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## **PLANT PROTECTION - Article**

# **Evaluation of forage grass resistance to Mahanarva fimbriolata (Stål)**

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**ABSTRACT:** The aim of this study was to select forage grasses with potential resistance to *Mahanarva fimbriolata* by assessing effects on insect. No-choice assays were conducted with 12 genotypes and laboratory-reared insects. The following parameters were assessed: mortality and duration of the nymphal stage; adult weight and longevity; pre-oviposition period; number of eggs/female; viability and duration of the embryonic period. As main result it was verified that the mortality of nymphs reared on *Panicum maximum* cv. Paredão is almost complete and that this forage grass may be

characterized as resistant to this insect by antibiosis. Further, the grasses can be divided into three groups following a cluster analysis: *P. maximum* cv. Aries, *Setaria sphacelata* cv. Kazungula, *Brachiaria humidicola*, *A. gayanus* and *P. maximum* cv. Aruana, which are insect-resistant grasses; *Brachiaria decumbens* cv. Basilisk, *B. dictyoneura*, *B. brizantha* cv. MG-4 and *B. ruziziensis*, which are moderately resistant grasses; and *B. brizantha* MG-5 and *B. brizantha* ecotype BB185, which are susceptible grasses. **Key words:** spittlebug, antibiosis, *Brachiaria*, host plant resistance.

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#### INTRODUCTION

The spittlebug *Mahanarva fimbriolata* (Stål) (Hemiptera: Cercopidae) is currently a major pest of mechanically harvested sugar cane, i.e., which is harvested without burning (Dinardo-Miranda 1999; Dinardo-Miranda et al. 2001; Dinardo-Miranda et al. 2004; Ravaneli et al. 2011). This species has also caused significant damage in forage grasses used for pasture establishment, thereby limiting the potential cattle production in Brazil. This damage is of concern to livestock producers, particularly those from the Brazilian Midwest and Northern regions (Valério 2006).

The damage caused to pastures by *M. fimbriolata* has not yet been estimated. It is difficult to calculate the damage caused by pests in this type of ecosystem because there is no direct effect on the final product (meat and milk); rather, the effect is on cattle feed (Pottinger 1976). One of the few studies of this effect was conducted by Holmann and Peck (2002), who, using a simulation model and analyzing two different ecosystems (dry and humid tropics) in Colombia, found that spittlebugs caused economic losses of US\$ 228-273 million/year and 39-47 million/year in the dry and humid tropics, respectively.

Due to the great damage caused by spittlebugs, there is a need for monitoring and control as necessary. The control of spittlebugs can be accomplished by several methods, all of which have advantages and disadvantages. Chemical control is considered to be economically unviable, as pasture is considered a low value crop per unit area; further, insecticides present problems of waste, and it is difficult to control the nymphs due to the presence of the foam they produce when feeding (Ferrufino and Lapointe, 1989; Valério 2005). Thus, it is preferable to use other control methods.

Among the alternative methods is biological control with *Metarhizium anisopliae* (Metsch.) Sorok. This species has been widely used for controlling Cercopidae in sugar cane from Northern to Southeastern Brazil, covering a treated area of approximately 357,058 ha (Alves et al. 2008). Another option is the use of resistant plants, which stands out because it is the most appropriate tool for these situations from an economic and environmental perspective and can be used with biological and cultural controls, thereby contributing to the integrated management of this pest (Nilakhe 1987; Vendramim 1990; Lapointe and

Sonoda, 2001; Valério 2005). Several studies are being developed toward the goal of selecting a grass that is resistant to both the nymphs and adults of spittlebugs (Ferrufino and Lapointe 1989; Valério et al. 1997; Cardona et al. 2004; López et al. 2009; Cardona et al. 2010).

The aim of this study was to select forage grasses with potential resistance to *M. fimbriolata*.

#### MATERIAL AND METHODS

Rearing of M. fimbriolata started in February 2011 after collection in the field, and the spittlebugs were maintained in the laboratory according to the technique described by Garcia et al. (2006), with some modifications: after infestation with newly hatched nymphs, the plants were packed in PVC tubes (10 cm diameter × 25 cm height) placed in a phytotron (at a temperature of 24 ± 6 °C, relative humidity of  $70 \pm 10\%$  and a photoperiod of 12 h). The nymphs remained in the phytotron until the emergence of the adults, and the plants were replaced when they showed yellowing of the leaves or loss of roots. After emergence, the adults were kept in cylindrical cages (made with pet bottles) to obtain eggs. These cages had two lateral and opposing openings ( $10 \text{ cm} \times 5 \text{ cm}$ ) for air exchange, two hooks made with wire near the base for the placement of elastics, which attached the cages to the Petri dish. A sugarcane plant was placed inside the cages to serve as a feed substrate. At the base of the plants, covering the soil, cotton pads were placed to serve as oviposition substrate. After oviposition, the eggs were removed from the cotton with a brush (n° 000) and kept in Petri dishes, lined with moist filter paper until the hatching of the nymphs.

A no-choice assay was performed with newly hatched nymphs from the stock colony. The grasses were sown in 500 mL pots using Basaplant substrate and fertilizer [Osmocote - 14-14-14 (N, P, K)] applied at 200 g for each 25 kg of substrate. Three-month-old plants were removed from the plastic container and re-planted in pots (15 cm high, 17.5 cm opening and 12 cm base), so that the roots remained partially exposed. After additional root development, 10 nymphs from the rearing colony were transferred to each plant with the aid of a brush. The experiment was replicated 10 times with each grass variety (treatment), resulting in a total of 100 nymphs per treatment. We tested the performance of *M. fimbriolata* 

on the following grasses: Brachiaria dictyoneura (Fig. & Not.) Stapf., B. humidicola (Rendle) Schweick cv. Llanero, B. ruziziensis (Germain & Edvard), B. decumbens Stapf. cv. Basilisk, B. brizantha (Hochstex A. Rich.) Stapf. cv. MG-4, B. brizantha (Hochst ex A. Rich.) Stapf. cv. MG-5 Vitória, Panicum maximum Jacq cv. Aruana, P. maximum Jacq cv. Aries, P. maximum Jacq cv. Paredão, Setaria sphacelata (Nandi) cv. Kazungula and Andropogon gayanus Kunth. Brachiaria brizantha (Hochstex A. Rich.) Stapf. ecotype BB185 was used as a control because this species demonstrated a high susceptibility to M. fimbriolata in preliminary trials. After infestation, the plants were kept in a greenhouse.

Evaluations (observations of the live nymphs) were initiated 30 days after infestation. When the nymphs reached the 5<sup>th</sup> instar, a gauze cage was placed over the pot so that at emergence, the adults were retained therein. After the 30<sup>th</sup> day, nymph mortality was assessed daily until the emergence of adults to determine the duration and survival of the nymphal stage.

After emergence of adults, the insects were weighed using an analytical balance and separated into couples, which were kept in cylindrical cages and fed with the same grass as the type from which they emerged. The plants offered to the insects were grown in 200 mL cups, using Basaplant substrate and Osmocote fertilizer [14-14-14 (N, P, K)] applied at 200 g for each 25 kg of the substrate. Covering the soil, cotton pads were placed to serve as oviposition substrate and the cotton pads were changed daily. At this stage, the pre-oviposition period, number of eggs/female and adult longevity were assessed. To evaluate the viability and the duration of the embryonic period, the eggs were removed from the cotton with a brush (n° 000) and placed in Petri dishes lined with filter paper, which was moistened daily.

To examine the effect of the grasses on the development of *M. fimbriolata*, the data were analyzed by analysis of variance (ANOVA). The assumptions of ANOVA were checked using the Box-Cox optimal transformation family (Box and Cox 1964), and the Hartley test (Hartley 1950) test was applied to check the homogeneity of variances. The experimental design was completely randomized. The treatments consisted of 12 grasses, and the variables analyzed were the duration and viability of the nymph stage, adult weight and longevity, pre-oviposition period, number of eggs/female and duration and viability of the embryonic stage. After the assumptions of the ANOVA were confirmed, an F-test

(p > 0.05) was applied to test for possible differences between the treatments. If the null hypothesis among the treatments was rejected, comparisons of the means were made using Tukey's test (p > 0.05). To classify the natural relationships that the biology data presented, a cluster analysis was performed, using the mortality and duration of the nymphal stage, adult weight, pre-oviposition period, adult longevity, number of eggs per female and duration and viability of the embryonic period. The data were analyzed using the computer program SAS (Release 9.1.3 Service Pack 2, SAS Institute Inc., 2003, Cary, NC, USA).

### **RESULTS AND DISCUSSION**

The survival of the nymphal stage of *M. fimbriolata* was affected by the genotypes, and all treatments differed from the reference of susceptibility, *Brachiaria brizantha* ecotype

**Table 1.** Mean  $\pm$  Standard Error (SE) of the mortality (%) of the nymphal stage of *Mahanarva fimbriolata* reared on different grasses (at a temperature of 24  $\pm$  6 °C, relative humidity of 70  $\pm$  10% and photoperiod of 12 h).

| Treatment                                | Mortality         | Total emerged insects |      |
|--|-------------------|-----------------------|------|
| ireatment                                | (%)               | Female                | Male |
| Panicum maximum cv. Paredão              | 98.0 ± 0.13 a     | 0                     | 2    |
| Andropogon gayanus                       | $86.0 \pm 0.54$ b | 6                     | 8    |
| Panicum maximum cv. Aruana               | 71,0 ± 0.55 b     | 13                    | 16   |
| Brachiaria<br>decumbens cv.<br>Basilisk  | 51.0 ± 0.98 c     | 19                    | 26   |
| Brachiaria brizantha<br>cv. MG-4         | 49.0 ± 0.98 c     | 22                    | 29   |
| Brachiaria ruziziensis                   | 47.0 ± 0.90 c     | 26                    | 27   |
| Panicum maximum cv. Aries                | 44.0 ± 0.86 c     | 19                    | 37   |
| Setaria sphacelata<br>cv. Kazungula      | 42.0 ± 0.65 c     | 30                    | 28   |
| Brachiaria<br>humidicola cv.<br>Llanero  | 35.0 ± 0.56 d     | 22                    | 43   |
| Brachiaria<br>dictyoneura                | 34.0 ± 0.48 d     | 28                    | 38   |
| Brachiaria brizantha<br>cv. MG-5 Vitória | 30.0 ± 0.77 d     | 29                    | 41   |
| Brachiaria brizantha<br>ecotype BB185    | 14.0 ± 0.87 e     | 38                    | 48   |

Means followed by the same letter in the column do not differ according to Tukev's test (p > 0.05).

BB185, which had the lowest mortality (14.0%) (Table 1). The highest mortality was found in P. maximum cv. Paredão (98.0%), which differed from the other treatments, followed by A. gayanus (86%) and P. maximum cv. Aruana (71.0%), which did not differ between themselves but differed from the other treatments for which the mortalities showed intermediate values ranging from 30.0 to 51.0%. Mortality is one of the parameters that classify a grass as resistant to spittlebugs. Cardona et al. (1999) consider a grass to be resistant, intermediate and susceptible when survival of the spittlebugs tested is lower than 30%, between 31-50%, or higher than 50%, respectively. Auad et al. (2007) observed significant differences in the survival of nymphs of Mahanarva spectabilis (Distant) reared in elephant grass genotypes, classifying the genotypes Cameroon of Piracicaba, Pioneiro, Cuba 169, Santa Rita, Mineiro Ipeaco, Colombian Elephant, Pinda Mercker and CNPGL 96-27-3 as resistant by antibiosis. In addition to mortality, other authors (Lapointe et al. 1992; Valério et al. 1997; Valério et al. 2004) have used the duration of the nymphal stage. To characterize a grass as resistant, the average mortality and duration values need to be above the average values of the group, summing the value of the corresponding standard deviation (Lapointe et al. 1992). Analyzing the mortality found in the present study, it is possible to assess the variation among the grasses tested for spittlebug resistance that affected the biological parameters, with the resistance being classified by the mechanism of antibiosis.

For P. maximum cv. Paredão, nymphal mortality was the only parameter analyzed as only two nymphs reached the adult stage, which restrain the analysis of the duration of the nymphal stage and other parameters relating to the adult spittlebug stage. Considering the other treatments, when analyzing males and females together, the longer duration of this phase was found in *A. gayanus* (54.1 days) and P. maximum cv. Aruana (46.5 days), where the recorded values differed from those of the others (Table 2). These species were followed by S. sphacelata cv. Kazungula, for which the nymphal stage lasted 42.0 days, not differing significantly from B. brizantha cv. MG-4 (39.3 days), P. maximum cv. Aries (39.0 days) and B. brizantha cv. MG-5 Vitória (38.6 days). The other treatments had values ranging between 36.2 and 38.6 days, which did not differ between the treatments or from those recorded for the suscetible control (*B. brizantha* ecotype BB185).

The studies that have been conducted to select genotypes resistant to spittlebugs consider the parameters mortality

and duration of the nymphal stage as the most important (Lapointe et al. 1992; Cardona et al. 1999). Following the criteria adopted by Lapointe et al. (1992) for the nymphal stage, the grasses selected as resistant include *P. maximum* cv. Paredão, *A. gayanus* and *P. maximum* cv. Aruana, which showed average mortality and duration values higher than those of the group averages and corresponding standard deviations (Table 3).

Most studies on the resistance of grasses to spittlebugs involve research only with the nymphal stage (Cosenza et al. 1989; Cardona et al. 2004). López et al. (2009) developed a methodology to evaluate the resistance of grasses to adult stages based on the observation of the visible leaf damage and the loss of chlorophyll, but they did not address the parameters adopted in the present study.

Difference was found in adult weights of males and females (Table 4). In the case of the females, lower weights were recorded in the genotypes P. maximum cv. Aruana and A. gayanus, with values of 5.3 and 5.4 mg, respectively. B. brizantha ecotype BB185, the susceptible control, had the heaviest weight value (9.2 mg). The lowest male weight was obtained for A. gayanus (4.4 mg), which differed from the weights of the others, except for those recorded for P. maximum cv. Aruana (4.8 mg), S. sphacelata cv. Kazungula (5.1 mg) and *P. maximum* cv. Aries (5.7 mg). In the same way as for females, B. brizantha ecotype BB185 had the heaviest weight value (7.4 mg). Similar results were observed when the males and females were analyzed together, for which the average values obtained for A. gayanus (5.0 mg) and P. maximum cv. Aruana (5.1 mg) differed from the others, with the exception of that of S. sphacelata cv. Kazungula (5.7 mg). The difference in weight found between the grasses can be explained by the quality of the food, which influences the growth and development of insects (Klein and Kogan 1974), thereby affecting the growth rate, development time, insect weight and survival and influencing the adult fertility, longevity, movement and competition capacity (Parra 1991).

For the parameter pre-oviposition, there was no difference between the means, which ranged from 4.0 to 5.0 days for *B. brizantha* cv. MG-5 Vitória and *B. dictyoneura*, respectively (Table 5). The number of eggs varied greatly among and within treatments. In addition to *P. maximum* cv. Paredão, two other treatments (*P. maximum* cv. Aruana and *A. gayanus*) were not included in the analysis, as only two repetitions were obtained in

these two genotypes, which is an insufficient sample size for comparison with the other genotypes. Among the other genotypes, the lowest values for the number of eggs were obtained for *P. maximum* cv. Aries, *B. humidicola* cv. Llanero and *S. sphacelata* cv. Kazungula (20.5, 31.2 and 31.4 eggs, respectively), and these values differ from

**Table 2.** Mean  $\pm$  Standard Error (SE) of the duration (days) of the nymphal stage of *Mahanarva fimbriolata* reared on twelve grasses (at a temperature of 24  $\pm$  6 °C, relative humidity of 70  $\pm$  10% and photoperiod of 12 h).

| Treatment                          | Duration (days)*                     |                                      |                            |  |
|------------------------------------|--------------------------------------|--------------------------------------|----------------------------|--|
|                                    | Female                               | Male                                 | Female + Male              |  |
| Panicum maximum cv. Paredão        | -                                    | $40.0 \pm 2.00 (n = 2)$              | -                          |  |
| Andropogon gayanus                 | 55.0 ± 2.66 a (n = 6)                | 52.9 ± 0.76 a (n = 8)                | 54.1 ± 1.50 a              |  |
| Panicum maximum cv. Aruana         | $46.8 \pm 1.83$ b (n = 13)           | $46.1 \pm 1.51$ b (n = 16)           | 46.5 ± 1.18 b              |  |
| Setaria sphacelata cv. Kazungula   | 42.7 ± 1.13 bc (n = 30)              | 41.2 ± 0.75 bc (n = 28)              | 42.0 ± 0.68 c              |  |
| Brachiaria brizantha cv. MG-4      | $40.9 \pm 0.86$ cd (n = 22)          | $37.6 \pm 0.77$ cd (n = 29)          | $39.3 \pm 0.68$ cd         |  |
| B.brizantha cv. MG-5 Vitória       | $39.0 \pm 1.10 \text{ cd (n = 29)}$  | $38.1 \pm 1.02$ cd (n = 41)          | $38.6 \pm 0.74  \text{cd}$ |  |
| Panicum maximum cv. Aries          | $38.8 \pm 1.14 \text{ cd (n} = 19)$  | $39.1 \pm 1.33$ cd (n = 37)          | $39.0 \pm 0.85  \text{cd}$ |  |
| Brachiaria dictyoneura             | $38.6 \pm 0.68 \text{ cd (n = 28)}$  | $36.5 \pm 0.62  \text{cd}  (n = 38)$ | 37.5 ± 0.49 d              |  |
| Brachiaria humidicola cv. Llanero  | $38.6 \pm 0.81  \text{cd}  (n = 22)$ | 37.2 ± 1.46 cd (n = 43)              | 38.6 ± 0.81 d              |  |
| Brachiaria decumbens cv. Basilisk  | $38.0 \pm 0.58$ cd (n = 26)          | $37.0 \pm 1.07 \text{ cd (n} = 19)$  | 37.5± 0.56 d               |  |
| Brachiaria brizantha ecotype BB185 | $36.8 \pm 0.87  d  (n = 38)$         | $35.6 \pm 0.41  d  (n = 48)$         | $36.2 \pm 0.48 \mathrm{d}$ |  |
| Brachiaria ruziziensis             | 36.8 ± 0.67 d (n = 26)               | 37.3 ± 1.10 cd (n = 27)              | 37.0 ± 0.63 d              |  |

<sup>\*</sup>Means followed by the same letter in the column do not differ according to Tukey's test (p > 0.05).

**Table 3.** Mean ± Standard Error (SE) of the mortality (%) and duration (days) of the *Mahanarva fimbriolata* nymphal stage on *Panicum maximum* cv. Paredão, *Andropogon gayanus* and *Panicum maximum* cv. Aruana compared with the averages of all of the grasses.

| Parameter                | The group average | P. maximum cv. Paredão | A. gayanus      | P. maximum cv. Aruana |
|--------------------------|-------------------|------------------------|-----------------|-----------------------|
| Mortality                | $50.1 \pm 6.92$   | $98.0 \pm 0.13$        | $86.0 \pm 0.54$ | 71.0 ± 0.55           |
| Duration (female)        | 41.1 ± 1.65       | -                      | 55.0 ± 2.66     | 46.8 ± 1.83           |
| Duration (male)          | $39.1 \pm 0.85$   | $40.0 \pm 2.00$        | $52.9 \pm 0.76$ | 46.1 ± 1.51           |
| Duration (female + male) | 39.9 ± 0.38       | 40.0 ± 2.00            | 54.1 ± 1.50     | 46.5 ± 1.18           |

**Table 4.** Mean  $\pm$  Standard Error (SE) of the average weight of the male and female *Mahanarva fimbriolata* from nymphs fed on twelve grasses (at a temperature of 24  $\pm$  6 °C, relative humidity of 70  $\pm$  10% and photoperiod of 12 h).

| Treatment —                           | Weight (mg)*                |                             |                            |
|---------------------------------------|-----------------------------|-----------------------------|----------------------------|
| Heaunent –                            | Female                      | Male                        | Female + Male              |
| Panicum maximum cv. Paredão           | -                           | $5.8 \pm 0.68 (n = 2)$      | -                          |
| Panicum maximum cv. Aruana            | $5.3 \pm 0.29$ a (n = 13)   | $4.8 \pm 0.33$ ab (n = 16)  | 5.1 ± 0.25 a               |
| Andropogon gayanus                    | $5.4 \pm 0.17$ a (n = 6)    | $4.4 \pm 0.35$ a (n = 8)    | $5.0 \pm 0.29$ a           |
| Setaria sphacelata cv. Kazungula      | $6.3 \pm 0.28$ ab (n = 30)  | $5.1 \pm 0.15$ ab (n = 28)  | 5.7 ± 0.18 ab              |
| Panicum maximum cv. Aries             | $6.9 \pm 0.40$ abc (n = 19) | $5.7 \pm 0.28$ abc (n = 37) | $6.2 \pm 0.27  bc$         |
| Brachiaria humidicola cv. Llanero     | $7.1 \pm 0.26$ bcd (n = 22) | $6.0 \pm 0.10$ bcd (n = 43) | $6.6 \pm 0.15  \text{bcd}$ |
| Brachiaria ruziziensis                | $7.9 \pm 0.57$ cde (n = 26) | $6.5 \pm 0.33$ cde (n = 27) | $7.1 \pm 0.45$ cde         |
| Brachiaria decumbens cv. Basilisk     | $8.1 \pm 0.36$ cde (n = 19) | $6.6 \pm 0.34$ cde (n = 2)  | 7.5 ± 0.44 cde             |
| Brachiaria dictyoneura                | $8.2 \pm 0.84$ cde (n = 28) | $6.7 \pm 0.13$ cde (n = 38) | 7.0 ± 0.43 de              |
| Brachiaria brizantha cv. MG-4         | $8.3 \pm 0.32$ cde (n = 22) | 7.1 ± 0.09 de (n = 29)      | 7.7 ± 0.15 de              |
| Brachiaria brizantha cv. MG-5 Vitória | $8.6 \pm 0.33$ de (n = 29)  | $7.0 \pm 0.26$ de (n = 41)  | 7.8 ± 0.28 de              |
| Brachiaria brizantha ecotype BB185    | $9.2 \pm 0.38$ e (n = 38)   | $7.4 \pm 0.18$ e (n = 48)   | 8.5 ± 0.35 e               |

<sup>\*</sup>Means followed by the same letter in the column do not differ according to Tukey's test (p > 0.05).

those recorded in the other genotypes, except for those of *B. decumbens* cv. Basilisk and *B. dictyoneura* (54.7 and 58.5 eggs, respectively). For the other genotypes, although the averages varied between 98.4 and 116.1 eggs, there was no difference compared to the control (134.5 eggs). Sujii et al. (2001) compared the effect of the native host with an exotic host plant on the fecundity of *Deois flavopicta* (Stal) and found that the size of the female is not correlated with its fecundity and that the reproductive capacity is associated to obtaining food by the female.

The longevity of females reared in *P. maximum* cv. Aries (5.2 days) differed from those observed for *B. brizantha* 

cv. MG-5 Vitória (8.9 days) and in the control (9.1 days). The other treatments showed intermediate values that did not differ significantly from the extremes noted above (Table 6). As for the males, the shortest lifespans were obtained for insects reared on *S. sphacelata* cv. Kazungula (4.8 days) and *P. maximum* cv. Aruana (4.9 days), and these values differed from the rest, with the exception of those of *P. maximum* cv. Aries (5.9 days), *B. humidicola* cv. Llanero (7.1 days), *B. ruziziensis* (7.6 days) and *B. decumbens* cv. Basilisk (8.0 days). For the other grasses, the average values ranged from 8.4 to 10.3 days and did not differ from the longevity observed in the control (8.5 days). Similar

**Table 5.** Mean  $\pm$  Standard Error (SE) of the pre-oviposition period (days) and number of eggs per *Mahanarva fimbriolata* female feeding on eleven grasses (at a temperature of  $24 \pm 6$  °C, relative humidity of  $70 \pm 10\%$  and photoperiod of 12 h).

| Treatment                             | Pre-oviposition*          | Number of eggs/female* |
|---------------------------------------|---------------------------|------------------------|
| Panicum maximum cv. Aruana            | -                         | 37**                   |
| Andropogon gayanus                    | -                         | 61**                   |
| Brachiaria dictyoneura                | $5.0 \pm 0.26$ a (n = 19) | 58.5 ± 12.58 ab        |
| Brachiaria humidicola cv. Llanero     | $4.7 \pm 0.18$ a (n = 13) | 31.2 ± 8.51 a          |
| Brachiaria ruziziensis                | $4.6 \pm 0.18$ a (n = 16) | 98.4 ± 18.63 bc        |
| Panicum maximum cv. Aries             | 4.5 ± 0.21 a (n = 12)     | 20.5 ± 4.79 a          |
| Brachiaria decumbens cv. Basilisk     | $4.4 \pm 0.18$ a (n = 16) | 54.7 ± 9.36 ab         |
| Brachiaria brizantha cv. MG-4         | $4.4 \pm 0.25$ a (n = 13) | 116.1 ± 20.24 bc       |
| Setaria sphacelata cv. Kazungula      | $4.2 \pm 0.20$ a (n = 10) | 31.4 ± 12.98 a         |
| Brachiaria brizantha ecotype BB185    | 4.2 ± 0.10 a (n = 17)     | 134.5 ± 15.65 c        |
| Brachiaria brizantha cv. MG-5 Vitória | 4.0 ± 0.14 a (n = 18)     | 108.3 ± 12.85 bc       |

<sup>\*</sup>Means followed by the same letter in the column do not differ according to Tukey's test (p > 0.05); \*\* Data not included in the analysis (only two repetitions were obtained in these two genotypes).

**Table 6.** Mean  $\pm$  Standard Error (SE) of longevity (days) of male and female *Mahanarva fimbriolata* feeding on eleven grasses (at a temperature of 24  $\pm$  6 °C, relative humidity of 70  $\pm$  10% and photoperiod of 12 h).

|                           | Longevity (days)*   |   |  |  |
|---------------------------|---|---|--|--|
|                           | Longevity (days)*   |   |  |  |
| Female                    | Male  | Female + Male   |  |  |
| 5.2 ± 0.50 a (n = 16)     | $5.9 \pm 0.57$ ab (n = 16)  | $5.6 \pm 0.38  ab$  |  |  |
| 5.8 ± 0.59 ab (n = 16)    | $4.8 \pm 0.41$ a (n = 16)   | 5.3 ± 0.36 a  |  |  |
| 6.4 ± 0.44 ab (n = 18)    | 7.1 ± 0.47 abc (n = 19)   | 6.8 ± 0.33 abc  |  |  |
| 6.4 ± 0.83 ab (n = 10)    | 4.9 ± 0.72 a (n = 9)  | 5.7 ± 0.57 ab   |  |  |
| 7.0 ± 1.73 ab (n = 4)     | $9.7 \pm 3.04$ bc (n = 4)   | $8.4 \pm 1.70 \text{ cd}$   |  |  |
| 7.1 ± 0.36 ab (n = 16)    | $8.0 \pm 0.68$ abc (n = 15)   | $7.5 \pm 0.39$ bcd  |  |  |
| 7.8 ± 0.91 ab (n = 18)    | $8.4 \pm 0.83$ bc (n = 20)  | $8.9 \pm 0.61  \text{cd}$   |  |  |
| 7.9 ± 0.82 ab (n = 18)    | $7.6 \pm 0.60$ abc (n = 20)   | 7.8 ± 0.47 bcd  |  |  |
| 8.5 ± 0.75 ab (n = 14)    | $10.3 \pm 0.68  \text{c}  (\text{n} = 14)$  | 9.7 ± 0.53 d  |  |  |
| $8.9 \pm 0.49$ b (n = 20) | $8.6 \pm 0.67$ bc (n = 19)  | 8.8 ± 0.42 cd   |  |  |
| 9.1 ± 0.94 b (n = 20)     | $8.5 \pm 0.65$ bc (n = 19)  | 9.0 ± 0.57 cd   |  |  |
|                           | $5.2 \pm 0.50 \text{ a } (n = 16)$<br>$5.8 \pm 0.59 \text{ ab } (n = 16)$<br>$6.4 \pm 0.44 \text{ ab } (n = 18)$<br>$6.4 \pm 0.83 \text{ ab } (n = 10)$<br>$7.0 \pm 1.73 \text{ ab } (n = 4)$<br>$7.1 \pm 0.36 \text{ ab } (n = 16)$<br>$7.8 \pm 0.91 \text{ ab } (n = 18)$<br>$7.9 \pm 0.82 \text{ ab } (n = 18)$<br>$8.5 \pm 0.75 \text{ ab } (n = 14)$<br>$8.9 \pm 0.49 \text{ b } (n = 20)$ | $5.2 \pm 0.50 \text{ a } (n = 16)$ $5.9 \pm 0.57 \text{ ab } (n = 16)$ $6.8 \pm 0.59 \text{ ab } (n = 16)$ $4.8 \pm 0.41 \text{ a } (n = 16)$ $6.4 \pm 0.44 \text{ ab } (n = 18)$ $7.1 \pm 0.47 \text{ abc } (n = 19)$ $6.4 \pm 0.83 \text{ ab } (n = 10)$ $4.9 \pm 0.72 \text{ a } (n = 9)$ $7.0 \pm 1.73 \text{ ab } (n = 4)$ $9.7 \pm 3.04 \text{ bc } (n = 4)$ $7.1 \pm 0.36 \text{ ab } (n = 16)$ $8.0 \pm 0.68 \text{ abc } (n = 15)$ $7.8 \pm 0.91 \text{ ab } (n = 18)$ $8.4 \pm 0.83 \text{ bc } (n = 20)$ $7.9 \pm 0.82 \text{ ab } (n = 18)$ $7.6 \pm 0.60 \text{ abc } (n = 20)$ $8.5 \pm 0.75 \text{ ab } (n = 14)$ $10.3 \pm 0.68 \text{ c } (n = 14)$ $8.9 \pm 0.49 \text{ b } (n = 20)$ $8.6 \pm 0.67 \text{ bc } (n = 19)$ |  |  |

<sup>\*</sup>Means followed by the same letter in the column do not differ according to Tukey's test (p > 0.05).

results were found when the males and females were analyzed together, as the shortest lifespan was obtained for insects from *S. sphacelata* cv. Kazungula (5.3 days), and these values differed from others, with the exception of those of *P. maximum* cv. Aries (5.6 days), *P. maximum* cv. Aruana (5.7 days) and *B. humidicola* cv. Llanero (6.8 days). *Setaria sphacelata* cv. Kazungula, *P. maximum* cv. Aries and *P. maximum* cv. Aruana were the only ones to show shorter longevity than that of the control (*B. brizantha* ecotype BB185), for which the value (9.0 days) did not differ from the other treatments with longevities ranging from 7.5 to 9.7 days.

The highest value for the embryonic period (17.9 days) occurred for the eggs of insects reared on *P. maximum* cv. Aries, differing from the results obtained for *B. ruziziensis* (15.9 days) and *S. sphacelata* cv. Kazungula (16.2 days) (Table 7). The other treatments showed intermediate values that did not differ from those of the extremes. The lowest value for egg viability was observed for *P. maximum* cv. Aries (54.7%), differing from the averages recorded for *B. ruziziensis*, *A. gayanus* and *B. dictyoneura* (90.5, 90.5 and 92.3%, respectively). The other treatments showed intermediate values that did not differ from those of the extremes.

**Table 7.** Mean  $\pm$  Standard Error (SE) of the viability and the embryo incubation period from eggs of adult *Mahanarva fimbriolata* reared on ten grasses (at a temperature of  $24 \pm 6$  °C, relative humidity of  $70 \pm 10$ % and photoperiod of 12 h).

| Treatment                             | Duration (days)*               | Viability (%)*                      |
|---------------------------------------|--------------------------------|-------------------------------------|
| Panicum maximum cv. Aries             | $17.9 \pm 0.53$ a (n = 32)     | $54.7 \pm 4.76$ a (n = 56)          |
| Brachiaria humidicola cv. Llanero     | 17.5 ± 0.50 ab (n = 98)        | 81.7 ± 12.15 ab (n = 112)           |
| Brachiaria decumbens cv. Basilisk     | $16.9 \pm 0.44$ ab (n = 213)   | $71.0 \pm 9.50$ ab (n = 300)        |
| Brachiaria brizantha cv. MG-4         | $16.7 \pm 0.27$ ab (n = 233)   | $82.0 \pm 8.13$ ab (n = 300)        |
| Brachiaria dictyoneura                | $16.6 \pm 0.16$ ab $(n = 278)$ | $92.3 \pm 0.59 \text{ b (n} = 300)$ |
| Andropogon gayanus                    | $16.5 \pm 0.33$ ab (n = 87)    | 90.5 ± 5.68 b (n = 98)              |
| Brachiaria brizantha ecotype BB185    | $16.5 \pm 0.28$ ab (n = 264)   | $88.0 \pm 6.11$ ab (n = 300)        |
| Brachiaria brizantha cv. MG-5 Vitória | $16.3 \pm 0.07$ ab (n = 126)   | $63.6 \pm 4.58$ ab (n = 211)        |
| Setaria sphacelata cv. Kazungula      | $16.2 \pm 0.51  b  (n = 100)$  | $62.2 \pm 21.47$ ab (n = 152)       |
| Brachiaria ruziziensis                | 15.9 ± 0.04 b (n = 269)        | $90.5 \pm 1.59  b  (n = 300)$       |

<sup>\*</sup>Means followed by the same letter in the column do not differ according to Tukey's test (p > 0.05).

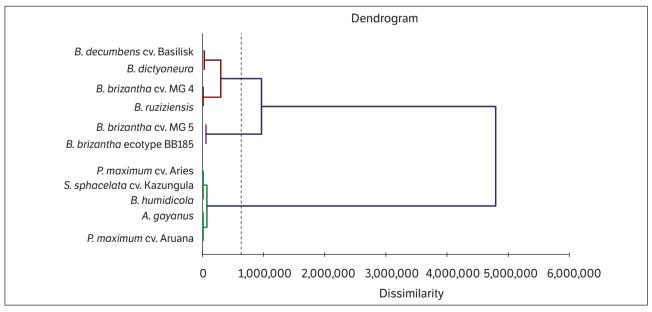


Figure 1. UPGMA clustering analysis based on biological data of Mahanarva fimbriolata on eleven grasses.

A dendrogram was generated and formed three distinct groups based on the dissimilarity of the tested grasses (Figure 1). From the results, it is possible to infer that the parameters that influenced the formation of the groups were the weight and longevity of the adults, especially the females. Group 1, which consisted of B. decumbens cv Basilisk, B. dictyoneura, B. brizantha cv. MG-4 and B. ruziziensis, behaved as intermediates for both the weight of the females (8.1, 8.2, 8.3 and 7.9 mg, respectively) and longevity of the females (7.1, 7.8, 8.5 and 7.9 days, respectively). Group 2 included B. brizantha MG-5 and B. brizantha ecotype BB185, which had the heaviest weights (8.6 and 9.2 mg, respectively) and longer lifespans (8.9 and 9.1, respectively) for the females. Finally, group three, which was formed by *P. maximum* cv. Aries, S. sphacelata cv. Kazungula, B. humidicola, A. gayanus and P. maximum cv. Aruana, showed the lightest weights (6.9, 6.3, 7.1, 5.4 and 5.3 mg, respectively) and shortest female longevities (5.2, 5.8, 6.4, 7.0 and 6.4 days, respectively).

Based on the results obtained for both the nymph and adult stages, it is possible to verify a negative influence on some biological aspects of *M. fimbriolata*. The high mortality and prolonging of the nymph stage allows the characterization of some grasses as promising source of resistance to this spittlebug due to the population reduction that these grasses caused in the present study. Other grasses, such as *P. maximum* 

cv. Aries, showed great potential for controlling adult stages despite not behaving as resistant to the nymphal stage, as it resulted in lighter adult weights, mainly related to the females, which, in turn, generated the least amount of eggs described in this paper. Combined with the lower adult longevity, the low number of eggs can drastically reduce the population of this insect. López et al. (2009) suggested that grasses with high levels of antibiosis to nymphs can be highly damaged by adults of the same species and recommended the inclusion of adult assessments as additional selection criteria in the development programs of new cultivars.

Studies examining forage grass resistance to *M. fimbriolata* are scarce, and due to the current increase in this species in grasslands, there is a need for further studies. The data obtained in the present study may lead to refinement and/ or adjustments to the methodology used in more in-depth studies that may even be conducted in the field.

#### CONCLUSION

*P. maximum* cv. Paredão can be characterized as resistant to *M. fimbriolata* by antibiosis in view of high mortality of the nymphs reared on this genotype.

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