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Effect of florpyrauxifen-benzyl concentration–exposure time on hygrophila and rotala

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ABSTRACT

Submersed vegetation in flood-control canals must be managed to facilitate the rapid movement of water during periods of heavy rainfall. Two of the most problematic and difficult-to-control macrophytes in south Florida's canals are hygrophila [*Hygrophila polysperma* (Roxb.) T. Anderson] and rotala [*Rotala rotundifolia* (Buch.-Ham. ex Roxb.) Koehne]. The submersed forms of these amphibious species quickly fill the water column and restrict water flow; thus the management of hygrophila and rotala is a major concern for resource managers. There are limited chemical control options for managing these species; therefore, the goal of these experiments was to evaluate florpyrauxifen-benzyl at varying concentration–exposure times for control of hygrophila and rotala. Submersed hygrophila and rotala plants were exposed to concentrations of florpyrauxifen-benzyl at 0, 2.5, 5, 10, or 20 $\mu g\ L^{-1}$ at 6-, 24-, or 48-h exposure times. Hygrophila dry weights decreased as florpyrauxifen-benzyl concentration and exposure time increased. Rotala was extremely sensitive to florpyrauxifen-benzyl; virtually all rotala biomass was eliminated by even the shortest exposure (6 h) to the lowest concentration (2.5 $\mu g\ L^{-1}$) of florpyrauxifen-benzyl. This research reveals the utility of florpyrauxifen-benzyl as a potential management option and provides guidance to resource managers charged with controlling hygrophila and rotala, especially in systems where flow can reduce exposure times.

Key words: arylpicolinate, canal weeds, CET, herbicide, *Hygrophila polysperma*, ProcellaCOR, *Rotala rotundifolia*

INTRODUCTION

Around 6.2 million people, representing around 30% of Florida's entire population, live in the Miami metropolitan area (hereafter MMA) which comprises Palm Beach, Broward, and Miami–Dade counties, the three most populated counties in the state (World Population Review 2019). The MMA has a total land area of 15,890 km^2, the majority of which is preserved by state or federal orders (i.e., the Everglades) with an estimated urbanized area of 2,890 km^2, resulting in a population density of 2,145 residents km^{-2}. This extremely high population density, coupled with south Florida's flat topography, low average elevation of <2 m above sea level (Ruggeri 2017), and frequent extreme weather events (e.g., tropical storms and hurricanes), makes floodwater management critical to avoid loss of life and property damage.

Flood-control canals are a major part of south Florida's landscape and must be maintained with minimal aquatic vegetation to allow rapid water movement during periods of heavy rainfall. Although a number of aquatic macrophytes often occupy these canals, hygrophila and rotala are two of the most problematic and difficult to control. Both species are native to Asia and are amphibious; their ability to grow under both submersed and emergent conditions allows them to fill the water column quickly and restrict water flow (Gettys and Della Torre 2020). This interferes with canal functionality, so reducing populations of hygrophila and rotala is a major concern for resource managers.

The first report of hygrophila in Florida occurred near Tampa in 1965, but the species was misidentified as a native species of twinflower (*Dyschoriste* sp.) until 1977 (Les and Wunderlin 1981), and by then had already become well established in south Florida's canals (Vandiver 1980). The species has dark-green lanceolate leaves that are attached in an opposite fashion to square stems (Les and Wunderlin 1981, Gettys and Enloe 2019). Hygrophila reproduces by seed and stem fragmentation, but the impact of seed production on invasion is not well understood (Sutton 1995).

Rotala was first reported in a Broward County, FL canal in 1996. Within a decade, the species had established large isolated populations throughout southern Florida (Burks et al. 2003, Jacono and Vandiver 2007) and is currently considered one of the most serious submersed macrophytes in the region's canal systems (M. Bodle, pers. comm.). The submersed form of rotala has dull red to green stems and leaves; leaf shape is lanceolate and attachment is opposite. Emergent rotala has rounded, bright green leaves arranged in an opposite manner on a pink to red stem (Gettys and Della Torre 2020). Little is known regarding seed biology of rotala, but the species reproduces very easily via fragmentation, which is likely the species' primary means of reproduction.

Mechanical harvesting is sometimes used to manage hygrophila and rotala in flood-control canals, but may not be operationally feasible because of the creation of fragments (which could facilitate spread). Additionally, all harvested plant material would need to be hauled away (which dramatically increases costs). As such, herbicides are often used for management of these weeds in south Florida canals and other infested waterbodies.

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Fast et al. (2009) reported that foliar applications of a number of aquatic herbicides provided excellent (>90%) control of emergent hygrophila, but did not evaluate treatments on the submersed form. Sutton et al. (1994, 1995) reported that single applications of 2.5, 3, or 4.5 mg ai L^{-1} potassium endothall or the amine salt of endothall at 0.2 mg ai L^{-1} provided only short-term control of submersed hygrophila, and populations had regrown to pretreatment levels within 3 mo after application. Vandiver and Timmer (2006) were able to control submersed hygrophila in static tanks with endothall, diquat, and auxinic herbicides, but only when plants were treated every 2 wk over a 10-wk period. In contrast, Haller and Gettys (2013) found that flumioxazin effectively eliminated submersed hygrophila at <200 μg ai L^{-1}, less than half the maximum label rate. The majority of herbicides labeled for aquatic use in the United States have been evaluated for efficacy on rotala; however, only fluridone (Della Torre et al. 2017), triclopyr, and 2,4-D (Puri and Haller 2010) have been documented to provide excellent (>90%) control of rotala in mesocosms trials.

The limited number of EPA-registered aquatic herbicides available for managing aquatic invasive species, in addition to best management practices to reduce the development of herbicide resistance, creates a critical need for evaluating newly registered aquatic herbicides to manage hygrophila and rotala. The most recent entry to the aquatic herbicide market is florpyrauxifen-benzyl [4-amino-3-chloro-6-(4-chloro-2-fluoro-3-methoxyphenyl)-5-fluoro-pyridine-2-benzyl ester] (ProcellaCOR®), which was developed by SePRO Corporation (Carmel IN) in partnership with Dow Agrosciences (Indianapolis, IN) (now Corteva Agriscience) and is an arylpicolinate synthetic auxin (HRAC group O/WSSA group 4) (Netherland and Richardson 2016). Florpyrauxifen-benzyl is classified as a reduced risk pesticide by the U.S. Environmental Protection Agency (EPA). As a reduced-risk pesticide, the EPA has deemed the pesticide to have a low impact on human health, a lower toxicity to nontarget organisms, a low potential for groundwater contamination, low use rates, low pest-resistance potential, and to be compatible with integrated pest management practices (EPA, 2021). The maximum label rate of ProcellaCOR® SC is 48 μg L^{-1} florpyrauxifen-benzyl (SePRO Corp. 2018).

A number of researchers (e.g., Netherland and Richardson 2016, Richardson et al. 2016, Beets and Netherland 2018, Haug 2018, Beets et al. 2019, Mudge and Netherland 2020) have evaluated the effects of florpyrauxifen-benzyl on a variety of aquatic plants, including alligatorweed (*Alternanthera philoxeroides*), lemon bacopa (*Bacopa caroliniana*), crested floatingheart (*Nymphoides cristata*), eelgrass (*Vallisneria americana*), fanwort (*Cabomba caroliniana*), monoecious and dioecious hydrilla (*Hydrilla verticillata*), lanceleaf sagittaria (*Sagittaria lancifolia*), parrotfeather (*Myriophyllum aquaticum*), and several related watermilfoils (*M. heterophyllum, M. sibiricum, M. spicatum, M. sibiricum* × *M. spicatum*), pickerelweed (*Pontederia cordata*), and waterwillow (*Justicia americana*). This body of work has revealed that florpyrauxifen-benzyl can be effective at much lower use rates (sometimes 50 to 100× lower—see Beets and Netherland 2018) than other herbicides such as 2,4-D, triclopyr, and endothall, resulting in a significant decrease in the volume of herbicide required for management of aquatic weeds. Some reports (e.g., Beets and Netherland 2018, Beets et al. 2019, Mudge and Netherland 2020) describe different exposure scenarios and suggest that required exposure times for effective control are relatively short, but do not report calculated ET values—the exposure time required to reduce a particular trait such as growth by a given percentage compared to nontreated control plants. The exposure time needed to reduce biomass of submersed species can be an important factor when choosing a management strategy, and that is especially true for canal weeds such as rotala and hygrophila, where significant flow can reduce exposure time. Potential low use rates, coupled with short exposure time requirements, make florpyrauxifen-benzyl a potential candidate for managing hygrophila and rotala in areas subject to high water exchange such as the canals they routinely invade. Therefore, the goal of these experiments was to determine the effects of florpyrauxifen-benzyl concentration-exposure times (CETs) on submersed hygrophila and rotala.

MATERIALS AND METHODS

Plant material was collected from existing populations of both species maintained in culture at the University of Florida Institute of Food and Agricultural Sciences Fort Lauderdale Research and Education Center. These experiments were conducted twice; for each run, 2-L HDPE pots without holes were filled with coarse masonry sand amended with 2 g L^{-1} of a controlled-release fertilizer.[1] Each pot was then planted with 10 unrooted 10-cm-long apical cuttings of either hygrophila or rotala. Planted pots were maintained under overhead irrigation until new growth was evident, then 12 (Run 1) or 9 (Run 2) pots of each species were moved to one of five 2.4-m HDPE tanks that were filled to a depth of ca. 50 cm with 1,514 L of pond water. Plant material was maintained under submersed conditions for 6 wk and was healthy and robust at the time of treatment.

We evaluated florpyrauxifen-benzyl[2] at four concentrations (2.5, 5, 10, and 20 μg L^{-1}) and three exposure times (6, 24, and 48 h). A nontreated control was also included. Run 1 plants were treated on 25 September 2017 and Run 2 plants were treated on 8 August 2018. Treatments were applied by mixing the appropriate volume of florpyrauxifen-benzyl with 8 L of pond water and then pouring the mixture around the inside perimeter of the specified tank to ensure even distribution. Six hours after treatment (HAT), four (Run 1) or three (Run 2) pots (replicates) of each species were randomly selected and pulled from each tank, labeled with the appropriate CET (for example, "rotala, 5 μg L^{-1}, 6 h") and moved to 68-L mesocosms filled with nontreated pond water. Each mesocosm housed all pots (replicates) of a single species that were treated with the same concentration of florpyrauxifen-benzyl and subjected to the same exposure time. This protocol was followed for the remaining exposure times (24 and 48 HAT), and all plants were maintained for 6 wk after treatment (WAT). Water pH was recorded at noon on the day of treatment and then weekly for the duration of these experiments. Data loggers[3] were

Table 1. Exposure time (h) needed to reduce biomass of plants treated with different concentrations of florpyrauxifen-benzyl by 50% (ET$_{50}$) or 90% (ET$_{90}$) compared to nontreated control plants. "–" = no live biomass at this concentration, unable to calculate ET$_{50}$ and ET$_{90}$.

	Concentration (µg L^{-1})							
	2.5		5		10		20	
Species	ET$_{50}$	ET$_{90}$	ET$_{50}$	ET$_{90}$	ET$_{50}$	ET$_{90}$	ET$_{50}$	ET$_{90}$
Hygrophila Run 1	203	673	50	164	45	151	–	–
Hygrophila Run 2	180	598	33	108	34	113	12	41
Rotala (pooled)	0.3	2.8	–	–	–	–	–	–

Table 2. Concentration (µg L^{-1}) of florpyrauxifen-benzyl needed to reduce biomass by 50% (EC$_{50}$) or 90% (EC$_{90}$) compared to nontreated control plants under different exposure times.

	Exposure time (h)					
	6		24		48	
Species	EC$_{50}$	EC$_{90}$	EC$_{50}$	EC$_{90}$	EC$_{50}$	EC$_{90}$
Hygrophila Run 1	13	44	20	68	9	30
Hygrophila Run 2	18	61	7	22	4	15
Rotala (pooled)	0.3	1.4	0.3	0.5	0.3	0.5

deployed to record temperature in two mesocosms with treated plants and two mesocosms with nontreated control plants every 10 min for the duration of each run.

All live aboveground biomass was destructively harvested 6 WAT (Run 1: 6 November 2017; Run 2: 19 September 2018), rinsed to remove algae and other debris, and transferred to a forced-air oven at 65 C for 1 wk before being weighed to obtain dry biomass. Dry biomass data were subjected to analysis of variance, and nonlinear regression was performed using exponential decay function.[4] Regression components were used to calculate ET$_{50}$ and ET$_{90}$ values (the effective time [hours of exposure] needed to reduce biomass by 50 and 90% compared to nontreated control plants) for each concentration of florpyrauxifen-benzyl. In addition, we calculated EC$_{50}$ and EC$_{90}$ values [the effective concentration (µg L^{-1}) of florpyrauxifen-benzyl needed to reduce biomass by 50 and 90% compared to nontreated control plants] for each exposure time.

RESULTS

There were no differences in water temperature or pH among mesocosms with treated or nontreated plants in either run. Mean temperature ranged from 27.7 to 28.1 C; minimum and maximum temperature ranges were 22.6 to 25.1 C and 31.0 to 33.2 C, respectively. Water pH on the day of treatment averaged 8.5 and ranged from 7.9 to 9.1 throughout the course of the experiments.

Analysis of variance performed on rotala data did not detect a run effect, so data from Runs 1 and 2 were combined. Rotala was extremely sensitive to florpyrauxifen-benzyl; all rotala biomass was eliminated at all CETs evaluated, with the exception of one replicate exposed to 2.5 µg L^{-1} for 6 h. Rotala treated with 2.5 µg L^{-1} florpyrauxifen-benzyl would require 0.3 or 2.8 h of exposure to attain a 50 or 90% reduction in biomass compared to nontreated plants; ET$_{50}$ and ET$_{90}$ estimates for rotala treated with other concentrations could not be calculated because all plant material treated with $>$2.5 µg L^{-1} florpyrauxifen-benzyl died at all exposure durations (Table 1). Rotala exposed to this auxin for 6 h would require a treatment concentration of 0.3 or 1.4 µg L^{-1} florpyrauxifen-benzyl to have a biomass reduction of 50 or 90% compared to nontreated plants, whereas plants exposed for 24 or 48 h would require a concentration of 0.3 or 0.5 µg L^{-1} to achieve 50 or 90% reductions (Table 2).

There were differences in hygrophila response between the runs, so these data were analyzed and are presented separately. Hygrophila treated with 2.5 µg L^{-1} florpyrauxifen-benzyl would require 203 (Run 1) or 180 (Run 2) h of exposure to attain a 50% reduction in biomass compared to nontreated plants (Table 1), whereas 90% reductions would be expected after 673 (Run 1) or 598 (Run 2) h of exposure. With the exception of plants treated with 10 µg L^{-1} florpyrauxifen-benzyl in Run 2, ET$_{50}$ and ET$_{90}$ values decreased as concentration increased. Similar to rotala, ET$_{50}$ and ET$_{90}$ estimates for hygrophila treated with 20 µg L^{-1} florpyrauxifen-benzyl in Run 1 could not be calculated because all plant material treated with this concentration died at all exposure times (Table 1). The EC$_{50}$ and EC$_{90}$ values of hygrophila exposed for 6 h in Run 1 were lower than those calculated for Run 2; in contrast, florpyrauxifen-benzyl concentrations needed to reduce biomass by 50 or 90% in plants exposed for 24 or 48 h in Run 1 were 3× (24 h) or 2× (48 h) higher than those in Run 2 (Table 2). With the exception of the 24-h exposure in Run 1, the concentration of florpyrauxifen-benzyl required to reduce biomass by 50 or 90% decreased as exposure time increased.

DISCUSSION

These studies reveal that both of these submersed weeds are susceptible to florpyrauxifen-benzyl. Rotala was extremely sensitive to florpyrauxifen-benzyl, with virtually all biomass eliminated at all CETs evaluated. With respect to the effect of concentration, rotala treated with 2.5 µg L^{-1} florpyrauxifen-benzyl would require 0.3 or 2.8 h of exposure to attain a 50 or 90% reduction in biomass compared to nontreated plants. In respect to the effect of exposure time, rotala exposed to this auxin for 6 h would require a treatment concentration of 0.3 or 1.4 µg L^{-1} florpyrauxifen-benzyl to have a biomass reduction of 50 or 90% compared to nontreated plants.

Hygrophila, although less sensitive than rotala, was still affected by these treatments. With respect to the effect of concentration, ET$_{50}$ and ET$_{90}$ values for hygrophila generally decreased as herbicide concentration increased. With respect to the effect of exposure time, the concentration of florpyrauxifen-benzyl required to reduce biomass by 50 or 90% decreased as exposure time increased. The reasons for hygrophila's differing response in Runs 1 and 2 are unclear, but could be due to a variety of factors. The half-life of the molecule in water ranges from 1 to 8 d, but is strongly influenced by environmental conditions (Heilman and

Getsinger 2018; Massachusetts Department of Agriculture 2019). The main driver for degradation of florpyrauxifen-benzyl is photolysis, with some degradation due to microbial activity and to hydrolysis in high (>9) pH waters (Heilman and Getsinger 2018). In addition, Beets and Netherland (2018) speculated that water temperature could influence florpyrauxifen-benzyl efficacy.

Initial water temperature and pH were similar in both runs of our experiments, so those factors are unlikely to have influenced our results. Daylength—and hours of exposure to sunlight after treatment—did differ between the runs. Treatments in both runs began at noon and were applied "low to high," with around 15 min between applications (e.g., the 2.5 $\mu g\ L^{-1}$ treatment was applied at noon, 5 $\mu g\ L^{-1}$ at 12:15, and so on). According to www.dateandtime.com, daylength during Run 1 ranged from 12 h 3 min (on day of treatment [DOT] 25 September 2017) to 11 h 2 min (on day of harvest [DOH] 6 November 2017). Sunset was at 7:12 P.M. on DOT, so treatments with 2.5 $\mu g\ L^{-1}$ florpyrauxifen-benzyl received around 7 h 12 min of sunlight after application, those with 5 $\mu g\ L^{-1}$ received around 6 h 58 min, and so on. Daylength during Run 2 ranged from 13 h 12 min (on DOT 8 August 2018) to 12 h 12 min (on DOH 19 September 2018). Sunset was 8:02 P.M. on DOT, so treatments with 2.5 $\mu g\ L^{-1}$ florpyrauxifen-benzyl received around 8 h 2 min of sunlight after application, those with 5 $\mu g\ L^{-1}$ received around 7 h 47 min, and so on. Based on this information, daylengths were around 1 hr longer throughout Run 2 than Run 1, and treatments in Run 2 were exposed to sunlight for around 50 min longer after application than those in Run 1.

If photolysis were responsible for differences between the runs, additional exposure to sunlight after application should increase herbicide degradation, resulting in longer exposure-time requirements (in other words, longer days → more exposure to sunlight → faster degradation → lower concentration → longer time requirement). Based on this model, ET values should have been higher in Run 2, because florpyrauxifen-benzyl received more exposure to sunlight. However, our data show the opposite, with ET values in Run 1 consistently higher than those in Run 2; in other words, hygrophila treated with the same concentration of florpyrauxifen-benzyl in both runs required longer exposure times in Run 1 to achieve similar reductions in biomass. It therefore seems unlikely that degradation via photolysis is responsible for differences between the runs. We cannot attribute run differences to pH, water temperature, or photolysis, but it is still clear that hygrophila is sensitive to florpyrauxifen-benzyl.

The body of work regarding the herbicidal effects of florpyrauxifen-benzyl is steadily growing. For example, Netherland and Richardson (2016) exposed crested floatingheart to florpyrauxifen-benzyl for 14 d under static conditions and calculated an EC_{50} of 5.6 $\mu g\ L^{-1}$ for biomass. Haug (2018) and Richardson et al. (2016) evaluated the effects of florpyrauxifen-benzyl on seven aquatic species and reported dry weight EC_{50} values ranging from <0.3 to >81 $\mu g\ L^{-1}$, but those plants were also maintained under static conditions, so exposure time was not evaluated. Beets and Netherland (2018) exposed four aquatic species to 12 to 48 $\mu g\ L^{-1}$ florpyrauxifen-benzyl for durations ranging from 24 h to 7 d ("static") and found that dry weights in most treatments were reduced compared to nontreated control plants, but they did not calculate EC or ET values. Beets et al. (2019) applied a range of florpyrauxifen-benzyl concentrations to nine submersed species for 6 h, 12 h, or static exposure periods and reported that dry weights of nonnative watermilfoils were greatly reduced by exposure to any CET, whereas native species were not negatively impacted; however, they did not report EC or ET values for the species under investigation. Mudge and Netherland (2020) treated crested floatingheart with a range of florpyrauxifen-benzyl concentrations over four exposure periods and reported that all but 1 CET resulted in >75% reduction in biomass (but they did not provide EC or ET values); also, in most cases, biomass decreased as exposure time increased within a concentration.

As shown above, most papers report the effects of this auxin on biomass as percent reduction compared to controls or as EC values, but information regarding exposure time requirements are lacking. Several authors did evaluate florpyrauxifen-benzyl exposure times (e.g., Beets and Netherland 2018, Beets et al. 2019, Mudge and Netherland 2020), but did not report ET values. The exposure time needed to reduce biomass of submersed species can be an important factor when choosing a management strategy, and that is especially true for canal weeds such as rotala and hygrophila, where significant flow can reduce exposure time. A number of reports describe the effects of concentration–exposure times of herbicides such as endothall (e.g., Netherland et al. 1991, Slade et al. 2008, Mudge and Theel 2011, Gyselinck and Courter 2015, Hunt et al. 2015, Dugdale et al. 2019), flumioxazin (Bultemeier et al. 2009, Mudge et al. 2012), and other active ingredients (Getsinger and Netherland 1997, Getsinger et al. 2011). Most reveal that biomass reductions increase and concentration and exposure times increase in species such as horned pondweed (*Zannichellia palustris*) (Gyselinck and Courter 2015), sago pondweed (*Stuckenia pectinata*) (Slade et al. 2008), hydrilla, Eurasian watermilfoil (Netherland et al. 1991, Mudge and Theel 2011), and other submersed weeds, but some (e.g., Hunt et al. 2015 and others) also report that low concentrations are effective if long exposure times are achieved.

This concept—that concentration and exposure time can be adjusted to compensate for prevailing conditions without loss in efficacy—may apply to florpyrauxifen-benzyl as well as other herbicides. For example, Mudge and Netherland (2020) treated crested floatingheart with a range of concentrations over four exposure periods and reported that in most cases, biomass decreased as exposure time increased within a concentration. We had similar results with hygrophila; our ET_{50} and ET_{90} values showed that the exposure time needed to reduce hygrophila biomass declined as florpyrauxifen-benzyl concentration increased. This suggests that it may be possible to achieve good control of canal weeds subjected to flow by increasing herbicide concentration, and this is our rationale for presenting both EC and ET values for rotala and hygrophila. For example, if a quiescent waterbody is treated with florpyrauxifen-benzyl for submersed weed control, it is likely that good control will result from low herbicide concentrations, as exposure time will be high. Conversely, when a canal or other high-flow aquatic system is targeted for weed control, exposure time will probably be greatly reduced and beyond the control of the resource manager. Herbicide concentration, however, can be increased (if allowed by the label) to compensate for short exposure times.

We show that rotala is extremely susceptible to florpyrauxifen-benzyl, so management of this species is likely to succeed with virtually any concentration–exposure time scenario. Hygrophila, although sensitive, required greater concentrations and longer exposure times than did rotala. Recall that hygrophila EC values decreased as exposure time increased. For example, in Run 2, our calculations show that hygrophila exposed to florpyrauxifen-benzyl for 6 h would require a concentration of 61 μg L^{-1} for a 90% reduction in biomass, whereas plants exposed for 24 or 48 h would need concentrations of 22 or 15 μg L^{-1}, respectively, for the same level of control. Thus, florpyrauxifen-benzyl should be applied at higher concentrations (up to the label maximum of 48 μg L^{-1}) to manage hygrophila in canal systems because the reduced exposure time due to flow will increase EC. Conversely, if water to be treated is still or can be impounded to increase exposure time, it may be possible to decrease florpyrauxifen-benzyl concentration without loss in efficacy.

This research adds to the body of knowledge regarding florpyrauxifen-benzyl concentration–exposure time requirements and provides new information about the effects of this auxin on the submersed weeds rotala and hygrophila. This also provides guidance that can inform management decisions under different CET scenarios and can serve as a framework for selecting appropriate concentrations of florpyrauxifen-benzyl based on field conditions, especially in systems where flow can reduce exposure times.

SOURCES OF MATERIALS

[1]Osmocote Plus 15-9-12; Everris—an ILC Fertilizers Company, Dublin, OH 43017.

[2]ProcellaCOR SC; SePRO Corp., Carmel, IN 46032.

[3]HOBO Water Temperature Pro v2 Data Logger; Onset Computer Corp., Bourne, MA 02532.

[4]SAS Software Version 9.3; SAS Institute, Cary, NC 27513.

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