Evaluation of Late-Season ManKocide Applications to Reduce *Xanthomonas hortorum* pv. *carotae* on Harvested Carrot Seed

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Abstract

Field plots were established to evaluate the efficacy of late-season applications of ManKocide for the suppression of *Xanthomonas hortorum* pv. *carotae* (*Xhc*) on harvested carrot seed. Two ManKocide applications were made in mid to late August 2014 in each of two fields, and harvested seed from the plots was assayed by plating dilutions of seed washes onto a semi-selective agar medium. Late season applications of ManKocide did not significantly reduce *Xhc* populations on harvested seed in this study.

Introduction

Bacterial blight of carrot, caused by the plant pathogenic bacterium *Xanthomonas hortorum* pv. *carotae* (*Xhc*), is a common disease of carrot wherever the crop is grown. The disease can affect carrot foliage, stems, umbels, and roots and can be seed-borne. Symptoms of bacterial blight include small, irregular, chlorotic areas on leaves that can manifest into water-soaked, necrotic lesions. Lesions can also occur on stems and petioles. Floral infections can result in blighted umbels, reduced seed yield, and reduced germination rates of harvested seed. Bacterial blight is of particular concern to carrot seed producers because *Xhc* is seedborne and seed treatments with hot water or disinfectants may not entirely eradicate the pathogen.

Carrot seed producers would like to reduce *Xhc* populations on harvested seed in order to minimize the need for hot water treatment and lessen the impact of bacterial blight on root crop producers. Copper-based bactericides such as ManKocide (mancozeb + copper hydroxide) are routinely applied multiple times each season to manage bacterial blight and increase seed quality. However, the effect of ManKocide on bacterial populations on leaves is generally short-term and it does not consistently reduce bacterial populations on seeds. The objective of this study was to evaluate the impact of late-season, pre-harvest applications of ManKocide on *Xhc* populations on harvested seed.

Materials and Methods

Twelve plots, each consisting of a single four-row set of female carrot plants, were established in two grower-cooperators' fields (#66 and #72) during the 2014 season. Ten umbels were collected from each plot in August to determine pre-treatment *Xhc* populations in both fields. Umbels from each plot were bulked according to field and subjected to an umbel wash assay on semi-selective XCS agar medium. Four 100 g subsamples of umbel tissue were soaked for 2 hours at room temperature in 1.5 L of sterilized PO_4 buffer (0.0125 M) containing Tween 20. After the soak the flasks were shaken by hand for 5 minutes. A 10-fold dilution series was prepared for each suspension ranging from 10^{-1} to 10^{-5} concentration using sterilized PO_4 buffer. A 0.1 ml aliquot of each dilution series was spread onto each of three plates of XCS agar medium for each dilution. The plates were incubated at 28°C in the dark and monitored for the development of

colonies typical of *Xhc*. The number of colonies typical of *Xhc* was counted after 6 days of incubation and suspect colonies were subcultured onto YDC agar medium to observe the development of growth typical of the pathogen.

Treatments included ManKocide (2.5 lb/acre) and a non-treated control that were replicated 6 times in a randomized complete block design. Each plot was treated with two passes in a total volume of approximately 50 gal/acre. Field 66 was treated on August 18 and August 25 and field 72 was treated on August 22 and August 29. Due to prolonged cool and wet weather, harvest of field 66 and field 72 were delayed until September 17 and October 4, respectively.

Plots were mechanically harvested according to standard grower practices and seed from each plot was cleaned by Central Oregon Seeds, Inc. before assaying. Seed from each plot was assayed for *Xhc* using a seed wash dilution plating assay similar to the umbel wash assay described above. Three 10 g subsamples were taken from each seed sample and soaked for 2 hours at room temperature in a 250 ml Erlenmeyer flask containing 100 ml of sterilized PO_4 buffer and one drop of Tween 20. After the soak the flasks were placed on a horizontal shaker set at 250 rpm for 5 minutes. A 10-fold dilution series was prepared for each suspension, plated on XCS agar, and incubated as described above. The number of colonies typical of *Xhc* was counted after 7 days of incubation.

Results and Discussion

Pre-treatment populations of *Xhc* on umbels were $>10^8$ CFU/g umbel tissue in field 66 and $>10^5$ CFU/g umbel tissue in field 72 (Table 1). *Xhc* populations on harvested seed were lower than on umbels collected earlier in the season, but significant differences were not observed between seed harvested from treated and non-treated plots (Table 1). *Xhc* levels in seed harvested from treated plots were slightly higher than *Xhc* levels recovered from seed harvested from non-treated plots but the differences were not significantly different. Cool, wet weather delayed the harvest of both fields for several weeks, so ManKocide applications were not as close to harvest as desired and it is possible that *Xhc* populations may have rebounded during the period between treatment and harvest.

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Table 1. Amount of *Xanthomonas hortorum* pv. *carotae* detected in two fields prior to ManKocide treatments and in plots that were not treated and treated with ManKocide in mid- to late August.

Field	Pre-treatment (± std. dev.)	Non-treated (± std. dev.)	Treated (± std. dev.)
66	2.08×10^8 ($\pm 2.43 \times 10^8$)	4.23×10^7 ($\pm 9.16 \times 10^6$)	4.91×10^7 ($\pm 8.80 \times 10^6$)
72	1.61×10^5 ($\pm 5.13 \times 10^4$)	6.25×10^2 ($\pm 7.19 \times 10^2$)	3.20×10^3 ($\pm 2.84 \times 10^3$)