

INVESTIGATION OF HERBICIDE RESISTANCE IN KENTUCKY ITALIAN RYEGRASS POPULATIONS

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INTRODUCTION

Italian ryegrass (*Lolium perenne* L. ssp. *Multiflorum*) also known as annual ryegrass is a primary weed pest for Kentucky small grain growers. Italian ryegrass is a highly competitive winter annual grass species that can cause significant competition and yield decreases in small grains. Italian ryegrass has been documented to decrease wheat yields by up to 60% when occurring at high densities. Control of annual ryegrass in small grains can be difficult, do to similarities in species growth habit and herbicide tolerance.

Increased adoption of no-tillage practices has increased the number of herbicide applications used to control winter annual weeds such as Italian ryegrass. The increase in herbicide applications has resulted in increased selection pressure and prevalence of herbicide-resistant Italian ryegrass. On a global scale Italian ryegrass ranks within the top five weed species with resistance to the greatest number of sites of action. Italian ryegrass has conferred resistance to eight different sites of action and is only surpassed by annual bluegrass, barnyardgrass, and rigid ryegrass which happens to be of the same genus.

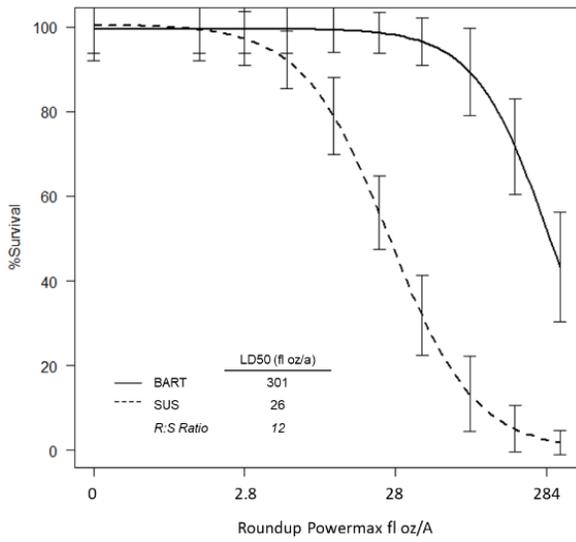
Italian ryegrass in Kentucky has previously been confirmed as resistant to mesosulfuron-methyl and pyroxsulam (ALS-inhibitors, SOA Group 2) and diclofop-methyl (ACCCase-inhibitor, SOA Group 1). In 2017 two separate populations of Italian ryegrass from Kentucky were submitted to the University of Kentucky weed science program for screening of herbicide resistance. The first population was suspected of glyphosate resistance and the second population contained two generations that were suspected of resistance to pinoxaden. The two populations were screened for resistance in greenhouses at the University of Kentucky Research

and Education Center in Princeton in the winter of 2017-2018.

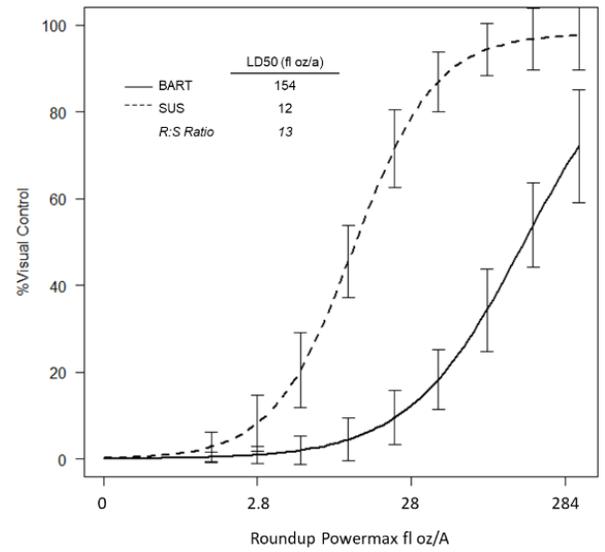
MATERIALS AND METHODS

The ryegrass seed sample suspected of glyphosate resistance contained one generation and was designated as “BART”. The population of ryegrass suspected as pinoxaden resistant contained two generations of seed that were designated as “WT2015” and “WT2017”. The two suspected ryegrass populations were screened in separate dose response studies in the greenhouse and each study was conducted twice. Within each dose response study the suspected resistant populations were screened against a known susceptible population. A cover crop ryegrass variety ‘Marshal’ was used as the known susceptible variety and is designated as “SUS”.

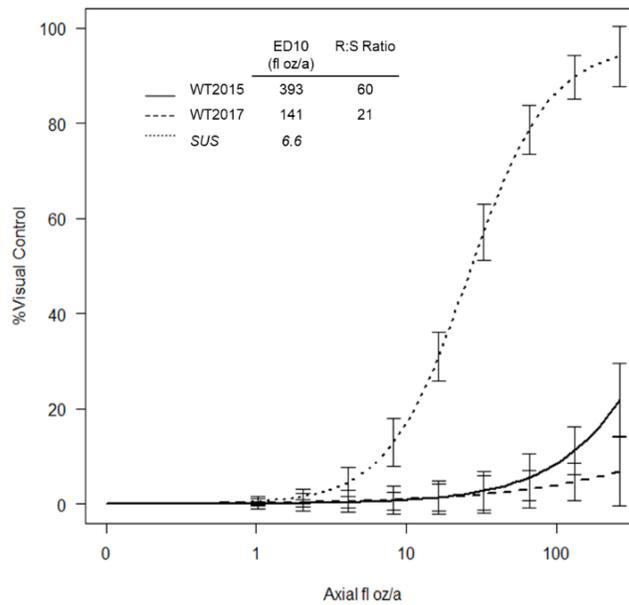
All ryegrass populations were planted in 4” greenhouse pots filled with a commercial grade potting mix. Ryegrass plants in the glyphosate dose response studies (BART and SUS populations) were thinned to three plants per pot following emergence, while plants in the pinoxaden studies (WT2015, WT2017, and SUS) were thinned to five plants per pot. Each population within a dose response study was subjected to 10 doses of glyphosate or pinoxaden respective of the study. Doses in the glyphosate response study ranged from 0 fl oz Roundup Powermax per acre to 350 fl oz Roundup Powermax per acre, with 22 fl oz Roundup Powermax per acre representing the 1X rate. Within the pinoxaden study doses ranged from 0 fl oz to 262 fl oz Axial XL per acre with 16 fl oz Axial XL per acre representing the 1X rate. Herbicide treatments were applied to the plants using an automated spray track spray in the greenhouse when plants reached the 1 to 2 tiller stage.



GRAPH 1. Percent survival dose response curves and ED50 values for BART and SUS ryegrass populations.



GRAPH 2. Visual control dose response curves and ED50 values for BART and SUS ryegrass populations.



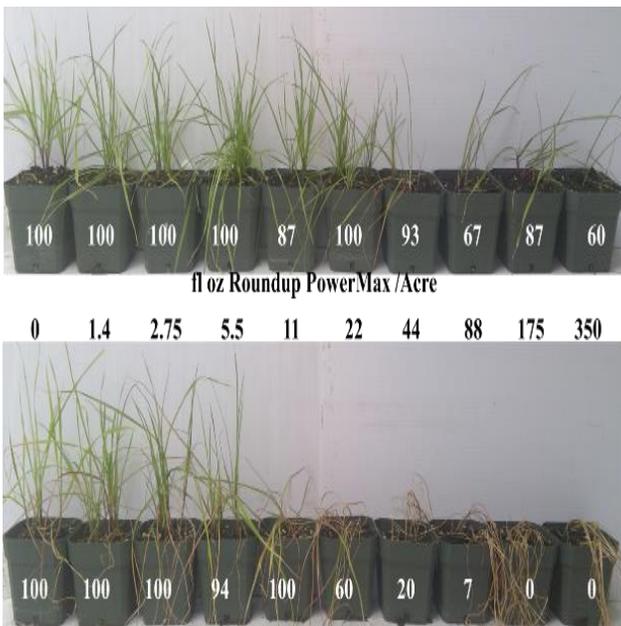
GRAPH 3. Visual control dose response curves and ED10 values for WT2015, WT2017, and SUS ryegrass populations

Following application control assessments were taken at 3 weeks and 4 weeks after application for the glyphosate dose response study and pinoxaden dose response study, respectively. A visual control rating was taken based on a 0 to 100 scale in which 0 represents no control or injury and 100 representing complete plant death. A count of surviving plants was also taken for a percentage survival assessment based on the known number of plants at time of application.

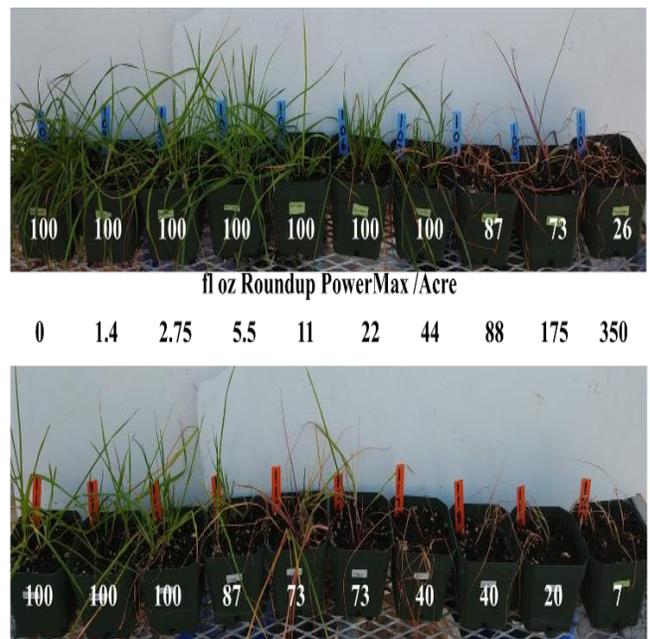
The dose response studies were setup in a factorial design with a randomized complete block layout and five replications. Dose response curves and estimated dose(ED) values were determined using the 'drc' package in R statistical software.

RESULTS
GLYPHOSATE DOSE RESPONSE STUDY

The BART population showed 26 to 60% survival at the highest rate of glyphosate of 350 fl oz Roundup Powermax as compared to the SUS which showed 0 to 7% survival (Pictures 1 & 2). ED50 values (Estimated Dose to control 50% of the population) based on survival percentage were 301 and 26 fl oz Roundup Powermax per acre for the BART and SUS populations, respectively (Graph 1). Visual control ratings resulted in an ED50 of 154 fl oz Roundup Powermax per acre for the BART population as compared to 12 fl oz Roundup Powermax per acre for the SUS population (Graph 2). When comparing the ED50 values of both the visual evaluations and survival percentages a difference of 12 to 13 fold is calculated. The results of this study indicates that a 12 to 13 fold dose is required to control the BART population as compared to a susceptible population.



Picture 1. Percentage ryegrass survival three weeks after application of Roundup Powermax in the first run of the glyphosate dose experiment.

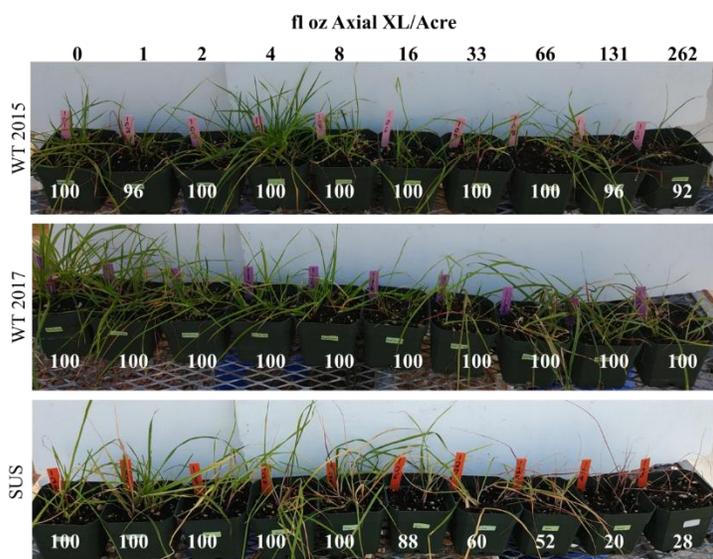


Picture 2. Percentage ryegrass survival three weeks after application of Roundup Powermax in the second run of the glyphosate dose experiment.

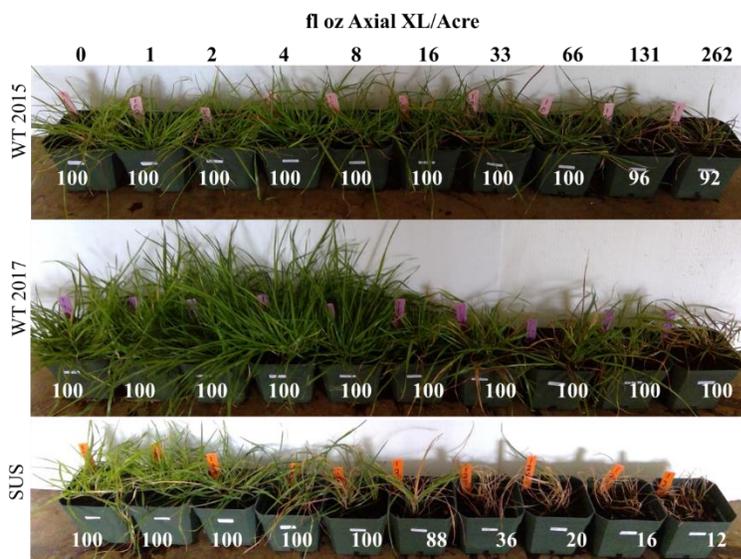
PINOXADEN DOSE RESPONSE STUDY

The WT2015 and WT2017 populations survived at a rate of 92 to 100% when receiving a 16X dosage of 262 fl oz Axial XL as compared to the SUS population which had 12 to 28% survival (Pictures 3 & 4). In the Pinoxaden dose response analysis an ED10 (Estimated Dose to control 10% of the population) was used rather than an ED50. The ED10 was used due to the fact that control of the WT2015 and WT2107 populations did not exceed 25% thus an ED above 25 would have to be extrapolated and potentially invalid.

Visual control ratings resulted in an ED10 of 393, 141, and 6.6 fl oz Axial XL per Acre for WT 2015, WT2017, and SUS, respectively (Graph 3). A resistance to susceptible ratio was calculated by comparing the ED10 of the WT2015 and WT2017 populations to the SUS population. The WT2015 populations had a 60 fold R to S ratio while the WT2017 population had a 21 fold R to S ratio. Results from this study indicate that a 21 to 60 fold dose of pinoxaden is required to control the WT2015 and WT2017 populations as compared to a susceptible population.



Picture 3. Percentage ryegrass survival four weeks after application of Axial XL in the first run of the pinoxaden dose experiment.



Picture 4. Percentage ryegrass survival four weeks after application of Axial XL in the second run of the pinoxaden dose experiment.

CONCLUSIONS

Results from the glyphosate dose response studies confirms the BART population as a glyphosate resistant population. This is the first population of ryegrass to be confirmed as glyphosate resistant in the state of Kentucky. Glyphosate-resistant ryegrass has been confirmed in seven states previously including bordering states of Tennessee and Arkansas. Further studies are to be conducted to confirm reproduction

of the resistance traits in future generations of this population.

Results from the pinoxaden dose response studies confirm a likely resistant population of ryegrass from two generations, although further work needs to be conducted with adjusted rates to achieve appropriate ED50 values. Pinoxaden resistant ryegrass has previously been confirmed in Arkansas and is suspected in many of the surrounding states.

The potential of loss of these two herbicides for control of Italian ryegrass is significant, especially for small grains growers. Pinoxaden is a primary post-emergence herbicide for many growers who are controlling ALS-resistant ryegrass, the loss of this tool eliminates all possible postemergence herbicides for this weed. While preemergence herbicide options are available, the loss of any tool in weed control is significant. The confirmation of glyphosate-resistant ryegrass in Kentucky is significant not only for small grain growers, but also corn and soybean farmers who rely on glyphosate for weed control. While there are alternate options to glyphosate for ryegrass control, again the loss of a herbicide as a weed control tool is

significant especially in a weed species that is globally known for conferring resistance to multiple herbicide sites of action.

FUTURE RESEARCH

As indicated previously the need for an adjusted pinoxaden dose-response is needed to appropriately calculate ED50 values for WT2015 and WT2017. Also further experiments screening for other herbicide sites of action are needed on all three populations to confirm potential multiple site of action resistance in each population.