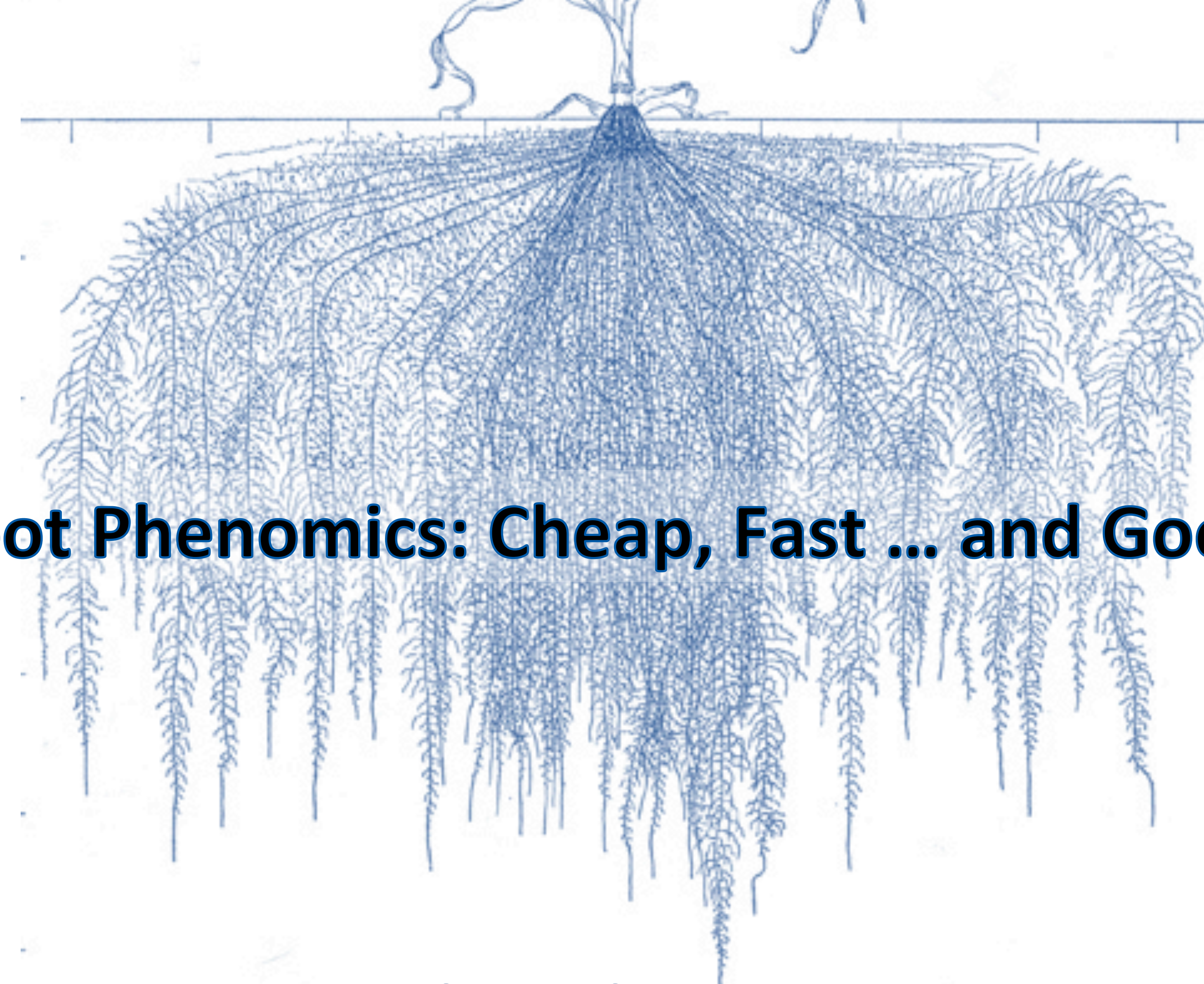
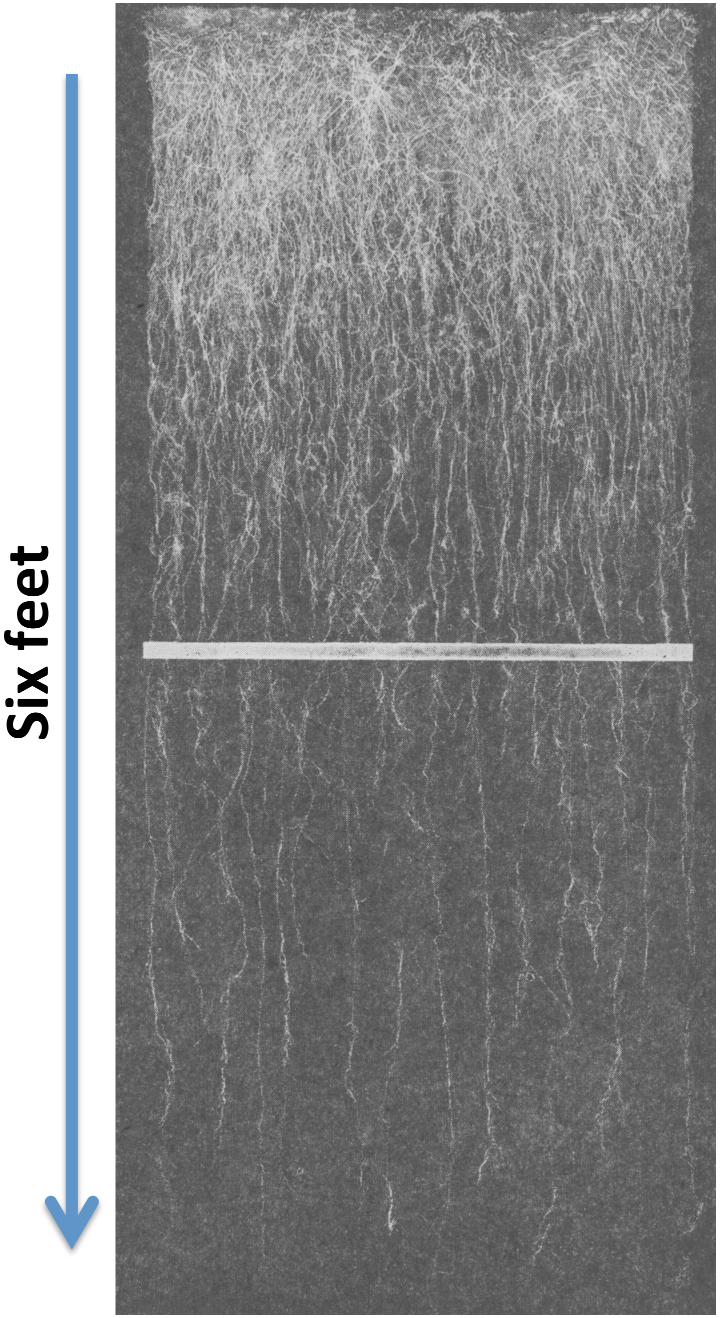
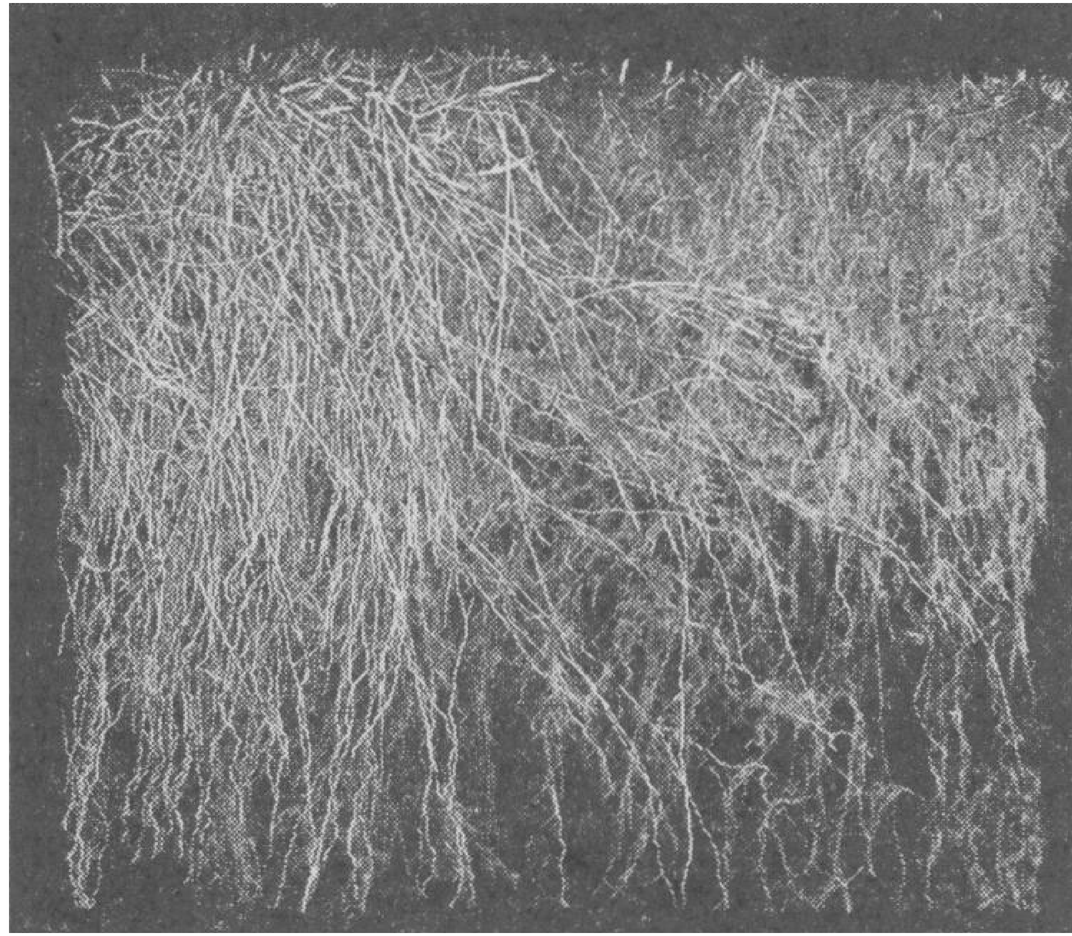


Root Phenomics: Cheap, Fast … and Good?

Christopher Topp
Donald Danforth Plant Science Center



Root interactions



N = few

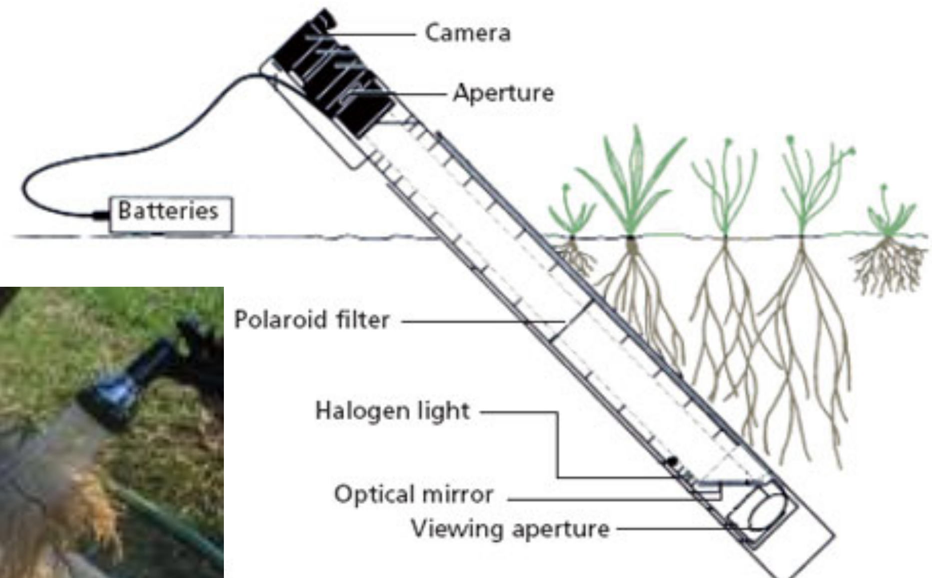
Weaver and Voigt Botanical Gazette 1950

Impact opportunity?

Better field phenotyping of roots

serc.carleton.edu

www.dpi.nsw.gov.au

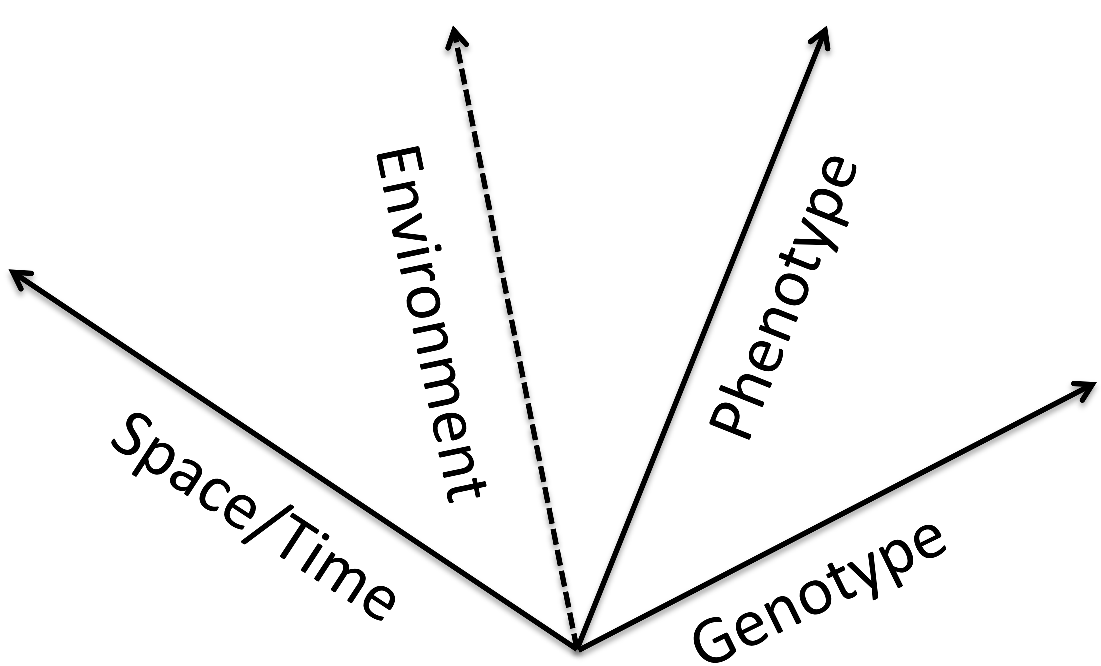


Poelman et al Plant and Soil 1996.

http://rps.psu.edu/indepth/roots.html

Phenotyping is vastly more dimensional than genotyping

There is not, and will not be, one all encompassing phenotyping tool



Phenotypes are many.
And depend on context.

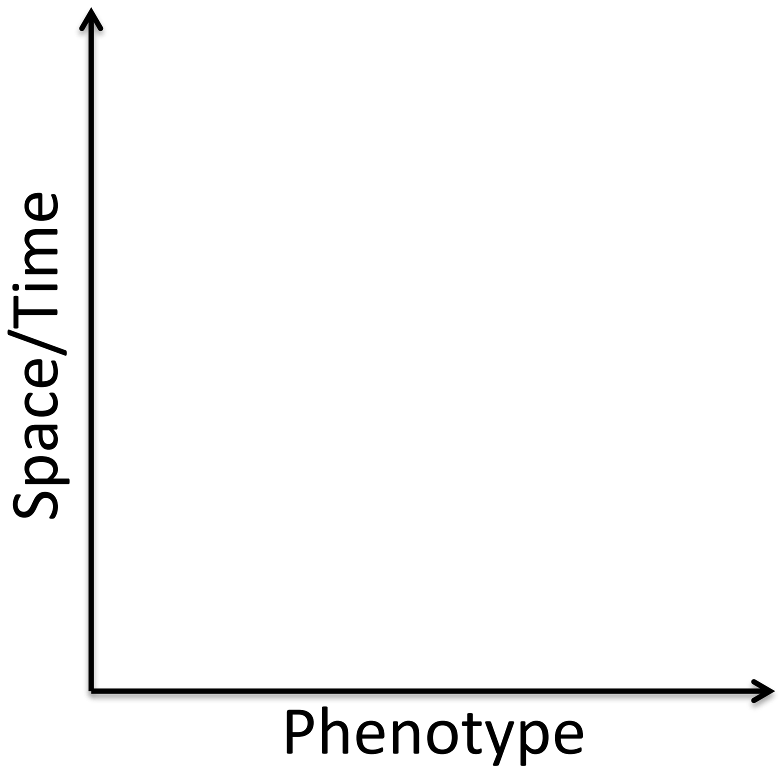
Phenotypes

Gene expression
Protein levels
Metabolite levels
Cells
Tissues
Organs – (roots, shoots, flowers)

Physiological: carbon allocation/
vascular function
Behavior/ environmental response
Plasticity
Growth rates
Macro and micro architecture
Et al.

→ Phenotype

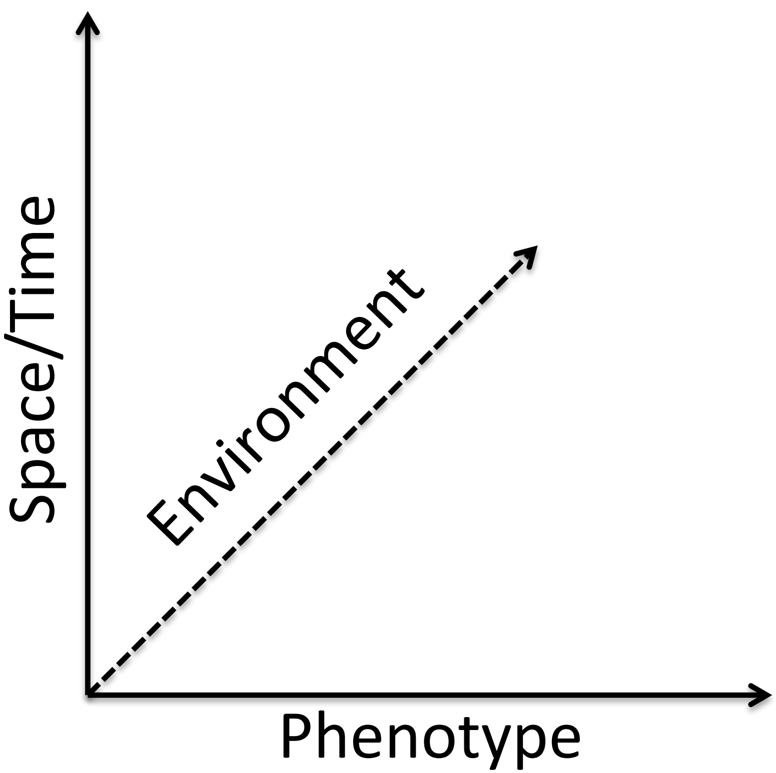
Phenotypes are many.
And depend on context.



Space/Time

Dynamic interactions
Development
Maturation/ Flowering time
Relative growth of organs
Circadian effects
Tradeoff constraints

Phenotypes are many.
And depend on context.

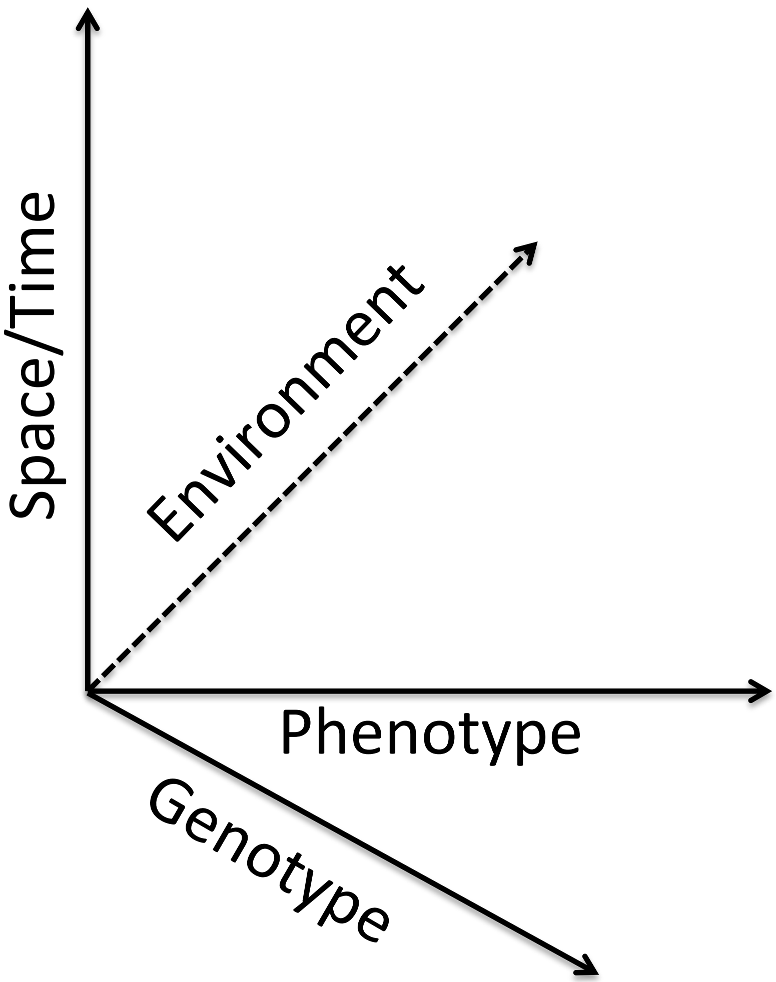


Environment

Abiotic
- Drought
- Light
- Ionic
- Osmotic
- Temperature
- Chemical

Biotic
- Pathogens
- Symbionts
- Competitiors
- Local Microbiome

Phenotypes are many.
And depend on context.

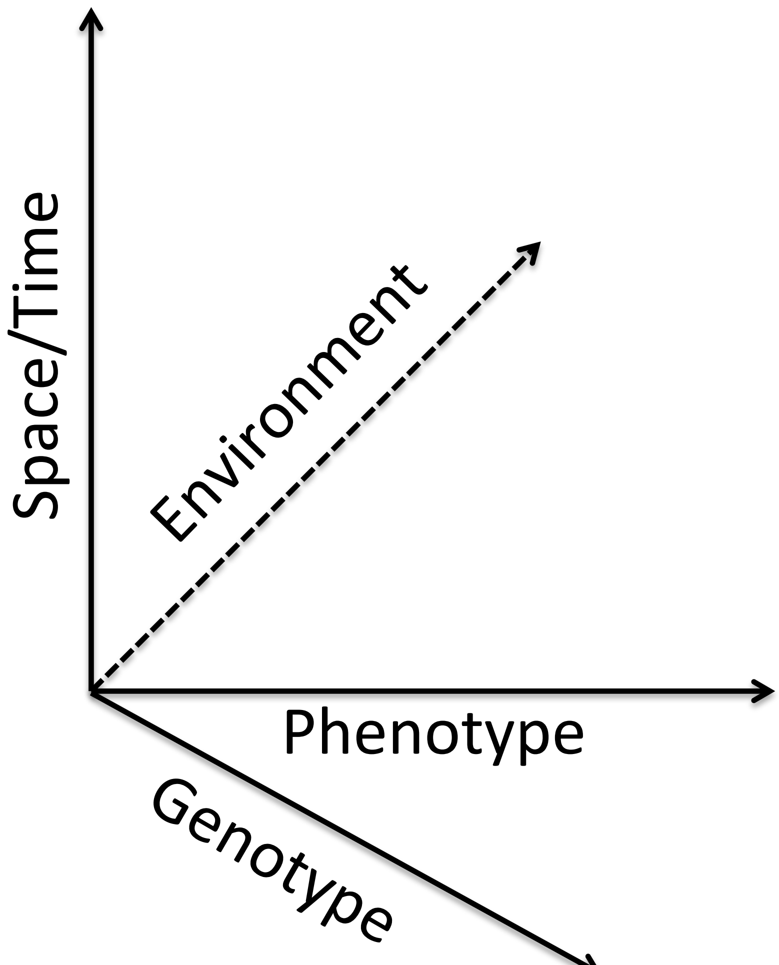


Genotype

> Incredible wealth of untapped information in natural variation

> Extremely powerful genetic resources have been developed

What phenotypes do we want to measure?
And in which contexts?



Sampling many levels

of many factors

and their interactions:

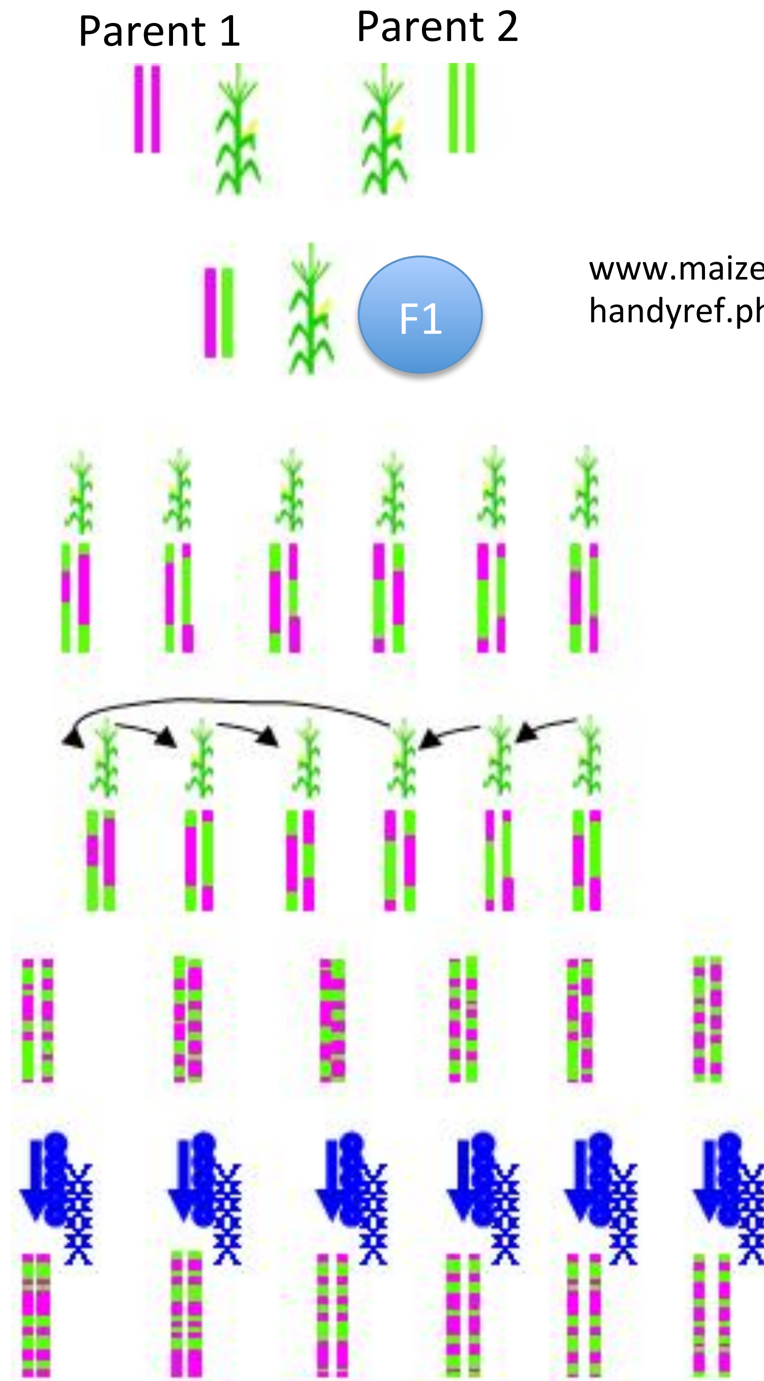
Requires high sampling

and high throughput!!!

We can leverage existing technologies in industrial engineering, robotics, computer vision/AI

Rick van de Zedde – Wageningen UR

‘High Resolution’ genetic resources are designed to rapidly associate phenotypes with the genes controlling them



www.maizegdb.org/handyref.php

<u>Several generations of intermating</u> →

Several generations selfing →

High throughput genotyping →

Root phenomics: major questions

1. How can we image roots?

2. What phenotypic variation exists and how can we quantify it?

3. What genetic and environmental factors condition root architecture, and what tradeoffs exist?

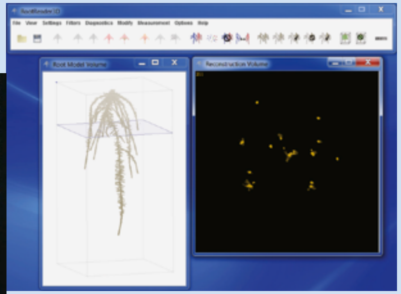
4. How do local (2D) growth decisions “add up” to global 3D shape?

5. What biological function does phenotypic variation have?

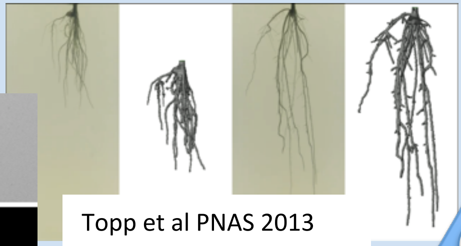
Phenotyping

Optical Tomography

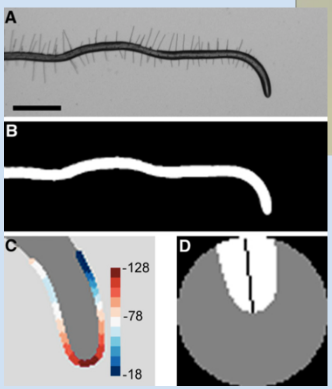
Iyer-Pascuzzi et al Plant Phys 2010



Clark et al Plant Phys. 2011

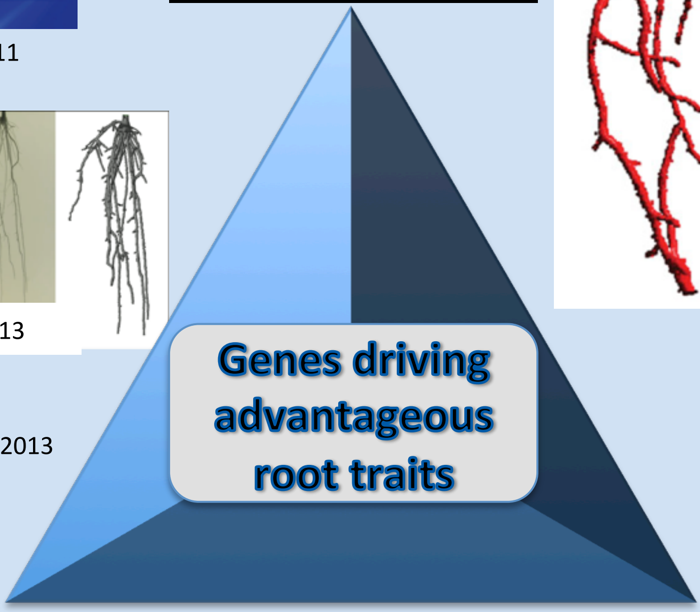


Topp et al PNAS 2013



Moore et al Genetics 2013

precision and accuracy

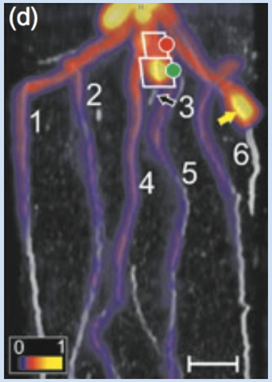


high throughput

environmental complexity

X-ray Computed Tomography
Mairhofer et al Plant Phys. 2012

MRI and PET
Schurr et al Plant Journal 2009

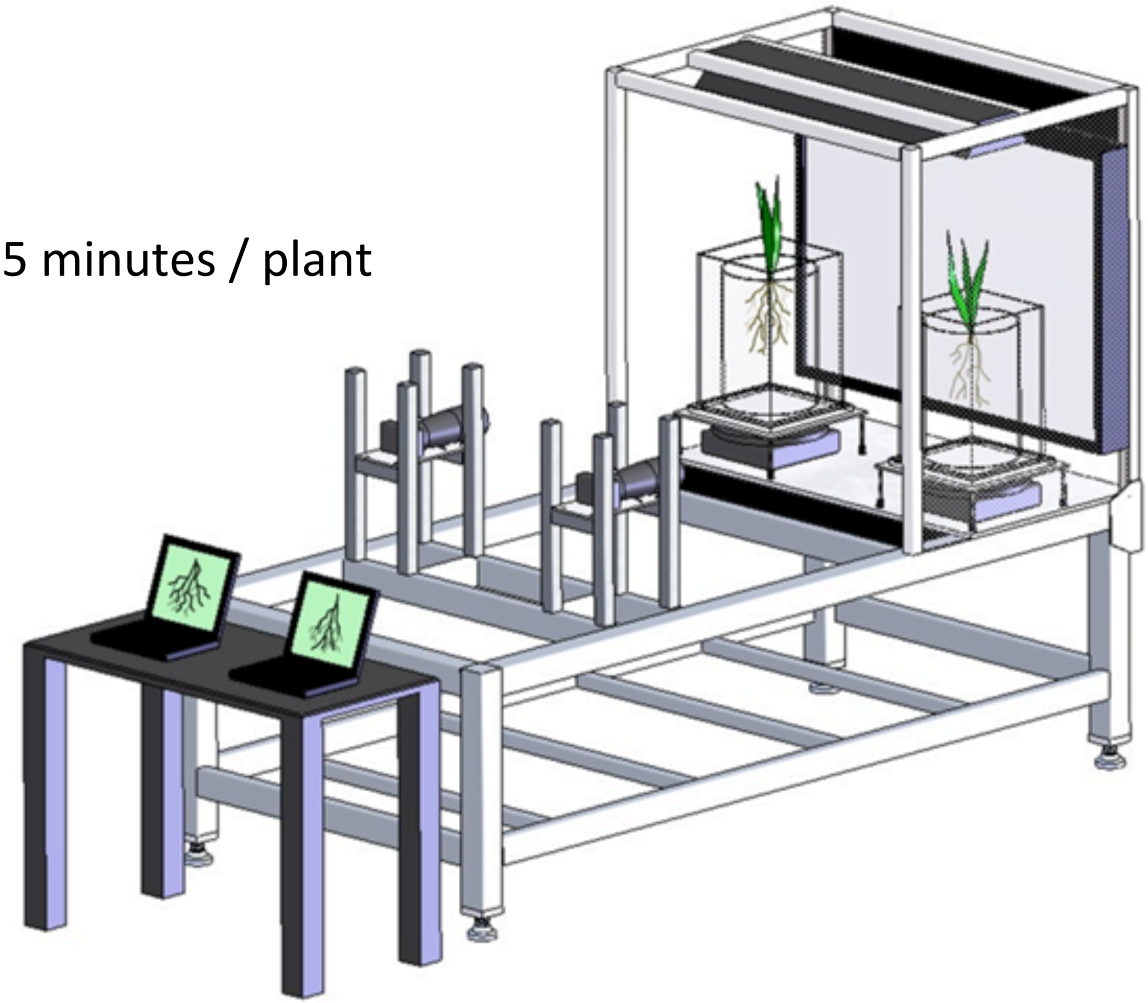


Automated rhizotron
Nagel Fun. Plant Biology 2012

Shovelomics
http://rps.psu.edu/indepth/roots.html

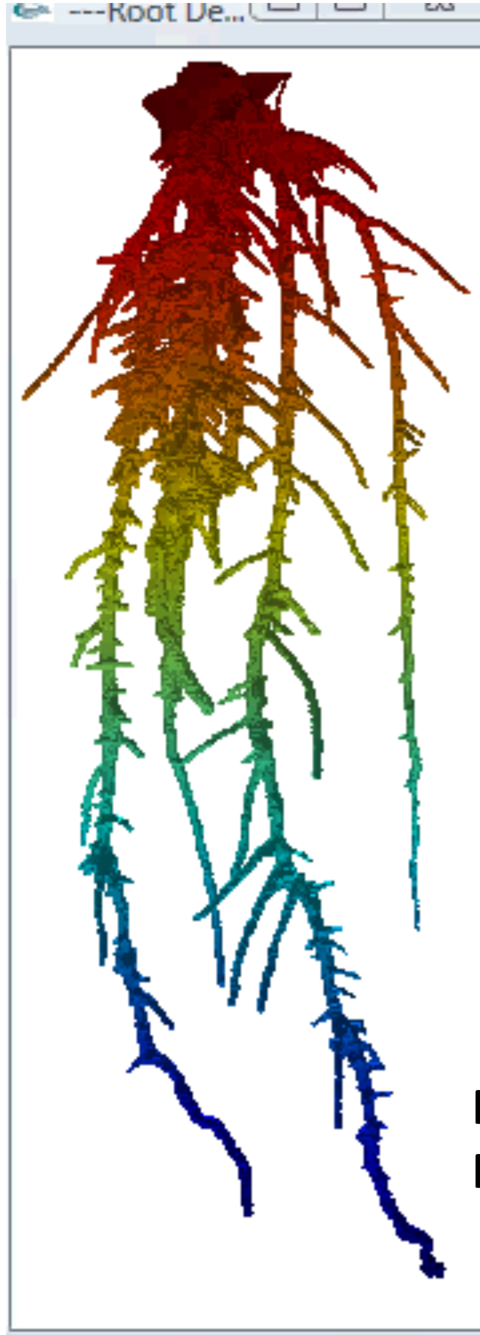
Optical Projection Tomography (OPT) platform for root phenotyping in 3D

Imaging time: ~5 minutes / plant



Tim Horn – mechanical engineer extraordinaire
Randy Clark – bio-engineer
Alex Bucksch and Joshua Weitz – physics and computer science

3-dimensional modelling of root architecture

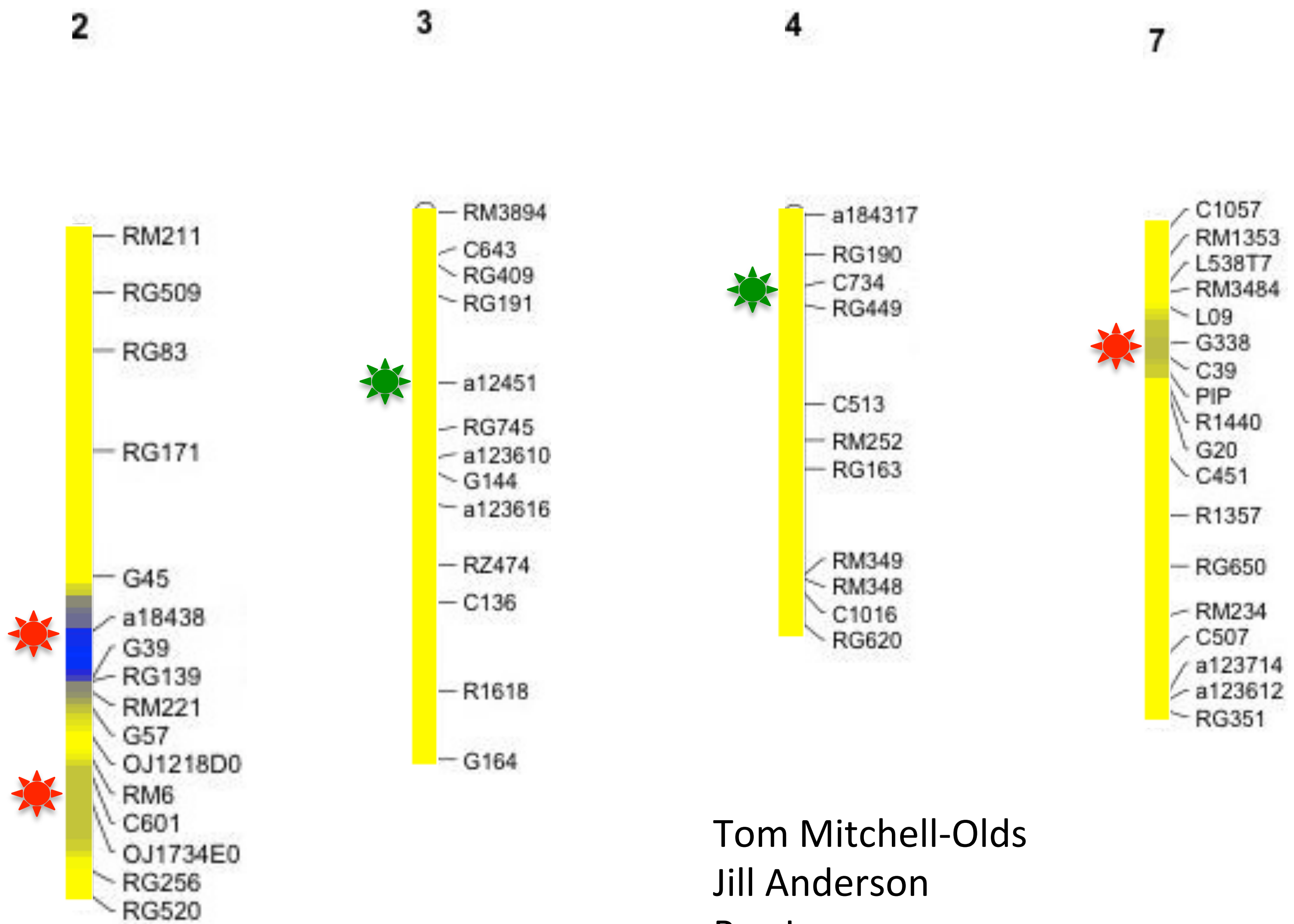


traits analyzed in 3D

1. median root number
2. maximum root number
3. root system volume
4. convex hull volume
5. solidity
6. surface area
7. bushiness
8. total root length
9. root system volume
10. specific root length
11. number of branches
12. et al.

Rootwork - Zheng et al ICCV 2011
RootReader3D - Clark et al Plant Phys 2011

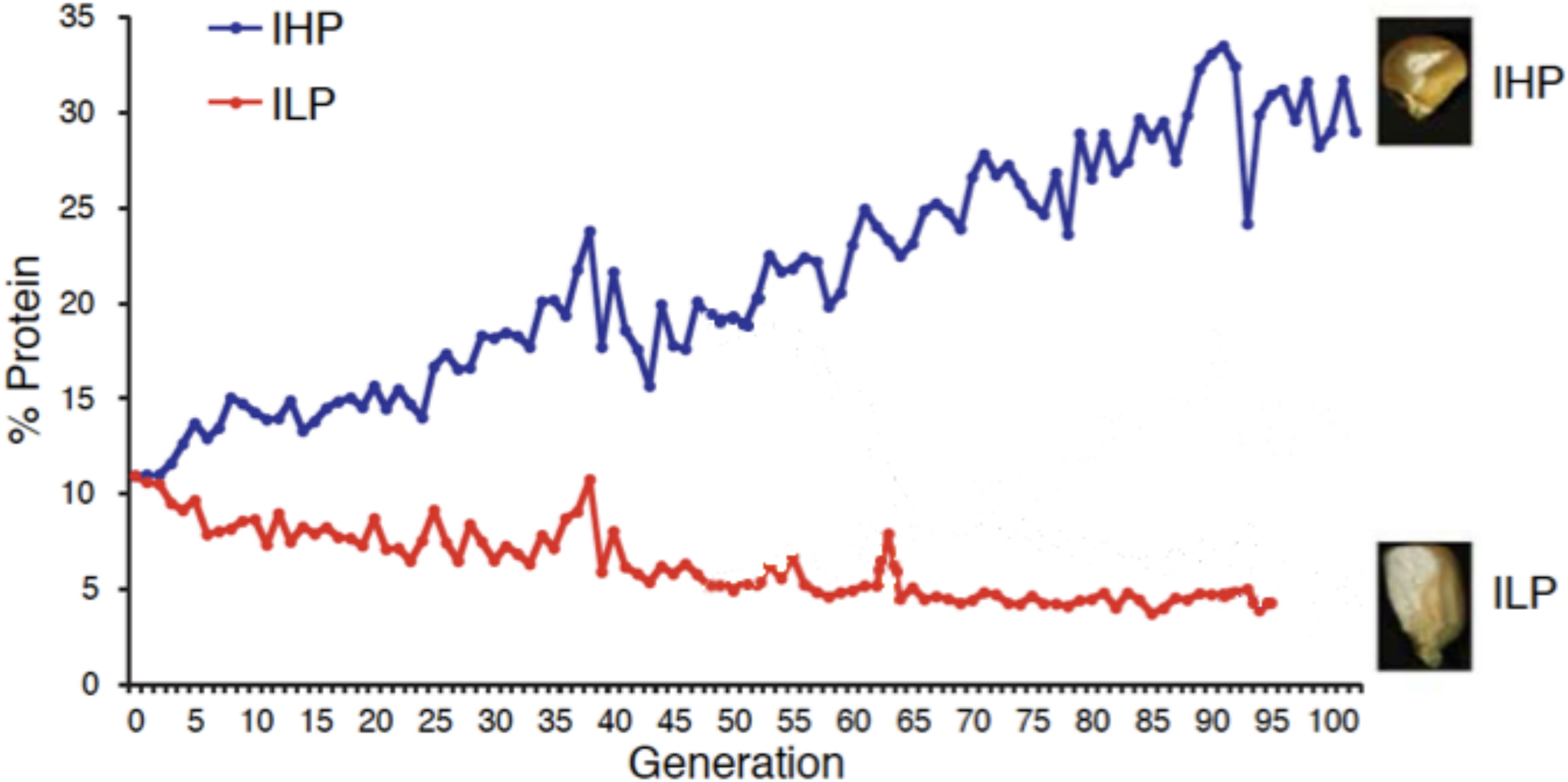
Image-based phenotyping allows the genetic basis of new 'traits' to be identified



Tom Mitchell-Olds
Jill Anderson
Ray Lee

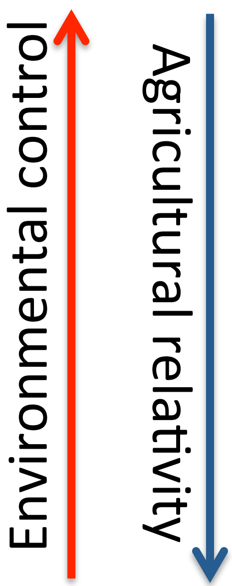
Illinois Long-term selection experiment

In >100 years of selection for seed protein content, Nitrogen uptake capacity was also strongly selected for



modified from Moose et al. TIPS 2004

Identifying the genetic basis of root architecture:
integrated root phenotyping with high-resolution germplasm

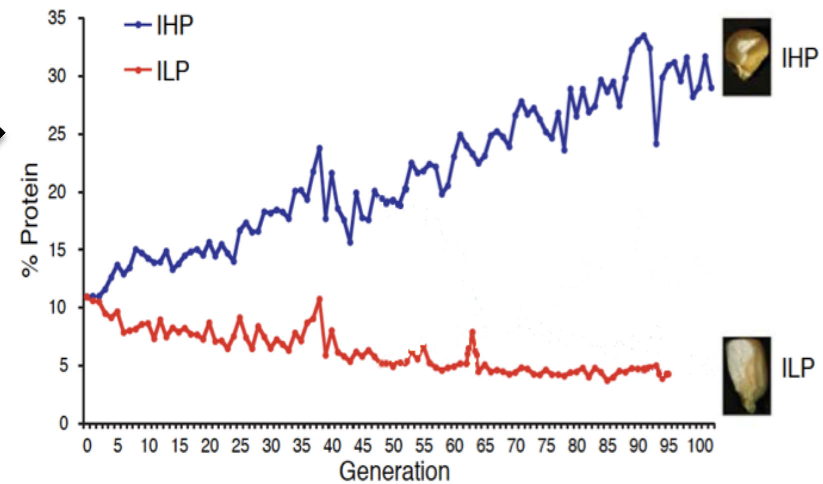


Root phenotyping methods

1. 3D gel imaging
2. X-ray CT in pots
3. Excavated root crowns:
4. minirhizotrons

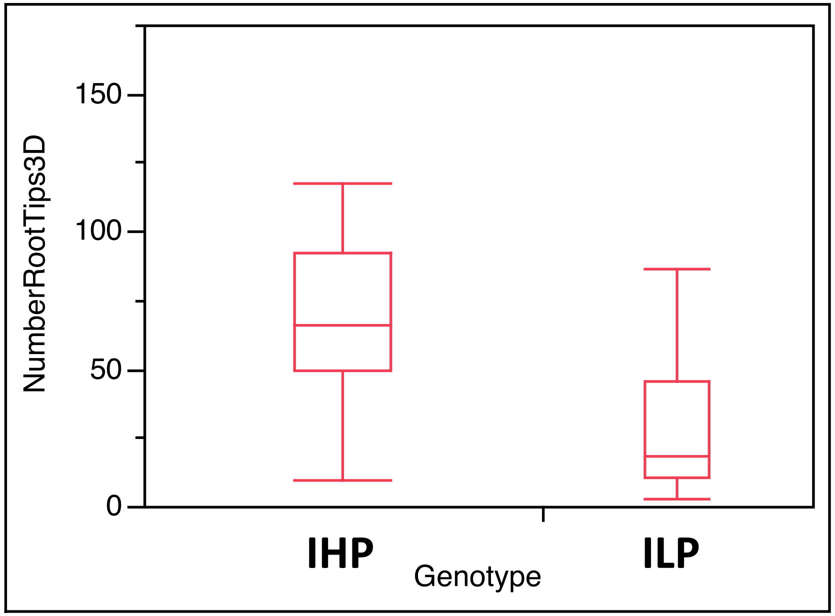
X

High-resolution germplasm with important agronomic traits



Characterizing potentially efficient high Nitrogen uptake root architectures in maize

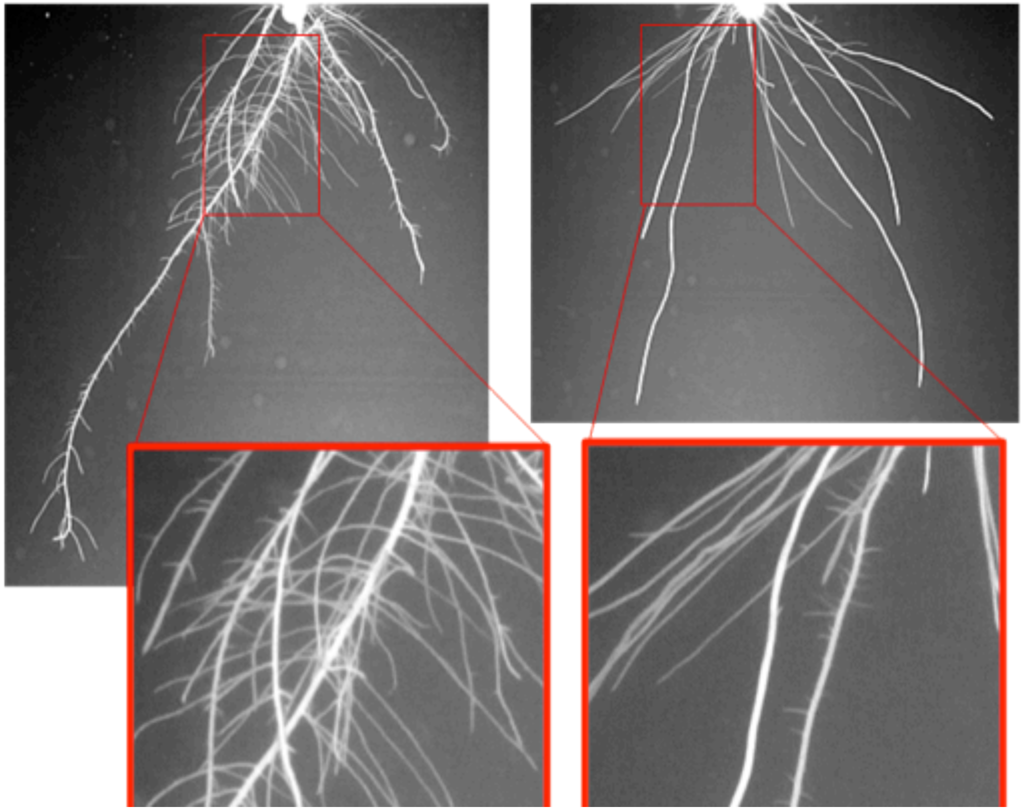
*# of root tips



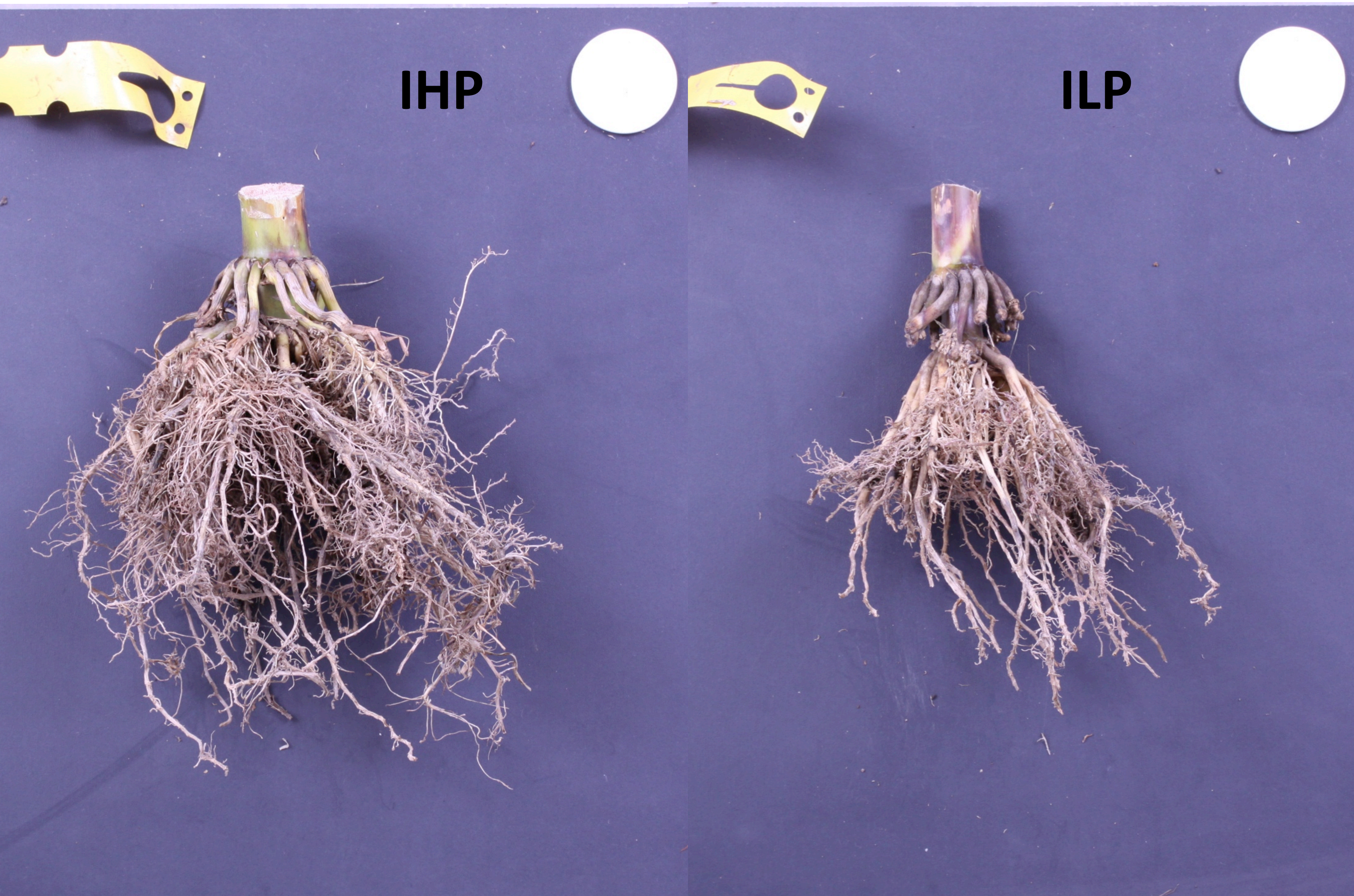
* = P < 0.05; Wilcoxon

IHP
High N uptake

ILP
Low N uptake



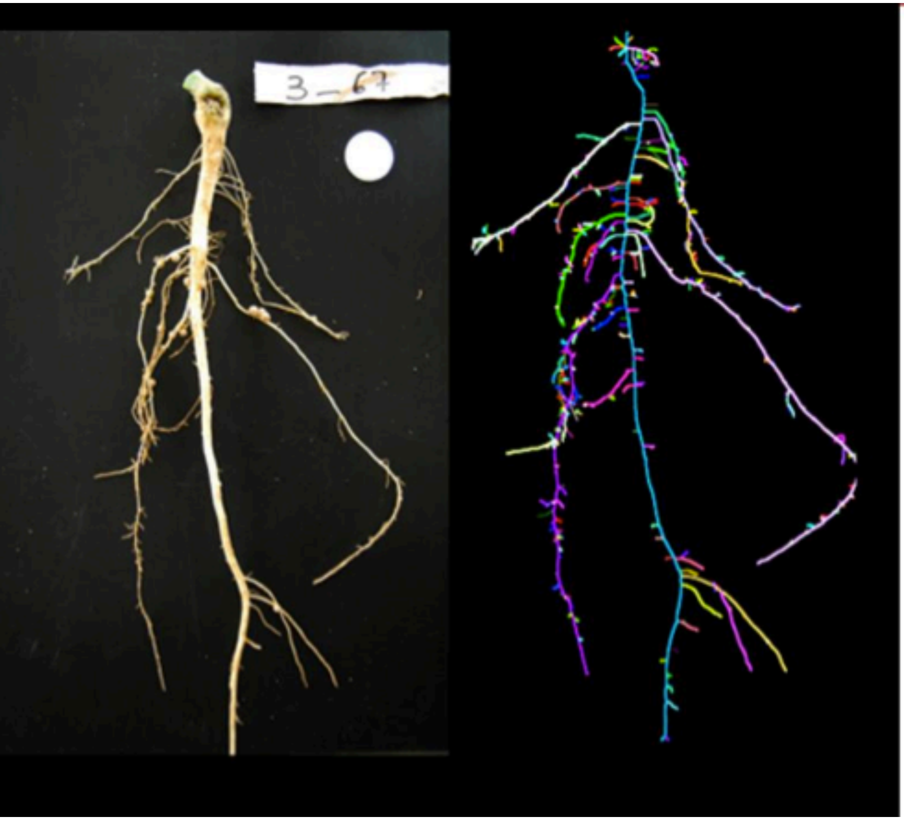
Field excavated maize roots

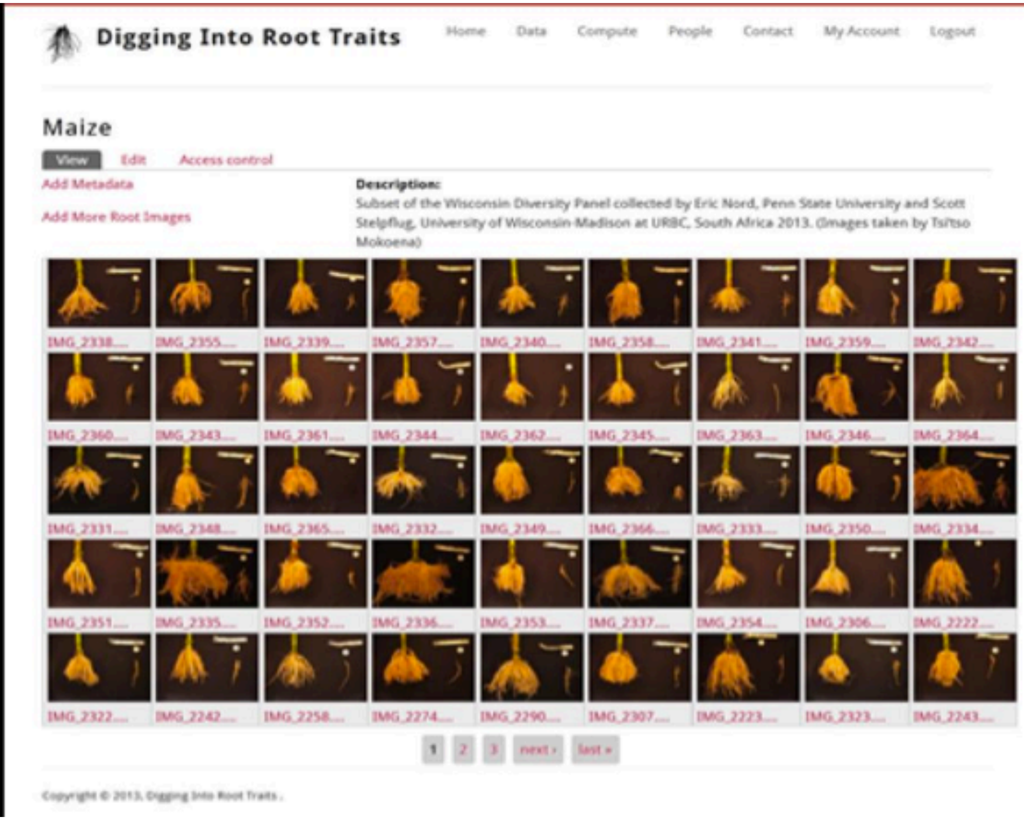


DIRT Pipeline: image based phenotyping of field excavated root samples:

Alex Bucksch and Joshua Weitz

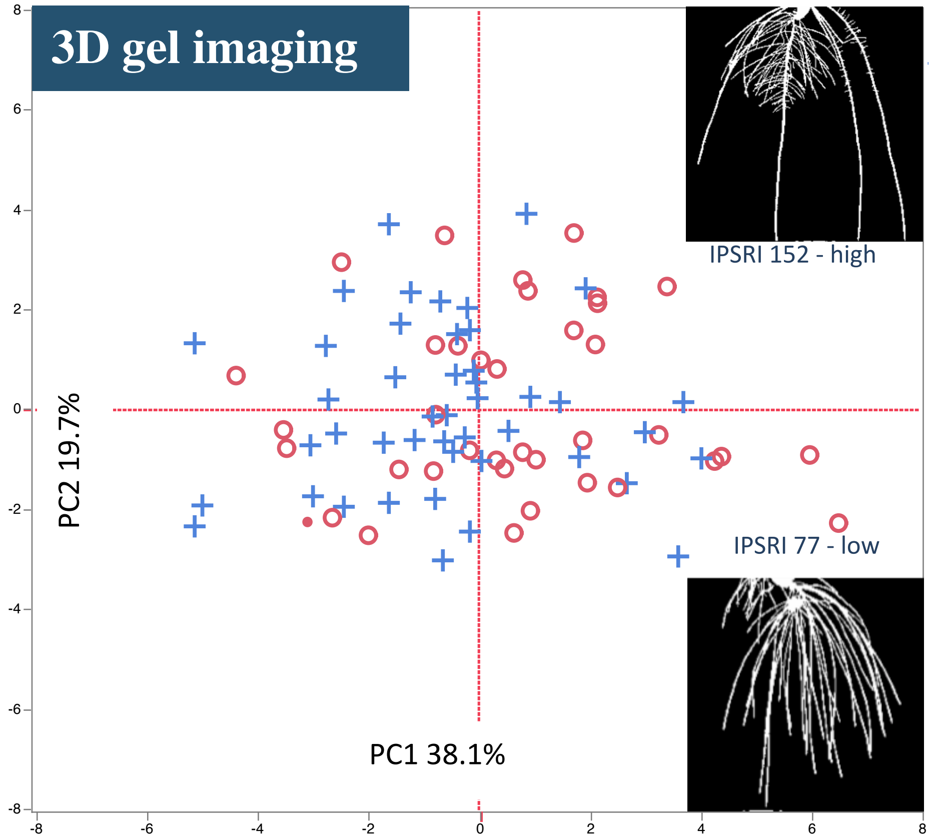
Jonathan Lynch and Eric Nord, Jimmy Burridge, Larry York

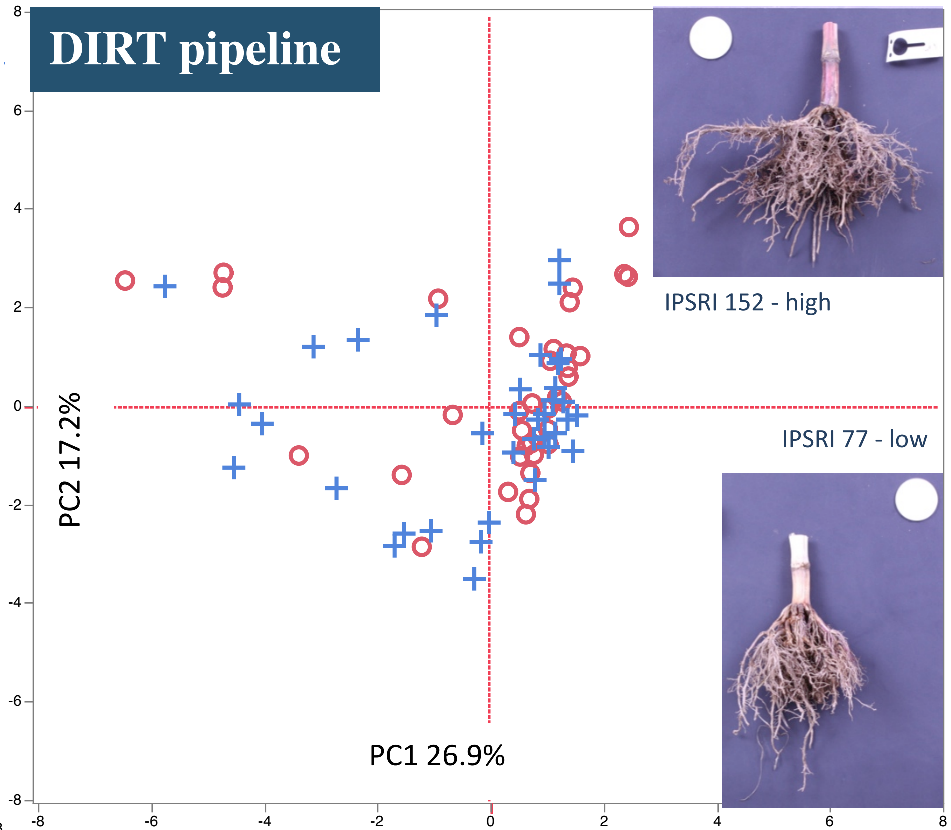




http://www.bucksch.nl/index.php/using-joomla

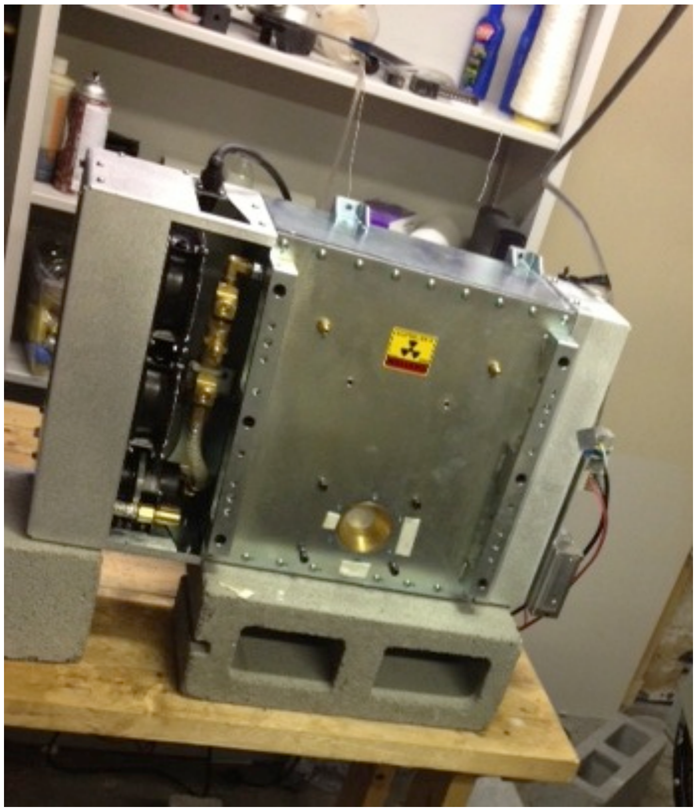
Converging lab and field phenotyping on high-resolution germplasm

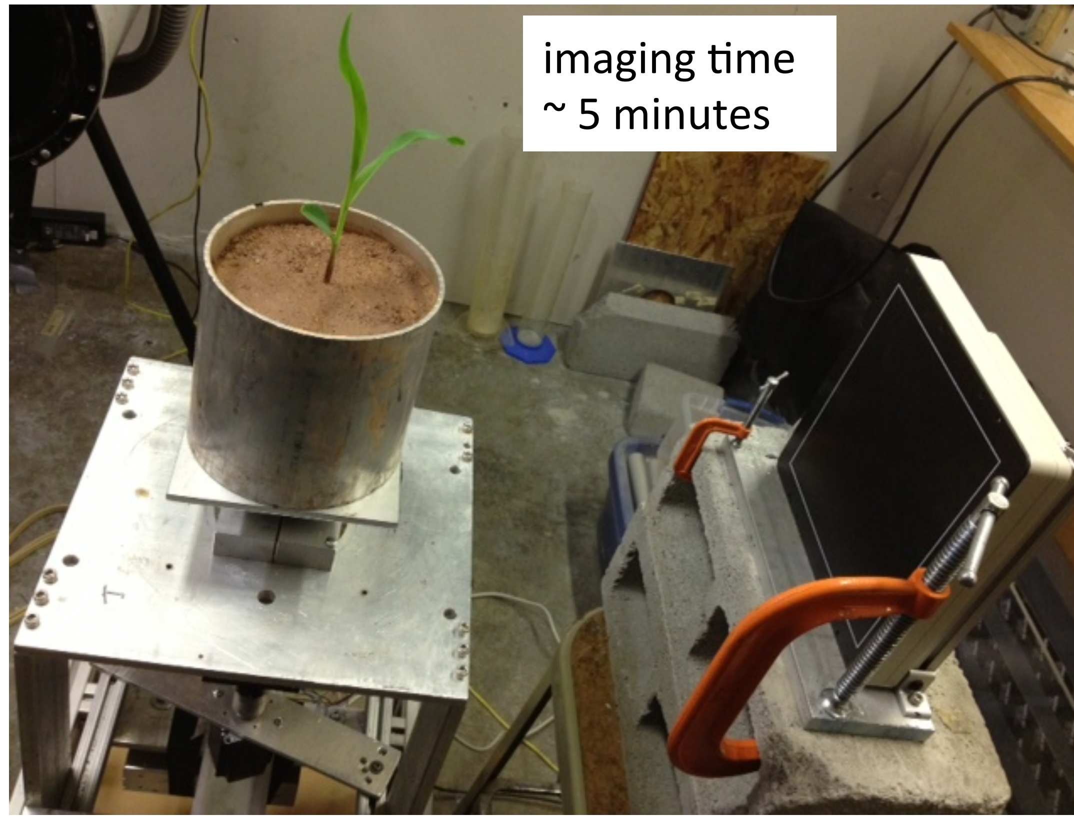




Intermediate phenotyping by X-ray CT:
realistic soils x controlled environment

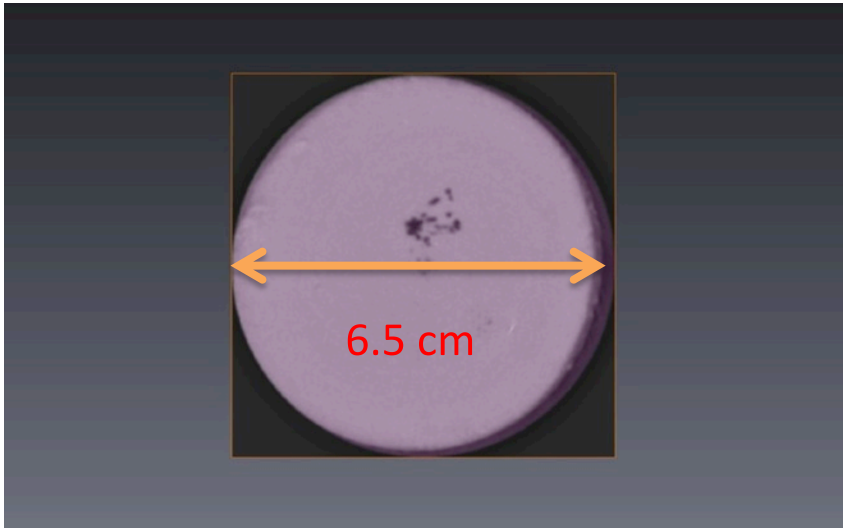
High-throughput X-ray computed tomography (X-ray CT) for comprehensive 3D shoot and root imaging in soils

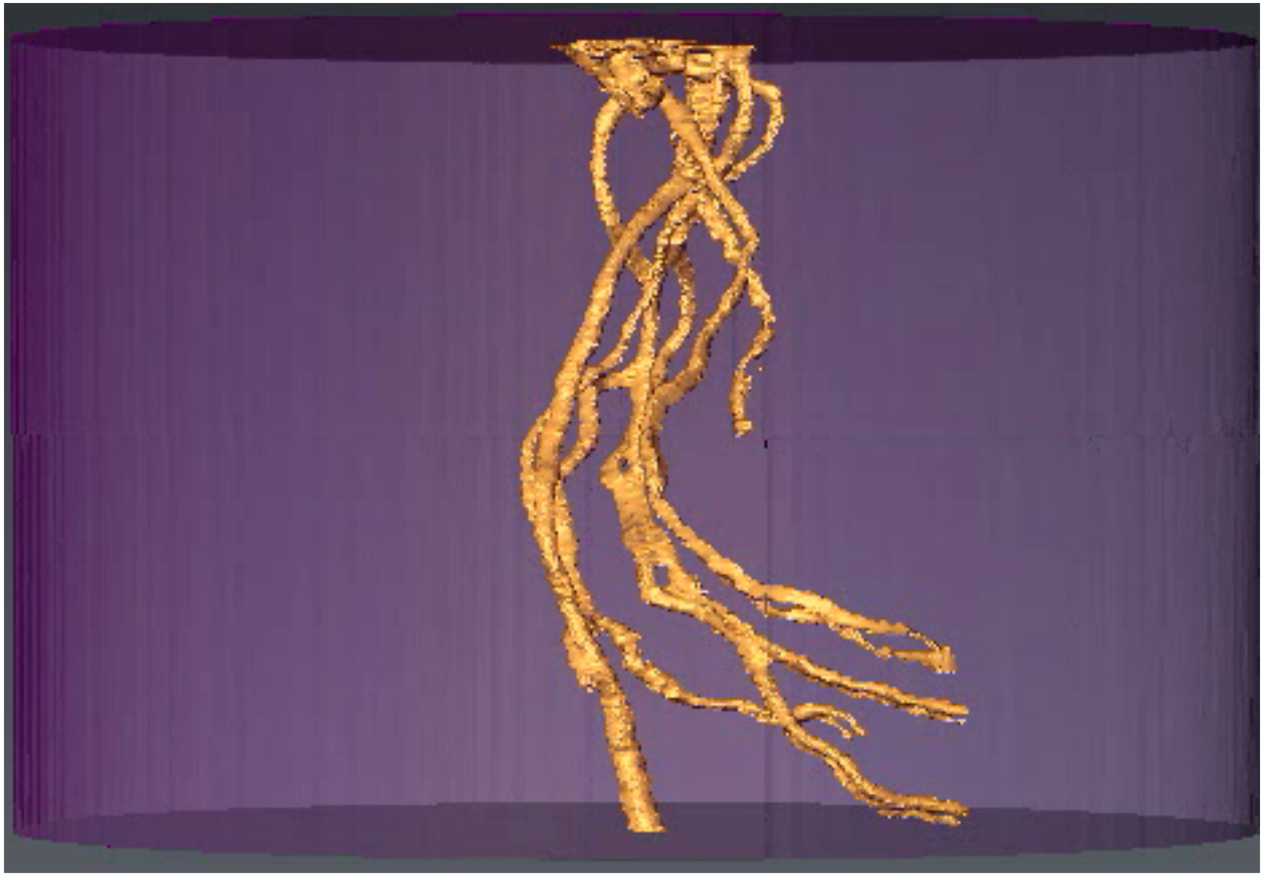




Daniel Goldman and Daria Moanenkova (GA Tech Physicists)

~ 10 day old rice root in fine sand particles

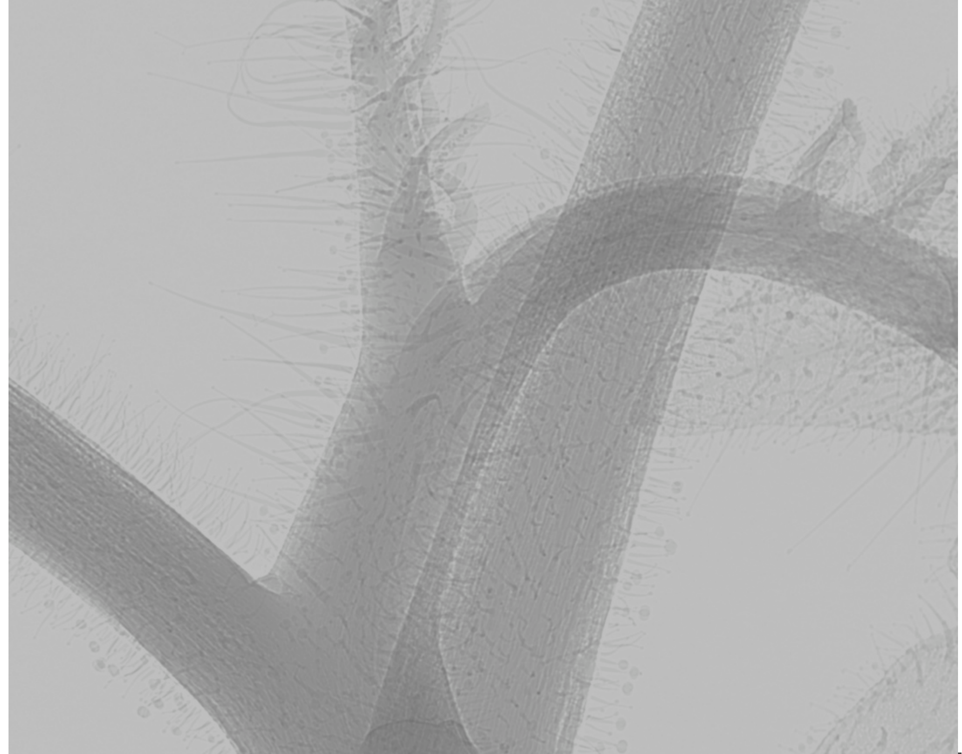


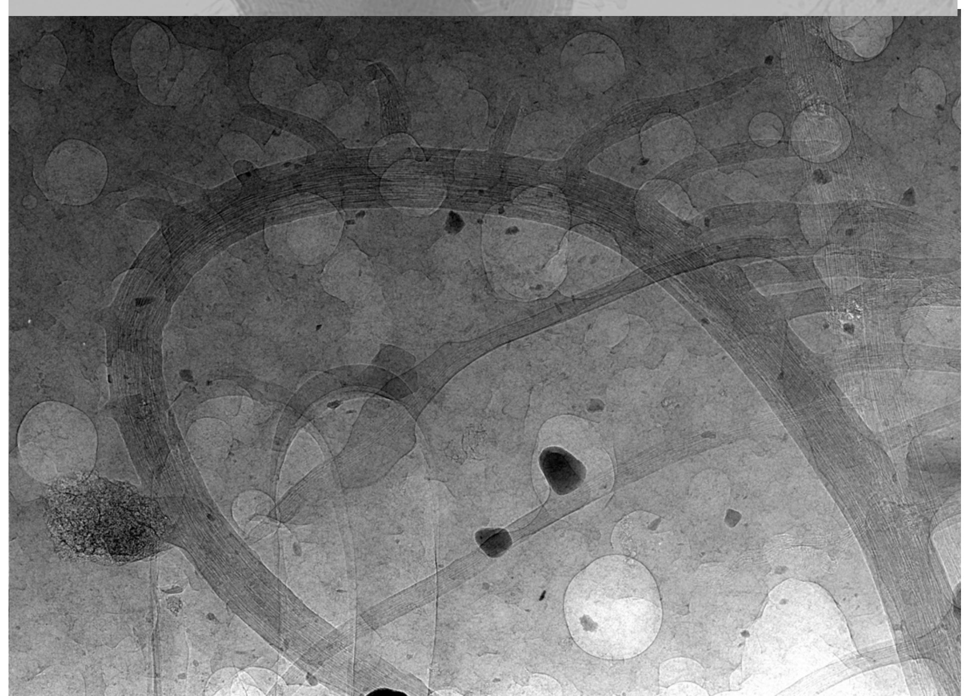


Micron resolution structures using X-ray CT

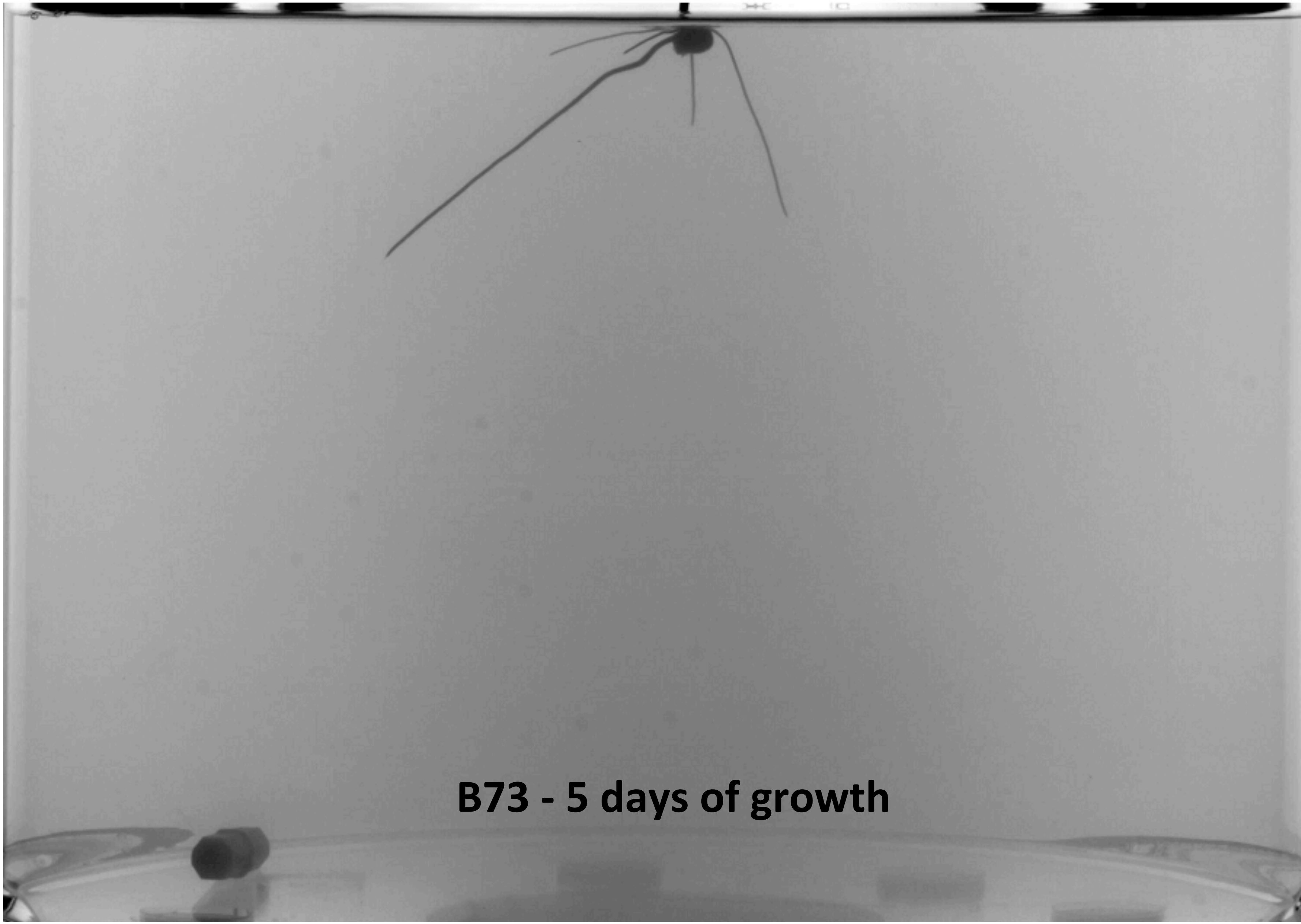
Mark Anastasio and Trey Garcon –
WashU Biomedical Engineering

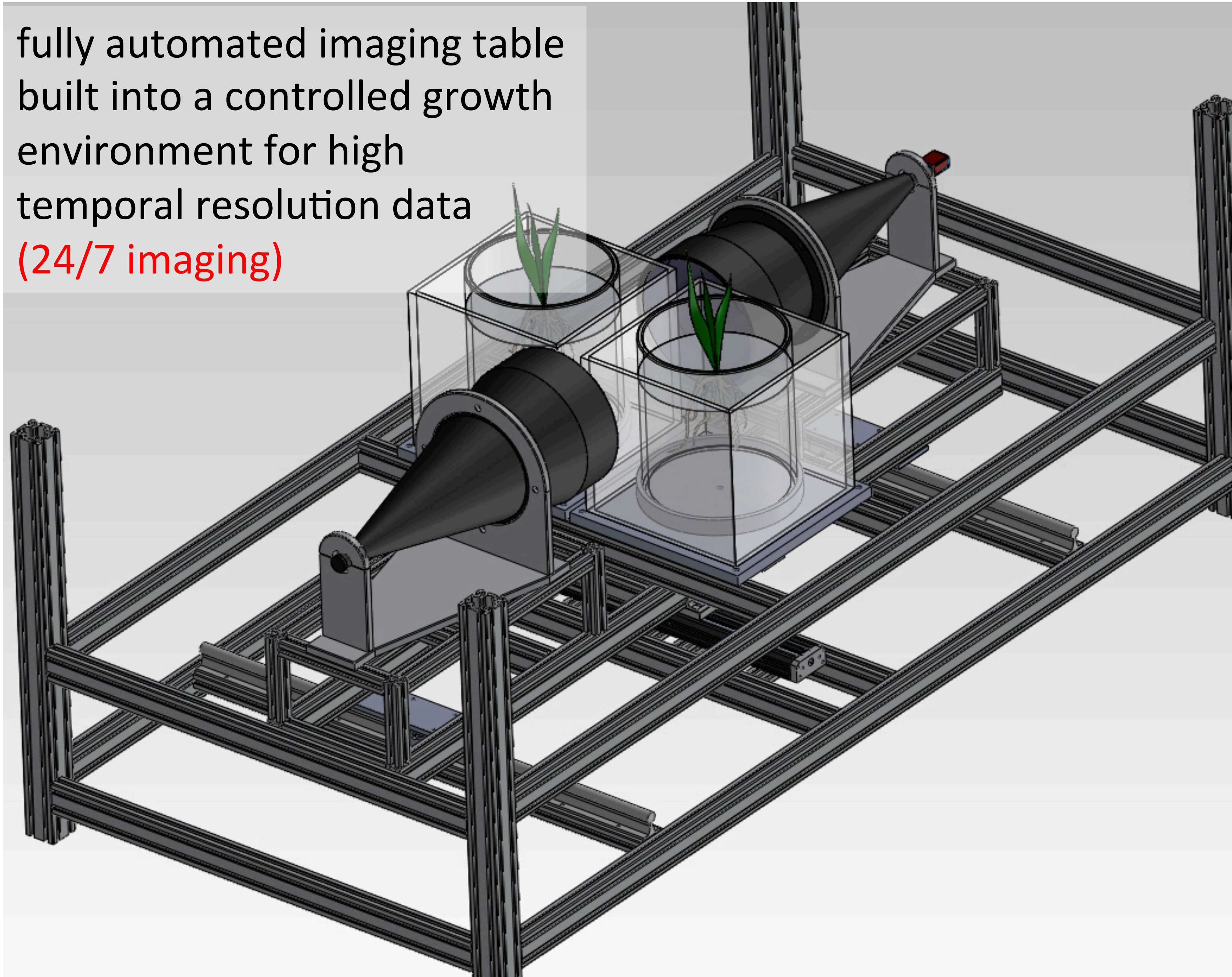
Dan Chitwood –
DDPSC

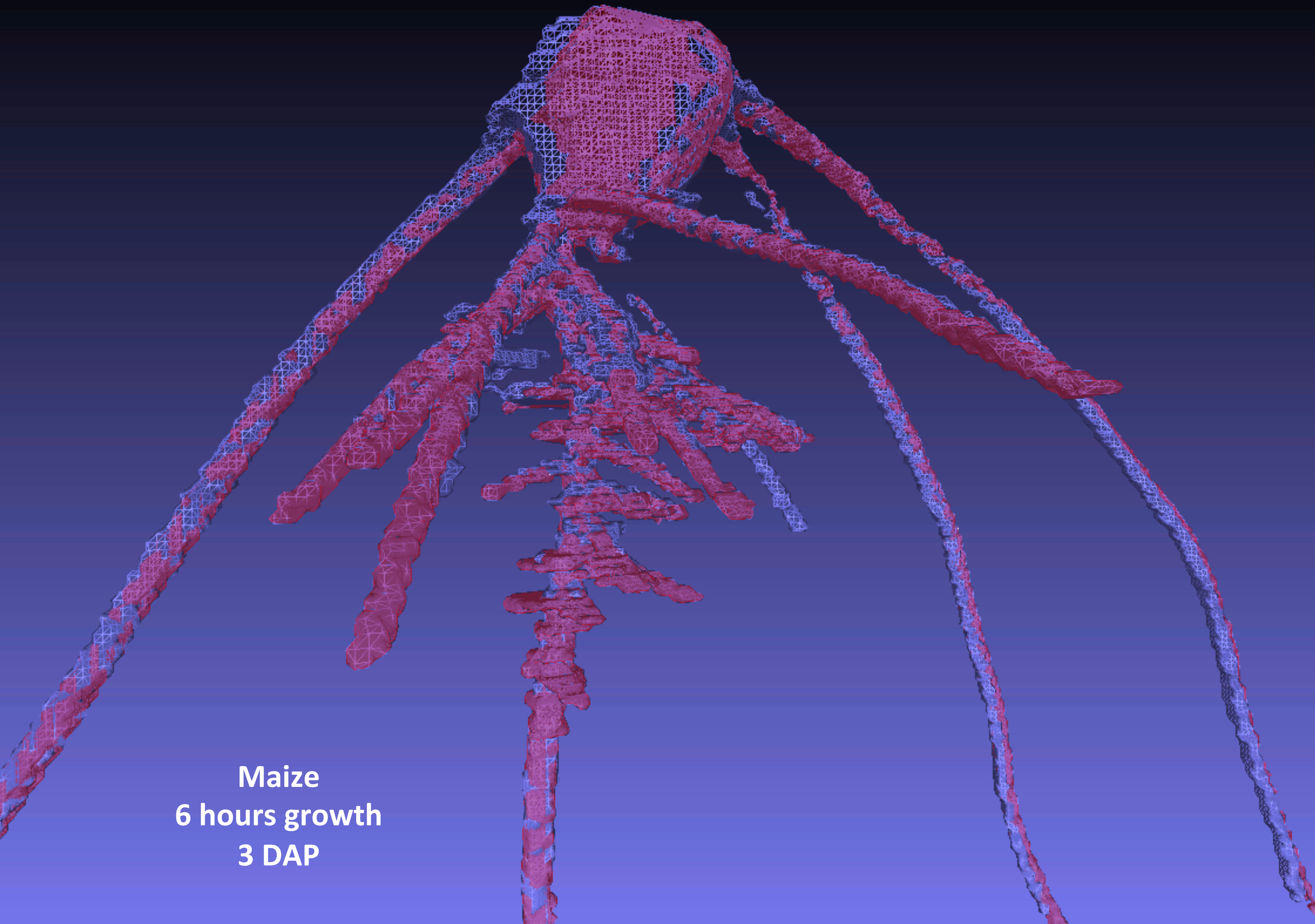




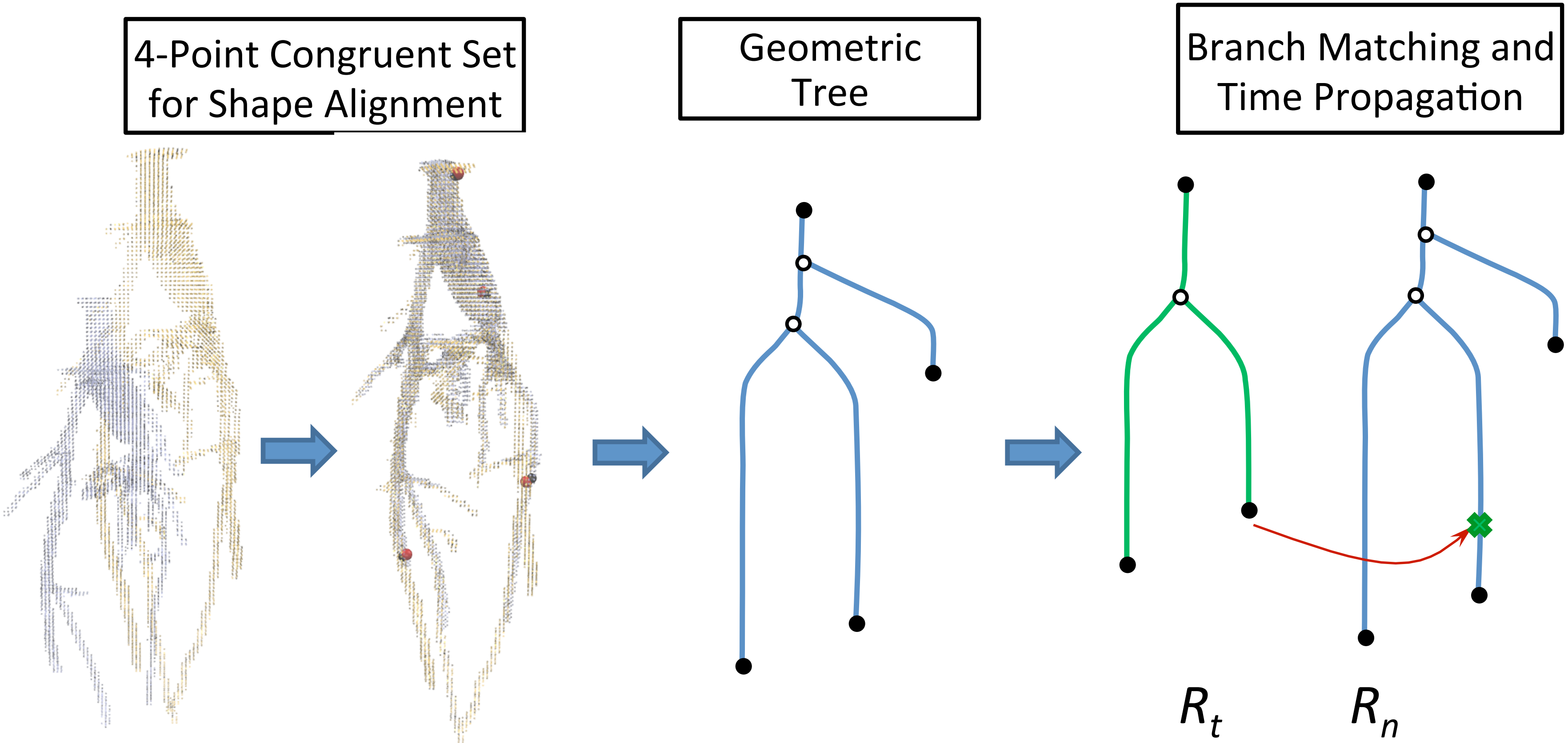
Capturing growth dynamics is key to understanding how roots interact with the environment




fully automated imaging table
built into a controlled growth
environment for high
temporal resolution data
(24/7 imaging)


Maize
6 hours growth
3 DAP

Growth model algorithm to quantify changes in RSA over time



Olga Symonova and Herbert Edelsbrunner, IST Austria

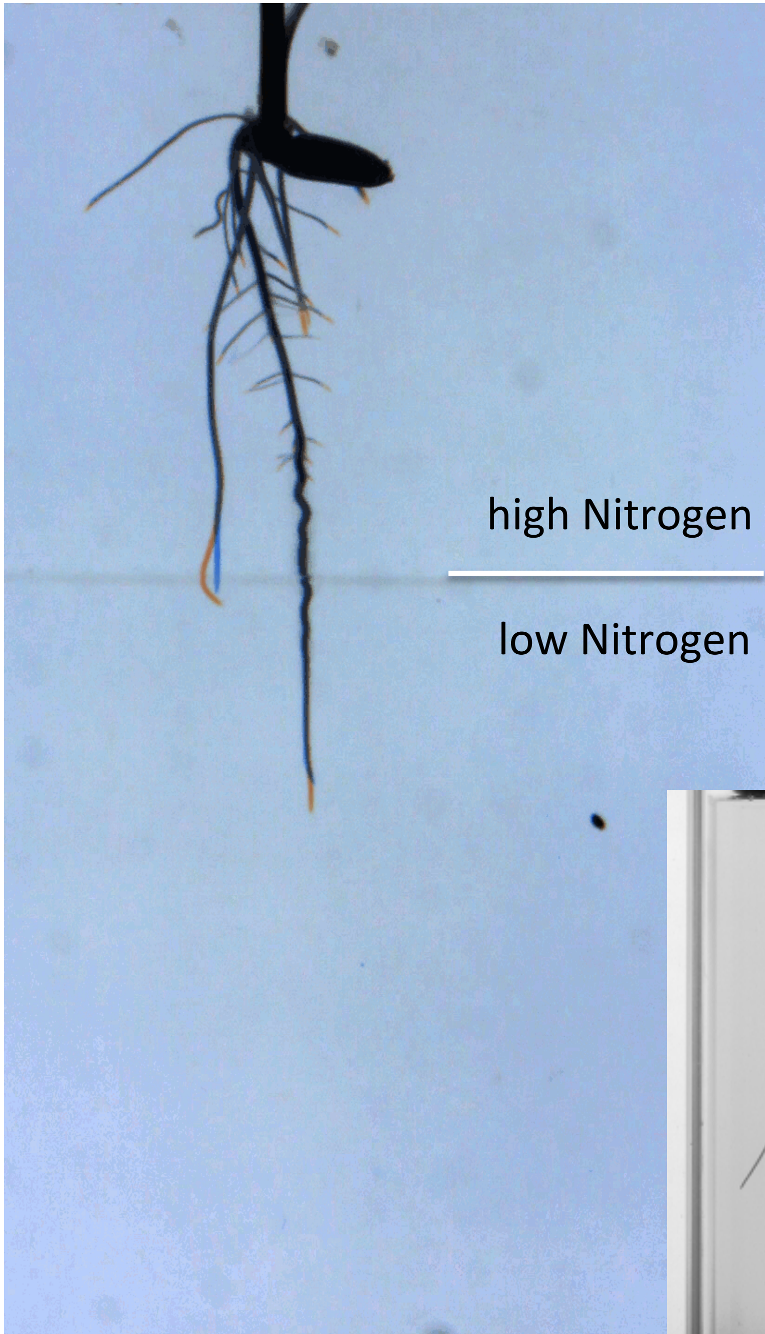
3D time series analysis analysis software:

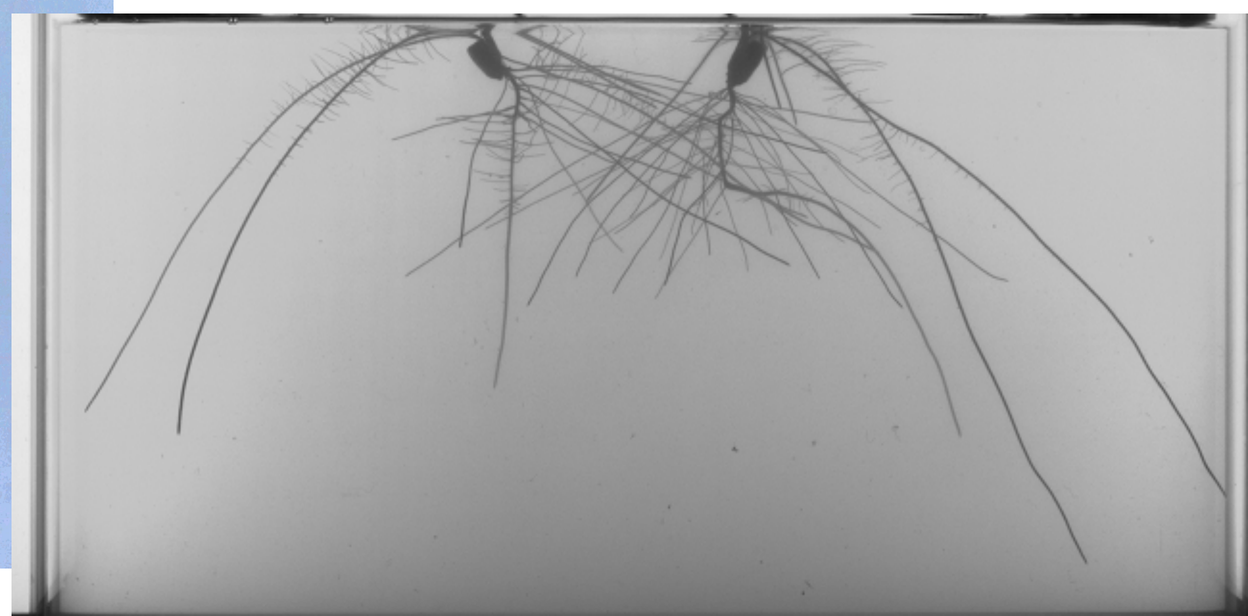
For each root at each time point:
length
width
Volume
root angle
root curvature
etc.

Hebert Edelsbrunner and
Olga Symonova

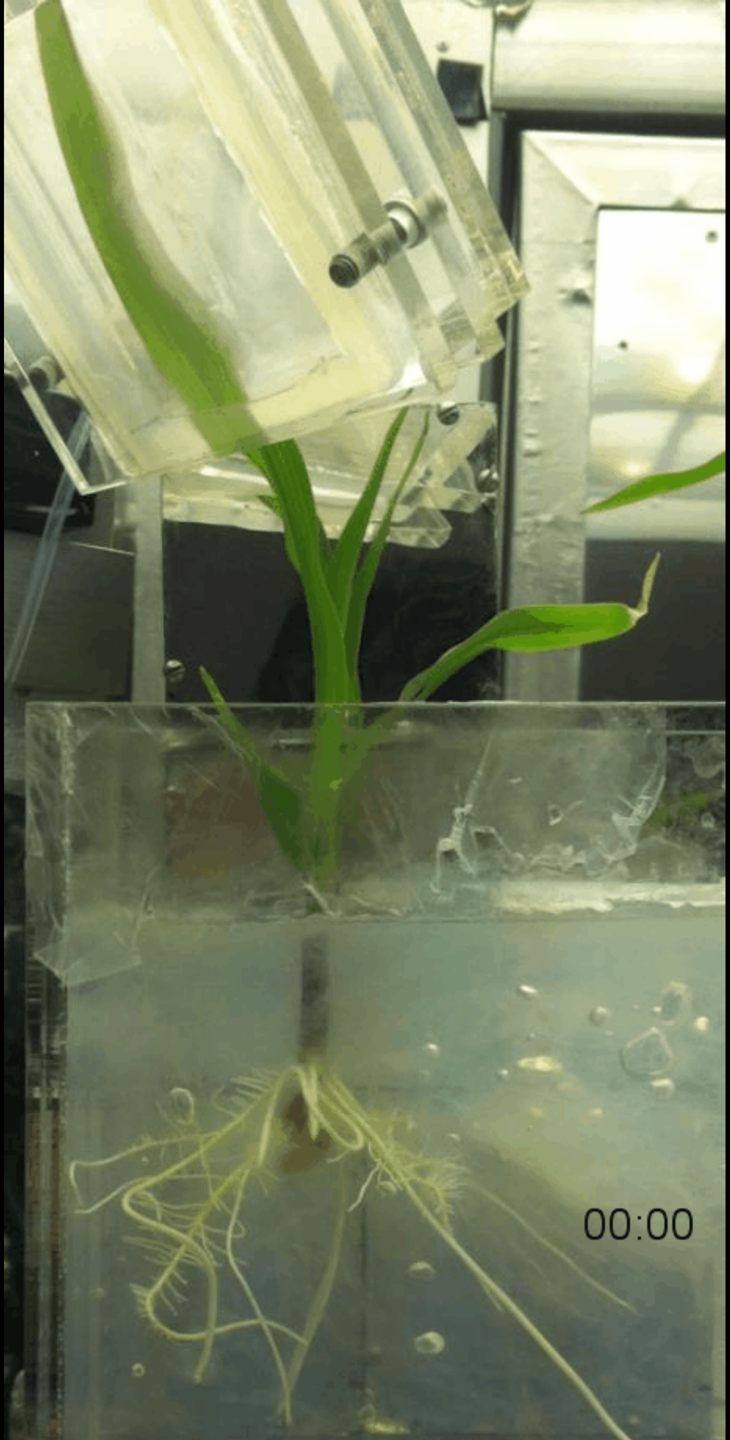
bi-hourly data set
25 reconstructions

How do roots respond to environmental challenge?





Positron Emission Tomography (PET): to image Carbon allocation and other dynamic physiological processes



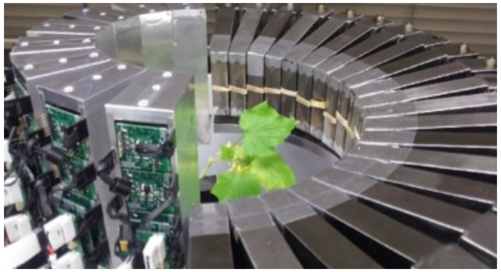
S. Lee, B. Kross, J. McKisson, J.E. McKisson, A.G. Weisenberger, W. Xi, C. Zorn, G. Bonito, C.R. Howell, C.D. Reid, A. Crowell, L. C. Cumberbatch, and M.F. Smith

PET imager integrated in a plant growth chamber

Plant PET System

Funded by a **NSF MRI Grant DBI-1040498**

A cucumber plant labeled with $^{11}CO_2$



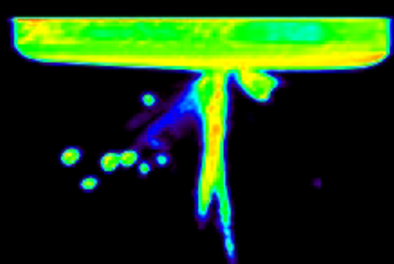
Yuan-Chuan Tai, Qiang Wang, Sergey Komarov,

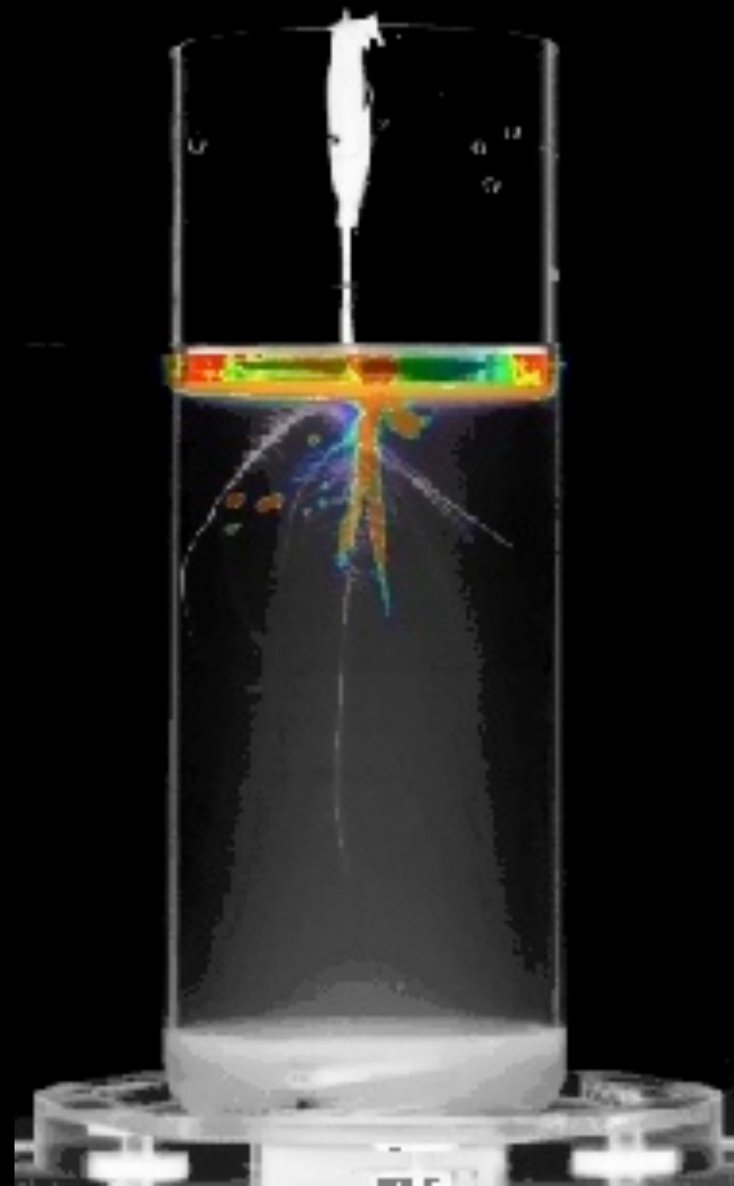
Aswin J Mathews, Ke Li, Jie Wen, Joseph A O'Sullivan

Washington University

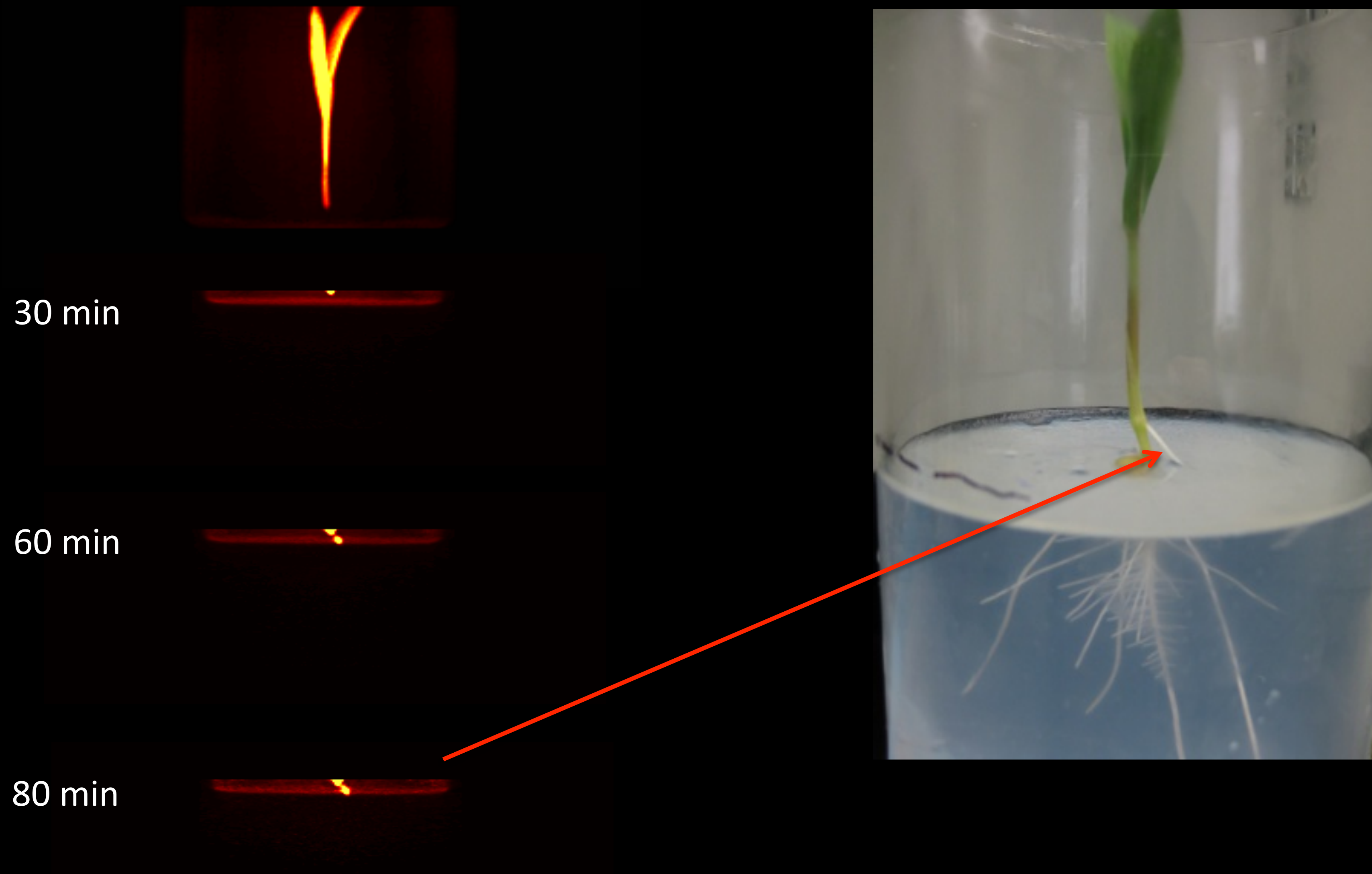
Department of Radiology

Department of Electrical and Systems Engineering





B73 – 3 dap

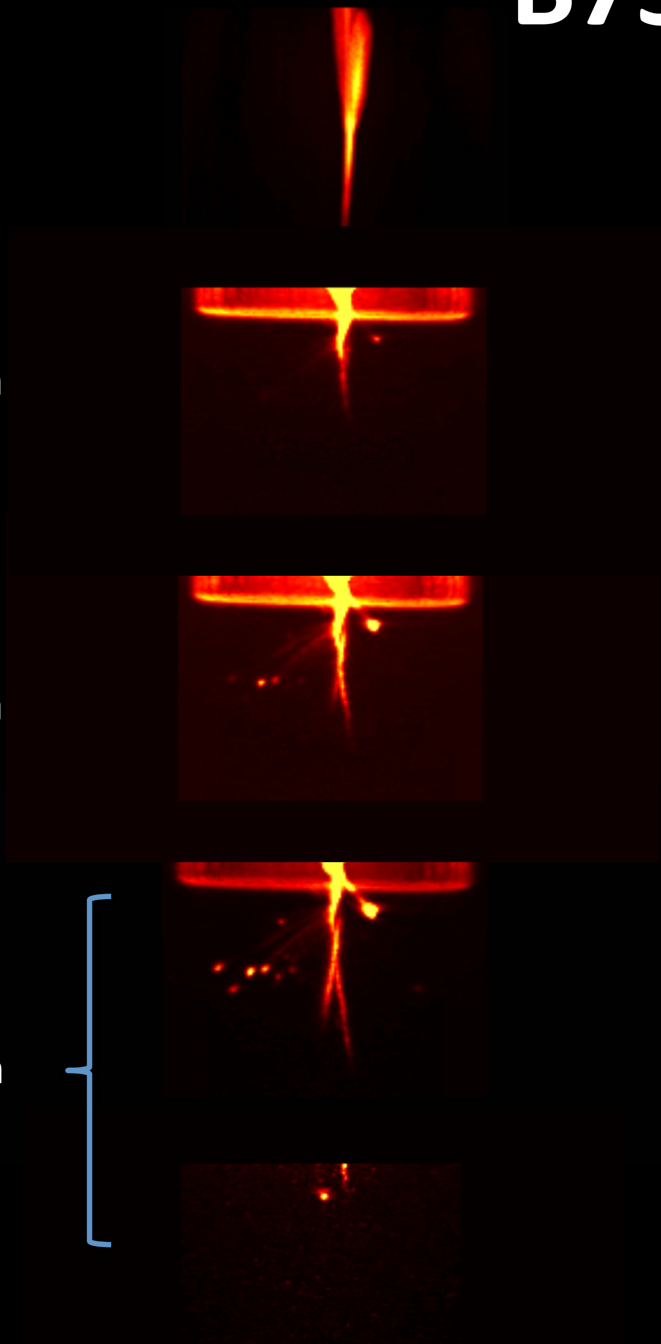


B73 – 4 dap

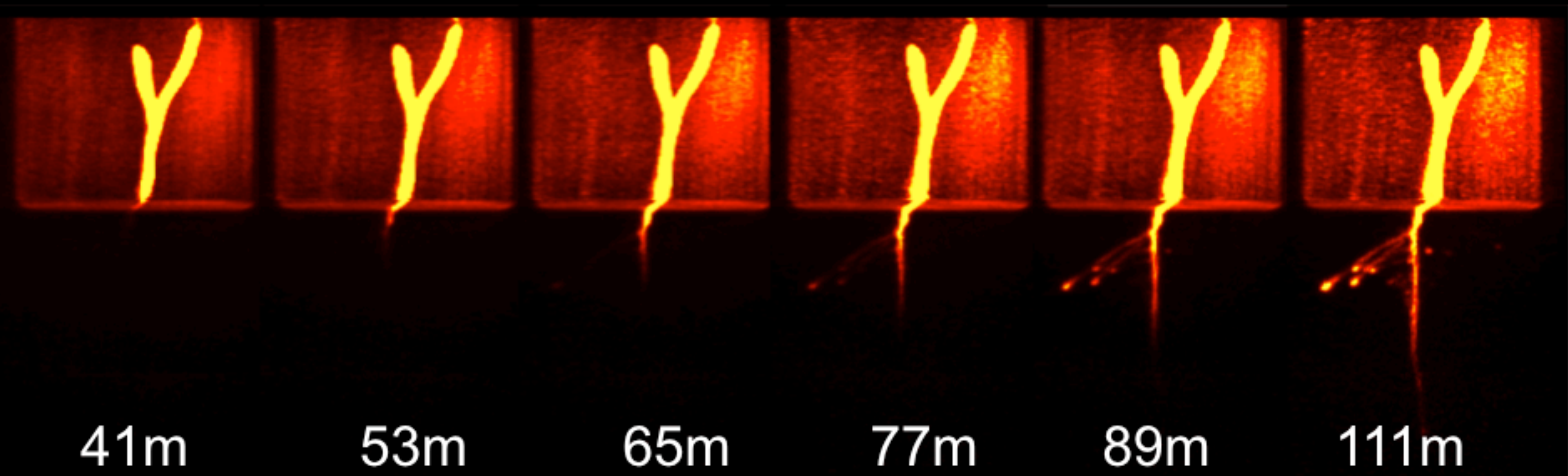
28 min

38 min

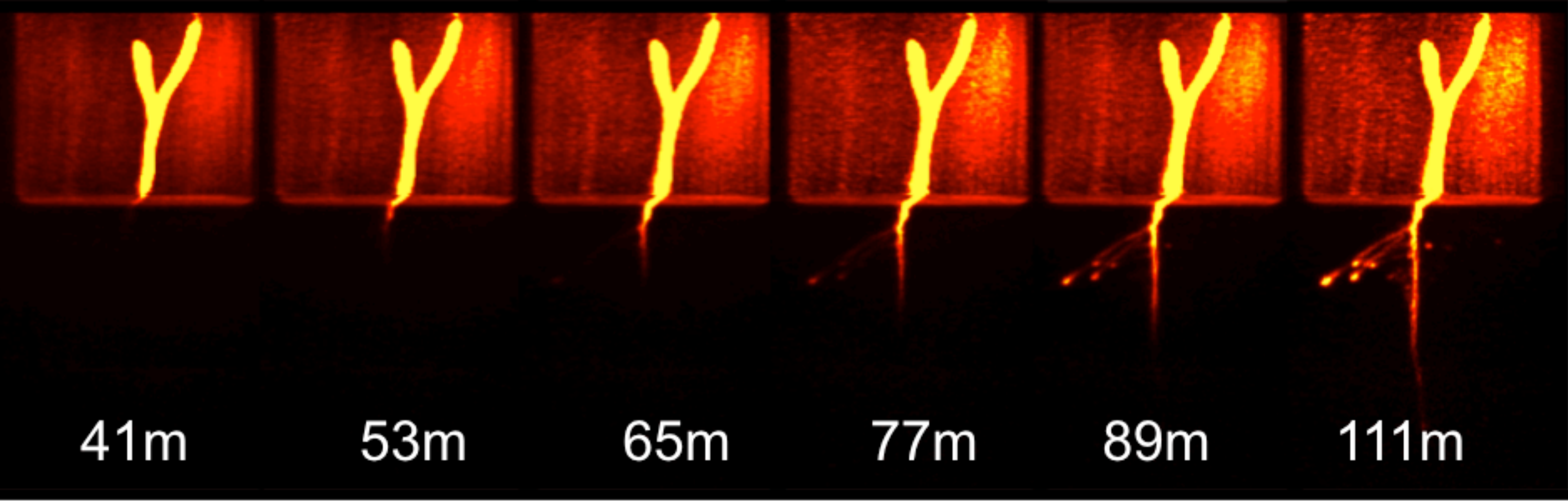
48 min

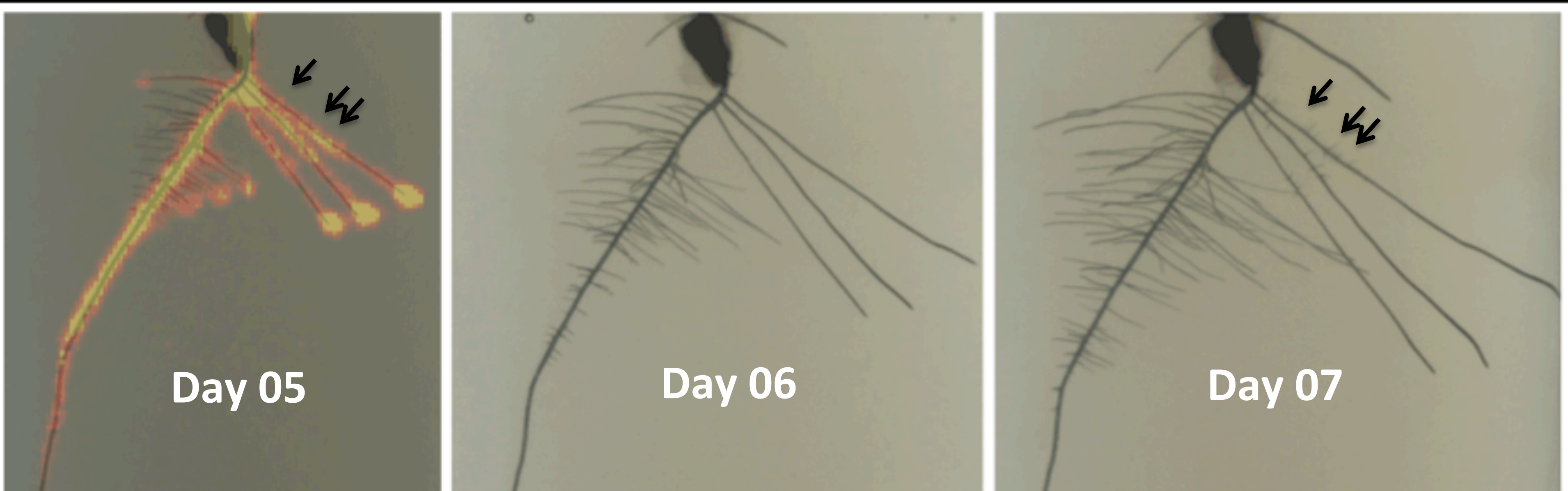


OPT-PET: physiological dynamics

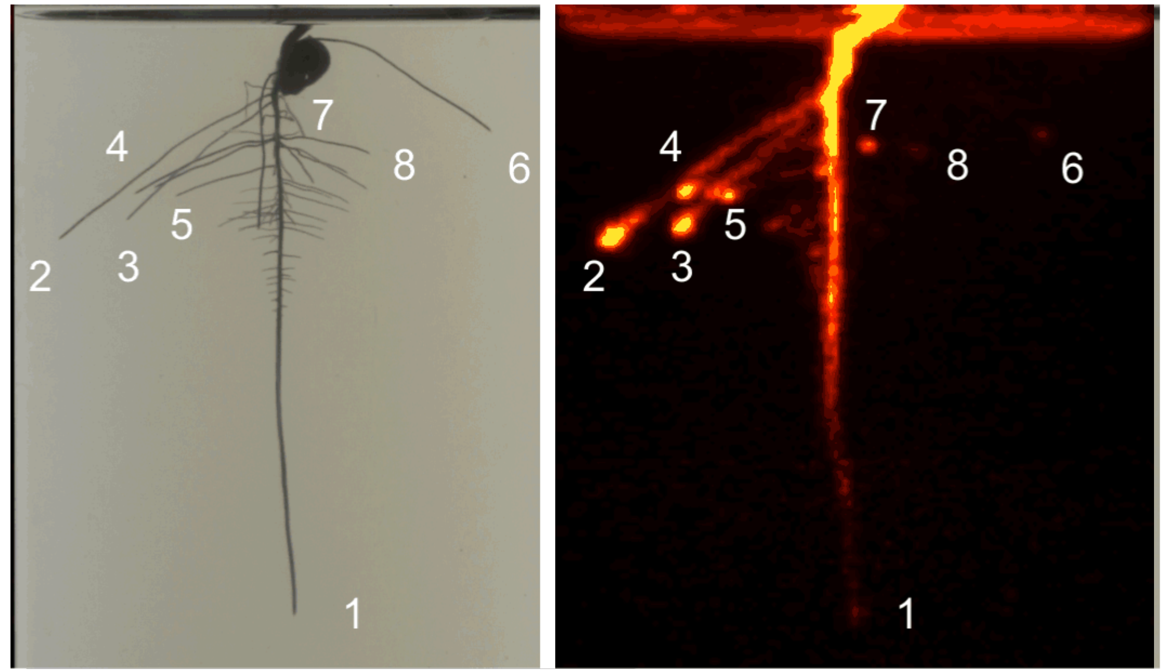


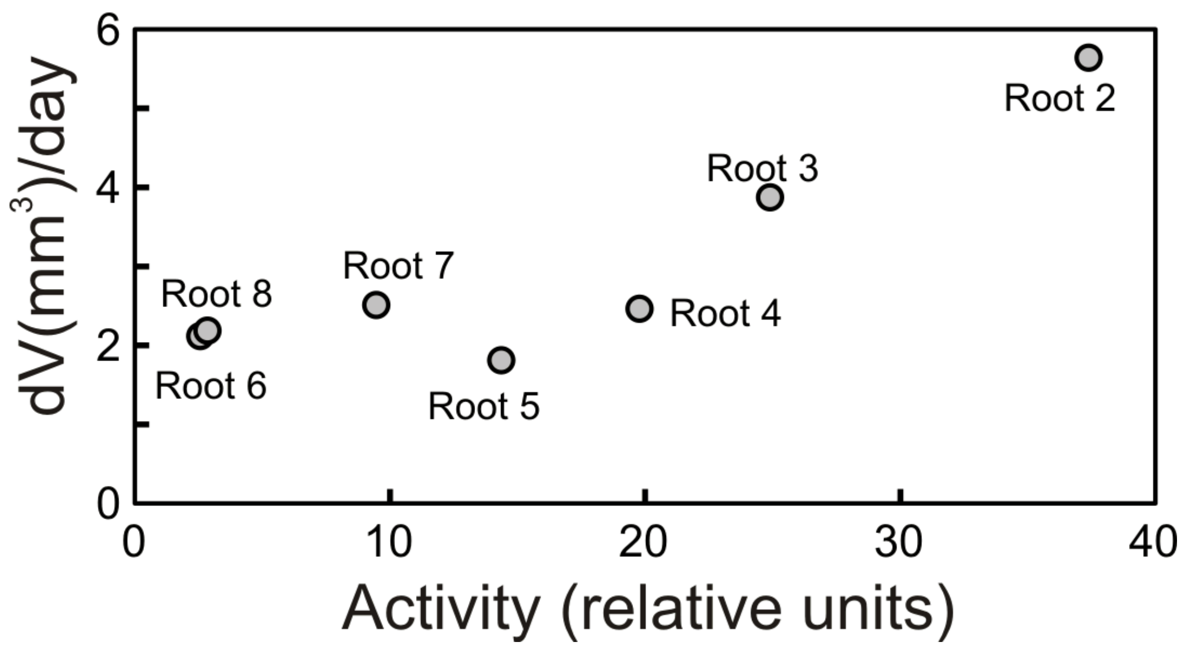
OPT-PET: morphological and physiological dynamics



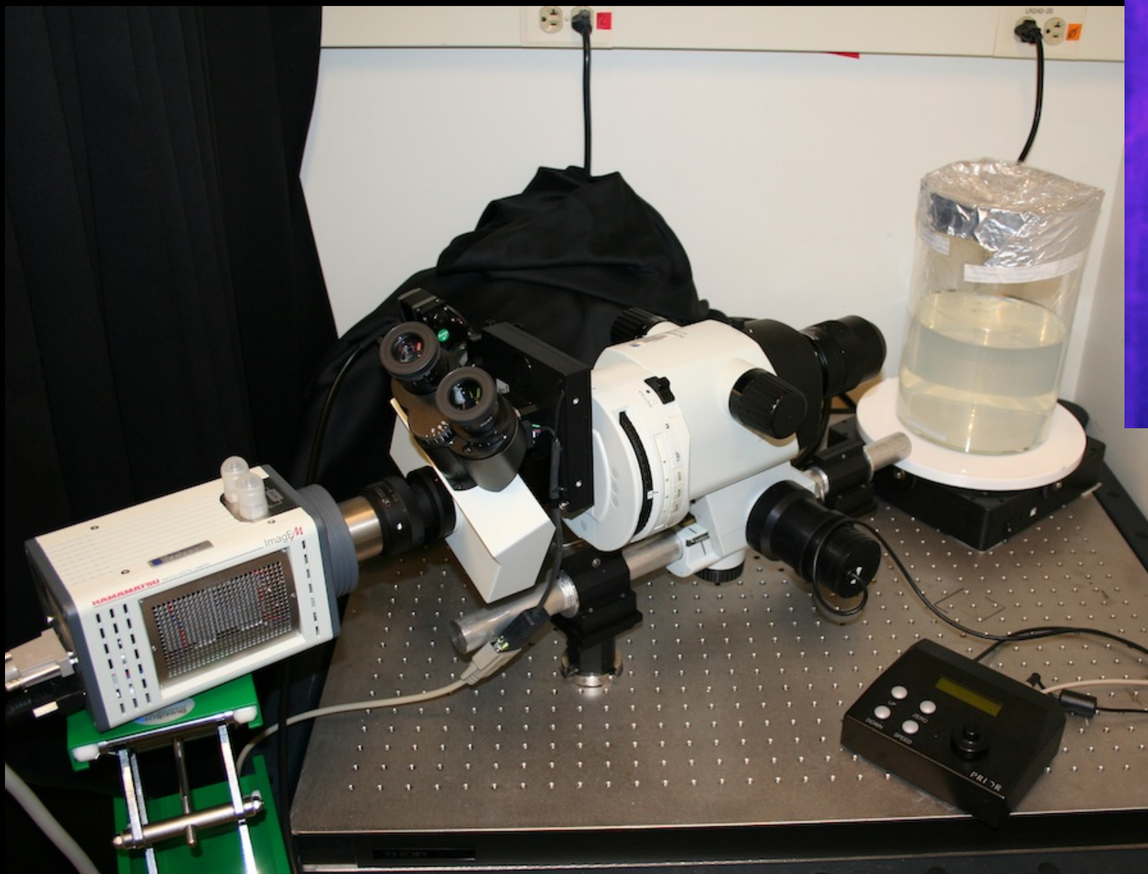


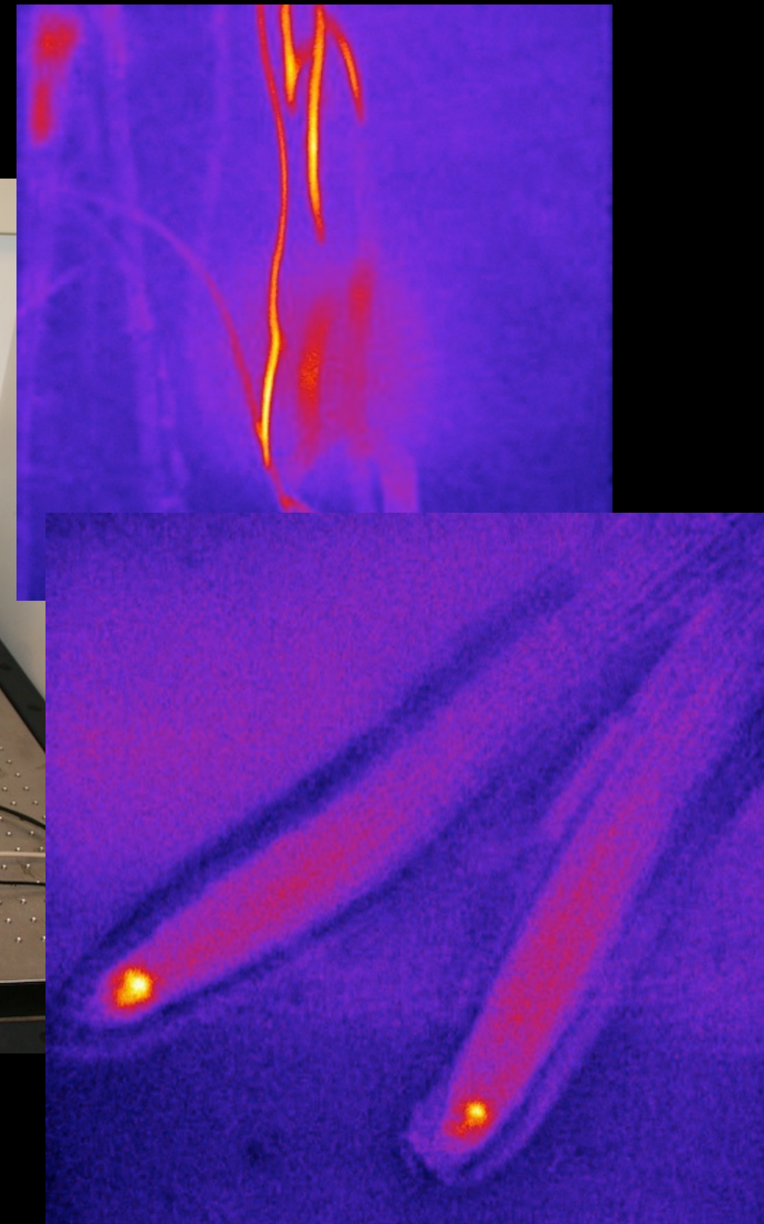
OPT-PET can be used to quantify baseline levels of carbon allocation: growth





Opportunity – molecular imaging over a large field of view





Bottlenecks and opportunities:

Plant phenomics is relatively nascent;
we lack expertise in tool development, data processing and analysis

Groups with the relevant expertise:

medical and industrial imaging
(engineers, physicists, computer scientists)

1. embed plant phenotyping in medical schools
2. leverage production agriculture for science
3. technology moves fast - focus on open source tools

This will happen at some level on a case by case, grant by grant basis, but a concerted institutional effort is required for large sustained payoffs

Acknowledgements

Philip Benfey (Duke)
Anjali Iyer-Pascuzzi (Purdue)
Paul Zurek
Olivia, Medha, Mike, Preston
Herbert Edelsbrunner (IST)
Olga Symonova
Ying Zheng
Joshua Weitz (GA Tech)
Alex Bucksch
Thomas Mitchell-Olds (Duke)
Jill Anderson
Cheng-Ruei Lee
Yuan-Chuan Tai (Wash U)
Sergey Komarov
Qiang Wang
Aswin Matthews
Leon Kochian (Cornell)
Randy Clark (Pioneer)
Jon Shaff
Dan Goldman (GA Tech)
Daria Moanenkova

Greg Bonito (Duke)
Chantal Reid (Duke)
Calvin Howell (Duke)
Drew Weisenberger (J-Labs)
Seungjoon Lee
Steve Moose (Illinois-UC)
and lab
Tim Horn (NC State)
Jode Edwards (IA State, USDA)
Mark Anastasio (Wash U)
Trey Garcon

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USDA-NIFA-AFRI