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Short communication

A single dominant gene confers resistance to *Fusarium oxysporum* f. sp. melonis race 1 in West Indian Gherkin (*Cucumis anguria* L.) accessions

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Abstract

To breed West Indian Gherkin (*Cucumis anguria* L.) cultivars with resistance to fusarium wilt, information about resistance is necessary. In this study, 24 accessions were inoculated with *Fusarium oxysporum* f. sp. *melonis* race 1, and 12 accessions showed similar reactions to immune (SI). The mode of inheritance of the resistance was investigated using the highly susceptible (HS) and SI accession, and the F_1 and F_2 plants. All F_1 were classified into SI, but the segregations were observed in the F_2. The number of SI and HS plants was in accordance with the expected 3 (SI):1 (HS) ratio. These results suggest that the resistance is controlled by a single dominant gene.

Keywords: *Cucumis anguria* L.; *Fusarium oxysporum*; Genetic resource; Inheritance mode; West Indian Gherkin

1. Introduction

West Indian Gherkin (*Cucumis anguria* L.), belongs to the subgenus *Melo*, and originated from southwestern Africa. The wild form is known as *C. anguria* var. *longaculeatus*, and is widely distributed in the northern parts of southern Africa. The fruits and leaves are consumed as vegetable in the United States, Brazil, India, and southern Africa (Mangan et al., 2008; Welman, 2003). Some studies have reported the susceptibility of West Indian Gherkin plants to certain melon (*Cucumis melo* L.) diseases such as powdery mildew (Alvarez et al., 2005; Lebeda, 1984; Pan and More, 1996), downy mildew (Pan and More, 1996), and fusarium wilt (Alvarez et al., 2005; Matsumoto et al., 2011). Among these diseases, fusarium wilt is the most serious disease. Once this disease colonizes a field, the pathogen survives in the soil, in crop residues, and in roots of crops grown in rotation; this increases the persistence of these pathogenic populations (Banihashemi and Dezeeuw, 1975; Gordon et al., 1989; Zuniga et al., 1997). It is caused by *Fusarium oxysporum* f. sp. *melonis*, and divided into four physiological races (0, 1, 2, and 1,2) based on its pathogenicity to three melon cultivars, 'Charentais T', 'Doublon', and 'CM 17187' (Risser et al., 1976). In these races, 1 and 2 are most prevalent and have been recognized since the 1930s (Zuniga et al., 1997). Therefore, control of these races is important for gherkin cultivation. Although some accessions resistance to race 2 in West Indian Gherkin, and the mode of inheritance of the resistant gene was reported (Alvarez et al., 2005; Matsumoto and Miyagi, 2012), few accessions resistance to race 1 were reported (Alvarez et al., 2005). Generally, resistant cultivars are effective in controlling such soil-borne diseases, and many resistant cultivars have been developed in melon (Tezuka et al., 2009). To date, no West Indian Gherkin cultivar with resistance to these diseases has been developed; however, high-yield cultivars have been previously created by cross breeding (Modolo and Costa, 2004). To breed new West Indian Gherkin cultivars with resistance to these races, resistant genetic resources are required.

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Furthermore, understanding the mechanism of mode of inheritance is important to create a breeding plan.

In this study, we evaluated the response of 24 West Indian Gherkin accessions to *F. oxysporum* f. sp. *melonis* race 1, and investigated the mode of inheritance using F_1 and F_2 generations developed by crossing the resistant and susceptible accessions.

2. Materials and methods

2.1. Fungal strain and plant material

The fungal strain used for inoculation was Mel 02221 and was classified as race 1 (Namiki et al., 2000). All the 24 wild West Indian Gherkin accessions collected from Southern and East Africa, and Latin America were obtained from the Germplasm Resources Laboratory (USDA, ARS, Beltsville, MD, USA) (Table 1). Three different melon cultivars, 'Charentais T', 'Doublon', and 'CM 17187', were used for race confirmation of the fungal strain Mel 02221. These cultivars were obtained from the Institut National de la Recherche Agronomique, France.

2.2. Artificial inoculation

Inoculation was conducted using a root dip method (Matsumoto et al., 2011). The fungal strain was cultured in 100 mL potato dextrose broth (PDB) in 300 mL flasks on a rotary shaker (ca. 120 rpm) for 1 week at 25 °C. The culture was filtered through two-ply gauze. Spore concentration was determined using a hemocytometer and was adjusted to the appropriate density by dilution with sterile distilled water. For artificial inoculation, seeds of the tested plants were sown in sterilized garden soil in plastic trays and grown in a growth chamber at 26–30 °C. Seedlings with fully expanded first true leaves were removed from the soil. Their roots were washed in tap water and dipped in a conidial suspension (10^7 spores per mL) of Mel 02221 for 15 s. Inoculated seedlings were transplanted in the sterilized fresh garden soil in new plastic pots and cultivated in the growth chamber at 23 °C (16 h photoperiod).

Table 1
Accessions of West Indian Gherkin used for the evaluation and response to *Fusarium oxysporum* f. sp. *melonis* race 1.

Accession	Disease severity [a]	Reaction [b]	Collected country [c]
Ames 22076	0 ± 0^{A}	SI	Zambia
Ames 23536	1.3 ± 0.5^{ABCD}	MR	South Africa
Ames 23541	1.1 ± 0.4^{ABCD}	MR	South Africa
Ames 23550	0.9 ± 0.4^{ABC}	MR	South Africa
Ames 23589	2.0 ± 0.4^{BCDE}	SU	South Africa
PI 147065	0 ± 0^{A}	SI	Brazil
PI 196477	2.7 ± 0.2^{E}	HS	Brazil
PI 233646	0 ± 0^{A}	SI	Ethiopia
PI 249895	2.2 ± 0.3^{DE}	SU	Zimbabwe
PI 249896	2.1 ± 0.3^{CDE}	SU	Zambia
PI 249897	0.3 ± 0.2^{A}	HR	Namibia
PI 320052	0 ± 0^{A}	SI	Ethiopia
PI 364475	0 ± 0^{A}	SI	South Africa
PI 390449	0 ± 0^{A}	SI	Ecuador
PI 438570	0 ± 0^{A}	SI	Guatemala
PI 438678	0.8 ± 0.3^{AB}	MR	Mexico
PI 438679	0 ± 0^{A}	SI	Mexico
PI 482383	0 ± 0^{A}	SI	Zimbabwe
PI 482386	1.7 ± 0.3^{BCDE}	SU	Zimbabwe
PI 482387	0 ± 0^{A}	SI	Zimbabwe
PI 482392	0 ± 0^{A}	SI	Zimbabwe
PI 494824	0.6 ± 0.3^{A}	MR	Zambia
PI 512091	0 ± 0^{A}	SI	Mexico
PI 542135	0.4 ± 0.3^{A}	HR	Botswana

[a] Means±SE., accessions with different letters are significantly different (Bonferroni multiple comparison *t* test following ANOVA, $P<0.05$).

[b] SI, HR, MR, SU, and HS indicates similar to immune, high resistance, moderately resistance, susceptible, and highly susceptible, respectively.

[c] The country names were according to the Germplasm Resources Information Network of USDA (http://www.ars-grin.gov/).

2.3. Disease severity evaluation

Disease severity was evaluated 21 days after inoculation by using a 0–3 scale (0 = no symptoms, 1 = beginning of symptom on leaves, 2 = leaves strongly affected, 3 = plant death) (Matsumoto et al., 2011), and each accession was classified into five reaction classes: 0 = similar reactions to immune (SI); 0.1–0.5 = highly resistance (HR); 0.6–1.5 = moderately resistance (MR); 1.6–2.5 = susceptible (SU); 2.6–3 = highly susceptible (HS) by means of disease severity scales.

A total of 10–20 plants were evaluated for each accession or melon cultivar. Results were expressed as mean values and standard errors. Disease severity was log-transformed and data were analyzed using least-squares analysis of variance (ANOVA). A post hoc Tukey-HSD test was performed to compare disease severity among the accessions. For these analyses, JMP statistical software (ver. 9.0.0; SAS Institute Inc., Cary, NC, USA) was used.

2.4. Mode of inheritance of the resistant gene

From the results of inoculation, two accessions (PI 196477 and PI 320052) were selected as the parental lines; PI 196477 as the HS accession and PI 320052 as the SI. F_1 and F_2 generations were obtained from a cross between PI 196477 and PI 320052. All each 50 plants of the F_1 and F_2 generation were inoculated. The observed ratios were used in the segregation of fit, with a significance level established at $P<0.05$.

3. Results and discussion

3.1. Evaluation of disease severity

Although all 'Charentais T' and 'Doublon' seedlings died and the mean disease severity was 3.0 and classified as HS, all 'CM 17187' seedlings were asymptomatic and the mean disease severity was 0 and classified as SI. According to previous reports, 'Charentais T' is HS to all races, 'Doublon' showed SI to races 0 and 2, and 'CM 17187' showed SI to races 0 and 1 (Risser et al., 1976). Therefore, the fungal strain Mel 02221 was confirmed as race 1 from the pathogenicity of the inoculated plants.