

Colonization of the Pacific by *Bembidion* beetles (Coleoptera: Carabidae), with description of

*Bembidion tahitiense*, sp. nov., from Tahiti, French Polynesia

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## Abstract

*Bembidion (Sloanephila) tahitiense*, sp. nov., is described from Mont Mauru, an isolated massif of Tahiti Nui volcano. Based upon evidence from seven genes (four nuclear protein-coding, one mitochondrial protein-coding, two nuclear ribosomal), its sister group is the Australian *B. jacksoniense* Guérin-Ménéville, with which it shares a synapomorphic spur on the ostium of the male genitalia. In contrast to *B. jacksoniense*, *B. tahitiense* is brachypterous, with rounded humeri, constricted posterior pronotal margins, and convex body form. Examination of the seven genes in two species of the Hawaiian subgenus *Nesocidium* Sharp reveals that the sister group of *Nesocidium* is subgenus *Zecillenus* Lindroth from New Zealand. These two subgenera belong to the *Ananotaphus* Complex, a clade inhabiting Australia, New Zealand, and Hawaii. The relationships of the second Hawaiian subgenus, *Gnatholymnaeum* Sharp, are less clear, although *Gnatholymnaeum* belongs to the *Bembidion* Series (along with *Sloanephila* and the *Ananotaphus* Complex). *Bembidion* beetles colonized the Society and Hawaiian Islands independently from source areas in the southwest Pacific. Based on parsimonious reconstructions of flight-wing configuration, the Tahitian and Hawaiian colonizations involved winged individuals. Colonization of the Society and Hawaiian Islands by carabid beetles of two other tribes—Platynini and Moriomorhini—follow the dispersal patterns hypothesized for *Bembidion*.

*Additional keywords:* biogeography, brachyptery, dispersal, speciation

*Running title:* *Bembidion (Sloanephila) tahitiense*, sp. nov.

## Introduction

Beetles of the genus *Bembidion* Latreille represent one of the more impressive radiations within Carabidae, given both the worldwide geographic scope of the taxon and the immense number of

constituent species that have evolved. There are over 1200 described species (Lorenz 2005), most in the Holarctic region, but with a notable center of diversity in temperate South America, especially in the mountains. Yet in spite of such diversity, *Bembidion* beetles have colonized very few archipelagos in the Pacific Ocean, and the group is not particularly diverse in mainland areas bordering the southwest Pacific. Although New Zealand hosts a moderate *Bembidion* fauna of 20 native species (Laroche and Larivière 2001), the native Australian *Bembidion* fauna totals only 10 species (Moore et al. 1987; Toledano 2005). Whereas many of the Pacific archipelagos composed of smaller, low-elevation islands totally lack *Bembidion* representation (Darlington 1970), several larger island groups host minimal endemic *Bembidion* diversity: *B. insularum* Andrewes is the sole *Bembidion* species precinctive to Fiji, and *B. hebridarum* Lindroth is the lone generic representative in Vanuatu (Lindroth 1980). Both of these species are placed in the subgenus *Desarmatocillenus* Netolitzky (Lindroth 1980), a taxon of ocean-shore species otherwise distributed in Australia, New Guinea, the Philippines, China and Japan. Within the Pacific, only the Hawaiian Islands host a sizable *Bembidion* fauna, with 23 native endemic species (Liebherr 2008). These are classified into two *Bembidion* subgenera: two species placed in subgenus *Gnatholymnaeum* Sharp, and 21 assigned to subgenus *Nesocidium* Sharp. Taxa in both subgenera are geographically restricted to Hawaii.

Herein we describe a new *Bembidion* species from the Society Islands, an archipelago previously not known to have been colonized by *Bembidion* beetles. The species exhibits a number of autapomorphies, mostly due to evolutionary reduction of its metathoracic flight-wing apparatus. Thus the elytral shape, convex elytral intervals, and narrow body overall are likely due to transformation of the metathorax into a flightless locomotory structure. Based on the results of a molecular phylogenetic analysis including representative *Bembidion* from across the southwest Pacific, we offer an hypothesis for the geographic origin of the ancestor of the present-day Society Island taxon. By including representatives of the Hawaiian subgenera, we also elucidate the patterns of dispersal from the southwest Pacific to both the Society and Hawaiian archipelagos.

## **Materials and methods**

### *Taxonomic material*

Specimens of the new Society Island *Bembidion* were collected in 2006 during a biological survey of the insects of Tahiti and Moorea islands. Members of the type series of the new species are deposited in the George Perrault Tahitian Carabidae collection, Muséum National d'Histoire

Naturelle, Paris (MNHN); the Cornell University Insect Collection, Ithaca (CUIC); and the Essig Museum of Entomology, University of California, Berkeley (EMEC). Additional comparative material was borrowed from the Museum of Comparative Zoology, Harvard University (MCZ) and the Oregon State Arthropod Collection, Oregon State University (OSAC).

#### *Taxonomic protocols*

Anatomical character analysis was accomplished using pointed, dry-mounted specimens. Habitus photos of whole specimens were made using a Microptics photographic apparatus with K2 series lenses and a Nikon D1X body. Two fibre-optic wands plus backlighting via a transmissible light stage from a third wand conveyed light to the specimen. Multiple images of varying focal depth were assembled with the Combine Z5 program (Hadley 2006). Male genitalia were examined after dissection, with specimens relaxed in near-boiling distilled water containing a drop of Kodak Photo-Flo® detergent, and dissected using minuten nadeln and watchmakers' forceps. Dissected parts were cleared in cold 10% KOH overnight, neutralized in dilute 10% acetic acid, and stored in glycerine. For some specimens, the genitalia were mounted in Euparal between paired circular cover slips that were attached to cardstock and pinned beneath the specimen. The male aedeagal median lobe in Euparal was photographed with a Leica Z6 and JVC KY-F75U camera, with a stack of photographs at different focal planes taken using Microvision's Cartograph® software. These photographs were then merged using the PMax procedure in Zerene Systems's Zerene Stacker®; the images thus potentially have some artifacts caused by the merging algorithm.

Type label data are transcribed in the description such that a single slash (/) separates lines of text on a label, and a double slash (//) indicates the start of text on a separate, lower label. The new species name is registered at Zoobank.org.

Two ratios are used in the taxonomic description: 1, ocular ratio, or the distance across the outer eye surface divided by the minimal distance between the eyes across the frons; and 2, MPW/BPW, i.e. the maximal pronotal width divided by the basal pronotal width measured between the angles laterally adjacent to the basal margin.

#### *DNA sequencing*

Genes studied, and abbreviations used in this paper, are: **28S** or 28S rDNA: 28S ribosomal DNA; **18S** or 18S rDNA: 18S ribosomal DNA; **COI**: cytochrome oxidase I; **wg**: *wingless*; **CAD**:

carbamoyl phosphate synthetase domain of the rudimentary gene; **ArgK**: arginine kinase; **Topo**: topoisomerase I.

DNA was extracted from specimens preserved in 95% ethanol using a Qiagen DNeasy Blood and Tissue Kit. Fragments for the seven genes were amplified using the Polymerase Chain Reaction on an Eppendorf Mastercycler Pro Thermal Cycler, using TaKaRa Ex Taq and the basic protocols recommended by the manufacturers. Primers and details of the cycling reactions used are given in Maddison (2012). The amplified products were then cleaned, quantified, and sequenced at the University of Arizona's Genomic and Technology Core Facility using a 3730 XL Applied Biosystems automatic sequencer.

Assembly of multiple chromatograms for each gene fragment and initial base calls were made with Phred (Green and Ewing 2002) and Phrap (Green 1999) as orchestrated by Mesquite's Chromaseq package (Maddison and Maddison 2011a; Maddison and Maddison 2011b) with subsequent modifications by Chromaseq and manual inspection. Multiple peaks at a single position in multiple reads were coded using IUPAC ambiguity codes.

#### *Taxon sampling for DNA studies*

We obtained DNA sequence data for all seven studied genes from the specimens of Hawaiian and Tahitian *Bembidion* listed in Table 1. DNA vouchers are housed in the David Maddison voucher collection at OSAC, with the exception of DNA voucher 2397 (*B. tahitiense*, sp. nov.), deposited in EMEC. Sequences have been deposited in GenBank with accession numbers KC460275 through KC460305.

In addition, Topo was sequenced from DNA vouchers 1700 (*B. (Ananotaphus) ateradustum* Liebherr) and 1439 (*B. (Zemettallina) hokitikense* Bates), and COI from voucher 1439; more details about those specimens are given in Maddison (2012). To these data were added sequences for the seven genes from other Bembidiina (data from Maddison 2012), for a total of 111 species (8 non-*Bembidion* Bembidiina, 13 *Bembidion* outside of the *Bembidion* Series, the 4 Pacific island taxa, and 86 other species of the *Bembidion* Series). Sequences from all 111 species were included for 28S, CAD, *wg*, and ArgK; for the other three genes sequence data is available for only some of the species (COI, 92 species; Topo, 90 species; 18S, 69 species). The species sampled are evident in Fig. 1; more details about GenBank accession numbers and voucher specimens for these species are given in Maddison (2012).

#### *Alignment and data exclusion*

Alignment was not difficult for any of the protein-coding genes. There were no insertions or deletions (indels) evident in the sampled CAD, ArgK, Topo, or COI sequences. In *wingless*, there were two small indels, restricted to only three taxa, separated from one another along the length of the sequence. There were three inserted nucleotides in *Bembidion (Zemetallina) parviceps* Bates, and six inserted nucleotides in a different region in the two species of subgenus *Omotaphus* Netolitzky sampled. These inserted nucleotides were excluded from analyses.

In contrast, the two ribosomal genes showed a moderate history of insertions and deletions. Both genes were first subjected to multiple sequence alignment in Opal version 2.1 (Wheeler and Kececioglu 2007), using default parameter values. Visual inspection suggested no needed improvements.

Sites were excluded if their data was present in less than 80 of the 111 sequenced species for 28S, CAD, wg, and ArgK; less than 75 of the 92 sequenced COI; less than 60 of the 90 sampled topoisomerase; less than 50 of the 69 sequenced 18S. This procedure served to exclude the partly sampled ends of each sequence, as well as almost all of the indel regions in the ribosomal genes. None of the included regions had obviously ambiguous alignments.

#### *Molecular phylogenetic analysis.*

Models of nucleotide evolution were chosen with the aid of jModelTest version 2.1.1 (Darriba et al. 2012; Guindon and Gascuel 2003). Among the models supported by RAxML, the model chosen for all genes by the Aikaike Information Criterion, Bayesian Information Criterion, and Decision Theoretic was GTR+I+ $\Gamma$ .

Likelihood analyses of nucleotide data were conducted using RAxML version 7.2.6 (Stamatakis 2006). Analyses were conducted on each gene individually, as well as a matrix of seven genes concatenated together partitioned by gene. For bootstrap analyses of the seven-gene matrix, 5000 replicates were conducted; for individual genes, 2000 replicates. In addition to these bootstrap analyses, a search for the maximum likelihood tree for the seven-gene matrix was conducted using 1000 search replicates.

Most-parsimonious trees (MPTs) were sought using PAUP\* (Swofford 2002). To search for most parsimonious trees, 2000 replicates were conducted, each beginning with a starting tree formed with the random addition sequence option, with each replicate saving no more than 25 trees. For parsimony bootstrap analyses in PAUP\*, 2000 bootstrap replicates were examined, each of which used a heuristic search with four replicates, each beginning with a starting tree

formed by the random addition sequence option, with TBR branch rearrangement, with each replicate saving no more than 25 trees; the estimated bootstrap values are reported as parsimony bootstrap percentages (PB).

### Phylogenetic Results

The maximum likelihood bootstrap tree and tree of highest likelihood found are shown in Figs 1 and 2 respectively. Table 2 shows the support for or against the clades numbered in Fig. 1. The four Pacific archipelago *Bembidion* species examined are all members of the *Bembidion* Series (Fig. 1), and fall in three distinct groups.

*Bembidion tahitiense*, sp. nov., is the sister group of *B. jacksoniense* Guérin-Méneville among the sampled species, a result supported by combined analyses and by each of the seven genes independently (Fig. 1, 2, Table 2), with bootstrap values being above 90 for all genes except COI.

Among the Hawaiian *Bembidion*, the two members of subgenus *Nesocidium* belong in the midst of taxa otherwise endemic to New Zealand and Australia, the *Ananotaphus* Complex of Maddison (2012). Within this complex, the two Hawaiian species of subgenus *Nesocidium* are strongly supported by multigene analyses to form the sister group of *Zecillenus*, a subgenus of ocean shore species restricted to New Zealand; this result is supported by 28S, CAD, *wg*, and ArgK data (Table 2). In turn, these two subgenera are in a larger clade (clade number 3 in Fig. 1) that also includes the Australian species in *Ananotaphus* Netolitzky (s. lat.), *B. ateradustum*, *B. errans* Blackburn, and *B. proprium* Blackburn (Table 2). The monophyly of the *Ananotaphus* Complex (clade 4 in Fig. 1) is strongly supported by the combined analyses (Table 2); there is some support in single-gene analyses from CAD, *wg*, and *topo*, and some support against the clade, most notably in arginine kinase.

The relationships of *B. (Gnatholymnaeum) blackburni* Sharp from Hawaii is less clear. There is moderate support from the combined analyses that it is the sister group of the *Ananotaphus* Complex (clade 5 in Fig. 1 and Table 2). The evidence for this comes from weak single-gene support from CAD and 18S. In addition, *B. blackburni* is within the *Ananotaphus* Complex in maximum likelihood and most parsimonious trees for ArgK and 28S. However, there is bootstrap support against a *Gnatholymnaeum-Ananotaphus* Complex clade in the topoisomerase data.

## Taxonomic Treatment

### Genus *Bembidion* Latreille

#### Subgenus *Sloanephila* Netolitzky

This species is placed in the subgenus *Sloanephila* with *B. (Sloanephila) jacksoniense* based on the phylogenetic relationships indicated by the DNA sequence data (Figs. 1 and 2, Table 2), in addition to the following shared morphological features: (1) two dorsal elytral setae that are situated free of elytral stria 3 in interval 3 (Figs 3, 5); (2) punctate striae 1–8 present at elytral midlength, with striae 2 and 4–6 absent basally and striae 2–7 absent from apical 1/6 of elytra (Figs 3, 6); (3) frontal grooves shallow, evident only near frontoclypeal suture where they are subparallel; (4) pronotum cordate, the posterior lateral seta situated far anterad the apparent hind angle (Figs 3, 7); (5) apex of ostium with a well-sclerotized spur (Figs 9, 11), a feature apparently unique within *Bembidion*, and a synapomorphy of the two species of *Sloanephila*.

#### *Bembidion (Sloanephila) tahitiense*, sp. nov.

##### *Material examined*

*Holotype male* (MNHN): French Polynesia: Tahiti Nui / Mt. Mauru lava tube / el. 705 m 18-IX-2006 lot 01 / 17°38.017'S 149°21.284'W / pyr. fog moss/liverworts / along stream J.K. Liebherr // HOLOTYPE / *Bembidion* / *tahitiense* / Liebherr & / Maddison 2012 (black-bordered red label).

*Paratype males* (CUIC, 1; EMEC, 1): same labeling except black-bordered yellow PARATYPE label. The EMEC specimen has in addition a pale green label with the following text: “David R. Maddison / DNA2397 / DNA Voucher”. CUIC male dissected with male genitalia in polyethylene vial on pin. EMEC male with male genitalia in a Euparal mount pinned beneath the specimen; specimen also DNA-extracted with DNA deposited in OSAC and EMEC.

##### Diagnosis

This new species shares well-developed isodiametric microsculpture on the vertex, an indistinctly margined metasternal process (Fig. 8), and a pale apical elytral lunule with its putative closest relative, *B. jacksoniense* (Fig. 4). *B. tahitiense* can be distinguished from *B. jacksoniense* by pronotal configuration, whereby the posterior lateral seta that marks the hind angle is situated

much further forward, approximately 28% of the pronotal length in front of the medial posterior margin (as opposed to 20% in *B. jacksoniense*); the edge of the pronotum mediad of the posterior lateral seta in *B. tahitiense* is thus almost parallel to the pronotal edge in front of the seta, as opposed to almost parallel to the medial posterior margin as in *B. jacksoniense*. Whereas *B. jacksoniense* is characterized by fully developed flight wings and broad elytral humeri, *B. tahitiense* is brachypterous, with the elytral humeri narrowly rounded, the elytral cuticle thicker, and the discal elytral intervals markedly convex. The apex of the median lobe is lamellate and broad in *B. tahitiense* (Fig. 9), in contrast to the much narrower apex in *B. jacksoniense* (Fig. 11).

#### Description

*Head.* Eyes convex, dorsomedial margins convergent on frons, ocular ratio 1.64; frontal grooves broad and shallow, median area of frons slightly, convexly elevated; mentum tooth little projected, lateral margins defining an obtuse angle, apex tightly rounded; epilobes typical for *Bembidion*, large and triangular.

*Prothorax.* Pronotum constricted basally, MPW/BPW = 1.68; posterior margin slightly convex between apparent hind angles; disc convex from lateral marginal depressions, but flat medially where median longitudinal impression is finely incised; anterior transverse impression shallow, defining a slightly convex, smooth anterior callosity, the discal surface behind impression slightly depressed; front angles not protruded, rounded behind; lateral marginal depression widest at midlateral pronotal seta, narrowed evenly to terminus at lateral margin of median base; basal pronotal seta associated with a convex expansion of the lateral margin (Fig. 7), the expansion defining a slightly sinuate margin immediately anterad seta, the margin behind seta—corresponding to excavated laterobasal pronotal margin of *B. jacksoniense* (Fig. 4)—markedly concave; prosternal process unmargined, ventral surface indistinctly depressed medially, posterior surface convexly protruded posterad narrow portion between procoxae.

*Elytra* obovate, humeri rounded, broadest at level of anterior lateral elytral series, progressively narrowed to apex; parascutellar seta present just mesad base of sutural stria 1; sutural interval 1 of similar convexity to intervals 2–5 basally, more convex, callous-like, meeting at an elevated suture on disc; anterior series of lateral elytral setae comprising four setae, the posterior seta separated from the third by twice the distance that separates the anterior three setae; posterior series of lateral elytral setae comprising four equidistantly separated setae; both apical and subapical elytral setae present (Fig. 6).



*Pterothorax.* Metepisternum moderately elongate, mesal margin along metasternum 1.75× distance measured perpendicularly from lateral margin to angle immediately laterad mesocoxa; metathoracic flight wing stenopterous, a club-shaped strap, of length 4× basal breadth with remnants of the Sc+R and M veins visible in the alar membrane.

*Abdomen.* Five ventrites visible, the plesiomorphically present suture between sternites III and IV (i.e., ventrites 2 and 3) indicated only as one of three short transverse depressions along the length of the fused sternites; ventrites 2–5 of males with a single seta each side of midline, ventrite 5 with an additional medial field of 6–8 very short, irregularly distributed setae.

*Colouration.* Frons and vertex rufopiceous dorsally with silvery reflection due to microsculpture, clypeus and labrum more rufous; mandibles rufoflavous, antepenultimate segment of maxillary palpi smoky flavous, apical two palpomeres smoky brunneous; antennomere 1 flavous, antennomeres 2–3 progressively darker to match smoky brunneous antennomeres 4–11; ventral surface of head capsule rufous; pronotal disc rufopiceous, median base more rufous to match concolorous, dark rufous elytral disc; elytral lateral marginal depression narrowly paler, rufobrunneous in anterior half of elytra, broadly flavous in apical 1/3 along flavous apical lunule, the lunule spanning intervals 5–8 along its anterolateral border; sutural interval concolorous with disc in basal half, rufoflavous to flavous in apical half; ventral body surface including coxae rufous, proepipleuron and elytral epipleuron contrastedly rufoflavous; legs including trochanters flavous, tibiae and tarsi with indistinct rufous cast.

*Microsculpture.* Frons, vertex and clypeus with upraised, granulate isodiametric sculpticells, the isodiametric microsculpture smoother on labrum (assessed at 125×); pronotal disc with indistinct microsculpture, isodiametric sculpticells best developed in depressed area just behind anterior transverse impression, isodiametric sculpticells in transverse rows traceable laterally and basally; discal elytral intervals glossy, indistinct isodiametric sculpticells present laterally in anterior half, entire apical portion glossy; metasternum with transverse sculpticells (Fig. 8); abdominal ventrites covered with well-developed transverse mesh microsculpture.

*Male genitalia.* Aedeagal median lobe robust, bilaterally flattened apically, with apex broadly rounded (Figs 9, 10); flagellum of central sclerite complex left lobe short, robust, ventral sclerite patch well defined, brush sclerite short; apex of ostium with a stout spur (Fig. 9); right paramere narrowed apically, but margins parallel to tightly rounded apex, two apical setae and a third more basally along ventral margin (Fig. 13); left paramere broad to bluntly rounded apex, two longer setae at ventroapical margin, a third smaller seta basally along ventral margin (Fig. 14).

### *Derivation of specific epithet*

The adjectival species epithet *tahitiense* signifies the island of Tahiti where this species was discovered.

### *Distribution and habitat*

The type specimens were discovered at 705 m elevation along the Faatautia River, Mont Mauru, eastern Tahiti Nui. The beetles were found in a thin layer of mosses and liverworts growing on the basaltic bank of the river. The habitat was at the lower end of an uncollapsed portion of lava tube, the remaining tube portion supporting a shade tolerant community along the river banks. The microhabitat was about 1 m above the water surface when the beetles were discovered, though this area was well under water at other times when this site was visited, the high water caused by extensive rainfall upslope. An adult specimen of *Mecyclothorax poro* Liebherr was also collected from the moss-covered bank housing the *Bembidion* beetles. The collecting site is figured in Liebherr (2012b, fig. 2A

[[http://www.pensoft.net/J\\_FILES/1/articles/3797/export.php\\_files/ZooKeys-227-063-g002.jpg](http://www.pensoft.net/J_FILES/1/articles/3797/export.php_files/ZooKeys-227-063-g002.jpg)].

### **Discussion**

Discovery of the first representative *Bembidion* species from the Society Islands raises the question of the biogeographic origin of its ancestral colonist. Based on current knowledge of *Bembidion* diversity in light of the above phylogenetic hypothesis, that origin was Australia, the present range of the sister species *B. jacksoniense*. That *B. jacksoniense* is a species characterized by a fully developed flight apparatus suggests any propagule drawn from *B. jacksoniense* populations would consist of flight-capable individuals. That the common ancestor of *B. jacksoniense* and *B. tahitiense* had fully developed wings is corroborated by the observation that their sister group (the species triplet *B. niloticum* Dejean, *B. foveolatum* Dejean, and *B. riverinae* Sloane), as well as the next most distantly related taxa (*B. pocularis* Bates and *B. sphaeroderum* Bates) (Fig. 1) all have full hind wings. It is thus most parsimonious to assume that the ancestral *Sloanephila* was also fully winged.

Based on relationships of the Tahitian *B. tahitiense* to the Australian *B. jacksoniense*, and of the Hawaiian *Nesocidium* species to an ancestral member of the Australian-New Zealand *Ananotaphus* Complex, the Society and Hawaiian archipelagoes have experienced independent colonizations by *Bembidion* species from the southwest Pacific. In Hawaii there were presumably two separate colonization events, one that founded subgenus *Gnatholymnaeum* Sharp and the second that represented the founding event for subgenus *Nesocidium* (Fig. 1). The ancestral

colonist for *Gnatholymnaeum* was winged, based on presence of fully developed flight wings in *B. (G.) spurcum* (Sharp), and the greater likelihood that wings were lost in *B. (G.) blackburni* (Sharp) rather than regained in *B. spurcum*. For *Nesocidium*, similar arguments suggest that the colonist was assuredly flight capable, as the more generalized Hawaiian *Nesocidium*—*B. ignicola* Blackburn, *B. pacificum* Blackburn, and *B. teres* Blackburn—are either monomorphically fully winged or wing-polymorphic (Liebherr 2008). For Hawaiian *Bembidion*, then, colonization also involved a winged ancestor.

The biogeographic histories of Hawaiian and Society Island taxa in the carabid tribes Platynini and Moriormorphini also involve direct colonization from the southwest Pacific without any stepping-stone relationships. The former comprises species of the Hawaiian endemic platynine genus *Blackburnia* Sharp (Liebherr and Zimmerman 2000; Liebherr 2001, 2003, 2007; Liebherr and Short 2007) versus the Tahitian platynine fauna comprising only three species (Perrault 1977). Hawaiian colonization by *Blackburnia* is hypothesized to have occurred 28–30 Mya, long before the origin of Kauai at 5.1 Mya, based on the lineage’s phylogenetic relationships to other Australian and Pacific taxa (Liebherr 2005). The most generalized Hawaiian *Blackburnia* are characterized by fully-developed flight wings (Liebherr and Zimmerman 2000). Conversely, the three platynine species in the Society Islands are arranged phylogenetically into two groups: 1, the “*Colpodes*” grade sister species pair “*C.*” *anachoreta* Fairmaire and “*C.*” *eremita* Fairmaire that both exhibit variously foreshortened though fully venated flight wings; and 2, the fully winged *Metacolpodes monticola* (Fairmaire) (Liebherr 2005). Tahitian colonization by these two platynine stocks was hypothesized to have occurred no more than 4.5 Mya, with both sets of species derived from a geographically widespread array of fully-winged species.

Species of the genus *Mecyclothorax* Sharp, tribe Moriormorphini, comprise a third pair of diverse carabid radiations to have evolved in the Society and Hawaiian islands. The genus *Mecyclothorax* exhibits greatest phylogenetic disparity in Australia and adjacent areas such as New Caledonia, New Guinea and New Zealand. Additionally, the Australian distribution of the *Mecyclothorax* sister group—*Amblytelus* Blackburn and allied genera (Liebherr 2011)—supports an Australian origin for the Pacific *Mecyclothorax* radiations. In Tahiti and Hawaii, *Mecyclothorax* species occupy the younger islands, with Hawaiian colonization most likely to have occurred on Maui Nui (Liebherr and Krushelnycky 2011). The founder of the Hawaiian radiation is hypothesized to have been one or more individuals of common, geographically widespread and flight-capable Australian species, *M. punctipennis* Macleay (Britton 1948). In the Society Islands, *Mecyclothorax* species are known only from the younger islands of Tahiti and Moorea (Perrault

1978a, 1978b; Liebherr 2012a). As in Hawaii, the colonizing propagule is hypothesized to have been drawn from Australia's *M. punctipennis*. Character-state distributions among all Tahitian and Hawaiian species support independent colonization of the Society and Hawaiian Islands by *M. punctipennis* (Liebherr 2012a). The island *Mecyclothorax* radiations differ from those of the Hawaiian and Tahitian platynine genera in that all island species are characterized by brachyptery. Nonetheless, among the wingless species of both radiations, the most generalized species are characterized by wing rudiments that are elongate, extended well beyond the posterior margin of the metathorax, and possessing rudimentary subcostal, radial, medial, and cubital wing veins. In contrast, islandic *Mecyclothorax* species with highly reduced elytral humeri—such as those observed in *B. tahitiense*—are characterized by vestigial flight wings that are much shorter, often bearing only remnants of the subcostal, radial, and medial veins. Thus, the results of evolutionary reduction of flight wings can be observed among the array of present-day island species, though a flight-capable colonizing taxon is not known for either island chain. That the original island colonists were fully winged in both instances is supported by parsimony reconstruction of wing state among the island colonists, the fully winged *M. punctipennis*, and its fully winged closest relatives: *M. ambiguus* (Erichson) of eastern Australia and *M. rotundicollis* (White) of New Zealand (Moore 1984). Based on monomorphically full flight wings in these latter three species, parsimony reconstruction assigns a macropterous flight-wing configuration to the common ancestor of *M. punctipennis* and both island chain colonizing propagules.

Colonization events in the Pacific involving carabid beetles have unambiguously involved winged colonizing taxa. Zimmerman (1948: 58) presented wind-borne dispersal as the major means by which southwest Pacific lineages have colonized the mid-Pacific. Gillespie et al. (2012) confirmed this directionality for flight-capable Austral-Pacific insects and birds, adding an easterly colonization route from the Americas that has been used by passively ballooning spiders. The sole alternative hypothesis for Pacific colonization appropriate for carabid beetles involves transport over water in drifting vegetation. For example, based on the occurrences of some Hawaiian *Mecyclothorax* “under bark, in moss on trees, or in hollow logs,” (Britton 1948: 108) proposed that *Mecyclothorax* beetles colonized Hawaii in this manner. If this were so, flight-wing condition may be discounted, opening up the option for brachypterous ancestral taxa to have spawned island colonizing propagules. Yet the common ancestors for both Society and Hawaiian Island carabid radiations are most parsimoniously reconstructed as macropterous. Moreover the Hawaiian and Tahitian *Bembidion* beetles are forest floor or riparian species, not log inhabitants, and so *Bembidion* cannot be accommodated in Britton's hypothesis.

The prevalence of winged dispersants establishing carabid faunas in the mid-Pacific seemingly flies in the face of the absence of stepping-stone dispersal. But, if macroptery is essential to colonization but wings are evolutionarily reduced upon a lineage's arrival and early diversification in isolated, ecologically stable island archipelagoes (Wollaston 1877, Southwood 1977), then it is very unlikely that one Pacific-island radiation would spawn a subsequent geographically more isolated radiation. Direct colonization from a continental source is repeatedly observed among Tahitian and Hawaiian *Bembidion*, platynine Carabidae and *Mecyclothorax*. Therefore we conclude that winged carabid ancestors have aerially crossed the Pacific to independently colonize, diversify, but never leave the Society and Hawaiian islands.

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**Table 1.** Specimens from which DNA was newly sequenced; all seven examined gene fragments were sequenced from each of these four specimens. # is the David Maddison DNA Voucher number.

Species	#	Locality and collecting information
<i>B. (Gnatholymnaeum) blackburni</i> (Sharp)	2219	USA: Hawaii: Oahu Waianae Mtns. Mt. Kaala summit, 1220 m el. 13-VI-2002, D.A. & J.T. Polhemus
<i>B.(Nesocidium) ignicola</i> (Sharp)	2220	USA: Hawaii: West Maui I. Mt. Eke 24.v.1997, C.P. Ewing el. 4400'
<i>B.(Nesocidium) waialeale</i> Liebherr	2577	USA: Hawaii: Kauai Waialeale summit gulch, 18.v.2005 lot 05. el. 1530 m pyr. fog mossy <i>Dubautia</i> . J.K.Liebherr
<i>B. (Sloanephila) tahitiense</i> n.sp.	2397	French Polynesia: Tahiti: Mount Mauru, 18-ix.2006, lot 01. el. 705m. pyr. fog moss/liverworts along stream. J.K. Liebherr

**Table 2.** Support for and against various clades; numbers on the far left correspond to clade numbers shown in Fig. 1. ML: Maximum likelihood analysis; P: parsimony analysis. Numbers indicate the bootstrap support expressed as a percentage; check marks indicate that the clade is present in the optimal (maximum likelihood or most parsimonious) trees but with bootstrap value below 50; x indicates that a contradictory clade was present in the optimal (maximum likelihood or most parsimonious) trees but with bootstrap value below 50; negative values indicate bootstrap support for a contradictory clade. Boxes in gray to black indicate support for the clade, and boxes in pink to red indicate support against that clade, with darker colors indicating stronger support. Blank boxes indicate no support for or against the clade because of lack of resolution in the inferred trees.

	Clade	Combined		28S		18S		COI	
		ML	P	ML	P	ML	P	ML	P
1	<i>Sloanephila</i>	100	100	99	100	99	93		
2	<i>Nesocidium</i> + <i>Zecillen</i> <i>us</i>	100	100	94	93	x		x	
3	<i>Nesocidium</i> + <i>Zecillen</i> <i>us</i> + <i>Ananotaphus</i>	99	100	x		x			
4	<i>Ananotaphus</i> Complex	100	99	x	x	x		x	x
5	<i>Ananotaphus</i> Complex + <i>B. blackburni</i>	84	57	x <sup>a</sup>	a			x	x

	Clade	CAD		wg		ArgK		Topo	
		ML	P	ML	P	ML	P	ML	P
1	<i>Sloanephila</i>	100	100	100	100	100	100	100	97
2	<i>Nesocidium</i> + <i>Zecillen</i> <i>us</i>	92	92	61	71	78	91	x	
3	<i>Nesocidium</i> + <i>Zecillen</i> <i>us</i> + <i>Ananotaphus</i>	86	89	86	93	x	x	x	
4	<i>Ananotaphus</i> Complex	79	84		54	-53	-55		
5	<i>Ananotaphus</i> Complex + <i>B. blackburni</i>	x		x		x <sup>a</sup>	x <sup>a</sup>	-67	-59

<sup>a</sup> *B. blackburni* sister to all or part of the New Zealand subgenera *Zemetallina* and *Zeplataphus* in maximum likelihood and most parsimonious trees

**Fig. 1.** Maximum likelihood bootstrap tree showing only those clades appearing in 75% of the bootstrap replicates. Thick lines mark those clades that are present in both the maximum likelihood bootstrap tree and parsimony bootstrap tree with bootstrap percentages  $\geq 90$ . Numbers below branches indicate maximum likelihood bootstrap percentage / parsimony bootstrap percentage (values less than 50 indicated by “-“); values are shown on only the clades of relevance for the discussion. Pacific island taxa shown in blue, other members of the *Bembidion* Complex in orange, and Australian and New Zealand members of the *Ananotaphus* Complex in green. Other members of the *Bembidion* Series are shown in black, and outgroups in gray.

**Fig. 2.** A portion of tree of highest likelihood found (only Clade 6 from Fig. 1 shown). Scale bar: 0.1, as reconstructed by RAxML. See legend of Fig. 1 for information about colors.

**Figs 3-4.** Habitus. 3, *B. tahitiense* holotype (MNHN). 4, *B. jacksoniense*; “Wiluna W.A. / ix 25 1931 //Australia / Harvard Exp. / Darlington” (MCZ). Scale bars 0.5 mm.

**Figs 5-6.** Elytra of *B. tahitiense*. 5, left elytron, discal seta ed5 (arrow) and surrounding area. 6, Apex of right elytron. Scale bars 0.1 mm.

**Figs 7-8.** *B. tahitiense*, paratype voucher number DNA2397 (EMEC). 7, pronotum. 8, center of metasternum. Scale bars 0.1 mm.

**Figs 9-12.** Aedeagi of *B. (Sloanephila)* species, left lateral aspect (9 and 11) and right lateral aspect (10 and 12). 9-10, *B. tahitiense*, paratype Maddison voucher number DNA2397 (EMEC); 11-12, *B. jacksoniense*, Australia: Queensland, Langlo Crossing, 26°07'S 145°40'E, Maddison voucher number V100682 (OSAC). Scale bars 0.1 mm.

**Figs 13-14.** Parameres, paratype male (CUIC). 13, right paramere, right lateral view. 14, left paramere, left lateral view. Scale bar 0.1 mm.