

Leaf and Shoot Apical Meristem Response to Abiotic Stress in Two Tomato Species

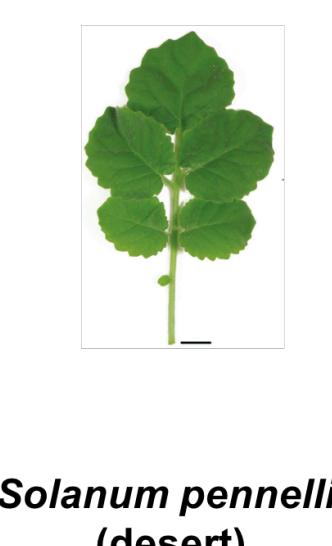
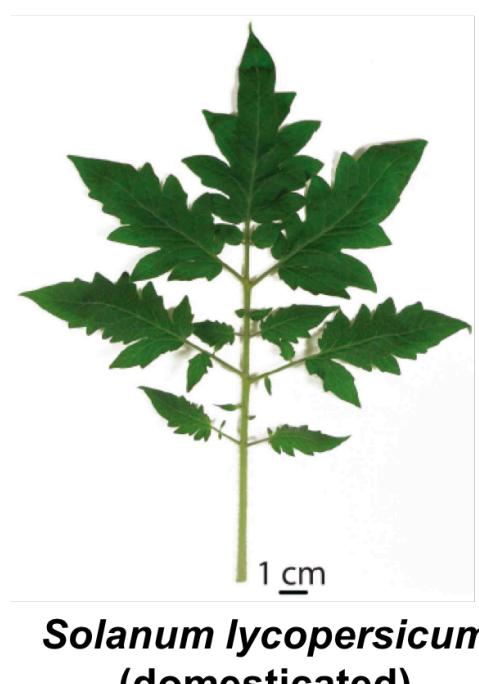
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Abstract

The recent irregularity in local climates has impacted agricultural productivity as crop yield and quality require specific ratios of rain, sunlight and warmth. To this end, fluctuations in water availability leave plants vulnerable to both desiccation and drowning. In order to investigate susceptible and tolerant responses to drought and waterlogging within the tomato family, we compared domesticated tomato (*Solanum lycopersicum*) and its wild desert relative (*Solanum pennellii*). By comparing the anatomical responses to water stress, we can identify structures that vary between the sensitive and tolerant species. Then by using molecular genetics we can identify the differences in the gene networks involved in responses to water stresses. To do this, we will utilize 35S-driven transgenic reporter lines (Isolation of Nuclei TAGged in specific Cell Types (INTACT)) to compare the transcriptome and the epigenetic regulation mechanisms in leaves and in the shoot apical meristems of plants challenged with drought and flooding. Having an understanding of the mechanisms that lead to abiotic stress tolerance will allow us to identify breeding strategies to create more robust crops.

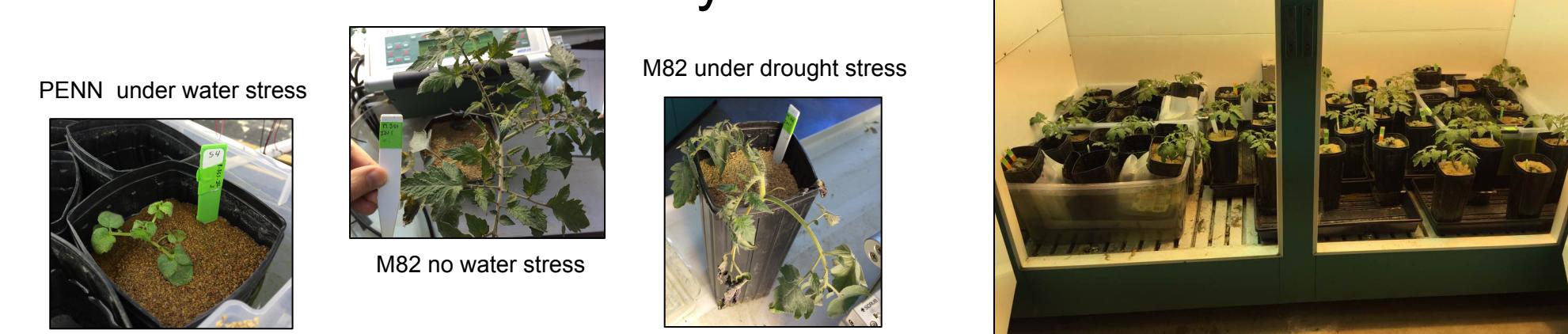
Introduction

The tomato clade originated in South America in the Andes mountain range and species thrive in diverse environments from deserts to salty, dry valleys to cold, damp mountain tops. *Solanum lycopersicum* (M82) was one of the first crop species to be domesticated and produces large, red fruits. Tomatoes have dark green compound leaves and are grown as an annual crop in temperate climates. *Solanum pennellii* ("PENN") is a wild species closely related to tomato that evolved in the desert. PENN has yellowish-green leaves that produce many noxious, defensive compounds and makes small, green, poisonous fruits when ripened. In order to investigate shoot responses to abiotic water stress, we conducted the following experiments.



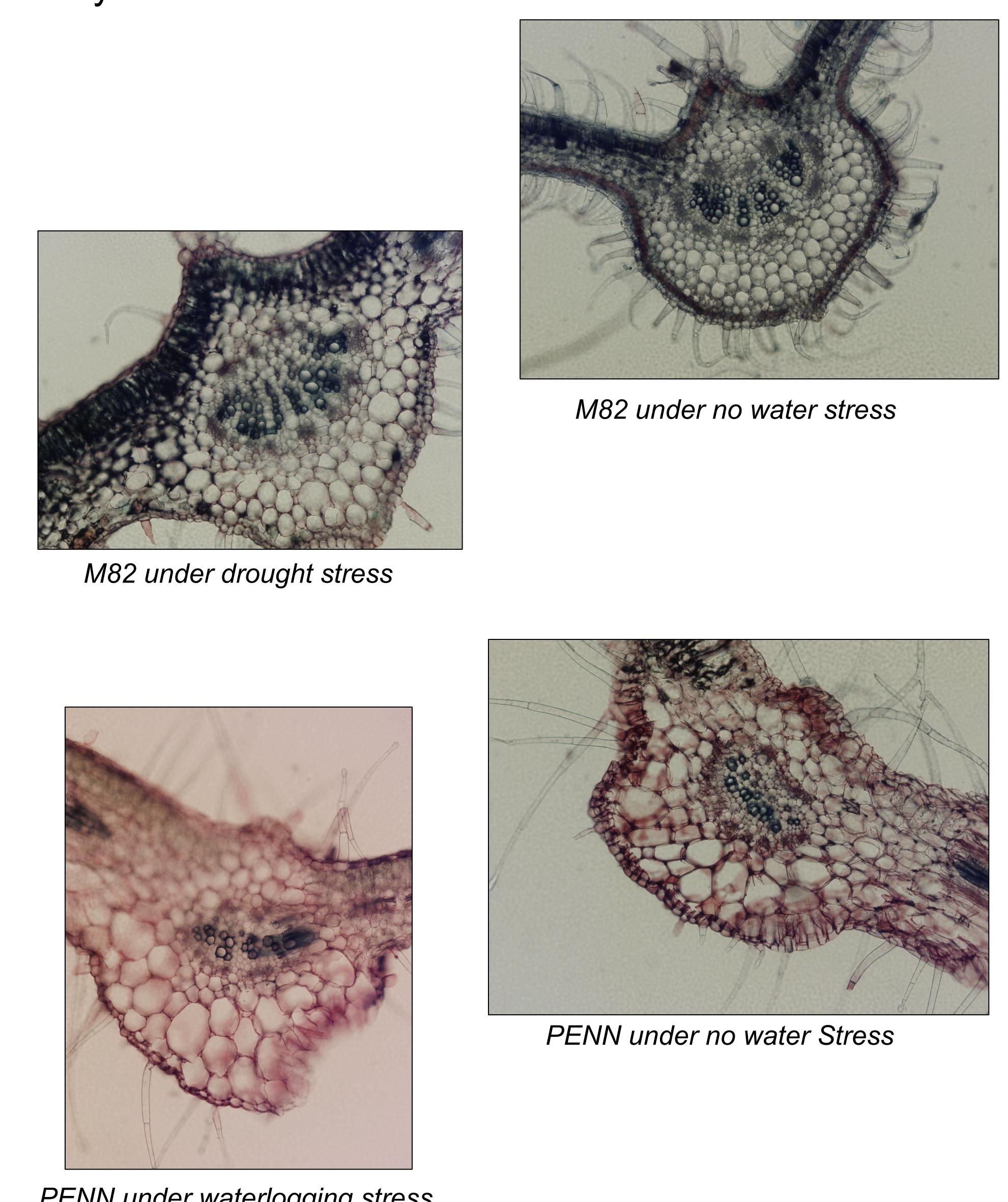
Methods

One week after germination, plants were transplanted into pots and randomized into treatment groups. Twenty days after transplanting, water stress conditions were imposed. After twelve days, the plants were phenotyped and harvested for further analysis.



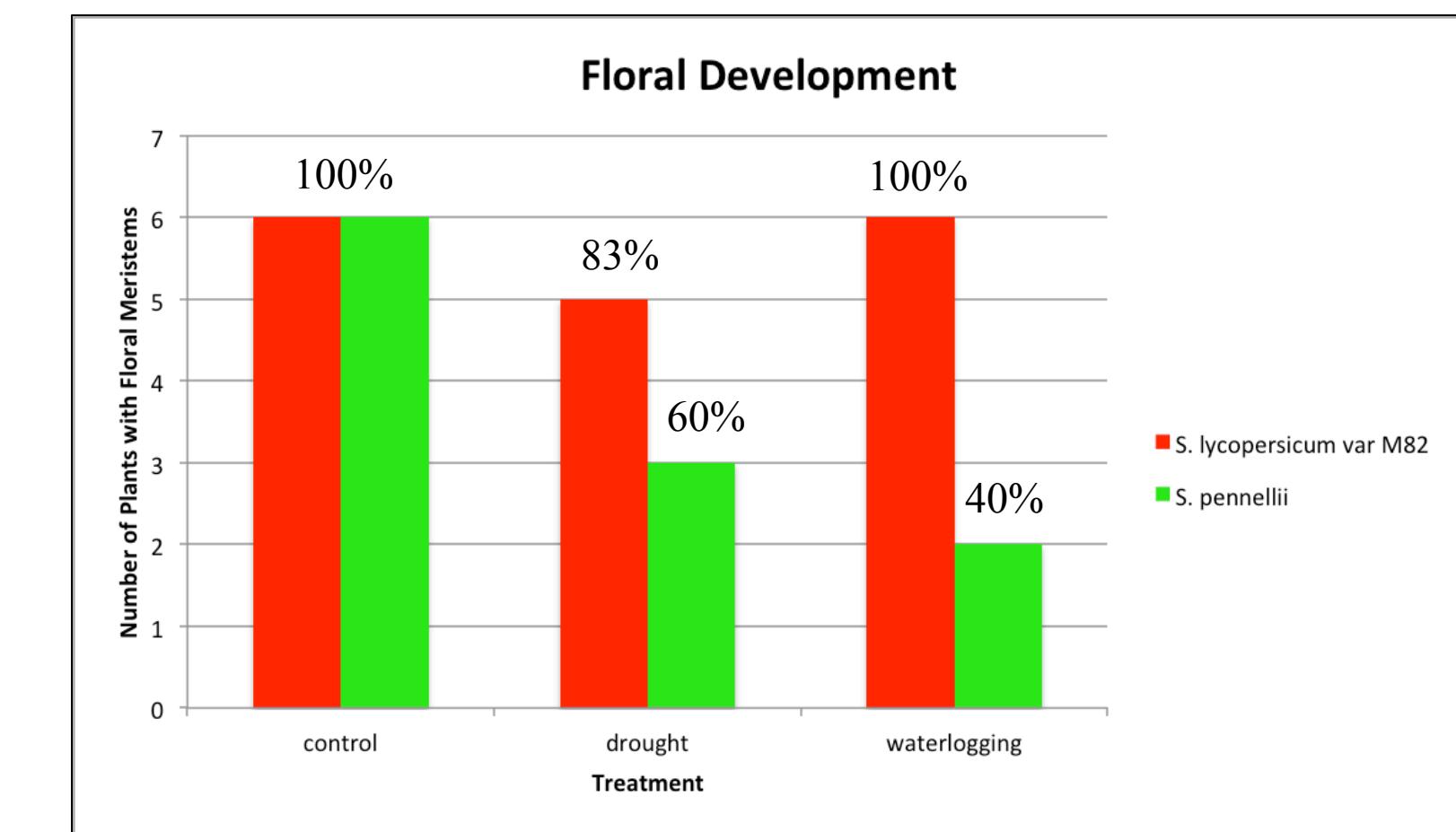
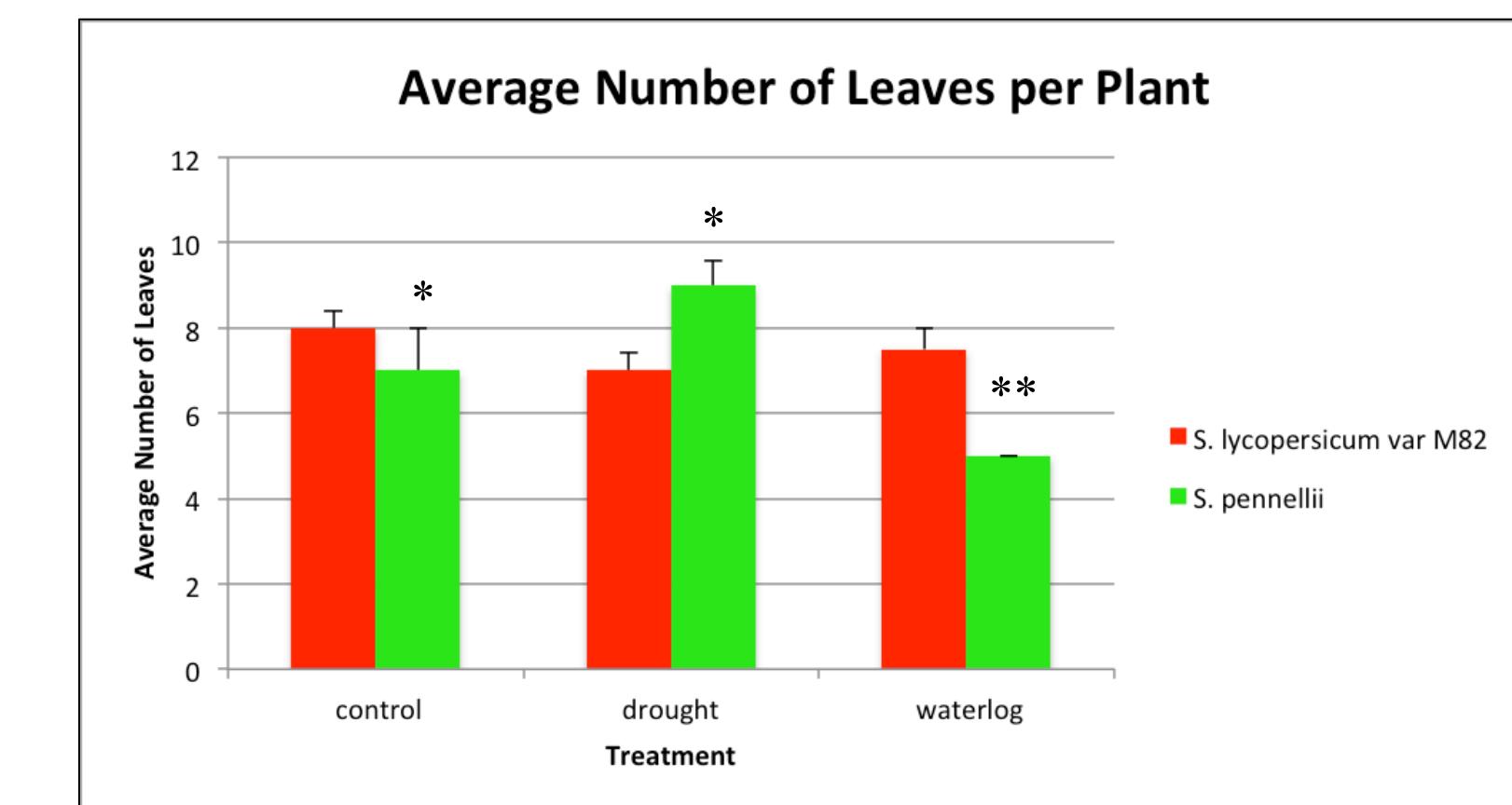
Anatomy

In order to see anatomical differences induced by stress, sections of tissue were cut from plants under all three conditions. Particularly of interest in shoot is the leaf mid vein. These pictures compare M82 under no water stress, M82 under drought stress, PENN under no water stress, and PENN under waterlogging stress. These sections were made using a vibratome and the vascular bundles were stained with 1% Toluidine Blue dye.

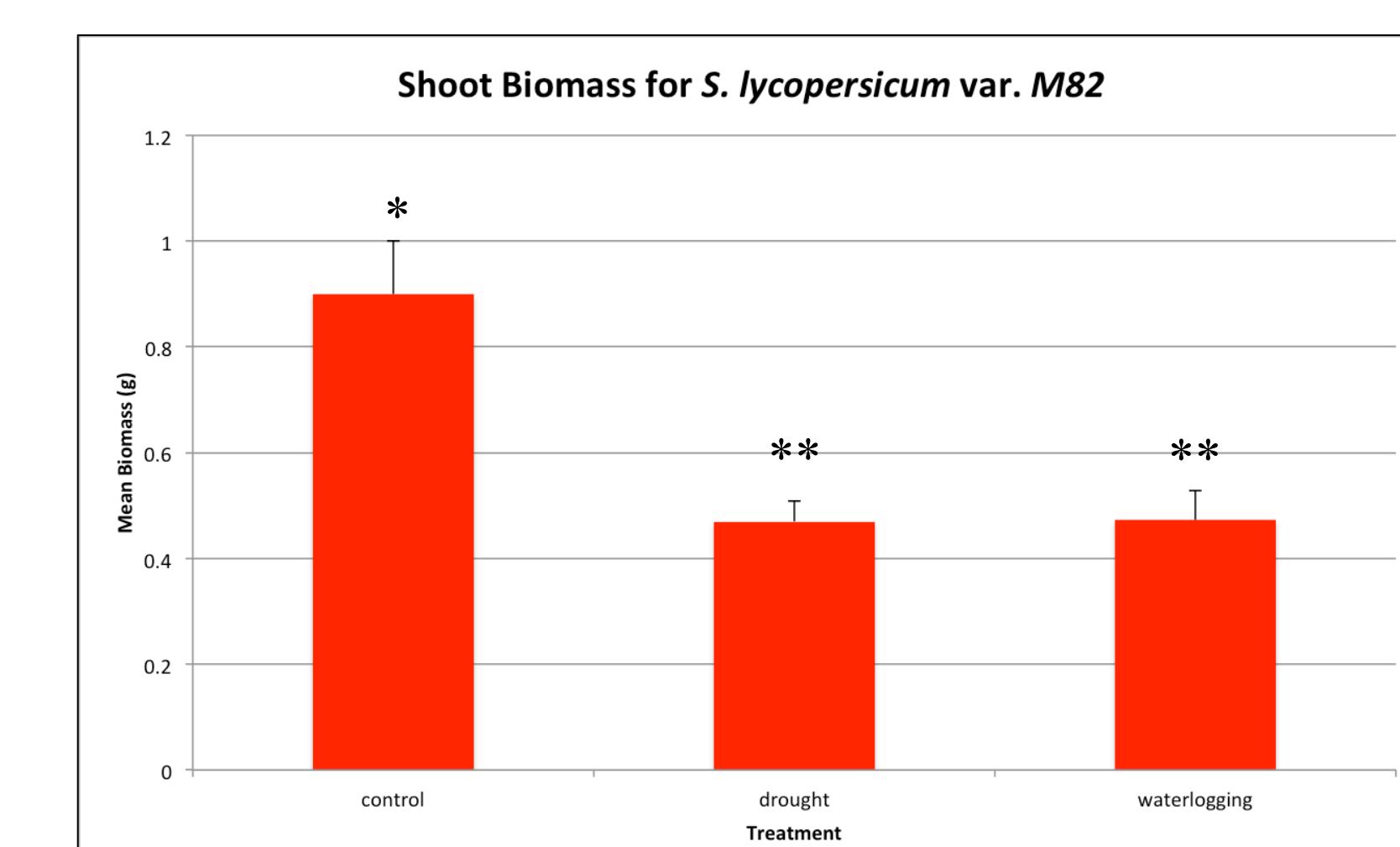


Results

After twelve days of stress, phenotypic measurements were collected.



While penn plants performed well under drought conditions, waterlogged penn plants produced significantly fewer mean leaves and fewer plants produced flower buds.



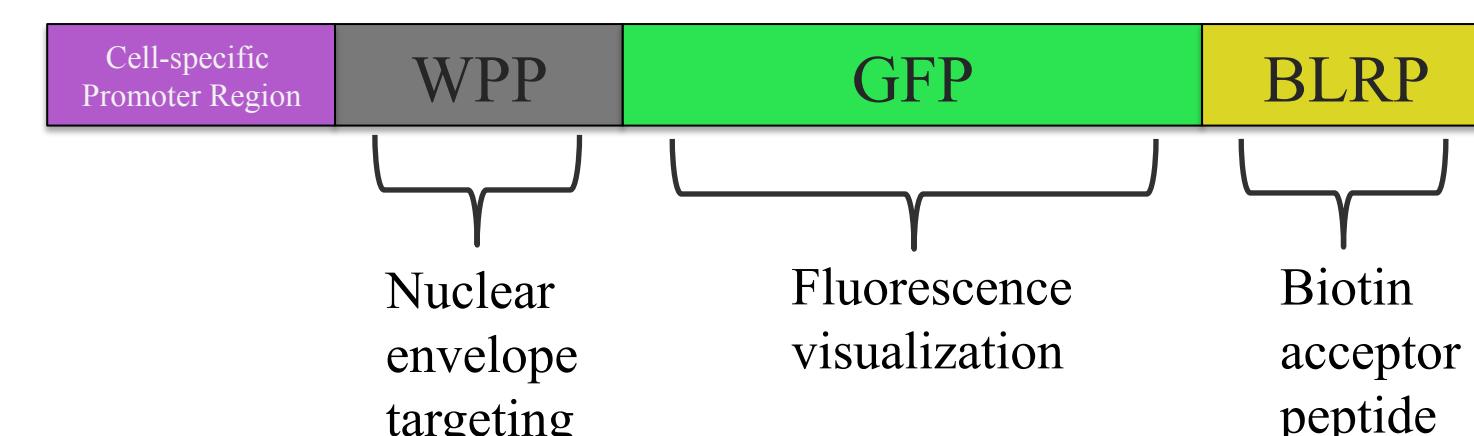
Though M82 retained much of its flowering capacity under waterlogging stress, the total shoot biomass was significantly reduced under both water stress conditions.

For each graph, * and ** denote different categories of results as determined by ANOVA comparisons.

Future Investigations

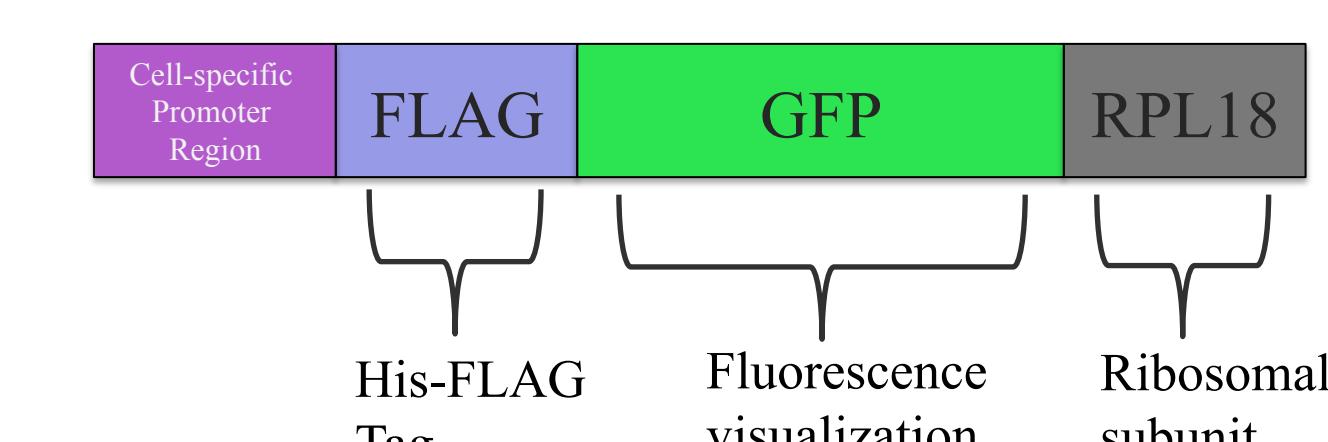
In order to more precisely understand the onset of genetic responses to water stress, the function of specific cell types during leaf development will be analyzed. To achieve this goal, two molecular methods will be utilized: Isolated Nuclei TAGged in specific Cell Types (INTACT) and Translating Ribosomal Affinity Purification (TRAP).

INTACT



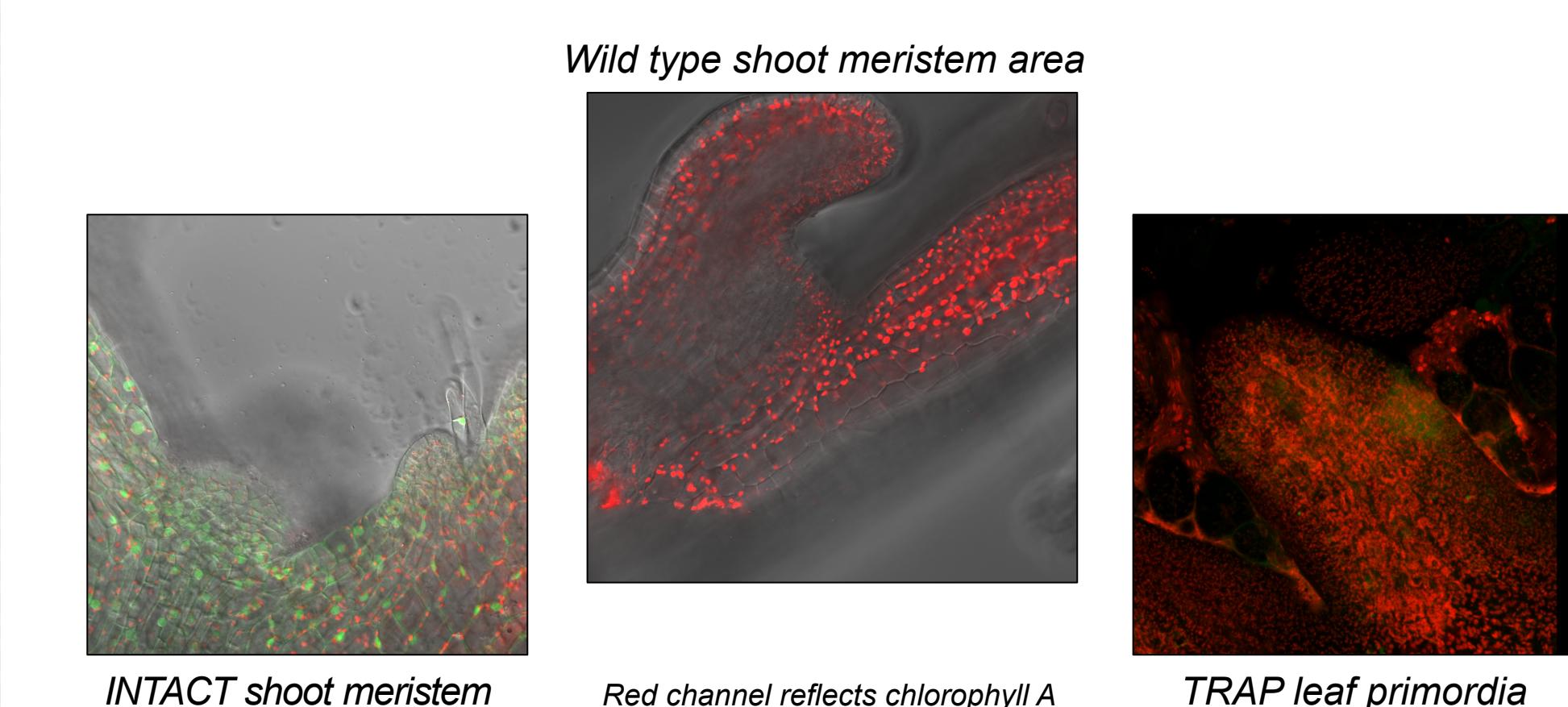
INTACT is a method of labeling the nuclear envelope allowing for isolation of the labeled nuclei to measure gene expression and chromatin features.

TRAP



The TRAP method tags ribosomes allowing pull down of ribosomes bound to messenger RNA (mRNA). This allows the analysis of the translome in genetically defined cell types.

Below are examples of stable T1 lines we are screening for proper expression patterns for both TRAP and INTACT. Future work will include water stress experiments on these transgenic lines.



References & Acknowledgements

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- Ron M, Kajala K, Pauluzzi G, et al Plant Physiology. 2014;166(2):455-469
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Plasticity



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