Plant Resistance

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Warm-Season (C4) Turfgrass Genotypes Resistant to Spittlebugs (Hemiptera: Cercopidae)

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Abstract

Screening for resistance to insect pests is one of the early stages of grass breeding programs. Pasture spittlebugs are sap-sucking insects that potentially cause severe damage to turfgrasses, including the loss of functional quality and perenniallity. The Brazilian flora has a large number of grass species with wide morphological variability and adaptability to different soil and climate conditions that can potentially be used as lawns. However, no study has screened turfgrass genotypes for resistance to spittlebug attack. In this study, we evaluated the intra- and interspecific variability of 35 turfgrass genotypes in the genera Paspalum, Axonopus, and Zoysia for resistance to the pasture spittlebugs, Deois flavopicta (Stal) and Notozulia entreriana (Berg) (Hemiptera: Cercopidae), as measured by damage scores, densities of nymphs and adults, and level of antibiosis resistance. Genotypes were grouped into three groups using cluster analysis and principal component analysis: GroupI had genotypes associated with low damage scores and high density of adult spittlebugs; GroupII had genotypes with intermediate damage scores and low density of nymphs and adults; and GroupIII was formed by genotypes with high damage scores and high nymph density. Intra- and interspecific genotypic variability was related to antibiosis resistance and morphological variation among genotypes with some indicating nonpreference resistance and others indicating tolerance resistance. Our results indicate that besides antibiosis resistance studies, it is essential to evaluate the morphological variability of grass genotypes when screening for resistance to insects. Further studies are needed to elucidate the intraspecific variability of Paspalum notatum Flüggé genotypes for resistance to spittlebug attack.

Key words: froghopper, germplasm resource, insect plant resistance, lawn, Paspalum

The expansion of urban green areas occupied by parks and gardens runs parallel to the social and economic development of communities. Thus, turfgrass lawns are major components of urban landscapes. In addition to the recreational and aesthetic benefits, turfgrass lawns play important environmental roles, including heat dissipation and temperature moderation, soil erosion control, carbon sequestration, and air pollution control (Beard and Green 1994, Alumai et al. 2009). Other benefits of turfgrasses include an overall better quality of life, enhanced physical health, and improved mental health, especially in densely populated urban areas (Beard and Green 1994, Grahn and Stigsdotter 2003).

The genus *Paspalum* L. (Poaceae) includes species and varieties that exhibit wide morphological variability and adaptability to different soil and climate conditions (Dos-Reis et al. 2010, Meirelles et al. 2013). For instance, *Paspalum vaginatum* Sw. is largely used on golf courses and athletic fields in North America (Brosnan and

Deputy 2009) because of its traffic tolerance and tolerance to variable soil pH and salinity. In Brazil, *Paspalum notatum* Flüggé, known commonly as bahiagrass or grama-batatais, is widely used for landscaping (Freitas 2002) because of its rusticity, rapid growth, water stress tolerance, low fertility requirements, and tolerance to traffic (Alencar 1949, Kissmann and Groth 1997).

Emerald grass (Zoysia japonica Steud) is native to Asia and has been widely used in Brazil as an ornamental plant in public and residential gardens and other recreational areas (Kojoroski-Silva et al. 2011), but it is also susceptible to spittlebug (Shortman et al. 2002). The genus Axonopus includes ~110 species distributed in tropical and subtropical regions, several of which are used as lawns (Hickenbick et al. 1975). Specifically, the carpetgrass Axonopus fissifolius (Raddi) Kuhlm. (syn. Axonopus affinis Chase), also known in Brazil as grama-São Carlos, is largely used in roadsides and as low-maintenance lawns in the southern United States, Australia, The use of grasses for landscaping purposes requires the screening for resistance to insect pests to ensure the long-term functional quality of lawns and to prevent the spread of pests into adjacent cultivated areas. Specifically, special attention should be given to the susceptibility of grass species to pasture spittlebugs (Hemiptera: Cercopidae), which are key pests of forage grasses in South America (Valério and Nakano 1988).

Spittlebugs are considered the most damaging pasture pests in Brazil (Gusmão et al. 2013a). Spittlebug nymphs suck sap from roots and excrete large amounts of liquid, forming a frothy, spittlelike mass that surrounds the young nymphs. The saliva of the sapsucking adults induces a phytotoxemia that causes the leaf blade and stem to yellow and die. Typically the plant reactions to the spittlebug attack are initially chlorotic spots and or stripes, progressing to a more severe damage, prevailing stripes or necrotic bands. In general, the leaves attacked by spittlebug die from the tips, with subsequently a twisted appearance (Valério and Nakano 1992). The main losses to lawns caused by spittlebugs include the loss of functional quality and perenniallity, whereas in forage grasses these include reduced biomass production, loss of palatability, and reduced carrying capacity of pastures (Valério and Nakano 1988).

Deois flavopicta Stal and Notozulia entreriana Berg (Hemiptera: Cercopidae) are two of the most important spittlebug species causing damage to grasses in Southeast and West Central Brazil. The population dynamics of the species is determined by climatic factors (temperature and relative humidity), which directly affect their development, especially the embryonic development of eggs, which remain in diapause during autumn and winter, a period characterized by low temperature and reduced rainfall (Pires et al. 2000).

Genetic resistance is the main method for spittlebug management, and the use of plants resistant to spittlebugs is an easily adopted and environmentally safe method compared to other management strategies such as chemical control (Valério et al. 1997, Auad et al. 2007). There are a lot of studies about genetic resistance involving spittlebugs on grass genotypes for pasture (Lapointe et al. 1992, Valério et al. 1997, López et al. 2009, Cardona et al. 2010). However, no studies have been done with turfgrass genotypes.

This study evaluated the intra- and interspecific genotypic variability of native turfgrasses of Brazil for resistance to attack by the spittlebug *D. flavopicta* and its antibiosis resistance levels to *N. entreriana*.

Materials and Methods

Trial 1. Resistance to Attack by the Spittlebug *D. flavopicta*

This first trial was conduced at Embrapa Southeast Livestock Station in São Carlos, SP, Brazil (21° 96'17" S, 47° 84'21" W, 856 m elevation) from January 2011 to July 2013 in field condictions.

Experimental Conditions

The soil in the experimental area had the following properties (0-20 cm profile): pH: 5.6; organic matter: 28 g/dm^3 ; resin P: 8.0 mg/dm³; K: 0.9 mmol/dm³; Ca: 20.0 mmol/dm³; Mg: 9.0 mmol/dm³; CEC: 61.0 mmol/dm³, base saturation: (V%) 49.0%; and 445.0, 79.0, and 476.0 g/kg of sand:silt:clay, respectively. Before seeding, the area was tilled, harrowed, and limed to raise the base saturation (V%) to 60.0%.

Plugs of turfgrass genotypes were planted 15.0 cm apart in 3- by 4-m plots in January 2011 and irrigated by sprinklers. Thirty days after planting, granular fertilizer (10N–10P–10K) was applied by hand at a rate of 100 kg/ha, and plots were irrigated weekly (10 mm) by sprinklers, except in rainy weeks. Plots were mowed to a 5-cm standard height when plot cover reached 100%. Throughout the study, $\frac{2}{3}$ of each plot was mowed to a height of 5.0 cm when plant height exceeded 7.5 cm, whereas the remaining $\frac{1}{3}$ was left unmowed.

Insect Infestation

Plots were naturally infested by a natural population of *D. flavopicta* from an adjacent infested field of *Urochloa decumbens* (Stapf) R.D. Webster, which is susceptible to spittlebug infestation.

Experimental Design and Sampling of Insects

Plots were arranged in a randomized complete block design with four replications of each genotype. In total, 35 genotypes were evaluated: 32 Paspalum spp. genotypes, two Axonopus spp. genotypes, and one positive control genotype (Emerald grass, Zoysia japonica Steud.) as function of its insect resistance (Table 1). Genotypes were provided by the Paspalum Germplasm Bank (BAG) at Embrapa Southeast Livestock Station in São Carlos, SP, and were selected for their small size and decumbent growth. The dependent variables were the number of D. flavopicta adults, number of spittle masses produced by nymphs, and the level of damage. The entire area of the plots was scored for damage levels using the 1-5 visual scale developed by Cardona et al. (1999): D₀-no visible damage (note 1); D₁₋₂₅-up to 25% of leaf area affected (note 2); D₂₆₋₅₀-from 26 to 50% of leaf area affected (note 3); D₅₁₋₇₅-from 51 to 75% of leaf area affected (note 4); and D₇₆₋₁₀₀—>76% of leaf area affected (note 5). Sampling was done monthly and the population density of D. flavopicta was estimated by counting the number of adults and nymphs. The sampling of adults was carried out using a sweep net (37-cm-diameter rim, 70cm-deep bag, and 120-cm-long handle) throughout the entire plot area (six sweeps per plot, starting from the boundary) due to insect dispersal ability. Nymphs were sampled in the mowed portion of plots by counting the number of spittle masses in one subsample of 0.25 m² area using a 50-cm metal quadrat.

Data Analysis

Data on proportion of damage level (D₀, D₁₋₂₅, D₂₆₋₅₀, D₅₁₋₇₅, and D₇₆₋₁₀₀), number of adults (Adult), and number of spittle masses (Nymph) for the 35 genotypes were standardized to $Z = (x - \mu)/\sigma$ and analyzed using two multivariate statistical techniques, cluster analysis (CA) and principal component analysis (PCA).

The K-means method of cluster analysis was used to form *K* clusters of genotypes according to level of damage and insect density. After *K* clusters were formed, a multivariate analysis of variance (MANOVA) followed by the *F*-test (K = 35; P < 0.01) was used to test the null hypothesis (H_0 : equality of groups). When H_0 was rejected, the Hotelling's T² test (P < 0.05) was applied to determine the difference between the mean vectors (centroids), two by two, of the *K* clusters formed.

PCA was used to complement CA and to illustrate the relationship between dependent variables and the genotype clusters formed. PCA was used to reduce the set of seven dependent variables (five damage levels, number of adults, and number of spittle masses) to two orthogonal principle components (PC), which are linear combinations of the original variables that correspond to the two largest eigenvalues extracted from the correlation matrix (Hair et al. 2005). All analysis was performed using Statistica, version 7.0, software (StatSoft, Tulsa, OK).

Table 1.	Description	of turforass	aenotypes	according	to stem type

Genotype	BRA Code	Specie name	Stem type ^{<i>a,b</i>}	Evaluated trial
G1	6173	Paspalum notatum Flüggé	Rhizome	1 and 2
G2	6203	Paspalum modestum	Stolons	1 and 2
G3	6301	Paspalum notatum Flüggé	Rhizome	1 and 2
G4	6378	P. denticulatum Trin.	Stolons-Rhizome, short internodes	1 and 2
G5	6467	Paspalum notatum Flüggé	Rhizome	1 and 2
G6	6491	Paspalum modestum Mez	Stolons	1 and 2
G7	6513	Paspalum notatum Flüggé	Rhizome	1
G8	7510	P. conjugatum P.J. Bergius	Stolons	1
G9	8681	Paspalum lepton Schult	Rhizome with long, thin internodes	1
G10	9989	Paspalum vaginatum Sw.	Stolons	1 and 2
G11	10006	Paspalum notatum Flüggé	Rhizome	1 and 2
G12	12254	Paspalum notatum Flüggé	Rhizome	1 and 2
G13	14842	P. subciliatum Chase	Stolons	1
G14	19178	Paspalum notatum Flüggé	Rhizome	1 and 2
G15	19470	Paspalum notatum Flüggé	Rhizome	1
G16	20648	Paspalum jesuiticum Parodi	Stolons	1
G17	54456-9	Paspalum dilatatum Poir.	Rhizome with short internodes	1
G18	21016	Paspalum indecorum Mez	Rhizome-Stolons, short internodes	1
G19	21547	Paspalum notatum Flüggé	Rhizome	1 and 2
G20	21563	Paspalum notatum Flüggé	Rhizome	1
G21	22594	Paspalum notatum Flüggé	Rhizome	1
G22	23523	Paspalum notatum Flüggé	Rhizome	1 and 2
G23	23558	Paspalum notatum Flüggé	Rhizome	1 and 2
G24	23566	Paspalum notatum Flüggé	Rhizome	1 and 2
G25	23591	Paspalum lepton Schult	Rhizome with long, thin internodes	1 and 2
G26	23728	Paspalum notatum Flüggé	Rhizome	1 and 2
G27	24236	Paspalum notatum Flüggé	Rhizome	1
G28	2658	Axonopus sp.	Stolons with rhizome	1
G29	25046	Paspalum notatum Flüggé	Rhizome	1
G30	25054	Paspalum lepton Schult.	Clumping with thin rhizomes	1
G31	25097	Paspalum lepton Schult.	Clumping with thin rhizomes	1
G32	54634-1	Paspalum lepton Schult.	Clumping with thin rhizomes	1
G33	Control	Zoysia japonica Steud.	Stolons with rhizome	1
G34	2691	A. fissifolius (Raddi) Kuhlm.	Stolons with rhizome	1
G35	25216	Paspalum notatum Flüggé	Rhizome	1

According to ^a Zuloaga and Morrone (2005) and ^b Kojoroski-Silva et al. (2011).

Trial 2. Antibiosis Resistance Levels to the Spittlebug *N. entreriana*

This second trial was conduced at Embrapa Beef Cattle Station in Campo Grande, MS, Brazil (20° 26'40.31″ S, 54° 43'20.4″ W, 531 m elevation) from March 2011 to May 2011 in greenhouse conditions. The antibioses resistance levels were evaluated for 16 turf-grasses genotypes, from the 35 evaluated in the first trial (Table 1).

Antibiosis Resistance Evaluation and Experimental Conditions

The antibiosis resistance levels were assessed by survival and duration of nymphal period parameters according to Lapointe et al. (1989). The experimental unit consisted on a plastic pot with a capacity of 2.5 kg of soil containing one plant. This plant was initially propagated by plugs of turfgrass genotypes in small plastic cups, one month before the start of the trial. After establishment, the seedlings were transferred to pots, which were capped with an aluminum lid, leaving a central opening for the exit of aerial plant portion. This procedure aimed firstly the stimulation of shallow roots emission, which are important to ensure the survival of newly hatched nymphs and secondly, to provide low aeration and luminosity, and high humidity environment to the nymphs.

The plants infestation was done one month after seedling establishment, using five spittlebug eggs of *N. entreriana* per pot and 10 repetitions for each turfgrass genotype. Close to the adult emergence, each pot was covered with a screened cage and the adults of spittlebug were collected daily as they emerged.

Data Analysis

The survival percentages of nymph and the nymphal time period of *N. entreriana* were calculated for the 16 turfgrasses genotypes, for which the same cluster correspondence (GI, GII, and GIII) obtained in the first trial were attributed. The data were submitted to variance analysis related to group effect using the GLM procedure of SAS, version 8.0 (SAS Institute, Inc., Cary, NC). The nymph survival percentages data were transformed to arcsine-square root (x/100), and the means separation were performed by pairwise comparisons of means by the t test of the least squares means (LSMeans) option of GLM, at 5% level of significance, using version 8.0 of SAS statistical software.

Results

Trial 1. Resistance Against Attack by the Spittlebug *D. flavopicta*

Due to the effect of climatic factors on insect population fluctuations, there was a high coefficient of variation in monthly data for insect attack variables (number of adults, number of spittle masses, and damage level). Thus, to reduce this variation, the mean values for each season was considered: autumn (April–June), winter (July–September), spring (October–December), and summer (January–March). Spittle masses, adult densities, and damage levels varied significantly during the study period (Fig. 1). The highest mean values were observed in the first year, followed by a strong reduction in subsequent periods.

The mean temperature and relative humidity (RH) data for the study site were within normal ranges for each season, with higher average temperatures in spring and summer and lower average temperatures in autumn and winter (Fig. 2). Even though the population density of *D. flavopicta* declined over the study period, cercopid attack preference was significantly different across the turfgrass genotypes: the highest damage scores were observed in the G₂ and G₆ genotypes of *P. modestum*, whereas the smallest number of insects and the lowest damage scores were observed in *Z. japonica* (G₃₃).

Cluster analysis grouped the 35 genotypes into three clusters, which were significantly different by the Hotelling's T^2 test (P < 0.05) and were defined as follows: GroupI (n = 14), low damage scores and high density of adults; GroupII (n = 12), intermediate damage scores and low densities of spittle masses and adults; and GroupIII (n = 9), high damage scores and high spittle masses density (Table 2).

PCA illustrated the relationship between the dependent variables and the genotype clusters. The projection of the seven insect attack variables and 35 turfgrass genotypes formed by the first and second principal components (PC₁ and PC₂) accounted for 69.33% of the total variance (Fig. 3).

The PC₁ was the axis of greatest contribution to formation of the plane (45.42% of the total variance). After the intermediate damage scores (D₂₆₋₅₀) variable, the number of *D. flavopicta* adults had the lower contribution to the formation of groups in the PC₁ axis (2%). On the other hand, the damage scores of D₀, D₁₋₂₅, D₅₁₋₇₅, and D₇₆₋₁₀₀ had high contribution to the formation of groups in the PC₁ axis with 15, 26, 25, and 24%, respectively. The spittle masses variable accounted for 6% of the PC₁ axis contribution. The three genotype groups formed by cluster analysis are also situated in the same plane. Additionally, the horizontal axis (PC₁) represents the increasing damage scores of genotype clusters (GroupI < GroupII < GroupII). GroupI, represented by genotypes with low damage scores (D₀ and D₁₋₂₅), consists of 11

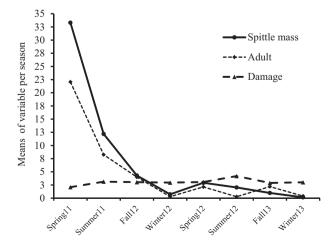


Fig. 1. Mean number of spittle masses, adults, and damage score of *D. flavo*picta over seasonal periods.

P. notatum genotypes, the *Axonopus fissifolius* genotype, the *Axonopus* sp. genotype, and the *Z. japonica* genotype. GroupII, represented by genotypes with intermediate damage scores (D_{26-50}), consists of seven *P. notatum* genotypes and one genotype each of *P. conjugatum*, *P. subciliatum*, *P. vaginatum*, *P. jesuiticum*, and *P. dilatum*. In addition, GroupIII, represented by genotypes with high damage scores (D_{51-75} and D_{76-100}) and high nymph density, is composed of all *P. lepton*, *P. modestum*, and *P. denticulatum* genotypes and one *P. indecorum* genotype. The absence of *P. notatum* genotypes in GroupIII is noteworthy.

Trial 2. Levels of Antibiosis Resistance Against the Spittlebug *N. entreriana*

Differences were observed between *Paspalum* genotypes in nymph survival and duration of nymphal period parameters (Table 3). The nymph survival ranged from 6.0 to 94.0% and the nymphal period lasted from 29.0 to 40.8 d. These results indicate resistance by antibiosis, considering that the genotypes with the lowest percentages of nymph survival were those with the longest nymphal periods,

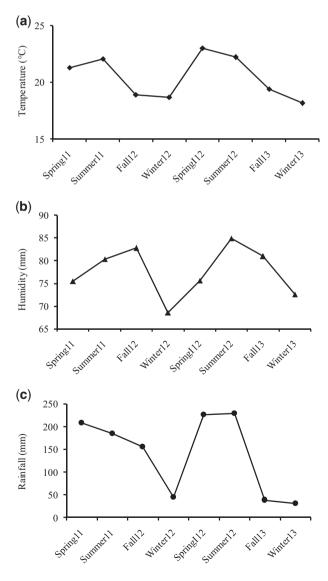


Fig. 2. Average temperature (a), average relative humidity (b), and rainfall (c) over the seasonal period from 2011 to 2013.

as verified for G₂₂ genotype, *P. notatum*, whose parameters were 6.0% and 40.8 d, respectively (Table 3). The grouping of genotypes according to the clustering realized in the first trial has shown that the less-attacked genotypes corresponded to those with higher levels of resistance by antibiosis in the second trial. From the 16 genotypes evaluated, the first seven with higher levels of antibioses (nymph survival equal or lower than 18% and nymphal period longer than 34 d) were clustered in GroupI. On the other hand, three of the four most susceptible genotypes (nymph survival equal or superior to 68% and nymphal period shorter than 29.5 d) were clustered in GroupIII. Five genotypes presented intermediate values of nymph survival (from 20 to 62%) and nymphal period (from 30 to 35 d), which were clustered in groups I and II (Table 3).

Significant differences between genotype groups in considering the antibiosis parameters are shown in Fig. 4. Nymph survival rate and nymphal period were different between groups I and III. These groups were formed by the most dissimilar genotypes in the first experiment.

Discussion

Deois flavopicta populations have been studied in areas near the study site since 2008. In the study area in particular, and in southeast Brazil in general, the species occurs at moderate population densities compared to regions with higher average temperatures Considering the regularity in mean temperature and RH in the field trial, it may be concluded that the reduction in the population density of *D. flavopicta* after the first year was not caused by climatic conditions and may be related to the increased resistance to spittlebug attack provided by the higher proportion of lignified stolons, rhizomes, and tillers in the grasses, and probably due to antibiosis resistance of *Paspalum* genotypes, as the resistance against *N. entreriana* verified in the second trial. Being native species, genotypes in the genus *Paspalum* generally have low susceptibility to pasture spittlebugs (Oliveira and Alves 1988, Da Costa et al. 2006, Gusmão et al. 2013b). Moreover, the increase in its natural enemy populations, including microhymenopteran egg parasitoids (Valério and Oliveira 2005) and nymph and adult predators (Sujii et al. 2002), may have contributed to the reduction in spittlebug numbers. Unfortunately, these data were not collected in this study.

Although only 45% of the evaluated genotypes were common to both trials, there was great similarity between levels of insect attack of the genotype groups formed in the first trial and the levels of antibiosis presented by genotypes in the second trial, especially considering groups I and III. Antibiosis resistance to *D. flavopicta* has been identified in *Paspalum* spp. accessions by Gusmão et al. (2013b). Evaluating 12 genotypes from different species, those researchers indicated that eight had antibiosis resistance similar to that found in

Table 2. Grouping of 35 genotypes by cluster analysis followed by Manova and Hotelling's T^2 test

Group	Ν	Selected genotypes	Characteristics	
Ι	14	G ₁ , G ₁₉ , G ₃ , G ₂₇ , G ₅ , G ₂₈ , G ₁₂ , G ₂₃ , G ₁₄ , G ₂₄ , G ₂₂ , G ₃₃ (control), G ₃₄ , and G ₃₅	Low damage scores and high density of adult	
II III		G ₂₆ , G ₂₁ , G ₁₅ , G ₁₆ , G ₁₀ , G ₁₇ , G ₈ , G ₂₉ , G ₁₁ , G ₁₃ , G ₇ , and G ₂₀ G ₂ , G ₆ , G ₄ , G ₃₀ , G ₃₁ , G ₉ , G ₂₅ , G ₃₂ , and G ₁₈	Intermediate damage scores and low density of spittle masses and adults High damage scores and high spittle masses density	

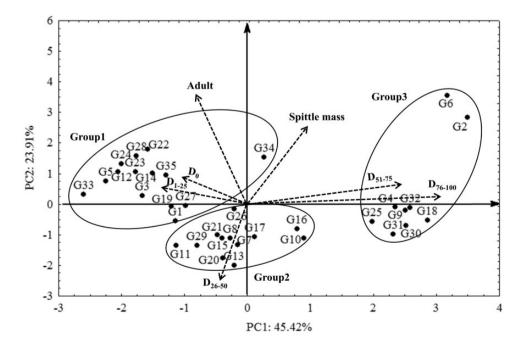


Fig. 3. Principal component analysis (PC₁ and PC₂) with 35 genotypes of grass and seven insect attack variables (Adult; Spittle masses; D_0 : no damage; D_{1-26} : damage from 1 to 25%; D_{26-50} : damage from 26 to 50%; D_{51-75} : damage from 51 to 75%, and D_{76-100} : damage from 76 to 100%) and the representation of the three generated groups (GroupI, GroupII, and GroupIII).

Table 3. Nymph survival and duration of nymphal period for *N. entreriana* in function of 16 *Paspalum* sp. genotypes grouped according to first trial

Genotype	Nymph survival (%)	Nymphal time (d)	Group
G22	6	40.8	Ι
G24	8	42.2	Ι
G1	10	43.2	Ι
G14	10	43.6	Ι
G12	12	41.5	Ι
G26	14	48.6	Ι
G23	18	34.1	Ι
G4	26	35.1	II
G19	34	34.3	Ι
G5	42	37.3	Ι
G11	44	36.8	II
G3	62	36.6	Ι
G25	68	29.6	III
G6	80	26.6	III
G2	94	28.6	III
G10	94	29.0	II

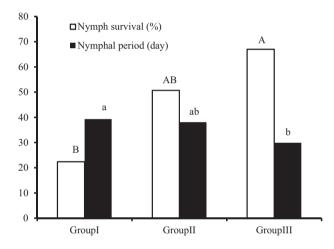


Fig. 4. Comparison between means of nymph survival and duration of nymphal period of *N. entreriana* in 16 genotypes of *Paspalum* sp. grouped according to the first trial. Means followed by different letters, capital (nymph survival) and lowercase (nymphal period) are significantly different by t test at P < 0.05.

Urochloa brizantha cv. Marandu, whereas the survival of nymphs was lower than the susceptibility pattern of *U. decumbens* cv. Basilisk for all genotypes investigated. Moreover, Oliveira and Alves (1988) reported that *Paspalum secans*, *P. coryphaeum* and *P. guenoarum* were more resistant to the cercopids *Deois incompleta* and *D. flavopicta* than exotic grasses from the genera *Andropogon*, *Brachiaria* (Urochloa), *Panicum*, and *Setaria*. Similar tolerance to the cercopid *N. entreriana* has been identified in four genotypes of *Paspalum* spp. and *U. brizantha* cv. Marandu, the latter of which has previously been identified as resistant to the insect (Da Costa et al. 2006).

Similar resistance of Z. *japonica* (G_{33}) to the attack of D. *flavopicta* was verified to *Prosapia bicincta* Say in the study of Shortman et al. (2002). It was mentioned that, even though Z. *japonica* was not susceptible to *P. bicincta*, this spittlebug species may potentially survive and cause significant damage to the plants. According to Valério and Nakano (1988), high damage levels are associated with the occurrence of adults. However, in this study the highest damage

score was unrelated to the greatest number of adults. The genotypes of GroupI were associated not only with low damage scores, but also with high density of adults. This contrary result may be explained by the dispersal ability of adults, and not necessarily by the greater attractiveness of these genotypes to adult spittlebugs, and is supported by the low association of GroupI genotypes with high damage scores and nymph density. In fact, *D. flavopicta* adults move mainly by short, low jumping flights, and females live within a 50–100 m radius from their emergence point (Sujii et al., 2000).

Paspalum notatum genotypes were associated with low damage scores and can be considered resistant to spittlebug attack. The presence of some *P. notatum* genotypes in GroupII may be explained by its phenotypic plasticity or intraspecific genetic variability (Cidade et al. 2013). Thus, the difference in damage scores between grass genotype groups is more evident between genotypes in GroupI and GroupIII. Besides the low level of antibiosis resistance, as verified to *N. entreriana*, the genotypes in the species *P. modestum*, *P. denticulatum*, and *P. lepton* were associated with high damage scores from *D. flavopicta* attack, possibly due to their morphological characteristics, and are rated susceptible. The *A. fissifolius* genotype (G₃₄, GroupI) was associated with high numbers of nymphs and adults and low damage scores and was rated tolerant to *D. flavopicta* attack.

The differences in damage scores among P. notatum, P. modestum, P. denticulatum, and A. fissifolius genotypes may be related to the type of stem and growth habit of each species, in addition to differences in antibiosis resistance levels. According to Coradin et al. (2011), P. notatum has horizontal, aboveground rhizomes that are pressed firmly to the ground and have short internodes, flat, folded, in-rolled leaves (Fig. 5a), which may be related to nonpreference resistance to spittlebug attack. Conversely, P. modestum (G2 and G6) plants have a prostrate, stoloniferous growth habit and superficial adventitious roots, glabrous, tender leaves and internodes (Fig. 5b and 5c). This genotypes were more heavily attacked by D. flavopicta, had high damage scores (first trial), presented high percentage of nymph survival, and short nymphal period of N. entreriana (second trial). Similar morphological characteristics are found in P. denticulatum (G₄) (Fig. 5d), another genotype with high damage scores, which is characterized by stoloniferous or rhizomatous stems, creeping or semicreeping cespitose habit, suberect, decumbent culms, open, compressed, and generally glabrous leaf sheaths, tender leaf blades, and membranous ligule (Coradin et al. 2011).

The tolerance of the *A. fissifolius* genotype (G_{34}) may be explained by the high levels of reserve carbohydrates stored in the stem and roots that are allocated aboveground to developing parts, and because this species can easily translocate reserve carbohydrates stored in roots to stems and damaged plant parts. In fact, *A. fissifolius* exhibited greater dry matter accumulation in roots, which may be a strong sink of photoassimilates, as well as greater leaf size, stolon thickness, and root/shoot ratio than Emerald (*Zoysia* sp.) and Tifton 419 (*Cynodon dactylon* x *C. transvaalensis*) grasses (Kojoroski-Silva et al. 2011). Fixed carbohydrate and nutrient reserves have long been considered important resources for regrowth after damage and may be responsible for the tolerance of plants to insect herbivore attack (Rosenthal and Kotanen 1994).

The results of this study support the findings of Ferrufino and Lapointe (1989), who reported that grass species that have unprotected, superficial adventitious roots and glabrous, tender culms and leaves are more heavily attacked by spittlebug nymphs and adults. Thus, as important as assessing the level of antibiosis is the evaluation of morphological variability of grass genotypes in breeding programs, especially considering the easiness and convenience of such comparisons. Nevertheless, because *P. notatum* genotypes exhibit



Fig. 5. Morphological characteristics of leaf sheaths, stems, and roots of lower insect attack grass genotypes—P. notatum (a) and largest insect attack—P. modestum (b and c) and P. denticulatum (d).

little morphological variation, further studies are needed to elucidate their intraspecific variability in the resistance against spittlebug attack.

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