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Survival and development of Colorado potato beetles on potatoes treated with phosphite

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A B S T R A C T

Phosphite is a general term used to describe the salts of phosphorous acid H_3PO_3. It is effective in suppressing a number of plant diseases caused by oomycetes and has been shown to reduce populations of several insect species. We investigated the effects of phosphite on the Colorado potato beetles in the field and laboratory. Beetle numbers and defoliation on phosphite-treated plots were lower compared to the control plots during one out of two years of the study. No phosphite effects were detected in the field during the second year of the study. However, larval mortality was significantly higher the second year in the laboratory when larvae were fed on potato foliage excised from the potato plants treated with phosphite in the field. Laboratory tests with excised leaves dipped in a solution of phosphite revealed lower beetle survivorship and prolonged development on the treated foliage. Because of its dual properties as a fungicide and an insecticide, as well as its low toxicity to vertebrates, phosphite is a potentially good fit for integrated pest management programs.

1. Introduction

The Colorado potato beetle (*Leptinotarsa decemlineata* Say) is a defoliating pest of potatoes, posing a management problem to farmers in numerous locations worldwide. Colorado potato beetles possess a suite of characteristics that make them particularly difficult to manage, including a diverse and flexible life history, adaptability, and high fecundity (Alyokhin et al., 2013). Pest management methods developed to date against this species provided short-term to medium-term solutions, but have made little progress toward sustainable crop protection (Casagrande, 1987; Alyokhin et al., 2013). This was in large part due to the high propensity of Colorado potato beetle populations to evolve resistance to a wide range of insecticides (Alyokhin et al., 2008). Since developing new chemistries is an increasingly difficult and expensive task, finding new ways of controlling resistant beetles is especially important.

Several species of oomycetes, such as late blight (*Phytophthora infestans* (Mont.) de Bary (Peronosporales: Pythiaceae)), pink rot (*Phytophthora erythroseptica* Pethyb.), and Pythium leak (*Pythium* spp., primarily *Pythium ultimum* Trow) cause serious diseases of potatoes (Stevenson et al., 2001, 2008). Of those, late blight epidemics usually present the most serious threat. *P. infestans* infections are often accompanied by high levels of precipitation, which impedes application of foliar fungicides. Other factors preventing the complete control of *P. infestans* include its resistance to fungicides, diversity of biotypes, and a lack of rotation of fungicidal active ingredients. *P. infestans* populations have developed resistance to two important fungicides — metalaxyl and mancozeb (Grünwald and Flier, 2005). As a result, *P. infestans* can completely defoliate a potato field within a few weeks from initial infection.

Phosphite is a general term used to describe the salts of phosphorous acid H_3PO_3 — the active ingredients in Phostrol® (Nufarm USA, Burr Ridge, Illinois) (Thao and Yamakawa, 2009). This group of compounds is effective in suppressing a number of plant diseases caused by oomycetes (Smillie et al., 1989; Guest and Grant, 1991; Anderson et al., 2006). Phosphite has a complex mode of action, both exhibiting direct toxicity to the pathogen, as well as indirectly inhibiting its growth through stimulation of host's defense responses (Saindrenan et al., 1998; Guest and Grant, 1991; Anderson et al., 2006; Daniel and Guest, 2006).

In addition to its fungicidal properties, phosphite has been shown to have some activity against several insect species. Following phosphite applications in field trials, Collins (1993) observed reduction in the populations of thrips, *Frankliniella* spp. (Thysanoptera: Thripidae), cotton aphids, *Aphis gossypii* Glover

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(Hemiptera: Aphididae), sweet potato whiteflies, *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae), and cotton leaf perforators, *Bucculatrix thurberiella* Busck (Lepidoptera: Bucculatricidae). Because of its dual activity against oomycetes and insects, phosphite can be a good fit in integrated pest management. However, little attention has been paid to its insecticidal properties since their original discovery by Collins (1993).

Preliminary field experiment conducted in 2008 on the Aroostook Research Farm revealed significant reduction in the Colorado potato beetle numbers at strips sprayed with Phostrol compared to the strips left untreated within the same potato field. Large larvae (third and fourth instars), which comprise the most damaging Colorado potato beetle life stage, were four times more abundant at the untreated strips. Consequently, defoliation was twice as severe for the untreated strips as for the strips sprayed with Phostrol®. In the present study, we further investigated the effects of phosphite on the Colorado potato beetles. A particular emphasis was placed on comparing the impacts on field-collected beetle strain with a known resistance to numerous insecticides with the impacts on a generally susceptible field strain.

2. Materials and methods

2.1. Field experiments

The study was done on the University of Maine Aroostook Research Farm, Presque Isle, Maine. The experiments were conducted on 17.7 m long and 4 row wide plots planted with potatoes ('Katahdin') grown in a three-year rotation with clover and small grains. The plots were arranged in a randomized complete block design with four replications per treatment. Approximately 1.8 m was left between the plots within each block, and blocks were spaced at approximately 3 m. Certified seed potato tubers were cut into 75–80 g pieces and hand-planted at ∼35 cm from each other within the rows and ∼90 cm between the rows. Fields were fertilized with 14-14-14 NPK fertilizer at planting and sprayed weekly with fungicides to control fungal diseases of potatoes.

Half of the plots were treated with phosphite (Phostrol®, Nufarm USA, Burr Ridge, Illinois) applied at 6270 g ai/ha using a tractor-mounted Century boom sprayer; three nozzles per row at 414 kPa pressure and flow rate of 458 L/ha. The other half of the plots were left untreated and used as control. The first application was made when approximately 50% of the Colorado potato beetle egg masses observed on the plots were in the process of hatching. The second application was made 10–14 days after the first application. In 2009, treatment plots were sprayed with phosphite on 9 and 23 July. In 2010, treatment plots were sprayed with phosphite on 30 June and 13 July.

Twenty plants were selected at random within each plot at weekly intervals and visually examined for the presence of Colorado potato beetles (Alyokhin et al., 2005). The number of adults, small larvae (first and second instars), large larvae (third and fourth instars) and egg masses were recorded on a whole-plant basis. Instars were determined based on the width of their pronotae (Boiteau and Le Blanc, 1992). At the time of harvest, tubers from a 3 m strip in the middle two rows of each plot were dug out by hand, pooled together, and weighed in the field.

Percent defoliation on experimental plots was visually estimated on the scale from 0.0 to 5.0, with 0.0 being no defoliation, and 5.0 being complete defoliation (Alyokhin et al., 2007). In 2010, the scale was adjusted to range from 0.0 to 10.0. To minimize potential human bias, the readings were done without reference to treatment. In 2009, defoliation readings were taken for each plot on 16, 23, and 29 July and 5 August. On 23 July, counts and defoliation readings were taken before the phosphite application. In 2010, defoliation readings were taken for each plot on 7, 14, 22, and 30 July.

Both insect counts and defoliation data were rank transformed (Conover and Iman, 1981) because results of the Wilk–Shapiro test (PROC UNIVARIATE, SAS Institute, 2009) revealed their non-normal distribution. Transformed data were analyzed using two-way repeated measures ANOVA (PROC MIXED, SAS Institute, 2009) with treatment and block considered to be the main factors. When interactions between the treatment and week of observations were significant, the data were further analyzed separately for each week using two-way ANOVA (PROC GLM, SAS Institute, 2009). Yield data followed normal distribution and were analyzed using two-sample *t*-tests (PROC TTEST, SAS Institute, 2009). Defoliation indices presented on the figures throughout the manuscript were converted to percent defoliation for the ease of interpretation.

2.2. Larval mortality on field-treated potato foliage

The study was conducted during the 2010 growing season. Potato leaves were collected on 2 July from potato plots sprayed with phosphite on 30 June and from the untreated control plots. Excised leaves were brought to the laboratory, inserted into floral pics with tap water, and placed inside ventilated transparent plastic containers (32 cm by 20 cm by 12.5 cm) lined with moistened paper towels. Six leaves were placed inside each container. Four containers with phosphite-treated foliage and four control containers with untreated foliage were used in the study.

Colorado potato beetle eggs were collected from untreated potato plots on Aroostook Research Farm and incubated until hatching in an environmental chamber (Percival Scientific, Perry, Iowa) maintained in the greenhouse at 20 ± 1 °C and 18L: 6D photoperiod. First instars were collected within 24 h of hatching and used in the experiment.

Thirty of the first instars were placed into each container using a soft brush. The containers were in a walk-in environmental chamber and maintained at 19 ± 1 °C and 18L: 6D photoperiod for four days. After that, the number of dead larvae was counted in each container. The data followed normal distribution and were analyzed using two-sample *t*-test (PROC TTEST, SAS Institute, 2009).

2.3. Larval development

The study was conducted in 2011 in our laboratory. Potato leaves were excised from potted potato plants ('Katahdin') grown in the greenhouse and inserted into individual floral picks with tap water. The leaves were dipped once for 1 s in 25 mL/L solution of phosphite (Phostrol®, Nufarm USA, Burr Ridge, Illinois) in distilled water. Control leaves were dipped in distilled water. To more closely approximate field conditions, 0.069 mL of Tween® was added both to phosphite solution and to water. After the treatment, foliage was allowed to air dry under the fume hood. Three leaves were then placed in a ventilated 5.7 L clear plastic, sealable shoe-storage box (34.54 by 20.32 by 12.7 cm) lined with paper towel. To provide additional insurance against larval escape, a five cm band of Fluon® (Northern Products, Inc., Woonsocket, RI) was painted around the rim of each container.

Larvae from two geographically isolated Colorado potato beetle populations were used in this experiment. One population originated from a commercial potato field in southern Maine and was highly resistant to multiple insecticides. Another population originated from Aroostook Research Farm and was generally susceptible (Alyokhin et al., 2006, 2007). Eggs were collected in the field and incubated until hatching as described above. Thirty of the first instars were then added to each box within 24 h of their hatching.