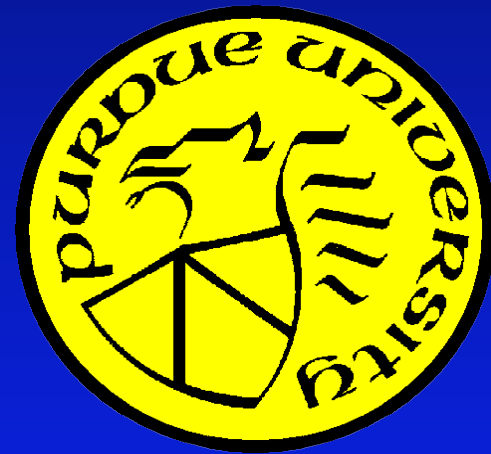
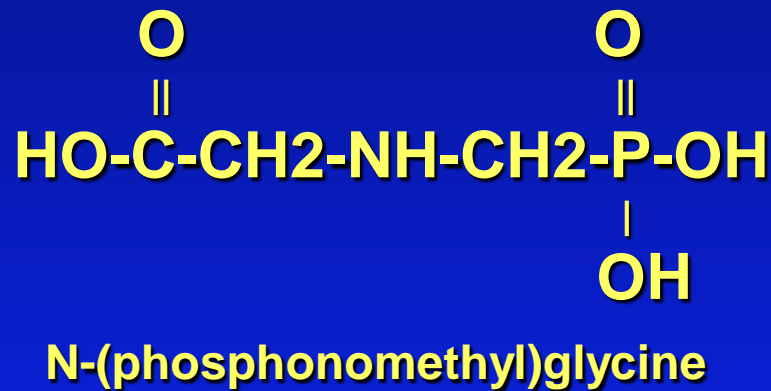
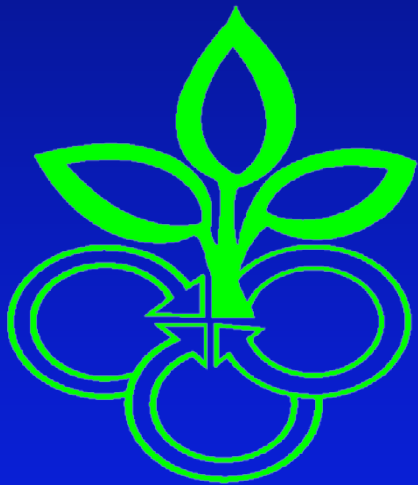


Glyphosate Effects on Diseases of Plants

Symposium: Mineral Nutrition and Disease Problems
in Modern Agriculture: Threats to Sustainability

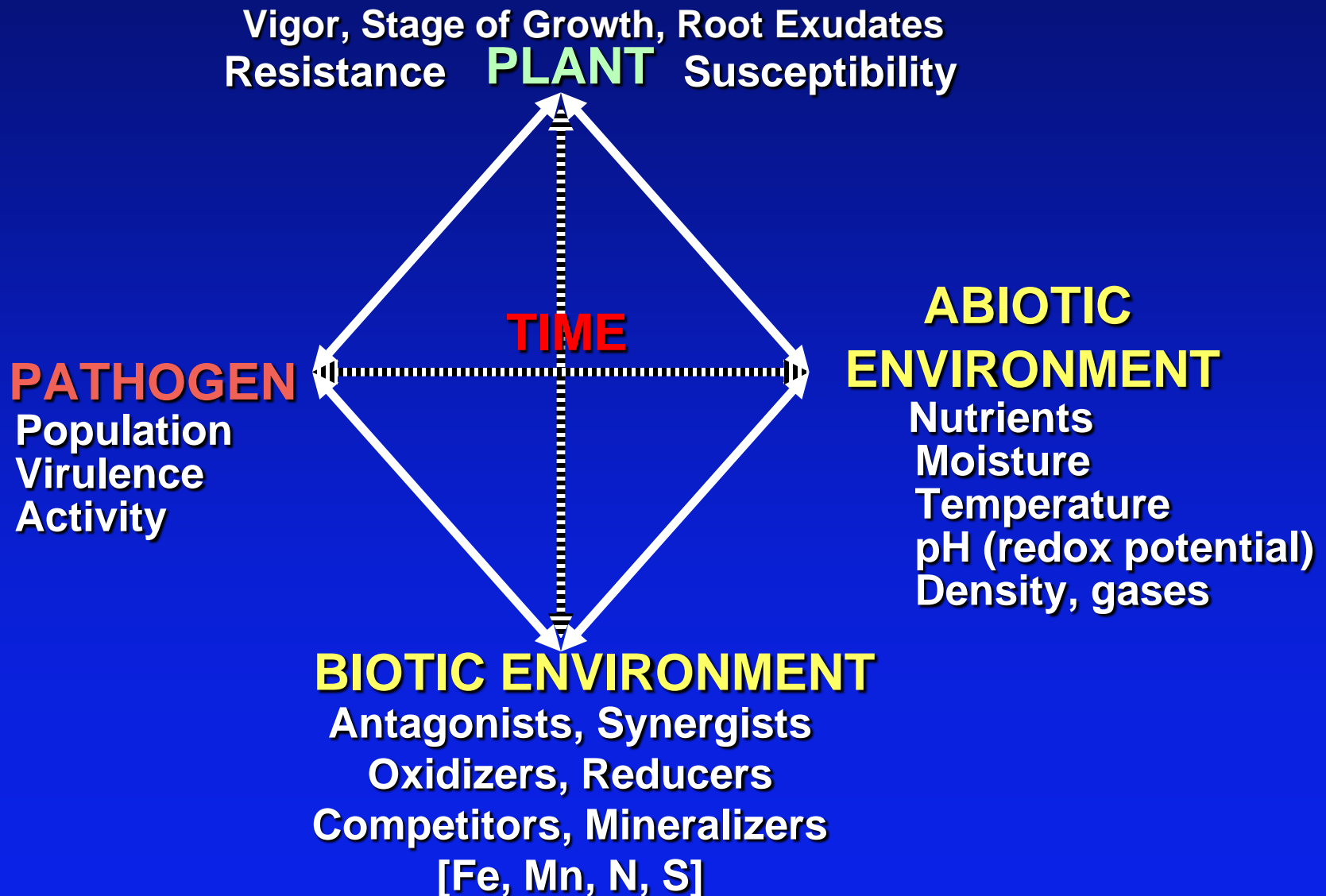


D. M. Huber, Emeritus Professor
Botany & Plant Pathology Department
Purdue University, West Lafayette, IN 47907

Glyphosate Effects on Diseases of Plants

- **Background - review**
 - Interacting factors for disease
 - Some cultural factors affecting nutrition and disease
- **Glyphosate**
 - Characteristics
 - Glyphosate resistance
 - Reported effects of glyphosate
- **Effect of glyphosate on disease**
 - Take-all root and crown rot of cereals
 - *Corynespora* root rot
 - *Marasmius* root rot of sugarcane
 - *Fusarium* head scab of cereals
 - Citrus variegated chlorosis (CVC)
 - Rust diseases
 - Rice blast
- **Mechanisms to reduce disease**
- **Conclusions**

INTERACTING FACTORS DETERMINING DISEASE SEVERITY



Changes in Agricultural Practices Change the Interactions

Crop Sequence

Biotic environment
Nutrition
Nitrification
Organic matter

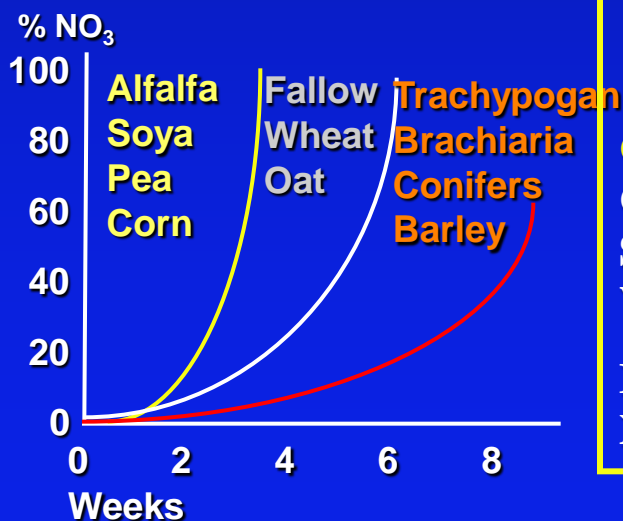
Tillage/No-till

Residue break down
Soil density/aeration
Pathogen survival
Nutrient distribution
Denitrification

Fertilization

Rate/form
Time applied
Source/assoc. ions
Inorganic
Organic

Effect of crop residue on nitrification

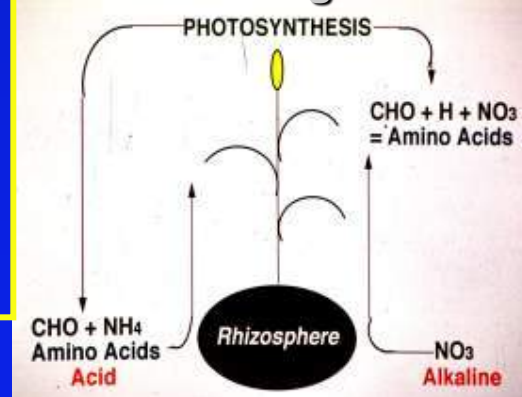


Crop sequence effect on Mn²⁺

Rotation	Extractable Mn
Continuous Corn	130 ppm
Continuous soybeans	64 pp,
Soybean, wheat, corn	91 ppm
Wheat, corn, soybean	79 ppm
Fall chisel	126 ppm
No-till	80 ppm



Metabolism of different forms of nitrogen



Factors Affecting N Form, Mn Availability and Severity of Some Diseases*

Soil Factor or Cultural Practice	Nitrification	Effect on: Mn Availability	Disease Severity
Low Soil pH	Decrease	Increase	Decrease
Green Manures(some)	Decrease	Increase	Decrease
Ammonium Fertilizers	Decrease	Increase	Decrease
Irrigation (some)	Decrease	Increase	Decrease
Firm Seed bed	Decrease	Increase	Decrease
Nitrification Inhibitors	Decrease	Increase	Decrease
Soil Fumigation	Decrease	Increase	Decrease
Metal Sulfides	Decrease	Increase	Decrease
High Soil pH	Increase	Decrease	Increase
Lime	Increase	Decrease	Increase
Nitrate Fertilizers	----	Decrease	Increase
Manure	Increase	Decrease	Increase
Low Soil Moisture	Increase	Decrease	Increase
Loose Seed bed	Increase	Decrease	Increase

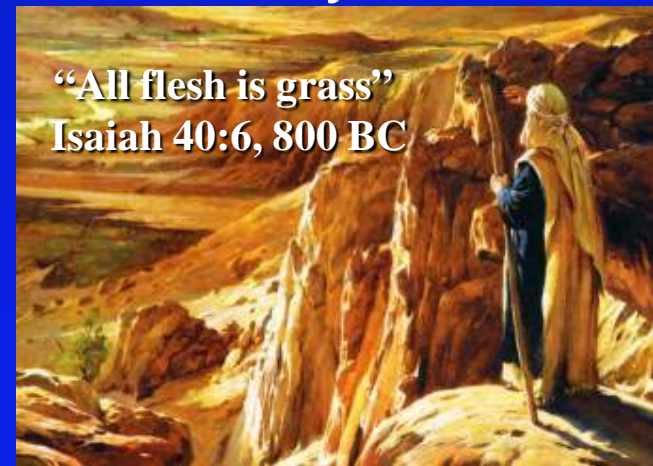
*Potato scab, Rice blast, Take-all, Phymatotrichum root rot, Corn stalk rot

Glyphosate Started Changing Agriculture 30+ Years Ago

The most widely used agricultural chemical!

- Broad-spectrum (non-selective) weed control
 - Paraquat, Tordon, Spike, salt
- Short “direct” residual activity
- Low direct mammalian toxicity
- Economical use
- TRANSGENIC PROTECTION - selectivity

**A very strong metal chelator with
Potential interaction with all life
Through mineral deprivation**



Some Characteristics of Glyphosate

- A chemical chelator

Small amount needed

Tightly bind mineral elements

Immobilizes Mn, Fe

- Non-specific herbicidal effect

- Tank mix impairs herbicidal activity

Chelating stability constants of glyphosate

Metal ion	$\frac{[ML]}{[M][L]}$	$\frac{[MHL]}{[M][H][L]}$	$\frac{[ML_2]}{[M][L_2]}$
Mg ²⁺	3.31	12.12	5.47
Ca ²⁺	3.25	11.48	5.87
Mn²⁺	5.47	12.30	7.80
Fe ²⁺	6.87	12.79	11.18
Cu ²⁺	11.93	15.85	16.02
Fe ³⁺	16.09	17.63	23.00



Glyphosate



Glyphosate + Zn tank mix



Some Chemical Chelators in Agriculture

- **Mn, Fe chelating compounds**
 - Piricularin, alpha-picolinic acid - rice blast toxin
 - Glyphosate - non-specific herbicide
 - Reducing activity - photosynthesis
- **Cu chelating compounds**
 - Nitrapyrin, methyl pyrazole - inhibit nitrification
 - Tordon herbicide - specific to broad-leaved plants
 - Oxidizing activity - (lacases, oxidases)
- **Various plant root exudates**
 - Induced with nutrient deficiency

Source of Chelators

- **Natural metabolites**

 - Plant root exudates - organic acids, siderophores

 - Microbial metabolites - organic acids, toxins

 - Soil organic matter

- **Synthetic compounds**

 - Herbicides - glyphosate, Tordon

 - Nitrification inhibitors - nitrapyrin

 - EDTA, DTPA, citric acid, amino acids

- **Important because micronutrients are the:**

 - Activators

 - Inhibitors

 - Regulators of plant physiological functions

Characteristic Effects of Glyphosate

- **Systemic in plants**

- A modified essential amino acid
 - Concentrates in meristematic tissues
 - Shoot and root tips
 - Reproductive structures

- **Distributed throughout the rhizosphere in root exudates**

- **Non-specific herbicidal effect**

- **Toxic to some soil microbes; stimulates others**

- Changes nutrient availability
 - Changes virulence of some pathogens



Some Microbial Interactions with Glyphosate

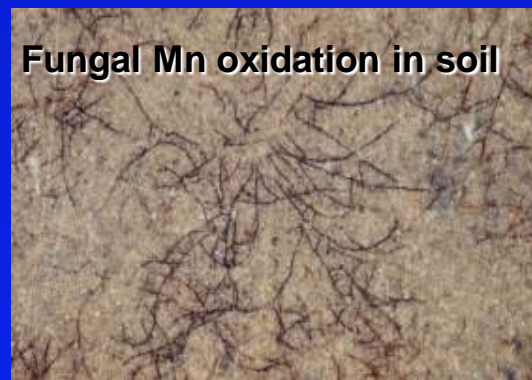
- Changes the soil microbial “balance”
- Toxic to beneficial organisms:
 - Rhizobium, Bradyrhizobium
 - Inhibits N-fixation
 - Mn reducing organisms (Biocontrol)
 - *Trichoderma* spp, *Bacillus* spp
 - Mychorrhizae
 - *Glomus mossea* - Zn, P uptake
- Stimulates:
 - Mn oxidizing organisms
 - *Fusarium*, other fungi
 - K sink immobilization
- Increases pathogens:



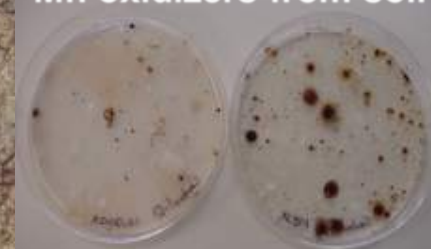
Manganese Availability

pH 5.2 to pH 7.8

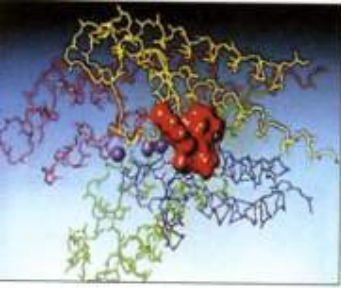
Rhizosphere biology



Mn oxidizers from soil

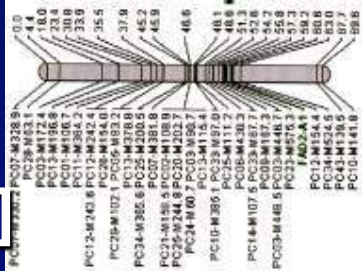


Control Glyphosate



Roundup Ready® Gene

[Greatly expanded usage of glyphosate]



- **Confers “tolerance” to glyphosate**
 - Alternate metabolic pathway introduced
 - Slows down some physiologic processes
 - Provided selective herbicidal activity
 - There are several “modifiers” possible
- **Changes physiology of the plant (N metabolism)**
- **Incomplete “protection” of meristematic and reproductive tissues - depends on:**
 - Time of application
 - Method of application
 - Crop species
- **Often causes a “Yield Drag”**



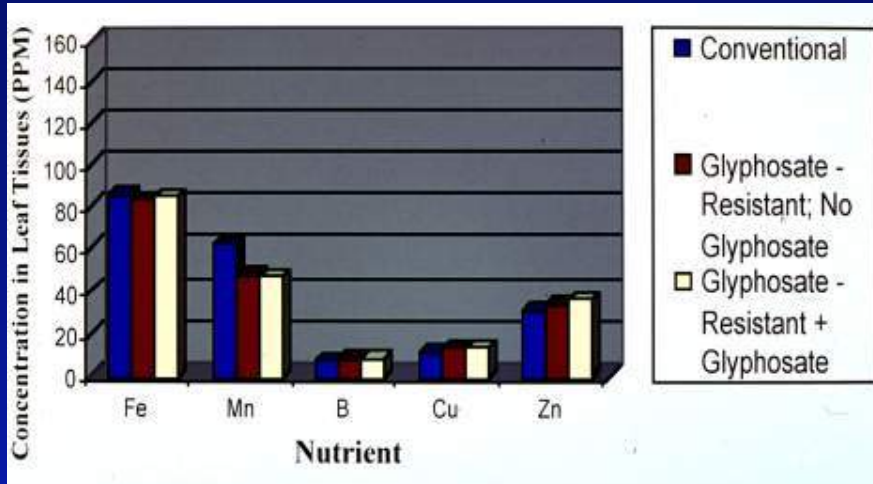
Mis-shaped cotton boll from glyphosate



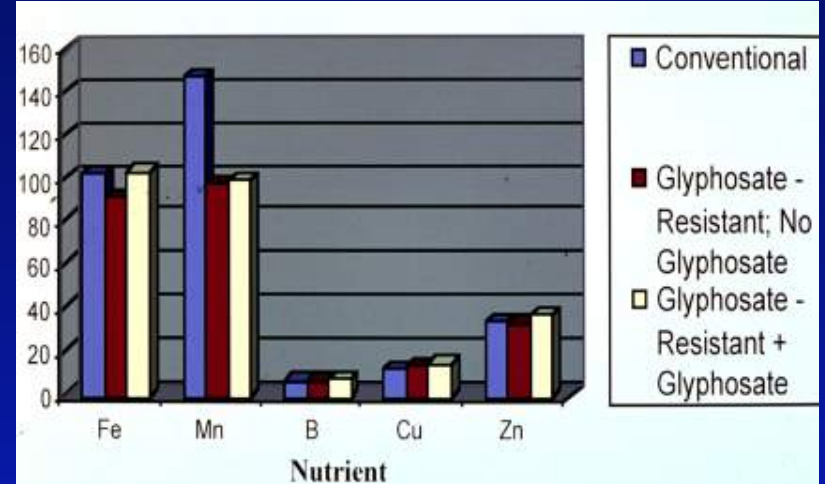
Normal

Glyphosate

“Glyphosate” Gene Effect on Mn Uptake



Soybean micronutrient concentrations



Corn micronutrient concentrations

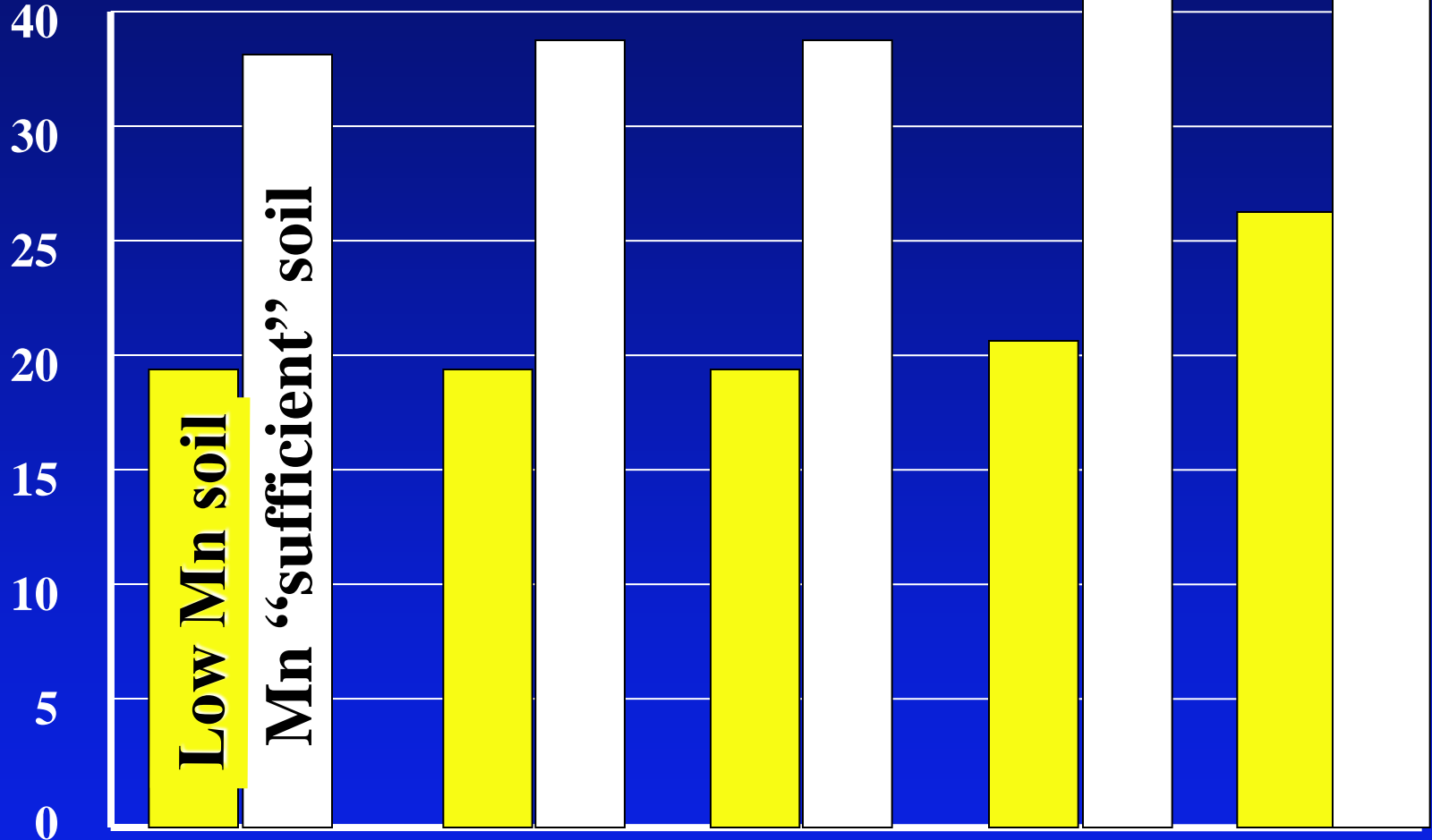
Mn Efficiency of Isogenic soybeans - after Gordon, 2007

Isoline:	KS4202		KS4202 RR		Difference*	
Mn applied (lb./a)	Yield (bu/a)	Tissue Mn (ppm)	Yield (bu/a)	Tissue Mn (ppm)	Yield (bu/a)	Tissue Mn (ppm)
0	76.9	75	64.9	32	-12.0	- 43
2.5	76.1	80	72.8	72	- 4.1	- 3
5.0	74.9	92	77.6	87	+ 0.7	+ 12
7.5	72.6	105	77.6	95	+ 0.7	+ 10

* Difference compared with 0 Mn of normal

Residual Chelation Effect of Glyphosate on Mn

PPM Mn in tissue



None - 4 days Same time + 4 days + 9 days
Time Mn Applied Relative to Glyphosate (UltraMax®)

REPORTED EFFECTS OF GLYPHOSATE

- **Reduced Mn & Fe uptake***

Root & foliage
[K reduced also]

- **Immobilization of Mn***

Translocation
Reduced physiological efficiency

- **Reduced root nodulation & N-fixation***

- **Soil Microflora changes - Root exudates**

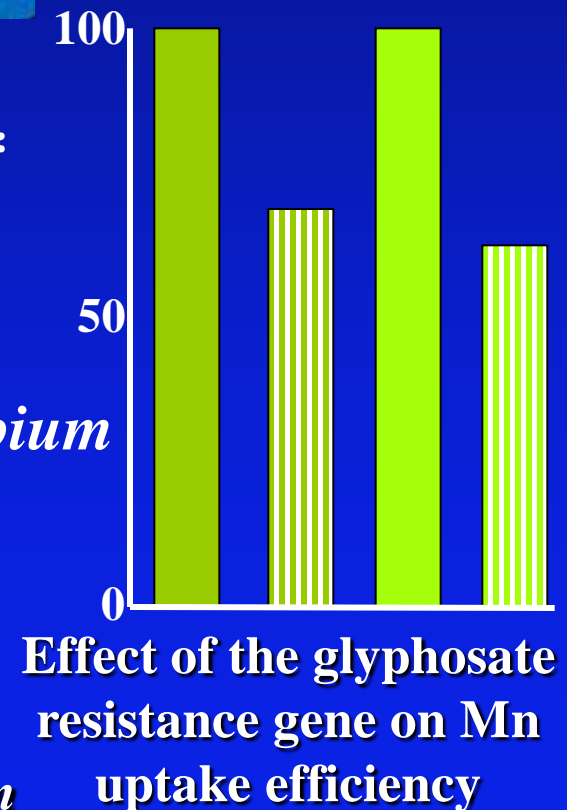
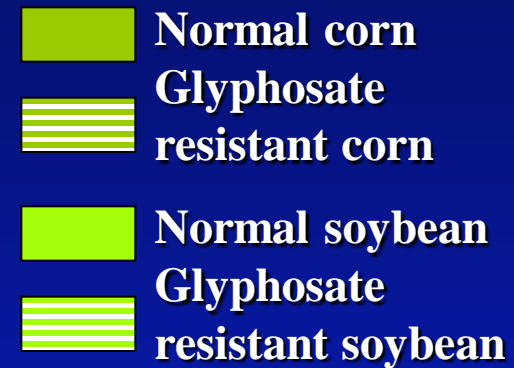
Stimulatory to *Fusaria*, oxidizers, etc.
Toxic to manganese reducers and *Rhizobium*

- **Increased drought stress***

- **Earlier maturity***

- **Interaction with some diseases***

*Can be modified by Mn or other micronutrient application



Some Diseases Increased by Glyphosate

Host plant	Disease	Pathogen
Apple	Canker	<i>Botryosphaeria dothidea</i>
Banana	Panama	<i>Fusarium oxysporum</i> f.sp. <i>cubense</i>
Barley	Root rot	<i>Magnaporthe grisea</i>
Beans	Root rot	<i>Fusarium solani</i> f.sp. <i>phaseoli</i>
Bean	Damping off	<i>Pythium</i> spp.
Bean	Root rot	<i>Thielaviopsis bassicola</i>
Canola	Crown rot	<i>Fusarium</i> spp.
Canola	Wilt (New)	<i>Fusarium oxysporum</i> , <i>F. avenaceum</i>
Citrus	CVC	<i>Xylella fastidiosa</i>
Cotton	Damping off	<i>Pythium</i> spp.
Cotton	Bunchy top	Manganese deficiency
Cotton	Wilt	<i>F. oxysporum</i> f.sp. <i>vasinfectum</i>
Grape	Black goo	<i>Phaeomoniella chlamydospora</i>
Melon	Root rot	<i>Monosporascus cannonbalus</i>
Soybeans	Root rot	<i>Corynespora cassicola</i>
Soybeans	Target spot	<i>Corynespora cassicola</i>
Soybeans	SDS	<i>Fusarium solani</i> f.sp. <i>glycines</i>
Sugarcane	Decline	<i>Marasmius</i> spp.
Tomato	Wilt (New)	<i>Fusarium oxysporum</i> f.sp. <i>pisi</i>
Various	Canker	<i>Phytophthora</i> spp.
Weeds	Biocontrol	<i>Myrothecium verucaria</i>
Wheat	Bare patch	<i>Rhizoctonia solani</i>
Wheat	Glume blotch	<i>Septoria</i> spp.
Wheat	Root rot	<i>Fusarium</i> spp.
Wheat	Head scab	<i>Fusarium graminearum</i>
Wheat	Take-all	<i>Gaeumannomyces graminis</i>



Some Diseases Reduced by Glyphosate

<u>Host plant</u>	<u>Disease</u>	<u>Pathogen</u>
Soybean	Rust	<i>Phycopsora pakyrhiza</i>
Wheat	Rust	<i>Puccinia graminis</i>

Plant Pathogens Affected by Glyphosate

Pathogen

Increase:

Botryospheara dothidea
Corynespora cassicola
Fusarium avenaceum
F. graminearum
F. oxysporum f. sp cubense
F. oxysporum f.sp (canola)
F. oxysporum f.sp. glycines
F. oxysporum f.sp. vasinfectum
F. solani f.sp. glycines
F. solani f.sp. phaseoli
F. solani f.sp. Pisi
Gaeumannomyces graminis
Magnaporthe grisea
Marasmius spp.

Pathogen

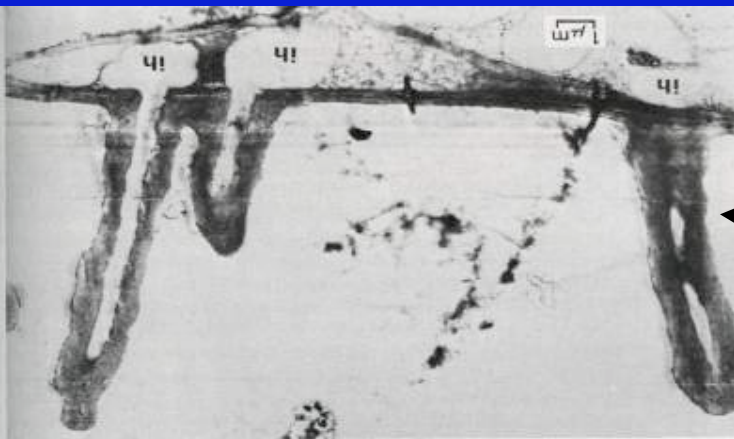
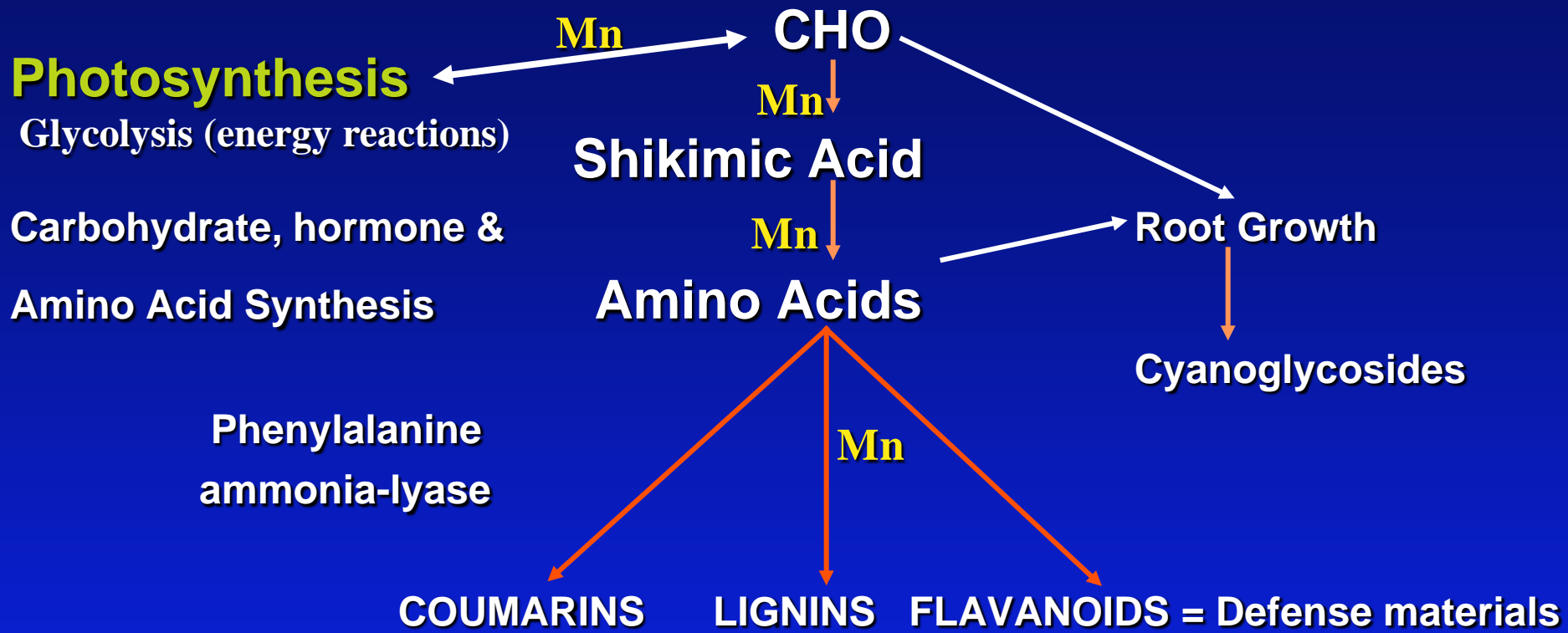
Monosporascus cannonbalus
Myrothecium verucaria
Phaeomoniella chlamydospora
Phytophthora spp.
Pythium spp.
Rhizoctonia solani
Septoria nodorum
Thielaviopsis bassicola
Xylella fastidiosa

Decrease (obligate pathogens):

Phykopsora pakyrhiza
Puccinia graminis

Abiotic increase: Mn deficiency diseases

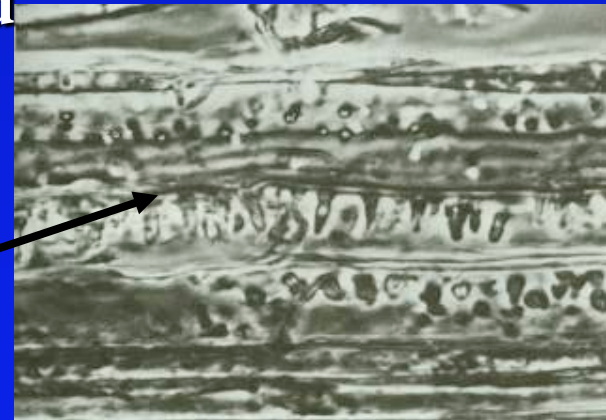
Physiologic Roles of Manganese



“Lignituber” formed in response to cell Penetration.

Wheat
Triticale

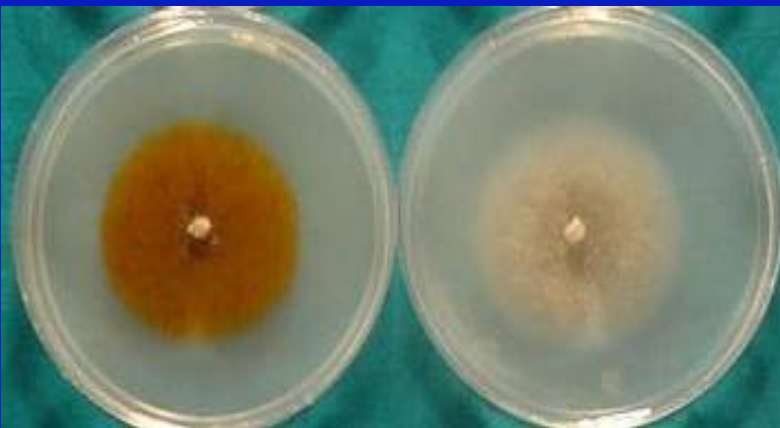
(After Skou, 1975)



Take-all of Cereals

- the Pathogen

- *Gaeumannomyces graminis* var *tritici*
- Common soilborne fungus - endemic world-wide
 - 600 “world” isolates were almost identical in peptidase profiles
 - Can distinguish *Gaeumannomyces graminis* var *tritici* from *G. graminis* var *graminis*
- Virulence associated with manganese oxidation
- Very high tolerance for Mn



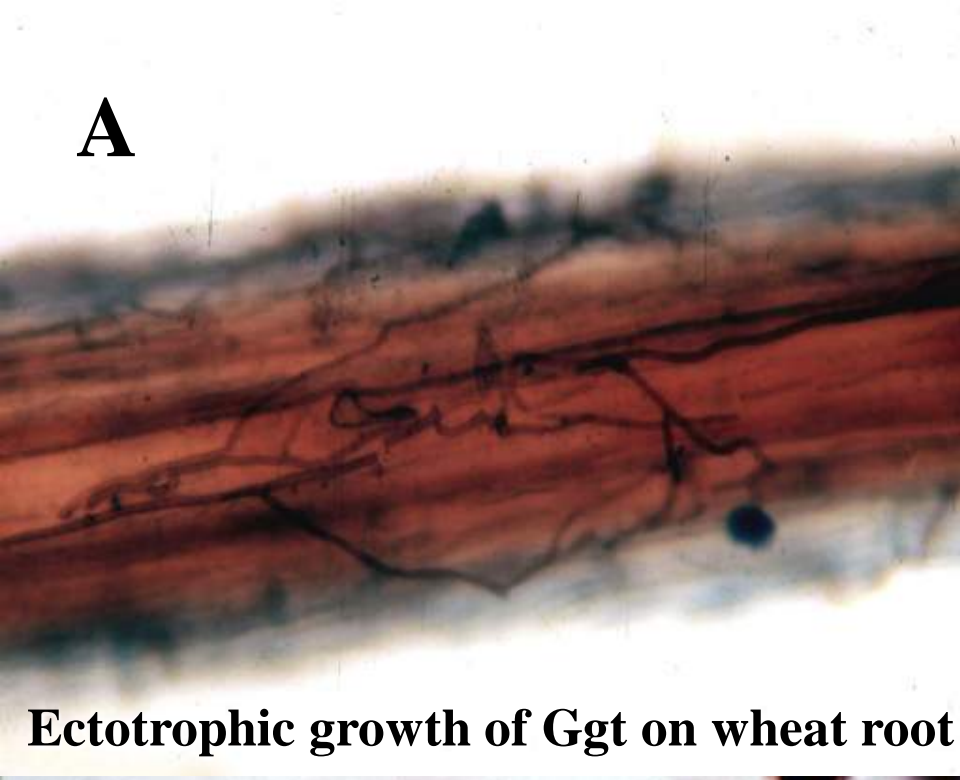
Mn oxidation **No oxidation**
Virulent **Avirulent**

	15C	25C
	to	to
	25C	15C

Temperature	15 C				25 C			
Isolate	X	A	B	C	X	A	B	C
Mn Oxid.	0	0	+	+	0	+	+	0

VIRULENCE AND MANGANESE OXIDATION

A



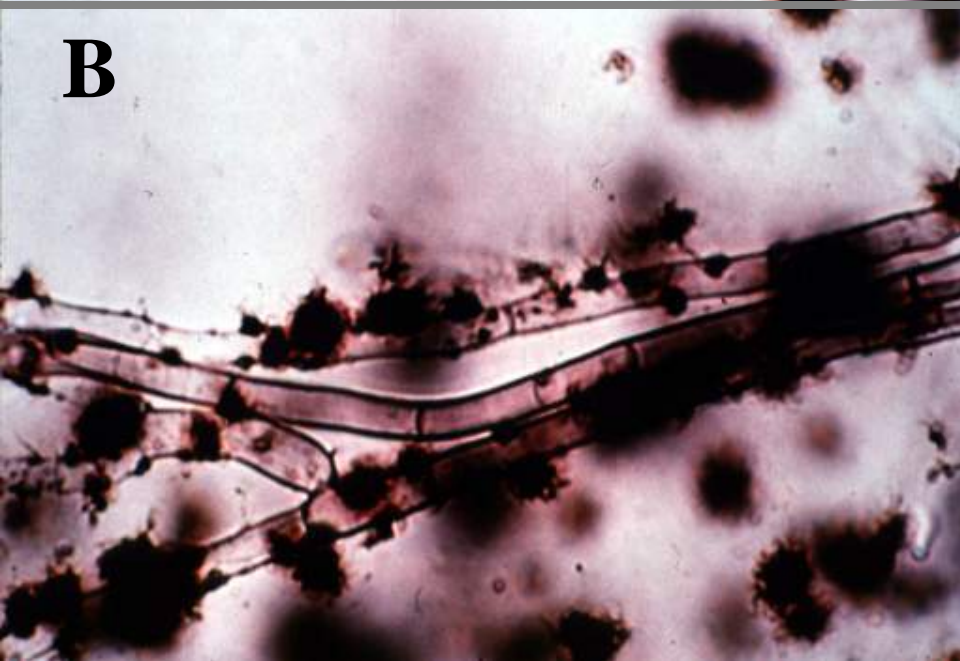
Ectotrophic growth of Ggt on wheat root

The Pathogen

Gaeumannomyces graminis

- A. Ectotrophic growth on root
“Runner” hyphae on wheat root
- B. Extracellular oxidation of Mn
- C. Dispersive X-ray microanalysis
of ectotrophic mycelium on root

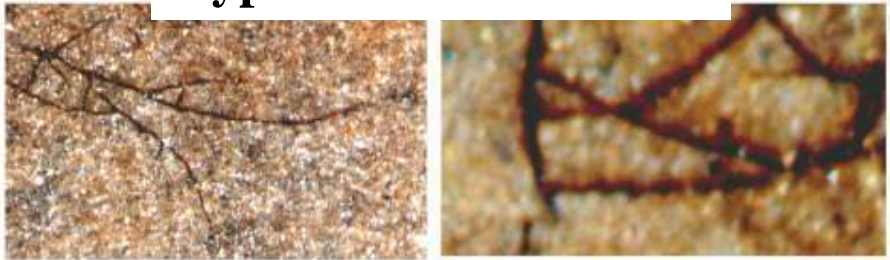
B



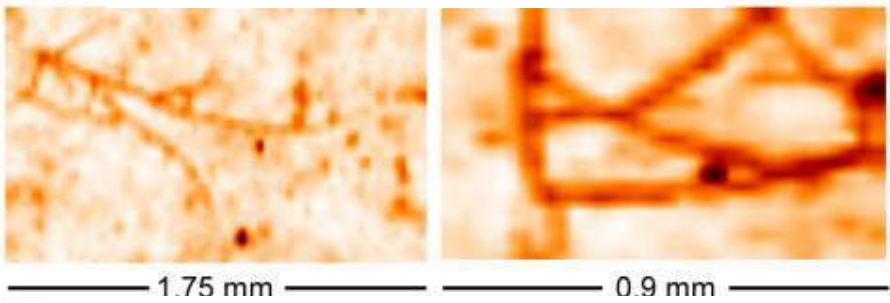
C



Hyphal networks in soil



XANES - MnO₂ distribution



More intense with high soil moisture

Gaeumannomyces oxidizes Mn in Soil, rhizosphere, and root tissue

MnO₂ in wheat root hair cell



Severe take-all spots in wheat



Severe Mn deficiency in double-crop Spybeans after severe take-all

Factors Affecting N Form, Mn Availability and Severity of Some Diseases*

Soil Factor or Cultural Practice	Nitrification	Effect on: Mn Availability	Disease Severity
Low Soil pH	Decrease	Increase	Decrease
Green Manures(some)	Decrease	Increase	Decrease
Ammonium Fertilizers	Decrease	Increase	Decrease
Irrigation (some)	Decrease	Increase	Decrease
Firm Seed bed	Decrease	Increase	Decrease
Nitrification Inhibitors	Decrease	Increase	Decrease
Soil Fumigation	Decrease	Increase	Decrease
Metal Sulfides	Decrease	Increase	Decrease
High Soil pH	Increase	Decrease	Increase
Lime	Increase	Decrease	Increase
Nitrate Fertilizers	----	Decrease	Increase
Manure	Increase	Decrease	Increase
Low Soil Moisture	Increase	Decrease	Increase
Loose Seed bed	Increase	Decrease	Increase

*Potato scab, Rice blast, Take-all, Phymatotrichum root rot, Corn stalk rot

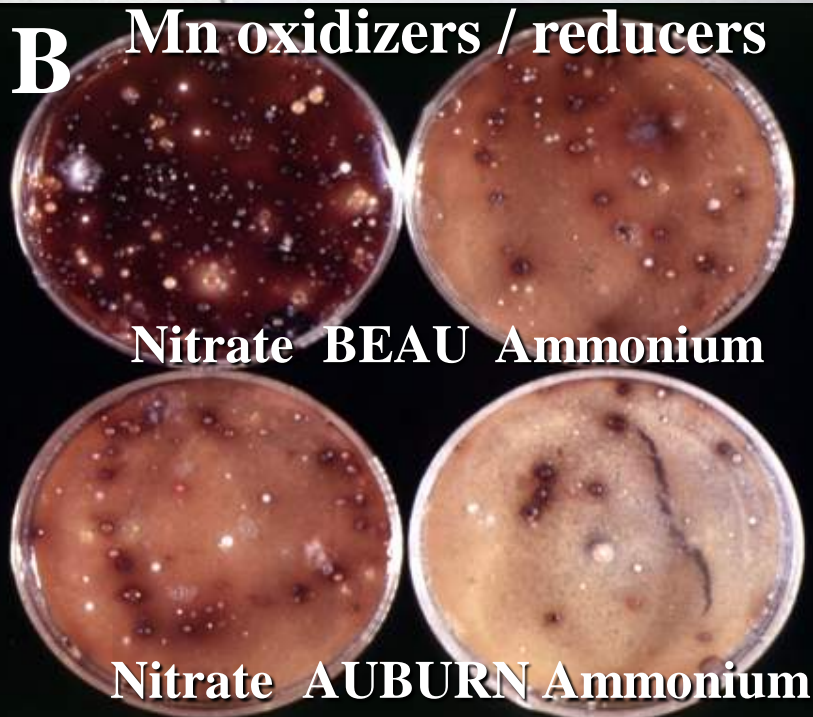


Effect of N form & inhibiting nitrification on take-all and rhizosphere Mn oxidizers

A. N form on Take-all

B. Manganese oxidizers

C. -/+ Nitrification inhibitor



Effect of Cultural Practices on Tissue Mn and Take-all

Cultural Condition	Mn*	TA index
Loose Seedbed	11.2	3.0
Firm Seedbed	19.3	2.4
Nitrification (normal)	8.9	3.2
Inhibiting Nitrification	17.2	2.0
Wheat-wheat- <u>wheat</u>	20.0	4.8
Wheat-oats- <u>wheat</u>	55.0	1.4
Oats-oats- <u>wheat</u>	76.0	0.5

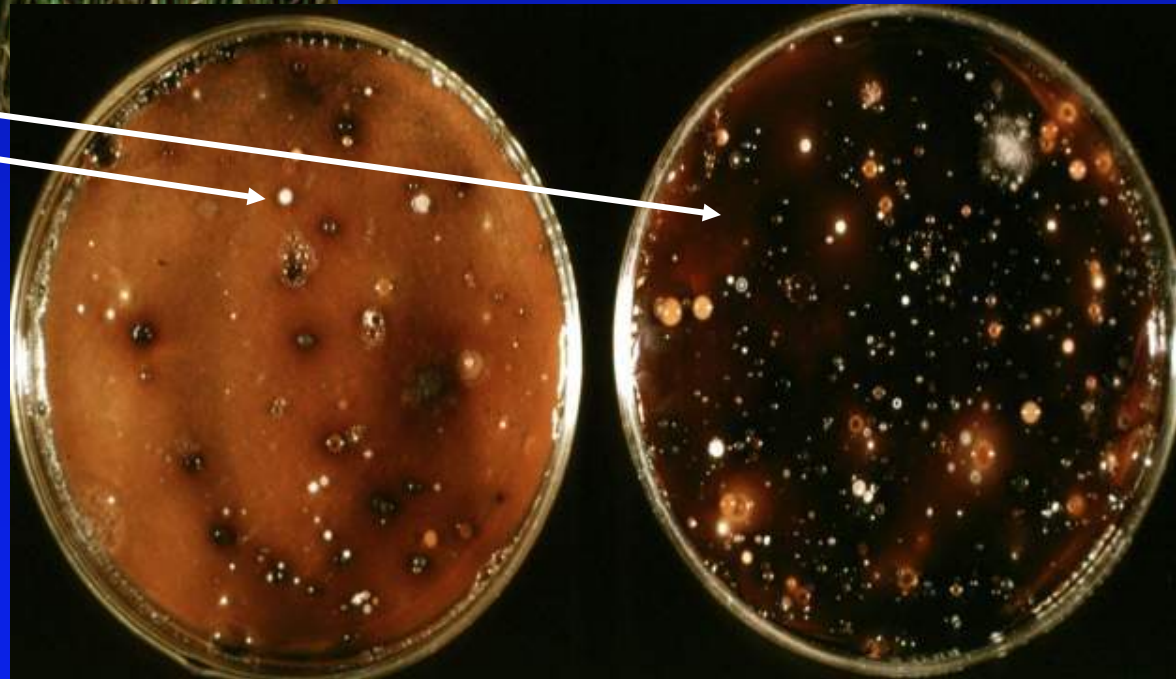


*Wheat tissue Mn, PPM; Take-all index = 1-5 (severe

Take-all and Populations of Mn-oxidizing Rhizosphere Bacteria



↑
Cattle
dung
(manure)



Impact of Glyphosate on Take-all

Take-all of wheat after
glyphosate to RR beans

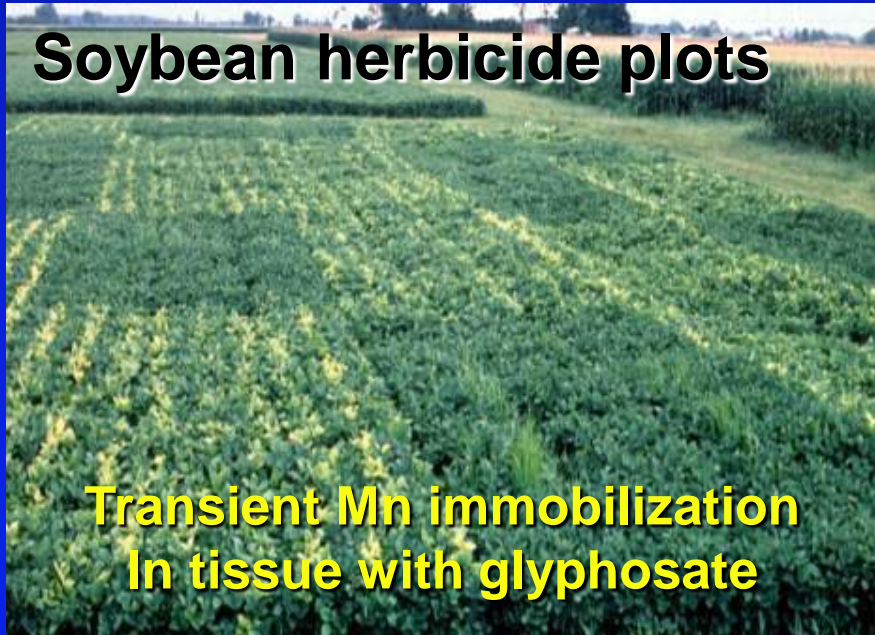


After
glyphosate

No
glyphosate



Soybean herbicide plots



Transient Mn immobilization
In tissue with glyphosate

Wheat after soybeans



After
glyphosate

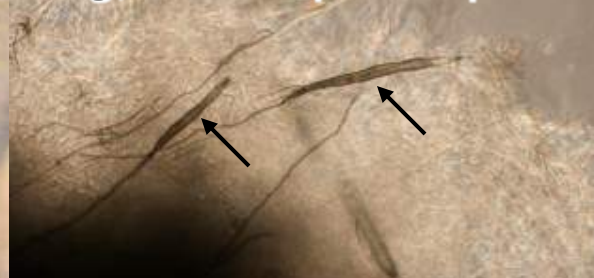
No
glyphosate

Corynespora Root Rot of Soybeans

- ❖ Caused by *Corynespora cassiicola*
- ❖ Dark brown to black rotted small lateral roots & hypocotyl
- ❖ Generally considered “root nibbler” - limited economics
- ❖ Can be severe & also as a foliage pathogen (target spot)



Long, multiseptate spores



Predisposing Effect of Glyphosate on *Corynespora* Root Rot of Soybean



Control

Inoculated

**Inoculated
+ foliar glyphosate**

Effect of Glyphosate from Root Exudates

- Stunted soybean plants adjacent to glyphosate-killed giant ragweed plants
- Very severe *Corynespora* root rot
- Dead ragweed is not a host for *Corynespora*



Dead ragweed plant



Surviving ragweed plant



4-6" 18" away

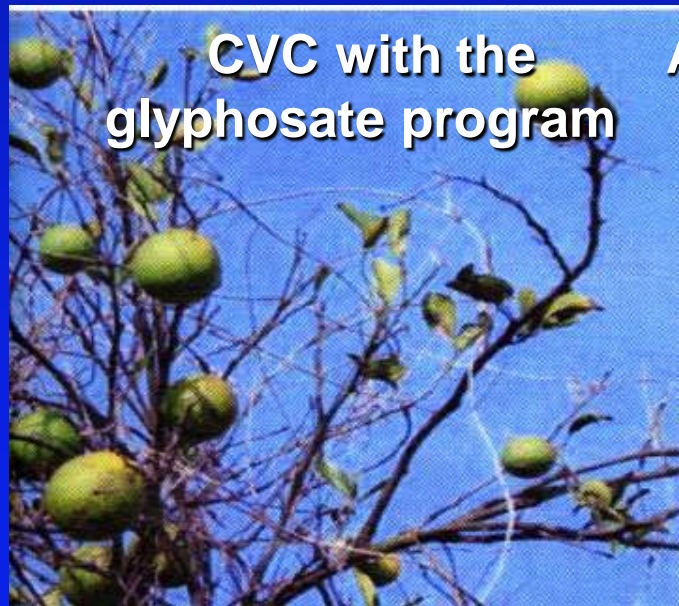
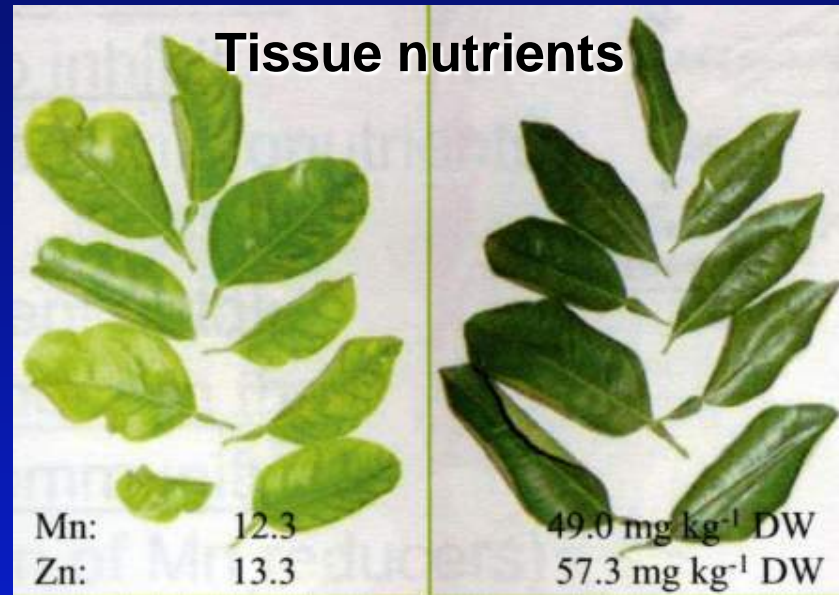
Citrus Variegated Chlorosis

Predisposition to CVC (*Xylella fastidiosa*) by glyphosate



Typical glyphosate
weed control

After T. Yamada



CVC with the
glyphosate program



Alternative mulch program
of T. Yamada

Fusarium Head Scab and Root Rot

- Caused by *Fusarium graminearum* & other *F.* spp.
 - Soilborne fungi
 - Stimulated by glyphosate
- Disease “requires” three “cardinal” conditions
 - Flowering (center of head outwards)
 - Moisture
 - Temperature > 26 C
- Temperature changes C:N ratio (physiology)
- Glyphosate induces similar changes (Mn, Fe, etc.)
- New “Cardinal” conditions:
 - Flowering
 - Moisture
 - Previously applied glyphosate



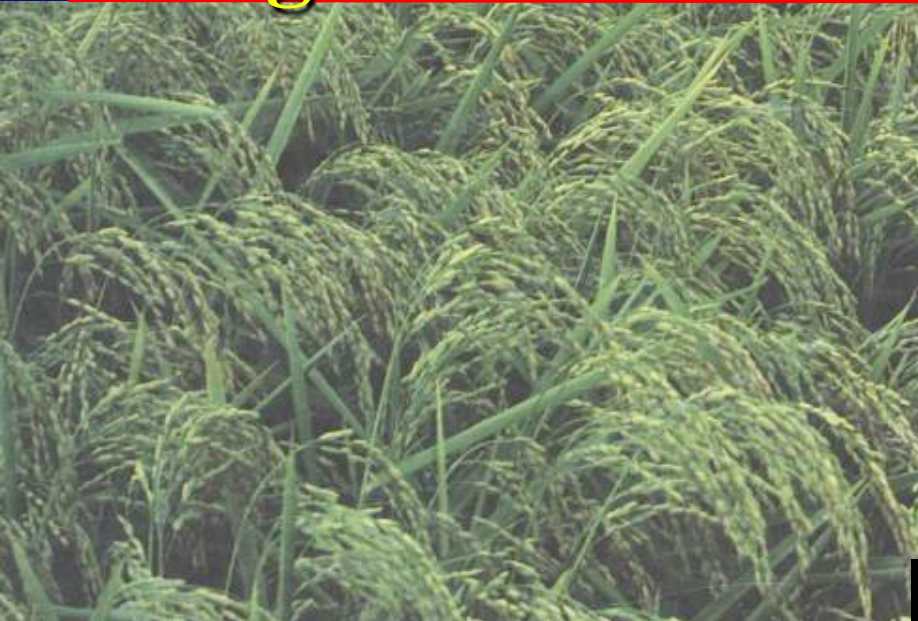
These changes also affect rust for “resistance”

Predisposition of Bean to Root Rot

- Non-nodulating isolines of beans are more resistant to root rot
- Glyphosate reduces nodulation and increases root rot
- Glyphosate increases manganese deficiency

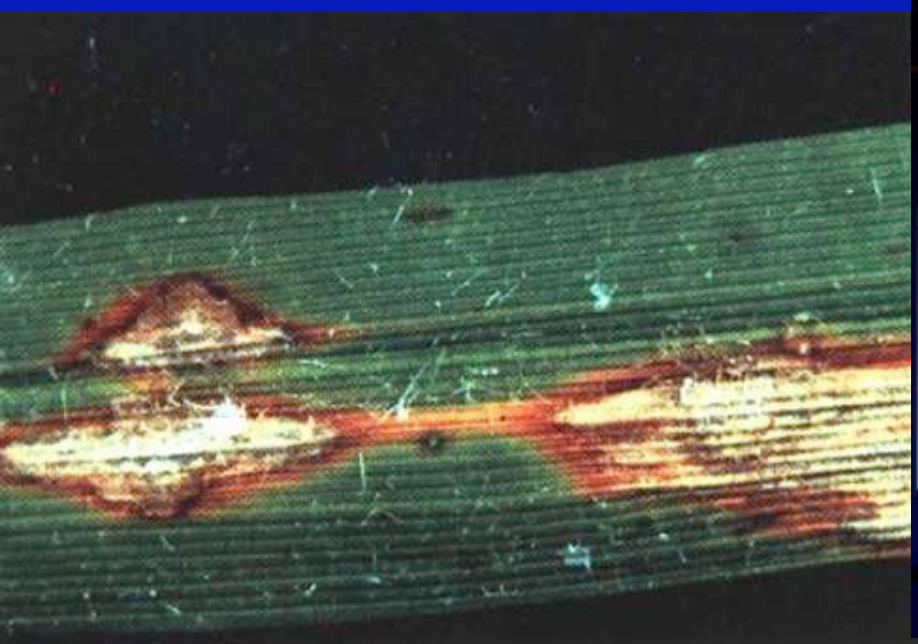


Manganese “Forms” in Blast Infected Rice



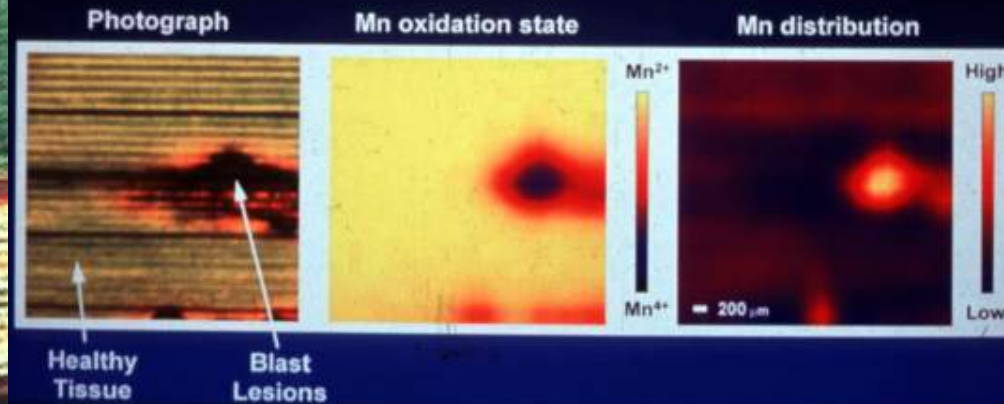
Rice blast, caused by *Pyricularia grisea* (*Magnaporthe grisea*)

Only oxidized Mn in lesion area



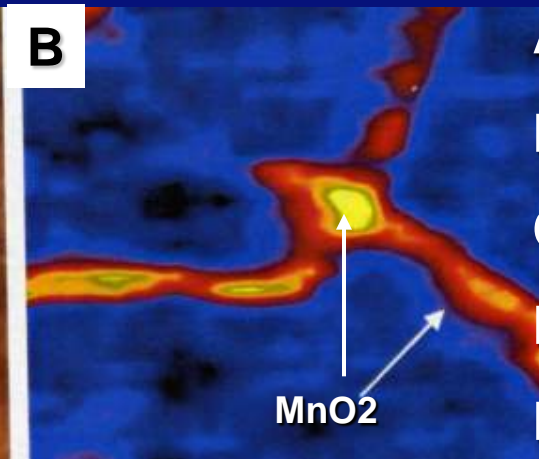
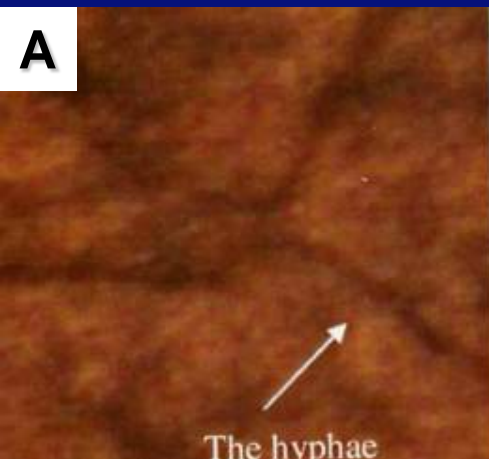
Manganese in Rice Blast Lesions

Rice leaves infected with *P. grisea*



Pathogen induced Mn deficiency in the infection court

Magnaporthe grisea is a strong Mn oxidizer



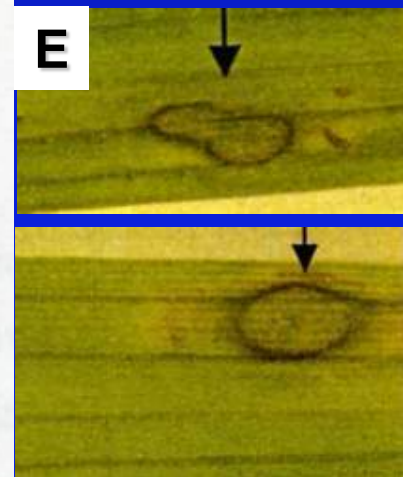
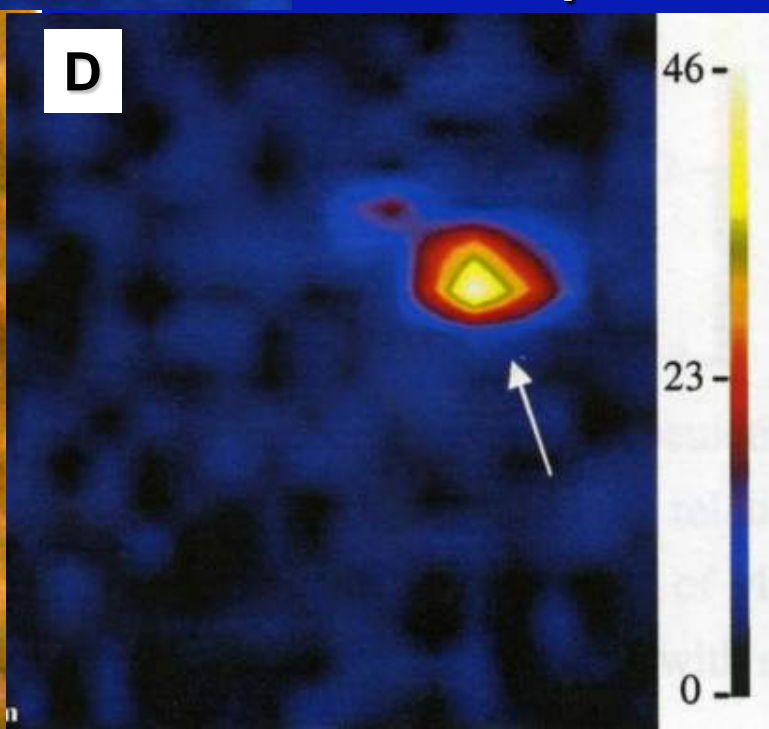
A. Mycelium on leaf surface

B. Micro XANES of MnO₂ in A

C. Blast lesion on leaf

D. XANES of MnO₂ in lesion

E. Lesion produced by toxin



Glyphosate is Reported to Control Rust Diseases

- **Increases resistance**
 - Specific N nutrients withheld
Glycine, phenylalanine, etc.
 - Amino acid inhibitors increased
- **Provides a 20-25 day effect**
- **Blocks specific peptidase activity**
- **May account for the more limited damage from soybean rust than anticipated in the U. S.**



Mechanisms by which Nutrients Reduce Disease

- **Increased Plant Resistance**
 - Physiology - phytoalexin, CHO, phenolic production
 - Defense- callus, lignin, cicatrix formation
- **Disease Escape, Increased Plant Tolerance**
 - Increased growth - roots, leaves
 - Shortened Susceptible stage
 - Compensation for disease damage
- **Modifying the environment**
 - pH, other nutrients
 - Rhizosphere interactions, nitrification, biological balance
- **Inhibited Pathogen Activity**
 - Reduced virulence
 - Direct effect on survival and multiplication
 - Biological control

Strategies to Reduce Mn Immobilization

❖ Amendment Micronutrient

Timing/formulation

Biological amendment

Bacillus, Trichoderma

❖ Detoxification

Calcium chelation - gypsum

Manganese

❖ Cultural practices

Increase Mn availability

Ammonium sources of N

Inhibit nitrification

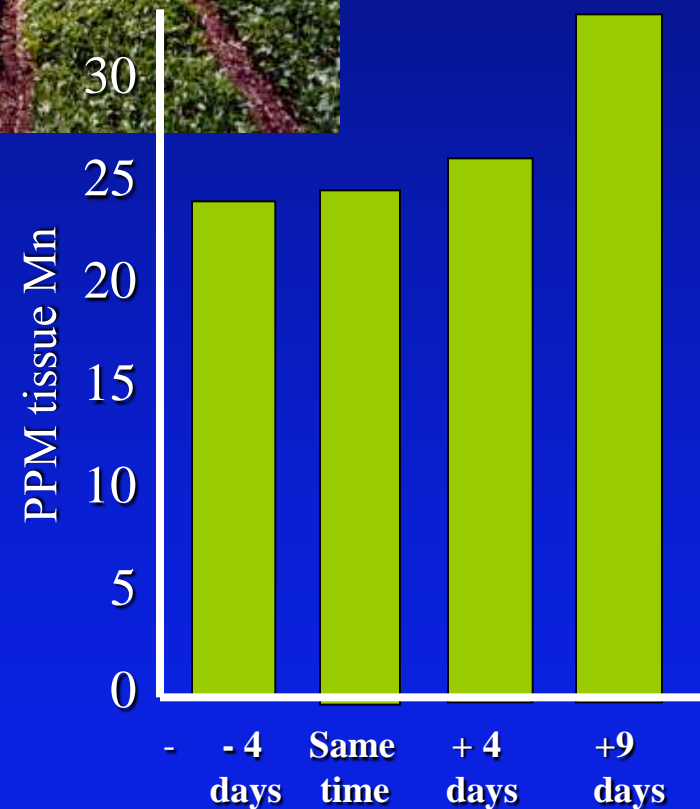
Crop sequence - after corn

Alternative weed control

Mulch

Reduce usage - chemistry

Reduce rates



Time Mn applied relative
to glyphosate (UltraMax®)

Interaction of Micronutrients with Glyphosate*

Micronutrient	Rate	Yield	% Weed control
Untreated control	None	46 a	0 a
Glyphosate** control	24 oz/a	57 b	100 e
Gly+MnCO ₃	0.5 #Mn/a	75 d	91 de
Gly+MnSO ₄	0.5 #Mn/a	70 cd	93 e
Gly+MnEDTA	0.25 #Mn/a	72 cd	100 e
Gly+Mn-AA	0.25 #Mn/a	67 c	85 d
Gly+ZnO	0.5 #Zn/a	49 ab	33 c
Gly+ZnChelate	0.25 #Zn/a	40 a	40 c
Gly+Zn+P	0.5 #Zn/a	41 a	20 b

* Glyphosate WeatherMax® formulation at 24 oz/a + AMS

Biological Amendments to Increase Mn

Microbes: *Bacillus (cereus)*, *Trichoderma (konigii)*

Concerns (other than Mn activity):

Tolerance of glyphosate

Timing

Method of application

Formulation

Safety

Treatment	<u>Corn yield (bu/a)</u>	
	<u>Rainfed</u>	<u>Irrigated</u>
None	176a	186a
Bio # 1	181ab	187a
Bio # 2	185b	186a

Detoxifying Glyphosate

➤ In meristematic/reproductive tissues

Mn, Si+Mn, Mn+Cu foliar fertilization

➤ In root exudates in soil

➤ Approach:

Broadcast:

Lime

Gypsum

Phosphorus

In furrow treatment:

Gypsum (CaSO₄)

Lime

Manganese

Ca + Mn

Effect of in-furrow treatments on Soybean tissue Mn		
Treatment	Rainfed	Irrigated
Lime	32a	29a
Gypsum	38b	36b

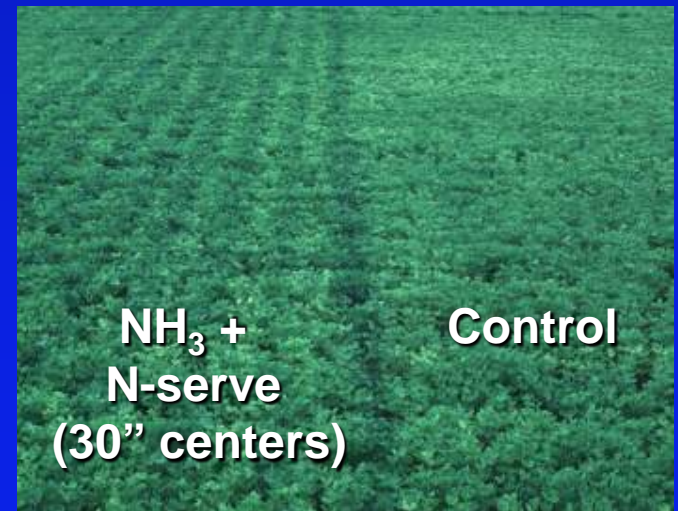
Modify Cultural Practices to Affect Mn Availability

- ✓ Crop sequence
- ✓ Firm seedbed
- ✓ Grass mulch
- ✓ Lower pH
- ✓ Moisture management
- ✓ Ammonium N
- inhibiting nitrification

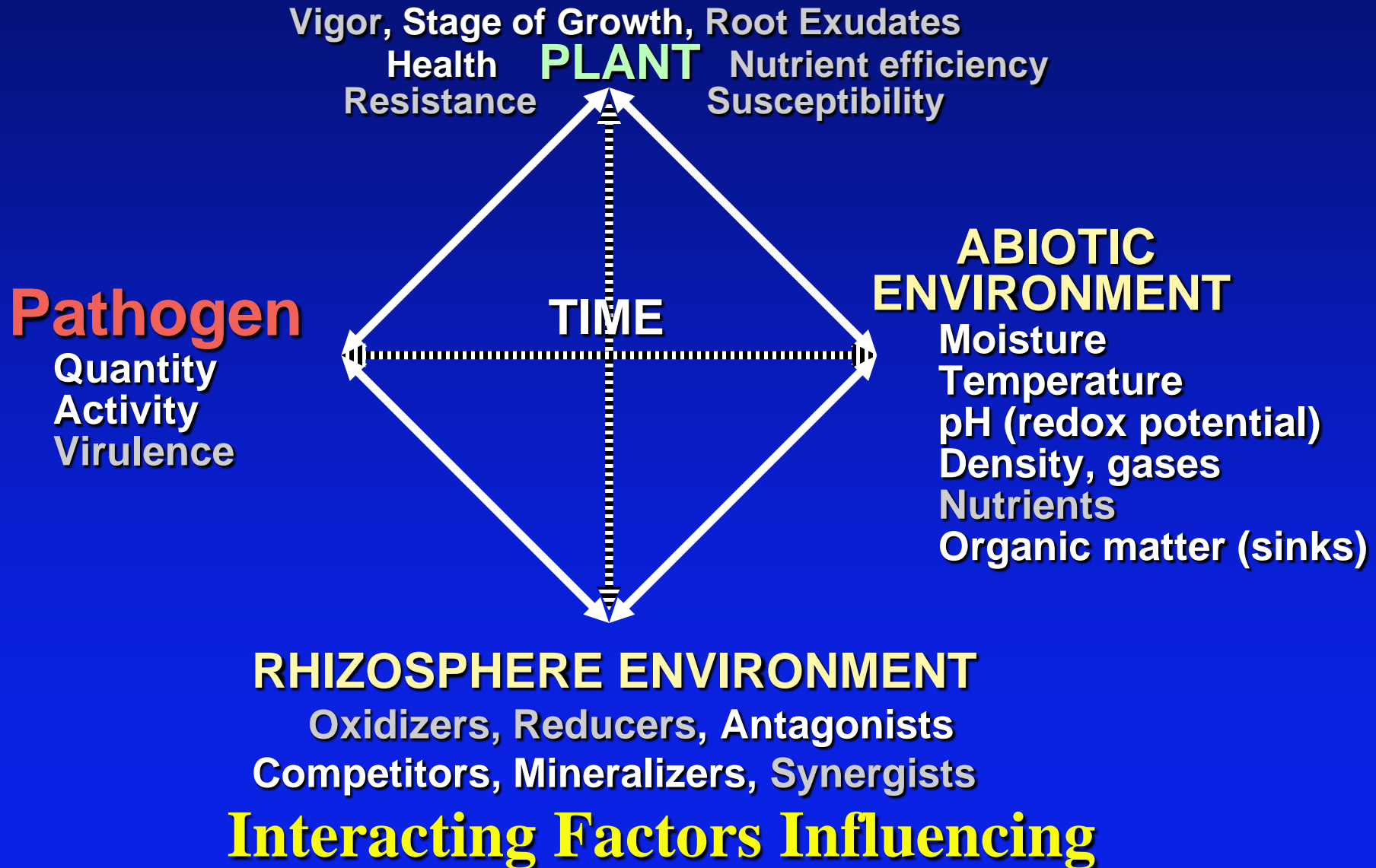
Residual effect of NH_3 for corn on Mn availability for soybean*

Treatment	Tissue Mn	Bean Yld (bu/a)
None	12.1	22
NH_3 only	14.3	26
NH_3+Mn	---	39
NH_3+NI	30.1	44
$\text{NH}_3+\text{NI}+\text{Mn}$	---	44

* NH_3 on 15" centers



GLYPHOSATE: A simple Compound with Profound Effects on Nutrients & Disease



Summary of Glyphosate Effects

- Physiology of the plant
 - Nutrient composition
 - Inorganic micronutrients
 - Organic - N compounds (amino acids, etc.)
 - Nutrient efficiency
 - Defense compounds
- Environment
 - Nutrient availability, form, uptake
 - Rhizosphere microbial activity and balance
- Pathogen
 - Virulence, biological synergy

Conclusions & Recommendations

- 1.** The glyphosate-resistance gene selectively reduces Mn uptake
Select cultivars with highest Mn efficiency
- 2.** Application of glyphosate reduces Mn translocation in tissues
Apply micronutrients 8+ days after glyphosate
- 3.** Glyphosate formulation and nutrient source influence uptake
Select formulations that are compatible for uptake
- 4.** Changes in rhizosphere biology are accumulative
Use cultural practices that minimize glyphosate impact
Use a non-systemic herbicide
- 5.** Glyphosate reduces root growth
Detoxify glyphosate in roots and rhizosphere
- 6.** Severity of some diseases increase with glyphosate
Use alternate weed control -Minimize glyphosate use