SOIL TEMPERATURE, GROWTH AND ECTOMYCORRHIZAL RELATIONSHIPS OF *QUERCUS VELUTINA* SEEDLINGS

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Abstract.--Black oak (*Quercus velutina* Lam.) seedlings inoculated with *Pisolithus tinctorius* (Pers.) Coker and Couch were grown in "air baths" to assess the influence of soil temperature on ectomycorrhizal development, root and shoot growth. The four soil temperature treatments imposed on the seedlings were 18°, 23°, 28° and 33°C. After an 18 week propagation period, soil temperature had significantly influenced both seedling growth and ectomycorrhizal development. As soil temperature was increased from 18° to 33°C, seedling root and shoot growth decreased. However, ectomycorrhizal development increased as the soil temperature increased with significant infection occurring only at 28° and 33°C. In a second experiment, *Pisolithus* inoculated and noninoculated black oak seedlings were grown in two soil temperature regimes, 26° and 31°C. Significant increases in seedling ectomycorrhizal development and root system length were found in the 31°C soil temperature regime. Soil temperature did not significantly affect seedling shoot growth.

Keywords: *Quercus velutina*, *Pisolithus tinctorius*, soil temperature

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INTRODUCTION

Producing oak (Quercus) seedlings in containers offers possibilities for increasing the slow growth that typically follows field planting (Johnson, 1979). However, problems in producing container-grown oaks include maximizing total root growth within the confines of a practical container volume, and balancing root and shoot development (Johnson, 1974). Although numerous environmental factors influence seedling development, manipulation of soil temperature may be one of the most useful techniques for balancing root and shoot growth and simultaneously stimulating ectomycorrhizal development in this genus. Maximizing ectomycorrhizal development of oak seedlings may be an effective method of increasing the root absorptive surface area, as well as providing other physiological benefits, which improve growth of newly planted oaks (Garrett et al., 1979).

Soil temperature has a pronounced effect on the root relationships and shoot growth of oak. Larson (1974) observed that shoot growth of northern red oak (*Quercus rubra* L.) was best at soil temperatures of 24°C but was greatly reduced below 18°C even though ambient air temperatures remained favorable. Larson (1974) and Teskey (1978) respectively found in red and white oak (*Quercus alba* L.) that root initiation and growth increased as temperatures increased from 13° to 24°C. Similarly, Malyshkin (1955) observed that some ectomycorrhizal infection occurred on oak seedlings at soil temperatures of 12-14°C. However, when the soil temperature was increased to 16-18°C, the intensity of ectomycorrhizal infection doubled.

In the two experiments reported here, we investigated the influence of growth medium temperature on ectomycorrhizal development and shoot and root growth of black oak (*Q. velutina* Lam.) seedlings inoculated with the fungal symbiont *Pisolithus tinctorius* (Pers.) Coker and Couch.

MATERIALS AND METHODS

Two investigations were conducted in adjacent glasshouses at the University of Missouri-Columbia. Unless otherwise stated, procedures for both studies were identical. All seedlings were exposed daily to 14-16 hours of sunlight under 50% green polypropylene shade cloth. Ambient greenhouse temperatures ranged from 21-33°C, and relative humidity ranged from 40-80%.

Black oak acorns were collected from six trees, surface sterilized in a 10% sodium hypochlorite solution and planted in Spencer-LeMaire Super 45 containers. Each container was divided into three 750 cc seedling cavities, which were filled with a 1:1 mixture of sterilized peat moss and vermiculite. The growth medium was inoculated with Pisolithus tinctorius by thoroughly mixing it with 17 cc of vegetative mycelia. The inoculum was produced during the previous four months in a mixture of vermiculite, peat moss, and modified Melin-Norkrans solution using methods described by Marx and Bryan (1975).

Four "air baths" were used to control the growth medium temperature of the study seedlings. The 0.4 m x 1.2 m x 2.7 m air baths consisted of plywood boxes that were heated or cooled by circulating fans and heating and cooling coils housed at the end of each insulated box (Behrns, 1979). Growth medium temperature was monitored continuously with thermistors and maintained within ± 1°C of a selected temperature within each box. Prior to fungal inoculation and acorn planting, the containers were sealed in the four wood frame air baths. In order to assure uniform spacing for all seedlings and adequate air flow around the containers, a 4 cm plywood strip was placed between rows of the containers. The containers were supported by perforated pressboard which permitted water drainage and allowed air to circulate throughout the bath.

In experiment I, four growth medium temperatures were imposed on Pisolithus inoculated seedlings: 18, 23, 28, and 33°C. Each air bath contained 256 seedlings and was arranged in a randomized complete block design with replications. Each temperature bath was considered a separate experiment in a series of four experiments (Snedecor and Cochran, 1976; page 375). To test differences among baths, the pooled error mean square from the separate experiments (baths) was used after testing for the equality of the error mean squares. Each of the seedlings was fertilized every three weeks with 75 ml of reagent grade nitrogen-phosphorus-potassium (NPK) solution (6-6-12 ppm). In experiment II, both Pisolithus inoculated and noninoculated seedlings were grown at growth medium temperatures of 26 and 31°C. Two air bath boxes were held at each temperature and half of the seedlings in each box were inoculated. This experiment was arranged in a randomized complete block design with four replications containing 74 inoculated and noninoculated seedlings each. During experiment II, the NPK (6-6-12 ppm) solution was applied bi-weekly to the growth medium. All seedlings received a uniform micronutrient solution weekly (Hewitt, 1966) and were watered as needed.

At the end of 18 weeks, the growth medium was washed from the roots, and the following measurements were made: shoot length and dry weight, diameter at the root collar, root dry weight, number of primary laterals, and number of primary laterals infected with Pisolithus. Data were analyzed by analysis of variance and the least significant difference test.

RESULTS

Experiment I

After an eighteen week propagation period, seedling survival was 100 percent in all four temperature regimes. However, the growth and morphology of the black oak seedlings were strongly influenced by the growth medium temperature. All morphological characteristics of the seedlings, with the exception of leaf area and shoot length, decreased in size or number as temperature increased (Table 1). Shoot length was the only seedling variable which did not differ significantly between temperature regimes. Leaf area increased as growth medium temperature increased up to 28°C and then decreased as temperatures increased to 33°C. Shoot dry weight and root collar diameter also significantly decreased as substrate temperatures increased. Seedling root weight and number of primary laterals at 18°C were nearly double that observed at 33°C.

Temperature significantly influenced the degree of ectomycorrhizal development by *Pisolithus tinctorius* (Table 1). No *Pisolithus* was found on seedlings grown at 18 and 23°C; small amounts of ectomycorrhizae were identified at 28°C; and significant amounts were present at 33°C. Over one-third of the primary laterals were colonized by *Pisolithus* at 33°C, while less than five percent of the laterals were ectomycorrhizal in the 28°C temperature regime. Ectomycorrhizal infection per seedling was variable ranging from less than 10 percent to 80 percent at 33°C. Microscopic examination of ectomycorrhizal roots revealed a greater mantle thickness and more expansive Hartig net development on seedlings grown at 33°C than those grown at 28°C.

Experiment II

The growth of *Pisolithus* inoculated and noninoculated black oak seedlings was influenced by both temperature and degree of ectomycorrhizal development (Table 2). Seedlings grown at both temperatures with *Pisolithus* ectomycorrhizae were taller, heavier, and had greater leaf surface area and root system development than the noninoculated control seedlings. Seedlings with *Pisolithus* ectomycorrhizae in the 31°C temperature regime developed significantly greater shoot and root weights and leaf surface area than those without ectomycorrhizae. Among the noninoculated seedlings, average growth for all measured variables decreased with increasing growth medium temperatures. Root system weights of the noninoculated seedlings grown at 26°C were also significantly greater than those grown at 31°C. Moreover, shoot length was greater at 26°C than at 31°C; however, the difference between the two treatments was not significant.

Significant amounts of *Pisolithus* ectomycorrhizae were observed on seedlings grown in both the 26 and 31°C temperature regimes (Table 2). However, ectomycorrhizal development at 31°C was two and one-half times greater than that observed in the 26°C regime. Ectomycorrhizal infection per seedling ranged from 20 to 80 percent at 31°C and from less than 5 to 40 percent at 26°C. No ectomycorrhizal structures were observed in the noninoculated treatments.

Table 1. Growth and ectomycorrhizal development of *Quercus velutina* seedlings colonized with *Pisolithus tinctorius* at four growth medium temperatures (Experiment I).

Seedling Growth Variable	Growth Medium Temperature °C			
	18	23	28	33
Shoot length (cm)	10.8a[a/]	11.2a	10.8a	10.8a
Leaf area (cm^2)	105.1a	141.2b	151.3b	137.5b
Root collar diameter (cm)	0.40a	0.37ab	0.35bc	0.34bc
Shoot dry weight (g)	0.38a	0.34ab	0.31bc	0.28c
Root dry weight (g)	2.95a	2.72a	2.18b	1.49c
Primary laterals (number)	35.0a	23.0b	25.0b	18.0c
Ectomycorrhizal laterals (%)	---	---	<5.0b	37.4a

[a/] Treatment means within a given variable that are followed by a common letter do not differ significantly at the 5 percent level based on the least significant difference test.

[b/] Percent of primary lateral roots with at least one *Pisolithus* infection point.

Table 2. Growth and ectomycorrhizal development of *Pisolithus* inoculated and noninoculated *Quercus velutina* seedlings at two growth medium temperatures (Experiment II).

Seedling Growth Variable	Growth Medium Temperature 26°C		31°C	
	Pisolithus inoculated	Noninoculated	Pisolithus inoculated	Noninoculated
Shoot length (cm)	12.7a[a]	12.2a	12.4a	12.1a
Leaf area (cm^2)	184.2b	165.0b	221.2a	186.9b
Root collar diameter (cm)	0.43a	0.43a	0.43a	0.44a
Shoot dry weight (g)	0.92 a	0.94a	0.93a	0.85b
Root dry weight (g)	3.76a	3.61a	3.52a	2.55b
Primary laterals (number)	37.0a	30.0bc	34.0ab	27.0
Ectomycorrhizal laterals (%)[b]	21.0b	---	53.9a	---

[a] Treatment means within a given variable that are followed by a common letter do not differ significantly at the 5 percent level based on the least significant difference test.

[b] Percent of primary lateral roots with at least one *Pisolithus* infection point.

DISCUSSION

A single optimum growth medium temperature for shoot and root growth of black oak seedlings was not clearly evident from our data. Reports in the literature also vary widely as to the influence of soil temperature on oak root growth. Larson (1974) observed that white oak roots grew well at 24°C. Howver, earlier reports by Larson (1970) and Bowen (1970) stated that numbers of newly initiated laterals in controlled environments increased in northern red oak as soil temperature approached 30°C. Teskey (1978) found that, under field conditions, roots of white oak grew most rapidly when soil temperatures ranged from 18 to 20°C. The results from both of our experiments indicated that maximum root growth probably occurs in the 18-26°C temperature range.

Gur et al. (1972) proposed that maximum shoot growth of woody plants generally occurs at the same temperature at which maximum root growth occurs. Our results appear to agree with this hypothesis. Shoot growth of both inoculated and noninoculated black oak seedlings was associated with increasing root growth within all temperature regimes. The largest seedlings were produced in the cooler growth medium temperatures (i.e., 18 and 26°C). Larson (1970, 1974) and Kramer (1958) also found that initial shoot growth of white and red oak seedlings was most rapid at soil temperatures below 26°C. However, in both experiments of the present study, leaf area of the mycorrhizal and nonmycorrhizal seedlings increased as growth medium temperatures increased to near 30°C. Similarly, Larson (1970, 1974) concluded that maximum leaf area development of white and northern red oak occurred at warmer soil temperatures (i.e., 28-30°C).

In experiment I, no significant amounts of *Pisolithus* ectomycorrhizae were detected at temperatures below 33°C. Moreover, ectomycorrhizal development was more than doubled as a result of the warmer growth medium temperature in experiment II. Marx et al. (1970) also found that *Pisolithus tinctorius* formed significantly greater amounts of ectomycorrhizae on *Pinus taeda* L. at high substrate temperatures of 34°C. Our observations agree with earlier reports from the Russian literature which suggest ectomycorrhizal development on oak is significantly improved as soil temperature increases (Malyshkin, 1955). Differences in seedling ability to form ectomycorrhizae at similar growth medium temperatures (i.e., 28 and 26°C in experiment I and II, respectively) were evident. The differential response of *Pisolithus tinctorius* to similar growth medium temperatures may be a result of relatively greater seedling size and vigor in experiment II. Seedlings in experiment II received the standard nutrient solution on a more frequent basis than experiment I and corresponding differences in seedling growth and vigor were evident. This suggests that even though a fungal symbiont like *Pisolithus tinctorius* may have a symbiotic potential over a wide range of temperatures, the degree of benefit to be gained by the host is determined by the physiological limitations of the host.

In the absence of ectomycorrhizae, the poor growth of black oak at high temperatures suggested that ectomycorrhizae may be essential to seedling survival and growth in soils of high temperature. Schramm (1966) also found that red and black oak seedlings with Pisolithus ectomycorrhizae were larger than control plants growing in anthracite mining spoils where soil surface temperatures approached 60°C. Marx et al. (1970) and Marx and Bryan (1971) observed that Pinus taeda seedlings infected with Pisolithus had a 96 percent survival rate at substrate temperatures of 40°C. They concluded that Pisolithus tinctorius adapts well to supra-optimal soil temperatures and can increase seedling tolerance to high soil temperatures. Our results also indicated that Pisolithus increases oak seedling tolerance to warm soil temperatures.

SUMMARY

Although cooler soil temperatures may favor maximum black oak root and shoot growth under controlled environmental conditions, warmer growth medium temperatures favor ectomycorrhizal development by Pisolithus tinctorius. Reports by Garrett et al. (1979) and Shemakhanova (1962) indicate that abundant ectomycorrhizal development on oak seedling root systems result in substantially improved growth following outplanting. Johnson (1979) and others have determined that producing oak seedlings of large size alone does not solve the problem of slow initial growth in the field. Rapid juvenile growth of newly planted oaks is dependent upon a well developed physiologically active root absroptive area. Therefore, it may be advantageous to provide warmer growth medium temperatures to improve ectomycorrhizal development of inoculated, container-grown oak seedlings during the seedling propagation phase.

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