# A SURVEY OF MYCORRHIZAS IN KANSAS BLUESTEM PRAIRIE AND ADJACENT FOREST

by

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## INTRODUCTION

This study was designed to ascertain the occurrence of mycorrhizas in prairie and forests near Manhattan, Kansas, in order to better understand the ecological interactions of species in these ecosystems.

Fungus-root organs or mycorrhizas (Frank, 1885) are natural phenomena occurring in those parts of root systems primarily responsible for absorption of nutrients. They are nonpathogenic associations between fungi and the roots of higher plants (Robinson, 1967) in which the fungal partners may not merely enhance but may be necessary for the growth of their associated higher plants. Possible explanations for the fungus-root association and the occurrence of root-surface and rhizosphere populations have been summarized by Harley (1969).

Although it is generally accepted that mycorrhizas affect the success of woody plants in forests (Went and Stark, 1968; Wilde, 1968; Harley, 1969), world-wide reports indicate that many grasslands are conspicuous in their dearth of mycorrhizas. University of Wisconsin studies indicate that trees, naturally or artificially established, do not grow normally if mycorrhizal fungi are absent. These have led Wilde (1968) to concur with White (1941) that the absence of tree mycorrhizal fungi is largely responsible for the persistence of the intra-zonal prairies in the humid climate of the American Midwest. But many other studies have shown that fire is chiefly responsible for the persistence of prairies. Burning is a process that is widely considered to be a major environmental factor in forming and maintaining prairies (Buell and Facey, 1960).

# SITES AND METHODS

## Areas

Riley County, Kansas, is part of the Flint Hills, a major segment of true prairie. Elevation of sites studied ranges from 311m (1019ft) to 402m (1320ft).

Physiographically a strongly dissected plain, the terrain is hilly with narrow divides bordered by steep slopes and rock outcrops. At Manhattan, rainfall averages about 81cm (32 inches) with about 75% of it falling during the growing season. The mean temperature for January is -2 C (28F) and for July is 25 C (78F) (U. S. Weather Bureau, 1956). Native vegetation is bluestem prairie (Andropogon-Panicum-Sorghastrum), northern floodplain forests (Populus-Salix-Ulmus), oak-hickory forests (Quercus-Carya) and a small area of cross timbers (Quercus-Andropogon) (Küchler, 1964).

As described by Fly (1949), the residual soils have developed from massive limestones, interbedded gray and yellow shales, and highly flinty or cherty limestones of the lower Permian formations. Most of the soils are Mollisols belonging to the Udoll (Prairie or Brunizem soils and some Reddish Prairie and Regosol soils) and Ustoll (Chernozem soils) suborders. Fertility is moderate to high in the dark, well-granulated silt loam or silty clay loam surface horizons that are slightly acid in reaction. Texture and consistency of subsoil, depth of soil, and degree of stoniness vary widely with the nature of the parent material and the degree of slope.

To represent a wide variety of conditions in the Riley County area where forest and prairie intermingle, specific sites were selected to include forest and prairie on both upland and lowland and a variety of soil types (Table 1). Selection of a variety of soils was included because previous studies (Harley and McCready, 1950; Melin and Nilsson, 1957; Went and Stark, 1968; Wilde, 1968) have shown a relationship between soils and mycorrhizas. Finally, heavily grazed, moderately grazed, and ungrazed grasslands, burned and unburned grasslands, and grazed and ungrazed forests were included. Previous studies (Rosendahl and Wilde, 1942; Wilde, 1958) revealed that forest soils subjected to cultivation and grazing retained mycorrhizal fungi.

Table 1. Location and characteristics of sites

Symbol	1 Name	Location in Kansas	Range Site	Vegetation	Management
Dry	Dwight-Irwin Complex 1-4% slope	Riley Co.,3 miles NW Manhattan, SW1/4 sec. 35, T 9S, R 7E	Claypan- clay upland	Bluestem Prairie (Andropogon-Panicum- Sorghastrum)	Park - disturbed
DrD	Dwight-Irwin Complex 1-4% slope	Riley Co.,5 miles NW Manhattan, W1/2 sec. 27, T 9S, R 7E	Claypan- clay upland	Bluestem Prairie	Unburned 1950- 1969, Grazed
蓋	Breaks-Alluvial Land Complex	Riley Co.,5 miles NW Manhattan, W1/2 sec. 27, T 9S, R 7E	Clay upland- loamy lowland	Bluestem Prairie	Unburned 1950- 1969, Grazed
Кa	Kahola silt loam	Riley Co.,1 mile N Manhattan, NW1/4 of NW1/4 sec.6,T 10S,R 8E	Loamy lowland	Bluestem Prairie	Unburned, Ungrazed
١٧	Ivan-Kennebec silt loam	Riley Co.,7 miles SW Manhattan, SE1/4 of SE 1/4 sec.2,T110S,R 7E	Loamy lowland	Northern Floodplain Forest (Populus- Salix-Ulmus)	Grazed
ಕ	Chase silty clay loam	Riley Co.,KSU Campus, SE1/4 of SW1/4 sec.7, T 9S, R 7E	Loamy lowland	Planted-lawn and shade trees	Lawn is mowed
BFM	Benfield-Florence Complex 5-20% slope	Riley Co.,3 miles NW Manhattan, SW1/4 sec. 35, T 9S, R 7E	Loamy upland	Prairie forest margin	Grazed
BfD	Benfield-Florence Complex 5-20% slope	Riley Co.,5 miles NW Manhattan, W1/2 sec. 27, E1/2 sec. 28, T 95, R 7E	Loamy upland	Bluestem Prairie	Burned and unburned, Grazed

Table 1. Continued

Symbol	1 Name	Location in Kansas	Range Site	Vegetation	Management
St	St Stony Steep land	Riley Co., 1 mile S Manhattan, W Cent. Sec. 29, T 10S,R 8E	Breaks	Oak-Hickory Forest (Quercus-Carya)	Grazed
Sa	Sarpy loamy fine sand	Riley Co.,3 miles SW Manhattan, W1/2 of SW 1/4 sec. 31,T 10S,R 7E	Sand	Trees, shrubs, and herbs on sand dunes	Nearly undis- turbed
권	Thurman loamy fine sand(tentative name) 6-12% slope	Pottawatomie Co.,7 miles E Manhattan, SE1/4 sec. 5, T 10S, R 9E	Sand	Cross Timbers (Quercus- Andropogon)	Occasionally burned
ThB	Thurman loamy fine sand(tentative name) 6-12% slope	Pottawatomie Co., 5 miles E Manhattan, SW1/4 of NE1/4 sec. 12, T 10S, R 8E	Sand	Northern Floodplain Forest	Highly diserreturbed by soil deposition and grazing

# Methods

Collections of specimens were made from June to September, 1971. Plant roots were carefully dug without removing adhering soil. They were taken to the laboratory in plastic bags where they were soaked in water until the soil mass softened and could be removed with a gentle water spray. Sections of desired roots were cut and further rinsed to wash away any remaining particles. Great care had to be taken to avoid excessive washing which would reduce surface mycelia. If microscopic study was not undertaken immediately, the roots were wrapped in wet paper toweling and stored in a refrigerator.

Woody species were a special problem because of the tannins in their root tissue. Several methods of clearing were tried. The first method (Bevege,1968) involved autoclaving the whole rootlets in potassium hydroxide, clearing them in sodium hypochlorite, and staining by autoclaving the tissue in alcohol lactophenol cotton blue as prepared by Shipton and Brown. This cleared the roots of tannin effectively without disrupting either plant or fungal tissue. It was difficult to obtain a good contrast of the blue-stained fungal structures against the pale host tissue. Thus this procedure was slow, and many trials were needed to adapt the basic methodology to particular species. The tissue became soft in this process and so hand-sliced microscopic sections were difficult to obtain with accuracy and without damage to tissue. The primary value of this method was in ascertaining location of fungal structures and the points of entry of surface hyphae on whole rootlets.

Another method used for clearing and staining woody plant roots as given in Gray's Formulary and Guide (1954) was developed by Garrett (1937) and Langeron (1942), Freshly washed root tissue was soaked overnight in a solution containing 4g of sodium hydroxide and 0.4g of bromothymol blue in 100ml water. The sodium hydroxide cleared the roots and the bromothymol blue stained the fungi. The con-

trast was not always the best but this method provided more laboratory work time because the clearing and staining went on without attention. An important asset of this method was the retention of turgidity by the roots so hand-sliced sections could be made more easily than after Bevege's method.

Usually the grass roots did not need clearing and staining was done with bromothymol blue. Grasses were difficult to work with because the matted and intertwining roots demanded collection in bulk form rather than as a single clump of grass. Following separation of the turf into component species and careful washing, the root systems were cut from their foliage and studied or fixed in formal acetic acid for later study (Nicolson, 1959). Whole mounts were used for most studies because the distribution and details of infection, if present, could be easily observed.

Microscopic study involved both whole mounts and transverse sections. The former were studied for the presence of dichotomous branching, root surface mycelia, and penetration into inner tissue. Cross sections were always made if the whole mount indicated fungal presence. Although microtome sections would have been more uniform in thickness, factors of time, cost, and personal competency were considered and free-hand transverse sections were selected as adequate for this study.

A straight-edge razor was used to make the sections. The roots were held between halves of tapioca root pith which had been preserved in 95% alcohol. Sections were transferred from water on the razor to a small dish with sectioning continuing until there were enough pieces to prepare a slide. Using a dissection microscope, the sections were transferred to a slide and mounted in lactophenol blue. Later Perma-mount was used on those slides to be retained on a semi-permanent basis.

Microscopic study of the cross sections revealed that some fungi were merely

in close proximity to the root but not attached to it. Sometimes hyphae were attached to the fine soil particles on the external surface of the root or enmeshed in root hairs. Fungi were determined to be mycorrhizal only if they had penetrated the root tissue. In most mycorrhizal species the fungi formed a mat in association with the outer layers of the root. Often there was a layered appearance and some occasional outgrowths. Internal penetration into the cortical cells was found in some species.

A list of species studied and their common names is given in Appendix Table 1. Identification of the higher plant species collected was checked with specimens in the Herbarium of Kansas State University. Both identification and scientific nomenclature for Gymnospermae are in accordance with Fernald (1950) and for Angiospermae with Barkley (1968). Common names are from Anderson and Owensby (1969). Vegetative categories in Küchler's Map of Potential Natural Vegetation of the Conterminous United States (1964) are used in Table 1 with the exception of the cultivated, prairie forest margin and sand dune areas which did not fit any of the categories. Soil nomenclature and range sites are from the unpublished Riley and Pottawatomie County Soil Surveys. The Riley County soil survey has been finished and the completed manuscript awaits publication. The Pottawatomie County soil survey is not yet complete so correlation of names has not yet been made. Symbols are soil-map symbols except for the added letters M, D, C, and B, in a few cases, to distinguish sites on the same soil type.

## RESULTS

# Mycorrhizal Species

Ten of the 31 tree species studied possessed mycorrhizas (Tables 2 and 3) but no true mycorrhizas were found in woody shrubs or herbs (Tables 4 to 7).

The trees possessed ectocellular, endocellular and ectoendotrophic mycorrhizas

Table 2. Species exhibiting mycorrhizas

Species	Туре
Juniperus virginiana	Endocellular mycorrhiza
Ostrya virginiana	Ectocellular mycorrhiza
Pinus ponderosa	Ectocellular mycorrhiza
Pinus strobus	Ectocellular mycorrhiza
oinus sylvestris	Ectoendotrophic mycorrhiza
latanus occidentalis	Ectocellular mycorrhiza
uercus borealis	Ectocellular mycorrhiza
uercus macrocarpa	Ectocellular mycorrhiza
uercus marilandica	Ectocellular mycorrhiza
uercus prinoides	Ectocellular mycorrhiza

Table 3. Trees examined for mycorrhizas

						Si	Sites					
Species	DrM	Oro	ਲ	Κa	I,	చ	BFM	BFD	St	Sa	ThC	ThB
Acer negundo					0							
Aesculus glabra var. sargentii					0							
Asimina triloba									0			
Carya cordiformis					0							
Catalpa speciosa									0			
Celtis occidentalis					0					0		0
Cercis canadensis					0				0			
Fraxinus pensylvanica									0	· ·		0
Gleditsia triacanthos					0			0		0		0
Gymnocladus dioica					0							
Juglans nigra									0	0		
Juniperus virginiana (young)		e.						0				
Juniperus virginiana				+		+	+		+			
Maclura pomifera							0	0			0	0
Morus alba												0
Morus rubra					0					0		0
Ostrya virginiana									+			

Table 3. Continued

						Sites	es					
Species	Dry	5	ਲ	κ	Ţ	క	BFM	BFD	St	Sa	ج ا	ThB
Pinus ponderosa						+						
Pinus strobus		8				+						
Pinus sylvestris						+						
Platanus occidentalis						+			+			
Populus deltoides	*3				0					0		0
Prunus persica												0
Ptelea trifoliata			8		0							
Quercus borealis var. maxima										S	+	
Quercus macrocarpa					+				+			
Quercus marilandica											+	
Quercus prinoides					+				+	0		0
Salix fragilis												0
Tilia americana					0					0		
Ulmus americana					0	0	0	0		0	0	0
Zanthoxylum americanum			27					0				0

Table 4. Shrubs and woody vines examined for mycorrhizas

						S	tes					
Species	DrM	Oro	滋	Z.	IV	క	BFM	Bf0	St	Sa	ThC	ThB
Amorpha fruticosa									0			
Cephalanthus occidentalis												0
Cornus drummondi	0		0	0			0	0	0	0	0	0
Euonymus atropurpureus									0			
Menispermum canadense					0				0			0
Parthenocissus quinquefolia					0				0		0	0
Prunus americana	0											
Prunus angustifolia											0	
Rhus aromatica var. serotina									0	0		
Rhus glabra									0	0		0
Rhus radicans												0
Ribes missouriense			0		0			0	0			0
Smilax hispida					0				0			0
Symphoricarpos orbiculatus	0				0			0	0		0	0
Vitis riparia											0	
Vitis vulpina									0			0

Grasses and grass-like plants examined for mycorrhizas (+) and peripheral fungi (\*) Table 5.

						S	Sites					
Species	DrM	Oro	滋	ъ	ľ	ಕ	BfM	BfD	St	Sa	윤	ThB
Andropogon gerardí	0	0	0					0		0		
Andropogon scoparius	0	0	0					0				
Bouteloua curtipendula		0	0					0	0			
Bouteloua gracilis		0										14)
Bouteloua hirsuta		0										
Bromus japonicus								0				
Bromus purgans					0				0			
Bromus secalinus				0								
Buchloe dactyloides		*										
Carex brevior			0									
Carex sp.		0	0		0			0		0		
Cenchrus longispinus										0		0
Cyperus filiculmis								0		0		
Cyperus schweinitzii										0		
Digitaria sanguinalis												0
Elymus canadensis			0		0			0	0			
Eragrostis spectabilis		0	0					0		0		0

Table 5. Continued

						SI	Sites				
Species	Dr.M	Ş	ਲ	ক্	2	క	B₽	B£0	\$ Sa	ThC	ThB
Eragrostis trichodes									0		0
Juncus interior		20	•					0			
Koeleria cristata		0						0			
Leptoloma cognatum											0
Muhlenbergia racemosa											0
Muhlenbergia schreberi											0
Panicum lanuginosum								*			
Panicum oligosanthes		*	*					*	*		
Panicum virgatum	*	*	*					*	*		
Paspalum ciliatifolium								0	0		0
Poa pratensis		0	0					0			
Schedonnardus paniculatus		0									
Setaria faberii											0
Setaria lutescens									0		0
Setaria viridis			0								
Sorghastrum nutans		0	0					0			
Spartina pectinata			0			•		0			

Table 5. Continued

						Si	Sites					
Species	Drw	DrD	쓢	κ	Ι	ర	BFM	8fD	St	Sa	BfM BfD St Sa ThC ThB	ThB
Sporobolus asper	0	0						0		0		
Sporobolus cryptandrus		0								0		0
Sporobolus heterolepis		0										
Sporobolus vaginiflorus var.			0									
Tridens flavus								0	0	0		0

Table 6. Forbs, except composites, examined for mycorrhizas

						5	Sitoc					
						5	3					
Species	DrM	0r0	ਲ	κā	ĭ	ಕ	BFM	BfD	St	Sa	Thc	ThB
Abutilon theophrasti												0
Acalypha ostryaefolia												0
Agastache nepetoides		12							0			
Amorpha canescens		0						0				
Amorpha fruticosa								0				
Asclepías speciosa	0											
Asclepías stenophylla			0									
Callirhoe alcaeoides								0		0		
Campanula americana					0				0			
Cassia fasiculata										0		
Chenopodium album			0		0							0
Chenopodium gigantospermum		×			0							0
Croton monanthogynus		0	0		pi.					0		
Desmodium glutinosum									0			
Desmodium illinoense	0											
Desmodium paniculatum									0			
Euphorbia dentata								0		0		0

Table 6. Continued

						Si	Sites					
Species	DrM	DrD	ਲ	Ka	)	క	BFM	BFD	ĸ	Sa	ThC	ThB
Euphorbia hexagona									0	0		0
Euphorbia nutans									0	0		
Gaura parviflora			0					0				
Geum canadense					0			0	0			0
Hackelia virginiana												0
Hypericum perforatum	0							0				
Ipomoea hederaceae												0
Kochia scoparia												0
Laportea canadensis					0							
Lespedeza capitata	0							0				
Lespedeza repens									0			
Linum sulcatum	0	0						0				
Oenothera serrulata	0											
Parietaria pensylvanica												0
Phryma leptostachya									0			
Physalis virginiana var. virginiana												
Phytolacca americana												0

Table 6. Continued

						Si	Sites					
Species	Dr.	Oro	滋	Кa	\	ਠ	BFM	BFD	St	Sa	TPC	ThB
Polygonum hydropiper					0							0
Salvia azurea var. grandiflora		0										
Scrophularia marilandica									0			
Solanum carolinense			0									0
Solanum nigrum										0		
Strophostyles helvola var. helvola												0
Strophostyles leiosperma										0		
Teucrium canadense var. virginicum			0					0				
Tradescantia bracteata										0		
Tragia urticifolia		0										
Verbascum blattaria								0				
Verbena stricta		0	0		0			0		0		0
Verbena urticifolia									0			
Viola sp.					0							

Table 7. Composites examined for mycorrhizas

						Si	Sites					
Species	Drw	P.O.	쑮	Ka	)I	ಕ	BFM	BfD	St	Sa	ThC	ThB
Achillea millefolium	0											
Ambrosia artemisiifolia												0
Ambrosia psilostachya		0	0		0			0		0		
Artemesia ludoviciana		0	0					0	0			
Aster drummondii									0			
Aster ericoides		0						0				
Aster sericeus								0				
Bidens bipinnata						73						0
Cirsium altissimum									0			
Conyza canadensis		0	0							0		0
Eupatorium altissimum											0	
Eupatorium rugosum					0				0			
Grindelia squarrosa var. squarrosa		0	95.									
Helianthus annuus										0		
Helianthus hirsutus									0			
Heterotheca subaxillaris										0		
Kuhnia eupatorioides var. corymbulosa	Sa		0							,		

Table 7. Continued

						Si	Sites					
Species	DrM	DrD	Bk	Ka	١٧	ಕ	BFM	BfD	St	Sa	Orm OrD Bk Ka Iv Ch BfM BfD St Sa ThC ThB	ThB
Lactuca floridana					0			N1 PRAGUI	0		· •	
Lactuca pulchella									0			
Lactuca serriola												0
Solidago canadensis									0			
Solidago missouriensis		0						0				ěl.
Vernonia baldwini var. interior		0	0		0			0	0			

as classified by Wilde and Lafond (1967). Ectocellular mycorrhizas develop short roots with a thick mantle and a Hartig net, intercellular mycelium forming in the cortex. Endocellular mycorrhizas lack the short roots but have intracellular hyphae. An irregular morphology is typical of ectoendotrophic mycorrhizas and may be characterized by an overproduction of mantle and Hartig net, an absence of the mantle, or the presence of both intercellular and intracellular mycelium with dichotomous short roots.

<u>Pinus sylvestris</u> best exhibited the phenomenon which Robinson (1967) terms dimorphism, that is, the repeated dichotomous divisions of the apical meristem before much growth can occur. A series of short, often much branched, mycorrhizal roots develop along the lateral roots as seen in Fig. 1. A transverse section, Fig. 2, shows the external sheath and Hartig net.

Ectocellular mycorrhizal short roots, a result of slower growth rate and differentiation of vascular tissue nearer the apex, were seen in Ostrya virginiana (Fig. 3) which showed negligible dimorphism. The mycelial covering of the root and some projecting hyphae were also seen. The fungi associated with ectocellular mycorrhizas are usually Basidiomycetes (Robinson, 1967) and Fig. 4 shows mycelial clamps characteristic of these fungi.

All mycorrhizal tree species were checked at least twice and some more often. Specimens were collected from both north- and south-facing slopes of the Stony Steep Land site (St) as well as the up and down slope sides of the tree-root systems. Microscopic examinations revealed that these factors did not affect the results which were consistently positive.

# Nonmycorrhizal Species

Although mycorrhizas were not found in grasses, grass-like plants, woody shrubs or forbs, hyphae were often found entangled with the roots of these plants. In <u>Buchloe dactyloides</u>, <u>Panicum lanuginosum</u>, <u>Panicum oligosanthes</u>, and <u>Panicum virgatum</u>, a particular hyphal type consistently occurred around

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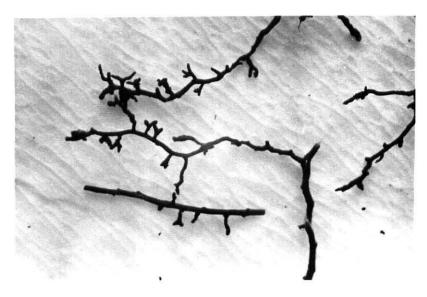


Fig. 1. Mycorrhizal roots of Pinus sylvestris showing typical dimorphic or "coralloid" mycorrhizas.

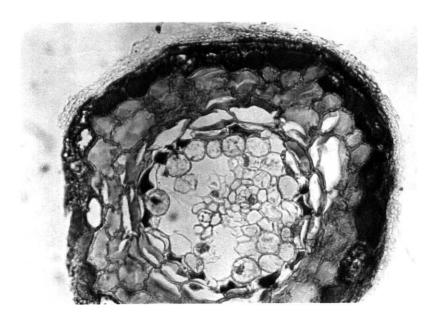


Fig. 2. Transverse section of ectoendotrophic root of Pinus sylvestris exhibiting enlarged cortical cells with both intracellular and intercellular mycelia.

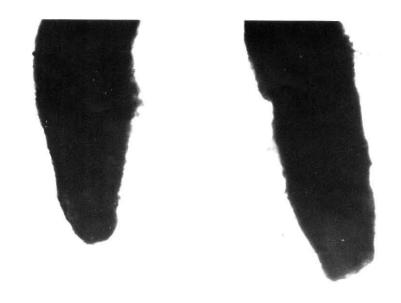


Fig. 3. Mycorrhizal short roots of Ostrya virginiana showing external hyphal sheath.

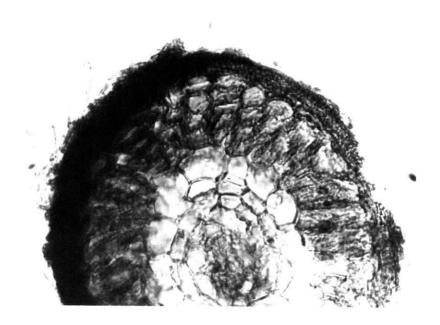


Fig. 4. Transverse section of Quercus macrocarpa with mycorrhizal sheath and hyphae exhibiting clamps, typical of Basidiomycetes.

but not attached to the roots. Usually only a single layer thick, the hyphae were easily distinguished from root tissue. These peripheral hyphae, sometimes referred to as rhizoclena, would be worth studying to learn if these fungi are important to the success of these grasses,

of the 157 species collected in one or more sites, all individuals of each species were consistently mycorrhizal or nonmycorrhizal except for Quercus prinoides and Juniperus virginiana. The only specimen of the redcedar to lack mycorrhizas was a tree less than lm tall in a grassland far from other trees. Two of the four Q. prinoides specimens showed no mycorrhizas but both nonmycorrhizal specimens were older root parts and collected at a depth of 40-50cm. Of the 147 nonmycorrhizal species only 4 showed a regular peripheral covering of hyphae and 30 species were found to show some hyphal presence but in an irregular pattern and amount. On 113 species few, if any, hyphae were seen. Since most species were negative, these findings give little evidence for or against the importance of soils and treatments on the occurrence of mycorrhizas.

# DISCUSSION

No species in prairie areas was mycorrhizal. A young juniper many meters from other trees lacked mycorrhizas. All other junipers checked were either in forests or at the edge of forests within a few meters of other trees. The absence of mycorrhizas on this young juniper in the prairie might be due to its young age or it might be because the mycorrhizal fungus was absent. Wilde (1968) supports as a possible explanation for lack of mycorrhizas the excretion of toxic substances by prairie plants which destroy mycorrhizal fungi that might otherwise be present, even in the absence of host plants (Curtis and Cottam, 1950; Persidsky, Loewenstein, and Wilde, 1965). They propose that these root exudates, composed of organic compounds, are inhibitors of fungus-root microorganism's physiological activities. Many nonmycorrhizal fungi must be

tolerant of these exudates because root specimens and prairie soils showed some hyphal presence.

Another possible explanation for the absence of mycorrhizas in the prairie species is the relationship found for mycorrhizas and soil fertility. Since the soils in this study are mostly Brunizem or Prairie soils, they can be expected to have an abundant supply of mineral nutrients. Previous studies indicate that mycorrhizas are formed mainly in humus-rich soils which are poor in nutrients (White, 1941; Robinson, 1967; Braga and Meyers, 1967; Went and Stark, 1968; Harley, 1969). Melin and Nilsson (1957) stated that without sufficient organic matter, mycorrhizas take sugars and growth substances from the roots and pass minerals, especially phosphorus, from the soil to the root, especially in soils deficient in minerals. In these soils with their abundant mineral and organic matter, the numerous roots and, in some cases, peripheral hyphae, would be adequate absorbers and the need for mycorrhizal functioning in nutrient absorption would be negligible.

Prairie soil texture apparently makes little difference in mycorrhizal presence or absence. The soils of the Dwight-Irwin complex are fine-textured with a thin, medium-textured surface layer and dense, fine-textured subsoils. No species collected on it or the stony Benfield-Florence complex of steeper slopes showed evidence of mycorrhizas. A third prairie soil, the Breaks-Alluvial complex, also did not exhibit support of mycorrhizal fungi in its deeper, silt loam surface layer. This agrees with results in Wisconsin on both fine- and coarse-textured soils (Wilde, 1968).

Many types of fungi are common to fertile soils and the local prairie soils were no exception. Soil particles were attached to roots by mycelia which also appeared to bind the soil particles together. <u>Buchloe dactyloides</u> and species of <u>Panicum</u> were particularly noticeable in the constant appearance of a particular type of hyphae around but not attached to the root. The epidermal layer

of the <u>Panicum</u> species was also characterized by numerous "outpocketings" or swellings on the external surface. These observations suggest that some prairie species may support a crypto-symbiosis type of rhizoclena.

Rhizoclena are superficial root-adherent mycelia contributing to mineral availability (Wilde and Lafond, 1967). These may be detectable and called phanero-symbiotic relationships or concealed and called crypto-symbiotic relationships in which the root epidermis is not penetrated by the extra-matrical, juxtaposed mycelia. The above mentioned grasses had extra-matrical mycelia but no mycelial sheath or other anatomical modifications. Perhaps these hyphae function to make available nutrients more accessible to these grasses rather than the usual function of making available nutrients that are present in deficient amounts. Perhaps these two genera produce fewer toxic exudates, thus supporting root-adherent mycelia.

Trappe (1969) has found that some fungi stimulate the growth of trees without actually forming mycorrhizas. This can help explain the lack of positive data on the trees surveyed in this area. Peripheral mycelia can perform the symbiotic function as effectively as do cortical mycelia (Wilde, 1954). They are known to contribute chelating agents which convert raw organic and mineral sources of nutrients into utilizable forms within the rhizosphere (Spryidakis, Chester, and Wilde, 1967). The growth of trees in soils lacking mycorrhizas may be the result of this type of fungal activity.

In forest sites, soil quality may be a decisive factor in the seeming absence of mycorrhizas. Since some positive mycorrhizal species were found at each forested site, it would seem feasible that other fungi capable of forming mycorrhizal relationships might be present. But because the soil is fertile and other fungi may stimulate tree growth without forming a mycorrhizal relationship, these fungi may not form such a relationship. This possibility needs further attention.

Quercus prinoides and J. virginiana were the only species that showed both positive and negative data. The fact that the juniper was a young tree and growing in the middle of a prairie where grass exudates might kill any mycorrhizas present might explain the absence of mycorrhizas. A possible explanation for the absence of mycorrhizas on Q. prinoides at two sites is the depth at which the roots were collected. On both sites, collections were obtained at 40-50cm depths and these roots were old. Gwynne-Vaughan (1937) reports that mycorrhizas have their greatest development on young roots, especially on those spreading horizontally in layers of decaying leaves. Hacskaylo (1971) notes that ectocellular associations, characteristic of woody plants, are usually confined to the upper several centimeters of the soil. A study of beech trees by Mystrik and Dominik (1969) indicated both depth and quantity of humus were influential factors. At a depth of 40-50cm the soil has less humus accumulation than aftthe surface.

This study indicates that in Riley County, Kansas, kind of soil and management practices have little, if any, effect on mycorrhizal relationships.

I found that many fungi which may have beneficial effects on the plant are in close association with the roots. It is interesting that <u>Juniperus</u> and species of <u>Quercus</u>, which showed positive mycorrhizal relationships in the forests, are the two most common trees in these ecosystems. The role of mycorrhizas in this frequency of occurrence needs further study.

In conclusion, I agree with Harley (1969) that the associations between roots and microorganisms which have been given the name mycorrhiza appear to be only the end terms of numerous less specialized tendencies for roots and microorganisms to set up balanced relationships with one another.

### SUMMARY

A survey of prairie and adjacent forest species was taken to determine

mycorrhizal presence or absence in order to better understand ecological interactions of these species in their ecosystems. Species showing mycorrhizas were:

Juniperus virginiana, Pinus ponderosa, Pinus strobus, Pinus sylvestris, Ostrya virginiana, Quercus borealis, Quercus macrocarpa, Quercus marilandica, and Quercus prinoides. Other trees, shrubs, grasses and grass-like plants, and forbs gave no positive data but indications of peripheral hyphae were seen on 4 grasses. Soil and management factors appear to have little effect in this particular location.

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Appendix Table 1. Scientific and common names of species studied, arranged by families.

	<u> </u>
Scientific Name	Common Name
Pinaceae	
Pinus strobus L.	White Pine
Pinus sylvestris L.	Scotch Pine
Pinus ponderosa Laws	Ponderosa Pine
Juniperus virginiana L.	Redcedar
Poaceae	
Andropogon scoparius Michx.	Little Bluestem
Andropogon gerardi Vitman	Big Bluestem
Bouteloua curtipendula (Michx.)Torr.	Sideoats Grama
Bouteloua hirsuta Lag.	Hairy Grama
Bouteloua gracilis (HBK.)Lag. ex Steud.	Blue Grama
Bromus purgans L.	Canada Brome
Bromus secalinus L.	Cheat
Bromus japonicus Thunb.	Japanese Brome
Buchloe dactyloides (Nutt.)Engelm.	Buffalo Grass
Cenchrus longispinus (Hack.)Fern.	Sandbur
Digitaria sanguinalis (L.)Scop.	Crabgrass
Elymus canadensis L.	Canada Wildrye
Eragrostis trichodes (Nutt.)Nash	Sand Lovegrass
Eragrostis spectabilis (Pursh)Steud.	Purple Lovegrass
Koeleria cristata (L.)Pers.	Prairie Junegrass
Leptoloma cognatum (Schultes)Chase	Fall Witchgrass
Muhlenbergia schreberi Gmel.	Nimblewill
Muhlenbergia racemosa (Michx.)BSP.	Green Muhly

Scribner Panicum

		The second secon
Appendix	Table 1	<ul> <li>Continued</li> </ul>
Unnellary	Idute	. Continueu

Panicum oligosanthes Schult.

Panicum lanuginosum Ell. Panicgrass

Panicum virgatum L. Switchgrass

Paspalum ciliatifolium Michx. Fringeleaf Paspalum

Poa pratensis L. Kentucky Bluegrass

Schedonnardus paniculatus (Nutt.)Trel. Tumblegrass

Setaria lutescens (Weigel) Hubbard Yellow Bristlegrass

<u>Setaria viridis</u> (L.)Beauv. Green Bristlegrass

<u>Setaria faberii</u> Herm. Giant Bristlegrass

Sorghastrum nutans (L.) Nash Indiangrass

Spartina pectinata Link Prairie Cordgrass

Sporobolus vaginiflorus (Torr.)Wood Puffsheath Dropseed var. neglectus (Nash)Shinners

Sporobolus asper (Michx.) Kunth var. asper Tall Dropseed

Sporobolus heterolepis Gray Prairie Dropseed

Sporobolus cryptandrus (Torr.)A. Gray Sand Dropseed

Tridens flavus (L.)Hitchc. Purpletop

Cyperaceae

Carex brevior (Dewey)Mackenzie Straw Sedge

Cyperus filiculmis Vahl Fern Flatsedge

<u>Cyperus schweinitzii</u> Torr. Schweinitz Flatsedge

Commelinaceae

Tradescantia bracteata Small. Bracted Spiderwort

Juncaceae

Juncus interior Wieg. Inland Rush

Liliaceae

Smilax hispida Muhl. Bristly Greenbriar

Salicaceae

Populus deltoides Marsh.

Eastern Cottonwood

Salix fragilis L.

Brittle Willow

Juglandaceae

Carya cordiformis (Wang.)K. Koch

Bitternut Hickory

Juglans nigra L.

Black Walnut

Betulaceae

Ostrya virginiana (Mill.)K. Koch

American Hophornbeam

Fagaceae

Quercus marilandica Muenchh.

Blackjack Oak

Quercus borealis Michx. var. maxima

(Marsh.) Ashe

Red Oak

Quercus macrocarpa Michx.

Bur Oak

Quercus prinoides Willd.

Dwarf Chinquapin Oak

Ulmaceae

Celtis occidentalis L.

Common Hackberry

Ulmus americana L.

American Elm

Moraceae

Maclura pomifera (Raf.)Schneid.

Osageorange

Morus alba L.

White Mulberry

Morus rubra L.

Red Mulberry

Urticaceae

<u>Laportea</u> canadensis (L.)Wedd.

Woodnettle

Parietaria pensylvanica Muhl. ex Willd.

Pellitory

Polygonaceae

Polygonum hydropiper L.

Marshpepper Smartweed

Chenopodiaceae

Chenopodium gigantospermum Aellen.

Mapleleaf Goosefoot

Chenopodium album L.

Lambsquarters

Kochia scoparia (L.)Schrad.

Kochia

Menispermaceae

Menispermum canadense" L.

Common Moonseed

Annonaceae

Asimina triloba (L.)Dunal

**Pawpaw** 

Saxi fragaceae

Ribes missouriense Nutt.

Missouri Gooseberry

Platanaceae

Platanus occidentalis L.

Sycamore

Rosaceae

Geum canadense Jacq.

White Avens

Prunus americana Marsh. var. americana

American Plum

Prunus angustifolia Marsh.

Chickasaw Plum

Prunus persica (L.)Batsch

Peach

Cassiaceae

Cassia fasiculata Greene

Showy Partridgepea

Cercis canadensis L.

Eastern Redbud

Gleditsia triacanthos L.

Common Honeylocust

Gymnocladus dioica (L.)K. Koch

Kentucky Coffeetree

Fabaceae

Amorpha fruticosa L.

Indigobush Amorpha

Amorpha canescens Pursh

Leadplant

Desmodium glutinosum (Muhl.)Wood

Largeflower Tickclover

Desmodium illinoense Gray

Desmodium paniculatum (L.)DC.

Lespedeza repens (L.)Bart.

Lespedeza capitata Michx.

<u>Strophostyles helvola</u> (L.)Britton

var. helvola

Strophostyles leiosperma (T. & G.)Piper

Linaceae

Rutaceae

Euphorbiaceae

Celastraceae

Linum sulcatum Riddell

Ptelea trifoliata L.

Zanthoxylum americanum Mill.

Acalypha ostryaefolia Riddell

Croton monanthogynus Michx.

Euphorbia nutans Lag.

Euphorbia hexagona Nutt.

Euphorbia dentata Michx.

Tragia urticifolia Michx.

Anacardiaceae

Rhus glabra L.

Rhus aromatica Ait. var. serotina

(Greene)Rehd.

Rhus radicans L.

Euonymus atropurpureus Jacq.

Aceraceae

Acer negundo L.

Illinois Tickclover

Panicled Tickclover

Creeping Lespedeza

Roundhead Lespedeza

Trailing Wildbean

Smoothseed Wildbean

Grooved Flax

di doved i lax

Common Hoptree

Common Pricklyash

Hophornbeam Copperleaf

Oneseed Croton

Spotted Euphorbia

Sixangle Euphorbia

Toothed Euphorbia

Nettleleaf Noseburn

Smooth Sumac

Aromatic Sumac

Poisonivy

Eastern Wahoo

Boxelder

Hippocastanaceae

Aesculus glabra Willd. var. sargentii Rehd. Western Buckeye

Vitaceae

Parthenocissus quinquefolia (L.)Planch.

Virginia Creeper

Vitis vulpina L.

Wild Grape

Vitis riparia Michx.

Wild Grape

Tiliaceae

Tilia americana L.

American Linden

Malvaceae

Abutilon theophrasti Medic.

Velvetleaf

Callirhoe alcaeoides (Michx.)A. Gray

Pale Poppymallow

Hypericaceae

Hypericum perforatum L.

Common St. Johnswort

Violaceae

Viola sp.

Violet

Oenotheraceae

Gaura parviflora Dougl.

Smallflower Gaura

Oenothera serrulata Nutt.

Serrateleaf Eveningprimrose

Cornaceae

Cornus drummondi Meyer

Roughleaf Dogwood

01eaceae

Fraxinus pensylvanica Marsh.

Red Ash

Asclepiadaceae

Asclepias stenophylla Gray

Narrowleaf Milkweed

Asclepias speciosa Torr.

Showy Milkweed

Convolvulaceae

Ipomoea hederaceae Jacq.

Ivyleaf Morningglory

Boraginaceae

Hackelia virginiana (L.)J.M. Johnston

Virginia Stickseed

Verbenaceae

Verbena urticifolia L.

White Verbena

Verbena stricta Vent.

Woolly Verbena

Lamiaceae

Agastache nepetoides (L.)Ktze.

Catnip Gianthyssop

Salvia azurea Lam. var. grandiflora Benth.

Pitcher Sage

Teucrium canadense L. var. virginicum (L.) Eaton

American Germander

Solanaceae

Physalis virginiana Miller var. virginiana

Virginia Groundcherry

Solanum nigrum L.

Black Nightshade

Solanum carolinense L.

Horsenettle

Scrophulariaceae

Scrophularia marilandica L.

Maryland Figwort

Verbascum blattaria L.

Moth Mullein

Bignoniaceae

Catalpa speciosa Warder

Catalpa

Phrymaceae

Phryma leptostachya L.

Lopseed

Rubiaceae

Cephalanthus occidentalis L.

Common Buttonbush

Caprifoliaceae

Symphoricarpos orbiculatus Moench

Buckbrush

## Campanul aceae

Campanula americana L.

American Bellflower

Asteraceae

Vernonia baldwini Torr. var. interior

(Small)Shubert

Baldwin Ironweed

Eupatorium altissimum L.

Eupatorium rugosum Houtt.

Tall Eupatorium

White Snakeroot

Kuhnia eupatorioides L. var. corymbulosa

T. & G.

Falseboneset

Aster drummondii Lindl.

Drummond Aster

Aster sericeus Vent.

Silky Aster

Aster ericoides L.

Heath Aster

Conyza canadensis (L.)Cronq.

Horseweed

Grindelia squarrosa (Pursh)Dunal

Curlycup Gumweed

var. squarrosa

Heterotheca subaxillaris (Lam.) Britt. & Rusby Camphorweed

Solidago missouriensis Nutt.

Missouri Goldenrod

Solidago canadensis L.

Canada Goldenrod

Ambrosia artemisiifolia L.

Common Ragweed

Ambrosia psilostachya DC.

Western Ragweed

Bidens bipinnata L.

Spanishneedles

Helianthus annuus L.

Common Sunflower

Helianthus hirsutus Raf.

Hairy Sunflower

Achillea millefolium L.

Common Yarrow

Artemesia ludoviciana Nutt. var. ludoviciana Louisiana Sagewort

Cirsium altissimum (L.)Spreng

Tall Thistle

Lactuca pulchella (Pursh)DC.

Chicory Lettuce

Lactuca serriola L.

Prickly Lettuce

Lactuca floridana (L.)Gaertn.

Florida Lettuce

# A SURVEY OF MYCORRHIZAS IN KANSAS BLUESTEM PRAIRIE AND ADJACENT FOREST

by

THERESE CATHERINE WETTA

B. S., Sacred Heart College, 1964

AN ABSTRACT OF A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Division of Biology

KANSAS STATE UNIVERSITY Manhattan, Kansas Occurrence of mycorrhizas in bluestem prairie and adjacent forest was surveyed in a variety of sites in Riley County, Kansas, in order to better understand the ecological interactions of species in these ecosystems. After careful removal of soil particles from roots in the laboratory, clearing and staining was done by soaking the fresh tissue overnight in a solution consisting of 4g NaOH and 0.4g bromothymol blue in 100ml H<sub>2</sub>O. Hand-sliced transverse sections and whole mounts were studied to determine presence or absence of mycorrhizas.

Of 31 tree species studied, the following 10 were found to possess mycorrhizas: Juniperus virginiana, Ostrya virginiana, Pinus ponderosa, Pinus strobus, Pinus sylvestris, Platanus occidentalis, Quercus borealis, Quercus macrocarpa, Quercus marilandica, and Quercus prinoides. No mycorrhizas were found in 16 shrubs, 39 grasses and grass-like plants, 23 composites, and 48 other herbs. Fungal penetration of root tissues determined a mycorrhizal relationship. In 4 grasses, Buchloe dactyloides, Panicum lanuginosum, Panicum oligosanthes, and Panicum virgatum, peripheral fungi were found in a regular pattern around the root, but the relationship was nonmycorrhizal. Some evidence of hyphae was seen regularly in 30 species but 113 showed none. Perhaps some of these fungi aid the plant similarly to mycorrhizas.

One young juniper showed negative results, perhaps due to its young age or its location in the prairie where the necessary fungus was absent. On 2 nonforest sites, Q. prinoides showed negative data but specimens were older roots and taken at a 40-50cm soil depth, conditions unfavorable for a mycorrhizal relationship.

Site selection included forest and prairie on both upland and lowland and a variety of soil types. Management variations of heavily grazed, moderately grazed, and ungrazed grasslands, burned and unburned grasslands, and grazed and ungrazed forests were studied. No differences were detected due to these

factors probably because most species lacked mycorrhizas.

Mycorrhizas are unnecessary for survival of prairie higher plants, perhaps because of the nutrient rich Brunizem prairie soils and the functioning of peripheral hypahe. The 2 most common trees in the upland forest of this area, juniper and oak, possess mycorrhizas.