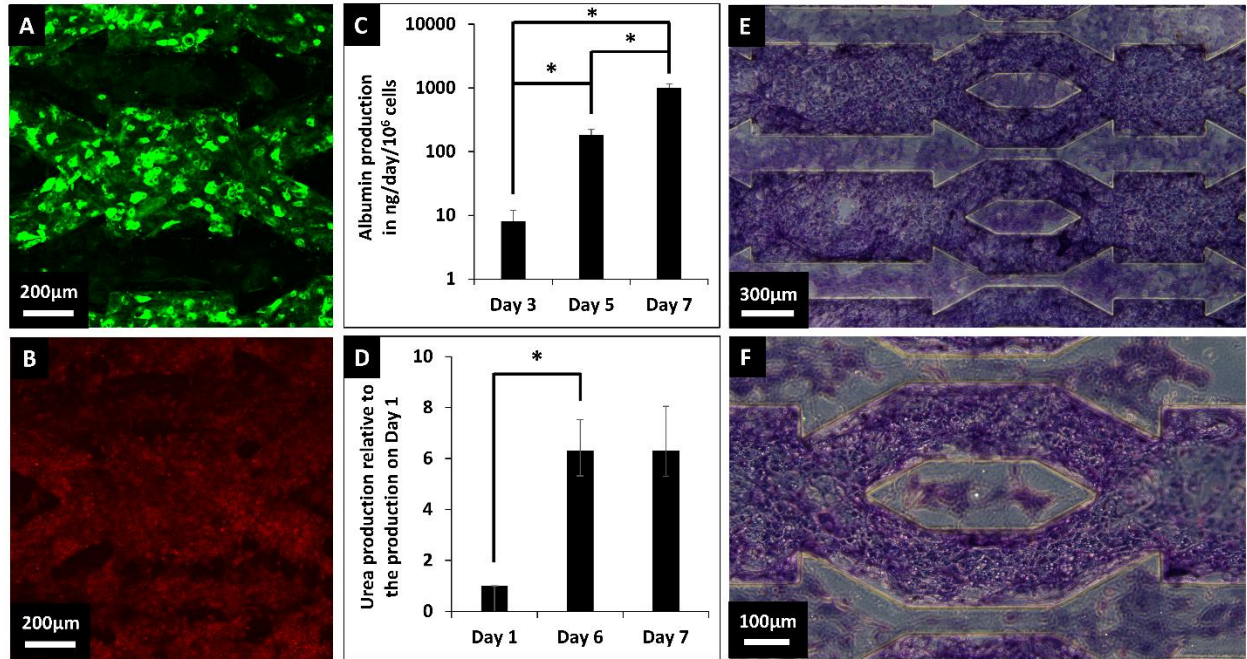
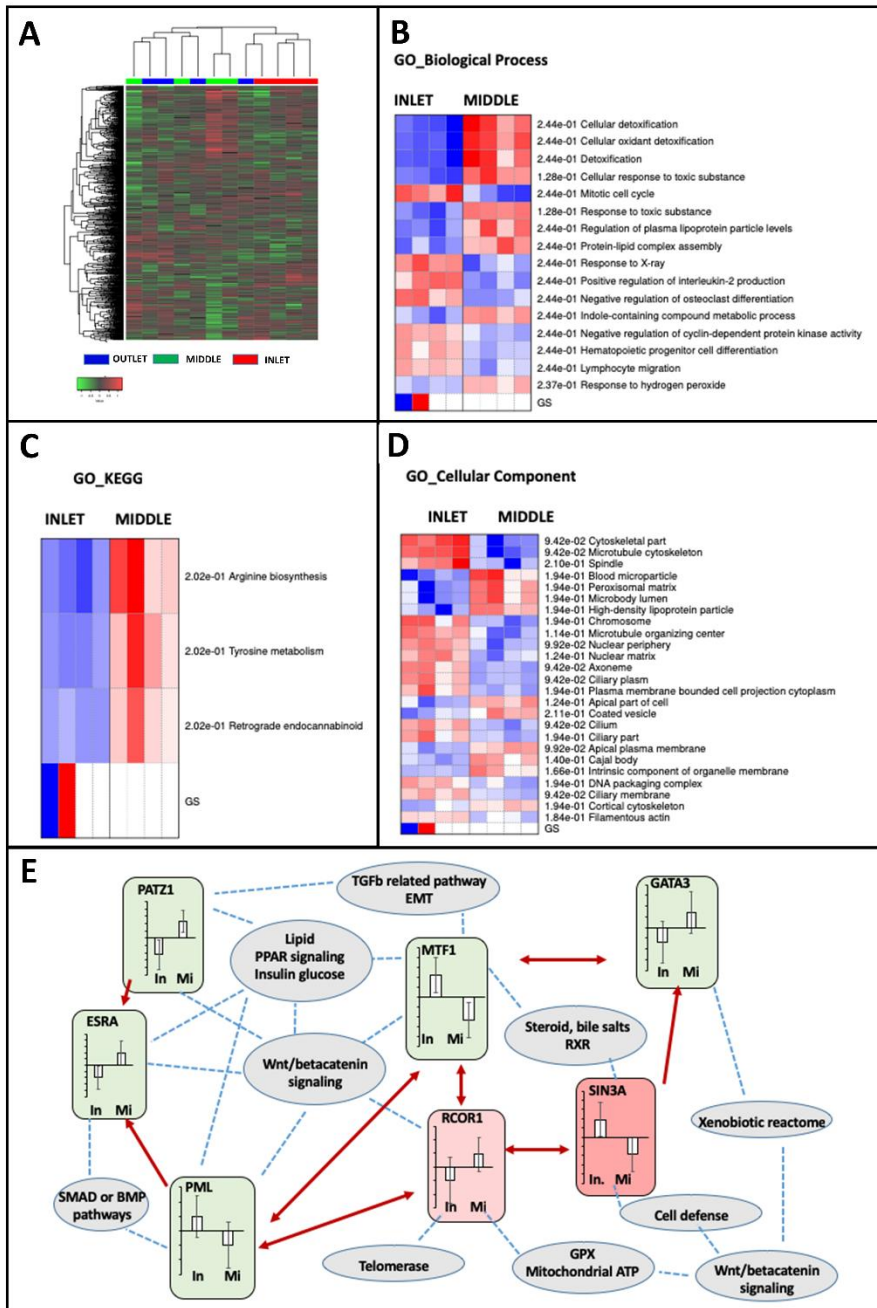


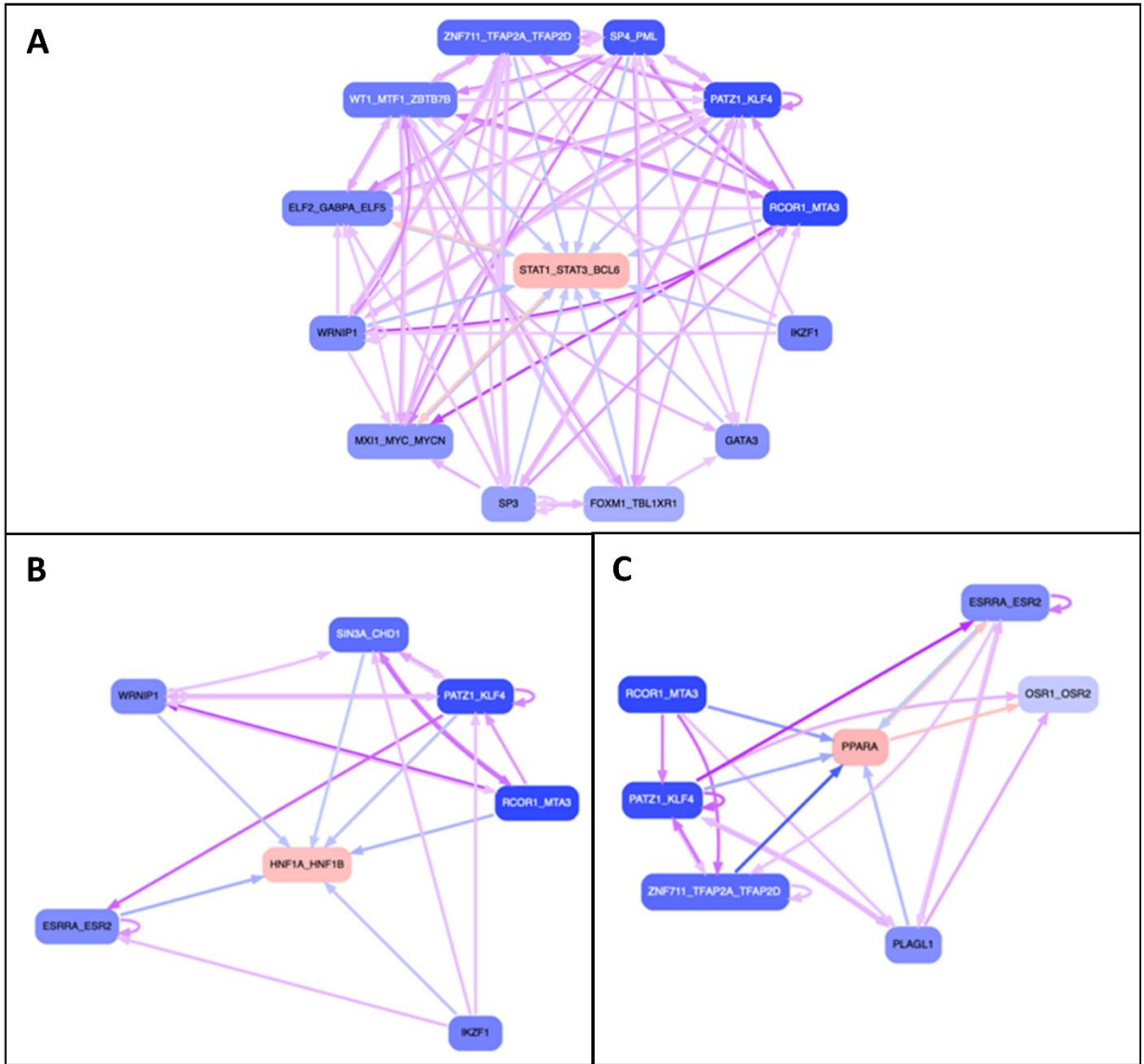
**Figure 1:** Cell morphologies observed in step 4 of the differentiation protocol: (A) after 24h of static culture in the biochip (B) after 24h of perfusion (C), after 72h of perfusion at the inlet and (D) in the middle area, and after 144h of perfusion (E) at the inlet and (F) in the middle area. Cell morphologies observed in step 4 of the differentiation protocol in Petri dishes on Day 1 (G) and Day 7 (H).



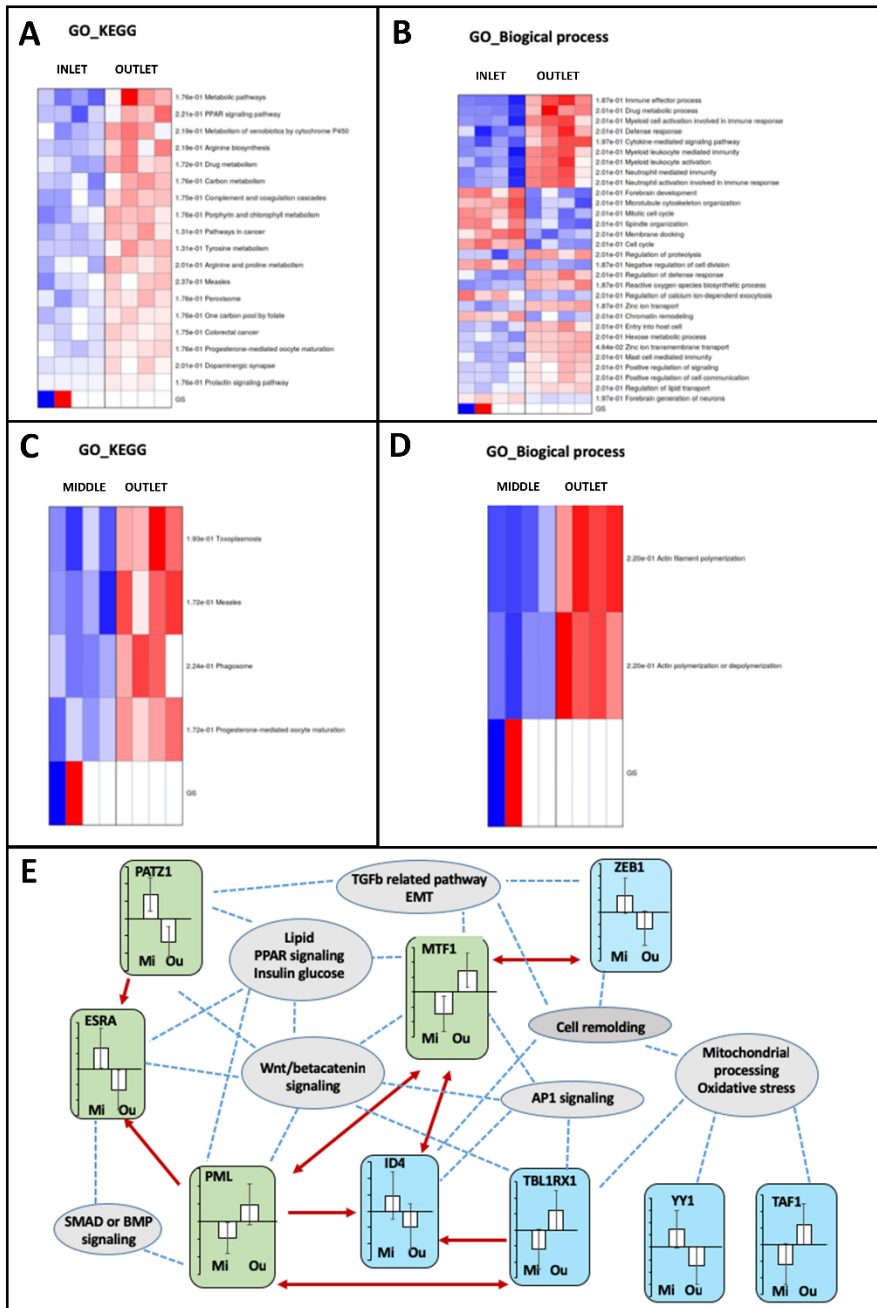
**Figure 2:** Immunostainings after 7 days of differentiation in step 4. Staining for Albumin (green, A) and CYP3A4 (red, B). Albumin (C) and Urea (D) productions measured in the cultures during step 4 of the differentiation protocol. PAS staining performed on samples at the end of the differentiation protocol (E-F).



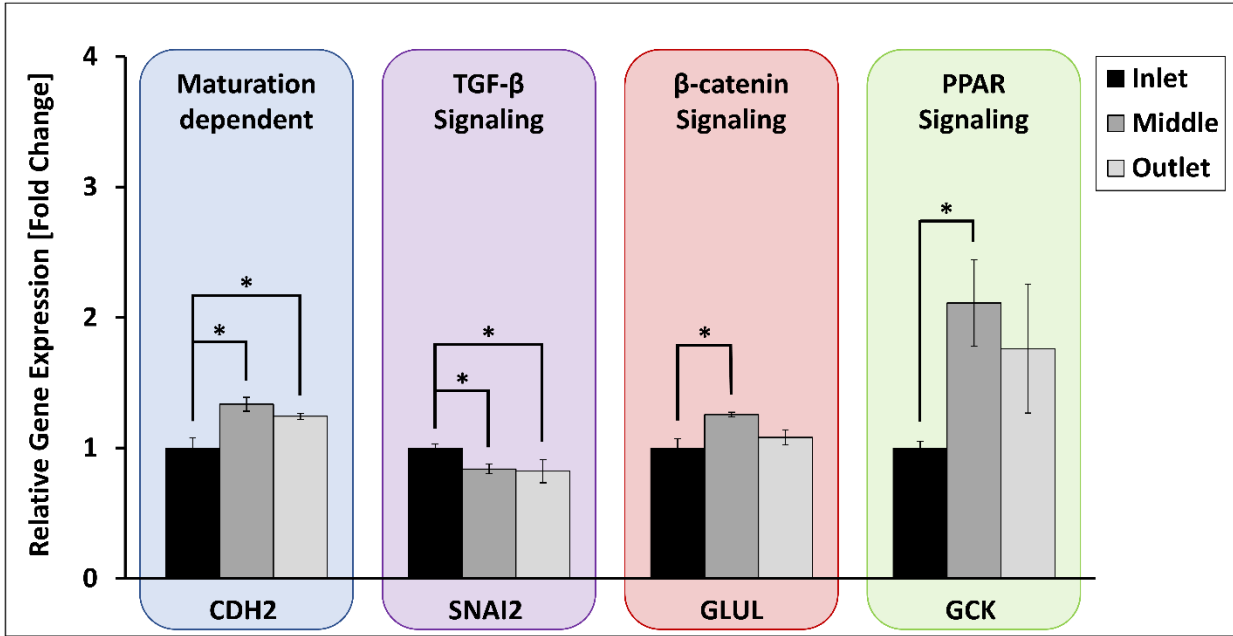
**Figure 3:** Heat map clustering of the Inlet vs Middle area vs Outlet comparison (A). GO biological process (B), GO KEGG pathway (C), and GO cellular compartment (D) extracted from iDEP7 post treatment on the comparison between Inlet and Middle area. Interaction network connecting the different TF motifs highlighted by MARA analysis and their target pathways in the comparison between Inlet and Middle area (E).



**Figure 4:** Interaction network of key TF motifs identified by Ingenuity Pathway Analysis upon comparison of cells collected from the middle area and the inlet.



**Figure 5:** GO KEGG pathway (A) and GO biological process (B) extracted from iDEP7 post treatment on the comparison between Inlet and Middle area. GO KEGG pathway (C) and GO biological process (D) extracted from iDEP7 post treatment on the comparison between Middle area and Outlet. Interaction network connecting the different TF motifs highlighted by MARA analysis and their target pathways in the comparison between Middle area and Outlet (E).



**Figure 6:** Expression levels of selected genes measured by RT-qPCR on samples collected from the inlet, the middle area and the outlet of the biochip.