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A New Species of *Cis* Latreille (Coleoptera: Ciidae) from the USA, with Comments on the Use by Ciidae of Stereaceae Fungi (Basidiomycota: Agaricomycetes: Russulales) as Hosts

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Abstract

Cis okennoni Lopes-Andrade and Ferro, **new species** (Coleoptera: Ciidae), is described from specimens found in Texas (type locality Fort Worth Nature Center and Refuge, Tarrant Co.), Alabama, Mississippi, and Oklahoma, USA. The new species is included in the *cayensis* species-group, and it is unique within this group in possessing single elytral punctation, an acute outer apical angle of the protibia, and males with the anterior edge of the pronotum projected into two conspicuous horns. The only known breeding record for *C. okennoni* is in basidiomes of *Xylobolus frustulatus* (Pers.) Boidin (Russulales: Stereaceae), from which the holotype and most of the type series were collected.

Key Words: *cayensis* species-group, ceramic or parchment fungus, minute tree-fungus beetle, taxonomy, *Xylobolus frustulatus*

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Introduction

Ciidae were last revised in North America north of Mexico by Lawrence (1971) and consisted of 85 species within 13 genera (Lawrence 1982; Souza-Gonçalves and Lopes-Andrade 2018b; Thayer and Lawrence 2002). No new native species have been described since then. However, Lawrence (1991) described three new extralimital species, *Cis chinensis* Lawrence, *Cis asiaticus* Lawrence, and *Orthocis auriculariae* Lawrence, from China and Thailand that had been imported into the USA in commercial dried fungi (Madenjian *et al.* 1993). Among these, only *C. chinensis* was reported to have become established in the USA (Souza-Gonçalves and Lopes-Andrade 2018b). In North America, the genus *Cis* Latreille is represented by 44 species (Lawrence 1971, 1974, 1982; Souza-Gonçalves and Lopes-Andrade 2018b; Thayer and Lawrence 2002), not including *C. asiaticus*.

In December 2016, specimens of an unidentified ciid were collected by Bob O'Kennon from ceramic or parchment fungus (*Xylobolus frustulatus* (Pers.) Boidin, Stereaceae) growing on post oak (*Quercus stellata* Wangenh., Fagaceae) near Fort Worth, Texas. A detailed morphological description of the fungus and information on development and ecology as well as observations about the association of the beetles with the fungus can be found in O'Kennon *et al.* (2018). The unidentified ciid was determined as an undescribed species, and

additional conspecifics from Alabama, Mississippi, and Oklahoma were made available to us by John F. Lawrence.

Herein, *Cis okennoni* Lopes-Andrade and Ferro, new species, is described. It fits well in the *cayensis* species-group, which includes *Cis cayensis* Lawrence, 1971 and *Cis niedhauki* Lawrence, 1971. The morphological affinities of the three species are briefly discussed, as well as the use of *Xylobolus* P. Karst. and other Russulales as host fungi by ciid beetles.

Material and Methods

Scanning electron microscopy was performed using a Hitachi SU6600. Specimens were not coated and observed with pressure 30 Pa, 10.0 kV. Images were taken using a Canon EOS 7D camera with Kenko extension tubes (12 mm, 20 mm, 36 mm) and a Canon MP-E 65-mm lens. The programs Helicon Remote version 3.9.7 W and Helicon Focus version 7.0.2 Pro (Helicon Software Ltd.) were used to obtain image stacks and render focused images. Resultant images were optimized using Adobe Photoshop CC 19.0.

Transcription of label data, dissection, and photography of sclerites and measurement of specimens followed the methods described by Araujo and Lopes-Andrade (2016). The fungus name was updated by consulting the database Index Fungorum (www.indexfungorum.org). Exemplar specimens from all localities were measured. Differences are given in "Variation", together with standard measurements (mean and standard deviation) and ratios. The following individuals were dissected: one male and one female from Tarrant Co., Texas; and one male from Marshall Co., Oklahoma. The holotype was not dissected.

Terms for external morphology and male terminalia of ciids follow Lawrence (2016), Lawrence *et al.* (2011), and Lopes-Andrade and Lawrence (2005, 2011), but see also Oliveira *et al.* (2013) for an explanation on the use of "tegmen". The following abbreviations are used for measurements (in mm) and ratios: BW (width of the anterior edge of the scutellar shield); CL (length of the antennal club measured from the base of the eighth antennomere to the apex of the tenth antennomere); EL (elytral length along the midline); EW (greatest width of the elytra); FL (length of the antennal funicle measured from the base of the third antennomere to the apex of the seventh antennomere); GD (greatest depth of the body measured in lateral view); GW (greatest diameter of the eye); PL (pronotal length along the midline); PW (greatest pronotal width); SL (length of the scutellar shield), TL (total length counted as EL+PL, *i.e.*, excluding head). The GD/EW and TL/EW ratios indicate the degree of body convexity and elongation, respectively.

Abbreviations for scientific collections are as follows:

ANIC	Australian National Insect Collection, CSIRO Entomology (Canberra, Australian Capital Territory, Australia, Adam Ślipiński)
BRIT	Botanical Research Institute of Texas (Fort Worth, Texas, USA, Tiana F. Rehman)
CELC	Coleção Entomológica do Laboratório de Sistemática e Biologia de Coleoptera da Universidade Federal de Viçosa (Viçosa, Minas Gerais, Brazil, Cristiano Lopes-Andrade)
CUAC	Clemson University Arthropod Collection (Clemson University, Clemson, South Carolina, USA, Michael L. Ferro)
FMNH	Field Museum of Natural History (Chicago, Illinois, USA, Margaret Thayer)
MCZ	Museum of Comparative Zoology (Harvard University, Cambridge, Massachusetts, USA, Brian D. Farrell)
NMNH	National Museum of Natural History (Smithsonian Institution, Washington, District of Columbia, USA, Terry Erwin)

Results

Cis okennoni Lopes-Andrade and Ferro, new species

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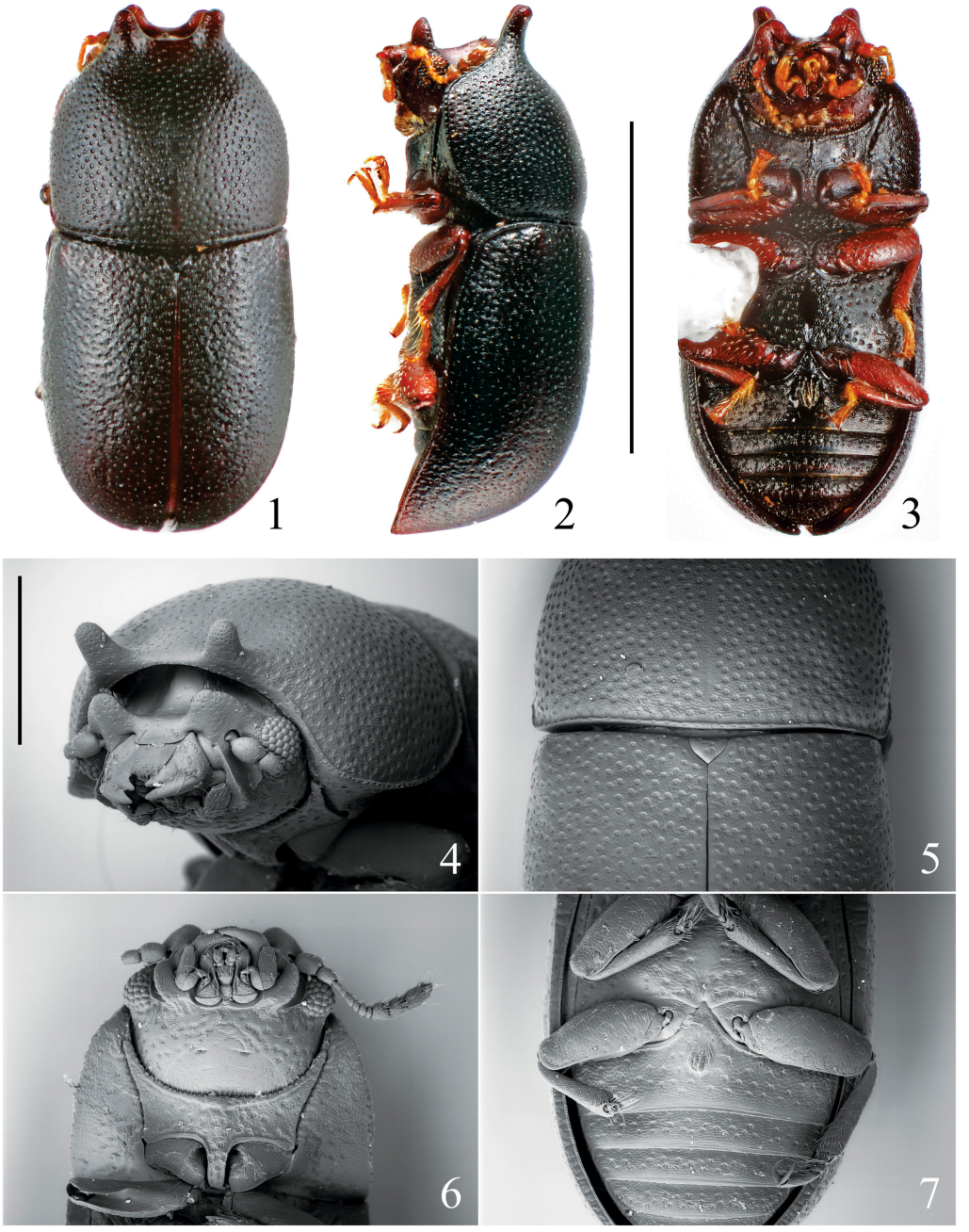
(Figs. 1–16)

Type Locality. Fort Worth Nature Center and Refuge (USA: Texas: Tarrant Co.), 32° 49' 42.60" N, 97° 49' 46.44" W.

Etymology. The species is named *okennoni* in honor of Bob O'Kennon, who collected most of the type series.

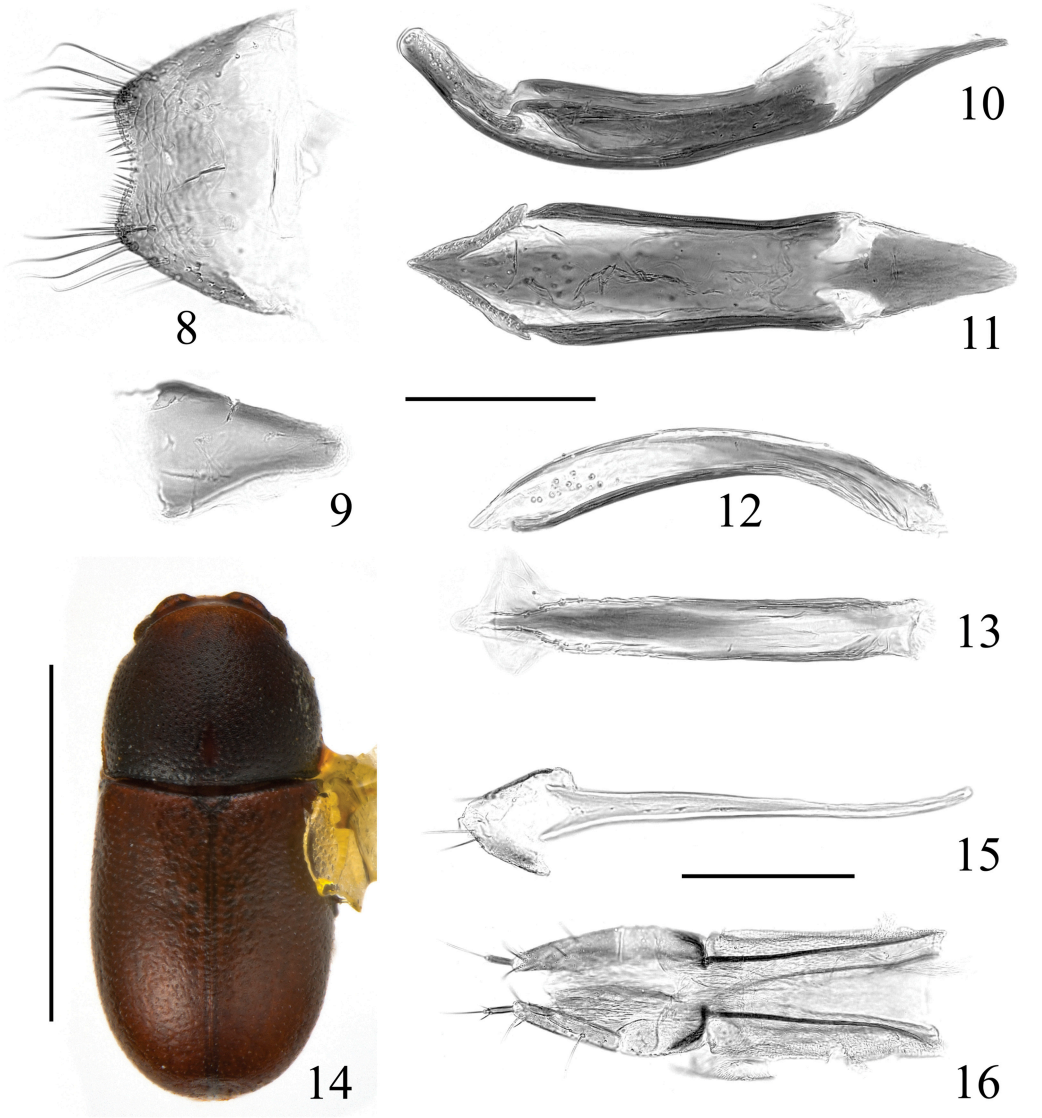
Diagnosis. Among species in the *C. cayensis* species-group, *C. okennoni* is unique in possessing single elytral punctation (Fig. 5), an acute outer apical angle of the protibia, and males with the anterior edge of the pronotum projected into two conspicuous horns (Figs. 1, 2, 4). A comparison of species in the *C. cayensis* species-group is provided in Table 1.

Description. Male Holotype. Adult fully pigmented and in good condition (Figs. 1–3). Measurements: TL = 1.56 mm, PL = 0.61 mm, PW = 0.69 mm, EL = 0.95 mm, EW = 0.75 mm, GD = 0.57 mm. Ratios: PL/PW = 0.88, EL/EW = 1.27, EL/PL = 1.56, GD/EW = 0.76, TL/EW = 2.08. **Body:** Oblong, convex; dorsum and venter dark



Figs. 1–7. *Cis okennoni*, **new species**. **1–3)** Holotype, dorsal, lateral and ventral views, respectively; Paratype from the type locality: **4)** Head and pronotum, oblique view; **5)** Part of pronotum and elytra; **6)** Head and prothorax, ventral view; **7)** Abdomen and part of thorax, ventral view. Scale bars = 1 mm (Figs. 1–3); 0.3 mm (Figs. 4–7).

brown; antennae, palpi, and tarsi dark reddish brown; dorsal vestiture of very minute setae about as long as 1 puncture width (Fig. 5); ventral vestiture of short, decumbent setae, usually longer than dorsal vestiture (Figs. 6, 7). **Head:** Only anterior portion visible from above (Fig. 1); anterior edge produced forward and slightly upward in 2 short, subtriangular plates, with rounded apices and



Figs. 8–16. *Cis okennoni*, **new species**. Male paratype from the type locality: **8)** Sternite VIII; **9)** Basal piece; **10–11)** Tegmen, lateral view and dorsal views, respectively; **12–13)** Penis, lateral view and dorsal views, respectively. Female paratype from Oklahoma: **14)** Dorsal habitus. Female paratype from the type locality: **15)** Spiculum ventrale, dorsal view; **16)** Ovipositor and associated parts, ventral view. Scale bars = 0.1 mm (Figs. 8–13); 1 mm (Fig. 14); 0.2 mm (Figs. 15–16).

separated by about 1 eye width (Figs. 3, 4); dorsum shiny, shallowly microreticulate, sparsely punctate; vertex elevated and convex at longitudinal midline (forming conspicuous protuberance), with a margined concavity at each side. **Antennae:** With 10 antennomeres (Fig. 6), lengths as follows (in mm, right antenna measured): 0.06, 0.04, 0.03, 0.02, 0.02, 0.02, 0.02, 0.04, 0.03, 0.06 (FL = 0.11 mm, CL = 0.16 mm, CL/FL = 1.45). **Eyes:** Suboval, coarsely faceted, with about 60–70 ommatidia; GW = 0.11. **Gula:** 0.52× as wide as head. **Pronotum:** Coarsely, confusedly punctate, narrow, longitudinal impunctate midline close to posterior edge (Fig. 5); punctures separated by about 1–2 puncture widths; interspaces markedly microreticulate; anterior edge produced forward and upward to form 2 slightly divergent horns (Figs. 1, 4) separated from each other at base by 1 eye width

Table 1. Comparative morphological differences among three *Cis* species in the *cayensis* species-group. Host-use groups *sensu* Orledge and Reynolds (2005).

Features	*C. cayensis*	*C. niedhauki*	*C. okennoni*
Anterior portion of vertex in male	2 lateral tubercles	2 lateral horns	2 lateral concavities
Posteromedian vertex in male	concavity	sharp conical tubercle	protuberance
Outer apical angle of protibia	rounded	rounded	acute
Anterior pronotal margin in male	simple	2 small tubercles	2 conspicuous horns
Elytral punctation	dual	dual	single
Elytral vestiture	dual	single	single
Abdominal sex patch in male	small	small	large
Host-use group	*Phellinus*	*Phellinus*	*Stereum*

and preceded by broad, shallow concavity; lateral edges barely crenulate, not explanate, not visible from above. **Scutellar shield:** Subtriangular, sparsely, shallowly punctate, interspaces shallowly microreticulate (Fig. 5); BW = 0.08; SL = 0.05. **Elytra:** Singly, confusedly punctate (Fig. 5), punctation slightly sparser than on pronotum; interspaces shallowly microreticulate. **Metathoracic wings:** Developed, apparently functional. **Hypomera:** Coarsely, shallowly, sparsely punctate (Fig. 6); interspaces microreticulate. **Prosternum:** Short (Fig. 6); tumid at longitudinal midline, subcarinate; coarsely, sparsely punctate; interspaces microreticulate. **Prosternal process:** Subparallel-sided, slightly curved inward (Figs. 3, 6); about as long as prosternum at midline beyond procoxae; apex rounded. **Protibiae:** Not expanded, devoid of spines; outer apical angle projected as acute tooth. **Meso- and metatibiae:** Not expanded; outer apical angle rounded; apical edge with row of spines. **Metaventrite:** 0.47× as long as abdominal ventrites together (Figs. 3, 7); coarsely punctate; interspaces microreticulate except for smooth, impunctate area at middle; discrimen about 1/4 length of metaventrite. **Abdominal ventrites:** Coarsely, shallowly punctate (Figs. 3, 7); interspaces microreticulate; length of ventrites (in mm, from base to apex at the longitudinal midline) as follows: 0.23, 0.08, 0.07, 0.07, 0.07; 1st abdominal ventrite with large, oval, margined, setose sex patch anteriorly that is 0.48× as long as ventrite, length 0.11 mm, transverse diameter 0.09 mm. **Male abdominal terminalia of paratypes:** Sternite VIII (Fig. 8) subtrapezoidal; posterior edge curved inwardly, with short setae; posterior sides rounded, bearing long slender setae. Tegmen (Figs. 10, 11) curved in lateral view (Fig. 10), concave dorsally, 4.35× as long as wide; anterior portion elongate, triangular; posterior portion triangular. Size and shape of basal piece (Fig. 9) similar to those of anterior portion of tegmen, 1.42× as long as wide. Penis (Figs. 12, 13) curved in lateral view, 0.77× as long as tegmen, 8× as long as wide; apex with subtriangular, membranous expansions.

Female. Head with convex vertex, devoid of any impression; anterocephalic edge (Fig. 14) slightly projecting, with shallow, round emargination at middle forming 2 short projections that are broadly rounded apically; gula 0.36× as wide as head. Pronotum with anterior edge broadly rounded; lateral edges more crenulate than in males. First abdominal ventrite devoid of sex patch. **Female abdominal terminalia:** Spiculum ventrale (Fig. 15) 0.94× length of paraprocts, gonocoxites, and gonostyli combined. Gonocoxites (Fig. 16) with 3 ventral lobes on each side; each apical lobe 1.32× as long as basal lobes together, with a slender gonostylus inserted apically. Paraprocts (Fig. 16) 1.26× length of gonocoxites (excluding gonostyli), with a pair of baculi. Proctiger about as long as paraprocts, with a pair of baculi.

Variation. Male measurements (n = 9, including holotype): TL = 1.28–1.90 mm (1.53±0.20), PL = 0.48–0.73 mm (0.61±0.09), PW = 0.55–0.85 mm (0.69±0.09), EL = 0.78–1.18 mm (0.92±0.13), EW = 0.63–0.88 mm (0.74±0.08), GD = 0.45–0.63 mm (0.56±0.06), PL/PW = 0.79–0.96 (0.88±0.05), EL/EW = 1.10–1.34 (1.25±0.08), EL/PL = 1.19–1.79 (1.52±0.18), GD/EW = 0.71–0.81 (0.76±0.03), TL/EW = 2.00–2.17 (2.08±0.06). Female measurements (n = 6): TL = 1.23–1.58 mm (1.40±0.15), PL = 0.43–0.58 mm (0.51±0.05), PW = 0.55–0.70 mm (0.64±0.06), EL = 0.78–1.05 mm (0.89±0.11), EW = 0.58–0.78 mm (0.70±0.07), GD = 0.48–0.60 mm (0.55±0.05), PL/PW = 0.75–0.84 (0.80±0.04), EL/EW = 1.07–1.40 (1.27±0.12), EL/PL = 1.48–2.00 (1.74±0.18), GD/EW = 0.76–0.83 (0.79±0.02), TL/EW = 1.79–2.13 (2.00±0.12). The secondary sexual features of males (vertexal elevation, anterocephalic plates, pronotal horns, and abdominal sex patch) vary in length, as occurs in several other species of *Cis*.

Type Material. Holotype: ♂ (in MCZ) labeled "USA: Texas: Tarrant Co. Fort Worth Nature Center N 32.8285, W −97.4629 21 December 2016 ex *Xylobolus frustulatus* on post oak log. col. B. O'Kennon \ *Cis okennoni* Lopes-Andrade & Ferro HOLOTYPUS [printed in red label]". Paratypes (13♂♂ and 9♀♀): 8♂♂ (4 CELC, 1 dissected; 4 CUAC, first three damaged, with database

identification labels CUAC000077871, CUAC000077872, CUAC000077873, and CUAC000077874) and 5♀♀ (2 CELC, 1 dissected; 1 MCZ; 2 CUAC with database identification labels CUAC000077875 and CUAC000077876), same locality data as holotype; 4♂♂ (1 ANIC; 1 CELC, dissected; 2 FMNH) and 2♀♀ (CELC, FMNH) "OKLAHOMA: Marshall Co.; Willis [printed] (2 mi W) 12.VII.1969 [handwritten] \ Butress Debris W. Suter leg. [printed] stump [handwritten]"; 1♀ (ANIC) "CHATTAHOOCHIE [sic] ST. PK. HOUSTON CO. ALA. IV-2-69 [handwritten \ J. F. Lawrence Lot. [printed] 2708 [handwritten] \ S. Peck collector [printed] \ STEREUM SUBPILEATUM [handwritten]"; 1♂ and 1♀ (NMNH) "Hancock Co. Miss [printed] 28.8 [handwritten]", the male additionally labeled "Coll Hubbard & Schwarz". All paratypes additionally labeled "*Cis okennoni* Lopes-Andrade & Ferro PARATYPUS [printed in yellow label]".

Host Fungi. There are only two host records, both in *Xylobolus*: *X. frustulatus*, a breeding record (criteria *sensu* Orledge and Reynolds 2005); and a record of a single female in *X. subpileatus* (Berk. and M. A. Curtis) Boidin, based on label data (see Type Material). *Xylobolus frustulatus* only occurs on *Quercus* L. (oaks) worldwide, but in the Arlington and Fort Worth area of Texas, only on *Q. stellata*. In addition, this fungus is a specialist on decorticated hard wood on decaying logs on ground sites and sometimes is present on many logs in a given area. Basidiomes of *X. frustulatus* are identified easily with pictures because of the ivory-colored, flattened ceramic-tile frustules. The beetles occur in older frustules that are weather-beaten and have lost their color and general habit. Fungal-beetle geographical distribution and collection records are associated with collectors who target oak woods. Most recently beetles were observed in *X. frustulatus* in Missouri at Lake of the Ozarks State Park, Camden County, Camp Clover Point, Missouri Mycological Society Foray, Sept. 30, 2017 (HWK, personal observation). Unfortunately, voucher specimens were not preserved for future study. *Quercus stellata* populations and habitats should be observed for this fungus and beetle in the following states: Kansas, Arkansas, Illinois, Indiana, Kentucky, Tennessee, North Carolina, South Carolina, and Georgia. The type locality on Greer Island at the Fort Worth Nature Center and Refuge had six *Q. stellata* logs with the fungus-beetle association, and three of these logs were transported to the Botanical Research Institute of Texas to study the life cycle stages of the beetles. A specimen of *X. frustulatus* was deposited in the BRIT herbarium as R. J. O'Kennon 31027 (BRIT barcode 59723).

Distribution. South-central to southeastern USA (Texas, Alabama, Mississippi, and Oklahoma) (Fig. 17).

Discussion

Specimens in the type series collected before 2016 were made available by John F. Lawrence (Australia), and the morphospecies was coded "*Cis* 135" by him. He also made available a series of specimens from Antiguo Morelos (Tamaulipas, Mexico); these specimens have a comparatively more elongate body and finer pronotal punctation, which suggests a separate species morphologically similar to *C. okennoni*. Males of this Mexican species have straighter pronotal horns, and the genitalia have a comparatively smaller basal piece and a tegmen with a narrower posterior portion. The *cayensis* species-group originally included species

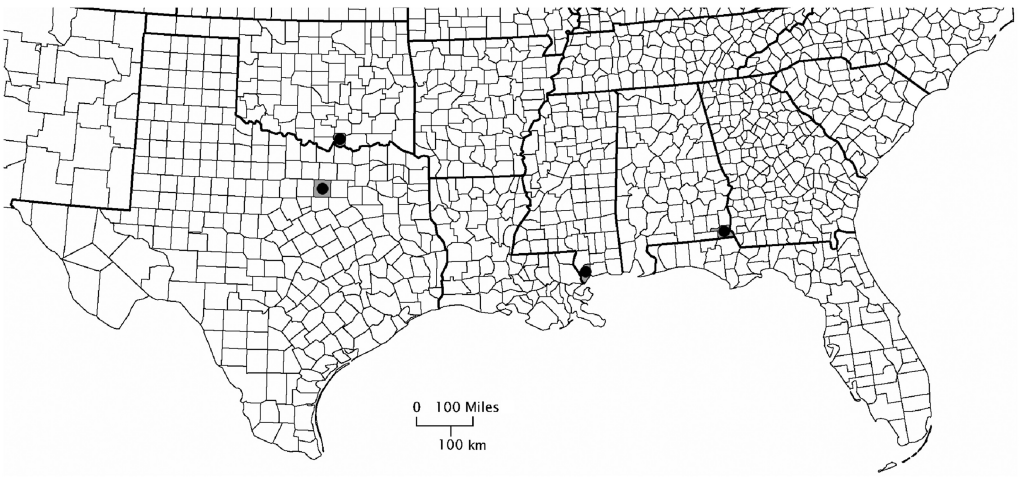


Fig. 17. Distribution map of *Cis okennoni*, **new species**.

with a moderately tumid and subcarinate prosternum, expanded and rounded outer apical angles of the protibiae, dual elytral punctation, and males bearing a conspicuous anteromedian impression on the vertex (Lawrence 1971). *Cis okennoni* differs in the acute outer apical angle of the protibiae, single elytral punctation, and the male vertex devoid of an anteromedian impression, but it has a similar prosternum and males bear a well-developed, conspicuous posteromedian elevation on the vertex similar to that of males *C. niedhauki* (absent in male *C. cayensis*). These latter two features and the noticeable general morphological similarity to *C. niedhauki* support the inclusion of *C. okennoni* in the *cayensis* species-group. The examined individuals of *C. okennoni* are usually larger (TL = 1.23–1.90 mm) than *C. cayensis* (TL = 1.02–1.37 mm) and *C. niedhauki* (TL = 1.00–1.35 mm), and the males usually have a larger abdominal sex patch. These species also differ in host use: *C. cayensis* and *C. niedhauki* are usually found in basidiomes of Hymenochaetaceae (Lawrence 1973; Orledge and Reynolds 2005), while *C. okennoni* is found in Stereaceae. The main differences between species in the *C. cayensis* species-group are provided in Table 1.

A common way to collect ciid beetles is to search for them in bracket fungi of the orders Polyporales and Hymenochaetales or by sifting leaf litter (CLA, personal observation). Fungi in the order Russulales, especially in Stereaceae, are usually not a target. Part of the type series of *C. okennoni* was observed feeding and breeding on the Stereaceae fungus *X. frustulatus* (O'Kennon *et al.* 2018, as *Cis* sp.). Besides *C. okennoni*, the only other ciid with breeding records in *Xylobolus* is *Ennearthron chujoi* Nakane and Nobuchi in *Xylobolus spectabilis* (Klotzsch) Boidin in Japan (Kawanabe 1995). Aside from *Xylobolus*, other breeding records of ciid beetles in Stereaceae are uncommon, and the following have been gathered from the scientific literature: *Cis stereophilus* Lawrence from southeastern USA, the unique North American ciid species found exclusively in species of *Stereum* Pers., mostly in *Stereum ostrea* (Blume and T. Nees) Fr. (Lawrence 1973); the polyphagous North American *Cis americanus* Mannerheim, with a few records in *Stereum hirsutum* (Willd.) Pers. (Lawrence 1973; Majka 2007); the polyphagous North American *C. levettei* (Casey) and *Cis subtilis* Mellié, with a few records in *S. ostrea* (Majka 2007); European species in the *festivus* species-group and *Orthocis alni* (Gyllenhal) in *Stereum rugosum* Pers. (Lawrence 1973; Orledge and Reynolds 2005); the polyphagous *Cis foveocephalus* Souza-Gonçalves and Lopes-Andrade, *Cis mpumalangaensis* Souza-Gonçalves and Lopes-Andrade, and *Cis neserorum* Souza-Gonçalves and Lopes-Andrade from South Africa (Souza-Gonçalves and Lopes-Andrade 2017, 2018a). Aside from Stereaceae, the other records of ciids in Russulales are of oligophagous and polyphagous North American species in *Heterobasidion annosum* (Fr.) Bref. (Bondarzewiaceae): as *Ceracis californicus* (Casey), *C. americanus*, *Cis creberrimus* Mellié, *C. levettei*, *Cis megastictus* Lawrence, *Dolichocis manitoba* Dury, and *Hadreule blaisdelli* (Casey) (Lawrence 1973; Majka 2007). Therefore, among these cases, only the North American *C. stereophilus* and the three European species in the *festivus* species-group breed exclusively or almost exclusively in *Stereum* species, while other ciids use Russulales fungi as an alternative host, not the main resource. It would be important to check whether *C. okennoni* is a specialist on *Xylobolus*.

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