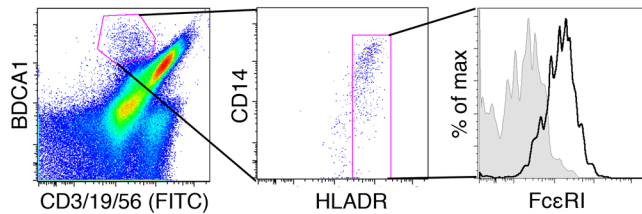


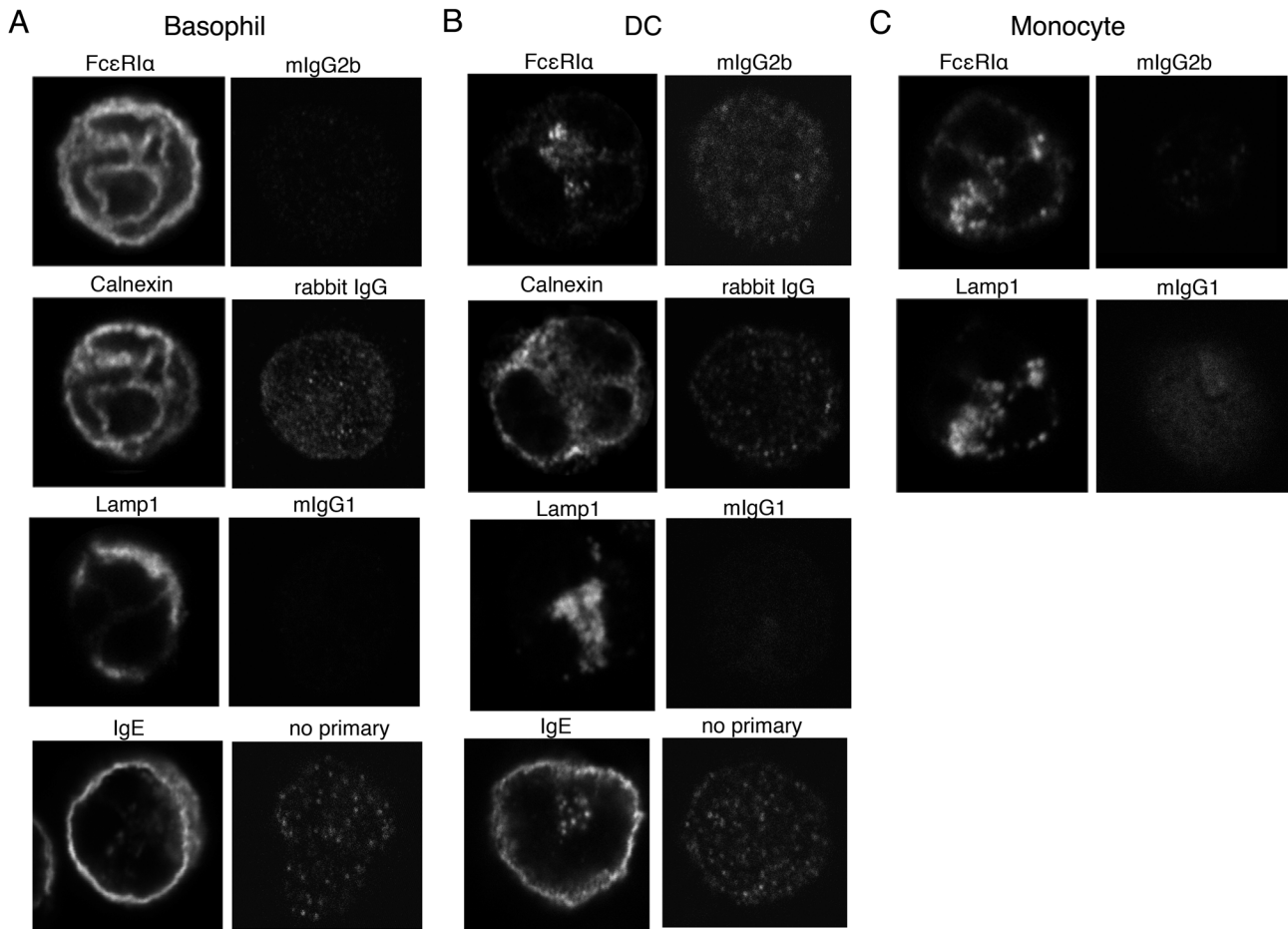
Subjects (n)	11
Age, mean $\pm$ SD	29.55 $\pm$ 3.47
Gender	
Male, n (%)	7 (64)
Female, n (%)	4 (36)
Race (n)	
Caucasian	6
Asian	5

**Supplemental Table 1: Human blood donors.**

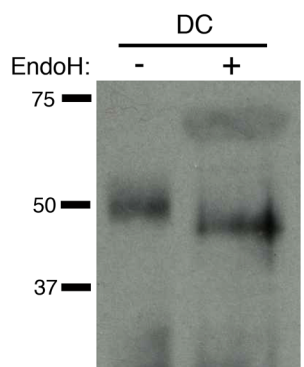


**Supplemental Figure 1: Expression of FcεRI in human lung BDCA1+ DCs.**

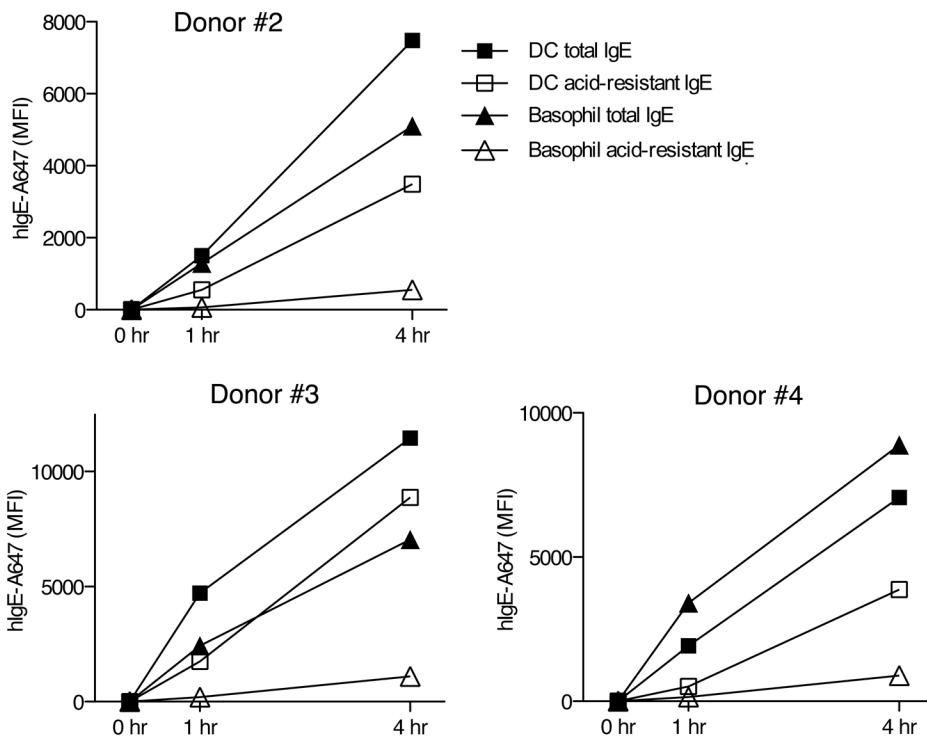
Live, singlet cells were stained with a cocktail of antibodies. BDCA1+ DCs were gated by selecting BDCA1+[CD3/19/56]- cells, then further selecting HLADR+ cells. BDCA1+ DCs are FcεRI+, as shown in the histogram on the right. Anti-hFcεRIα antibody (CRA-1) is black, and isotype control is shown in grey. Data shown are from one representative donor of 4 tested.



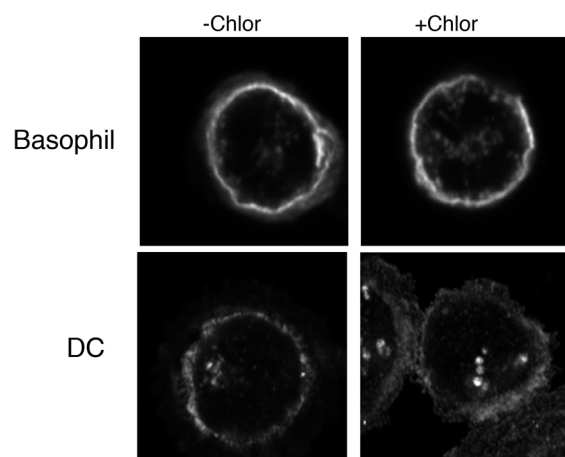
**Supplemental Figure 2: Isotype control stains for confocal microscopy of human cells.** Basophils (**A**), DCs (**B**), and monocytes (**C**) were isolated as per Methods and were stained with antibodies indicated (left panels) or matched isotype control antibodies (right panels). Note that for basophils and DCs, a 'no primary' antibody control was used for IgE.



**Supplemental Figure 3: Second representative Western blot of FcεRIα.** DCs were isolated according to methods and some immunoprecipitated FcεRIα was treated with EndoH. The star indicates EndoH band which cross-reacts with FcεRIα antibody.

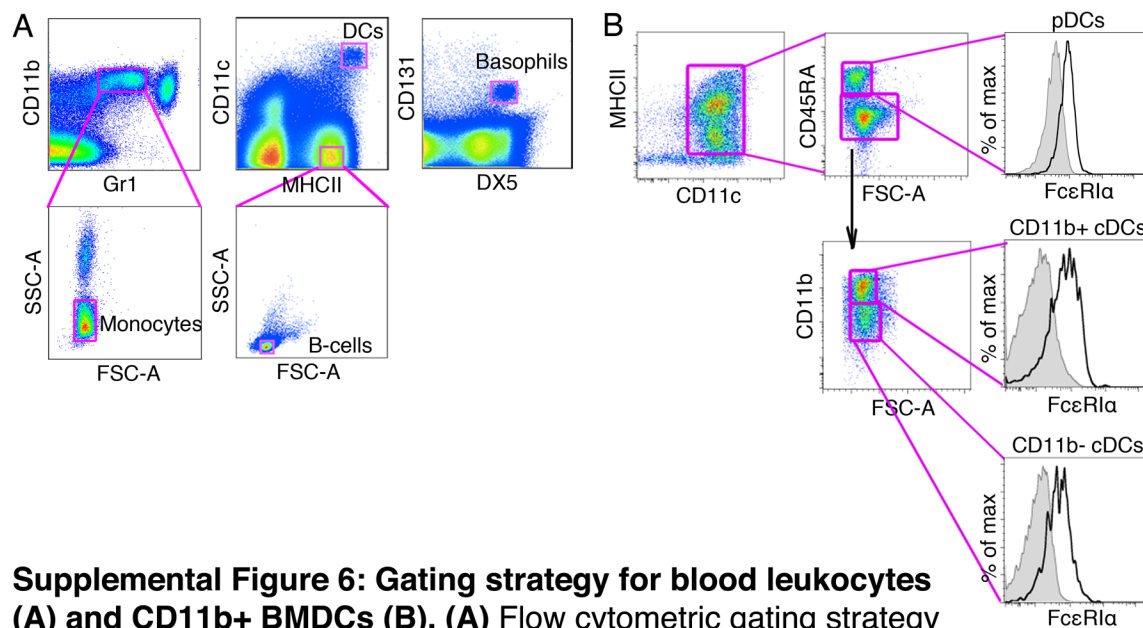


**Supplemental Figure 4: hlgE-A647 binding and internalization by DCs and basophils of three additional healthy blood donors.** Experiments were run in parallel with that shown in Figure 3C.

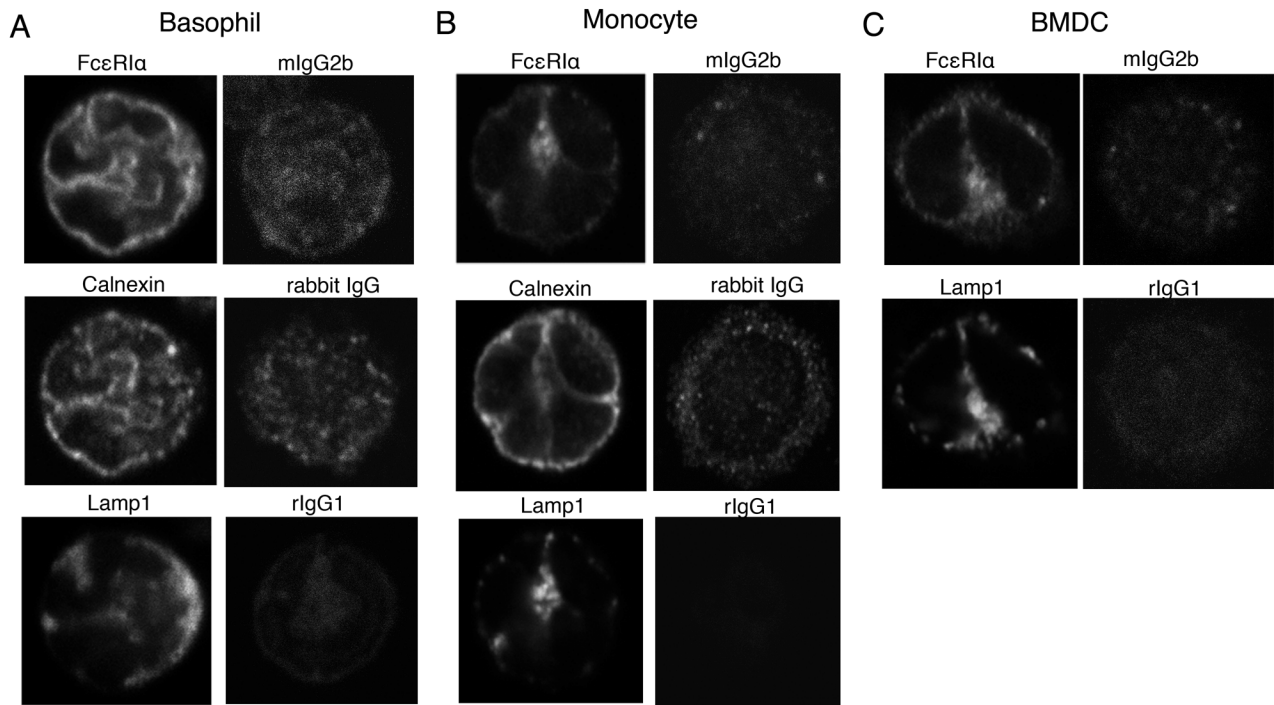


**Supplemental Figure 5: The effect of chloroquine on IgE-containing intracellular compartments.**

Basophils and DCs were incubated for 8 hrs at 37° C with or without 0.5  $\mu$ M chloroquine (Chlor) and stained with anti-IgE antibody for confocal microscopy. The same laser intensity was employed for chloroquine-treated and untreated cells. Images are representative from 2 unique donors.

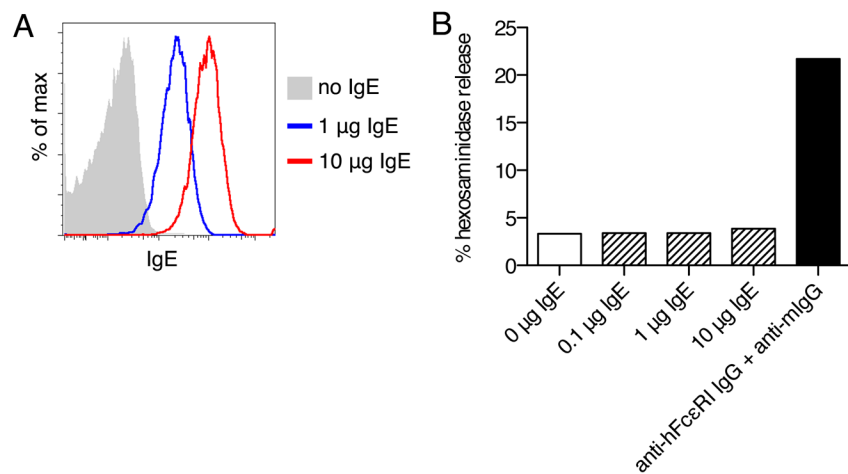


**Supplemental Figure 6: Gating strategy for blood leukocytes (A) and CD11b+ BMDCs (B).** (A) Flow cytometric gating strategy used for the experiment described in Figure 4A. (B) Bone marrows were isolated from hFcεRIα-Tg mice and cultured for 6 days in the presence of 100 ng/mL Flt3L. Generated cells were stained using a cocktail of antibodies and each DC subset was gated as shown. Expression of FcεRI (Black) was also determined compared to isotype control (grey).

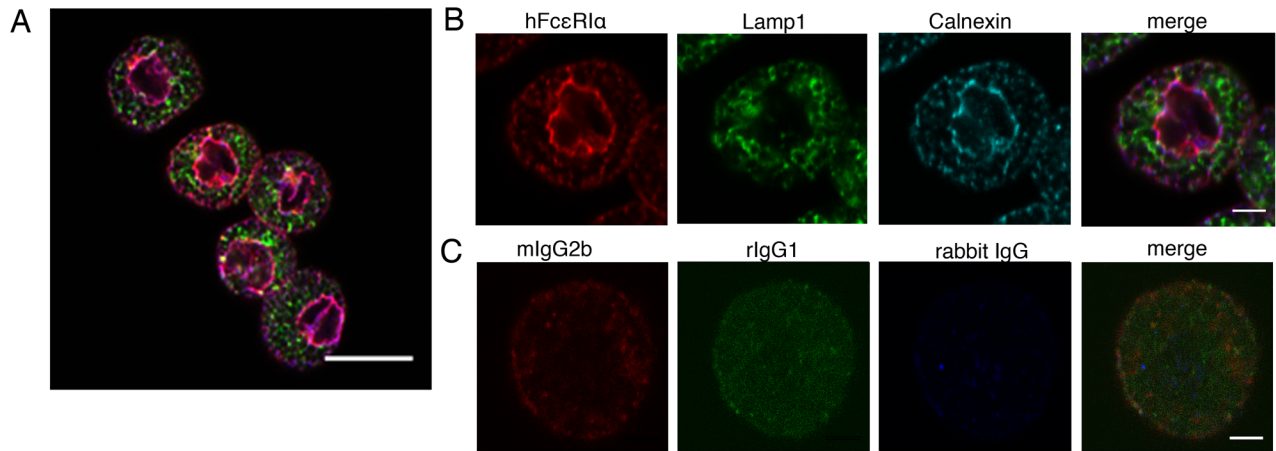


**Supplemental Figure 7: Isotype controls for confocal microscopy of mouse blood cells and BMDCs.** Blood Basophils (A), monocytes (B), and CD11b<sup>+</sup> Flt3L-derived BMDCs (C) were isolated as described in methods and were stained with antibodies indicated (left columns) or with the corresponding isotype controls (right columns). Images were taken with the same laser intensity and gain for the antibodies indicated compared to isotype controls.

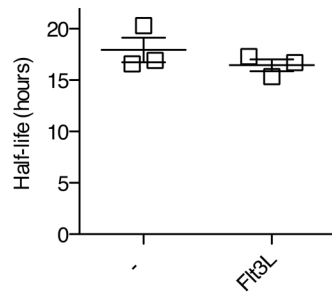




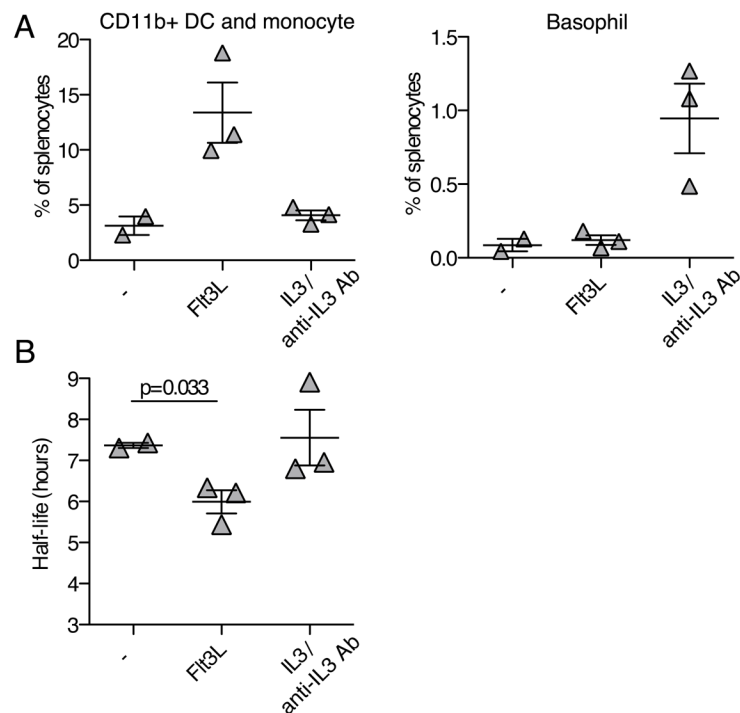
**Supplemental Figure 8: Purified human IgE (Abcam) does not degranulate hFcεRI-expressing bone-marrow derived mast cells (BMMCs).** BMMCs cultured from hFcεRIα-transgenic mice were incubated with varying concentrations of human IgE (Abcam) or anti-hFcεRI IgG/anti-mouse IgG Ab complex. **(A)** Binding of IgE was determined by flow cytometry. **(B)** Degranulation was determined by hexosaminidase assay.



**Supplemental Figure 9: hFcεRIα in peritoneal mast cells of hFcεRI-Tg mice is located in the ER, not in the lysosome.** Peritoneal mast cells were isolated from peritoneal lavage cells of hFcεRIα-transgenic mice by flow-cytometric sorting based on expression of c-kit and side-scatter properties. Sorted cells were placed on coverslips and stained as described in methods for hFcεRIα, Lamp1, and calnexin, then imaged by confocal microscopy at 60x. **(A)** A group of mast cells. hFcεRIα is in red, Lamp1 is in green, and Calnexin is in blue. Size bar is 10 μm. **(B)** One mast cell imaged at higher magnification. Note that Calnexin is changed to cyan in single-color format for ease of visualization. Size bar is 2.5 μm. **(C)** Isotype controls for stains shown in (B). Images were taken with the same laser intensity and gain for the antibodies indicated compared to isotype controls.



**Supplemental Figure 10: Flt3L does not accelerate serum hlgE clearance in Tg<sup>-</sup> mice.** Half-lives of human IgE in Tg<sup>-</sup> mice or Tg<sup>-</sup> mice implanted with Flt3L-transfected B16 melanomas. Half-lives are not significantly different as measured by Student's T-test.



**Supplemental Figure 11: Flt3L, but not IL-3, accelerates serum IgE clearance in 10:90 (Tg<sup>+</sup>:Tg<sup>-</sup>) mixed chimeric mice. (A)** The percentage of [CD11b<sup>+</sup> DCs + monocytes] or basophils in spleen was determined by flow cytometry. Data are plotted as mean ± SEM. **(B)** Serum human IgE half-lives were determined as described in Figure 6. Significance was determined by Student's T-test.