Glyphosate Herbicides How Do They Kill Plants?

Jim Rahe
Professor Emeritus,
Simon Fraser University,
Burnaby B.C. Canada
email: rahe@sfu.ca

Acknowledgements

Guri Johal
PhD, 1988
Pioneer documentation of GSI

Andre Levesque
PhD, 1990
LD 50's, Molecular diagnostics for Pythium

Rolando Descalzo
PhD, 1996
Ecology of GSP

Lixing Liu
PhD, 1995
Physiology of GSI

Outline of presentation

I: Contribution of root rot fungi to herbicidal activity of glyphosate

II: Ecology of Pythium spp. as glyphosate synergistic fungi

III: How glyphosate predisposes roots of bean seedlings to colonization by root fungi

Glyphosate herbicides - Roundup, et al.

- Non-selective
- Absorbed and translocated
- Negligible residual activity in most soils
- Water soluble
- Low mammalian toxicity

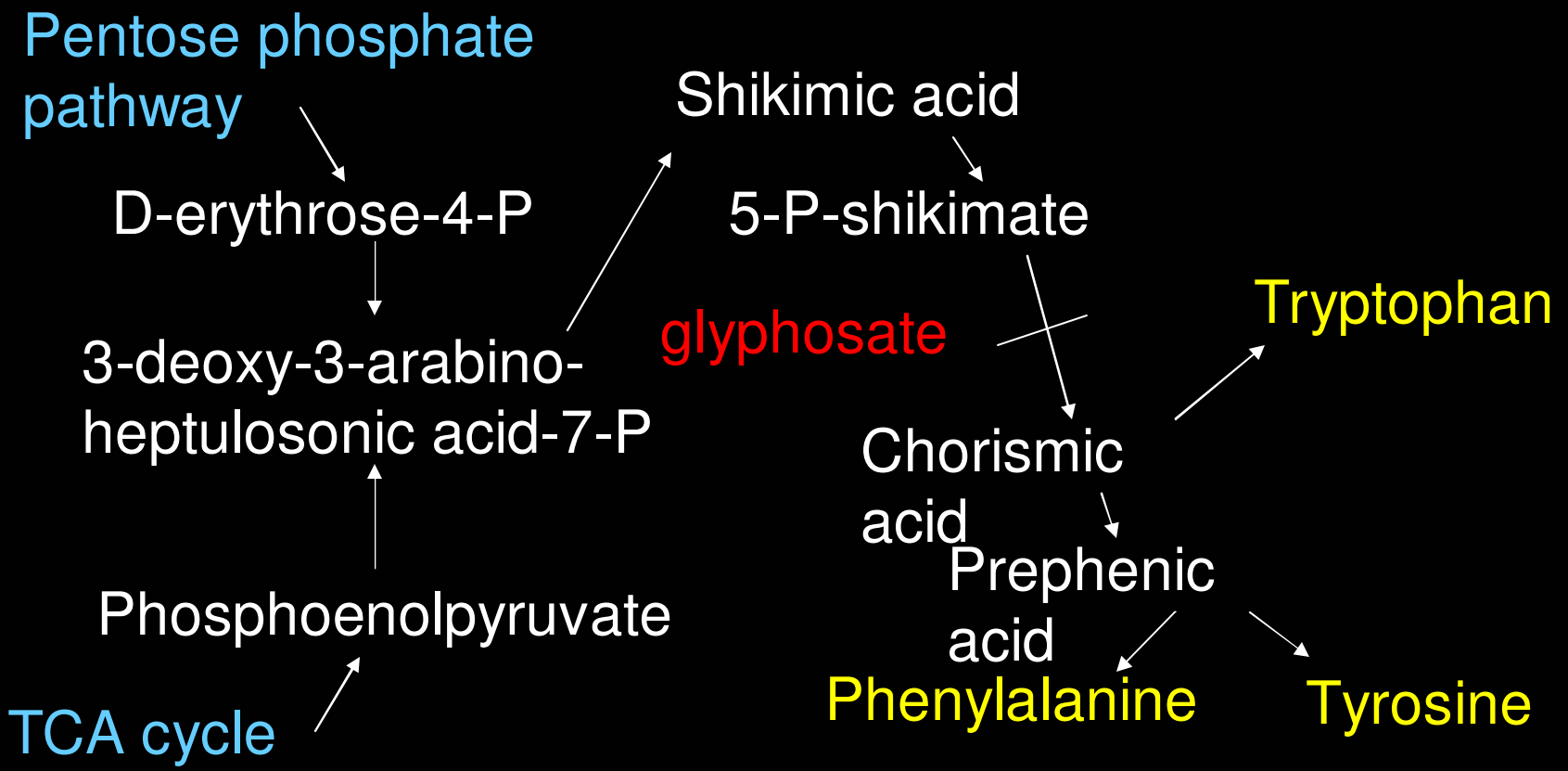
Glyphosate herbicides - uses

- Directed sprays
- Preplant cleanup
- Chemfallow
- Crop desiccation
- Injection (forestry)
- Glyphosate-tolerant GMP’s

Glyphosate herbicides - behavior in plants

- Foliar absorbed
- Slow to kill
- Activity mainly in roots
- Shikimic acid pathway targeted
- Long term effects of sublethal doses

Shikimic acid pathway and some of its main products



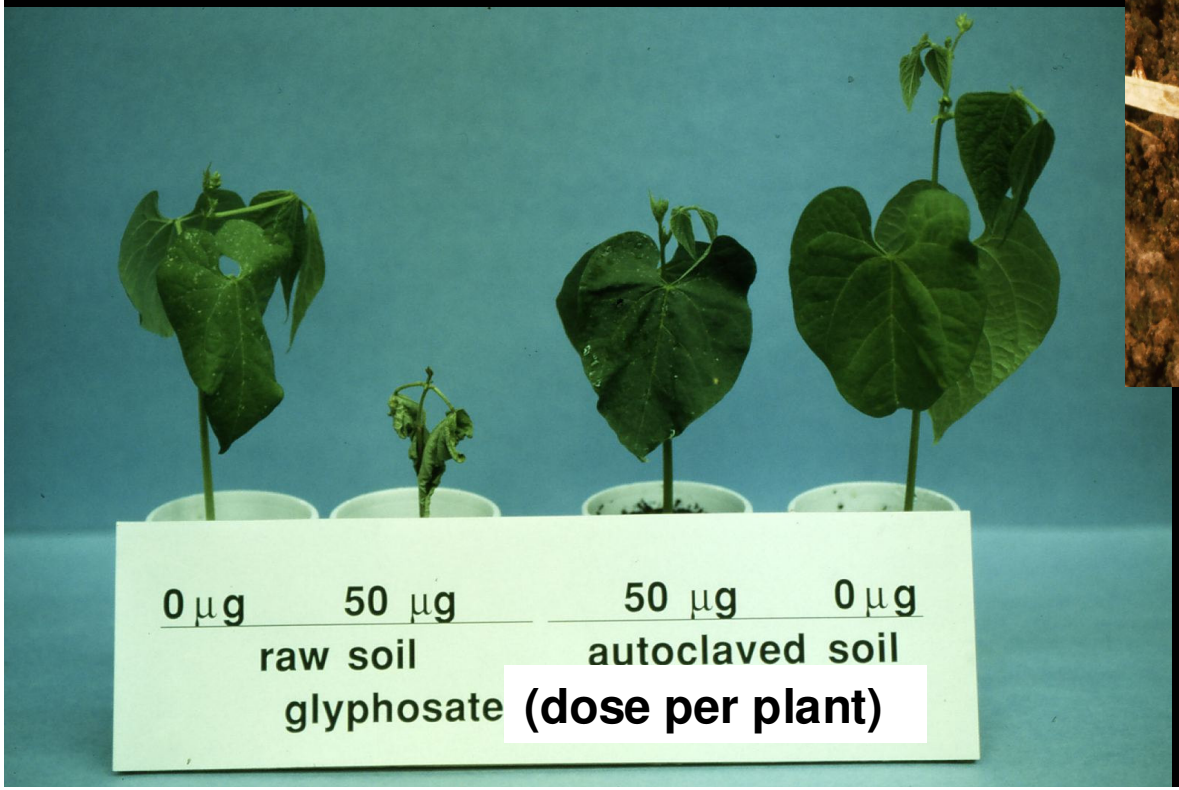
Some questions about the herbicidal effect of glyphosate

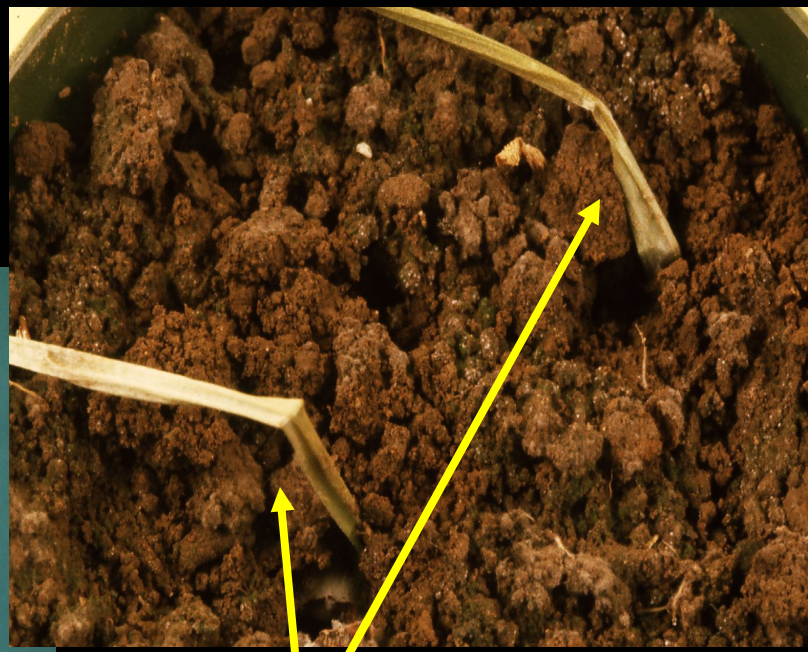
If death of treated plants is directly due to inhibition of the shikimic acid pathway . . .

Why does glyphosate have to be translocated to roots to be effective?

Why do effects of sublethal doses on perennial plants sometimes appear a year after exposure, and persist for two or more years?

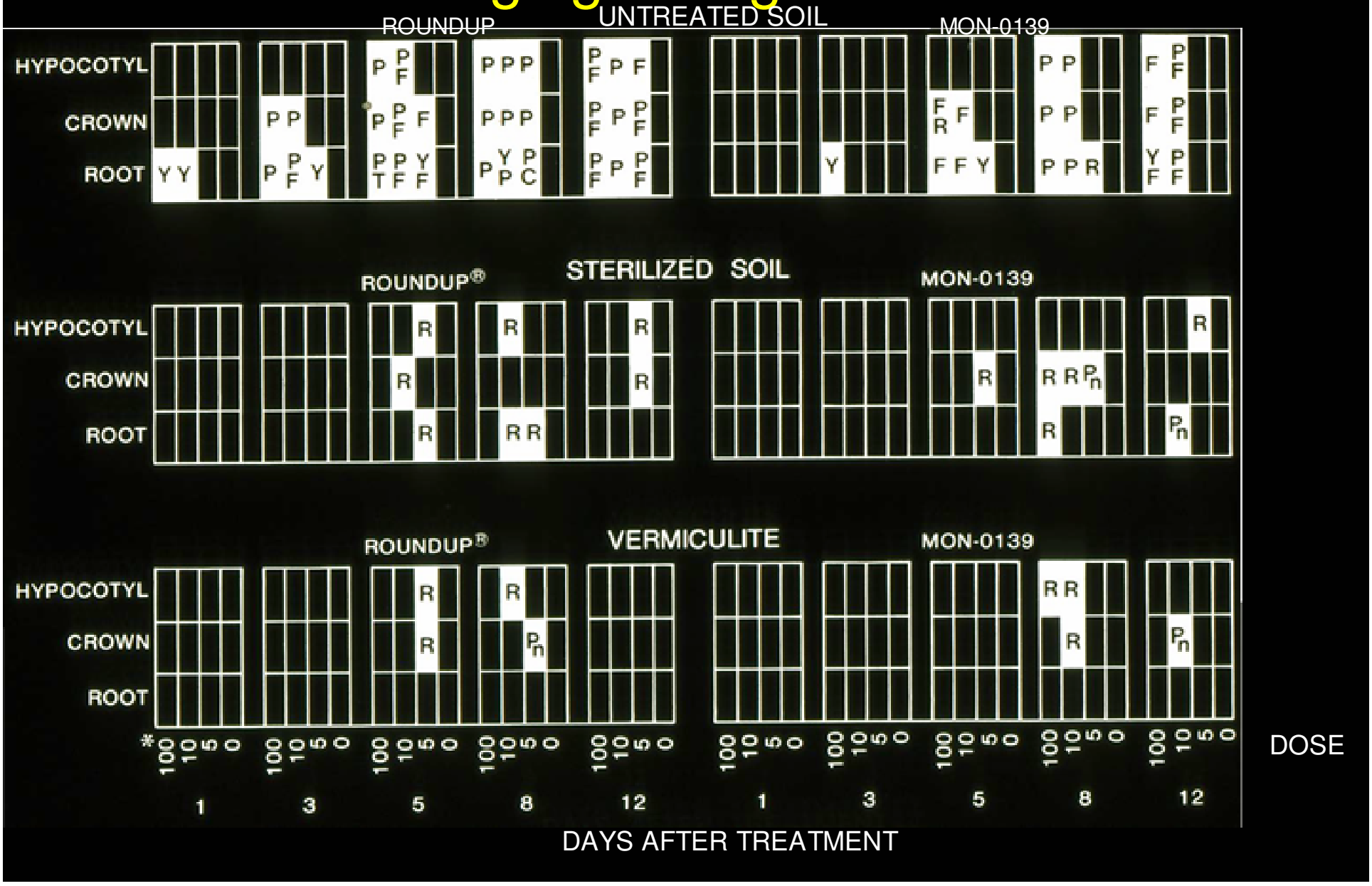
Effect of soil sterilization on herbicidal activity of glyphosate





Symptoms on seedlings killed by glyphosate in untreated soil appear similar to symptoms of damping off caused by Pythium spp.

Fungal colonization of roots of glyphosate treated bean seedlings growing in untreated soil

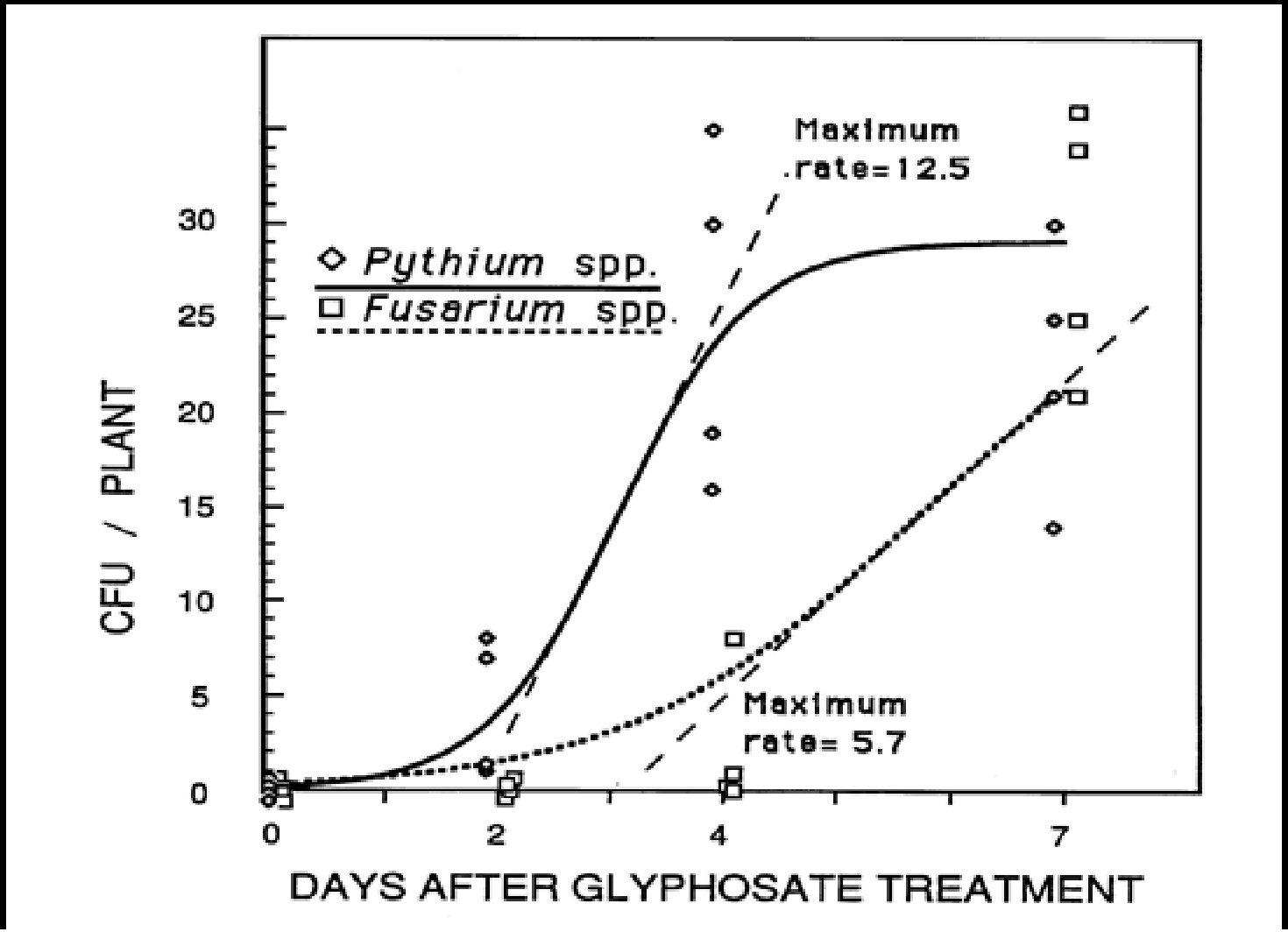


Particulate, Heat-Sensitive Factors From Untreated Soil Restore Herbicidal Activity of Glyphosate to Seedlings Growing in Sterilized Soil

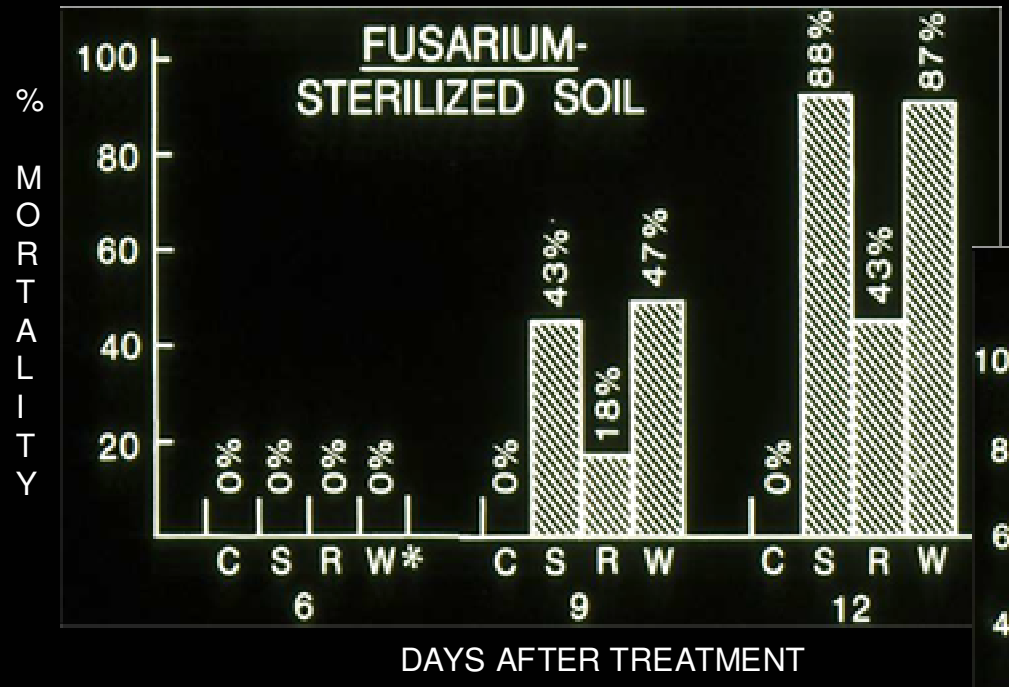
% Mortality in Bean Seedlings Grown in Autoclaved Soil Amended with Extracts from Untreated Soil

Extract:	Raw	Filtered	Autoclaved
Glyphosate	100	0	0
Control	0	0	0

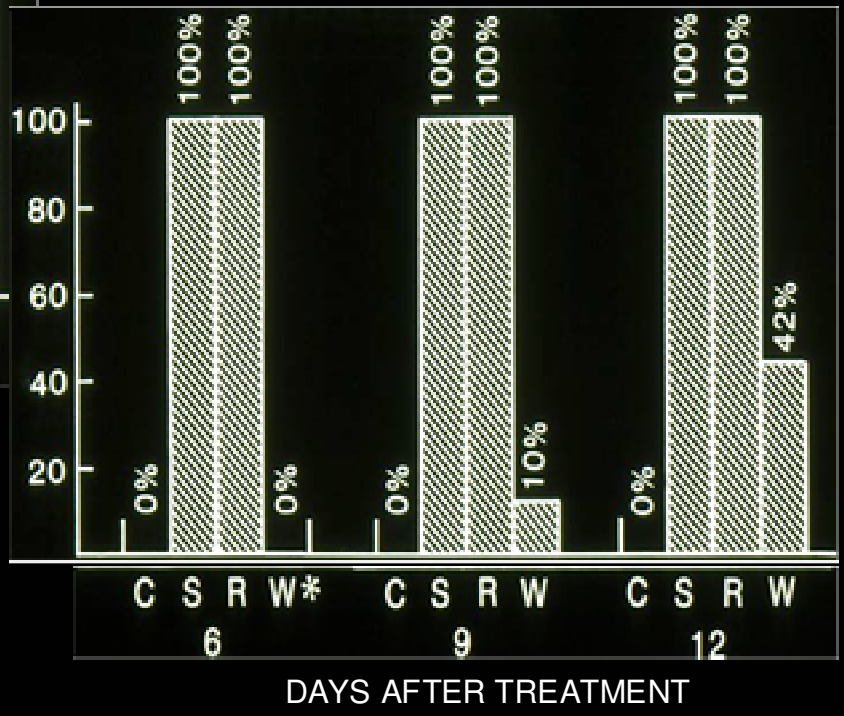
Rates of Colonization of Roots of Bean Seedlings Growing In Untreated Soil by Pythium and Fusarium Following Foliar Treatment With Glyphosate



Fusarium and Pythium restore herbicidal activity of glyphosate to bean seedlings growing in sterilized soil



PYTHIUM-
STERILIZED SOIL



* FUNGUS APPLIED BY
C = CONTROL
S = SEED TREATMENT
R = ROOT DIP
W = WOUNDING

Effect of Ridomil on % Mortality on Bean Seedlings Growing in Different Media, 12 Days After Treatment with Glyphosate

	Pythium - no Ridomil - no	Pythium - yes Ridomil - no	Pythium - yes Ridomil - yes
Sterilized soil	0*	100	0
Vermiculite	0	100	0
Untreated Soil	100	100	88

Conclusion: Ridomil, a systemic fungicide specific for Oomycete fungi, negates the ability of Pythium to restore herbicidal activity of glyphosate to seedlings growing in sterilized soil or soilless media (vermiculite). Ridomil does not protect seedlings growing in untreated soil where Fusarium and other non-Oomycete root rot fungi are present.

Summary of Evidence that Pythium and Fusarium Contribute to Herbicidal Activity of Glyphosate in Bean Seedlings

Herbicidal activity is reduced in sterilized soils, and in soilless plant growth media

Pythium and Fusarium spp. rapidly colonize roots of glyphosate treated plants growing in non sterile soil

Adding Pythium or Fusarium restores herbicidal activity of glyphosate to seedlings growing in sterilized soil or soilless media

Metalaxyl blocks restoration of activity when Pythium is used to amend sterilized soil

II: Ecology of Pythium spp. as glyphosate synergistic fungi

o Glyphosate treated bean and wheat seedlings were used to bait Pythium from diverse agricultural soils

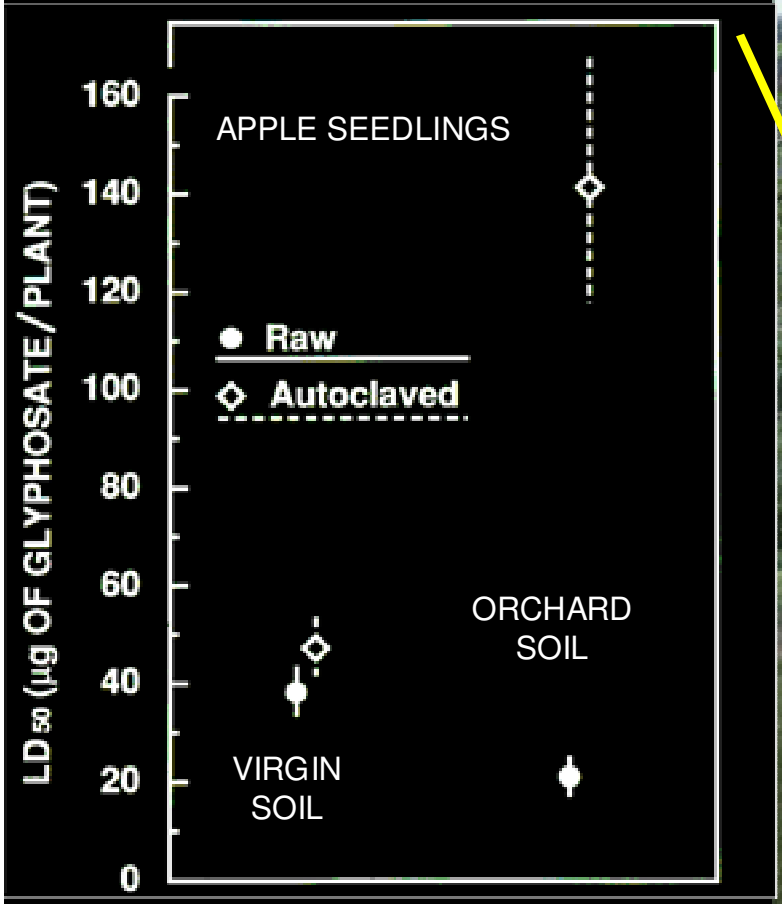
o Pythium isolates from beans were assigned to 15 RFLP groups; isolates from wheat were assigned to 14 RFLP groups.

o Representatives of each RFLP group were tested for efficacy as glyphosate synergists on various monocot and dicot plant species.

Number of isolates of different species and groups of Pythium obtained from roots of glyphosate treated bean seedlings grown in five different soils

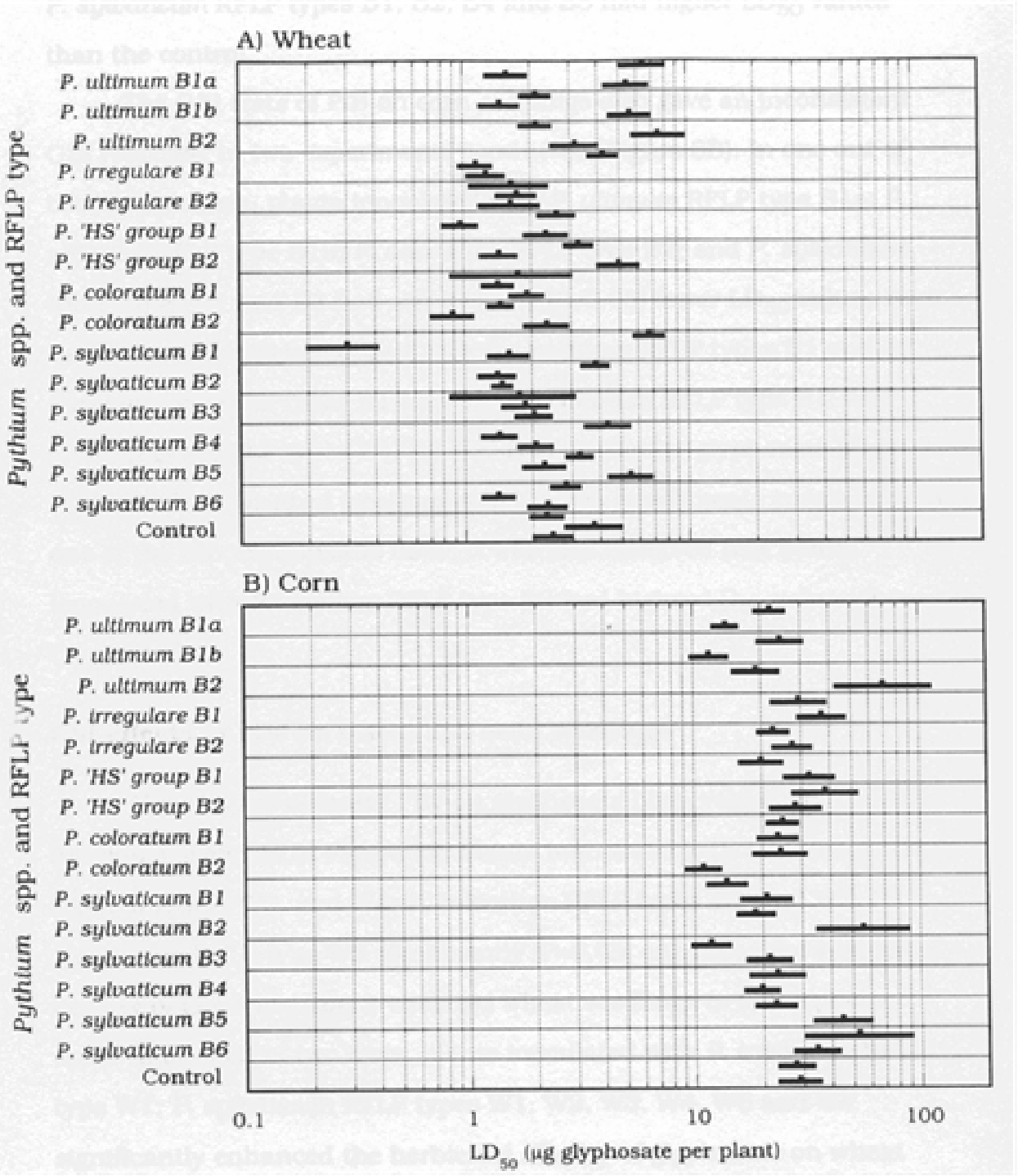
Pythium spp. and groups	Arid climate			Humid climate		Σ
	virgin	replant	crown rot	loam	muck	
P. ultimum	0	4	7	9	6	26
P. irregulare	0	7	0	1	0	8
P. sylvaticum	2	0	6	1	5	14
P. coloratum	0	0	0	0	3	3
P. "G" group	0	1	3	3	4	11
P. 'HS' group	0	0	1	0	2	3
Σ	2	12	17	14	20	65

GSI IN VIRGIN AND ORCHARD SOIL COMPARED



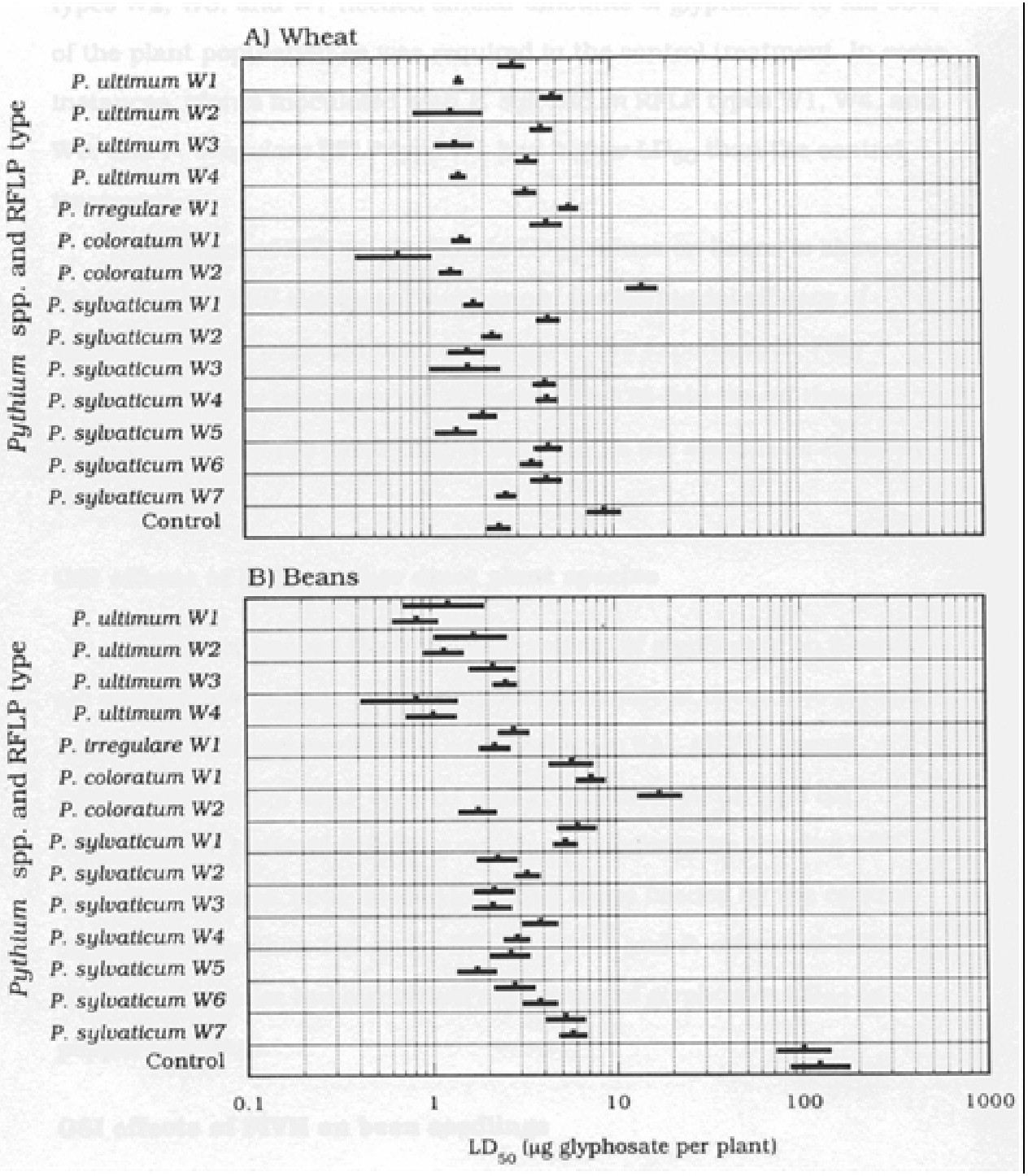
Pythium isolates and GSI

efficacy of 15 isolates from bean, tested on wheat and corn seedlings



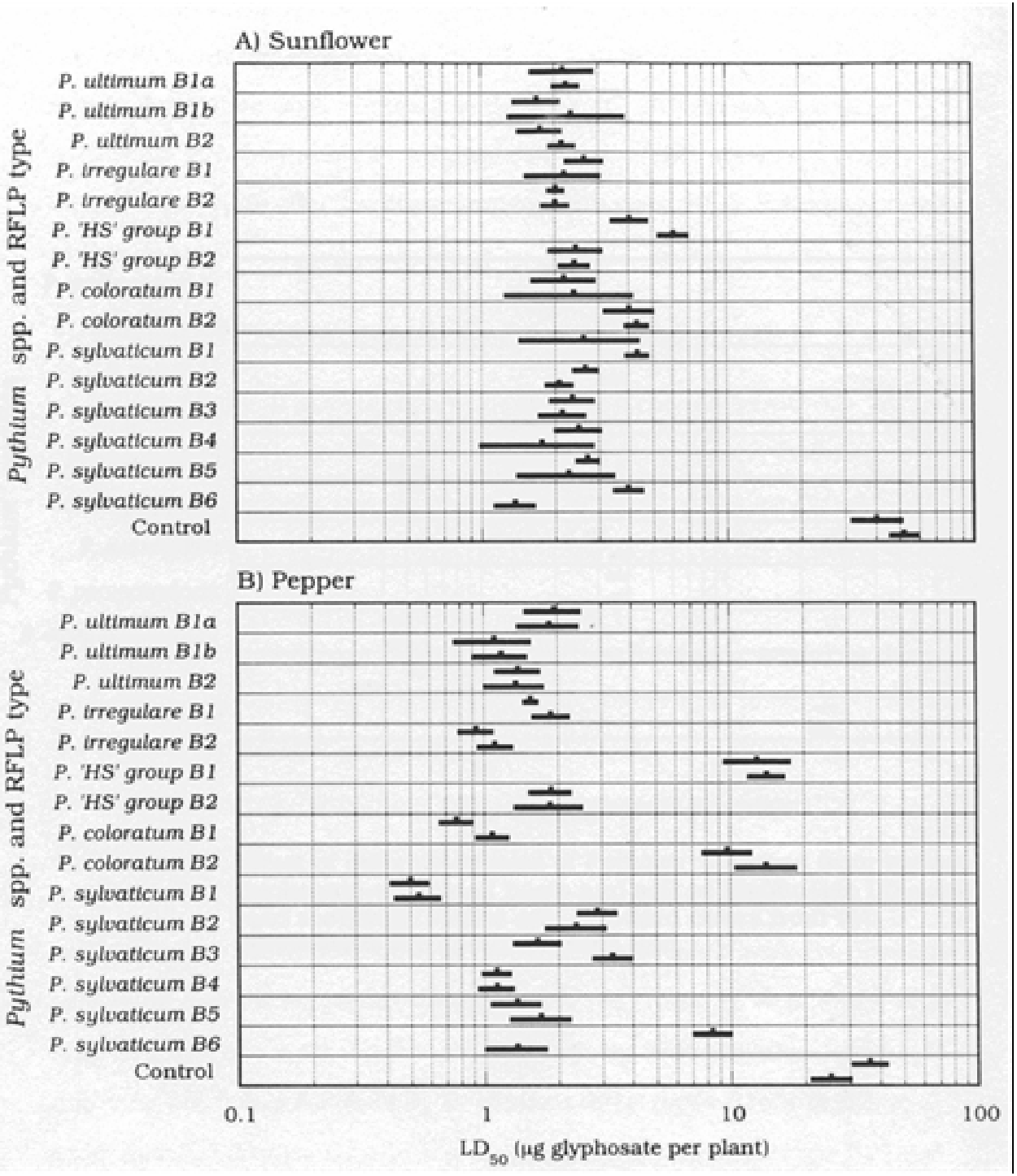
Pythium isolates and GSI

efficacy of 14 isolates from wheat, tested on wheat and bean seedlings



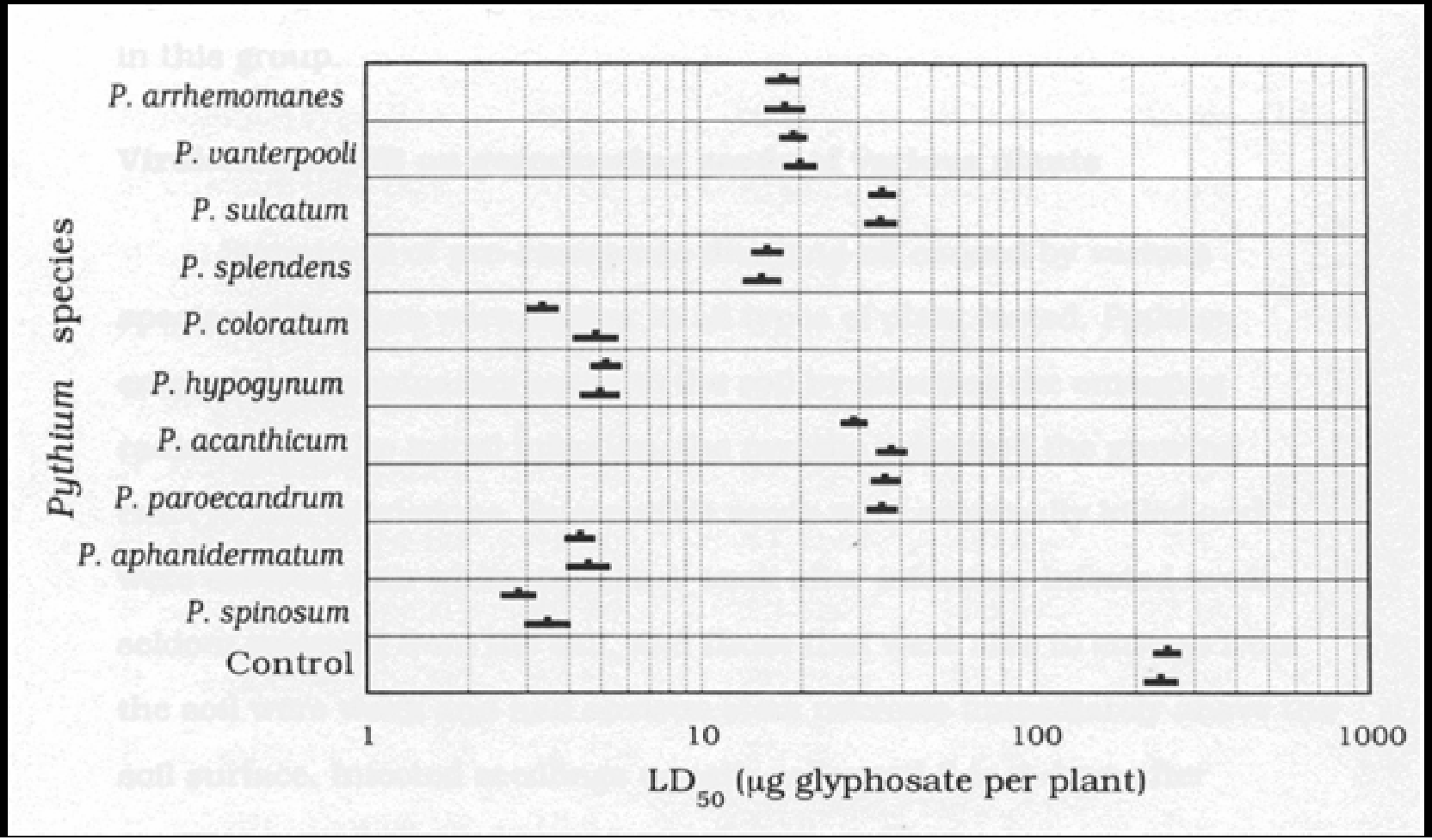
Pythium isolates and GSI

efficacy of 15 isolates from bean, tested on sunflower and pepper seedlings



Pythium isolates and GSI

efficacy of isolates from type cultures, tested on bean seedlings



Pythiaceous fungi as glyphosate synergists - Summary

o GSP were present in and isolated from diverse soil types.

o All isolates and species tested were able to enhance herbicidal efficacy of glyphosate on at least some of the plant species used for testing.

o Individual isolates were typically active on several different plant species.

o Pythium GSI was generally strong (10X-50X) on dicot species, and weak (2X-3X) on monocot species.

III: How Does Glyphosate Predispose Plant Roots to Colonization by Soil Fungi?

Lixing Liu

Studies of Mechanisms of Predisposition by Glyphosate of Bean Roots (Phaseolus vulgaris L.) to Colonization by Pythium spp.

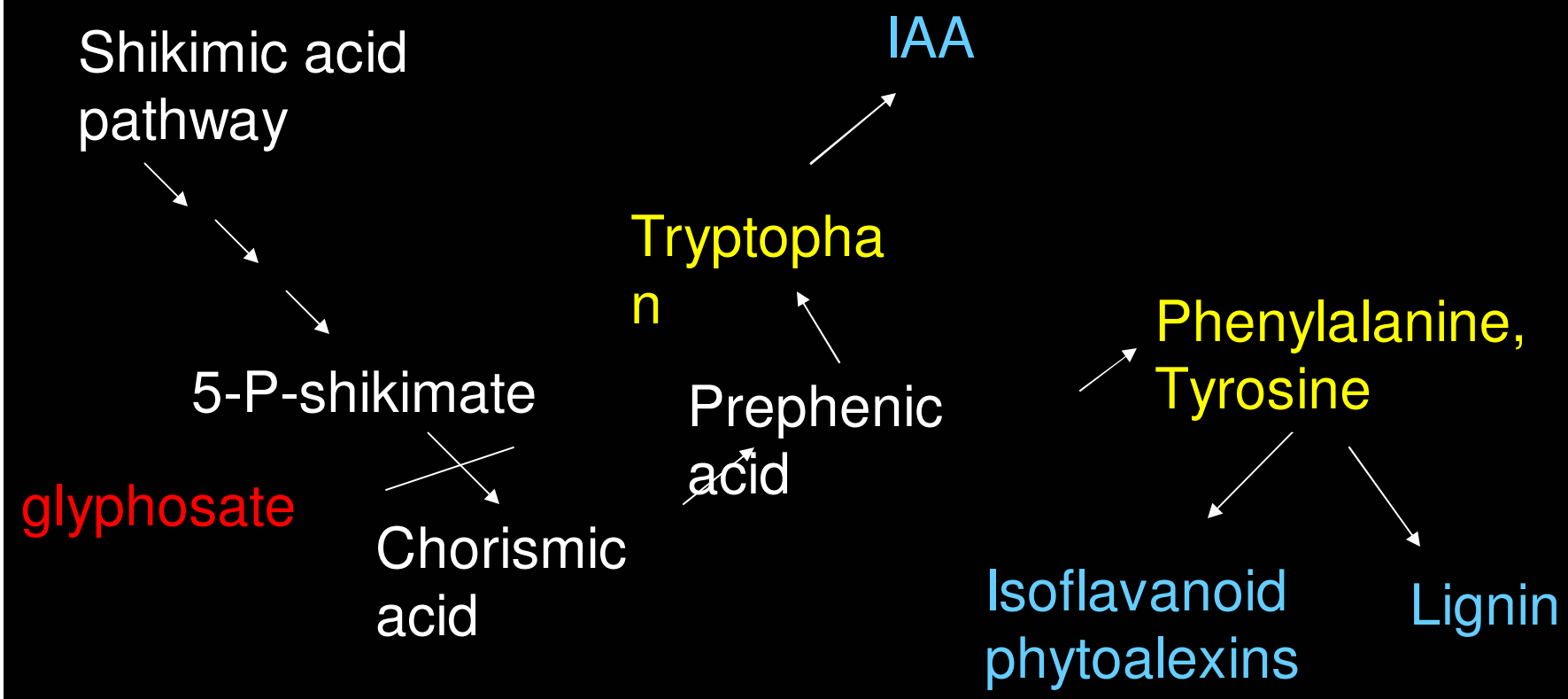
PhD Thesis, 1995, Simon Fraser University

How Does Glyphosate Predispose Plant Roots to Colonization by Soil Fungi?

Four hypotheses

- glyphosate directly stimulates growth of Pythium
- glyphosate enhances root exudation
- glyphosate blocks phytoalexin production
- glyphosate interferes with induced lignification

Shikimic acid pathway and some of its secondary metabolites



Effect of formulations of glyphosate herbicides with and without surfactant on percent germination of sporangia of *Pythium ultimum*

	ppm glyphosate				
	0.0	0.1	1.0	10	100
Formulation*					
Accord	30	35	41	35	28
Roundup	31	34	39	15	0

*Accord contains no surfactant; Roundup contains proprietary surfactant

Conclusions:

1. Glyphosate @ 1 ppm stimulates germination of sporangia
2. Inhibition @ 10, 100 ppm by Roundup formulation probably caused by surfactant

How does 1 ppm glyphosate relate to concentrations of glyphosate in soil solution?

Rates of application; adsorption to soil colloids;
losses over time due to microbial degradation and leaching

How much water in 1 ha plow depth?

1 ha = 1 x 10^4 m^2; = 1 x 10^8 cm^2

1 x 10^8 cm^2 x 17.5 cm = 1.75 x 10^9 cc/ha plow depth

@ bulk density = 1.4, ha plow depth = 2.5 x 10^9g

Water content of different soils typically 30% - 50% w/w

@ -1bar matric potential

Therefore, water content of ha plow depth

= 0.7-1.2 x 10^6 L (for simplicity, let's assume 1 x 10^6L/ha)

Usual application rates 0.9-2.7 L a.I.(glyphosate) /ha.

Therefore, ~1 ppm in soil water IF no adsorption to soil colloids;
if adsorption is 99.9%, ~1 ppb at time of application

Growth (colony diameters, mm @ 24 h) of *Pythium ultimum* in water agar and corn meal agar amended with Roundup and Accord

	ppm glyphosate				
	0.0	0.1	1.0	10	100
Formulation					
Accord					
water agar	43	43	42	44	41
corn meal agar	44	44	45	46	46
Roundup					
water agar	45	45	42	32	13
corn meal agar	48	47	47	46	44

Effect of root exudates from glyphosate-treated and untreated bean seedlings on fungal germination and germ tube growth

	Glyphosate	Control
Germination		
Pythium ultimum	50.5%	36.0%
Fusarium oxysporum	54.0%	43.1%
Germ tube length		
Pythium ultimum	60.5µ	44.6u
Fusarium oxysporum	19.4µ	15.6u

Effect of root exudates from bean seedlings treated with glyphosate or water on growth of germ tubes of *Pythium ultimum* and *FORL**

Water bathing roots changed at time of treatment

	control	glyphosate
P. ultimum	44.6	60.5
FORL*	15.6	19.4

Water bathing roots not changed at time of treatment

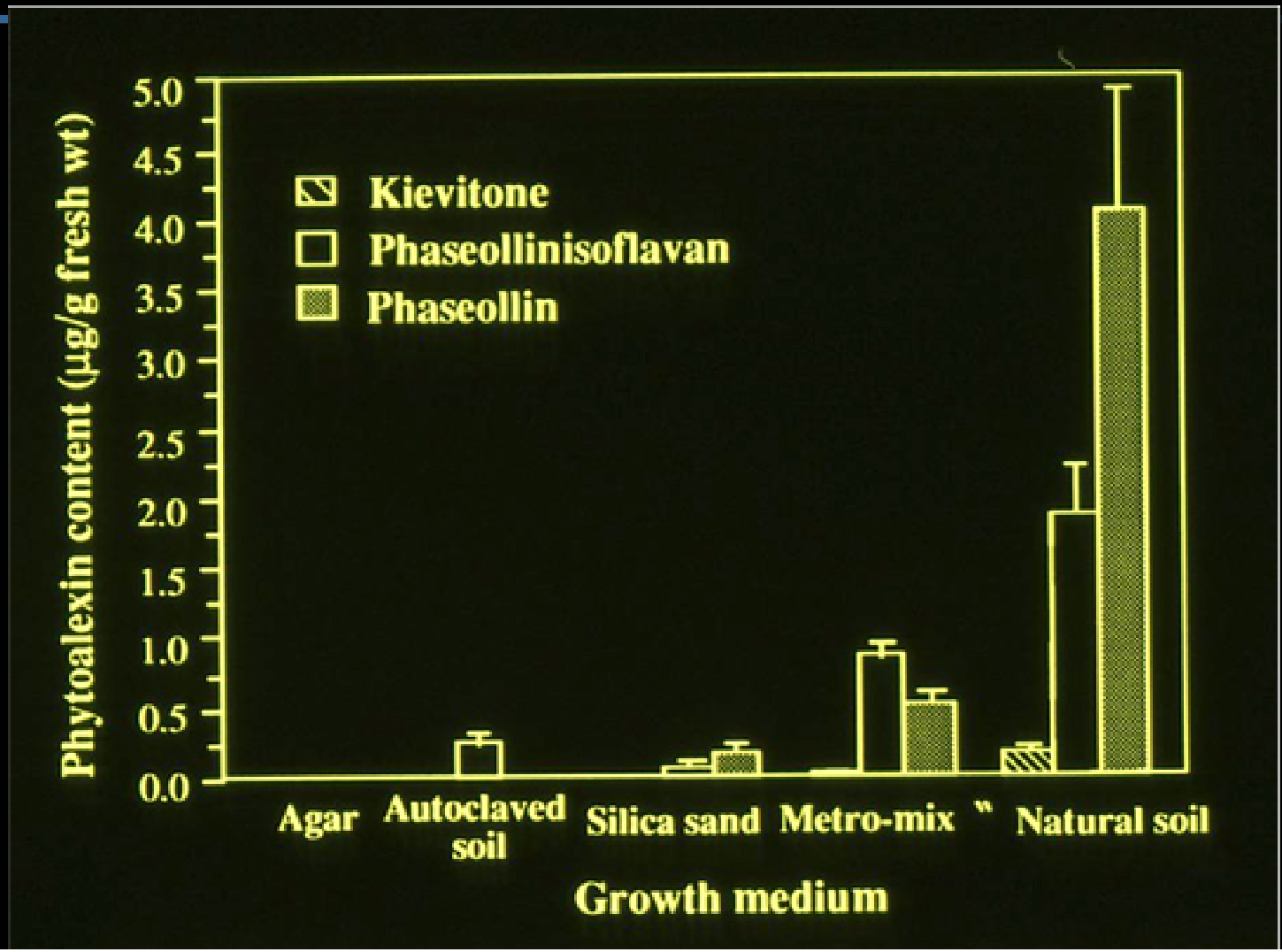
	control	glyphosate
P. ultimum	84.8	82.6
FORL*	29.9	29.5

** Fusarium oxysporum* f.sp. *radicis lycopersici*

Effect of root exudates collected at different times after treatment of bean seedlings with glyphosate on sporangial germination and germ tube growth by *Pythium ultimum*

Exudate collected X hours after Treatment	Germination (%)		Germ tube length	
	control	treatment	control	treatment
0	36	32	11.5	12.0
6	34	42	11.5	11.0
12	31	39	12.0	16.0
24	34	36	15.5	17.5

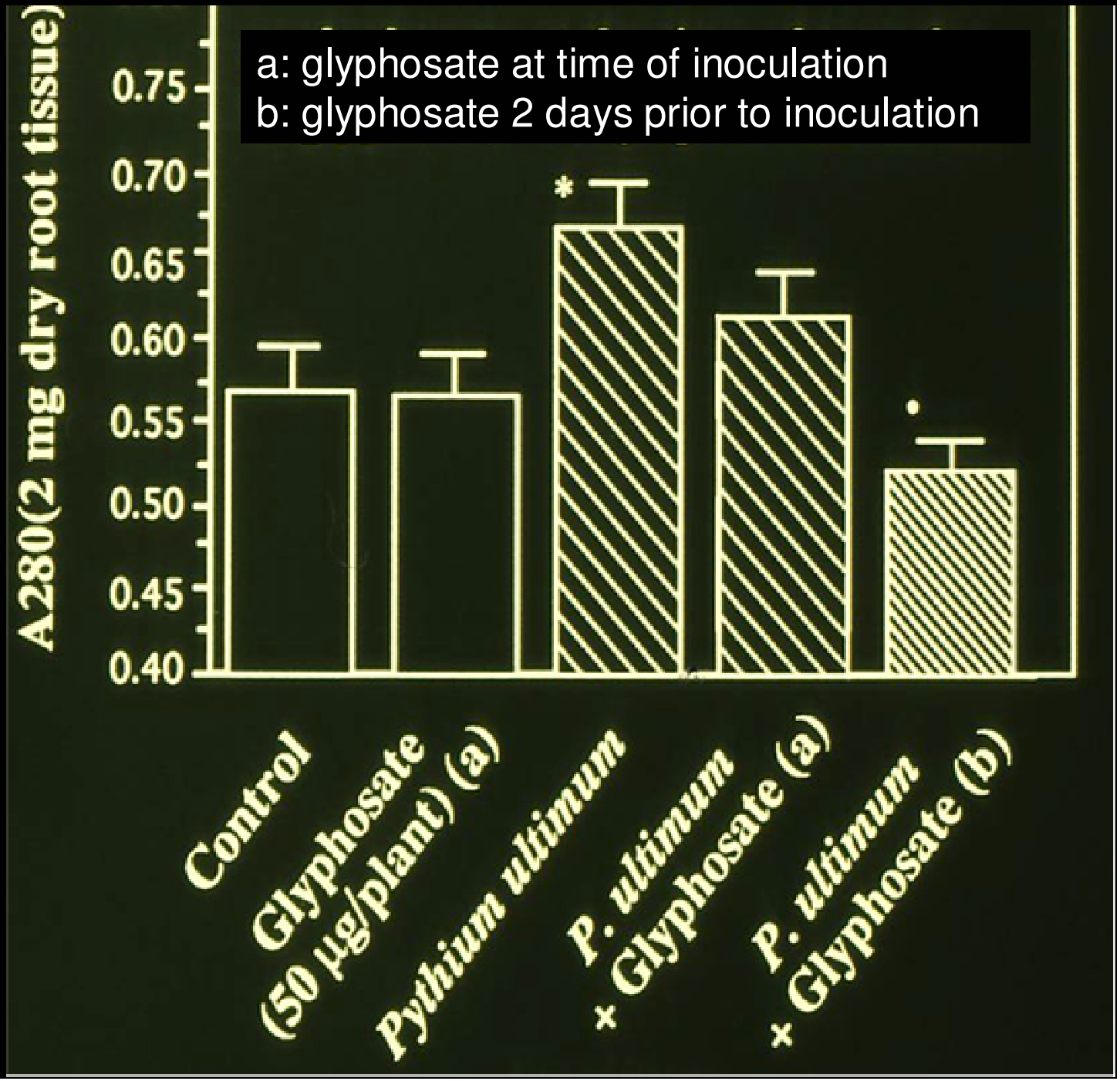
Phytoalexin content of roots of bean seedlings grown in different media



Effect of glyphosate on phytoalexin production by roots of bean seedlings

Treatment	Phaseollin (υg/g fresh wt)
Control	3.5
Glyphosate	3.4
Pythium ultimum	13.7
P. ultimum + glyphosate	10.0

Effect of glyphosate on induced lignification in roots of bean seedlings



Summary - I

Research by Johal, Levesque, Descalzo, Liu, Rahe provides conclusive evidence that glyphosate predisposes plant roots to colonization by root rot fungi such as Pythium and Fusarium within 2-3 days of treatment

These fungi, ubiquitous in agricultural soils, contribute significantly to the herbicidal efficacy of glyphosate on dicot seedlings.

Summary - II

Predisposition of roots to colonization by low doses of glyphosate may explain why

- glyphosate has to be translocated to roots to be effective.

- effects of sublethal doses on perennial plants sometimes appear a year after exposure, and persist for two or more years.

Whole Root Plating to Determine # of CFU Per Plant