

Technical Committee

TC/59/20

Fifty-Ninth Session  
Geneva, October 23 and 24, 2023Original: English  
Date: September 29, 2023**PARTIAL REVISION OF THE TEST GUIDELINES FOR MELON***Document prepared by an expert from France**Disclaimer: this document does not represent UPOV policies or guidance*

1. The purpose of this document is to present a proposal for a partial revision of the Test Guidelines for Melon (document TG/104/5 Rev. 2).

2. The Technical Working Party for Vegetables (TWV), at its fifty-seventh session<sup>1</sup>, considered a proposal for a partial revision of the Test Guidelines for Melon (*Cucumis melo* L.) on the basis of documents TG/104/5 Rev. 2 and TWV/57/22 "Partial revision of the Test Guidelines for Melon" and proposed the following changes (see document TWV/57/26 "Report", paragraph 70):

- (a) Revision of Characteristics 69.1 to 69.4 "Resistances to *Fusarium oxysporum* f. sp. *melonis* (Fom) - races 0, 1, 2, and 1.2";
- (b) Revision of explanation Ad. 69 "Resistances to *Fusarium oxysporum* f. sp. *melonis* (Fom) - races 0, 1, 2, and 1.2" in Chapter 8.2 "Explanations for individual characteristics";
- (c) Revision of Characteristics 70.1 to 70.5 "Resistances to *Podosphaera xanthii* (Px) - races 1, 2, 3, 5, 3.5";
- (d) Revision of explanation Ads. 70.1 to 70.3, 71 "Resistances to *Podosphaera xanthii* (Px), Resistance to *Golovinomyces cichoracearum* (*Erysiphe cichoracearum*), race 1 (Powdery mildew) Gc (Ec)" in Chapter 8.2 "Explanations for individual characteristics";
- (e) Inclusion of characteristics from the Table of Characteristics in the Technical Questionnaire

3. The proposed new wording is presented below. The proposed changes are presented in highlight and underline (insertion) and ~~strike through~~ (deletion) in the Annex to this document (in English only).

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<sup>1</sup> held in Antalya, Türkiye, from May 1 to 5, 2023, in hybrid format

Proposed revision of Characteristics 69.1 to 69.4 “Resistances to *Fusarium oxysporum* f. sp. *melonis* (Fom) - races 0, 1, 2, and 1.2”

	English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
<b>69. VG</b>	<b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom)</b>	<b>Résistance à <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom)</b>	<b>Resistenz gegen <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom)</b>	<b>Resistencia al <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom)</b>		
<b>69.1 (+)</b>	<b>Race 0 (Fom: 0)</b>	<b>Race 0 (Fom: 0)</b>	<b>Pathotyp 0 (Fom: 0)</b>	<b>Raza 0 (Fom: 0)</b>		
<b>QL</b>	absent	absente	fehlend	ausente	Atos, Charentais T	1
	present	présente	vorhanden	presente	Cadence, Charentais Fom-2, Dibango, Jubilo, Karakal, Védrantais	9
<b>69.2 (+)</b>	<b>Race 1 (Fom: 1)</b>	<b>Race 1 (Fom: 1)</b>	<b>Pathotyp 1 (Fom: 1)</b>	<b>Raza 1 (Fom: 1)</b>		
<b>QL</b>	absent	absente	fehlend	ausente	Atos, Charentais T, Védrantais	1
	present	présente	vorhanden	presente	Cadence, Charentais Fom-2, Dibango, Jubilo, Karakal	9
<b>69.3 (+)</b>	<b>Race 2 (Fom: 2)</b>	<b>Race 2 (Fom: 2)</b>	<b>Pathotyp 2 (Fom: 2)</b>	<b>Raza 2 (Fom: 2)</b>		
<b>QL</b>	absent	absente	fehlend	ausente	Atos, Charentais Fom-2, Charentais T, Dibango, Marianna	1
	present	présente	vorhanden	presente	Cadence, Charentais Fom-1, Jubilo, Karakal, Perlita, Védrantais	9
<b>69.4 (+)</b>	<b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Race 1.2 (Fom: 1.2)</b>	<b>Résistance à <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Race 1.2 (Fom: 1.2)</b>	<b>Resistenz gegen <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Pathotyp 1.2 (Fom: 1.2)</b>	<b>Resistencia al <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Raza 1.2 (Fom: 1.2)</b>		
<b>QL</b>	absent	absente	fehlend	ausente	Graffio, Prity, Virgos	1
	present	présente	vorhanden	presente	Isabelle, Kyriel, Lunasol, Meliance, Piboule	9

Proposed revision of explanation Ad. 69 “Resistances to *Fusarium oxysporum* f. sp. *melonis* (Fom) - races 0, 1, 2, and 1.2” in Chapter 8.2 “Explanations for individual characteristics”

Ads. 69: 69.1 - 69.3: Resistance to *Fusarium oxysporum* f. sp. *melonis*, races 0, 1 and 2 (Fom: 0, Fom: 1, Fom: 2)

1.	Pathogen	<i>Fusarium oxysporum</i> f. sp. <i>melonis</i> races 0, 1, and 2																														
2.	Quarantine status	No																														
3.	Host species	Melon - <i>Cucumis melo</i>																														
4.	Source of inoculum	e.g. GEVES (FR) <sup>2</sup>																														
5.	Isolate	e.g., Reference strain validated in an inter-laboratory test <sup>3, 4</sup> Fom:0 - Strain MLZ = MAT/REF/04-07-01-03-02 <sup>2</sup>  Fom: 1 - Strain FOM 26 = MAT/REF/04-07-01-01 <sup>2</sup>  Fom: 2 - Strain F185																														
6.	Establishment isolate identity	The most recent table is available through ISF at <a href="https://www.worldseed.org/our-work/plant-health/differential-hosts/">https://www.worldseed.org/our-work/plant-health/differential-hosts/</a> <i>Situation July 2019</i>																														
<table border="1"> <thead> <tr> <th>Differential host</th> <th>Gene present</th> <th>Fom: 0*</th> <th>Fom: 1*</th> <th>Fom: 2*</th> <th>Fom: 1.2*</th> </tr> </thead> <tbody> <tr> <td>Charantais T*</td> <td>-</td> <td>S</td> <td>S</td> <td>S</td> <td>S</td> </tr> <tr> <td>Védraçais*, Doublon*</td> <td><i>Fom-1</i></td> <td>HR</td> <td>S</td> <td>HR</td> <td>S</td> </tr> <tr> <td>Charantais Fom-2*, CM17187*</td> <td><i>Fom-2</i></td> <td>HR</td> <td>HR</td> <td>S</td> <td>S</td> </tr> <tr> <td>Isabelle*</td> <td><i>Polygenic?</i></td> <td>HR</td> <td>HR</td> <td>HR</td> <td>IR</td> </tr> </tbody> </table> <p>S = susceptible; HR = highly resistant; IR = intermediate *differential hosts and isolates that are used by the seed sector</p> <p style="text-align: right;">Courtesy of Worldseed.org website</p>			Differential host	Gene present	Fom: 0*	Fom: 1*	Fom: 2*	Fom: 1.2*	Charantais T*	-	S	S	S	S	Védraçais*, Doublon*	<i>Fom-1</i>	HR	S	HR	S	Charantais Fom-2*, CM17187*	<i>Fom-2</i>	HR	HR	S	S	Isabelle*	<i>Polygenic?</i>	HR	HR	HR	IR
Differential host	Gene present	Fom: 0*	Fom: 1*	Fom: 2*	Fom: 1.2*																											
Charantais T*	-	S	S	S	S																											
Védraçais*, Doublon*	<i>Fom-1</i>	HR	S	HR	S																											
Charantais Fom-2*, CM17187*	<i>Fom-2</i>	HR	HR	S	S																											
Isabelle*	<i>Polygenic?</i>	HR	HR	HR	IR																											
7.	Establishment pathogenicity	use susceptible melon varieties																														
8.	Multiplication inoculum																															
8.1	Multiplication medium	on agar medium – e.g., Potato Dextrose Agar, Malt agar at 20°C to 25°C																														
8.2	Multiplication variety	-																														
8.3	Plant stage at inoculation	-																														
8.5	Inoculation method	-																														
8.6	Harvest of inoculum	7–10-day-old culture																														
8.7	Check of harvested inoculum	-																														
8.8	Shelf life /viability inoculum	Between 4 to 8 h or keep cool to prevent spore germination																														
9.	Format of the test																															
9.1	Number of plants per genotype	at least 30 plants, it is important to have at least 5 non-inoculated plants per genotype to be able to judge growth reduction																														
9.2	Number of replicates	At least e.g. 3 replicates (3 x10)																														


<sup>2</sup> [matref@geves.fr](mailto:matref@geves.fr)



<sup>3</sup> Harmores 3 CPVO project ([https://cpvo.europa.eu/sites/default/files/documents/report\\_harmores\\_3\\_final\\_meeting\\_v0\\_0.pdf](https://cpvo.europa.eu/sites/default/files/documents/report_harmores_3_final_meeting_v0_0.pdf))

<sup>4</sup> ISF EG DRT Fom: 2 resistance in Melon – [Link to include](#) – *not yet available*

9.3	Control varieties	
9.3.1	Control varieties for race 0	Resistance absent: Charentais T Resistance present: Charentais Fom-2, Védrantais
9.3.2	Control varieties for race 1	Resistance absent: Charentais T, Védrantais Resistance present: Charentais Fom-2
9.3.3	Control varieties race 2	Resistance absent: Marianna Resistance present: Perlita, Charentais Fom-1, Védrantais
9.4	Test design	3 replicates of 10 plants to allow statistical analysis (in different trays) and at least 5 non-inoculated plants per genotype.
9.5	Test facility	glasshouse or climatic room
9.6	Temperature	- Fom: 0 and Fom: 1: 18 - 24°C - Fom: 2: 24°C
9.7	Light	- Fom: 0 and Fom: 1: At least 12h - Fom: 2: 16h
9.9	Special measures	- Fom: 0 and Fom: 1: Recommend having really 18°C at night and not above 24°C during the day.
10.	Inoculation	
10.1	Preparation inoculum	Scrape spore cultures with water from agar medium (see 8.1) or optional multiplication on liquid medium (e.g., Messiaen (1991) synthetic liquid medium, sucrose 50g/L, on permanent agitator-shaker or aerated Czapek-Dox culture medium for 5-7 days at room temperature). <i>Remark:</i> Beware of toxin productions by some isolates (see remark under 13.)
10.2	Quantification inoculum	$4 \times 10^5$ to $1 \times 10^6$ sp /mL
10.3	Plant stage at inoculation	cotyledon expanded
10.4	Inoculation method	Plant at the inoculation stage are harvested carefully, roots and hypocotyls are immersed in spore suspension for 2-15 min; trimming of roots is an option; transplant in trays.
10.5	First observation	1 <sup>st</sup> notation: symptoms on Resistance absent (susceptible) control at classes 2 and 3 with a strong proportion at class 3
10.6	Second observation	A second notation can be necessary to re-evaluate some unclear varieties
11.	Observations	
11.1	Method	Visual observation

11.2 Observation scale

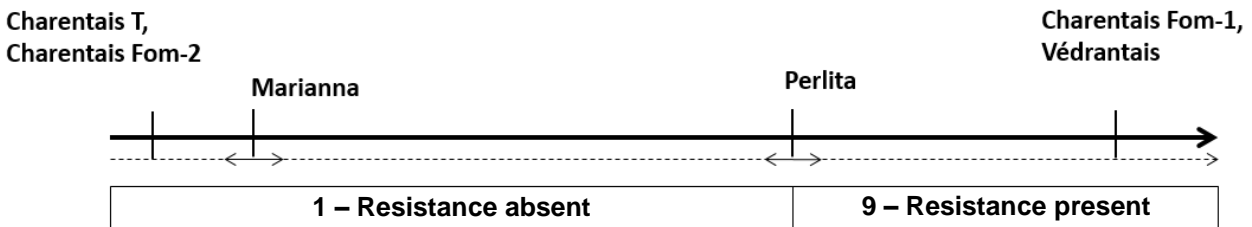
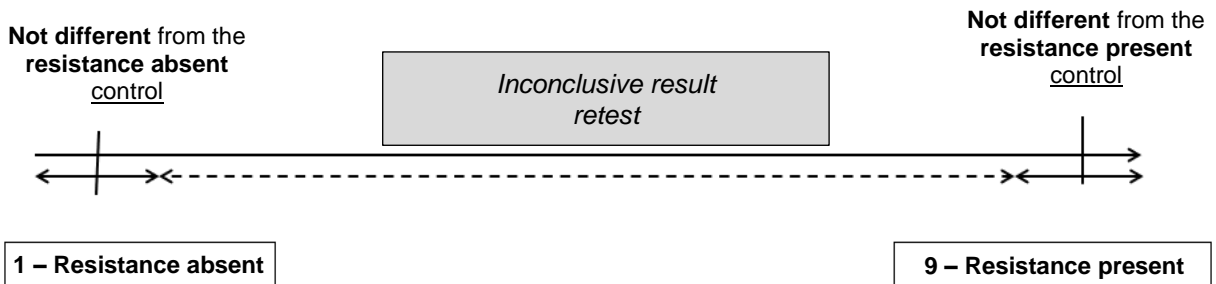
non-inoculated plant	Class 0	Class 1
At least 5 plants	Healthy plant: no symptoms of yellowing and wilting, could be some growth reduction due to inoculation stress compared to mock. Sometimes in the mock we can observe some yellowing, different from the symptoms of <i>Fusarium</i>	Light symptoms of yellowing/wilting
		

Class 2	Class 3	
typical symptoms: yellowing, wilting and necrosis, stunting (growth stopped)	Death of plant (Dead)	
		 <p>Other symptoms of vein clearing could be difficult to judge. It is advised to make a later notation to observe the evolution of these symptoms over the time.</p>

Courtesy of GEVES-SNES in the framework of CPVO Harmores project.

11.3	Validation of test	<p>Validation on controls.</p> <p>In case of the Fom: 0 and Fom:1 tests: Controls expected response: Resistance absent: most of the plants at classes 2 and 3 Resistance present: most of the plants at classes 0 and 1, sometimes very few plants at classes 2 or 3.</p> <p>In case of the Fom: 2 test Controls expected response:</p> <ul style="list-style-type: none"> <li>• Susceptible controls, with UPOV characteristic state 'Resistance absent', should have most of the plants in observation classes 2 or 3, and few or no plants in observation classes 0 or 1. <ul style="list-style-type: none"> <li>◦ Marianna, the susceptible control is less susceptible than Charentais Fom-2, Charentais T</li> </ul> </li> <li>• Resistant controls should have most of the plants in observation classes 0 or 1, and few or no plant in observation classes 2 or 3.</li> </ul> <p>Perlita, the lower threshold resistance control, should have at least some plants in observation class 1, 2, or 3. It has to be less resistant than Charentais Fom-1, Védrantais.</p>
11.4	Off-types	-
12.	Interpretation of data in terms of UPOV characteristic states	<p>In case of varieties with a response between the susceptible (resistance absent) and the resistant control, repeat the test- In case of confirmation of the result, the variety will be judged heterogeneous. In case of unclear results, retest or test in another lab.</p>

Resistance to Fom: 0 and Fom: 1



13.	Critical control points	<p>For race 2, the control Perlita, with the <i>Fom-3</i> gene, allows to validate the capacity of the isolate to partially attack this variety.</p> <p>In the case of inoculum increased in e.g. Messiaen (1991) synthetic liquid medium, on permanent agitator-shaker, inoculum can be used after 5 to 7 days.</p> <p>For race 0 and 1, dilution 1/12 is recommended, while it must not be less than 1/20 for race 2. At a lower dilution (higher concentration of the medium), it has been observed that toxins released in the medium by the race 2 can cause some yellowing of melon plants, even if they are resistant. Alternatively, spores can be “washed” by resuspending a mass of spores collected on a Millipore filter with vacuum force.</p>
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Ad 69.4: Resistance to *Fusarium oxysporum* f. sp. *melonis* race 1.2 (Fom: 1.2)




1.	Pathogen	<i>Fusarium oxysporum</i> f. sp. <i>melonis</i> race 1.2 (Fom: 1.2)																														
2.	Quarantine status	No																														
3.	Host species	Melon - <i>Cucumis melo</i> L.																														
4.	Source of inoculum	GEVES (FR) <sup>5</sup>																														
5.	Isolate	e.g., Reference strain validated in an inter-laboratory test <sup>6</sup> Fom: 1.2 - Strain TST = MAT/REF/04-07-01-04 <sup>2</sup>																														
6.	Establishment isolate identity	The most recent table is available through ISF at <a href="https://www.worldseed.org/our-work/plant-health/differential-hosts/Situation%20July%202019">https://www.worldseed.org/our-work/plant-health/differential-hosts/Situation July 2019</a>																														
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Differential host	Gene present	Fom: 0*	Fom: 1*	Fom: 2*	Fom: 1.2*																											
Charantais T*	-	S	S	S	S																											
Védrantais*, Doublon*	<i>Fom-1</i>	HR	S	HR	S																											
Charantais Fom-2*, CM17187*	<i>Fom-2</i>	HR	HR	S	S																											
Isabelle*	<i>Polygenic?</i>	HR	HR	HR	IR																											
7.	Establishment pathogenicity	use susceptible melon varieties																														
8.	Multiplication inoculum																															
8.1	Multiplication medium	on agar medium e.g., Potato Dextrose Agar, Sabouraud, at 20°C to 25°C																														
8.2	Multiplication variety	-																														
8.3	Plant stage at inoculation	-																														
8.5	Inoculation method	-																														
8.6	Harvest of inoculum	4-10 day-old culture																														
8.7	Check of harvested inoculum	-																														
8.8	Shelf life/viability inoculum	-																														
9.	Format of the test																															
9.1	Number of plants per genotype	30 plants per variety plus 5 non-inoculated controls																														
9.2	Number of replicates	At least 3 x 10 plants, in different trays																														
9.3	Control varieties	Resistance absent: Virgos Resistance present: Piboule and Lunasol and Isabelle (Isabelle is expected to have a lower disease index (DI) (= higher resistance than Piboule and Lunasol). Piboule and Lunasol are both needed to illustrate the lower level of resistance. Their resistance is based on other genetics and may have different levels in different labs.																														
9.4	Test design	3 replicates of 10 plants to allow statistical analysis (in different trays) and at least 5 non-inoculated plants per genotype.																														
9.5	Test facility	glasshouse or climatic room																														
9.6	Temperature	18-24°C																														
9.7	Light	at least 12h																														

<sup>5</sup> [matref@geves.fr](mailto:matref@geves.fr)<sup>6</sup> Harmores 3 CPVO project ([https://cpvo.europa.eu/sites/default/files/documents/report\\_harmores\\_3\\_final\\_meeting\\_v0\\_0.pdf](https://cpvo.europa.eu/sites/default/files/documents/report_harmores_3_final_meeting_v0_0.pdf))



10.	Inoculation	
10.1	Preparation inoculum	Scrape cultures with water on agar medium (see 8.1) or optional multiplication on liquid medium (e.g., Potato Dextrose Broth (PDB), Czapek-Dox culture medium for 7 days at room temperature and darkness or Messiaen (1991) synthetic liquid medium, sucrose 50 g/L, on permanent agitator-shaker, at room-temperature, inoculum can be used after 5 to 7 days)
10.2	Quantification inoculum	$1 \times 10^5$ - $1 \times 10^6$ sp/mL, depending on inoculation method (see 10.4) and lab conditions
10.3	Plant stage at inoculation	cotyledons expanded, first leaf emerging
10.4	Inoculation method	One of two methods can be used for inoculation. - Absorption: Absorption of a suspension of spores, e.g., 700mL of a suspension at $1.10^5$ sp/mL for 50 plants in a tray 30 cm*30 cm. - Injection: Injection of a suspension of spores into the soil at the base of the plant, e.g., 5mL at $10^6$ sp /mL per plant.
10.7	Final observations	1 <sup>st</sup> notation: symptoms on susceptible control at least at class 3 [generally 10-21 dpi]. A 2 <sup>nd</sup> notation can be necessary to reevaluate some unclear varieties.
11.	Observations	
11.1	Method	Visual observation
11.2	Observation scale	

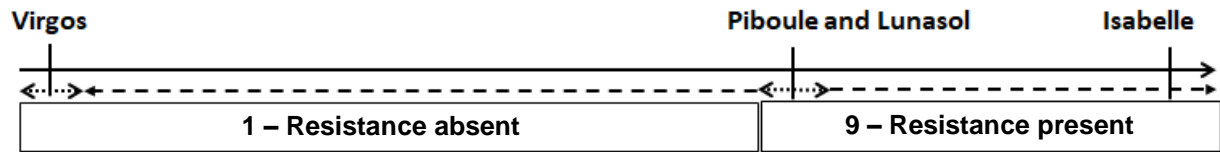
Non-inoculated plants	Class 0	Class 1
Varieties must be compared to the non-inoculated plants.	Healthy plant, the whole plant is green or at the same level than the mock. Just a light yellowing can be accepted on the mock	Light level of symptoms, light yellowing on cotyledons and/or leaves without necrosis
		

Class 2	Class 3	Class 4
Moderate level of symptoms, yellowing on cotyledon and/or leaves, starting of necrosis and wilting but not extended	Severe symptoms of yellowing and/or wilting on cotyledons and/or leaves with extended necrosis	Dead plant, no green leaf part or hypocotyl is dry
		

Courtesy of GEVES-SNES in the framework of CPVO Harmores project.

11.3	Validation of test	<p>Validation on controls. Controls expected response:</p> <ul style="list-style-type: none"> <li>- Resistance present: Most plants in classes 0 and 1, in some cases with few plants in 2, 3, 4. Low level of disease index (DI) generally below 40%. A difference of disease index is generally observed between Piboule and Lunasol compared to Isabelle</li> <li>- Resistance absent: Most plants in classes 3 and 4, in some cases with few plants at class 0, 1, or 2. Very high disease index (DI) above 80%.</li> </ul>
11.4	Off-types	-
12.	Interpretation of data in terms of UPOV characteristic states	<p>Interpretation of varieties depending on controls (figure 1) Note 1 = Resistance absent Note 9 = Resistance present</p> <p>Quantitative analysis is based on the disease index (DI) AND the distribution of plants per class compared to the controls</p> <p>The varieties statistically similar to the resistant controls or with a lower disease index (DI) have to be judged as resistant.</p> <p>The varieties between the susceptible and the resistant controls have to be judged as susceptible.</p> <p>If not clear, the use of statistics is highly recommended.</p>

Resistance to Fom:1-2:



$$DI = \frac{(N0 * 0) + (N1 * 1) + (N2 * 2) + (N3 * 3) + (N4 * 4)}{(N0 + N1 + N2 + N3 + N4) * 4} * 100$$

Nx : number of plants at class x

Figure 1: disease index (DI) formula

Proposed revision of Characteristics 70.1 to 70.5 “Resistances to *Podosphaera xanthii* (Px) - races 1, 2, 3, 5, 3.5”

70.	VG	Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i> ) (Powdery mildew)	Résistance à <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i> ) (oïdium)	Resistenz gegen <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i> ) (Echter Mehltau)	Resistencia a <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i> ) (Oidio)		
70.1		<b>Race 1 (Px: 1)</b>	<b>Race 1 (Px: 1)</b>	<b>Pathotyp 1 (Px: 1)</b>	<b>Raza 1 (Px: 1)</b>		
	(+)						
QN		absent or low	absente ou faible	fehlend oder gering	ausente o baja	Védrantais	1
		medium	moyenne	mittel	media	Escrito	2
		high	élevée	hoch	alta	Arum	3
70.2		<b>Race 2 (Px: 2)</b>	<b>Race 2 (Px: 2)</b>	<b>Pathotyp 2 (Px: 2)</b>	<b>Raza 2 (Px: 2)</b>		
	(+)						
QN		absent or low	absente ou faible	fehlend oder gering	ausente o baja	Védrantais	1
		medium	moyenne	mittel	media	Escrito, Pendragon	2
		high	élevée	hoch	alta	Arum	3
70.3		<b>Race 3 (Px: 3)</b>	<b>Race 3 (Px: 3)</b>	<b>Pathotyp 3 (Px: 3)</b>	<b>Raza 3 (Px: 3)</b>		
	(+)						
QN		absent or low	absente ou faible	fehlend oder gering	ausente o baja	Védrantais	1
		medium	moyenne	mittel	media	Arago, Durango	2
		high	élevée	hoch	alta	Arum	3
70.4		<b>Race 5 (Px: 5)</b>	<b>Race 5 (Px: 5)</b>	<b>Pathotyp 5 (Px: 5)</b>	<b>Raza 5 (Px: 5)</b>		
	(+)						
QN		absent or low	absente ou faible	fehlend oder gering	ausente o baja	Védrantais	1
		medium	moyenne	mittel	media	Arago, Durango	2
		high	élevée	hoch	alta	Arum	3
70.5		<b>Race 3-5 (Px: 3.5)</b>	<b>Race 3-5 (Px: 3.5)</b>	<b>Pathotyp 3-5 (Px: 3.5)</b>	<b>Raza 3-5 (Px: 3.5)</b>		
	(+)						
QN		absent or low	absente ou faible	fehlend oder gering	ausente o baja	Védrantais	1
		medium	moyenne	mittel	media	Arago, Durango	2
		high	élevée	hoch	alta	Arum	3

Revision of explanation Ads. 70.1 to 70.3, 71 “Resistances to *Podosphaera xanthii* (Px), Resistance to *Golovinomyces cichoracearum* (Gc) (Powdery mildew)” in Chapter 8.2 “Explanations for individual characteristics”

Ads. 70.1 to 70.5: Resistance to *Podosphaera xanthii* (Px) (ex *Sphaerotheca fuliginea*) (Powdery mildew) races 1, 2, 3, 5, 3.5 (Px: 1, 2, 3, 5, 3.5)

Ad. 71: Resistance to *Golovinomyces cichoracearum* (Gc) (*Erysiphe cichoracearum*) (Powdery mildew), race 1 (Gc: 1)

1.	Pathogen	Powdery mildew: <i>Podosphaera xanthii</i> (ex <i>Sphaerotheca fuliginea</i> ) races 1, 2, 3, 5 and 3.5 <i>Golovinomyces cichoracearum</i> (ex <i>Erysiphe cichoracearum</i> ) race 1
2.	Quarantine status	No
3.	Host species	Melon - <i>Cucumis melo</i> L.
4.	Source of inoculum	GEVES (FR) <sup>7</sup>
5.	Isolate	e.g., Reference strain validated in an inter-laboratory test <sup>8</sup> Px: 1 - Strain Sm 3 = MAT/REF/04-07-03-01 <sup>7</sup> Px: 2 - Strain S87-7 = MAT/REF/04-07-03-02 <sup>7</sup> Px: 3 - Strain 00Sm39 = MAT/REF/04-07-03-04-02 <sup>7</sup> Px: 5 - Strain 98Sm65 = MAT/REF/04-07-03-03-01-02 <sup>7</sup> Px: 3.5 - Strain 04Sm2 = MAT/REF/04-07-03-05-01 <sup>7</sup>  Gc: 1 - Strain GEVES = MAT/REF/04-07-02-01) <sup>3</sup>
6.	Establishment isolate identity	on differentials (table 1)

Table 1:

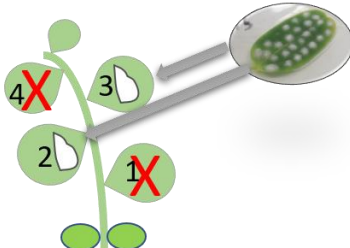
Races of *Podosphaera xanthii* (Px) and *Golovinomyces cichoracearum* (Gc), J. McCreight and M. Pitrat



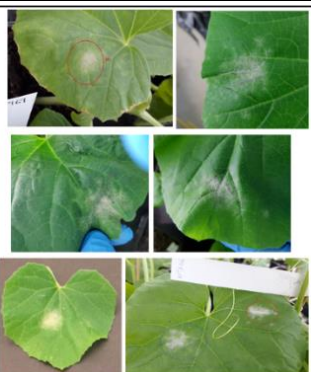


	Race 0	<i>Podosphaera xanthii</i>						<i>Golovinomyces cichoracearum</i>	
		Race 1	Race 2	Race 3	Race 4	Race 5	Race 3.5	Race 0	Race 1
Iran H	S	S	S	S	S	S	S	S	S
Védrantais	R	S	S	S	S	S	S	R	S
PMR45	R	R	S	S	S	S	S	R	S
WMR29	R	R	R	R	S	S	S	R	S
Edisto 47	R	R	R	R	R	S	S	R	S
MR-1, PI124112	R	R	R	R	R	R	R	R	R
PMR5	R	R	R	S	S	R	S	R	R
Nantais Oblong	R	S	S	S	S	S	S	R	R

<sup>7</sup> [matref@geves.fr](mailto:matref@geves.fr)

<sup>8</sup> Harmores 3 CPVO project ([https://cpvo.europa.eu/sites/default/files/documents/report\\_harmores\\_3\\_final\\_meeting\\_v0\\_0.pdf](https://cpvo.europa.eu/sites/default/files/documents/report_harmores_3_final_meeting_v0_0.pdf))

7.	Establishment pathogenicity	use susceptible melon varieties								
8.	Multiplication inoculum									
8.1	Multiplication medium	Melon plantlets								
8.2	Multiplication variety	Susceptible variety, for example Védraçais. For higher isolates like 3.5 or 5, a variety with broken resistance is recommended to keep the isolate pure.								
8.3	Plant stage at inoculation	Cotyledon								
8.5	Inoculation method	Sowing in substrate, for example soil or disinfected peat inside a closed mini glasshouse. When the cotyledons have expanded, remove them from the plant. Disinfect the cotyledons by soaking them for 3 minutes in a mercuric chloride solution (0.05%) or in sodium hypochlorite solution. Rinse them with sterilized water. Dry the cotyledons with sterile paper towel, then place them in Petri dishes with the following medium: <table style="margin-left: 40px; border: none;"> <tr> <td>Sucrose</td> <td>10g</td> </tr> <tr> <td>Mannitol</td> <td>20g</td> </tr> <tr> <td>Agar</td> <td>5g</td> </tr> <tr> <td>Distilled water</td> <td>1 liter</td> </tr> </table> Scatter conidia on the cotyledons and blow them or deposit conidia at the surface of cotyledons. Incubate the inoculated cotyledons in Petri dishes for example at 23°C during 14 hours in the light and at 18°C during 10 hours in the dark or 17°C permanently under very low light intensity. 9 to 11 days after the inoculation, the cotyledons will be covered with conidia and can be used as an inoculum.	Sucrose	10g	Mannitol	20g	Agar	5g	Distilled water	1 liter
Sucrose	10g									
Mannitol	20g									
Agar	5g									
Distilled water	1 liter									
8.6	Harvest of inoculum	Sporulation on cotyledons								
8.8	Shelf life /viability inoculum	Maximum 1 to 1.5 months after the inoculation.								
9.	Format of the test									
9.1	Number of plants per genotype	At least 20 plants per variety and controls, 5 plants for other differentials.								
9.2	Number of replicates	-								
9.3	Control varieties	For <i>Podosphaera xanthii</i> (Px) race 1, resistance <ul style="list-style-type: none"> <li>• absent or low: Védraçais</li> <li>• medium: Escrito</li> <li>• high: Arum</li> </ul> For <i>Podosphaera xanthii</i> (Px) race 2, resistance: <ul style="list-style-type: none"> <li>• absent or low: Védraçais</li> <li>• medium: Escrito, Pendragon</li> <li>• high: Arum</li> </ul> For <i>Podosphaera xanthii</i> (Px) races 3, 5, 3.5, resistance: <ul style="list-style-type: none"> <li>• absent or low: Védraçais</li> <li>• medium: Arago, Durango</li> <li>• high: Arum</li> </ul> For <i>Golovinomyces cichoracearum</i> (Gc) race 1, resistance: <ul style="list-style-type: none"> <li>• absent or low: Védraçais</li> <li>• medium: Anasta</li> <li>• high: Cézanne</li> </ul>								
9.4	Test design	Include differentials to validate the race (at least 5 plants per differentials) and compare the level of sporulation.								
9.5	Test facility	Climatic chamber or greenhouse								
9.6	Temperature	20-24°C								
9.7	Light	At least 12 hours								

10.	Inoculation	
10.1	Preparation inoculum	-
10.2	Quantification inoculum	-
10.3	Plant stage at inoculation	Whole plants at 3-4 true leaf fully expanded stage. Inoculation on the leaves 2 and 3 indicated on the diagram below. 
10.4	Inoculation method	Take spores from a cotyledon already covered with conidia and deposit them on a leaf. Different isolates can be tested on the same plant (or the same leaf) if the local deposit is well separated from each other and if a mark indicates the place of the deposit. <small>Courtesy of GEVES-SNES in the framework of CPVO Harmores project.</small>
10.7	Final observations	The date of notation should be chosen based on expected symptoms on the three controls. Sporulation should be well expressed on the susceptible control.
11.	Observations	
11.1	Method	Visual observation of sporulation
11.2	Observation scale	

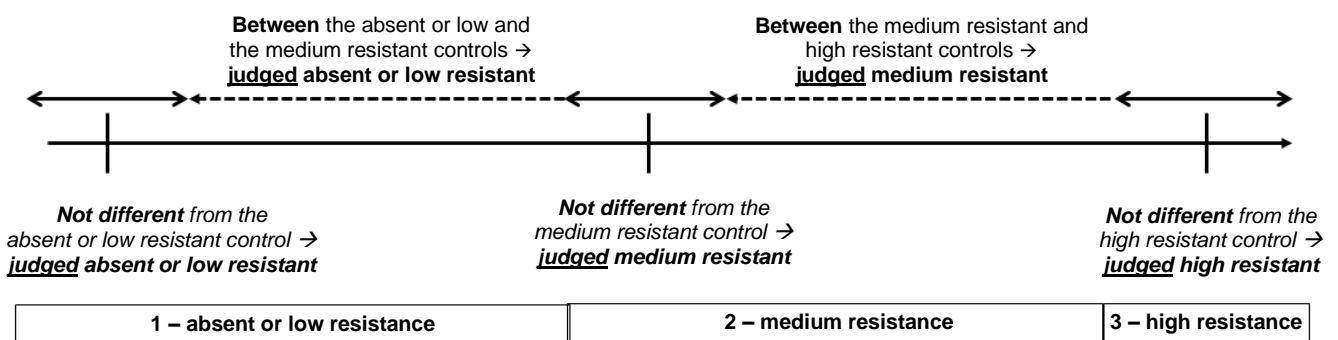
Class 1: No development of the fungus (no mycelium or dead mycelium) or no sporulation	Class 3: weak sporulation	Class 5: moderate sporulation	Class 9: strong sporulation
			
	Example of contamination by environment on the susceptible control, test not validated		

Courtesy of GEVES-SNES in the framework of CPVO Harmores project.

11.3	Validation of test	<p>Validation on controls.</p> <p>Additional information for expected responses of <i>Podosphaera xanthii</i> controls</p> <p>Resistance absent or low</p> <ul style="list-style-type: none"> <li>Plants at class 9, or most of the plants at class 9 and few plants at class 5 (high disease index).</li> <li>Few plants at class 3 but in this case the resistant controls should be all at class 1 and the intermediate resistant control at classes 3 and 1.</li> <li>No plants at class 1.</li> </ul> <p>Resistance medium</p> <ul style="list-style-type: none"> <li>Between the resistant and the susceptible control.</li> <li>Generally, plants at classes 3 and 5.</li> </ul> <p>Resistance high</p> <ul style="list-style-type: none"> <li>Plants at class 1, or most of the plants at class 1 and few plants at class 3 (very low disease index).</li> <li>Plants at class 3 but in this case the susceptible control should be all at class 9.</li> <li>No plants at classes 5 or 9.</li> </ul>
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11.4	Off-types	-
12.	Interpretation of data in terms of UPOV characteristic states	<p>Interpretation of varieties depending on controls (figure 1)</p> <p>Resistance Note 1 = absent or low Note 2 = medium Note 3 = high</p> <p>Quantitative analysis is based on the disease index AND the distribution of plants per class compared to the controls.</p> <p>Additional information for <i>Podosphaera xanthii</i> controls: The varieties between the intermediate resistant and the resistant control have to be judged as intermediate resistant (because they are not resistant enough to be considered resistant). The varieties between the susceptible and the intermediate resistant control have to be judged as susceptible (because they are not resistant enough to be considered intermediate resistant).</p>

Resistance to Px:



$$DI = \frac{(N1*0)+(N3*1)+(N5*2)+(N9*3)}{(N1+N3+N5+N9)*3} * 100$$

NX: Number of plants at class X

Figure 1: disease index formula

13.	Critical control points	To avoid cross contamination, it is advised to not produce inoculum of different races in the same room.
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Inclusion of characteristics from the Table of Characteristics in the Technical Questionnaire

4. The TWV is invited to consider the inclusion of the following characteristics in the TQ (characteristics for inclusion indicated in highlight and underline):

Char. No.	(*)	Characteristic Name
12	(*)	Inflorescence: sex expression (at full flowering)
13		<u>Young fruit: hue of green color of skin</u>
14	(*)	<u>Young fruit: intensity of green color of skin</u>
24	(*)	<u>Fruit: length</u>
25	(*)	<u>Fruit: diameter</u>
28	(*)	Fruit: shape in longitudinal section
29	(*)	Fruit: ground color of skin
31		<u>Fruit: hue of ground color of skin</u>
32		<u>Fruit: density of dots</u>
36	(*)	Fruit: density of patches
38	(*)	Fruit: warts
43	(*)	Fruit: grooves
45		<u>Fruit: depth of grooves</u>
47	(*)	<u>Fruit: creasing of surface</u>
48	(*)	Fruit: cork formation
49	(*)	<u>Fruit: thickness of cork layer</u>
50	(*)	Fruit: pattern of cork formation
51	(*)	Fruit: density of pattern of cork formation
54	(*)	Fruit: main color of flesh
60	(*)	Seed: length
62		<u>Seed: shape</u>
63	(*)	Seed: color
68	(*)	Shelf life of fruit
69.1	(*)	Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Race 0 ( <u>Fom: 0</u> )
69.2	(*)	Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Race 1 ( <u>Fom: 1</u> )
69.3	(*)	Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Race 2 ( <u>Fom: 2</u> )
69.4		<u>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Race 1.2 (Fom: 1.2)</u>
70.1		<u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) (Px: 1)</u>
70.2		<u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) Race 2 (Px: 2)</u>
70.3		<u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) Race 3 (Px: 3)</u>
70.4		<u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) Race 5 (Px: 5)</u>
70.5		<u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) Race 3-5 (Px: 3,5)</u>
71		<u>Resistance to <i>Golovinomyces cichoracearum</i> (<i>Erysiphe cichoracearum</i>) (Powdery mildew) Race 1 (Gc: 1)</u>
72		<u>Resistance to colonization by <i>Aphis gossypii</i></u>
73		<u>Resistance to <i>Zucchini yellow mosaic virus</i> (ZYMV)</u>
74.1		<u>Resistance to <i>Papaya ringspot virus</i> (PRSV) Guadeloupe strain</u>
74.2		<u>Resistance to <i>Papaya ringspot virus</i> (PRSV) E2 strain</u>
75		<u>Resistance to <i>Melon necrotic spot virus</i> (MNSV) Strain 0 (MNSV: 0)</u>
76		<u>Resistance to <i>Cucumber mosaic virus</i> (CMV)</u>

5. The detailed changes to the TQ are presented in highlight and underline (insertion) and ~~strike through~~ (deletion) in the Annex to this document (in English only).

PROPOSED CHANGES PRESENTED IN HIGHLIGHT  
(in English only)

Proposed revision of Characteristics 69.1 to 69.4 “Resistances to *Fusarium oxysporum* f. sp. *melonis* (Fom) - races 0, 1, 2, and 1.2”

	English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
<b>69. A</b>	<b>VG</b>	<b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom)</b>	<b>Résistance à <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom)</b>	<b>Resistenz gegen <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom)</b>	<b>Resistencia al <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom)</b>	
		-----	-----	-----	-----	-----
<b>69.1</b>		<b>Race 0 (Fom: 0)</b>	<b>Race 0 (Fom: 0)</b>	<b>Pathotyp 0 (Fom: 0)</b>	<b>Raza 0 (Fom: 0)</b>	
		(+) (+)				
<b>QL</b>	absent	absente	fehlend	ausente	Jaune Canari 2 Atos, Charentais T	1
	present	présente	vorhanden	presente	Cadence, Charentais Fom-2, Dibango, Jador, Jubilo, Karakal, Védrantais	9
		-----	-----	-----	-----	-----
<b>69.2</b>		<b>Race 1 (Fom: 1)</b>	<b>Race 1 (Fom: 1)</b>	<b>Pathotyp 1 (Fom: 1)</b>	<b>Raza 1 (Fom: 1)</b>	
		(+) (+)				
<b>QL</b>	absent	absente	fehlend	ausente	Jaune Canari 2 Atos, Charentais T, Védrantais	1
	present	présente	vorhanden	presente	Arapaho, Jador, Rubbens Cadence, Charentais Fom-2, Dibango, Jubilo, Karakal	9
		-----	-----	-----	-----	-----
<b>69.3</b>		<b>Race 2 (Fom: 2)</b>	<b>Race 2 (Fom: 2)</b>	<b>Pathotyp 2 (Fom: 2)</b>	<b>Raza 2 (Fom: 2)</b>	
		(+) (+)				
<b>QL</b>	absent	absente	fehlend	ausente	Arapaho, Jaune Canari 2, Rubbens Atos, Charentais Fom-2, Charentais T, Dibango, Marianna	1
	present	présente	vorhanden	presente	Anasta, Cléo, Jador, Cadence, Charentais Fom- 1, Jubilo, Karakal, Perlita, Védrantais	9
		-----	-----	-----	-----	-----
<b>69.4 B</b>	<b>VG</b>	<b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Race 1.2 (Fom: 1.2)</b>	<b>Résistance à <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Race 1.2 (Fom: 1.2)</b>	<b>Resistenz gegen <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Pathotyp 1.2 (Fom: 1.2)</b>	<b>Resistencia al <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> Raza 1.2 (Fom: 1.2)</b>	
		(+) (+)				
<b>QN</b>	absent	absente	fehlend	ausente	Graffio, Prity, Virgos	1
<b>QL</b>	present	présente	vorhanden	presente	Isabelle, Kyriel, Lunasol, Meliance, Piboule	9
	moderately resistant	moyennement résistant	mäßig resistent	moderadamente resistente	Lunasol	2
	highly resistant	hautement résistant	hochresistent	altamente resistente	Dinero, Isabelle	3

Proposed revision of explanation Ad. 69 “Resistances to *Fusarium oxysporum* f. sp. *melonis* (Fom) - races 0, 1, 2, and 1.2” in Chapter 8.2 “Explanations for individual characteristics”

Ads. 69-A: 69.1 - 69.3: Resistance to *Fusarium oxysporum* f. sp. *melonis*, races 0, 1 and 2 (Fom: 0, Fom: 1, Fom: 2)

1.	Pathogen	<i>Fusarium oxysporum</i> f. sp. <i>melonis</i> races 0, 1, and 2
2.	Quarantine status	No
3.	Host species	Melon - <i>Cucumis melo</i>
4.	Source of inoculum	e.g. GEVES (FR) <sup>9</sup>
5.	Isolate	<u>Fom: 0, Fom: 1, Fom: 2</u> e.g., Reference strain validated in an inter-laboratory test <sup>10, 11</sup> Fom:0 - Strain MLZ = MAT/REF/04-07-01-03-02 <sup>2</sup>  Fom: 1 - Strain FOM 26 = MAT/REF/04-07-01-01 <sup>2</sup>  Fom: 2 - Strain F185
6.	Establishment isolate identity	<u>use differential varieties:</u> <u>Test on differential hosts (potentially including Durango, see 13.).</u> The most recent table is available through ISF at <a href="https://www.worldseed.org/our-work/plant-health/differential-hosts/">https://www.worldseed.org/our-work/plant-health/differential-hosts/</a> <u>Situation July 2019</u>

Differential host	Gene present	Fom: 0*	Fom: 1*	Fom: 2*	Fom: 1.2*
Charantais T*	-	S	S	S	S
Védantais*, Doublon*	<i>Fom-1</i>	HR	S	HR	S
Charantais Fom-2*, CM17187*	<i>Fom-2</i>	HR	HR	S	S
Isabelle*	<i>Polygenic?</i>	HR	HR	HR	IR

S = susceptible; HR = highly resistant; IR = intermediate

\*differential hosts and isolates that are used by the seed sector

Courtesy of Worldseed.org website

	Gene	Race 0	Race 1	Race 2
Charentais T		S	S	S
Védantais	<i>Fom-1</i>	R	S	R
Charentais Fom-2	<i>Fom-2</i>	R	R	S
Isabelle, Jador		R	R	R

7.	Establishment pathogenicity	use susceptible melon varieties
8.	Multiplication inoculum	
8.1	Multiplication medium	on agar medium – e.g., Potato Dextrose Agar, <u>Malt agar at 20°C to 25°C</u>
8.2	Multiplication variety	-
8.3	Plant stage at inoculation	-
8.4	<del>Inoculation medium</del>	<del>on liquid medium</del>
8.5	Inoculation method	-
8.6	Harvest of inoculum	<u>7–10-day-old culture</u>

<sup>9</sup> [matref@geves.fr](mailto:matref@geves.fr)

<sup>10</sup> Harmores 3 CPVO project ([https://cpvo.europa.eu/sites/default/files/documents/report\\_harmores\\_3\\_final\\_meeting\\_v0\\_0.pdf](https://cpvo.europa.eu/sites/default/files/documents/report_harmores_3_final_meeting_v0_0.pdf))

<sup>11</sup> ISF EG DRT Fom: 2 resistance in Melon – Link to include – *not yet available*

8.7	Check of harvested inoculum	-
8.8	Shelf life /viability inoculum	Between 4 to 8 h or keep cool to prevent spore germination
9.	Format of the test	
9.1	Number of plants per genotype	at least 20 at least 30 plants, it is important to have at least 5 non-inoculated plants per genotype to be able to judge growth reduction
9.2	Number of replicates	At least e.g. 3 replicates (3 x10)
9.3	Control varieties	Jaune Canari 2 (susceptible) Védrantais, Arapaho, Rubbens, Anasta, Cleo (resistant, depending on the considered race)

	Gene	Race 0	Race 1	Race 2
Jaune Canari 2		S	S	S
Védrantais	Fom-1	R	S	R
Arapaho, Rubbens	Fom-2	R	R	S
Anasta, Cleo		R	R	R

9.3.1	Control varieties for race 0	Resistance absent: Charentais T Resistance present: Charentais Fom-2, Védrantais
9.3.2	Control varieties for race 1	Resistance absent: Charentais T, Védrantais Resistance present: Charentais Fom-2
9.3.3	Control varieties race 2	Resistance absent: Marianna Resistance present: Perlita, Charentais Fom-1, Védrantais
9.4	Test design	3 replicates of 10 plants to allow statistical analysis (in different trays) and at least 5 non-inoculated plants per genotype.
9.5	Test facility	glasshouse or climatic room
9.6	Temperature	- Fom: 0 and Fom: 1: 18-25 24°C - Fom: 2: 24°C
9.7	Light	- Fom: 0 and Fom: 1: At least 12h - Fom: 2: 16h
9.8	Season	all seasons
9.9	Special measures	optional: shading (no direct sunlight during 12 h after inoculation) - Fom: 0 and Fom: 1: Recommend having really 18°C at night and not above 24°C during the day.
10.	Inoculation	
10.1	Preparation inoculum	aerated culture 7-10 days, e.g., Czapek-Dox broth some isolates need filtration or centrifugation resuspend the pelleted spores in demineralized water Scrape spore cultures with water from agar medium (see 8.1) or optional multiplication on liquid medium (e.g., Messiaen (1991) synthetic liquid medium, sucrose 50g/L, on permanent agitator-shaker or aerated Czapek-Dox culture medium for 5-7 days at room temperature). Remark: Beware of toxin productions by some isolates (see remark under 13.)
10.2	Quantification inoculum	spore count; adjust to 10 <sup>6</sup> -10 <sup>7</sup> per mL 4x10 <sup>5</sup> to 1x10 <sup>6</sup> sp /mL
10.3	Plant stage at inoculation	cotyledon expanded
10.4	Inoculation method	soaking of the root system in a suspension of liquid medium of fungus at least 30 sec - 5 min Plant at the inoculation stage are harvested carefully, roots and hypocotyls are immersed in spore suspension for 2-15 min; trimming of roots is an option; transplant in trays.

10.5	First observation	<del>7 days post inoculation</del> 1 <sup>st</sup> notation: symptoms on Resistance absent (susceptible) control at classes 2 and 3 with a strong proportion at class 3
10.6	Second observation	<del>14–20 days post inoculation</del> A second notation can be necessary to re-evaluate some unclear varieties
<del>10.7</del>	<del>Final observations</del>	<del>20 days post inoculation</del>
11.	Observations	
11.1	Method	Visual <u>observation</u> , <u>comparative</u>
11.2	Observation scale	
	<del>[1] absent</del>	<del>Growth retardation in combination with yellowing or wilting cotyledons (useful for judging the severity of the attack), possible internal vessel browning, death of plant.</del>
	<del>[9] present</del>	<del>no symptoms</del>

non-inoculated plant	Class 0	Class 1
At least 5 plants	Healthy plant: no symptoms of yellowing and wilting, could be some growth reduction due to inoculation stress compared to mock. Sometimes in the mock we can observe some yellowing, different from the symptoms of <i>Fusarium</i>	Light symptoms of yellowing/wilting

Class 2	Class 3	
typical symptoms: yellowing, wilting and necrosis, stunting (growth stopped)	Death of plant (Dead)	
		<p>Other symptoms of vein clearing could be difficult to judge. It is advised to make a later notation to observe the evolution of these symptoms over the time.</p>

Courtesy of GEVES-SNES in the framework of CPVO Harmores project.

11.3	Validation of test	<p><del>on standards</del></p> <p>Validation on controls.</p> <p>In case of the Fom: 0 and Fom:1 tests: Controls expected response: Resistance absent: most of the plants at classes 2 and 3 Resistance present: most of the plants at classes 0 and 1, sometimes very few plants at classes 2 or 3.</p> <p>In case of the Fom: 2 test Controls expected response:</p> <ul style="list-style-type: none"> <li>• Susceptible controls, with UPOV characteristic state 'Resistance absent', should have most of the plants in observation classes 2 or 3, and few or no plants in observation classes 0 or 1. <ul style="list-style-type: none"> <li>○ Marianna, the susceptible control is less susceptible than Charentais Fom-2, Charentais T</li> </ul> </li> <li>• Resistant controls should have most of the plants in observation classes 0 or 1, and few or no plant in observation classes 2 or 3.</li> </ul> <p>Perlita, the lower threshold resistance control, should have at least some plants in observation class 1, 2, or 3. It has to be less resistant than Charentais Fom-1, Védrentais.</p>
11.4	Off-types	-
12.	Interpretation of data in terms of UPOV characteristic states	<p><del>QL</del></p> <p><u>In case of varieties with a response between the susceptible (resistance absent) and the resistant control, repeat the test.</u> <u>In case of confirmation of the result, the variety will be judged heterogeneous.</u> <u>In case of unclear results, retest or test in another lab.</u></p>

Resistance to Fom: 0 and Fom: 1

Not different from the  
resistance absent  
control

Inconclusive result  
retest

Not different from the  
resistance present  
control



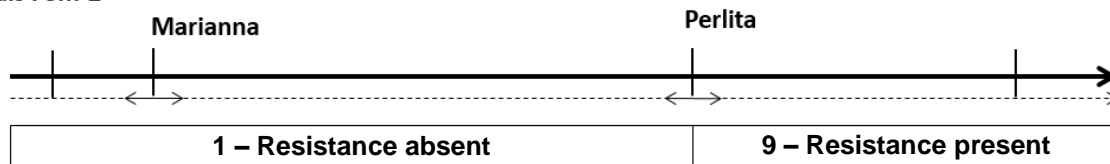
1 – Resistance absent

9 – Resistance present

Resistance to Fom: 2

Charentais T,  
Charentais Fom-2

Charentais Fom-1,  
Védrantais



13.	Critical control points	<p><del>For Race 1.2 the modified protocol on the next page should be used.</del></p> <p>For race 2, the control Perlita, with the <i>Fom-3</i> gene, allows to validate the capacity of the isolate to partially attack this variety.</p> <p>In the case of inoculum increased in e.g. Messiaen (1991) synthetic liquid medium, on permanent agitator-shaker, inoculum can be used after 5 to 7 days.</p> <p>For race 0 and 1, dilution 1/12 is recommended, while it must not be less than 1/20 for race 2. At a lower dilution (higher concentration of the medium), it has been observed that toxins released in the medium by the race 2 can cause some yellowing of melon plants, even if they are resistant. Alternatively, spores can be “washed” by resuspending a mass of spores collected on a Millipore filter with vacuum force.</p>
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Ad 69-B 69.4: Resistance to *Fusarium oxysporum* f. sp. *melonis* race 1.2 (Fom: 1.2)

1.	Pathogen	<i>Fusarium oxysporum</i> f. sp. <i>melonis</i> race 1.2 (Fom: 1.2)																														
2.	Quarantine status	No																														
3.	Host species	Melon - <i>Cucumis melo</i> L.																														
4.	Source of inoculum	GEVES (FR) <sup>12</sup> , Naktuinbouw (NL)																														
5.	Isolate	Fom: 1.2 (moderately aggressive): TST strain e.g., Reference strain validated in an inter-laboratory test <sup>3</sup> Fom: 1.2 - Strain TST = MAT/REF/04-07-01-04 <sup>2</sup>																														
6.	Establishment isolate identity	use differential varieties: Védrantais, Virgos (susceptible) Lunasol (moderately resistant) Dinero, Isabelle (highly resistant) The most recent table is available through ISF at <a href="https://www.worldseed.org/our-work/plant-health/differential-hosts/Situation%20July%202019">https://www.worldseed.org/our-work/plant-health/differential-hosts/Situation July 2019</a>																														
<table border="1"> <thead> <tr> <th>Differential host</th> <th>Gene present</th> <th>Fom: 0*</th> <th>Fom: 1*</th> <th>Fom: 2*</th> <th>Fom: 1.2*</th> </tr> </thead> <tbody> <tr> <td>Charantais T*</td> <td>-</td> <td>S</td> <td>S</td> <td>S</td> <td>S</td> </tr> <tr> <td>Védrantais*, Doublon*</td> <td><i>Fom-1</i></td> <td>HR</td> <td>S</td> <td>HR</td> <td>S</td> </tr> <tr> <td>Charantais Fom-2*, CM17187*</td> <td><i>Fom-2</i></td> <td>HR</td> <td>HR</td> <td>S</td> <td>S</td> </tr> <tr> <td>Isabelle*</td> <td><i>Polygenic?</i></td> <td>HR</td> <td>HR</td> <td>HR</td> <td>IR</td> </tr> </tbody> </table> <p>S = susceptible; HR = highly resistant; IR = intermediate *differential hosts and isolates that are used by the seed sector</p> <p style="text-align: right;">Courtesy of Worldseed.org website</p>			Differential host	Gene present	Fom: 0*	Fom: 1*	Fom: 2*	Fom: 1.2*	Charantais T*	-	S	S	S	S	Védrantais*, Doublon*	<i>Fom-1</i>	HR	S	HR	S	Charantais Fom-2*, CM17187*	<i>Fom-2</i>	HR	HR	S	S	Isabelle*	<i>Polygenic?</i>	HR	HR	HR	IR
Differential host	Gene present	Fom: 0*	Fom: 1*	Fom: 2*	Fom: 1.2*																											
Charantais T*	-	S	S	S	S																											
Védrantais*, Doublon*	<i>Fom-1</i>	HR	S	HR	S																											
Charantais Fom-2*, CM17187*	<i>Fom-2</i>	HR	HR	S	S																											
Isabelle*	<i>Polygenic?</i>	HR	HR	HR	IR																											
7.	Establishment pathogenicity	use susceptible melon varieties																														
8.	Multiplication inoculum																															
8.1	Multiplication medium	on agar medium e.g., Potato Dextrose Agar, Sabouraud, at 20°C to 25°C																														
8.2	Multiplication variety	-																														
8.3	Plant stage at inoculation	-																														
8.4	Inoculation medium	on liquid medium																														
8.5	Inoculation method	-																														
8.6	Harvest of inoculum	4-10 day-old culture																														
8.7	Check of harvested inoculum	-																														
8.8	Shelf life/viability inoculum	-																														
9.	Format of the test																															
9.1	Number of plants per genotype	at least 30 30 plants per variety plus 5 non-inoculated controls																														
9.2	Number of replicates	e.g., At least 3 x 10 plants, in different trays																														
9.3	Control varieties	Resistance absent: Virgos Resistance present: Piboule and Lunasol and Isabelle (Isabelle is expected to have a lower disease index (DI) (= higher resistance than Piboule and Lunasol). Piboule and Lunasol are both needed to illustrate the lower level to intermediate resistance of resistance. Their resistance is based on other genetics and may have different levels in different labs.																														
	[1] susceptible	Védrantais, Virgos																														
	[2] moderately resistant	Lunasol (the lowest accepted level)																														

<sup>12</sup> [matref@geves.fr](mailto:matref@geves.fr)



<sup>3</sup> Harmores 3 CPVO project ([https://cpvo.europa.eu/sites/default/files/documents/report\\_harmores\\_3\\_final\\_meeting\\_v0\\_0.pdf](https://cpvo.europa.eu/sites/default/files/documents/report_harmores_3_final_meeting_v0_0.pdf))



	<del>[3] highly resistant</del>	<del>Dinero, Isabelle, Jador</del>
9.4	Test design	<del>3 replicates of 10 plants to allow statistical analysis (in different trays) and at least 5 non-inoculated plants per genotype.</del>
9.5	Test facility	<del>glasshouse or climatic room</del>
9.6	Temperature	<del>18-25 24°C</del>
9.7	Light	<del>at least 12h</del>
9.8	Season	<del>All seasons in a climatic room / in a greenhouse be aware of the strong environmental effect: winter could be too severe and summer could be too mild.</del>
9.9	Special measures	<del>optional shading (no direct sunlight during 12 h after inoculation)</del>
10.	Inoculation	
10.1	Preparation inoculum	<del>aerated culture 7-10 d old — e.g.: Czapek-Dox broth Scrape cultures with water on agar medium (see 8.1) or optional multiplication on liquid medium (e.g., Potato Dextrose Broth (PDB), Czapek-Dox culture medium for 7 days at room temperature and darkness or Messiaen (1991) synthetic liquid medium, sucrose 50 g/L, on permanent agitator-shaker, at room-temperature, inoculum can be used after 5 to 7 days)</del>
10.2	Quantification inoculum	<del>spore count; adjust to <math>2 \cdot 10^4 - 10^5</math> per ml <math>1 \times 10^5 - 1 \times 10^6</math> sp/mL, depending on inoculation method (see 10.4) and lab conditions</del>
10.3	Plant stage at inoculation	<del>cotyledons expanded, first leaf emerging</del>
10.4	Inoculation method	<del>soaking of the trays in spore suspension; 700 ml for a tray with 25 - 30 plants, plants are not uprooted One of two methods can be used for inoculation. - Absorption: Absorption of a suspension of spores, e.g., 700mL of a suspension at <math>1 \cdot 10^5</math> sp/mL for 50 plants in a tray 30 cm*30 cm. - Injection: Injection of a suspension of spores into the soil at the base of the plant, e.g., 5mL at <math>10^6</math> sp /mL per plant.</del>
10.5	First observation	<del>7 - 14 days post inoculation</del>
10.6	Second observation	<del>14 - 21 days post inoculation</del>
10.7	Final observations	<del>21 - 28 days post inoculation 1<sup>st</sup> notation: symptoms on susceptible control at least at class 3 [generally 10-21 dpi]. A 2<sup>nd</sup> notation can be necessary to reevaluate some unclear varieties.</del>
11.	Observations	
11.1	Method	<del>Visual observation, comparative</del>
11.2	Observation scale	<del>symptoms:</del>
	<del>[1] susceptible</del>	<del>Védrantais: growth retardation, yellow cotyledons, drying, possible internal vessel browning, death of the plant</del>
	<del>[2] moderately resistant</del>	<del>Symptoms may be present, but the level of expression must be distinctly lower than the susceptible control variety. = the lowest level of resistance is defined by the behavior of Lunasol</del>
	<del>[3] highly resistant</del>	<del>Symptoms may be present, but the level of expression must be lower than the moderately control variety Lunasol.</del>

Non-inoculated plants	Class 0	Class 1
Varieties must be compared to the non-inoculated plants.	Healthy plant, the whole plant is green or at the same level than the mock. Just a light	Light level of symptoms, light yellowing on cotyledons and/or leaves without necrosis

	yellowing can be accepted on the mock	
		

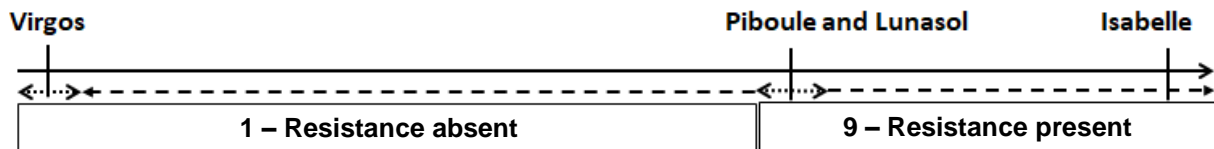
Class 2	Class 3	Class 4
Moderate level of symptoms, yellowing on cotyledon and/or leaves, starting of necrosis and wilting but not extended	Severe symptoms of yellowing and/or wilting on cotyledons and/or leaves with extended necrosis	Dead plant, no green leaf part or hypocotyl is dry
		

Courtesy of GEVES-SNES in the framework of CPVO Harmores project.

11.3	Validation of test	<p>Validation on controls. Controls expected response:</p> <ul style="list-style-type: none"> <li>- Resistance present <del>Intermediate Resistant</del>: Most plants in classes 0 and 1, in some cases with few plants in 2, 3, 4. Low level of disease index (DI) generally below 40%. A difference of disease index is generally observed between Piboule and Lunasol compared to Isabelle</li> <li>- Resistance absent <del>Susceptible</del>: Most plants in classes 3 and 4, in some cases with few plants at class 0, 1, or 2. Very high disease index (DI) above 80%.</li> </ul>
11.4	Off-types	<p><del>calibrate with Lunasol</del></p> <ul style="list-style-type: none"> <li>-</li> </ul>

12.	Interpretation of data in terms of UPOV characteristic states	<p><b>QN</b></p> <p>Interpretation of varieties depending on controls (figure 1)</p> <p>Note 1 = Resistance absent <del>Intermediate resistance absent = susceptibility</del></p> <p>Note 9 = Resistance present <del>Intermediate resistance present</del></p> <p>Quantitative analysis is based on the disease index (DI) AND the distribution of plants per class compared to the controls</p> <p>The varieties statistically similar to the <del>intermediate resistant controls</del> or with a lower disease index (DI) have to be judged as <del>intermediate resistant</del>.</p> <p>The varieties between the susceptible and the <del>intermediate resistant controls</del> have to be judged as susceptible. (<del>not resistant enough to be considered</del>)</p> <p>If <del>not clear results</del> are not clear, the use of statistics is highly recommended <del>suggested</del>.</p>
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Resistance to Fom:1-2:



$$DI = \frac{(N0 * 0) + (N1 * 1) + (N2 * 2) + (N3 * 3) + (N4 * 4)}{(N0 + N1 + N2 + N3 + N4) * 4} * 100$$

Nx : number of plants at class x

Figure 1: disease index (DI) formula

13.	Critical control points	<p>A moderately aggressive type of Fom: 1.2 should be used as this is likely to show the difference between the presence and absence of resistance most clearly.</p> <p>There are two types of <i>Fusarium oxysporum</i> f. sp. <i>melonis</i>, Fom:1.2, viz. Fom: 1.2y which is a yellowing type with yellowing symptoms on leaves and another type and Fom: 1.2w which is a wilt type with wilting symptoms on leaves.</p>
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Proposed revision of Characteristics 70.1 to 70.5 "Resistances to *Podosphaera xanthii* (Px) - races 1, 2, 3, 5, 3.5"

70.	VG	Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i> ) (Powdery mildew)	Résistance à <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i> ) (oïdium)	Resistenz gegen <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i> ) (Echter Mehltau)	Resistencia a <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i> ) (Oidio)		
70.1		<b>Race 1 (Px: 1)</b>	<b>Race 1 (Px: 1)</b>	<b>Pathotyp 1 (Px: 1)</b>	<b>Raza 1 (Px: 1)</b>		
(+)							
QN		susceptible absent or low	sensible absente ou faible	anfällig fehlend oder gering	susceptible ausente o baja	Jaune Canari 2, Védrantais	1
		moderately resistant medium	moyennement résistant moyenne	mäßig resistent mittel	moderadamente resistente media	Escrito	2
		highly resistant high	hautement résistant élevée	hochresistent hoch	altamente resistente alta	Anasta, Cézanne Arum	3
70.2		<b>Race 2 (Px: 2)</b>	<b>Race 2 (Px: 2)</b>	<b>Pathotyp 2 (Px: 2)</b>	<b>Raza 2 (Px: 2)</b>		
(+)							
QN		susceptible absent or low	sensible absente ou faible	anfällig fehlend oder gering	susceptible ausente o baja	Galoubet, Védrantais	1
		moderately resistant medium	moyennement résistant moyenne	mäßig resistent mittel	moderadamente resistente media	Escrito, Pendragon	2
		highly resistant high	hautement résistant élevée	hochresistent hoch	altamente resistente alta	Anasta, Cézanne Arum	3
70.3		<b>Race 3 (Px: 3)</b>	<b>Race 3 (Px: 3)</b>	<b>Pathotyp 3 (Px: 3)</b>	<b>Raza 3 (Px: 3)</b>		
(+)							
QN		susceptible absent or low	sensible absente ou faible	anfällig fehlend oder gering	susceptible ausente o baja	Védrantais	1
		moderately resistant medium	moyennement résistant moyenne	mäßig resistent mittel	moderadamente resistente media	Nettune Arago, Durango	2
		highly resistant high	hautement résistant élevée	hochresistent hoch	altamente resistente alta	Batista, Godiva Arum	3
70.4		<b>Race 5 (Px: 5)</b>	<b>Race 5 (Px: 5)</b>	<b>Pathotyp 5 (Px: 5)</b>	<b>Raza 5 (Px: 5)</b>		
(+)							
QN		susceptible absent or low	sensible absente ou faible	anfällig fehlend oder gering	susceptible ausente o baja	Védrantais	1
		moderately resistant medium	moyennement résistant moyenne	mäßig resistent mittel	moderadamente resistente media	Hugo, Pendragon-Arago, Durango	2
		highly resistant high	hautement résistant élevée	hochresistent hoch	altamente resistente alta	Arapaho Arum	3
70.5		<b>Race 3-5 (Px: 3.5)</b>	<b>Race 3-5 (Px: 3.5)</b>	<b>Pathotyp 3-5 (Px: 3.5)</b>	<b>Raza 3-5 (Px: 3.5)</b>		
(+)							
QN		susceptible absent or low	sensible absente ou faible	anfällig fehlend oder gering	susceptible ausente o baja	Védrantais	1
		moderately resistant medium	moyennement résistant moyenne	mäßig resistent mittel	moderadamente resistente media	Cisco Arago, Durango	2
		highly resistant high	hautement résistant élevée	hochresistent hoch	altamente resistente alta	90625 Arum	3

Revision of explanation Ads. 70.1 to 70.3, 71 “Resistances to *Podosphaera xanthii* (Px), Resistance to *Golovinomyces cichoracearum* (Gc) (Powdery mildew)” in Chapter 8.2 “Explanations for individual characteristics”

Ads. 70.1 to 70.3 5: Resistance to *Podosphaera xanthii* (Px) (~~ex~~ *Sphaerotheca fuliginea*) (Powdery mildew) Px (~~Sf~~) races 1, 2, 3, 5, 3.5 (Px: 1, 2, 3, 5, 3.5)

Ad. 71: Resistance to *Golovinomyces cichoracearum* (Gc) (*Erysiphe cichoracearum*) (Powdery mildew), race 1 (Gc: 1)

1.	Pathogen	Powdery mildew: <i>Podosphaera xanthii</i> ( <del>ex</del> <i>Sphaerotheca fuliginea</i> ) races 1, 2, 3, 5 and 3.5 <i>Golovinomyces cichoracearum</i> ( <del>ex</del> <i>Erysiphe cichoracearum</i> ) race 1 <del>Only <i>Podosphaera xanthii</i> was validated in Harmores 3 project.</del>
2.	Quarantine status	No
3.	Host species	Melon - <i>Cucumis melo</i> L.
4.	Source of inoculum	GEVES (FR) <sup>13</sup>
5.	Isolate	Px: races 1, 2, 3, 5 and 3.5; e.g., Reference strain validated in an inter-laboratory test <sup>6</sup> Px: 1 - Strain Sm 3 = MAT/REF/04-07-03-01 <sup>7</sup> Px: 2 - Strain S87-7 = MAT/REF/04-07-03-02 <sup>7</sup> Px: 3 - Strain 00Sm39 = MAT/REF/04-07-03-04-02 <sup>7</sup> Px: 5 - Strain 98Sm65 = MAT/REF/04-07-03-03-01-02 <sup>7</sup> Px: 3.5 - Strain 04Sm2 = MAT/REF/04-07-03-05-01 <sup>7</sup>  Gc: 1 - Strain GEVES = MAT/REF/04-07-02-01) <sup>3</sup>
6.	Establishment isolate identity	on differentials (table 1)

	Powdery Mildew					
	<i>Podosphaera xanthii</i> ( <i>Sphaerotheca fuliginea</i> )					<i>Golovinomyces cichoracearum</i> ( <i>Erysiphe cichoracearum</i> )
	race 1	race 2	race 3	race 5	race 3-5	race 1
Védreantais	S	S	S	S	S	S
Nantais Oblong	S	S	S	S	S	R
PMR 45	R	S	S	S	S	S
Edisto 47, WMR 29	R	R	R	S	S	S
PI 124112, 90625	R	R	R	R	R	R
PMR 5	R	R	S	R	S	R
PI 414723	R	R	IR	R	R/IR	R

Legend: S – susceptible (high sporulation); R – resistant (low sporulation), IR (moderately resistant)

<sup>3</sup> [matref@geves.fr](mailto:matref@geves.fr)

<sup>6</sup> Harmores 3 CPVO project ([https://cpvo.europa.eu/sites/default/files/documents/report\\_harmores\\_3\\_final\\_meeting\\_v0\\_0.pdf](https://cpvo.europa.eu/sites/default/files/documents/report_harmores_3_final_meeting_v0_0.pdf))

Table 2:

Races of *Podosphaera xanthii* (Px) and *Golovinomyces cichoracearum* (Gc), J. McCreight and M. Pitrat

	<i>Podosphaera xanthii</i>							<i>Golovinomyces cichoracearum</i>	
	Race 0	Race 1	Race 2	Race 3	Race 4	Race 5	Race 3.5	Race 0	Race 1
Iran H	S	S	S	S	S	S	S	S	S
Védrantais	R	S	S	S	S	S	S	R	S
PMR45	R	R	S	S	S	S	S	R	S
WMR29	R	R	R	R	S	S	S	R	S
Edisto 47	R	R	R	R	R	S	S	R	S
MR-1, PI124112	R	R	R	R	R	R	R	R	R
PMR5	R	R	R	S	S	R	S	R	R
Nantais Oblong	R	S	S	S	S	S	S	R	R

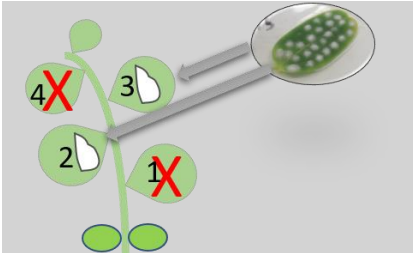
7.	Establishment pathogenicity	use susceptible melon varieties								
8.	Multiplication inoculum									
8.1	Multiplication medium	<del>detached cotyledon in Petri-dish on 0.35 – 0.5% Agar, 1-2% mannitol, possible add of 1% sucrose</del> Melon plantlets								
8.2	Multiplication variety	<del>susceptible varieties</del> Susceptible variety, for example Védrantais. For higher isolates like 3.5 or 5, a variety with broken resistance is recommended to keep the isolate pure.								
8.3	Plant stage at inoculation	<del>young, unfolded cotyledon; decontaminated with e.g., 0.05% mercuric chloride or 3 to 5% bleach (NaClO + NaCl)</del> Cotyledon								
8.4	<del>Inoculation medium</del>	Air								
8.5	Inoculation method	<del>scatter conidia on the cotyledons transferred by blowing</del> Sowing in substrate, for example soil or disinfected peat inside a closed mini glasshouse. When the cotyledons have expanded, remove them from the plant. Disinfect the cotyledons by soaking them for 3 minutes in a mercuric chloride solution (0.05%) or in sodium hypochlorite solution. Rinse them with sterilized water. Dry the cotyledons with sterile paper towel, then place them in Petri dishes with the following medium: <table style="margin-left: 40px;"> <tr> <td>Sucrose</td> <td>10g</td> </tr> <tr> <td>Mannitol</td> <td>20g</td> </tr> <tr> <td>Agar</td> <td>5g</td> </tr> <tr> <td>Distilled water</td> <td>1 liter</td> </tr> </table> Scatter conidia on the cotyledons and blow them or deposit conidia at the surface of cotyledons. Incubate the inoculated cotyledons in Petri dishes for example at 23°C during 14 hours in the light and at 18°C during 10 hours in the dark or 17°C permanently under very low light intensity. 9 to 11 days after the inoculation, the cotyledons will be covered with conidia and can be used as an inoculum.	Sucrose	10g	Mannitol	20g	Agar	5g	Distilled water	1 liter
Sucrose	10g									
Mannitol	20g									
Agar	5g									
Distilled water	1 liter									
8.6	Harvest of inoculum	<del>use cotyledons with strong sporulation</del> Sporulation on cotyledons								
8.7	Check of harvested inoculum	<del>check presence of spores</del>								
8.8	Shelf life /viability inoculum	<del>on cotyledon, 17-23°C, under very low light intensity; maximum storage time is 15 days, after the inoculation</del> Remark: In case of longer-term preservation, inoculate locally with a few spores, store at 14°C/12h low light per day Maximum 1 to 1.5 months after the inoculation.								

9.	Format of the test	
9.1	Number of plants per genotype	at least 16 plants At least 20 plants per variety and controls, 5 plants for other differentials.
9.2	Number of replicates	e.g., 3
9.3	Control varieties	-

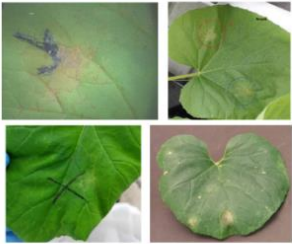




	Powdery Mildew					<i>Golovinomyces cichoracearum</i>
	<i>Podosphaera xanthii</i>					
	race 1	race 2	race 3	race 5	race 3-5	race 1
Susceptible	Jaune Canari 2, Védrantais	Galoubot, Védrantais	Védrantais	Védrantais	Védrantais	Védrantais
moderately resistant	Escrito	Escrito, Pendragon	Nettuno	Hugo, Pendragon	Ciseo	Anasta
highly resistant	Anasta, Cézanne	Anasta, Cézanne	Batista, Godiva	Arapaho	90625	Heliobel

		<p><u>For <i>Podosphaera xanthii</i> (Px) race 1, resistance</u></p> <ul style="list-style-type: none"> <li>absent or low Susceptible: Védrantais</li> <li>medium Intermediate resistant: Escrito</li> <li>high Resistant: Arum</li> </ul> <p><u>For <i>Podosphaera xanthii</i> (Px) race 2, resistance:</u></p> <ul style="list-style-type: none"> <li>absent or low Susceptible: Védrantais</li> <li>medium Intermediate resistant: Escrito, Pendragon</li> <li>high Resistant: Arum</li> </ul> <p><u>For <i>Podosphaera xanthii</i> (Px) races 3, 5, 3.5, resistance:</u></p> <ul style="list-style-type: none"> <li>absent or low Susceptible: Védrantais</li> <li>medium Intermediate resistant: Arago, Durango</li> <li>high Resistant: Arum</li> </ul> <p><u>For <i>Golovinomyces cichoracearum</i> (Gc) race 1, resistance:</u></p> <ul style="list-style-type: none"> <li>absent or low Susceptible: Escrito, Score, Védrantais</li> <li>medium Intermediate resistant: Flores, Anasta</li> <li>high Resistant: Cézanne, Heliobel, Théo</li> </ul>
9.4	Test design	leaf discs placed on 0.4% agar with 1-4% mannitol and possible add of 0.003% benzimidazole Include differentials to validate the race (at least 5 plants per differentials) and compare the level of sporulation.
9.5	Test facility	climatic room Climatic chamber or greenhouse
9.6	Temperature	20-24°C
9.7	Light	12 to 24h darkness after inoculation At least 12 hours
9.8	Season	-
9.9	Special measures	Inoculation tower needed for even distribution of dry spores.
10.	Inoculation	
10.1	Preparation inoculum	-
10.2	Quantification inoculum	-



10.3	Plant stage at inoculation	<p><u>Routine method:</u> leaf disks, 2 cm in diameter, from young plants.  <u>Complementary method, if necessary:</u> young plants          Whole plants at 3-4 true leaf fully expanded stage. Inoculation on the leaves 2 and 3 indicated on the diagram below.</p>  <p>Courtesy of GEVES-SNES in the framework of CPVO Harmores project.</p>
10.4	Inoculation method	<p><u>Routine method:</u> on leaf disks: inoculation tower needed for even distribution of dry spores.  <u>Complementary method:</u> take spores from a cotyledon covered with conidia and deposit them on a leaf or blow the spores from a cotyledon.          Take spores from a cotyledon already covered with conidia and deposit them on a leaf. Different isolates can be tested on the same plant (or the same leaf) if the local deposit is well separated from each other and if a mark indicates the place of the deposit.</p>
10.5	<u>First observation</u>	8-10 days post inoculation
10.6	<u>Second observation</u>	-
10.7	Final observations	<p>11-12 days post inoculation          The date of notation should be chosen based on expected symptoms on the three controls. Sporulation should be well expressed on the susceptible control.</p>
11.	Observations	
11.1	Method	Visual <u>observation of sporulation</u>
11.2	Observation scale	
	[1] susceptible	medium or intense sporulation all over the leaf disc surface
	[2] intermediate	weak sporulation all over the surface or isolated colonies on more than 10% of the surface
	[3] resistant	isolated colonies on less than 10% of the surface or no sporulation

Class 1: No development of the fungus (no mycelium or dead mycelium) or no sporulation	Class 3: weak sporulation	Class 5: moderate sporulation	Class 9: strong sporulation
			
 <p>Example of contamination by environment on the susceptible control, test not validated</p>			



11.3	Validation of test	<p><del>on controls</del></p> <p><u>Validation on controls.</u></p> <p><u>Additional information for expected responses of <i>Podosphaera xanthii</i> controls</u></p> <p><u>Resistance absent or low</u></p> <ul style="list-style-type: none"> <li>Plants at class 9, or most of the plants at class 9 and few plants at class 5 (high disease index).</li> <li>Few plants at class 3 but in this case the resistant controls should be all at class 1 and the intermediate resistant control at classes 3 and 1.</li> <li>No plants at class 1.</li> </ul> <p><u>Resistance medium</u></p> <ul style="list-style-type: none"> <li>Between the resistant and the susceptible control.</li> <li>Generally, plants at classes 3 and 5.</li> </ul> <p><u>Resistance high</u></p> <ul style="list-style-type: none"> <li>Plants at class 1, or most of the plants at class 1 and few plants at class 3 (very low disease index).</li> <li>Plants at class 3 but in this case the susceptible control should be all at class 9.</li> <li>No plants at classes 5 or 9.</li> </ul>
11.4	Off-types	-
12.	Interpretation of data in terms of UPOV characteristic states	<p><del>QN</del></p> <p><u>Interpretation of varieties depending on controls (figure 1)</u></p> <p><u>Resistance</u></p> <p><u>Note 1 = Resistance absent or low = susceptibility</u></p> <p><u>Note 2 = medium-Intermediate resistance present</u></p> <p><u>Note 3 = high Resistance present</u></p> <p><u>Quantitative analysis is based on the disease index AND the distribution of plants per class compared to the controls.</u></p> <p><u>Additional information for <i>Podosphaera xanthii</i> controls:</u></p> <p><u>The varieties between the intermediate resistant and the resistant control have to be judged as intermediate resistant (because they are not resistant enough to be considered resistant).</u></p> <p><u>The varieties between the susceptible and the intermediate resistant control have to be judged as susceptible (because they are not resistant enough to be considered intermediate resistant).</u></p>
<u>Resistance to Px:</u>		
$DI = \frac{(N1*0)+(N3*1)+(N5*2)+(N9*3)}{(N1+N3+N5+N9)*3} * 100$		
<p>NX: Number of plants at class X</p> <p><b>Figure 2: disease index formula</b></p>		
13.	Critical control points	<p><u>To avoid cross contamination, it is advised to not produce inoculum of different races in the same room.</u></p>

Inclusion of characteristics from the Table of Characteristics in the Technical Questionnaire

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5. Characteristics of the variety to be indicated (the number in brackets refers to the corresponding characteristic in Test Guidelines; please mark the note which best corresponds).

Characteristics	Example Varieties	Note
<b>5.1</b>		
<b>(12)</b>		
<b>Inflorescence: sex expression (at full flowering)</b>		
monoecious	Alpha, Categoría	1[ ]
andromonoecious	Piel de Sapo	2[ ]
<b>5.2</b>		
<b>(13)</b>		
<b>Young fruit: hue of green color of skin</b>		
<u>whitish green</u>	<u>Geasol</u>	1[ ]
<u>yellowish green</u>	<u>Fimel</u>	2[ ]
<u>green</u>	<u>Lucas</u>	3[ ]
<u>greyish green</u>	<u>Spanglia</u>	4[ ]
<b>5.3</b>		
<b>(14)</b>		
<b>Young fruit: intensity of green color of skin</b>		
<u>very light</u>	<u>Solarking</u>	1[ ]
<u>very light to light</u>		2[ ]
<u>light</u>	<u>Fimel</u>	3[ ]
<u>light to medium</u>		4[ ]
<u>medium</u>	<u>Eros</u>	5[ ]
<u>medium to dark</u>		6[ ]
<u>dark</u>	<u>Galia</u>	7[ ]
<u>dark to very dark</u>		8[ ]
<u>very dark</u>	<u>Edén</u>	9[ ]

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Characteristics	Example Varieties	Note
<b>5.4</b> <b>(24)</b> <b>Fruit: length</b>		
<u>very short</u>	<u>Doublon, Golden Crispy</u>	1[ ]
<u>very short to short</u>		2[ ]
<u>short</u>	<u>Topper, Védrantais</u>	3[ ]
<u>short to medium</u>		4[ ]
<u>medium</u>	<u>Marina, Spanglia</u>	5[ ]
<u>medium to long</u>		6[ ]
<u>long</u>	<u>Categoría, Toledo</u>	7[ ]
<u>long to very long</u>		8[ ]
<u>very long</u>	<u>Katsura Giant, Valdivia</u>	9[ ]
<b>5.5</b> <b>(25)</b> <b>Fruit: diameter</b>		
<u>very narrow</u>	<u>Banana, Golden Crispy</u>	1[ ]
<u>very narrow to narrow</u>		2[ ]
<u>narrow</u>	<u>Alpha, Maestro</u>	3[ ]
<u>narrow to medium</u>		4[ ]
<u>medium</u>	<u>Categoría, Galia</u>	5[ ]
<u>medium to broad</u>		6[ ]
<u>broad</u>	<u>Albino, Kinka</u>	7[ ]
<u>broad to very broad</u>		8[ ]
<u>very broad</u>	<u>Noir des Carmes</u>	9[ ]

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Characteristics	Example Varieties	Note
<b>5.2.6</b> <b>(28)</b>	<b>Fruit: shape in longitudinal section</b>	
ovate	De Cavaillon, Piolín	1[ ]
medium elliptic	Piel de Sapo	2[ ]
broad elliptic	Corin, Sardo	3[ ]
circular	Alpha, Galia	4[ ]
quadrangular	Zatta	5[ ]
oblate	Jívaro, Noir de Carmes	6[ ]
obovate	Cganchi	7[ ]
elongated	Alficoz, Banana	8[ ]
<b>5.3.7</b> <b>(29)</b>	<b>Fruit: ground color of skin</b>	
white	Albino, Honey Dew	1[ ]
yellow	Amarillo-Canario, Edén, Galia, Passport, Solarking	2[ ]
green	Gohyang, Piel de Sapo	3[ ]
grey	Geaprince, Geamar, Romeo, Sirio, Supporter, Védrantais	4[ ]
<b>5.8</b> <b>(31)</b>	<b>Fruit: hue of ground color of skin</b>	
<u>absent or very weak</u>	<u>Amarillo-Canario, Albino, Piel de Sapo, Sirio</u>	<u>1[ ]</u>
<u>whitish</u>	<u>Romeo</u>	<u>2[ ]</u>
<u>yellowish</u>	<u>Geaprince, Supporter</u>	<u>3[ ]</u>
<u>orange</u>	<u>Edén</u>	<u>4[ ]</u>
<u>ochre</u>	<u>Passport</u>	<u>5[ ]</u>
<u>greenish</u>	<u>Geamar, Honey Dew, Solarking</u>	<u>6[ ]</u>
<u>greyish</u>	<u>Gohyang</u>	<u>7[ ]</u>

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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Characteristics	Example Varieties	Note
<b>5.9</b> <b>(32)</b> <b>Fruit: density of dots</b>		
<u>absent or very sparse</u>	<u>Charentais</u>	1[ ]
<u>very sparse</u>		2[ ]
<u>sparse</u>		3[ ]
<u>sparse to medium</u>		4[ ]
<u>medium</u>	<u>Petit Gris de Rennes</u>	5[ ]
<u>medium to dense</u>		6[ ]
<u>dense</u>	<u>Piel de Sapo</u>	7[ ]
<u>dense to very dense</u>		8[ ]
<u>very dense</u>	<u>Albino</u>	9[ ]
<b>5.4 10</b> <b>(36)</b> <b>Fruit: density of patches</b>		
absent or very sparse	Rochet	1[ ]
<u>very sparse to sparse</u>		2[ ]
sparse		3[ ]
<u>sparse to medium</u>		4[ ]
medium	Braco	5[ ]
<u>medium to dense</u>		6[ ]
dense	Piel de Sapo	7[ ]
<u>dense to very dense</u>		8[ ]
very dense	Oranje Ananas	9[ ]
<b>5.5 11</b> <b>(38)</b> <b>Fruit: warts</b>		
absent	Piel de Sapo	1[ ]
present	Zatta	9[ ]

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:	
Characteristics	Example Varieties	Note	
<b>5.6 12</b> <b>(43)</b>	<b>Fruit: grooves</b>		
absent or very weakly expressed	Piel de Sapo, Arava	1[ ]	
weakly expressed	Total, Hobby	2[ ]	
strongly expressed	Védrantais, Galia	3[ ]	
<b>5.13</b> <b>(45)</b>	<b>Fruit: depth of grooves</b>		
<u>very shallow</u>	<u>Amber</u>	1[ ]	
<u>very shallow to shallow</u>		2[ ]	
<u>shallow</u>	<u>Galia</u>	3[ ]	
<u>shallow to medium</u>		4[ ]	
<u>medium</u>	<u>Alpha</u>	5[ ]	
<u>medium to deep</u>		6[ ]	
<u>deep</u>	<u>Panamá, Supermarket</u>	7[ ]	
<u>deep to very deep</u>		8[ ]	
<u>very deep</u>	<u>Noir des Carmes,</u> <u>Sucrin de Tours</u>	9[ ]	
<b>5.14</b> <b>(47)</b>	<b>Fruit: creasing of surface</b>		
<u>absent or very weak</u>	<u>Védrantais</u>	1[ ]	
<u>very weak to weak</u>		2[ ]	
<u>weak</u>	<u>Melchor, Sirocco</u>	3[ ]	
<u>weak to medium</u>		4[ ]	
<u>medium</u>	<u>Costa, Piolín</u>	5[ ]	
<u>medium to strong</u>		6[ ]	
<u>strong</u>	<u>Tendral Negro</u>	7[ ]	
<u>strong to very strong</u>		8[ ]	
<u>very strong</u>	<u>Balbey, Kirkagac</u>	9[ ]	
<b>5.7 15</b> <b>(48)</b>	<b>Fruit: cork formation</b>		
absent	Alpha	1[ ]	
present	Dalton	9[ ]	

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Characteristics	Example Varieties	Note
<b>5.16</b> <b>(49)</b> <b>Fruit: thickness of cork layer</b>		
<u>very thin</u>	<u>Amarillo Oro</u>	1[ ]
<u>very thin to thin</u>		2[ ]
<u>thin</u>	<u>Riosol, Védrantais</u>	3[ ]
<u>thin to medium</u>		4[ ]
<u>medium</u>	<u>Marina</u>	5[ ]
<u>medium to thick</u>		6[ ]
<u>thick</u>	<u>Geamar, PMR 45</u>	7[ ]
<u>thick to very thick</u>		8[ ]
<u>very thick</u>	<u>Honey Rock, Perlita</u>	9[ ]
<b>5.8 17</b> <b>(50)</b> <b>Fruit: pattern of cork formation</b>		
dots only	Hermes, Védrantais	1[ ]
dots and linear	Jivaro, Topper	2[ ]
linear only	Futuro, Riosol	3[ ]
linear and netted	Anatol, Chantal	4[ ]
netted only	Galia, Perlita	5[ ]
<b>5.9 18</b> <b>(51)</b> <b>Fruit: density of pattern of cork formation</b>		
very sparse	Alpha, Amarillo Oro	1[ ]
<u>very sparse to sparse</u>		2[ ]
sparse	Védrantais	3[ ]
<u>sparse to medium</u>		4[ ]
medium	Regal, Vital	5[ ]
<u>medium to dense</u>		6[ ]
dense	Galia, Geamar	7[ ]
<u>dense to very dense</u>		8[ ]
very dense	Honey Rock, Perlita	9[ ]

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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Characteristics	Example Varieties	Note
<b>5.10 19</b> <b>Fruit: main color of flesh</b> <b>(54)</b>		
white	Piel de Sapo	1[ ]
greenish white	Galia	2[ ]
green	Radical	3[ ]
yellowish white	Guaraní	4[ ]
orange	Védrantais	5[ ]
reddish orange	Magenta	6[ ]
<b>5.11 20</b> <b>Seed: length</b> <b>(60)</b>		
very short	Geumssaraki, Golden Crispi	1[ ]
<u>very short to short</u>		<u>2[ ]</u>
short	Elario, Katsura Giant	3[ ]
<u>short to medium</u>		<u>4[ ]</u>
medium	Arava, Sancho	5[ ]
<u>medium to long</u>		<u>6[ ]</u>
long	Amarillo Oro, Toledo	7[ ]
<u>long to very long</u>		<u>8[ ]</u>
very long	Albino	9[ ]
<b>5.21</b> <b>Seed: shape</b> <b>(62)</b>		
<u>not pine-nut shape</u>	<u>Toledo</u>	<u>1[ ]</u>
<u>pine-nut shape</u>	<u>Piel de Sapo</u>	<u>2[ ]</u>
<b>5.12 22</b> <b>Seed: color</b> <b>(63)</b>		
whitish	Amarillo Oro s.b.	1[ ]
cream yellow	Galia, Piel de Sapo	2[ ]



TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
Characteristics	Example Varieties	Note
<b>5.13 23</b> <b>(68)</b> <b>Shelf life of fruit</b>		
very short	Charentais	1[ ]
<u>very short to short</u>		2[ ]
short	Galia	3[ ]
<u>short to medium</u>		4[ ]
medium	Clipper	5[ ]
<u>medium to long</u>		6[ ]
long	Piel de Sapó	7[ ]
<u>long to very long</u>		8[ ]
very long	Tendral Negro	9[ ]
<b>5.14 24</b> <b>(69.1)</b> <b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom) – Race 0</b> <b>(Fom: 0)</b>		
absent	<u>Jaune Canari 2</u> <u>Atos, Charentais T</u>	1[ ]
present	<u>Cadence, Charentais Fom-2,</u> <u>Dibango, Jador, Jubilo, Karakal,</u> <u>Védrantais</u>	9[ ]
<b>5.15 25</b> <b>(69.2)</b> <b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom) - Race 1</b> <b>(Fom: 1)</b>		
absent	<u>Jaune Canari 2</u> <u>Atos, Charentais T, Védrantais</u>	1[ ]
present	<u>Arapaho, Jador, Rubbens</u> <u>Cadence, Charentais Fom-2,</u> <u>Dibango, Jubilo, Karakal</u>	9[ ]
<b>5.16 26</b> <b>(69.3)</b> <b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> (Fom) - Race 2</b> <b>(Fom: 2)</b>		
absent	<u>Arapaho, Jaune Canari 2,</u> <u>Rubbens-Atos,</u> <u>Charentais Fom-2, Charentais T,</u> <u>Dibango, Marianna</u>	1[ ]
present	<u>Anasta, Cléo, Jador, Cadence,</u> <u>Charentais Fom-1, Jubilo,</u> <u>Karakal, Perlita, Védrantais</u>	9[ ]
<b>5.27</b> <b>(69.4)</b> <b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>melonis</i> - Race 1.2 (Fom: 1.2)</b>		
<u>susceptible</u>	<u>Jaune Canari 2,</u> <u>Védrantais, Virgos</u>	4[ ]
<u>moderately resistant</u>	<u>Lunasol</u>	2[ ]
<u>highly resistant</u>	<u>Dinero, Isabelle</u>	3[ ]
<u>absent</u>	<u>Graffio, Prity, Virgos</u>	1[ ]
<u>present</u>	<u>Isabelle, Kyriel, Lunasol,</u> <u>Meliance, Piboule</u>	9[ ]
<u>not tested</u>		[ ]

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Characteristics	Example Varieties	Note
<b>5.28</b> <b>(70.1)</b> <b><u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) - Race 1 (Px: 1)</u></b>		
susceptible <u>absent or low</u>	Jaune Canari 2, Védrantais	1 [ ]
moderately resistant <u>medium</u>	Escrito	2 [ ]
highly resistant <u>high</u>	Anasta, Cézanne Arum	3 [ ]
not tested		[ ]
<b>5.29</b> <b>(70.2)</b> <b><u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) - Race 2 (Px: 2)</u></b>		
susceptible <u>absent or low</u>	Galoubet, Védrantais	1 [ ]
moderately resistant <u>medium</u>	Escrito, Pendragon	2 [ ]
highly resistant <u>high</u>	Anasta, Cézanne Arum	3 [ ]
not tested		[ ]
<b>5.30</b> <b>(70.3)</b> <b><u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) - Race 3 (Px: 3)</u></b>		
susceptible <u>absent or low</u>	Védrantais	1 [ ]
moderately resistant <u>medium</u>	Nettune Arago, Durango	2 [ ]
highly resistant <u>high</u>	Batista, Gédiva Arum	3 [ ]
not tested		[ ]
<b>5.31</b> <b>(70.4)</b> <b><u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) - Race 5 (Px: 5)</u></b>		
susceptible <u>absent or low</u>	Védrantais	1 [ ]
moderately resistant <u>medium</u>	Hugo, Pendragon Arago, Durango	2 [ ]
highly resistant <u>high</u>	Arapaho Arum	3 [ ]
not tested		[ ]
<b>5.32</b> <b>(70.5)</b> <b><u>Resistance to <i>Podosphaera xanthii</i> (Px) (ex <i>Sphaerotheca fuliginea</i>) (Powdery mildew) - Race 3-5 (Px: 3.5)</u></b>		
susceptible <u>absent or low</u>	Védrantais	1 [ ]
moderately resistant <u>medium</u>	Ciseo Arago, Durango	2 [ ]
highly resistant <u>high</u>	90625 Arum	3 [ ]
not tested		[ ]

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Characteristics	Example Varieties	Note	
<b>5.33</b> <b>(71)</b> <u>Resistance to <i>Golovinomyces cichoracearum</i> (<i>Erysiphe cichoracearum</i>) (Powdery mildew) Race 1 (Gc: 1)</u>			
susceptible	<u>Escrito, Score, Védrantais</u>	1	[ ]
moderately resistant	<u>Flores, Anasta</u>	2	[ ]
highly resistant	<u>Cézanne, Héliobel, Théo</u>	3	[ ]
not tested		[ ]	
<b>5.34</b> <b>(72)</b> <u>Resistance to colonization by <i>Aphis gossypii</i></u>			
absent	<u>Védrantais</u>	1	[ ]
present	<u>AR Hale's Best Jumbo, AR Top Mark, Godiva, Héliobel, Virgos</u>	9	[ ]
not tested		[ ]	
<b>5.35</b> <b>(73)</b> <u>Resistance to <i>Zucchini yellow mosaic virus</i> (ZYMV)</u>			
absent	<u>Cardillo, Générís, Jador, Védrantais</u>	1	[ ]
present	<u>Hannah's Choice, Lunaduke</u>	9	[ ]
not tested		[ ]	
<b>5.36</b> <b>(74.1)</b> <u>Resistance to <i>Papaya ringspot virus</i> (PRSV) - Guadeloupe strain</u>			
absent	<u>Védrantais</u>	1	[ ]
present	<u>Hannah's Choice</u>	9	[ ]
not tested		[ ]	
<b>5.37</b> <b>(74.2)</b> <u>Resistance to <i>Papaya ringspot virus</i> (PRSV) - E2 strain</u>			
absent	<u>Hannah's Choice, Védrantais</u>	1	[ ]
present	<u>WMR29</u>	9	[ ]
not tested		[ ]	
<b>5.38</b> <b>(75)</b> <u>Resistance to <i>Melon necrotic spot virus</i> (MNSV) Strain 0 (MNSV: 0)</u>			
absent	<u>Védrantais</u>	1	[ ]
present	<u>Cyro, Primal, Virgos, Yellow Fun</u>	9	[ ]
not tested		[ ]	

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Characteristics	Example Varieties	Note
<b><u>5.39</u></b> <b><u>(76)</u></b> <b><u>Resistance to <i>Cucumber mosaic virus</i> (CMV)</u></b>		
<u>absent</u>	<u>Cézanne, Dalton</u>	<u>1[ ]</u>
<u>present</u>	<u>Lunaduke, Virgos</u>	<u>9[ ]</u>
<u>not tested</u>		<u>[ ]</u>

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