Tuberculosis is one of the leading causes of morbidity and mortality globally. Vitamin D directly influences the body’s immunological response to tuberculosis by modulating the production of certain antimicrobial peptides and the release of cytokines. Vitamin D deficiency has been extensively described in patients with tuberculosis. Currently, it is a recognised risk factor for tuberculosis. Genetic polymorphisms of the vitamin D receptor may influence host susceptibility to tuberculosis and response to antituberculosis therapy. This review article explores the close association between tuberculosis and vitamin D, with the aim of forming a strong basis for further interventional and genetic studies on patients with active tuberculosis.

Introduction

Tuberculosis remains a substantial global public health concern, especially in low- and middle-income countries. In 2011, the World Health Organization estimated that there were approximately 8.8-million incident cases of tuberculosis and roughly 1.45-million deaths due to tuberculosis globally. Approximately, one third of the world population (two billion) is latently infected with *Mycobacterium tuberculosis*, which may progress to active tuberculosis later.1 Approximately 1.1-million cases of tuberculosis worldwide are human immunodeficiency virus (HIV)-associated. The greatest disease burden of HIV-co-infection is documented in sub-Saharan Africa.2

Sources and metabolism of vitamin D

Vitamin D is a fat-soluble vitamin that is derived principally from exposure to sunlight and diet in humans. It occurs mainly in the form of two biologically inert precursors, i.e. vitamin D3 (cholecalciferol) and vitamin D2 (ergocalciferol). The latter is derived from plants and is produced exogenously by the irradiation of ergosterol. It enters the circulation through the diet.3

Vitamin D3 is formed when the cutaneous 7-dehydrocholesterol is exposed to solar ultraviolet B, and then later converted to previtamin D3. Through a heat-dependent process, previtamin D3 is instantly converted to vitamin D.

Vitamin D then undergoes hydroxylation in the liver to form 25-hydroxyvitamin D [25(OH)D], which is the major circulating form of vitamin D that is used to determine an individual’s vitamin D status. Additional renal hydroxylation of 25(OH)D, mediated by the 1-α-hydroxylase enzyme, occurs to form the biologically active form of vitamin D, i.e. 1,25-dihydroxyvitamin D [1,25(OH)2D] or calcitriol.3-4

Mechanisms of vitamin D and tuberculosis interaction: the immunological perspective

Vitamin D deficiency has been demonstrated to be associated with an increased risk of tuberculosis5 and a defective cell-mediated immune response to *M. tuberculosis* infection.6

A growing body of evidence suggests that vitamin D metabolites have very potent immunomodulatory properties. They also have significant immunological roles, like activation of monocytes, suppression of lymphocyte proliferation, cytokine synthesis and increasing the apoptosis of the macrophages that are infected with *M. tuberculosis*.7-8

Activated macrophages, lymphocytes and dendritic cells abundantly express vitamin D receptors (VDRs), suggesting a close relationship between vitamin D and the immune system.9-10 These immune cells also actively express the 1-α-hydroxylase enzyme, which unlike the renal 1-α-hydroxylase enzyme that is largely regulated by serum calcium concentrations, is dependent upon the immune
activation of the Toll-like receptors (TLRs) for its regulation. TLRs are mediators of innate immunity which are essential for microbial recognition of the macrophages and dendritic cells.  

Infection of the alveolar macrophages with *M. tuberculosis* results in activation of the macrophage TLR 1/2, which forms an integral part of the innate immune system. This activation is associated with increased expression of VDR and 1α-hydroxylase enzyme, as well as ensuing augmentation of the hydroxylation process of 25(OH)D3 into 1,25(OH)2D3 or calcitriol.

Increased production of calcitriol enhances the production of cathelicidin, an antimicrobial peptide which facilitates direct killing of the *M. tuberculosis*. This peptide is abundantly expressed by the alveolar macrophages, lymphocytes, neutrophils and epithelial cells. Cathelicidin also causes reduced intracellular viability of *M. tuberculosis* bacilli, by inducing increased fusion of the phagosomes and lysosomes within the infected macrophages.

Vitamin D also induces generalised antimycobacterial activity via other non-specific mechanisms. Macrophages that are infected with *M. tuberculosis* in the presence of adequate calcitriol concentrations produce high levels of nitric oxide (NO). In addition to its renowned vascular functions, NO also functions as a reactive oxygen species which exerts direct mycobacterial killing effects within the infected alveolar macrophages. In vitro, vitamin D also inhibits bacterial growth in macrophages infected with *M. tuberculosis* by stimulating the production of hydrogen peroxide and macrophage differentiation.

The extrapulmonary dissemination of tuberculosis and severe tuberculosis disease occurs owing to the ability of the *M. tuberculosis* bacilli to increase the expression and release of matrix metalloproteinases (MMPs) by the infected macrophages. MMPs are enzymes that cause increased degradation of components of the pulmonary extracellular matrix. They are also associated with the extrapulmonary spread of tuberculosis and extensive pulmonary cavitations in patients with tuberculosis. Studies have shown that in active form, vitamin D inhibits the secretion of MMPs, and increases the production of their inhibitors (the tissue inhibitor of metalloproteinase 1) in patients with tuberculosis. Therefore, vitamin D has a role to play in preventing severe pulmonary tuberculosis disease and extensive extrapulmonary tuberculosis spread.

**The role of vitamin D in tuberculosis management**

Vitamin D was extensively used in the management of active tuberculosis during the pre-antibiotic era. This was first demonstrated by Charpy and Dowling, who used vitamin D in the management of lupus vulgaris (cutaneous tuberculosis).

Vitamin D supplementation has been demonstrated to improve immunity against tuberculosis in the contacts of adult patients with tuberculosis. A study by Martineau et al showed that a single dose of 0.25 mg of oral vitamin D significantly enhanced immunity to *M. tuberculosis* in the contacts of tuberculosis-infected people for six weeks.

Some studies have shown that vitamin D supplementation is associated with clinical and radiological improvement and faster sputum smear conversion in patients with tuberculosis. Significant clinical, sonographic and radiological improvement was noted in study participants who received both oral vitamin D and antituberculosis therapy, compared to those receiving antituberculosis therapy alone, at eight weeks in a randomised controlled trial by Morcos et al (conducted in Egypt on 24 children with tuberculosis who were on antituberculosis therapy).

In another randomised controlled trial by Nursyam et al, on 67 Indonesian patients with tuberculosis on antituberculosis treatment, additional vitamin D therapy resulted in more rapid sputum smear conversion and radiological improvement in patients in the vitamin D supplementation group, than in those in the placebo group. Patients in the former group (n = 34, 100%) and only 25 patients in the latter group (76.7%) had sputum smear conversion at six weeks (p-value 0.002). Chest X-ray radiological improvement at six weeks was noted in 87.5% of patients in the vitamin D group, and in 65% of the patients in the placebo group.

However, other studies have reported contrasting results in relation to rate and time of sputum smear conversion in patients with tuberculosis who were supplemented with vitamin D. In a multicentre randomised controlled trial by Martineau et al on 126 patients on standard tuberculosis therapy, oral vitamin D supplementation in the dose of 2.5 mg at baseline, and on days 14, 28 and 42 did not significantly affect time to sputum culture conversion in the study participants collectively (adjusted hazard ratio 1.39, 95% confidence interval [CI]: 0.90–2.16, p-value 0.14). However, faster sputum smear conversion was observed in participants with the *TaqI* VDR polymorphism on genotyping (adjusted hazard ratio 8.09, 95% CI: 1.36–48.01, p-value 0.02).

Delays in the resolution of immunopathological inflammatory responses have been associated with increased mortality in patients with respiratory conditions. Vitamin D can be used as an effective adjuvant to antimicrobial therapy to reduce mortality in such patients because of its potent immunomodulatory role and ability to increase the resolution of pulmonary inflammation.

This effect of increasing the resolution of inflammation was demonstrated in a randomised controlled trial by Coussens et al on 95 patients on antituberculosis therapy to treat smear-positive tuberculosis. In this study, oral vitamin D supplementation was associated with significant suppression of concentrations of circulating inflammatory markers, like
C-reactive protein, interferon-gamma (IFN-\(\gamma\)), IFN-\(\gamma\)-inducible chemokine ligands, CXCL9 and CXCL10, metalloproteinase-9 (MMP-9), and antigen-stimulated chemokine ligand 5, interleukin-4 and IFN-alpha.

In one published case report, antituberculosis therapy, with simultaneous correction of vitamin D deficiency using oral vitamin D supplementation, resulted in clinical and microbiological improvement in an African American female patient who presented with refractory, drug-susceptible pulmonary tuberculosis.33 Therefore, oral vitamin D can be used as an adjuvant to antituberculosis therapy in such clinical circumstances.

Additionally, multivitamin supplementation, including vitamin D, reduced mortality by 50% in patients with HIV who were co-infected with tuberculosis in a randomised controlled trial on Tanzanian subjects with tuberculosis.34 Conversely, a randomised controlled trial by Wejse et al, carried out in Guinea Bissau on 365 adult patients with tuberculosis, documented no improvement in clinical outcome, weight gain nor a reduction in mortality, following vitamin D supplementation in doses of 100 000 IU at baseline, and at five and eight months subsequent to the initiation of antituberculosis therapy.35 The negative findings noted in this study could probably be explained by the suboptimal vitamin D doses used.

**Vitamin D levels in patients with active tuberculosis infection**

Low vitamin D levels have been well described in patients with tuberculosis.36 The earliest case control studies by Davies et al in London37 and Kenya38 noted lower 25(OH)D levels in participants.39-43 In Africa, where hypovitaminosis D is the most prevalent worldwide,44 varying frequencies of vitamin D deficiency in patients with active tuberculosis have been reported. Cross-sectional studies from Uganda,45 Tanzania46 and Guinea Bissau47 documented a low prevalence of 7.3%, 10.6% and 11%, respectively, while studies from Malawi48 and South Africa49 reported a high prevalence of 42% and 62.7%, respectively. This disparity in prevalence can probably be explained by the contrasting methods of vitamin D measurement and operational definitions of vitamin D deficiency used in the different studies, and the varying prevalence of HIV co-infection and dietary habits in the study participants.

**Vitamin D receptor genetic polymorphisms, susceptibility to tuberculosis and clinical tuberculosis outcomes**

Genetic polymorphisms involving the VDRs may influence susceptibility to tuberculosis and response to antituberculosis treatment.50 The commonly occurring polymorphisms of VDRs are restriction fragment length polymorphism of BsmI, ApaI and TaqI and the exon 2 splice site FokI polymorphism.

The TaqI TT VDR genotype has been shown to be associated with tuberculosis resistance and confers protection against tuberculosis disease, as seen in some native communities in West Africa51 and Paraguay.52 By contrast, a case control study that was conducted in India documented an increase in tuberculosis susceptibility in patients with the TaqI TT VDR genotype, especially in women.53

Wilkinson et al studied the influence of vitamin D deficiency and VDR polymorphisms, defined by the presence of restriction endonuclease sites for TaqI, BsmI, and FokI, on susceptibility to tuberculosis in adult Gujarati Asians in West London, an immigrant population with a high tuberculosis incidence.55 One hundred and twenty-six untreated patients with tuberculosis and 116 healthy tuberculosis contacts of Hindu and Gujarati origin, residing in London, were enrolled in this hospital-based, case control study. Although no significant independent association was noted between VDR genotypes and tuberculosis disease, the combination of genotype TT/Tt and vitamin D deficiency was associated with tuberculosis disease (odds ratio (OR) 2.8, 95% CI: 1.2-6.5, p-value 0.017). The presence of genotype ff, or undetectable vitamin D concentrations, was strongly associated with disease (OR 5.1, 95% CI: 1.4-18.4, p-value 0.015).

In another case control study on 103 patients aged 15-45 years with confirmed tuberculosis corresponding with age- and sex-matched healthy controls in Lima Peru, neither of the VDR polymorphisms was significantly associated with susceptibility to active tuberculosis [FokI FF genotype, compared to Ff or ff genotypes (OR 1.32, 95% CI: 0.55-3.18, p-value 0.538); [TaqI Tt or tt genotype, compared to TT genotype (OR 0.61, 95% CI: 0.29-1.30, p-value 0.199)].54

Increased severity of tuberculosis has been associated with the increased production of MMP-9.20 The TaqI T allele has been associated with decreased production of tissue inhibitor of metalloproteinase 1, a potent natural inhibitor of MMP-9,55 thereby increasing the risk of severe and aggressive tuberculosis disease in patients with that VDR polymorphism.

The association between VDR polymorphisms and response to tuberculosis treatment remains inconclusive. A significantly higher probability of sputum mycobacterial culture conversion during tuberculosis treatment was noted in participants with TaqI TT genotype [adjusted relative risk (RR) 4.28, 95% CI: 1.88-9.75, p-value 0.001] and FokI FF genotype (adjusted RR 2.35, 95% CI: 1.15-4.8, p-value 0.020) in a cohort study of 78 adult patients with confirmed tuberculosis in Lima Peru.54

In a similar study by Babb et al on a cohort of adult patients with tuberculosis in Western Cape, South Africa, a quicker response to tuberculosis chemotherapy was predicted with ApaI AA genotype and TaqI T-containing genotypes. However, no association between the VDR genotypes and tuberculosis
was observed in the case control study of 249 newly diagnosed South African patients with tuberculosis and 352 healthy controls.66

The recent most updated meta analysis and systematic review of 23 eligible studies, which examined the association between tuberculosis and certain genetic VDR polymorphisms, reported varying results.57 This was because of the diverse populations studied. Most included studies were underpowered.

Of the studies involving Asian subjects, the FokI ff genotype showed a strong positive association with active tuberculosis, while the TaqI and ApaI polymorphisms showed a trivial association. None of the VDR genetic polymorphisms were significantly related to increased susceptibility to tuberculosis in the studied African or South American populations.

The association between VDR polymorphisms and susceptibility to tuberculosis still remains very inconclusive. Hence, large and adequately powered studies that explore this area are justified.

Conclusion
There is now adequate compelling evidence that low vitamin D levels increase the risk and severity of tuberculosis disease. There is a great need for additional well designed studies to investigate the significance of vitamin D supplementation in patients with tuberculosis, especially in Africa where it is the most prevalent. Better designed and powered studies that explore the association between VDR genetic polymorphisms, tuberculosis susceptibility and treatment response are warranted.

Conflict of interest
The authors declare no commercial or other association that might pose a conflict of interest with regard to this study.

Declarations
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References


Review: What is the link between vitamin D and tuberculosis?