Open Forum Infectious Diseases

REVIEW ARTICLE

Clofazimine in Nontuberculous Mycobacterial Infections: A Growing Niche

Sarah A. McGuffin,[1] Paul S. Pottinger,[1] and James P. Harnisch[2]

Divisions of [1]Allergy & Infectious Diseases and [2]Dermatology, University of Washington, Seattle

Infection secondary to rapidly growing mycobacteria (RGM) is associated with significant morbidity and mortality, especially in individuals with underlying structural lung disease or immune compromise. Such infections, particularly those caused by the *Mycobacterium abscessus* group, are challenging to treat due to high virulence, antibiotic resistance, and the lack of effective and tolerable therapies. Although novel antimycobacterials are under development, clofazimine—a drug historically administered as part of multidrug therapy regimens for *Mycobacterium leprae*—holds promise as a chemotherapeutic for the treatment of RGM. The history, pharmacologic properties of clofazimine, as well as in vitro and in vivo studies against RGM are described here and highlight a potential new niche for an old drug.

Keywords. clofazimine; *Mycobacterium abscessus*; nontuberculous mycobacteria; rapidly growing mycobacteria.

Over the past decades, nontuberculous mycobacteria (NTM), particularly rapidly growing mycobacteria such as the *Mycobacterium abscessus* group, have emerged as important human pathogens. These organisms cause infections that are often difficult to treat, have a high rate of recurrence, demonstrate a high incidence of multidrug resistance, require prolonged treatment regimens, and can be associated high fatality rates in certain populations [1–9]. Nontuberculous mycobacteria-associated pulmonary infections are the most common and severe clinical manifestation, particularly in patients with structural lung disease [4, 5, 10]. Until novel chemotherapies with improved efficacy and tolerability become available, there is a growing niche for the antileprosy drug clofazimine in the treatment of pulmonary NTM.

NONTUBERCULOUS MYCOBACTERIA

Although *Mycobacterium tuberculosis* is the most widely recognized species of the genus mycobacteria, there is increasing clinical interest in NTM. This is likely due to aging populations in many countries, the recognition that NTM lung disease is encountered with increasing frequency in the non-human immunodeficiency virus (HIV) population, especially those with bronchiectasis or impaired host immunity, as well as the congruent growth in the worldwide HIV epidemic [4, 9–11]. The expanding use of immunosuppressive drugs and improved methodology for NTM isolation and identification from clinical specimens heightens the importance of these infections in medical practice [1, 4].

Taxonomy and Microbiology

In 1959, Runyon [12] proposed the first classification for NTM species, categorizing human isolates of NTM into 4 groups based on growth rate, colony morphology, and pigmentation: (1) *M tuberculosis* complex, (2) *Mycobacterium leprae*, (3) rapidly growing NTM, and (4) slowly growing NTM. Enhanced species identification with nucleic acid probes, species-specific polymerase chain reaction, ribosomal ribonucleic acid sequencing, high-performance liquid chromatography, and mass spectrometry has decreased the reliance on the Runyon [12] classification strategy as the sole means of species identification, although this phenotypic testing strategy can provide a preliminary clue as to the identity of an NTM in the early stages of diagnostic evaluation [3, 4, 7, 13, 14]. The taxonomic classification of certain species has recently become more complex with the subspeciation of *M abscessus* (now called the *M abscessus* group) into *M abscessus* subsp *abscessus*, *M abscessus* subsp *massiliense*, and *M abscessus* subsp *bollettii* based on antibiotic resistance phenotypes and *rpoB* and *erm*(41) genotypes [3, 7, 13–15]. Although clinically important, unfortunately, the required molecular methods for such differentiation are not routinely available in most mycobacteriology laboratories, identification can be challenging even in laboratories with the most advanced diagnostic tools, and no codified or authoritative guidelines exist for "official" taxonomic designations [7, 14]. Currently, more than 150 NTM species have been identified [13].

Epidemiology and Disease

Most NTM species are ubiquitous in the environment worldwide and can cause colonization, pseudo-outbreaks in

Received 4 April 2017; editorial decision 13 July 2017; accepted 19 July 2017.

Correspondence: S. A. McGuffin, MD, 1959 NE Pacific Street Box 356421, Seattle, WA 98195-6421 (mcguffin@uw.edu).

Open Forum Infectious Diseases®

DOI: 10.1093/ofid/ofx147

healthcare settings, and infection [4, 5, 10]. Reports of NTM infection have increased worldwide over the last 2 decades, an increase which exceeds that expected due to improved detection and diagnosis alone [5, 16]. In the United States, the most frequently reported clinically significant species are *Mycobacterium avium* complex (MAC), *M abscessus* group, *Mycobacterium fortuitum*, and *Mycobacterium kansasii* [4, 10, 16]. The pathologic potential of these mycobacteria is influenced by the immunologic status of the patient, the site of infection, and the presence of underlying structural/anatomic disease [4, 9, 10]. Recent reports suggest that in many areas of the United States, the prevalence of NTM pulmonary disease exceeds that caused by *M tuberculosis* [1, 4, 5, 9, 11]. Pulmonary disease is the most common clinical manifestation and most frequently occurs in older adults and individuals with underlying structural lung disease (chronic obstructive pulmonary disease, bronchiectasis, cystic fibrosis, pneumoconiosis) [1, 4, 6, 10, 11]. Symptoms, physical examination findings, and imaging are nonspecific, and thus the American Thoracic Society/Infectious Diseases Society of America diagnostic criteria are key when evaluating a patient with suspected NTM lung disease [4]. Other clinical manifestations include disseminated disease, ophthalmic infections, localized skin and soft-tissue infections, central nervous system infections, otitis media, lymphadenitis, and abdominal infections [2, 4, 9, 16].

The Need for New Antimicrobials

Although MAC is the mycobacterial pathogen most commonly isolated from respiratory samples, infection secondary to rapidly growing NTM, especially the *M abscessus* group, is particularly challenging due to high virulence, antibiotic resistance, and the lack of effective and tolerable therapies [1, 3–5, 7]. Not every positive culture for NTM requires treatment, but there have been numerous reports of clinical deterioration and death associated with persistent recovery of NTM, particularly *M abscessus* group from lower respiratory specimens [4]. The overall paucity of successful treatment options is particularly daunting because *M abscessus* group represents up to 18% of NTM respiratory isolates in patients with cystic fibrosis [4].

There is little published information regarding treatment options for pulmonary infections secondary to *M abscessus* group, and no antibiotic regimens, even based on in vitro susceptibilities, have been shown to reliably produce clinical response or long-term sputum conversion [3–5, 15–17]. *Mycobacterium abscessus* group is uniformly resistant to the standard antituberculous agents [3, 4]. Inducible macrolide resistance undermines the cornerstones of treatment of other NTM pulmonary infections such as MAC and *M kansasii* [3–5, 7]. Most mycobacteria are intrinsically resistant to glycopeptides such as vancomycin [16]. Intrinsic resistance to β-lactams, acquired resistance to tetracyclines, and intrinsic and acquired resistance to fluoroquinolones have also been observed [7, 16]. Although antibiotics such as linezolid and tigecycline have moderate in vitro activity against *M abscessus* group, in vivo studies have yielded disappointing results (Table 1), and long-term administration is frequently associated with intolerable side effects [3, 4]. The only predictably curative therapy for "focal" pulmonary disease due to *M abscessus* group consists of surgical resection of the involved lung combined with multidrug chemotherapy therapy [4, 16]. Currently, recommended multidrug regimens consist of amikacin plus cefoxitin or imipenem and an oral macrolide (if sensitive) for at least 2–4 months [4, 5, 7]. However, acquired and intrinsic aminoglycoside and macrolide resistance exists, and the significant side effects and toxicities common with aggressive and prolonged parenteral therapy limit long-term use and tolerability [3, 4, 7, 16, 17]. Currently, the Clinical and Laboratory Standards Institute recommends in vitro testing with amikacin, cefoxitin, ciprofloxacin, clarithromycin, doxycycline, sulfamethoxazole, and imipenem against all RGM [4, 16, 18].

Cure rates of pulmonary *M abscessus* group disease range from 30% to 50% [17]. Given the current lack of viable antibiotic options, many experts view pulmonary infection with *M abscessus* group as (1) a chronic and incurable infection for most patients and (2) an area in need of new antimicrobial agents [4, 5]. Although novel antimicrobials are under investigation, an old drug may fill this emerging need in the treatment of NTM pulmonary disease.

CLOFAZIMINE

Discovery

In 1957, Barry and colleagues [19, 20] described a new series of pigments derived from lichens that demonstrated high antituberculous activity. The agents were called riminophenazines for their phenazine core and imine moiety, and the most potent of these agents was B-663 [19, 20]. The pharmaceutical company Novartis subsequently further developed B-663 and sold it under the International Nonproprietary Name clofazimine (brand name Lamprene). Despite high activity against *M tuberculosis* in vitro and in mouse infections, good oral absorption in a micronized form, and deposition and persistence in adipose and cells of the reticuloendothelial system, the compound was found to be ineffective against tuberculosis in humans [19, 21]. When Browne and Hogerzeil [22] theorized that a compound such as clofazimine that accumulates inside macrophages would be effective against "intracellular diseases" such as leprosy, its current role as a staple in the treatment of leprosy was realized. In 1982, a World Health Organization (WHO) Study Group recommended treating multibacillary leprosy with a multidrug regimen consisting of dapsone, rifampicin, and clofazimine [23].

Mechanism of Action

Clofazimine possesses both antimicrobial and anti-inflammatory properties [21]. It is active against both slowly

Table 1. In Vitro Studies of Clofazimine Activity Versus Nontuberculous Mycobacteria

Study	Mycobacteria	Drug	MIC Range (μg/mL)	MIC_{50} (μg/mL Clofazimine)	Notes
Ausina et al [47]	*Mycobacterium fortuitum* (N = 28)	Clofazimine	≤0.25–1	0.5	
	Mycobacterium chelonae subsp *chelonae* (N = 13)	Clofazimine	≤0.25–8	1	
	Mycobacterium fallax (N = 10)	Clofazimine	≤0.25–5	0.5	
Shen et al [2][a]	*Mycobacterium abscessus* group (N = 117 or 40)	Clofazimine	<0.03125–2 (N = 117) 0.25–0.5 (N = 40)	0.25 (N = 117)	99.1% of isolates with MIC <1μg/mL
		Amikacin	4–64 (N = 40)		
		Clofazimine + amikacin	<0.03125 (N = 40)		Synergism seen in 100% of isolates
	M fortuitum (N = 48)	Clofazimine	0.125–8	0.25	91.7% with MIC <1 μg/mL
		Amikacin	0.5–8		
		Clofazimine + amikacin	<0.03125–4		Synergism seen in 6.25% of isolates (no interaction in 91.75% of isolates)
	M chelonae (N = 20)	Clofazimine	0.25–0.5	0.5	100% with MIC <1 μg/mL
		Amikacin	4–16		
		Clofazimine + amikacin	<0.03125		Synergism seen in 100% of isolates
van Ingen et al [48][a]	*M abscessus* subsp *abscessus* (N = 342)	Clofazimine		≤0.5	
		Amikacin		16	
		Clofazimine + amikacin (N = 68)		≤0.5	Synergism seen in 82% of isolates
	M abscessus subsp *bollettii* (N = 48)	Clofazimine		≤0.5	
		Amikacin		8	
		Clofazimine + amikacin (N = 9)		≤0.5	Synergism seen in 67% of isolates
	M chelonae (N = 57)	Clofazimine		≤0.5	
		Amikacin		8	
		Clofazimine + amikacin (N = 5)		≤0.5	Synergism seen in 80% of isolates
	M fortuitum (N = 44)	Clofazimine		≤0.5	
		Amikacin		≤2.0	
		Clofazimine + amikacin (N = 1)		≤0.5	Synergism seen in 100% of isolates
Singh et al [49][a]	*M abscessus* group (N = 42)	Clofazimine		2 (N = 42)	
		Tigecycline		4 (N = 42)	
		Clofazimine + tigecycline	0.03–4 (N = 19)		Synergism seen in 42% of isolates

Abbreviations: MIC, minimum inhibitory concentration; MIC_{50}, MIC 50%; subsp, subspecies.

[a]For synergy testing, Shen et al [2] used amikacin concentrations of one fourth the amikacin MIC, van Ingen et al [48] used amikacin concentrations of 2 μg/mL, and Singh et al [49] used tigecycline concentrations of 4 μg/mL. In all studies, not all isolates of mycobacterium underwent synergy testing.

growing and rapidly growing mycobacteria and most Gram-positive bacteria in vitro [21, 24]. The exact mechanism(s) of antimicrobial action have yet to be fully elucidated, although the cell membrane seems to be the primary site of action [21]. One putative mechanism of action theorizes that the molecule stimulates phospholipase A_2 activity, resulting in an accumulation of detergent-like lipophospholipids and thus disrupting fundamental cellular functions [21, 24, 25]. It is also proposed that clofazimine may disrupt cell membranes via interaction with intracellular redox cycling, leading to the generation of antimicrobial reactive oxygen species [21, 25]. It has also been hypothesized to inhibit cell replication by binding to the guanine bases of deoxyribonucleic acid [25].

Pharmacokinetics/Pharmacodynamics

Due to its lipophilic nature, clofazimine accumulates in macrophages and adipose cells, thus conferring a long half-life of 65–70 days and an important role as an antimicrobial agent for the treatment of mycobacterial diseases that require prolonged courses of therapy [2, 26]. It also accumulates in the adrenals, heart, liver, pancreas, kidney, spleen, bone marrow, and lamina propria of the jejunum [21, 27, 28]. Oral absorption varies, ranging from 45% to 62%, and is increased when taken with

food [27, 28]. Clofazimine is metabolized by glucoronidation in the liver, partially excreted via the bile, and only minimally excreted in the urine, sputum, sebum, and sweat [25, 27, 28]. It does not cross the blood-brain barrier but does cross the placenta and is excreted in breast milk [21, 24, 25, 27, 28]. It is Pregnancy Category C, but there is a paucity of reports of use for multidrug-resistant tuberculosis (MDR-TB) in pregnancy, and scant data exist for later life teratogenicity [27–31]. Within the noted case reports, only 5 pregnant patients with MDR-TB received clofazimine as part of multidrug therapy, and all infants were considered normal at birth [29, 30]. No significant issues have been reported with clofazimine use as part of multidrug therapy for leprosy during pregnancy [31]. There are no known significant drug-drug interactions.

Uses

In the United States, clofazimine is currently only approved by the US Food and Drug Administration (FDA) for the treatment of leprosy [27, 28, 32]. In the United States, the National Hansen's Disease Program (NHDP) protocol for the treatment of multibacillary leprosy differs both in dosage and duration from the WHO recommendations (rifampicin 600 mg daily, dapsone 100 mg daily, and clofazimine 50 mg daily for 24 months vs rifampicin 600 mg once a month, dapsone 100 mg daily, clofazimine 50 mg daily, and 300 mg once a month for 12 months) [33, 34]. However, clofazimine is not commercially available in the United States and has until recently necessitated an Investigational New Drug (IND) request directly to the FDA [32, 34, 35]. It has also (1) been used in combination with other antimicrobials in the treatment of MAC and (2) as second-line treatment of MDR-TB and extensively drug-resistant TB (XDR-TB) [24, 36–45]. Although previously categorized as a WHO Group 5 drug (one with unclear efficacy), clofazimine is now part of Group C after its importance was underscored when van Deun and colleagues [26, 38, 45] demonstrated its efficacy as part of a shorter regimen for MDR-TB, previously known as the Bangladesh regimen. Due to its anti-inflammatory properties, clofazimine has also been used and/or investigated for use in neutrophilic dermatoses, severe pyoderma gangrenosa, disseminated granuloma annulare, idiopathic panniculitis nodularis, and autoimmune diseases, plus lymphocytic dermatoses such as discoid lupus erythematosus [21, 25, 46]. The mechanism for this immune modulation has not been elucidated.

Adverse Effects

Skin, hair, and corneal changes typically involve reddish to brownish-black to even an orange-pink discoloration. The dyschromia is more apparent in fair-skinned individuals exposed to sunlight. Although slowly reversible, the discoloration may take months to years to resolve [21, 25, 26]. Breast milk, feces, sweat, and urine may become discolored. Relatively common (reported in >10% of patients) adverse reactions include gastrointestinal (GI) disorders (nausea, vomiting, abdominal pain, diarrhea), ichthyosis, and dry skin [21, 27, 28, 32]. Decreased visual acuity, eye irritation/dryness, rash or pruritus, and weight loss occur in 1% to 10% of patients. Less common but more serious adverse effects include enteropathy potentially complicated by intestinal obstruction, GI bleeding, and splenic infarction. The usual leprosy treatment dose is 50 mg daily. Clofazimine has an additional important immunomodulation benefit via the suppression of the Type 2 reaction of erythema nodosum leprosum (ENL) in the treatment of leprosy. Clofazimine takes 4–6 weeks to saturate the reticuloendothelial system to yield this immunosuppressive benefit. Up to 300 mg daily can often be tolerated for the treatment of ENL, but 100 mg daily is the usual dose due to GI intolerance [27, 28, 33, 34]. Higher doses given beyond 4 months may lead to crystal enteropathy and malabsorption. This side effect usually presents with diffuse vague and/or severe abdominal discomfort. Imaging studies usually show edema of the small bowel with effacement of the mucosa. Discontinuation of the drug usually leads to a relatively rapid improvement, although in exceptional circumstances the enteropathy has been fatal [27, 39, 40]. Long-term use as with leprosy often leads to edema of the lower legs. The mechanism may be due to crystal deposition within the lymphatics.

Although adverse effects involving the integumentary and GI systems are not uncommon and have been reported to occur in approximately one third of patients taking clofazimine for the treatment of mycobacterial infections, the severity is not typically significant enough to prompt discontinuation of clofazimine [24, 26, 32, 35, 39, 40].

NICHE FOR CLOFAZIMINE WITH NONTUBERCULOUS MYCOBACTERIA

The rate of lung infection with NTM, especially RGM such as *M abscessus* group, appears to be increasing and is associated with significant morbidity and mortality, especially among patients with underlying structural lung disease such as cystic fibrosis [4–6, 10]. Despite this, the cure rate remains dismal even with prolonged treatment using various multidrug regimens. Although a few laboratories are working on the development of novel antimycobacterial agents, the focus is generally on antituberculous agents. There exists a niche for the use of clofazimine against RGM, especially *M abscessus* group, as evidenced by the in vitro and in vivo results reported by multiple investigators. Of note, many researchers report inconsistency in minimum inhibitory concentration (MIC) results, and the relationship between in vitro susceptibilities and clinical response is still under investigation [4, 5, 15–17]. A possible explanation may be that clofazimine saturates many of the body tissues and has a long half-life leading to a concentration considerably higher than the NTM MIC.

In Vitro Studies

Early in vitro studies revealed that rapidly growing mycobacteria were susceptible to clofazimine [47]. In the last 10 years,

3 large in vitro studies of clofazimine against RGM isolates showed excellent results. Shen et al [2] reported an MIC of <1 μg/mL in 180 isolates of *M abscessus* group, *M fortuitum*, and *Mycobacterium chelonae* and noted synergism between clofazimine and amikacin. van Ingen et al [48] found that 97% of 564 isolates of 21 species of RGM had an MIC <1 μg/mL and also observed synergism between clofazimine and amikacin. Singh et al [49] found clofazimine to have the lowest MIC 50 among 7 antibiotics studied on 67 isolates of *M abscessus* group and noted synergism between clofazimine and tigecycline.

In Vivo Studies

In fly models, clofazimine extended the lifespan of *Drosophila* infected with *M abscessus* group by 2–3 days in comparison to control groups that received no treatment [50]. However, the results with clofazimine were not as impressive as those observed with tigecycline, followed by clarithromycin and linezolid.

Obregón-Hanao et al [5] demonstrated in mice lacking interferon-gamma that clofazimine treatment afforded a statistically significant reduction in the bacterial burden in the lung, spleen, and liver after 8 days of treatment. These anti-mycobacterial effects were augmented by the addition of bedaquiline. Significant reduction in bacterial burden in the lungs was not seen when amikacin, clarithromycin, or ciprofloxacin alone were used for 8 days. In severe combined immunodeficiency mouse models (lacking functional lymphocytes and natural killer cells), treatment with clarithromycin, clofazimine, or bedaquiline each individually significantly reduced bacterial burden in the lung, spleen, and liver. Again, the most significant reduction was observed with the combination treatment of clofazimine and bedaquiline together. However, it is also worth noting that other studies have demonstrated low-level cross-resistance between clofazimine and bedaquiline in *M tuberculosis* due to mutations in *the rv0678* gene, a regulator of the MmpL5 and MmpS5 multisubstrate efflux pumps, and/or mutations within the aminopeptidase gene *pepQ* (rv3525c) [24, 51–53]. Per Basic Local Alignment Search Tool (BLAST) search, PepQ, MmpL5, and MmpS5 homologs exist in a number of other mycobacteria including *M fortuitum*, *Mycobacterium intracellulare*, and *M leprae*, and thus similar mutations could confer cross-resistance.

A recent retrospective review by Yang et al [54] examined the clinical, radiographic, and microbiologic outcomes of patients with *M abscessus* group lung disease who received clofazimine as part of an initial or salvage multidrug regimen. They found that after 12 months of a clofazimine-containing regimen, 81% of patients exhibited treatment response based on symptomatic improvement, 31% demonstrated radiographic improvement, and 24% achieved sputum-negative culture conversion. These improvements were all greater in the group receiving clofazimine as part of an initial drug regimen, although not always to a degree that was statistically significant. Another group evaluated quality of life of patients undergoing treatment for *M abscessus* group lung disease and found that although quality of life was improved at various follow-up time points, and positively associated with an isolate susceptible to imipenem-cilastin and with lung resection surgery, there was neither a positive nor negative association with clofazimine use or any other of the other evaluated antibiotics [55]. There is limited additional data regarding clinical outcomes of patients with *M abscessus* group pulmonary disease treated with clofazimine [8, 56].

Clofazimine-containing regimens have also been used and studied in the treatment of MDR-TB and XDR-TB, and they were found to promote cavity closure, accelerate sputum culture conversion, and improve treatment success rates [26, 32, 40, 41, 45]. Clofazimine has also been evaluated in the treatment of MAC lung disease and bacteremia. Clinical results have been inconsistent, although additional clinical trials are underway [36, 42–44, 57].

Synergy

Numerous studies suggest a synergistic effect between clofazimine and other antimicrobials such as amikacin, tigecycline, clarithromycin, isoniazid, ethambutol, pyrazinamide, linezolid, and bedaquiline against various species of both rapidly growing and slowly growing NTM [2, 5, 17, 24, 32, 49, 58, 59]. A biologic explanation may be that via its cell wall-destabilizing properties, clofazimine allows for increased influx of other antibiotics with intracellular targets [17, 40]. In addition, clofazimine may help prevent acquired resistance to aminoglycosides and macrolides by preventing point mutations in drug targets and activation of drug-efflux pumps [58]. The optimum combination of antimicrobials remains to be established, and this likely varies with the species/subspecies in question, the anatomical site involved, and the immune status of the patient.

Difficulty Obtaining

Since 2004, clofazimine has not been available in the United States for any FDA-approved indication other than treatment of leprosy, except via a single-patient IND process with the FDA, and then the drug has been distributed by the NHDP [35]. As of January 2017, Novartis and the FDA have agreed to an "intermediate-sized" IND protocol, which may simplify the process. Interested healthcare providers begin by contacting Novartis directly (1-888-669-6682). The process remains burdensome. In addition to completing paperwork required by Novartis and the FDA, physicians must obtain written consent from patients and approval from an Institutional Review Board because the prescription is considered to be experimental for this off-label purpose. In most cases, delivery of the medication takes approximately 2 weeks from approval of the application. Clofazimine is not approved for children or pregnant patients on this IND. Male and female patients must be using highly effective birth control during and up to 4 months after stopping treatment. From January 1, 2005 through the end of 2011

the FDA approved 649 single patient IND applications for the treatment of NTM [35]. Data regarding the new Novartis IND protocol are not available.

CONCLUSIONS

Nontuberculous mycobacteria disease is increasing in prevalence and represents an important health issue, particularly for predisposed individuals. Advances are needed in the treatment of nearly all NTM infections, particularly RGM such as *M abscessus* group, to provide more effective, simple, and tolerable treatment options. The repurposing of the "old" drug clofazimine may fill this niche. Given the multitude of clinical variables possible (NTM species/subspecies, site of infection, patient immune status and comorbidities, accompanying antimicrobials, adjunctive nonpharmacologic therapies, etc.), need for long-term follow-up, and limits of the observational studies performed to date, additional studies are needed to help elucidate this potential new role for clofazimine. The efficacy and safety of clofazimine should be further tested as part of multidrug regimens for the treatment of RGM.

Acknowledgments

We thank Dr. Diana Willaims for assistance in obtaining information regarding PepQ, MmpL5, and MmpS5 homologs.

Financial support. This effort was supported by the University of Washington Internal Medicine Residency Training Program.

Potential conflicts of interest. All authors: No reported conflicts of interest.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References

1. Winthrop KL, McNelley E, Kendall B, et al. Pulmonary nontuberculous mycobacterial disease prevalence and clinical features: an emerging public health disease. Am J Respir Crit Care Med **2010**; 182:977–82.
2. Shen GH, Wu BD, Hu ST, et al. High efficacy of clofazimine and its synergistic effect with amikacin against rapidly growing mycobacteria. Int J Antimicrob Agents **2010**; 35:400–4.
3. Benwill JL, Wallace RJ Jr. *Mycobacterium abscessus*: challenges in diagnosis and treatment. Curr Opin Infect Dis **2014**; 27:506–10.
4. Griffith DE, Aksamit T, Brown-Elliott BA, et al. An official ATS/IDSA statement: diagnosis, treatment, and prevention of nontuberculous mycobacterial diseases. Am J Respir Crit Care Med **2007**; 175:367–416.
5. Obregón-Henao A, Arnett KA, Henao-Tamayo M, et al. Susceptibility of *Mycobacterium abscessus* to antimycobacterial drugs in preclinical models. Antimicrob Agents Chemother **2015**; 59:6904–12.
6. Fleshner M, Olivier KN, Shaw PA, et al. Mortality among patients with pulmonary non-tuberculous mycobacteria disease. Int J Tuberc Lung Dis **2016**; 20:582–7.
7. Nessar R, Cambau E, Reyrat JM, et al. *Mycobacterium abscessus*: a new antibiotic nightmare. J Antimicrob Chemother **2012**; 67:810–8.
8. Jarand J, Levin A, Zhang L, et al. Clinical and microbiologic outcomes in patients receiving treatment for *Mycobacterium abscessus* pulmonary disease. Clin Infect Dis **2011**; 52:565–71.
9. Mirsaeidi M, Machado RF, Garcia JG, Schraufnagel DE. Nontuberculous mycobacterial disease mortality in the United States, 1999–2010: A population-based comparative study. PLoS One **2014**; 9:1–9.
10. Prevots DR, Marras TK. Epidemiology of human pulmonary infection with nontuberculous mycobacteria: a review. Clin Chest Med **2015**; 36:13–34.
11. Stout JE, Koh WJ, Yew WW. Update on pulmonary disease due to non-tuberculous mycobacteria. Int J Infect Dis **2016**; 45:123–34.
12. Runyon EH. Anonymous mycobacteria in pulmonary disease. Med Clin North Am **1959**; 43:273–90.
13. Tortoli E. Microbiological features and clinical relevance of new species of the genus *Mycobacterium*. Clin Microbiol Rev **2014**; 27:727–52.
14. Griffith DE, Brown-Elliott BA, Benwill JL, Wallace RJ Jr. *Mycobacterium abscessus*. "Pleased to meet you, hope you guess my name." Ann Am Thorac Soc **2015**; 12:436–9.
15. Griffith DE. *Mycobacterium abscessus* subsp abscessus lung disease: 'trouble ahead, trouble behind…'. F1000Prime Rep **2014**; 6:107.
16. Brown-Elliott BA, Nash KA, Wallace RJ Jr. Antimicrobial susceptibility testing, drug resistance mechanisms, and therapy of infections with nontuberculous mycobacteria. Clin Microbiol Rev **2012**; 25:545–82.
17. van Ingen J, Boeree MJ, van Soolingen D, Mouton JW. Resistance mechanisms and drug susceptibility testing of nontuberculous mycobacteria. Drug Resist Updat **2012**; 15:149–61.
18. Woods GL. Susceptibility testing for mycobacteria. Clin Infect Dis **2000**; 31:1209–15.
19. New drug for leprosy. Br Med J **1969**; 1:797–8.
20. Barry VC, Belton JG, Conalty ML, et al. A new series of phenazines (rimino-compounds) with high antituberculosis activity. Nature **1957**; 179:1013–5.
21. Cholo MC, Steel HC, Fourie PB, et al. Clofazimine: current status and future prospects. J Antimicrob Chemother **2012**; 67:290–8.
22. Browne SG, Hogerzeil LM. "B 663" in the treatment of leprosy. Preliminary report of a pilot trial. Lepr Rev **1962**; 33:6–10.
23. World Health Organization. Chemotherapy of leprosy for control programmes. WHO Technical Report Series, No. 675. Geneva, Switzerland. **1982**. Available at: http://apps.who.int/iris/bitstream/10665/38984/1/WHO_TRS_675.pdf. Accessed 1 June 2017.
24. Cholo MC, Mothiba MT, Fourie B, Anderson R. Mechanisms of action and therapeutic efficacies of the lipophilic antimycobacterial agents clofazimine and bedaquiline. J Antimicrob Chemother **2017**; 72:338–53.
25. Arbiser JL, Moschella SL. Clofazimine: a review of its medical uses and mechanisms of action. J Am Acad Dermatol **1995**; 32:241–7.
26. Gopal M, Padayatchi N, Metcalfe JZ, O'Donnell MR. Systematic review of clofazimine for the treatment of drug-resistant tuberculosis. Int J Tuberc Lung Dis **2013**; 17:1001–7.
27. Novartis. Lamprene® Prescribing Information. NDA 19-500/S-010. Novartis. East Hanover, NJ. 2002. Available at: https://www.accessdata.fda.gov/drugsatfda_docs/label/2003/19500slr010_lamprene_lbl.pdf. Accessed 1 June 2017.
28. Food and Drug Administration. LAMPRENE ® (clofazimine) capsules, Full prescribing information. Reference ID: 3956651. U.S. Food and Drug Administration. Silver Spring, MD. Available at: https://www.accessdata.fda.gov/drugsatfda_docs/label/2016/019500s013lbl.pdf. Accessed 1 June 2017.
29. Drobac PC, del Castillo H, Sweetland A, et al. Treatment of multidrug-resistant tuberculosis during pregnancy: long-term follow-up of 6 children with intrauterine exposure to second-line agents. Clin Infect Dis **2005**; 40:1689–92.
30. Shin S, Guerra D, Rich M, et al. Treatment of multidrug-resistant tuberculosis during pregnancy: a report of 7 cases. Clin Infect Dis **2003**; 36:996–1003.
31. Ozturk Z, Tatliparmak A. Leprosy treatment during pregnancy and breastfeeding: a case report and brief review of literature. Dermatol Ther **2017**; 30:e12414.
32. Hwang TJ, Dotsenko S, Jafarov A, et al. Safety and availability of clofazimine in the treatment of multidrug and extensively drug-resistant tuberculosis: analysis of published guidance and meta-analysis of cohort studies. BMJ Open **2014**; 4:e004143.
33. World Health Organization. WHO Model Prescribing Information: Drugs Used in Leprosy. Geneva, Switzerland. World Health Organization, 1998. Available at: http://apps.who.int/medicinedocs/pdf/h2988e/h2988e.pdf. Accessed 1 June 2017.
34. National Hansen's Disease (Leprosy) Program. Recommended Treatment Regimens. US Department of Health and Human Services. Washington DC. Available at: https://www.hrsa.gov/hansensdisease/diagnosis/recommendedtreatment.html. Accessed 1 June 2017.
35. Vaidya P, O'Shaughnessy E, Mauer R, et al. Clofazimine use in patients with mycobacterial infections under Single Patient Investigational New Drug (SPIND) [poster]. ID Week 2012 (San Diego, CA).
36. Jo KW, Kim S, Lee JY, et al. Treatment outcomes of refractory MAC pulmonary disease treated with drugs with unclear efficacy. J Infect Chemother **2014**; 20:602–6.
37. World Health Organization. WHO treatment guidelines for drug-resistant tuberculosis, 2016 update, annexes 4, 5, and 6. Geneva, Switzerland: World Health Organization; **2016**. Available at: http://www.who.int/tb/areas-of-work/drug-resistant-tb/MDRTBguidelines2016.pdf. Accessed 1 June 2017.
38. Van Deun A, Maug AK, Salim MA, et al. Short, highly effective, and inexpensive standardized treatment of multidrug-resistant tuberculosis. Am J Respir Crit Care Med **2010**; 182:684–92.
39. Arbiser JL, Moschella SL, Ausina V, et al. Clinical and microbiologic outcomes in patients receiving treatment for *Mycobacterium abscessus* pulmonary disease. J Antimicrob Chemother **2014**; 20:1–9.
40. Tang S, Yao L, Hao X, et al. Clofazimine for the treatment of multidrug-resistant tuberculosis: prospective, multicenter, randomized controlled study in China. Clin Infect Dis **2015**; 60:1361–7.

41. Piubello A, Harouna SH, Souleymane MB, et al. High cure rate with standardised short-course multidrug-resistant tuberculosis treatment in Niger: no relapses. Int J Tuberc Lung Dis **2014**; 18:1188–94.
42. Jarand J, Davis JP, Cowie RL, et al. Long-term follow-up of *Mycobacterium avium* complex lung disease in patients treated with regimens including clofazimine and/or rifampin. Chest **2016**; 149:1285–93.
43. Field SK, Cowie RL. Treatment of *Mycobacterium avium-intracellulare* complex lung disease with a macrolide, ethambutol, and clofazimine. Chest **2003**; 124:1482–6.
44. Cariello PF, Kwak EJ, Abdel-Massih RC, Silveira FP. Safety and tolerability of clofazimine as salvage therapy for atypical mycobacterial infection in solid organ transplant recipients. Transpl Infect Dis **2015**; 17:111–8.
45. Aung KJ, Van Deun A, Declercq E, et al. Successful '9-month Bangladesh regimen' for multidrug-resistant tuberculosis among over 500 consecutive patients. Int J Tuberc Lung Dis **2014**; 18:1180–7.
46. Gurfinkel P, Pina JC, Ramos-e-Silva M. Use of clofazimine in dermatology. J Drugs Dermatol **2009**; 8:846–51.
47. Ausina V, Condom MJ, Mirelis B, et al. In vitro activity of clofazimine against rapidly growing nonchromogenic mycobacteria. Antimicrob Agents Chemother **1986**; 29:951–2.
48. van Ingen J, Totten SE, Helstrom NK, et al. In vitro synergy between clofazimine and amikacin in treatment of nontuberculous mycobacterial disease. Antimicrob Agents Chemother **2012**; 56:6324–7.
49. Singh S, Bouzinbi N, Chaturvedi V, et al. In vitro evaluation of a new drug combination against clinical isolates belonging to the *Mycobacterium abscessus* complex. Clin Microbiol Infect **2014**; 20:O1124–7.
50. Oh CT, Moon C, Park OK, et al. Novel drug combination for *Mycobacterium abscessus* disease therapy identified in a *Drosophila* infection model. J Antimicrob Chemother **2014**; 69:1599–607.
51. Hartkoorn RC, Uplekar S, Cole ST. Cross-resistance between clofazimine and bedaquiline through upregulation of MmpL5 in *Mycobacterium tuberculosis*. Antimicrob Agents Chemother **2014**; 58:2979–81.
52. Xu J, Wang B, Hu M, et al. Primary clofazimine and bedaquiline resistance among isolates from patients with multidrug-resistant tuberculosis. Antimicrob Agents Chemother **2017**; 61:pii: e00239-17.
53. Almeida D, Ioerger T, Tyagi S, et al. Mutations in pepQ confer low-level resistance to bedaquiline and clofazimine in *Mycobacterium tuberculosis*. Antimicrob Agents Chemother **2016**; 60:4590–9.
54. Yang B, Jhun BW, Moon SM, et al. A clofazimine-containing regimen for the treatment of *Mycobacterium abscessus* lung disease. Antimicrob Agents Chemother **2017**; 61:pii: e02052-16.
55. Czaja CA, Levin AR, Cox CW, et al. Improvement in quality of life after therapy for *mycobacterium abscessus* group lung infection. A prospective cohort study. Ann Am Thorac Soc **2016**; 13:40–8.
56. Winthrop K. Phase 2 Study of Clofazimine for the Treatment of Pulmonary Mycobacterium Avium Disease. U.S. National Institutes of Health. ClinicalTrials.gov identifier: NCT02968212. 2017. Available at: https://clinicaltrials.gov/ct2/show/NCT02968212. Accessed 2 June 2017.
57. Chaisson RE, Keiser P, Pierce M, et al. Clarithromycin and ethambutol with or without clofazimine for the treatment of bacteremic *Mycobacterium avium* complex disease in patients with HIV infection. AIDS **1997**; 11:311–7.
58. Ferro BE, Meletiadis J, Wattenberg M, et al. Clofazimine prevents the regrowth of *Mycobacterium abscessus* and *Mycobacterium avium* type strains exposed to amikacin and clarithromycin. Antimicrob Agents Chemother **2016**; 60:1097–105.
59. Zhang Z, Li T, Qu G, et al. In vitro synergistic activity of clofazimine and other antituberculous drugs against multidrug-resistant *Mycobacterium tuberculosis* isolates. Int J Antimicrob Agents **2015**; 45:71–5.