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Supplemental Information

Targeted Activation of Cystic Fibrosis

Transmembrane Conductance Regulator

Olga Villamizar, Shafagh A. Waters, Tristan Scott, Sheena Saayman, Nicole Grepo, Ryan Urak, Alicia Davis, Adam Jaffe, and Kevin V. Morris

Targeted activation of Cystic Fibrosis Transmembrane Conductance Regulator

Table S1. Primers, gRNA sequences and BGas-Gapmer.

Name	Sequence (5'-3')
CFTR F	CTCATGGGATGTGATTCTTTCGACC
CFTR R	TTTCTGTCCAGGAGACAGGAGCAT
BGas DRT	TTGATGGTTAAGCAGCTGGTGCC
BGas F	GTGCCAGCACAGAATCCCTCAAT
BGas R	GAATGTTTGCATTTGGTGATCGGG
Beta Actin F	CACCAACTGGGACGACAT
Beta Actin R	ACAGCCTGGATAGCAACG
gRNA 3 sequence (+)	GGGCCGGACCAGGCAGCACT
gRNA 3 sequence (-)	AGTGCTGCCTGGTCCGGCCC
BGas Gapmer	G*T*G*GTATAAAAGATAAT*T*A*T
Scramble	G*A*T*ATATGAATAATGAG*T*A*T

DRT: Directional Reverse Transcriptase primer

Supplemental Figure S1

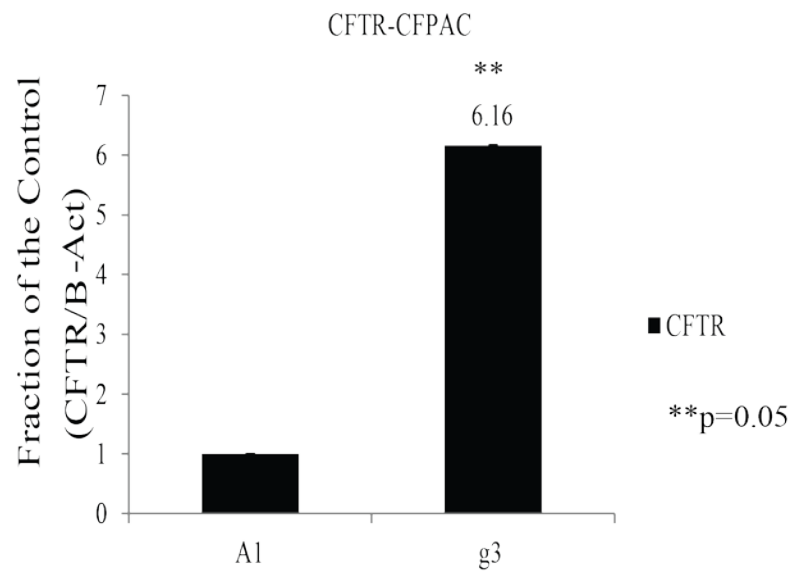
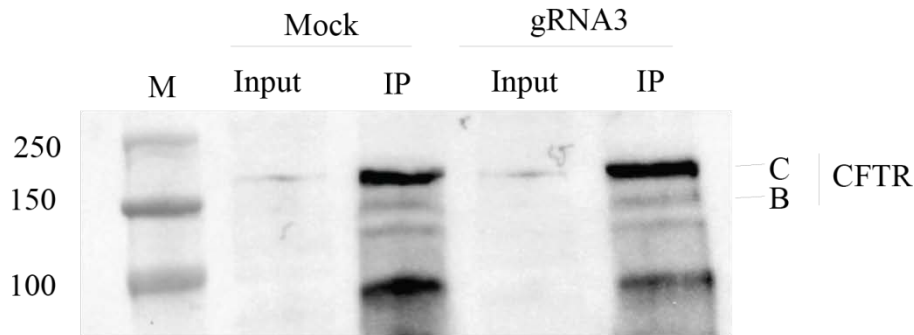


Figure S1. Activation of CFTR expression by dCas9/VPR. CFTR promoter directed guide RNA g3, activates CFTR mRNA expression in CFPAC cells with (F508del-CFTR). Experiments were performed in triplicate in cells shown with the standard error of the means and p values from a paired two-sided T-test, *p 0.01, **p < 0.05

Supplemental Figure S2

A



B

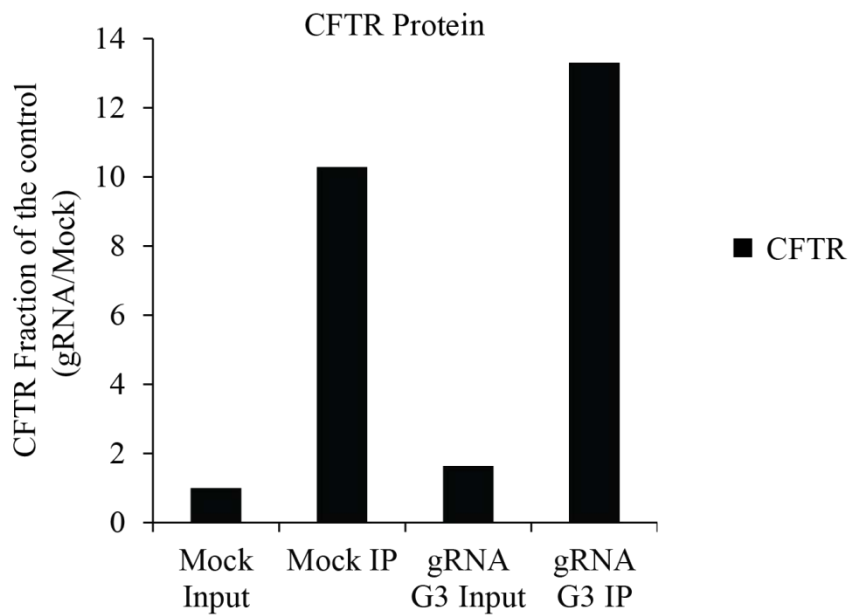


Figure S2. Activation of CFTR expression by dCas9/VPR. (A) Western blot (n=1) showing CFTR protein levels increased of the mature form of CFTR (Band C) and small amounts of the immature form of CFTR (band B) after transiently transfection of A549 (WT-CFTR) cells with g3RNA/dcas9-VPR as determined by (B) optical density analysis. Immunoprecipitation (IP).

Supplemental Figure S3

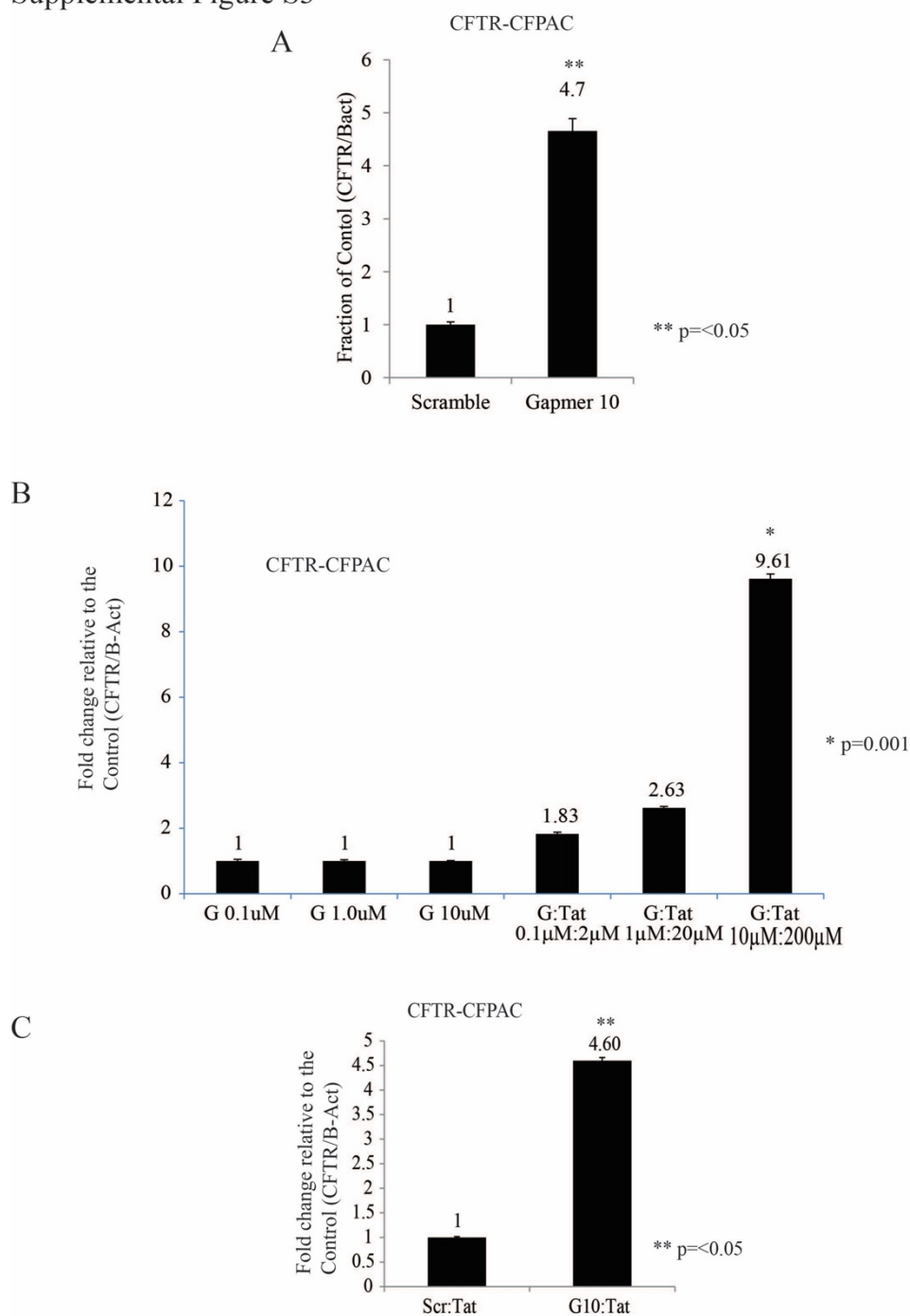
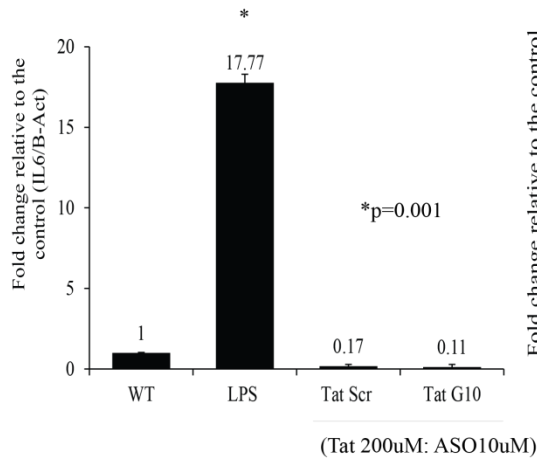


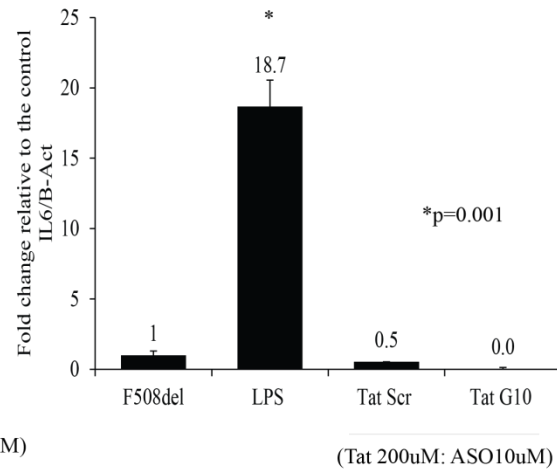
Figure S3. BGas repression results in increase expression of CFTR (A) Effect of Gapmer 10 in CFPAC cells. (B) Dose titration of Gapmer-Tat in CFPAC Cells. (C) CFTR expression after treatment with Gapmer-Tat CPP in CFPAC cells. Experiments were performed in triplicate in cells shown with the standard error of the means and p values from a paired two-sided T-test, *p=0.001, **p < 0.05.

Supplemental Figure S4

A.



B.



C.

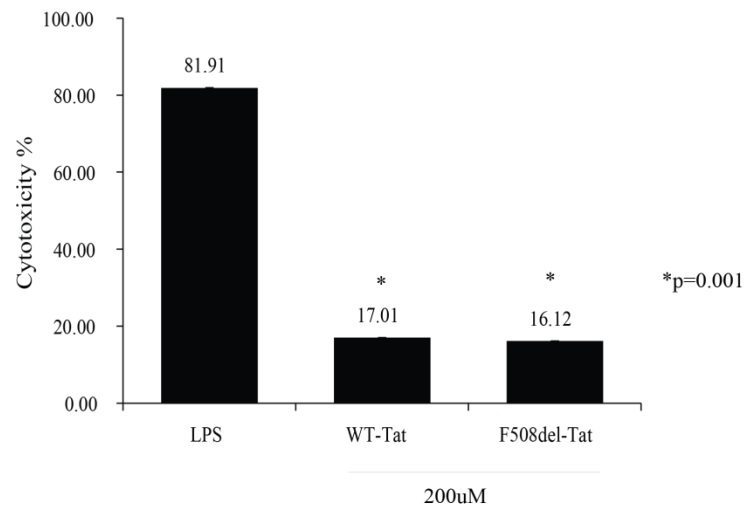
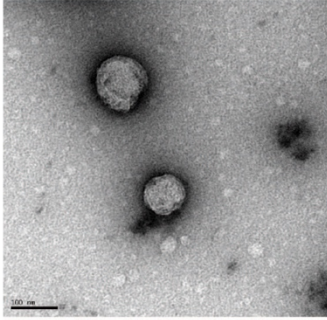


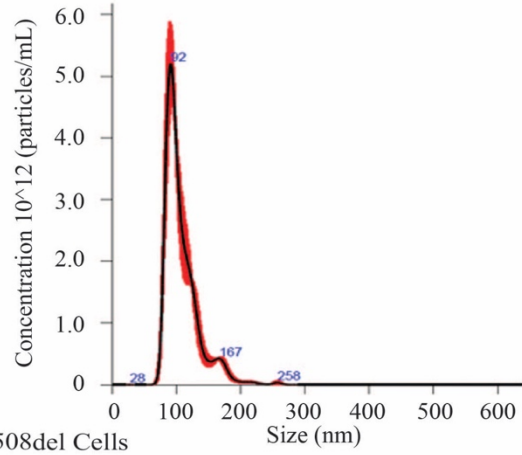
Figure S4. Immunogenicity and cytotoxicity of Gapmer-Tat CPP particles (A) IL-6 expression after treatment with Gapmer- Scramble- Tat. **(B)** Cytotoxicity of Tat-CPP. Lipopolysaccharide (LPS)-treated cells as a positive control. Experiments were performed in triplicate in cells shown with the standard error of the means and p values from a paired two-sided T-test, *p=0.001,

Supplemental Figure S5

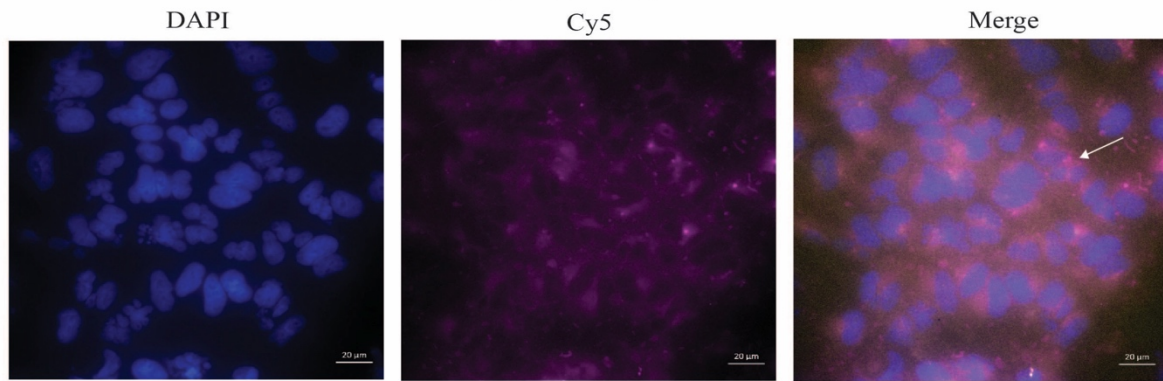
A



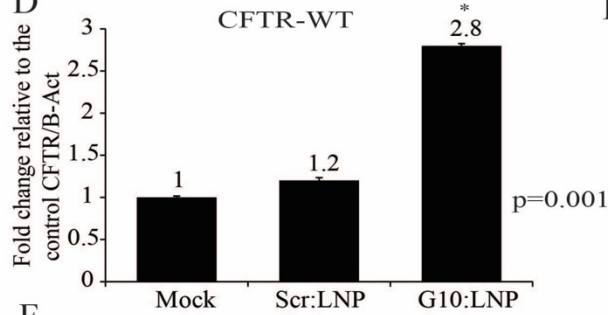
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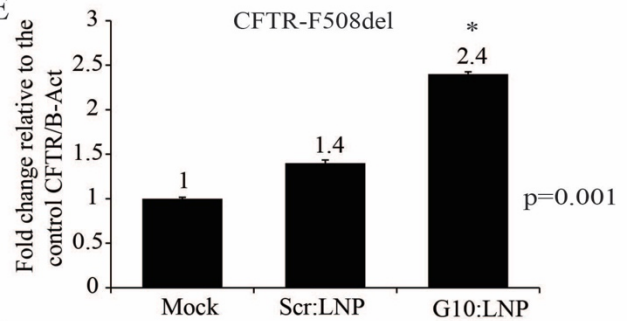
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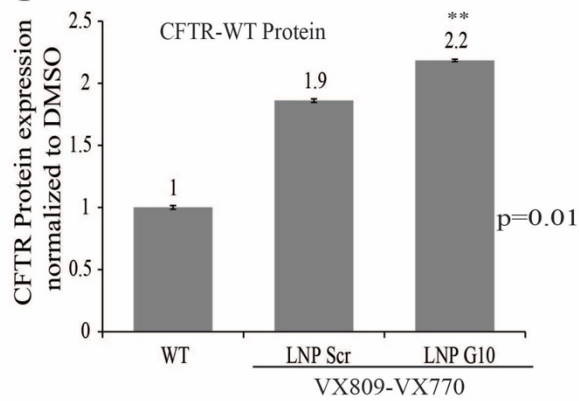
D



E



F



G

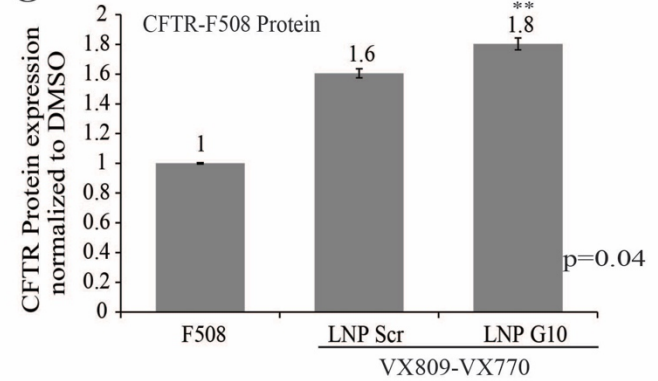


Figure S5. LNP-mediated delivery of Gapmer enters Human Nasal cells and increase expression of CFTR. (A) TEM micrographs of LNP measured by using Nanosight NS 300 system. The histogram represents particle size distribution. (B) Light scope analysis of uptake of Gapmer10-Cy5 LNP in to nasal cells. (C-D) qRT-PCR of CFTR mRNA levels in (C) nasal cells from healthy donor WT and (D) nasal cells from CF patient with F508del mutation. (E-F) CFTR ELISA showing CFTR protein levels expression increased after treated with BGas-Gapmer in (E) nasal cells from healthy donor –WT and (F) nasal cells from CF patient with F508del mutation treated with Cy5-Gapmer loaded LNP. Scale bar: 20 μ m. Experiments were performed in triplicate in cells shown with the standard error of the means and p values from a paired two-sided T-test, *p=0.001, **p < 0.05.