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PLANT PROTECTION
Short Communication

Phosphite for the root rot (*Rhizoctonia solani*) management in common bean and compatibility with *Rhizobium tropici*

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ABSTRACT: Phosphites can be considered an additional strategy to be included in disease management programs. Therefore, this study aims to evaluate the application of potassium phosphite (KPhi) in common bean seeds, in the reduction of intensity of root rot (*Rhizoctonia solani*) and in the compatibility with *Rhizobium tropici*, a nitrogen-fixing bacterium. The emergence speed index (ESI) and the percentage of total emergence were evaluated in seeds inoculated with *R. solani* and treated with KPhi. The number of nodules per root system, dry nodule mass, quantified the compatibility Phi x R. tropici, dry mass, nitrogen content, and nitrogen accumulation in the shoots. KPhi promoted a higher ESI and increase in plant stand compared to the control treatment. The number of nodules formed and the nitrogen content and accumulation in the KPhi treatments were similar to control (seeds inoculated with *R. tropici*). Thus, the treatment of seeds with KPhi provides control of *R. solani* and is compatible with *R. tropici*.

Key words: Phaseolus vulgaris L., emergence speed index, seeds.

INTRODUCTION

The common bean crop (*Phaseolus vulgaris* L.) is subject to attack by several pathogens that can be transmitted by seeds (Mahmoud et al. 2013). Root rot and plant tipping are diseases caused by the fungus *Rhizoctonia solani* Kuhn that result in damage to bean plants (Carvalho et al. 2011). The fungus causes lesions in the basal part of the hypocotyl and in the main root of the seedlings, causing decreased stand.

In many areas where bean cultivation is planted every year, it is necessary to treat seeds with fungicides to prevent diseases that affect the crop. However, the environmental impact of these products must be highlighted in addition to the risk of incompatibility between fungicides and nitrogen-fixing bacteria of *Rhizobium genera* (Araújo and Araújo 2006; Barbosa and Gonzaga 2012).

The use of phosphite can be considered an additional strategy to be included in disease management programs, to reduce the intensive use of fungicides (Lobato et al. 2008) and has been investigated mainly in the control of common bean diseases as white mold and anthracnose (Deliopoulos et al. 2010; Fagundes-Nacarath et al. 2018; Gadaga et al. 2017). According Carmona et al. (2018), phosphites can a feasible alternative to fungicide seed treatments against *Pythium* that causes damping-off in soybean. Phosphites exert a complex mode of action against fungi and oomycetes, which involves direct action, such as by inhibiting sporulation or reducing the growth rate, and indirect effects, such as rapid and strong stimulation of plant defense mechanisms, both in the presence and absence of the pathogen (Deliopoulos et al. 2010).

Thus, this study aims to evaluate the effects of potassium phosphite (KPhi), applied to common bean seeds on the control *R. solani* and its compatibility with *Rhizobium tropici*, a nitrogen-fixing bacterium.

METHODS

The assay was conducted in a greenhouse. Seeds of the commom bean cultivar Pérola were disinfected with 1% sodium hypochlorite for 30 seconds, washed with sterile distilled water, and then kept in a laminar flow hood until dry. The seeds were inoculated with *R. solani* by the water restriction method, described by Machado et al. (2012).

The fungus was obtained from infected seeds and isolated in 9 cm diameter Petri dishes, containing potato-dextrose-agar (PDA) culture medium. After 3 days of growth, mycelial discs were transferred to a 9 cm Petri dish containing PDA medium with mannitol added (71.3 $g \cdot L^{-1}$ PDA) and with the water potential adjusted to -1.0 MPa, using SPPM software (Micheland Radcliffe, 1995). The colonies were incubated at 21 ± 2 °C with a 12 hour photoperiod for 3 days. The seeds were deposited on the colonies and remained in contact with the fungus for 48 hours.

The treatments evaluated were: KPhi P_2O_5 33.6% w/w and K_2O 29% w/w, at a dose of 5.0 $mL \cdot kg^{-1}$ seeds; fungicide (phenylurea - 25% w/v) at a dose of 3.0 $mL \cdot kg^{-1}$ seeds; untreated and inoculated seeds (inoculated control), and untreated and uninoculated seeds (uninoculated control). After treatment, the seeds were placed in a laminar flow hood for 2 hours. The seeds were deposited in 48 × 29 × 10 cm polyethylene boxes containing sterile sand. A randomized block experimental design was used with four replicates, in which each plot consisted of a box with 50 seeds. The number of emerged seedlings was counted daily to calculate the emergence speed index (ESI), as proposed by Maguire (1962). The percentage of total emergence of plants in each box, was calculated on the 14th day after sowing. The statistical analysis was performed using Sisvar software version 5.1 (Ferreira 2011).

The experiment to verify the compatibility of KPhi application with the nitrogen-fixing bacterium *R. tropici* was performed in a greenhouse. Seeds of the common bean cultivar Pérola were disinfected with 1% sodium hypochlorite for 30 seconds and six successive washes were performed in sterile distilled water.

The treatments tested were: 1: inoculant (*R. tropici*–based peat inoculum containing the strains SEMIA 4077 and SEMIA 4088, with a guarantee of 3.0×10^9 CFU g^{-1}) at a dose of 2 $g \cdot kg^{-1}$ seeds; 2: KPhi (P_2O_5 33.6% w/w and K_2O 29% w/w) at a dose of 5.0 $mL \cdot kg^{-1}$ seeds + inoculant (2 $g \cdot kg^{-1}$); 3: fungicide (carboxanilide at 20% w/v and dimethyldithiocarbamate at 20% w/v) at a dose of 2.5 $mL \cdot kg^{-1}$ seeds + inoculant (2 $g \cdot kg^{-1}$); 4: common bean seeds non-inoculated with *R. tropici* but receiving nitrogen fertilization (350 $mg \cdot plant^{-1}$, ammonium nitrate source); 5: common bean seeds non-inoculated with *R. tropici* and without nitrogen fertilization. The seeds were sown in pots (2.5 dm3) containing substrate.

The experiment was conducted in a randomized block design, with four repetitions and an experimental plot consisting of a pot containing two plants. The pots were periodically irrigated with the nutrient solution, proposed by Hoagland and Arnon (1950) with nitrogen restriction. Approximately 50 days after sowing, at the beginning of flowering (stage R5), the number of nodules per root system was counted, and the dry mass of nodules, the dry shoot weight, and the nitrogen content and accumulation were evaluated in the shoots. The statistical analyses were performed in Sisvar software version 5.1 (Ferreira 2011).

RESULTS AND DISCUSSION

The fungicide and KPhi treatments had 98.5% and 96.0% emerged plants, respectively, which were similar to each other (Fig. 1). In the inoculated control treatment, the percentage of emerged plants was 68.5%. Compared to that of the inoculated control, the plant stand provided by the seed treatment with phenylurea and with KPhi were 43.8% and 40.1% higher, respectively. The fungicide treatment yielded a significantly higher ESI than the other treatments (Fig. 1). The treatment with KPhi was similar to the non-inoculated control and provided a higher ESI than the inoculated control.

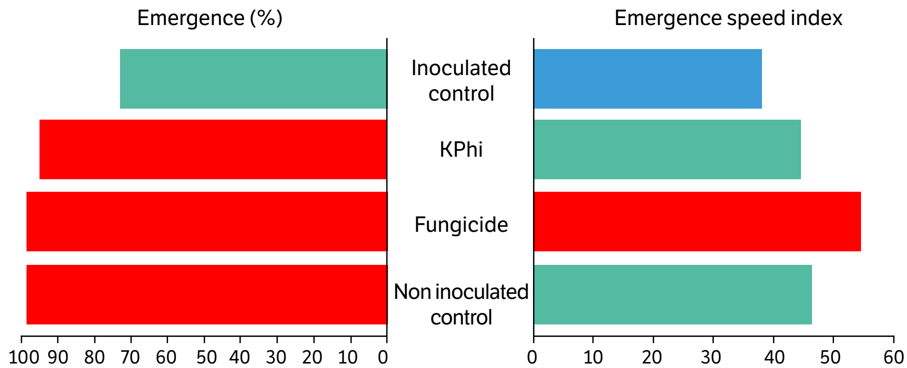

Figure 1. Effect of potassium phosphite (KPhi) on the emergence percentage and on the emergence speed index (ESI) of common bean seedlings. Bars followed by the same color do not differ statistically from each other by the Scott-Knott test (p≤0.05).

Source: Elaborated by the authors.

Maintenance of plant stand is a fundamental condition to achieve the desired productivity of the common bean crop. The main consequences of *R. solani* infection include reduced stand and seedling vigor, with consequent loss of productivity (Souza et al. 2009). The KPhi treatment led to a lower *R. solani* intensity in inoculated common bean seeds, resulting in greater plant emergence. KPhi was associated with a higher ESI than the inoculated control and a similar ESI to the non-inoculated control, indicating that it did not show phytotoxic action on common bean seeds. A high ESI is important to reduce the exposure time of tissues susceptible to pathogens present in the seed itself or in the soil. In the potato crop, the treatment of tubers with KPhi has reduced late blight in tubers inoculated with *Phytophthora infestans*, resulting in 90% seedling emergence, while in inoculated and untreated tubers, the plants did not emerge. KPhi has also reduced the rot caused by *F. solani* by 75% and the severity of *R. solani* by 25% (Lobato et al., 2008). According Carmona et al. (2018), MnPhi and KPhi are feasible alternatives as seed treatments to control Pythium damping-off in soybean.

In the assay to verify the effects of KPhi on nodulation, dry nodule mass, dry shoot mass, shoot nitrogen content, and shoot nitrogen accumulation, there was no significant difference between the inoculant, KPhi + inoculant, and fungicide + inoculant treatments in the number of nodules (Fig. 2).

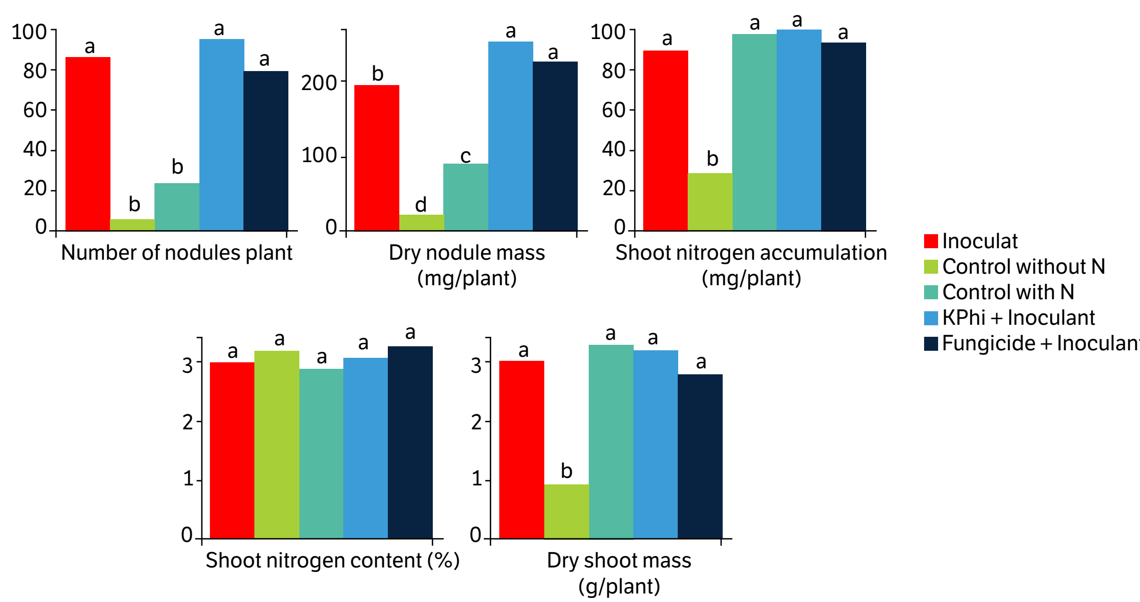

Figure 2. Number of nodules, dry nodule mass, dry shoot mass, shoot nitrogen content, and shoot nitrogen accumulation of bean plants, after the treatment of seeds with potassium phosphite (KPhi) or fungicide (carboxanilide + dimethyldithiocarbamate). Bars followed by the same letter, do not differ statistically from each other by the Scott-Knott test (p≤0.05).

Source: Elaborated by the authors.

The treatments without inoculant application showed low nodulation. KPhi + inoculant and fungicide + inoculant showed higher dry mass of nodules than inoculant treatment. The shoot dry mass and shoot nitrogen accumulation did not differ between the inoculant, control with nitrogen, KPhi + inoculant, and fungicide + inoculant treatments (Fig. 2), indicating that seed treatment with KPhi is not detrimental to the development of the shoot or to nitrogen fixation in bean plants. All treatments showed efficient nitrogen accumulation and shoot growth compared to the control without the addition of nitrogen fertilizer.

KPhi was compatible with *R. tropici*, as it did not affect nodulation. The treatment of common bean seeds with the fungicide carboxanilide + dimethyldithiocarbamate did not affect the formation of nodules, either. The fungicides are usually applied before inoculation to reduce the toxic effects on nitrogen-fixing bacteria (Alcântara Neto et al. 2014). According to Barbosa and Gonzaga (2012), most fungicide combinations, indicated for seed treatment, reduce nodulation and biological nitrogen fixation.

The nodulation process is controlled, in large part, by the exchange of signals between the symbiotic bacteria and the host plant. In the initial stages of root nodule formation, the host species releases signals in its rhizosphere, mainly phenolic compounds (flavonoids) and betaines, which when detected by the bacteria trigger the coordinated expression of a series of nodulation genes (Krishnan et al. 1995; Mercante et al. 2002).

CONCLUSION

In summary, this study provides information regarding the application of KPhi to seeds is a promising technique, to integrate into disease management methods, for common bean crops.

AUTHORS' CONTRIBUTION

Conceptualization: Resende, M.L.V. and Costa, B.H.C.; **Methodology:** Costa, B.H.C., Monteiro, A.C.A and Botega, G.P.; **Investigation:** Costa, B.H.C, Botelho, D.M.S.; Resende, A.R.M. and Pereira, M.H.B.; **Writing – Original Draft:** Costa, B.H.C., Monteiro, A.C.A; Botelho, D.M.S. and Botega, G.P.; **Writing – Review and Editing:** Resende, M.L.V.; Botelho, D.M.S. and Monteiro, A.C.A; **Funding Acquisition:** Resende, M.L.V.; **Resources:** Resende, M.L.V.; **Supervision:** Resende, M.L.V.

DATA AVAILABILITY STATEMENT

All data sets were generated or analyzed in the current study.

FUNDING

Not applicable.

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