

Phenotype Meeting

5.3.2024

Sha Library EMS Screen Overview

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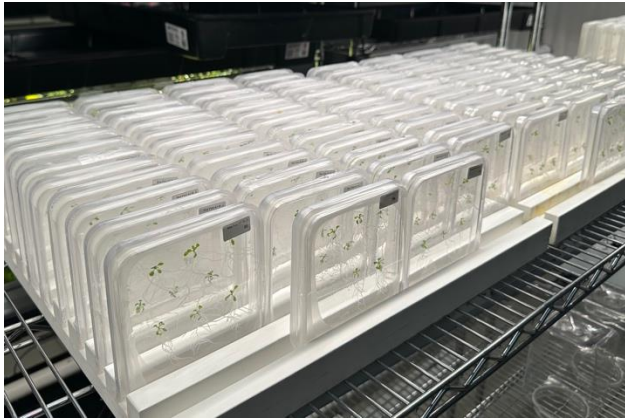
EMS Screen Pipeline

Harvested as 135 pools (September 2023)



Seed to seed growth

Weekly waves of 990 mutants in 200 plates each



Mutants identified in gel at 14 days

Plate Layout

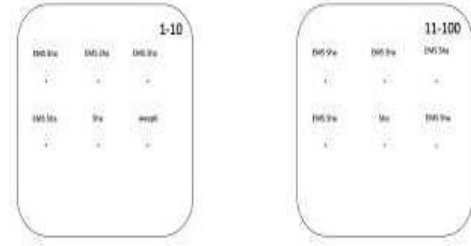
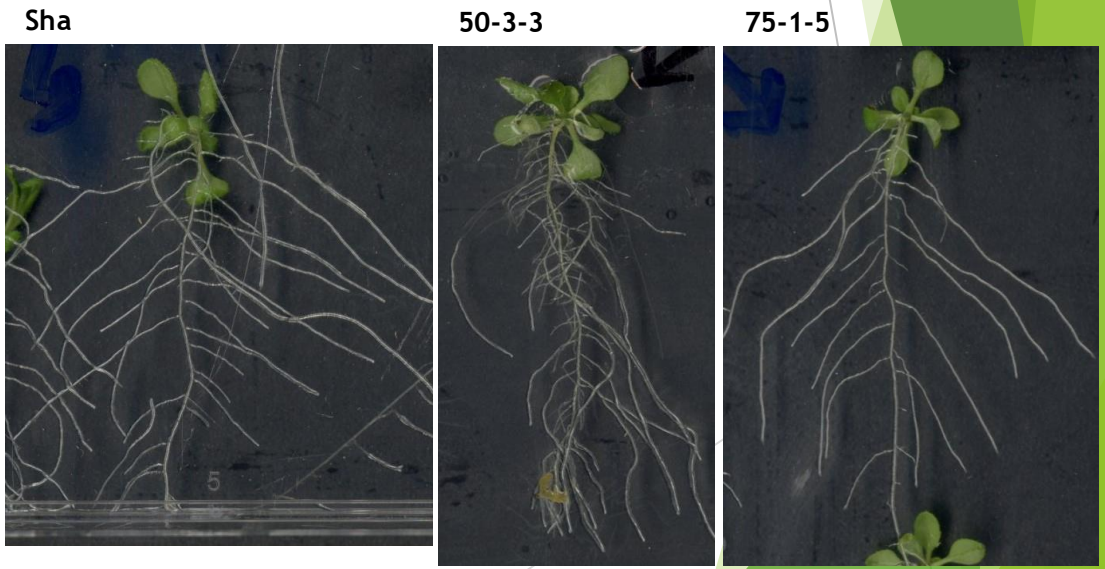


Plate labels are as follows: Plate # Pool #, Wave #



600+ interesting steep architecture phenotypes so far



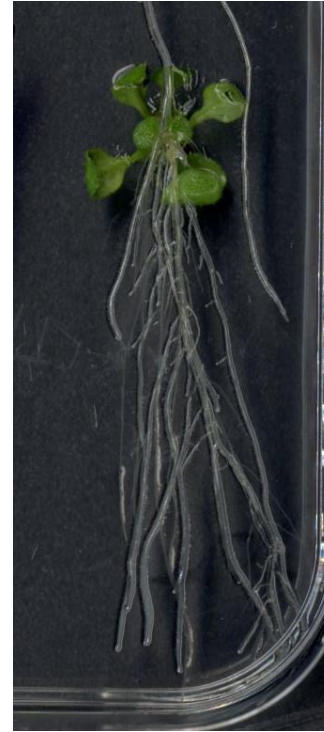
Steep Root System Architecture (RSA) mutants and Sha are grown to seed. Mutants are labeled by: Pool #- Mutant #- Wave #

Typical RSA of Sha (Control) Versus weep6 (Steep RSA Phenotype Control)

Sha



weep6

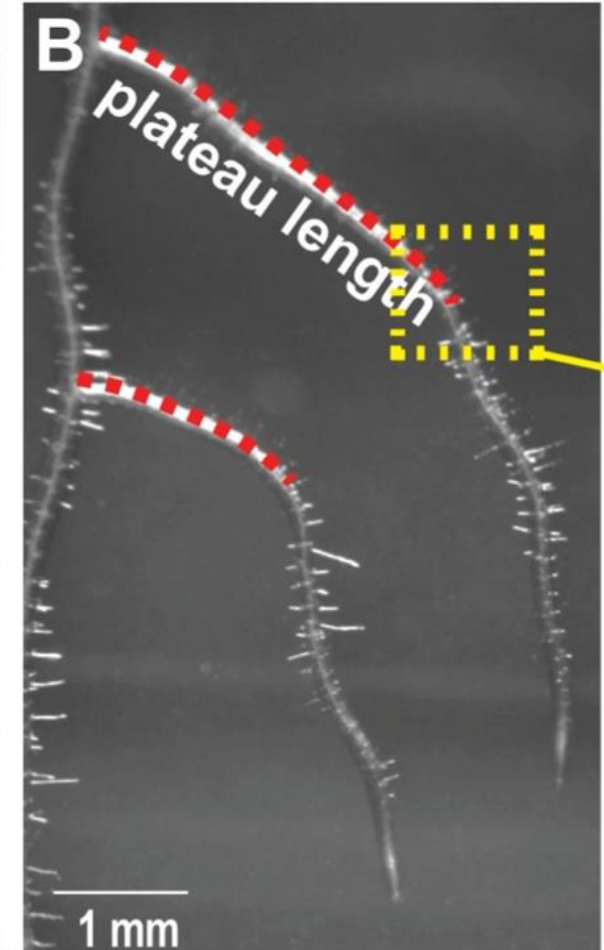
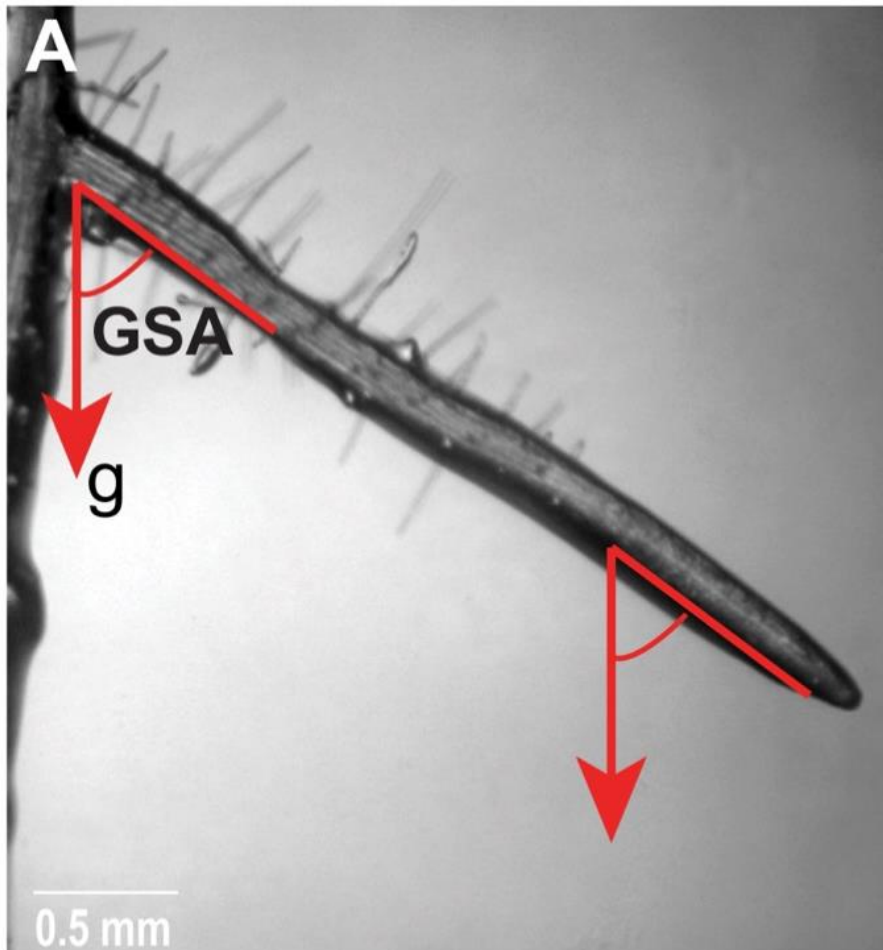


-With this screen we are trying to identify new genes that control root system architecture. For HPI purposes, we are especially interested in steep RSA because those can determine deep rooting.

-A good example of a steep root system architecture is the weep6 mutant and we use it as a control in our plates.

-We also use Sha seedlings as controls of non-mutagenized individuals

Factors That Determine RSA Steepness: LRiGSA and Plateau Phase



- The steepness of a RSA can be determined by different factors, one is small initial lateral root angles (LRiGSA) and a good example of this is weep6
- Another factor is the length or duration of the plateau phase. A shorter plateau phase determines a steeper RSA.



626 Mutants Found
So Far

Steep LRiGSA Mutant: 10-10-8

Sha



weep6



10-10-8



Note: the Mutant ID is (Pool # - Mutant # - Wave #)

Steep LRiGSA Mutant: 32-6-10



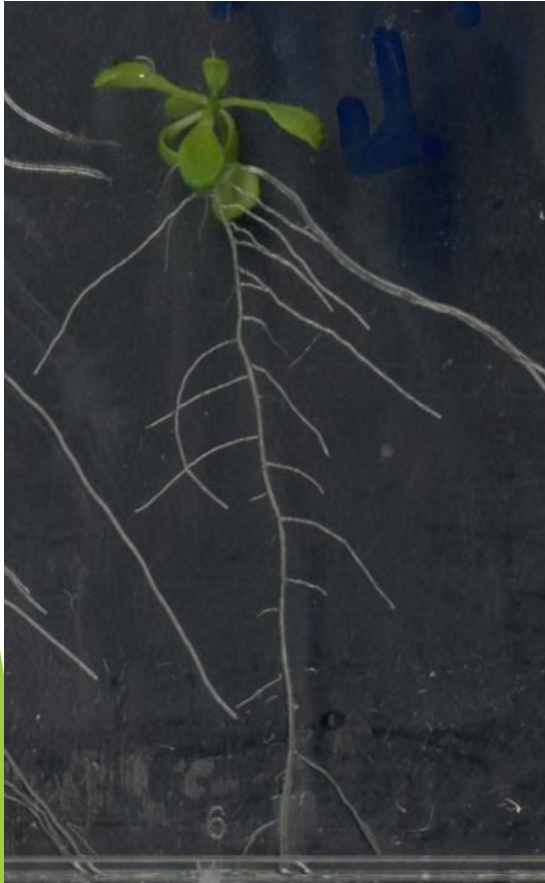
32-6-10

weep6

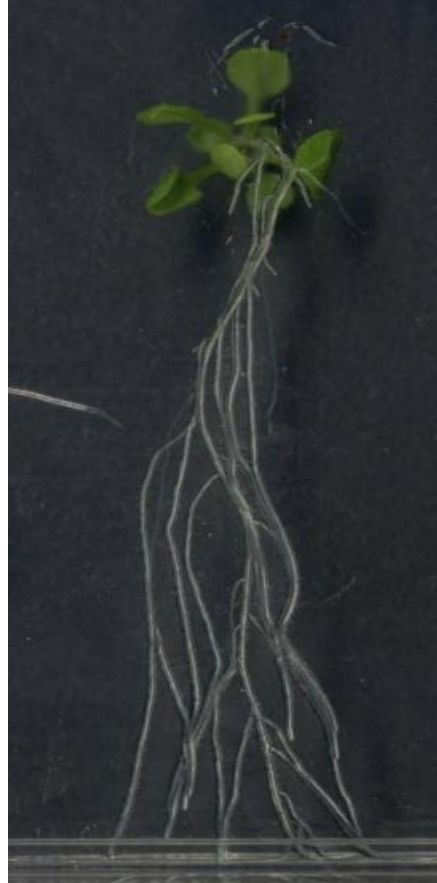
Sha

Steep LRiGSA Mutant: 15-3-9

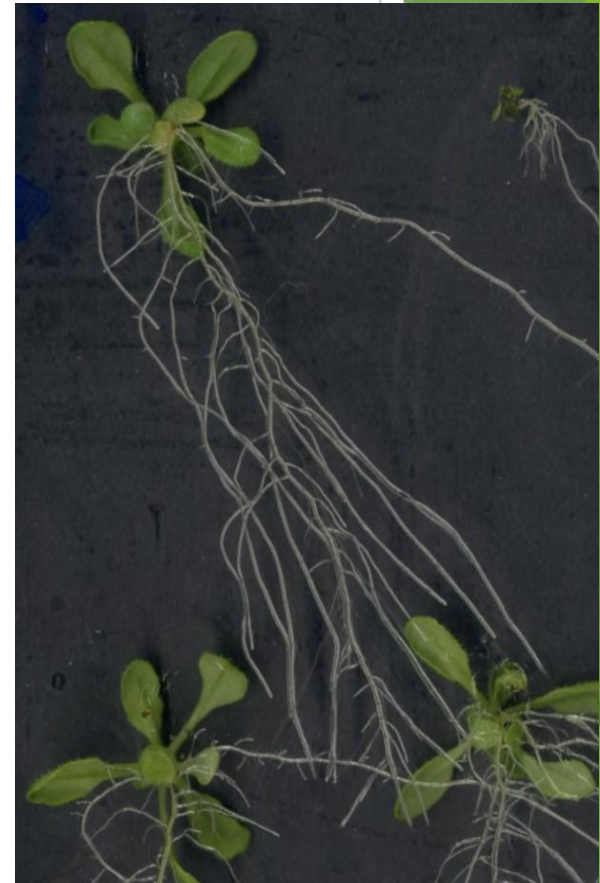
Sha



weep6



15-3-9



Note: the Mutant ID is (Pool # - Mutant # - Wave #)

Steep LRiGSA Mutant: 22-5-9

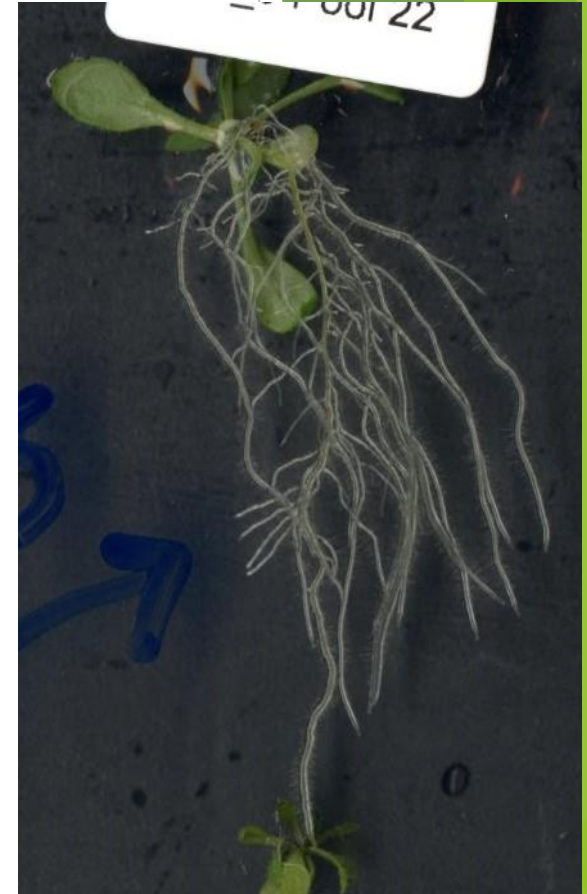
Sha



weep6



22-5-9



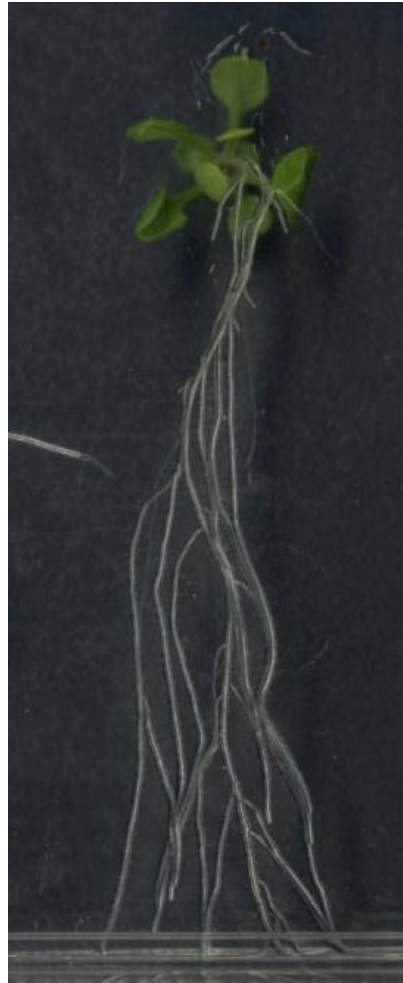
Note: the Mutant ID is (Pool # - Mutant # - Wave #)

Steep LRiGSA Mutant: 31-7-9

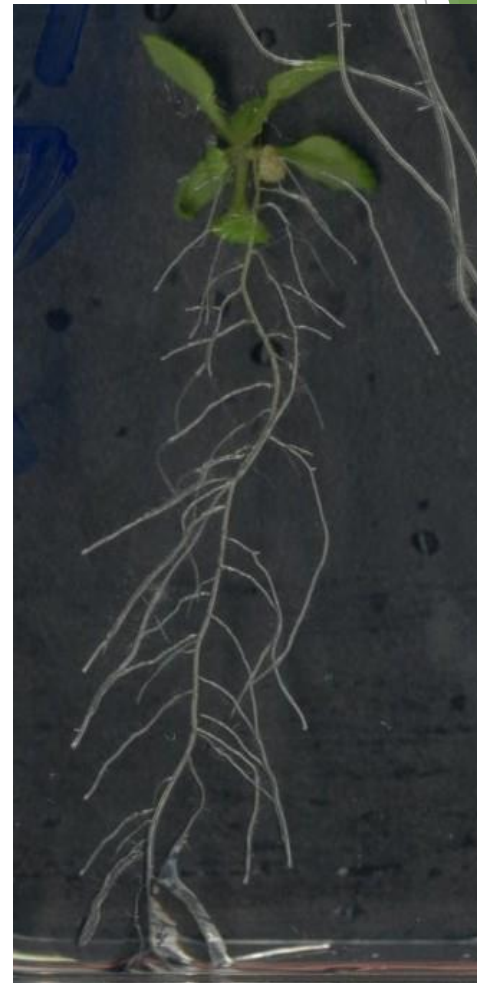
Sha



weep6



31-7-9



Note: the Mutant ID is (Pool # - Mutant # - Wave #)

Plateau Length Mutant: 26-3-9



26-3-9

26-3-9 mutant has an RSA that distinguishes it from basically every other seedling on the plate and not only the parental Sha control

Sha

Plateau Length Mutant: 49-2-10



Sha-like mutant

49-2-10

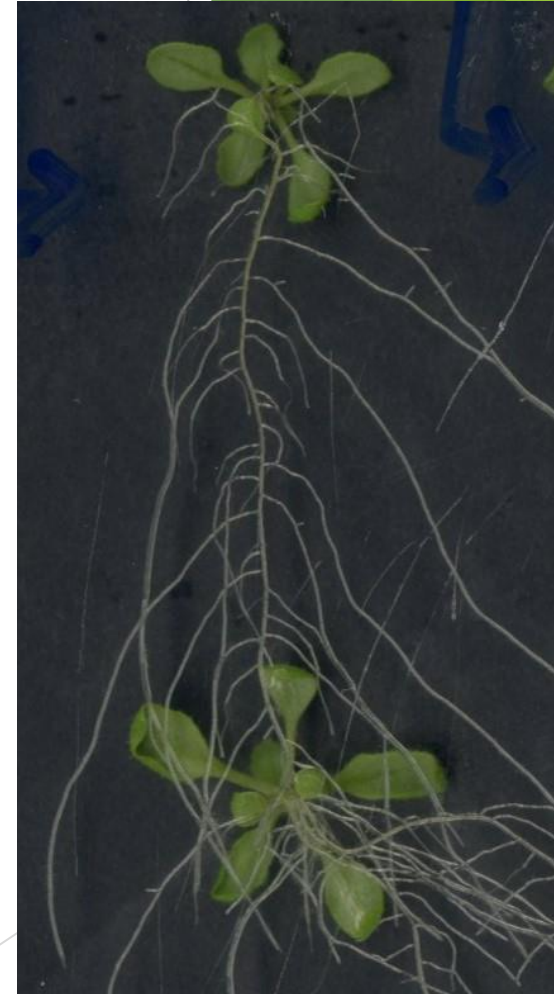
49-2-10: The Sha control on that plate is not great because it grew much less than normal, the contrast with the mutant above is very clear

Plateau Length Mutant: 13-8-9

Sha

weep6

13-8-9



Note: the Mutant ID is (Pool # - Mutant # - Wave #)

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Steep LRiGSA and Plateau Length Mutant: 18-1-9

Sha



weep6



18-1-9



This mutant could be a combination of both steeper LRiGSA and a shorter plateau phase.

EMS Summary

- ▶ Mutagenesis Occurred September 2023, Waves began January 2024 through present
- ▶ Total Number of Waves: **12 so far**
- ▶ Total Number of Mutants Screened: **11,380 so far**
- ▶ Total Number of Steep Architecture Mutants Found So Far: **626 so far**
- ▶ The total rate of discovery oscillates between: **1% and 16%**
- ▶ A problem in the EMS Screen is that not all mutants transferred after identification survive in soil.
 - ▶ For example, in Wave 7 the survival rate of transplanted mutants was: **46%**

Thank You

- ▶ Acknowledgements:

- ▶ Wolfgang Busch
- ▶ Michel Ruiz
- ▶ Melissa Baez
- ▶ Juan Gonzalez
- ▶ Natalie Gonzalez
- ▶ Shane Hunt
- ▶ Petra Banuet
- ▶ Charlie Pope