The Role of Lipid and the Benefit of Statin in Augmenting Rifampicin Effectivity for a Better Leprosy Treatment

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Abstract

BACKGROUND: Although leprosy remains as a serious disease of the skin and nervous system, the current treatment is still lacking in its effectiveness.

AIM: This literature review will explore the association of lipid and leprosy, as well as the potential of statin and other lipid-lowering agents as adjunctive drugs to combat leprosy.

MATERIALS AND METHODS: Articles were searched through the PubMed, EBSCOhost, and Google Scholar with the keywords: immunomodulation, lipid-body, lipids, leprosy, Mycobacterium leprae, pathogenesis, rifampin or rifampicin, and statins. A manual searching is also carried out to find an additional relevant information to make this literature review more comprehensive.

RESULTS: The literatures showed that lipids are highly correlated with leprosy through alterations in serum lipid profile, metabolism, pathogenesis, and producing oxidative stress. Statins can diminish lipid utilization in the pathogenesis of leprosy and show a mycobactericidal effect by increasing the effectiveness of rifampicin and recover the function of macrophages. In addition, Statins have anti-inflammatory properties which may aid in preventing type I and II reactions in leprosy. Standard multidrug therapy might reduce the efficacy of statins, but the effect is not clinically significant. The statin dose-response curve also allows therapeutic response to be achieved with minimal dose.

CONCLUSION: The various pleiotropic effects of statins make it a potential adjunct to standard treatment for leprosy in the future.

Introduction

Leprosy, commonly known as Hansen's disease or Morbus Hansen (MH), is a chronic granulomatous infection caused by *Mycobacterium leprae*, which predominantly manifests in the skin and nervous system. Leprosy is generally dreaded due to its potential to cause body deformities and disabilities. The initial symptoms of leprosy often manifest only as hypopigmented macule. In a leprosy reaction, other general clinical signs may appear, such as chills, anorexia, sometimes accompanied by an enlarged liver-spleen, nephritis, and neuritis [1]. In 2016, the global prevalence of leprosy was 0.29 per 10,000 population,[2] while in Indonesia, the figure was higher, 0.7 per 10,000 population in 2017 [3]. There were 16,826 new cases in 2016 and 15,920 new cases in 2017, placing Indonesia as the country with the third-highest incidence of leprosy in the world after India and Brazil [2]. Late diagnosis of leprosy can cause irreversible neurological disability, and its clinical manifestations are often stigmatized, thus affecting the physical, economic, and social life of the sufferer. Early diagnosis and effective treatment are needed to prevent complications [4].

Several challenges arise in leprosy eradication, such as increasing drug resistance cases due to withdrawal from multidrug therapy (MDT), therapy failure after completing MDT resulting relapse cases, and MDT has not been effective in killing persistent leprosy bacteria ultimately [4]. In a prospective survey of leprosy endemic countries from 2009-2015, leprosy's global resistance rate was 8% of the 2000 reported cases. Of these cases, 74 were resistant to rifampin, 87 to dapsone, and 21 to ofloxacin [5]. Twenty patients were resistant to the multidrug rifampin and dapsone, and four patients resistant to the multidrug ofloxacin and dapsone in Brazil, India, and Indonesia [5].

A notable characteristic of most relapsing or reactivation leprosy cases is the presence of granulomatous infiltration regardless of treatment, indicating drug resistance. One possible solution to counter the resistance is drug combinations which may also reduce the rate of recurrence, increase the bactericidal effect, and exert anti-inflammatory effects on leprosy [6]. In various studies, statins have several pleiotropic effects, such as potential bacterial growth inhibitors [7], [8], [9], [10], augmentation of other bactericidal drugs [11], and anti-inflammatory effects [12], [13], [14], [15], [16], [17].

This article aims to provide a comprehensive insight to the link between lipid and leprosy, as well as a more in-depth discussion of statins' role in leprosy treatment, focusing their benefits on enhancing the mycobactericidal effects of rifampicin and its potential as an anti-inflammatory for various leprosy inflammatory pathways. Hopefully, this review will lead to further research into the investigation of statins as a therapeutic adjuvant in the leprosy program.

Methods

This narrative review was arranged in December 2020 by searching the 20 years of sources (due to limited literature) regarding more effective leprosy treatment using the PubMed, EBSCOhost, and Google Scholar databases journal to manual hand searching through references in articles in a snowball manner. We used the keywords combination "body-lipids," "immunomodulation," "pathogenesis," "leprosy," "lipids," "_Mycobacterium leprae_," "rifampin," and "statins" using Booleans AND/OR/NOT, as well as asterisk, parentheses, and quotation marks to enhance specificity and sensitivity.

We obtained 448 hits in PubMed, and 140 appears in EBSCO, which were screened for abstracts and titles using inclusion criteria: Articles in the form of original articles with evidence levels 1-5, published in English, correlated with writing papers to discuss lipid relationships, leprosy, and statins, and in publications in the past 20 years. Due to the limited number of studies, the authors decided the inclusion criteria should not be too strict concerning publication time, level of evidence, and study design. The article was excluded if it is a double, its publication is older than 2000, only discuss tuberculosis-type mycobacteria without leprosy, and is not related to the topic. At the end of the selection process, 15 articles were reviewed and assessed for validity, importance, and applicability aspect using the checklist of appraisal from the Joanna Briggs Institute, University of Adelaide 2017 [18]. Our findings are summarized in a table which notes the evidence level based on the Oxford Center for Evidence-Based Medicine [19].

We investigated (1) the role of lipids in leprosy, (2) leprosy immunopathogenesis related to lipid bodies, (3) leprosy treatment challenges, (4) benefits of statins in leprosy by several mechanisms, and (5) the pharmacology aspect of statin on co-administration with MDT. Supporting literature was also searched manually with various relevant keywords without limiting publication year. From the collection of obtained articles and references, a comprehensive literature review is synthesized.

Results

Through our systematic approach to obtain the literatures, most of the studies are considered valid, necessary, and applicable to this review even though we assessed the level of evidence of which mainly in low-level evidence (ranging from 4 to 5) due to no available systematic review or meta-analysis yet [18]. The evidence we collected are published in the year 2000 to 2020 and comprised of seven cross-sectional studies and two case series with the level of evidence 4, three experimental studies _in vivo_, and three bench types of research with evidence level in the low position [19]. However, the outcomes which described in all studies are essential and opens a new perspective of leprosy treatment by lipids control.

The result of the studies has been sum up in Table 1. Among the 15 studies, we summarize that the main topics discussed related to lipid and leprosy include: (1) Serum lipid profile alterations in leprosy in six studies [20], [21], [22], [25], [27], [28] (2); lipid metabolism in leprosy and role of their metabolites [23], [24], [32]; (3) lipid body formation and immunopathogenesis [10], [30], [31]; (4) lipid related to oxidative stress in leprosy [26]; and (5) the impact of statin in leprosy treatment regarding evidence of lipid involvement in the immunopathogenesis of leprosy [11]. We found some correlations; lipid metabolism and formation of lipid body or lipid droplet (LB) may also reflect on changes to overall blood lipid profile, and more severe variants of leprosy manifestation show the most changes to lipid metabolism. However, some studies show inconsistent data about changes in serum profile regarding types of leprosy. The lipid metabolism and lipid body formations may be related to the overproduction of stressful oxidative agents which more are prone to cause inflammation. Statins can potentially solve all lipid roles in the pathogenesis of MH as lipid-lowering agent and anti-inflammation as a pleiotropic effect.

Discussion

The role of lipid in leprosy infection

Five most apparent facts elucidated by the obtained studies are: Alterations of serum lipid profile in leprosy patients [20], [21], [22], [25], [27], [28], lipid metabolism and its metabolites [23], [24], [32], lipid bodies formation [10], [30], [31], and oxidative stress result from lipid metabolism enhanced by MH is proportional to the severity of the infection and the survival of _M. leprae_ itself [26]. Furthermore, interestingly statin found has benefit in leprosy treatment regarding lipid involvement in the immunopathogenesis of leprosy [11].

Table 1: Summary findings of studies related between leprosy and lipid metabolism

Authors, Year	Study design	LoE	Outcome measured	Details of methods	Sample size	Finding related lipid metabolism in leprosy
Lemes *et al.*, 2020 [20]	Cross-sectional	4	HDL composition and biological activities from PB and MB leprosy patient before and after MDT	Sample from control, MB, and PB patients that has and has not gone through MDT is tested for plasma Cho profile	39 samples: 12 controls; MB leprosy patients (8 pre-MDT and 6 post-MDT); PB leprosy patients (6 pre MDT and 7 post-MDT); plus one sample from liver tissue of MB leprosy	HDL lower in pre-MDT MB leprosy compared to control group (p≤0.0001) and significantly higher in post-MDT compared to pre-MDT MB leprosy. Overall MB leprosy patients show lower level of HDL-cholesterol HDL in MB pre-MDT patients impaired in function compared to control as anti-oxidative stress, anti-inflammatory, and a lower cholesterol acceptor capacity (p ≤ 0.05) MB pre-MDT patients showed lower concentrations of ApoA-1 (major HDL protein), compared to control (p ≤ 0.0001) and to post-MDT recovery (p ≤ 0.001). Hepatic involvement in leprosy is indicated by observed bacilli in the liver of a MB patient
Silva DSD *et al.*, 2018 [21]	Cross-sectional	4	Lipid-like clot formed in leprosy patients	Plasma samples are taken and analyzed for the presence of clots	638 leprosy patients and generated clots from 6 patients uninfected by *Mycobacterium leprae* as control	Among 638 patients, 35 patients (5.48%) are found having lipid-like clot (leprosum clot), 16 of which (45.7%) is LL pole. Leprosum clot is commonly correlated with the MB pole (p < 0.0001). Leprosum clot had significantly higher lipid content (p < 0.0001) compared to control; and present ten-fold more ChoE and TGL than control clots MB leprosy has low levels of serum total Cho and HDL, while TGL, LDL and VLDL were normal
Sarwar *et al.*, 2016 [22]	Cross-sectional	4	Lipid profile in MB and PB leprosy patients	Measurement of total Cho, LDL, HDL, and TGL	Newly diagnosed leprosy patients (24 MB and 18 PB), plus 30 controls	LDL, total Cho, and TGL was significantly decreased in both PB and MB patients (p < 0.05) HDL significantly increased (p < 0.05) in both MB and PB patients HDL in patients is increased might be due to host response to fix impaired innate immune response through role of oxidative stress scavenger molecules
Amaral *et al.*, 2013 [23]	Cross-sectional	4	Level of metabolites present in leprosy patients before and after MDT	Analysis and profiling serum metabolomic of leprosy patients before and after complete MDT	29 LL patients, 29 BT patients, 10 controls	PUFA metabolism is highly affected in leprosy, namely omega-3 and omega-6 FA, suppressing inflammation Analysis of highly infected skin lesions revealed changes on the levels of lipids derived from lipase activity, such as PUFA, indicating high lipid turnover. No statistical significance was found (p > 0.05). Comments: Lack of statistical significance may be due to low sample size
Al-Mubarak *et al.*, 2011 [24]	Cross-sectional	4	Serum metabolomics in LL patients	50 μL from each serum sample was taken to analyzed by UPLC-MS to measure primary metabolomic EPA, DHA, and AA	23 Patients serum, comprising: with BI < 1 (10 patients) and BI > 4 (13 patients)	Significant increases in n-3 PUFAs (EPA and DHA), and the n-6 PUFA AA in the high-BI patients with the median values of EPA (2.6-fold increase), DHA (1.6-fold increase), and AA (2-fold increase), were found to be greater in patients with high-BI compared to the low-BI patients EPA and DHA have anti-inflammatory properties, while AA has both pro- and anti-inflammatory activities. The increase of several lipids in high-BI patients may imply the biological pathways These PUFAs could serve as biomarkers, useful for infection susceptibility evaluation, early diagnosis, and as monitor for disease progression
Nwosu *et al.*, 2001 [25]	Cross-sectional	4	Total Cho and TGL from blood samples	Leprosy patients are given questionnaire to determine dietary habits and blood samples were taken.	Unclear, only stated 30 control subjects	There is significant difference in total Cho (p < 0.05) and TGL (p < 0.005) between leprosy group. TGL diminishes in spectrum of LL>BL>TT Comments: Little of the samples or the result can be inferred from it.
Bhadwat *et al.*, 2000 [26]	Cross-sectional	4	Plasma MDA as a biochemical marker of lipid peroxidation and activity of SOD	Blood samples are taken to analyzed Inclusion Criteria: Diagnosed with leprosy clinically and bacterially, age 31-40 y.o. Control group must not be on any drugs Exclusion criteria: smoking, RA, DM, concomitant bacterial infections	70 leprosy patients including 18 of LL, 16 of BL, 10 of BB, 14 of BT and 12 of TT patients. Also 15 normal subjects as a control groups	MDA was lowest (3.225 ± 0.24) in TT cases and highest (4.516 ± 0.54) in LL cases indicating increased lipid peroxidation in LL due to high rate of ROI, meanwhile scavenging activity by antioxidant is not adequate. Also it can be caused by cell-mediated immunity in TT stronger than LL The reverse is true for SOD activity level, highest in TT (34.05 ± 2.46 units/mg Hb) and lowest (25.84 ± 5.67 units/mg Hb) in LL. Erythrocyte SOD is an important catalytic protein which specifically scavenges superoxide radicals and become one of the major antioxidant enzyme. In LL level of SOD weaker than TT The mean ratio of MDA/SOD was lowest in TT (0.095) while it was highest (0.185) in LL (p < 0.001). Results points to an increase in oxidative stress particularly in LL cases. Comments: This studies implies correlation of lipid peroxidation, antioxidant level (e.g HDL), and leprosy related stress oxidative especially in LL
Silva *et al.*, 2017 [27]	Case series	4	Lipid profile and CVD outcomes in leprosy patients during treatment	Data which taken from PB and MB leprosy patients after undergoing MDT: Socio-demographic data, anthropometry, food intake, and lipid profile Inclusion criteria: Completed MDT 2 – 5 years ago; follow up regularly Exclusion criteria: Not match with age; incomplete data; has DM, HIV, recurrent or reactional state of leprosy, diagnosed dyslipidemia before leprosy; use of corticosteroid	84 adults, ranging in age 20 – 60 y.o with PB and MB leprosy after treatment	On PB and MB leprosy respectively: 　Mean LDL (mg/dL) was 116.9 and 121 　Male HDL was 51.7 and 52.8 　Female HDL was 50.2 and 53.5 　Total Cho was 193.8 and 203.5 　TGL was 123.4 and 147.4 Comments: 　This study excluded patients diagnosed with dyslipidemia before leprosy 　The risk of leprosy patients developing CVD was low after treatment considering the lipid profile might be due to use of lipid in ML infection massively as a carbon source to survive 　However, a lot of patients are grouped to be high risk for CVD due to excess weight, so it is essential to implement healthy lifestyle in leprosy patients

(Contd...)

Table 1: (*Continued*)

Authors, Year	Study design	LoE	Outcome measured	Details of methods	Sample size	Finding related lipid metabolism in leprosy
Gupta *et al.*, 2002 [28]	Case-series	4	Total cholesterol, LDL, VDL, HDL, chylomicrons	Study group are tested for leprosy clinically, bacteriologically, and immunologically. After grouping, 5 ml of blood is drawn and analyzed for Cho content	50 untreated leprosy cases: 21 cases of BT, 12 of BB, 7 of BL, 9 of LL, and 1 case of TT leprosy	The serum TGL is lower in TT, no changes in BT or BB, and not significantly increased in BL and LL patients The total Cho is lower in TT increased and decreased in BB, BT, BL, and LL cases significantly VLDL is raised in LL leprosy significantly, not significantly in BB cases, and in normal value in TT, BT, and BL patients HDL is decreased significantly in BT and LL cases and insignificantly decrease in BL, within normal in TT and BB Comments: No correlation is drawn in this study between lipid fractions and the type of leprosy. The result in this study seems like not consistent with other studies
Miyamoto Y *et al.*, 2016 [29]	Experimental *in vivo*	5	Intracellular metabolite in ML infected mice by CE-MS analysis	ML strain is injected to nude mice footpad and their metabolite profile is identified, then compared with *M. bovis* culture	Three mice infected with ML and three uninfected mice.	There are 193 ML-derived metabolites; 84 of which are common to ML and *M. bovis*, and 22 of them have 10-fold difference in mean ratio Central carbon metabolism of ML markedly decreased, suggesting certain metabolic characteristic to carbon that is unique to ML Comments: In this study the role of lipid is unclear, only mention role of carbon as a part of lipid chains
Lobato *et al.*, 2014 [11]	Experimental *in vivo*	5	Effect of rifampin with atorvastatin in bacterial viability and cholesterol profile in infected mice	Mice are divided into 6 groups: (1) control; (2) treated by 40 mg/kg atorvastatin; (3) treated by 80 mg/kg atorvastatin; (4) treated by 10 mg/kg rifampin; (5) treated by 1 mg/kg rifampin; and (6) treated by 1 mg/kg rifampin plus 80 mg/kg atorvastatin.	6 groups of mice with unclear number of mice included	Statin reduce (75%) intracellullar mycobacterial viability but does not alter mycobacterial invasion High dose of atorvastatin alone (80 mg/kg/day) was effectively reduced ML replication and only this high dose showed significant decreased of plasma Cho levels compared to control Atrovastatin or simvastatin synergizes with rifampin for its antibacterial effect and reduced plasma Cho compared to rifampin alone (p < 0.01). The inhibition of Cho de novo synthesis which essential for cell wall of mycobacteria might kill them by carbon deprivation and make them more vulnerable to rifampin
Mattos *et al.*, 2010 [30]	Experimental *in vivo*	5	LB in macrophages and possible mechanism to its formation	Macrophages are obtained from infected and uninfected mice, which are evaluated using microscope, FACS cytometry, and IHC analysis of ADRP	C57BL/6J mice with unclear of number and four skin biopsies of LL patients	Both Cho and ChoE were found higher in LL skin biopsies compared to control (p < 0.05). It can be concluded that ML infection triggers a modulatory response to Cho homeostasis, leading to its intracellular accumulation as LB, enriched witj free Cho and ChoE LB formation per macrophages are found to be significantly higher in infected murine and human ML-infected (p < 0.05) Several host genes: ADRP, CD36, and selective lipid receptor (TLR2, TLR6, and cytoskeleton), are up regulated which are essential for lipid metabolism and LB formation by ML-infected cells
Mattos *et al.*, 2014 [10]	Bench research	5	Level of LDL receptors expression and enzymes for Cho biosynthesis in lepromatous tissues	Using PCR, fluorescence, TEM, bacterial viability kit, and flow-cytometry to investigate lipid accumulation in ML-infected tissues; intracellular Cho sources in macrophages; SREBP expression; level of LDL receptor; LB in macrophages; and viability of ML	Skin biopsies of 6 LL patients and 4 BT patients, not stated clearly number of sample	ML can induce the formation of LB in infected macrophages Cho is one of the host lipid molecules that is higher in ML-infected macrophages, but FA and TGL are higher in uninfected cells Expression of LDL receptors and enzymes involved in biosynthesis of Cho was higher in LL tissues compared to BT lesions SREBP transcriptional factors as a key regulator of Cho biosynthesis and uptake of cellular Cho were also found to be higher in LL tissues Macrophages infected by ML synthesize Cho with higher capacity and sequester exogenous LDL Both Cho de novo synthesis inhibition by statins and depletion of exogenous Cho was found to reduce intracellular survival of ML There is significantly higher ADRP expression in LL compared to BT samples
Mattos *et al.*, 2011 [31]	Bench research	5	Parameters of biogenesis in LB formation by ML in SC and how it contributes to the innate immune response	Nerve biopsy samples, strain from infected mice footpads, and Schwann cell cultures are examined with IHC analysis and LB staining.	Nerve biopsy specimens from LL patients, SC cultures, and C57BL/6 mice with unclear number of samples	The induction of LB formation in SC requires the uptake of live ML in manner depend on dose and time in level of multiplicity of infection of 50 after 24 – 48 h TLR6 has more critical role than TLR2 to induce LD formation in SC ML-induced LB biogenesis associated with high PGE2 and IL-10 secretion, and decreased number of IL-12 and NO production in ML-infected SC Fatty acid synthase inhibitor C-75 nullifies the effect of ML on immunoinflammatory mediators production, thus enhancing the anti-mycobacterial ability of SC
Cruz *et al.*, 2008 [32]	Bench research	5	Lipid metabolism profile in leprosy	Examination of host gene expression across the disease spectrum	Skin biopsies of leprosy patients, not stated clearly number of sample	To survive, ML evade the host's immune system and uses the host's metabolic pathways to obtain necessary nutrients, which includes lipid (FA and phospholipids) In LL lesions, it is preferred host lipid metabolism genes such as a group of phospholipases (host-derived oxidized phospholipids and one specific oxidized phospholipid PEIPC) Lipid metabolism accounts for 11% of the upregulated gene in LL lesions, 4-fold more sizeable than TT. Lipid metabolism is significantly higher than protein metabolism in LL lesions, and the reverse is true for TT lesions Mycobacterial inhibits innate immune responses using the accumulated host-derived oxidized phospholipids. HDL enhances immune response instead as a scavenger of oxidized phospholipids. However, HDL from LL patients was found less functional compared to that of TT patients

AA: Arachidonic acid, ADRP: Adipose differentiation-related protein, ApoA-1: Apolipoprotein A-I, BB: Borderline borderline, BI: Bacterial index, BT: Borderline tuberculoid, BL: Borderline lepromatous, CD36: Cluster of differentiation 36 gene, CE-MS: Capillary electrophoresis-mass spectrometry, Cho: Cholesterol, ChoE: Cholesterol ester, CVD: Cardiovascular disease, DHA: Docosahexaenoic acid, DM: Diabetes mellitus, EPA: Eicosapentaenoic acid, FA: Fatty acids, FACS: Fluorescence-activated cell sorting, HDL: High-density lipoprotein, HIV: Human immunodeficiency virusm, IHC: Immunohistochemistry, IL: Interleukin, LB: Lipid bodies also known as lipid droplets, LDL: Low-density lipoproteins, LL: Lepromatous leprosy, LoE: Level of evidence, *M. bovis: Mycobacterium bovis*, MB: Multibacillary leprosy, MDA: Malondialdehyde, MDT: Multidrug therapy, ML: Mycobacterium leprae, NO: Nitric oxide, PB: Paucibacillary leprosy, PCR: Polymerase chain reaction, PEIPC: 1-Palmitoyl-2-(5,6-epoxyisoprostane E2)-sn-glycerol-3-phosphorylcholine, PGE2: Prostaglandin E2, PUFA: Polyunsaturated fatty acids, RA: Rheumatoid arthritis, ROI: Reactive oxygen intermediates, SC: Schwann cells, SOD: Superoxide dismutase, SREBP: Sterol regulatory element-binding proteins, TEM: Transmission electron microscopy, TGL: Triglycerides, TLR: Toll-like receptors, TT: Tuberculoid leprosy, UPLC-MS: Ultrahigh pressure liquid chromatography mass spectrometry, VLDL: Very low-density lipoproteins.

Related to serum lipid profile in leprosy, a study from Sarwar *et al*. [22] found decreased total cholesterol, low-density lipoproteins (LDL), and triglyceride as well as an increased high-density lipoproteins (HDL) on newly diagnosed multibacillary (MB) and paucibacillary (PB) leprosy patients. A Study from Lemes *et al*. [20] compares HDL serum level between pre- and post-MDT treatment. The result shows lower HDL level in pre-MDT MB leprosy than the control group. Conversely, higher level of HDL was observed in post-MDT MB leprosy than on standard control. They also observed lower expression of apolipoprotein A-1 (ApoA-1) in pre-MDT MB leprosy compared to health control. Changes expression of ApoA-1 also found in *M. leprae* infected hepatic cells compared to non-infected, possibly due to the improvement of immune response and increased production of HDL that scavenge oxidative stress. Interestingly though, HDL from MB pre-MDT patients showed dysfunction, having their anti-inflammatory and anti-oxidative stress properties impaired, and their cholesterol acceptor capacity reduced [20].

Previous study from Cruz *et al*. [32] also shows HDL dysfunction in both LL and tuberculoid leprosy (TT), although the dysfunction is more apparent with the former. The dysfunction leads to reduction of dendritic cell differentiation instead of its promotion, thus increasing bacterial survivability. A study of Nwosu *et al*. [25] showed that total cholesterol and triglyceride among leprosy groups were significantly different, specifically in triglycerides decreasing in the spectrum of LL > borderline lepromatous (BL) > TT [25]. Meanwhile, study of Gupta *et al*. found an inconsistent lipid profile among leprosy groups and no correlations between lipids fractions and the type of leprosy [28]. The mechanism behind the changes of serum lipid profile is due to the usage of host lipids for virulence and growth of *M. leprae*. Interestingly, this lipid profile is also related with cardiovascular disease (CVD) risk in leprosy patients. Silva DSD *et al*. on their study mentioned that leprosum lipid-like clot contains higher levels of lipid, ten-fold more cholesterol esters, and triglycerides in line with a lower total serum of cholesterol and HDL; meanwhile, other lipid profiles are standard. Leprosum clot is formed by several mechanisms, such as (1) induction of antiphospholipid antibodies by *M. leprae*, (2) activation of anaphylatoxins (complement C4), and (3) induction of inter-alpha-trypsin inhibitor family heavy chain-related protein, all of which are associated with vascular abnormalities and procoagulant exacerbation [21]. On the other hand, Silva *et al*. [27] came to a different result, in that alteration in lipid profile of leprosy patients is not correlated to CVD risk.

Changes in serum lipids are possibly related to lipid metabolism and its metabolites which also occurs in leprosy infection. Regarding lipid metabolism, carbon chain and *M. leprae*-derived metabolites decreases in leprosy, according to the study of Miyamoto *et al*. [29] This study also revealed that not only carbon chains, amino acids were also affected. A study by Cruz *et al*.

[32] concludes that to survive, *M. leprae* impairs the host immune system and reach host lipid metabolic pathways such as a group of phospholipases and upregulated lipid metabolism in LL, which is greater than TT. In addition, similar results were found when examining host gene expression across different leprosy spectrum; lipid metabolism is higher than protein metabolism in lepromatous cases but lower in tuberculoid cases. These findings indicate that the more severe variants of leprosy manifestation show the most changes to lipid metabolism. Further related to lipid metabolites, a study from Amaral *et al*. [23] shows that polyunsaturated fatty acids (PUFA) is the most affected in leprosy (omega-3 and omega-6 fatty acids), which has a role in suppressing inflammation even though there is no statistical significance. Meanwhile, Al Mubarak *et al*. [24] shows that n-3 PUFAs, such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), as well as n-6 PUFA, that is, arachidonic acid (AA), are increased significantly in cases with high bacterial index (BI), such as with MB patients. Compared to low-BI patients, EPA increases 2.6-fold, DHA increases 1.6-fold, and AA increases 2-fold, in high-BI patients. This study also shows that PUFA holds potential as a serum metabolomic marker in LL for susceptibility to infection, diagnosis, and progression of the disease. It is discussed that promotion of anti-inflammatory lipids such as DHA, protectin D1, and EPA may enable *M. leprae* to thrive with little interference by the host's immune response. At the same time, interaction of pro-inflammatory eicosanoids and *M. marinum* and *M. tuberculosis* posits that the pathogen's survival and capacity to multiply are reinforced by the presence of these eicosanoids. This interaction is mainly found in the early stages [23], [24].

Lipids metabolized by *M. leprae* are made available through the formation of LBs which accumulate in a larger structure into lipid bodies. The study by Mattos *et al*. [30] revealed that the LB formation is higher in macrophages *M. leprae*-infected; and several genes play a role in lipid metabolism and LB formation, such as adipose differentiation-related protein (ADRP), cluster of differentiation 36 gene (CD36), toll like receptors 6 (TLR6) in higher expression than TLR2. It turns out that LB are found more abundant in infected cells with dependence on dose and time, as revealed in the study by Mattos *et al*. [31] On Schwann cells, as predilection for leprosy primary cells, it was found that LB is more abundant in highly infected cells within 24–48 h, and the biogenesis of LB is associated with Prostaglandin E2 (PGE2) and Interleukin 10 (IL-10). LB is also manifest in greater number in the more severe spectrum of leprosy. In nutritional recruitment and formation of LB, there is a role for LDL receptors, a key regulator of cholesterol biosynthesis, that is., sterol regulatory element-binding proteins, and uptake capacity of cellular cholesterol, which turns out to be higher in LL tissues [10], [30]. Another notable finding is that these changes to lipid metabolism appear to be more prominent in lepromatous patients than tuberculoid, as shown by Mattos *et al*. [10] whose study

indicates that LDL receptor expression, as well as key regulators of cholesterol biosynthesis is found to be higher in LL compared to in BT.

Lipids, especially cholesterol, are used as an energy source and a resource for mycobacterial lipid synthesis. Thus, lesions with more mycobacterium, such as in LL, would require more of the host's lipids. In addition, LB is required to be formed before mycobacterium species enters a dormant state. Aside from resource provision, it appears that cholesterol deposits also protect mycobacterium from antibacterial agents by reducing its permeability. In addition, cholesterol accumulation can be found in entry sites of _M. leprae_ to macrophages, indicating its function to ensure survival within macrophages. The accumulated cholesterol recruits coronin-1A, which in turn prevents the fusion of phagosome and lysosome [10], [30], [31].

In its pathogenesis, the overall lipid metabolism in _M. leprae_ infection causes oxidative stress to increase, especially in MB patients. A vital tangent to eradicating _M. leprae_ involving the macrophages was brought by Bhadwat _et al._ [26] whose study shows that the oxidative stress marker (MDA) is present higher in LL cases compared to TT, and the opposite is true for SOD activity. This correlates with lipid peroxidation and dysfunction of macrophages associated with low SOD and higher MDA. Overall, the two processes would promote the growth of _M. leprae_ within the host.

Thus, one may hypothesize that if plasma lipid metabolism was changed, then attempts to correct the lipid profile might be beneficial in treating leprosy patients. Lobato _et al._ [11] experimented with leprosy-infected mice treated with rifampin and with or without atorvastatin. The result shows that statin appears to reduce mycobacterial viability and overall synergizes with rifampin for its antibacterial effect.

Unique features of macrophage lipid bodies and their implications in the pathogenesis

of leprosy

As already discussed in the previous discussion of lipid bodies, when macrophages capture, ingest and destroy leprosy bacilli in phagosomes, simultaneously there is also the formation of lipid-rich organelles called LB and accumulate to form foamy macrophages [33]. LB are formed from the host lipid substrate in the form of droplets surrounding _M. leprae_ with the composition of sterol esters and triacylglycerols [34]. Surrounding it, there is a hemimembrane layer of phospholipids and proteins with various biological functions [35]. LB-specific structural proteins, namely, the PAT family (Peripilin, ADRP, and TIP47) are the peripheral structures of the LB walls. These protein participate in the regulation of intracellular lipid metabolism [36], [37] in conjunction with lipid metabolism enzymes, membrane trafficking protein Rab family (regulator of vesicular traffic and organelle interactions), reticulum endoplasm (RE) protein, and molecular chaperones [38], [39]. Capsules or lipid layers in LB contain mycoserosoic acid from phthiocerol dimycocerosates (PDIM) and phenolic glycolipids [34]. Lipid phenolic glycolipid-1 (PGL-1) was also dominant in the cell wall of _M. leprae_, which provides immunological specificity in its interaction with Schwann's laminin cells and stimulation of suppressor T cells in LL [31], [40].

Biogenesis of LB in macrophages, as illustrated in Figure 1, starts with introducing microbes by TLR2 and 6 (in Schwann cells only TLR6), which makes them associated with macrophages, triggering phagocytosis, and the formation of LBs [31], [40]. Then, the infected macrophages produce reactive oxygen species (ROS) and oxidize LDL to subsequently bind to surface-amplified receptors in the macrophage nucleus to increase uptake of oxidized fatty acids from host cells. _M. leprae_ in the phagosome will attract LBs to form LB and eventually enter a dormant and resistant period. Furthermore, infected macrophages will react positively to ADRP so that _M. leprae_ can strengthen the edges of its cell walls [40].
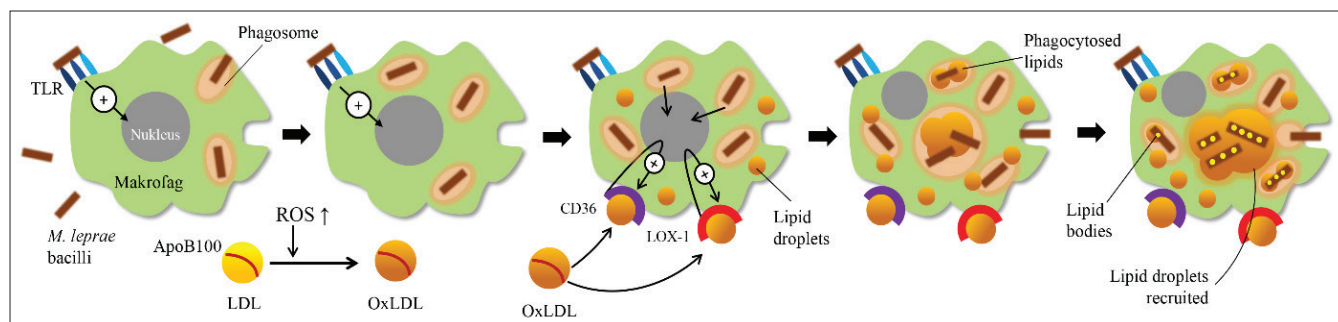
Figure 1: Proposed model for the induction of lipid body biogenesis in macrophages by M. leprae. Lipid body biogenesis begins with introducing microbes by TLR2 and TLR6 (in Schwann cells only TLR6), which triggers phagocytosis and the formation of lipid droplets. The binding of M. leprae by TLR2 and TLR6, which heterodimerize, allows leprosy to produce subsequent lipid droplets. Infected macrophages produce ROS and oxidize LDL to form OxLDL. Furthermore, OxLDL binds to the receptor type 1 scavenger CD36 and LOX1, resulting in amplification of the macrophage nucleus's surface receptor expression to increase OxLDL uptake from host cells. M. leprae in the phagosome will attract lipid droplets to form LB and eventually enter a period of dormancy and resistance [40]. ApoB100: Apolipoprotein B100, LDL: Low-density lipoprotein, OxLDL: Oxidized LDL, TLR: Toll-like receptor, LOX1: Oxidized LDL receptor-1

The lipid body role in leprosy's pathogenesis is quite significant because LB stores AA, which acts as a substrate for forming intracellular second messenger to activate host cells in a series of inflammatory reactions. Excessive inflammatory response in leprosy becomes a problem of discomfort, primarily when it manifests on the patient's skin. Eicosanoids in LB also contributes to the survival mechanism of intracellular bacteria in host cells by increasing the concentration of PGE2 in macrophages as an inhibitor of the T-helper 1 (Th1) type immune response and the production of nitric oxide (NO) free radicals, thus creating a suitable environment for the growth of *M. leprae* [40].

In addition, LB's lipid content can serve as a source of carbon and nutrients for mycobacterium, which allows its survival in cells. LB-phagosome interaction is considered as a strategy for *M. leprae* to access host lipids during infection [31]. LB organelles can also facilitate the delivery of other nutrients into the phagosome, such as iron and mycobactin (lipophilic siderophores from mycobacterium); their availability is guaranteed and promotes the growth of leprosy bacilli [31].

The presence and importance of LB in the mycobacterium group make it an attractive target for new drug recommendations in leprosy's treatment. Another promising target for inhibition is the esterase-secreting cell wall utilizing and degrading host fat cells such as Rv0183 and LipY [41]. Future studies focusing on the lipid metabolism of *M. leprae*, which is regulated by host lipid metabolism genes during infection, is crucial because *M. leprae* is highly dependent on the availability of lipids from its host cells [32].

Current leprosy treatment challenges

In 1981, the World Health Organization (WHO) introduced MDT, namely, rifampin, clofazimine, and dapsone, as first-line drugs to treat MH [4], [42]. Patients take this combination of prescriptions every month under close supervision for 6–9 months in PB and 12–18 months in MB leprosy. Minocycline, ofloxacin, and clarithromycin are second-line drugs. MDT is beneficial for preventing resistance and reducing the spread of infection. However, in a long treatment period, low patient's compliance is still a problem. This is particularly significant in regard with rifampin in the MDT regimen; although it works as a potent bactericidal agent, but heavily relies on consistent administration to be effective [4], [5], [42]. In addition, patients who received leprosy treatment for an inadequate amount of time might actually suffer from worse inflammatory reaction [43].

Another problem with MDT is its lack of ability to eliminate the whole leprosy pathogens [4], [43], [44]. A study from the WHO involving populations who have finished their MDT regiment and tested negative with AFB smear found a risk of relapse of 0.77% for MB and 1.07% for PB within 9 years. When referring to person-years calculations, the figures are 0.02–0.8% for MB and 0.65–3% for PB [44]. In 2015, out of 103 countries with relapsing leprosy, 46 countries reported an average of 3039 cases of recurrence in the country. In Indonesia in 2015, 526 of 20,160 cases of leprosy were reported as recurrences [2]. Another study proved that recurrence occurred in 39% of patients with BI ≥ 4+ after taking routine MDT for 24 months, and 67% of patients who received MDT treatment [45].

The relapse risk and resistance trend to rifampin and dapsone associated with upregulation of the *rpoB* and *folP1* genes are also increasing [46]. Second-line drugs are very good at coping with resistance but due to the high cost, their use is severely limited [46]. Based on previously presented statistical data, the authors argue that new drugs are needed to be added to the current MDT regimen. The authors found a variety of literature that describes statins' potential to help the healing process of leprosy patients.

The role of statins in addressing the challenges of leprosy treatment

Potential of statins as mycobactericidal agents

Various studies revealed that statins can serve as an antibacterial agent [7], [8]. Two meta-analyses have shown that statins can reduce the likelihood of death in patients with infection (OR 0.71; 95% CI; 0.64–0.78) and pneumonia (OR 0.66; 95% CI; 0.55–0.79) [7], [8]. Various studies have also confirmed the inhibitory effects of Gram-positive and harmful bacteria's growth, both *in vitro* and *in vivo*, but the mechanism is still unknown. It is suspected that the mechanism of death in Gram-positive bacteria by statins is due to inhibition of the bacterial mevalonic pathway in its survival. Gram-negative bacteria are independent of the mevalonic pathway but remain inhibited through an unknown mechanism [7], [8], [9], [11].

Statins more specifically inhibit the growth of *M. leprae* as it depends in intracellular lipid to survive. The study of Mattos *et al*. [10] proved the role of lipids in *M. leprae* through cell culture from LL skin lesion biopsy using AIM-V medium and RPMI-1640-FCS (Roswell Park Memorial Institute supplemented with Fetal Calf Serum). Based on this study, the viability of *M. leprae* in AIM-V liquid medium is reduced by 50% when compared to its growth in the RPMI-1640-FCS culture medium. The AIM-V medium is known to have a composition of L-glutamine, streptomycin sulfate, and gentamicin sulfate without cholesterol. In contrast, the combination RPMI-1640 and FCS medium still contains cholesterol and other compounds such as glucose, phenol pH indicator, sodium chloride, sodium bicarbonate, magnesium sulfate, disodium phosphate, potassium chloride, and calcium nitrate [10], [47]. The study showed that exogenous LDL lipids are required at least partially for *M. leprae* to survive. In the human

body, this phenomenon is supported by the behavior of _M. leprae_, which can increase the de novo synthesis of its host lipids, while macrophages infected with _M. leprae_ experience disruption of lipid homeostasis. In _in vitro_ experiments, _M. leprae_ can increase the LDL receptor excitability, which causes macrophages to have a higher LDL cholesterol intake [10].

Through the accumulation of LB in macrophages, _M. leprae_ can also avoid several components of the human immune system by inhibiting phagosome and lysosome fusion. Furthermore, pathogenic mycobacteria can use lipid membranes to manipulate macrophages by recruiting permissive macrophages and avoiding bactericidal macrophages expressing inducible NO synthase (iNOS) [10], [11]. Mycobacteria avoid bactericidal macrophages using lipid cell-surface-associated PDIM. PDIM lipids will hide the pathogen-associated molecular pattern of mycobacterium so that the TLR pathway cannot trigger bactericidal macrophages call [30], [31], [48]. Finally, _M. leprae_ avoids the macrophage immune system by its ability to escape from the phagosome through mediating the secretion of early secretory antigenic target 6 (ESAT-6) which is also owned by _M. tuberculosis_ [49].

Aside from its defense against macrophages, _M. leprae_ also inhibits the maturation pathway and activation of dendritic cells by _M. leprae_ PGL-1 expression, a unique ability that is not shared by all mycobacterium (i.e., tuberculosis and bacille Calmette-Guerin) [50], [51]. Besides, increased PGE2, which suppresses gamma interferon (IFN-γ) of _M. leprae_-infected macrophages, PGL-1 can also polarize DC to become the DC2 subset [50]. DC consists of two subsets, namely, monocyte/spleen-derived DC1 cells, which are lymphoid origin, and myeloid/plasmacytoid-derived DC2 cells, which each encourage naive Th0 to differentiate into Th1 and Th2 with cytokine support. DC1 cells express CD8a$^+$ and produce large amounts of IL-12, which creates a conducive environment for differentiation DC1 [52], [53], [54]. However, to encourage Th1 differentiation, DC1 cells can still do so without IL-12 [52], [53]. DC2 express CD11b and produce very little IL-12 [54].

The immune-suppressing effect of the PGL-1 or PGE2 molecule is dose-dependent and determines the activation of DC activation or suppression. The balance between activation and suppression of DC maturation determines whether Th1 or Th2 type immunity will respond to infected DC cells. The Th1 and Th2 immunity is also associated with the clinical manifestations of leprosy; tuberculoid leprosy will develop Th1 immunity with a bacterial load that is usually little on skin lesions. In contrast, lepromatous leprosy usually has a high number of bacilli and spreads throughout the skin, and is more dominated with Th2 [50].

Interestingly, statin appears to also aid macrophage to attack mycobacteria by improving phagosome maturation via inhibition of cholesterol (squalene) and isoprenoid (geranylgeraniol) synthesis in the host mevalonic pathway so that autophagy can take place, as was also observed in _M. tuberculosis_ [10], [11], [55].

The benefits of stopping the lipid supply for the survival of _M. leprae_ have been demonstrated through studies comparing the viability of _M. leprae_ in monocytes with various medium conditions: Presence/absence of cholesterol; presence/absence of lovastatin; and the presence/absence of a lovastatin drug vehicle [10]. The study showed that lovastatin significantly reduced the viability of _M. leprae_ ($p < 0.05$) with mean fluorescence intensity (MFI) of 58.9 ± 3 versus control MFI of 84.5 ± 2.7. The administration of a lovastatin vehicle alone does not significantly impact the viability of _M. leprae_ [10]. The mechanism of statins as mycobactericidal agents is summarized in Figure 2.

Potential statins to increase the effectiveness of rifampicin

In WHO MDT regiment for treating pauci- and multi-bacillary leprosy, rifampicin (rifampin) is always included [56]. Rifampicin works by inhibiting two of the five subunits of the RNA polymerase enzyme in the transcription process so in the end, RNA elongation stops [57]. However, the effect of rifampin is deterred by the lipid accumulation in the bacterial cell wall which reduces its permeability. Statins, as anti-lipid drugs, function to reduce body cholesterol. This will in turn diminish the supply of cholesterol in the bacterial cell wall, making _M. leprae_ more sensitive to rifampin [58], [59].

Research in animal model by Lobato _et al._ [11] shows that atorvastatin had a better adjuvant effect on rifampin than simvastatin. Nonetheless, both individually remained a good mycobactericidal agents in reducing the viability of _M. leprae_ significantly ($p < 0.01$).

Potential of statins as anti-inflammatory in leprosy

Statins are used for their anti-inflammatory effects in CVD and various infectious diseases such as _M. tuberculosis_ and _M. leprae_ [12]. The anti-inflammatory effect originates from the reduction neutrophils and macrophages' recruitment by increasing the level of peroxisome proliferator-activated receptor alpha (PPAR-α), which is an anti-inflammatory nuclear receptor. In addition, statins, especially simvastatin, can downregulate protein kinase C alpha (PKC-α) signaling pathway in neutrophils and macrophages, which stimulates PPAR-α to inhibit the inflammatory response [12]. PKC-α is known to regulate a molecular switch between transactivation and transrepression activity of PPARα. This phenomenon was investigated by Paumelle _et al._, in wild type mice that had PPAR-α compare to a modified mice which did not have PPAR-α. Simvastatin 50 mg/kg was able to reduce
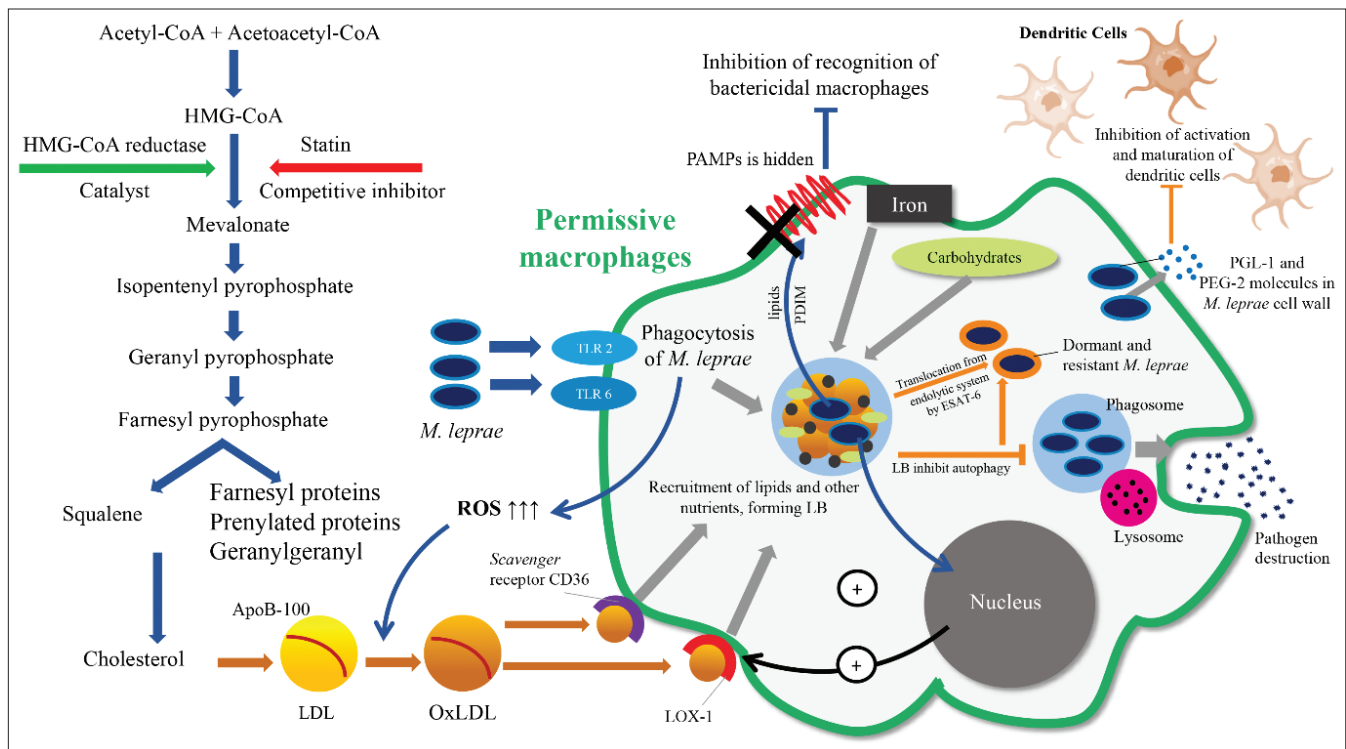
Figure 2: Proposed mechanism of statins as mycobactericidal agents through inhibition of the lipid body formation pathway. M. leprae requires lipids, iron, carbohydrates, and other nutrients from the host in the formation of lipid bodies to make them dormant and resistant; statins as competitive inhibitors of HMG-CoA reductase can stop mevalonate synthesis from the de novo lipid synthesis pathway of the host so that it will reduce lipid intake. in leprosy, restores the ability of macrophages to autophagy and causes the mycobacterium to die [10], [11], [40], [48], [49], [50], [55]. ApoB100: Apolipoprotein B100, ESAT-6: Early secretory antigenic target 6, HMG-CoA: 3-Hydroxy-3-methylglutaryl coenzyme A, LDL: Low-density lipoprotein, LOX1: Lectin-like oxidized low-density lipoprotein receptor-1, PAMP: Pathogen-associated molecular pattern, PDIM: Phthiocerol dimycocerosates, PGE-2: Prostaglandin E2, PGL-1: Phenolic glycolipid-1, ROS: Reactive oxygen species

neutrophil recruitment and swelling in the footpad of wild type mice, but not in mice without PPAR-α and the group that was given only the vehicle. The effect of simvastatin is dose-dependent; the higher the dose, the more significant the neutrophils' decrease, and the increased anti-inflammatory effectiveness [12].

M. leprae infection produces symptoms that arise as an inflammatory reaction. In TT leprosy, the pro-inflammatory Th17 cells become more dominant than the T-regulator (Treg), which is immunosuppressive. The target of recovery in chronic inflammation is to create a higher proportion of Treg among T cells and suppression of Th17. Statins were found to have the ability to do both of these things [13]. This Treg/Th17 balance regulation is closely related to the complex regulation of molecular proteins in Suppressor of Cytokine Signaling (SOCS) monocytes as well as signal transducer and activator of transcription (STAT). Both play a role in the differentiation of helper T cells from naive cells to develop into their functional subtypes (including Th1, Th2, Th17, and Treg) that depend on the cytokines they express [60]. SOCS proteins regulate the transcription of several cytokines and the expression of MHC Class II and co-stimulatory molecules [16]. The presence of SOCS1 and SOCS3 plays a role in regulating Th1/Th17 balance and Treg integrity, as shown in Figure 3.

In a more dominant Th1 immune response, STAT1 activation will induce SOCS3 protein expression to inhibit STAT3 activity, suppressing the development of Th17 and Th1 to beget relatively higher amount in equilibrium. On the other hand, SOCS3 can inhibit Th1 when overexpressed by suppressing the IL-12-mediated STAT4 activation pathway. Deletions of SOCS3 are a double-edged sword, as it also increase the production of IL-10 and transforming growth factor beta (TGF-β), thereby also inhibiting Th1. It is different when the Th17 immune response is more dominant, then SOCS1 will be highly expressed and inhibits the further development of Th1, which is mediated by IFN-γ so that Th17 will be relatively higher in the Th1/Th17 balance [60].

The differentiation of naïve CD4$^+$ T cells into Tregs will be triggered when specific cytokines and proteins are present such as TGF-β and forkhead box P3 (Foxp3$^+$). In the Tregs environment, SOCS1 levels will always be high and have a role in suppressing inflammatory conditions mediated by STAT1, STAT3, and STAT5 (high levels of cytokines IL-2, IL-6, and IFN-γ). Excessive activation of STAT5 can increase Treg counts because STAT5 directly regulates Foxp3$^+$ expression. Conversely, if the activation of STAT1 and STAT3 is excessive, then Foxp3$^+$ expression will be lost by an unknown mechanism and cause the production of IFN-γ and IL-17 to be high, respectively. Thus, SOCS1
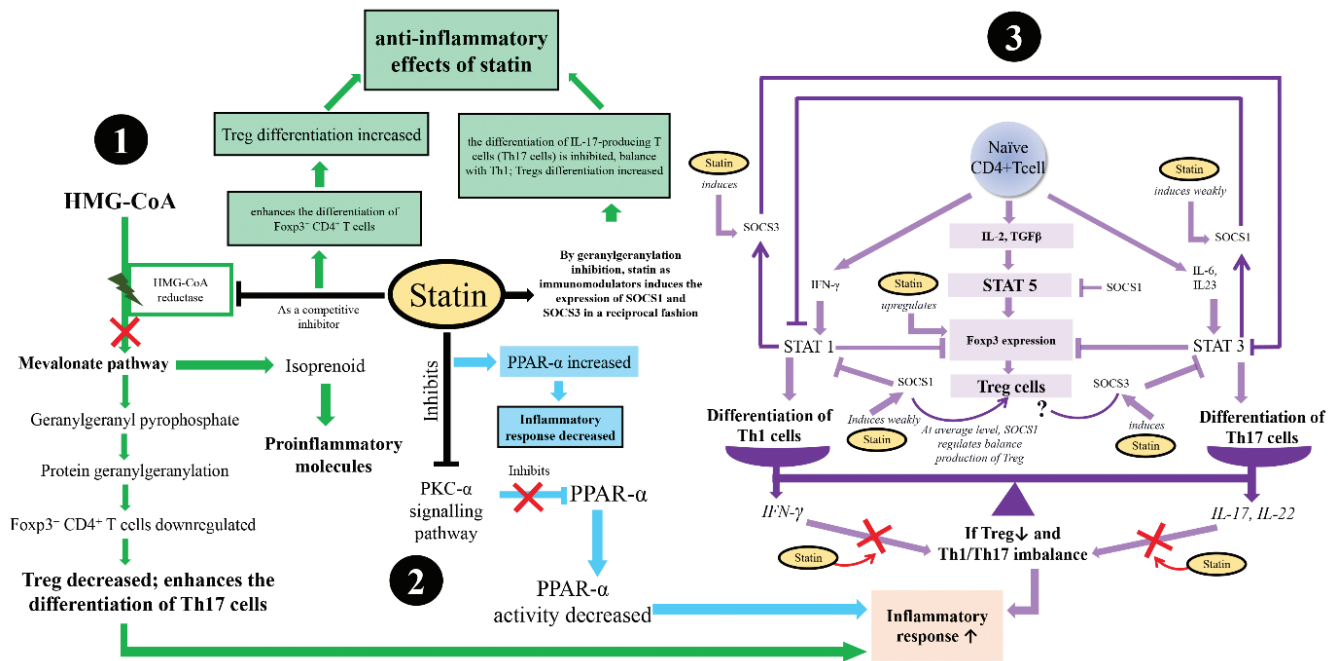
Figure 3: Proposed anti-inflammatory mechanism of statins on various signaling pathways. Statins act as competitive inhibitors of HMG-CoA reductase, which cause (1) the mevalonic lipid synthesis pathway to stop so that Foxp3+ expression upregulated, making Treg more dominant than Th17 for decreased inflammation; also isoprenoid inflammatory mediators are lowered so that the production of pro-inflammatory molecules is low. (2) Statins also inhibit the PKC-α signaling pathway so that PPAR-α expression is increased to reduce the recruitment of neutrophils and other inflammatory cells. (3) Statins are involved in the regulation of Th1/Th17/Treg balance through the mechanism of the SOCS and STAT protein complexes. Statin inhibits phosphorylation of STAT3 through upregulation of SOCS3, and inhibits phosphorylation of STAT1 through induce expression of SOCS1 so that Th1/Th17 formation can be balanced and inflammation is reduced. SCOS1 is always highly expressed to maintain the balance of STAT5, STAT3, and STAT1 to control the integrity of the Treg balance [12], [13], [14], [15], [16], [50], [60], [61]. Foxp3+: Forkhead Box P3, HMG-CoA: 3-Hydroxy-3-methyl-glutaryl-coenzyme A, IFN-γ: Gamma interferon, IL: Interleukin, PKC-α: Protein kinase C alpha, PPAR-α: Peroxisome proliferator-activated receptor-alpha, SOCS: Suppressor of cytokine signaling, STAT: Signal transducer and activator of transcription); Th: T-helper, Treg: T-regulator

in Tregs acts as a "guardian" of the balance between the number of Tregs and the Treg function [60].

Simvastatin is known to affect on SOCS and STAT through the protein geranylgeranylation process which regulates the balance between Th17 cells and Foxp3+ CD4+ T cells differentiation as a marker of Treg lineage [61]. Foxp3+ expression can be increased by simvastatin by inhibiting the protein geranylgeranylation so that the induction of Treg increases [14], [61]. This geranylgeranylation inhibition will induce upregulation of SOCS3 expression and weakens Th17 cell differentiation through reduction of pro-inflammatory interleukins produced by Th17 such as IL-17, IL-21, and IL-22 [15], [16]. SOCS3 also inhibits both STAT1 and STAT3, though the inhibition is stronger and more specific to STAT3. Statin's induction effect is also present with SOCS1 expression but weaker. SOCS1 is somewhat more specific to STAT1 inhibition [16].

As a result of the induction of SOCS3, there is an inhibition of STAT3 signaling and a decrease in the formation of IL-6 and IL-23, both play a role in the differentiation of Th17, accompanied by feedback on increasing the differentiation of Foxp3+ CD4+ T cells for induction of Treg regulation. Although it is clear that inhibition of geranylgeranylation is an integral part for SOCS3 expression upregulation, the entirety of the upregulation mechanism remains unclear

[61]. STAT3 itself, if not inhibited, can cause Foxp3+ expression to downregulate. Thus, the inhibition of protein geranylgeranylation is essential to decrease the differentiation of pro-inflammatory Th17 cells and increase Treg regulation for inflammatory balance [16].

In another way, simvastatin can inhibit the differentiation of Th17 cells, which play an essential role in the pathogenesis of other autoimmune and inflammatory disease models through inhibition of 3-hydroxy-3-methyl-glutaric-coenzyme A (HMG-CoA) reductase in the mevalonate pathway. This results in reduced isoprenoid synthesis as a mediator in the path of formation of inflammatory molecules [17], [61]. Several studies have proved this phenomenon using lovastatin, which was found to reduce the expression of different pro-inflammatory molecules, including tumor necrosis factor-alpha (TNF-α), IL-6, and iNOS [17], [61]. Atorvastatin has also been shown to suppress the differentiation of pro-inflammatory Th1 cells [61]. The various mechanisms of inhibition of inflammation by statins are summarized in Figure 3 [12], [13], [14], [15], [16], [50], [60], [61].

Anti-inflammatory effect of statin: From bench to clinic

The Ridley-Jopling classification assesses the clinical condition and immunity status of a patient by

three primary forms of leprosy; TT, LL, and borderline leprosy [62]. Regarding the inflammatory process, TT leprosy patients usually have milder symptoms, as one or more hypopigmented macules may be numb due to the death of the underlying nerves. The lepromatous type has more severe symptoms than the TT type and is easily transmitted due to the high burden of the BI. LL patients have many skin patches spread throughout the body, accompanied by numbness and muscle weakness. Apart from the skin, leprosy can also affect the nose, kidneys, and male reproductive organs. Meanwhile, the manifestation of borderline type leprosy is between the two former types [62].

The immune response to inflammation induced by *M. leprae* varies in the form of a type one or two reaction to the immune response. This difference in response is related to changes in the body's immune system. Type one reaction involves type IV hypersensitivity, especially in TT leprosy [24]. The type one reaction involves the Th-1 cell response, which produces IFN-γ and IL-2 [62]. There is also an increase in TNF-α and activation of CD4$^+$ cells and increased macrophage activity. In addition to being responded to by Th1, Th17 cells will also react to leprosy infection by secreting IL-17A, IL-17, IL-21, and IL-22 [62]. This will cause inflammation and tissue destruction of the patient. Based on a literature review, statins are thought to have an excellent anti-inflammatory effect and can potentially prevent type I reactions in tuberculoid leprosy by suppressing Th17. This hypothesis is based on simvastatin's effect in inhibiting the differentiation of naïve CD4$^+$ T cells into Th17 and reducing pro-inflammatory interleukins' secretion produced by those cells such as IL-17, IL-21, and IL-22 [15], [16].

In contrast to TT, the LL leprosy immune mechanism is a type two reaction involving a type III hypersensitivity mechanism due to deposition of immune complexes, especially in the skin. The deposits raise the level of TNF-α, and increase the number of neutrophil infiltration, as occurs in LL leprosy [13]. Some studies show lovastatin in particular to be very promising in reducing TNF-α [17].

Study of pharmacology and safety of statins on leprosy MDT administration

Rifampin is a potent inducer of CYP450 enzymes, especially CYP3A4 [63]. Administration of rifampin and statins through the same enzyme metabolic pathway can speed up statin elimination, causing reduced effectiveness. Rifampin is also an inhibitor of Organic Anion Transporter Protein B1/B3 (OATP1B1/OATP1B3), seemingly exacerbating statin concentration reduction due to co-adminsitration [63]. Giving rifampin with simvastatin can reduce 94% of the area under curve (AUC) for simvastatin with a similar effect on lovastatin and 80% on atorvastatin due to the shared metabolic pathway by CYP3A4. Although with a lesser impact, AUC reduction also occurs in fluvastatin by 50% and Pravastatin by 30% [63], [64]. However, rifampicin does not affect rosuvastatin pharmacokinetics, and even increases AUC in pitavastatin, possibly due to OATP1B1 rifampicin inhibition [63].

Although the presence of rifampin slightly decreases statins' cholesterol-lowering efficacy, the statin dose-response curve is flattened, so that two-thirds of the maximum response can be achieved with only a quarter of the total dose. Therefore, the clinical significance of the rifampin drug interaction as an inducer of the metabolic enzyme CYP3A4 does not severely impact the overall efficacy [64]. In addition, rifampin is administered as MDT with dapsone or clofazimine, whose interaction patterns decrease the metabolism of statins, in contrast to rifampin [65], [66]. Should statin viability become a concern, it is still possible to adjust statin dose when giving MDT to reach optimal effect due to some drug interaction between rifampin and statin in the level of the liver metabolic pathway. However, future research is needed to establish an optimal dose of statin when co-administration as adjuvant therapy for MDT of leprosy [63], [64].

At present, available data regarding the effect of statin administration are minimal in leprosy patients, most only exist in tuberculosis. Tuberculosis infection has several immunopathogenesis similarities with leprae, as they are both caused by mycobacterium species, and current studies on tuberculosis cases present statin as a promising therapeutic agent in two publications by Tahir *et al*. [67] and Alffenaar *et al.* [68]. *In vivo* studies in mice also showed statin adjuvant in MDT-TB shorten the treatment time and reduce recurrence rates [55], [69].

Conclusion

Lipid metabolism holds a key role in the immunopathogenesis of leprosy, so much so that the patient's lipid profile is significantly altered. As such, denying *M. leprae* of its access to lipid could potentially be a new therapeutic approach to support the current MDT regiment. Statins have various properties that can help cure leprosy more effectively through several mechanisms regarding lipid metabolism involvement in *M. leprae* infection. The inhibitory effect of statin on mevalonate lipid synthesis can reduce the supply of lipids for the survival of *M. leprae*, improve the phagocytosis performance of macrophages, and increase bacterial cell walls permeability to rifampin. Statins also have an excellent anti-inflammatory effect that may prevent type I reaction in tuberculoid leprosy with Th17 suppression and prevent the type II reaction in lepromatous leprosy by decreasing TNF-α. These properties synergize with the current MDR regiment.

However, to date, research linking the effects of statins to leprosy in humans is still limited, thus further research is needed.

References

1. Wibawa T, Satoto TB. Magnitude of neglected tropical diseases in Indonesia at postmillennium development goals era. J Trop Med. 2016;2016:5716785. https://doi.org/10.1155/2016/5716785

2. World Health Organization. Global leprosy update, 2016: Accelerating reduction of disease burden. Relev Epidemiol Hebd. 2017;92(35):501-19.

3. Kemenkes RI. Hapuskan Stigma dan Diskriminasi terhadap Kusta (In Indonesian). InfoDatin Pusat Data dan Informasi Kementrian Kesehatan RI. Indonesia: Kementerian Kesehatan Republik Indonesia; 2018. p. 1-11. Available from: https://www.pusdatin.kemkes.go.id/resources/download/pusdatin/infodatin/infoDatin-kusta-2018.pdf. https://doi.org/10.32922/jkp.v8i2.186. [Last accessed on 2021 Mar 19].

4. Naaz F, Mohanty PS, Bansal AK, Kumar D, Gupta UD. Challenges beyond elimination in leprosy. Int J Mycobacteriol. 2017;6(3):222-8. https://doi.org/10.4103/ijmy.ijmy_70_17
   PMid:28776519

5. Cambau E, Saunderson P, Matsuoka M, Cole ST, Kai M, Suffys P, et al. Antimicrobial resistance in leprosy: Results of the first prospective open survey conducted by a WHO surveillance network for the period 2009-15. Clin Microbiol Infect. 2018;24(12):1305-10. https://doi.org/10.1016/j.cmi.2019.01.004
   PMid:29496597

6. Trindade MA, Benard G, Ura S, Ghidella CC, Avelleira JC, Vianna FR, et al. Granulomatous reactivation during the course of a leprosy infection: Reaction or relapse. PLoS Negl Trop Dis. 2010;4(12):1-5. https://doi.org/10.1371/journal.pntd.0000921
   PMid:21200422

7. Ma Y, Wen X, Peng J, Lu Y, Guo Z, Lu J. Systematic review and meta-analysis on the association between outpatient statins use and infectious disease-related mortality. PLoS One. 2012;7(12):e51548. https://doi.org/10.1371/journal.pone.0051548
   PMid:23284711

8. Chopra V, Rogers MA, Buist M, Govindan S, Lindenauer PK, Saint S, et al. Is statin use associated with reduced mortality after pneumonia? A systematic review and meta-analysis. Am J Med. 2012;125(11):1111-23. https://doi.org/10.1016/j.amjmed.2012.04.011
   PMid:22835463

9. Haeri MR, White K, Qharebeglou M, Ansar MM. Cholesterol suppresses antimicrobial effect of statins. Iran J Basic Med Sci. 2015;18(12):1253-6.
   PMid:26877857

10. Mattos KA, Oliveira VC, Berrêdo-Pinho M, Amaral JJ, Antunes LC, Melo RC, et al. Mycobacterium leprae intracellular survival relies on cholesterol accumulation in infected macrophages: A potential target for new drugs for leprosy treatment. Cell Microbiol. 2014;16(6):797-815. https://doi.org/10.1111/cmi.12279
    PMid:24552180

11. Lobato LS, Rosa PS, Da Silva Ferreira J, Da Silva Neumann A, Da Silva MG, Nascimento DC, et al. Statins increase rifampin mycobactericidal effect. Antimicrob Agents Chemother. 2014;58(10):5766-74. https://doi.org/10.1128/aac.01826-13

PMid:25049257

12. Paumelle R, Blanquart C, Briand O, Barbier O, Duhem C, Woerly G, et al. Acute antiinflammatory properties of statins involve peroxisome proliferator-activated receptor-alpha via inhibition of the protein kinase C signaling pathway. Circ Res. 2006;98(3):361-9. https://doi.org/10.1161/01.res.0000202706.70992.95
    PMid:16397146

13. Chapel H, Haeney M, Misbah S, Snowden N. Essentials of Clinical Immunology. 6th ed. Chicester: Wiley-Blackwell; 2014. p. 350. Available from http://www.med-mu.com/wp-content/uploads/2018/06/Essentials-of-Clinical-Immunology-6E-Chapel-Haeney-Misbah-_-Snowden.pdf. https://doi.org/10.1046/j.1365-2249.1997.4291322.x. [Last accessed on 2021 Mar 19].

14. Kim YC, Kim KK, Shevach EM. Simvastatin induces Foxp3+ T regulatory cells by modulation of transforming growth factor-beta signal transduction. Immunology. 2010;130(4):484-93. https://doi.org/10.1111/j.1365-2567.2010.03269.x
    PMid:20408897

15. Zhang X, Tao Y, Troiani L, Markovic-Plese S. Simvastatin inhibits IFN regulatory factor 4 expression and Th17 cell differentiation in CD4+ T cells derived from patients with multiple sclerosis. J Immunol. 2011;187(6):3431-7. https://doi.org/10.4049/jimmunol.1100580
    PMid:21856936

16. Zhang X, Jin J, Peng X, Ramgolam VS, Markovic-Plese S. Simvastatin inhibits IL-17 secretion by targeting multiple IL-17-regulatory cytokines and by inhibiting the expression of IL-17 transcription factor RORC in CD4+ lymphocytes. J Immunol. 2008;180(10):6988-96. https://doi.org/10.4049/jimmunol.180.10.6988
    PMid:18453621

17. Pahan K, Sheikh FG, Namboodiri AM, Singh I. Lovastatin and phenylacetate inhibit the induction of nitric oxide synthase and cytokines in rat primary astrocytes, microglia, and macrophages. J Clin Invest. 1997;100(11):2671-9. https://doi.org/10.1172/jci119812
    PMid:9389730

18. The Joanna Briggs Institute. The Joanna Briggs Institute Critical Appraisal Tools: Checklist for Analytical Cross Sectional Studies. South Australia: Joanna Briggs Institute, University of Adelaide; 2017. p. 1-7. Available from: https://www.joannabriggs.org/sites/default/files/2019-05/JBI_Critical_Appraisal-Checklist_for_Analytical_Cross_Sectional_Studies2017_0.pdf. https://doi.org/10.11124/01938924-201109481-00003 [Last accessed on 2021 Mar 19].

19. Oxford CEEBM. Oxford Centre for Evidence-Based Medicine: Levels of Evidence. Center for Evidence-based Medicine; 2009. Available from: https://www.cebm.ox.ac.uk/resources/levels-of-evidence/oxford-centre-for-evidence-based-medicine-levels-of-evidence-march-2009. [Last accessed on 2021 Mar 19]. https://doi.org/10.1136/ebm.14.3.69-a

20. Lemes RM, Silva CA, Marques MÂ, Atella GC, Nery JA, Nogueira MR, et al. Altered composition and functional profile of high-density lipoprotein in leprosy patients. PLoS Negl Trop Dis. 2020;14(3):1-24. https://doi.org/10.1371/journal.pntd.0008138
    PMid:32226013

21. da Silva DS, Teixeira LA, Beghini DG, da Silva Ferreira AT, de Berredo Moreira Pinho M, et al. Blood coagulation abnormalities in multibacillary leprosy patients. PLoS Negl Trop Dis. 2018;12(3):1-21.
    PMid:29565968

22. Sarwar G, Sultana V, Gul A, Ara J. Comparative study of lipid profile in multibacillary and paucibacillary leprosy patients. J Bahria Univ Med Dent Coll. 2016;6(1):43-6.

23. Amaral JJ, Antunes LC, de Macedo CS, Mattos KA, Han J, Pan J, *et al*. Metabonomics reveals drastic changes in anti-inflammatory/pro-resolving polyunsaturated fatty acids-derived lipid mediators in leprosy disease. PLoS Negl Trop Dis. 2013;7(8):e2381. https://doi.org/10.1371/journal.pntd.0002381
    PMid:23967366

24. Al-Mubarak R, Vander Heiden J, Broeckling CD, Balagon M, Brennan PJ, Vissa VD. Serum metabolomics reveals higher levels of polyunsaturated fatty acids in lepromatous leprosy: Potential markers for susceptibility and pathogenesis. PLoS Negl Trop Dis. 2011;5(9):e1303. https://doi.org/10.1371/journal.pntd.0001303
    PMid:21909445

25. Nwosu C, Nwosu S. Abnormalities in serum lipids and liver fuction in Nigeria patients with leprosy. JOMIP. 2001;2:5-10.

26. Bhadwat VR, Borade VB. Increased lipid peroxidation in lepromatous leprosy. Indian J Dermatol Venereol Leprol. 2000;66(3):121-5.
    PMid:20877051

27. Silva RV, de Araújo RS, Aarão TL, da Silva Costa PD, Sousa JR, Quaresma JA. Correlation between therapy and lipid profile of leprosy patients: Is there a higher risk for developing cardiovascular diseases after treatment? Infect Dis Poverty. 2017;6(1):1-7. https://doi.org/10.1186/s40249-017-0295-1
    PMid:28457229

28. Gupta A, Koranne R, Kaul N. Study of serum lipids in leprosy. Indian J Dermatol Venereol Leprol. 2002;68(5):262-6.
    PMid:17656962

29. Miyamoto Y, Mukai T, Matsuoka M, Kai M, Maeda Y, Makino M. Profiling of intracellular metabolites: An approach to understanding the characteristic physiology of *Mycobacterium leprae*. PLoS Negl Trop Dis. 2016;10(8):1-12. https://doi.org/10.1371/journal.pntd.0004881
    PMid:27479467

30. Mattos KA, D'Avila H, Rodrigues LS, Oliveira VG, Sarno EN, Atella GC, *et al*. Lipid droplet formation in leprosy: Toll-like receptor-regulated organelles involved in eicosanoid formation and *Mycobacterium leprae* pathogenesis. J Leukoc Biol. 2010;87(3):371-84. https://doi.org/10.1189/jlb.0609433
    PMid:19952355

31. Mattos KA, Oliveira VG, D'Avila H, Rodrigues LS, Pinheiro RO, Sarno EN, *et al*. TLR6-driven lipid droplets in *Mycobacterium leprae*-infected schwann cells: Immunoinflammatory platforms associated with bacterial persistence. J Immunol. 2011;187(5):2548-58. https://doi.org/10.4049/jimmunol.1101344
    PMid:21813774

32. Cruz D, Watson AD, Miller CS, Montoya D, Ochoa MT, Sieling PA, *et al*. Host-derived oxidized phospholipids and HDL regulate innate immunity in human leprosy. J Clin Invest. 2008;118(8):2917-28. https://doi.org/10.1172/jci34189
    PMid:18636118

33. Alter A, Grant A, Abel L, Alcaïs A, Schurr E. Leprosy as a genetic disease. Mamm Genome. 2011;22(1-2):19-31. https://doi.org/10.1007/s00335-010-9287-1
    PMid:20936290

34. Elamin AA, Stehr M, Singh M. Lipid droplets and *Mycobacterium leprae* infection. J Pathog. 2012;2012:361374.
    PMid:23209912

35. Melo RC, Dvorak AM. Lipid body-phagosome interaction in macrophages during infectious diseases: Host defense or pathogen survival strategy? PLoS Pathog. 2012;8(7):e1002729. https://doi.org/10.1371/journal.ppat.1002729
    PMid:22792061

36. Bickel PE, Tansey JT, Welte MA. PAT proteins, an ancient family

37. of lipid droplet proteins that regulate cellular lipid stores. Biochim Biophys Acta. 2009;1791(6):419-40. https://doi.org/10.1016/j.bbalip.2009.04.002
    PMid:19375517

37. Degang Y, Akama T, Hara T, Tanigawa K, Ishido Y, Gidoh M, *et al*. Clofazimine modulates the expression of lipid metabolism proteins in *Mycobacterium leprae*-infected macrophages. PLoS Negl Trop Dis. 2012;6(12):e1936. https://doi.org/10.1371/journal.pntd.0001936
    PMid:23236531

38. Wan HC, Melo RC, Jin Z, Dvorak AM, Weller PF. Roles and origins of leukocyte lipid bodies: Proteomic and ultrastructural studies. FASEB J. 2007;21(1):167-78. https://doi.org/10.1096/fj.06-6711com
    PMid:17135363

39. Huang S, Jiang L, Zhuang X. Possible roles of membrane trafficking components for lipid droplet dynamics in higher plants and green algae. Front Plant Sci. 2019;10:207. https://doi.org/10.3389/fpls.2019.00207
    PMid:30858860

40. Stehr M, Singh M. Lipid inclusions in mycobacterial infections. In: Tuberculosis-Current Issues in Diagnosis and Management. India: InTech; 2013. p. 31-46.

41. Daleke MH, Cascioferro A, de Punder K, Ummels R, Abdallah AM, van der Wel N, *et al*. Conserved Pro-Glu (PE) and Pro-Pro-Glu (PPE) protein domains target LipY lipases of pathogenic mycobacteria to the cell surface via the ESX-5 pathway. J Biol Chem. 2011;286(21):19024-34. https://doi.org/10.1074/jbc.m110.204966
    PMid:21471225

42. World Health Organization. Effectiveness of MDT: FAQ. Leprosy Elimination. Geneva: World Health Organization; 2020. Available from: https://www.who.int/lep/mdt/effectiveness/en. [Last accessed on 2020 Dec 30]. https://doi.org/10.4269/ajtmh.2009.81.330

43. Fajardo TT, Villahermosa L, Pardillo FE, Abalos RM, Burgos J, Dela Cruz E, *et al*. A comparative clinical trial in multibacillary leprosy with long-term relapse rates of four different multidrug regimens. Am J Trop Med Hyg. 2009;81(2):330-4.
    PMid:19635893

44. The Leprosy Unit. Risk of relapse in leprosy. The leprosy unit, WHO. Indian J Lepr. 1995;67(1):13-26.
    PMid:7622926

45. Malathi M, Thappa DM. Fixed-duration therapy in leprosy: Limitations and opportunities. Indian J Dermatol. 2013;58(2):93-100. https://doi.org/10.4103/0019-5154.108029
    PMid:23716796

46. Nakata N, Kai M, Makino M. Mutation analysis of the *Mycobacterium leprae* folP1 gene and dapsone resistance. Antimicrob Agents Chemother. 2011;55(2):762-6. https://doi.org/10.1128/aac.01212-10
    PMid:21115799

47. Rosdiana A, Hadisaputri YE. Review artikel: Studi pustaka tentang prosedur kultur sel (in Indonesian). Farmaka. 2016;14(1):236-49.

48. Cambier CJ, Takaki KK, Larson RP, Hernandez RE, Tobin DM, Urdahl KB, *et al*. Mycobacteria manipulate macrophage recruitment through coordinated use of membrane lipids. Nature. 2014;505(7482):218-22. https://doi.org/10.1038/nature12799
    PMid:24336213

49. Geluk A, van Meijgaarden KE, Franken KL, Subronto YW, Wieles B, Arend SM, *et al*. Identification and characterization of the ESAT-6 homologue of *Mycobacterium leprae* and T-cell cross-reactivity with *Mycobacterium tuberculosis*. Infect Immun. 2002;70(5):2544-8. https://doi.org/10.1128/

iai.70.5.2544-2548.2002

PMid:11953394

50. Murray RA, Siddiqui MR, Mendillo M, Krahenbuhl J, Kaplan G. *Mycobacterium leprae* inhibits dendritic cell activation and maturation. J Immunol. 2007;178(1):338-44. https://doi.org/10.4049/jimmunol.178.1.338

    PMid:17182571

51. Spencer JS, Brennan PJ. The role of *Mycobacterium leprae* phenolic glycolipid I (PGL-I) in serodiagnosis and in the pathogenesis of leprosy. Lepr Rev. 2011;82(4):344-57. https://doi.org/10.47276/lr.82.4.344

    PMid:22439275

52. Viney JL. Dendritic cell subsets: The ultimate T cell differentiation decision makers? Gut. 1999;45(5):640-1. https://doi.org/10.1136/gut.45.5.640

    PMid:10517895

53. Rissoan MC, Soumelis V, Kadowaki N, Grouard G, Briere F, de Waal Malefyt R, *et al*. Reciprocal control of T helper cell and dendritic cell differentiation. Science. 1999;283(5405):1183-6. https://doi.org/10.1126/science.283.5405.1183

    PMid:10024247

54. Lambrecht BN, Prins JB, Hoogsteden HC. Lung dendritic cells and host immunity to infection. Eur Respir J. 2001;18(4):692-704.

    PMid:11716176

55. Parihar SP, Guler R, Khutlang R, Lang DM, Hurdayal R, Mhlanga MM, *et al*. Statin therapy reduces the *Mycobacterium tuberculosis* burden in human macrophages and in mice by enhancing autophagy and phagosome maturation. J Infect Dis. 2014;209(5):754-63. https://doi.org/10.1093/infdis/jit550

    PMid:24133190

56. World Health Organization. WHO Model Prescribing Information: Drugs used in Bacterial Infections: Sexually Transmitted Diseases: Gonorrhoeaea. Geneva: World Health Organization Regional Office for the Eastern Mediterranean; 1995. Available from: https://www.apps.who.int/iris/handle/10665/37143. [Last accessed on 2020 Jul 05].

57. Unissa AN, Hanna LE. Molecular mechanisms of action, resistance, detection to the first-line anti tuberculosis drugs: Rifampicin and pyrazinamide in the post whole genome sequencing era. Tuberculosis (Edinb). 2017;105:96-107. https://doi.org/10.1016/j.tube.2017.04.008

    PMid:28610794

58. Brzostek A, Pawelczyk J, Rumijowska-Galewicz A, Dziadek B, Dziadek J. *Mycobacterium tuberculosis* is able to accumulate and utilize cholesterol. J Bacteriol. 2009;191(21):6584-91. https://doi.org/10.1128/jb.00488-09

    PMid:19717592

59. Mira E, León B, Barber DF, Jiménez-Baranda S, Goya I, Almonacid L, *et al*. Statins induce regulatory T cell recruitment via a CCL1 dependent pathway. J Immunol. 2008;181(5):3524-34. https://doi.org/10.4049/jimmunol.181.5.3524

    PMid:18714025

60. Yoshimura A, Suzuki M, Sakaguchi R, Hanada T, Yasukawa H. SOCS, inflammation, and autoimmunity. Front Immunol. 2012;3:1-9. https://doi.org/10.3389/fimmu.2012.00020

    PMid:22566904

61. Kagami SI, Owada T, Kanari H, Saito Y, Suto A, Ikeda K, *et al*. Protein geranylgeranylation regulates the balance between Th 17 cells and Foxp3+ regulatory T cells. Int Immunol. 2009;21(6):679-89. https://doi.org/10.1093/intimm/dxp037

    PMid:19380384

62. de Lima Fonseca AB, do Vale Simon M, Cazzaniga RA, de Moura TR, de Almeida RP, Duthie MS, *et al*. The influence of innate and adaptative immune responses on the differential clinical outcomes of leprosy. Infect Dis Poverty. 2017;6(1):1-8. https://doi.org/10.1186/s40249-016-0229-3

    PMid:28162092

63. Hylton Gravatt LA, Flurie RW, Lajthia E, Dixon DL. Clinical guidance for managing statin and antimicrobial drug-drug interactions. Curr Atheroscler Rep. 2017;19(11):46. https://doi.org/10.1007/s11883-017-0682-x

    PMid:28990114

64. Neuvonen PJ, Niemi M, Backman JT. Drug interactions with lipid-lowering drugs: Mechanisms and clinical relevance. Clin Pharmacol Ther. 2006;80(6):565-81. https://doi.org/10.1016/j.clpt.2006.09.003

    PMid:17178259

65. Dapsone. Drug Bank; 2020. Available from: https://www.drugbank.ca/drugs/DB00250. [Last accessed on 2020 Dec 30].

66. Clofazimine. Drug Bank; 2020. Available from: https://www.drugbank.ca/drugs/DB00845. [Last accessed on 2020 Dec 30].

67. Tahir F, Bin Arif T, Ahmed J, Shah SR, Khalid M. Anti-tuberculous effects of statin therapy: A review of literature. Cureus. 2020;12(3):e7404. https://doi.org/10.7759/cureus.7404

    PMid:32337130

68. Alffenaar JW, Akkerman OW, Van Hest R. Statin adjunctive therapy for tuberculosis treatment. Antimicrob Agents Chemother. 2016;60(11):7004. https://doi.org/10.1128/aac.01836-16

    PMid:28045667

69. Dutta NK, Bruiners N, Pinn ML, Zimmerman MD, Prideaux B, Dartois V, *et al*. Statin adjunctive therapy shortens the duration of TB treatment in mice. J Antimicrob Chemother. 2016;71(6):1570-7. https://doi.org/10.1093/jac/dkw014

    PMid:26903278