Otitis Media and Otomastoiditis Caused by *Mycobacterium massiliense* (M. abscessus subsp. bolletii)

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We describe two patients with otologic infections caused by *Mycobacterium massiliense* (M. abscessus subsp. bolletii) which were identified using *erm*(41) PCR, 23S rRNA, and rpoB gene sequence analysis. They were middle-aged adults with underlying otologic diseases and were treated successfully with clarithromycin-based combination regimens for 3 and 9 months, respectively.

TABLE 1 Antimicrobial susceptibilities of four isolates of *M. massiliense* (M. abscessus subsp. bolletii) to 15 antimicrobial agents determined by the broth microdilution method

<table>
<thead>
<tr>
<th>Isolate no.</th>
<th>AMC</th>
<th>FOX</th>
<th>CRO</th>
<th>FEP</th>
<th>IPM</th>
<th>CIP</th>
<th>MXF</th>
<th>DOX</th>
<th>MIN</th>
<th>LZD</th>
<th>TOB</th>
<th>AMK</th>
<th>TGC</th>
<th>SXT</th>
<th>CLA</th>
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<tbody>
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<td>&gt;64/32</td>
<td>&gt;64/32</td>
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</table>

Values for days 5 and 14 are shown.

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The *Mycobacterium abscessus* complex comprises three closely related subspecies, namely, *massiliense*, *bolletii*, and *abscessus* (*sensu stricto*) (1). Identification of *M. abscessus* complex members to the species level depends on the sequencing analysis of several genes, including the *erm*(41) gene, the 23S rRNA gene, and several housekeeping genes (e.g., *rpoB* and *hsp65*), which is not possible in many laboratories (1, 7, 9). A previous report also indicated that *erm*(41) PCR can be efficiently used to simply differentiate *M. massiliense* from *M. abscessus* and *M. bolletii* and inconsistency between *rpoB* and *hsp65* sequence analyses could be found (7). Recently, the taxonomic status of *M. massiliense* has come under debate and currently *M. abscessus* subsp. *bolletii* is probably preferred (10). Infections due to *M. massiliense* (*M. abscessus* subsp. *bolletii*) include postsurgical infections, cutaneous infections, pulmonary infections, and central nervous system infections (3, 8, 11, 12, 14). Otitis media and otomastoiditis caused by nontuberculous mycobacteria (NTM) are common in children, and members of the *M. abscessus* complex are the pathogens most frequently isolated from patients with these diseases (5, 17). In a recent study of 10 patients with otomastoiditis, van Ingen et al. reported that the causative pathogen in all of the patients was *M. abscessus* (*sensu stricto*) (17).

From 2000 to 2010, eight patients with otitis media or otomastoiditis whose clinical (biopsy and ear discharge) specimens were positive for *M. abscessus* complex members were treated at the National Taiwan University Hospital. Biopsy or ear discharge specimens from the eight patients were processed and pretreated for mycobacterial cultures as previously described (13). *M. abscessus* complex isolates were identified to the subspecies level by screening for the presence of the *erm*(41) gene, as well as sequencing of the 23S rRNA and *rpoB* (306 bp) genes (7). The MICs of 15 antimicrobial agents for the four *M. massiliense* (*M. abscessus* subsp. *bolletii*) isolates were determined using the Sensititre RAPMYCO panel test (TREK Diagnostic Systems, Magellan Biosciences, West Sussex, United Kingdom). The MICs of all of the agents tested were read on day 5 after incubation, and those of clarithromycin were read after extended incubation (on day 14) (15).

Of the eight *M. abscessus* complex isolates, four were confirmed to be *M. abscessus* (*sensu stricto*) and the other four were confirmed to be *M. massiliense* (*M. abscessus* subsp. *bolletii*) by *erm*(41) PCR (397 bp) and sequence analysis of the 23S rRNA (accession number F1358489.1, 99% similarity) and *rpoB* genes (100% similarity) (7). The four patients with *M. abscessus* (*sensu stricto*) otitis media have been previously described (6).

The MICs of the 15 agents for the four *M. massiliense* (*M. abscessus* subsp. *bolletii*) isolates are shown in Table 1. Amoxicillin-clavulanic acid, cephalosporins, imipenem, tetracyclines, lincomycin, fluoroquinolones, and aminoglycosides were not active
against any of the four isolates. The clarithromycin MICs for the four isolates were 0.12 to 1.0 μg/ml. The clarithromycin MIC for one isolate (a colonizer from patient 3) was >16 μg/ml, and that for the other three isolates was 0.5 μg/ml (read on day 14 of incubation).

The *M. massiliense* (*M. abscessus* subsp. *bolletii*) isolates from two of the four patients (patients 3 and 4) with positive response to clarithromycin were considered contaminants or colonizations instead of infections because of the good response to topical treatment alone and negative acid-fast staining and one single positive culture from multiple specimens (two specimens from each of the two patients) (Table 2). Patient 1 was a 53-year-old male who suffered from chronic otitis media due to *Aspergillus niger* and had received a mastectomy plus a tympanoplasty 1 year ago. Otorrhea, hearing impairment, and vertigo developed 3 months after the surgery. Three cultures from the right ear discharges all grew *M. abscessus*, which was then confirmed to be *M. massiliense* (*M. abscessus* subsp. *bolletii*). The patient received clarithromycin (500 mg twice daily) plus ciprofloxacin (750 mg twice daily) for 9 months. Repeated cultures of samples from the right ear were negative at the end of treatment, and his status remained uneventful for 4 years without any sequelae.

Patient 2 was a 58-year-old male patient with a history of perforation of the tympanic membrane. He presented to the hospital with otorrhea, hearing impairment, and Gradenigo’s syndrome (6, 15). He first received a petrosectomy, a mastoidectomy, and a tympanoplasty, followed by combination therapy with clarithromycin (500 mg twice daily), ciprofloxacin (750 mg twice daily), and ethambutol (800 mg/day) for 3 months. Acid-fast bacilli were found in one ear discharge specimen that was also culture positive for *M. massiliense* (*M. abscessus* subsp. *bolletii*). At the end of treatment, cultures became negative for mycobacteria. The patient experienced the complication of abducens nerve palsy during the 3-year follow-up after the completion of treatment (6).

The incidence of otitis media and otomastoiditis caused by NTM is increasing (17). The *M. abscessus* complex, *M. chelonae*, *M. fortuitum*, and the *M. avium-M. intracellulare* complex have been reported to cause otologic infections (4). In our previous report, members of the *M. abscessus* complex were the most common causes of otitis media, followed by *M. chelonae* (6). In the present study, infection due to *M. massiliense* (*M. abscessus* subsp. *bolletii*) was found in one-third of the six patients with infections caused by members of the *M. abscessus* complex (excluding patients 3 and 4). This finding differs from that reported by van Ingen et al., who found that none of the otologic infections due to the *M. abscessus* complex were caused by *M. massiliense* (*M. abscessus* subsp. *bolletii*) (17).

Common features of otomastoiditis caused by *M. massiliense* (*M. abscessus* subsp. *bolletii*) and *M. abscessus* (sensu stricto) included the presence of underlying otologic disease and the need for prolonged antibiotic combination therapy along with surgical intervention (17). Most of the *M. massiliense* isolates were susceptible to clarithromycin (2, 7). Kim et al. reported that clarithromycin-resistant *M. massiliense* isolates invariably had a point mutation at the adenine, A2058 or A2059, in the peptidyltransferase subunit.

### Table 2

<table>
<thead>
<tr>
<th>Patient no.</th>
<th>Age (yr)</th>
<th>Sex</th>
<th>Ear colonization by the organism</th>
<th>Underlying medical condition</th>
<th>Initial presentation</th>
<th>Surgical interventions</th>
<th>Sequelae</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>53</td>
<td>M</td>
<td>M. massiliense (M. abscessus subsp. bolletii)</td>
<td>None</td>
<td>Otorrhea, hearing impairment, vertigo</td>
<td>Petrosectomy, mastoidectomy, tympanoplasty for chronic otitis media</td>
<td>Left facial palsy</td>
</tr>
<tr>
<td>2</td>
<td>58</td>
<td>M</td>
<td>M. massiliense (M. abscessus subsp. bolletii)</td>
<td>None</td>
<td>Otorrhea, hearing impairment, Gradenigo’s syndrome</td>
<td>Mastoidectomy, tympanoplasty for chronic otitis media</td>
<td>None</td>
</tr>
<tr>
<td>3</td>
<td>45</td>
<td>M</td>
<td>M. abscessus (M. abscessus subsp. bolletii)</td>
<td>None</td>
<td>Otorrhea, hearing impairment</td>
<td>Petrosectomy, mastoidectomy, tympanoplasty for chronic otitis media</td>
<td>None</td>
</tr>
<tr>
<td>4</td>
<td>40</td>
<td>F</td>
<td>M. chelonae (M. chelonae)</td>
<td>None</td>
<td>Otorrhea</td>
<td>Petrosectomy, mastoidectomy, tympanoplasty for chronic otitis media</td>
<td>None</td>
</tr>
<tr>
<td>5</td>
<td>51</td>
<td>F</td>
<td>M. avium (M. avium)</td>
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<td>None</td>
<td>Topical ofloxacin</td>
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</tr>
<tr>
<td>6</td>
<td>59</td>
<td>M</td>
<td>M. fortuitum (M. fortuitum)</td>
<td>None</td>
<td>None</td>
<td>CLI, Ciprofloxacin, clarithromycin, ethambutol (3 mo)</td>
<td>None</td>
</tr>
</tbody>
</table>

Note: CLI = Ciprofloxacin, clarithromycin, ethambutol.
caused by *M. abscessus* (*sensu stricto*) has been reported predominately in children (17) while all of the patients described in this report and in our previous report are adults (6). Although the tigecycline MICs were within the 0.5- to 1-μg/ml range in our study, the clinical efficacy of this agent needs to be investigated. Systemic antibiotics may be warranted if otomastoiditis, osteomyelitis, or central nervous system invasion develops (12, 16).

In conclusion, we discovered two patients with otologic infections due to *M. massiliense* (*M. abscessus* subsp. *bolletii*). *M. massiliense* (*M. abscessus* subsp. *bolletii*) should be considered when treating patients with otomastoiditis or otitis media, and molecular techniques are warranted for identification to the species level.

REFERENCES