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Cathelicidins and defensins antimicrobial host defense peptides in the treatment of TB and HIV: Pharmacogenomic and nanomedicine approaches towards improved therapeutic outcomes

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Abstract

Tuberculosis (TB) and human immunodeficiency virus (HIV) represent a significant burden of disease on a global scale. Despite improvements in the global epidemic status, largely facilitated by increased access to pharmacotherapeutic interventions, slow progress in the development of new clinical interventions coupled with growing antimicrobial resistance to existing therapies represents a global health crisis. There is an urgent need to expand the armamentarium of TB and HIV therapeutic strategies. Host mediated immune responses represent an untapped reservoir of novel approaches for TB and HIV. Antimicrobial peptides (AMPs) are an essential aspect of the immune system. Cathelicidins and defensins AMPs have been studied for their potential applications in TB and HIV therapeutic interventions. Genetic polymorphism across different population groups may affect endogenous expression or activity of AMPs, potentially influencing therapeutic outcomes. However, certain genetic polymorphisms in autophagy pathways may alter the downstream effects of nano-delivery of cathelicidin. On the other hand, certain

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CRediT authorship contribution statement

AD and RDC conceived the idea. PND, AG, RDC and NK researched the literature and wrote the manuscript. AD, RDC and RC reviewed and revised the manuscript.

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Further Reading

genetic polymorphisms in beta-defensins may provide a protective role in reducing HIV-1 mother-to-child-transmission. Pharmaceutical development of cathelicidins and defensins is disadvantaged with complex challenges. Nanoparticle formulations improve pharmacokinetics and biocompatibility while facilitating targeted drug delivery, potentially minimising the risk of immunogenicity or non-specific haemolytic activity. This review aims to explore the potential viability of using cathelicidins and defensins as novel pharmacotherapy in the management of TB and HIV, highlight potential pharmacogenomic implications in host mediated immunity and AMP therapeutic applications, as well as propose novel drug delivery strategies represented by nanomedicine for AMPs.

Keywords

Antimicrobial peptides; Cathelicidin; Defensins; Nanoparticle drug delivery systems; Pharmacogenomics; Tuberculosis and HIV

1. Introduction

Pathogens are responsible for a significant proportion of the burden of disease observed in the human population today. *Mycobacterium tuberculosis* (*M.tb*) and the Human Immunodeficiency Virus (HIV) are among the major causes of morbidity and mortality globally [45]. *M.tb* which causes tuberculosis (TB) was the leading cause of death from a single infectious agent before the Covid-19 global pandemic and consistently represents a significant global epidemic with approximately a quarter of the world's population infected. An estimated 10 million individuals were diagnosed with *M.tb* as of 2020 before the outbreak of the covid-19 global pandemic which impacted diagnosis patterns ([83] a). Sub-Saharan Africa has the highest disease burden of HIV [75] and approximately half of TB cases globally are reported in the BRICS (Brazil, Russia, India, China and South Africa) countries ([83] a). Furthermore, approximately 38 million people were infected and living with HIV as of 2020 with 1.5 million new infections reported for that same year [75]. TB and HIV represent a common combined burden of disease in high risk populations and the former is the leading cause of death among people living with HIV, where one in three HIV related deaths are due to TB co-infection (UNAIDS, 2020). People living with HIV are 18 times more likely to contract TB and the risk of reactivation of TB for people living with latent TB is 5–10%, with an increased risk in patients living with HIV [84]. In South Africa, HIV and TB form part of the quadruple burden of diseases which include obesity and non-communicable diseases, poor maternal and child health and violence [29].

Antimicrobial peptides (AMPs) are an essential component of the innate immune system and represent novel therapeutic interventions for TB and HIV. AMPs are short, cationic and hydrophobic peptides and play an integral role in the host mediated immune response. Endogenously, genetically encoded AMPs are expressed in host immune cells to eliminate invading pathogens [42]. Cathelicidin and defensin AMPs endogenously fulfil important roles as effectors in the innate immunity [91], primarily responsible for direct inhibition of pathogens as well as modulating innate and adaptive immune responses [71]. Instead of administering antibiotics to treat infected patients, therapeutic interventions may involve

using an inducing agent to trigger AMP expression at a desired site and at the right time. Following the rationale that the endogenous expression of AMPs in host immune cells is induced in response to invading pathogens, it follows that AMPs form a compelling natural basis for novel antimicrobial agents. This can be either directly through exogenous AMPs or indirectly through AMPs inducers. However, most AMPs do not possess ideal physicochemical and biopharmaceutical properties *e.g.* potency, stability, selectivity. This presents a notable challenge in pharmaceutical applications where formulations of AMPs may be desired. Application would require a series of modifications and enhancements during the development process. Peptide nanoparticles could be used as specific drug delivery and target systems. Where the intracellular induction and expression of pathways involved with AMPs is concerned, genetic polymorphism and variations thereof present an extra dimension of complexity where potential pharmacotherapeutic applications may be involved.

In this review we discuss AMPs for TB and HIV, the potential pharmacogenomic implications as well as the applications of nanoparticle drug delivery systems to address the physicochemical and biopharmaceutical challenges faced by AMPs. The goal is to highlight the interface between pharmacogenomic and drug delivery aspects for AMPs for these diseases, with a view to support the development of precision AMP nanomedicine formulations.

1.1. Pathogenesis and limitations of TB and HIV pharmacotherapy

M.tb is an airborne disease and can be contracted by anyone who inhales a dispersed particle which can be suspended in room air for several hours, increasing the risk for transmission. The TB trajectory can be described as the primary infection when the inhaled particle is deposited in the lungs, followed by the latent phase when the immune system is still able to suppress the replication process and finally the acute phase where patients become symptomatic and acutely ill [73]. *M. tb* typically infects the lungs, however infection can manifest at other extrapulmonary sites. TB is a preventable disease and can be successfully cured through appropriate pharmacotherapeutic interventions which involve a six-month multidrug regimen with a fixed dose combination tablet [83]. In contrast, HIV is a chronic condition requiring lifelong therapy. HIV pathogenesis involves the rapid replication of the contracted virus followed by the debilitation of the host immune system, primarily the destruction of the host's CD4 + immune cells. Clinically these markers are used as indicators of disease progression, namely the viral load (VL) and the CD4 + cell count, respectively. Untreated, HIV may progress to a severe immunosuppressed stage of HIV known as acquired immunodeficiency syndrome (AIDS). An impaired immune system due to depletion of key immune cells results in increased morbidity and potentially, an increased risk of mortality in the affected host as they are susceptible to invading pathogens and potentially even latent ones such as TB. Fortunately, HIV can be managed through antiretroviral therapy (ART) which is largely accessible globally with approximately 26 million people living with HIV having access to ART as of 2020 [74].

People with HIV/TB co-infection are challenged with a high pill burden, which increases with the treatment of drug resistant TB and second line ART. Other factors that may

complicate treatment regimens include duration of therapy, adverse effects, stigma, mental health and financial burdens[72]. Immune reconstitution inflammatory syndrome (IRIS) and drug induced liver injury are other complications the medical team faces in HIV and TB treatment which can lead to mortality and requires a change in therapy [77,55]. Drug-drug and drug- gene -interactions need special consideration in this population [47]. Tables 1 and 2 summarise the mechanisms of action and limitations of current TB and HIV therapies. Rifampicin TB based regimens require dose optimization of ART. For example, given that rifampicin is a strong inducer of CYP450 enzymes and UGT1A1, when co-administered with a commonly used integrase inhibitor dolutegravir, twice daily *versus* once daily dosing of dolutegravir is required to account for this drug-drug interaction.

Incorporating pharmacogenomics into practice enables prescribers to follow a personalized medicine approach which can ultimately optimize therapy, reduce adverse effects and drug-drug interactions[56]. Patients living with HIV and/or TB are one of many patient groups who can benefit from pharmacogenomics implementation.

According to the World Health Organisation [81], approximately half a million people developed rifampicin-resistant TB (RR-TB) in 2019 of which 3.3% of new cases and 17.7% of previously treated cases had multidrug resistant TB/RR-TB. This observed trend in the development of multidrug resistant *M.tb* could potentially minimise the pool of viable pharmacotherapeutic interventions against TB. The incidence and prevalence of drug-resistant TB presents a challenge to the effective management of TB and a threat to global health. Research into novel TB therapeutic interventions promises a rejuvenation of TB pharmacotherapy. There is an urgent need to expand and improve the armamentarium of TB therapy. HIV drug resistance (HIVDR) exists globally and is steadily increasing. Further, patients living with HIV may have an increased risk for developing drug resistant TB [40,21]. In a study conducted in South Africa, HIVDR was detected in 27% of virally unsuppressed individuals with a higher prevalence in those who discontinued therapy [46]. Generally, antimicrobial resistance represents an imminent global health crisis, and HIVDR threatens the effective management of HIV [74]. HIVDR, particularly to efavirenz and/or nevirapine, represents the highest reported incidents of resistance in patients [74]. More than HIVDR, the effects of adherence on pharmacotherapeutic outcomes is much more pronounced and represents a persistent threat to the management of HIV. Poor adherence may be attributed to adverse effects associated with ART. Research into novel ART as well as novel formulation strategies for existing ART may provide therapeutic alternatives which may broaden the spectrum of effective HIV management strategies. For example, injectable therapies such as cabotegravir and rilpivirine hold various benefits including potentially improved adherence and patient satisfaction for stable patients living with HIV due to only monthly required injectable administrations of this particular ART [19,70]. However, in many countries with a high TB burden, this therapy is not yet available and contra-indicated with rifampicin use, since this coadministration could potentially result in subtherapeutic cabotegravir and rilpivirine drug concentrations leading to treatment failure [38].

1.2. AMPs in therapy

AMPs boast a broad spectrum of antimicrobial activity across gram-negative and gram-positive bacteria. Over 2800 AMPs isolated from natural sources are currently registered on the Antimicrobial Peptide Database (APD). Out of those only two groups of AMPs have been found in humans. These are broadly classified into two principal categories, namely cathelicidins and defensins [36]. Antimicrobial activity is a consequence of their physical characteristics. Multiple mechanisms for the bactericidal action of AMPs have been postulated. hypothetical mechanisms of action of these peptides, including the cell membrane damage, intracellular bactericidal mechanism, the inhibition of the synthesis of macromolecules, the damage of the organelles to cause DNA fragmentation, the inhibition of enzyme activity, and antimicrobial effect *via* participating in immune regulation [4,36,65]. The largely often cited explanation suggests bactericidal activity through membrane interaction that neutralises the charge of the bacterial cell membrane. AMPs with their amphiphilic α -helical structure consisting of hydrophobic and hydrophilic halves interact with the bacterial lipid bilayer membrane when they adsorb onto the bacterial surface and undergo conformational changes [4,36]. An amphipathic structure affects the specificity of their membrane activity. This is apparent in the observed target selectivity of AMPs for anionic bacterial membranes compared to more zwitterionic host cell membranes [42]. Antimicrobial peptides can destroy the membrane structures of bacteria, resulting in the massive exudation of cell contents and ultimately leading to the death of bacterial cells [36].

Interactions among positive charges and negative charges allow the cationic AMPs to bind to the outer structures of the cell membranes. This mechanism of cell membrane damage generally involves the positively charged AMPs selectively binding onto the surface of the negatively charged membrane of the bacterial cell before proceeding to destroy bacterial membranes by the membrane perforation or non-perforation mode. The membrane perforation mode can be classified into four hypothetical models including the barrel-stave model, the carpet model, the toroidal-pore model, and the aggregated channel model, while the non-perforation model predicts that AMPs bind to the surface of the bacterial cell membranes to cause the cell death by disrupting intracellular physiological processes, such as DNA replication, transcription, or protein synthesis [36].

Three electrostatic models that explain the AMPs destructive action on bacterial membranes have been hypothesized. First, the carpet model proposes that peptides remain in parallel conformation within the bi-layers and upon adequate coverage, a cleanser effect is generated which devastates the membrane. The second is the barrel stave model, where the peptides are supposedly introduced perpendicularly into the bi-layers, ultimately coalescing to create a pore. The third, the toroidal pore model, proposes that peptides are presented perpendicularly into the lipid bilayer to produce a territorial membrane curvature, where a pore is formed alongside phospholipid head bunches[65]. This mechanism confers specificity to the antimicrobial activity of AMPs, they can exert antimicrobial effects without damaging endogenous cells likely due to the α -helix surface of AMPs that have positive charges which conveniently interact with negatively charged membranes of microbes, while, in contrast, eukaryotic cellular membranes are composed of un-charged neutral phospholipids, sphingomyelins and cholesterol [36]. Practically AMPs display

good water solubility and thermal stability as well as being small enough to facilitate low-cost synthesis and production. This, partially, makes them favourable candidates for drug design and development. Conversely, naturally occurring AMPs exhibit a short-half and notable susceptibility to proteolysis making them biologically unstable. Cytotoxicity and the potential for immunogenicity and non-specific haemolytic activity are some limitations for clinical development and application [36].

1.3. Cathelicidin, vitamin D3 and autophagy in TB pathogenesis and treatment

Cathelicidins make up a broad range of antimicrobial defence proteins, however, the only human cathelicidin identified thus far is the Human Cationic Antimicrobial Protein (hCAP-18). Cathelicidin primarily contains an inactive antimicrobial domain and a signal peptide, which is a cathelin-like domain; the latter of which is a precursor protein from which the C-terminal peptide, LL-37, is cleaved. Upon being released from the pro-region through the action of proteinase 3, LL-37 becomes an active antimicrobial agent [71,89]. LL-37 is a versatile host protein that has been shown to have a number of pleiotropic functions such as chemotactic, angiogenic and wound-healing activities [5].

LL-37 has been identified in a number of cells including keratino-cytes, monocytes, mast cells and neutrophils. It plays a multifaceted role in innate immune cell responses. This includes the induction of interleukin 8 (IL-8), upregulation of CXCR4, CCR2 and IL-8RB and acts as a chemoattractant for neutrophils and monocytes. Increased binding of calcitriol to the vitamin D receptor (VDR) in macrophages is associated with the induction of LL-37, contributing to the macrophage's antimycobacterial activity [79]. Using the microtubule associated protein 1 light chain (LC3), Yuk and colleagues were able to monitor the development of autophagosomes in human monocytes treated with 1, 25-(OH)₂D₃. Contrasting this enhanced formation of autophagosomes against decreased autophagosomes in monocytes treated with an autophagy inhibitor, the study indicates that 1,25-(OH)₂D₃ not only induces autophagy, but is also the specific antimicrobial apparatus necessary for pathogen eradication. Cathelicidin is a key mediator for 1,25-(OH)₂D₃ dependent autophagy and is essential for the maturation and success of the autophagic pathway in antimicrobial activity against *M.tb* as illustrated in Fig. 1 and 2 [5,71,79,89].

Autophagy is an integral intracellular homeostasis pathway generally reserved for the degradation of damaged cellular components, misfolded proteins and other cytosolic debris. However, in the event of a microbial insult, autophagic pathways are observed as a central component of the intracellular defense, specifically, through the capture and degradation of immature phagosomes that harbour *M.tb*. [89]. Autophagy has been shown to play a significant role in host-mediated immunity against infection with intracellular *M.tb* [68,89]. Ideally, the induction of autophagy against intracellular *M.tb* should culminate in successful colocalization of autophagosomes with the phagosomes harbouring *M.tb* as shown in Fig. 2. Previous studies have shown that AMPs are central to vitamin D₃-mediated innate immunity, particularly against intracellular infection of *M.tb* [89]. Both *in vitro* and *in vivo* studies have shown enhanced phagocytosis and eradication of *M.tb* by macrophages as a result of treatment with vitamin D (Fig. 1). Notably a number of pathogens including *M.tb* have resistance mechanism(s) to neutralize AMPs. Several

pathogenic proteases have been documented that may degrade LL-37[78]. A study by Padhi et al. investigating the *M. tb* lipoprotein LprE_{Mtb}, particularly the significance in *M.tb* resistance mechanisms and virulence in macrophages, demonstrates that LprE_{Mtb} plays a significant role in facilitating *M.tb* intracellular survival. LprE_{Mtb} was shown to undermine the expression of CAMP through the down-regulation of CYP27B1 and VDR expression, both significant components of the p38-CYP27B1-VDR signaling pathway for CAMP expression. Subsequently permitting *M.tb* to evade autophagic eradication [54]. Continued studies of pathogenic mechanisms of resistance may elucidate strategies to circumvent their effects.

Vitamin D deficiency has been associated with susceptibility to develop active TB in infected individuals. Early treatment methods for TB used before the development of antibiotics involved exposure to sunlight and vitamin D supplementation [8,63,79]. Selvaraj *et. al* observed therapeutic benefits of vitamin D in treatment of TB, wherein infected patients who received vitamin D supplementation showed sputum clearance and radiological improvements relative to the control group [63]. Adequate vitamin D3 levels are required for sufficient expression of the AMP cathelicidin and pertinent for antimicrobial activity against *M.tb* [50,89].

Multiple studies report the active form of vitamin D3, 1,25-(OH)₂D3, as inducing autophagy in human monocytes through the expression of cathelicidin. Vitamin D3-induced autophagy has been shown to enhance innate immunity response against infection with intracellular *M.tb* [5,89]. 1,25-(OH)₂D3 concentrations (~20 nM) typically used in *in-vitro* studies to efficiently induce cathelicidin expression or antimicrobial activity against *M.tb* in human monocyte and macrophage cultures, are notably higher than endogenous serum levels. This would typically present concerns regarding the formulation of safe dosages, however studies have indicated sufficient induction of autophagy with normal physiological concentrations. Based on studies published on 1, 25-(OH)₂D3-induced autophagy, as far as autophagic eradication of intracellular *M.tb* is concerned, 1,25-(OH)₂D3 and LL-37 are invariably linked and interdependent.

Inducing the expression of cathelicidin through VDRs presents an exciting avenue for pharmaceutical applications that can enhance macrophage defenses against *M.tb* through autophagy. Treatment of macrophages with TLR agonists has been observed to increase vitamin D receptors in the cells resulting in increased production of LL-37 and other VitD responsive genes [71].

1.4. Defensins in HIV pathogenesis and treatment

Defensins are a family of small cationic antimicrobial peptides associated with the innate immune system. Defensins have been observed to have effective antimicrobial activity against both gram-negative and gram-positive bacteria, fungi as well as enveloped and non-enveloped viruses [5,91]. There is increasing evidence suggesting that all defensin subfamilies also have an inhibitory effect against HIV-1 [91]. Mammalian defensins are generally classified into one of three subfamilies. They may be either α , β , or θ defensins. This is based primarily on the relative size and pattern of their disulphide linkage. Further classification takes into account sites of expression as well as biological activity (Table 3)

[71]. Only two of these subfamilies are expressed in humans namely the α -defensins and the β -defensins [71, 91]. Structurally, defensins are generally observed as β -sheets with three intramolecular cysteine-disulfide bonds and are primarily distinguished by the distribution and connection of six cysteine residues [91]. At least six human α -defensins have been identified and these are distinguished as either enteric or myeloid α -defensins where the enteric α -defensins are primarily produced in the gut and the genitourinary tract while the latter are primarily associated with neutrophils. The myeloid subclass of the α -defensins is generally found in primary granules of neutrophils which are produced in the bone marrow. These constitute the human α -defensins 1–4 also referred to as human neutrophil peptides (HNP 1–4) on account of their significant abundance in neutrophils. Although primarily derived from neutrophils HNP 1–4 can also be found in granulocytes, natural killer (NK) cells, B-cells, $\gamma\delta$ T-cells, and monocytes/macrophages [18]. Generally HNPs are packaged in azurophil granules of neutrophils where they are constitutively expressed and released in large quantities during neutrophil activation [71]. HNPs have been detected in the placenta, intestinal mucosa, saliva, and cervical mucus plugs that occlude the uterine cervix, although the cell source producing these HNPs remains to be elucidated. Elevation of α -defensins has been observed in vaginal fluid from patients with lower and upper genital tract infection, suggesting some role for α -defensins in mucosal immunity against infection *in vivo* [18].

Enteric α -defensins, also referred to as human α -defensins (HD 5–6), are mainly produced by Paneth cells (PCs) in the small intestines, but have also been detected in the salivary gland, stomach and eyes [52,91]. Human β -defensins are primarily produced by epithelial cells. While HBD-1 is constitutively expressed by epithelial cells, HBD-2, and HBD-3 have been observed to be induced in response to viruses, bacteria, microbial products or pro-inflammatory cytokines such as tumor necrosis factor (TNF) and interleukin (IL)– 1 [71,80,91]. Regarding tissue distribution, HBD-1 and HBD-2 have been detected in oral and nasal mucosa, lung, plasma, salivary gland, small and large bowel, stomach, skin, eye, mammary gland, urogenital tracts and kidney [91]. The θ -defensins are only expressed in a select group of non-human primate species. Three θ -defensins have been found in leukocytes and bone marrow of rhesus macaque. Although RNA transcripts homologous to the rhesus θ -defensin gene, namely, DEFT, are found in human bone marrow, they contain a premature stop codon in the upstream signal sequence, which abolishes subsequent translation of the θ -defensin peptide. Retrocyclin is a synthetic circular peptide based on the sequence of the mature peptide encoded by the human θ -defensin pseudogene [91]. This gives the synthetic θ -defensin Retrocyclin a distinct cyclic peptide structure unlike the α and β -defensins.

There are different modes and determinants as far defensins binding to viruses to produce an antiviral effect. The inhibitory effect of defensins against HIV-1 has been reported *in-vitro*. Inhibition of HIV replication using α -defensins was first reported in 1993 using synthetic α -defensins obtained from guinea-pigs, rabbits and rats. The synthetic α -defensins were able to inhibit HIV-1 infection following viral entry. Defensin binding to viral proteins could disrupt receptor interactions essential for viral entry into the host cells. HNP1–4 interact with HIV-1's gp41 and gp120 and the cell surface receptor CD4 to produce an inhibitory effect [65]. Recently native α -defensins 1–3 purified from human neutrophils were shown to inhibit replication of R5 and X4 strains of HIV-1 at a 50% inhibitory concentration (IC50) of

0.5–22 μM . Pretreatment of cells with α -defensin-1, followed by wash-out prior to infection, blocks HIV-1 infection, indicating that direct inactivation of virions is not required for its inhibitory effect. The range of α -defensins reported to result in significant inhibition of HIV-1 ranges from the low micromolar to approximately 60 μM (~200 $\mu\text{g/mL}$). At the latter dose range, α -defensins may display some cytotoxic effects as well [18].

Further studies suggest at least a dual mechanism in the α -defensins anti-HIV activity. They can inhibit HIV-1 replication by a direct inactivation of the virus as well by affecting the target cells. However, it is important to note that while in the absence of serum, α -defensins can act directly on the virus, this effect is negated by the presence of serum or significant increase in virus particles. These cellular effects appear to block viral replication following entry and reverse transcription in CD4 T-cells including steps prior to completion of nuclear import as well as transcription [18,91].

While investigating the potential of hBDs 1–3 to inhibit HIV-1 in human monocyte-derived macrophages, Barucha et al. demonstrate that hBD2 and hBD3 inhibit HIV-1 in macrophages *in-vitro* in a dose-dependent manner. Through this study they demonstrate that this hBD2 mediated inhibition of HIV-1 occurs post-entry through several mechanisms. Notably, binding to and signalling through various CCR type that results in induction of anti-retroviral restriction factors of the APOBEC3 family. This study elucidates the capacity for defensins, specifically hBD2, to inhibit HIV-1 through several avenues beyond primary human PBMCs or CD4 + T cells[12].

There is some evidence to suggest defensin activity against *M.tb* in respiratory epithelia. While macrophages typically contain negligible amounts of HNPs, they have been observed to ingest neutrophils and take up the HNPs therein. The acquired HNPs may co-localize with *M.tb* in early endosomes and contribute towards inhibition of bacterial growth [71]. However, recent discoveries suggest some notable unfavourable effects of defensins that may promote viral infection under physiological conditions [86]. A study by Valere et al. demonstrated that HNP1, which is extensively associated with multifaceted anti-HIV activity, is also capable of disrupting epithelial integrity promoting HIV traversal across epithelial barriers, which may facilitate increased viral infection and dissemination[76]. Current evidence for the activity of defensins against HIV is largely based on *in vitro* studies, the challenge for future research is to determine the relevance of these observed *in vitro* trends *in vivo*.

1.5. Genetic polymorphisms affecting the activity of cathelicidin and defensins in TB and HIV therapy

The binding of 1,25-(OH)₂D₃ to VDR in immune cells creates a conformational change in the receptor, forming a heterodimer with the retinoic acid receptor (RXR). In the nucleus, this VDR/RXR complex activates the expression of Vitamin D Response Elements (VDRE) which are responsible for the upregulation of LL-37 (cathelicidin) production (Fig. 3). Studies show that under conditions of decreased serum 1,25-(OH)₂D₃ levels, the magnitude of LL-37 upregulation is reduced, limiting the immune response of macrophages in killing intracellular *M. tb* [2,8,13].

1.6. Vitamin D receptor (VDR)

Polymorphisms in the VDR gene, located on chromosome 12, are associated with variable functionality which may impact the downstream binding effects of 1,25-(OH)₂D₃. These include *Apal* (rs7975232), *BsmI* (rs1544410), and *TaqI* (rs731236), found in strong linkage disequilibrium near the 3′-untranslated region. They alter mRNA stability through VDR mRNA transcription polyadenylation. The *FokI* (rs2228570) polymorphism, located near the start codon of VDR, is associated with functional effects. In particular, the long variant (f allele) is associated with less activity (*i.e.* transactivation of VDR) than the short variant (F allele). Polymorphisms in *Apal*, *BsmI*, *TaqI*, and *FokI* have been associated with regulation of LL-37 production in various ethnic populations [30,44]. These polymorphisms, that lead to decreased VDR expression, have been shown to have subsequent decreased production of LL-37 post vitamin D administration and *vice versa* (Fig. 3). Furthermore, multiple clinical studies have shown mixed results regarding the association of various VDR polymorphisms and their susceptibility to TB in different populations [7,9,14,17,31,35,37,43,44,53,57,69]. It is unclear why these differences exist but additional well-designed, larger-scale epidemiological studies among other ethnicities are warranted. Overall, these VDR polymorphisms may potentially affect the use of vitamin D AMPs in soliciting an immune response of macrophages (*i.e.* regulation of cathelicidin production) in killing intracellular *M.tb*. However, delivering AMPs such as LL-37 (cathelicidin) as opposed to inducing its production, could avoid and bypass these potential drug-gene interactions associated between vitamin D and VDR function.

1.7. Beclin-1

The protein Beclin-1, also called autophagy-related 6 protein or ATG6, plays a role in multiple cellular processes and is integral in the induction of autophagy. Because of this, it may be associated with TB and HIV outcomes ([16,33,88]). Transcription of Beclin-1 and ATG5 by 1,25-(OH)₂D₃ is promoted when cathelicidin is present, leading to the down regulation of mTOR ([20,67]). As a result, autophagy is induced, and mycobacteria are suppressed [67]. Chung et al. report that although vitamin D and cathelicidin lead to HIV autophagy, it does lead to HIV-1 inhibition [20]. As ATG6 is located downstream of LL-37 and its expression is dependent on the presence of LL-37 [20], it is unclear whether genetic variants in the Beclin-1 promoter region (rs60221525, rs116943570, rs34882610, and rs34037822 and c.-933delG [32]) will impact its overall function in inducing autophagy. Additional studies are needed to understand the role of using LL-37 in the presence of these polymorphisms in Beclin-1 and its potential impact on overall management of TB and HIV.

1.8. Autophagy-related 5 (ATG5)

Located downstream of LL-37, autophagy-related 5 protein (ATG 5) is involved in the autophagy pathway, specifically in the formation of autophagic vesicles [87]. Similar to Beclin-1, expression of ATG5 is induced by 1,25-(OH)₂D₃ in the presence of LL-37, leading to the down regulation of mTOR and mycobacterial suppression ([20,67]). ATG5 is found on chromosome 5q12 and has numerous single nucleotide polymorphisms [64]. One of these SNPs includes rs2245214, which Songane et al. and Cucu et al. both concluded

that ATG5 was not associated with susceptibility to TB. However, the role of ATG5 polymorphisms in TB is not well understood and more robust studies are warranted.

1.9. Immune-related GTPase family M 1 (IRGM1)

Immune-related GTPase family M (IRGM1) is a protein that regulates autophagy and has been shown to have a potential role in TB susceptibility [6]. Located on chromosome 5q33.1, the IRGM gene encodes 5 exons and is involved in the formation of the autophagosome ([67]). The IRGM gene promoter region has numerous polymorphisms including rs4958843, rs10065172, and rs4958846, which may alter the expression of the gene ([10,34]). Studies by Barahi et al. and King et al. report that single nucleotide polymorphisms in IRGM1 have been associated with increased susceptibility to TB in the Iranian and African American populations, respectively. King et al. concluded that although the rs10065172 polymorphism was associated with susceptibility to TB, it did not affect the expression of IRGM1 in peripheral blood mononuclear cells but may be tissue dependent [34]. Alternatively, some studies have shown the rs10065172 polymorphism to be associated with decreased susceptibility to TB in the Korean and Asian populations ([66,85]. Due to conflicting outcomes of IRGM1 in studies involving different populations, additional research is needed to determine the role of genetic polymorphisms in IRGM1 in TB infection.

Overall, current literature suggests that genetic polymorphisms in VDR might impact the effectiveness of using AMPs involving vitamin D for the management of TB. However, these potential gene-drug interactions could be overcome by delivering AMPs such as LL-37 (cathelicidin). It is unclear whether downstream genetic polymorphisms in Beclin-1, ATG5 and IGRM1 will impact the effectiveness of using AMPs such as LL-37 for management of TB and HIV.

1.10. DEFB1 (human beta defensins 1 (hBD-1))

The DEFB1 (8p23.1) gene encodes for human beta defensins 1 (hBD-1). Relevant functional polymorphisms in this gene that may modulate expression include the following: the -52 G>A (rs1799946), 44 C>G (rs1800972) and -20 G>A (rs11362) at the 5' untranslated region (5' UTR) and the c.* 87 A>G (rs1800971) at the 3' UTR. In a recent study by Zupin et al. they observed that the DEFB1 c.* 87 A allele was associated with decreased susceptibility towards HIV-1 mother-to-child transmission (MTCT) in a Zambian population. Previous studies have also shown associations between DEFB1 polymorphisms and HIV-1 infection among other ethnic groups ([15,41,58,61,62]). Although no association was found between maternal DEFB1 polymorphisms and plasma viral load (Zupin et al.), constitutive expression of hBD-1 at the mucosal surface including placenta may account for the potential benefits this may have for preventing HIV-1 MTCT [58,60]. Zupin et al. propose that the DEFB1 c.* 87 G allele and the DEFB1 ACGA haplotype, more commonly representative among children with HIV, might have decreased mRNA DEFB1 expression and hBD-1 levels resulting in an increased risk of acquiring HIV-1 infection. Additional studies are warranted to fully understand the role of DEFB1 gene polymorphisms in HIV-1 MTCT. These study findings also suggest the potential beneficial role that antimicrobial peptides such as beta defensins may have in curbing the HIV epidemic.

2. Precision peptide nanomedicine for delivery of AMPs for TB and HIV therapy

While continued research into AMPs and their potential pharmacotherapeutic value promises alternative novel therapeutic interventions for infectious diseases, naturally occurring cathelicidins and defensins present with various disadvantages that complicate their development as clinically applicable pharmaceuticals. These problems include the nonspecific haemolytic activity on human cells, osmotic sensitivity, rapid turnover in the human body, and high price of production [4]. Clinical development is also disadvantaged by the peptide structure which is chemically unstable and highly susceptible to various biological proteases [3,39]. Therefore viable clinical applications of AMPs would require some preconditions to be met, *i.e.* stability in the human body, transportation, targeted delivery, controlled release, and immunogenicity of these peptides must be improved. Developing safe and practical formulations for oral delivery, for example, may be complicated by enzymatic degradation. Several gastrointestinal enzymes such as aminopeptidases, exopeptidases, and carboxypeptidases rapidly break down amino acids immediately inactivating the peptides [4].

Efforts to overcome the challenges presented by AMPs have focused on improving their half-life and overall biological stability. Notable approaches include chemical alterations such as cyclization or lipidation, design of peptidomimetics, synthesis of hybrid peptides, or novel formulation strategies using nanocarriers, engineered particles of size range 1–1000 nm. The prospect of encapsulating and delivering AMPs in nanoparticle delivery systems presents a unique avenue for viable clinical application of the otherwise biologically unstable AMPs [39]. Carrier systems affect the pharmacokinetic properties, potentially improve bio adhesion, biodegradation, and biocompatibility. Another advantage of carrier systems is target specificity, preventing AMPs disseminating to the environment of other unaffected cells [4].

Encapsulation of therapeutics ideally implies protecting the therapeutic from degradation, improving its absorption across biological barriers, controlling the release rate and facilitating targeted delivery of the therapeutic agent to cellular targets [23,25,90]. Encapsulating AMPs into nanoparticles would maintain their chemical integrity and improve peptide stability, protecting them from enzymatic degradation and deactivation. Beyond enhanced stability and integrity, nanoparticles could facilitate controlled release, mitigate adverse effects, improve biofilm penetration as well as allow for localized activity following administration [3,39]. Nanocarriers can improve dissolution rates of poorly soluble or insoluble drugs and facilitate controlled release, both of which may improve the bioavailability. Extending duration of action, reduced frequency of administration and minimised risk for drug-drug interactions makes nanocarriers a particularly favourable option for peptide formulation, particularly in TB and HIV co-infection. Nanoparticle formulations for AMPs may include, but are not limited to liposomes, micelles, polymeric nanoparticles, or lipid nanoparticles or lipid-polymer hybrid nanoparticles.

Nanoparticles can direct encapsulated therapeutic agents to target cells. This would improve concentrations of the desired peptide at the site of action and reduce off-target

effects [24,25,90]. This may be particularly advantageous where genetic polymorphisms in susceptible individuals or groups may affect endogenous expression, production or overall function of the AMPs. The ideal outcome for encapsulation is targeted action at the affected tissue. There are two strategies for targeted nanoparticle drug delivery. One strategy relies on active targeting where the carrier system typically possesses a ligand for the target tissue. The other strategy is more passive and relies on the carrier system targeting affected, typically inflamed tissues, where it is likely to accumulate at relatively higher concentrations [90].

There are a few studies investigating nanoparticle encapsulation and therapeutic activity of AMPs. Gold nanoparticles (AuNPs) have demonstrated potent antimicrobial activity and used to counteract multidrug resistant bacteria, yet they retain low toxic activity against mammalian cells, and the fact that they do not induce bacterial resistance [4]. Core-shell magnetic nanoparticles, typically employed in theranostics, have been demonstrated to have unique biological properties and the potential for bactericidal activity achieved through the destruction of the bacterial cell membrane [49]. A recent study investigating potential synergistic antibacterial effects of core-shell magnetic nanoparticles with LL-37 against *Staphylococcus aureus* and *Pseudomonas aeruginosa* demonstrated a potentiated effect. Where LL-37 was combined with gold coated core-shell magnetic nanoparticles, significantly increased antibacterial activity was observed, at doses of 2 µg/mL and 4 µg/mL this combination managed to inhibit bacterial growth of tested bacterial strains, indicating a 64-fold and 32-fold decrease in the original MIC value of LL-37 [49].

Recent studies investigate the use of novel formulation strategies such as lipidation for AMPs where a fatty acid is added to the N- or C-terminal affinity which facilitates membrane permeability [28]. AMPs loaded into nanocarriers may be easier to deliver to important sites of pathogenic assault such as phagocytic cells [3,28,39]. Matougui et al. [39] reported the encapsulation of LL-37, AA230, an amphipathic arenicin-3 derivative of 21 amino acid length as well as DPK-060, a 20 amino acid derivative of human kininogen in reverse micelle lipid nanocarriers, achieving good encapsulation efficiency. Where MICs of the native peptides were first determined against seven selected bacterial strains, nanoformulated LL-37 was shown to have comparable MIC to the unformulated peptide indicating the preservation of antimicrobial activity following nanoformulation. Where stability against proteolysis was concerned, the nanoformulations succeeded to ensure protection of the LL-37 relative to the naked peptide. The kinetics of LL-37 degradation were significantly slower, when adsorbed on the surface or encapsulated in the core of the lipid nanocapsules [39].

This study and similar studies elucidate proof of concept for viable formulation of AMPs as nanoparticles. Further studies into nanoformulation of AMPs particularly where the biological activity and relative safety of encapsulated AMPs *in vivo* is concerned, are therefore important and an essential foundation for future clinical development.

3. Conclusion and future directions

Significant challenges exist in the treatment of HIV and TB and there is a need for therapeutic innovation. Extensive research into AMPs and their potential therapeutic application has been conducted to date and more research is currently being carried out. This review presents insight for AMPs as part of the innate immune response to be active in the eradication of invading pathogens that include endemic pathogens responsible for TB and HIV. Much of the research done on the AMP LL-37 (cathelicidin) indicates activity against TB and presents the prospect of a viable therapeutic alternative to existing interventions. Likewise defensins have demonstrated extensive antiviral activity against HIV-1 through varied mechanisms, demonstrated in not only in CD4 + T cells, but other key cells including macrophages and PBMCs. A more accurate and precise understanding of the AMPs and their antimicrobial mechanisms of action through continued research will facilitate more efficient clinical development and application. Using AMPs such as LL-37 (cathelicidin) could avoid and bypass potential drug-gene interactions associated between vitamin D and VDR function. However, additional studies are warranted to further elucidate the role of using LL-37 in the presence of downstream polymorphisms in Beclin-1, ATG5 and IGRM1 and its potential impact on overall management of TB and HIV. Furthermore, additional research is needed to confirm the beneficial role of DEFB1 gene polymorphisms in prevention of HIV-1 MTCT. Research into AMPs is largely relegated to small scale, mostly *in-vitro* studies. *In-vivo* and clinical studies focused on safe and viable pharmaceutical application may elucidate potential approaches towards broad therapeutic use. Where stability may have presented an issue with viable therapeutic applications, research into nanoparticles as a possible avenue of formulation is currently scarce, but could chart a potential path forward. Peptide nanoparticles could be used as specific drug delivery and target systems. They have a promising future in the treatment of different bacterial and viral infections.

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Data Availability

No data was used for the research described in the article.

Abbreviations:

1,25-OH2D3	1,25-dihydroxycholecalciferol/Calcitriol
AIDS	Acquired Immunodeficiency Syndrome
AMP	Antimicrobial Peptide
ART	Anti-retroviral Therapy
CYP27B1	Cytochrome P27B1

CYP450	Cytochrome P450
HDP	Host Defense Peptides
HIV	Human Immunodeficiency Virus
IRIS	Immune reconstitution inflammatory syndrome
LprE	Putative lipoprotein
PBMCs	Peripheral Blood Mononuclear cells
TB	Tuberculosis
M.tb	Mycobacterium tuberculosis
MTCT	mother-to-child transmission
UGT1A1	Uridine diphosphate glucuronosyltransferase 1A1
VDR	Vitamin D Receptor
VL	Viral Load
WHO	World Health Organisation

References

- [1]. Ajbani K, Shetty A, Mehta A, Rodrigues C, Rapid diagnosis of extensively drug-resistant tuberculosis by use of a reverse line blot hybridization assay, *J. Clin. Microbiol* 49 (7) (2011) 2546–2551, 10.1128/JCM.02511-10. [PubMed: 21613436]
- [2]. Akimbekov NS, Ortoski RA, Razzaque MS, Effects of sunlight exposure and vitamin D supplementation on HIV patients, *J. Steroid Biochem Mol. Biol* 200 (2020), 105664, 10.1016/j.jsbmb.2020.105664. [PubMed: 32229174]
- [3]. Alamdaran M, Movahedi B, Mohabatkar H, Behbahani M, In-vitro study of the novel nanocarrier of chitosan-based nanoparticles conjugated HIV-1 P24 protein-derived peptides, *J. Mol. Liq* 265 (2018) 243–250.
- [4]. Amerikova M, Pencheva El-Tibi I, Maslarska V, Bozhanov S, Tachkov K, Antimicrobial activity, mechanism of action, and methods for stabilisation of defensins as new therapeutic agents, *Biotechnol. Amp. Biotechnol. Equip* 33 (1) (2019) 671–682.
- [5]. Arranz-Trullén J, Lu L, Pulido D, Bhakta S, Boix E, Host antimicrobial peptides: the promise of new treatment strategies against tuberculosis, *Front. Immunol* (2017) 8.
- [6]. Aravindan PP, Host genetics and tuberculosis: theory of genetic polymorphism and tuberculosis, *Lung India* 36 (3) (2019) 244–252, 10.4103/lungindia.lungindia_146_15. [PubMed: 31031349]
- [7]. Areeshi MY, Mandal RK, Akhter N, Panda AK, Haque S, Evaluating the association between TaqI variant of vitamin D receptor gene and susceptibility to tuberculosis: a meta-analysis, *Toxicol. Int* 21 (2) (2014) 140–147, 10.4103/0971-6580.139791. [PubMed: 25253922]
- [8]. Ayelign B, Workneh M, Molla MD, Dessie G, Role of vitamin-D supplementation In TB/HIV Co-infected patients, *Infect. Drug Resist* 13 (2020) 111–118. Published 2020 Jan 10. doi:10.2147/IDR.S228336.
- [9]. Babb C, van der Merwe L, Beyers N, et al. , Vitamin D receptor gene polymorphisms and sputum conversion time in pulmonary tuberculosis patients, *Tuberculosis* 87 (4) (2007) 295–302, 10.1016/j.tube.2007.03.001. [PubMed: 17449323]

- [10]. Bahari G, Hashemi M, Taheri M, Naderi M, Eskandari-Nasab E, Atabaki M, Association of IRGM polymorphisms and susceptibility to pulmonary tuberculosis in Zahedan, Southeast Iran, *ScientificWorldJournal* 2012 (2012), 950801, 10.1100/2012/950801. [PubMed: 23049477]
- [11]. E Beauduy C, Winston LG Antimycobacterial Drugs. In: Katzung BG, Vanderah TW. eds. *Basic & Clinical Pharmacology*, 15e. McGraw Hill; 2021. Accessed April 25, 2022. <https://accesspharmacy.mhmedical.com/content.aspx?bookid=2988§ionid=250602139>.
- [12]. Bharucha J, Sun L, Lu W, Gartner S, Garzino-Demo A, Human beta-defensin 2 and 3 Inhibit HIV-1 replication in macrophages, *Front. Cell. Infect. Microbiol* (2021) 11.
- [13]. Bizzaro G, Antico A, Fortunato A, Bizzaro N, Vitamin D and Autoimmune Diseases: Is Vitamin D Receptor (VDR) Polymorphism the Culprit? *Isr. Med Assoc. J* 19 (7) (2017) 438–443.
- [14]. Bornman L, Campbell SJ, Fielding K, et al. , Vitamin D receptor polymorphisms and susceptibility to tuberculosis in West Africa: a case-control and family study, *J. Infect. Dis* 190 (9) (2004) 1631–1641, 10.1086/424462. [PubMed: 15478069]
- [15]. Braidia L, Boniotto M, Pontillo A, Tovo PA, Amoroso A, Crovella S, A single-nucleotide polymorphism in the human beta-defensin 1 gene is associated with HIV-1 infection in Italian children, *AIDS* 18 (2004) 1598–1600. [PubMed: 15238780]
- [16]. Campbell GR, Spector SA, Vitamin D inhibits human immunodeficiency virus type 1 and Mycobacterium tuberculosis infection in macrophages through the induction of autophagy, *PLoS Pathog.* 8 (5) (2012), e1002689, 10.1371/journal.ppat.1002689. [PubMed: 22589721]
- [17]. Cao Y, Wang X, Cao Z, Cheng X, Vitamin D receptor gene *FokI* polymorphisms and tuberculosis susceptibility: a meta-analysis, *Arch. Med Sci* 12 (5) (2016) 1118–1134, 10.5114/aoms.2016.60092. [PubMed: 27695504]
- [18]. Chang T, Vargas J, DelPortillo A, Klotman M, Dual role of α -defensin-1 in anti-HIV-1 innate immunity, *J. Clin. Investig* 115 (3) (2005) 765–773. [PubMed: 15719067]
- [19]. Chounta V, Overton ET, Mills A, et al. , Patient-reported outcomes through 1 year of an HIV-1 clinical trial evaluating long-acting cabotegravir and rilpivirine administered Every 4 or 8 Weeks (ATLAS-2M), *Patient* 14 (6) (2021) 849–862. [PubMed: 34056699]
- [20]. Chung C, Silwal P, Kim I, Modlin RL, Jo EK, Vitamin D-cathelicidin axis: at the crossroads between protective immunity and pathological inflammation during infection, *Immune Netw.* 20 (2) (2020), e12, 10.4110/in.2020.20.e12. Published 2020 Feb 11. [PubMed: 32395364]
- [21]. Cox H, Salaam-Dreyer Z, Goig GA, Nicole MP, Menardo F, Dippenaar A, et al. , Potential contribution of HIV during first-line tuberculosis treatment to subsequent rifampicin-monoresistant tuberculosis and acquired tuberculosis drug resistance in South Africa: a retrospective molecular epidemiology study, *Lancet Microbe* 2 (11) (2021), e584, 10.1016/S2666-5247(21)00144-0. [PubMed: 34766068]
- [22]. Department of Health and Human Services (DHHS). Panel on antiretroviral guidelines for adults and adolescents. Guidelines for the use of antiretroviral agents in adults and adolescents with HIV. Available at <https://clinicalinfo.hiv.gov/sites/default/files/guidelines/documents/AdultandAdolescentGL.pdf>. (Accessed 26 April 2022).
- [23]. Dube A, Nanomedicines for infectious diseases, *Pharm. Res* 36 (4) (2019) 1–2.
- [24]. Dube A, Lemmer Y, Hayeshi R, Balogun M, Labuschagne P, Swai H, Kalombo L, State of the art and future directions in nanomedicine for tuberculosis, *Expert Opin. Drug Deliv* 10 (12) (2013) 1725–1734. [PubMed: 24102208]
- [25]. Dube A, Semete-Makokotlela B, Ramalapa B, Reynolds J, Boury F, Nanomedicines for the Treatment of Infectious Diseases: Formulation. Delivery and Commercialization Aspects. In *Green Synthesis in Nanomedicine and Human Health*, CRC Press., 2021, pp. 271–290.
- [26]. Food & Drug Administration (FDA). FDA warns about increased risk of ruptures or tears in the aorta blood vessel with fluoroquinolone antibiotics in certain patients. 2018a <https://www.fda.gov/drugs/drug-safety-and-availability/fda-warns-about-increased-risk-ruptures-or-tears-aorta-blood-vessel-fluoroquinolone-antibiotics> (Accessed 25 April 2022).
- [27]. Food & Drug Administration (FDA). FDA reinforces safety information about serious low blood sugar levels and mental health side effects with fluoroquinolone antibiotics; requires label changes. 2018b <https://www.fda.gov/drugs/drug-safety-and-availability/fda-warns->

[about-increased-risk-ruptures-or-tears-aorta-blood-vessel-fluoroquinolone-antibiotics](#) (accessed 26 April 2022).

- [28]. Gao X, Ding J, Liao C, Xu J, Liu X, Lu W, Defensins: The natural peptide antibiotic, *Adv. Drug Deliv. Rev* 179 (2021), 114008. [PubMed: 34673132]
- [29]. Hofman K, Shabir M, The unanticipated costs of COVID-19 to South Africa's quadruple disease burden, *South Afr. Med. J* 110 (8) (2020) 689–699, 10.7196/SAMJ.2020.v110i8.15125.
- [30]. Jiang X, Kiel DP, Kraft P, The genetics of vitamin D, *Bone* 126 (2019) 59–77, 10.1016/j.bone.2018.10.006. [PubMed: 30316967]
- [31]. Kang TJ, Jin SH, Yeum CE, et al. , Vitamin D receptor gene *TaqI*, *BsmI* and *FokI* polymorphisms in korean patients with tuberculosis, *Immune Netw.* 11 (5) (2011) 253–257, 10.4110/in.2011.11.5.253. [PubMed: 22194708]
- [32]. Kazachkova N, Raposo M, Ramos A, Montiel R, Lima M, Promoter variant alters expression of the autophagic BECN1 Gene: implications for clinical manifestations of machado-joseph disease, *Cerebellum* 16 (5–6) (2017) 957–963, 10.1007/s12311-017-0875-4. [PubMed: 28699106]
- [33]. Killian MS, Dual role of autophagy in HIV-1 replication and pathogenesis, *AIDS Res Ther.* (2012), 10.1186/1742-6405-9-16, 2012;9(1):16. Publ. [PubMed: 22606989]
- [34]. King KY, Lew JD, Ha NP, et al. , Polymorphic allele of human IRGM1 is associated with susceptibility to tuberculosis in African Americans, *PLoS One* 6 (1) (2011), e16317, 10.1371/journal.pone.0016317. Published 2011 Jan 21. [PubMed: 21283700]
- [35]. Lee YH, Song GG, Vitamin D receptor gene *FokI*, *TaqI*, *BsmI*, and *ApaI* polymorphisms and susceptibility to pulmonary tuberculosis: a meta-analysis, *Genet. Mol. Res* (2015), 10.4238/2015.August.7.21, 2015;14(3): 9118–9129. Publ. [PubMed: 26345844]
- [36]. Lei J, , Sun L, , Huang S, Zhu C, Li P, He J, Mackey V, Coy D, He Q, 2022. The antimicrobial peptides and their potential clinical applications. [online] PubMed Central (PMC). Available at: <<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6684887/>> [Accessed 28 April 2022].
- [37]. Liu W, Cao WC, Zhang CY, et al. , VDR and NRAMP1 gene polymorphisms in susceptibility to pulmonary tuberculosis among the Chinese Han population: a case-control study, *Int. J. Tube Lung Dis* 8 (4) (2004) 428–434.
- [38]. Fernandez C, van Halsema C, Evaluating cabotegravir/rilpivirine long-acting, injectable in the treatment of HIV infection: emerging data and therapeutic potential, *HIV/AIDS* 11 (2019) 179–192, 10.2147/HIV.S184642.
- [39]. Matougui N, Groo A, Umerska A, Cassisa V, Saulnier P, A comparison of different strategies for antimicrobial peptides incorporation onto/into lipid nanocapsules, *Nanomedicine* 14 (13) (2019) 1647–1662. [PubMed: 31293215]
- [40]. Mesfin YM, Hailemariam D, Biadgign S, Kibret KT, Association between HIV/AIDS and multi-drug resistance tuberculosis: a systematic review and meta-analysis, *PLOS ONE* 9 (2) (2014), e89709, 10.1371/journal.pone.0082235.
- [41]. Milanese M, Segat L, Pontillo A, Arraes LC, de Lima Filho JL, Crovella S, DEFB1 gene polymorphisms and increased risk of HIV-1 infection in Brazilian children, *AIDS* 20 (2006) 1673–1675. [PubMed: 16868452]
- [42]. Mishra B, et al., 'Host defense antimicrobial peptides as antibiotics: design and application strategies', in: *Current Opinion in Chemical Biology*, 38, Elsevier Ltd., 2017, pp. 87–96, 10.1016/j.cbpa.2017.03.014. [PubMed: 28399505]
- [43]. Mahmoud AA, Ali AHK, Vitamin D receptor gene polymorphism and 25 hydroxy vitamin D levels in Egyptian patients with pulmonary tuberculosis, *Egypt J. Chest Dis. Tube* 63 (2014) 651–655, 10.1016/j.ejcdt.2014.02.015.
- [44]. Mohammadi A, Khanbabaie H, Nasiri-Kalmarzi R, Khademi F, Jafari M, Tajik N, Vitamin D receptor *ApaI* (rs7975232), *BsmI* (rs1544410), *FokI* (rs2228570), and *TaqI* (rs731236) gene polymorphisms and susceptibility to pulmonary tuberculosis in an Iranian population: A systematic review and meta-analysis, *J. Microbiol Immunol. Infect* 53 (6) (2020) 827–835, 10.1016/j.jmii.2019.08.011. [PubMed: 31740220]
- [45]. Montales MT, Chaudhury A, Beebe A, Patil S, Patil N, HIV-associated TB syndemic: a growing clinical challenge worldwide, *Front. Public Heal* 3 (2015) 281.

- [46]. Moyo S, Hunt G, Zuma K, Zungu M, Marinda E, et al. , HIV drug resistance profile in South Africa: Findings and implications from the 2017 national HIV household survey, PLOS ONE 15 (11) (2020), e0241071, 10.1371/journal.pone.0241071. [PubMed: 33147285]
- [47]. Mukonzo J, Aklillu E, Marconi V, Schinazi RF, Potential drug–drug interactions between antiretroviral therapy and treatment regimens for multi-drug resistant tuberculosis: Implications for HIV care of MDR-TB co-infected individuals, Int. J. Infect. Dis 83 (2019) 98–101, 10.1016/j.ijid.2019.04.009. [PubMed: 30991140]
- [48]. Namdar R, Tuberculosis Peloquin C, Pharmacotherapy: A Pathophysiologic Approach, 11e, in: DiPiro JT, Yee GC, Posey L, Haines ST, Nolin TD, Ellingrod V (Eds.) 2022, McGraw Hill, 2020. Accessed April 25, <<https://accesspharmacy.mhmedical.com/content.aspx?bookid=2577§ionid=223398313>> .
- [49]. Niemirowicz K, Piktel E, Wilczewska A, Markiewicz K, Durna B, W tek M, Puszkarz I, Wróblewska M, Nikli ska W, Savage P, Bucki R, Core-shell magnetic nanoparticles display synergistic antibacterial effects against *Pseudomonas aeruginosa* and *Staphylococcus aureus* when combined with cathelicidin LL-37 or selected ceragenins, Int. J. Nanomed Volume 11 (2016) 5443–5455.
- [50]. Nijnik A, Hancock R, The roles of cathelicidin LL-37 in immune defences and novel clinical applications, Curr. Opin. Hematol 16 (1) (2009) 41–47. [PubMed: 19068548]
- [51]. Nugraha RV, Yunivita V, Santoso P, Aarnoutse RE, Ruslami R, Clofazimine as a treatment for multidrug-resistant tuberculosis: a review, Sci. Pharm 89 (2021) 19, 10.3390/scipharm89020019.
- [52]. Pace B, Lackner A, Porter E, Pahar B, The Role of Defensins in HIV Pathogenesis, Mediat. Inflamm 2017 (2017) 1–12.
- [53]. Panda S, Tiwari A, Luthra K, Sharma SK, Singh A, Association of Fok1 VDR polymorphism with Vitamin D and its associated molecules in pulmonary tuberculosis patients and their household contacts, Sci. Rep 9 (1) (2019) 15251. Published 2019 Oct 24. doi:10.1038/s41598-019-51803-8. [PubMed: 31649297]
- [54]. Padhi A, Pattnaik K, Biswas M, Jagadeb M, Behera A, Sonawane A, Mycobacterium tuberculosis LprE suppresses TLR2-dependent cathelicidin and autophagy expression to enhance bacterial survival in macrophages, J. Immunol 203 (10) (2019) 2665–2678. [PubMed: 31619537]
- [55]. Pillaye JN, Marakalala MJ, Khumalo N, Spearman W, Ndlovu H, Mechanistic insights into antiretroviral drug-induced liver injury, Pharm. Res Perspect 8 (4) (2020), e00598, 10.1002/prp2.598.
- [56]. Primorac D, Bach-Rojecky L, Vadunec D, Juginovi A, Žuni K, Matiši V, et al. , Pharmacogenomics at the center of precision medicine: challenges and perspective in an era of Big Data, Pharmacogenomics 21 (2) (2020) 141–156, 10.2217/pgs-2019-0134. [PubMed: 31950879]
- [57]. Rathored J, Sharma SK, Singh B, et al. , Risk and outcome of multidrug-resistant tuberculosis: vitamin D receptor polymorphisms and serum 25(OH)D, Int J. Tube Lung Dis 16 (11) (2012) 1522–1528, 10.5588/ijtld.12.0122.
- [58]. Ricci E, Malacrida S, Zanchetta M, Montagna M, Giaquinto C, De Rossi A, Role of beta-defensin-1 polymorphisms in mother-to-child transmission of HIV-1, J. Acquir Immune Defic. Syndr 51 (2009) 13–19. [PubMed: 19390326]
- [59]. Safrin S, Antiviral Agents, in: Katzung BG, Vanderah TW (Eds.), Basic and Clinical Pharmacology, 15e, McGraw Hill, 2021. Available from: <https://accesspharmacy.mhmedical.com/content.aspx?bookid=2988§ionid=250602382> [Accessed 25 April 2022.].
- [60]. Schneider JJ, Unholzer A, Schaller M, Schafer-Korting M, Korting HC, Human defensins, J. Mol. Med 83 (2005) 587–595. [PubMed: 15821901]
- [61]. Segat L, Brandao LA, Guimaraes RL, Crovella S, Are defensin beta 1 gene polymorphisms associated with HIV infection and virus replication? AIDS 23 (2009) 647–649, author reply 9–50. [PubMed: 19516112]
- [62]. Segat L, Milanese M, Boniotto M, Crovella S, Bernardon M, Costantini M, Alberico S, Italian Group SHIViO, Gynecology. DEFB-1 genetic polymorphism screening in HIV-1 positive

- pregnant women and their children, *J. Matern Fetal Neonatal Med* 19 (2006) 13–16. [PubMed: 16572527]
- [63]. Selvaraj P, Prabhu Anand S, Harishankar M, Alagarasu K, Plasma 1,25 dihydroxy vitamin D3 level and expression of vitamin D receptor and cathelicidin in pulmonary tuberculosis, *J. Clin. Immunol* 29 (4) (2009) 470–478. [PubMed: 19219539]
- [64]. Shao Y, Chen F, Chen Y, et al. , Association between genetic polymorphisms in the autophagy-related 5 gene promoter and the risk of sepsis, *Sci. Rep* 7 (1) (2017) 9399. Published 2017 Aug 24. doi:10.1038/s41598-017-09978-5. [PubMed: 28839236]
- [65]. Solanki S, Singh P, Kashyap P, Sansi M, Ali S, Promising role of defensins peptides as therapeutics to combat against viral infection, *Microb. Pathog* 155 (2021), 104930. [PubMed: 33933603]
- [66]. Song JH, Kim SY, Chung KS, et al. , Association between genetic variants in the IRGM gene and tuberculosis in a Korean population, *Infection* 42 (4) (2014) 655–660, 10.1007/s15010-014-0604-6. [PubMed: 24595493]
- [67]. Songane M, Kleinnijenhuis J, Alisjahbana B, et al. , Polymorphisms in autophagy genes and susceptibility to tuberculosis, *PLoS One* 7 (8) (2012), e41618, 10.1371/journal.pone.0041618. [PubMed: 22879892]
- [68]. Songane M, Kleinnijenhuis J, Netea M, van Crevel R, The role of autophagy in host defence against *Mycobacterium tuberculosis* infection, *Tuberculosis* 92 (5) (2012) 388–396. [PubMed: 22683183]
- [69]. Siregar Y, Sinaga B, Is there any impact of VDR gene polymorphism in bataks ethnic to have tuberculosis? *J. Health Sci* (2017) 5, 10.17265/2328-7136/2017.01.001.
- [70]. Swindells S, Lutz T, Van Zyl L, et al. , Week 96 extension results of a phase 3 study evaluating long-acting cabotegravir with rilpivirine for HIV-1 treatment, *AIDS* 36 (2) (2022) 185–194. [PubMed: 34261093]
- [71]. Teclé T, Tripathi S, Hartshorn K, Review: defensins and cathelicidins in lung immunity, *Innate Immun.* 16 (3) (2010) 151–159. [PubMed: 20418263]
- [72]. Thomas BE, Shanmugam P, Malaisamy M, Ovung S, Suresh C, Subbaraman R, Adinarayanan S, Nagarajan K, Psycho-socio-economic issues challenging multidrug resistant tuberculosis patients: a systematic review, *PLoS One* 11 (1) (2016 25), e0147397, 10.1371/journal.pone.0147397. [PubMed: 26807933]
- [73]. Tierney D, Nardell EA 2018. Tuberculosis - MSD Manual Professional Edition. [online]. Kenilworth, USA: Merck & Co; 2018. Available from: <https://www.msdmanuals.com/professional/infectious-diseases/mycobacteria/tuberculosis-tb> [Accessed 26 April 2022].
- [74]. UNAIDS. UNAIDS data 2020. 2020. Available from: <https://www.unaids.org/en/resources/documents/2020/un aids-data> [Accessed 9 February 2022].
- [75]. UNAIDS. Global HIV & AIDS statistics - fact sheet. 2022. Available from: <https://www.unaids.org/en/resources/fact-sheet> [Accessed 9 February 2022].
- [76]. Valere K, Rapista A, Eugenin E, Lu W, Chang T, Human alpha-defensin HNP1 increases HIV traversal of the epithelial barrier: a potential role in STI-mediated enhancement of HIV transmission, *Viral Immunol.* 28 (10) (2015) 609–615. [PubMed: 26379091]
- [77]. Walker NF, Scriven J, Meintjes G, Wilkinson RJ, Immune reconstitution inflammatory syndrome in HIV-infected patients, *HIV AIDS Res. Palliat. Care* 7 (2015) 49–64, 10.2147/HIV.S42328.
- [78]. Wang G, Narayana J, Mishra B, Zhang Y, Wang F, Wang C, Zarena D, Lushnikova T, Wang X, Design of antimicrobial peptides: progress made with human cathelicidin LL-37, *Adv. Exp. Med. Biol* (2019) 215–240.
- [79]. White J, 2010. Vitamin D as an inducer of cathelicidin antimicrobial peptide expression: Past, present and future. *The Journal of Steroid Biochemistry and Molecular Biology*, 121(1–2), pp.234–238. [PubMed: 20302931]
- [80]. Weinberg A, Quiñones-Mateu M, Lederman M, Role of Human β -defensins in HIV Infection, *Adv. Dent. Res* 19 (1) (2006) 42–48. [PubMed: 16672548]
- [81]. World Health Organization (WHO). Global Tuberculosis Report 2020. 2020a. Geneva, WHO.
- [82]. WHO. WHO operational handbook on tuberculosis, Module 4: treatment - drug-resistant tuberculosis treatment. Geneva: World Health Organization; 2020b.

- [83]. WHO. Global tuberculosis report 2021. 2021a. Geneva: World Health Organization.
- [84]. WHO. Tuberculosis. 2021b. Available from: <https://www.who.int/news-room/fact-sheets/detail/tuberculosis> [Accessed 9 February 2022].
- [85]. Xie H, Li C, Zhang M, Zhong N, Chen L, Association between IRGM polymorphisms and tuberculosis risk: a meta-analysis, *Medicine* 96 (43) (2017), e8189, 10.1097/MD.00000000000008189. [PubMed: 29068986]
- [86]. Xu D, Lu W, Defensins: a double-edged sword in host immunity, *Front. Immunol* (2020) 11.
- [87]. Ye X, Zhou XJ, Zhang H, Exploring the role of autophagy-related gene 5 (ATG5) yields important insights into autophagy in autoimmune/autoinflammatory diseases, *Front Immunol.* 9 (2018) 2334, 10.3389/fimmu.2018.02334. Published 2018 Oct 17. [PubMed: 30386331]
- [88]. Yu Q, Zhu X, Di W, Jiang Y, High beclin-1 expression in human alveolar macrophage significantly correlate with the bacteriologic sterilization in pulmonary tuberculosis patients, *Clin. Lab* 62 (6) (2016) 1003–1007, 10.7754/clin.lab.2015.150833. [PubMed: 27468561]
- [89]. Yuk JM, et al., ‘Vitamin D3 Induces Autophagy in Human Monocytes/Macrophages via Cathelicidin’, in: *Cell Host and Microbe*, 6, Elsevier Ltd., 2009, pp. 231–243, 10.1016/j.chom.2009.08.004. [PubMed: 19748465]
- [90]. <https://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.904.2127&rep=rep1&type=pdf>, 2013. (Accessed 23 June 2021).
- [91]. Chang TL, Klotman ME, Defensins: natural anti-HIV peptides, *AIDS reviews* 6 (3) (2004) 161–168. [PubMed: 15595433]

Further Reading

- [1]. Ahmed A, Siman-Tov G, Hall G, Bhalla N, Narayanan A, Human Antimicrobial Peptides as Therapeutics for Viral Infections, *Viruses* 11 (8) (2019) 704.
- [2]. Austin CM, Ma X, Graviss EA, Common nonsynonymous polymorphisms in the NOD2 gene are associated with resistance or susceptibility to tuberculosis disease in African Americans, *J Infect Dis* 197 (12) (2008) 1713–1716, doi:10.1086/588384. [PubMed: 18419343]
- [3]. Dini C, Bianchi A, The potential role of vitamin D for prevention and treatment of tuberculosis and infectious diseases, *Annali dell’Istituto Superiore di Sanità* 48 (3) (2012) 319–327.
- [4]. Gupta S, Bhatia G, Sharma A, Saxena S, Host defense peptides: An insight into the antimicrobial world, *Journal of Oral and Maxillofacial Pathology* 22 (2) (2018) 239. [PubMed: 30158778]
- [5]. Luong K, Nguyen L, Impact of Vitamin D in the Treatment of Tuberculosis, *The American Journal of the Medical Sciences* 341 (6) (2011) 493–498. [PubMed: 21289501]
- [6]. Mehlotra RK, Zimmerman PA, Weinberg A, Defensin gene variation and HIV/AIDS: a comprehensive perspective needed, *J. Leukoc. Biol* 99 (2016) 687–692. [PubMed: 26957215]
- [7]. Shapira Y, Agmon-Levin N, Shoenfeld Y, Mycobacterium Tuberculosis, Autoimmunity, and Vitamin D, *Clinical Reviews in Allergy & Immunology* 38 (2–3) (2009) 169–177.
- [8]. Stockton JC, Howson JM, Awomoyi AA, McAdam KP, Blackwell JM, Newport MJ, Polymorphism in NOD2, Crohn’s disease, and susceptibility to pulmonary tuberculosis, *FEMS Immunol Med Microbiol* 41 (2) (2004) 157–160, 10.1016/j.femsim.2004.02.004. [PubMed: 15145460]
- [9]. Wang C, Chen ZL, Pan ZF, et al., NOD2 polymorphisms and pulmonary tuberculosis susceptibility: a systematic review and meta-analysis, *Published* 28 (2013).
- [10]. Zhao M, Jiang F, Zhang W, et al. , A novel single nucleotide polymorphism within the NOD2 gene is associated with pulmonary tuberculosis in the Chinese Han, Uyghur and Kazak populations, *Published* 14 (2012).
- [11]. Cucu MG, Streat I, et al. , Polymorphisms in autophagy genes and active pulmonary tuberculosis susceptibility in Romania, *Rev. Romana Med Lab* 25 (2017) 47–53, 10.1515/rmlm-2017-0002.
- [12]. HIV Drug Resistance Report 2019. Geneva, Switzerland: World Health Organization; 2019.
- [13]. Lafferty M, Sun L, Christensen-Quick A, Lu W, Garzino-Demo A, Human beta defensin 2 selectively inhibits HIV-1 in highly permissive CCR6+CD4+ T cells, *Viruses* 9 (5) (2017) 111.

- [14]. Liu P, Modlin R, Human macrophage host defense against mycobacterium tuberculosis, *Curr. Opin. Immunol* 20 (4) (2008) 371–376. [PubMed: 18602003]
- [15]. Méndez-Samperio P, The human cathelicidin hCAP18/LL-37: a multifunctional peptide involved in mycobacterial infections, *Peptides* 31 (9) (2010) 1791–1798. [PubMed: 20600427]
- [16]. Pan H, Dai Y, Tang S, Wang J, Polymorphisms of NOD2 and the risk of tuberculosis: a validation study in the Chinese population, *Int J. Immunogenet* 39 (3) (2012) 233–240, 10.1111/j.1744-313X.2011.01079.x. [PubMed: 22212192]

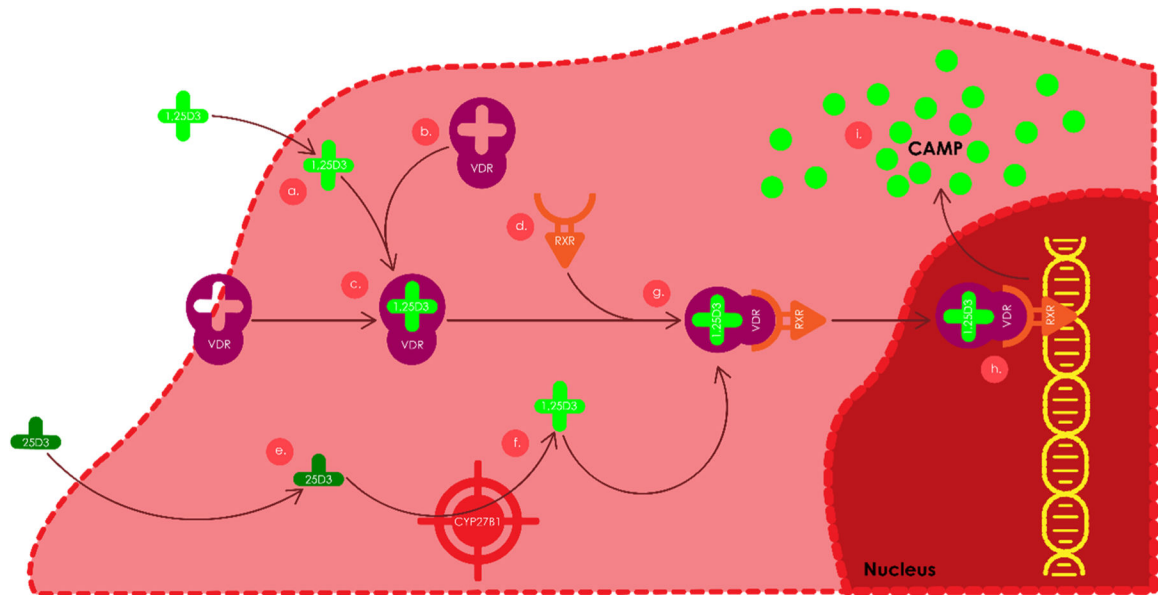


Fig. 1.

Intracellular pathways for AMP cathelicidin induction. (a.) 1,25-(OH)₂D₃ may be directly transported into the affected cell from an extracellular reservoir or a precursor, 25 hydroxyvitamin (e.) D₃ may be converted in the cytoplasm to 1,25-(OH)₂D₃ by the enzyme CYP27B1(f.). (c.)1,25-(OH)₂D₃ binds upregulated VDR. (d.) (g.) Bound VDR+ 1,25-(OH)₂D₃ binds RXR before being transported to the nucleus where it induces the expression of hCAMP (h.) (i.).

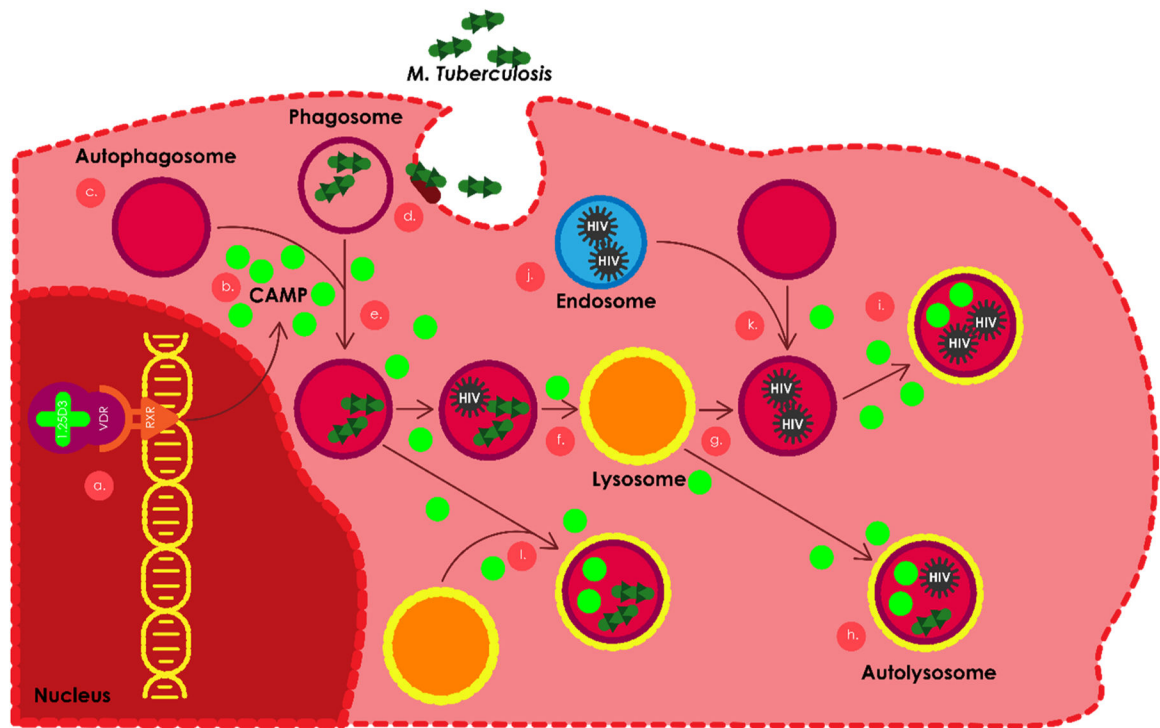


Fig. 2.

(a.) VDR+ 1,25-(OH)₂D₃ +RXR complex facilitates the expression of CAMP (b). Macrophages capture and retain pathogens through phagocytosis (d.) CAMP facilitates activation of phagosomes (e.) containing *Mtb* or endosomes containing HIV (j) (k) into autophagosomes (c.). Autophagosomes may fuse (g.) with lysosomes to form pathogen degrading autolysosomes (f.) (l.). Autophagic degradation of pathogen (HIV or *Mtb*) (h.) (i.).

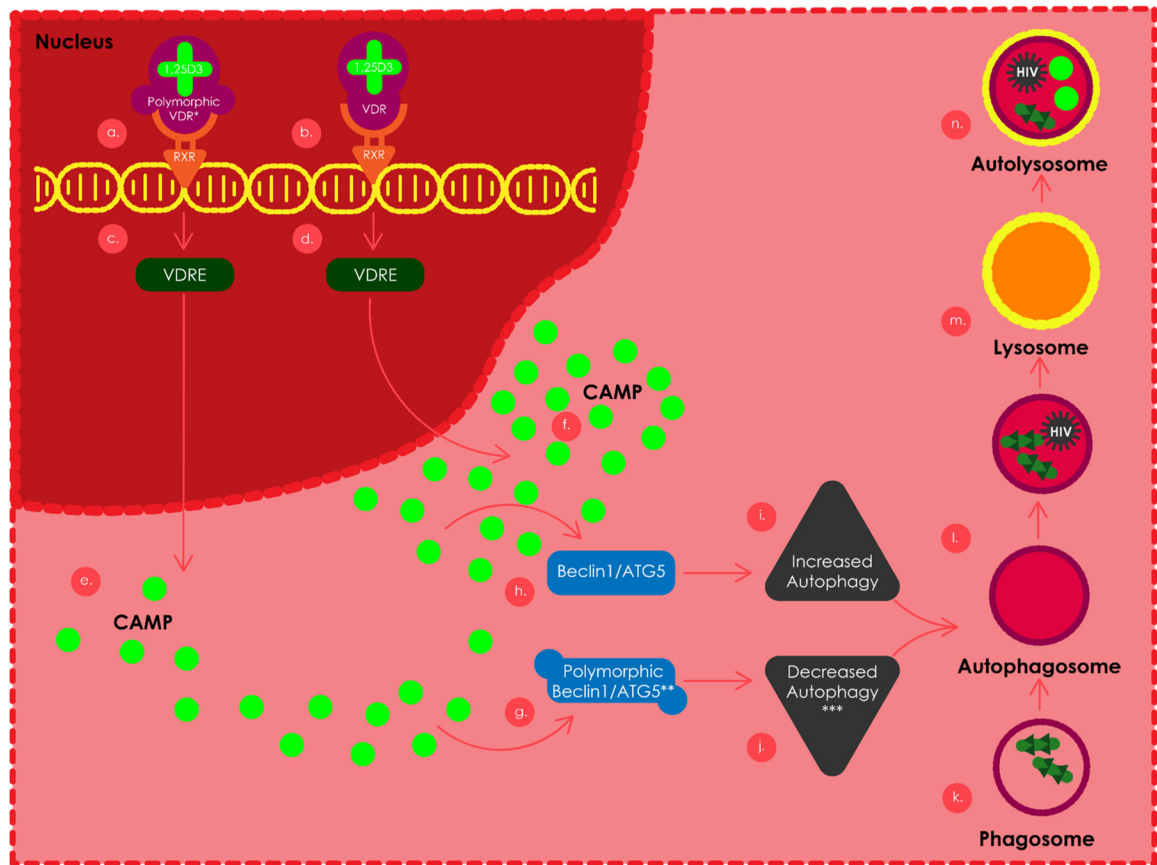


Fig. 3. Polymorphism in VDR may affect the activation of VDRE and the subsequent production of CAMP (LL-37). Reduced CAMP results in reduced induction of autophagy. Polymorphisms in Beclin 1/ATG5 may affect the induction of autophagy through Vitamin D or CAMP due to impaired response to the latter.

Table 1

Mechanism of action and limitations of current TB therapies.

Pharmacotherapy	Mechanism of action	Limitations	References
<i>TB medicines for drug sensitive TB</i>			
Rifamycin (rifampicin, rifabutin, rifapentine)	RNA synthesis inhibitor. (bactericidal)	Drug-drug interactions due to strong induction of CYP450 (rifampicin > rifabutin, rifapentine). Adverse effects include hepatotoxicity. Rifampicin resistance (<i>rpoB</i> gene mutation).	[1,11]
Isoniazid	Cell wall synthesis inhibition due to inhibition of mycolic acid. (bactericidal)	Adverse effects include hepatotoxicity and peripheral neuropathy due to vitamin B6 deficiency, with a higher risk in slow acetylators. Isoniazid resistance (<i>inhA</i> and <i>katG</i> gene mutation)	[1,11]
Ethambutol	Cell wall synthesis inhibitor due to inhibition of mycobacterial arabinosyl transferases. (bacteriostatic)	Adverse effects include retrobulbar neuritis leading to red-green color blindness. Requires renal dose adjustments in renal impairment. Bacteriostatic mechanism of action.	[11]
Pyrazinamide	Cell membrane inhibitor. (bacteriostatic)	Adverse effects may include hepatotoxicity and hyperuricemia.	[11]
<i>TB medicines for drug resistant TB and/or second line therapy</i>			
Fluoroquinolones (moxifloxacin, levofloxacin, gatifloxacin)	DNA synthesis inhibitor.	Adverse effects include aortic aneurysm, tendonitis, tendon rupture, hypoglycemia, mental health side effects (disturbance in attention, disorientation, agitation, nervousness, memory impairment, delirium). Fluoroquinolone resistance (<i>gyrA</i> gene mutations)	[1,26,27]
Aminoglycosides (kanamycin, amikacin, streptomycin)	Protein synthesis inhibitors.	Adverse effects include ototoxicity and nephrotoxicity. Only available as injectables. Aminoglycoside resistance (<i>rrs</i> gene mutation)	[1,11]
Thionamides (ethionamide, prothionamide)	Cell wall synthesis inhibitor.	Adverse effects include gastric irritation which is dose dependent.	[48,11]
P-aminosalicylic acid (PAS)	Folate synthesis antagonist.	Adverse effects include hypersensitivity reactions.	[11]
Clofazimine	Not clearly understood. Multiple mechanisms have been explored-release of toxic enzymes and reactive oxygen, competing with a cofactor in mycobacteria (menaquinone).	Adverse effects include skin discoloration, QT prolongation, and gastrointestinal intolerance.	[48,51,82]
Cycloserine, Terizidone	Cell wall synthesis inhibitor.	Adverse effects include peripheral neuropathy, central nervous system side effects (depression, psychosis, lethargy).	[48,82]
Linezolid	Protein synthesis inhibitor.	Adverse effects include bone marrow suppression and neuropathy. Risk of serotonin syndrome when used with selective serotonin reuptake inhibitors.	[11]
Bedaquiline	Inhibits ATP synthase pump depleting energy stores.	Drug-drug interactions (CYP3A4 metabolism). Adverse effects include hepatotoxicity and QT prolongation. High pill burden.	[11]
Nitro-imidazole derivatives (delamanid, pretomanid)	Inhibit mycolic acid synthesis.	Adverse effects include QT prolongation and hepatotoxicity.	[48,92]


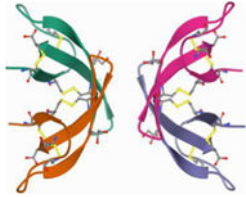
Table 2



Mechanism of action and limitations of current HIV therapies.

Pharmacotherapy	Mechanism of action	Limitations	References
Nucleoside reverse transcriptase inhibitors (abacavir, lamivudine, emtricitabine, tenofovir disoproxil fumarate (TDF), tenofovir alafenamide (TAF), zidovudine)	Inhibits HIV reverse transcriptase preventing RNA to DNA synthesis.	Abacavir: HLA-B* 5701 screening required before initiation due to the risk for hypersensitivity if a carrier. TDF: worsening effects on bone and kidney relative to TAF. TAF: weight gain, worsening lipids, contraindications with rifamycins. Zidovudine: Macrocytic anemia, neutropenia, fatigue, lipodystrophy.	[22,59]
Non-nucleoside reverse transcriptase inhibitors (efavirenz, nevirapine, rilpivirine, etravirine, doravirine)	Inhibits HIV reverse transcriptase by allosteric binding to enzyme preventing RNA to DNA synthesis.	Efavirenz: central nervous system adverse effects including exacerbation of depression, and QT prolongation; drug-drug interactions (CYP3A4, CYP2B6 inducer); food requirement (empty stomach); low genetic barrier to resistance. Nevirapine: adverse effects such as hepatotoxicity and rash; drug-drug interactions (CYP3A4, CYP2B6 inducer); low genetic barrier to resistance. Rilpivirine: central nervous system adverse effects and QT prolongation; contraindications with proton pump inhibitors; food requirement (take with food); low genetic barrier to resistance. Etravirine: twice daily dosing; multiple drug-drug interactions (CYP3A4 inducer and CYP2C9/ CYP2C19 inhibitor).	[22,59]
Protease inhibitors (atazanavir, lopinavir/ritonavir, darunavir)	Inhibits HIV protease preventing cleavage of polyprotein into individual subunits responsible for assembly of new virions.	Multiple drug-drug interactions; increased pill burden for patients requiring twice daily administration (<i>i.e.</i> treatment-experienced patients); food requirement with atazanavir and darunavir (take with food) to improve bioavailability. Atazanavir: hyperbilirubinemia Lopinavir/ritonavir: gastrointestinal adverse effects (diarrhea), QT prolongation and lipodystrophy. Darunavir: sulfonamide (rash), gastrointestinal adverse effects (nausea, vomiting, diarrhea) and hepatotoxicity.	[59,22]
Integrase strand transfer inhibitors (bictegravir, dolutegravir, raltegravir, elvitegravir, cabotegravir)	Inhibits HIV integrase preventing integration of HIV DNA into CD4 cell genome (<i>i.e.</i> preventing formation of HIV DNA provirus in viral reservoirs).	Bictegravir: diarrhea, nausea, headache and weight gain. Dolutegravir: insomnia, headache, weight gain and hepatotoxicity. Raltegravir: headache, insomnia, muscle weakness, rash, nausea, diarrhea, weight gain; low genetic barrier to resistance. Elvitegravir: nausea, diarrhea, headache; food requirement (take with food); low genetic barrier to resistance. Cabotegravir: headache, nausea, abnormal dreams, anxiety, insomnia, depressive disorders and hepatotoxicity.	[59,22]
Entry inhibitors (fostemsavir, ibalizumab, maraviroc, enfuvirtide)	Fostemsavir: gp-120 directed attachment inhibitor Ibalizumab: humanized IgG4 anti-CD4 monoclonal antibody Maraviroc: CCR5 antagonist Enfuvirtide: binds to HIV envelope glycoprotein gp41 preventing fusion of HIV with CD4 cell membrane	Fostemsavir: adverse effects include nausea, QT prolongation, increase in liver function tests, bilirubin elevation, sleep disturbance and dizziness; twice daily dosing. Ibalizumab: requirement for intravenous administration; cost (expensive); adverse effects include diarrhea, dizziness, nausea, rash and hypersensitivity. Maraviroc: requirement for CCR5 tropism testing (only effective for CCR5 tropic virus); twice daily dosing; hepatotoxicity. Enfuvirtide: requirement for twice daily subcutaneous administration; injection site reactions common.	[22]

Overview of antimicrobial peptides: three-dimensional structure, site of expression, and antimicrobial activity.

Table 3

Antimicrobial peptide	Three dimensional peptide structure*	Site of expression	Antimicrobial activity	References
Cathelicidin (<i>LL-37</i>)		Neutrophil specific granules and epithelial cells. Inducible in immune cells through vitamin D receptor	Broad spectrum antimicrobial activity (<i>active against gram – and gram + strains</i>) including <i>M.tb</i>	[5,8,50]
α -defensins (HNP 1–4)		Constitutively expressed in azurophilic granules of neutrophils. Found in granulocytes, NK cells, B-cells, $\gamma\delta$ T-cells or monocytes/macrophages	HIV- 1 (in <i>vitro</i>) Serum dependent and salt concentration independent	[4,91]

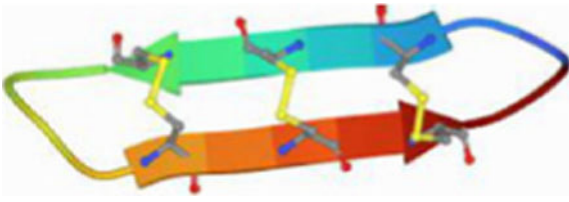
Antimicrobial peptide	Three dimensional peptide structure*	Site of expression	Antimicrobial activity	References
α -defensins (<i>HDS</i> and <i>HD6</i>)		Constitutively expressed in Paneth cells. Found in the salivary gland, stomach and eyes	HIV- 1 (in <i>vitro</i>) Serum dependent and salt concentration independent	[4,18,91]
β -defensins (<i>hBD 1-4</i>)		Epithelial cells hBD 1 expressed constitutively. hBD 2-4 are inducible e.g. pathogenic induction. Found in oral and nasal mucosa, lung, plasma, salivary gland, GI, skin, eye, mammary gland, UGT and kidney	HIV- 1 (in <i>vitro</i>) Serum dependent and salt concentration dependent	[4,18,91]

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Antimicrobial peptide	Three dimensional peptide structure*	Site of expression	Antimicrobial activity	References
θ -defensins (<i>Retrocyclins</i>)		Synthetic derivatives obtained from Primate bone marrow.	HIV- 1 (<i>in vitro</i>)	[4,9]

* The structures shown are those of LL-37, HNPI, HD6, hBD2 and retrocyclin-2 obtained on the PDB with ID#s: 2K60, 3GNY, 1ZMQ, IFD3, and 2LZI respectively.