



Supporting Information

for *Small*, DOI: 10.1002/sml.201601155

A Size-Selective Intracellular Delivery Platform

May Tun Saung, Armon Sharei, Viktor A. Adalsteinsson, Nahyun Cho, Tushar Kamath, Camilo Ruiz, Jesse Kirkpatrick, Nehal Patel, Mari Mino-Kenudson, Sarah P. Thayer, Robert Langer, Klavs F. Jensen, Andrew S. Liss, and J. Christopher Love**

Supplemental Figures

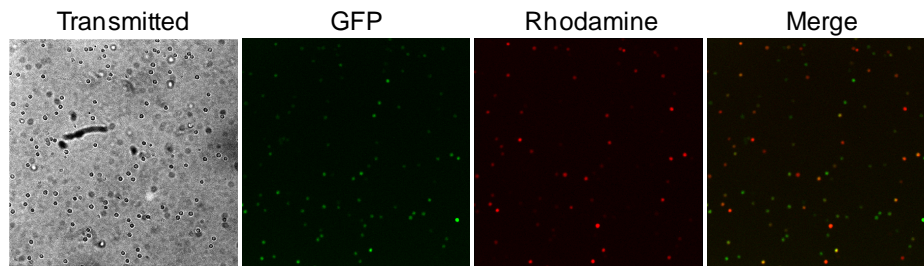


Fig. S1 Intracellular delivery of a dextran fluorophore. GFP-expressing PANC-1 cells spiked in whole blood were delivered rhodamine-conjugated dextran, and isolated by FACS. Representative images of FACS-purified cells are shown.

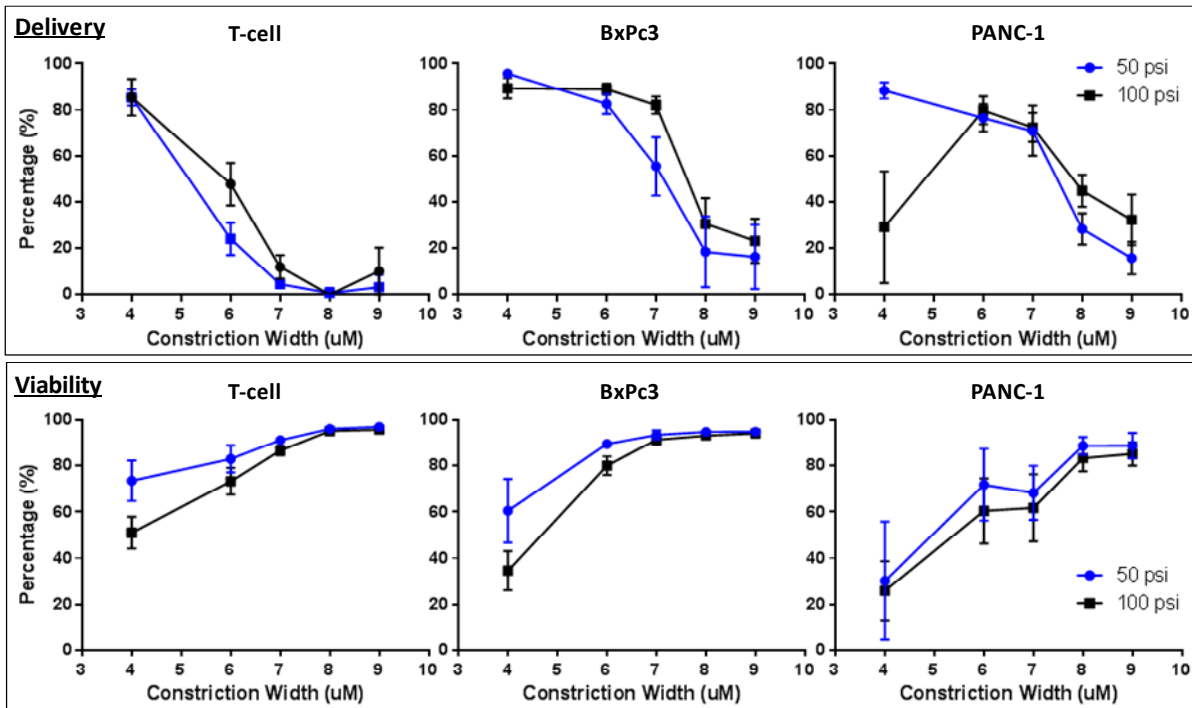


Fig. S2 The effect of pressure on intracellular delivery and cell viability. T-cells, BxPC3 and PANC-1 cells were delivered rhodamine-conjugated dextran with varying pressure (50 vs 100 psi) using platforms with different constriction widths. Percentage delivery (top panel) and cell viability (bottom panel) were determined. The average and standard deviation of at least 3 independent experiments are shown.

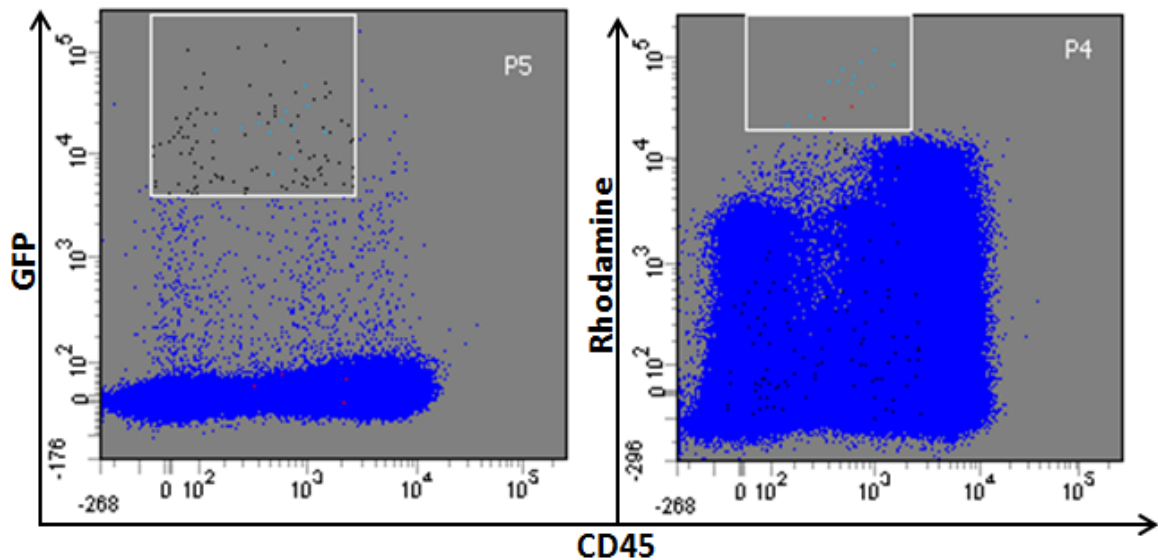


Fig. S3 Specificity of capturing PANC-1 cells from whole blood. GFP-expressing PANC-1 cells spiked into whole blood at low concentration (200 cells/mL). PANC-1 cells tagged with the rhodamine fluorophore were independently verified based on GFP fluorescence. Representative FACS plot demonstrating 75% specificity in capturing PANC-1 cells. The P4 gate [high rhodamine, low CD45 region] was used as a basis for sorting high candidate CTCs, and P5 [high GFP, low CD45 region] was used to sort for GFP-expressing PANC-1 cells. The light blue dots within P5 are accurate hits (i.e. cells that are present within both P4 & P5 gates), such that those are GFP-expressing PANC-1 cells with intra-cellular rhodamine; 75% of the cells within P5 were accurate hits. False positive hits are red, and false negative hits are black. Within a mixture of about 8×10^5 viable cells, 12 GFP-expressing PANC-1 cells were isolated using the size-selective delivery platform.

Sample Sequenced	TP53 (p.R273H)		Kras (p.G12D)	
	Alternate Frequency	Total Reads	Alternate Frequency	Total Reads
Healthy Donor Germline	0.00	180	0.00	113
50 Panc-1 Cells, Flow Sort #1	1.00	45	0.83	476
50 Panc-1 Cells, Flow Sort #2	1.00	32	0.31	391
110 Labeled Cells, 2k / mL Panc-1 Blood Spike	0.97	68	0.63	439
61 Labeled Cells, 200 / mL Panc-1 Blood Spike	0.00*	10	0.31	160
Panc-1 gDNA	1.00	105	0.65	203

Fig. S4 Targeted sequencing results from PANC-1 spiking studies. Targeted sequencing results from cells sorted based on the Rhodamine⁺CD45⁻ gate from spiking studies with PANC-1 cells, including the respective controls. For two genes, TP53 and KRAS, the alternate allele frequency and total depth at each position is presented. *denotes low coverage.