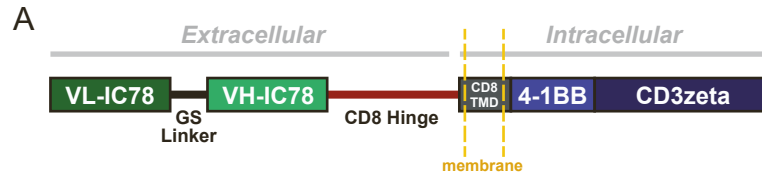


Supplemental information

**Preclinical specificity & activity of a fully
human 41BB-expressing anti-CD19 CART-
therapy for treatment-resistant autoimmune disease**

Binghao J. Peng, Andrea Alvarado, Hangameh Cassim, Soprina Guarneri, Steven Wong, Jonathan Willis, Julia SantaMaria, Ashley Martynchuk, Victoria Stratton, Darshil Patel, Chien-Chung Chen, Yan Li, Gwendolyn K. Binder, Rebecca Dryer-Minnerly, Jinmin Lee, and Samik Basu



B

QAVLTQPPSVSEAPRQRVTISCSGSSSNIGNNAVSWYQQLPGKAPKLLIYYDLLPSGVSDRFSGSKSGTSASLAISGLQSEDEADYYCAAWDDSLNGWVF
 GGGTKVTVLGGGGGGGGGGGGSEVQLVQSGAEVKKPGESLKISCKGSGYSFTSYWIGWVRQMPGKGLEWMGIIPGDS DTRYSPSFQGGQVTISADK
 SISTAYLQWSSLKASDTAMYCARLSYSWSSWYWDFWGQGLTVTVSS

Figure S1. CABA-201. (A) Schematic of CABA-201 CAR construct. (B) Protein sequence of IC78 binder used in CABA-201 CAR construct.

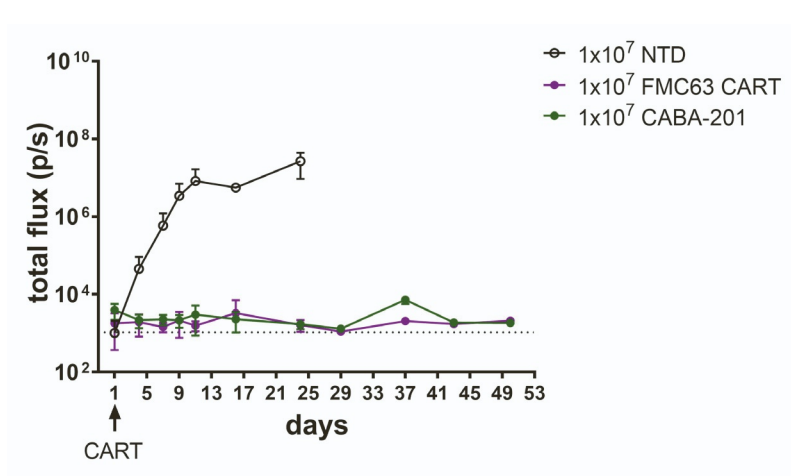


Figure S2. CABA-201 demonstrates activity in vivo. Luciferase-expressing Nalm6 cells (1×10^6 cells per mouse) were injected intravenously into NSG mice, followed 5 days later by injection with 1×10^7 NTD T-cells, FMC63 CART cells, or CABA-201 cells. Bioluminescence imaging quantification of Nalm6 CD19⁺ cells after FMC63 CART, CABA-201 or NTD treatment for days 1, 4, 7, 9, 11, 16, 24, 29, 37, 43, and 50 were plotted as mean \pm SD for each group (n=12 for NTD and n=18 for FMC63-CART and CABA-201 group). NTD T-cells treated group mice were terminated on day 28 for humane reasons.

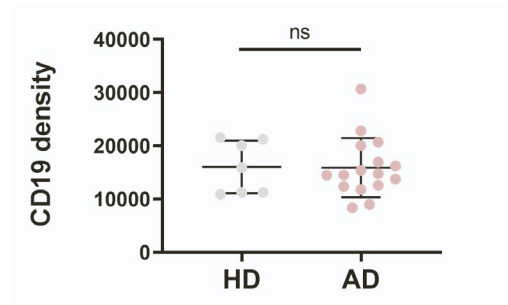


Figure S3. CD19 surface expression is comparable between healthy and autoimmune disease donors. CD19 surface expression on isolated B cells from healthy donors (n=7) and various autoimmune disease patients (n=16) were quantified using phycoerythrin (PE) quantification beads (see Methods). Each dot represents CD19 density (number of CD19 per cell) per donor. Mean \pm SD are shown for HD (healthy donors) and AD (autoimmune disease patients). Autoimmune disease indication includes SLE (n=4), RA (n=9), and chronic inflammatory demyelinating polyneuropathy (CIDP, n=3). Statistical differences between HD and AD were determined by Mann-Whitney test. ns, not significant.

Table S1. Transduction and expansion of CABA-201 cells derived from autoimmune disease donors. Transduction efficiency of CAR and cell expansion during generation of CABA-201 cells from multiple autoimmune disease patient materials and types are shown. Donors with less than 5-fold cell expansions were largely due to poor quality of frozen PBMC samples.

Donor ID	Indications	Transduction Efficiency (%)	Cell expansion (fold)
mcPV Donor 1	Mucocutaneous pemphigus vulgaris	73.8	3.4
mcPV Donor 2		65.9	6.8
SLE Donor 1	Systemic lupus erythematosus	75.8	0.5
SLE Donor 2		67.5	5.22
SLE Donor 3		80.7	5.46
MS Donor 1	Multiple sclerosis	68.2	15.9
RA Donor 1	Rheumatoid arthritis	56.1	12.5
RA Donor 2		38.2	7.9
SSc Donor 1	Systemic sclerosis	14.6	7.4
SSc Donor 2		25.8	20.8
Myositis Donor 1	Idiopathic inflammatory myopathies	38.1	2.5
Myositis Donor 2		55.0	13.4
Myositis Donor 3		47.2	7.0
Myositis Donor 4		74.5	2.1
Myositis Donor 5		52.8	22.1