Supplementary Material (ESI) for Molecular BioSystems

Boolean Transfer Functions for the *Mtb*–host interactome

Supplementary Material

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Summary

In this text, we discuss all the Boolean transfer functions that have been employed in our model of the *Mtb*–host interactome. These Boolean transfer functions represent the inter-dependence of the various nodes in the model on one another. We first discuss the components of innate immunity, followed by the cytokines, the components of the adaptive immune response and finally bacteria, and the bacterial virulence factors.

Critical components of immune system Several components of the immune system are always present, viz. T cells, B cells, mast cells, macrophages, neutrophils, endothelial cells, etc. These are not updated at each step, i.e., they remain 'on', as initialised. Instead, there are activated forms of most of these components, which come into play.

Macrophages are versatile cells found in practically every tissue in the body, where they participate in an overwhelming array of biological processes. They are the sentinels of the immune system [1]. Lymphocytes (B lymphocytes and T lymphocytes) are produced in the bone marrow, and they circulate in the blood and lymphatic systems, and reside in various lymphoid organs [2]. T cells contribute significantly to the anti-mycobacterial adaptive immune response [3]. Neutrophils, a type of granulocytes are produced in the bone marrow and released into the peripheral blood and circulate for a few hours before migrating into the tissues [2].

1 Innate Immunity

Antigen presentation *Antigen_presentation* = Bacteria and Antigen_processing and Random*
Antigen presentation follows antigen processing. There is quite a bit of uncertainty in this process, particularly for *Mtb*, which is encapsulated as 'Random' [3–6]. When a 'Random' element is involved in a transfer function, it is taken as on or off during the simulations based on a uniform random distribution. It can be expected that the 'Random' element would evaluate to 'True', roughly 50% of the time, and 'False' otherwise.

TLR signalling *TLR_signalling* = (Macrophage or Activated_phagocytic_cells or Dendritic_cells or Mast_cells) and ((Bacteria or PIM) and not (ManLAM and Random))*
TLRs stimulate host-defence mechanisms [7]. TLR2 and TLR4 have been implicated in the activation of macrophage by mycobacteria [8]. TLR stimulation in macrophages up-regulates phagocytosis of bacteria and apoptotic cells. Mycobacterial components can activate cells through hetero-dimers of TLR1 and TLR2, as well as through TLR4 and TLR6 [9]. All TLRs except TLR3 signal through the MyD88 pathway, leading to activation of the NF-κB gene transcription program and production of pro-inflammatory cytokines [10]. TLR signalling may be actuated by macrophages, APCs, DCs or mast cells in the presence

of bacteria. PIM promotes this process. It is inhibited by ManLAM, although it is possible that at times, ManLAM may not bind to TLR, which is represented by the 'Random' component [5; 7–12].

CR MR other signalling *CR_MR_other_signalling* = (Macrophage or Activated_phagocytic_cells or Dendritic_cells) and (Bacteria and Random)*
Bacteria can bind to the complement receptors, mannose receptors and other receptors, like the DC-SIGN receptor in case of DCs, which triggers subsequent signalling, the mechanisms of which are not well-characterised [10; 13–17].

signalling molecules *signalling_molecules* = Bacteria and Random*
These signalling molecules are produced in the presence of bacteria, through mechanisms that are not very well-defined [4].

Macrophage *Macrophage* = Macrophage or Th1RC*
Macrophages are always present in the host. Th1RC promotes the influx of macrophages at the site of action [2].

Activated DCs *Activated_Dendritic_cells* = (Dendritic_cells and Bacteria) or Activated_phagocytic_cells or (Dendritic_cells and Bacteria and (Th1RC or Th2RC))*
Immature DCs, upon stimulation by bacteria, get activated and mature in the lymph nodes. APCs, Th1RC and Th2RC also aid in activating DCs [16; 18–20].

Phagocytosis *Phagocytosis* = Bacteria and (Macrophage or Activated_phagocytic_cells or Dendritic_cells)*
Phagocytosis is a type of endocytosis, the general term for the uptake of material from its environment by the cell. Phagocytosis involves the expansion of the cell's plasma membrane around the particulate material, which may include whole pathogenic microorganisms, to form large vesicles called phagosomes [2]. Phagocytosis initiates the innate immune response, which in turn orchestrates the adaptive immune response [21]. Phagocytosis is initiated when bacteria bind to the macrophage, APCs or DCs [2; 4; 5; 20; 22].

Activated phagocytic cells *Activated_phagocytic_cells* = Bacteria and ((Phagocytosis and CR_MR_other_signalling) or Pro_inflammatory_cytokines or TcRC or (CD1r_T_cells and IFN_gamma) or (Macrophage and Chemokine_signalling) or (T_cells and (IL_1 or IL_4 or (IFN_gamma and TNF_alpha) or IL_10 or IFN_alpha or TNF_beta)))*
Phagocytosis of bacteria and the subsequent signalling activates the phagocytic cells. Alternatively, PICs, TcRC, cytokines released by T cells, such as IL-1, IL-4, IL-10, IFN-α, TNF-β and IFN-γ and TNF-α in synergy can activate the phagocytic cells. Chemokine signalling stimulates the macrophage to recruit APCs [2; 4; 14; 15; 23; 24]. The cytokines (IFN-γ) released by the CD1-restricted T cells contribute to the cell-mediated immunity by activating phagocytic cells [25].

PICs *Pro_inflammatory_cytokines* = ((Activated_phagocytic_cells and (TNF_alpha or IL_1 or GM_CSF)) or (Phagocytosis and TLR_signalling and not ManLAM)) and (not (Th2RC or IL_6 or IL_10 or TGF_beta) or Random)*
PICs are secreted on phagocytosis followed by TLR signalling, which is inhibited by ManLAM. PICs may also be secreted by APCs on stimulation by cytokines such as TNF-α, IL-1 or GM-CSF. The production of PICs is inhibited by anti-inflammatory cytokines such as Th2RC, IL-6, IL-10 or TGF-β. A balance between the effects of PICs and anti-inflammatory cytokines is thought to determine the outcome of disease [26]. This is accounted for by the 'Random' factor, which permits the activation of PICs, even in the presence of anti-inflammatory cytokines [5; 10; 27].

ROI *ROI* = Activated_phagocytic_cells and Pro_inflammatory_cytokines and not (SodA or SodC or BpoB or KatG or SecA2 or ManLAM)*
ROIs are released by APCs, under the influence of PICs. The detrimental effect of ROIs on the bacteria is mitigated by the various bacterial defence components, such as SodA, SodC, BpoB, KatG, SecA2 and ManLAM [27–29].

RNI *RNI* = Activated_phagocytic_cells and Pro_inflammatory_cytokines and not KatG*
RNIs are released by APCs, under the influence of PICs. The detrimental effect of RNIs on the bacteria is mitigated by KatG [3; 4; 28; 30].

Cathelicidin *Cathelicidin* = Bacteria and Macrophage and TLR_signalling*
TLR2-mediated activation of macrophages upregulated the expression of Vitamin D receptor and Vitamin-D-1-hydroxlyase genes, leading to induction of antimicrobial peptide, cathelicidin, as well as its co-localisation to intracellular vacuoles containing mycobacterial cells. Cathelicidin significantly inhibits the growth of *Mtb* [20; 31]. Infection of macrophage with bacteria, followed by TLR signalling leads to the production of cathelicidin [32].

Phagolysosome formation *Phagolysosome_formation* = (Bacteria or PIM) and Phagocytosis and (not (ManLAM or PknG or LAM or SapM or Urease) or Random)*
Phagosomes containing viable, virulent mycobacteria show the presence of early endosomal markers such as transferrin receptor, MHC class II molecules, and the ganglioside GM1 and exclude late endosomal markers such as the proton ATPase, mannose-6-phosphate receptor and the lysosomal protease cathepsin D, Rab7, LAMP-1 and, LAMP-2 [33; 34]. Virulent mycobacteria maintain the phagolysosome as a habitable environment by preventing normal vacuole acidification through the exclusion of the vesicular proton-ATPase [28]. Some mycobacterial phagosomes can proceed to develop to the more mature stages of the phagolysosome [7]. The initial analyses of Rabs on mycobacterial phagosomes have indicated that *Mtb* phagolysosome biogenesis arrest occurs between the stages controlled by the early endosomal GTPase Rab5 and its late endosomal counterpart Rab7 [35].

Phagocytosis of bacteria leads to the formation of phagosome, which fuses with the lysosome to form the phagolysosome. PIM enhances this process, while the bacterial virulence factors such as ManLAM, PknG, LAM, SapM and urease inhibit this process. It has been stated that phagolysosome formation may take place despite the inhibitory action of the various factors listed above; this is accounted for by the 'Random' component [20; 33; 34; 36–39].

Antigen processing *Antigen_processing* = ((Macrophage and Phagolysosome_formation and not (Ag85CX or FAP or LP_19kDa or Urease or LprG)) and Random) or (Dendritic_cells and Bacteria)*
Phagolysosome formation in the macrophage leads to antigen processing. This is inhibited by various bacterial virulence factors such as Antigen-85 complex, FAP, 19kDa lipoprotein, urease or LprG. In some cases, even after phagolysosome formation, antigen processing may not happen, which is encoded by using 'and Random', in the transfer function [3; 4; 6; 20; 34; 40; 41]. DCs are also involved in antigen processing. Cross priming of T cells by apoptotic vesicles released from infected macrophages for subsequent uptake and presentation by DCs. This detour pathway includes not only a mechanism of antigen distribution, but describes infection-induced apoptosis as a key prerequisite for CD8+ T cell activation due to the nature of phagosomally enclosed pathogens [42].

2 Cytokines

IL-1 *IL_1* = (Activated_T_cells or Eosinophils or Activated_phagocytic_cells or (Macrophage and Bacteria))*

and not (IL_6 or IL_10 or TGF_beta)
IL-1 is inhibited by various cytokines such as IL-6, IL-10 and TGF-β. It is produced by T cells or macrophages in the presence of bacteria, eosinophils and APCs [2; 4; 5; 28; 43].

IL-2 *IL_2* = Activated_T_cells or Th1_cells or Eosinophils or Activated_Dendritic_cells*
IL-2 is produced by T cells and DCs in the presence of bacteria, as well as the Th 1 cells and eosinophils [2; 4].

IL-3 *IL_3* = Th1_cells or Th2_cells*
IL-3 is produced by both Th1 and Th2 cells [2; 28; 44].

IL-4 *IL_4* = (Activated_T_cells or Th2_cells or Eosinophils or Basophils or Activated_Mast_cells) and not IFN_gamma*
IL-4 is inhibited by IFN-γ. It is produced by T cells and mast cells when exposed to bacteria, as well as the other immune cells, such as eosinophils, basophils and Th2 cells [2; 4; 5].

IL-5 *IL_5* = (Th2_cells or Eosinophils or Activated_Mast_cells) and not IFN_gamma*
IL-5 is also inhibited by IFN-γ. It is produced by mast cells in the presence of bacteria, Th2 cells and eosinophils [2; 4].

IL-6 *IL_6* = Activated_phagocytic_cells or Activated_T_cells or Activated_Dendritic_cells or Eosinophils or Activated_Mast_cells or (Macrophage and Bacteria)*
IL-6 is produced by APCs and eosinophils, as well as macrophages, T cells, DCs and mast cells in the presence of bacteria [2; 4; 5; 28; 43; 45].

IL-8 *IL_8* = Macrophage and (Bacteria or LAM) and TLR_signalling*
IL-8 is produced by macrophages upon stimulation by bacteria or LAM, through TLR signalling [4; 5; 43].

IL-10 *IL_10* = (Th2_cells or Activated_T_cells or Activated_phagocytic_cells or (Activated_Dendritic_cells and ManLAM) or (Macrophage and (Bacteria or ManLAM or LAM))) and not IFN_gamma*
IL-10 is inhibited by IFN-γ. It is produced by Th2 cells and activated T cells and phagocytic cells. It is also produced by activated DCs, where it is actuated by ManLAM. It is also produced by macrophages containing bacteria, where it is actuated by both ManLAM and LAM [4; 5; 19; 43].

IL-12 *IL_12* = Activated_T_cells or (Activated_Dendritic_cells and not (ManLAM or LAM)) or Activated_phagocytic_cells or (Macrophage and (Bacteria or LP_19kDa))*
IL-12 is a regulatory cytokine which connects the innate and adaptive host response to mycobacteria, by activating the naïve T cells [5; 46]. IL-12 is produced by activated T cells, activated DCs, APCs as well as macrophages containing bacteria. The 19kDa lipoprotein enhances the production of IL-12 in macrophages containing bacteria. The production of IL-12 by activated DCs is inhibited by both ManLAM and LAM [4; 5; 11; 19; 20].

IL-13 *IL_13* = Th2_cells or Eosinophils*
IL-13 is produced by Th2 cells, as well as eosinophils [47].

IL-18 *IL_18* = Activated_Dendritic_cells or Activated_T_cells or Activated_phagocytic_cells*
IL-18 is produced by activated DCs, activated T cells and APCs [5].

GM-CSF *GM_CSF* = Activated_phagocytic_cells or Activated_T_cells or Th1_cells or Th2_cells*
GM-CSF is produced by APCs, activated T cells, as well as Th1 and Th2 cells [7; 20; 48].

IFN-α *IFN_alpha* = Activated_T_cells or Activated_Dendritic_cells*
IFN-α is produced by activated T cells or activated DCs [2; 19].

IFN-γ *IFN_gamma* = (Activated_T_cells or Th1_cells or Tc_cells or (CD1r_T_cells and Antigen_presentation) or Activated_Dendritic_cells or Activated_phagocytic_cells or ((Macrophage and Bacteria) and not ManLAM)) and not (TGF_beta and IL_10)*
IFN-γ is a central factor in the activation of anti-mycobacterial activities of macrophages, and thus crucial for protection against tuberculosis [7]. Production of IFN-γ is critical in the control of *Mtb* infection, whether produced early in infection as a by-product of the activation of immune defence mechanisms, or by Ag-specific T cells following the induction of specific immunity [8]. IFN-γ is produced by several cells [2; 7; 28; 49], viz. activated T cells, Th1 cells, Tc cells, activated DCs, APCs and macrophages (where it is inhibited by ManLAM [34]). It is inhibited by TGF-β and IL-10 [4; 5]. CD1-restricted T cells produce IFN-γ upon stimulation with mycobacterial antigens [25; 50].

TNF-α *TNF_alpha* = Activated_Neutrophils or Th1_cells or Tc_cells or Eosinophils or Basophils or Activated_Mast_cells or Activated_T_cells or Activated_phagocytic_cells or (((Macrophage and Bacteria) or LAM or LP_19kDa) and not ManLAM)*
TNF-α leads to the recruitment of monocytes and lymphocytes from the blood and the development of the inflammatory process. It also helps in granuloma formation [28]. TNF-α is required for the induction of apoptosis in response to infection with *Mtb* [5]. TNF-α is produced by various cells, viz. neutrophils, Th 1 and Tc cells, eosinophils, basophils and activated mast [45] and T cells. It is also produced by APCs and macrophages [19; 28]. ManLAM inhibits the production of TNF-α by both APCs and macrophages [11], while LAM and the 19kDa lipoprotein [20; 51] promote production of TNF-α by both APCs and macrophages [2; 4; 5; 7; 43; 49].

TNF-β *TNF_beta* = Th1_cells or Activated_T_cells or Tc_cells*
TNF-β is produced by Th1 cells, Tc cells and activated T cells [7].

TGF-β *TGF_beta* = ((Macrophage and Bacteria) or Activated_phagocytic_cells) and (ManLAM or LAM)*
TGF-β is produced by macrophages and APCs, in the presence of either ManLAM or LAM [2; 4; 5].

Chemokine signalling *Chemokine_signalling* = Bacteria and (Macrophage or Neutrophils or Activated_phagocytic_cells)*
Chemokines are small chemo-attractant cytokines that control a wide variety of biological and pathological processes, ranging from immuno-surveillance to inflammation and from viral infection to cancer [52]. Chemokine signalling here represents the complex signalling mechanisms initiated by chemokines, since the contribution of individual chemokines is difficult to evaluate. Various chemokines such as CCL2, CCL3 and CCL5 are produced, contributing to chemokine signalling, by cells such as macrophages, neutrophils and APCs, in the presence of bacteria [4; 5; 28; 53].

3 Adaptive Immunity

It must be noted that the nodes involved in adaptive immunity are all activated only after a delay of δ_{AI}.

T cells *T_cells* = T_cells or (CD1r_T_cells and IFN_gamma)*
IFN-γ released by CD-restricted T cells enhances T cell proliferation [25].

TC Differentiation *TC_Differentiation* = Bacteria and (Activated_Dendritic_cells and (IL_12 or IL_6 or IL_18 or IFN_gamma))*
This is a critical step in the immune response, promoted by DCs in the presence of cytokines such as IL-12, IL-6, IL-18 or IFN-γ [16; 19].

B cell signalling *B_cell_signalling* = B_cells and Bacteria and Random*
The cytokines released through this process play a role in the activation of T cells. The mechanism is again not very well understood [24].

Activated T cells *Activated_T_cells* = Bacteria and ((Antigen_presentation and Phagocytosis) or Activated_phagocytic_cells or (Activated_Dendritic_cells and TC_Differentiation) or (B_cells and B_cell_signalling) or (Neutrophils and TNF_alpha) or (T_cells and (IL_2 or IL_4 or IL_6 or IFN_gamma or IFN_alpha)))*
This is another critical step in the adaptive immune response. Naïve T cells are activated when they recognise an antigen-MHC complex on an appropriate antigen presenting cell or target cell. Activation depends on a signal induced by engagement of TCR complex and a co-stimulatory signal induced by the CD28-B7 interaction [2]. Upon antigen presentation, the naïve T cells get activated. DCs also play a major role in the activation and differentiation of T cells. The various cytokines release by the T cells, viz. IL-2, IL-4, IL-6, IFN-γ and IFN-α activate the T cells in an autocrine fashion. Neutrophils, in the presence of TNF-α, B cells, on signalling, and APCs also play a role in the activation of T cells [20; 54].

Th1 cells *Th1_cells* = (Bacteria and (T_cells and (IL_12 or IL_18))) or (Macrophage and Chemokine_signalling)*
Th1 cells are important in the control of tuberculosis infection as they produce the cytokines IFN-γ and TNF-α [28; 40]. T cells in the presence of bacteria, upon stimulation by IL-12 or IL-18 differentiate into Th1 cells. Macrophages, in the presence of chemokines increase the population of Th1 cells [7; 19; 52; 55].

Th2 cells *Th2_cells* = (Bacteria and T_cells and IL_4) or (Macrophage and Chemokine_signalling)*
Th2 cells are a type of effector T cells, which are usually characterised by less stringent activation requirements, increased expression of cell adhesion molecules and production of soluble effector molecules [2]. T cells in the presence of bacteria, upon stimulation by IL-4 differentiate into Th2 cells. Macrophages, in the presence of chemokines increase the population of Th2 cells [7; 19; 52; 55].

Tc cells *Tc_cells* = Activated_T_cells or Th1RC*
CD8+ T cells (Tc cells or cytotoxic T cells) have been suggested to play a special role in the human immune response to *Mtb* by injecting anti-mycobacterial effector molecules such as granulysin into the target cell [6].

T cells can either differentiate into Th or Tc cells; the differentiation into Tc cells is promoted by Th1RC, as well as DCs, on antigen presentation [16; 56].

CD1-restricted T cells *CD1r_T_cells* = T_cells and Bacteria and Random*
The lipid antigen presenting molecule, CD1 stimulates a repertoire of unique CD1-restricted T cells. These cells appear to go through processes of negative and positive selection in the thymus similar to MHC-restricted T cells. Some CD1-restricted T cells have been found to possess the co-receptors CD4 or CD8, while other CD1-restricted T cells have been found to be double negative for the CD4 and CD8 co-receptors [50].

Th1RC *Th1RC* = (Th1_cells and IL_12) and not Th2RC*
Th1RCs are produced by Th1 cells in the presence of IL-12. Th2RCs inhibit Th1RCs [2; 4; 7; 39].

Th2RC *Th2RC* = ((Th2_cells and IL_4) and not Th1RC) or Activated_Mast_cells*
Th1RCs are produced by Th2 cells in the presence of IL-4. Th1RCs inhibit Th2RCs. Activated mast cells also produce Th2RCs [2; 4].

TcRC *TcRC* = Tc_cells and (Th1RC or (Activated_Dendritic_cells and Antigen_presentation))*
TcRCs are produced by Tc cells on stimulation by Th1RCs. Antigen presentation by activated DCs also induces the production of TcRCs [2].

Eosinophils *Eosinophils* = T_cells and Chemokine_signalling and (IL_3 or GM_CSF)*
Eosinophils are recruited by T cells, on chemokine signalling and the cytokines IL-3 or GM-CSF [16; 57].

Basophils *Basophils* = T_cells and Chemokine_signalling and (IL_3 or IL_5 or GM_CSF)*
Basophils are recruited by T cells, on chemokine signalling and the cytokines IL-3, IL-5 or GM-CSF [55; 57].

Activated Neutrophils *Activated_Neutrophils* = Neutrophils and Bacteria and (signalling_molecules or (T_cells and (IL_4 or IL_8 or IFN_gamma or ((TNF_alpha or TNF_beta) and IL_1))))*
Neutrophils are always present in circulation; they are activated in the presence of bacteria, on stimulation by various signalling molecules, and by cytokines released by the T cell, such as IL-4, IL-8, IFN-γ, or the synergistic action of IL-1 and TNF-α or TNF-β [4; 24; 58].

Activated Mast cells *Activated_Mast_cells* = Mast_cells and (Bacteria or (IL_4 or IL_5 or IL_13) or TLR_signalling)*
Mast cells are inflammatory cells typically found in relatively large numbers in the mucosa of the respiratory, gastrointestinal and urinary tracts and near blood or lymphatic vessels [45]. Bacteria, TLR signalling and cytokines such as IL-4, IL-5 and IL-13, act as stimulants of mast cells [45].

Apoptosis *Apoptosis* = ((Bacteria and Macrophage and TNF_alpha) and not (IL_10 or (RNI and PknE))) or ((Fas_FasL_pathway or Perforin_Granulysin) and not (NuoG or ManLAM))*
Apoptosis is a type of programmed cell death involving a series of biochemical events leading to characteristic cell morphology and death. Apoptotic cell death is characterised by several cellular changes, including loss of membrane symmetry and mitochondrial potential, membrane blebbing, and rapid and profound nuclear damage resulting in chromatin condensation and nuclear fragmentation [59]. Apoptosis is controlled by a complex machinery comprising various cellular components. Macrophage infected with bacteria can undergo apoptosis on stimulation by TNF-α. Apoptosis is inhibited by IL-10 and PknE, which responds to the nitric oxide stress in macrophages. Apoptosis can also happen as a result of the Fas-FasL pathway or the production of Tc cell related cytotoxins, viz. perforin and granulysin. NuoG and ManLAM block the initiation of apoptosis through either of these mechanisms [5; 34; 49; 60–63].

Perforin Granulysin *Perforin_Granulysin* = ((Tc_cells and ((IL_2 and IL_6) or IL_1)) or CD1r_T_cells) and Antigen_presentation*
These are Tc cell related cytotoxins, that are produced by Tc cells on antigen presentation, followed by the stimulation through IL-1 or the cytokines IL-2 and IL-6, in synergy [49]. The lysis of target cells by CD1-restricted T cells depends on the release of granules like perforin and granulysin [25].

Fas-FasL pathway *Fas_FasL_pathway* = ((Tc_cells and (IFN_gamma or IL_2)) or CD1r_T_cells) and Antigen_presentation*
During the Fas-based cytotoxic response, the cytotoxic cell produces FasL upon recognition of the target cell. FasL on the cytotoxic cell cross-links the Fas receptor on the target cell and induces the intrinsic suicide program of the target cell. Each FasL trimer binds three Fas receptor molecules on the surface of the target cell. The complex of Fas receptor, FADD (cytosolic adapter protein) and caspase-8 is called the Death Inducing Signaling Complex (DISC). Self-activation of caspase-8 activates downstream caspases, committing the cell to apoptosis [64].

This pathway is initiated by antigen presentation to Tc cells, followed by the stimulation through either of the cytokines, IFN-γ or IL-2 [49]. CD1-restricted T cells lyse target cells through the Fas-FasL pathway [25].

Inflammatory molecules *Inflammatory_molecules* = (Eosinophils or Basophils or Activated_Mast_cells) and (Chemokine_signalling or (IL_8 or IL_3 or IL_1 or GM_CSF))*
Eosinophils, basophils and activated mast cells, produce on stimulation by chemokine signals or cytokines such as IL-8, IL-3, IL-1 and GM-CSF, various inflammatory molecules, which may include granules, ROIs and cytokines [45].

Endothelial cells *Endothelial_cells* = Endothelial_cells or (Activated_phagocytic_cells and (TNF_alpha and IL_1))*
Endothelial cells are always present; they are activated by the cytokines TNF-α and IL-1, released by APCs [4].

Fibroblasts *Fibroblasts* = Activated_phagocytic_cells and ((TNF_alpha and IL_1) or TGF_beta or Chemokine_signalling)*
Fibroblasts are recruited by APCs, on stimulation by cytokines such as TGF-β or TNF-α and IL-1 in synergy or chemokine signalling [4; 26].

4 Bacterial Virulence Factors

Bacteria *Bacteria* = ((Bacteria and (Macrophage or Activated_phagocytic_cells)) and not (ROI or RNI or Cathelicidin or Inflammatory_molecules)) or (Bacteria and not (Phagolysosome_formation or Apoptosis))*
Bacteria on the right hand side of the transfer function imply the need for bacteria in a previous run, if there are to be bacteria in the current run. Bacteria remain viable in the macrophage or APCs in the absence of phagolysosome formation, apoptosis, or molecules such as ROIs, RNIs, cathelicidin and inflammatory molecules [30; 32; 57; 62; 65; 66].

ManLAM *ManLAM* = Bacteria and ManLAM*
The abundance of ManLAM on the surface of *Mtb* would be a determinant for the outcome — survival versus intracellular killing of mycobacteria [34]. ManLAM significantly interferes with the host defence mechanisms, like phagosome maturation arrest, scavenging free oxygen radicals, and directly inhibiting macrophage response and TNF-α and IFN-γ production in macrophages [11; 20; 34]. It is an important virulence factor of *Mtb*, that plays a crucial role in defending against the various immune mechanisms of the host. It is always present in the pathogen.

LAM *LAM* = Bacteria and LAM*
LAM is a phosphatidylinositol-anchored lipoglycan composed of a mannan core with oligoarabinosyl-containing side chains with diverse biological activities [11].

PknG *PknG* = Bacteria and Phagocytosis*
PknG, a protein kinase affects the intracellular traffic of *Mtb* in macrophages. PknG is released by the bacteria within the macrophage cytosol by an unknown mechanism and can be efficiently inhibited by specific kinase inhibitors. Since the kinase activity of PknG is absolutely required for its activity in blocking lysosomal delivery, PknG presumably functions through the phosphorylation of a host factor, thereby preventing its normal function in phagosome-lysosome fusion [9; 37].

PknE *PknE* = Bacteria and PknE*
PknE, a serine/threonine kinase, is important for the survival of *Mtb*. It prevents apoptosis by interfering with the host signalling pathways [61].

SapM *SapM* = Bacteria and SapM*
SapM, a PI3P phosphatase, is involved in the PI3P depletion at the mycobacterial phagosome, thus blocking the association of FYVE proteins with phagosomes [9; 39].

19kDa Lipoprotein *LP_19kDa* = Bacteria and LP_19kDa*
19kDa lipoprotein, anchored in the cell wall of *Mtb*, has been implicated in various immunological responses [3; 11; 34]. 19kDa lipoprotein interacts with host APC via TLR1 and TLR2, leading to antigen processing and MHC II expression, turning what is normally regarded as a pro-inflammatory pathway into an anti-inflammatory one [12; 56].

Ag85CX *Ag85CX* = Bacteria and Ag85CX*
Ag85 complex (Ag85 A, B, C) demonstrate varying degrees of fibronectin binding and have been suggested to play an important role in macrophage uptake of the mycobacteria [28].

FAP *FAP* = Bacteria and FAP*
The attachment and internalisation of several mycobacterial species to their host cell is dependent on bacterial attachment to fibronectin, and FAP (Rv1860) has been proposed as the bacterial mediator of this process [40].

Urease *Urease* = Bacteria and Phagocytosis*
Mycobacterial urease, an enzyme that hydrolyses urea to carbon dioxide and ammonia, has the potential to be active within the host cell, thereby leading to inadequate acidification of the MHC class II compartment and processing of class II complexes [67]. Ammonia generated by the action of urease may be of importance in alkalinising the micro-environment of the organism and in preventing phagosome-lysosome fusion. Urease may provide a source of nitrogen for biosynthesis [33; 34]. Urease is expressed only after bacteria undergo phagocytosis [67].

PIM *PIM* = Bacteria and PIM*
PIM is present on the cell surface of *Mtb* [68]. Mycobacterial pro-inflammatory PIM induce the fusion of granuloma macrophage into multi-nucleated giant cells [69].

NuoG *NuoG* = Bacteria and NuoG*
nuoG of *Mtb*, which encodes a subunit of the type I NADH dehydrogenase complex, is a critical bacterial gene for inhibition of host cell death [60].

LprG *LprG* = Bacteria and LprG*
LprG, a 24kDa lipoprotein found in the *Mtb* cell wall, is a TLR2 agonist [20; 41].

BpoB *BpoB* = Bacteria and Activated_phagocytic_cells*
BpoB, a peroxidase enzyme, is involved in the neutralisation of reactive radicals [29]. BpoB is expressed only after the bacteria undergo phagocytosis [29].

SodA *SodA* = Bacteria and Activated_phagocytic_cells*
SodA (Fe), a superoxide dismutase enzyme, is among the major extracellular proteins released by *Mtb* during growth. It is exported in an active form via a signal peptide-independent pathway that has not been fully characterised [30].

SodC *SodC* = Bacteria and Activated_phagocytic_cells*
SodC (Cu-Zn), also a superoxide dismutase enzyme, is essential for survival of *Mtb* in macrophages [29; 30]. SodC is expressed only after bacteria undergo phagocytosis [29].

KatG *KatG* = Bacteria and Activated_phagocytic_cells*
KatG, catalase-peroxidase-peroxynitritase enzyme converts hydrogen peroxide to water and oxygen and can also break down peroxynitrate, which is a dangerous reaction product of superoxide and nitric oxide. KatG is expressed only after the bacteria undergo phagocytosis [27].

SecA2 *SecA2* = Bacteria and Activated_phagocytic_cells*
SecA2 protein of *Mtb* is an accessory secretion factor that promotes secretion of a subset of proteins that include superoxide dismutase (SodA) and catalase peroxidase (KatG). SecA2 is expressed only after the bacteria undergo phagocytosis [9; 27].

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