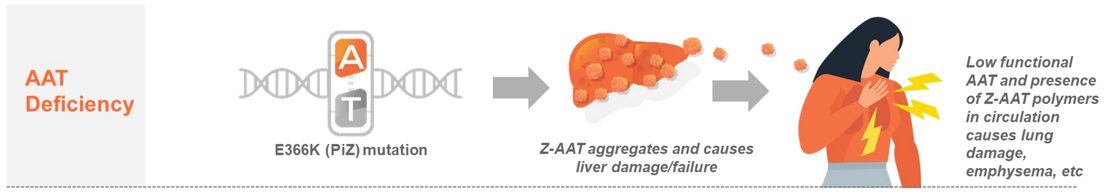
BEAM-302 decreases hepatic aggregates of mutant AAT and increases circulating functional AAT in rodent models of Alpha-1 Antitrypsin Deficiency

Ayan Banerjee, Lo-I Cheng, Dominique Leboeuf, Brianna Bannister, Steven Boule, Courtney Sawyer, Valerie Winton, Jerry Decker, Carrie Curtis, Dongyu Chen, Karen Cao, Delai Chen, Bo Yan, Ronak Shah, Yi Yu, Sarah Smith, Michael Packer, Giuseppe Ciaramella

Liver and Lung Manifestations of Alpha-1 Antitrypsin Deficiency



Could correction of the PiZ mutation via BEAM-302 lipid-nanoparticle-mediated base editing in the liver resolve both liver and lung pathologies[1]?

Precise Correction and Bystander Editing in PiZZ patient fibroblasts

Figure 1 Allele frequencies assessed by high-throughput targeted amplicon sequencing in primary PiZZ fibroblasts (GM11423, Coriell) transfected with base editor mRNA and gRNA. In addition to precise correction (7G), a significant amount of linked bystander editing (5G+7G) was also observed. 7G editing results in WT protein; 5G+7G editing results in expression of D365G protein.

D365G AAT Protein Binds and Inhibits Elastase *In Vitro*

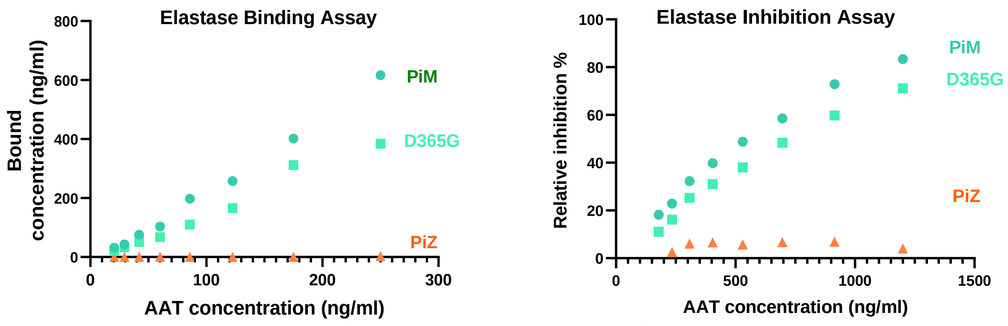


Figure 2. Elastase binding and inhibition assay with recombinant AAT protein (PiM, D365G and PiZ)

In Vivo Correction of the PiZ Mutation with Base Editing in Mice

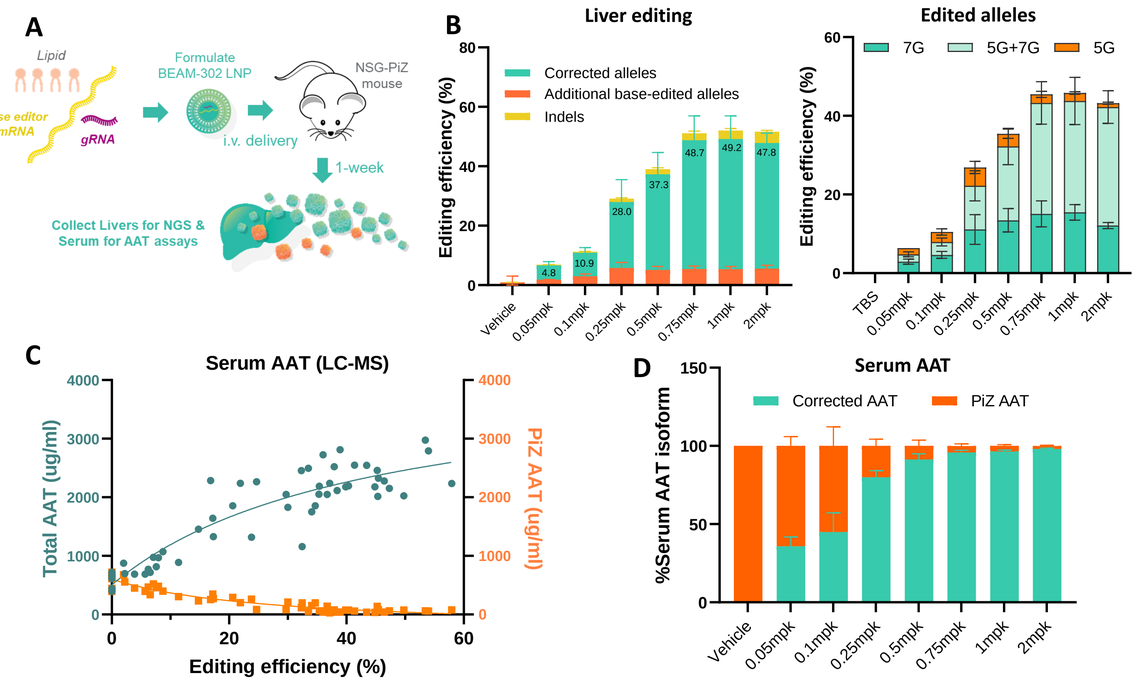


Figure 3 A. Experimental design with BEAM-302 LNP dosed intravenously in adolescent NSG-PiZ mice. **B.** Relative frequencies of base-edited alleles in the liver of NSG-PiZ mice after single administration of increasing dose levels of BEAM-302. **C.** Relationship between liver base editing and serum concentrations of total AAT and PiZ AAT as measured by liquid chromatography-mass spectrometry (LC-MS). **D.** Percentages of corrected AAT and PiZ AAT proteoforms.

In Vivo Correction Increases Functional Human AAT in Serum

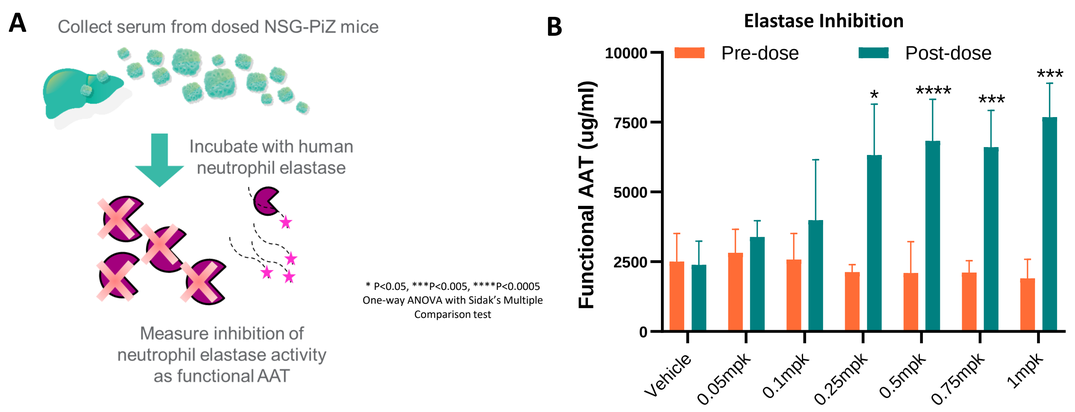


Figure 4 A. Schematic of neutrophil elastase inhibition assay used to determine levels of functional AAT in mouse serum. **B.** Functional AAT levels reflecting neutrophil elastase inhibitory capacity in serum of NSG-PiZ mice collected pre-dose or one-week after a single administration of increasing dose levels of BEAM-302.

In Vivo Correction Reduces PiZ Globule Burden in Mouse Liver

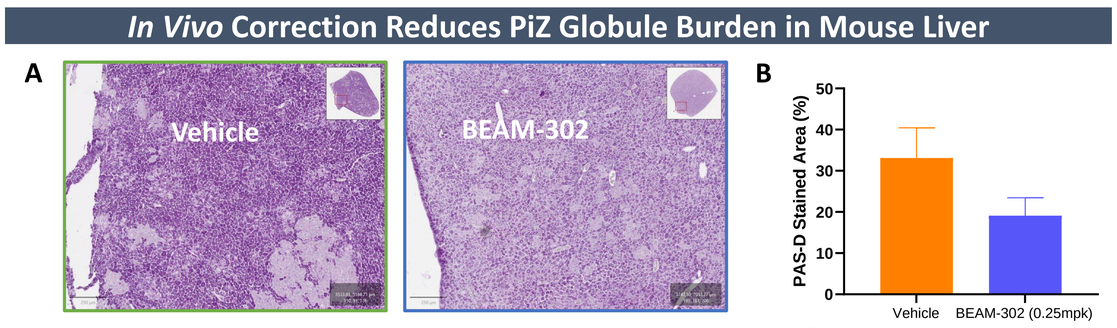


Figure 5 A. Representative Periodic Acid-Schiff-Diastase (PAS-D) stained sections of NSG-PiZ mouse livers dosed with vehicle or 0.25mpk BEAM-302. **B.** Images at 40x magnification were subject to color thresholding (ImageJ) to calculate % PAS-D stained area.

Durable Editing and Increased Functional AAT in NSG-PiZ Mice

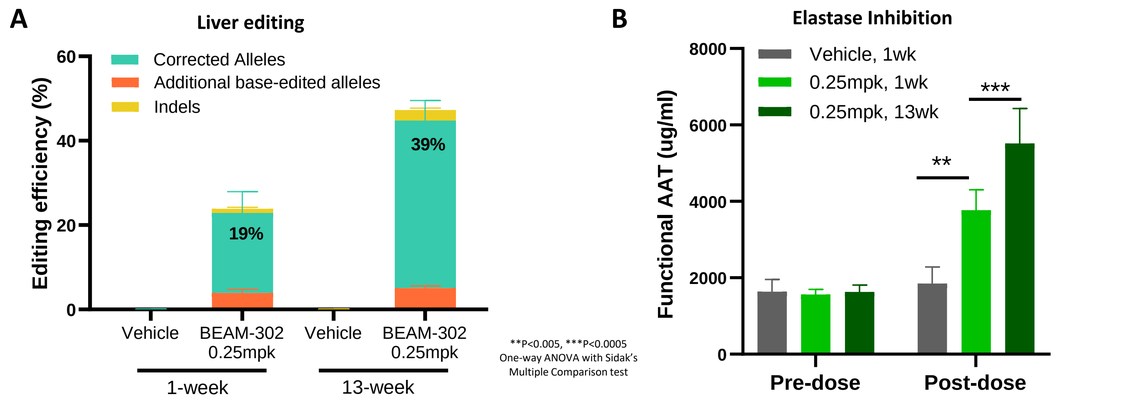


Figure 6 A. Relative frequencies of base-edited alleles in livers of NSG-PiZ mice 1-week or 13-weeks after a single administration of 0.25mpk BEAM-302. **B.** Functional AAT levels in serum of NSG-PiZ mice collected pre-dose, one-week or 13-weeks after a single administration of 0.25mpk BEAM-302.

Repeat Dosing of BEAM-302 Increases Liver Editing and Serum AAT

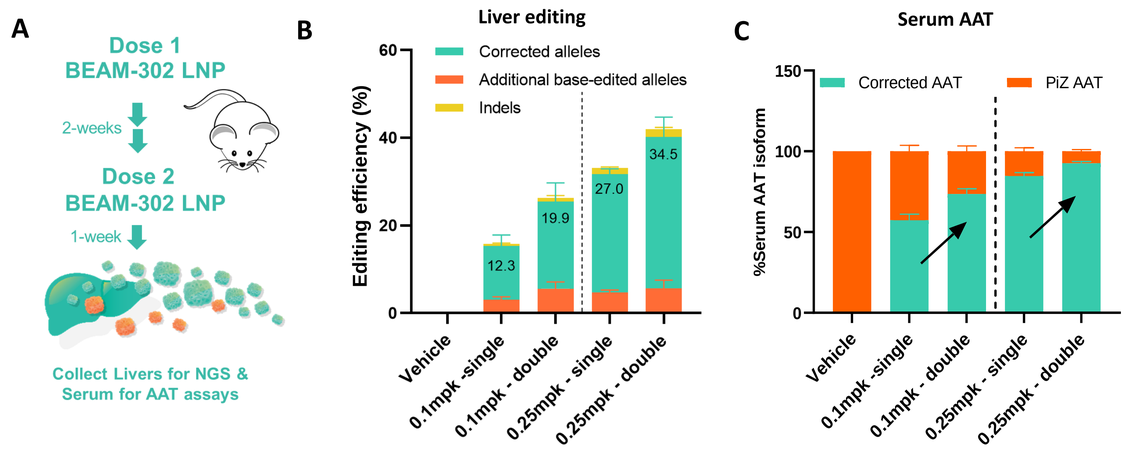


Figure 7 A. Experimental design with BEAM-302 LNP dosed intravenously in adolescent NSG-PiZ mice. **B.** Relative frequencies of base-edited alleles in the liver of NSG-PiZ mice after a single or repeat administration of BEAM-302 at two sub-saturating dose levels. **C.** Percentages of circulating corrected AAT and PiZ AAT proteoforms in NSG-PiZ mice.

Generation of a Novel Knockout Knock-in Humanized PiZ-Rat

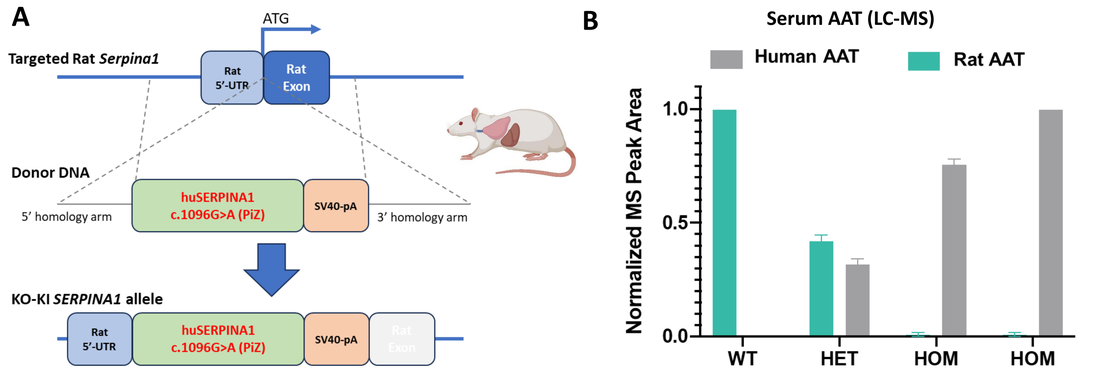


Figure 8 A. Experimental design to generate a PiZ rat model that expresses human PiZ *SERPINA1* cDNA under control of the endogenous rat *Serpina1* promoter. Insertion of the human PiZ *SERPINA* cDNA knocks out expression of rat AAT. **B.** Detecting rat AAT and human PiZ AAT with LC-MS in serum collected from wildtype (WT), heterozygous (HET) or two founder homozygous (HOM) PiZ rats.

In Vivo Correction of the PiZ Mutation with Base Editing in PiZ-Rats

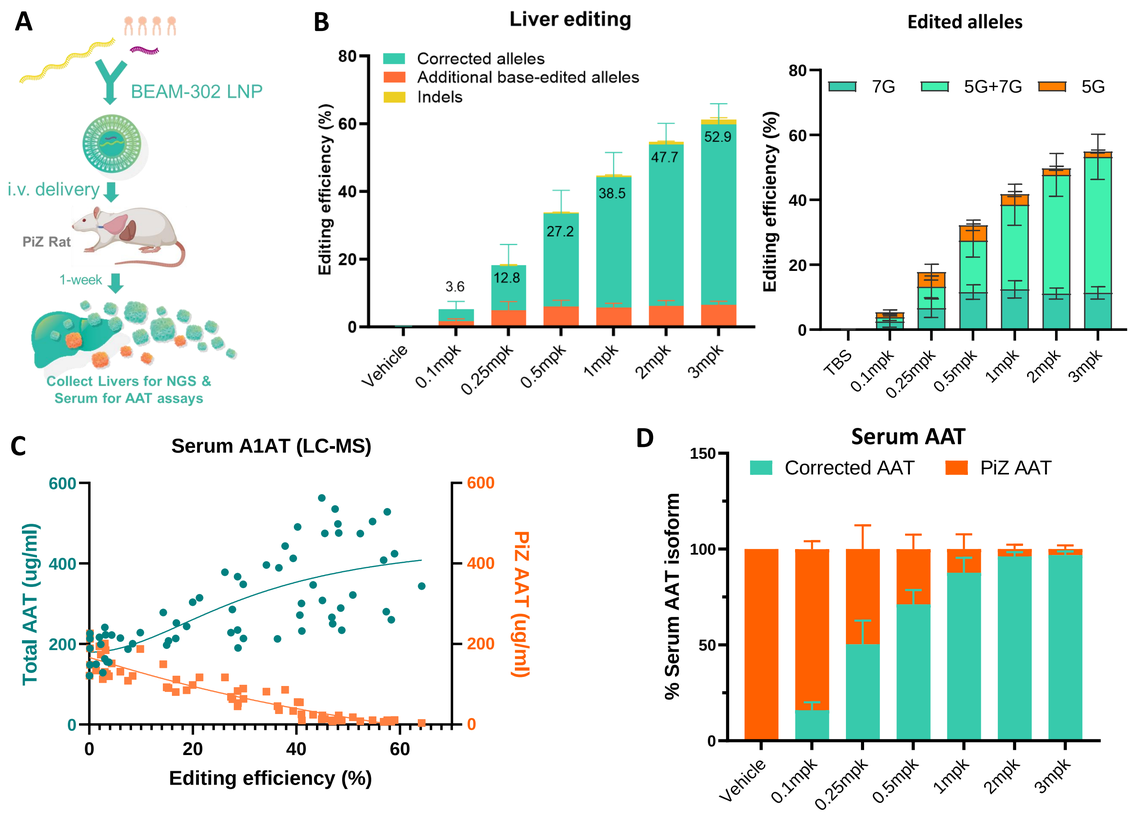


Figure 9 A. Experimental design with editor mRNA and gRNA formulated in BEAM-302 LNP and dosed intravenously in adolescent PiZ rats. **B.** Relative frequencies of base-edited alleles in the liver of PiZ rats after single administration of increasing dose levels of BEAM-302 **C.** Relationship between liver base editing and serum concentrations of total AAT and PiZ AAT as measured by LC-MS **D.** Percentages of circulating corrected AAT and PiZ AAT proteoforms in PiZ rats.

Future Directions

Complete CTA/IND enabling studies, file a CTA/IND application and initiate a first-in-human clinical study in AATD patients in early 2024.

References and Disclosures

1. Stoller JK, Aboussouan LS. Am J Respir Crit Care Med. 2012;185(3):246-259. doi:10.1164/rccm.201108-1428CI
2. Carlson JA, Rogers BB, Sifers RN, Finegold MJ, Clift SM, DeMayo FJ, Bullock DW, Woo SL. Accumulation of PiZ alpha 1-antitrypsin causes liver damage in transgenic mice. J Clin Invest. 1989 Apr;83(4):1183-90. doi: 10.1172/JCI113999. PMID: 2784798; PMCID: PMC303805.

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