungi which were associated with the roots of the trees. In fact, only part of the community was studied, in that the fungi were considered but the ageing trees remained the same. Thus, succession as conceived here may differ from other approaches where complete phytocoenoses are usually studied. Nevertheless, a succession of fungi is clearly present in the sense of the above-mentioned definition as fungal populations colonize and disappear from the studied sites according to general rules.

Information on the succession of ectomycorrhizal fungi during forest development has been mainly derived from experimental stands of *Betula pendula* Roth with trees up to 15 years old (e.g. Last et al. 1987). Continuous change in the ectomycorrhizal flora over the years has been convincingly shown by both the appearance of sporocarps and the occurrence of different types of ectomycorrhizas (Gibson and Deacon 1988). The mycelia of early colonizers was found to expand from the tree stem with the growing root system at a rate of approximately 0.1 to 0.2 m/year (Last et al. 1984). After some years, ectomycorrhizal fungi tend to

occur in circles around trees, the early colonizers near the margin of the root system, the late colonizers near the trunk (Mason et al. 1982).

In order to explain this sequence, experiments were carried out with different species in various conditions. It appeared that fungi appearing early and late in the succession were both able to form mycorrhizas on seedlings in axenic laboratory conditions. However, only one group, termed "early-stage fungi", were able to colonize seedlings from basidiospores in unsterile conditions (e.g. Deacon et al. 1983; Mason et al. 1984). Early-stage fungi are considered as *r*-strategists, generally combining a relatively low carbon demand, a rapid mycelial growth, the production of relatively small sporocarps and occurrence in soils with nutrients mainly in the inorganic pool (Dighton and Mason 1985). Early-stage fungi are the only ectomycorrhizal species in very young, even-aged stands, e.g. on afforested arable land and in silvicultures of the clear-cutting system. The first sporocarps in stands of *Betula* appeared 2 years after planting of seedlings (Mason et al. 1987). The early-stage fungi comprise relatively few species, mainly of the genera *Laccaria*, *Hebeloma*, *Inocybe* and *Thelephora*. Some species are considered to be restricted exclusively to young trees (e.g. *Laccaria tortilis*, *Hebeloma crustuliniforme*, *Thelephora terrestris*), whereas others are also observed in older stands (Dighton and Mason 1985).

"Late-stage fungi", on the other hand, are considered as *K*-strategists, generally having a higher carbohydrate demand, a slower mycelial growth, the formation of hyphal strands, on average larger sporocarps and growing in environments where nutrients are mainly in the organic pool (Dighton and Mason 1985). They are not able to colonize roots of seedlings from basidiospores under unsterile conditions, but at least some species are able to colonize young trees vegetatively from ectomycorrhizal roots of neighbouring mature trees (Fleming 1984). In stands of *Betula pendula*, sporocarps of the first late-stage fungi already appear 4 years after planting and are dominant on roots of trees 10 years old or older (Last et al. 1987). The great majority of ectomycorrhizal fungi belongs to this category, for instance *Paxillus involutus* and all species of *Boletus* and its allies, *Russula*, *Lactarius*, *Cortinarius* and *Amanita*.

Comparative mycocoenological studies were carried out by Keizer in the period 1986–1988 in roadside verges planted with common oak (*Quercus robur* L.) and beech (*Fagus sylvatica* L.), referred to below as *Quercus* and *Fagus*, respectively. The aims of this research were: (1) to describe the variation in macromycete communities in this habitat; (2) to compare a classification of macromycete communities with that of communities of green plants; (3) to determine the main factors which contribute to differences in composition of mycocoenoses; (4) to compare the macromycete communities in roadside verges including trees with those of related forest communities.

From studies on succession of ectomycorrhizal fungi it has become evident that one of the main factors de-

termining the composition of the ectomycorrhizal flora is the age of the associated host trees (e.g. Dighton and Mason 1985; Mason et al. 1987).

Therefore, tree age was chosen as one of the variables studied in roadside verges. The age of the trees in these plots ranged from 10 to 140 years (in 1988). The selection of plots with trees of different age classes was restricted to roadside verges in open landscapes ("open" plots) with oak on relatively poor soils, belonging to the *Hypochaeris radicata-Quercus* type (Keizer 1993).

Investigation of succession phenomena by this method is of course only indirect, because spatially separate units do not necessarily represent instant pictures from one and the same time series. The data should, therefore, be interpreted with care. On the other hand, direct observations on succession series of this duration are practically impossible (Oldeman 1990). Moreover, the course of the process in roadside verges may be strongly influenced by changing environmental factors, including the deposition of pollutants (Fellner 1988; Arnolds 1991) and changed use of these verges. We have tried to meet most objections connected with indirect observations by the selection of plots with relatively homogeneous soil conditions, in one vegetation type and in a rather small area (approximately 1800 km<sup>2</sup>). Oak trees planted along roads are taken from nurseries when they are approximately 10 years old. Thus, a methodological advantage of roadside verges (and most other tree plantations) above natural forest stands is that trees along a road are even-aged.

All plots with *Quercus* belonging to other vegetation types as well as those with *Fagus* had only mature trees over 50 years old. Detailed results of these studies have been published elsewhere, including classification of the mycocoenoses in roadside verges with *Quercus* and *Fagus*, correlations between mycocoenological characteristics and environmental variables, and a comparative study on roadside verges planted with trees in relation to forest communities with the same tree species (Keizer 1993).

## Material and methods

In 1986, a total of 53 plots in roadside verges planted with *Quercus robur* were selected in Drenthe, in the northeast of the Netherlands. The area lies about 10–20 m above sea level and has a cool-temperature climate (average precipitation 781 mm/year, mean temperature in January 1.2° C, and in July 15.9° C). All plots were situated on acidic, pleistocene sands with a variable organic matter content and a more or less disturbed soil profile due to road construction. The trees were of local provenance.

The plots were 100 m long and the width varied between 1.5 and 6.5 m with 9–35 trees per plot. All sporocarps of macromycetes were counted by species at intervals of 3–4 weeks in the period August–November during the years 1986–1988. In most plots, the position of all sporocarps of ectomycorrhizal fungi was indicated on detailed maps (scale 1:100) during each visit. Soil profiles and some soil-chemical characteristics were determined in all plots (for details see Keizer 1993). In addition, the communities of green plants were described and classified according to

the Braun-Blanquet method (e.g. Westhoff and Van der Maarel 1973).

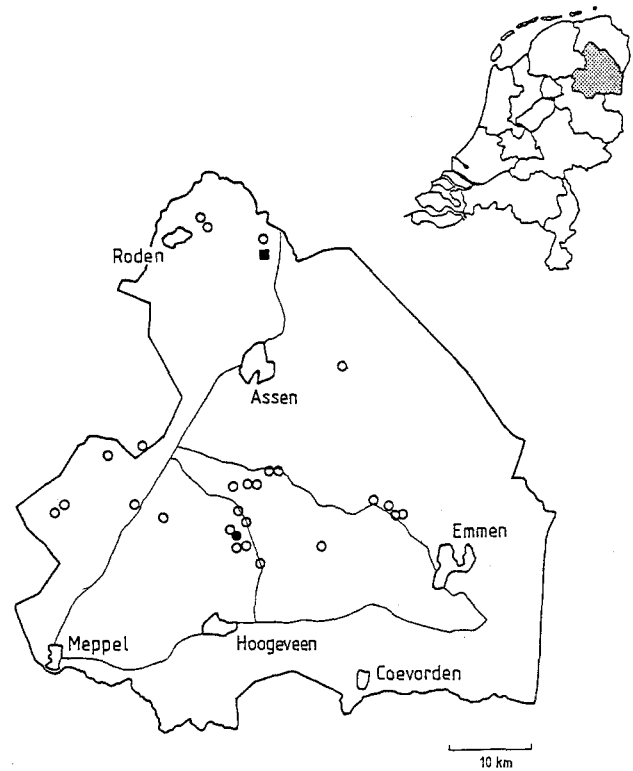
Plots belonging to the *Hypochoeris radicata-Quercus* type were divided into three age classes of the trees: "young" (10–20 years; 5 plots), "medium old" (20–50 years; 11 plots) and "old" (50–140 years; 10 plots). The age of the trees was determined with an increment borer. The *Hypochoeris radicata-Quercus* type is characterized as roadside verges in open landscapes with a short, low-productive, grass-rich herb layer on rather dry to dry oligotrophic to mesotrophic soils.

In addition to the 26 plots of the *Hypochoeris radicata* type (indicated in Fig. 1), 23 plots belong to the *Anthriscus sylvestris* type, open to half-open roadside verges on slightly richer soils, and four plots to the *Mnium hornum* type, shady plots along roads through forest stands. All plots of these types belong to the "old" age class.

On the basis of vegetation relevés, phytocoenological classifications were carried out for the roadside verges for ectomycorrhizal and saprotrophic fungi independently, using the computer program TWINSPIN (Hill 1979; Jongman et al. 1989). The methods and results are discussed in extenso by Keizer (1993). The data on saprotrophic fungi are left out of consideration in this paper. The plots were divided into three types. Some phytocoenological and environmental characteristics are summarized in Table 1. In the data presented here, species found in less than three plots were omitted.

Two parameters are used: (1) the presence-degree, i.e. the percentage of plots of a type in which a certain species was found; (2) the average maximum numbers of sporocarps per visit over 3 years. The maximum numbers of sporocarps (or carpophores) encountered in one visit during 3 years is considered as the most reliable expression of the potential fruiting capacity of a species in a plot (Arnolds 1981; Barkman 1987). Consequently, the average maximum numbers of sporocarps is the best expression of the potential fruiting capacity of a species in a set of plots belonging to a certain vegetation type.

A species is considered to be differential for a certain age class when: (1) its presence-degree is at least twice the presence-degree in other classes, or (2) its average maximum number is at



**Fig. 1.** The geographical situation of the studies plots. ○ = plots in roadside verges planted with common oak of the *Hypochoeris radicata-Quercus* type; ■ = Eelde Meteorological Station; ● = Wijster Biological Station

least three times that in other classes. The combination of presence and abundance characters in the criteria for differential species prevents the application of statistical tests.

**Table 1.** Some phytocoenological and environmental characteristics of three types of roadside verges with planted oak trees

	Phytocoenological type		
	<i>Hypochoeris radicata-Quercus</i> type	<i>Anthriscus sylvestris-Quercus</i> type	<i>Mnium hornum-Quercus</i> type
Number of plots	26	23	4
Ages of trees (years, 1988)	10–114	26–146	102–144
Phytocoenological characteristics			
Av. no. of plant species ( $\pm$ SD)	37 $\pm$ 9	29 $\pm$ 7	19 $\pm$ 5
Av. coverage herb layer (% , $\pm$ SD)	75 $\pm$ 19	72 $\pm$ 17	35 $\pm$ 11
Av. coverage moss layer (% , $\pm$ SD)	14 $\pm$ 16	0.5 $\pm$ 2.0	6 $\pm$ 4.7
Some differential species	<i>Hypochoeris radicata</i> <i>Leontodon autumnalis</i> <i>Luzula campestris</i>	<i>Anthriscus sylvestris</i> <i>Stellaria media</i> <i>Poa trivialis</i>	<i>Mnium hornum</i> <i>Deschampsia flexuosa</i> <i>Hypnum cupressiforme</i>
Environmental characteristics <sup>a</sup>			
Av. litter layer (cm, $\pm$ SD)	0.2 $\pm$ 0.5	1.1 $\pm$ 1.3	5.5 $\pm$ 3.8
Av. pH-CaCl <sub>2</sub> ( $\pm$ SD)	4.2 $\pm$ 0.4	4.0 $\pm$ 1.0	3.3 $\pm$ 0.4
Av. Ellenberg N-indication value ( $\pm$ SD) <sup>b</sup>	5.1 $\pm$ 1.0	6.7 $\pm$ 0.6	4.0 $\pm$ 1.7
Av. hours direct sunshine (October, $\pm$ SD)	6.9 $\pm$ 4.2	3.0 $\pm$ 2.9	0
Exposure	Open, bordered by fields	Open to shady, bordered by fields, forest at one side or in forests	Shady, situated in forests

<sup>a</sup> Soil profile: all disturbed sandy soils with variable organic layers

<sup>b</sup> Nitrogen availability-indicating values are assigned to plant species according to Ellenberg (1979). The plots are character-

ized by the median of these values of the occurring plant species. The presented averages are the averages of the medians of the plots that belong to a phytocoenological type

The nomenclature of phanerogams is according Heukels and Van der Meijden (1983), of bryophytes according to Margadant and During (1982), and of fungi mainly according to Kreisel (1987). A complete list of fungi and taxonomic and nomenclatural notes on critical and rare species are presented in Keizer (1993). The names of vegetation types in roadside verges are according to those presented in Keizer (1993) and are only preliminary, without official syntaxonomic status.

## Results

The occurrence of ectomycorrhizal fungi in plots belonging to different age classes of the *Hypochaeris radicata-Quercus* type is presented in Table 2. No ectomycorrhizal fungi were found in plot Q54.

For comparison, data on the average maximum sporocarp numbers in the *Anthriscus sylvestris-Quercus* and *Mnium hornum-Quercus* types of roadside verges are included. These types only comprise plots with trees over 50 years old. In addition, data are presented on the average maximum numbers of sporocarps of all species, the average numbers of species per plot, and the average dry weights of sporocarps.

The number of ectomycorrhizal species in the *Hypochaeris radicata-Quercus* type was found to increase significantly with increasing tree age. The average number of species in old plots of this type was much larger than in plots of the *Anthriscus sylvestris-Quercus* and *Mnium hornum-Quercus* types with old trees. The number of sporocarps was much higher in medium-old than in young plots, but lower in the old plots of the *Hypochaeris radicata-Quercus* type. The latter plots produced more sporocarps than the *Anthriscus sylvestris-Quercus* and *Mnium hornum-Quercus* types, but the differences were less pronounced than for numbers of species.

The relationship between tree age and number of ectomycorrhizal species in the plots of the *Hypochaeris radicata-Quercus* type is illustrated in Fig. 2. Although the variation is rather large, there is a significant, positive correlation between tree age and numbers of species ( $r=0.569$ ;  $n=26$ ,  $P<0.01$ ). The large variation is mainly due to the considerable heterogeneity of this vegetation type.

The relationship between tree age and total numbers of sporocarps is indicated in Fig. 3. The variation is very large indeed and no significant correlation between these characteristics is found. The higher averages in medium-aged plots can be mainly attributed to two plots with very high sporocarp counts.

The species in Table 2 are arranged according to their preference for age classes of trees within the *Hypochaeris radicata-Quercus* type. Six groups of differential species are distinguished.

Two species with a preference for young trees (group I) are *Laccaria tortilis* and *L. proxima*. Four other species, viz *Hebeloma pallidoluctuosum*, *Inocybe lanuginosa*, *Naucoria bohemica*, *Thelophora terrestris*, probably also belong to this group but are listed in group VI (no preference) because of their low frequencies (one out of four investigated plots). *Thelephora*

*terrestris* was found with higher presence-degree in old plots of the *Mnium hornum* type.

Five species are differential for young and medium-aged trees compared to old trees, viz *Hebeloma mesophaeum*, *Inocybe lacera*, *I. lanuginella*, *Laccaria bicolor* and *Scleroderma areolatum* (group II).

A group of 10 species was found most often in plots of medium-aged trees. These 10 fungi include *Amanita muscaria*, *Cortinarius saniosus* and *Chalciporus piperatus* (group III). Some fungal species were present in almost all plots but were most abundant in this age class, viz *Lactarius quietus* and *Laccaria laccata*. However, the latter two species also constantly occurred at high densities in the old plots of the *Anthriscus sylvestris-Quercus* type.

Six species occurred more in plots with medium-aged and old trees within the *Hypochaeris radicata-Quercus* type than in plots with young trees, e.g. *Russula parazurea* and *R. amoenolens* (group IV). Three species of this group had a higher presence-degree and/or abundance in old plots of the *Anthriscus sylvestris-Quercus* type (*Russula parazurea*, *Xerocomus chrysenteron*) or *Mnium hornum* type (*Lactarius theiogalus*).

The largest group of 34 taxa are differential for old trees, among which *Amanita citrina*, *A. rubescens*, *Russula atropurpurea* and *R. nigricans* (group V) are common species. Interestingly, 20 of these taxa were characteristic for the *Hypochaeris radicata-Quercus* type and consequently less frequent in the *Anthriscus sylvestris-Quercus* and *Mnium hornum-Quercus* types, despite the occurrence of trees of comparable age. Only three species in group V are differential for the *Mnium hornum-Quercus* type and none for the *Anthriscus sylvestris-Quercus* type.

Twenty species showed no preference for any age class within the *Hypochaeris radicata-Quercus* type (group VI). *Hebeloma helodes* and *Naucoria bohemica* were the only species with fairly high presence-degrees in this group. The remaining species occurred only in one to three plots of this vegetation type, and no conclusions can be drawn. Thirteen species of this group are considered differential for the *Anthriscus sylvestris-Quercus* or *Mnium hornum-Quercus* type. The numbers of species differential for each group belonging to different genera of fungi are presented in Table 3.

A comparison of the three vegetation types with old trees (Table 2, columns 3–5) also reveals large differences in ectomycorrhizal flora. These differences have been extensively described and discussed by Keizer (1993) and are only summarized in this paper. The average number of species per plot was considerably lower in the *Anthriscus sylvestris-Quercus* type than in the *Hypochaeris radicata-Quercus* type and lowest in the *Mnium hornum-Quercus* type. The numbers of sporocarps were also lower in these types, but the differences to the *Hypochaeris radicata-Quercus* type were less pronounced. Among 80 species studied, the largest group of 27 species had a preference for the *Hypochaeris radicata-Quercus* type. Twelve species are differential for the *Anthriscus sylvestris-Quercus* type

**Table 2.** Average maximum numbers (AMN) of sporocarps per 1000 m<sup>2</sup> and percentage-degrees (%) of ectomycorrhizal fungi in roadside verges planted with *Quercus robur*, belonging to different age classes and vegetation types, in Drente, The Netherlands. Only species occurring in more than two plots were included. The following vegetation types are distinguished: HQ, *Hypochaeris radicata-Quercus* type; AQ, *Anthriscus sylvestris-Quercus* type; MQ, *Mnium hornum-Quercus* type. The following differential

species groups are distinguished within plots with old trees (>50 years old) belonging to different vegetation types: H, differential of the *Hypochaeris radicata-Quercus* type; A, differential of the *Anthriscus sylvestris-Quercus* type; M, differential of the *Mnium hornum-Quercus* type; HA, differential of the *Hypochaeris radicata-Quercus* and the *Anthriscus sylvestris-Quercus* type; AM, differential of *Anthriscus sylvestris-Quercus* and the *Mnium hornum-Quercus* type; O, not differential

Ectomycorrhizal fungus	Age of trees (years), vegetation type										Differential species group
	<20, HQ		20–50, HQ		>50, HQ		>50, AQ		>50, MQ		
	%	AMN	%	AMN	%	AMN	%	AMN	%	AMN	
<b>I. Preference for young trees</b>											
<i>Laccaria proxima</i>	100	221	36	18	50	16	38	11	75	98	O
<i>Laccaria tortilis</i>	50	167	0	0	10	1	0	0	0	0	O
<b>II. Preference for young and medium-aged trees</b>											
<i>Hebeloma mesophaeum</i>	50	5	9	59	0	0	14	3	0	0	A
<i>Inocybe lanuginella</i>	25	14	27	12	0	0	0	0	0	0	O
<i>Inocybe lacera</i>	50	10	36	12	0	0	5	0	25	18	O
<i>Laccaria bicolor</i>	25	11	18	1	0	0	10	1	25	3	O
<i>Scleroderma areolatum</i>	100	10	82	22	40	2	52	8	0	0	HA
<b>III. Preference for medium-aged trees</b>											
<i>Amanita muscaria</i>	0	0	36	23	0	0	0	0	0	0	O
<i>Boletus edulis</i>	0	0	36	40	40	9	10	1	0	0	H
<i>Chalciporus piperatus</i>	0	0	27	4	0	0	0	0	0	0	O
<i>Clavulina coralloides</i>	0	0	64	66	20	44	33	74	25	59	O
<i>Clitopilus prunulus</i>	0	0	27	19	20	1	5	0	0	0	H
<i>Cortinarius violilamellatus</i>	0	0	9	275	10	4	0	0	0	0	O
<i>Cortinarius saniosus</i>	0	0	55	47	40	7	10	1	0	0	H
<i>Laccaria laccata</i>	100	108	100	380	90	115	90	429	100	117	A
<i>Lactarius quietus</i>	25	2	91	516	100	153	95	63	100	139	O
<i>Russula pectinatoides</i>	0	0	45	29	20	8	38	8	0	0	HA
<b>IV. Preference for medium-aged or old trees</b>											
<i>Cortinarius flexipes</i>	0	0	18	2	30	4	19	2	0	0	HA
<i>Cortinarius erythrinus</i>	0	0	27	23	40	13	19	3	0	0	H
<i>Inocybe mixtilis</i>	0	0	27	8	20	5	5	0	0	0	H
<i>Lactarius theiogalus</i>	0	0	18	53	30	21	43	17	75	223	M
<i>Russula amoenolens</i>	25	1	82	70	100	59	90	33	0	0	HA
<i>Russula parazurea</i>	25	2	55	19	60	22	100	30	50	18	H
<i>Xerocomus chrysenteron</i>	0	0	27	1	50	3	71	10	25	1	A
<b>V. Preference for old trees</b>											
<i>Amanita citrina</i>	0	0	0	0	60	8	33	2	25	2	H
<i>Amanita rubescens</i>	0	0	18	5	90	21	71	4	100	16	O
<i>Amanita fulva</i>	0	0	0	0	60	2	14	1	50	4	O
<i>Cantharellus cibarius</i>	0	0	0	0	40	19	10	2	0	0	H
<i>Cortinarius privignus</i>	0	0	0	0	20	20	0	0	0	0	H
<i>Cortinarius striaeopilus</i>	0	0	27	6	50	87	29	82	25	3	HA
<i>Cortinarius subserotipes</i>	0	0	0	0	20	22	5	2	0	0	H
<i>Cortinarius hinnuleus</i>	0	0	9	0	50	42	14	3	0	0	H
<i>Cortinarius obtusus</i>	0	0	0	0	20	2	5	0	0	0	O
<i>Cortinarius paleaceus</i>	0	0	27	5	30	124	33	21	25	1	H
<i>Hebeloma longicaudum</i>	25	10	9	17	40	11	0	0	0	0	H
<i>Hebeloma truncatum</i>	0	0	0	0	20	16	0	0	0	0	H
<i>Inocybe assimilata</i>	0	0	9	1	20	2	29	5	25	7	HA
<i>Inocybe napipes</i>	0	0	0	0	20	2	10	1	50	4	M
<i>Laccaria amethystea</i>	0	0	9	23	40	163	33	25	0	0	H
<i>Lactarius serifluus</i>	0	0	0	0	50	44	38	19	0	0	H
<i>Lactarius chrysorrhoeus</i>	0	0	9	3	60	77	24	15	0	0	H
<i>Otidea bufonia</i>	0	0	0	0	20	4	10	10	0	0	HA
<i>Paxillus involutus</i>	0	0	9	1	30	12	48	27	100	82	M
<i>Russula vesca</i>	0	0	0	0	30	1	19	2	0	0	HA
<i>Russula graveolens</i>	0	0	0	0	60	30	24	10	0	0	H
<i>Russula nigricans</i>	0	0	0	0	100	118	52	11	0	0	H
<i>Russula cyanoxantha</i>	0	0	9	1	40	7	33	3	25	1	O
<i>Russula graveolens f. purp.</i>	0	0	0	0	20	1	14	1	0	0	O

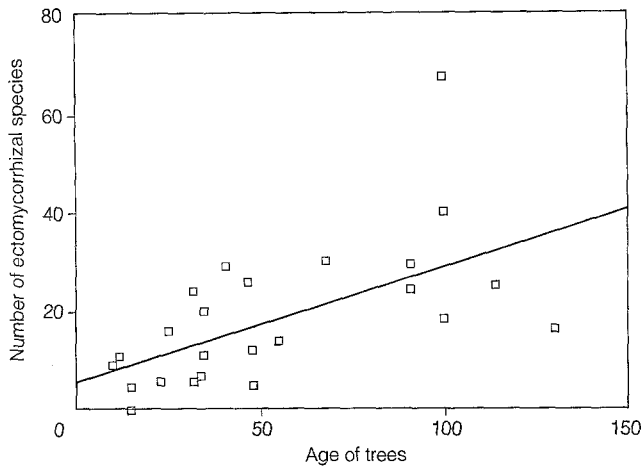
Table 2 (continued)

Ectomycorrhizal fungus	Age of trees (years), vegetation type										Differential species group
	<20, HQ		20-50, HQ		>50, HQ		>50, AQ		>50, MQ		
	%	AMN	%	AMN	%	AMN	%	AMN	%	AMN	
<i>Russula fragilis</i>	0	0	73	11	100	69	43	8	25	2	H
<i>Russula velenovskyi</i>	0	0	0	0	40	5	10	5	0	0	H
<i>Russula graveolens f. cica.</i>	0	0	0	0	30	7	14	1	0	0	H
<i>Russula atropurpurea</i>	0	0	0	0	80	19	43	9	50	3	H
<i>Russula ochroleuca</i>	0	0	9	1	30	2	29	1	100	147	M
<i>Russula odorata</i>	0	0	9	0	70	8	5	0	0	0	H
<i>Scleroderma citrinum</i>	0	0	18	3	60	7	38	9	100	312	O
<i>Tricholoma saponaceum</i>	0	0	0	0	20	4	5	2	0	0	H
<i>Tricholoma sulphureum</i>	0	0	0	0	20	1	5	12	0	0	H
<i>Xerocomus badius</i>	0	0	9	1	60	8	38	6	75	10	O
VI. No preference or rare species											
<i>Amanita spissa</i>	0	0	0	0	10	2	10	3	0	0	O
<i>Cortinarius umbrinolens</i>	0	0	18	1	10	3	0	0	0	0	O
<i>Cortinarius anomalus</i>	0	0	9	1	10	1	10	0	0	0	O
<i>Cortinarius lanatus</i>	0	0	0	0	0	0	19	4	25	5	AM
<i>Cortinarius helveolus</i>	0	0	0	0	10	6	10	19	0	0	A
<i>Hebeloma pallidoluctuosum</i>	25	5	0	0	10	1	0	0	0	0	O
<i>Hebeloma helodes</i>	25	25	36	54	30	23	29	6	0	0	H
<i>Inocybe griseoililacina</i>	0	0	9	1	20	8	5	92	0	0	HA
<i>Inocybe lanuginosa</i>	25	5	9	5	10	0	0	0	0	0	O
<i>Inocybe petiginosa</i>	0	0	0	0	10	10	14	35	0	0	A
<i>Inocybe maculata</i>	0	0	0	0	0	0	19	18	0	0	A
<i>Inocybe albomarginata</i>	0	0	0	0	10	1	19	7	0	0	A
<i>Lactarius camphoratus</i>	0	0	0	0	20	1	19	7	50	8	M
<i>Leotia lubrica</i>	0	0	9	73	10	1	5	1	25	7	O
<i>Naucoria bohemica</i>	50	6	64	9	30	3	0	0	0	0	H
<i>Pseudocraterellus sinusus</i>	0	0	0	0	0	0	14	31	0	0	A
<i>Russula ionochlora</i>	0	0	0	0	0	0	29	3	0	0	A
<i>Russula grisea</i>	0	0	0	0	0	0	19	1	0	0	A
<i>Russula laurocerasi</i>	0	0	0	0	0	0	14	2	0	0	A
<i>Russula emetica</i>	0	0	0	0	0	0	5	0	50	5	M
<i>Thelephora terrestris</i>	25	14	9	1	10	0	5	0	50	17	M
<i>Xerocomus rubellus</i>	0	0	0	0	10	0	14	3	0	0	A
<i>Xerocomus porosporus</i>	0	0	0	0	0	0	19	2	0	0	A
Age of trees (years), vegetation type											
	<20, HQ		20-50, HQ		>50, HQ		>50, AQ		>50, MQ		
Number of plots	4		11		10		21		4		
Av. no. of species per plot	8		15		28		20		16		
Av. no. of sporocarps per plot	614		1932		1619		1264		1324		

(often relatively weak) and only seven species for the *Mnium hornum-Quercus* type. The remaining 34 species are either not differential (24 species) or differential for a combination of two types.

In the present study, sporocarps of ectomycorrhizal fungi were indicated as spots on detailed maps during each visit. Calculations were made of the average sporocarp distance for some selected species (belonging to early- and late-stage fungi and present in sufficiently large numbers) to the nearest tree and to the margin of the pavement in one plot (plot Q1, Oranjekanaal near Odoornerveen) with trees 100 years old in 1986 (Table 4). The average distances from the trees was indeed

larger for the early-stage fungi *Laccaria laccata* and *Hebeloma helodes* than for the four late-stage fungi studied. However, roadside verges are a habitat liable to deform the patterns to be tested, since the root systems of trees are restricted to a narrow strip between the road and the canal and since an environmental gradient is present perpendicularly to the road. *L. laccata* and *H. helodes* were also found at the greatest distance from the pavement, so that the influence of tree roots is of uncertain significance. It is striking that sporocarps of *Russula odorata* were found very close to the road, a phenomenon also observed in other roadside verges.



**Fig. 2.** The relationship between the number of ectomycorrhizal species and tree age in plots of the *Hypochaeris radicata-Quercus* type



**Fig. 3.** The relationship between the number of sporocarps of ectomycorrhizal fungi and tree age in plots of the *Hypochaeris radicata-Quercus* type

**Table 3.** Numbers of taxa in different genera of ectomycorrhizal fungi belonging to various groups of differential taxa for three age classes of trees in roadside verges planted with *Quercus robur* in Drente, The Netherlands. The group of differential taxa were distinguished in the *Hypochaeris radicata-Quercus* type only, according to Table 1: group I: differential taxa of young trees (<20

## Discussion

### *Changes in numbers of species and sporocarps*

In open roadside verges, the average number of ectomycorrhizal species increased with the age of the trees. Old trees had more differential symbionts than young and medium-aged trees. The numbers of productivity of sporocarps showed a maximum in plots with medium-aged trees. The initial increase can partly be understood in view of the extension of the root system of the trees. After approximately 30 years, the soil is entirely occupied by fine roots (Keizer, unpublished observation). Hence, the striking succession of species in later stages cannot be explained by root extension alone.

Our results can be compared with published data on numbers of ectomycorrhizal fungi in forest stands of different age. Ricek (1981) studied ectomycorrhizal macromycetes in 19 stands of *Picea abies* in Germany on former pastures with trees varying in age from 4 to 40 years and summarized his results by indicating a species only if present during this primary succession; no information on presence-degrees or abundance in stands or groups of stands is given. Exact conclusions about species occurrences per stand can, therefore, not be drawn. In stands with trees less than 5 years old, he observed only three ectomycorrhizal species, increasing to six in the age class 5–10 years, 17 in the age class 10–15 years, 29 in the age class 15–20 years (in which canopy closure was usually reached), and up to 48 in the age class 20–25 years. In older stands, the total number of species gradually decreased to 36 in stands of 40–45 years.

Hintikka (1988) compared the ectomycorrhizal fungi in 25 plots (750 m<sup>2</sup>) with *Pinus sylvestris* of different ages in Finland (Table 5). He found the minimal average number of species (10) and sporocarps (179) in young stands (5–15 years), increasing to 21 species and

years); group II: differential taxa of young and medium-aged trees; group III: differential taxa of medium-aged trees (20–50 years); group IV: differential taxa of medium-aged and old trees; group V: differential taxa of old trees (>50 years); group VI: indifferent and rare taxa

	Group(s) of differential species				Total
	I + II	III + IV	V	VI	
Total number of differential taxa	10	16	34	20	80
<i>Laccaria</i>	3	1	1	—	5
<i>Inocybe</i>	3	1	2	4	10
<i>Hebeloma</i>	2	—	2	1	5
<i>Scleroderma</i>	1	—	1	—	2
<i>Boletus, Xerocomus, Chaliciporus</i>	—	3	1	1	5
<i>Lactarius</i>	—	2	2	1	5
<i>Cortinarius</i>	—	4	6	4	14
<i>Amanita</i>	—	1	3	1	5
<i>Russula</i>	—	3	11	4	18
<i>Tricholoma</i>	—	—	2	—	2
Other genera	1	1	3	4	9

**Table 4.** Average distances of sporocarps of various ectomycorrhizal fungi from the trunk of the nearest tree and to the edge of the pavement in the year 1986 in a roadside verge planted with *Quercus robur* 100 years old (in 1988) near Zwiggelte, The Netherlands

	<i>n</i>	Distance from nearest tree (m)	Distance from road surface (m)
Fungi of the canopy closure phase			
<i>Laccaria laccata</i>	330	3.5 ± 0.7	4.5 ± 0.2
<i>Hebeloma helodes</i>	105	3.2 ± 0.5	4.3 ± 0.5
Fungi of the biostatic phase			
<i>Russula odorata</i>	15	3.0 ± 1.3	0.9 ± 0.6
<i>Russula fragilis</i>	126	2.3 ± 0.6	3.3 ± 0.6
<i>Lactarius chrysorrheus</i>	160	2.0 ± 0.9	2.5 ± 0.8
<i>Russula nigricans</i>	27	1.9 ± 0.8	2.6 ± 0.8

**Table 5.** Presence-degree (%) of selected ectomycorrhizal fungi in plots (250 × 3 m) in stands of different ages of *Pinus sylvestris* on sandy soils near Helsinki, Finland (data from Hintikka 1988) and in The Netherlands (data from Termorshuizen 1990)

	Finland				Netherlands			
	No. of plots	Age class (years)	7	8	4	6	14	19
Occurrence mainly near young trees (5–15 years)								
<i>Thelephora terrestris</i>	7	5–15	57	—	—	—	—	—
<i>Laccaria laccata</i> s.l.	7	5–15	100	62	25	—	100	88
Occurrence mainly near (young and) rather young trees (20–30 years)								
<i>Inocybe</i> sp.	8	20–30	29	25	—	—	—	—
<i>Suillus luteus</i>	8	20–30	14	37	—	—	—	—
<i>Gomphidius roseus</i>	8	20–30	29	50	—	—	38	—
Occurrence mainly near young to medium-aged trees (30–50 years)								
<i>Amanita muscaria</i>	4	30–50	71	62	100	16	38	—
<i>Paxillus involutus</i>	4	30–50	43	37	50	—	100	56
Occurrence mainly near (rather young and) medium-aged trees (20–50 years)								
<i>Boletus edulis</i>	6	20–50	—	37	25	—	—	—
<i>Cortinarius semisanguineus</i>	6	20–50	—	75	50	16	75	—
<i>Tricholoma auratum</i>	6	20–50	—	75	50	16	—	—
<i>Hygrophorus hypothejus</i>	6	20–50	—	75	50	—	25	—
<i>Rozites caperata</i>	6	20–50	—	75	50	16	—	—
<i>Russula xerampelina</i>	6	20–50	29	75	75	33	—	—
Occurrence mainly near medium-aged and old trees (>70 years)								
<i>Suillus variegatus</i>	6	>70	43	100	100	83	25	—
<i>Russula emetica</i>	6	>70	—	25	25	16	13	13
<i>Russula vinosa</i>	6	>70	14	100	100	100	—	—
<i>Russula paludosa</i>	6	>70	—	62	50	83	—	—
<i>Russula decolorans</i>	6	>70	14	50	100	100	—	—
<i>Cortinarius collinius</i>	6	>70	14	37	75	83	—	—
No preference for age class								
<i>Lactarius rufus</i>	19		86	100	100	100	100	31
<i>Suillus bovinus</i>	19		86	75	75	67	75	13

470 sporocarps in stands of between 20 and 30 years old, and gradually decreasing again to 13 species and 208 sporocarps in stands older than 70 years.

Jansen (1991) investigated ectomycorrhizal fungi in 25 plots (1000 m<sup>2</sup>; 500 m<sup>2</sup> in seven plots in young stands) in plantations of the introduced *Pseudotsuga menziesii* (Mirb.) Franco in the Netherlands. The average numbers of species (16) and sporocarps (5500) were highest in young plots before canopy closure (age 8–18 years) and strongly decreased in older stands, nine species and 560 sporocarps in stands between 20 and 36 years old, eight species and 280 sporocarps in stands between 41 and 54 years old. This author also

observed a strong reduction of the numbers of ectomycorrhizal root tips (from an average of 128/100 cm<sup>3</sup> in young stands to 30/100 cm<sup>3</sup> in old stands) and mycorrhizal frequency (from 70% in young stands to 8% in old stands).

The results of these three investigations all show the same trends, i.e. an initial increase in species numbers and probably also of sporocarp numbers until a maximum is reached at an age of 30–40 years in the studied tree species, and a subsequent decrease.

Termorshuizen and Schaffers (1991) studied a total of 33 plots (1050 m<sup>2</sup>) in planted forests of *Pinus sylvestris* on sandy soils. They found trends comparable to



the preceding authors in plantations of the first and second rotation: in young stands of the first rotation (4–10 years), the average number of species was six, of sporocarps 1190; in old stands (50–80 years) these figures were three and 120, respectively. In young stands (6–11 years) of second or third rotation, an average of 11 species was found with 1860 sporocarps per plot; in old stands (50–80 years), three species with 390 sporocarps. Some 60-year-old, slowly growing pine forests in coastal dunes on sand poor in humus differed greatly, containing an average of 11 species and producing an average of 2720 sporocarps. They also found a negative correlation between the numbers of mycorrhizas and tree age, but the mycorrhizal frequency only varied between 97 and 100%.

The steady increase in the number of ectomycorrhizal species with the age of the trees found in roadside verges with *Quercus* was not observed in the studies mentioned above. The results also conflict with the model for ectomycorrhizal succession proposed by Dighton and Mason (1985) that predicts an increase of species diversity until canopy closure and a strong decrease thereafter. The possible reasons for this discrepancy are discussed below.

#### *Changes in species composition*

Our observations on ectomycorrhizal fungi in road verges with planted oaks of different age can be partly explained using the concepts of early- and late-stage fungi. The five species found to be differential for young stands (<20 years) are all considered to be early-stage fungi (Dighton and Mason 1985). Most other species in these stands also belong to this category but are also present in road verges with older trees and in older forest stands, e.g. *Laccaria laccata* (Dighton et al. 1986). Only three typical late-stage fungi were found in young plots, each of them in only one plot at a very low density: *Russula parazurea*, *R. amoenolens* and *Lactarius quietus*. In addition, two species regularly found in young plots have not yet been classified as early- or late-stage, viz *Scleroderma areolatum* and *Naucoria bohemica*. In view of their occurrence in the field, they may very well belong to the group of early-stage fungi (in the sense of Dighton and Mason 1985).

A noticeable difference between ectomycorrhizal succession in *Betula* and *Quercus* is the duration of different phases. Early-stage fungi are dominant with *Quercus* in trees up to 20 years old and with *Betula* in trees younger than about 6 years (Last et al. 1987). It is quite possible that this phenomenon is related to the longevity of the trees: less than 100 years for *Betula* and over 1000 years for *Quercus*. The predominance of early-stage fungi with *Quercus* trees of 10–20 years may also be caused by the maintenance of ectomycorrhizal fungi already present in the tree nursery and transported on the tree roots to the roadside verge. Dighton et al. (1986) also found in stands of *Pinus contorta* and *Picea sitchensis* in Northumberland (Great Britain)

that sporocarps of early-stage fungi were dominant in slightly older stands (4–10 years) than in previous experiments with *Betula*. He suggested that this difference depended on both the tree species and the environmental conditions.

Two early-stage fungi, *Laccaria proxima* and *Thelephora terrestris*, were present outside of the young *Hypochaeris radicata-Quercus* stands in old plots of the *Mnium hornum-Quercus* type and with comparable frequency. This suggests that ectomycorrhizal succession is not (only) dependent on the age of the trees but also on soil conditions (Mason et al. 1987; see below).

Roadside verges with medium-old and old oak trees were dominated by late-stage fungi (Tables 2 and 3). However, boletes were most often found with medium-old trees, most *Russula* and *Amanita* species with old trees. The distinction between early- and late-stage fungi can often not be made at the generic level. For instance, most *Laccaria* species are typical early-stage fungi but *L. amethystea* is differential for old trees in our plots; *Inocybe lanuginella* and *I. lacera* were already present in young stands, but *I. napipes* and *I. assimilata* grew exclusively in old stands (Table 1). The succession in stands of *Betula* has only been investigated during the first 15 years (e.g. Last et al. 1987). Dighton et al. (1986) analysed the succession in stands of *Pinus contorta* and *Picea sitchensis* up to a stand age of 27 years. It is evident that ectomycorrhizal development is not restricted to younger stands, at least of *Quercus robur*. The largest number of differential species are conspicuously characteristic of roadside verges with trees over 50 years old. Likewise, species characteristic for oak trees older than 130 years may exist but this age class has not so far been investigated. Considerable ecological, and possibly physiological, differentiation apparently exists within the group of late-stage fungi.

Other data confirm the prolonged succession of ectomycorrhizal fungi after the establishment of late-stage fungi. Data by Hintikka (1988), rearranged by the present authors into groups similar to those for the roadside verges with *Quercus*, are illustrative of the succession in forests of *Pinus sylvestris* on poor, sandy soils (Table 5). Species of *Laccaria*, *Thelephora* and *Inocybe* were important in young stands, *Boletus* and *Suillus* were best represented in rather young and medium-old stands, whereas *Russula* species dominated medium-old and old stands. Species characteristic for stands older than 70 years were reported. Roadside verges with *Quercus* and *Pinus* forests have only a few species in common. Some species show a similar preference, for instance *Thelephora terrestris* for young, *Boletus edulis* for medium-old and *Russula emetica* for old trees. However, striking differences exist: *Laccaria laccata* (sensu lato) is frequent under old oak trees but absent from old pine forest; *Amanita muscaria* and *Paxillus involutus* are present in many young pine forests but in road verges with *Quercus* they are characteristic for medium-aged and old trees, respectively.

Comparison of data on the presence-degree of species found by Hintikka (1988) and observations by Termorshuizen (1990) in young and old stands of *Pin-*

*us sylvestris* on sandy soils in the Netherlands (Table 5, columns 5 and 6) shows that many species characteristic for older stands in Finland behave quite differently in the Netherlands. In the latter country, some are even restricted to young forests, e.g. *Cortinarius semisanguineus* and *Suillus variegatus*. On the other hand, *Laccaria laccata* sensu lato was present in most old stands in the Netherlands.

Comparable results were obtained by Jansen (1991) for stands of *Pseudotsuga menziesii* in The Netherlands. The early-stage fungi *Laccaria proxima* and *Thelephora terrestris* were observed in most stands of all age classes (from 8 to 54 years), whereas late-stage fungi such as *Cortinarius semisanguineus*, *Paxillus involutus*, *Russula ochroleuca* and *Xerocomus badius* were present in at least 70% of the stands younger than 20 years.

### Changes in weight of sporocarps

The average dry weights of sporocarps of the species found in the three age classes of roadside verges of the *Hypochaeris radicata-Quercus* type were determined in order to test the conclusion of Dighton and Mason (1985) that sporocarps of early-stage fungi are usually smaller than those of late-stage fungi.

The average dry weights of sporocarps of the species encountered in roadside verges with *Quercus* were  $0.3 \pm 0.2$  g in young plots,  $4.5 \pm 11.8$  g in medium-aged plots and  $1.8 \pm 0.9$  g in old plots (Table 2). Only the difference between young and old plots was significant ( $P < 0.01$ , *t*-test). The high value and very large standard deviation in plots with medium-aged trees is mainly due to the occurrence of *Boletus edulis* with heavy sporocarps in some of these plots. Our results agree with the observations of Dighton and Mason (1985). However, the conclusion that sporocarps in young stands are on average lighter has no general validity. Termorshuizen (1991) found the highest average sporocarp weight in young *Pinus sylvestris* (4–14 years) stands in the Netherlands. Therefore, the concepts of early-stage fungi and fungi growing in young stands are not identical.

### Conclusions

From our investigation of roadside verges with oak trees of different ages and other studies on succession of ectomycorrhizal fungi in different habitats, it is evident that the course of this succession is not uniform in different habitats.

Probably the most usual process in forest stands is a fairly rapid increase in species richness and sporocarp productivity during approximately 30–40 years and a more gradual decrease afterwards to an intermediate, rather constant level. In the beginning, the ectomycorrhizal flora is dominated by early-stage fungi, which to a large extent are gradually replaced by late-stage fungi,

some late-stage fungi being more characteristic for rather young or medium-aged stands, others of old ones. This process was observed in pine forests in Finland (Hintikka 1988; Table 5).

However, in the Netherlands maximum species richness and sporocarp productivity in coniferous forests are reached in young stands up to approximately 15 years old. Part of the early-stage fungi also live in mature stands, whereas some late-stage species, found in older stands elsewhere, are mostly found in young stands in the Netherlands (Jansen 1991; Termorshuizen 1991).

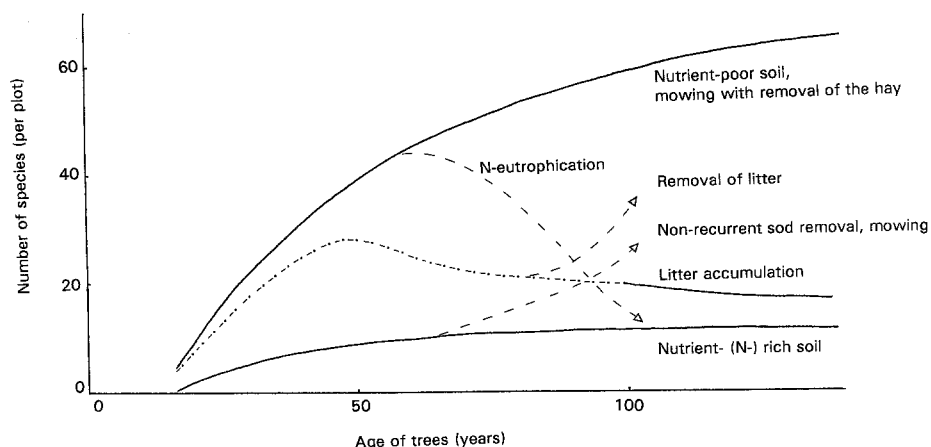
On the contrary, in roadside verges planted with *Quercus*, the species richness steadily increased with the age of the trees and the sporocarp abundance remained at a high level. Some early-stage fungi were confined to roadside verges with young trees, others were also present near old trees. Among the late-stage fungi, a strong differentiation was observed in connection with the age of their partner trees.

These observations do not support the conclusions of Gibson and Deacon (1988), who suggested that succession of ectomycorrhizal fungi is primarily determined by the age of the tree root system. Alvarez et al. (1979) demonstrated that in the presence of an organic layer over the mineral soil growth and survival of *Abies concolor* seedlings was worse than in soil lacking this layer. They attributed this to an inhibitory effect of the organic layer on mycorrhizas. Thus soil factors may play a decisive role in the course of succession, as suggested by Mason et al. (1987).

Environmental conditions in coniferous forests in the Netherlands mainly differ from those in Finland in the high deposition of acidifying substances and nitrogen (on average  $50 \text{ kg N ha}^{-1} \text{ year}^{-1}$ ; Erisman and Heij 1991) and a probably related accumulation of coarse, slowly decaying litter (Kuyper 1989; Jansen 1991). These factors are thought to be responsible for a strong reduction of sporocarp production in mature stands (Arnolds 1991) and an acceleration of ectomycorrhizal succession, so that some fungi normally found in mature stands now occur in young stands.

The high level of air pollution can also directly influence the nutritional status of trees, causing a change in the allocation pattern and/or quantity of carbohydrates in the tree. This might result in a lower availability of carbohydrates in the roots and consequently a changed species composition and a lower sporocarp production in some species (Termorshuizen 1990).

Roadside verges with oak trees in open landscapes in the Netherlands mainly differ from forest stands in the absence of a litter layer. Like forests, they are exposed to "acid rain", but the fertilizing effects of the litter are, at least in part, compensated by removal of litter by the wind. Furthermore, the sward is removed from a part of the roadside verges by mowing. The absence of a litter layer and/or loss of nitrogen from the system would thus lead to an extended succession in which early-stage fungi tend to maintain themselves longer and numerous late-stage fungi appear in the course of the succession.



**Fig. 4.** A model indicating the probable course of succession of ectomycorrhizal fungi in roadside verges with ageing trees under various circumstances. —, Observed; — · —, inside forests, after Dighton and Mason (1985); ---, probable course

However, the importance of nitrogen and litter and their interaction are not yet sufficiently known. It has been experimentally demonstrated that the use of nitrogen fertilizers reduces the species richness of ectomycorrhizal fungi in roadside verges (Keizer 1993), which is in good agreement with the effect of fertilizer application in forest (Kuyper 1989; Termorshuizen 1992). It has also been demonstrated that removal of sods in forests leads to an increase in sporocarp numbers of ectomycorrhizal fungi (Baar and Kuyper 1993). In addition, roadside verges form a relatively dynamic habitat where periodical riding, trampling and road (re)construction may locally create "pioneer" circumstances. In such places, species usually confined to early successional stages can presumably either maintain themselves or re-establish themselves repeatedly.

In view of the above, it is probable that a negative effect of N deposition on ectomycorrhizal fungi acts via a change in soil properties. In this context it is interesting that shady roadside verges with old trees situated in forests (*Mnium hornum-Quercus* type), where litter accumulation is only slightly reduced relatively to the forest itself and usually no management is carried out, on average contained only half the number of ectomycorrhizal species found in the open, exposed *Hypochaeris radicata-Quercus* type (Table 2). In addition, the *Mnium hornum-Quercus* type had few differential species and its ectomycorrhizal flora was much more similar to that of typical *Quercus* forests (Keizer 1993).

Some mature forest types are also reported to be extremely rich in ectomycorrhizal fungi, e.g. oak forests on acid sand dunes (*Dicrano-Quercetum*) in the Netherlands (Jansen 1984), pine forests on sand dunes (*Cladonio-Pinetum*) in eastern Germany (Sammler 1988; Wöldecke and Wöldecke 1990), and beech forest on limestone slopes (*Carici-Fagetum*) in western Germany (Jahn 1986). These forest types have only a few features in common, in particular their occurrence in hilly areas where part of the litter and nutrients are removed from the system by natural causes and only a thin organic top soil is developed, and that the forest floor is overgrown mainly with mosses and only a few herbaceous plants. Many characteristic species of these

forest types are differential species for roadside verges with old trees in open landscapes, e.g. *Russula graveolens*, *Tricholoma saponaceum* and *Lactarius vellereus* (Keizer 1993).

We expect the mycorrhizal flora in the *Hypochaeris radicata-Quercus* type to change towards that of the *Mnium hornum-Quercus* type if litter and nutrients accumulate for some reason, and thus that species richness and composition change due to soil succession rather than aging of trees.

Dighton and Mason (1985) developed a model of ectomycorrhizal succession in which species richness increases from young to medium-aged stands, then strongly decreases in old stands, to reach a very low final level. This model does not explain the series found by us. At least three different processes must be distinguished in connection with litter accumulation and nitrogen deposition. The different pathways of succession are schematically presented in Fig. 4. According to this model, in nutrient-poor soils the species number increases with the age of the trees provided that no accumulation of litter and nutrients takes place. Litter accumulation and eutrophication reduce the species number. On the other hand, nature management measures like litter removal, removal of sods or yearly mowing with removal of the hay may increase the species number in situations with litter accumulation or eutrophication.

The underlying mechanism may be as follows: Young trees need more mineral nutrients (nitrogen) than old trees, and this is rapidly available in young, mineral soils with a low organic matter content. Early-stage fungi (relatively few "pioneer" species) are adapted to these circumstances. As the stand ages, more root substrate and more different (micro)habitats become available. In the course of time, an organic layer develops containing a large proportion of the nutrients. The trees need relatively less nutrients because of a more efficient internal circulation of nitrogen (Cole and Rapp 1981; Chapin 1986). This in combination with the presence of an organic layer will lead to a somewhat lower species number.

Some symbiotic fungi of this stage are capable of releasing nutrients from soils with a high organic mat-

ter content more effectively than the early-stage fungi (Read 1991). However, a wide variation between species must exist in this ability, as many species may be found near old trees in mineral soils (although at these sites probably more organic material is present than in young mineral soils, due to the presence of dead tree and plant roots). The strong decrease in many ectomycorrhizal fungi from aging and old forests in the Netherlands and their maintenance in foreign (old) forests and in the *Hypocharis radicata-Quercus* type of roadside verges indicate that the quality of the organic material in Dutch forests has become unfavourable for many ectomycorrhizal fungi.

The concepts of early- and late-stage fungi are primarily based on physiological characteristics of species and indeed are useful to understand early phases of primary forest succession. However, they are not appropriate to describe ectomycorrhizal succession under field conditions over a longer period since: 1) some early-stage fungi are restricted to young trees but others are maintained on the root systems of old trees (Table 2 group I, II); 2) some late-stage fungi appear already with young trees (Table 5); 3) seedlings near mature trees may be infected by late-stage fungi (Last et al. 1987); 4) late-stage fungi are dominant during some 90–95% of the lifetime of a tree and can be divided into several groups living on root systems of different age classes.

We propose the following schematic classification of ectomycorrhizal species in view of their appearance during forest development. The groups are not primarily based on age classes of trees but on general changes in forest architecture (Oldeman 1990), since the duration of successional stages seems to depend on the maximum lifetime of the tree species and soil fertility in the stand. This classification is based on supposed "typical" succession series and may be altered under the influence of environmental factors (cf. Fig. 4). Many species may occupy an intermediate position or occur during two or more stages (Tables 2, and 5):

1. Ectomycorrhizal fungi of the innovation phase: fungi colonizing root systems of very young trees in the absence of mature trees; low degree of host-specific association (e.g. *Betula* and *Pinus*  $\pm 1$ –5 years; *Quercus*  $\pm 1$ –10 years); e.g. *Thelephora terrestris*, *Laccaria tortilis*)
2. Ectomycorrhizal fungi of the canopy closure phase: fungi mainly growing in open stands of young trees without canopy closure; moderately low degree of host-specific association (e.g. *Betula*  $\pm 5$ –10 years, *Pinus*  $\pm 5$ –15 years; *Quercus*  $\pm 10$ –20 years); e.g. *Suillus luteus*, *Lactarius pubescens*, some *Inocybe* species
3. Ectomycorrhizal fungi of the aggradation phase: fungi mainly growing in stands around canopy closure and some years afterwards; moderately high degree of host-specific association (e.g. *Betula*  $\pm 10$ –25 years, *Pinus*  $\pm 15$ –40 years; *Quercus*  $\pm 25$ –50 years); e.g. *Cortinarius semisanguineus*, *Boletus edulis*
4. Ectomycorrhizal fungi of the late biostatic phase: fungi mainly growing in mature stands; high degree of

host-specific association (e.g. *Betula*  $\pm 25$ –50 years; *Pinus*  $\pm 40$ –100 years; *Quercus*  $\pm 50$ –150 (?) years); e.g. *Amanita rubescens*, *Russula nigricans*, *R. decolorans*

5. Ectomycorrhizal fungi of the degradation phase: fungi mainly growing in stands with senescent trees (e.g. *Betula*  $\pm 50$ –100 years; *Pinus* >100 years; *Quercus* >150 (?) years). Examples unknown, possibly *Podoscypha multizonata* (doubtfully ectomycorrhizal, cf. Jahn and Müller 1976)

6. Persistent ectomycorrhizal fungi: fungi occurring from the innovation phase to phase 3 or 4; low degree of host-specific association, e.g. *Laccaria laccata*

It is evident that changes in species richness and composition during ectomycorrhizal succession are not only influenced by the structure of stands and the age of the trees but also by e.g. the soil conditions, tree species and former land use (primary or secondary succession). The interactions between these factors are complex and still poorly understood. More data on the composition of the ectomycorrhizal flora in stands of different age classes growing in various environments are indispensable to a better understanding of ectomycorrhizal successions. In addition, field and laboratory experiments are needed to unravel the physiological and ecological differences between groups of late-stage fungi appearing at different moments during stand development.

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