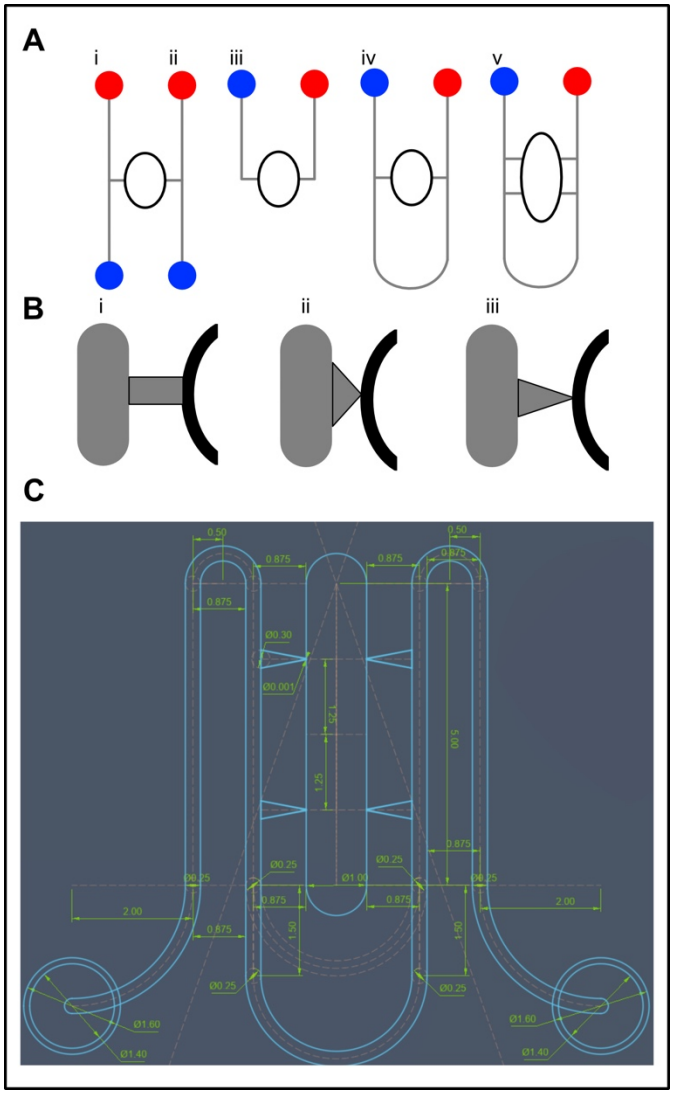
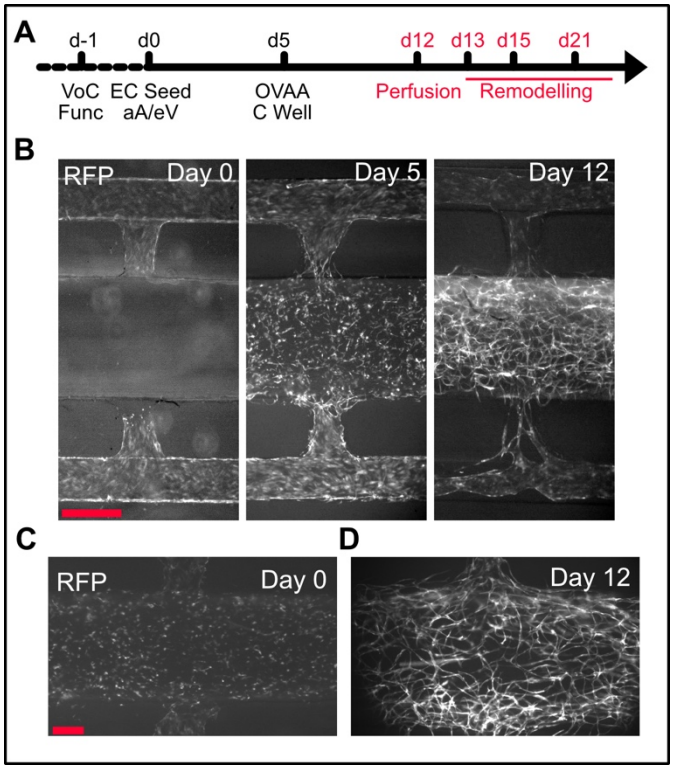
Electronic Supplementary Material (ESI) for Lab on a Chip.

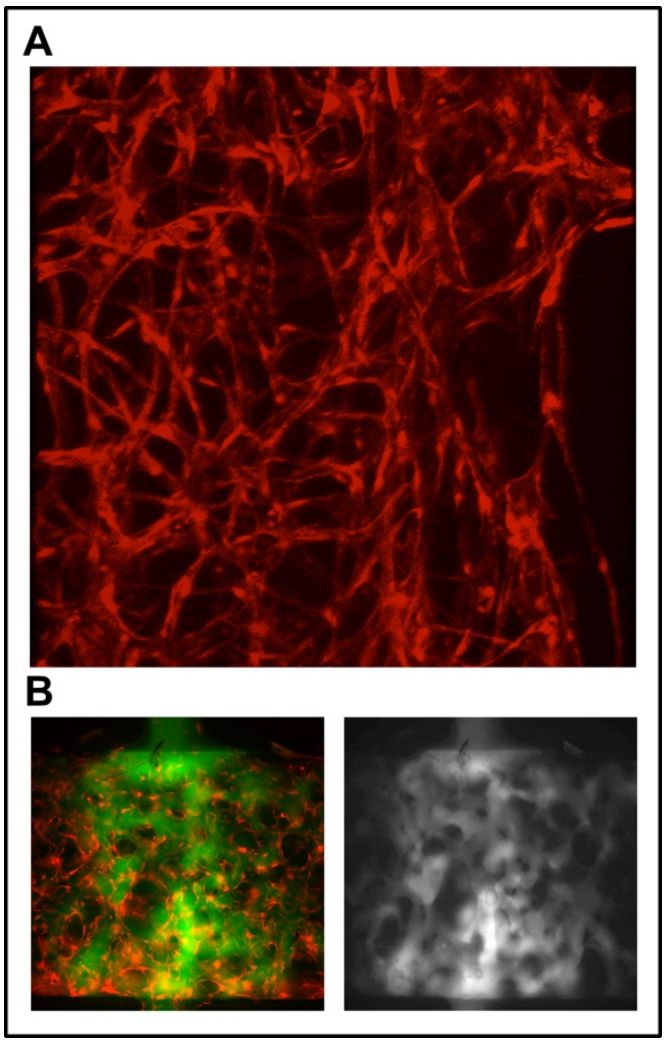


Supplementary figure 1: LoC device design optimisation. A) Different design of the side channel length and configuration. Designs that were considered include: independent (i-ii) afferent arteriole (aA) and efferent venule (eV), direct total perfusion (iii) in the central well, or partially divided (iv, v) flow into the central well. B) The junction between aA and eV can be designed to have a 90° angle (i), a bigger angle (ii) to receive more flow or a longer length (iii). C) CAD design of the LoC. Measures are expressed in mm.

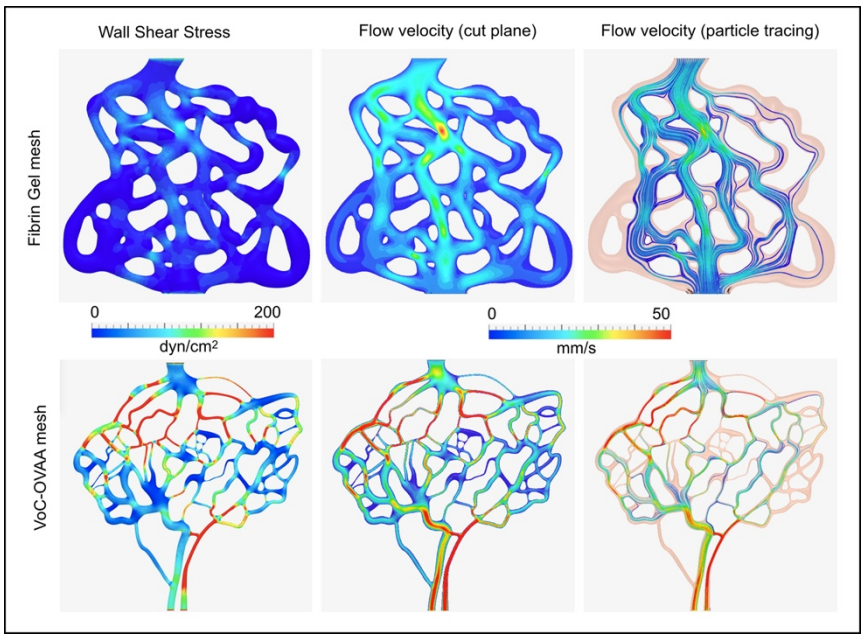
Supplementary information



Supplementary figure 2: VoC timeline and versatility. A) Timeline for the creation of perfused vasculature-on-chip. The VoC is first coated with fibronectin to promote the adhesion of ECs in the side channels. At day 0, ECs are seeded in the side channels (aA and eV) and left in culture for 5 days to coat the channels on all sides. At day 5, the co-culture of ECs and FBs is seeded in the central well and cultured in static conditions for 7 days to promote capillary network formation. At day 12, the VoC can be linked to the microfluidic system and continuously perfused. B) Photographs of the VoC system with HUVEC-RFP at day 0, 5 and 12 of culture in co-culture with HDF. Scale bar: 500µm. C) Photographs of the VoC system with HUVEC-RFP at day 0 and after 12 days (D) in co-culture with MSCs. Scale bar: 250µm.



Supplementary figure 3: A) 3D projection of the vascular network formed by HUVEC-RFP in the central well of the VoC-OVAA after 7 days of culture, before perfusion. B) composite image (RFP-HUVEC + mAb-647) and individual mAb-647 channel of perfused HUVEC networks in fibrin hydrogels (Cfr Fig 1).



Supplementary Fig 4: CFD analysis of microvascular networks generated in fibrin hydrogels (upper panels) and OVAAA (lower panels). Rightmost panels show top view of the 3D mesh coloured according to wall shear stresses magnitude at each face (7.5M faces/mesh). Centre panels show flow velocities on a plane crossing the middle of all vessels. Rightmost panels show top view of particle trace analysis across the networks orange shading highlights the network, coloured traces indicate particle routes and velocities. In each panel the flow runs from top to bottom.

	3D Printing	Photolithography	Notes
Resolution *	67µm (15µm)	<1µm	
Features height **	>1mm	<200µm	
Post processing	PFOTS	PFOTS	3DP moulds do not require repeating post porcessing
Reproducibility	Good	High	
Durability (moulds) ***	High	Moderate	Features on 3DP moulds are only marginally affected by peeling forces. Only relevant of features with high aspect ratio.
Cost/chip ****	Low	Medium/high	
Lab Environment	Chemical Lab	Clean room	
Equipment cost	Medium	High	
Photomask sourcing	Not required		
* Number outside parenthesis refers to equipment used in this work. Number in brackets refers to the maximum resolution achieved with DLP technology to date.			
** For both technologies maximum features height is limited by aspect ratio but SU-8 photholytography is further limited by thickness of phothoresist achievable by spin-coating. 3DP allows creating features with different heights on the same mould which is technically challenging with SU-8 photolithography.			
*** Differently from 3DP, moulds created by SU-8 photolythography on silicon wafers are fragile, require repeating PFOTS coating every few replicas, and are subject to features detachment.			
**** Include lower production costs of the mould and higher durbility leading to less reagents consumption and "man-hours"			

Table 1: Comparison between SU-8 photolithography and 3D printing for the manufacture of moulds for PDMS soft lithography.