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Highlights of 2024: $\gamma\delta$ T cells and bad blood

Matthias Eberl^{1,2,*}

¹*Division of Infection and Immunity, School of Medicine, Cardiff University, Cardiff, UK;*

²*Systems Immunity Research Institute, Cardiff University, Cardiff, UK*

* **Correspondence:** eberlm@cf.ac.uk

Abstract

In this manuscript for the ‘Highlights of 2024’ series, this is me trying to summarise recent discoveries regarding the emerging regulatory and effector roles of $\gamma\delta$ T cells during sepsis, both in human patients and in mouse models. The new findings not only aid our understanding of key immunological and pathophysiological pathways that hit differently during the course of the disease but may also have relevance in the clinic.

Main text

Sepsis is a treacherous condition that is recognised as a global health priority by the World Health Organization. It causes one in five deaths worldwide, totalling around 11 million people in 2017; almost half of all estimated sepsis cases occur in children under 5 years of age.¹ Call it what you want – historically often referred to as ‘blood poisoning’ or ‘septicaemia’, *i.e.* the presence of micro-organisms in the blood – the definition of sepsis has been agreed to mean a dysfunctional host response to infection that culminates in life-threatening organ failure.^{2,3} That’s when individuals with suspected sepsis require swift intervention and tailored patient management. Yet, despite the undoubted clinical need, timely diagnosis of sepsis and the initiation of targeted therapy the moment you know it is sepsis remains unsatisfactory, in part because the labyrinth between intersecting pro-inflammatory and anti-inflammatory pathways combined with the modulatory effects of medication such as vasopressors, antibiotics and corticosteroids and other interventions makes it difficult to understand, and exploit, the immunological basis of the condition.⁴

While the reputation of unconventional T cells as immunological players in infectious disease is forever and always overlooked, several significant discoveries regarding the emerging regulatory and effector roles of $\gamma\delta$ T cells during sepsis were reported in 2024. $\gamma\delta$ T cells are set apart from classical $CD4^+$ and $CD8^+$ T cells in that they are typically not restricted by antigenic peptides presented by molecules of the major histocompatibility complex, with the $\gamma\delta$ T cell receptor itself being the only permanent marker of $\gamma\delta$ T cells (as already expertly reviewed in 1989 by Tonegawa *et al.*). The novel and intriguing findings, both in human sepsis patients^{5,6,7} and in mouse models of sepsis,^{8,9} not only provide clean insight into key immunological pathways in the disease process but they may also have diagnostic and prognostic relevance as well as implications for new interventions, so it goes.

$\gamma\delta$ T cells as indicators of the infecting organism

Gram-negative organisms are associated with poorer outcomes in sepsis patients with first-hit infections.¹⁰ As a result, predicting the causative organism at the point of admission, long before the availability of microbiological culture results, is of prime importance for early risk stratification and the commencement of targeted therapy. Using machine learning to interrogate a comprehensive set of clinical parameters, plasma biomarkers and flow cytometry data of blood immune cells, Burton *et al.* were able to define immune signatures that predicted the 90-day mortality of patients with severe sepsis, as well as identified the underlying cause of the infection.⁵ Of particular relevance, frequencies of circulating V δ 2⁺ $\gamma\delta$ T cells had a profound impact on the prediction of Gram-negative infections, alongside other variables including the total neutrophil count in blood. Although this points to a possible host response to microorganisms capable of producing the isoprenoid intermediate (*E*)-4-hydroxy-3-methyl-but-2-enyl pyrophosphate (HMB-PP), a highly potent activator V δ 2⁺ T cells produced by most Gram-negative bacteria but not by sepsis-causing Gram-positive bacteria such as streptococci and staphylococci, the situation may be evermore so complex.

Of note, other investigations did not find such clear-cut differences depending on the causative organism. For instance, Grimaldi *et al.* reported reduced levels of circulating $\gamma\delta$ T cells in sepsis patients compared with healthy controls, and saw a difference in levels of mucosal-associated invariant T (MAIT) cells but not $\gamma\delta$ T cells between patients with streptococcal and patients with non-streptococcal infections.¹¹ Similarly, some studies found that mortality in sepsis patients was associated with a decrease in blood $\gamma\delta$ T cells^{12,13} but found no differences in lymphocyte subsets in patients with Gram-positive versus Gram-negative infections.¹² Taken together, these observations identify $\gamma\delta$ T cells as potent responders and potential effectors in acute sepsis, yet more needs to be done to dissect their exact contribution.

Organism-specific expansion of foetal-derived $\gamma\delta$ T cells in neonatal sepsis

Over 22 years after the description of HMB-PP as the one most potent microbial activator of human V δ 2⁺ T cells,¹⁴ our understanding of $\gamma\delta$ T cell responses to different micro-organisms is far from closure. In order to investigate $\gamma\delta$ T cell responses during acute bacterial infection, Giannoni and Sanchez Sanchez *et al.* recruited children with blood-culture proven sepsis, focusing on microbiologically confirmed cases of infections by the HMB-PP producing Gram-negative bacterium *Escherichia coli*, and by the HMB-PP-deficient Gram-positive bacteria *Staphylococcus aureus* and *Streptococcus pneumoniae*.⁶ Unexpectedly, the authors found that infection not only with *E. coli*, but even more so also with *S. aureus*, led to a selective expansion of a foetal TRDV2 repertoire in children younger than two years of age but not in older children or adults (**Figure 1**). In contrast, they did not detect any such effect of *S. pneumoniae* on the TRDV2 repertoire at any age. While the lack of responsiveness to *S. pneumoniae* might be anticipated given the absence of HMB-PP in streptococci, the striking response to HMB-PP-deficient *S. aureus* is all too well puzzling and may point to either the existence of additional, and so far, invisible bacterial metabolites with activity on V δ 2⁺ T cells, or to an indirect activation via superantigens or soluble factors. In this regard, the findings by Giannoni and Sanchez Sanchez *et al.* nicely complement previous reports on the high infidelity of human V δ 2⁺ T cell responses to *S. aureus* even in the absence of HMB-PP.^{15,16}

An immunoregulatory role of foetal-derived $\gamma\delta$ T cells in neonatal sepsis

A clue for the potential role of foetal-derived V δ 2⁺ $\gamma\delta$ T cells in early life comes from the investigation by León-Lara *et al.* who identified a population of activated HLA-DR^{hi} CD83⁺ $\gamma\delta$ T cells that was preferentially expanded in neonates with sepsis.⁷ Functional assays revealed that *in vitro* stimulation for seven days with HMB-PP did induce expression of CD83 on V δ 2⁺ T cells in preterm and term neonates but not on V δ 2⁺ T cells from adults. In contrast, only V δ 2⁺

T cells from adults but not from preterm or term babies upregulated expression of the co-stimulatory molecule CD86 in the presence of HMB-PP, and induced proliferation of allogeneic CD4⁺ T cells. These findings may point toward a regulatory role of neonatal Vδ2⁺ T cells as they may be unable to differentiate into fully functional antigen-presenting cells and shape long-lived adaptive immunity,¹⁷ unlike their adult counterparts that readily give rise to distinct CD4⁺ T cell effector subsets.¹⁸ Taken together, the findings by both Giannoni and Sanchez Sanchez *et al.* and León-Lara *et al.* identify unique responses by foetal-derived γδ T cells that only the young display and that are lost at an older age, thereby extending previous reports on neonatal sepsis within a fortnight after birth.¹⁹ Further studies will require functional experiments using antigen-presenting Vδ2⁺ T cells from infected individuals,²⁰ with preliminary findings already indicating that Vδ2⁺ T cells from adult sepsis patients may actually be less equipped to drive CD4⁺ T cell responses compared to Vδ2⁺ T cells from healthy donors.²¹

Organism-specific γδ T cell responses in mouse models

Of note, the delicate responsiveness of γδ T cells to distinct microbial organisms as demonstrated by Burton *et al.* and by Giannoni and Sanchez Sanchez *et al.* is replicated in mouse models, even though the identity of the actual microbial antigens there remains a blank space (murine γδ T cells are unable to respond to HMB-PP). Witt *et al.* showed elegantly how *Streptococcus agalactiae* and *Escherichia coli* induce distinct effector γδ T cell responses during neonatal sepsis induced upon intraperitoneal injection of bacteria into C57BL/6 pups.⁸ γδ T cells rapidly responded both to *S. agalactiae* and *E. coli* bloodstream infections, yet gave rise to markedly distinct immune responses within 18 hours of infection (**Figure 1**). *E. coli* induced IL-17 producing γδ T cells, in a TCR-dependent manner; in contrast, *S. agalactiae* led to production of IFN-γ by γδ T cells, independently of TCR signalling. Moreover, while γδ T cells could be found in the brain of neonatal mice infected with either organism, the quality of

the ensuing neuroinflammation was markedly different, with only *E. coli* leading to a pronounced infiltration of monocytes and neutrophils to the brain. The underlying molecular mechanisms and antigens remain to be defined, especially with regard to the functional dichotomy between IFN- γ and IL-17 producing $\gamma\delta$ T cells in infection.²²

$\gamma\delta$ T cells as modulators of lung injury

The detrimental role of IL-17 producing $\gamma\delta$ T cells exacerbating the inflammatory response to neonatal *E. coli* infection shown by Witt *et al.* was in agreement with the observation by Xie *et al.* of a sepsis-induced migration of IL-17⁺ $\gamma\delta$ T cells from the small intestine to the lung.⁹ A substantial proportion of patients with sepsis develop acute respiratory distress syndrome characterised by difficulty to breathe, fluid build-up and lung injury. Using a model of polymicrobial sepsis induced by caecal ligation and puncture (CLP) in C57BL/6 mice, Xie *et al.* proposed that targeting of pulmonary IL-17 might in fact represent a promising adjunct therapy against acute lung injury in sepsis and demonstrated that the drug esketamine, the (*S*)-enantiomer of ketamine, downmodulated pulmonary Wnt/ β -catenin signalling, thereby inhibiting the migration of small intestinal $\gamma\delta$ T cells into the lung. Whether such a treatment may also affect the $\gamma\delta$ T cell-driven neuroinflammation studied by Witt *et al.* is currently unclear. Moreover, the exact role of IL-17⁺ $\gamma\delta$ T cells in the lung of septic mice deserves closer inspection, with conflicting data from similar CLP mouse studies where IL-17⁺ $\gamma\delta$ T cells infiltrating the lung and liver in fact exerted a protective role.^{23,24}

Outlook

Long story short, these $\gamma\delta$ highlights of 2024 represent a further step toward understanding the pathophysiology of sepsis, and to potentially exploiting this knowledge in the clinic. However, as throughout all eras in science, each answer immediately poses a series of new questions.

What are the microbial antigens recognised by $\gamma\delta$ T cells, other than HMB-PP? Where do $\gamma\delta$ T cells get activated and where do they migrate upon activation? Do they proliferate outside secondary lymphoid tissues, at the site of inflammation? What is the role of antigen-presenting $\gamma\delta$ T cells in infection? How do $\gamma\delta$ T cell responses towards microbes and other insults change from birth to adulthood? Under which circumstances are $\gamma\delta$ T cells protective, and when do they exacerbate the situation or help tolerate it? Are they innocent bystanders or powerful masterminds? To what extent do mouse models replicate the situation in human patients? And possibly most importantly, dear reader – how can we standardise experimental protocols and define clinical cohorts better so that findings are reproducible across multiple studies and become timeless and untouchable?

If we are ready for it and can address all these points in style, the $\gamma\delta$ T cell field will be more than just a welcome distraction and can in our wildest dreams have real impact in patients with severe infections. Wouldn't it be a wonderful thing if we were able to state that everything has changed based on this research of ours? This is what we came for.

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Conflict of interest

I declare no conflicts of interest.

Author contributions

ME! Conceptualisation; writing – original draft; writing – review and editing.

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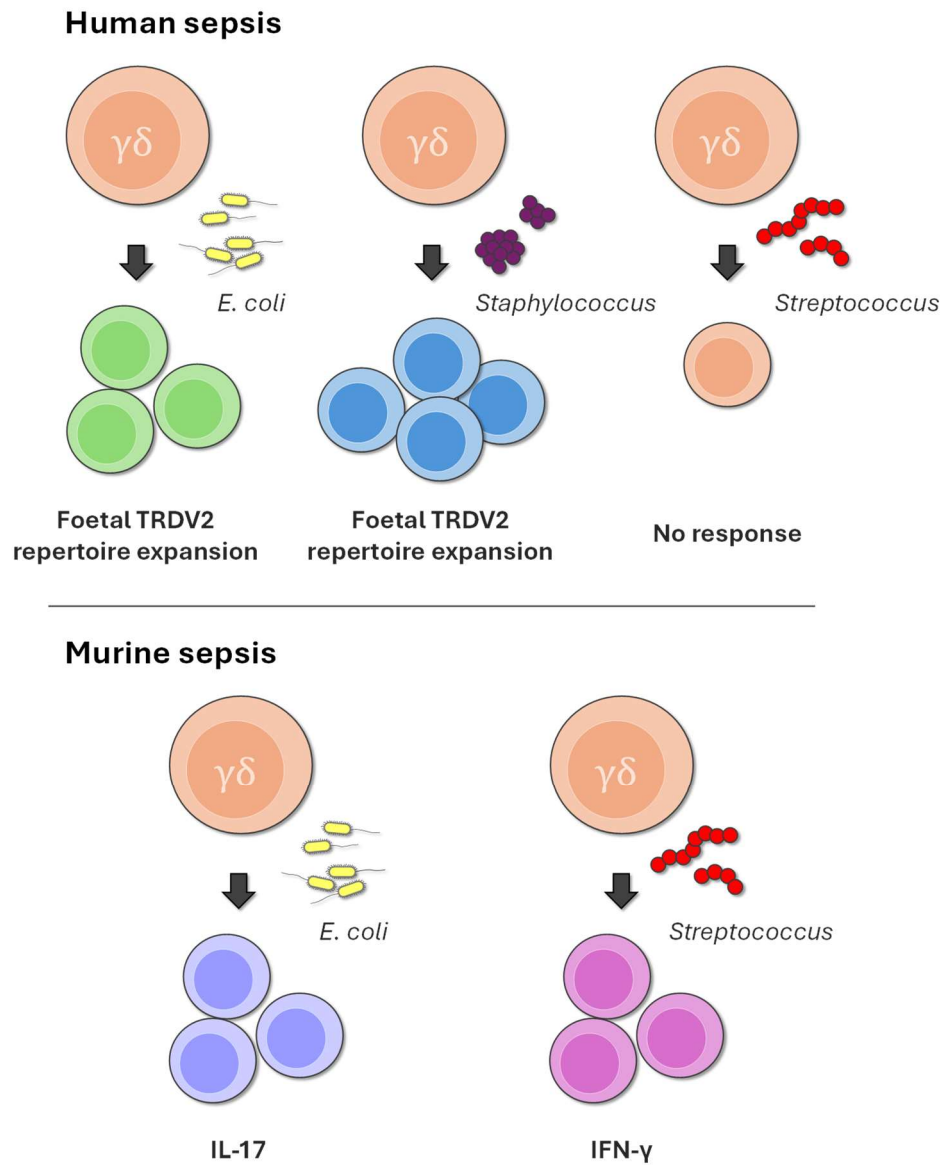


Figure 1. Depiction of $\gamma\delta$ T cell responses in neonatal sepsis (tailored version according to the Highlights of 2024 papers featured in this article).⁵⁻⁹