

# European Journal of Immunology

**Supporting Information**

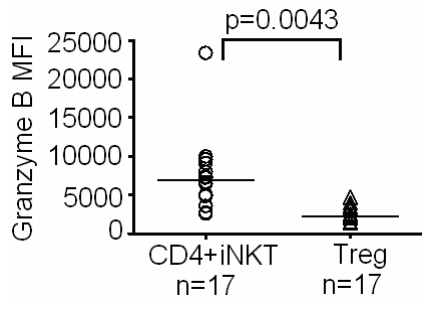
**for**

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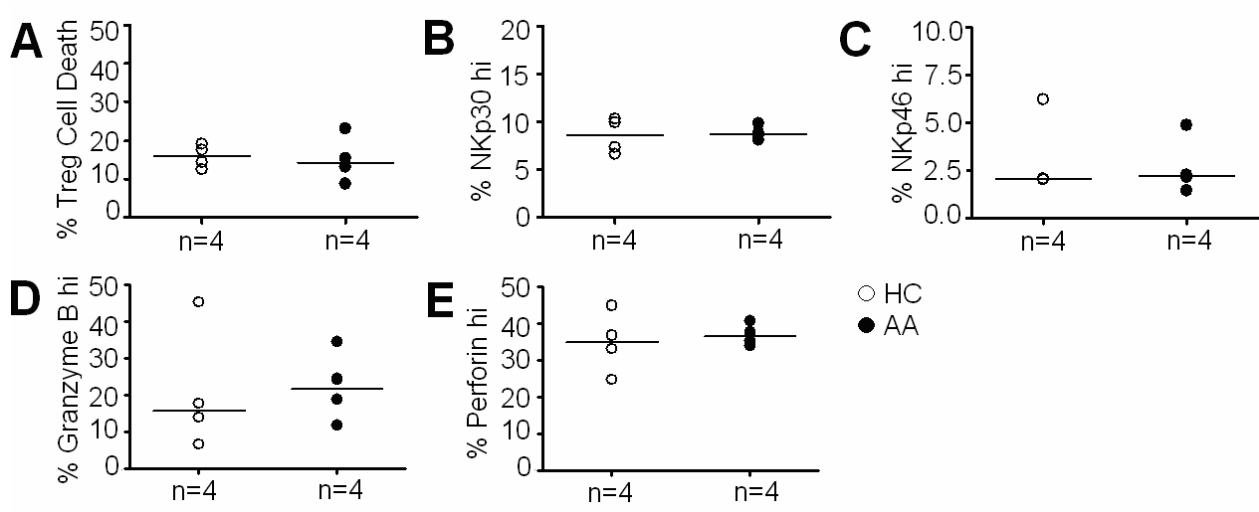
**Increased cytotoxicity of CD4<sup>+</sup> invariant NKT cells against CD4<sup>+</sup>CD25<sup>hi</sup>CD127<sup>lo/-</sup>  
regulatory T cells in allergic asthma**

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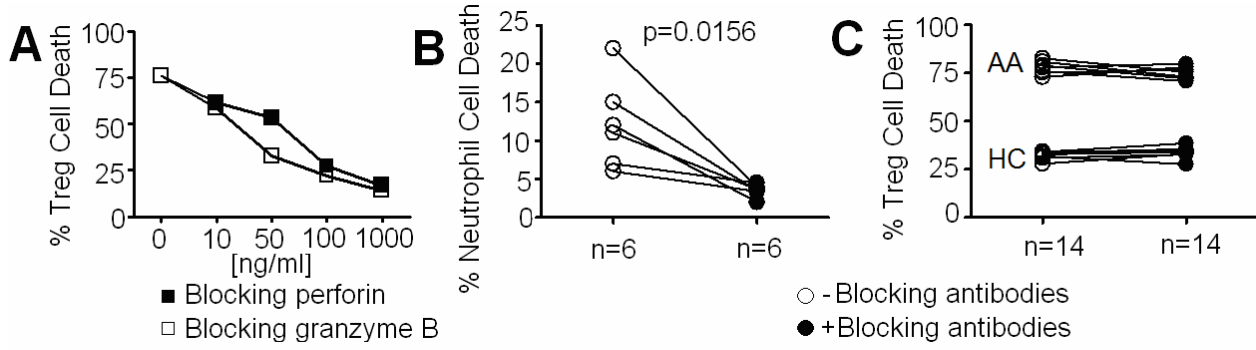
**SUPPLEMENTARY FIGURE 1**



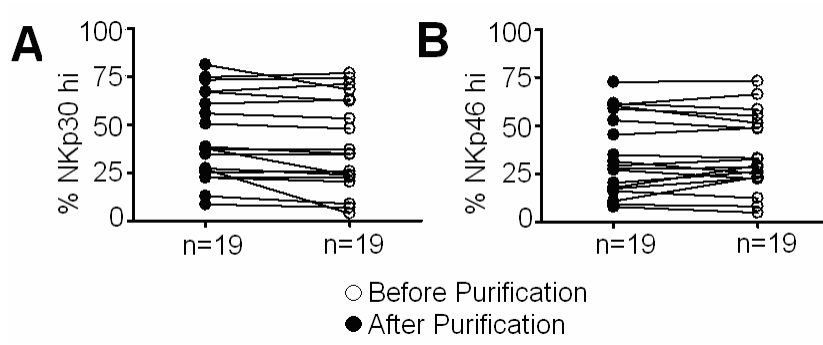
**SUPPLEMENTARY FIGURE 2**



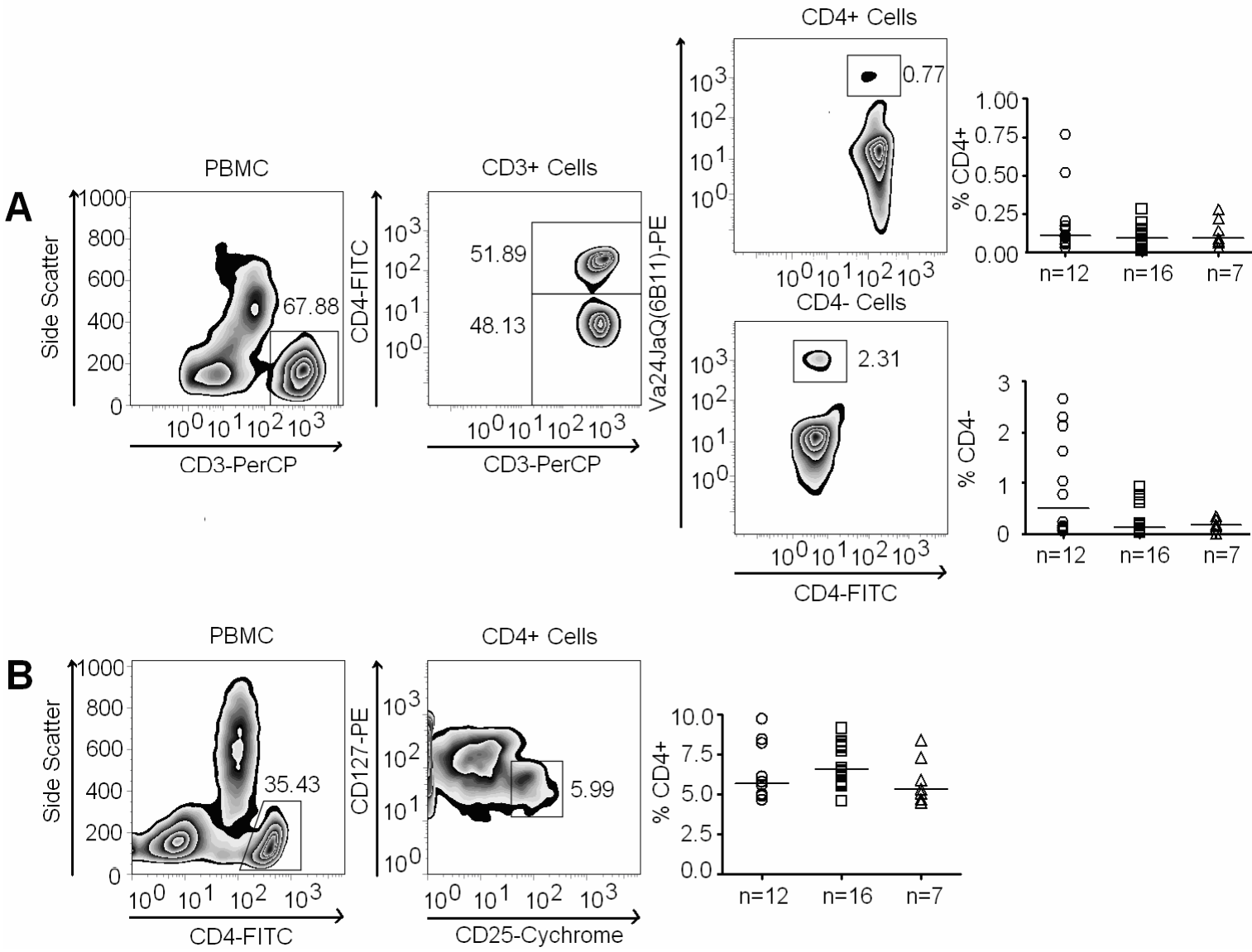
**SUPPLEMENTARY FIGURE 3**



**SUPPLEMENTARY FIGURE 4**



**SUPPLEMENTARY FIGURE 5**



**SUPPLEMENTARY FIGURE 1. Expression of granzyme B in CD4<sup>+</sup> iNKT and Treg.** Paired t-tests were used for statistical analysis. P values represented results from t-tests. Samples represent data from HC.

**SUPPLEMENTARY FIGURE 2. Phenotype and cytotoxicity of CD4<sup>-</sup> iNKT.** **A.** Percentages of target cell death in killing assays of CD4<sup>-</sup> iNKT against autologous Treg targets. **B.** Percentage distribution of CD4<sup>-</sup>NKp30<sup>hi</sup> iNKT. **C.** Percentage distribution of CD4<sup>-</sup>NKp46<sup>hi</sup> iNKT. **D.** Percentage distribution of CD4<sup>-</sup>granzyme B<sup>hi</sup> iNKT. **E.** Percentage distribution of CD4<sup>-</sup>perforin<sup>hi</sup> iNKT. Unpaired t-tests were used for statistical analysis. Horizontal bars represented median values.

**SUPPLEMENTARY FIGURE 3. Effects of inhibitors of cell death pathways on CD4<sup>+</sup> iNKT cytotoxicity.** **A.** Titration of blocking antibodies against granzyme B and perforin in killing assays of CD4<sup>+</sup> iNKT against Treg. An AA representative was used in the titration experiment. **B.** Positive control for FasL blocking experiments. Neutralizing FasL antibodies blocked serum-derived FasL mediated apoptosis of neutrophils from juvenile idiopathic neutropenic subjects. **C.** Effects of an NKp30 blocking antibody in killing assays of CD4<sup>+</sup> iNKT against Treg. Data were collected from equal numbers of HC and AA subjects. Paired t-tests were used for statistical analysis. P values represented results from t-tests.

**SUPPLEMENTARY FIGURE 4. Effects of magnetic purification on the expression of natural cytotoxicity receptors in CD4<sup>+</sup> iNKT.** **A.** Expression of NKp30 on CD4<sup>+</sup> iNKT before and after purification. **B.** Expression of NKp46 on CD4<sup>+</sup> iNKT before and after purification. Data were collected from 7 HC, 7 AA, and 5 NA subjects. Paired t-tests were used for statistical analysis.

**SUPPLEMENTARY FIGURE 5. Distribution of iNKT and Treg subsets in peripheral blood.** **A.** Percentage distribution of CD4<sup>+</sup> iNKT and CD4<sup>-</sup> iNKT subsets. **B.** Percentage distribution of Treg. ANOVA was used for statistical analysis. Horizontal bars represented median values.