

## BRIEF NOTE

# Vesicular-Arbuscular Mycorrhizal Status of Spring Ephemerals in Two Ohio Forests<sup>1</sup>

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**ABSTRACT.** Nineteen spring ephemerals (7 monocots and 12 dicots) were surveyed for vesicular-arbuscular mycorrhizae (VAM) in two Ohio forests during spring 1993 and 1994. Eleven of the nineteen species sampled displayed VAM development which was consistent between years and sites, except for *Cardamine concatenata* (Brassicaceae). Patterns of VAM occurrence primarily reflected the taxonomic classification of the sampled species. In this study, all monocots sampled were mycorrhizal, while only 25% of the dicots developed VAM relationships. The occurrence of VAM in the dicots primarily reflected their taxonomy as well. All Ranunculaceae species were mycorrhizal while those in the other five dicot families were not.

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## INTRODUCTION

Approximately 95% of all plant species form mycorrhizal associations (Trappe 1987). Of those mycorrhizal associations, herbaceous species typically form the vesicular-arbuscular mycorrhizal (VAM) type (Brundrett 1991). The occurrence of VAM among herbaceous taxa has been well documented in a variety of ecosystems including forests (Trappe 1987). However, surveys of VAM occurrence within specific phenological groups within forest ecosystems are scant (however, see Brundrett and Kendrick 1988). One phenological group in forest ecosystems is the spring ephemerals. Spring ephemerals are species which complete their above ground activity before canopy closure in moist, rich temperate forests throughout the world (Grubb and Marks 1989). The purpose of this project was to examine VAM occurrence in spring ephemerals in two Ohio forests and to compare it with that reported in the literature for taxa of other phenological groups in similar forests.

## MATERIALS AND METHODS

Nineteen species of spring ephemerals from seven families were sampled in two Ohio forests during 1993 and 1994. One study site was the 40 ha Bohannon Preserve located in eastern Delaware County, OH (40°21'N, 82°56'W). It was an older growth, upland beech-maple forest which has not been disturbed since approximately 1900. Specific vegetation and soil information for the Bohannon Preserve is described in Boerner and Koslowsky (1989). The other site was the Wright State University (WSU) woods located in northwestern Greene County (39°45'N, 84°03'W). This site was an older growth (100+ years), oak-maple forest dominated by *Acer saccharum* Marsh., *Quercus rubra* L., and *Quercus alba* L. Specific vegetation and soil information is described in DeMars and Runkle (1992) and DeMars and Boerner (1995).

In the field, sampling was conducted by extracting six individuals of each spring ephemeral species present at each site per sampling event (see Table 1 for dates). Following extraction, plants were placed in plastic bags with soil intact for transport to the laboratory. Samples were taken from the upland stand of each site as described in Boerner and Koslowsky (1989) and DeMars and Boerner (1995) to avoid possible topographic variations. In the laboratory, roots were rinsed, separated, cleared and stained using trypan blue (Phillips and Hayman 1970). Roots were preserved with a 1:1:10 FAA (formalin:acetic acid:ethanol) solution and stored in plastic tissue cassettes under refrigeration.

VAM development was examined in five randomly selected 2-3 cm root segments (including root tips) per sampled plant. VAM development was defined as the presence of any internal nonseptate hyphae, vesicles, arbuscules, or chlamydospores. The percentage of root length colonized (PRLC) by mycorrhizal fungi was estimated for each segment (Giovanetti and Mosse 1980). The mean PRLC was then computed for each plant and averaged for the entire sample ( $N = 6$  plants per species per sampling event).

## RESULTS AND DISCUSSION

Eleven of the nineteen spring ephemeral species examined for VAM development were mycorrhizal (Table 1). The patterns of VAM development between sites and years were consistent and apparent in all cases. Consequently, statistical analysis was not deemed necessary to describe obvious patterns among species.

All the monocot species examined in this study were mycorrhizal (Table 1). The high percent root length colonized (PRLC) in the Liliaceae probably reflected their root morphology. These species have magnolioid root systems characterized by minimal root hair development (Baylis 1975). The low numbers of root hairs increase their dependence upon zygomycetous VAM resulting in mycotrophy (mycorrhizal dependence) as described by Trappe (1987). Other studies have indicated similar results for some liliaceous spring

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TABLE 1

Mean percent root length colonized (PRLC) by vesicular-arbuscular mycorrhizal fungi in spring ephemerals (N = 6 individuals per species per sampling event) in the Bohannon Woods Preserve (BW) and the Wright State University Woods (WSU).

Spring Ephemeral Species	Family	Site	Date	Mean PRLC
<b>Monocots</b>				
<i>Allium tricoccum</i> Ait.	Liliaceae	BW	4-22-93	56
			4-25-94	64
		WSU	4-23-93	53
			4-19-94	57
<i>Erythronium albidum</i> Nutt.		WSU	4-23-93	63
			4-19-94	61
<i>Erythronium americanum</i> Ker Gawler		BW	4-22-93	59
			4-25-94	54
<i>Ornithogalum umbellatum</i> L. #		WSU	4-23-93	66
			4-19-94	52
<i>Trillium flexipes</i> Raf.		WSU	5-16-93	59
			5-09-94	63
<i>Trillium grandiflorum</i> (Michx.) Salisb.		BW	5-23-93	50
			5-19-94	61
<i>Trillium sessile</i> L.		BW	5-23-93	72
			5-19-94	59
		WSU	5-16-93	63
			5-09-94	67
<b>Dicots</b>				
<i>Erigenia bulbosa</i> (Michx.) Nutt.	Apiaceae	BW	4-03-93	0
			4-05-94	0
<i>Cardamine concatenata</i> (Michx.) O. Schwarz	Brassicaceae	BW	4-22-93	0
			4-25-94	0
		WSU	5-16-93	15
			5-09-94	0
<i>Cardamine dissecta</i> (Leavenw.) Al-Shehbaz		BW	4-22-93	0
			4-25-94	0
<i>Cardamine douglassii</i> Britton		BW	4-22-93	0
			4-25-94	0
		WSU	4-23-93	0
<i>Cardamine rhomboidea</i> (Pers.) DC.		WSU	4-19-94	0
<i>Dicentra canadensis</i> (Goldie) Walp.	Fumariaceae	BW	4-22-93	0
			4-25-94	0
<i>Dicentra cucullaria</i> (L.) Bernh.		BW	4-22-93	0
			4-25-94	0
<i>Floerkea proserpinacoides</i> Willd	Limnanthaceae	BW	4-22-93	0
			4-25-94	0
<i>Claytonia virginica</i> L.	Portulacaceae	BW	4-22-93	0
			4-25-94	0
		WSU	4-23-93	0
			5-16-93	0
			4-19-94	0
			5-09-94	0
<i>Anemone virginiana</i> L.	Ranunculaceae	BW	4-22-93	24
			4-25-94	34
		WSU	5-16-93	49
			5-09-94	38
<i>Anemonella thalictroides</i> (L.) Spach.		WSU	5-16-93	63
			5-09-94	58
<i>Ranunculus ficaria</i> L. #		WSU	5-16-94	23

# indicates a non-native species.

ephemerals with magnolioid root systems; Brundrett and Kendrick (1988) observed 83% PRLC for *Allium tricoccum*, 67% PRLC for *Erythronium americanum*, and 84% PRLC for *Trillium erectum* and *Trillium grandiflorum* in Ontario, while DeMars and Boerner (1995) reported 60.2% PRLC for *Trillium flexipes* in Ohio.

The distribution of VAM development in the dicots examined in this study reflected their taxonomic classification also. All Ranunculaceae species were mycorrhizal while only one sample from the other dicot families were (Table 1). Trappe (1987) indicated that the order Ranunculales is highly mycotrophic as are most other terrestrial orders of the subclass Magnoliidea. The exception to this is the Papaverales in which 61% of all species examined were scored as nonmycorrhizal (Trappe 1987). In the present study both *Dicentra canadensis* and *D. cucullaria* (Fumariaceae/Papaverales) were nonmycorrhizal, lending support to the weakly mycotrophic status of the order. Brundrett and Kendrick (1988) also observed similar patterns for species they sampled in both these orders.

The orders Caryophyllales (subclass Caryophyllidae) and Capperales (subclass Dilleniidae) have also been deemed weakly mycotrophic by Trappe (1987). In the present study *Claytonia virginica* (Portulacaceae/Caryophyllales) and *Cardamine* spp. (Brassicaceae/Capperales) were nonmycorrhizal with the exception of *Cardamine concatenata* in the 1993 WSU sample. The nonmycorrhizal status of these species probably reflected their more ruderal life histories and the production of mustard oil glucosides, glucosinolates, and other defensive chemicals (Trappe 1987). However, it has not been definitively demonstrated that such phytochemicals prevent the establishment of VAM (Schreiner and Koide 1993).

The observation of VAM development in the 1993 WSU sample of *C. concatenata* probably indicates an "apparent" tendency to form VAM during its senescence period when root integrity becomes compromised (DeMars and Boerner 1995). The formation of VAM in the nonmycotrophic Brassicaceae has been reported by numerous authors (Harley and Harley 1987, Medve 1983, Newman and Redell 1987, DeMars and Boerner 1994), but its significance has often been questioned (Hirrel et al. 1978, Tester et al. 1987, Koide and Schreiner 1992) as most authors suggest that it is nonfunctional due to its occurrence in senescent roots.

In the present study, *Erigenia bulbosa* (Apiaceae/Apiales) was nonmycorrhizal in all samples even though its family has shown a high degree of mycotrophy (Trappe 1987). The nonmycorrhizal status in this species may also reflect chemical defenses; however, cold temperatures may have prevented VAM development. *E. bulbosa* is the earliest spring ephemeral to become active. Its above ground activity is limited to March and April in the study forests, a time when soil temperatures are low. Temperature has been shown to play a major role in VAM development. Hayman (1983), Chilvers and Daft (1982), and Anderson et al. (1987) have shown limited VAM development at temperatures as high as 10° C, and Smith and Bowen (1979) have reported lack

of VAM initiation at slightly lower temperatures.

In *Floerkea proserpinacoides* (Limnanthaceae/Geraniales), the lack of VAM development may have been a result of chemical defense compounds similar to those found in members of the Brassicaceae. Gleason and Cronquist (1991) reported that *Floerkea* sp. produces mustard oils also.

Overall, the levels of PRLC in species of the mycotrophic taxa (Trappe 1987) corresponded well with those reported for other mycotrophic, forest spring ephemerals (Brundrett and Kendrick 1988, DeMars and Boerner 1995) and for summer-green herbs (Mayr and Godoy 1989) such as *Geranium robertianum* L. (Boerner 1990), *Geranium maculatum* L. and *Polygonatum pubescens* (Walter) Elliot (Boerner 1986), and *Smilacina racemosa* (L.) Desf. (Brundrett and Kendrick 1988, DeMars and Boerner 1995). In conclusion, the mycorrhizal status of spring ephemerals in the two sites in this study reflected their taxonomy. This is not surprising as mycotrophy (VAM dependence) originated from coevolutionary plant-fungal interactions in certain taxa and from interactions which would have resulted in preclusion of such fungi in other taxa (Trappe 1987).

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