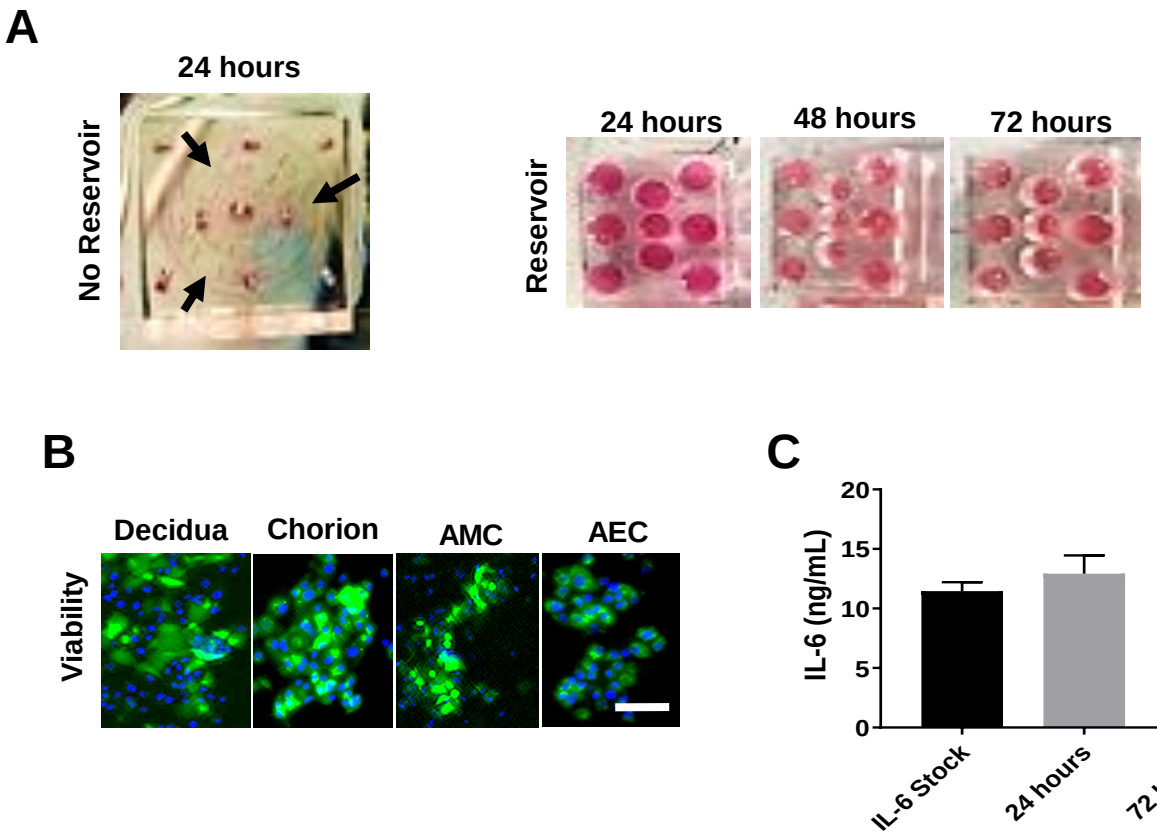
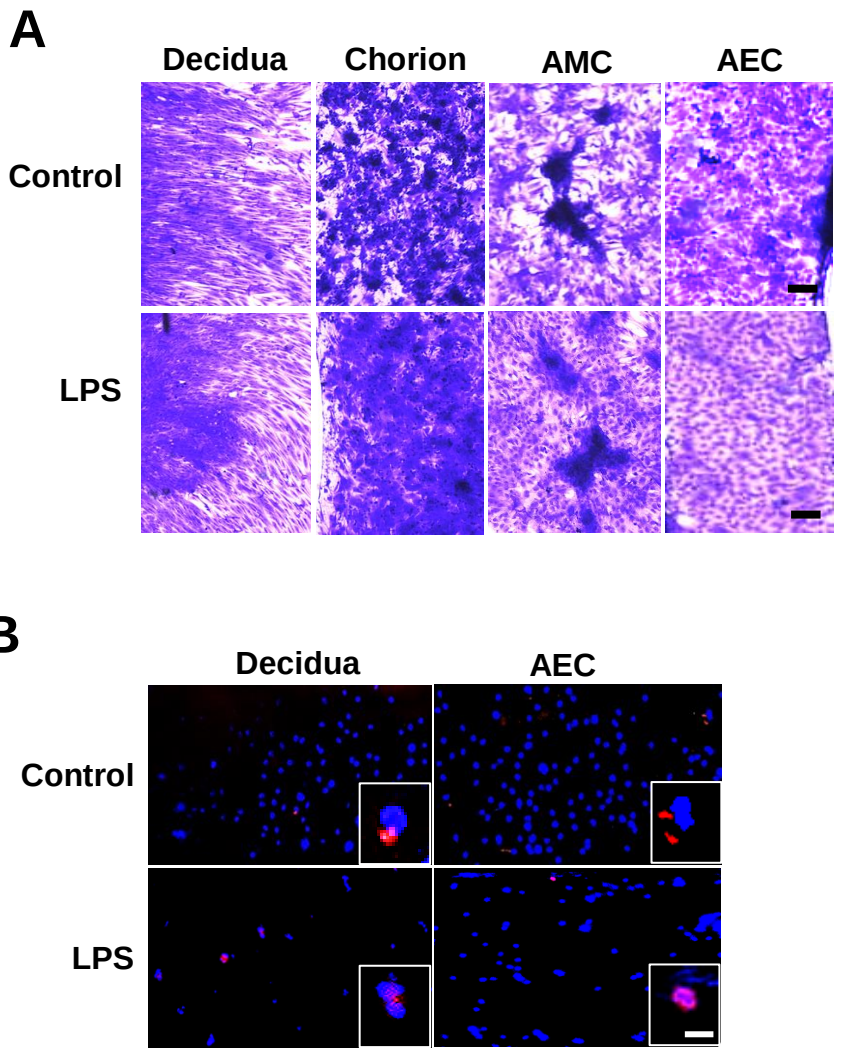


Supplemental Figure 1: Initial FMi-OOC device characterization for testing cell culture and media collection capabilities.

A) The FMi-OOC device without the top media reservoir layer, showing fast evaporation of media within 24 h, where bubbles in the cell culture chambers can be seen (black arrows). The FMi-OOC device with the on-chip reservoir layer attached on top, which enabled maintaining media levels within the cell culture chambers and reservoirs for up to 72 h. Scale bar = 50 µm.

B) Example images of cells grown within the FMi-OOC for 72 h, showing good viability as indicated by strong calcein AM stain (green) but no ethidium homodimer-1 stain (red).

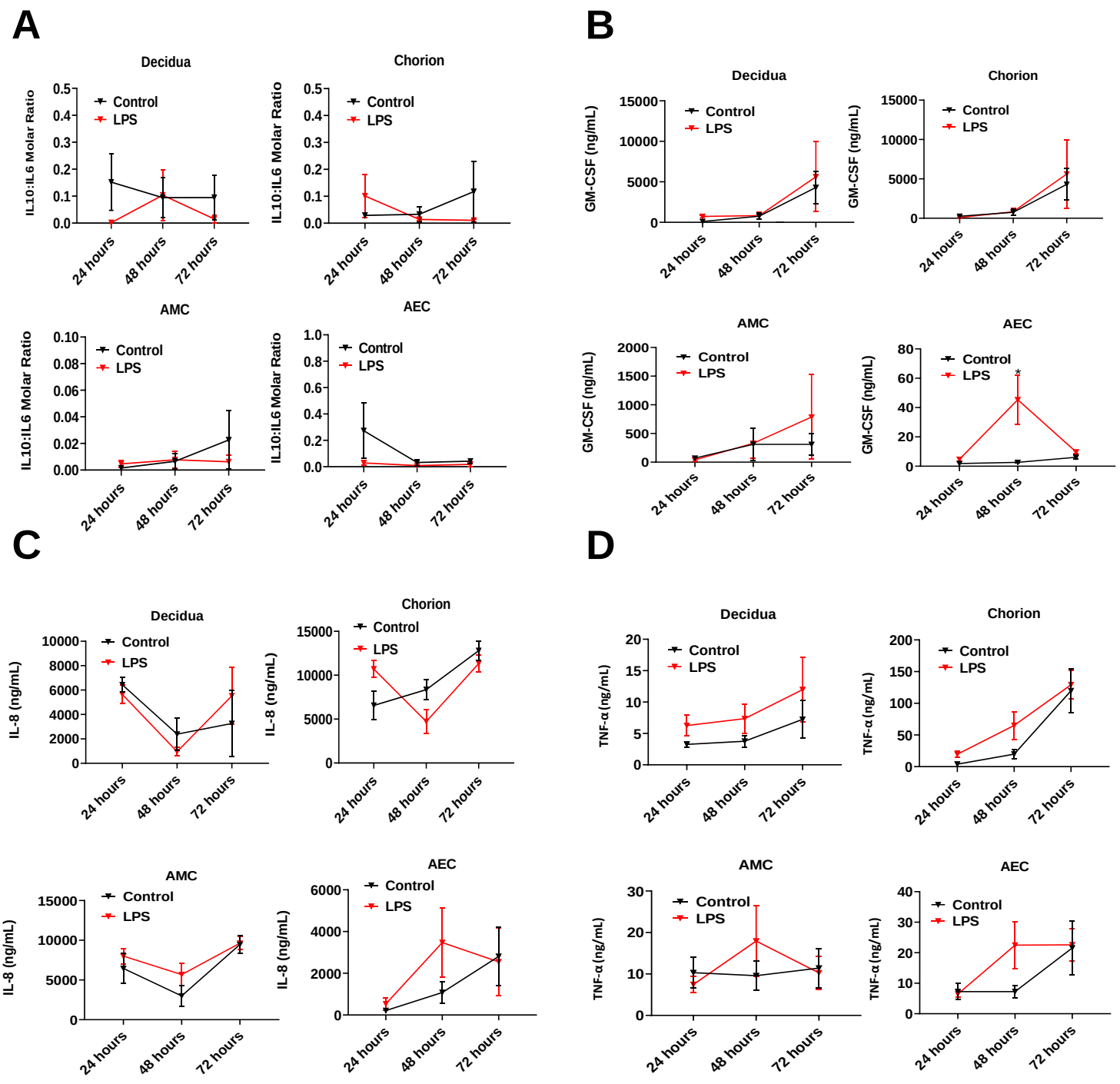
C) ELISA measured pro-inflammatory cytokine IL-6 concentrations within the FMi-OOC, showing no difference over a 72 h period, indicating no or limited absorption of cytokines to the PDMS device or reservoir system (*n* = *3*). Values are expressed as mean ± SEM.



Supplemental Figure 2: LPS-induced NF-κB translocation but showing no cell death within the FMi-OOC system.

A) Maternal and fetal cells remained viable, as noted by the retention of crystal violet stain, even after LPS treatment on the maternal side for 72 h. Scale bar = 50 µm.

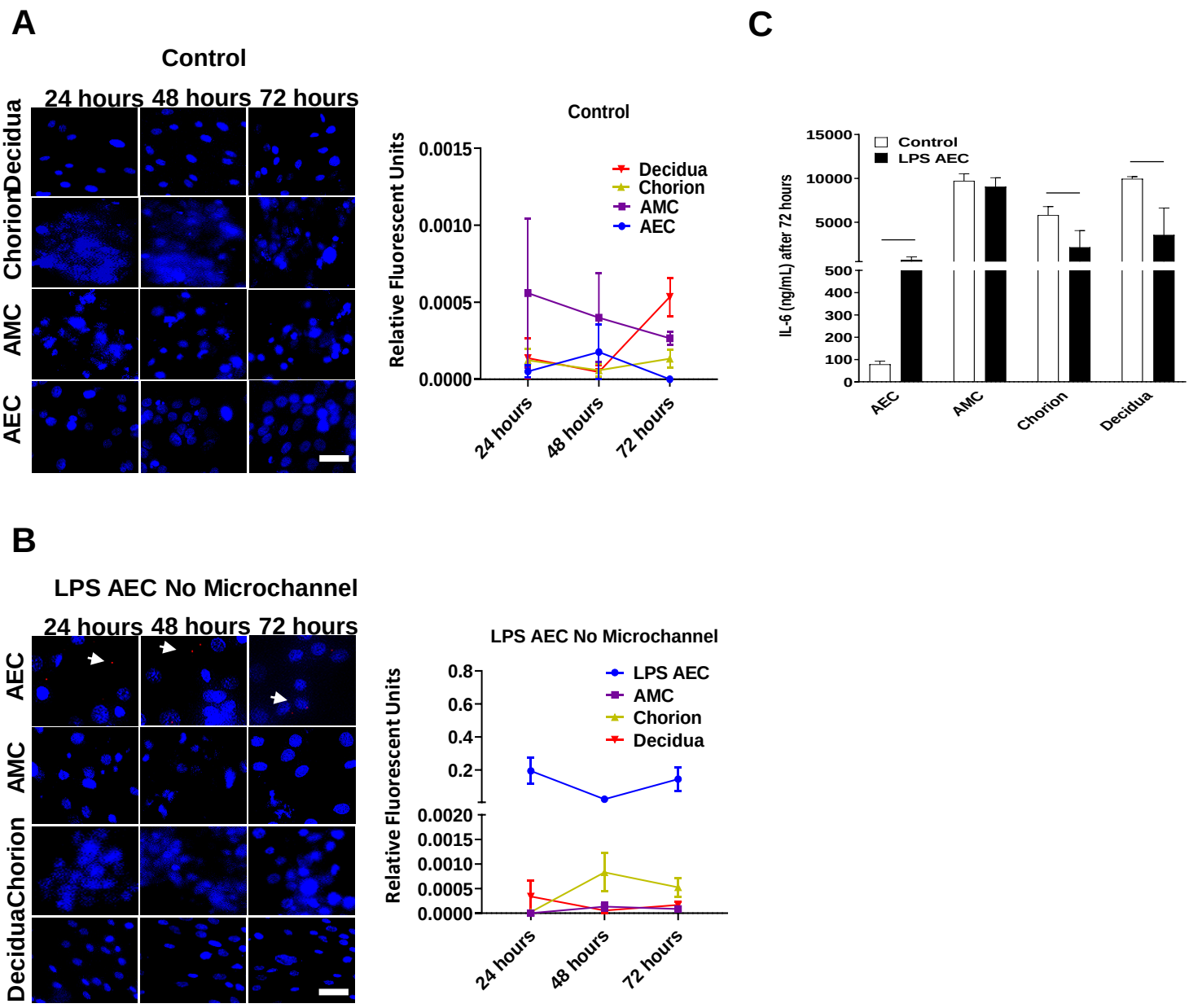
B) LPS-treated decidua and AECs within the FMi-OOC induced NF-kB (red) translocation from the cytoplasm to the nucleus, documenting its activation (*n* = 3). Scale bar = 10 µm. Blue – DAPI, Red – NF-kB.



Supplemental Figure 3: Production and propagation of pro- and anti-inflammatory mediators in the FMi-OOC.

A) Multiplex measured pro- and anti-inflammatory cytokines, IL-6 and IL-10, converted to their molar ratios and graphed for each cell type and time point (*n = 4)*.

B) Multiplex measured pro-inflammatory cytokine GM-CSF (AEC: *P=0.032*), **C)** IL-8, and **D)** TNF-α in each cell culture chamber after 24 h, 48 h, and 72 h (*n* = 4). Values are expressed as mean ± SEM.



Supplemental Figure 4: Experimental controls for the LPS propagation studies within the FMi-OOC system

A) To ensure that we did not have any non-specific binding of LPS antibodies, control FMi-OOCs were stained following the immunocytochemistry protocol but without the primary antibody. LPS was not detected in any cell or time point as seen by no red LPS dots (*n = 3)*. Scale bar = 10 µm. Blue – DAPI, Red – LPS. Values are expressed as mean intensities ± SEM. **B-C)** To confirm that our model relied on LPS trafficking through the interconnecting microchannels to mimic ascending infection *in utero*, an FMi-OOC was designed without microchannel connections between the cell culture chambers. AECs treated with LPS (100 ng/mL) (red; white arrows) for 72 h neither propagated LPS nor inflammation **(C)** to other cell chambers due to the lack of microchannels. However, AECs were found to have LPS at all time points **(B)** and showed higher inflammation after 72 h compared to other cell chambers **(C).** The line above the bar graph means at least 2-fold change. Scale bar = 10 µm. Blue – DAPI, Red – LPS. Values are expressed as mean intensities ± SEM.