Supplemental Information

Exosome-Mediated Small RNA Delivery:

A Novel Therapeutic Approach for Inflammatory

Lung Responses

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Supplemental Figures

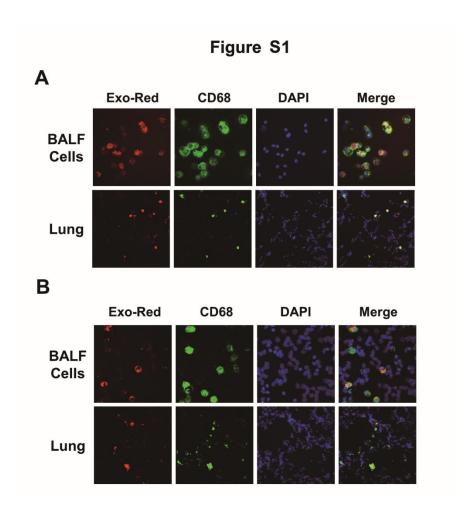


Figure S1. Distribution of Exo-Red labeled serum EXOs after intratracheal instillation.

(A and B) Purified serum EXOs (100 μ g in 50 μ L PBS) labeled with Exo-Red were administrated to non-pretreated mice (A) or LPS pretreated mice (B) intratracheally (n = 4 for each group). Mice were sacrificed 24 hours after EXOs administration. Immunofluorescent staining of macrophages was performed in BALF cells and lungs sections using an antibody against CD68. The nuclei were stained with DAPI.

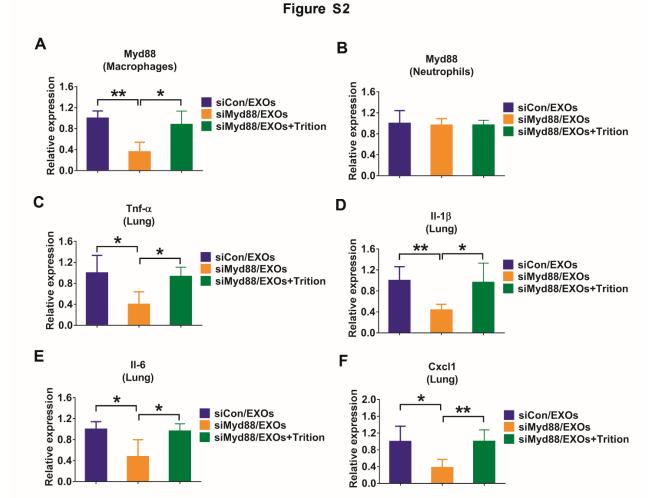


Figure S2. The integrity of serum EXOs are crucial for siRNA/miRNA delivery. (A-F) Mice were pretreated with 1 μ g LPS intratracheally. After 3 hours, 100 μ g intact serum EXOs transfected with 100 pmol siRNA control (siCon/EXOs), Myd88 siRNA (siMyd88/EXOs) or 100 μ g 0.075% Triton X-100 pre-lysed EXOs transfected with 100 pmol Myd88 siRNA (siMyd88/EXOs+Triton) were given to each mouse (n=5 for each group). 24 hours later, the level of Myd88 was detected in sorted macrophages (A) and neutrophils (B) from BALF cells. Relative mRNA levels of TNF- α (C), Il-1 β (D), Il-6 (E) and Cxcl1 (F) in the lung were measured. Results represent means \pm SD. *P < 0.05, **P < 0.01.

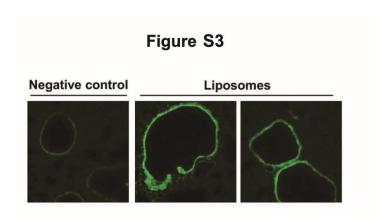


Figure S3. Distribution of liposomes after intratracheal instillation.

Liposomes were generated using Lipofectamine 2000 reagent and labeled with fluorescent dye. Immunofluorescent pictures showing the distribution of liposomes in murine lungs were taken using fluorescence microscopy 24 hours after intratracheal instillation.