

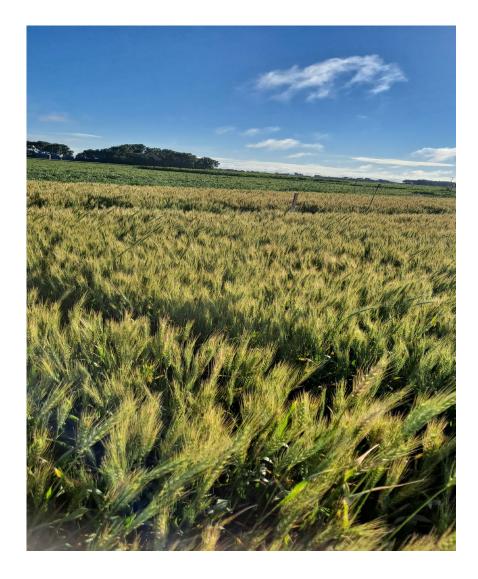
### 2023 NUTS & BOLTS SESSION

### Welcome!

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### Fusarium graminearum Isolates

#### **Develop a collection of the isolates**

- Collect infected heads
- Surface sterilizing the infected kernels
- Medias: KOMADA, Potato Dextrose Agar and
- **Carnation Leaf Agar**
- Storage of single spore isolates on soil or silica gel





## **Inoculum Production**

#### Macroconidial Suspension Inoculum

- Select the isolates(s)
- Grow isolates on Mung Bean Agar
- Wash the plates
- Count the spore concentration using a hemocytometer





## **Inoculum Production**

- Dilute inoculum to get desired spore concentration
- Inoculum can be stored in the freezer in 1 L plastic bottles for future use or in small vials for greenhouse inoculation



## **Inoculum Production**

### **Colonized Corn Inoculum**

- Grow the isolates on Mung Bean Agar
- Mix 1 kg of corn with 1 L of water in a pan
- Autoclave the pan one hour, 2 times
- Inoculate the sterile grain media with 4-5 plates
- of *Fusarium graminearum* cultures using sterile technique
- Check the pans after 2-3 weeks of incubation
- Use immediately or store the pans in a cold room until used for inoculation or you can dry the colonized grain down





### **Inoculation Methods**

#### **1- Spray Inoculation**

- Head emergence/anthesis
- apply 1 -2 applications
- Use 50,000 to 200,000 spore/ml



## **Inoculation Methods**

#### **2- Applying infected corn kernels**

- Apply infected corn kernels from tillering through boot
- Inoculation Rate varies by location, dependent on environmental variables.
- You will likely need to adjust the inoculation rate according to your experience over time .
- Dry inoculum will take longer to produce mature spores compared to fresh inoculum.



## Mist Irrigation System

- Misting design
  Pumps, pipes, nozzles
- **Misting schedule** Frequency, duration



#### Stay vigilant!

# FHB Rating

Types of resistance

Type I: Resistance to initial infection Type II: Resistance to disease spread within a spike



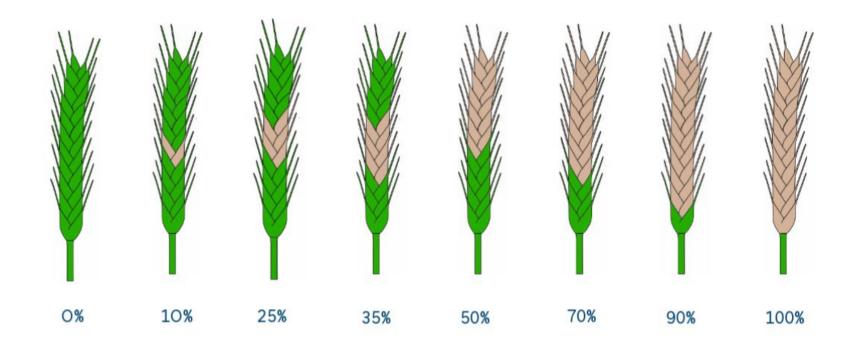




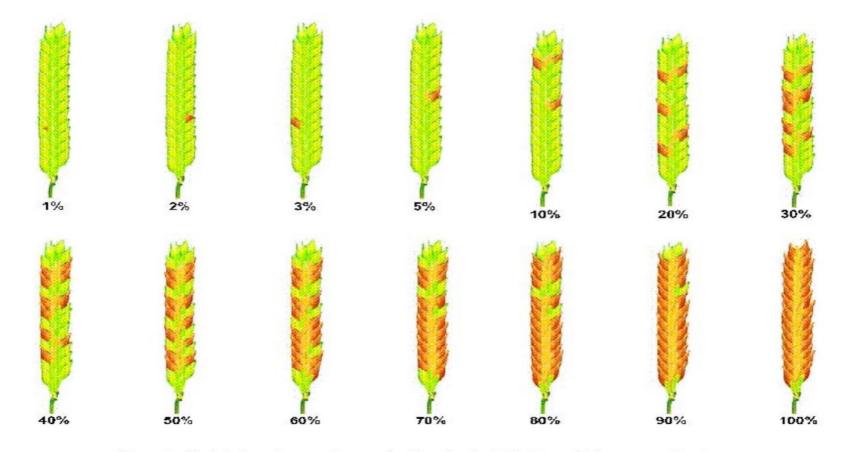
### Incidence/ Severity/ FHB Index

- Incidence proportion of diseased spikes (number of spikes with nonzero severity divided by the total number of spikes sampled).
- Severity average proportion of diseased spikelets per spike on *diseased spikes*.
- FHB Index (Scale of 0 to 100 or 0 to 9) average proportion of diseased spikelets per spike (sum of the proportion of diseased spikelets per spike divided by the total number of spikes sampled, *including those with zero severity*).

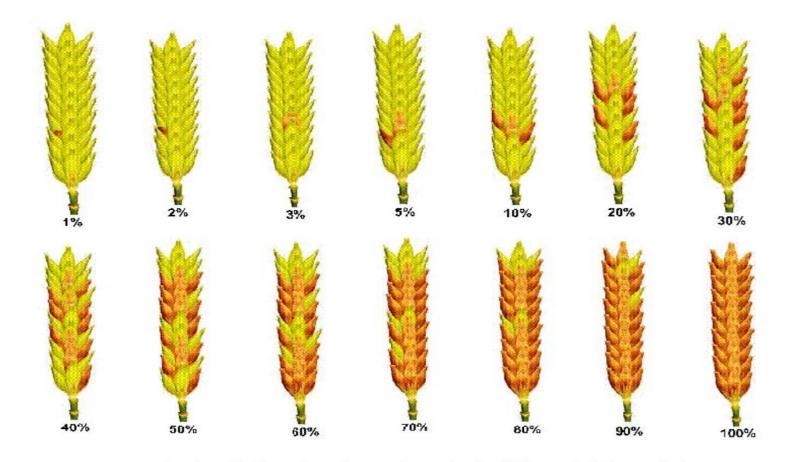
## Rating FHB- Severity (%) Wheat



### Rating FHB- Severity (%) 2-row Barley



### Rating FHB- Severity (%) 6-row Barley



### VSK Assessment

- Wait one week after harvest before scoring (allows grain to dry)
- Mix the grain before subsampling as damaged kernels tend to rise to the top
- Scoop subsample into a petri dish or weighing boat and fill till top is level
- 2 or 3 people should do the assessment and compare the results

### VSK Assessment



### Scoring FDK

### (Fusarium Damaged Kernels)

#### Kernel Damage



Tombstone, white and

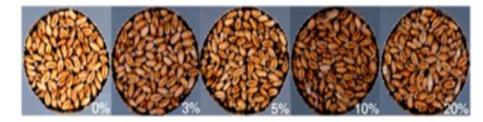
chalky (like limestone)

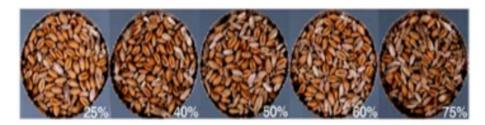
Healthy, plump and amber-colored

> Shriveled, note color difference: shiny

Raisin? Damaged, not likely due to Fusarium

Pink, covered in F. graminearum mycelium FDK Scale by Engle, De Wolf & Lipps; Ohio State







Greenhouse inoculation of *Fusarium graminearum* 

- Point Inoculation
- Spray Inoculation



Toxin levels generally higher in the greenhouse than field.





# Harvesting Samples

- Harvest 1-2 feet of row from center of plot
- Thresh harvested heads on belt thresher
- Clean the sample using a seed cleaner
- Obtain a 100g sample using a divider
- Send samples to a testing lab



### **Cleaning Samples**



**Belt Thresher** 



Kornservice seed cleaner

### **Toxin Analysis**

- Obtain ~ 100 g of representative sample using a grain divider
- Clean the sample
- Grind the samples (10 to 100 grams)
- Send the samples to a testing lab for deoxynivalenol (don) measurement

# Thank you!



### Questions?

For follow-up questions, please contact: Beheshteh Zargaran zarga001@umn.edu

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U.S. Wheat & Barley Scab Initiative