## Role of Micronutrients in Plant Resistance to Diseases

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SIMPÓSIO SOBRE RELAÇÕES ENTRE NUTRIÇÃO MINERAL E INCIDÊNCIA DE DOENÇAS DE PLANTAS





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### Plant Diseases: Global Constraint to Crop Production

- Crop plants are frequently subjected to number of diseases and pests, leading to large economic losses in crop production globally.
- Chemical control of disease (e.g., use of fungicides) is a widely applied approach to minimize disease-related problems in crop production.

# Problems associated with chemical control:

- -Threat to ecosystem and human health,
- Increases in production costs and
- Induction of pathogen resistance to chemicals following their long-term use.
- Not always successful

### Need for alternative approaches

- Breeding
- Biological control
- Management

Irrespective of the approaches applied in disease control, mineral nutritional status of plants substantially affects plant resistance and tolerance to pathogens, and must be considered in all type of disease management systems.

Increasing evidence is available showing that mineral nutrient deficiency or imbalance in mineral nutrient supply (e.g., excess of N and low supply with K and micronutrients..) greatly increases susceptibility of crop plants to pathogen attack.

Mineral nutritional status of plants is very often ignored or unrecognized in practical agriculture and even in breeding programs.

### This is especially true for micronutrients

(Graham and Webb, 1991, SSSA Publications; Huber and Graham; 1999, Howarth Press)

### **EXUDATION OF ORGANIC COMPOUNDS AND PATHOGENIC INFECTION**

One of the important factors in infection of plant tissues is the availability of exudates released from the cells into apoplast of root or leaf tissues.

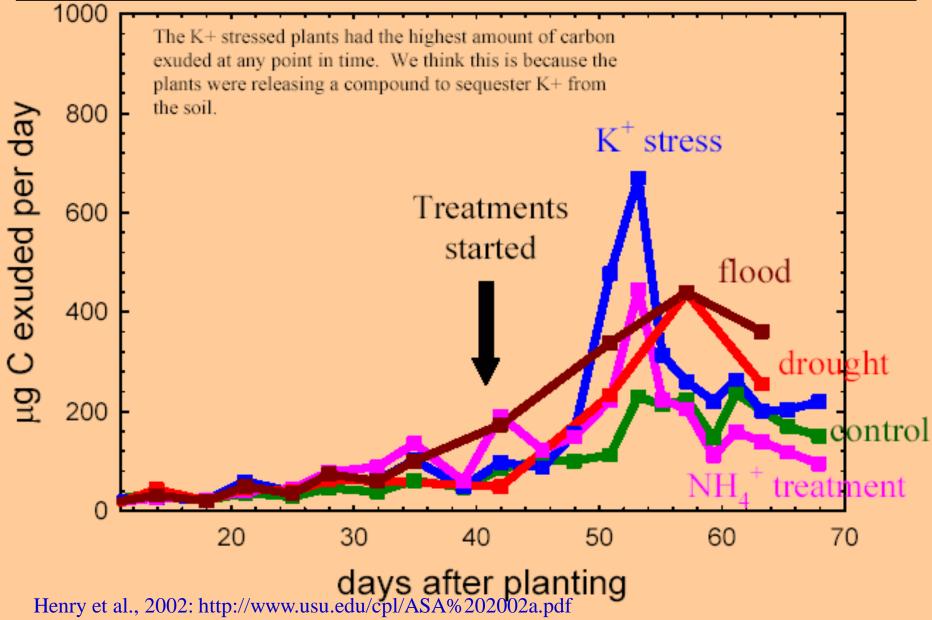
Exudates such as carbohydrates and amino acids are decisive factors in attraction and rapid spread of pathogens in plant tissues. Release of exudates from cells is highly dependent on structural integrity (stability) of cell membranes.

Any impairment in structural integrity of cell membranes induces membrane permeability and extensive release of exudates

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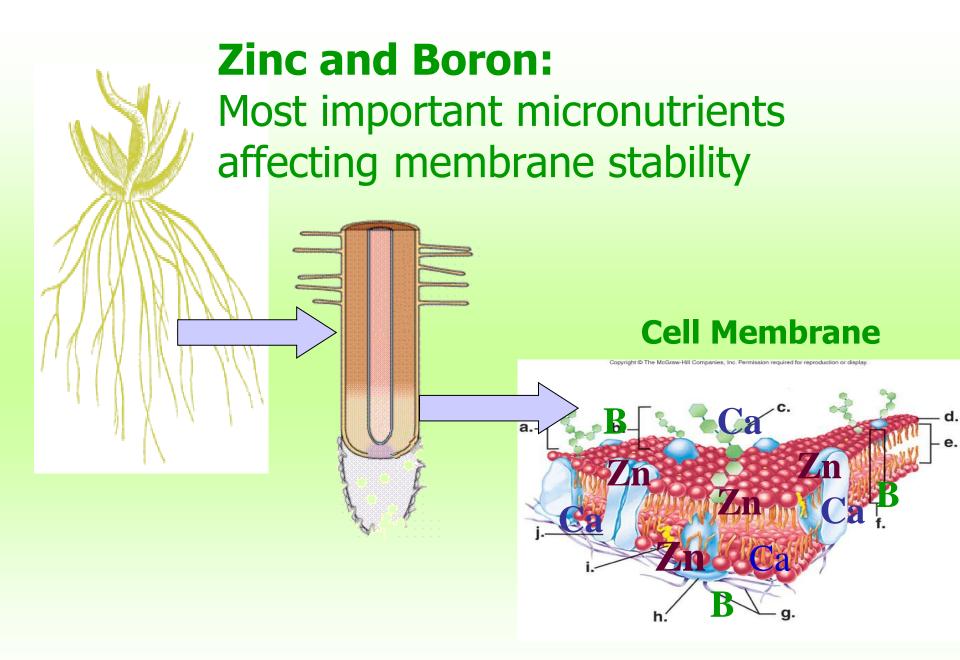
#### Several Abiotic Stress Factors Induce Membrane Permeability



# Cumulative carbon exuded per gram dry plant

	mg C exuded per g dry plant		Percent
	Average	Std. dev.	of control
control	2.6	0.4	
NH4+	2.3	0.1	90
K+	3.7	0.6	144
flood	3.8	0.9	145
drought	4.4	0.5	170

Henry et al., 2002: http://www.usu.edu/cpl/ASA%202002a.pdf

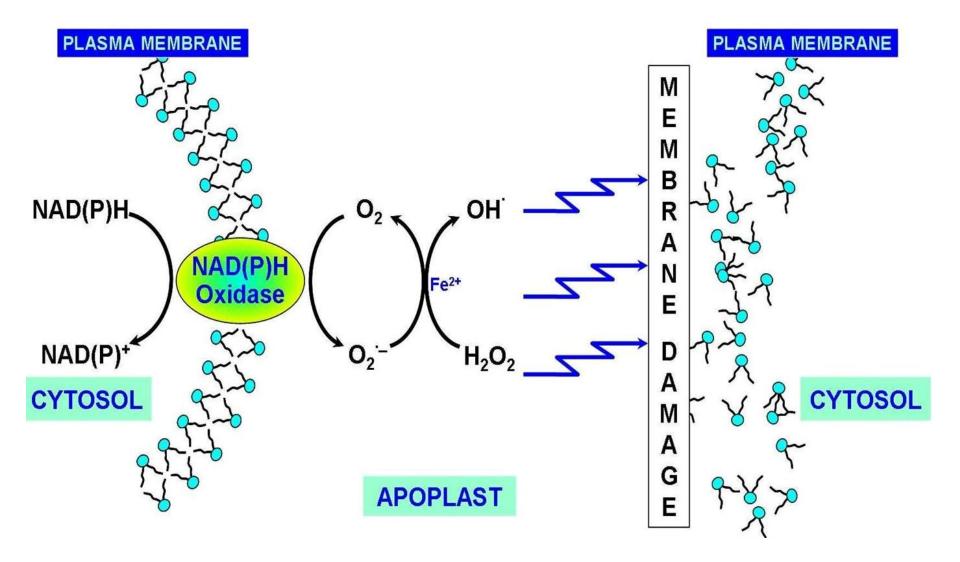


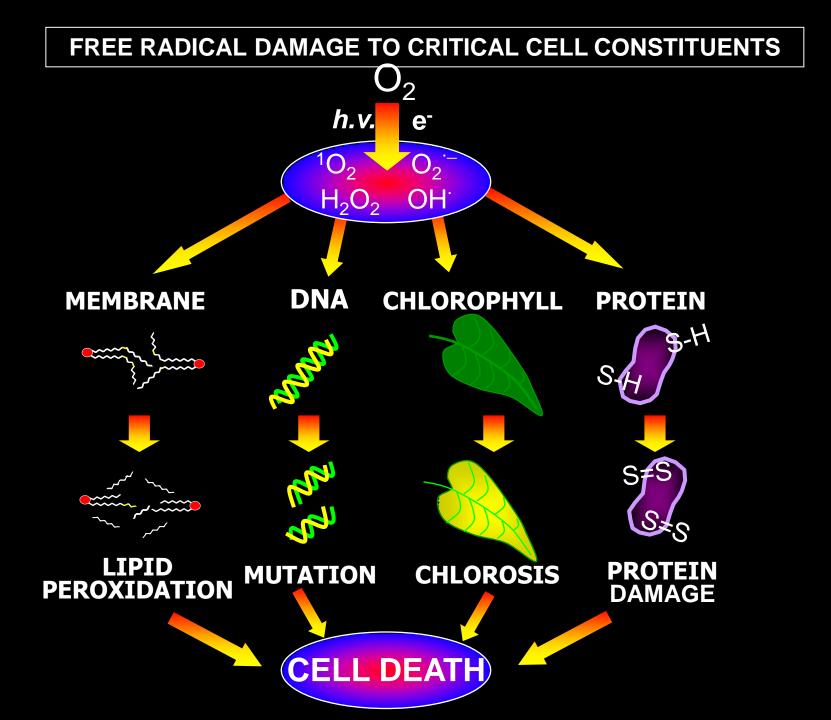
#### Root exudation of organic compounds in cotton, wheat and apple at different Zn supplies

Zn Treatment	Amino acids	Sugars	Phenolics
	(µg g <sup>-1</sup> root 6h <sup>-1</sup> )		
		COTTON	
-Zn	165	751	161
+Zn	48	375	117
		WHEAT	
-Zn	48	615	80
+Zn	21	315	34
		APPLE	
-Zn	55	823	350
+Zn	12	275	103

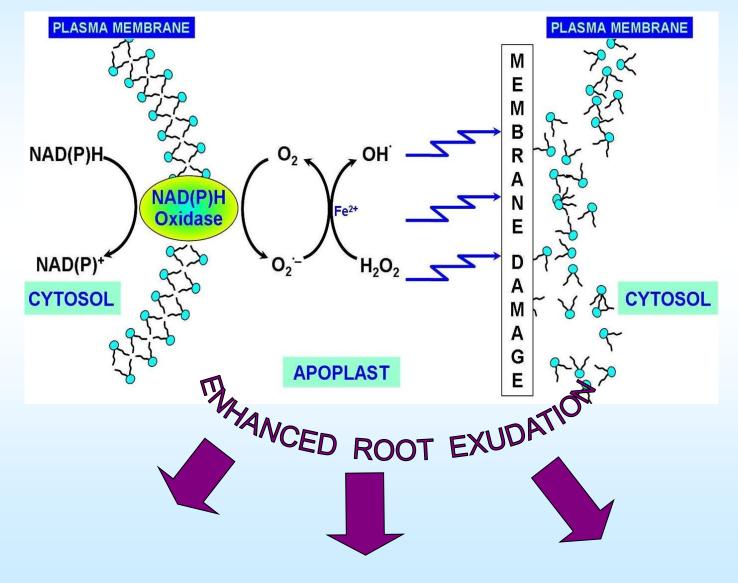
Cakmak and Marschner, 1988, J. Plant Physiol.

#### Zinc Deficiency-Induced NADPH-Dependent Superoxide Radical Generation and Membrane Damage





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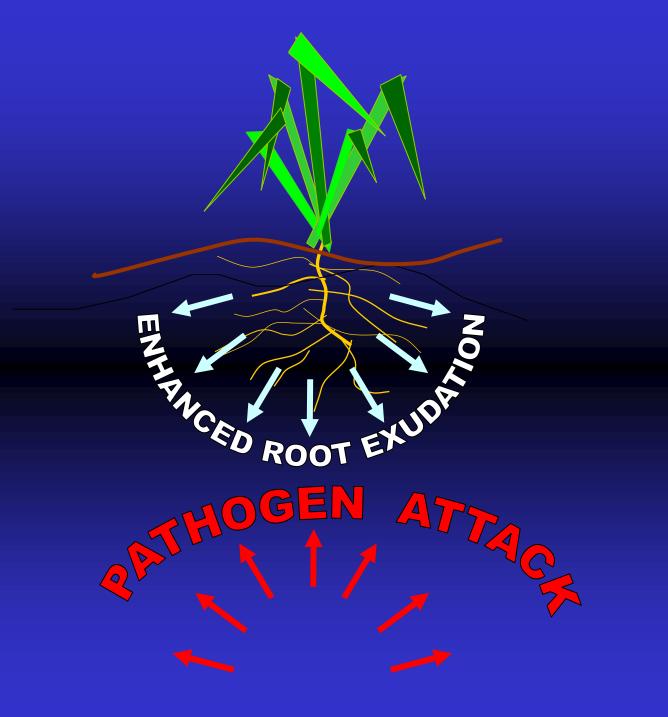


### Boron Deficiency-Induced Membrane Permeability

# Leakage of sucrose, phenolics and amino acids from sunflower leaves as influenced by B supply

B supply	Sucrose	Phenolics	Amino acids
(µM)		(µg g <sup>-1</sup> FW [2h] <sup>-1</sup> )	
0.01	900	79	163
0.20	440	72	122
1.0	70	17	33
20.0	20	13	23

Cakmak et al., 1995, Physiol. Plant.

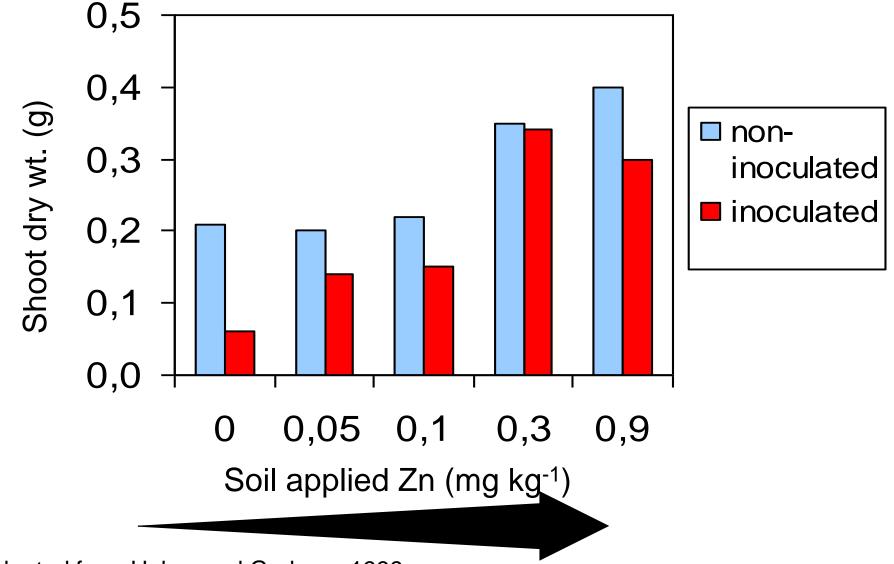


#### Effect of Zn application on phytophtora zoospores on roots of two different Eucalyptus

	Species		
Zn supply	E. marginata	E. sieberi	
	(No./mm <sup>2</sup> root)		
+Zn	<b>4</b> ±1	<b>89</b> ±13	
-Zn	<b>44</b> ±8	<b>489</b> ±48	

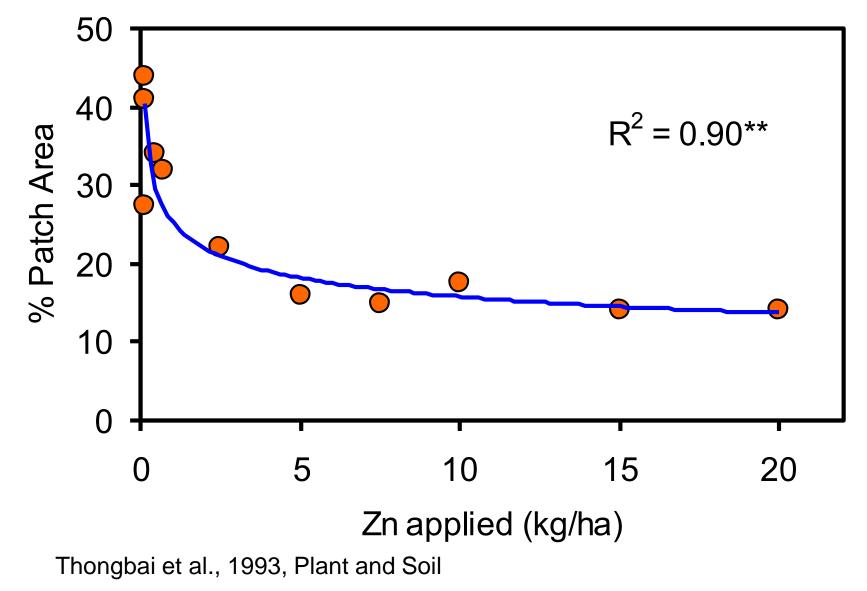
Graham and Webb, 1991

# Effect of inoculation with *R. solani* on shoot dry weight of *M. truncatula* plants as affected by increasing Zn supply

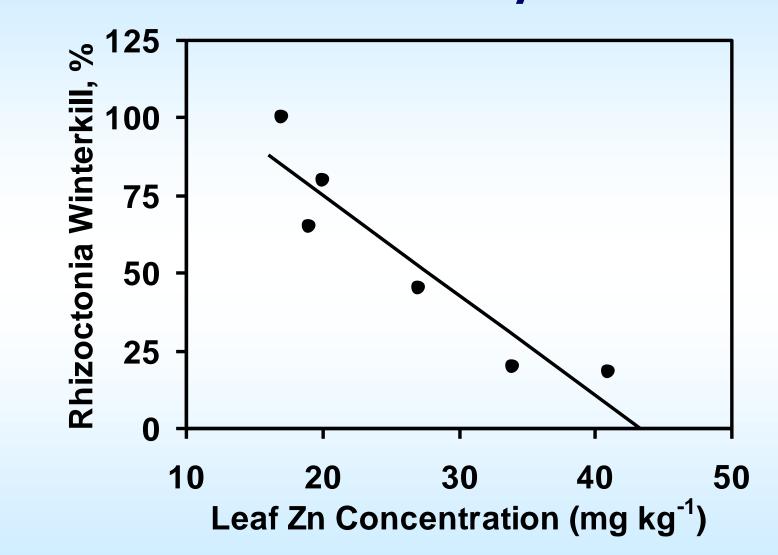


Adapted from Huber and Graham, 1999

#### **Correlation between Zn application and bare patch caused by Rhizoctonia in wheat**

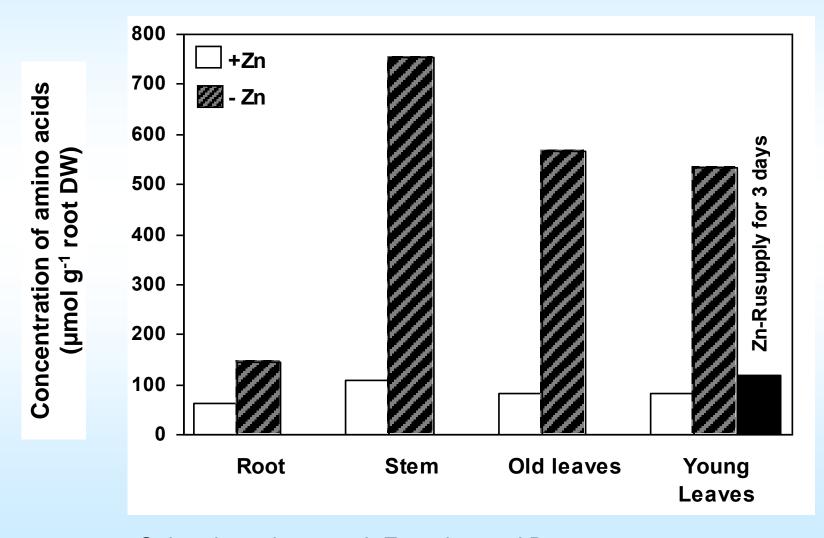


#### Relationship between leaf Zn concentration and Winterkill caused by Rhizoctonia



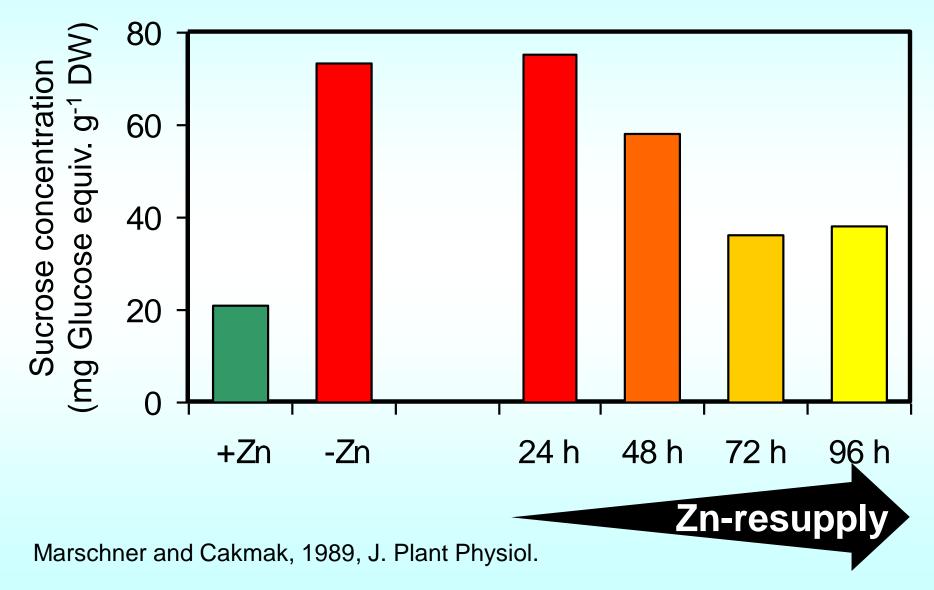
adapted from Huber and Graham, 1999

#### Free amino acids in bean leaves at different Zn treatments

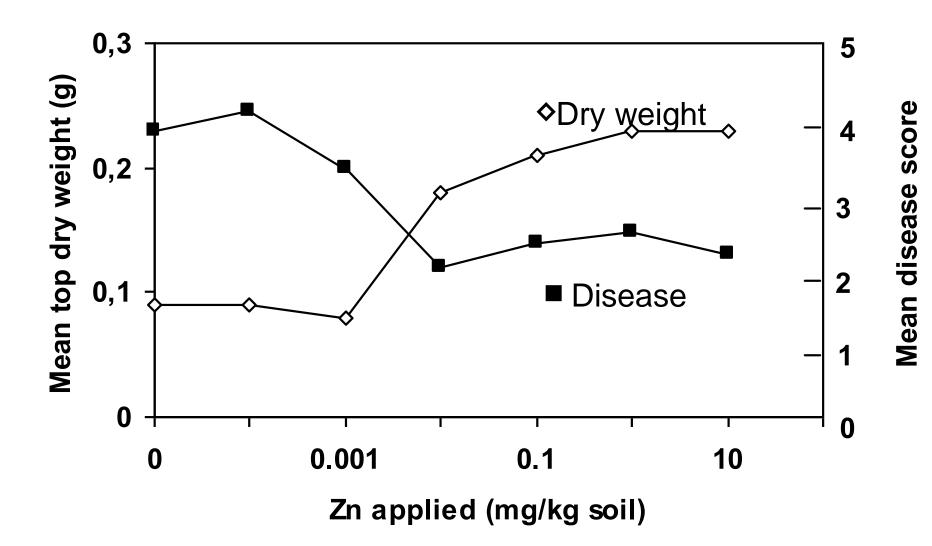


Cakmak et al. 1989, J. Experimental Botany

# Sucrose concentration in leaves at different Zn treatments



# Relationship between Zn supply, shoot growth and disease score in wheat



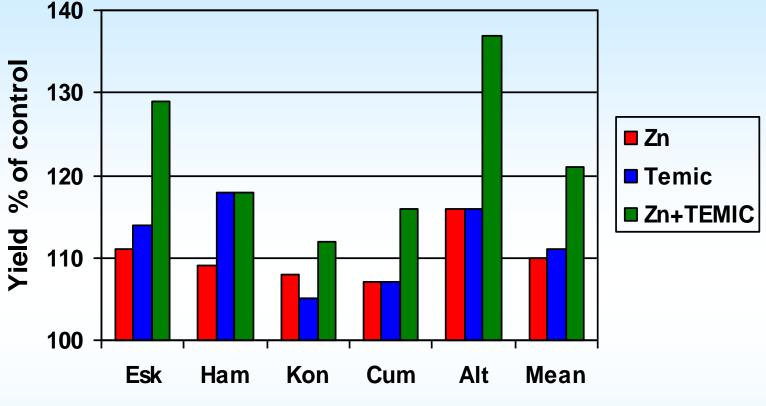
# Zn inhibits root nematode invasion in different crop species

(Shaukat and Siddiqui, (2003, *Lett. Appl. Microbiol*. 36: 392-398; Siddiqui et al., 2002, *J. Phytopathol*. 150: 569-575; Hamid et al., 2003, *Lett. Appl. Microbiol*. 36: 239-244).

The suppression of nematode penetration by adding Zn was most distinct when Zn was applied in combination with diseasesuppressive strains of Pseudomonas.

It seems that Zn is able to stimulate production of antimicrobial compounds by disease-suppressive strains of *Pseudomonas fluorescens* 

#### Zinc improves effect of nematicide



#### LOCATIONS

Effect of 90 kg ZnSO4 and Temik (T: nematicid) Application on grain yield of Bezostaya at 5 locations in Central Anatolia. Control: grain yield without Zn and Temik application

(Unpublished results; data from M.Kalayci and F. Yildirim)

#### Effect of increasing Zn supply on root infection/penetration of *M. phaseolina*, *F. solani* and *R. Solani* with and without *Ps. aeruginosa* IE-6S+ biological control agent

	Inf	fection (%)	
Zn concentration (mg/kg soil)	M. phaseolina	F. solani	R. solani
Without IE-6S <sup>+</sup>			
0	55	88	42
0.2	55	92	33
0.4	42	75	55
0.8	33 🗡	66 🗡	22 🗡
1.6	33	66	17
With IE-6S <sup>+</sup>	-	-	-
0	42	77	33
0.2	33	66	17
0.4	44	58	22
0.8	33 🗸	44 🔶	8 🗸
1.6	18	50	8

Siddique et al., 2002, J. Phytopathol.

### Effect of increasing Zn supply on nematode population with and without *Ps. aeruginosa* IE-6S<sup>+</sup> biological control agent

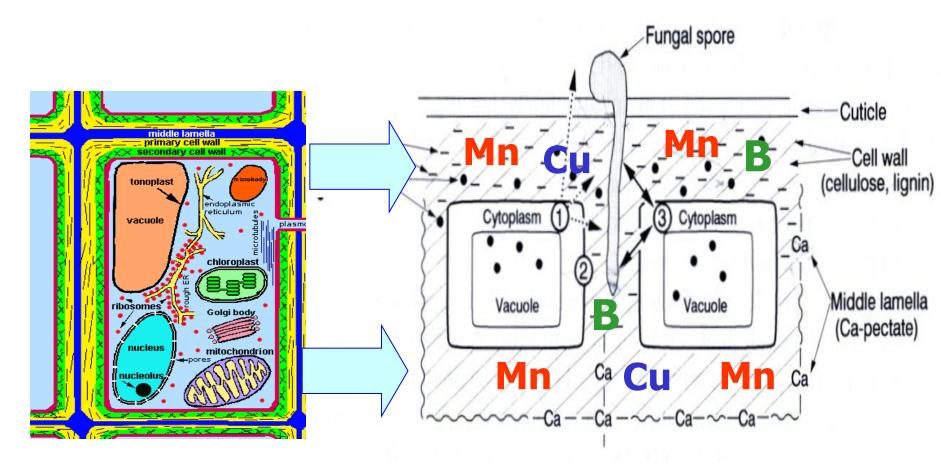
Zn concentration (mg/kg soil)	Nematode population Per gram root	
Without IE-6S⁺		
0	150 🛛	
0.2	163	
0.4	140	
0.8	124	
1.6	117 🔶	
With IE-6S⁺		
0	97	
0.2	85	
0.4	81	
0.8	70	
1.6	65	

Siddique et al., 2002, J. Phytopathol.

Manganese is also highly effective micronutrient in plant resistance to diseases

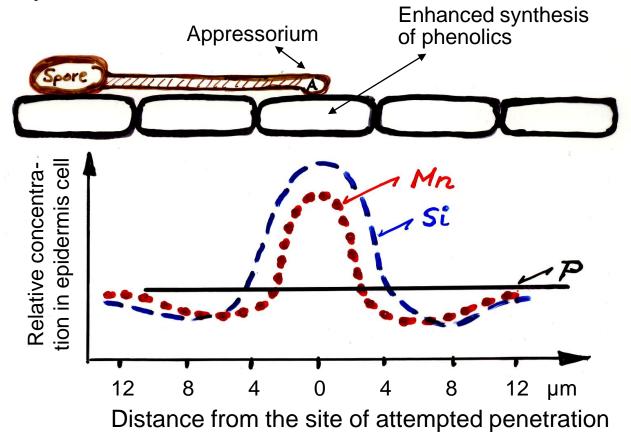
By affecting cell wall composition and lignin synthesis Mn suppresses penetration of pathogens into plant tissue.

# Penetration of a fungal hypha in to the cell wall/apoplasm and role of micronutrients

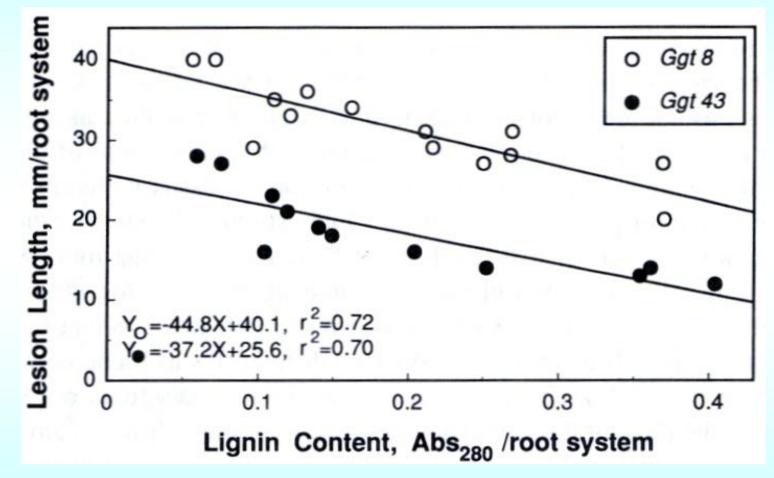


Of specific interest is the <u>short term, very localized</u> <u>redistribution of Mn and Si</u> in leaves in relation with the attempted penetration of a pathogen hyphae in a leaf cell and the subsequent rapid enhanced biosynthesis of phenolics for increased disease resistance. (Leusch and Buchenauer, 1988)

This interaction of Si, Mn and phenolics, however, is not well studied, yet.

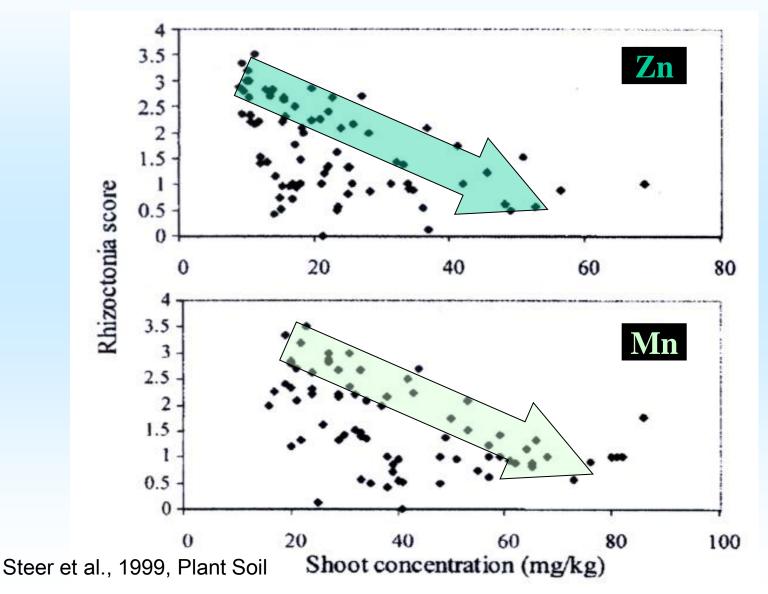


#### Relationship between root lignin content and total length of root-stellar lesions caused by *Ggt* isolates 8 (high virulent) and 43 (weakly virulent)



Rengel et al., 1994

#### Relationship between shoot Zn and Mn with *Rhizoctonia* infection rate

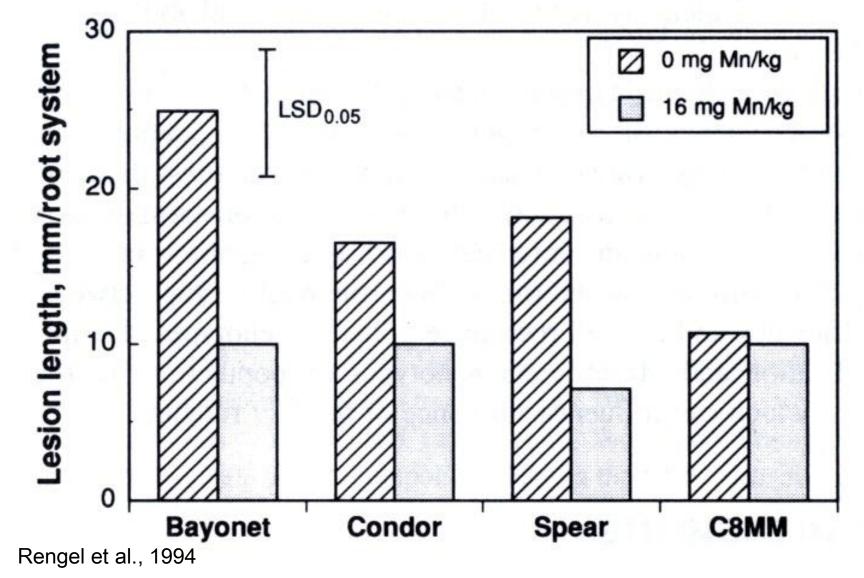


#### **Root lesions and lignin content in root tissue of four wheat genotypes at different Mn treatments**

Variable	Total length of <i>Ggt</i> lesions (mm)	Lignin content <sup>a</sup> (Abs <sub>280</sub> /root system)
Mn, mg/kg s	soil	
0	38	0.14
3	28	0.12
30	23	0.25
300	22	0.28
Genotype <sup>b</sup>		
Bayonet	30	0.14
Millewa	27	0.16
Aroona	26	0.22
C8MM	23	0.27

Rengel et al., 1994

#### Resistance of wheat genotypes to take-all in a Mn-deficient soil with and without Mn supply



Suppression of take-all can get achieved also by a rotation with oat as a precrop, resulting in a better Mn status of wheat



Take-all of wheat after wheat (front) versus wheat after oat (back)

Effect of oat as precrop in the wheat-oat rotation (black oat more effective than white oat, Integrata Coop, Brazil)

Don Huber 2002

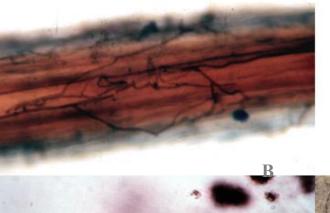
Mn-deficiency enhanced takeall in wheat monoculture

Crop Sequence	Tissue Mn (mg/kg DN	Disease Index* /I)	Yield (kg/ha)
Wheat-wheat-wheat	t 20	4.2	1450
Wheat-oats-wheat	55	1.8	3900
Oats-oats-wheat	<u>76</u>	1.0	4160

\* Rating of disease symptoms

Enhanced Mn concentrations in wheat after oat as cause for or consequence of suppressed take-all fungus

### Gaeumannomyces graminis



A. Runner hyhae on wheat rootB. Oxidation of Mn in mediaC. Oxidation of Mn in soil



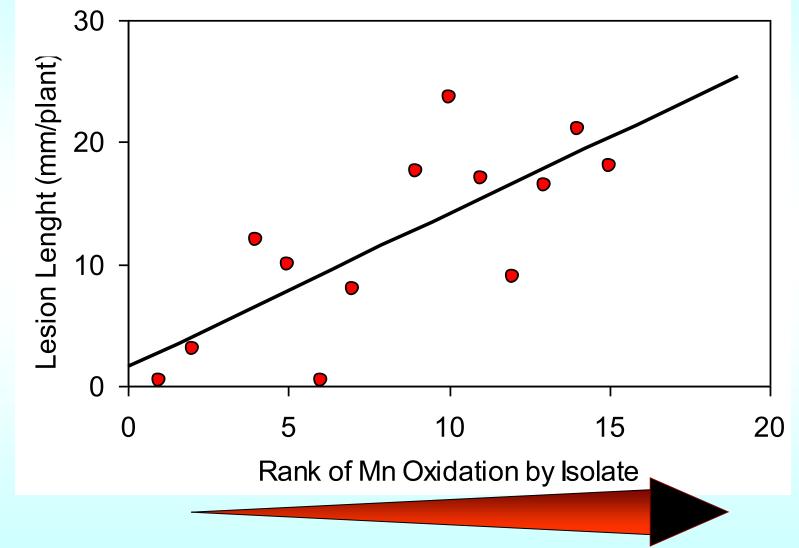
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Suppression of take-all <u>as cause</u> of an enhanced Mn mobilization due to specific Mn reducing bacteria promoted by exudates of oat *or* enhanced Mn acquisition by wheat <u>as consequence</u> of suppressed take-all fungus (common rotation effect), which is a strong Mn oxidizer on the root surface of wheat

(Gerretsen, 1937; Timoni, 1946)

(D. Huber, 2002 pers. Comm.)

### **Relationship between take-all lesions in wheat root and Mn-oxidation capacity of** *Ggt* **isolates**



Graham and Webb, 1991

# Comparision of Mn-oxidizing capacity with virulence of selected *Ggt* isolateas

	First experiment		Second experiment		
Ggt isolate <sup>a</sup>	Mn-oxidation rank <sup>b</sup>	Lesion length <sup>c</sup> (mm/plant)	Mn-oxidation rank <sup>b</sup>	Lesion length <sup>c</sup> (mm/plant)	
Controld	0	0	0	0	
82	2	4	1	0	
233	7	8	14	27	
43	11	17	5	2	
8 (Subcultured	d) 13	17	11	4	
500	14	21	20	39	

Rengel et al., 1994

#### The role of Cu in disease resistance

+ Cu Lignin (DAO) Cell wall strength



*Cu containing enzymes*: Ascorbate oxidase (AO) Diamine oxidase (DAO) Polyphenol oxidase (PPO)

Influence of Cu supply on cell wall composition of young wheat leaves (Robson et al., New Phytol. 89, 361-373; 1981)

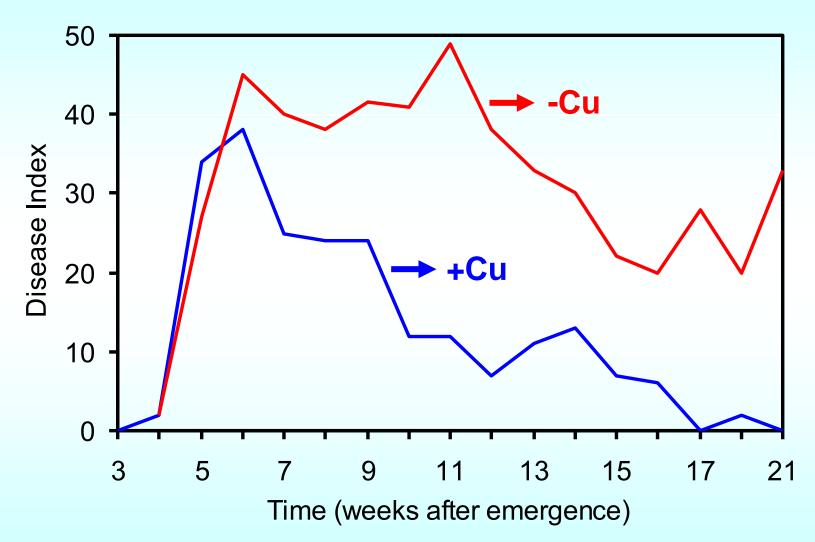
Treatment	Cu concentration (mg/kg DW)	cell wall content (%)	cellulose	all composition hemicellulose of cell wall)	
– Cu	1.0	42.9	55.3	41.4	3.3
+ Cu	7.1	46.2	46.8	46.7	6.5

## Effect of increasing Cu supply on root infection and lesions caused by *G. graminis*

Cu supply	Fresh weight yield	Roots infected	Lesions
µg/pot	g/plant	%	No./g root FW
0	3.1	66	6.5
50	6.6	62	4.0
400	10.0	49	2.4
800	9.7	58	3.2
1600	9.9	51	2.2

Graham and Webb, 1991

## Effect of Cu application on powdery mildew disease in wheat



Graham and Webb, 1991

### Conclusions

Mineral nutritional status of plants with micronutrients is crucial component in whole resistance mechanism of plants against pathogens and should be considered in all type of management systems.

Micronutrients contribute also to efficiency of biological control agents as demonstrated for Zn and Mn. A special attention should also be paid to increasing micronutrient concentration in seeds.

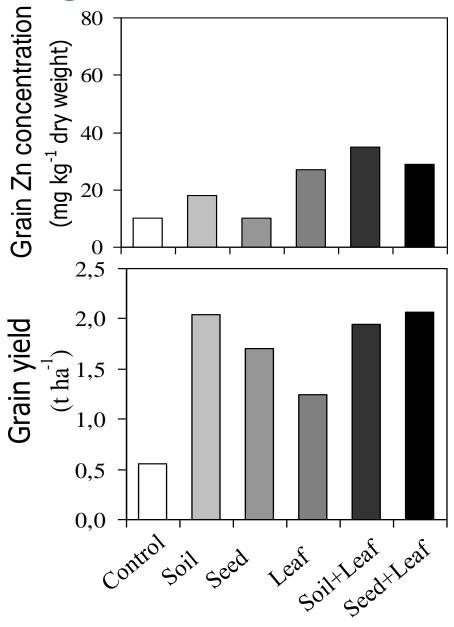
It has been shown that a high level of micronutrients (especially Zn, Mn, B) in seeds at early growth stage significantly prevents pathogenic infection, improves seedling vigor and ensures a good field establishment.

## Effect of Micronutrients on Seed Quality





## Effect of Different Zn-applications on grain yield and grain Zn concentration



Source: Yilmaz et al., 1997; J. Plant Nutr.21:2257-2264

### Influence of Seed Zn Content on Growth of Bread Wheat in a Zinc-Deficient Soil in Central Anatolia

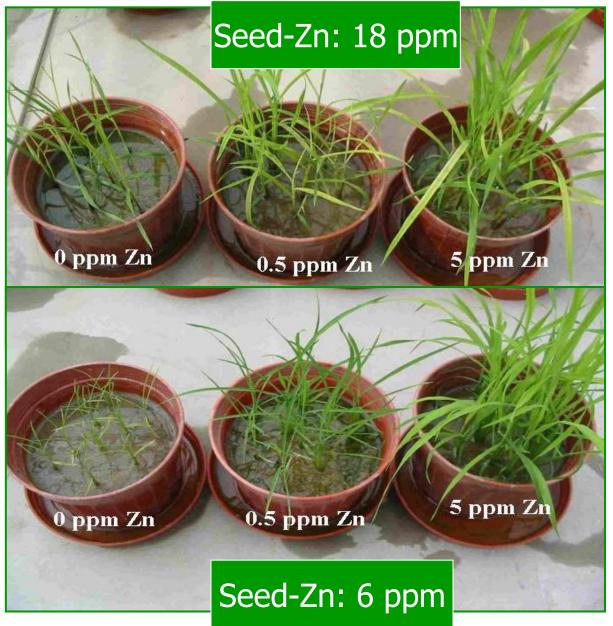
### **0.36** μg **Zn seed**<sup>-</sup>

**0.80** μg **Zn seed**-1

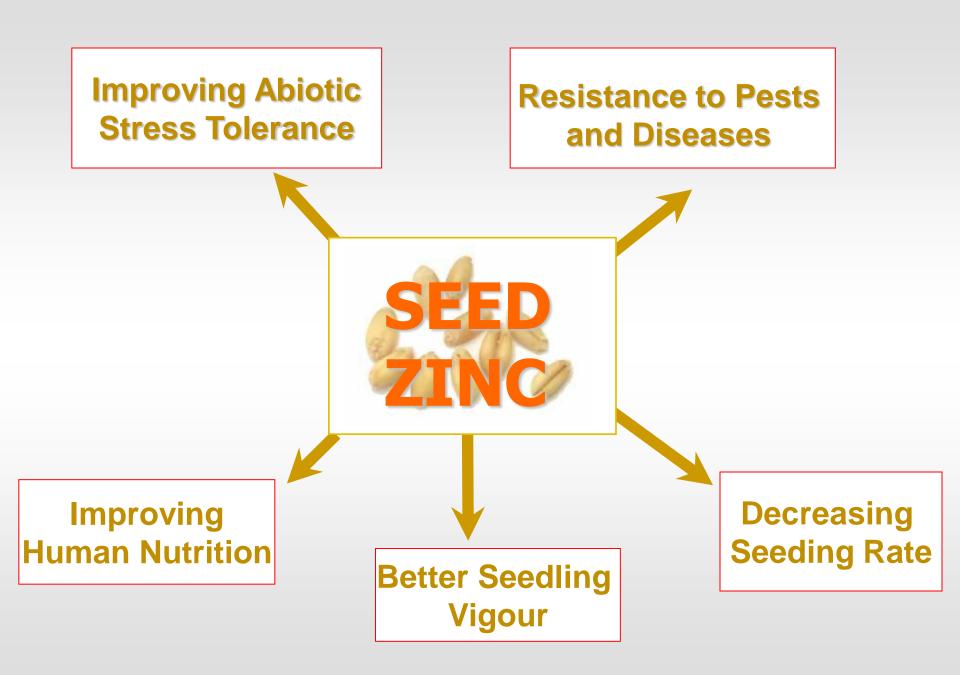


Source: Ekiz et al., 1998, J. Plant Nutr.

### **Rice Cultivars Growing in a Zn-Deficient Soil**

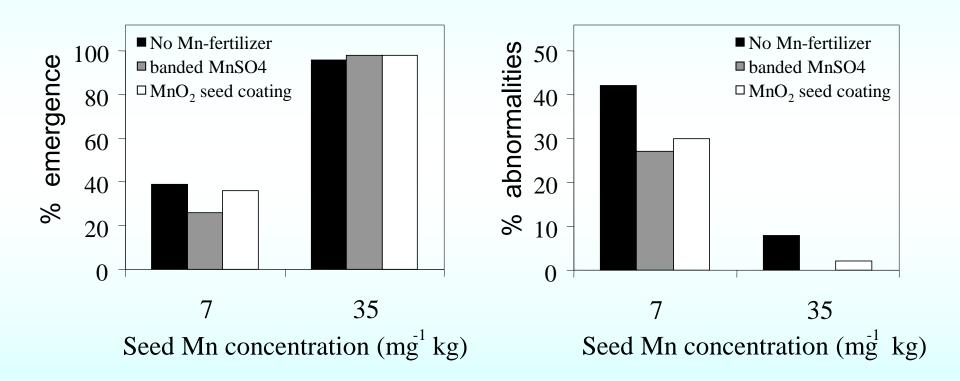


(Unpublished results)

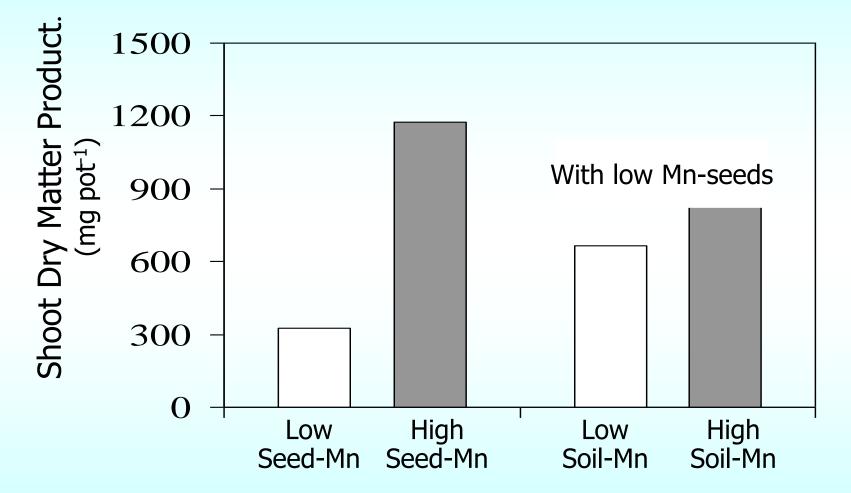


### Effect of Seed-Mn and Mn-fertilization on emergence and seedling vigor

(Source: Longnecker et al, Crop Sci. 1996; 36: 355-361)

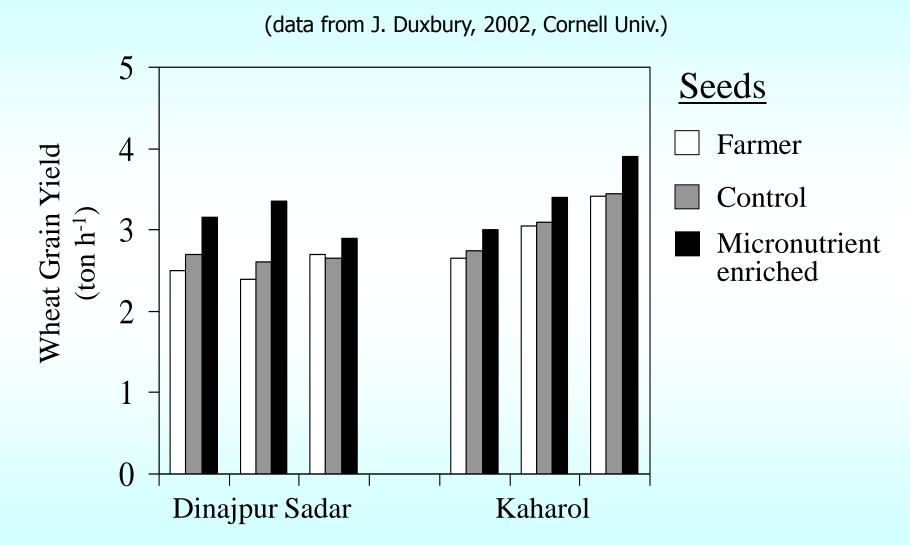


### Effect of Seed-Mn and Soil-Mn on Shoot Dry Matter Production of Wheat



Source: Moussavi-Nik et al., 1997; In: Plant Nutrition-For Sustainable Food Production and Environment, Kluwer Academic Publishers, pp. 267-268.

### **Effect of Micronutrient Enriched Wheat Seeds on Yield in different Farmer Fields at 2 Locations in Bengladesh**



CHALLENGE: Improving seeds with mineral nutrients is highly important at early growth stage for crop plants to prevent pathogenic infection, improve seedling vigor and ensure a good field establishment.

Pay attention to SEED NUTRITION to have a good NUTRIENT PROFILE in seeds