

## Poor Control of Cucurbit Powdery Mildew Associated with First Detection of Resistance to Cyflufenamid in the Causal Agent, *Podosphaera xanthii*, in the United States

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Accepted for publication 20 July 2018.

Fungicide resistance is a critical aspect of managing *Podosphaera xanthii* because fungicides at risk of resistance development are essential for achieving adequate control. Also, this pathogen has demonstrated it is accurately classified by the Fungicide Resistance Action Committee (<http://www.frac.info/>) as high risk for developing resistance. Torino, which has cyflufenamid (FRAC code U6) as the active ingredient, was registered for control of powdery mildews in the United States in July 2012. At that time, resistance already had been documented in *Podosphaera xanthii* in the United States to benzimidazole fungicides (FRAC code 1), triadimefon, the first-generation demethylation inhibitor (DMI) fungicide (FRAC code 3), quinone outside inhibitor (QoI) fungicides (FRAC code 11), and boscalid in the pyridine carboxamide chemical group of succinate dehydrogenase inhibitors (SDHIs) (FRAC code 7) (McGrath et al. *in press*). Torino label use directions for managing resistance are more restrictive than other fungicides labeled for cucurbit powdery mildew: only two applications are allowed in a 12-month period, with consecutive applications not recommended. In 2012 there were effective DMI fungicides (e.g., triflumizole) to use in a fungicide program with Torino and also quinoxifen (FRAC code 13) for nonedible peel cucurbits. Metrafenone (FRAC code U8) was registered for cucurbit powdery mildew in 2014. Fluopyram in the SDHI pyridinyl-ethyl-benzamide chemical group (FRAC code 7) was registered for use on watermelon in 2012 and other cucurbit crop types in 2016. Resistance to quinoxifen was documented in the United States in 2015 (McGrath 2017).

Torino exhibited poor efficacy compared with some other fungicide treatments when applied alone 6 or 9 times on a weekly schedule in fungicide evaluations conducted in North Carolina in 2016 (Adams and Quesada-Ocampo 2017) and in New York in 2017 (McGrath and Sexton 2018), which contrasted greatly with excellent control obtained in previous evaluations including one conducted in New York in 2011 (McGrath and Hunsberger 2012). In all three experiments, Torino and several other fungicides were tested alone to determine their relative efficacy for a fungicide program and to determine if resistant isolates were present and their selection would affect efficacy.

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**Funding:** This work was supported by the National Institute of Food and Agriculture, U.S. Department of Agriculture, Hatch under NYC-153409.

\*The e-Xtra logo stands for “electronic extra” and indicates that one supplementary figure is published online.

Isolates of *P. xanthii* were collected on Long Island, New York, at the end of the 2017 growing season for a multiyear project monitoring pathogen sensitivity and resistance to fungicides. They were from two commercial field plantings of Halloween pumpkin (*Cucurbita pepo*) and plots in the fungicide evaluation. One additional isolate had been collected in fall 2016.

A leaf disk bioassay (Supplementary Fig. S1) was used to determine the ability of isolates to tolerate cyflufenamid and other fungicides. Pumpkin (cv. Gold Challenger) seedlings at the cotyledon leaf stage (about 7 days old) were sprayed with formulated fungicides in a fume hood, the treated plants were dried overnight, and then disks were cut from cotyledons and placed on water agar in Petri plates with four sections. Six disks with the same treatment were placed in each section. Each plate used to test an isolate had three treatments plus a nontreated control. Isolates were maintained on detached cotyledons placed on water agar in Petri dishes. A glass pipette, with the tip melted closed to permit sterilization by dipping in ethanol, was used to remove spores from these cotyledon cultures and transfer them to each disk center. Isolate growth was assessed about 7 and 14 days later, when the control treatment typically had good growth of the pathogen. Results usually were based on the later assessment. An isolate was considered insensitive (tolerant) to a particular fungicide concentration if it was able to grow and produce spores on at least half of the disks.

Of the 37 isolates from the research plots, 27 were tolerant of 10 ppm of cyflufenamid and 17 were tolerant of 50 ppm of cyflufenamid. In contrast, none of the 73 isolates collected on Long Island in 2007 were able to tolerate 0.5 ppm of cyflufenamid (McGrath and Miazzi, *unpublished*). An isolate able to tolerate 50 ppm of cyflufenamid is considered resistant because it is not expected to be controlled by Torino, considering 53 ppm is the concentration of cyflufenamid when applied at the label rate and 468 liters/ha. These 17 resistant isolates were from plants treated with Torino or other fungicide(s), as well as untreated plots. Isolates sensitive to 50 ppm of cyflufenamid were collected from all of these treatments and also plots treated with Pristine. Neither grower used Torino; however, three of eight isolates from one commercial field were resistant to cyflufenamid. All 15 isolates from the commercial fields were sensitive to boscalid, which these growers also had not applied. Resistance was detected in previous years when boscalid fungicides were being used. An isolate collected in fall 2016 was tested and found to be resistant to cyflufenamid. It was collected from a research plot treated with Quintec in a fungicide efficacy experiment.

Sensitivity to other fungicides was also examined, because multifungicide-resistant isolates of *P. xanthii* have been detected previously on Long Island. Most (13) of the 17 cyflufenamid-resistant isolates were also resistant to boscalid. Three isolates

resistant to cyflufenamid were also resistant to quinoxyfen (active ingredient in Quintec). Six of seven cyflufenamid-resistant isolates tested were found to be resistant to QoI fungicides. Resistance to this chemistry has been common in isolates tested previously. Two of these isolates were also resistant to boscalid and quinoxyfen. Isolates like these, which are resistant to four unrelated fungicide chemistries (FRAC codes 7, 11, 13, and U6), have thwarted efforts to manage fungicide resistance through alternating among chemistries and have increased the challenge to effectively manage powdery mildew in cucurbit crops. All 42 isolates tested were found to be sensitive to 150 ppm of metrafenone (active ingredient in Vivando). Five isolates tested were sensitive to 40 ppm of myclobutanil (active ingredient in Rally, FRAC code 3).

This is the first known report of resistance to cyflufenamid in *P. xanthii* in the United States. In Italy, decline in cyflufenamid efficacy associated with a shift in this pathogen's sensitivity was detected just 1 year after the fungicide was approved for commercial use (Pirondi et al. 2014).

#### Literature Cited

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