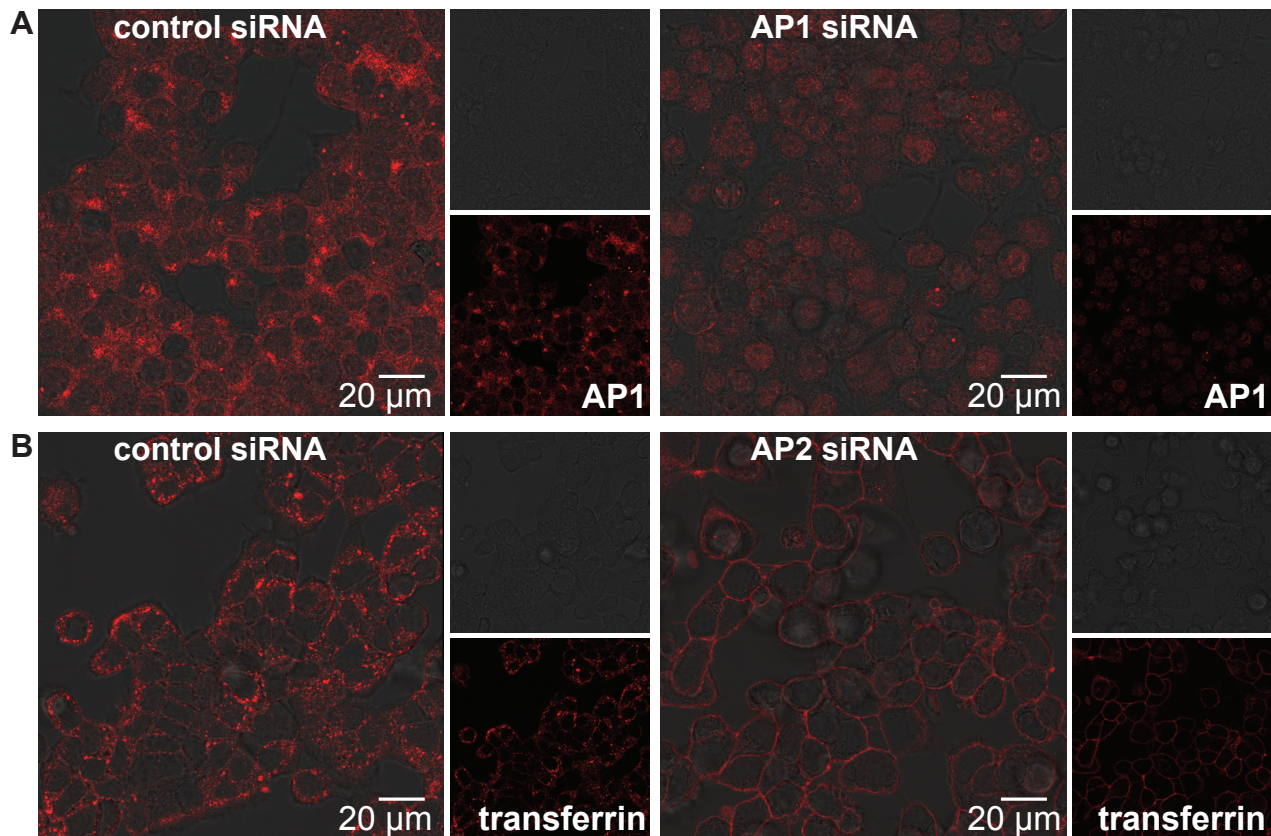


**Supplemental Fig 1.** Expression levels of UNC93B1-mCitrine variants in different stable cell lines and RNA responsiveness of HEK cells. **(A)** HEK cell lines shown in Fig. 1C were analyzed for UNC93B1-mCitrine expression by Western blotting. **(B)** EBV-immortalized human B cells and **(C)** human UNC93B1 KO THP-1 monocytes shown in Fig. 1D or E, respectively, were analyzed for UNC93B1-mCitrine expression levels via flow cytometry. **(D)** HEK cells stably expressing TLR7 or 8 and UNC93B1-mCitrine WT or AxxA were stimulated with 20  $\mu$ M TLR7- or 8-specific single-stranded RNA and analyzed for IL-8 secretion. Data are representative of two independent experiments.



**Supplemental Fig 2.** Validation of AP1 and AP2 knockdown. **(A)** Control siRNA and AP1 siRNA treated cells from Fig. 4 were fixed with 4% PFA, permeabilized with 0.5% Saponin, stained for AP1 (BD Transduction Laboratories, 611329) and analyzed by confocal microscopy. **(B)** Control siRNA and AP2 siRNA treated cells from Fig. 4 were incubated for 10 min with transferrin-Alexa647 (1 μg/ml), fixed with 4% PFA and analyzed by confocal microscopy. Data are representative of three independent experiments.