



# Influence of Physical and Morphological Factors On the Preference and Colonization of *Bemisia Tabaci* MED in Soybean Genotypes

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Received: 14 November 2023 / Accepted: 15 January 2024 / Published online: 19 February 2024  
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## Abstract

The whitefly, *Bemisia tabaci* Mediterranean (MED), is an invasive pest of several crops, including soybeans. The objective of this study was to evaluate the resistance of soybean genotypes to *B. tabaci* MED, in addition to the influence of possibly related physical and morphological factors. A no-choice test was carried out with 90 soybean genotypes. Subsequently, 35 materials were selected for further no-choice and multiple-choice tests. Trichomes and leaf color of plants were observed, with the aim of correlating these factors with the preference and colonization of *B. tabaci* MED. The genotypes KS 4202, TMG 1188 RR, M 7739 IPRO, 65165 IPRO, and PI 229358 were the least preferred by adults of *B. tabaci* MED. In the multiple-choice test, the lowest numbers of eggs and nymphs per square centimeter were observed for the genotypes Dowling, PI 229358, IAC 24, KS 4202. The genotypes IAC 19, TMG 1288 RR, TMG 1182 RR, 99R09, Dowling, and TMG 2375 IPRO presented the lowest numbers of eggs and nymphs in the no-choice assay. Plants with higher trichome density were preferred by adults of *B. tabaci* MED and, consequently, were more heavily colonized by these insects. Plants with leaves of lower luminosity and reduced green and yellow intensity were more attractive to the whiteflies. In summary, genotypes IAC 24, IAC 19, Dowling, 99R09, TMG 1182 RR, TMG 1288 RR and TMG 2375 IPRO exhibited lower colonization by *B. tabaci* MED in both assays, thus indicating their potential as promising sources of resistance to *B. tabaci* MED.

**Keywords** Host plant resistance · Trichome · Colorimetry · Antixenosis · Antibiosis

## Introduction

The sweetpotato whitefly, *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae), is composed of cryptic (sibling) species group that are morphologically indistinguishable, requiring the use of molecular markers for species identification (De Barro et al. 2011; Boykin and De Barro

2014; Brown et al. 2023). Two of them are considered invasive, the Middle East-Asia Minor 1 (MEAM1) (known as B biotype and *Bemisia argentifolii* Bellows & Perring (Bellows et al. 1994)) and the Mediterranean (MED, known as Q biotype) and considered as the *B. tabaci* sensu stricto (Tay et al. 2012; Brown et al. 2023). These insects cause damage to several species of cultivated plants, including soybean, cotton, tomato, and bean (Vieira et al. 2011; De Barro et al. 2011; Ramos et al. 2018).

The *B. tabaci* MEAM1 was first reported in Brazil in 1991 and is predominantly present (Lourenção and Nagai 1994; De Moraes et al. 2018). Since its introduction, this insect has gained notoriety as a pest in soybean crops, with losses that can reach up to 30% of crop productivity (Vieira et al. 2011, 2013). The MED species has a more recent occurrence in Brazil, being first reported in 2014, associated with ornamental plants and greenhouse-grown vegetables (Barbosa et al. 2015). However, it is known that MED already occurs in soybean open fields in the states of São Paulo and Paraná (Brazil) (Bello et al. 2021).

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Among the damages caused by *Bemisia tabaci*, the phloem sap-sucking by both nymphs and adults can be mentioned, resulting in direct damage to the plant's physiology (Zaidi et al. 2017; Perring et al. 2018; De Lima Toledo et al. 2021; Schutze et al. 2022; Farina et al. 2022). Indirect damage is also attributed to this pest. The release of honeydew by these insects promotes the growth of *Capnodium* spp fungi, causing sooty mold, which significantly reduces the total photosynthetic surface of the plant (Cameron et al. 2013; Cuthbertson and Vänninen 2015). *Bemisia tabaci* also acts as a powerful vector of plant viruses (Gilbertson et al. 2015). In Brazil, the transmission of the soybean stem necrosis virus (*Cowpea mild mottle virus*—CpMMV) can be carried out efficiently by MED and MEAM1, intensifying the problems caused by whiteflies to this crop (Bello et al. 2019).

The primary strategy for controlling whiteflies is the use of synthetic insecticides. However, several cases of loss of susceptibility have already been reported (Dângelo et al. 2017; Bielza et al. 2018; Hopkinson et al. 2020; Zhou et al. 2020; Wang et al. 2020; Du et al. 2023), particularly for *B. tabaci* MED, which exhibits higher levels of resistance due to its higher detoxification capacity when compared to MEAM1 (Horowitz et al. 2005; Sun et al. 2013; He et al. 2018), making it less susceptible to this type of control. Therefore, other management strategies must be adopted to reduce populations of this pest.

The use of resistant plants can play a crucial role in managing *B. tabaci* in soybean crops (Cruz and Baldin 2017). Resistant plants can exhibit antibiosis, affecting the biology of the pest, and antixenosis, influencing the behavior of the insects, resulting in the reduction of their populations (Canassa et al. 2020; Morando et al. 2021; Santos et al. 2023). There are also tolerant plants that do not affect the biology and behavior of the insects, and even with a high incidence of the pest, these plants can recover and remain productive (Smith 2005; Baldin et al. 2019). Several studies have been conducted to assess the resistance of soybean genotypes to *B. tabaci* MEAM1, leading to the identification of some sources of resistance based on antixenosis and/or antibiosis (Valle Do and Lourenção 2002; Vieira et al. 2011, 2016; Cruz and Baldin 2017; Baldin et al. 2017). However, to date, there has been a lack of studies aimed at evaluating the resistance of soybean genotypes to *B. tabaci* MED.

Several factors contribute to the expression of plant resistance to insects. Plant defense mechanisms include various morphological characteristics, such as trichomes, surface waxes, and leaf hardness (Smith 2005; Baldin et al. 2019). Additionally, the color spectrum expressed by plants can influence their attractiveness as a host, which may be decisive in attracting or repelling insects (Santos et al. 2020). Given the potential of MED as a pest and the lack of infor-

mation on sources of resistance in soybean plants in Brazil, this study aims to identify soybean genotypes resistant to this invasive pest, focusing on antixenosis and/or antibiosis. Furthermore, it seeks to understand aspects related to resistance by characterizing plant trichomes and leaf coloration.

## Materials and Methods

### *Bemisia Tabaci* MED Rearing

The initial population of *B. tabaci* MED was obtained from a greenhouse-grown pepper crop in São Miguel do Arcanjo, São Paulo. Species confirmation using the mtCOI analysis with primer pair Bem23F and Bem23R, followed the protocols described by De Barro et al. (2003). The insects were kept in metal cages (3×3×2.5 m), with the sides covered with anti-aphid mesh and the roof covered with transparent plastic and shade cloth. Poinsettia plants (*Euphorbia pulcherrima* (Willd.)), bell peppers (*Capsicum annuum* L.), and ornamental peppers (*Capsicum* spp.) were provided as a source of food and shelter. These plants were grown in plastic pots (2.5 L) and were periodically irrigated and replaced as needed.

### Obtaining Soybean Genotypes

Initially, 90 soybean genotypes were evaluated (Table 1). The plants were grown in plastic pots (2.5 L) containing a substrate composed of soil (dark red latosol), sand, and organic matter (cured cattle manure) in a 1:1:1 ratio. The substrate was fertilized according to the crop recommendations (Cantarella et al. 2022). Plants at the V3/V4 (three or four nodes on the main stem with fully developed leaves) phenological stage (Fehr and Caviness 1977) were used in all trials, which were kept in a greenhouse, free from insect infestation.

### Screening

The screening assay was conducted in a greenhouse, where plants of 90 different soybean genotypes were individually placed inside metal cages covered with *voile* fabric (35 cm in diameter×55 cm in height) and infested with 50 couples of *B. tabaci* MED.

At 21 days after infestation (21 DAI), six leaflets (two from each third of the plant) were removed for the counting of the number of eggs and nymphs present on the abaxial surface of the leaves, using a stereoscopic microscope (40×). Subsequently, the leaf area was measured using a LI 3000A leaf area meter (LI-COR Inc., Lincoln, NE, USA) to determine the number of eggs and nymphs per cm<sup>2</sup> (Baldin et al. 2005; Cruz and Baldin 2017). The assay was con-

**Table 1** Mean ( $\pm$  SE) number of eggs and nymphs per  $\text{cm}^2$  of *Bemisia tabaci* MED on ninety soybean genotypes and their respective origins

Genotype	Eggs/ $\text{cm}^2$	Nymphs/ $\text{cm}^2$	Origin
TMG 1180 RR	2.03 $\pm$ 0.17 e	0.46 $\pm$ 0.03 f	Tropical Melhoramento & Genética
Ultra BMX	1.25 $\pm$ 0.23 f	0.64 $\pm$ 0.12 f	Brasmax Genética
TMG 1288 RR	0.86 $\pm$ 0.09 g	0.67 $\pm$ 0.10 f	Tropical Melhoramento & Genética
Anta 82 RR	1.25 $\pm$ 0.18 f	0.72 $\pm$ 0.09 f	Tropical Melhoramento & Genética
TMG 1182 RR	1.37 $\pm$ 0.12 f	0.79 $\pm$ 0.06 f	Tropical Melhoramento & Genética
TMG 2375 IPRO	0.44 $\pm$ 0.05 g	0.88 $\pm$ 0.19 f	Tropical Melhoramento & Genética
BRS 1003 IPRO	0.74 $\pm$ 0.16 g	0.89 $\pm$ 0.12 f	Embrapa
IAC 78-2318	1.44 $\pm$ 0.19 f	0.90 $\pm$ 0.08 f	Instituto Agronômico de Campinas/IAC
99R09	0.70 $\pm$ 0.13 g	0.91 $\pm$ 0.17 f	Pioneer Seeds
TMG 7262 RR	1.57 $\pm$ 0.14 e	0.92 $\pm$ 0.13 f	Tropical Melhoramento & Genética
65I65 RSF IPRO	1.40 $\pm$ 0.15 f	0.93 $\pm$ 0.03 f	Brasmax Genética
ST 721 IPRO	0.66 $\pm$ 0.10 g	0.93 $\pm$ 0.12 f	SoyTech
FTS Campo Mourão RR	0.72 $\pm$ 0.15 g	0.97 $\pm$ 0.09 f	FT Sementes
95R95 IPRO	2.36 $\pm$ 0.29 d	0.97 $\pm$ 0.09 f	Pioneer Seeds
BRB 15—237.527	1.04 $\pm$ 0.05 g	0.97 $\pm$ 0.14 f	Embrapa
AS 3680 IPRO	1.41 $\pm$ 0.19 f	1.02 $\pm$ 0.08 f	Agroeste
TMG 2378 IPRO	0.63 $\pm$ 0.06 g	1.05 $\pm$ 0.09 f	Tropical Melhoramento & Genética
IAC 24	1.98 $\pm$ 0.14 e	1.05 $\pm$ 0.07 f	IAC
Dowling (PI 548663)	1.84 $\pm$ 0.24 e	1.08 $\pm$ 0.06 f	USDA (USA)
KS 4202	1.09 $\pm$ 0.18 f	1.08 $\pm$ 0.27 f	University of Nebraska (USA)
M 8866 IPRO	0.57 $\pm$ 0.10 g	1.14 $\pm$ 0.40 f	Monsoy
DS 6217 IPRO	1.81 $\pm$ 0.25 e	1.15 $\pm$ 0.35 f	Brevant Seeds
50I52 RSF IPRO	0.91 $\pm$ 0.04 g	1.15 $\pm$ 0.08 f	Brasmax Genética
TMG 1188 RR	1.16 $\pm$ 0.24 f	1.16 $\pm$ 0.12 f	Tropical Melhoramento & Genética
Conquista	1.62 $\pm$ 0.22 e	1.21 $\pm$ 0.09 f	Embrapa
BRS 391	1.16 $\pm$ 0.19 f	1.23 $\pm$ 0.18 f	Embrapa
P98Y51	1.38 $\pm$ 0.18 f	1.36 $\pm$ 0.20 e	Pioneer Seeds
TMG 2286 IPRO	1.33 $\pm$ 0.17 f	1.36 $\pm$ 0.20 e	Tropical Melhoramento & Genética
BRS 8381	3.28 $\pm$ 0.06 c	1.36 $\pm$ 0.25 e	Embrapa
BRS 539	4.69 $\pm$ 0.12 b	1.37 $\pm$ 0.20 e	Embrapa
96Y90 RR	0.66 $\pm$ 0.12 g	1.41 $\pm$ 0.19 e	Pioneer Seeds
NS 7901 RR	1.68 $\pm$ 0.18 e	1.52 $\pm$ 0.15 e	Nidera Seeds
BMX Potência RR	1.25 $\pm$ 0.06 f	1.52 $\pm$ 0.18 e	Brasmax Genética
ADV 4681 IPRO	0.62 $\pm$ 0.06 g	1.53 $\pm$ 0.17 e	Advanta Seeds
NA 5909	0.99 $\pm$ 0.17 g	1.53 $\pm$ 0.22 e	Nidera Seeds
M 5917 IPRO	0.69 $\pm$ 0.06 g	1.56 $\pm$ 0.14 e	Monsoy
IAC 17	1.34 $\pm$ 0.13 f	1.56 $\pm$ 0.24 e	IAC
IAC 19	1.33 $\pm$ 0.16 f	1.56 $\pm$ 0.16 e	IAC
TMG 7063 IPRO	1.88 $\pm$ 0.22 e	1.58 $\pm$ 0.25 e	Tropical Melhoramento & Genética
TMG 4377	1.30 $\pm$ 0.15 f	1.59 $\pm$ 0.26 e	Tropical Melhoramento & Genética
Desafio RR	1.01 $\pm$ 0.07 g	1.60 $\pm$ 0.12 e	Brasmax Genética
BMX Bônus IPRO 8579 RSF	1.81 $\pm$ 0.30 e	1.60 $\pm$ 0.16 e	Brasmax Genética
Coodetec 208	2.29 $\pm$ 0.16 d	1.61 $\pm$ 0.18 e	Coodetec
TMG 7067 IPRO	0.99 $\pm$ 0.04 g	1.62 $\pm$ 0.28 e	Tropical Melhoramento & Genética
BRS 543 RR	1.04 $\pm$ 0.08 g	1.67 $\pm$ 0.16 e	Embrapa
TMG 7058 IPRO	1.26 $\pm$ 0.24 f	1.70 $\pm$ 0.09 e	Tropical Melhoramento & Genética
55I57 RSF IPRO	3.27 $\pm$ 0.13 c	1.71 $\pm$ 0.33 e	Brasmax Genética
PI 227687	5.46 $\pm$ 0.17 a	1.72 $\pm$ 0.21 e	Japan
D75-10169	1.80 $\pm$ 0.18 e	1.78 $\pm$ 0.14 e	IAC
Coodetec 2820	3.36 $\pm$ 0.36 c	1.79 $\pm$ 0.22 e	Coodetec

**Table 1** (Continued)

Genotype	Eggs/cm <sup>2</sup>	Nymphs/cm <sup>2</sup>	Origin
IAC 23	1.83±0.25 e	1.86±0.15 e	IAC
TMG 7260 IPRO	2.29±0.17 d	1.86±0.25 e	Tropical Melhoramento & Genética
NS 7780 IPRO	1.90±0.14 e	1.92±0.19 e	Nidera Seeds
Jackson (PI 548657)	2.61±0.25 d	1.98±0.17 e	USDA (USA)
TMG 7062 IPRO	1.53±0.21 f	1.98±0.12 e	Tropical Melhoramento & Genética
IAC 100	2.33±0.17 d	2.01±0.30 e	IAC
TMG 2379 IPRO	1.84±0.15 e	2.01±0.21 e	Tropical Melhoramento & Genética
TMG 2383 IPRO	0.99±0.15 g	2.02±0.30 e	Tropical Melhoramento & Genética
M 9144	2.58±0.33 d	2.03±0.13 e	Monsoy
TMG 7363 RR	0.96±0.11 g	2.19±0.08 d	Tropical Melhoramento & Genética
TMG 4182	1.28±0.18 f	2.22±0.07 d	Tropical Melhoramento & Genética
L1-1-01	2.39±0.23 d	2.22±0.21 d	ESALQ/USP
M 5947 IPRO	1.80±0.12 e	2.24±0.36 d	Monsoy
BRS 284	1.22±0.30 f	2.32±0.17 d	Embrapa
BRS 399 RR	1.24±0.07 f	2.35±0.14 d	Embrapa
TMG 7061 IPRO	1.83±0.29 e	2.43±0.18 d	Tropical Melhoramento & Genética
TMG 2185 IPRO	3.42±0.28 c	2.45±0.34 d	Tropical Melhoramento & Genética
CZ 48B32 IPRO	1.64±0.20 e	2.46±0.30 d	Credenz
PI 274453	1.60±0.24 e	2.48±0.18 d	Japan
TMG 1179 RR	1.59±0.16 e	2.53±1.31 d	Tropical Melhoramento & Genética
UX 2569-159	1.70±0.20 e	2.55±0.29 d	University of Nebraska (USA)
PI 171451	1.32±0.24 f	2.59±0.24 c	Japan
TMG 4185	2.45±0.24 d	2.65±0.31 c	Tropical Melhoramento & Genética
TMG 2165 IPRO	1.94±0.36 e	2.69±0.37 c	Tropical Melhoramento & Genética
TMG 132 RR	2.26±0.25 d	2.70±0.28 c	Tropical Melhoramento & Genética
CD 2728 IPRO	2.50±0.18 d	2.72±0.27 c	Brevant Seeds
TMG 133 RR	2.26±0.11 d	2.74±0.15 c	Tropical Melhoramento & Genética
96R29 IPRO	3.23±0.20 c	2.81±0.36 c	Pioneer Seeds
BRS 523	1.39±0.22 f	2.89±0.27 c	Embrapa
IAC 18	1.93±0.26 e	2.90±0.20 c	IAC
TMG 2381 IPRO	2.42±0.19 d	2.91±0.28 c	Tropical Melhoramento & Genética
97R50 IPRO	2.14±0.32 d	3.03±0.30 b	Pioneer Seeds
NS 6700 IPRO	1.80±0.19 e	3.07±0.07 b	Nidera Seeds
PI 274454	2.78±0.20 c	3.23±0.19 b	Japan
TMG 7161 RR	1.23±0.18 f	3.25±0.15 b	Tropical Melhoramento & Genética
NS 7007 IPRO	1.69±0.26 e	3.31±0.11 b	Nidera Seeds
PI 229358	1.87±0.35 e	3.56±0.19 b	Japan
IAC 74-2832	4.32±0.26 b	3.57±0.20 b	IAC
M 8644 IPRO	1.25±0.17 f	3.61±1.66 b	Monsoy
M 7739 IPRO	5.23±0.41 a	4.57±0.24 a	Monsoy
<i>p</i>	<0.0001	<0.0001	–

Means followed by the same letter in the column do not differ significantly according to the Scott-Knott test ( $p > 0.05$ )

ducted in a completely randomized experimental design with 90 treatments (genotypes) and four replications. At the end of this bioassay, 35 genotypes were selected and used in the subsequent experiments.

### Multiple-choice Assay

To assess the preference of *B. tabaci* MED, pots containing plants of each genotype were randomly arranged in a circle inside metal cages, similar to those described for the rearing. The plants were spaced 15 cm apart from each other to prevent contact between their leaves. Subsequently, the

whiteflies were released from the ground and at the center, at a ratio of 50 couples per genotype (Baldin et al. 2005; Cruz and Baldin 2017). After 24, 48, and 72 h from the infestation, the number of adults present on the abaxial surface of six leaflets (two from each third of the plant) was recorded with the aid of a mirror.

At 48 DAI, six leaflets were collected from each plant to quantify the number of eggs, nymphs and exuviae left by adults after emergence (emerged adults) present on the abaxial surface with the aid of a stereoscopic microscope (40×). After counting, the leaf area was measured to determine the number of eggs, nymphs, and adults emerged per cm<sup>2</sup>. The assay was conducted in a randomized block design, with 35 treatments (genotypes) and eight replications. Each cage, containing the pots of all genotypes and insects, was considered a replicate.

### No-choice Assay

This experiment was conducted following the methodology used in the screening assay. However, the assessments of the number of eggs, nymphs, and emerged adults from the plants were performed at 48 DAI. A completely randomized design was used, with 35 treatments (genotypes) and four replications.

### Trichome Analysis

The density, angle, and length of trichomes were assessed on leaflets from the middle third of the plants. These assessments were carried out in the central region of each leaflet, on the right side of the central vein. Trichome density was quantified by counting the number of trichomes present in 1 cm<sup>2</sup> of the abaxial side of the leaflets, under a stereoscopic microscope, with 40× magnification. With the aid of a Hirox high-resolution stereoscopic microscope (KH-8700, Hirox), the length and inclination angle in relation to the 90° angle formed between the trichome and the leaf surface were measured using the Scandium software. A completely randomized design was used, with 35 treatments (genotypes) and eight replications, and each leaflet was considered a replication.

To obtain detailed images of the trichomes, leaflet samples were subjected to a JEOL JSM-IT300 LV scanning electron microscope (Tokyo, Japan) at 20kV, and the images were digitized (Fig. 2).

### Colorimetric Leaf Analysis

The assessment of color parameters was conducted on the adaxial side of leaflets from the middle third of the plants, through reflectance in the CIE color space using a Minolta Color Reader 300 colorimeter, which determines the

L\* (luminosity), a\* (green color intensity), and b\* (yellow color intensity) parameters. The L\* value can range from 0 (black) to 100 (white). The a\* value is represented by positive numbers when the object is red and negative numbers when the object is green. The value of b\* is positive when the object is yellow and negative when it is blue. The assay was carried out in a completely randomized design with 35 treatments (genotypes) and eight replications, with each leaflet considered as one replication.

### Statistical Analysis

Initially, the normality of residuals and homogeneity of variances were assessed using the Shapiro-Wilk and Bartlett tests, respectively. When the assumptions of normality and homogeneity were accepted, the data were subjected to analysis of variance, and the means were compared using the Scott-Knott test ( $p > 0.05$ ). When the assumptions were not met, a two-step cluster analysis was performed, starting with the fastclus procedure (PROC FASTCLUS, SAS) to identify the initial clusters, followed by the cluster procedure (PROC CLUSTER, SAS) using a hierarchical structure. The fastclus procedure uses Euclidean distances, where cluster centers are based on least squares estimates (LSE). In this clustering method, also called the 'k-means' model, the cluster centers are the means of the observations assigned to each cluster when the algorithm is run to complete convergence between the clusters. Each iteration reduces the least squares criterion until convergence is achieved, and the groups are defined. Clusters were characterized based on analysis of variance and  $\chi^2$  analyses using cluster variables as outcomes of cluster membership. Finally, the differences between the variables were verified through the contrast test between the groups, using generalized linear models (PROC GLM-contrast statement, SAS).

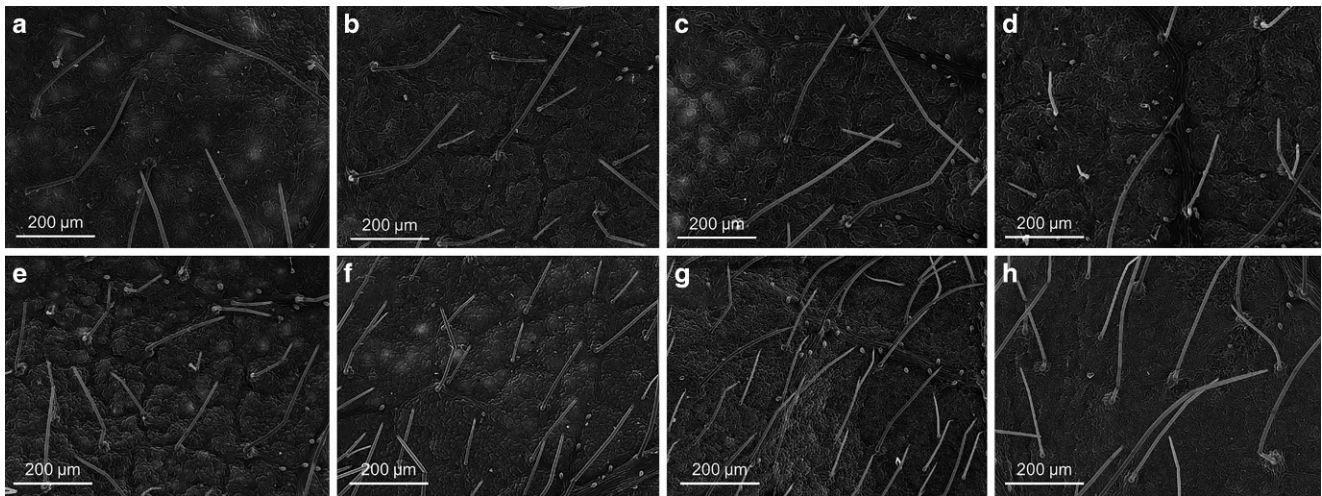
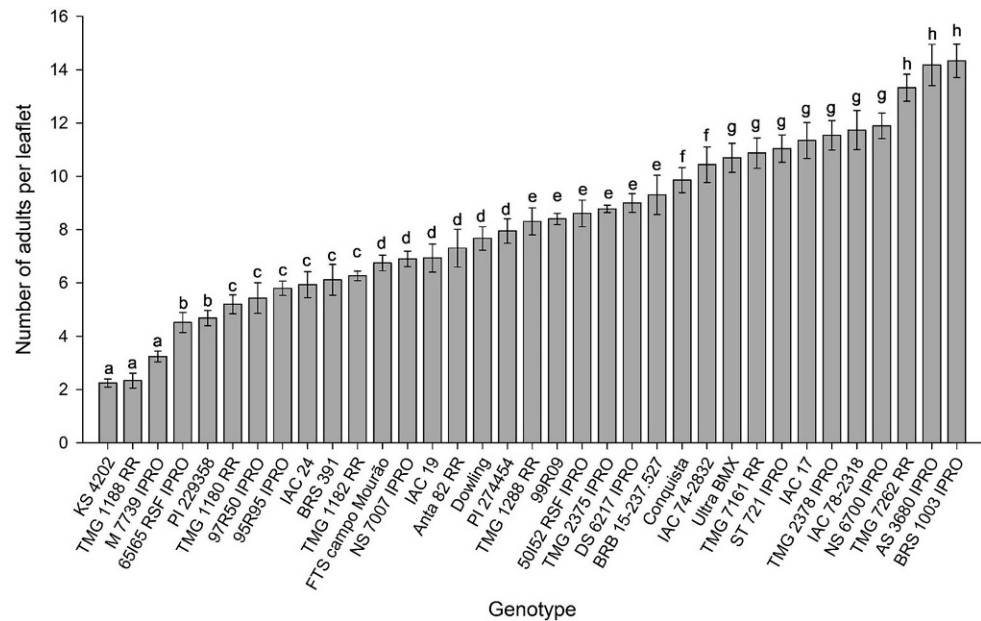
## Results

### Screening

In the preliminary tests, there was a significant difference between the treatments regarding the number of eggs and nymphs per cm<sup>2</sup> after 21 days of infestation (DAI) (Table 1). Genotypes with lower oviposition rates had fewer than 1.04 eggs per cm<sup>2</sup>, while genotypes such as PI 227687, M 7739 IPRO, BRS 539, and IAC 74-2832 had higher number of eggs, ranging from 5.46 to 4.32 eggs per cm<sup>2</sup>. On the other hand, 26 genotypes had an average number of nymphs per cm<sup>2</sup> lower than 1.23, which significantly differed from other treatments. The genotypes M 7739 IPRO, M 8644 IPRO, IAC 74-2832, PI 229358, NS 7007 IPRO, TMG 7161 RR, PI 274454, NS 6700 IPRO, and 97R50 IPRO showed



**Fig. 1** Mean ( $\pm$  SE) adult count of *Bemisia tabaci* MED per leaflet across 35 soybean genotypes after 24, 48, and 72 h of infestation in a free-choice experiment conducted in a greenhouse. Means followed by the same letter do not differ significantly from each other using the Scott-Knott test ( $p > 0.05$ )



**Fig. 2** Scanning electron microscopy of trichomes on the abaxial surface of soybean leaflets: **a** IAC 24, **b** Dowling, **c** PI 229358, **d** TMG 1288 RR, **e** BRS 1003 IRPO, **f** FTS Campo Mourão RR, **g** Ultra BMX, and **(H)** 99R09. From **(a)** to **(d)**, genotypes with the lowest trichome density, from **(e)** to **(h)**, genotypes with the highest trichome density

greater infestation by nymphs, with numbers varying between 4.57 to 3.03 nymphs per  $\text{cm}^2$ .

Based on these results, 35 genotypes were selected for subsequent tests, considering the number of nymphs per  $\text{cm}^2$  as a selection criterion. Twenty-five genotypes with fewer nymph infestations (considered more resistant) and eight with greater infestations (considered more susceptible) were chosen. Additionally, the genotypes IAC 17 and IAC 19 were included in subsequent trials due to their history of resistance to other pests (Canassa et al. 2017; Souza et al. 2017; Coelho et al. 2020; Ongaratto et al. 2021). The genotypes M 8866 IPRO and M 8644 IPRO (among the

least and most infested, respectively) were not used in subsequent trials due to low seed availability.

### Multiple-choice Assay

In the choice test, the genotypes KS 4202, TMG 1188 RR, M 7739 IPRO, 65165 IPRO, and PI 229358 were the least preferred by adults of *B. tabaci* MED, with less than 4.68 insects per leaflet (Fig. 1). On the other hand, the genotypes Ultra BMX, TMG 7161 RR, ST 721 IPRO, IAC 17, TMG 2378 IPRO, IAC 78-2318, NS 6700 IPRO, TMG 7262 RR, AS 3680 IPRO, and BRS 1003 IPRO attracted the highest number of insects per leaflet (10.69 to 14.34 insects/leaflet).

**Table 2** Mean ( $\pm$  SE) number of eggs, nymphs and adults emerged from *Bemisia tabaci* MED per  $\text{cm}^2$  in 35 soybean genotypes, after 48 DAI, in a multiple-choice assay, in a greenhouse

Genotype	Eggs/ $\text{cm}^2$	Nymphs/ $\text{cm}^2$	Emerged adults/ $\text{cm}^2$
TMG 1182 RR	1.09 $\pm$ 0.03 c	1.09 $\pm$ 0.05 e	0.41 $\pm$ 0.05 e
IAC 17	1.31 $\pm$ 0.06 c	1.29 $\pm$ 0.10 e	2.03 $\pm$ 0.18 c
BRS 1003 IPRO	2.38 $\pm$ 0.23 b	1.30 $\pm$ 0.05 e	1.19 $\pm$ 0.10 d
Dowling	0.06 $\pm$ 0.02 d	1.33 $\pm$ 0.08 e	0.95 $\pm$ 0.06 d
TMG 1188 RR	0.97 $\pm$ 0.03 c	1.35 $\pm$ 0.06 e	0.50 $\pm$ 0.05 e
FTS Campo Mourão RR	0.75 $\pm$ 0.04 c	1.40 $\pm$ 0.08 e	0.62 $\pm$ 0.07 d
NS 7007 IPRO	0.99 $\pm$ 0.04 c	1.42 $\pm$ 0.09 e	0.46 $\pm$ 0.04 e
BRS 391	0.50 $\pm$ 0.05 c	1.53 $\pm$ 0.09 e	0.73 $\pm$ 0.08 d
95R95 IPRO	1.15 $\pm$ 0.15 c	1.56 $\pm$ 0.10 e	1.63 $\pm$ 0.17 c
KS 4202	0.31 $\pm$ 0.02 d	1.69 $\pm$ 0.10 e	1.87 $\pm$ 0.12 c
PI 229358	0.11 $\pm$ 0.03 d	1.71 $\pm$ 0.16 e	2.11 $\pm$ 0.14 c
IAC 24	0.22 $\pm$ 0.03 d	1.73 $\pm$ 0.12 e	1.64 $\pm$ 0.14 c
99R09	0.91 $\pm$ 0.07 c	1.90 $\pm$ 0.19 e	0.98 $\pm$ 0.06 d
TMG 1288 RR	1.04 $\pm$ 0.09 c	1.92 $\pm$ 0.19 e	0.85 $\pm$ 0.04 d
TMG 7161 RR	2.08 $\pm$ 0.09 b	2.07 $\pm$ 0.15 e	0.80 $\pm$ 0.08 d
65I65 RSF IPRO	2.77 $\pm$ 0.24 b	2.22 $\pm$ 0.19 d	0.79 $\pm$ 0.11 d
TMG 1180 RR	0.51 $\pm$ 0.06 c	2.41 $\pm$ 0.16 d	0.76 $\pm$ 0.06 d
PI 274454	1.97 $\pm$ 0.11 b	2.68 $\pm$ 0.11 d	3.36 $\pm$ 0.19 a
DS 6217 IPRO	2.44 $\pm$ 0.13 b	2.69 $\pm$ 0.17 d	1.76 $\pm$ 0.11 c
M 7739 IPRO	1.46 $\pm$ 0.14 c	2.75 $\pm$ 0.14 d	0.34 $\pm$ 0.03 e
50I52 RSF IPRO	0.48 $\pm$ 0.02 c	2.80 $\pm$ 0.17 d	2.09 $\pm$ 0.06 c
AS 3680 IPRO	1.30 $\pm$ 0.15 c	2.89 $\pm$ 0.35 d	3.20 $\pm$ 0.14 a
TMG 2375 IPRO	0.43 $\pm$ 0.01 d	2.96 $\pm$ 0.26 d	0.93 $\pm$ 0.06 d
97R50 IPRO	0.66 $\pm$ 0.05 c	3.00 $\pm$ 0.29 d	1.54 $\pm$ 0.11 c
TMG 7262 RR	2.28 $\pm$ 0.15 b	3.55 $\pm$ 0.13 d	2.49 $\pm$ 0.18 b
IAC 19	0.22 $\pm$ 0.02 d	3.58 $\pm$ 0.24 d	1.70 $\pm$ 0.14 c
Anta 82 RR	0.09 $\pm$ 0.02 d	3.63 $\pm$ 0.25 d	1.08 $\pm$ 0.06 d
NS 6700 IPRO	0.41 $\pm$ 0.04 d	3.84 $\pm$ 0.30 d	1.88 $\pm$ 0.20 c
Conquista	0.27 $\pm$ 0.04 d	3.85 $\pm$ 0.27 d	1.20 $\pm$ 0.07 d
IAC 74-2832	0.93 $\pm$ 0.05 c	4.29 $\pm$ 0.22 c	3.74 $\pm$ 0.21 a
BRB 15-237.527	2.05 $\pm$ 0.09 b	4.32 $\pm$ 2.53 c	1.55 $\pm$ 0.18 c
IAC 78-2318	1.32 $\pm$ 0.11 c	4.90 $\pm$ 0.21 c	1.31 $\pm$ 0.13 d
Ultra BMX	3.51 $\pm$ 0.24 b	5.70 $\pm$ 0.19 c	1.97 $\pm$ 0.19 c
TMG 2378 IPRO	5.40 $\pm$ 0.49 a	7.38 $\pm$ 0.32 b	1.19 $\pm$ 0.12 d
ST 721 IPRO	4.71 $\pm$ 0.32 a	9.46 $\pm$ 0.67 a	3.36 $\pm$ 0.17 a
<i>P</i>	<0.0001	<0.0001	<0.0001

\*Means followed by the same letter in the column do not differ significantly by the contrast test ( $p > 0.05$ )

There were significant differences between treatments for the number of eggs, nymphs, and adults emerged per  $\text{cm}^2$  at 48 DAI (Table 2). The lowest oviposition rates were observed in the genotypes Dowling, Anta 82 RR, PI 229358, IAC 19, IAC 24, Conquista, KS 4202, NS 6700 IPRO, and TMG 2375 IPRO, with an average equal to or lower than 0.43 eggs/ $\text{cm}^2$ . The genotypes TMG 2378 IPRO and ST 721 IPRO showed the highest averages for oviposition (5.40 and 4.71 eggs/ $\text{cm}^2$ , respectively) and nymph colonization (7.38 and 9.46 nymphs/ $\text{cm}^2$ , respectively). The lowest nymph colonization rates were observed in the genotypes TMG 1182 RR, IAC 17, BRS 1003 IPRO, Dowling, TMG 1188 RR, FTS Campo Mourão RR, NS 7007 IPRO, BRS 391, 95R95

IPRO, KS 4202, PI 229358, IAC 24, 99R09, TMG 1288 RR, and TMG 7161 RR, with averages ranging from 1.09 to 2.07 nymphs/ $\text{cm}^2$ . Adult emergence was lower ( $\leq 0.50$  emerged adults/ $\text{cm}^2$ ) in the genotypes M 7739 IPRO, TMG 1182 RR, NS 7007 IPRO and TMG 1188 RR when compared to the others.

### No-choice Assay

The number of eggs/ $\text{cm}^2$  in the 35 soybean genotypes evaluated ranged from 0.07 to 1.77 at 48 days after infestation (DAI) (Table 3). Genotypes IAC 19, TMG 1288 RR, TMG 1182 RR, Conquista, 99R09, Dowling, NS 7007 IPRO,

**Table 3** Mean ( $\pm$  SE) number of eggs, nymphs, and emerged adults of *Bemisia tabaci* MED per cm<sup>2</sup> on 35 soybean genotypes after 48 DAI in a no-choice assay, in a greenhouse

Genotype	Eggs/cm <sup>2</sup>	Nymphs/cm <sup>2</sup>	Emerged adults/cm <sup>2</sup>
TMG 1288 RR	0.15 $\pm$ 0.02 d	1.43 $\pm$ 0.20 d	0.88 $\pm$ 0.11 e
IAC 19	0.07 $\pm$ 0.03 d	1.69 $\pm$ 0.25 d	1.71 $\pm$ 0.32 d
TMG 2375 IPRO	0.33 $\pm$ 0.02 d	1.98 $\pm$ 0.07 d	1.16 $\pm$ 0.02 d
Dowling	0.29 $\pm$ 0.04 d	2.19 $\pm$ 0.41 d	2.13 $\pm$ 0.15 c
99R09	0.26 $\pm$ 0.03 d	2.40 $\pm$ 0.30 d	1.47 $\pm$ 0.19 d
IAC 24	0.41 $\pm$ 0.05 c	2.41 $\pm$ 0.16 d	1.27 $\pm$ 0.17 d
DS 6217 IPRO	0.62 $\pm$ 0.06 c	2.52 $\pm$ 0.47 d	1.48 $\pm$ 0.29 d
TMG 1182 RR	0.23 $\pm$ 0.04 d	2.64 $\pm$ 0.06 d	1.18 $\pm$ 0.17 d
Ultra BMX	0.58 $\pm$ 0.08 c	3.10 $\pm$ 0.08 d	0.99 $\pm$ 0.09 e
IAC 78-2318	0.68 $\pm$ 0.08 c	3.31 $\pm$ 0.26 d	2.31 $\pm$ 0.22 c
M 7739 IPRO	0.81 $\pm$ 0.05 c	3.80 $\pm$ 0.59 c	1.33 $\pm$ 0.09 d
TMG 1180 RR	0.73 $\pm$ 0.11 c	3.80 $\pm$ 0.44 c	1.05 $\pm$ 0.07 e
BRS 1003 IPRO	0.67 $\pm$ 0.07 c	3.80 $\pm$ 0.35 c	1.41 $\pm$ 0.15 d
TMG 7262 RR	0.69 $\pm$ 0.03 c	4.04 $\pm$ 0.34 c	0.65 $\pm$ 0.07 e
BRS 391	0.49 $\pm$ 0.04 c	4.07 $\pm$ 0.53 c	1.63 $\pm$ 0.12 d
TMG 2378 IPRO	0.73 $\pm$ 0.04 c	4.07 $\pm$ 0.36 c	0.75 $\pm$ 0.09 e
TMG 7161 RR	0.73 $\pm$ 0.09 c	4.40 $\pm$ 0.26 c	1.58 $\pm$ 0.04 d
FTS Campo Mourão RR	0.80 $\pm$ 0.03 c	4.42 $\pm$ 0.40 c	2.02 $\pm$ 0.28 c
BRB 15-237.527	0.59 $\pm$ 0.08 c	4.49 $\pm$ 0.35 c	1.78 $\pm$ 0.17 d
Conquista	0.24 $\pm$ 0.06 d	4.54 $\pm$ 0.55 c	2.21 $\pm$ 0.13 c
95R95 IPRO	0.59 $\pm$ 0.03 c	4.55 $\pm$ 0.35 c	1.67 $\pm$ 0.08 d
97R50 IPRO	0.41 $\pm$ 0.07 c	4.56 $\pm$ 0.44 c	1.52 $\pm$ 0.16 d
NS 7007 IPRO	0.31 $\pm$ 0.08 d	4.76 $\pm$ 0.21 c	1.64 $\pm$ 0.33 d
IAC 17	0.56 $\pm$ 0.06 c	4.77 $\pm$ 0.45 c	2.73 $\pm$ 0.27 b
AS 3680 IPRO	0.47 $\pm$ 0.09 c	4.83 $\pm$ 0.24 c	1.69 $\pm$ 0.14 d
Anta 82 RR	0.57 $\pm$ 0.08 c	4.86 $\pm$ 0.44 c	2.58 $\pm$ 0.20 b
PI 274454	1.05 $\pm$ 0.09 b	4.87 $\pm$ 0.22 c	1.54 $\pm$ 0.09 d
TMG 1188 RR	1.25 $\pm$ 0.01 b	5.83 $\pm$ 0.22 b	4.21 $\pm$ 0.35 a
65165 RSF IPRO	0.97 $\pm$ 0.09 b	6.66 $\pm$ 0.57 b	1.44 $\pm$ 0.01 d
IAC 74-2832	1.69 $\pm$ 0.18 a	7.07 $\pm$ 0.44 b	2.04 $\pm$ 0.09 c
KS 4202	1.05 $\pm$ 0.07 b	7.13 $\pm$ 0.68 b	2.51 $\pm$ 0.37 b
ST 721 IPRO	1.77 $\pm$ 0.26 a	7.33 $\pm$ 0.89 b	3.19 $\pm$ 0.22 b
50I52 RSF IPRO	0.97 $\pm$ 0.05 b	7.51 $\pm$ 0.96 b	2.80 $\pm$ 0.40 b
NS 6700 IPRO	1.64 $\pm$ 0.22 a	10.10 $\pm$ 0.42 a	2.73 $\pm$ 0.07 b
PI 229358	1.06 $\pm$ 0.05 b	10.60 $\pm$ 0.68 a	3.27 $\pm$ 0.21 b
<i>P</i>	<0.0001	<0.0001	<0.0001

\*Means followed by the same letter in the column do not differ significantly by the contrast test ( $p > 0.05$ )

and TMG2375 IPRO had the lowest averages (ranging from 0.07 to 0.33 eggs/cm<sup>2</sup>). In contrast, NS 6700 IPRO, IAC74-2832, and ST 721 IPRO were the most oviposited genotypes, with an average number of eggs/cm<sup>2</sup> equal to or greater than 1.64. The number of nymphs/cm<sup>2</sup> varied from 1.43 to 3.31 among the genotypes TMG 1288 RR, IAC 19, TMG 2375 IPRO, Dowling, 99R09, IAC 24, DS 6217 IPRO, TMG 1182 RR, Ultra BMX, and IAC 78-2318, which were the least colonized. The most colonized genotypes were PI 229358 and NS 6700 IPRO (10.60 and 10.10 nymphs/cm<sup>2</sup>, respectively), followed by 50I52 RSF IPRO (7.51 nymphs/cm<sup>2</sup>), ST 721 IPRO (7.33 nymphs/cm<sup>2</sup>), KS 4202 (7.13 nymphs/cm<sup>2</sup>), IAC 74-2832

(7.07 nymphs/cm<sup>2</sup>), 65165 IPRO (6.66 nymphs/cm<sup>2</sup>), and TMG 1188 RR (5.83 nymphs/cm<sup>2</sup>). Considering the number of emerged adults/cm<sup>2</sup>, the lowest averages were obtained in the genotypes TMG 7262 RR, TMG 2378 IPRO, TMG 1288 RR, Ultra BMX, and TMG 1180 RR. The genotype TMG 1188 RR presented 4.21 emerged adults/cm<sup>2</sup>, which was the highest value among the treatments. KS 4202, Anta 82 RR, IAC 17, NS 6700 IPRO, 50I52 RSF IPRO, ST 721 IPRO and PI229358 also showed high emergence of adults and did not differ from each other.



**Table 4** Mean ( $\pm$  SE) density ( $\text{cm}^2$ ), size ( $\mu\text{m}$ ), inclination angle ( $^\circ$ ) of trichomes and colorimetric parameters ( $L^*$ ,  $a^*$  e  $b^*$ ) of 35 soybean genotypes

Genotype	Trichome			Colorimetric parameters		
	Density ( $\text{cm}^2$ ) <sup>a</sup>	Length ( $\mu\text{m}$ ) <sup>b</sup>	Angle of inclination ( $^\circ$ ) <sup>a</sup>	$L^*$ <sup>a</sup>	$a^*$ <sup>a</sup>	$b^*$ <sup>b</sup>
IAC 24	85.38 $\pm$ 6.17 e	893.37 $\pm$ 38.95 b	70.56 $\pm$ 2.06 c	38.55 $\pm$ 0.07 b	-12.53 $\pm$ 0.29 e	17.18 $\pm$ 0.38 c
Dowling	98.63 $\pm$ 5.47 e	823.50 $\pm$ 17.39 c	72.05 $\pm$ 3.37 c	38.83 $\pm$ 0.61 b	-14.91 $\pm$ 0.45 c	22.27 $\pm$ 0.26 b
PI 229358	114.88 $\pm$ 3.61 e	1096.32 $\pm$ 43.62 a	65.44 $\pm$ 1.98 c	38.98 $\pm$ 0.26 b	-14.61 $\pm$ 0.29 c	20.93 $\pm$ 0.48 b
TMG 1288 RR	115.50 $\pm$ 7.72 e	710.89 $\pm$ 28.07 d	67.43 $\pm$ 2.95 c	37.69 $\pm$ 0.36 c	-15.61 $\pm$ 0.12 b	21.70 $\pm$ 0.28 b
Anta 82 RR	135.38 $\pm$ 8.23 d	996.56 $\pm$ 25.38 a	77.96 $\pm$ 1.64 b	42.18 $\pm$ 0.20 a	-16.00 $\pm$ 0.09 b	24.93 $\pm$ 0.17 a
PI 274454	139.75 $\pm$ 9.26 d	812.75 $\pm$ 18.72 c	58.37 $\pm$ 2.55 d	40.99 $\pm$ 0.42 a	-15.37 $\pm$ 0.23 c	22.04 $\pm$ 0.26 b
TMG 1182 RR	141.00 $\pm$ 10.72 d	821.96 $\pm$ 41.88 c	66.52 $\pm$ 3.34 c	36.90 $\pm$ 0.17 c	-12.27 $\pm$ 0.22 e	17.99 $\pm$ 0.15 c
97R50 IPRO	154.25 $\pm$ 9.04 d	898.45 $\pm$ 30.87 b	79.54 $\pm$ 2.08 b	39.53 $\pm$ 0.45 b	-15.08 $\pm$ 0.25 c	21.14 $\pm$ 0.33 b
TMG 2375 IPRO	159.88 $\pm$ 4.56 d	984.30 $\pm$ 38.77 a	70.59 $\pm$ 1.62 c	38.25 $\pm$ 0.28 c	-13.86 $\pm$ 0.17 d	19.49 $\pm$ 0.29 b
TMG 2378 IPRO	163.00 $\pm$ 6.91 d	859.10 $\pm$ 31.40 c	78.55 $\pm$ 1.49 b	38.19 $\pm$ 0.21 c	-13.47 $\pm$ 0.17 d	20.04 $\pm$ 0.30 b
TMG 1188 RR	167.75 $\pm$ 8.65 d	1025.7 $\pm$ 28.99 a	62.84 $\pm$ 1.61 c	41.89 $\pm$ 0.22 a	-16.11 $\pm$ 0.24 b	25.88 $\pm$ 0.62 a
IAC 17	170.50 $\pm$ 6.42 d	971.75 $\pm$ 19.18 b	55.11 $\pm$ 1.59 d	39.13 $\pm$ 0.27 b	-13.80 $\pm$ 0.13 d	20.59 $\pm$ 0.31 b
AS 3680 IPRO	179.88 $\pm$ 9.61 c	815.36 $\pm$ 32.41 c	75.22 $\pm$ 1.54 b	37.26 $\pm$ 0.33 c	-13.41 $\pm$ 0.22 d	20.39 $\pm$ 0.34 b
50I52 RSF IPRO	180.63 $\pm$ 3.94 c	1078.76 $\pm$ 32.74 a	72.51 $\pm$ 1.77 c	38.78 $\pm$ 0.45 b	-13.87 $\pm$ 0.50 d	21.17 $\pm$ 0.39 b
BRS 391	181.38 $\pm$ 11.91 c	882.18 $\pm$ 30.78 c	62.96 $\pm$ 1.49 c	38.95 $\pm$ 0.50 b	-13.27 $\pm$ 0.29 d	20.15 $\pm$ 0.39 b
Conquista	183.63 $\pm$ 10.50 c	904.74 $\pm$ 33.51 b	64.05 $\pm$ 4.48 c	36.93 $\pm$ 0.45 c	-11.99 $\pm$ 0.32 e	17.67 $\pm$ 0.41 c
65I65 RSF IPRO	192.50 $\pm$ 5.45 c	902.59 $\pm$ 40.79 b	74.00 $\pm$ 0.87 b	39.67 $\pm$ 0.28 b	-15.30 $\pm$ 0.08 c	21.87 $\pm$ 0.19 b
TMG 1180 RR	210.88 $\pm$ 14.65 c	736.43 $\pm$ 34.32 d	66.94 $\pm$ 2.28 c	41.27 $\pm$ 0.28 a	-16.89 $\pm$ 0.15 a	23.87 $\pm$ 0.39 a
KS 4202	213.63 $\pm$ 11.49 c	824.40 $\pm$ 22.43 c	75.63 $\pm$ 1.86 b	37.08 $\pm$ 0.35 c	-12.89 $\pm$ 0.28 d	18.74 $\pm$ 0.39 c
BRB 15-257.324	218.50 $\pm$ 7.98 c	693.93 $\pm$ 30.00 d	73.08 $\pm$ 1.64 c	38.02 $\pm$ 0.23 c	-14.22 $\pm$ 0.13 d	19.85 $\pm$ 0.23 b
IAC 19	221.25 $\pm$ 7.80 c	991.00 $\pm$ 17.87 a	64.75 $\pm$ 1.65 c	38.77 $\pm$ 0.24 b	-14.67 $\pm$ 0.20 c	21.01 $\pm$ 0.37 b
TMG 7262 RR	221.50 $\pm$ 11.13 c	867.15 $\pm$ 36.48 c	77.59 $\pm$ 2.01 b	36.24 $\pm$ 0.33 c	-12.33 $\pm$ 0.24 e	16.68 $\pm$ 0.28 c
IAC 78-2318	228.00 $\pm$ 12.94 c	795.33 $\pm$ 48.65 c	69.42 $\pm$ 2.22 c	38.14 $\pm$ 0.37 c	-13.71 $\pm$ 0.13 d	19.74 $\pm$ 0.37 b
TMG 7161 RR	235.75 $\pm$ 10.17 c	855.16 $\pm$ 24.09 c	64.12 $\pm$ 1.49 c	42.48 $\pm$ 0.31 a	-16.34 $\pm$ 0.12 a	25.28 $\pm$ 0.32 a
NS 6700 IPRO	240.63 $\pm$ 9.81 b	906.92 $\pm$ 35.45 b	70.66 $\pm$ 2.33 c	39.02 $\pm$ 0.24 b	-14.15 $\pm$ 0.19 d	19.38 $\pm$ 0.40 b
95R95 IPRO	241.75 $\pm$ 12.24 b	929.62 $\pm$ 46.00 b	86.06 $\pm$ 1.19 a	40.25 $\pm$ 0.32 b	-15.61 $\pm$ 0.16 b	23.41 $\pm$ 0.42 a
ST 721 IPRO	254.50 $\pm$ 9.57 b	676.64 $\pm$ 34.41 d	67.87 $\pm$ 1.03 c	37.78 $\pm$ 0.43 c	-12.70 $\pm$ 0.30 d	17.90 $\pm$ 0.33 c
DS 6217 IPRO	255.75 $\pm$ 11.40 b	953.52 $\pm$ 69.78 b	72.83 $\pm$ 1.90 c	38.76 $\pm$ 0.25 b	-13.08 $\pm$ 0.25 d	18.95 $\pm$ 0.46 c
M 7739 IPRO	261.50 $\pm$ 12.80 b	845.30 $\pm$ 33.57 c	67.78 $\pm$ 3.09 c	37.41 $\pm$ 0.52 c	-14.39 $\pm$ 0.29 c	19.35 $\pm$ 0.67 b
NS 7007 IPRO	265.88 $\pm$ 14.16 b	985.53 $\pm$ 15.43 a	67.50 $\pm$ 2.03 c	39.12 $\pm$ 0.24 b	-14.56 $\pm$ 0.17 c	21.70 $\pm$ 0.41 b
IAC 74-2832	267.50 $\pm$ 13.03 b	705.82 $\pm$ 17.89 d	42.97 $\pm$ 1.10 e	40.09 $\pm$ 0.18 b	-14.90 $\pm$ 0.20 c	21.33 $\pm$ 0.46 b
99R09	270.50 $\pm$ 8.74 b	1028.41 $\pm$ 59.30 a	66.46 $\pm$ 3.68 c	36.33 $\pm$ 0.28 c	-12.25 $\pm$ 0.28 e	18.23 $\pm$ 0.35 c
Ultra BMX	292.25 $\pm$ 7.98 b	1063.48 $\pm$ 56.02 a	67.08 $\pm$ 1.61 c	38.92 $\pm$ 0.41 b	-13.42 $\pm$ 0.23 d	19.18 $\pm$ 0.41 b
FTS Campo Mourão	331.88 $\pm$ 12.89 a	793.99 $\pm$ 32.07 c	56.03 $\pm$ 1.19 d	39.90 $\pm$ 0.49 b	-15.15 $\pm$ 0.34 c	23.20 $\pm$ 0.32 a
BRS 1003 IPRO	349.88 $\pm$ 11.86 a	939.97 $\pm$ 40.51 b	69.26 $\pm$ 1.39 c	36.93 $\pm$ 0.66 c	-13.99 $\pm$ 0.17 d	19.15 $\pm$ 0.41 b
<i>P</i>	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

<sup>a</sup> Means followed by the same letter in the column do not differ significantly by the contrast test ( $p > 0.05$ )

<sup>b</sup> Means followed by the same letter in the column do not differ significantly by the Scott-Knott test ( $p > 0.05$ )

## Trichome Analysis

The trichome density ranged from 85.38 to 349.88/cm<sup>2</sup> among the genotypes. The highest trichome densities were observed in the genotypes BRS 1003 IPRO and FTS Campo Mourão (349.88 and 331.88/cm<sup>2</sup>, respectively) (Table 4). IAC 24, Dowling, PI 229358, and TMG 1288 RR exhibited the lowest trichome means per cm<sup>2</sup> (Fig. 2), with values varying from 85.38 to 115.50. Shorter trichomes were observed in the genotypes ST 721 IPRO, BRB 15-257.327,

IAC 74-2832, TMG 1288 RR, and TMG 1180 RR (676.64 to 736.43  $\mu\text{m}$ ). Whereas the genotypes TMG 2375 IPRO, NS 7007 IPRO, IAC 19, Anta 82 RR, TMG 1188 RR, 99R09, Ultra BMX, 50I52 RSF IPRO, and PI 229358 have longer trichomes than the other treatments ( $\geq 984.30 \mu\text{m}$ ). The inclination of the trichomes varied between 42.97 and 86.06°. The inclination of the trichomes varied between 42.97 and 86.06°, with the genotype IAC 74-2832 having more inclined trichomes compared to 95R95 IPRO, which had more erect trichomes.

**Table 5** Spearman or Pearson<sup>a</sup> correlation coefficients ( $r$ ) and probabilities ( $P$ ) between *Bemisia tabaci* MED parameters and trichome characteristics and colorimetric parameters of soybean genotypes

Parameter	Coef- ficient	Adults	Trichome			Colorimetric parameters		
			Density	Length ( $\mu\text{m}$ )	Angle of incli- nation ( $^\circ$ )	L*	a*	b*
Adults	$r$	–	0.20	–0.03 <sup>a</sup>	0.06	–0.21	–0.25 <sup>a</sup>	–0.25
	$P$	–	0.0006	0.58 <sup>a</sup>	0.33	0.0003	<0.0001 <sup>a</sup>	<0.0001
Number of eggs/cm <sup>2</sup>	$r$	0.40	0.37	–0.06	0.07	–	–	–
	$P$	<0.0001	<0.0001	0.33	0.23	–	–	–
Number of nymphs/cm <sup>2</sup>	$r$	0.38	0.13	–0.01	0.15	–	–	–
	$P$	<0.0001	0.03	0.83	0.01	–	–	–
Number of emerged adults/cm <sup>2</sup>	$r$	0.37	0.01	–0.04	0.06	–	–	–
	$P$	<0.0001	0.89	0.46	0.29	–	–	–

### Colorimetric Analysis

The luminosity (L\*) varied between the materials analyzed, with L\* values varying between 36.24 and 42.48 (Table 4). The genotypes PI 274454, TMG 1180 RR, TMG 1188 RR, Anta 82 RR and TMG 7161 RR exhibited higher luminosity. The smallest green color intensities (a\*) were observed for TMG 1180 RR and TMG 7161 RR (–16.89 and –16.34, respectively), followed by TMG 1188 RR, Anta 82 RR, TMG1288 RR and 95R95 IPRO. On the other hand, the genotypes Conquista, 99R09, TMG 1182 RR, TMG 7262 RR and IAC 24 showed higher intensity for this parameter. The genotypes FTS Campo Mourão, 95R95 IPRO, TMG 1180 RR, Anta 82 RR, TMG 7161 RR and TMG 1188 RR expressed greater yellow color intensity (b\*) (23.20–25.88), while TMG 7262 RR, IAC 24, Conquista, ST 721 IPRO, TMG 1182 RR, 99R09, KS 4202 and DS 6217 IPRO had the lowest averages for this parameter (ranging from 16.68 to 18.95).

### Correlations

According to the calculated coefficients ( $r$ ), significant correlations were identified among some of the studied interactions (Table 5). A positive correlation was observed between the number of adults per leaflet and the number of eggs ( $r=0.40$ ;  $P<0.0001$ ), nymphs ( $r=0.38$ ;  $P<0.0001$ ), and emerged adults/cm<sup>2</sup> ( $r=0.37$ ;  $P<0.0001$ ) in the multiple-choice test. Additionally, the number of adults per leaflet exhibited a positive correlation with trichome density ( $r=0.37$ ;  $P=0.0006$ ) and a negative correlation with the color parameters L\* ( $r=-0.21$ ;  $P<0.0003$ ), a\* ( $r=-0.25$ ;  $P<0.0001$ ), and b\* ( $r=-0.25$ ;  $P<0.0001$ ). Positive correlations were found between the number of eggs ( $r=0.37$ ;  $P<0.0001$ ) and nymphs/cm<sup>2</sup> ( $r=0.13$ ;  $P=0.03$ ) and trichome density.

### Discussion

*Bemisia tabaci* MED was recently introduced in Brazil, and its occurrence in soybean-producing areas represents a new threat to this crop (Barbosa et al. 2015; Bello et al. 2021). The use of resistant plants can help in the management of this pest. This management strategy is highly effective in controlling arthropod pests due to its easy adoption, persistence, specificity, cumulative effect, low cost, and compatibility with other control methods, in addition to being less aggressive to the environment and producers (Baldin et al. 2019). No-choice assays with 90 soybean genotypes demonstrated different levels of infestation by *B. tabaci* MED. The variation in the host plant colonization process may occur due to the expression of chemical, physical and morphological factors that may limit the feeding and development of arthropod pests (Powell et al. 2006; War et al. 2012).

Preference and colonization by *B. tabaci* MED were investigated in multiple-choice and no-choice, using the most resistant and most susceptible genotypes initially selected in a screening test. In the multiple-choice test, the genotypes KS 4202, TMG 1188 RR, M 7739 IPRO, 65165 IPRO, and PI 229358 were less preferred by *B. tabaci* MED adults, revealing possible expression of antixenosis. Host plant selection by insects is a complex process in which visual, olfactory, and tactile stimuli are involved. Guided by volatile compounds or the color of the substrate, for example, insects can be attracted towards the host, starting or not feeding and colonization (Schoonhoven et al. 2005).

In this study, negative correlations were observed between color parameters (L\*, a\*, b\*) and the preference of adult insects for soybean genotypes. The genotypes that expressed lower luminosity (L\*) and lower intensity of green (a\*) and yellow (b\*) were the most attractive to *B. tabaci* MED. Contrasting results were observed in studies with the cryptic species *B. tabaci* MEAM1 regarding the intensity of green. Bean genotypes that exhibit a higher intensity of

green color were more attractive to this species (Santos et al. 2020). Similarly, the establishment of MEAM1 was positively correlated with higher levels of green in eggplant and cotton genotypes (Hasanuzzaman et al. 2016; Prado et al. 2016). On the other hand, Santos et al. (2023) observed that the preference of *B. tabaci* MED for tomato genotypes was not correlated with the coloration of the plants.

The genotypes Dowling, PI 229358, KS 4202, and IAC 24 exhibited low colonization by eggs and nymphs after 48 DAI in a multiple-choice test. Furthermore, the genotypes TMG 1182 RR, IAC 17, BRS 1003 IPRO, TMG 1188 RR, FTS Campo Mourão RR, NS 7007 IPRO, BRS 391, 95R95 IPRO, KS 4202, 99R09, TMG 1288 RR, and TMG 7161 RR also showed a low number of nymphs. These results suggest a possible expression of resistance through antixenosis and/or antibiosis in these genotypes. In addition to the low occurrence of nymphs, the genotypes TMG 1188 RR and TMG 1288 RR exhibited low emergence of adults, further supporting the hypothesis of the occurrence of antibiosis.

Morphological and chemical characteristics can have a significant impact on the colonization process of host plants by whiteflies (Li et al. 2023). Hairiness, for example, is one of the factors related to plant resistance to insects (Baldin et al. 2017; Santos et al. 2020, 2023). In this study, it was possible to observe that the high trichome density in soybean is related to a higher preference for adults of *B. tabaci* MED and, consequently, a higher number of eggs and nymphs/cm<sup>2</sup>. Similar results were observed for *B. tabaci* MEAM1, indicating that this leaf characteristic is an important factor for the establishment of different cryptic species of *B. tabaci* (Valle Do and Lourenção 2002; Vieira et al. 2011). In fact, the high density of trichomes can favor the process of insect colonization in plants, as, in addition to possibly creating a microclimate favorable to oviposition, trichomes can assist insects to remain fixed on plants in unfavorable climatic conditions, such as intense rain and winds, or even hinder the action of natural enemies (Li et al. 1987; Butter and Vir 1989; Valle Do and Lourenção 2002; Vieira et al. 2011).

In the no-choice test, the genotypes TMG 1288 RR, IAC 19, TMG 2375 IPRO, Dowling, 99R09 and TMG 1182 RR stood out for presenting a low number of eggs and nymphs. Furthermore, similar to the multiple-choice test, IAC 24 is among the genotypes with the lowest colonization by nymphs in the no-choice test. Based on the assays conducted in this study, it is noted that some soybean genotypes allow low or high colonization by *B. tabaci* MED in both evaluation conditions (with and without choice), confirming their susceptibility or expression of antixenosis and/or antibiosis. This is the case of the genotypes ST 721 IPRO, IAC 742832 and NS 6700 IPRO, which present high colonization by eggs, nymphs and/or high emergence of

adults, being among the most susceptible genotypes among the 35 evaluated. On the other hand, the genotypes IAC 24, IAC 19, Dowling, 99R09, TMG 1182 RR, TMG 1288 RR, TMG 2375 IPRO generally showed low numbers of insects per cm<sup>2</sup>, being considered resistant due to antixenosis and/or antibiosis.

Several studies report the resistance of the genotypes IAC 24 and IAC 19 to a wide range of insects. For *B. tabaci* MEAM1, for example, both genotypes caused a reduction of more than 50% in adult emergence (Vieira et al. 2016), in addition, IAC 24 presented low attractiveness and a reduced number of eggs for this pest (Valle et al. 2012). In other studies, the expression of resistance by IAC 24 was reported for *Anticarsia gemmatalis* (Fugi et al. 2005; Ongaratto et al. 2021). Both genotypes also negatively affected the development, larval viability, and pupal weight of *A. gemmatalis* (Ongaratto et al. 2021). Other examples are found in the literature, with results revealing the resistance of the IAC 24 and/or IAC 19 genotypes to *Helicoverpa armigera*, *Dichelops melacanthus*, *Euschistus heros*, *Piezodorus guildinii*, among other insect pests (Silva et al. 2014; Canassa et al. 2017; Souza et al. 2017; Coelho et al. 2020). Here, IAC 24 and IAC 19 were less preferred or colonized by *B. tabaci* MED, corroborating the results obtained in previous studies.

The genotypes IAC 17, TMG 1188 RR, KS 4202 and PI 229358 showed different patterns of colonization between the multiple-choice and no-choice tests. When insects were allowed to choose, these genotypes were less preferred and/or colonized by *B. tabaci* MED. However, the opposite was observed in the no-choice assay, where these genotypes were revealed as potential hosts for *B. tabaci* MED. In order to manage this pest, results such as these must be taken into consideration, since confinement trials, where there is no opportunity for choice, more closely represent the real field situation, particularly in large-scale monoculture areas of the host plant (Baldin et al. 2019).

As expected, positive correlations were observed between the number of adults per leaflet and the number of eggs, nymphs, and emerged adults per cm<sup>2</sup> in the multiple-choice test. However, this pattern was not observed for the genotype BRS 1003 IPRO. Although this genotype is among the most preferred by *B. tabaci* MED, the high number of adults per leaflet did not result in high colonization by nymphs after 48 DAI. Results of this type may occur due to the expression of antibiosis, resulting in greater mortality of nymphs in this material. The genotype BRS 1003 IPRO was developed with the Block technology, which, despite not affecting nymphal development or the survival of stink bug nymphs and adults, (Oliveira et al. 2022), confers tolerance to the attack of these insects on soybean pods, allowing high productivity even under high infestations of this group of pests (Lucini et al. 2021; Oliveira et al. 2022). However,

in the present study, BRS 1003 IPRO showed resistance to MED due to the possible expression of antibiosis.

The results presented here reveal the resistance of soybean genotypes to *Bemisia tabaci* MED. Among the genotypes that showed higher resistance to *B. tabaci* MED are 99R09, TMG 1182 RR, TMG 1288 RR, and TMG 2375 IPRO, which are commercially available to farmers. These materials can be integrated into other management strategies in areas where there is a high incidence of *Bemisia tabaci* MED, aiding in the maintenance of pest populations below the economic damage threshold.

## Conclusion

The preference and colonization of soybean genotypes by *B. tabaci* MED may be attributed to factors such as leaf color, trichome density, and chemical substances. The genotypes IAC 24, IAC 19, Dowling, 99R09, TMG 1182 RR, TMG 1288 RR, TMG 2375 IPRO, and BRS 1003 IPRO exhibited resistance due to antixenosis and/or antibiosis. Genotypes that are less preferred and colonized are considered as sources of resistance and may be useful in genetic breeding programs with the aim of developing varieties more resistant to *B. tabaci* MED. Cultivars with good productivity and carrying adequate levels of resistance can be recommended for planting in areas with infestations of this insect.

**Funding** This study was funded by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior—Brasil (CAPES)—Finance Code 001 and CNPq (Process n. 140918/2020-5).

## Declarations

**Conflict of interest** A.P. Santana Lima, E.L. Lopes Baldin, T.L. Braga dos Santos, A. da Silva Santana, I. Rubio Cabral, A. Marques Pinheiro, R. Krause Sakate and A.L. Lourenção declare that they have no competing interests.

**Ethical standards** For this article no studies with human participants or animals were performed by any of the authors.

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