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Effect of plant sex (dioecism) on the performance of *Apocnemidophorus pipitzi* (Coleoptera: Curculionidae), a stem boring weevil of Brazilian peppertree, *Schinus terebinthifolia*

**James P. Cuda**,*, Judy L. Gillmore, and Bolivar R. Garcete-Barrett*

Brazilian peppertree, *Schinus terebinthifolia* Raddi (Sapindales: Anacardiaceae) (Zona 2015), a perennial woody plant, is one of the worst invasive weeds in Florida because of its wide distribution, adaptation to various habitats, and demonstrated negative impacts on biodiversity (Cuda et al. 2006; Manrique et al. 2013; FLEPPC 2017). Introduced into Florida from South America as a landscape ornamental in the late 19th century, Brazilian peppertree escaped cultivation and currently dominates entire ecosystems in central and south Florida (Manrique et al. 2013; Rodgers et al. 2014). This invasive shrub grows rapidly via hybrid vigor (Williams et al. 2007; Geiger et al. 2011), tolerates a broad range of moisture and salinity conditions (Ewe and Sternberg 2002, 2003; Mytinger & Williamson 1987), exhibits allelopathy (Morgan & Overholt 2005; Donnelly et al. 2008), and is a prolific seed producer (Ewel et al. 1982; Spector & Putz 2006). In the 1980s, Brazilian peppertree was targeted for biological control because its aggressive nature was consistent with escape from natural enemies (Williams 1954).

Brazilian peppertree is a dioecious species (Burdon & Marshall 1981), and the presence of distinct male and female plants in a typical stand or population has important implications for its management. For instance, when resources are limited, effective control of Brazilian peppertree can be accomplished by matricide, which is the selective control of reproductively mature female plants to stop seed production (Cuda et al. 2006). Biological control is another management option but dioecy may present some unique challenges. Brazilian peppertree and other *Schinus* spp. synthesize toxic phenolic compounds (e.g., alkylcatechols and alkyfliescorinals) in their leaves to protect them from herbivores (Aguilar-Ortigoza & Sosa 2004; Wheeler et al. 2014). Furthermore, the expression of other secondary compounds (e.g., volatile terpenoids), presumably produced by Brazilian peppertree in response to herbivory (Barbosa et al. 2007; Wheeler et al. 2014), may be plant sex dependent. For instance, Campbell et al. (1980) observed the following, “When we stripped...leaves in Brazil we found that only known female plants displayed the familiar pungent odor when the leaves were crushed. Male plants did not have any odor ... Upon our return to the U.S., we applied the same test to female and male plants and got the same results in most cases ... What may be happening is that many chemical components of the fruit ... become concentrated at the growing points of female plants.” Only female Brazilian peppertrees synthesize cardanol, an alkylphenol produced in the fruit that causes dermatitis in sensitive individuals (Stahl et al. 1983). Therefore, it is conceivable that natural selection favored the synthesis of a higher concentration of secondary compounds in female plants relative to males to deter herbivory and ensure their survival.

In Mar 2007, a weevil identified as *Apocnemidophorus pipitzi* (Faust) (Coleoptera: Curculionidae) was collected as a potential biological control agent of Brazilian peppertree in the Itapúa Province of Paraguay (Cuda et al. 2016). Adults feed mainly on the upper surface of subterminal leaflets, where they produce a characteristic notching pattern. Females deposit eggs in the stems and larvae feed under the bark where they damage the vascular cambium. To test the hypothesis that female Brazilian peppertree plants may be better defended chemically than their male counterparts, we compared the performance (adult feeding and survival) of the specialist herbivore *A. pipitzi* on foliage from both male and female plants.

Plants were propagated at the University of Florida, Entomology & Nematology Department nursery using methods described by Cuda et al. (2016). In total, 20 adult weevils were caged with a fresh cut bouquet of either male or female terminal shoots with about 25 leaflets placed in a vial containing water. Plant sexes were differentiated by examination of the flowers (Gioeli & Langeland 1997). Cages (Bug Dorm® collapsible cages 30.5 cm × 30.5 cm × 30.5 cm, BioQuip® Products, Rancho Dominguez, California, USA) were located in a rearing room at the Entomology Department Containment Laboratory. Temperature, RH, and photoperiod were 25.8 to 27.2 °C, 53 to 57% RH, and 12:12 h L:D, respectively. Cages containing female bouquets were separated from those with male plant bouquets. Bouquets were replaced with fresh plant material about 3 times per week until all the adults were dead. As plant material was removed, feeding damage on leaf tissues was measured in mm² under a dissecting microscope. Weevil mortality was quantified using LT₅₀ (lethal time required to kill 50% of the weevils per cage). There were 3 cages per treatment containing bouquets of either male or female plants, and data were analyzed with a paired t-test (JMP 2012; de Winter 2013). All data are reported as $\bar{x} \pm$ SEM.

Male plants sustained significantly higher feeding damage compared to female plants (Fig. 1). Weevils consumed on average 392.68 ± 76.12 cm² of leaf tissue from male plants compared to 303.44 ± 53 cm² for female plants ($t = 3.05; df = 4; P = 0.027$). Furthermore, adults survived significantly longer on male plants compared to female plants (Fig. 2). Weevil longevity (LT₅₀) was almost twice as long (91.67 ± 12

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days) on male plants compared to female plants (52.33 ± 6.8 days) (t = 2.71; df = 4; P = 0.029). These results suggest that female Brazilian peppertree plants are better defended chemically than male plants.

Sex-related differences in herbivore damage, with male plants suffering higher levels of feeding damage than female plants, have been documented in other plant systems (Bawa 1980, Cornelissen & Stiling 2005). Furthermore, in contrast to other studies differences in herbivore survival also were observed between plant sexes. Female Brazilian peppertree plants probably contain higher levels of volatile terpenoids, which are feeding deterrents or toxicants. At least 7 volatile terpenoids have been isolated from Schinus haplotypes A and B found in Florida (Wheeler et al. 2014). Herbivore induced, differential expression of these compounds evidently affords seed bearing female plants greater protection from herbivory. Further studies are needed to quantify differences in secondary plant chemistry profiles in male and female Brazilian peppertree plants.

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Summary

Brazilian peppertree, Schinus terebinthifolia Raddi (Sapindales: Anacardiaceae), is an ornamental dioecious shrub introduced from South America that has become one of the major invasive weeds in Florida. Anecdotal evidence from the literature suggests that crushed leaves from female plants produce a more pungent aroma, indicating these plants may be better defended chemically than their male counterparts. To test this hypothesis, we compared the performance (adult feeding and survival) of the South American stem boring weevil Apocnemidophorus pipitzi (Faust) (Coleoptera: Curculionidae) by caging insects on foliage collected from both male and female Brazilian peppertree plants. Results showed that weevils consumed significantly less leaf tissue from female plants compared to male plants. Furthermore, the LT50 (average time required to cause 50% mortality of the weevils) was significantly shorter when weevils fed exclusively on female leaf tissue. Our findings confirmed male-biased plant herbivory on Brazilian peppertree by one of its folivorous natural enemies.

Key Words: Plant-insect interactions; toxic compounds; weed biocontrol; sex-biased herbivory

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