

Sorghum genotypes with potential resistance to the aphid *Melanaphis sorghi* (Hemiptera: Aphididae)

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ABSTRACT

The sorghum aphid *Melanaphis sorghi* (Hemiptera: Aphididae) has become the primary threat to crop production in various parts of the world. Although the combined use of organosynthetic insecticides and resistant hybrids is a common strategy in countries like the USA, Brazilian farmers face the challenge of a lack of registered active ingredients and resistant cultivars against *M. sorghi*. This study aimed to evaluate grain sorghum genotypes for resistance to *M. sorghi* infestation. In the initial screening phase, 28 genotypes were evaluated, from which eight were selected and grouped as: i) resistant to *M. sorghi* (BRS373, CMSXS3017, BRS310, and SC110 [control]); and ii) susceptible (CMSXS3012, 1822043, 2116017, and AG 1085), for further assessment and determination of productivity in subsequent stages. The trial was conducted in a greenhouse under controlled humidity and temperature, with three controlled artificial infestations and injury assessments every seven days. Aphid infestation primarily reduced the plant fresh weight of the plant and grain size, negatively affecting grain weight per panicle. Without *M. sorghi* infestation, the hybrid genotypes AG 1085, 1822043, and CMSXS3012 showed the highest grain weight; however, they were susceptible to aphids when infested. Under infestation, the most resistant genotypes with the highest grain weight values were CMSXS3017, SC110, BRS373, and BRS310. The SC110 lineage was minimally affected by aphid infestation and should be used as a source of resistance in breeding programs for aphid tolerance. The results of this study may be used to evaluate productivity losses due to sorghum aphid infestation and to propose potential resistant genetic materials. Combined with other integrated management tactics, these findings can make sorghum aphid control more precise and sustainable.

Introduction

The sorghum aphid *Melanaphis sorghi* (Hemiptera: Aphididae), previously identified as the sugarcane aphid *Melanaphis sacchari* (Nibouche et al., 2021), is considered the primary phytosanitary problem for sorghum cultivation in the Americas (Nibouche et al., 2018; Avellar et al., 2023). Its successful invasion and establishment in these areas are attributed to its adaptability to a wide range of climatic conditions and ecosystems (Armstrong et al., 2015; Haar et al., 2019), its broad host range (Bowling et al., 2016), and its efficient reproductive and dispersal capacity, which present challenges for effective control, leading to significant economic losses (Souza and Davis, 2020; Harris-Shultz and Ni, 2021).

The sorghum aphid feeds on phloem sap, which is rich in sugars, from healthy leaf and stem tissues (Royer et al., 2015). Feeding injuries

caused by *M. sorghi* also result in the production of large amounts of honeydew, a sugary substance that promotes the growth of dark sooty mold fungi on plant surfaces. This creates favorable conditions for phytopathogens, reducing photosynthesis in affected leaves (Bowling et al., 2016; Pekarcik and Jacobson, 2021). Additionally, plants may exhibit symptoms such as chlorosis and necrosis of leaves, stunted growth, delayed or hindered panicle emergence, and reduced or arrested plant development, directly affecting crop yield (Haar et al., 2019; Wilson et al., 2020; Lahiri et al., 2021).

Considering the highly destructive and reproductive capacity of the pest, which has intensified with rising global temperatures, assertive field management of *M. sorghi* is essential. Host plant resistance, combined with other integrated management tactics, is a crucial tool in *M. sorghi* control. Resistant cultivars provide a baseline of protection, suppressing population growth rates, reducing injuries, and improving grain yield (Pekarcik and Jacobson, 2021). Therefore, it is urgent to

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study sorghum plant defenses that can be incorporated into breeding programs for resistance to sorghum aphids.

Plant resistance to herbivorous insects and other arthropods is a complex phenomenon involving a variety of plant traits that reduce herbivore damage. These resistance mechanisms are often categorized into three primary types: antixenosis, antibiosis, and tolerance (Painter, 1951). Understanding these mechanisms is crucial for developing sustainable pest management strategies and breeding resistant crop varieties. Antixenosis, also referred to as non-preference, describes plant characteristics that deter herbivores from colonizing, feeding, or ovipositing on a particular host (Dudareva et al., 2013; Smith et al., 2021; Zhang et al., 2022). These characteristics can involve a range of physical and chemical plant attributes. Antibiosis involves plant traits that negatively affect herbivore biology, such as survival, growth, development, or reproduction, after the herbivore has begun to feed on or utilize the plant (Dahlman and Hibbs, 1975). These effects are typically mediated by toxic or antinutritional compounds present in the plant tissue. Finally, the tolerance differs from antixenosis and antibiosis in that it does not affect herbivore behavior or biology. Instead, tolerance describes the ability of a plant to withstand or recover from herbivore damage without significant yield or fitness reduction (Painter, 1951). Tolerant plants can maintain their productivity despite sustaining a level of herbivore damage that would severely affect susceptible plants (Rao et al., 2020).

While the combined strategy of resistant hybrids and organosynthetic insecticides has been validated in various studies in the USA (Lahiri et al., 2021; Pekarcik and Jacobson, 2021; Uyi et al., 2022; Vasquez et al., 2025), there are no registered organosynthetic insecticides for *M. sorghi* control in sorghum crops in Brazil. Thus, investigations into resistant sorghum cultivars should be prioritized. Initial steps in plant breeding focus on identifying resistance in regionally adapted commercial materials already bred for other characteristics, especially grain yield. Only after this preliminary evaluation, and if no source of resistance is found, the next step is to introduce resistance from exotic or less-enhanced material.

Studies indicate that different sorghum genotypes exhibit significantly distinct phenotypic responses influenced by aphid infestation (Armstrong et al., 2015; Bayoumy et al., 2016). Given the current scenario of high field infestations by the sorghum aphid, it is necessary to find genotypes with potential resistance traits to recommend them in sorghum integrated pest management and breeding programs. This study aimed to evaluate sorghum genotypes (pre-commercial and commercial) for resistance levels to the sorghum aphid *Melanaphis sorghi*.

Materials and methods

Plants

The trials were conducted in a controlled greenhouse environment with an average temperature of $26 \pm 2^\circ\text{C}$ and relative humidity of $60 \pm 10\%$.

Initially, 28 grain sorghum genotypes, including varieties and pre-launch hybrids from the Embrapa Milho e Sorgo (Brazil) germplasm bank were evaluated. After the screening phase, eight genotypes were selected: three commercial, four in the pre-launch phase, and one resistant lineage (=standard). The selected grain sorghum hybrids included BRS373, CMSXS3017, AG1085, CMSXS3012, BRS310, 1822043, and 2116017, as well as the resistant SC110 lineage (=standard). Among these, four genotypes (BRS373, CMSXS3017, BRS310, and SC110) were identified as more resistant to *M. sorghi*, while four (CMSXS3012, 1822043, 2116017, and AG1085) were classified as susceptible.

Transplantation was performed in 18-liter pots filled with Tropstrato Hortaliças® commercial substrate to evaluate plant characteristics

such as plant fresh weight, plant dry weight, 1000-grain weight (with 10 replicates =plants), and grain yield to assess the damage caused by aphids. Each pot was irrigated individually as needed to maintain substrate saturation throughout the trial.

Insects and plant infestation

Populations of *M. sorghi* were collected from both greenhouse-grown (under controlled conditions at $25 \pm 2^\circ\text{C}$, $65 \pm 10\%$ relative humidity, and a photoperiod of 16 hours light and 8 hours dark) and field-grown plants. Artificial infestation was performed 19 days after sowing. Leaf fragments of approximately 4 cm^2 , taken from infested plants in the greenhouse, were used to introduce aphids to the experimental plants.

Infestation percentage and the severity of aphid-induced injuries were evaluated at 21 days after each infestation cycle. At the end of the experiment, the aphid population was controlled with foliar application of Verter® insecticide (Sulfoxaflor: Sulfoximines) at a rate of 70 ml/ha to prevent further damage and allow the completion of the crop cycle.

A five-grade scale was employed to assess infestation, progressing from Grade 1 (20%) with few aphids and no exuviae, to Grade 2 (40%) where exuviae became apparent. Grade 3 (60%) denoted a moderate aphid population, numerous exuviae, and the emergence of symptoms like leaf discoloration, with large colonies exceeding 50 aphids per leaf. Grade 4 (80%) was characterized by a large aphid population, abundant exuviae, and symptom expression both centrally and marginally on the leaves. Finally, Grade 5 (100%) represented a plant overwhelmed by aphids and exuviae, exhibiting severe symptoms throughout, and the beginning of leaf death. (Fernandes et al., 2021). Genotypes that did not survive until the end of the experiment were classified as “dead plants.” The uninfested control group of plants consisted of the same materials (i.e., eight treatments with five replicates). To manage potential infestations, Verter® insecticide was applied throughout the crop cycle at a dose of 0.4 L/ha.

Statistical analysis

Preliminary data analysis included the Shapiro-Wilk test to assess normality and the Levene test to evaluate homogeneity of variance, both at a 5% significance level. The presence of outliers in the response variables (=outliers) was also assessed using box-plot visualizations. Ensuring assumptions were met is necessary when applying the use of parametric tests.

A completely randomized design (CRD) was used, with explanatory variables (x's) being *M. sorghi* infestation and sorghum cultivar, and response variables (y's) including dry matter weight, plant fresh weight, grain weight, and 1000-grain weight (in grams). The plants were maintained in the greenhouse until the panicle reached the maturation stage for harvesting. An initial Analysis of Variance (ANOVA) was performed, followed by the Scott-Knott test for mean comparisons at a 5% significance level, provided that the assumptions for such models were satisfied.

For preliminary data analysis, model fitting, and graph generation, the R statistical environment (R Development Core Team, 2024, v.4.4.1) was used. The packages MASS, car, and rstatix were employed for model adjustments, while ggplot2 was used for generating graphs. The numerical dataset, scripts for model fitting, and graph generation will be stored in an open-access GitHub repository to enhance the reproducibility and verification of results by other researchers (Gandrud, 2020).

Results

The evaluation of *M. sorghi* injury in the 28 sorghum genotypes was assessed 21 days after infestation ($F = 4.69$, $df = 236$, $p < 0.001$).

The choice of genotypes to be evaluated in the next stage was based on the results in Table 1.

The infestation of the sorghum aphid *M. sorghi* reduced dry matter by 0.8 times ($F = 82.0$; $df = 1$; $p < 0.001$; Fig. 1a), plant fresh weight by 0.5 times ($F = 129.9$; $df = 1$; $p < 0.001$; Fig. 1b), grain weight by 0.5 times ($F = 214.9$; $df = 1$; $p < 0.001$; Fig. 1c), and 1000-grain weight by 0.5 times ($F = 202.0$; $df = 1$; $p < 0.001$; Fig. 1d) in sorghum plants.

In the absence of *M. sorghi* infestation (Fig. 2, green bars), the genotypes with the highest dry matter values, in descending order, were 1822043, CMSXS3012, 2116017, and AG1085 ($F = 15.2$; $df = 7$; $p < 0.001$; Fig. 2a). For plant fresh weight, the highest values were observed in 1822043, CMSXS3012, AG1085, and 2116017 ($F = 10.6$; $df = 7$; $p < 0.001$; Fig. 2b). For grain weight, the highest values were for AG1085, 1822043, and CMSXS3012 ($F = 5.2$; $df = 7$; $p < 0.001$; Fig. 2c), and for the 1000-grain weight, the highest value was observed in AG1085 ($F = 13.4$; $df = 7$; $p < 0.001$; Fig. 2d).

In the presence of *M. sorghi* infestation (Fig. 2, orange bars), cultivars with the highest dry matter values, in descending order, were CMSXS3012, AG1085, CMSXS3017, BRS373, 2116017, and 1822043 ($F = 4.6$; $df = 7$; $p < 0.001$; Fig. 2a). For plant fresh weight, the highest values were in 1822043, BRS373, AG1085, CMSXS3012, and CMSXS3017 ($F = 2.6$; $df = 7$; $p = 0.019$; Fig. 2b). For grain weight, the highest values were for CMSXS3017, SC110 (=resistant standard), BRS373, and BRS310 ($F = 3.5$; $df = 7$; $p = 0.002$; Fig. 2c), and for the 1000-grain weight, the highest values were observed in SC110, 1822043, CMSXS3012, BRS373, and AG1085 ($F = 2.9$; $df = 7$; $p = 0.011$; Fig. 2d).

Discussion

The study assessed the reduction in production and the resistance levels of sorghum genotypes to the sorghum aphid *M. sorghi*. The genotypes were grouped into two categories: the first being genotypes less susceptible to the attack of the sorghum aphid, based on the highest values of the production component, grain weight: CMSXS3017, SC110, BRS373, and BRS310; and the second group being susceptible genotypes, with lower grain weight values: AG1085, 2116017, 1822043, and CMSXS3012. Hybrids AG1085, 1822043, and CMSXS3012 were among the most productive without infestation but proved susceptible when exposed to *M. sorghi*, which resulted in significant reductions in grain size.

The SC110 lineage (=resistant control) demonstrated a unique response in which despite a reduction in plant fresh weight caused by the aphids, there was no impact on grain production, as reflected in the stable grain and 1000-grain weights. This suggests that SC110 may exhibit a tolerance mechanism, allowing it to withstand injury without yield loss. Tolerance is a plant response that does not affect insect behavior or development. It plays a crucial role in breeding resistant cultivars since insects have a great ability to develop resistant biotypes to plants. This defense mechanism also raises the economic damage threshold for crops, reducing the immediate need for other control tactics (Reese et al., 1994).

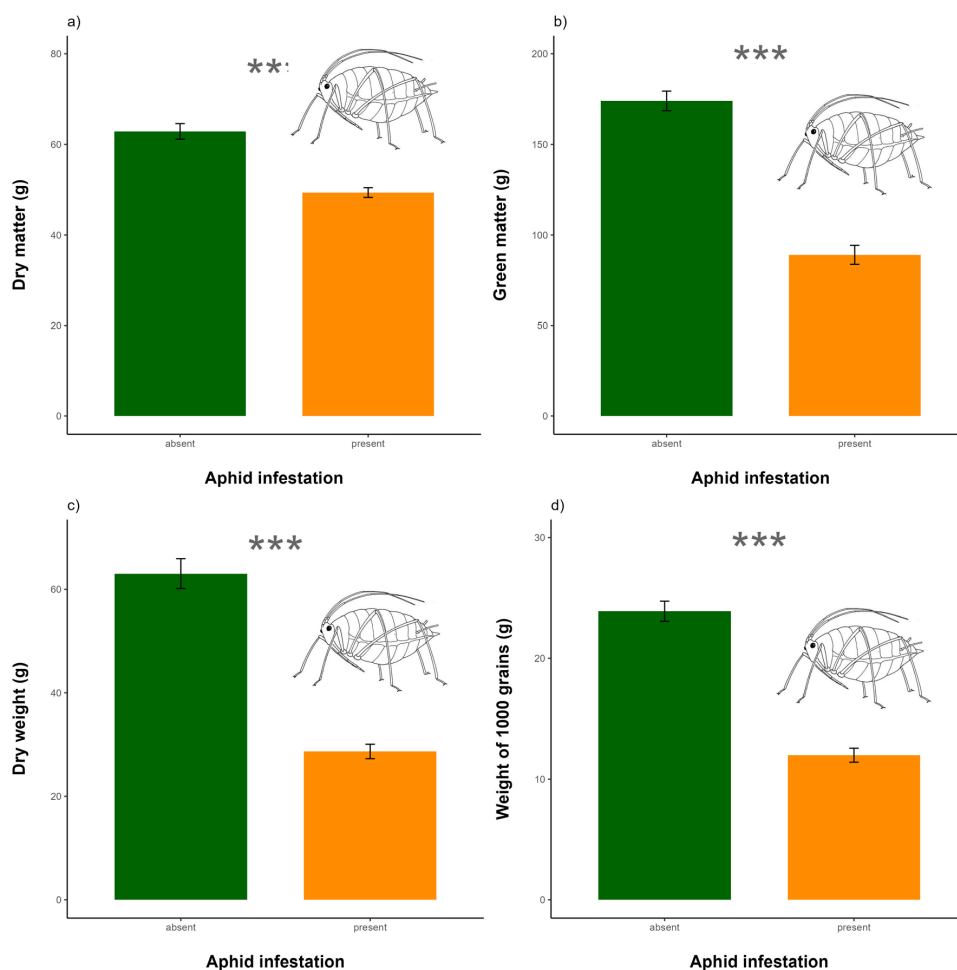


Figure 1 Infestation by the sorghum aphid *Melanaphis sorghi* reduced (a) dry matter, (b) plant fresh weight, (c) grain weight, and (d) 1000-grain weight (mean $\pm$ standard deviation) in sorghum plants. (***) statistically significant difference at 1% significance level for F-test). July 2024. Sete Lagoas, MG, Brazil.

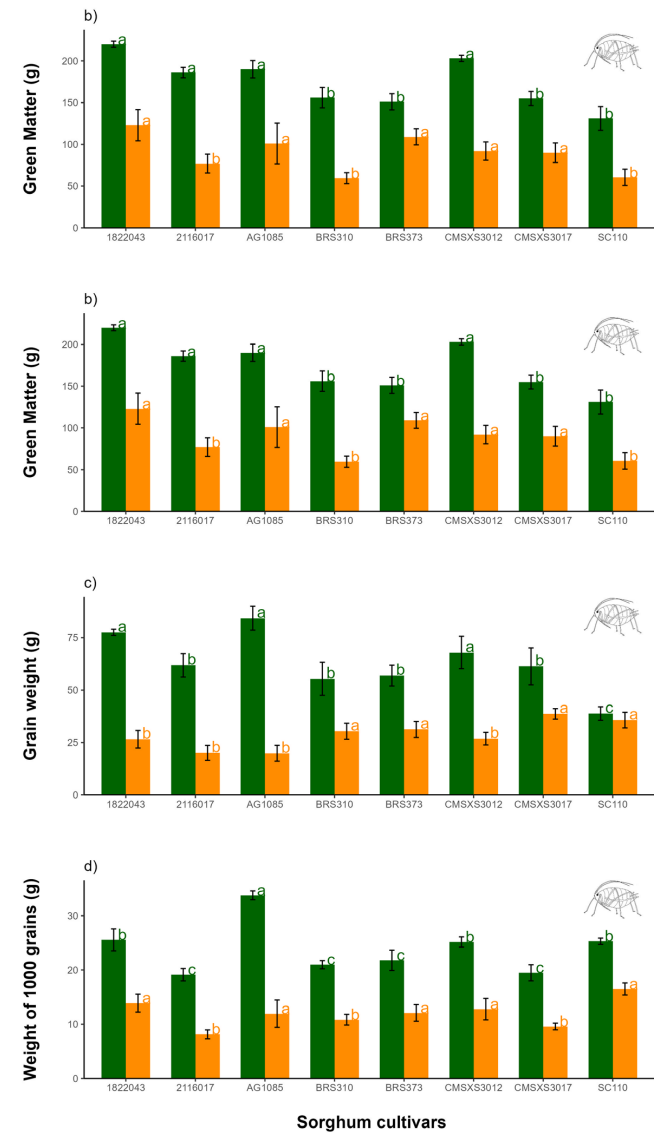


Figure 2 Mean $\pm$ standard deviation of (a) dry matter, (b) plant fresh weight, (c) grain weight, and (d) 1000-grain weight for eight sorghum cultivars under absent and present infestation of the aphid *Melanaphis sorghi*. (Bars followed by the same letter of the same color do not differ statistically by the Scott-Knott post hoc test at 5% significance). July 2024. Sete Lagoas, MG, Brazil.

Phenotypic plasticity responses of sorghum genotypes to *M. sorghi* infestation varied significantly. Our results show that *M. sorghi* infestation affects critical components such as dry matter, plant fresh weight, grain weight, and 1000-grain weight. High aphid densities can cause physiological stress, including soot mold (*Capnodium* spp.) incidence, leading to symptoms like wilting, chlorosis, and tissue necrosis (Bowling et al., 2016), which can reduce production by over 50% in susceptible hybrids.

In other regions, such as the southern USA, Asia, Africa, Oceania, and other Latin American countries, *M. sorghi* is a pest of economic relevance, causing significant yield losses (Singh et al., 2004; Peña-Martínez et al., 2016; Brewer et al., 2017; Szczepaniec, 2018; Lahiri et al., 2021). In Brazil, the lack of registered insecticides for sorghum poses additional challenges for controlling this pest.

Research efforts have focused on identifying and selecting resistant plants (Armstrong et al., 2015; Bayoumy et al., 2016; Nibouche et al., 2021). Resistant cultivars provide baseline protection by suppressing

Table 1
Mean ($\pm$ standard deviation) of *Melanaphis sorghi* injury at 21 days after the initial infestation in a greenhouse. Means followed by the same letter do not differ by the Scott-Knott test at 5% significance.

Hybrid	Mean $\pm$ SE
AG 1085	72.0 $\pm$ 26.6 a
1822043	68.0 $\pm$ 18.7 a
2116017	66.0 $\pm$ 28.8 a
CMSXS 3021	59.0 $\pm$ 22.4 a
1822044	58.0 $\pm$ 29.0 a
CMSXS 3019	58.0 $\pm$ 28.6 a
1921008	57.0 $\pm$ 28.3 a
1822048	53.0 $\pm$ 25.8 b
1920012	52.0 $\pm$ 27.4 b
2116019	51.0 $\pm$ 24.7 b
2116037	51.0 $\pm$ 2.8 b
CMSXS 3016	50.0 $\pm$ 24.9 b
CMSXS 3011	50.0 $\pm$ 21.6 b
CMSXS 3020	46.0 $\pm$ 28.8 b
1921007	45.0 $\pm$ 21.7 b
CMSXS 3004	44.0 $\pm$ 19.6 b
2120043	42.0 $\pm$ 21.5 b
BRS 3318	42.0 $\pm$ 23.0 b
SC 110	39.0 $\pm$ 7.4 b
CMSX S3012	37.0 $\pm$ 21.6 b
1527057	34.0 $\pm$ 9.7 b
1920007	34.0 $\pm$ 22.2 b
CMSXS 3014	33.3 $\pm$ 5.8 b
BRS 373	33.0 $\pm$ 18.9 b
CMSXS 3017	32.0 $\pm$ 23.5 b
BRS 310	21.0 $\pm$ 7.4 b

aphid population growth rates, reducing injury and damage, and directly benefiting grain yield (Lahiri et al., 2021). Evaluation of resistance mechanisms of *M. sacchari* in grain sorghum and forage sorghum, such as tolerance, antibiosis and antixenosis (Armstrong et al., 2017; Paudyal et al., 2019; Gordy et al., 2021). As well as integrated control tactics, such as biological and chemical methods (Brewer and Elliott, 2004).

As shown in our results, Gordy et al. (2021) also reported that infestation during sorghum panicle formation resulted in delayed development and malformed grains. Furthermore, infestation during grain filling reduced grain size and weight, causing cumulative yield losses due to multiple stressors, including aphids as biotic stress agents.

This study provides a foundation for future field research on *M. sorghi* population ecology and interactions with the studied hybrids. Plant-insect interactions vary by environment. Some research groups have also focused their efforts in this direction, seeking to understand the different levels of resistance of hybrids to the sorghum aphid (Bowling et al., 2016; Haar et al., 2019; Souza and Davis, 2020; Gordy et al., 2021). Resistance is strongly influenced by climatic factors like temperature and water regimes. Therefore, considering systematic changes in the climate, studies on environmental drivers of infestation intensity are essential for pest management.

Conclusion

In conclusion, *M. sorghi* infestation impacts dry matter, plant fresh weight, grain weight, and 1000-grain weight in the studied hybrids. The hybrids CMSXS3017, BRS373, and BRS310, along with the SC110 lineage, showed higher grain weight values, with SC110 (=control) maintaining yield despite reduced plant fresh weight.

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Conflicts of interest

The authors declare that there is no conflict of interest to disclose.

Author contribution statement

AFC, SMM, CBM and MAMF conceived the study. AFC, SMM, CBM, ICMO and MPMJ designed the methodology. MAMF performed the formal analysis. AFC, SMM, CBM, PTNM, MAMF, ICMO and MPMJ prepared the original draft. AFC, SMM, CBM, PTNM and MAMF reviewed and edited the manuscript. All authors read and approved the final version of the manuscript.

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