#### THE EFFECT OF PARAQUAT AND NAPROPAMIDE APPLICATION ON THE

COLONIZATION OF SEEDLING ROOTS BY MYCORRHIZAL FORMING FUNGI 1/

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#### INTRODUCTION

That mycorrhizae are beneficial in a variety of situations has been well documented. Beneficial effects of herbicide usage are also well documented. These two management operations come together in a very unique way in the forest nursery. This is the place of seedling inoculation which will apparently have a continuous impact on future forest productivity of major proportions. The nursery is also the epitome of the most intensive cultural practices ever applied to the forest seedlings. Herbicides are an important factor in this cultural operation and promise even greater use potential with restrictive budgets and increasing labor costs. However, this interactive potential is essentially unexplored.

There are numerous reasons for examining this herbicide-mycorrhizal relationship, The mode of action of paraquat lends itself to application before tree seed germination, a preemergence treatment, as well as postemergent directed applications. Paraquat is not generally considered to have residual soil activity since it is adsorbed by clay. However, forest nurseries tend to be located on sandy soils where adsorptive sites are extremely limited. Lack of adsorption coupled with intense irrigation during seedling germination make it very possible to move the paraquat ions into the soil profile. This possibility along with the inherent mammalian toxicity of the herbicide, 120 mg/kg of paraquat ion, creates the potential of destroying the inoculated mycorrhizal fungi.

### OBJECTIVES

1. Determine the effects of paraquat and napropamide on the degree of hardwood seedling root colonization by the mycorrhizae forming fungi, <u>Pisolithus</u> <u>tinctorius</u> and <u>Glomus fasiculatus</u>.

2. Determine the effect of paraquat and napropamide on growth of seedlings inoculated or not inoculated with mycorrhizal forming fungi.

3. Correlate degree of mycorrhizal root colonization to physical growth parameters.

#### METHODS AND STATISTICAL DESIGN

The study is composed of two separate phases. Phase I is a greenhouse study designed to compare the growth response of mycorrhizal and nonmycorrhizal seedlings grown under controlled conditions and to determine the effects of a herbicide, paraquat, on the degree of mycorrhizal formation. Phase II is a field study which will both complement and supplement the greenhouse study and was installed at Vallonia, Indiana.

Only the Field Study will be reported.

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Field Study

Tree species in the tests were red oak <u>(Quercus rubra)</u> and black walnut <u>(Juglans</u> nigra).

Paraquat at 1 and 2 kg/ha active chemical or napropamide at 3 and 6  $_{kg/ha}$  active chemical were applied as preemergence treatments in May 1978 prior to seedling germination. Herbicide plots were 5 meters long and bed width.' Each treatment was replicated 3 times in a completely randomized design.

Inclusion of napropamide was based on 3 years of tests in Indiana state nurseries. This also provided a relative comparison on mycorrhizal affects and weed control.

within herbicide plots, prior to seeding or chemical treatment, plots 1/2 meter long and bed width were inoculated with mycorrhizal forming fungi. Beds to contain <u>Juglans nigra</u> (blank walnut) were inoculated with <u>Glomus fasiculatus</u> at a rate of 2000 spores/ft2, while beds to be planted to <u>Quercus rubra</u> (red oak) were inoculated with <u>Pisolithus tinctorius</u> at a rate of 200 ml/ft<sup>2</sup>

At the end of the growing season (October 3, 1978), ten seedlings from each inoculated or non-inoculated plot were sampled. Data included height, caliper, dry weight of stem and root, and degree of fungal colonization, i.e., mycorrhizal development.

Soil samples were taken before inoculation but after fumigation with methyl bromide and at time of harvest. The samples established fertility levels and fungal populations.

#### RESULTS

For red oak, the % root colonization was significantly greater in the inoculated control versus the non-inoculated control plots (Table 1). Percentage colonization in the inoculated plots was unaffected by napropamide even at the 6 lb. rate but decreased with additions of paraquat and was lowest for the combined Paraquat-napropamide treatment (2 and 6 kg active/ha, respectively). Percentage root colonization (Table 2) was significantly correlated with stem height (r = 0.574) and root/shoot ratio (r = 0.631).

Table 1. Effect of paraquat and napropamide on root colonization (%) of red oak seedlings by <u>Pisolithus</u> <u>tinctorius</u> fungus. (Levels of paraquat and napropamide are expressed in kg of active chemical per hectare).

				PARAQ	UAT		
		0		1		2	
N		_ <u>I</u>	<u>N-I</u>		<u>N-I</u>	I	N-I
N A P R O	0	24a*	13b	16b	12b	12b	10ь
P A M	3	22a	13b				
I D E	6	19ab	8bc			5c	5c

Values not followed by the same letter are significantly different ( $\alpha$  = .05)

Table 2. Correlation coefficients between percent root colonization by <u>Pisolithus tinctorius</u> fungi and growth variables for red oak seedlings.

Dependent Variable	Independent Variable	Correlation (r Value)
	Stem Height	a) 0.574* b) 0.423*
Percent Colonization	Stem Diameter	a) 0.115 b) 0.083
a) Inoculated plots	Stem Weight	a) 0.178 b) 0.163
b) All plots	Root Weight	a) 0.197 b) 0.236
	R/S Ratio	a) -0.631* b) -0.205

\*Correlations significant at  $\alpha$  = .05 level

Generally, the application of either herbicide or a rate increase resulted in a decrease in seedling height or stem dry weight when compared to inoculated control (Table 3 and 4). Seedlings in noninoculated plots responded similarly but appeared to be less sensitive to the chemical treatments. For all chemical treatments, on the average, the inoculated seedlings were larger. Root dry weight was unaffected by addition of chemicals but, on the average, seedlings in inoculated plots exhibited greater root dry weights.

Table 3. Effect of paraquat and napropamide on stem height (cm) of red oak seedlings inoculated (I) or not inoculated (N-I) with <u>Pisolithus tinctorius</u> fungus. (Levels of paraquat and napropamide are expressed in kg of active chemical per hectare).

				PARAQ	UAT		
		0		1		2	-
		I	N-I	I	N – I	I	N – I
N A P R	0	41.79a <sup>*</sup>	34.17ab	35.50ab	33.76ab	27.77b	34.82ab
0 P A M	3	36.32ab	33.87ab				
I D E	6	26.94b	19.87c			23.29bc	18.47c

Values not followed by the same letter are significantly different ( $\alpha = .05$ )

Table 4. Effect of paraquat and napropamide on stem weight (gm) of red oak seedlings inoculated (I) or not inoculated (N-I) with <u>Pisolithus tinctorius</u> fungus. (Levels of paraquat and napropamide are expressed in kg of active chemical per hectare).

				PARAQU	TAT			
		0		1		2	2	
М		I	N-I	I	<u>N-I</u>	I	<u>N-I</u>	
N A P R	0	3.75a <sup>*</sup>	3.00ab	3.01ab	2.71b	2.33b	2.18b	
O P A M	3	3.21ab	2.35b					
I D E	6	1.99bc	2.17b			1.80c	2.26b	

Values not followed by the same letter are significantly different ( $\alpha = .05$ )

For black walnut, the % colonization with G. <u>fasiculatus</u> of the control inoculated plots was significantly greater than the non-inoculated control. The addition of paraquat and/or napropamide did not significantly reduce the % colonization by G. <u>fasiculatus</u> except at 2 kg active/hectare of paraquat (Table 5).

Table 5. Effect of paraquat and napropamide on the colonization (%) of black walnut roots inoculated (I) or not inoculated (N-I) with <u>Glomus fasiculatus</u> fungi. (Values for paraquat and napropamide are given in kg of active chemical per hectare).

			PARA	QUAT		
	0		1		2	
	_I	<u>N-I</u>		N-I		<u>N-I</u>
0	16a <sup>*</sup> (26)	7b (17)	15a (28)	13a (30)	6b (22)	5b (6)
3	15a (18)	13a (21)				
6	12a (26)	5b (10)			14a (27)	9ab (13)
	3	I       0     16a*       (26)       3     15a       (18)	$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c cccc} 0 & & & \\ \hline I & N-I & I \\ 0 & 16a^{*} & 7b & 15a \\ (26) & (17) & (28) \\ 3 & 15a & 13a \\ (18) & (21) \end{array}$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$

Values not followed by the same letter are significantly different ( $\alpha$  = .05)

The seedlings were colonized by another species of Glomus sp. which could not be identified. The % colonization of this unknown species plus the % colonization by G. <u>fasiculatus</u> are represented by the parenthetical values (Table 5). Total colonization appeared to be unaffected by either chemical or the combined treatment. Significant correlations between percent colonization and the measured growth variables are presented in Table 6. The significant negative correlations indicate an inverse relationship between % colonization and stem and root weight. Possibly the greater the % colonization the more susceptible the seedling to herbicide injury.

# Table 6. Correlation coefficients (r values) between the physical growth parameters and percent colonization total and with G. <u>fasiculatus</u>.

	Co1	onization (%)
	Total	<u>Glomus</u> fasiculatus
Stem Height	-0.143	0.372
Stem Diameter	0.085	0.151
Stem Weight	-0.577*	0.213
Root Weight	-0.641*	-0.591*
R/S Ratio	0.099	0.072

Significant correlations at  $\alpha = .05$ 

stem weight and height of inoculated plots were significantly increased by applications of 1 kg active/acre paraquat or 3 kg active/ acre napropamide. Both variables were significantly reduced by the 2 kg active/acre paraquat and 6 kg napropamide combination treatment (Table 7 and 8).

Table 7. Effect of paraquat and napropamide on the stem weight (gm) of black walnut seedlings inoculated (I) or not inoculated (N-I) with <u>Glomus fasiculatus</u> fungi. (Values for paraquat and napropamide are kg of active chemical per hectare).

				PARAQ	TAU			
		0			1	2		
N		I	<u>N-I</u>	I	N-I	I	<u>N-I</u>	
A P R	0	27.0a*	22.4ab	27.6a	15.6b	15.3b	15.2b	
N A P R O P A M I D E	3	27.0a	14.4b	-				
I D E	6	20.5ab	19.7ab			19.6ab	17.3b	

Values not followed by the same letter are significantly different ( $\alpha = .05$ )

Table 8. Effect of paraquat and napropamide on the stem height (cm) of black walnut seedlings inoculated (I) or not inoculated (N-I) with <u>Glomus fasiculatus</u> fungi. (Values for paraquat and napropamide are kg active chemical per hectare).

	0			1		2
	_ <u>I</u>	<u>N-I</u>		N-I	I	N-I
0	50b <sup>*</sup>	45bc	83a	60.5ab	53b	45bc
3	73a	68ab				
6	63ab	54b			35c	34c

Values not followed by the same letter are significantly different ( $\alpha$  = .05)

Root dry weight did not differ significantly between inoculated and non-inoculated plots for a given herbicide treatment except for paraquat at 1 kg active/ha and napropamide at 3 kg. For all herbicide treatments, on the average, the root weights of inoculated seedlings was greater (Table 9). Both stem and root weight was significantly correlated with % colonization of G. <u>fasiculatus</u>. Table 9. Effect of paraquat and napropamide on the root weight (gm) of black walnut seedlings inoculated (I) or not inoculated (N-I) with <u>Glomus fasiculatus</u> fungi. (Values for paraquat and napropamide are given in kg of active chemical per hectare).

	(	)		1	2	
		<u>N-I</u>	I	N-I	I	N- I
	10.8b <sup>*</sup>	8.9bc	12,6ab	12.2ab	7.6bc	5.5
3	14.4a	12.3ab				
1	12.5ab	10.7b			5.9c	5.1

Values not followed by the same letter are significantly different ( $\alpha = .05$ )

## DISCUSSION

Biologically significant mycorrhizal colonization of hardwood seedlings in the nursery can be achieved by inoculation with <u>Pisolithus</u> <u>tinctorius</u> or <u>Glomus fasiculatus</u>. The level of colonization (24% for P. <u>tinctorius</u> and 16% for G. <u>fasiculatus</u>) is relatively low compared to the results reported for other species. These low levels may be attributed to the high fertility levels maintained throughout the study period especially for available P (Table 10).

# Table 10. Soil chemical analysis of the Vallonia tree seedling nursery.

	Dat	e
Component	5/20/78	10/3/78
$NO_3 - N$	17.0	11.5
NH4 - N	14.6	13.2
Р	53	68
К	380	340
Ca	1240	1180
Mg	210	206
pH (water)	5.8	5.6

Chemical (values in ppm)

Similarly, the lack of significant correlations between percentage colonization and the physical growth parameters for either tree species could be influenced by the relatively high level of available nutrients. It has been documented that percentage colonization and the subsequent correlation to plant growth parameters is poor for plants grown under fertile conditions.

The large seeded species such as walnut and red oak have relatively large quantities of stored "food" in the cotyledons and may not be as receptive to mycorrhizal colonization as many of the small seeded species. Consequently their dependency on available nutrients from the soil may not be as important as small seeded species that demonstrate a higher receptivity to mycorrhizal formation and subsequent growth correlations.

#### SUMMARY

Biologically significant mycorrhizal colonization of roots of red oak and black walnut can be obtained with <u>Pisolithus tinctorius</u> and Glomus <u>fasiculatus</u>, respectively. Correlation between percentage colonizaon and the growth parameters is low but may be attributed to the seed characteristics of these species in combination with high fertility levels maintained throughout the study.

Chemical weed control with paraquat or napropamide can reduce the percentage of root colonization by these mycorrhizal forming fungi but paraquat is more effective.