## Intracellular calcium dynamics of lymphatic endothelial and muscle cells co-

## cultured in a Lymphangion-Chip under pulsatile flow

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## SUPPLEMENTARY INFORMATION



## LMC monoculture

**Supplementary Fig. 1 LMC calcium dynamics in monoculture while exposed to step shear**. Calcium dynamics of devices with monoculture of LMCs near lumen boundary and away from the flow field (50-100 μm distance) while exposed to shear step with 10 dyne/cm<sup>2</sup> amplitude.



**Supplementary Fig. 2 Lumen cross-section size with and without LMCs.** (A) Micrographs of lumen cross-section and (B) the ratio of lumen height (b) over lumen width (a) (i.e., b/a) for devices with and without LMCs.

Supplementary Video 1 3D confocal micrograph of a quarter of the lumen formed of a confluent monolayer of LECs (green) surrounded by multiple layers of LMCs (red) embedded in 3D ECM made using GLP technique. The cells were fixed and stained for Lyve-1 (green), F-actin (red), and nuclei (blue) after 4 days of cell culture.

Supplementary Video 2 Timelapse fluorescent video of LECs intracellular calcium content (Fluo-4) while the Lymphangion-Chip lumen was exposed to step shear profile. There is a sudden rise in calcium signal following by a gradual decay.

Supplementary Video 3 Timelapse fluorescent video of LMCs intracellular calcium content (Fluo-4) while the Lymphangion-Chip lumen was exposed to step shear profile. There is a sudden rise in calcium signal following by a gradual decay.