



**FIGURE S1.** (A) Densitometric quantification of the immunoblotting gel data presented in Fig. 1C (in text) using Quantity one software. (B) MH-S cells were transfected with Ctrl siRNA or Atg7 siRNA respectively, combined with LC3-RFP plasmid for 24 h and then infected with PAO1-GFP (MOI=10:1) for 2 h. LC3 congregations were found in Ctrl siRNA transfected cells upon Pa infection. Data are representative from three independent experiments. Scale bar=5  $\mu$ m. (C) Bacteria internalization assay in MH-S cells 30 min after infection with pretreatment of DPI or NOC-18 either with Ctrl siRNA or Atg7 siRNA transfection, repectively. Average values and SDEVs were calculated from triplicate samples.



NOS2

Α

Atg7



+

Ctrl SiRNA SZ SiRNA

Т

—

+

+

+

С

NOS2

 $\beta$ -actin

200

150·

100

50-

+

Ра

Merge



FIGURE S2. (A) MH-S cells were transfected with Ctrl siRNA or Atg7 siRNA at 50 nM for 24 h, respectively. The cells were infected with PAO1 for 2 h (MOI=10). Atg7 and NOS2 were detected by immunostaining. (B-D) MH-S cells were transfected with Ctrl siRNA or NOS2 siRNA, respectively. The cells were infected as above. (B) The expression of NOS2 was detected by real-time qPCR. (C) NOS2 knock-down was determined by immunoblotting. (D) ELISA was used to measure IFN- $\gamma$  in MH-S cell supernatant. (E) MH-S cells were transfected as above, and pretreated with DPI (5  $\mu$ M) or NOC-18 (100  $\mu$ M) for 30 min, then infected with PAO1. ELISA was used to measure IFN- $\gamma$  in cell supernatant. (F) Cells were transfected as above, and pretreated with STAT1 inhibitor Fludara (50  $\mu$ M, 2 h). pSTAT1 $\alpha$  was immunostained for nuclear translocation detection. The data are representative of three independent experiments. Scale bar=10 µm. (Average values and SDEVs were calculated from triplicate samples; \*, p < 0.05 by one-way ANOVA with Turkey's post-hoc).



**FIGURE S3.** MH-S cells were transfected with Ctrl, Atg5 or Beclin1 siRNA at 50 nM, respectively. Protein lysates prepared 24 hours post-transfection were analyzed for Atg5 (A) and Beclin1(B) expression by immunoblotting. The data are representative of three independent experiments. (C) MH-S cells were pretreated with ABH (150  $\mu$ M), DPI (5  $\mu$ M), APO (100  $\mu$ M) and NAC (100  $\mu$ M) for 30 min, respectively. EuTc assay was used to measure  $H_2O_2$  after Pa infection (MOI=10, 2 h). (D) MH-S cells were transfected with Ctrl siRNA or SHP2 siRNA, respectively. Cells were pretreated with  $H_2O_2$  (10 mM, 30 min). The cell were then infected with Pa as above. The expression of NOS2 was detected by realtime qPCR. (E) WT mice and  $atg7^{-/-}$  mice were infected with  $1 \times 10^7$  CFU of PAO1. After 24 h, BAL fluids were collected. The pro-inflammatory cytokines were determined by ELISA, respectively. Data are showed as means  $\pm$  SD of three independent experiments (\*, p < 0.05; \*\*, p < 0.01). One-Way ANOVA with Turkey's post-hoc. RFU, relative fluorescence units.



**FIGURE S4.** (A) WT mice and  $atg7^{-/-}$  mice were infected with  $1 \times 10^7$  CFU of PAO1. After 24 h, organs were homogenized in PBS and used for assessing bacterial colonies. (B) MPO activity in lung, liver, spleen, and kidney of the mice. (C) Levels of lipid peroxidation were observed in the lung, liver, kidney and spleen tissues as assessed by the thiobarbituric acid-reactive substance assay. (D) WT mice and  $atg7^{-/-}$  mice were treated with AG (100 mg/kg body weight), NOC-18 (10 mg/kg body weight), or PBS 1 h before pulmonary Pa infection. Lungs were homogenized for RNA isolation 24 h post infection. NOS2 mRNA was detected by qPCR. (E) NOS2 mRNA expression in different organs were determined after Pa infection. The data are shown as means ± SD from three mice (one-way ANOVA (Tukey's post hoc); \*, p<0.05; \*\*, p<0.01). RLU, relative luciferase units.