FIRST REPORTS OF THE INVASIVE GRASS-FEEDING LEAFHOPPER BALCLUTHA RUBROSTRIATA (MELICHR) (HEMIPTERA: CICADELLIDAE) IN THE UNITED STATES

James N. Zahniser, Steven J. Taylor, and Jean K. Krejca

ABSTRACT: The invasive leafhopper Balclutha rubrostrata (Melichar) was found in abundance in Bexar County, Texas. An arthropod diversity study along highway right-of-way in September and October, 2008 identified B. rubrostrata as the single most abundant species. This is the first peer-reviewed publication of this invasive species in the mainland United States. Other records of this species in the U.S. are discussed. Diagnostic characters of the adults are provided and illustrated, and the nymph is described and illustrated for the first time. Genomic DNA was extracted from one adult and one nymph specimen, and the mitochondrial COI gene was sequenced for each and yielded identical sequences, thus confirming the association of the nymph and adult. Sequences are deposited in GenBank for future use in diagnostic or other studies.

KEYWORDS: leafhopper, invasive species, Texas, Macrostelini, Deltocephalinae, COI, PCR primer

Leafhoppers in the genus Balclutha Kirkaldy (Cicadellidae: Deltocephalinae: Macrostelini) typically feed on grasses and are common and sometimes highly abundant in grasslands. The genus occurs worldwide and contains 111 valid species and one species, B. incissa Matsumura (= B. hortensis), is a serious pest of groundnut in India. The subject of this study, B. rubrostrata (Melichar), has recently been reported as a carrier of the sugarcane white leaf phytoplasma in Thailand (Hanboonsong et al., 2006). This species has a very wide distribution, being recorded from many parts in the Old World including Australia, several southeast Asian islands, Japan, India, and several African countries and is known in the New World from Puerto Rico, the U.S. Virgin Islands, Cuba, and Hawaii.

Balclutha species are typically small (3-4 mm), slender, and greenish-yellow, pale leafhoppers, sometimes with orange or reddish longitudinal stripes or markings. Species can be identified by using one of several taxonomic treatments of the group, including Blocker (1967), Knight (1987), and Webb and Villaste (1994), all of which include descriptions of the adult of B. rubrostrata. The main species-level diagnostic characters for species of Balclutha are found on the male pygofer and aedeagus.

The objectives of this report are to document the first records of this species in the continental U.S., to provide diagnostic features of the adult and of the nymph for the first time, and to make DNA sequence data available for future use in diagnostic or other studies.

METHODS

Collections were made as part of a study examining the arthropod diversity of an area of highway right-of-way that is within critical habitat designated for the endangered cave beetle, Rhadine infernalis (Barr) (USFWS 2003). The site is near Caracol Creek Coon Cave and designated as Critical Habitat Unit 16. The study area where these collections were obtained is approximately 500 m long and 65 m wide and immediately adjacent to Loop 1604, a highway around the city of San Antonio, Bexar County, Texas. Sampling took place between September 29, 2008, and October 19, 2008, and included sweep netting just before sunset, blacklighting 2-4 hours after sunset, and a Malaise trap that was checked and emptied approximately seven times in a fourteen-day period. The plant community consists of grasses, shrubs, and the scattered trees that represent a diversity of native and a few non-native species. The herbaceous plant community is dominated by the invasive grass Bothriochloa ischaemum (L.) Keng (King Ranch bluestem), with woody species including Colubrina texensis (Torr. & Gray) Gray (hognum), and Quercus fusiformis Small (Texas live oak).

Malaise trap samples were also available from another locality in Texas (Hays Co.: Driftwood) and were collected approximately once every two weeks between January 31, 2006, and August 13, 2006. Leafhoppers were extracted from these samples and identified.

Morphological terminology follows Oman (1949). Leafhopper abdomens were cleared in KOH solution, rinsed in water, and suspended in glycerin. Digital photographs were taken with a Q Imaging Micropublisher 3.3 digital camera mounted on an Olympus SZX12 stereo microscope and with a Nikon D1x digital SLR camera configured with lenses by Microptic. Digital Lab XLT system. Photographs were modified with Adobe Photoshop CS.

Two DNA extractions were performed - one of an adult male and one of a nymph suspected to be the nymph of B. rubrostrata; both specimens were from the Bexar Co. locality. DNA extraction was performed with a DNeasy Tissue Kit (Qiagen, Inc.). Two PCR reactions were performed for each extraction using the primers in Table 1. PCR products were amplified in a 25 μl total reaction volume using Taq Polymerase (Promega Corp.), held first for 5 min. at 94°C, then 40 cycles of 94°C for 45 sec., 55°C for 1.5 min., and 72°C for 2 min., then a final elongation step at 72°C for 5 min., and held at 10°C before being removed from the cycler. Double-stranded PCR products were checked for quality by running 5 μl of the product on a 1% agarose gel, stained with GelGreen, and imaged under visible light with a Dark Reader Transilluminator. Successfully amplified products were purified using a GeneClean III Kit. Both strands were sequenced using ABI Prism BigDye Terminator Kit version 3 (PE Applied Biosystems). Sequencing products were run on an ABI 3730XL capillary sequencer. Contigs were assembled in Sequencher 4.7.
Table 1. Primers used to amplify fragments of COI.

<table>
<thead>
<tr>
<th>Primer Name</th>
<th>Sequence</th>
<th>Reference</th>
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<tbody>
<tr>
<td>1517 (forward)</td>
<td>5'-CWA CAA AYC ATA AGG ACA TYG GAA C-3'</td>
<td>new*</td>
</tr>
<tr>
<td>2191 (reverse)</td>
<td>5'-CCG GTG AAA ATT AAA ATA TAA ACT TC-3'</td>
<td>Simon et al. (1994)¹</td>
</tr>
<tr>
<td>2195 (forward)</td>
<td>5'-TTG ATT TTT TGG TCA TCC AGA AGT-3'</td>
<td>Simon et al. (1994)</td>
</tr>
<tr>
<td>3014 (reverse)</td>
<td>5'-TCC AAT GCA CTA ATC TGC CAT ATT A-3'</td>
<td>Simon et al. (1994)</td>
</tr>
</tbody>
</table>

* Created based on conserved regions of Drosophila yakuba, Locusta migratoria, Philaenus spumarius, Triatoma sp., Homodolica vitripennis, and Blatella germanica COI sequences obtained from GenBank.

¹ Highlighted sequence changed from 'C' in Simon et al. (1994) to 'T' based on observed leafhopper sequences.

RESULTS AND DISCUSSION

The sampling in Bexar Co., TX yielded 18,162 individual invertebrates, 85% of which (15,000+) individuals were identified as Balclutha rubrostriatia (Melichar), thus making this species by far the dominant invertebrate collected by these methods at the site. Once identified, internet searches revealed that this species had been detected elsewhere recently in Texas. On the website BugGuide.net (http://bugguide.net/node/view/87190), this species (identified as B. rubrofasciata) was identified and photographed from Texas: Kerr County: Kerrville on 19.X.2006 (photographed by Ed Johnson) and from Texas: Travis Co.: Austin on 12.X.2006 (photographed by Mike Quinn, TexasEnto.net). The malaise trap samples from Driftwood, TX yielded many specimens of B. rubrostriatia. In these samples, the species was present from 31.I.2006 to 11.II.2006, absent from 12.II.2006 to 28.V.2006, present from 29.V.2006 to 4.VII.2006, abundant from 5.VII.2006 to 4.VIII.2006, and present from 5.VIII.2006 to 13.VIII.2006.

Five specimens of B. rubrostriatia were also found in the Illinois Natural History Survey (INHS) Insect Collection - one specimen previously recorded from US Virgin Islands: St. Thomas (27.XI.1947), 3 specimens newly reported here from Florida: Key Largo (28.IV.1990), and one specimen from Texas: Kleberg Co.: Kingsville (southwest of Corpus Christi; 8.VI.1971). Elsewhere in the US, this species previously has been recorded from Hawaii: Honolulu, where 6 specimens were taken in a blacklight at December, 1974 (anonymous, 1974; identified by J.W. Beardsley).

Because this species has been captured multiple times in recent years and because it was so abundant in some of the collections, it appears that this species is well-established in Texas. At this time, it is uncertain whether the records from Key Largo, FL, and Kingsville, TX, are stray specimens that may have been blown in or transported from the Caribbean Islands or whether they may be from unrecorded established populations. The Florida State Collection of Arthropods (FSCA) and the Texas A&M University Insect Collection (TAMU) were searched for specimens of B. rubrostriatia. One specimen tentatively assigned to this species was found at FSCA from St. Lucie Co., FL (Jan. 27, 1987), and none were found at TAMU. The absence of records from Texas from 1971 to 2006 and high populations recorded in 2006 and after seem to suggest that the earlier record from Kingsville may have been incidental. It is not known how it arrived in Texas, but perhaps it was transported by wind from its nearest known established locality, Cuba, or another Caribbean island. In another species of Balclutha, B. pauxila Lindberg, long distance dispersal has been documented. Swarms of this species were reported to have descended on Ascension Island from its presumed African mainland source (Ghauri, 1983). Alternatively, if B. rubrostriatia was already established in Florida, it may have dispersed by movement of populations over land to Texas. The distribution of B. rubrostriatia indicates that this is a tropical to warm temperate species, and should it spread further in the US, it will likely be confined to the southern states.

Although there is no evidence of B. rubrostriatia being a pest in the continental United States, its dominance at the Bexar Co. site (85% of the individuals between September and October) suggests that it has the capacity to alter the structure of grassland invertebrate communities in central Texas and perhaps elsewhere (Crowl et al., 2008), which may have as yet undetermined consequences (Strayer et al., 2006; Vila et al., 2009). The dominance of this species at this site also is consistent with the hypothesis that disturbance associated with the roadside habitat might be a factor in explaining the abundance of a new invader (Hobbs and Huenneke, 1992).

Primer pair 1517/2191 yielded fragments ~600 base pairs (bp) in length, and primer pair 2195/3014 yielded fragments ~825 bp in length. An overlapping region of ~10 bp between the two fragments allowed them to be aligned to each other and to other known leafhopper COI sequences. Sequences from the adult and nymph were identical, confirming the identity of the nymph. Full COI sequence are available from the NCBI GenBank database, accession #’s FJ824034 (adult) and FJ824035 (nymph).

Below, diagnostic characters of the adult are given and illustrated (Figs. 1, 2, 5-7) and the nymph is described and illustrated for the first time (Figs. 3, 4). The small-case letters after the publication dates in the synonymy given below correspond to those listed in Metcalf (1964).

SYSTEMATIC Entomology

Balclutha rubrostriatia (Melichar) Figs. 1-7

Original description: Gnathodus rubrotinctus Melichar 1903b: 208
= Gnathodus rubrotinctus Melichar 1905a: 304
= Nesostes sanguinescens Kirkaldy 1906c: 344
= Nesostes sordidor Kirkaldy 1906c: 344
= Balclutha rubrovittata Matsumura 1914a: 168
= Typhlocyba rubrostratiata Distant 1918b: 102
Description of Nymph. Overall pigmentation ranges from relatively lightly to relatively darkly pigmented (specimen in Figs. 3-4 is darkly pigmented). Body color generally stramineous. Crown somewhat square-shaped; with broad brown or fuscous longitudinal stripes next to eyes, very faint in lightly pigmented specimens; median portion of crown ivory. Face with anterodorsal part of frontoclypeus with brown pigmentation, without or with light pigmentation on posteroventral part of frontoclypeus and upper part of genae, clypellus, lorae, and lower part of genae with brown pigmentation. Bases of antennae with brown pigmentation. Pronotum with lateral stripes continuing from crown. Wing pads fuscous. Legs darkly pigmented at least on tarsi, fenur and tibia often with dark pigmentation; hind tibia nearly always darkly pigmented. Abdominal tergites with broad ivory coloring medially, with broad lateral brown stripes with red coloration on posterior margin. Abdominal sternites more or less pigmented medially, more posteriorly situated segments with darker pigmentation. Abdominal tergites with 6 longitudinal rows of setae and some more slender setae at lateral margin. Genital segment with several setae.


Voucher specimens from Bexar and Hays Cos. are deposited in the INHS Insect Collection.

ACKNOWLEDGEMENTS

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LITERATURE CITED


