Effective Capture of Circulating Tumor Cells from a Transgenic Mouse Lung Cancer Model using Dendrimer Surfaces Immobilized with anti-EGFR

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Table S1. Dissociation rate constants of PEGylated and dendrimer-coated surfaces functionalized with anti-EGFR, obtained by observing the surface detachment of the captured ED-1 and ED1-SC cells over static agitation. The dissociation rate constants (k_d) were calculated based on the number of the remaining cells on the surface upon agitation, relative to the initial number of bound cells before agitation, indicating relative binding strengths between the cells and the surfaces. Our data show that the binding between the cancer cells and the anti-EGFR-dendrimer-immobilized surface is substantially more stable than that on the anti-EGFR-PEG-coated surface.

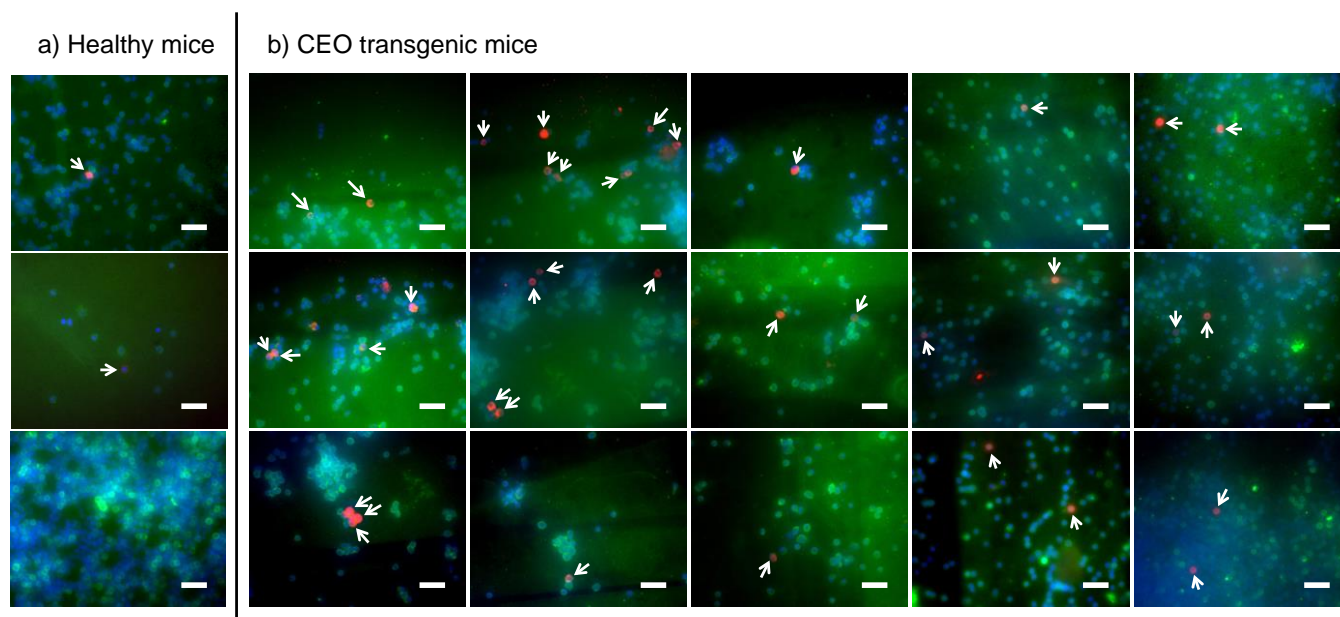| Cell lines	k_d [min^{-1}]		Fold enhancement
	PEG	G7	
ED-1	2.81	1.89	1.5
ED1-SC	1.14	0.89	1.4

Table S2. Histology results and the measured numbers of CTCs captured from blood samples of healthy controls and transgenic mice.

a) Healthy Controls

Identification #	CTC#	Histology	Age
C1	1.14	Normal lung	~ 3 months
C2	3.22	Normal lung	~ 3 months
C3	6.67	Normal lung	~ 6 months
C4	7.6	Normal lung	~ 6 months
C5	6.75	Normal lung	~ 6 months
C6	0.75	Normal lung	~ 6 months

b) Transgenic Mice

Identification #	CTC#	Histology	Age	Gender
1	74	Macroscopic tumor	~ 28 months	Female
2	29.67	Macroscopic tumor	~ 21 months	Male
3	142.75	Macroscopic tumor	~ 26 months	Male
4	50.22	Macroscopic tumor	~ 25 months	Female
5	110.33	Macroscopic tumor	~ 28 months	Female
6	45.57	Macroscopic tumor	~ 9 months	Male
7	74.78	Macroscopic tumor	~ 9 months	Male

| a) Healthy mice | b) CEO transgenic mice |

Figure S1. Representative fluorescence images of the captured CTCs from the blood samples of (a) healthy and (b) transgenic mice using an anti-EGFR-G7 dendrimer-immobilized surface. CTCs are identified as cytokeratin-positive (red fluorescence), CD45-negative (green fluorescence), and DAPI (blue fluorescence)-positive cells, which are marked with white arrows on the images. Leukocytes are defined as cytokeratin-negative, CD45-positive, and DAPI-positive cells. The images show that significantly more CTCs are captured from the transgenic mouse blood, compared to the healthy control. Scale bar = 100 µm

Table S3. Histology results and the measured numbers of CTCs captured from blood samples of vehicle-treated transgenic mice, control locked nucleic acid-treated transgenic mice, and anti-miR-31 locked nucleic acid-treated transgenic mice.

a) Vehicle-treated transgenic mice (Control)

Identification #	CTC#	Histology
1	6	Focal dysplasia, mild lymphocytic pneumonitis
2	3.11	Normal lung
3	55.11	Papillary adenocarcinoma, grade 2
4	23.67	Normal lung
5	2.67	Normal lung

b) Control locked nucleic acid (C-LNA)

Identification #	CTC#	Histology
1	86.57	Normal lung
2	48.33	Papillary adenocarcinoma, grade 2
3	60.44	Papillary adenocarcinoma, grade 2
4	19.56	Normal lung
5	16.33	Normal lung

c) Anti-miR-31 locked nucleic acid (a31-LNA)

Identification #	CTC#	Histology
1	7.5	Mild lymphocytic pneumontis
2	2.67	Normal lung
3	3.56	Carcinoid vs adenocarcinomama, grade 1
4	7.13	Normal lung
5	11.11	Normal lung
6	19.67	Normal lung