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Mycobacterium abscessus: a new antibiotic nightmare

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The intrinsic and acquired resistance of *Mycobacterium abscessus* to commonly used antibiotics limits the chemotherapeutic options for infections caused by these mycobacteria. Intrinsic resistance is attributed to a combination of the permeability barrier of the complex multilayer cell envelope, drug export systems, antibiotic targets with low affinity and enzymes that neutralize antibiotics in the cytoplasm. To date, acquired resistance has only been observed for aminoglycosides and macrolides, which is conferred by mutations affecting the genes encoding the antibiotic targets (*rrs* and *rrl*, respectively). Here we summarize previous and recent findings on the resistance of *M. abscessus* to antibiotics in light of what has been discovered for other mycobacteria. Since we can now distinguish three groups of strains belonging to *M. abscessus* (*M. abscessus sensu stricto*, *Mycobacterium massiliense* and *Mycobacterium bolletii*), studies on antibiotic susceptibility and resistance should be considered according to this new classification. This review raises the profile of this important pathogen and highlights the work needed to decipher the molecular events responsible for its extensive chemotherapeutic resistance.

Keywords: natural, acquired, resistance

Introduction

Mycobacterium abscessus is a rapidly growing mycobacteria (RGM) first described by Moore and Frerichs in 1953.[1] However, it was only in 1992, after its separation from the *Mycobacterium chelonae* group, that *M. abscessus* acquired the recognition that it is an important human pathogen responsible for a wide spectrum of soft tissue infections, disseminated infection in immunocompromised patients and a contraindication to lung transplantation.[2] *M. abscessus* is now considered the prominent *Mycobacterium*, along with *Mycobacterium avium*, involved in broncho-pulmonary infection in patients with cystic fibrosis or chronic pulmonary disease.[3–6] Several outbreaks of *M. abscessus* skin and soft tissue infections have also recently been reported, demonstrating this organisms importance in healthcare-associated infections, including surgical tourism.[7–10] The major threat posed by this species is mainly due to its resistance to antibiotics, which is of major concern in public health institutions.[11] Indeed, *M. abscessus* is one of the most resistant organisms to chemotherapeutic agents.[12] Elucidating the molecular mechanisms responsible for this particular trait has become an increasing research focus, particularly after the genome sequence became available in 2009.[13] Interestingly, genome analysis has revealed that *M. abscessus* shares a number of common characteristics with some slow-growing mycobacteria (SGM), and this has led to intriguing questions such as: (i) are the resistance mechanisms similar to those found in SGM; and (ii) what additional characteristics of this organism make it particularly resistant to antibiotic therapy? Antibiotic resistance in mycobacterial species can be either natural or acquired, and for the latter, resistance is not reported to be provided by genes introduced by transmissible genetic elements such as plasmids and transposons, but by spontaneous mutation at targeted genes in response to the presence of antibiotics. The absence of reports of plasmid-encoded antibiotic resistance is due in part to the problem of discerning added resistance by extrachromosomal genetic determinants against the very high intrinsic antibiotic resistance of mycobacteria.

Recently the *M. abscessus* species has been subclassified into three new species on the basis of *rpoB* sequences: *M. abscessus sensu stricto*, *Mycobacterium massiliense* and *Mycobacterium bolletii*.[14] They constitute what is now called the *M. abscessus* group, or *M. abscessus sensu lato*. Further taxonomic studies have shown that differentiation of the three species is not straightforward; they share ribosomal sequences and multilocus sequencing approaches cannot clearly assign clinical strains to one of the three species.[7,9,15,16] These species or subspecies,

however, can also differ from each other in their antibiotic resistance phenotype and genotype, indicating that studies on precisely identified strains are warranted.[17,18] For instance, clarithromycin susceptibility is observed for *M. massiliense*, whereas resistance is observed in *M. bolletii*.[17]

Antibiotic susceptibility and efficacy

Infections due to the *M. abscessus* group are difficult to treat because these mycobacteria are intrinsically resistant to not only the classical anti-tuberculous drugs, but also to most of the antibiotics that are currently available.[12,19–21] Few drugs have *in vitro* activity against *M. abscessus* (Table 1). Modal MICs are below the tissue or serum levels only for clarithromycin, aminoglycosides, cefoxitin, tigecycline and TMC-207. However, some strains appear much more susceptible to some drugs, and this may relate to the difference in the subspecies within the *M. abscessus* group.

In the 1990s clarithromycin became the drug of choice for *M. abscessus* infections and therapeutic successes were reported.[4,22,23] Recommendations are now to combine clarithromycin with one aminoglycoside (usually amikacin) and one other injectable drug such as cefoxitin or imipenem.[2] Clinical efficacy of this multidrug therapy is still controversial, with success for some patients and failure for others.[20,24]

Natural resistance

A number of mechanisms are responsible for natural resistance of *M. abscessus* and other mycobacterial species to drugs, including slow growth, the presence of a waxy impermeable cell wall, which acts as a physical (size exclusion) and a chemical (hydrophobic) barrier, drug export systems and genetic polymorphism of targeted genes.

The mycobacterial cell envelope

The role of the mycobacterial cell envelope in conferring resistance to drugs has been extensively studied. In 1990 Jarlier and Nikaido indicated the essential role that the lack of permeability of the cell envelope played in making *M. chelonae* (grouped at that time in the same species with *M. abscessus*) resistant to antibiotics.[25] In the case of the β-lactams, the *M. chelonae* cell envelope drastically reduced the influx of β-lactam antibiotics and, together with the low level β-lactamase activity, was sufficient to explain the low activity of β-lactams against the *M. chelonae* group.[25] It is also likely that the low permeability of the cell envelope of *M. abscessus* (acting in synergy with aminoglycoside-modifying enzymes) plays a role in aminoglycoside resistance.[25] The existence of the cell wall barrier also explains the intrinsic resistance of mycobacterial cells to acids and alkalis.[26] A key feature of the mycobacterial cell envelope is its high lipid content (up to 60% of the dry weight of the bacteria), which is considered to be the main factor contributing to its low permeability.[27]

The mycobacterial cell envelope plays a crucial role in protecting the cell against toxic extracellular compounds. The presence of porins enable the rapid passage of potentially lethal amounts of compounds and hydrophilic antibiotics through the envelope.[28] Once internalized the antibiotics can reach their target in the cytoplasm and activate the expression of potential drug resistance genes. It is well documented that the cell envelope acts synergistically with antibiotic-inducible internal systems in competing against the effects of the drugs.[29] This internal system, known as the 'intrinsic resistome', includes efflux pumps, antibiotic-modifying/inactivating enzymes, target-modifying enzymes and genes conferring metal resistance (Table 2).

Antibiotic-modifying/inactivating enzymes

M. abscessus produces enzymes that potentially degrade or modify antibiotics, which can result in their inactivation.

Table 1. Antibiotic susceptibility of *M. abscessus* as defined by MIC

Antibiotic	*n*	Modal MIC (mg/L)	MIC range (mg/L)	Percentage susceptibility[a]	References
clarithromycin	48, 74	0.03	0.03–16	83, 99	89, 90
cefoxitin	48, 74	32	16–128	11, 99	89, 90
imipenem	48, 74	8	1–64	8, 55	89, 90
ciprofloxacin	48, 74	2	0.016–8	44, 57	89, 90
levofloxacin	21	32	8–64		91
moxifloxacin	21	16	2–32	73	91
doxycycline	48, 20	32, >128	0.06–32, 2–>128	8, 5	90, 92
tigecycline	20	0.12	≤0.06–1	100	92
minocycline	20	>64	0.25–>64	5	92
tetracycline	20	64	4–>128	10	92
linezolid	98	32	0.5–128	23	93
sulfamethoxazole	48, 74	256	4–256	12, 1	89, 90
isepamicin	117	8	4–>128	96	94
tobramycin	21, 117, 74	16, 8	8–32, 4–>128	95, 36	91, 94, 90
amikacin	48, 117	2, 16	0.25–128, 4–>128	94, 87	90, 94
TMC-207	1	0.25			95
clofazimine	117	0.5	0.25–1	99	96

[a]According to breakpoints defined in Griffith *et al.*[2] and Woods *et al.*[97]

Table 2. Synopsis of the genes and the possible mechanisms involved in natural resistance of *M. abscessus*

Antibiotic	Locus and genes	Proteins involved	Mechanism of resistance
hydrophilic antibiotics			selective permeability of cell envelope
aminoglycosides	MAB_4395 MAB_0327, MAB_0951 MAB_3637c, MAB_4910c, MAB_4395	aminoglycoside 2-*N*-acetyltransferase aminoglycoside phosphotransferases	antibiotic-modifying enzymes
rifampicin	MAB_0951	rifampicin ADP-ribosyltransferase	
β-lactams	MAB_2875	β-lactamase	antibiotic-degrading enzymes
macrolides	*erm(41)* gene MAB_2297	23S RNA methyltransferase	target-modifying enzymes
several antibiotics	scattered in genome	ABC transporters MmpL family	efflux pumps
metal compounds	plasmid pMMV23 MAB_p05c, MAB_06c	mercury operon regulator *Mer*R, mercury reductase; *ars* operon	efflux pumps/detoxification

M. abscessus possesses a rifampicin ADP-ribosyltransferase, as well as a mono-oxygenase that may be involved in resistance to rifampicin.[13] The fast-growing *Mycobacterium smegmatis* is naturally resistant to rifampicin, although no mutation in the target gene *rpo*B has been reported.[30] Quan *et al*.[31] reported in 1997 that a ribosylation mechanism is responsible for the inactivation of rifampicin and represents the principal contributor to the low susceptibility of *M. smegmatis* to rifampicin. It is conceivable that the same phenomenon could well operate in *M. abscessus*, since no mutation has been reported in the *rpoB* gene from *M. abscessus* clinical isolates resistant to rifampicin.[32] *M. abscessus* also contains enzymes that could modify aminoglycoside drugs by transferring acetyl or phosphate residues on key positions within the antibiotic, rendering them inactive.[13] *M. abscessus* contains an aminoglycoside 2-*N*-acetyltransferase and several homologs of aminoglycoside phosphotransferases. Acetyltransferases and phosphotransferases from *M. smegmatis* and *Mycobacterium tuberculosis* have been reported to confer aminoglycoside resistance.[33,34] Antibiotic-degrading enzymes, for example, β-lactamases, can also assist some mycobacterial species to nullify the effect of antibiotics and thus confer resistance to β-lactam antibiotics.[29] Genetic analysis has revealed the presence of β-lactamase-encoding genes in *M. abscessus* and in SGM including *M. tuberculosis*.[13,35]

Target-modifying enzymes

Macrolide antibiotics are generally used to treat infections caused by non-tuberculous mycobacteria (NTM).[2,36] However, *M. abscessus* infections tend to respond poorly to macrolide chemotherapy. Recent reports demonstrate that intrinsic resistance to macrolides in *M. abscessus* clinical isolates is due to the expression of a novel inducible *erm* gene, *erm(41)* (MAB_2997), which is induced by macrolides and confers resistance to clarithromycin and erythromycin.[37] Furthermore, the same gene has been shown to confer resistance to clindamycin and telithromycin in *M. smegmatis*, although *M. abscessus* is naturally resistant to these two agents by a mechanism that is independent of *erm* gene induction.[37]

Efflux pumps

Active efflux mechanisms represent potentially one of the causative factors of antibiotic resistance in mycobacteria.[38,39] Efflux pump mechanisms have a physiological role protecting bacteria against toxic molecules and maintaining cell homeostasis and physiological balance through export of toxins or metabolites to the extracellular environment.[39] *M. abscessus* encodes protein members of the major facilitator family ABC transporters and mycobacterial membrane protein large (MmpL) families.[13] The ABC-type multidrug transporters use ATP energy to pump drugs out of the cell and can be classified either as importers (uptake of extracellular molecules) or exporters (remove substrates to the external environment).[40,41]

The MmpL transporter family is involved in lipid transport to the membrane and encode resistance, nodulation and cell division (RND) proteins, which are a family of multidrug resistance pumps that recognize and mediate the transport of a diverse group of compounds (cationic, anionic or neutral), including various drugs, metals and fatty acids.[42] These proteins mediate transport across the cytoplasmic membrane using the proton motive force of the transmembrane electrochemical proton gradient.[43] Genes for members of the MmpL transporter family are distributed throughout the *M. abscessus* genome, but their role in this species has yet to be established. Recent studies have attributed a drug resistance function to the MmpL family.[13] Pasca *et al*.[44] demonstrated that the *mmpL7* gene from *M. tuberculosis* confers a high level of resistance to isoniazid when overexpressed in *M. smegmatis* and the resistance level was significantly decreased in the presence of efflux inhibitors. However, Domenech *et al*.[42] constructed *M. tuberculosis* mutant strains with 11 of 13 of the *mmpL* genes inactivated and reported that drug susceptibility of these mutants to a broad spectrum of agents was unaltered. This led the authors to suggest that,

unlike their function in other organisms, these proteins do not play a significant role in the intrinsic drug resistance of *M. tuberculosis*.

The P55 efflux pump was also shown to be involved in natural resistance in *M. tuberculosis*, since after deletion of the corresponding gene the bacteria became more susceptible to toxic compounds including rifampicin and clofazimine.[45] Of note, this pump was inhibited by carbonyl cyanide *m*-chlorophenylhydrazone (CCCP) and valinomycin.[45]

Transcriptional regulator whiB gene family

M. abscessus is equipped with a family of transcriptional regulators potentially involved in conferring drug resistance (the *whiB* gene family). This family is exclusively present in the actinomycetes (there are six *whiB* genes within *M. abscessus*) and *Streptomyces* genomes.[46] The WhiB proteins are putative transcription factors involved in the regulation of significant cellular processes such as cell division, pathogenesis and responses to oxidative stress, and the presence of a helix-turn-helix motif indicates a DNA binding role.[47–50] The *whiB7* gene in *M. tuberculosis* has been shown to be induced by exposure to subinhibitory concentrations of antibiotic. Microarray analysis demonstrated that upon subinhibitory exposure to tetracycline, the expression of a cluster of genes was dependent on the induction of *whiB7*.[51] Other *M. tuberculosis whiB* family members have also exhibited conditional up-regulation in response to environmental changes.[52,53] The *M. tuberculosis* null mutant *whiB7* is hypersusceptible to a large spectrum of antibiotics, and *whiB7* null mutants of *M. smegmatis* and *M. bovis* also show the same susceptibility pattern.[46] Other members of the *whiB* family have also been shown to be involved in drug resistance. Geiman *et al.*[51] studied the transcription of *whiB* genes in *M. tuberculosis* and demonstrated *whiB2* is responsive to antimicrobial stress. The expression of *whiB2* was stimulated by exposure to a spectrum of antibiotic agents (isoniazid, ethambutol and cycloserine) that inhibit cell wall biosynthesis in mycobacteria.

Genetic polymorphism of target genes

The presence of variant nucleotides within conserved genes targeted by drugs has been associated with establishment of a correlation between genotype and susceptibility to drugs within NTM. Two examples are highlighted by ethambutol and fluoroquinolone resistance in NTM.[54,55] *M. abscessus* exhibits intrinsic high-level resistance to ethambutol (MICs >64 mg/L), and much of this resistance is due to the presence of variant nucleotides within the conserved *emb*B ethambutol resistance-determining region (ERDR) (Figure 1).[55] The mycobacterial *emb*CAB operon encodes arabinosyl transferases, which are putative targets for ethambutol. Mutations in *emb*B have been associated with resistance to ethambutol in *M. tuberculosis*.[56] Transfer of the *emb* region carrying the variant allele to the drug-susceptible *M. smegmatis* resulted in a 500-fold increase in the MICs to ethambutol.[55] Sequencing of the conserved ERDRs of 13 NTM strains allowed the identification of a unique variant sequence that was associated with ethambutol resistance. When compared with ethambutol-susceptible *M. tuberculosis*, three NTM strains—*Mycobacterium leprae*, *M. chelonae* and *M. abscessus*—had isoleucine substituted with glutamine at position 303 and leucine substituted with methionine at position 304 (I303Q and L304M) (Figure 1). This variation conferred intrinsic high-level resistance to ethambutol in the three strains.

Species	ERDR	MIC (mg/L)	Phenotype
M. tuberculosis	SDDY**ILGM**ARVADHAGYMSN	2.5	S
M. chelonae	SDDY**QMGM**AR**T**A**E**HAGYM**A**N	64	R
M. abscessus	SDDY**QMGM**AR**T**A**E**HAGYM**A**N	64	R
M. leprae	SDDY**QMQM**AR**T**A**D**H**S***GYM**A**N*	64	R

Figure 1. Comparison between ethambutol phenotype and genetic polymorphism at the ERDR in EmbB with gene polymorphisms indicated at positions 303 (I303Q) and 304 (L304M). S, susceptible; R, resistant. Adapted from Sreevatsan *et al.*[56]

Another example of the role of gene polymorphism in conferring drug resistance has been observed with the fluoroquinolones. Fluoroquinolones have been used in combination with other anti-mycobacterial agents for infection caused by *M. abscessus*.[57] Guillemin *et al.*[54] studied intrinsic resistance to fluoroquinolones in NTM by determining the sequences of conserved regions known as quinolone resistance-determining regions (QRDRs) in the DNA gyrase subunits GyrA and GyrB (the regions targeted by quinolones). It was shown that the presence of alanine at position 83 (Ala-83) (*Escherichia coli* numbering) within GyrA QRDR and arginine and asparagine at positions 447 and 464 (Arg-447 and Asn-464), respectively, within GyrB QRDR confer resistance to fluoroquinolones in *M. abscessus* (MICs >8 mg/L) as well as in *M. avium*, *Mycobacterium intracellulare*, *Mycobacterium marinum* and *M. chelonae* (Figure 2).[58]

Mercury resistance

Bacterial resistance to inorganic and organic mercury compounds (HgR) has been studied extensively in eubacteria.[59] The genes encoding the proteins responsible for mercury resistance occur naturally on the chromosome and on plasmid and transposable elements.[59] Two major components are required to confer bacterial resistance to mercury: the regulator MerR and the major detoxification enzyme MerA. The resistance of some mycobacterial strains is related to the presence of a megaplasmid probably containing mercury resistance genes because the mercury resistance is carried by 'transferable' elements.[60,61] *M. abscessus* contains a 23 kb mercury resistance plasmid that is 99% identical to pMM23 from *M. marinum*. This plasmid carries a *mer* operon with mercury operon regulator *Mer*R (MAB_p05c) and a mercury reductase (MAB_06c), which probably confers resistance to a wide range of organomercury compounds.[13] Although the mechanism of mercury resistance has been well characterized in other eubacterial species, further studies are needed to decipher the mechanism of mercury resistance in *M. abscessus*.

Acquired resistance

Acquired resistance as a result of genotypic changes within mycobacterial clinical isolates does not appear to involve

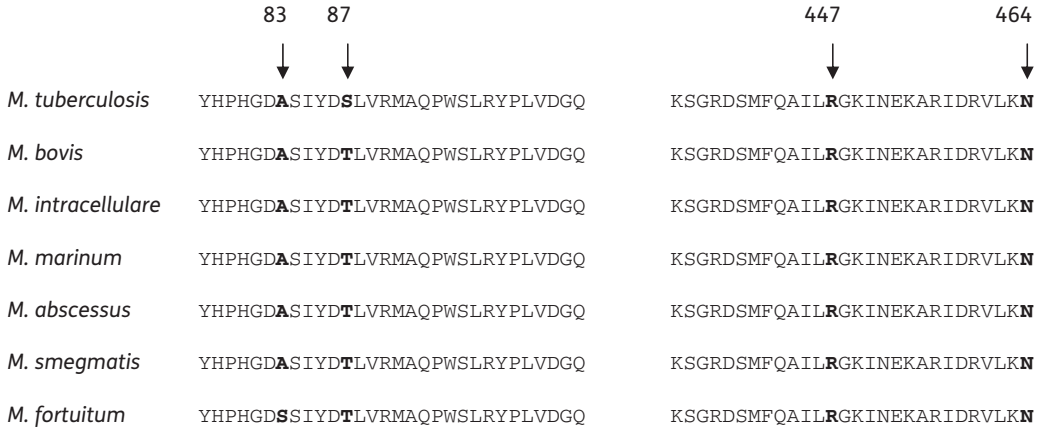

Figure 2. Alignment of the peptide sequences of the QRDRs of GyrA and GyrB from mycobacterial species. The GyrA QRDR extends from amino acid residues 77 to 106, and the GyrB QRDR extends from amino acid residues 436 to 464, in the numbering system used for *E. coli*.

mobile genetic elements such as plasmid and transposons, although some genetic transfer within mycobacterial species cannot be entirely excluded.[62] Spontaneous mutations affecting the key targets of antibiotics are frequently associated with drug resistance in mycobacterial species, but resistance may result from alteration in the function of more than one gene.[63] Alteration in chromosomal gene function represents the main mechanism of acquired resistance in clinical strains of mycobacteria species; however, other mechanisms can be involved because several reports have indicated the absence of mutations in drug target genes.[39,64,65]

Aminoglycoside resistance

The 2-deoxystreptamine aminoglycosides (kanamycin, amikacin, gentamicin and tobramycin) are important drugs for the treatment of multidrug-resistant *M. tuberculosis* and NTM infection.[66] This class of antibiotic targets the 16S rRNA in the rRNA operon, thereby inhibiting protein synthesis by interfering with the proofreading process, causing errors in synthesis with premature termination.[67] *M. abscessus* possesses one copy of the rRNA operon, making the likelihood of phenotypic expression of a single mutation more likely. Prammananan *et al.*[68] reported that a spontaneous single mutation affecting the 16S rRNA of clinical isolates of *M. abscessus* was associated with resistance to 2-deoxystreptamine aminoglycosides. They showed that adenine substituted by guanine at position 1408 (A1408G) (*E. coli* numbering) within the 16S rRNA is responsible for the high level of resistance of *M. abscessus* clinical isolates to kanamycin, amikacin and tobramycin (MICs >1000 mg/L). The same mutation conferred resistance to *in vitro* isolates of *M. abscessus* to 2-deoxystreptamine aminoglycosides.[68] Recently we reported the presence of four mutations affecting the 16S rRNA (T1406A, A1408G, C1409T and G1491T) (*E. coli* numbering) that conferred high-level resistance to kanamycin, amikacin (for A1408G, C1409T and G1491T) and gentamicin (Figure 3). Other researchers have associated these mutations with aminoglycoside resistance in different mycobacterial species and in other microorganisms.[69–73]

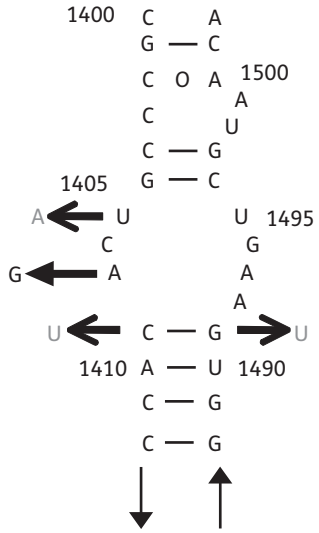

Figure 3. Secondary structure model of *E. coli* 16S rRNA showing different mutations that confer resistance to kanamycin and other 2-deoxystreptamine aminoglycosides.

Macrolide resistance

Macrolides represent another class of antibiotics that target the rRNA operon, preventing peptidyltransferase from adding the peptidyl group attached to tRNA to the next amino acid and inhibiting ribosomal translocation.[74] Macrolide drugs (mainly azithromycin, clarithromycin, erythromycin and roxithromycin) are used for the treatment of NTM infections, including *M. abscessus*, *M. avium*, *M. intracellulare* and *M. chelonae*.[75–77] Bacterial resistance to macrolides occurs by post-transcriptional methylation of the 23S bacterial ribosomal RNA, thereby inhibiting drug attachment. This acquired resistance results in cross-resistance to macrolides, lincosamides and streptogramins.[76] *M. abscessus* infections tend to respond poorly to macrolide-based chemotherapy due to inducible and acquired resistance mechanisms.[37] The involvement of an inducible ribosome methylase *erm(41)* gene conferring high-level resistance in clinical isolates of *M. abscessus* to clarithromycin

Table 3. Acquired resistance described in *M. abscessus*

Antibiotics	Gene involved	Mutations	Protein	References
aminoglycosides	*rrs*	A1408G, T1406A, C1409T, G1491T	16S RNA	98
macrolides	*rrl*	A2058G, A2058C, A2058T, A2059T, A2059C, A2059G	23S RNA	17, 82
fluoroquinolones	*gyrA*	Ala90Val	gyrase A subunit	87

(MICs >32 mg/L) and other macrolides has been reported.[37] Resistance to macrolides acquired by mutation in the *rrl* gene encoding the 23S rRNA generally occurs in mycobacterial species, although it does not occur with any recognizable incidence in other bacterial species.[78–81] Wallace *et al.*[82] studied a group of 800 patients infected by *M. abscessus* that had either disseminated disease or chronic lung disease and found 18 patients (2.3%) were infected with clarithromycin-resistant organisms (MICs >4 mg/L). The resistant isolates were recovered after clarithromycin monotherapy, and sequencing of the gene encoding the 23S rRNA peptidyltransferase region revealed the presence of a point mutation involving adenine at position 2058 (38%) and 2059 (62%) (Table 3).[82]

M. abscessus group: *M. abscessus* (sensu stricto), *M. massiliense* and *M. bolletii*

M. abscessus, *M. bolletii* and *M. massiliense* are closely related species currently identified by the sequencing of the *rpoB* gene and other housekeeping genes.[83] There are few reports on the pathogenic traits of *M. bolletii* and *M. massiliense*, although they have a broad drug resistance profile similar to *M. abscessus*.[84–86] However, differences have been reported in the susceptibility patterns of the three species.[17,18] For example, *M. massiliense* was reported to be susceptible to doxycycline, whereas *M. abscessus* and *M. bolletii* were resistant, although this difference is debatable. Also, these species differ with respect to specific *erm(41)* features and intrinsic clarithromycin susceptibility patterns. *M. massiliense*, which harbours a truncated *erm(41)* gene, is intrinsically susceptible to clarithromycin, whereas *M. abscessus sensu stricto* contains a complete *erm(41)* gene.[18] Strains identified as *M. abscessus* with a C28 polymorphism are associated with clarithromycin susceptibility, whereas a T28 polymorphism is associated with clarithromycin resistance. *M. bolletii*, which contains the T28 polymorphic *erm(41)* gene, was shown to be clarithromycin resistant.[17]

Recently Monego *et al.*[87] investigated *M. massiliense* clinical isolates for their susceptibility to ciprofloxacin. They reported high resistance to ciprofloxacin, mediated by a mutation at codon 90 within the *gyrA* gene.[87]

Concluding remarks

M. abscessus has acquired the reputation of being the most virulent and chemotherapy-resistant member of the RGM group.[88] This notoriety has drawn the attention of several research groups to study this organism in order to decipher its secrets. The development of genetic methods to study *M. abscessus* represent a major breakthrough in this regard, along with the availability of the *M. abscessus* genome, which opens new perspectives in the analysis of the pathogenesis and evolution of this organism. Gene conservation between *M. abscessus* and other mycobacterial pathogens is high, so it is likely that discoveries associated with antibiotic resistance in this RGM will facilitate our understanding of the mechanisms responsible for treatment failure in other mycobacterial species, such as the highly feared SGM pathogens of the *M. tuberculosis* complex. Treatment of infections due to *M. abscessus* complex may benefit from molecular identification within the complex since *M. massiliense* appears more susceptible than *M. abscessus sensu stricto* and *M. bolletii*.[86] If susceptibility testing shows sensitivity after prolonged incubation (14 days), this may predict a favourable outcome with a combination therapy of clarithromycin and amikacin, possibly combined with cefoxitin or moxifloxacin.[58,86] Therapeutic studies on infections involving strains that have been precisely identified and tested against these latter drugs, and also new anti-tuberculous drugs such as tigecycline and TMC-207, are necessary to improve treatment outcomes.

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Transparency declarations

None to declare.

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