

## Protocol I: Anatomical mapping of cellulose in plant tissues

This protocol describes a non-destructive method of anatomical mapping of cellulose structures in plant tissues.

### Solutions and Reagents:

Carbotrace™ is provided as concentrated solution. The following common reagents are required (not supplied):

- Phosphate buffered saline (PBS), pH 7.4

### Assay Procedure:

- Prepare thin sections of plant tissue.
- Dilute Carbotrace in PBS 1:1000.
- Apply diluted Carbotrace™ generously on the plant tissue. Use enough liquid (ca 0.5 ml) to prevent the sections from drying out during incubation. Incubate for 30 min.
- Wash 2 x 5 min in PBS.
- Mount sections for microscopic analysis.

### Fluorescence Microscopy:

- **Carbotrace™680:** Excite at 535 nm and detect emission using a 575-620 nm bandpass filter. Excitation at 635 nm and detection using a 655-755 nm bandpass filter is also possible. Alternatively, you can use the standard settings for AlexaFluor™635. The broad optical spectrum allows custom settings to be applied, using an excitation range of 530-565 nm and a detection range of 600-800 nm.

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