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

Supplementary Information

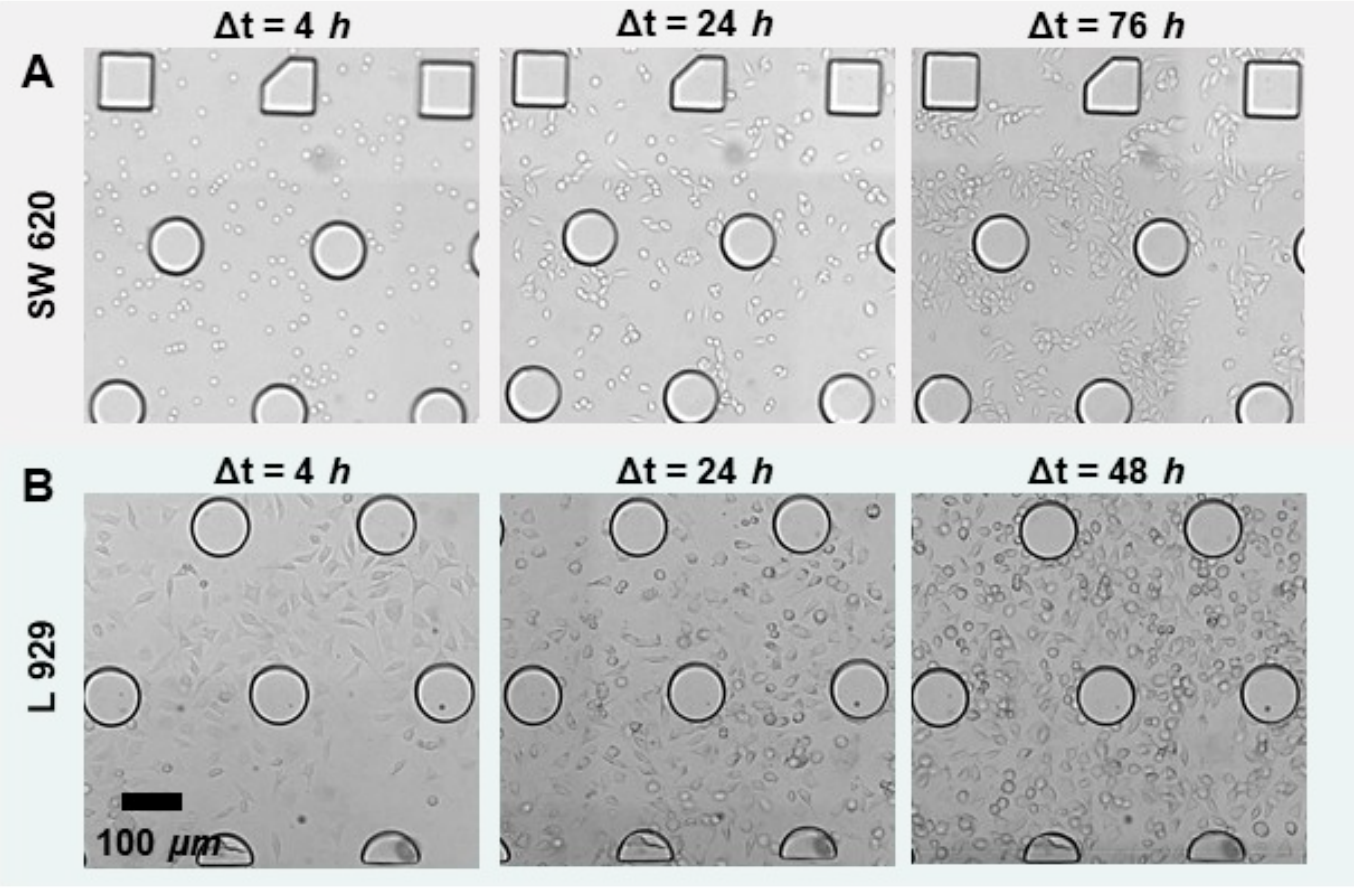

Figure S1 – Cell culture of human colorectal adenocarcinoma SW620 (ECACC, ECACC 87051203) cells and L929 murine fibroblasts (ECACC, ECACC 85011425) at different timepoints. Both cell lines were cultured in Dulbecco's Modified Eagle Medium (DMEM) cell culture medium (Gibco). Culture medium was supplemented with 10% heat inactivated fetal bovine serum (Biowest) and 100 *U/mL* penicillin and 100 *μg/mL* streptomycin (Gibco) and maintained at 37 ^{o}C, 5% CO_2 in a saturated atmosphere.

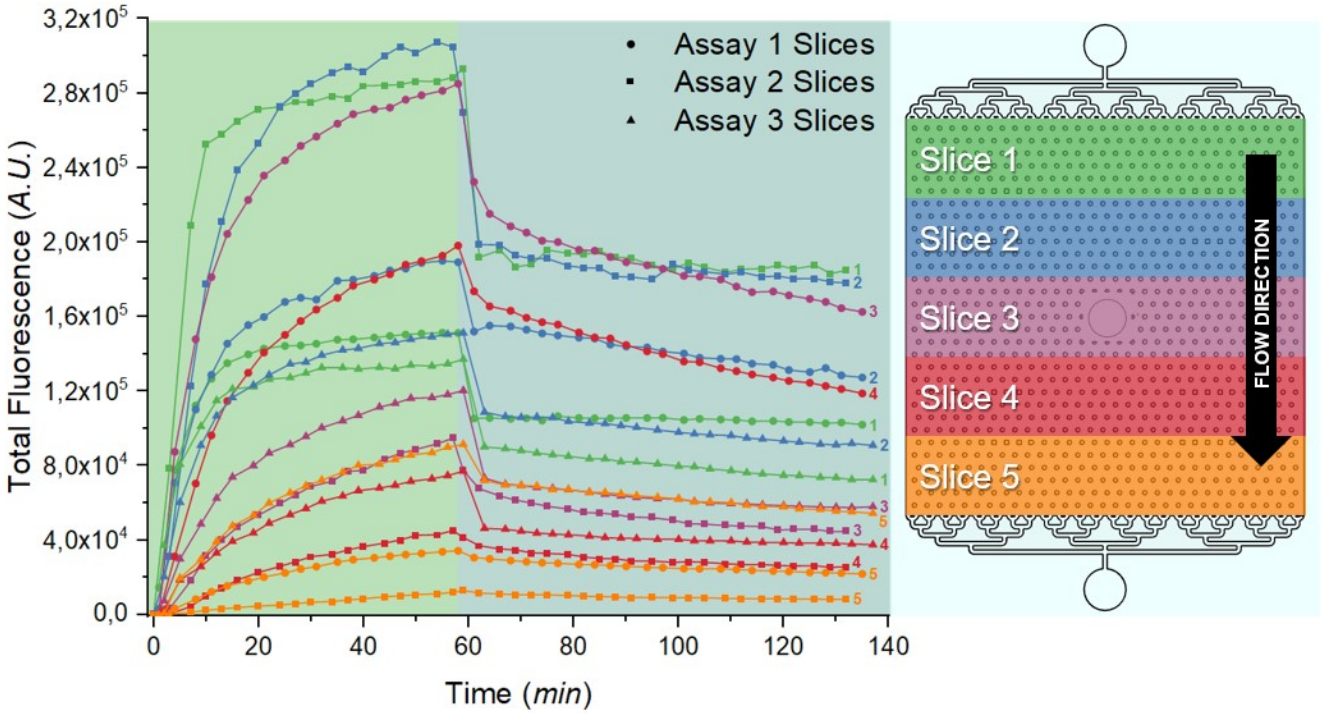

Figure S2 – Fluorescent signal acquired after the injection of anti-CXCR4 labeled phages. The microfluidic device was divided into five slices, numbered from the inlet (1) to the outlet (5). From 0-60 *minutes* it is possible to follow the capture and from 60-140 *minutes* it is possible to see the wash with culture medium. In this figure, it is possible to observe that the first slices, generally, capture more phages with the curves with different behavior being associated with cell distribution heterogeneity.

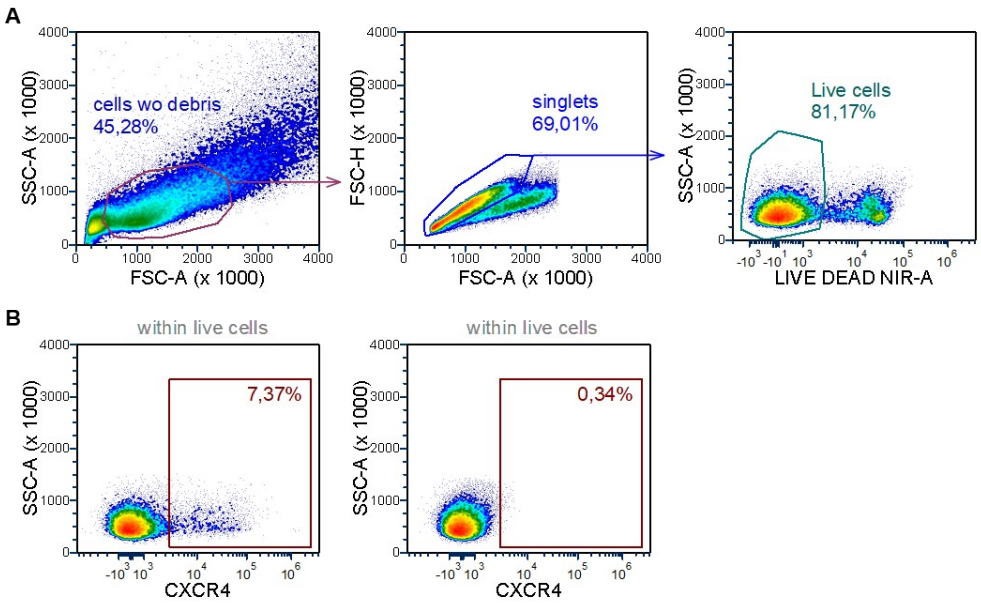

Figure S3 – Gating strategy for flow cytometry data (A) and CXCR4 surface expression in HCT116 cells (B, left panel; right panel is the control without mAb anti-CXCR4).

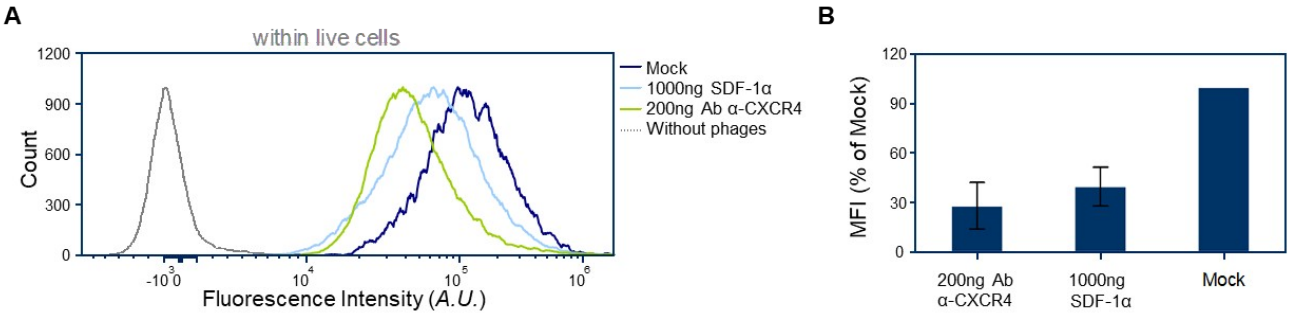

Figure S4 – Phage binding assessment upon SDF-1α (1000 *ng*) or mAb α-CXCR4 (200 *ng*) pre-incubation using flow cytometry. A ratio of 1.0×10^4 phages/cell was used to a total of 2.0×10^5 HCT116 cells. (A) Representative histogram showing fluorescence intensity corresponding to phage binding upon each treatment. (B) Median fluorescence intensity (MFI) for phage binding after treatment with 200 *ng* of mAb α-CXCR4 or 1000 *ng* of SDF-1α, using flow cytometry. Data is represented as mean of fluorescence (±SDs) relative to mock condition

Table S1 – Data input and output from GraphPad Prism 7.0. To calculate the KD, the model Association then dissociation (non linear regression). To use this model the values HotNM (phage concentration in *nM*, row 3) and Time0 (time at which dissociation was initiated, row 7) were manually defined.

Nonlin fit		A	B	C
		Assay 1	Assay 2	Assay 3
		Y	Y	Y
1	Association then dissociation			
2	Best-fit values			
3	HotNM	= 0.9963	= 0.9963	= 0.9963
4	Kon	133276269	120853076	136292771
5	Koff	0.006566	0.004981	0.00667
6	Bmax	742941	989235	598692
7	Time0	= 62	= 62	= 62
8	NS	-125080	-183296	-126560
9	KD	4.927e-011	4.122e-011	4.894e-011
10	Std. Error			
11	Kon	15439436	11115797	35878912
12	Koff	0.0007568	0.0005209	0.00186
13	Bmax	40752	49154	98078
14	NS	44096	53614	108556
15	KD	1.042e-011	7.533e-012	2.546e-011
16	95% CI (profile likelihood)			
17	Kon	107293894 to 164233913	100265212 to 144974475	80986081 to 220261140
18	Koff	0.005199 to 0.008247	0.004001 to 0.006142	0.003772 to 0.01203
19	Bmax	663978 to 826060	894387 to 1093520	436648 to 844686
20	NS	-213499 to -41410	-296827 to -79946	-391079 to 55805
21	Goodness of Fit			
22	Degrees of Freedom	37	37	37
23	R square	0.9287	0.9506	0.768
24	Absolute Sum of Squares	48214610596	45747517622	86277980718
25	Sy.x	36098	35163	48289
26	Constraints			
27	HotNM	HotNM = 0.9963	HotNM = 0.9963	HotNM = 0.9963
28	Kon	Kon > 0	Kon > 0	Kon > 0
29	Koff	Koff > 0	Koff > 0	Koff > 0
30	Time0	Time0 = 62	Time0 = 62	Time0 = 62