

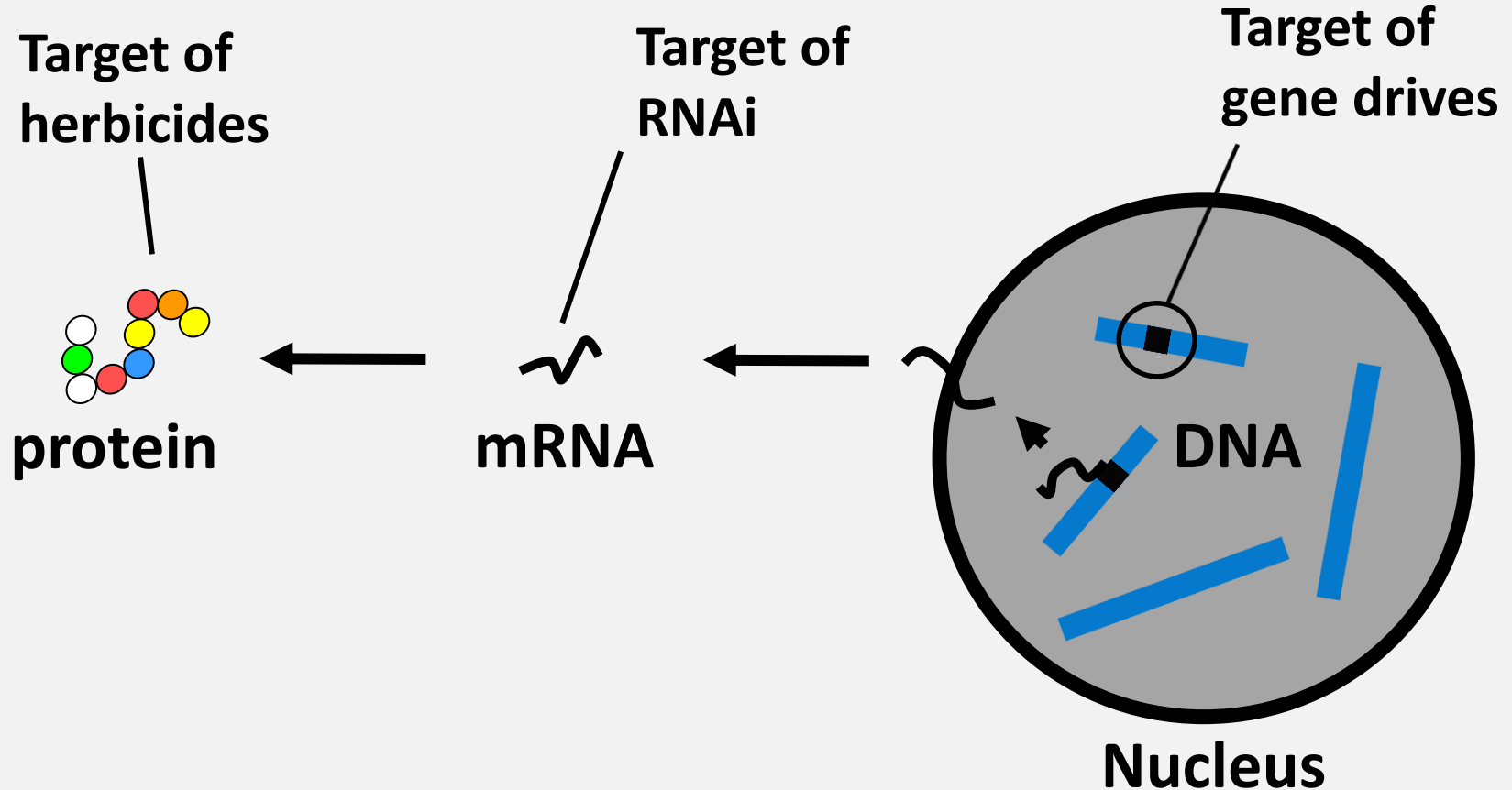
# **Weed Genetics Project Update**

**Michael Christoffers, Ph.D.**

**Department of Plant Sciences**

**North Dakota State University**

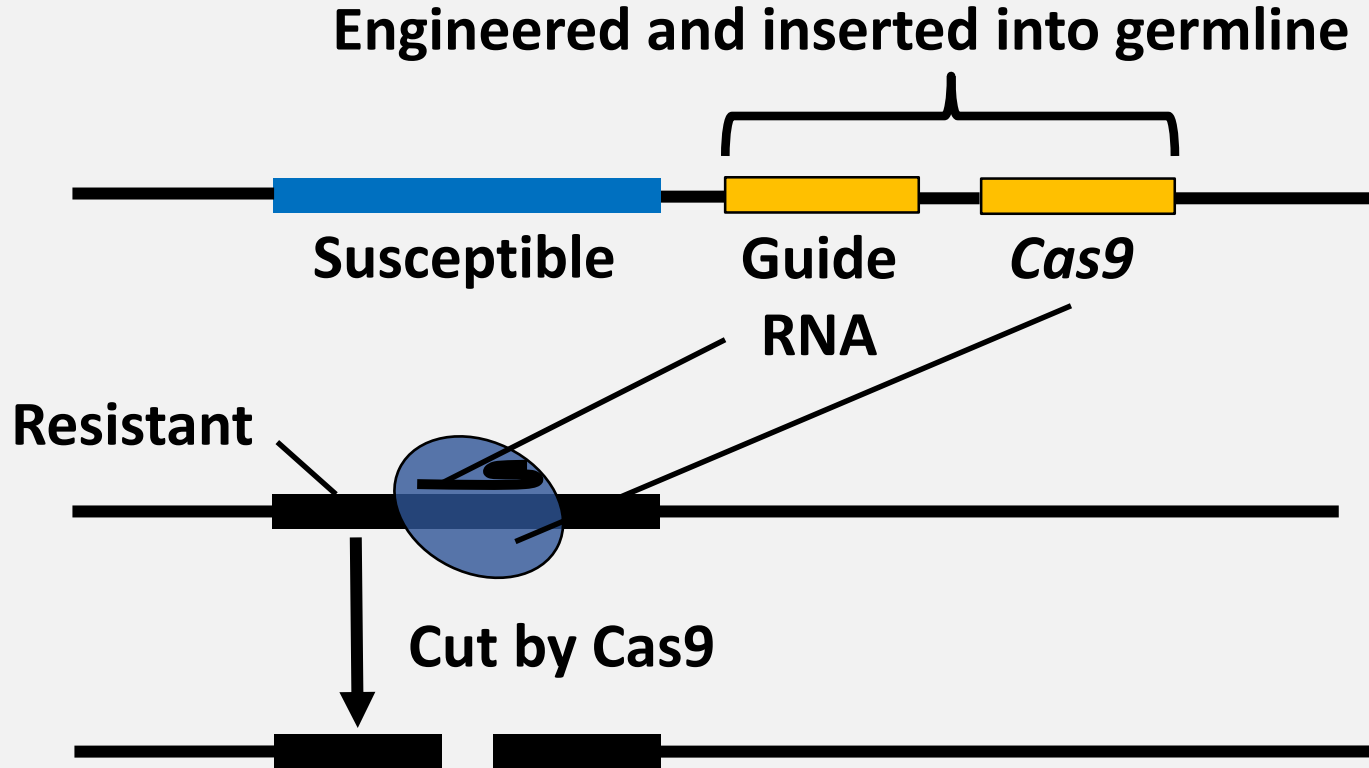
# Genetic Biocontrol



# Genetic Biocontrol: Gene Drive Update



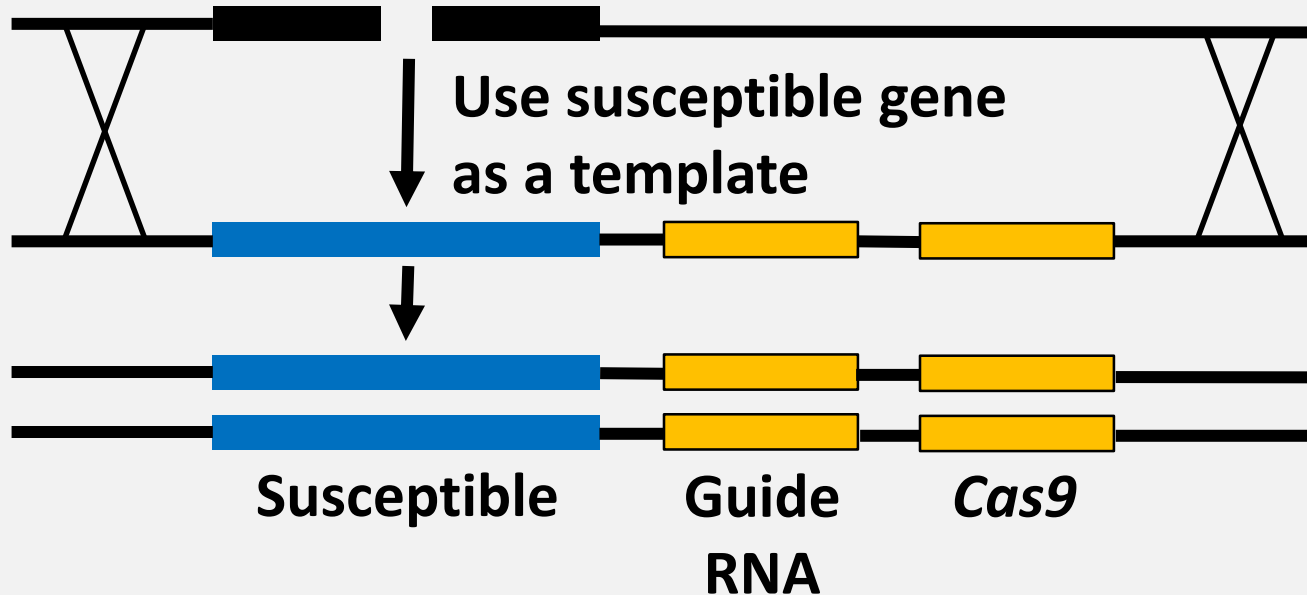
# Example: Reversion of Pesticide Resistance



# Example: Reversion of Pesticide Resistance

Natural repair of the cut, one of two possibilities:

- ① Use susceptible gene as a template



# Example: Reversion of Pesticide Resistance

Natural repair of the cut, one of two possibilities:

- ② Join the broken ends together

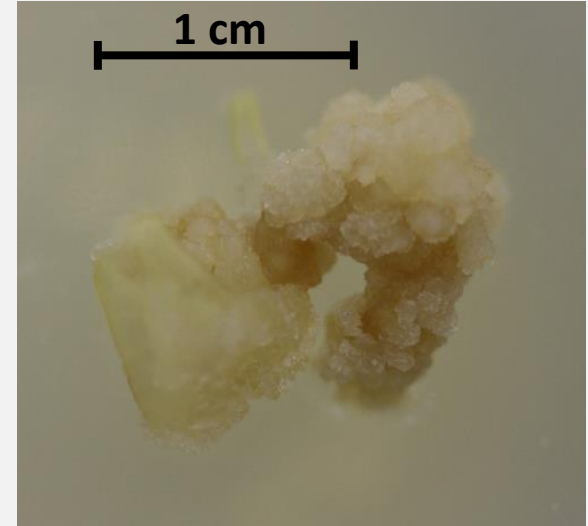


# Waterhemp Tissue Culture



**Germinated seeds**

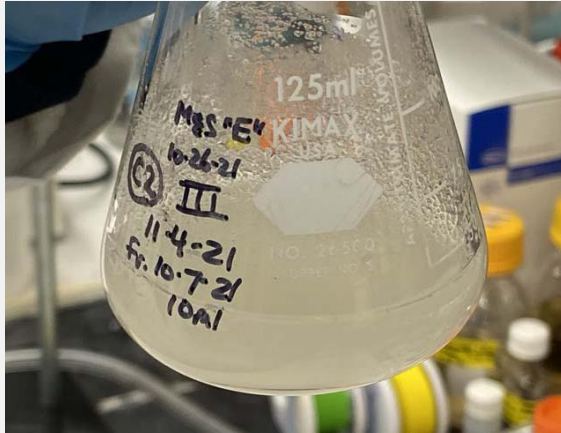
**Remove a portion  
of the stem  
(hypocotyl)**



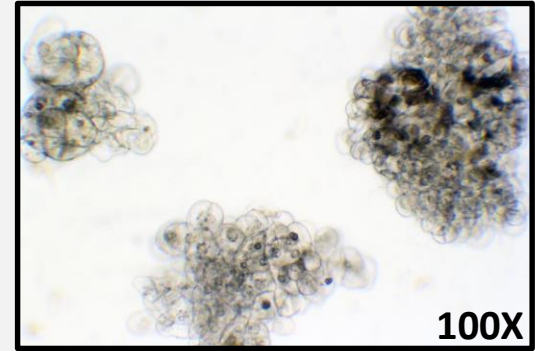
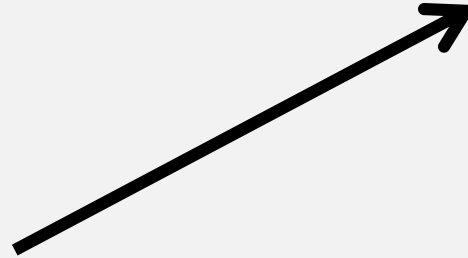
**Callus tissue**

# Waterhemp Cell Suspension Culture

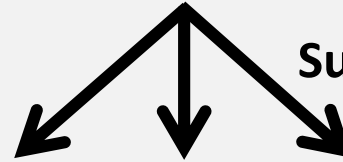
Callus  
tissue



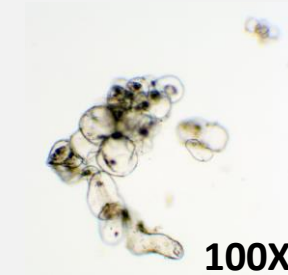
Cell suspension culture



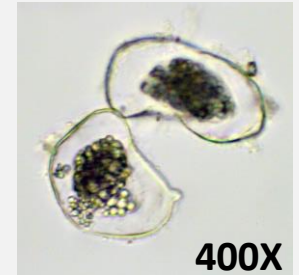
Subculture



100X

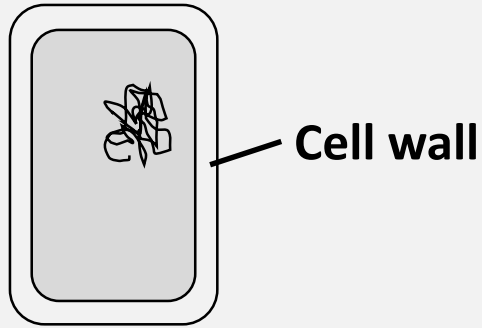


100X




400X

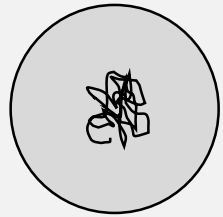
# Successful Production of Waterhemp Protoplasts



**Degrade  
(remove)  
cell walls**



A large, bold, black arrow pointing downwards, indicating the process of degrading the cell wall.

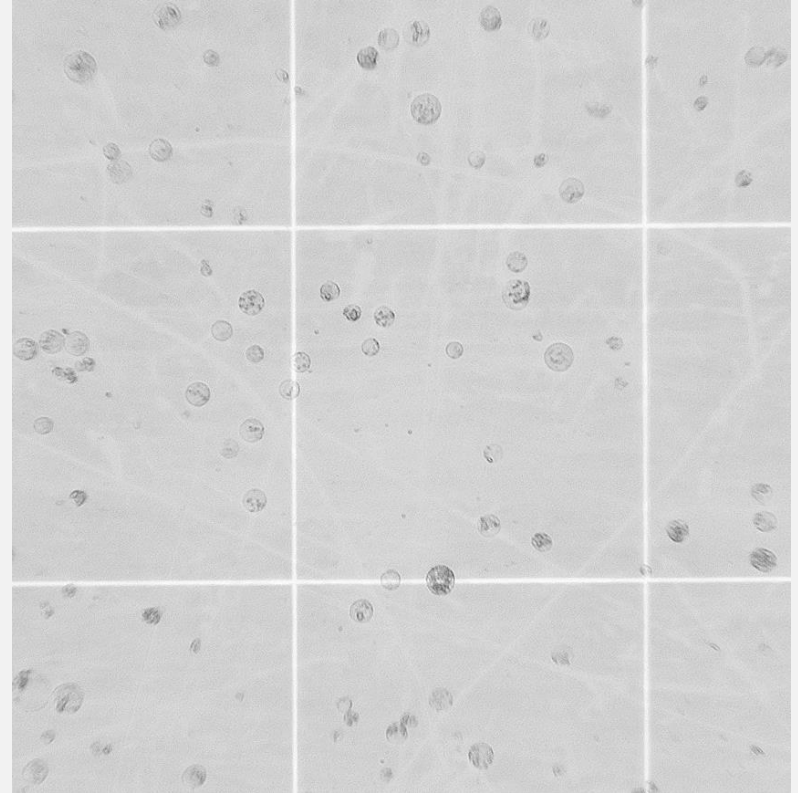


**Protoplast**

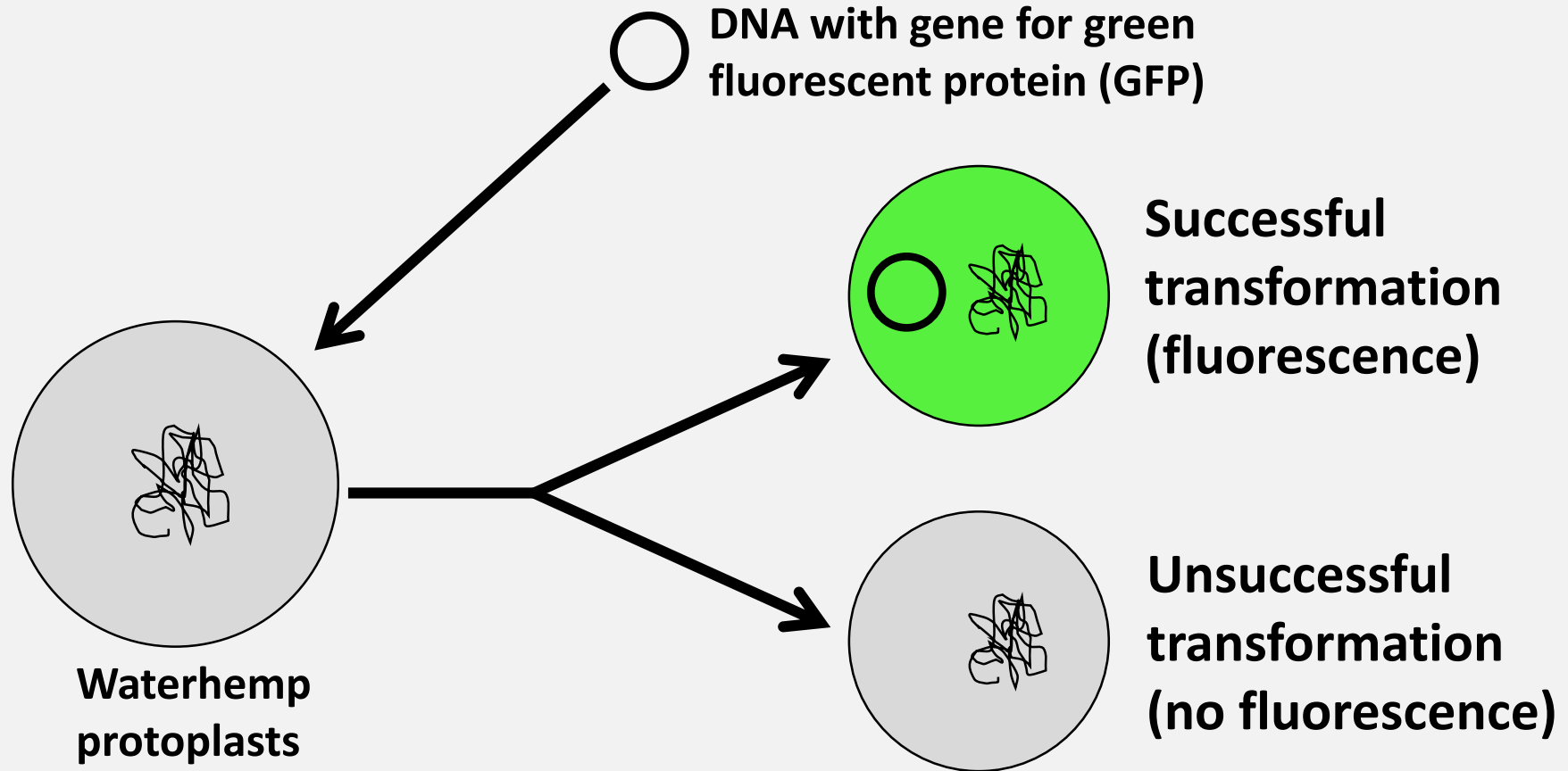
**Waterhemp  
protoplast**



**Waterhemp protoplasts**

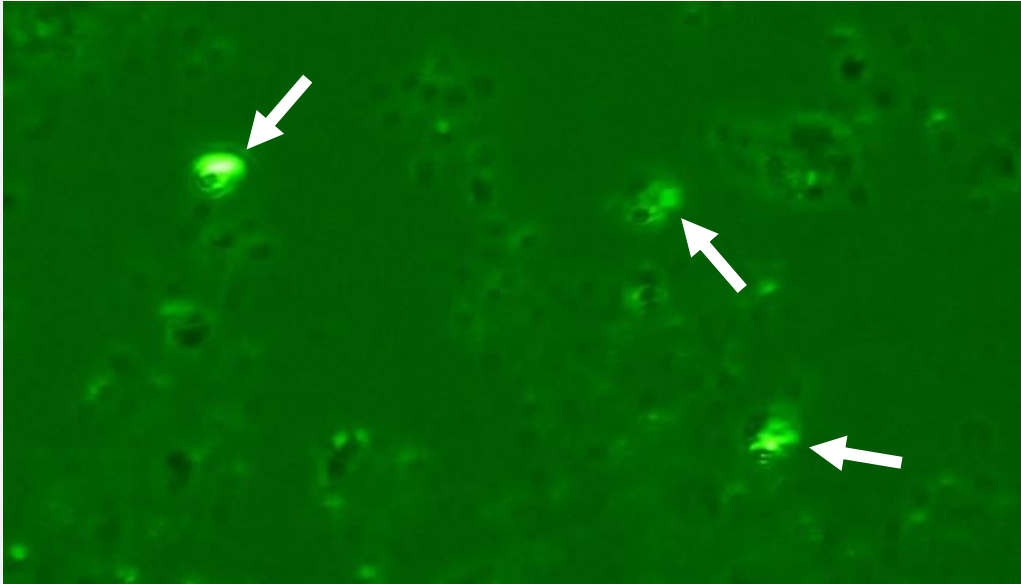


# Protoplast Transformation



# Transformation of Waterhemp Protoplasts

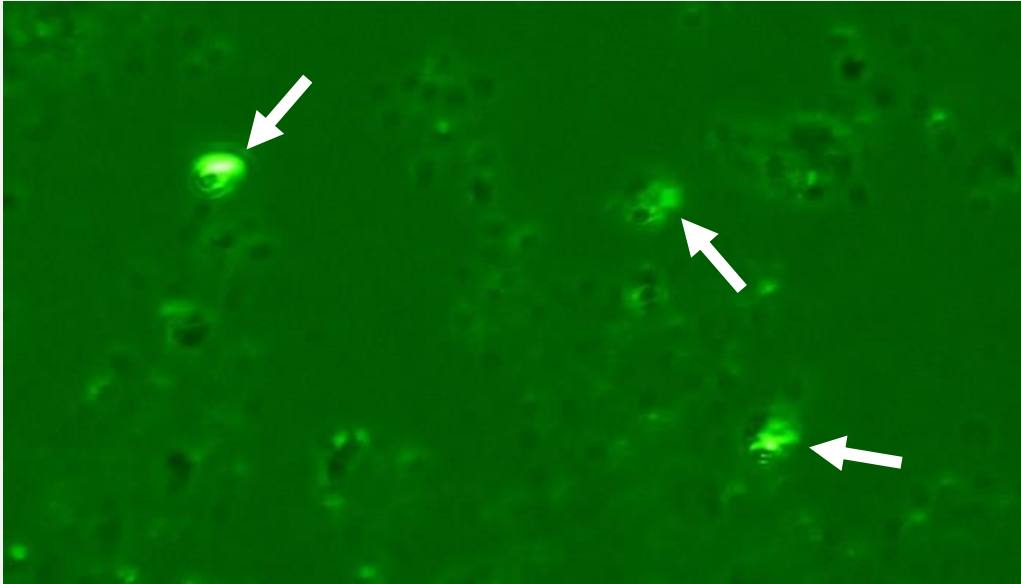
Waterhemp protoplasts after transformation with the gene for GFP



- Arrows indicate likely expression of GFP in waterhemp protoplasts
- Additional confirmation and optimization is needed

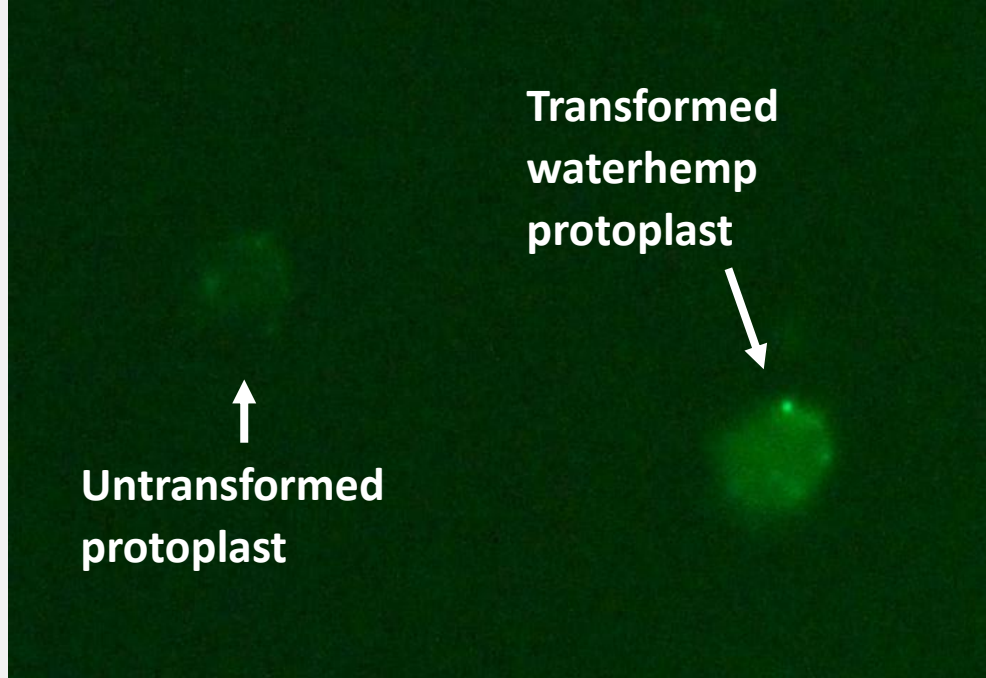
# Transformation of Waterhemp Protoplasts

Waterhemp protoplasts after transformation with the gene for GFP



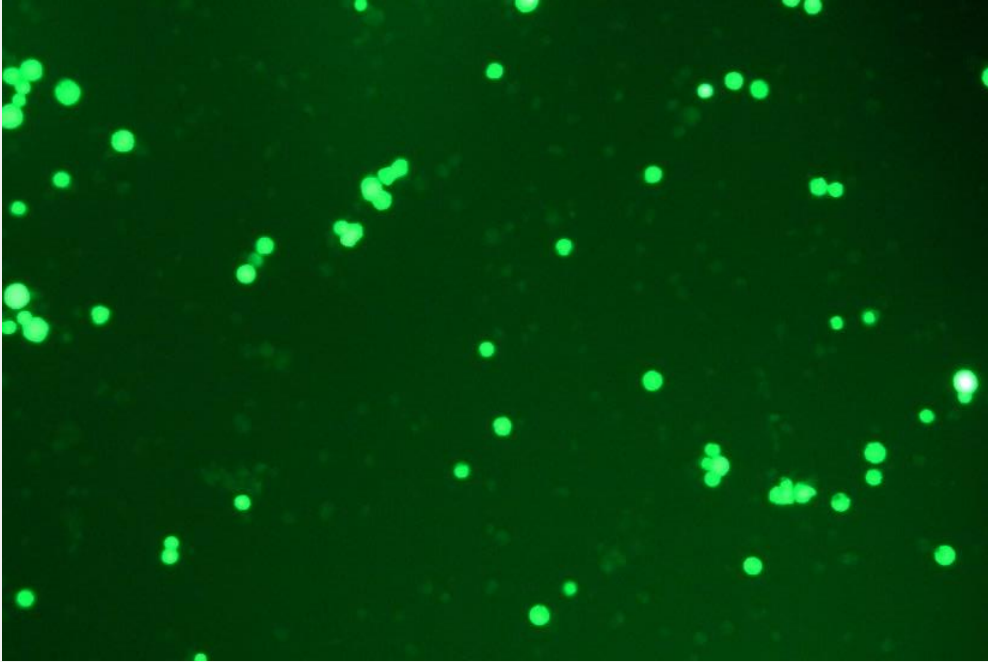
- ~~• Arrows indicate likely expression of GFP in waterhemp protoplasts~~
- Additional confirmation and optimization is needed

# Protoplast Transformation with Protein



Green fluorescence indicates successful transformation of waterhemp protoplasts with protein (Cas9/GFP fusion protein)

# Testing for Oxidative Stress



In this case, green fluorescence is bad

Indicates that  
waterhemp  
protoplasts are under  
oxidative stress

# **Acknowledgments**

- **Robert Sabba and Peter Beerbower**
- **ND Agricultural Experiment Station**
- **ND Corn Council**
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- **ND State Board of Agricultural Research and Extension – Soybean**
- **USDA – National Institute of Food and Agriculture**