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| International Union for the Protection of New Varieties of Plants |  |

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| Technical Working Party for VegetablesFifty-Fourth SessionBrasilia, Brazil, May 11 to 15, 2020 | TWV/54/5Original: EnglishDate: April 14, 2020 |

Partial revision of the Test Guidelines for Vegetable Marrow, Squash

Document prepared by experts from France

Disclaimer: this document does not represent UPOV policies or guidance

 The purpose of this document is to present a proposal for a partial revision of the Test Guidelines for Vegetable Marrow, Squash (document TG/119/4 Corr. 2).

 The Technical Working Party for Vegetables (TWV), at its fifty-third session, held in Seoul, Republic of Korea, from May 20 to May 24, 2019, agreed that the Test Guidelines for Vegetable Marrow, Squash (document TG/119/4 Corr. 2) be partially revised to add new Characteristics “Resistance to *Zucchini yellow mosaic virus*” and “Resistance to *Watermelon mosaic virus*” (see document TWV/53/14 Rev. “Report”, paragraph 91 and Annex III).

 The following changes are proposed:

1. Addition of new Characteristic 82 “Resistance to *Zucchini yellow mosaic virus* (ZYMV)” at the end of the Table of Characteristics
2. Addition of an explanation Ad. 82. “Resistance to *Zucchini yellow mosaic virus* (ZYMV)” in Chapter 8.2 “Explanations for individual characteristics”
3. Addition of new Characteristic Characteristic 83 “Resistance to *Watermelon mosaic virus* (WMV)” at the end of the Table of Characteristics
4. Addition of an explanation Ad. 83 “Resistance to *Watermelon mosaic virus* (WMV)” in Chapter 8.2 “Explanations for individual characteristics”

## Proposal to add new Characteristic 82 “Resistance to Zucchini yellow mosaic virus (ZYMV)” at the end of the Table of Characteristics

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| --- | --- | --- | --- | --- | --- | --- |
|  | English | français | Deutsch | español | Example VarietiesExemplesBeispielssortenVariedades ejemplo | Note/Nota |
| **82**. | Resistance to *Zucchini yellow mosaic virus* (ZYMV)  | **Resistance au *Zucchini yellow mosaic virus* (ZYMV)** | **Resistenz gegen *Zucchini yellow mosaic virus* (ZYMV)** | **Resistencia a *Zucchini yellow mosaic virus* (ZYMV)** |  |  |
|  | susceptible | sensible | anfällig | sensible | Cora | 1 |
|  | moderatly resistant | modérément résistante | mäßig resistent | moderadamente resistente | Mirza | 3 |
|  | resistant | résistante | resistent | resistente | Mikonos | 5 |

## Proposal to add explanation Ad. 82. “Resistance to *Zucchini yellow mosaic virus* (ZYMV)” in Chapter 8.2 “Explanations for individual characteristics”

Ad. 82: Resistance to *Zucchini yellow mosaic virus* (ZYMV)

|  |  |  |
| --- | --- | --- |
| 1. | Pathogen | *Zucchini yellow mosaic virus* (ZYMV) |
| 2. | Quarantine status | No |
| 3. | Host species | *Cucurbita pepo* L. |
| 4. | Source of inoculum | GEVES[[1]](#footnote-2) (FR) |
| 5. | Isolate | e.g. strain E9 |
| 6. | Establishment isolate identity | - |
| 7. | Establishment pathogenicity | Symptoms on susceptible squash variety |
| 8. | Multiplication inoculum |  |
| 8.1 | Multiplication medium | Living plant |
| 8.2 | Multiplication variety | e.g. Cora |
| 8.3 | Plant stage at inoculation | - |
| 8.4 | Inoculation medium | - |
| 8.5 | Inoculation method | - |
| 8.6 | Harvest of inoculum | - |
| 8.7 | Check of harvested inoculum | - |
| 8.8 | Shelflife/viability inoculum | - |
| 9. | Format of the test |  |
| 9.1 | Number of plants per genotype | At least 20 |
| 9.2 | Number of replicates | At least 2 |
| 9.3 | Control varieties | susceptible : Coramoderatly resistant: Mirzaresistant: MikonosUp to now, no complete resistance is identified. The two intermediate and resistant controls are necessary to validate the agressiveness of the test. |
| 9.4 | Test design | add non inoculated plants |
| 9.5 | Test facility | Climatic room or greenhouse |
| 9.6 | Temperature | e.g. 22°C or 24°C/18°C |
| 9.7 | Light | 12h-16h |
| 9.8 | Season |  |
| 9.9 | Special measures | - |
| 10. | Inoculation |  |
| 10.1 | Preparation inoculum | 1 g leaf with symptoms with 4 mL of PBS with carborundum (400 mg) and activated carbon (400 mg) or similar buffer, homogenize |
| 10.2 | Quantification inoculum | - |
| 10.3 | Plant stage at inoculation | First expanded leaf |
| 10.4 | Inoculation method | Rubbing with virus suspension |
| 10.5 | First observation | 14 days post-inoculation |
| 10.6 | Second observation | - |
| 10.7 | Final observations | 21 days post-inoculation |
| 11. | Observations |   |
| 11.1 | Method | Visual observation |
| 11.2 | Observation scale | Class 0: no symptomsClass 1: few chlorotic *patches**Class 2: many chlorotic patches**Class 3: large chlorotic areas (some patches on young leaves)**Class 4: mosaic and weak vein banding*Class 5: deformation and vein banding |
| 11.3 | Validation of test | Results should be compared with results of controls and are depending of the aggressiveness of the test and the distribution of the plants over the categories. The analysis of raw data of the couple ZYMV / Squash is planned in the Pathostat tool (free statistic analysis dedicated to quantitative disease resistances) [https://pathostat.geves.fr](https://pathostat.geves.fr/) |
| 11.4 | Off-types | - |
| 12. | Interpretation of data in terms of UPOV characteristic states | Classes 0, 1 are commonly judged as resistant – Note 5Classes 2, 3 are commonly judged as moderately resistant – Note 3Classes 4 and 5 are commonly judged as susceptible – Note 1 |
| 13. | Critical control points | Recommended dates of notation should be adapted depending on expression of symptoms on controls. Environmental conditions can have an effect on the expression of symptoms over time. In this case a second notation could be necessary.  |



## Proposal to add new Characteristic Characteristic 83 “Resistance to *Watermelon mosaic virus* (WMV)” at the end of the Table of Characteristics

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | English | français | Deutsch | español | Example VarietiesExemplesBeispielssortenVariedades ejemplo | Note/Nota |
| **83**. | Resistance to *Watermelon mosaic virus* (WMV) | **Resistance au *Watermelon mosaic virus* (WMV)** | **Resistenz gegen *Watermelon mosaic virus* (WMV)** | **Resistencia a *Watermelon mosaic virus* (WMV)** |  |  |
|  | susceptible | sensible | anfällig | sensible | Cora | 1 |
|  | moderatly resistant | modérément résistante | mäßig resistent | moderadamente resistente | Sofia | 3 |
|  | resistant | résistante | resistent | resistente | Mikonos, Syros | 4 |

## Proposal to add explanation Ad. 83 “Resistance to *Watermelon mosaic virus* (WMV)” in Chapter 8.2 “Explanations for individual characteristics”

Ad. 83: Resistance to *Watermelon mosaic virus* (WMV)

*Proposed new wording*

|  |  |  |
| --- | --- | --- |
| 1. | Pathogen | *Watermelon mosaic virus* (WMV) |
| 2. | Quarantine status | No |
| 3. | Host species | *Cucurbita pepo* L. |
| 4. | Source of inoculum | GEVES[[2]](#footnote-3) (FR) |
| 5. | Isolate | e.g. strain LL1A |
| 6. | Establishment isolate identity | - |
| 7. | Establishment pathogenicity | Symptoms on susceptible squash variety |
| 8. | Multiplication inoculum |  |
| 8.1 | Multiplication medium | Living plant |
| 8.2 | Multiplication variety | e.g. Cora |
| 8.3 | Plant stage at inoculation | - |
| 8.4 | Inoculation medium | - |
| 8.5 | Inoculation method | - |
| 8.6 | Harvest of inoculum | - |
| 8.7 | Check of harvested inoculum | - |
| 8.8 | Shelflife/viability inoculum | - |
| 9. | Format of the test |  |
| 9.1 | Number of plants per genotype | At least 20 |
| 9.2 | Number of replicates | At least 2 |
| 9.3 | Control varieties | susceptible: Coramoderately resistant: Sofia (moderate resistant control of lower level)moderately resistant: Mikonos or Syros (moderate resistant controls of higher level)Up to now, no complete resistance is identified. The two levels of intermediate resistant controls are necessary to validate the agressiveness of the test. |
| 9.4 | Test design | add non inoculated plants |
| 9.5 | Test facility | Climatic room or greenhouse |
| 9.6 | Temperature | e.g. 22°C or 24°C/18°C |
| 9.7 | Light | 12h-16h |
| 9.8 | Season |  |
| 9.9 | Special measures | - |
| 10. | Inoculation |  |
| 10.1 | Preparation inoculum | 1 g leaf with symptoms with 4mL of PBS with carborundum (400mg) and activated carbon (400mg) or similar buffer, homogenize |
| 10.2 | Quantification inoculum | - |
| 10.3 | Plant stage at inoculation | First expanded leave |
| 10.4 | Inoculation method | Rubbing with virus suspension |
| 10.5 | First observation | 14 days post-inoculation |
| 10.6 | Second observation | - |
| 10.7 | Final observations | 21 days post-inoculation |
| 11. | Observations |   |
| 11.1 | Method | Visual observation |
| 11.2 | Observation scale | Class 0: no symptomsClass 1: few chlorotic patchesClass 2: many chlorotic patchesClass 3: large chlorotic areas (some patches on young leaves)Class 4: mosaic, weak vein bandingClass 5: deformation and vein banding |
| 11.3 | Validation of test | Results should be compared with results of controls and are depending of the aggressiveness of the test and the distribution of the plants over the categories. *The analysis of raw data of the couple WMV / Squash is planned in the Pathostat tool ( free statistic analysis dedicated to quantitative disease resistances)* [*https://pathostat.geves.fr*](https://pathostat.geves.fr/) |
| 11.4 | Off-types | - |
| 12. | Interpretation of data in terms of UPOV characteristic states | Classes 0, 1, 2 are commonly judged as resistant – Note 4Classes 2, 3, 4 are commonly judged as intermediate resistant – Note 3Classes 4 and 5 are commonly judged as susceptible – Note 1 |
| 13. | Critical control points | Recommended dates of notation should be adapted depending on expression of symptoms on controls. Environmental conditions can have an effect on the expression of symptoms over time. In this case a second notation could be necessary. |



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1. matref@geves.fr [↑](#footnote-ref-2)
2. matref@geves.fr [↑](#footnote-ref-3)