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Mycobacterium abscessus disease in lung transplant recipients: Diagnosis and management

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ABSTRACT

Mycobacterium abscessus complex (*MAbsC*) disease in lung transplant recipients is increasingly being recognized as an important cause of graft function decline and suboptimal outcomes. Lung transplant recipients appear to be at the highest risk of *MAbsC* among solid organ transplant recipients, as they have more intense immunosuppression, and the organisms preferentially inhabit the lungs. *MAbsC* is the most resistant species of rapidly growing mycobacteria and difficult to treat, causing considerable mortality and morbidity in immunocompetent and immunosuppressed patients. Herein we describe the risk factors, epidemiology, clinical features, diagnostics, and treatment strategies of *MAbsC* in lung transplant candidates and recipients.

1. Introduction

Mycobacterial infections remain a significant cause of morbidity and mortality in lung transplant recipients [1]. Therefore, careful scrutiny of potential transplant recipients is necessary. “Chronic infection with highly virulent and/or resistant microbes” is considered an absolute contraindication to lung transplant, while colonization with the same is considered a relative contraindication [2]. One such infection that remains controversial is *Mycobacterium abscessus* complex (*MAbsC*) pulmonary infections. This group of non-tuberculous mycobacteria (NTM) infections includes *M. abscessus subsp. abscessus* (MAA), *M abscessus subsp. bolletii* (MAB) and *M. abscessus subsp. massiliense* (MAM) [3,4].

Immunocompetent individuals without structural lung disease usually do not develop NTM pulmonary disease [5,6]. However, patients with immunosuppressive conditions – such as solid organ transplant recipients – are at the highest risk for pulmonary and disseminated NTM disease. NTM that frequently cause pulmonary disease in lung transplant recipients include *Mycobacterium avium* complex (MAC) followed by *MAbsC* [7–9]. In addition, some experts consider that NTM disease is an under-recognized cause of graft dysfunction in lung transplant recipients [5,10].

MAbsC are part of the rapidly growing mycobacteria (RGM) associated with significant pathogenicity, and they account for 65–80% of RGM isolates in the United States [11,12]. MAA disease is increasingly recognized as the most virulent of the NTM infections [13]. MAA pulmonary disease is associated with severe infection and rapid decline in lung function when compared to other NTM pulmonary infections [9]. MAA is among the most antibiotic-resistant NTM species and, thus, most difficult to treat and cure, and unfortunately, it is associated with considerable morbidity and mortality in the lung transplant recipients [14].

2. Epidemiology

NTM are ubiquitously found in the environment, including in soil, dust, natural water sources and municipal water supplies [11]. NTM also colonize the skin, respiratory and gastrointestinal secretions in humans [15]. The number of NTM species recognized today has increased to more than 140 species [11,16,17]. Improved mycobacterial laboratory methodology using molecular laboratory techniques has resulted in identification that is more accurate and increased recognition of NTM lung disease [3,11]. *MAbsC* and other RGM colonize organic surfaces and produce growth in solid media within seven days [12,18]. RGM are able to survive in treated water sources like municipal tap water as they are resistant to many commonly used disinfectants including chlorine, glutaraldehyde, chloramine, ozone and organomercurials [12]. Human disease is mostly acquired from

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environmental exposure [11] However, patient-to-patient and fomite-related transmission of *MAbsC*, including (MAM) have been reported, highlighting the ability of these organisms to cause outbreaks and to disseminate globally [19,20].

The exact incidence of *MAbsC* pulmonary disease in lung transplant recipients is not known as reporting of NTM infections is not mandatory, unlike TB. Incidence and prevalence of *MAbsC* disease appears to be increasing [12]. The increased prevalence could be related to an increase in the frequency of transplants, more potent immunosuppressive regimens, improved diagnostic procedures and increased vigilance and surveillance for these organisms [6,15,21]. Lung transplant recipients are at highest risk among the solid organ transplant recipients for NTM infections as the organisms preferentially inhabit the lungs and lung transplant recipients have more intense immunosuppression [21,22]. In one study, cumulative incidence for *MAbsC* disease was thought to be somewhere around 0.33% [5]. *MAbsC* disease in lung transplant recipients are more frequently reported as allograft infection (pleuroparenchymal disease) followed by skin, soft tissue and disseminated disease [5,22]. The majority of *MAbsC* disease in lung transplant recipients occurs in the first 8 months after transplantation. This might be due higher intensity immunosuppression used during this period [22]. *MAbsC* infections are reported more frequently from southern coastal states that include Florida and Texas which also appear to be the major endemic areas. [11,12,23].

3. Risk factors

The pathophysiology of NTM diseases in otherwise healthy subjects appears to be related to a number of factors including the pathogenicity of the NTM species, environmental exposures and complex host-genetic factors associated with ciliary function, immune function, connective tissue pathways, and the transmembrane conductance regulator (CFTR) [17,24]. Other risk factors associated with the development of NTM pulmonary disease in immunocompetent patients include structural lung disease and upper gastrointestinal tract diseases. Similar to other NTM infections, *MAbsC* lung disease is more common in patients with chronic obstructive pulmonary disease and emphysema, bronchiectasis, cystic fibrosis (CF), alpha-1 anti-trypsin deficiency, pneumoconiosis, sarcoidosis and pulmonary tuberculosis [5,11,14,23]. Moreover, there is very little information regarding the nature of the host immune response to *MAbsC* infections from animal models and human studies [25]. Reduced Th1 and Th2 cell immune responses associated with upregulated Th17 response have been reported in patients with *MAbsC* pulmonary disease compared to healthy controls [26]. In a small study of CF patients, an increased frequency of antigen-specific peripheral blood T cells expressing CD40L(+) but not IL-2(−) was found mostly in patients with *MAbsC* pulmonary disease [27]. This limited immune profiling information suggests that *MAbsC* infections are associated with alterations in some specific pathways of cell-mediated immunity that may contribute to disease progression. In addition, esophageal dysmotility and gastroesophageal reflux are considered important risk factors for the development of *MAbsC* lung disease [11,23]. Some experts believe that true colonization with *MAbsC* does not exist; patients who have minimal clinical features have minimal disease [14]. In pulmonary transplant recipients, the potential risk factors for the development of *MAbsC* disease include immunosuppression and the development of structural lung disease over time due to chronic lung allograft dysfunction [7,22].

Routine screening for NTM infection is recommended in all patients with CF due to the high prevalence of NTM pulmonary disease in this population [12]. Pre-transplant isolation of *MAbsC* in CF patients is a major risk factor for *MAbsC*-related cervical lymphadenitis, wound infection, and mediastinal abscess, which can progress to disseminated disease after organ transplantation (Table 1) [5,9,13,14,19,28,29,30]. The candidacy of CF patients who have history of *MAbsC* disease should be very carefully evaluated in experienced centers as some would consider isolation of *MAbsC* as a relative contraindication to lung transplantation [2,28].

4. Microbiology

RGM are acid-fast bacilli (AFB) and stain as gram-positive beaded rods on gram stain. Fluorochrome (auramine or auramine-rhodamine) is the preferred method of acid-fast staining. Broth and solid media are recommended for *MAbsC* culture. Rapid identification of *MAbsC* subspecies can be performed by molecular methods [12].

Table 1
A summary of reported cystic fibrosis/bronchiectasis patients with pre-transplant isolation of *M.* abscessus complex receiving bilateral lung transplants.

Author	Diagnosis	Age/sex	N	Site	Recurrence (days post)	Site of recurrence	Outcome	Cause of death
Aitken et al. [19]	CF	22/M	1	Sputum positive	74 days		Died	Overwhelming M abscessus infection
Chernenko et al. [5]	CF,	29/M	2	Lung	1 month	Lung	Died 2 months	Sepsis from pseudomonas and aspergillus.
	Bronchiectasis	57/F			5 months	breasts	Died 14 months	Sepsis from C diff colitis
Taylor et al. [28]	CF	21/F	1	Lung	19 months	Right breast	Died	Disseminated M abscessus
Gilljam et al. [29]	CF	10/M, 28/F, 26/F	3	Lung	Few weeks Few weeks Few weeks	Surgical incision, Osteomyelitis of sternum, disseminated M abscessus	All 3 alive after prolonged abx course	
Zaidi et al. [13]	CF	17/M	1	Lung	50 days	Sternal wound infection	Died 87 days after tx	
Qvist et al [30]	CF	39/M	6	Lung	1 year	BAL positive	Alive	
		22/M				Substernal abscess	Alive	
		30/M				Deep tissue infection	Alive	
		22/F					Died 19 days	ARDS
		26/F					Died 3years	CLAD
		29/M					Died 2 months	Invasive aspergillus
Lobo et al. [9]	CF	22/F	3	Lung	6 months	Mediastinal abscess	Died	NTM sepsis
		32/M			2 months	Empyema, sternal OM	Died	BOS
		19/F				Empyema, sternal OM	Alive	

All the patients had pre-transplant *MAbsC* disease. Most common listing diagnosis was CF. These patients had recurrence after transplantation. Most common site for recurrence was sternum, mediastinum and soft tissue infections. N = number of cases; CF = cystic fibrosis; F = female; M = male; OM = osteomyelitis; ARDS = adult respiratory distress syndrome; CLAD = chronic lung allograft dysfunction. BOS = bronchiolitis obliterans syndrome.

4.1. Erythromycin methylase gene (erm gene)

Macrolides bind to 50s ribosomal subunit and inhibit peptide synthesis. Erythromycin methylase *(erm)* genes prevent the binding of macrolides to ribosomes and diminish the inhibitory activity of these agents. The *erm* gene is the primary mechanism of innate and clinically significant macrolide resistance in *MAbsC*, [3]. When MAA or MAB isolates are exposed to macrolide, the *erm* gene activity is induced with subsequent *in vivo* macrolide resistance. This may not be apparent by the initial *in vitro* MIC of the organism for the macrolide. This might be one of the mechanisms for the discrepancy between *in vitro* susceptibility results and *in vivo* response. In order to unmask this inducible macrolide resistance, the NTM isolate should be incubated with a macrolide for up to 14 days to induce this *erm* gene. Standard incubation for 3 days used to determine minimal inhibitory concentration (MIC) is not enough to induce *erm* gene resistance. As a result, the isolate might falsely show *in vitro* susceptibility to macrolide if it is not incubated for 14 days [3,16,31]. DNA sequencing has demonstrated that MAM have a large deletion in *erm* gene, which is associated with a nonfunctional gene and macrolide susceptible phenotype for these isolates. On the other hand, MAA and MAB lack this deletion and the *erm* gene is functional causing macrolide resistance. [3]. Hence, MAM is susceptible to macrolides and have better treatment outcomes at eradication compared to MAA where 80% of isolates are macrolide resistant and are very difficult to treat and eradicate [18,32,33] (Table 2).

Table 2
Mycobacterium abscessus subspecies and their macrolide resistance patterns.

Name	***erm*** gene functional	
M. abscessus subsp. abscessus (MAA)	Yes	Macrolide resistance
M. abscessus subsp. bolletii (MAB)	Yes	Macrolide resistance
M. abscessus subsp. massiliense (MAM)	No	Macrolide susceptible

Reference [3].

5. Pretransplant screenings and management

The transplant community lacks consensus in how to treat *MAbsC* isolation in a potential lung transplant candidate. Some argue *MAbsC* isolation prior to lung transplantation is a relative contraindication to lung transplantation [28]. However, this is controversial because local control and infection clearance is possible in recurrent cases [9,34]. Moreover, CF patients with pre-existing *MAbsC* pulmonary disease have successfully achieved expected survival rates but they are at risk of developing difficult-to-treat soft-tissue infections [30]. *MAbsc* infection is an absolute contraindication to lung transplant in some transplant centers due to concerns for increased morbidity, mortality, and risk of person-to-person spread after transplantation. There is consensus that all lung transplant candidates with structural lung disease (e.g. bronchiectasis) and risk factors for NTM should be screened for mycobacteria and NTM disease. This will identify more patients with pretransplant *MAbsc* disease. Type of screening will be dependent on the severity of underlying lung disease. For patients on high-flow oxygen supplementation, bronchoalveolar lavage may not be feasible, hence sputum smear testing for AFB stain and mycobacteria cultures should be performed [8].

There are no standardized guidelines for diagnosis and management in the pre-transplant evaluation. In order to reduce transplant risks, experts recommend eradication of NTM infection should be attempted prior to lung transplantation, and all foreign bodies should be removed at the time of lung transplantation which include indwelling lines and even some suggest the removal of breast implants [28,30,34]. Patients should be adequately treated with multidrug regimens. Pre-transplant patients who have three sputum samples negative for AFB stain and mycobacteria cultures for more than one year are considered cleared and could be listed [9]. However, in the case series described by Lobo *et al* only 3 of the 13 CF patients with *MAbsC* disease pre- transplant were able to clear the infection despite aggressive therapy (mean duration of multidrug treatment was 10 months), demonstrating the difficulty of clearing *MAbsC* disease.

In situations where listing cannot be delayed, the multidrug regimen could be continued in the post-transplant phase for 6 weeks or more while the surveillance for *MAbsC* isolation should continue with BAL, sputum and wound cultures [9]. This is particularly important since those lung transplant recipients are a high risk to develop *MAbsC* wound infection, sternal abscess, skin, soft tissue and disseminated disease. [9,28,29]. Moreover, AFB stains and mycobacteria cultures should be obtained from the donor bronchial washings, bronchoalveolar lavage if feasible and intraoperative samplings should be encouraged in individuals at risk.

During lung transplantation surgery, removal of the diseased lungs probably serves as a means of decreasing “disease burden”. The surgery in addition to aggressive antimicrobial treatment can help achieve disease eradication [9]. Moreover, the native lung in single lung transplant recipients could act as a potential source of infection that could infect the allograft [8]. Hence, patients with a history of pretransplantation *MAbsC* disease should be considered for bilateral lung transplantation to decrease disease burden and prevent reinfection of the graft.

Following transplantation, repeated surface cleaning of clinic and equipment and isolation procedures should occur between patients to avoid transmission of *MAbsC*. Transplant centers should also consider placing patients with *MAbsC* disease in negative pressure rooms and contact isolation during clinic visits as patient-to-patient and fomite-related transmission of *MAbsC*, including (MAM) have been reported [19,20].

6. Immunosuppression in lung transplantation

It has to be recognized that the immunosuppression required following lung transplantation is much higher than that of any other solid organ transplantation [35]. This might relate to endogenous factors (large number of donor-derived dendritic cells capable of stimulating T cells) and exposure to environmental antigens which might be responsible for triggering frequent rejections [35]. Immunosuppression is used to reduce both acute and chronic rejection in lung transplant recipients. This higher level of immunosuppression contributes to higher incidence of opportunistic infections, including *MAbsC*, in lung transplant recipients when compared to other solid organ transplantation.

7. Clinical features of *M Abscessus* complex disease in lung transplant recipients

Disease in immunosuppressed lung transplant recipients ranges from pulmonary disease, lymph node involvement, localized cutaneous lesions, thoracotomy wound infection, sternal osteomyelitis, empyema to disseminated infections [6,7,21]. Most common initial presentation is pulmonary disease followed by cutaneous lesions [5,21,36].

7.1. Pulmonary disease

Patients with pulmonary disease could present with cough, fever, chills, chest pain, dyspnea, expectoration, and/or hemoptysis. Others may present with non-specific symptoms like fatigue, malaise and weight loss. Some patients are asymptomatic. Nearly half of the patients present with decline in lung function testing [5,12]. Empyema has also been described in some patients with *MAbsC* infection after lung transplantation [37].

7.2. Skin and soft tissue disease

Cutaneous disease due to *MAbsC* could be from (1) surgical wound infection or (2) may be the initial presentation of disseminated disease [7,11,21]. Therefore, skin lesions have to be followed very closely [21]. Lesions have been described in upper or lower extremities, buttocks, breast and very rarely in multiple sites. Cutaneous lesions could occur before or after the allograft infection [5]. Lesions typically present as painful erythematous or violaceous nodules that may form abscesses. Some of these abscesses may open up to form ulcers [5]. A skin biopsy or aspirate for histopathologic examination is required to make a definitive diagnosis.

MAbsC is the most common NTM that causes surgical site infection in solid organ transplant recipients [22]. This is a very challenging complication to treat. Interestingly this has been seen more commonly in cystic fibrosis patients who were colonized with *MAbsC* prior to transplantation [9,13,29,30].

Disseminated disease presents as multiple draining cutaneous nodules or abscesses. There is no obvious portal of entry. Patients can also present with chronic cervical lymphadenopathy. The organism can be isolated in blood culture, tissue biopsy or aspirated material [12] (Fig. 1 and Fig. 2).

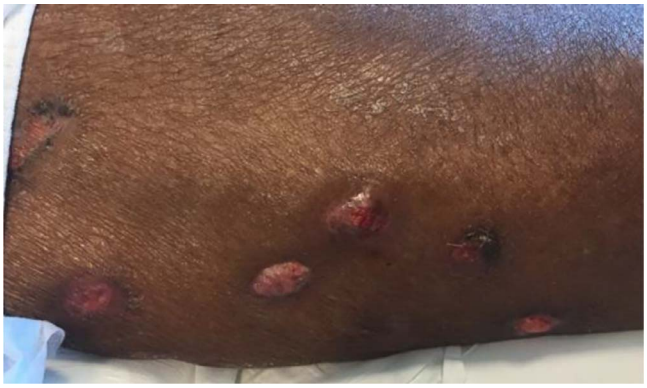

Fig. 1. Skin lesions, right lower extremity in a patient with disseminated *M abscessus* subsp. *bolletii* (MAB).

8. Radiological findings

The majority of patients present with pulmonary nodules, fibrocavitary disease, nodular bronchiectasis, or combination of those features. Common findings in chest high-resolution computed tomography (HRCT) include multifocal bronchiectasis with multiple small nodules and/or cavitary lesions [5,7]. Other findings in chest radiograph (CXR) and chest HRCT include focal consolidation, patchy multinodular opacities, as well as reticulonodular, interstitial and alveolar infiltrates with upper lobe predominance [12]. The pulmonary nodules are visualized on CXR in only 50% of the cases. Hence a HRCT of the chest should be performed if there is a high index of suspicion [5] (Fig. 3).

9. Diagnosis

The diagnosis of NTM lung disease should include presence of compatible symptoms, microbiological and radiological evidence as recommended by the ATS and IDSA guidelines for the diagnosis and treatment of NTM (Table 3) [11]. All three criteria must be met before the diagnosis of NTM lung disease is made. The evaluation of all patients suspected of NTM lung disease should include three or more sputum specimens obtained early in the morning and on three separate days for AFB analysis along with a CXR or chest HRCT (if no cavitation present on CXR) and exclusion of other pulmonary disorders like TB or malignancy. When performing expectorated sputum samples, patients should not drink or rinse their mouth with tap water prior to collection to minimize the risk of environmental contamination. Sputum can be induced with hypertonic saline nebulization if patient is unable to produce sputum. If unable to obtain sputum, bronchoalveolar lavage (BAL) with or without biopsy can be performed. [11,12]

As NTM are ubiquitous organisms, contamination of the specimens can occur during collection, transport and processing of the sample. Hence, a single positive expectorated culture is not adequate to make a definitive diagnosis of NTM lung disease. On the other hand, specimens obtained from bronchoalveolar lavage have less likelihood of environmental contamination; hence, a single positive culture obtained from BAL is considered adequate for microbiological diagnosis [11,12]. In lung transplant recipients diagnosis is established by bronchoalveolar lavage (BAL) in the majority and rarely by biopsy [5].

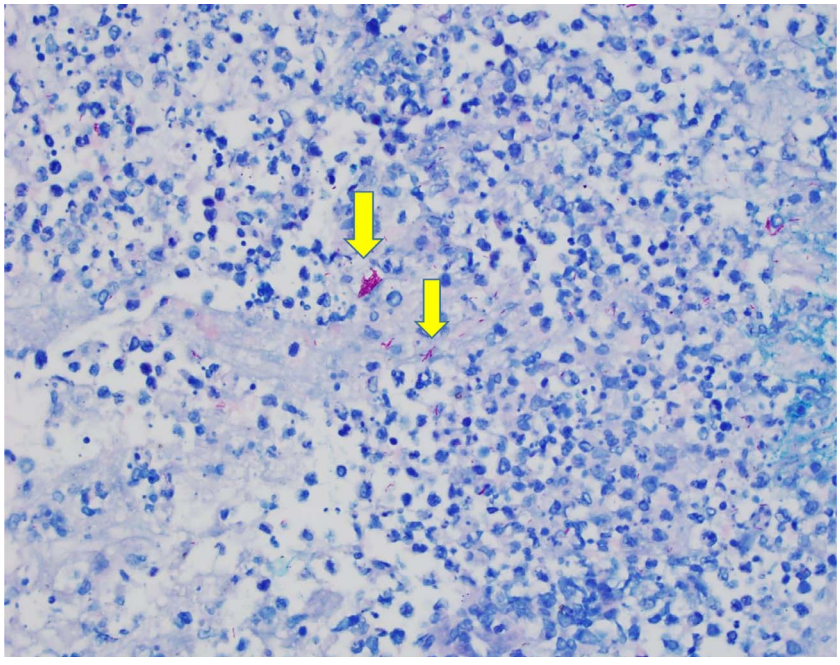

Fig. 2. Ziehl–Neelsen stain (acid fast stain) of the biopsy reveals numerous rod shaped acid fast bacilli (arrows, 400X).

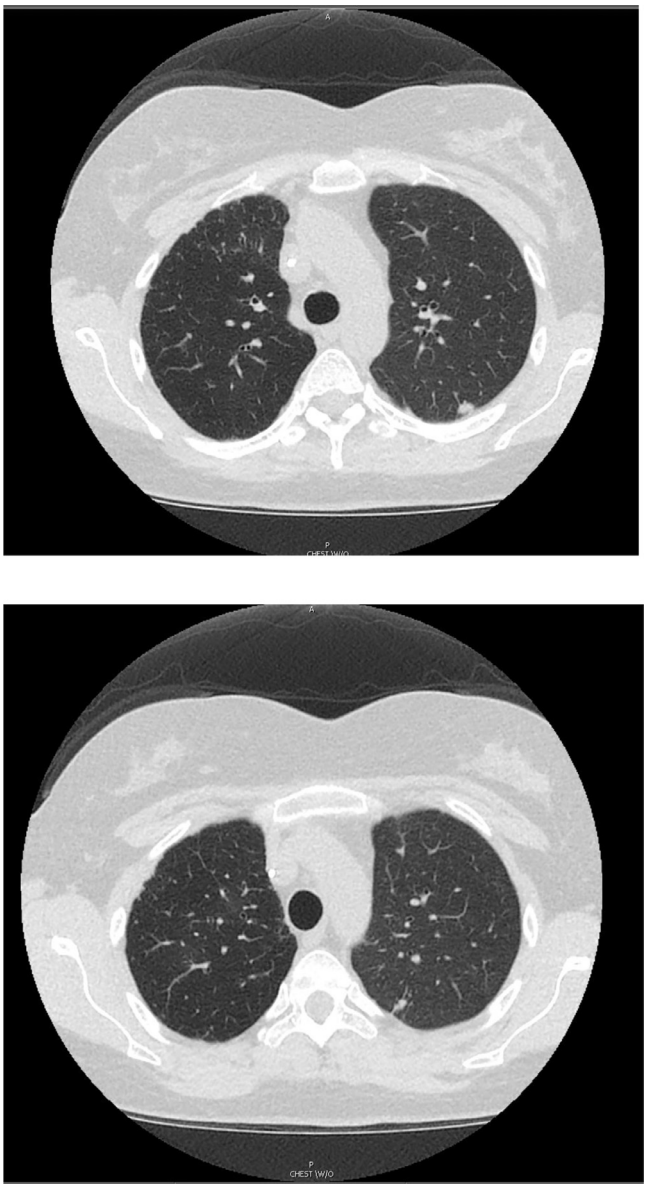

Fig. 3. Multiple left upper lobe nodules 5 years after lung transplantation in a patient with *MAbsC* disease.

Table 3
ATS/IDSA diagnostic criteria for NTM pulmonary disease.

1. Consistent clinical features along with
2. Radiological findings compatible with NTM along with
3. One of the following microbiological findings:
   a) Two or more positive sputum cultures from different samples
   b) One positive culture from bronchoalveolar lavage
   c) Positive culture from lung biopsy or biopsy with mycobacterial features

Diagnostic criteria for NTM pulmonary disease as set forth by the 2007 ATS/IDSA guidelines (Table 3). All three criteria have to satisfy for diagnosis of NTM lung disease [11].

The presence of NTM in one bronchoalveolar lavage sample or two separate sputum samples in conjunction with compatible radiographic or CT imaging and clinical symptoms satisfies the ATS diagnostic criteria and constitutes NTM disease [11]. Moreover, approximately 20% of patients with *MAbsC* infection will also develop infection or disease due to *M. avium* complex. [14]. Importantly, one has to be very careful in extrapolating the ATS/IDSA criteria to immunosuppressed lung transplant recipients, as these criteria have not been validated in lung transplant recipients [38].

Table 4
Most frequent adverse effects of the antimicrobials used to treat *M. abscessus* complex infections.

Antimicrobial agent	Adverse reaction	Monitoring
Aminoglycosides	Nephrotoxicity especially when concurrently used with calcineurin inhibitors.	Trough levels, serum creatinine, urinalysis
	Hearing loss, ototoxicity, vestibular toxicity, tinnitus	Audiometry
Cefoxitin	Neutropenia, thrombocytopenia,	CBC weekly
	AST, ALT elevation, Rash	Weekly Liver enzymes
Imipenem	Neutropenia	CBC weekly
Linezolid	Bone marrow suppression,	CBC weekly,
	Optic neuropathy, Peripheral neuropathy	Ophthalmology for optic neuropathy
Tigecycline	Nausea, vomiting, diarrhea	
Azithromycin, Clarithromycin	Nausea, diarrhea, Liver toxicity, Ototoxicity, QT interval prolongation	Liver function test, Audiometry, EKG
Bedaquiline	Nausea, arthralgia, QT interval prolongation	EKG
Clofazimine	Nausea, vomiting, diarrhea, Red-brown skin discoloration, QT interval prolongation	EKG

10. Treatment

Once the diagnosis of *MAbsC* pulmonary disease is established, the next step would be to decide if the patient's clinical condition warrants treatment and what antimicrobials to treat the patient with. Treatment decisions are made on a case-by-case basis and this would take into consideration the presence of other co-morbid conditions and the presence of other organ dysfunction. One has to decide if the treatment goal is curative, chronic suppressive therapy or palliative therapy [39]. Other considerations include the risks and benefits of therapy, the tolerability of treatment, the toxicities associated with the multi-drug regimen (Table 4). Drug interactions, especially with immunosuppressive medications should also be taken into consideration (See Table 5). If the patient is symptomatic and there are signs of clinical progression or a decline in graft function one should elect to treat. If possible the intensity of immunosuppression should be decreased. This might be difficult in the first year after transplantation. Coexisting viral infections like CMV infections should be treated [6]. In fact, some have suggested that the development of disease after lung transplantation could be an indicator for excessive immunosuppression since the great majority of the patients who die after *MAbsC* disease, succumb to other causes [8]. In patients with pre-transplant *MAbsC* disease, aggressive induction therapy with antithymocyte globulin should be avoided and post-transplant calcineurin target trough levels should be lowered if feasible. However, lowering immunosuppression to attempt to improve immune function to facilitate the control of infection should be balanced against the risk of organ rejection [34]. Every attempt should be to optimize nutritional status of these patients. In patients with impaired quantitative IgG levels, gamma globulin supplementation could be considered [29]. Along these lines, effective treatment for comorbid risk factors for NTM disease, including treatment for GERD and dysphagia can result in improvement of their *MAbsC* lung disease [11]. Patients with co-existing bronchiectasis should also receive optimal chest physiotherapy, including the use of bronchodilators, hypertonic saline nebulizations and the use of flutter valve, postural drainage, and/ or high-frequency chest wall oscillation (*i.e.* Vest) therapy [40–42].

Antimicrobial treatment of *MAbsC* is very demanding and

Table 5
Drug interactions between antimicrobials used to treat *M* abscessus complex infections and immunosuppressive agents.

Antibiotic	Cyclosporine	Tacrolimus	Sirolimus
Azithromycin	Weak inhibition of CYP450 (weaker than Clarithromycin) leading to increase of cyclosporine levels	Weak inhibition of CYP450 leading to increase of tacrolimus levels	No known interaction
Aminoglycosides	Increased risk of nephrotoxicity	Increased risk of nephrotoxicity	No known interaction
Clarithromycin	Inhibition of CYP450 leading to increase of cyclosporine levels	Inhibition of CYP450 leading to increase of tacrolimus levels	Inhibition of CYP450 leading to increase of sirolimus levels
Fluoroquinolones	Weak inhibition of CYP450 leading to increase of cyclosporine levels	Weak inhibition of CYP450 leading to increase of tacrolimus levels	No interaction

Reference [15].

Table 6
Antimicrobial dosage examples and route of administration.

Antibiotic	Route	Dose and frequency
Amikacin	Intravenous	10–15 mg/kg daily, target peak serum levels 20–25 mg/ml range
Clarithromycin	Oral	500 mg twice daily
Azithromycin	Oral	250–500 once mg daily
Cefoxitin	Intravenous	200 mg/kg or 2 to 4 g twice or three times daily with a maximum of 12 g/daily
Imipenem	Intravenous	500 mg to 1 g two to four times daily
Tigecycline	Intravenous	25–50 mg daily
Linezolid	Oral	600 mg twice daily
Clofazimine	Oral	100 mg daily

References [11,32].

challenging for a number of reasons. *MAbsC*, in particular MAA and MAB, are resistant to many antimicrobial agents [4,16,43]. *In vitro* antibiotic susceptibilities correlation with *in vivo* response is supported by limited data [16,18,36]. The Clinical and Laboratory Standards Institute recommends testing the RGM for susceptibility to macrolides, aminoglycosides, cefoxitin, imipenem, tigecycline, fluoroquinolones, linezolid and doxycycline. *In vitro* studies have shown *MAbsC* isolates being sensitive to parenteral amikacin (90%), cefoxitin (70%), imipenem (50%), but these drug susceptibility rates vary depending on the geographic location. [4,5,39]

In recent years, it has become clear that identification of the mycobacteria to the species and subspecies level is very important. If *MAbsC* subspecies determination is not available, it will be very important to know if the *MAbsC* isolate has an active or inducible *erm* gene. The ability of the mycobacterial laboratory to provide subspecies level identification and presence or absence of an inducible *erm* gene will help the treatment plan immensely [16]. It is much more difficult to treat MAA and MAB than to treat MAM. The absence of inducible resistance to macrolides makes MAM more susceptible to these key antimicrobials in the antibiotic regimen [18,33]. Moreover, clarithromycin induction of the *erm* gene is greater than with the use of azithromycin, which explains a higher induction of macrolide resistance with clarithromycin than with azithromycin in MAA but not in MAM [44]. Hence, azithromycin might be more effective against MAA than clarithromycin whereas both macrolides might be equally effective against MAM. In the United States, there is more lung disease caused by MAA than MAB [31]. Many centers use azithromycin as a drug to prevent or slow obstructive CLAD (chronic lung allograft dysfunction), and thus, macrolide susceptibility and *erm* gene phenotypic testing should be obtained if information of *MAbsC* subspecies is not readily available. Moreover, Azithromycin is a less potent cytochrome P 450 (CYP450) inhibitor than clarithromycin, which is an important consideration in order to design an antimicrobial regimen with less likelihood to interact with other patient's drugs [6].

10.1. Treatment of pulmonary disease

The eventual goal of treatment of *MAbsC* lung disease as in other NTM lung diseases is symptomatic improvement, resolution of radiologic findings, improvement in graft function, and achievement of 12 months of negative sputum cultures while on therapy [11,12].

Table 7
Maintenance immunosuppression examples in lung transplantation.

	0–12 months post lung transplantation	12–24 months post lung transplantation	> 24 months post lung transplantation	Adverse effects to monitor
Calcineurin inhibitor	*12 h trough level*	*12 h trough level*	*12 h trough level*	Nephrotoxicity, hypertension, hypercholesterolemia, hyperkalemia, hypomagnesemia, neurotoxicity (tremor, headache, PRES), thrombotic microangiopathy
Tacrolimus or	10–14	8–12	6–12	
Cyclosporine (Neoral)	250–350 ng/ml	200–300 ng/ml	100–200 ng/ml	
Plus				
Antimetabolite	*Dose*			
Azathioprine or	100–150 mg PO qd			Leukopenia, thrombocytopenia, anemia, hepatotoxicity, pancreatitis
Mycophenolate mofetil or	1000 mg PO bid			Cytopenia, GI intolerance
Myfortic	720 mg PO bid			Cytopenia, GI intolerance
Plus				
Steroid	*Dose*	*Dose*	*Dose*	Hyperglycemia, diabetes mellitus, weight gain, hypertension, hyperlipidemia, osteoporosis, cataracts
Prednisone	10–20 mg	5–10 mg	5 mg	
And Or				
mTOR inhibitors	*24 h trough level*	*24 h trough level*	*24 h trough level*	Delayed wound healing, bronchial dehiscence, proteinuria, pneumonitis, hypertriglyceridemia, leukopenia, thrombocytopenia, venous thromboembolism
Sirolimus*	10–15 ng/ml	10–15 ng/ml	5–8 ng/ml	

* *Note.* Early post-transplant use of sirolimus is contraindicated due to reported anastomotic dehiscence.

Monotherapy with macrolides is not recommended because of the risk of emergence of macrolide resistance [15]. The antimicrobial agents with the best *in vitro* activity against *MAbsC* include amikacin, clarithromycin, tigecycline and cefoxitin. Half of *MAbsC* isolates will demonstrate at least intermediate susceptibility to linezolid and imipenem. [12,45].

The ATS/IDSA guidelines recommend combination of intravenous amikacin plus cefoxitin or imipenem and oral macrolide for at least 2–4 months [11]. One small series of 41 non-transplant patients with *MAbsC* treated according to these guidelines demonstrated a treatment success rate of 80.5% at one year with a 10% relapse rate [46]. Another case series of 22 non-transplant patients on various treatment regimens reported less successful outcomes of only 36.4% improvement at one year [47]. However, these recommendations are for non-immunosuppressed host. These recommendations were published before the era of *MAbsC* subspeciation and *erm* gene phenotypic testing. One has to be cautious extrapolating the 2007 ATS/IDSA guidelines to immunosuppressed lung transplant recipients since optimal duration of treatment in this population is not known. Current evidence is limited to a few case series and retrospective studies in which treatment durations were often in the 6–9 months range but treatments of 12–24 months with microbiological response by 3–6 months have been reported [5,7,8,29,48]. Prolonged therapy is needed to achieve microbiologic clearance and radiologic improvement. Moreover, disease recurrence has been reported to occur even in patients who have received prolonged treatment [5]. If possible, reduction in immunosuppression should be considered but this decision should be weighed against the potential risk of organ rejection and allograft dysfunction. If reduction in immunosuppression is not possible or if high disease burden exists, (disseminated disease or smear positive lung disease) prolonged therapy or lifelong suppression should be considered [6].

Aspergillus sp and *Pseudomonas aeruginosa* infections occur frequently after treatment of *MAbsC* disease with prolonged use of multidrug regimen. In one study 70% of patients treated for *MAbsC* disease developed concurrent infections with *Aspergillus, Pseudomonas, Clostridium difficile* or *Stenotrophomonas* [5].

Treatment adherence is a very important factor in the success of the treatment regimen, and this has to be emphasized with appropriate patient education and close monitoring in a multidisciplinary environment. Complete microbiological clearance and/or cure from *MAbsC* lung disease may not be achievable for some patients with the current antimicrobial agents, due to side effects and poor tolerability of medications. Long-term antimicrobials regimens designed with the goal of minimizing adverse effects of the medications while suppressing the infection can be considered [39]. Prolonged course of intravenous antibiotics with or followed by oral macrolide, clofazimine, and/or oral linezolid with inhaled amikacin can be used to suppress the infection. Periodic antibiotics can be considered for some to provide suppressive therapy, but long-term sputum conversion is rare and risk of developing additional drug resistance can increase [12]. In patients with *MAbsc* empyema, in addition to surgical drainage of pleural space, prolonged intravenous multidrug regimen should be considered. This has to be followed by long term oral suppressive therapy [9].

Adjuvant surgical resection with optimal antimicrobial therapy is recommended and associated with the best chance of cure for immunocompetent patients with localized *MAbsC* pulmonary disease [39]. However, a surgical treatment option can be challenging in lung transplant recipients with allograft dysfunction. In selected patients with localized disease and adequate lung function, surgical resection of the diseased portion of the lung following initial treatment with antimicrobial agents to decrease the disease burden can offer the best chance of cure [11,12].

Clofazamine may be a treatment option for macrolide resistant or intolerant regimen. Inhaled amikacin may have some benefits in patients with refractory *MAbsC* disease although its role is still not very clear [36,49]. However, the combination of clofazamine with amikacin or clarithromycin is synergistic *in vitro* [50], therefore and at least in theory, the combined use of inhaled amikacin with clofazimine and a macrolide might enhance the microbiological control of the infection at the airway and cavitary disease levels. Bedaquiline also has been used as salvage therapy in few refractory MAA cases [51].

10.2. Treatment of skin, soft tissue and bone disease

Skin and soft tissue infection from *MAbsC* in transplant recipients can be localized or disseminated, which is associated with considerable morbidity. Diagnosis is established by tissue biopsy, cultures and abscess drainage. *In vitro* susceptibility pattern would aid in the choice of the antimicrobials. In non-immunosuppressed host, multidrug regimen that includes a macrolide (for macrolide-susceptible strains), intravenous amikacin plus high-dose cefoxitin or imipenem is recommended by the ATS/IDSA guidelines. For severe disease, four months of combination therapy is recommended. In case of osteomyelitis, the therapy is extended to 6 months, and at least 2 months beyond the resolution of all clinical signs of infection [11,12,39]. However, the successfully cured immunosuppressed lung transplant patients received a combination of antimicrobials including amikacin for 6 months or longer [29]. Surgical drainage of abscesses and debridement of extensive disease should be performed. Surgical treatment has been recommended for cases with drug resistance and/or when drug therapy is challenging [39]. Surgical drainage of the localized cutaneous disease with parenteral antibiotics is also thought to be best treatment strategy for cutaneous infection [12]. Antimicrobial therapy alone for cutaneous infection might not be sufficient due to poor penetration [5]. Some of these infections require several or more surgical drainages [9]. Antibiotic monotherapy might produce resistance hence combination therapy is generally recommended [5]. Chronic suppressive antimicrobial therapy might be required [7]. Topical amikacin cream has been used in skin and soft tissue infections, though the efficacy is not clear [9].

10.3. Toxicities of treatment

Treatment of *MAbsC* in lung transplant recipients can be challenging due drug toxicities and to drug-to-drug interactions of antimicrobials with immunosuppressants and other medications used in these patients [34] (Tables 4 and 5). In fact, a transplant pharmacist assistance can be very helpful to carefully plan and dose antimicrobials and to very closely monitor antirejection drug and antimicrobials levels in these patients (Tables 5–7). Aminoglycosides can increase the nephrotoxicity when administered concomitantly with calcineurin inhibitors [5]. Use of systemic aminoglycosides may cause nephrotoxicity (15%), ototoxicity (37%) and vestibular toxicity (9%). Nephrotoxicity and vestibular toxicity are reversible in majority of the cases. The toxicity is not different when daily dosing was compared to three times weekly dosing. Older age, larger cumulative dose, longer duration of treatment are associated with risk of ototoxicity [52].

11. Survival and outcome

Development of *MAbsC* disease after lung transplantation is associated with allograft dysfunction and higher mortality [8]. About 60% of lung transplant recipients with *MAbsC* disease were cured with combination antimicrobial therapy, while 40% died. *MAbsc* disease was the cause of death in some, but the greater proportion of the patients who died, succumbed to non-NTM infections, including sepsis from *Aspergillus, Pseudomonas,* and *Clostridium difficile colitis* [5,8].

12. Conclusion

MAbsC is a group of ubiquitous organisms that can cause significant morbidity, including chronic pulmonary infections and skin and soft

tissue infections in lung transplant recipients, which can be challenging to treat in this patient population [36]. The overall incidence of NTM seems also to be increasing, and providers involved in the care of lung transplant candidates and recipients should screen patients at risk and have high clinical suspicion for these mycobacterial infections. AFB stains and mycobacteria cultures should be obtained from the donor bronchial washings if feasible and intraoperative samplings should be encouraged in individuals at risk. Lung transplant practitioners should consider routine mycobacterial testing for abnormal pulmonary lesions and during surveillance. BAL testing for AFB smear and culture should be performed as part of post-transplant evaluations for early diagnosis and prompt management. Any suspicious skin lesions should be biopsied and subjected to special staining and microbiological cultures accordingly.

MAbsC disease prior to lung transplantation in a potential recipient is not an absolute contraindication to transplantation at all centers, but there is still controversy since *MAbsC* disease is associated with a significant risk factor for post transplantation skin and soft tissue infections and disseminated disease [29]. Every attempt should be made to eradicate the infection with prolonged multidrug regimen prior to transplantation. [9,30]. Moreover, *MAbsC* disease in an immunosuppressed lung transplant recipient is very challenging to treat and cure. The ATS/IDSA statement and recent expert reviews provide excellent guidelines for treatment of NTM disease in immunocompetent patients but those have not been validated in lung transplant recipients. In fact, antimicrobial treatment for *MAbsC* disease is not only challenging due to mycobacterial drug resistance but also by significant drug-to-drug interactions with immunosuppression medications, and potential toxicities of these agents. In this context, future research directions should not only focus on the development of new antimicrobials for *MAbsC* disease but also in better understanding the nature of host immune response to this emerging infection in order to improve preventive and treatment strategies, and ultimately clinical outcomes in this population.

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Conflict of interest

None of the authors have any conflict of interest.

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