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INSIGHTS

First impressions count in TB

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***Mycobacterium tuberculosis* drives expression of type I IFN–mediated neutrophil accumulation, which limits interaction between CD4 T cells and macrophages. Failure to limit type I IFN very early in the interaction between Mtb and immune cells allows rapid progression of disease (Branchett et al. https://doi.org/10.1084/jem.20250466; Gern et al. https://doi.org/10.1084/jem.20250161).**

In this issue of *JEM*, the importance of the very earliest interaction between *Mycobacterium tuberculosis* (Mtb), the causative agent of tuberculosis (TB), and the immune cells of the lung is shown to be critical in determining the long-term outcome of disease (Branchett et al., 2025). The battle is shown to be not so much between the bacteria and the lung but between innate-derived type I IFN and acquired expression of type II IFN—and timing is all.

Why do we care about these early interactions? We care because while nearly 11 million people fall ill with TB every year, we do not know why infection appears inconsequential for 95% of those infected (WHO, 2024). Not knowing what the underpinning mechanisms driving disease are, makes it difficult to intervene—essentially, how do you improve on a 95% success rate of the primary immune response? In people (and animals), the essential protective components of TB immunity are expressed effectively in the majority of infectious foci, but occasionally a site of infection fails to express immunity despite ongoing control within other foci in the same organ (Gideon et al., 2015; Rich, 1944). Why immunity is expressed under some conditions and not others has been investigated using animal models of aerosol infection with Mtb (Kramnik and Beamer, 2016); these models allow hypothesis testing to define the mechanistic pathways driving disease (Cooper, 2014). As the tools available for unbiased spatial and single-cell analysis have improved, the animal models have become more sensitive and specific and are now providing detailed insight into the early events in TB (Branchett et al., 2025; Gern et al., 2025). It is now possible to define the interaction between innate and acquired immune cells at the earliest stages of infection and to define how these interactions influence local expression of immunity.

Branchett and colleagues have used in-depth transcriptomic analyses to compare the earliest response to infection between two mouse models—one where infection is contained and inflammation is regulated (C57BL/6) and one where bacterial growth is not contained and inflammation is progressively damaging (C3HeB/FeJ) (Branchett et al., 2025). Susceptibility of the C3HeB/FeJ mice is linked to an overexuberant type I IFN response (Moreira-Teixeira et al., 2020) resulting from the absence of the type I IFN regulator, Sp140 (Ji et al., 2021), while resistance in C57BL/6 mice depends upon a robust type II IFN response (Cooper et al., 1993; Pearl et al., 2001). Counterintuitively, Branchett and colleagues found that at the earliest time points following infection, the lungs of the resistant C57BL/6 contained higher bacterial numbers than the susceptible C3HeB/FeJ (Branchett et al., 2025). Unbiased transcriptional analysis showed that the C57BL/6 mice also had a more robust and rapid transcriptional response, including significant expression of type I IFN–stimulated genes, while the C3HeB/FeJ exhibited a sluggish transcriptional response relative to the C57BL/6 that was characterized by neutrophil and inflammatory myeloid signatures (Branchett et al., 2025). In a recent *JEM* article (Gern et al., 2025), using a model wherein concomitant immunity is induced by a contained Mtb infection in the ear (Nemeth et al., 2020), a similar neutrophil dominant response in C3HeB/FeJ progressed without hindrance in the primary infection, but this response plateaued in the mice with concomitant immunity. Together, these articles illustrate the critical importance of timing and balance in the early response to Mtb in the lung. Both susceptible and resistant mice express a type I IFN response and recruit neutrophils to the lung, but this response is balanced and fails to progress if countered by a strong type II IFN response in both the C57BL/6 mice and C3HeB/FeJ mice with established immunity.

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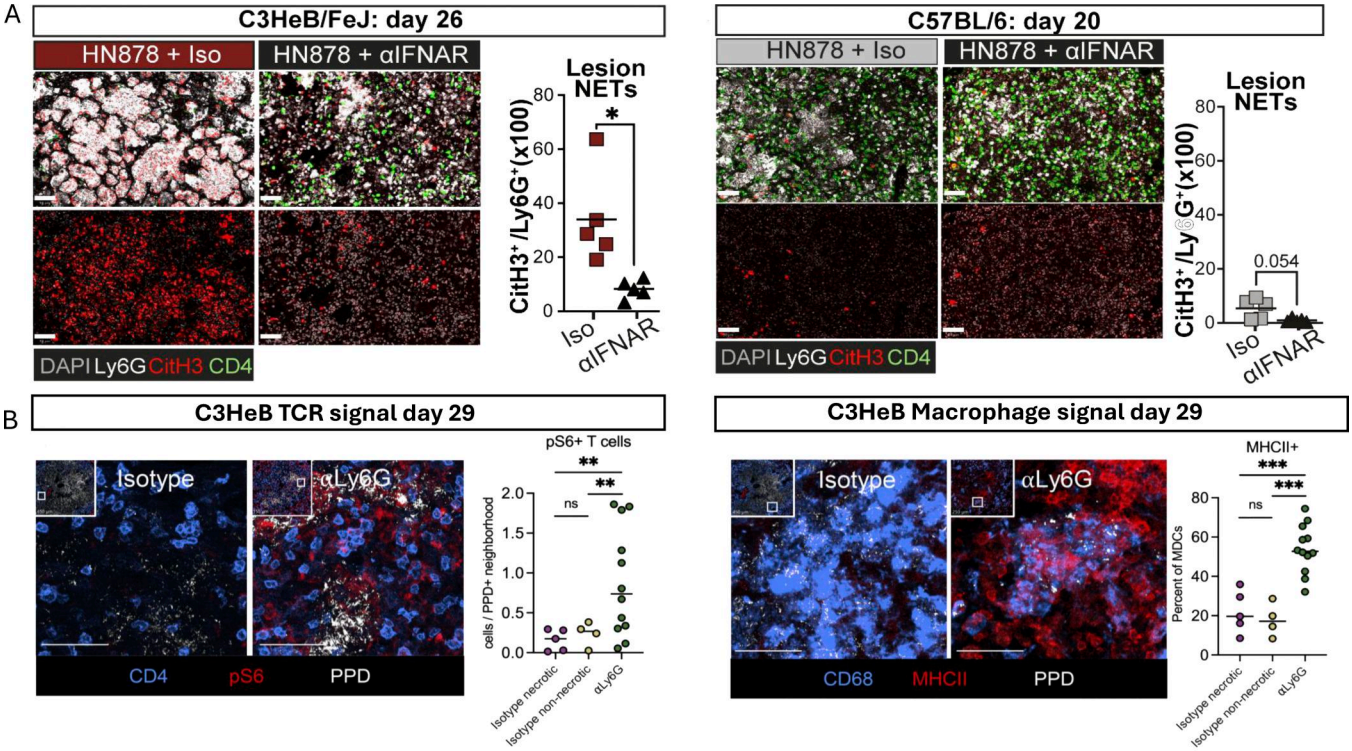

JEM | Journal of Experimental Medicine

Specific early intervention changes cell–cell interactions during TB. (A) Early type I IFN signaling promotes neutrophil swarming and limits CD4+T cell numbers in TB lesions of both relatively resistant and highly TB-susceptible mice—representative images showing reduced CitH3 (neutrophil NET [neutrophil extracellular trap] deposition) in C3HeB/FeJ (left) and C57BL/6 (right) mice with quantification of CitH3 NET staining relative to Ly6G staining in lung lesions following anti-IFNAR treatment (Branchett et al., 2025). Scale bars: 50 µm; *, P < 0.05. (B) Neutrophils limit T cell macrophage interaction—representative confocal images and quantification showing increased pS6+ T cells (TCR signaling) and increased MHCII+ in monocyte-derived macrophages (activation) following αLy6G administration to remove neutrophils (Gern et al., 2025). Scale bars: 50 µm. **P < 0.01, ***P < 0.001, and ns, P ≥ 0.05. MDC, monocyte-derived cell; PPD, Mtb antigen-bearing cells.

How are these cytokine signals balanced? Both studies used single-cell transcriptomic analysis to define the behavior of single cells and to infer cell–cell interactions and recruitment mechanisms. The outcomes from both studies support the hypothesis that CD4 T cell IFNγ production activates macrophages in the lung and that this is associated with reduced bacterial burden and development of non-necrotic lesions (Branchett et al., 2025; Gern et al., 2025). More intriguingly, the data sets provide insight into the pathways mediating the accumulation of cells. In particular, the importance of macrophage and neutrophil recruitment in the lungs of the resistant and susceptible mice highlights how the balance between a fulminant innate neutrophil response and a restrained macrophage response can define lesion progression within a single site. In both recent studies, the CXCL2–CXCR2 pathway for neutrophil recruitment was expressed early, but the ongoing and expansive expression of CXCL2 from both neutrophil and macrophage populations in C3HeB/FeJ mice was associated with continued recruitment of neutrophils at the expense of macrophages and T cells. Comparison between the lungs from the resistant and susceptible mice highlighted increased *Cxcl9*, *Cxcl10*, and *Cxcl16* recruitment of T cells in resistant mice and *Cxcl2*, *Ccl3*, and *Ccl4* recruitment of inflammatory neutrophils in susceptible mice (Branchett et al., 2025).

Although single-cell sequencing and flow cytometry allow us to assess the size and nature of responses to Mtb, they do not demonstrate the physical cell/cell interactions between cells in situ. Our understanding of how neutrophils limit immunity in the lung has been clarified in these recent studies by comparing the relative location and function of key immune cells between resistant and susceptible models using spatial analysis. In both recent *JEM* articles, neutrophil accumulation is associated with reduced accumulation of CD4 T cells and increased bacterial accumulation; this is beautifully illustrated in the lung lesion images within both papers (see figure). We know that removal of neutrophils from mice susceptible to TB is protective (Dorhoi et al., 2010; Dorhoi et al., 2014), and these new studies demonstrate for the first time that it is the very early balance between neutrophil accumulation and its effect on CD4 T cell accumulation and function that defines the inflammatory outcome of Mtb infection. Removal of CD4 T cells in the concomitant immunity model of resistance in C3HeB/FeJ mice results in development of necrotic lesions (Gern et al., 2025), and removal of neutrophils from both susceptible models allows closer association and better interaction between CD4 T cells and macrophages (Branchett et al., 2025; Gern et al., 2025) (see figure).

Animal models allow us to define the relationships between immune cell and the development of disease in complex

systems such as TB. Type I IFN and neutrophil signatures are associated with active TB (Berry et al., 2010), and the causal relationship between these elements and TB susceptibility has been demonstrated by Branchett and colleagues. Blocking of type I IFN signaling very early following infection in both C57BL/6 and C3HeB/FeJ mice resulted in reduced bacterial burdens and reduced neutrophil accumulation in the C57BL/6 mice. Interestingly, the size of the neutrophil influx was not strongly altered when type I IFN signaling was blocked in the C3HeB/FeJ mice, but there was an increase in accumulation of CD4 T cells and maturation and activation of the monocyte-derived macrophages (Branchett et al., 2025) (see panel B of figure). As inflammation progresses in the susceptible C3HeB/FeJ mice, the accumulating neutrophils differ in phenotype compared to those in the C57BL/6 lungs. The expansion of this neutrophil phenotype is limited in the absence of type I IFN signaling, supporting the important hypothesis that the inability to limit type I IFN signaling results not only in more neutrophils but a more damaging type of neutrophil. It is not just excess neutrophils, but the type of neutrophils that are in excess, that is important.

What do these studies mean for future work on TB pathogenesis and for development of interventions that control TB? How do we take advantage of our new understanding of the importance of cellular dynamics and regulation of the type I IFN signaling in mediating immunity to TB? We do not know when people are infected and so cannot deliver host-directed therapy to alter early events, and we are unlikely to be able to influence these early events by conventional vaccination. If we cannot change the initial events, then we should focus on determining how best to drive long-lived T cells that can be rapidly recruited to the lung, that can regulate type I IFN production, limit neutrophil accumulation and maturation, and persist in a lesion where neutrophils may be present. One further element that both the recent *JEM* articles highlight is that the pathways identified in these refined models may be further modulated by genetic background, indeed both genetic background and other environmental influences will impact the role of these pathways in the human responses to TB. While these articles have provided significant insight into TB pathogenesis, we still must maintain focus on TB vaccine design via collaborative efforts such as those mediated by TB Vaccine Initiative (https://tbvi.eu/), the Collaboration for TB Vaccine Development (https://www.ctvd.org/), and VALIDATE (https://validate.web.ox.ac.uk/home)—join the effort.

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References

Berry, M.P.R., et al. 2010. *Nature*. https://doi.org/10.1038/nature09247
Branchett, W.J., et al. 2025. *J. Exp. Med.* https://doi.org/10.1084/jem.20250466
Cooper, A.M. 2014. *Cold Spring Harb. Perspect. Med.* https://doi.org/10.1101/cshperspect.a018556
Cooper, A.M., et al. 1993. *J. Exp. Med.* https://doi.org/10.1084/jem.178.6.2243
Dorhoi, A., et al. 2010. *J. Exp. Med.* https://doi.org/10.1084/jem.20090067
Dorhoi, A., et al. 2014. *Eur. J. Immunol.* https://doi.org/10.1002/eji.201344219
Gern, B.H., et al. 2025. *J. Exp. Med.* https://doi.org/10.1084/jem.20250161
Gideon, H.P., et al. 2015. *PLoS Pathog.* https://doi.org/10.1371/journal.ppat.1004603
Ji, D.X., et al. 2021. *Elife*. https://doi.org/10.7554/eLife.67290
Kramnik, I., and G. Beamer. 2016. *Semin. Immunopathol.* https://doi.org/10.1007/s00281-015-0538-9
Moreira-Teixeira, L., et al. 2020. *Nat. Commun.* https://doi.org/10.1038/s41467-020-19412-6
Nemeth, J., et al. 2020. *PLoS Pathog.* https://doi.org/10.1371/journal.ppat.1008655
Pearl, J.E., et al. 2001. *Cell. Immunol.* https://doi.org/10.1006/cimm.2001.1819
Rich, A. 1944. The Pathogenesis of Tuberculosis. Charles C. Thomas, Baltimore.
WHO. 2024. Global Tuberculosis Report 2024. WHO. https://www.who.int/publications/i/item/9789240101531. (accessed September 27, 2025).