

## Supplementary Figure 1. Peg-and-socket interactions in proximity to the pericyte nucleus.

(A) A cropped region of the data set, including part of the pericyte process with serrated edges, was taken for detailed examination. (B) Surface model of the endothelium alone with yellow outline showing region of pericyte overlap. Magnified view (dashed box) shows endothelial sockets and broadly distributed, shallow grooves created by the pericyte process. The under-surface of the pericyte process reveals pegs (green), numbered to match their corresponding endothelial sockets. The appearance of one pericyte peg (#3) in the raw 2D data is marked with a black arrowhead. (C) The surface model is rotated to the left 90 degrees to the left from panel B. Magnified view (dashed square) reveals sockets and deeper impression made along the lateral edge of the process. The appearance of one claw-shaped pericyte peg (#1) in the raw 2D data (boxed region enlarged) is marked with an arrowhead. (D) The endothelial model is rotated to the left another 30 degrees from panel C to display peg-and-socket interactions on the opposing side of the lumen.

A



1 Persists through multiple image slices in Z-stack



- 2 Protrusion causes clear indentation of apposing cell membrane beyond its normal shape (red line)
- 3 Surrounded on all sides by the membrane of the apposing cell type

## В

## **Examples of exclusions**

Convoluted interactions-hard to decipher structure



Superficial and not surrounded on all sides



## Supplementary Figure 2. Criterion for verified pericyte or endothelial pegs.

(A) To be included as a pericyte or endothelial peg in examination of the MICrONS data set, the three criterion listed must have been fulfilled. (B) Examples of faux pegs that were excluded from the final data set.



**Supplementary Figure 3. Mesh pericyte in MICrONS data set.** (A) Raw annotations and graphical representation of pericyte and endothelial ultrastructures from the 3D rendered MICrONS electron microscopy (EM) data set showing a mesh pericyte (red) along the brain vasculature. The edges of pericyte processes are annotated (red dots) to outline the boundary of the pericyte processes. (B) Close up and (B') electron microscopic images highlight the tendency for pericyte pegs (yellow) and endothelial pegs (dark blue) to flank along the endothelial junctions (light blue dotted line). (C) Close up and (C') electron microscopic images demonstrate how secondary pericyte processes envelope the vasculature near the endothelial cell nuclei (blue).



**Supplementary Figure 4. Pericyte secondary processes associate with endothelial nuclei. (A,B,C)** Three examples of showing the emergence of secondary process (white arrows) from a thin-stand pericyte process to interact with an underlying endothelial cell nucleus (yellow arrow).