

# CLONAL TESTS OF NEW COTTONWOOD SELECTIONS FROM THE SOUTHEAST

Jonathan Paul Jeffreys, Samuel B. Land, Jr., Emily B. Schultz, and Andrew J. Londo<sup>1</sup>

**Abstract**—One hundred “new” clones and 20 “check” clones were established with unrooted cuttings during March–April 2003 in a second-stage clonal trial in Missouri and Georgia. The new clones had been selected for 2-year superiority in *Melampsora* leaf rust resistance, height growth, and diameter growth during first-stage rooted cutting trials. All 120 clones were vegetatively multiplied in a cutting production nursery at Stoneville, MS, and cuttings from this nursery were used for the two second-stage trials. Results from second-year measurements of *Melampsora* leaf rust infection, height, and d.b.h. indicate that geographic patterns of genetic variation were apparent at both sites. Location effects were present for all traits except d.b.h. for the 120 clones tested, and additional gains from selection among clones within origins can be attained for these traits.

## INTRODUCTION

Eastern cottonwood (*Populus deltoides* Bartr. ex Marsh. var. *deltoides*) is the fastest growing native commercial forest species in North America (Cooper and van Haverbeke 1990). This rapid growth has led to the establishment of poplar plantations worldwide. Success of these plantations begins with the selection of the correct seed sources and/or clones. However decisions on which seed source or clone to use should not be made until thorough testing has been conducted (Zobel and Talbert 1984). Planting trees from the wrong seed source or clone can be financially costly and time-consuming due to delayed mortality or poor growth.

Genetic improvement of eastern cottonwood has been undertaken in several regions of the United States (Mohn 1973, Nelson and Tauer 1987, Wilcox and Farmer 1967, Ying and Bagley 1976). The U.S. Department of Energy has funded research on the development of high-yielding, short-rotation woody crops of *Populus* for energy and fiber production (Wright and Tuskan 1997). However, relatively few improved clones have been released for use in the Southeastern United States. The purpose of the present study is to select superior performing clones based on performance at a fiber-farm location in southeast Missouri and at a location in east Georgia.

## MATERIALS AND METHODS

Land and others (2001) divided the southeast region east of the Mississippi River into three subregions: Southeast Atlantic (SA), East Gulf (EG), and East Central (EC) (fig. 1). These were used in sampling the region's wild population of eastern cottonwood. Open-pollinated seeds were collected from mother trees in natural stands on various rivers within each subregion. The seeds were germinated and vegetatively multiplied as containerized rooted cuttings for four first-stage field trials (Warwell and others 1999). These field sites were located in Florida (30° 32.5' N, 84° 35' W), Alabama (32° 02' N, 88° 07' W), North Carolina (35° 58' N, 77° 09' W), and Missouri (32° 02' N, 89° 46' E). One hundred clones were selected for *Melampsora* leaf rust resistance and height and diameter growth based on second-year performance. An

additional 20 “check” clones from former trials of the USDA Forest Service and Oklahoma State University were chosen to include with these 100 “new” clones in second-stage field trials.

The 120 clones were vegetatively multiplied during 2002, in a cutting production nursery at Mississippi State University's (MSU) Delta Research and Extension Center in Stoneville, MS. On February 10 and 11, 2003, cuttings were collected from the 120 clones. These 12-inch unrooted hardwood cuttings were planted in second-stage clonal trials in Scott County, MO (MO), and Richmond County, GA (GA).

The MO location was planted on April 8, 2003, while the GA location was planted on March 12–13, 2003. The clones were planted in a randomized complete block design. Each location was divided into four replications and planted with two trees per clone per replication. Two cuttings of each clone were planted at each position to help insure survival. Border rows were planted at the same time around the studies at each location.

Measurements of *Melampsora* leaf rust infection were taken in both September (Y2rust1) and October (Y2rust2) of 2004. Severity of rust infection was scored according to the amount of urediospores (orange powder) visible on the leaf and the amount of leaf curl present. Scores ranged from 1 to 4, with 1 representing a clone with no visible infection and 4 representing a tree that was heavily infected and almost completely defoliated. Therefore clones with lower mean rust scores are preferred. Also, measurements were taken in October 2004 for height (Y2ht) and d.b.h. (Y2dbh). Clones that had not reached a height of 4.5 feet were assigned a d.b.h. of 0.1 inch. A performance level was calculated for each trait of each clone by subtracting the clone mean for the trait from the location mean of that trait and dividing by the standard deviation. Performance levels for d.b.h., height, and *Melampsora* leaf rust were combined to obtain the overall performance level for a clone. The overall clone performance levels were used to identify the best-performing clones for each location.

Analyses of variance for a random model were conducted for the randomized complete block design at each location. The

<sup>1</sup> Graduate Student and Professors, respectively, Mississippi State University, Department of Forestry, Mississippi State, MS 39762.

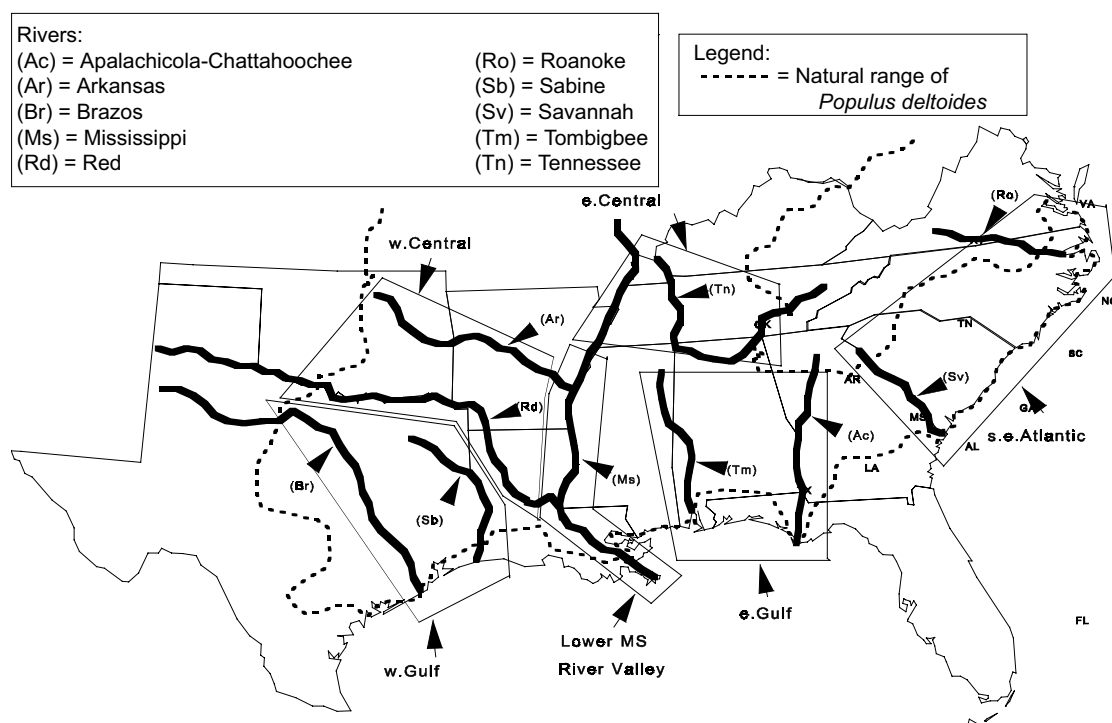


Figure 1—Map of subregions and some river systems from which open pollinated seed and cuttings were collected in 1995-1998. One-hundred “new” clones in this study came from the three eastern subregions: E. Gulf, E. Central, and S.E. Atlantic.

Tukey-Kramer test of ranked means was used to test differences among locations and among selection types (checks and new clones) for each trait.

## RESULTS AND DISCUSSION

### Location Effects

Locations differed significantly for all traits except d.b.h. in the analyses of variance, but even d.b.h. was significantly different if locations were considered fixed effects in the

Tukey-Kramer test (tables 1 and 2). Clones were taller, had slightly larger d.b.h., and had less leaf rust at the GA location. The larger amount of rust at the MO location could be due to adjacent cottonwood plantings that are present on the fiber farm. The MO location also had drip irrigation. This increase in moisture and humidity may provide a better environment for rust infection. A large incidence of *Melampsora* allows better detection of differences among clones in resistance, so the MO location was particularly helpful for this purpose.

Table 1—Analysis of variance<sup>a</sup> for test locations, replications within each location, clones, within clone groups, and locations by clone groups for *Melampsora* leaf rust infections, d.b.h. growth and height growth

Source of variation	DF	Measurement traits							
		Y2Rust1		Y2Rust2		Y2dbh		Y2Ht	
		MS	F-test <sup>b c</sup>	MS	F-test <sup>b c</sup>	MS	F-test <sup>b c</sup>	MS	F-test <sup>b c</sup>
Locations (L)	1	70.4	49.0**	106	31.9**	45.8	2.39 <sup>ns</sup>	8710	11.9*
Reps in locations	6	0.68	5.86**	2.17	13.8**	18.1	24.4**	718	91.1**
Clones	119	1.18	1.55*	2.34	1.99**	2.07	2.11**	47.3	4.09**
-new vs. check	1	3.42	29.7**	12.6	78.7**	0.09	0.12 <sup>ns</sup>	97.5	12.4**
-w/in new	99	1.03	9.00**	1.91	11.9**	1.98	2.68**	42.8	5.43**
-w/in checks	19	1.90	16.5**	4.07	25.4**	2.62	3.54**	68.2	8.64**
L x clones	119	0.76	6.61**	1.17	7.47**	0.98	1.32**	11.6	1.47**
-L x (new vs. checks)	1	7.45	64.8**	14.4	89.9**	1.35	1.82 <sup>ns</sup>	3.71	0.47 <sup>ns</sup>
-L x w/in new	99	0.67	5.80**	1.09	6.81**	1.08	1.46**	11.2	1.41**
-L x w/in checks	19	0.83	7.20**	0.85	5.31**	0.39	0.53 <sup>ns</sup>	14.3	1.81*
Error	1428	0.115		0.157		0.743		7.894	

<sup>a</sup> Model is completely random

<sup>b</sup> ns = non-significant at the 0.05 level

<sup>c</sup> \* = significant at the 0.05 level; \*\* = significant at the 0.01 level

**Table 2—Location means and results<sup>a</sup> of Tukey-Kramer test of ranked means for d.b.h. growth, height growth, September rust score, and October rust score for 100 “new” clones, 20 “check” clones, and location means for all 120 “new and check” clones combined**

Trait	Type of clones	Test location		Clone type over both locs.
		GA	MO	
D.b.h. (in.)	Checks	2.38b	1.92a	2.16S
	New	2.28b	1.99a	2.14S
	Loc. means	2.30B	1.98A	2.15
Height (ft.)	Checks	20.4e	15.9d	18.2U
	New	19.8e	15.1c	17.6T
	Loc. means	19.9D	15.3C	17.7
Rust (Sept.)	Checks	1.80f	2.51h	2.14W
	New	1.88f	2.23g	2.05V
	Loc. means	1.87E	2.28F	2.06
Rust (Oct.)	Checks	2.21i	3.12k	2.64Y
	New	2.23i	2.67j	2.44X
	Loc. means	2.23G	2.75H	2.47

<sup>a</sup> Means followed by the same letter and case for each trait are not significantly different (5% probability level)

### Checks vs. New Clones and Interactions with Locations

Selection types (checks vs. new clones) were not significantly different for d.b.h., but height was greater and rust infection was greater for the checks than new clones in the combined analyses over both locations (tables 1 and 2). Also, there were significant location-by-selection type interactions. The check clones had greater performance levels than the new select clones at the GA location, probably because of the greater d.b.h. and height growth by the checks (table 2). However, at the MO location, the new select clones had better performance levels than the check clones. This discrepancy was probably due to greater rust resistance by the new clones than the check clones at the MO location.

### Variation Among Check Clones

Significant variation existed within the check clones for all traits over both locations and for each individual location (table 1). The best five check clones for each location are listed in table 3 with the subregion, area (upland or bottom-

land), river, state of origin, and performance score. There were three check clones (ST111733, ST111234, and S7C8) that were represented in the top five checks at both locations. The top two “check” clones (ST111733 and S7C8) were the same rank at both locations. The geographic sources are different for these top “check” clones. ST111733 is from the Mississippi River in Mississippi, while S7C8 is from the Brazos River in the West Gulf subregion of Texas. The lack of change in rank indicates that clone-by-location effects may not be important for the best performers.

When the performance levels for the individual traits were compared, ST111733 performed better at the GA location due to high rust resistance and superior height growth. For the MO location, ST111733 was the top performer due to superior height growth, (which was higher than the height growth score for the GA location). However, performance of this clone for d.b.h. growth and rust resistance was only average. The second best performing “check” clone for each location was S7C8. This clone was second at the GA location

**Table 3—Performance levels and origins for the five best performing “check” clones at each location**

Location	Clone ID	Subr.	Area	River	State	Performance
GA	ST111733	LM	B	Mississippi	MS	4.1
	S7C8	WG	B	Brazos	TX	3.0
	ST111234	LM	B	Mississippi	MS	2.9
	ST904401	LM	B	Mississippi	MS	1.8
	ST111412	LM	B	Mississippi	MS	1.4
MO	ST111733	LM	B	Mississippi	MS	2.7
	S7C8	WG	B	Brazos	TX	2.4
	ST-148	LM	B	Mississippi	MS	2.2
	S7C1	WG	B	Brazos	TX	1.9
	ST111234	LM	B	Mississippi	MS	0.5

due to superior d.b.h. growth. Its height growth performance was somewhat above average, but it had only average rust resistance. For the MO location, S7C8 was second due to superior height growth, (which was better than the GA location) and average d.b.h. growth. S7C8 had a poor rust resistance score (0.02) at the MO location, but the height performance score is what pushed it ahead of the other “check” clones.

### Variation Among New Clones

Significant variation existed among the “new” 100 select clones for all traits in the combined analyses over locations (table 1). However, there were also significant locations-by-clones-within-new-selections interactions. The top 10 clones for each location are listed in table 4 along with the subregion, area, river, state of origin, and performance scores. There were 3 clones (MS072C-7, MS093-1, and MS094-1) that were in the top 10 at both sites. These three clones were from the Tombigbee River system in Alabama and the Apalachicola River in Florida, and they were all from bottomland areas.

The best 3 clones for the GA location were not in the top 10 clones at the MO location, and vice versa. This indicates that some clones show greater genetic x environment interaction than others. This also indicates that the interaction between locations and new clones may be more important than that for checks.

The best “new” clone for the GA location was MS093-6, which is from a bottomland area in the Tombigbee River system in Alabama (East Gulf subregion). This clone was superior because of a high d.b.h. growth performance level. None of the remaining clones in the top 10 at this location were within 1.5 standard deviations of MS093-6’s d.b.h.

performance level. The 3 clones that were in the top 10 at both locations were ranked differently at each location. At the GA location, MS094-1 was ranked highest of the three. This ranking was due to the clone’s rust resistance. MS093-1 was second of the three due to slightly above average scores for all three traits. MS072C-7 was third at this location due to slightly above average rust resistance and d.b.h. growth. However, this clone had a below-average performance level for height growth.

The best “new” clone for the MO location was MS095A-6, which is from a bottomland area in the Escambia River system in Florida (East Gulf subregion). This clone’s superiority was due to a high height growth performance level, high rust resistance, and a better than average d.b.h. growth performance. MS093-1 was the top performer of the 3 clones that were in the top 10 at both locations. This was due to good height growth and slightly above average d.b.h. growth and rust resistance. MS072C-7 came in second of the three due to a high rust score and average d.b.h. and height growth. MS094-1, which was the best of the three at the GA location, was the worst of the three at the MO location. However, at this location, it still had good rust resistance and height growth, but the d.b.h. growth performance score was below average.

With the exception of one unknown clone, the best clones for the MO location consisted of trees from the Tombigbee River in southwest Alabama and the Escambia and Apalachicola River systems in northwest Florida. The Tombigbee River system in both Alabama and Mississippi was also the primary source for the GA location’s best clones. For the 2 locations combined, >50 percent of the top performing clones came from the Tombigbee River system.

**Table 4—Performance levels and origins for the 10 best performing “new” clones at each location**

Location	Clone ID	Subr.	Area	River	State	Performance
GA	MS093-6	EG	B	Tombigbee	AL	3.2
	MS096-2	EG	B	Tombigbee	MS	2.6
	MS086-6	EG	U	Chattahoochee	AL	2.4
	MS094-1	EG	B	Tombigbee	AL	2.3
	MS093-1	EG	B	Tombigbee	AL	2.1
	MS154A-1	SA	U	Saluda	SC	2.0
	MS118-4	EG	U	Tombigbee	AL	2.0
	MS130B-1	SA	B	Pee Dee	SC	1.2
	MS118-3	EG	U	Tombigbee	AL	1.2
	MS072C-7	EG	B	Apalachicola	FL	1.1
MO	MS095A-6	EG	B	Escambia	FL	4.0
	MS094-4	EG	B	Tombigbee	AL	3.1
	MS105-5	EG	B	Tombigbee	AL	2.5
	MS119-1	EG	U	Tombigbee	AL	2.3
	MS092-5	EG	B	Tombigbee	AL	2.2
	MS093-1	EG	B	Tombigbee	AL	2.2
	MS006-4			Unknown		2.2
	MS072C-7	EG	B	Apalachicola	FL	2.0
	MS094-1	EG	B	Tombigbee	AL	2.0
	MS093-5	EG	B	Tombigbee	AL	1.9

## Comparison of Best Clones from Checks and New Clones Combined

The best performing check clone (ST111733) for the GA location performed better than the top performing "new" clone (MS093-6) for that location. However, S7C8 did not perform better than the best "new" clone. The 2 superior check clones did, however, perform better than the 3 new clones that occurred in the top 10 at both locations. The two best "new" clones (MS095A-6 and MS094-4) at the MO location performed better than the best two check clones. However, the top 2 check clones performed better than the 3 new clones that occurred in the top 10 at both locations.

The 5 clones of choice for the entire southeast would be the 3 new clones that occurred in the top 10 at both locations and the 2 best check clones. The 3 "new" clones, while not the best at each location, have shown the ability to perform better across the southeast than 90 percent of the 100 select clones. The clones of choice for the MO location, and other locations similar to that location, would be the two top check clones along with the top three "new" clones (MS095A-6, MS094-4, and MS105-5) at that location. The clones of choice for the GA location, and locations similar to it, would be the top two check clones that occurred at both locations, the third best performing check clone (ST111234) at GA, and the top two "new" clones (MS093-6 and MS096-2) at that location.

## SUMMARY AND CONCLUSIONS

There were significant differences between the GA and MO locations for all traits except d.b.h., when locations were considered random. Clones were taller, had slightly larger d.b.h.s, and had less rust infection at the GA location. Mean performance of "check" clones was better than mean performance of "new" clones at the GA location, while the "new" clones had better performance levels than the "checks" at the MO location. There were three "check" clones that were in the top five "checks" at the two locations. However, relative contributions of rust resistance, d.b.h., and height to the high overall performance level of these three check clones differed at the two locations. There were three "new" clones that performed well at both locations. These were in the top 10 new clones at each location, but they were not among the top 3 ranking new clones at either location. The top 3 "new" clones in rank for the GA location were not in the top 10 "new" clones at the MO location, and likewise the top 3 "new" clones at the MO location were not in the top 10 "new" clones at the GA location. The clones of choice for the GA location

are the three "check" clones (ST111733, ST111234, and S7C8) and the top two "new" clones (MS093-6 and MS096-2). The ideal clones for the MO location would be the top two "check" clones (ST111733 and S7C8) and the top three "new" clones (MS095A-6, MS094-4, and MS105-5). All of these new-clone selections come from the Tombigbee, Escambia, and Apalachicola Rivers in south Alabama and northwest Florida.

## ACKNOWLEDGMENTS

This manuscript is publications #FO-280 of the Forest and Wildlife Research Center, Mississippi State University.

## LITERATURE CITED

- Cooper, D.T.; van Haverbeke, D. 1990. *Populus deltoides* Bartr. ex Marsh Eastern Cottonwood. In: Burns, R.M.; Honkala, B.H., eds. *Silvics of North America, volume 2. Hardwoods. Agric. Handb. 654.* Washington, DC: U.S. Department of Agriculture, Forest Service: 530-543.
- Land, S.B., Jr.; Stine, M.; Ma, X. [and others]. 2001. A tree improvement program for eastern cottonwood in the Southeastern United States. Proceedings of the 26<sup>th</sup> biennial southern forest tree improvement conference. Athens, GA: The University of Georgia: 84-93.
- Mohn, C.A. 1973. Practical breeding of cottonwood in the northcentral region. In: Joint Proceedings of the 10<sup>th</sup> Lake States forest tree improvement conference and the 7<sup>th</sup> Central States forest tree improvement conference. Gen. Tech. Rep. NC-3. St. Paul, MN: U.S. Department of Agriculture, Forest Service, North Central Forest Experiment Station: 35-39.
- Nelson, C.D.; Tauer, C.G. 1987. Genetic variation in juvenile characters of *Populus deltoides* Bartr. from the southern great plains. *Silvae Genetica*. 36(5-6): 216-221.
- Warwell, M.V.; Alker, G.R.; Rockwood, D.L [and others]. 1999. Culture and genetic influences on greenwood cutting propagation of a new collection of eastern cottonwood in the South. In: Proceedings of the 25<sup>th</sup> biennial southern forest tree improvement conference. New Orleans, LA: Louisiana State University: 174-176.
- Wilcox, J.R.; Farmer, R.E., Jr. 1967. Variation and inheritance of juvenile characters of eastern cottonwood. *Silvae Genetica*. 16: 162-165.
- Wright, L.L.; Tuskan, G.A. 1997. Strategy, results and directions for woody crop research funded by the US Department of Energy. In: Proceedings of the 1997 pulping conference. Atlanta, GA: TAPPI Press: 791-799.
- Ying, C.C.; Bagley, W.T. 1976. Genetic variation of eastern cottonwood in an eastern Nebraska provenance study. *Silvae Genetica*. 25: 67-73.
- Zobel, B.J.; Talbert, J.T. 1984. The importance of source of seed in tree improvement programs. In: Applied forest tree improvement. New York: John Wiley and Sons, Inc.: 76-116.