EVALUATION OF DIFFERENT BANANA GENOTYPES FOR RESISTANCE TO PANAMA DISEASE

AVALIACAO DE DIFERENTES GENÓTIPOS DE BANANEIRA QUANTO A RESISTENCIA AO MAL-DO-PANAMÁ

Michel GARCEZ¹; Juliana Araújo Santos MARTINS²; Ernesto José Resende RODRIGUES²

1. Graduando em Engenharia Agronômica, Instituto Federal do Triângulo Mineiro – IFTM, Uberlândia, MG, Brasil, garcez2002@gmail.com; 2. Engenheiro Agrônomo, Professor, Doutor, Instituto Federal do Triângulo Mineiro – IFTM, Uberlândia, MG, Brasil.

ABSTRACT: Panama disease, or fusarium wilt disease, caused by the fungus *Fusarium oxysporum* f.sp. *cubense* is a disease causing great losses in banana production worldwide, hindering production in certain areas of the world. In Brazil, banana crop has suffered great losses due to favorable soil and climate conditions for the development of pests and diseases and, also, by the low technological level used in most growing areas of the country. Therefore, the use of genetic resistance for the control of this disease becomes a promising alternative, making possible the cultivation of bananas in some areas that had been decimated by this pathogen. Thus, this study evaluated the performance of banana genotypes subjected to artificial inoculation with *F. oxysporum* f.sp. *cubense* in a nursery and in the field. The genotypes Tropical, Pacovan, Caipira, Japira, Princesa and Fhia-18, besides 'Prata Anã', as a susceptible control, were evaluated in a completely randomized design, with 4 replications, and each replication consisted of two plants. The seedling were also subjected to a field test in a completely randomized design, with three replications. Disease severity, for external symptoms, was evaluated 50 days after inoculation, using a rating scale in which, all plants, except for Fhia-18, which did not show symptoms, had initial yellowing of older leaves. Field evaluations consisted of measuring plant height and diameter, and after harvesting the banana bunches, internal symptoms were observed in the pseudostems. Cultivars Prata-Anã and Caipira had greater susceptibility to the disease.

KEYWORDS: Fusarium wilt disease. Musa spp. Genetic resistance. Fusarium oxysporum f.sp. cubense

INTRODUCTION

Banana cultivation is agricultural activity of great importance for Brazil, fourth major producer in the world, with 6.97 million tons produced in 508.800 ha, and average yield of 13.7 t ha⁻¹ (FAO, 2007). The average of the 10 countries with greatest yield is $46.3 \text{ t} \text{ ha}^{-1}$, much above world average, which is $18.4 \text{ t} \text{ ha}^{-1}$. Such low yield of national banana crop, below the world average, and much lower than its potential, is due to the cultivars used in Brazil, but also to the technological level adopted by producers and the widespread incidence of pests and diseases in the crop. One of the major fungal diseases, limiting cultivation of 'Maçã' cultivar and became serious in some important growth areas of 'Prata' cultivar, is Panama disease, a soilborne disease, where the pathogen can survive for many years, turning some regions inapt for banana cultivation (SILVA et al., 2011).

While the difficulties for banana cultivation increase due to diseases, Brazilian consumers, as well as those elsewhere in the world, become more aware and concerned with the presence of contaminants and chemical residues (pesticides), besides the nutritional value of the food. Society demands that, simultaneously, conserve natural resources and provide healthier products, without compromising technological levels of food safety already reached. There is a potential market for fruits obtained with reduced use of pesticides, within programs of certification and surveillance. Banana producers in Brazil cannot be oblivious to world demand for sustainability of production systems.

Considering that the use of genetic resistance for disease control in bananas is a highly promising alternative, this study evaluated the performance of 7 banana genotypes subjected to artificial inoculation with *Fusarium oxysporum* f.sp. *cubense* in the greenhouse and field.

MATERIALS AND METHODS

The experiments were done in a nursery and in the field, in 2012 and 2013, respectively, at the Instituto Federal do Triangulo Mineiro, Uberlândia -MG coordinates: 18°45'51"S and 48°17'21"W; 655m above sea level.

The fungus isolate was obtained from pseudo stem fragments with typical symptoms of Panama disease, from Uberlândia. Small fragments were disinfected with alcohol 70%, for 30 seconds and transferred to sodium hypochloride 1.5%, for 1-

2 minutes. Subsequently, the fragments were rinsed twice in sterile distilled water, placed over sterile paper towels and transferred to Petri plates containing potato-dextrose-agar (PDA). After fungal growth was observed, fragments of mycelium were asseptically transferred to other Petri plates to obtain pure cultures and to test tubes containing PDA, for subsequent studies.

Fusarium oxysporum f. sp. cubense was incorporated to a sterile mixture of rice:sand:water, in the proportion 4:2:3. This mixture, after sterilization by autoclaving for 30 minutes had adequate consistency for homogeneous incorporation of the inoculum, after cooling by the addition of mycelial disks of F. oxysporum f. sp. cubense grown in the Petri plates. The pathogen was cultivated in 250-mL erlenmeyers containing 125 mL of substrate. The flasks were vigorously shaken every other day to assure homogeneous fungal growth. An inoculum suspension was prepared, after eight days of incubation, by diluting the mixture with sterile water. The suspension was adjusted to 10^6 conidia mL⁻¹ with a Neubauer chamber (RODRIGUES et al., 1993).

Banana seedlings were inoculated by dipping the roots in the spore suspension for 60 seconds. The seedlings were transplanted to 10-L and transferred to the nursery. The pots experimental design was completely randomized, with five replications. Plants were removed from the pots fifty days after inoculation, washed, and the rhizome was split for the evaluations, according to the scale proposed by Cordeiro et al. (1993): (0) no vascular discoloration; (1) isolated discoloration spots in the vascular cambium (CV); (2) discoloration of up to 1/3 of CV; (3) discoloration between 1/3 and 2/3 of CV; (4) discoloration greater than 2/3 of CV; and (5) complete discoloration of CV. Disease severity of external symptoms was evaluated using a rating scale with five classes: 1 no symptoms; 2 - initial yellowing of old leaves; 3 yellowing of old leaves and initial discoloration of voung leaves; 4 – intense vellowing of all leaves; 5 - dead plant (RODRIGUEZ et al., 2011). The field experiment was done in 2013, in a completely randomized design, with 7 treatments and 3 replications. A total of 3 evaluations were cone, considering plant height pseudo-stem diameter, and external and internal symptoms of Panama disease disease, caused by Fusarium oxysporum f.sp. cubense. Crop management was done with planting and side dressing fertilization, according to technical recommendations. Data were transformed to $\sqrt{x} + 0.5$. The results were submitted to analysis of variance and the averages compared by the Tukey test at 5% probability.

Seedlings used in the experiment were obtained by tissue culture, and were about 45 days old. Six genotypes with different resistance degrees against Panama disease, besides 'Prata-Anã', a susceptible control (Borges et al., 2010) were used.

RESULTS AND DISCUSSION

Data obtained in external and internal evaluations for the nursery experiment are shown in Table 1. The genotypes did not vary significantly on Panama disease symptom expression.

Disease severity, from the external symptoms, evaluated 50 days after inoculation, with a rating scale, showed that all plants had initial yellowing of older leaves. Internally, all genotypes had isolated discoloration spots in the vascular cambium of the rhizome.

Given that a genotype rating below 1.4 is considered resistant (CORDEIRO et al., 1993), no genotype in this study was resistant. Even the genotypes considered resistant presented internal disease symptoms. However, according to Rishbeth and Naylor (1957), infection can be more restricted to the roots in resistant cultivars, due to a more effective reaction of hos cells, or the presence of toxic substances against the pathogen. This was observed in all genotypes evaluated.

Plants of genotype Prata-Anã had low disease severity, i.e., all plants survived inoculation, in contrast with what was expected from a genotype considered as susceptible.

	Panama disease		
Constynes	Internal	External	
Genotypes	symptoms	symptoms	
Tropical	1,8 a	2,0 a	
Pacovan Ken	1,6 a	2,0 a	
Caipira	2,4 a	2,2 a	
Japira	1,6 a	2,0 a	
Princesa	2,4 a	2,2 a	
Fhia-18	2,0 a	1,2 a	
Prata-Anã	2,0 a	1,6 a	
C.V. (%)	16,10	16,0	

Table 1. Disease severity in banana genotypes, caused by Fusarium oxysporum f. sp. cubense.

Averages followed by the same letter in the column do not differ statistically by the Tukey test at 5% significance. Data transformed by $\sqrt{x + 0.5}$

This study was fundamental to determine the causal agent of the disease, by Koch's postulates, proving that the pathogen causing banana leaf dropping, leaf yellowing, and fissures in the pseudostem is the fungus *F. oxysporum* f.sp. *cubense*.

Tables 2. present the values of p	plant height and diameter,	respectively.

Genotypes	Average	Tukey	Genotypes	Average	Tukey
Pacovan Ken	363,55	a	Fhia - 18	16,99	а
Princesa	327,88	ab	Pacovan Ken	16,95	a
Japira	322,66	ab	Princesa	16,27	ab
Caipira	321	ab	Caipira	15,65	ab
Tropical	256,44	bc	Japira	15,40	ab
Fhia-18	213,66	с	Prata-Anã	14,91	ab
Prata-anã	210	с	Tropical	13,85	b
C.V. (%)	21,82		C.V. (%)	11,46	

Averages followed by the same letter in the column do not differ statistically by the Tukey test at 5% significance. Data transformed by $\sqrt{x + 0.5}$

Cultivar Pacovan Ken was the tallest one, thus this cultivar was little affected by inoculation of the fungus. A study done by Silva et al (2011), showed that cultivar Pacovan Ken also presented the best ratings for resistance against Mal do Panamá. Another characteristic of this cultivar is its acceptance by consumers for its flavor similar to cultivar Prata. Therefore, besides its high resistance agaist mal do panamá it is well accepted by the consumer market (GARRUTI, 2006). Plant diameter also was evaluated, and cultivars Pacovan Ken and FHIA-18 had the greatest values, similar to each other. Such results were also observed by Donato et al. (2009), who found that cultivar FHIA-18 had the greatest ratings for pseudo-stem height and diameter, while cultivar Prata-Anã had the lowest ones. Therefore, cultivars Prata-anã and Caipira were the ones presenting fissures in the pseudo-stem and, after harvesting, were split to analyze internal symptoms, presented Evaluation of different banana...

redish color of the vascular tissue, a characteristic symptom of the fungus *F. oxysporum* f.sp. *cubense*.

CONCLUSIONS

All the genotypes presented internal and external disease symptoms.

Cultivars Prata-anã and Caipira were the ones presenting greater susceptibility to the disease caused by *Fusarium oxysporum* f.sp. *cubense*, since they had fissures in the pseudo-stem and leaf yellowing, external symptoms characteristic of Panama disease

RESUMO: Alguns problemas da bananicultura mundial se tornaram também problemas para o Brasil. A doença fúngica de solo, que limita o cultivo da banana 'Maçã' é de difícil controle e tornou-se grave problema em algumas importantes áreas de cultivo é o mal-do-Panamá, uma das mais importantes doenças da bananeira, causada pelo fungo *Fusarium oxysporum* f.sp. *cubense*, provoca elevadas perdas na produção e pode persistir no solo por muitos anos. Portanto o objetivo desse trabalho é avaliar o comportamento de genótipos de bananeira quando submetidos à inoculação artificial de *Fusarium oxysporum* f.sp. *cubense* em situação de viveiro de mudas e situação de campo. Foram avaliados os genótipos Tropical, Pacovan, Caipira, Japira, Princesa e Fhia-18, além da 'Prata Anã' como testemunha (suscetível), em delineamento inteiramente casualizado, com 4 repetições, sendo duas plantas por repetição. As mudas também foram submetidas a teste de campo em que o delineamento experimental foi inteiramente casualizado com 3 repetições. A severidade da doença quanto aos sintomas externos foi avaliada aos 50 dias após a inoculação utilizando escala de notas, na qual todas as plantas, com exceção da Fhia-18 que não apresentou sintomas, apresentaram amarelecimento inicial nas folhas velhas.No teste de campo foram avaliados, altura e diâmetro das plantas e após a retirada dos cachos, foi observado os sintomas internos do pseudocaule onde as cultivares Prata-Anã e Caipira apresentaram maior suscetibilidade a doença.

PALAVRAS-CHAVE: Mal-do-Panamá. Bananicultura, Resistência genética. Fusarium oxysporum f.sp.cubense.

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