Evaluation of Chinese Centipedegrasses and other Turfgrass Taxa for Potential Resistance to Twolined Spittlebug, *Prosapia bicincta* (Say) [1]

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Abstract Warm season turf taxa of centipedegrass [*Eremochloa ophiuroides* (Munro) Hack], bermudagrass [*Cynodon* L.C. Rich. spp.], St. Augustinegrass [*Stenotaphrum secundatum* (Walt.) Kuntze], and zoysiagrass [*Zoysia* Willd. spp.] were evaluated for tolerance to adult twolined spittlebug (*Prosapia bicincta* Say) feeding in choice and no-choice experiments, and for their ability to support nymphal development (antibiosis potential). Among 133 selections evaluated, few showed evidence of potential antibiosis and/or improved tolerance over commercially available cultivars. Most of the centipedegrass taxa evaluated were susceptible to the spittlebug. However, some potential antibiosis among Chinese centipedegrass taxa was identified, and there was a gradient in the ability to tolerate spittlebug feeding. Among centipedegrasses, TC 358 and TC 362 showed moderate tolerance and recovery in no-choice and choice trials. The most tolerant bermudagrasses in no-choice trials were 00 - 23, 03 - 14, and 03 - 15. Centipedegrasses overall were the best hosts for nymph development, but TC 379, TC 422 and *E. ciliaris* did not support nymph development, and TC 341 and TC 399 showed very low numbers of nymphs. The bermudagrasses 00 - 23 and 00 - 28 and the St. Augustinegrasses T638 and Mercedes failed to support nymph development.

Key Words twolined spittlebug, *Prosapia bicincta,* host plant resistance, turfgrass, bermudagrass, centipedegrass, seashore paspalum, zoysiagrass

Centipedegrass, *Eremochloa ophiuroides* (Munro) Hack., is a warm season turfgrass that grows well on a wide range of soil types. It has great potential for expanded use for lawns, roadsides, and commercial properties because it requires less management and fertilization to produce an acceptable turf than most warm season turfgrasses (Hanna 1995). Centipedegrass is indigenous to Southeast Asia and was introduced into the United States in 1916. Since its introduction, it has become widely used in the Southeast where it is vulnerable to infestation and damage by the twolined spittlebug, *Prosapia bicincta* (Say). Deployment of pest-resistant plants in production and landscape settings provides a proactive solution to key pest problems in a manner acceptable to the general public (Braman et al. 2000c).

The spittlebug genus, *Prosapia* Fennah, occurs in North America, with most species occurring in Mexico and Central America (Hamilton 1977). *Prosapia bicincta* has been reported from Florida to Maine (Byers 1965) and as far west as Texas and

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Arkansas in the United States (Braman and Ruter 1997). Cercopids (at least 8 species) limit the establishment of *Brachiaria* (Trin.) Griseb. spp. grasses in the South and Central American rangeland. Intensive breeding programs for resistance are underway (LaPointe et al. 1992). Nymphs and adult twolined spittlebugs are opportunistic xylem feeders, with reports of more than 40 hosts (Fagan and Kuitert 1969). The twolined spittlebug is a recognized pest of bermudagrass (*Cynodon* L.C. Rich.) pastures (Byers 1965, Fagan and Kuitert 1969, Taliaferro et al. 1973). Damage has been reported on other grasses such as Pangola grass, *Digitaria decumbens* Stent. and St. Augustinegrass, *Stenotaphrum secundatum* (Walt.) Kuntze (Genung and Green 1974). Economic damage also has been reported on ornamental hollies, *Ilex opaca* L., *I. cornuta burfordii* De France, and *I. cassine* L. (Braman and Ruter 1997), and on southern lawns planted with centipedegrass or bermudagrass (Braman and Abraham 2012, Potter and Braman 1991).

Forage grass response to various spittlebugs has been investigated (Cardona et al. 1999, LaPointe et al. 1992, Miles et al. 1995, Stimmann and Taliaferro 1969) and in another study, (Shortman et al. 2002) potential resistance to *P. bicincta* was evaluated among 56 turfgrass genotypes. In that study, greenhouse and laboratory bioassays identified differences in spittlebug survival and development, host preference and damage levels, and turfgrass tolerance to and ability to recover from spittlebug induced injury. However, all centipedegrasses demonstrated high levels of susceptibility, followed by bermudagrasses, seashore paspalums (*Paspalum vaginatum* Swartz) and zoysiagrasses (*Zoysia* Willd. spp.). Average nymphal survival to the adult stage in that study ranged from 1.5 - 78.1%. Development required 38.1 - 62.0 days depending on plant taxa under greenhouse conditions.

Liu et al. (2002) reported the collection of centipedegrass from 58 sites in central and south China. These taxa, found growing well under a diversity of conditions, were classified into the following groups: dwarf and aggressive, coarse and aggressive, dwarf and slow-growing and intermediate. Here, we report the relative resistance of 133 turfgrass taxa, including 55 Chinese centipedegrass selections and 9 centipedegrass breeding lines, to the twolined spittlebug.

Materials and Methods

Insects and plants. Adult twolined spittlebugs were field collected from local residential and commercial landscapes in Spalding Co., GA. The spittlebugs were maintained on turfgrass in acrylic cylinders (Pioneer Plastics, Inc.; Dixon, KY 279C, 10.5 cm diameter X 21 cm height). Adults were placed in environmental chambers (Percival Scientific, Inc.; Perry, IA) and maintained at 24°C, 85% RH and on a 15:9 (L:D) h. Adults were provided with moistened filter paper as oviposition sites. Eggs were collected daily, placed on moistened filter paper in 10-cm Petri dishes and maintained in environmental chambers under the same conditions.

Turfgrass taxa evaluated included 55 Chinese centipedegrass accessions, 8 centipedegrass breeding lines, 1 fringed centipedegrass taxon [*Eremochloa ciliaris* (L.) Merr.], two seeded bermudagrasses, 57 triploid bermudagrasses, 6 St. Augustinegrasses, and 4 zoysiagrasses. Some of these grasses have already been evaluated for their relative resistance to mole crickets, *Scapteriscus* Scudder spp. (Braman et al. 1994, Braman et al. 2000b) and fall armyworms, *Spodoptera frugiperda* (J.E. Smith) (Braman et al. 2000a, Braman et al. 2002b). Grasses (24 replicates of 133 plant selections) were planted in Metro mix 300 potting media (Scotts-Sierra Horticultural, Marysville, OH) into single cell cone-tainers (SC-10 Super cells 3.8 cm diameter X 21 cm height, 98 cells per tray, 30X61X18 cm, Stuewe & Sons, Inc., Corvallis, OR).

No-choice tolerance evaluation. Tolerance tests were conducted by caging adults on plants in a randomized complete block design with 4 replications of each of the 133 grasses. Grass survival and appearance were compared with uninfested plants. One male and one female *P. bicincta* of undetermined age were confined to single plant cone-tainer units using the fiber milk test sleeves (milk test filters, Kleen-test Products, Milwaukee, WI). An identical set of plants was caged, but did not receive spittlebugs. A third set of plants received no cage and no spittlebugs. Spittlebugs were confined to the plants for 7 d. At the end of the exposure period, cages and spittlebugs were removed. Numbers of green versus number of brown stems were counted for every entry in each of the 3 categories: cage with spittlebugs, cage without spittlebugs, and no cage-no spittlebug. Data expressed as percentages of brown stems were transformed (arcsin square root transformation) prior to analysis of variance (ANOVA) using the general linear model procedure (PROC GLM) of SAS (SAS Institute 2003). Means were separated using the least significant difference test (LSD). Species-related differences in tolerance to adult spittlebug injury were identified using the LSD test following a significant ANOVA.

Free choice tolerance evaluation. Sixty eight turfgrass taxa (64 centipedegrasses and 1 each of bermudagrass, St. Augustinegrass, zoysiagrass and seashore paspalum) were evaluated again in tolerance/preference tests in cages in a greenhouse. Twelve wooden- frame 1 m^3 cages covered with 32- mesh nylon screen were infested with 70 adult spittlebugs. The adults were allowed to settle and feed on the 68 grasses which had been arranged in a randomized complete block design within the cages (=reps). Damage ratings were taken at 4 and 30 d postinfestation. Grasses in their cone-tainers were assigned a rank from 0 (no damage) to 100 (completely brown). Grasses were irrigated, but not fertilized, during the course of the trial. Spittlebugs were not removed from the cages. Data were transformed (arcsin square root transformation) prior to analysis using the GLM procedure and mean separation was by LSD (SAS Institute 2003). Species-related differences in tolerance to/ non preference by adult spittlebugs were identified using the LSD test following a significant ANOVA.

Evaluation for potential antibiosis. Eggs from the spittlebug colony were held until eyespot stage, extracted from filter paper and transferred to filter paper wedges. Wedges containing 3 eggs were transferred to single plants. Hatching spittlebugs were confined to tubes using fiber sleeves (milk test filters, Kleentest Products, Milwaukee, WI). Two trials with 3 replications each of the 133 genotypes were arranged in a randomized complete block design, one in a greenhouse and the other in a growth chamber. The grasses were grown with a weekly fertilization program using a water-soluble general purpose fertilizer (Peter's 20 - 20 - 20; The Scotts Company, Marysville, OH). Plants were observed for the presence of twolined spittlebug spittle masses, nymphs, and adults. Nymphal growth and survival were evaluated. Number of nymphs surviving to day 30 among grass taxa was compared using GLM procedure, followed by mean separation by LSD (SAS Institute 2003). Species-related differences in development were identified using the LSD test following a significant ANOVA.

Results

No-choice tolerance evaluation. A wide range in adult spittlebug-induced injury was observed in no-choice tests among the grass taxa evaluated (Table 1). Turfgrass

Table 1. Mean ± SE damage /impact evaluation of *P. bicincta* adults feeding for 7 days on *Eremochloa*, *Cynodon*, *Stenotaphrum* and *Zoysia* spp. genotypes in a greenhouse no-choice trial.

Grass Type	Genus	Mean % brown stems	No. of accessions with score (% brown stems)				
			1 (0 - 5)	2 (5 - 30)	3 (30 - 60)	4 (60 - 80)	5 (80 - 100)
Centipede*	*Eremochloa*	65.9 ± 2.0 A	0	1	24	23	15
Bermuda	*Cynodon*	61.9 ± 2.2 A	0	4	26	12	17
St. Augustine	*Stenotaphrum*	16.7 ± 5.1 B	2	3	1	0	0
Zoysia	*Zoysia*	21.1 ± 7.4 B	1	3	0	0	0

* One accession (*E. ciliaris*) failed to survive.

Means ± SE followed by the same letter are not significantly different, $LSD_{0.05}$.

species varied in ability to tolerate spittlebug feeding ($P < 0.0001$). Tolerance also varied by turf taxa within a species ($P < 0.0001$). Damage ranged from 1.9% on 'Mercedes' St. Augustinegrass to 97.3% on TC 400 centipedegrass when 2 spittlebugs were allowed to feed for 7 d on fully-rooted 3.8 cm diam. grass plugs. Damage to centipede and bermudagrass on average was similar, 65.9 and 61.9% respectively. St. Augustinegrass and zoysiagrass were less severely damaged by spittlebugs, expressing 16.7 and 21.1% injury on average, respectively (Table 1). Thirteen of 64 centipedegrasses displayed at least 50% normal growth after injury, with the remaining 51 being more severely damaged and, therefore, not discussed. The 13 taxa were TC 341, TC 342, TC 344, TC 346, TC 347, TC 354, TC 358, TC 362, 'TifBlair', TC 390, TC 423, TC 438, and TC 441 in this no- choice trial. Twenty of the 59 bermudagrasses met this criterion: 00 - 10, 00 - 14, Tifway, 00 - 18, 00 - 21, 00 - 22, 00 - 23, 00 - 27, 03 - 12, 03 - 14, 03 - 15, 03 - 17, 03 - 18, 03 - 19, 03 - 20, 03 - 21, 03 - 23, ST-3, ST-4 and TifGrand. All of the St. Augustine and zoysiagrasses evaluated maintained at least 50% of their normal green growth (Fig. 1). Caging grasses had little to no effect on zoysiagrass, St. Augustinegrass and bermudagrass entries, resulting in 0, 0, and 0.1% brown stems due to caging, respectively, among the 3 species. Caging centipedegrass in the absence of spittlebugs resulted in a 6.7% browning compared with uncaged grasses that were not infested (data not presented).

Free-choice tolerance/preference evaluation. When a heavy infestation of adults had free access to 68 different grass taxa, heavy damage was sustained by most centipedegrasses within 4 d (Table 2). Damage ranged from 4.4% on the *Paspalum* selection to 89.5% on TC 341 ($P < 0.0001$). By day 30, many selections showed 100% damage, however, 44 of the 68 selections evaluated demonstrated slight to substantial recovery ($P < 0.0001$). Within genera, centipede and bermudagrass expressed 72.5 and 58.0% damage by day 4, respectively ($P < 0.0001$). St. Augustine and zoysia were again less severely impacted at 36.4 and 40.9% damage (Table 3). By day 30, the zoysiagrass was showing less damage than it had at day 4 ($F = 80.8$; df = 11,4; $P < 0.0001$). Centipedegrasses showing moderate tolerance and recovery included TC 324, TC 325, TC 326, TC 338, TC 358, TC 359, TC 360, TC 361, TC 362, TC 396, TC 400, and TC 401. Taxa that continued to display improved

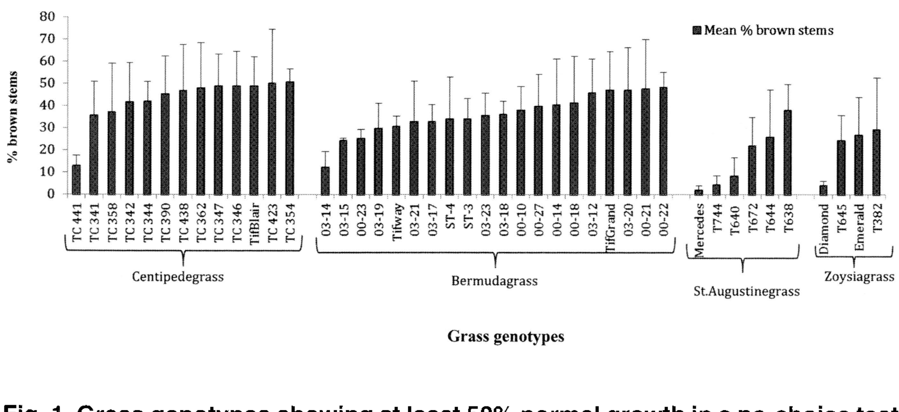

Fig. 1. Grass genotypes showing at least 50% normal growth in a no-choice test after feeding by 2 adult *Prosapia bicincta* for 7 days.

Table 2. Mean ± SE damage /impact evaluation of *P. bicincta* adults feeding for 7 and 30 days on *Eremochloa, Cynodon, Stenotaphrum, Zoysia* and *Paspalum* spp. genotypes in a greenhouse free-choice trial.

Grass Type	Genus	Mean damage rating of infested plants- day 4	Mean damage rating of infested plants- day 30
Centipede	*Eremochloa*	72.5 ± 0.7 A	91.7 ± 0.8 A
Bermuda	*Cynodon*	58.0 ± 7.6 AB	95.1 ± 3.4 A
St. Augustine	*Stenotaphrum*	36.4 ± 5.0 C	52.0 ± 8.6 B
Zoysia	*Zoysia*	40.9 ± 4.4 BC	36.2 ± 7.9 B
Paspalum	*Paspalum*	4.4 ± 1.8 D	15.8 ± 0.4 C

Means ± SE followed by the same letter are not significantly different, $LSD_{0.05}$.

resistance/tolerance to heavy spittlebug infestation by day 30 were TC 325, TC 400 and TC 401 (Fig. 2).

Evaluation for potential antibiosis. Centipedegrass was the best host for spittlebug nymphal development among the 133 taxa in the greenhouse evaluation ($P < 0.0001$). On average, half of the eggs placed on centipedegrass produced healthy nymphs surviving to day 30 on centipedegrass ($P < 0.0001$) (Table 3). However, the centipedegrasses TC 379, TC 422 and *E. ciliaris* did not support nymph development, and TC 341 and TC 399 showed very low numbers of nymphs. Numbers of nymphs on bermudagrasses were significantly lower than those on centipedegrasses, and 00 - 23 and 00 - 28 appeared most unfavorable for nymphal development with no nymphs recorded at 30 d. Among St. Augustinegrasses T638 and 'Mercedes' failed to support nymphal development. The other St. Augustinegrasses and all of the zoysiagrasses showed low numbers of nymphs (Fig. 3).

Discussion

This study evaluated the response of 133 warm season turfgrass taxa to feeding and nymph development of the twolined spittle bug *P. bicincta*. Our selection of

Table 3. Mean number (of three original eggs) of twolined spittlebugs surviving to day 30 on *Eremochloa, Cynodon, Stenotaphrum* and *Zoysia* spp. genotypes in no-choice trials (averages over two trials).

Grass Type	Genus	Mean no. nymphs alive on day 30
Centipede	*Eremochloa*	1.4 ± 0.1 A
Bermuda	*Cynodon*	0.8 ± 0.1 B
St. Augustine	*Stenotaphrum*	0.4 ± 0.1 C
Zoysia	*Zoysia*	0.8 ± 0.2 B

Means ± SE followed by the same letter are not significantly different, $LSD_{0.05}$.

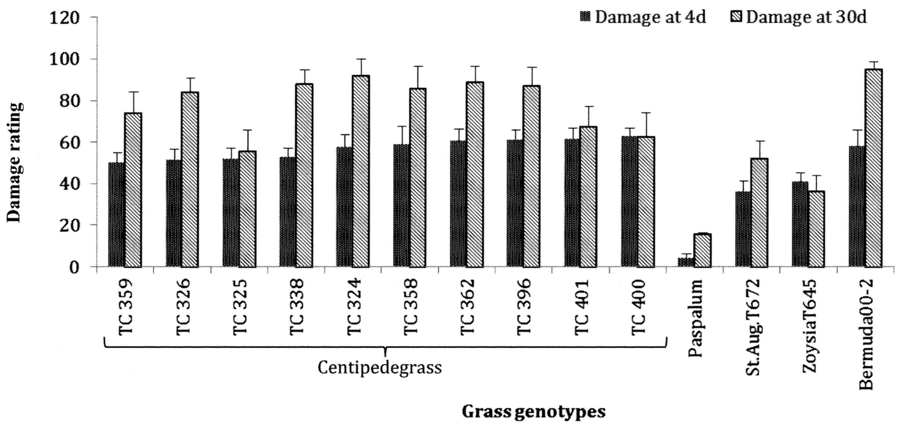

Fig. 2. Grass genotypes showing moderate tolerance and recovery after heavy spittlebug (*Prosapia bicincta*) infestation (70 adults) in a free-choice test.

experimental grasses included taxa of centipedegrass, bermudagrass, zoysiagrass and St. Augustinegrass, which provided a good representative framework for comparison. Several taxa showed improved tolerance and evidence of potential antibiosis, as indicated by mortality of nymphs, over commercially available cultivars. Among the 4 genera evaluated in no-choice trials, centipedegrass and bermudagrass appeared to be more susceptible to *P. bicincta* than St. Augustinegrass and zoysiagrass, in terms of damage caused by feeding. Centipedegrass also appeared highly susceptible in the free choice trials, but some taxa displayed good tolerance and recovery. Overall, the grasses in the free choice trials maintained a better appearance on day 4, than on day 30 because of the intense infestation pressure that was used in this experiment. Taxa that were able to show less damage under these conditions at day 4 were interpreted to have resistance potential. The centipedegrasses that showed this potential in both no-choice and choice trials were TC 358 and TC 362. However, the centipedegrasses that showed antibiosis potential (as indicated by nymph mortality)

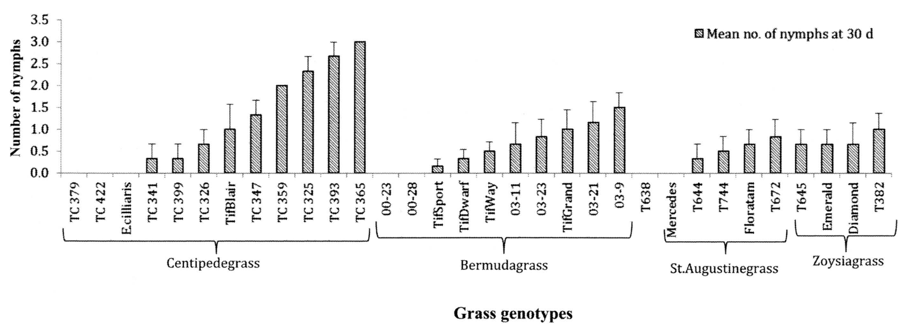

Fig. 3. Variation in spittlebug (*Prosapia bicincta*) nymph emergence on different grass genotypes in no-choice test.

were TC 379, TC 422, *E. ciliaris,* TC 341 and TC 399. Of these, TC 341 showed moderate tolerance and recovery in no-choice trials, but not in free-choice trials.

Cardona et al. (2004) discuss the inherent risk in deploying tolerance alone to spittlebugs (rather than in combination with antibiosis) in their evaluation of *Brachiaria* spp. grasses for resistance to 5 species of spittlebugs in tropical America. When large contiguous areas are planted with grasses that have demonstrated tolerance, spittlebugs may build to high population levels causing severe damage. Although that discussion concerned forage grasses, the concept is relevant for large contiguous plantings of managed turfgrasses where entire residential neighborhoods, commercial properties or sod farms may be planted with centipedegrass. What is needed is tolerance coupled with antibiosis properties capable of causing significant mortality. Information on the specific attributes of different turfgrass taxa will be useful to breeders trying to integrate these characters in hybrid turfgrasses. The interaction of resistant plants and natural enemies of the key pests targeted by deployed resistance must be understood for successful development of sustainable strategies (Braman et al. 2002a, Braman et al. 2003, Braman et al. 2004). Further work on resistance mechanisms and their effect at all trophic levels will help direct future breeding studies.

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References Cited

Braman, S. K. and C. M. Abraham. 2012. Two-lined spittlebug, Pg. 77-79. *In* R. L. Brandenburg and C.P. Freeman [eds.], Handbook of Turfgrass Insects 2nd Edition, Entomological Society of America, Lanham, MD.

Braman, S. K. and J. M. Ruter. 1997. Preference of twolined spittlebug for *Ilex* species, hybrids and cultivars. J. Environ. Hortic. 15: 211-214.

Braman, S. K., R. R. Duncan and M. C. Engelke. 2000a. Evaluation of turfgrass selections for resistance to fall armyworms (Lepidoptera: Noctuidae). HortScience 35: 1268-1270.

Braman, S. K., A. F. Pendley and N. Corley. 2002a. Influence of commercially available wildflower mixes on beneficial arthropod abundance and predation in turfgrass. Environ. Entomol. 31: 564-572.

Braman, S. K., A. F. Pendley, R. N. Carrow and M. C. Engelke. 1994. Potential resistance in zoysiagrasses to tawny mole crickets (Orthoptera, Gryllotalpidae). Fla. Entomol. 77: 301-305.

Braman, S. K., R. R. Duncan, W. W. Hanna and W. G. Hudson. 2000b. Evaluation of turfgrasses for resistance to mole crickets (Orthoptera: Gryllotalpidae). HortScience 35: 665-668.

Braman, S. K., R. R. Duncan, W. W. Hanna and C. M. Engelke. 2003. Arthropod predator occurrence and performance of *Geocoris uliginosus* (Say) on pest-resistant and susceptible turfgrasses. Environ. Entomol. 32: 907-914.

Braman, S. K., R. R. Duncan, W. W. Hanna and M. C. Engelke. 2004. Turfgrass species and cultivar influences on survival and parasitism of fall armyworm. J. Econ. Entomol. 97: 1993-1998.

Braman, S. K., J. G. Latimer, R. D. Oetting, R. D. McQueen, T. B. Eckberg and M. Prinster. 2000c. Management strategy, shade, and landscape composition effects on urban landscape plant quality and arthropod abundance. J. Econ. Entomol. 93: 1464-1472.

Braman, S. K., R. R. Duncan, M. C. Engelke, W. W. Hanna, K. Hignight and D. Rush. 2002b. Grass species and endophyte effects on survival and development of fall armyworm (Lepidoptera: Noctuidae). J. Econ. Entomol. 95: 487-492.

Byers, R. A. 1965. Biology and control of a spittlebug, *Prosapia bicincta* (Say), on Coastal Bermudagrass. Tech. Bull. NS 42 Georgia Agric. Exp. Stn. 26 pp.

Cardona, C., J. W. Miles and G. Sotelo. 1999. An improved methodology for massive screening of *Brachiaria* spp. genotypes for resistance to *Aeneolamia varia* (Homoptera: Cercopidae). J. Econ. Entomol. 92: 490-496.

Cardona, C., P. Fory, G. Sotelo, A. Pabon, G. Diaz and J. W. Miles. 2004. Antibiosis and tolerance to five species of spittlebug (Homoptera: Cercopidae) in *Brachiaria* spp.: Implications for breeding for resistance. J. Econ. Entomol. 97: 635-645.

Fagan, E. B. and L. C. Kuitert. 1969. Biology of the two-lined spittlebug, *Prosapia bicincta*, on Florida pastures (Homoptera: Cercopidae). Fla. Entomol. 52: 199-206.

Genung, W. G. and V. E. Green. 1974. Food habits of the meadowlark in the Everglades in relation to agriculture. Environ. Entomol. 3: 39-42.

Hamilton, K. G. A. 1977. Review of the world species of *Prosapia* Fennah (Rhynchota: Homoptera: Cercopidae). Can. Entomol. 109: 621-630.

Hanna, W. W. 1995. Centipedegrass - diversity and vulnerability. Crop Sci. 35: 332-334.

LaPointe, S. L., M. S. Serrano, G. L. Arango, G. Sotelo and F. Córdoba. 1992. Antibiosis to spittlebugs (Homoptera: Cercopidae) in accessions of *Brachiaria* spp. J. Econ. Entomol. 85: 1485-1490.

Liu, J., W. Hanna, E. Elsner and J. Zhou. 2002. Germplasm investigation of centipedegrass in the People's Republic of China. Plant Genet. Resour. Newsl. 132: 1-4.

Miles, J. W., S. L. LaPointe, M. L. Escandon and G. Sotelo. 1995. Inheritance of resistance to spittle bug (Homoptera: Cercopidae) in interspecific *Brachiaria* spp. hybrids. J. Econ. Entomol. 88: 1477-1481.

Potter, D. A. and S. K. Braman. 1991. Ecology and management of turfgrass insects. Annu. Rev. Entomol. 36: 383-406.

SAS Institute. 2003. SAS System, version 9.3 for Windows. SAS Institute, Cary, NC.

Shortman, S. L., S. K. Braman, R. R. Duncan, W. W. Hanna and M. C. Engelke. 2002. Evaluation of turfgrass species and cultivars for potential resistance to twolined spittlebug (Hemiptera: Cercopidae). J. Econ. Entomol. 95: 478-486.

Stimmann, M. W. and C. M. Taliaferro. 1969. Resistance of selected accessions of Bermudagrass to phytotoxemia caused by adult two-lined spittle bugs. J. Econ. Entomol. 62: 1189-1190.

Taliaferro, C. M., D. B. Leuck and M. W. Stimmann. 1973. Tolerance of *Cynodon* clones to phytotoxemia caused by the two-lined spittlebug. Crop Sci. 9: 765-766.