

Rapid Communication**The Asian hemipteran *Brachyplatys subaeneus* (Westwood, 1837) (Hemiptera: Heteroptera: Plataspidae) in a protected area in Panama**Dumas Gálvez^{1,2,3,4,*}, Daniel Murcia-Moreno⁵, Yostin Añino⁶ and Carlos Ramos^{4,7}¹Programa Centroamericano de Maestría en Entomología, Universidad de Panamá, Panamá²Smithsonian Tropical Research Institute, Panamá³Sistema Nacional de Investigación, Panamá⁴Coiba AIP, Panamá⁵Centro Regional de Azuero, Universidad de Panamá, Panamá⁶Museo de Invertebrados G.B. Fairchild, Universidad de Panamá, Panamá⁷Departamento de Genética y Biología Molecular, Universidad de Panamá, Panamá

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OPEN ACCESS**Abstract**

We report the occurrence of *Brachyplatys subaeneus* (Westwood, 1837) on a new host plant species: the leguminose *Macroptilium longepedunculatum*. We discovered the bugs in the mouth of the river San Juan in Coiba National Park in Panama. We present the morphological and barcode species level identification. This insect pest normally attacks plants of economic importance within cultivated habitats and our report possibly highlights for the first time the ability of *B. subaeneus* to invade natural habitats.

Key words: Coiba, invasive species, pest, national park**Introduction**

The growing trade and expansion worldwide have facilitated the invasion and worldwide distribution of many insects considered pests, causing diseases to humans and agricultural crops (Bebber et al. 2014). The Asian hemipteran *Brachyplatys subaeneus* (Westwood, 1837) is an example of a pest with the potential to spread across tropical and subtropical regions (Rédei 2016). For instance, it was recently introduced to the Americas (Aiello et al. 2016; identification correction by Rédei 2016) and considered a pest for a variety of crops with reports from Panama (Añino et al. 2018; Lowry et al. 2013; Rédei 2016), Costa Rica (Carmona-Ríos 2019), Dominican Republic (Pérez-Gelabert et al. 2019), Colombia (Forero 2021), Ecuador (Añino et al. 2020), United States (Eger et al. 2020), Guadalupe (Streito and Étienne 2020), including reports on GBIF for Mexico, Jamaica and Colombia (www.gbif.org). Moreover, predictive models of distribution based on environmental variables foresee its spread across all the region between the south of the United States, Venezuela, Peru and north of Brazil (Anses 2021).



Figure 1. Location of Coiba National Park (square) in Panama, including site of the discovery of *Brachyplatys subaeneus* (blue circle) and approximate locations of previous reports as registered by Añino et al. (2018, red circle) and Zachrisson et al. (*pers. comm.*; orange circle). Map of Panama: Free vector map.

It is expected that its distribution will follow the spatial distribution of plantations that the bug normally attacks, which is the case reported by Añino et al. (2018) in plantations of pigeon pea (*Cajanus cajan*) in Panama. Overall, this bug has been reported attacking 34 species of host plants contained in 14 families (Añino et al. 2018, 2020; Rédei 2016; Zachrisson et al. *pers. comm.*). In fact, a review of all the host species reveals that almost all of them are species of economic importance cultivated for agriculture, forestry or as ornamentals. However, less is known about the capacity of the *B. subaeneus* to invade natural habitats, as those in protected areas. Here, we report *B. subaeneus* attacking a new host plant—*Macroptilium longepedunculatum* (Fabaceae)—within a protected area in the Coiba National Park, an island located on the Pacific side of Panama. We used morphological and Cytochrome Oxidase I sequences for barcode specimens identification.

Materials and methods

The discovery of the *B. subaeneus* occurred during field work (August 16, 2021) for sampling spiders in Coiba National Park, a protected area composed of 38 islands, which is a World Heritage Site (UNESCO 2005, Figure 1), with high endemism of plants, invertebrates and vertebrates (Ridgely and Gwyne 1992; Olson 2007; González et al. 2010; Guzman and Breedy 2012; Mendez-Carvajal 2012; Roubik and De Camargo 2012; Miranda et al. 2013; Flores et al. 2016). Except for the MiAmbiente station and the old prison camp, most of the island is unoccupied. We found the bugs specifically on the main island, in the mouth of the river San Juan (7°27.589'N; 81°43.343'W). An estuary and coastal ecosystem composed of low shrub vegetation and mangroves (*Rhizophora mangle* and *Laguncularia racemosa*). We discovered nymphs and adults when sampling, in a little beach, with a beating tray under shrubs at chest-height. The host *Macroptilium longepedunculatum* (Fabaceae) is a climber plant found growing on the ground (sandy soil) and over shrubs (Figure 2a) and belongs



Figure 2. Details of growth habit of the host plant *Macroptilium longepedunculatum* (a) and details on the morphology of *Brachyplatys subaeneus* (top: dorsal view; bottom: ventral view, b). Photographs by Dumas Gálvez.

to the *Macroptilium gracile* complex (Berlingeri et al. 2020). Inspection of plants isolated from the rest of the vegetation allowed us to confirm that this was in fact the host plant. We placed the sampled bugs in 97% ethanol for further identification in the laboratory and DNA extraction.

DNA was extracted using EZNA Tissue DNA kit (Omega Bio-tek, Inc. Georgia, USA) following the manufacturer's protocol. A 654-bp fragment of the COI was amplified with universal primers LCO1490 (5'-GGTCA ACAATCATAAAGATATTGG-3') and HCO2198 (5'-TAAACTTCAG GGTGACCAAAAATCA-3') (Folmer et al. 1994).

PCR amplification was performed in 30 μ l reactions containing 2 μ l of template DNA (~ 200 ng), 15 μ l of 2X PCR Taq Mastermix (Omega, Bio-Tek), 0.5 μ l (10 mM) of each primer and 12 μ l nuclease free water. The thermal profile was 5 min at 94 $^{\circ}$ C, and 35 cycles of 45 s at 94 $^{\circ}$ C, 45 s at 52 $^{\circ}$ C, followed by final extension of 5 min at 72 $^{\circ}$ C.

Amplified DNA fragments of the expected size were visualized in agarose gels containing GelRed (Biotium, catalogue 41003) and sent to Psomagen (Psomagen, Inc., Maryland, USA) for purification and sequencing. Sequences were edited and aligned using SEQUENCHER 5.4.6 software (Gene Codes Corporation, Ann Arbor, MI, USA). COI sequence was verified for the respective open reading frame and search in NCBI-BLAST (www.ncbi.gov) and BOLD Systems (www.barcodinglife.org) to find the closest related sequences.

Results

Our sampled individuals matched *B. subaeneus* described by Rédei (2016, Figure 2b). Mounted vouchers of the collected bugs were deposited at the

collection of the Programa Centroamericano de Maestría en Entomología at the University of Panama.

The sequences generated from two specimens were deposited in Genbank with accession number OK560114 and OK560115. The Blastn and BOLD results indicate that both sequences match the Genbank sequences with accession numbers MW847232 and KT447152 from samples identified as *Brachyplatys subaeneus* with identity values ranging between 98 and 99%. Furthermore, the default BOLD phylogenetic tree constructed with Kimura 2 parameter distance model clusters both sequences in the *B. subaeneus* clade.

Based on the number of bugs observed per sampling bout and the spatial distribution of the plant in the field site (roughly 30,000 m²), we estimate that the abundance of the bug in that area is at least of several thousand individuals.

Discussion

All previous reports of *B. subaeneus* in Panama were from the mainland in association to cultivated plants, particularly *Cajanus cajans* (Añino et al. 2018). Our finding of a new leguminous host plant is in line with the apparent preference of *B. subaeneus* for this family of plants (Añino et al. 2018, 2020; Anses 2021). Moreover, its occurrence in a native plant within a protected area indicates that it can persist in natural habitats as well. In that case, the development of measurements for population control of this bug in cultivated areas should consider the proximity to natural habitats that may provide refuge for the bug, perhaps similar to other systems (Coffey and Johnston 1997). Natural habitats would provide resources and an escape from human control measurements (e.g. biological control, Hawkins et al. 1993). Alternatively, or parallelly, this bug may pose a serious threat for native plants in protected areas (Liu et al. 2020), together with competing with native insects (e.g. Freitas et al. 2009).

Origins of the invasion remain unknown, and it is surprising that we found the bugs in a remote area of the island, with no human presence. Nevertheless, it is possible that the bug first established in the old prison camp which is around 6.0 km from the discovery site. In this old prison camp, prisoners used to culture *Cajanus cajans* among other plants (*pers. comm.* by island staff) which may have provided the initial source for establishment. However, the prison was shut down in 2004 and it is unknown when the last plantations of *C. cajans* were still present. Still, we did not detect the bug around areas with infrastructures (old prison, MiAmbiente station) where the plantations may have been. Given that the shortest distance to the mainland is approximately 23 km, the most likely entrance of the bug in the park was via boats transporting supplies for the staff in the island. Supporting this hypothesis is the fact that one of the authors (DG) also discovered the invasive ant *Tapinoma melanocephallum* in the island (MiAmbiente station), a species that can be frequently transported in cargos in sea and rivers (Wetterer 2009).

Our finding highlights the potential that protected areas offer a refuge for insect pests and raises the question on how to control pest populations inside protected areas where standard methods of control are prohibited (e.g., pesticides) or with risk of causing more issues (e.g. exotic species as biological control, Camacho-Cervantes et al. 2017). Therefore, our results point out at the need to develop new concepts on refuge theory (Berryman and Hawkins 2006) for invasive species that are able to colonize both crops and natural habitats (the refuge), using a large set of hosts and without or few local enemies other than humans (e.g., parasitoids).

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Authors' contribution

Sampling by DG and DM. Morphological species identification by YA. DNA barcoding by CR. Writing of draft by DG and DM. Editing of manuscript by all authors.

Ethics and permits

Sampling in the park was carried out under the permit ARB-017-2021.

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