New Phytologist

Supporting Information Figs S1–S6 and Tables S1–S5

Article title: **Comparative genomics of *Fusarium oxysporum* f. sp. *melonis* reveals the secreted protein recognized by the *Fom-2* resistance gene in melon**

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The following Supporting Information is available for this article:

Fig. S1 Fom001 is a race 1 isolate.

Fig. S2 *Fom* isolates cluster according to VCG and geographic location, but not according to race.

Fig. S3 PCR of *Fom* effector candidate genes on genomic and cDNA.

Fig. S4 Protein alignment of AvrFom2, *Colletotrichum higginsianum* ChEC13 and *Pyrenophora tritici-repentis* ToxA.

Fig. S5 Melon plants harbouring the *Fom-2* resistance gene are resistant to race 2 isolates that are genetically complemented with *AVRFOM2*.

Fig. S6 *AVRFOM2* is absent in race 1,2 isolates.

Table S1 Confirmation of *Fom* races

Table S2 Coverage of the Fom001 reference genome by the Illumina paired-end sequencing reads of each Fom isolate

Table S3 Reported SNPs for sequence read mapping to Fom004 genome

Table S4 Average read depth coverage of candidate effector genes

Table S5 Primers used in this study

Table S6 Sequence read average coverage of Fom001 contigs (see separate Excel file)

Fig. S1 Fom001 is a race 1 isolate. Ten-day-old melon seedlings without (Cha-T) or with either the *Fom-1* (Cha-Fom1) or the *Fom-2* (Cha-Fom2) resistance gene were inoculated with conidia of Fom001 (NRRL26406). Pictures were taken 10 d after inoculation. Bars, 15 cm.

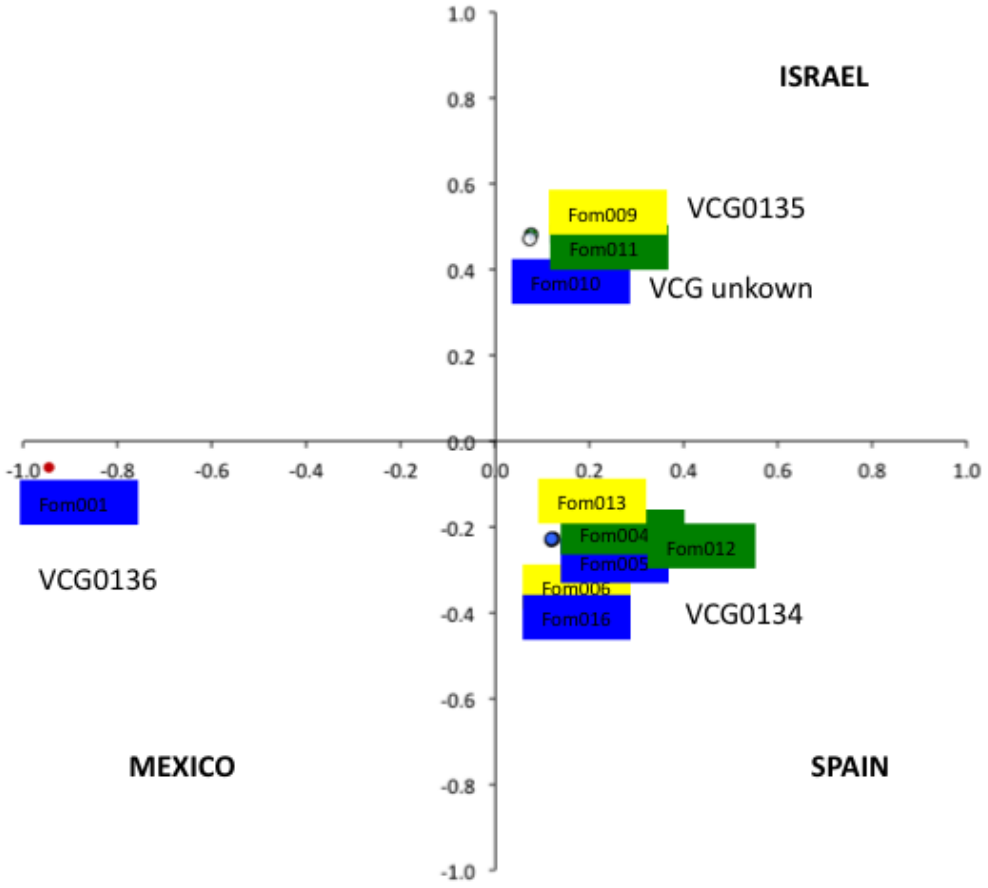

Fig. S2 *Fom* isolates cluster according to VCG and geographic location, but not according to race. The Illumina paired-end sequencing reads were mapped to the Fom004 genome using BWA (version 0.7.4-r385 mem) (Li, 2013). Single-nucleotide polymorphisms were called using GATK (see the Materials and Methods section). The variant positions were merged with Tabix v0.2.6 bgzip, tabix, and VCFtools v0.1.9 vcf-merge, to conduct a Princincipal Components Analysis (PCA) using smartPCA version 9102.

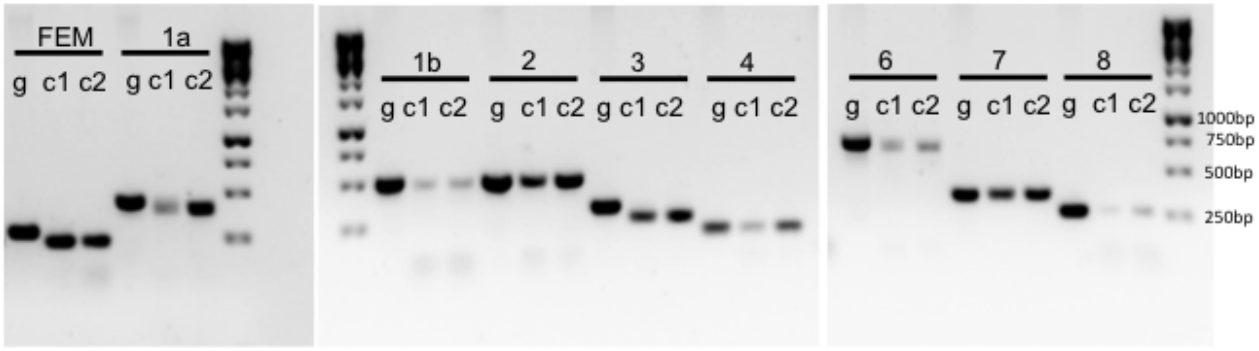

Fig. S3 PCR of *Fom* effector candidate genes on genomic and cDNA. Genomic DNA (g) was isolated from Fom005 grown on potato dextrose agar (PDA) plates. Revere transcript-PCR was performed on cDNA made from RNA that was isolated from 5-d-old NO_3-cultures (c1) or Fom005-infected melon Cha-T seedlings (c2). *FEM*, *Fusarium extracellular matrix* gene (Schoffelmeer *et al.*, 2001). Numbers indicate the effector candidates. Effector candidate 5 is absent in Fom005.

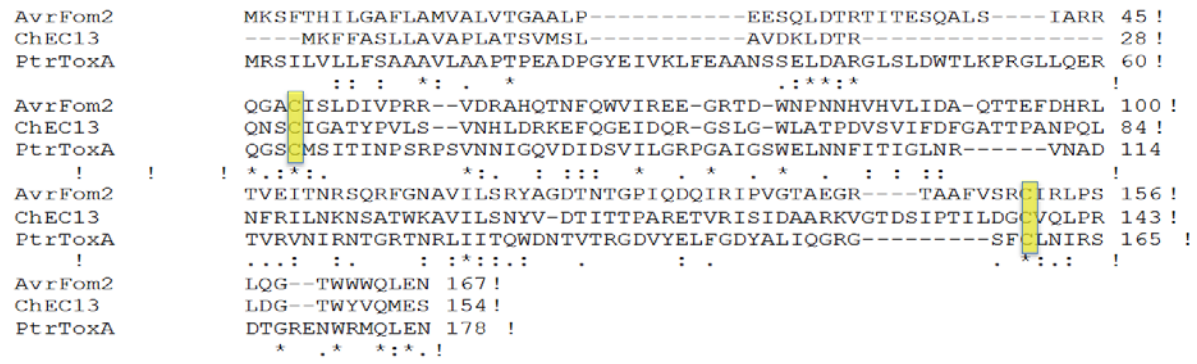

Fig. S4 AvrFom2 is weakly similar to ChEC13 and PtrToxA. Protein sequences of AvrFom2 (FOMG_19011), *Colletotrichum higginsianum* ChEC13 (CCF70887.1) and *Pyrenophora tritici-repentis* ToxA (ADY05382.1) were aligned with ClustalW2. Cysteine residues that form a cysteine knot in ToxA are marked with yellow boxes.

Fig. S5 Melon plants harbouring the *Fom-2* resistance gene are resistant to race 2 isolates that are ectopically transformed with *AVRFOM2*. Ten-day-old melon seedlings without (Cha-T) or with the *Fom-2* (Cha-*Fom2*) resistance gene were inoculated with conidia of Fom006, Fom009, Fom013 or the ectopic transformants Fom006T5, Fom009T5, Fom013T10. Pictures were taken 9 d after inoculation. Bars, 15 cm.

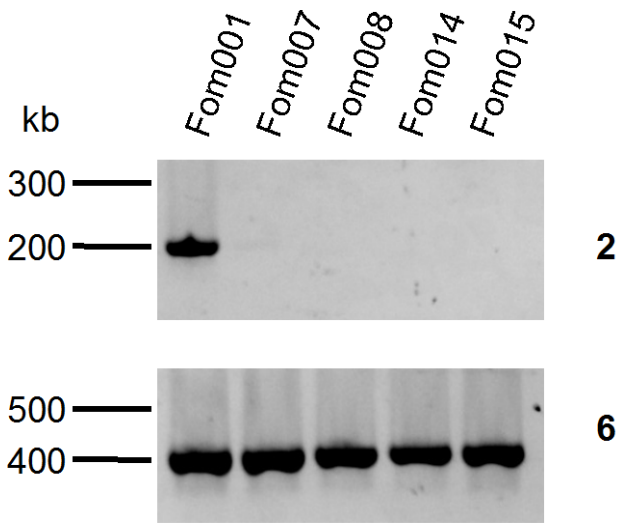

Fig. S6 *AVRFOM2* is absent in race 1,2 isolates. PCR of *AVRFOM2* and effector candidate *6* was performed on genomic DNA of Fom001 and four different race 1,2 isolates (Fom007, Fom008, Fom014, Fom015) grown for 5 d on PDA plates.

New Phytologist

Table S1 Confirmation of *Fom* races

Fom isolate	Melon cultivar	Green leaves, no wilting symptoms (%)	Severe wilting symptoms (%)	Dead (%)	Reported race	Our race determination
Fom001	Cha-T	0	0	100	-	**race1**
	Cha-Fom1	10	30	60		
	Cha-Fom2	100	0	0		
Fom004	Cha-T	10	0	90	race 0	**race 0**
	Cha-Fom1	100	0	0		
	Cha-Fom2	100	0	0		
Fom005	Cha-T	0	0	100	race 1	**race 1**
	Cha-Fom1	0	0	100		
	Cha-Fom2	100	0	0		
Fom006	Cha-T	0	0	100	race 2	**race 2**
	Cha-Fom1	100	0	0		
	Cha-Fom2	0	0	100		
Fom009	Cha-T	0	100	0	race 2	**race 2**
	Cha-Fom1	100	0	0		
	Cha-Fom2	0	90	10		
Fom010	Cha-T	70	30	0	race 1	**race 1**
	Cha-Fom1	0	100	0		
	Cha-Fom2	100	20	80		
Fom011	Cha-T	30	70	0	race 0	**race 0**
	Cha-Fom1	80	20	0		
	Cha-Fom2	100	0	0		
Fom012	Cha-T	0	50	50	race 0	**race 0**
	Cha-Fom1	80	20	0		
	Cha-Fom2	100	0	0		
Fom013	Cha-T	0	20	80	race 2	**race 2**
	Cha-Fom1	90	10	0		
	Cha-Fom2	0	30	70		
Fom016	Cha-T	0	0	100	race 1	**race 1**
	Cha-Fom1	0	0	100		
	Cha-Fom2	100	0	0		
MOCK	Cha-T	100	0	0		
	Cha-Fom1	100	0	0		
	Cha-Fom2	100	0	0		

Table S2 Coverage of the Fom001 reference genome by the Illumina paired-end sequencing reads of each *Fom* isolate

	Fom001	**Fom004**	**Fom005**	**Fom006**	**Fom009**	**Fom010**	**Fom011**	**Fom012**	**Fom013**	**Fom016**
x coverage[1]	106	139	86	92	142	90	91	87	90	92
Mapped reads[2]	83	74	77	79	70	75	77	81	81	79
Nonspecific matches[3]	1.28	1.46	1.44	1.4	1.93	1.91	1.53	1.28	1.17	1.3
Nonperfect matches[2]	33.77	64.03	65.33	65.6	67.83	66.59	68.84	64.96	66.02	66.04
Fraction of reference covered	1	0.86	0.86	0.85	0.84	0.82	0.84	0.85	0.83	0.85

The isolates were sequenced by Illumina using either 180 bp insert size libraries (Fom001), a combination of 170 bp and 500 bp insert size libraries (Fom006, Fom009, Fom010, Fom011, Fom012, Fom013, Fom016) or a combination of 170 bp, 500 bp and 5 kb insert size libraries (Fom004, Fom005). All reads were mapped to the annotated genome assembly of Fom001 . Nonspecific matches are an indication of repetitive sequences, nonperfect matches are an indication of sequence variation.

[1]Genome coverage of Fom001 genome (54 Mb) by sequencing reads. [2]Percentage of all reads.

[3]Percentage of all mapped reads.

Table S3 Reported single nucleotide polymorphisms (SNPs) for sequence read mapping to *F. oxysporum* f. sp. *melonis* Fom004 genome

Fom isolate	nt aligned w/ BWA[1]	Depth	GATK UG REF[2]	SNPs	Breadth of coverage (%)
Fom001	6179406543	112.68	**45,528,110**	**832,991**	84.54
Fom004	5621035770	102.50	**54,067,241**	**6,769**	98.60
Fom005	4595916870	83.80	**53,449,088**	**11,695**	97.48
Fom006	4918505400	89.69	**51,844,025**	**16,345**	96.42
Fom009	7396783110	134.88	**47,739,494**	**379,627**	89.46
Fom010	4525896690	82.53	**47,156,275**	**382,904**	88.38
Fom011	4569965640	83.33	**47,712,092**	**379,364**	89.41
Fom012	2890633230	52.71	**51,863,015**	**17,394**	96.46
Fom013	3107482740	56.66	**50,943,725**	**16,922**	94.75
Fom016	3092398650	56.39	**52,241,571**	**17,431**	97.16

[1]Burrows-Wheeler Aligner (BWA, version 0.7.4-r385 mem) (Li, 2013). [2]Genome Analysis Toolkit (GATK, version 2.7-4-g6f46d11) UnifiedGenotyper with reference (REF) base calling enabled and the haploid genotyper ploidy setting.

Table S4 Average read depth coverage of candidate effector genes

Super-contig	Start	End	Name	Mean base count per isolate									
				Fom 001	Fom 004	Fom 005	Fom 006	Fom 009	Fom 010	Fom 011	Fom 012	Fom 013	Fom 016
160	4001	4556	Fom_1A	43.86	83.17	61.42	77.63	0	59.86	0	69.07	77.86	83.28
450	1946	2494	Fom_1B	45.28	86.80	76.07	83.27	81.87	207.80	80.25	75.22	87.99	78.60
236	7624	8127	Fom_2	39.73	85.64	66.77	0.01	0	134.72	72.52	77.25	0	79.89
755	769	1160	Fom_3	29.48	77.04	63.83	77.7	83.46	126.13	83.52	70	71.38	165.46
900	1097	1291	Fom_4	84.45	131.31	124.51	157.97	195.05	365.76	213.78	141.13	144.04	151.02
692	502	1135	Fom_5	121.96	0	0	0	0	0	0	0	0	0
716	194	1044	Fom_6	80.52	225.96	170.82	218.73	204.51	241.34	210.46	204.81	202.56	210.95
449	2701	3147	Fom_7	39.91	78.3	130.41	75.34	79.22	138.70	76.34	0	77.00	88.28
584	671	1009	Fom_8	104.59	174.22	132.76	185.30	152.31	76	146.38	155.63	162.66	175.97
337	1016	1453	Fom_LysM	23.32	78.95	55.58	71.04	70.51	60.24	65.73	79.6	75.65	77.74
226	11358	12005	Fom_SIX6	38.13	95.51	71.62	76.7	0.07	0	0	76.01	84.83	85.37

Table S5 Primers used in this study

Primer no.	Orientation	Sequence (5'–3')	Target gene
157	Sense	ATGAAGTACACTCTCGCTACC	FEM1
158	Anti-sense	GGTGAAAGTGAAAGAGTCACC	FEM1
4520	Sense	GCACGCCATGACGTTGAGAA	Fom effector 1A
4521	Anti-sense	CTAGGAGCCTCTTTGATTGC	Fom effector 1A
4522	Sense	ATCCCATATGGCACTGGGCAA	Fom effector 1B
4523	Anti-sense	CTAGTTATGATTGGCGATAAC	Fom effector 1B
4524	Sense	ATGAAGAGCTTCACCCACATCC	Fom effector2
4525	Anti-sense	GCTGCCACCACCATGTGCCT	Fom effector2
4526	Sense	ATGAGGTTGCCACGGACAACTG	Fom effector3
4527	Anti-sense	GTACTTGCTTAGGTTGGGGTTC	Fom effector 3
4528	Sense	ATGCTCAATATGAAGATCTTTGC	Fom effector4
4529	Anti-sense	TTAACCACGGCAGCGGTCTTC	Fom effector4
4533	Sense	AATGCATCGCCTGCCGAAG	Fom effector 6
4534	Anti-sense	TAAGTCCGAATGTAAAGTCG	Fom effector 6
4535	Sense	ACTGGTCATGGCACAGTCTAA	Fom effector 7
4536	Anti-sense	TCATAGCGAATGTGCCAGAAG	Fom effector 7
4537	Sense	CCAGCTGTACTAACACATGCA	Fom effector8
4538	Anti-sense	TCAAATTGCCCGTAGCTGGAT	Fom effector 8

References

Li H. 2013. Aligning sequence reads, clone sequences and assembly contigs with BWA-MEM. *arXiv*: 1303.3997v2 [q-bio.GN].

Schoffelmeer EAM, Vossen JH, van Doorn AA, Cornelissen BJC, Haring MA. 2001. FEM1, a *Fusarium oxysporum* glycoprotein that is covalently linked to the cell wall matrix and is conserved in filamentous fungi. *Molecular Genetics and Genomics* **265:** 143–152.