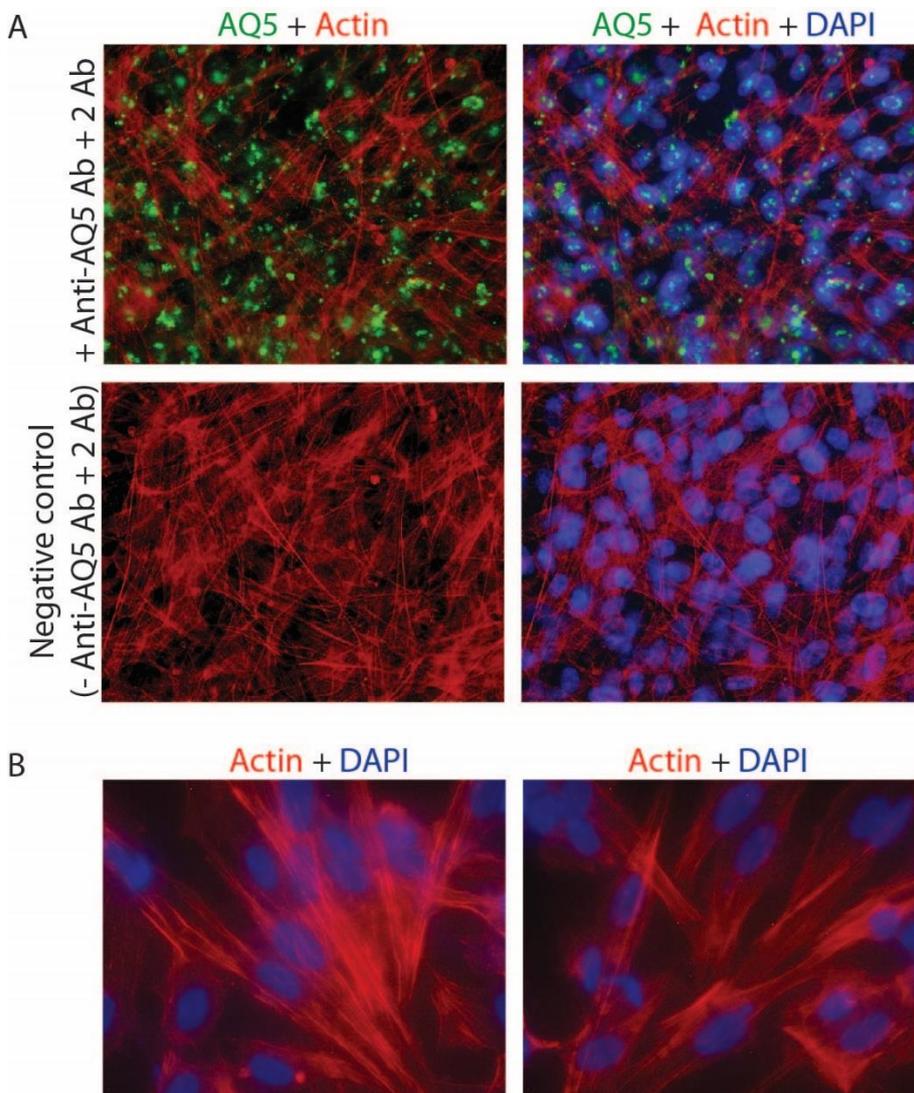
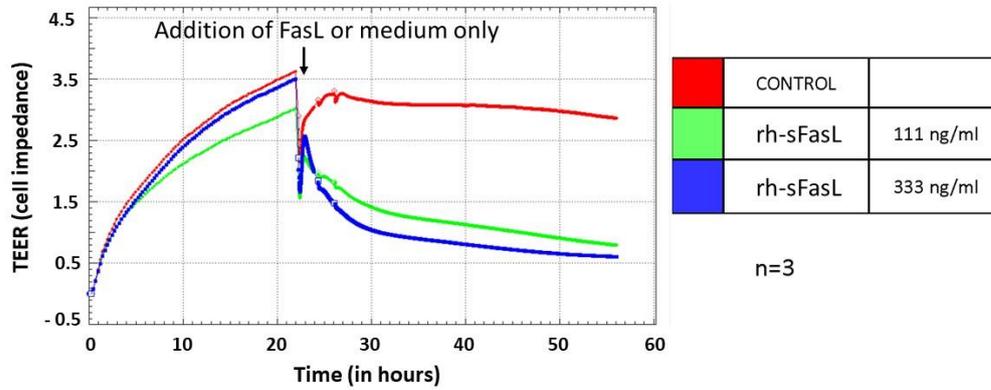


SUPPLEMENTARY FIGURES

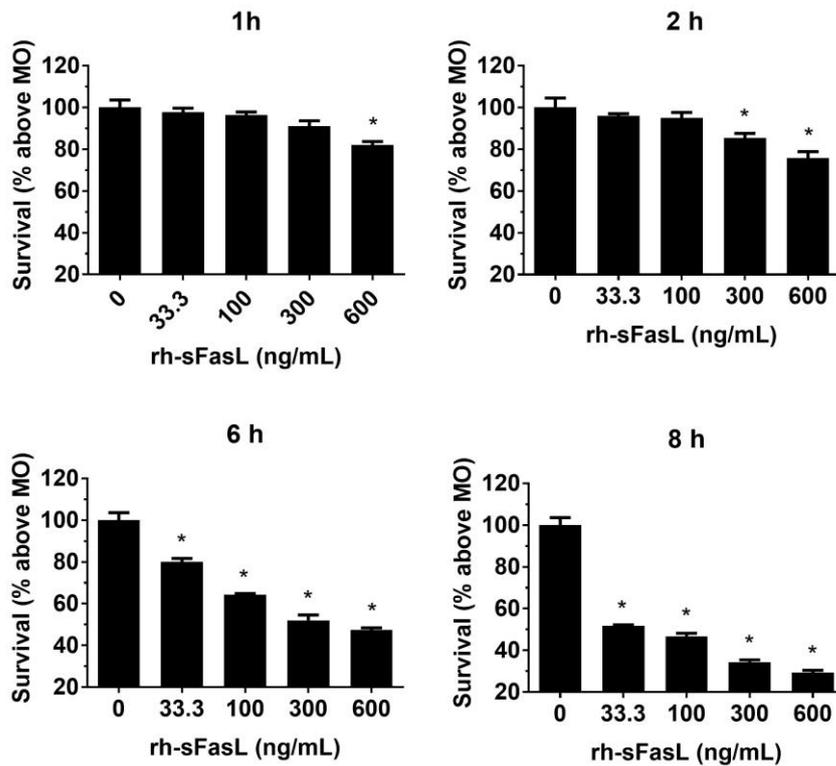
SUPPLEMENTARY FIGURE 1. (A) Representative immunofluorescence images show the expression of AQ5 (green signal) and actin fibers (red signal) in human pulmonary alveolar epithelial cell monolayers. For AQ5 detection, we used a primary anti-human AQ5 antibody (rabbit monoclonal-EPR3747) and a secondary antibody (goat anti-rabbit Alexa Fluor 488-IgG H&L), both obtained from Abcam. The negative control of the immunofluorescence technique, in which the incubation with the primary anti-AQ5 antibody was omitted, shows no green signal. Original image magnification X200. **(B)** Fluorescence staining of actin fibers (red signal) in human pulmonary alveolar epithelial cell monolayers. For actin detection, the cells were stained with Alexa Fluor 568 Phalloidin (Invitrogen). Original image magnification X400. Blue signal corresponds to the DAPI staining of the cell nuclei.



SUPPLEMENTARY FIGURE 2. Transepithelial permeability of human pulmonary alveolar epithelial cell monolayers assessed by the Transepithelial Electrical Resistance (TEER) method before and after incubation with two different concentrations of recombinant human soluble FasL (111 ng/ml and 333 ng/ml) or with medium only (Control). Please, note that sFasL decreased the TEER of these monolayers already at 2 h of incubation.



SUPPLEMENTARY FIGURE 3. Time-course analysis of cell survival in human pulmonary alveolar epithelial cell monolayers treated with recombinant human soluble FasL (rh-sFasL). The graphs show the percentage of survival of human pulmonary alveolar epithelial cells at different time points (1, 2, 6 and 8 h) after treatment with various concentrations of rh-sFasL. Cells with medium only were used as controls. Results from 3 separate experiments performed in duplicate. Results expressed in mean±SD. *P < 0.05 vs medium only (rh-sFasL= 0 ng/mL).



SUPPLEMENTARY FIGURE 4. Effects of genistein at different doses on IL8 expression and survival in human pulmonary alveolar epithelial cells treated with recombinant human soluble FasL (rh-sFasL). The cells were preincubated with different doses of genistein for 1.5 h. Then, rh-sFasL (300 ng/ml) or medium only was added to the cells. The expression of IL8 in the supernatant and the percentage of cell survival were measured 2 h later. 30 μ M was the lowest dose of genistein that prevented the increase in the levels of IL8 induced by sFasL without causing cell death.

