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RESISTANCE TO ALTERNARIA BROWN SPOT OF NEW CITRUS HYBRIDS[1]

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ABSTRACT- Alternaria brown spot (ABS) disease is caused by the fungus of *Alternaria alternata f. sp. citri*, which causes injury in leaves, branches and fruits of citrus. The action of the pathogen is directly related to the presence of toxin receptors in susceptible genotypes. The objective of this study was to characterize a population of citrus hybrids obtained from controlled crosses between Pêra de Abril sweet orange and the hybrid of Murcott tangor x Pêra sweet orange (TM x LP 163) for response to ABS through the *in vitro* inoculation of fungal spores in young detached leaves. The fungus was isolated from the lesions of Murcott tangor fruits that exhibited ABS symptoms. Two hundred thirty-five hybrids were evaluated, and 70 (30%) showed different levels of disease symptoms on detached leaves after 72 hours of inoculation with the fungus, and 165 (70%) were asymptomatic. The frequency of segregation observed (165R:70S) and high level of heritability (h^2_g = 0.91) suggest that few genes may be involved in controlling the inheritance of ABS resistance in citrus.

Index terms: *Alternaria alternata*, genetic breeding, Murcott tangor, Pêra de Abril, sweet orange.

RESISTÊNCIA DE NOVOS HÍBRIDOS DE CITROS À MANCHA MARROM DE ALTERNARIA

RESUMO - A mancha marrom de alternaria (MMA) é uma doença causada pelo fungo *Alternaria alternata f. sp. citri*, que causa lesões em frutos, folhas e ramos de citros. A ação do patógeno está diretamente relacionada à presença de receptores de toxina em genótipos suscetíveis. O objetivo deste estudo foi caracterizar uma população de híbridos de citros obtidos a partir de cruzamentos controlados entre a laranja Pêra de Abril e o híbrido de tangor Murcott x laranja Pêra (TM x LP 163) para resposta à MMA através da inoculação *in vitro* de esporos do fungo em folhas jovens destacadas. Um isolado do fungo foi obtido das lesões de frutos de tangor Murcott que exibiram sintomas de MMA. Duzentos e trinta e cinco híbridos foram avaliados, sendo que 70 (30%) apresentaram níveis diferentes de sintomas de doença em folhas destacadas após 72 horas da inoculação com o fungo e 165 (70%) foram assintomáticos. A frequência de segregação observada (165R:70S) e alta herdabilidade (h^2g = 0,91) sugerem que poucos genes podem estar envolvidos no controle da herança da resistência à MMA em citros.

Termos de indexação: *Alternaria alternata,* melhoramento genético, tangor Murcott, Pêra de Abril, laranja

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INTRODUCTION

The main diseases that affect the Brazilian citrus industry include tristeza (RODRIGUES et al., 2014), citrus canker (GRAHAM et al., 2004), citrus variegated chlorosis (COLETTA FILHO et al., 2007), leprosis (BASTIANEL et al., 2010) and huanglongbing (greening) (COLETTA FILHO et al., 2004). With respect to mandarins specifically, we highlight alternaria brown spot (ABS), which is caused by *Alternaria alternata* f. sp. *citri*, that affects the main commercial varieties and their hybrids in the country, such as Ponkan mandarin (*Citrus reticulata* Blanco) and Murcott tangor (*C. reticulata* x *Citrus sinensis* Osbeck) (AZEVEDO et al., 2015).

ABS was first identified in Australia in Emperor mandarin (*C. reticulata* Blanco) around 1903, and the disease soon spread to other places in the world; it is now present in several countries, such as Turkey, Spain, Italy, South Africa, the USA, China, Brazil and Argentina (STUART et al., 2009; CUENCA et al., 2013, 2016; HUANG et al., 2015). In Brazil, the disease was recorded in Rio de Janeiro in 2001 (GOES et al., 2001) in Dancy mandarin, and it later spread to other states, such as Minas Gerais, Rio Grande do Sul and São Paulo (SPOSITO et al., 2003; STUART et al., 2009).

ABS is mainly disseminated through conidia (asexual spores) of the fungus, and the process of colonization involves the production of host-specific toxin (HST) (TSUGE et al., 2013). ABS symptoms occur in leaves, branches and fruits. The symptoms initially appear as small lesions that expand by the action of the toxin and cause intense defoliation during periods of increased infection (TIMMER et al., 2003). The toxin type ACT produced by *A. alternata f. sp. citri* is widely described in the literature and is specific to a particular host range (KOHMOTO et al., 1991; PEEVER et al., 1999), including mandarins and some hybrids.

In fine branches, small cortical lesions occur, with or without a chlorotic halo. In mature fruits, necrotic spots 1 to 10 mm in diameter occur, and the external quality of these fruits is severely reduced, depreciating them for consumption (TIMMER et al., 2003; TSUGE et al., 2013; BASSIMBA et al., 2014).

Currently, ABS constitutes the main disease of mandarins (*C. reticulata*) and their hybrids; the main varieties planted in orchards in the state of São Paulo, including Ponkan mandarin and Murcott tangor, are highly susceptible (AZEVEDO et al., 2010). ABS incidence leads to reduced productivity (PRATES, 2004; AZEVEDO et al., 2010; BASSANEZI et al., 2014) and, consequently, economic loss (CHUNG, 2012).

The resistance of citrus plants to ABS has been observed in species of citrus such as sweet orange (STUART et al., 2009), willow leaf mandarins and mandarins like Fremont, Thomas, Clementina and Cravo (REIS et al., 2007; SOUZA et al., 2009; PACHECO et al., 2012).

Few studies have investigated the origin of resistance and susceptibility of citrus genotypes to ABS. Susceptibility is likely simple and controlled by dominant inheritance (DALKILIC et al., 2005; GULSEN et al., 2010; CUENCA et al. 2013, 2016). Thus, the “A” allele would be dominant for susceptibility, and “a” would be recessive for disease resistance.

Studies conducted using populations of triploid hybrids (CUENCA et al., 2013) and diploid hybrids (CUENCA et al., 2016) confirmed these results, and the authors suggested that Murcott tangor and Orlando tangelo are both susceptible and heterozygous and that willow leaf mandarins (*C. deliciosa* Tenore) and clementines (*C. clementina*) are resistant and homozygous recessive.

The present study aimed to characterize ABS resistance in a population of 235 citrus hybrids from crosses between Pêra de Abril sweet orange and a hybrid of Murcott tangor x Pêra sweet orange (TM x LP163).

MATERIAL AND METHODS

A population of 235 hybrids was obtained in 2010 from controlled crossings between Pêra de Abril sweet orange (*C. sinensis*) and TM x LP 163. The latter is a hybrid between Murcott tangor (*C. reticulata* x *C. sinensis*) x Pêra sweet orange (*C. sinensis*), produced in 2010. The population was available for evaluation in the greenhouse at the Sylvio Moreira Citrus APTA Center of the Agronomic Institute (IAC).

Isolates of *A. alternata f. sp. citri* were obtained from injured tissues of Murcott tangor fruits collected from plants grown in the field at the Sylvio Moreira APTA Citrus Center of the Agronomic Institute (IAC). Leaf tissue with characteristic lesions was cut into small pieces with a scalpel. Surface disinfection was then performed by immersion in commercial ethyl alcohol (70%) for 1 minute followed by immersion in 2% sodium hypochlorite for 2 minutes, after which the samples were transferred to Petri dishes containing potato dextrose agar medium (200 g potato, 20 g dextrose, 15 g L^{-1} agar).

To prevent the growth of other fungal contaminants, fungicide with carbendazim (640 mg L^{-1}) as the active ingredient was added. To avoid the growth of bacteria, 50 µg mL^{-1} tetracycline was added to the culture medium. The plates were maintained under a photoperiod of 12 hours and a temperature of 25 °C (SASSERON, 2008).

After seven days, media composed of 30 g of calcium carbonate, 20 g of sucrose and 20 g of agar was prepared to a volume of 1 L and poured into Petri plates. Fifty 5-mm-wide discs with mycelial tissue growth were then transferred to this medium to induce sporulation. This material was maintained at 27 °C for 5 days under a 12-hour photoperiod. The identity of the isolates was confirmed via the optical microscopy of asexual structures.

For *in vitro* inoculation, young leaves were collected from all hybrids and their parents. The ABS-susceptible Dancy mandarin and Murcott tangor varieties and the ABS-resistant Fremont mandarin variety (AZEVEDO et al., 2010) served as control genotypes. Three leaves per inoculated plant were placed in a Petri dish with moistened filter paper and cotton, which were previously sterilized. The leaves were sprayed with 2 mL of a suspension containing the spores at a concentration of 10^5 conidia mL^{-1} according to the methodology described by Peever et al. (1999) and Canihos et al. (1999). The experiment was conducted in a growth chamber at 27 °C under a 12-hour photoperiod. The experimental design was completely randomized with three replicates per genotype, and each replicate was represented by one Petri dish.

The evaluations of symptoms caused by the fungus were performed at 24, 48 and 72 hours after inoculation by observing the presence of typical symptoms of the disease. The determination of severity was assessed for a subset of three leaves per treatment using a diagrammatic scale including nine levels of severity in leaves expressed as the percentage of infected area (0, 0.3, 3.5, 8.0, 15, 34, 61, 80, 90 and 97%) (MARTELLI et al., 2016).

To better understand the disease progress over time, it was calculated the area under the disease progress curve (AUDPC). Based on the results, a representative histogram of the population in relation to ABS severity was constructed using the software R.

The severity of the disease data at the three times of evaluation was used to calculate the AUDPC as follows:

$$AUDPC = \Sigma [((y1 + y2) / 2) * (t2 - t1)]$$

where y1 and y2 are two consecutive assessments carried out at times t1 and t2, respectively. This calculation is a statistical analysis that allows the verification of the behavior of the disease by the progress curve (BERGAMIN, 1995) from mathematical models used previously.

The AUDPC values were used to calculate genetic parameters such as variation (genetic and environmental) and heritability. For this, the program SELEGEN - REML/BLUP was used (RESENDE and SILVA, 2014).

RESULTS AND DISCUTION

Table 1 shows the severity values assessed using the diagrammatic scale and the AUDPC. In the first evaluation, 24 hours after the inoculation of detached leaves *in vitro*, leaf lesions were observed in 54 hybrids (22.9%) and in varieties used as positive controls (Murcott tangor and Dancy mandarin), as expected (Figure 1). After 48 hours of inoculation, 54 hybrids were considered symptomatic, and 14 new hybrids showed symptoms typical of the disease. At this evaluation stage, a higher percentage of affected leaf area in the leaves where symptoms started was observed at 24 hours. Values ranging from 0.3% (24 hours) to 61% (48 hours) of the leaf area with symptoms were also observed. Forty-eight hours after inoculation, a 22.98% (24 hours) to 28.94% increase in the percentage of individuals showing symptoms was observed.

After 72 hours, an increase in the area of necrosis on the leaves in which symptoms began within 24 hours was observed, and the hybrid PAx163-3 showed maximum severity, as assessed by the diagrammatic scale (97%). Throughout the population, 165 (70%) asymptomatic individuals were observed, and 70 (30%) other individuals showed different levels of symptoms (Figure 2). Studies on inheritance of ABS resistance in citrus were based on crosses between mandarins varieties (CUENCA et al., 2013) and between mandarin and sweet orange (CUENCA et al., 2016). These studies stated that the inheritance of ABS resistance in citrus is controlled by a single recessive allele (DALKILIC et al., 2005; GULSEN et al., 2010; CUENCA et al., 2013). Thus, the “A” allele would be dominant for susceptibility, and “a” would be recessive for disease resistance (CUENCA et al., 2013, 2016). Therefore, segregation is expected in progeny arising from crosses between resistant and heterozygous ABS-susceptible parents or even between two heterozygous ABS susceptible ones. In the present work, the parents Pêra de Abril sweet orange and the hybrid TM x LP 163, were asymptomatic, 165 (70%) asymptomatic individuals of the progeny were observed and 70 (30%) individuals showed different levels of symptoms (Figure 2). Thus, the segregation observed in F_1 plants contrasts with that reported before (DALKILIC et al., 2005; GULSEN et al., 2010; CUENCA et al., 2013, 2016) and suggested that two genes were involved in genetic control of this trait, rather than a single gene. Then, we suggested that due to a complete dominance for both gene pairs, only if both kinds of dominant alleles were present the susceptible phenotype appears. When one gene is homozygous recessive or both genes are homozygous recessive, the susceptible phenotype is hidden. Further studies should be conducted to elucidate the genetic mechanisms involved in sweet orange or in other varieties that do not respond to a HST produced by the pathogen.

High heritability (h^2_g = 0.91) and genotypic variability expressed by the genotypic variation coefficient (*Cvgi* = 228.25) (Table 2) were observed in the present work, indicating that the selection of resistant plants in the progeny can be successful. Namely, the hybrids have adequate genetic variability for selection.

TABLE 1- Severity (%) and the area under the disease progress curve (AUDPC) of alternaria brown spot evaluated after inoculation with *A. alternata f. sp. citri* in hybrids, parents and citrus varieties.

Genotype	Hours after inoculation						AUDPC		Genotype	Hours after inoculation						AUDPC	
	24 h		48 h		72h					24 h		48 h		72h			
Fremont	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 180	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 1	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 184	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 10	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 185	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 100	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 186	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 101	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 188	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 102	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 193	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 104	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 195	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 108	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 196	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 11	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 197	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 110	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 203	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 112	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 204	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 113	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 21	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 117	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 212	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 119	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 213	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 121	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 215	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 122	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 220	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 123	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 222	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 124	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 223	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 125	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 224	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 126	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 226	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 127	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 234	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 128	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 235	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 130	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 236	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 132	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 238	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 133	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 239	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 134	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 240	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 138	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 241	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 140	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 242	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 143	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 243	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 144	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 244	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 145	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 247	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 146	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 248	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 147	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 25	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 149	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 250	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 15	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 251	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 151	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 252	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 152	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 253	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 153	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 254	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 154	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 255	0.0	aA	0.0	aA	0.0	aA	0.0	a

continued...

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PAx163 156	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 257	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 157	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 258	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 158	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 26	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 159	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 260	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 160	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 261	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 162	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 263	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 164	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 264	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 165	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 265	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 166	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 266	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 167	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 267	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 168	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 269	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 169	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 271	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 170	0.3	aA	0.0	aA	0.0	aA	0.0	a	PAx163 273	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 171	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 276	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 172	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 277	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 173	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 279	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 174	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 28	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 175	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 280	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 177	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 283	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 179	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 29	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 33	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 78	0.3	aA	0.3	aA	0.3	aA	14.4	a
PAx163 35	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 13	0.0	aA	0.3	aA	3.5	aA	49.2	b
PAx163 38	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 218	0.0	aA	0.3	aA	3.5	aA	49.2	b
PAx163 39	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 93	0.0	aA	0.3	aA	3.5	bB	49.2	b
PAx163 4	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 272	1.4	aA	1.4	aA	5.0	bB	109.2	c
PAx163 40	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 275	0.0	aA	3.5	bB	3.5	bB	126.0	c
PAx163 41	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 56	0.0	aA	3.5	bB	3.5	bB	126.0	c
PAx163 42	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 111	0.3	aA	3.5	bB	3.5	bB	129.6	c
PAx163 44	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 69	0.0	aA	0.3	aA	10.3	cB	131.2	c
PAx163 45	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 231	0.3	aA	3.5	bB	8.0	cC	183.6	d
PAx163 46	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 105	3.9	bA	3.9	bA	3.9	bA	188.8	c
PAx163 47	0.0	aA	0.2	aA	0.2	aA	0.0	a	PAx163 262	1.3	aA	3.8	bB	7.7	bC	199.2	c
PAx163 48	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 12	0.0	aA	2.4	bB	12.7	dC	210.9	d
PAx163 49	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 115	0.2	aA	3.9	bB	10.3	cC	220.8	d
PAx163 5	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 141	3.9	bA	3.9	bA	8.8	cB	247.6	d
PAx163 50	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 249	1.4	aA	5.0	cB	10.3	cC	260.4	d
PAx163 51	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 281	0.0	aA	3.5	bB	15.0	dC	264.0	d
PAx163 52	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 62	0.0	aA	3.5	bB	15.0	dC	264.0	d
PAx163 53	0.3	aA	0.3	aA	0.3	aA	0.0	a	PAx163 27	2.3	bA	5.4	cB	10.1	cC	279.6	d
PAx163 55	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 176	0.3	aA	6.5	cB	12.7	dC	311.6	d
PAx163 57	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 67	0.3	aA	0.3	aA	27.7	fB	342.8	e
PAx163 6	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 76	0.0	aA	8.0	dB	15.0	dC	372.0	e
PAx163 60	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 221	0.3	aA	10.3	dB	10.3	cB	375.6	e
PAx163 61	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 31	0.3	aA	8.0	dB	15.0	dC	375.6	e
PAx163 63	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 34	0.0	aA	5.0	cB	21.3	eC	376.0	e
PAx163 64	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 58	0.0	aA	5.0	cB	21.3	eC	376.0	e
PAx163 65	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 37	0.3	aA	8.0	dB	21.3	eC	451.6	e
PAx163 66	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 217	0.0	aA	12.7	eB	27.7	fC	636.0	f
PAx163 68	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 14	3.5	bA	12.7	eB	27.7	fC	678.0	f
PAx163 7	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 198	0.3	aA	10.3	dB	43.0	hC	767.6	f
PAx163 70	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 214	0.3	aA	15.0	fB	34.0	gC	771.6	f
PAx163 71	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 74	1.4	aA	10.3	d	43.0	hC	780.4	f
PAx163 72	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 211	0.0	aA	15.0	fB	45.7	hC	908.0	f
PAx163 75	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 109	0.3	aA	12.7	eB	52.0	iC	931.6	f
PAx163 79	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 22	0.3	aA	12.7	eB	64.7	jC	1083.6	g
PAx163 8	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 23	0.3	aA	12.7	eB	64.7	jC	1083.6	g
PAx163 80	0.0	aA	0.0	aA	0.0	aA	0.0	a	PAx163 54	1.3	aA	16.3	eB	56.7	hC	1087.2	g

continued...

continuation...

PAx163 81	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 82	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 83	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 84	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 85	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 86	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 87	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 88	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 89	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 90	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 91	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 94	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 95	0.0	aA	0.0	aA	0.0	aA	0.0	a
Pêra de Abril	0.0	aA	0.0	aA	0.0	aA	0.0	a
TMxLP163	0.0	aA	0.0	aA	0.0	aA	0.0	a
PAx163 237	0.0	aA	0.0	aA	0.3	aA	3.6	a
PAx163 103	0.1	aA	0.1	aA	0.1	aA	4.8	a
PAx163 201	0.1	aA	0.1	aA	0.1	aA	4.8	a
PAx163 205	0.1	aA	1.4	aA	5.0	aA	9.4	b
PAx163 182	0.2	aA	0.2	aA	0.2	aA	9.6	a
PAx163 274	0.2	aA	0.3	aA	0.3	aA	13.2	a
PAx163 129	0.3	aA	0.3	aA	0.3	aA	14.4	a
PAx163 207	0.3	aA	0.3	aA	0.3	aA	14.4	a
PAx163 230	0.3	aA	0.3	aA	0.3	aA	14.4	a
PAx163 43	0.3	aA	15.0	fB	61.0	jC	1095.6	g
PAx163 259	1.4	aA	15.0	fB	61.0	jC	1108.4	g
PAx163 118	1.4	aA	12.7	eB	73.7	kC	1204.4	g
PAx163 199	0.3	aA	15.0	fB	80.0	lC	1323.6	h
PAx163 202	0.3	aA	15.0	fB	80.0	lC	1323.6	h
PAx163 32	0.3	aA	15.0	fB	80.0	lC	1323.6	h
PAx163 99	0.3	aA	15.0	fB	80.0	lC	1323.6	h
PAx163 229	1.2	aA	21.3	gB	70.7	mC	1374.0	h
PAx163 191	3.5	bA	21.3	gB	70.7	kC	1402.0	h
PAx163 216	0.0	aA	15.0	fB	90.0	mC	1440.0	h
PAx163 120	0.3	aA	21.3	gB	83.3	lC	1515.6	h
PAx163 19	5.4	bA	27.7	hB	73.7	kC	1613.2	i
PAx163 268	0.3	aA	34.3	hB	67.3	jC	1635.2	h
PAx163 106	0.3	aA	27.7	hB	86.7	mC	1707.6	i
PAx163 227	0.3	aA	27.7	hB	86.7	m	1707.6	i
PAx163 2	0.3	aA	34.0	iB	76.0	kC	1731.6	i
PAx163 131	3.5	bA	27.7	hB	86.7	mC	1746.0	i
Murcott	3.5	bA	34.0	iB	83.3	lC	1858.0	j
PAx163 17	3.5	bA	34.0	iB	90.0	mC	1938.0	j
PAx163 256	0.3	aA	36.7	iB	89.0	mC	1951.6	j
PAx163 3	0.3	aA	34.0	iB	97.0	mC	1983.6	j
Dancy	5.0	bA	43.0	jB	86.7	mC	2132.0	j
PAx163 98	0.3	aA	52.0	kB	94.7	mC	2387.6	j

[1]Means followed by the same letters in the column (lower) and in the line (capital) do not differ by the Scott-Knott test ($p > 0.05$).

TABLE 2 - Estimated genetic parameters based on AUDPC values of 235 F_1 hybrid progeny crossing between Pêra de Abril sweet orange vs TM x LP 163.

Genetic parameters	AUDPC
h^2_g	0.91 ± 0.10
CVgi (%)	228.25
CVe (%)	70.88
Mean	215.86

[1]Legend: h^2_g heritability of individual parcels in the broad sense, ie, the genotypic effects; *CVgi* (%): genotypic coefficient of variation in percent; *CVe* (%) environmental variation coefficient as a percentage; Mean: overall mean of the experiment.

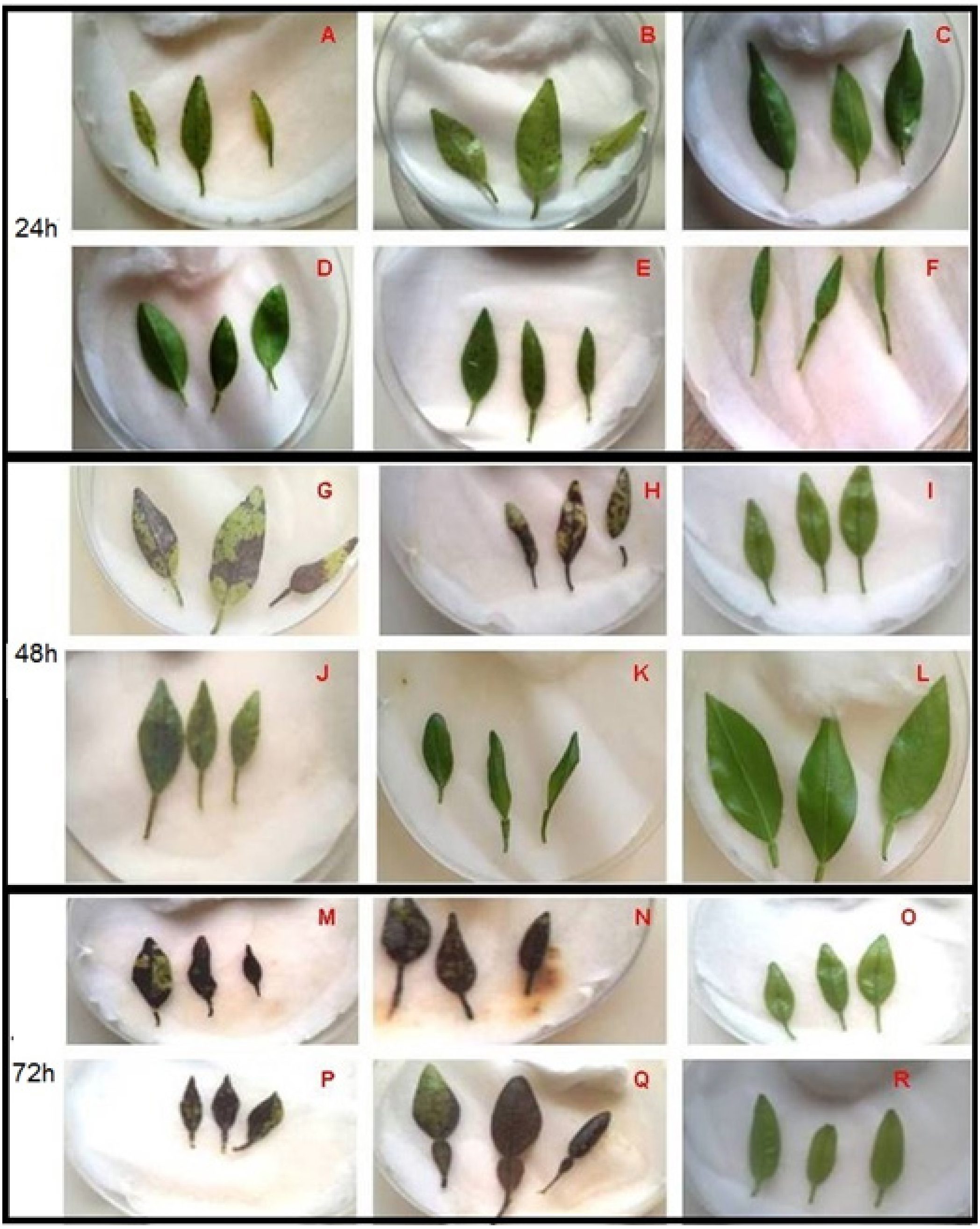

FIGURE 1- Leaves of hybrids showing typical lesions caused by *A. alternata:* 24 h after inoculation. A - Dancy (Susceptible - S), B - Murcott (S), C - Fremont (Resistant - R), D - PAx163 85 (R), E - PAx163 67 (S), F- PAx163 131 (S). 48 h after inoculation, G - Dancy, H - Murcott, I - Fremont, J - PAx163 37 (S), K - PAx163 69 (S) e L - PAx163 269 (R). 72 h after inoculation, M - Dancy, N - Murcott, O - Fremont, P - PAx163 67 (S), Q - PAx163 268 (S) and R - PAx163 80 (R).

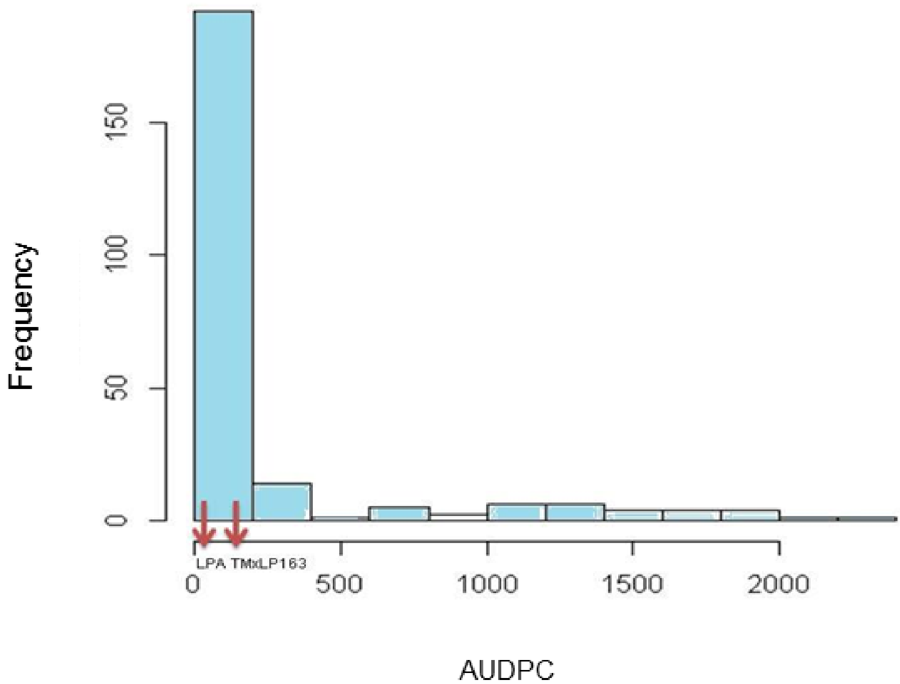

FIGURE 2- Histogram of frequency distribution of AUDPC means of 235 hybrids evaluated.

CONCLUSION

The frequency of segregation observed (165R:70S) and high heritability ($h^2_g = 0.91$) suggest that few genes may be involved in controlling the inheritance of ABS resistance in citrus.

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