

Figure S1: Cytotoxic effect of ferulic acid on dendritic cells. On day 6 of culture, bone marrow-derived dendritic cells (DCs) were treated with various concentrations of FA for 72 h. Untreated DCs were pulsed with medium alone as the control. These cells were incubated with 10 μ l of 5 mg/mL of MTT for another 4 h, and then solubilized in DMSO. The amount of reduction was measured using a microplate reader at 570 nm. Results from three independent experiments are shown and are expressed as the mean \pm SEM. *** p < 0.001 vs. control DCs.

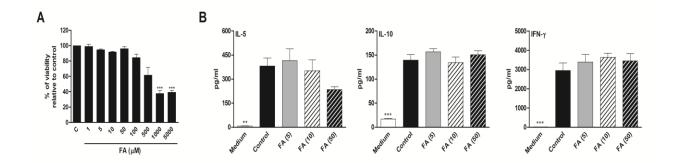


Figure S2: The direct effect of ferulic acid on CD4⁺ T cells. (A) Cytotoxicity effect of FA on T cells. T cells were treated with different doses of FA for 72 h. Cell viability was detected by using MTT assay. Untreated T cells were as the control. Results from three independent experiments are shown and are expressed as the mean \pm SEM. *** p < 0.001 vs. control T cells. (B) The direct effect of FA on T-cell cytokine production. T cells (1 × 10⁶ cells/well) were cultured in 24-well plates and stimulated with anti-CD3 (1 µg/ml)/anti-CD28 (1 µg/ml) antibodies in the presence of FA (5, 10, and 50 µM) for another 72 h. T cells stimulated with anti-CD3/anti-CD28 alone as the control. The culture supernatants were collected and analyzed by an ELISA. Results from triplicate experiments are shown and are expressed as the mean \pm SEM. *** p < 0.01, **** p < 0.001 vs. the control group.

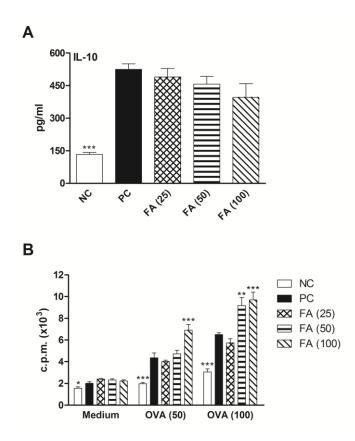


Figure S3: Treatment with ferulic acid enhanced the allergen-specific T-cell proliferative response in ovalbumin (OVA)-induced asthmatic mice. (A) Splenocytes (10^7 cells/ml) from FA-treated and control groups were stimulated with 50 µg/ml OVA in 24-well plates, and culture supernatants were collected after 72 h. Levels of IL-10 cytokine production was analyzed by an ELISA. Results are expressed as the mean \pm SEM (n = 8 in each group). *** p < 0.001 vs. the PC group. (B) 3 x 10^5 spleen cells/well were stimulated with OVA (50 and 100 µg/ml) *in vitro*. After 5 days of culture, cell proliferation was estimated by [3 H]-thymidine incorporation assay. Results are expressed as the mean \pm SEM (n = 8 in each group). * p < 0.05, *** p < 0.001 vs. the PC group.