Yield Increases in Burrowing Nematode-infested Anthurium with Fluopyram and Trifloxystrobin Applications

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SUMMARY. Burrowing nematode (*Radopholus similis*) causes severe stunting and yield reduction in anthurium (*Anthurium andraeanum*) cut flower production. Two field trials were conducted at commercial grower farms to test the efficacy of fluopyram or fluopyram + trifloxystrobin for managing burrowing nematodes. Nematode population densities in roots and cinder media were evaluated during the trial in addition to cut flower yield and canopy cover. In the first trial, the nematode population in roots was reduced by 57% after two applications of fluopyram 3 months apart. As plant health improved, the increasing anthurium root weight supported higher nematode populations. After 14 months, fluopyram-treated plots had 43% more green canopy cover and a 53% increase in flower production compared with the untreated control plots. At a second location, population densities of burrowing nematode were reduced in roots after one application of fluopyram + trifloxystrobin and remained low with quarterly applications. Nematode populations were initially reduced in fluopyram-treated plots followed by a resurgence as demonstrated in the other trial. Ten months after the initial treatment, flower yield was greater in fluopyram + trifloxystrobin-treated plots with more large and extra-large flowers produced. Canopy cover was 45% and 22% greater with fluopyram + trifloxystrobin and fluopyram applications, respectively. Fluopyram shows potential for management of burrowing nematodes in anthurium by improving plant vigor and cut flower production.

Burrowing nematode (*Radopholus similis*) is a major pest of many important agricultural

crops throughout subtropical and tropical regions. An endoparasitic migratory nematode, burrowing nematode spends its life inside the root where it feeds and reproduces (Duncan and Moens, 2006). Commonly known for its infestations on citrus (*Citrus* sp.) and banana (*Musa acuminata*), burrowing nematode is a pest of quarantine importance (Class A in California, A2 in Europe) that results in strict regulations on potted plant exports from infested regions.

In Hawaii, burrowing nematode causes anthurium decline with limited options for management (Aragaki et al., 1984). Dark lesions visible on

anthurium (*Anthurium andraeanum*) roots during the initial stages of infestation lead to root rot, blackening, and eventual destruction of the root system. The overall plant height as well as leaf development are retarded by infestations of burrowing nematode (Sipes and Lichty, 2002). Fewer flowers are produced because flowers are subtended by each new leaf. Commercial cultivars average less than six leaves per flowers per year per plant in noninfested conditions. Infested plants become severely stunted, resulting in yield reductions of up to 50% (Sipes and Lichty, 2002). Screening of commercial anthurium cultivars and hybrids in Hawaii revealed no obvious resistance to burrowing nematode (Wang et al., 1997).

Anthuriums are the most important cut flower in the Hawaiian floriculture industry with a farm gate value of \$2.7 million (U.S. Department of Agriculture and Hawaii Department of Agriculture, 2019). Even so, maximum production is not being achieved due to yield reductions and high production costs associated with bacterial blight and plant-parasitic nematodes. Historically commercial anthurium growers have treated nematode-infested fields with fenamiphos, but since its voluntary removal from the market (U.S. Environmental Protection Agency, 2011), no post-plant nematicides have been demonstrated effective in managing anthurium decline.

Fluopyram was first introduced as a fungicide in 2007 and later as a nematicide in 2014. Due to the efficacy of this chemical, significant increases in yield and quality were demonstrated in field tests with several agriculturally important crops (Jeschke, 2016). Fluopyram inhibits the mitochondrial respiratory chain in plant-parasitic nematodes, resulting

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Units

To convert U.S. to SI, multiply by	U.S. unit	SI unit	To convert SI to U.S., multiply by
29.5735	fl oz	mL	0.0338
3.1833	fl oz/1000 ft ²	L·ha ⁻¹	0.3141
0.3048	ft	m	3.2808
2.54	inch(es)	cm	0.3937
16.3871	inch ³	cm ³	0.0610
1	micron(s)	μm	1
28.3495	oz	g	0.0353
70.0532	oz/acre	g·ha ⁻¹	0.0143
(°F - 32) ÷ 1.8	°F	°C	(°C × 1.8) + 32

in a depletion of energy and immobilization of the organism. In-furrow applications of fluopyram combined with imidacloprid reduced root-knot (*Meloidogyne incognita*) and reniform nematode (*Rotylenchulus reniformis*) population densities in cotton (*Gossypium* sp.) plantings (Lawrence et al., 2014). Fluopyram suppressed sting nematodes (*Belonolaimus longicaudatus*) in turf with effects being observed for 6 to 8 months after the final application (Crow et al., 2017).

The systemic nature of fluopyram allows it to penetrate the roots and target migratory endoparasitic nematodes. In tea (*Camellia sinensis*) plantations, fluopyram at 600 to 800 g·ha⁻¹ was found to be effective for control of burrowing nematode (Mohotti et al., 2018). Yield increases of 45% were observed with fluopyram treatments in banana infested with burrowing and banana root nematodes [*Pratylenchus coffeae* (Vawa et al., 2019)].

Fluopyram could be useful for limiting damage caused by plant-parasitic nematodes in anthurium cut flower production. This research focuses on examining the potential of two fluopyram formulations, Indemnify and Luna Sensation (Bayer CropScience, Research Triangle Park, NC) against burrowing nematode in field grown anthurium. Indemnify is registered as a fungicide with nematicidal properties on turfgrass and contains only the active ingredient fluopyram (34.5%). Luna Sensation contains fluopyram (21.4%) and trifloxystrobin (21.4%) as active ingredients and is registered as a fungicide for use on fruit and vegetable crops. The specific objectives of this study were to assess their efficacy in 1) reducing burrowing nematode populations and 2) increasing cut flower yields.

Materials and methods

TRIAL 1. A field trial was conducted at a commercial anthurium farm in Hilo, HI, with a 2-year-old established planting of 'Starlight' anthurium inside a shadehouse with 78% shade. Plants were growing in beds of volcanic cinder medium on the ground, a typical growing medium for anthurium farmers in Hawaii. All plants were stunted from a natural infestation of burrowing nematode before the initiating the experiment. Plants received natural

rainfall of ≈380 cm per year and were supplemented by overhead irrigation during drought. Average temperature in the shadehouse was 23 °C with 90% average relative humidity. The two treatments evaluated in the field trial were a fluopyram application (Indemnify) and an untreated control. Each treatment plot was 15 × 4 ft and contained 52 plants. Treatment plots were replicated four times and arranged in a randomized complete block design. Two border rows of anthurium were placed in between treated and untreated plots.

The trial was initiated on 20 Apr. 2017 and concluded on 20 July 2018. Five applications were administered during that time. The treated plots were sprayed with fluopyram alone at a rate of 0.14 fl oz/1000 ft². The application was applied to the foliage until runoff and repeated once every 3 months. The untreated control was sprayed with tap water.

Nematode sampling was conducted before the first fluopyram application and every month thereafter for 14 months. Anthurium roots and cinder from the rhizosphere were collected from six plants per plot and composited. At the laboratory, all roots were separated from the cinder media. Subsamples of ≈10 g of roots (wet weight) and 100 cm³ of cinder were taken from each sample. Roots or cinder were placed individually on Baermann funnels for 72 h (Walker and Wilson, 1960). Nematodes were concentrated on a 20-μm-pore sieve, collected in 25 mL of tap water, and counted under an inverted light microscope (DM1L; Leica, Wetzlar, Germany).

Anthurium cut flowers were harvested weekly and graded according to industry standards by the grower as corsage (3.5–3.9 inches), small (4.0–4.5 inches), medium (4.5–4.9 inches), and large (5.0–5.9 inches) width. At the termination of the trial, 14 months after the initial fluopyram application, photographs were taken of three subsections of each plot measuring 4 × 5 ft from a horizontal plane 5 ft above the ground. Using the imaging application Canopeo (Oklahoma State University, Stillwater, OK), percent canopy cover was quantified and compared among treatments (Patrignani and Ochsner, 2015).

TRIAL 2. A field trial was conducted at a second commercial anthurium

farm in Pahoia, HI, commencing on 2 Aug. 2017 and ending on 20 July 2018 with four applications administered during the trial. Treatments were tested on 'Midori' anthurium planted 18 months before the initiation of the trial. Growing conditions were as described in Trial 1. Plants were naturally infested with burrowing nematode. Each plot contained 60 plants with four plots per treatment. Treatment plots were divided by borders of six rows of anthurium plants.

Treatments consisted of fluopyram at a rate of 0.14 fl oz/1000 ft², fluopyram + trifloxystrobin at 0.30 fl oz/1000 ft² of each, and an untreated water control. The products were applied foliarly, and applications were made by spraying to runoff once every 3 months.

Nematode sampling was performed before the first treatment to assess initial nematode population densities, and sampling was conducted every 3 months thereafter immediately before fluopyram applications. Root and cinder samples were collected from each plot and processed as described earlier.

Weekly harvesting and grading of cut flowers were carried out by the farmer as described in Trial 1 with the addition of an extra-large (≥6.0 inches width) standard. Photographs were obtained, and the plant canopy was quantified using Canopeo as previously described.

STATISTICAL ANALYSIS. Nematode data from each trial were subjected to time × treatment analysis of variance (ANOVA). When an interaction was significant, treatments were analyzed by time sections. Otherwise data were subjected to repeated measure over time using PROC GLM in SAS (version 9.4; SAS Institute, Cary, NC). Anthurium yield data and plant canopy cover were subjected to one-way ANOVA. All means were separated by least significant difference.

Results

TRIAL 1. Monthly monitoring of burrowing nematode population densities from anthurium roots revealed lower nematode densities in fluopyram-treated plots compared with untreated control plots during the first 6 months after initial nematicide application ($P \leq 0.01$) with a population reduction of 57% compared with the

Table 1. Population of burrowing nematode recovered from ‘Starlight’ anthurium treated with fluopyram at 0.14 fl oz/1000 ft² (0.446 L·ha⁻¹).

Treatment	Burrowing nematodes (no.)			
	1–6 mo. after first application		7–14 mo. after first application	
	Roots (no./10 g) ^z	Cinder (no./100 cm ³) ^z	Roots (no./10 g)	Cinder (no./100 cm ³)
Fluopyram	30 b ^y	22 a	36 a	38 a
Untreated	68 a	34 a	29 a	24 b

^z1 nematode/10 g = 45.3592 nematodes/lb, 1 nematode/100 cm³ = 283.1684 nematodes/ft³.

^yTreatments with the same letter within a column are not different ($P > 0.05$) according to least significant difference test.

Table 2. ‘Starlight’ anthurium cut flowers harvested over 14 mo. in plots treated with fluopyram at 0.14 fl oz/1000 ft² (0.446 L·ha⁻¹).

Treatment	Flower grades ^z				Total
	Corsage	Small	Medium	Large	
Fluopyram	902 a ^y	471 a	219 a	36 a	1628 a
Untreated	679 b	295 b	89 b	2 a	1065 b

^zAnthurium spathes are measured by width to determine flower grade: corsage = 3.5–3.9 inches, small = 4.0–4.5 inches, medium = 4.5–4.9 inches, large = 5.0–5.9 inches; 1 inch = 2.54 cm.

^yTreatments with the same letter within a column are not statistically different according to least significant difference test ($P > 0.005$).

Table 3. Population of burrowing nematode recovered from ‘Midori’ anthurium treated with fluopyram + trifloxystrobin (0.30 fl oz/1000 ft² of each) or fluopyram (0.14 fl oz/1000 ft²) over a 12-mo. period.^z

Treatment	Burrowing nematodes (no.)	
	Root (no./10 g) ^z	Cinder (no./100 cm ³) ^z
Fluopyram + trifloxystrobin	3 b ^y	5 a
Fluopyram	26 ab	18 a
Untreated	32 a	21 a

^z1 fl oz/1000 ft² = 3.1833 L·ha⁻¹, 1 nematode/10 g = 45.3592 nematodes/lb, 1 nematode/100 cm³ = 283.1684 nematodes/ft³.

^yTreatments with the same letter within a column are not different ($P > 0.05$) according to least significant difference test.

control (Table 1). In the following 8 months, no differences in the number of nematodes extracted from roots were observed between treatments ($P \geq 0.05$). No differences in population densities of burrowing nematode were detected in cinder during the first 6 months of the trial ($P \geq 0.05$). More nematodes were recovered from fluopyram-treated plots in the remaining months ($P \leq 0.05$).

After two fluopyram applications, weekly cut flower yield improved and began to increase with subsequent applications. Over the 14-month trial, fluopyram-treated plots produced 53% more cut flower stems than untreated plots [1628 vs. 1065 stems ($P \leq 0.01$)]. The size of harvested flowers also improved with 145% more medium-sized and 1700% more large flowers from treated plots (Table 2).

Plant vigor at the end of the trial indicated a 43% greater percentage of

green canopy cover in fluopyram-treated vs. untreated plots ($P \leq 0.01$). More robust roots were observed on the surface of cinder beds along with an increase in foliage production in plots treated with fluopyram. Root sampling was more efficient in treated plots due to the availability of additional roots per plant.

TRIAL 2. Plots treated with fluopyram + trifloxystrobin started with the highest nematode population densities in the roots before fluopyram applications. The results showed that after one treatment, the number of burrowing nematodes in the roots dropped lower than all other treatments and remained low with quarterly applications [$P \leq 0.01$ (Table 3)]. Nematode populations in the roots were initially reduced in fluopyram-treated plots followed by an increase as demonstrated in Trial 1. Because no

interaction between sampling data and fluopyram treatments were found in ANOVA, population densities of burrowing nematode in roots over a 12-month period were combined in a repeated-measure analysis. The number of burrowing nematodes in roots differed only among fluopyram + trifloxystrobin-treated plots and untreated controls ($P \leq 0.05$). No differences in burrowing nematode population densities were detected among treatments in cinder ($P \geq 0.05$).

Over the 10 months that yield data were taken, no difference was observed in total flower yield between fluopyram-treated plots and untreated control plots ($P \geq 0.05$). An increase in flower size was observed with more extra-large flowers harvested in fluopyram + trifloxystrobin plots compared with untreated controls ($P \leq 0.05$). With increasing applications, yields in treated plots improved. During the last month of data collection, fluopyram + trifloxystrobin-treated plots had a higher overall yield than untreated plots ($P \leq 0.01$) and a greater number of large ($P \leq 0.01$) and extra-large flowers ($P \leq 0.05$) (Table 4).

Plant vigor increased with both fluopyram applications [$P \leq 0.01$ (Fig. 1)]. Fluopyram + trifloxystrobin-treated plots had greater green canopy cover (86%) than fluopyram and the untreated control. Fluopyram-treated plots had more plant growth than untreated plots with 72% green canopy cover compared with 59% in the control.

Discussion

Quarterly fluopyram and fluopyram + trifloxystrobin applications to field grown anthurium initially decreased burrowing nematode populations in the roots and increased canopy cover, flower size, and flower yield over time. A marketable increase in cut flower production was observed in ‘Starlight’ (Trial 1) after just two applications of fluopyram. In Trial 2, quarterly applications of fluopyram + trifloxystrobin resulted in increased cut flower size and yield. Plants treated with fluopyram in the second trial also demonstrated a yield and size increase, although this was not statistically different from the other treatments. Some inconsistencies existed in the yield data with

Table 4. ‘Midori’ anthurium cut flowers harvested during month 10 after four applications of fluopyram + trifloxystrobin (0.30 fl oz/1000 ft² of each) or fluopyram (0.14 fl oz/1000 ft²).^z

Treatment	Flower grades ^y					Total
	Corsage	Small	Medium	Large	Extra large	
Fluopyram + trifloxystrobin	3 a ^x	15 a	29 a	29 a	21 a	97 a
Fluopyram	6 a	23 a	25 a	17 ab	8 ab	79 ab
Untreated	6 a	22 a	19 a	7 b	0 b	54 b

^z1 fl oz/1000 ft² = 3.1833 L·ha⁻¹.

^yAnthurium spathes are measured by width to determine flower grade: corsage = 3.5–3.9 inches, small = 4.0–4.5 inches, medium = 4.5–4.9 inches, large = 5.0–5.9 inches, extra large ≥6.0 inches; 1 inch = 2.54 cm.

^xTreatments with the same letter within a column are not different ($P > 0.05$) according to least significant difference test.

Fig. 1. Green canopy cover (on the left) measured using imaging analysis (on the right) of ‘Midori’ anthurium in (A) untreated, (B) treated with fluopyram at 0.14 fl oz/1000 ft² (0.446 L·ha⁻¹), and (C) treated with fluopyram + trifloxystrobin at 0.30 fl oz/1000 ft² (0.955 L·ha⁻¹) of each.

regard to the performance of fluopyram between Trials 1 and 2, which could be attributed to the nematode tolerance/susceptibility of the two

cultivars. Whereas susceptibility of ‘Starlight’ to burrowing nematode has not been assessed, ‘Midori’ used in Trial 2 demonstrated moderate

tolerance to burrowing nematode among 17 commercially grown cultivars evaluated in a bioassay (Wang et al., 1998).

Evaluating burrowing nematode populations in the cinder was not a good indicator of nematicide efficacy in this study due to the endoparasitic nature of the nematode. Taking samples from cinder, which has larger pore spaces than regular soil, even in close proximity to the root system showed no significance among treatments in both trials. Extracting burrowing nematodes from roots, however, was useful in detecting the nematode-suppressive effects of fluopyram.

The first two applications of fluopyram caused a reduction in the population densities of burrowing nematode in roots in both trials. After the third application, the trend began to reverse as plants in treated plots exhibited increasing plant vigor and could support higher nematode populations. At the same time, roots of untreated plants became increasingly necrotic with limited living tissue to support nematode feeding. In other anthurium studies, fewer roots also resulted in lower nematode reproduction (Sipes and Lichty, 2002; Wang et al., 1997).

Initial symptoms of burrowing nematode infestations are necrotic root lesions, which are accelerated into extensive root rots by secondary pathogens and saprophytes (Uchida et al., 2003). Although not identified or evaluated in this study, parasitic oomycetes (*Pythium*) and plant-parasitic fungi (*Rhizoctonia solani* and *Colonectria crotalariae*) have previously been isolated from anthurium roots at the University of Hawaii’s College of Tropical Agriculture and Human Resources Agricultural Diagnostic Service Center (B. Bushe, unpublished data). The fungicidal effects of fluopyram and trifloxystrobin in this trial could have caused mortality to these fungal organisms and delayed the progression of root decomposition. Although pathogenic fungi were found in advanced stages of root rot in declining plants, applications of fungicide alone did not reduce disease severity (Aragaki et al., 1984). Aragaki et al. (1984) also reported that fungi were not recovered from young necrotic lesions, suggesting that burrowing nematodes

were the cause of initiating disease symptoms.

Whether applied as a nematicide or a fungicide, fluopyram appeared to improve the plant health and yield of anthurium in the presence of burrowing nematode. Although nematode populations in the media were not representative of its nematode-suppressive effects, plant vigor, flower size, and cut flower yield were significantly improved with fluopyram and fluopyram + trifloxystrobin applications. Fluopyram appears to have potential in mitigating the damage caused by burrowing nematodes and shows promise as a management tool in anthurium field production.

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