

Supplementary Materials: Therapeutic Effect of Exogenous Truncated IK Protein in Inflammatory Arthritis

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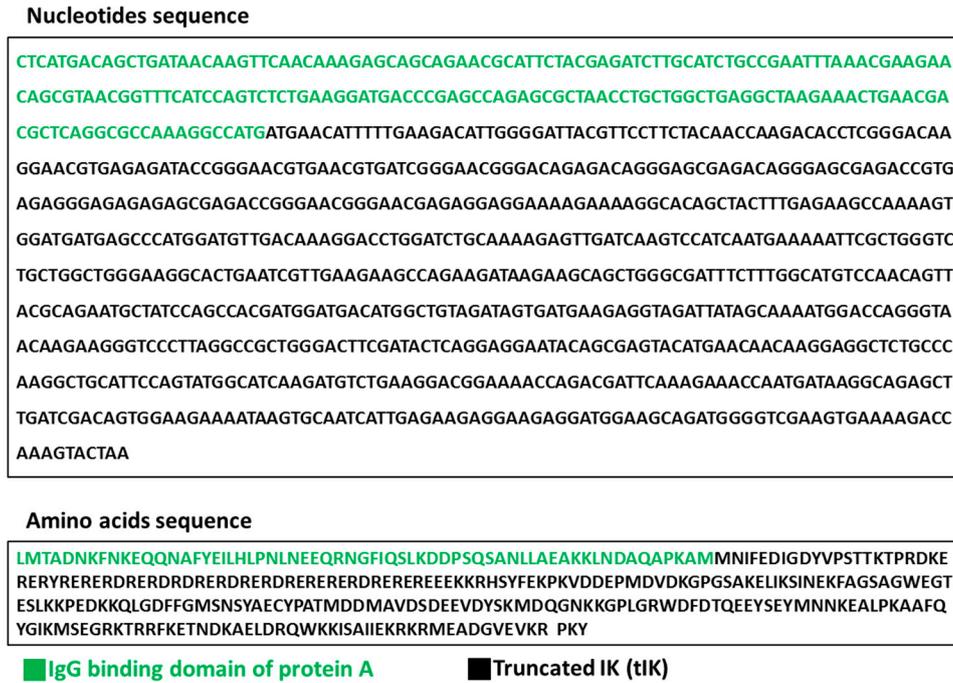


Figure S1. Nucleotide and amino acid sequences of tIK fused with IgG binding domain.

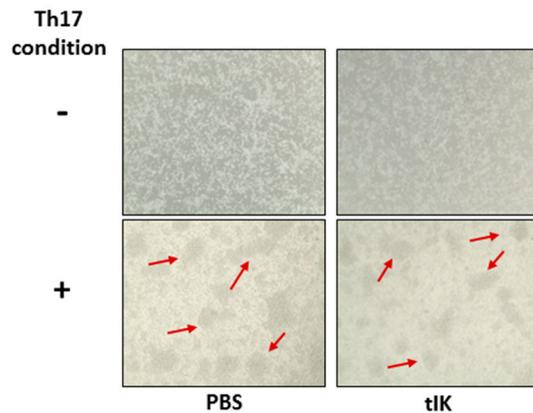


Figure S2. Cell clumps seen in Th17-polarizing medium. CD4⁺ T cells isolated from 7-week-old Balb/c mice using magnetic activated cell sorting were cultured in Th17-polarizing medium or control medium. After 3 days, the cells formed clumps only in Th17-polarizing medium. The red arrows point to the clumps.

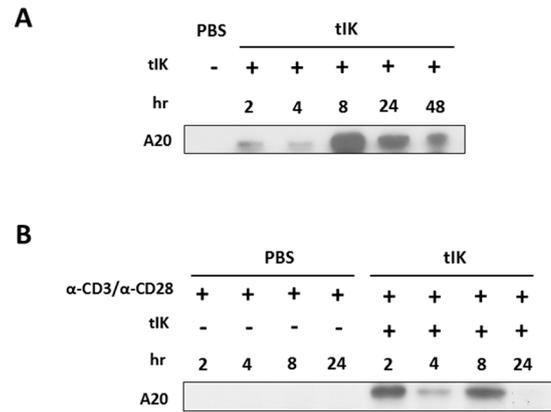


Figure S3. The time course of A20 induction by treatment with exogenous tIK protein. (A) CD4⁺ T cells isolated and cultured as described for Figure 4(A). At 2, 4, 8, 24, and 48 h posttreatment with tIK protein, the cells were harvested and lysed using RIPA buffer containing a protease inhibitor cocktail. A20 was detected in the total extracted protein by Western blot analysis. (B) CD4⁺ T cells were isolated and stimulated as described for Figure 4(B). At 2, 4, 8, and 24, h posttreatment with tIK protein, total protein from cells were extracted and used for Western blot analysis as described for Supplementary Figure 3(A).

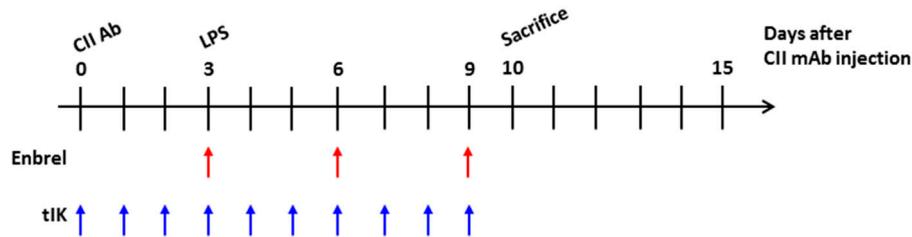
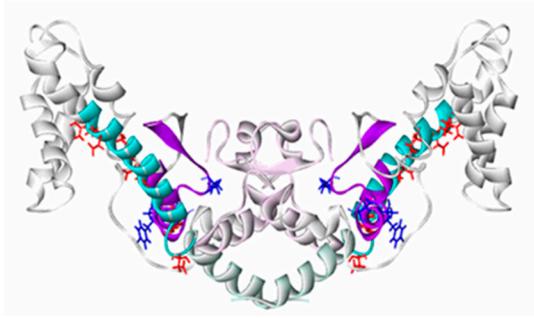
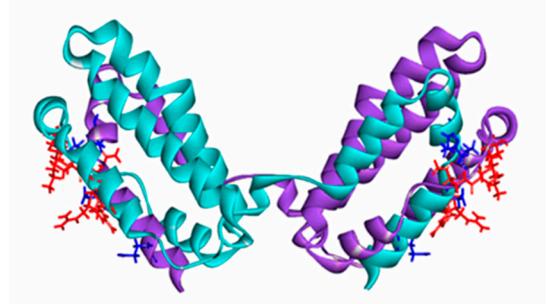


Figure S4. Detailed schedule for induction of CAIA in mice, and the schedule for treatments with tIK protein or Enbrel. CAIA was induced by an intravenous injection of monoclonal antibody against type II collagen into DBA/1J mice. At day 3 after the monoclonal antibody injection, LPS was injected to stimulate the. PBS (vehicle) or tIK protein (tIK) was injected every day; Enbrel was injected every 3 days.



tIK protein



IL-10

Figure S5. 3D structure analysis of tIK protein. The 3D structure of tIK protein and IL-10 dimer were analyzed *in silico* using PyMol Molecular Graphic System (DeLano Scientific LLC).