

## Challenge MY2208-3

August 2022

### Bronchoalveolar lavage (BAL): *Aspergillus terreus*

#### HISTORY

This challenge was sent as a simulated bronchoalveolar lavage (BAL) sample from a 74 year old patient with chest pain. Laboratories were expected to isolate and identify *Aspergillus terreus*.

#### CMPT QC/QA/Statistics

All Mycology samples are produced at CMPT according to CMPT internal protocols.

*The samples are assessed for homogeneity and stability using in-house quality control methods and random selection of samples before and during production, and post sample delivery. The number of random samples selected is 15% of the total production batch.*

The sample was verified by a reference laboratory. *Aspergillus terreus* was isolated as a pure culture.

The challenge sample lot was confirmed to be homogeneous and stable for 46 days.

*All challenge components have in-house assigned values based on the most clinically appropriate result; the most clinically appropriate result is determined by expert committee evaluation. No further statistical analysis is performed on the results.*

#### SURVEY RESULTS

10/10 (100%) processing labs correctly reported the organism at least to the genus level. 7 of those labs identified the species as *terreus* or *terreus* complex (Table 1).

#### COMMENTS ON RESULTS

The correct answer here was *Aspergillus terreus* or *Aspergillus terreus* species complex. The strain sent to participating laboratories was not known to have multiple colony variants, however, colony variants are known to occur with *Aspergillus terreus*.<sup>1</sup>

Given that the strain was isolated from a BAL sample, it was expected that laboratories would report the organism identification to the species level or refer the organism to a reference laboratory for species level identification. Species-level identification is important because different *Aspergillus* species have different susceptibility profiles. For example, amphotericin B is not recommended for the treatment of *Aspergillus terreus* infections.<sup>2</sup>

#### IDENTIFICATION

*Aspergillus terreus* is a fast-growing mold producing mature colonies in 3 days. Colonies are tan to cinnamon (with sugar) brown, with a velvety or powdery texture. Reverse is yellow to tan.

Microscopically, it presents septate hyphae; conidiophores are smooth and relatively short. Phialides are biserial, with the metula and phialide being equal in length, forming only on the upper half of the vesicle. Conidia are round, smooth, 2 – 2.5µm in diameter.<sup>3</sup>

Round solitary conidia are commonly produced along sides of hyphae submerged in medium.

In contrast to other *Aspergillus* species, *A. terreus* produces round solitary conidia—also called aleurioconidia—along sides of hyphae.<sup>4</sup> These accessory conidia play a potential role in pathogenesis and dissemination.<sup>5,6</sup>

#### Grading

Reporting *Aspergillus terreus* was graded acceptable.

Reporting *Aspergillus* species was graded acceptable.

#### Grading guidelines

**Acceptable:** Report is technically correct and clinically appropriate. *Slide or Culture or Susceptibility is correctly identified.*

**Acceptable\*\*:** Report contains deficiencies, but within the limits of acceptable. *Culture may be identified correctly only to the class, or genus level. Nomenclature errors would not likely lead to errors in clinical judgement or decision making.*

**Unacceptable:** Report contains errors that could result in incorrect interpretation and actions. *Significant error.*

**Ungraded:** Report cannot be fairly assessed.

Table 1. Identification results

Reported	Labs	Grade
<i>Aspergillus terreus</i> +/- complex	7	Acceptable
<i>Aspergillus</i> species, refer	1	Acceptable
<i>Aspergillus</i> species possibly two strains, refer	1	Acceptable
<i>Aspergillus</i> species	1	Acceptable**
Sample not normally processed	1	Ungraded
<b>Total</b>	<b>11</b>	

## CLINICAL RELEVANCE

*Aspergillus* species are widespread in the environment found in a wide variety of habitats, such as soil, compost, and dust.<sup>3,7</sup>

*Aspergillus* species cause a group of diseases known as aspergillosis; the form the disease takes usually depends on the underlying immune status of the infected individual. The main syndromes include allergic bronchial pulmonary aspergillosis, chronic necrotizing *Aspergillus* pneumonia, aspergilloma, and invasive diseases.<sup>3,6</sup>

The primary means of acquisition of *Aspergillus* is the inhalation of airborne conidia, which are released from environmental sources and contaminated food.<sup>6</sup>

*Aspergillus terreus* is an emerging opportunistic fungus whose clinical incidence has increased in recent years.<sup>8</sup>

*A. terreus* has been recognized as a cause of invasive aspergillosis, allergic bronchopulmonary aspergillosis, chronic aspergillosis, chronic obstructive pulmonary disease exacerbation, aspergilloma, otitis externa, keratitis, and others.<sup>6,9-11</sup>

*A. terreus* accounts for nearly 4% of all cases of invasive aspergillosis<sup>12</sup> and of special concern is the high mortality by this species and the fact that most representatives are amphotericin B (AmB) resistant.<sup>8</sup>

Culture and microscopy in sputum or bronchoalveolar lavage fluid in combination with antigen- and possibly molecular-based assays are used to diagnose aspergillosis.<sup>6</sup>

The diagnosis of aspergillosis by blood culture is rare as the *Aspergillus* hyphal mass that develops in the lumen during angioinvasion remains in place until the force of blood flow causes hyphal breakage, allowing the mass to circulate. The likelihood that a blood culture would capture this event is rare.

However, as *A. terreus* have the ability to discharge a steady series of unicellular spores (adventitious sporulation) into the bloodstream, a blood culture positive for *A. terreus* should not be ignored.<sup>5</sup>

## ANTIFUNGAL SUSCEPTIBILITY

While CLSI has published epidemiologic cutoff values (ECVs) for *Aspergillus terreus* (CLSI M57S, 4<sup>th</sup> Ed., 2022), there are no interpretive criteria published for this organism. Reporting out the MIC/MEC without an interpretation or the MIC/MEC using the ECV is appropriate for *Aspergillus terreus*. ECVs differentiate strains with acquired resistance mechanisms to common antifungal agents, however their clinical significance is not yet well understood<sup>13</sup>. Currently, the only mold with a clinical breakpoint published is *Aspergillus fumigatus* and voriconazole. Laboratories that follow the CLSI broth dilution method for mold susceptibility testing can use the CLSI ECVs when reporting out their susceptibility results.

EUCAST has interpretive criteria for isavuconazole, itraconazole, and posaconazole against some *Aspergillus terreus* spp.<sup>8,13</sup>

Additionally, they have published epidemiologic cutoff values (ECOFFs)<sup>11</sup>. Laboratories must follow the EUCAST method for mold susceptibility testing if the EUCAST interpretive criteria or ECOFFs are used.

While *A. terreus* species complex strains may test with low MICs to Amphotericin B, these have no correlated to positive clinical outcomes. CLSI recommends against testing amphotericin B for this organism and it is not a recommended antifungal agents for *A. terreus* species complex infections.

Usually, *A. terreus* is fully susceptible against azole drugs displaying MICs lower than epidemiologic cutoff values.<sup>6</sup>

**A recent and extensive review on *A. terreus* complex has been published by the ASM (see ref. 4)**

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