



# Eavesdropping on sexual vibratory signals of stink bugs (Hemiptera: Pentatomidae) by the egg parasitoid *Telenomus podisi*

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Males and females of the Neotropical brown stink bug, *Euschistus heros*, communicate with vibratory songs transmitted through plants. In this study, we examined the responses of the egg parasitoid *Telenomus podisi* to vibratory signals of these bugs. *Telenomus podisi* females responded with orientated movements to the vibratory signals of female songs on plants and on an artificial substrate, whereas male songs, duets or continuous pure tone vibratory signals failed to elicit any response. In addition, *T. podisi* females had a higher turning rate when stimulated with female songs, indicating some effect on their kinetic behaviour. This is the first demonstration of a parasitoid eavesdropping on the sexual vibratory signals of insects. The potential mechanism of this directionality and its adaptive significance are discussed.

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Animal communication is the exchange of information between individuals. In this exchange, one individual (signaller) transmits information to others (receivers), with both signaller and receivers being expected to benefit (Greenfield 2002). Sexual communication in particular is characterized by the use of conspicuous, long-range, nondirectional signals (Mougeot & Bretagnolle 2000) that can be intercepted by eavesdroppers to extract information. Unintended or 'illegitimate' receivers can be competitors, predators or parasites (Burk 1982; Zuk & Kolluru 1998; Mougeot & Bretagnolle 2000; Deecke et al. 2005).

Signals that are used for sexual communication can also be used by predators and parasitoids to locate prey/hosts, thereby increasing the risk of mortality for the signaller, especially for males, which are the main signallers (Burk 1982). This exploitation has been well documented for

insects and birds that use visual signals, for arthropods, reptiles, birds and mammals (bats and opossums) that use acoustic signals and for insects (parasitoids and predators) that use chemical signals (Zuk & Kolluru 1998 and references therein; Mougeot & Bretagnolle 2000; Müller & Robert 2002).

Predation pressure can have important implications for the evolution of prey communication. Antipredatory behaviour, especially in acoustic communication, includes the production of signals when predators are not active, a reduction in the duration of calling, the cessation of calling when predators are present or singing in choruses (Ryan et al. 1981; Zuk & Kolluru 1998; Mougeot & Bretagnolle 2000). Other forms of antipredatory behaviour can include changes in signal characteristics (Verrel 1991; Rotenberry et al. 1996), switching from an advertisement call to a lower-intensity courtship call (Greenfield 2002) and supplementing acoustic communication or shifting towards substrate-borne vibratory communication (Belwood & Morris 1987).

In parasitoids, which are known to eavesdrop on the chemical and acoustic signals of their hosts, host location includes several steps that are mediated by different types of cues. Chemical cues are used mainly

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on a long-range scale (Vinson 1985; Godfray 1994), whereas over short distances, visual and mechanical cues may also be included (Vinson 1985; Godfray 1994). This information can be used separately, hierarchically or in a combined, integrated multimodal signal (Fischer et al. 2001).

To date, there are no records of parasitoids eavesdropping on the substrate-borne vibratory communication of insect hosts. However, several reports have shown that some species of parasitoid wasps (Braconidae, Eulophidae and Pteromalidae) locate their hosts, such as miners, stem and fruit borers, seed predators and litter dwellers that live in concealed habitats, by using the vibrations produced during host movement, foraging or other activities (Meyhöfer et al. 1997; Meyhöfer & Casas 1999; Broad & Quicke 2000; Vilhelmsen et al. 2001). Additionally, some orussid (Vilhelmsen et al. 2001) and ichneumonid (Wäckers et al. 1998; Broad & Quicke 2000) parasitoid wasps use echolocation to find their hosts.

Egg parasitoids, especially *Telenomus* spp. and *Trissolcus* spp. (Hymenoptera: Scelionidae), are the most common natural enemies of pentatomid bugs. These parasitoids use chemical signals from plants (Colazza et al. 2004; Moraes et al. 2005b) and cues from host chemical communication (Mattiacci et al. 1993; Medeiros et al. 1997; Borges et al. 1998; Colazza et al. 1999; Conti et al. 2003) for long-range orientation, whereas chemical and visual cues are used for short-range orientation and host selection (Sales et al. 1980; Bin et al. 1993; Borges et al. 1999, 2003; Colazza et al. 1999; Conti et al. 2003). The use of vibratory signals for host location has not yet been demonstrated for any scelionid parasitoid.

Stink bugs (Hemiptera: Pentatomidae) communicate with substrate-borne vibratory signals during mating. Species- and sex-specific songs have been described in several species (Čokl & Virant-Doberlet 2003; Gogala 2006), and different vibratory songs have recently been described and correlated with reproductive behaviour in the Neotropical species *Acrosternum impicticorne* (Stål), *Euschistus heros* (F.), *Piezodorus guildinii* (Westwood) and *Thyanta perditor* (F.) (Moraes et al. 2005a).

Vibratory communication may have evolved in response to predation pressures (Henry 1994), and may be a strategy that prevents the hosts from being perceived by predators (Belwood & Morris 1987). However, specialized natural enemies (such as scelionid egg parasitoids in stink bugs) can evolve mechanisms to breach this gap and use these signals to search for hosts.

In this work, the hypothesis that the scelionid wasp *Telenomus podisi* (Ashmead) uses the vibratory signals of pentatomid bugs when searching for hosts was tested using the vibratory songs of the Neotropical brown stink bug, *Euschistus heros* (F.). In addition, we also examined the influence of a gradient in signal intensity on the movement of the wasps to the source of vibration. We chose *E. heros* because it is the main host of *T. podisi* (Corrêa-Ferreira & Moscardi 1995; Medeiros et al. 1997; Corrêa-Ferreira 2002) and it is the main soybean pest in Brazil (Panizzi & Rossi 1991).

## METHODS

### Insects

Individuals of *T. podisi* were obtained from a laboratory colony started from adults collected near the Embrapa Genetic Resources and Biotechnology Laboratory in Brasilia, DF, Brazil (15°47'S, 47°55'W). The insects were maintained in an environmental chamber in plastic cages (25-cm<sup>2</sup> angle-necked tissue culture flasks; ICN Biomedicals, Irvine, California, U.S.A.). Host (*E. heros*) eggs were glued onto cardboard strips and exposed to parasitoids for 24 h and then removed and placed in glass tubes (7.5 × 1.3 cm) for incubation. Before their use in the bioassays, adult parasitoids were housed for 24–48 h in plastic cages to allow mating.

Nymphs and adults of *E. heros* were reared on sunflower seeds (*Helianthus annuus* L.), soybeans (*Glycine max* (L.) Merrill), raw peanut seeds (*Arachis hypogaea* L.) and green beans (*Phaseolus vulgaris* L.) in separate containers. The eggs were collected daily and kept separately in petri dishes until eclosion. Both insect species were reared on a 14-h photophase at 26.0 ± 1°C and 65 ± 10% relative humidity.

### Vibratory Signals

Parasitoids were stimulated artificially by continuous, pure-tone signals of 130 Hz (artificial signal) produced with Sound Forge 4.5 software (Sonic Foundry Inc., Madison, Wisconsin, U.S.A.) and by pre-recorded, naturally produced songs obtained from virgin and sexually mature *E. heros* adults, 15–25 days after the final moult (Costa et al. 1998). Female songs, male songs and duet songs were used. Five recordings of each song type obtained from different individuals were used in the experiments and each song was used in at least 5–10 bioassays. Control tests using no stimulus (no vibration) were also done.

Naturally produced *E. heros* songs were recorded from bugs singing on a nonresonant loudspeaker membrane and the signals captured by the loudspeaker were digitalized (Aardvark-Direct Pro 24/9; Aardvark Computer Systems, Ann Arbor, Michigan, U.S.A.) and stored on a computer using CoolEdit Pro software (Syntrillium Software 2001, Fort Wayne, Indiana, U.S.A.). The female stimulatory song consisted of short pulses (median pulse duration of 948 ms; Moraes et al. 2005a) in which a female started vibratory communication (females song 1, FS1, as defined by Moraes et al. 2005a). In contrast, male stimulatory song consisted of two types of male songs: male song 1 (MS1), which was characterized by short pulses with a mean pulse duration of 1756 ms, and male song 2 (MS2), which was characterized by long pulses with a mean pulse duration of 4407 ms (Moraes et al. 2005a). Male songs were normally produced in response to female pulses, and male song pulses were characterized by two spectral peaks within the dominant frequency domain (Fig. 1). A duet stimulatory song consisted of female song pulses that alternated with or were often superimposed between short (MS1) and long (MS2) male pulses that were produced in an irregular pattern.

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