

Supporting Information

Capture, Release and Culture of Circulating Tumor Cells in Pancreatic Cancer Patients using an Enhanced Mixing Chip

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Figure S1

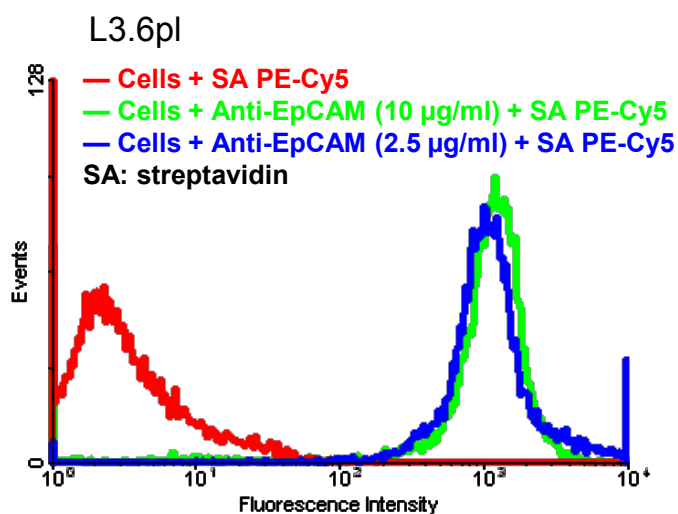


Figure S1. Flow cytometry test of anti-EpCAM binding with L3.6pl cells. Streptavidin phycoerythrin Cy5 (SA PE-Cy5) was used to label the biotinylated anti-EpCAM.

Figure S2

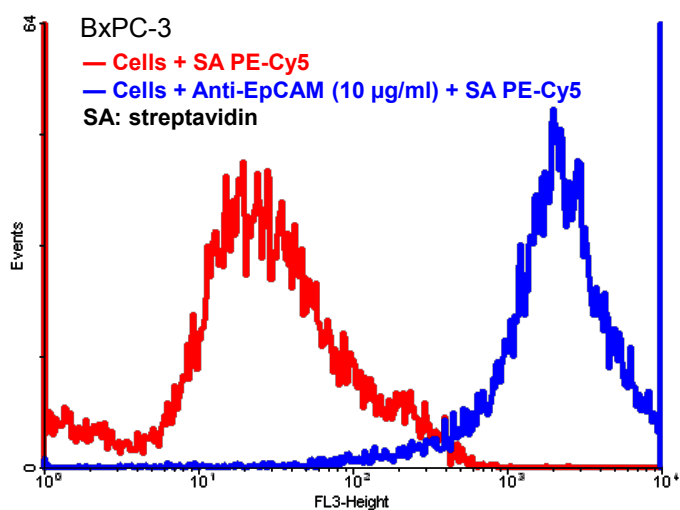


Figure S2. Flow cytometry test of anti-EpCAM binding with BxPC-3 cells.

Figure S3:

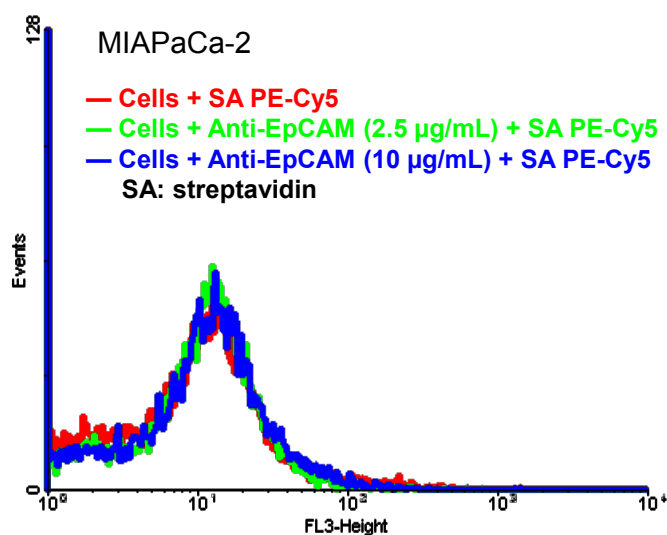


Figure S3. Flow cytometry test of the binding behavior between anti-EpCAM with MIAPaCa-2 cells. Data shows that anti-EpCAM does not bind with MIAPaCa-2 cells, indicating that MIAPaCa-2 cells do not express EpCAM.

Figure S4:

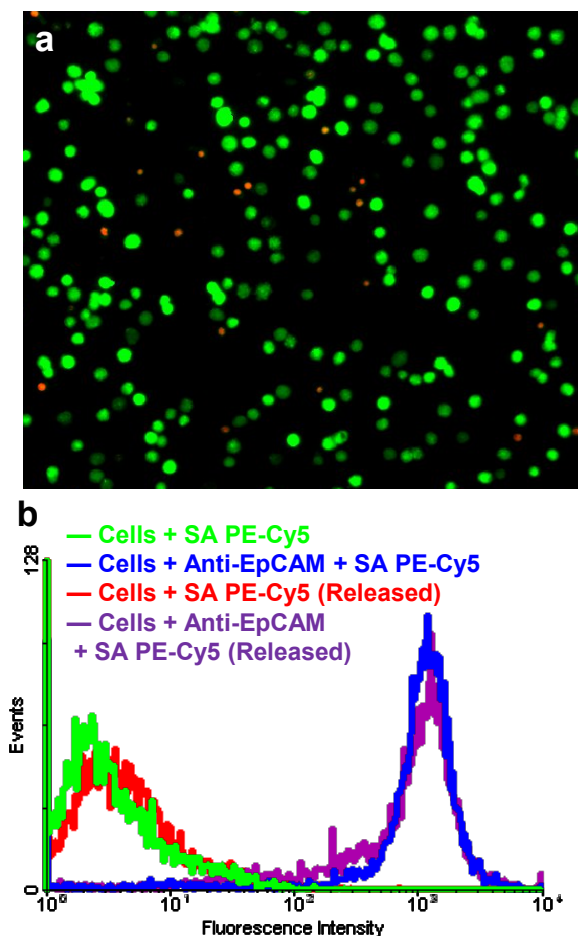


Figure S4. a) Fluorescence image of the L3.6pl cells after capture and release with PI/AO staining. The orange (red merged with green) color indicates nonviable cells (PI and AO staining), while the green color alone indicates viable cells (AO staining alone); b) Flow cytometry test shows that the captured and then released L3.6pl cells maintain their binding capability with anti-EpCAM, without any differences compared to normal L3.6pl cells.

Table S1. Comparison of this work with those in the literature.

References	Device	Capture efficiency	Purity*	Throughput*	Cell viability	Release	Culture
1	CTC chip	65%	~50%	0.5-1 h/mL	$98.5 \pm 2.3\%$	No	No
2	HB-chip	~91.8%	Higher than [1]	~0.83 h/mL	$95\% \pm 0.6\%$	No	No
3	Sinusoidal channel	>97%	NA*	~0.5 h/mL	NA	Yes	No
4	GEDI chip	85-97%	$68 \pm 6\%$	1 h/mL	NA	No	No
5	3D-nanopillar & Mixer	>95%	NA	1 h/mL	84-91%	No	No
This work	GEM chip	>90%	~84%	0.28 h/mL	~89%	Yes	Yes

Note*: 1) NA indicates “not available” or “not applicable”. 2) Throughput is determined by the flow rate, which is inversely proportional to the time required to process sufficient amount of sample that contains detectable number of CTCs. The time required to process 1 mL of sample is listed. 3) Purity varies with the target/control cell ratio and depends on whether obtained from buffer system or whole blood; thus purity here is just for reference not for comparison.

Table S2. Quantification of CTCs and “double positive” cells per mL of blood among 18 samples from patients with metastatic pancreatic cancer.

Sample No.	Cancer type	Volume processed (mL)	Raw number of CTCs	CTCs/mL	“Double positive” cells/mL
1	Pancreas	2	4	2	1
2	Pancreas	4	14	4	0
3	Pancreas	2	9	5	3
4	Pancreas	1	2	2	6
5	Pancreas	2	2	1	1
6	Pancreas	2	0	0	2
7	Pancreas	2	5	3	4
8	Pancreas	1	2	2	0
9	Pancreas	2	4	2	5
10	Pancreas	4	19	5	2
11	Pancreas	2	5	3	0
12	Pancreas	2	4	2	0
13	Pancreas	4	5	1	1
14	Pancreas	4	15	4	3
15	Pancreas	4	16	4	2
16	Pancreas	4	29	7	4
17	Pancreas	2	6	3	1
18	Pancreas	2	2	1	0

Table S3. Quantification of CTCs in healthy donor blood.

Healthy Sample	1	2	3	4	5	6	7	8	9
Number of CTCs	0	0	0	0	0	0	0	0	0

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