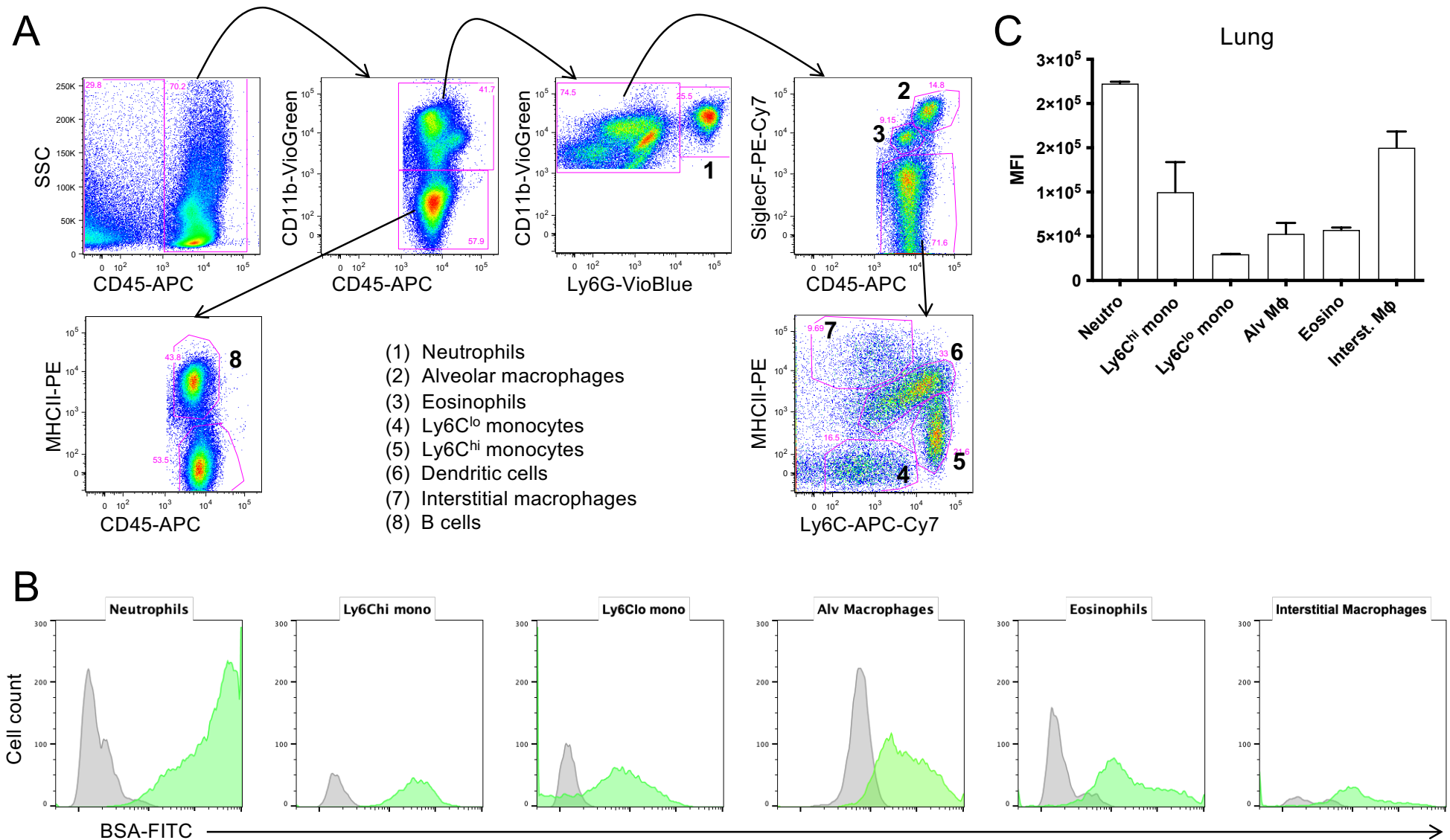
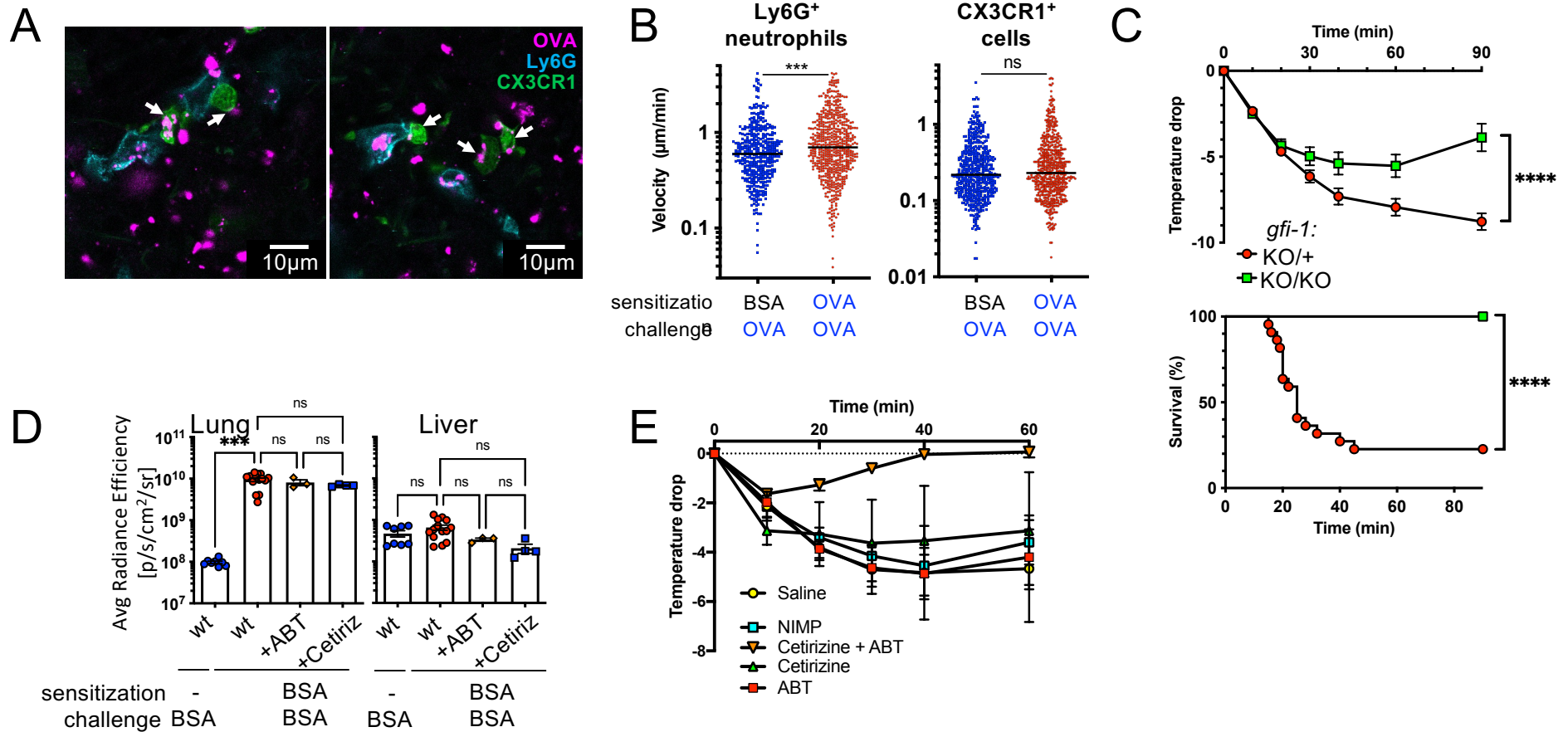


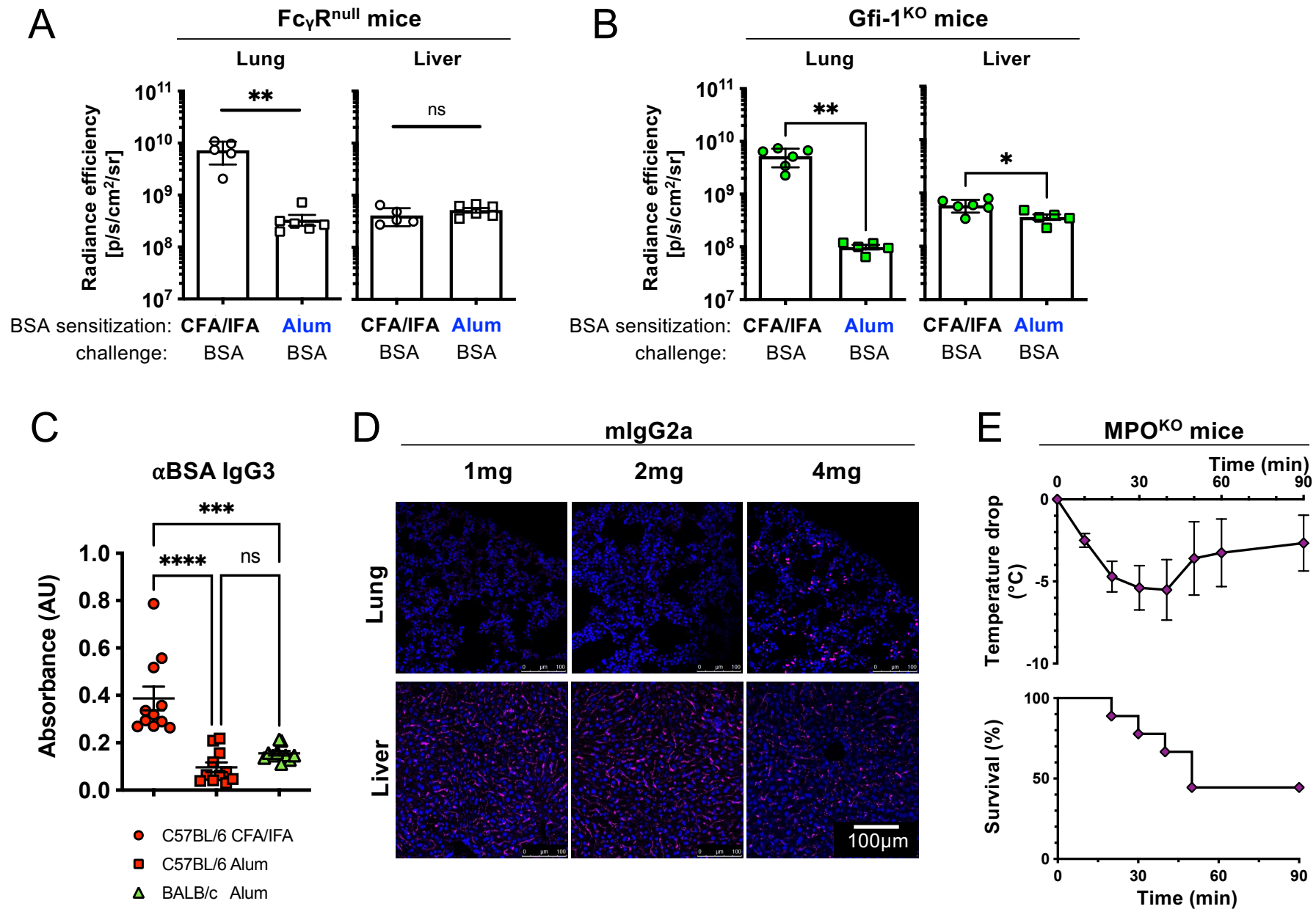
**Supplemental Figure 1. Sensitization-specific antigen accumulation in the lung during active systemic anaphylaxis.** A, In vivo quantification of antigen in C57BL/6 mice sensitized or not (-) with BSA in CFA/IFA, 5 min after challenge with indicated fluorescent antigens. B-D, Post-challenge ex vivo quantification of BSA-VT680 in explanted organs of C57BL/6 sensitized or not with BSA in CFA/IFA, and challenged with (C) 500  $\mu\text{g}$  BSA, or (D) 500, 50 or 10  $\mu\text{g}$  BSA. Data are pooled from 2 independent experiments ( $n = 5-14$  per group). Individual measurements, mean  $\pm$  SEM and p values are indicated: ns: non-significant; \* $P < .05$  or \*\*\*\* $P < .0001$ . (A-B) Data are representative of 3 independent experiments ( $n = 3$  per group). (C-D) Data are pooled from 2 independent experiments, Individual measurements, mean  $\pm$  SEM and p-values are indicated: ns, non-significant; \*\*\* $P < .001$  or \*\*\*\* $P < .0001$ .



**Supplemental Figure 2. Antigen accumulation in various lung cells following antigen challenge.** A-B, Flow cytometry gating strategy (A) and example histograms of BSA-FITC intensity (B) for BSA-FITC post-challenge of lung cells in C57BL/6 mice sensitized with BSA in CFA/IFA. C, Flow cytometry quantification of mean fluorescence intensity of BSA-FITC among the indicated cell populations in the lung 15 min post-challenge with BSA-FITC in C57BL/6 mice sensitized with BSA in CFA/IFA. Data are pooled from 2 independent experiments.



**Supplemental Figure 3. Antigen-capture and velocity of cells in the lung during active systemic anaphylaxis, and mechanisms of anaphylaxis following alum immunizations.** A-B, (A) Two-photon microscopy images of fresh lung slices, and (B) intra-tissular velocity ( $\mu\text{m}/\text{min}$ ) of neutrophils (Ly6G-stained cells) or CX3CR1<sup>+</sup> cells recorded over 39 min on fresh lung slices, 5 min post-challenge with OVA in CX3CR1-GFP mice immunized with OVA in CFA/IFA; Mice were injected with anti-Ly6G-BV421 mAb to label neutrophils; (A) arrows indicate evidence of antigen uptake by CX3CR1<sup>GFP</sup> cells; (B) Individual measurements for each cell are represented for neutrophils (BSA/OVA n=421; OVA/OVA n=601) and CX3CR1<sup>+</sup> cells (BSA/OVA n=555; OVA/OVA n=601) as well as the median and p values: ns: non-significant, \*\*\*P < .001. C, Temperature drop (top) and survival (bottom) during experimental anaphylaxis in *gfi-1*<sup>-/-</sup> mice (n=12) and *gfi-1*<sup>+/-</sup> littermate controls (n=20). Data are pooled from 2 independent experiments. D, Post-challenge quantification of BSA-VT680 in explanted organs of C57BL/6 mice immunized with BSA in CFA/IFA injected with saline (wt), PAF-R antagonist (ABT) or histamine-receptor antagonist (cetirizine; Cetiriz). Individual measurements, mean  $\pm$  SEM and p values are indicated: ns: non-significant, \*\*\*P < .001 or \*\*\*\*P < .0001. E, Post-challenge changes in body temperature of C57BL/6 mice immunized with BSA in CFA/IFA injected with saline, histamine-receptor antagonist (cetirizine; Cetiriz) in combination or not with PAF-R antagonist (ABT), or anti-Ly6G mAb (NIMP). Data are represented as mean  $\pm$  SEM and are representative of at least 2 independent experiments (n $\geq$ 3).



**Supplemental Figure 4. Antigen retention in Fc $\gamma$ R<sup>null</sup> and Gfi-1<sup>KO</sup> mice following alum or CFA/IFA immunization, and IgG2a antibody mediated antigen retention in the lung.** A-B, Post-challenge ex vivo quantification of BSA-VT680 in explanted organs of (A) Fc $\gamma$ R<sup>null</sup> mice (n $\geq$ 5) and (B) Gfi-1<sup>KO</sup> mice (n $\geq$ 5) immunized with BSA in alum or CFA/IFA. C, Serum anti-BSA IgG3 titers in C57BL/6 or BALB/c mice immunized with BSA in CFA/IFA or alum 10 days after the last immunization. D, 15 min post-challenge immunofluorescence on frozen sections during passive systemic anaphylaxis in C57BL/6 mice. Arrows indicate antigen aggregates. E, Temperature drop (top) and survival (bottom) during experimental anaphylaxis in MPO<sup>-/-</sup> mice (n=9) immunized with BSA in CFA/IFA and challenged with BSA. Data are pooled from 2 independent experiments. (A-C) Individual measurements, mean  $\pm$  SEM are represented as mean  $\pm$  SEM and are representative of at least 2 independent experiments, with p values indicated: ns: non-significant, \*P < .05, \*\*P < .01, \*\*\*P < .001 or \*\*\*\*P < .0001.