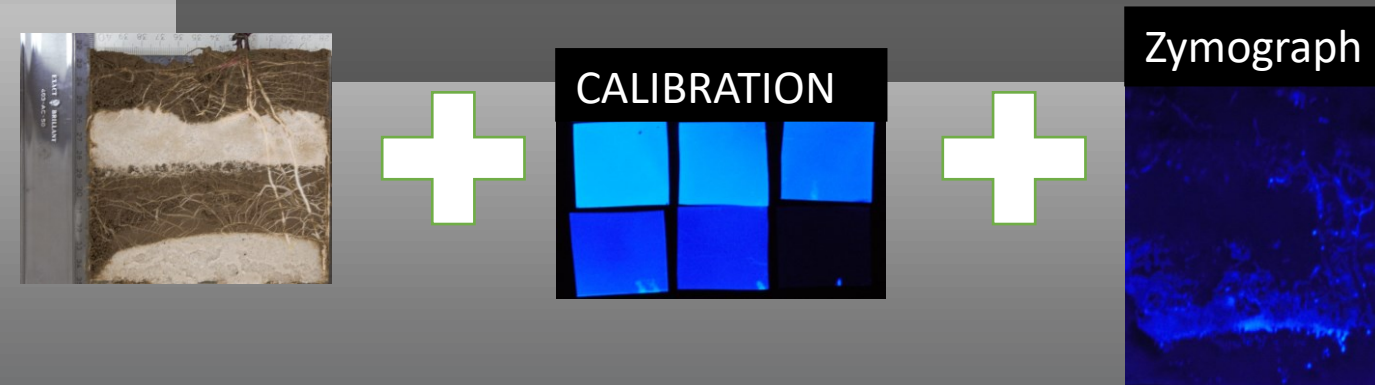


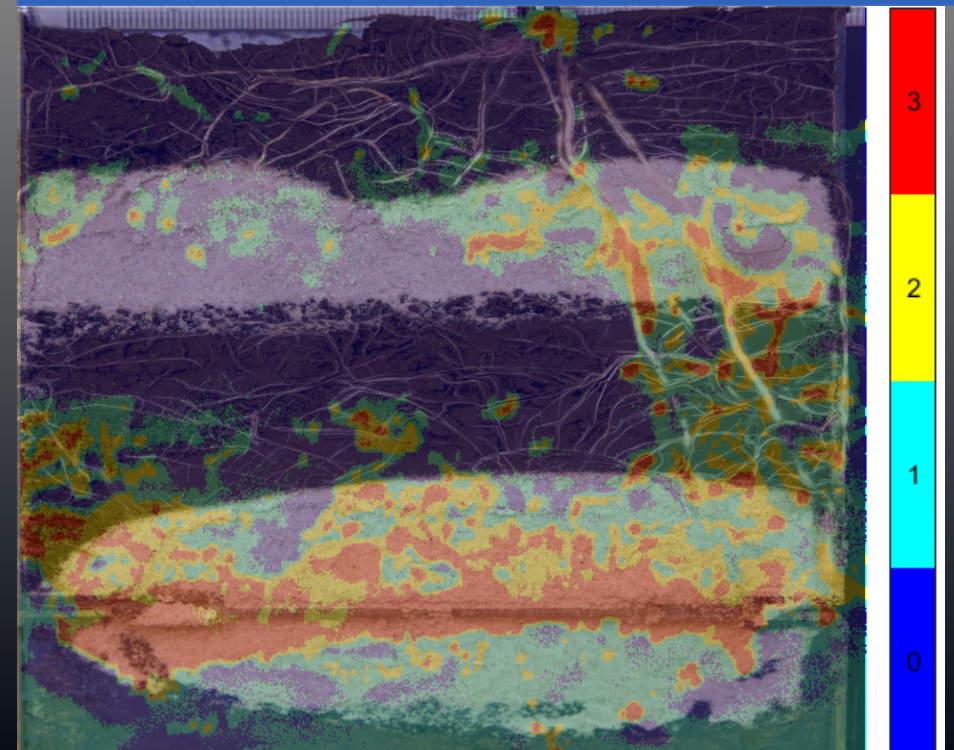
Examples
for soil zymography

Dr. Sebastian Loepmann
CAU Kiel

Image processing in soil zymography



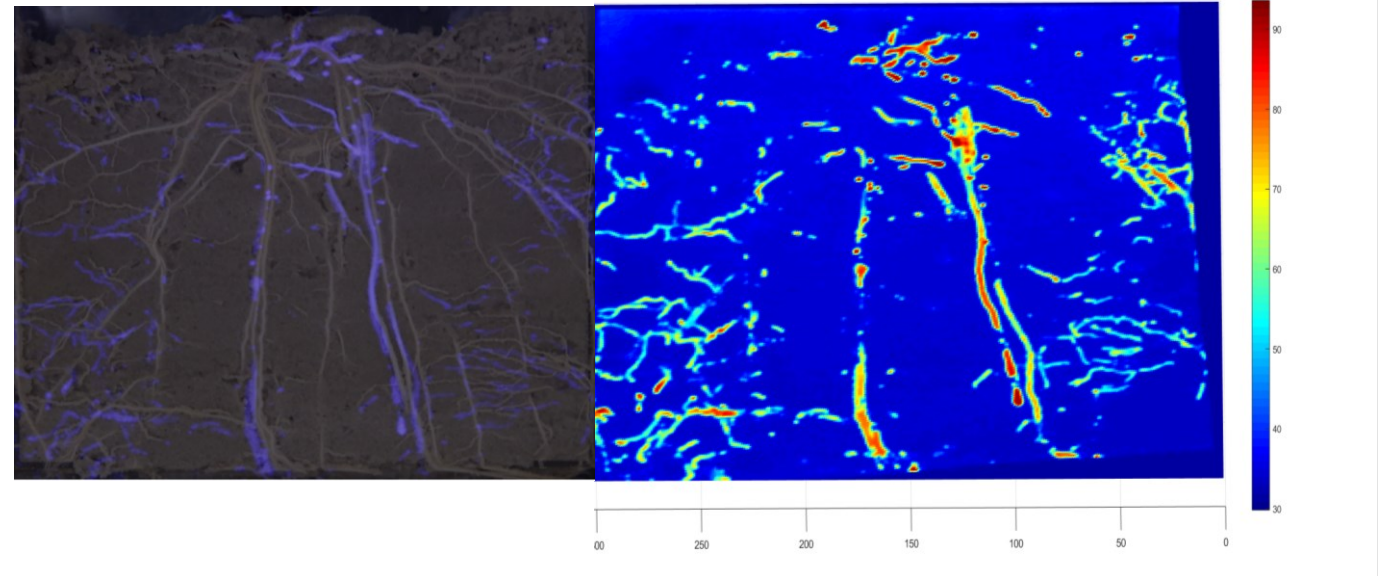
Standardized image clustering for hot spot detection



Aligned day-light and clustered image of the spatial distribution of enzyme activity in the rhizosphere of maize in a mix of layer derived from soil and sand.

ROOT-O-MAT

Zymograph of leucine aminopeptidase activity measured in a rhizobox filled with sand and planted with maize



aligned zymograph

calculated enzyme activity after proper calibration

All image processing steps in one application!
Fast and standardized workflow!
Basic data statistics and data export for further analysis

Day-light image



SOIL

SAND

zymograph image

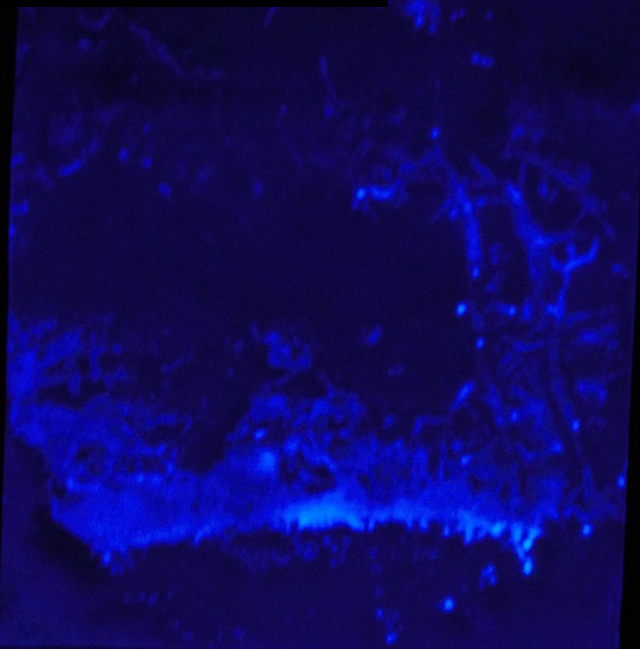
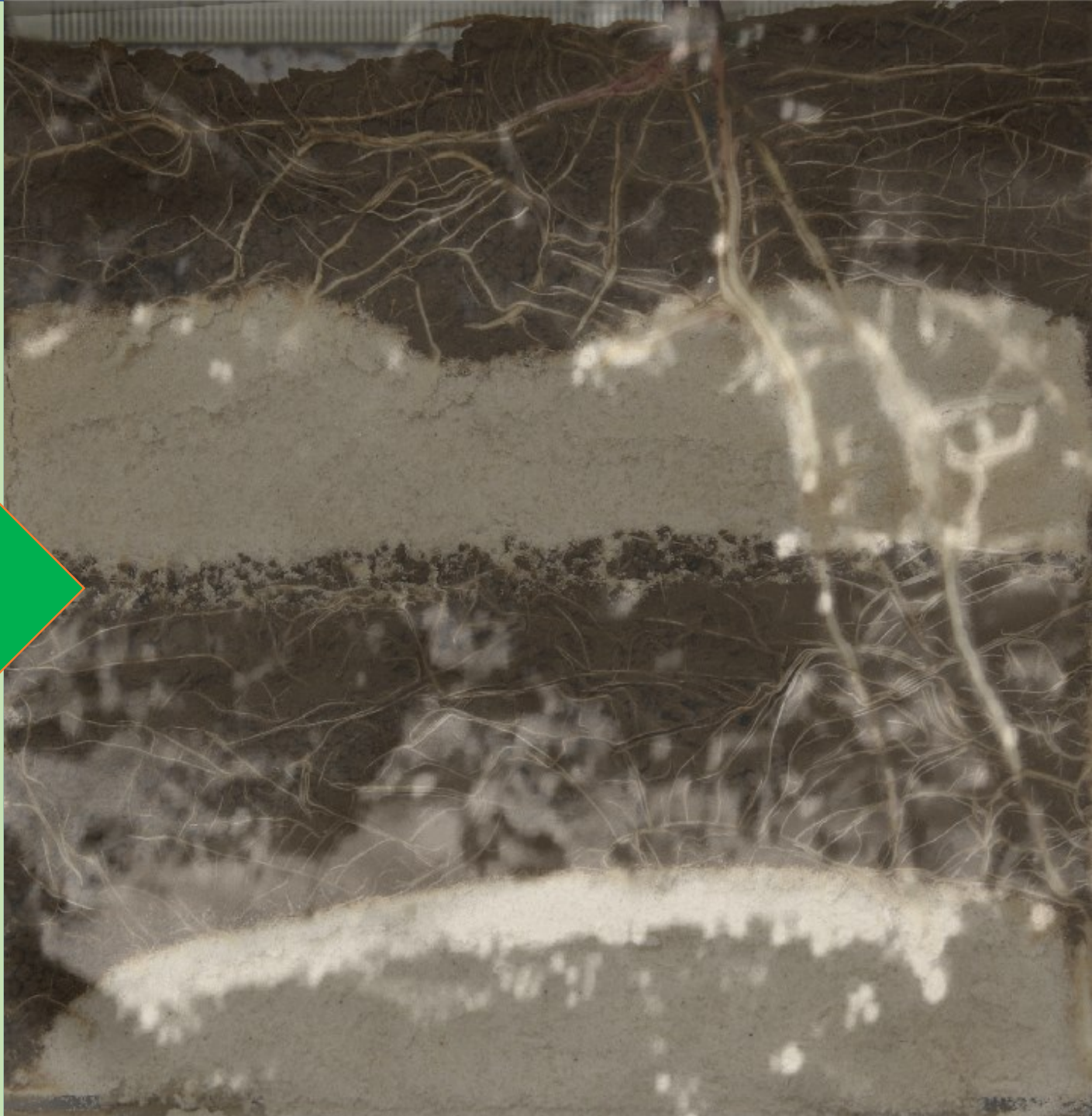
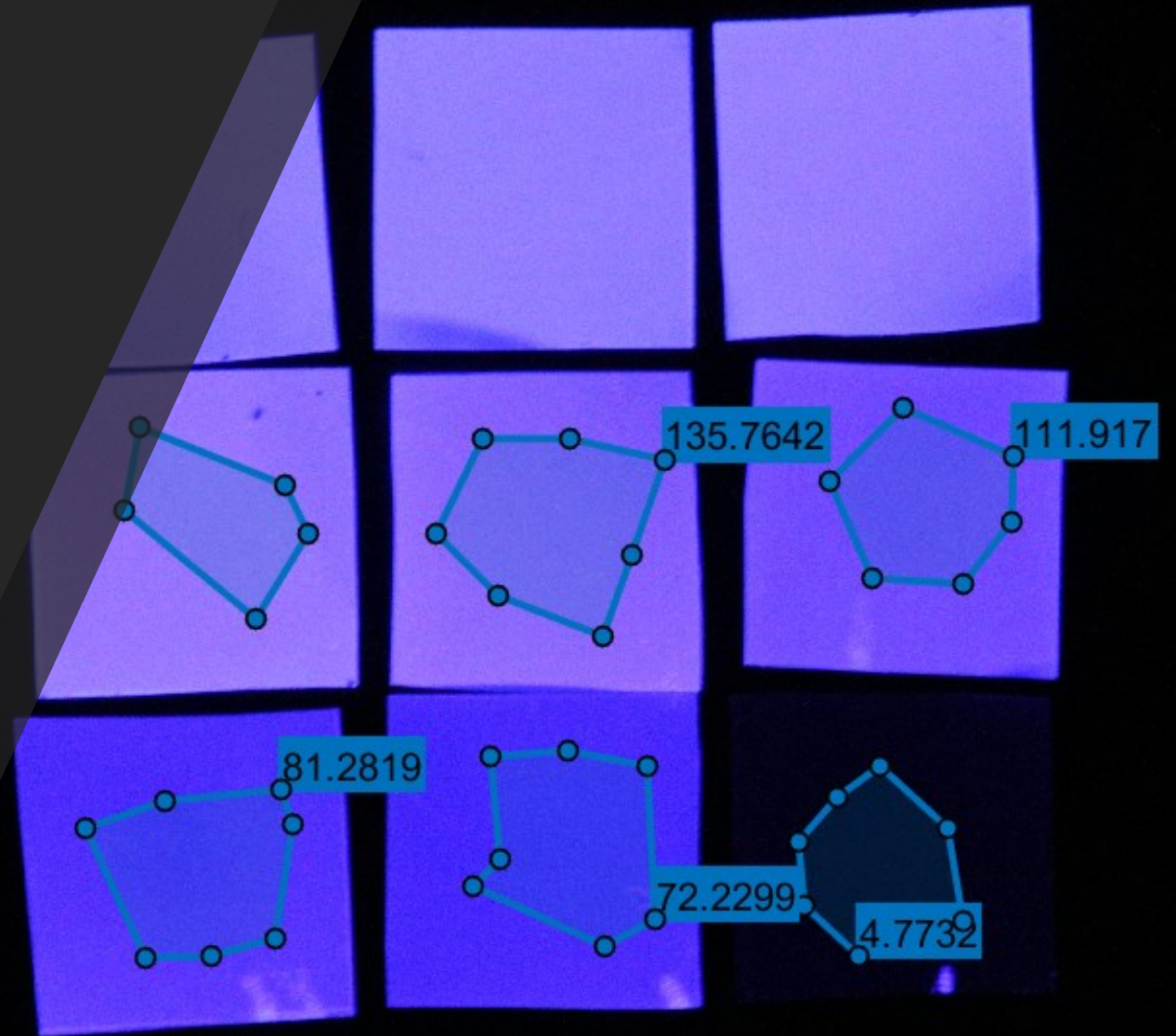


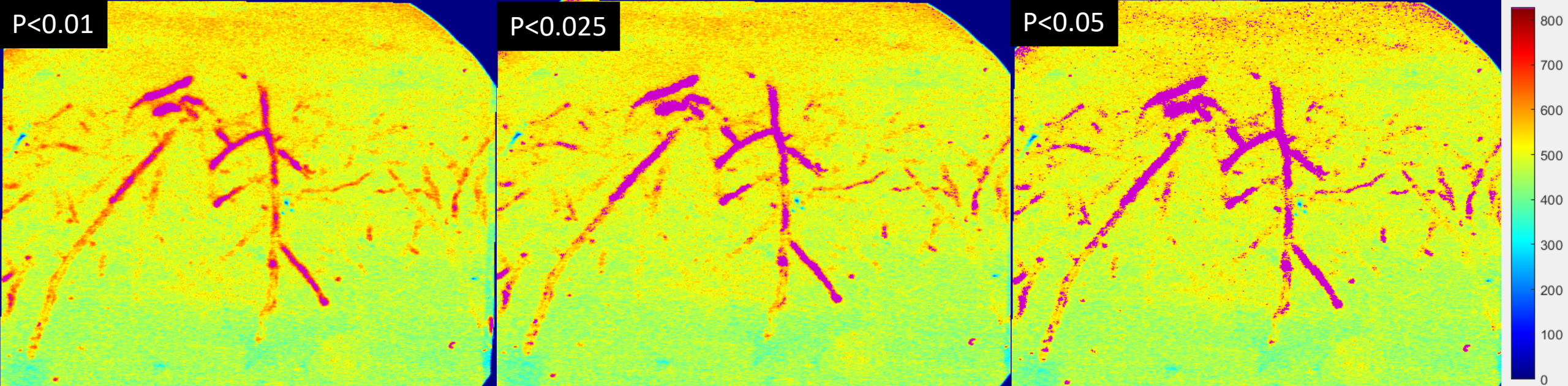
Image registration and
Conversion to grey
scale values



Root-o-Mat calibration of 2D Imaging techniques

- Simple AOI selection for calibration detection
- Manual entering of data into table
- Multiple calibrations are possible
- linear and non-linear calibration
- Background correction of images
- Basic fit statistics
- Export calibration curves





Manual Clustering

Mode

Hot Spots

Cold Spots

704.6

0.01 0.025

0.1 0.05

0.25 0.5

min 0 max 828

zsand

▶

ROOT-O-MAT – Hot and cold spot detection

#Image saturation biases your results

Automatic Clustering (Otsu Method)

Select Image

zsand

No. of Classes

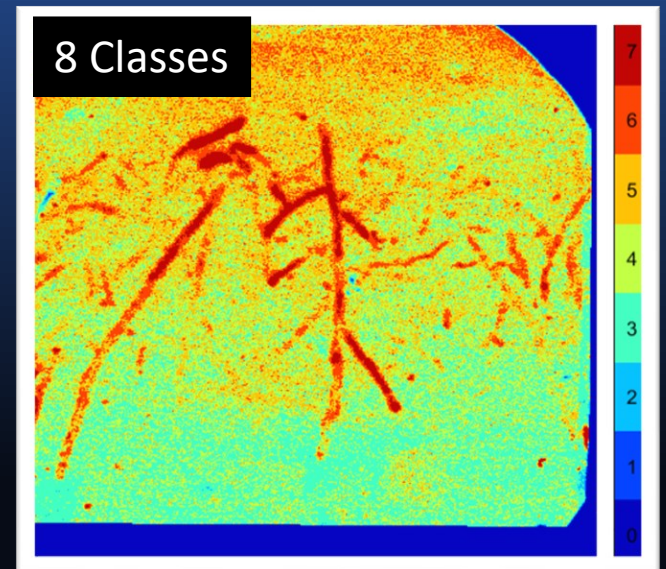
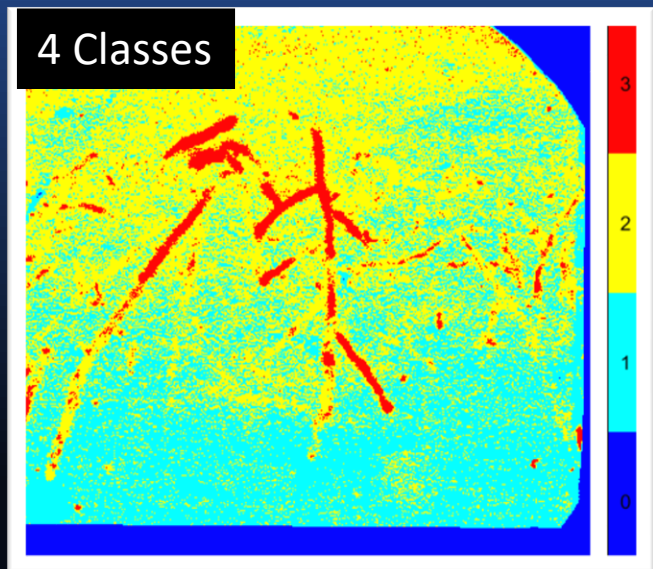
Select Image for Comparison

z sand

8

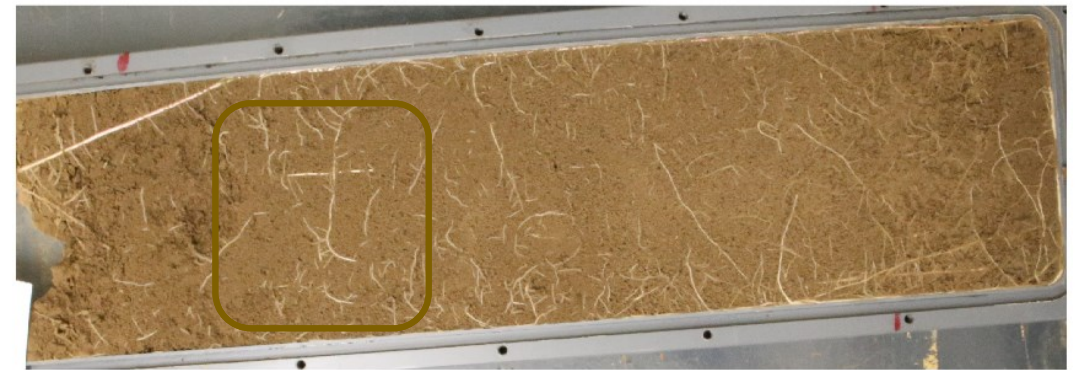
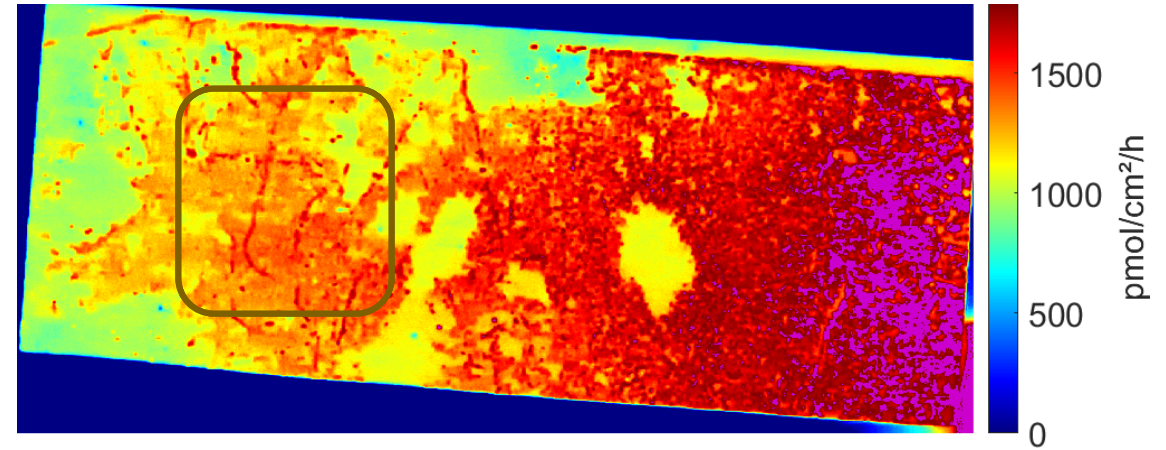
Compare

Statistics



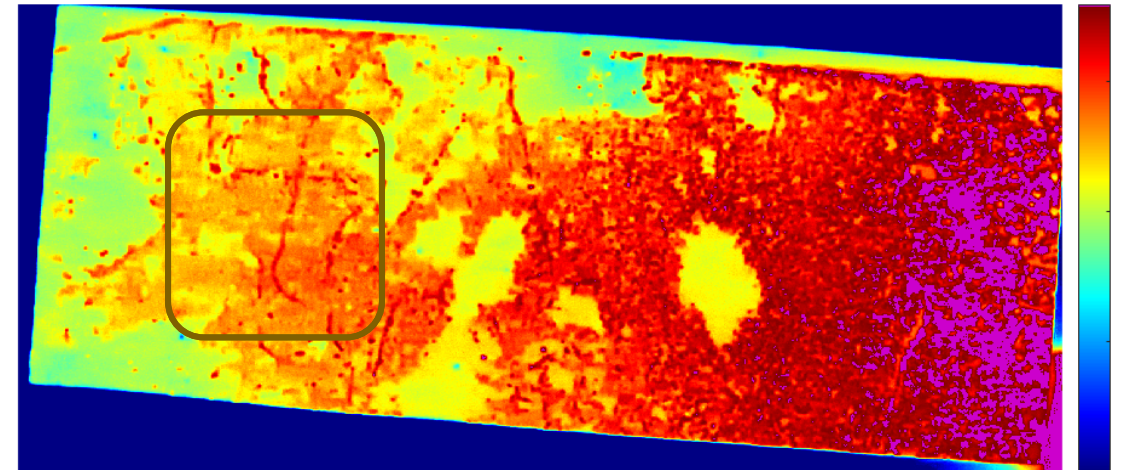
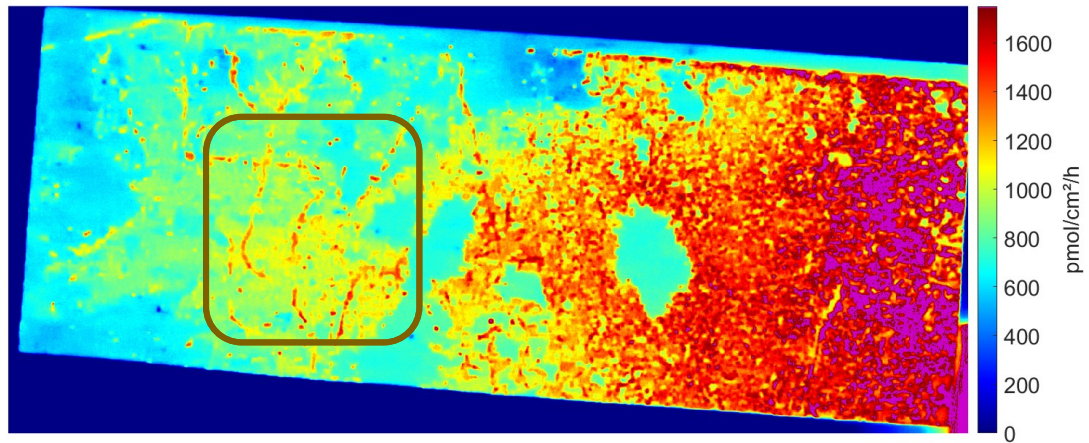
Acid phosphatase

The violet color depicts saturated pixels (max DN) in the resulting image of the calibrated zymographs.

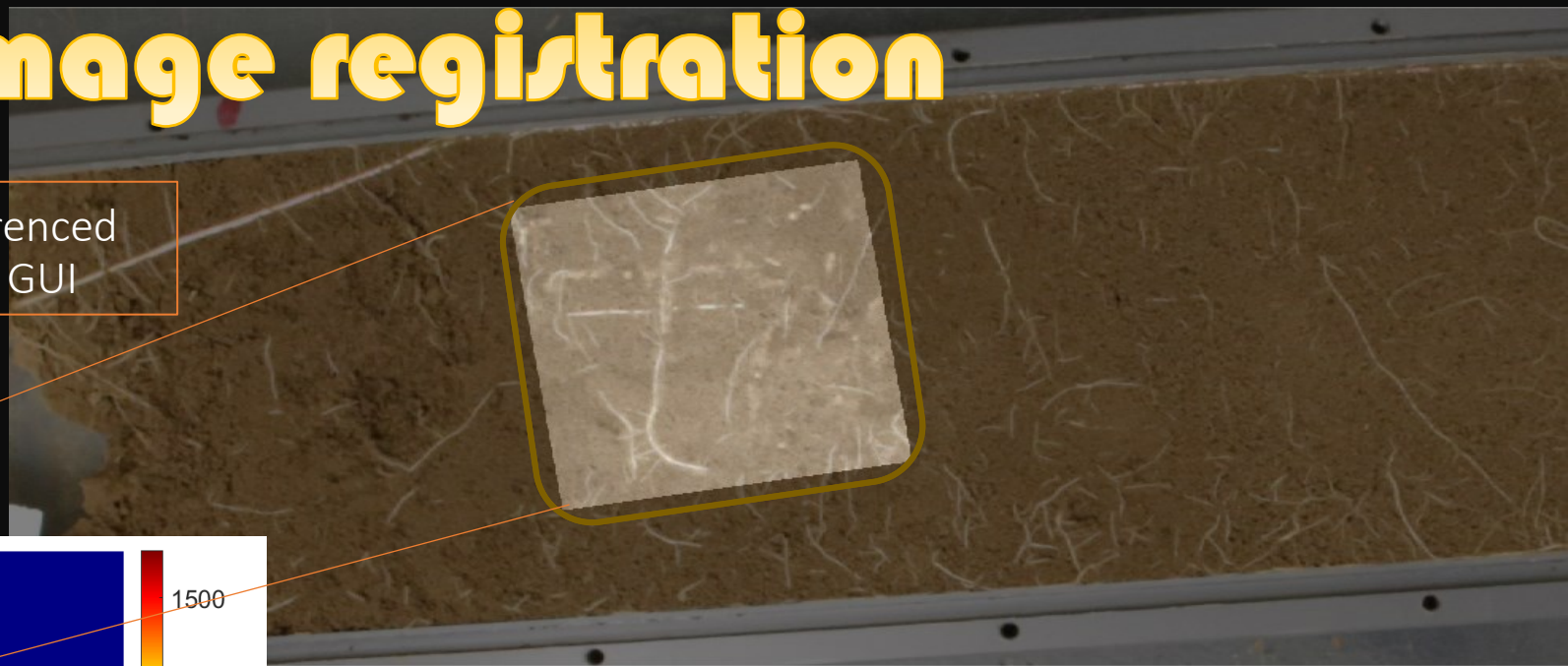


Acid phosphatase

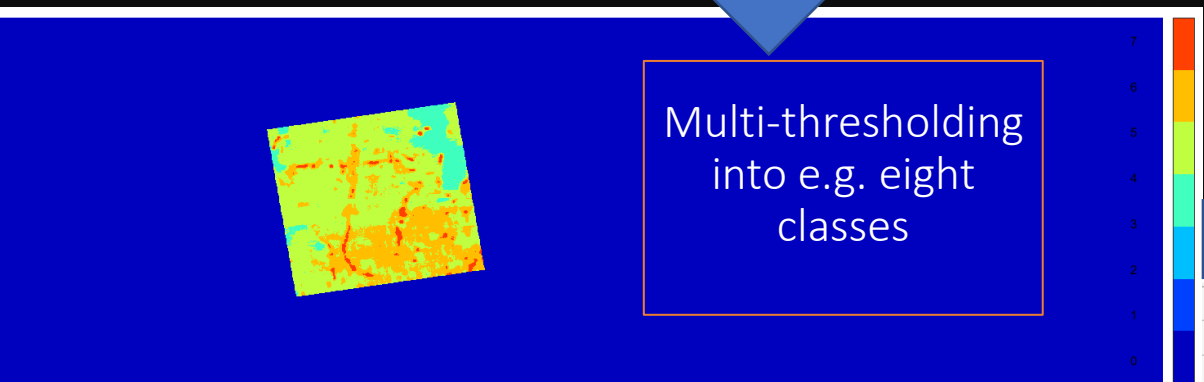
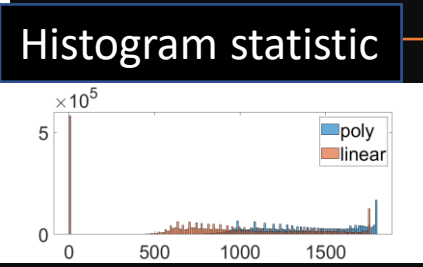
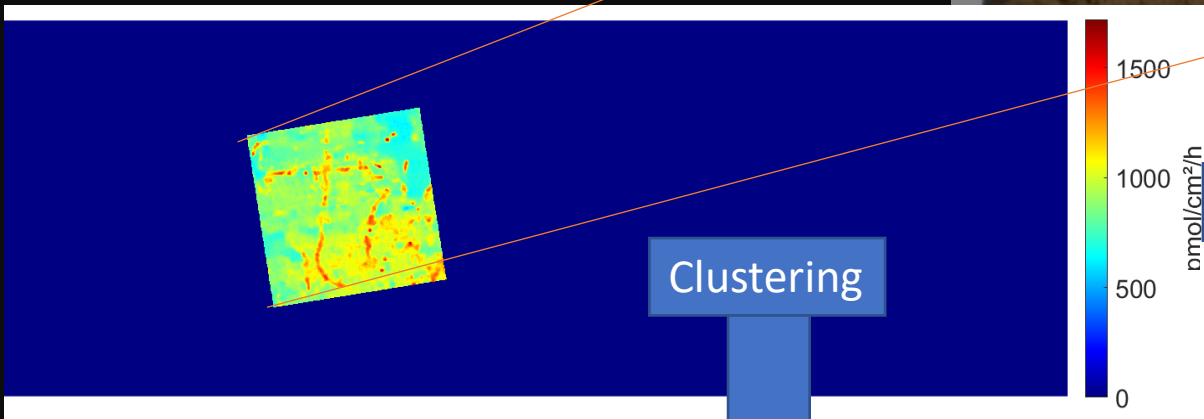
Comparison between linear and polynom fit (identically scaled). In the subsoil of the rhizotrone a higher moisture content compared to the topsoil may have induced higher enzyme activity.



ROOT-O-MAT – Image registration



AOI was converted to grey scale image and referenced to the day-light image via control point selection GUI



AOI statistic: linear vs. polynom fit

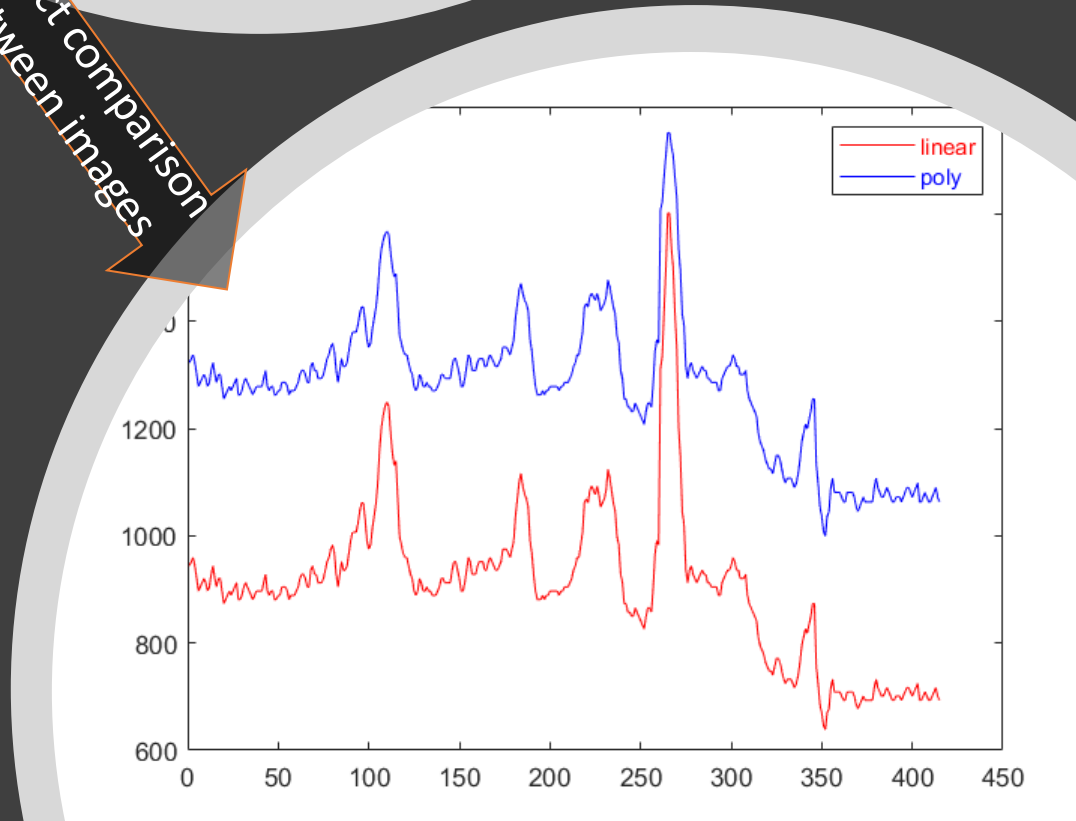
	mean Image #1	std Image #1	saturated #1	mean Image #2	std Image #2	saturated #2
1	1053.94	336.58	7494.51	1372.21	267.12	7697.21

Manual drawing of profile line

Enzyme activity along a profile line determined for a single rhizotrone calibrated by linear (red) and polynom (blue) fit



Direct comparison between images




Draw Profile Line

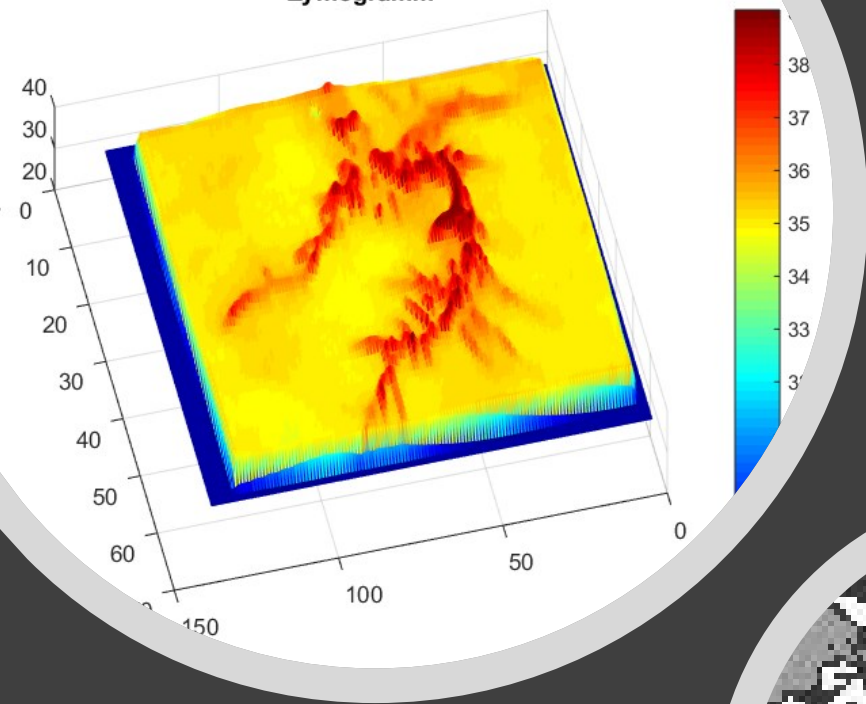
Select Image #1: linear

Select Image #2: poly

Select Background Image (optional): rh



Zymogramm

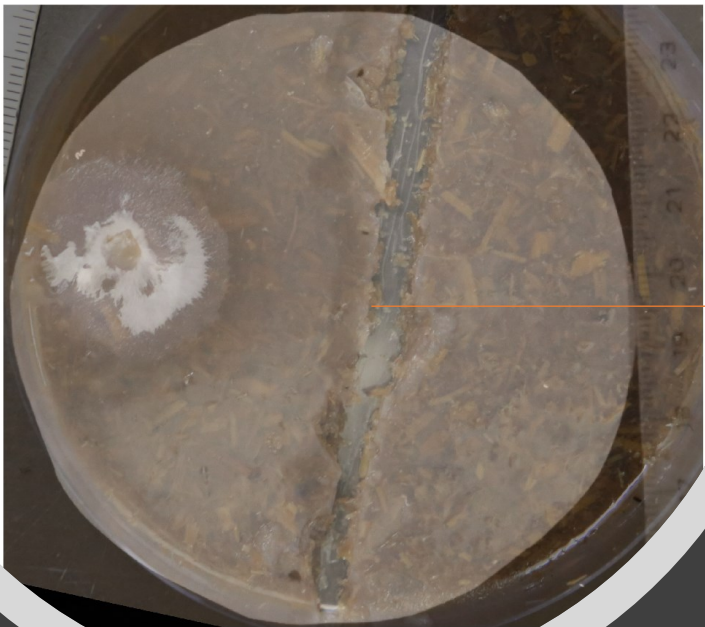


ROOT-O-MAT

We planted a little tree on agarose and analyzed the enzyme activity by zymography without any soil.



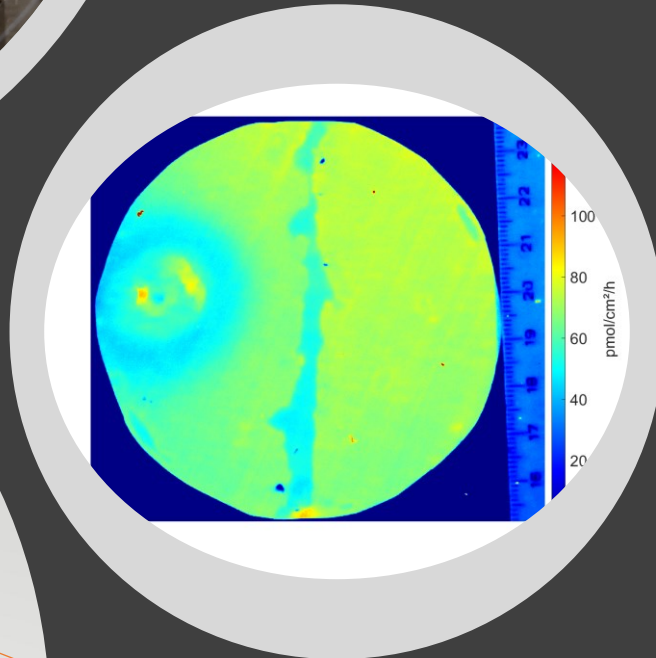
ROOT-O-MAT



Registered and
grey scale converted
zymograph

The barrier without agarose

Fungi grown on agarose, and we measured the enzyme activity by zymography without any soil.



Two areas of interest for comparison