Considering Interface Concavity in Spongy Mesophyll Segmentation
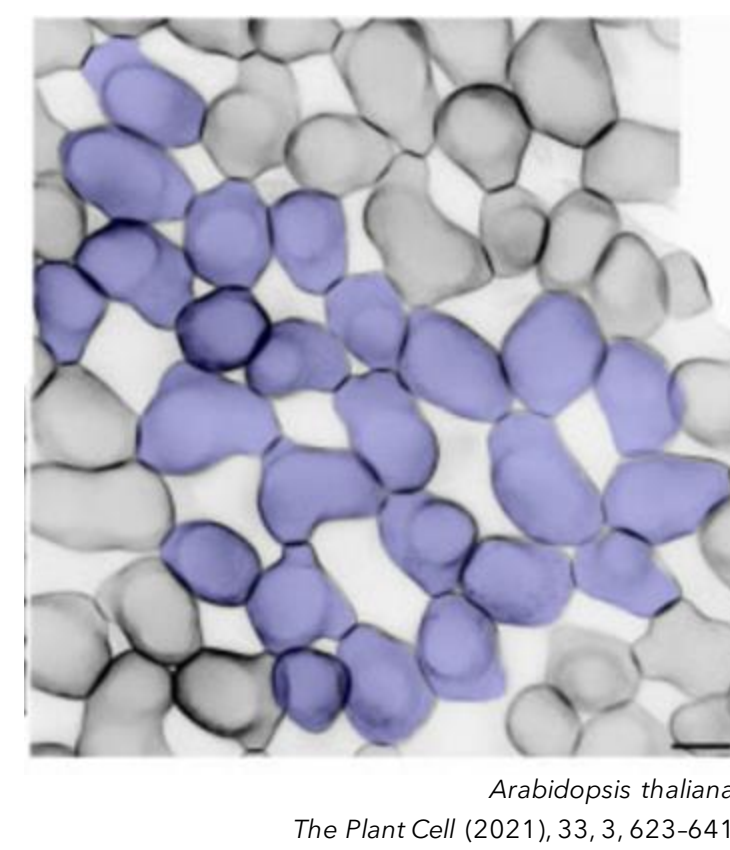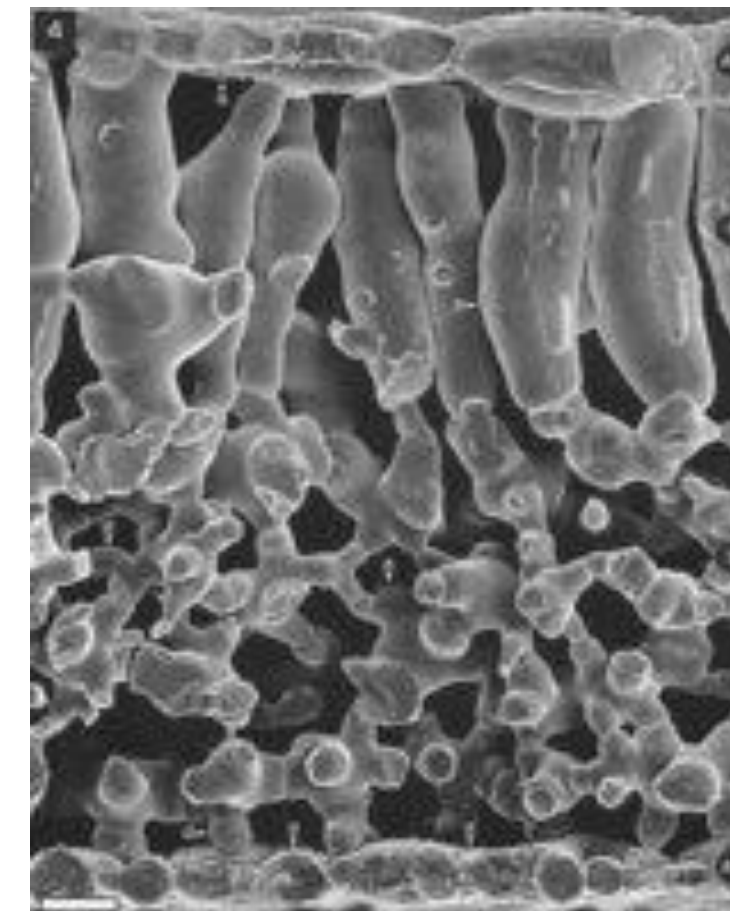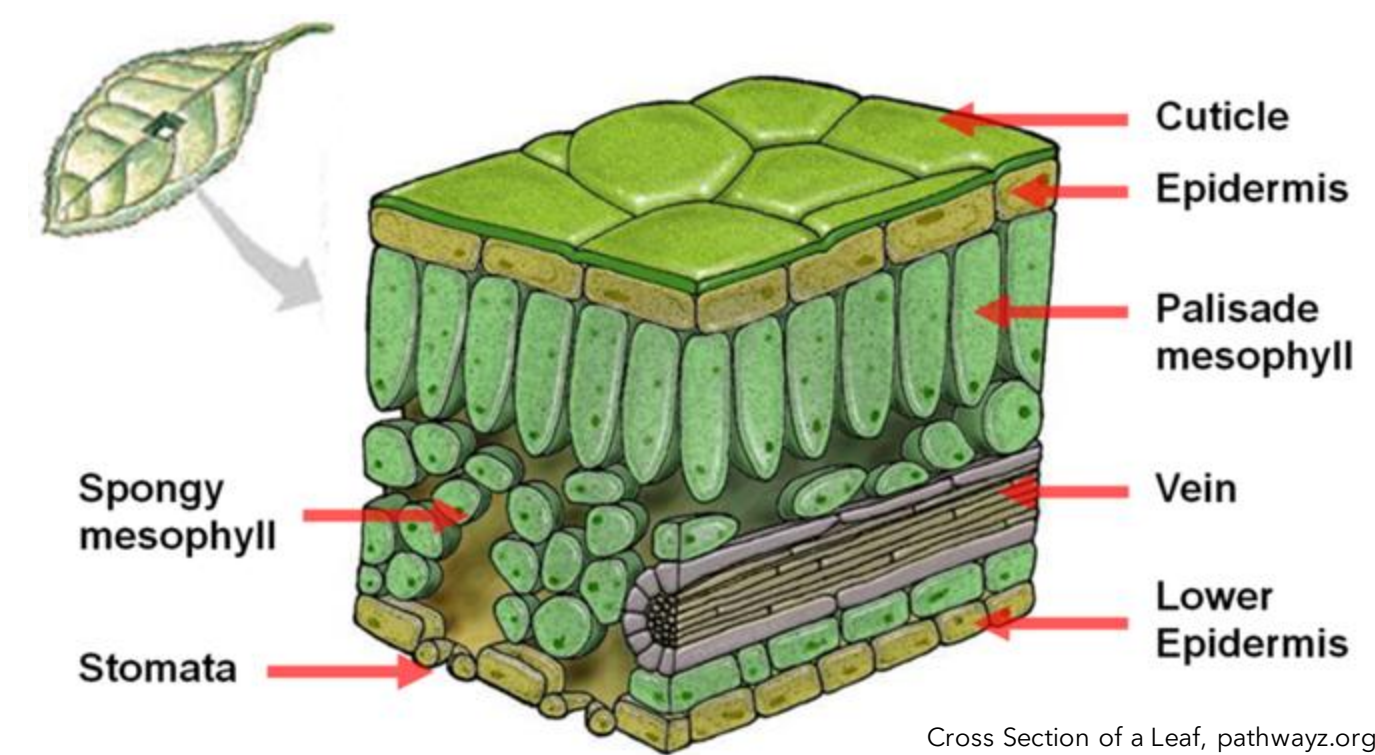
Evan A. Cook[1,3], Arthur K. MacKeith[2,3], Allison E. Culbert[2,3], Joy Pajarla[2,3], Craig R. Brodersen[6], Adam B. Roddy[7], Mark D. Shattuck[8], and Corey S. O'Hern[2,3,4,5]

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Introduction and Overview

Cross Section of a Leaf, pathwayz.org

Phaseous vulgaris
Planta (1987), 172, 20-37

Arabidopsis thaliana
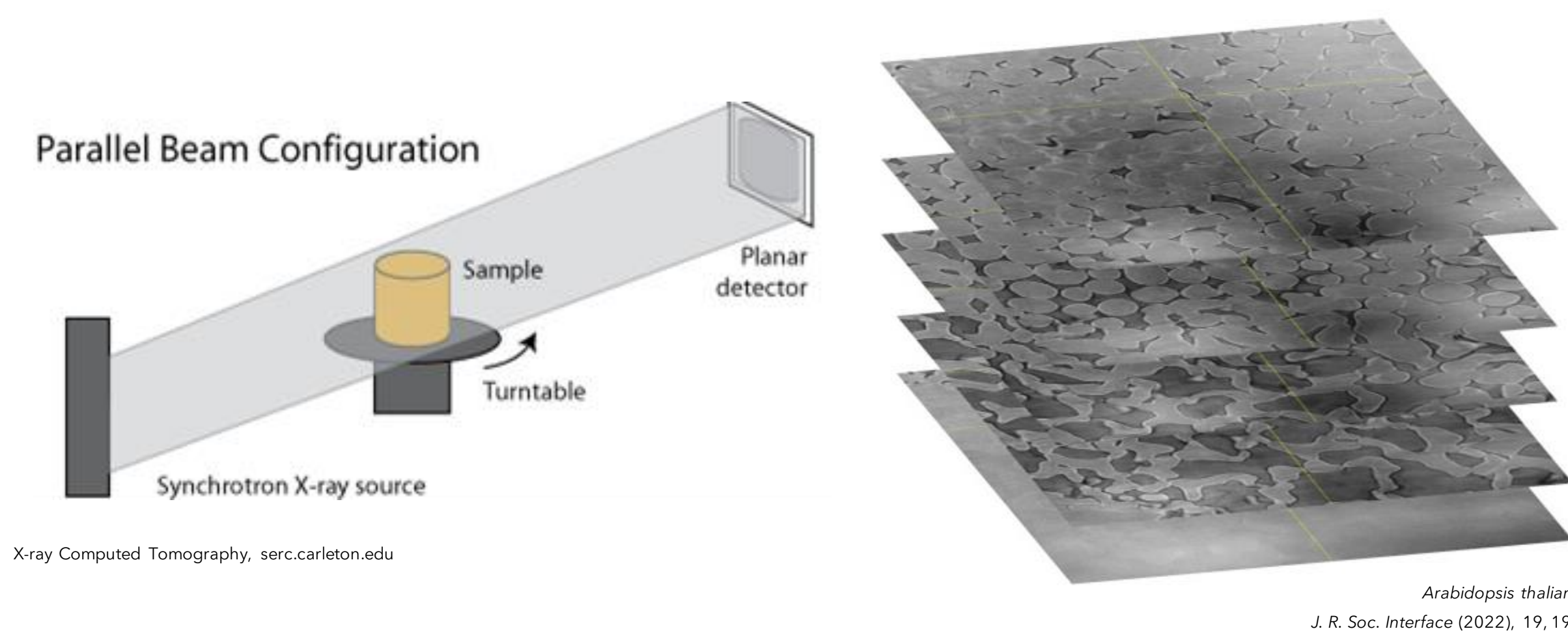*The Plant Cell* (2021), 33, 3, 623-641

Questions

- How can we maximize usable cell count in **segmenting microCT scans** for use in later studies?
- How do our eyes identify separate cells, and how might that inform our algorithm design?
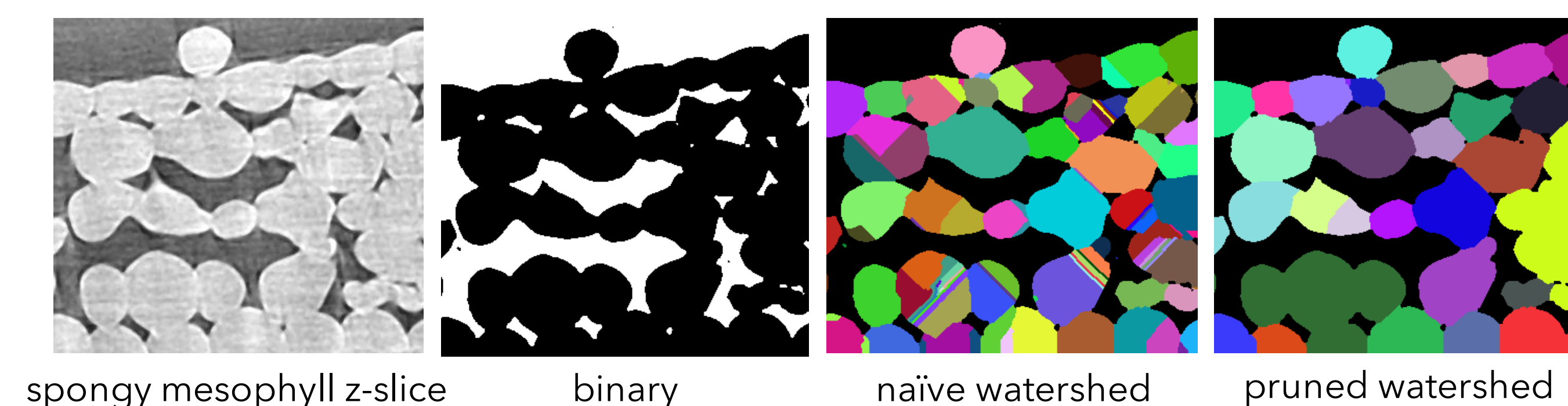
Goals

- Designing and implementing a **new 3D segmentation algorithm**
- Testing this algorithm's **accuracy** and **viability** towards use in further studies

microCT Scans

X-ray Computed Tomography, serc.carleton.edu

Arabidopsis thaliana
J. R. Soc. Interface (2022), 19, 197

Existing Segmentation Algorithm

spongy mesophyll z-slice   binary   naïve watershed   pruned watershed


Camellia yunnanensis

Summary

- Algorithm is generally accurate, but can struggle when parsing compact clusters.
- Pruning stage can produce significant undersegmentation. Oversegmentation is preferable in the human verification stage.
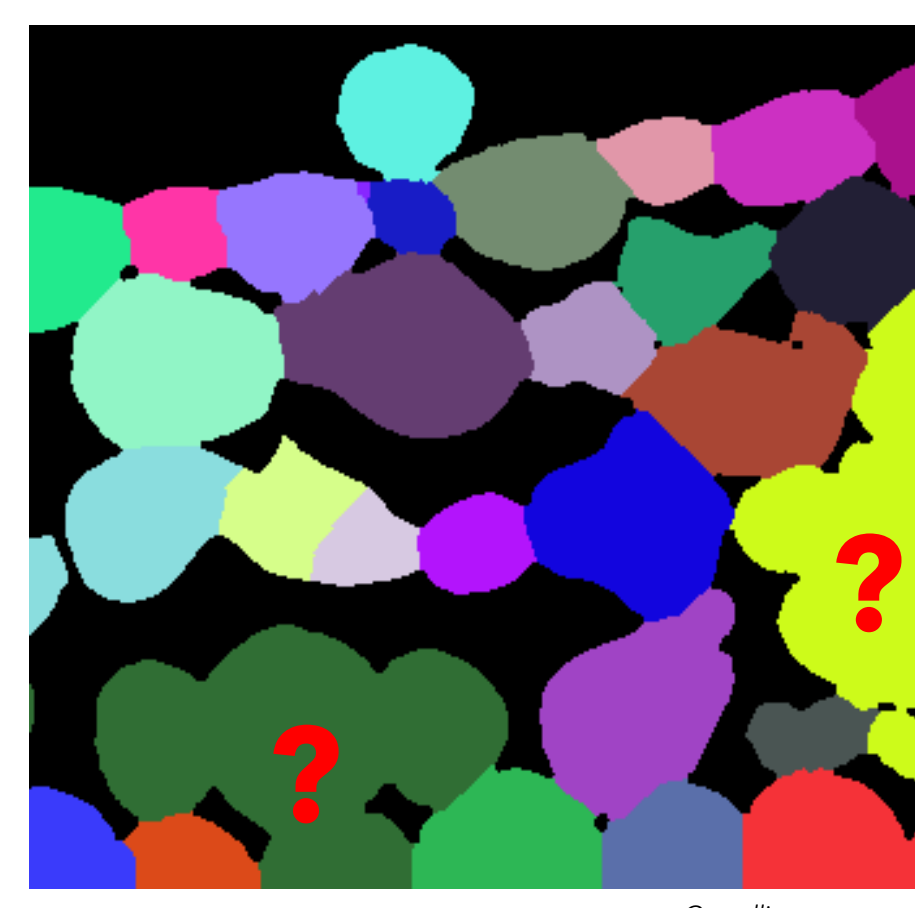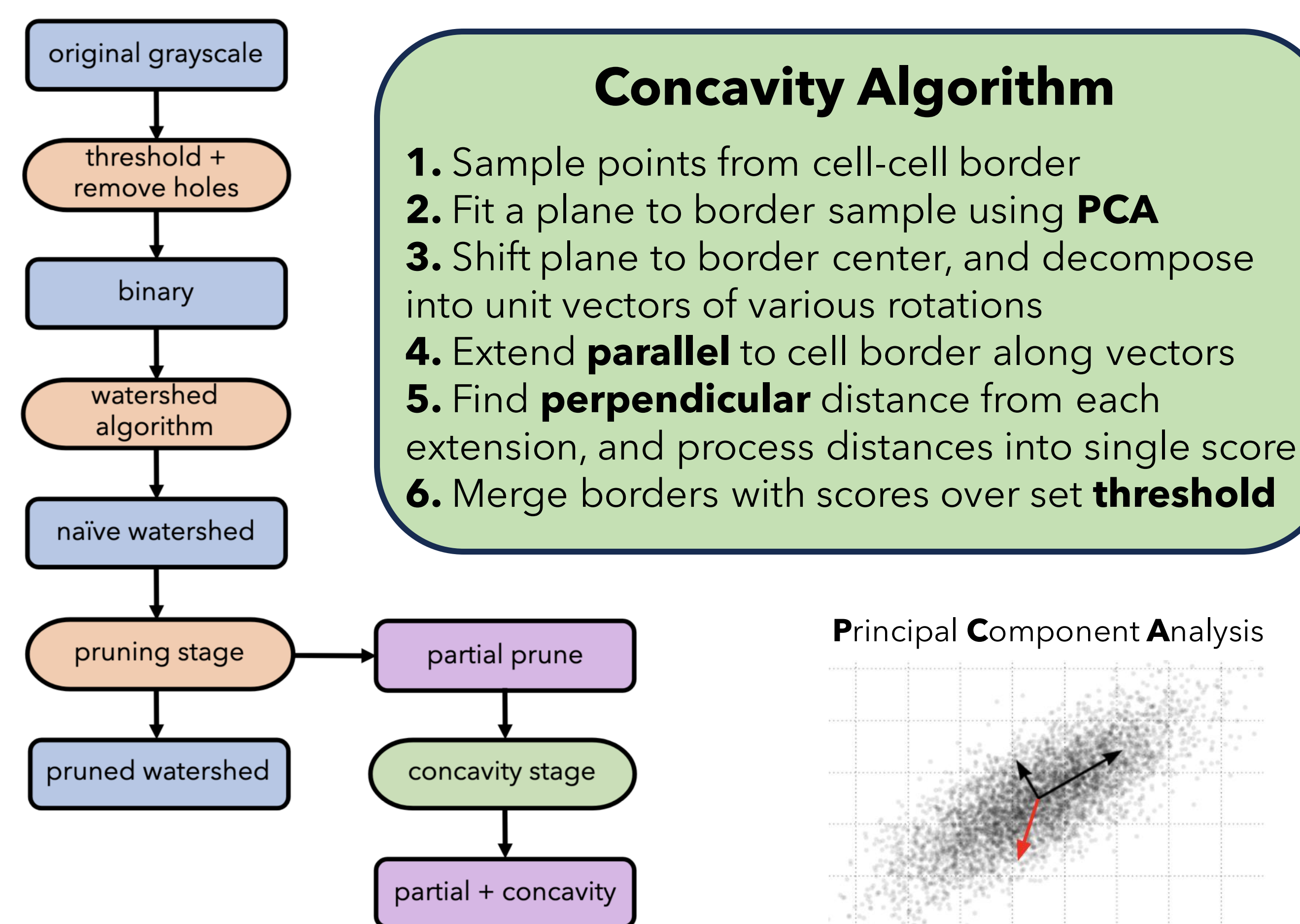
Concavity Intuition in 2D

- One property of spongy mesophyll is observed **necking** of cell-cell interfaces. When assessing prospective borders, actual borders will usually be **more concave** than false borders. As such, we generate a quantitative measure of concavity:

concave border → **small** distance   **convex** border → **large** distance
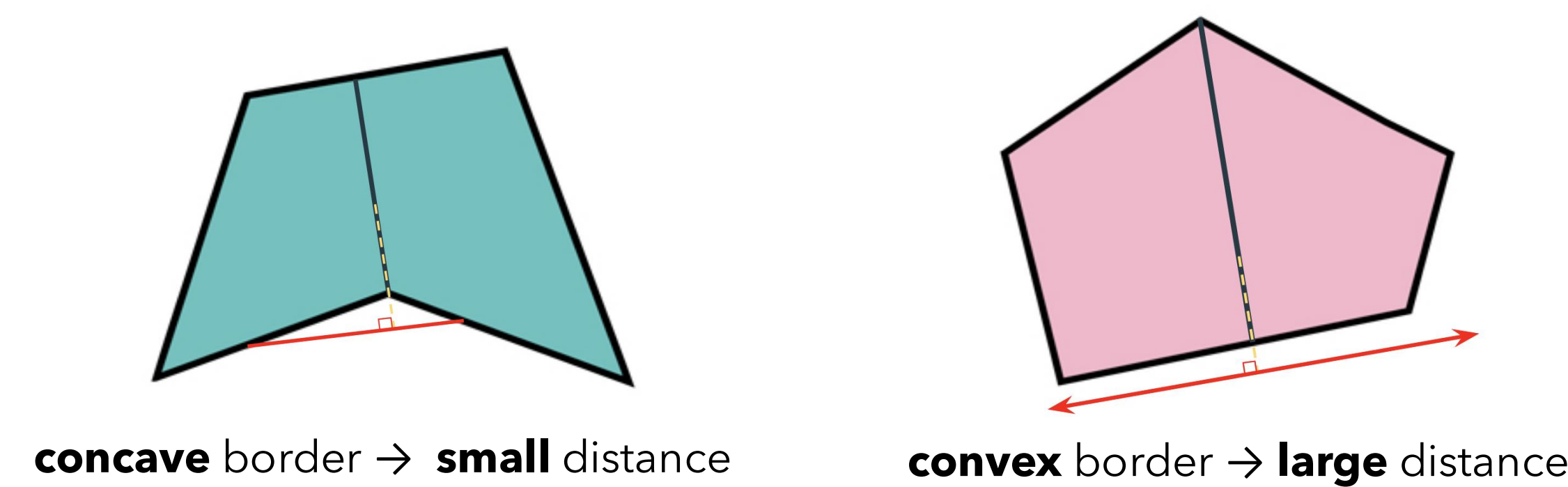
- From the corner, we step **parallel** to the cell border, then measure **perpendicular** distance to cell matter. Large distances indicate convex borders, which are usually false and should be merged.
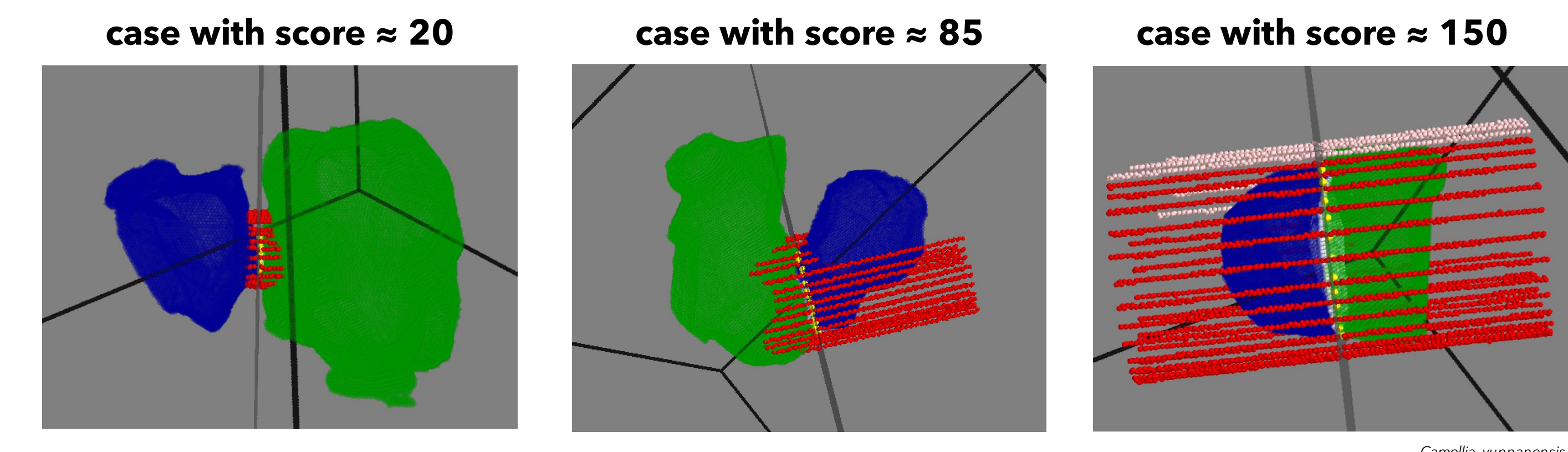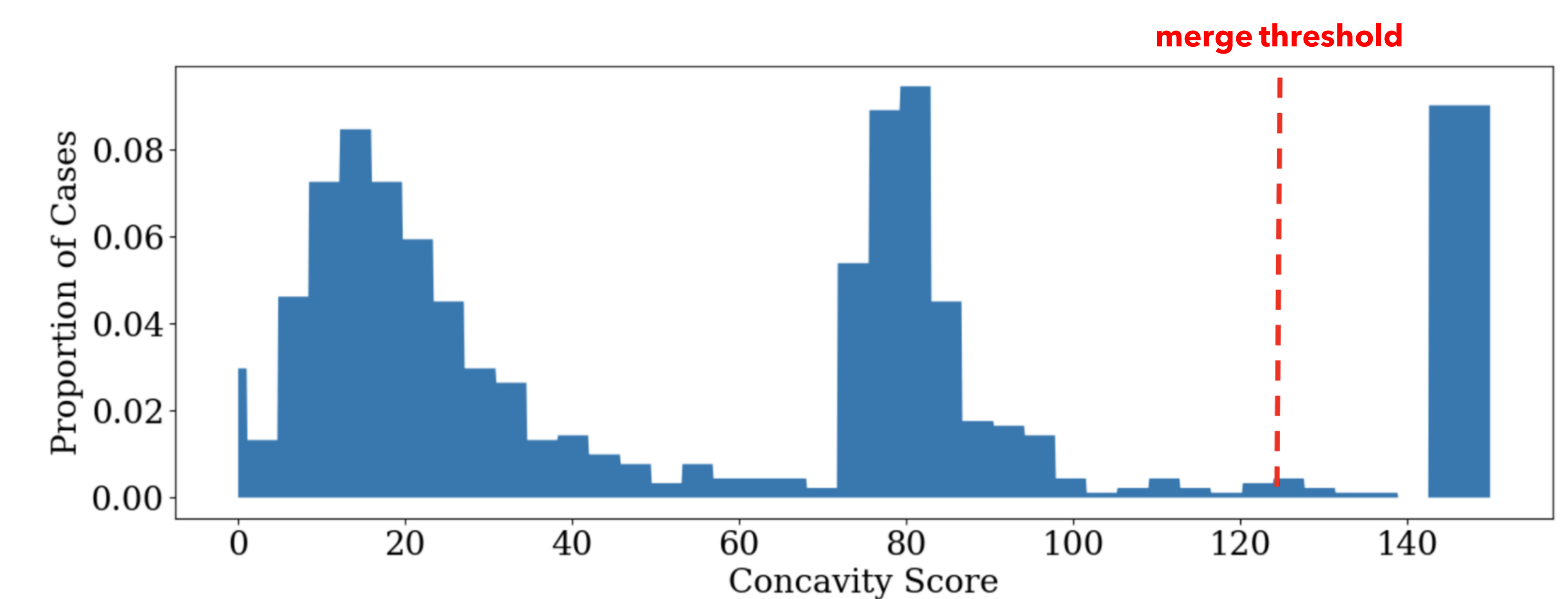
Core Algorithm in 3D

original grayscale → threshold + remove holes → binary → watershed algorithm → naïve watershed → pruning stage → pruned watershed; pruning stage → partial prune → concavity stage → partial + concavity

Concavity Algorithm

1. Sample points from cell-cell border
2. Fit a plane to border sample using **PCA**
3. Shift plane to border center, and decompose into unit vectors of various rotations
4. Extend **parallel** to cell border along vectors
5. Find **perpendicular** distance from each extension, and process distances into single score
6. Merge borders with scores over set **threshold**

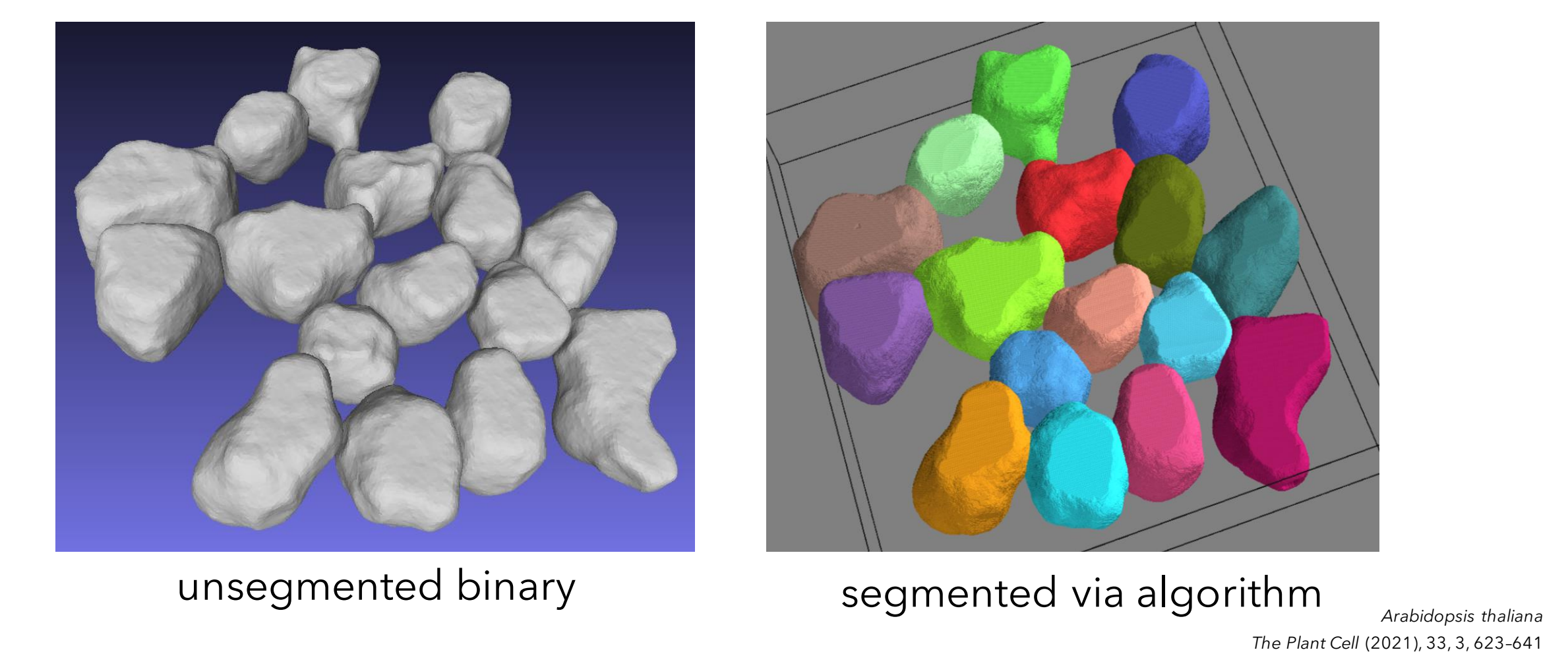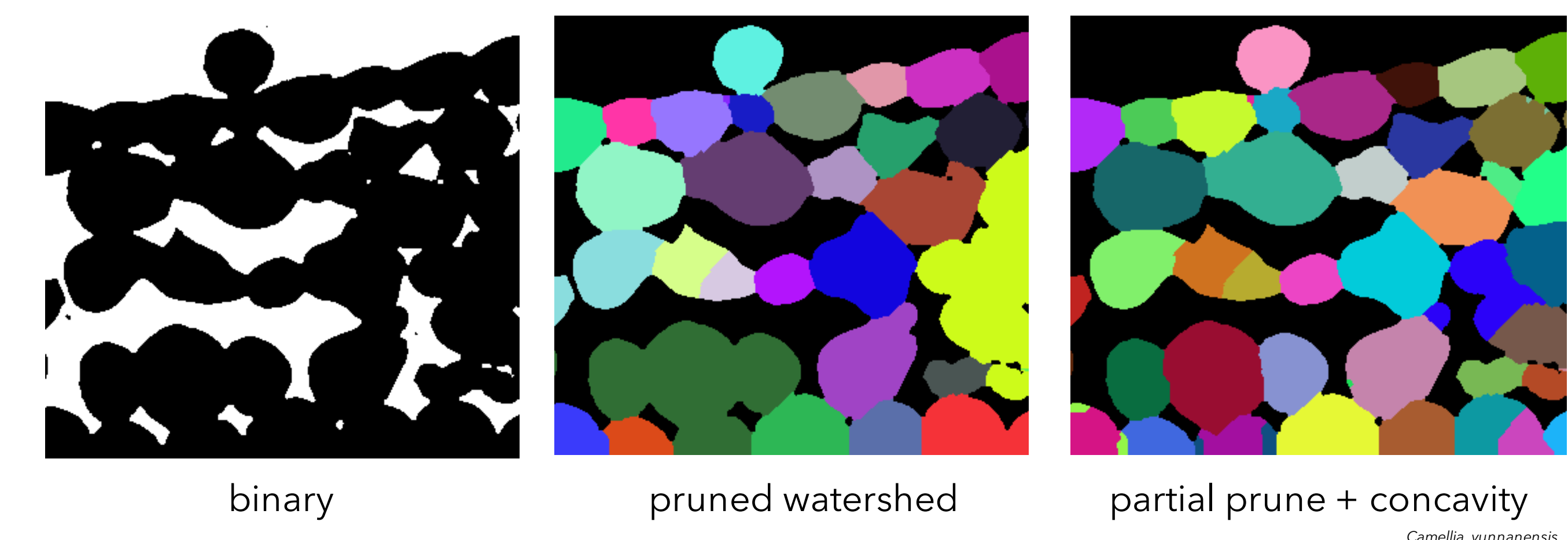**P**rincipal **C**omponent **A**nalysis

Acknowledgements

References

[1] Cross Section of a Leaf, pathwayz.org
[2] *Planta* (1987), 172, 20-37
[3] *The Plant Cell* (2021), 33, 3, 623–641
[4] X-ray Computed Tomography, serc.carleton.edu
[5] *J. R. Soc. Interface* (2022), 19, 197

Algorithm Behavior

merge threshold

case with score ≈ 20   case with score ≈ 85   case with score ≈ 150

Camellia yunnanensis

Sample Output

binary   pruned watershed   partial prune + concavity

Camellia yunnanensis

unsegmented binary   segmented via algorithm

Arabidopsis thaliana
The Plant Cell (2021), 33, 3, 623-641

Future Work

Our algorithm displays promising accuracy on test datasets (above), addressing observed shortcomings of the current algorithm. Our next steps are to increase the algorithm's robustness, and to investigate its viability with additional datasets and future studies.

(1) Analyze more of the 40 species for which we have leaf or flower data
(2) Automate the setting of tuning parameters in the algorithm
(3) Assess accuracy with more complicated datasets/ground truths
(4) Apply increased dataset of confidently segmented cells to the calculation of useful metrics (volume, SA, porosity, etc.)