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Supplemental information

Rapid GPR183-mediated recruitment

of eosinophils to the lung after

Mycobacterium tuberculosis infection

Andrea C. Bohrer, Ehydel Castro, Claire E. Tocheny, Maike Assmann, Benjamin Schwarz, Eric Bohrnsen, Michelle A. Makiya, Fanny Legrand, Kerry L. Hilligan, Paul J. Baker, Flor Torres-Juarez, Zhidong Hu, Hui Ma, Lin Wang, Liangfei Niu, Zilu Wen, Sang H. Lee, Olena Kamenyeva, Tuberculosis Imaging Program, Keith D. Kauffman, Michele Donato, Alan Sher, Daniel L. Barber, Laura E. Via, Thomas J. Scriba, Purvesh Khatri, Yanzheng Song, Ka-Wing Wong, Catharine M. Bosio, Amy D. Klion, and Katrin D. Mayer-Barber



Eosinophil and neutrophil staining and gating strategy in rhesus macaques:

Representative FACS plots and NHP granulocyte gating strategy in BAL and WB for rhesus macaque eosinophils (orange gate) and neutrophils (blue gate).

Bohrer/Castro et al. Supplemental Figure S2, related to Figure 2



Granulocyte and myeloid cell gating strategy and kinetics of granulocytes after Mtb infection in mice:

(A) Representative FACS plots and gating strategies in WT B6 mouse lung 14 days after aerosol (100-300 CFU) infection in WT B6 mice for eosinophils (orange), neutrophils (blue), AM (green) and pan-myeloid monocytes/macrophages/DC (black)

(B) Total cell number of lung parenchymal (CD45 i.v. negative) eosinophils and neutrophils over time after aerosol infection in WT mice (n=6-25 per time-point, M+F, 95%CI, SEM, 2-3 experiments per time-point)

(C) E/N ratio in lung over course of Mtb infection in WT mice (n=6-25 per time-point, 2-3 experiments per time-point)



Eotaxin levels in airways and lungs of Mtb infected rhesus macaques and mice:

(A) Eotaxin levels in BAL of rhesus macaques over the course of Mtb infection, dotted line represents limit of detection, Eotaxin-2 (CCL24) was undetectable (n=3, d7-14 time points are highlighted in grey)
(B) EPX and MBP (major-basic-protein) levels in BAL of Mtb infected rhesus macaques (n=3) across all timepoints correlated with E/N ratio in same samples (Spearman)

(C) Lung tissue gene expression by qRT-PCR for Ccl11, Ccl24 and Ccl26 after low dose (100-300 CFU) aerosol Mtb infection in B6 mice (n=5-12, 2 experiments)



GPR183 expression in human TB lesion:

(A) Flow cytometric gating strategy of human lung TB lesions for GPR183 expression (n=5)

(B) qRT-PCR of 18FDG PET/CT low or high signal intense human TB lung lesions for CH25H and GPR183 (n=4, Wilcoxon-matched pairs)



GPR183 expression on eosinophils is dispensable for lung migration after SARS-CoV-2 or in allergic asthma: (A) Frequency of lung parenchymal eosinophils in WT and Gpr183-/- competitive mixed BM chimeric mice prior to infection (M, n=13, 2 experiments)

(B) Quantification of CD45 i.v. negative WT or Gpr183-/- eosinophils from competitive mixed BM chimeric mice three to five days (F+M, n=9, 2 experiments) post intranasal SARS-CoV2 infection (B.1.351, Wilcoxon-matched pairs not significant)

(C) Quantification of CD45 i.v. negative WT or Gpr183-/- eosinophils from competitive mixed BM chimeric mice (2 experiments) after OVA-Alum immunization and three (F, n=4) or five days (M, n=5) post intranasal OVA challenge (Wilcoxon-matched pairs not significant, dotted black line shows average CD45 i.v. negative eosinophils or neutrophils in uninfected mixed BM chimeric mice)