

Supplementary Information

Effective lung-targeted RNAi in mice with peptide-based delivery of nucleic acid

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This Supplementary Information includes:

Figs. S1 to S8

Fig S1. Transfection of Lungs and Liver with NF55 / pmCherry

Fig S2. Biodistribution of pDNA after formulation with PF14 at 24h after i.v. injection

Fig S3. H&E from normal UT mouse and from the animals who were injected with PF14 or NF55 in complex with pDNA

Fig S4. The map of the pshTNF plasmid

Fig S5. Lung transfection with siTNF or pshTNF inhibits LPS-induced inflammation: H&E lung sections

Fig S6. The effect of knocking down TNF in LPS-induced inflammation: the pathology scores

Fig S7. Lung transfection with siTNF or pshTNF inhibits asthma: H&E lung sections

Fig S8. The effect of knocking down TNF in OVA/ALUM-induced asthma: the pathology scores

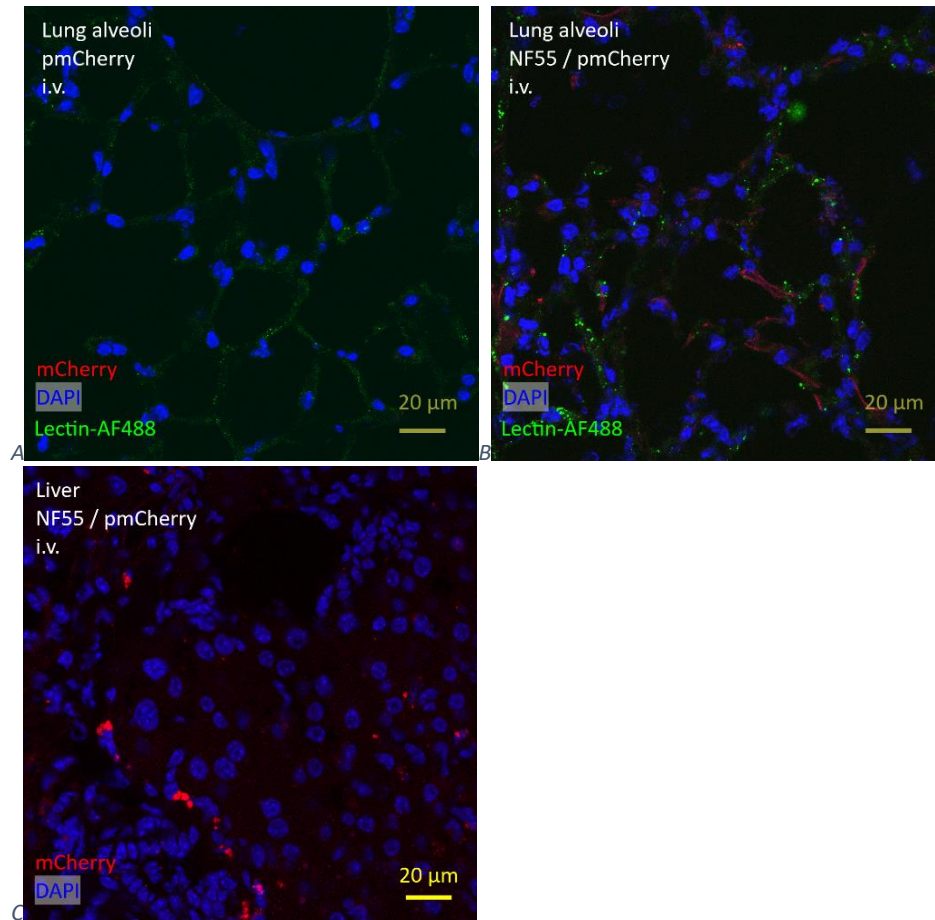


Figure S1. (A) Unvectorized naked pmCherry in lungs. (B) NF55 / pmCherry in Lungs. (C) NF55 / pmCherry in Liver.

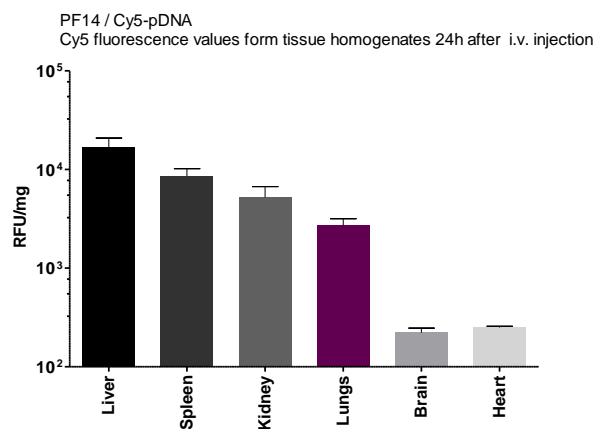
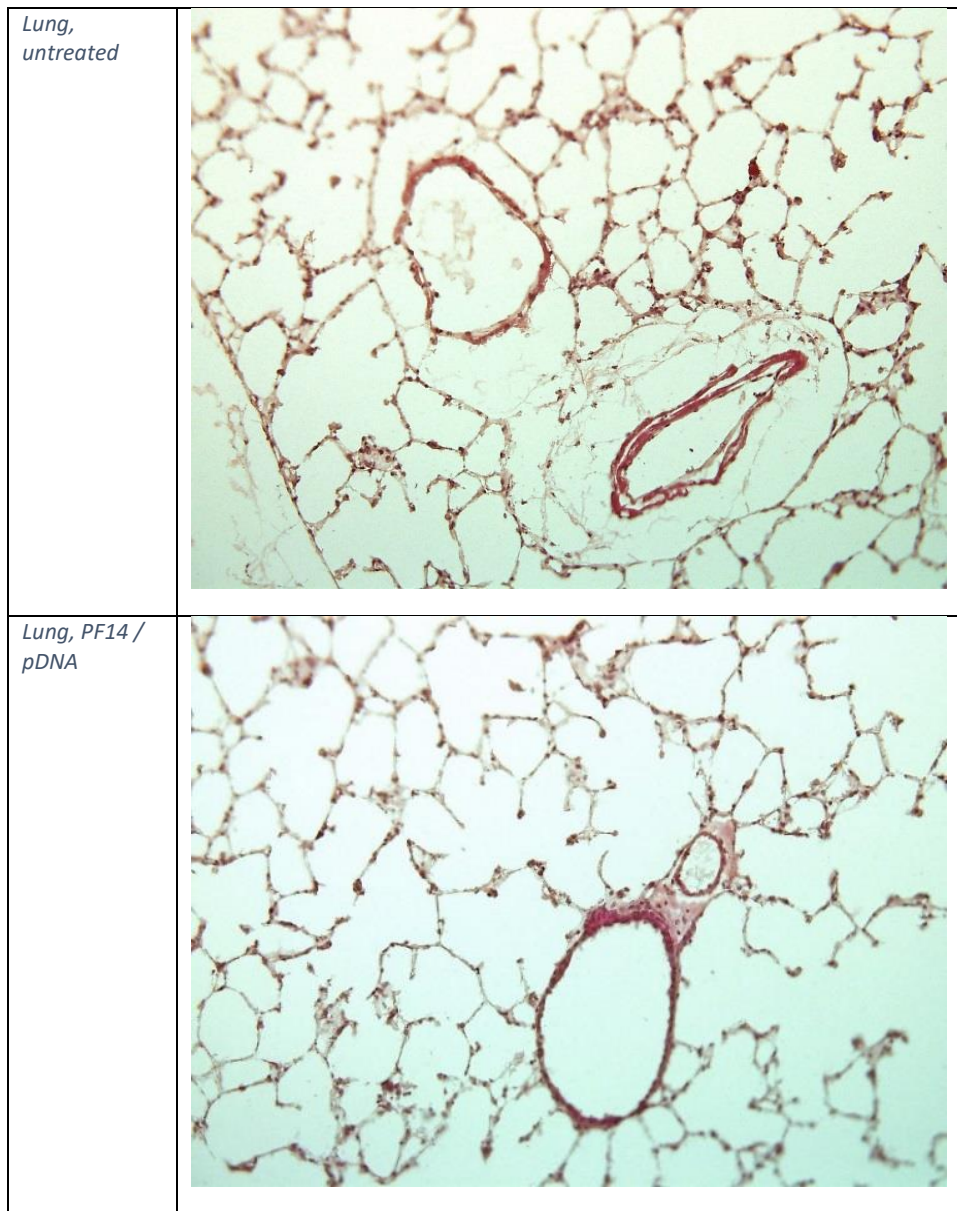


Figure S2. Biodistribution of pDNA after formulation with PF14 at 24h after i.v. injection. The majority of pDNA physically accumulates in liver.



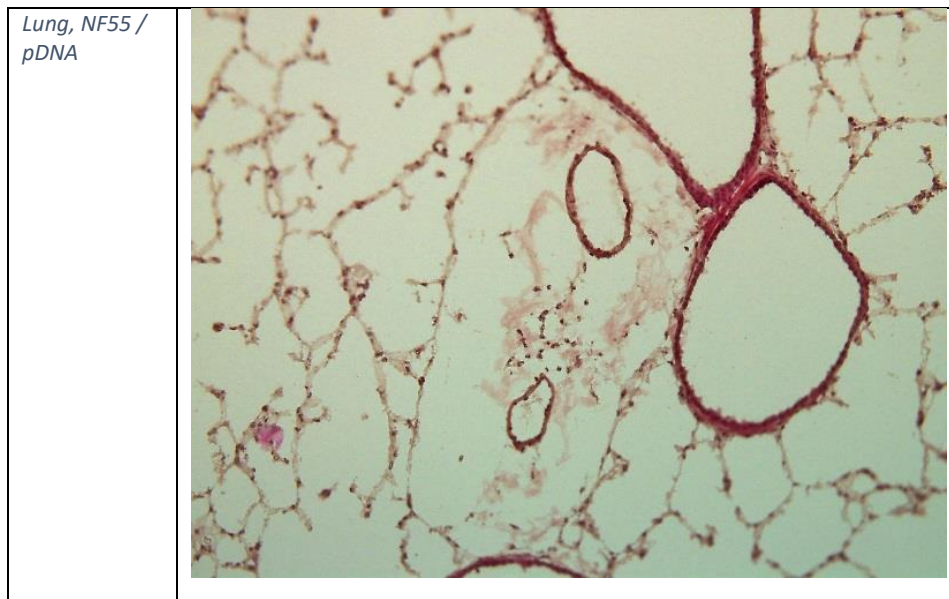


Figure S3. H&E from normal UT mouse and from the animals who were injected with PF14 or NF55 in complex with pDNA.

pshTNF (2981 bp)

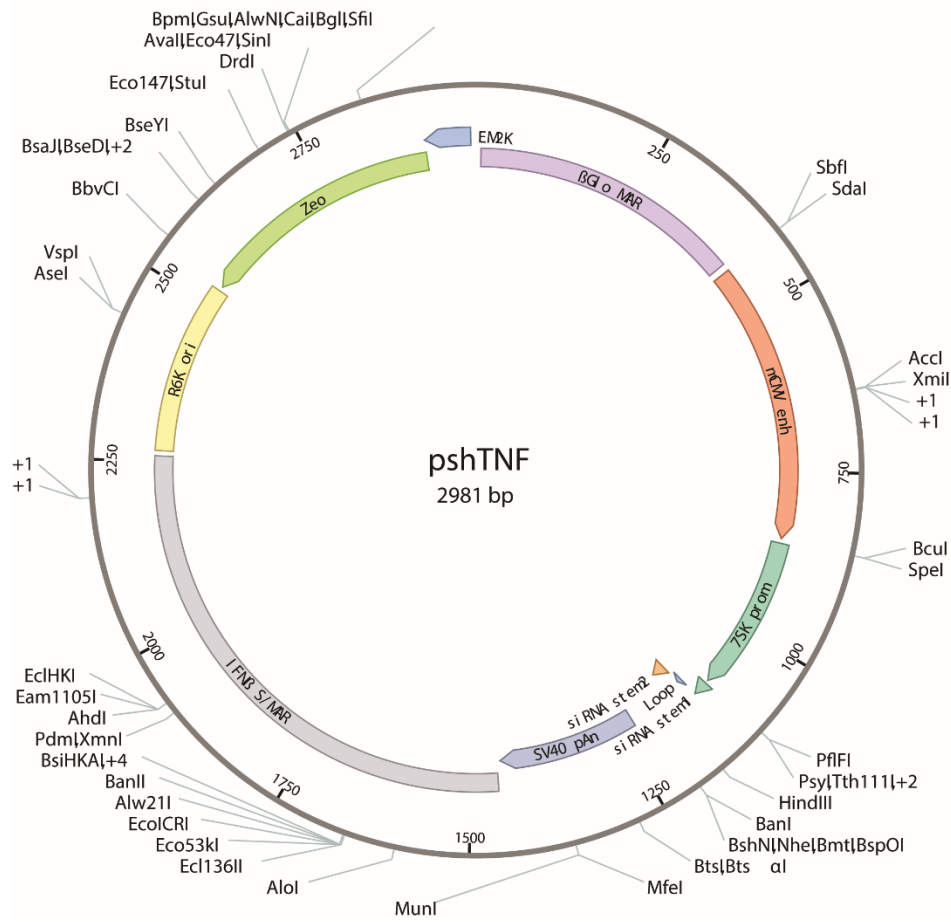
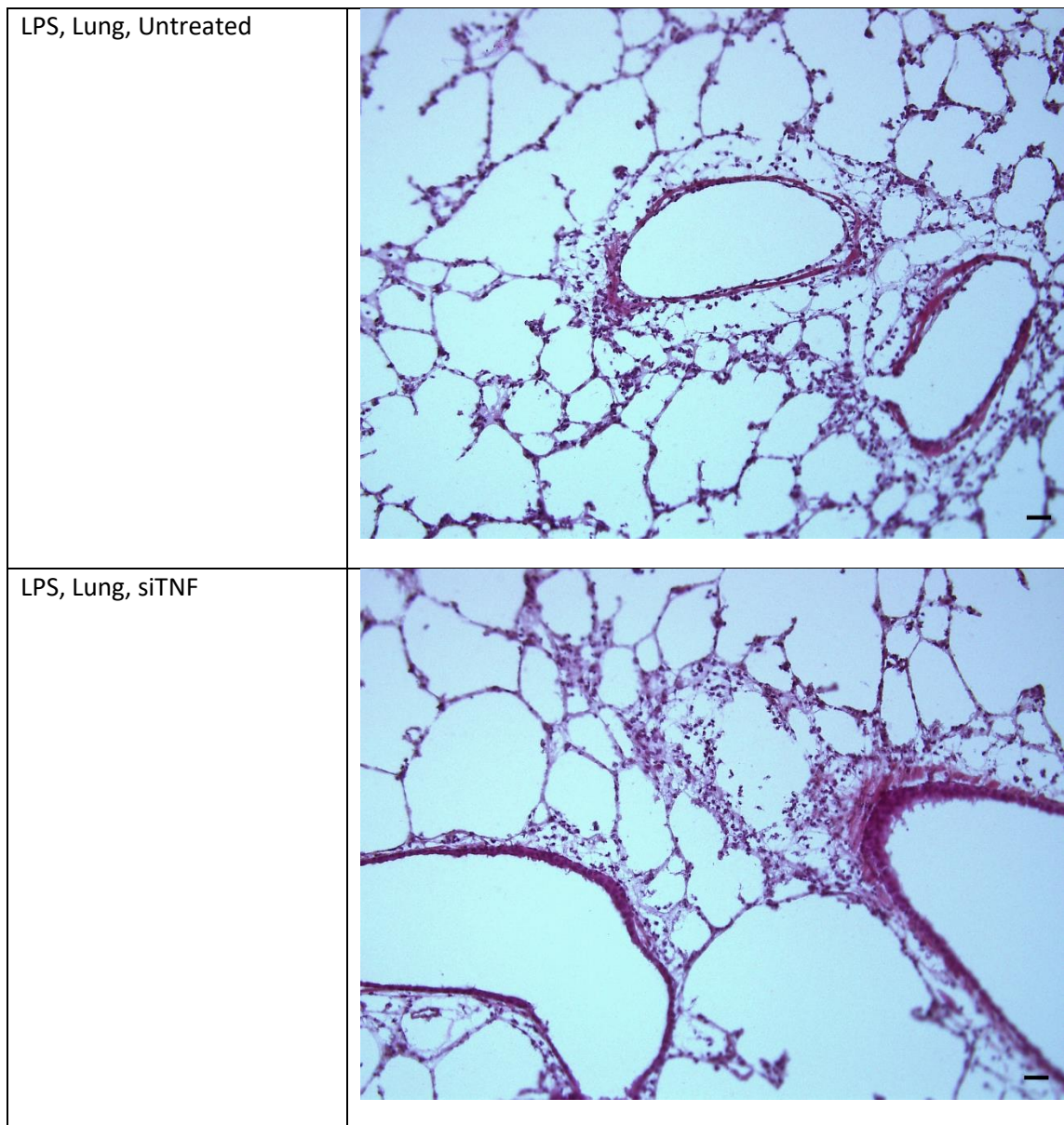
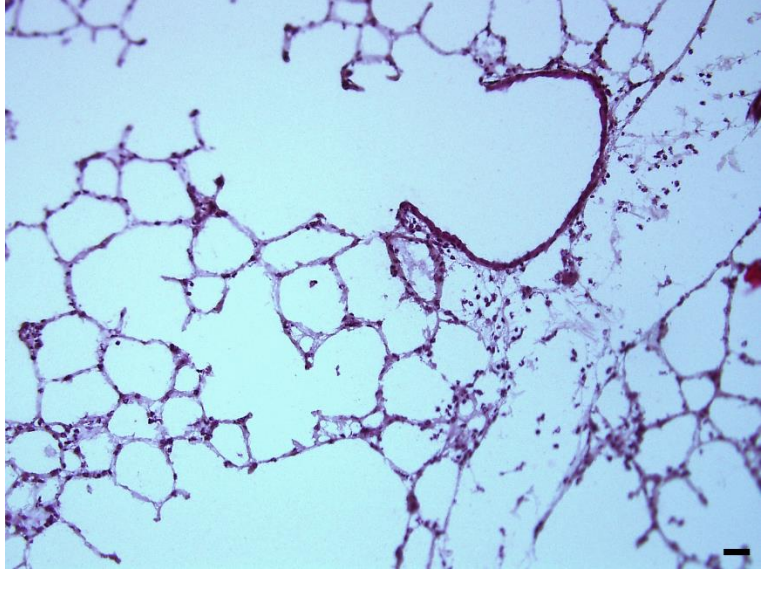
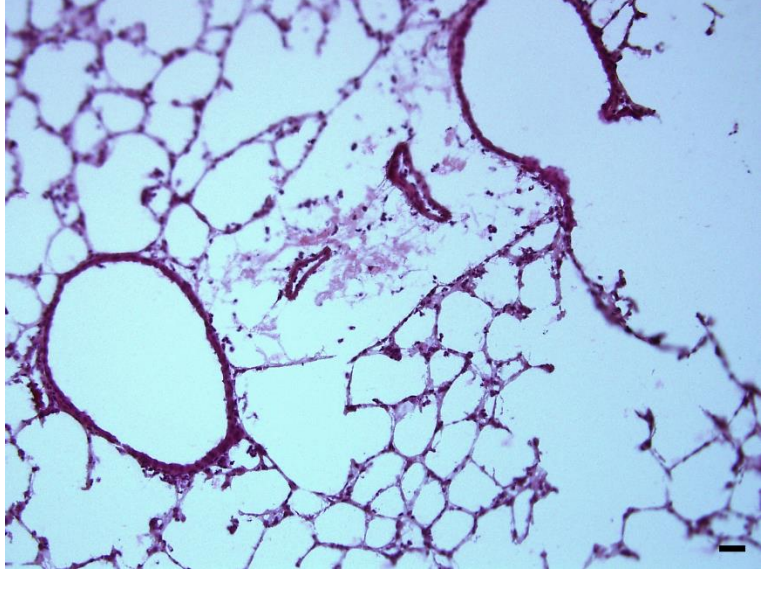


Figure S4. Design of the sh-expressing plasmid: the pshTNF plasmid map.



<p>LPS, Lung, PF14 / siTNF</p>	
<p>LPS, Lung, NF55 / siTNF</p>	

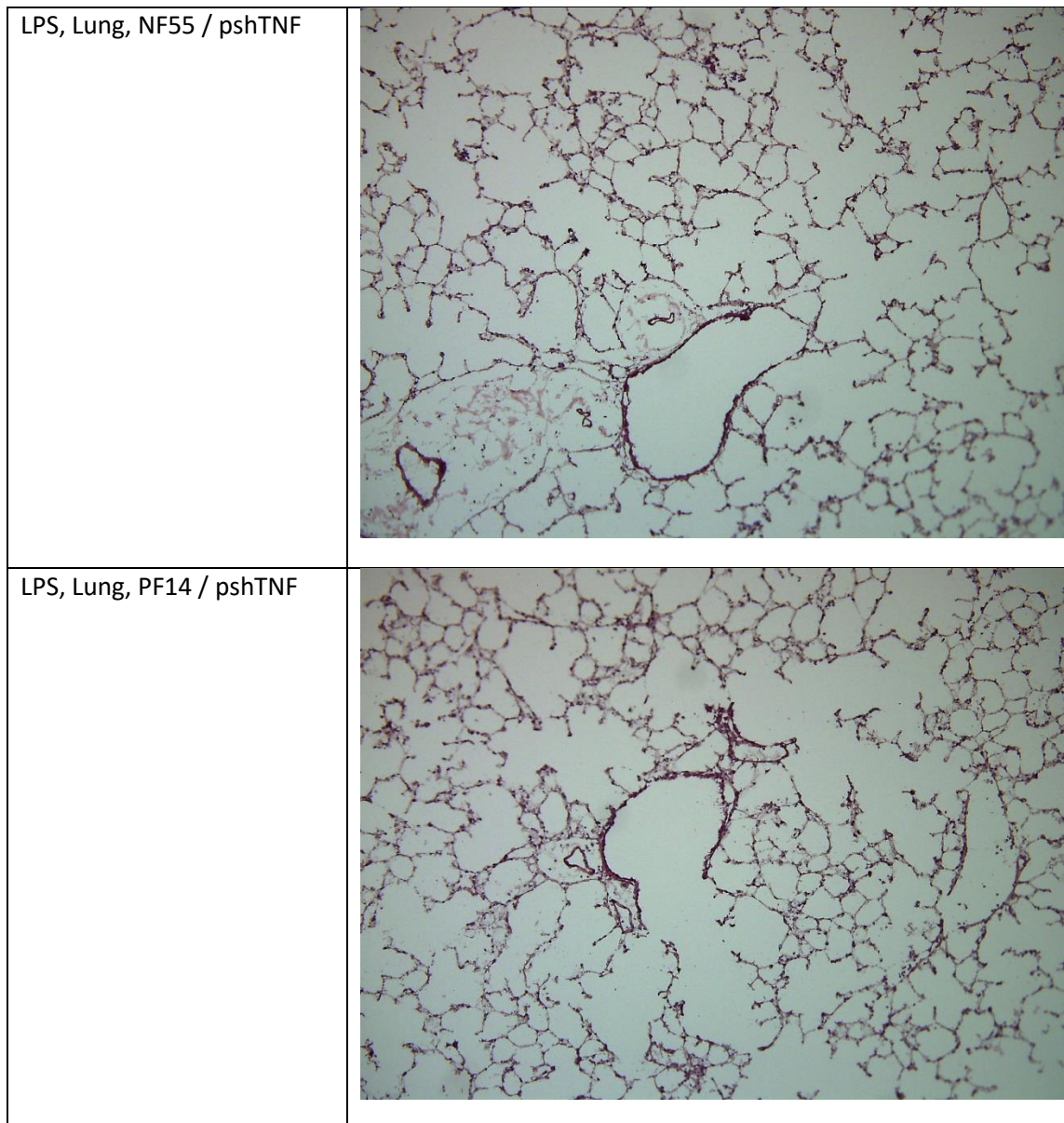


Figure S5. Lung transfection with siTNF or pshTNF inhibits LPS-induced inflammation. H&E stained lung sections of representative individuals. LPS (2.5 mg/kg intranasal) was administered to Balb/C mice at 0h and +24h. siTNF or pshTNF was administered once i.v. at +1h. Lungs were harvested at +28h, cryosectioned, stained with H&E.

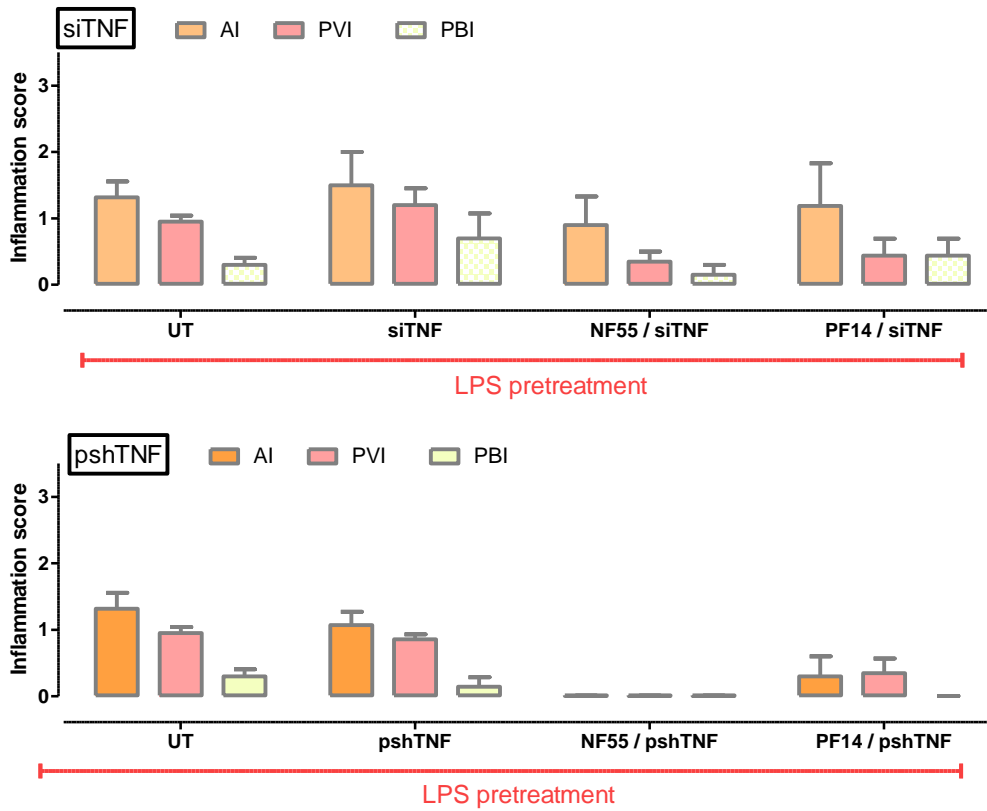
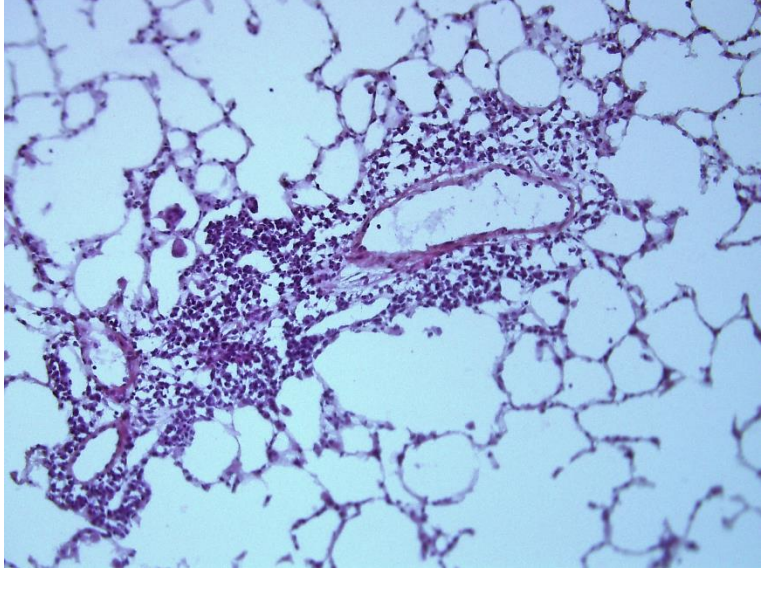
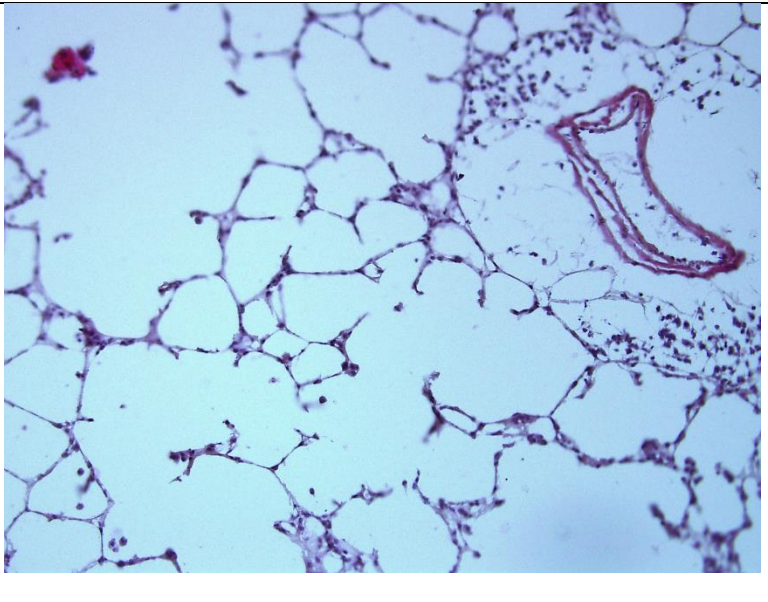
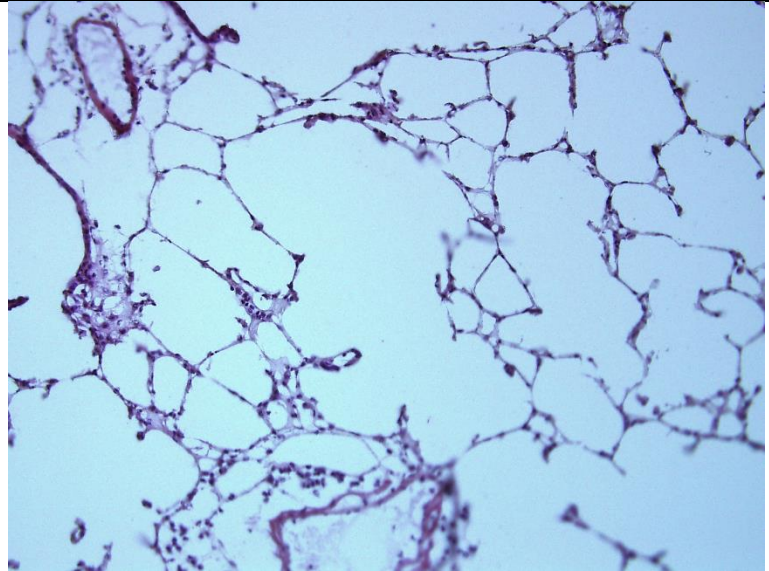


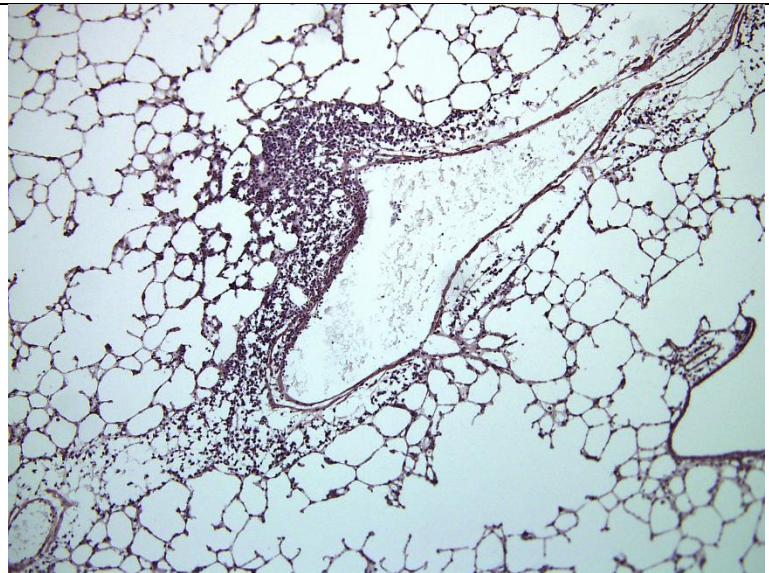
Figure S6. The effect of knocking down $TNF\alpha$ in mice with LPS-induced inflammation by either siTNF or pshTNF. Inflammation was scored by a pathologist: PVI = perivascular infiltration, PBI = peribronchial infiltration, AI = alveolar infiltration.

<p>OVA/ALUM, Lung, pshTNF</p>	 A histological micrograph of lung tissue stained with hematoxylin and eosin (H&E). The image shows a cross-section of lung parenchyma with alveolar spaces. There is a prominent, dense cluster of inflammatory cells, likely macrophages and lymphocytes, infiltrating the interstitial space and partially obscuring the alveolar architecture. The nuclei of these cells are stained dark purple, while the surrounding tissue and alveolar walls are stained pink.
<p>OVA/ALUM, Lung, PF14 / pshTNF</p>	 A histological micrograph of lung tissue stained with hematoxylin and eosin (H&E). The image shows a cross-section of lung parenchyma with alveolar spaces. Compared to the top panel, there is a significant reduction in the density of inflammatory cell infiltration. The alveolar structure is more clearly visible, and the overall appearance is less inflamed. A small, distinct structure, possibly a blood vessel or a small airway, is visible on the right side of the image.

OVA/ALUM, Lung, NF55 / pshTNF



OVA/ALUM, Lung, siTNF



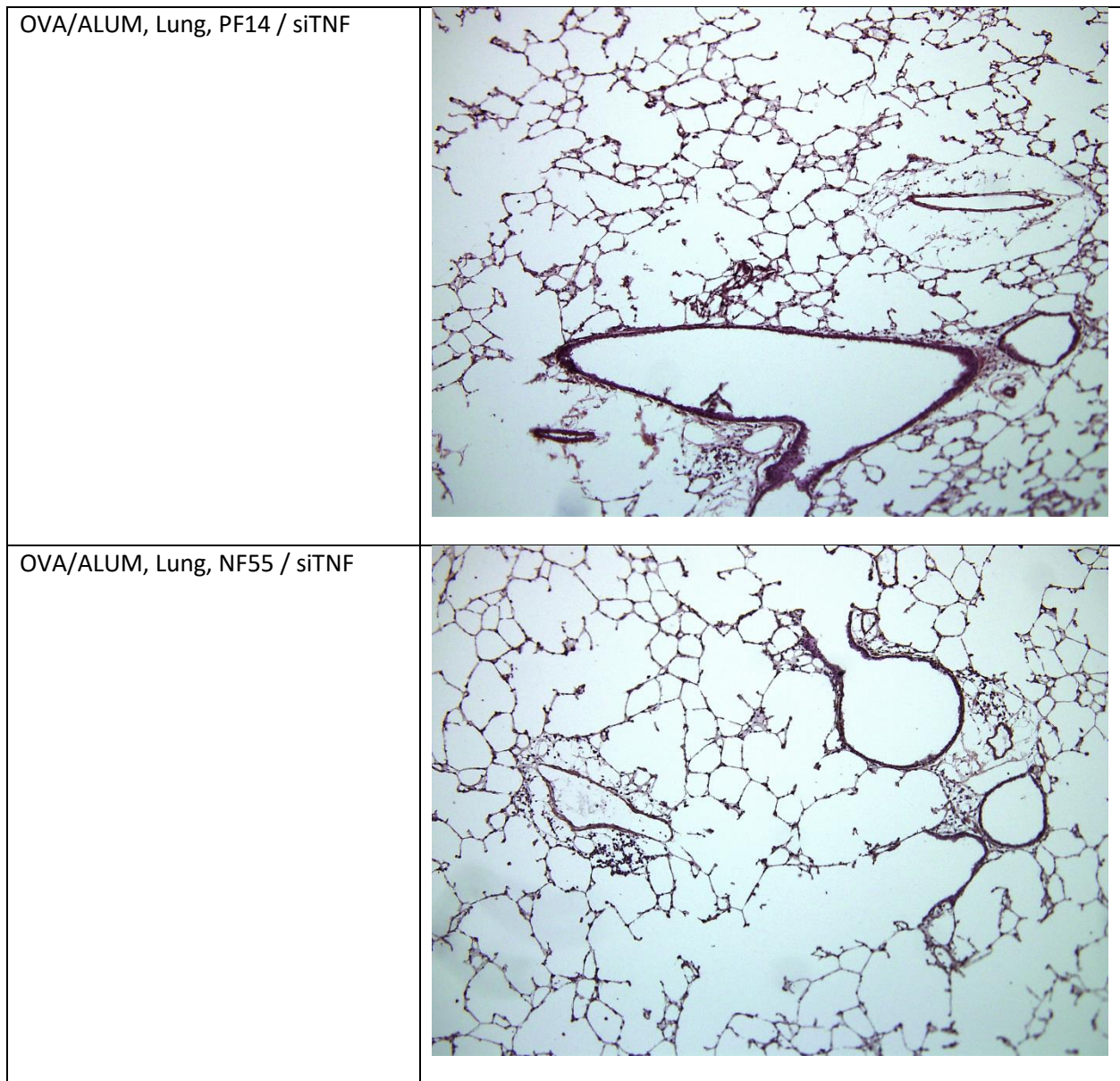


Figure S7. Lung transfection with siTNF or pshTNF inhibits OVA/ALUM-induced inflammation in a model of asthma in mice. H&E stained lung sections of representative individuals.

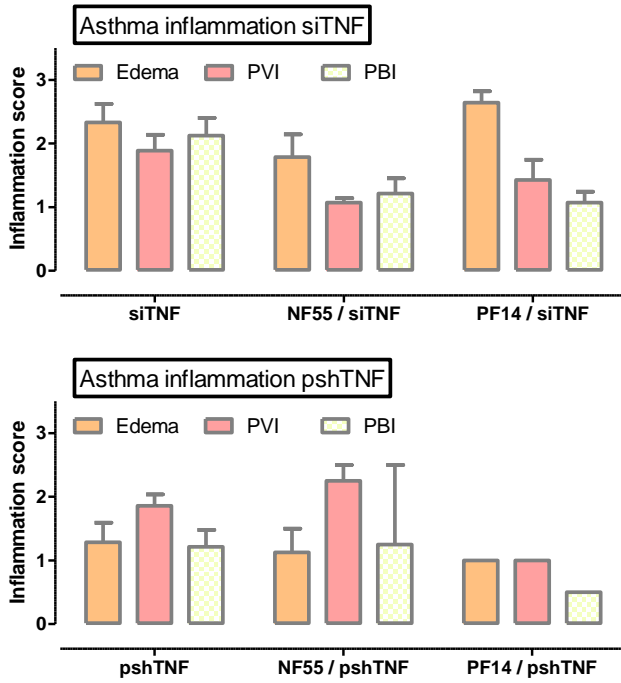


Figure S8. The effect of knocking down $TNF\alpha$ in mice with OVA/ALUM-induced asthma by either siTNF or pshTNF. Inflammation was scored by a pathologist: PVI = perivascular infiltration, PBI = peribronchial infiltration, AI = alveolar infiltration.