

Stridulatory file and calling song of two populations of the tropical bush cricket *Eneoptera surinamensis* (Orthoptera, Gryllidae, Eneopterinae)

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ABSTRACT. This study characterizes the calling song and ultramorphology of the stridulatory file of two geographically isolated populations of the tropical bush cricket *Eneoptera surinamensis* (De Geer, 1773) from city of Foz do Iguçu, state of Paraná, and town of Rio Claro, state of São Paulo, Brazil, distant 1,000 Km from each other. The teeth are shell-shaped, the larger ones are distributed in the medium region of the file, decreasing gradually in size towards the edges. Specimens from Foz do Iguçu have a file with 82 ± 9.8 teeth, length = $1.89 \text{ mm} \pm 0.15$ with 43.76 ± 5.94 teeth per mm ($n=15$). Specimens from Rio Claro present a file with 87 ± 9.81 , length = 1.96 ± 0.19 mm with 44.52 ± 4.61 teeth per mm ($n=15$). Statistical differences found between the two populations are not significant. The calling song is an uninterrupted trill that alternates two sets of notes distinct for its temporal features.

KEYWORDS. Grylloidea, bioacoustic, sonogram, *pars stridens*.

RESUMO. Fileira estridulatória e som de chamado de duas populações do grilo tropical *Eneoptera surinamensis* (Orthoptera, Gryllidae, Eneopterinae). Este estudo caracteriza a ultramorfologia da nervura estridulatória e o som de chamado de duas populações de *Eneoptera surinamensis* (De Geer, 1773) de Foz do Iguçu, Paraná e Rio Claro, São Paulo, distantes cerca de mil quilômetros. Os dentes que compõem a nervura estridulatória têm forma de concha, os maiores presentes na região mediana da fileira, reduzindo gradualmente de tamanho em direção às extremidades. Os espécimes de Foz do Iguçu têm a nervura estridulatória com $82 \pm 9,8$ dentes, sendo $43,76 \pm 5,94$ ($n=15$) por milímetro, enquanto os de Rio Claro $1,96 \pm 0,19$ mm de comprimento, $87 \pm 9,81$ dentes e $44,52 \pm 4,61$ dentes por mm ($n=15$). As diferenças observadas entre as populações não são estatisticamente significantes. O som de chamado é composto por um trill contínuo que alterna dois conjuntos de notas, com padrões temporais distintos.

PALAVRAS-CHAVE. Grylloidea, bioacústica, sonograma, *pars stridens*.

The acoustic repertoire of crickets includes different kinds of songs, such as calling, courtship, copulation and aggressiveness (ALEXANDER, 1957a,b, 1960, 1961, 1968). According to ALEXANDER (1962), crickets present the most complex acoustic systems among the invertebrates and the calling song is the best understood due to its taxonomic features (ALEXANDER, 1957a; WALKER, 1964; OTTE & ALEXANDER, 1983; DAVID *et al.*, 2003).

Cricket acoustic signals are produced when forewings (tegmen) are raised and rubbed together. The right tegmen presents a file of teeth (*pars stridens*), which is scraped by a structure named *plectrum*, located on the left tegmen. The energy of the impact of the *plectrum* on the teeth promotes the vibration of the specialized areas of the tegmen, such as the harp and mirror, which engender sound pulses (WALKER, 1962; WALKER & CARLYSLE, 1975; LEROY, 1979; KOCH *et al.*, 1987; BENNETT-CLARK, 1989).

The specialized structures of the tegmen that produce and amplify the acoustic signals can vary between species (WALKER & CARLYSLE, 1975), and consequently may be employed as taxonomic characters.

According to WALKER & CARLYSLE (1975), the morphology of teeth is similar among individuals of the same species and highly diversified among different subfamilies. *Endecous itatibensis* Rehn, 1918 has

triangular teeth with a smooth surface (E. Zefa, unpublished data); *Allonemobius fasciatus* (De Geer, 1773) has laminar and smooth teeth; *Anurogryllus arboreus* Walker, 1973 has thick teeth, rounded off and with longitudinal notches on their surface; the teeth of *Gryllus ovisopis* Walker, 1974 are similar to those of *A. fasciatus*, but more curved in shape (WALKER & CARLYSLE, 1975). A remarkable case is *Eneoptera guyanensis* Chopard, 1931, whose stridulatory file presents two morphological groups of teeth, one of them containing 50-60 large and widely separated teeth, while the other set bears the same number of teeth, but these are smaller and set closer together (DESUTTER-GRANDCOLAS, 1998).

The teeth density (number of tooth per millimeter) is another feature that varies among different species. Contrasting cases may be observed in *Anaxipha latipennis* (Walker, 1869) and *Oecanthus exclamationis* Davis, 1907, with 173 and 19 teeth/mm, respectively (WALKER & CARLYSLE, 1975).

ROBILLARD & DESUTTER-GRANDCOLAS (1995) presented a taxonomic revision of *Eneoptera*, including information on calling songs and teeth number of stridulatory file of *Eneoptera surinamensis* (De Geer, 1773), *E. fasciatus* (Scudder, 1869), *E. guyanensis* Chopard, 1931, *E. gracilis* Robillard, 2005, and *E. nigripedis* Robillard, 2005.

Eneoptera surinamensis lives in open areas of rain forest domain (along roads, in natural or man-made clearings). Both sexes are found near the ground, in the leaf litter or sometimes on bushes, from where males sing (ROBILLARD & DESUTTER-GRANDCOLAS, 2005).

The aim of this work was to characterize the ultramorphology and the morphometry of the stridulatory file of the bush cricket *E. surinamensis* and to correlate its features with the calling song. We particularly address the hypothesis that *E. surinamensis* specimens from the two geographically distant populations would present some divergence in stridulatory file morphology and morphometry.

MATERIAL AND METHODS

The specimens here analyzed were found on shrubs and leaf litter of Semi-deciduous Forest, located at town of Foz do Iguaçu, state of Paraná, Brazil (25°27'S; 54°34'W), and at the Campus of the UNESP – Rio Claro, state of São Paulo, Brazil (22°23'S - 47°32'W). These sites are 1,000 km distant from each other.

All specimens were deposited in the collection of the Departamento de Biologia, Instituto de Biociências - UNESP, Rio Claro.

To test the hypothesis that *E. surinamensis* specimens from the two populations would present some divergence in stridulatory file, three parameters were measured: total number of teeth, stridulatory file length, and number of teeth per millimeter. The stridulatory file of fifteen specimens from each locality was analyzed with a photomicroscope using the software Leica Qwin. The results were submitted to statistical test *t* – Student. The ultra-morphology of the stridulatory file of six specimens from each locality was photographed with a scanning electron microscope.

Seven specimens from Foz do Iguaçu and one from Rio Claro were acoustically analyzed. Sound recordings were made in the field from August to November 2003 at Foz do Iguaçu and on October 10th, 2004 at Rio Claro.

Recordings were made with a Nagra E tape recorder at tape speed of 19 cm/s. Songs were analyzed with Avisoft-SASLab Light software, digitalized at sampling frequency 22050 Hz, resolution: 16 bit; FFT: 256, frame 12.50%, windows: hamming, overlap: 99.1%. We analyzed 20 seconds of the calling song of each specimen. Two song frequency variables were measured - the maximum and the minimum frequencies - and two temporal variables - time duration of the note and the elapsing time between notes. The sequence of the calling song of the *E. surinamensis* from Rio Claro here analyzed is available at the site "Orthoptera Species File" (EADES *et al.*, 2006).

To analyze the acoustic signals, it was considered that each tooth of the file emits a sonorous pulse and, in addition, that a set of pulses emitted during the closing movement of the tegmen produces a note [=phonotome (LEROY, 1979) or syllable].

RESULTS

The teeth of stridulatory file are arranged linearly and inclined from the outer to the inner side of the tegmen

(Fig. 1). Each tooth is shell-shaped (Figs. 2, 3); the larger ones are distributed in the medium region of the file, with a gradual reduction in size towards the extremities.

The smaller teeth located at the outer (Fig. 4) and inner (Fig. 5) ends of the file show an asymmetric morphology, irregular alignment and intermittent distribution.

Parallel to the anterior side of the file occurs a dense accumulation of microtrichias, forming a strip that is broadest where the biggest teeth are located (Figs. 1, 4, 5). There is a gradual reduction in the accumulation of microtrichias following the decrease in teeth size towards the file endings. There are no microtrichias where the smaller, asymmetric and intermittent teeth are located (Figs. 2, 3).

Close to the outer end of the file (postcubital area) there is a hair plate (Fig. 1).

Specimens from Foz do Iguaçu have a file with 82 ± 9.8 teeth, including the minor and asymmetric ones, with the length of the file equal to $1.89 \text{ mm} \pm 0.15$, which is equivalent to 43.76 ± 5.94 teeth per mm. The population from Rio Claro presents a file with 87 ± 9.81 teeth, with 1.96 ± 0.19 mm of length, and 44.52 ± 4.61 teeth per mm. Statistical test had disclosed that the differences found between the two populations are not significant ($p > 0.05$).

The specimens from both places had been observed stridulating during the first hours of the morning, at nightfall and during the night. The calling song is a trill that alternates two distinct sets of notes, one of them (section *a*) with 0.6 s and the other one (section *b*) with 1.4 s (Fig. 6).

The section *a* includes 46 ± 3.74 (42 - 52, $n=5$) notes, each note with a duration of 0.009 s; the elapsed time between notes is 0.004 s (Fig. 7). The section *b* includes 80.8 ± 2.79 (77 - 85, $n=6$) notes, with 0.012 s and the elapsed time between them of 0.007 s (Fig. 8).

In the section *a*, each note is composed of 24 pulses and in the section *b*, of 31 pulses (Figs. 9, 10, respectively).

The minimum frequency of the calling song of specimens from Foz do Iguaçu is $2,706.7 \pm 97.9$ Hz (2497 - 2756, $n=7$) and the maximum frequency is $3,284.1 \pm 153.5$ Hz (3,014 - 3,445, $n=7$). The minimum frequency of the specimen from Rio Claro is 2,325 Hz and the maximum frequency is 3,186 Hz.

DISCUSSION

The number of teeth varies considerably among and within the species of *Eneoptera*, being 74 to 90 in *E. surinamensis*, 51 to 59 in *E. gracilis*, 97 to 107 in *E. nigripedis* and 108 to 126 in *E. guyanensis*. The latter two species have the stridulatory file with two distinct morphologic groups of teeth (ROBILLARD & DESUTTER-GRANDCOLAS, 2005).

The specimens here analyzed have the stridulatory file with a similar number of teeth as the specimens of *E. surinamensis* studied by ROBILLARD & DESUTTER-GRANDCOLAS (2005).

The teeth at both endings of the stridulatory file of *E. surinamensis*, with asymmetric morphology, irregular

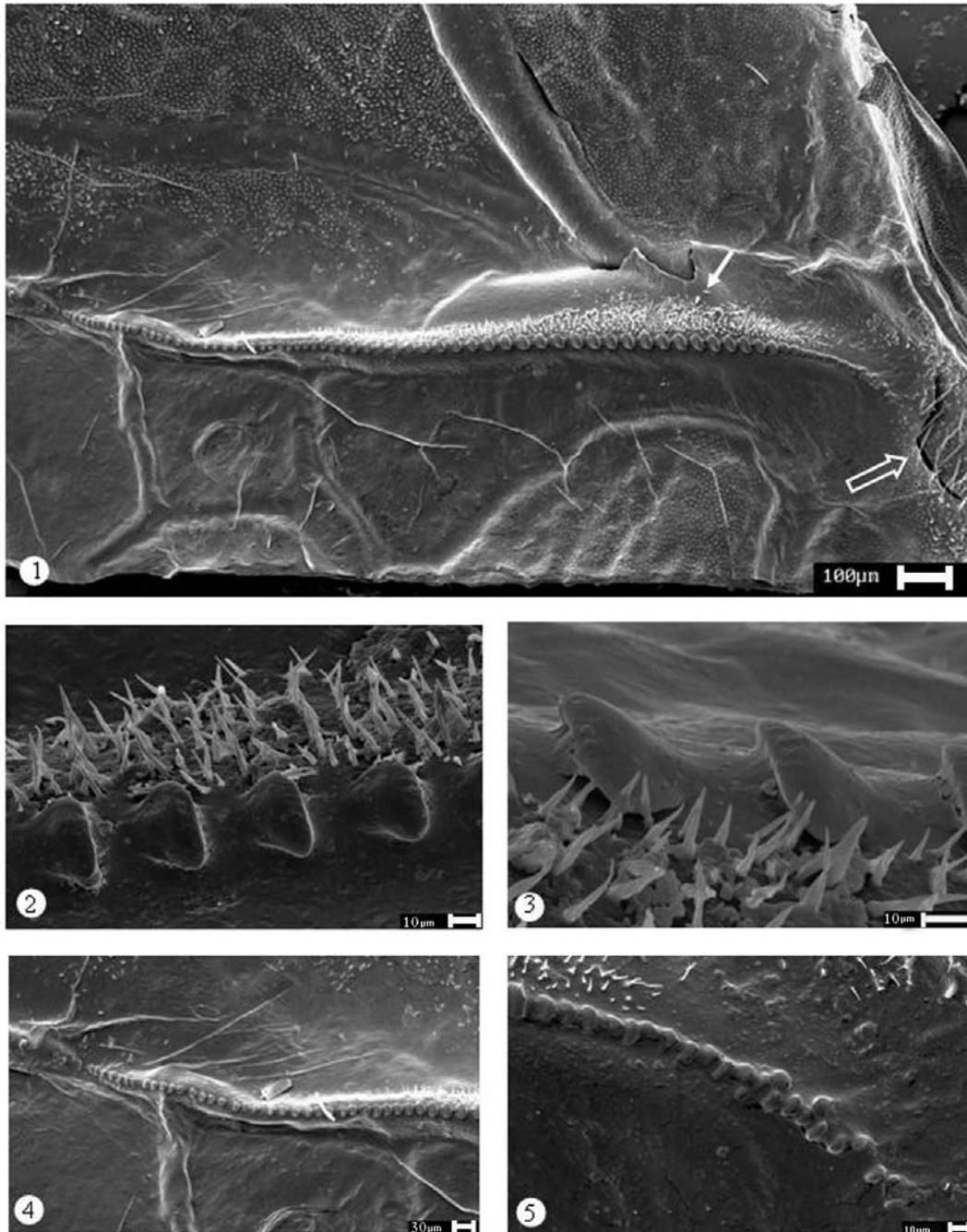
alignment and intermittent distribution are probably not scraped by the plectrum and consequently they are not involved in the production of the acoustic signals during stridulation. These kind of teeth had also been observed in the Phalangopsidae *Endecous itatibensis* Rehn, 1918, *Endecous betariensis* Mello & Pellegatti-Franco, 1998 and in several species of the genus *Gryllus*. The sonorous pulse are produced by the larger teeth located at the medium region of the stridulatory file.

The two sections (*a* and *b*) of the *trill* of *E. surinamensis* have distinct temporal features. In the

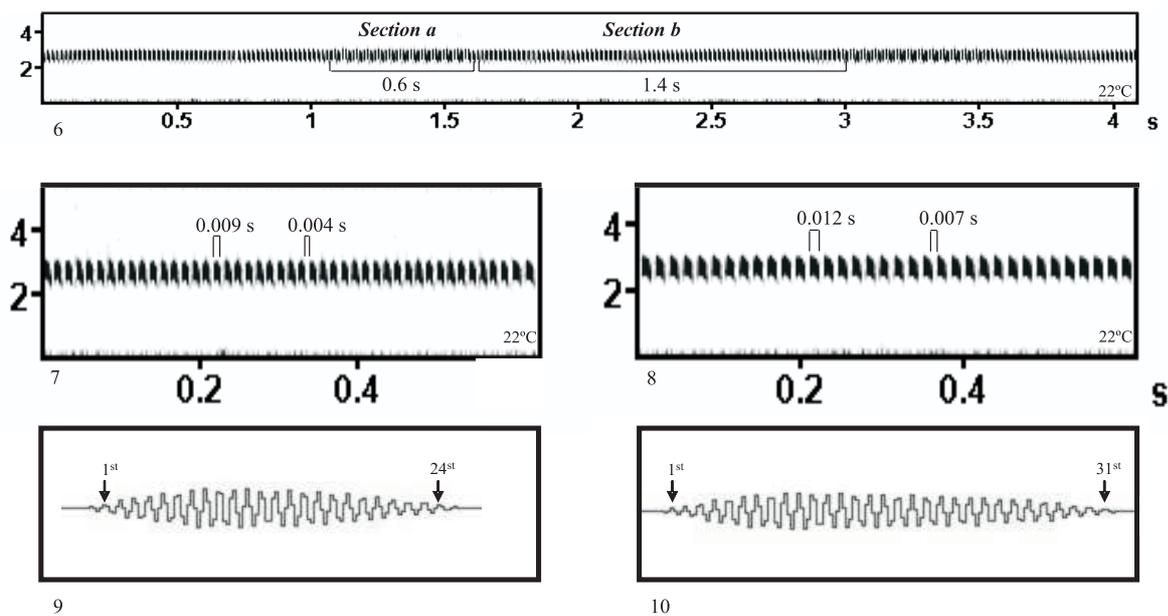
section *a*, each note has 24 pulses and in the section *b*, 31 pulses. Each pulse is produced by a tooth stroke (WALKER & CARLYSLE, 1975; LEROY, 1979), consequently the notes at the section *a* are produced by the action of 24 teeth and those of section *b* for 31 teeth.

The elapse time between notes is shorter in the section *a* (0.004 s) than in section *b* (0.007 s), due to the fact that the plectrum covers a shorter distance during the wing opening movement.

There is no modulation, contrasting with pattern verified by ROBILLARD & DESUTTER-GRANDCOLAS (2005)



Figs. 1-5. Scanning Electron Microscope image of the stridulatory file teeth of the right tegmen of *Eneoptera surinamensis* (De Geer, 1773): 1, stridulatory file teeth (white arrow shows the microtrichia and the hollow arrow indicates the hair plate); 2, 3, ventral and lateral view of the teeth in the mid portion of the file, respectively; 4, 5, smaller teeth located at the right and left ends of the file, respectively.



Figs. 6-10. Sonograms of the calling song of *Eneoptera surinamensis* (De Geer, 1773): 6, trill with two distinct sets of notes: section *a* and *b*; 7, trill sequence with notes of 0.009s and interval time between notes of 0.004 s; 8, trill sequence with notes of 0.012 s and interval time between notes of 0.007s (frequency in kHz); 9, 10 oscillogram showing one note expanded from 1st to 24th pulse and from 1st to 31st pulse, respectively.

for *E. guyanensis*. This closely-related species shows a calling song with a *trill* alternating low pitch (around 4,000 Hz) and high pitch (around 14,000 Hz). This modulation does occur due to a stridulatory file divided into two sections containing different kinds of teeth, one section with teeth very widely separated and the other one with teeth very close together (DESUTTER-GRANDCOLAS, 1998). *Eneoptera surinamensis* presents teeth morphology and linear distribution similar to that *E. guyanensis*' section file with widely separated teeth, and both produce lower frequencies.

The microtrichias located at the tegmen of *E. surinamensis* here analyzed had also been verified by DESUTTER-GRANDCOLAS (1995, 1998) in *E. guyanensis*, *Lerneca fuscipennis* (Saussure, 1874), *Oecanthus pellucens* (Scopoli, 1763) and *Gryllus campestris* Linnaeus, 1758. In *E. surinamensis* the microtrichias are distributed parallel the 31 teeth involved in the stridulating process.

The postcubital hair plate observed in *E. surinamensis* controls the tegmen opening angle and normally prevents the tegmen from overlapping left over right instead of right over left (ELLIOTT *et al.* 1982; ELLIOTT & KOCH, 1983; ELLIOTT, 1983; SCHÄFFNER & KOCK, 1987).

The stridulatory apparatus is the structure that produces the acoustic signals directly involved in the reproductive process of the crickets. The variations of these elements in isolated populations may indicate a behavioral and consequently reproductive isolation, thus contributing to the speciation process.

The wide geographic distribution of *E. surinamensis* in the Neotropical region, as listed by ROBILLARD & DESUTTER-GRANDCOLAS (2005) and supported by our field observation, could result in occurrence of

several isolated populations. The developed functional wings of *E. surinamensis* must allow high vagility and consequently high gene flow. Although specimens here analyzed are distant 1,000 km from each other, file features and sonograms have similar composition, showing that these elements remain conserved in both populations.

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