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Mycorrhiza

The influence of host plant, nitrogen fertilization and fungicide application on the abundance and seasonal dynamics of vesicular-arbuscular mycorrhizal fungi in arable soils of northern Germany

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Abstract. The present investigation examines whether the crop plant, increased nitrogen (N) fertilization and fungicide application influence the pattern of vesicular-arbuscular mycorrhizal fungi (VAM) populations. For this purpose, two arable field locations in Lower Saxony (Hotteln and Langreder) were chosen and the formation of mycorrhiza, spore density, number of infectious propagules (MPN) and frequency of spore types within VAM populations were investigated. The influence of crop plants was examined over two cultivation periods (1986/1987 and 1987/1988) in Hotteln, comparing winter wheat, winter barley and sugar beet. The effects of increased N fertilization and fungicide application were investigated on winter wheat in Langreder in the cultivation period 1988 only. Both the frequency of mycorrhizal infection and the spore dynamics in soil differed with the crops grown. Spore density and MPN increased until harvesting when host plants (winter wheat, winter barley) were cultivated, whereas both diminished with a non-host plant (sugar beet). Different spore types increased or decreased, according to the plant species grown, but the predominating types of the location remained constant. Increased N fertilization caused marginal inhibition of mycorrhizal infection and sporulation on winter wheat, whereas both leaf and base application of fungicides resulted in minor increases in mycorrhizal colonization of roots and sporulation in soil. Both increased N fertilization and fungicide application distinctly decreased the sporulation of one type in May, but the characteristic compositions of the VAM populations remained unchanged.

Key words: Host plant – Nitrogen fertilization – Fungicide application – Vesicular-arbuscular mycorrhizae – Spore diversity

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Introduction

Many reports document the influence of diverse factors (substrate, host plant, pH, temperature, fertilization, water regime, light, plant protectives) on the occurrence of vesicular-arbuscular mycorrhizae (VAM) under greenhouse conditions (Peuss 1958; Kruckelmann 1975; Schenk and Kinloch 1980; Sieverding 1981; Tommerup and Briggs 1981; Tommerup 1984; Hayman and Tavares 1985). Which of these factors have an impact on VAM populations under field conditions, especially in agricultural soils, has been little investigated.

Previous experiments have shown patterns of VAM populations characteristic of the soil texture (Land and Schönbeck 1991). However, according to the literature cited, mycorrhizal infection of plants and the spore density in soil can be influenced by the crop plant and crop rotation (Winter 1951; Sieverding and Leihner 1984; Daniels Hetrick and Bloom 1986; Harinikumar and Bagyaraj 1989). In addition to crop plants that are intensively colonized by VAM fungi, such as maize or flax, there are also non-mycorrhizal crops. Whether this difference has an influence on the characteristic VAM population in soil, and to what extent, is still unknown.

Furthermore, increased fertilization (Jensen and Jacobsen 1980) and pesticide application, especially herbicides (Sieverding and Leihner 1984; Tommerup and Briggs 1984) and fungicides (Menge 1982), can inhibit VAM fungi. Unbalanced nitrogen (N) fertilization may distinctly inhibit mycorrhizal colonization and the elimination of host plants by application of herbicides can result in a lower inoculum density of VAM fungi. The effect of fungicides, however, depends on the application site and the active substance used. Direct application to soil usually inhibits mycorrhizal colonization (Nemec 1980; Ocampo and Hayman 1980), whereas leaf applications may even result in an increase (Dehne 1985, 1986).

The present study examined the influence of the crop plant, N fertilization and fungicide application on patterns of VAM fungi by monitoring the inocula, root colonization and composition of the mycorrhizal populations.

Table 1. Composition and characteristics of sites investigated

Parameter	Sites	
	Langreder	Hotteln
pH	7.5	7.3
Organic matter (C%)	1.6	3.1
Sand (%)	1.8	4.5
Silt (%)	87.8	82.3
Clay (%)	10.4	12.2
Soil texture	Clayey silt	Clayey silt
Soil type	Gley-like para-brown earth	Degraded black earth

Table 2. Crop rotation of plots investigated at Hotteln

Plot	Crop		
	1985/1986	1986/1987	1987/1988
1	Winter wheat	Winter barley cv. Tapir	Sugar beet cv. Regina
2	Winter wheat	Winter wheat cv. Kanzler	Sugar beet cv. Regina
3	Winter barley	Sugar beet cv. Regina	Winter wheat cv. Rektor

Materials and methods

Sampling sites

Investigations on the influence of the crop plants used were carried out over two cultivation periods (1986/1987 and 1987/1988) near Hotteln (Table 1). During the first cultivation period (1987/1988), winter wheat and winter barley were compared, the previous crop on both plots being winter wheat. In the subsequent cultivation period (1987/1988), sugar beet and winter wheat were planted (Table 2). Fertilizer and pesticides were applied according to usual practice. Investigations on the influence of N fertilization and fungicide application were carried out near Langreder (Table 1), where many soil parameters were similar to those in Hotteln. In the cultivation period 1987/1988, standard N fertilization was compared with increased N fertilization, with and without fungicide application (Tables 3 and 4, respectively). According to the Landwirtschaftliche Untersuchungs- und Forschungsanstalt, Landwirtschaftskammer Hannover (LUFA), the contents of potassium and phosphorus were very high at the beginning and at the end of the cultivation period on both sites. Soil temperature at a depth of 15 cm was continously measured and plotted by a soil thermograph.

Spore extraction and root examination

On five to eight dates during each cultivation period, five samples were taken per date and location from the surface soil layer (0–30 cm). Each sample was prepared for measuring two different parameters: spore number and type classification, and the percentage of mycorrhizal roots.

Spores were isolated by wet sieving (Gerdemann and Nicolson 1963) of the soil samples (38–350 µm) followed by sucrose-density centrifugation (Fang et al. 1983). The supernatant containing the spores was then carefully decanted into a sieve and the spores were washed, transferred into petri dishes and sorted into types under a dissecting microscope. Only intact spores were counted. Classification was based on colour, size, shape, surface structure, general nature of the spore contents and hyphal attachment (Table 5). Because of intersample variation in soil moisture, 20 g of each sample was dried and all spore counts were normalized to 100 g of dry soil to obtain a uniform basis for comparison.

Roots were washed out of 400 ml soil to estimate the percentage of mycorrhizal colonization. The rate of infection was determined microscopically for 100 root segments, each 1 cm long.

The "most probable number" (MPN) was determined by diluting the original soil samples (100 ml) eight times, each time fourfold, with sterilized soil from the same site (Cochran 1950; Alexander 1965). Five replicates were prepared at each level of dilution. Three plants of *Tagetes erecta* were planted in each pot and cultivated for 6 weeks before roots were washed out and stored in alcohol-formaldehyde-acetic acid fixative (Gerlach 1969) until staining with trypan blue (Land 1990) to estimate mycorrhizal infection. The presence or absence of VAM was determined for the whole root system under a dissecting microscope (50×). Calculation of MPN was carried out according to Fisher and Yates (1974).

Data analysis

Data were processed by analysis of variance. Percentage data were arcus-sinus-root transformed (Sachs 1974), and pair comparisons made by a Student's *t*-test ($P \leq 0.05$).

Results

The influence of crop plants on VAM fungi

Mycorrhizal colonization. In the first cultivation period (1986/1987), mycorrhizal colonization in both winter crops began in April (Fig. 1) after a soil temperature of 5° C was reached at a depth of 15 cm (Fig. 2). Infection frequency attained a maximum of 60% in both crops. In winter wheat, the maximum VAM infection was reached

Table 3. N fertilization of winter wheat cv. Kanzler at Langreder during cultivation period 1987/1988

Date of N fertilization	N (kg/ha) as calcium ammonium nitrate	
	Standard	Increased
30 March 1988	93	93
5 May 1988	60	90
10 June 1988	80	110
Total N fertilization	233	293

Table 4. Fungicide applications to winter wheat cv. Kanzler at Langreder during cultivation period 1987/1988

Date	Fungicide application (trade name, active substance)
29 April 1988	1.5 l/ha Sportak, alpha (prochloraz, carbendazim)
19 May 1988	1.0 l/ha Simbo (fenpropimorph, propiconazole)
14 June 1988	0.5 l/ha Bayfidan (triadimenol)
	4.0 l/ha Dyrene (anilazine)

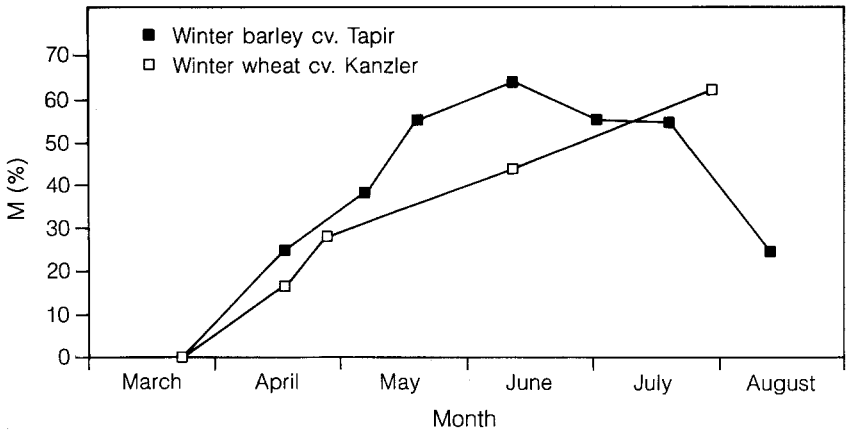

Fig. 1. The frequency of mycorrhizal infection (*M*) at Hotteln in two different crop plants during the cultivation period 1986/1987

Table 5. Spore type classification based on optical attributes and identified species. w, White; y, yellow; h, hyaline; g, gold; br, brown

Type	Colour	Surface	Size (µm)	Characteristics	Identified species
1	w	Smooth	50–100	Granular contents; straight hyphal attachment	*Glomus aggregatum* *G. albidum*
2	w-y	Rough	≥100	Granular contents; hyphal attachment often absent	*G. albidum* *G. aggregatum*
3	h	Smooth	≥150	Thick-walled; hyphal attachment often absent	*G. aggregatum* *G. albidum*
4	br	Smooth	40–130	Hyphal attachment often absent	*G. etunicatum* *G. caledonium*
5	br	Smooth	40–80	Transparent; oval; thick, brown wall	*G. intraradix* *G. fasciculatum*
6	br	Smooth	80–150	Thick-walled	*G. etunicatum* *G. caledonium*
7	g-br	Smooth	100–150	Transparent; straight hyphal attachment	*G. intraradix*
8	g-br	Smooth	100–150	Thick; brown wall; septum	*G. etunicatum* *G. caledonium*
9	y	Smooth	50–100	Thin-walled; transparent; fragile	*G. intraradix* *G. fasciculatum*
10	g-br	Smooth or peridium	≥150	Hyphal attachment straight to funnel shaped; septum; large lipid drops; thick walled	*G. caledonium* *G. mosseae*
11	br	Smooth	40–80	Hyphal attachment absent; reddish resplendent	?
12	y-br	Rough	≥120	Dull; dirty	*G. caledonium* *G. mosseae*

at flowering in June and decreased thereafter until harvest. In contrast, the frequency of mycorrhizal roots in winter barley increased steadily from the beginning of stem elongation (April) until harvesting at the end of July.

Comparison with the second cultivation period (1987/1988) showed that sugar beet as expected was not a host for VAM fungi (Fig. 3). Mycorrhizal colonization of winter wheat cv. Rektor had similar kinetics to those of cv. Kanzler in the previous cultivation period, but the infection level was distinctly lower.

Inoculum density in soil. Apart from germinating chlamydospores, other structures such as mycelium in soil or in roots and vesicles are also able to infect plant roots. Therefore, both the number of spores and MPN were determined in the vegetation period 1988. As for mycorrhizal infection, the spore density was measured on five to eight dates during the vegetation period in order to investigate the population dynamics. However, MPN was determined only at the beginning and the end of the vegetation period 1988.

Spore densities increased markedly from sowing until harvesting with winter wheat and winter barley. The dynamics of spore density for both crops differed between cultivation periods, as did mycorrhizal infection. During the cultivation period 1986/1987 (Fig. 4), spore densities remained unchanged until stem elongation in May. For winter barley they then decreased and increased again towards the end of the cultivation period; in winter wheat, spore density showed a steady increase from April to harvesting in August. Nevertheless, both plots contained similar numbers of spores at harvesting, independent of the dynamics during the vegetation period.

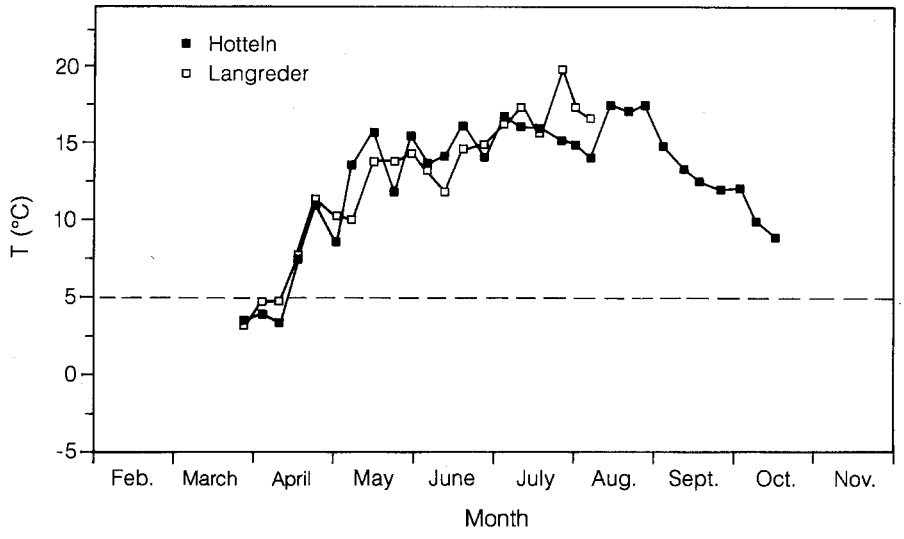

Fig. 2. Weekly average soil temperature (T) at a depth of 15 cm for the two different field locations during the cultivation period 1988

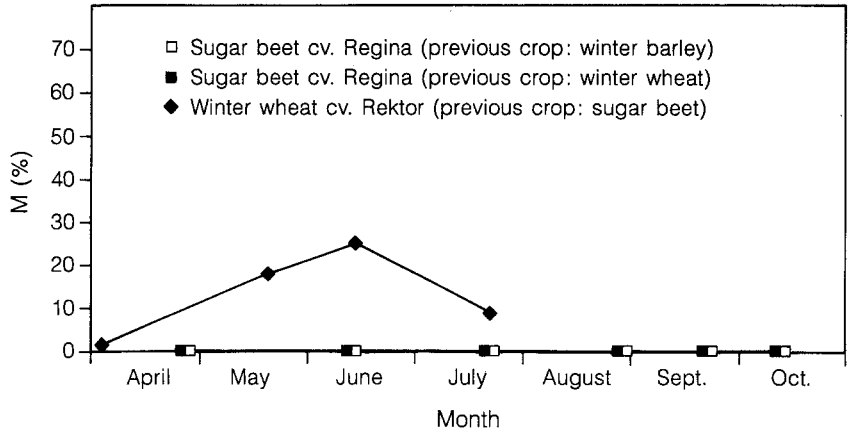

Fig. 3. The frequency of mycorrhizal infection (M) at Hotteln for two different crop plants during the cultivation period 1987/1988

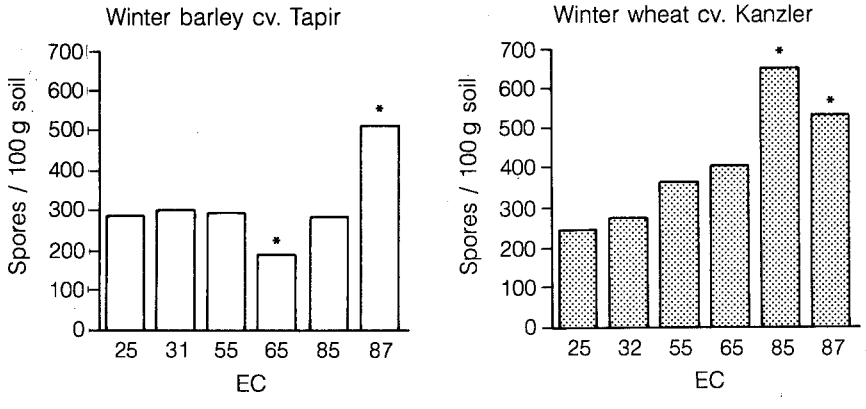

Fig. 4. Vesicular-arbuscular mycorrhizal (VAM) spore density in winter barley cv. Tapir and winter wheat cv. Kanzler at Hotteln during the cultivation period 1986/1987. *EC*, Eucarpia stages of development (after Zadoks et al. 1974). *Asterisks* indicate spore densities significantly different from that at EC 25

Crops were rotated in the following cultivation period (1987/1988) and sugar beet was cultivated on the experimental plots. Therefore, a third plot was added for cultivating winter wheat. The spore number decreased by about 70% from sowing until harvesting in sugar beet (plot 1 in Fig. 5). In winter wheat, sporulation increased especially towards the end of cultivation (Fig. 5), showing the same trend as the winter wheat of the previous cultivation period (Fig. 4). The number of infectious units increased considerably from sowing to harvesting (Table 6). Only a rather minor proportion of total infectious structures were chlamydospores at the end of the cultivation period. In contrast, spore density and MPN decreased during the cultivation of sugar beet and the number of spores at harvesting corresponded almost with the total infectious units.

Distribution of chlamydospore types within VAM populations. Both the level of mycorrhizal colonization and the kinetics of changes in spore density in the soil varied

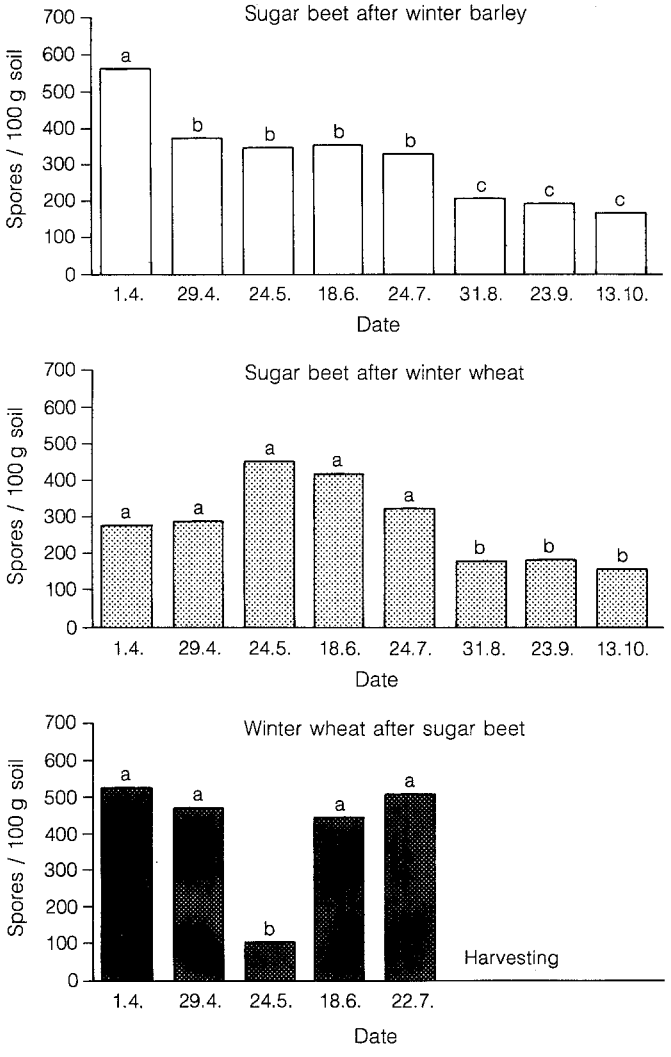

Fig. 5. VAM spore density in soil of two different cultures at Hotteln during the cultivation period 1988. *a*, *b*, *c* indicate significantly different spore densities

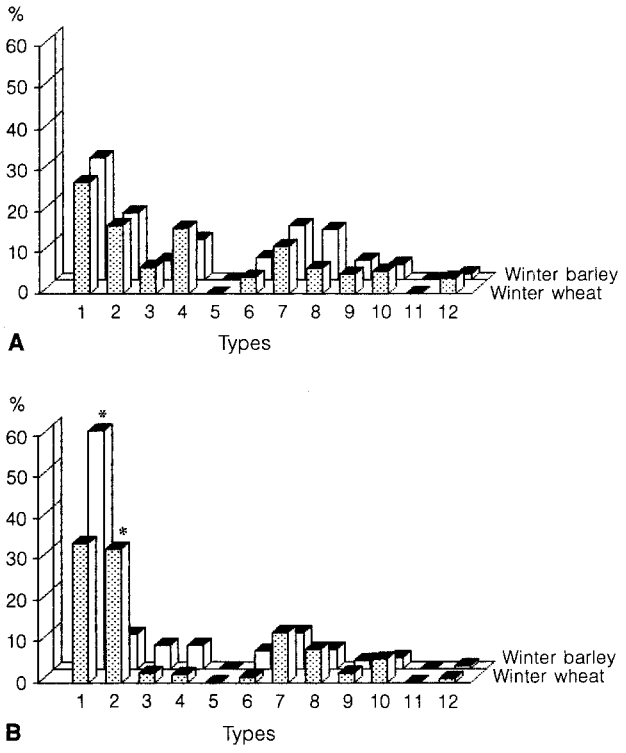

Fig. 6. Percentage of spore types within the VAM population present in each of two plots at Hotteln cultivated with different crops at the beginning (**A**) and at the end (**B**) of the cultivation period 1986/1987. *Asterisks* indicate significant differences in abundance of one type between the two crops

in relation to the plants cultivated. The object here was to investigate whether this resulted in different compositions of VAM populations.

The distribution of VAM types within populations of winter wheat and winter barley were similar at the beginning of the cultivation period 1986/1987. Spore types 1, 2 and 7 were most abundant (Fig. 6A). The same types were predominant at harvesting, but the percentage of type 2 within the population significantly decreased while that of type 1 distinctly increased in winter barley (Fig. 6B). The increase of type 1 in winter barley at the end of cultivation period 1987/1988 was based on completely different spore number kinetics during the vegetation period, with higher sporulation at harvesting than for winter wheat. In contrast, the decrease of type 2 in winter barley was based on a lower sporulation rate during the whole vegetation period than for winter wheat (Fig. 7).

During the cultivation period 1987/1988, sugar beet as a non-host of mycorrhizal fungi provided the opportunity to study viability of different spore types. At the beginning of the cultivation period, significant differences between sugar beet and winter wheat were apparent for types 1 and 7 (Fig. 8A). At harvesting, type 1 drastically decreased, whereas the percentages of types 2, 3 and 7 within the population, but not the total spore number of these types, increased in sugar beet (Fig. 8B). In winter wheat, only the sporulation of type 7 de-

Table 6. Most probable number (MPN) and spore density in spring and autumn 1988 on two plots at Hotteln

Crop rotation	Field	Spring		Autumn	
		MPN (per 100 g soil)	Spores (per 100 g soil)	MPN (per 100 g soil)	Spores (per 100 g soil)
Winter wheat/sugar beet	2	1854	278	649	156
Sugar beet/winter wheat	3	511	526	22058	507

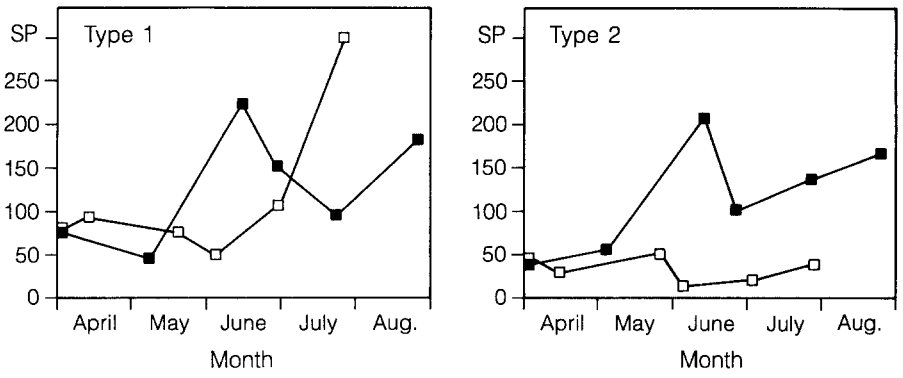

Fig. 7. Number of spores of type 1 and 2 during the cultivation period 1986/1987 in winter wheat and winter barley at Hotteln. *SP*, Spores/100 g dried soil. □, Winter barley cv. Tapir; ■, winter wheat cv. Kanzler

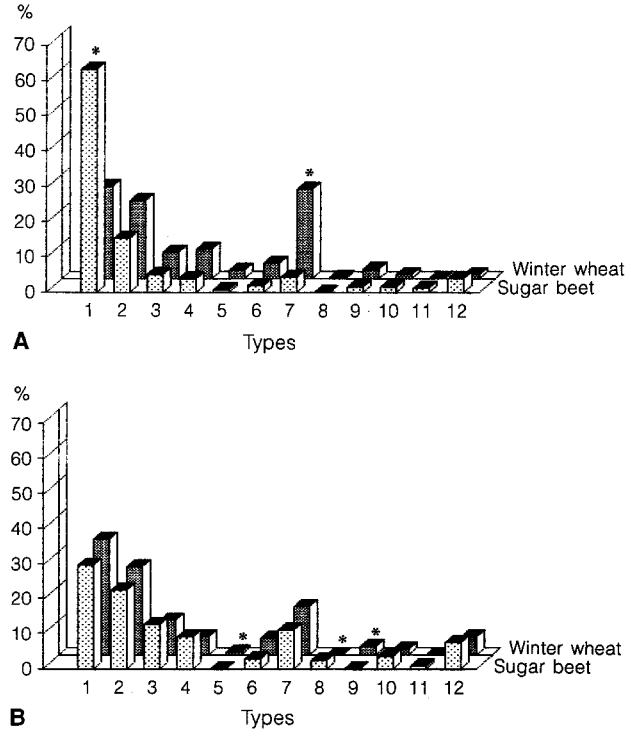

Fig. 8. Percentage of spore types within the VAM population present in each of two plots at Hotteln cultivated with different crops at the beginning (**A**) and at the end (**B**) of the cultivation period 1987/1988. *Asterisks* indicate significant differences in abundance of one type between the two crops

creased, while the proportions of the other types remained unchanged. Nevertheless, types 1, 2 and 7 predominated in both crops at the end of the cultivation period 1987/1988.

Except for sugar beet, all crops cultivated were highly colonized by VAM fungi, although levels of infection and the seasonal dynamics were variable. The spore densities of the different cereals varied during the cultivation periods, but had approximately the same maxima at harvesting. Cultivation of a non-host reduced the total spore density in soil, although different spore types varied in their viability. Independent of the crop plant cultivated, the spore types characteristic for Hotteln (1, 2 and 7) remained predominant.

Influence of N fertilization and fungicides on VAM fungi

Mycorrhizal colonization. Mycorrhizal development was similar on all plots investigated (Fig. 9) and increased N fertilization reduced the level of VAM infection to a negligible extent. The application of fungicides increased mycorrhizal colonization at both levels of N fertilization (Fig. 9), but was especially marked in the plot with increased N fertilization.

Spore density in soil. Here the object was to investigate whether increased N fertilization or fungicide application influenced the sporulation of VAM fungi. The dynamics of spore densities in soil (Fig. 10) was similar to those of mycorrhizal colonization. Application of fungicides increased sporulation of VAM fungi, whereas increased N fertilization had a minor inhibitory effect.

By the time of harvesting of winter wheat, the spore density in soil had distinctly increased in all plots investigated. In contrast, the number of infectious propagules remained unchanged or was lower (Table 7).

Distribution of chlamydospore types within VAM populations. Increased N fertilization or application of fungicides may cause a shift in the population pattern of VAM fungi. Since fertilization levels were not changed until April, the effect of this factor was only investigated at the end of the cultivation period 1988.

Analysis of the VAM population at harvesting showed that increased N fertilization especially influenced two types with low abundancy (types 4 and 10 in Fig. 11). Furthermore, the relative frequencies of the most abundant types (1 and 2) within the VAM population were changed, although these types remained predominant. From the dynamics of the absolute spore counts of the four types mentioned, it is seen that sporulation of types 4 and 10 remained almost unchanged after increased N fertilization, whereas sporulation of type 2 was slightly reduced at harvesting (Fig. 12). In this case, the changes in relative abundancy were not based on differences in spore number of these types, but on changes of the total spore density in soil or on changes in spore number of other types. Only type 1 spores usually showed a high sporulation rate in May, the time additional N fertilization was applied. This resulted in a significant reduction of sporulation of type 1 (Fig. 12).

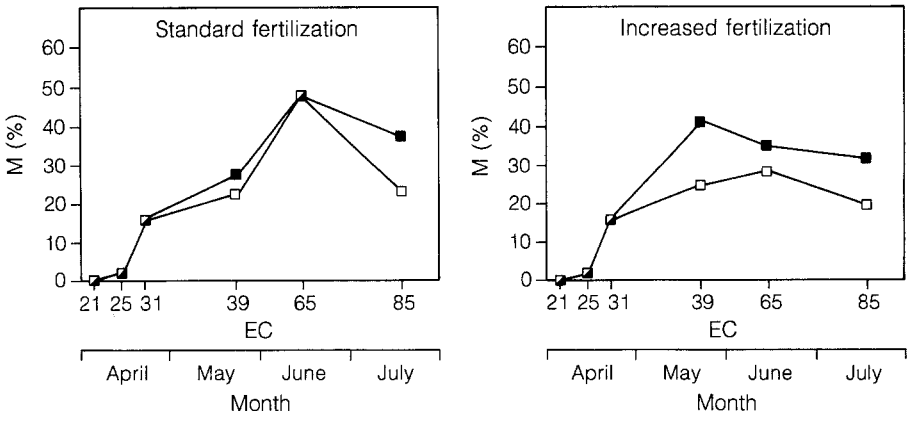

Fig. 9. Frequency of mycorrhizal infection (*M*) in winter wheat cv. Kanzler at two levels of N fertilization with and without fungicide application at Langreder during the cultivation period 1988. *EC*, Eucarpia stages of development (after Zadoks et al. 1974). ■, With fungicides; □, without fungicides

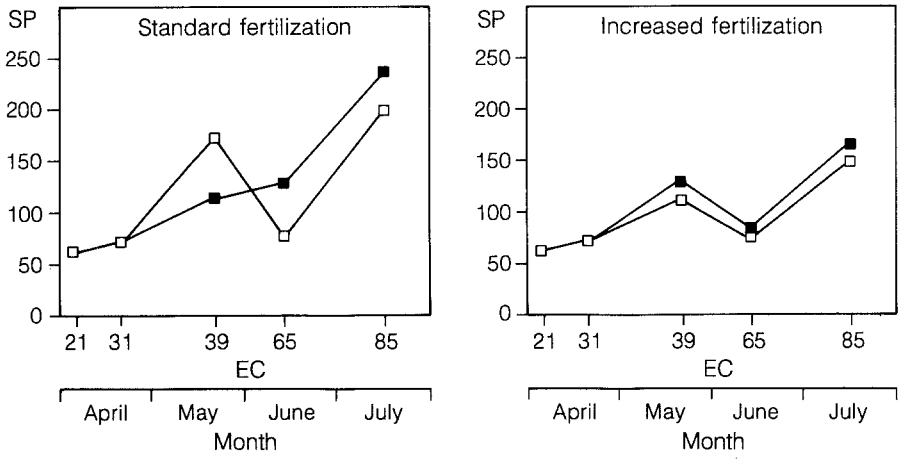

Fig. 10. VAM spore density in winter wheat cv. Kanzler at two levels of N fertilization with and without fungicide application during cultivation period 1988 at Langreder. *EC*, Eucarpia stages of development (after Zadoks et al. 1974); *SP*, spores/100 g dried soil. ■, With fungicides; □, without fungicides

Table 7. MPN and spore density in soil with winter wheat cv. Rektor at Langreder at the beginning of the vegetation period 1988 (spring) and in autumn after N fertilization and fungicide application. N, Standard fertilization; +N, increased fertilization; F, fungicide application. The numbers in parentheses are the lowest and highest MPN values at the 95% confidence limit

Fertilization	Spores/100 g soil		MPN/100 g soil	
	Spring	Autumn	Spring	Autumn
N	70	197	1954 (914–4174)	580 (320–1460)
+N		148		2022 (946–4320)
N, F		233		2076 (971–4436)
+N, F		165		1591 (745–3401)

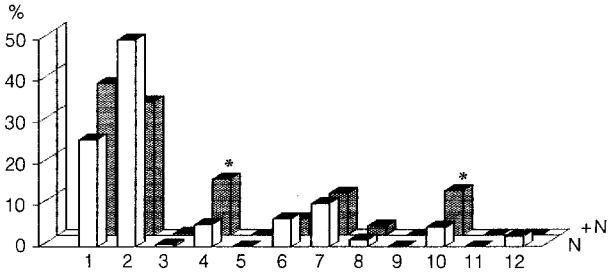

Fig. 11. Percentage of spore types within the VAM population present in winter wheat cv. Kanzler at the end of the cultivation period 1988 at Langreder with standard (*N*) and increased N fertilization (+*N*) without fungicide application. *Asterisks* indicate significant differences in abundance of one type between the two crops

Spore numbers at harvesting were similar in both treatments.

After fungicide application in combination with standard N fertilization, the percentage of type 2 within VAM populations decreased whereas types 1 and 10 increased (Fig. 13). Looking at the real spore counts of these three types during the vegetation period 1988, the spore number of type 10 remained unchanged after fungicide application, whereas sporulation of type 2 was slightly reduced at harvesting (Fig. 14). Like increased N fertilization, fungicide application significantly reduced sporulation of type 1 in May (Fig. 14). Nevertheless, the number of type 1 spores at harvesting was approximately the same in both treatments.

The results indicate that increased N fertilization reduced and fungicide application increased mycorrhizal colonization of plant roots and sporulation in soil to a

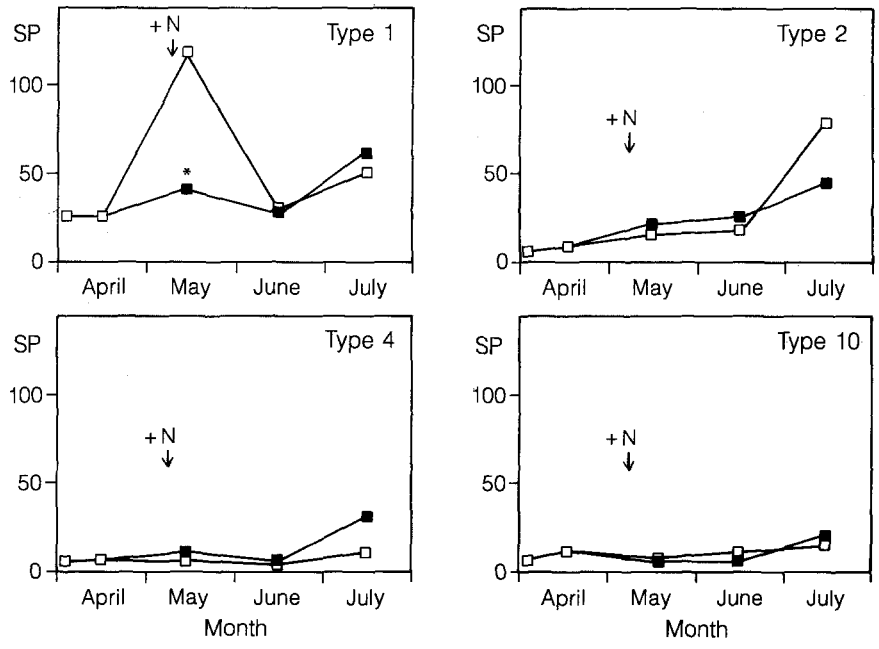

Fig. 12. VAM spore density of the most abundant types in winter wheat cv. Kanzler during the cultivation period 1988 at Langreder with standard and increased N fertilization without fungicide application. *+N*, Date of first increased N fertilization. *SP*, Spores/100 g dried soil. □, Standard fertilization; ■, increased fertilization

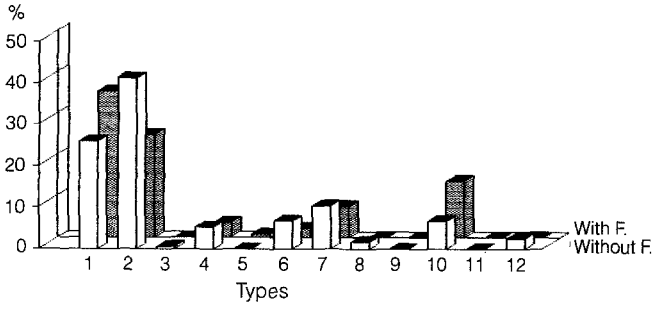

Fig. 13. Percentage of spore types within the VAM population present in winter wheat cv. Kanzler at the end of the cultivation period 1988 at Langreder with and without fungicide application (*F*) and with standard N fertilization

negligible extent. However, one type showing intensive sporulation at the beginning of these treatments was distinctly but transiently affected. The highest total spore densities were obtained at the end of the cultivation periods, but the characteristic spore types (1 and 2) remained predominant.

Discussion

The chosen crop plants influenced mycorrhizal development but not total spore densities in soil. This result disagrees with reports by Strubble and Skipper (1988) and Simpson and Daft (1990), who described an effect on both spore density and VAM development. However, these authors compared plants belonging to different families and the present investigation was restricted to members of the Graminaceae. A change in host plant promoted or repressed certain VAM fungi and thus the influence was expressed not in total spore density but in the composition of VAM populations. These results agree with those of Daniels Hetrick and Bloom (1986),

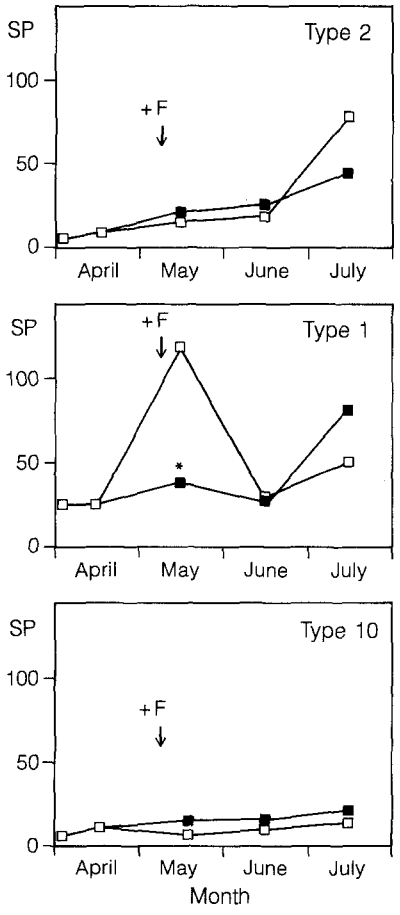

Fig. 14. VAM spore density of the most abundant types in winter wheat cv. Kanzler during the cultivation period 1988 at Langreder with and without fungicide application and with standard N fertilization. *Asterisks* indicate spore density significantly different at a given date; *+F*, date of first fungicide application; *SP*, spores/100 g dried soil. ■, With fungicides; □, without fungicides

who reported high sporulation rates of a few VAM fungi only on certain host plants. However, the effects observed in the present investigation were based on a change in the abundance of type 2 in favour of type 1, but the species *Glomus aggregatum* and *G. albidum* could be identified for both types. In summary, of the characteristic spore types of Hotteln (types 1, 2 and 7), types 1 and 2 remained predominant (Land and Schönbeck 1991).

Nevertheless, crop rotation may have a major impact on the occurrence of VAM fungi when it involves frequent or repeated cropping of non-hosts. Cultivation of sugar beet distinctly decreased the number of infectious units (by 65%) and spores (by 50%) within one cultivation period. Similar effects were described by Harinikumar and Bagyaraj (1988) for mustard, although Hayman (1974) found no effect within a period of 1 year. The differences observed by different authors probably depend on variations in microbial activity and the VAM species present. Weed contamination of non-host plots may also allow reproduction of VAM fungi.

In the present investigation, one of the most abundant spore types (type 1) had thin walls and were rapidly decomposed; other types with thick, pigmented walls (types 4, 10) or those sporulating in roots (type 7) remained viable in soil over one cultivation period. Despite low inoculum densities at harvesting, mycorrhizal infection of host plants began in April of the subsequent cultivation period in all plots investigated. This agrees with observations by Baltruschat and Dehne (1982). However, frequent or repeated cultivation of non-hosts within a close crop rotation may result in high losses of infectious structures, thus depressing mycorrhizal development.

In agricultural soils, VAM fungi are exposed to different cultivation practices. High levels of phosphorous (P) supply, particularly in unbalanced fertilization, may result in the pronounced depression of mycorrhizal development (Gnekow 1988). This effect can be attributed to lower levels of root exudation due to increased P content of the plant tissue (Bowen 1985), and thus to reduced positive chemotropism of fungal hyphae (Koske 1982). Increased N fertilization also changes the physiology and morphology of plants with possible consequences for mycorrhizal colonization. In winter wheat, some authors observed a decrease of VAM infection in plant roots and sporulation in soils after the supply of high levels of nitrogen (Hayman 1970; Kruckelmann 1975; Jensen and Jacobsen 1980; Baltruschat and Dehne 1982). Opinions as to how far these effects are influenced by the soil type are rather contradictory. Most authors reported more pronounced negative effects in sandy than in heavy and fertile soils. However, Kruckelmann (1975) found the converse effect. In the heavy soil (clayey silt) investigated in the present study, increased N fertilization reduced mycorrhizal colonization and sporulation to only a minor extent. This effect was also reflected in root length (data not presented) and an explanation in terms of "disease escape" can thus be excluded. The reductions in mycorrhizal colonization and spore production were probably caused by changes in amount or composition of root exudates resulting in lower attractivity of roots for VAM fungi as suggested in relation to P fertilization. The absence of the typical sporulation of type 1 (*G. albidum* and *G. aggregatum*) in spring was possibly an indirect influence of nitrogen on sporulation, since nitrogen causes many physiological changes in plants. In summary, N fertilization had only a minor influence on the characteristic patterns of VAM populations at harvesting.

In the present work, fungicide application increased mycorrhizal development and sporulation. In contrast, there are many reports of negative effects of fungicides with a wide action spectrum on mycorrhizal development (Ocampo and Hayman 1980; Dodd and Jeffries 1989). These investigations were mostly evaluated under controlled conditions with fungicides applied as soil drenches. Germination and hyphal growth of VAM fungi were reduced after such treatments. However, in practice most fungicides are applied onto leafs and only small amounts reach the soil. Furthermore, in these latter studies fungicides were applied during the cultivation period when VAM fungi are often already established in roots and protected against directly effects of fungicides. In fact, the lower sensitivity of an established mycorrhiza to fungicides has been reported (Dehne 1985; Dodd and Jeffries 1989). Dehne (1985) reported increase in mycorrhizal infection after leaf application of triadimefon, which belongs, together with the active substance applied in the present investigation (triadimenol), to the triazoles. Such an increase may possibly be the result of better plant health. The enlargement of assimilating area by mildew and rust protection can result in a better supply of assimilates to roots and consequent improvements in the development of VAM fungi (Strubble and Skipper 1988). The observed effect could also be side effects of the fungicide on plant physiology (Förster et al. 1980).

Sporulation of *G. aggregatum* and *G. albidum* (type 1) was reduced directly after fungicide application and the same effect was observed after increased N fertilization. Apparently these species were especially sensitive to changes in the soil and within the plants. Tommerup and Briggs (1981) already described the different sensitivities of various VAM species to several fungicides.

N fertilization, fungicide application and the crop plant all influenced mycorrhizal infection and spore dynamics, but the characteristic patterns of the VAM populations were not changed within one cultivation period. Relative to the effects of soil texture (Land and Schönbeck 1991), the three factors investigated here are of minor importance.

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