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Variation in the response of melon genotypes to *Fusarium oxysporum* f.sp. *melonis* race 1 determined by inoculation tests and molecular markers

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Screening of genotypes of melon (*Cucumis melo*) for resistance to wilt caused by *Fusarium oxysporum* f.sp. *melonis* is often characterized by wide variability in their responses to inoculation, even under carefully controlled conditions. The variability at the seedling stage of 17 genotypes susceptible to race 1 was examined in growth-chamber experiments. Disease incidence varied from 0 to 100% in a genotype-dependent manner. Using four combinations of light (60 and 90 μE m^{-2} s^{-1}) and temperatures of (27 and 31°C), only light intensity showed a statistically significant effect. Marker-assisted selection for fusarium resistance breeding using cleaved amplified polymorphic sequence (CAPS) and sequence-characterized amplified region (SCAR) markers were compared using a single set of genotypes that included 24 melon accessions and breeding lines whose genotype regarding the *Fom-2* gene was well characterized. The practical value of the markers for discriminating a range of genotypes and clarifying the scoring of phenotypes was also tested using a segregating breeding population which showed codominant SCAR markers to be useful in marker-assisted selection.

Keywords: fusarium wilt, molecular markers, resistance

Introduction

Fusarium wilt of melons, caused by *Fusarium oxysporum* f.sp. *melonis*, is a limiting factor for melon production worldwide (Mas *et al*., 1981; Martyn & Gordon, 1996). Four races of the pathogen have been described, designated 0, 1, 2 and 1–2 (Mas *et al*., 1981). The most effective means of controlling this soilborne disease is to use resistant cultivars (Martyn & Gordon, 1996), and two wilt resistance genes, *Fom-1* and *Fom-2*, have been identified in melons (Mas *et al*., 1981; Martyn & Gordon, 1996). *Fom-1* confers resistance to races 0 and 2 of the pathogen, *Fom-2* confers resistance to races 0 and 1, and the two genes are extensively used in breeding programmes. Successful breeding depends on the availability and quality of resistance sources, and on effective methods for inoculating the plants and assessing their response to the disease. The expression of fusarium wilt symptoms following artificial inoculation is affected by the virulence of the pathogen isolates (Namiki *et al*., 1998), the genetic background of the plant (Mas *et al*., 1981), and environmental factors such as temperature and light intensity (Cohen *et al*., 1996). Ambiguous disease expression results of inoculation tests may lead to the inefficient selection of individual resistant plants, which could delay the breeding programmes. Like other breeders, this research group has encountered situations in which inadequate discrimination between genetically resistant and 'escaping' plants resulted in the loss of fusarium resistance during the breeding process (Y.B. and R.C., unpublished results). Molecular markers linked to the desired genes might alleviate this problem by ensuring the chromosome segment that carries the resistance gene is introgressed and maintained even when the inoculation test is not very reliable.

An additional advantage of molecular markers relates to the possibility of simultaneous selection of genes for resistance to two or more pathogens, or pathogen races. Inoculation with one *F. oxysporum* f.sp. *melonis* race may confer cross-protection (Mas *et al*., 1981; Alabouvette & Couteaudier, 1992; Freeman *et al*., 2001), thus impeding further selection of the same plant for resistance to an additional race. For example, it was found that melon plants that survived inoculation with race 1 did not show any disease symptoms following subsequent inoculation with race 2 (R.C., unpublished results).

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Several molecular markers linked to the *Fom-2* gene have been described recently. A RAPD fragment, E07-1·25, was derived from the susceptible cultivar Vedrantais, and was linked at a distance of 1·6 cM to the *Fom-2* gene in a segregating F_2 population that was developed from the cross Vedrantais × PI 161375 (Baudracco-Arnas & Pitrat, 1996). A cleaved amplified polymorphic sequence (CAPS) marker was developed from the E07-1·25 RAPD fragment by Zheng *et al.* (1999), who cloned and sequenced the RAPD fragment from both parents, and found that the PCR-amplicon from the susceptible parent could be digested with restriction endonuclease *Bcl*I, while the resistant-parent allele could be digested with *Bss*SI. A codominant CAPS marker was obtained only by combining both restriction enzymes. Wang *et al.* (2000) reported the isolation of an AFLP AGG/CCC fragment that cosegregated with *Fom-2* in a population of 60 BC1 plants derived from the cross between the genotypes MR-1 (resistant) and Ananas Yoqne'am (susceptible). Another codominant marker based on sequence-characterized amplified regions (SCARs), designated AM, was developed by specific amplification of marker alleles that ranged in size between the sizes of the resistant and susceptible parents. Similarly, another SCAR, namely FM, was developed by the same authors on the basis of a cosegregating RAPD band, 596-1. The efficiency of these markers is compared here for the first time in the same genetic material.

The objectives of the present study were: (i) to document the variation in the response of well defined melon genotypes to inoculation with *F. oxysporum* f.sp. *melonis* race 1; (ii) to demonstrate the difficulties in screening for fusarium resistance by means of an inoculation test, as compared with the use of molecular markers; and (iii) to compare the potential of *Fom-2*-linked molecular markers as tools for the unambiguous selection of resistant plants.

Materials and methods

Response of melon genotypes to fusarium wilt and to pathogen isolates

Twenty-four melon genotypes, representing various PI accessions, cultivars and breeding lines (mostly of the Galia market type) that are commonly used in this research group's breeding programmes, were selected for this study (Table 1). The response of these lines to *Fusarium oxysporum* f.sp. *melonis* race 1 had been characterized previously (Table 1). Seeds of the tested genotypes were

Table 1 Melon genotypes and their source

Genotype name and market type	Abbreviation	Resistance genes[a]	Seed source
Noy Amid (Yellow canary)	NA	None (susceptible)	Cultivar (Hazera, Israel)
Noy Yizre'el (Ha'Ogen)	NY	None (susceptible)	Cultivar (Hazera, Israel)
Krymka (Galia)	KRY	None (susceptible)	Breeding line (ARO)[b]
Ein Dor (Ananas)	ED	None (susceptible)	Cultivar (Hazera, Israel)
Eshkolit Ha'Amaqim (Galia)	ES	None (susceptible)	Cultivar (ARO)
Ananas Yoqne'am (Ananas)	AY	None (susceptible)	Cultivar (Hazera, Israel)
Arava (Galia) F_1 Hybrid	AR	None (susceptible)	Cultivar (Hazera, Israel)
Freeman cucumber (pickling melon)	FRC	*Fom-2*	Breeding line (ARO)
PI 161375	PI	*Fom-2*	Breeding line (ARO)
F65 (Galia)	F65	*Fom-2*	Breeding line (ARO)
I4-6-2-B (Galia)	I4	*Fom-2*	Breeding line (ARO)
Védrantais (Charentais)	VED	*Fom-1*	Cultivar (INRA, France)
Dulce (American cantaloupe)	DUL	*Fom-1*	Cultivar USA
FM 025 (Galia)	FM 025	*Fom-1*	Breeding line (ARO)
FM 018 (Galia)	FM 018	*Fom-1*	Breeding line (ARO)
FM 004 (Galia)	FM 004	*Fom-1*	Breeding line (ARO)
FM 023 (Galia)	FM 023	*Fom-1*	Breeding line (ARO)
FM 014 (Galia)	FM 014	*Fom-1*	Breeding line (ARO)
FM 024 (Galia)	FM 024	*Fom-1*	Breeding line (ARO)
Doublon (Charentais)	DOU	*Fom-1*	Cultivar (INRA, France)
Hemed (Ha'Ogen)	HEM	*Fom-1*	Commercial cultivar (Hazera, Israel)
Maqdimon (Galia) F_1 Hybrid	MK	Heterozygous for *Fom-2*	Commercial cultivar (Hazera, Israel)
Omega (5080) (Galia) F_1 Hybrid	Omega	Heterozygous for *Fom-1*, *Fom-2*	Commercial cultivar (Nunza, the Netherlands)
Caruso (5093) (Galia) F_1 Hybrid	Caruso	Heterozygous for *Fom-1*, *Fom-2*	Commercial cultivar (Nunza, the Netherlands)

[a]Susceptible, relates to races 0, 1 and 2; *Fom-1*, resistant to races 0 and 2; *Fom-2*, resistant to races 0 and 1.
[b]ARO, Agricultural Research Organization, Israel.

sown in sandy soil. Two days after emergence, the seedlings were removed from the soil and washed under running tap water. The roots were pruned to approximately half length, and seedlings were inoculated by dipping the roots in a conidial suspension (10^6 conidia mL^{-1}) of *F. oxysporum* f.sp. *melonis* race 1 for 2 min. The pathogens used were isolated from diseased melon plants grown in Bet Netufa Valley in northern Israel. The pathogens were maintained on yeast extract medium at 27°C in the dark. Conidial suspensions for seedling inoculation were prepared by macerating 1-week-old cultures with 100 mL water (Cohen *et al*., 1996). Inoculated plants were transplanted into 250 mL pots containing new, disease-free, sandy soil. The inoculated plants were maintained in the growth room under four different combinations of conditions: 27°C and light intensity of 60 μE m^{-2} s^{-1}, 31°C and 60 μE m^{-2} s^{-1}, 27°C and 90 μE m^{-2} s^{-1}, and 31°C and 90 μE m^{-2} s^{-1}. For each combination of genotype and environmental regime, 35 plants were grown (seven plants per pot × five pots). The number of wilted plants was recorded twice a week and disease incidence was calculated. The effects of light intensity and temperature on disease incidence in each cultivar were analysed using the Student–Newman–Keuls multiple range test.

The main effects of all factors (genotype, light intensity and temperature) on disease incidence, and their interactions, were analysed by applying three-way ANOVA by means of the SUPER ANOVA software on a Macintosh computer. The effects of different pathogen isolates on disease incidence were also evaluated. Ten susceptible cultivars and breeding lines (NA, ED, AY, VED, DOU, KRY, DUL, ES, HEM, NY; Table 1) were inoculated with two different isolates of *F. oxysporum* f.sp. *melonis* race 1. Inoculated plants were maintained in a growth chamber at 31°C and 90 μE m^{-2} s^{-1}. Each cultivar by isolate combination included 35 plants, comprising five replicates (pots), each containing seven plants. The experiment was terminated 21 days after inoculation, and disease incidence was recorded and analysed using the Student–Newman–Keuls multiple range test.

Comparison of selection efficacy based on an inoculation test and a molecular marker

A Galia-type breeding population, obtained by selfing an individual plant heterozygous for the *Fom-2* locus during a breeding programme (106 plants), was inoculated with *F. oxysporum* f.sp. *melonis* race 1 following the above procedure. The plants were maintained in a growth chamber at 31°C and 90 μE m^{-2} s^{-1}. The number of wilted plants and of those showing leaf necrosis was recorded. All plants were sampled for DNA isolation and for marker analysis, in order to compare the results of the DNA tests with those of the phenotypic evaluation.

Molecular marker analysis

The markers applied in this study were: (i) the CAPS markers (E07 cut with either *Bcl*1 or *Bss*S1) as described by Zheng *et al*. (1999); and (ii) two SCAR markers (AM and FM) as described by Wang *et al*. (2000).

DNA for PCR amplification was isolated from all the melon genotypes, either from root tissue by means of a mini-procedure described by Fulton *et al*. (1995), or from leaf tissue using the small-scale method described by Wang *et al*. (1993). Thirty nanograms of plant genomic DNA were used for all PCR analyses, and the PCR protocols were according to Zheng *et al*. (1999) and Wang *et al*. (2000), respectively.

Results

Variable response of melon genotypes to fusarium wilt under different environmental conditions

The responses of 24 melon genotypes (Table 1) to fusarium wilt were evaluated under four different combinations of temperature and light intensity. The seven resistant hybrids and breeding lines, with the *Fom*-2 gene, were fully resistant to the pathogen under all four regimes (Fig. 1F).

Major differences in disease incidence among the 17 susceptible genotypes tested were observed in each of the environmental combinations (Table 2). Overall, the genotypes exhibited a very wide range of disease incidence (0–100%). For example, cultivars NA and ED were highly susceptible, and exhibited typical wilting symptoms under all four environmental regimes (Fig. 1), whereas most of the other genotypes exhibited low levels of disease incidence, at least under certain conditions, despite their documented lack of *Fom-2* resistance alleles. In addition, 10 susceptible melon cultivars were inoculated with two race 1 isolates to confirm that the response variability was a genetic phenomenon and not due to variations in isolate virulence. Disease incidence ranged from 97–100% in Noy Amid (NA) to 6–13% in Noy Yizre'el (NY). This disease incidence variability among the melon genotypes, and its trend from the most susceptible to the relatively tolerant ones, was maintained with the two fungal isolates tested. Plants varied in their cotyledon or leaf necrosis symptoms, and not all wilted (Fig. 1b–e). Analysis of the variance of the effects of genotype, temperature and light intensity revealed that the effect of temperature was not significant, whereas that of light intensity was statistically significant (Table 3). The interaction between genotype and temperature was also highly significant. Statistical analysis of the effect of the environmental conditions on disease incidence of each genotype (Table 2) revealed that six out of the 17 susceptible genotypes tested (FM 004, VED, HEM, DUL, AR and AY) exhibited a significant effect of the environmental conditions on disease incidence. The effect, however, varied among genotypes, which demonstrated the genotype–environment interaction. For example, the lowest disease incidence for HEM was evident under 31°C and 60 μE m^{-2} s^{-1}, the environmental regime associated with the highest disease incidence in AR. Also in the other susceptible genotypes, differences of up to 30% between treatments were found, although they were not statistically significant.

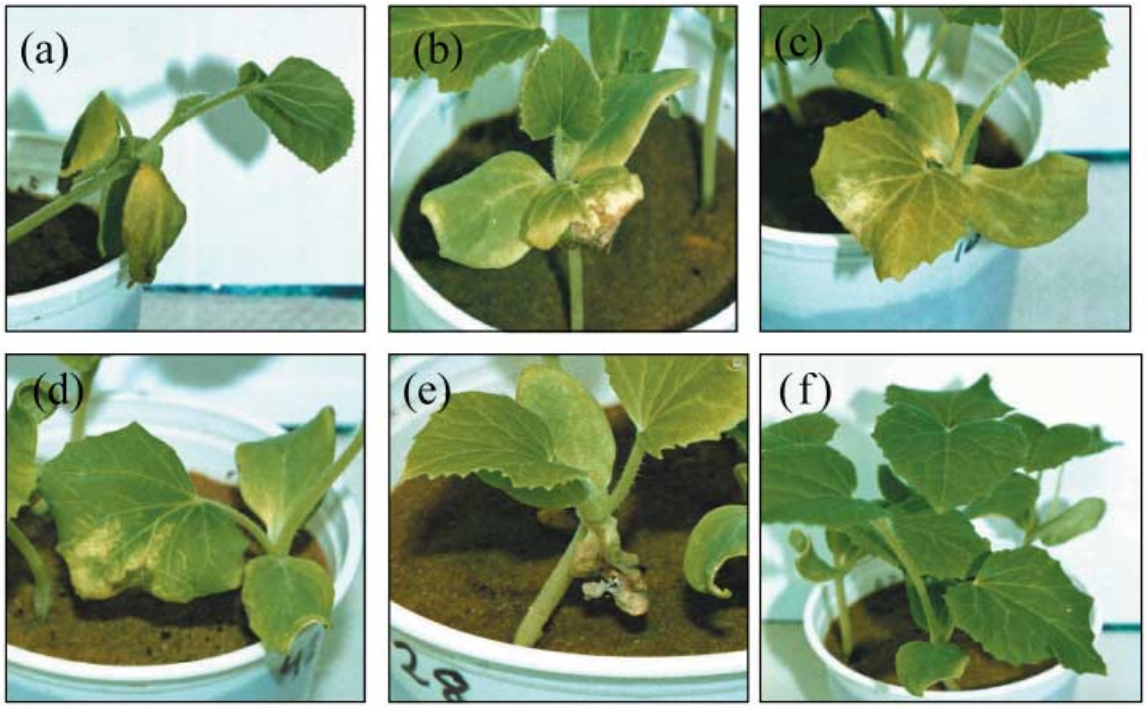


Figure 1 Symptoms caused by *Fusarium oxysporum* f.sp. *melonis* race 1 in melon under artificial inoculation. (a) Typical wilting of the susceptible cultivar Ananas Ein Dor (ED). (b–e) Non-wilting plants, identified as susceptible by the markers, exhibiting various degrees of cotyledon and leaf necrosis symptoms. (f) Symptomless plants that are either genetically resistant or 'escapes'.

Table 2 Disease incidence (%) of susceptible melon genotypes (number ≈35), inoculated with *Fusarium oxysporum* f.sp. *melonis* race 1 under four different environmental conditions

Temp. (°C)	Light intensity (μE m^{-2} s^{-1})	Disease incidence (%)[a]																
		NA[b]	NY	KRY	ED	ES	AY	AR	VED	DUL	025	018	004	023	014	024	DOU	HEM
31	60	92a	11a	62a	96a	26a	86a	45a	80a	65ab	51a	12a	13b	11a	24a	40a	80a	3b
31	90	100a	47a	59a	97a	32a	58b	41a	82a	83a	66a	43a	56a	27a	41a	13a	nt[c]	41a
27	60	97a	28a	54a	84a	50a	96a	0b	35b	41b	43a	30a	35ab	28a	28a	16a	65a	13b
27	90	97a	24a	53a	93a	63a	72a	11b	63a	50b	56a	43a	39ab	36a	45a	34a	61a	19b

[a]In each column, disease incidence values marked with the same letter are not significantly different (P = 0·05). The hybrids Makdimon (MK), Omega and Caruso, and the lines Freeman cucumber (FRC), PI 161375, F65 and I4-6-2-B, having the gene *Fom-2* (Table 1), were completely resistant, exhibiting no wilting under any environmental conditions.
[b]Letters refer to genotype names; numbers refer to FM breeding lines.
[c]nt, not tested.

Table 3 Analysis of variance for the effect of genotype, light intensity and temperature on *Fusarium* wilt incidence in melon seedlings

Source	df	SS	MS	F	P
Genotype	15	182404·50	12160·04	35·0	0·0001
Light	1	7003·19	7003·19	20·18	0·0001
Temperature	1	1094·43	1094·43	3·15	0·0769
Light × genotype	15	9333·80	622·25	1·79	0·0359
Genotype × temperature	15	22832·40	1522·16	4·38	0·0001
Light × temperature	1	97·59	97·59	0·28	0·5963
Light × temperature × genotype	15	9823·08	654·87	1·88	0·0248
Residual	253	87783·75	346·97		

Table 4 Expression of symptoms in a susceptible and resistant cultivar and segregating population of melon inoculated with *Fusarium oxysporum* f.sp. *melonis* (race 1) and genotype analysis using SCAR markers

Host	Symptoms	Number of plants			Expected ratio	χ^2	P
		Susceptible	Resistant[a]				
Ein Dor (susceptible)	Wilting	21	3				
Maqdimon (resistant)	Wilting	0	17				
Segregating population	Wilting	8	98		1 : 3	17·22	<0·001
	Wilting + leaf necrosis	13	93		1 : 3	9·17	<0·01
	Genotyping with SCAR markers	*fom-2* / *fom-2*[b]	*fom-2* / *Fom-2*	*Fom-2* / *Fom-2*			
	AM and FM	23	53	30	1 : 2 : 1	0·94	0·65

[a]Plants showing no symptoms (resistant or escaping plants).
[b]*Fom-2*, resistant allele; *fom-2*, susceptible allele.

Use of molecular markers for genotype identification in the response of the *Fom-2* locus

Four markers that had been found in previous, separate studies to be associated with resistance to *Fom-2* were compared for the first time in one study. The 24 genotypes were scored with each of these markers (Fig. 2).

CAPS Marker E07

DNA of each of the 24 genotypes was amplified using the E07-specific primers, followed by digestion with either *Bcl*1 or *Bss*S1. According to Zheng *et al.* (1999), the resistant parent allele is cleaved by *Bss*S1. However, of the four resistant breeding lines tested in the present study, only PI 161375 could be digested by this enzyme. The CAPS band was therefore cleaved with *Bcl*I, which specifically digested the susceptible parent and obtained a dominant marker that differentiated the homozygous resistant genotypes from plants that were either susceptible or heterozygous (Fig. 2a). All four homozygous resistant genotypes were successfully identified by this assay, indicating the applicability of this marker.

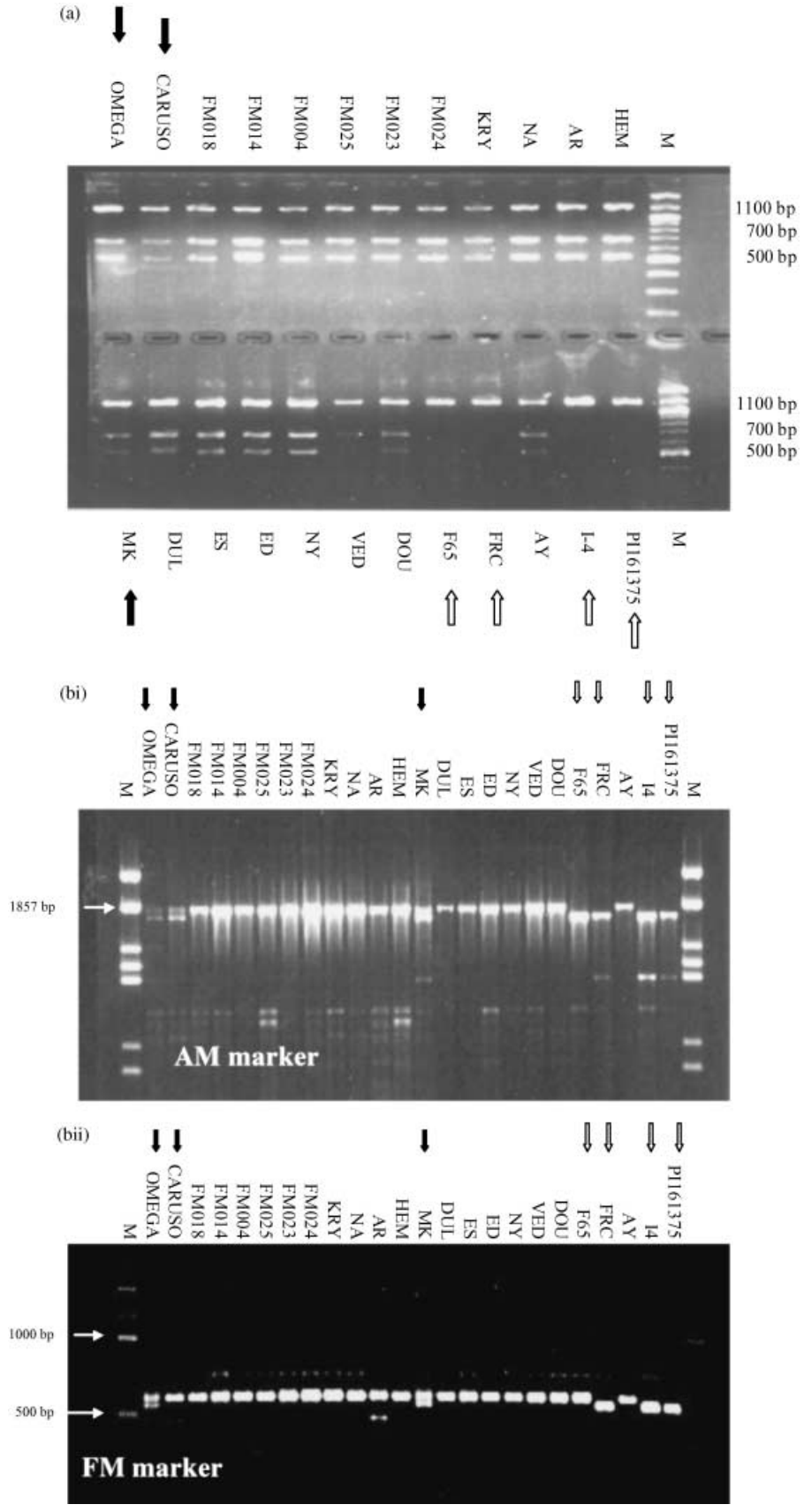
SCAR markers FM and AM

The FM marker was capable of identifying all homozygous and heterozygous genotypes that presented the susceptible allele for this marker (Fig. 2b), except for the resistant line F65 and the F_1 hybrid Caruso derived from it. The AM marker, on the other hand, was the most discriminatory: it identified all the resistant genotypes and also separated the homozygous from the heterozygous ones (Fig. 2b). The only disadvantage encountered with this marker involved DNA extraction; the small-scale method described above was used successfully for the FM primer, but was not efficient with this marker, probably because of its large size (*c.* 1700 bp). This necessitated the use of the more laborious mini-procedure for DNA isolation.

Use of markers in a segregating breeding population

A total of 106 plants from a segregating breeding population were inoculated with *F. oxysporum* f.sp. *melonis* race 1. Only eight plants exhibited typical wilt symptoms (Fig. 1a), and five other plants exhibited various leaf and cotyledon necrosis symptoms but were not wilted (Fig. 1b–e). The inoculation results obtained did not fit the expected 3 : 1 segregation ratio (Table 4), demonstrating once again the reliability problem inherent in using inoculation results for the selection procedure. Two tightly linked molecular markers were used to assess the genotypes of the plants around the *Fom-2* locus. Each of the 106 individuals of the tested population was scored for the FM and AM SCAR markers. The genotypic scores obtained with the two markers were compatible: according to the molecular test, 23 plants were identified as susceptible, while 53 and 30 plants were found to be heterozygous-resistant and homozygous-resistant, respectively. Ten plants out of the 93 that were identified as resistant on the basis of wilting and necrosis symptoms were, in fact, very likely to be 'escapes', as indicated by the

Figure 2 Marker-assisted selection of 24 melon genotypes. (a) E07-CAPS marker. PCR products were obtained by using E07-specific primers, digested with *Bcl*I restriction enzyme and separated on 1% agarose gel. A single, uncut PCR product identified the four homozygous-resistant genotypes, while digested products identified either the heterozygous-resistant or the susceptible lines. M, 100 bp ladder size markers. Genotypes that are homozygous-resistant to fusarium race 1 are indicated by white arrows; heterozygous-resistant by black arrows. (b) SCAR markers (i) AM and (ii) FM. M, molecular weight standards: PBR322 DNA digested with *Alw*44I and *Mva*I. AM differentiates all resistant genotypes from all susceptible ones; FM failed to differentiate resistant lines F65 and Caruso. Genotypes that are homozygous-resistant to race 1 are indicated by white arrows; heterozygous-resistant by black arrows.


(a)
OMEGA
CARUSO
FM018
FM014
FM004
FM025
FM023
FM024
KRY
NA
AR
HEM
M
1100 bp
700 bp
500 bp
1100 bp
700 bp
500 bp
MK
DUL
ES
ED
NY
VED
DOU
F65
FRC
AY
I-4
PI161375
M
(bi)
1857 bp
AM marker
(bii)
1000 bp
500 bp
FM marker

tightly linked markers. In other backgrounds the escape rate would have been even higher (Table 4).

Discussion

Environmental factors can play an important role in the development of fusarium wilt in melons, both in the field and following artificial inoculation; however, it is difficult to discern the influence of each factor (Mas *et al*., 1981). Horsfall & Dimond (1957) suggested that some wilt diseases are 'low-sugar diseases' while others are 'high-sugar diseases', that is, susceptibility to them is enhanced by low or high sugar content in the tissue, respectively. Light intensity was used by Cohen *et al*. (1996) to manipulate sugar levels in plants: disease incidence was significantly higher in melon plants grown under reduced light conditions than in those grown under high light intensity, which suggested that fusarium wilt of melon is a low-sugar disease. In the present study, however, the environmental effect on disease incidence was not consistent among all the genotypes tested. Consequently, it is not possible to suggest a combination of temperature and light intensity that would enhance the expression of disease symptoms, and so facilitate screening for disease resistance, in all the genotypes. It seems that the effects of light intensity and other environmental factors are complicated and also depend on the genetic background of the genotype tested. In addition, the present results suggest that the response of the various genotypes to the disease was not affected by possible variations in the virulence of the isolates. For example, the NA and ED cultivars, which are known to be very susceptible, exhibited 84–100% wilting under all the environmental conditions tested. Cultivar ED served as a reliable susceptible control in all experiments and breeding programmes.

The relatively low disease incidence in many of the susceptible genotypes and breeding lines cannot be explained on the basis of currently available knowledge about the genetic control of resistance to races 0 and 1 of *F. oxysporum* f.sp. *melonis*. A possible explanation is that modifier genes might segregate in the susceptible genotypes, and thus play a role in the response to inoculation and cause variation among incidental plants. In presumably pure lines that would not be expected to segregate, infection phenomena may have accounted for the differences between wilting and nonwilting individuals. There may be a 'physiological threshold' that must be overcome by the pathogen in order to cause the collapse of the susceptible plants, and in those plants with less-susceptible genetic backgrounds, minor environmental or internal fluctuations might determine whether such a threshold is crossed.

Marker-assisted selection is proving to be a useful tool in breeding for disease resistance. In this study four markers were applied to a diverse collection of 24 melon genotypes that included homozygous-resistant, heterozygous-resistant, and susceptible genotypes. The RAPD marker (E07) that was tightly linked to *Fom-2* (1·6 cM) was tested in a diverse array of melon genotypes by Zheng & Wolf (2000), who showed that this marker could discriminate successfully among most genotypes. Zheng *et al*. (1999) reported that the CAPS marker derived from E07 displayed codominant segregation when two restriction enzymes were used, but failed to recognize several resistant genotypes due to incomplete digestion, regardless of restriction reaction conditions. As suggested by Zheng *et al*. (1999), the incomplete digestion of the PCR amplicon probably resulted from the amplification of a mixture of related sequences by these PCR primers. At least half the resistant or heterozygous genotypes tested by Zheng *et al*. (1999) were not digested by *Bss*SI. In the present study, the restriction enzyme *Bss*SI could digest only the DNA of PI 161375, and not that of the other three homozygous-resistant genotypes tested. Only one restriction enzyme, *Bcl*I, was therefore applied. This enzyme digested the susceptible allele, allowing the identification of all the homozygous-resistant genotypes but not the separation of the heterozygous-resistant plants from the susceptible ones (Fig. 2). Selection of only homozygous-resistant plants and omission of the heterozygous-resistant ones could lead to the loss of useful genotypes during the breeding cycles. Moreover, the CAPS marker assay requires both PCR and restriction digestion, and this extra step could be a disadvantage in a large-scale breeding programme.

The codominant SCAR markers AM and FM, which are the most tightly linked markers reported, are simpler to analyse and resulted in good identification of the genotypes tested (Wang *et al*., 2000). Of the genotypes tested in the present study, the FM marker failed to identify the F65 breeding line and its derivative, the F_1 hybrid Caruso, whereas the AM marker successfully identified all the homozygous- and heterozygous-resistant genotypes. A possible recombination between the two markers and the gene should not be ignored, although low misprediction rates were demonstrated for both of them (Wang *et al*., 2000).

The comparison between phenotypic scoring of a segregating population and molecular evaluation of the genotype highlights the difficulty in identifying resistant plants on the basis of inoculation alone. Plants that might have been considered resistant on the basis of disease evaluation were identified as susceptible by the molecular test. Such a mistake could lead to a loss of resistance or to a delay in a breeding programme.

Markers to be used in marker-assisted selection need to be simple, fast and cost-effective. The data presented here suggest that the FM marker is preferable in breeding programmes that do not include line F65 as a parent, whereas the AM marker should be used if F65 is a chosen resistant parent.

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