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Pollination effectiveness of opportunistic Galápagos birds compared to that of insects: From fruit set to seedling emergence

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PREMISE OF THE STUDY: Relying on floral traits to assess pollination systems has been shown to underestimate the ecological service that novel mutualisms can provide. Although vertebrates with opportunistic food habits are common on islands, usually feeding upon flowers of entomophilous species (ES), little is known about how effective they are as pollinators. In a previous study, we had reported that native insectivorous and frugivorous Galápagos birds commonly visit ES flowers, without assessing whether they act as pollinators. Here we investigate this by focusing on three typically ES (*Cryptocarpus pyriformis*, *Waltheria ovata*, *Cordia lutea*) and one mostly ornithophilous species (OS), *Opuntia echios*.

METHODS: The quantitative component (QNC: the product of floral visit frequency and number of flowers contacted) and qualitative components (QLC: fruit and seed set, fruit length, and mass and proportion of seedling emergence) of pollination effectiveness of birds was compared with that of insects.

KEY RESULTS: Birds were not quantitatively important pollinators compared to insects. However, selective exclusion experiments in the four plant species revealed that all qualitative components of fitness improved when both birds and insects visited the flowers. Our study is the first to confirm pollination effectiveness of ES by native opportunistic birds.

CONCLUSIONS: The Galápagos pollination systems are probably more generalized than previous data suggested and, given that ES dominate the flora of this archipelago, we argue that, contrary to expectation, birds might have an important role in maintaining the reproductive success and diversity of plant communities.

KEY WORDS *Cryptocarpus pyriformis*; *Cordia lutea*; *Opuntia echios*; ornithophily; *Waltheria ovata*.

On oceanic islands, specialized pollinators are typically underrepresented, particularly insects, and plants tend to be more generalized in their pollination requirements compared to mainland relatives (Barrett et al., 1996; Traveset et al., 2016). Furthermore, island species experience an increase in their abundance and trophic niche expansion, developing novel and opportunistic mutualisms as a result of an 'interaction release' (sensu Traveset et al., 2015), which is mainly due to food shortage, reduced predation risk, and low interspecific competition. Thus, for instance, typical insectivorous or frugivorous vertebrates (mainly birds and lizards) may be forced to consume nectar and pollen on islands (Grant and Grant, 1981; Olesen and Valido, 2003; Valido et al., 2004). Because these opportunistic vertebrates are not strictly dependent on flower resources, a high insect or fruit availability may result in low dependence on nectar and pollen and hence in reduced flower visitation. However, not all flower visitors are pollinators and not every pollinator grants the same reproductive success (Ne'eman et al., 2010; Ballantyne et al., 2015; Gorenflo et al., 2017). Moreover, variation in visitation frequency to plants (i.e., quantity component of pollination effectiveness, hereafter QNC) may or may not be correlated with plant reproductive success (i.e., quality component, hereafter QLC) (Mayfield et al., 2001; Rodríguez-Rodríguez and Valido, 2008; Gorenflo et al., 2017). The two components of pollination effectiveness have therefore to be assessed in order to properly compare effectiveness between pollinator guilds, especially in those assemblages with marked interspecific differences in life modes (e.g., birds vs. insects) (Rodríguez-Rodríguez et al., 2013).

The vast majority of angiosperms exhibit floral traits (e.g., morphology, color, size, reward, phenology) that suggest evolutionarily specialized pollination by a particular group of pollinators yet are ecological generalists (sensu Ollerton et al., 2007). On many oceanic islands, there is a high frequency of small, bowl-shaped, drab flowers (i.e., typically entomophilous species, hereafter ES) and instead low representation of large, tubular, showy flowers (i.e., typically ornithophilous species, hereafter OS) (Müller, 1881; Stewart, 1911; Carlquist, 1974). Despite the predominance of ES within insular floras, previous attempts have focused on OS to confirm pollination effectiveness (hereafter PE) of opportunistic nectar-feeding birds (Rodríguez-Rodríguez and Valido, 2008, 2011; Le Péchon et al., 2013; Rodríguez-Rodríguez et al., 2013; Fernández de Castro et al., 2017). To our knowledge, only one study has previously documented effectiveness in ES of nonspecialist birds that stopover on Ventotene Island (Mediterranean Sea), finding that birds are more important than insects when the latter are scarce (Cecere et al., 2011). Hence, the ecological service of novel mutualistic agents can be underestimated when simply relying on morphological floral traits (Kelly et al., 2010; Anderson et al., 2016). Such service needs to be known given the global importance of vertebrate pollinators maintaining diversity and plant reproductive success (Ratto et al., 2018).

On the Galápagos, most angiosperms are ES and self-pollinated mainly due to the paucity of insect pollen vectors (Chamorro et al., 2012). The evaluation of their pollination systems has depended heavily on the concept of floral traits; solely insect pollination has been examined, whereas visits by opportunistic feeders have been discounted (McMullen, 1999). However, recent data have revealed that native granivorous/frugivorous and insectivorous birds commonly visit flowers of Galápagos plants (mainly ES) and can even act as double mutualists by consuming both flower rewards and fruits of the same plants (Traveset et al., 2015; Olesen et al., 2018). It remains unknown, though, whether bird–flower visitations to such plants result in effective pollination. Here, we focused on four plant species of this oceanic archipelago: three typically ES: *Cryptocarpus pyriformis* (Nyctaginaceae) Kunth, *Waltheria ovata* (Sterculiaceae) Cav., and *Cordia lutea* (Boraginaceae) Lam. known to be visited by insects (McMullen, 2012; Traveset et al., 2013), and one OS, *Opuntia echios* (Cactaceae) J. T. Howell, with mixed pollination system (birds and bees) (Grant and Grant, 1981). Our study aimed at assessing whether Galápagos birds act as effective pollinators, and compared their QNC (floral visitation frequency and number of flowers contacted) and their QLC (fruit and seed set, fruit size and mass, proportion of seedling emergence, time to seedling emergence and growth rate) with those of insects. If birds act as effective pollinators, their exclusion should result in a reduction of the qualitative subcomponents. Moreover, such reduction in QLC should be greater in OS, given that ES might depend only marginally on bird pollination. The two quantitative subcomponents were used to plot the location of birds and insects on a quantitative component landscape (sensu Schupp et al., 2010).

MATERIAL AND METHODS

Study site and potential bird pollinators

This study was carried out in the arid vegetation zone of Santa Cruz Island, which is the most extensive of all zones (≈60% of total land area) and harbors the highest proportion of endemic species (Guézou

et al., 2010). The studied populations were located in the vicinity of Puerto Ayora: *Cryptocarpus pyriformis*, *Cordia lutea*, and *O. echios* at approximately 10 m a.s.l. (0° 44′ 26.5″ S, 90° 18′ 16.8″ W) and *W. ovata* at approximately 30 m a.s.l. (0° 44′ 19.8″ S, 90° 19′ 23.5″ W) (see https://arcg.is/1TiSr4). Other common species occurring in the study area are: *Cordia leucophlyctis* Hook. f., *Jasminocereus thouarsii* (F.A.C.Weber) Backeb., *Maytenus octogona* D.C, *Tournefortia pubescens* Hook. f., *T. rufo-sericea* Hook. f. and *Scutia spicata* Weberbauer.

The Galápagos climate is characterized by a hot/wet season (January–May) and a cold/dry season (June–December). Although the former normally provides enough food resources, in some years there is little rain, and plant and insect availability is correspondingly sparse, particularly in the arid zone (mean rainfall: 277 mm, mean temperature: 24°C; Trueman and d'Ozouville, 2010).

There are nine common land bird species in the arid zone of Santa Cruz: the three ground finches (*Geospiza fuliginosa*, *G. fortis*, and *G. magnirostris*) feed preferentially on seeds and fruits, while *G. scandens* relies mostly on floral rewards; the tree finch (*Camarhynchus parvulus*) feeds on fruits and insects, the vegetarian finch (*Platyspiza crassirostris*) on leaves and fruits, the Galápagos mockingbird (*Mimus parvulus*) on fruits, insects, lizards and small rodents, whereas the yellow warbler (*Dendroica petechia*) and the Galápagos flycatcher (*Myiarchus magnirostris*) are mainly insectivorous (Grant and Grant, 2008; Heleno et al., 2013; De León et al., 2014; Traveset et al., 2015). However, recent data have revealed that all these species commonly feed upon flowers (Traveset et al., 2015; Olesen et al., 2018).

Study plant species

The four species are within the pollination network in the arid zone of Santa Cruz (Traveset et al., 2013): *Cryptocarpus pyriformis*, *W. ovata*, and *Cordia lutea* are native to the archipelago and the endemic *O. echios* var. *gigantea* is exclusively found in the southern side of Santa Cruz (Wiggins and Porter, 1971). They all are perennial with hermaphroditic flowers, and the stigma and anthers are located in the uppermost part of the corolla (Appendix S1; see the Supplemental Data with this article). The monotypic *Cryptocarpus pyriformis* is a shrub with tiny bowl-shaped white flowers (2 mm long) in terminal and auxiliary panicles, tightly clustered, and one-seeded dry fruits (1.5 mm); its flowers are visited by several introduced ants and an endemic moth (*Aetole galapagoensis*). *Waltheria ovata* is a 0.5–2 m tall shrub with funnel-form dark yellow flowers (6–7 mm) in terminal and axillary clusters and one-seeded dry fruits (2–3 mm). Its flowers are visited by the endemic Galápagos carpenter bee (*Xylocopa darwini*) and an introduced wasp (*Polistes versicolor*) (McMullen, 1999; Chamorro et al., 2012; Traveset et al., 2013). No information exists on how effective the insects visiting those two species are as pollinators. Similarly, there are no published records of bird visits to *C. pyriformis* and *W. ovata*, although their pollen was detected in samples gathered from beaks of *G. fuliginosa*, *G. fortis*, *Camarhynchus parvulus*, *Mimus parvulus*, *D. petechia*, and *Myiarchus magnirostris* (Traveset et al., 2015). *Cordia lutea* is a small tree up to 8 m tall with funnel-form bright yellow flowers (4 cm) in cymes and fleshy fruits (8–15 mm, normally 1 seed). Moths such as the Galápagos sulfur butterfly (*Phoebis sennae*) and *X. darwini* are its most effective pollinators, however, the last often acts as nectar robber, making a hole at the base of the corolla and removing nectar without entering the flower (McMullen, 2012). Its strong flower scent, mean maximum nectar volume and

concentration (24 μl and 19%), and diurnal and nocturnal flower anthesis, suggest that *C. lutea* is entirely entomophilous (McMullen, 2012). Lastly, *O. echios* is a columnar cactus up to 12 m tall with solitary saucer-shaped bright yellow flowers (5–10 cm) and fleshly multiseeded fruits (3–7 cm). *Opuntia* spp. are visited by both insects and birds, but mostly by the latter and it is thus considered here as an OS. Its flowers are primarily visited by the cactus finch, *G. scandens,* but also by *G. fuliginosa*, *G. fortis*, *G. magnirostris*, *Camarhynchus parvulus*, *P. crassirostris*, *Mimus parvulus*, *D. petechia*, and *Myiarchus magnirostris*. The open structure of its flowers also allows visits of insects such as *X. darwini*, *P. versicolor*, and introduced ants (Traveset et al., 2013). A previous study by Grant and Grant (1981) confirmed PE of *G. scandens* and *G. conirostris* visiting *Opuntia echios* var. *echios* and *O. helleri*, on Daphne Major and Genovesa islands, respectively. This cactus is considered self-incompatible (Grant and Grant, 1981) and *Cordia lutea* self-compatible (McMullen, 2012), while no information is available for *Cryptocarpus pyriformis* and *W. ovata*.

Census of flower visitors

In March 2016, one flower/inflorescence of five different individuals of each species was marked at bud stage and monitored every six hours until senescence. The study species presented diurnal and nocturnal anthesis, thus enabling potential visits from both nocturnal and diurnal animals (see Results). Direct observations of flower visitors were made during two consecutive flowering periods in 2016 and 2017. Diurnal censuses were carried out between 6:00 h and 17:59 h (approx. same number of census per hour), encompassing the daytime interval during which both birds and diurnal insects are active (total observation period = 28 h/species). A total of 514 randomly selected plants at comparable stages of flowering were observed during periods of 30 min, of which 15 min were dedicated to insect monitoring from a distance of about 2 m, and 15 min to bird monitoring from a distance of about 5 m with 10×42 binoculars (Olimpus, United Kingdom). The order of insect and bird monitoring was reversed at each consecutive observation. Nocturnal censuses were carried out from 18:00 h to 00:00 h, covering the period of highest night-time activity (18:00–21:00h; McMullen, 2012), and monitoring a total of 65 plants for 15 min with a red lamp (total observation period = 4 h/species). For each visit, we recognized birds and insects at the species level whenever possible and recorded their foraging behavior (e.g., legitimate visits, nectar robbing, florivory, stigma stripping), number of legitimate visits per 15 min per plant (i.e., the visitor entered the corolla and came into contact with the reproductive organs of the flowers) and number of flowers/inflorescences contacted by each pollinator species during a census, standardized by number of flowers/inflorescences observed, number of census per plant species, and specific flower abundance. After observations from a distance, flowers were inspected at close range to confirm the insect species or to collect them for further identification. Identifications were performed at the Charles Darwin Research Station (CDRS, Puerto Ayora, Santa Cruz) using the insect reference collection available at this institution.

Assessment of bird pollination

To investigate the pollination role of all floral visitors (birds and insects), in March 2016, we used selective exclusion experiments in individual plants of approximately similar size. Plant height and width from ground level (or diameter at breast height in the case of

O. echios) and mean flower availability (i.e., mean number of flowers opened to pollinators, monitored weekly over the time span of the exclusion experiments) were measured for each individual, because these variables might be relevant criteria used by birds/insects when visiting flowers. Within each individual, the same number of inflorescence buds (approximately) of *Cryptocarpus pyriformis* and *W. ovata*, or flower buds of *Cordia lutea* (flowers already open were removed) were randomly assigned to three different treatments (3 replicates / 3 treatments / 10 individual plants/species): (1) control treatment, in which flowers were left open to pollination by both birds and insects; (2) bird exclusion treatment by a 25 mm wire mesh cage that allowed only insects to visit the flowers; and (3) autogamy treatment, in which total pollinator exclusion was ensured by a 1 mm cloth mesh bag surrounding the flower/inflorescence to check the plant's ability to self-pollinate (see images of exclusion experiments in Appendix S2). An additional treatment was used in *O. echios* (2 replicates / 4 treatments / 11 individual plants): (4) nocturnal exposure treatment by removing the wire meshes after sunset (18:00 h) and replacing them before sunrise (06:00 h), so large hawkmoths could visit the flowers. Because *Cordia lutea* produces a relatively low level of fruits (McMullen, 2012), we had to increase the number of replicates of this species successively (using a similar number of flowers in each replicate) until each treatment produced at least 10 fruits per individual (15 replicates / 3 treatments / 18 individual plants). This was possible because individuals produced flowers all year round.

To assess whether the mesh cage used in bird-excluded flowers might have also affected insect visits, two camera traps (Ecotone digital trail HE-50, Gdynia, Poland), each deployed for two consecutive days (normal sensor, 15 s video at each trigger approximately 2 s apart; 10 s delay between each trigger), were placed at five different bird-excluded flower/inflorescence (1.5 m from the flower) per species. In addition, we performed direct observations (2 h, eight plants observed/species) of insects visiting excluded flowers, and compared number of visits and number of flowers contacted between bird-excluded and control flowers.

After fruit ripening, the total number of fruits and seeds produced were collected, and length, width and mass of 10 randomly selected fruits and 10 seeds of each replica were measured by the same observer (SHP) using a digital caliper (precision 0.01 mm) and a digital scale (precision 0.01 g). For each treatment, reproductive success was assessed as the ratio between number of fruits/number of flowers (fruit set) and the number of seeds per fruit (seed set). Within each individual, the three replicates of the same treatment were pooled and 10 seeds were randomly selected to subsequently conduct germination experiments (June 2016: *Cryptocarpus pyriformis* and *W. ovata*; December 2016: *Cordia lutea* and *O. echios*). Seeds were randomly arranged in space and sown individually in wells filled with organic fertilized peat moss mix (Berger, BM2, Saint-Modeste, Canada) and constant moisture at the greenhouse of the CDRS. Seedling emergence was recorded daily and seedling height once a week, and the experiments ran for five months. Results are expressed as: (1) proportion of seedling emergence (i.e., proportion of total seeds that emerged), (2) seedling emergence time (i.e., mean number of days to seedling emergence), and (3) seedling growth rate (i.e., increase in plant height per unit time) per treatment and species.

Data analysis

To examine whether the wire bird exclusion might have also affected insect visits, differences in number of visits and number of

flowers contacted between bird-excluded and control flowers were tested using generalized linear models ('glm' function from the package 'stats') in R v.3.4.3 (R Core Team, 2017), fitting data to a Poisson distribution with "treatment" and "plant species" as explanatory variables.

Differences among pollinators were tested using generalized linear mixed models (GLMMs) including each quantitative and qualitative subcomponent as dependent variables in separate models. The GLMMs were conducted using the glmer function of the R package 'lme4', followed by Tukey post hoc tests with the glht function of the package 'multcomp'. Number of visits and number of flower contacted (QNC) were fitted to a Poisson family, including "year" as a random factor and "pollinator class" (Aves or Insecta) as a fixed factor. A total of six estimates of plant reproductive success (QLC) were tested: fruit set or seed set, fruit length or fruit mass (depending upon plant species), proportion of seedling emergence, time to seedling emergence and seedling growth rate; in these models, treatment and mean flower availability were included as fixed explanatory variables. For fruit and seed set (binomial error distribution), and fruit length and mass (Poisson error distribution) we used individual plants and replicates as nested random effects to account for inherent differences among individuals and replicates. For proportion of seedling emergence (binomial error distribution) and time to seedling emergence and seedling growth rate (Poisson error distribution), we used individual plants as random effect. Prior to the analyses, we correlated fruit length, width and mass, and seed length, width and mass with proportion of seedling emergence using the rcorr function of the R package 'Hmisc'. We selected the variable most strongly associated with proportion of seedling emergence as an estimation of plant reproductive success: fruit length in *Cryptocarpus pyriformis* ($p < 0.001$) and *Cordia lutea* ($p = 0.023$), and fruit mass in *W. ovata* ($p = 0.013$) and *O. echios* ($p = 0.001$). Plant height and width were not included in the analyses because they were highly correlated with mean flower availability (all $p \leq 0.01$). Power analyses for the GLMMs were conducted using the powerSim function from the R package 'simr' to determine if our lowest sample size of each plant species (see Results) had sufficient power to detect meaningful differences in the qualitative subcomponents among treatments (Green and MacLeod, 2016).

The QNC was calculated as the product of the number of legitimate visits and the number of flowers contacted per 15 min per plant. Mean ± SD values were used to plot the location of bird and insect species on the two-dimensional quantitative effectiveness landscape using the effectiveness_plot function of the 'effect.lndscp' package (Jordano, 2014).

Lastly, to compare mean QLC values between the three ES and the OS, one generalized linear model for each subcomponent was performed with "treatment" and "plant species" as fixed factors.

RESULTS

The blooming peak of *Cryptocarpus pyriformis* and *W. ovata* extended between late March to mid-April, but flowers were found from January to May (mean = 925.0 ± 516.3 SD and 792.0 ± 556.9, respectively). *Cordia lutea* reached the blooming peak in mid-April and five sampled individuals had flowers all year-round (mean = 49.7 ± 53.3). The blooming period of *O. echios* started in late November, peaked between mid-December and mid-January and extended until late March (mean = 9.4 ± 10.9).

On average, the anthesis of the four species lasts 1–3 days; specifically, *Cryptocarpus pyriformis* 3.0 days ± 0.5, *W. ovata* 1.0 ± 0.2 days, *Cordia lutea* 1.5 ± 0.2 days and *O. echios* 2.0 ± 0.2 days.

Quantitative importance of flower visitors

Bird flower visitors comprised eight passerine species, which acted as potential pollinators of at least one of the study species (Table 1). Most bird visits to flowers were legitimate (92%); birds landed on the branch (or flower in *O. echios*) and rapidly introduced the beak into the corolla contacting the stigma and anthers, usually visiting more than one flower/inflorescence each time. All illegitimate visits (8%) were made by *Camarhynchus parvulus* to *Cordia lutea* flowers and *G. scandens* to *O. echios* flowers (see Appendix S3). The former species usually strips flowers (80% of visits to *Cordia lutea*, $n = 267$) to reach the nectar at the base of the corolla where nectaries are located. This behavior was also observed in all visits made by *P. crassirostris* to *C. lutea* ($n = 385$ flowers), thus this bird was considered as nectar robber. *Geospiza scandens* was seen snipping the tip of stamens and stigmas of *O. echios* flowers (37% of visits). Regarding insect flower visitors, at least 39 species were identified (Table 1). Virtually none of the visits by *X. darwini* to *C. lutea* flowers resulted in successful pollination as this species thieved the nectar piercing the corolla base, and seldom contacted reproductive organs (7% of visits).

Cryptocarpus pyriformis—Birds visited flowers less frequently than insects ($z = -3.40$, df = 1, $p < 0.001$), however, they did not differ significantly in the number of flowers contacted ($z = -1.55$, df = 1, $p = 0.123$) ($n = 122$ censuses; Table 1). Quantitatively, birds were not very important pollinators, while insects varied greatly in the QNC landscape mainly because of variation in the number of flowers contacted (Fig. 1). *Geospiza fuliginosa* and *G. fortis* had the highest quantitative scores (≥ 18) with intermediate values for both quantitative subcomponents, while *G. scandens* and *D. petechia* showed the lowest scores (<2) for both subcomponents. Two introduced wasps (*P. versicolor* and *Brachygastra lecheguana*) had the highest scores in number of flowers contacted (>120), whereas three introduced ants (*Tapinoma melanocephalum*, *Wasmannia auropunctata*, and *Brachymyrmex heeri*) had the highest scores in number of visits (Appendix S4). Birds visited flowers mostly early in the morning (06:00–08:00 h) and then around 15:00 h, whereas insects visited during the late morning (10:00–11:00 h) and late afternoon (16:00–18:00 h) (Fig. 2).

Waltheria ovata—Birds made fewer visits to flowers than insects ($z = -5.11$, df = 1, $p < 0.001$) but no differences were found in the number of flowers contacted between the two assemblages ($z = -1.74$, df = 1, $p = 0.058$) ($n = 126$; Table 1). As observed in *Cryptocarpus pyriformis*, the quantitative effectiveness of birds was low, whereas that of insects was contingent on the variation in the number of flowers contacted (Fig. 1). *Camarhynchus parvulus* and *G. fuliginosa* had the highest quantitative scores (>1.9), but with low values of both quantitative subcomponents. The best-scored insects (>130) were the endemic *X. darwini*, which contacted the highest number of flowers, and *P. versicolor*, which made visits most frequently (Appendix S5). The number of bird visits ranged between 0 and 0.02 visits/hour throughout the day. Insects reached the visitation peak at 09:00 h (0.09 visits/hour) and decreased their activity during the day. Alien ants (*Camponotus conspicuus zonatus* and *Paratrechina longicornis*) were also important nocturnal floral visitors (Fig. 2).

TABLE 1. Mean number (± SD) of legitimate visits per 15 min per plant, and of flowers/inflorescences contacted by each pollinator species during a census, standardized by number of flowers observed, number of census per plant species and specific flower abundance, during two consecutive flowering periods in 2016 and 2017 on Santa Cruz, Galapagos.

Order	Family	Visitor species	*Cryptocarpus pyriformis* n visits	n flowers	*Waltheria ovata* n visits	n flowers	*Cordia lutea* n visits	n flowers	*Opuntia echios* n visits	n flowers
Passeriformes		*Camarhynchus parvulus*	1.00 ± 0.00	3.40 ± 3.80	1.00 ± 0.00	2.69 ± 2.31	1.00 ± 0.00	0.07 ± 0.19		0.17 ± 0.07
		Dendroica petechia	1.00 ± 0.00	0.64 ± 0.70			1.33 ± 0.52	0.41 ± 0.41		0.12 ± 0.03
		Geospiza fortis	1.67 ± 1.15	12.37 ± 12.94			1.00 ± 0.00	0.21 ± 0.12	9.25 ± 5.70	0.17 ± 0.07
		Geospiza fuliginosa	1.75 ± 0.96	10.11 ± 13.32	1.00 ± 0.00	2.33 ± 2.37	1.18 ± 0.73	0.94 ± 1.42	12.25 ± 9.39	0.12 ± 0.03
		Geospiza magnirostris							6.00 ± NA	0.06 ± NA
		Geospiza scandens	1.00 ± 0.00	0.50 ± 0.70			1.00 ± 0.00	0.42 ± 0.35	20.89 ± 36.50	0.14 ± 0.13
		Mimus parvulus			1.00 ± 0.00	0.25 ± 0.12			2.66 ± 0.58	0.07 ± 0.01
		Myiarchus magnirostris								
TOTAL BIRDS			1.44 ± 0.82	6.73 ± 10.69	1.00 ± 0.00	0.63 ± 2.30	1.16 ± 0.57	0.26 ± 0.76	16.51 ± 29.97	0.08 ± 0.11
Blattodea	Blattellidae	*Chorisoneura carpenteri*	1.00 ± NA	0.24 ± NA	1.00 ± NA	0.40 ± NA				
Coleoptera		*Coleoptera* sp. 1							1.00 ± NA	0.26 ± NA
	Chrysomelidae	*Acanthoscelides manleyi*								
		Acanthoscelides sp.					1.00 ± 0.00	0.53 ± 0.75		
	Coccinellidae	*Cycloneda sanguinea*	1.00 ± NA	2.33 ± NA	1.00 ± NA	0.19 ± NA	1.00 ± NA	1.55 ± NA		
		Rodolia cardinalis					1.00 ± NA	1.28 ± NA		
	Tenebrionidae	*Lobopoda galapagoensis*							1.00 ± NA	0.26 ± NA
Total Coleoptera										
Diptera		*Diptera* sp. 1	1.00 ± NA	2.33 ± NA	1.00 ± 0.00	0.29 ± 0.15	1.00 ± 0.00	0.63 ± 0.64		
	Sarcophagidae	*Oxysarcodexia taitensis*	2.00 ± NA	2.33 ± NA	3.00 ± NA	1.18 ± NA				
		Tricharaea sp.	1.00 ± 0.00	1.64 ± 1.63	1.00 ± NA	9.33 ± NA				
	Syrphidae	*Ornidia obesa*								
		Pseudodoros clavatus	1.00 ± NA	1.17 ± NA						
Total Diptera			1.25 ± 0.50	1.82 ± 1.37	2.00 ± 1.41	4.75 ± 6.47				
Hemiptera		Hemiptera sp. 1	1.00 ± NA	0.63 ± NA	1.00 ± NA	0.38 ± NA				
Hymenoptera	Apidae	*Xylocopa darwini*	1.00 ± NA	1.00 ± NA	4.39 ± 3.96	30.11 ± 49.37	1.25 ± 0.50	1.19 ± 1.32	5.61 ± 10.12	0.43 ± 0.31
	Formicidae	*Brachymyrmex heeri*	3.75 ± 2.63	4.64 ± 3.55	3.25 ± 2.63	1.34 ± 0.87	4.00 ± 4.24	0.31 ± 0.23	7.36 ± 10.19	0.13 ± 0.11
		Camponotus conspicuus zonatus	2.25 ± 2.09	26.58 ± 28.55	1.00 ± 0.00	0.51 ± 0.38	1.25 ± 0.50	0.08 ± 0.67	1.10 ± 0.32	0.32 ± 0.19
		Monomorium floricola	2.80 ± 2.49	0.86 ± 0.91	4.00 ± 0.82	1.95 ± 2.91	6.72 ± 4.28	3.74 ± 5.29	14.37 ± 9.21	0.20 ± 0.21
		Paratrechina longicornis	2.33 ± 2.16	4.16 ± 2.91	1.00 ± 0.00	1.10 ± 1.19	2.40 ± 1.34	3.16 ± 3.81	1.00 ± NA	0.33 ± NA
		Tapinoma melanocephalum	3.75 ± 3.59	23.44 ± 62.00	15.00 ± 0.71	0.73 ± 0.08	6.32 ± 4.78	7.21 ± 12.37	7.80 ± 4.21	0.40 ± 0.35
		Wasmannia auropunctata	3.11 ± 2.20	8.17 ± 10.83			5.40 ± 3.36	1.69 ± 2.14	19.33 ± 16.07	0.29 ± 0.08
	Total Formicidae		3.09 ± 2.79	16.36 ± 40.62	2.57 ± 1.87	1.27 ± 1.59	5.64 ± 4.39	4.63 ± 8.72	9.00 ± 9.49	0.26 ± 0.23
	Pentomidae	*Nezara* sp.			1.00 ± NA	0.38 ± NA				
	Sphecidae	*Bycertis* sp.			1.00 ± 0.00	0.38 ± 2.28				
	Vespidae	*Brachygastra lecheguana*	2.55 ± 1.51	45.40 ± 58.47	4.00 ± 3.83	15.37 ± 13.59	7.00 ± NA	13.60 ± NA		
		Polistes versicolor	2.92 ± 2.50	65.89 ± 85.33	7.67 ± 5.86	16.89 ± 7.83	1.50 ± 0.71	0.53 ± 0.60	1.50 ± 0.71	0.27 ± 0.28
		Vespidae sp. 1			1.00 ± NA	2.67 ± NA				
	Total Vespidae		2.77 ± 2.11	57.51 ± 74.63	5.10 ± 4.53	15.82 ± 11.71	3.33 ± 3.21	4.89 ± 7.55	1.50 ± 0.71	0.27 ± 0.28
Total Hymenoptera			2.95 ± 2.58	27.16 ± 54.50	3.93 ± 3.72	20.50 ± 40.13	5.31 ± 4.34	4.46 ± 8.44	8.11 ± 9.56	0.29 ± 0.25
Lepidoptera		Lepidoptera sp. 1	1.00 ± NA	0.23 ± NA	1.50 ± 0.71	5.84 ± 3.12	1.00 ± NA	2.00 ± NA		
		Lepidoptera sp. 2			1.00 ± NA	2.80 ± NA				
	Gelechiidae	*Agnippe omphalopa*	1.00 ± NA	15.33 ± NA						
	Geometridae	*Disclisioprocta stellata*			3.86 ± 2.91	12.06 ± 13.00				

(continued)

TABLE 1. (continued)

Order	Family	Visitor species	Cryptocarpus pyriformis		Waltheria ovata		Cordia lutea		Opuntia echios	
			n visits	n flowers	n visits	n flowers	n visits	n flowers	n visits	n flowers
	Hesperiidae	Urbanus galapagensis	1.00 ± 0.00	1.62 ± 1.48	1.00 ± NA	0.61 ± NA	1.33 ± 0.82	2.90 ± 1.19		
	Lycaenidae	Hemiargus ramon	1.00 ± NA	1.17 ± NA	4.61 ± 3.15	3.43 ± 3.43				
		Leptotes parrhasioides	1.00 ± 0.00	1.47 ± 1.08	1.00 ± 0.00	0.47 ± 0.33	1.00 ± NA	0.10 ± NA		
	Total Lycaenidae				3.95 ± 3.17	2.89 ± 3.30	1.00 ± NA	0.10 ± NA		
	Noctuidae	Anticarsia gemmatalis			1.00 ± 0.00	3.16 ± 0.67				
	Pieridae	Phoebis sennae					1.00 ± 0.00	1.50 ± 2.12		
		Podisus sp. 1			1.00 ± 0.00	7.93 ± 8.58				
	Pyralidae	Asciodes gordialis	1.50 ± 0.71	1.39 ± 1.27	4.00 ± 1.41	15.33 ± 1.11			1.00 ± NA	0.43 ± NA
	Sphingidae	Manduca rustica galapagensis			1.00 ± NA	0.67 ± NA	1.00 ± 0.00	0.85 ± 0.20		
Total Lepidoptera			1.14 ± 0.38	3.25 ± 5.40	3.30 ± 2.87	3.25 ± 5.40	1.15 ± 0.55	1.93 ± 1.72	1.00 ± NA	0.43 ± NA
Orthoptera		nymphal Halmenus sp.	1.00 ± NA	0.76 ± NA	2.67 ± 0.58	5.41 ± 7.25				
Phasmida		Phasmida sp. 1	1.50 ± 0.70	19.78 ± 14.77						
TOTAL INSECTS			2.60 ± 2.43	21.17 ± 48.37	3.57 ± 3.35	12.98 ± 30.91	4.55 ± 4.25	3.89 ± 7.70	7.78 ± 9.47	0.25 ± 0.25

Cordia lutea—Birds visited flowers less frequently ($z = -7.99$, df = 1, $p < 0.001$) and contacted fewer flowers than insects ($z = -2.30$, df = 1, $p = 0.023$) ($n = 124$). Quantitatively, neither birds nor insects were very frequent pollinators (Fig. 1), except one introduced wasp (*B. lecheguana*) and four introduced ants (*Tapinoma melanocephalum*, *Monomorium floricola*, *Wasmannia auropunctata*, and *Paratrechina longicornis*), which varied in the QNC landscape mainly because of variation in the number of flowers contacted. *Wasmannia auropunctata* was the best-scored insect (>7.5), whereas *G. fuliginosa* was the highest scored bird (>1, Appendix S6). Flower visitation by birds was generally low (0.02 visits/hour) whereas that of insects was relatively high (0.07 visits/hour), although both assemblages showed the highest visitation activity around midday (12:00 h). Nocturnal insects, mainly ants and moths, peaked activity at 23:00 h (Fig. 2).

Opuntia echios—Birds visited flowers more frequently ($z = 12.95$, df = 1, $p < 0.001$), but a smaller number of flowers than insects ($z = -3.90$, df = 1, $p = 0.002$) ($n = 142$). Unlike the entomophilous species, variation in the QNC landscape of this OS was mainly driven by variation in the number of visits, and this pattern was found for both birds and insects (Fig. 1). *Geospiza scandens* had the highest quantitative score (3), followed by *G. fortis* (1.7) and *G. fuliginosa* (1.6). Three alien ants, *Wasmannia auropunctata* (5.5), *Tapinoma melanocephalum* (3.2), and *Monomorium floricola* (3), and the endemic bee (*X. darwini*, 2.5), were the best-scored insects (Appendix S7). Both bird and insect visitation activity peaked early in the morning (06:00–07:00 h), and then again within the period 14:00–15:00 h (Fig. 2).

Qualitative importance of flower visitors

Camera traps recorded four insect visits to bird-excluded flowers of *Cryptocarpus pyriformis*, five to *W. ovata*, three to *Cordia lutea*, and six to *O. echios*. Various unsuccessful attempts by *G. scandens* to access *O. echios* excluded flowers were also recorded, showing the strong dependence of this bird on this cactus for food. No differences were found in either the number of insect visits or contacted flowers between bird-excluded and control flowers ($z = 0.7$, df = 1, $p = 0.330$ and $z = 0.5$, df = 1, $p = 0.460$, respectively, $n = 64$ plants), confirming the effectiveness of bird exclusion experiments allowing insects but not birds to access the flowers.

Cryptocarpus pyriformis—The large majority of fruits (89%) from the autogamy treatment were empty ($n = 9290$). Significant differences among treatments were detected in seed set ($z = 6.2$, $p < 0.001$, $n = 360$ inflorescences), fruit length ($z = 14.5$, $p < 0.001$, $n = 900$ fruits), and proportion of seedling emergence ($z = 3.2$, $p < 0.001$; $n = 300$ seeds, all df = 2). Specifically, control flowers had larger fruits and higher seed set than flowers from either the exclusion or autogamy treatment. Moreover, bird-excluded flowers set larger fruits and more seeds per fruit than autogamous flowers. Also, control seeds germinated more than either excluded or autogamous seeds, whereas no differences in seedling emergence time and growth rate were observed among treatments ($p \geq 0.05$; Table 2, Fig. 3).

Waltheria ovata—Most fruits (97%) from the autogamy treatment were empty ($n = 6216$). Significant differences among treatments were found in seed set ($z = 7.9$, $p < 0.001$, $n = 360$),
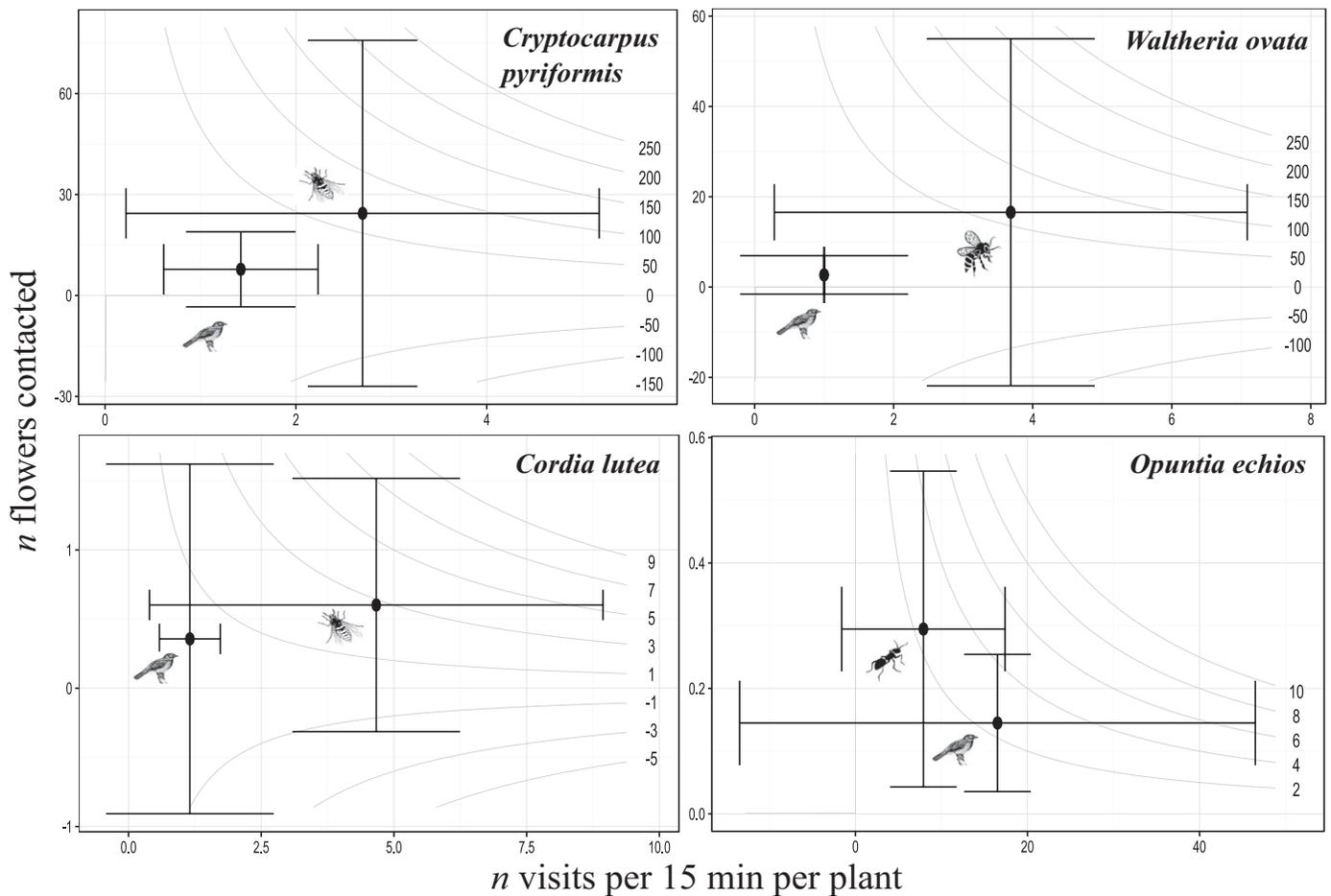
FIGURE 1. The distribution of birds and insects on the quantitative component pollination landscape for *Cryptocarpus pyriformis*, *Waltheria ovata*, *Cordia lutea*, and *Opuntia echios*. Isoclines represent all combinations of mean (± SD) number of visits per 15 min and per plant and number of flowers/inflorescences contacted during a census, standardized by number of flowers observed, number of census per plant species, and specific flower abundance during two consecutive flowering periods in 2016 and 2017 on Santa Cruz, Galápagos.

fruit mass ($z = 14.1$, $p < 0.001$, $n = 900$) and proportion of seedling emergence ($z = 4.1$, $p < 0.001$; $n = 300$, all df = 2). Control flowers had heavier fruits and higher seed set than flowers from either the exclusion or autogamy treatment, and excluded flowers set heavier fruits and more seeds per fruit than autogamous flowers. Moreover, control seeds and excluded seeds germinated more than autogamous seeds, whereas no differences in seedling emergence time and growth rate were found among treatments ($p \geq 0.8$; Table 2, Fig. 3).

Cordia lutea—Only 0.3% of flowers ($n = 5430$) from the autogamy treatment produced fruits, although all those fruits produced seeds. Significant differences among treatments were observed in fruit set ($z = 14.1$, $p < 0.001$, $n = 810$) and fruit length ($z = 2.2$, $p < 0.001$; $n = 314$, all df = 2). Control flowers had greater fruit set and larger fruits than either excluded or autogamous flowers. Excluded flowers set also more fruits and larger fruits than autogamous flowers. No differences among treatments were observed in any of the germination variables ($p \geq 0.3$; Table 2, Fig. 3).

Opuntia echios—All *O. echios* flowers set fruits and 76% autogamous fruits were empty ($n = 17$). Significant differences among

treatments were detected in seed set ($z = 18.1$, $p < 0.001$, $n = 88$), fruit mass ($z = 5.5$, $p < 0.001$, $n = 88$) and proportion of seedling emergence ($z = 4.0$, $p < 0.001$; $n = 440$, all df = 3). Control flowers set more seeds per fruit than either excluded or autogamous flowers, and also than those exposed to nocturnal floral visitors. Flowers exposed to nocturnal insects (e.g., moths like *Manduca rustica*) set more seeds than those from which these were excluded or where exclusion of floral visitors was total. Lastly, bird-excluded flowers showed higher seed set than autogamous flowers. Moreover, control fruits were heavier than those from the exclusion, autogamy, or nocturnal treatment, whereas no differences in fruit mass were observed between the last three treatments. Control seeds and nocturnal exposed seeds germinated in higher proportions than those from exclusion or autogamy treatment, whereas no differences in seedling emergence time and growth rate were found among treatments ($p \geq 0.05$; Table 2, Fig. 3).

As expected, when birds were excluded, the OS showed a higher reduction of seed set ($z = -8.5$, $p < 0.001$) and proportion of seedling emergence ($z = -7.2$, $p < 0.001$) than ES. Specifically, only insects set 1.4 times fewer seeds in *Cryptocarpus pyriformis*, 2.2 in *W. ovata*, and 2.6 in *O. echios* compared to the control seeds (birds and insects). Likewise, seedling emergence was reduced 1.5 times in
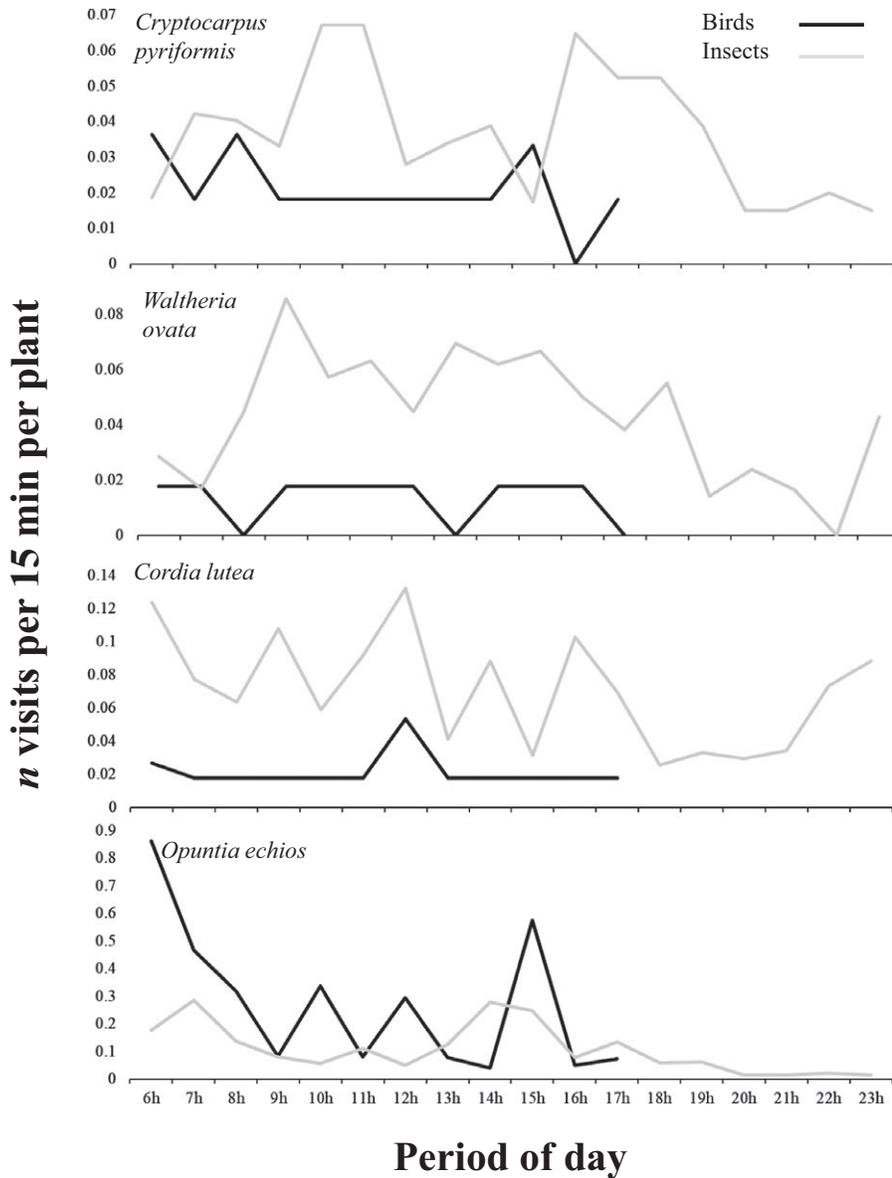
FIGURE 2. Variation in mean number of legitimate visits per 15 min per plant to flowers of *Cryptocarpus pyriformis*, *Waltheria ovata*, *Cordia lutea*, and *Opuntia echios*, performed by birds (black) and insects (gray), during two consecutive flowering periods in 2016 and 2017 on Santa Cruz, Galápagos.

C. pyriformis, 0.8 times in *W. ovata* and *Cordia lutea*, and 1.6 times in *O. echios* (Table 2 and Appendix S8).

Post hoc power analyses suggested that our lowest sample size had 100% (95% CI: 92.9–100.0) power to detect differences in *Cryptocarpus pyriformis*, *W. ovata*, and *Cordia lutea*, and 83.4% (95% CI: 74.4–96.3) in *O. echios* (all $p = 0.05$, 50 simulations).

DISCUSSION

The lower position occupied by birds in the quantitative pollination landscape indicates that they are quantitatively less effective pollinators than insects. However, insect species varied greatly in the quantitative component of pollination effectiveness with only *X.*

darwini and a few alien wasps and ants being relatively effective. Despite the low quantity of pollination service provided by opportunistic birds to entomophilous species, birds were unexpectedly important for effective reproduction. Thus, pollination systems on the Galápagos are probably more generalized than previous data suggested, opportunistic birds being relevant for both ES and OS within the flora of this archipelago.

Pollination effectiveness in entomophilous species

Our findings confirming that Galápagos birds act as effective pollinators are specially relevant on this archipelago where birds show a plastic feeding behavior visiting a variety of flowering plants (106 species, Traveset et al., 2015) and acting even as double mutualists for 13 species (Olesen et al., 2018). Birds can actually be potential pollinators of other ES, which apparently, seem to dominate the Galápagos flora (Stewart, 1911; McMullen, 1999). Previous studies in ES have reported exclusive pollination by insects, e.g., in *Aloe inconspicua* (Asphodelaceae) (Hargreaves et al., 2008) or hummingbirds and insects being equally effective as pollinators, e.g., in *Isertia laevis* (Rubiaceae) (Wolff et al., 2003). To the best of our knowledge, only one study documented opportunistic birds (European warblers) drinking flower nectar while on an island they used as a "rest stop" along their migration routes, showing they are more effective as pollinators than insects (Cecere et al., 2011). Our study, however, is the first to confirm pollination effectiveness by native opportunistic birds in entomophilous species.

Although the three ES were visited by six passerines in total, *G. fuliginosa* was the most important quantitatively. Interestingly, this endemic bird visits more plant species ($n = 68$), and is more generalized and abundant than any other passerine on the Galápagos (Dvorak et al., 2012; Traveset et al., 2015; Olesen et al., 2018). Thus, density compensation is likely to explain its highest visit frequency and, thus, greatest niche expansion of *G. fuliginosa* to include floral rewards. Previous studies have shown that variation in mutualist assemblages across years can lead to differences in the quantitative effectiveness landscape (Schupp et al., 2010 and references therein). We tested this hypothesis in the study plant species, but there were no interannual differences in number of visits and flowers contacted (see Appendix S9 for further details). The weak nectar dependence of opportunistic Galápagos birds and availability of alternative food resources, such as insects and fruits, could explain their reduced scores in the quantitative effectiveness landscape.

TABLE 2. Mean values ± SD of 11 variables used to assess the quality component of pollination effectiveness in *Cryptocarpus pyriformis*, *Waltheria ovata*, *Cordia lutea* and *Opuntia echios* by three selective exclusion experiments (control, bird exclusion, and autogamy treatments) on Santa Cruz, Galápagos, from March 2016 to April 2017.

		Fruit set	Seed set	Fruit length	Fruit width	Fruit mass	Seed length	Seed width	Seed mass	Seedling emergence	Time to seedling emergence	Seedling growth rate
Cryptocarpus pyriformis	C	0.54 ± 0.23	0.29 ± 0.26	2.43 ± 0.39	1.55 ± 0.34	0.76 ± 0.56	1.18 ± 0.23	0.81 ± 0.24	0.03 ± 0.01	29.00 ± 45.61	29.61 ± 10.61	0.10 ± 0.11
	E	0.58 ± 0.22	0.21 ± 0.20	2.06 ± 0.31	1.21 ± 0.28	0.59 ± 0.26	1.08 ± 0.22	0.71 ± 0.23	0.02 ± 0.01	19.00 ± 39.43	33.95 ± 8.81	0.13 ± 0.10
	A	0.58 ± 0.28	0.17 ± 0.16	1.95 ± 0.37	1.39 ± 0.97	0.53 ± 0.38	0.90 ± 0.22	0.52 ± 0.14	0.01 ± 0.01	1.10 ± 10.48	31.36 ± 10.06	0.08 ± NA
Waltheria ovata	C	0.57 ± 0.23	0.18 ± 0.15	4.39 ± 0.40	1.82 ± 0.25	0.76 ± 0.36	1.94 ± 0.23	1.4 ± 0.14	0.05 ± 0.02	32.00 ± 46.88	47.50 ± 8.35	0.03 ± 0.02
	E	0.55 ± 0.22	0.12 ± 0.10	4.04 ± 0.34	1.63 ± 0.28	0.52 ± 0.19	1.77 ± 0.23	1.11 ± 0.20	0.04 ± 0.03	40.00 ± 49.24	44.04 ± 8.60	0.03 ± 0.02
	A	0.41 ± 0.21	0.04 ± 0.07	3.91 ± 0.46	1.57 ± 0.21	0.35 ± 0.16	1.85 ± 0.35	1.09 ± 0.58	0.02 ± 0.02	7.35 ± 26.29	45.33 ± 28.01	0.02 ± 0.01
Cordia lutea	C	0.11 ± 0.12	1.00 ± NA	10.73 ± 1.56	12.85 ± 2.17	1.27 ± 0.61	8.29 ± 1.03	5.68 ± 1.00	0.17 ± 0.07	29.71 ± 45.86	50.2 ± 20.01	0.14 ± 0.12
	E	0.05 ± 0.10	100 ± NA	10.49 ± 1.34	12.51 ± 2.02	1.20 ± 0.50	7.68 ± 1.10	5.19 ± 0.88	0.14 ± 0.05	37.25 ± 48.59	42.47 ± 24.52	0.16 ± 0.16
	A	0.00 ± 0.04	100 ± NA	8.77 ± 4.31	9.89 ± 3.05	0.86 ± 0.43	7.98 ± 1.13	4.77 ± 0.87	0.11 ± 0.05	31.25 ± 47.87	42.20 ± 18.96	0.11 ± 0.07
Opuntia echios	C	1.00 ± NA	0.95 ± 0.22	52.36 ± 12.00	32.54 ± 6.67	18.98 ± 11.48	2.98 ± 0.57	2.45 ± 0.30	2.76 ± 2.42	32.00 ± 46.88	25.72 ± 4.78	0.01 ± 0.00
	E	100 ± NA	0.36 ± 0.48	48.57 ± 8.34	30.55 ± 6.33	15.13 ± 8.55	2.72 ± 0.29	2.32 ± 0.28	1.50 ± 1.63	20.00 ± 40.20	25.90 ± 7.17	0.01 ± 0.01
	NE	100 ± NA	0.85 ± 0.21	53.00 ± 9.12	31.33 ± 5.80	15.94 ± 11.25	2.57 ± 0.42	2.25 ± 0.37	1.97 ± 2.20	29.00 ± 45.60	21.79 ± 5.13	0.01 ± 0.01
	A	100 ± NA	0.91 ± 0.28	50.02 ± 9.45	27.20 ± 5.44	9.71 ± 7.11	2.50 ± 0.48	2.13 ± 0.42	2.30 ± 1.82	16.13 ± 37.39	22.00 ± 1.41	0.01 ± 0.00

The insect assemblage varied greatly in the quantity of pollination service provided to the study plants. Most insect species relied on *Cryptocarpus pyriformis*, *W. ovata*, and *Cordia lutea* only marginally, resulting in low visitation rates, whereas only a few species provided a relatively high quantity of pollination. Invasive wasps and ants were undoubtedly the most quantitatively important pollinators. We often observed these species touching the stamens and stigmas and walking on the anthers while visiting the flowers. This behavior suggests that they may act as pollinators, although further research is needed to properly test this hypothesis. It is also unknown whether they have negative effects on native pollinators because of competition for floral rewards. No evidence of asynchrony in the daily variation in number of visits was observed between insects and native birds (Fig. 2).

The analysis of the qualitative component of pollination effectiveness confirmed, however, that the three ES considerably improved reproductive success when both birds and insects visited the flowers. This finding supports that uncommon floral visitors can be efficient pollinators (reviewed in Fenster et al., 2004; King et al., 2013). Flowers visited by birds and insects produced larger and heavier fruits and more seeds than did those from which birds were excluded, suggesting that opportunistic birds provide an important pollination service. In addition, birds may enhance female fecundity through effective pollen flow because seedlings coming from control flowers of *Cryptocarpus pyriformis* germinated in higher percentages than those coming from either insect- or self-pollinated flowers (Fig. 3). This suggests that birds promote xenogamous crosses, what increases plant fitness. On the other hand, the differences observed between bird exclusion and autogamy treatments also suggest that insect pollinators are important to ensure the pollination of the study species, acting as a "fail-safe" system (sensu Aguilar-Rodríguez et al., 2016).

Autogamy showed to contribute little to fruit and seed production in all four study species, although it is another alternative to ensure reproduction, especially because of the scarcity of pollinators on the Galápagos (Linsley, 1966; Rick, 1966; Chamorro et al., 2012; Traveset et al., 2013). Hence, in self-compatible hermaphroditic species, we may expect a more relaxed selection by pollinators on floral traits and lower predictability of pollination syndromes than in self-incompatible species (Rosas-Guerrero et al., 2014). Pollination syndromes in insular species are probably less adjusted than on continents because species might have to widen their niches to maintain their populations, which was reflected, for instance, in their flowering throughout the year. Moreover, the lower predictability of pollination syndromes on islands might also be related to the higher densities of plant and opportunistic nectar-feeding species associated to low diversity levels (Gentry, 1988). Our findings suggest that the exclusion of birds results into a reduction of QLC, especially in the OS. But, with the available information so far, it is difficult to assess whether insects or birds represent the most important selective force on plant pollination traits. Because none of the ES is endemic, we do not expect different traits from their mainland relatives. However, it is possible that some traits (e.g., nectar) have evolved in response to selective pressures exerted by opportunistic nectarivorous birds living on the Galápagos. The possibility that opportunistic birds on islands exert a selective force strong enough to induce changes in the evolution of characters related
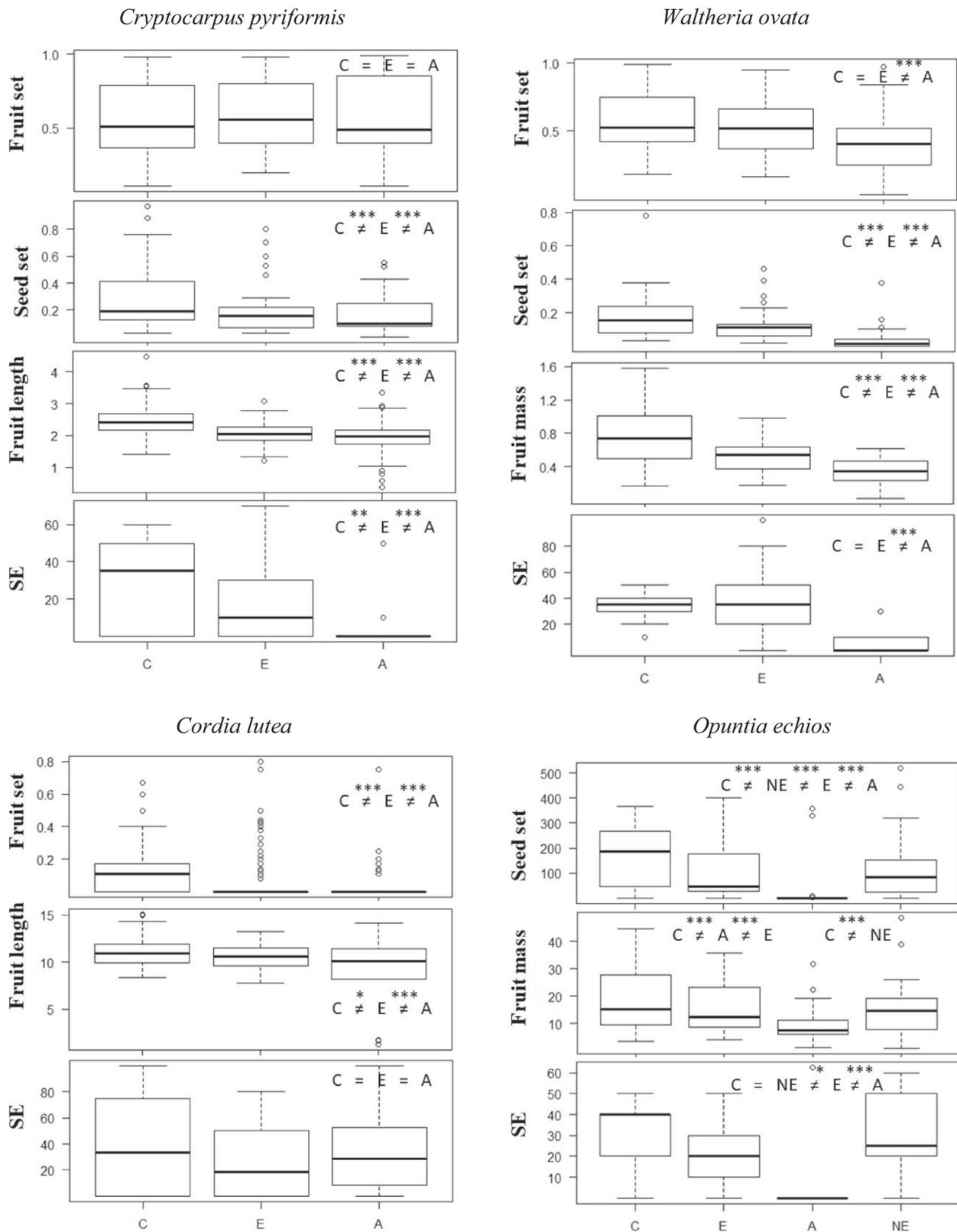
FIGURE 3. Mean values (± SD) of fruit set or seed set, fruit length or fruit mass, proportion of seedling emergence (SE) of each study species and treatment (C: control, E: bird exclusion, A: autogamy, NE: nocturnal exposure). Significant values of post-hoc comparisons: * $p = 0.01$; *** $p < 0.001$. All flowers of *Opuntia echios* produced fruits (100% fruit set), all fruits of *Cordia lutea* had seeds (100% seed set).

with plant-pollinator interactions certainly deserves further investigation.

Our exclusion experiments did not include an "only bird treatment" to disaggregate the QLC of birds from that of insects and thus to support that insects play a minor role in the reproduction of ES. An "only bird treatment", or flowers excluded after a single visit, would be needed to plot pollinator species in the two dimensional pollination effectiveness landscape and identify the most effective assemblage (Rodríguez-Rodríguez et al., 2013; Fernández de Castro et al., 2017; Schupp et al., 2017). Moreover, birds and insects have marked interspecific differences in life modes, which may affect the dynamics of pollen transfer within and between plants (Krauss et al., 2017). Nevertheless, the large reduction in seed set of *Cryptocarpus pyriformis* and *W. ovata*, and fruit set in the case of *Cordia lutea*, when birds are excluded, suggests that they may have evolved with birds as significant pollinators.

Pollination effectiveness in ornithophilous species

Both birds and insects were quantitatively effective pollinators of *O. echios*. Even though five bird species were identified as pollinators of this ornithophilous species, *G. scandens* was certainly the most important, although *G. fortis* and *G. fuliginosa* also provided relatively high quantity of pollination effectiveness. During the census, we observed *G. scandens* snipping the stamens and stigmas (37% of visits) and opening floral buds, as had previously been reported by Grant and Grant (1981). This behavior did not affect the flowers of our exclusion experiments, but it is known to prevent ovule fertilization and seed development (Grant and Grant, 1981).

Alien ants and the endemic *X. darwini* were quantitatively the most effective insect pollinators of *O. echios*. Bees were already suggested as the main pollinators of this species (Linsley, 1966; Rick, 1966). Our findings, however, are consistent with those of Grant and Grant (1981) on Daphne Major who documented a higher seed set when both birds and bees had access to the flowers than when bees were the sole visitors. These authors, nonetheless, recognized the possibility that bees visited covered flowers less often than uncovered ones. By means of camera traps, however, here we refused such possibility because no differences in insect visits were found between excluded and control flowers.

CONCLUSIONS

Despite the remarkable representation of ES and ecological generalization of island floras, and their depauperate pollinator faunas, very little is known about the role of opportunistic birds as legitimate pollinators on such species. Our study constitutes the first evidence that the native Galápagos birds are indeed effective pollinators of ES. Moreover, given the dominance of ES, we suggest that birds might have a key role maintaining the reproductive success and diversity of the Galápagos plant communities.

Previous evaluations of the pollination system of Galápagos plant species have disregarded the role of opportunistic birds. Because such birds are not strictly dependent on flower resources, a high insect or fruit availability may reduce plant visitation. However, our study revealed that the low flower visitation frequency by birds, compared to insects, did not preclude them from being effective pollinators. Therefore, future studies on the pollination system of Galápagos plant species should not simply rely on morphological floral traits, but should consider that opportunistic birds can also play an important role as selective factors as well as contributing to maintain plant reproductive success.

DATA ACCESSIBILITY

The data set used by this article is deposited into figshare at https://doi.org/10.6084/m9.figshare.6142373.

SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

LITERATURE CITED

Aguilar-Rodríguez, P., T. Krömer, J. García-Franco, and G. MacSwiney. 2016. From dusk till dawn: nocturnal and diurnal pollination in the epiphyte *Tillandsia heterophylla* (Bromeliaceae). *Plant Biology* 18: 37–45.

Anderson, S. H., D. Kelly, A. W. Robertson, and J. J. Ladley. 2016. Pollination by Birds. *In* Why Birds Matter: Avian Ecological Function and Ecosystem Services, 73. University of Chicago Press, Chicago.

Ballantyne, G., K. C. Baldock, and P. G. Willmer. 2015. Constructing more informative plant–pollinator networks: visitation and pollen deposition networks in a heathland plant community. *Proceedings of the Royal Society B* 282: 20151130.

Barrett, S. C., B. Emerson, and J. Mallet. 1996. The reproductive biology and genetics of island plants. *In* Evolution on Islands, 725–733. Oxford University Press, Oxford.

Carlquist, S. 1974. Island biology. Columbia University Press, New York.

Cecere, J., L. Cornara, F. Spina, S. Imperio, and L. Boitani. 2011. Birds outnumber insects in visiting *Brassica* flowers on Ventotene Island (central Mediterranean). *Life and Environment* 61: 145–150.

Chamorro, S., R. Heleno, J. M. Olesen, C. K. McMullen, and A. Traveset. 2012. Pollination patterns and plant breeding systems in the Galápagos: a review. *Annals of Botany* 110: 1489–1501.

De León, L., J. Podos, T. Gardezi, A. Herrel, and A. Hendry. 2014. Darwin's finches and their diet niches: the sympatric coexistence of imperfect generalists. *Journal of evolutionary biology* 27: 1093–1104.

Dvorak, M., B. Fessl, E. Nemeth, S. Kleindorfer, and S. Tebbich. 2012. Distribution and abundance of Darwin's finches and other land birds on Santa Cruz Island, Galápagos: evidence for declining populations. *Oryx* 46: 78–86.

Fenster, C. B., W. S. Armbruster, P. Wilson, M. R. Dudash, and J. D. Thomson. 2004. Pollination syndromes and floral specialization. *Annual review of ecology, evolution, and systematics* 35: 375–403.

Fernández de Castro, A. G., J. C. Moreno-Saiz, and J. Fuertes-Aguilar. 2017. Ornithophily for the nonspecialist: Differential pollination efficiency of the Macaronesian island paleoendemic *Navaea phoenicea* (Malvaceae) by generalist passerines. *American Journal of Botany* 104: 1556–1568.

Gentry, A. H. 1988. Changes in plant community diversity and floristic composition on environmental and geographical gradients. *Annals of the Missouri Botanical Garden* 75: 1–34.

Gorenflo, A., T. Diekötter, M. van Kleunen, V. Wolters, and F. Jauker. 2017. Contrasting pollination efficiency and effectiveness among flower visitors of *Malva sylvestris*, *Borago officinalis* and *Onobrychis viciifolia*. *Journal of Pollination Ecology* 21: 62–70.

Grant, B., and P. R. Grant. 1981. Exploitation of *Opuntia* cactus by birds on the Galápagos. *Oecologia* 49: 179–187.

Grant, P. R., and B. R. Grant. 2008. How and why species multiply: the radiation of Darwin's finches. Princeton University Press, Oxfordshire, UK.

Green, P., and C. J. MacLeod. 2016. SIMR: an R package for power analysis of generalized linear mixed models by simulation. *Methods in Ecology and Evolution* 7: 493–498.

Guézou, A., M. Trueman, C. E. Buddenhagen, S. Chamorro, A. M. Guerrero, P. Pozo, and R. Atkinson. 2010. An extensive alien plant inventory from the inhabited areas of Galápagos. *PLoS ONE* 5: e10276.

Hargreaves, A., L. Harder, and S. Johnson. 2008. *Aloe inconspicua*: the first record of an exclusively insect-pollinated aloe. *South African Journal of Botany* 74: 606–612.

Heleno, R. H., J. M. Olesen, M. Nogales, P. Vargas, and A. Traveset. 2013. Seed dispersal networks in the Galápagos and the consequences of alien plant invasions. *Proceedings of the Royal Society B* 280: 2012–2112.

Jordano, P. 2014. An R package for plots of effectiveness landscapes in mutualisms: effect.lndscp. Available at: https://github.com/pedroj/effectiveness_pckg [Accessed March 7, 2018].

Kelly, D., J. J. Ladley, A. W. Robertson, S. H. Anderson, D. M. Wotton, and S. K. Wiser. 2010. Mutualisms with the wreckage of an avifauna: the status of bird pollination and fruit-dispersal in New Zealand. *New Zealand Journal of Ecology* 34: 66.

King, C., G. Ballantyne, and P. G. Willmer. 2013. Why flower visitation is a poor proxy for pollination: measuring single-visit pollen deposition, with implications for pollination networks and conservation. *Methods in Ecology and Evolution* 4: 811–818.

Krauss, S. L., R. D. Phillips, J. D. Karron, S. D. Johnson, D. G. Roberts, and S. D. Hopper. 2017. Novel consequences of bird pollination for plant mating. *Trends in Plant Science* 22: 395–410.

Le Péchon, T., M. Sanchez, L. Humeau, L. D. Gigord, and L.-B. Zhang. 2013. Vertebrate pollination of the endemic *Trochetia granulata* (Malvaceae) on Réunion. *Journal of Tropical Ecology* 29: 353–356.

Linsley, E.G. 1966. Pollinating Insects of the Galápagos Islands. *In* The Galápagos Proceedings of the Symposia of the Galápagos International Scientific Project, University of California Press, Los Angeles.

Mayfield, M. M., N. M. Waser, and M. V. Price. 2001. Exploring the 'most effective pollinator principle' with complex flowers: bumblebees and *Ipomopsis aggregata*. *Annals of Botany* 88: 591–596.

McMullen, C. K. 1999. Flowering plants of the Galápagos. Cornell University Press, Ithaca, NY, USA.

McMullen, C. K. 2012. Pollination of the heterostylous Galápagos native, *Cordia lutea* (Boraginaceae). *Plant Systematics and Evolution* 298: 569–579.

Müller, H. 1881. Die Alpenblumen, ihre Befruchtung durch Insekten und ihre Anpassung an dieselbe. Wilhelm Englemann, Leipzig.

Ne'eman, G., A. Jürgens, L. Newstrom-Lloyd, S.G. Potts, and A. Dafni. 2010. A framework for comparing pollinator performance: effectiveness and efficiency. *Biological Reviews* 85: 435–451.

Olesen, J. M., C. F. Damgaard, F. Fuster, R. H. Heleno, M. Nogales, B. Rumeu, K. Trøjelsgaard, et al. 2018. Disclosing the double mutualist role of birds on Galápagos. *Scientific Reports* 8: 57.

Olesen, J. M., and A. Valido. 2003. Bird pollination in Madeira island. *Ardeola* 50: 67–69.

Ollerton, J., A. Killick, E. Lamborn, S. Watts, and M. Whiston. 2007. Multiple meanings and modes: on the many ways to be a generalist flower. *Taxon* 56: 717–728.

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

Ratto, F., B. I. Simmons, R. Spake, V. Zamora-Gutierrez, M. A. MacDonald, J. C. Merriman, C. J. Tremlett, et al. 2018. Global importance of vertebrate pollinators for plant reproductive success: a meta-analysis. *Frontiers in Ecology and the Environment* 16: 82–90.

Rick, C.M. 1966. Some Plant-Animal Relations on the Galápagos Islands. *In* The Galápagos Proceedings of the Symposia of the Galápagos International Scientific Project, University of California Press, Los Angeles.

Rodríguez-Rodríguez, M. C., and A. Valido. 2008. Opportunistic nectar-feeding birds are effective pollinators of bird-flowers from Canary Islands: experimental evidence from *Isoplexis canariensis* (Scrophulariaceae). *American Journal of Botany* 95: 1408–1415.

Rodríguez-Rodríguez, M. C., and A. Valido. 2011. Consequences of plant-pollinator and floral-herbivore interactions on the reproductive success of the Canary Islands endemic *Canarina canariensis* (Campanulaceae). *American Journal of Botany* 98: 1465–1474.

Rodríguez-Rodríguez, M. C., P. Jordano, and A. Valido. 2013. Quantity and quality components of effectiveness in insular pollinator assemblages. *Oecologia* 173: 179–190.

Rosas-Guerrero, V., R. Aguilar, S. Martén-Rodríguez, L. Ashworth, M. Lopezaraiza-Mikel, J. M. Bastida, and M. Quesada. 2014. A quantitative review of pollination syndromes: do floral traits predict effective pollinators? *Ecology Letters* 17: 388–400.

Schupp, E. W., P. Jordano, and J. M. Gómez. 2010. Seed dispersal effectiveness revisited: a conceptual review. *New Phytologist* 188: 333–353.

Schupp, E. W., P. Jordano, and J. M. Gómez. 2017. A general framework for effectiveness concepts in mutualisms. *Ecology Letters* 20: 577–590.

Stewart, A. 1911. A botanical survey of the Galápagos Islands. *Proceedings of the California Academy of Science* 4: 7–228.

Traveset, A., R. Heleno, S. Chamorro, P. Vargas, C. K. McMullen, R. Castro-Urgal, M. Nogales, et al. 2013. Invaders of pollination networks in the Galápagos Islands: emergence of novel communities. *Proceedings of the Royal Society of London B: Biological Sciences* 280: 20123040.

Traveset, A., J. M. Olesen, M. Nogales, P. Vargas, P. Jaramillo, E. Antolín, M. M. Trigo, and R. Heleno. 2015. Bird-flower visitation networks in the Galápagos unveil a widespread interaction release. *Nature Communications* 6: 6376.

Traveset, A., C. Tur, K. Trøjelsgaard, R. Heleno, R. Castro-Urgal, and J. M. Olesen. 2016. Global patterns of mainland and insular pollination networks. *Global Ecology and Biogeography* 25: 880–890.

Trueman, M., and N. d'Ozouville. 2010. Characterizing the Galápagos terrestrial climate in the face of global climate change. *Galápagos Research* 67: 26–37.

Valido, A., Y. L. Dupont, and J. M. Olesen. 2004. Bird–flower interactions in the Macaronesian islands. *Journal of Biogeography* 31: 1945–1953.

Wiggins, I. L., and D. M. Porter. 1971. Flora of the Galápagos Islands. Stanford University Press, Stanford, California.

Wolff, D., M. Braun, and S. Liede. 2003. Nocturnal versus diurnal pollination success in *Isertia laevis* (Rubiaceae): a sphingophilous plant visited by hummingbirds. *Plant Biology* 5: 71–78.