Multiple factors affect pest and pathogen damage on 31 *Populus* clones in South Carolina ☆

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Abstract

Populus species and hybrids have many practical applications, but there is a paucity of data regarding selections that perform well in the southeastern US. We compared pest susceptibility of 31 *Populus* clones over 3 years in South Carolina, USA. Cuttings were planted in spring 2001 on two study sites. Clones planted in the bottomland site received granular fertilizer yearly and irrigation the first two years only, while those on the sandy, upland site received irrigation and fertilization throughout each growing season. Foliar damage by the cottonwood leaf beetle (*Chrysomela scripta*), cottonwood leafcurl mite (*Tetra lobulifera*), and poplar leaf rust (*Melampsora medusae*) was visually monitored several times each growing season. Damage ratings differed significantly among clones, and clonal rankings changed from year to year. Irrigation increased *C. scripta* and *M. medusae* damage, but had no effect on *T. lobulifera* damage. Certain clones received greater pest damage at a particular study site. Temporal damage patterns were evident among individual clones and on each site. At the upland site, OP367 and 7300502 were highly resistant to all three pests; I45/51 was highly resistant to *C. scripta* and *M. medusae*; NM6 and 15–29 were highly resistant to *M. medusae*; and 7302801 was highly resistant to *T. lobulifera* and *M. medusae*. At the bottomland site, NM6, Eridano, I45/51, and 7302801 were highly resistant to all three pests; clone 7300502 was highly resistant to *M. medusae* only. Based on this preliminary 3-year study of pest damage levels, we would recommend clones NM6, Eridano, I45/51, OP367, 15–29, 7302801, 7300502, and Kentucky 8 for use in this region.

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Keywords: *Chrysomela scripta*; Clone; Irrigation; *Melampsora medusae*; Temporal variation; *Tetra lobulifera*

1. Introduction

With production rates far greater than could be obtained naturally, wood and wood products from intensively managed forest plantations may reduce harvest pressure

on native forests [1,2]. Additionally, species such as *Populus* [3] and *Salix* [4] can be used for phytoremediation, carbon sequestration, erosion control, and biomass production.

Genotypes of *Populus* are desirable for intensive management given their rapid growth characteristics and established propagation methodologies [5]. A major focus of *Populus* breeding programs is on producing high-yielding genotypes [6]. If not careful in selection schemes there is a potential loss of pest resistance or tolerance if the breeding strategy focuses too strictly on growth alone [7,8]. Growth is the most common variable used in the selection process, but mortality [9], rootability [10], and pest susceptibility [11–13] should also be included among the selection criteria.

The monocultural environment created by intensive management creates an ideal situation for pest infestations

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[14,15]. *Populus* selections are particularly susceptible to a suite of insect and disease pests [16–19]. Three pests are of major importance in the southeastern US: the cottonwood leaf beetle, *Chrysomela scripta* F. (Coleoptera: Chrysomelidae), the cottonwood leafcurl mite, *Tetra lobulifera* (Keifer) (Acari: Eriophyidae), and leaf rust, *Melampsora medusae* Thuem. (Basidiomycetes: Uredinales).

C. scripta is the most economically important pest of intensively managed *Populus* in the eastern US [19]. This multivoltine folivore can cause extensive leaf loss [20], and *C. scripta* feeding has been shown to reduce stem volume by over 70% in plantation-grown *Populus* [21]. *T. lobulifera* is cosmopolitan over much of the US [22] and can be a serious pest in the Southeast [23]. Certain *Populus* clones have been removed from commercial production due to high susceptibility to *T. lobulifera* (RJ Rousseau, pers. commun.). *Melampsora* leaf rust is a potentially lethal disease to young *Populus* [24–26]. Its alternate host lifestyle and lack of viable control tactic has made breeding rust resistance into new *Populus* selections a priority [16,25].

Several *Populus* selection trials have taken place in the Mississippi Delta region [27,28], but virtually no published information exists regarding *Populus* pest susceptibility in the southeastern US coastal plain. Our objectives were to evaluate pest resistance among a range of top performing *Populus* clones in the southeastern US. We visually measured leaf damage on several selections from the southeastern, northwestern, and north-central US on two contrasting study sites in South Carolina. We hypothesized that (1) clones would differ in their susceptibility to pest damage, (2) increased pest damage would occur on trees receiving irrigation, (3) pest damage would differ between study sites, and (4) pest damage would exhibit significant temporal variation over the course of a growing season.

2. Materials and methods

2.1. Study location

The trial was conducted on two sites (termed D-Area and SRWC) located on the US Department of Energy Savannah River Site, a National Environmental Research Park, near Aiken, SC, USA (33°23' N, 81°40' E). The climate is humid continental, with warm, dry summers and mild winters; temperature varied little between the sites and years, but mean annual rainfall was slightly greater at D-Area. Additional site details can be found in WSRC [29], Coleman et al. [30], and Coyle et al. [31].

2.2. Plant material

A total of 18 and 31 pure species and hybrid *Populus* clones were used at D-Area and SRWC, respectively [31]. Clones chosen represented a range of growing regions and genotypes, with particular emphasis on those reported or expected to do well in the southeastern US. Dormant

hardwood cuttings were stored at 3 °C and soaked in water 48 h prior to planting to promote optimum rooting [32].

2.3. Study design and treatments

Specific details are available in Coyle et al. [31]; a brief description follows. Both sites were planted in spring 2001; D-Area with four blocks of 18 clones at 1.3 × 1.3 m spacing, and SRWC with two blocks of 31 clones at 2.5 × 2.5 m spacing. D-Area clones S7C15, 52–225, and Eridano were planted in 36-tree plots; all others were planted in 16-tree plots. Two blocks at D-Area received irrigation during the 2001 and 2002 growing seasons at a rate of 1.5 and 2.0 cm wk⁻¹, respectively, while control blocks received 0.1 cm wk⁻¹ irrigation. Trees received only rainwater in 2003. Granular fertilizer (112 kg N ha⁻¹) plus micronutrients was applied to D-Area blocks each spring. SRWC clones 110531, 112127, ST70, ST71, ST109, and ST260 were planted in eight-tree plots due to material availability; all others were planted in 16-tree plots. Trees at SRWC received 3.0 cm wk⁻¹ irrigation and were fertilized at a rate of 160 kg N ha⁻¹ annually. Both study sites were kept weed free throughout the experiment.

2.4. Pest damage assessment

Pest damage was visually monitored several times each growing season on the interior four trees per clonal plot. Where a clonal plot was comprised of only eight individuals (two rows of four trees), the center four trees were monitored. We recorded *C. scripta* defoliation using a 0–4 rating scale [33] where 0 = no *C. scripta* defoliation and 4 = severe (>75%) defoliation and/or terminal mortality on leaf plastochron index (LPI) 0–8. The LPI is a leaf-numbering system whereby the most apical leaf on a terminal or branch with a lamina length of ≥3.0 cm is termed “LPI 0”. Leaves are numbered positively moving toward the stem and negatively away from the stem [34]. *C. scripta* damage was recorded every year on both sites. *T. lobulifera* damage was recorded on LPI 0–12 using a 0–6 rating scale, where 0 = no *T. lobulifera* damage and 6 = extreme foliar curl and discoloration, >75% defoliation, or a dead terminal [23]. *T. lobulifera* damage was recorded in 2001 and 2002 at D-Area, and in 2002 at SRWC. Leaf rust was assessed using the Schreiner [35] scale, where values are assigned based on rust intensity and percent infestation; the values are then multiplied to obtain a final damage rating. *M. medusae* damage was recorded every year at D-Area, and at SRWC in 2002 and 2003.

2.5. Statistical analyses

Pest damage ratings were analyzed separately each season using a repeated measures analysis of variance (Proc MIXED; SAS Inc., Cary, NC). Damage ratings were recorded at different time intervals each growing season;

therefore, no comparisons between years were made. Means were compared using the Tukey's *t*-test ($\alpha = 0.05$).

At D-Area, we were most interested in the main effects of clone, irrigation, and time. The clone $\times$ irrigation, clone $\times$ time, and clone $\times$ irrigation $\times$ time interactions also were examined to determine if irrigated clones received less pest damage and to determine if pest damage varied over the course of a growing season. Since all trees at the SRWC site received the same treatment, we were only interested in the effects of clone, time, and the clone $\times$ time interaction. Finally, we compared 2003 *C. scripta* and *M. medusae* damage on clones at D-Area receiving the irrigation treatment with the same clones at SRWC (hereafter referred to as “common clones”; see [31]) to examine the effects of site, clone, and the site $\times$ clone interaction on damage levels.

A single control S7C1 and D105 clone survived at D-Area; these clones are not included in any analysis, but data are presented for comparison. By 2003, the only WV416 clones surviving at D-Area were in irrigated plots; therefore, this clone is not included in the 2003 D-Area analysis. At SRWC, no S7C1 or 112830 clones survived; thus they are not included in any analysis. Data from D105 was used only in the 2002 analysis, as this clone experienced 100% mortality by 2003.

3. Results

3.1. Clone effects

Damage by *C. scripta*, *T. lobulifera*, and *M. medusae* varied significantly among clones each year at both study

sites (Table 1). Overall, damage at both sites was consistent over three years, as indicated by the small changes in clonal rankings (Tables 2 and 3). Certain clones did, however, experience dramatic changes in damage rankings. For instance, *C. scripta* damage rankings at SRWC increased greatly on Eridano and 184-411 from 2002 to 2003 (Table 3).

3.2. Irrigation effects

Irrigation had a significant effect on pest damage ratings (Table 1). Greater *C. scripta* damage occurred on irrigated trees every year (Fig. 1). A significant clone $\times$ irrigation interaction occurred for *C. scripta* in 2001 and 2003, while a marginally significant interaction occurred in 2002 (Table 1). Greater *C. scripta* defoliation occurred on irrigated clones 110804 and S7C15 every year, whereas the irrigated NM6 trees had significantly less defoliation each year. Irrigated clone 7300502 had significantly less defoliation than control trees in 2002 and 2003. Kentucky 8 had greater *C. scripta* damage on trees receiving irrigation in 2003.

Irrigation alone had no effect on *T. lobulifera* damage, but the clone $\times$ irrigation interaction was significant each year (Table 1). No effect was observed in most clones; however, significantly less *T. lobulifera* damage occurred on clones 110804 and ST66 receiving irrigation in 2001 and 2002 (data not shown).

M. medusae damage was greatest on irrigated trees each year (Fig. 1), and damage was significantly affected by the clone $\times$ irrigation interaction (Table 1). Nearly all clones receiving irrigation showed increased *M. medusae* foliar

Table 1
ANOVA statistics for pest damage ratings during a 3-yr *Populus* clone trial in South Carolina

Pest	Source	D-Area			SRWC		
		2001	2002	2003	2001	2002	2003
<i>C. scripta</i>	Clone	****	**	****	dnr	****	****
	Irrigation	****	***	****	dnr	na	na
	Clone $\times$ irrigation	****	*	****	dnr	na	na
	Time	****	****	****	dnr	****	****
	Clone $\times$ time	***	*	****	dnr	****	****
	Clone $\times$ irrigation $\times$ time	ns	ns	ns	dnr	na	na
<i>T. lobulifera</i>	Clone	****	****	dnr	dnr	****	dnr
	Irrigation	ns	ns	dnr	dnr	na	dnr
	Clone $\times$ irrigation	***	**	dnr	dnr	na	dnr
	Time	****	****	dnr	dnr	****	dnr
	Clone $\times$ time	****	****	dnr	dnr	****	dnr
	Clone $\times$ irrigation $\times$ time	**	**	dnr	dnr	na	dnr
<i>M. medusae</i>	Clone	****	****	****	dnr	****	****
	Irrigation	****	***	****	dnr	na	na
	Clone $\times$ irrigation	****	***	****	dnr	na	na
	Time	****	****	****	dnr	****	****
	Clone $\times$ time	****	****	****	dnr	****	****
	Clone $\times$ irrigation $\times$ time	****	****	****	dnr	na	na

ns, not significant; * $P = 0.10 > 0.05$; ** $P = 0.05 > 0.01$; *** $P = 0.01 > 0.001$; **** $P < 0.001$; na, not applicable; dnr, data not recorded.

Table 2

Mean pest damage^a and rankings at D-Area over a 3-yr *Populus* clone trial in South Carolina

Pest	Clone	2001	Rank	2002	Rank	2003	Rank
<i>C. scripta</i>	S7C1	0.20	1	0.17	1	0.21	1
	7302801	0.24	2	0.22	2	0.22	2
	NM6	0.37	3	0.32	4	0.28	3
	WV415	0.44	4	0.28	3	0.33	4
	Eridano	0.55	6	0.56	11	0.39	5
	D105	0.60	7	0.45	8	0.45	6
	Kentucky 8	0.52	5	0.33	5	0.48	7
	I45/51	0.68	9	0.53	9	0.48	8
	WV94	0.72	10	0.38	7	0.51	9
	WV99	0.78	11	0.59	14	0.51	10
	7300502	0.67	8	0.33	6	0.53	11
	S7C15	0.86	14	0.58	12	0.53	12
	110804	0.83	13	0.54	10	0.58	13
	15–29	0.91	16	0.64	16	0.59	14
	S13C20	0.80	12	0.58	13	0.60	15
	WV416	0.89	15	0.64	17	0.68	16
	ST66	0.95	17	0.64	15	0.68	17
	WV316	1.43	18	1.00	18	0.88	18
<i>T. lobulifera</i>	NM6	0.00	1	0.00	1		
	I45/51	0.03	2	0.03	5		
	Eridano	0.10	6	0.13	8		
	15–29	0.07	3	0.08	7		
	7302801	0.20	9	0.25	9		
	D105	0.08	4	0.00	2		
	Kentucky 8	0.10	7	0.03	3		
	S13C20	0.13	8	0.04	6		
	WV415	0.09	5	0.03	4		
	7300502	0.37	15	0.25	10		
	S7C1	0.40	16	0.25	13		
	WV94	0.34	14	0.35	16		
	WV99	0.33	13	0.28	15		
	110804	0.51	17	0.46	18		
	WV416	0.31	11	0.25	11		
	S7C15	0.32	12	0.28	14		
	WV316	0.23	10	0.25	12		
	ST66	0.67	18	0.42	17		
<i>M. medusae</i>	NM6	0.01	1	0.00	1	0.01	1
	15–29	0.33	3	0.11	3	0.19	2
	I45/51	0.44	4	0.03	2	0.27	3
	7302801	0.18	2	0.11	4	0.47	4
	WV416	0.84	5	0.36	7	0.55	5
	Eridano	1.53	6	0.17	5	0.93	6
	7300502	4.37	7	0.33	6	2.91	7
	WV316	4.57	8	0.96	9	3.05	8
	Kentucky 8	7.72	10	1.83	12	5.65	9
	ST66	9.67	12	1.75	11	6.49	10
	S7C15	10.04	13	1.18	10	6.71	11
	110804	9.66	11	4.93	14	6.85	12
	S7C1	13.07	15	5.92	15	8.17	13
	S13C20	12.47	14	0.88	8	8.31	14
	D105	5.36	9	4.80	13	10.48	15
	WV99	16.93	17	12.75	18	11.59	16
	WV415	17.40	18	8.47	17	13.59	17
	WV94	14.58	16	5.98	16	14.44	18

^aDamage rating scale for *C. scripta* was from 0 to 4 on LPI 0 to 8, for *T. lobulifera* was from 0 to 6 on LPI 0 to 12, and for *M. medusae* was from 0 to 100 based on percent foliage infested and infestation severity.

Table 3

Mean pest damage^a and rankings at SRWC over a 3-yr *Populus* clone trial in South Carolina

Pest	Clone	2002	Rank	2003	Rank
<i>C. scripta</i>	OP367	0.00	1	0.95	1
	I45/51	0.00	1	1.06	2
	311–93	0.00	1	1.13	3
	7300502	0.36	5	1.20	4
	ST71	0.58	11	1.22	5
	ST261	0.43	8	1.23	6
	WV94	0.68	13	1.28	7
	S13C20	0.50	9	1.33	8
	ST109	0.76	14	1.35	9
	15–29	0.32	4	1.38	10
	Kentucky 8	0.90	16	1.38	10
	S7C15	0.37	6	1.43	11
	52–225	1.00	17	1.46	12
	ST260	0.50	9	1.50	13
	WV415	0.53	10	1.50	13
	ST66	0.60	12	1.50	13
	112127	1.20	20	1.50	13
	NM6	0.06	2	1.54	14
	184–411	0.27	3	1.60	15
	WV416	0.50	9	1.61	16
	110531	0.77	15	1.65	17
	WV316	1.18	19	1.65	17
	110804	0.60	12	1.67	18
	Eridano	0.40	7	1.70	19
	WV99	1.23	21	1.79	20
	7302801	1.05	18	1.84	21
	ST70	1.40	22	2.00	22
	ST264	1.80	23	2.03	23
	D105 ^b	0.40	7	na	
	S7C1 ^c	na	na	na	
	112830 ^c	na	na	na	
<i>T. lobulifera</i>	7302801	0.25	1		
	OP367	0.25	1		
	ST109	0.28	2		
	WV99	0.30	3		
	52–225	0.30	3		
	S7C15	0.31	4		
	ST264	0.32	5		
	184–411	0.33	6		
	7300502	0.36	7		
	D105	0.40	8		
	15–29	0.40	8		
	112127	0.40	8		
	ST260	0.40	8		
	ST261	0.43	9		
	WV94	0.44	10		
	WV415	0.47	11		
	I45/51	0.48	12		
	ST66	0.50	13		
	Eridano	0.50	13		
	ST71	0.50	13		
<i>M. medusae</i>	110804	0.53	14		
	WV316	0.55	15		
	110531	0.55	15		
	S13C20	0.56	16		
	ST70	0.60	17		
	NM6	0.66	18		
	Kentucky 8	0.80	19		
	WV416	0.83	20		
	311–93	1.10	21		
	S7C1 ^c	na			
	112830 ^c	na			

Table 3 (continued)

Pest	Clone	2002	Rank	2003	Rank
<i>M. medusae</i>	NM6	0.0	1	0.0	1
	15–29	0.0	1	0.0	1
	184–411	0.0	1	0.0	1
	311–93	0.0	1	0.0	1
	52–225	0.0	1	0.0	1
	I45/51	0.0	1	0.1	2
	7302801	0.0	1	0.2	3
	OP367	0.1	2	0.3	4
	ST70	0.0	1	0.6	5
	WV416	0.0	1	0.9	6
	7300502	3.4	4	2.8	7
	Eridano	4.5	5	3.0	8
	ST261	4.7	6	3.6	9
	S7C15	2.8	3	3.7	10
	ST66	7.5	8	6.3	11
	S13C20	7.2	7	6.7	12
	110531	10.2	10	7.5	13
	ST71	9.3	9	8.1	14
	WV99	16.8	12	11.7	15
	ST264	17.4	13	11.9	16
	Kentucky 8	15.8	11	12.0	17
	ST260	30.3	14	20.3	18
	WV316	32.7	15	22.6	19
	112127	33.0	16	23.4	20
	WV94	35.9	17	26.0	21
	WV415	36.7	18	28.3	22
	110804	49.0	19	32.3	23
	ST109	52.2	20	36.1	24
	D105 ^b	55.0	21	na	
	S7C1 ^c	na		na	
	112830 ^c	na		na	

^aDamage rating scale for *C. scripta* was from 0 to 4 on LPI 0 to 8, for *T. lobulifera* was from 0 to 6 on LPI 0 to 12, and for *M. medusae* was from 0 to 100 based on percent foliage infested and infestation severity.

^bClone D105 suffered 100% mortality prior to the 2003 growing season.

^cClones S7C1 and 112830 had 100% mortality prior to the initiation of pest damage monitoring.

damage. Clones WV94, WV415, and 110814 receiving irrigation all had significantly higher *M. medusae* damage levels each year, whereas clones S7C15, ST66, and Kentucky 8 receiving irrigation had higher damage levels in 2001 and 2003. Clone WV99 had lower *M. medusae* damage on trees receiving irrigation in 2002.

3.3. Site effects

In 2003, mean *C. scripta* damage ratings were significantly greater ($P < 0.001$) at SRWC (3.13 ± 0.03) compared to D-Area (2.88 ± 0.03), while *M. medusae* damage did not differ between the sites ($P = 0.64$). Greater *C. scripta* damage ratings on six clones at SRWC compared to D-Area were indicated by a significant site $\times$ clone interaction ($P < 0.001$). Three of these clones were hybrids (Eridano, NM6, and 15–29) while three were pure *P. deltoides* (S7C15, WV99, and S13C20). The site $\times$ clone interaction also was significant for *M. medusae* damage ($P < 0.001$);

Fig. 1. Effects of irrigation on (A) *C. scripta* and (B) *M. medusae* damage levels at D-Area during the 2001–2003 growing seasons. Means ($\pm$ SE) within a combination of pest and year sharing a letter are not significantly different (Tukey's HSD, $\alpha = 0.05$).

clones WV94 and 7300502 had higher damage ratings at D-Area and SRWC, respectively.

3.4. Time effects

Within each growing season, pest damage changed significantly at both sites (Table 1). At D-Area, *C. scripta* and *M. medusae* damage was significantly greater at the end of the 2001 and 2002 growing seasons, but in 2003 *C. scripta* and *M. medusae* damage was greatest in mid- and early summer, respectively (Fig. 2). *T. lobulifera* damage peaked in both mid-summer and late fall during 2001, but only in mid-summer 2002.

At SRWC, *C. scripta* damage was greatest in August each year, and stayed high through September in 2003 (Fig. 3). In 2002, *T. lobulifera* damage declined throughout the season until peaking in October (Fig. 3). *M. medusae* damage increased throughout the 2002 growing season, but peaked in mid-summer 2003 (Fig. 3).

Significant clone $\times$ time interactions occurred at D-Area in 2001 and 2003 for *C. scripta*, and every year for *T. lobulifera* and *M. medusae* (Table 1). All clone $\times$ time interactions at SRWC were highly significant (Table 1), indicating that pest damage on many clones changed during the growing season. Pest damage on individual clones fell into one of four general patterns throughout a growing season: (1) damage was relatively even (e.g., clones WV316 and 110804), (2) damage began low, then steadily escalated (e.g., clone Kentucky 8), (3) damage peaked once (e.g., clone 184–411), and (4) damage peaked twice (e.g.,

Fig. 2. Temporal pest damage at D-Area from 2001 to 2003: (A) *C. scripta*, (B) *T. lobulifera*, (C) *M. medusae*. Means ($\pm$ SE) within a year sharing a letter are not significantly different (Tukey's HSD, $\alpha = 0.05$). Damage ratings for *Tetra lobulifera* were not recorded in 2003.

clones OP367 and ST70). Clone $\times$ irrigation $\times$ time interactions at D-Area were significant each year for *T. lobulifera* and *M. medusae* only (Table 1).

4. Discussion

4.1. Clone effects

Intensively managed *Populus* plantations are subject to defoliation by a wide variety of pests [19], and a wide range of susceptibility occurs among clones and hybrids [11,12,18,25,36–38]. *C. scripta* preference [39] and damage [21] varies greatly among *Populus* clones and hybrids, as does that of *T. lobulifera* [23] and *M. medusae* [36,37]. Our study found that several clones of various *Populus* hybrids

and pure *P. deltoides* had low susceptibility to all three pests monitored. In addition, many clones performed very well with respect to two out of three pests (e.g., D105 at D-Area and 7302801 at SRWC).

Low overall pest pressure occurred throughout the duration of this study for each of the three pests. Most average *C. scripta* and *T. lobulifera* damage ratings were <1 at D-Area and <2 at SRWC. Only at SRWC did we see mean rust ratings over 30. Mean damage ratings >3 are common under heavy *C. scripta* defoliation pressure [20], while a *T. lobulifera* outbreak caused mean damage ratings of almost 4 [23]. Rust damage ratings of 100 have been observed in other studies [40], occasionally resulting in tree mortality [24]. When pest damage levels are low, it can be difficult to determine if certain clones perform better than

Fig. 3. Temporal pest damage at SRWC from 2002 to 2003: (A) *C. scripta*, (B) *T. lobulifera*, (C) *M. medusae*. *T. lobulifera* was not monitored in 2003. Means ($\pm$ SE) within a year sharing a letter are not significantly different (Tukey's HSD, $\alpha = 0.05$). Damage ratings for *Tetra lobulifera* were not recorded in 2003.

others; thus greater interpretation is required in these situations.

4.2. Irrigation effects

The effect of plant water stress on herbivore performance is an ongoing debate [41–43]. Generally speaking, damage and performance (i.e., body size, fecundity, developmental rate) of folivores is not consistently

affected, and that of sap-feeders is reduced on water stressed woody plants. Conversely, fungal pathogens tend to exhibit increased damage and greater performance in moist environments. Results from this study concur with these general trends. Although we did not explicitly measure pest performance, folivore (*C. scripta*) damage levels were highly variable on irrigated and control trees. Damage by *C. scripta* was inconsistent within years and among clones. It can be difficult to assess treatment (or in

our case, clonal) differences when pest populations (and subsequent damage levels) are low as seen previously with the Nantucket pine tip moth, *Rhyacionia frustrana* (Comstock) (Lepidoptera: Tortricidae) [14]. Daane and Williams [44] found *Erythroneura variabilis* Beamer (Homoptera: Cicadellidae) leafhopper populations on grapevines linearly related to irrigation amounts. However, we found increased sap-feeder (*T. lobulifera*) damage on non-irrigated trees in all but two clones. Previous research on *Populus* near our study site found the effects of irrigation on *T. lobulifera* damage to be inconsistent [23]. Results from this study do not support the argument that irrigation increases sap-feeder damage. With the exception of clone WV99 in 2002, where significant clone $\times$ irrigation interactions occurred, the highest *M. medusae* damage always occurred on the irrigated trees.

4.3. Site effects

Several clones experienced greater *C. scripta* damage at SRWC compared to D-Area. The D-Area site was located in a bottomland area on sandy loam soil, whereas the SRWC site was on an upland sandy site surrounded by a pine forest [30]. Greater beetle recruitment most likely occurred at SRWC due to greater *Populus* foliage in surrounding plantations, and this may have influenced *C. scripta* population pressure in our study. Ostry and McNabb [36] found site to be a significant influence on disease occurrence and severity in the north-central US. However, the site did not affect *M. medusae* damage levels, but certain clones were more susceptible at a particular site. Caution should be exercised when interpreting these data, as site comparisons were made during 1 year only.

4.4. Time effects

Inter-year variation was small, as clones with low susceptibility remained so throughout the study (especially at D-Area). But some exceptions did occur (e.g., NM6 and Eridano susceptibility to *C. scripta* at SRWC). These patterns also were seen with canker ratings in Argentina, where some clones remained highly ranked between years 3 and 10 and some changed rankings substantially [45].

Often, pest damage ratings are taken at a single time during a study [11,46] or growing season [13,37]. Herbivore damage is known to exhibit temporal variation [47]. Previous work with *C. scripta* [48], *T. lobulifera* [23] and *M. medusae* [24] has indicated changing damage levels over the course of a single growing season. Thus, adequate sampling on a temporal scale must occur to ensure accurate data. Results from our study confirm the importance of taking multiple damage ratings over the course of a season. Single sampling dates may misrepresent what is actually occurring, and consequently may misrepresent clonal performance.

5. Conclusions

Based on these preliminary data, hybrid clones NM6, Eridano, I45/51, OP367, and 15–29 and *P. deltoides* clones 7302801, 7300502, and Kentucky 8 all had generally low pest susceptibility. At SRWC, OP367 and 7300502 were highly resistant to all three pests; I45/51 was highly resistant to *C. scripta* and *M. medusae*; NM6 and 15–29 were highly resistant to *M. medusae*; and 7302801 was highly resistant to *T. lobulifera* and *M. medusae*. At D-Area, NM6, Eridano, I45/51, and 7302801 were highly resistant to all three pests; clone 7300502 was highly resistant to *M. medusae* only. The effects of irrigation on insect performance and damage were very situational, and results from this study neither disputed nor supported the plant stress hypothesis [43]. However, increased damage ratings by *C. scripta* and *M. medusae* occurred on irrigated clones. Care must be taken to match clones to sites on which their performance will be optimized. Finally, knowledge of temporal variation in pest damage patterns is crucial to accurate pest monitoring.

Ultimately, several variables must be taken into consideration when evaluating and selecting *Populus* clones for further testing or deployment. We monitored pest susceptibility in this study, and survival and growth previously [31] on these clones. Clones I45/51, Eridano, and NM6 exhibited excellent survival rates [31], and exhibited low pest susceptibility. WV416 grew well at both sites, but was susceptible to *C. scripta* and *T. lobulifera*. S13C20 and Kentucky 8 grew well at D-Area, and were relatively resistant to all three pests. Hybrids 184–411 and 52–225 grew well at SRWC, and were resistant to *T. lobulifera* and *M. medusae*.

M. medusae currently cannot be managed using cultural or chemical techniques; therefore, resistance to this pest is very important. If cultural management strategies were applied for *C. scripta* and *T. lobulifera* when necessary, clones WV416, S13C20, 184–411, 52–225, and Kentucky 8 may warrant additional evaluation in the southeastern US coastal plain.

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