Phytopathology/ Original Article

Pathogenicity of novel *Monosporascus* species in accessions of melon varietal groups

Abstract – The objective of this work was to evaluate the pathogenicity of the following *Monosporascus* species: *Monosporascus brasiliensis*, *Monosporascus caatinguensis*, *Monosporascus mossoroensis*, *Monosporascus nordestinus*, and *Monosporascus semiaridus* in comparison with *Monosporascus cannonballus*, as well as the reaction to these pathogens of the A-16, C-32, 'Goldex', and 'Hales Best Jumbo' melon accessions, which belong to the acidulus, cantalupensis, conomon, and inodorus varietal groups, respectively. Vine decline severity was evaluated based on root damage and on the root dry matter reduction index. All studied *Monosporascus* species caused damage to the melon accessions, but only *M. brasiliensis*, *M. nordestinus*, and especialy *M. caatinguensis* were considered virulent. The A-16 accession shows higher resistance to *M. nordestinus*, *M. caatinguensis*, and *M. cannonballus*, whereas 'Goldex' presents susceptibility to *M. caatinguensis*, *M. nordestinus*, and *M. semiaridus*. The *M. brasiliensis*, *M. caatinguensis*, *M. mossoroensis*, *M. nordestinus*, and *M. semiaridus* species present varying levels of pathogenicity and diferent levels of infection severity, with *M. semiaridus* having the highest severity, and *M. cannonballus*, the lowest.

Index terms: *Cucumis melo*, germplasm, vine decline, virulence.

Patogenicidade de novas espécies de *Monosporascus* em acessos de grupos varietais de melão

Resumo – O objetivo deste trabalho foi avaliar a patogenicidade das seguintes espécies de *Monosporascus: Monosporascus brasiliensis*, *Monosporascus caatinguensis*, *Monosporascus mossoroensis*, *Monosporascus nordestinus* e *Monosporascus semiaridus*, em comparação com *Monosporascus cannonballus*, bem como a reação a esses patógenos dos acessos de melão A-16, C-32, 'Goldex' e 'Hales Best Jumbo', os quais pertencem aos grupos varietais acidulus, cantalupensis, conomon e inodorus, respectivamente. A severidade do declínio das ramas foi avaliada com base nos danos às raízes e no índice de redução da matéria seca das raízes. Todas as espécies de *Monosporascus* estudadas causaram danos aos acessos de melão, mas apenas *M. brasiliensis*, *M. nordestinus* e, especialmente**,** *M. caatinguensis* foram consideradas virulentas. O acesso A-16 apresenta maior resistência a *M. nordestinus***,** *M. caatinguensis* e *M. cannonballus*, enquanto 'Goldex' apresenta susceptibilidade a *M. caatinguensis*, *M. nordestinus* e *M. semiaridus*. As espécies *M. brasiliensis*, *M. caatinguensis*, *M. mossoroensis*, *M. nordestinus* e *M. semiaridus* apresentam variados níveis de patogenicidade e diferentes níveis de severidade da infecção, com *M. semiaridus* tendo a maior severidade, e *M. cannonballus* a menor.

Termos para indexação: *Cucumis melo*, germoplasma, declínio das ramas, virulência.

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Introduction

The melon vine decline is a complex cucurbit syndrome caused by several species of soil-borne fungi, including *Monosporascus cannonballus* Pollack & Uecker, *Acremonium cucurbitacearum* Alfaro-García, W. Gams & García-Jim. 1996, *Plectosporium tabacinum* (J.F.H. Beyma) M.E. Palm, W. Gams & Nirenberg 1995, and *Rhizopycnis vaga* D.F. Farr [as 'vagum'], in Farr, Miller & Bruton (Chilosi et al., 2008). Symptoms typically appear at the fruit maturity stage, which include initial yellowing and deterioration of the vine leaves, progressive defoliation, and partial or total collapse of the vines (Picó et al., 2008; Cluck et al., 2009). This disease has been causing many losses around the world, especially in melon (*Cucumis melo* L.) and watermelon [*Citrullus lanatus* (Thunb.) Matsum. & Nakai] crops (Sales Jr et al., 2003, 2010; Al-Mawaali et al., 2013; Yan et al., 2016; Markakis et al., 2018; Negreiros et al., 2019; Sales Júnior et al., 2019).

Five novel *Monosporascus* species associated with the roots of *Boerhavia diffusa* L. and *Trianthema portulacastrum* Linn. were described in the Brazilian Northeastern region, namely *Monosporascus brasiliensis*, *Monosporascus caatinguensis*, *Monosporascus mossoroensis*, *Monosporascus nordestinus*, and *Monosporascus semiaridus*, as described by Negreiros et al. (2019), posing potential threats to cucurbit crops (Cavalcante et al., 2020) and non-cucurbit crops, such as cowpea, jack bean, corn, sorghum, and bell pepper (Tavares et al., 2023). Althogh this information has been reiterated, the specific impact of these five new species on melon crops remains largely unexplored, requiring studies to assess the reaction of melon crops to *Monosporascus* species in Brazil, in order to develop resistant cultivars.

The objective of this work was to evaluate the pathogenicity of the following *Monosporascus* species: *M. brasiliensis*, *M. caatinguensis*, *M. mossoroensis*, *M. nordestinus*, and *M. semiaridus* in comparison with *M. cannonballus*, as well as the reaction to these pathogens of the A-16, C-32, 'Goldex', and 'Hales Best Jumbo' melon accessions, belonging to acidulus, cantalupensis, conomon, and inodorus varietal groups, respectively.

Materials and Methods

The research was conducted in a greenhouse located on the campus of Universidade Federal Rural do Semi-Árido (UFERSA), in the municipality of Mossoró, state of Rio Grande do Norte, Brazil. Mossoró has a dry, very hot climate, classified as BSwh' according to the Köppen classification. The rainy season extends from summer to autumn, and the rainiest months are February, March, and Abril, with an average temperature of 27.5°C, an average annual rainfall of 670 mm, and an average relative humidity of 68.9%. The experimental period was from August 5, 2019 to September 23, 2019, during which the maximum and minimum temperatures were 34.4°C and 19°C, respectively, with an average of 26.7°C.

The melon genotypes evaluated included two commercial cultivars: Hales Best Jumbo (HBJ) and Goldex, as well as two accessions: A-16, C-32 syn. Pat 81, from the cucurbit germplasm bank at UFERSA. 'HBJ', 'Goldex', A-16, and C-32 belong to cantalupensis, inodorus, acidulous, and conomon varietal groups, respectively.

The isolates of *Monosporascus* spp. used were *M. brasiliensis* (CMM 4839), *M. caatinguensis* (CMM 4833), *M. mossoroensis* (CMM 4857), *M. nordestinus* (CMM 4846), *M. semiaridus* (CMM 4830), and *M. cannonballus* (CMM 2386), which were obtained from Profa. Maria Menezes Culture Collection of Phytopathogenic Fungi (CMM) at Universidade Federal Rural de Pernambuco (UFRPE).

The experiment followed a completely randomized design with five replicates, in a factorial arrangement of four melon acessions × six *Monosporascus* species, resulting in 24 treatments or pathosystems.

The seeds of the melon accessions were sown in trays with substrate autoclaved two times at 120°C with a 24-hour interval, composed of a 2:1 ratio of sand and Topstrato HT Hortaliças (Vida Verde, Mogi Mirim, SP, Brazil), which was also used to fill 500 mL pots for plant cultivation.

The seeds of the accessions from the Cucurbit Germplasm Bank of UFERSA were set to germinate in non-inoculated Topstrato. To produce the inocula, the mycelia of each *Monosporascus* species was multiplied in separate Petri dishes in sterilized potato dextrose agar (PDA) medium, using an adapted method by Ben Salem et al. (2015). The culture media containing the mycelia from each Petri dish was diluted in 300 mL

of sterile distilled water and mixed using a blender. A 10 mL mycelial suspension of each *Monosporascus* species was inoculated into the substrate compound by 2:1 ratio of sand and Topstrato, which was incubated for seven days; then the seedlings of the melon accessions with 15 days old were transplanted to 500 mL pots, kept in a greenhouse, and irrigated daily.

The evaluations were performed 50 days after transplanting. The aerial part of the plants was removed, the roots were kept, and the substrate was washed off from the roots with tap water. The analyzed variables were: vine decline severity, using a rating scale proposed by Armengol et al. (1999), and root dry matter. The rating scale ranged from 0 to 4, in which: 0 means no symptoms; 1, mild discoloration, or <10% root decay; 2, moderate discoloration, or 25–35% root decay; 3, death of secondary roots, or 50% root decay; and 4, total root necrosis, or plant death. The average reaction was calculated by summing the scores of each genotype and dividing them by the total number of evaluated plants. The following genotype classes were: 0, for similar to immune; 0.1–1.0, for highly resistant; 1.1–2.0, for moderately resistant; 2.1–3.0, for susceptible; and 3.1–4.0, for highly susceptible (Sales Júnior et al., 2019).

As the severity variable did not have normally distributed residuals, the original values were transformed using the Aligned Rank Transformed (ART) method for non-parametric factorial analyses (Wobbrock et al., 2011). Analysis of variance was performed for the severity rank variable and for the root dry matter reduction index ($RI_{(DM)}$), calculated using the following equation: $RI_{(DM)} = (DM_u - DM_i)/(DM_u)$, where DM_u is the dry matter of the uninoculated access; and DM_i, the dry matter of the access inoculated with one of the species. The Scott-Knott's test, at 5% probability, was used for grouping the means of the accessions and species. The Spearman correlation coefficient was estimated to assess the association between severity and dry matter index.

The GGE Biplot method, proposed by Yan & Kang (2003), assesses the non-additive effects, specifically interactions, between pathogens and plant hosts. The GGE Biplot model considers multiplicative effect, this is, the effect of the genotype and the genotype-species interaction, without separating them, which is described as: $Y_{ij} - \mu - \alpha_i - \beta_j = g_{i1}e_{i1} + g_{i2}e_{i1} + e_{ij}$, where Y_{ij} is the performance of genotype i in species j; μ is

the overall mean of the observations; α_i is the main effect of genotype i; β_j is the main effect of species j; g_{i1} and e_{i1} are the principal scores of genotype i and species j, respectively; g_{i2} and e_{i1} are the secondary scores of genotype i and species j, respectively; e_{ij} is the residual that cannot be explained by both effects.

All statistical analyses were performed using the R software (R Core Team, 2020).

Results and Discussion

Althouth the results did not show differences in the effect of melon accessions to the *Monosporascus* species studied, it was found an effect on the melon accessions response to the infection. This result is due to the diversity of plant materials assessed for severity and the influence of the accession-species interaction, which resulted in varied responses of the accessions to the six inoculated *Monosporascus* species.

The accessions were grouped according to the average ranks of infection severity of melon accessions to *Monosporascus* spp. inoculation (Table 1). Two groups were formed with the *M. brasiliensis* inoculation: the first was composed by the A-16 and 'HBJ' accessions, representing the highest average ranks of severity; and the second one consisted of C-32 and 'Goldex'. Two other groups of accessions were composed by *M. caatinguensis* and *M. nordestinus* inoculation, in which the A-16 accession presented the lowest average rank of severity.

A-16 was the most resistant to the *Monosporascus* species evaluated. The accessions composed a single group with *M. cannonballus*, *M. mossoroensis*, and *M. semiaridus* inoculation. When inoculated in A-16, the fungal species grouped into two categories (Table 1): the first group comprised *M. brasiliensis*, *M. mossoroensis*, and *M. semiaridus*, showing higher aggressiveness. In 'HBJ', the species were divided into a primary group consisting of the most aggressive ones: *M. brasiliensis* and *M. nordestinus*. Subsequently, the remaining species formed the second group. However, when pathogens were inoculated in C-32 and 'Goldex', they formed a single group (Table 1).

In a related study on the interaction of 'Titanium' with *Monosporascus* spp., symptoms of vine decline were noted, confirming their pathogenicity to melons (Cavalcante et al., 2020). 'Titanium' displayed susceptibility, with disease severity ranging from 1.2

to 1.6%. However, the authors found no significant variations in terms of disease severity and reductions in fresh and dry root weights (Cavalcante et al., 2020).

According Marquez et al. (2023) the susceptibility of 'TAM-Uvalde' to *M. cannonballus* is linked to the production of hydroxyproline and gamma-aminobutyric acid (GABA), serving as susceptibility markers for vine decline. In contrast, the resistant genotype, USDA PI 124104, exhibited elevated levels of the amino acids glycine and glutamine after the inoculation, consequently, these amino acids could potentially serve as resistance markers for vine decline.

All accessions studied in this work presented moderate or total resistance to *M. cannonballus* (Table 2), which is among the most frequently isolated in melon roots in the Brazilian semiarid region, along with *Macrophomina phaseolina* (Tassi) Goid. and *Fusarium solani* (Mart.) Sacc. (Ambrósio et al., 2015), whose literature on resistance is lacking.

The accessions were classified according to the reaction severity to the six inoculated species (Table 2). A-16 was highly resistant to *M. nordestinus*, *M. caatinguensis*, and *M. cannonballus*, moderately resistant to *M. mossoroensis* and *M. semiaridus*, but susceptible to *M. brasiliensis*. C-32 was susceptible to *M. caatinguensis*, but moderately resistant to the other species. 'Goldex' was susceptible to *M. caatinguensis*, *M. nordestinus*, and *M. semiaridus*, and moderately resistant to *M. brasiliensis*, *M. cannonballus*, and *M.*

mossoroensis. 'HBJ' was susceptible to *M. brasiliensis* and *M. nordestinus* and moderately resistant to *M. cannonballus*, *M. caatinguensis*, *M. mossoroensis*, and *M. semiaridus*.

In one of the first efforts to identify sources of resistance to *M. cannonballus*, Mertely et al. (1993) concluded that 'HBJ', 'Honey Dew Green Flesh', 'Improved', 'Cruiser', 'Durango', PI 12411, and 'Laredo' were tolerant to this fungal species. However, 'HBJ' was susceptible to *M. brasiliensis* and *M. nordestinus.*

In Spain, under field conditions and through greenhouse artificial inoculation, the accession Pat 81 (*C. melo* ssp. *agrestis*) presented a high level of tolerance (Iglesias & Nuez, 1998; Iglesias et al., 2000), playing an important role in a breeding program involving backcrosses and leading to the development of resistant lines in the piel de sapo group (Fita et al., 2009). The accession C-32, derived from Pat 81, presented susceptibility to *M. caatinguensis* and moderate resistance to the other species. Therefore, C-32 can be a source of resistance; however, for the first time, it presented susceptibility to a *Monosporascus* species.

'Goldex' was susceptible to *M. caatinguensis*, *M. nordestinus*, and *M. semiaridus* (Table 2), presented high values of root dry matter reduction index (Table 3), and is highly susceptible to powdery mildew [*Podosphaera xanthii* (Castagne) U. Braun & Shishkoff] and leafminer (*Liriomyza sativae*

Table 1. Scott-Knott's test to average ranks and original averages of the severity of melon (*Cucumis melo*) accessions with *Monosporascus* spp. inoculation[1].

Species	Accession[2]				Average
	A-16	C-32	'Goldex'	'HBJ'	
Monosporascus brasiliensis	80.5Aa	38.3Ba	53.8Ba	84.8Aa	64.4
Monosporascus cannonballus	29.5Ab	55.9Aa	65.0Aa	47.1Ab	49.4
Monosporascus caatinguensis	14.5Bb	78.1Aa	78.1Aa	59.8Ab	57.7
Monosporascus mossoroensis	50.9Aa	71.4Aa	64.7Aa	48.8Ab	61.2
Monosporascus nordestinus	14.5Bb	62.6Aa	80.2Aa	96.0Aa	63.3
Monosporascus semiaridus	71.4Aa	55.9Aa	78.1Aa	70.0Ab	68.9
Average	45.1	60.4	69.9	67.9	
Effect[3]	df_1	df_2	Test (Type III – Wald)		
			F		χ^2
Accessions (A)	3	96	4.84**		14.53**
Species (S)	5	96	1.08ns		5.42ns
A × S	15	96	3.01**		45.07**

[1]Means followed by equal letters, uppercase in the rows and lowercase in the columns, do not differ from each other by Scott-Knott's test, at 5% probability. [2]'HBJ', 'Hales Best Jumbo'. [3]df1, degrees of freedom of the numerator; and df2, degrees of freedom of the denominator. * and **Significant at 5 and 1% probability by the F-test and Chi-square (χ^2) test, respectively; nsNonsignificant.

Blanchard). However, it is still the preferred one due to its high quality and long shelf life, which turns out to be the melon with the largest cultivated area in the Brazilian semiarid region, with approximately 22,000 ha per year in the past 20 years. Therefore, despite the potential fragility of the hybrid, a strategy is to obtain yellow melon cultivars resistant to the main pathogens with a 'Goldex' background.

The dry matter reduction index of the roots showed no significant effect in either accessions or species; however, it was observed a notable effect of the interaction among factors (Table 3). In the case of *M. brasiliensis*, accessions formed two distinct groups: A-16 constituting the first group and the remaining accessions comprising the second. When subjected to *M. cannonballus* infection, accessions were again divided into two groups based on the extent of root dry matter reduction. 'Goldex' was in the first group, exhibiting the greatest reduction, while the other accessions formed the second group. With *M. mossoroensis* inoculation, A-16 had the lowest reduction, segregating it from the other accessions with higher averages, which formed another group. For *M. nordestinus*, accessions were grouped into two, with the highest averages being performed by 'Goldex' and 'HBJ', similarly occurring for *M. caatinguensis* and *M. semiaridus*.

Monosporascus brasiliensis species caused a reduction of more than 50% in the A-16 root dry matter, so it was allocated in a separate group (Table 3). The species were gathered in the same group when inoculated the accession C-32. The *M. cannonballus*, *M. brasiliensis*, *M. mossoroensis*, and *M. nordestinus* species were gathered in the same group, because they were more virulent with 'Goldex'. The species were combined into two groups when

Table 2. Melon (*Cucumis melo*) accession reaction to six species of *Monosporascus*.

Species	Accession[1]			
	A-16	C-32	'Goldex'	'HBJ'
Monosporascus brasiliensis	SU	MR	MR	SU
Monosporascus cannonballus	HR	MR	MR	MR
Monosporascus caatinguensis	HR	SU	SU	MR
Monosporascus mossoroensis	MR	MR	MR	MR
Monosporascus nordestinus	HR	MR	SU	SU
Monosporascus semiaridus	MR	MR	SU	MR

[1]SU, susceptible; HR, highly resistant; MR, moderately resistant; and 'HBJ', 'Hales Best Jumbo'.

Table 3. Root matter reduction index of melon (*Cucumis melo*) accession inoculated with *Monosporascus*.[1]

Species	Accession[2]			
	A-16	C-32	'Goldex'	'HBJ'
	Root reduction index			
Monosporascus brasiliensis	0.53Aa	0.38Ba	0.45Ba	0.26Bb
Monosporascus cannonballus	0.30Bb	0.24Ba	0.58Aa	0.10Bb
Monosporascus caatinguensis	0.17Ab	0.17Ba	0.32Ab	0.27Ab
Monosporascus mossoroensis	0.24Bb	0.46Aa	0.40Aa	0.36Ab
Monosporascus nordestinus	0.26Bb	0.21Ba	0.46Aa	0.59Aa
Monosporascus semiaridus	0.39Ab	0.29Aa	0.25Ab	0.23Ab
Effect	df_1	df_2	Test (F of Snedecor)	
Acession (A)	3	96	2.01[ns]	
Species (S)	5	96	2.25[ns]	
A x S	15	96	1.91*	

[1]Means followed by equal letters, uppercase in the rows and lowercase in the columns, do not differ from each other by Scott-Knott's test, at 5% probability. [2]'HBJ', 'Hales Best Jumbo'; df1, degrees of freedom of the numerator; and df2, degrees of freedom of the denominator. * and **Significant by Snedecor's test, at 5 and 1%, respectively. [ns]Nonsignificant.

inoculated 'HBJ', in which the first group was formed by *M. nordestinus*, more virulent, while the second group was formed by the other species.

A-16, which had the lowest root dry matter reduction index (Table 3), has fruits with high mesocarp firmness, high titratable acidity, low content of soluble solids (Dantas et al., 2015), and is resistant to *Myrothecium roridum* Tode (Nascimento et al., 2012). It belongs to the acidulus group, whose varieties are cultivated in India and Sri Lanka. Several genotypes within this group, such as PI 313970 (90625), PI 164323, PI 164723, and Kekiri, present resistance to various viruses and diseases, such as powdery mildew and downy mildew [*Pseudoperonospora cubensis* (Berk. & M.A. Curtis) Rostovzev], as well as insects, such as *Aphis gossypii* Glover (Dhillon et al., 2011; Pitrat, 2016).

Infection caused by *Monosporascus* spp. is contingent upon imbalanced soil conditions and exacerbated by elevated temperatures, such as 35°C in the greenhouse. Furthermore, in the field, intensive melon cultivation system results in the onset of vine decline, and the absence of appropriate crop rotation practices allows these fungi to extend beyond their natural habitat, like spontaneous plant roots such as those of *B. diffusa* and *T. portulacastrum* (Negreiros et al., 2019), causing damage to cucurbit crops.

In the biplot graph (Figure 1), the two axes together explained 97.25% of the total variation in the accession-species interaction, and the polygon formed was composed of four vertices. In each vertex, there is an inoculated accession, which has the highest average rank in its respective sector, being, therefore, the one with the highest severity. The first vertex shows that the species *M. nordestinus*, *M. brasiliensis*, and *M. semiaridus* were more virulent to 'HBJ' in relation to the others. The second vertex indicates that 'Goldex' has the highest average rank in relation to *M. caatinguensis*. The third vertex presents that C-32 was associated with the highest average rank (highest severity) when inoculated with *M. cannonballus* and *M. mossoroensis*. The fourth vertex shows that A-16 did not interact with the species and showed less severity, consequently, it presented higher performance.

The Biplot modeling also can be set in the opposite direction, i.e., setting the species in the vertices. Therefore, the six species were set into five vertices of the polygon of the Biplot graph (Figure 1). In the counterclockwise direction: the species *M. brasiliensis*,
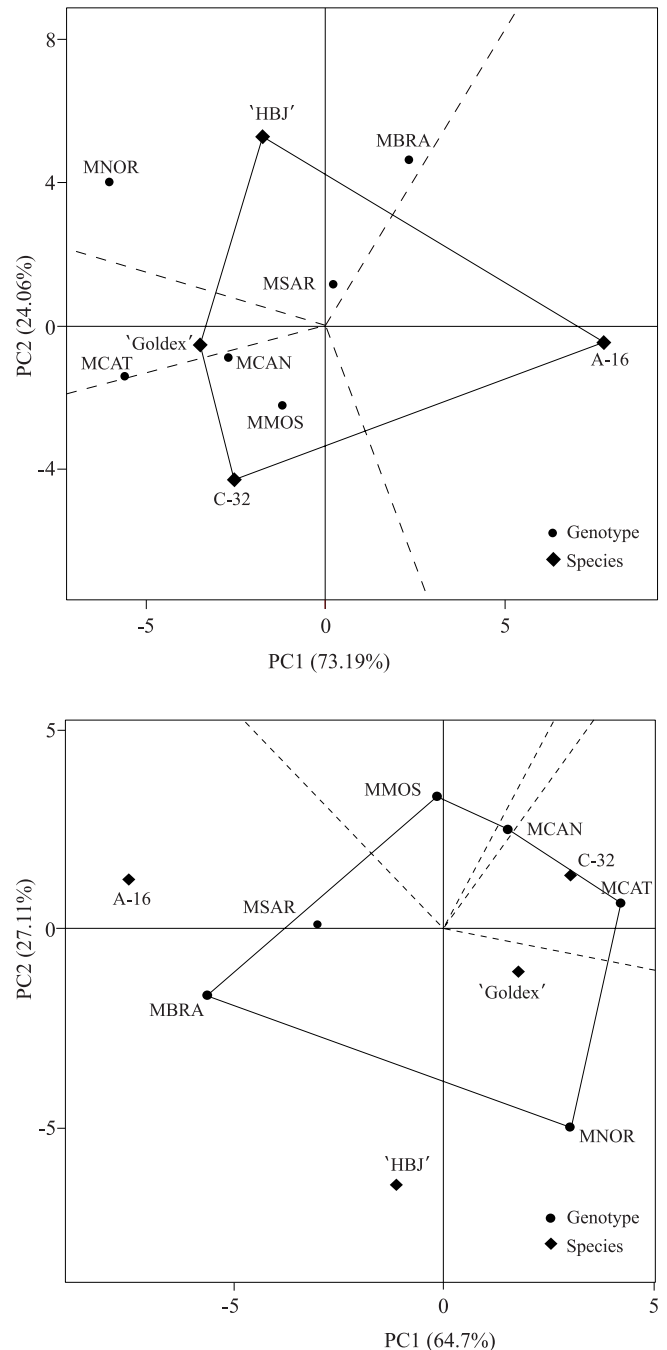

Figure 1. GGE biplot showing: A, the distribution of the melon (*Cucumis melo*) genotypes (on the vertices) inoculated with six species of *Monosporascus* spp.; and B, the distribution of the species of the genus *Monosporascus* (on the vertices) inoculated in four genotypes of melon ('HBJ', 'Goldex', A-16, and C-32). MBRA, *Monosporascus brasiliensis*; MCAN, *Monosporascus cannonballus*; MCAT, *Monosporascus caatinguensis*; MMOS, *Monosporascus mossoroensis*; MNOR, *Monosporascus nordestinus*; and MSAR, *Monosporascus semiaridus*.

in the second vertex, interacted more with A-16; *M. nordestinus*, in the third vertex, interacted with 'Goldex' and 'HBJ'; and *M. caatinguensis* species, located in the fourth vertex, interacted only with C-32. The latter species presented greater virulence because it was more to the right-hand side on the x-axis, i.e., it can be seen as the species that best interacted with C-32, due to the proximity in the graph indicating good association between pathogen and accession. Constrastingly, *M. brasiliensis* had less virulence because it was more to the left-hand side on the x-axis, followed by *M. semiaridus*. The other species showed higher severity, due to the greater distance from the origin (o) on the y-axis. The length of the vector can also reflect high disease incidence and good level of discriminative ability of the varietal groups in regard of the pathogens.

The representation of species at various distances from the origin along the y-axis highlights their diverse associations with genotypes, demonstrating their virulence within the experimental environment. This biplot analysis avoids redundancy and enhances understanding of complex interactions. In contrast to the behavior of varietal groups, the absence of *M. semiaridus* in a distinct vertex of the biplot (Figure 1) can be attributed to intricate interactions between species and accessions in the experimental context (Yan & Falk, 2002).

To date, there are no known reports on the virulence of other species of *Monosporascus* in melon varietal groups, being known only vine decline caused by the species *M. cannonballus* and *M. eutypoides* worldwide (Castro et al., 2020). Despite this, five novel species of *Monosporascus* isolated from weed roots of *B. diffusa* and *T. portulacastrum*, usually found in melon production fields in the Northeastern region of Brazil, were identified (Negreiros et al., 2019).

Therefore, the present work is the first report of the pathogenicity and virulence of the species *M. brasiliensis*, *M. caatinguensis*, *M. mossoroensis*, *M. nordestinus*, and *M. semiaridus* in melon varietal groups, as well as observing that the species have a different behavior according to each accession. It also found that the species *M. cannonballus*, notoriously the agent of vine decline worldwide (Sarpeleh, 2008; Al-Mawaali et al., 2013; Yan et al., 2016; Markakis et al., 2018; Sales Júnior et al., 2019) and the novel species *M. mossoroensis* were the least virulent, since

none of the accessions was classified as susceptible to both species. Particularly, A-16 was highly resistant to *M. cannonballus*, and of moderate or high resistance when inoculated with each of these species. Nevertheless, the other species of *Monosporascus* caused a susceptibility reaction in, at least, one of the accessions (Table 2).

An important aspect for breeding programs is that the tolerance in melon to *M. cannonballus* is strictly related to the root system. In the present work, only the root dry matter reduction index of the genotypes was estimated. There was a positive and significant association between the severity and the root dry matter reduction index, that is, the greater the severity, the lower the dry matter of the roots of the inoculated plants when compared with the non-inoculated plants (Figure 2). Crosby et al. (2000) observed that resistant cultivars have higher averages of total root length, root diameter, number of root branches, number of thin roots (0.0–0.5 mm), and number of small roots (0.5–1.0 mm) compared with susceptible cultivars. The tolerance of the C-32 may be explained by the high vigor and pronounced branching of its root system. This accession has high root mass, even infected, when compared to the susceptible 'Pioñet' (Dias et al., 2002).

Several methods can be developed to evaluate the virulence of pathogens, by observing the severity of attacks on plant tissues, such as damage to the hypocotyl, in primary and secondary roots and reduction of leaf area (Bruton et al., 2000). Therefore, several inoculation methods can be adopted, such as inoculating the soil with agar colonized with the fungus (Tsay & Tung, 1995; Martyn & Miller, 1996; Pivonia et al., 1997), oat husks mixed with sand in pots (Mertely et al., 1993; Karlatti et al., 1997; Pivonia et al., 1997), which proved the effectiveness of the different inoculation methods. In the present study, it was used a mycelial suspension, since some species of *Monosporascus* could not be induced to sporulation (Negreiros et al., 2019).

Significant values were found in practically all accessions and species by means of Spearman's correlations coefficients, but there were some exceptions, such as C-32 (Figure 2), which may indicate that the increase in severity does not cause a significant reduction in dry matter. The positive sign indicates the two variables grow in the same direction, this is, they are directly proportional. When the severity increases,
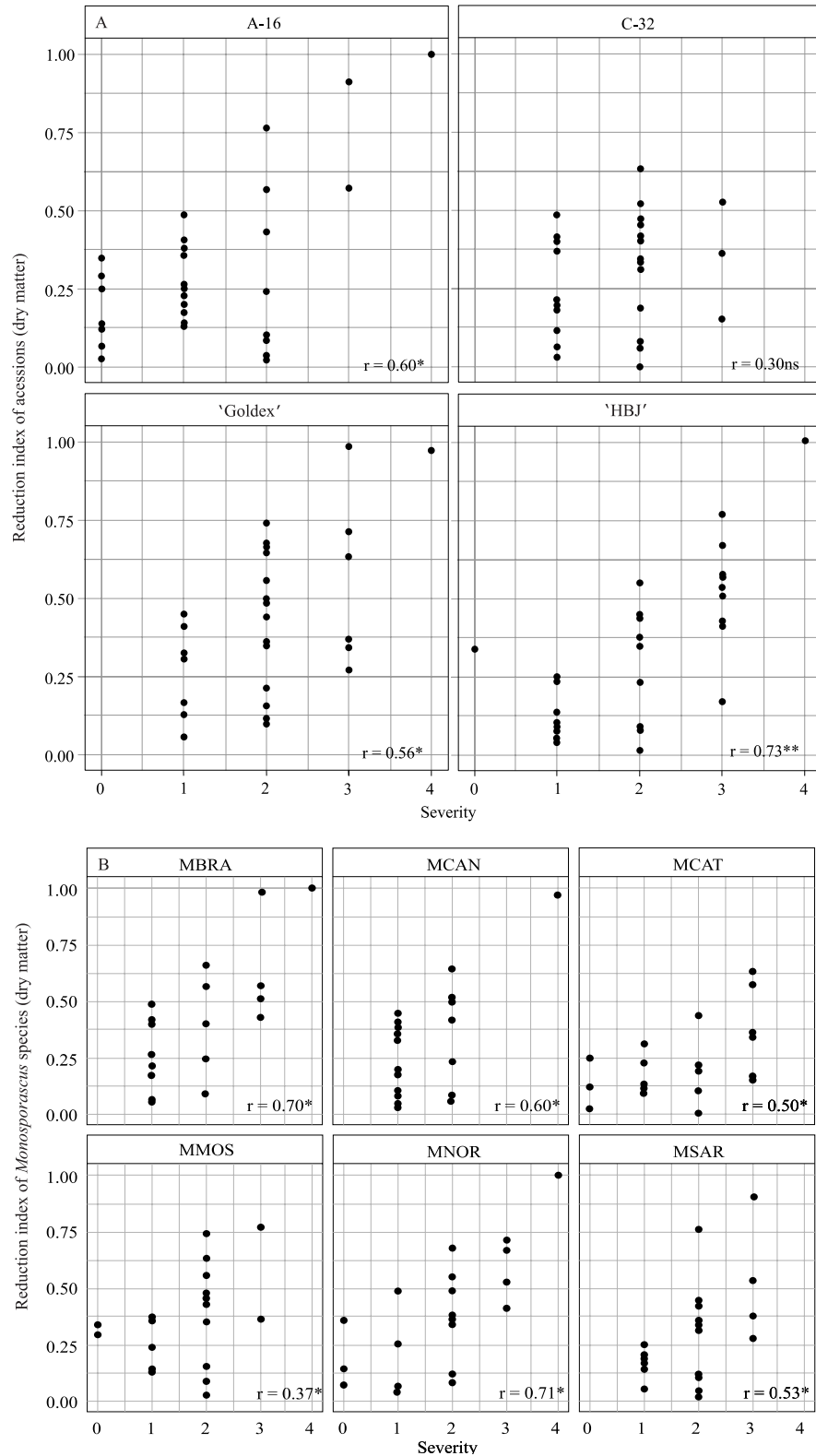
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Figure 2. Estimations of the coefficient of Spearman's correlation between the severity and the root matter reduction index among each melon (*Cucumis melo*) accession (A) and species of *Monosporascus* spp. (B). 'HBJ', 'Hales Best Jumbo'; MBRA, *Monosporascus brasiliensis*; MCAN, *Monosporascus cannonballus*; MCAT, *Monosporascus caatinguensis*; MMOS, *Monosporascus mossoroensis*; MNOR, *Monosporascus nordestinus*; and MSAR, *Monosporascus semiaridus*.

there is a reduction in the dry matter of the plant. The correlation, considering all the data, also followed the same fashion, positive and significant value (r=0.58*).

This study confirmed the susceptibility of the melon groups cantalupensis, inodorus, acidulous, and conomon to five new *Monosporascus* spp. Given the natural presence of these species in melon-producing regions, managing melon vine decline requires awareness of these new species, thus, it is important to produce cultivars resistant to vine decline.

Future breeding strategies should aim at enhancing resistance against pathogens so future research should focus on developing yellow melon cultivars resistant to major pathogens, particularly to specific *Monosporascus* species. Continued investigation into the host-pathogen dynamics will contribute to the development of resilient melon varieties for sustainable cultivation.

Conclusions

1. The A-16 melon (*Cucumis melo*) accession shows higher resistance to *Monosporascus nordestinus*, *Monosporascus caatinguensis*, and *Monosporascus cannonballus*, whereas 'Goldex' presents susceptibility to *M. caatinguensis*, *M. nordestinus*, and *Monosporascus semiaridus*.

2. The species *Monosporascus brasiliensis*, *M. caatinguensis*, *Monosporascus mossoroensis*, *M. nordestinus*, and *M. semiaridus* present varying levels of pathogenicity and diferent levels of infection severity, with *M. semiaridus* having the highest severity, and *M. cannonballus* the lowest.

Acknowledgments

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