

# Portfolio Presentation

## A Hierarchical Zero-Inflated Gamma Analysis of Chemical Efficacy on Soil-Borne Pathogens in Soybean

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# Introduction

- Soybean is a major food and oilseed crop, valued for its high protein content and use in both human and livestock diets.
- In 2013, global soybean production reached 268 million metric tons across 70 countries, led by the U.S. and Brazil (each 31%), and Argentina (19%).
- By 2024/2025 (USDA), Brazil, USA, and Argentina are expected to account for 80% of global soybean output, with Brazil alone producing 40%.
- Soybean yield losses in US vary annually due to environmental conditions, farming practices, and varietal disease resistance.
- In 2022, 75% of U.S. soybean production came from 13 Midwestern and Northeastern states, which also suffered 71.3% of the nation's total yield loss.
- Seedling diseases due to Fusarium, Pythium, Phomopsis and Rhizoctonia are one of the major causes of soybeans loss in 2022

## Nebraska Context

- In Nebraska, the largest producer of beef and pork, Soybeans is one of the major ingredient used in the beef and pork production.
- However, Nebraska experiences an estimated annual loss exceeding 9 million bushels due to pathogenic organisms.
- Pathogens such as *Colletotrichum truncatum*, *Fusarium virguliforme*, *Macrophomina phaseolina*, *Pythium irregulare*, *Rhizoctonia solani*, *Sclerotinia sclerotiorum*, are major contributors to soybean seedling diseases, which leads to decrease in soybeans yield.

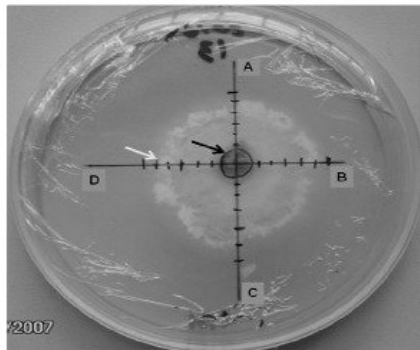
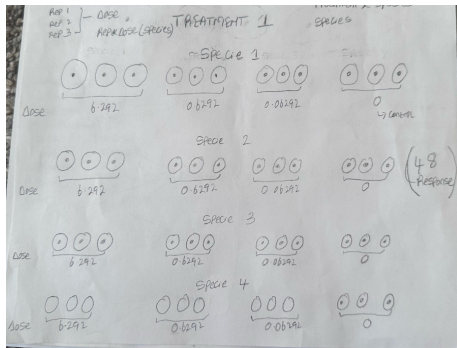
## Study Objective

- Thus, this study seek to investigate the efficacy of chemical applications in controlling soil-borne pathogens of soybean and
- look at the tolerance of the various pathogens to fungicide at different dose level.

# Materials and Method

## Experimental design

- The fungicide used was incorporated into the growth medium (petri dish) to ensure there was homogeneous distribution of this chemical across the different fungal species.
- This setup is called Poisoned food and it mimics how the different fungicide would react with the pathogens in natural soil conditions.



# Materials and Method

## Fungal Species:

- *Diaporthe longicolla*
- *Fusarium oxysporum*
- *Fusarium solani*
- *Rhizoctonia solani*

## Fungicide Treatments and Dosages ( $\mu\text{g/ml}$ ):

	DelaroComplete (Proth+Trif+Fluop)	Endura (Boscalid)	Quadris (Azoxystrobin)	Topguard (Flutriafol)	Topguard EQ (Flut+Azoxys)
High	6.292	4.011	6.1036	5.7975	4.9128
Medium	0.6292	0.4011	0.61036	0.57975	0.49128
Low	0.06292	0.04011	0.061036	0.057975	0.049128
Control	0	0	0	0	0

*Note: Each fungal species get each fungicide treatment at all dosage levels.*

# Two-Part Model: Zero-Inflated Gamma

**Motivation:** The radial growth rate of pathogens is semicontinuous—many zeros and a positively skewed distribution. Modelling semicontinuous data with true zero is done in two steps

- First part models the probability that an outcome is non-zero
- Second part models the value of an outcome given it is greater than zero

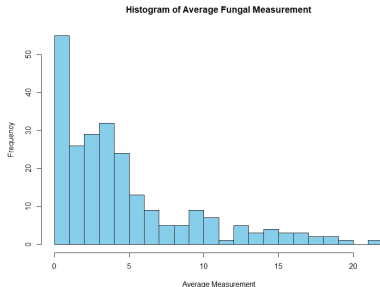
## Model Specification:

$$f(y) = (1 - p_\beta)I(y = 0) + p_\beta G_\theta(y \mid y > 0)$$

- $\beta$ : parameter used in modelling the probability of positive response
- $\theta$ : mean and dispersion parameter of the conditional distribution of the positive response
- $p_\beta = P(Y_i > 0 \mid X_i)$ , probability of positive response, modeled via logistic regression  
 $\text{logit}(p_\beta) = X_i\beta$
- $G_\theta(y \mid y > 0)$ : Distribution of positive responses, modeled using Gamma regression with log link,

$$Y_i \sim \Gamma(\exp(X_i\tau), \nu)$$

$\beta$  and  $\tau$  is the parameter associated with  $X$  in the logistic and Gamma portion of the model.



# Model Structure

## Split-Plot Factor Nested in Whole-Plot Factor

Source of Variation (SV)	df
Treatment	$(5 - 1) = 4$
Dose(Treatment)	$(4 - 1) \times 5 = 15$
Species	$(4 - 1) = 3$
Treatment * Species	$(5 - 1) \times (4 - 1) = 12$
Species * Dose(Treatment)	$3 \times 15 = 45$
Error	$(3 - 1) \times (4 \times 4 \times 5) = 160$

## Model Specification:

$$\log(E[Y_{ijkl} | Y_{ijkl} > 0]) = \mu + \text{Species}_i + \text{Treatment}_j + (\text{Species} \times \text{Treatment})_{ij} + (1 | \text{Dose:Treatment}) + (1 | \text{Species:Dose:Treatment}) \quad (1)$$

## Where:

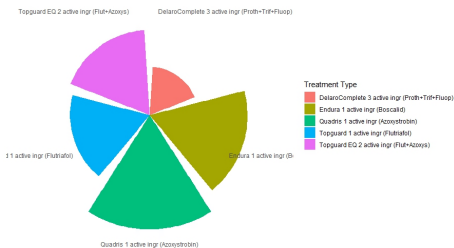
- $Y_{ijkl}$ : Average measurement for the  $i^{\text{th}}$  species,  $j^{\text{th}}$  treatment,  $k^{\text{th}}$  dose,  $l^{\text{th}}$  dish
- $\mu$ : Baseline log mean fungal growth
- $e_{ijkl} \sim \text{Gamma}(\alpha, \beta)$ : Error term from the Gamma distribution
- $(1 | \text{Dose:Treatments})$ : Random variability among doses within treatments.
- $(1 | \text{Species:Dose:Treatments})$ : Random variability for species across dose-treatment combinations.



# Exploratory Data Analysis

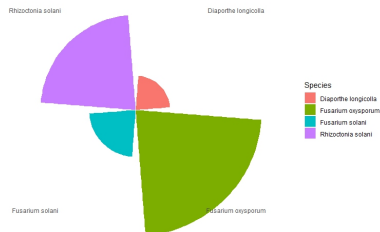
This section provides a visual summary of the data to understand the distribution, of average fungal growth across treatment and species.

Average Measurement by Treatment



(a) Fungal Growth by Treatment

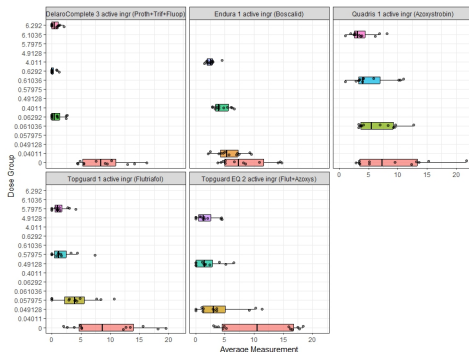
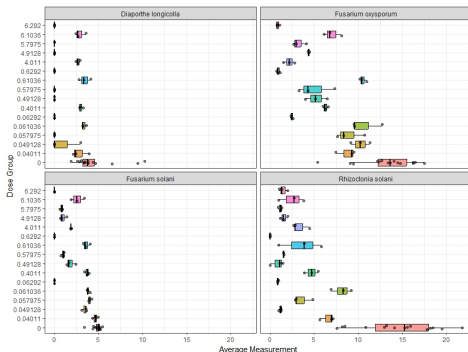
Average Measurement by Species



(b) Fungal Growth by Species

# Exploratory Data Analysis

This section provides a visual summary of the distribution of average fungal growth measurements across different fungicide dose levels, split by fungal species and treatment.



(c) Fungal Growth by Species and Dose

(d) Fungal Growth by Treatment and Dose

# Advanced Analysis: Random and Fixed Effects Structures

## Random Effects:

- Explains growth variability due to hierarchical structure:

```
Random effects:
Conditional model:
Groups              Name      Variance Std.Dev.
Dose:Treatments     (Intercept) 0.3823  0.6183
Species:Dose:Treatments (Intercept) 0.1007  0.3173
Number of obs: 239, groups: Dose:Treatments, 20; Species:Dose:Treatments, 80

Dispersion estimate for Gamma family (sigma^2): 0.0586
```

## Fixed Effects from Gamma Model:

- Marginal species  $\times$  treatment interaction ( $p > 0.05$ ).

```
Analysis of Deviance Table (Type III Wald chisquare tests)

Response: Avg_Measurement
              Chisq Df Pr(>Chisq)
(Intercept)    1.6882  1    0.19384
Species        1.9564  3    0.58150
Treatments      4.8059  4    0.30780
Species:Treatments 18.7860 12    0.09382 .
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

# Advanced Analysis: Interactions and Pairwise Comparisons

## Species × Treatment Interactions: Pairwise comparisons between species and treatment:

Table 3: Interaction between Species and Treatments on Average Growth

Species	Treatments	df	Avg Growth	SE	LCL	UCL	p
Diaporthe longicolla	(Proth+Trif+Fluop)	3	1.96	1.01	0.711	5.38	0.4564
Fusarium oxysporum	(Proth+Trif+Fluop)	3	2.2	1.0	0.614	4.081	0.3657
Fusarium solani	(Proth+Trif+Fluop)	3	1.2	0.618	0.435	3.29	0.8737
Rhizoctonia solani	(Proth+Trif+Fluop)	3	2.05	0.778	0.975	4.31	0.3553
Diaporthe longicolla	(Boscalid)	1	3.27	1.1	1.651	6.5	0.9614
Fusarium oxysporum	(Boscalid)	1	6.52	1.31	3.255	10.65	0.1217
Fusarium solani	(Boscalid)	1	5.63	1.3	2.85	10.2	0.1703
Rhizoctonia solani	(Boscalid)	1	5.61	1.19	2.874	11.27	0.2824
Diaporthe longicolla	(Azoxystrobin)	1	3.27	1.1	1.631	6.55	0.9614
<b>Fusarium oxysporum</b>	<b>(Azoxystrobin)</b>	<b>1</b>	<b>9.9</b>	<b>3.51</b>	<b>4.941</b>	<b>19.83</b>	<b>0.0115</b>
Fusarium solani	(Azoxystrobin)	1	3.61	1.28	1.803	7.24	0.8737
Rhizoctonia solani	(Azoxystrobin)	1	5.99	2.2	1.184	11.99	0.1632
Diaporthe longicolla	(Flutriafol)	1	1.05	0.52	0.399	2.77	0.5177
Fusarium oxysporum	(Flutriafol)	1	6.61	2.34	2.3	13.25	0.0894
Fusarium solani	(Flutriafol)	1	3.9	0.77	2.03	6.4	0.3336
Rhizoctonia solani	(Flutriafol)	1	1.92	0.646	0.905	4.04	0.2261
Diaporthe longicolla	(Flut+Azoxys)	2	1.89	0.785	0.835	4.27	0.3336
Fusarium oxysporum	(Flut+Azoxys)	2	2.87	0.84	1.184	4.75	0.0913
Fusarium solani	(Flut+Azoxys)	2	3.01	0.87	1.055	6.05	0.1187
Rhizoctonia solani	(Flut+Azoxys)	2	2.6	0.926	1.298	5.23	0.6934

Table 4: Pairwise Differences in Fungal Growth Under Different Treatments

Pairwise Comparison	Ratio	SE	LCL	UCL	p-value
Diaporthe longicolla (Azoxystrobin) / Fusarium oxysporum (Azoxystrobin)	0.33	0.081	0.1384	0.787	0.0011
Fusarium oxysporum (Azoxystrobin) / Fusarium solani (Azoxystrobin)	2.741	0.672	1.1489	6.537	0.0063
Fusarium oxysporum (Azoxystrobin) / Diaporthe longicolla (Flutriafol)	9.412	5.73	1.0886	81.381	0.0313
Diaporthe longicolla (Flutriafol) / Fusarium oxysporum (Flutriafol)	0.159	0.0674	0.0354	0.714	0.0024
Fusarium oxysporum (Flutriafol) / Fusarium solani (Flutriafol)	3.26	0.8	1.3664	7.776	0.0003
Diaporthe longicolla (Flut+Azoxys) / Fusarium oxysporum (Flut+Azoxys)	0.236	0.0773	0.0736	0.754	0.0018
Fusarium oxysporum (Flut+Azoxys) / Fusarium solani (Flut+Azoxys)	3.378	0.829	1.416	8.058	0.0001
Fusarium oxysporum (Flut+Azoxys) / Rhizoctonia solani (Flut+Azoxys)	3.075	0.759	1.2826	7.374	0.0009

Only extracted significant pairwise difference

- Significant interaction between *Fusarium oxysporum* and Quadris 1 active (Azoxystrobin)
- Marginal significant interactions between *Fusarium oxysporum* and Topguard 1 active (Flutriafol), Topguard EQ 2 active (Flutriafol + Azoxystrobin)

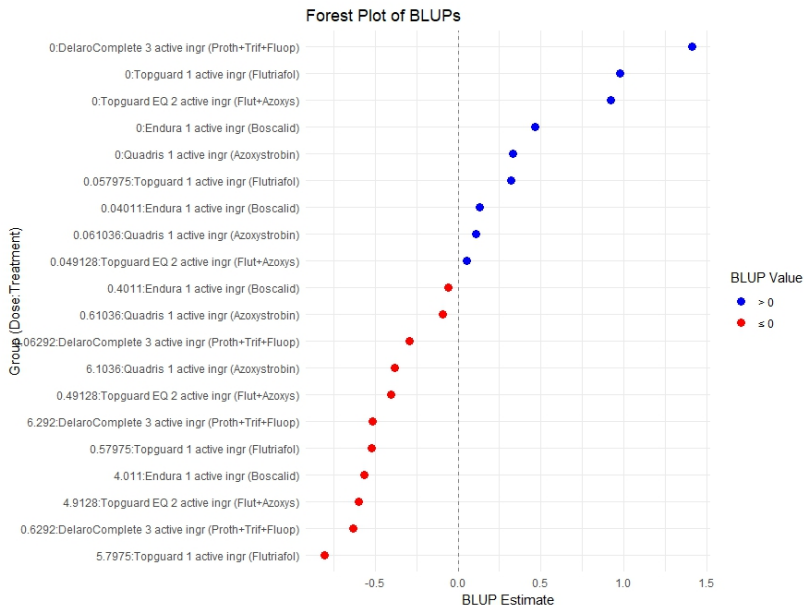
# Best Linear Unbiased Prediction: Dose within Treatment

Table 5: Best Linear Unbiased Prediction for Dose nested within Treatment

Dose:Treatment	BLUP
0:DelaroComplete 3 active (Proth+Trif+Fluop)	1.4118
0.06292:DelaroComplete 3 active (Proth+Trif+Fluop)	-0.2938
0.6292:DelaroComplete 3 active (Proth+Trif+Fluop)	-0.6315
6.292:DelaroComplete 3 active (Proth+Trif+Fluop)	-0.5171
0:Endura 1 active (Boscalid)	0.4644
0.04011:Endura 1 active (Boscalid)	0.1312
0.4011:Endura 1 active (Boscalid)	-0.0614
4.011:Endura 1 active (Boscalid)	-0.5659
0:Quadris 1 active (Azoxystrobin)	0.3327
0.061036:Quadris 1 active (Azoxystrobin)	0.1095
0.61036:Quadris 1 active (Azoxystrobin)	-0.0933
6.1036:Quadris 1 active (Azoxystrobin)	-0.3806
0:Topguard 1 active (Flutriafol)	0.9781
0.057975:Topguard 1 active (Flutriafol)	0.3181
0.57975:Topguard 1 active (Flutriafol)	-0.5203
5.7975:Topguard 1 active (Flutriafol)	-0.8084
0:Topguard EQ 2 active (Flut+Azoxys)	0.9204
0.049128:Topguard EQ 2 active (Flut+Azoxys)	0.0501
0.49128:Topguard EQ 2 active (Flut+Azoxys)	-0.4068
4.9128:Topguard EQ 2 active (Flut+Azoxys)	-0.5986

- **DelaroComplete:** Control dose has high growth, but growth decreases as dose increase.
- **Endura, Quadris, Topguard, and Topguard EQ 2:** Control and low dose was predicted to increase growth above average, while other doses decrease growth.

# Best Linear Unbiased Prediction: Dose within Treatment



# BLUP: Species crossed with dose nested within treatment

Table 6: Best Linear Unbiased Prediction for Dose nested within Treatment for *Diaporthe longicolla*

Dose:Treatment				BLUP
0:DelaroComplete	3	active		-0.000616511
(Proth+Trif+Fluop)				
0.06292:DelaroComplete	3	active		0
(Proth+Trif+Fluop)				
0.6292:DelaroComplete	3	active		0
(Proth+Trif+Fluop)				
6.292:DelaroComplete	3	active		0
(Proth+Trif+Fluop)				
0:Endura 1 active (Boscalid)				-0.041562794
0.04011:Endura 1 active (Boscalid)				-0.237818396
0.4011:Endura 1 active (Boscalid)				-0.021039568
4.011:Endura 1 active (Boscalid)				0.298317876
0:Quadris 1 active (Azoxystrobin)				-0.282390186
0.061036:Quadris 1 active (Azoxystrobin)				-0.062961318
0.61036:Quadris 1 active (Azoxystrobin)				0.121451677
6.1036:Quadris 1 active (Azoxystrobin)				0.221821793
0:Topguard 1 active (Flutriafol)				-0.000574947
0.057975:Topguard 1 active (Flutriafol)				0
0.57975:Topguard 1 active (Flutriafol)				0
5.7975:Topguard 1 active (Flutriafol)				0
0:Topguard EQ 2 active (Flut+Azoxys)				-0.271722003
0.049128:Topguard EQ 2 active (Flut+Azoxys)	EQ	2	active	0.265117379
(Flut+Azoxys)				
0.49128:Topguard EQ 2 active (Flut+Azoxys)	EQ	2	active	0
(Flut+Azoxys)				
4.9128:Topguard EQ 2 active (Flut+Azoxys)				0

- DelaroComplete and Topguard 1: decrease at control but stable from low to high dose;
- Endura 1 active: decrease from control to medium but increase at high dose.
- Quadris: decrease at control and low, increase at medium and high dose.
- Topguard EQ: decrease at control, increase at low dose, stable at medium and high dose.

# BLUP: Species crossed with dose nested within treatment

Table 7: Best Linear Unbiased Prediction for Dose nested within Treatment for *Fusarium oxysporum*

Dose:Treatment			BLUP
0:DelaroComplete	3	active	0.106100059
(Proth+Trif+Fluop)			
0.06292:DelaroComplete	3	active	0.355692318
(Proth+Trif+Fluop)			
0.6292:DelaroComplete	3	active	-0.166327429
(Proth+Trif+Fluop)			
6.292:DelaroComplete	3	active	-0.299732171
(Proth+Trif+Fluop)			
0:Endura 1 active (Boscalid)			0.278496151
0.04011:Endura 1 active (Boscalid)			0.136230476
0.4011:Endura 1 active (Boscalid)			0.019989513
4.011:Endura 1 active (Boscalid)			-0.436805234
0:Quadris 1 active (Azoxystrobin)			-0.129392723
0.061036:Quadris 1 active (Azoxystrobin)			-0.034205375
0.61036:Quadris 1 active (Azoxystrobin)			0.128827724
6.1036:Quadris 1 active (Azoxystrobin)			0.032736257
0:Topguard 1 active (Flutriafol)			-0.268141692
0.057975:Topguard 1 active (Flutriafol)			-0.017637166
0.57975:Topguard 1 active (Flutriafol)			0.200686125
5.7975:Topguard 1 active (Flutriafol)			0.083511652
0:Topguard EQ 2 active (Flut+Azoxys)			-0.153415918
0.049128:Topguard EQ 2 active (Flut+Azoxys)	EQ	2 active	0.164391125
0.49128:Topguard EQ 2 active (Flut+Azoxys)	EQ	2 active	-0.014750217
4.9128:Topguard EQ 2 active (Flut+Azoxys)	EQ	2 active	0.004023557

- DelaroComplete: increase at control and low, decrease at medium and high.
- Endura 1 active: increase at control, low and high, decrease at high.
- Quadris and Topguard 1: decrease at control and low but decrease at medium and high.
- Topguard EQ: decrease at control and medium, increase at low and high



# BLUP: Species crossed with dose nested within treatment

Table 8: Best Linear Unbiased Prediction for Dose nested within Treatment for *Fusarium solani*

Dose:Treatment			BLUP
0:DelaroComplete	3	active	-0.000625527
(Proth+Trif+Fluop)			
0.06292:DelaroComplete	3	active	0
(Proth+Trif+Fluop)			
0.6292:DelaroComplete	3	active	0
(Proth+Trif+Fluop)			
6.292:DelaroComplete	3	active	0
(Proth+Trif+Fluop)			
0:Endura 1 active (Boscalid)			-0.13874485
0.04011:Endura 1 active (Boscalid)			0.110523464
0.4011:Endura 1 active (Boscalid)			0.080927519
4.011:Endura 1 active (Boscalid)			-0.05479903
0:Quadris 1 active (Azoxystrobin)			-0.088966249
0.061036:Quadris 1 active (Azoxystrobin)			-0.04005418
0.61036:Quadris 1 active (Azoxystrobin)			0.078676058
6.1036:Quadris 1 active (Azoxystrobin)			0.048275351
0:Topguard 1 active (Flutriafol)			-0.062226152
0.057975:Topguard 1 active (Flutriafol)			0.308842891
0.57975:Topguard 1 active (Flutriafol)			-0.158526749
5.7975:Topguard 1 active (Flutriafol)			-0.089786457
0:Topguard EQ 2 active (Flut+Azoxys)			-0.160570939
0.049128:Topguard EQ 2 active (Flut+Azoxys)	2	active	0.272726683
0.49128:Topguard EQ 2 active (Flut+Azoxys)	2	active	0.124419505
4.9128:Topguard EQ 2 active (Flut+Azoxys)	2	active	-0.236335868

- DelaroComplete: decrease at control but stable from low to high dose.
- Endura 1 active and Topguard EQ 2: decrease at control and high, increase at low and medium.
- Quadris: decrease at control and low, increase at medium and high.
- Topguard 1: decrease at control, medium and high, increase at medium.

# BLUP: Species crossed with dose nested within treatment

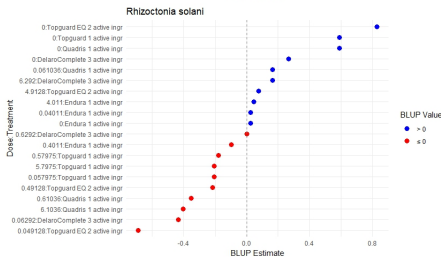
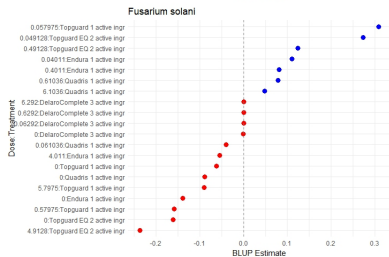
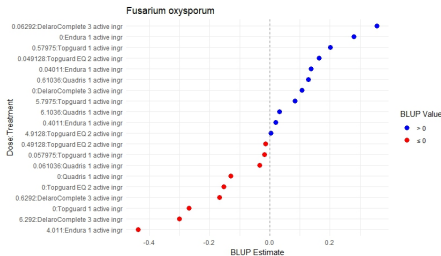
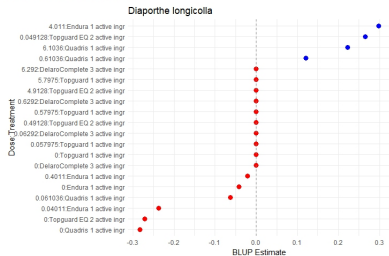
Table 9: Best Linear Unbiased Prediction for Dose nested within Treatment for *Rhizoctonia solani*

Dose:Treatment			BLUP
0:DelaroComplete (Proth+Trif+Fluop)	3	active	0.266911298
0.06292:DelaroComplete (Proth+Trif+Fluop)	3	active	-0.433046666
0.6292:DelaroComplete (Proth+Trif+Fluop)	3	active	0
6.292:DelaroComplete (Proth+Trif+Fluop)	3	active	0.163567614
0:Endura 1 active (Boscalid)			0.02410454
0.04011:Endura 1 active (Boscalid)			0.025606511
0.4011:Endura 1 active (Boscalid)			-0.09605766
4.011:Endura 1 active (Boscalid)			0.044278398
0:Quadris 1 active (Azoxystrobin)			0.588356584
0.061036:Quadris 1 active (Azoxystrobin)			0.166050995
0.61036:Quadris 1 active (Azoxystrobin)			-0.353527076
6.1036:Quadris 1 active (Azoxystrobin)			-0.403055423
0:Topguard 1 active (Flutriafol)			0.588507333
0.057975:Topguard 1 active (Flutriafol)			-0.20745378
0.57975:Topguard 1 active (Flutriafol)			-0.179171739
5.7975:Topguard 1 active (Flutriafol)			-0.206618072
0:Topguard EQ 2 active (Flut+Azoxys)			0.828083859
0.049128:Topguard (Flut+Azoxys)	EQ	2 active	-0.689032837
0.49128:Topguard (Flut+Azoxys)	EQ	2 active	-0.216787688
4.9128:Topguard EQ 2 active (Flut+Azoxys)			0.074685212

- DelaroComplete: increase at control and high, decrease at low, stable at medium.
- Endura 1 active: increase at control, low and high, decrease at medium.
- Quadris: increase at control and low, decrease at medium and high.
- Topguard 1: increase at control, decrease at low, medium and high.
- Topguard EQ: increase at control and high, decrease at low, and medium.

# BLUP: Species crossed with dose nested within treatment

Forest Plot of BLUPs by Species



# Conclusion

- *F. oxysporum* differed significantly in growth response compared to species: *F. solani*, *D. longicolla*, *R. solani*) under the following treatment **Quadris 1 active** (Azoxystrobin), **Topguard 1 active** (Flutriafol), and **Topguard EQ 2 active** (Flut+Azoxys) .
- At control dose, delarocomplete treatment has the highest growth, followed by Topguard 1 active, Topguard EQ 2 active, Endura 1 active, Quadris 1 active.
- Topguard 1 active, Topguard EQ 2 active, Endura 1 active, Quadris 1 active continued to grow at low dose but delarocomplete decreased in growth.
- for species crossed with dose nested in treatments, responses varied by species and fungicide, indicating strong species-specific interaction patterns.
- Notable differences for *F. oxysporum*, which showed variable trends depending on treatment and dose.
- Overall, effective fungal control depends on both selecting the right treatment and optimizing dose for the specific species involved.

# Questions?

# References

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