

Impact of an invasive oak gall wasp on a native butterfly: a test of plant-mediated competition

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Abstract. Phytophagous insects commonly interact through shared host plants. These interactions, however, do not occur in accordance with traditional paradigms of competition, and competition in phytophagous insects is still being defined. It remains unclear, for example, if particular guilds of insects are superior competitors or important players in structuring insect communities. Gall-forming insects are likely candidates for such superior competitors because of their ability to manipulate host plants, but their role as competitors is understudied. We investigate the effect of invasive populations of an oak gall wasp, *Neuroterus saltatorius*, on a native specialist butterfly, *Erynnis propertius*, as mediated by their shared host plant, *Quercus garryana*. This gall wasp occurs at high densities in its introduced range, where we stocked enclosures with caterpillars on trees that varied in gall wasp density. Biomass production of butterflies was lower in enclosures on high-density than on low-density trees because overwintering caterpillars were smaller, and fewer of them enclosed into adults the following spring. To see if the gall wasp induced changes in foliar quality, we measured host plant quality before and after gall induction on 30 trees each at two sites. We found a positive relationship between gall wasp density and the percentage change in foliar C:N, a negative relationship between gall wasp density and the percentage change in foliar water at one site, and no relationship between the percentage change in protein-binding capacity (i.e., phenolics) and gall-wasp density. Additionally, there was a negative relationship between foliar quality and butterfly performance. Our results provide evidence for a plant-mediated impact of an invasive oak gall wasp on a native butterfly and suggest that gall wasps could act as superior competitors, especially when they occur at high densities.

Key words: *cynipids*; *Erynnis propertius*; *gall-forming insect*; *induced defenses*; *interspecific competition*; *invasive species*; *Neuroterus saltatorius*; *plant-mediated interactions*; *plant–insect interactions*; *Quercus garryana*; *range expansion*.

INTRODUCTION

The importance of competition between phytophagous insects has a controversial history (Denno et al. 1995, Kaplan and Denno 2007; e.g., Davidson and Andrewartha 1948, Lawton and Strong 1981, Schoener 1983). On one hand, the world is green (Hairston et al. 1960), many phytophagous insects coexist, and they often lack aggressive behaviors (e.g., Ross 1957, Rathcke 1976), suggesting that food is abundant and competition between phytophagous insects is a rare force in nature. More recently, however, researchers have suggested that this view is over-simplified, and competitive interactions between phytophagous insects are more common than previously thought (e.g., Faeth 1986, Denno et al. 2000, Van Zandt and Agrawal 2004). These interactions are frequently mediated by a shared host plant and do not occur by exploitative competition. Interactions are often temporally and spatially separated and can occur between nonrelated insects and between different feeding guilds (Damman 1993, Denno et al.

1995, Denno and Kaplan 2007, Kaplan and Denno 2007). Observations of competition in insects are redefining traditional theories of competition, and continuing to do so is an important focus for population and community ecology.

While indirect interactions between phytophagous insects are often asymmetrical (Denno et al. 1995, Kaplan and Denno 2007), it remains unclear if certain guilds of insects are superior competitors, and therefore play large roles in shaping insect communities. Herbivores feeding in the spring, for example, have been shown to influence insect community structure of herbivores that occur later in the season (Faeth 1986, Wold and Marquis 1997, Lill and Marquis 2003, Van Zandt and Agrawal 2004). Attention also has been paid to sap-sucking insects due to their ability to affect the source–sink dynamics of assimilates and nutrients (Larson and Whitham 1991, Inbar et al. 1995, Denno et al. 2000). Gall-formers also are good candidates for competitive superiority because they can manipulate plant metabolism and create galls that act as sinks for assimilates and nutrients (Price et al. 1987, Stone and Schönrogge 2003). However, their role as competitors is understudied (Nykänen and Koricheva 2004, Kaplan

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and Denno 2007; except see, Inbar et al. 1995, Fritz et al. 1986, Fritz and Price 1990, Foss and Rieske 2004).

Gall-formers include both sap-sucking insects (e.g., aphids, whiteflies) and chewers (e.g., gall midges, sawflies, oak wasps). Previous reviews have found that competitive interactions were more frequent with sucking (77%) than with chewing (51%) herbivores (Denno et al. 1995). A recent meta-analysis, however, found no differences in the prevalence of competition between these two groups (Kaplan and Denno 2007). These conflicting results likely occur because there are a limited number of studies in particular subguilds. For example, in these reviews, gall-forming chewers were lumped with free-living chewers. Yet these groups function differently in the way that they induce plant responses (e.g., Ollerstam and Larsson 2003) and they could therefore differ in their competitive effects.

There are ~3000 species of galling-forming insects in five orders (Stone et al. 2002). Cynipid wasps (Hymenoptera) are the second most diverse family after the Cecidomyiidae midges, and the majority of cynipids are obligate parasites on oaks (Stone et al. 2002). There are ~1000 species of oak gall wasps globally, with the majority occurring in the Nearctic (Cornell 1983, Stone et al. 2002). Classical and current studies of the role of host plant quality in shaping insect communities have been conducted in oak systems (e.g., Feeny 1970, Faeth 1986, Wold and Marquis 1997, Lill and Marquis 2003, Forkner et al. 2004), but the role of cynipids in influencing oak insect communities is largely unknown. A few studies have focused on intraspecific interactions between cynipid species (Hartley and Lawton 1992, Hartley 1998), and fewer on paired interspecific and interguild interactions (Foss and Rieske 2004), with equivocal results as to the influence of gall-formers as competitors.

Galls formed by cynipid wasps are the most sophisticated of all gall-formers (Cornell 1983). Their galls are induced by the insect and contain several layers of tissue including a protective outer layer and an inner layer of nutritive tissue (Stone and Schönrogge 2003). Galls can act as nutrient sinks, sequestering nutrients from surrounding leaf tissue to the nutritive tissue of the gall (e.g., Paclt and Hassler 1967, McCrea and Abrahamson 1985, Price et al. 1987, Bronner 1992; except see Hartley 1998). Gall-formers can also increase the production of secondary chemicals, such as tannins, actively concentrating them in the outer protective layer of the gall while keeping them out of the nutritive tissue (Hartley 1998, Nyman and Julkunen-Tiitto 2000, Allison and Schultz 2005). Little is known about how the manipulation of nutrients and phenols by gall-formers affects nearby foliage and thus other herbivores. Given the prevalence and diversity of cynipids and their ability to affect host plant quality, their role in influencing insect communities warrants further exploration.

We focused this study on *Neuroterus saltatorius* (Edwards), a cynipid foliar gall wasp that recently

expanded its range. Like many species that establish in a new area, *N. saltatorius* occurs in higher abundance than it does in its native range (Smith 1995, Duncan 1997; K. M. Prior, *unpublished data*). This high density provides an opportunity for particularly strong effects on native community members and the opportunity to investigate the effect of density on competitive interactions. For example, outbreaking phytophagous insects have been found to have large effects on host plant quality (e.g., Schultz and Baldwin 1982). These large effects could have detrimental impacts on the fitness of native community members that are not adapted to large changes in host plant quality. As climate change and humans continue to move species around the globe, it is increasingly important that we understand the role of novel community members on insect communities.

This study provides an example of an interspecific plant-mediated interaction in an understudied guild that has the potential for influencing oak insect communities. In particular, we investigate if invasive populations of *N. saltatorius* affect the performance of a native specialist butterfly, *Erynnis propertius* (Scudder and Burgess) (Lepidoptera: HesperIIDae), as mediated through interactions with their shared host plant, *Quercus garryana* Dougl. ex Hook (Fagaceae). Specifically, we address if gall wasp density affects butterfly performance; if this gall wasp causes induced changes in its host plant; and if host plant quality plays a role in affecting butterfly performance. This study contributes to a young but growing literature that is redefining theories of competition for phytophagous insects. Examples from understudied guilds such as gall-formers are integral to the formation of a new competition paradigm.

METHODS

Study system

The native range of *N. saltatorius* is from north Texas to mainland Washington (Duncan 1997; see map in Appendix A). Its hosts are a variety of white oak species, with *Q. garryana* being its only host in northern Oregon, Washington, and British Columbia (Duncan 1997). *Neuroterus saltatorius* was first recorded on Vancouver Island, British Columbia in 1982 just north of Victoria (Smith 1995, Duncan 1997). Since then, it has been found as far north as the range limit of *Q. garryana* (K. M. Prior, *personal observation*). *Neuroterus saltatorius* occurs in higher densities in its introduced range, being 1.6 times higher in density at sites in British Columbia (average over two years) compared to sites in Southern Puget Sound, Washington (K. M. Prior, *unpublished data*).

Neuroterus saltatorius has two generations: a sexual (gamic) generation that forms cluster galls of males or females on leaves in the spring, and an asexual (agamic) generation that forms single spherical galls (1 mm) on the underside of leaves starting in mid-June. Agamic galls develop through the summer, drop from leaves, and overwinter in the leaf litter (Smith 1995). The



PLATE 1. (Left) *Neuroterus saltatorius* breaking out on its host plant, *Quercus garryana*, in its introduced range on Vancouver Island, British Columbia. (Right) A caterpillar of the specialist butterfly, *Erynnis propertius*, which develops on *Q. garryana* leaves contemporaneously with *N. saltatorius* galls. Photo credits: K. M. Prior.

agamic generation occurs at higher density than the gamic generation (Smith 1995). Chlorotic leaf spotting around the site of gall attachment occurs in the agamic generation, and necrosis of continuous portions of the leaf occurs when gall density is high (i.e., as in British Columbia) (see Appendix B). This necrosis is likely under control by the gall wasp (Stone et al. 2002), but it may also be an induced response by the plant, as has been implicated for other gall-formers (Fernandes 1990, Ollerstam and Larsson 2003).

Erynnis propertius is a specialist butterfly on *Quercus* spp. and is threatened in British Columbia. It occurs on several oak species in the southern portion of its range, but only on *Q. garryana* in British Columbia (Pelini et al., *in press*). Its flight occurs from April through June, with peak flight in May. Caterpillars develop in leaf-folds between May and September. In the fall, *E. propertius* enters a quiescent state in its sixth instar where it overwinters in a rolled up leaf and pupates and ecloses the following spring (Prior et al. 2009). Caterpillars of *E. propertius* and agamic galls of *N. saltatorius* develop contemporaneously on leaves from early June through the end of August (Appendix B), and we predict that agamic galls affect the foliar quality of leaves that *E. propertius* caterpillars forage on throughout their development (see Plate 1).

Assessing the relationship between gall wasp density and butterfly performance

In 2007, we conducted an experiment at a 6-ha site in British Columbia where there was substantial variation in *N. saltatorius* density among trees (S1; in Appendix A). Gall wasp density is patchy among trees (e.g., Eliason and Potter 2001, Egan and Ott 2007), and we used this variation to test the effect of gall wasp density on butterfly performance. The amount of foliar damage caused by the gall wasp is a good surrogate of gall wasp

density (Appendix C). In mid-June 2007, we chose nine trees at S1 and assigned each to one of three density categories by assessing gall wasp damage on a large number of leaves at eye level. If the majority (i.e., over half) of the leaves had 0–10 galls, or less than ~25% gall-damaged leaf area, we assigned the tree to the “low” density category. If the majority of the leaves had a lot of galls on leaves, but damage was spotty (e.g., ~50% gall-damaged leaf area), we assigned the tree to the “moderate” density category. If the majority of the leaves had continuous portions of gall-damaged leaf area (e.g., ~75%), we assigned the tree to the “high” density category (see Appendix C for images). We performed photo-imaging analysis on a subset of leaves from these trees in late July to verify the assignment of these trees to density categories and to quantify leaf damage as a continuous variable. We placed four enclosures over four different branches on each tree ($n = 9$). Enclosures were 41 cm in diameter, 60 cm long, and covered with fine Dacron mesh (7250C [Bioquip, Rancho Dominguez, California, USA]). Competitors and predators were repeatedly removed from enclosures.

Females of *E. propertius* were caught on the wing at S2, where this species is abundant (Hellmann et al. 2008, Prior et al. 2009; Appendix A). Eggs were collected from adults, and caterpillars were reared on potted oaks in a greenhouse until their second instar. We then stocked six to seven caterpillars in three of the enclosures on each tree, with one enclosure per tree being left empty (i.e., control) to assess gall damage and foliar quality (see *Assessing the relationship between gall wasp density and host plant quality*). Each enclosure contained enough leaves so that the quantity of leaves was not limiting to caterpillars (as in Hellmann et al. 2008, Pelini et al. 2009). There were no differences in the initial size of caterpillars among trees (one-way ANOVA: $F_{8,18} = 0.83$; $P = 0.585$). We used a nested design, of multiple

enclosures per tree, as opposed to replicating large numbers of trees, because host plant quality can differ among trees (e.g., Forkner et al. 2004). The power of our nested design and replication was adequate as it detected differences among density categories (see *Results*).

The length and width of caterpillars were repeatedly measured throughout their growing season, and volume was calculated as the volume of a cylinder. This is a good surrogate for body mass (Hellmann et al. 2008, Pelini et al. 2009; Appendix D). In October 2007, we measured caterpillars in their overwintering stage (i.e., sixth instar) (Prior et al. 2009). We allowed caterpillars to overwinter in a greenhouse with conditions set to mimic southern Vancouver Island, British Columbia, using the average maximum and minimum temperatures and photoperiods calculated over the last five years from a local weather station (48°39' N, 123°26' W). In the spring of 2008, caterpillars were checked daily, and as each individual pupated, volume measurements were taken and individuals were weighed. Adults were dried in a drying oven at 60°C for 48 hours, and volume and dry mass without wings was measured. We measured butterfly performance as butterfly production, defined as the change in biomass in each enclosure from the second instar to adult. We calculated this metric by dividing the sum of adult dry mass by the sum of the volume of the initial caterpillars (milligrams per cubic millimeter) in each enclosure. This metric captures both growth and survivorship of individuals (Hellmann et al. 2008; see Appendix D).

At the end of July, when damage is most apparent on leaves, we took pictures of 10 randomly chosen leaves within the three butterfly enclosures on each tree. We used the photo-imaging analysis program APS Assess to quantify *N. saltatorius* leaf damage (Lamari 2002; see Appendix C). We also randomly sampled 10 leaves from control enclosures on each tree to quantify damage and assess foliar quality (as described in the next section).

Assessing the relationship between gall wasp density and host plant quality

To examine the effect of *N. saltatorius* on host plant quality, we assessed the relationship between gall wasp density and foliar quality. In 2007, we collected leaves from 30 trees spread throughout S1, and 30 trees at an additional 6-ha site (S3; see Appendix A) in late July. We randomly collected 10 leaves from each tree and pooled these leaves to represent damage and quality at the tree level. In 2008, we collected leaves before and after agamic gall induction to see if gall wasp density depends on constitutive foliar quality, and if gall wasps induce changes in foliar quality. “Early-season” collections were conducted in mid-May before gamic female wasps had laid eggs. “Late-season” collections occurred in late July after agamic galls had been developing for ~1.5 months. The percentage change in water, C:N, and

total phenolics was calculated as the difference between late and early-season samples, divided by early-season samples, for each tree. An increase or decrease in the percentage change in quality with increasing gall wasp density would indicate that the gall wasp induces changes in foliar quality.

To assess foliar quality, we calculated the wet and dry mass of each tree sample to determine water content. In 2007, leaves were oven dried at 60°C for 48 hours. In 2008, leaves were shipped on dry ice, stored at –80°C, and freeze-dried for 48 hours. Photographs were taken from the late-season samples, and percentage damage was calculated using APS Assess, as described in *Assessing the relationship between gall wasp density and butterfly performance*. We then removed the petiole, midvein, and damaged area of each dried leaf, because we assumed that caterpillars ate only green leaf material. Samples were ground in a Wiley mill (Thomas Scientific, Swedesboro, New Jersey, USA) (mesh size 40; 0.425 mm). Percentage carbon and nitrogen were determined by the use of an elemental analyzer (ESC 4010 CHNSO Analyzer [Costech Analytical Technologies Incorporated, Valencia, California, USA]). Three analytical replicates per tree sample were conducted. The protein-binding capacity, which is a measure of foliar phenolics, of each tree sample was measured using the radial diffusion method (Hagerman 1987, e.g., Wold and Marquis 1997, Foss and Rieske 2004). Each sample was run three times on three different plates (20- μ L aliquots/well) and the average squared ring diameter was calculated. Tannic acid standard was used (403040 [Sigma-Aldrich Incorporated, St. Louis, Missouri, USA]) to determine the equivalent tannic acid concentration (TAE; in milligrams). Protein binding capacity was only measured in 2008 leaves.

Statistical analyses

We conducted a nested ANOVA on percent damage in the experiment among density categories and among trees within density categories. We also performed a nested ANOVA on butterfly production where production was arcsine square-root transformed. Since percent damage (i.e., density) in each enclosure was quantified, damage also could be treated as a continuous variable, and a linear regression between percent damage and production was conducted using the mean percent damage and production of enclosures on each tree.

Separate linear regressions were conducted between percent damage and each foliar quality component, percentage of water, C:N, and TAE (mg) from the 30 trees collected at each site. We made transformations on variables when necessary. First, to see if *N. saltatorius* responds to differences in constitutive foliar quality, we regressed foliar quality from early-season leaves on percent damage from late-season leaves. Second, to see if *N. saltatorius* induces changes in foliar quality, we regressed percent damage on the percent change in foliar

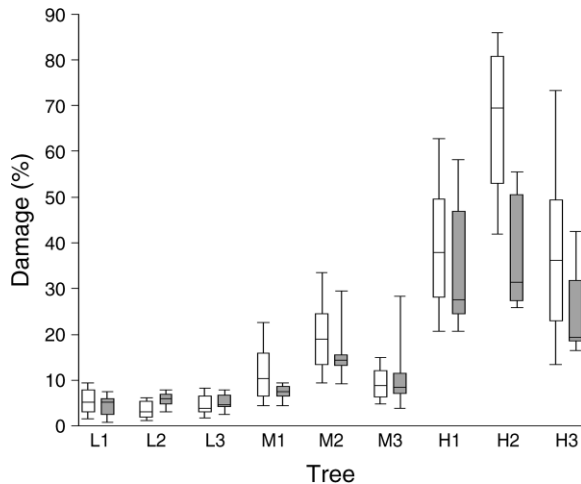


FIG. 1. Box plot of damage caused by *Neuroterus saltatorius* on experimental trees that were grouped into low (L), moderate (M), and high (H) gall-density categories. White bars represent the percent damage of leaves in enclosures with caterpillars. Leaves from the three enclosures per tree are pooled. Gray bars represent the percent damage of leaves in control enclosures on the same tree. Percent damage was digitally assessed from 7–10 leaves per enclosure. Top and bottom bars represent the 90th and 10th percentile, respectively. The top and bottom of the boxes represent the 75th and 25th percentile, respectively, and the horizontal bar within the box represents the median.

quality. We also conducted regressions between percent damage and foliar quality in late-season leaves.

To determine if host plant quality was related to butterfly performance, we conducted a principal components analysis (PCA) to collapse percent damage, C:N, and water content into a single measure of foliar quality using leaves collected from the control enclosures. We did not include TAE, because it was not measured in these leaves. A linear regression was conducted between PC1 (quality) and butterfly production ($n = 9$). Finally, linear regressions were also conducted between percent damage and percentages of nitrogen and carbon, separately, to see how damage affected these elements differently.

All statistical tests were carried out with SYSTAT 12 (Systat Software, Chicago, Illinois, USA). ANOVAs were carried out using General Linear Model, Type III sums of squares, and PCA was carried out using Factor Analysis.

RESULTS

A nested ANOVA on percent damage in the experimental trees revealed larger differences among density categories ($F_{2,6} = 35.37$, $P < 0.0001$) than among trees within categories ($F_{6,18} = 5.26$, $P = 0.003$; Fig. 1). A nested ANOVA of butterfly production revealed differences among density categories ($F_{2,6} = 4.91$; $P = 0.020$) with lower production in the high- vs. low-density trees ($P = 0.024$, Tukey's post hoc test; Fig. 2). However, we found no differences among trees within density

categories ($F_{6,18} = 0.96$; $P = 0.478$). Differences in production were driven by differences in overwintering caterpillar volume ($F_{2,6} = 6.93$, $P = 0.029$) with smaller caterpillars in the high- vs. low-density trees ($P = 0.008$; Tukey's post hoc test; see Appendix D for details). There was also a marginal reduction in survivorship to adulthood ($F_{2,6} = 4.55$, $P = 0.063$; Appendix D). In addition, there was a negative relationship between percent damage and butterfly production ($R^2 = 0.88$, $P < 0.0001$; $n = 9$; Fig. 3).

Percent damage was higher in trees that had low C:N in early-season collections at S1 and S3. At S3, there was also a positive relationship between percentage of water in early-season leaves and percent damage. There was no relationship between TAE in early-season leaves and percent damage at S1 or S3 (Table 1). At S1, late-season trees with a high percent damage also had high C:N, but there was a lot of variation in water with respect to damage. At S3, however, there was a significant positive relationship between percent damage and C:N, and a significant negative relationship with water content. There was a significant positive relationship between TAE and percent damage in late-season leaves at S3 (Table 1). There was a highly significant positive relationship between percent damage and the percentage change in C:N before and after agamic gall induction at both sites. However, only at S3 was there a significant negative relationship between percent damage and the percentage change in water. There was no relationship between percent damage and the percentage change in TAE (Table 1).

PC1 accounted for 80% of the variance in the PCA and had positive loadings for percent damage (0.97) and C:N (0.84), and a negative loading for water (−0.87). There was a negative relationship between PC1 (quality) and butterfly production ($R^2 = 0.49$, $P = 0.034$; $n = 9$;

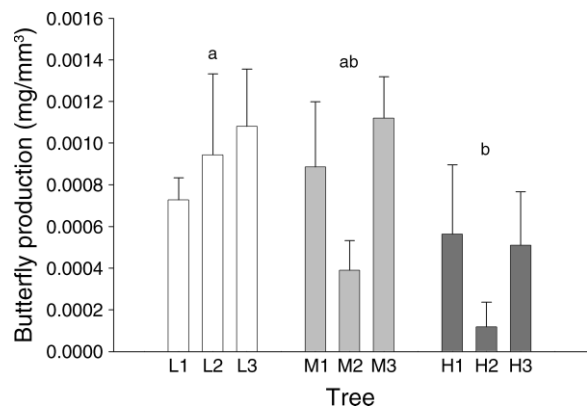


FIG. 2. Butterfly production of *Erynnis propertius* in low (white bars), moderate (light gray bars), and high (dark gray bars) gall-density trees. Each bar is the average of three enclosures (+SE). Letters represent statistically significant differences ($P < 0.05$) in butterfly production among density categories. Untransformed data are shown (see *Methods: Assessing the relationship between gall wasp density and butterfly performance*).

FIG. 3. Linear regression between percent damage caused by *Neuroterus saltatorius* and butterfly production of *Erynnis propertius*. Each point represents the mean of three enclosures on a tree (\pm SE). White circles represent low-density trees; gray, moderate-density trees; and dark gray, high-density trees. Percent damage was estimated digitally from 10 leaves in each caterpillar enclosure. Untransformed data are shown (see *Methods*).

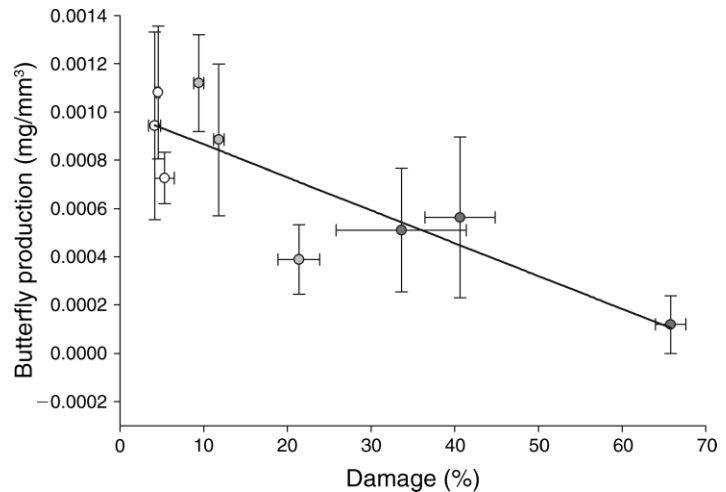


Fig. 4). Percent carbon increased with percent damage ($R^2 = 0.50$, $P = 0.033$), and although not statistically significant, there was a trend toward decreased percent nitrogen with increasing percent damage ($R^2 = 0.36$, $P = 0.085$).

DISCUSSION

Our study suggests that an indirect, plant-mediated competitive interaction is occurring between an invasive gall-former and a native specialist butterfly. Such interspecific competition has not been well documented for gall-forming wasps. We found that butterfly performance decreased with increasing gall wasp density, and that this competitive interaction was likely driven by reduced plant quality in the form of increased carbon, and reduced nitrogen and water. Nitrogen and water are essential nutrients for the development of phytophagous insects (Scriber 1977, Scriber and Slansky 1981), and gall-formers have been shown to concentrate

nutrients in their gall tissue (Paclt and Hassler 1967, Bronner 1992). Our findings suggest that gall-formers could act as superior competitors and that more studies in this guild are needed to advance our understanding of competition among phytophagous insects.

Our results differed from previous studies of interactions with gall-forming chewers. For example, we found evidence of a competitive interaction, but one previous study found evidence of a positive interaction between a stem gall midge and an aphid (Nakamura et al. 2003). This positive interaction occurred because shoots with the gall midge produced a secondary flush of leaves that had high water and nitrogen content. In addition, we found that *N. saltatorius* reduced foliar nitrogen. In contrast, another study found that pin oak (*Quercus palustris*) quality was different between trees infested with the stem cynipid galls, but that heavily infested trees had higher nitrogen content and tannin concentrations (Foss and Rieske 2004). In this same study gypsy

TABLE 1. Linear regressions between percent damage caused by an invasive oak gall wasp, *Neuroterus saltatorius*, on late-season leaves and three measures of foliar quality at two sites (S1 and S3) in 2007 and 2008 on Vancouver Island, British Columbia.

Site, season, % change	Water content (%)			C:N			TAE (mg)		
	R^2	β	P	R^2	β	P	R^2	β	P
S1									
Late-season 2007†	0.095		0.104	0.611	+	< 0.001
Early-season 2008‡	0.067		0.222	0.200	-	0.029	0.014		0.659
Late-season 2008†	0.082		0.175	0.393	+	0.002	0.132		0.202
% change 2008†	0.122		0.120	0.511	+	< 0.001	0.203		0.106
S3									
Late-season 2007†	0.427	-	< 0.001	0.582	+	< 0.001
Early-season 2008‡	0.289	+	0.007	0.206	-	0.026	0.016		0.594
Late-season 2008†	0.296	-	0.006	0.273	+	0.009	0.200	+	0.048
% change 2008†	0.396	-	0.001	0.633	+	< 0.001	0.086		0.210

Notes: Percent damage and foliar quality are assessed from 10 leaves collected from 30 trees of the host *Quercus garryana* at each site. Symbols in the " β " columns represent the direction of the slope of the regression line (for significant regressions only). Statistically significant regressions ($P < 0.05$) are shown in boldface type. TAE stands for tannic acid equivalent.

† Independent variable is percent damage; dependent variable is foliar quality.

‡ Independent variable is quality in early-season leaves; dependent variable is percent damage.

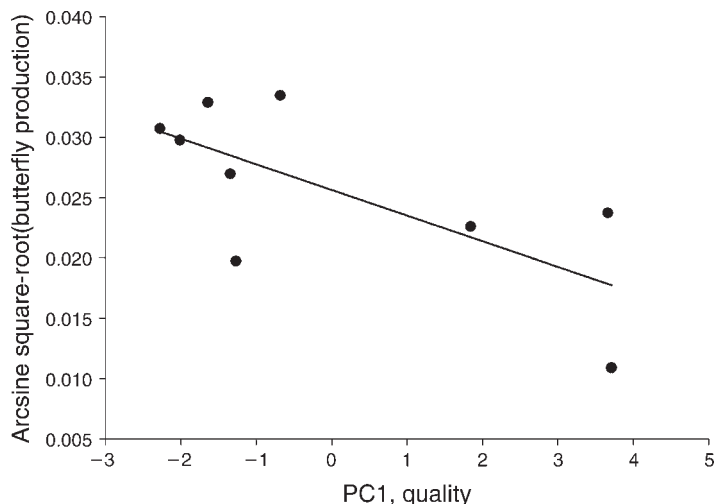


FIG. 4. Linear regression between PC1 (quality) and butterfly production of *Erynnis propertius*. Each data point represents a tree ($n = 9$). Production is measured as the average production of three enclosures on a tree, and PC1 (quality) is measured as the first principal component of quality components measured in control enclosures on each tree. See (Methods: Assessing the relationship between gall wasp density and host plant quality). Butterfly production is arcsine square root-transformed, where the original units of butterfly production are milligrams per cubic millimeter.

moth caterpillars preferred, but did not have higher performance, on ungalled foliage. However, host plant quality was not measured before and after gall induction in this study, so stem galls could have been responding to constitutive foliar quality rather than affecting foliar quality.

We predicted that *N. saltatorius* would interact with *E. propertius* because agamic gall development and caterpillar development occur contemporaneously over ~3–4 months (Appendix B). While we facilitated this interaction in our enclosures, we suspect that this interaction occurs in nature, because we have observed caterpillars of *E. propertius* on high-density trees in the wild (K. Prior, *personal observation*). It is also likely that *E. propertius* caterpillars cannot move among trees, given their small and sessile nature, and that they develop on trees where their eggs were laid. We do not know if *E. propertius* females prefer to oviposit on certain oak trees, but it would be unlikely that females ovipositing in May are able to accurately assess future host plant quality for larvae developing over ~3–4 months (Craig and Itami 2008). Further, trees with high foliar nutritional quality during butterfly oviposition have high gall densities and low nutritional quality later in the season. If butterflies prefer nutritional trees (e.g., Prudic et al. 2005), we would expect that *E. propertius* would prefer the same trees that support high gall wasp densities. *Erynnis propertius* and *N. saltatorius* may not overlap in nature, however, if female butterflies and female wasps prefer to oviposit on trees with different phenolic profiles or differentially cue into some other unknown environmental variable.

We found that butterfly production decreased on high-density trees relative to low-density trees (Fig. 2) because fewer individuals that developed on high-density trees survived over winter and eclosed into adults. Caterpillars foraging on high-density trees gained less biomass by the end of larval development (i.e.,

overwintering stage), and this could have contributed to low overwinter survival. For example, we found that mortality was 7% higher between overwintering and pupation, and 30% higher between pupation and eclosion for caterpillars developing on high-density trees than on low- or moderate-density trees. Holometabolous insects need to reach a minimum threshold size before they pupate (Nijout 1975), and other studies also have found indirect effects of host plant quality on survivorship mediated by body size (e.g., Stamp and Bowers 1991). Furthermore, overwintering and diapause is metabolically demanding, and larger individuals with greater nutrient reserves often have higher post-diapause fitness (Hahn and Denlinger 2007).

Our results suggest an important role of foliar quality in mediating this competitive interaction. We found consistent patterns of increased C:N on high-density trees (Table 1, Fig. 4). Foliage with high C:N provides low-quality food for caterpillars, as nitrogen is a limiting nutrient and an increase in carbon could be a result of increased carbon-based compounds such as tannins and lignin (Scriber and Slansky 1981). Increases in C:N could result from increasing carbon or decreasing nitrogen. While our results suggest increasing carbon and possible decreases in nitrogen, the relationships are relatively weak, and further investigation into the separate components of C:N is necessary. Changes in C:N could be controlled by the gall-former or by the plant. There is increasing evidence that gall-formers can manipulate nutrients and metabolites to their benefit (Stone and Schönrogge 2003), but little is known about induced plant responses to gall-formers (except see Ollerstam and Larsson 2003). Foliar nitrogen loss has been shown to occur as an induced response to oak defoliators (e.g., Faeth 1986, Wold and Marquis 1997) and could also occur via resorption in response to tissue necrosis (Aerts 1996). Increased foliar carbon could be due to increased metabolism of carbon-based metabolites (e.g., Wold and Marquis 1997) or increases in

photosynthetic rates (e.g., Bagatto et al. 1996). Future investigations into the mechanisms of changed plant quality are needed to fully understand the competitive effects of gall-formers.

Our study cannot rule out other factors that could both influence gall wasp density and butterfly performance. Phenolics, for example, have been implicated in playing an antifungal role in gall-formers (e.g., Taper and Case 1987). Responses of insects adapted to plants containing tannins are variable (see Ayres et al. 1997), and although an oak specialist, we do not know if *E. propertius* is able to cope with tannins. Although we found that carbon increased, we found no relationship between gall wasp density and phenolics (Table 1). We only measured total phenolics, however, and each species could respond differently to different phenolic compounds. Measuring how the full suite of compounds mediates this interaction would be an interesting avenue for future research. A range of other plant traits and environmental variables also could influence gall-former distribution such as plant age, plant vigor, phenology, parasitoids, and genotype (Stone et al. 2002); however, it is unclear if or how these factors would also negatively affect *E. propertius*.

Our finding that *N. saltatorius* likely changes the nutritional quality of *Q. garryana* provides additional evidence that foliar nutrients contribute to this interaction. Host plant quality can be influenced by a host of genetic, abiotic, and biotic factors (Herms and Mattson 1992), and there also could be changes in these other factors that induced responses in individual trees. For example, we found that water content both affected and was affected by *N. saltatorius* at S3 but not at S1 (Table 1), suggesting that other unknown factors had a stronger effect on foliar water content at S1 than the gall-former. *Quercus garryana* occurs in both deep-soil woodland (e.g., S3) and shallow-soil (e.g., S1) rocky outcrop habitats in British Columbia, and soil moisture could limit foliar water content at S1. Given that we surveyed 30 trees at two different sites and observed strong relationships between density and the change in foliar nutrients before and after gall induction provides convincing evidence that *N. saltatorius* modifies its host plant.

Our study assumes that induced responses occur at the tree level, but they could be more localized (e.g., Faeth 1986, Wold and Marquis 1997). We did not test if wasp damage induced local or systematic responses, but we did observe that gall distribution was patchier among trees than within trees (K. Prior, *personal observation*). We tried to capture quality at the tree level by collecting leaves that represented the damage level observed on the majority of the tree. Furthermore, caterpillars had decreased production on high-density trees, even though they were able to forage on leaves that range in gall damage within an enclosure. This indicates that induced responses may be systematic (at least at the branch level), that caterpillars could have little preference for

low-damage leaves, or that options to forage on unaffected leaves are limited on high-density trees.

Recall that *N. saltatorius* occurs at higher densities in British Columbia than it does in its native range, and we found that host plant quality and caterpillar performance decreased with increasing density of this species. This circumstance suggests that this wasp likely has little effect on *E. propertius* populations in its native range. *Erynnis propertius* is already threatened in British Columbia due to habitat loss (Prior et al. 2009), and will likely be further affected by climate change (Pelini et al. 2009). Our study highlights the importance of also understanding the effects of newly emerging insect populations on native insect communities, particularly in taxonomic groups or guilds with the potential to modify resource quality or availability.

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APPENDIX A

Range map of *Quercus garryana* and map of study sites on Vancouver Island, British Columbia (*Ecological Archives* E091-232-A1).

APPENDIX B

Phenology of *Neuroterus saltatorius* and *Erynnis propertius* on Vancouver Island, British Columbia (*Ecological Archives* E091-232-A2).

APPENDIX C

Using *Neuroterus saltatorius* foliar damage as a surrogate for density, and assignment of damage categories for the butterfly (*Erynnis propertius*) performance experiment (*Ecological Archives* E091-232-A3).

APPENDIX D

Measuring the performance of *Erynnis propertius*: size, survivorship, and butterfly production (*Ecological Archives* E091-232-A4).