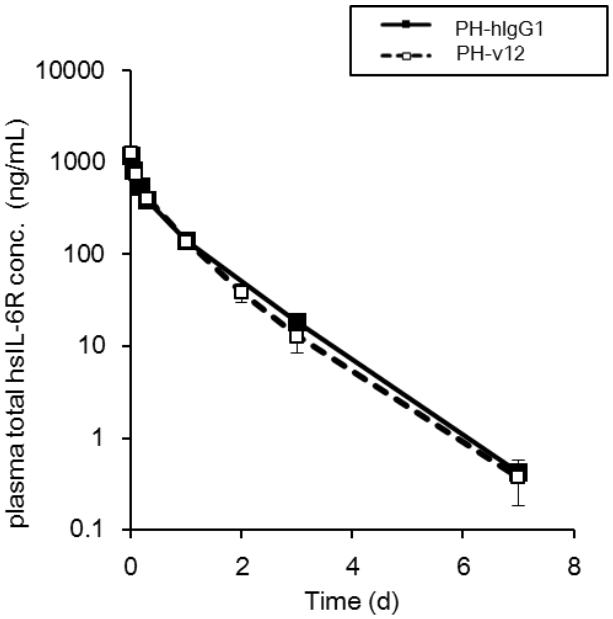
Supplemental Figure 1-A



Supplemental Figure 1-B

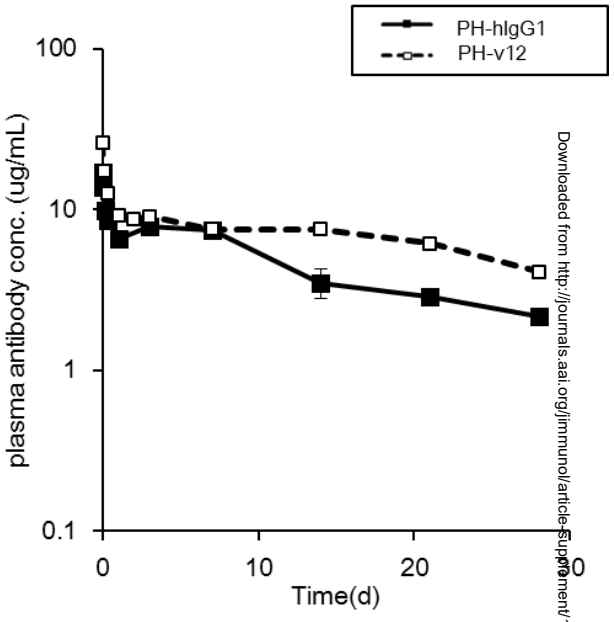
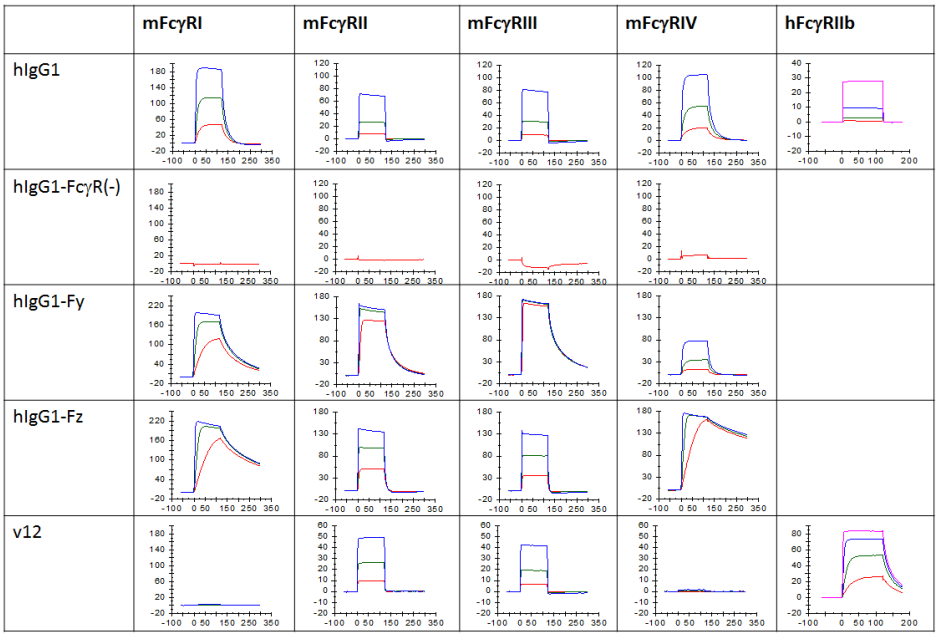


Figure S1: A pH-dependent binding antibody with Fc engineering to selectively increase hFcγRIIb binding did not enhance antigen clearance in wild-type mice

The effect of antibodies on the total hsIL-6R plasma concentration was evaluated in a co-injection model. hsIL-6R and antibody were intravenously administered as single doses of 50 μg/kg for hsIL-6R and 1 mg/kg for antibody. PH-hIgG1 and PH-v12 were each co-injected with hsIL-6R in wild-type mice and time profiles of (A) total hsIL-6R plasma concentration and (B) antibody plasma concentration are shown. Each data point represents the mean ± s.d. (n=2 or 3 each)

Supplemental Figure 2-A



Supplemental Figure 2-B

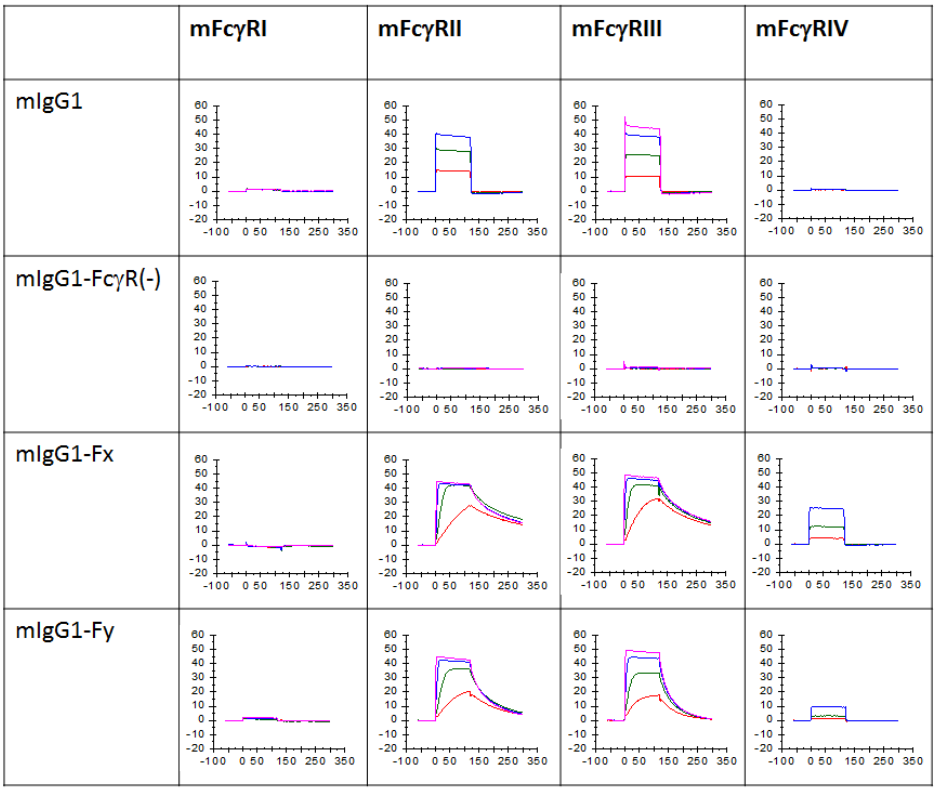


Figure S2: Sensorgrams of Fc variants binding to FcγRs

SPR sensorgrams for representative set of Fc variants as described in Table I, II are shown. X-axis represents time (sec) and y-axis represents binding response (RU). Each analysis were conducted as described in Materials and Method under several different concentrations of FcγR.

Supplemental Figure 3

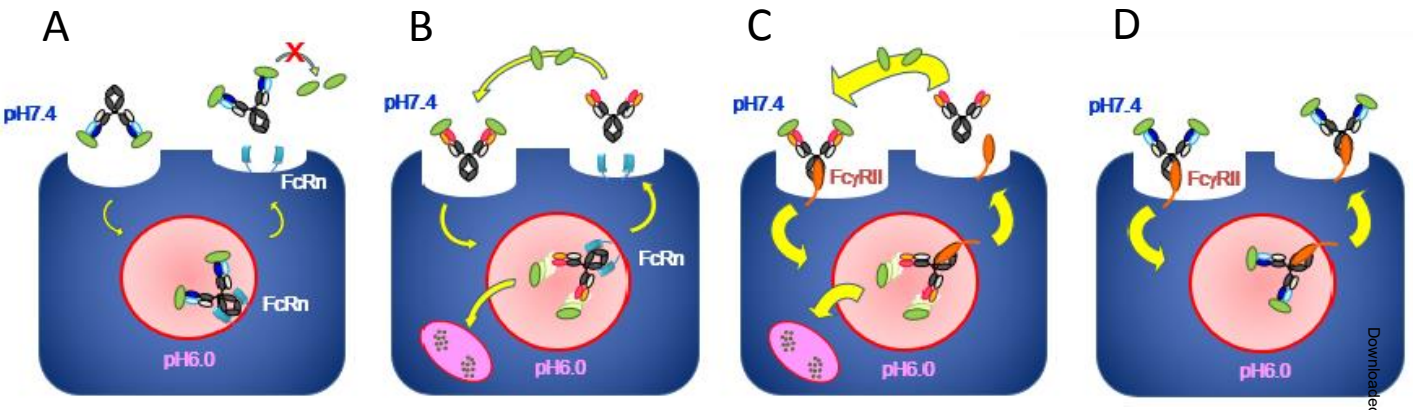


Figure S3:Proposed mode of action of pH-dependent antibody in comparison with conventional antibody. (A) Conventional antibody bound to soluble antigen is slowly taken up by nonspecific pinocytosis. Since wild-type IgG1 antibody has weak FcγRII binding affinity, this interaction is diminished under high endogenous IgG competition. Antigen-antibody complex binds to FcRn in acidic endosome, and is recycled back to the cell surface and released from FcRn back to circulation. (B) pH-dependent binding antibody (recycling antibody) bound to soluble antigen is also nonspecifically taken up by pinocytosis as conventional antibody, and binds to FcRn in acidic endosome, while antigen is dissociated from the antibody, transferred into lysosome and degraded. Antibody is recycled back to the cell surface by FcRn, released from FcRn back to circulation and binds to another antigen, allowing single antibody to bind to antigen multiple times. (C) pH-dependent antibody with FcγRII binding capability bound to soluble antigen is taken up by FcγRII-mediated endocytosis. In acidic endosome, antigen is dissociated from the antibody, transferred into lysosome and degraded. Antibody is recycled back to the cell surface and released from FcγRII back to circulation to bind to another antigen. Enhanced FcγRII binding allows rapid FcγRII-mediated uptake and enhances lysosomal antigen degradation rate. (D) Conventional antibody with FcγRII binding capability bound to soluble antigen is taken up by FcγRII-mediated endocytosis. Antigen stay bound to the antibody in acidic endosome, and antigen-antibody complex is recycled back to the cell surface and released from FcγRII back to circulation. Although enhanced FcγRII binding allows rapid FcγRII-mediated uptake, but it does not significantly enhances lysosomal antigen degradation rate.