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Mycorrhiza

Influence of different soil types on abundance and seasonal dynamics of vesicular arbuscular mycorrhizal fungi in arable soils of North Germany

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Summary. The present investigation examines whether characteristic mycorrhiza occur in intensively used agricultural soils of different types. For this purpose, three arable soils in the north of Germany were chosen and the formation of mycorrhiza, spore density and the frequency of spore types within populations were investigated over two cultivation periods (1986/1987 and 1987/1988). Soil type influenced spore density as well as the percentage of mycorrhizal colonization of roots during each cultivation period, whereby high spore density was not necessarily connected with intensive mycorrhizal development. Although the level of mycorrhizal colonization increased most rapidly in silty sand, the highest level of infection was observed in barley roots in clayey soil. At harvest, infection levels differed in the soils investigated, but spore density was equal at all three sites. Each soil type displayed a characteristic distribution of spore types within a population; this distribution remained unchanged over the two cultivation periods.

Key words: Soil type – Winter barley – Vesicular arbuscular mycorrhizal infection – Spore density – Spore diversity

Introduction

Many reports document the occurrence of vesicular arbuscular (VA) mycorrhiza in diverse crops of intensively cultivated arable soils of temperate zones (Winter 1951; Kruckelmann 1975; Dehne 1981; Baltruschat and Dehne 1982; Hayman 1982; Jeffries et al. 1988). These fungi can increase plant growth under low-fertility conditions and, of particular interest for well-fertilized agricultural soils, can improve tolerance towards different kinds of stress such as drought (Allen and Boosalis 1983), or resistance towards root pathogens (Baltruschat and Schönbeck 1975; Schönbeck 1978; Caron 1989).

In a relatively short growing season, such effects clearly depend on early infection, which again is related to sufficient inoculum density and effective vesicular arbuscular mycorrhizal (VAM) fungi in the soil. There are, however, few quantitative and qualitative studies on inoculum density and population dynamics of VAM fungi in agricultural soils of temperate zones. The literature includes indications that tillage, fertilization and application of plant protection products may inhibit mycorrhiza. So far it is not known if soil type influences the composition of VAM populations. Thus, ecological studies are of fundamental importance before considering the possibilities of increasing indigenous VAM populations or of introducing more efficient species or strains into a field crop with a view to improving or stabilizing a high yield. The present study carried out a detailed evaluation of mycorrhizal populations in different soil types by monitoring the inocula, root colonization, and composition of the mycorrhizal populations.

Materials and methods

Sampling sites

Investigations were carried out over two cultivation periods (1986/1987 and 1987/1988) on three sites with different soil types in North Germany (Table 1). In 1986/1987 winter barley cv. 'Tapir' was cultivated at all three sites, whereas in 1987/1988 sugar beet (site 3), maize (site 2) and spring barley (site 1) were grown, in accordance with the practice of crop rotation. Prior to the experiments, oats (site 1) and winter wheat (site 2 and 3) had been grown on these sites (Table 2). According to Landwirtschaftliche Untersuchungs- und Forschungsanstalt, Landwirtschaftskammer Hannover (LUFA) the nitrogen, phosphate and potassium contents were high in all three field locations, due to adequate fertilization of the crops grown.

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Table 1. Composition, characteristics and yield of sites investigated

Parameter	Sites		
	Riesenbeck (soil 1, podzol)	Ahnsen (soil 2, para-brown earth)	Hotteln (soil 3, degraded black earth)
pH	5.3	6.4	7.3
Organic matter (C%)	3.3	3.4	3.1
Sand (%)	81.4	8.4	4.5
Silt (%)	17.6	88.4	82.3
Clay (%)	1.0	3.1	12.2
Soil texture	Silty sand	Silt	Clayey silt
Yield (winter barley)	45 dt/ha	60 dt/ha	90 dt/ha

Table 2. Crop rotation at sites investigated

Site	Soil type	Previous crop	Crop 1986/1987	Crop 1987/1988
Riesenbeck	1	Oats	Winter barley	Spring barley
Ahnsen	2	Winter wheat	Winter barley	Maize
Hotteln	3	Winter wheat	Winter barley	Sugar beet

Spore extraction and root examination

Five samples per date and location were taken from the surface soil-layer (0–30 cm) on eight dates during each cultivation period. Each sample was prepared for three different purposes: spore counting and classification into types; estimation of percentage of mycorrhizal roots; and cultivation, concentration and separation of VAM fungi for identification.

Spores were isolated by wet sieving (Gerdemann and Nicolsen 1963) 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 and transferred to petri dishes before being sorted into types under a binocular microscope. Only intact spores were counted. Classification was based on colour, size, shape, surface structure, general nature of the spore contents and hyphal attachment. Because of inter-sample variations in soil moisture, 20 g of each sample was dried and spore counts were recalculated for 100 g of dry soil to optain a uniform basis for comparison.

Roots were washed out from 400 ml soil to estimate the percentage of mycorrhizal colonization. The rate of infection was determined microscopically for 100 root segments, each of 1 cm length. The indigenous VAM fungi of the arable soils were maintained in pot culture on *Tagetes erecta* under controlled conditions. For taxonomic identification cultures with numerous of uniform spores were obtained by two steps; plants were first inoculated with infected root segments, and spores were subsequently selected from these cultures for further propagation (Schenck and Perez 1988).

Data analysis

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

Results

Mycorrhizal colonization

In autumn 1986, only sporadic infection by VAM fungi was observed on barley (Fig. 1). In all three soils investigated, distinct mycorrhizal colonization started in April after tillering. Further mycorrhizal development during the period of vegetation varied from site to site. In soil 1 (silty sand) the percentage of mycorrhizal colonization first increased rapidly until ear emergence at the end of May, and then decreased until harvesting, whereas in soils 2 and 3 frequency of mycorrhizal roots increased steadily from stem elongation (April) until harvesting at the end of July. Infection levels attained a high frequency (40–60%) in soils 1 and 3, while in soil 2 a maximum of only 30% of roots were mycorrhizal. Figure 2 shows soil temperatures at a depth of 15 cm during the cultivation period 1986/1987.

Spore density in soil

The frequency of mycorrhizal infection, particularly the rapid colonization of roots at the beginning of a cultivation period, depends, among other factors, on the inoculum density of a particular soil. Therefore, following the general practice, spores were counted as a parameter of inoculum density. As for mycorrhizal infection, spore density was also ascertained for each sample in order to determine the population dynamics.

Spore density increased distinctively from sowing until harvesting in soils 1 and 3 (Fig. 3). The dynamics of spore density varied, as did the mycorrhizal infection at the different sites (Fig. 3). During the cultivation period of 1986/1987, spore density first increased, then decreased at stem elongation in soils 1 (silty sand) and 3 (clayey silt), increasing again towards the end of the cultivation period; in soil 2, spore density varied only slightly. All three soils contained comparable numbers of spores at harvesting, independent of the dynamic during the vegetative period.

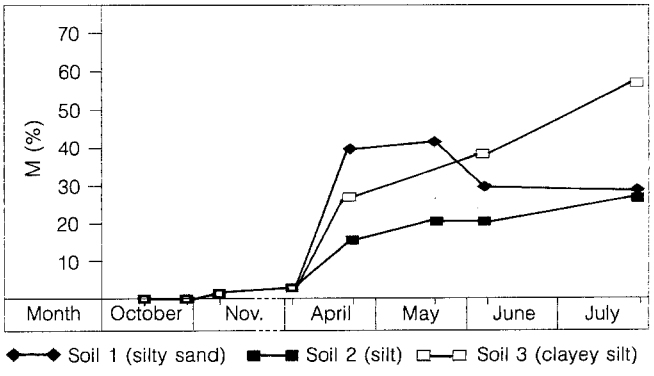

Fig. 1. Frequency of mycorrhizal infection (*M*) in winter barley, cv. 'Tapir', in three different arable soil types during cultivation period 1986/1987

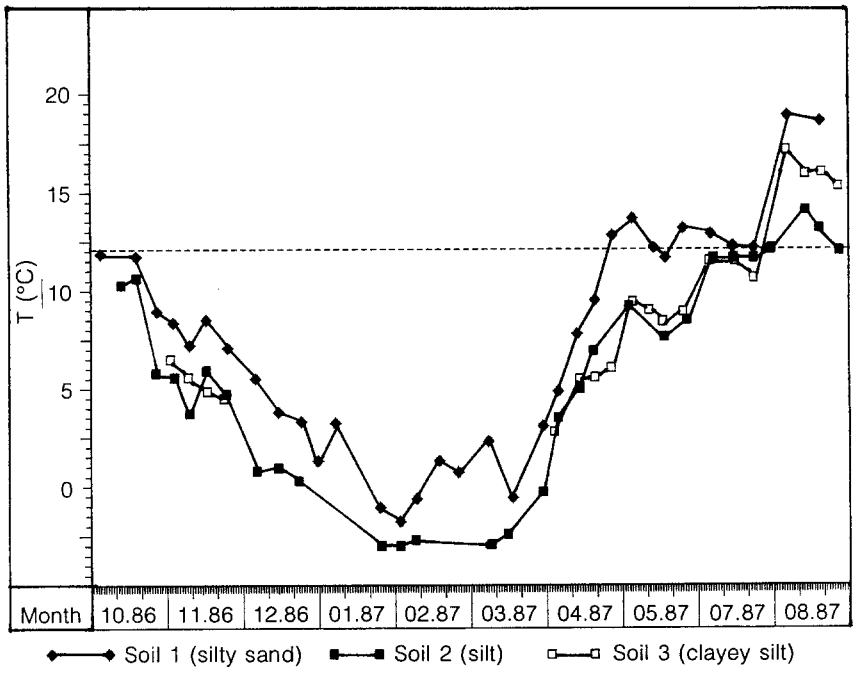

Fig. 2. Weekly average soil temperature at 15 cm depth for the different field locations during cultivation period 1986/1987

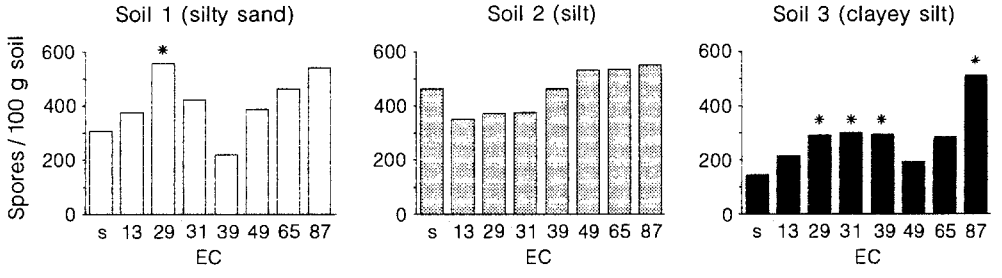

Fig. 3. VAM spore density in winter barley cv. 'Tapir' in three different soils during cultivation period 1986/1987. *s*, Sowing; *EC*, "Eucarpia" stages of development (after Zadoks et al. 1974). *Asterisks* indicate spore density significantly different from that at time of sowing of crop

Distribution of chlamydospore types and species within populations

Here, the object was to investigate if the varying dynamics of spore densities in the soils examined were correlated with different compositions of VAM populations. Different VAM species, possibly varying in their abilities to rapidly and intensively colonize the roots of winter barley, may predominate in different soils. We therefore isolated VAM spores from field soils, classified them into 12 types, and determined the percentage of each type within each population.

During the first cultivation period the abundance of each spore type differed between soil types (Fig. 4A). In soil 3 (clayey silt) VAM types 1, 2 and 7 predominated, with type 1 comprising the major proportion (40%) of the total number of spores. Soil 1 (silty sand) contained four characteristic types (4, 5, 6 and 10); the most abundant (type 6) constituted up to 20% of the total number. In soil 2 (silt), types 6 and 7 dominated, the latter accounting for up to 30% of the total spores. Type 6 was also most abundant in soil 1, while type 7 was one of the predominant VAM types in soil 3.

Crop rotation in the following vegetative period resulted in different crop plants being cultivated on the experimental sites in 1987/1988 (Table 3). One of these was a non-host (sugar beet), leading to a decrease in the total number of spores (Table 3). The distribution of spore types, however, remained unchanged (Fig. 4B).

The separation of spores into different types on the basis of morphological characteristics admits a certain possibility of errors: the appearance of spores varies according to soil type and age, so that in some cases one and the same species appears in diverse types and on the other hand one type includes more than one species. Only seven species of 12 classified types could be cultivated and subsequently identified. Table 4 shows the association of different *Glomus* species with characteristic types.

Discussion

In the arable soils investigated, only VAM fungi of the genus *Glomus* were recovered. According to the literature, many species of this genus seem to be adapted to fertile soils with a high nutrient level (Hayman and Stovold 1979; Jensen and Jacobsen 1980; Young et al. 1985; Jeffries et al. 1988). In contrast, the genera *Scutellospora*, *Acaulospora* and *Gigaspora* are more abundant in low-nutrient or nutrient-binding soils (Redhead 1977; Koske 1987; Gemma et al. 1989). Thus, VAM

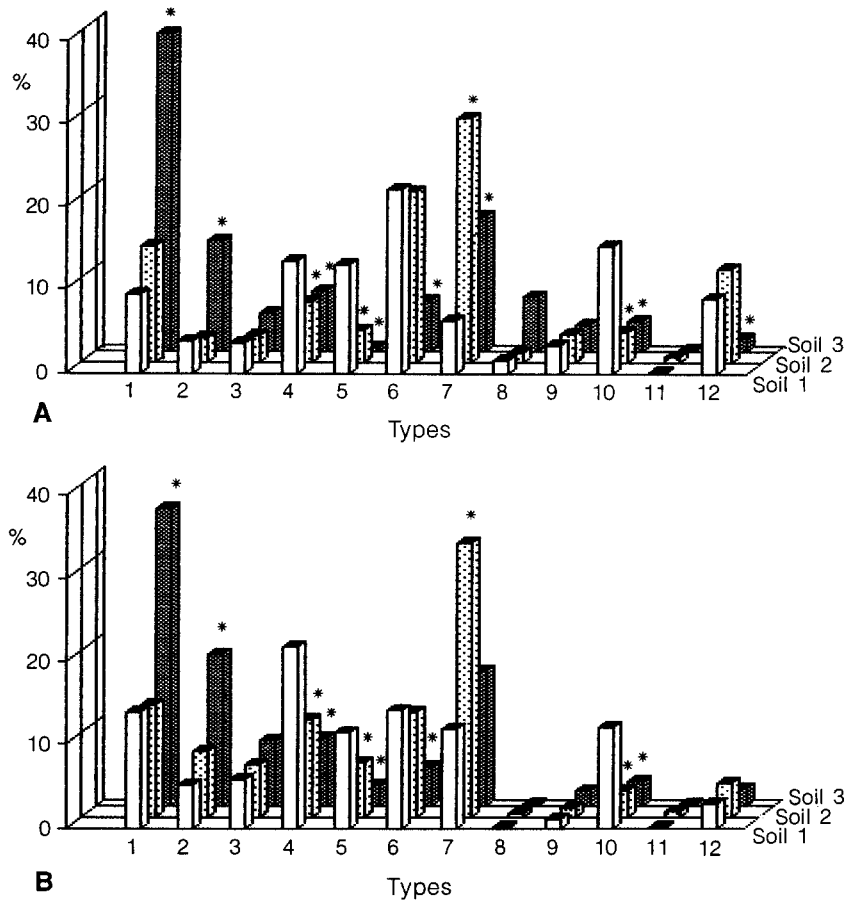

Fig. 4A, B. Percentage of spore types within the VAM population present in each of three different arable soils during cultivation periods 1986/1987 (**A**) and 1987/1988 (**B**). Data are averages of eight dates from each growing season; soil 1, silty sand; soil 2, silt; soil 3, clayey silt. *Asterisks* indicate significant differences in density of spore types in soils 2 and 3 from that of soil 1

Table 3. Development of mycorrhizal infection and spore density over cultivation period 1987/1988 at three sites investigated

Soil	Crop plant	Mycorrhizal infection			Spores/100 g dry soil	
		Start	Maximum	Harvest	Sowing	Harvest
1	Spring barley	April/May	30%	40%	359	563
2	Maize	June/July	39%	14%	464	582
3	Sugar beet		0%	0%	563	168

Table 4. Taxonomic identification of characteristic spore types at sites investigated

Site, soil	Types	Species (*Glomus*)
Riesenbeck		
1	4, 6	*Gl. etunicatum* and *Gl. caledonium*
	5	*Gl. intraradices* and *Gl. fasciculatum*
	10	*Gl. mosseae* and *Gl. caledonium*
Ahnsen		
2	6	*Gl. etunicatum* and *Gl. caledonium*
	7	*Gl. intraradices*
Hotteln		
3	1, 2	*Gl. aggregatum* and *Gl. albidum*
	7	*Gl. intraradices*

fungi are obviously adapted to specific climatic and nutritional circumstances.

Although fertilization had resulted in equal nutrient levels for all three soil types, soil texture (and therefore permeability to water and air), pH, temperature and adsorption capacity still differed from soil to soil. All these parameters probably influence VAM fungi.

The mycorrhizal colonization levels of the crop plants were similar to those documented in other investigations of arable soils in Germany, England and Denmark (Hayman and Stovold 1979; Jensen and Jacobsen 1980; Dodd and Jeffries 1986; Baltruschat 1987).

Mycorrhizal colonization in spring did not commence until the soil temperature had reached 5° C, which is also described as the critical temperature for spore germination and mycorrhizal infection by other authors (Tommerup 1984a; Buwalda et al. 1985; Trent et al. 1988). Further mycorrhizal development was re-

lated to soil temperature, which has, according to Furlan and Fortin (1973), a distinct influence on the rate and intensity of infection by VAM fungi. Silty sand (soil 1), for example, reached a temperature of 12° C about 8 weeks earlier than soils 2 and 3.

The course of mycorrhizal development corresponded to a typical sigmoid curve, consisting of a lag-phase and a subsequent increase, sometimes succeeded by a plateau and a decrease at the end of the vegetation period. Furlan and Fortin (1973) described a similar course of development, but did not, however, mention a decrease. The shape of the curve is probably related to a decrease in young roots over time, since only young roots can be infected by VAM fungi (Römer and Schilling 1986), and to a decrease in host carbohydrates, as these are mainly translocated to the ears of the crop.

Spore densities in autumn were several times higher than those recorded in comparable soils of Lower Saxony and England (Hayman 1974; Kruckelmann 1975), but were similar to those recorded from Wales (Hayman and Stovold 1979) and Denmark (Jensen and Jacobsen 1980).

Intensive sporulation was observed at the end of each cultivation period. Hayman (1982), who also observed this, supposed it to be connected with decreasing root growth and the beginning of senescense.

Maximum spore density was unaffected by soil type. As mentioned already, mycorrhizal infection and spore density displayed seasonal variations, which differed from soil to soil. Nevertheless, there was no correlation between mycorrhizal colonization of roots and spore density in any type of soil.

Each soil type displayed a characteristic pattern of population. Each of the 12 spore types was present in all three soils, but at different frequencies. So each soil contained some characteristic species. Differences were especially pronounced between soils 1 (silty sand) and 3 (clayey silt); soil 2 (silt) occupied an intermediate position. The VAM populations investigated possibly included more than the seven species identified. As many as 13 species have been described for one field soil (Land 1990), indicating that species diversity is probably much greater than recorded in the present work. However, direct identification of single spores from field soils is open to many possible errors.

Separation of VAM fungi by means of mycorrhizal roots possibly resulted in only rapidly infecting or very abundant fungi being selected from the total population present. The literature contains but few reports on the diversity of VAM populations. Often only the most abundant types or species are described (Kruckelmann 1975; Jensen and Jacobsen 1980; Gnekow 1988). Only Dalpe et al. (1986), evaluating an orchard soil, also identified seven species in a study similar to the present work.

The soils investigated varied in several parameters, so that it was impossible to exactly determine the influencing factors. The following considers the importance of each factor separately.

PH seems to be of minor importance, because although it was near neutrality in soils 2 and 3, the patterns of VAM populations differed significantly at these two sites. Furthermore, reports in the literature suggest that the species described in the present work are not affected by pH values ranging from 5.5 to 7.5 (Wang et al. 1985; Gemma et al. 1989).

A positive correlation between the frequency of VAM fungi and the percentage of organic matter in the soil has often been proposed (Howard 1948; Hayman 1974; Harinikumar and Bagyaraj 1989). The present investigation did not support this, since the percentage of organic matter was similar for all three soils (3.1%–3.4%).

VAM fungi are possibly mainly influenced by soil texture, affecting the partial atmospheric pressure and moisture, which might, in turn, inhibit germination or hyphal growth of VAM fungi (Reid and Bowen 1979; Le Tacon et al. 1983; Tommerup 1984b). Perhaps species differ in their sensitivity to these factors, so that any one characteristic VAM population develops according to the prevailing soil texture.

Summarizing, inoculum density was sufficiently high for rapid and intensive mycorrhizal colonization of the host plants. The soil type determined the composition of the VAM populations. However, to what extent the effectiveness of the predominant species and the characteristic composition of VAM populations may be influenced by fertilization, the application of plant protection products, or crop rotation has yet to be shown.

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