REVISTA BRASILEIRA DE FRUTICULTURA
Published by the Brazilian Society of Fruit Crops
ISSN 0100-2945

Biotechnology - Original Article - Edited by: Alexandre Pio Viana

Doses and intervals of application of potassium phosphite for the control of passion fruit scab

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Abstract – Passion fruit scab (PFS), which is caused by species of the fungal complex *Cladosporium cladosporioides*, occurs both in protected environments and in the field. In view of the importance of the passion fruit scab, this study aimed to evaluate the effect of different intervals and doses of potassium phosphite in the greenhouse and in the field on the incidence of the disease. To evaluate the best dose of potassium phosphite, three experiments were carried out, two in a greenhouse and one in the field. The best application interval was evaluated in another experiment in a greenhouse. The variables evaluated were the incidence of plants with PFS symptoms (%) and the incidence of defoliated plants (%) in all experiments. The harvested fruits were evaluated for their mass (g), diameter (cm), soluble solids, acidity and chlorophyll content. The present work allowed to conclude that the dose of 0.1% of potassium phosphite was the best to control the passion fruit scab in a greenhouse, the treatment with an interval of 28 days of application showed a lower incidence of scab in seedlings at 35 days after inoculation and that potassium phosphite did not interfere in the quality parameters of fruits and leaves of passion fruit evaluated.

Index terms: *Passiflora edulis*, *Cladosporium cladosporioides*, seedlings.

Doses e intervalos de aplicação de fosfito de potássio para o controle da verrugose do maracujazeiro-azedo

Resumo – A verrugose do maracujazeiro-azedo (*Passiflora edulis*), causada por espécies do complexo fúngico *Cladosporium cladosporioides* (CCSC), é uma das doenças mais importantes da cultura, ocorrendo tanto em ambiente protegido quanto em campo. A doença é típica de tecidos jovens de maracujazeiro,;entretanto, ela se manifesta em todo o ciclo da cultura. O efeito das substâncias genericamente chamadas de fosfitos, sobre diversas doenças de plantas, tem sido amplamente estu-

Rev. Bras. Frutic., v.45, e-300 DOI: https://dx.doi.org/10.1590/0100-29452023300
Received 15 Sep, 2022 • Accepted 27 Feb, 2023 • Published Jul/Aug, 2023. Jaboticabal - SP - Brazil.

dado. Tendo em vista a importância da verrugose para o maracujazeiro-azedo, este trabalho teve por objetivo avaliar o efeito de diferentes intervalos e doses de fosfito de potássio em casa de vegetação e em campo, sobre a incidência da doença. Para avaliar a melhor dose de fosfito de ,oram realizados três experimentos, dois em casa de vegetação e um a campo. O melhor intervalo de aplicação foi avaliado em outro experimento, em casa de vegetação. As variáveis avaliadas foram à incidência de plantas com sintomas de verrugose (%) e a incidência de plantas desfolhadas (%) em todos os experimentos. Os frutos colhidos foram avaliados quanto à sua massa (g), diâmetro (cm), sólidos solúveis, acidez e teor de clorofila (A, B e total). O presente trabalho permitiu concluir que a dose de 0,1% de fosfito de potássio foi a melhor para controlar a verrugose do maracujazeiro-azedo em casa de vegetação; que o tratamento com intervalo de 28 dias de aplicação apresentou a menor incidência de plântulas com verrugose, aos 35 dias após a inoculação e que o fosfito de potássio não interferiu nos parâmetros de qualidade dos frutos e folhas de maracujazeiro-azedo avaliados.

Termos para indexação: *Passiflora edulis*, *Cladosporium cladosporioides*, mudas.

Introduction

Passion fruit (*Passiflora edulis*) scab (PFS), which is caused by species of the fungal complex *Cladosporium cladosporioides* (CCSC) (ROSADO et al., 2019), is one of the most important diseases of the crop. It occurs both in protected environment and in the field. In seedling production nurseries, the disease cause great losses, as it delays the plant development due to the decrease in the emission of new leaves (PERUCH; SCHROEDER, 2018). The disease is typical of young passion fruit tissues (GÓES, 1998; PERUCH et al., 2009), however it manifests itself throughout the crop cycle. In seedlings, it affects cotyledon leaves, true leaves and the stem. On the leaves, it exhibit circular lesions, which begin as translucent and increase in size, acquiring a dry appearance and brown color. After the latency period of the fungus, a grayish mass appears in the center of the lesion, which are its spores. The spots may increase in size and damage a large part of the leaf area, or different lesions may form on the same leaf and coalesce. Even in the first month of development, the grayish growth of the fungus on the stem is observed, causing the apex to bend. The high incidence of the disease in the first 30 days of age cause intense defoliation and the death of the plants, making the production of seedlings unfeasible.

The control of scab in a protected environment can be carried out mainly using fungicides, however, resistant *Cladosporium* spp. populations can increase, when the products are not correctly applied. In the field, chemical control is also the main form of controlling the disease, which can appear on leaves, flower buds and fruits. In the latter, as cortical lesions, similar to warts. Such symptoms reduce its commercial value, and may lead to abortion (PERUCH; SCHROEDER, 2018).

The effect of substances generically called phosphites, to refer to phosphoric acid salts (H_3PO_3), on various plant diseases has been widely studied. Phosphites have been shown to be effective for disease control in some crops, such as fruit and vegetables, among others (SILVA, et al., 2011). For the apple crop, for example, the use of potassium phosphite has shown a high rate of control of apple scab (*Venturia inaequalis*) on leaves and fruits (BONETI; KATSURAYAMA, 2015). These substances are known to be foliar fertilizers that have an additional antifungal action, being interesting because they have less toxicity than traditionally used fungicides (SANHUEZA, 2018). For the passion fruit, Junqueira et al. (2011) verified a reduction in the severity of warts in a treatment with potassium phosphite in the field. Due the importance of PFS for passion fruit, this work aimed to evaluate the effect of different intervals and doses of potassium phosphite in the greenhouse and in the field on the incidence of the disease.

Material and Methods

Obtaining *the inoculum and inoculation*

The inoculum was obtained by direct isolation from the leaves of 'SCS Catarina' sour passion fruit seedlings with scab symptoms. The fungus spores were transferred to PDA (Potato-Dextrose-Agar) culture medium, which was placed in a B.O.D type incubator at a temperature of 25°C±2 for 14 days. From the first isolation, the fungus was maintained in the laboratory from successive subcultures to the PDA culture medium.

Colonies with 14 days of age were used for the inoculation of passion fruit seedlings. To produce the inoculum, 20 mL of sterilized distilled water (miliQ) was added to the colonies and a glass slide was used to scrape the surface of the colony. The spore count of the resulting suspension was performed in a Neubauer chamber with the aid of an optical microscope. The spore suspension was calibrated to 6 x 10^7 spores/ml.

Different doses of potassium phosphite for the control of PFS in the greenhouse and in the field

Two experiments were carried out in a greenhouse, which comprised 5 treatments and three replications. The treatments were as follows: untreated control, potassium phosphite at 0.10%, 0.20%, 0.40% and 0.60%. In Experiment 1, the treatment 0.80% was added. The experimental unit consisted of a tray containing 120 'SCS Catarina' sour passion fruit seedlings.

In both experiments, the trays were kept in a greenhouse with natural CCSC infestation and two sprays of potassium phosphite were carried out, one at 45 and another at 60 DAS (days after sowing). In Experiment 1, the plants were evaluated at 7 DASA (days after the second application), at 14 DASA and at 21 DASA. In Experiment 2, the plants were evaluated at 7 DAFA (days after the first application), at 14 DAFA and at 21 DAFA. The variables analyzed were: incidence of plants with symptoms (%) and incidence of defoliated plants (%). In Experiment 3, the most effective dose of potassium phosphite for PFS control was evaluated under field conditions. The treatments were the same as in those tests carried out in a greenhouse. The experimental plot consisted of rows with four passion fruit plants. The incidence of the disease was obtained through natural infection. The analyzed variables were number of spotted leaves, number of scab lesions, number of lesions per leaf, incidence of infected plants (%) and incidence of defoliated plants (%). The harvested fruits were evaluated for their mass (g), diameter (cm), soluble solids, acidity and chlorophyll content (A, B and total).

Different application intervals of potassium phosphite for PFS control

In experiment 4, the most effective potassium phosphite application intervals at a concentration of 0.10% were evaluated in a greenhouse. The experiment consisted of 5 treatments: 7, 14, 21 and 28 days of interval between applications and the non-inoculated control, and 4 replications. The experimental plot consisted of a tray with 120 'SCS Catarina' sour passion fruit seedlings. Three days after potassium phosphite application, all experimental plots were artificially inoculated with spores of the CCSC isolate. Artificial inoculation was chosen in this experiment because the incidence of the disease was low in the greenhouse. The variables analyzed were the incidence of plants with scab symptoms and the incidence of defoliated plants. Both calculated as a percentage of the total number of plants per plot.

Experimental design and data analysis

The experimental design of the experiments was completely randomized. The variables obtained were subjected to analysis of variance (ANOVA) followed by Tukey's test at 5% probability, with the aid of the statistical program R (R DEVELOPMENT CORE TEAM, 2022).

Results and Discussion

In Experiment 1, the dose of 0.10% provided the lowest incidence of plants with symptoms of scab, with significantly smaller differences than in the untreated control, at 14 and 21 DASA (Figure 1). The 0.10% dose in Experiment 2 also led to lower incidences of PFS in two evaluation times, but earlier, at 7 and 14 DAFA (Figure 2). Similar to what happened in Experiment 1 at 14 DAFA, the dose of 0.10% in Experiment 2 led to a significantly lower incidence of defoliated plants than the control, in the evaluation carried out at 21 DAFA (Figure 3). Experiment 1 showed no significant difference between the doses at 7 DASA, regarding the incidence of PFS. For Experiment 2, the dose of 0.10% did not show difference in relation to the control regarding the incidence of PFS at 21 DAFA, however the dose of 0.20% was the only one that showed difference in relation to the control, at this time of evaluation.

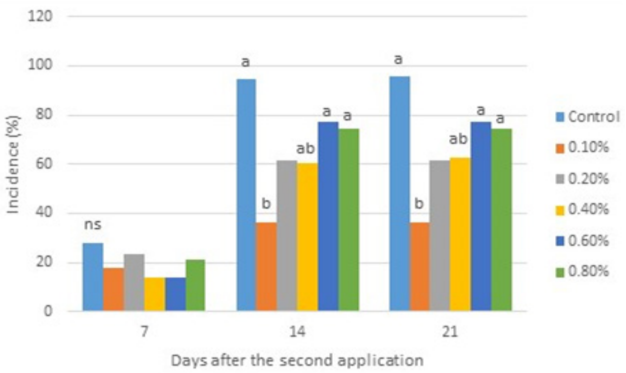

Figure 1 - Incidence (%) of plants with PFS symptoms at 7, 14 and 21 days after the second application of different doses of potassium phosphite. ns – not significant. Means followed by the same letter in each evaluation period do not differ according to the Tukey Test at 5% significance.

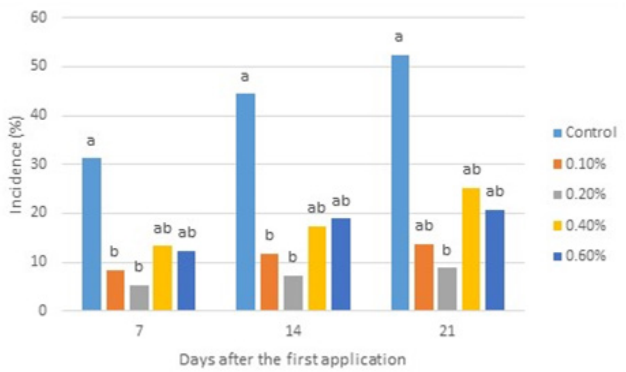

Figure 2 - Incidence (%) of plants with PFS symptoms at 7, 14 and 21 days after the first application of different doses of potassium phosphite. Means followed by the same letter in each evaluation period do not differ according to the Tukey Test at 5% significance.

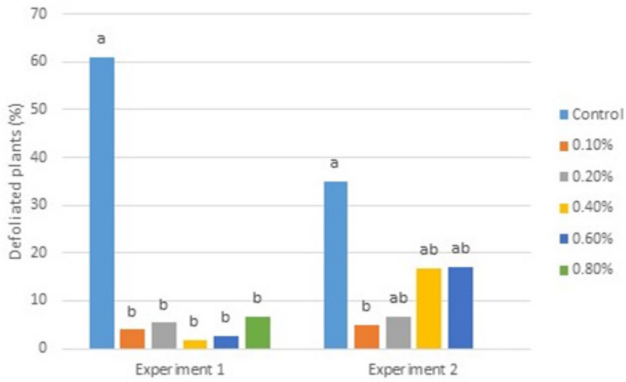

Figure 3 – Defoliated plants (%) in Experiment 1 and Experiment 2 at 14 days after the second application and at 21 days after the first application of different doses of potassium phosphite, respectively. Means followed by the same letter in each experiment do not differ according to the Tukey Test at 5% significance.

Such results demonstrate that the dose of 0.10% of potassium phosphite is the most effective for the control of scab in young passion fruit seedlings under greenhouse conditions. The fact that the effect was verified earlier in Experiment 2 can be explained by a lower CCSC inoculum pressure, when compared to Experiment 1, in which the disease showed an incidence close to 100% at 21 DASA and an incidence of plants defoliated above 60% in the control, compared to 52.45% and 35.02% for the two variables, respectively, in Experiment 2.

The 0.10% dose is already recommended by the manufacturer for fruit trees in general as fertilizer. Knowledge about its effect on PFS will allow management strategies to be established in order to determine the ideal moment for its application, which the results of this work suggest should be done at the beginning of the appearance of the first symptoms.

In Experiment 3, no statistical difference was observed between treatments regarding the variables number of spotted leaves (NSL), number of lesions (NL), number of lesions per leaf (NL/leaf) and incidence of infected plants (INC) (Tables 1 , 2 and 3), except for the evaluation 7 days after the fourth application, in which the evaluation of the number of lesions per leaf was higher for the concentration of

0.60%, however this result was due to a lower incidence of diseased leaves in this treatment (Table 2). Despite this result, the NL in untreated plants was higher in most of the evaluations carried out. Statistical differences were not found for fruit mass (FM), largest fruit diameter (LFD), smallest fruit diameter (SFD), soluble solids (SS), acidity (AC) and chlorophyll (A, B and total) (Table 4), however, the higher incidence of the disease in untreated plants suggests that the greater weight of damage is related to the visual quality of the fruit due to the presence of greater amounts of fungal inoculum in these plants.

Table 1 - Number of spotted leaves (NSL), number of lesions (NL), number of lesions per leaf (NL/F) and incidence of infected plants (INC), in percentage, as a function of potassium phosphite dose (D%).

D%	14 DAFA				21 DAFA				35 DAFA			
	NSL	NL	NL/F	INC	NSL	NL	NL/F	INC	NSL	NL	NL/F	INC
Test	1.25^{ns}	13.5	6	18.75	0.75	9.25	3	6.25	16.75	128	3	50
0.10	0.75	8.5	3	6.25	1.5	7.25	1	6.25	16	52.5	3	68.75
0.20	1.75	8.75	2	18.75	0.5	1.5	2	12.5	9.25	27.25	2	56.25
0.40	0.75	4.25	2	12.5	0.5	5.75	3	6.25	4.75	54.25	8	43.75
0.60	0.5	4.50	5	12.5	0	0	0	0	13.25	46.5	2	68.75

DAFA = Days after the first application. ^{ns} – not significant, in the column, according to the Tukey Test at 5% significance.

Table 2 - Number of spotted leaves (NSL), number of lesions (NL), number of lesions per leaf (NL/F) and incidence of infected plants (INC), in percentage, as a function of potassium phosphite dose (D %).

D(%)	21 DATA				7 DAFOA			
	NSL	NL	NL/F	INC	NSL	NL	NL/F	INC
Test	9^{ns}	42.75	3	50	8	37.75	3b	69
0.10	3.5	10	2	38	8	41.25	3b	50
0.20	3.75	23	4	38	4.25	19.5	3b	25
0.40	2	9.25	2	31	1.5	6.25	4b	38
0.60	5	25	4	63	2.25	40	19a	56

DATA = Days after the third application; DAFOA = Days after the fourth application. ^{ns} – not significant. Means followed by the same letter, in the column, do not differ according to the Tukey Test at 5% significance.

Table 3 - Number of spotted leaves (NSL), number of lesions (NL), number of lesions per leaf (NL/F) and incidence of infected plants (INC), in percentage, as a function of potassium phosphite dose (D %).

D%	14 DAFOA				21 DAFOA			
	NSL	NL	NL/F	INC	NSL	NL	NL/F	INC
Test	21^{ns}	114	4	69	20	113	3	100
0.10	10	36	3	63	8	24	3	88
0.20	6	24	4	50	7	24	3	69
0.40	4	14	3	44	4	9	3	50
0.60	5	17	3	50	3	6	2	63

DAFOA = Days after the fourth application. ^{ns} – not significant, in the column, according to the Tukey Test at 5% significance.

Table 4 - Fruit mass (FM), largest fruit diameter (LFD), smallest fruit diameter (SFD), soluble solids (SS), acidity (AC) and chlorophyll (A, B and total), depending on the dose of potassium phosphite (D%).

D%	FM (g)	LFD (cm)	SFD (cm)	SS	AC	Cl A	Cl B	Cl Tot
Test	380.84^{ns}	11.67	9.40	11.5	4.29	43.9	14.32	58.23
0.10	330.76	11.1	9.22	11.4	4.75	42.89	13.56	56.45
0.20	348.70	11.50	9.54	12.1	4.75	39.65	11.02	50.66
0.40	350.19	11.56	9.39	9.91	4.99	39.09	11.51	50.60
0.60	351.35	11.39	9.55	11.4	4.83	38.59	10.67	49.26

^{ns} – not significant, in the column, according to the Tukey Test at 5% significance.

The results of Experiment 4 show that there was no significant difference between the 0.10% potassium phosphite application intervals up to 28 days after inoculation (DAI), both for the variable of incidence of seedlings with scab symptoms and for defoliated seedlings. However, at 35 DAI, the treatment with an interval of 28 days of application had the lowest incidence of seedlings with scabs (Table 5). CCSC inoculum pressure present in the area, the occurrence of optimal environmental conditions for the development of the fungus and the time of product application may have interfered with the effect of the product at other evaluation times. The incidence of defoliated seedlings was lower in all treatments with phosphite when compared to the control, however, no statistical difference was observed between them.

Table 5 - Incidence of passion fruit seedlings with PFS symptoms (INC) and defoliated seedlings (DEF), in percentage, treated with 0.1% potassium phosphite at different application intervals (INT): 7, 14, 21 , and 28 days, evaluated at 7, 14, 21, 28 and 35 days after inoculation (DAI).

INT	7 DAI		14 DAI		21 DAI		28 DAI		35 DAI	
	INC	DES	INC	DES	INC	DES	INC	DES	INC	DES
Test	37.15^{ns}	16.3	48.1	35.21	51.24	37.94	62.73	57.24	56.31ab	41.9
7	27.34	9.98	37.05	27.8	32.83	21.52	54.46	39.00	50.63ab	26.43
14	26.87	8.8	36.1	24.95	32.34	21.4	64.04	48.16	62.75a	40.4
21	26.28	12.84	29.12	25.83	33.92	29.17	44.07	39.94	41.82ab	24.18
28	19.08	8.03	25.84	14.5	28.26	16.02	40.18	33.59	36.03b	25.95

^{ns} – not significant, in the column, according to the Tukey Test at 5% significance.

Difficulties in identifying yellow passion fruit genotypes and related species resistant to CCSC have been frequently observed in the literature. Oliveira et al. (2013) found that all tested accessions belonging to three passion fruit species (*P. edulis*, *P. alata* and *P. cincinnata*) were classified as moderately resistant or susceptible. Only four *P. setacea* accessions were considered immune or resistant. Due to the difficulty in obtaining genotypes with resistance to CCSC and the environmental and fungicide resistance risks, the evaluation of alternative measures for managing the disease is justified. The use of biostimulant substances has intensified in recent years due to the demand for agriculture with less environmental impact. Biostimulant substances can be defined as a technology that allows the plant to be able to overcome biotic or abiotic stresses that negatively impact productivity (SANTOS-JIMENEZ et al., 2022). Several biostimulant substances have shown promising results for the management of PFS. Humic acids, for example, may have an effect on the activation of genes related to passion fruit defense, in addition to other substances such as fungal cell wall glycoproteins (galactomannan peptide – pGM) (SANTOS-JIMENEZ et al., 2022) and acibenzolar (WILLINGHAM et al., 2002), these specifically on PFS. The use of potassium phosphites, in turn, appears as an option, given its efficacy reported in the literature for scab and other passion fruit diseases (JUNQUEIRA et al., 2011) and the results obtained in the present study.

Conclusions

The best dose to control scab on passion fruit was 0.10% in a greenhouse.

Potassium phosphite did not interfere with the quality parameters of the fruits and leaves of passion fruit evaluated.

In the present work, it was possible to observe that the treatment with potassium phosphite in the 28-day interval of application presented the lowest incidence of seedlings with scabs at 35days after inoculation.

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