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3 **Resistance of chestnut trees to infestation by Asian chestnut gall wasp**

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13 **Abstract**

14 Asian chestnut gall wasp (*Dryocosmus kuriphilus*) was introduced into Georgia
15 (USA) in 1975 and has been spreading north throughout the range of American
16 chestnut (*Castanea dentata*). This pest is now present throughout most of Tennessee.

17 In 2003, it was found near Cleveland, Ohio and has been spreading south from there.

18 In 1995, hybrid chestnuts with *C. dentata* female parents and two Ozark chinquapin X
19 Chinese chestnut male parents (*C. ozarkensis* X *C. mollissima*) were planted in North

20 Carolina, where this introduced insect is now naturalized. Of the 93 trees planted, 69

21 survived four years, and 36 were still surviving after 14 years. The surviving trees were

22 evaluated for the presence of Asian chestnut gall wasp galls in 2006 and 2009. After 14

23 years, the progeny of one male parent had 67% survival and of the other, 16% survival.

1 Among the survivors, 31 had no wasp galls in 2009 and 4 had 10 or fewer galls. The
2 four female parents (American chestnuts and half-sibs) were assumed to be fully
3 susceptible, so the genes controlling resistance to infestation cannot be cytoplasmic. If
4 resistance is conferred by one or two nuclear genes, resistance can easily be
5 transferred into timber chestnuts and orchard chestnut cultivars.

6

7 **INTRODUCTION**

8 Asian chestnut gall wasp, *Dryocosmus kuriphilus*, (Payne et al. 1976; Rieske, 2007) has
9 now spread from the state of Georgia, where it was introduced in 1974, to most of the
10 southern growing area of chestnut trees in the United States, and to the vicinity of
11 Cleveland, OH (Rieske, 2007; Stehli, 2003; Stehli, 2006). American and Chinese
12 chestnuts (*Castanea dentata* and *C. mollissima*) all appear to be susceptible to this pest
13 (Payne et al 1976). At present there are few predators to stop gall wasp from moving
14 throughout the range of American chestnut trees (Cooper et al., 2007). Eggs are laid in
15 leaf and flower buds, the developing galls kill the leaves and flowers, and infestations
16 can result in tree death. The pest was found in northern Italy in 2002, and European
17 chestnut (*C. sativa*) and many commercial hybrids were infested with levels serious
18 enough to result in significant tree mortality (Sartor et al., 2009). No pesticides are
19 currently registered for control of this insect. In 1995, hybrid chestnuts were planted in
20 the state of North Carolina, in cooperation with the U.S. Forest Service, to test the trees
21 for resistance to infestation in an area where Asian chestnut gall wasp was present.

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23

1 MATERIALS AND METHODS

2 The research plot was established at 760 m elevation in the Southern Appalachian
3 Mountains (35°30'N, 82°37'W) about 16 km southwest of Asheville, NC where both
4 chestnut blight disease (caused by *Cryphonectria parasitica*) and Asian chestnut gall (*D.*
5 *kuriphilus*) were present. The study site was located along an edge of a large (>4 ha)
6 forest opening that had been cleared of vegetation in 1994. The trees surrounding the
7 study site were primarily scarlet oak (*Quercus coccinea*) and white oak (*Q. alba*), with
8 an understory of red maple (*Acer rubrum*) and sourwood (*Oxydendrum arboreum*);
9 scattered root sprouts of American chestnut (*C. dentata*) were also present. The study
10 site was 0.05 ha, consisting of five rows spaced 3.05 m apart, and trees were spaced
11 1.5 m apart within each row.

12 The female parents of the hybrids planted were four different American chestnut
13 trees that were half-sibs, and the male parents (called M1 and M2) were two different
14 trees of *C. ozarkensis* X *C. mollissima* (Ozark chinquapin crossed with Chinese
15 chestnut; the Ozark chinquapin and Chinese chestnut parents of these two trees were
16 not the same) (Table 1) and both male parents had good resistance to chestnut blight
17 disease. The hybrids were from hand-pollinated crosses made in 1993 in Connecticut
18 (seed planted in April 1994). A total of 93 seedlings were dug in April 1995 and
19 delivered (bare-root) to the planting site in early May 1995. We used a completely
20 randomized experimental design consisting of single-tree plots and no perimeter buffer
21 row. Competing vegetation in the acidic (pH<5.5) silty to sandy loam sediment material
22 consisted mostly of blackberry (*Rubus* spp.) canes, which were cut at ground level
23 during the late winter for the first four years of the study. Most of the seedlings had

1 grown to sapling size by 1999 and had formed a closed canopy that shaded competing
2 vegetation. The trees were examined in the fall of 2006 (Anagnostakis et al., 2009) and
3 2009, and the presence of chestnut blight disease cankers on the trunks and galls on
4 terminal branches were recorded.

5

6 **RESULTS**

7 In 1999, 26 of the 44 hybrids with M1, and 43 of the 49 hybrids with M2 were still alive.
8 By 2006, 17 of 44 and 36 of 49 survived; by 2009, the survival rate fell to 7 of 44 and 29
9 of 49 respectively for M1 and M2 progeny. In 2009, all but one of the survivors had
10 cankers of chestnut blight disease on their trunks (i.e., one tree had no cankers) and
11 seven had died to the ground and had large sprouts with small blight cankers.

12 In 2006, 36 of the total 53 survivors had few (one to 10) or no wasp galls. In
13 2009, 35 of the 36 survivors had few or no wasp galls (Table 2).

14

15 **DISCUSSION**

16 Asian chestnut gall wasp has been a serious problem in Japan on Japanese chestnut
17 (*C. crenata*) for many years, and efforts have been made to select cultivars with some
18 tolerance to the insect. Kotobuki and his colleagues (1984) made 26 crosses (770 total
19 progeny) of Japanese cultivars of chestnut, and rated them for presence of galls. When
20 rated after 5 or 6 years, 50% or more of the progeny of 11 crosses had "no galls on
21 tree" or "very small number of galls on weak twigs." Cultivar 'Ginrei' conferred
22 resistance to progeny five times when used as a female parent, and twice when used as
23 a pollen parent. Other cultivars used in crosses had fewer resistant progeny. The

1 authors did not offer any conclusions about genetic control of resistance to gall wasp
2 infestation. However, Kotobuki said that significantly more offspring had resistance if
3 the parents were cultivars with good resistance than if the parents were less resistant.
4 Sartor *et al.* (2009) reported that cultivar 'Bouche de Betizac' (a Japanese X European
5 hybrid) showed good resistance to infestation in their tests. None of these cultivars are
6 available in our collection, and no current information is available about how well the
7 resistance has held up in the presence of Asian chestnut gall wasp in Japan.

8 We expected genes from the Chinese chestnut parents to impart partial
9 resistance to chestnut blight disease to some of the hybrids. This would allow some
10 trees to survive long enough to be rated for gall wasp resistance. Chinese chestnut
11 trees have never been reported to have any resistance to gall wasp infestation.
12 Because gall wasp resistance was observed in Allegheny and Chinese chinquapins (*C.*
13 *pumila* and *C. henryi*) (Anagnostakis, personal observation), we used male parents that
14 were Chinese chestnut crossed with Ozark chinquapin (*C. ozarkensis*), which is a
15 timber tree (unlike *C. pumila*) with more winter hardiness than *C. henryi* and some
16 resistance to chestnut blight disease (Anagnostakis, personal observation). If the Ozark
17 chinquapins had genes for resistance to gall wasp infestation in their cytoplasm, the *C.*
18 *ozarkensis* X *C. mollissima* trees used as male parents would have carried these genes,
19 but none of our planted hybrids would have any resistance, since American chestnuts
20 (the mother-trees) all appear to be susceptible. If nuclear genes for resistance were
21 dominant, the resistance phenotypes should be segregating among the progeny. We
22 would expect half of the progeny to be resistant, if resistance was controlled by a single

1 dominant gene; one out of four if two dominant genes, or one out of eight if there were
2 three dominant genes.

3 Since there was segregation for susceptibility to gall wasp infestation among our
4 surviving hybrids, the genes for resistance to infestation cannot be cytoplasmic. The
5 numbers of survivors are too small to determine the exact number of resistance genes
6 in the male parents. The 28 of the 36 surviving hybrids with good to moderate chestnut
7 blight disease resistance all had few or no galls.

8 New crosses will be made and tested in the future. Larger numbers of progeny
9 will help us determine whether resistance to Asian chestnut gall wasp is simply
10 inherited. If so, that resistance can easily be transferred by breeding to timber and
11 orchard chestnut types at risk.

12

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18

19 **Literature Cited**

20 Anagnostakis SL (1998) Chestnuts in Georgia. Annu. Rept. Northern Nut Growers
21 Assoc. 89:5-14

22

1 Anagnostakis SL, Clark S, McNab H (2009) Preliminary report on the segregation
2 of resistance in chestnuts to infestation by oriental chestnut gall wasp. Acta Hort.
3 #815, ISHS, A. Soylu and C. Mert eds., p.33-37
4
5 Cooper WR, Rieske LK (2007) Community associates of an exotic gallmaker,
6 *Dryocosmus kuriphilus*, (Hymenoptera: Cynipidae) in eastern North America. Ann.
7 of the Ent. Soc. Amer. 100:236-244
8
9 Kotobuki K, Machida Y, Sato Y, Kajiura I, Kozono T (1984) Genetics of the
10 resistance to the Chestnut Gall Wasp (*Dryocosmus kuriphilus* Yasumatsu), harvest
11 date, mean nut weight, and the characteristics of selected clones of chestnut: results
12 of the fourth chestnut breeding program. (in Japanese) Bull. Fruit Tree Res. Stn.
13 11:43-53
14
15 Payne JA, Green RA, Lester CD (1976) New nut pest: an Oriental Chestnut Gall
16 Wasp in North America. Annu. Rep. Northern Nut Growers Assoc. 67:83-86
17
18 Rieske LK (2007) Success of an exotic gallmaker, *Dryocosmus kuriphilus*, on
19 chestnut in the USA: a historical account. Bull. Euro. and Mediter. Pl. Prot. Org.
20 37:172-174
21
22 Sartor C, Botta R, Mellano MG, Beccaro GL, Bounous G, Marinoni DT, Quacchia A,
23 Alma A (2009) Evaluation of susceptibility to *Dryocosmus kuriphilus* Yasumatsu

1 (Hymenoptera: Cynipidae) in *Castanea sativa* Miller and in hybrid cultivars. Acta
2 Hort. #815, ISHS, A. Soylu and C. Mert eds., p289-297

3

4 Stehli R (2003) Oriental chestnut gall wasp found in N.E. Ohio. The Nutshell
5 57:1&19

6

7 Stehli R (2006) Oriental Chestnut Gall Wasp in Ohio – an update. The Nutshell
8 60:27

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 2 Table 1. Source of the hybrid chestnut trees planted in 1995 in North Carolina in an
 3 area where Asian chestnut gall wasp (*Dryocosmus kuriphilus*) and chestnut blight
 4 disease (*Cryphonectria parasitica*) were present.

Female parent*	Male parent 1	#trees planted	#survivors 2009
American 1	Ozark chinquapin 1 X Chinese 1	6	1
American 2	Ozark chinquapin 1 X Chinese 1	18	1
American 3	Ozark chinquapin 1 X Chinese 1	20	5
	Male parent 2		
American 4	Ozark chinquapin 2 X Chinese 2	49	29

5
 6 *The four American trees were half-sibs from a female tree in a woodlot in Rocky Hill,
 7 CT; Ozark chinquapins 1 and 2 were from Russelville, Arkansas, planted 1936 in
 8 Hamden, CT; Chinese chestnut 1 was a graft of Luther Burbank's 'Miracle', planted
 9 about 1949 in Hamden, CT; Chinese chestnut 2 was Plant Introduction #70315 planted
 10 1930 in Hamden, CT.

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 12

1 Table 2. Number of trees with chestnut blight cankers (caused by *Cryphonectria*
 2 *parasitica*) on their trunks and number with Asian chestnut gall wasp (*Dryocosmus*
 3 *kuriphilus*) galls on terminal branches, in 2006 and 2009. Trees had two different male
 4 parents (M1 and M2) and the female parents were American chestnuts that were half-
 5 sibs. All were planted in North Carolina in 1995.

Male	Blight cankers on trunks*				Galls on terminal branches**					
	2006		2009		2006			2009		
	+	-	+	-	0	1-10	≥11	0	1-10	≥11
M1	17	0	7	0	6	7	2	6	1	0
M2	20	16	28	1	5	18	13	25	3	1

6
 7 *Chestnut blight disease cankers present (+) or absent (-) on trunks; **Wasp gall
 8 numbers: 0 – no galls detected on branch terminals, 1 – 10 galls noted on branch
 9 terminals, 11 or more galls noted on branch terminals

10