Influence of Ginsenosides on the Pathogenicity of *Ilyonectria*

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UNIVERSITY · CANADA

Ginseng and Ginseng Replant Disease (GRD)

- American ginseng (*Panax quinquefolius*) roots used in Traditional Chinese Medicine
- Subject to GRD when planted in a former ginseng garden
- Severe root rot resulting in poor germination and poor stand establishment

Image edited by Jessica Sinka

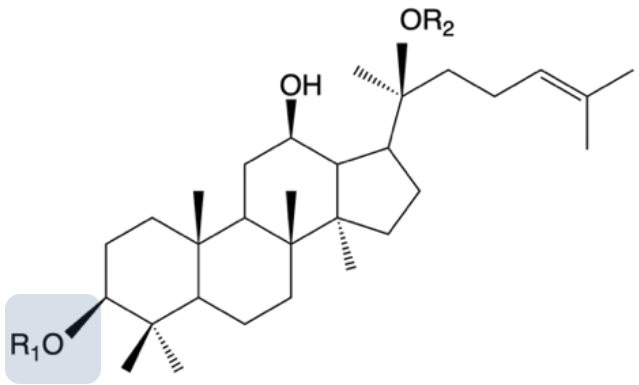
Contributors to GRD

Pathogens

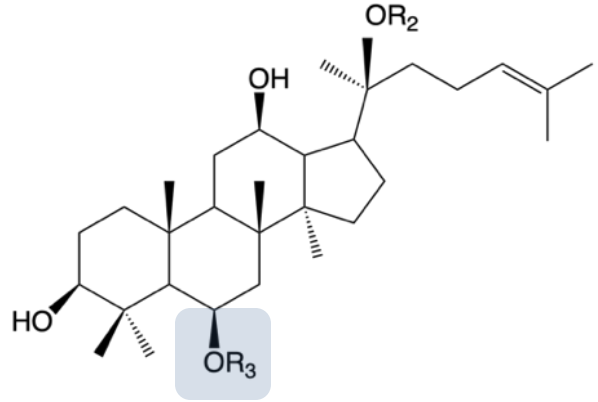
- *Fusarium* spp., *Globisporangium* (*Pythium)* spp., *Phytophthora* spp., *Ilyonectria* spp.
- Primarily *Ilyonectria mors-panacis*

Ginsenosides

- Triterpenoid saponins



Protopanaxadiol (PPD)



Protopanaxatriol (PPT)

Contributors to GRD

Ilyonectria-ginsenosides interaction ?

Pathogens

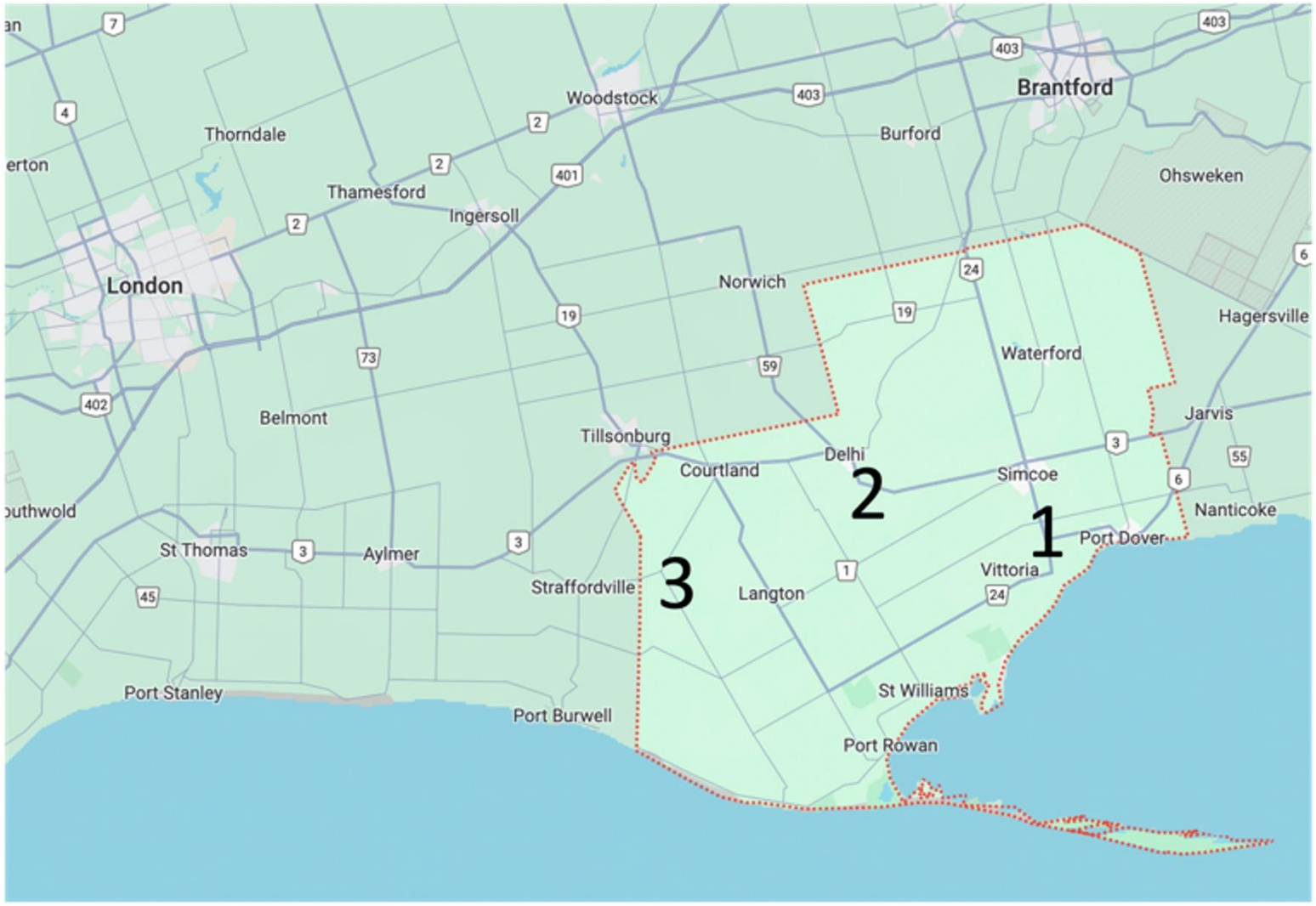
- *Fusarium* spp., *Globisporangium* (*Pythium)* spp., *Phytophthora* spp., *Ilyonectria* spp.
- Primarily *Ilyonectria mors-panacis*

Ginsenosides

- Triterpenoid saponins
- Fungitoxic to some organisms and stimulants of others

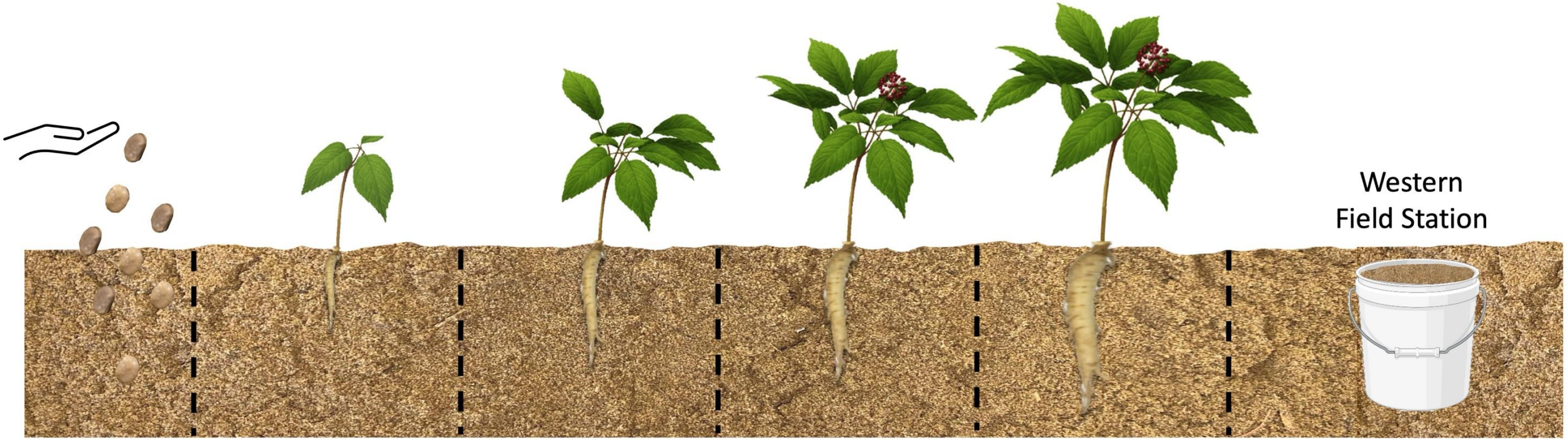
Do ginsenosides influence the pathogenicity of *Ilyonectria*?

Are ginsenosides present in ginseng garden soils?



Norfolk County, Ontario

Are ginsenosides present in ginseng garden soils?



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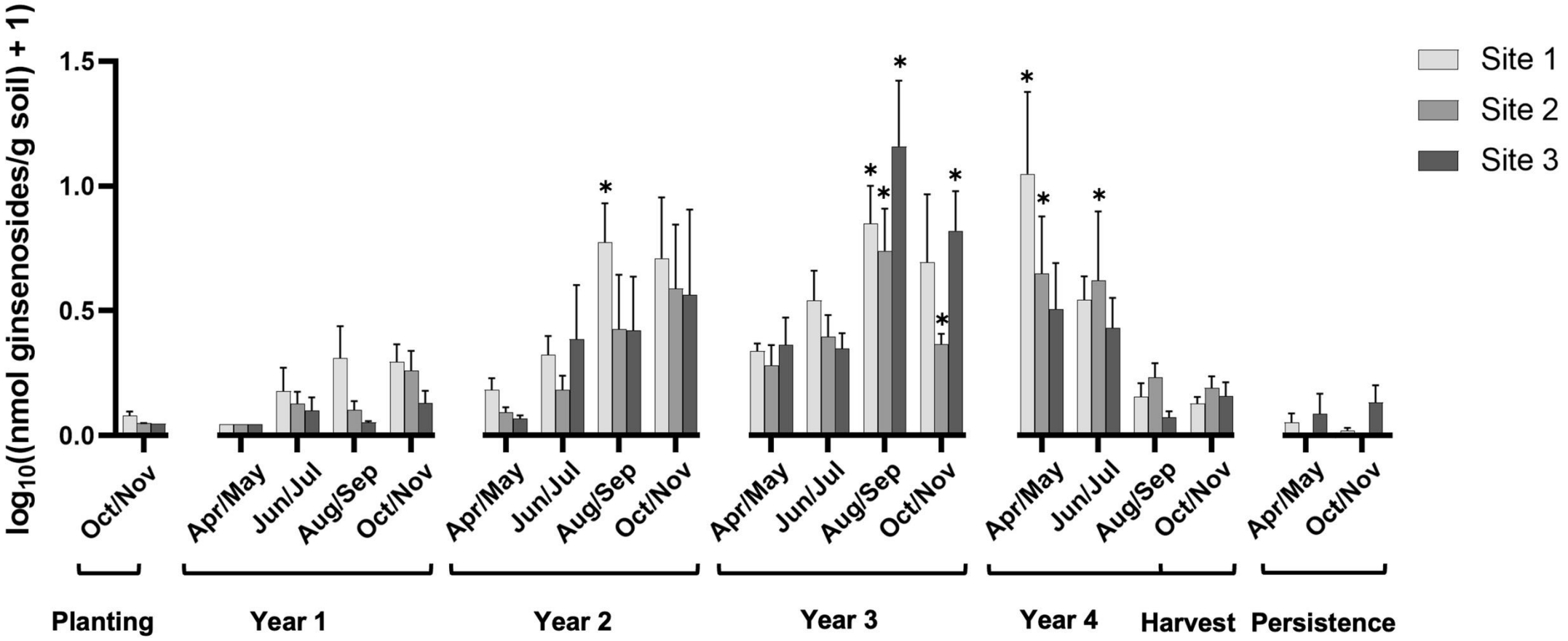
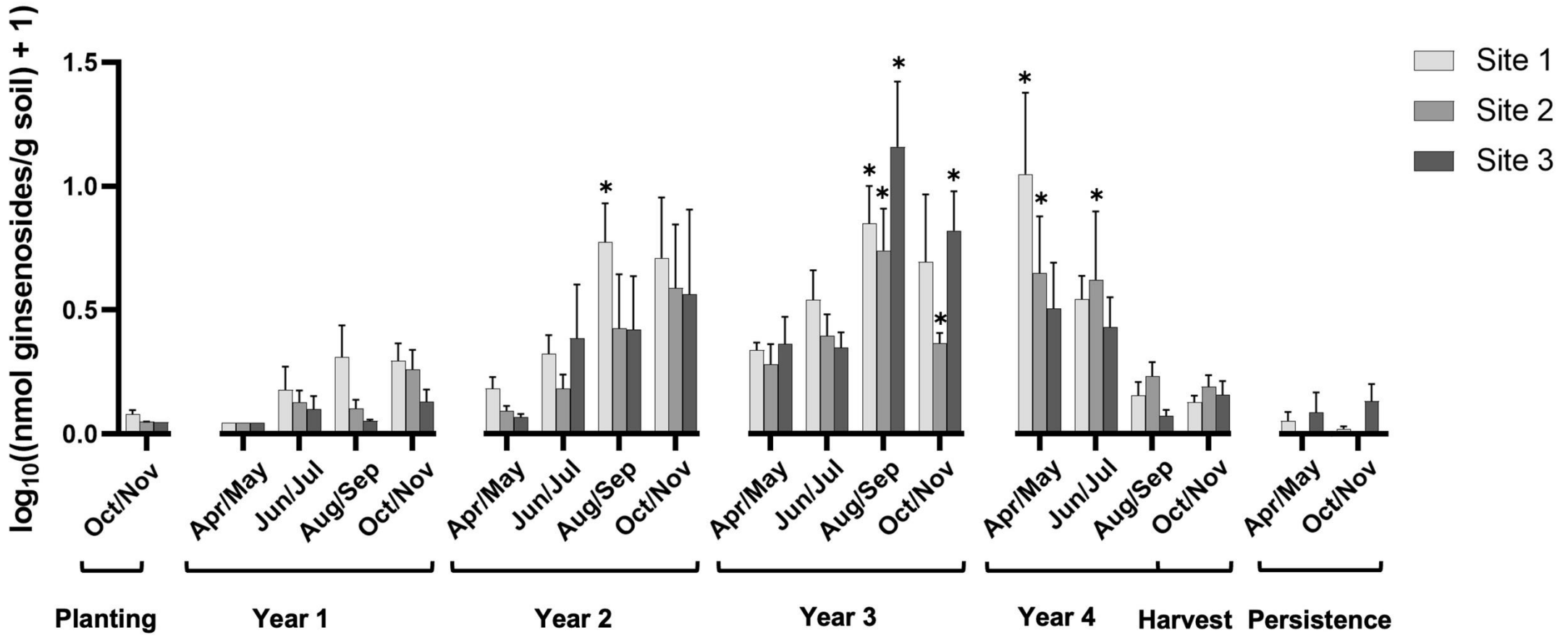


*p<0.05, statistical analysis using Friedman test, with Dunn's post-hoc test

Are ginsenosides present in ginseng garden soils?



Ginsenosides accumulate in **soils** annually for the first three years but, decline during the fourth year.

Ginsenosides are likely **not a direct contributor** to GRD and instead, establish the conditions for GRD to occur.

Do Ginsenosides Influence the Pathogenicity of *Ilyonectria*?

Objective 1

Determine disease severity in ginseng roots inoculated with different isolates of *Ilyonectria* with and without **previous exposure to ginsenosides**.

Objective 2

Determine whether different isolates of *Ilyonectria* **metabolize ginsenosides** differently and correlate the findings with virulence results from objective 2.

Objective 1

Determine disease severity in ginseng roots inoculated with different isolates of *Ilyonectria* with and without **previous exposure to ginsenosides**.

Ilyonectria Number	Species	Host Plant	Location
I.sp.1	*C. destructans*	*Pseudotsuga menziesii* (Douglas fir)	British Columbia, Canada
I.sp.2	*C. destructans*	*Picea glauca* (White Spruce)	Quebec, Canada
I.sp.3	*C. destructans*	***Panax quinquefolius* (American ginseng)**	Ontario, Canada
I.sp.4	*C. destructans*	*Poa pratensis* (Blue grass)	Alberta, Canada
I.sp.5	*C. destructans*	*Pseudotsuga menziesii* (Douglas fir)	British Columbia, Canada
I.sp.6	*C. destructans*	*Prunus cerasus* (Montomorency sour cherry)	Ontario, Canada
I.sp.7	*C. destructans*	***Panax quinquefolius* (American ginseng)**	Ontario, Canada
I.sp.8	*C. destructans*	***Panax quinquefolius* (American ginseng)**	Ontario, Canada
I.sp.9	*C. destructans*	***Panax* sp.**	Nagano Prefecture, Japan
I.sp.10	*C. destructans*	***Panax* sp.**	Nagano Prefecture, Japan
I.sp.11	*C. destructans*	***Panax quinquefolius* (American ginseng)**	Ontario, Canada
I.sp.12	*C. destructans*	***Panax quinquefolius* (American ginseng)**	Ontario, Canada

Cabral et al., (2016) noted different *Ilyonectria* species identification for several of the same isolates.

Objective 1

Determine disease severity in ginseng roots inoculated with different isolates of *Ilyonectria* with and without **previous exposure to ginsenosides**.

***Ilyonectria* Number**	**Species**	**Host Plant**	**Location**
I.sp.1	*I. rufa*	*Pseudotsuga menziesii* (Douglas fir)	British Columbia, Canada
I.sp.2	*I. rufa*	*Picea glauca* (White Spruce)	Quebec, Canada
I.sp.3	***I. mors-panacis***	***Panax quinquefolius* (American ginseng)**	Ontario, Canada
I.sp.4	***I. mors-panacis***	*Poa pratensis* (Blue grass)	Alberta, Canada
I.sp.5	*I. rufa*	*Pseudotsuga menziesii* (Douglas fir)	British Columbia, Canada
I.sp.6	*I. robusta*	*Prunus cerasus* (Montomorency sour cherry)	Ontario, Canada
I.sp.7	***I. mors-panacis***	***Panax quinquefolius* (American ginseng)**	Ontario, Canada
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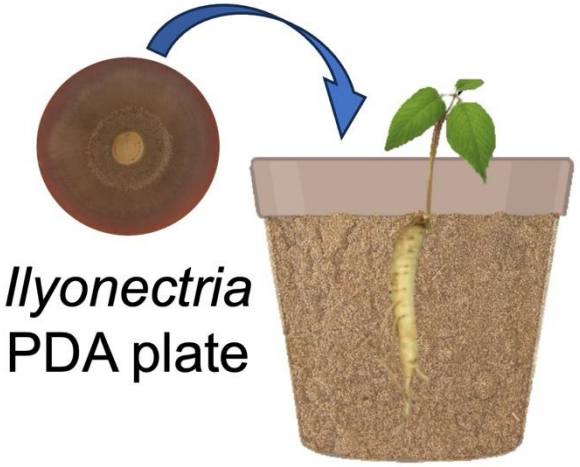
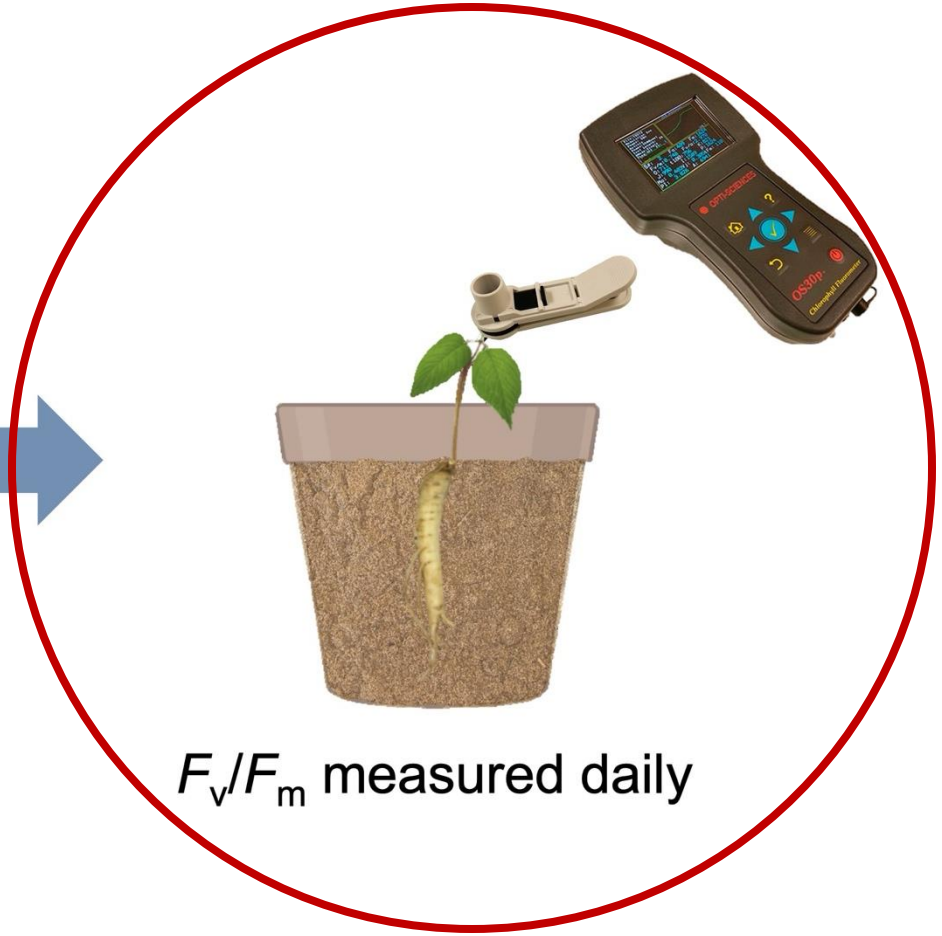


Ilyonectria PDA plate

One year-old American ginseng seedlings, per 12 *Ilyonectria* isolates X 5 replicates



F_v/F_m measured daily

Disease load scored (disease severity index)

28-days

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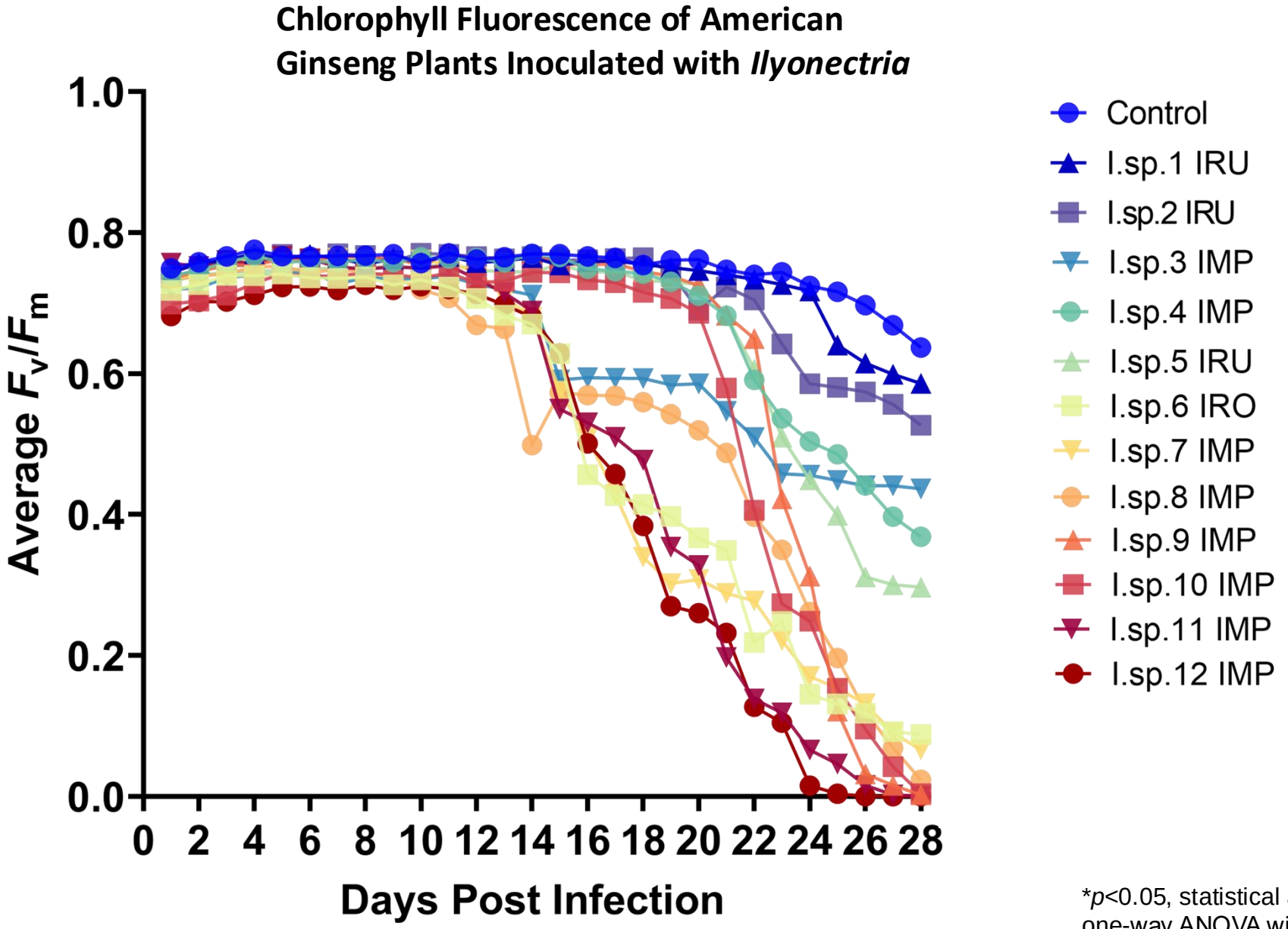

IRU = *I. rufa*
IMP = *I. mors-panacis*
IRO = *I. robusta*

*p<0.05, statistical analysis at 28-dpi using one-way ANOVA with Dunnett's post-hoc test

Objective 1

Determine disease severity in ginseng roots inoculated with different isolates of *Ilyonectria* with and without **previous exposure to ginsenosides**.

Ilyonectria **Number**	**Virulence**	***Ilyonectria*** **Species**	**Host Plant**	**Location**
I.sp.1	Low	*I. rufa*	*Pseudotsuga menziesii* (Douglas fir)	British Columbia, Canada
I.sp.2	Low	*I. rufa*	*Picea glauca* (White Spruce)	Quebec, Canada
I.sp.3	Low	*I. mors-panacis*	*Panax quinquefolius* (American ginseng)	Ontario, Canada
I.sp.4	Low	*I. mors-panacis*	*Poa pratensis* (Blue grass)	Alberta, Canada
I.sp.5	Low	*I. rufa*	*Pseudotsuga menziesii* (Douglas fir)	British Columbia, Canada
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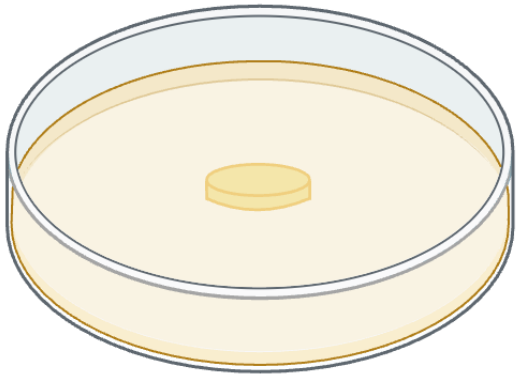
Determine disease severity in ginseng roots inoculated with different isolates of *Ilyonectria* with and without **previous exposure to ginsenosides**.

1



Ilyonectria grown on ginsenoside-infused PDA for 4 plate transfers

2

1 dpi

3

16 dpi

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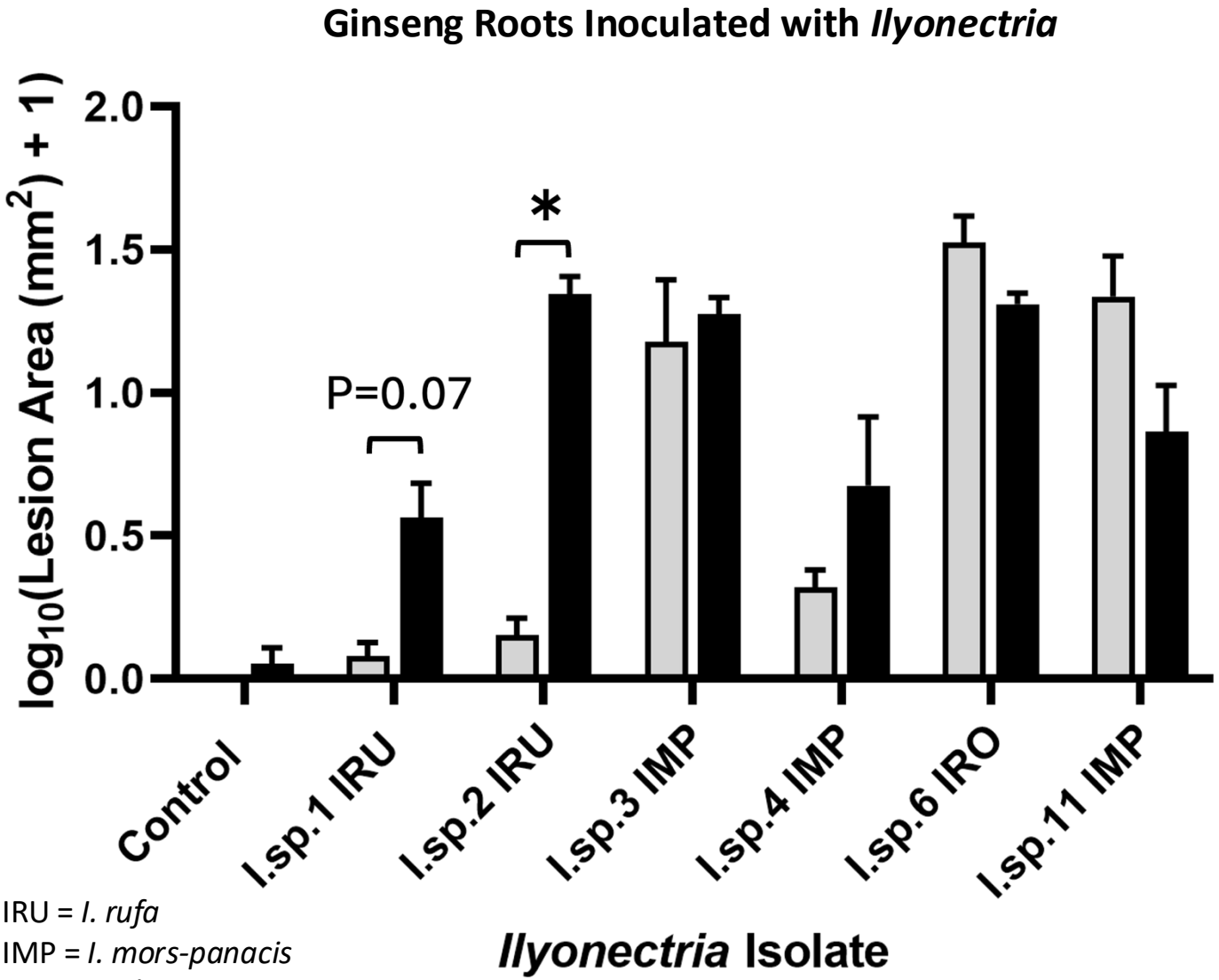

IRU = *I. rufa*
IMP = *I. mors-panacis*
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I.sp.2 IRU
Without Ginsenoside Exposure

I.sp.2 IRU
With Ginsenoside Exposure

Do Ginsenosides Influence the Pathogenicity of *Ilyonectria*?

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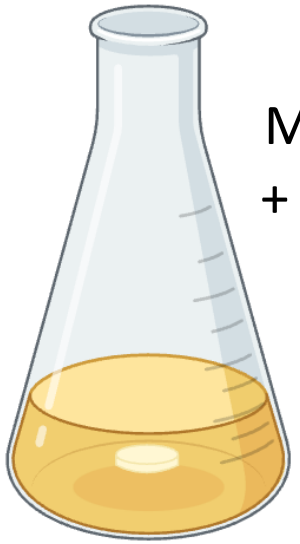
1 Liquid Culture



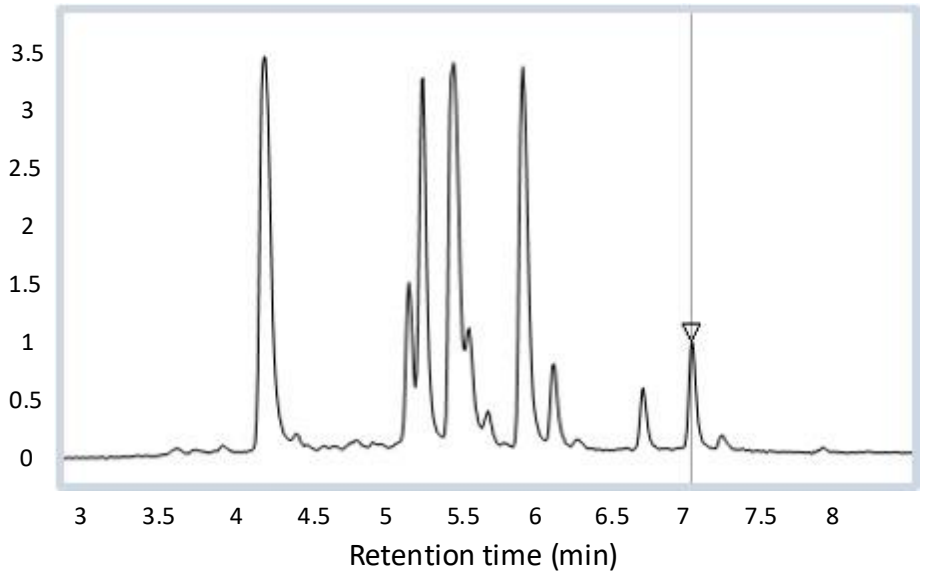
Minimal broth
+ ginsenosides

Ilyonectria PDA plug

7 days

2 LCMS

3 Quantify Ginsenosides



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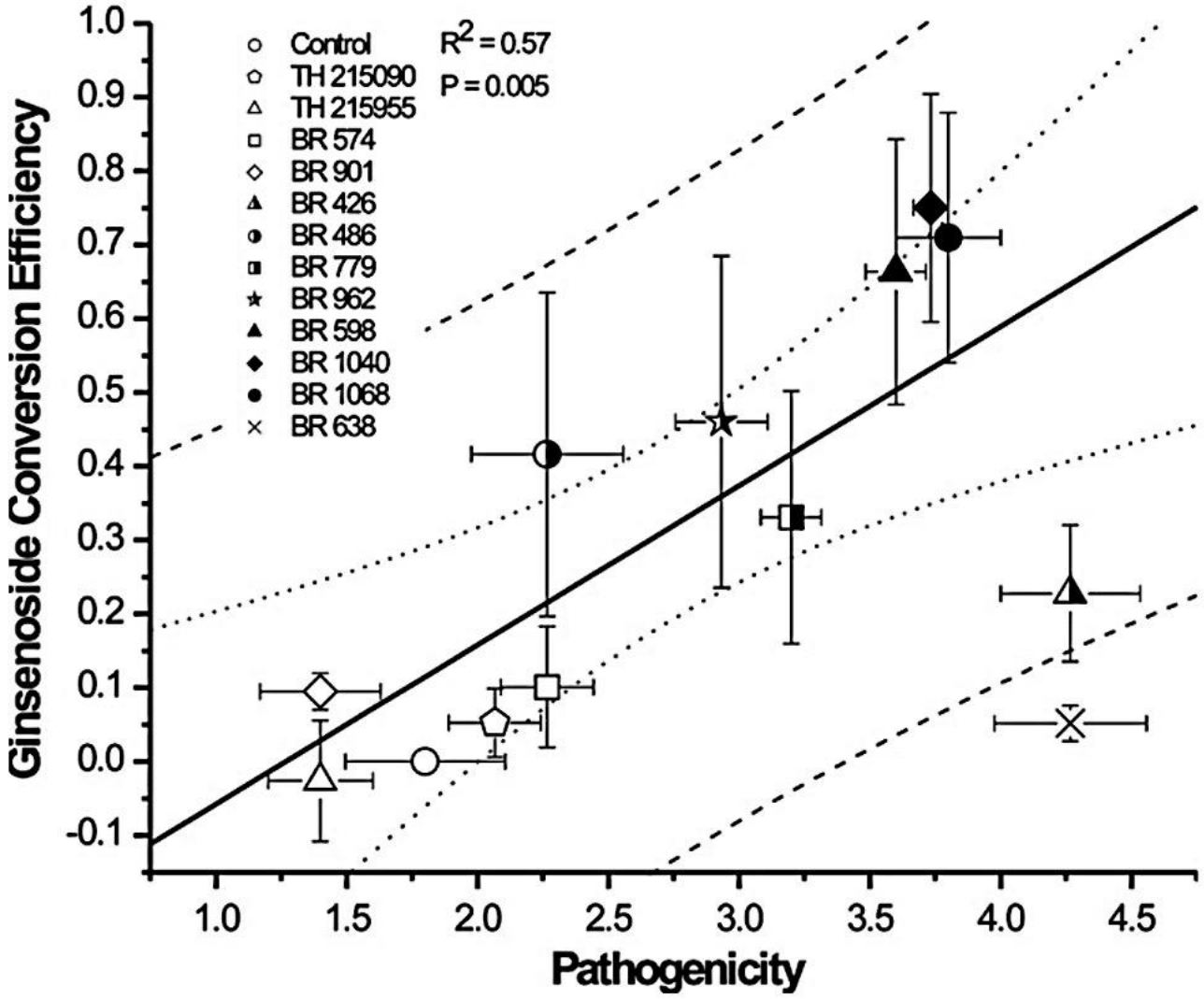

Globisporangium irregulare

(previously *Pythium irregulare*)

- Pathogenic towards American ginseng
- Correlation between metabolism of ginsenosides x pathogenicity

Ivanov, D., and Bernards, M.A. (2012). Phytochemistry, *78*, 44–53

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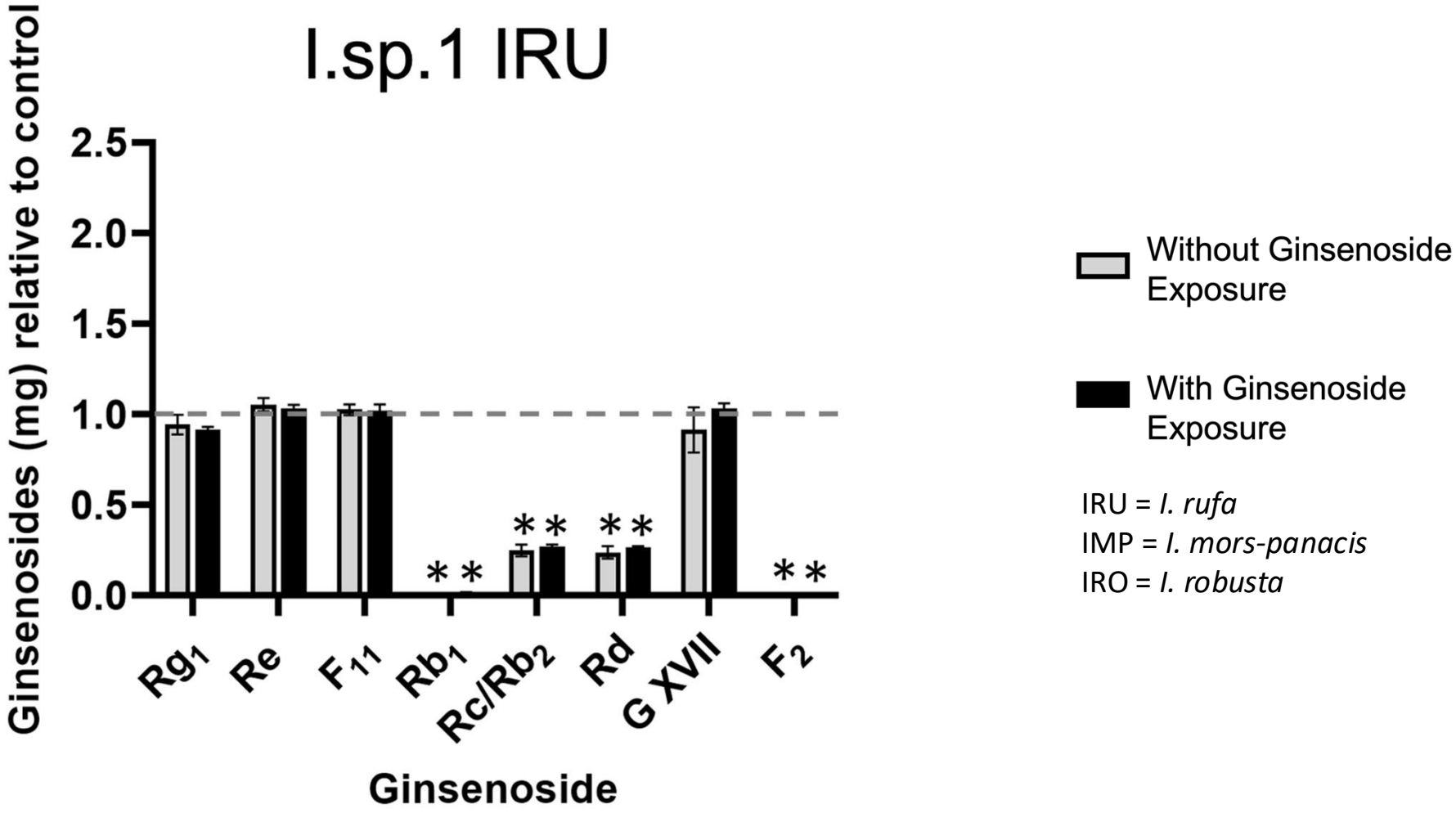

*p<0.05, two-way ANOVA with Dunnett's test
#p<0.05, two-way ANOVA with Šidak's test

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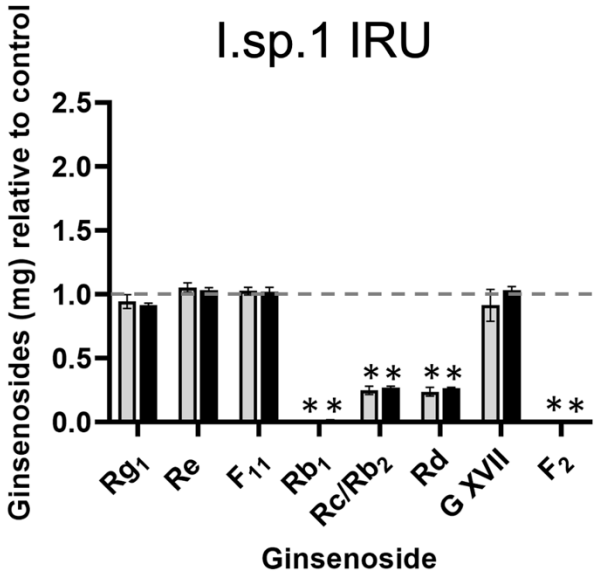
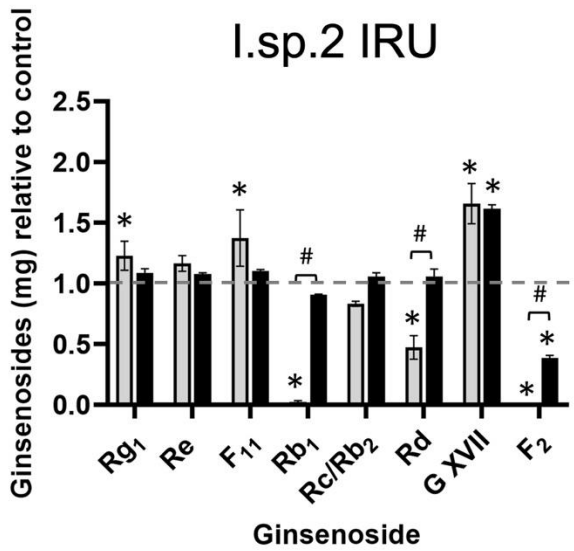
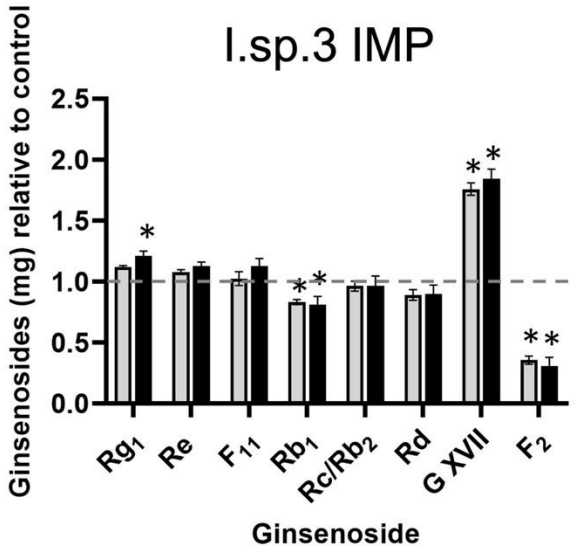
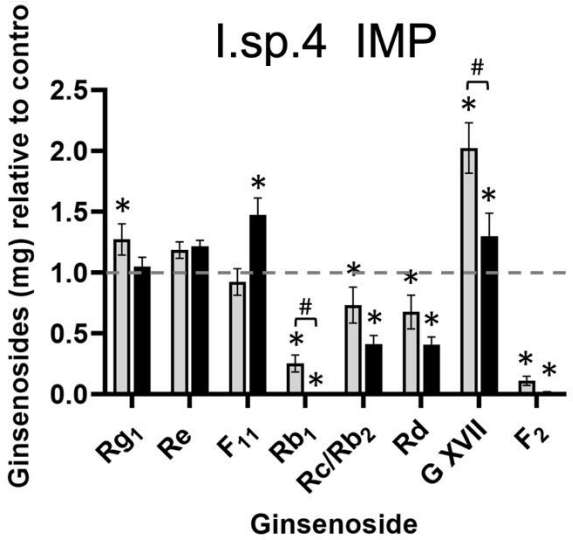
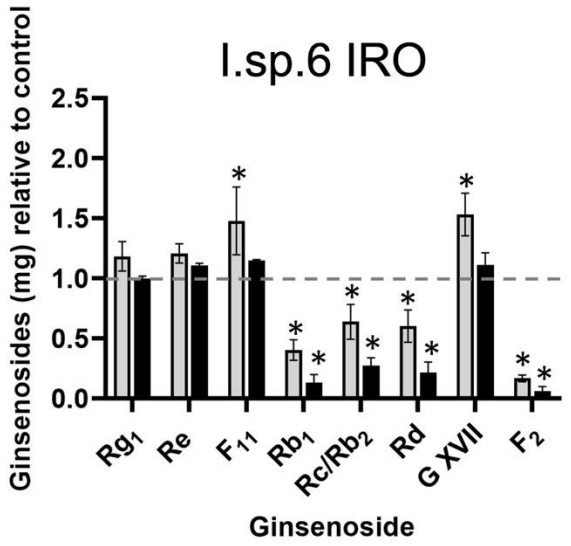
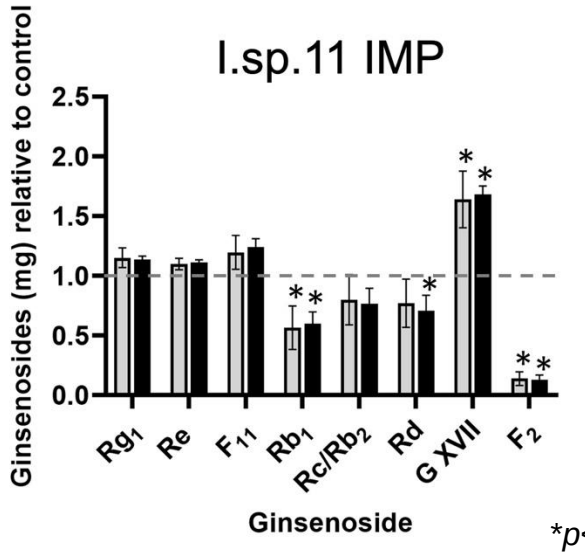












Without Ginsenoside Exposure

With Ginsenoside Exposure

IRU = *I. rufa*
IMP = *I. mors-panacis*
IRO = *I. robusta*

*p<0.05, two-way ANOVA with Dunnett's test
#p<0.05, two-way ANOVA with Šidak's test

Summary: Ginsenosides DO influence *Ilyonectria* Pathogenicity

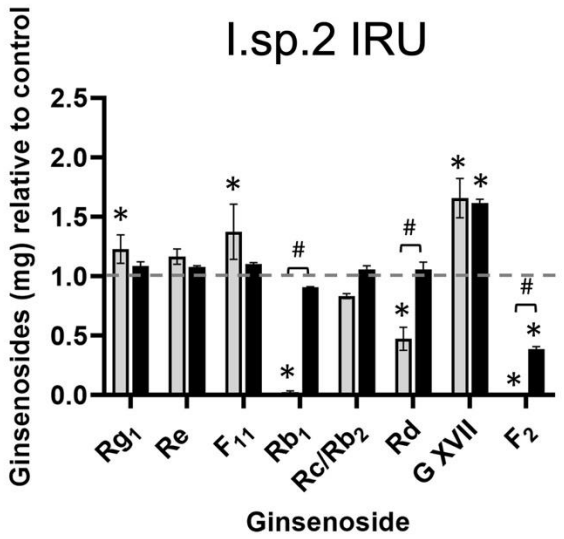
1

I.sp.2 Without Ginsenoside Exposure

I.sp.2 With Ginsenoside Exposure

- *I. rufa* virulence can increase post-ginsenoside exposure
- This likely occurs in later years of ginseng cultivation

2



- All *Ilyonectria* isolates tested can metabolize select ginsenosides
- No correlation between virulence x metabolism of ginsenosides

Research regarding GRD should continue to investigate **how** *Ilyonectria* virulence increases post-ginsenoside exposure.

Acknowledgments

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