N O T E

Presence of Tibial Spurs as a Male Sexual Character for *Galerucella calmariensis* (Coleoptera: Chrysomelidae)[1]

Allard A. Cossé[2]

Crop Bioprotection Research, National Center for Agricultural Utilization Research, USDA/ARS, 1815 N. University Street, Peoria, IL 61604 USA

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The leaf beetle, *Galerucella calmariensis* (L.), (Coleoptera: Chrysomelidae) is an effective biological control agent of the introduced perennial wetland plant, purple loosestrife, *Lythrum salicaria* L. (Malecki et al. 1993, BioScience 43: 680-686). The beetle is native to Europe but has been established in the United States. A pheromone could be a useful tool in monitoring the dispersal of *G. calmariensis,* especially during the early stages of beetle introductions in unpopulated stands of purple loosestrife.

During our investigation into possible pheromone component(s) for *G. calmariensis,* by chemical and electrophysiological comparisons of male and female volatiles, it was imperative to correctly identify and separate the sexes prior to volatiles collection. *Galerucella calmariensis* can be sexed by close examination of the ventral side of the last abdominal segment (Manguin et al. 1993, Ann. Entomol. Soc. Am. 86: 397-410). The apex of the last abdominal segment of male beetles has a more broadened v-shape indentation than that of the female. This sexual dimorphism might be a general guideline for sexing *G. calmariensis,* but this characteristic is somewhat subjective and not completely reliable. The discovery of male-specific tibial spurs made the sexing of *G. calmariensis* a simpler process. However, *Galerucella* is a relatively large genus, and the presence or absence of tibial spurs might vary from species to species.

The beetles were obtained from a laboratory culture maintained by the Illinois Natural History Survey, Champaign, IL, and kept on purple loosestrife according to methods described by Verlarde et al. (2002, BioControl 47: 587-601). Individual beetles were placed in Pasteur pipettes and examined under a Nikon SMZ-2B stereomicroscope. The beetles crawled into the tip of the pipettes until stopped by the decreasing diameter. Each was then held immobile by a wooden applicator stick placed gently against the end of the abdomen. It was easy to observe the beetles at different angles by rotating and tilting the pipettes. Beetles placed their tarsi against

[1]Received 10 March 2003; accepted for publication 18 October 2003.
[2]email address (cosseaa@ncaur.usda.gov).

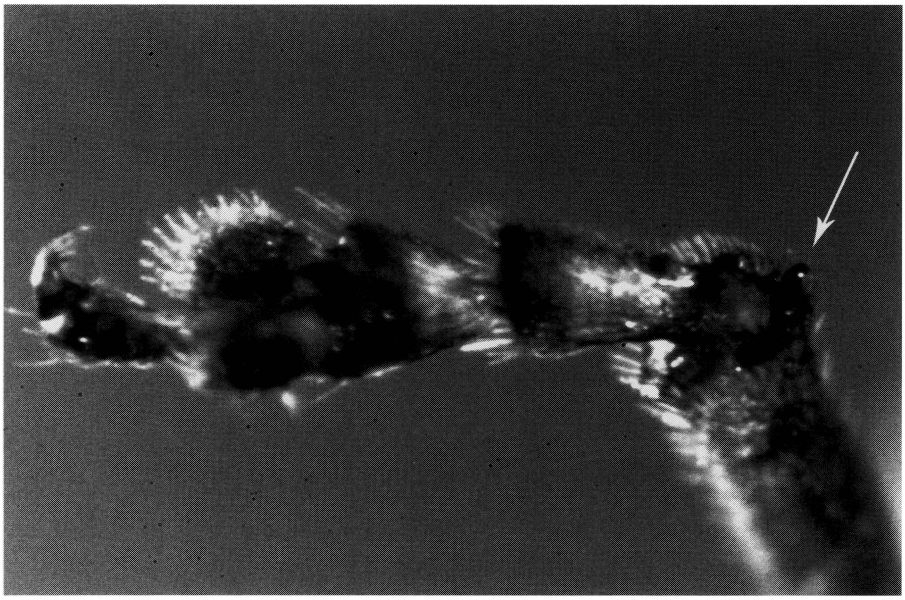

Fig. 1. Stereomicroscope image (30x) of the metatarsi and apex of the metatibia of male *G. calmariensis*. Arrow denotes the presence of a tibial spur.

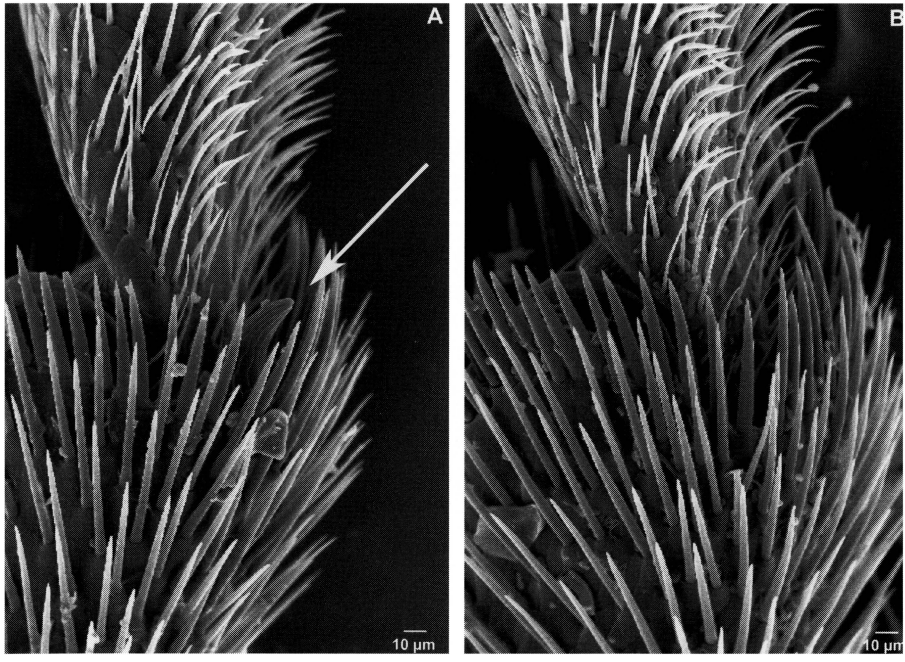

Fig. 2. Laterial view showing the presence and absence of a single spur on the apex of metatibia of male (A) and female (B) *G. calmariensis*. Arrow denotes the presence of a tibial spur.

the inner glass surface which gave a clear view of the tip of the tibia. Close examination (20-30x) showed that some of the beetles had a single black-colored spur at the distal end of the tibia (Fig. 1). These spurs were only observed on the meso- and metatibia and were clearly contrasted against the light brown color of the legs. Those beetles that showed tibial spurs were determined to be all males, those without the spurs were determined to be all female, following dissection and examination of the genitalia.

Scanning electron micrographs were taken using a JEOL JSM-6400V scanning microscope (Fig. 2A,B). The male tibial spur can be clearly observed as a curved cone-shaped structure in between the hairs on the apex of the tibia. These striated spurs (approximately 60 µm in length) were absent on the female tibia.

Using the presence of tibial spurs as a sexual characteristic, two workers were able to sex 500 *G. calmariensis* beetles in 2 h. A random sample of 25 beetles with spurs (males) and 35 beetles without spurs (females) were dissected and their genitalia were examined. The result of the dissections showed a 100% accurate sex determination.

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