Supplemental Information

Adenovirus-Antibody Complexes

Contributed to Lethal Systemic

Inflammation in a Gene Therapy Trial

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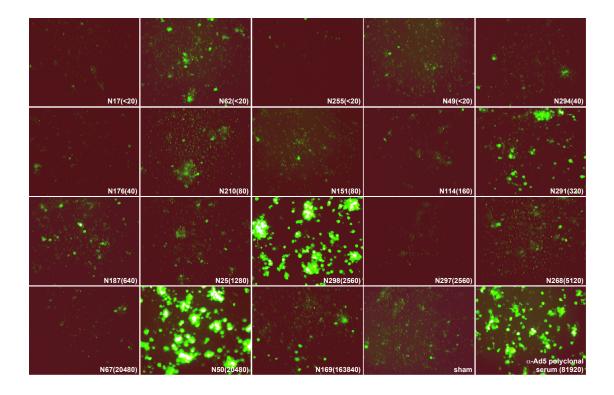


Figure S1. Pre-existing NAb levels do not correlate with enhancement of DC transduction, see Table 1. Human serum from individuals with a range of NAb titers were pre-complexed with Ad5-expressing GFP before being added to cells. We imaged the cells 48 h later with an inverted fluorescent microscope equipped with a digital camera. Each panel shows the serum identification numbers and the levels of NAb titer (within brackets).

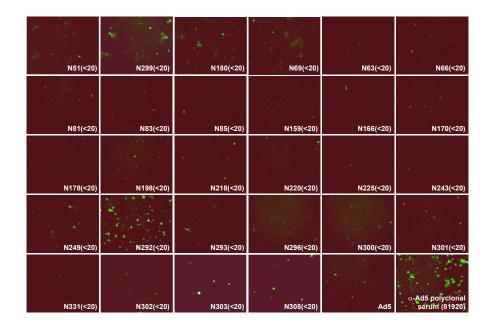


Figure S2. Screening of naïve human serum for enhancement of DC transduction, see Table 1. Human sera lacking NAbs (titers <20) were screened for enhancement of DC transduction by Ad5. We imaged cells 48 h later using an inverted fluorescent microscope equipped with a digital camera. The numbers inside each panel show the serum identification number and the levels of NAb titer (within brackets).

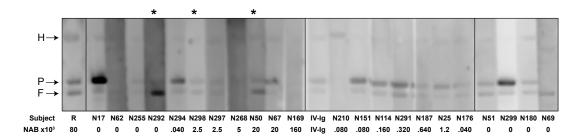


Figure S3. Immunoblotting of serum samples from healthy donors, see Table 1. Ad5 proteins were resolved on a 4–12% polyacrylamide gel and probed with serum from healthy donors. Serum sample identification numbers and the NAb titer are provided below the gel. Arrows indicate Ad5 hexon (H), penton (P), or fiber (F) bands. Reactivity with rabbit antiserum (R) is also shown on the blot. Digital images of blots performed under similar conditions were spliced together to generate the composite image. Vertical lines separate the different experiments. (*) Indicates serum samples that enhance transduction.

Supplemental Table 1. Activation of Human DCs by Whole Blood from Human Subjects.

Analyte	Donor	Cells	Ad5	B1	B2	В3	B4	B5	B6	019
GFP (MFI)	1	1	8		58				41	147
	2	1	22	83	68	83	51	49	82	158
	3	1	18		73				82	177
	4	1	38	107	65	102	113	91	106	132
CD80 (Fold Change)	1	1	1.7		2.5				2.4	2.5
	2	1	2.4	1.7	3.4	2.3	2.1	1.5	2.3	4.6
	3	1	1.4	1.6	1.9			1.6	1.6	2.5
	4	1	1.6	1.6	1.6	1.3	1.3	1.4	1.1	2.6
	1	205	335		675				730	926
IL-6	2	149	1338	1425	1435	1649	1893	1924	1861	2513
(pg/ml)	3	0	204	181	245			160	185	1690
	4	0	587	680	338	833	1103	829	1246	1558