GENETIC VARIATION OF BLACK LOCUST SEEDLINGS ON RECLAIMED SURFACE MINE SOILS[1]

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Abstract.--Surface mining of coal frequently is done on forestry-based industrial lands in West Virginia. The resulting plant competition following reclamation inhibits the reestablishment of natural hardwood forests. Black locust (Robinia pseudoacacia L.) is a successful, fast growing hardwood when planted on these sites. Studies were established to determine how much genetic variability exists in black locust and which sources are the most desirable for planting on surface mined lands. Three experimental plantings have been established in Greenbrier County, WV, since 1984 from seed collected throughout the range of black locust. The oldest plantation has 30 seedlots from across the species' entire range. Genetic variation has been observed in leaf nutrient and chlorophyll variation. Height variation has also been observed. No geographic patterning has been associated with the variation. Selected seedlots can be identified which are expected to be superior sources.

INTRODUCTION

The state of West Virginia has an abundance of natural resources including forests and minerals. Over 75% of the state is covered with natural hardwood forests (Bones 1978). These forests may be managed for valuable hardwood lumber or converted to pine plantations for fiber production. Coal is another abundant natural resource in the state is frequently surface mined on forestry-based industrial lands.

Surface mines are recontoured soon after the mining operation is completed. The heterogenous artificial soils which cover the site are quickly regenerated with plants to prevent soil erosion. Herbaceous plants which are often used are legumes including vetch, clover, and Lespedeza and grasses (oats, fescue, and millet). These plants usually colonize and dominate the new sites very quickly, and the resulting plant competition inhibits the reestablishment of natural hardwood forests. Forestry operations on surface mined lands are greatly hampered due to the difficulty of forest regeneration, thus removing large acreages of forestry land from productivity for many years.

Numerous experimental trials of replanting hardwood and conifer species on reclaimed surfaces have been made. Black locust (Robinia pseudoacacia L.) has been demonstrated to be a successful, fast-growing hardwood when planted on these sites (Brown 1973). One of the most widely planted tree species in the world (Keresztesi 1980), it is a rapidly growing, shade-intolerant species which grows well on a range of soil qualities. The wood is naturally resistant to decay and resists swelling and contracting with moisture changes, and as a legume it improves soil quality by fixing atmospheric nitrogen. Black locust has been used in the forest industry as a source of railroad ties, fenceposts, mine timbers, stakes, boxes, and other items. Fencepost-sized trees can be grown in 15 to 20 years (Kellogg 1933). The wood has a high specific gravity of about 0.73 ovendry (Panshin and de Zeeuw 1970) and is one of the most efficient wood sources in the paper-pulping process.

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The best growth of black locust has been found in West Virginia (Fowells 1965). Because of its extensive use on strip mined soils, a great opportunity exists to improve the surface mined land quality while concurrently producing wood for wood products by establishing plantings of black locust on these lands.

Most forest tree species exhibit genetic variation in height and volume growth. Considerable gain has been made in recent years by identifying superior sources of many tree species. Kennedy (1983) has reported genetic variation among sources of black locust in seed length, seed length/width ratio, seedling height, rachis length, and timing of spring bud break. Provenance tests of over 100 locust seedlots have been established in West Virginia since 1984. The purposes of these tests are to determine how much genetic variability exists in black locust and which sources are the most desirable for planting on surface mined land. Recommendations from field performance results of these tests will not be available for another 3 to 9 years. Physiological traits which may reflect expected field performance can be measured immediately, however. The basic photosynthetic pigments, chlorophyll A and chlorophyll B, can easily be measured in a laboratory using spectrophotometric techniques (MacKinney 1941). These chemicals are primary components of carbon assimilation, and variation in their concentrations may indicate limits in the amount of possible growth. Concentrations of the basic nutrients N, P, Ca, Mg, and K may also limit seedling development. Studies of these traits were made to provide genetic information in advance of useful growth data. Knowledge of the genetic variation in leaf physiology will lead to a better understanding of tree growth and of ways to improve the species.

PROCEDURES

Plantation Establishment

Seed collections were made throughout the range of black locust during the years 1981 to 1984. Three experimental plantations of seedlings on surface mined lands have been established in Greenbrier County, WV (table 1).

Greenhouse-grown seedlings were produced at Westvaco's West Virginia Research Center in Rupert, WV. The nursery-grown seedlings were produced at the Parsons, West Virginia State Nursery. Each planting is composed of five or six blocks within which all seedlots are represented and randomly distributed. Measurements of height and survival are made annually. Relative heights were expressed as a percentage of the plantation mean.

Plant materials for the physiological studies were obtained from the 1984 Bellburn planting. Table 2 lists geographic

Table 1.--Location, elevation, and establishment data of experimental black locust plantations, Greenbrier County, WV.

Site	Ele.	Year Estab.	No. of Seedlots	Seedling type
Bellburn #1	823 m	1984	30	Greenhouse-grown
Clearco	1070 m	1985	26	Greenhouse-grown
Bellburn #2	823 m	1986	117	Nursery-grown, 1-0 Stock

origins for each seedlot. Prior to the 1985 growing season, after one year in the field, the seedlings ranged in height from 6 to 120 cm.

Chlorophyll Analysis

Two times during the 1985 growing season, on June 12 and August 23, leaf samples were collected on randomly selected, fully developed leaves to perform chlorophyll concentration analyses. A paper punch was used to make a 0.266 cm^2 leaf disk of each sampled leaf. Tissues were collected only on the leaflet blade, avoiding the midrib. Leaf disks were immediately stored in a glass vial which was transported to the lab inside an ice chest. All vials were stored in the freezer as soon as they were returned to Westvaco's West Virginia Research Laboratory. Chlorophyll analyses were similar to those described by Arnon (1949). Disks were homogenized in 3 ml of 80% acetone for 10 min. The mixture was transferred to graduated centrifuge tubes, diluted to 10 ml with 80% acetone, and centrifuged for 20 min at high speed. The supernatant was measured at 645 and 633 nm wavelength with a Beckman Spectronic 21 spectrophotometer using an 80% acetone blank (MacKinney 1941). Chlorophyll A (Chl. A) and chlorophyll B (Chl. B) concentrations were determined by the following equations:

$$\text{Chl. A (mg/L)} = 12.7\ D_{663} - 2.69\ D_{645}$$

$$\text{Chl. B (mg/L)} = 22.9\ D_{645} - 4.68\ D_{663}$$

$$\text{Total chlorophyll (mg/L)} = 20.2\ D_{645} + 8.02\ D_{663}$$

where D_{645} and D_{663} are the optical density readings at 645 and 663 nm, respectively. Concentrations in the extract solutions were adjusted for total area of leaf disks used in the extraction. Final leaf concentrations were given as $\mu g/cm^2$ leaf area.

Table 2.--Geographic origins of seedlots in the 1984 Bellburn #1 black locust seedlot test.

Seedlot	City	County	State	North Latitude		West Longitude		Elevation, m
683	Blacksburg	Pulaski	VA	37°	09'	80°	33'	536
687	Blacksburg	Montgomery	VA	37	17	80	27	689
688	Gay	Meriwether	GA	33	01	84	35	244
692	Gay	Meriwether	GA	32	58	84	41	259
696	Dadeville	Tallapoosa	AL	32	50	85	48	198
699	Dadeville	Tallapoosa	AL	32	50	85	48	201
700	Dadeville	Tallapoosa	AL	32	50	85	48	201
701	Grant	Marshall	AL	34	31	86	15	372
703	Gurley	Madison	AL	34	46	86	30	238
707	Hiawassee	Towns	GA	34	52	83	43	622
712	Hiawassee	Towns	GA	34	55	83	42	597
715	Lake City	Campbell	TN	36	17	84	12	372
716	Caryville	Campbell	TN	36	18	84	13	323
720	Staunton	Augusta	VA	38	05	79	05	469
722	Staunton	Augusta	VA	38	13	79	03	475
730	Umpire	Howard	AR	34	17	94	03	268
732	Umpire	Howard	AR	34	15	94	04	244
734	Licking	Texas	MO	37	34	91	53	366
742	Chestnut Mound	Smith	TN	36	13	85	50	213
743	Elmwood	Smith	TN	36	13	85	53	165
748	Robbinsville	Graham	NC	35	17	83	46	695
750	Bryson City	Swain	NC	35	25	83	28	543
754	Gneiss	Macon	NC	35	06	83	16	884
756	Gneiss	Macon	NC	35	08	83	17	645
797	Univ. Park	Centre	PA	40	48	75	55	384
799	Univ. Park	Centre	PA	40	49	77	56	421
808	Frostburg	Allegany	MD	39	39	78	59	640
811	Frostburg	Allegany	MD	39	37	78	52	488
812	Elkins	Randolph	WV	38	54	79	42	725
815	Harman	Randolph	WV	38	54	79	33	847

Nutrient Analysis

A collection of leaves was made on June 12, 1985, in order to perform a nutrient variation analysis. One fully mature leaf randomly selected from each of the five trees of a seedlot was collected within each block. If leaf samples were too small to make a minimum 1 g ovendry sample, additional leaves were collected from trees within the block. One hundred and seventy-five samples were collected, dried, and analyzed at the Westvaco Forest Science Laboratory for N, P, Ca, Mg, and K.

Simple correlations of seedlot mean values for nutrient and chlorophyll concentrations and height growth were made to determine possible patterns.

RESULTS

Seedlot Chlorophyll Variation

Chlorophyll A concentrations were consistently higher than chlorophyll B concentrations across all seedlots during both collection times. Chl. A comprised over 60 percent of total chlorophylls, and showed a slight increase in the August collection. Mean seedlot values in the June measurements ranged from 22 to 29 $\mu g/cm^2$ for chlorophyll A and from 13 to 19 $\mu g/cm^2$ for chlorophyll B (table 3). The range of August values was 21 to 35 and 13 to 21 $\mu g/cm^2$ for chlorophyll A and B, respectively. Significant seedlot differences in Chl. A concentrations existed in both the June and August measurements (table 4). Differences among seedlots for Chl. B were only observed in the August collection. The ratio between Chl. A and Chl. B did not vary between seedlots. This ratio was higher in the August collection, however, due to an increase in Chl. A levels.

For a specific collection period, the values of Chl. A, Chl. B and total chlorophylls were highly correlated. Seedlots number 699 from Alabama and 812 from West Virginia had the highest chlorophyll levels in the June collection. They still had high rankings in the August collection, but seedlot 815 from West Virginia, which had a much lower level in the June collection, had the highest August levels. Seedlot 799 decreased in total chlorophyll from the June to August collections. Analysis of variance indicated no interaction between seedlot and date of collection (table 4). The ratio between Chl. A and Chl. B showed no consistent relationship across seedlots in both collections.

There was no apparent geographic pattern to the chlorophyll measurement variation. The highest June values were in

Table 3.--Concentrations of chlorophylls and the ratio of chlorophyll A to chlorophyll B in the leaves of thirty black locust seedlots.

Seedlot	Chlorophyll A			Chlorophyll B			Total Chlorophyll			Chl. A/Chl. B Ratio		
	June	August	Avg.	June	August	Avg.	June	August	Avg.	June	August	Avg.
	- - - - - - - - - - - - - - - - $\mu g/cm^2$ - - - - - - - - - - - - - - - -											
683	25a-e*	24b-e	25b-h	15	14b	15cd	41a-d	38cde	40b-h	1.65	1.67	1.66
687	22e	23cde	22h	14	14b	14d	36d	37cde	36gh	1.55	1.61	1.58
688	23b-e	21de	22h	15	13b	14d	38bcd	34de	36h	1.55	1.59	1.57
692	22de	29a-d	26b-h	14	18ab	16bcd	36cd	47a-e	41b-h	1.59	1.68	1.63
696	27a-d	29a-e	28a-d	17	17ab	17abc	44abc	46a-e	45a-d	1.60	1.64	1.62
699	29a	33ab	31a	19	20a	19a	48a	53ab	51a	1.59	1.61	1.60
700	25a-e	29a-e	27a-g	15	18ab	16bcd	39a-d	46a-e	43b-h	1.67	1.64	1.65
701	26a-e	23cde	25b-h	16	14b	15bcd	42a-d	38cde	40b-h	1.66	1.64	1.65
703	24a-e	24b-e	24c-h	16	14b	15bcd	40a-d	39cde	39b-h	1.56	1.68	1.62
707	25a-e	22cde	24d-h	16	14b	15bcd	41a-d	36cde	39c-h	1.57	1.65	1.60
712	23b-e	22cde	23fgh	14	13b	14d	38bcd	36cde	37fgh	1.64	1.67	1.66
715	24a-e	26a-e	25b-h	15	15b	15bcd	39a-d	41b-e	40b-h	1.61	1.70	1.66
716	26a-e	22cde	24d-h	16	14b	15bcd	42a-d	36cde	39b-h	1.60	1.58	1.59
720	23b-e	25b-e	24d-h	14	15b	15cd	36cd	40b-e	38d-h	1.63	1.64	1.63
722	24a-e	27a-e	25b-h	15	16ab	15bcd	39bcd	42a-e	40b-h	1.65	1.70	1.67
730	26a-e	29a-e	27a-f	16	17ab	17bcd	41a-d	46a-e	44a-g	1.64	1.69	1.67
732	25a-e	28a-e	26b-h	16	18ab	16bcd	40a-d	45a-e	42b-h	1.57	1.57	1.57
734	23b-e	22cde	22h	14	13b	14d	37bcd	35cde	36h	1.58	1.64	1.61
742	23b-e	24b-e	23d-h	15	13b	14d	38bcd	37cde	38e-h	1.57	1.75	1.66
743	25a-e	30abc	27a-g	16	18ab	17abc	41a-d	48abc	44a-f	1.54	1.65	1.59
748	24a-e	27a-e	26b-h	16	15ab	15bcd	40a-d	42a-e	41b-h	1.57	1.71	1.65
750	26a-e	26a-e	26b-h	15	16ab	15bcd	41a-d	41b-h	41b-h	1.67	1.67	1.67
754	23b-e	22cde	23gh	14	13b	14d	37bcd	35cde	36h	1.62	1.70	1.66
756	22de	25b-e	23d-h	14	15ab	15cd	36cd	41b-e	38d-h	1.55	1.63	1.58
797	22cde	29a-d	26b-h	13	18ab	16bcd	35d	47a-e	42b-h	1.68	1.63	1.65
799	26a-e	20e	23e-h	16	13b	15cd	42a-d	33e	38e-h	1.62	1.56	1.59
808	28ab	27a-e	27a-f	16	17ab	17bcd	44a-d	44a-e	44a-g	1.69	1.59	1.64
811	28abc	31abc	29ab	16	18ab	17abc	44a-d	49abc	46abc	1.70	1.70	1.70
812	28ab	30a-d	29abc	18	18ab	18ab	46ab	48a-d	47ab	1.59	1.67	1.63
815	23b-e	35a	28a-e	15	21a	17abc	38bcd	55a	45a-e	1.57	1.70	1.62
Over-all	25	26	25	15	16	16	40	42	41	1.61	1.65	1.63

*Values within a column followed by the same letter are not significantly different at 5% by Duncan's Multiple Range Test.

seedlots from Alabama, West Virginia, and Maryland. The lowest June values were seedlots from Virginia, Georgia, North Carolina, and Pennsylvania. The August values similarly indicated no geographic patterning to the variation.

The lack of a clear-cut geographic pattern to the variation is significant because most tree species do show such variation. It is possible that natural variation has been severely disturbed by artificial regeneration techniques during the past several hundred years.

Leaf Nutrient Variation

Leaf nutrient levels also varied between seedlots. Of the five nutrients routinely analyzed, only calcium showed no seedlot source differences (table 5). Significant block differences for N and Ca levels may have been due to the artificial mixing of soils associated with strip mining.

Correlation analysis of individual samples indicates that nitrogen levels were proportional to phosphorus, calcium, and potassium levels. Phosphorus levels also correlated positively with magnesium and potassium. Although seedlot differences were significant for four nutrients, the ranges of values were not great (table 6). Seedlot 688 from Meriwether County, GA, had the highest values for most nutrients. Seedlot 815 from Randolph County, WV, had the lowest nitrogen levels. As with the chlorophyll variation, no geographic patterns could be determined among the seedlots.

Height Growth Variation

The most recent data are presented in table 7 for 56 seedlots. Statistically distinct differences in growth can be seen in each study. At the Bellburn #1 location, seedlots from Tennessee, Alabama, and West Virginia had the best growth by age 3 yrs. In the Clearco study, lot BN4194,

Table 4.--Analysis of variance of black locust leaf chlorophyll concentrations.

Source	DF	Chlorophyll A Type III Sums of Squares	Chlorophyll A F	Chlorophyll B Type III Sums of Squares	Chlorophyll B F	Chl. A/Chl. B Type III Sums of Squares	Chl. A/Chl. B F
Spring Collection							
Seedlot	29	682.45	1.70 *	232.38	1.34 NS	0.3450	0.98 NS
Block	5	200.32	2.90 *	84.13	2.81 *	0.1608	2.65 *
Error	136	1879.02	-	814.81	-	1.6482	-
Fall Collection							
Seedlot	29	2015.60	1.85 *	725.91	2.21 **	0.3307	1.05 NS
Block	5	413.69	2.20 NS	80.68	1.37 NS	0.2822	5.21 **
Error	127	4782.61	-	1498.85	-	1.3757	-
Combined Measurements							
Seedlot	29	1811.19	2.13 **	671.50	2.32 **	0.4135	1.16 NS
Block	5	138.39	1.17 NS	26.73	0.65 NS	0.1079	1.70 NS
Error A	140	4109.91	1.25 NS	1398.32	1.21 NS	1.7195	0.97 NS
Collection	1	115.16	4.88 *	6.76	0.82 NS	0.1132	8.94 **
S X C	29	941.14	1.38 NS	333.38	1.39 NS	0.3194	0.87 NS
Error B	128	3017.97	-	1057.76	-	1.6212	-

*, ** - Differences between means are significant at 5% and 1%, respectively.
NS - No significant differences between means.

Table 5.--Statistical analysis of variance of black locust seedlot leaf nutrient levels.

Degrees of Freedom	Source: Seedlot 29	Block 5	Error 140
Nitrogen			
Type III Sum of Squares	5.64	1.23	14.95
F	1.82*	2.32*	-
Phosphorus			
Type III Sum of Squares	0.06	0.01	0.12
F	2.23**	1.29NS	-
Calcium			
Type III Sum of Squares	0.43	0.28	1.68
F	1.24NS	4.58**	-
Magnesium			
Type III Sum of Squares	0.13	0.01	0.26
F	2.39**	1.56NS	-
Potassium			
Type III Sum of Squares	2.15	0.06	4.11
F	2.53**	0.43NS	-

* - Differences significant at 5%.
** - Differences significant at 1%.
NS - Differences not significant.

which was provided by the Soil Conservation Service, but originated in West Virginia, performed excellently. The Bellburn #2 study showed additional good material from Virginia. Seedlot # 716 from Tennessee performed well in both of the Bellburn plantings. Individual tree selection is necessary for an efficient genetic improvement program.

Although seedlot differences exist, we have not observed any geographic pattern from which we can make collection recommendations. This observation is consistent with those made by other researchers who have concluded that little geographic variation can be associated with genetic variation in black locust (Bongarten 1985; Kennedy 1983). Individual tree selection is necessary for an efficient genetic improvement program.

Relationship Between Traits

Mean seedlot values for each trait measured in this report were compared to determine possible relationships which may exist. Although considerable variation occurred in the data, simple linear regressions did point out some expected and unexpected relationships.

Seedlots with higher levels of nitrogen tended to have higher phosphorus ($r = 0.54$) and potassium levels ($r = 0.37$). Phosphorus levels also paralleled magnesium levels. The results are similar to those observed with individual sample correlations. An unexpected result was the negative relationship between certain nutrients and chlorophyll concentrations and height

Table 6.--Leaf nutrient variation among thirty seedlots of black locust seedlings.

Seedlot	Nitrogen	Phosphorus	Calcium	Magnesium	Potassium
			-% Tissue-		
683	3.15 a-e	0.21 a-e	0.58	0.24 cde	1.62 b-e
687	2.80 de	0.19 cde	0.51	0.22 cde	1.52 cde
688	3.48 a	0.25 a	0.62	0.30 ab	1.96 a
692	3.04 a-e	0.19 cde	0.46	0.21 e	1.80 ab
696	3.26 a-d	0.19 cde	0.50	0.19 e	1.57 b-e
699	3.36 ab	0.20 cde	0.53	0.20 e	1.67 bcd
700	2.99 b-e	0.18 de	0.58	0.21 de	1.59 b-e
701	3.07 a-e	0.18 de	0.49	0.21 e	1.64 bcd
703	3.03 a-e	0.19 cde	0.53	0.24 a-e	1.62 b-e
707	3.24 a-d	0.21 a-e	0.57	0.19 e	1.64 bcd
712	3.06 a-e	0.20 b-e	0.46	0.20 e	1.57 b-e
715	3.15 a-e	0.19 de	0.59	0.22 cde	1.76 abc
716	3.07 a-e	0.17 e	0.51	0.20 e	1.68 bcd
720	3.14 a-e	0.19 de	0.48	0.21 e	1.67 bcd
722	3.36 ab	0.22 a-d	0.53	0.27 a-d	1.50 de
730	3.01 a-e	0.21 a-e	0.51	0.20 e	1.72 bcd
732	3.19 a-e	0.21 a-e	0.50	0.19 e	1.56 b-e
734	3.25 a-d	0.24 ab	0.55	0.30 a	1.51 cde
742	3.19 a-e	0.18 de	0.58	0.25 a-e	1.65 bcd
743	3.34 ab	0.20 cde	0.59	0.22 de	1.63 bcd
748	2.82 cde	0.20 cde	0.59	0.22 de	1.43 de
750	3.16 a-e	0.22 a-d	0.48	0.24 a-e	1.53 cde
754	3.12 a-e	0.23 abc	0.53	0.28 abc	1.64 bcd
756	3.38 ab	0.23 abc	0.50	0.22 cde	1.62 bcd
797	3.28 abc	0.21 a-e	0.57	0.24 b-e	1.69 bcd
799	2.99 b-e	0.19 de	0.59	0.21 de	1.38 e
808	3.20 a-d	0.21 a-e	0.63	0.23 cde	1.55 cde
811	3.07 a-e	0.21 a-e	0.60	0.22 de	1.73 bcd
812	2.86 cde	0.19 cde	0.56	0.22 cde	1.64 bcd
815	2.72 e	0.18 de	0.62	0.24 a-e	1.49 de
Mean	3.12	0.20	0.54	0.22	1.62

* Values within a column followed by a common letter are not significantly different at 5% by Duncan's Multiple Range Test.

growth. Seedlot values for magnesium tended to decrease with increasing chlorophyll levels. Height increments were negatively related to phosphorus (r = -.60) and magnesium levels (r = -.68).

The various chlorophyll measurements tended to show positive relationships between seedlots, as could be expected. They also exhibited a positive relationship with height growth. The June measurements were more strongly related to overall seasonal growth than August levels. This relationship is supported by the variation in June foliage coloration. Seedlots which had the highest total chlorophyll in June generally had greater growth rates during the season (fig. 1). Some seedlots may produce chlorophyll earlier in the season which may allow some growth advantage. This possible relationship between June chlorophyll levels and overall seasonal growth should be verified as it may be a useful selection tool.

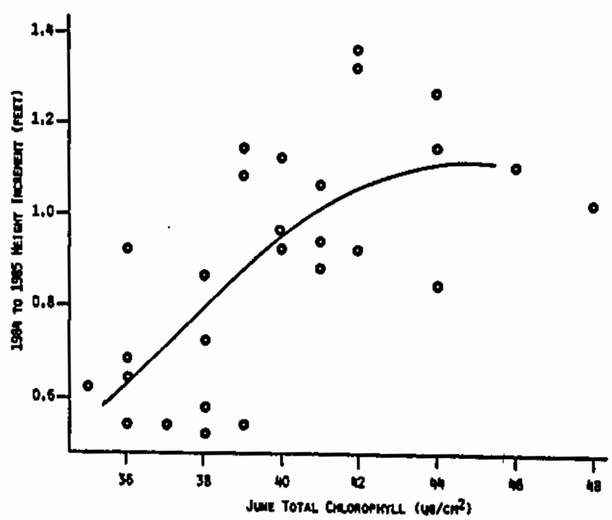

Figure 1.--Distribution of seedlot mean values for total June chlorophyll and height increment during the 1985 Growing season. The curvilinear regression accounts for 50% of the variation.

Table 7.--Relative heights of 56 black locust seedlots planted on 3 sites in West Virginia. Seedlots within a planting site which are followed by a common letter are not significantly different at a 95% probability.

Seedlot	Bellburn #1 3 yr old	Clearco 2 yr old	Bellburn #2* 1 yr old
	-Relative Heights and Differences- Percent of Plantation Mean		
683	89 A-E	--- ---	106 B-Q
687	81 CDE	--- ---	130 A-D
688	92 A-E	--- ---	94 E-X
692	83 B-E	--- ---	109 A-O
696	115 A-D	--- ---	87 J-X
699	106 A-E	--- ---	--- ---
700	113 A-D	--- ---	100 C-S
701	121 AB	--- ---	71 R-Y
703	103 A-E	--- ---	124 A-F
707	99 A-E	--- ---	100 C-S
712	103 A-E	--- ---	106 B-Q
715	105 A-E	--- ---	103 B-S
716	125 A	--- ---	134 AB
720	79 DE	--- ---	95 E-W
722	79 DE	--- ---	92 E-X
730	105 A-E	--- ---	111 A-N
732	95 A-E	--- ---	84 K-X
734	71 E	--- ---	--- ---
742	109 A-D	--- ---	--- ---
743	103 A-E	--- ---	112 A-N
748	105 A-E	--- ---	63 T-Y
750	85 B-E	--- ---	93 E-X
754	83 CDE	--- ---	90 F-X
756	92 A-E	--- ---	98 D-S
797	100 A-E	--- ---	105 B-R
799	95 A-E	--- ---	125 A-D
808	95 A-E	--- ---	111 A-N
811	113 A-D	--- ---	107 B-Q
812	117 ABC	--- ---	106 B-Q
815	100 A-E	--- ---	102 B-S
838	--- ---	93 CDE	60 XY
839	--- ---	112 BCD	112 A-M
840	--- ---	80 DEF	85 K-X
841	--- ---	92 C-F	89 G-X
842	--- ---	90 C-F	73 Q-Y
845	--- ---	98 CDE	114 A-L
846	--- ---	98 CDE	84 K-X
847	--- ---	122 BC	93 E-X
848	--- ---	79 DEF	113 A-L
849	--- ---	101 CDE	95 E-W
852	--- ---	56 F	--- ---
856	--- ---	71 EF	--- ---
857	--- ---	85 C-F	--- ---
858	--- ---	115 BCD	--- ---
860	--- ---	83 DEF	120 A-I
861	--- ---	111 BCD	119 A-I
862	--- ---	72 EF	96 D-U
863	--- ---	83 DEF	88 H-X
865	--- ---	93 CDE	97 D-T
866	--- ---	81 DEF	62 WXY
867	--- ---	96 CDE	100 C-S
868	--- ---	92 C-F	78 N-Y
869	--- ---	85 C-F	108 A-O
4191	--- ---	138 AB	--- ---
4192	--- ---	101 CDE	--- ---
4194	--- ---	157 A	--- ---

*Only those seedlots common to the other plantings are listed.

CONCLUSIONS

This research in black locust seedlot variations has led to the following conclusions:

1. The concentrations of chlorophylls A and B in the leaves of black locust seedlings vary seasonally, increasing from low June levels to the highest levels in August.

2. Concentrations of chlorophylls in black locust exhibit considerable seedlot variation. The variation has no geographic patterning associated with it. Chlorophyll A had a consistently higher concentration than chlorophyll B and increased in concentration during the season.

3. There are no significant interactions between seedlot chlorophyll levels and time of collection.

4. The ratio between chlorophyll A and chlorophyll B did not vary between seedlots. It did increase during the growing season as chlorophyll A levels increased and chlorophyll B did not.

5. Leaf nutrient levels of N, P, Mg, and K did vary significantly between seedlots. No geographic patterning was associated with this variation.

6. Simple correlation analysis of the various traits measured indicates a positive relationship between June chlorophyll levels and overall seasonal growth. Seedlot variation in the rates of chlorophyll development may yield a growth advantage and possibly could be used as a selection criterion in black locust improvement.

7. Seedlots do vary significantly in their rate of growth. Several seedlots have been identified which are superior to others when planted on surface mined lands. As the technology to reproduce superior black locust genotypes develops, owners of reclaimed surface mines may be able to select and plant only the best material for their sites.

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