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Complex inter-island colonization and peripatric founder speciation promote diversification of flightless *Pachyrhynchus* weevils in the Taiwan–Luzon volcanic belt

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Abstract

Aim: We investigated the spatial and temporal patterns of diversification among colourful and flightless weevils, the *Pachyrhynchus orbifer* complex, to test the stepping-stone hypothesis of colonization across the Taiwan–Luzon volcanic belt.

Location: Southeast Asia.

Methods: The phylogeny of the *P. orbifer* complex was reconstructed from a multilocus data set of mitochondrial and nuclear genes using maximum likelihood in RAxML and Bayesian inference in MRBAYES. Likelihood-based tests in CONSEL were used to evaluate alternative tree topologies. Divergence times were estimated in BEAST based on a range of mutation rates. Ancestral range and biogeographical history were reconstructed using Bayesian binary MCMC (BBM) methods in RASP and in BioGeoBEARS. Demographic histories were inferred using the extended Bayesian skyline plot (EBSP). Species boundaries were tested using BPP.

Results: The phylogeny of the *P. orbifer* complex indicated strong support for seven reciprocally monophyletic lineages grouped by current island boundaries (Camiguin, Fuga, Dalupiri, Calayan, Babuyan, Orchid and Yaeyama Islands), except for a sister Green + Itbayat lineage. Complex and stochastic colonization of *P. orbifer* was inferred to have involved both northward and southward directions with short- and long-distance dispersal events, which are strongly inconsistent with the strict stepping-stone hypothesis. Divergence time estimates for all extant island lineages (<1 Myr of Middle Pleistocene) are much more recent than the geological ages (22.4–1.7 Myr) and subaerial existence (*c.* 3 Myr) of the islands. The statistically delimited seven cryptic species imply that the diversity of *Pachyrhynchus* from small peripheral islands continues to be largely under-estimated.

Main conclusions: The non-linear, more complex spatial and temporal settings of the archipelago and stochastic dispersal were probable key factors shaping the colonization history of the *P. orbifer* complex. Speciation of the *P. orbifer* complex may have occurred only between islands, indicating that peripatric speciation through the founders of stochastic dispersals was the major evolutionary driver.

KEYWORDS
cryptic species, Kuroshio current, long-distance dispersal, oceanic islands, stepping-stone hypothesis, the Philippines, weevils

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1 | INTRODUCTION

Islands are excellent natural systems to study the diversification of organisms because of their range of ages, sizes and well-studied geology (Carson, 1983; Losos & Ricklefs, 2009). Effective overwater dispersal and subsequent successful establishment in novel habitats are fundamental processes in generating endemic island biodiversity (MacArthur & Wilson, 1967; Whittaker, Triantis, & Ladle, 2008). The colonization of remote oceanic islands by continental species is mainly an interaction between the relative isolation of islands and the vagility of the organisms (Gillespie & Roderick, 2002). Therefore, more remote islands usually have fewer colonizers than nearer islands, and vagile organisms often colonize more islands than their less mobile counterparts. Colonization of remote oceanic island also depends on the available ecological space, with the most successful colonization occurring in the youngest islands within an isolated archipelago (Whittaker & Fernández-Palacios, 2007).

Inter-island dispersal in an isolated oceanic archipelago may produce a highly concordant colonization pattern consistent with the chronological sequence of the origin of its islands (The progression rule; Funk & Wagner, 1995; Juan, Emerson, Oromí, & Hewitt, 2000; Shaw & Gillespie, 2016). The "stepping-stone by island age model" of asymmetric colonization from geologically older to younger islands is one of the common modes of colonization in several well-documented archipelagos, showing a clear linear age sequence of the origin of islands, such as the Hawaiian and Galápagos Islands in the Pacific Ocean (Funk & Wagner, 1995; Gillespie, 2004; Parent, Caccone, & Petren, 2008) and the Canary Islands in the Atlantic (Juan et al., 2000; Planas & Ribera, 2014). However, some observations of stochastic dispersal have suggested that the unpredictability of inter-island colonization can be linked to the geological history of the islands in a more complex manner (Gillespie et al., 2012). The pattern of long-distance dispersal across oceanic barriers, which was previously perceived as a rare and almost impossible event, may be equally probable as stepwise dispersal across islands given that "non-standard" dispersal mechanisms such as strong storms and ocean currents often drive these complex and highly stochastic processes (Nathan, 2006).

Situated in a unique biogeographical junction between the Asian and Philippine faunal regions known as the extension of Kano's Line ("Neo-Wallace Line," Kano, 1941), the Taiwan–Luzon volcanic belt (Figure 1a) is currently one of the least studied groups of oceanic islands. Endemic species of the Taiwan–Luzon volcanic belt were hypothesized to have originated through northward stepping-stone dispersal from observations of a decreasing number of shared Luzon species when moving from southern to northern islands (Oliveros, Ota, Crombie, & Brown, 2011; Ota & Huang, 2000). This northward "stepping-stone by island distance" model of colonization may have been facilitated by the regionally dominant surface Kuroshio current, which originates at the equator and flows northerly towards the Philippines, Taiwan–Luzon volcanic belt and Japanese islands. The Kuroshio current is thought to be a strong oceanic mechanism in shaping the distribution of tropical sea grasses (Kuo, Kanamoto, Iizumi, Aioi, & Mukai, 2006) and population genetic structures of skinks in the Ryukyu Archipelago (Kurita & Hikida, 2014). However, the stepping-stone model of colonization in the Taiwan–Luzon volcanic belt has never been rigorously tested using a molecular phylogeographical approach, with the exception of some phylogenetic studies of shrews (Esselstyn & Oliveros, 2010), birds (Oliveros & Moyle, 2010) and geckos (Siler, Oaks, Cobb, Ota, & Brown, 2014), which indicated a direction of colonization from either Northern Taiwan (shrews) and Batanes (birds) or Southern Luzon (geckos). Nevertheless, these earlier studies were limited by representatives from only a few islands across the Taiwan–Luzon volcanic belt to fully address the direction and stepwise pattern of the stepping-stone hypothesis.

The Taiwan–Luzon volcanic belt consists of a group of remote volcanic islands adjacent to the continental shelf of mainland Asia (Figure 1a). This archipelago contains two parallel island chains (Eastern and Western) in a north–south direction, which were all created de novo, without any connections to nearby larger islands (Luzon and Taiwan) or the Asian mainland since their emergence (Voris, 2000). The deep ocean channels surrounding these islands separate them from neighbouring Luzon by >25 km to the south (Babuyan Islands), and >20 km from the continental island of Taiwan to the west (Green Island). The Balintang Channel between the Babuyan and Batanes Islands is *c*. 75 km wide. North of Batanes Islands, approximately 150 km across the Bashi Channel, lies Orchid Island. Although the two island chains are separated by only 50 km just North of Luzon and merge into a single island chain near Batan, most islands in the Western chain (Orchid, Itbayat, Sabtang, Calayan, Dalupiri and Fuga) are much older and mainly composed of volcanic rocks of the Miocene to Pliocene age (>3.5 Myr) (Figure 1c) (Yang et al., 1996). In contrast, all of the currently active volcanoes are in the Eastern chain (Green, Babuyan, Batan, Camiguin), where the majority of islands are younger and have origins in the Quaternary (<2.8 Myr) (Figure 1c) (Osozawa et al., 2012; Yang et al., 1996). Most of the islands of the Taiwan–Luzon volcanic belt were not connected to each other or to the Asian mainland during the Pleistocene glacial period (Voris, 2000), minimizing the effect of recent climatic changes on regional phylogeographical patterns (e.g. Nakamura et al., 2014). The current elevation of these islands ranges from several hundred metres in most islands to over a thousand metres in Batan (Yang et al., 1996) (Figure 1a). They were likely subaerial during the high sea levels of the Pleistocene interglacial period (Voris, 2000). Within each island chain, there is a general chronological order of island origins beginning from the older southern islands towards the younger northern islands (Yang et al., 1996) (Figure 1a, c). However, the geomorphologic evidence indicates that most islands of the Taiwan–Luzon volcanic belt were submarine and only uplifted above sea level *c*. 3 Ma (Yang et al., 1996). Therefore, for islands older than 3 Myr, the chronological sequence of emergence may not necessarily follow a south–north direction of geographical arrangement.

We set out specifically to test the stepping-stone model of colonization in a group of endemic weevils, the *Pachyrhynchus orbifer*

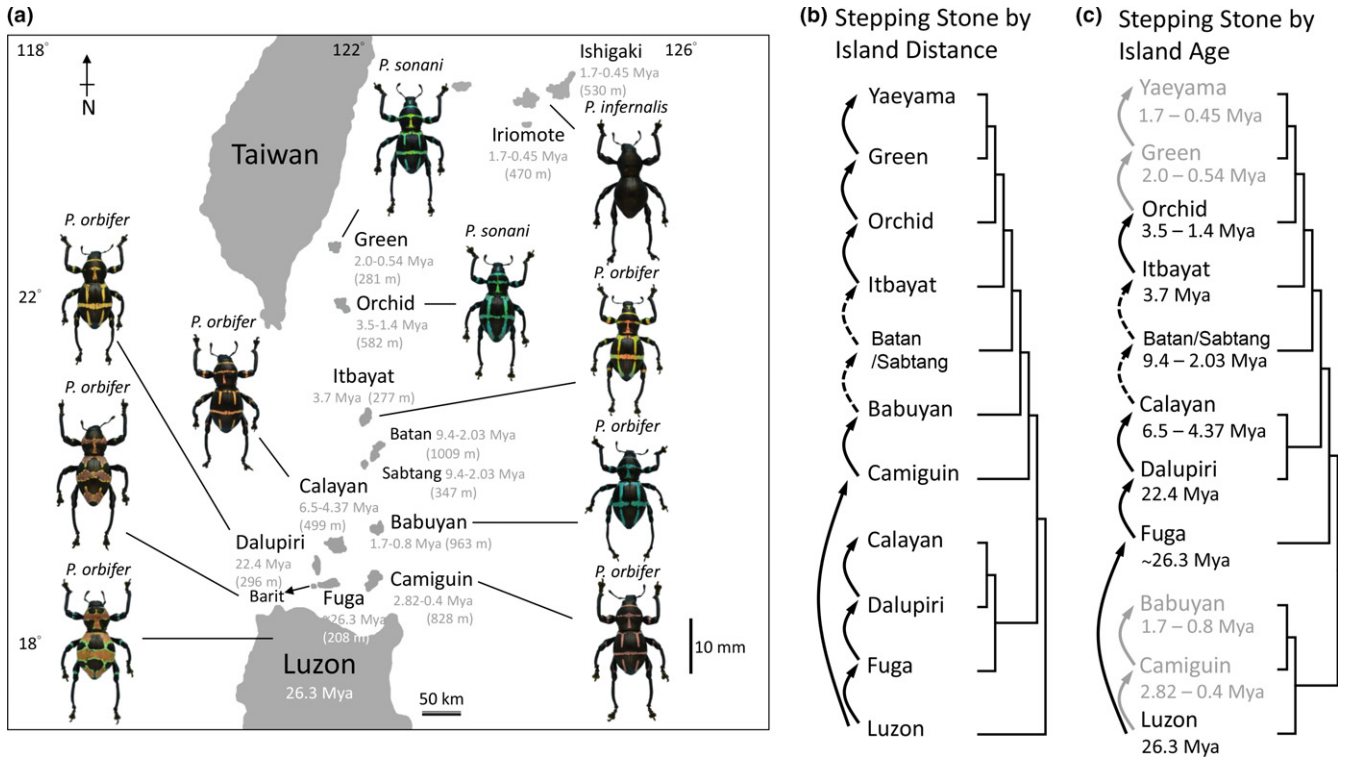

FIGURE 1 (a) Map of the Taiwan–Luzon volcanic belt showing the elevation of the islands (Yang et al., 1996 and University of Texas Libraries, http://www.lib.utexas.edu/maps/ams/japan/) and representative species of the *Pachyrhynchus orbifer* complex in the Taiwan–Luzon volcanic belt. (b) Predicted tree topology derived from the hypothesis of stepping-stone by island distance. (c) Predicted tree topology derived from the hypothesis of stepping-stone by island age (geological ages derived from Yang et al., 1996 and Osozawa et al., 2012). The islands of the Taiwan–Luzon volcanic belt only became subaerial *c*. 3 Ma (Yang et al., 1996). Solid arrows are predicted directions of colonization; dotted arrows are unobserved colonization events of the islands currently with no *P. orbifer*. Grey arrows are predicted colonization events following the emergence of the islands (progression rule) [Colour figure can be viewed at wileyonlinelibrary.com]

Waterhouse, 1841 complex (Insecta: Coleoptera: Curculionidae), from the Taiwan–Luzon volcanic belt. *Pachyrhynchus* is a group of plant-feeding insects decorated with aposematic colours and with a large number of endemic species in the Philippine Archipelago (Schultze, 1923; Tseng, Lin, Hsu, Pike, & Huang, 2014). The *P. orbifer* complex represents an excellent set of organisms to test the stepping-stone hypothesis, not only because of the existence of closely related endemic species on nearly every sizable island across the archipelago, but also due to three intriguing adaptations that may facilitate their cross-ocean dispersal through rafting on floating vegetation by oceanic currents: (1) *Pachyrhynchus* adults are wingless and have completely fused elytra; (2) they have the ability to draw and keep air under their elytra, allowing them to float on water for at least several hours (Schultze, 1923); (3) their eggs are inserted into plant tissues, and the larvae live and feed inside the stems of the host plants. Recent studies suggested that flightless weevils were able to colonize remote archipelagos of Southeast Asia and the Pacific via stepping-stone islands (Claridge, Gillespie, Brewer, & Roderick, 2017; Machado, Rodríguez-Expósito, López, & Hernández, 2017; Tänzler et al., 2016).

We reconstructed the phylogeny, colonization history, divergence times and historical demography of the *P. orbifer* complex using a multi-locus data set to test the following specific predictions from stepping-stone hypotheses by island distance (Figure 1b) and by island age (Figure 1c). (1) The hypothesis of stepping-stone by island distance predicts that the topology of the phylogeny and inferred colonization history are consistent with an asymmetric colonization from the nearest southern islands (Fuga and Camiguin) to the farthest northern islands (Yaeyama) (Figure 1b, solid arrows), which may have been promoted by the northerly flowing Kuroshio current. Because the Taiwan–Luzon volcanic belt is a double arc island chain, the weevils were expected to disperse northwards initially from Northern Luzon to Fuga of the western arc and Camiguin of the eastern arc, and then either to colonize Dalupiri and Calayan from Fuga of the western arc, or to colonize Babuyan, Itbayat, Orchid, Green and Yaeyama Islands in sequence. Conversely, the age progression from older to younger islands only occurred in the Luzon/Camiguin/Babuyan and in the Orchid/Green/Yaeyama Islands (Figure 1c, grey arrows) because all the other geologically older islands became subaerial *c*. 3 Ma. The remaining tree topologies predicted by age progression (Figure 1c, solid arrows) are the same and cannot be distinguished from those predicted by island distance (Figure 1b). (2) If the colonization was initiated sequentially from southern to northern islands by geographical distance, then we predicted that the divergence times of species from the

southern islands were longer than those of the species from their neighbouring northern islands in sequence (Figure 1b, c). If the colonization occurred via the progression of island age, we predicted that the divergence time of species in the Luzon/Camiguin/Babuyan and Orchid/Green/Yaeyama Islands were near the time when these islands emerged (Figure 1c, grey arrows). (3) If more ancient colonization occurred in the southern (Figure 1b) or older (Figure 1c) islands, species on these island were predicted to have a history of a stable or declining population size, whereas species from the northern (Figure 1b) or younger (Figure 1c) islands showed a demographic expansion following a population bottleneck (recent colonization).

Collecting trips to the Batanes and Babuyan Islands revealed island populations of *P. orbifer* that displayed similar stripe patterns, but different colours, in extant species (Figure 1a). The extensive and characteristic stripe and colour variation of endemic island populations suggest that the widespread *P. orbifer* (Schultze, 1923) might comprise a number of cryptic species, necessitating closer investigation of its species boundary. A recent study of *P. sonani* from Orchid and Green islands based on combining morphological, molecular and ecological data supported two distinct species by island origin (Chen et al., 2017). Therefore, we used coalescent-based species delimitation methods to test the hypothesis that each island population of the *P. orbifer* complex forms a distinct species.

2 | MATERIALS AND METHODS

2.1 | Taxon sampling

Pachyrhynchus weevils were collected from 11 locations between Northern Luzon and Southern Ryukyus, including the Cagayan Province (Claveria), Babuyan Islands (Camiguin, Fuga, Dalupiri, Calayan and Babuyan), Batanes Islands (Itbayat), Orchid Island, Green Island and Yaeyama Islands (Ishigaki and Iriomote) (Figure 1a, Appendix S1 in supporting information). Our samples comprised three known (Northern Luzon, Batanes and Fuga Island) (Kano, 1936; Schultze, 1923) and four newly recorded populations of *P. orbifer* Waterhouse, 1841 and closely related species (*P. sonani* Kono, 1930 and *P. infernalis* Kano, 1897). Specimens of closely related *P. speciosus* Waterhouse, 1841 (Samar Island), *P. tobafolius* Kano, 1929 (Orchid Island) and *P. nobilis yamianus* Kano, 1929 (Orchid Island) were used as outgroups for the phylogenetic analyses.

2.2 | DNA extraction and sequencing

Genomic DNA was extracted from the legs or thoracic muscles (DNA Mini Kit, Favorgen Biotech, Taiwan). The DNA fragments of two mitochondrial (cytochrome c oxidase subunit 1, *CO1*; NADH dehydrogenase subunit 2, *ND2*) and two nuclear genes (elongation factor 1-α, *EF1*-α; ribosomal internal transcribed spacer, *ITS*) were amplified and sequenced using a combination of newly designed and published primers (PCR, sequencing, sequence editing and alignment, Appendix S2).

2.3 | Phylogenetic analyses and topology test

Phylogenetic relationships were reconstructed using maximum likelihood (ML) in RAxML-VI-HPC (Stamatakis, 2006) and Bayesian inference (BI) in MRBAYES 3.2.2 (Ronquist et al., 2012) (Appendix S3). Phylogenetic data were deposited in the Dryad Digital Repository (https://doi.org/10.5061/dryad.25fc4). We evaluated the topological prediction (Figure 1b) using likelihood-based methods in CONSEL 0.1i (Shimodaira & Hasegawa, 2001). Per site log likelihoods of alternative tree topologies (constrained topology of the stepping-stone hypothesis, Figure 1b versus the topology of our best tree, Figure 2) were estimated from ML heuristic searches of 1,000 generations using RAxML-VI-HPC (Stamatakis, 2006). The *p*-value of the tests was calculated with 10,000 bootstrap replicates. The topological hypotheses with a p-value $< .05$ were rejected.

2.4 | Divergence time estimation

The divergent times were estimated in BEAST 1.8.2 (Drummond, Suchard, Xie, & Rambaut, 2012), based on an uncorrelated lognormal relaxed clock model. The substitution models were unlinked among gene partitions. The upper and lower bounds of the mutation rate in mitochondrial DNA (mtDNA) were applied, based on the standard (0.0115 mutations site^{-1} Myr^{-1}; Brower, 1994) and a revised molecular clock for insects (0.0177 mutations site^{-1} Myr^{-1}; Papadopoulou, Anastasiou, & Vogler, 2010), with the caveat that the rates of molecular evolution might be accelerated in flightless weevils (Tänzler et al., 2016). The substitution rate of *EF1*-α and *ITS* followed the mutation rate of synonymous substitution for nuclear genes (0.016 mutations site^{-1} Myr^{-1}; Moriyama & Gojobori, 1992) (Appendix S4).

2.5 | Ancestral range and historical dispersal

The ancestral range and biogeographical history were reconstructed using Bayesian binary Markov chain Monte Carlo (MCMC) (BBM) (Ronquist & Huelsenbeck, 2003) methods implemented in RASP 3.2 (Yu, Harris, Blair, & He, 2015) and a R package, "BioGeoBEARS" (Matzke, 2013a). The analysis used the best trees generated from the BI analyses (Figure 2), with the range of the specimens coded as the island of origin (Appendix S6).

2.6 | Population demographic history and species delimitation

The demographic histories of weevils on 10 islands (Claveria, Camiguin, Fuga, Dalupiri, Calayan, Babuyan, Itbayat, Orchid, Green and Yaeyama Islands) were inferred from multi-locus data using coalescent-based extended Bayesian skyline plot (EBSP) (Heled & Drummond, 2008) implemented in BEAST 1.8.2 (Drummond et al., 2012) (Appendix S8). The species boundaries were estimated using the multi-species coalescent model (MSC) and reversible-jump Markov chain Monte Carlo (rjMCMC) algorithms in BPP 3.1 (Yang, 2015) (Appendix S7).

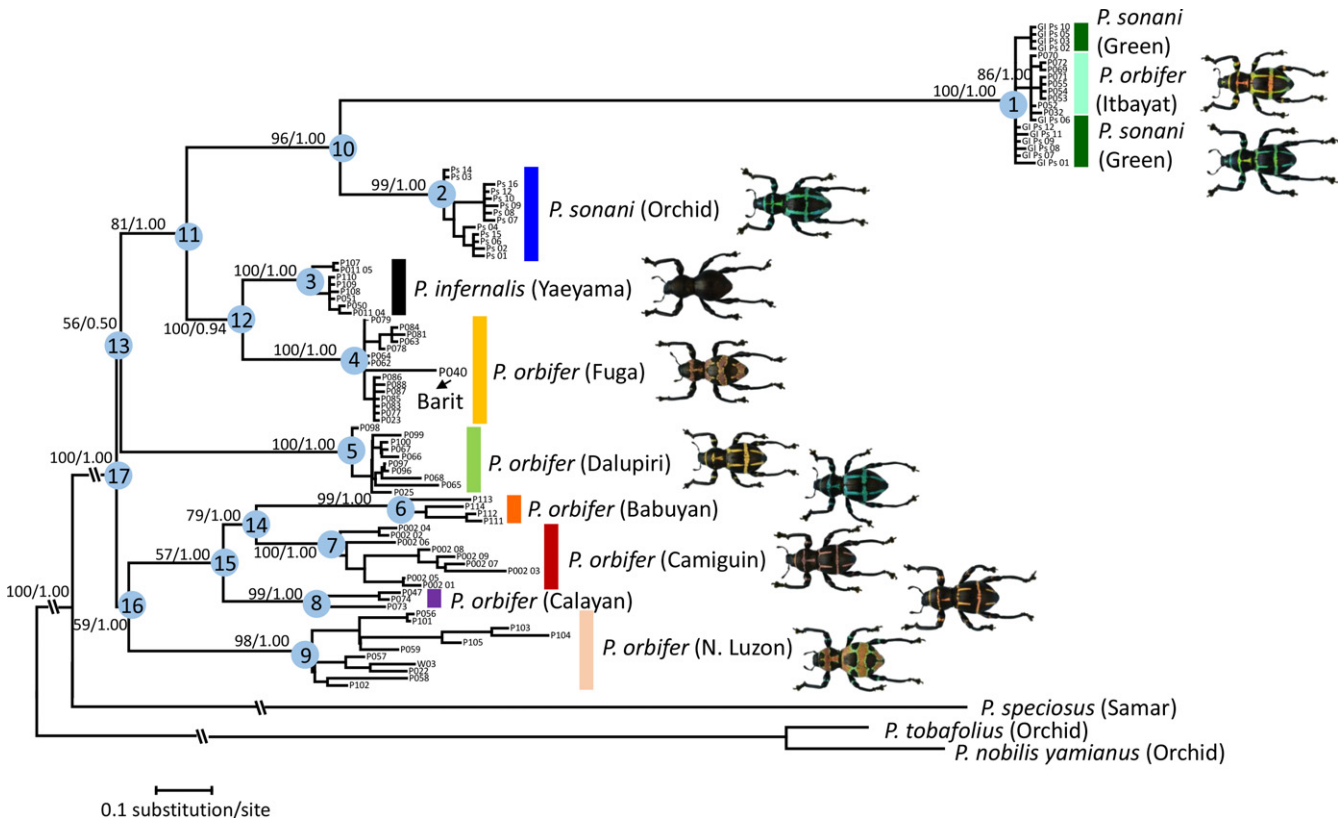

FIGURE 2 Phylogeny of *Pachyrhynchus orbifer* complex of the Taiwan–Luzon volcanic belt reconstructed from the combined data using maximum likelihood (ML) analyses of 1,000 bootstrap replicates of the rapid hill-climbing algorithm in RAxML-VI-HPC and Bayesian inference (BI) of 1 × 10^8 generations of Markov chain Monte Carlo (MCMC) processes in MRBAYES v3.2.2. Numbers near the nodes are branch support values of the likelihood bootstrap (LB)/Bayesian posterior probability (BPP). Numbers in blue dots are node numbers of the tree [Colour figure can be viewed at wileyonlinelibrary.com]

3 | RESULTS

3.1 | Phylogeny and topological test of the stepping-stone hypothesis

A sequence matrix of 4,050 base pairs was obtained from 93 ingroup and three outgroup taxa. The *CO1*, *ND2* and *ITS* gene trees all resulted in well-resolved phylogenetic relationships, with the majority of the lineages clustered according to the island of origin (Appendix S5). In contrast, the *EF1-α* gene tree showed only a few clusterings by island origins (Green + Itbayat, Dalupiri and Fuga) but limited phylogenetic structure among island lineages. The phylogeny of the combined data indicated a well-resolved tree with strong branch supports (LB/BPP > 99%/1.0) for seven reciprocally monophyletic lineages grouped by current island boundaries (Camiguin, Fuga, Dalupiri, Calayan, Babuyan, Orchid and Yaeyama Islands), except for a sister Green + Itbayat lineage (Figure 2, node 1). Because the island populations were reciprocally monophyletic, we also reconstructed the species tree using coalescence-based analyses in *BEAST (Appendix S4). The majority of the sister relationships of the species tree were similar to that of the tree inferred from concatenated data, with the inconsistency of tree topologies that are not well supported. Maximum likelihood topological tests (AU, KH, SH, WKH and WSH) of phylogenetic hypotheses all significantly supported the topology of the best tree (Table 1; Figure 2) and rejected the tree topology derived from the two stepping-stone hypotheses ($p < .05$; Figure 1b, c).

3.2 | Divergence time and colonization history

The divergence time estimation provided a mean estimate of *c.* 0.91 Ma [95% confidence interval (CI): 0.61–1.31 Ma] for the origin of the *P. orbifer* complex (Figure 3, node 17). The most recent common ancestors (MRCA) of all nine extant island lineages of the *P. orbifer* complex (Figure 3, node 1–8 and 18) fell within the timeframe of the late Ionian to Tarantian (in the late Pleistocene) between 0.27 and 0.05 Ma (Figure 3, node 7, 95% CI: 0.12–0.48 Ma; node 18, 95% CI: 0.01–0.12 Ma), with the majority of island lineages originating *c.* 0.2 Ma. The youngest island lineage was found only *c.* 50,000 years ago in Itbayat (Figure 3, node 18). The majority of the southern island lineages (Calayan, Camiguin, Babuyan and Dalupiri, but not Fuga) were older than that of their northern counterparts (Itbayat, Green and Yaeyama Islands), except for Orchid Island among the older island lineages (Figure 3). BioGeoBEARS analyses showed that the best-fit model was DIVA-LIKE + J (Ln*L* = −37.6159, AIC = 81.2317) (Table 2). This model inferred a

TABLE 1 Maximum likelihood values and statistics calculated in RAxML-VI-HPC and CONSEL for alternative tree topologies of the stepping-stone hypothesis (Figure 1b, c) and the best phylogenetic tree of the *Pachyrhynchus orbifer* complex in the Taiwan–Luzon volcanic belt (Figure 2)

Hypotheses	Likelihood	AU	KH	SH	WKH	WSH
Stepping-stone by island distance	−12341.086	1e−011	0	0	0	0
Stepping-stone by island age	−12305.422	1e−015	0	0	0	0
Best tree	−11815.909	1	1	1	1	1

AU, approximately unbiased test; KH, Kishino–Hasegawa test; SH, Shimodaira–Hasegawa test; WKH, weighted Kishino–Hasegawa test; and WSH, weighted Shimodaira–Hasegawa test.

process of combining dispersal, extinction and peripatric events for diversification among extant islands, and highlighted the essential process of founder speciation for generating endemic diversity in these oceanic islands. Ancestral range reconstruction of BBM and DIVA-LIKE + J provided comparable results in recently diverged nodes (Figure 3). Among deeper nodes, only BBM distinguished higher probable ancestral areas, whereas the DIVA-LIKE + J model showed ambiguous reconstruction (Figure 3). We therefore interpreted the colonization history based on the results of the BBM analyses. Two major historical colonization routes were identified for the *P. orbifer* complex (Figure 4, solid versus open arrows); the first route (solid arrows) suggested that the ancestral *P. orbifer* probably originated from Northern Luzon [given an equally probable ancestral range of Luzon (32.4%) and Dalupiri (32.4%)] and then first split and moved to Dalupiri Island *c.* 0.84 Ma (Figure 3, node 13, 95% CI: 0.56–1.22 Ma; Figure 4a, dispersal 1). The descendent lineage of Dalupiri Island dispersed remotely to Northern Yaeyama Island (Figure 3, node 11, Yaeyama: 20.19%) *c.* 0.76 Ma (95% CI: 0.49–1.10 Ma; Figure 4a, dispersal 3), then spread southward to Green Island (Figure 3, node 10, Green: 47.9%, Orchid: 40.21%) *c.* 0.53 Ma (95% CI: 0.34–0.80 Ma; Figure 4b, dispersal 5) and later travelled a long distance from Yaeyama southward to Fuga Island (Figure 3, node 12, Fuga: 45.75%) *c.* 0.47 Ma (Figure 3, node 12, 95% CI: 0.23–0.78 Ma; Figure 4c, dispersal 7). The ancestral lineage of Orchid Island originated from Green Island (Figure 3, node 2, Green: 99.7%) *c.* 0.23 Ma (Figure 3, node 2, 95% CI: 0.08–0.46 Ma; Figure 4d, dispersal 8), and later the ancestral Green Island lineage dispersed southward towards Itbayat Island only 50,000 years ago (Figure 3, node 18; Figure 4d, dispersal 9). The second colonization route (open arrows) indicated that the ancestral *P. orbifer* dispersed from Northern Luzon to Calayan Island (Figure 3, node 16, Northern Luzon: 71.64%; node 15, Calayan: 67.09%) *c.* 0.76 Ma (Figure 3, node 16, 95% CI: 0.49–1.14 Ma; Figure 4a, dispersal 2), subsequently colonized nearby Camiguin Island (Figure 3, node 15, Camiguin 41.68% and Babuyan 41.55%) *c.* 0.64 Ma (Figure 3, node 15, 95% CI: 0.39–1.00 Ma; Figure 4b, dispersal 4), and later spread to the neighbouring Babuyan Island nearly 0.50 Ma (Figure 3, node 14, 95% CI: 0.27–0.80 Ma; Figure 4c, dispersal 6). We inferred alternative colonization histories to assess the robustness of our interpretation of the data (Appendix S6). The eight alternative colonization scenarios contained on average of five long distance (between non-neighbouring islands) and four short-distance dispersals (between neighbouring islands), and four northward and five southward events.

3.3 | Species status and demographic history

The BPP species delimitation suggested a total of 10 species corresponding to current island boundaries (Figure 3, nodes marked with an asterisk) and supported by high posterior probability (0.99–1.0; Appendix S7). The results indicate that previously recognized *P. sonani* might consist of two cryptic species that originated from Green and Orchid Island (Figure 3, nodes 1 and 2) (Chen et al., 2017). Other lineages with similar colour stripes from neighbouring (N. Luzon versus Fuga; Camiguin versus Calayan) and distant islands (Babuyan versus Green/Orchid) were each assigned to putative species (Figure 3; Appendix S7). EBSP analyses suggested that the 95% HPD of the number of demographic population size changes included zero in nine island populations (except Claveria of N. Luzon; Appendix S8), indicating that historical sizes of these populations largely remained constant during the time period. The population of *P. orbifer* from Northern Luzon underwent recent population growth starting *c.* 0.1 Ma (Appendix S8). The populations of Green, Itbayat, Babuyan, Camiguin and Dalupiri Islands had the highest frequency of one population size change, suggesting only a minor trend towards a recent increase in the size of these populations.

4 | DISCUSSION

4.1 | Rejecting the strict stepping-stone hypothesis

Our analyses show that the divergence time of all extant island lineages of the *P. orbifer* complex fall within the last 1 Myr. This timeframe of diversification is much more recent than the geological age of the islands (average: 22.4–1.7 Myr) and the subaerial existence of most older islands (*c.* 3 Myr). Because all the islands likely have been present when the weevils first colonized; therefore, we did not expect the weevils to follow the island age progression. Indeed, the results of divergence time showed that the colonization of the *P. orbifer* complex did not follow the island age progression. The tree topology of the *P. orbifer* phylogeny also suggested inconsistency with the expectation of age progression from older to younger islands, except for Camiguin and Babuyan islands. Therefore, our results reject the hypothesis of stepping-stone by island age for *P. orbifer*.

Although most island lineages of the Southern Taiwan–Luzon volcanic belt are older than that of their northern counterparts, indicating the noticeable effect of a northward stepping-stone by island distance, the temporal order of origin of these island lineages did not follow a linear sequence of spatial arrangement in the

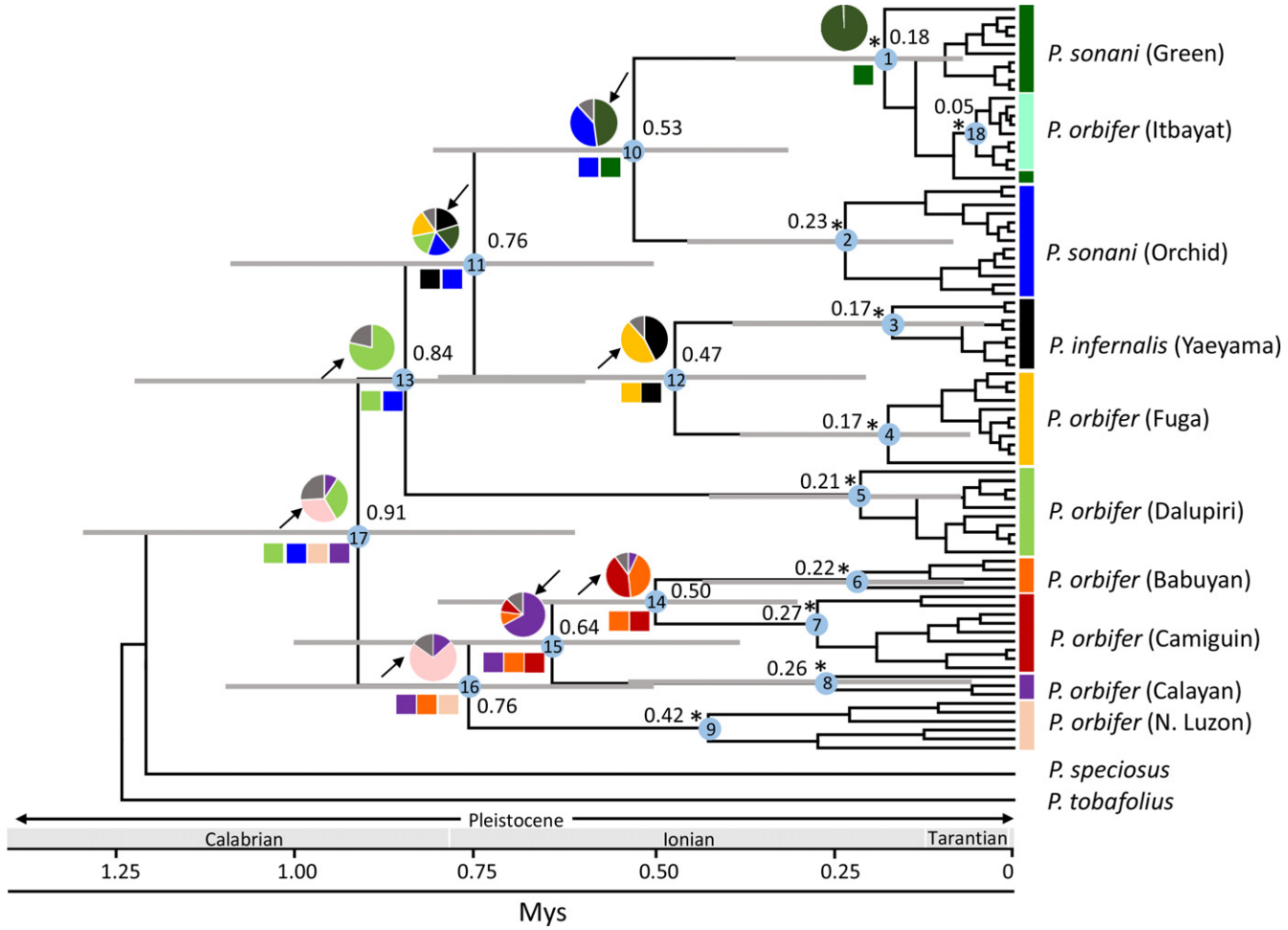

FIGURE 3 Divergence time, species delimitation and ancestral area of the *Pachyrhynchus orbifer* complex in the Taiwan–Luzon volcanic belt. *Pachyrhynchus speciosus* and *Pachyrhynchus tobafolius* were used as outgroups. The pie charts specify the relative probability of the ancestral area of nodes based on the BBM model, with arrows indicating the highest posterior probability of the reconstructed area. The colour squares represent the most probable ancestral area estimated from the best-fit model DIVA-LIKE + J in BioGeoBEARS. An asterisk indicates the Bayesian posterior probability calculated in BPP, where all members within a lineage belonging to one species is ≥0.99 (Appendix S7). Numbers in blue dots are node numbers of the tree [Colour figure can be viewed at wileyonlinelibrary.com]

TABLE 2 Model selection of ancestral area reconstruction of BioGeoBEARS for the *Pachyrhynchus orbifer* complex in the Taiwan–Luzon volcanic belt

	Log likelihood	Number of parameters	*d*	*e*	*j*	AIC
DEC	−45.863	2	0.053081	0.173657	0	95.72594
DEC + J	−42.7932	3	1.00E−12	0.145533	0.004753	91.58633
DIVALIKE	−37.6794	2	0.052299	1.00E−12	0	79.35875
DIVALIKE + J	−37.6159	3	0.042183	1.00E−12	0.000869	81.2317
BAYAREALIKE	−60.8371	2	0.061572	1.503288	0	125.6743
BAYAREALIKE + J	−56.7928	3	0.012976	1.439617	0.003612	119.5857

archipelago. The results of historical demography suggest that none of the island lineages showed recognizable population fluctuations, and there was no apparent pattern of southern stable versus northern expansion among island lineages. Together, these findings establish that the direction, sequence, timing and demographic history of the colonization of the *P. orbifer* complex were not correlated with the geo-historical organization of the islands in a south–north succession, as predicted from the strict stepping-stone hypothesis by distance. Alternatively, our findings indicate that the *P. orbifer* complex of the Taiwan–Luzon volcanic belt had a complex colonization history of both northward and southward directions with short- and long-distance dispersal events.

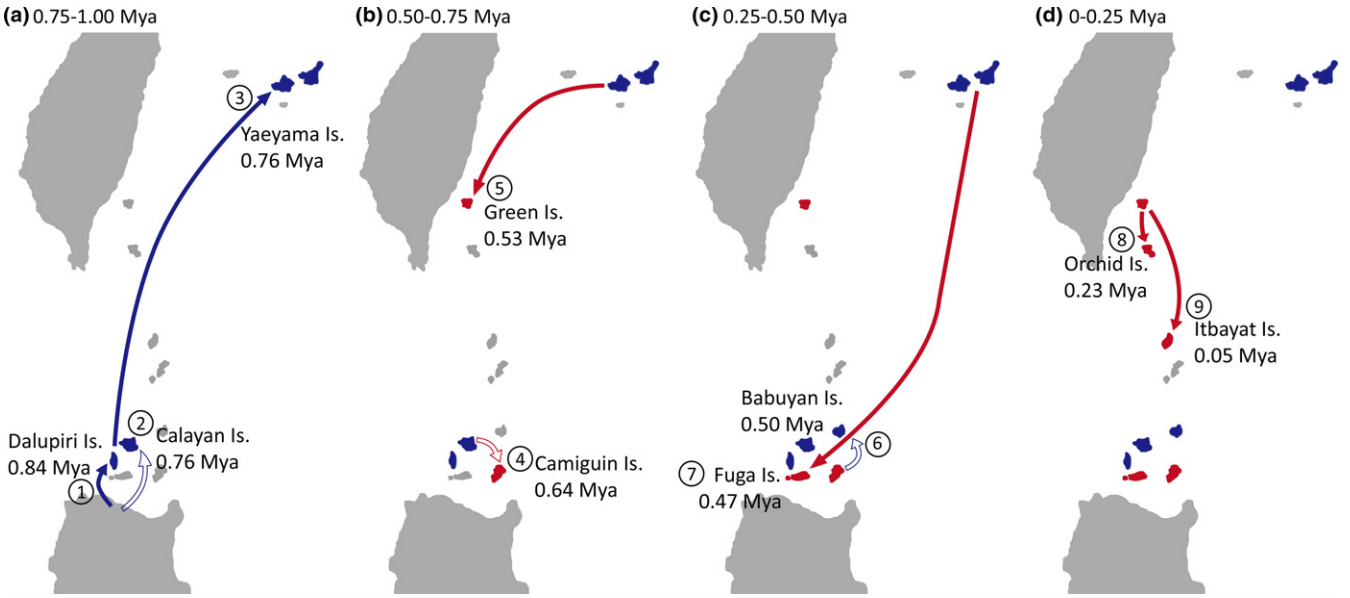

FIGURE 4 Inferred dispersal history of the *Pachyrhynchus orbifer* complex across the Taiwan–Luzon volcanic belt. (a) 0.75–1.00 Ma: the first two colonization events from Luzon to Dalupiri (0.84 Ma) and Calayan (0.76 Ma), and then a long-distance dispersal from Dalupiri to Yaeyama (0.76 Ma); (b) 0.50–0.75 Ma: one southward dispersal from Calayan to the neighbouring island of Camiguin (0.64 Ma), and the other dispersal event from Yaeyama to Green Island (0.53 Ma); (c) 0.25–0.50 Ma: one northward dispersal from Camiguin to adjacent Babuyan Island (0.50 Ma), and one southward long-distance dispersal from Yaeyama to Fuga (0.47 Ma); and (d) 0–0.25 Ma: two southward dispersals including one dispersal from Green Island to Orchid Island (0.23 Ma), and the other from Green Island to Itbayat (0.05 Ma). Blue and red arrows represent northward and southward dispersal, respectively. Solid and open arrows indicate the two major dispersal routes [Colour figure can be viewed at wileyonlinelibrary.com]

Pachyrhynchus weevils are wingless, sedentary insects with a considerable degree of host plant specialization that precludes the mobility of organisms as a possible explanation for the inferred complex colonization history. Insular *P. orbifer* is a recent lineage that diverged during the Pleistocene, when all extant islands of the archipelago had emerged above sea level. Thus, the non-linear, more complex spatial and temporal settings of the Taiwan–Luzon volcanic belt and stochastic dispersal were probable key factors shaping the colonization history of the *P. orbifer* complex.

4.2 | Complex inter-island colonization and peripatric founder speciation as major drivers of diversification

One of the most striking patterns of *Pachyrhynchus*'s diversification is the apparent stochastic nature of inter-island colonization. Nevertheless, diversification of the *P. orbifer* complex in the Taiwan–Luzon volcanic belt appears to have occurred only between islands. This study shows that colonization of new islands always leads to speciation among *Pachyrhynchus* lineages. These findings therefore strongly suggest that peripatric speciation through founders of stochastic dispersals was the major evolutionary driver of diversification. Peripatric speciation by founder events can be a predominant mode of speciation on oceanic islands, given prolonged periods of isolation between ancestral and founder populations, and through the effect of genetic drift and environmental heterogeneity among islands (Mayr, 1954; Templeton, 1980). Our finding of no closely related *Pachyrhynchus* species inhabiting the same island indicates that the process of in situ speciation within individual islands was less important than inter-island diversification. Ecological differentiation within an island does not seem to have occurred among any lineages of the *P. orbifer* complex in the Taiwan–Luzon volcanic belt.

The second unique feature of *Pachyrhynchus*'s diversification is the evident long-distance dispersal between the northern and southern edges of their distribution. At least three of the nine evolutionarily successful colonization events accomplished were caused by long-distance dispersal (i.e. dispersal between non-neighbouring extant islands in the archipelago; Figure 4, dispersal events 3, 7 and 9) (alternative scenarios, Appendix S6). This finding reinforces the notion that a single extreme long-distance dispersal event could be equally or more probable than multiple shorter dispersal events (i.e. the stepping-stone model), given that probabilities multiply in a chain of independent consecutive rare events (Nathan, 2006). Within the Taiwan–Luzon volcanic belt, long-distance dispersal by skipping the stepping-stone islands in between has been inferred for the Philippine bulbuls (*Microscelis amaurotis*) from Japan to the Babuyan and Batanes Islands (Oliveros & Moyle, 2010), and for Kikuchi's geckos (*Gekko kikuchii*) from Luzon to Orchid Island (Siler et al., 2014). However, the prevalence of long-distance dispersal in the Taiwan–Luzon volcanic belt shown herein and in earlier studies requires further examination by comparative analyses of colonization histories and community assemblies across taxonomic groups (Shaw & Gillespie, 2016).

The enormous diversity of weevils is often attributed to adaptation and co-evolution to the complexity and diversity in phenotypes, life histories and habitats of flowering plants (McKenna, Sequeira, Marvaldi, & Farrell, 2009). Available host plant records of the *P. orbifer* complex (Appendix S9) suggest that inter-island diversification of these weevils is almost invariably associated with shifts of host plant range or to distantly related host plants. We hypothesize that, in addition to founders of stochastic dispersals, ecological adaptation to specialized host plants similar to *Cratopus* weevils of Mauritius (Kitson et al., 2013) may also have played an important role in differentiation among insular *Pachyrhynchus* species in the Taiwan–Luzon volcanic belt. However, the incomplete host records and occasional host records of cultivated plants currently limit our interpretation of co-evolution under a phylogenetic framework between the *P. orbifer* complex and their host ranges.

4.3 | Probable mechanisms of overwater dispersal of remote oceanic islands

Our analyses indicated that flightless *Pachyrhynchus* weevils were capable of both short- and relatively long-distance dispersal across the ocean, despite being able to stay afloat on water for only several hours (Schultze, 1923). Recent studies on a few flightless weevils have suggested that they have been able to colonize numerous remote archipelagos and islands likely due to their long and complex histories in the regions (Claridge et al., 2017; Machado et al., 2017; Tänzler et al., 2016). However, the mechanism and transporting vectors of effective long-distance dispersal across oceanic barriers remain elusive for these weevils. One potential mechanism of cross-oceanic dispersal is drifting on floating vegetation, wood and debris due to oceanic currents. Rafting of terrestrial organisms to remote oceanic islands has been reported from a range of floating substrates and across all major oceans of the world (Peck, 1994; Thiel & Gutow, 2005). In particular, live adult weevils of *Sphenophorus* sp. and *Macrancylus linearis* were found on drifting debris offshore of Puerto Rico (Heatwole & Levins, 1972). Therefore, it is probable that *Pachyrhynchus* weevils may have colonized the Taiwan–Luzon volcanic belt through rafting as adults on floating substrates, or as eggs, larvae and pupae inside the stems and fruits of their host plants. Larvae of wood-boring beetles such as *Pachyrhynchus* weevils are among the most frequent insects found on floating items on the open sea (Heatwole & Levins, 1972). This wood-boring habit may facilitate their isolation from saltwater, thereby increasing their survival at sea. One of the host plants of *Pachyrhynchus* weevils, the fish poison tree (*Barringtonia asiatica*), has rather large box-shaped fruit with thick spongy fibrous layers. The fruit is extremely water-resistant and buoyant, and possibly represents adaptations for sea dispersal via the ocean current (Tsou & Mori, 2002). These specialized fruits of *B. asiatica* could serve as floating vehicles and simultaneously as a food source during the oceanic journeys of larval *Pachyrhynchus*.

The regional dominant Kuroshio current may also have been important in facilitating northward rafting of *Pachyrhynchus* weevils across the Taiwan–Luzon volcanic belt, especially the relatively long-distance dispersal event (Dalupiri to Yaeyama Islands, >600 km). Nevertheless, only four of nine historical colonization events in *Pachyrhynchus* weevils have been northward dispersals, and three of the these four events were relatively short-distance dispersals between neighbouring islands at the southern end of the Taiwan–Luzon volcanic belt (Figure 4). The remaining historical colonization events were all southbound dispersals, including two long-distance dispersal events from Yaeyama Islands to Fuga (*c.* 650 km) and from Green to Itbayat Island (*c.* 215 km). These findings indicate other unidentified mechanisms besides Kuroshio current, such as prevailing north-eastern monsoons, strong typhoon winds or stochastic movements (largely moving north–south direction) of surface flows in the region (Stommel & Yoshida, 1972), which could have directed the historical southward colonization routes of *Pachyrhynchus* weevils.

4.4 | Cryptic species diversity and conservation of island endemics

The diversification of invertebrate fauna in the Philippines was poorly studied until recently (Su, Wang, Villanueva, Nuñeza, & Lin, 2014), especially for species colonizing the smaller islands at the fringe of the archipelago. Our study provides the first compelling evidence of a recent diversification of invertebrates across small oceanic islands lying at the northern periphery of the Philippine archipelago between Taiwan and Luzon. In contrast to earlier taxonomic arrangements of *Pachyrhynchus* weevils based mainly on colour pattern (Kano, 1929; Schultze, 1923; but see Yoshitake, 2013), we have evidence of seven cryptic, but divergent, monophyletic species of *P. orbifer*, as delimitated through statistical inferences from genetic data. Species of *P. orbifer* complex with similar colouration were sometimes not the most closely related sister taxa (e.g. *P. sonani* of Orchid Island and *P. orbifer* of Babuyan Island; *P. orbifer* of Fuga and Northern Luzon). These findings indicate that colour markings of the *P. orbifer* complex can evolve rapidly and show little phylogenetic conservatism, such as the loss of colour markings in *P. infernalis* on Yayeyama Island and the frequent evolutionary transitions of stripe colours between sister *Pachyrhynchus* species. We propose that all seven identified *P. orbifer* should be recognized as candidate species based on the monophyletic lineages, the statistical species limits, and in combination with their isolated distributions, host plant ranges and colour variation.

The most remarkable scenario of cryptic diversification in this study was obtained from the surprisingly large genetic divergence between morphologically identical *P. orbifer* of Barit Island (Figure 2, P040) and Fuga Island (average genetic distances of four genes, 0.3 ± 0.1% vs. 0.1 ± 0.04% between Itbayat and Green Island). These two islands currently are only 1.2 km apart (Figure 1a) but were unified into a single larger island during the Pleistocene glacial cycles. The

substantial genetic divergence between weevils of these two extremely close neighbouring islands suggests a strong isolation effect of the oceanic barrier or priority effects due to the order and time of colonization (De Meester, Vanoverbeke, Kilsdonk, & Urban, 2016; Fukami, 2015). This high genetic differentiation at fine spatial scales highlights that the diversity of cryptic species of *Pachyrhynchus* weevils on smaller oceanic islands may still be largely underestimated (e.g. flightless *Trigonopterus* weevils in Bali, Tänzler, Toussaint, Suhardjono, Balke, & Riedel, 2014). Conservation of insular species and endangered populations in their peripheral ranges is crucial for preserving rare alleles and unique phenotypes. These rare alleles and localized phenotypes are of particular interest because they may be responsible for the adaptation of endemic island species to local habitats.

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DATA ACCESSIBILITY

DNA sequences: GenBank accession numbers, KU953405-KU953778. Phylogenetic data: Dryad Digital Repository, https://doi.org/10.5061/dryad.25fc4.

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REFERENCES

Brower, A. (1994). Rapid morphological radiation and convergence among races of the butterfly *Heliconius erato* inferred from patterns of mitochondrial DNA evolution. *Proceedings of the National Academy of Sciences of the United States of America*, *91*, 6491–6495.

Carson, H. L. (1983). Chromosomal sequences and interisland colonizations in Hawaiian *Drosophila*. *Genetics*, *103*, 465–482.

Chen, Y.-T., Tseng, H.-Y., Jeng, M.-L., Su, Y.-C., Huang, W.-S., & Lin, C.-P. (2017). Integrated species delimitation and conservation implications of an endangered weevil *Pachyrhynchus sonani* (Coleoptera: Curculionidae) in Green and Orchid Islands of Taiwan. *Systematic Entomology*, *42*, 796–813.

Claridge, E. M., Gillespie, R. G., Brewer, M. S., & Roderick, G. K. (2017). Stepping-stones across space and time: Repeated radiation of Pacific flightless broad-nosed weevils (Coleoptera: Curculionidae: Entiminae: *Rhyncogonus*). *Journal of Biogeography*, *44*, 784–796.

De Meester, L., Vanoverbeke, J., Kilsdonk, L. J., & Urban, M. C. (2016). Evolving perspectives on monopolization and priority effects. *Trends in Ecology & Evolution*, *31*, 136–146.

Drummond, A. J., & Bouckaert, R. R. (2015). *Bayesian evolutionary analysis with BEAST*. Cambridge: Cambridge University Press.

Drummond, A. J., & Rambaut, A. (2007). BEAST: Bayesian evolutionary analysis by sampling trees. *BMC Evolutionary Biology*, *7*, 214.

Drummond, A. J., Suchard, M. A., Xie, D., & Rambaut, A. (2012). Bayesian phylogenetics with BEAUti and the BEAST 1.7. *Molecular Biology and Evolution*, *29*, 1969–1973.

Esselstyn, J. A., & Oliveros, C. H. (2010). Colonization of the Philippines from Taiwan: A multi-locus test of the biogeographic and phylogenetic relationships of isolated populations of shrews. *Journal of Biogeography*, *37*, 1504–1514.

Folmer, O., Black, M., Hoeh, W., Lutz, R., & Vrijenhoek, R. (1994). DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology*, *3*, 294–299.

Fukami, T. (2015). Historical contingency in community assembly: Integrating niches, species pools, and priority effects. *Annual Review of Ecology, Evolution, and Systematics*, *46*, 1–23.

Funk, V. A., & Wagner, W. L. (1995). Biogeographic patterns in the Hawaiian Islands. In V. A. Funk, & W. L. Wagner (Eds.), *Hawaiian biogeography: Evolution on a hot spot archipelago* (pp. 379–419). Washington: Smithsonian Institution.

Gillespie, R. G. (2004). Community assembly through adaptive radiation in Hawaiian spiders. *Science*, *303*, 356–359.

Gillespie, R. G., Baldwin, B. G., Waters, J. M., Fraser, C. I., Nikula, R., & Roderick, G. K. (2012). Long-distance dispersal: A framework for hypothesis testing. *Trends in Ecology & Evolution*, *27*, 47–56.

Gillespie, R. G., & Roderick, G. K. (2002). Arthropods on islands: Colonization, speciation, and conservation. *Annual Review of Entomology*, *47*, 595–632.

Heatwole, H., & Levins, R. (1972). Biogeography of the Puerto Rican Bank: Flotsam transport of terrestrial animals. *Ecology*, *53*, 112–117.

Heled, J., & Drummond, A. J. (2008). Bayesian inference of population size history from multiple loci. *BMC Evolutionary Biology*, *8*, 289.

Heled, J., & Drummond, A. J. (2010). Bayesian inference of species trees from multilocus data. *Molecular Biology and Evolution*, *27*, 570–580.

Hernández-Vera, G., Mitrović, M., Jović, J., Toševski, I., Caldara, R., Gassmann, A., & Emerson, B. C. (2010). Host-associated genetic differentiation in a seed parasitic weevil *Rhinusa antirrhini* (Coleptera: Curculionidae) revealed by mitochondrial and nuclear sequence data. *Molecular Ecology*, *19*, 2286–2300.

Huang, J. P., & Lin, C. P. (2010). Diversification in subtropical mountains: Phylogeography, Pleistocene demographic expansion, and evolution of polyphenic mandibles in Taiwanese stag beetle, *Lucanus formosanus*. *Molecular Phylogenetics and Evolution*, *57*, 1149–1161.

Juan, C., Emerson, B. C., Oromí, P., & Hewitt, G. M. (2000). Colonization and diversification: Towards a phylogeographic synthesis for the Canary Islands. *Trends in Ecology & Evolution*, *15*, 104–109.

Kano, T. (1929). Description of three new species of Curculionidae of the genus *Pachyrrhynchus* Germar from the island of Botel-Tobago. *Entomological Society of Japan*, *3*, 237–238.

Kano, T. (1936). Some problems concerning the biogeography of Kôthshô (5). *Geographical Review of Japan*, *10*, 59–83.

Kano, T. (1941). Biogeography of the island of Kôtôsho (Botel Tobago) with special reference to the New-Wallace Line. In *The Institute of the Pacific, Section of Sciences*, (Ed.), *Greater South Seas: Its culture and its soil* (pp. 219–323). Tokyo: Kawade Shobou.

Kitson, J. J., Warren, B. H., Vincent Florens, F. B., Baider, C., Strasberg, D., & Emerson, B. C. (2013). Molecular characterization of trophic ecology within an island radiation of insect herbivores (Curculionidae: Entiminae: *Cratopus*). *Molecular Ecology*, *22*, 5441–5455.

Kuo, J., Kanamoto, Z., Iizumi, H., Aioi, K., & Mukai, H. (2006). Seagrasses from the Nansei Islands, Southern Japanese Archipelago: Species composition, distribution and biogeography. *Marine Ecology*, *27*, 290–298.

Kurita, K., & Hikida, T. (2014). Divergence and long-distance overseas dispersals of island populations of the Ryukyu five-lined skink, *Plestiodon marginatus* (Scincidae: Squamata), in the Ryukyu Archipelago, Japan, as revealed by mitochondrial DNA phylogeography. *Zoological Science*, *31*, 187–194.

Landis, M. J., Matzke, N. J., Moore, B. R., & Huelsenbeck, J. P. (2013). Bayesian analysis of biogeography when the number of areas is large. *Systematic Biology*, *62*, 789–804.

Lanfear, R., Calcott, B., Ho, S. Y., & Guindon, S. (2012). PartitionFinder: Combined selection of partitioning schemes and substitution models for phylogenetic analyses. *Molecular Biology and Evolution*, *29*, 1695–1701.

Lim, H. C., & Sheldon, F. H. (2011). Multilocus analysis of the evolutionary dynamics of rainforest bird populations in Southeast Asia. *Molecular Ecology*, *20*, 3414–3438.

Losos, J. B., & Ricklefs, R. E. (2009). Adaptation and diversification on islands. *Nature*, *457*, 830–836.

MacArthur, R. H., & Wilson, E. O. (1967). *The theory of island biogeography*. Princeton, NJ: Princeton University Press.

Machado, A., Rodríguez-Expósito, E., López, M., & Hernández, M. (2017). Phylogenetic analysis of the genus *Laparocerus*, with comments on colonisation and diversification in Macaronesia (Coleoptera, Curculionidae, Entiminae). *ZooKeys*, *651*, 1–77.

Matzke, N.J. (2013a). BioGeoBEARS: BioGeography with Bayesian (and likelihood) evolutionary analysis in R Scripts, CRAN: The Comprehensive R Archive Network, Vienna, Austria. http://cran.r-project.org/package=BioGeoBEARS.

Matzke, N. J. (2013b). Probabilistic historical biogeography: New models for founder-event speciation, imperfect detection, and fossils allow improved accuracy and model-testing. *Frontiers of Biogeography*, *5*, 242–248.

Matzke, N. J. (2014). Model selection in historical biogeography reveals that founder-event speciation is a crucial process in island clades. *Systematic Biology*, *63*, 951–970.

Mayr, E. (1954). Change of genetic environment and evolution. In J. Huxley, A. C. Hardy, & E. B. Ford (Eds.), *Evolution as a process* (pp. 157–180). London: Allen and Unwin.

McKenna, D. D., Sequeira, A. S., Marvaldi, A. E., & Farrell, B. D. (2009). Temporal lags and overlap in the diversification of weevils and flowering plants. *Proceedings of the National Academy of Sciences of the United States of America*, *106*, 7083–7088.

Moriyama, E. N., & Gojobori, T. (1992). Rates of synonymous substitution and base composition of nuclear genes in *Drosophila*. *Genetics*, *130*, 855–864.

Nakamura, K., Kokubugata, G., Rubite, R. R., Huang, C. Jr, Kono, Y., Yang, H. A., … Peng, C. I. (2014). In situ glacial survival at the northern limit of tropical insular Asia by a lowland herb *Begonia fenicis* (Begoniaceae). *Botanical Journal of the Linnean Society*, *174*, 305–25.

Nathan, R. (2006). Long-distance dispersal of plants. *Science*, *313*, 786–788.

Oliveros, C. H., & Moyle, R. G. (2010). Origin and diversification of Philippine bulbuls. *Molecular Phylogenetics and Evolution*, *54*, 822–832.

Oliveros, C., Ota, H., Crombie, R., & Brown, R. (2011). The Herpetofauna of the Babuyan group of islands, northern Philippines. *Scientific Papers of the Natural History Museum of the University of Kansas*, *43*, 1–20.

Osozawa, S., Shinjo, R., Armid, A., Watanabe, Y., Horiguchi, T., & Wakabayashi, J. (2012). Palaeogeographic reconstruction of the 1.55 Ma synchronous isolation of the Ryukyu Islands, Japan, and Taiwan and inflow of the Kuroshio warm current. *International Geology Review*, *54*, 1369–1388.

Ota, H., & Huang, W.-S. (2000). *Mabuya cumingi* (Reptilia: Scincidae): An addition to the herpetofauna of Lanyu Island, Taiwan. *Current Herpetology*, *19*, 57–61.

Papadopoulou, A., Anastasiou, I., & Vogler, A. P. (2010). Revisiting the insect mitochondrial molecular clock: The mid-Aegean trench calibration. *Molecular Biology and Evolution*, *27*, 1659–1672.

Parent, C. E., Caccone, A., & Petren, K. (2008). Colonization and diversification of Galápagos terrestrial fauna: A phylogenetic and biogeographical synthesis. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *363*, 3347–3361.

Peck, S. B. (1994). Sea-surface (pleuston) transport of insects between islands in the Galápagos Archipelago, Ecuador. *Annals of the Entomological Society of America*, *87*, 576–582.

Planas, E., & Ribera, C. (2014). Uncovering overlooked island diversity: Colonization and diversification of the medically important spider genus *Loxosceles* (Arachnida: Sicariidae) on the Canary Islands. *Journal of Biogeography*, *41*, 1255–1266.

Rambaut, A., Suchard, M.A., Xie, D., & Drummond, A.J. (2014). Tracer v1.6. Available from http://beast.bio.ed.ac.uk/Tracer.

Ree, R. H., Moore, B. R., Webb, C. O., & Donoghue, M. J. (2005). A likelihood framework for inferring the evolution of geographic range on phylogenetic trees. *Evolution*, *59*, 2299–2311.

Ree, R. H., & Smith, S. A. (2008). Maximum likelihood inference of geographic range evolution by dispersal, local extinction, and cladogenesis. *Systematic Biology*, *57*, 4–14.

Ronquist, F. (1997). Dispersal-vicariance analysis: A new approach to the quantification of historical biogeography. *Systematic Biology*, *46*, 195–203.

Ronquist, F., & Huelsenbeck, J. P. (2003). MrBayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics*, *19*, 1572–1574.

Ronquist, F., Teslenko, M., van der Mark, P., Ayres, D. L., Darling, A., Höhna, S., … Huelsenbeck, J. P. (2012). MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology*, *61*, 539–542.

Schultze, W. (1923). A monograph of the pachyrrhynchid group of the Brachyderinae, Curculionidae: Part 1. *The Philippine Journal of Science*, *23*, 609–673.

Shaw, K. L., & Gillespie, R. G. (2016). Comparative phylogeography of oceanic archipelagos: Hotspots for inferences of evolutionary process. *Proceedings of the National Academy of Sciences of the United States of America*, *113*, 7986–7993.

Shimodaira, H., & Hasegawa, M. (2001). CONSEL: For assessing the confidence of phylogenetic tree selection. *Bioinformatics*, *17*, 1246–1247.

Siler, C. D., Oaks, J. R., Cobb, K., Ota, H., & Brown, R. M. (2014). Critically endangered island endemic or peripheral population of a widespread species? Conservation genetics of Kikuchi's gecko and the global challenge of protecting peripheral oceanic island endemic vertebrates. *Diversity and Distributions*, *20*, 756–772.

Simon, C., Buckley, T. R., Frati, F., Stewart, J. B., & Beckenbach, A. T. (2006). Incorporating molecular evolution into phylogenetic analysis, and a new compilation of conserved polymerase chain reaction primers for animal mitochondrial DNA. *Annual Review of Ecology, Evolution, and Systematics*, *37*, 545–579.

Stamatakis, A. (2006). RAxML-VI-HPC: Maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics*, *22*, 2688–2690.

Stommel, H. M., & Yoshida, K. (1972). *Kuroshio: Its physical aspects*. Tokyo: University of Tokyo Press.

Su, Y. C., Wang, J. F., Villanueva, R. J. T., Nuñeza, O. M., & Lin, C.-P. (2014). Hopping out of Mindanao: Miocene-Pliocene geological processes and cross-island dispersal as major drivers of diversity for Philippine treehoppers. *Journal of Biogeography*, *41*, 1277–1290.

Tamura, K., Stecher, G., Peterson, D., Filipski, A., & Kumar, S. (2013). MEGA6: Molecular evolutionary genetics analysis version 6.0. *Molecular Biology and Evolution*, *30*, 2725–2729.

Tänzler, R., Toussaint, E. F., Suhardjono, Y. R., Balke, M., & Riedel, A. (2014). Multiple transgressions of Wallace's Line explain diversity of

flightless *Trigonopterus* weevils on Bali. *Proceedings of the Royal Society of London B: Biological Sciences*, *281*, 20132528.
Tänzler, R., Van Dam, M. H., Toussaint, E. F., Suhardjono, Y. R., Balke, M., & Riedel, A. (2016). Macroevolution of hyperdiverse flightless beetles reflects the complex geological history of the Sunda Arc. *Scientific Reports*, *6*, 18793.
Templeton, A. R. (1980). The theory of speciation via the founder principle. *Genetics*, *94*, 1011–1038.
Thiel, M., & Gutow, L. (2005). The ecology of rafting in the marine environment. II. The rafting organisms and community. *Oceanography and Marine Biology: An Annual Review*, *43*, 279–418.
Tseng, H.-Y., Lin, C.-P., Hsu, J.-Y., Pike, D. A., & Huang, W.-S. (2014). The functional significance of aposematic signals: Geographic variation in the responses of widespread lizard predators to colourful invertebrate prey. *PLoS ONE*, *9*, e91777.
Tsou, C.-H., & Mori, S. A. (2002). Seed coat anatomy and its relationship to seed dispersal in subfamily Lecythidoideae of the Lecythidaceae (the Brazil nut family). *Botanical Bulletin of Academia Sinica*, *43*, 37–56.
Voris, H. K. (2000). Maps of Pleistocene sea levels in Southeast Asia: Shorelines, river systems and time durations. *Journal of Biogeography*, *27*, 1153–1167.
Weekers, P. H. H., De Jonckheere, J. F., & Dumont, H. J. (2001). Phylogenetic relationships inferred from ribosomal ITS sequences and biogeographic patterns in representatives of the genus *Calopteryx* (Insecta: Odonata) of the West Mediterranean and adjacent West European zone. *Molecular Phylogenetics and Evolution*, *20*, 89–99.
Whittaker, R. J., & Fernández-Palacios, J. M. (2007). *Island biogeography: Ecology, evolution, and conservation*. New York: Oxford University Press.
Whittaker, R. J., Triantis, K. A., & Ladle, R. J. (2008). A general dynamic theory of oceanic island biogeography. *Journal of Biogeography*, *35*, 977–994.
Yang, Z. (2015). The BPP program for species tree estimation and species delimitation. *Current Zoology*, *61*, 854–865.
Yang, T. F., Lee, T., Chen, C. H., Cheng, S. N., Knittel, U., Punongbayan, R. S., & Rasdas, A. R. (1996). A double island arc between Taiwan and Luzon: Consequence of ridge subduction. *Tectonophysics*, *258*, 85–101.
Yang, Z., & Rannala, B. (2014). Unguided species delimitation using DNA sequence data from multiple loci. *Molecular Biology and Evolution*, *31*, 3125–3135.
Yoshitake, H. (2013). A new genus and two new species of the tribe Pachyrhynchini (Coleoptera: Curculionidae) from Palawan Island, the Philippines. *Esakia*, *53*, 1–8.
Yu, Y., Harris, A. J., Blair, C., & He, X. (2015). RASP (Reconstruct Ancestral State in Phylogenies): A tool for historical biogeography. *Molecular Phylogenetics and Evolution*, *87*, 46–49.

BIOSKETCHES

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Author contributions: C.-P.L. designed the study; H.-Y.T., W.-S.H., M.-L.J., R.J.T.V., O.M.N. and C.-P.L. carried out the fieldwork; H.-Y.T generated the molecular data; H.-Y.T and C.-P.L. conducted the phylogenetic and statistical analyses; and C.-P.L. and W.-S.H. led the writing with input from H.-Y.T and M.-L.J.

SUPPORTING INFORMATION

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

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