Invited Review

Immunology of Tuberculosis:
Clinical and Research Perspectives

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BACKGROUND

The interest in tuberculosis (TB) must not wane especially when we look at the statistics currently available. It is estimated that one-third of the world’s population is infected with Mycobacterium tuberculosis (M. tuberculosis)1. In Africa and Asia, the annual rate of infection is about 2-3%. It is similar in Malaysia where TB remains a major cause of morbidity and mortality. Tuberculosis remains an enigma either from a clinical viewpoint or its immunopathogenesis as to how to contain the organism. It has remained with us from the dawn of history, recognized in skeletons from the stone age and in mummified corpses in the old kingdoms of Egypt, spreading in Western Europe in the Middle ages after the plague and becoming an epidemic during the era of urbanization and industrialization in the 18th and 19th centuries2.

Is there a better strategy than an epidemiological method to control TB? Is complete eradication of TB, as seen in the case of smallpox really an attainable goal? Is anti-tuberculous therapy adequate although the decline of TB in the western world in the late 1940s was attributed to the discovery, development and widespread use of anti-tuberculous drugs? The currently used vaccine, the attenuated Mycobacterium bovis (BCG) does not consistently prevent pulmonary infection although it does reduce fatal disseminated tuberculous diseases3. As BCG is less effective in preventing late reactivation and pulmonary TB, BCG immunization has not contributed to controlling the spread of TB3.

The efficacy of BCG vaccination against adult tuberculosis remains controversial; it is this very group that the prevalence is highest and disease most infectious, being a post-primary type of TB. In large case-controlled and randomized placebo-controlled trials, the protection conferred by BCG has varied enormously in different areas, ranging from nil to nearly 80% in the British MRC trial4,5. As in most old diseases which persisted, there are more questions than answers. The inability to control tuberculosis despite the availability of effective relatively inexpensive therapy is one of the greatest failures of our time as had been highlighted by Starke et al. 20046.

There is a need to have a hard re-look at pathogenesis especially immunopathogenesis of tuberculosis. An explanation as to how mycobacteria adapt and evade the host immune system still eludes us. Understanding its immunopathogenesis would lead towards better diagnostic methods, preventive measures as in a newer vaccine better than BCG, and of course more effective therapy, developing newer biologicals and drugs that require shorter duration of treatment, thus reducing patient non compliance.
Immunity to *M. tuberculosis* infection is like a double-edged sword denoting a delicate balance between protection and disease. At one end, the immune system could protect the host by containing the infection and on the other hand it causes tissue damage propagating disease. Thus the spectrum of manifestation of infection of the host after exposure to *M. tuberculosis* is quite diverse ranging from exposed but uninfected, latent infection (infected but asymptomatic) and disease (infected and symptomatic). This variety of clinical presentations is the result of interactions of (a) the organism, (b) the host, and (c) the environment.

This article shall cover immunological aspects of tuberculosis emphasizing current clinical and research issues. A brief account of the organism, the host, environment and zoonotic aspects that relate to the transmission and control of the disease is also included.

**ORGANISM**

The determining factors that influence disease outcome include the the size of the inoculum and the strain especially if pathogenic. The larger the size of the inoculum, the more likely it is to overwhelm the immune system and develop disease. *M. tuberculosis* has adapted to evading the host immune responses and has the ability to remain latent in the host tissue itself. Pathogenic *M. tuberculosis* variants are more efficient at evasion than the less pathogenic variety.

Variations of strains are known to have different impacts on the host; variation of strains is the result of repetitive DNA sequence and insertional elements which contribute to its DNA polymorphism. As an example, an investigation into a outbreak of newly identified genetic strain (now known as CDC1551) had an unusually high transmission in humans and was significantly more virulent in animal models than other clinical isolates[6]. In the study of McShane et al. H878 strain was found to be hypervirulent and mice infected with this strain failed to induce a Th1-type immune response with lower IFN γ and TNFα in lungs[6]. The development of Th1-type immune response is essential for protective immunity against *M. tuberculosis* which is an intracellular pathogen residing in a macrophage. Activation of macrophages, primarily by IFNγ is central to protective immune response in humans and mice. If sequence diversity results in differing ability to induce a host Th-type response, then this may be the explanation of differences in pattern of disease.

**THE HOST**

Host immunity is a paramount determinant of the spectrum of disease manifestation that would ensue. Immunosuppressed states favour disease especially the severe forms. For this reason, infants, malnourished states, and secondary immune deficiencies (HIV infections, transplant/cancer patients on cytotoxics) tend to suffers disseminated tuberculosis including tuberculous meningitis. Primary immunodeficiencies (e.g. SCID, chronic muco cutaneous candidiasis, antibody deficiency & Hyper IgM) found locally are also not exempted. (unpublished data).

Host defence could also be genetically determined. Individuals with HLADR2 are prone to progression of disease when infected while those with HLA DRB1*1502 do not[7]. There are recent findings to show the existence of a deficient immune response specific to *M.
Tuberculosis and related organisms, akin to TB specific immune deficiency. In these individuals, there are gene defects making them susceptible to tuberculosis. Amongst them are: (i) Gamma Interferon (IFN γ) receptor deficiency (IFNγR1, IFNγR2); (ii) impairment IFN-γ mediated immunity viz Interleukin 12 deficiency (IL12-p40); and (iii) IL12β1 receptor deficiency[8].

ENVIRONMENT

Poverty, over-crowding, poor sanitation and illiteracy remain the other factors in the underdeveloped world that prevent control of TB. Illiteracy and poverty are interrelated, both of which could lead to nutritional deficiency and with it, impairment of host defence. We have seen the relationship between malnutrition and TB. The main defect in protein calorie malnutrition is impairment of T cell immunity besides defects in humoral and cytokine path ways[34]. Host resistance against intracellular organisms (such as M. tuberculosis) is dependant on T cell immunity, hence the explanation of malnutrition predisposing one to TB. Meanwhile over-crowding favours transmission of the bacilli, increasing exposure leading to disease. Overcoming these limitations of the environment would contribute to successful control of TB.

IMMUNOPATHOGENESIS OF TUBERCULOSIS

Protective immunity to tuberculosis is still not completely known although cellular immune response plays a dominant role which is dependant on interplay between phagocytic cells of the monocytic lineage and T lymphocyte[9]. Initial exposure stimulates an immune response which serves to restrict growth and spread. If growth continues, M. tuberculosis disseminates via lymphatic and blood, setting up new foci of infection in other tissues. In other words, the pathogenesis of infection with M. tuberculosis is the consequence of the inability of the macrophage to contain the mycobacteria and the failure of T cells to provide uniformly prolonged protection[10].

The major access to the host is via inhalation; almost all infection by M. tuberculosis is mediated through droplet nuclei (aerosolised droplet) which is dry while being air-borne but able to reach terminal air passage. A cough from a diseased individual produces about 3000 droplets which is the equivalent to an exhalation of a diseased individual talking for 5 minutes; the room is still contagious even after the affected diseased individual has left the room[11]. As few as 5 bacilli is enough to establish a primary focus which is initially seen as multiplication of bacilli in the host alveolar macrophages

Pre immune Pulmonary Defense

Alveolar macrophages form the first pulmonary defence phagocytosing the inhaled Mycobacteria tuberculosis and killing the microbes (Figure 1). Nonetheless alveolar macrophages are not efficient in killing as other mononuclear macrophages thus mycobacteria may survive and multiply. Lysis and death of alveolar macrophages as a result of increasing bacterial load frees the mycobacteria which are ingested by mononuclear phagocyte and dendritic cells which also act as antigen presenting cells. Dendritic cells, being migratory, appear to play a critical role in dissemination[12]. In the macrophage, the bacilli in endocytic
vacuoles of macrophage cause fusions of phagolysosome which provide an environment adverse to the pathogens which being acidic in nature contain reactive oxygen intermediates (ROI), lysosome, and toxic peptides.

**ANTIGEN SPECIFIC IMMUNE RESPONSES (MAINLY IN LYMPH NODES)**

*Primary Response*

Macrophages containing mycobacterium migrate to the regional lymph nodes where interaction of naïve CD4 with antigen presenting cells (macrophage or dendritic cells) cause proliferation of antigen specific CD4 cells which can differentiate into Th1 or Th2 cells according to the cytokine they produce. The CD4 (Th1) type response is characterized by the release of IL 2, IFN γ, IL12, IL18 and TNF α of which IFN γ predominantly activate macrophages to kill intracellular mycobacteria limiting spread of the bacilli. Most infected children with Th1 response are associated with hilar adenopathy and pulmonary infiltrate on the chest X ray\[13\]. Th1 responses are associated with resistance whereas predominant Th2 responses are associated with progress of disease with severe manifestation including pulmonary and even disseminated forms of tuberculosis *viz* miliary form and tuberculous meningitis\[13\]. The CD4 Th2 responses are associated with release of IL4, IL5 and IL10.

*Secondary Response*

Persistence of mycobacteria induces the development of secondary response whereby memory T cells are formed which further augments the ongoing cell mediated immunity. These *M. tuberculosis* reactive CD45 RO/memory T cells are responsible for delayed type hypersensitivity (DTH) which contribute to a positive tuberculin reaction in some\[12\].

Containment of mycobacteria may not be complete and dissemination to the rest of the tissues of the body occurs. The mycobacteria infected dendritic cells being migratory in nature provide the means for dissemination of the bacilli.

In some, attempts at containment are reflected by granuloma formation in tissues\[14\]. This occurs when mycobacteria in a macrophage persist causing chronic stimulation of T cells and its concomitant release of cytokines. This environment induces the macrophage to develop into epitheloid cells which may form multinucleate giant cells. This aggregate of epitheloid cells with surrounding CD4 and CD8 T cells form a granuloma which is dependant on TNF α released by activated macrophage and antigen specific T cells\[9\]. The centre of the granuloma may undergo necrosis with loss of cellular definition and formation of amorphous caseous material. Residual extracellular mycobacteria have difficulty replicating in this anaerobic environment. However, depression of cellular immunity may cause *mycobacteria* to multiply with reactivation of the disease\[9\].

*Other Lymphocyte Subsets*

Varieties of T cell subsets (other than CD4) also contribute to integrated responses to mycobacteria as inferred in studies of gene knockout (GKO) mice. They include T cells bearing αβ –T cell receptors (TCR) CD8 + phenotype(CD4 cells also bear αβ –T cell receptors ), γδ T cells and CD1 restricted T cells.
**Figure 1.** Stages of immune activity in tuberculosis: antigen specific, cell mediate response to mycobacteria

**CD8 + T cells**

The presence of high frequency of IFN γ-secreting CD8 T cells in healthy individuals exposed to TB may indicate its positive role in protectivity. Furthermore, these subsets have been known to persist for more than 2 years in such patients. It is thought that CD8 cytotoxic T cells contribute to killing of contained *mycobacteria* via granule mediated pathway whilst the cytokine IFN γ secreted, activates macrophage to kill its intracellular *mycobacteriae*\(^{[15]}\).

The incubation period from the period of exposure and inhalation until skin sensitivity (tuberculin sensitivity ) is between 3 weeks to 3 months (incubation period is shorter with a large inoculum)\(^{[2]}\).

The advent of tuberculin sensitivity may indicate development of DTH with its associated inflammatory response and tissue damage. Our own study implicated T cells as contributory to skin tuberculin sensitivity (Mantoux) whereby a population of T (CD3) cell subset in peripheral blood lymphocytes of Malaysian children with tuberculosis correlated with the degree of tuberculin sensitivity (Figure 2: unpublished data).

**γδ T cells**

This subset may play a role in early response to *Mycobacterium*. This population is present in sufficient numbers in naïve tuberculin negative individuals and gives measurable *in vitro* proliferative response to *mycobacteriae*\(^{[12]}\). Like CD8+ T cells, γδ T cells are capable of lysing infected macrophage\(^{[7]}\) by granule pathway and reducing the viability of contained bacteria\(^{[15]}\). This subset also secretes cytokine IFN γ, IL2, IL4, IL10 and is activated during

![Figure 2](image-url)

**Figure 2.** Scatter plot of T (D3) cells vs tuberculin skin sensitivity in Malaysian children

*Source:* Lokman M Noh.
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pulmonary TB, increasing in frequency but declining after treatment\textsuperscript{[15,16]}. Subsequently $\alpha\beta$ T cells are attracted to site of infection as a result of chemo-attractant secreted by $\gamma\delta$ T. Cytokines activate $\alpha\beta$ T\textsuperscript{[13]} following which clones of reactive memory T cells develop (CD45RO cells) into Th1 type lymphocytes. Memory T cells are often associated with tuberculin positivity and with it, decreased risk of exogenous reinfection\textsuperscript{[13]}.

\textbf{CD1 Restricted T Cells}

CD1-restricted T cells recognizing mycobacterial lipids (mainly cell wall components) have been identified in humans but not in mice\textsuperscript{[12]}. The different forms of CD1 survey different intracellular compartments and so encounter products of \textit{M. tuberculosis} in different ways\textsuperscript{[15]}.

\textbf{Macrophage Killing Mechanism}

Although IFN $\gamma$-induced killing of macrophages containing \textit{M. leprae} is via oxidative burst mechanism, it cannot be said that similar mechanisms adequately explain killing of \textit{M. tuberculosis} infected macrophages. Although reactive N metabolite is implicated in killing of activated macrophage in mouse, its corollary in humans remains controversial because of its inability to reproduce a similar \textit{in vitro} culture system that reproducibly demonstrates high output nitric oxide (NO) production in human macrophage\textsuperscript{[17]}. Destructive tissue responses reflect active immune responses that fail to eliminate \textit{M. tuberculosis} as a result of recognition of structural rather than secreted mycobacterial protein. Predominant Th2 response may cause severe manifestation of disease including progressive pulmonary TB and miliary /tuberculous meningitis\textsuperscript{[13]}.

\textbf{CYTOKINES AND PATHOGENESIS OF TUBERCULOSIS} \textsuperscript{17}

The interaction of T cells, macrophage and NK cells elaborates cytokines that ultimately leads to recruitment and activation of macrophages\textsuperscript{[12]}. The presence of \textit{M. tuberculosis} in macrophages stimulates it to produce IL1, TNF $\alpha$, IL12, and IL18. These cytokines induce NK cells, $\gamma\delta$ T, CD4 and CD8 T cells to produce IFN $\gamma$ which is microbicidal to intracellular Mycobacteria besides enhancing release of IL12 and TNF$\alpha$ by macrophages. The role of IFN $\gamma$ and IL12 should not be underestimated in pathogenesis; it is reported repeatedly that low production of IFN $\gamma$ and IL12 is often observed in patients with active TB\textsuperscript{[10]}. Meanwhile IL1 and TNF $\alpha$ contribute to formation of granuloma.

It is known that 60\% of active pulmonary TB is associated with immunosuppression as evidenced by suppression of \textit{in vitro} T cell response to \textit{M. tuberculosis} antigen. This suppression of T cell response correlates with the extent of tuberculosis radiologically, and is associated with loss of of cutaneous DTH response (i.e. PPD skin test). The molecular basis of suppression during active tuberculosis has been identified to be predominantly transformation growth factor(TGF) $\beta$ \textsuperscript{[10]}. Monocytes from patients with tuberculosis produced an increased amount of this immunosuppressive cytokine TGF $\beta$; neutralizing of TGF $\beta$ enhances \textit{M. tuberculosis} induced IFN $\gamma$ production\textsuperscript{[18]} TGF $\beta$ is a strong inhibitor of endothelial and epithelial cell growth and in both promotes production of collagen matrix and enhances tissue degradation through production of macrophage collagenase\textsuperscript{[14]}. In experimental animals, systematic administration of TGF$\beta$ results in cachexia and fibrosis\textsuperscript{[19]}.
**IL 6 and IL10: Other Cytokines Associated with Immunosuppression**

IL6 has a potential role of suppression of T cell responses\[^{35}\]. IL6 reduces binding of TNF α to murine macrophages and antagonizes antimycobacterial activity of TNF α in macrophages infected with *M. avium*. Addition of IL6 to human monocytes also enhances intracellular and extracellular mycobacterial growth\[^{36}\].

IL 10, an anti-inflammatory cytokine produced by by macrophage and T cells during *M. tuberculosis* infection, possesses macrophage deactivating properties, including down regulation of IL12 leading to a decrease in IFN γ by T cells. IL10 inhibits CD4 T cell response either directly or by inhibiting antigen presenting cell function (APC) of cells infected with mycobacteria\[^{37}\].

An increased mycobacterial load in the host often leads to a state of immunosuppression leading to severe manifestation of disease with reduced tuberculin sensitivity. This is the reason why the Mantoux test lacks sensitivity and specificity, and that not all TB diseases show a positive Mantoux reaction.

**ISSUES IN PATIENT CARE**

**Tuberculosis in Children**

Tuberculosis in children although less frequent than in adults remains a grave concern as the mortality and morbidity are higher, especially in the first 5 years\[^{20}\]. The younger the child the higher the risk of disease when exposed. As much as 43 % of children younger than 1 year, 25 % between 1-5 years and 15 % at adolescence develop active disease at some time after primary infection\[^{13,21}\]. Compared to adults, the risk is 5-20%. As high as 60% of children, younger than 5 years, develop tuberculous meningitis as a complication\[^{21}\] more frequently than in adults.

The likely explanation for this is a different immune response to *M. tuberculosis* in individuals at different ages\[^{12,22}\]. There exists a qualitative difference in alveolar macrophage in which there is reduced ability to kill intracellular organisms which is more pronounced especially in neonates and children than in adults. Furthermore chemotaxis reaches adult level only by age 6-10 years. There are more naïve T cells in children (90%) compared to adults (60%) which results in poor response to presented antigens and less competency in cytokine production compared to memory T cells (compared to adults, IFN gamma 10%; TNF 25-50 % of adult’s capabilities)\[^{22,23,56}\].

It is therefore an imperative to diagnose TB in children early and to treat them adequately. However, one faces uncertainty for the gold standard for diagnosis of TB which is a positive culture or a positive AFB is 10- 50% \[^{24,25}\] (as low as 10 % in a Malaysian series\[^{19}\]). One often considers (a) clinical symptoms (b) characteristic radiological changes, and (c) a positive tuberculin test as a basis for firm diagnosis of childhood TB. The presence of exposure to a TB individual strengthens the diagnosis. However, in a few occasions fewer than two of those three parameters are positive, and not to treat such children is not without dire consequences for they may develop complications of tuberculosis.

The onus then is to develop better diagnostic methods. There is a need to extend beyond tuberculin test(sensitivity to PPD) to diagnose tuberculous infection. Many factors contribute
to its sensitivity making it a test of moderate sensitivity and specificity; 30% of TB children are Mantoux negative\textsuperscript{[20]}; equally disturbing is that a false positive occurs due to a previous BCG, or exposure to non tuberculous mycobacteria infection.

Efforts are now directed towards specific immune diagnosis in tuberculosis to surpass the low specificity of PPD as a skin test. The greatest drawback of PPD is that most protein components in this substance are shared between mycobacterial species with unrelated bacteria species e.g. BCG, non-tuberculous \textit{Mycobacterium}. This compromises the specificity of the test as previous exposure to BCG or non tuberculous mycobacteria may cause a positive skin test to PPD. To overcome this, there is a need to identify and select the region of \textit{M. tuberculosis} genome that is absent in BCG and non tuberculous mycobacteria in the newer \textit{M. tuberculosis} antigens. Such an antigen has been successfully isolated in the form of low molecular mass antigen viz ESAT-6 \textsuperscript{[25]}. Work has been done to show the potential of such reagents for a more sensitive and specific diagnosis of tuberculosis as either a skin test or \textit{in vitro} testing of cells in blood.

A novel approach has been to use the PPD antigen for an \textit{in vitro} detection of IFN \(\gamma\) secreting lymphocytes sensitized by mycobacterial antigen. The assay is done as a lymphoid stimulation assay with PPD, most conveniently directly in whole blood for 24 h followed by ELISA detection of IFN\(\gamma\) after stimulation. ESAT-6 is now replacing PPD in such an assay and is further modified into an ELISpot testing. This showed higher sensitivity and specificity than the tuberculin skin test \(\gamma\textsuperscript{[26]}\).

\textit{Improving Therapy}

The long duration of anti-TB treatment is often the cause of poor compliance and thus contributing to MDR TB (multiresistant TB). Although no breakthrough into drugs that inactivate \textit{M. tuberculosis} and shorten disease duration is in sight, biologicals viz IFN\(\gamma\) has been used experimentally. Current drugs are efficient in killing actively growing mycobacteria but are less efficient against quiescent non dividing organisms. It is conceivable that this persistence of non dividing bacteria in the host is the reason for the prolonged therapy and the study of their metabolism may therefore identify potential targets for a novel class of drugs. Isocitrate lyase (ICL) has been identified as one such target on the basis of its essential role of persistence of \textit{M. tuberculosis} in murine models\textsuperscript{[27]}. Screening for ICL inhibitors is underway.

The transfer factor (TF) is another candidate which may potentiate standard anti-TB treatment with added advantage of shortening duration. Transfer factor (or leukocyte dialysate) is a low molecular weight (mw) dialyzable product from immune cells that are able to transmit the ability to express delayed type hypersensitivity (DTH) and cell mediated immunity (CMI) from sensitized donor to non immune recipients. Although the exact molecular nature or its action is unknown, studies have found that TF is a low mw protein (<5 kD protein) that can be purified to a high degree of homogeneity\textsuperscript{[28, 29]}.

Studies have also shown the potential benefits of transfer factor in TB, an advancement from previous observed benefits in primary immunodeficiency. Studies in mice, using dialysate (TF) from 21 days \textit{M. tuberculosis} infected mice (coinciding with peak protection and when granuloma reach maturation), have shown increased Th1 responses, IFN \(\gamma\) thus
evoking an efficient reconstitution of cell mediated immunity (CMI) correlating with increased survival of *M. tuberculosis* infected mice\(^{28}\). Such studies also showed protection when transfer factor from healthy individuals with tuberculin reactivity was used. It is conceivable that the benefits of transfer factor will be similar in man, making anti TB therapy duration to less than the conventional 6 months a distinct possibility.

**Towards Defining a Vaccine Better than BCG**

BCG vaccine has its limitations in the prevention of tuberculosis although it is considered still efficacious against the disseminated form and TB meningitis. It has failed to control the increase of new cases worldwide and the disease remains a world problem\(^{30,31}\). There is therefore an urgent need to develop a vaccine better than BCG. The strategy appears to be towards two of the most promising approaches-viz recombinant BCG and DNA vaccine\(^{29}\).

Whole organism vaccine (e.g. BCG) consisting of many antigenic determinants (epitopes) stimulates various immune responses some of which are irrelevant to protectivity in the host. Subunit vaccines appear to be more attractive as they consist of less diverse protein (secreted and non secreted), lipid, and carbohydrate components of *M. tuberculosis* in various formulations. They have the potential to be specific, defined, and safe\(^{32}\). Their disadvantage is limited persistence *in vivo* and whether they can induce lasting cellular immunity or not. Potential immunodominant epitopes of *M. tuberculosis* defined by sera or T cells have been identified. Nonetheless, this subunit proteins have not been able to provide better protection than BCG. Furthermore they are expensive to produce besides requiring multiple boosters\(^{33}\).

**Recombinant BCG**

It is now possible to construct an immunodominant epitope of *M. tuberculosis* and to insert it into the BCG genome; the BCG now acts as a vehicle for the subunit vaccine. Such work is also being done in Malaysia; it involves constructing recombinant BCG containing 2 T cell epitopes of ESAT–6 antigen of *M. tuberculosis*\(^{33}\). Microbial vectors other than mycobacteria, including attenuated strains of Salmonella and vaccinia expressing mycobacterial antigens, are being tested in animal models. The former has the potential to induce mucosal immunity, and the latter to induce CTLs (cytotoxic T lymphocytes)\(^{32}\).

DNA vaccines encoding a variety of *M. tuberculosis* antigens have shown significant protection in mice; in principle it involves direct injection of plasmid DNA encoding specific antigen or epitopes of *M. tuberculosis* that will provide protective immunity\(^{33}\). Advantages include ease of production, the low cost, and the ability to induce long-lasting cellular immune responses. In a recent study, tuberculosis relapse after chemotherapy of infected mice was prevented by post-exposure vaccination with a DNA vaccine encoding the 65-kD antigen; significantly, this effect was not obtained with BCG. These observations suggest the exciting possibility that DNA vaccination might be useful in preventing reactivation of latent tuberculosis – an enormous reservoir of contagion for which no effective or practicable intervention currently exists\(^{32}\).

It is without doubt that the study of immunopathogenesis would not have significantly advanced, if not for the contribution of studies in animals, especially *in vivo* studies which
would have been unethical in humans. Tuberculosis in animals or zoonotic tuberculosis is as relevant to man as in veterinary medicine. One has to be cognizant of the fact that transmission of disease from animals to man and vice versa may become relevant if it is not so presently. A discourse into this subject is both relevant as well as timely.

Zoonotic tuberculosis (TB) caused by *Mycobacterium bovis* is present in animals in most developing countries where surveillance and control activities are often inadequate or unavailable. Therefore, many epidemiologic and public health aspects of this infection remain largely unknown and Malaysia is no exception.

Zoonotic TB is clinically indistinguishable from TB caused by *M. tuberculosis*. In countries where bovine TB is uncontrolled, most human cases occur in young persons and result from drinking or handling contaminated milk. Cervical lymphadenopathy, intestinal lesions, chronic skin TB (*lupus vulgaris*) and other nonpulmonary forms are particularly common. Such cases may, however, be also caused by *M. tuberculosis*. Little is known of the relative frequency with which *M. bovis* causes nonpulmonary TB in developing countries because of limited laboratory facilities for the culture and typing of these bacilli. Nonetheless non tuberculous mycobacterial infection such as *M. bovis* is known to have occurred in cellular immunodeficiencies even in Malaysia (pers. comm.).

Livestock farmers, animal scientists and veterinarians may acquire the disease by inhaling cough spray from infected cattle. They may develop typical pulmonary TB. These patients may in turn infect other animals. However, evidence on human-to-human transmission is limited and anecdotal. Grange and Yates[38] reported survey findings in the United States, Scandinavia and South England whereby approximately half of the post primary cases are pulmonary and the remainder involve other nonpulmonary sites, notably cervical lymph nodes[39]. For reasons that are not clear, the incidence is higher, approximately 20% in ethnic minority populations.

Very limited data on zoonotic TB are available from Asian countries including Malaysia. Much information on the epidemiologic patterns of zoonotic TB has been obtained in this century from developed countries. However, some striking epidemiologic differences related to both animal and human populations in developing countries require particular attention. Only some of these differences are discussed in this article.

**Animal Reservoirs**

The widespread distribution of zoonotic TB in domesticated and wild animals shows an extensive reservoir of this microorganism. The spread of the infection from affected to susceptible animals in both developed and developing countries is most likely to occur when wild and domesticated animals share grazing grounds[39]. Wild animal TB represents a permanent reservoir of infection and poses a serious threat to control and elimination programs.

**Milk Production and Animal Husbandry**

Milk production has increased in most developing countries as a consequence of greater demand for milk for human consumption. This increased demand for milk has led to an increase in the number of productive animals and milk imports and intensification of animal production through the introduction of more productive exotic breeds[40]. In developing
countries, bovine TB infects a higher proportion of exotic dairy breeds (*Bos taurus*) than indigenous zebu cattle (*Bos indicus*) and crossbred beef cattle\[^{[41]}\]. Where extensive management is more common, animal crowding such as in water ponds, dip tanks, cattle markets still plays a major role in the spread of the disease.

**Control Measures and Programs**

The basic strategies required for control and elimination of bovine TB are well known and well defined\[^{[42]}\]. However, because of financial constraints, scarcity of trained professionals, lack of political will, as well as the underestimation of the importance of zoonotic TB in both the animal and public health sectors by national governments and donor agencies, control measures are not implemented adequately in most developing countries.

Bovine TB does not often justify the emergency measures required for other zoonotic diseases. Successful conduct of a test-and-slaughter policy requires sustained cooperation of national and private veterinary services, meat inspectors, and farmers, as well as adequate compensation for services rendered. Only a few developing countries can adhere to these requirements.

The full economic implications of zoonotic TB are overlooked in many developing countries where the overall impact of the disease on human health and animal production needs to be assessed. Where foot-and-mouth disease has been eliminated, bovine TB and other existing disease infections such as brucellosis have become important because of the impact on the meat and live animal export trade. Bovine TB and brucellosis also limit the development and expansion of the dairy industry at the regional level.

**Animal Vaccination and Research Developments**

Vaccination of animals against TB would be a viable strategy in two disease control situations. First, in domesticated animals in developing countries and second in wildlife and reservoirs where test-and-slaughter programs have failed particularly in developed countries.

Many issues need to be addressed before vaccination becomes a realistic option for control of disease in cattle and other animals. Of highest priority is the development of a highly effective vaccine. The results obtained globally with Bacillus Calmette-Guerin (BCG) have been suboptimal, and efficacy has varied considerably from region to region\[^{[43]}\]. In addition, the delivery of the vaccine poses a few problems in domesticated animals and is fraught with difficulties in wild animals. Moreover, vaccination itself may comprise diagnostic tests. A vaccine that induces tuberculin reactivity would invalidate the key diagnostic tool used in control programs. Evaluation of the protective efficacy of a new vaccine often poses serious difficulties. Traditionally, the guinea pig and mouse have been used for this purpose, but the information gained has been of little value. Recent work has indicated that the deer may well prove a suitable mammal for evaluating new vaccines and optimum delivery systems\[^{[44]}\].

The successful control of TB worldwide, not attainable presently, would become a reality if we take stock of a premise that a change of approach is imperative that is to take a fresh look at not just the immunopathogenesis and its correlation with clinical TB but also from a perspective of both animals and man.
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REFERENCES


[27] Young DB. Ten years of research progress and what’s to come.Tuberculosis 2003;83:77–81.


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