

SUPPLEMENTAL MATERIAL

Tran et al., <http://www.jem.org/cgi/content/full/jem.20130110/DC1>**Table S1.** Summary of IHC staining for FAP in various human tumors

Sample ID	Cancer type	Tumor characteristics	% stroma	% of stromal cells that are FAP+	FAP intensity
2230	Melanoma	Metastatic	30	>50	3+
3436-1	Melanoma	Metastatic	10	>50	2+
3581	Melanoma	Metastatic	50	>50	3+
3584	Melanoma	Metastatic	20	>50	1-2
3591-2	Melanoma	Metastatic	70	>50	3+
*3593	Melanoma	Metastatic	20	>50	2-3+
3612	Melanoma	Metastatic	20	>50	3+
3614	Melanoma	Metastatic	30	>50	3+
3620	Melanoma	Metastatic	80	>50	3+
0090	Colorectal adenocarcinoma	Moderately differentiated	40	>50	3+
*0212	Colorectal adenocarcinoma	Poorly differentiated metastatic	60	>50	3+
0403	Colorectal adenocarcinoma	Moderately differentiated	30	>50	3+
0663	Mammary carcinoma	Mucinous	30	>50	2+
0879	Mammary carcinoma	Invasive ductal	90	<50	3+
*0951	Mammary carcinoma	Invasive ductal	60	>50	3+
0264	Pancreatic adenocarcinoma	Poorly differentiated	90	>50	2-3+
*0586	Pancreatic adenocarcinoma	Moderately differentiated	80	>50	3+
0986	Pancreatic adenocarcinoma	Moderately differentiated	60	>50	3+

*Shown in Fig. 1 (A-D).

Table S2. Genetic sequence of the codon-optimized FAP5-CAR construct

CAR component	Sequence (5'–3')
5' flanking sequence (contains XhoI restriction site, Kozak sequence, and start codon)	CTCGAGCCGCCACCATGGTT
Human GM-CSFR leader sequence	TTGCTTGTGACCAGCCTCCTGCTCTGTGAACTGCCTCATCTGCACTTCTGTTGATCCCA
FAP5 scFv heavy chain	CAGGTGCAGTCCAGCAGAGTGCGCAGAGCTCGCTCGCCAGGCGCTTCTGTGAATCTGAGTTGTAAGG CCTCCGGATATACTTTACGAACAACGGCATCACTGGCTGAAGCAGCGGACCGGCCAGGGCCTGGAGTG GATCGGCGAAATATACCCCGGTCACAAACACTCTCTATAACGAGAAGTTAAGGGCAAAGCAACTCTGA CCGCGGACAGGTCCTTAACACAGCCTATATGGAGCTGAGAAGCITGACGAGTGAGGACTCCGCTGTCTAT TTTTGCGCCGAACTCTGACCGCTCTTTTGGTGGGGCCAGGGCAGCTCGTGACCGTAAGTGCG
218 linker	GGCTCCTAGCGGTTCCGGCAAACCTGGCAGCGGAGAAGGCAGCACAAAGGG
FAP5 scFv light chain	CAGATCGTCTGACGAGTCTCCAGCCATCATGAGCGCCTCACCCGGCGAAAAGGTGACCATGACCTGCTC AGCCTCTTCTGGTGTGAATTCATGCACTGGTACCAGCAAAAAAGTGGGACCTCCCTAAAAGGTGGATCTT CGATACCAGCAAACCTGGCTTCTGGCGTCCCGCAAGGTTAGCGGCTCTGGTCCGGCACATCATAAGCCT GACGATCAGCAGCATGGAGGCAGAAGACGACGCTACTTACTGCCAGCAATGGAGCTTTAACCCACCTA CTTTCGGAGGAGGAACAAAGCTGGAAATAAAAAGA
Joining sequence (contains NotI restriction site)	GCGGCCGCA
Mouse CD8 hinge and transmembrane domain	ACTACTACCAAGCCAGTGTGCGAACTCCCTCACCTGTGCACCCTACCGGGACATCTCAGCCCCAGAGACC AGAAGATTGTCGGCCCCGTGGCTCAGTGAAGGGGACCGGATTGGACTTCGCCTGTGATATTTACATCTGGG CACCCTTGGCCGAACTCTGCGTGGCCCTTCTGCTGTCCITGATCATCACTCTCATCTGCTACCACAGGAGCCGA
Mouse CD28 signaling domain	AATAGTAGAAGGAACAGACTCCTCAAGTACTACCATGAACATGACTCCCGGAGGCCTG GGCTCACTCGAAAGCCTTACCAGCCCTACGCCCTGCCAGAGACTTTGCAGCGTACCGCCCC
Mouse 4-1BB signaling domain	AAATGGATCAGGAAAAATCCCCACATATTCAAGCAACCATTTAAGAAGACCACTGGAGCAGCTC AAGAGGAAGATGCTGTAGCTGCCGATGTCCACAGGAAGAAGAAGGAGGAGGAGGCTATGAGCTG
Mouse CD3-ζ signaling domain	AGAGCAAATTCAGCAGGAGTGACAGAGACTGCTGCCAACCTGCAGGACCCCAACCAGCTCTACAATGAGC TCAATCTAGGGCGAAGAGAGGAATATGACGCTTGGAGAAGAAGCGGGCTCGCGATCCAGAGATGGGAG GCAAACAGCAGAGGAGGAGGAACCCCAAGGAAGGCGTATACAATGCACTGCAGAAAAGACAAGATGGCA GAAGCCTACAGTGAGATCGGCACAAAAGGCGAGAGGCGGAGAGGCAAGGGGCACGATGGCCTTTACCA GGGTCTCAGCACTGCCACCAAGGACACCTATGATGCCCTGCATATGCAGACCCTGGCCCTCGCTAA

The FAP5 scFv sequence was obtained from the United States Patent Application Publication: US 2009/0304718 A1. The variable heavy chain sequence comprises of amino acids 20 to 136 of SEQ ID NO: 1 from the patent application. The variable light chain sequence comprises of amino acids 23 to 129 of SEQ ID NO: 2 from the patent application.

Table S3. Genetic sequence of the codon-optimized Sibro-CAR construct

CAR construct	Sequence (5'–3')
5' flanking sequence (contains XhoI restriction site, Kozak sequence, and start codon)	CTCGAGCCGCCACCATGGTT
Human GM-CSFR leader sequence	CTCTTGTAACCAAGTCTGTTGCTCTGCGAACTGCCCAACCCGGCCCTCCTCTTGATTCCG
Sibrotuzumab scFv heavy chain	CAGGTACAGCTTGTGCAGTCCGGCGCTGAGGTCAAAAAACCAGGCGCCAGCGTTAAGGTGTCATGCAAG ACTAGCAGGTATACATTCAGTGAATACACAATACACTGGGTGCGACAGGCTCCCGGGCAAAGACTGGAGT GGATTGGGGGCATAAACCCCAACAATGGGATCCCGAATTATAATCAGAAATTAAGGGTCGGGTGACAAT CACAGTAGACTAGCGCATCAACCGCTACATGGAGCTCAGTCCCTTAGGTCTGAAGACACAGCAGTT TATTACTGCGCAAGGCGCCGCATCGCTACGGCTATGACGAAAGGTCATGCTATGGACTATTGGGGGCAAG GGACACTCGTGAAGTGTCTCATCA
218 linker	GGAAGTACTAGTGGCTCAGGAAAGCCCGGATCCGGGGAAGGATCTACGAAGGGG
Sibrotuzumab scFv light chain	GACATTGTCATGACGAGAGTCTGACAGTCTCGCCGTGCTCTTGGGAGAGCGGGCAACAATCAATTGTA AAAGCAGCCAATCTCTCTGTACAGCCGGAACCAGAAAGAACTACCTCGCCTGGTACCAGCAGAAACCCG GTCAGCCGCCAAGCTCCTGATTTTTGGGCCAGCACTCGGAAAGCGGTGCCCCGACAGATTTCCGG ATCTGGCTTCGGGACAGATTTTACCCTGACGATCTCATCACTCAGGCAGAGGATGTGGCCGCTACTACTG TCAACAGTATTTTCTACCCCTCACTTTCGGCCAAGGAACCAAGGTGGAGATTA
Joining sequence (contains NotI restriction site)	GCGGCCGCA
Human CD8 hinge and transmembrane domain	TTCGTGCCGGTCTTCTGCCAGCGAAGCCACCACGACGCCAGCGCCGCGACCAACAACCCGGCGCCC ACCATCGCGTCGACGCCCTGTCCCTGCGCCAGAGGCGTGCCGGCCAGCGGGGGGGCGCAGTGC ACACGAGGGGGCTGGACTTCGCCTGTGATATCTACATCTGGGCGCCCTTGGCCGGGACTTGTGGGTCT TCTCTGTCACTGGTTATCACCCCTTACTGCAACCACAGGAAC
Human CD28 signaling domain	AGGAGTAAGAGGAGCAGGCTCTGCACAGTGACTACATGAACATGACTCCCCGCGCCCCG GGCCACCCGCAAGCATTACCAGCCCTATGCCCCACCACGCGACTTCGACGCTATCGCTCC
Human 4-1BB signaling domain	CGTTTCTGTGTTAAACGGGGCAGAAAGAACTCCTGTATATTTCAAACAACCATTTATGAGACCA GTACAAACTACTCAAGAGGAAGATGGCTGTAGCTGCCGATTTCCAGAAGAAGAAGAGGAGGATGTGAAGT
Human CD3-ζ signaling domain	AGAGTGAAGTTCAGCAGGAGCGCAGACGCCCCCGCTACCAGCAGGGCCAGAACCAGCTCTATAACGA GCTCAATCAGGACGAAGAGAGGAGTACGATGTTTTGGACAAGAGACGTGGCCGGGACCCTGAGATGG GGGAAAGCCGAGAAGGAAGAACCTCAGGAAGGCCTGTACAATGAACTGCAGAAAGATAAGATGGC GGAGGCTACAGTGAGATTGGGATGAAAGGCGAGCGCCGAGGGGCAAGGGGCACGATGGCCTTAC CAGGGTCTCAGTACAGCCCAAGGACACCTACGACGCCCTTACATGCAAGGCCCTGCCCCCTCGCTAA

The Sibrotuzumab (BIB1) scFv sequence was obtained from the United States Patent Application Publication: US 2003/0103968 A1. The variable heavy and light chain sequences are derived from SEQ ID NO: 12, and SEQ ID NO: 2 of the patent application, respectively.