10.1.c Wob juncto 63.2.a Vo 1107/2009 juncto 39.2.a Vo 178/200

Comparison of infectivity efficiency after high-pressure spraying of V10 with carborundum and with synthetic sand

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Document Infectivity data of V10 high-pressure spraying with carborundum and synthetic sand, 10 April 2017

Period 2015

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Introduction

V10 is a microbiological crop protection product against virulent variants of Pepino mosaic virus (PepMV). V10 contains a mixture of two active substances, VX1 and VC1. Both are mild isolates of PepMV. After inoculation of young tomato plants with V10 and multiplication of VX1 and VC1 during two weeks, plants are protected against virulent variants of PepMV. V10 can be applied in the spraying solution in combination with carborundum as abrasive to facilitate virus infection in the leaves by mechanical inoculation. In this study the efficiency of V10 to infect plants with carborundum is compared with V10 and using synthetic sand as abrasive.

Aim

Comparison of infectivity efficiency after high-pressure spraying of V10 with carborundum and with synthetic sand.

Trial site and period

The trials were conducted in greenhouses in the municipality Westland, the Netherlands in 2015.

Treatments

Young tomato plants were inoculated with V10 by high-pressure spraying using carborundum or synthetic sand as abrasive to facilitate infection of the plants. This was performed in week 18, 20 and 28 of 2015.
After 2 or 3 weeks the percentages of infected plants were determined by ELISA (SPV A517, GAC). Real-time PCR was used to determine the presence of VX1 and VC1 (SPV A518, GAC)

Materials:

- Two batches of V10 were used, named V10N20150210 and V10N20150602. Batches were isolated from tobacco plants without further purification step (high nicotine content). V10 contains the mild PepMV isolate VC1 and the mild PepMV isolate VX1. Both batches contain phoshate as buffer.
- As abrasives were used carborundum (Silicon carbide, particle size 17 µm; article number F400/17, Saint-Gobain) or synthetic sand (Aluminium oxide, particle size 54-73 µm; article number 08.01.09, Eijkelkamp Soil & Water).
- The used young tomato plants were three to four weeks old after sowing and between 10 and 30 cm high. The used cultivars were [redacted]. In total eight groups of approximately 6500 plants were used for inoculaton.

Used methods

Groups of 6500 young tomato were inoculated by high-pressure spraying using carborundum or synthetic sand as abrasive.

Used inoculation protocol:

- Apply onto young tomato plants (10-30 cm in height), before planting. The plants should be free of pepino mosaic virus at the time of application.
- Shake product well before use within the container.
- Prepare 2% dilution of the product (1 L V10 per 50 litres of water) with cold water (approximately 8 °C) and add 8 grams of carborundum or synthetic sand per litre of water. Mix thoroughly.
- Apply 0.5 L spray liquid per m^2 of plant bed.
- Boom height should be approximately 10-15 cm above the crop. Diluted product should be sprayed at 12-15 bar (measured at spray boom).

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Assessments

To determine the percentage of infected plants, 10 plants of each group were tested after two or three weeks by ELISA whether they were infected with PepMV (SPV A517). According to the Quality Control of the operating procedure for the preparation of VX1 and VC batches (Valto SOP1), the percentage of plants that are infected should be at least 70%. To determine the presence of VX1 and VC1 in the infected plants, real-time PCR was used (SPV A518).

Results

The four groups of tomato plants which were inoculated with V10 and carborundum were all infected for 100% (Table 1). The four groups of tomato plants which were inoculated with V10 and synthetic sand were infected for 100%, 100%, 80% and 80%, respectively (Table 2). All eight groups were infected with VX1 as well as with VC1 (Table 1 and 2).

Table 1. Percentage of infected plants and the presence of VX1 and VC1 after inoculation of young tomato plants with V10 by high-pressure spraying with carborundum.

Groups of tomato plants	Cultivar	V10 batch	Week of inoculation	Infectivity (%)	VX1 detected	VC1 detected
1	[redacted]	V10N20150210	Wk 18, 2015	100	yes	yes
2		V10N20150210	Wk 18, 2015	100	yes	yes
3		V10N20150210	Wk 20, 2015	100	yes	yes
4		V10N20150602	Wk 28, 2015	100	yes	yes

Table 2. Percentage of infected plants and the presence of VX1 and VC1 after inoculation of young tomato plants with V10 by high-pressure spraying with synthetic sand.

Groups of tomato plants	Cultivar	V10 batch	Week of inoculation	Infectivity (%)	VX1 detected	VC1 detected
1	[redacted]	V10N20150210	Wk 18, 2015	100	yes	yes
2		V10N20150210	Wk 18, 2015	100	yes	yes
3		V10N20150210	Wk 20, 2015	80	yes	yes
4		V10N20150602	Wk 28, 2015	80	yes	yes

Statistics

Table 3. Averages and standard deviation (SD) of infectivity efficiencies after high-pressure spraying of V10 with carborundum or synthetic sand.

Used abrasive	Average Infectivity (%)	SD (%)
Carborundum	100	0
Synthetic sand	90	12

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Conclusion

All four groups of plants sprayed with carborundum and all four groups of plants sprayed with synthetic sand meet the quality control requirement of an infectivity of at least 70%.

Discussion

Not all groups were infected for 100%, because two of the four groups with synthetic sand were infected for 80%. By experience with previous inoculations and also by general knowledge about mechanically transmitted viruses, it is known that within two weeks further all tomato plants will be infected by crop handling if the virus is present in a large portion of the plants in a greenhouse.

References:

SPV A517: Protocol for the qualitative determination of PepMV by DAS-ELISA (Groen Agro Control).
SPV A518: Protocol for the determination of indices of VC1, VX1 and PepMV by qRT-PCR (Groen Agro Control).
SOP1 Valto: Operating procedure for the preparation of VX1 and VC1 batches (Valto).