

# Immunolocalization of arabinoxylan in wheat bran for the effect of different treatments



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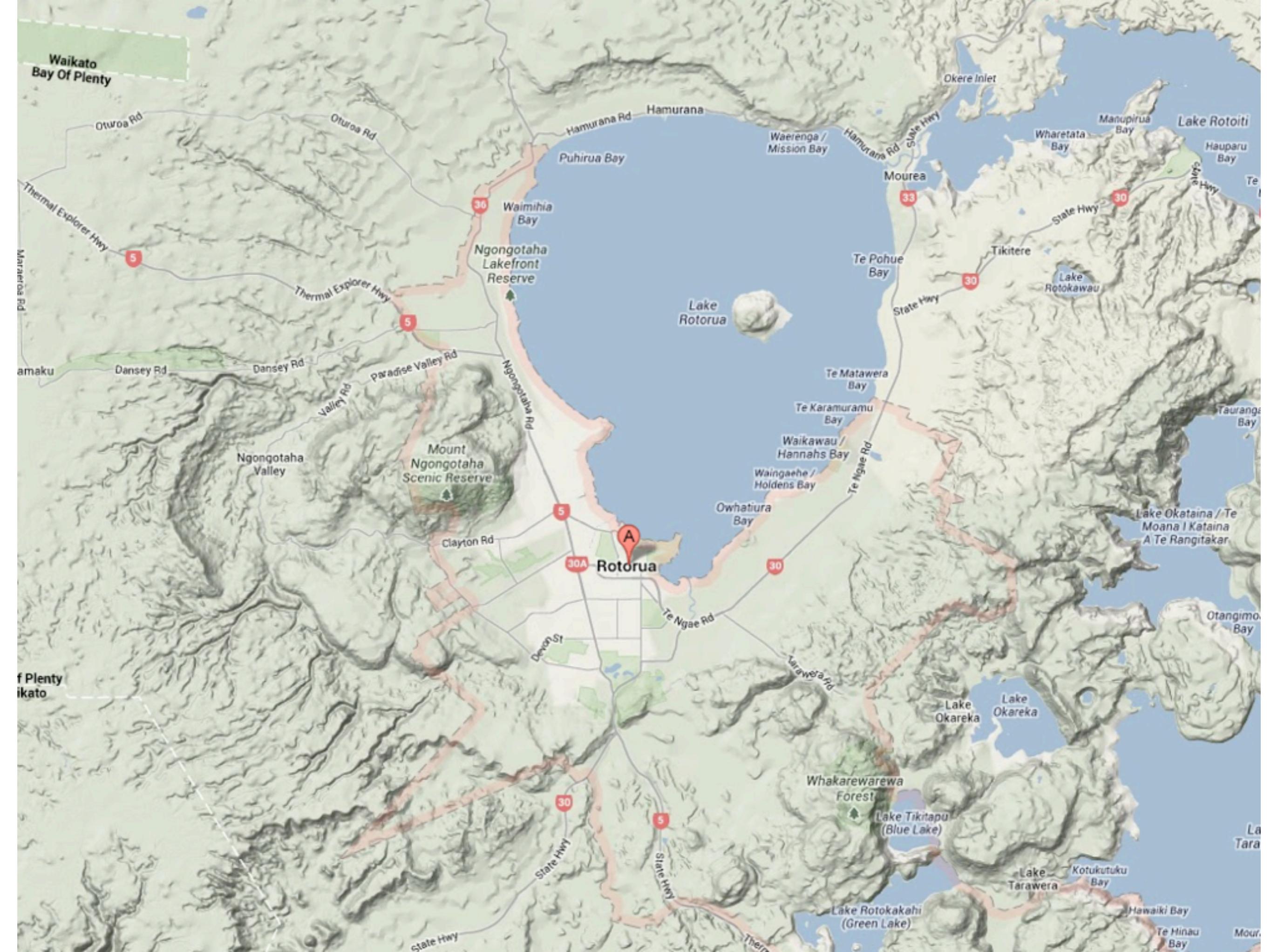
SCION   
forests · products · innovation

 cost











Sala St

Sala St

Froude St

Nelson St

30

Forest Research Village Rd

Nursery Rd

Nursery Rd

Nursery Rd

Hinganoa Drive

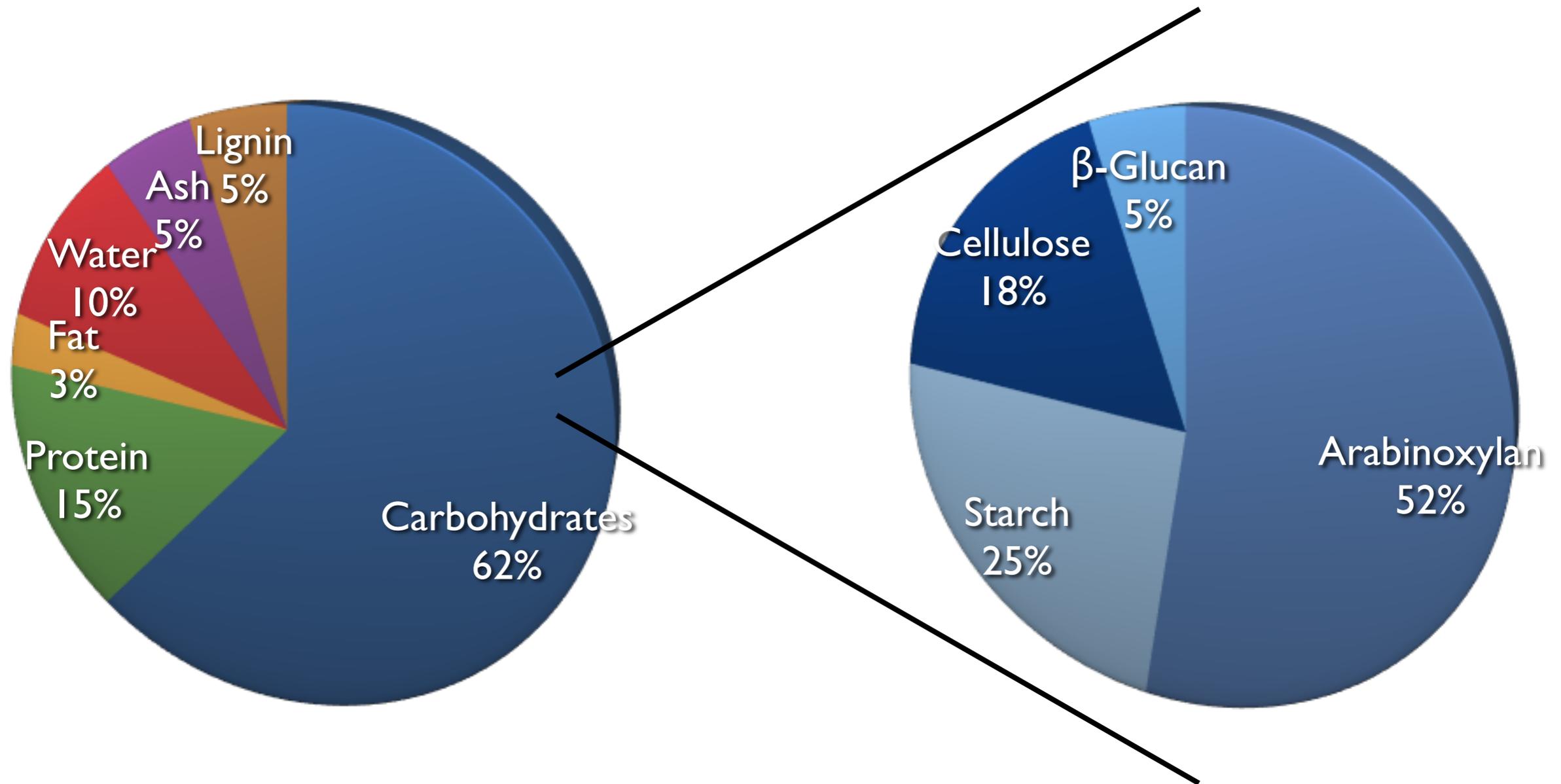
A





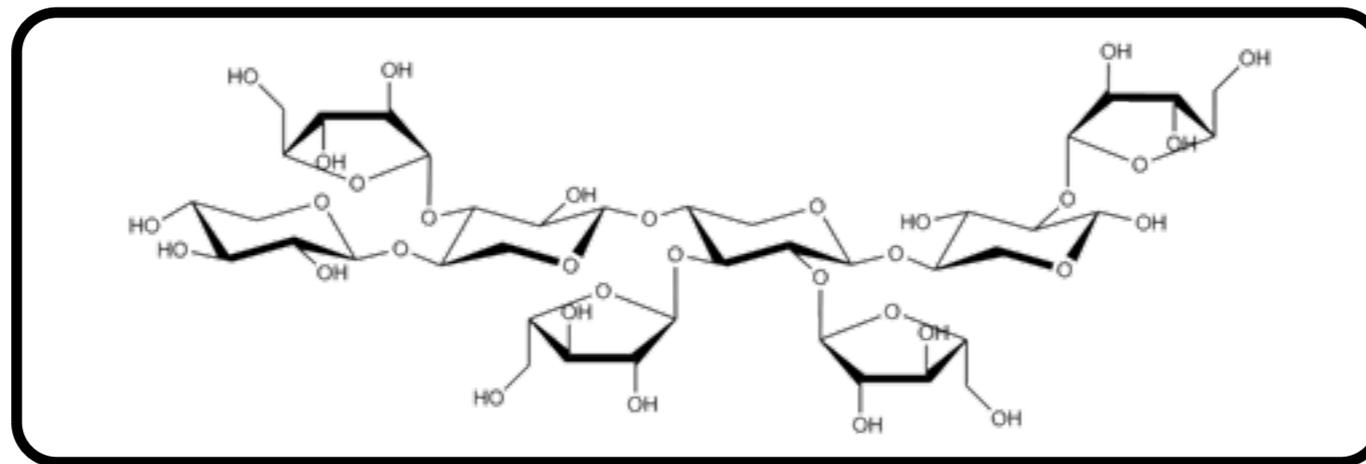


# Composition of wheat bran



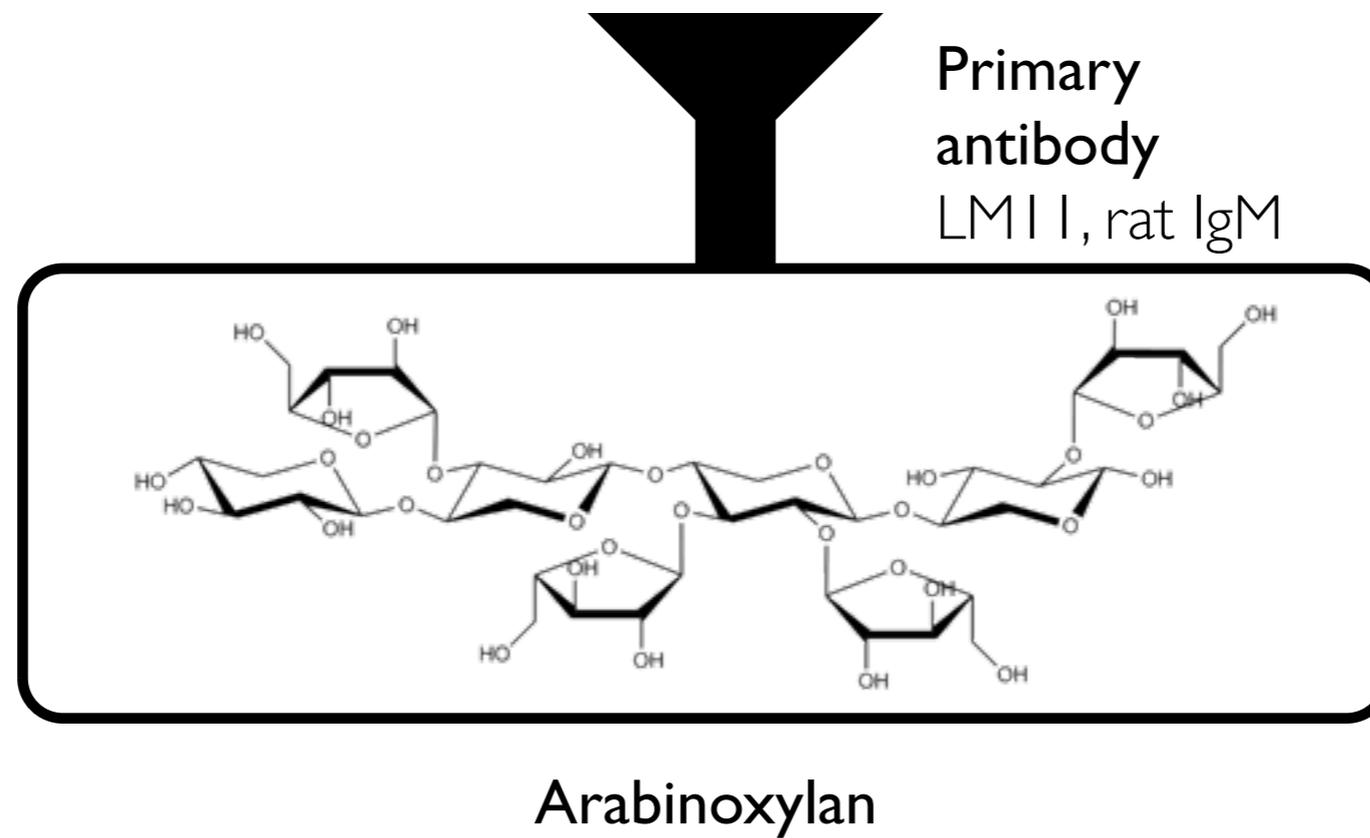
Source: Sun, Liu, Qu, Li; The Effects of Wheat Bran Composition on the Production of Biomass-Hydrolyzing Enzymes by *Penicillium decumbens*; Applied Biochemistry and Biotechnology 2007

# Immunolabeling

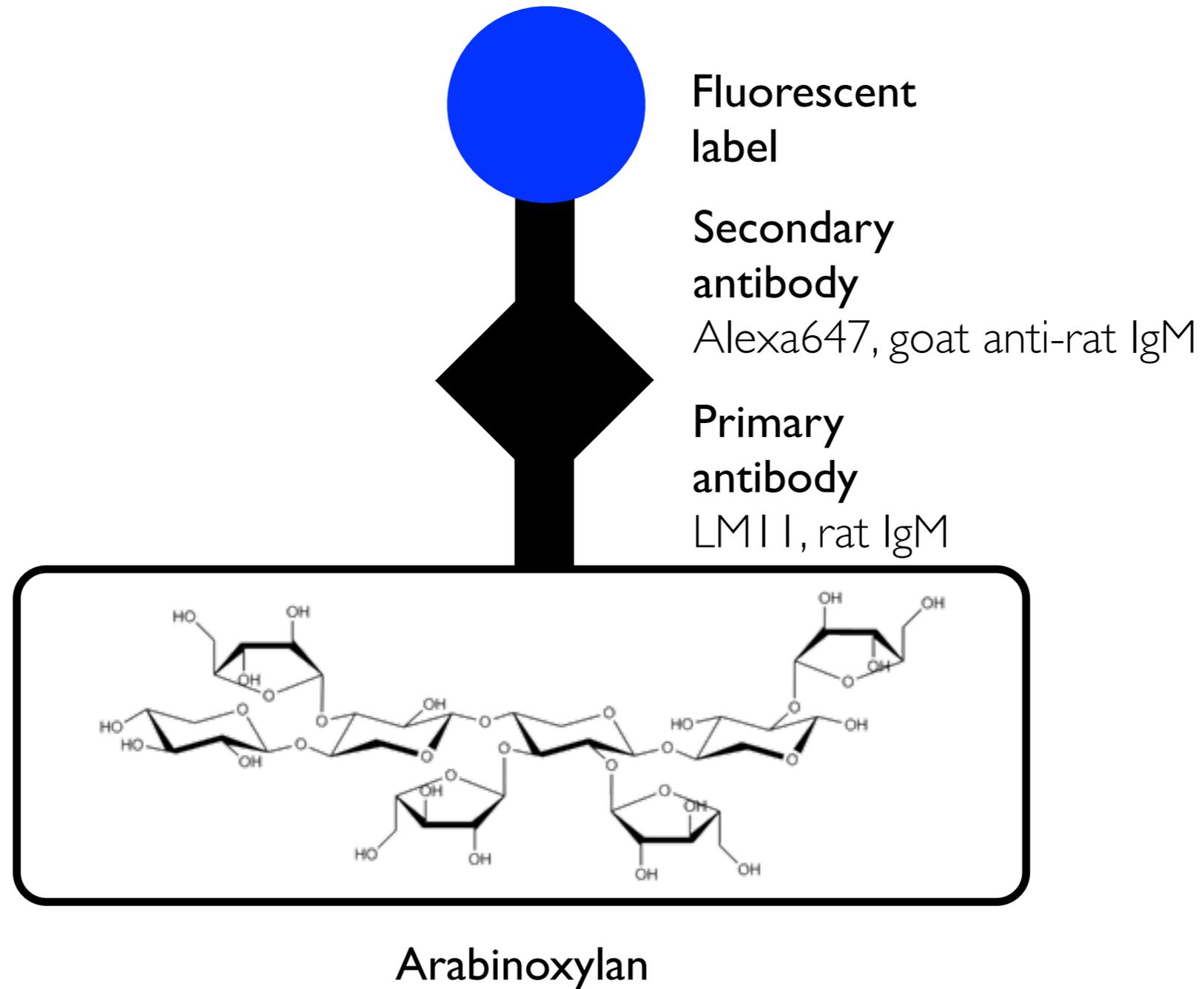


Arabinoxylan

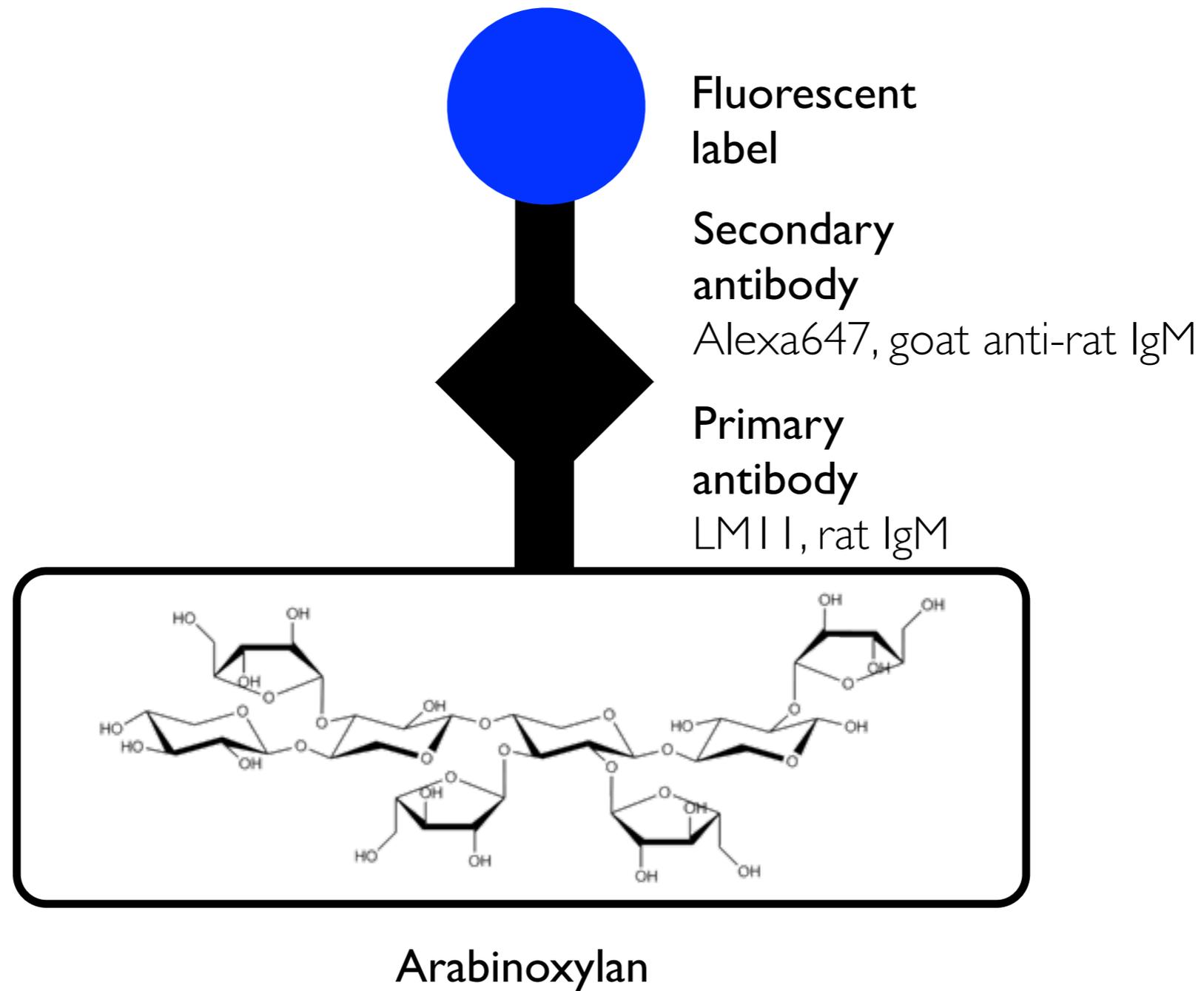
# Immunolabeling



# Immunolabeling

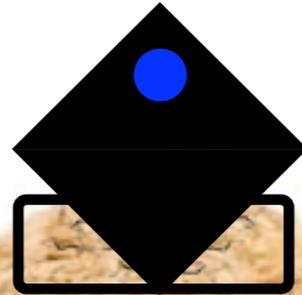


# Immunolabeling



# Immunolabeling

- Labeling of sample surface
- “Topographic ELISA”

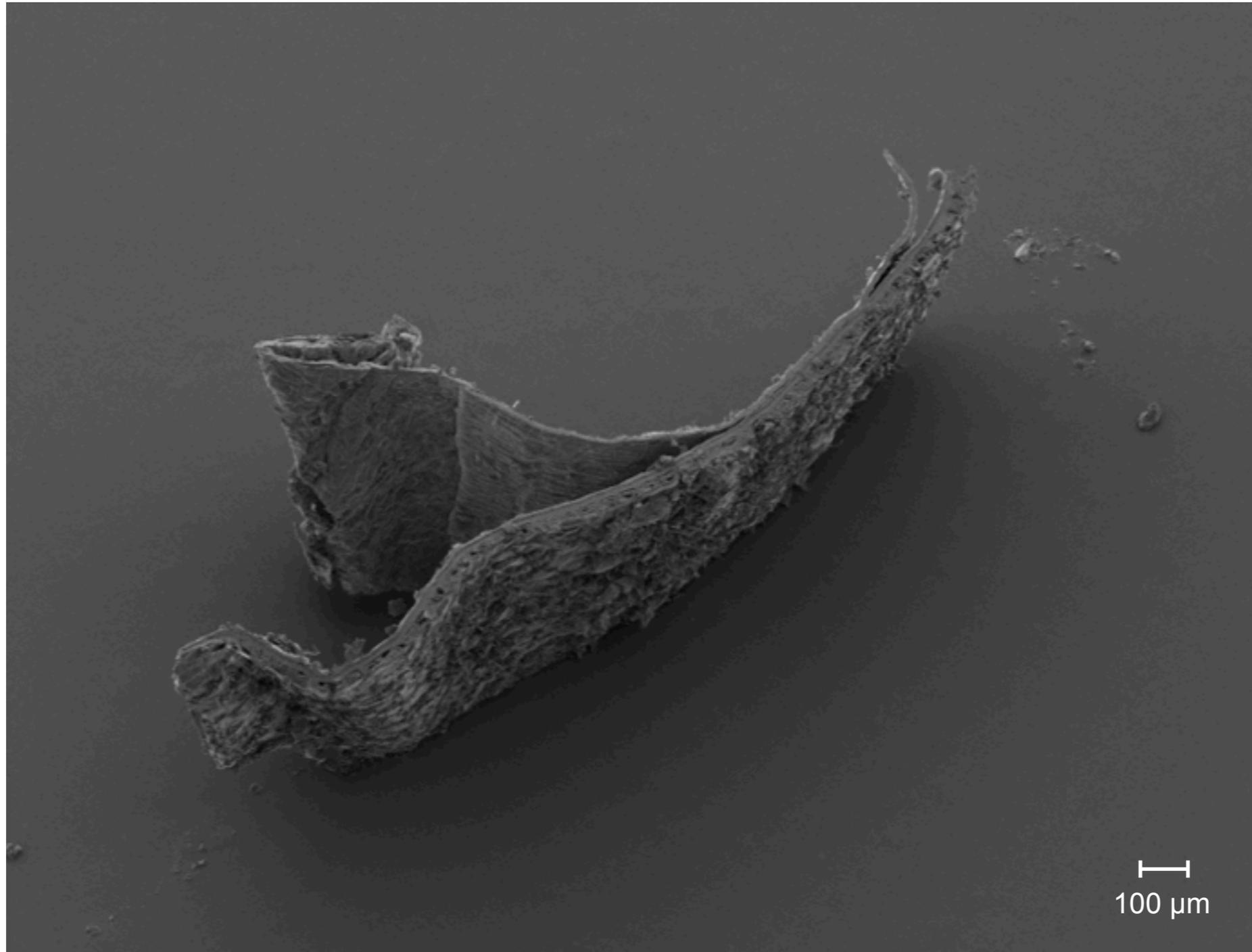


# Sample preparation

## Sample treatments

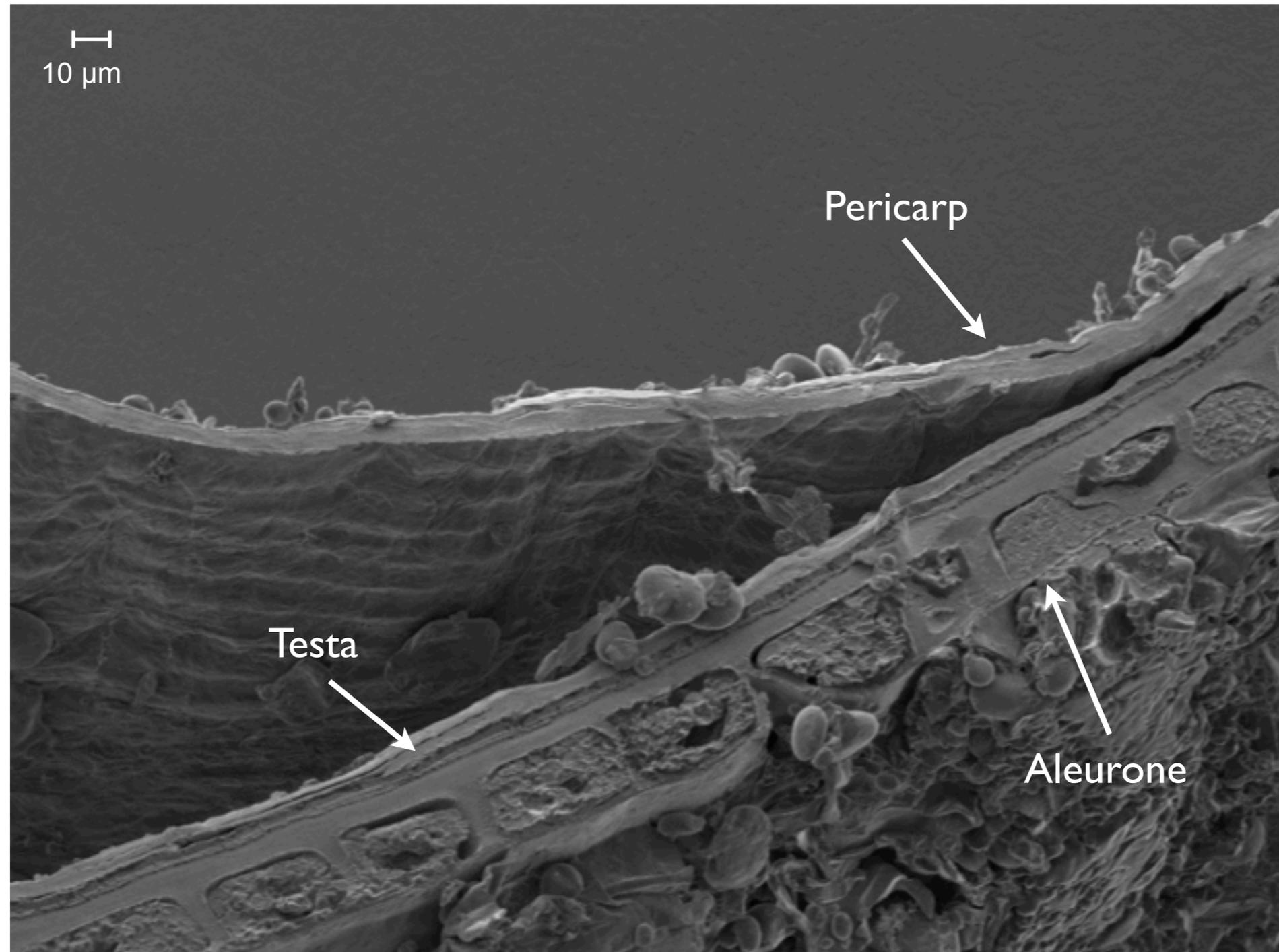
Mechanical	Chemical	Biological
Ball-milling	Acid	Fermentation
Extrusion	Lye	Enzymes
	Peroxide	

# Sample preparation



Scanning electron image of an untreated wheat bran flake

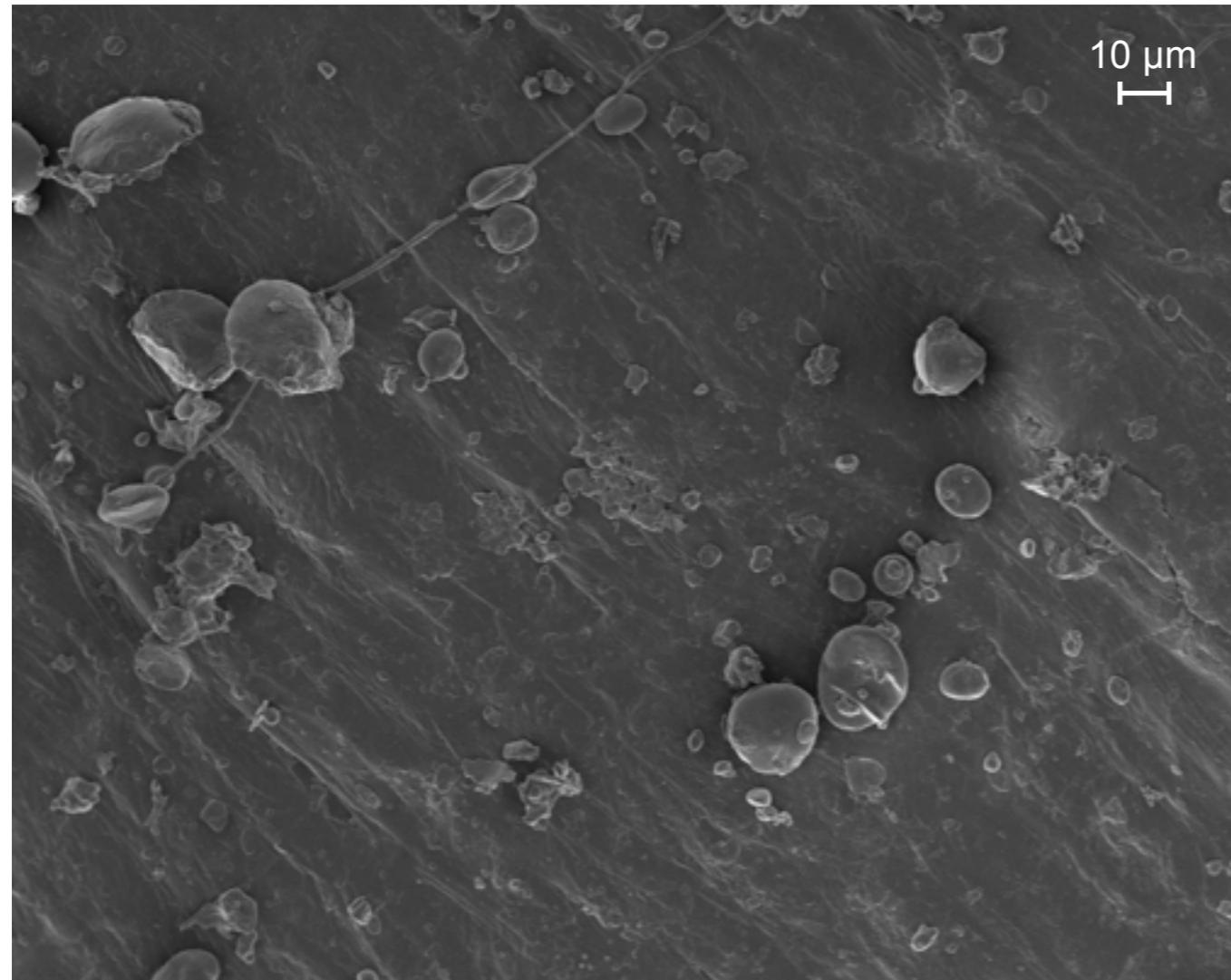
# Sample preparation



Scanning electron image of an untreated wheat bran flake  
(close-up)

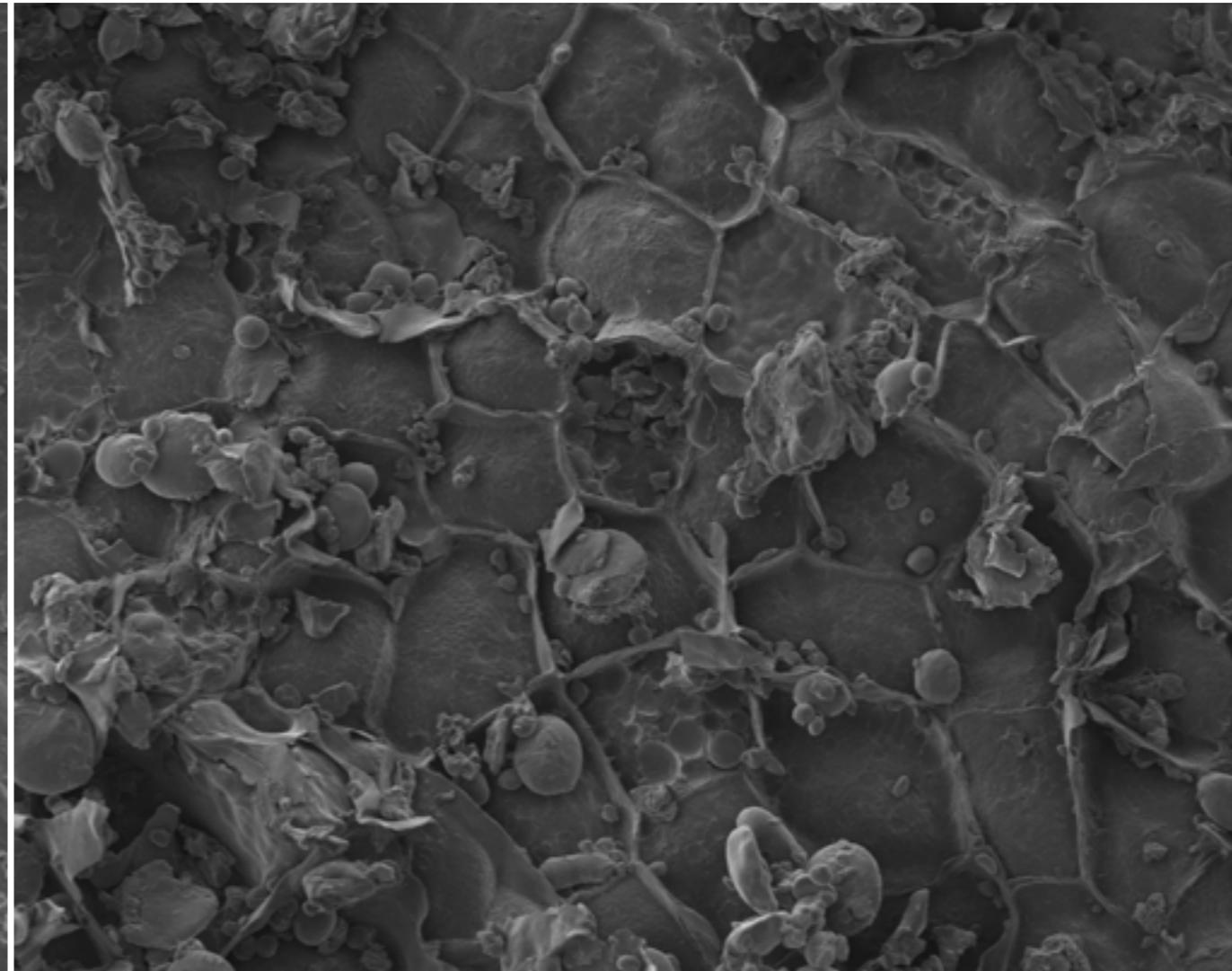
# Sample preparation

Top-view



**Pericarp**

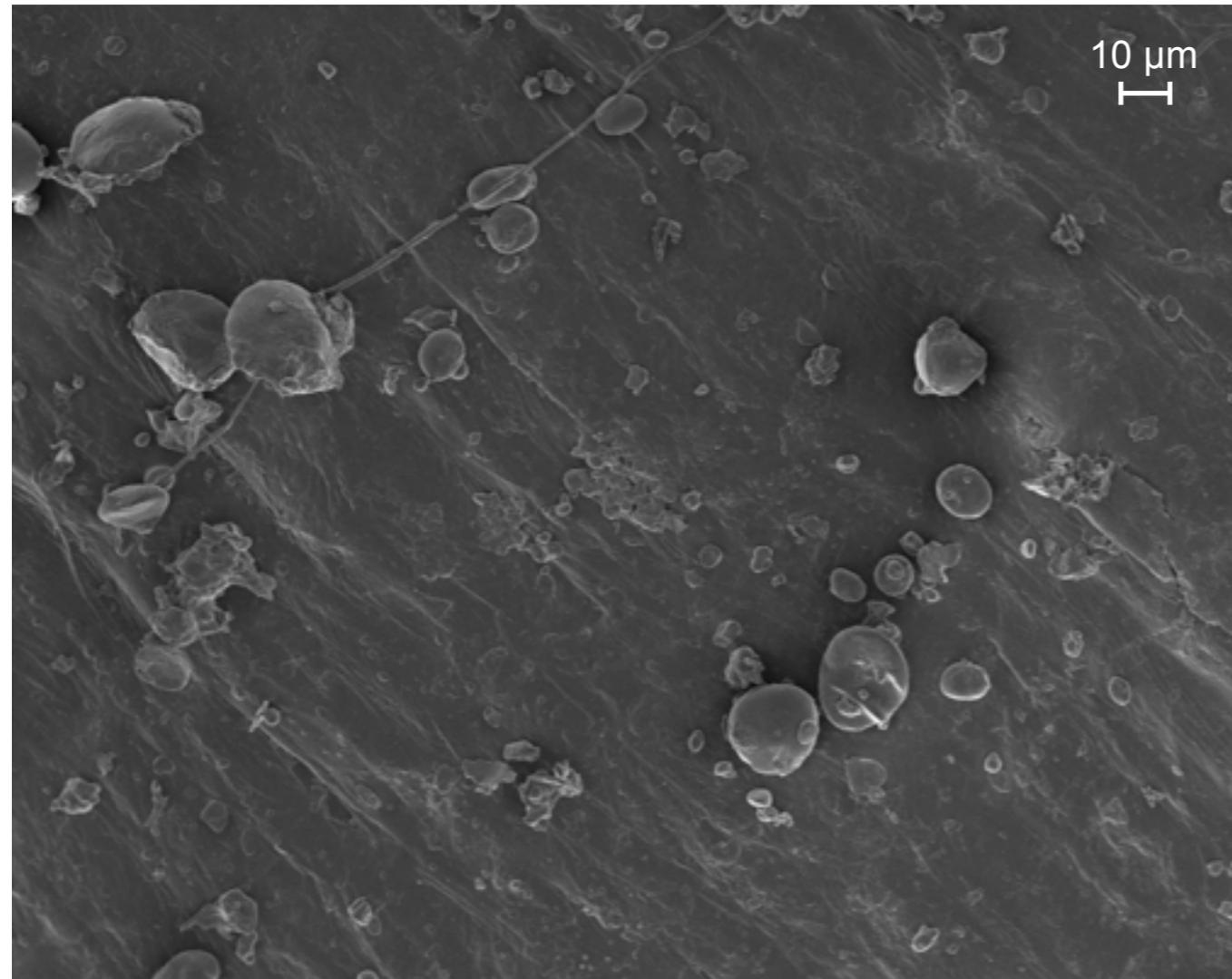
Bottom-view



**Aleurone**

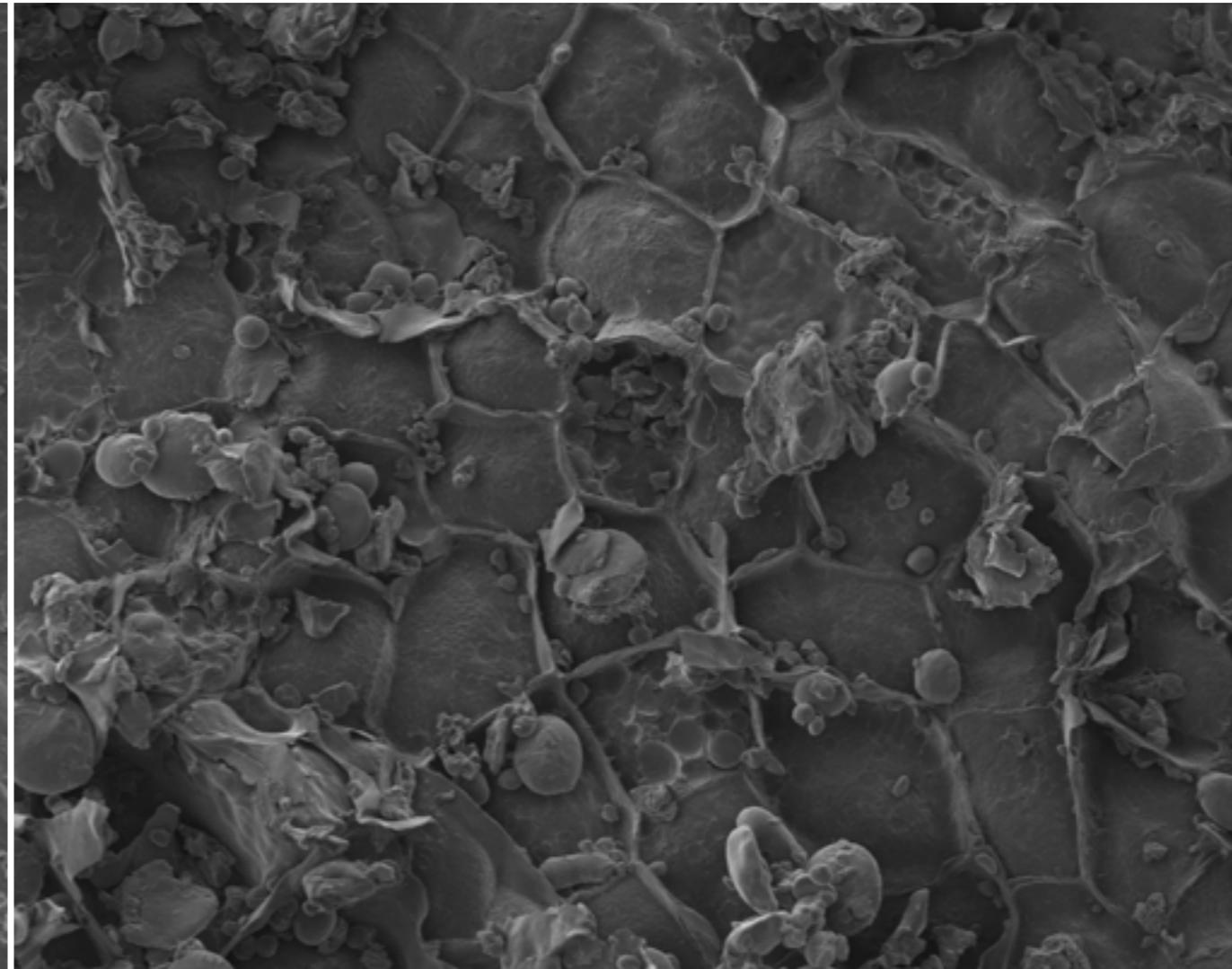
# Sample preparation

Top-view



Pericarp

Bottom-view



Aleurone

Very limited information

# Sample preparation



Bran flakes embedded in paraffin wax

# Sample preparation



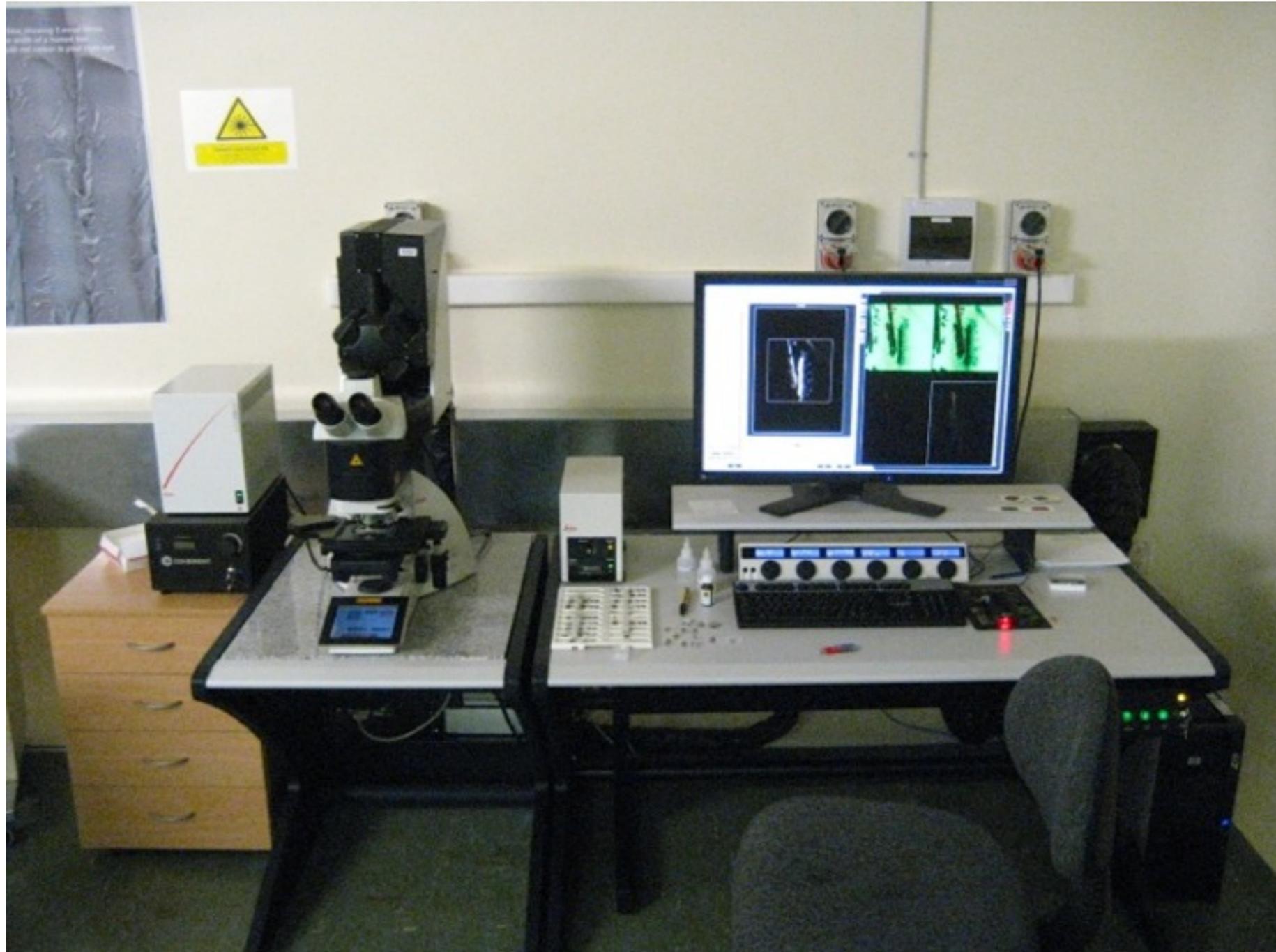
Bran flakes embedded in paraffin wax

# Sample preparation



Wax blocks mounted on slides

# Sample preparation



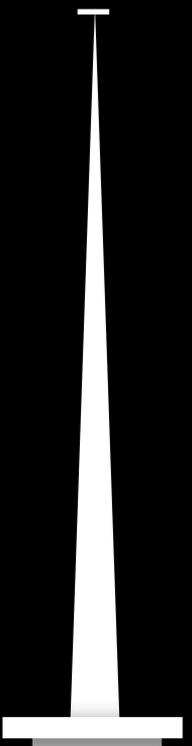
**Confocal laser scanning microscope**  
Leica SP5 MA II

Auto-  
fluorescence

488 nm

561 nm

Auto-  
fluorescence



Alexa647  
label

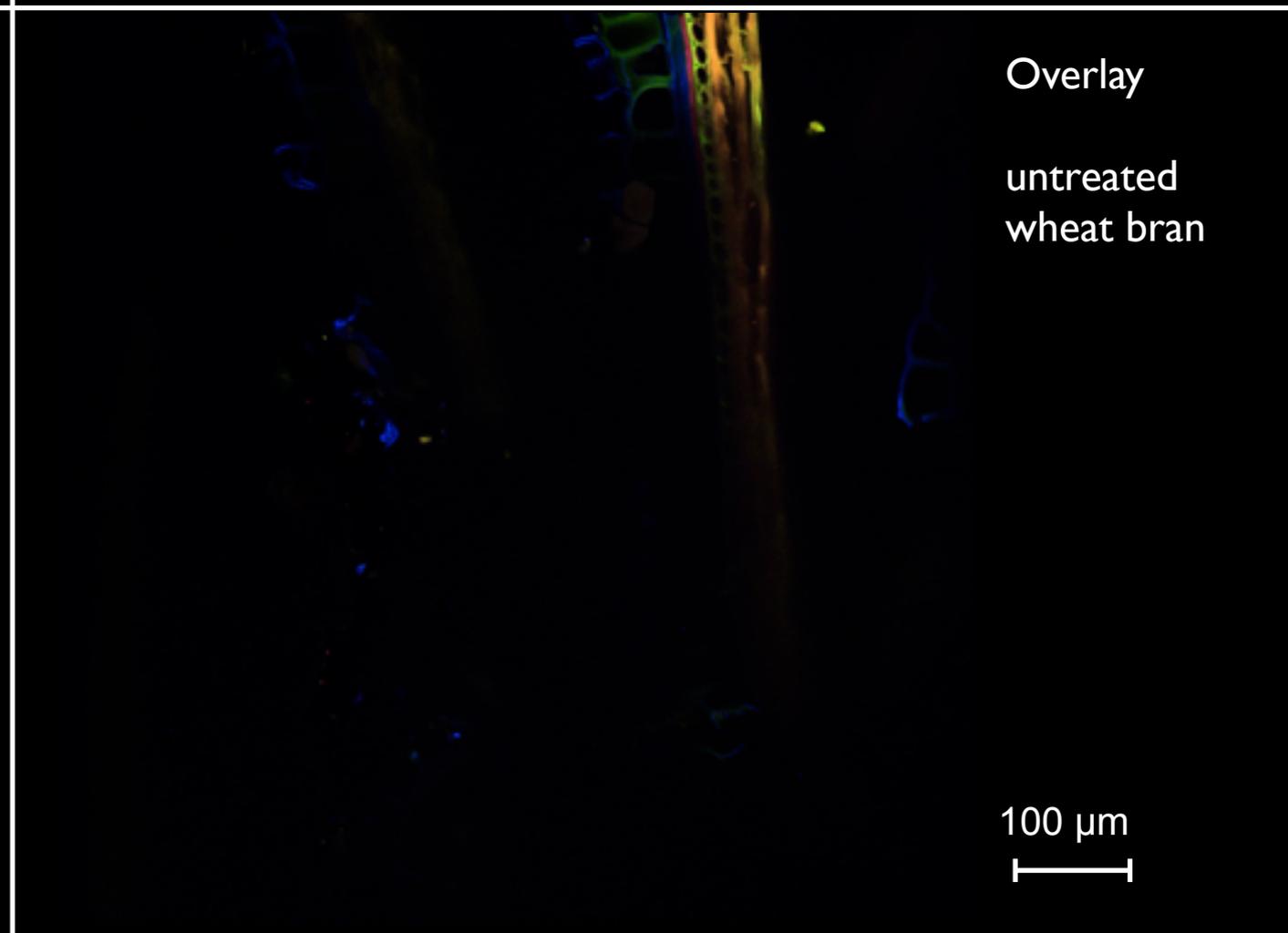
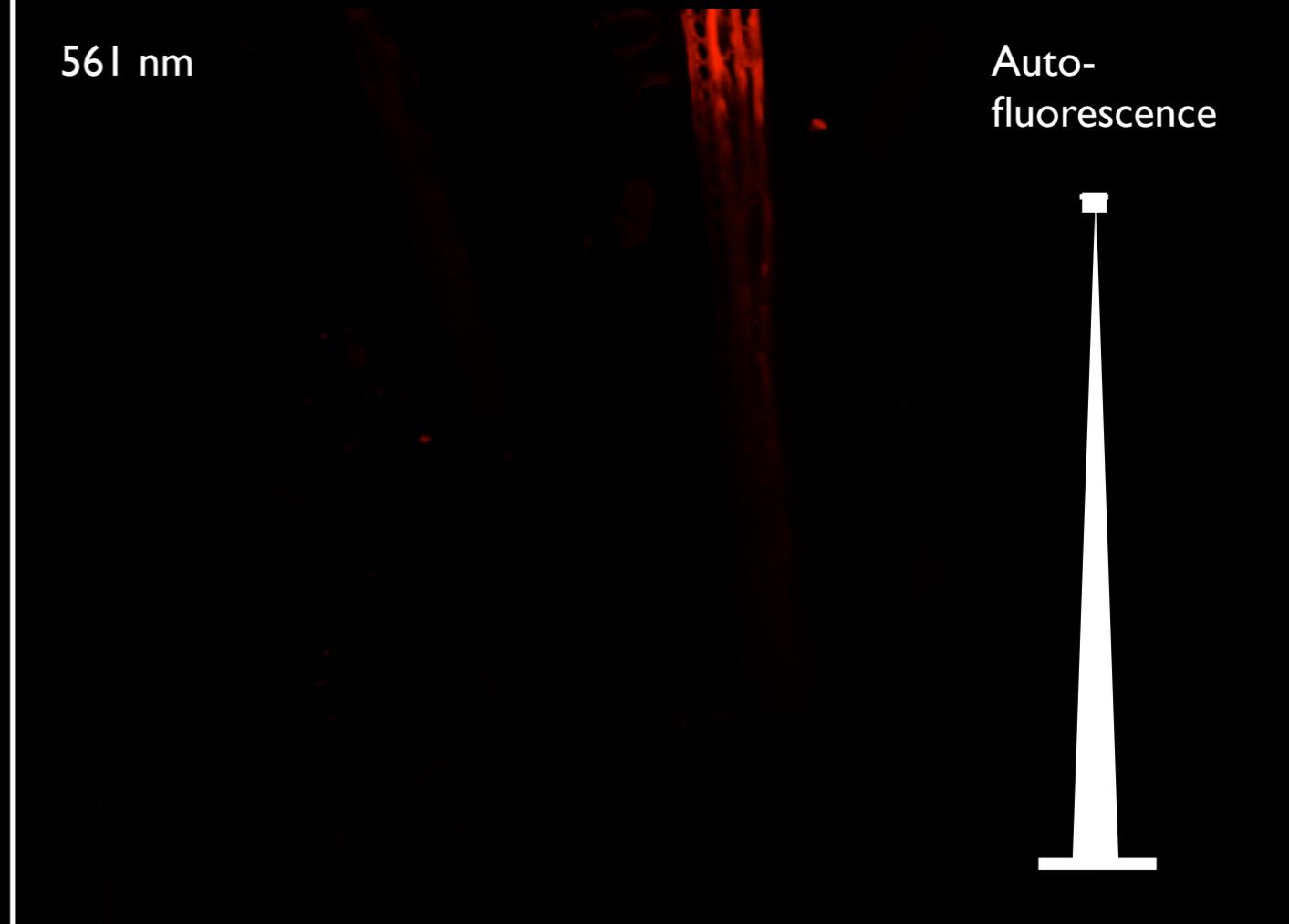
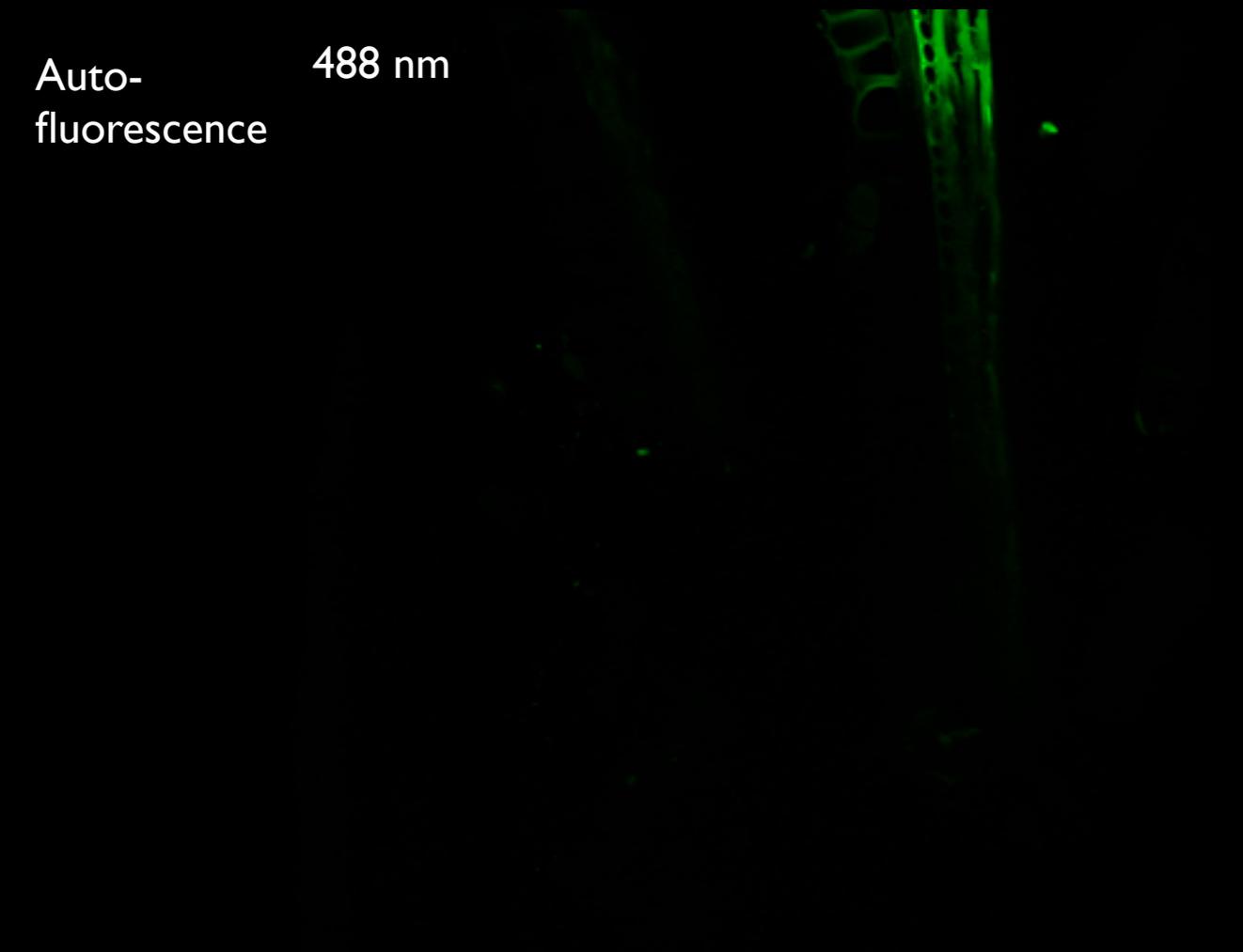
633 nm

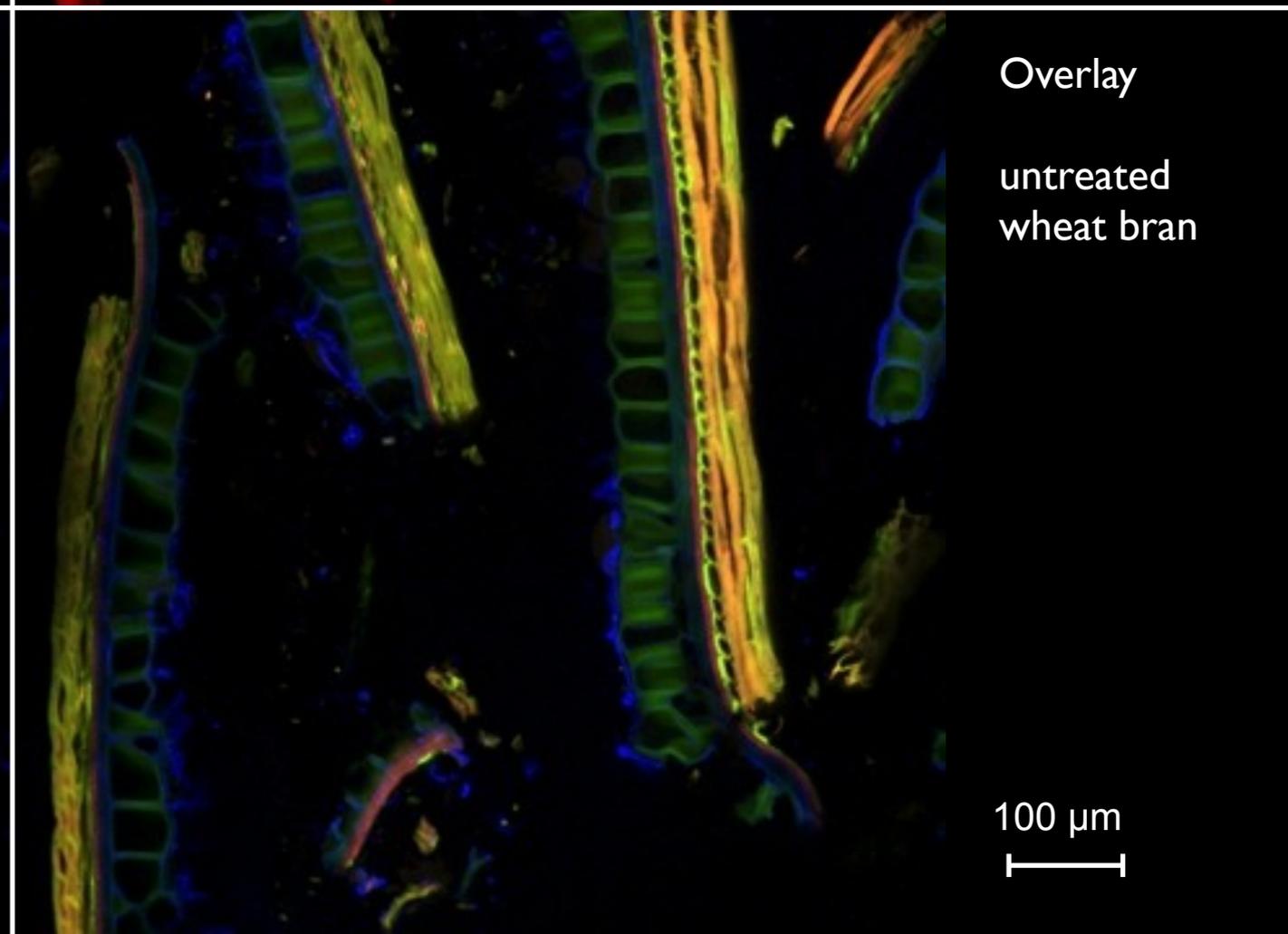
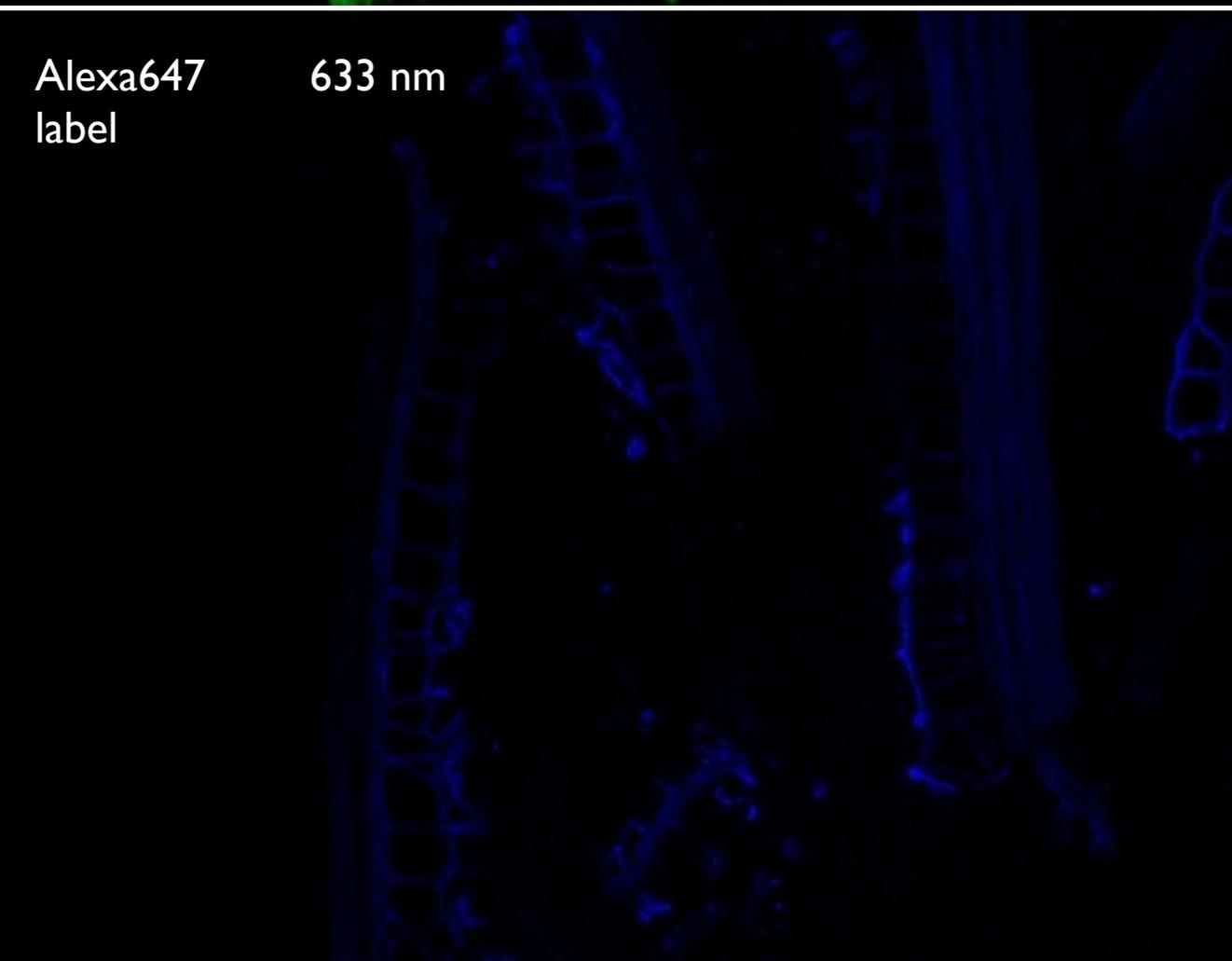
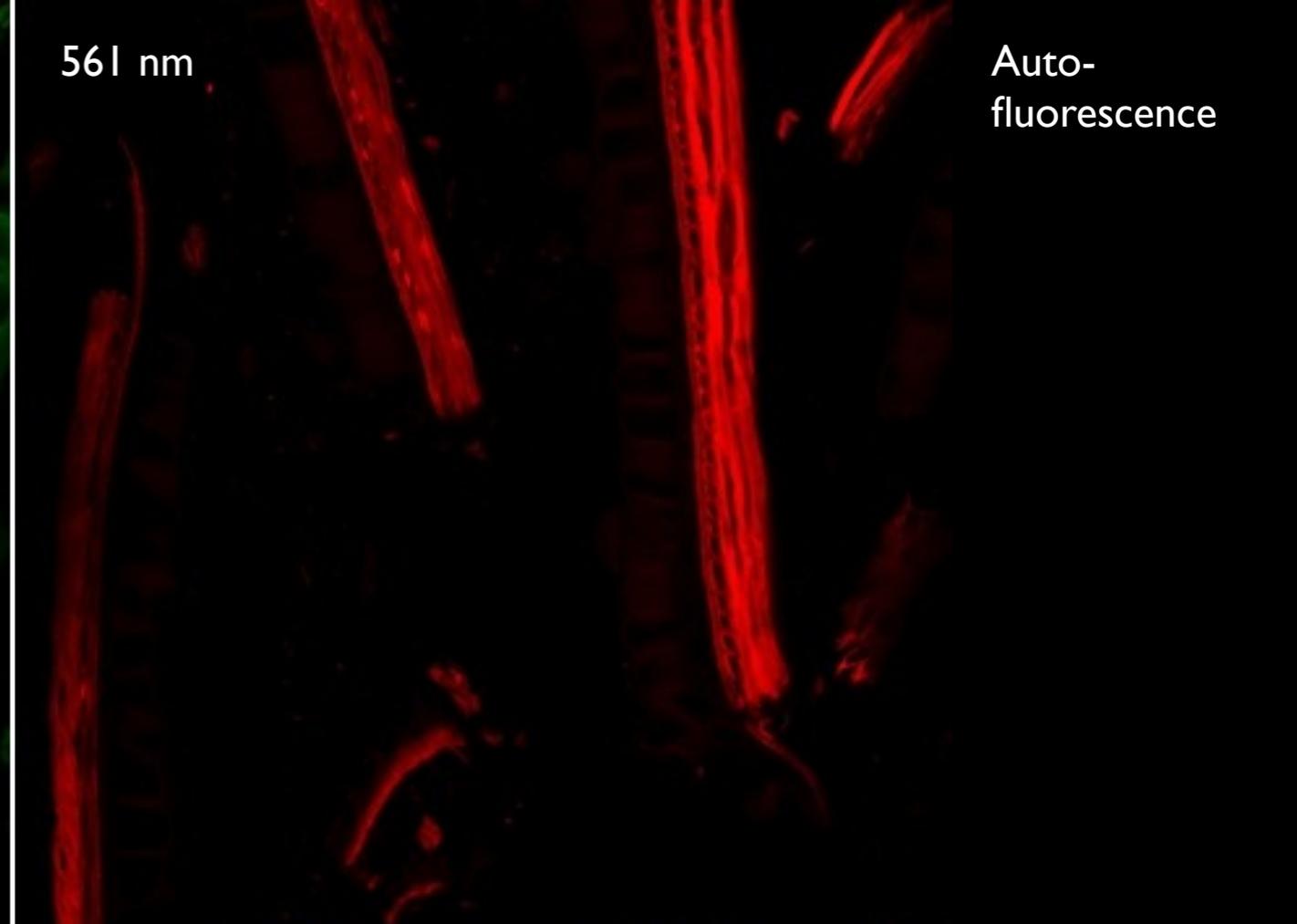
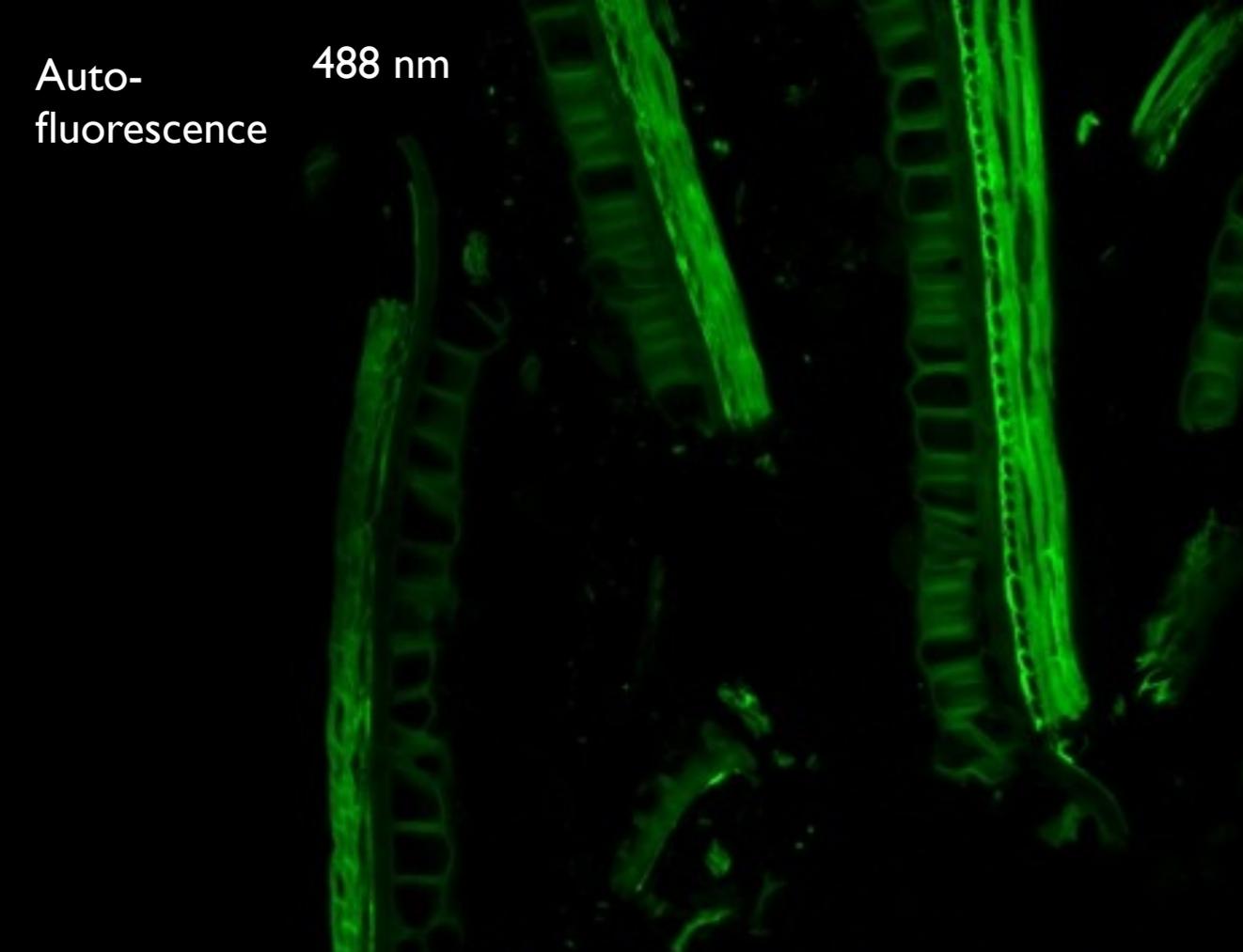
Overlay

untreated  
wheat bran

100  $\mu$ m

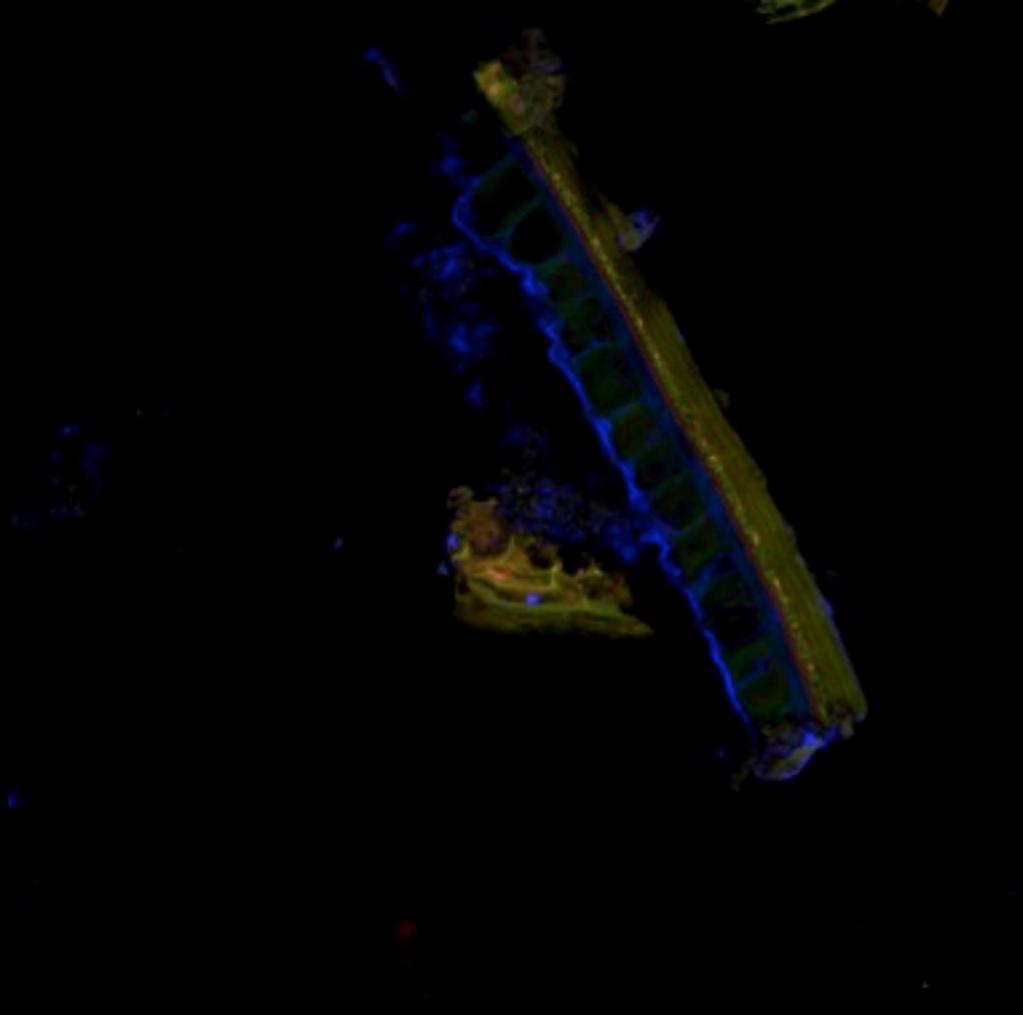






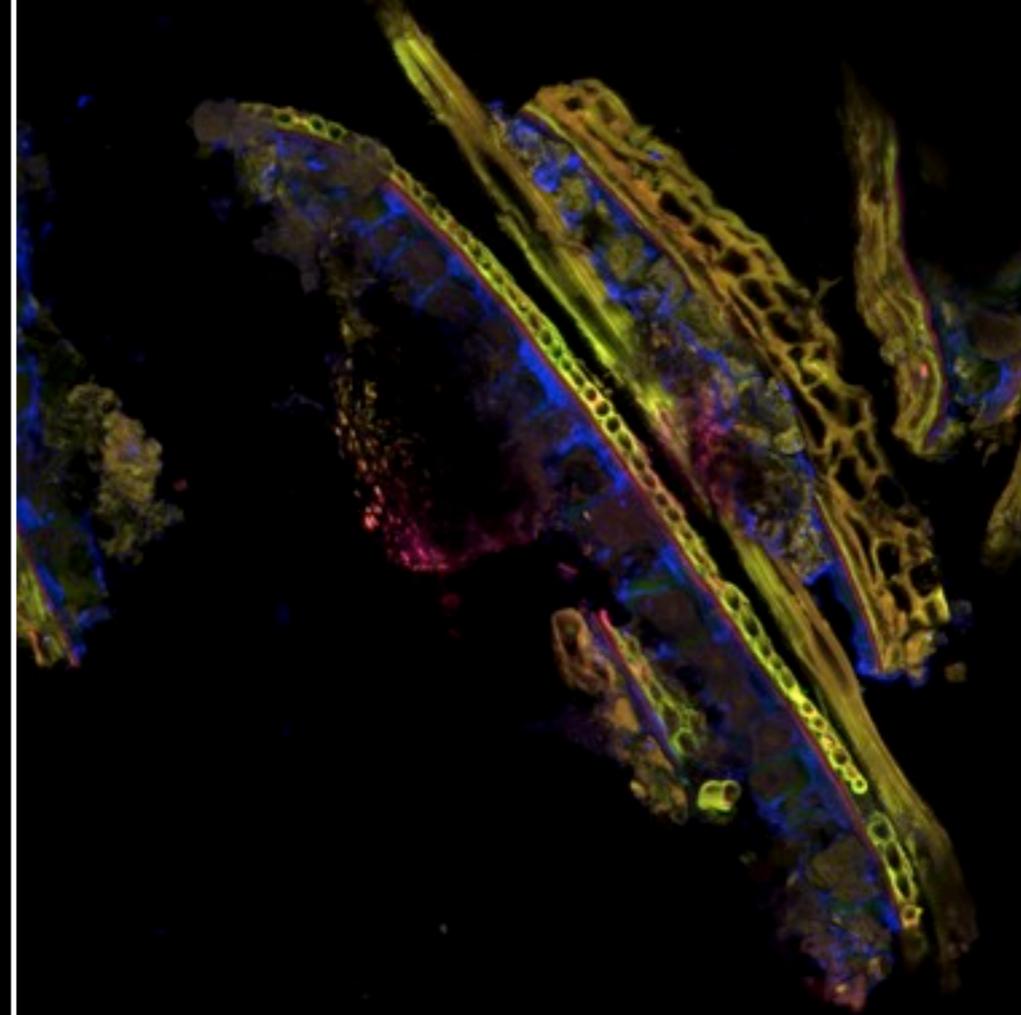
Esterase

1% (w/w)  
RT  
5 days



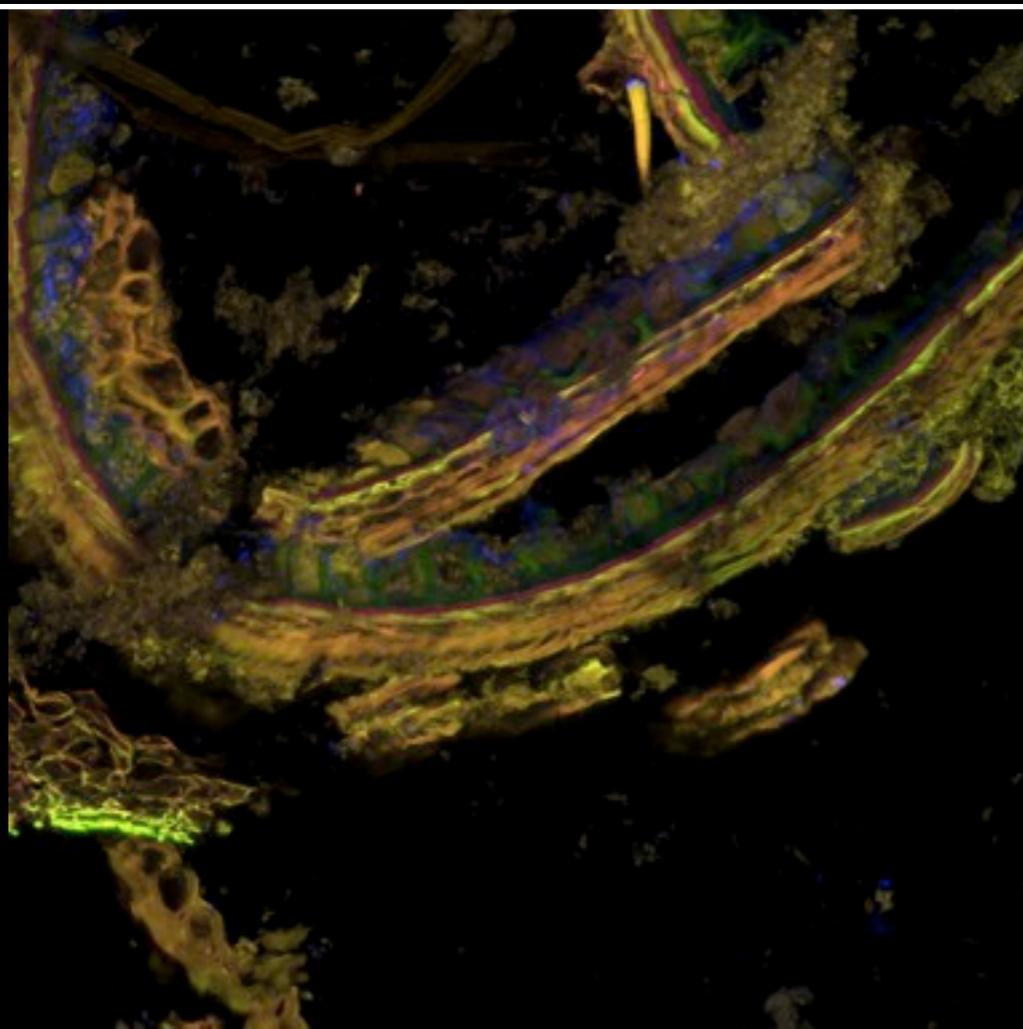
Xylanase

1% (w/w)  
RT  
5 days



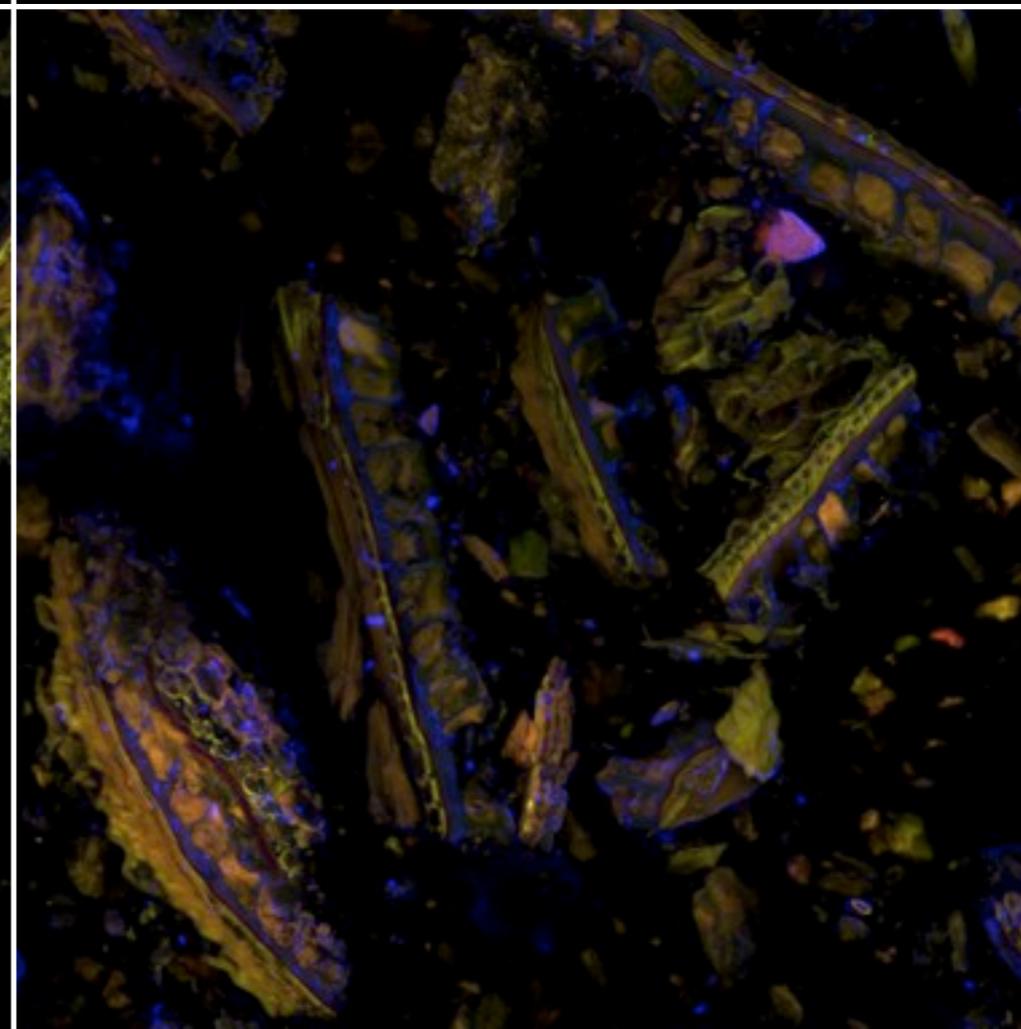
Esterase  
+Xylanase

1% (w/w)  
RT  
5 days



Cellulase  
+Xylanase

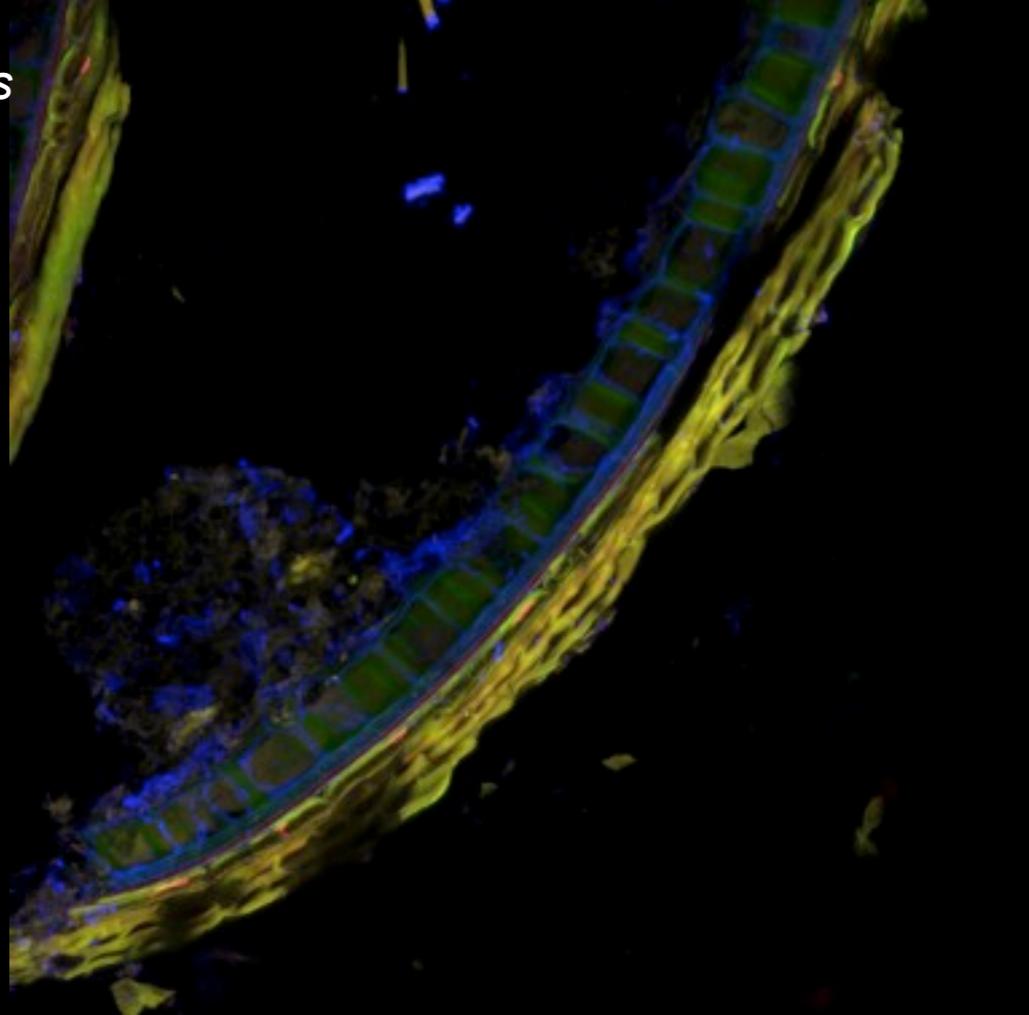
1% (w/w)  
RT  
5 days



100  $\mu$ m  
└───┘

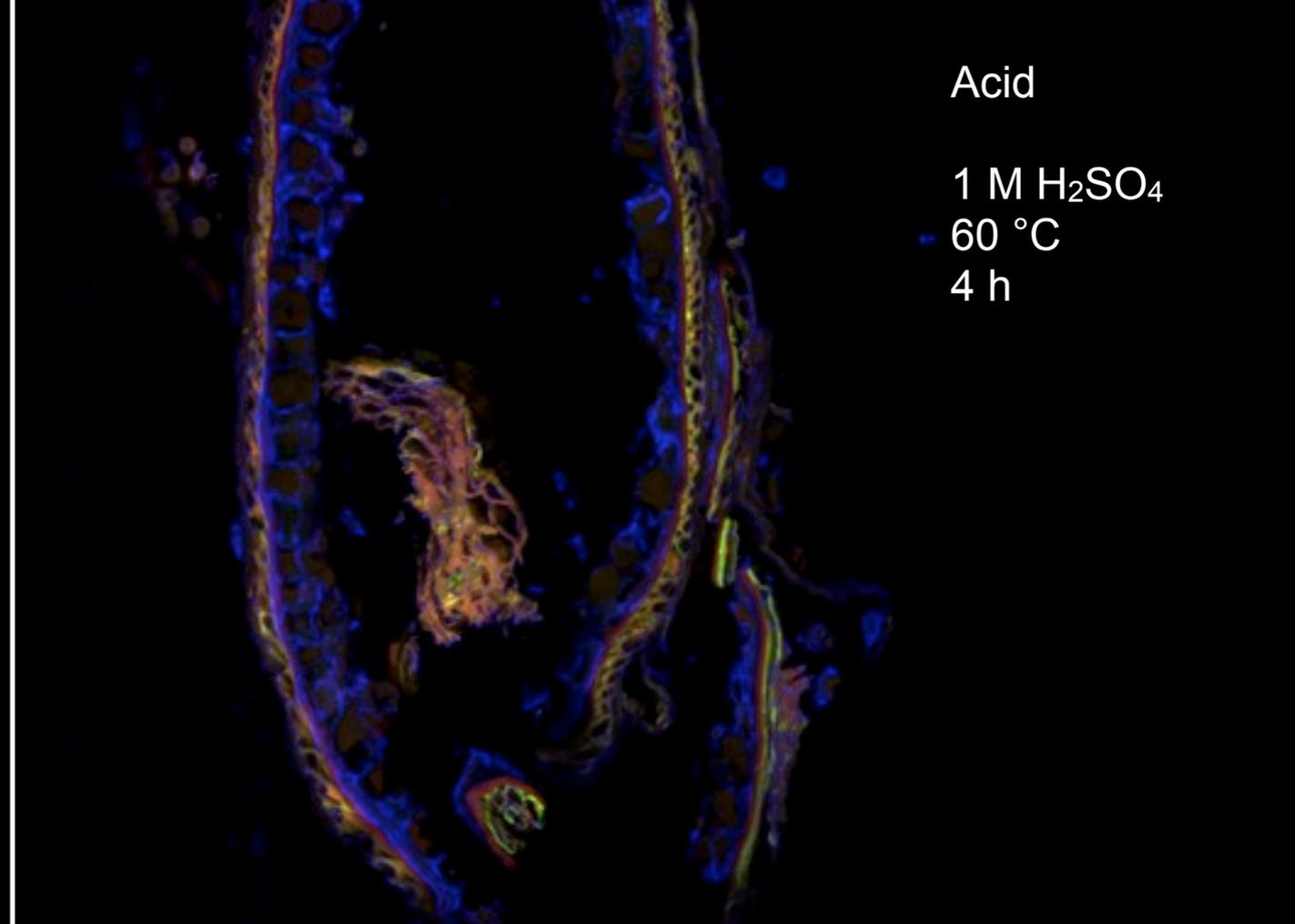
*Lactobacillus plantarum*

2% (w/w)  
RT  
5 days



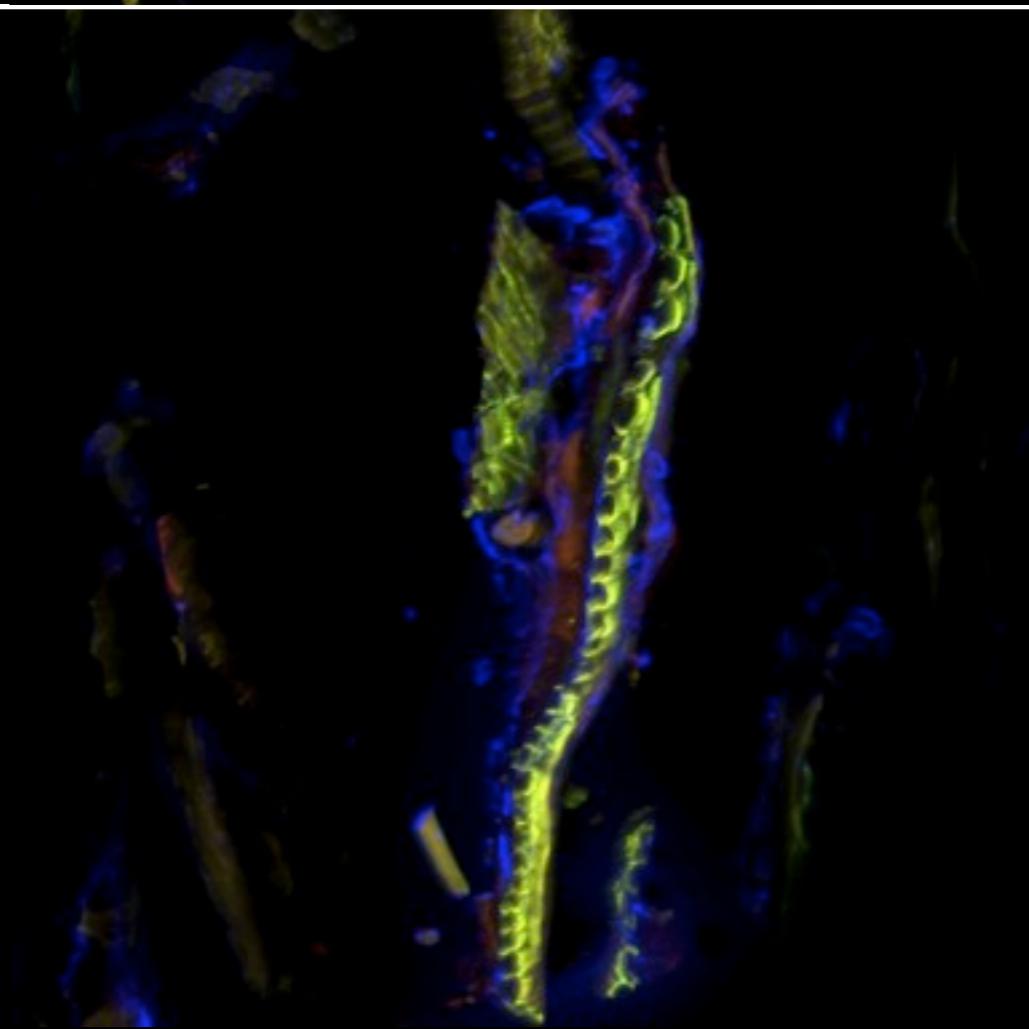
Acid

1 M H<sub>2</sub>SO<sub>4</sub>  
60 °C  
4 h



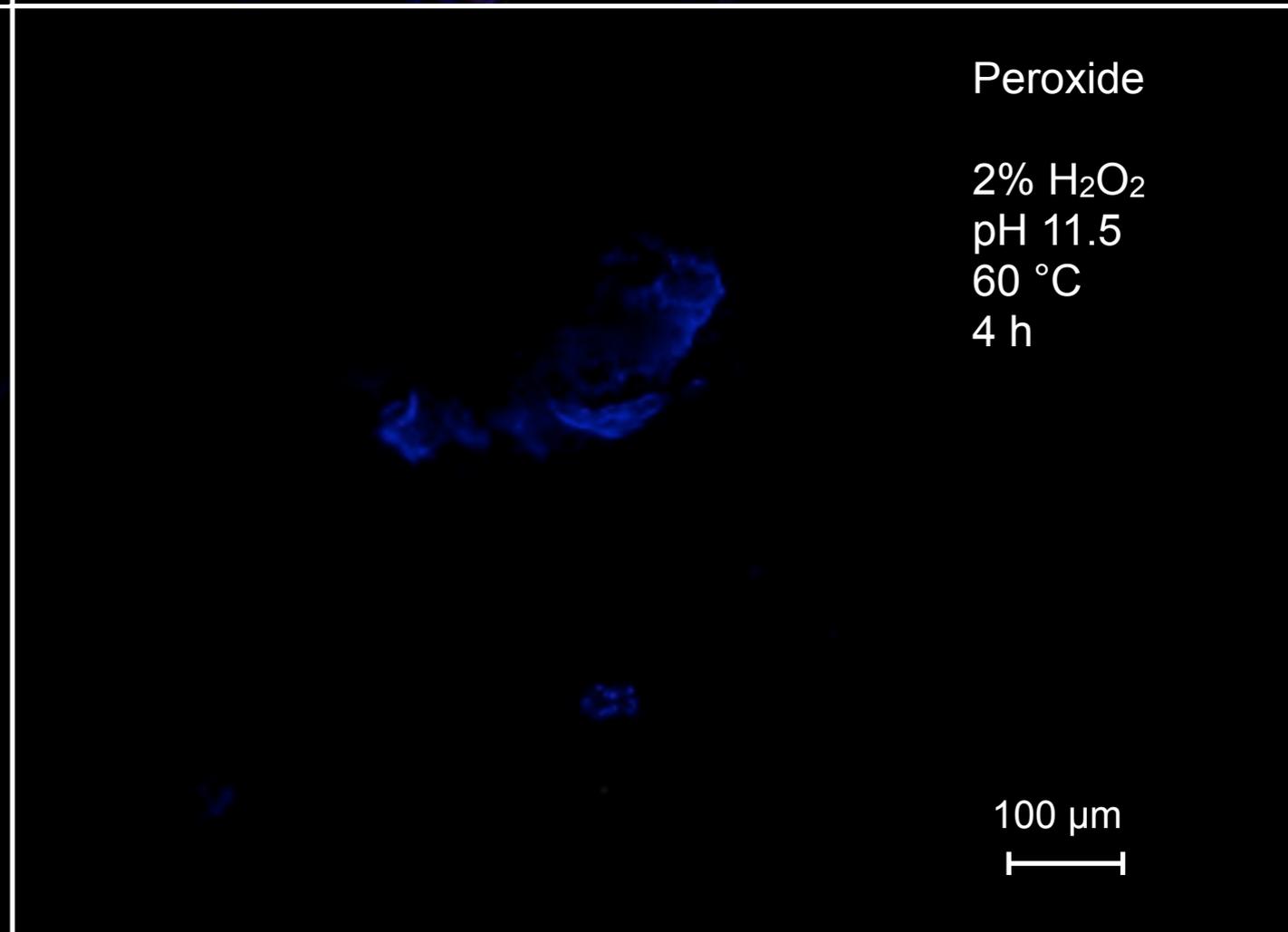
Lye

1 M NaOH  
60 °C  
4 h

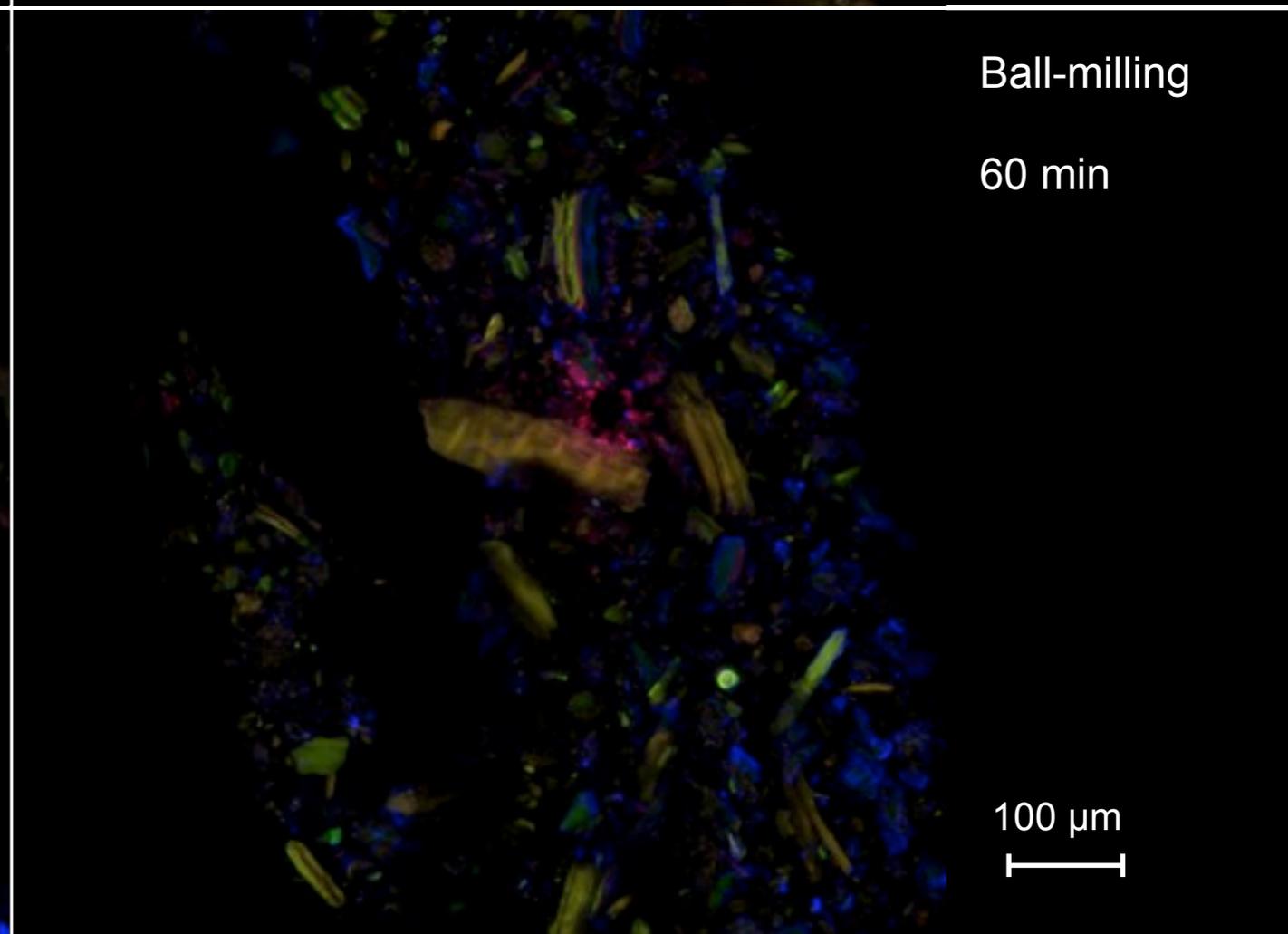
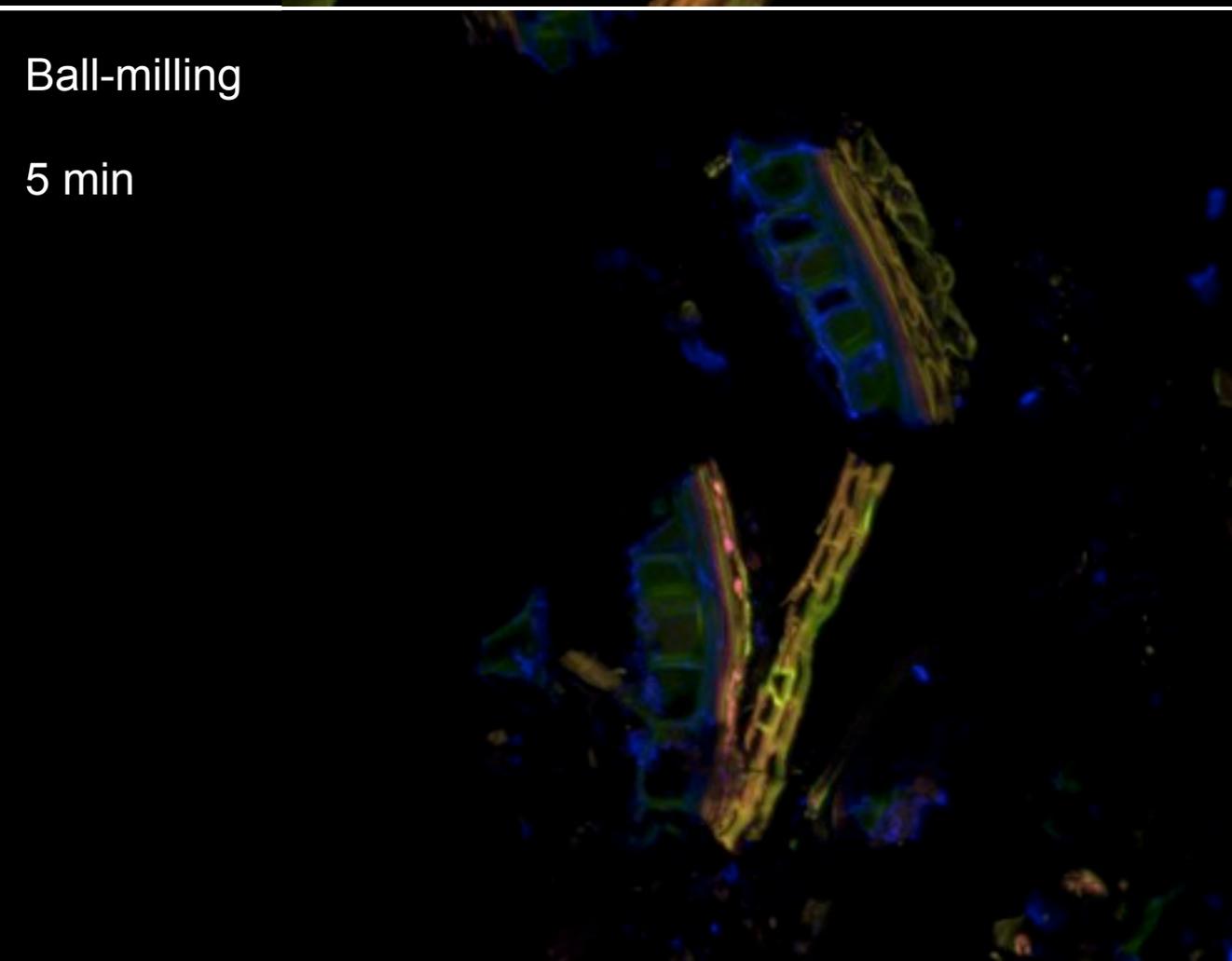
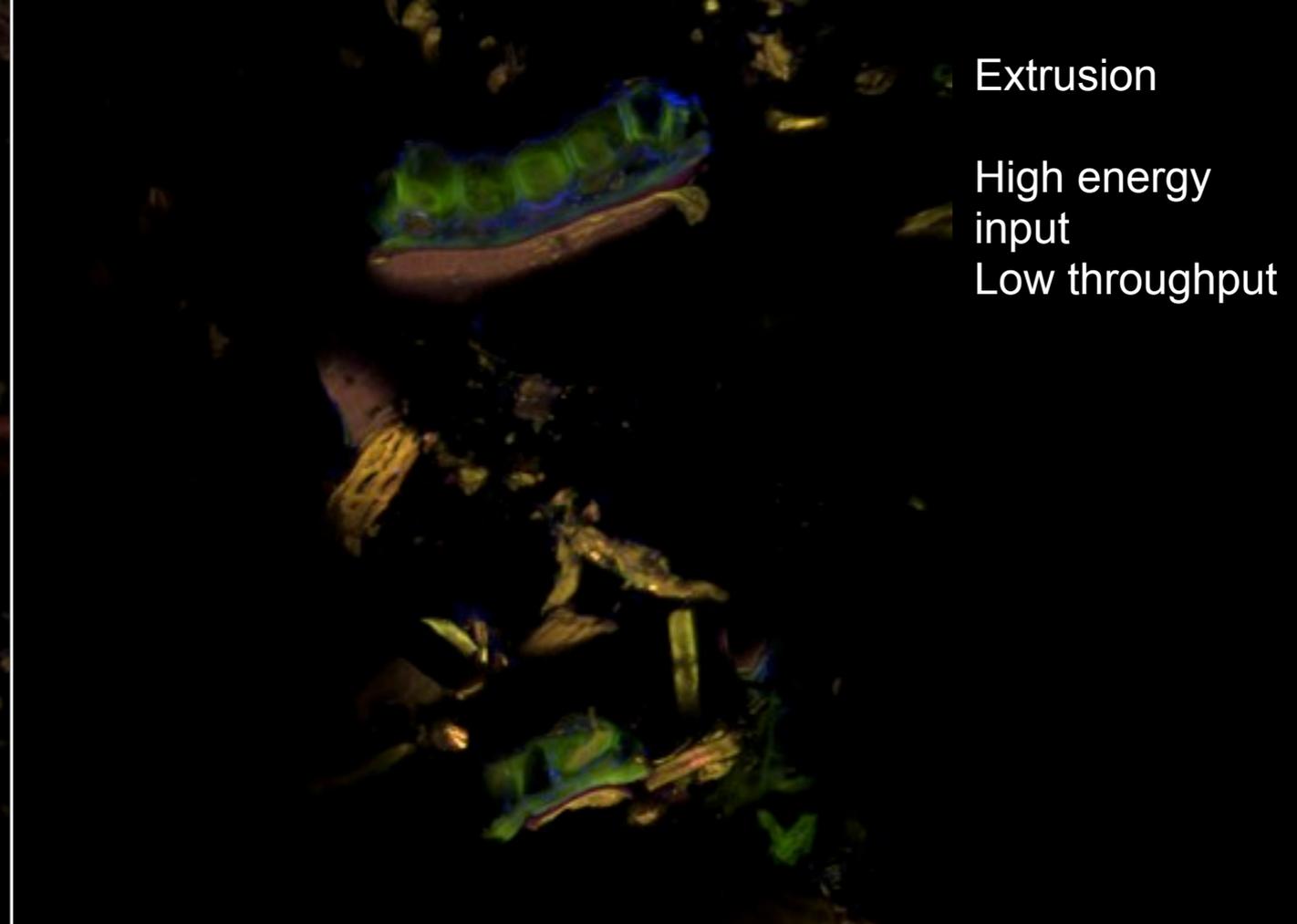
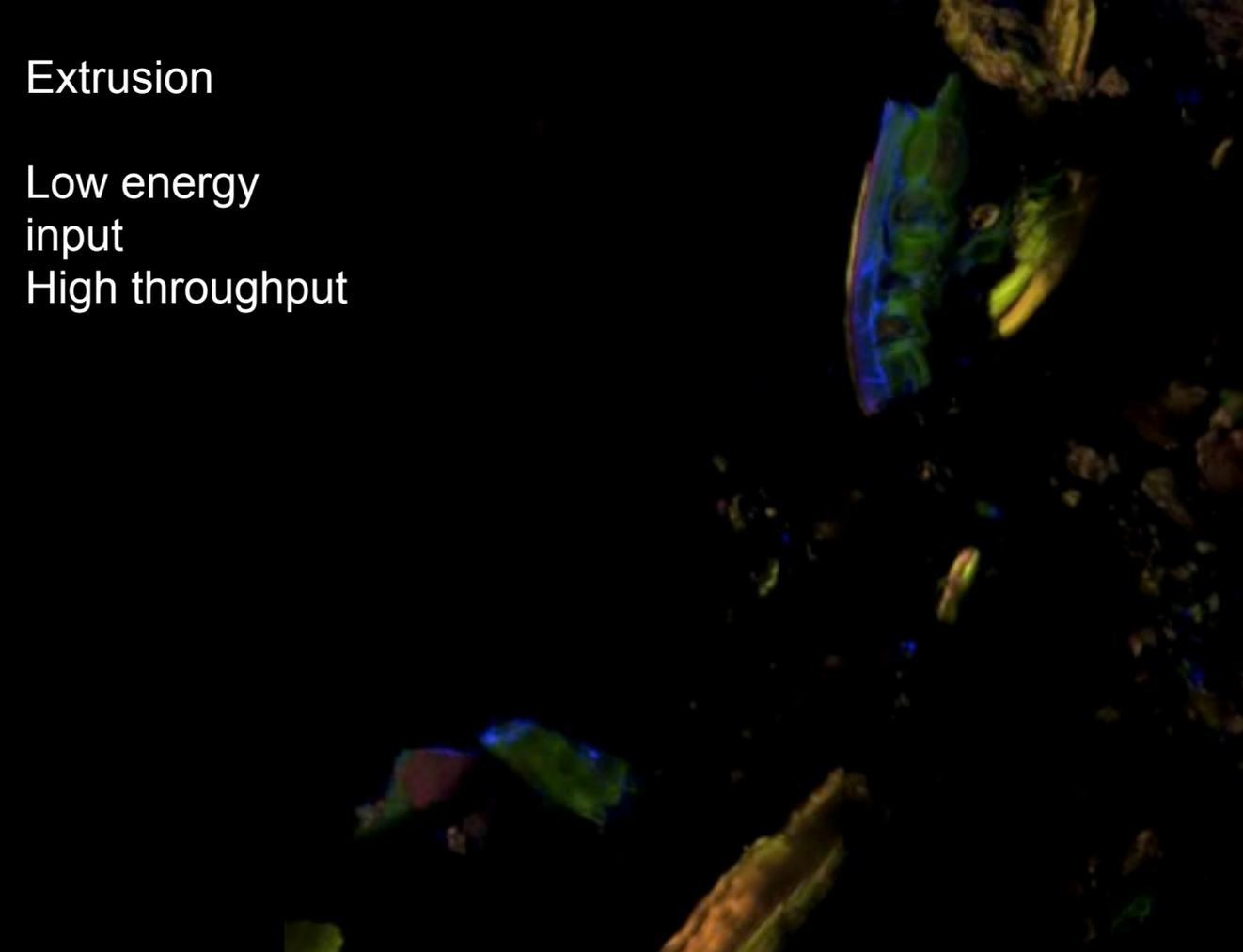


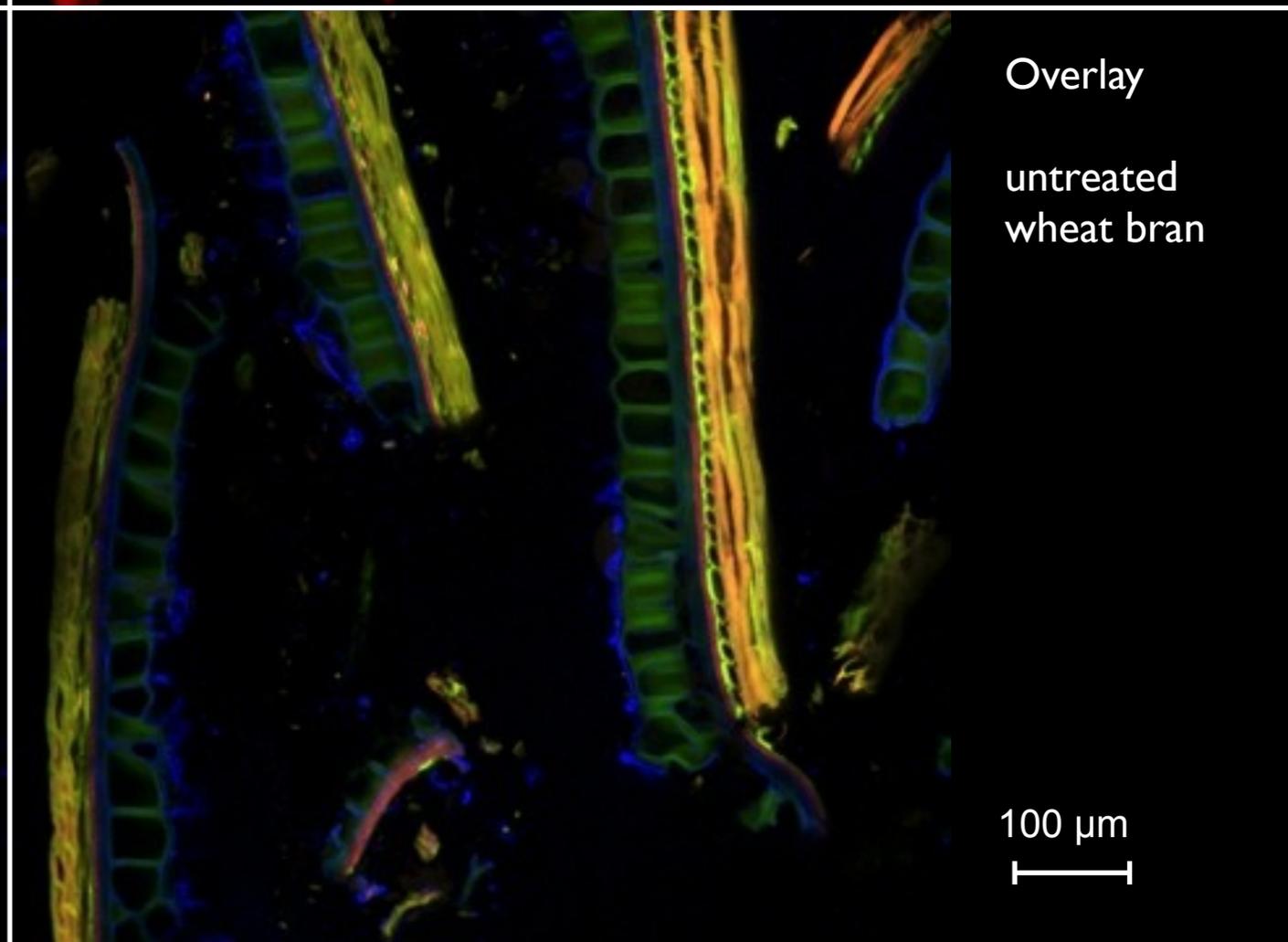
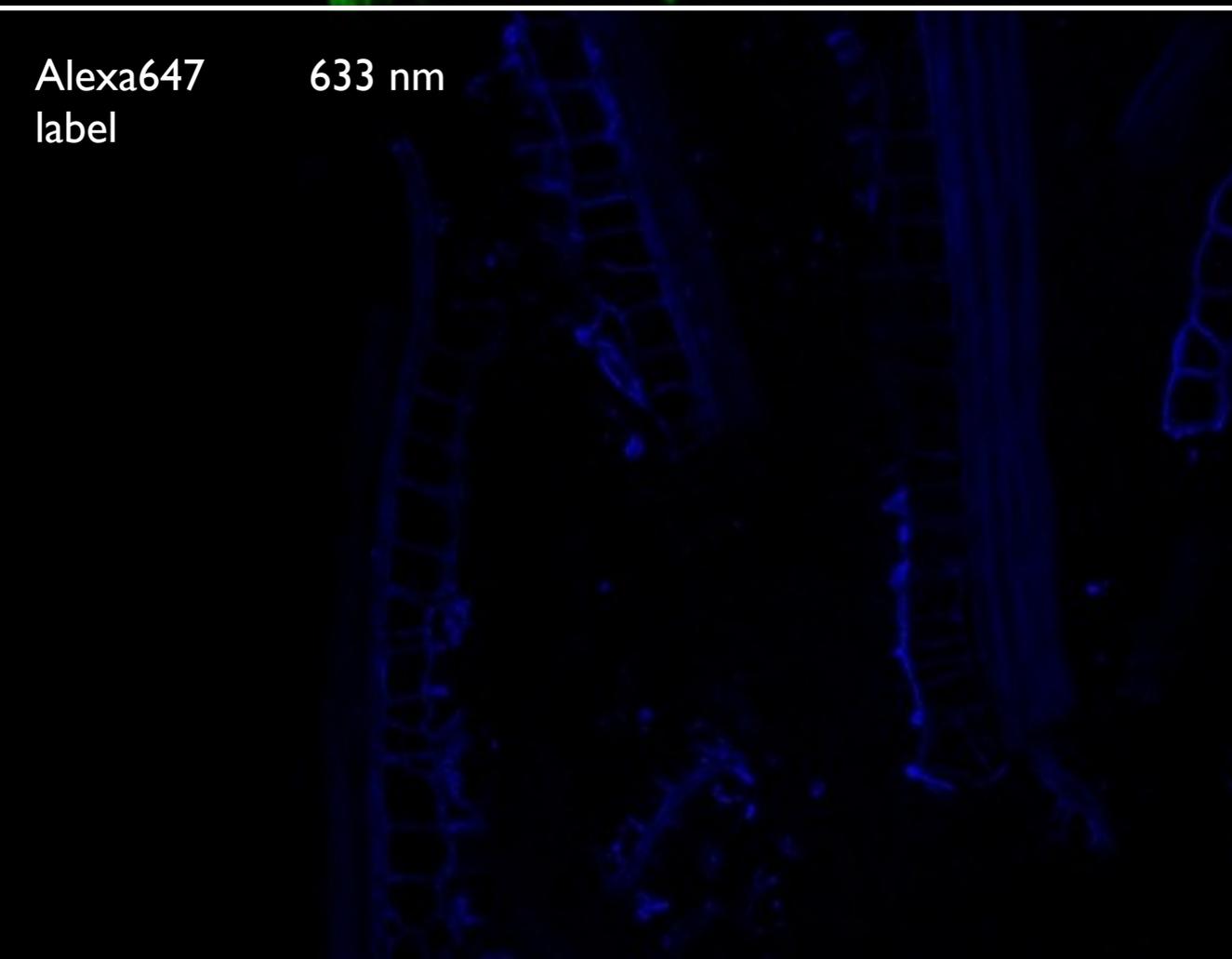
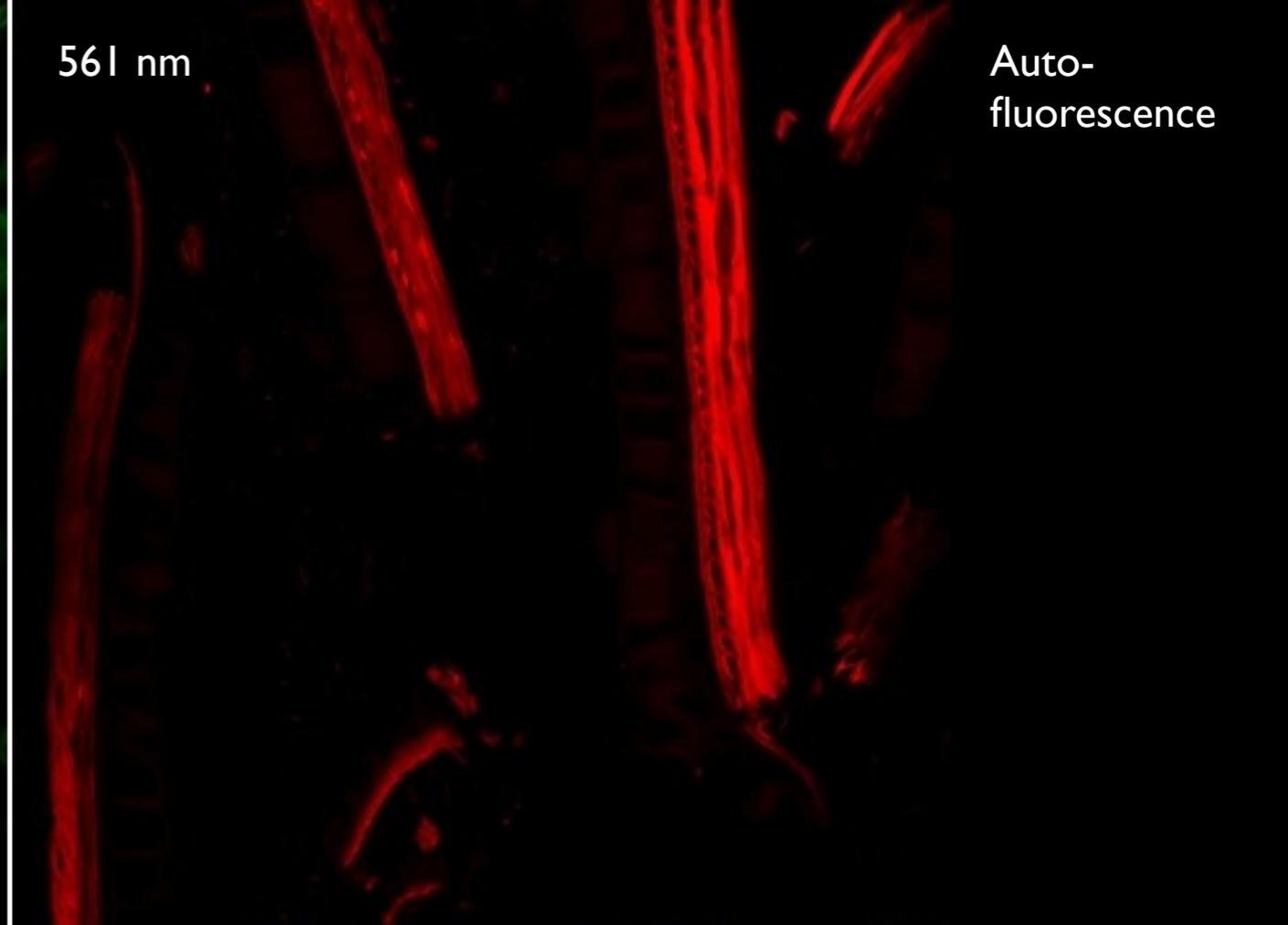
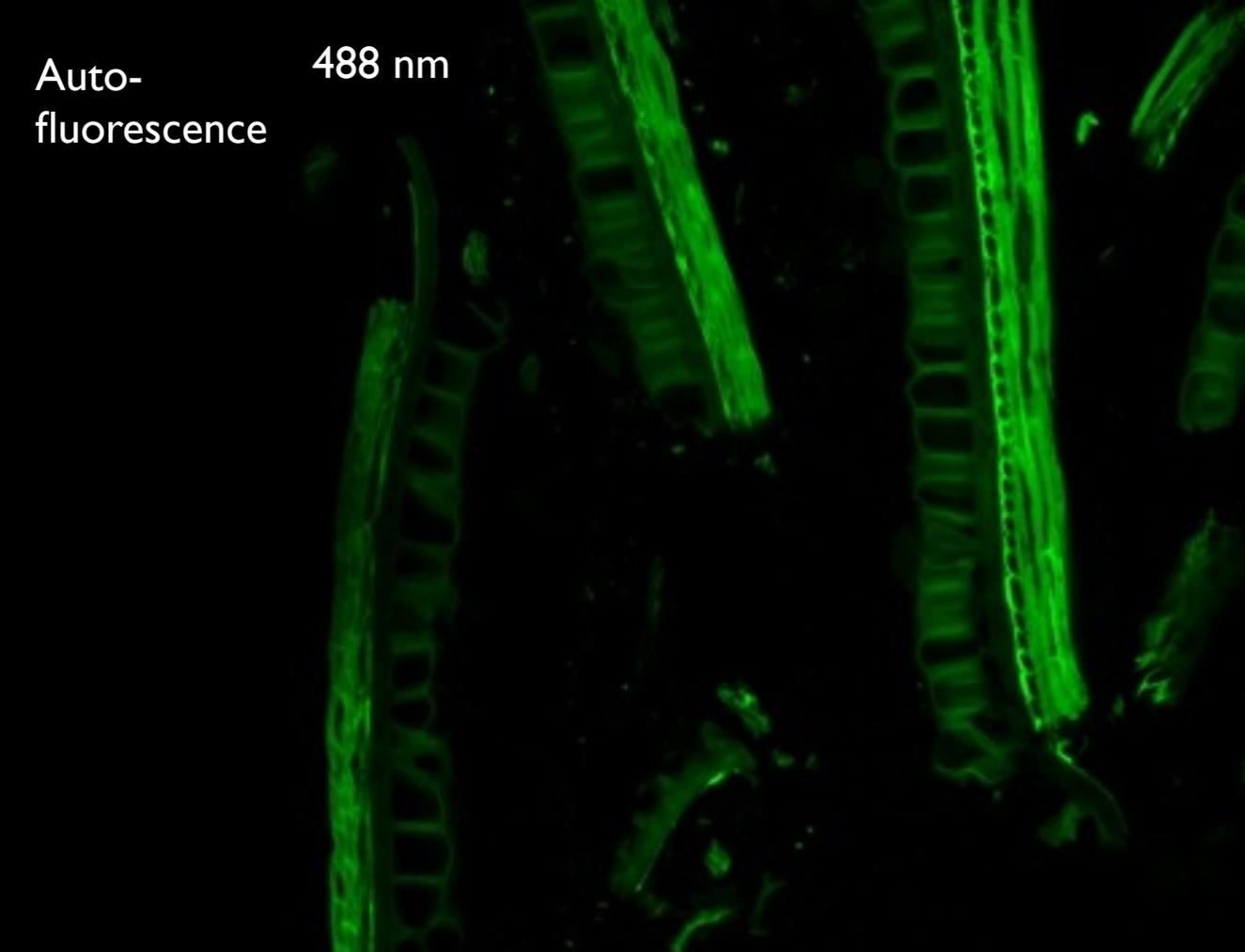
Peroxide

2% H<sub>2</sub>O<sub>2</sub>  
pH 11.5  
60 °C  
4 h



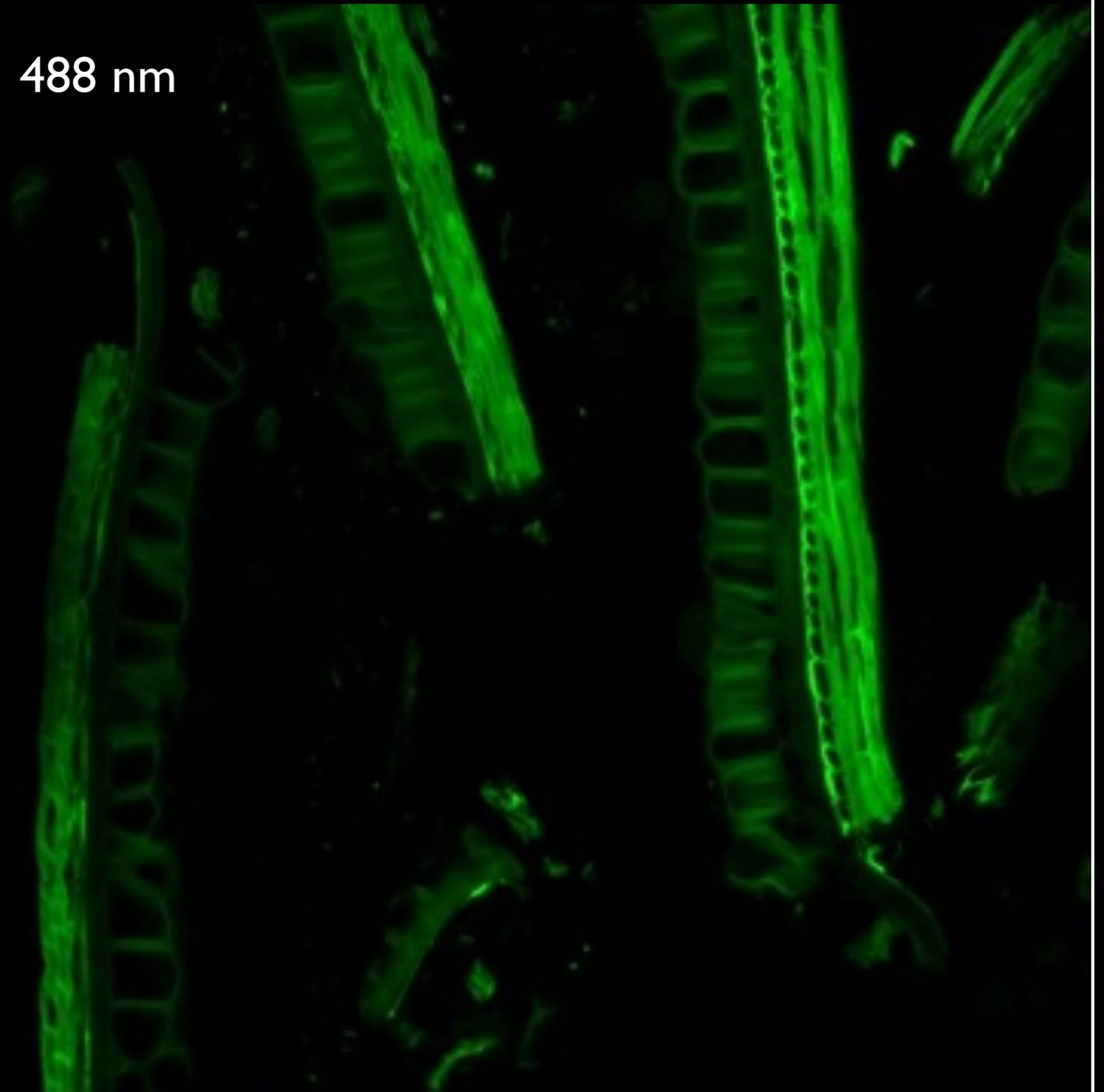
100 μm  
└───┘





Auto-  
fluorescence

488 nm



Auto-  
fluorescence

488 nm

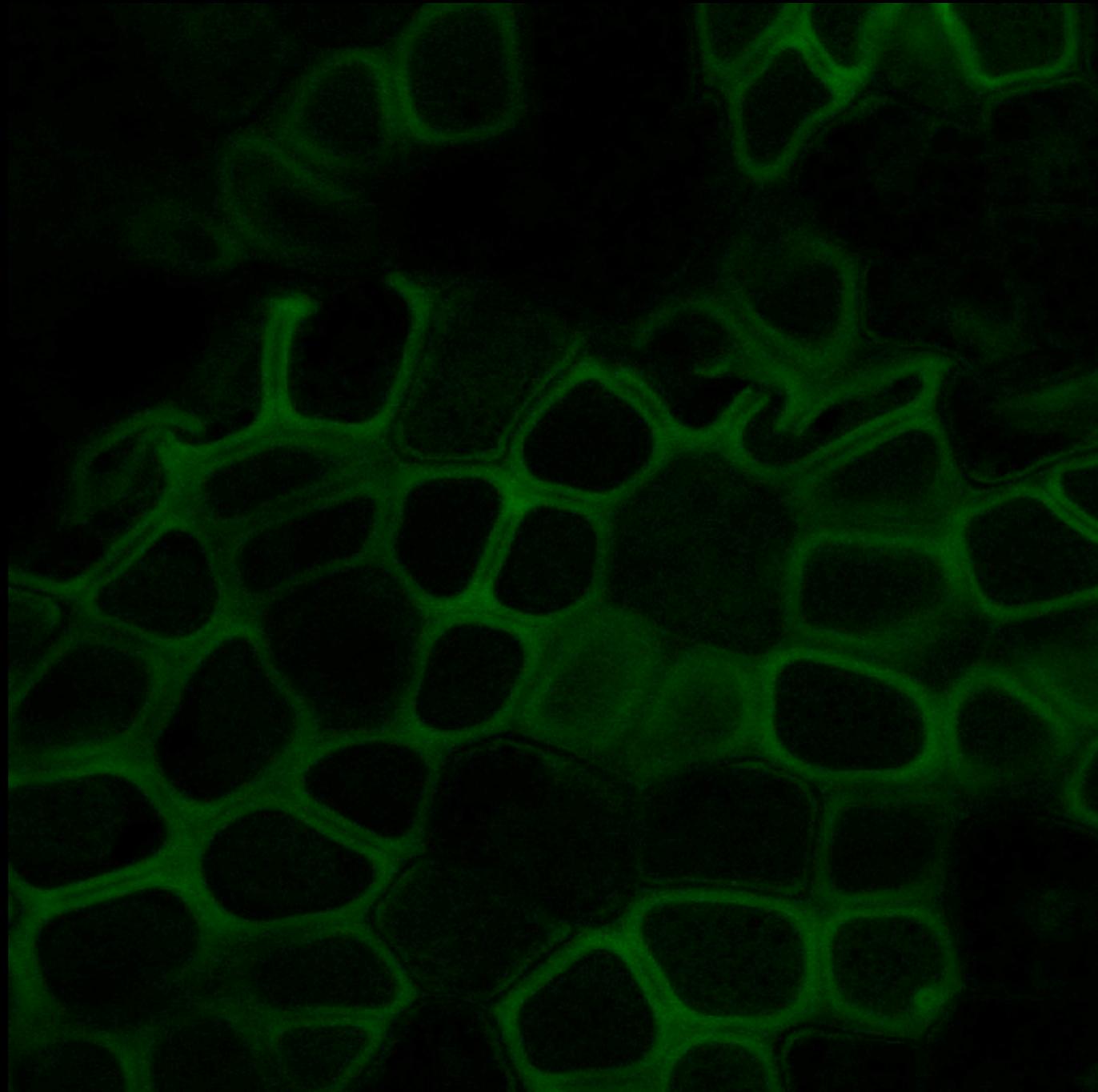


Excitation 488 nm

Untreated  
wheat bran



Excitation 488 nm



Untreated  
wheat bran

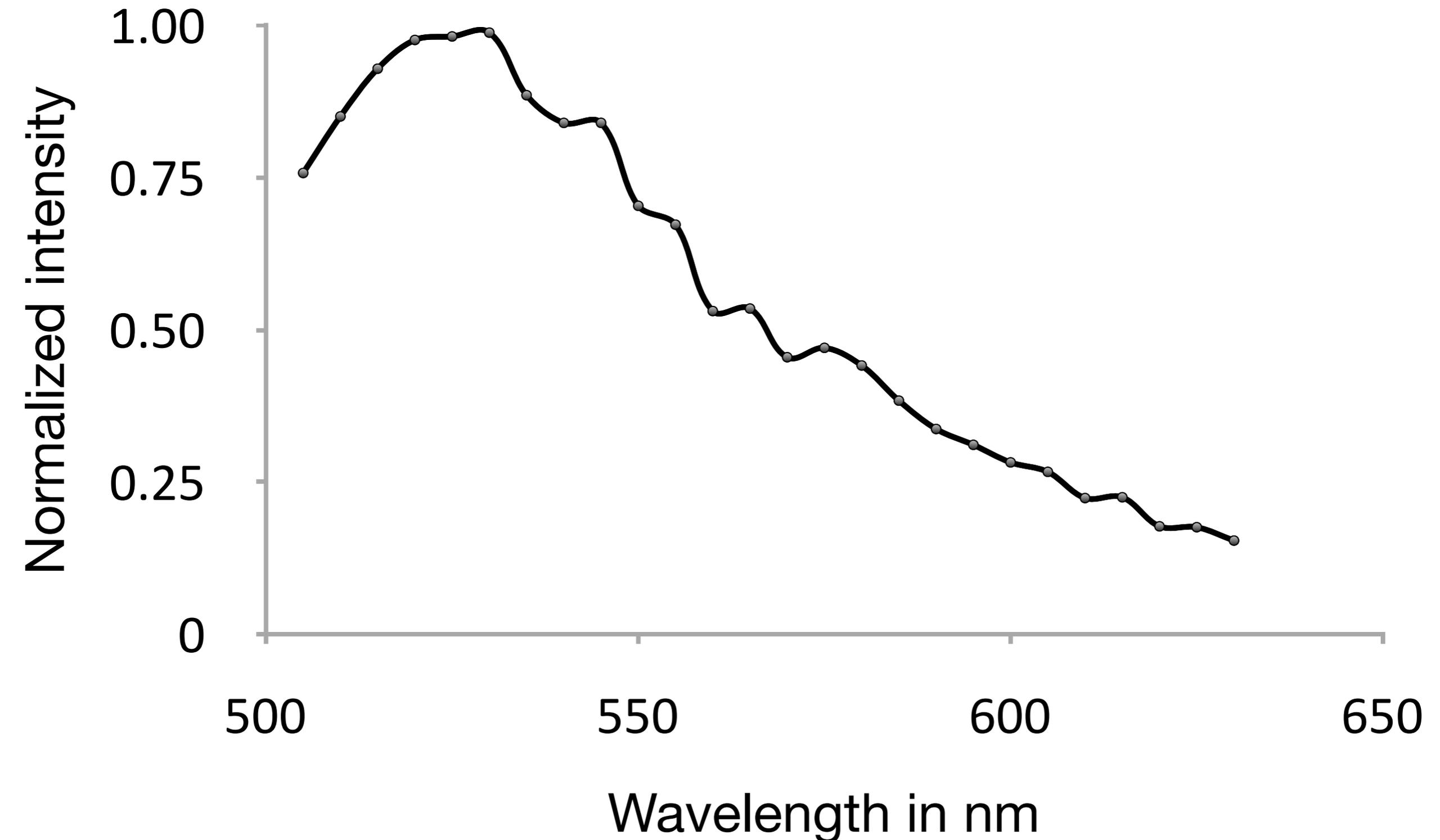
505 nm

630 nm

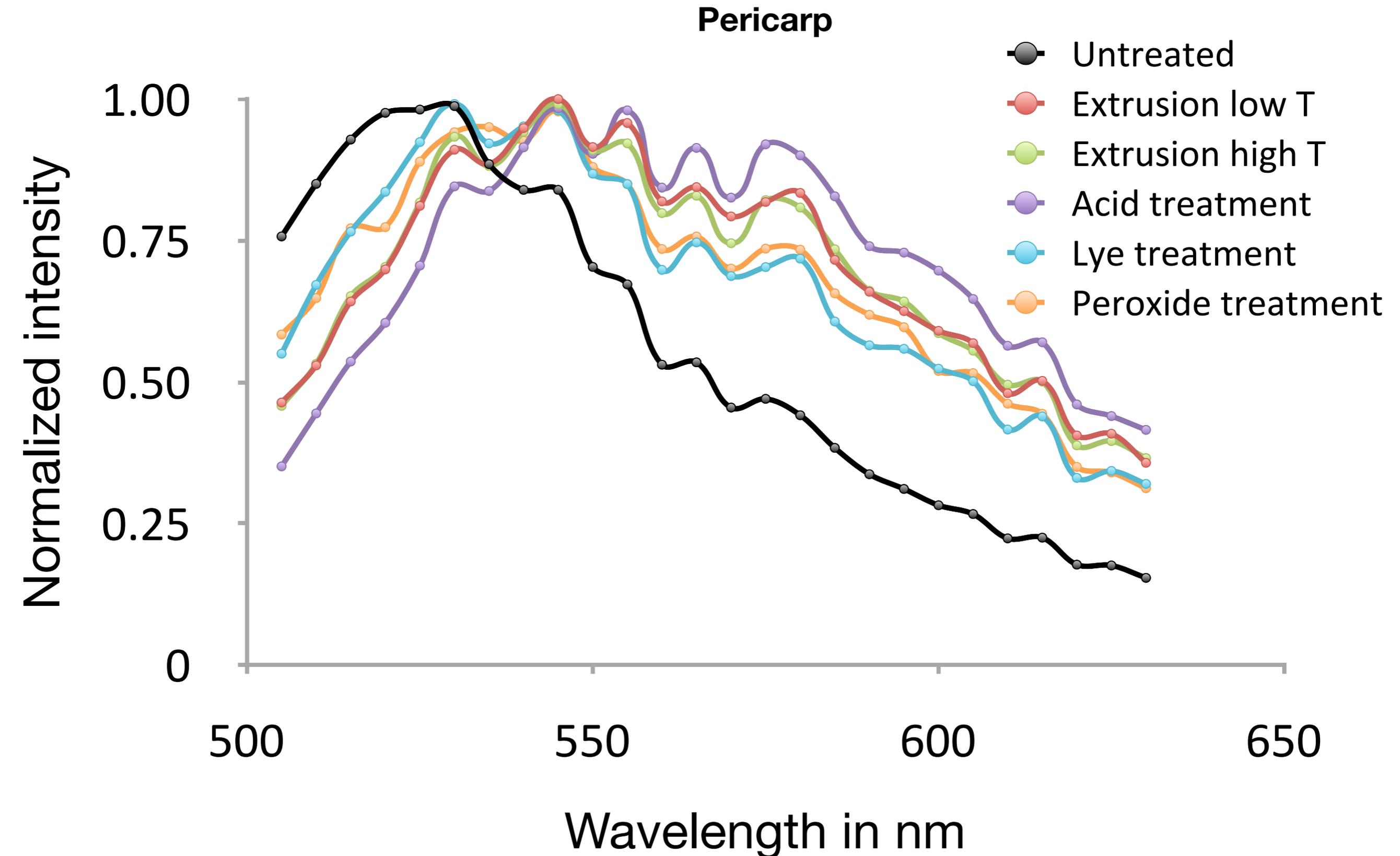
# Lignin auto-fluorescence spectrometry

# Lignin auto-fluorescence spectrometry

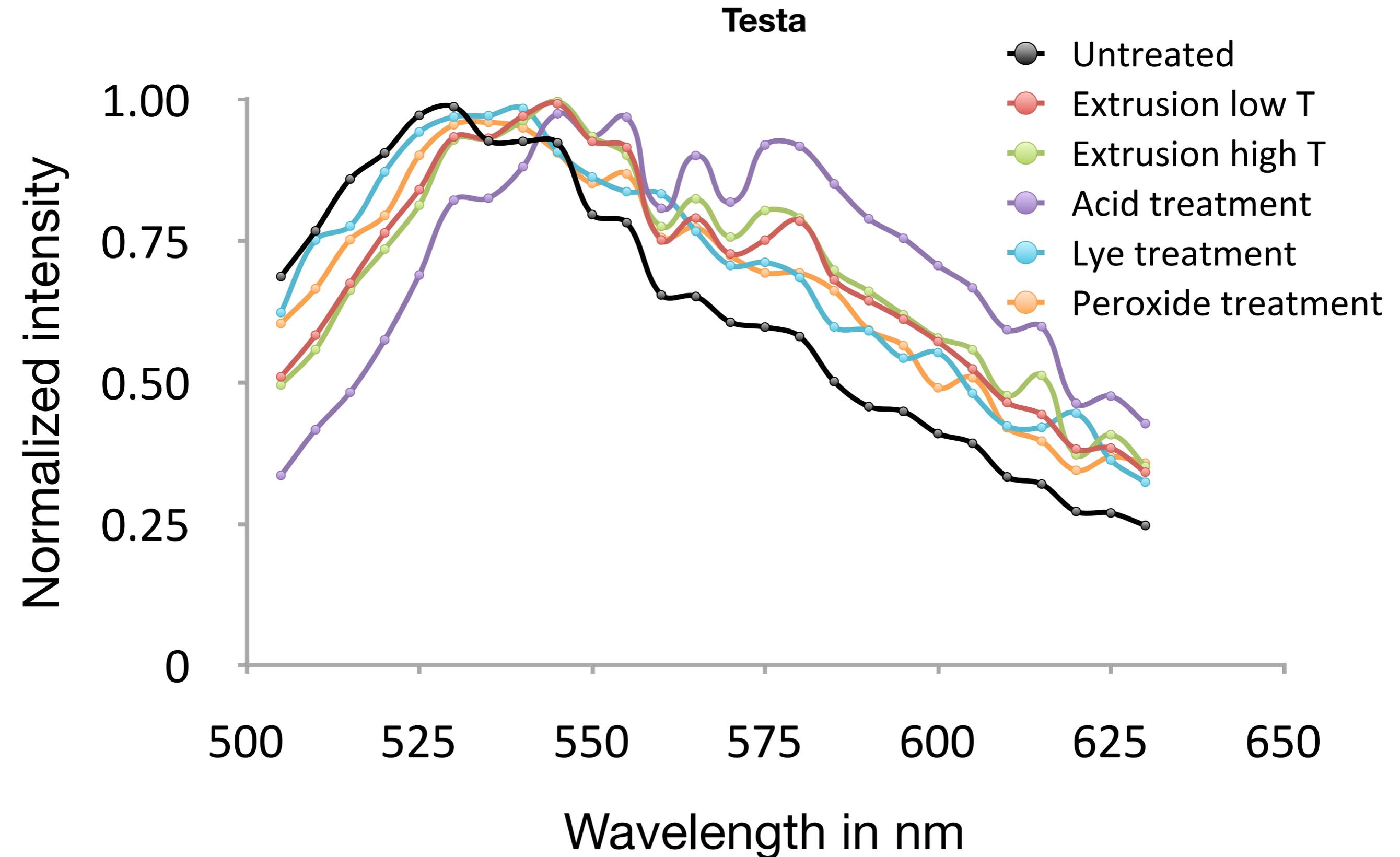
Pericarp



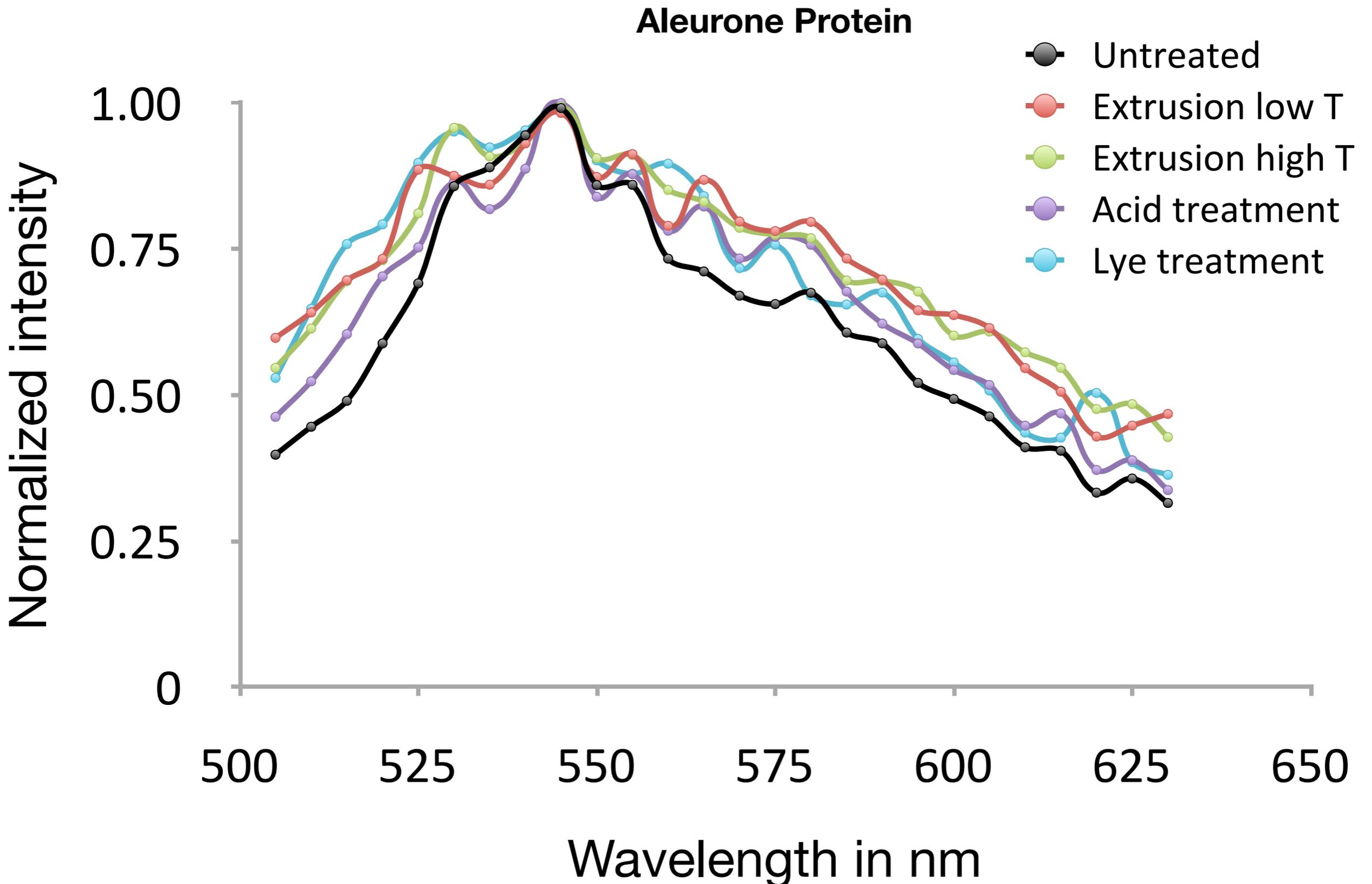
# Lignin auto-fluorescence spectrometry



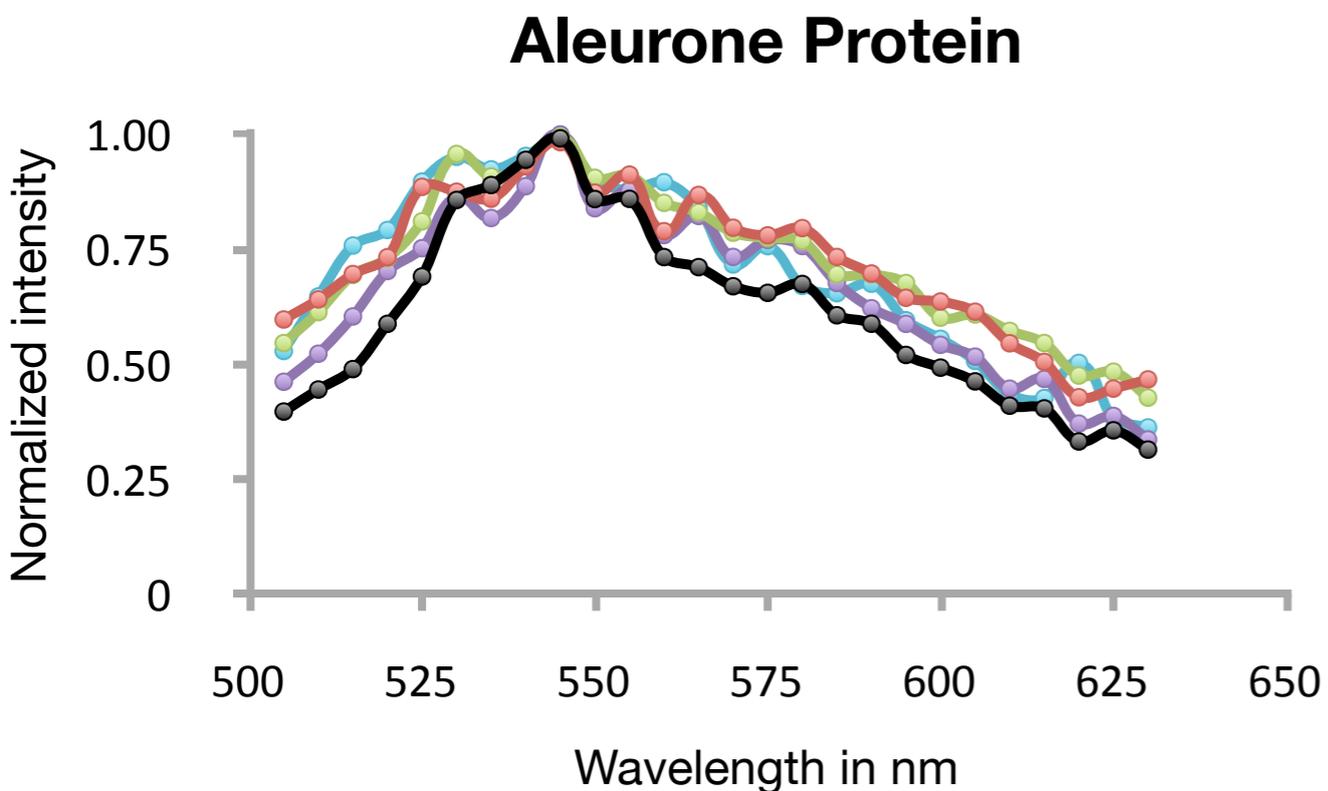
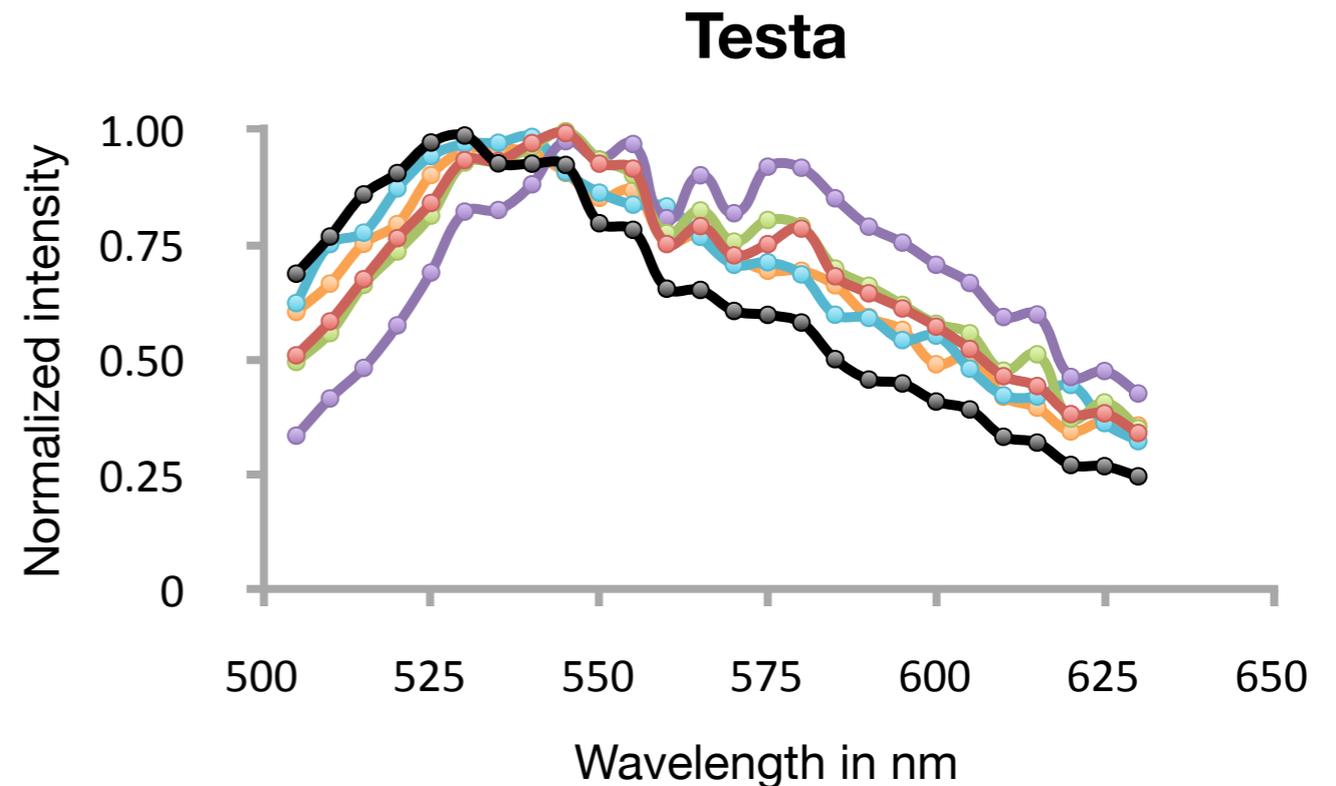
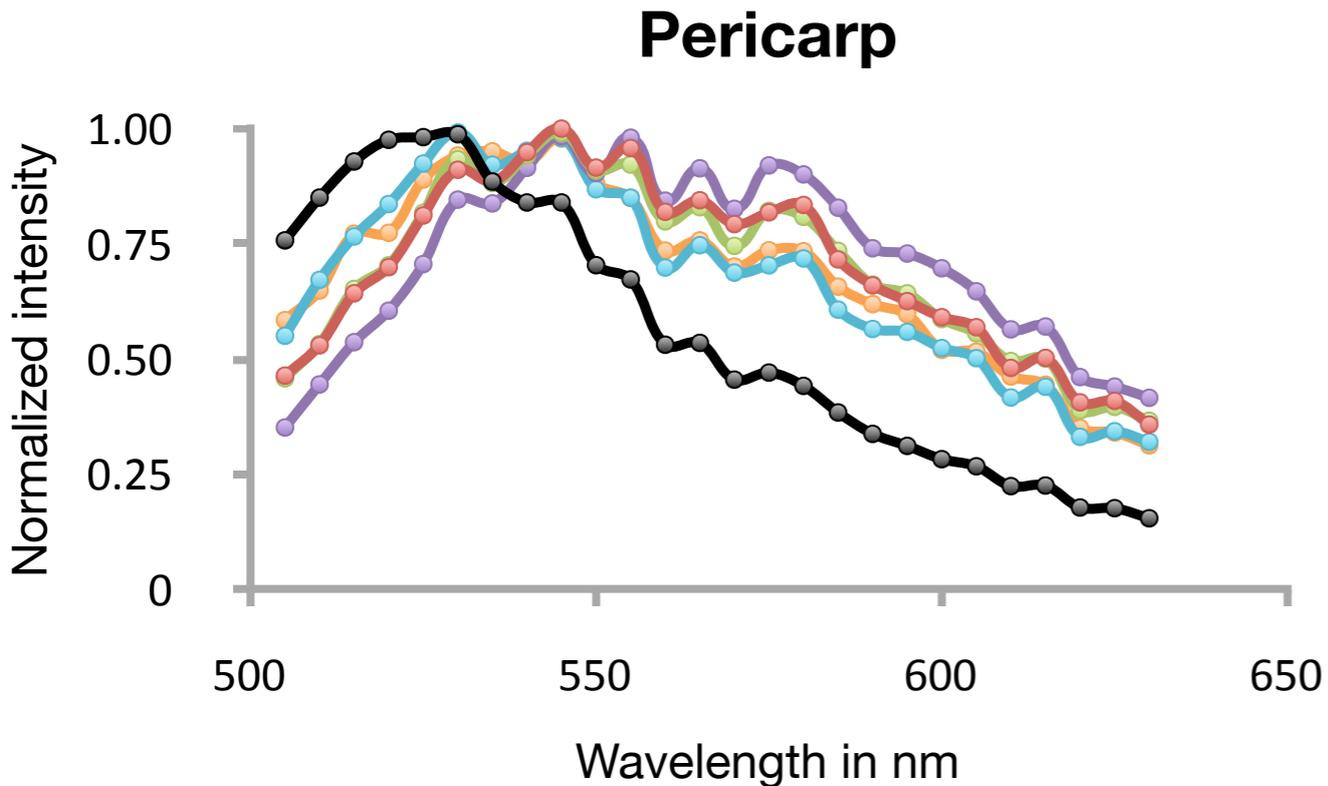
# Lignin auto-fluorescence spectrometry



# Lignin auto-fluorescence spectrometry



# Lignin auto-fluorescence spectrometry



## Conclusion

We see a shift in relative fluorescence intensity, but we don't know what we're looking at and we don't know what it changes into, nor how and why.

**Thank you for your  
attention**