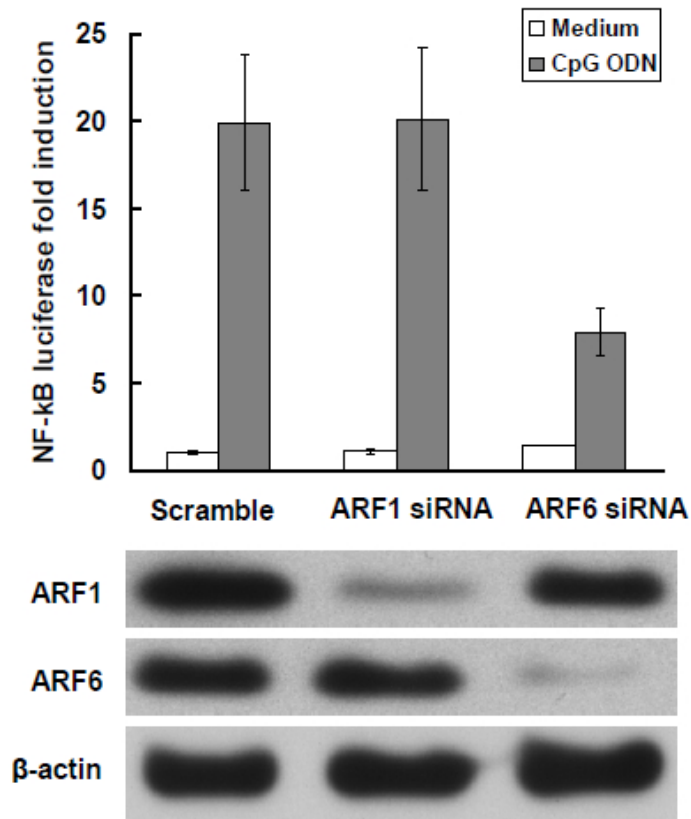
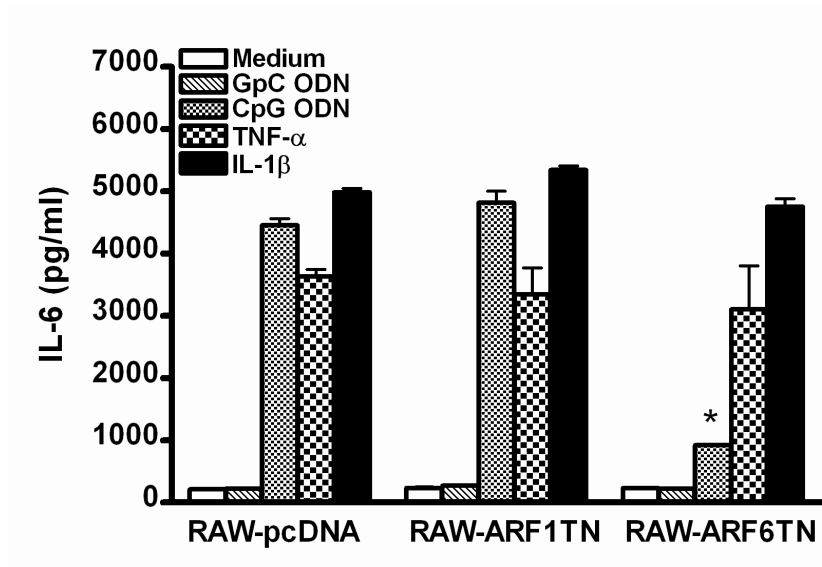


## Supplemental Figure 1



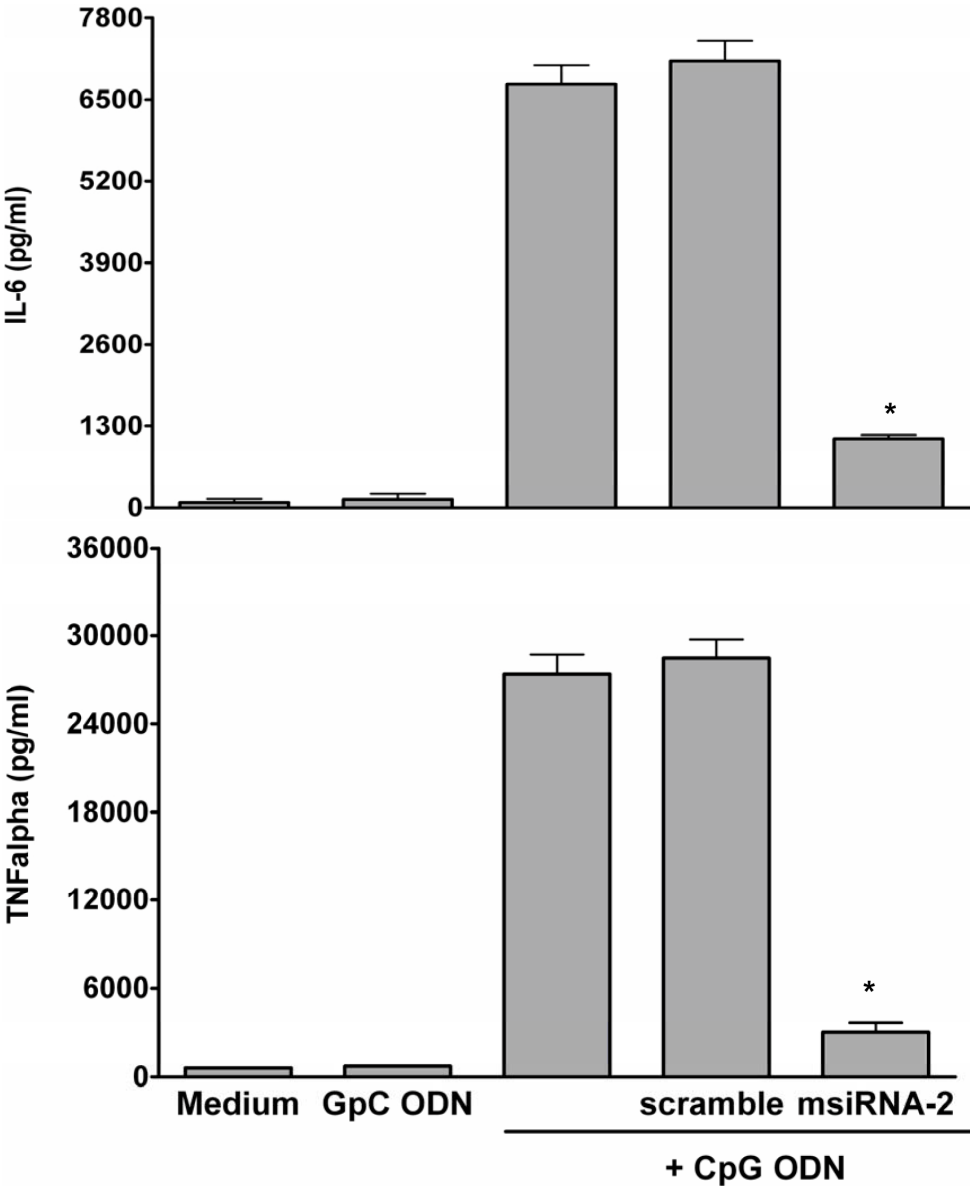
**Figure S1. ARF6 siRNA impair NF- $\kappa$ B activation induced by CpG ODN.** HEK293T transfectants, stably transfected with mouse TLR9 (293T-mTLR9), were transiently transfected with scramble siRNA, human ARF1 siRNA or human ARF6 siRNA plus p5 $\times$ NF- $\kappa$ B-luciferase 48 h, followed by CpG ODN (1.0  $\mu$ M) stimulation for 8 h. NF- $\kappa$ B luciferase activities were then measured. In addition, the protein level of ARF6 was determined by western blotting with anti-ARF6.

## Supplemental Figure 2



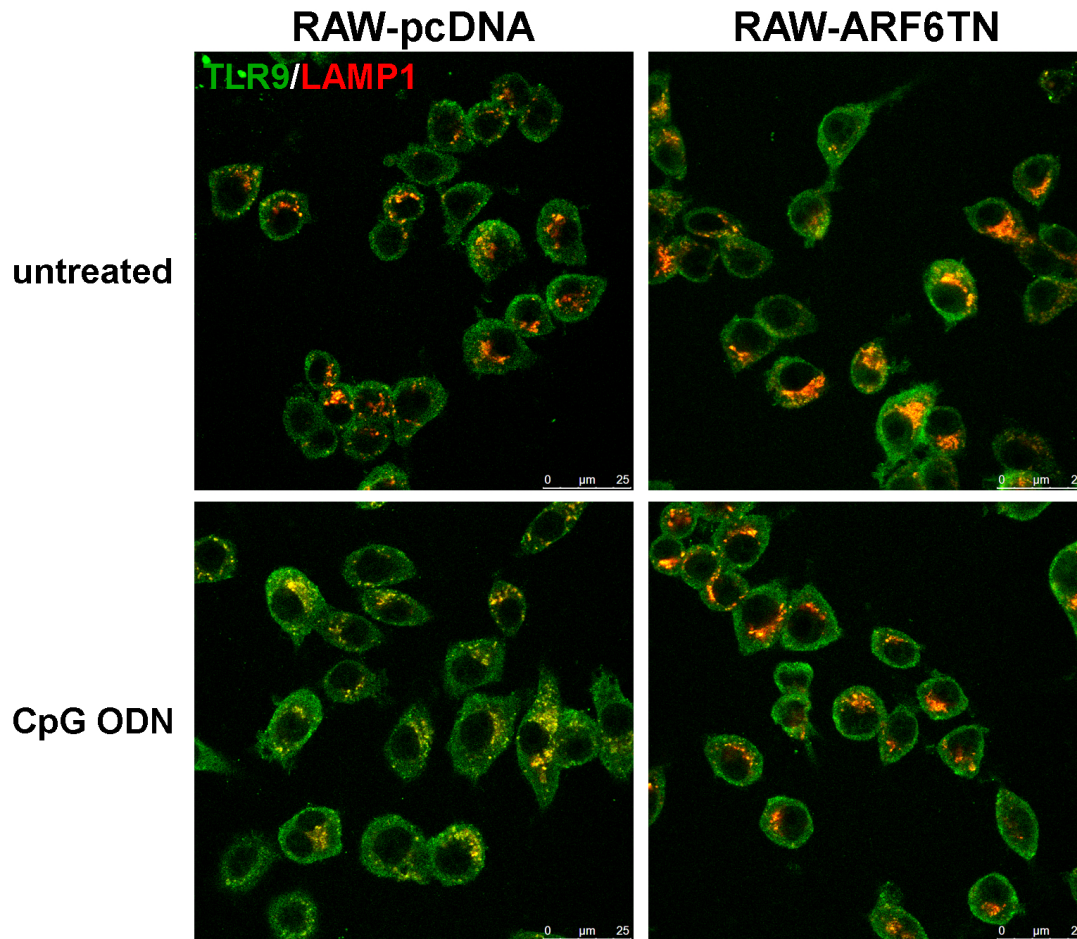
**Figure S2. ARF6 is indispensable for IL-1 $\beta$ - or TNF- $\alpha$ -induced IL-6 production.** RAW-pcDNA, RAW-ARF1T31N and RAW-ARF6T27N cells were stimulated with medium alone, GpC ODN, CpG ODN, IL-1 $\beta$  (20 ng/ml) or TNF- $\alpha$  (10 ng/ml) for 20 h. IL-6 levels in the culture supernatants were measured by ELISA. Data represent the mean  $\pm$  SD of 3 experiments.

### Supplemental Figure 3



**Figure S3. ARF6 siRNA impaire production of IL-6 and TNF- $\alpha$  induced by CpG ODN.** Scramble siRNA or ARF6 siRNA-2 (msiRNA-2) transfected spleen monocytes/macrophages were incubated in medium or medium supplemented with 1.0  $\mu$ M GpC ODN or CpG ODN for 20 h as indicated. IL-6 and TNF- $\alpha$  levels in the culture supernatants were measured by ELISA. Data represent the mean  $\pm$  SD of 3 experiments. \* $P < 0.001$ .

## Supplemental Figure 4



**Figure S4. Stable expression of ARF6T27N interferes with TLR9 trafficking.** RAW-pcDNA or RAW-ARF6T27N cells were treated with or without CpG ODN (1.0  $\mu$ M) for 2 h. Cells were then fixed, permeabilized and labeled with anti-TLR9 (green, FITC) and anti-LAMP1 (red, Dylight 549) antibodies. The histograms depict cross-line scans of the fluorescence intensities of the merged panel. These images are representative of 3 independent experiments. Scale bar, 25  $\mu$ m.