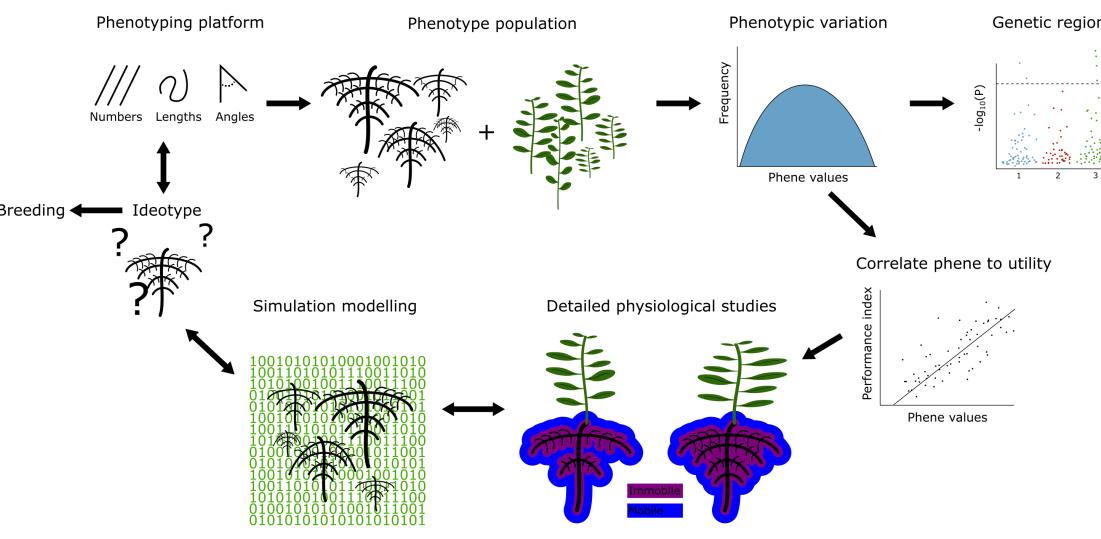
# OBLERESEARCH INSTITUTE

## **Root Functional Phenomics: Using High-Throughput** Phenotyping and Physiological Experimentation to **Understand Soil Resource Acquisition**

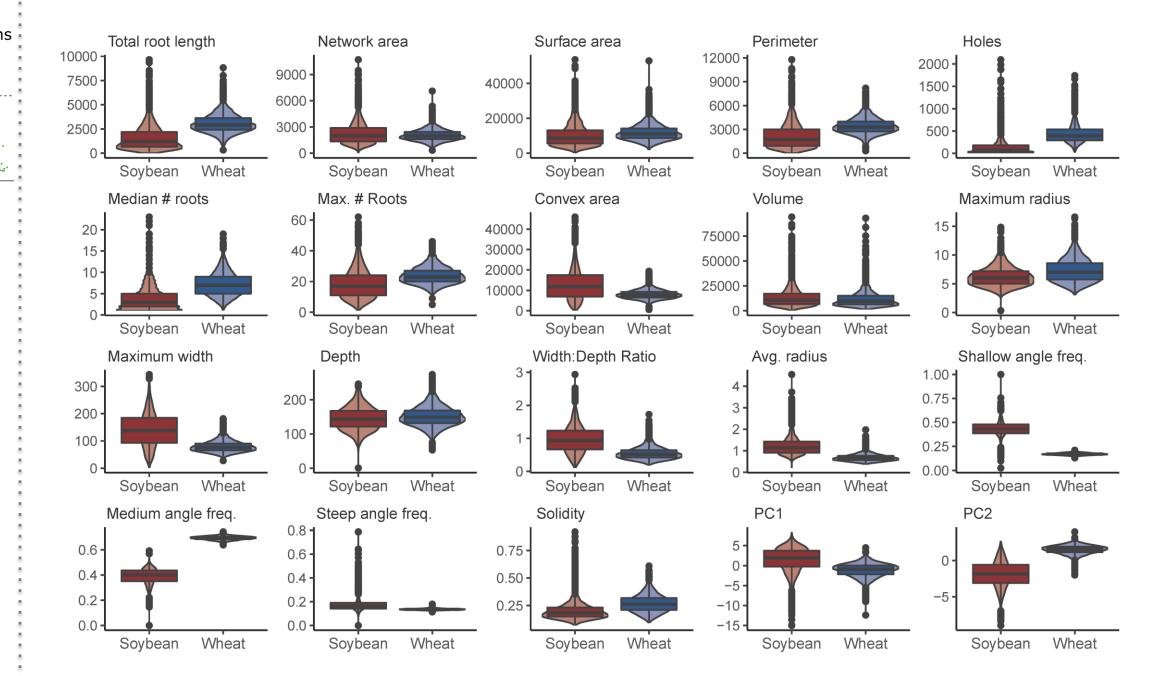
Larry M. York, Haichao Guo, Anand Seethepalli, Chakradhar Mattupalli, and Carolyn A. Young

## Functional phenomics: An emerging field integrating phenotyping, physiology, and bioinformatics



Roots are the interface of plants with the soil, yet little is known about which root phenes, or elemental units of phenotype, have the greatest impact on soil resource acquisition. Root functional phenomics addresses this knowledge gap by using a pipeline consisting of: ideotype development, phenotyping platform design, phenotyping populations, genetic analysis, statistical tests of root relations to crop performance, and the use of targeted physiology experiments and simulation modelling to validate root phene utility.

### Root system architectural differences between soybean and wheat

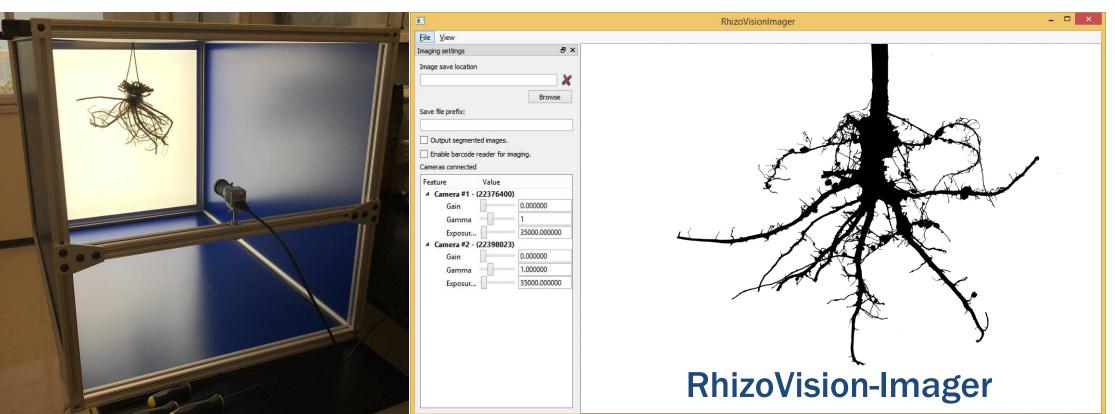


### Manipulating nodal root number by excision increases root length, shoot mass, and nitrogen uptake

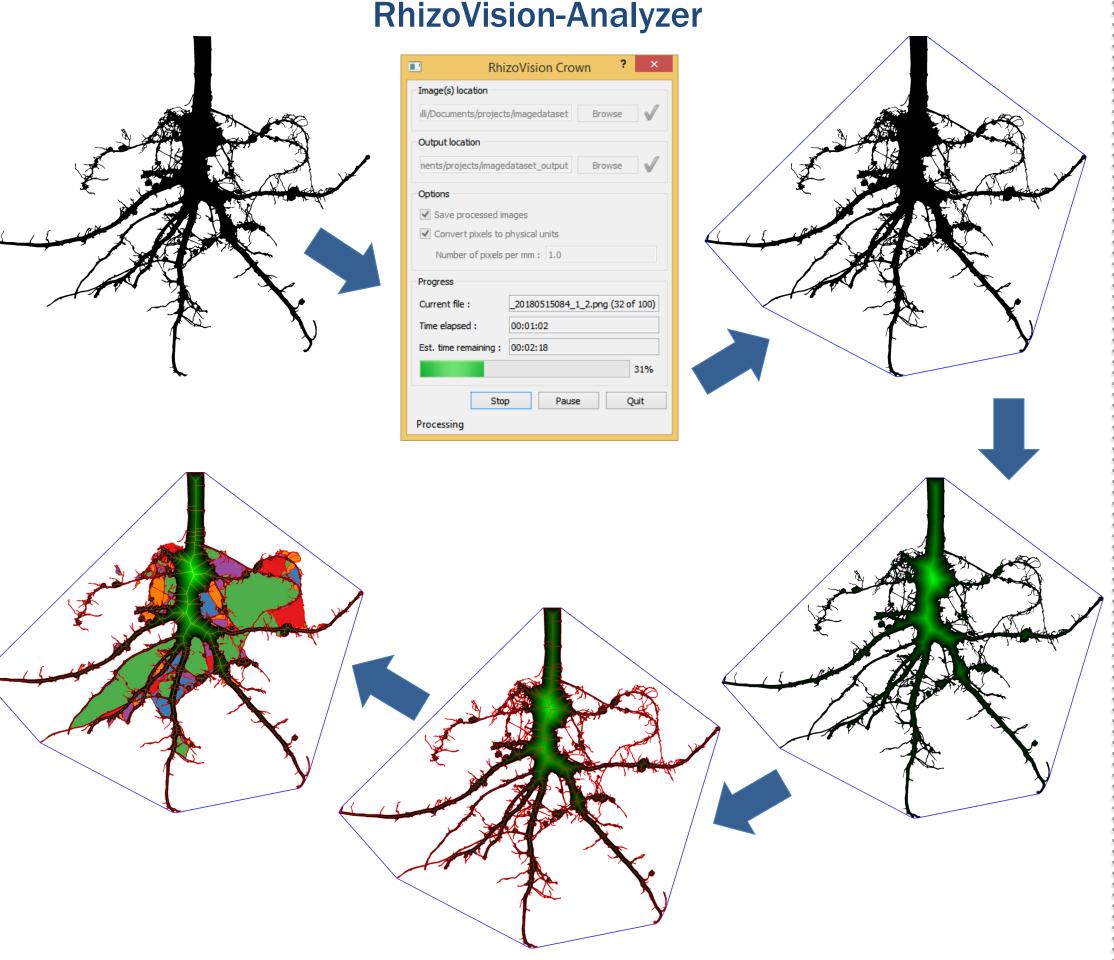


Maize B73 was planted in 150 cm tall mesocosms with 15 cm internal diameters. A plastic sleeve was inserted into each mesocosm then filled with a growth media consisting of sand, vermiculite, and perlite. Hoagland's solution with either high nitrogen (HN) or low nitrogen (LN) levels was added to the respective mesocosms, and were used to fertigate the same mesocosms throughout the experiment. Nodal roots were excised as they emerged over time to reach target cutting levels of 0%, 33%, and 67% of roots removed. At 42 days of growth, shoots were harvested, the sleeve removed, the roots were washed clean, then processed for scanning. This research was performed by Dr. Haichao Guo (pictured above, contact: hguo@noble.org).

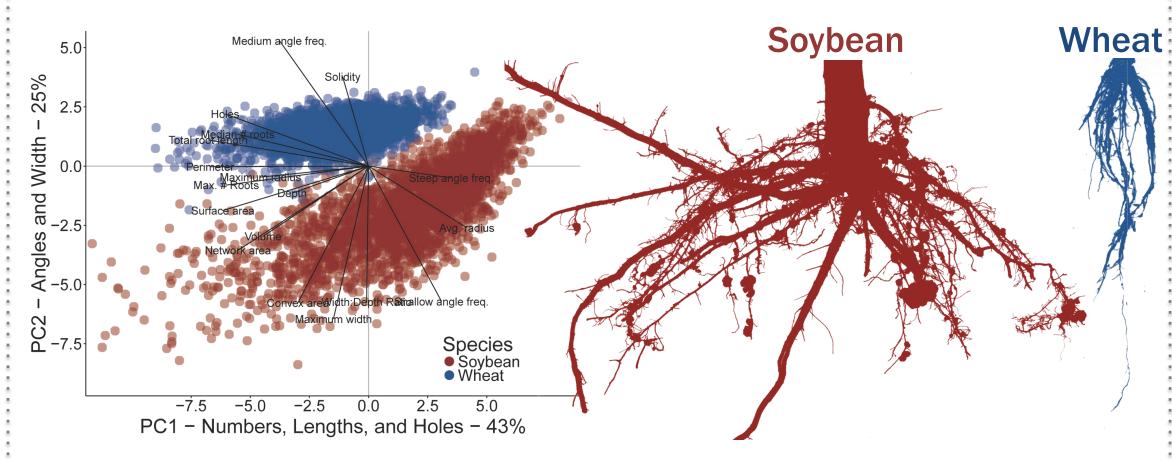
### RhizoVision-Crown: An integrated hardware and software phenotyping platform for root crowns



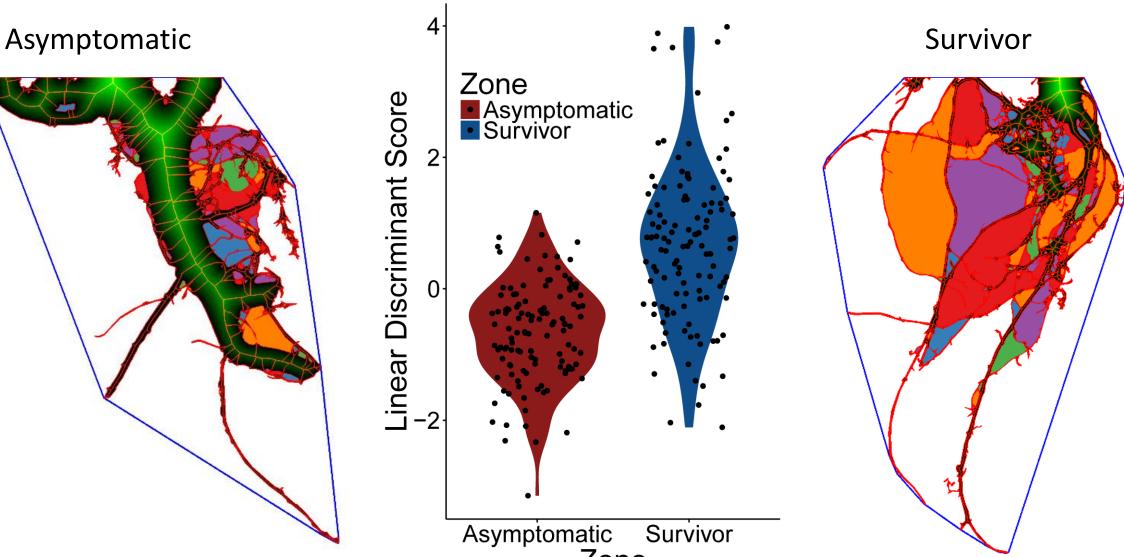
The RhizoVision-Crown phenotyping platform is constituted by both custom hardware and software solutions. The hardware platform consists of a 60 cm x 60 cm LED ceiling light panel, a root holder, a monochrome CMOS machine vision camera, and a barcode scanner. In total, these technologies allow fast and precise root crown phenotyping and can be employed in the field using a battery or in the lab. The raw images from the camera are quasi-segmented and only need simple thresholding for binary segmentation. The RhizoVision-Imager software allows changing camera settings, choosing the save location, live view, and the choice to use a barcode scanner for triggering image acquisition and storing filenames with the sample ID.

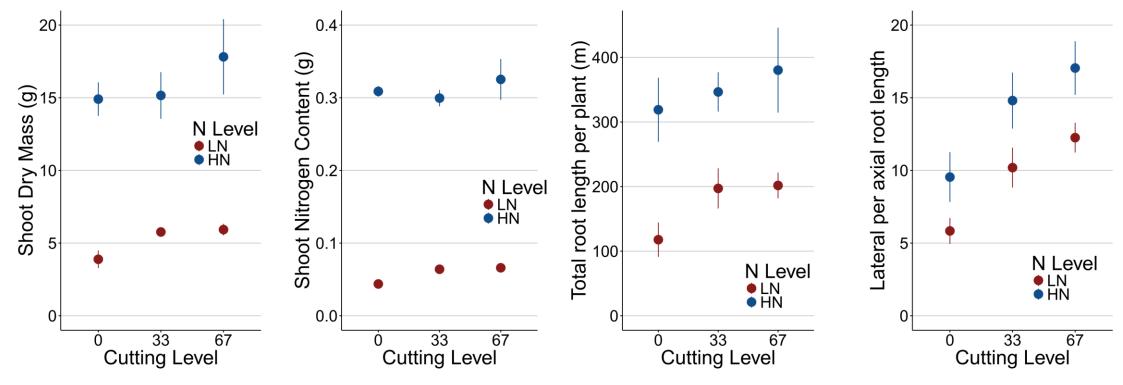


Soybean and wheat root crowns were excavated from field plots of mapping populations in Missouri and Oklahoma, respectively. Collaborators include Felix Fritschi and Xuefeng Ma.

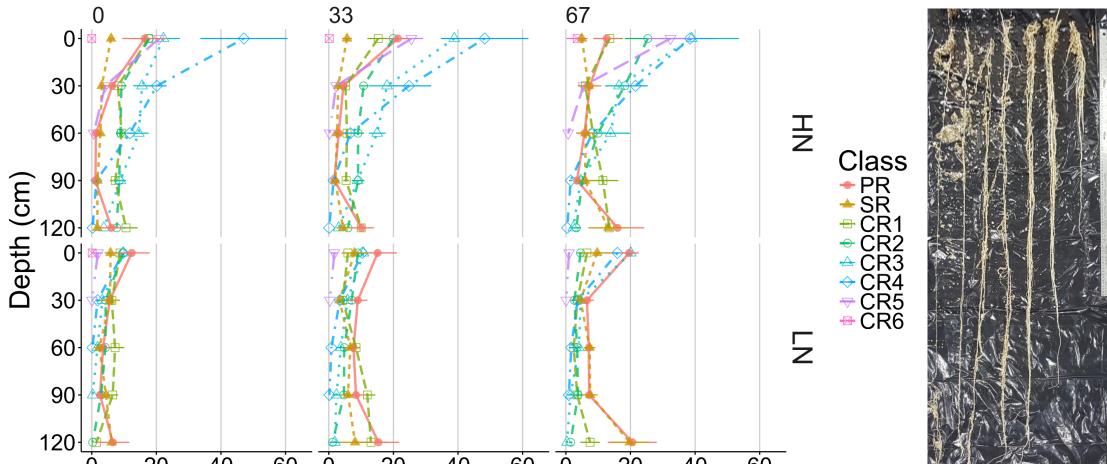


Root crown architectural changes due to surviving a root rot disease include compensatory growth of laterals





In low nitrogen mesocosms, decreasing the nodal root number led to greater shoot mass, shoot nitrogen content, total root length per plant, and an increase of the lateral per axial root length, indicating compensation of root length through greater lateral root length and increased shoot investment.



The RhizoVision-Analyzer software accepts either quasi-segmented greyscale images or binary segmented images and performs a suite of image analytics in batch mode. The outputs are a CSV data file and the optional 'feature images.' Feature images depict the distance map, skeleton, perimeter, convex hull, and the holes (disconnected components). Other image features such as radius and angle profiles are included in the CSV file.

40 60 0 20 40 60 0 20 40 60 Root length (m) 20

After washing, the root system was separated by axial root class, including primary, seminal, and nodal root whorls from oldest to youngest (left to right in above photograph). Each root class was separated into 5 layers of 30 cm depth then scanned. In general, compensation of root growth after excising nodal roots was greater for the oldest parts of the root system, including primary, seminal, and first whorl nodal roots.

#### Manipulative experiment confirms simulation results

This study confirms that reduced nodal root number may be a beneficial breeding target with regards to resource acquisition, however the tradeoff with possible increases in lodging needs to be investigated. Simulation studies using SimRoot first indicated the likely role reduced nodal root number would have (right), and this study confirms the compensation of root growth via longer axial roots and much longer lateral roots predicted by SimRoot.

#### Acknowledgements

The Root Phenomics Laboratory is supported by the Noble Research Institute, The Samuel Roberts Noble Foundation, and the USDA.

Phymatotrichopsis omnivora is a soil-borne fungus causing Phymatotrichopsis Root Rot (PRR)

disease. The host range of P. omnivora includes several dicotyledonous plants including perennial forage crops such as alfalfa. PRR disease can limit the ability to establish profitable alfalfa stands in areas where this disease is prevalent, such as southern Oklahoma and Texas. The pathogen attacks alfalfa roots causing wilting and rapid death. The disease manifests as circular to irregular shaped areas in the field. Root crowns were sampled along transects in four separate disease rings including surviving plants from within the ring, and asymptomatic plants from without. Root crowns were imaged using the RhizoVision-Crown platform and linear discriminate analysis was used to optimally classify Asymptomatic vs Survivor root crowns based on the multivariate data.

Zone  Asymptomatic Survivor									
Total root 2600 2400 2200 2000	length •	Network	area	Lower roc 2900 2800 2700 2600 2500 2500 2400	ot area	Computat 1.64 1.60 1.56 1.52 1.52 1.48	ion •	Surfa 18000 17000 16000 15000 14000	ce area
Perimeter 3000- 2800- 2600- 00 2400- 00 2200	•	Holes 175- 150- 125- <b>ф</b>	¢	E 2400 Median # 5.50 5.25 5.00 €	roots •	1.48 Max. # Ro 18 17 16 15 ↓ ↓	oots •		ex area
C Volume C 40000 C 35000 S 30000	¢	Maximur 10.5- 10.0- 9.5- 9.0- ∳	n radius	Maximum 135 130 125 120 120 115	width •	Depth 160- 150- 140	¢	Width 0.90 0.88 0.86 0.84	:Depth Ratio
Avg. radiu 4 25000 Avg. radiu 4 1.4 4 1.3 1.2 1.1	is ¢	Fine root 0.64 0.62 0.60 0.58	t freq.	Medium ro 0.20 0.19 0.18 0.17 0.16	oot freq.	0.275 0.250 0.225 0.200 0.175	ot freq.	Shallo 0.1650 0.1625 0.1600 0.1575	ow angle freq.
Medium a 0.696 0.692 0.688 0.684	ngle freq.	0.152 0.150 0.148	ngle freq.	0.25 0.24 0.23 0.22 0.22 Z	one	PC1 0.4 0.0 -0.4 -0.4 -0.8	¢	0.6 0.3 0.0 -0.3 -0.6	•



Nodal Roots Nodal Roots

Zone