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A new species of planthopper in the genus *Haplaxius* from Osa Peninsula in Costa Rica (Hemiptera: Fulgoroidea: Cixiidae)

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Abstract

Haplaxius is a large genus of New World cixiid planthoppers. The genus is of particular interest because *Haplaxius crudus* can transmit palm infecting phytoplasmas and the recent discovery of additional *Haplaxius* on palms during survey work highlights the need to fully understand the diversity of this genus on palms. Herein, a new species, *Haplaxius cotinga* **sp. n.**, is described from the Osa Peninsula in Costa Rica. This species is most similar to *H. deleter* from southern Panama, from which it differs mostly by features of the male terminalia. Molecular data for the cytochrome *c* oxidase subunit I (COI), 18S rRNA, and histone 3 (H3) gene is provided and demonstrates supplemental support for placing the novel taxon in *Haplaxius*.

Key words: new species, taxonomy, phylogenetics, planthopper

Resumen

Haplaxius es un género numeroso de chicharritas del Nuevo Mundo pertenecientes a la familia cixiidae. El género es de particular interés porque *Haplaxius crudus* puede transmitir los fitoplasmas que afectan a las palmeras. El reciente descubrimiento de otras especies bajo el mismo género *Haplaxius* en las palmeras durante un trabajo de investigación, resalta la necesidad de conocer más detalladamente la diversidad de este género. En este documento se describe una nueva especie, *Haplaxius cotinga* **sp. n.**, para la Península de Osa en Costa Rica. Esta especie es más similar a *H. deleter* del sur de Panamá y se diferencia principalmente por las características de su genitalia masculina. Se proporcionan datos moleculares para la subunidad I del citocromo c oxidasa (COI), el ARNr 18S y el gen de la histona 3 (H3), los que respaldan la ubicación del nuevo taxón en *Haplaxius*.

Introduction

Haplaxius (Cixiidae: Oecleini) is a large genus (66 species) of New World planthoppers spread from southern Canada to northern South America, including the Caribbean (Barrantes *et al*. 2021, Bourgoin 2022). *Haplaxius* is defined by features of the forewing, face and hind leg; the bases of longitudinal veins Sc+R and MP form a common

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stalk so that only two veins appear to arise from the basal cell (a tribal feature, Muir 1922), frontal view of frons diamond-shaped with median ocellus, nearly straight frontoclypeal border, and lacking lateral teeth on hind tibia (Emeljanov 2007). *Haplaxius* may be heterogenous based on high variability in the male terminalia (see Kramer (1979). Recently, the genus *Myxia* Bahder & Bartlett 2019 was described as similar to *Haplaxius* but differing according to molecular data (viz. COI and 18S) and morphology (especially the male terminalia, most notably the medioventral lobe of the pygofer being triangular, the phallobase prolonged into projection ventrad of the aedeagus, and the anal tube short, stout and distally downcurved) (Bahder *et al.* 2019, Echavarria *et al.* 2021a, b).

Haplaxius is of particular interest because *H. crudus* is the vector of the lethal yellowing (LY) phytoplasma, '*Candidatus* Phytoplasma palmae' (Howard & Thomas 1980) and lethal bronzing (LB) phytoplasma, '*Ca.* P. aculeata' (Mou *et al*. 2022). The ability of other species of *Haplaxius* to transmit these phytoplasmas seems likely, but has not been established. However, the discovery of new species of *Haplaxius* from palms (e.g., Bahder *et. al.* 2020, Barrantes *et. al.* 2021) emphasizes the need to better understand the diversity of palm associated planthoppers and their importance in phytoplasma transmission ecology.

During an expedition to the Osa Peninsula in Costa Rica in 2021, a *Haplaxius* was collected at a light trap and subsequently found to be a new species. Here this novel species is described and sequence data generated for cytochrome *c* oxidase subunit I (COI), 18S rRNA, and histone 3 (H3) genes for constructing an updated phylogeny among available New World Oecleini is provided.

Materials and methods

Locality and specimen collection. Individuals of the novel taxon were aspirated directly from the sheet of a mercury vapor light trap and were immediately transferred to 95% ethanol. Specimens were collected (permit no. SINAC-ACOSA-D-R-060-2021) at La Cotinga Biological Station on 7-VII-2021, Puntarenas province, Costa Rica (8.621825, -83.478819), and exported from Costa Rica under permit number CUSBSE-659-2021, brought to the U.S.A. under permit number P526-170201-001. All specimens collected were measured, photographed and dissected using a Leica M205 C stereoscope and Leica DFC25 camera. Images of specimens and all features photographed were generated using the LAS Core Software v4.12. Voucher specimens, including primary types, are stored at the University of Florida—Fort Lauderdale Research and Education Center (FLREC) in Davie, FL, U.S.A and the Florida State Collection of Arthropods (FSCA) in Gainesville, FL, U.S.A.

Morphological terminology and identification. Morphological terminology generally follows Kramer (1979) except with male terminalia nomenclature updated after Bourgoin (1988) and Bourgoin & Huang (1990) and forewing venation following Bourgoin *et al*. (2015). New taxa are to be attributed to Bahder and Bartlett.

Dissections and DNA extraction. The terminalia that were dissected also served as the source of tissue for DNA extraction. The terminal end of the abdomens was removed and placed directly into a solution of tissue lysis buffer (buffer ATL) and proteinase K (180 µl ATL and 20 µl proteinase K) from the DNeasy® Blood and Tissue Kit (Qiagen). The abdomen was left to lyse for 24 hours at 56°C. Following lysis, eluate was transferred to a new 1.5 ml microcentrifuge tube and DNA extraction proceeded as per the manufacturer's instructions. The terminalia were then immersed in 200 µl of buffer ATL and 200 µl of buffer AL from the same kit and placed at 95°C for 24 hours to remove fat, wax, and residual tissue. The cleared genitalia were then used for morphological characterization and photography.

PCR parameters, sequence data, and analysis. To obtain COI, 18S, and H3 sequence data, previously published primers were used in all PCR reactions (Table 1). PCR reactions contained 5x GoTaq Flexi Buffer, 25 mM $MgCl_2$, 10 mM dNTP's, 10 mM of each primer, 10% PVP-40, and 2.5U GoTaq Flexi DNA Polymerase, 2 µl DNA template, and sterile dH_20 to a final volume of 25 µL. Thermal cycling conditions for all loci involved were as follows: 2 min initial denaturation at 95°C, followed by 35 cycles of 30 sec denaturation at 95°C, 30 sec annealing, and extension at 72°C. Specific annealing temperatures and extension times for respective loci are presented in Table 1. Products were visualized on a 1.5% agarose gel stained with GelRed (Biotium). PCR products of the appropriate size were purified using the ExoSAP-IT™ Express PCR Product Cleanup Reagent per the manufacturers' protocol (ThermoFisher Scientific, Waltham, Massachusetts, USA). Purified PCR product was quantified using a NanoDrop Lite Spectrophotometer (ThermoFisher Scientific, Waltham, Massachusetts, USA) and sequenced using the SeqStudio Genetic Analyzer (Applied Biosystems). Contiguous files were assembled using DNA Baser (Version

4.36) (Heracle BioSoft SRL, Pitesti, Romania), aligned using ClustalW as part of the package MEGA7 (Kumar *et al.* 2016). Maximum Likelihood trees were generated using the Bootstrap method at 1,000 replicates based on the Tamura-Nei model for both the COI, 18S, and H3 loci as well as the consensus tree with concatenated data for COI and 18S data. A matrix of pairwise differences using number of differences among 18S and COI a subset of taxa within each genus was calculated with MEGA7 (Kumar *et al.* 2016). The bootstrap method was used for variance estimation at 1,000 replicates and using the p-distance model.

TABLE 1. Primers used to amplify corresponding gene regions used to assess placement of novel taxon and PCR parameters for each locus.

Locus	Primer	Direction	Sequence (5'-3')	Annealing	Extension	Reference
COI	COI_D1_F	Forward	GGAACWATAAGAAGWATAATYATYCG	40˚C	1 min. 30	Humphries
	C1-J-2195RC	Reverse	ACTTCTGGATGACCAAAAAATCAA		sec.	*et al.* 2021
18S	18SF	Forward	ACTGTCGATGGTAGGTTCTG	50˚C	2 min.	Bahder
	18SR	Reverse	GTCCGAAGACCTCACTAAA			*et al.* 2019
H3	H3F	Forward	CAGACGGCBMGKAARTCSACC	55˚C	30 sec.	Echavarria
	H3R	Reverse	GTKACHCKCTTRGCGTGRAT			*et al.* 2021a

Taxon sampling. For molecular comparisons, *Haplaxius crudus* (Van Duzee, 1907), *H. dougwalshi* Bahder & Bartlett 2020, *H. skarphion* (Kramer, 1979), *H. pocococo* Bahder & Bartlett 2021, *H. pictifrons* (Stål, 1862), *H. lunatus* (Van Duzee, 1909) were used for in-group comparisons and *Oecleus borealis* Van Duzee, 1912, *O. dormido* Bahder & Bartlett 2022, *O. mackaspringi* Bahder & Bartlett 2019, *Myxia belinda* Bahder & Bartlett 2019, *M. delta* (Kramer, 1979), *M. hernandezi* Bahder & Bartlett 2021, *M. baynardi* Bahder & Bartlett 2021, *Nymphocixia unipunctata* Van Duzee, 1923, *N. caribbea* Fennah, 1971 and *Melanoliarus chuliotus* (Ball, 1934) were used as outgroups. GenBank Accession numbers for all included taxa are provided in Table 2.

TABLE 2. Molecular taxon sampling and GenBank accession numbers.

		GenBank Accession No.			
Taxon	Locality	COI	18S	H3	Collection
Haplaxius crudus	Costa Rica	MT080284	MT002393	MZ274037	FLREC
Haplaxius dougwalshi	Costa Rica	MT080284	MT002395	MZ297815	FLREC
Haplaxius lunatus	Florida, U.S.A.	OM264285	OM258692	OM262388	FLREC
Haplaxius skarphion	Costa Rica	MT900603	MT892907	MZ274039	FLREC
Haplaxius pocococo	Costa Rica	MW086873	MW086509	OM262387	FLREC
Haplaxius pictifrons	Delaware, U.S.A.	MT946292	MN200098	MZ274038	FLREC
Myxia belinda	Costa Rica	MT900605	MN200095	MZ274041	FLREC
Myxia delta	Costa Rica	MT900602	MT892907	MZ274042	FLREC
Myxia hernandezi	Costa Rica	MZ234085	MZ262449	MZ274043	FLREC
Myxia baynardi	Costa Rica	MT900604	MT892909	MZ274040	FLREC
Nymphocixia unipunctata	Florida, U.S.A.	OM264284	OM258690	OM262389	FLREC
Nymphocixia caribbea	Jamaica	MT080286	MT002394	MZ274044	FLREC
Oecleus borealis	Florida, U.S.A.	OM264286	OM258691	OM262390	FLREC
Oecleus dormido	Costa Rica	OM264283	OM258693	OM262392	FLREC
Oecleus mackaspringi	Jamaica	MN488999	MN422261	MZ274045	FLREC
Melanoliarus chuliotus	Florida, U.S.A.	OM264287	OM258689	OM262392	FLREC

Systematics

Family Cixiidae Spinola, 1839

Subfamily Cixiinae Spinola, 1839

Tribe Oecleini Muir, 1922

Genus *Haplaxius* Fowler, 1904

Type species: *Haplaxius laevis* Fowler, 1904 (type species designation by Caldwell 1946: 203)

Diagnosis (modified from Bahder *et al*. 2020). Small to average size cixiids (3.2–6.4 mm); head in dorsal view narrower than pronotum, eyes large (elongate-oval, diagonally truncate posteriorly, usually with ventral emargination at antenna); vertex elongate, moderately broad (among Oecleini), vertex disc slightly concave, sides and apex carinate, median carina usually absent, apex variably produced beyond eyes (usually weakly projected). In lateral view, apex of head bluntly angled, ocellus beneath eye (anterior to antenna). In frontal view, sides of frons concave ("flared") and carinate, midline of frons carinate, interrupted near frontoclypeal suture by ocellus (ocellus sometimes absent), clypeus subtriangular with lateral margins and midline carinate. Antennal scape reduced, pedicel robust, flagellum beadlike basally and filamentous distally. Pronotum narrow, with irregular ridges and distinct paranotal region, length shortest at midline, posterior margin indented; tegulae evident. Mesonotum tricarinate; longitudinal midlength of mesonotum about two times that of vertex. Hind tibiae without lateral spines. Forewings tectiform, usually hyaline or transparent, but sometimes infuscated or patterned, veins usually with small setae-bearing pustules (tubercles). Male pygofer usually longest on ventral margin, hind margin variably produced. Aedeagus asymmetrical and elaborated with projections and processes, vertical connective articulating base of aedeagus with gonostyli (genital styles). Gonostyli symmetrical and usually simple. Anal tube symmetrical or asymmetrical, with processes from one or both ventral margins and sometimes with median ventral projections.

Haplaxius cotinga Bahder & Bartlett sp. n.

(Figures 2–9)

Type locality. La Cotinga Biological Station (8.621825, -83.478819), Puntarenas province, Costa Rica (Fig. 1).

FIGURE 1. General location of La Cotinga Biological Station (red circle) on the Osa Peninsula in Costa Rica where *Haplaxius cotinga* **sp. n.** was collected.

Diagnosis. A pale species (light green in life (Fig. 2), yellowish when preserved (Fig. 3), with whitish unmarked face and large reddish marking on abdominal terga. Male terminalia bearing gonostyli with sclerotized subapical dorsal ridge. Phallobase complex, bearing large, sinuous ventral projection with hooked apex and a large, elongate dorsal process. Anal segment in lateral view stout and of moderate length, ventral margin with large median process at midlength, apex elongate, strongly curved ventrad.

FIGURE 2. Live adult female of *Haplaxius cotinga* **sp. n.**

Description. *Color*. In life, body pale green, fading to yellow (in ethanol), nearly white on posterior 2/3 of vertex disc and genae excluding temple (yellow), ocelli (yellow with reddish highlights), and antennae (yellow); legs mostly white along with portions of pleuron and paranota; mesonotum paler on posterior margin and between lateral carinae. Forewings transparent, broadly but weakly infuscate except pale along leading margin (humeral region to pterostigma). Abdomen with dorsal portion of terga reddish (approximately from 2nd-5th apparent abdominal tergum).

Morphology. Body length (including wings), males: 2.99 mm (n = 1), females 2.99–3.02 mm (n= 5). Head. In dorsal view (Fig. 4A), slightly anteriorly projecting, anterior margin convex, posterior margin concave with median convexity (giving appearance of pair of concave dentations; Fig. 4A), lateral carinae foliate, disc convex, vertex length at midline about twice width at widest point (at posterior margin); widest basally, narrowing slightly at distal margin (Fig. 4A). In lateral view (Fig. 4B), head anteriorly rounded (slightly compressed), somewhat projected at level of ventral margin of compound eye (Fig. 4B). Face (in frontal view—Fig. 4C) with frons plus clypeus collectively ovate, lateral carinae foliate, median carina distinct, forked between compound eyes, frons distinctly expanding from between compound eyes to below level of antennae, constricting slightly at frontoclypeal suture; frontoclypeal suture straight; median ocellus small, translucent. Clypeus roughly triangular with median carina weak. Antennae bulbous, scape narrow, ring-like; pedicle spheroid, bearing many irregularly placed sensory plaques, flagellum elongate and bristle-like with a bulbous base. Lateral ocelli distinct, anterior to antennae beneath compound eye near leading margin.

Thorax. In dorsal view, pronotum narrow, convex at anterior margin, concave at posterior margin (Fig. 5A), median carina indistinct, lateral carinae extending to ventral margin at 2/3 length to ventrolateral margin. Mesonotum tricarinate, median carina evident, reaching posterior margin, lateral carinae subparallel for most of length, diverging posteriorly (Fig. 5A). Spinulation of hind leg 6-6-6.

Forewing. Forewing veins with setal pits (Fig. 6), apex of clavus past wing midlength (fusion of PCu and A1 in basal third of wing), composite vein Pcu+A1 reaching wing margin well before claval apex; fork of R (forming C1 cell) near wing midlength just distad of CuA fork; forewing branching pattern: RA 1-branched, RP 3-branched, MP 5-branched, CuA 2-branched with CuA connected with CuP by icu crossvein. A small cell, spurious, present along the distal margin of C5 at level of claval apex (absent in 3/5 females).

FIGURE 3. Adult male habitus of *Haplaxius cotinga* **sp. n.**; (A) dorsal view, and (B) lateral view, scale = 1 mm.

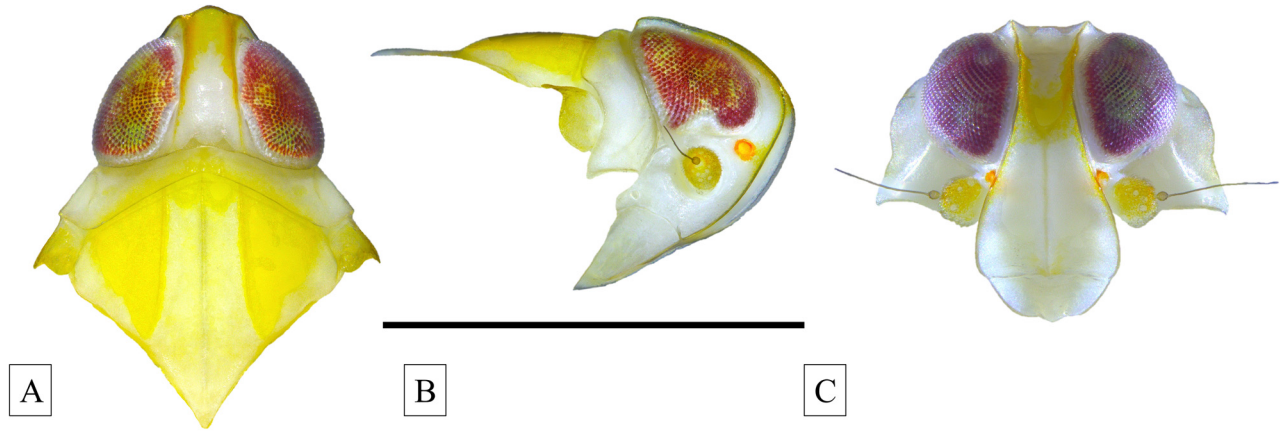

FIGURE 4. Adult male *Haplaxius cotinga* **sp. n.**; (A) head, pronotum and mesonotum dorsal view, (B) head, pronotum, and mesonotum lateral view, and (C) head and pronotum frontal view; scale = 1 mm; italics = crossvein, green = cell.

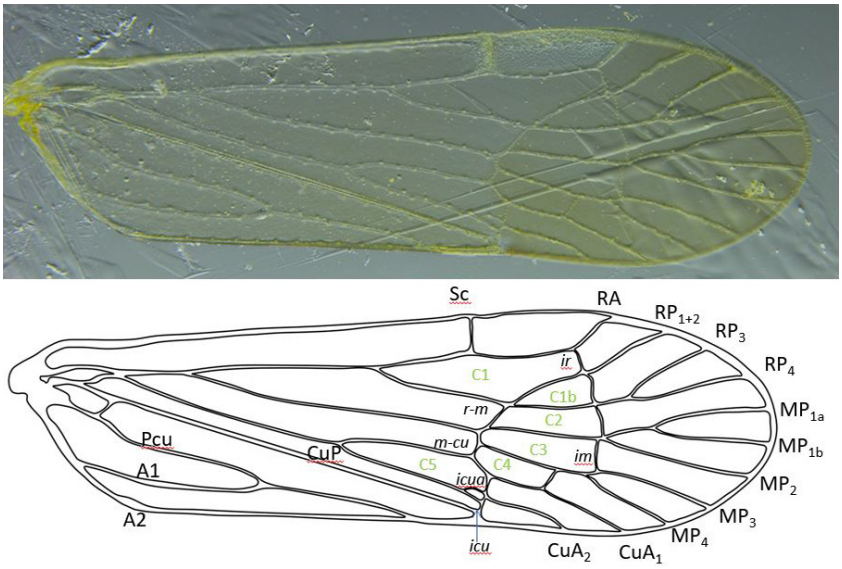

FIGURE 5. Forewing venation of *Haplaxius cotinga* **sp. n.**; black = vein, italics = crossvein, green = cell.

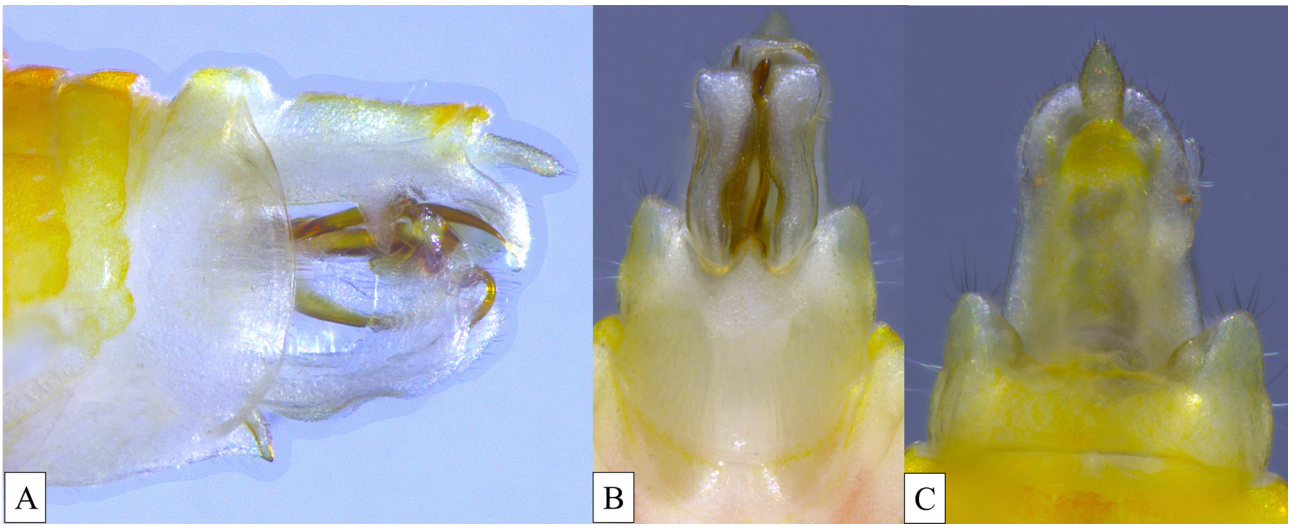

FIGURE 6. Male *Haplaxius cotinga* **sp. n.** terminalia; (A) lateral view, (B) ventral view, and (C) dorsal view.

Terminalia. Pygofer, in lateral view, irregularly triangular, narrowest at dorsal margin, widest at ventral margin, anterior margin sinuate, posterior margin convex (Fig. 7A, 8A). Medioventral process (in lateral view) scoop-like, in ventral view, spade-like to sagittate, apex acutely pointed (Fig. 7B, 8B). Gonostyli in lateral view strongly expanded distally, sinuate ventrally, dorsally strongly curved to bulbous apex (Fig. 7A, 8A), dorsoapical margin with two sclerotized dorsal projections, one arising approximately at 2/3 length and second at apex, lightly sclerotized between (Fig. 7A, 8A); in ventral view, inner and outer margins irregularly sinuate, narrowest basally, expanding approximately to twice the width of basal half, apically truncate, inner margins with irregular median serrulations in distal half (Fig. 7B, 8B). Aedeagus approximately straight (in lateral view, Figs. 9A, 10A) for most of length, then angled downward to ventral lobe (A1) followed by dorsal inflection (A2). Phallotheca incompletely enclosing aedeagus, mostly membranous with sclerotized portions, bearing large projection traversing aedeagus dorsum to form large dorsal process (P1), ventral sclerotization with large apical strongly curved process (P2) (Figs. 7 & 8). Anal segment in lateral view stout (not 'stalklike', viz. Kramer 1979) moderate length, ventral margin with large median process at midlength, apex elongate, strongly curved ventrad (Figs. 5A, 6A); in dorsal view broad with rounded apex (Figs. 5C, 6C). Anal column tubular, elongate.

Plant associations. Unknown, collected at light trap. Habitat was an abandoned pasture/grazing land being re-forested with hardwood trees.

Distribution. Costa Rica (Puntarenas), Osa Peninsula.

Etymology. The specific name given is in reference to La Cotinga Biological Station where the specimens were collected.

Material examined. Holotype male "Costa Rica, Puntarenas / La Cotinga Biological Station / 7.VII.2021 / Light trap near dorms / Coll.: B.W.Bahder // Holotype / *Haplaxius cotinga* ♂" (FLREC); Paratypes, La Cotinga Biological Station [7.VII.2021] (1 male, 1 female—FSCA, 1 female—FLREC).

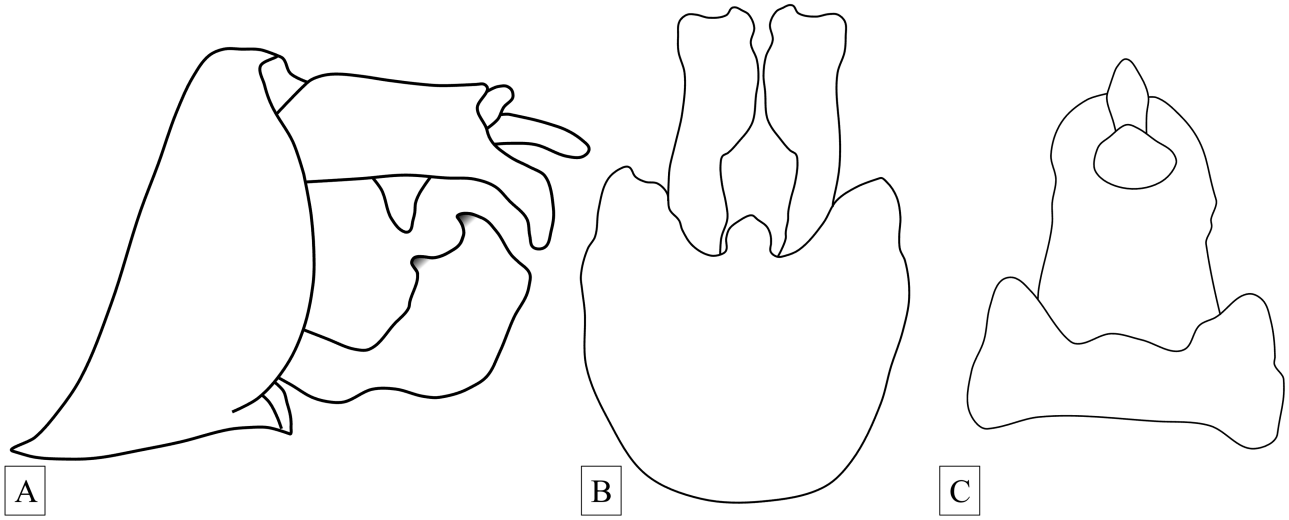

FIGURE 7. Male *Haplaxius cotinga* **sp. n.** terminalia line-art; (A) lateral view, (B) ventral view, and (C) dorsal view.

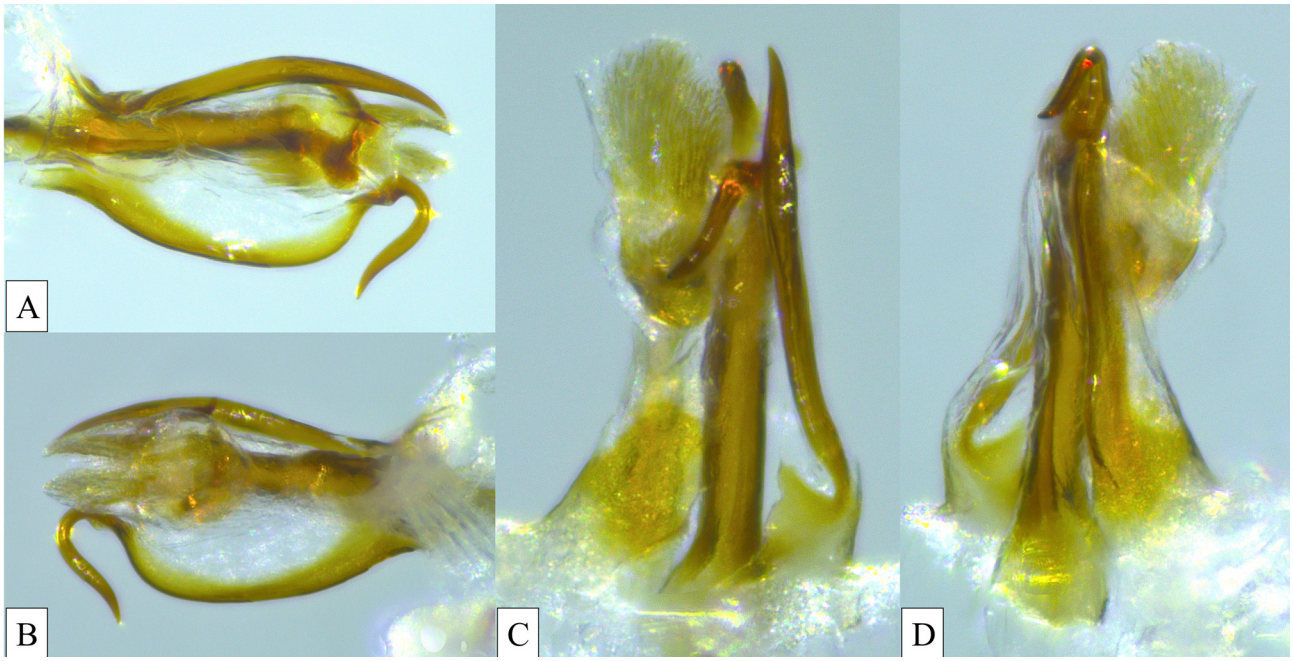

FIGURE 8. Aedeagus of *Haplaxius cotinga* **sp. n.**; (A) left lateral view, (B) right lateral view, (C) dorsal view, and (D) ventral view.

Sequence Data. For the COI gene, a 646 bp product was generated (GenBank Accession No. ON763279), for 18S, a 1,403 bp product was generated (GenBank Accession No. ON758370) and for the H3 gene, a 347 bp product was generated (GenBank Accession No. ON755134). Based on the phylogenetic analyses performed for the 18S and H3 gene, there is strong bootstrap support for the monophyly of *Haplaxius*, 97 and 94 respectively (Fig. 9). *Haplaxius cotinga* **sp. n.** resolves within this clade for both 18S and H3 but separate from all other *Haplaxius* species analyzed. The placement of *Haplaxius cotinga* **sp. n.** is further supported in the consensus tree based on all three loci (Fig. 9D), where it resolves with weak support (49) adjacent to *H. pictifrons*. For the COI, there is weak bootstrap support at all branches (<80) except in the instances of closely related taxa (Fig. 9A). The pairwise comparison based on the 18S gene shows that the average level of variation among species within a genus is 0.77%

(SE±0.1) for *Haplaxius*, 0.37% (SE±0.1) for *Oecleus*, 1% (SE N/A) for *Nymphocixia* and 0.63% (SE±0.3) for *Myxia* based on the taxa analyzed (Table 3). The average variation among genera for the 18S gene is 2.1% (SE±0.1), ranging from 1.3% to 2.7% with *Haplaxius cotinga* **sp. n.** differing on average by 0.53% (SE±0.2) from other species of *Haplaxius*, 1.77% (SE±0.1) from species of *Oecleus*, 1.5% (SE±0.2) from species of *Nymphocixia* and 1.9% (SE±0.0) from species of *Myxia* (Table 3). The pairwise comparison based on the COI gene shows an average of 15.6% intrageneric variability whereas there was on average, 18.9% intergeneric variability (Table 4). While there does appear to be some distinction at the generic level for COI based on nucleotide variability, this distinction is not as pronounced and consistent as that seen with 18S, which to some extent, is also reflected in the phylogenies. In addition, there are taxa where this value overlaps (*M. belinda* and *M. hernandezi* as well as with *Nymphocixia*), further highlighting that COI is not a suitable marker for phylogenies in the currently analyzed Oecleini but should be reserved for species delimitation and population genetic studies.

TABLE 3. Pairwise comparison showing estimates of evolutionary divergence between sequences based on the 18S rRNA gene for *Haplaxius cotinga* **sp. n.** demonstrating intrageneric (orange) and intergeneric (blue) variability; the number of base differences per site from between sequences are shown. Standard error estimate(s) are shown above the diagonal and were obtained by a bootstrap procedure (1000 replicates).

		1	2	3	4	5	6	7	8	9	10	11
1	*Haplaxius cotinga* **sp. n.**		0.002	0.002	0.004	0.003	0.003	0.004	0.003	0.004	0.003	0.004
2	*Haplaxius pocococo*	0.007		0.002	0.004	0.004	0.004	0.004	0.004	0.004	0.004	0.004
3	*Haplaxius crudus*	0.004	0.005		0.004	0.004	0.004	0.004	0.003	0.004	0.003	0.003
4	*Oecleus borealis*	0.019	0.025	0.021		0.002	0.001	0.004	0.004	0.004	0.004	0.004
5	*Oecleus mackaspringi*	0.016	0.022	0.019	0.006		0.002	0.004	0.004	0.004	0.004	0.004
6	*Oecleus dormido*	0.018	0.023	0.019	0.001	0.004		0.004	0.004	0.004	0.004	0.004
7	*Nymphocixia unipunctata*	0.017	0.022	0.019	0.026	0.024	0.025		0.003	0.004	0.004	0.004
8	*Nymphocixia caribbea*	0.013	0.016	0.013	0.024	0.021	0.022	0.010		0.004	0.004	0.004
9	*Myxia hernandezi*	0.019	0.022	0.019	0.025	0.025	0.023	0.027	0.022		0.001	0.003
10	*Myxia delta*	0.019	0.021	0.019	0.025	0.024	0.024	0.027	0.021	0.001		0.003
11	*Myxia belinda*	0.019	0.023	0.019	0.023	0.022	0.022	0.026	0.022	0.009	0.009	

TABLE 4. Pairwise comparison showing estimates of evolutionary divergence between sequences based on the COI gene for *Haplaxius cotinga* **sp. n.** demonstrating intrageneric (orange) and intergeneric (blue) variability; the number of base differences per site from between sequences are shown. Standard error estimate(s) are shown above the diagonal and were obtained by a bootstrap procedure (1000 replicates).

		1	2	3	4	5	6	7	8	9	10	11
1	*Haplaxius cotinga* **sp. n.**		0.015	0.015	0.015	0.016	0.015	0.016	0.015	0.017	0.017	0.016
2	*Haplaxius pocococo*	0.153		0.014	0.015	0.016	0.015	0.017	0.016	0.016	0.016	0.016
3	*Haplaxius crudus*	0.139	0.139		0.016	0.016	0.016	0.016	0.016	0.016	0.017	0.017
4	*Oecleus borealis*	0.164	0.166	0.186		0.015	0.015	0.017	0.016	0.017	0.017	0.017
5	*Oecleus mackaspringi*	0.188	0.168	0.181	0.159		0.014	0.017	0.016	0.016	0.016	0.017
6	*Oecleus dormido*	0.166	0.161	0.168	0.146	0.141		0.016	0.016	0.016	0.015	0.016
7	*Nymphocixia unipunctata*	0.164	0.190	0.175	0.201	0.197	0.177		0.015	0.017	0.016	0.017
8	*Nymphocixia caribbea*	0.161	0.168	0.175	0.181	0.190	0.177	0.164		0.017	0.016	0.018
9	*Myxia hernandezi*	0.221	0.193	0.192	0.206	0.177	0.190	0.223	0.204		0.013	0.015
10	*Myxia delta*	0.208	0.190	0.193	0.206	0.192	0.177	0.197	0.203	0.120		0.017
11	*Myxia belinda*	0.177	0.192	0.206	0.204	0.199	0.206	0.232	0.212	0.186	0.192	

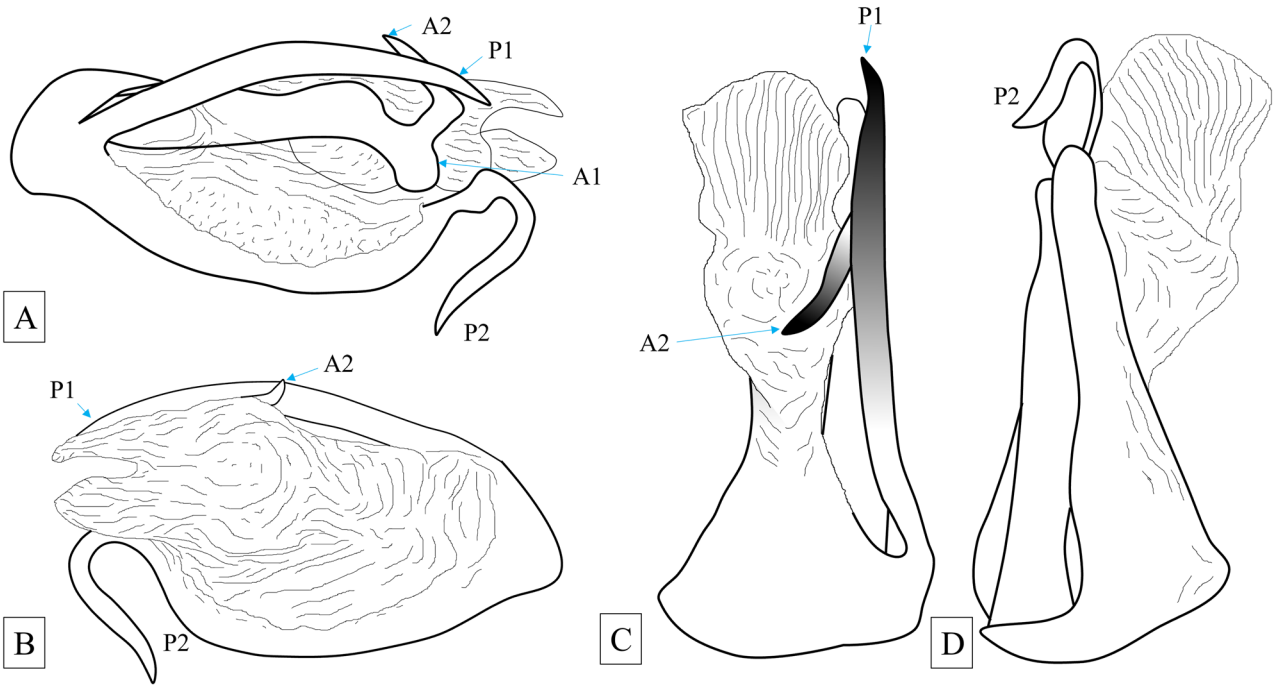

FIGURE 9. Aedeagus line-art for *Haplaxius cotinga* **sp. n.**; (A) left lateral view, (B) right lateral view, (C) dorsal view, and (D) ventral view.

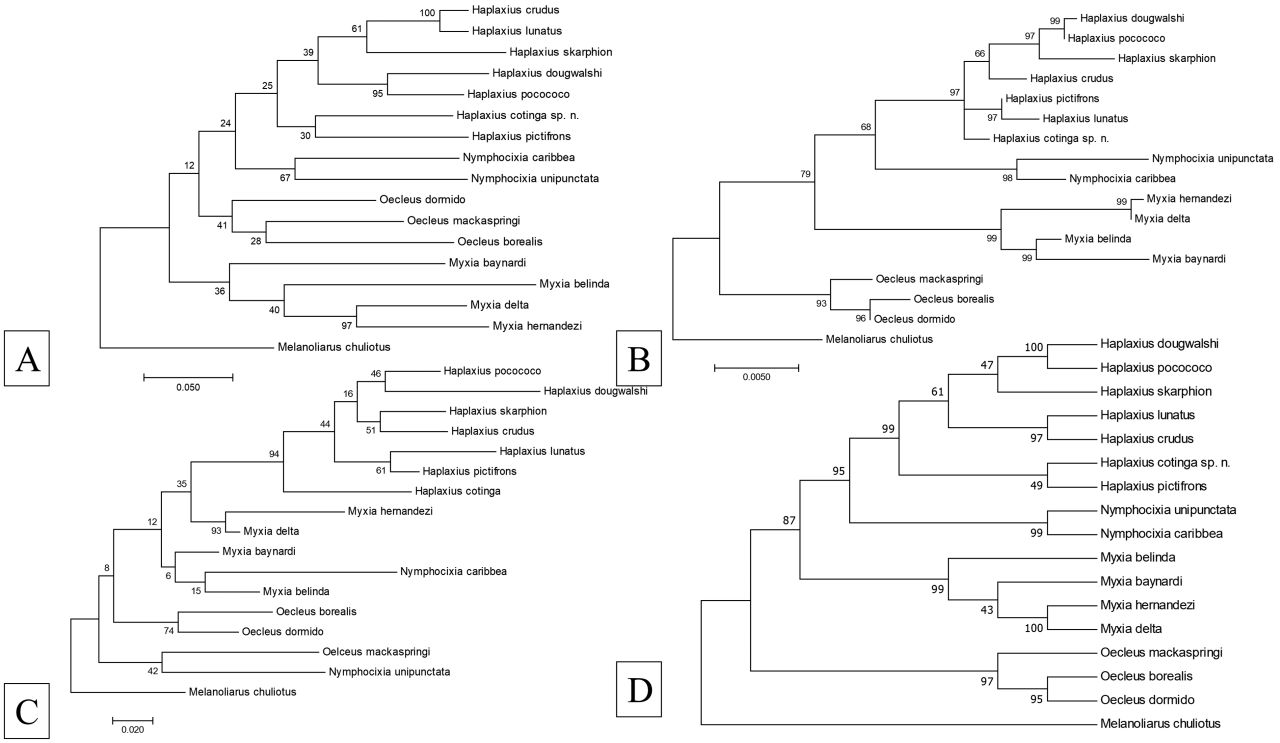

FIGURE 10. Maximum likelihood phylogenetic tree based on 1,000 replicates: A) COI gene, B) 18S rRNA gene, C) H3 gene, and D) consensus tree of concatenated COI, 18S, and H3 sequences; scale bar = percent nucleotide difference.

Remarks. Morphological characters and molecular data presented support the placement of the novel taxon in *Haplaxius*. The closest species to *Haplaxius cotinga* **sp. n.** appears to be *H. deleter* Kramer, 1979 (described from southern Panama). Both species have the same general form of the terminalia (especially the gonostyli and aedeagus), and both are similar in general coloration (*H. deleter* described by Kramer 1979: 369 as "ground color of head and pronotum pale chalky green, anterior portion of crown lightly washed with orange, face unmarked…"). The

two species would key in different directions in Kramer (1979) because *H. cotinga* **sp. n.** has a median projection on the anal tube that is absent in *H. deleter* (*H. deleter* instead bears a quadrate process about midlength of the left lateral margin of the anal tube, absent in *cotinga* **sp. n.**). In *H. deleter*, the gonostyli appear more slender with the dorsal projections much more separated, longer and less curved than observed in *H. cotinga* **sp. n.** Additionally, the curvature of the apical process of the aedeagus is different where it is angled distad in *H. deleter* and cephalad in *H. cotinga* **sp. n.** and the ventral process of the phallobase is distinctly angled caudad in *H. deleter* and less sinuate whereas the process in *H. cotinga* **sp. n.** is strongly sinuate, curving cephalad. Finally, in *H. cotinga* **sp. n.** the flagellar apex is bifurcated in lateral view, where is rounded in *H. deleter*. Other features that distinguish *H. cotinga* **sp. n.** from *H. deleter* is the size (*H. deleter* adult males reported at 5.0 mm while *H. cotinga* **sp. n.** adult males are around 3 mm with wings).

Discussion

Haplaxius cotinga **sp. n.** represents the 8th *Haplaxius* species recorded from Costa Rica (viz., *H. akko* (Kramer, 1979), *H. crudus*, *H. dougwalshi*, *H. pocococo*, *H. phylax* (Kramer, 1979), *H. simplicatus* Caldwell, 1946 and *H. skarphion* are also reported). *Haplaxius* is an important genus because of its potential as phytoplasma vector, especially among palms. Host associations are reported from very few *Haplaxius* species, although *H. crudus*, *H. hochae* O'Brien, 2006, *H. rubidus* (Ball, 1933) and *H. skarphion* are reported from Arecaceae (Caldwell 1946, Kramer 1979, O'Brien 2006, Bahder *et al*., 2020). The discovery of a new species of *Haplaxius* adds helpful data for evaluating the evolution and phylogeny of this interesting group of planthoppers, especially because this species appears to be morphologically distinct from all known *Haplaxius* (with the exception of *H. deleter*) while resolving well within *Haplaxius* based on current molecular markers. While there is no data on other species of *Haplaxius* being competent palm-infecting phytoplasma vectors, aside from *H. crudus*, other palm feeding *Haplaxius* species may yet be regionally important for transmission of local strains of phytoplasma.

Similar to *Haplaxius cotinga* **sp. n.** and *H. deleter* is a species encountered in specimens from La Selva Research Station (Heredia Province, Costa Rica). This species was initially thought to be *H. deleter* but differs in features of the terminalia, most notably that the process on the right lateral margin of the anal tube is triangular in the La Selva specimens but quadrate in *H. deleter.* We have, so far, been unable to obtain sequence data from available La Selva specimens, and *H. deleter* is unavailable, but we hope to further consider these taxa in future work.

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References

Ball, E.D. (1933) The genus *Myndus* Stål in North America (Homoptera: Fulgoridae). *Journal of the Washington Academy of Sciences*, 23, 478–484.

Ball, E.D. (1934) The genus *Oliarus* and its allies in North America (Homoptera: Fulgoridae). *Journal of the Washington Academy of Sciences*, 24, 268–276.

Bahder, B.W., Bartlett, C.R., Barrantes, E.A.B, Echavarria, M.A.Z., Humphries, A.R., Helmick, E.E., Goss, E.M. & Ascunce, M.S. (2019) A new genus and species of cixiid planthopper (Hemiptera: Auchenorrhyncha: Fulgoroidea) from the Reserva Privada el Silencio de Los Angeles Cloud Forest in Costa Rica. *Zootaxa*, 4701 (1), 65–81. https://doi.org/10.11646/zootaxa.4701.1.5

Bahder, B.W., Barrantes, E.A.B., Echavarria, M.A.Z., Mou, D.-F., Helmick, E.E. & Bartlett, C.R. (2020) A new species of planthopper in the genus *Haplaxius* (Hemiptera: Auchenorrhyncha: Fulgoroidea: Cixiidae) on palms in Costa Rica and a new country record for *Haplaxius skarphion*. *Zootaxa*, 4767 (4), 543–552. https://doi.org/10.11646/zootaxa.4767.4.4

Barrantes, E.A.B., Echavarria, M.A.Z., Bartlett, C.R., Helmick, E.E. & Bahder, B.W. (2021) A new species of planthopper in the genus *Haplaxius* (Hemiptera: Auchenorrhyncha: Fulgoroidea: Cixiidae) from coconut palm (*Cocos nucifera*) in Costa Rica. *Zootaxa,* 4963 (3), 412–422.

https://doi.org/10.11646/zootaxa.4963.3.2
Barrantes, E.A.B., Echavarria, M.A.Z., Bartlett, C.R., Helmick, E.E. & Bahder, B.W. (2022) A new species of *Oecleus* (Hemiptera: Auchenorrhyncha: Fulgoroidea: Cixiidae) from the Caribbean coast of Costa Rica and a reassessment of the generic status of *Nymphomyndus*. *Zootaxa*, 5133 (4), 509–526.
https://doi.org/10.11646/zootaxa.5133.4.3
Bourgoin, T. (1988) A new interpretation of the homologies of the Hemiptera male genitalia illustrated by the Tettigometridae (Hemiptera, Fulgoromorpha). *In*: Vidano, C. & Arzone, A. (Eds.). *Proceedings of the 6th Auchenorrhyncha Meeting, Turin, Italy, 7–11 September 1987*. Consiglio Nazionale delle Ricerche, IPRA, Rome, pp. 113–120.
Bourgoin, T. (2022) FLOW (Fulgoromorpha Lists on The Web): a world knowledge base dedicated to Fulgoromorpha. Version 8. Updated 22 May 2022. Available from: http://hemiptera-databases.org/flow/ (accessed 9 June 2022)
Bourgoin, T. & Huang, J. (1990) Morphologie comparée des genitalia mâles des Trypetimorphini et remarques phylogénétiques (Hemiptera: Fulgoromorpha: Tropiduchidae). *Annales de la Société entomologique de France*, 26, 555–564.
Bourgoin, T., Wang, R.R., Asche, M., Hoch, H., Soulier-Perkins, A., Stroiński, A., Yap, S. & Szwedo, J. (2015) From micropterism to hyperpterism: recognition strategy and standardized homology-driven terminology of the forewing venation patterns in planthoppers (Hemiptera: Fulgoromorpha). *Zoomorphology*, 134 (1), 63–77.
https://doi.org/10.1007/s00435-014-0243-6
Caldwell, J.S. (1946) Notes on *Haplaxius* Fowler with descriptions of new species (Homoptera, Cixiidae). *Proceedings of the Entomological Society of Washington,* 48, 203–206.
Echavarria, M.A.Z., Barrantes, E.A.B., Bartlett, C.R., Helmick, E.E. & Bahder, B.W. (2021a) A new species of planthopper in the genus *Myxia* (Hemiptera: Auchenorrhyncha: Cixiidae) from the Reserva Privada el Silencio de Los Angeles Cloud Forest in Costa Rica. *Zootaxa,* 4915 (3), 351–363.
https://doi.org/10.11646/zootaxa.4915.3.4
Echavarria, M.A.Z., Barrantes, E.A.B., Bartlett, C.R., Helmick, E.E. &Bahder, B.W. (2021b) A new species of *Myxia* (Hemiptera: Auchenorrhyncha: Cixiidae) collected on palms from the Reserva Privada el Silencio de Los Angeles Cloud Forest in Costa Rica. *Zootaxa*, 5027 (3), 417–428.
https://doi.org/10.11646/zootaxa.5027.3.7
Emeljanov, A.F. (2007) New and little known taxa of the family Cixiidae (Homoptera, Fulgoroidea). *Entomologicheskoe Obozrenie*, 86 (1), 107–131. [in Russian, English translation *Entomological Review*, 87 (3), 287–308 (2007)]
https://doi.org/10.1134/S0013873807030062
Fennah, R.G. (1971) Fulgoroidea from the Cayman Islands and adjacent areas. *Journal of Natural History*, 5, 299–342.
https://doi.org/10.1080/00222937100770241
Fowler, W.W. (1904) Order Rhynchota. Suborder Hemiptera-Homoptera. (Continued). *Biologia Centrali-Americana*, 1, 85–124.
Howard, F.W. & Thomas, D.L. (1980) Transmission of palm lethal decline to *Veitchia merrillii* by a planthopper *Myndus crudus*. *Journal of Economic Entomology*, 73 (5), 715–717.
https://doi.org/10.1093/jee/73.5.715
Humphries, A.R., Ascunce, M.S., Goss, E.M., Helmick, E.E., Bartlett, C.R, Myrie, W., Barrantes, E.A.B., Zumbado, M.A.Z., Bustillo, A.E. & Bahder, B.W. (2021) Genetic variability of *Haplaxius crudus* based on the 5' region of the cytochrome *c* oxidase subunit I gene shed light on the epidemiology of palm lethal decline phytoplasmas. *Phytofrontiers*, 1 (3), 127–134.
https://doi.org/10.1094/PHYTOFR-12-20-0048-R
Kramer, J.P. (1979) Taxonomic study of the planthopper genus *Myndus* in the Americas (Homoptera: Fulgoroidea: Cixiidae). *Transactions of the American Entomological Society*, 105 (3), 301–389.
Kumar, S., Stecher, G. & Tamura, K. (2016) MEGA7: Molecular Evolutionary Genetics Analysis version 7.0 for bigger datasets. *Molecular Biology and Evolution*, 33, 1870–1874.
https://doi.org/10.1093/molbev/msw054
Mou, D.-F., Di Lella, B.A., Halbert, S.E., Bextine, B., Helmick, E.E. & Bahder, B.W. (2022). Acquisition and transmission of the lethal bronzing phytoplasma by *Haplaxius crudus* using infected palm spear leaves and artificial feeding media. *Phytopathology (in press)*.
https://doi.org/10.1094/PHYTO-03-22-0079-R
Muir, F.A.G. (1922) New Malayan Cixiidae (Homoptera). *Philippine Journal of Science*, 20, 111–119.
Myrie, W., Helmick, E.E., Bartlett, C.R., Bertaccini, A. & Bahder, B.W. (2019) A new species of planthopper belonging to the genus *Oecleus* Stål, 1862 (Hemiptera Fulgoroidea: Cixiidae) from coconut palm (*Cocos nucifera* L) in Jamaica. *Zootaxa*, 4712 (1), 127–137.
https://doi.org/10.11646/zootaxa.4712.1.9
O'Brien, L.B. (2006) Three new species of Neotropical Cixiidae (Hemiptera: Fulgoromorpha). *Russian Entomological Journal*, 15 (3), 311–314.
Spinola, M. (1839) Essai sur les Fulgorelles, sous-tribu de la tribu des Cicadaires, ordre des Rhyngotes. *Annales de la Société Entomologique de France*, 8, 133–337.
Stål, C. (1862) Novae vel minus cognitae Homopterorum formae et species. *Berliner Entomologische Zeitschrift*, 6, 303–315.
Van Duzee, E.P. (1907) Notes on Jamaican Hemiptera: A report on a collection of Hemiptera made on the Island of Jamaica in

the spring of 1906. *Bulletin of the Buffalo Society of Natural Sciences*, 8 (5), 3–79.
Van Duzee, E.P. (1909) Observation of some Hemiptera taken in Florida in the spring of 1908. *Bulletin of the Buffalo Society of Natural Sciences*, 9, 149–230.
Van Duzee, E.P. (1912) Hemipterological gleanings. *Bulletin of the Buffalo Society of Natural Sciences*, 10, 477–512.
Van Duzee, E.P. (1923) Expedition of the California Academy of Sciences to the Gulf of California in 1921—The Hemiptera (True bugs, etc.). *Proceedings of the California Academy of Sciences*, Series 4, 12, 123–200.