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Host Innate Immune Response to *Mycobacterium tuberculosis*

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This review focuses on recent progress in our understanding of *Mycobacterium tuberculosis* survival in macrophages, the interaction of *M. tuberculosis* with Toll-like receptors (TLRs) and the establishment of the link between innate and adaptive immunity, and TLRs and interferon-γ-mediated antimicrobial pathways in macrophages. We also propose a paradigm that TLR2 signaling regulates the magnitude of the host Th1 response leading to either *M. tuberculosis* persistence and latent infection or replication and disease.

KEY WORDS: *Mycobaterium tuberculosis*; innate immunity; TLR2; dendritic cells; macrophages.

INTRODUCTION

The naissance of innate immunity was the description of macrophage phagocytosis by Metchnikoff (1). Today, the science of innate immunity is more than just phagocytosis. The innate immune response comprises several different cell types, has its own receptor system to recognize the presence of pathogens, and is a key to the initiation of an adaptive immune response in the host. No wonder, successful pathogens have evolved ways to evade innate immune killing in order to find a niche in the host. In this review, we will discuss host innate immunity generated in response to the pathogen, *Mycobacterium tuberculosis* (Mtb).

Mtb infects approximately one-third of the world's population (2). Close to eight million new cases of tuberculosis occur each year, accounting for approximately 7% of all deaths and 26% of all avoidable adult deaths in developing countries (3). Despite the implementation of TB control programs, case rates continue to soar where the prevalence of HIV infection is high (4). The situation is further complicated by a worldwide increase in drug resistant and MDR-TB, and the recent reports of XDR TB (5). Thus, the resurgence of TB truly constitutes a global health crisis (6).

Tuberculosis begins with the inhalation of Mtb-containing aerosols into the pulmonary alveoli. Here, the bacteria bind to phagocytic receptors and enter resident alveolar macrophages, dendritic cells, and monocytes recruited from the bloodstream. Besides expressing phagocytic receptors, macrophages and dendritic cells also express Toll-like receptors (TLRs) that recognize conserved molecular patterns expressed on pathogens (7–9). Ligation of TLRs by these pathogen-specific ligands initiates a signal transduction pathway in the host cell that culminates in the activation of NFκb and the induction of cytokines and chemokines (10) that are crucial to eliciting the adaptive immune response against the pathogen. Consequently, activation of TLR is an important link between innate cellular response and the subsequent activation of adaptive immune defense against microbial pathogens.

As depicted in Fig. 1, a small percentage of individuals, despite exposure to Mtb, remain uninfected, most likely due to the expression of high innate immunity. However, in the majority of individuals who are exposed to Mtb, the innate response cannot protect from infection, and effector Th1 cytokines of the adaptive immune response are necessary to restrict bacterial growth and mediate protection. The adaptive immunity generated in these people, although protective, nonetheless does not induce sterilizing immunity. These individuals therefore remain latently infected, and are vulnerable to disease reactivation when their immune surveillance weakens or when their immune response is compromised. Reactivation tuberculosis contributes significantly to the morbidity and mortality associated with the disease (11, 12), and is believed to account for a substantial portion of TB cases in HIV-infected individuals (13). In another small proportion of individuals, infection leads directly to primary tuberculosis due to a

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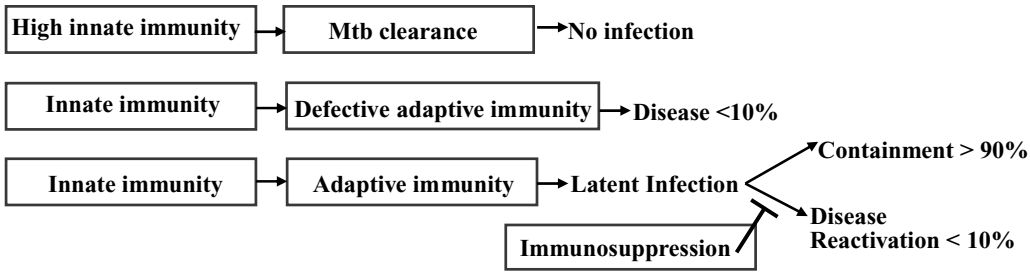

Fig. 1. Three possible outcomes of Mtb exposure. In some individuals, Mtb is eliminated by the host immediately upon inhalation. The frequency and the cause of such spontaneous healing are not certain. In the second and the largest group of individuals, infection is contained as a result of successful granuloma formation, a function of strong innate and adaptive immune response by the host, which results in latent infection. In this group, reactivation of latent infection can occur due to factors such as aging or the immunocompromised status of the host. In a small number of infected individuals, adaptive immunity fails and they develop primary tuberculosis.

failure of their adaptive immune response to control the initial bacterial replication.

In the last several years, we have gained a better appreciation regarding host innate immune response to Mtb, although much needs to be learned regarding the nature of the innate response that prevents establishment of Mtb infection. This review focuses on our current understanding of the regulation of host innate immune response to Mtb and how it interfaces with the adaptive immune response. The review is not all-inclusive, and only areas where substantial progress has been made will be addressed. Specifically, we will discuss (i) Mtb entry and subsequent survival inside macrophages; (ii) Mtb interaction with innate receptors, specifically TLRs; (iii) Mtb-induced cytokine and chemokine induction in macrophages and dendritic cells; (iv) maturation and migration of dendritic cells—a key step that links innate and adaptive anti-Mtb immunity; (v) modulation of innate immunity by effectors of the adaptive immune response. Finally, in closing, we will explore the evolving paradigm that innate immune response generated in the host in response to Mtb infection is not only important for initiating anti-Mtb immunity, but the innate response concomitantly also activates regulatory pathways in the host. Whether the regulatory mechanisms activated are important for controlling the magnitude of the host immune response to prevent immunopathology, or whether this is a virulence strategy for the TB bacillus to persist in the host will be discussed.

MTB PHAGOCYTOSIS AND SURVIVAL INSIDE MACROPHAGE

Macrophages lining the alveolar spaces of the lungs represent the first line of defense upon aerosol infection of the host with Mtb. *In vitro* studies have implicated complement receptor (CR)3 as a major receptor on macrophages for phagocytosis of Mtb (14). Nonetheless, several other macrophage surface receptors, such as CR1, CR4, Mannose receptor, CD14, and Scavenger receptors can also recognize and bind Mtb *in vitro* (15). Pulmonary surfactant protein A (Sp-A) too enhances Mtb uptake by human macrophages (16, 17). In this context, it is worth noting that although CR3 was determined as the primary mode of macrophage entry for Mtb *in vitro*, mice lacking CR3 exhibited similar bacterial burden and host response to that of CR3-sufficient mice (18). Cholesterol accumulates at the site of Mtb entry into macrophages, and depleting cells of cholesterol prevent Mtb internalization (19). This indicates that perhaps cholesterol accumulation around phagocytic receptors rather than the nature of the receptor itself dictates Mtb uptake. Whether *in vivo* a similar relocation of cholesterol occurs at sites of Mtb entry into macrophages and whether it provides a distinct advantage to the TB bacillus remains to be determined.

Once inside the host cell, Mtb successfully evades destruction by the innate microbicidal machinery. Armstrong and Hart in papers published in the early 1970s (20, 21) shaped our understanding of how Mtb might persist in the host. Their work showed that Mtb vacuoles did not fuse with the lysosomal compartment. Substantive work from several laboratories has built on this seminal observation to provide a detailed insight into the molecular events that arrest the maturation of the Mtb phagosome and prevent its further biogenesis and acquisition of lysosomal components. The early trafficking pattern of the Mtb phagosome is normal and exhibits fusion with certain early endosomal compartments, since both iron (22, 23) and glycosphingolipids (24) are found associated with the Mtb phagosome. The arrested phagosome, however, lacks the vacuolar ATPase and lysosomal hydrolases (25). Subsequent studies that monitored the trafficking pattern

of the Mtb phagosome showed that the arrest occurs between the acquisition of the endocytic vesicles Rab5 and Rab7. The Mtb phagosome is associated with Rab5 (26), but not Rab7 (27), and furthermore exhibits reduced recruitment of the early endosomal autoantigen 1 (EEA1) (28), an effector molecule of Rab5 required for organelle tethering and delivery of lysosomal hydrolases, cathepsins, and vacuolar ATPases from the trans golgi network to the phagosome. The Mtb phagosome also lacks a specific type III phosphatidylinositol 3-kinase, hVPs34 (29) whose activation product phospatidylinositol 3-phosphate aids the retention of EEA1 to the endosomal membrane (29).

How and what components of Mtb block the Mtb phagosome from undergoing the typical phagosome biogenesis? The arrest of the Mtb phagosome at the Rab 5 stage and its inability to proceed through the maturation pathway, at least partly, results from Mtb-induced inhibition of sphingosine kinase activity and subsequent Ca^{2+} signaling pathway in the cell, a step necessary for recruitment of hVPs35 to membranes of organelles (30). Another study reported that maturation of the Mtb phagosome is impeded because Mtb suppresses phagosomal actin assembly (31). Yet another study determined that the Mtb phagosome arrest was dependent on its initial fusion with early endosomes and acquisition of iron (32). Manosylated lipoarabinamannan (ManLAM), the Mtb analog of host phophatidylinositol-3 phosphate is responsible for actively inhibiting Mtb phagosome from fully maturing and acquiring lysosomal hydrolases (33, 34). Indeed, it has been shown that phagocytosis of Mtb via binding of its cell surface LAM to the mannose receptor on human macrophages led to the non-fusogenic phenotype of the Mtb phagosome (35). Interestingly, other Mtb lipids such as PIMs enhance Mtb phagosome fusion with early endosomes, possibly providing the phagosome access to host nutrients (36). Besides lipids, a protein from Mtb, protein kinase G (PknG) has also been implicated in interfering with the transfer of Mtb phagosome to the lysosomal compartment (37).

Our understanding of Mtb phagocytosis, the biogenesis of Mtb phagosome, and intracellular growth of Mtb is almost entirely derived from studies examining interaction of Mtb with macrophages. However, it is necessary to also understand the handling of Mtb inside dendritic cells, since in response to an aerosol challenge with Mtb, dendritic cells take up Mtb, and as will be discussed later, are crucial to linking the innate and adaptive immune responses. Besides the expression of CR3 and mannose receptor, dendritic cells are endowed with an additional phagocytic receptor for binding Mtb. Dendritic cells express the C-type lectin, DC-SIGN (DC-specific intercellular adhesion molecule-grabbing nonintegrin), and Mtb can bind DC-SIGN through manLAM expressed on their surface. A comparative analysis of Mtb survival within human macrophages and dendritic cells revealed that unlike macrophages, dendritic cells did not support intracellular growth of Mtb (38, 39). Despite being a key player in the innate response to Mtb, we know very little regarding the trafficking of Mtb vacuole inside dendritic cells; except for a study that reported that endosomal trafficking is significantly reduced compared to that in macrophages (39). Clearly, more detailed studies examining the intracellular fate of Mtb inside dendritic cells are needed.

It must be emphasized that Mtb replication in the macrophage is also controlled at the level of host factors. Expression of a candidate gene *IntracellularPathogenResistance* (*lpr1*) within the *sst1* locus limits Mtb multiplication in the host (40). Variants in the human equivalent of the lpr1 gene SP110 were shown to be associated with genetic susceptibility to TB in a study of families in West Africa (41). Another association study in human TB, also in West Africa, however, found no association of human pulmonary TB with SP110 variants (42). Undoubtedly, more studies in genetically different populations are needed to equivocally determine the role of SP100 and other potential candidate genes in susceptibility to TB.

MTB INTERACTION WITH TLRS

Engagement of TLR by Mtb ligands is an early event in the interaction of Mtb with its host cell. Accumulating data indicate that Mtb expresses a large repertoire of TLR2 ligands. The 19-kDa lipoprotein (LpqH), a secreted antigen of Mtb, was the first Mtb ligand shown to interact specifically with TLR2 to induce TNFα and nitric oxide production from both murine and human macrophages (43). In addition, the 19-kDa lipoprotein is a major inducer of interleukin (IL)-12 production in human monocytes (43). LprA (Rv1270) (44) and LprG (Rv1411c) (45) are two other mycobacterial lipoproteins that are TLR2 agonists. In addition to lipoproteins, lipomannan (46) and phosphatidyl-*myo*-inositol mannoside (PIM) (47, 48) also interact with TLR2 to initiate cellular activation (48). However, with regards to PIMs, Abel *et al.* (49) demonstrated that PIM structures can also elicit cellular activation via TLR4. They showed that PIM was able to induce NFκB activation in a dose-dependent manner in stable TLR4 and MD-2 Ba-F3 transformants. A systematic biochemical characterization of four acyl forms of lipomannan (LM) from *M. bovis* BCG that differed in their degree of acylation, indicated that only the triacylated LM was a potent TLR2 agonist (50).

Interestingly, ManLAM derived from virulent Mtb fails to activate either TLR2 or TLR4-transfected cells (51). In contrast, AraLAM purified from fast-growing mycobacteria is capable of TLR2-mediated cellular activation (51).

Studies aimed at determining the requirement of TLR4 in controlling Mtb infection following an aerosol challenge showed that lack of TLR4 did not compromise host resistance to TB (52, 53). However, a high dose of Mtb infection did lead to enhanced susceptibility in the absence of TLR4 signaling (49). It is interesting that despite a large collection of TLR2 agonists on the TB bacillus, murine studies indicate that TLR2 is not essential for host resistance against tuberculosis. In a model of low-dose aerosol infection, TLR2 (53, 54) deficiency did not affect host defense against Mtb infection. However, in one of the two studies (53) with high-dose aerosol infection, a role for TLR2 in host resistance was revealed. The TLR2-deficient mice were not compromised in their ability to induce Th1 immunity, but on the contrary, exhibited exaggerated immunopathology. *In vitro* studies have shown that engagement of TLR2 with Mtb ligands induces inhibition of macrophage MHC class II antigen presentation (55) and also blocks macrophage responsiveness to IFNγ (56, 57). Together with the *in vivo* studies, these *in vitro* findings that TLR2 signaling negatively modulates macrophage functions point to the need for future studies designed to examine whether the negative signaling from TLR2 curtails Th1 activation, and whether this is important for balancing protection and immunopathology in the host.

Our studies examining the *in vitro* interaction of TLR with live Mtb reported that in response to Mtb, dendritic cells secreted copious amount of IL-12, while the secretion was limited in infected macrophages. The study also reported that Mtb induced rapid and significantly higher remodeling at the IL-12p40 promoter in dendritic cells in comparison to macrophages. The mechanism behind the differential remodeling at the IL-12p40 promoter and subsequent IL-12 release was shown to be due to differences in TLR usage by macrophages and dendritic cells. Mtb induced IL-12 secretion from dendritic cells in a TLR9-dependent manner while in macrophages it was TLR2-dependent (58). Consistent with this, the greatest effect on the progression of tuberculosis disease was seen in mice doubly deficient in TLR2 and TLR9 (59).

Although IFNγ is undoubtedly necessary for resistance against Mtb infection (60), it is of interest that there exist antimycobacterial pathways that are independent of IFNγ and are induced by TLR in human macrophages. For example, it has been known for some time that activation of the Vitamin D3 pathway controls Mtb replication in human macrophages (61). Also, it had been documented that Vitamin D deficiency is a risk factor for tuberculosis (62). Only recently, however, Modlin and colleagues deciphered the mechanism for Vitamin-D3-mediated antimicrobial pathway. They demonstrated that TLR2-mediated activation of macrophages upregulated the expression of Vitamin D receptor and Vitamin-D-1-hydroxylase genes, leading to the induction of the antimicrobial peptide, cathelicidin (63). The study from Modlin's group also showed that African American individuals who are more susceptible to Mtb infection and disease were not efficient in inducing the antimicrobial peptide, cathelicidin. The TLR2-mediated innate mechanism of mycobacterial killing provides a scientific basis for tuberculosis treatment of a century ago: exposure to sunlight. Other TLR-induced killing mechanisms may also participate in the innate response. For instance, CpG, an activator of the TLR9-pathway, also induces rapid antimycobacterial responses in macrophages, in a phospholipase D-dependent manner (64). These innate mechanisms for killing Mtb provoke future investigations of whether individuals who never become infected with Mtb have the capacity to activate these pathways and overpower the Mtb-induced block in phagosome maturation.

MTB-INDUCED UPSURGE OF CYTOKINE AND CHEMOKINE SECRETION

A major consequence of Mtb interaction with the TLRs on macrophages and dendritic cells is the burst in cytokine and chemokine secretion. The induction of these effector molecules regulates the formation of the granuloma and is responsible for initiating and shaping the adaptive immune response to Mtb. The contribution of adaptive immunity to the evolving tubercle granuloma in the lung will not be elaborated in this review. The reader is referred to other reviews on the topic (65–67).

Cytokines Important for Induction of Th1 Immunity

Clearly, induction of cellular Th1 immunity is critical for protection against tuberculosis as evidenced by enhanced disease in the HIV-infected (68) and from experiments of nature where individuals carrying defective genes for IFNγR and IL-12R (69) are exquisitely susceptible to intracellular pathogens, including mycobacteria. Currently, there are three well-defined cytokines that steer naïve T cells toward Th1 commitment (70). IL-12 was the first cytokine to be described with potent Th1 promoting attributes, followed by the discovery of IL-23

(shares the p40 component with IL-12) and the recent addition of IL-27 to this list. Work from several laboratories has revealed that the three cytokines together orchestrate Th1 responses, with IL-12 being the prototypic and dominant cytokine that affects both the induction and maintenance of Th1 immunity. IL-23, on the other hand, has activities on memory T cells and IL-27, secreted prior to IL-12 by antigen-presenting cells, is involved in Th1 initiation.

In patients with tuberculosis pleuritis, a clinical form of disease that is mostly self-healing, high IL-12 levels were found in the pleural fluid (71). Two studies comparing murine macrophages and dendritic cells demonstrated that dendritic cells release significantly higher IL-12 than did macrophages in response to live Mtb (72, 73). *In vitro*, Mtb-infected dendritic cells also primed naïve T cells toward Th1 development, while macrophages did not; though, IL-23-secreting macrophages were capable of inducing Mtb-specific Th1 response (74).

Early studies in the murine model of tuberculosis clearly demonstrated that the cytokine IL-12 that is necessary to drive Th1 responses and IFNγ—the effector molecule of the Th1 response—were both necessary for protection against Mtb infection. Mice deficient in the p40 component of IL-12 or in IFNγ (GKO) were both highly susceptible to Mtb infection. Exogenous supplementation of IL-12 at the onset of disease led to reduction in bacterial burden and delayed the lung pathology in the relatively susceptible Balb/C strain of mice (75). However, IL-12 supplementation did not lead to enhanced protection in GKO mice indicating that IFNγ is downstream of IL-12 and is the effector molecule mediating protection in the host. In another study (76), the role of endogenous IL-12 was studied by neutralization with anti-IL-12 antibodies. It was found that in Balb/C mice neutralization of IL-12 at the onset of infection led to disruption in the ability of the host to contain infection; however, neutralization of the cytokine after the onset of infection (third week) did not affect bacterial replication. This suggests that the presence of IL-12 is more critical during early infection when anti-Mtb adaptive immunity is being shaped toward Th1-type. However, in later studies, reconstitution of IL-12-p40 gene-deficient mice with recombinant IL-12 only during the early phase of infection was determined not to be sufficient to provide long-term immunity, despite early control of bacterial growth. Transfer of immune CD4 T cells from Mtb-infected wild-type mice to Rag −/− provided immunity against infection. However, similar reconstitution of immune CD4 T cells into Rag−/− mice that were also deficient in IL-12-p40 failed to induce protection. This provides experimental evidence that sustained production of IL-12 throughout the course of infection is necessary to maintain antibacterial immunity in the host (77). A reason why this study differed from the previous where IL-12 was shown not to be necessary for long-term immunity may probably be due to the presence of residual IL-12 activity in the neutralization experiments.

To further characterize whether susceptibility to Mtb infection in the absence of p40 is due to the lack of biologically active IL-12 or is a consequence of defective IL-23 production, mice deficient in the specific components of IL-12 and IL-23, p35 and p19, respectively, were studied. Mice lacking p19 were able to control Mtb infection as well as the wild-type mice (78, 79). Mice lacking p35 were able to control bacterial replication slightly better than the p40-deficient mice (79, 80), but in comparison to the p19 knockout mice exhibited significantly higher bacterial burden. Mice doubly deficient in p35 and p19 genes were as susceptible as the p40-gene-deficient mice (79). Exogenous delivery into the lung of IL-23 via adenoviral vectors enhanced anti-Mtb immunity, upregulated IL-17 expression, and reduced bacterial burden in the lungs of Mtb-infected mice (81). Together, these data indicate that IL-23 is less critical for protection against Mtb, and only provides a moderate level of protection to the host in the absence of biologically active IL-12. On the other hand, IL-12 has a far more vital role in the generation of protective anti-Mtb immunity.

IL-27 is also an IL-12-related cytokine and WSX-1 is a component of the IL-27R complex (82). IL-27/IL-27R signaling, interestingly, exhibits both pro- and anti-inflammatory properties. Infection of WSX-1−/− mice with Mtb revealed that, in the absence of IL-27R signaling, there was a reduced bacterial burden accompanied by enhanced CD4 infiltration into the lungs (83). Another group examining Mtb infection in the same WSX-1−/− mice observed increased IL-12-p40 and TNFα expression and enhanced IFNγ production from CD4 T cells. This group also reported reduced Mtb burden in the lungs of infected WSX-1 knockout mice in comparison with wild-type mice. Despite restricted bacterial growth, the WSX-1 knockout mice succumbed to infection due to exaggerated immunopathology, a scenario similar to what was first reported with *Toxoplasma gondii* infection in this strain of knockout mice. Under conditions where pathogens do not induce a Th1 response in the host, for example in *Leishmania major* infection of Balb/c mice, absence of WSX-1 resulted in the generation of protective Th1 response with concomitant Th2 downregulation in the host (84). A tenet for future perusal is that the anti-inflammatory activity of IL-27 is perhaps more critical than its Th1 promoting activity in response to pathogens that have high Th1-inducing potential (85, 86).

TNF

TNFα plays an important role in regulating the pathology of tuberculosis (87). TNFα exists in both soluble and membrane bound forms and signals through TNFαR. Mtb infection leads to TNFα secretion by macrophages, dendritic cells, and T cells (60). Secretion of TNFα by Mtb-infected macrophages is a potent mechanism to induce killing of Mtb via generation of reactive nitrogen intermediates in conjunction with IFNγ (88). An attribute of membrane TNFα is to induce apoptosis of the Mtb-infected alveolar macrophages (89), and thereby indirectly contribute to the reduction of bacterial burden. TNFα's ability to induce alveolar macrophage apoptosis may also be important in the cross-presentation of Mtb antigens for CD8 cytotoxic T cell priming (90). It has also been suggested that inhibition of TNFα-mediated macrophage apoptosis is a virulence strategy of Mtb. Avirulent H37Ra induced TNFα-dependent macrophage apoptosis, while virulent H37Rv released soluble TNFR2 that reduced TNFα activity and subsequent apoptosis of macrophages (91). Although it needs to be examined in more detail, TNFα has been shown to support the growth of Mtb in human monocytes and macrophages (92).

The requirement for TNFα in host defense against Mtb infection was demonstrated in studies which showed that mice treated with antibody to TNFα became more susceptible to BCG infection and exhibited malformed granulomas (93). Mtb infection of mice lacking TNF receptor or neutralization of TNFα activity in mice also led to the failure to control bacterial replication resulting in enhanced susceptibility (94). This study indicated that TNFα contributed to maintaining host resistance by inducing the production of reactive nitrogen intermediates by macrophages. Later studies have indicated that TNFα also participates in setting the chemokine circuitry in the developing granuloma. Mice lacking TNFα had reduced chemokine expression in lung granulomas (95–97) and this resulted in reduced T cell infiltration into the lungs and a failure to form a productive granuloma.

For the most part, the immunological forces that control reactivation remain ill defined, except for the knowledge that TNFα is a major player (98). Studies in a murine latent model of tuberculosis from the Chan and Flynn laboratories clearly demonstrated that neutralization of TNFα during the latent/persistent phase induced reactivation in C57BL/6 mice, as indicated by the enhanced bacterial burden in the lungs (99). Further, histological examination of lung tissue from TNFα-neutralized animals revealed a disorganized granuloma with indications of a lack of cellular turnover and increased fluid accumulation. NOS2 expression was attenuated, while IL-10 expression was upregulated in the lungs. Immunohistochemical analysis indicated an increased presence of apoptotic T cells and macrophages in the lung, a feature not seen previously in other reactivation models. The importance of TNF in maintaining Mtb in a chronic/persistent phase in mice has been corroborated in humans. It has been observed that the use of anti-TNFα antibody in patients undergoing treatment for rheumatoid arthritis has resulted in reactivation of tuberculosis in some latently infected individuals (100, 101).

Mtb-Induced Chemokines

In vitro and *in vivo* studies provide evidence for the participation of chemokines in the control of TB. It is present in the innate and adaptive immune response to Mtb (96, 102). Mtb infection of both human and murine macrophages results in the secretion of a large number of chemokines, including CCL2, CCL3, CCL7, CCL12, CXCL2, and CXCL10 (96). A comparative chemokine expression analysis showed that lung interstitial macrophages from a susceptible mouse strain expressed significantly high levels of CXCL13 and CXCL14, while higher expression of CXCL9 and CXCL0 was found in macrophages from the resistant strain of mice (103). Regulation of chemokine production in macrophages is predominantly regulated by TNFα. Mtb infection of macrophages leads to the production of TNFα which, in turn, regulates the secretion of a plethora of chemokines from macrophages, including CCL2, CCL3, CCL4, CCL5, CXCL10, and CXCL13 (104). Mtb-infected dendritic cells also secrete chemokines, including CXCL9, CXCL10, CCL3, and CCL4. CXCL10 secretion was IFNα-dependent and in conjunction with CXCL9 and CXCL3 acted to recruit inflammatory cells to the site of Mtb infection (105).

Studies to examine the role of chemokine and chemokine receptors in host resistance against Mtb infection has led to conflicting results, in great part due to redundancy in the function of chemokines and their receptors. In a mouse model of tuberculosis, it has been shown that the first step in recruitment of cells into the lung, specifically recruitment of immature dendritic cells and monocytes to the site of infection, is mediated by CCR2 and, as a consequence, CCR2−/− mice (106) are more susceptible to Mtb infection. These mice also show defective recruitment of dendritic cell to the draining lymph nodes resulting in delayed and reduced priming of naive T cells. However, a later study (107) indicated that susceptibility of CCR2 knockout mice to Mtb infection was dose dependent. As seen with high dose infection, a low-dose aerosol challenge of the CCR2 knockout mice with

Mtb also resulted in reduced cellular migration to the lungs and delayed priming. However, there was no change in bacterial burden or susceptibility to infection. Using chimeric mice, where either the myeloid or the lymphoid compartment was lacking CCR2, it was determined that expression of CCR2 on macrophages and dendritic cells was important for the recruitment of T cells to lungs (108). However, mice deficient in CCL2/MCP-1, which is a ligand for CCR2, do not show reduced susceptibility to Mtb infection, thereby indicating that *in vivo* other chemokines such as CCL7, CCL8, and CCL12 can compensate for the lack of CCL2/MCP-1 (109).

In addition to its expression on granulocytes and macrophages, CCR5 is also present on immature dendritic cells and Mtb modulates its expression. Indeed, mycobacterial Hsp70 can interact with CCR5 on immature dendritic cells and induce their maturation and the interaction also induces IL-12 secretion from dendritic cells (110). Despite CCR5-mediated IL-12 production and the enhanced production of CCR5 ligands, MIP-1α, MIP-1β, and RANTES in the lungs of Mtb-infected mice, absence of CCR5 did not affect the ability of the host to control bacterial replication in the lung (111, 112). The latter study, in addition, observed that CCR5−/− mice exhibited increased bacterial burden in the draining lymph nodes (112). The intriguing possibility that CCR5 signals impede Mtb-bearing dendritic cell migration resulting in enhanced accumulation of Mtb in the draining lymph nodes needs further scrutiny.

CCR7 expression on cells guides their migration to the draining lymph nodes where its cognate ligands CCL19 and CCL21 are present. Indeed, Mtb infection upregulates CCR7 expression on dendritic cells, but absence of CCR7 did not enhance bacterial replication (113). Although it must be noted that the granulomas of CCR7−/− mice had altered granuloma architecture with enhanced inflammation and a lack of follicular B cell architecture. How these changes in the granuloma affect host resistance is still not clear. As discussed in the next section, Mtb-infected dendritic cells migrate to the draining lymph nodes to initiate an immune response. Therefore, it would be important to determine what chemokine/receptor gradient controls the migration of Mtb-infected dendritic cells from lungs to draining lymph nodes.

CXCR3 is a chemokine receptor preferentially expressed on activated Th1 cells and regulates their migration in response to ligands CXCL10, CXCL9, and CXCL11. Given the importance of Th1 in host resistance against TB, C57BL/6 mice deficient in CXCR3 were studied following Mtb infection. Despite the ability of CXCR3 to regulate the migration of Th1 cells, absence of the receptor did not affect Mtb replication in the host. The CXCR3 knockout mice, however, did exhibit a neutrophil deficit in the granuloma. The consequence to host resistance of reduced neutrophils in the granuloma remains unclear (114). Contrary findings were reported in a recent study that examined mice lacking CXCR3 on the BALB/c background. In this study, the CXCR3-deficient mice exhibited heightened resistance to Mtb infection in the chronic phase when compared with wild-type mice (115), and the mice also had enhanced T cell activation (115). The authors of the paper suggest that enhanced resistance in BALB/cCXCR3 knockout mice could have resulted from the absence of the immunosuppressive CXCR3–CXCL10 chemokine gradient. Certain chemokine gradients, including CXCR3–CXCL10, have recently been recognized as immunosuppressive and to interfere with the formation of the immunological synapse (116). Together, these studies highlight the emerging recognition that the chemokine circuitry activated during Mtb infection may not only regulate cellular recruitment, but also directly impact on the function of immune cells. The studies also highlight the role of genetic differences in regulating chemokine functions.

IL-10

Dendritic cells and macrophages in response to Mtb produce the immunosuppressive and anti-inflammatory cytokine IL-10. Interestingly, dendritic cells secrete substantial IL-12 in response to Mtb infection and can prime naive T cells toward Th1-type, despite concomitant secretion of IL-10 (73, 117). IL-10-secreting CD8 suppressor/regulatory T cells are associated with susceptibility to Mtb infection (118) and T cell expressing both IFNγ and IL-10 have been isolated from the bronchoalveolar lavage fluid of TB patients (119). Additionally, depressed T-cell IFNγ responses in pulmonary tuberculosis was shown to be associated with the induction of IL-10 from monocytes (120). Interestingly, in patients with pleural TB, considered as the resistant and self-healing form of the disease, IL-10 is found along with IFNγ at sites of infection in the pleural fluid (121). *In vitro*, IL-10 downregulates the production of IL-12 in human monocytes infected with Mtb (122). Also, IL-10 down modulates the activity of CD4 and CD8 T cells via downregulation of costimulatory molecules on macrophages (123). In addition, IL-10 inhibits the proliferation of IFNγ producing T cells and $\gamma\delta$ T cells (124). Although absence of IL-10 did not enhance resistance to Mtb infection in IL-10 knockout mice (125), transgenic over-expression of IL-10 resulted in reactivation of chronic disease (126). Similarly, expression of human IL 10 in mice under the control of MHC II promoter enhanced the susceptibility to disease, independent of

T-cell-derived IL-10. The Mtb-infected macrophages from these transgenic mice exhibited reduced antimycobacterial capacity (127). That IL-10 may have a role in TB is suggested by the fact that polymorphism in murine SLC11 A1, a tuberculosis susceptibility locus, has been associated with variation in IL-10 production (128). Together, these data suggest that IL-10 is induced by Mtb and suppresses the generation anti-Mtb immunity. However, Th1 cytokines are often found along with IL-10. Perhaps the relative quantities of the two cytokines determine whether Th1 immunity is suppressed or not.

DENDRITIC CELL MATURATION, MIGRATION, AND ANTIGEN PRESENTATION

Recent work from several laboratories has focused on dissecting the role of dendritic cells in Mtb infection. Upon its interaction with Mtb, dendritic cells undergo a repertoire of phenotypical changes, a process termed as maturation. This process, which is TLR-dependent, brings forth three major phenotypic changes in dendritic cells: upregulation of costimulatory molecules-CD40, B7.1 and B7.2, heightened expression of adhesion molecules, and upregulation of chemokine receptor—CCR7. Whereas immature dendritic cells are efficient at Mtb phagocytosis and exhibit enhanced microbicidal property, maturation endows them with the role of an efficient antigen presenter and initiator of adaptive immune responses (129).

Following Mtb phagocytosis and concomitant TLR activation, the next step in the development of host immunity is the transport of pathogen from the lung to the draining lymph nodes, where the matured dendritic cells can present antigen to naive T cells and initiate the process of adaptive immune response. Although Mtb uptake and engagement of TLR signaling for cellular activation occurs in macrophages and dendritic cells, only the latter cell type was shown to acquire the capacity to upregulate CCR7 expression and migrate to draining lymph nodes (130). Consistent with this study that tracked intratracheally instilled cells, endogenous lung dendritic cells also exhibited similar migratory property. Following intratracheal infection of mice with GFP-expressing BCG, dissemination of mycobacteria from the lung was initiated by the migration of infected dendritic cells to the draining lymph nodes (131), despite predominant infection of alveolar macrophages. Another study in BCG-infected mice also demonstrated that dendritic cells, and not macrophages, were the antigen-presenting cells responsible for priming naïve T cells (132). Direct evidence for the role of dendritic cells as the priming APC for initiating pulmonary immunity came from the study where mice depleted of CD11c + dendritic cells exhibited delayed CD4 responses to Mtb and worsening disease (133). It has been argued that in addition to ferrying antigen for T cell priming, migration of dendritic cells to the lymph nodes may also aid in Mtb dissemination (131).

Are other cell types involved in transporting Mtb antigens to the draining lymph nodes? Indeed, a recent report implicates neutrophils as the carrier of live BCG following intradermal vaccination from peripheral tissue to the DLN capsule (134). Whether neutrophils participate in antigen transport during a pulmonary infection with Mtb would be worth investigating, particularly since neutrophils appear to marginally influence early immune responses and the architecture of the ensuing granuloma (135, 136). Following Mtb infection, there is an influx of macrophages and dendritic cells from the periphery into the lung. The relative contribution of interstitial versus the newly recruited dendritic cells in Mtb transport from lung to the draining lymph nodes is also not clear. However, absence of CCR2 was shown to impair macrophage and dendritic cells trafficking into the infected lungs resulting in susceptibility to Mtb infection (108), suggesting that dendritic cells recruited to the lung may also function to carry Mtb to the draining lymph nodes.

Collective data indicate that dendritic cells are the antigen-presenting cells that migrate to the draining lymph nodes, and process and present Mtb antigens on MHC Classes I and II to naïve $CD4^+$ and $CD8^+$ cells, respectively. This review will not address the mechanisms of antigen processing and presentation and other molecular events controlling T cell activation. However, we discuss here one study from Kaufman's group that demonstrated a detour pathway for how antigens of Mtb, presumably confined within the phagosome, are delivered to Class I molecules. This study demonstrated that dendritic cells take up apoptotic vesicles containing Mtb, the vesicles are then degraded in an endosomal-dependent manner and Mtb antigens are cross-presented on MHC Class I molecules to CD8 T cells (137).

Since maturation and migration of dendritic cells is such a key step in linking innate and adaptive immunity, it is not surprising that Mtb negatively interferes with this step. IL-1β release from mycobacteria-infected antigen-presenting cells inhibits dendritic cell maturation (138) and virulent Mtb has been reported to impair the maturation of monocyte derived dendritic cells (139). The migration of dendritic cells appears to be regulated to some degree by IL-12p40 homodimers (140). Whether IL-10, which can impede dendritic cell migration (141), functions by downregulating the p40 homodimers is worth considering. In addition, one also needs

to examine whether in human infection Mtb interference with the dendritic cell maturation and migration process is dependent on the degree of virulence of the infecting clinical strain and its ability to induce IL-10 production.

MODULATION OF INNATE IMMUNITY BY EFFECTORS OF THE ADAPTIVE IMMUNE RESPONSE

In effect, despite the early induction of chemokines and cytokines from macrophages, Mtb is able to skillfully avoid the innate antimicrobial defense mechanisms of the macrophage and find a safe niche in the phagosome for intracellular growth. IFNγ, an effector molecule of the adaptive immune response, halts this unimpeded growth of Mtb in the macrophage (60). Although it is clear that IFNγ is highly effective in restricting mycobacterial growth in macrophages (60), the mechanisms through which this is achieved is not fully understood. NOS2-mediated antibacterial pathway is one mechanism that has been extensively studied in the murine model of tuberculosis, wherein it has been demonstrated that IFNγ, in conjunction with TNF, upregulates NOS2 and the production of reactive nitrogen intermediates within the phagolysosome, resulting in Mtb killing (142). Nitric oxide also reacts with glutathione to form s-nitrosoglutathione that is toxic to Mtb (143). Despite this accumulated evidence of IFNγ-mediated antimycobacterial activity in the murine model, the Mtb killing mechanism in human macrophages is less clear (144). Individuals with mutations in the IFNγ receptor are more susceptible to mycobacterial infections suggest that IFNγ-mediated antimycobacterial pathways are active in human macrophages (69). Alveolar macrophages of tuberculosis patients express NOS2, and isolation of NOS2-expressing macrophages exhibit antimycobacterial activity *in vitro*, which can be abolished in the presence of NOS2 inhibitors (145). Thus, whether NOS2-mediated pathway is active in human macrophages remains an open question and awaits better methodologies for studying the enzyme.

Recent evidence indicates that IFNγ induced LRG-47, a GTP-binding protein has a principal role in the ability of the host to control Mtb replication, since mice lacking LRG-47 are highly susceptible to Mtb infection (146). Based on several elegant works, autophagy, originally defined as a cellular homeostatic process, is emerging as a powerful host defense machinery of innate immune cells (147). IFNγ, through enhancement of LRG-47 activity, was shown to induce autophagy in Mtb-infected macrophages, which resulted in revoking the restriction on Mtb phagosome maturation and delivery of the antimicrobial contents of the lysosomal compartment (148). Whether autophagy or more specifically "immunophagy," (a term coined by Deretic to define the specialized function of autophagy in host immunity (147)) restricts Mtb growth in human macrophages and whether innate immune signaling pathways such as TLRs induce immunophagy are all very significant questions that need to be addressed.

OTHER INNATE IMMUNE CELLS

Besides macrophages and dendritic cells, $\gamma\delta$ T cells, NK cells, and NKT cells also participate in the innate immune response to TB. Murine studies have indicated that the induction of $\gamma\delta$ T cell in the immune response against TB precedes that of conventional CD4 and CD8 cells and hence plays an important role in modulating the effector response against tuberculosis. Following infection, the early recruitment of cells to the lung is mediated by the chemokines, CXCL2 and CXCL10, and the cytokine, IL-12 released by macrophages and dendritic cells in the lungs (149). Once activated, $\gamma\delta$ T cells secrete IFNγ and TNFα. The production of these cytokine strengthens the bactericidal capacity of macrophages by induction of NOS2. Recently, it has been shown that $\gamma\delta$ T cells secrete IL 17 in response to IL 23 secreted by dendritic cells, thereby implicating them as a main player in the resistance against infection at the initial stage (150). Response of mice deficient in $\gamma\delta$ T cells to Mtb infection is dependent on the dose and the route of infection. These knockout mice are able to contain Mtb infection with a low inoculum; however, infection with a higher inoculum of Mtb administered intravenously resulted in the formation of pyogenic granulomas, indicating that a role for these cells is perhaps in cellular traffic during infection (151, 152). In these experiments with high dose of virulent Mtb, the lung pathology indicated enhanced migration of neutrophils and increased size of granuloma thereby indicating a role of $\gamma\delta$ T cells in granuloma formation and mycobacterial containment (152).

The antigen specificity of murine $\gamma\delta$ has not been well studied. However, it has been shown that murine $\gamma\delta$ cells do not respond to phosphate antigens, which are recognized by human $\gamma\delta$ cells. Upon contact with Mtb, $\gamma\delta$ cells have been shown to secrete IL-2 and exhibit cytolytic function and hence involved in innate immune effector mechanism (153). In both humans and primates, antigen-specific $\gamma\delta$ cells recognizing phosphoantigens have been documented and have been shown to generate a memory response (154, 155). Loss of Vγ9+/V$\gamma\delta$2+ subset of $\gamma\delta$ T cells was shown to be correlated with tuberculosis (156).

NK cells are recruited to the lungs early during Mtb infection. There they are known to expand and become a primary source of IFNγ. Activated NK cells are known to cause lysis of infected macrophage by utilizing NK cell receptor in TLR-dependent manner (157). NK cell depletion studies have shown no change in bacterial burden (158). However, recent studies (159) indicate that NK cells provide resistance during early Mtb infection via production of IFNγ. The IFNγ activated macrophage in a NOS2-dependent manner and also by regulating neutrophil migration to the lung for controlling lung inflammation. Future experiments defining the exact contribution of NK cells and the IFNγ secreted from them to innate immunity would help understand whether NK cell activation has a role in preventing infection following exposure to Mtb.

NKT cells are TCR-expressing T cells which also express the NK cell marker NK1.1. In mice, NKT cells are mainly represented by Vα14 NKT cells, while in humans, there is a homologous population of Vα24 NKT cells. NKT cells are known to recognize nonpeptide antigens in the context of CD1d. Role of NKT cells in tuberculosis has been studied in both humans and mice. Human Vα24-restrcited NKT cells are activated by α-galactosylceramide. CD1d-restricted NKT in the presence of α-galactosylceramide cells restrict the growth of Mtb in a granulolysin-dependent manner (160). It has been shown that NKT cells induce a granulomatous response to a glycolipid fraction of Mtb cell wall (161). This finding is further supported by the fact that α-galactosylceramide-activated NKT cells contribute to enhanced resistance against Mtb infection (162).

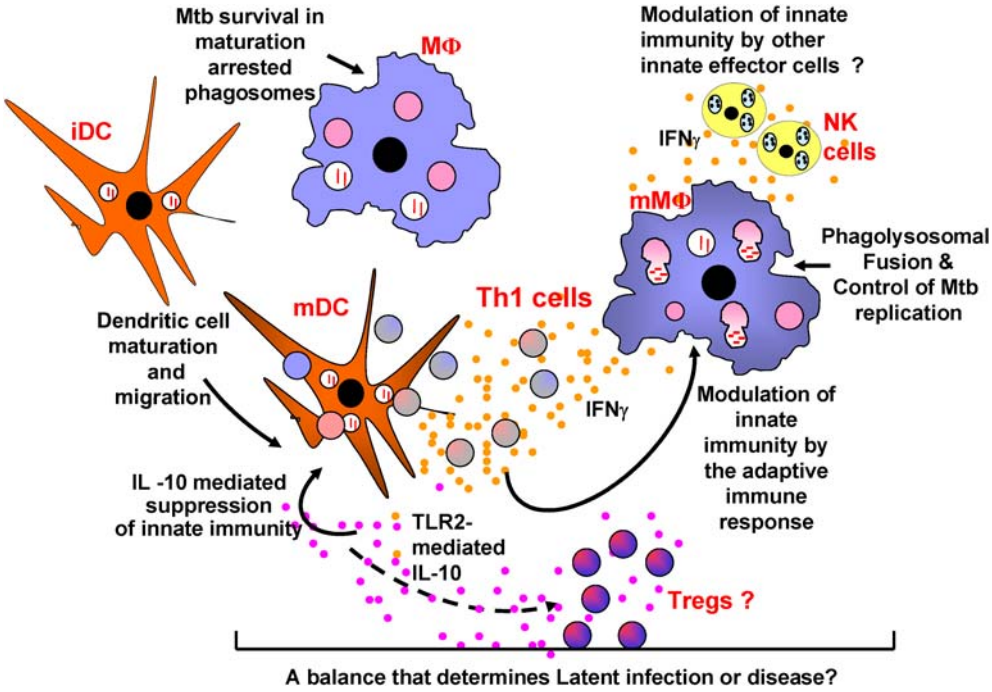

Fig. 2. A paradigm for how the innate immune response to Mtb regulates the adaptive immune response. Initially, Mtb survives and replicates inside macrophages since it can prevent fusion of its phagosome with the lysosomal compartment. Concomitantly, dendritic cells capture Mtb, undergo maturation, and migrate to the draining lymph nodes. Adaptive response is initiated in the draining lymph nodes wherein naïve antigen-specific T cells are primed by dendritic cells to Th1 and cytotoxic effector cell types. Mycobactericidal function of macrophages is dependent on IFNγ, initially produced by innate immune cells such as NK cells and later on provided by effector T cells. The secreted IFNγ promotes phagolysosomal fusion and enhances Mtb killing. TLR2-mediated innate IL-10 is released during the induction of innate immune response and subsequent Th1 induction. The role of the innate IL-10 is to control the magnitude of the Th1 response by either down modulating antigen-presentation function or by inducing T regulatory cells. The TLR2/IL-10 axis, on the one hand, is important for allowing Mtb to achieve the latent state and, on the other hand, may also cause excessive immunosuppression leading to disease. The *unbroken lines* indicate that experimental evidence is available and *dashed lines* indicate that it is speculative and is an area for future investigation.

There is still much to learn regarding the contribution of $\gamma\delta$ T cells, NK cells, and NKT cells to the overall innate immune protection against TB in humans. A next step is to study these cells in the context of protection against clinical strains of Mtb in humans.

CLOSING THOUGHTS

An emerging principle in intracellular parasitism is that successful pathogens such as Mtb have acquired the ability to persist in the host without always inducing disease and mortality (163). The strategy on the part of Mtb is to induce sufficient Th1 immunity in the host to control its replication but not result in its complete eradication. The advantage to the host is minimal collateral damage to lung tissue. Thus, Mtb remains dormant in the host for decades, in a sort of symbiotic relationship. Under certain altered conditions in the host, Mtb will reactivate and cause immunopathology such as lung cavitation, which increases its infectivity and thereby maintains the cycle of transmission to new hosts.

Although Mtb interacts with several different TLRs on host cells, we posit that to establish a latent infection Mtb specifically usurps the innate TLR2 signaling in the host to blunt Th1 immune responses. Supporting evidence for the paradigm include (i) Mtb possess a large gamut of ligands for TLR2; (ii) Mtb/TLR2 interaction suppresses macrophage functions; (iii) innate IL-10 secretion by dendritic cells and macrophages in response to live Mtb is TLR-2 dependent; and (iv) absence of TLR2 results in exaggerated immunopathology in the host. The mechanisms for limiting Th1 response may include inhibition of antigen-presentation functions and induction of T regulatory cells (Fig. 2).

A corollary to the paradigm is that virulent strains can tip the balance toward immunosuppression using the same TLR2 signaling pathway. Although there is no evidence that TLR2 interaction is necessary for protection against Mtb disease, it must be pointed out that these conclusions are drawn from studies performed with laboratory strains of Mtb. It is highly probable that the interaction of clinical strains of Mtb with the TLR2 complex does result in potent immunosuppression in the host. In this regard, it would be interesting to determine if the immunosuppressive cytokines induced by the phenolic glycolipid of the virulent Beijing strains (164) is TLR2-mediated, and whether the induction of T regulatory cells present in TB patients (165, 166) is TLR2-dependent.

Clearly, future studies should investigate if the differing interaction of Mtb clinical strains with the TLR2/IL-10 axis is the control switch that determines whether the outcome of Mtb exposure is latent infection or tuberculosis disease.

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REFERENCES

1. Metchnikoff E: Immunity to Infective Diseases. London, Cambridge University Press, 1905
2. World Health Organization: Anti-Tuberculosis Drug Resistance in the World: The WHO/IUTLD Global Project on Anti-Tuberculosis Drug Resistance Surveillance. Geneva, Switzerland, World Health Organization, 2001
3. Murray JF: Tuberculosis and HIV infection: A global perspective. Respiration 65:335, 1998
4. De Cock KM, Chaisson RE: Will DOTS do it? A reappraisal of tuberculosis control in countries with high rates of HIV infection. Int J Tuberc Lung Dis 3:457, 1999
5. Pablos-Mendez A, Raviglione MC, Laszlo A, Binkin N, Rieder HL, Bustreo F, Cohn DL, Lambregts-van Weezenbeek CS, Kim SJ, Chaulet P, Nunn P: Global surveillance for antituberculosis-drug resistance, 1994–1997. World Health Organization–International Union against Tuberculosis and Lung Disease Working Group on Anti-Tuberculosis Drug Resistance Surveillance. N Engl J Med 338:1641, 1998
6. Raviglione MC, Snider DE, Jr, Kochi A: Global epidemiology of tuberculosis. Morbidity and mortality of a worldwide epidemic. JAMA 273:220, 1995
7. Medzhitov R, Janeway CJ: The Toll receptor family and microbial recognition. Trends Microbiol 10:452, 2000
8. Kaisho T, Akira S: Critical roles of Toll-like receptors in host defense. Crit Rev Immunol 20:393, 2000
9. Takeda K, Kaisho T, Akira S: Toll-like receptors. Annu Rev Immunol 21:335, 2003
10. Aderem A, Ulevitch RJ: Toll-like receptors in the induction of the innate immune response. Nature 406:782, 2000
11. Stead WW: Pathogenesis of a first episode of chronic pulmonary tuberculosis in man: Recrudescence of residuals of the primary infection or exogenous reinfection? Am Rev Respir Dis 95:729, 1967
12. Stead WW: The pathogenesis of pulmonary tuberculosis among older persons. Am Rev Respir Dis 91:811, 1965
13. Selwyn PA, Hartel D, Lewis VA, Schhoenbaum EE, Vermuns SH, Klein RS, Walker AT, Freidland GH: A prospective study of the risk of tuberculosis among intravenous drug users with human immunodeficiency virus infection. N Engl J Med 320:545, 1989
14. Schlesinger LS, Bellinger-Kawahara CG, Payne NR, Horwitz MA: Phagocytosis of *Mycobacterium tuberculosis* is mediated by human monocyte complement receptors and complement C3. J Immunol 144:2771, 1990
15. Ernst JD: Macrophage receptors for *Mycobacterium tuberculosis*. Infect Immun 66:1277, 1998
16. Beharka AA, Gaynor CD, Kang BK, Voelker DR, McCormack FX, Schlesinger LS: Pulmonary surfactant protein A up-regulates activity of the mannose receptor, a pattern recognition receptor expressed on human macrophages. J Immunol 169:3565, 2002

17. Gaynor CD, McCormack FX, Voelker DR, McGowan SE, Schlesinger LS: Pulmonary surfactant protein A mediates enhanced phagocytosis of *Mycobacterium tuberculosis* by a direct interaction with human macrophages. J Immunol 155:5343, 1995
18. Hu C, Mayadas TN, Tanaka K, Chan J, Salgame P: *Mycobacterium tuberculosis* infection in complement receptor 3-deficient mice. J Immunol 165:2596, 2000
19. Gatfield J, Pieters J: Essential role for cholesterol in entry of mycobacteria into macrophages. Science 288:1647, 2000
20. Armstrong JA, Hart PD: Response of cultured macrophages to *M. tuberculosis* with observations of fusion of lysosomes with phagosomes. J Exp Med 134:713, 1971
21. Armstrong JA, Hart PD: Phagosome–lysosome interactions in cultured macrophages infected with virulent tubercle bacilli. Reversal of the usual nonfusion pattern and observations on bacterial survival. J Exp Med 142:1, 1975
22. Sturgill-Koszycki S, Schaible UE, Russell DG: Mycobacterium-containing phagosomes are accessible to early endosomes and reflect a transitional state in normal phagosome biogenesis. EMBO J 15:6960, 1996
23. Clemens DL, Horwitz MA: The *Mycobacterium tuberculosis* phagosome interacts with early endosomes and is accessible to exogenously administered transferrin. J Exp Med 184:1349, 1996
24. Russell DG, Dant J, Sturgill-Koszycki S: *Mycobacterium avium*- and *Mycobacterium tuberculosis*-containing vacuoles are dynamic, fusion-competent vesicles that are accessible to glycosphingolipids from the host cell plasmalemma. J Immunol 156:4764, 1996
25. Sturgill-Koszycki S, Schlesinger PH, Chakraborty P, Haddix PL, Collins HL, Fok AK, Allen RD, Gluck SL, Heuser J, Russell DG: Lack of acidification in *Mycobacterium phagosomes* produced by exclusion of the vesicular proton-ATPase. Science 263:678, 1994
26. Clemens DL, Lee BY, Horwitz MA: Deviant expression of Rab5 on phagosomes containing the intracellular pathogens *Mycobacterium tuberculosis* and *Legionella pneumophila* is associated with altered phagosomal fate. Infect Immun 68:2671, 2000
27. Via LE, Deretic D, Ulmer RJ, Hibler NS, Huber LA, Deretic V: Arrest of mycobacterial phagosome maturation is caused by a block in vesicle fusion between stages controlled by Rab5 and Rab7. J Biol Chem 272:13326, 1997
28. Fratti RA, Backer JM, Gruenberg J, Corvera S, Deretic V: Role of phosphatidylinositol 3-kinase and Rab5 effectors in phagosomal biogenesis and mycobacterial phagosome maturation arrest. J Cell Biol 154:631, 2001
29. Deretic V, Vergne I, Chua J, Master S, Singh SB, Fazio JA, Kyei G: Endosomal membrane traffic: Convergence point targeted by *Mycobacterium tuberculosis* and HIV. Cell Microbiol 6:999, 2004
30. Kusner DJ: Mechanisms of mycobacterial persistence in tuberculosis. Clin Immunol 114:239, 2005
31. Anes E, Kuhnel MP, Bos E, Moniz-Pereira J, Habermann A, Griffiths G: Selected lipids activate phagosome actin assembly and maturation resulting in killing of pathogenic mycobacteria. Nat Cell Biol 5:793, 2003
32. Kelley VA, Schorey JS: Mycobacterium's arrest of phagosome maturation in macrophages requires Rab5 activity and accessibility to iron. Mol Biol Cell 14:3366, 2003
33. Fratti RA, Chua J, Vergne I, Deretic V: Mycobacterium tuberculosis glycosylated phosphatidylinositol causes phagosome maturation arrest. Proc Natl Acad Sci USA 100:5437, 2003
34. Chua J, Vergne I, Master S, Deretic V: A tale of two lipids: *Mycobacterium tuberculosis* phagosome maturation arrest. Curr Opin Microbiol 7:71, 2004
35. Kang PB, Azad AK, Torrelles JB, Kaufman TM, Beharka A, Tibesar E, DesJardin LE, Schlesinger LS: The human macrophage mannose receptor directs *Mycobacterium tuberculosis* lipoarabinomannan-mediated phagosome biogenesis. J Exp Med 202:987, 2005
36. Vergne I, Fratti RA, Hill PJ, Chua J, Belisle J, Deretic V: *Mycobacterium tuberculosis* phagosome maturation arrest: Mycobacterial phosphatidylinositol analog phosphatidylinositol mannoside stimulates early endosomal fusion. Mol Biol Cell 15:751, 2004
37. Walburger A, Koul A, Ferrari G, Nguyen L, Prescianotto-Baschong C, Huygen K, Klebl B, Thompson C, Bacher G, Pieters J: Protein kinase G from pathogenic mycobacteria promotes survival within macrophages. Science 304:1800, 2004
38. Bodnar KA, Serbina NV, Flynn JL: Fate of *Mycobacterium tuberculosis* within murine dendritic cells. Infect Immun 69:800, 2001
39. Tailleux L, Neyrolles O, Honore-Bouakline S, Perret E, Sanchez F, Abastado JP, Lagrange PH, Gluckman JC, Rosenzwajg M, Herrmann JL: Constrained intracellular survival of *Mycobacterium tuberculosis* in human dendritic cells. J Immunol 170:1939, 2003
40. Pan H, Yan BS, Rojas M, Shebzukhov YV, Zhou H, Kobzik L, Higgins DE, Daly MJ, Bloom BR, Kramnik I: Ipr1 gene mediates innate immunity to tuberculosis. Nature 434:767, 2005
41. Tosh K, Campbell SJ, Fielding K, Sillah J, Bah B, Gustafson P, Manneh K, Lisse I, Sirugo G, Bennett S, Aaby P, McAdam KP, Bah-Sow O, Lienhardt C, Kramnik I, Hill AV: Variants in the SP110 gene are associated with genetic susceptibility to tuberculosis in West Africa. Proc Natl Acad Sci USA 103:10364, 2006
42. Thye T, Browne EN, Chinbuah MA, Gyapong J, Osei I, Owusu-Dabo E, Niemann S, Rusch-Gerdes S, Horstmann RD, Meyer CG: No associations of human pulmonary tuberculosis with Sp110 variants. J Med Genet 43:e32, 2006
43. Brightbill HD, Libraty DH, Krutzik SR, Yang RB, Belisle JT, Bleharski JR, Maitland M, Norgard MV, Plevy SE, Smale ST, Brennan PJ, Bloom BR, Godowski PJ, Modlin RL: Host defense mechanisms triggered by microbial lipoproteins through toll-like receptors. Science 285:732, 1999
44. Pecora ND, Gehring AJ, Canaday DH, Boom WH, Harding CV: *Mycobacterium tuberculosis* LprA is a lipoprotein agonist of TLR2 that regulates innate immunity and APC function. J Immunol 177:422, 2006
45. Gehring AJ, Dobos KM, Belisle JT, Harding CV, Boom WH: *Mycobacterium tuberculosis* LprG (Rv1411c): A novel TLR-2 ligand that inhibits human macrophage class II MHC antigen processing. J Immunol 173:2660, 2004
46. Quesniaux VJ, Nicolle DM, Torres D, Kremer L, Guerardel Y, Nigou J, Puzo G, Erard F, Ryffel B: Toll-like receptor 2 (TLR2)-dependent-positive and TLR2-independent-negative regulation of proinflammatory cytokines by mycobacterial lipomannans. J Immunol 172:4425, 2004
47. Gilleron M, Himoudi N, Adam O, Constant P, Venisse A, Riviere M, Puzo G: Mycobacterium smegmatis phosphoinositols-glyceroarabinomannans. Structure and localization of alkali-labile and alkali-stable phosphoinositides. J Biol Chem 272:117, 1997
48. Jones BW, Means TK, Heldwein KA, Keen MA, Hill PJ, Belisle JT, Fenton MJ: Different Toll-like receptor agonists induce distinct macrophage responses. J Leukoc Biol 69:1036, 2001
49. Abel B, Thieblemont N, Quesniaux VJ, Brown N, Mpagi J, Miyake K, Bihl F, Ryffel B: Toll-like receptor 4 expression is required to control chronic *Mycobacterium tuberculosis* infection in mice. J Immunol 169:3155, 2002

50. Gilleron M, Nigou J, Nicolle D, Quesniaux V, Puzo G: The acylation state of mycobacterial lipomannans modulates innate immunity response through toll-like receptor 2. Chem Biol 13:39, 2006
51. Means TK, Lien E, Yoshimura A, Wang S, Golenbock DT, Fenton MJ: The CD14 ligands lipoarabinomannan and lipopolysaccharide differ in their requirement for Toll-like receptors. J Immunol 163:6748, 1999
52. Kamath AB, Alt J, Debbabi H, Behar SM: Toll-like receptor 4-defective C3H/HeJ mice are not more susceptible than other C3H substrains to infection with *Mycobacterium tuberculosis*. Infect Immun 71:4112, 2003
53. Reiling N, Holscher C, Fehrenbach A, Kroger S, Kirschning CJ, Goyert S, Ehlers S: Cutting edge: Toll-like receptor (TLR)2- and TLR4-mediated pathogen recognition in resistance to airborne infection with *Mycobacterium tuberculosis*. J Immunol 169:3480, 2002
54. Sugawara I, Yamada H, Li C, Mizuno S, Takeuchi O, Akira S: Mycobacterial infection in TLR2 and TLR6 knockout mice. Microbiol Immunol 47:327, 2003
55. Noss EH, Pai RK, Sellati TJ, Radolf JD, Belisle J, Golenbock DT, Boom WH, Harding CV: Toll-like receptor 2-dependent inhibition of macrophage class II MHC expression and antigen processing by 19-kDa lipoprotein of *Mycobacterium tuberculosis*. J Immunol 167:910, 2001
56. Fortune SM, Solache A, Jaeger A, Hill PJ, Belisle JT, Bloom BR, Rubin EJ, Ernst JD: *Mycobacterium tuberculosis* inhibits macrophage responses to IFN-gamma through myeloid differentiation factor 88-dependent and -independent mechanisms. J Immunol 172:6272, 2004
57. Banaiee N, Kincaid EZ, Buchwald U, Jacobs WR, Jr, Ernst JD: Potent inhibition of macrophage responses to IFN-gamma by live virulent *Mycobacterium tuberculosis* is independent of mature mycobacterial lipoproteins but dependent on TLR2. J Immunol 176:3019, 2006
58. Pompei L, Jang S, Zamlynny B, Ravikumar S, McBride A, Hickman SP, Salgame P: Disparity in interleukin-12 release in dendritic cells and macrophages in response to *Mycobacterium tuberculosis* is due to utilization of distinct Toll-like receptors. J Immunol, 2007 (in press)
59. Bafica A, Scanga CA, Feng CG, Leifer C, Cheever A, Sher A: TLR9 regulates Th1 responses and cooperates with TLR2 in mediating optimal resistance to *Mycobacterium tuberculosis*. J Exp Med 202:1715, 2005
60. Flynn JL, Chan J: Immunology of tuberculosis. Annu Rev Immunol 19:93, 2001
61. Denis M: Killing of *Mycobacterium tuberculosis* within human monocytes: Activation by cytokines and calcitriol. Clin Exp Immunol 84:200, 1991
62. Wilkinson RJ, Llewelyn M, Toossi Z, Patel P, Pasvol G, Lalvani A, Wright D, Latif M, Davidson RN: Influence of vitamin D deficiency and vitamin D receptor polymorphisms on tuberculosis among Gujarati Asians in west London: A case–control study. Lancet 355:618, 2000
63. Liu PT, Stenger S, Li H, Wenzel L, Tan BH, Krutzik SR, Ochoa MT, Schauber J, Wu K, Meinken C, Kamen DL, Wagner M, Bals R, Steinmeyer A, Zugel U, Gallo RL, Eisenberg D, Hewison M, Hollis BW, Adams JS, Bloom BR, Modlin RL: Toll-like receptor triggering of a vitamin D-mediated human antimicrobial response. Science 311:1770, 2006
64. Auricchio G, Garg SK, Martino A, Volpe E, Ciaramella A, De Vito P, Baldini PM, Colizzi V, Fraziano M: Role of macrophage phospholipase D in natural and CpG-induced antimycobacterial activity. Cell Microbiol 5:913, 2003
65. Russell DG: Who puts the tubercle in tuberculosis? Nat Rev Microbiol 5:39, 2007
66. Ulrichs T, Kaufmann SH: New insights into the function of granulomas in human tuberculosis. J Pathol 208:261, 2006
67. Salgame P: Host innate and Th1 responses and the bacterial factors that control *Mycobacterium tuberculosis* infection. Curr Opin Immunol 17:374, 2005
68. Estaquier J, Idziorek T, Weiping Z, Emilie D, Farber C-M, Bourez J-M, Ameisen JC: T helper type 1/T helper type 2 cytokines and T cell death: Preventive effect of IL-12 on activation-induced and CD95 (Fas/Apo-1)-mediated apoptosis of $CD4^+$ T cells from human immunodeficiency virus-infected persons. J Exp Med 182:1759, 1995
69. Rosenzweig SD, Holland SM: Defects in the interferon-gamma and interleukin-12 pathways. Immunol Rev 203:38, 2005
70. Brombacher F, Kastelein RA, Alber G: Novel IL-12 family members shed light on the orchestration of Th1 responses. Trends Immunol 24:207, 2003
71. Zhang M, Gong J, Iyer DV, Jones BE, Modlin RL, Barnes PF: T cell cytokine responses in persons with tuberculosis and human immunodeficiency virus infection. J Clin Invest 94:2435, 1994
72. Giacomini E, Iona E, Ferroni L, Miettinen M, Fattorini L, Orefici G, Julkunen I, Coccia EM: Infection of human macrophages and dendritic cells with *Mycobacterium tuberculosis* induces a differential cytokine gene expression that mosulates T cell response. J Immunol 166:7033, 2001
73. Hickman SP, Chan J, Salgame P: *Mycobacterium tuberculosis* induces differential cytokine production from dendritic cells and macrophages with divergent effects on naive T cell polarization. J Immunol 168:4636, 2002
74. Verreck FA, de Boer T, Langenberg DM, Hoeve MA, Kramer M, Vaisberg E, Kastelein R, Kolk A, de Waal-Malefyt R, Ottenhoff TH: Human IL-23-producing type 1 macrophages promote but IL-10-producing type 2 macrophages subvert immunity to (myco)bacteria. Proc Natl Acad Sci USA 101:4560, 2004
75. Flynn J, Goldstein M, Triebold K, Sypek J, Wolf S, Bloom B: IL-12 increases resistance of BALB/c mice to *Mycobacterium tuberculosis* infection. J Immunol 155:2515, 1995
76. Castro A, Silva R, Appelberg R: Endogenously produced IL-12 is required for the induction of protective T cells during *Mycobacterium avium* infections in mice. J Immunol 155:2013, 1995
77. Feng CG, Jankovic D, Kullberg M, Cheever A, Scanga CA, Hieny S, Caspar P, Yap GS, Sher A: Maintenance of pulmonary Th1 effector function in chronic tuberculosis requires persistent IL-12 production. J Immunol 174:4185, 2005
78. Khader SA, Pearl JE, Sakamoto K, Gilmartin L, Bell GK, Jelley-Gibbs DM, Ghilardi N, deSauvage F, Cooper AM: 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 175:788, 2005
79. Chackerian AA, Chen S-J, Brodie SJ, Mattson JD, McClanahan TK, Kastelein RA, Bowman EP: Neutralization or absence of the interleukin-23 pathway does not compromise immunity to mycobacterial infection. Infect Immun 74:6092, 2006
80. Cooper AM, Kipnis A, Turner J, Magram J, Ferrante J, Orme IM: Mice lacking bioactive IL-12 can generate protective, antigen-specific cellular responses to mycobacterial infection only if the IL-12 p40 subunit is present. J Immunol 168:1322, 2002

81. Happel KI, Lockhart EA, Mason CM, Porretta E, Keoshkerian E, Odden AR, Nelson S, Ramsay AJ: Pulmonary interleukin-23 gene delivery increases local T-cell immunity and controls growth of *Mycobacterium tuberculosis* in the lungs. Infect Immun 73:5782, 2005
82. Hunter CA: New IL-12-family members: IL-23 and IL-27, cytokines with divergent functions. Nat Rev Immunol 5:521, 2005
83. Pearl JE, Khader SA, Solache A, Gilmartin L, Ghilardi N, de-Sauvage F, Cooper AM: IL-27 signaling compromises control of bacterial growth in mycobacteria-infected mice. J Immunol 173:7490, 2004
84. Artis D, Johnson LM, Joyce K, Saris C, Villarino A, Hunter CA, Scott P: Cutting edge: Early IL-4 production governs the requirement for IL-27-WSX-1 signaling in the development of protective Th1 cytokine responses following Leishmania major infection. J Immunol 172:4672, 2004
85. Villarino A, Hibbert L, Lieberman L, Wilson E, Mak T, Yoshida H, Kastelein RA, Saris C, Hunter CA: The IL-27R (WSX-1) is required to suppress T cell hyperactivity during infection. Immunity 19:645, 2003
86. Villarino AV, Huang E, Hunter CA: Understanding the pro- and anti-inflammatory properties of IL-27. J Immunol 173:715, 2004
87. Stenger S: Immunological control of tuberculosis: Role of tumour necrosis factor and more. Ann Rheum Dis 64(Suppl 4):iv24, 2005
88. Chan J, Xing Y, Magliozzo R, Bloom B: Killing of virulent *Mycobacterium tuberculosis* by reactive nitrogen intermediates produced by activated murine macrophages. J Exp Med 175:1111, 1992
89. Keane J, Balcewicz-Sablinska MR, Remold HG, Chupp GL, Meek BB, Fenton MJ, Kornfeld H: Infection by *Mycobacterium tuberculosis* promotes human alveolar macrophage apoptosis. Infect Immun 65:298, 1997
90. Winau F, Weber S, Sad S, de Diego J, Hoops SL, Breiden B, Sandhoff K, Brinkmann V, Kaufmann SHE, Schaible UE: Apoptotic vesicles crossprime CD8 T cells and protect against tuberculosis. Immunity 24:105, 2006
91. Balcewicz-Sablinska MK, Keane J, Kornfeld H, Remold HG: Pathogenic *Mycobacterium tuberculosis* evades apoptosis of host macrophages by release of TNF-R2, resulting in inactivation of TNF-alpha. J Immunol 161:2636, 1998
92. Engele M, Castiglione K, Schwerdtner N, Wagner M, Bolcskei P, Rollinghoff M, Stenger S: Induction of TNF in human alveolar macrophages as a potential evasion mechanism of virulent *Mycobacterium tuberculosis*. J Immunol 168:1328, 2002
93. Kindler V, Sappino A-P, Grau GE, Piguet P-F, Vassalli P: The inducing role of tumor necrosis factor in the development of bactericidal granulomas during BCG infection. Cell 56:731, 1989
94. Flynn JL, Goldstein MM, Chan J, Triebold KJ, Pfeffer K, Loenstein CL, Schreiber R, Mak TW, Bloom BR: Tumor necrosis factor is required in the protective immune response against *Mycobacterium tuberculosis* in mice. Immunity 2:561, 1995
95. Roach DR, Bean AGD, Demangel C, France MP, Briscoe H, Britton WJ: TNF regulates chemokine induction essential for cell recruitment, granuloma formation, and clearance of mycobacterial infection. J Immunol 168:4620, 2002
96. Algood HMS, Chan J, Flynn JL: Chemokines and tuberculosis. Cytokine Growth Factor Rev 14:467, 2003
97. Algood HMS, Lin PL, Flynn JL: Tumor necrosis factor and chemokine interactions in the formation and maintenance of granulomas in tuberculosis. Clin Infect Dis 41:S189, 2005
98. Tufariello JM, Chan J, Flynn JL: Latent tuberculosis: Mechanisms of host and bacillus that contribute to persistent infection. Lancet Infect Dis 3:578, 2003
99. Mohan VP, Scanga CA, Yu K, Scott HM, Tanaka KE, Tsang E, Tsai MM, Flynn JL, Chan J: Effects of tumor necrosis factor alpha on host immune response in chronic persistent tuberculosis: Possible role for limiting pathology. Infect Immun 69:1847, 2001
100. Keane J, Gershon S, Wise RP, Mirabile-Levens E, Kasznica J, Schwieterman WD, Siegel JN, Braun MM: Tuberculosis associated with infliximab, a tumor necrosis factor {alpha}-neutralizing agent. N Engl J Med 345:1098, 2001
101. Gardam MA, Keystone EC, Menzies R, Manners S, Skamene E, Long R, Vinh DC: Anti-tumour necrosis factor agents and tuberculosis risk: Mechanisms of action and clinical management. Lancet Infect Dis 3:148, 2003
102. Peters W, Ernst JD: Mechanisms of cell recruitment in the immune response to *Mycobacterium tuberculosis*. Microbes Infect 5:151, 2003
103. Orlova MO, Majorov KB, Lyadova IV, Eruslanov EB, M'lan CE, Greenwood CMT, Schurr E, Apt AS: Constitutive differences in gene expression profiles parallel genetic patterns of susceptibility to tuberculosis in mice. Infect Immun 74:3668, 2006
104. Tessier P, Naccache P, Clark-Lewis I, Gladue R, Neote K, McColl S: Chemokine networks *in vivo*: Involvement of C-X-C and C-C chemokines in neutrophil extravasation *in vivo* in response to TNF-alpha. J Immunol 159:3595, 1997
105. Lande R, Giacomini E, Grassi T, Remoli ME, Iona E, Miettinen M, Julkunen I, Coccia EM: IFN-alpha beta released by *Mycobacterium tuberculosis*-infected human dendritic cells induces the expression of CXCL10: Selective recruitment of NK and activated T cells. J Immunol 170:1174, 2003
106. Peters W, Scott HM, Chambers HF, Flynn JL, Charo IF, Ernst JD: Chemokine receptor 2 serves an early and essential role in resistance to *Mycobacterium tuberculosis*. Proc Natl Acad Sci USA 98:7958, 2001
107. Scott HM, Flynn JL: *Mycobacterium tuberculosis* in chemokine receptor 2-deficient mice: Influence of dose on disease progression. Infect Immun 70:5946, 2002
108. Peters W, Cyster JG, Mack M, Schlondorff D, Wolf AJ, Ernst JD, Charo IF: CCR2-dependent trafficking of F4/80dim macrophages and CD11cdim/intermediate dendritic cells is crucial for T cell recruitment to lungs infected with *Mycobacterium tuberculosis*. J Immunol 172:7647, 2004
109. Kipnis A, Basaraba RJ, Orme IM, Cooper AM: Role of chemokine ligand 2 in the protective response to early murine pulmonary tuberculosis. Immunology 109:547, 2003
110. Floto RA, MacAry PA, Boname JM, Mien TS, Kampmann B, Hair JR, Huey OS, Houben ENG, Pieters J, Day C, Oehlmann W, Singh M, Smith KGC, Lehner PJ: Dendritic cell stimulation by mycobacterial Hsp70 is mediated through CCR5. Science 314:454, 2006
111. Badewa AP, Quinton LJ, Shellito JE, Mason CM: Chemokine receptor 5 and its ligands in the immune response to murine tuberculosis. Tuberculosis 85:185, 2005
112. Algood HM, Flynn JL: CCR5-deficient mice control *Mycobacterium tuberculosis* infection despite increased pulmonary lymphocytic infiltration. J Immunol 173:3287, 2004
113. Kahnert A, Hopken UE, Stein M, Bandermann S, Lipp M, Kaufmann SHE: *Mycobacterium tuberculosis* triggers formation of lymphoid structure in murine lungs. J Infect Dis 195:46, 2007
114. Seiler P, Aichele P, Bandermann S, Hauser AE, Lu B, Gerard NP, Gerard C, Ehlers S, Mollenkopf HJ, Kaufmann SH: Early

granuloma formation after aerosol *Mycobacterium tuberculosis* infection is regulated by neutrophils via CXCR3-signaling chemokines. Eur J Immunol 33:2676, 2003
115. Chakravarty SD, Xu J, Lu B, Gerard C, Flynn J, Chan J: The chemokine receptor CXCR3 attenuates the control of chronic *Mycobacterium tuberculosis* infection in BALB/c mice. J Immunol 178:1723, 2007
116. Bromley SK, Peterson DA, Gunn MD, Dustin ML: Cutting edge: Hierarchy of chemokine receptor and TCR signals regulating T cell migration and proliferation. J Immunol 165:15, 2000
117. Jang S, Uematsu S, Akira S, Salgame P: IL-6 and IL-10 induction from dendritic cells in response to *Mycobacterium tuberculosis* is predominantly dependent on TLR2-mediated recognition. J Immunol 173:3392, 2004
118. Boussiotis VA, Tsai EY, Yunis EJ, Thim S, Delgado JC, Dascher CC, Berezovskaya A, Rousset D, Reynes JM, Goldfeld AE: IL-10-producing T cells suppress immune responses in anergic tuberculosis patients. J Clin Invest 105:1317, 2000
119. Gerosa F, Nisii C, Righetti S, Micciolo R, Marchesini M, Cazzadori A, Trinchieri G: $CD4^+$ T cell clones producing both interferon-gamma and interleukin-10 predominate in bronchoalveolar lavages of active pulmonary tuberculosis patients. Clin Immunol 92:224, 1999
120. Hirsch CS, Toossi Z, Othieno C, Johnson JL, Schwander SK, Robertson S, Wallis RS, Edmonds K, Okwera A, Mugerwa R, Peters P, Ellner JJ: Depressed T-cell interferon-gamma responses in pulmonary tuberculosis: Analysis of underlying mechanisms and modulation with therapy. J Infect Dis 180:2069, 1999
121. Barnes PF, Lu S, Abrams JS, Wang E, Yamamura M, Modlin RL: Cytokine production at the site of disease in human tuberculosis. Infect Immun 61:3482, 1993
122. Fulton SA, Cross JV, Toossi ZT, Boom WH: Regulation of interleukin-12 by interleukin-10, transforming growth factor-beta, tumor necrosis factor-alpha, and interferon-gamma in human monocytes infected with *Mycobacterium tuberculosis* H37Ra. J Infect Dis 178:1105, 1998
123. De La Barrera S, Aleman M, Musella R, Schierloh P, Pasquenelli V, Garcia V, Abbate E, Sasiain M, Del C: IL-10 down-regulates costimulatory molecules on *Mycobacterium tuberculosis*-pulsed macrophages and impairs the lytic activity of CD4 and CD8 CTL in tuberculosis patients. Clin Exp Immunol 138:128, 2004
124. Rojas RE, Balaji KN, Subramanian A, Boom WH: Regulation of human $CD4^+$ alpha beta T-cell-receptor-positive (TCR^+) and gamma delta TCR^+ T-cell responses to *Mycobacterium tuberculosis* by interleukin-10 and transforming growth factor beta. Infect Immun 67:6461, 1999
125. North RJ: Mice incapable of making IL-4 or IL-10 display normal resistance to infection with *Mycobacterium tuberculosis*. Clin Exp Immunol 113:55, 1998
126. Turner J, Gonzalez-Juarrero M, Ellis DL, Basaraba RJ, Kipnis A, Orme IM, Cooper AM: *in vivo* IL-10 production reactivates chronic pulmonary tuberculosis in C57BL/6 mice. J Immunol 169:6343, 2002
127. Feng CG, Kullberg MC, Jankovic D, Cheever AW, Caspar P, Coffman RL, Sher A: Transgenic mice expressing human interleukin-10 in the antigen-presenting cell compartment show increased susceptibility to infection with *Mycobacterium avium* associated with decreased macrophage effector function and apoptosis. Infect Immun 70:6672, 2002
128. Awomoyi AA, Marchant A, Howson JM, McAdam KP, Blackwell JM, Newport MJ: Interleukin-10, polymorphism in SLC11A1 (formerly NRAMP1), and susceptibility to tuberculosis. J Infect Dis 186:1808, 2002
129. Buettner M, Meinken C, Bastian M, Bhat R, Stossel E, Faller G, Cianciolo G, Ficker J, Wagner M, Rollinghoff M, Stenger S: Inverse correlation of maturity and antibacterial activity in human dendritic cells. J Immunol 174:4203, 2005
130. Bhatt K, Hickman SP, Salgame P: Cutting edge: A new approach to modeling early lung immunity in murine tuberculosis. J Immunol 172:2748, 2004
131. Humphreys IR, Stewart GR, Turner DJ, Patel J, Karamanou D, Snelgrove RJ, Young DB: A role for dendritic cells in the dissemination of mycobacterial infection. Microbes Infect 8:1339, 2006
132. Jiao X, Lo-Man R, Guermonprez P, Fiette L, Deriaud E, Burgaud S, Gicquel B, Winter N, Leclerc C: Dendritic cells are host cells for mycobacteria *in vivo* that trigger innate and acquired immunity. J Immunol 168:1294, 2002
133. Tian T, Woodworth J, Skold M, Behar SM: *in vivo* depletion of $CD11c^+$ cells delays the $CD4^+$ T cell response to *Mycobacterium tuberculosis* and exacerbates the outcome of infection. J Immunol 175:3268, 2005
134. Abadie V, Badell E, Douillard P, Ensergueix D, Leenen PJ, Tanguy M, Fiette L, Saeland S, Gicquel B, Winter N: Neutrophils rapidly migrate via lymphatics after *Mycobacterium bovis* BCG intradermal vaccination and shuttle live bacilli to the draining lymph nodes. Blood 106:1843, 2005
135. Pedrosa J, Saunders BM, Appelberg R, Orme IM, Silva MT, Cooper AM: Neutrophils play a protective nonphagocytic role in systemic *Mycobacterium tuberculosis* infection of mice. Infect Immun 68:577, 2000
136. Tsai MC, Chakravarty S, Zhu G, Xu J, Tanaka K, Koch C, Tufariello J, Flynn J, Chan J: Characterization of the tuberculous granuloma in murine and human lungs: Cellular composition and relative tissue oxygen tension. Cell Microbiol 8:218, 2006
137. Schaible UE, Winau F, Sieling PA, Fischer K, Collins HL, Hagens K, Modlin RL, Brinkmann V, Kaufmann SH: Apoptosis facilitates antigen presentation to T lymphocytes through MHC-I and CD1 in tuberculosis. Nat Med 9:1039, 2003
138. Makino M, Maeda Y, Mukai T, Kaufmann SH: Impaired maturation and function of dendritic cells by mycobacteria through IL-1beta. Eur J Immunol 36:1443, 2006
139. Hanekom WA, Mendillo M, Manca C, Haslett PA, Siddiqui MR, Barry C, III, Kaplan G: *Mycobacterium tuberculosis* inhibits maturation of human monocyte-derived dendritic cells in vitro. J Infect Dis 188:257, 2003
140. Khader SA, Partida-Sanchez S, Bell G, Jelley-Gibbs DM, Swain S, Pearl JE, Ghilardi N, deSauvage FJ, Lund FE, Cooper AM: Interleukin 12p40 is required for dendritic cell migration and T cell priming after *Mycobacterium tuberculosis* infection. J Exp Med 203:1805, 2006
141. Demangel C, Bertolino P, Britton WJ: Autocrine IL-10 impairs dendritic cell (DC)-derived immune responses to mycobacterial infection by suppressing DC trafficking to draining lymph nodes and local IL-12 production. Eur J Immunol 32:994, 2002
142. Flynn JL, Chan J: Immune evasion by *Mycobacterium tuberculosis*: Living with the enemy. Curr Opin Immunol 15:450, 2003
143. Dayaram YK, Talaue MT, Connell ND, Venketaraman V: Characterization of a glutathione metabolic mutant of *Mycobacterium tuberculosis* and its resistance to glutathione and nitrosoglutathione. J Bacteriol 188:1364, 2006
144. Thoma-Uszynski S, Stenger S, Takeuchi O, Ochoa MT, Engele M, Sieling PA, Barnes PF, Rollinghoff M, Bolcskei PL,

Wagner M, Akira S, Norgard MV, Belisle JT, Godowski PJ, Bloom BR, Modlin RL: Induction of direct antimicrobial activity through mammalian Toll-like receptors. Science 291:1544, 2001
145. Nozaki Y, Hasegawa Y, Ichiyama S, Nakashima I, Shimokata K: Mechanism of nitric oxide-dependent killing of *Mycobacterium bovis* BCG in human alveolar macrophages. Infect Immun 65:3644, 1997
146. MacMicking JD, Taylor GA, McKinney JD: Immune control of tuberculosis by IFN-gamma-inducible LRG-47. Science 302:654, 2003
147. Deretic V: Autophagy as an immune defense mechanism. Curr Opin Immunol 18:375, 2006
148. Gutierrez MG, Master SS, Singh SB, Taylor GA, Colombo MI, Deretic V: Autophagy is a defense mechanism inhibiting BCG and *Mycobacterium tuberculosis* survival in infected macrophages. Cell 119:753, 2004
149. Ferrero E, Biswas P, Vettoretto K, Ferrarini M, Uguccioni M, Piali L, Leone BE, Moser B, Rugarli C, Pardi R: Macrophages exposed to *Mycobacterium tuberculosis* release chemokines able to recruit selected leucocyte subpopulations: Focus on $\gamma\delta$ cells. Immunology 108:365, 2003
150. Lockhart E, Green AM, Flynn JL: IL-17 production is dominated by {gamma}{delta} T cells rather than CD4 T cells during *Mycobacterium tuberculosis* infection. J Immunol 177:4662, 2006
151. Mogues T, Goodrich ME, Ryan L, LaCourse R, North RJ: The relative importance of T cell subsets in immunity and immunopathology of airborne *Mycobacterium tuberculosis* infection in mice. J Exp Med 193:271, 2001
152. Saunders BM, Frank AA, Cooper AM, Orme IM: Role of gamma delta T cells in immunopathology of pulmonary *Mycobacterium avium* infection in mice. Infect Immun 66:5508, 1998
153. Munk M, Gatrill A, Kaufmann S: Target cell lysis and IL-2 secretion by gamma/delta T lymphocytes after activation with bacteria. J Immunol 145:2434, 1990
154. Chen ZW: Immune regulation of [gamma][delta] T cell responses in mycobacterial infections. Clin Immunol 116:202, 2005
155. Shen Y, Zhou D, Qiu L, Lai X, Simon M, Shen L, Kou Z, Wang Q, Jiang L, Estep J, Hunt R, Clagett M, Sehgal PK, Li Y, Zeng X, Morita CT, Brenner MB, Letvin NL, Chen ZW: Adaptive immune response of Vgamma 2Vdelta 2^+ T cells during mycobacterial infections. Science 295:2255, 2002
156. Li B, Rossman M, Imir T, Oner-Eyuboglu A, Lee C, Biancaniello R, Carding S: Disease-specific changes in gamma delta T cell repertoire and function in patients with pulmonary tuberculosis. J Immunol 157:4222, 1996
157. Vankayalapati R, Garg A, Porgador A, Griffith DE, Klucar P, Safi H, Girard WM, Cosman D, Spies T, Barnes PF: Role of NK cell-activating receptors and their ligands in the lysis of mononuclear phagocytes infected with an intracellular bacterium. J Immunol 175:4611, 2005
158. Junqueira-Kipnis AP, Kipnis A, Jamieson A, Juarrero MG, Diefenbach A, Raulet DH, Turner J, Orme IM: NK cells respond to pulmonary infection with *Mycobacterium tuberculosis*, but play a minimal role in protection. J Immunol 171:6039, 2003
159. Feng CG, Kaviratne M, Rothfuchs AG, Cheever A, Hieny S, Young HA, Wynn TA, Sher A: NK cell-derived IFN-{gamma} differentially regulates innate resistance and neutrophil response in T cell-deficient hosts infected with *Mycobacterium tuberculosis*. J Immunol 177:7086, 2006
160. Gansert JL, Kiebler V, Engele M, Wittke F, Rollinghoff M, Krensky AM, Porcelli SA, Modlin RL, Stenger S: Human NKT cells express granulysin and exhibit antimycobacterial activity. J Immunol 170:3154, 2003
161. Apostolou I, Takahama Y, Belmant C, Kawano T, Huerre M, Marchal G, Cui J, Taniguchi M, Nakauchi H, Fournie J-J, Kourilsky P, Gachelin G: Murine natural killer cells contribute to the granulomatous reaction caused by mycobacterial cell walls. PNAS 96:5141, 1999
162. Chackerian A, Alt J, Perera V, Behar SM: Activation of NKT cells protects mice from tuberculosis. Infect Immun 70:6302, 2002
163. Aliberti J, Bafica A: Anti-inflammatory pathways as a host evasion mechanism for pathogens. Prostaglandins Leukot Essent Fatty Acids 73:283, 2005
164. Reed MB, Domenech P, Manca C, Su H, Barczak AK, Kreiswirth BN, Kaplan G, Barry CE, III: A glycolipid of hypervirulent tuberculosis strains that inhibits the innate immune response. Nature 431:84, 2004
165. Ribeiro-Rodrigues R, Resende Co T, Rojas R, Toossi Z, Dietze R, Boom WH, Maciel E, Hirsch CS: A role for $CD4^+CD25^+$ T cells in regulation of the immune response during human tuberculosis. Clin Exp Immunol 144:25, 2006
166. Guyot-Revol V, Innes JA, Hackforth S, Hinks T, Lalvani A: Regulatory T cells are expanded in blood and disease sites in patients with tuberculosis. Am J Respir Crit Care Med 173:803, 2006