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Herbivory and tree mortality across a pinyon pine hybrid zone

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Abstract We examined the abundances of three common insect herbivores on pure and hybrid pinyon pines along a 250-km transect in west-central Arizona, United States. Using six morphological traits, we developed a hybrid index to classify trees as pure *Pinus californiarum*, hybrid, or pure *Pinus edulis*. The insects (the stem-boring moth, *Dioryctria albovittella*, the scale insect, *Matsucoccus acalyptus*, and several species of pitch moths that produce wounds on the trunk and branches) exhibited different distributional patterns across tree types. Stem-boring moths were significantly more abundant on trees at “hybrid” sites compared to trees at “pure” sites. In addition, within hybrid sites, hybrids supported significantly more moth larvae than pure trees of either species. These two patterns support the hybrid susceptibility hypothesis in which hybrid breakdown results in increased susceptibility to herbivory. In contrast to stem-borers, there were significantly more pitch moth wounds on trees at pure *P. californiarum* sites than at hybrid and pure *P. edulis* sites. Within the hybrid zone, pitch moth abundance was equal on pure *P. californiarum* and hybrids, and both were significantly greater than on pure *P. edulis*. These within-site comparisons support the dominance hypothesis where hybrid resistance differs from one tree species, but not the other. Scale insects exhibited the most restricted distribution; over the 250 km transect they were found only in the hybrid zone. This supports the hybrid susceptibility and/or the stress hypothesis (i.e., species at the edge of their range suffer greater stress and are more susceptible to herbivory). We summed the mean numbers of these three common herbivores across sites and found that hybrid sites supported 2.1 and 3.9 times more herbivores than pure *P. californiarum* and *P. edulis* sites, respectively. Furthermore, tree mortality was on average, 35 times greater within the hybrid zone compared to pure zones of each species and was associated with the cu-

mulative abundance of herbivores ($r^2 = 0.646$). Regardless of whether this mortality is due to insect infestation, stress or a combination of both, these results suggest that hybrid zones are important arenas of natural selection.

Key words *Dioryctria albovittella* · Herbivory · Hybrid *Matsucoccus acalyptus* · *Pinus*

Introduction

Hybridization in plants is widespread (Grant 1981) and may have been the mechanism of speciation for 30–80% of extant angiosperm species (e.g., Stace 1987). Ecologists have recently begun to examine how the patterns of plant hybridization in nature might affect other organisms. Several studies have found increased phytophage abundance on hybrid plants relative to parentals (Drake 1981a,b; McClure 1985; Whitham 1989; Barker 1990; Floate and Whitham 1993; Floate et al. 1993; Ericson et al. 1993), but fewer (Mitchell et al. 1974; McClure 1985; Boecklen and Spellenberg 1990) and intermediate numbers also have been found (Manley and Fowler 1969; McClure 1985; Aguilar and Boecklen 1992). Additionally, mixed responses have been found in studies where the abundances of numerous phytophages have been examined in a single system (Fritz et al. 1994; Morrow et al. 1994; Whitham et al. 1994).

Because plant hybrid zones are genetically diverse (Riesberg and Wendel 1993) and exhibit great variation in resistance to herbivores (Paige et al. 1990; Paige and Capman 1993), they have the potential to influence herbivore evolution and vice versa (Moran and Whitham 1988; Whitham 1989; Moran 1991; Floate and Whitham 1993). Hybridization in plants may also influence diverse organisms and affect species interactions (e.g., plant pathogens: Ericson et al. 1993; Fritz et al. 1994; Whitham et al. 1994; mutualisms: Floate and Whitham 1994; nesting of birds: Martinsen and Whitham 1994). Additionally, because some insect species appear to be

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restricted to plant hybrid zones, conservation issues need to be considered (Whitham et al. 1991, 1994).

Few studies have examined hybridization in conifers as it affects herbivory (Manley and Fowler 1969; Mitchell et al. 1974; McClure 1985). Because resistance characteristics of conifers may differ from the more widely studied broadleaf hybrids (e.g., oleoresin defenses versus tannins), general pattern(s) of herbivore response to hybrid conifers are unknown. Here, we compared the abundances of the stem-boring moth (*Dioryctria albobittella*), pitch moths (in the genera *Dioryctria*, *Synanthedon* or *Vespa mima*) and the scale insect (*Matsucoccus acalyptus*) on pure and hybrid pinyon pines (*Pinus californiarum* X *P. edulis*). In view of their intermediate needle morphology (a mixture of one- and two-needled fascicles), pinyon pines in west-central Arizona have been identified as hybrid by several investigators (Lanner 1974; Bailey 1987; Malusa 1992; Lanner and Phillips 1992).

Numerous hypotheses have been proposed to describe patterns of herbivory on hybrid plants relative to their parental species. Here, we examine four genetic-based hypotheses described by Fritz et al. 1994 regarding the resistance traits of hybrids. These hypotheses are: (1) The additive hypothesis, which predicts that hybrids will not differ from the mean resistance of the parents; (2) The dominance hypothesis, where hybrid resistance differs significantly from the mean resistance of both parents, but not from one of the parents; (3) The hybrid susceptibility hypothesis, which predicts higher herbivore densities and/or higher herbivore performance on hybrids versus parental species; and (4) The hybrid resistance hypothesis, which predicts that hybrid plants will be more resistant than either parent. The occurrence of these patterns in nature likely depends upon the genetic resistance mechanisms involved (e.g., dominant or recessive, monogenic or polygenic; Gallun and Khush 1980) and the pattern of hybrid introgression (e.g., F1 sterility, unidirectional or bidirectional; Keim et al. 1989; Floate and Whitham 1993). These two factors will maintain or disrupt coadapted gene complexes and affect genetic interactions such as those occurring in heterosis.

An additional hypothesis, the stress hypothesis, proposes that increased environmental stress at the edge of a species' range (e.g., water and nutrient deficits, temperature extremes) may result in increased herbivory in the hybrid zone (Whitham et al. 1994). Although plant stress is often associated with increased herbivory (e.g., White 1969), the reverse situation may be common. Vigorous plants suffering little stress are the preferred hosts for many herbivores (Price 1991). Furthermore, environmental and genetic hypotheses are not mutually exclusive; genotype by environment interactions may also influence plant resistance (Tingey and Singh 1980).

Using this hybrid system, we address the following questions:

1. What is the geographic pattern of hybridization between two pinyon pine species along a 250-km transect as determined by morphological analyses?

2. What are the geographic patterns of herbivory across pure and hybrid zones?

3. Within the hybrid zone, how does herbivory vary among pure and hybrid trees?

4. Is tree mortality related to the cumulative level of herbivory across sites?

Questions 2 and 3 allow us to distinguish between the relative importance of site differences (e.g., greater stress at the boundary of a species' distribution) and genetics in affecting herbivore distributions.

Methods

Morphological analyses

We chose 15 sites that spanned approximately 250 km from pure *P. californiarum* populations near Kingman (site 1) in western Arizona, USA to pure *P. edulis* populations near Flagstaff (site 15) in central Arizona. At each site, 20 trees were permanently marked, and we collected a small branch (approx. 20 shoots) from the west side of the tree at a height of approximately 2 m. In the laboratory, a total of 100 fascicles were haphazardly taken from all parts of each branch and pooled. We measured, counted, or estimated from these 100 needles the following characters that have been used in most taxonomic studies of pinyon pines (Little 1968; Bailey 1987; Malusa 1992; Lanner and Phillips 1992): (1) percent one-needled fascicles, (2) length of the longest needle, (3) number of stomatal lines on the longest needle, (4) number of resin ducts in the longest needle, (5) fascicle sheath length of a representative 1st-year needle (as these needles are typically the only ones with intact sheaths), and (6) the amount of curl back of the fascicle sheath measured in (5) was categorized as 1 if it was estimated to be <120°, 2 if it was 120–240°, and 3 if it was >240°.

We used stepwise discriminant function analysis (Jennrich and Sampson 1983) to identify morphological characters that varied most among the 15 sites and to create a morphological hybrid index based upon the values of the canonical variable that best separated *P. californiarum*, hybrids, and *P. edulis* trees. We examined the values of the first canonical variable for trees at pure sites of both species where there were no trees of intermediate morphologies and used these ranges of values to classify trees within the hybrid zone as pure or hybrid. This canonical variable explained 90.8% of the total dispersion of the trees in discriminant space.

Insect censuses

On the same trees used for morphological measurements, we performed 1-min surveys of each insect. For *D. albobittella*, we counted the number of dead shoots per tree that resulted from the tunneling of their larvae. For *M. acalyptus*, we counted the number of scales observed on the needles of each tree, and for pitch moths, we counted the number of wounds exuding large amounts of resin from the tree trunk and major branches. Although we have not been able to identify this species, we are confident that these wounds result from insect infestation because of the presence of numerous tunnels found beneath the wounds. The pitch moth species likely to create this damage are found in the genera *Dioryctria*, *Synanthedon* and/or *Vespa mima* (Cain et al. 1983; Cranshaw et al. 1992).

We censused stem-boring moths and scale insects from 1990 to 1992 at sites 1–11, except in 1991 when sites 3, 4 and 6 were inaccessible. In 1992, sites 12–15 were added to our transect to better represent pure *P. edulis* in our insect surveys. Even though scale insects were absent from all sites in 1992, because they scar needles, and pinyon pines retain their needles for 6–7 years, we could census the damage from previous years of scale attack. Resin wounds were counted in 1992 at all sites.

Statistical analyses

As other studies have found that different hybrid classes (e.g., F1 versus backcross hybrids) support different abundances and species of herbivores (e.g., Morrow et al. 1994), we initially separated hybrid pinyons into classes. Based upon the value of the first canonical variable, we classified trees with scores between -2.0 and 0 as "backcrosses" to *P. edulis*, scores between 0.1 and 2.0 as "F1 type hybrids", and scores between 2.1 and 4.0 as "backcrosses" to *P. californiarum*. We recognize that these classifications may be artificial, but used them for convenience. We compared average stem-borer abundance, pitch moth abundance in 1992 and the cumulative abundance of stem-boring moths and pitch moths among these hybrid tree classes using the Kruskal-Wallis test (Conover 1980). Because no significant differences were found among classes of hybrids in any comparison (stem-boring moth: $T = 2.94$, $P = 0.23$, $df = 2$; pitch moth: $T = 0.8$, $P = 0.67$, $df = 2$; cumulative stem-boring moth and pitch moth: $T = 2.53$, $P = 0.28$, $df = 2$), all hybrid classes were pooled for the remaining analyses.

Among sites, we compared the number of dead shoots from stem-boring moths, the number of wounds from pitch moths, and the number of scales counted in timed surveys. The mean number of insects or damaged plant parts counted during the study was used for each tree (data from sites 12–15 are not averages as they were only censused in 1992). Following a rejection of the null hypothesis in a Kruskal-Wallis test, we performed a nonparametric contrast (Daniel 1990) of insect abundance on trees pooled from hybrid sites (6–11) versus pure sites (1–5 and 12–15) of each tree species. The absolute value of the difference in mean rank sums was compared to a critical value determined by multiplying $Z_{\alpha^*/2(k-1)}$ by the square root of $N(N+1)/12 \cdot (1/n_i + 1/n_j)$ where α^* is the experiment-wise error rate set at 0.10%, k is the number of groups (3) and N is the sum of the two sample sizes being contrasted (n_i, n_j).

Within the hybrid zone, we compared the average number of dead shoots from 1990 to 1992 and the number of pitch moth wounds in 1992 between pure and hybrid trees using the Kruskal-Wallis test. We classified each tree according to the value of the first canonical variable as described above. We also performed nonparametric contrasts using the methods described above to determine if the levels of herbivory were significantly different between hybrid and pure trees in the hybrid zone.

To better visualize overall trends in herbivore population abundances, for each of the 15 sites we summed the mean number of scale insects, the mean number of shoots killed by stem-borers, and the mean number of pitch moth wounds observed in 1992. This resulted in a cumulative score of herbivore abundance for each site. We also calculated a cumulative score for the different tree types within the hybrid zone classified as described above. We compared the cumulative numbers at pure and hybrid sites, and pure and hybrid individuals within the hybrid zone, again using the Kruskal-Wallis test.

Tree mortality

In 1991, we quantified tree mortality at each site by walking a linear transect from north to south through the stand. Each tree encountered on the transect (within 5 m of the observer) was classified as live or dead. We walked until 100 trees had been encountered, and compared percent tree mortality among sites using a χ^2 goodness of fit test (Conover 1980). Although all trees that were dead had resin clumps characteristic of pitch moth attack (Cain et al. 1983; Cranshaw et al. 1992), we could not be certain that the moth was the actual cause of death. To determine if there was a predictive relationship between cumulative herbivory and percent tree mortality across the 15 sites, we regressed percent mortality on cumulative herbivory.

Results

Tree morphology

Tree morphology varied significantly among sites (MANOVA $P < 0.05$), where percent one- and two-needled fascicles best discriminated among sites as it was included first in the stepwise discriminant analysis. This character changed from near 100% single needles to near 100% double needles from west to east. Although needle number entered the model first, the other characters also entered the model except for curl index, indicating that they added significantly to the separation of sites. A plot of site centroids shows three groupings where sites 1–5, 6–11 and 12–15 are together (Fig. 1) representing pure *P. californiarum*, hybrids and pure *P. edulis*, respectively. Insect abundances among these groupings will be contrasted below.

The values for the first canonical variable did not overlap between pure populations of each species (Fig. 2A) and showed a continuous distribution through the hybrid zone (Fig. 2B) suggesting bidirectional introgression between species. Figure 2B shows that within the hybrid zone some trees had pure morphologies. This allowed us to contrast herbivory on pure and hybrid trees within a site and thus factor out potential site effects.

Stem-borer abundance 1990–1992

Abundance of the stem-boring moths varied significantly among sites ($T = 71.51$, $P < 0.0001$, $df = 14$) where trees at hybrid sites (6–11) supported significantly more moths than trees at pure sites (Fig. 3A; $Z = 44.0 > Z_{crit,0.05} = 17.7$). These data are consistent with both the hybrid susceptibility and stress hypotheses.

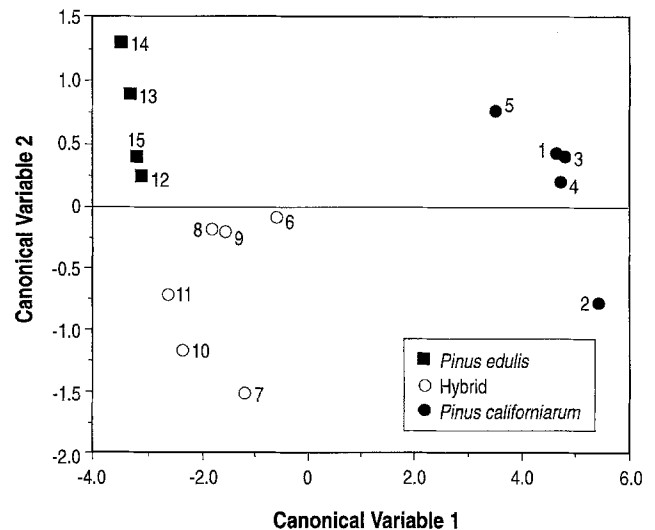


Fig. 1 A plot of site centroids in canonical space based upon six morphological measurements for each tree. Sites 1–5, 6–11 and 12–15 appear to be grouped

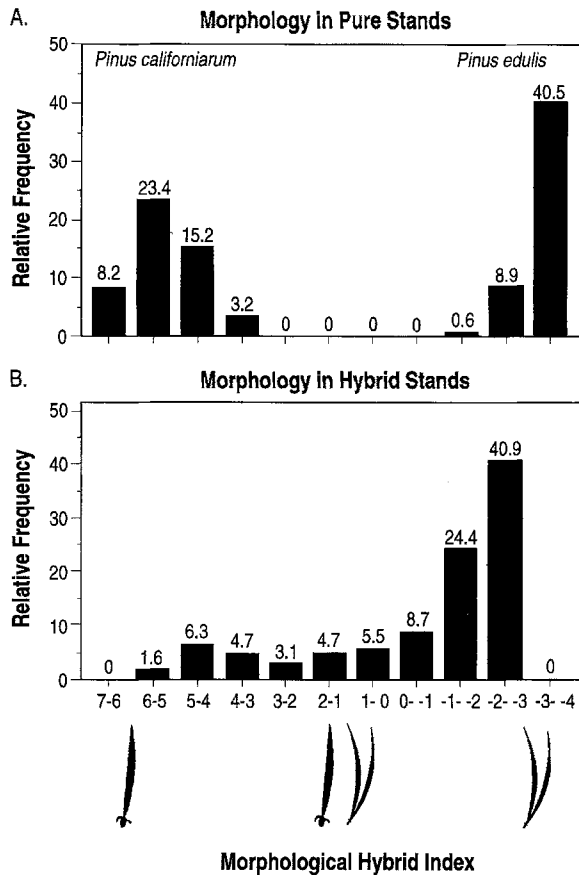


Fig. 2 A Frequency distributions of the morphological hybrid index for individual trees show that there is clear separation between pure stands of *Pinus californiarum* and *P. edulis*. B The frequency distribution of hybrid indices of trees within the hybrid zone shows that many of the trees exhibit similar morphologies to pure *P. edulis* trees, but that there is a continuum of morphological types into the pure *P. californiarum* morphotypes

Within the hybrid zone, the abundance of stem-boring moths also varied significantly among tree types (Fig. 3B; $T = 14.99$, $P = 0.0047$, $df = 2$) where hybrid trees suffered significantly greater shoot mortality than either parental species (hybrid: *P. californiarum* contrast; $Z = 24.9 > Z_{crit,0.05} = 10.1$; hybrid: *P. edulis* contrast; $Z = 18.7 > Z_{crit,0.05} = 8.8$). Thus, even when environmental stress or site effects were held constant, hybrids suffered greater shoot mortality from stem-boring moths relative to pure trees of both species. These data support the hybrid susceptibility hypothesis and suggest that hybrids suffer breakdown in resistance to the stem-boring moth.

Pitch moth abundance

Although mean pitch moth abundance did not appear to vary significantly between hybrid and pure *P. californiarum* sites (Fig. 4A), comparisons based on ranks shows that there were significantly more wounds on trees at pure *P. californiarum* sites than hybrid sites ($T = 51.8$, $P < 0.0001$, $df = 14$). Furthermore, the number of pitch moth wounds on trees at hybrid sites did not differ from

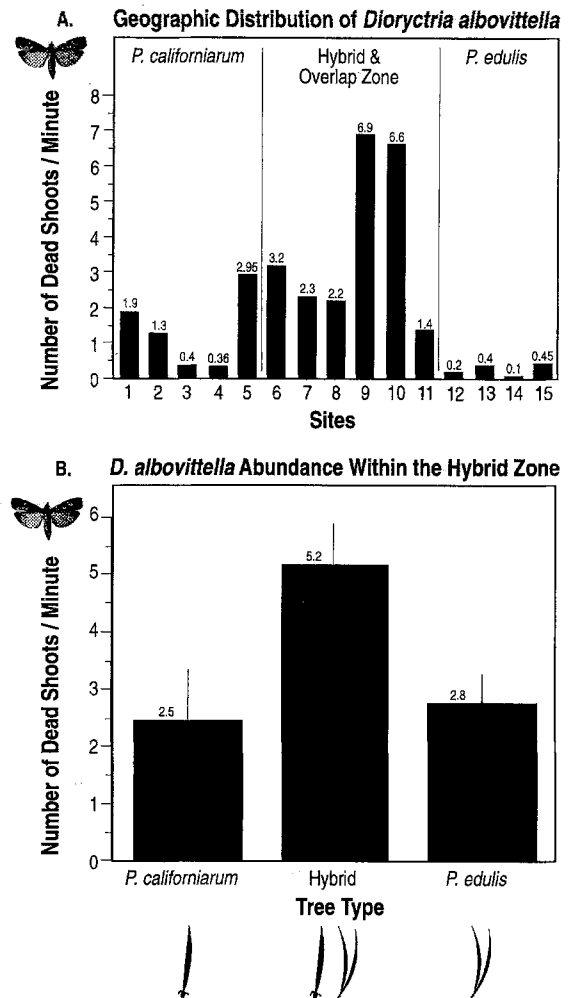


Fig. 3 A Stem-boring moth abundance was greatest at hybrid sites compared to pure sites as shown by the mean number of dead shoots counted per minute across the 15-site transect. B Within the hybrid zone, stem-boring moth abundance was greatest on hybrid trees compared to pure trees of both species as shown by the mean number of dead shoots counted per minute. Bars indicate the mean ± 1 standard error of the mean

those recorded at pure *P. edulis* sites (*P. californiarum*-hybrid contrast; $Z = 36.7 > Z_{crit,0.05} = 12.0$; hybrid-*P. edulis* contrast; $Z = 10.48 < Z_{crit,0.05} = 11.7$).

Within the hybrid zone, hybrids and pure *P. californiarum* had equal numbers of wounds (Fig. 4B; $T = 21.6$, $P < 0.001$, $df = 2$) and both had significantly more wounds than pure *P. edulis* (Fig. 4B; hybrid-*P. californiarum* contrast $Z = 9.9 < Z_{crit,0.05} = 10.8$; hybrid-*P. edulis* contrast; $Z = 12.7 > Z_{crit,0.05} = 12.0$). Thus, within the hybrid zone where site effects are largely eliminated, these data most closely support the dominance hypothesis where one parental species is more susceptible to a herbivore, but not the other. Although these analyses show significant differences between categories and support the dominance hypothesis, we emphasize that the magnitude of the differences are not great and question their biological relevance. Nevertheless, this pattern differs from that of the stem-boring moth and suggests that many herbi-

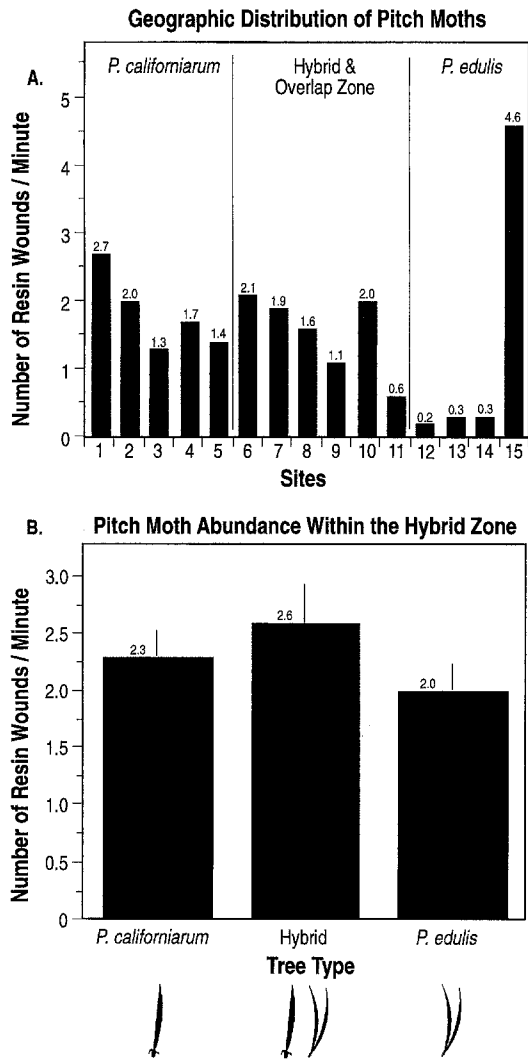


Fig. 4 **A** Although pitch moths do not appear to vary in abundance across sites, analyses argue that they were significantly more abundant at pure *P. californiarum* sites than either hybrid or pure *P. edulis* sites (see text). Abundance was recorded as the mean number of pitch moth wounds counted per minute across the 15-site transect. **B** Within the hybrid zone, pitch moth abundance was greatest on *P. californiarum* and hybrid trees compared to *P. edulis* trees as shown by the mean number of pitch moth wounds counted per minute. Bars indicate the mean ± 1 standard error of the mean

vore species should be examined before generalities of hybrid/herbivore interactions can be made.

Scale insect abundance across sites 1990–1991

The abundance of scales varied significantly among sites (Fig. 5; $T = 69.36$, $P < 0.0001$, $df = 14$); scales were found only on trees at hybrid sites 8, 9 and 10. Our observation that scales could only be found within the hybrid zone suggests that hybrid zones may be important refugia for some insects during periods of population decline (note that scales went extinct in 1992 even at the hybrid sites). Although these results are consistent with both the hy-

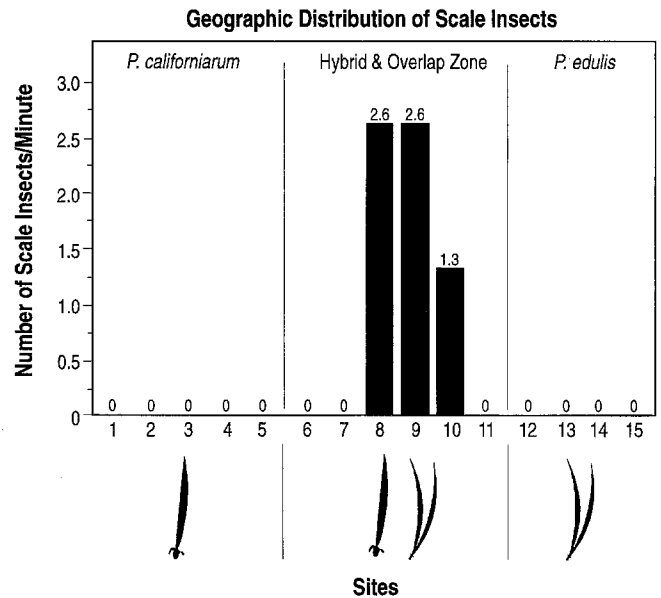


Fig. 5 Scale insects were only found at three sites at the center of the hybrid zone. Mean number of scale insects counted per minute across the 15-site transect are shown

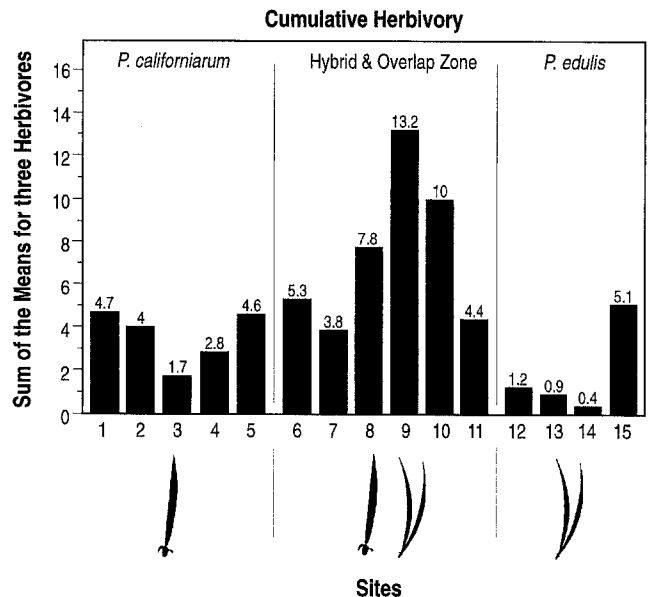


Fig. 6 A histogram of the cumulative number of all three herbivores across the 15-site transect shows that more herbivores are found in the hybrid zone compared to pure zones

brid susceptibility and stress hypotheses, we have no data on how scales are distributed among pure and hybrid trees within the hybrid zone and thus cannot discriminate between these two hypotheses.

Cumulative herbivore abundance

The cumulative abundance of the three herbivores examined in this study varied significantly among sites

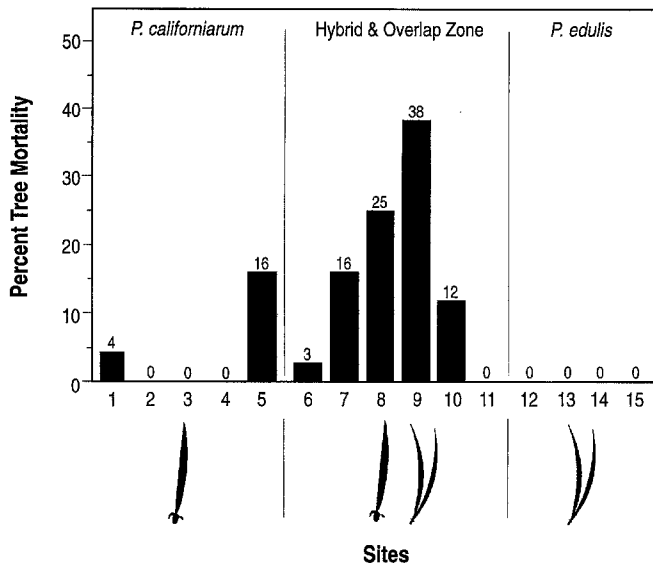


Fig. 7 Percent tree mortality varied significantly across sites and was highest at hybrid sites (χ^2 goodness of fit, $X^2 = 57.4$, $df = 14$, $P < 0.05$)

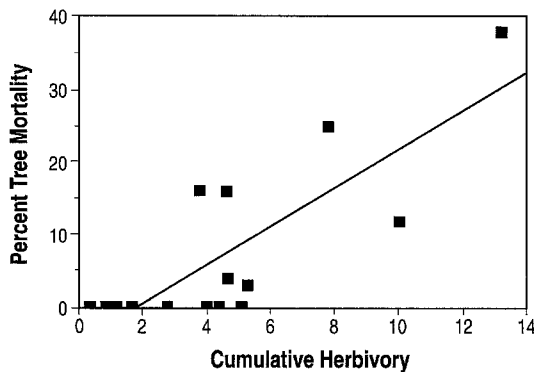


Fig. 8 There is a significant positive relationship between the cumulative level of herbivory and the level of tree mortality across sites. Each point represents a site ($y = -4.88 + 2.68x$, $r^2 = 64.6\%$, $P < 0.001$)

(Fig. 6; $T = 16.2$, $df = 14$, $P < 0.001$) and trees in the hybrid zone supported significantly more herbivores than trees at either pure zone (hybrid-*P. californiarum* contrast; $Z = 9.03 > Z_{crit.,0.05} = 3.78$, hybrid-*P. edulis* contrast; $Z = 14.12 > Z_{crit.,0.05} = 3.69$).

Although trees in the hybrid zone generally supported greater numbers of herbivores than trees in the pure zone, there were no significant differences in cumulative herbivore loads among pure and hybrid tree types within the hybrid zone ($T = 1.38$, $P = 0.41$, $df = 2$). This lack of pattern within the hybrid zone is not surprising as all three herbivores exhibited different distributions and different mechanisms are implicated in affecting the abundance of each.

Tree mortality

Tree mortality varied significantly across sites where the greatest mortality was found at hybrid sites 8 and 9

(Fig. 7). Here, there was a maximum of 38% mortality at site 9 and an average of 15.7% across the hybrid sites. The average levels of mortality at the pure sites were 4% and 0% at *P. californiarum* and *P. edulis* sites, respectively.

Cumulative herbivory and tree mortality

The level of cumulative herbivory was significantly correlated with tree mortality across the 15 sites (Fig. 8). While these results imply a causal relationship between herbivory and tree mortality, we caution that other factors such as greater environmental stress at the boundary of a species' distribution may contribute to this pattern.

Discussion

In this system, the hybrid susceptibility hypothesis was supported in three of seven analyses, and both the dominance and stress hypotheses were supported in two of seven analyses. Of our five initial hypotheses, three appear to be involved in affecting the distributions of the herbivores we examined. The abundance of stem-boring moths supported the hybrid susceptibility (twice) and stress (once) hypotheses, pitch moth abundance supported the dominance hypothesis (twice) while scale insect distribution supported both the stress (once) and hybrid susceptibility (once) hypotheses. These results agree with other recent studies (Fritz et al. 1994, *Salix* spp.; Whitham et al. 1994, *Eucalyptus* spp.; Morrow et al. 1994, *Eucalyptus* spp.), showing that diverse taxa, members of the same guild and even closely related species respond differently to hybridization by their host plants.

Variation in hybrid susceptibility

Variation in resistance among hybrid classes can be great and may even exceed the variation in host use that has been observed between species. In their studies of a natural cottonwood hybrid swarm Paige et al. (1990) and Paige and Capman (1993) found that F1 and backcross-1 type hybrids were nearly totally resistant to the gall aphid, *Pemphigus betae*, while complex backcrosses ranged from highly resistant to highly susceptible. In another system, Morrow et al. (1994) found that 33 species of galling insects on eucalypts were more likely to vary in abundance among hybrid classes than they were between pure species. Variation in hybrid resistance and susceptibility is also supported by agricultural studies (Maxwell and Jennings 1980).

In some systems, herbivore preferences for a specific host species results in predictable patterns of hybrid class use. For example, in their analyses of 40 insect and fungal taxa in a hybrid eucalypt swam in Australia, Whitham et al. (1994) found that 73% were significantly more abundant in the hybrid zone than in pure zones. Within the hybrid swarm herbivores and fungi that spe-

cialized on *Eucalyptus risdonii* were most abundant on backcross hybrids with *E. risdonii*, while species that specialized on *E. amygdalina* were most abundant on backcross hybrids with *E. amygdalina*. Thus, for specialist taxa, the most used hybrid class was the one phenotypically most similar to the preferred host species (i.e., the phenotypic affinity hypothesis; Whitham et al. 1994). This pattern has been observed in two other eucalypt hybrid swarms (Morrow et al. 1994). Such host specificity also suggests that insects might be used as traits in plant systematics to discriminate between difficult taxa (Floate and Whitham 1995).

In contrast to the above studies, we did not observe significant differences in herbivore abundance among pinyon hybrid classes. The use of fewer taxonomic characters may have affected our ability to discriminate between hybrid classes and suggests that molecular techniques might be used to gain greater resolving power. For example, using relatively few markers, Paige and Capman (1993) found that some cottonwoods that appeared to be pure based upon morphology were actually hybrid. Floate et al. (1994), however, argued that to gain the necessary resolving power, 50 or more genetic markers (Keim et al. 1992) were needed to separate hybrid classes, and that failure to do this may render genetic studies less accurate than studies based upon morphology.

Stress and genetic impacts on herbivory

Several studies have found plants and animals to suffer greater environmental stress at the edges of their range (Bunce et al. 1979; Brussard 1984; Parsons 1991) and at least one study has found a conifer to suffer greater herbivory at the edge of its range (McClure 1985). In this pinyon pine system, the hybrid sites are at the edges of the ranges of both *P. edulis* and *P. californiarum*. Thus, increased susceptibility to herbivory due to stress (e.g., White 1969; Waring and Cobb 1992) could in part contribute to our observed patterns of increased insect abundance (Fig. 6) and higher tree mortality in the hybrid zone (Fig. 7). This possibility is likely considering the experiments of N. Cobb, S. Mopper, C. Gehring, M. Caouette, K. Christensen, T.G. Whitham (unpublished work). In comparisons of treatment and control trees growing in the stressful cinder soils of Sunset Crater, they found that trees receiving supplemental water and fertilizer produced significantly more resin defenses and suffered significantly reduced herbivory from the stem-boring moth *Dioryctria albobittella*.

In addition to the effects of stress on plant susceptibility to stem-borers, two lines of evidences support a genetic involvement as well. First, using many of the same trees as N. Cobb, S. Mopper, C. Gehring, M. Caouette, K. Christensen, T.G. Whitham (unpublished work), Mopper et al. (1991) found that trees resistant and susceptible to *D. albobittella* differed significantly in both allelic frequencies and heterozygosity. Second, in our present study, when stress is largely eliminated as a fac-

tor (i.e., comparisons within a site should make stress uniform), we found that *D. albobittella* was significantly more concentrated on hybrids than on either pinyon species. Because these combined studies show that both environmental stress and genetics affect herbivory on *Pinus edulis* in northern Arizona, we also expect this to be true within the hybrid zone where both genetic variation and environmental stress are likely to be high.

Although these separate studies show that stress and genetics affect herbivory on pinyon pine, how these two factors potentially interact has not yet been examined. Another study, however, found that stress and genetics interact to affect pinyon growth. Cobb et al. (1994) examined the growth rates of three different pinyon genotypes for the glycerate dehydrogenase locus, an enzyme implicated in drought tolerance. In the water and nutrient stressed cinder soils of Sunset Crater, the (SS) homozygotes grew at twice the rate of the (FF) homozygotes, and the (FS) heterozygotes were intermediate, but not significantly different from the (FF) homozygotes. Furthermore, the slowest growing genotypes were significantly less abundant in mature trees compared to juvenile trees suggesting that selection had favored the (SS) homozygotes in this stressful environment.

While the causes of pinyon tree mortality have not yet been experimentally determined, the observed high levels of mortality of mature trees in the hybrid zone (Figs. 7 and 8) suggest that hybrid zones are important arenas where the genetic template of plant populations are tested against both environmental and biotic agents. Further studies of such natural systems may prove valuable in studying the ecology and evolution of plant-herbivore interactions, and have applied value as hybrids are used extensively in agriculture, forest plantations and in ornamental plantings.

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References

- Aguilar JM, Boecklen WJ (1992) Patterns of herbivory in the *Quercus grisea* X *Quercus gambelii* species complex. *Oikos* 64:498-504
- Barker JF (1990) Sunflower trichome defenses avoided by a sunflower stem weevil, *Cylindrocopturus adspersus* LeConte (Coleoptera: Curculionidae). *J Kansas Ent Soc* 63:638-641
- Bailey DK (1987) A study of *Pinus* subsection cembroides I: the single-needled pinyons of the Californias and Great Basin. *Notes R Bot Gard Edinb* 44:275-310
- Boecklen WJ, Spellenberg R (1990) Structure of herbivore communities in two oak (*Quercus* spp.) hybrid zones. *Oecologia* 85:92-100
- Brussard PF (1984) Geographic patterns and environmental gradients: the central-marginal model in *Drosophila* revisited. *Annu Rev Ecol Syst* 15:25-64
- Bunce JA, Chabot BF, Miller LN (1979) Role of annual leaf carbon balance in the distribution of plant species along an elevational gradient. *Bot Gaz* 140:288-294

- Cain R, Cota J, Ward C (1990) Conifer pests in New Mexico. New Mexico State University Cooperative Extension Service, Las Cruces
- Cobb N, Mitton JB, Whitham TG (1994) Genetic variation associated with chronic water and nutrient stress in pinyon pine. *Am J Bot* 81:936–940
- Cranshaw W, Leatherman D, Kondratieff B (1993) Insects that feed on Colorado trees and shrubs (Bulletin 506A). Colorado State University Cooperative Extension, Ft. Collins
- Conover WJ (1980) Practical nonparametric statistics. 2nd edn. Wiley, New York
- Daniel WW (1990) Applied nonparametric statistics. 2nd edn. PWS-Kent, Boston
- Drake DW (1981a) Reproductive success of two *Eucalyptus* hybrid populations I. Generalized seed output model and comparison of fruit parameters. *Aust J Bot* 29:25–35
- Drake DW (1981b) Reproductive success of two *Eucalyptus* hybrid populations II. Comparison of predispersal seed parameters. *Aust J Bot* 29:37–48
- Ericson L, Burdon JJ, Wennstrom A (1993) Interspecific host hybrids and phalacrid beetles implicated in the local survival of smut pathogens. *Oikos* 68:393–400
- Floate K, Whitham TG (1993) The “hybrid-bridge” hypothesis: host shifting via plant hybrid swarms. *Am Nat* 141:651–662
- Floate K, Whitham TG (1994) Aphid-ant interaction reduces chrysomelid herbivory in a cottonwood hybrid zone. *Oecologia* 97:215–221
- Floate K, Whitham TG (1995) Insects as traits in plant systematics: their use in discriminating between hybrid cottonwoods. *Can J Bot* (in press)
- Floate KD, Kearsley MJC, Whitham TG (1993) Elevated herbivory in plant hybrid zones: *Chrysomela confluens*, *Populus* and phenological sinks. *Ecology* 74:2025–2034
- Floate KD, Whitham TG, Keim P (1994) Morphological versus genetic markers in classifying hybrid plants. *Evolution* (in press)
- Fritz RS, Nichols-Orians CM, Brunsfeld SJ (1994) Interspecific hybridization of plants and resistance to herbivores: hypotheses, genetics, and variable responses in a diverse herbivore community. *Oecologia* 97:106–117
- Gallun RL, Khush GS (1980) Genetic factors affecting expression and stability of resistance. In: Maxwell FG, Jennings PR (eds) *Breeding plants resistant to insects*. Wiley, New York, pp 63–85
- Grant V (1981) *Plant speciation*, 2nd edn. Columbia University Press, New York
- Jennrich R, Sampson P (1983) Stepwise discriminant analysis. In: Dixon WJ (ed) *BMDP statistical software*. University of California Press, Berkeley, pp 519–537
- Keim P, Paige KN, Whitham TG, Lark KG (1989) Genetic analysis of an interspecific hybrid swarm of *Populus*: occurrence of unidirectional introgression. *Genetics* 123:557–565
- Keim P, Beavis W, Schupp J, Freestone R (1992) Evaluation of soybean RFLP marker diversity in adapted germ plasm. *Theor Appl Genet* 85:205–212
- Lanner RM (1974) Natural hybridization between *Pinus edulis* and *Pinus monophylla* in the American Southwest. *Silv Genet* 23:108–116
- Lanner RM, Phillips AM (1992) Natural hybridization and introgression of pinyon pines in northwestern Arizona. *Int J Plant Sci* 153:250–257
- Little EL (1968) Two new pinyon varieties from Arizona. *Phytologia* 17:329–341
- Malusa J (1992) Phylogeny and biogeography of the pinyon pines (*Pinus* subsect. *Cembroides*). *Syst Bot* 17:42–66
- Manley AM, Fowler DP (1969) Spruce budworm defoliation in relation to introgression in red and black spruce. *For Sci* 15:365–366
- Martinsen GD, Whitham TG (1994) More birds nest in hybrid cottonwoods. *Wilson Bulletin* 106:474–481
- Maxwell FG, Jennings PR (1980) *Breeding plants resistant to insects*. Wiley, New York
- McClure MS (1985) Patterns of abundance, survivorship, and fecundity of *Nuculaspis tsugae* (Homoptera: Diaspididae) on *Tsuga* species in Japan in relation to elevation. *Environ Entomol* 14:413–415
- Mitchell RG, Johnson NE, Wright KH (1974) Susceptibility of 10 spruce species and hybrids to the white pine weevil (=sitka spruce weevil) in the pacific northwest USDA For Serv Res Note PNW-225
- Mopper S, Mitton JB, Whitham TG, Cobb NS, Christensen KM (1991) Genetic differentiation and heterozygosity in pinyon pine associated with resistance to herbivory and environmental stress. *Evolution* 45:989–999
- Moran NA, Whitham TG (1988) Evolutionary reduction of complex life cycles: loss of host alternation in *Pemphigus* (Homoptera: Aphididae). *Evolution* 42:717–728
- Moran NA (1991) Phenotype fixation and genotypic diversity in the complex life cycle of the aphid *Pemphigus betae*. *Evolution* 45:957–970
- Morrow PA, Whitham TG, Potts BM, Ladiges P, Ashton DH, Williams JB (1994) Gall-forming insects concentrate on hybrid phenotypes of *Eucalyptus*. In: Baranchikov YN, Price PW, Mattson WJ Jr (eds) *Gall-forming insects: ecology, physiology and evolution*. USDA For Serv North Central For Exp St, GTRNC-174, pp 121–134
- Paige KN, Capman WC (1993) The effects of host-plant genotype, hybridization, and environment on gall-aphid attack and survival in cottonwood: the importance of genetic studies and the utility of RFLPs. *Evolution* 47:36–45
- Paige KN, Keim P, Whitham TG, Lark KG (1990) The use of restriction fragment length polymorphisms to study the ecology and evolutionary biology of aphid-plant interactions. In: Campbell RK, Eikenbary RD (eds) *Aphid-plant genotype interactions*. Elsevier, Amsterdam, pp 69–87
- Parsons PA (1991) Evolutionary rates: stress and species boundaries. *Annu Rev Ecol Syst* 22:1–18
- Price PW (1991) The plant vigor hypothesis and herbivore attack. *Oikos* 62:244–251
- Riesberg LH, Wendel JF (1993) Introgression and its consequences in plants. In: Harrison RG (ed) *Hybrid zones and the evolutionary process*. Oxford University Press, New York, pp 70–109
- Stace CA (1987) Hybridization and the plant species. In: Urbanska KM (ed) *Differentiation patterns in higher plants*. Academic Press, New York, pp 115–127
- Tingey WM, Singh SR (1980) Environmental factors influencing the magnitude and expression of resistance. In: Maxwell FG, Jennings PR (eds) *Breeding plants resistant to insects*. Wiley, New York, pp 87–113
- Waring GL, Cobb NS (1992) The impact of plant stress on herbivore population dynamics. In: Bernays EA (ed) *Insect-plant interactions*. CRC Press, Boca Raton, FL, pp 167–226
- White TCR (1969) An index to measure weather-induced stress of trees associated with outbreaks of psyllids in Australia. *Ecology* 50:905–909
- Whitham TG (1989) Plant hybrid zones as sinks for pests. *Science* 244:1490–1493
- Whitham TG, Morrow PA, Potts BM (1991) Conservation of hybrid plants. *Science* 254:779–780
- Whitham TG, Morrow PA, Potts BM (1994) Plant hybrid zones as centers of biodiversity: the herbivore community of two endemic Tasmanian eucalypts. *Oecologia* 97:481–490