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Associational susceptibility of a native shrub induced by context-dependent attraction of an invasive herbivore

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Abstract. Indirect interactions among native and invasive species are notoriously difficult to predict. Here, we apply theory from trait-mediated indirect effects and plant–insect interactions to explain the outcomes of multiple invader interactions. The present study investigates the roles of herbivore preference and plant ontogeny in mediating associational effects among a southern California native shrub, *Atriplex canescens*, an invasive annual forb, *Brassica tournefortii*, and an invasive stink bug, *Bagrada hilaris*. *Ba. hilaris* have been observed to form dense aggregations on *A. canescens* in late spring following senescence of *Br. tournefortii*, with subsequent increased mortality of *A. canescens*. A manipulative experiment found *Ba. hilaris* recruited to *A. canescens* in approximately 70 times greater numbers when neighbored by *Br. tournefortii* than when alone. *Ba. hilaris* nymph production, while minimized experimentally, occurred only on *Br. tournefortii*, suggesting *A. canescens* is not of sufficient quality for *Ba. hilaris* reproduction. In greenhouse preference trials, *Ba. hilaris* exhibited an overall preference for *Br. tournefortii* over *A. canescens;* however, the magnitude of preference depended on ontogenetic stage of *Br. tournefortii*. Overall, our results suggest significant potential for associational susceptibility mediated by a combination of *Ba. hilaris* preference for reproductive *Br. tournefortii*, substantial aggregation of *Ba. hilaris* onto *Br. tournefortii*, and a marked decline in *Ba. hilaris* preference for *Br. tournefortii* with advancing ontogeny—triggering spillover onto neighboring plants. Our results demonstrate an important role for multiple invaders and phenologically driven trait changes in mediating associational susceptibility.

Key words: associational effects; context-dependent preference; herbivore attraction; herbivore recruitment; phenology; plant senescence; spillover herbivory; trait-mediated indirect effects.

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INTRODUCTION

Invasive species are among the most significant threats to global biodiversity (Elton 1958, Vitousek et al. 1996, Mack et al. 2000), having wrought devastating impacts on native ecosystems around the world (Crooks 2002, Vilà et al. 2011). The impacts of invasive plants can be characterized generally as modifications to the local abiotic environment experienced by native species (Crooks 2002), or to the existing network of biotic interactions in which native species are embedded (Levine et al. 2003, Vilà et al. 2011). In the latter case, direct interactions between an invader and a native species may also have indirect effects on other organisms in the community. The enormity of potential indirect effects by invasive plants makes them critical to the study of invasion ecology (White et al. 2006).

Associational effects are a class of indirect effects in which consumer–resource interactions are modified by the identity or diversity of neighboring individuals (Barbosa et al. 2009, Underwood et al. 2014). For plants, associational susceptibility occurs when a focal plant experiences greater herbivore damage due to neighboring plant identity

or diversity (Letourneau 1995); conversely, associational resistance occurs when herbivore damage is reduced (Tahvanainen and Root 1972). Both associational susceptibility and associational resistance are thought to be ubiquitous interactions in plant communities (Barbosa et al. 2009), though their importance among taxa and variability across plant communities remains poorly understood (Underwood et al. 2014).

Indirect effects of invasive species on native species are notoriously difficult to predict. To begin with, indirect interactions among co-existing species generate dense, reticulate interaction webs that produce emergent properties (Ohgushi et al. 2012). Further, despite concerted efforts among ecologists to pinpoint characteristics that predispose some species to become invasive, reliable predictions of invasiveness largely rely on estimates of propagule pressure over species traits (Simberloff 2009). In other words, generalizable predictions based upon the traits of interacting species remain elusive. For example, the bio-control weevils, *Rhinocyllus conicus* and *Larinus planus*, introduced in North America to control *Carduus nutans* (musk thistle) attack less-preferred native thistles in proximity to target weeds. These insects increase both oviposition and feeding rates on native thistles with increasing density and proximity of the invasive musk thistle (Rand et al. 2004, Russell et al. 2007). Such studies challenge an assumption of the preference–performance hypothesis (Gripenberg et al. 2010): That reduced preference for and performance on non-target host plants minimize the risk of impacts by introduced insect herbivores on non-target hosts (Suckling and Sforza 2014, Catton et al. 2015). In reality, insect herbivores may utilize sub-optimal plants even when preferred plants are present, generating higher-order impacts on native plant density and diversity, and long-term effects on plant population dynamics and community structure (Crawley 1989, Marquis 1992). Damage to native plants by invasive herbivores can indirectly facilitate invasive plants, which underscores the necessity of research on co-occurring invasive species (Simberloff and Von Holle 1999, Kuebbing et al. 2013).

The study of associational effects in invasion biology is nascent, with much of the work to date conducted on plant species pairs with similar life histories, in which direct antagonistic interactions are expected to be important (Rand 2003, Rand et al. 2004, Russell et al. 2007, Kim and Underwood 2014, Catton et al. 2015). Associational effects among dissimilar species pairs, in which both the strength and direction of interactions may vary greatly through time, remain largely unexplored. Because perennial plants persist through shifts in annual plant community composition within seasons and across years, at any time they may neighbor plants with highly similar or dissimilar phenology, plant architecture, defensive chemistry, and resultant herbivore communities. As a result, associational effects may vary in their impact on plant populations with the phenology and ontogenetic stage of one or more plant species.

As in other indirect interactions, associational effects manifest via density-mediated and trait-mediated interactions among herbivores and plants. Density-mediated associational susceptibility occurs as a result of dense plants supporting inflated herbivore populations that later spill over onto a neighbor plant (Rand 2003, Rand et al. 2004). Trait-mediated associational susceptibility occurs when some trait in a neighboring plant (independent of density or abundance) influences some change in herbivore behavior, increasing the herbivore impact on a focal plant (Karban 2010, Pearse et al. 2012). Density-mediated and trait-mediated interactions are not necessarily mutually exclusive, and relative herbivore preference for each plant modulates the strength of associational susceptibility (Rand 2003).

The present study investigates the role of herbivore preference in associational effects among invasive and native plants of differing life histories. This system is comprised of the invasive annual forb, *Brassica tournefortii* Gouan (Brassicaceae; Sahara mustard), the native perennial shrub, *Atriplex canescens* (Pursh) Nutt. (Chenopodiaceae; four-winged saltbush), and an invasive insect herbivore, *Bagrada hilaris* Burmeister (Heteroptera: Pentatomidae; painted bug). *Br. tournefortii* is a noxious forb from the Mediterranean region of northern Africa and southern Europe that has been present in California since at least 1927 (Minnich and Sanders 2000). Its success stems from an early, rapid phenology; dense stands appear following fall rains and begin flowering as early as 40 days post-emergence (Marushia et al. 2012). By late winter, mature individuals produce hundreds of siliques

containing thousands of seeds. *A. canescens* is a trioecious, wind-pollinated shrub native to western North America (McArthur et al. 1992, Ogle et al. 2012). It is a foundation species associated with increased density and diversity of native annual plants, and provides shelter, food, and water for insects, small mammals, reptiles, granivorous birds, deer, and sheep (Bestelmeyer and Wiens 2001). Finally, *Ba. hilaris* is an aposematically colored stink bug native to eastern and southern Africa, Middle East, and Asia, where it is a major agricultural pest of brassicaceous crops and some grains (Palumbo et al. 2016). *Ba. hilaris* first appeared in the western hemisphere in June 2008 in California (Palumbo and Natwick 2010) and is now present throughout much of the western United States and portions of Mexico (Bundy et al. 2012, Palumbo et al. 2016, Torres-Acosta and Sánchez-Peña 2016).

Although *Ba. hilaris* is an oligophage that is most often associated with plants in the family Brassicaceae (Reed et al. 2013, Lambert and Dudley 2014, Palumbo et al. 2016), including weedy mustards, it has been observed to form dense aggregations on *A. canescens* in late spring following seasonal depletion and senescence of local *Br. tournefortii* populations in southern California deserts (Barrows 2014). Many of the attacked *A. canescens* in the Coachella Valley National Wildlife Refuge and in Anza-Borrego Desert State Park suffered mortality in succeeding months (Barrows 2014). These field observations suggest that *Br. tournefortii* senescence may trigger intense *Ba. hilaris* herbivory on *A. canescens*. As a dominant shrub associated with increased native biodiversity and the endangered Coachella Valley fringe-toed lizard (*Uma inornata* Cope), any increase in *A. canescens* mortality risk warrants close examination.

Phenology-driven host-plant switching is common among stink bugs (Kiritani et al. 1965, Panizzi 1997). While field observations of *Ba. hilaris* abundance and distribution are consistent with patterns of associational susceptibility, the phenomenon of annual plants triggering herbivore-induced mortality of perennial plants has not previously been documented in natural ecosystems. Further, it is unclear whether potential *A. canescens* susceptibility is density-mediated (i.e., herbivore spillover due to high densities and proximity of *Br. tournefortii* neighbors) or trait-mediated (i.e., advancing *Br. tournefortii* ontogeny triggers changes in *Ba. hilaris* relative plant preference; Kim and Underwood 2014, Underwood et al. 2014). Accordingly, we used a set of greenhouse and manipulative field experiments to address the following questions: (1) Does *A. canescens* experience greater *Ba. hilaris* recruitment when neighbored by *Br. tournefortii*? (2) Does *Ba. hilaris* exhibit differential preference between *Br. tournefortii* and *A. canescens*? (3) Is *Ba. hilaris* plant preference modified by *Br. tournefortii* ontogeny?

Materials and Methods

Experiments were conducted at the Citrus Research Center and Agricultural Experiment Station (CRC-AES), adjacent to the University of California, Riverside campus in Riverside, California, USA. The experiment station encompasses 510 acres of citrus orchards, greenhouses, experimental farmland, and open space.

Bagrada hilaris *recruitment*

To elucidate whether *A. canescens* experiences greater recruitment of *Ba. hilaris* in the presence of neighboring *Br. tournefortii*, we observed the response of local *Ba. hilaris* to *A. canescens* with and without *Br. tournefortii* neighbors. Ten sites were identified throughout CRC-AES to deploy experimental plants, each site at least 50 m away from the nearest other site. Although the flight range of *Ba. hilaris* has not yet been clearly determined, 50 m is greater than the 3–6 m flights commonly observed at peak daytime temperatures for this species (Huang et al. 2012). At each site, we placed two potted *A. canescens* plants 10 m from one another. One *A. canescens* was randomly designated the treatment plant, and the other the control plant. Two potted *Br. tournefortii* individuals were placed alongside treatment *A. canescens*, such that all neighboring pots were in physical contact. Control *A. canescens* plants had no neighbors. We watered all plants to field capacity 2–3 times weekly, as needed. Mortality of any experimental plants within a site prompted replacement of all *A. canescens* and *Br. tournefortii* plants at that site. Twice weekly, we examined all plants for the presence of *Ba. hilaris*, counted the number of nymphs or adults, and manually removed and added them to greenhouse colonies for use in other experiments.

We explicitly limited the numerical response (i.e., reproduction) of *Ba. hilaris* to *Br. tournefortii* by removing the recruited insects from plants at each census. Reproduction and population growth would likely increase the strength of any observed associational effects by increasing the density of insect herbivores. Thus, our experimental design is a conservative test of the potential for associational effects. This design was employed to allow for detection of patterns in initial *Ba. hilaris* recruitment (i.e., attraction) onto experimental plants. Subsequent behaviors, as well as responses to varying plant quality and phenology, were largely excluded by design and addressed in later studies (Lillian 2017). If trait-mediated associational susceptibility of *A. canescens* is important, we expect greater frequencies of *Ba. hilaris* on *A. canescens* with *Br. tournefortii* neighbors than on *A. canescens* without neighbors.

We analyzed relative *Ba. hilaris* attraction and recruitment onto the 10 sets of plants over 40 censuses in two complementary analyses. First, variation among the three plant types in the summed total of *Ba. hilaris* collected over the duration of the field experiment was assessed by negative binomial generalized linear models for adults and for nymphs, separately with R package MASS (Venables and Ripley 2002, Crawley 2007). Nymphs were excluded from further analysis due to limited data and the minimal contribution of nymphs to recruitment via dispersal (Panizzi et al. 1980). Next, we analyzed variation in seasonal presence/absence of *Ba. hilaris* adults on *A. canescens* alone versus *A. canescens* with *Br. tournefortii* neighbors by a generalized linear mixed-effects model with a fixed effect of plant type, a random effect of replicate identity, and a binomial error distribution with R package lme4 (Crawley 2007). This design was used to capture autocorrelation due to repeated measurements made on individual plants over time. We analyzed all data using R version 3.3.2 (R Core Team 2017).

Bagrada hilaris *preference*

Next, we assessed the relative preference of *Ba. hilaris* for *A. canescens* versus *Br. tournefortii* using a series of choice experiments. Choice arenas were composed of a 40 cm^3 insect cage (BugDorm 2, Megaview Science, Taiwan) containing one mature *A. canescens* potted in 7.5-L round pot of UC mix #3 soil and one *Br. tournefortii* potted in 2.5-L tree pot (Stuewe & Sons, Tangent, Oregon, USA) of UC mix #3 soil (http://agops.ucr.edu/soil/). Plants were watered to field capacity three times weekly. To assess the role of *Br. tournefortii* ontogeny in mediating preference, we manipulated the ontogenetic stage of presented *Br. tournefortii* as a factor with three levels: rosette (basal leaves only), bolting (flowers and fruits present), and senescent (no new buds present, and plant browning). A total of 66 replicate choice trials were conducted, each with a unique *Ba. hilaris* individual and unique *Br. tournefortii* and *A. canescens* plants. *Br. tournefortii* life stages were replicated as follows: 23 at the rosette stage, 30 that were bolting, and 13 senescent. Because it was not feasible to confidently assay *Ba. hilaris* oviposition given the size of the cages and amount of plant biomass, we instead tracked insect position within cages over time as a proxy for some combination of orientation and feeding preference (Sisterson 2008, Zeilinger et al. 2014). A single un-mated adult, *Ba. hilaris*, was introduced into the experimental arena within two d of eclosion. We placed the subject insect on the center floor of the experimental arena and then recorded its position within the arena at six censuses: 30 min, 4, 24, 28, 72, and 76 h post-introduction. For analysis, we recorded the insects as being present at a given census on *A. canescens*, *Br. tournefortii*, or in some other location (e.g., cage wall, cage floor, pot surface).

We assessed *Ba. hilaris* relative preference via a series of related analyses. First, the influence of *Br. tournefortii* life stage on variation in *Ba. hilaris* first choice (among *Br. tournefortii*, *A. canescens*, or other at the 30-min post-introduction census) was analyzed by multinomial logistic regression, with *Br. tournefortii* life stage included as a fixed effect in R package MASS (Venables and Ripley 2002). We followed this by three pairwise Fisher's exact tests assessing variation in *Ba. hilaris* choices only between *Br. tournefortii* and *A. canescens*, for each *Br. tournefortii* life stage separately (Crawley 2007). Next, we assessed variation in *Ba. hilaris'* choices upon each of six censuses by generalized estimator equations with a binomial error structure, logit link function, and exchangeable correlation structure with R package geepack (Højsgaard et al. 2005). *Br. tournefortii* life stage was included as a fixed effect and replicate ID as a random

effect. Wald tests were used to assess significance of the overall model and model coefficients.

Results

In the field study of *Ba. hilaris* recruitment over 40 sampling dates between July and November 2014, we collected a total of 5962 *Ba. hilaris* adults and 335 nymphs from experimental plants. Data from a single site were removed prior to analysis due to a lack of *Ba. hilaris* detection on any experimental plants deployed at that site over the duration of the experiment. We conducted analyses utilizing data from the remaining nine sites. Over the duration of the experiment, the total number of *Ba. hilaris* nymphs collected was greatest on *Br. tournefortii*, significantly more than on *A. canescens* with neighbors and without (Fig. 1; $\chi^2 = 48.471$, df = 2, $P < 0.001$). Further statistical analyses assessed *Ba. hilaris* adult abundance exclusively, as nymph recruitment was explicitly limited by experimental design. The total number of *Ba. hilaris* adults collected was also greater on *Br. tournefortii* relative to *A. canescens* with neighbors and without (Fig. 2; $\chi^2 = 31.992$, df = 2, $P < 0.0001$). *Ba. hilaris* was also present significantly more often on *A. canescens* with neighbors than on *A. canescens* without neighbors ($\chi^2 = 42.106$, df = 1, $P < 0.0001$). On average, *Ba. hilaris* abundance was 70 times greater on *A. canescens* with neighbors than on control *A. canescens* and was more than threefold more abundant on *Br. tournefortii* than on neighbored *A. canescens*. *Ba. hilaris* was most prevalent in the field between early August and mid-October (Fig. 3).

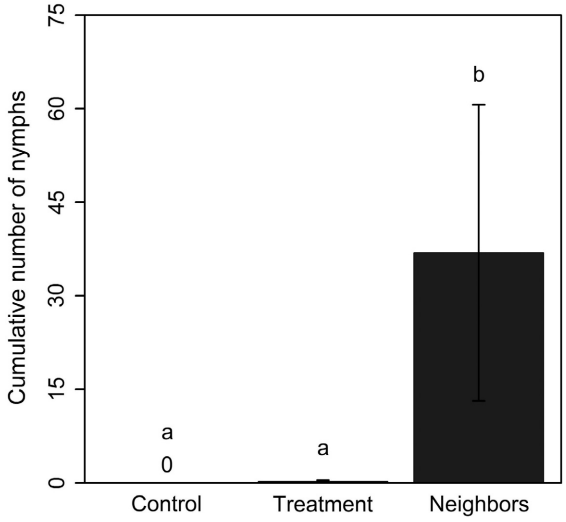


Fig. 1. Mean (±SE) cumulative number of *Bagrada hilaris* nymphs collected over 40 censuses from nine replicate plants of *Atriplex canescens* alone (Control), *A. canescens* with neighboring *Brassica tournefortii* (Treatment), and *Br. tournefortii* (Neighbors). Different letters denote significant differences among groups.

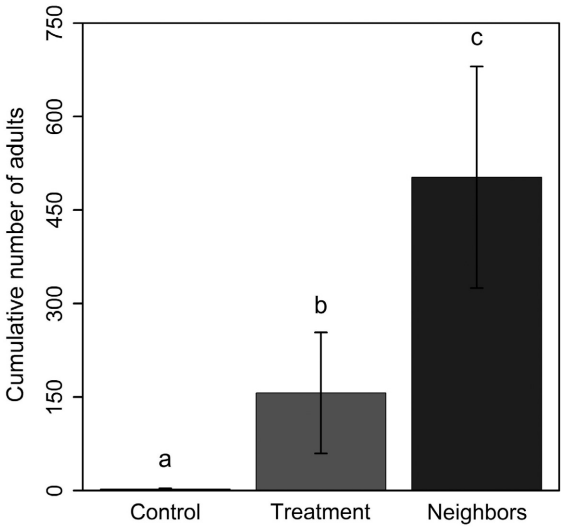


Fig. 2. Mean (±SE) cumulative number of *Bagrada hilaris* adults collected from *Atriplex canescens* alone (Control), *A. canescens* with neighboring *Brassica tournefortii* (Treatment), and *Br. tournefortii* (Neighbors). Different letters denote significant differences among groups.

For the choice experiment, in 32 of 66 trials, *Ba. hilaris* did not choose either plant species by the initial 30 min census; of these, 21 eventually chose one of the two plants. The life history stage of *Br. tournefortii* presented in the experiment had a significant effect on *Ba. hilaris* first choice ($\chi^2 = 12.941$, df = 4, $P = 0.0116$). Although two of the three stages showed a trend consistent with initial preference for *Br. tournefortii* over *A. canescens* (Fig. 4), post hoc pairwise comparison Fisher's exact tests comparing frequencies within each of the *Br. tournefortii* life stages found no significant differences in the frequency of *Br. tournefortii* or *A. canescens* choices. The longitudinal patterns of *Ba. hilaris* choices within replicate trials varied significantly among *Br. tournefortii* life history

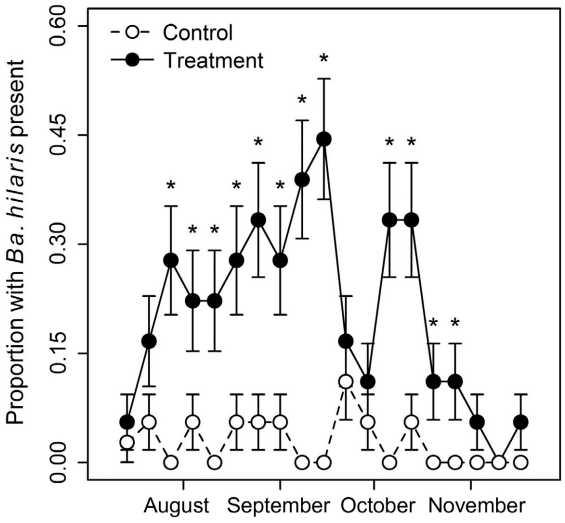


Fig. 3. Proportion (±SE) of *Atriplex canescens* alone (Control) vs. *A. canescens* with neighboring *Brassica tournefortii* (Treatment) with the presence of *Bagrada hilaris* over time. Census dates averaged by week for clarity. Neighboring *Br. tournefortii* (not shown) had *Ba. hilaris* present on all individual plants at nearly every census date. Asterisks denote pairs of censuses for which the treatments differed significantly.

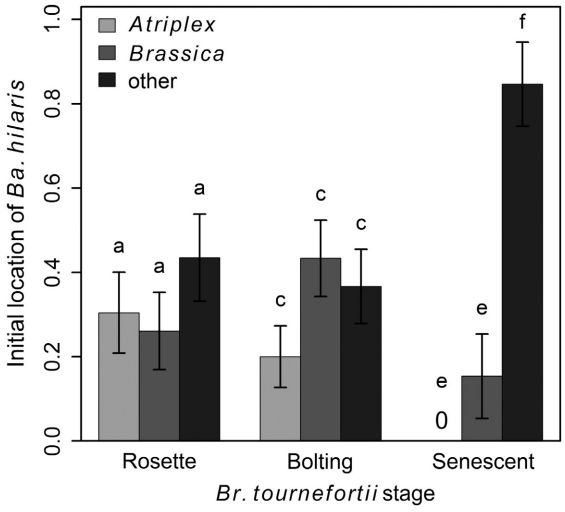


Fig. 4. Proportion (±SE) of *Bagrada hilaris* individuals found at each of three possible locations in choice trials: on *Atriplex canescens*, on *Brassica tournefortii*, or on an other surface. Trials consisted of an individual *Ba. hilaris* introduced into arenas containing one *A. canescens* and one *Br. tournefortii* plant at one of three possible ontogenetic stages: rosette in which only basal leaves are present, bolting in which flowers and fruits are present, and senescent in which the plant has discontinued growth and is browning. Different consecutive lowercase letters denote significant differences among locations within a *Br. tournefortii* stage.

stages (χ^2 = 8.26, df = 2, P = 0.016). *Ba. hilaris* was present on *Br. tournefortii* more often than on *A. canescens*, and this pattern was observed most strongly when *Br. tournefortii* were in the bolting stage, followed by rosette, and with the weakest preference for senescent *Br. tournefortii* plants (Fig. 5). Indeed, when restricted to *A. canescens* and senescent *Br. tournefortii*, we most often found *Ba. hilaris* on neither plant.

Discussion

This study investigated the underlying role of herbivore–plant preference in contributing to associational effects among invasive and native plants of divergent life histories. Collectively, our results suggest the potential for associational susceptibility of the native shrub *A. canescens* that is driven in part by attraction of the invasive herbivore *Ba. hilaris* to the invasive annual mustard *Br. tournefortii* growing in close proximity, followed by herbivore spillover after senescence of mustard plants. It has been suggested in agricultural systems that *Ba. hilaris* infest some crop plants simply due to their proximity to alternative brassicaceous hosts (e.g., *Brassica* crops or weeds in field margins; Lambert and Dudley 2014, Palumbo et al. 2016). Thus, the patterns of associational susceptibility presented here may be consistent with those observations from agriculture, while also providing insights into the contributing factors for apparent declines in native plant populations (Barrows 2014).

In seeking host plants, insect herbivores use volatile chemicals emitted by plants, as well as visual and gustatory cues to recognize and accept suitable hosts (Bruce et al. 2005, Chapman 2009). Differences in apparent suitability among plant species may result in substantially different patterns of herbivore recruitment. Our field experiment showed *Ba. hilaris* occurrence on *A. canescens* depended on whether *Br. tournefortii* neighbors were present. These results provide further evidence of brassicaceous plants being favored hosts for *Ba. hilaris* (Reed et al. 2013,

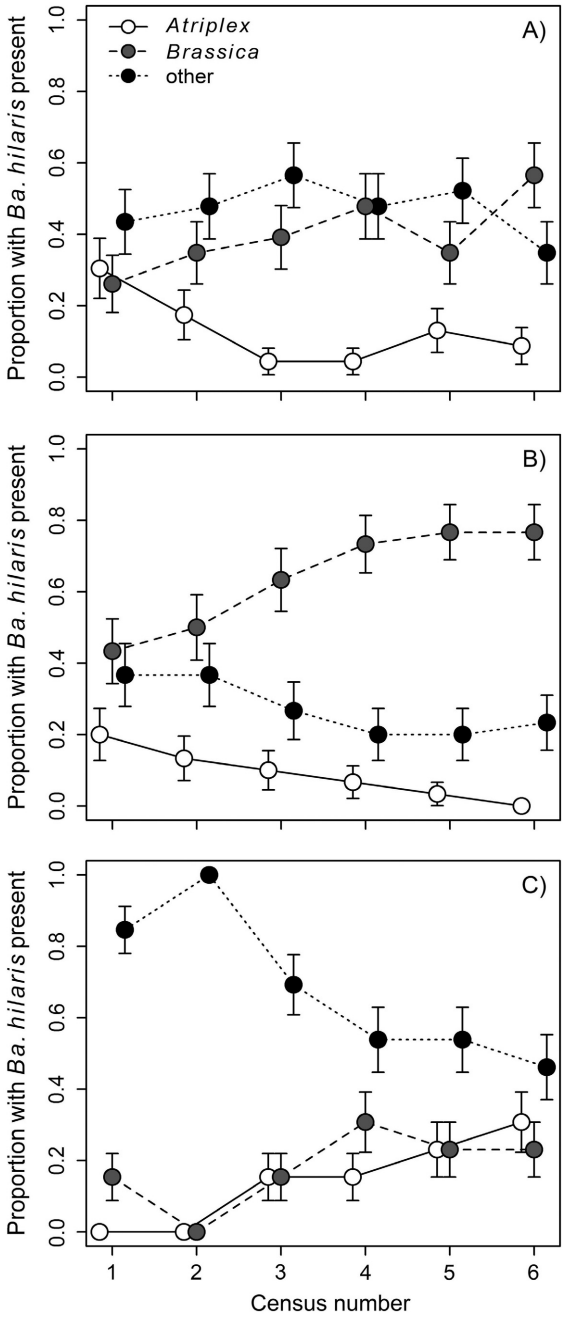


Fig. 5. Proportion (±SE) of *Bagrada hilaris* found at each of three possible locations (on *Atriplex canescens*, *Brassica tournefortii*, and an other surface) over six censuses in choice trials between *A. canescens* and *Br. tournefortii* in one of three ontogenetic stages: (A) *Br. tournefortii* in rosette stage, (B) *Br. tournefortii* in bolting stage, (C) *Br. tournefortii* in senescent stage. Points offset slightly for clarity.

Lambert and Dudley 2014, Palumbo et al. 2016), while also suggesting that their presence may promote herbivore recruitment onto nearby less-preferred hosts. It is worth noting that in the experiment, total plant density and *Br. tournefortii* neighbor presence are confounded. This design was intended to capture herbivore recruitment to *A. canescens* in a context that reflects the natural history of the system, in which neighboring plants are sparse at uninvaded sites or are dominated by numerous *Br. tournefortii* plants at invaded sites (Barrows et al. 2009, Barrows 2014). Yet, it also limits our ability to parse out effects of neighboring plant density from plant identity. However, if recruitment was driven purely by plant density, there should be roughly equal insect occurrence on *Br. tournefortii* and on their neighboring *A. canescens*, and approximately a threefold difference in total herbivores between treatments. Instead, on average, there were nearly twice as many total *Ba. hilaris* adults per plant on *Br. tournefortii* as on neighboring *A. canescens* and a nearly 300-fold difference in total adults between treatments. The relative differences were even more pronounced for nymphs. Thus, although plant density may have influenced the observed patterns, the inherent attractiveness of mustards (Lambert and Dudley 2014) appears to be a driving force in *Ba. hilaris* recruitment in our experiment.

The conclusion that neighbor plant identity was an important element of *Ba. hilaris* recruitment on *A. canescens* is supported by the results from our greenhouse preference experiment. In those trials, *Ba. hilaris* was found more often overall on *Br. tournefortii* compared to *A. canescens*. However, preference was driven primarily by bolting *Br. tournefortii* and largely disappeared with senescent *Br. tournefortii*. The design of our trials was comparable to other studies that use the specific location of herbivores over time or their leaving rates from a given plant as an indication of relative preference (Zeilinger et al. 2014). Although egg laying was not quantified in these preference bioassays, it is worth noting that the large differences in the number of *Ba. hilaris* nymphs in the field experiment may be consistent with a higher oviposition preference for *Br. tournefortii*. *Ba. hilaris* feeding damage is apparent on the two plant species (Lillian 2017), suggesting

that both are potential host plants, but they differ substantially in their quality for *Ba. hilaris* in a way that is consistent with their relative attractiveness (Lillian 2017). Moreover, the results of our preference trials suggest that ontogenetic shifts in host phenology or quality may alter herbivore preference hierarchies. Ontogenetic variation in insect herbivory has been well documented and is generally associated with patterns in plant defense and nutritional quality (Awmack and Leather 2002, Barton and Koricheva 2010). Senescing plant tissues rapidly decline in quality by exporting nutrients (e.g., sugars and amino acids) to storage or reproductive tissues (Thomas 2013), and most herbivorous insects are incapable of utilizing senescing and senescent plant tissue (White 2015). Such ontogenetic shifts in herbivore preference, regardless of the specific mechanism, are relevant to association effects because they may serve as a trigger for herbivore spillover.

The differences in *Ba. hilaris* abundance among treatments in the recruitment experiment are consistent with *Ba. hilaris* spillover from more attractive *Br. tournefortii* (Rand et al. 2004). Pestiferous stink bugs in various crop systems are known to switch from wild or weedy plants on to agricultural crops when they become available (Kiritani et al. 1965, Panizzi 1997). Further, phenological shifts in insect herbivore feeding and aggregation behavior have been documented in some other natural systems; Bay checkerspot butterfly (*Euphydryas editha bayensis*) caterpillars more readily switch from the common *Plantago erecta* to patchy *Castilleja exserta* when *P. erecta* plants senesce rapidly (Hellmann 2002), and seed-feeding true bugs (*Corimelaena extensa*) switch from feeding on abundant *Nicotiana obtusifolia* in early spring to feeding in high densities on the more rare *Nicotiana attenuata* in late spring, resulting in increased insect fitness and decreased *N. attenuata* fitness (Stanton et al. 2016). We found spillover to be most dramatic in late August and September, when rapid declines in *Br. tournefortii* quality were also most difficult to minimize experimentally. *Br. tournefortii* exhibits a ruderal life history strategy (Grime 1977), completing its life cycle rapidly, especially under stressful abiotic conditions (Marushia et al. 2012). While experimental plants were promptly replaced when they began to senesce, late in the summer, *Br. tournefortii* showed signs of senescence (e.g., wilting, yellowing) within days of deployment, likely succumbing to water and heat stress in addition to *Ba. hilaris* herbivory. This, along with consistently greater recruitment onto *Br. tournefortii* over *A. canescens*, is consistent with a trait-mediated indirect effect, in which the switch in *Ba. hilaris* foraging behavior exerts a negative impact on *A. canescens*.

The temporal patterns of peak *Ba. hilaris* abundance we observed corroborate what has been observed in the field (Barrows 2014): *Ba. hilaris* feed on *Br. tournefortii* preferentially and subsequently spill over onto *A. canescens*. *Br. tournefortii* exhibits an advanced phenology (Marushia et al. 2012) relative to *A. canescens* and is available as an early food resource to *Ba. hilaris*. However, *Br. tournefortii* also exhibits rapid ontogeny (Marushia et al. 2012), reaching maturity and senescence earlier than related weeds and co-occurring native plants, including *A. canescens*. In this way, we predict that *Br. tournefortii* inflicts both direct and indirect impacts on invaded communities. Its advanced phenology relative to native annuals confers a beneficial priority effect, largely excluding natives (Barrows et al. 2009, Wolkovich and Cleland 2011, Hulton VanTassel et al. 2014), and its rapid ontogeny may buffer *Br. tournefortii* populations against harsh abiotic conditions in short growing seasons and provide abundant food resources to *Ba. hilaris* populations (Marushia et al. 2012). Both features serve to facilitate *Ba. hilaris*, but only for a short period until *Br. tournefortii* senesce, after which alternative host plants must be sought out.

Our results also indicate that peak *Ba. hilaris* abundance on *A. canescens* coincided with seasonal peaks in overall *Ba. hilaris* density on *Br. tournefortii*, which is consistent with a density-mediated indirect effect in which associational susceptibility is driven by inflated herbivore populations spilling over onto neighboring plants. Yet, the experimental design employed is insufficient to estimate the magnitude of density-mediated effects—for a few reasons. First, we purposefully removed *Ba. hilaris* at each census, which likely suppressed recruitment stemming from local reproduction. This was done to be able to estimate more precisely the potential role of neighbor traits, or attractiveness, in associational effects. Second, *Br. tournefortii* were replaced upon the detection of senescence. Again, this was done

to minimize impacts on herbivore attraction stemming from declining *Br. tournefortii* quality through time. Finally, only one level of neighbor density was utilized. The *Br. tournefortii* density utilized here is considerably less than that observed in the field (Barrows et al. 2009, Hulton VanTassel et al. 2014), likely underestimating any density-mediated indirect effects. Additional studies are needed to determine whether varying neighbor density influences the strength of the associational effect (Underwood et al. 2014), as well as the role that potential differences in plant quality play in the magnitude of density-mediated effects (Lillian 2017). Given the strong results observed despite these intentional constraints, we expect *A. canescens* to experience heavier spillover herbivory under natural conditions, which may be relevant to long-term population dynamics and management decisions in the southwest United States where both *Br. tournefortii* and *Ba. hilaris* co-occur.

Associational susceptibility is a complex indirect interaction, influenced by the traits of species involved (e.g., plant defense, herbivore feeding mode), as well as their numeric status (e.g., relative density and frequency). Our results demonstrate that the role of annual plants in indirectly impacting perennial plants is complex and may be important for invasive species management and native species conservation. Indeed, in years of above-average precipitation promoting high *Br. tournefortii* density, *Ba. hilaris* population outbreaks may be more likely and *A. canescens* may be at drastically increased risk. This suggests that environmental conditions that would otherwise be beneficial to *A. canescens* (i.e., increased yearly precipitation) may instead promote extreme negative impacts on this native foundation species. Evaluating this prediction will require further investigation into the population-level responses of *Ba. hilaris* to *Br. tournefortii* and *A. canescens*, and the strength of direct and indirect effects on each plant (Lillian 2017).

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Literature Cited

Awmack, C. S., and S. R. Leather. 2002. Host plant quality and fecundity in herbivorous insects. Annual Review of Entomology 47:817–844.

Barbosa, P., J. Hines, I. Kaplan, H. Martinson, A. Szczepaniec, and Z. Szendrei. 2009. Associational resistance and associational susceptibility: having right or wrong neighbors. Annual Review of Ecology, Evolution, and Systematics 40:1–20.

Barrows, C. W. 2014. Sahara mustard, *Brassica tournefortii*: trophic impacts on a desert sand dune community. Page 34 *in* American Association for the Advancement of Science – Pacific Division Annual Meeting. Riverside, California. June 17–20, 2014. AAAS Pacific Division, Ashland, Oregon, USA.

Barrows, C. W., E. B. Allen, M. L. Brooks, and M. F. Allen. 2009. Effects of an invasive plant on a desert sand dune landscape. Biological Invasions 11:673–686.

Barton, K. E., and J. Koricheva. 2010. The ontogeny of plant defense and herbivory: characterizing general patterns using meta-analysis. American Naturalist 175:481–493.

Bestelmeyer, B. T., and J. A. Wiens. 2001. Ant biodiversity in semiarid landscape mosaics: the consequences of grazing vs. natural heterogeneity. Ecological Applications 11:1123–1140.

Bruce, T. J. A., L. J. Wadhams, and C. M. Woodcock. 2005. Insect host location: a volatile situation. Trends in Plant Science 10:269–274.

Bundy, C. S., T. R. Grasswitz, and C. Sutherland. 2012. First report of the invasive stink bug *Bagrada hilaris* (Burmeister) (Heteroptera: Pentatomidae) from New Mexico, with notes on its biology. Southwestern Entomologist 37:411–414.

Catton, H. A., R. G. Lalonde, and R. A. De Clerck-Floate. 2015. Nontarget herbivory by a weed biocontrol insect is limited to spillover, reducing the chance of population-level impacts. Ecological Applications 25:517–530.

Chapman, R. F. 2009. Foraging and food choice in phytophagous insects. Pages 99–141 *in* J. D. Hardege, editor. Chemical ecology. EOLSS Publications, Oxford, UK.

Crawley, M. J. 1989. Insect herbivores and plant population dynamics. Annual Review of Entomology 34:531–564.

Crawley, M. J. 2007. The R Book. John Wiley and Sons, Chichester, UK.

Crooks, J. A. 2002. Characterizing ecosystem-level consequences of biological invasions: the role of ecosystem engineers. Oikos 97:153–166.

Elton, C. S. 1958. The ecology of invasions by animals and plants. Methuen & Co, London, UK.

Grime, J. P. 1977. Evidence for the existence of three primary strategies in plants and its relevance to ecological and evolutionary theory. American Naturalist 111:1169–1194.

Gripenberg, S., P. J. Mayhew, M. Parnell, and T. Roslin. 2010. A meta-analysis of preference-performance relationships in phytophagous insects. Ecology Letters 13:383–393.

Hellmann, J. J. 2002. The effect of an environmental change on mobile butterfly larvae and the nutritional quality of their hosts. Journal of Animal Ecology 71:925–936.

Højsgaard, S., U. Halekoh, and J. Yan. 2005. The R package geepack for generalized estimating equations. Journal of Statistical Software 15:1–11.

Huang, T., D. A. Reed, T. M. Perring, and J. C. Palumbo. 2012. Diel activity and behavior of *Bagrada hilaris* (Hemiptera: Pentatomidae) on desert cole crops. Journal Economic Entomology 106:1726–1738.

Hulton VanTassel, H. L., A. M. Hansen, C. W. Barrows, Q. Latif, M. W. Simon, and K. E. Anderson. 2014. Declines in a ground-dwelling arthropod community during an invasion by Sahara mustard (*Brassica tournefortii*) in aeolian sand habitats. Biological Invasions 16:1675–1687.

Karban, R. 2010. Neighbors affect resistance to herbivory - a new mechanism. New Phytologist 186: 564–566.

Kim, T. N., and N. Underwood. 2014. Plant neighborhood effects on herbivory: Damage is both density and frequency dependent. Ecology 96:1431–1437.

Kiritani, K., N. Hokyo, K. Kimura, and F. Nakasuji. 1965. Imaginal dispersal of the southern green stink bug, *Nezara viridula* L., in relation to feeding and oviposition. Japanese Journal of Applied Entomology and Zoology 9:291–297.

Kuebbing, S. E., M. A. Nuñez, and D. Simberloff. 2013. Current mismatch between research and conservation efforts: the need to study co-occurring invasive plant species. Biological Conservation 160:121–129.

Lambert, A. M., and T. L. Dudley. 2014. Exotic wildland weeds serve as reservoirs for a newly introduced cole crop pest, *Bagrada hilaris* (Hemiptera: Pentatomidae). Journal of Applied Entomology 138:795–799.

Letourneau, D. K. 1995. Associational susceptibility: effects of cropping pattern and fertilizer on Malawian bean fly levels. Ecological Applications 5:823–829.

Levine, J. M., M. Vilá, C. M. D. Antonio, J. S. Dukes, K. Grigulis, and S. Lavorel. 2003. Mechanisms underlying the impacts of exotic plant invasions. Proceedings of the Royal Society of London B: Biological Sciences 270:775–781.

Lillian, S. 2017. Associational susceptibility of a native shrub, *Atriplex canescens*, mediated by an invasive annual forb, *Brassica tournefortii*, and invasive stinkbug, *Bagrada hilaris*. Dissertation. University of California, Riverside, California, USA.

Mack, R. N., D. Simberloff, W. M. Lonsdale, H. Evans, M. Clout, and F. A. Bazzaz. 2000. Biotic invasions: causes, epidemiology, global consequences, and control. Ecological Applications 10:689–710.

Marquis, R. J. 1992. Selective impact of herbivores. Pages 301–325 *in* R. S. Fritz and E. L. Sims, editors. Plant resistance to herbivores and pathogens: ecology, evolution, and genetics. University of Chicago Press, Chicago, Illinois, USA.

Marushia, R. G., M. L. Brooks, and J. S. Holt. 2012. Phenology, growth, and fecundity as determinants of distribution in closely related nonnative taxa. Invasive Plant Science and Management 5: 217–229.

McArthur, E. D., D. C. Freeman, L. S. Luckinbill, S. C. Sanderson, and G. L. Noller. 1992. Are trioecy and sexual lability in *Atriplex canescens* genetically based? Evidence from clonal studies. Evolution 46:1708–1721.

Minnich, R. A., and A. C. Sanders. 2000. *Brassica tournefortii*. Pages 68–72 *in* C. C. Bossard, J. M. Randall and M. C. Hoshovsky, editors. Invasive plants of California's wildlands. University of California Press, Berkeley, California, USA.

Ogle, D. G., L. St. John and D. Tilley. 2012. Plant Guide for fourwing saltbush (*Atriplex canescens*). USDA Natural Resources Conservation Service, Aberdeen, ID Plant Materials Center. 83210-0296.

Ohgushi, T., O. J. Schmitz, and R. D. Holt. 2012. Introduction. Pages 1–6 *in* T. Ohgushi, O. J. Schmitz and R. D. Holt, editors. Trait-mediated indirect interactions: ecological and evolutionary perspectives. Cambridge University Press, Cambridge, UK.

Palumbo, J. C., and E. T. Natwick. 2010. The Bagrada bug (Hemiptera: Pentatomidae): a new invasive pest of cole crops in Arizona and California. Plant Health Progress 49:2–4.

Palumbo, J. C., T. M. Perring, J. G. Millar, and D. A. Reed. 2016. Biology, ecology, and management of an invasive stink bug, *Bagrada hilaris*, in North America. Annual Review of Entomology 61:453–473.

Panizzi, A. R. 1997. Wild hosts of Pentatomids: ecological significance and role in their pest status on crops. Annual Review of Entomology 42:99–122.

Panizzi, A. R., M. H. M. Galileo, H. A. O. Gastal, J. F. F. Toledo, and C. H. Wild. 1980. Dispersal of *Nezara viridula* and *Piezodorus guildinii* nymphs in soybeans. Environmental Entomology 9:293–297.

Pearse, I. S., et al. 2012. Complex consequences of herbivory and interplant cues in three annual plants. PLoS ONE 7:1–10.

R Core Team. 2017. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

Rand, T. A. 2003. Herbivore-mediated apparent competition between two salt marsh forbs. Ecology 84:1517–1526.

Rand, T. A., F. L. Russel, and S. M. Louda. 2004. Local- vs. landscape-scale indirect effects of an invasive weed on native plants. Weed Technology 18:1250–1254.

Reed, D. A., J. C. Palumbo, T. M. Perring, and C. May. 2013. *Bagrada hilaris* (Hemiptera: Pentatomidae), an invasive stink bug attacking cole crops in the Southwestern United States. Journal of Integrated Pest Management 4:1–7.

Russell, F. L., S. M. Louda, T. A. Rand, and S. D. Kachman. 2007. Variation in herbivore-mediated indirect effects of an invasive plant on a native plant. Ecology 88:413–423.

Simberloff, D. 2009. The role of propagule pressure in biological invasions. Annual Review of Ecology, Evolution, and Systematics 40:81–102.

Simberloff, D., and B. Von Holle. 1999. Positive interactions of nonindigenous species: Invasional meltdown? Biological Invasions 1:21–32.

Sisterson, M. S. 2008. Effects of insect-vector preference for healthy or infected plants on pathogen spread: insights from a model. Journal of Economic Entomology 101:1–8.

Stanton, M. A., J. Preßler, C. Paetz, W. Boland, A. Svatoš, and I. T. Baldwin. 2016. Plant-mediated pheromone emission by a hemipteran seed feeder increases the apparency of an unreliable but rewarding host. New Phytologist 211:113–125.

Suckling, D. M., and R. F. H. Sforza. 2014. What magnitude are observed non-target impacts from weed biocontrol? PLoS ONE 9:1–12.

Tahvanainen, J. O., and R. B. Root. 1972. The influence of vegetational diversity on the population ecology of a specialized herbivore, *Phyllotreta cruciferae* (Coleoptera: Chrysomelidae). Oecologia 10:321–346.

Thomas, H. 2013. Senescence, ageing and death of the whole plant. New Phytologist 197:696–711.

Torres-Acosta, R. I., and S. R. Sánchez-Peña. 2016. Geographical distribution of Bagrada hilaris (Hemiptera : Pentatomidae) in Mexico. Journal of Entomological Science 51:165–167.

Underwood, N., B. D. Inouye, and P. A. Hambäck. 2014. A conceptual framework for associational effects: When do neighbors matter and how would we know? Quarterly Review of Biology 89:1–19.

Venables, W. N., and B. D. Ripley. 2002. Modern applied statistics with S, 4th edition.. Springer-Verlag, New York, New York, USA.

Vilà, M., J. L. Espinar, M. Hejda, P. E. Hulme, V. Jarošík, J. L. Maron, J. Pergl, U. Schaffner, Y. Sun, and P. Pyšek. 2011. Ecological impacts of invasive alien plants: a meta-analysis of their effects on species, communities and ecosystems. Ecology Letters 14:702–708.

Vitousek, P. M., C. M. D' Antonio, L. L. Loope, and R. Westbrooks. 1996. Biological invasions as global environmental change. American Scientist 84:468–478.

White, T. C. R. 2015. Senescence-feeders: a new trophic sub-guild of insect herbivores. Journal of Applied Entomology 139:11–22.

White, E. M., J. C. Wilson, and A. R. Clarke. 2006. Biotic indirect effects: a neglected concept in invasion biology. Diversity and Distributions 12: 443–455.

Wolkovich, E. M., and E. E. Cleland. 2011. The phenology of plant invasions: a community ecology perspective. Frontiers in Ecology and the Environment 9:287–294.

Zeilinger, A. R., D. M. Olson, and D. A. Andow. 2014. A likelihood-based biostatistical model for analyzing consumer movement in simultaneous choice experiments. Environmental Entomology 43:977–988.