Published in final edited form as:

Trends Immunol. 2011 February ; 32(2): 66–72. doi:10.1016/j.it.2010.12.001.

The cellular response to mycobacteria: balancing protection and pathology

Egídio Torrado, Richard T. Robinson, and Andrea M. Cooper

Trudeau Institute, Inc., 154 Algonquin Ave., Saranac Lake, NY 12983, USA.

Abstract

There has been a recent increase in our understanding of T cell responses during mycobacterial infection; however we have not yet identified the protective mechanisms capable of mediating vaccine-induced protection in the lung. Novel approaches have allowed the determination of the kinetics and location of naïve T cell activation, the factors impacting expansion of antigen-specific T cell responses, and the factors impacting the balance between protective and immunopathological consequences during the chronic stages of infection. With new and more efficient vaccination strategies urgently needed, the integration of these data will result in improved vaccine strategies.

T cells: a central component of immunity to mycobacteria

The outcome of mycobacterial infections is dependent on the interaction between the bacteria and the host's innate and acquired immune system. During *Mycobacterium tuberculosis* (Mtb) infection, these interactions can lead to pathogen control, establishment of latent infection or progressive disease [1]. Recent use of the zebrafish model infected with *Mycobacterium marinum*, suggests that innate responses can contribute to the control of mycobacterial growth [2] however despite this, T cells are activated when a person is exposed to Mtb and are an essential component of the protective response [3,4]. There are several important questions with regard to T cell activation and function in mycobacterial disease. The first is the role of the bacteria in limiting the early activation and accumulation of T cells into the infected site. In addition, the chronic nature of mycobacterial infection results in constant activation of T cells and recent studies have addressed the consequences of chronic stimulation in terms of both T cell biology and with regard to control of immunopathological consequences. As new vaccines are developed and enter clinical trials [5], it is critical to understand the mechanisms of T cell activation and the factors regulating maintenance of the cellular response at the infection site. To help in this regard, we discuss advances in our understanding of T cell responses during mycobacterial infection; with particular focus on the mouse model of TB. We discuss how T cell responses are initiated and maintained during infection, what immunoregulatory pathways slow these responses and the immunopathological consequences of disrupting the balance between regulatory and effector responses in the infection site.

© 2010 Elsevier Ltd. All rights reserved.

Corresponding author: acooper@trudeauinstitute.org, Tel: 518-891-3080, Fax: 518-891-5126.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

T cell priming is slow to occur during *M. tuberculosis* infections

Following aerosol infection with Mtb, bacilli are deposited in the lower airways of the lung. The bacteria grow in an apparent uncontrolled manner for the first 21 days, at which point growth is controlled by the acquired immune response [3,4]. IFN γ -producing T cells, which are required to activate phagocytic host cells, are first detected in the lung at day 14 post-infection, but accumulation in sufficient numbers to limit Mtb proliferation does not occur until around day 20. During systemic infections, the accumulation of T cells in the lung is faster [4]. The basis for the slow accumulation of T cells in the lung during aerosol infections is as yet unclear.

Using T cell receptor (TCR) transgenic (TCRTg) mice that express a TCR specific for Mtb antigen, different groups have demonstrated that CD4 T cell priming occurs in the draining lymph nodes (dLN) of the lung 7 to 10 days post aerosol infection [6–8]. Specifically, expression of phenotypic markers associated with the first steps of T cell priming, correlate with the temporal arrival of bacteria into the dLN, and only after this do TCRTg CD4 T cells begin to proliferate and migrate to the lung [6,7]. These data suggest that the optimal conditions for T cell activation are unique to the dLN and do not occur in the lung (figure 1). Indeed, when different APC populations were isolated from lungs of Mtb-infected mice and used to stimulate naïve antigen-specific CD4 T cells, they were poor inducers of cytokine production by these T cells, when compared to the same populations isolated from the dLN [9]. Interestingly, dLN APCs that did not harbor Mtb bacilli were the most efficient at inducing cytokine production by CD4 T cells [9], suggesting that Mtb-infected phagocytes from the lungs are not efficient at priming naïve T cells.

Because Mtb grows exponentially in the lung from day 3 post-infection [10], it is still not clear why T cells see antigen only after day 8 post-infection. It is possible that Mtb is phagocytosed by a population of cells that are unable to migrate to the dLN or alternatively, the infection itself is inhibiting this migration. To examine these hypotheses, inflammation in the lung environment has been induced by delivering LPS to infected animals; however this did not accelerate T cell priming [8]. On the other hand, increasing the infection dose did enhance the response by 1–2 days [7]. These data suggest that bacteria are initially phagocytosed by non-motile cells rather than that they actively inhibit the migration of the infected cell to the dLN (figure 1).

Together these data suggest that T cell responses are initiated slowly during TB as a consequence of the limited antigen availability and ability of the infected cells to move to the dLN. Furthermore, central memory immune cells circulating through the dLN will also fail to see antigen until 8 days following infection. In contrast, if a new vaccine could generate effector memory cells that persist in the lung tissue, these cells could respond to infection as soon as it occurs and thereby limit bacterial growth sooner.

Modulation of the microenvironment by mycobacteria: does it affect T cell priming?

The slow kinetics of T cell activation might be a key element in the ability of Mtb to establish infection. Modulation of the environment by actively growing bacteria, both at the site of infection and at the site of T cell priming is likely to play a role in both the slow induction and expression of immunity.

Mtb appears to be adept at modulating phagocyte function in the absence of acquired immunity, with virulent strains of Mtb able to prevent initial elimination by inhibiting both maturation of the phagolysosome and apoptosis within the host phagocyte [11–13]. Mtb

inhibits apoptosis and promotes necrotic death of infected macrophages by stimulating the production of lipoxins (LO), such as lipoxin A₄ (LXA₄), that inhibit the production of prostaglandin E₂ which is required for protection of the mitochondrial inner membrane (figure 1) [14–16]. This is a key activity for increasing survival of the bacteria and recent work has shown that inhibition of apoptosis might also impact T cell activation [17]. Specifically, infected macrophages from mice deficient in 5-lipoxygenase, which cannot synthesize LXA₄, undergo more apoptosis, and when transferred into wild-type mice, induce a greater accumulation of Mtb-specific CD4 and CD8 T cells in the lung than do wild-type macrophages [17]. While these pro-apoptotic macrophages are as efficient as wild-type macrophages at stimulating TCRTg OT-I cells when pulsed with peptide alone, the enhanced T cell response after Mtb infection resulted from DC uptake and cross-presentation of the antigen cargo contained in the apoptotic vesicles (figure 1) [17]. Indeed, when caspase activation was pharmacologically blocked and apoptotic death reduced, there was a significant reduction in the frequency of antigen-specific CD8 T cells in the lung [17]. Taken together with the inability of bacteria to arrive in the dLN before day 8, one could speculate that it is the destruction of the infected non-motile cells that provides the antigen cargo for the motile DCs. That the nature of this destruction and the subsequent ability of these motile cells to stimulate naïve T cells are actively modulated by the bacteria is testament to the pernicious nature of the interaction between host and this pathogen (figure 1). The nature of the cells that are first infected and the reason for their lack of motility should be determined.

T cell priming and accumulation in the infected site might also be delayed through the induction of regulatory T cells (Tregs). The role of Tregs has been analyzed in Mtb-infected humans and during experimental infections [18–20]. Tregs expand in the lung during Mtb infection in mice, and upon their deletion, the bacterial burden is reduced at early stages of infection [18] but not during chronic infection [21]. Neutralization of Tregs increases the frequency of IFN γ -producing T cells in the lung [18,21], suggesting that these cells are capable of impairing the protective immune response to Mtb. These studies, however, were done in polyclonal cell populations, and until recently it has been unclear whether Mtb-specific Tregs develop during infection. With the use of transfer models and Mtb-specific TCRTg CD4 T cells, pathogen-specific Tregs were shown to expand from a population of pre-existing foxp3⁺ Tregs in the dLN of the lung; however, Mtb-specific Tregs had a higher propensity to proliferate when compared to effector T cells [22]. Importantly, these Tregs develop only after bacteria are transported to the dLN, suggesting that both effector and regulatory responses are initiated at the same time (figure 1). When Mtb-specific Tregs were transferred into wild-type mice, the number of Mtb-specific T cells in dLN was reduced, suggesting that specific Tregs reduce T cell responses by delaying either priming or proliferation of newly activated cells [22]. Moreover, accumulation of Mtb-specific T cells was also delayed in the lung, resulting in higher bacterial burdens [22]. While the mechanism by which Tregs delay T cell priming during TB is not understood, previous work suggest that the interaction of Tregs with DCs can result in the down regulation of co-stimulatory molecules in DCs, impairing their capacity to prime naïve T cells [23,24]. In addition, Tregs can make IL-10 which can delay accumulation of effector T cells in the lung during Mtb infection [25]. IL-10-deficient mice express higher levels of CXCR3 binding chemokines [25], it is possible therefore that the improved accumulation of antigen-specific T cells in the lungs of the mice with reduced Tregs results from reduced chemokine expression as well as reduced expansion of activated effectors (figure 1).

Together the bacteria and the host both appear to actively regulate the early immune response to infection with the bacteria delaying the response by subverting the activation of sufficient T cells while the host generates Tregs and IL-10 production.

Unconventional T cell subsets during mycobacterial infection

Unconventional T cells vary greatly between mouse and humans, and hence their role is not well understood in TB. $\gamma\delta$ T cells, for instance, are important during granuloma development upon BCG and Mtb infection [26,27], possibly through IL-17 production (Box 1 and figure 2) [28]. Interestingly, IL-17-deficient mice have enhanced susceptibility to high dose intratracheal Mtb infection [27] but not low dose aerosol infection [29,30], suggesting that $\gamma\delta$ T cell-derived IL-17 may be essential for optimal protection during high dose challenge. Indeed, mice that are deficient in the liver X receptor (LXR), which is involved in lipid homeostasis and inflammation, appear to be susceptible to high dose intratracheal infection and this is linked to deficiency in the early neutrophil IL-17 response [31]. It would be interesting to know whether this LXR-dependent IL-17 response required $\gamma\delta$ T cells. Further research is required to define the impact of infection dose and route on the role of both IL-17 and $\gamma\delta$ T cell responses in early protection.

The recently identified mucosally associated invariant T (MAIT) lymphocytes (see box 1) respond to a presenting molecule that is monomorphic and which is presented by professional APCs and infected epithelial cells [32]. In humans, MAIT cells are enriched in the lungs of healthy or latently infected individuals and are reduced in the blood of patients with active TB [32,33] while mice infected with *Mycobacterium abscessus* are more susceptible in the absence of MAIT cells [33]. These data suggest that MAIT cells could have an important role in cell-mediated immune responses to mycobacterial infection and may be a unique target for vaccination or immune intervention.

Since the mycobacterial cell wall is lipid-rich, it is not surprising that CD1-restricted T cells are among the unconventional T cell subsets that respond to infection. However, the majority of CD1-restricted T cells do not exist in mice and thus definitive answers about the role of these cells during mycobacterial infection remain elusive (Box 1). Recently, the use of a transgenic mouse model expressing the group 1 CD1 genes CD1a, CD1b and CD1c (CD1Tg) under their endogenous human promoters has been used to study the role of group 1 CD1-restricted T cells during TB [34]. Group 1 CD1 molecule expression in CD1Tg mice was found to be similar to the pattern observed in humans, and APCs isolated from these mice were able to activate human group 1 CD1-restricted cell lines specific to Mtb lipid antigens. CD1-restricted T cell clones generated upon immunization with Mtb antigens responded to group 1 CD1+ APCs by producing IFN γ and lysing group 1 CD1+ targets. Some of the generated cell lines responded to Mtb lipids while the majority reacted in the absence of endogenous antigen, suggesting that recognition of self-lipids is a property of CD1-restricted T cells. Following aerosol Mtb infection, lipid antigen-specific T cell responses were observed 3 and 4 weeks after challenge, and this response was abrogated in the presence of anti-group 1 CD1 monoclonal antibodies [34]. An interesting feature of these group 1 CD1-restricted T cells was the ability to respond more rapidly and robustly upon secondary antigen challenge [34], a feature of acquired immunity and not observed in group 2 NKT cells. Indeed, NKT cells are activated during mycobacterial infection and probably exert their effector function before MHC-restricted T cells (Box 1); however, they rapidly contract and become unresponsive to TCR stimulation [35]. Nevertheless, pharmacological activation of NKT cells with a single treatment of α galactosylceramide significantly prolongs survival of Mtb-infected mice [36–38], suggesting a potential protective role for these cells if effectively manipulated.

These data suggest that antigen-specific CD1-restricted T cells are induced during infection and could contribute to the protective immune response. Unlike the MHC, CD1 molecules exhibit low levels of polymorphism in their antigen-binding domain, making them attractive

as vaccine components that would be recognized widely in a genetically diverse human population.

Overall it appears that several distinct populations of unconventional T cell populations are primed during mycobacterial infection. The capacity of these cells to directly recognize infection and, in some cases, to respond more rapidly during a secondary response makes them a valuable subject of study that will contribute to rational vaccine design. Moreover, since some of the unconventional T cell populations can lyse infected cells they could act to enhance conventional T cell priming by promoting phagocytosis of bacilli by newly recruited APCs (figure 1).

Maintenance of T cell responses during chronic mycobacterial infection

Sustained T cell responses are essential to keep mycobacterial infection at check; however data from different chronic infection models suggest that, in settings of chronic antigen stimulation, T cells can undergo exhaustion [39,40]. The question of how effector cell populations are maintained in TB has been the subject of recent study. From other fields it has been shown that expression of specific surface markers indicates functional exhaustion and terminal differentiation of T cells in chronic disease states. During Lymphocytic Choriomeningitis Virus (LCMV) infection, CD8 T cells that progress through a substantial number of cell divisions express Killer cell Lectin-like Receptor subfamily G member 1 (KLRG1) on their surface [40]. In addition, expression of Programmed Death-1 (PD-1) is also selectively up regulated by exhausted CD8 T cells and blockade of the PD-1 ligand 1 increases the number and frequency of antigen-specific CD8 T cells to LCMV [39]. Thus, KLRG1 and PD-1 expression are associated with proliferative senescence and terminally differentiated cells.

During experimental Mtb infections, different populations of CD4 T cells express PD-1 and KLRG1 [41]. Both KLRG1 and PD-1 T cell subsets proliferate and produce cytokines; however, PD-1+ CD4 T cells have a higher proliferative potential and lower capacity to produce cytokines when compared to KLRG1+ CD4 T cells that have lower proliferative capacity but are more efficient at producing cytokines (figure 2) [41]. In transfer studies, PD-1+ CD4 T can become KLRG1+, whereas KLRG1+ CD4 T cells do not change their phenotype and fail to persist in Mtb-infected animals [41]. These data show that PD-1 and KLRG1 expression on CD4 T cells during TB is not associated with exhaustion or terminally differentiation, but rather reflects functionally distinct subsets of effector CD4 T cells. Interestingly, PD-1 deficient mice are highly susceptible to Mtb infection, developing extensive CD4 T cell responses and pathology [42,43]. Furthermore, PD-1 deficiency in CD4 T cells is sufficient to recapitulate the phenotype of PD-1 deficient mice [43] demonstrating that the PD-1 pathway is critical for regulating the pathological response of CD4 T cells during Mtb infection.

Since antigen-specific effector T cell numbers remain relatively stable during the chronic phase of Mtb infection, these data suggest that either effector T cells have an unlimited proliferation capacity or the effector T cell pool must be maintained by a source of new effector T cells. It was suggested that the PD-1+ cells could represent a self-replicating pool within the antigen-specific population that seeds the more differentiated KLRG1+ population [41]. A second possibility is that new thymic emigrants serve to replenish the effector T cell pool (figure 2). However, it has been shown that the frequency and proliferative potential of CD4 T cells and antigen-specific CD4 T cells during Mtb infection in both normal and adult-thymectomized mice is very similar [44], suggesting that the thymus is not an essential requirement for the maintenance of the effector cell population at least in the early phase of the disease. It is possible that thymic emigrants might only be

required to replenish peripheral effector T cells late during chronic infection; more research is required to clarify this subject.

The question of new thymic emigrants entering the response is important because during *Mycobacterium avium* or Mtb infections the thymus can become infected [45,46]. In a very interesting model performed to address the protective role of T cells that developed after thymic infection, the thymic lobes from *M. avium*-infected TCR α -deficient mice were transplanted under the kidney capsule of athymic nude mice [45]. In this situation only T cells generated after *M. avium* infection of the thymus could enter the response in the periphery. It was shown that infection and growth of *M. avium* in the thymus compromises the T cell response to mycobacterial antigens as T cells in the periphery are impaired in their ability to produce IFN γ in response to *M. avium* antigens. This response was specific for the infecting pathogens, as Ag85-specific T cell, but not OVA-specific T cell, numbers were reduced upon immunization with the cognate antigen in adjuvant [45]. Thus it appears that mycobacterial infection of the thymus has the capacity to specifically compromise the antigen response in the periphery. While it is still not clear whether new thymic emigrants are essential to the response during mycobacterial infections, these data are clearly relevant to situations where new thymic emigrants are required to replenish T cell pools in the periphery, such as during recovery from extensive lymphopenia after HIV infection.

Regulation of T cell responses during chronic mycobacterial infection: implications for immune-mediated pathology

Despite robust innate and adaptive immune responses, bacterial persistence in the host is a key feature of mycobacterial infections. The chronic nature of mycobacterial infections requires constant expression of cellular immunity to maintain mycobacterial proliferation at check. On the other hand, strong cellular responses can have immunopathological consequences with implications for the survival of the host and transmission of Mtb. Specifically, in susceptible individuals the granuloma develops to become necrotic, allowing Mtb to be spread upon coughing. While the mechanisms of the damage are not well understood, mycobacterial infections are excellent models to study the balance between protective immunity and immune-mediated pathology, because this balance is maintained for prolonged periods of time.

The cytokine IFN γ is central for macrophage recruitment, activation and control of mycobacterial proliferation. In the absence of IFN γ , or IFN γ -signaling, both mice and humans are extremely susceptible to mycobacteria [1,47]. This susceptibility is not only associated with low phagocyte activation but also with the elevated accumulation of granulocytes in the infection foci [10]. Indeed, both *M. avium* and *M. leprae*-infected IFN γ -deficient mice show very mild increase in bacterial numbers but they have robust granulocyte recruitment to the infection foci [48,49]. Recent studies using chimeric mice wherein nonhematopoietic cells are unable to respond to IFN γ have dissected the function of this cytokine in regulating granulocyte accumulation in response to mycobacteria [50]. Specifically, infection of these mice with Mtb results in extensive immunopathological consequences in the lung that are associated with elevated Th17 responses and an inability of epithelial and endothelial cells to express Indoleamine 2,3-dioxygenase and generate tryptophan catabolites [50]. It was suggested by in vitro data that under normal conditions the Th17 cells was restrained by tryptophan metabolite production. Further, it seemed that in the absence of IFN γ signaling there was an increase in IL-23 at the inflamed site and that this aided the survival and effector function of the Th17 cells [50].

The impact of local IL-23 production on immunopathological consequences was highlighted again in recent work. By revaccination of mice with BCG following Mtb challenge the

IL-17 response was enhanced without a change in the protective response [51]. This IL-17 response was associated with higher local expression of IL-23 expression and was absent in IL-23-deficient mice [51]. That the increase in inflammation and granulocyte influx in this model was dependent upon IL-17 was shown by loss of these responses during anti-IL-17 treatment [51]. These data suggest that high antigen challenge in Mtb-infected mice can result in expansion of the ongoing IL-17 response that results in enhanced tissue damage. It would be of interest to determine whether the enhanced IL-23- and IL-17-dependent pathology requires the LXR pathway as was described above for the early IL-17 response in the high dose intratracheal model [31]. Both the revaccination study and the IFN γ study support the contention that a balance between Th1 and Th17 responses needs to be maintained during chronic mycobacterial infection and that disrupting this fine balance has important immunopathological consequences (figure 2).

The role of IL-27 in the balance between pathology and protection is interesting. In the absence of IL-27 there is elevated mononuclear inflammation during TB and better ability to control bacterial growth [52,53]. IL-27 induces T-bet expression from CD4 T cells, therefore enhancing IFN γ production on a per cell basis [52,54]. This is relevant since high levels of IFN γ can be detrimental for T cell survival during mycobacterial infections by altering the susceptibility to apoptotic signals [55]. On the other hand, IL-27 induces IL-10 from activated CD4 T cells such that in its absence less IL-10 may be present in the effector site [56]. It is possible that these apparently antagonist effects of IL-27 on T cells are dependent on the activation state of the cell, thus IL-27 may act first to induce T-bet and IFN γ secretion and then act by enhancing IL-10 to protect the cell from the damaging effects of IFN γ . Further research is required to determine the role of IL-27, IL-23 and IL-12 in the balance of protective and pathologic responses and their effect on the progressive development of T cells during mycobacterial infections (figure 2).

Concluding remarks

T cells are a critical component of the protective immune response to mycobacteria however, their priming is relatively slow and this allows bacteria to grow unrestrainedly and induce inflammation in the absence of the modulating effect of the acquired response. It is important to future rational vaccine design that we dissect the mechanisms that prevent Mtb antigens from being efficiently presented to T cells, as this is an essential step for alerting circulating memory cells to accumulate in the lung and thereby limit Mtb growth. Furthermore, understanding what impacts the maintenance of T cell responses at the site of infection and the balance between different T cell subsets is essential to our ability to vaccinate in such a way as to maintain effective immune protection with minimal pathologic consequences for the host. This balance needs to be taken in consideration for vaccine design, as a vast percentage of the human population has come in contact with Mtb. Disrupting this balance by inappropriate vaccination can have tremendous consequences for host survival and pathogen transmission.

General features of unconventional T cell populations that respond to mycobacteria

Unconventional T cells that respond to Mtb include CD1-restricted T cells, $\gamma\delta$ T cells and MAIT cells. Unlike MHC, CD1 molecules bind self and foreign lipid antigens that are surveyed by CD1-restricted T cell populations expressing $\gamma\delta$ or $\alpha\beta$ TCRs [57]. Group II CD1-restricted T cells (CD1d) are the only population present in mice, which lack the genes encoding group I CD1 (CD1a, CD1b and CD1c). In humans, group I CD1-restricted T cells recognize a variety of lipid antigens from mycobacteria [58] and have a broad range of effector functions, including IFN γ secretion and cytotoxicity towards CD1+ targets [57,58]. CD1-mediated lipid presentation is likely to be more rapid than

MHC-mediated peptide presentation. Indeed, MHC molecules traffic from the phagosome to the cell surface with rapid kinetics that can precede antigen availability and loading whereas CD1 molecules continually recycle between these sites, independently of the maturation status of the APC [59]. This might lead to an earlier recognition of the mycobacteria-infected APC by CD1-restricted T cells. Unlike group I CD1-restricted T cells, the array of microbial lipids presented to NKT cells in the context of CD1d is limited. To date, phosphatidylinositol mannoside (PIM) is the only mycobacterial-derived antigen identified that can be recognized in a CD1d-restricted fashion [60]. Recognition of PIM induces IFN γ secretion and cytotoxicity by splenic NKT cells [60]. $\gamma\delta$ T cells are an important source of IL-17 and IFN- γ during mycobacterial infection; however, their role seems to vary depending on the mycobacterial strain and route/dose of infection used. In the high dose intratracheal BCG model, $\gamma\delta$ T cells produce IL-17 that is required for early formation of the granuloma and in the absence of $\gamma\delta$ T cells or IL-17, mice are more susceptible to this route of infection [27]. On the other hand, upon low dose aerosol Mtb infection $\gamma\delta$ T cell-deficient mice have elevated accumulation of neutrophils in the lung, with minor changes in bacterial burden [26]. MAIT cells recognize a variety of bacterial components via a monomorphic MHC class Ib molecule (MR1) in a very rapid manner [32,33,61]. They are expanded from a naïve population following birth in response to gut flora [32,33,61]. These cells, although never exposed to Mtb, respond rapidly to delipidated cell wall preparations of mycobacteria [32,33].

Acknowledgments

The Trudeau Institute, Inc., NIH grants AI67723, AI69121 and AI46530 and an American Lung Association De Souza award, supports our work.

References

1. Casanova J, Abel L. Genetic dissection of immunity to mycobacteria: the human model. *Annu. Rev. Immunol* 2002;20:581–620. [PubMed: 11861613]
2. Tobin DM, et al. The *Ita4h* locus modulates susceptibility to mycobacterial infection in zebrafish and humans. *Cell* 2010;140:717–730. [PubMed: 20211140]
3. Cooper A. T cells in mycobacterial infection and disease. *Curr. Opin. Immunol* 2009;21:378–384. [PubMed: 19646851]
4. Cooper A. Cell mediated immune responses in tuberculosis. *Annu. Rev. Immunol* 2009;27:393–422. [PubMed: 19302046]
5. Kaufmann SH. Novel tuberculosis vaccination strategies based on understanding the immune response. *J. Intern. Med* 2010;267:337–353. [PubMed: 20433580]
6. Wolf A, et al. Initiation of the adaptive immune response to *Mycobacterium tuberculosis* depends on antigen production in the local lymph node, not the lungs. *J. Exp. Med* 2008;205:105–115. [PubMed: 18158321]
7. Reiley W, et al. ESAT-6-specific CD4 T cell responses to aerosol *Mycobacterium tuberculosis* infection are initiated in mediastinal lymph nodes. *Proc. Natl. Acad. Sci. USA* 2008;105:10961–10966. [PubMed: 18667699]
8. Gallegos A, et al. Delayed protection by ESAT-6-specific effector CD4+ T cells after airborne *M. tuberculosis* infection. *J. Exp. Med* 2008;205:2359–2368. [PubMed: 18779346]
9. Wolf AJ, et al. *Mycobacterium tuberculosis* infects dendritic cells with high frequency and impairs their function in vivo. *J. Immunol* 2007;179:2509–2519. [PubMed: 17675513]
10. Cooper AM, Khader SA. The role of cytokines in the initiation, expansion, and control of cellular immunity to tuberculosis. *Immunol. Rev* 2008;226:191–204. [PubMed: 19161425]
11. Bhatt K, Salgame P. Host innate immune response to *Mycobacterium tuberculosis*. *J. Clin. Immunol* 2007;27:347–362. [PubMed: 17364232]

12. Chen M, et al. A mechanism of virulence: virulent *Mycobacterium tuberculosis* strain H37Rv, but not attenuated H37Ra, causes significant mitochondrial inner membrane disruption in macrophages leading to necrosis. *J. Immunol* 2006;176:3707–3716. [PubMed: 16517739]
13. Flynn J, Chan J. Immunology of tuberculosis. *Annu. Rev. Immunol* 2001;19:93–129. [PubMed: 11244032]
14. Chen M, et al. Lipid mediators in innate immunity against tuberculosis: opposing roles of PGE2 and LXA4 in the induction of macrophage death. *J. Exp. Med* 2008;205:2791–2801. [PubMed: 18955568]
15. Divangahi M, et al. *Mycobacterium tuberculosis* evades macrophage defenses by inhibiting plasma membrane repair. *Nat. Immunol* 2009;10:899–906. [PubMed: 19561612]
16. Behar SM, et al. Evasion of innate immunity by *Mycobacterium tuberculosis*: is death an exit strategy? *Nat. Rev. Microbiol* 2010;8:668–674. [PubMed: 20676146]
17. Divangahi M, et al. Eicosanoid pathways regulate adaptive immunity to *Mycobacterium tuberculosis*. *Nat. Immunol* 2010;11:751–758. [PubMed: 20622882]
18. Scott-Browne J, et al. Expansion and function of Foxp3- expressing T regulatory cells during tuberculosis. *J. Exp. Med* 2007;204:2159–2169. [PubMed: 17709423]
19. Marin ND, et al. Regulatory T cell frequency and modulation of IFN-gamma and IL-17 in active and latent tuberculosis. *Tuberculosis (Edinb)* 2010;90:252–261. [PubMed: 20594914]
20. Garg A, et al. Mannose-capped lipoarabinomannan- and prostaglandin E2-dependent expansion of regulatory T cells in human *Mycobacterium tuberculosis* infection. *Eur. J. Immunol* 2008;38:459–469. [PubMed: 18203140]
21. Ozeki Y, et al. Transient role of CD4+CD25+ regulatory T cells in mycobacterial infection in mice. *Int. Immunol* 2010;22:179–189. [PubMed: 20139174]
22. Shafiani S, et al. Pathogen-specific regulatory T cells delay the arrival of effector T cells in the lung during early tuberculosis. *J. Exp. Med* 2010;207:1409–1420. [PubMed: 20547826]
23. Onishi Y, et al. Foxp3+ natural regulatory T cells preferentially form aggregates on dendritic cells in vitro and actively inhibit their maturation. *Proc. Natl. Acad. Sci. USA* 2008;105:10113–10118. [PubMed: 18635688]
24. Wing K, et al. CTLA-4 control over Foxp3+ regulatory T cell function. *Science* 2008;322:271–275. [PubMed: 18845758]
25. Redford P, et al. Enhanced protection to *Mycobacterium tuberculosis* infection in IL-10-deficient mice is accompanied by early and enhanced Th1 responses in the lung. *Eur. J. Immunol* 2010;40:2200–2210. [PubMed: 20518032]
26. D'Souza CD, et al. An anti-inflammatory role for $\gamma\delta$ T lymphocytes in acquired immunity to *Mycobacterium tuberculosis*. *J. Immunol* 1997;158:1217–1221. [PubMed: 9013962]
27. Okamoto Yoshida Y, et al. Essential role of IL-17A in the formation of a mycobacterial infection-induced granuloma in the lung. *J. Immunol* 2010;184:4414–4422. [PubMed: 20212094]
28. Torrado E, Cooper AM. IL-17 and Th17 cells in tuberculosis. *Cytokine Growth Factor Rev* 2010;21:455–462. [PubMed: 21075039]
29. Khader S, et al. IL-23 compensates for the absence of IL-12p70 and is essential for the IL-17 response during tuberculosis but is dispensable for protection and antigen-specific IFN-gamma responses if IL-12p70 is available. *J. Immunol* 2005;175:788–795. [PubMed: 16002675]
30. Aujla S, et al. Th17 cells and mucosal host defense. *Semin. Immunol* 2007;19:377–382. [PubMed: 18054248]
31. Korf H, et al. Liver X receptors contribute to the protective immune response against *Mycobacterium tuberculosis* in mice. *J. Clin. Invest* 2009;119:1626–1637. [PubMed: 19436111]
32. Gold M, et al. Human mucosal associated invariant T cells detect bacterially infected cells. *PLoS Biol* 2010;8:e1000407. [PubMed: 20613858]
33. Le Bourhis L, et al. Antimicrobial activity of mucosal-associated invariant T cells. *Nat. Immunol* 2010;11:701–708. [PubMed: 20581831]
34. Felio K, et al. CD1-restricted adaptive immune responses to *Mycobacteria* in human group 1 CD1 transgenic mice. *J. Exp. Med* 2009;206:2497–2509. [PubMed: 19808251]

35. Chiba A, et al. Rapid NKT cell responses are self-terminating during the course of microbial infection. *J. Immunol* 2008;181:2292–2302. [PubMed: 18684918]
36. Sada-Ovalle I, et al. α -galactosylceramide as a therapeutic agent for pulmonary *Mycobacterium tuberculosis* infection. *Am. J. Respir. Crit. Care Med* 2010;182:841–847. [PubMed: 20508216]
37. Sada-Ovalle I, et al. Innate invariant NKT cells recognize *Mycobacterium tuberculosis*-infected macrophages, produce interferon- γ , and kill intracellular bacteria. *PLoS Pathog* 2008;4:e1000239. [PubMed: 19079582]
38. Chackerian A, et al. Activation of NKT cells protects mice from tuberculosis. *Infect. Immun* 2002;70:6302–6309. [PubMed: 12379709]
39. Barber DL, et al. Restoring function in exhausted CD8 T cells during chronic viral infection. *Nature* 2006;439:682–687. [PubMed: 16382236]
40. Joshi NS, et al. Inflammation directs memory precursor and short-lived effector CD8(+) T cell fates via the graded expression of T-bet transcription factor. *Immunity* 2007;27:281–295. [PubMed: 17723218]
41. Reiley WW, et al. Distinct functions of antigen-specific CD4 T cells during murine *Mycobacterium tuberculosis* infection. *Proc. Natl. Acad. Sci. USA* 2010;107:19408–19413. [PubMed: 20962277]
42. Lazar-Molnar E, et al. Programmed death-1 (PD-1)-deficient mice are extraordinarily sensitive to tuberculosis. *Proc. Natl. Acad. Sci. USA* 2010;107:13402–13407. [PubMed: 20624978]
43. Barber DL, et al. CD4 T cells promote rather than control tuberculosis in the absence of PD-1 mediated inhibition. *J. Immunol.* 2010 in press.
44. Winslow G, et al. Persistence and turnover of antigen- specific CD4 T cells during chronic tuberculosis infection in the mouse. *J. Immunol* 2003;170:2046–2052. [PubMed: 12574375]
45. Nobrega C, et al. Dissemination of mycobacteria to the thymus renders newly generated T cells tolerant to the invading pathogen. *J. Immunol* 2010;184:351–358. [PubMed: 19949112]
46. Nobrega C, et al. The thymus as a target for mycobacterial infections. *Microbes Infect* 2007;9:1521–1529. [PubMed: 18062904]
47. Cooper AM, et al. Disseminated tuberculosis in interferon gamma gene-disrupted mice. *J. Exp. Med* 1993;178:2243–2247. [PubMed: 8245795]
48. Adams L, et al. The study of *Mycobacterium leprae* in IFN- γ gene disrupted mice as a model to explore the immunopathological spectrum of leprosy. *J. Inf. Dis* 2002;185 suppl 1:S1–S8. [PubMed: 11865434]
49. Cooper AM, et al. IFN- γ and NO in mycobacterial disease: new jobs for old hands. *Trends Microbiol* 2002;10:221–226. [PubMed: 11973155]
50. Desvignes L, Ernst JD. Interferon- γ -responsive nonhematopoietic cells regulate the immune response to *Mycobacterium tuberculosis*. *Immunity* 2009;31:974–985. [PubMed: 20064452]
51. Cruz A, et al. Pathological role of Interleukin 17 in mice subjected to repeated BCG vaccination after infection with *Mycobacterium tuberculosis*. *J. Exp. Med* 2010;207:1609–1616. [PubMed: 20624887]
52. Pearl JE, et al. IL-27 signaling compromises control of bacterial growth in mycobacteria-infected mice. *J. Immunol* 2004;173:7490–7496. [PubMed: 15585875]
53. Holscher C, et al. The IL-27 receptor chain WSX-1 differentially regulates antibacterial immunity and survival during experimental tuberculosis. *J. Immunol* 2005;174:3534–3544. [PubMed: 15749890]
54. Villarino A, et al. Understanding the pro- and anti-inflammatory properties of IL-27. *J. Immunol* 2004;173:715–720. [PubMed: 15240655]
55. Li X, et al. IFN-gamma acts directly on activated CD4+ T cells during mycobacterial infection to promote apoptosis by inducing components of the intracellular apoptosis machinery and by inducing extracellular proapoptotic signals. *J. Immunol* 2007;179:939–949. [PubMed: 17617585]
56. Yoshida H, Miyazaki Y. Regulation of immune responses by Interleukin-27. *Immunol. Rev* 2008;226:234–247. [PubMed: 19161428]
57. Gumperz JE, Brenner MB. CD1-specific T cells in microbial immunity. *Curr. Opin. Immunol* 2001;13:471–478. [PubMed: 11498304]

58. Cohen NR, et al. Antigen presentation by CD1: lipids, T cells, and NKT cells in microbial immunity. *Adv. Immunol* 2009;102:1–94. [PubMed: 19477319]
59. Hava D, et al. Evasion of peptide, but not lipid antigen presentation, through pathogen-induced dendritic cell maturation. *Proc. Natl. Acad. Sci. USA* 2008;105:11281–11286. [PubMed: 18685099]
60. Fischer K, et al. Mycobacterial phosphatidylinositol mannoside is a natural antigen for CD1d-restricted T cells. *Proc. Natl. Acad. Sci. USA* 2004;101:10685–10690. [PubMed: 15243159]
61. Martin E, et al. Stepwise development of MAIT cells in mouse and human. *PLoS Biol* 2009;7:e54. [PubMed: 19278296]

Figure 1. T cell priming is slow to occur during Mtb infection

Very early upon challenge resident phagocytes in the lung phagocytose Mtb. The limited migration of these cells to the dLN allows Mtb to grow and modulate its environment. This modulation results in delayed T cell priming and reduced efficacy of the response in the lung. Death of infected phagocytes is an important link to acquired immunity, specifically apoptosis of the host phagocyte allows for the antigen cargo contained in the apoptotic vesicles to be picked up by DCs which then traffic to the dLN and more efficiently prime naïve T cells [16,17]. Mtb-specific Tregs are induced in the dLN and limit effector T cell priming and their accumulation in the lung, possibly via IL-10 [22,25].

Figure 2. Maintaining a balanced T cell response during chronic mycobacterial infection limits tissue damage

Different cell populations initiate granuloma formation following high dose challenge with $\gamma\delta$ T cells producing IL-17 that enhances neutrophil migration to the lung [28]. Neutrophils and IL-17 induce chemokine gradients that guide newly primed T cells to the lung and into the granuloma. Initially, distinct T cell subsets have high proliferative potential, produce cytokines and express high levels of PD-1 [41], which is required to regulate the pathological potential of the activated cells [43]. Constant IL-12 and IL-27 signaling can lead to more differentiated cell populations with lower proliferative potential but enhanced capacity to produce cytokines. When the balance between different T cell populations is shifted or extensive cell death occurs, elevated IL-23 expression can enhance ongoing IL-17 responses with excessive neutrophil recruitment and important pathological consequences [50,51]. New thymic emigrants may be required to enter the response at and they may impact the Th1/Th17 balance, however further research is required to clarify their role during infection.