



Original Research Article

Absence of native flower visitors for the endangered Hawaiian mint *Stenogyne angustifolia*: Impending ecological extinction?

Clare E. Aslan^{a, b, *}, Christina T. Liang^c, Aaron B. Shiels^d, William Haines^e

^a Landscape Conservation Initiative, Northern Arizona University, Flagstaff, AZ, 86011, USA

^b Conservation Science Partners, Flagstaff, AZ, 86011, USA

^c Institute of Pacific Islands Forestry, USDA Forest Service, Hilo, HI, 96720, USA

^d National Wildlife Research Center, USDA, Fort Collins, CO, 80521, USA

^e Center for Conservation Research and Training, University of Hawaii, Honolulu, HI, 96822, USA

ARTICLE INFO

Article history:

Received 11 June 2018

Received in revised form 27 October 2018

Accepted 27 October 2018

Keywords:

Creeping mint

Flower pollination treatments

Flower visitation observations

Island endemic

Lasioglossum

Mutualism disruption

ABSTRACT

If an organism becomes rare enough that it no longer participates in certain interspecific interactions, it can be said to have become *ecologically extinct*, even though it is still present. This form of extinction is much less recognized than global extinctions, although it may have ramifications for ecological community function. Here, we describe a case of possible or pending ecological extinction of an endemic Hawaiian plant. We performed over 120 h of systematic flower visitation observations of the endangered Hawaiian mint, *Stenogyne angustifolia*, in its wild habitat. The robust size and open shape of *S. angustifolia* flowers, along with their high accessibility, visibility, and nectar content, suggest that they are adapted to animal-mediated pollination. However, only one flower visitor was observed at our focal high-elevation study site: an individual of the non-native bee species *Lasioglossum impavidum*. Experimental pollination treatments indicate that *S. angustifolia* is self-compatible and demonstrates some autogamy, setting fruit and seed in the absence of pollinators. However, experimental additions of pollen increased fruit production, indicating that plants are pollen-limited and that lack of pollinators carries a reproductive cost for this species. Ecological communities throughout Hawaii are highly modified, and the distribution and diversity of the native pollinator community that occurred with *S. angustifolia* prior to these changes are wholly unknown. Nevertheless, the lack of visitation by native pollinators and extremely rare visitation by non-native pollinators suggest that this plant is today contributing little to pollination networks in its high-elevation habitat.

© 2018 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

1. Introduction

Widespread population declines across taxonomic groups and ecosystem types result from global change processes such as habitat loss and fragmentation, biological invasions, and climate change (Brook et al., 2008). *Ecological extinction* occurs when a species becomes rare enough that it no longer interacts meaningfully with the broader ecological community, even

* Corresponding author. Landscape Conservation Initiative, Northern Arizona University, PO Box 5767, Flagstaff, AZ, 86011, USA.

E-mail address: clare.aslan@nau.edu (C.E. Aslan).

though the species itself persists (Estes et al., 1989; Redford, 1992; McConkey and Drake, 2006). Disruption of key interactions as a result of ecological extinction could drive extinction cascades, where species or populations are directly threatened by functional loss (McConkey and Drake, 2006; McConkey & O'Farrill, 2015; Estes et al., 2016).

Although a large number of species interact with threatened and endangered partners (Aslan et al., 2013a), only those that are strongly dependent on those partners for survival or reproduction face possible extinction themselves as a result of ecological extinction (Brodie et al., 2014). Many traits act to limit such dependence (e.g., generalization in morphology or behavior, phenotypic plasticity, adaptive capacity) (Aslan et al., 2016), reducing the probability of extinction cascades. On the other hand, traits such as self-compatibility and autonomous autogamy in plants, which may reduce their dependence on pollinators in the short term, have the capacity to enable persistence of a species but only under conditions of inbreeding that may not be conducive to long-term stability (Vellend et al., 2006; Brys et al., 2011).

Key benefits exchanged in mutualism can include propagule transportation, reproduction and gene flow, water and nutrients, and protection from natural enemies (Bond, 1994; Kiers et al., 2010). Most mutualists interact with more than one partner taxon to obtain these benefits; it is rare that an organism is fully dependent on a single mutualist species for any particular function (Johnson and Steiner, 2000; Blüthgen et al., 2007). For an individual to experience complete mutualism disruption, therefore, it is necessary for all partners capable of interacting with that individual to become locally absent, a case of multiple ecological, local, or global extinctions (Estes et al., 1989; Aslan et al., 2013a).

Island-based ecological communities are often simplified relative to continental communities, comprising only those species and functional groups that successfully reached and established on the island over the course of its history (Loope et al., 1988; Fattorini, 2009). This may be particularly true under current conditions, given high rates of extinction among some island-based taxa (Clavero and García-Berthou, 2005; Kaiser-Bunbury et al., 2010). As a result, diversity is often low within any particular guild on an island (Cowie and Holland, 2006; but see notable adaptive radiation exceptions, e.g., Givnish et al., 2009). Some island species act as super-generalists: occupying broad niches, as a result of low competition, and interacting with large numbers of partners (Olesen et al., 2002). In many interaction networks, however, low diversity within a guild results in a lower number of partners per interacting species is on islands compared with continents (Aslan et al., 2013a). This low number of partners reduces redundancy within interaction networks, such that complete mutualism disruption is more probable in these systems than on mainlands (Aslan et al., 2013a). Evidence of complete mutualism disruption has been documented in cases of large birds and bats becoming ecologically or globally extinct due to overhunting in Tonga (Meehan et al., 2002), forest bird loss on Guam as a result of the brown tree snake introduction (Rogers et al., 2017), and widespread declines of nectarivorous birds in Hawaii due to introduction of avian diseases (Boyer, 2008). In these cases, the losses of animals are substantial and far-reaching enough that many individual plants no longer overlap spatially or temporally with mutualists and have therefore lost all seed dispersal (Tonga, Guam) or pollination (Hawaii) services. Some seed dispersers and pollinators in these cases have become ecologically extinct: they persist in limited locations, but at such small population sizes that they no longer contribute functionally to the broader community (Wiles et al., 2003). Ecological extinction has been documented for certain interactions when population sizes are still relatively large: for example, due to density dependent behaviors, flying foxes in the Pacific cease to be effective seed dispersers as populations decline below a certain threshold, but the bats are still relatively common (McConkey and Drake, 2006). Disruption of such mutualisms could leave plants with reduced capacity for reproduction, gene flow, habitat colonization, and adaptation (Howe and Miriti, 2004; Aguilar et al., 2006; Gallagher et al., 2015), and result in reduced or negative population growth.

To better understand this context of complex ecological interactions and threats to key functions in ecosystems, it is necessary to conduct systematic field studies of ongoing interactions among rare species. Here, we performed flower visitation observations, manual pollination treatments, and nectar sampling to investigate ongoing pollination of the endemic Hawaiian mint *Stenogyne angustifolia*. *Stenogyne angustifolia* currently occurs in only a few locations on Hawai'i Island, all of which are actively managed by public agencies in an attempt to reverse the decline of the species. We set out to identify the pollinators most critical for *S. angustifolia* outcrossing and to determine the plant's breeding system, with the goal of informing recovery efforts for the species. This examination of a critical ecological interaction for a very rare species illuminates the increasing risk of ecological extinction in highly modified systems worldwide.

2. Materials and methods

2.1. Study species

Stenogyne angustifolia is a scandent vine occupying 'a'ā and pāhoehoe lava flows on Mauna Kea and Mauna Loa between 1500 m and 1800 m in elevation on Hawai'i Island (USFWS, 1993). One of 23 *Stenogyne* species in the Hawaiian archipelago (Wagner et al., 2012), *S. angustifolia* is listed as Endangered under the U.S. Endangered Species Act, with a global population estimated at 5000–7500 individuals, all located in the dry forest of upper elevation Mauna Loa (Evans et al., 2002). (The species has not yet been evaluated on the IUCN Red List of Threatened Species, but four congeners appear on that list and all are Critically Endangered; eight congeners are listed as federally Endangered by the US Fish & Wildlife Service.) Flowers of *S. angustifolia* are 2–3 cm long, tubular, yellow to deep fuchsia in color, and borne along the vine-like stems such that they are frequently close to the ground (Fig. 1). Flowers are produced throughout the year and followed by production of black fleshy fruits, comprised of up to four nutlets, approximately five weeks after flower senescence. Due to their size, shape, and color, the flowers of *S. angustifolia* superficially appear adapted for pollination by nectarivorous birds, like the plant's congeners



Fig. 1. Photo of *Stenogyne angustifolia* flowers, demonstrating their robust size and tubular shape.

(Renner, 1998; Wood and Oppenheimer, 2008); as a monophyletic clade characterized by curved, tubular, reddish flowers (Lindqvist and Albert, 2002), it may be that all members of the *Stenogyne* genus were historically bird-pollinated. However, no native nectarivorous birds currently occur in the open, dry forest habitat where *S. angustifolia* occurs on Hawai'i Island. Anecdotal reports have suggested that flowers of *S. angustifolia* are also visited by various insects, although prior to this study there was no attempt to determine whether those insects were acting as pollinators (USFWS, 1993). Our observation methods were designed to enable detection of flower visitation by potential pollinators of any size or guild.

2.2. Study sites

Field observations for this study took place on the US Department of Defense's Pōhakuloa Training Area (PTA), in a site located between 1500 m and 1800 m on the flanks of Mauna Loa on the Island of Hawai'i. We worked within a PTA conservation area that is fenced and has been off-limits to unauthorized personnel for several decades; the area is therefore protected from both non-native, browsing ungulates and most forms of human disturbance. As a result of these protections, a relatively large number (20 species) of Endangered and Threatened plants occur within PTA, and the military actively manages these species for their long-term conservation.

Within PTA, *S. angustifolia* occurs in tropical dry forest dominated by māmane (*Sophora chrysophylla*) and naio (*Myoporum sandwicense*). The substrate comprises lava flows of both 'a'ā and pāhoehoe, ranging in age from 1000 to 5000 years. Extensive invasion of *P. setaceum* in the area imposes a continual risk of grass-fed fire, but the lack of ungulate browsing and propensity of *S. angustifolia* to spread vegetatively have facilitated a robust local population of *S. angustifolia* over the past decade (USFWS, 2012).

2.3. Flower visitation observations

We conducted systematic flower visitation observations to determine the primary potential pollinators for *S. angustifolia*. Flower visitors cannot be assumed to be pollinators, since many visitors are poor pollen transporters (Schemske and Horvitz, 1984); nonetheless, visitation observations can provide a pollinator candidate pool, with frequent visitors likely among the most important pollinators for a given plant. We observed flowering *S. angustifolia* individuals at PTA from March 2015 through February 2016. Observations took place weekly throughout that time period. Each observation consisted of three 50-min periods, separated by 10-min rest periods to prevent observer fatigue. Observations beginning in early morning, late morning, early afternoon, and late afternoon were conducted at equal frequencies, and a small number of additional night observations (4 h total in 30-min blocks distributed over 8 dates throughout the year) were conducted as time and base access allowed. Prior to observations, each observer was first trained in Hawaiian insect identification and in flower morphology and

pollination biology. Each 50-min observation was divided into 10-min blocks. Each such block began with a one-minute scan sampling period, during which the observer remained at a fixed location and noted all visitors interacting in any way with the visible *S. angustifolia* flowers. The observer recorded the number of individuals of each visitor species interacting with each flower as well as the total number of plants and open flowers visible from the observer's vantage point. For the remaining nine minutes of the block, the observer employed focal individual observations (after Renne et al., 2000; Aslan et al., 2013b), observing a single plant visitor at a time and recording all behaviors of that visitor (number of plants visited, number of flowers visited, number of flowers nectar-robbed, pollen harvesting, nectar harvesting, visible pollen transport, etc.) as long as it was visible. The observer recorded the total amount of time over which each visitor was observed. The protocol stipulated that after the visitor was lost from view or 180 s had passed (whichever occurred first), the observer select another focal individual to observe if possible.

To analyze these data, we used scan samples to calculate the average number of individuals per visitor species per open flower per unit time, and focal individual observations to calculate the average number of plants and flowers visited per minute per individual of each visitor species. These two values were then multiplied to generate an overall Visitor Importance value (VI) for each visitor species (Renne et al., 2000; Aslan, 2015). Using this index, a visitor emerges as particularly important if they are either present and interacting with flowers a high percentage of the time or if they interact with a large number of flowers when present.

In all, we conducted 120.67 h of observations at PTA, spread over 53 separate observation occasions, with three-hour observation start times ranging from 6:10 a.m. to 6:35 p.m. Because of the extremely low number of flower visits observed via this effort at PTA (a total of 1 visit over the course of the year), we conducted an additional 20.5 h of observations in spring 2016 at Pu'u Wa'awa'a Forest Reserve, a lower-elevation site to the west of PTA where *S. angustifolia* has been outplanted in relatively high densities and where bees had been anecdotally reported interacting with the outplanted individuals. Start times of observations for this supplementary effort ranged from 10:08 a.m. to 4:04 p.m.

2.4. Pollination treatments

Although our observations detected remarkably low flower visitation (see Section 3.0), we frequently observed fruit production by *S. angustifolia* at PTA. To determine the degree to which *S. angustifolia* is dependent on pollinators for fruit and seed set, we performed manual pollination treatments on flowers in July and October 2015, in January and April 2016, and in June 2017. Treatments included: (a) a pollinator exclusion treatment in which individual flower buds were covered to exclude pollinators and determine the degree to which plants exhibit autonomous autogamy and whether absence of pollinators carries a reproductive cost; (b) an exclusion control wherein the same type of cover was applied to individual flowers in bud stage, pollen from up to three different plant individuals located in the same local population was delivered manually when the flower opened, and the flower was covered once more until senescence, to determine whether the cover itself affected fruit and seed set; (c) a flower supplementation treatment in which pollen from up to three different plant individuals located in the local population was delivered manually to open flowers to evaluate fruit and seed after known pollination; and (d) an unmanipulated treatment in which flowers were marked with small lengths of embroidery thread tied around their pedicels and then allowed to develop without interference, to evaluate average fruit and seed set under existing pollination conditions. We attempted several designs of flower covers for exclusion treatments because covers were frequently lost from developing flowers as a result of the narrow bud shape and windy conditions. Successful exclusions were carried out using two designs: first, wide plastic drinking straws (~1-cm diameter), closed at the end with staples and perforated with small pinholes to allow airflow and cooling while excluding visiting animals; and second, small envelopes that were closed at the ends with fabric tape and were constructed of nylon wedding tulle fabric of loose weave to allow airflow but exclude all visiting animals. Exclusion devices remained on flowers until the corollas had fully wilted and then were removed to allow fruits to develop normally. There was no difference between the two cover types in fruit or seed production, so results from the two cover designs were pooled in data analysis.

Because *S. angustifolia* is Endangered, we minimized our treatment sample size to minimize our potential impact to the reproduction of our study plants. We aimed to apply treatments to 10 *S. angustifolia* plants, with each treatment replicated on three flowers per plant. However, few plants produced 12 flowers in usable reproductive stage at any given time. Additionally, we were committed to handling flowers as gently as possible to minimize our impact, and this care certainly increased the total number of exclusion covers that were lost due to wind. As a result, our successful treatments were unbalanced across plants, with a total of 28 plants receiving treatments, and many receiving fewer than three replicates of each treatment. Quantile-quantile plots indicated that the dataset violated assumptions of normality (Wood, 2010); we therefore used nonparametric Kruskal-Wallis tests to determine whether significant differences in fruit and seed set among treatments were evident, and Dunn's tests for multiple comparisons to determine which pairs of treatments exhibited significant differences. All data were analyzed in the statistical software environment R version 2.14.1 (R Development Core Team, 2012), with significance accepted at $p \leq 0.05$.

We calculated the Index of Self-Incompatibility (ISI) as the ratio of fruit set by bagged flowers to fruit set after hand-pollination (Zapata and Arroyo, 1978). We calculated the Pollen Limitation Index (PLI) as: $1 - (\text{fruit or seed set of unmanipulated flowers})/(\text{fruit or seed set of hand-pollinated flowers})$ (Larson and Barrett, 2000). The PLI indicates the amount to which ambient pollen transfer is limiting reproduction; a value of 0 indicates no pollen limitation and a value of 1 indicates complete pollen limitation (Larson and Barrett, 2000). Calculating this index for both fruit set (as fruits produced per flower)

and seed set (as seeds produced per fruit) enabled us to detect pollen limitation whether it resulted in reduced numbers of fruit overall or reduced number of seeds in those fruits that were produced.

2.5. Nectar analysis

Since nectar sugar composition can be indicative of pollinator preferences and thus indicate the pollinator functional groups likely to interact with a plant, we took nectar samples from *S. angustifolia* flowers and used high performance liquid chromatography to determine their component sugars. We used 0.90 mm-diameter capillary tubes to extract nectar from seven wild individuals, probing the floral nectar chambers and blowing the resulting nectar into 1.5 mL microcentrifuge tubes. Tubes were capped and frozen until they could be returned to the laboratory for analysis. For analysis, samples were diluted four times with distilled water. The ratios of sugars in the nectar were determined by high performance liquid chromatography using a Shimadzu Model 20 HPLC. Ratios of sugars in the nectar were determined by examining the area under the curves of sugar peaks compared to pure standards.

3. Results

3.1. Flower visitation observations

In 120.67 h of observation (724 10-min observation blocks) of *S. angustifolia* flowers at PTA, we observed just one individual visitor: the non-native bee *Lasioglossum impavidum*, which visited an individual at 13:10 in the afternoon on 28 July 2015. In all, 640 open flowers were observed at PTA in the course of those 724 observation blocks. In contrast, in 20.5 h of observation at Pu'u Wa'awa'a, we observed two visiting *Lasioglossum impavidum* individuals and six non-native *Ceratina cf. dentipes* individuals. In all, we observed 281 open flowers at Pu'u Wa'awa'a. The Visitor Importance (VI) value for *L. impavidum* at PTA was 0.000066, whereas the VI for *L. impavidum* at Pu'u Wa'awa'a was 0.014 and the VI for *C. cf. dentipes* at Pu'u Wa'awa'a was 0.032.

3.2. Pollination treatments

In all, pollination treatments were administered to 55 flowers on 28 plants. Fruit set (defined as fruits produced per flower) across all treatments varied significantly (Kruskal-Wallis test, chi-square = 9.56, $p = 0.023$) (Fig. 2). Hand-supplemented flowers set significantly more fruit than bagged, full-exclusion flowers (Dunn's test, $p = 0.023$) and fruits from unmanipulated flowers (Dunn's test, $p = 0.048$). No other pairwise treatment comparisons differed significantly.

The ISI value was 0.19, indicating self-compatibility but limited autogamy; flowers set fruit in the absence of pollinator visits (i.e., in bagged treatments), but at a lower rate than when pollen was manually delivered to them (i.e., in hand-supplementation treatments), indicating that lack of pollinators carries a reproductive cost. The PLI value based on fruit set was 0.48, indicating an intermediate level of pollen limitation (i.e., hand-supplementation doubled the set of fruit relative to unmanipulated flowers, although each fruit that was produced following these two treatments produced on average approximately equal numbers of seeds).

3.3. Nectar analysis

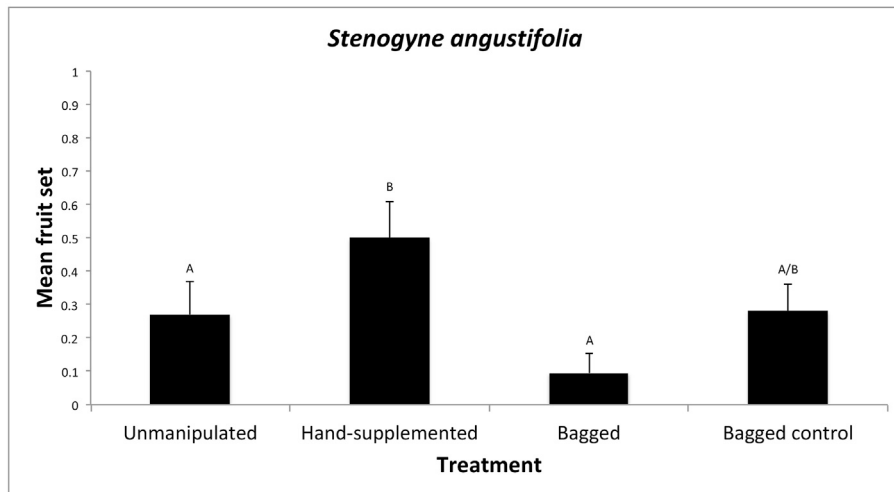
Nectar analysis revealed that sugar composition is hexose-dominated, with glucose and fructose present in roughly equal amounts (mean glucose 148.2 ± 36.7 mg/mL; mean fructose 154.4 ± 39.8 mg/mL) and far less sucrose (mean sucrose 35.9 ± 20.5 mg/mL). Individual samples contained hexose:sucrose ratios ranging roughly from 5:1 to 4:1. As additional indicators of flower attractiveness to potential pollinators, flowers did not carry detectable odor, but each flower contained a small visible pool of nectar, and nectar samples per flower ranged in volume from 2.5 to 13.0 μ L.

4. Discussion

We observed only one species visiting *S. angustifolia* in its wild population and two in the lower elevation outplanted population. Both visitor species were non-native bees. Observations were performed year-round and at all times of day, representing a much higher investment of time than is typical for flower visitation observation studies (e.g., Schemske and Bradshaw, 1999; Johnson et al., 2002; Hanna et al., 2013). For comparison, just 55 h of observation of the relatively common Hawaiian endemic *Argemone glauca* in the same PTA study site yielded 257 visit detections comprising nine insect taxa, including both native and non-native species (unpubl. data).

We cannot know what the historical pollination regime was for *S. angustifolia* since there are no relevant historical records. However, it is logical given the plant's attractive floral resources and endemism within Hawaii that *S. angustifolia* has likely partnered readily with native pollinators in the past. With regard to the two non-native visitors, *L. impavidum* is not rare at PTA, and we have repeatedly observed it visiting other plant species (unpubl. data). The dearth of visits to *S. angustifolia* suggests that *S. angustifolia* may not be very attractive to *L. impavidum*. *Ceratina cf. dentipes*, on the other hand, is primarily a

a.



b.

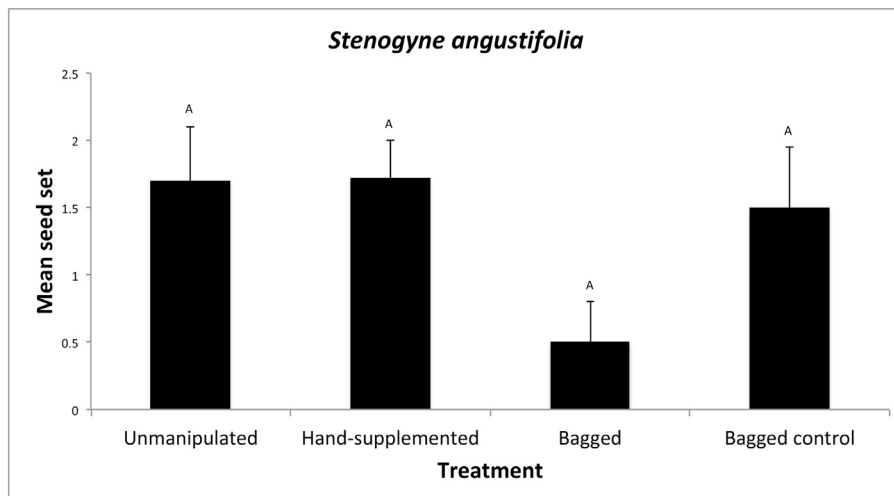


Fig. 2. *Stenogyne angustifolia* flower treatment results, including (a) fruit set (as fruits produced per flower) and (b) seed set (as seeds produced per fruit). Treatments, which per performed at the Pōhakuloa Training Area, included open or unmanipulated flowers, hand-supplementation with pollen, bagged self-fertilized flowers, and bagged control flowers bagged as buds and hand-cross-pollinated when open and receptive. Different letters above bars indicate significant differences between treatments.

coastal species, so its absence at PTA and presence at Pu'u Wa'awa'a might be explained by its distribution and the fact that it is found at lower elevations on Hawai'i Island (Magnacca and King, 2013).

Although the ISI results indicated that *S. angustifolia* is self-compatible and demonstrates autogamy, bagged flowers set significantly less fruit than hand-pollinated flowers, suggesting that access to pollinators is important for maximizing the reproductive output of *S. angustifolia*. This is concerning in light of the plant's rarity and the lack of native flower visitors observed in this study; *S. angustifolia* partners so rarely with even common non-native insect visitors in its wild range that it is largely detached from the local pollinator network and may thus be essentially ecologically extinct in certain interspecific interactions, no longer functioning meaningfully in the community.

The results of nectar analysis were consistent with pollination by either native Hawaiian honeycreepers or native Hawaiian bees, both of which prefer hexose-rich nectar such as that of *S. angustifolia* (Baker and Baker, 1983; Lammers and Freeman, 1986; Pender et al. 2014). The flower morphology appears conducive to pollination by birds: flowers are long and tubular and usually occur in various shades of red. It is impossible to know what native birds might have visited *S. angustifolia* in dry forests in the past. It is likely, however, that such birds are now globally extinct (Boyer, 2008). Native bees do persist in PTA, but their distributions are patchy; we observed them interacting with other endemic plants but whether because they do not

spatially overlap with *S. angustifolia* or because they are not attracted to it (due perhaps to shape, size, color, or the low growth form of the plant), they did not visit wild *S. angustifolia* flowers.

We speculate that the observed dearth of native pollinator visitation to *S. angustifolia* may be related to low population size of the plant. We have, however, also performed observations on *Tetramolopium arenarium*, another endemic plant species of similarly small population size in the study site, and detected a wider diversity (7 taxa) of flower visitors (unpubl. data). Clearly, rarity alone does not inevitably result in ecological extinction from interactions. Nevertheless, in the case of *S. angustifolia* it may be that small population sizes of the plants in their wild range, coupled with flower characteristics, have reduced the functional contributions of the species in the plant/pollinator community to the level of ecological extinction (Altrichter et al., 2012; Valiente-Banuet et al., 2015; Estes et al., 2016). That is, in the context of the full ecological community at the PTA, which contains an active network of native and non-native pollinators and native and non-native plants (these authors, unpubl. data), *S. angustifolia* may have become irrelevant as a resource for extant native and non-native pollinators in the system.

In many cases, non-native pollinators are present in high densities (e.g., Shay et al., 2016) and across varied habitat types and wide geographic ranges, increasing the probability that they will encounter and transfer pollen for even a rare endemic such as *S. angustifolia*. In spite of this, however, the rate of visitation by non-native bees to *S. angustifolia* observed in this study was remarkably low in comparison to pollination rates quantified in other systems, including in Hawaii (e.g., Aslan et al., 2013b). If the non-natives visiting *S. angustifolia* are not able to effect sufficient outcrossing, the species in our study site may be reproducing via self-fertilization. Self-compatibility and autogamy can be advantageous in colonization of new habitats, where they enable individual plants to reproduce and establish populations even if the arrival of a mate is unlikely (Baker, 1955). Colonization of the Hawaiian Islands was likely much easier for self-compatible plant species, and self-compatibility is strongly represented among the Hawaiian flora (Sakai et al., 1995). Following initial colonization of the islands, many species may have persisted via inbreeding for many generations, a process that has the potential to largely erase deleterious alleles from a population via a phenomenon known as purging (Byers and Waller, 1999; Crnokrak and Barrett, 2002). If *S. angustifolia* or its ancestors experienced purging, that may have enabled their persistence under conditions of exclusive inbreeding. However, under global change, inbreeding could hamper a species' ability to successfully adapt to changing environmental conditions (Jump and Penuelas, 2005; Fox and Reed, 2010). Our pollination treatments found that bagged flowers produced fewer fruits on average than the other treatments, indicating a quantitative cost to the absence of pollinators, but the quality of seeds produced via autonomous autogamy has not been examined.

Potential conservation measures in light of extremely low outcrossing for *S. angustifolia* could include bolstering of populations of potential native and non-native pollinators, hand-pollination, and outplanting in lower elevation sites with higher diversity of native and non-native pollinators alike. Such conservation efforts, which leverage biotic interactions and a mixed native/non-native mutualist community, are rooted in an attempt to understand novel ecosystems and ensure their future functioning in spite of transformed community composition. In light of future uncertainty, undertaking such tasks can be daunting. Yet without such active attempts to bolster and maintain critical functions in the face of ecological extinctions, the consequences could be cascading losses of native populations.

Acknowledgements

We extend our deep appreciation to H. Coad, M. Donofrio, J. Felt, J. Smith, and A. Wagner for many hours of field support. We thank everyone at the PTA Natural Resources Office for their logistical support and assistance. This work was performed under USFWS Native Endangered Species Recovery Permit: TE-28360B-0; Hawaii DLNR-DOFAW Permit for Threatened & Endangered Plant Species: P-201; and Hawaii DLNR-DOFAW Permit for Native Invertebrate Research: FHM17-428. Funding for this research was provided by the Department of Defense Strategic Environmental Research and Development Program [grant number RC-2432]. We are deeply grateful to R. Paull for laboratory analysis assistance and to K. Magnacca for bee identification assistance. We thank D. Drake for invaluable comments on an earlier draft of this manuscript.

References

- Aguilar, R., Ashworth, L., Galetto, L., Aizen, M.A., 2006. Plant reproductive susceptibility to habitat fragmentation: review and synthesis through a meta-analysis. *Ecol. Lett.* 9, 968–980.
- Altrichter, M., Taber, A., Beck, H., Reyna-Hurtado, R., Lizarraga, L., Keuroghlian, A., Sanderson EW, E.W., 2012. Range-wide declines of a key Neotropical ecosystem architect, the Near Threatened white-lipped peccary *Tayassu pecari*. *Oryx* 46, 87–98.
- Aslan, C.E., 2015. Pollination of the Endangered Arizona hedgehog cactus (*Echinocereus arizonicus*). *Am. Midl. Nat.* 173, 61–72.
- Aslan, C.E., Zavaleta, E.S., Tershy, B., Croll, D., 2013a. Mutualism disruption threatens global plant biodiversity: a systematic review. *PLoS One* 8, e66993.
- Aslan, C.E., Zavaleta, E.S., Tershy, B., Croll, D., Robichaux, R.H., 2013b. Imperfect replacement of native species by non-native species as pollinators of endemic Hawaiian plants. *Conserv. Biol.* 28, 478–488.
- Aslan, C.E., Bronstein, J.L., Rogers, H.S., Gedan, K.B., Brodie, J., Palmer, T.M., Young, T.P., 2016. Leveraging nature's backup plans to incorporate interspecific interactions and resilience into restoration. *Restor. Ecol.* 24, 434–440.
- Baker, H.G., 1955. Self-compatibility and establishment after "long-distance" dispersal. *Evolution* 9, 347–349.
- Baker, H.G., Baker, I., 1983. A brief historical review of the chemistry of floral nectar. In: Bentley, B., Elias, T. (Eds.), *The Biology of Nectaries*. Columbia University Press, New York, pp. 126–152.
- Blüthgen, N., Menzel, F., Hovestadt, T., Fiala, B., Blüthgen, N., 2007. Specialization, constraints, and conflicting interests in mutualistic networks. *Curr. Biol.* 17, 341–346.
- Bond, W.J., 1994. Do mutualisms matter? assessing the impact of pollinator and disperser disruption on plant extinction. *Philos. Trans. R. Soc. Lond. Ser. B Biol. Sci.* 344, 83–90.

- Boyer, A.G., 2008. Extinction patterns in the avifauna of the Hawaiian Islands. *Divers. Distrib.* 14, 509–517.
- Brodie, J.F., Aslan, C.E., Rogers, H.S., Redford, K.H., Maron, J.L., Bronstein, J.L., Groves, C.R., 2014. Secondary extinctions of biodiversity. *Trends Ecol. Evol.* 29, 664–672.
- Brook, B.W., Sodhi, N.S., Bradshaw, C.J.A., 2008. Synergies among extinction drivers under global change. *Trends Ecol. Evol.* 23, 453–460.
- Brys, R., De Crop, E., Hoffmann, M., Jacquemyn, H., 2011. Importance of autonomous selfing is inversely related to population size and pollinator availability in a monocarpic plant. *Am. J. Bot.* 98, 1834–1840.
- Byers, D.L., Waller, D.M., 1999. Do plant populations purge their genetic load? Effects of population size and mating history on inbreeding depression. *Annu. Rev. Ecol. Systemat.* 30, 479–513.
- Clavero, M., García-Berthou, E., 2005. Invasive species are a leading cause of animal extinctions. *Trends Ecol. Evol.* 20, 110.
- Cowie, R.H., Holland, B.S., 2006. Dispersal is fundamental to biogeography and the evolution of biodiversity on oceanic islands. *J. Biogeogr.* 33, 193–198.
- Crnokrak, P., Barrett, S.C.H., 2002. Perspective: purging the genetic load: a review of the experimental evidence. *Evolution* 56, 2347–2358.
- Estes, J.A., Duggins, D.O., Rathbun, G.B., 1989. The ecology of extinctions in kelp forest communities. *Conserv. Biol.* 3, 252–264.
- Estes, J.A., Burdick, A., Doak, D.F., 2016. Sea otters, kelp forests, and the extinction of Steller's sea cow. *Proc. Natl. Acad. Sci. Unit. States Am.* 113, 880–885.
- Evans, S.A., Faucette, D., Henderson, S., Kawakami, K., Schnell, L., Trainer, J., 2002. Annual Report for the Ecosystem Management Program Pohakuloa Training Area. Island of Hawaii. December 2002.
- Fattorini, S., 2009. On the general dynamic model of oceanic island biogeography. *J. Biogeogr.* 36, 1100–1110.
- Fox, C.W., Reed, D.H., 2010. Inbreeding depression increases with environmental stress: an experimental study and meta-analysis. *Evolution* 65, 246–258.
- Gallagher, R.V., Makinson, R.O., Hogbin, P.M., Hancock, N., 2015. Assisted colonization as a climate change adaptation tool. *Austral Ecol.* 40, 12–20.
- Givnish, T.J., Millam, K.C., Mast, A.R., Paterson, T.B., Theim, T.J., Hipp, A.L., Henss, J.M., Smith, J.F., Wood, K.R., Sytsma, K.J., 2009. Origin, adaptive radiation and diversification of the Hawaiian Lobeliads (Asterales: Campanulaceae). *Proc. Biol. Sci.* 276, 407–416.
- Hanna, C., Foote, D., Kremen, C., 2013. Invasive species management restores a plant–pollinator mutualism in Hawaii. *J. Appl. Ecol.* 50, 147–155.
- Howe, H.F., Miriti, M.N., 2004. When seed dispersal matters. *Bioscience* 54, 651–660.
- Johnson, S.D., Steiner, K.E., 2000. Generalization versus specialization in plant pollination systems. *Trends Ecol. Evol.* 15, 140–143.
- Johnson, S.D., Edwards, J., Carbutt, P.C., 2002. Specialization for hawkmoth and long-proboscid fly pollination in *Zaluzianskya* section *Nycterinia* (Scrophulariaceae). *Bot. J. Linn. Soc.* 138, 17–27.
- Jump, A.S., Penuelas, J., 2005. Running to stand still: adaptation and the response of plants to rapid climate change. *Ecol. Lett.* 8, 1010–1020.
- Kiers, E.T., Palmer, T.M., Ives, A.R., Bruno, J.F., Bronstein, J.L., 2010. Mutualisms in a changing world: an evolutionary perspective. *Ecol. Lett.* 13, 1459–1474.
- Lammers, T.G., Freeman, C.E., 1986. Ornithophily among the Hawaiian Lobelioideae (Campanulaceae): evidence from floral nectar sugar compositions. *Am. J. Bot.* 73, 1613–1619.
- Larson, B.M., Barrett, S.C., 2000. A comparative analysis of pollen limitation in flowering plants. *Biol. J. Linn. Soc.* 69, 503–520.
- Lindqvist, C., Albert, V.A., 2002. Origin of the Hawaiian endemic mints within North American *Stachys* (Lamiaceae). *Am. J. Bot.* 89, 1709–1724.
- Loope, L.L., Hamann, O., Stone, C.P., 1988. Comparative conservation biology of oceanic archipelagos: Hawaii and the Galapagos. *Bioscience* 38, 272–282.
- Magnacca, K.N., King, C.B.K., 2013. Assessing the Presence and Distribution of 23 Hawaiian Yellow-faced Bee Species on Lands Adjacent to Military Installations on O'ahu and Hawai'i Island. Technical Report No. 185. Pacific Cooperative Studies Unit. University of Hawai'i, Honolulu, Hawai'i.
- McConkey, K.R., Drake, D.R., 2006. Flying foxes cease to function as seed dispersers long before they become rare. *Ecology* 87, 271–276.
- McConkey, K.R., O'Farrell, G., 2015. Cryptic function loss in animal populations. *Trends Ecol. Evol.* 30, 182–189.
- Meehan, H.J., McConkey, K.R., Drake, D.R., 2002. Potential disruptions to seed dispersal mutualisms in Tonga, Western Polynesia. *J. Biogeogr.* 29, 695–712.
- Olesen, J.M., Eskildsen, L.L., Venkatasamy, S., 2002. Invasion of pollination networks on oceanic islands: importance of invader complexes and endemic super generalists. *Div. Dist.* 8, 181–192.
- Pender, R.J., Morden, C.W., Paull, R.E., 2014. Investigating the pollination syndrome of the Hawaiian lobeliad genus *Clermontia* (Campanulaceae) using floral nectar traits. *Am. J. Bot.* 101, 201–205.
- R Development Core Team, 2012. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.
- Redford, K.H., 1992. The empty forest. *Bioscience* 42, 412–422.
- Renne, I.J., Gauthreaux Jr., S.A., Gresham, C.A., 2000. Seed dispersal of the Chinese tallow tree (*Sapium sebiferum* (L.) Roxb.) by birds in coastal South Carolina. *Am. Midl. Nat.* 144, 202–215.
- Renner, S.S., 1998. Effects of habitat fragmentation on plant pollinator interactions in the tropics. In: Newbery, D.M., Prins, H.H.T., Brown, N.D. (Eds.), *Dynamics of Tropical Communities: 37th Symposium of the British Ecological Society*. Cambridge University Press, p. 339.
- Rogers, H.S., Buhle, E.R., HilleRisLambers, J., Fricke, E.C., Miller, R.H., Tewksbury, J.J., 2017. Effects of an invasive predator cascade to plants via mutualism disruption. *Nature Comm.* 8, 14557.
- Sakai, A.K., Wagner, W.L., Ferguson, D.M., Herbst, D.R., 1995. Biogeographical and ecological correlates of dioecy in the Hawaiian flora. *Ecology* 76, 2530–2543.
- Schemske, D.W., Bradshaw, H.D., 1999. Pollinator preference and the evolution of floral traits in monkeyflowers (*Mimulus*). *Proc. Natl. Acad. Sci. Unit. States Am.* 96, 11910–11915.
- Schemske, D.W., Horvitz, C.C., 1984. Variation among floral visitors in pollination ability: a precondition for mutualism specialization. *Science* 225, 519–521.
- Shay, K., Drake, D.R., Taylor, A.D., Sahli, H.F., Euaparadorn, M., Akamine, M., Imamura, J., Powless, D., Aldrich, P., 2016. Alien insects dominate the plant–pollinator network of a Hawaiian coastal ecosystem. *Pac. Sci.* 70, 409–429.
- USFWS, 1993. Recovery Plan for *Haplostachys haplostachya* and *Stenogyne angustifolia*. September 20, 1993.
- USFWS, 2012. *Stenogyne angustifolia* (No common name). 5-Year review summary and evaluation. Available at: <https://ecos.fws.gov/ecp0/profile/speciesProfile?sld=1591>. (Accessed October 2017).
- Valiente-Banuet, A., Aizen, M.A., Alcántara, J.M., Arroyo, J., Cocucci, A., et al., 2015. Beyond species loss: the extinction of ecological interactions in a changing world. *Funct. Ecol.* 29, 299–307.
- Vellend, M., Verheyen, K., Jacquemyn, H., Kolb, A., Van Calster, H., Peterken, G., Hermy, M., 2006. Extinction debt of forest plants persists for more than a century following habitat fragmentation. *Ecology* 87, 542–548.
- Wagner, W.L., Herbst, D.R., Lorence, D.H., 2012. Flora of the Hawaiian Islands Website. <http://botany.si.edu/pacificislandbiodiversity/hawaiianflora/index.htm>. (Accessed October 2017).
- Wiles, G.J., Bart, J., Beck, R.E., Aguon, C.F., 2003. Impacts of the brown tree snake: patterns of decline and species persistence in Guam's avifauna. *Conserv. Biol.* 17, 1350–1360.
- Wood, S.N., 2010. Statistical inference for noisy nonlinear ecological dynamic systems. *Nature* 466, 1102–1104.
- Wood, K.R., Oppenheimer, H., 2008. *Stenogyne kauaulaensis* (Lamiaceae), a new species from Maui, Hawaiian islands. *Novon A.J. Bot. Nomencl.* 18, 544–549.
- Zapata, T.R., Arroyo, M.T.K., 1978. Plant reproductive ecology of a secondary deciduous tropical forest in Venezuela. *Biotropica* 10, 221–230.