



## Supporting Information

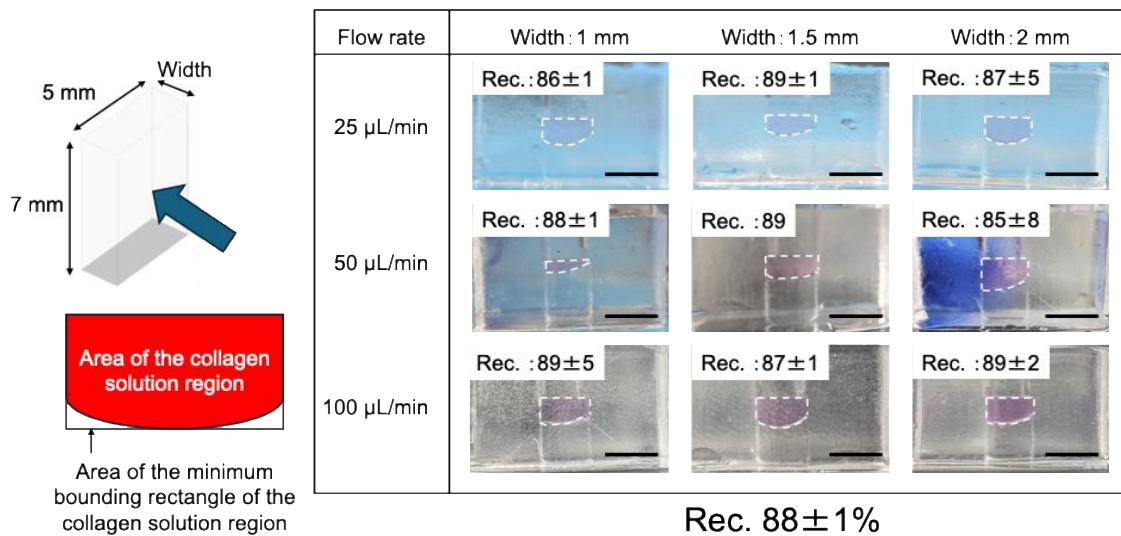
for

### **Interface-engineered Caco-2 cell culture on a collagen-coated liquid–liquid interface in a microfluidic device**

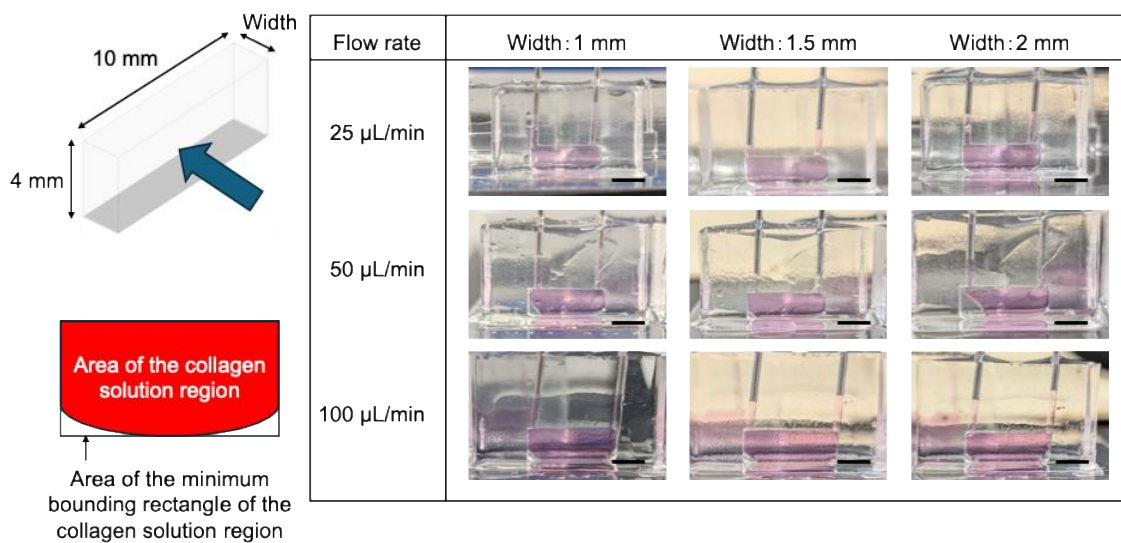
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*Beilstein J. Nanotechnol.* **2026**, *17*, 760–768. [doi:10.3762/bjnano.17.53](https://doi.org/10.3762/bjnano.17.53)

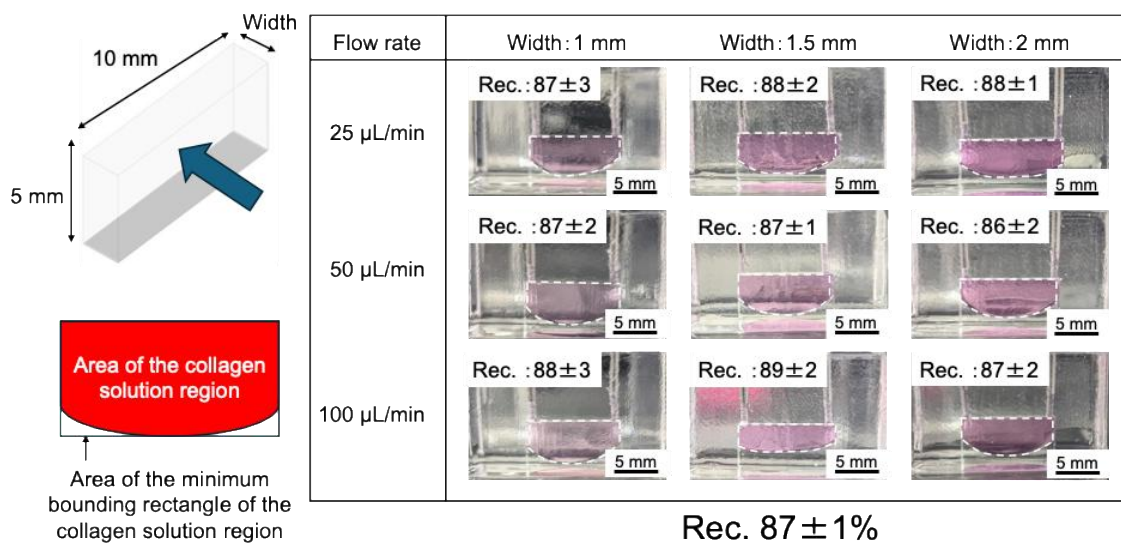
## Additional figures



**Figure S1:** Rectangularity of the collagen solution region with respect to channel dimension ( $7 \times 5 \times \text{width}$ ) and collagen solution introduction flow rate. Collagen region was imaged by smartphone followed by the direction indicated by arrow. Rectangularity is abbreviated as “Rec”.



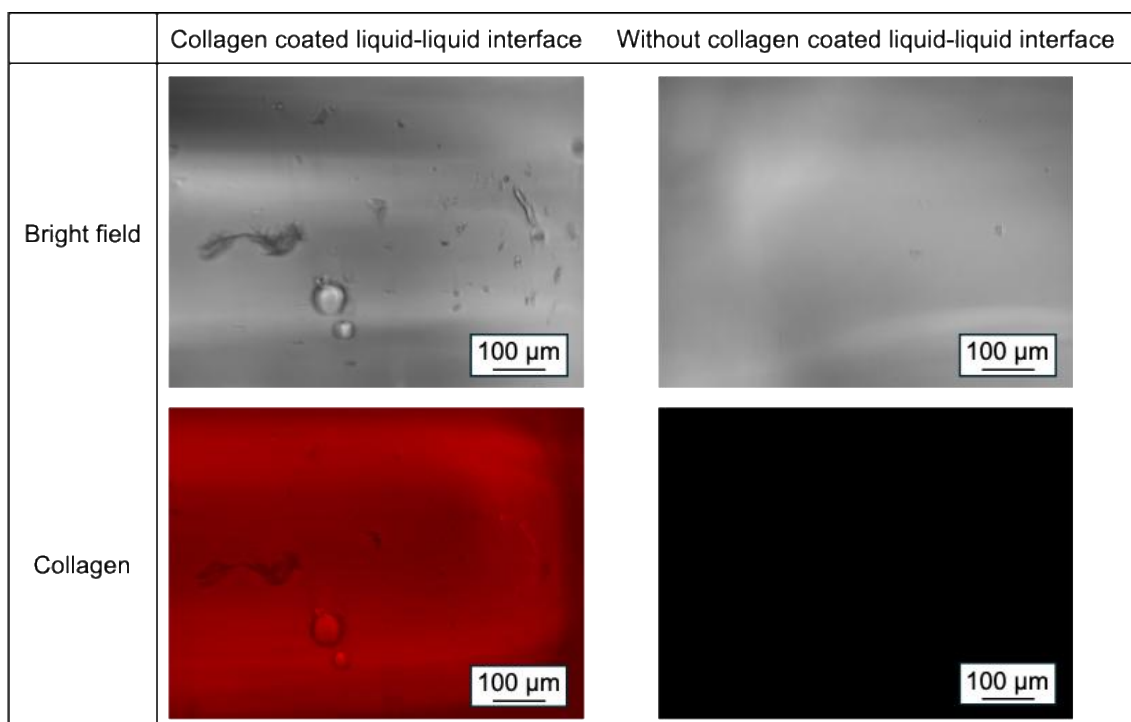
**Figure S2:** The collagen solution region with respect to channel dimension ( $4 \times 10 \times \text{width}$ ) and collagen solution introduction flow rate. Collagen region was imaged by smartphone followed by the direction indicated by arrow. For a channel height of 4 mm, almost all of the FC-43 was replaced by the collagen solution.



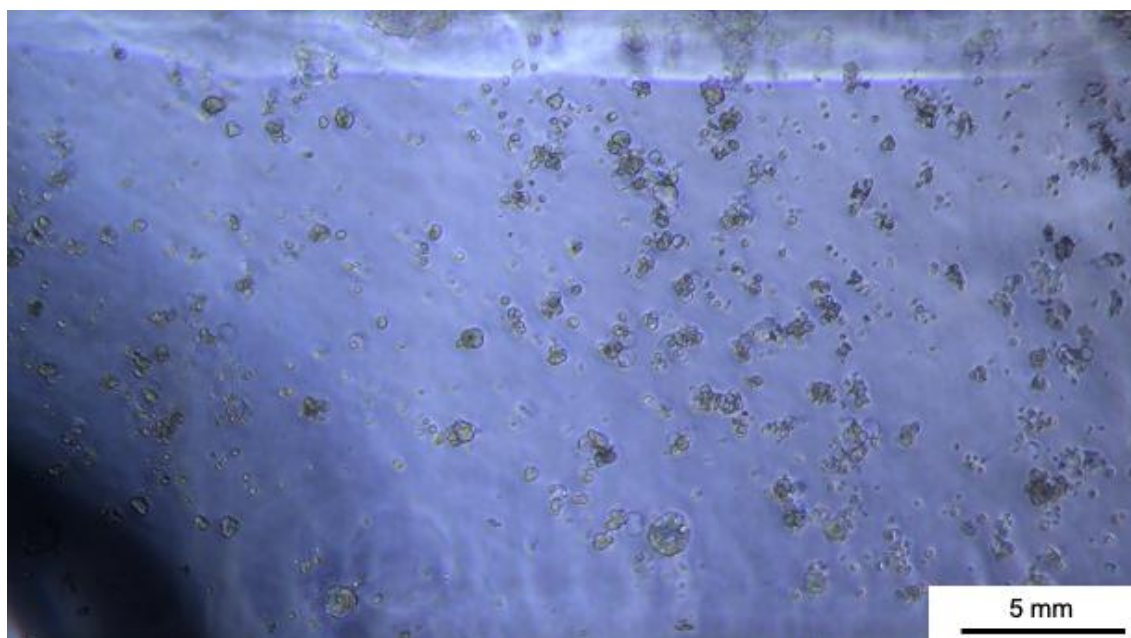
**Figure S3:** Rectangularity of the collagen solution region with respect to channel dimension (5 × 10 × width) and collagen solution introduction flow rate. Collagen region was imaged by smartphone followed by the direction indicated by arrow. Rectangularity is abbreviated as “Rec”.

	Left	Center	Right	Confluency [%]
Device1				93
Device2				99
Device3				94

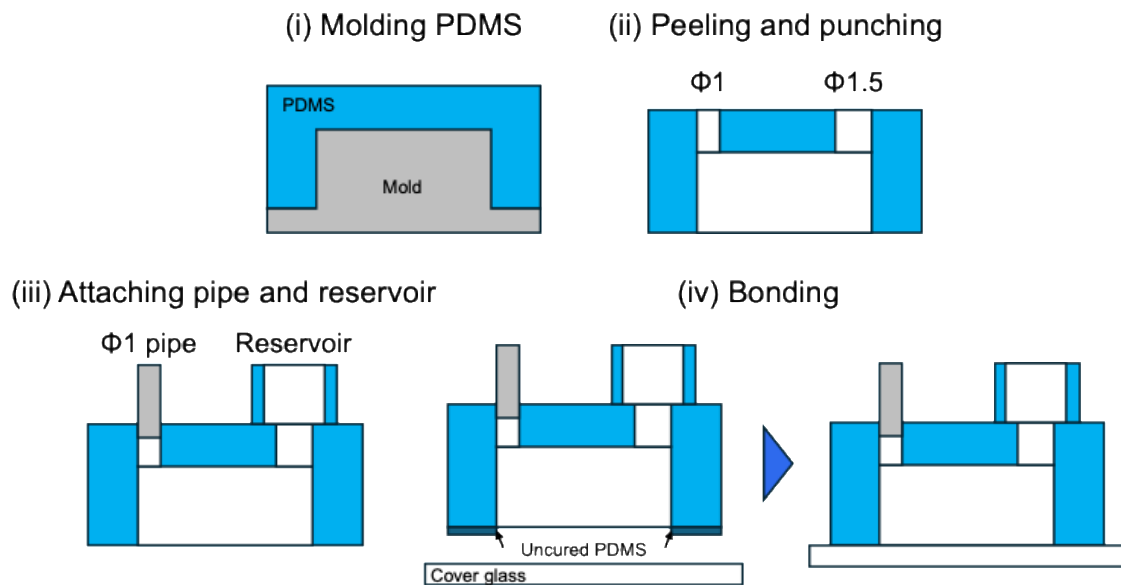
**Figure S4:** Caco-2 cell monolayers formed on the liquid-liquid interface. Images of the left, center and right regions of the channel were acquired on day 7 of culture. The confluency for each independent device is indicated.



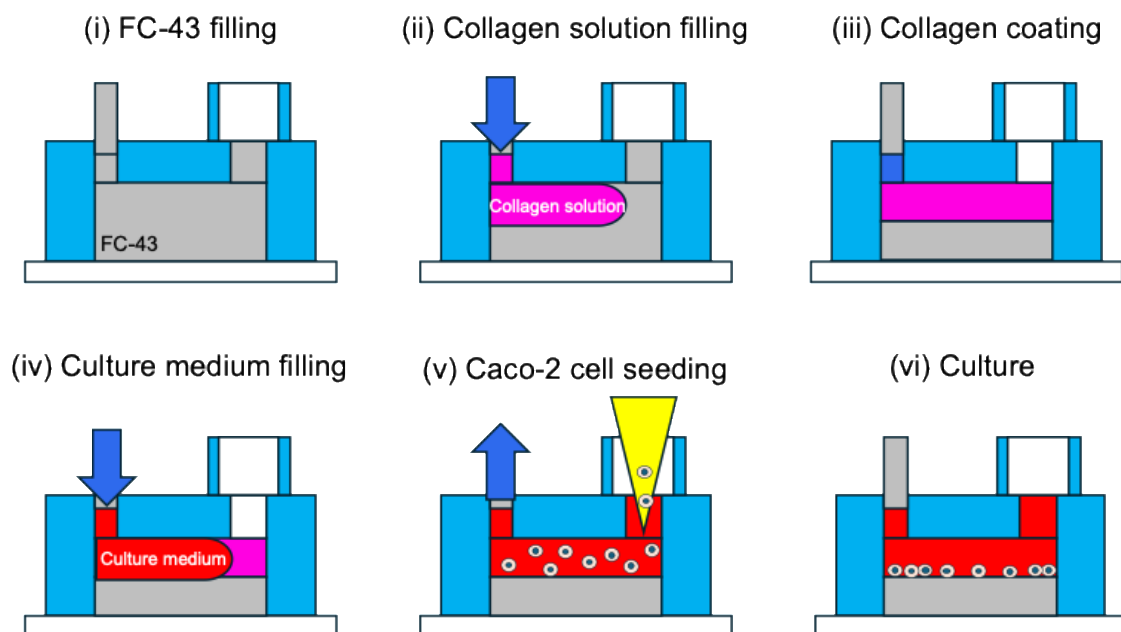
**Figure S5:** Immunofluorescence images of liquid-liquid interface with collagen layer and without collagen layer. The fluorescently labeled anti-collagen antibody emitted red fluorescence by binding to the collagen layer coated on the liquid-liquid interface.



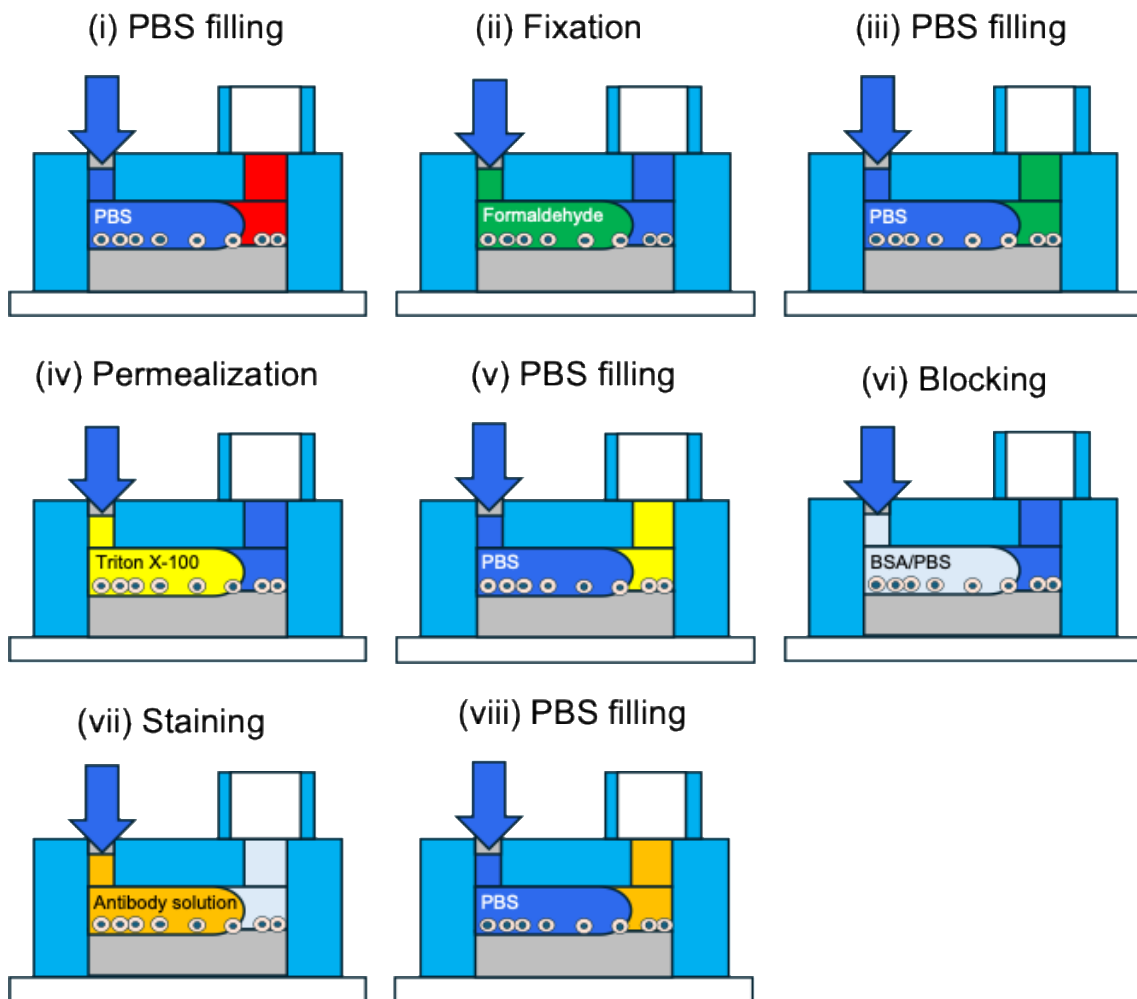
**Figure S6:** Caco-2 cells seeded onto a liquid-liquid interface without collagen coating. The image was captured 2 days post-seeding.



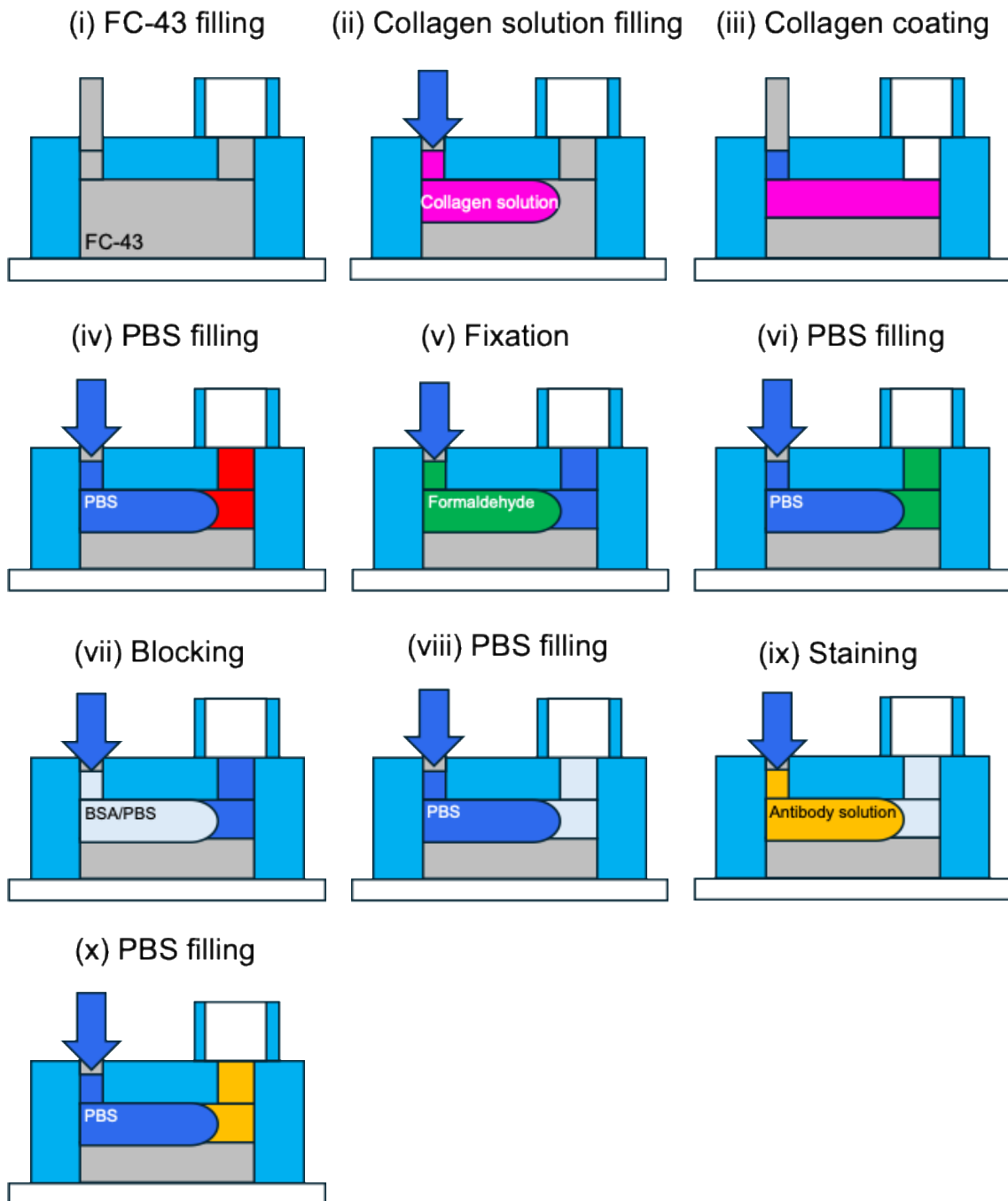
**Figure S7:** Fabrication procedure of the microfluidic device. (i) Molding PDMS. (ii) Peeling and punching. (iii) Attaching pipe and reservoir. (iv) Bonding.



**Figure S8:** Caco-2 cell culture procedure in the microfluidic device. (i) FC-43 filling. (ii) Collagen solution filling. (iii) Collagen coating. (iv) Culture medium filling. (v) Caco-2 cell seeding. (vi) Culture.



**Figure S9:** Immunostaining procedure in the microfluidic device. (i) PBS filling. (ii) Fixation. (iii) PBS filling. (iv) Permealization. (v) PBS filling. (vi) Blocking. (vii) Staining. (viii) PBS filling.



**Figure S10:** Immunostaining procedure for the collagen layer coated on the liquid-liquid interface. (i) FC-43 filling. (ii) Collagen solution filling. (iii) Collagen coating. (iv) PBS filling. (v) Fixation. (vi) PBS filling. (vii) Blocking. (viii) PBS filling. (ix) Staining. (x) PBS filling.