A phenotyping system quantifies pollen populations during heat stress using high-throughput microscopy and computer vision

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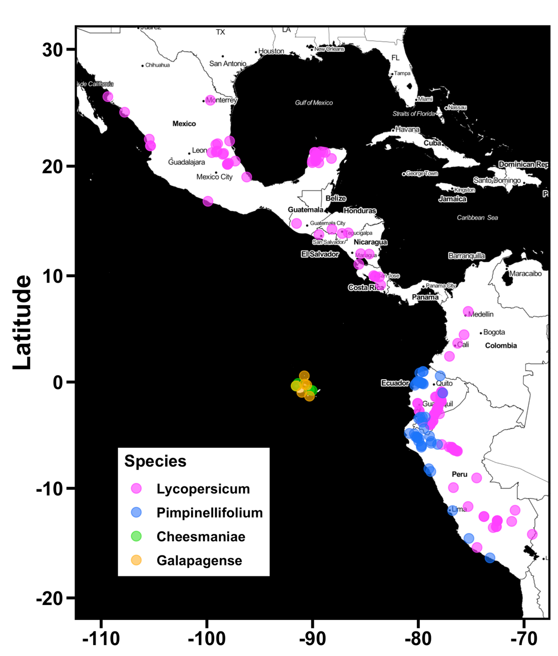
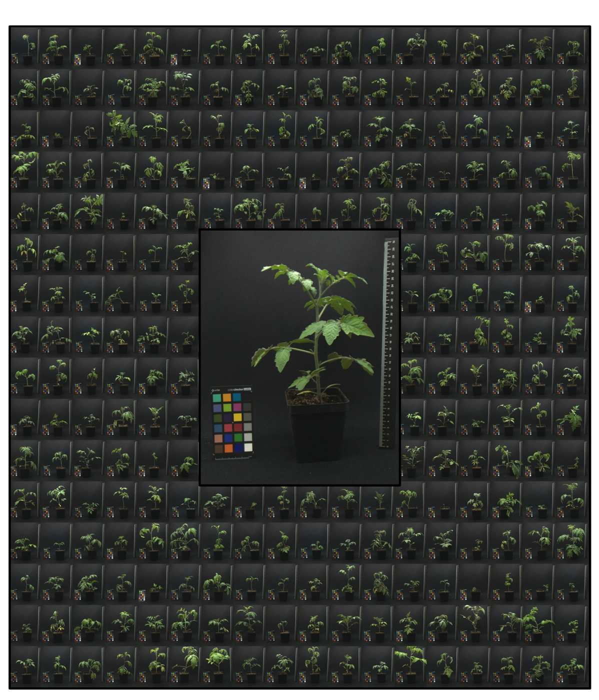
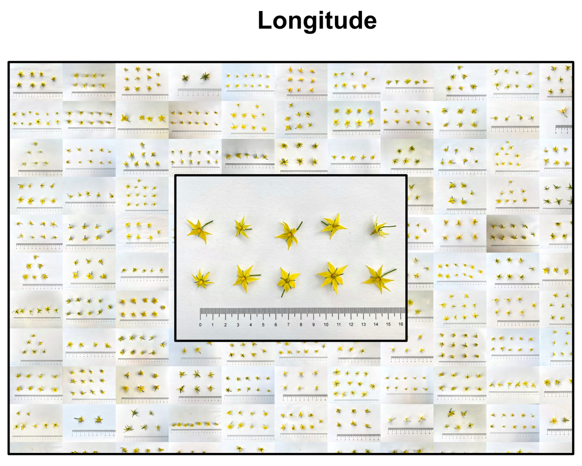
Abstract

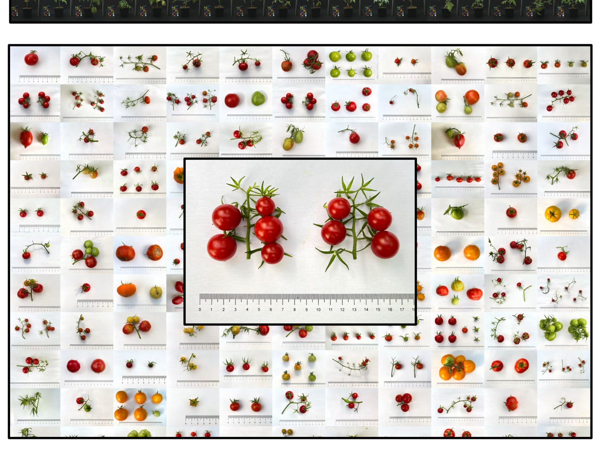
Plant reproduction is sensitive to heat stress. Pollen tube growth can be accelerated or arrested by high temperatures, leading to unstable tubes, failed sperm cell delivery, and ultimately crop yield loss. Pollen growth dynamics have historically been observed on the scale of individual pollen grains, but there are only a few studies surveying pollen populations across genotypes and environmental conditions. We have developed a phenotyping system that quantifies tomato pollen characteristics on a large scale and under varied heat stress conditions. In this system, we combined high-throughput bright-field microscopy with automated object detection and tracking to quantify pollen phenotypes. We used this method to survey pollen from a diverse panel of 220 genome-sequenced tomato and close wild relative accessions. Pollen from these accessions showed a wide variety of responses to heat stress across measured phenotypes, suggesting genetic components influence heat stress responses that can be used to improve tomato productivity in challenging climates. This method can be readily adapted to pollen from different species, providing a way to rapidly characterize molecular mechanisms of pollen functions and identify thermotolerant alleles for enhanced reproduction in flowering plants.

220 accessions capture genetic diversity of tomato and wild relatives

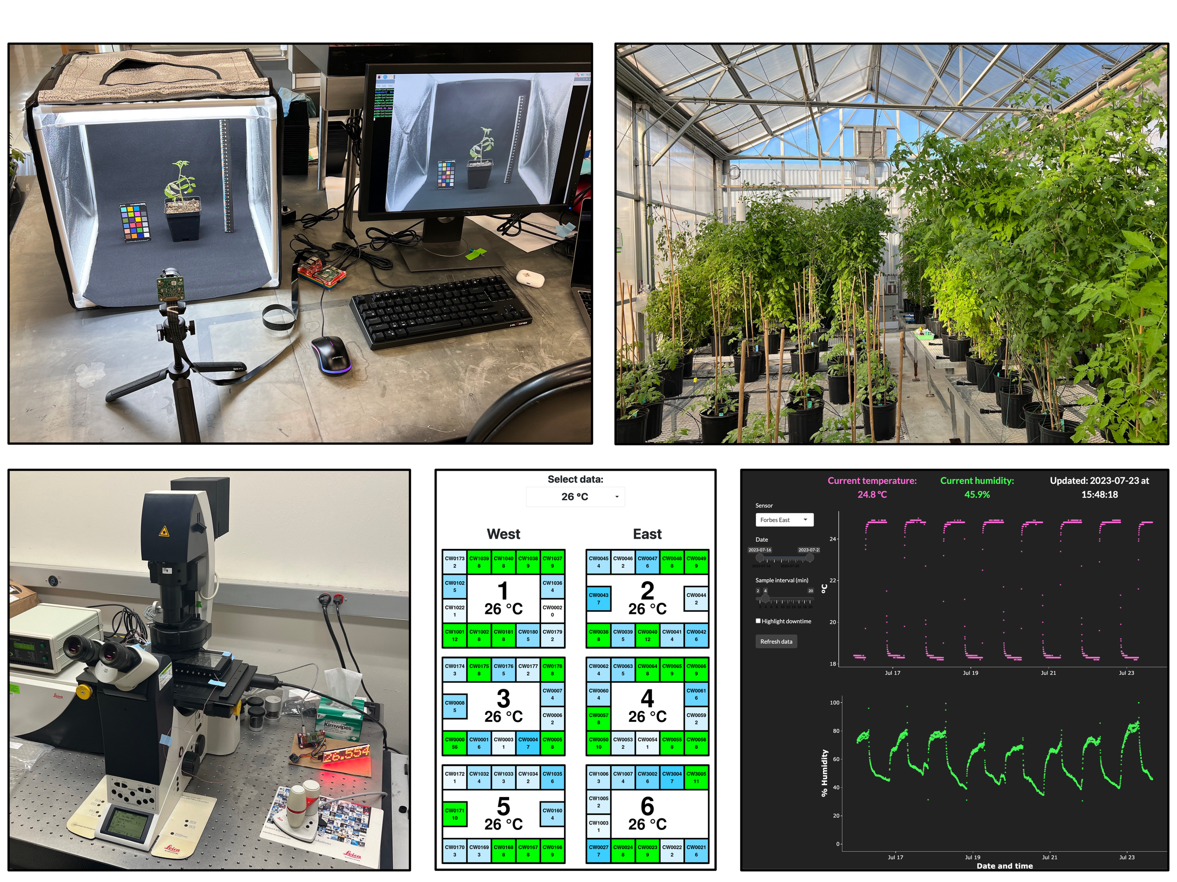








Open-source phenotyping systems, sensors, and web apps



Acknowledgements

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This poster is available online:

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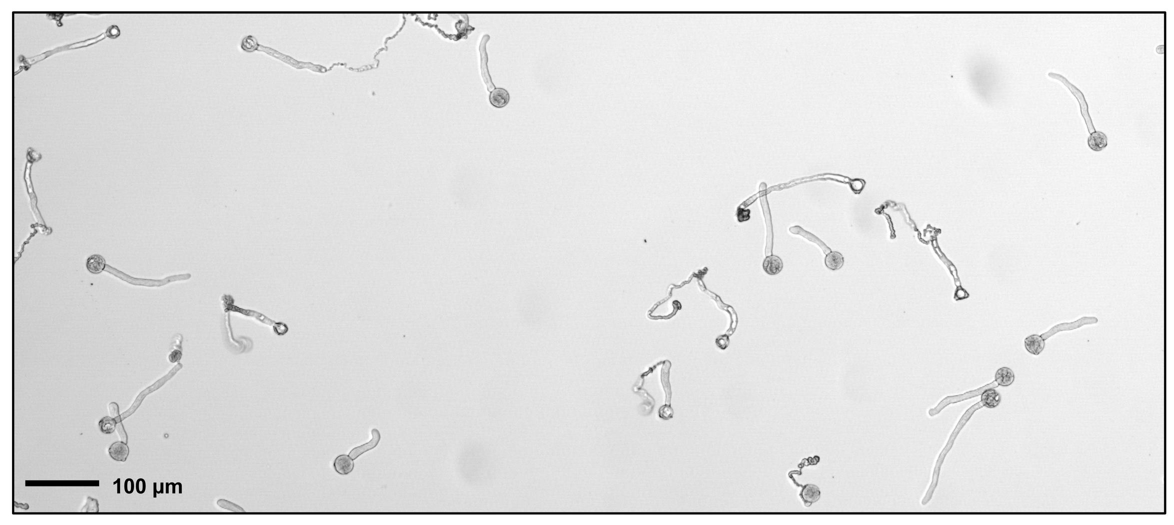
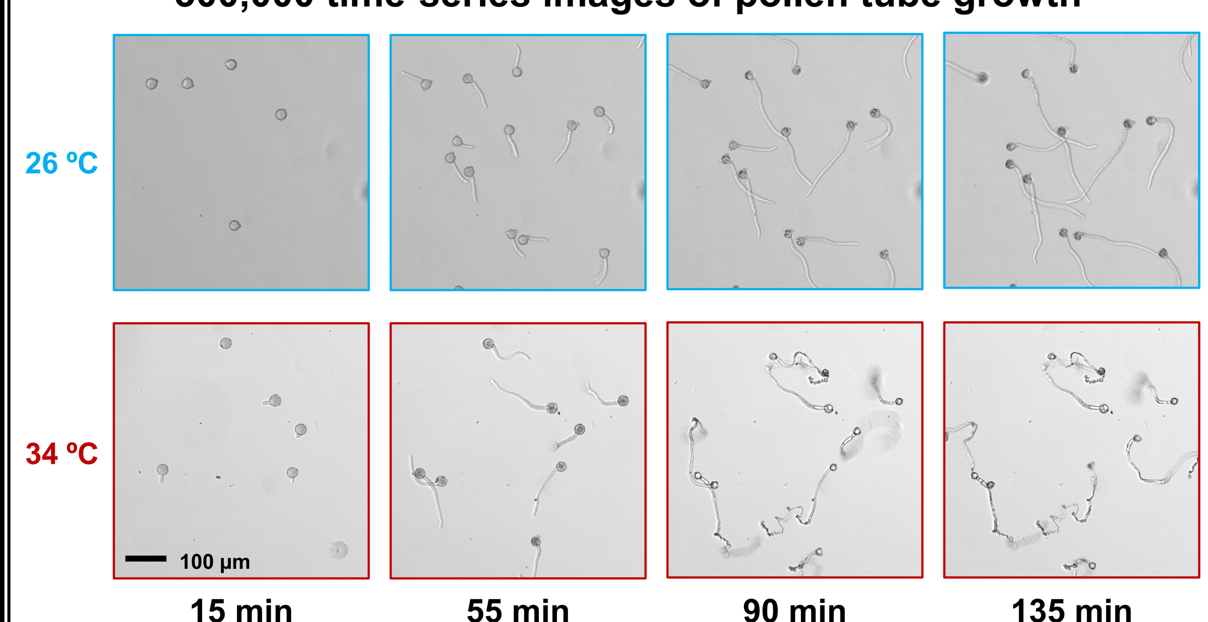
Heat stress disrupts pollen tube growth



~300,000 time-series images of pollen tube growth



A convolutional neural network was trained to label pollen features

CenterNet Hourglass104

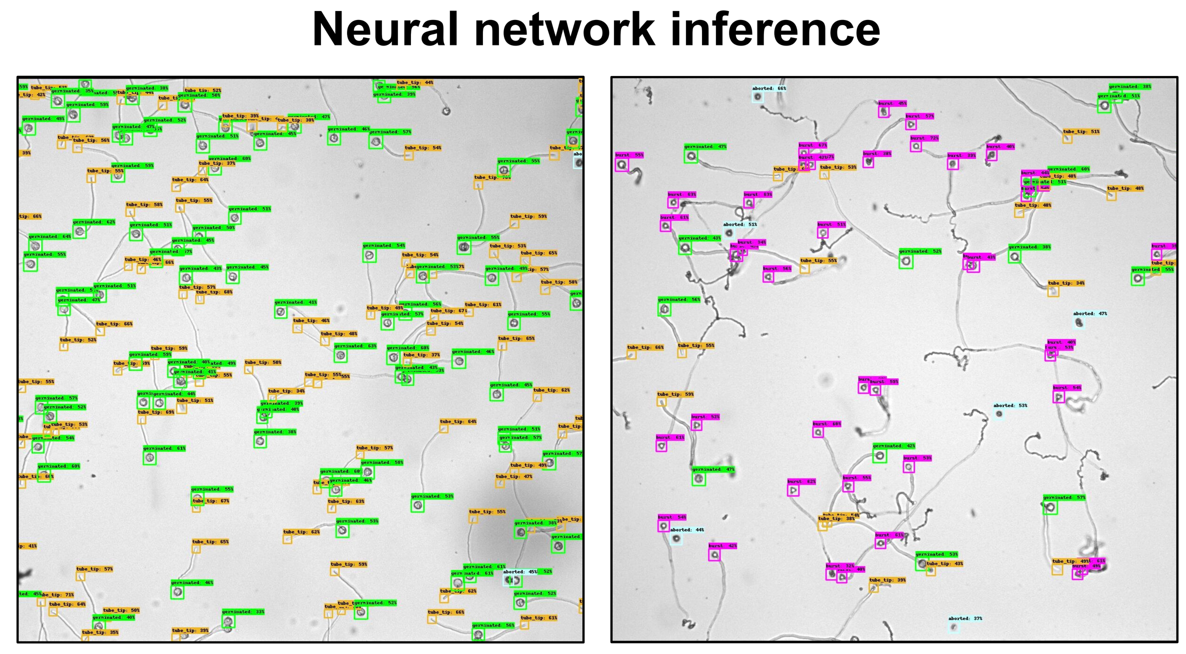
- 1000 labeled images
  - ~80,000 labels
- 8 minutes per image labeling
- Trained on Nvidia A100 for 6 hours
- Inference on 4 Nvidia V100s
  - 19 hours for ~300,000 images
  - 2,000x human speed

TensorFlow

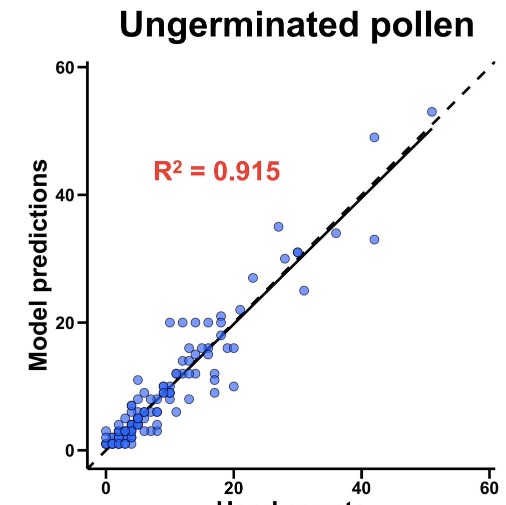
The University of Arizona High Performance Computing

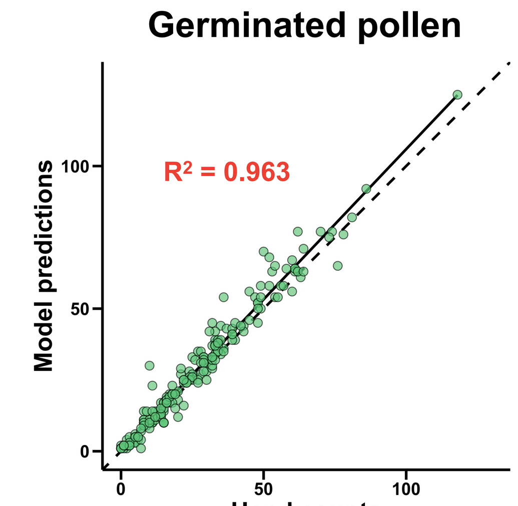
Jetstream2

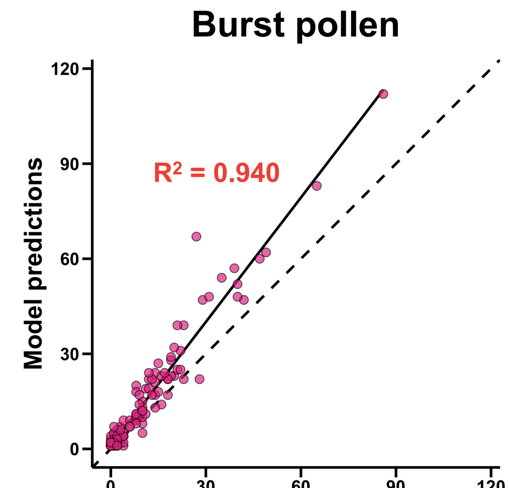
Neural network inference

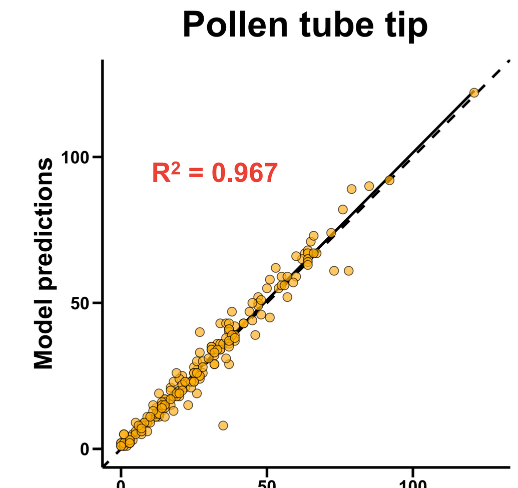


The neural network is accurate



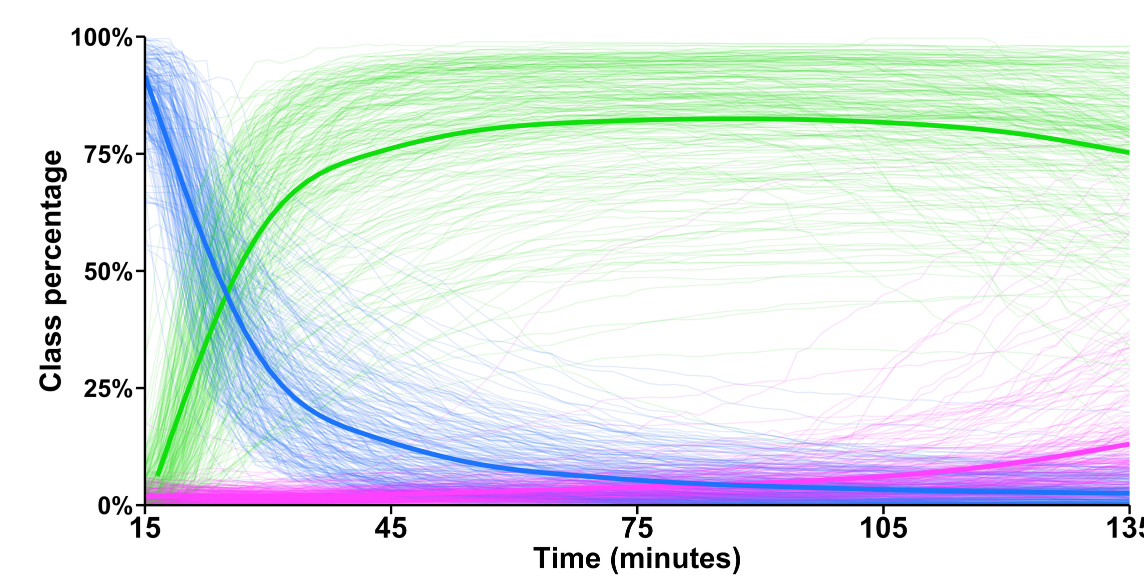




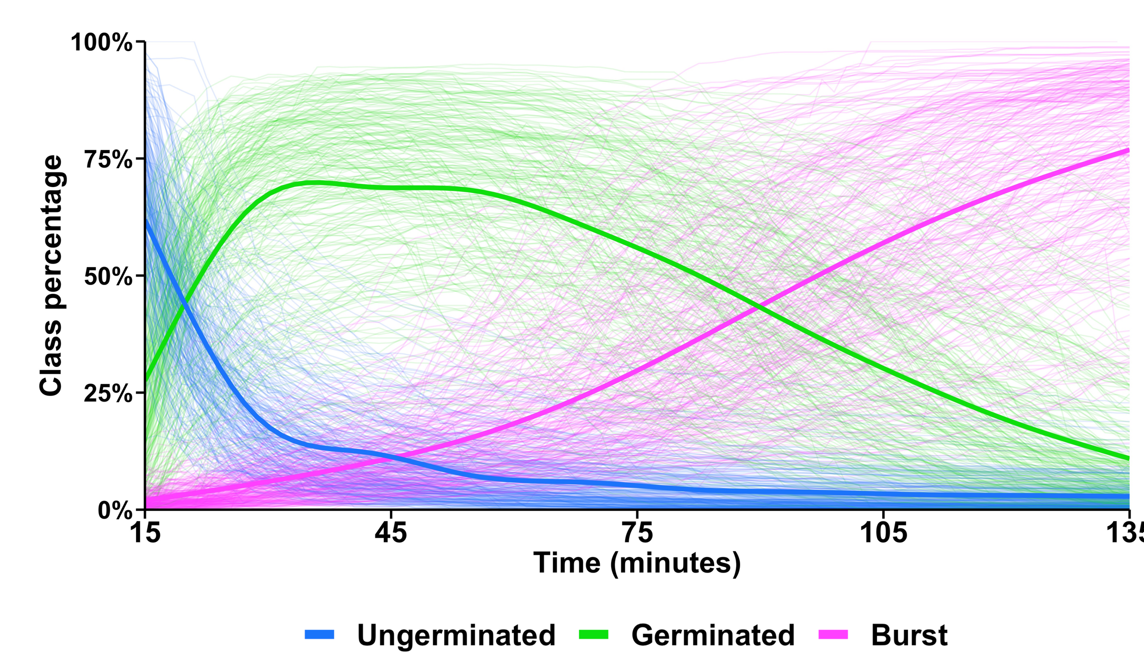


Pollen populations were measured by the phenotyping system

Pollen classes by accession at 26 °C



Pollen classes by accession at 34 °C



Additional phenotypes were quantified by tracking tube tips

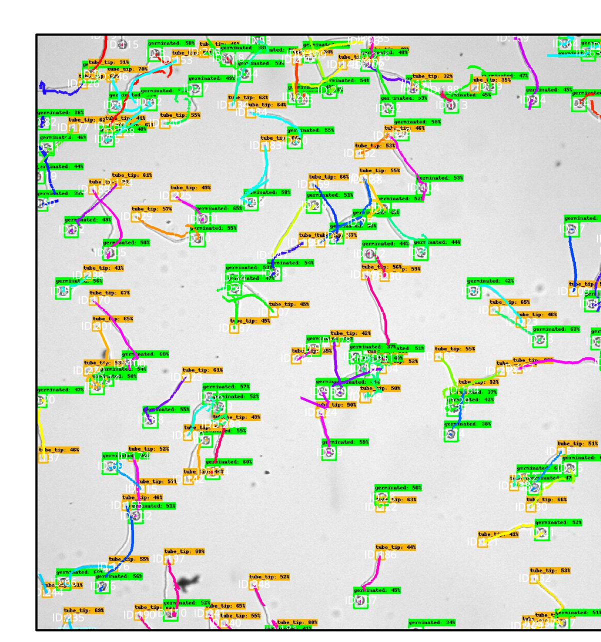
Multiple object tracking

- Modified version of Bayesian Tracker reconstructs pollen and tube tip trajectories.
- Tracks revised with pollen tube growth characteristics.

btrack

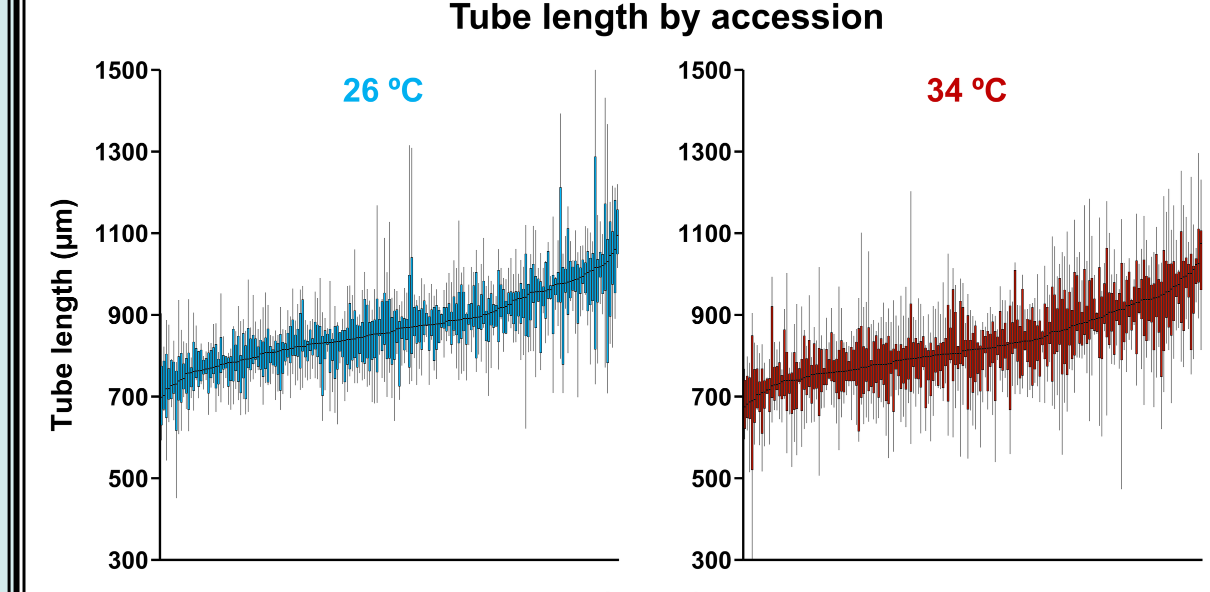
Lowe Lab, UCL

https://github.com/quantumjot/btrack



Pollen tube phenotypes are affected by heat stress

Tube length by accession



Tube growth speed by accession