

## Description of Additional Supplementary Files

**File name:** Supplementary Movie 1

**Description:** Video demonstrating the simplicity of mounting specimens on the OTLS system, as well as real-time previewing of specimen(s), followed by automated imaging of the specimen(s).

**File name:** Supplementary Movie 2

**Description:** Time-lapse video demonstrating the operating principles of the OTLS system (image tiling as well as multiple specimens). Shown are 12 ECI-cleared human prostate biopsies, stained with TO-PRO3 (nuclear) and Eosin (cytoplasmic).

**File name:** Supplementary Movie 3

**Description:** Surface blend volume rendering of one of the ECI-cleared human prostate biopsies shown in **Fig. 3a**. Color channels are TO-PRO3 (cyan) and Eosin (purple).

**File name:** Supplementary Movie 4

**Description:** Z-stack of benign glands in a ECI-cleared human prostate biopsy. The TO-PRO3 and Eosin channels are pseudo-colored to mimic conventional H&E staining.

**File name:** Supplementary Movie 5

**Description:** Maximum intensity volume rendering of a TDE-cleared GFP-expressing mouse brain slice.

**File name:** Supplementary Movie 6

**Description:** Maximum intensity volume renderings of the bronchial tree (EpCAM, shown in yellow) in a Ce3D-cleared mouse lung

**File name:** Supplementary Movie 7

**Description:** Z-stack of a small sub-region of an entire ExM-cleared mouse kidney specimen. Color channels are DAPI (white), WGA-lectin (magenta), Coll IV (cyan), and Podxl (green).

**File name:** Supplementary Movie 8

**Description:** Surface blend volume rendering of a small sub-region of an entire ExM-cleared mouse kidney specimen

**File name:** Supplementary Movie 9

**Description:** Fly through of a blood vessel for the expanded mouse kidney shown in Supplementary Videos 8 and 9.

