INFLUENCE OF FAMILY GENOTYPE AND SOIL pH ON GROWTH AND CHLOROSIS OF PIN OAK SEEDLINGS[1]

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Abstract.--The progenies of 14 families of pin oak (Quercus palustris Muenchh.) were grown for 18 weeks in sand culture in the greenhouse. Seven green and seven chlorotic phenotypes, growing along city street parkways, were selected as parents. Seedling progeny were arranged in a replicated factorial design and watered daily with a modified Hoagland's solution adjusted to pH 4.0, 5.5 or 7.0. This solution contained adequate concentrations of all nutrients except iron which was limited to 3 ppm.

The degree of chlorosis varied significantly among families grown at pH 4.0, and 5.5 but not at 7.0. In general, chlorosis increased with increasing pH. Height growth of progeny of different parents varied significantly for the different pH treatments. Overall seedling height declined significantly with increasing pH.

Progenies from green and chlorotic parents were combined separately and compared. Seedlings from green parents were significantly greener than those of chlorotic parents only at pH 7.0. However, height growth of seedlings of chlorotic parents was significantly greater at this pH. No significant effects were found at pH 4.0 and 5.5.

INTRODUCTION

Pin oak has been widely planted as an ornamental throughout the midwest and northeast because it has a desirable form and leaf texture. Many trees have been planted by homeowners and by municipalities along city street parkways as is common in Fort Wayne, Indiana where this study was conducted.

Pin oak is confined to wet, acidic sites in its native habitat and these conditions enhance the uptake of reduced iron and several other trace elements. Offsite plantings in more aerated and alkaline soils often result in severe iron chlorosis and premature death (Neely 1973).

In a geographic study on pin oak, Berrang and Steiner (1980) found significant differences in iron chlorosis among geographic populations and among offspring of parents within populations. Populations from the northwest portion of the range were generally more resistant to chlorosis. Since parental effects are often large, selecting outstanding parent trees growing in stressed city habitats may prove to be effective for producing chlorosis resistant seedlings. Parkways represent stressful sites since they are often composed of heterogenous soils which are generally infertile and alkaline in pH in this locality.

The objectives of this study were to assess the genetic variation of iron chlorosis of offspring from selected parents of pin oak and to determine the effects of iron deficiency on seedling growth.

MATERIALS AND METHODS

A total of 153 trees, the majority growing in city street parkways, were rated for chlorosis during the summer of 1984. Trees were scored on a 1 to 10 scale. Individuals exhibiting severe overall chlorosis and necrosis were rated 1 and trees with all dark green leaves were rated 10.

The 1984 acorn crop was generally good and seed was collected from 80 trees, but many highly chlorotic individuals did not produce sufficient seed to be included in the study. In addition,

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four soil samples, 0.2cm diameter x 12-14cm long were obtained under each tree. The 3cm surface portion of the sample was excluded. The soil core samples were equally spaced around the tree and located 3/4 of the distance out from the center of the crown radius when possible.

Filled acorns were placed in perforated plastic bags filled with moist peat moss and stored under refrigeration.

Seeds of seven green parents and seven chlorotic parents were selected for study. The remaining seedlots were outplanted on a nearby alkaline site. However, this field experiment was terminated because of poor survival due to drought.

The 14 seedlots selected for this study were germinated in mid-May and grown in 2 liter pots filled with acid-washed silica sand. Seedlings were grown in a greenhouse for 18 weeks under natural daylight photoperiods and near ambient temperatures. They were watered daily with an excess of modified Hoagland's solution containing 156 ppm nitrogen and 3 ppm iron (Hoagland and Arnon 1950). The solution was adjusted to pH 4.0, 5.5 and 7.0 by the addition of H_2SO_4, KOH and 3:1 saturated $Ca(OH)_2$ solution to 1M KOH, respectively.

For the first 3 weeks of growth, all seedlings were watered with a nutrient solution adjusted to pH 5.5 and containing 1 ppm Fe EDTA. At the end of this period, the cotyledons were removed, and 5 seedlings of each family were assigned one of the pH treatments.

After 6 weeks of growth, many seedlings became chlorotic including several red oak seedlings of the same age. Therefore, the iron content of the nutrient solution was increased to 3 ppm using a combination of 2 ppm Fe EDTA and 1 ppm $FeCl_3$.

The study design was a 14 x 3 factorial arrangement (14 families and 3 pH levels) with 5 seedling replications for each pH level. Pots were randomly arranged within pH treatments.

At the completion of the study, seedlings were measured for stem height and visually rated for chlorosis. Leaf color was scored by two people and averaged for subsequent analysis. The last flush of growth was scored using a 1 to 10 scale as described earlier.

Data were analyzed by analysis of variance, and variation about the ranked means was examined by Duncan's New Multiple Range Test. T-tests were used to compare pooled means of offspring from chlorotic and normal parents.

RESULTS AND DISCUSSION

Chlorosis.--Differences in chlorosis ratings due to solution pH were significant ($P < 0.01$). As expected, the severity of chlorosis increased as pH increased. Family differences within a pH treatment varied significantly at pH 4.0 and 5.5 but not at 7.0 (Table 1).

Table 1.--Influence of pH and iron stress on chlorosis and height of pin oak families.[1]

	pH of nutrient solution		
	4.0	5.5	7.0
	(mean chlorosis rating)[2]		
chlorosis rating	7.44*[3]	36.27**	5.04
	(mean height [cm])		
stem height	29.6**	28.0*	27.0*

1 Each value is a mean of 70 trees (14 families x 5 replicates).
2 severe chlorosis = 1; dark green = 10.
3 *, ** = Difference among families significant at the 0.05 and 0.01 level of probability, respectively.

At pH 4.0 the overall mean was 7.4. Individual family chlorosis ratings ranged from 6.0, which is somewhat less than 50% of the leaf surface showing some chlorosis, to 9.6 in which the majority of the leaves are dark green. At pH 5.5 and 7.0 the mean chlorosis ratings decreased to 6.3 (5.2-7.0) and 5.0 (4.0-6.2), respectively.

Chlorosis ratings of parents appear to have little value in predicting the chlorosis value of progeny at pH 4.0 (Table 2). Approximately equal numbers of chlorotic and green parents are found above and below the treatment mean value of 7.4. However, as iron stress increases, 5 of 8 families of green parents were above the treatment mean at pH 5.5, and at pH 7.0 4 of 6 families above the mean were offspring of green parents.

The rankings of families varied considerably especially between pH 4.0 and 5.5. However, with increasing iron stress at the higher pH treatments, a majority of the same families remained above or below the treatment means. Across all solution reactions 3 families, 4, 10, 12, were consistently above the treatment means and 3, 6, 8, 9 remained below the means.

In the case of parent 4, which was considerably chlorotic, (60% to 70% of the leaf area was yellow) the above average performance of its offspring was somewhat unexpected. However, chlorosis may be due to several different environmental factors other than the potential to adsorb iron from alkaline soils. Additional factors that may contribute to chlorosis are: high levels of soil P or other mineral deficiencies

Table 2.--Mean chlorosis ratings of half-sib families grown at 3 nutrient solution levels.

Parent no. and chlorosis rating	Parent soil pH	pH of nutrient solution 4.0	5.5	7.0
		mean chlorosis rating[2]		
6-2[1]	7.6	6.0[a]	6.2ab[3]	4.6a
3-2	7.7	6.2a	5.2a	5.0a
8-9	7.5	6.6ab	5.2a	4.4a
9-8	8.1	6.8ab	5.4ab	4.8a
5-2	7.4	7.0ab	6.0ab	5.2a
14-8	6.8	7.0ab	7.0b	5.6a
7-1	7.8	7.2ab	6.6ab	4.0a
13-9	7.7	7.2ab	6.8ab	6.0a
11-8	7.7	7.6abc	6.8ab	4.2a
12-10	7.3	7.6abc	7.0b	6.0a
10-9	7.7	8.0abc	6.4ab	6.2a
1-2	7.4	8.6bc	5.8ab	5.0a
2-2	7.8	8.8bc	6.4ab	4.2a
4-3	7.6	9.6c	7.0b	5.4a
$\bar{x}$	7.6	7.4	6.3	5.0

[1] chlorosis rating: 1 = severe chlorosis; 10 = dark green
[2] Each value is a mean of 5 trees.
[3] Means within a column followed by a common letter are not significantly different at the 5% level of probability trees with more consistent performance.

such as Mn and Zn (Messenger 1983), unfavorable soil texture and aeration, stress imposed by disease or insects and damaged or poorly developed root systems. In this study few feeder roots were observed in the soil samples of chlorotic trees, but roots were common in samples under healthy individuals. I presume that chlorosis eventually leads to a loss of root volume, but could other perturbations that result in the loss of roots cause chlorosis? The better than expected chlorosis resistance of progeny of parent 4 could also be due to favorable genetic contributions of male parent(s).

Parents 8 and 9, which exhibited healthy green phenotypes, produced progeny which performed below average. This suggests that these parents were not genetically superior in chlorosis

These results indicate that as iron stress increases, genetic differences in iron chlorosis

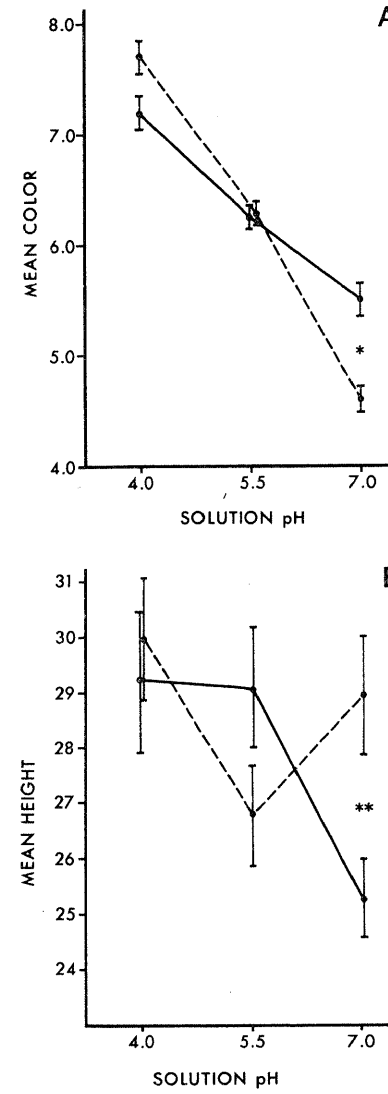

Figure 1.--Mean chlorosis rating and height of offspring of 7 green parents (——) and 7 chlorotic parents (---). Each point represents the mean of 35 trees and the length of the bar is the standard error. *, ** = Differences between means are significant at the 0.05 and 0.01 level of probability, respectively.

resistance and/or the pollen parent contributed to the lower level of chlorosis resistance of the progeny.

Progenies of chlorotic parents and green parents were combined separately and each pair of treatment means was analyzed by a Student's t-test. The only significant difference was found at pH 7.0 in which the seedlings of green rated parents were greener than those of chlorotic parents ($P < .05$) (Figure 1A). However, the selection of any

one parent does not insure more chlorosis resistant offspring.

These results indicate that as iron stress increases, genetic differences in iron chlorosis among and within families are more defined. It was not unusual for 1 out of 5 seedlings of any average family to vary widely in chlorosis resistance. Such variation suggests that full sib family selection may be necessary to obtain trees with more consistent performance. Since crossing in oaks would be slow and difficult, more immediate results could be made through vegatative propagation of superior individuals.

Stem Height.--The growth of seedlings from individual families varied significantly among solution pH treatments ($P<.01$) and within treatments (Table 1). Overall there was a general decrease in growth as pH increased. The mean family heights at pH 4.0, 5.5 and 7.0 were 29.6 cm, 28.0 cm and 27.0 cm, respectively. The greater supply of available iron at pH 4.0 improved both growth and uniformity of seedling height within families. As iron stress increased with increasing pH, genotypic differences in stress susceptibility were more clearly expressed and heights became more variable.

The rankings of families across pH treatments were inconsistent. Only families 8 and 11 remained consistently above the treatment means and families 3, 5, 13 and 14 were consistently below treatment means (Table 3).

The combined mean height of families derived from chlorotic parents (28.8 cm) was significantly greater than those of green rated parents (25.3 cm) at pH 7.0 (Figure 1B). Height differences at pH 4.0 and 5.5 were not significant. At pH 4.0 families of chlorotic parents were less than 1 cm taller than families of green parents. At pH 5.5 the order was reversed and the mean height of offspring from green parents was 29.1 cm compared to 26.8 cm for families of chlorotic parents.

Soil pH and Chlorosis.--The pH of 76 soil samples collected under parent trees was determined. The pH ranged from 6.10 to 8.10 and the mean was 7.55. Chlorosis ratings were found to be weakly negatively correlated with soil pH ($r^2 = -0.275$, $p < .05$). Thus darker green trees, with higher ratings, were generally associated with lower soil pH levels. This suggests that the selection of outstanding phenotypes growing in above average soil pH levels may be a useful aid in screening individuals for the genetic improvement of chlorosis resistance in pin oak.

In this study, chlorotic parents and green parents had identical mean soil pH levels of 7.6.

Table 3.-- Mean height of half-sib families grown at 3 nutrient solution pH levels.

Parent no. and chlorosis rating	Parent soil pH	pH of nutrient solution 4.0	5.5	7.0
		mean height (cm)[2]		
14-8[1]	6.8	24.2a	24.6a[3]	23.6ab
10-9	7.7	24.6a	32.8ab	22.2a
1-2	7.4	25.2ab	26.0a	30.6de
9-8	8.1	26.4ab	26.0a	28.2bcde
5-2	7.4	27.2abc	25.8a	24.0ab
7-1	7.8	27.2abc	27.6ab	33.2fg
3-2	7.7	29.2abc	26.6ab	20.4a
13-9	7.7	29.2abc	24.6a	24.6abc
6-2	7.6	31.0abc	26.2a	25.2abcd
11-8	7.7	31.2abc	30.4ab	30.4de
4-3	7.6	33.0abc	24.4a	37.2g
2-2	7.8	33.2abc	26.6ab	24.4ab
8-9	7.5	35.2bc	35.2c	30.0cde
12-10	7.3	37.2c	34.6bc	24.2ab
$\bar{x}$	7.6	29.6	28.0	27.0

[1] color rating: 1 = severe chlorosis; 10 = dark green
[2] Each value is a mean of 5 trees.
[3] Means within a column followed by a common letter are not significantly different at the 5% level of probability.

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