Challenge MY2304-3

April 2023

Eye: *Fusarium* species

HISTORY

This challenge was sent as a simulated eye swab sample from a 65 year old patient with eye infection. Laboratories were expected to isolate and identify *Fusarium* species.

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. *Fusarium* species was isolated as a pure culture.

The challenge sample lot was confirmed to be homogeneous and stable for 48 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

9/11 (82%) participants reported *Fusarium* species, one lab reported *Fusarium oxysporum* complex, and one participant reported *Acremonium* spp. (Table 1).

COMMENTS ON RESULTS

The hyphae of *Acremonium* are generally narrower than that of *Fusarium*, approximately <2µm in width smaller.

Like *Acremonium, Fusarium* can produce small conidiophores with 1 or 2 celled conidia singly or in clusters of similar size on the tips of the phialides. As a result of this characteristic, it may be difficult to differentiate the two species especially in early culture growth. However, in mature growth, *Fusarium* can also produce unbranched or branched coniophores with phialides that produce large sickle or canoe shaped macroconidia, with 3 to 5 septa. [1]

Grading

Reporting *Fusarium* species was graded Acceptable.

Reporting *Acremonium* species was graded Unacceptable.

IDENTIFICATION

Fusarium species can be recovered easily on routine media, and there are no specific growth requirements. The colony is usually white and cottony but can develop a pink or violet centre; the reverse is usually light (Figure 1) however, *Fusarium* species can exhibit a remarkable degree of variation with respect to morphological and physiological characteristics. [1,2]

Microscopic morphology

Fusarium species may produce three types of spores called macroconidia, microconidia, and chlamydospores.

Macroconidia. Macroconidia of *Fusarium* species can be sickle- or canoe-shaped and they may show considerable variation within individual species (Figure 2). [1,2]

Microconidia. Their presence or absence is important for species differentiation; if microconidia are present, the shape and the mode of formation (singly, in false heads only, or in false heads and chains) are important identification factors.

The morphology of the conidiophores bearing the microconidia is also a primary taxonomic character. These conidiophores may be either monophialides only or both monophialides and polyphialides in a given species producing microconidia.

Chlamydospores may be present in some species; they may be formed singly, in pairs, in clumps, or in chains, with either rough or smooth walls. [1,2]

Table 1. Identification results

Reported	Labs	Grade
Fusarium species *+/- refer*	9	Acceptable
Fusarium oxysporum complex	1	Acceptable
Acremonium spp.	1	Unacceptable
Total	**11**	

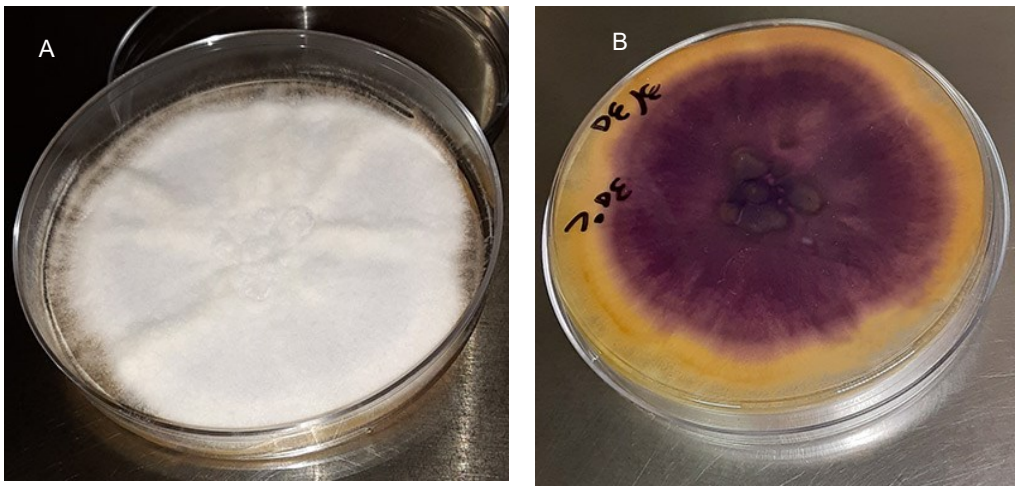

Figure 1. Colony of *Fusarium species* on Sabouraud's dextrose agar. A: front B: reverse

Rapid diagnosis to inform prompt and appropriate treatment is critical to the successful clinical management of fungal keratitis.

PCR has emerged as both a sensitive and specific test for the diagnosing fungal keratitis, [3,4] however the accuracy of PCR to diagnose fungal keratitis is dependent on adequate sampling and the primers used. Some methods allow for rapid species identification aiding in the selection of appropriate treatment as certain species, like *Fusarium solani*, have shown to have a worse prognosis and higher resistance to voriconazole. [5]

Matrix-assisted laser desorption/ionization time of flight-mass spectrometry (MALDI-ToF MS) is a rapid and reliable tool for the identification of microorganisms, allowing the identification of fungal isolates within minutes. [6,7] Several studies have been conducted on *Fusarium* species with high success rates of 82%–99%.[8] It is important particularly for species that are uncommonly recovered that each laboratory validate their own MALDI-ToF MS system (usually at least five isolates) of the suspected species before using this method to report such strains to the species level.

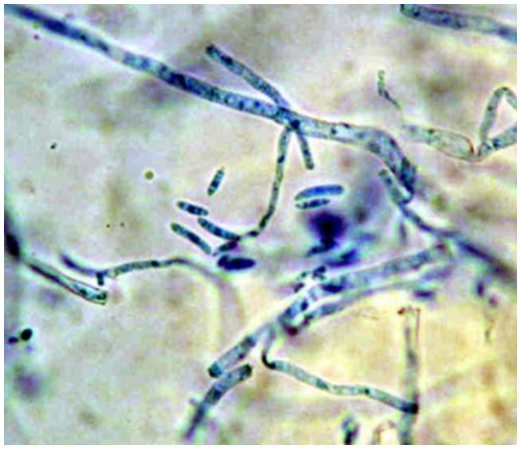

Figure 2. *Fusarium species*, cotton-blue stain under magnification of 400X.

ANTIFUNGAL SUSCEPTIBILITY

In vitro susceptibility testing of *Fusarium* is becoming increasingly important because of frequency and diversity of infections and because resistance profiles are species-specific. Reference methods for antifungal susceptibility testing have been established by Clinical and Laboratory Standards Institute (CLSI) and European Committee on Antimicrobial Susceptibility (EUCAST).

Because different *Fusarium* species show different tendencies in their susceptibility against various antifungal compounds,[9] susceptibility testing should be included in particularly in severe infections.

Espinel-Ingroff et al.[10] established epidemiological cutoff values (ECVs) for some species in order to distinguish wild-type from non-wild-type populations. Although clinical interpretative breakpoints for in vitro antimicrobial susceptible testing for *Fusarium* are not yet established, epidemiological cutoff values for three *Fusarium* species have been suggested. [10]

Fusarium susceptibility to voriconazole is variable,[11] amphotericin B has shown to be most active agent against clinical and reference strains

Due to the poor prognosis obtained with monotherapy, combination therapy may be considered in severe *Fusarium* species infections. [12]

CLINICAL RELEVANCE

Mycotic or fungal keratitis is a severe and potentially blinding infection of the cornea and is considered an ophthalmic emergency.[13]

Accurate diagnosis remains challenging as it is frequently not possible to clinically distinguish bacterial and fungal mycotic keratitis. Obtaining a detailed clinical history and alerting the laboratory of the suspected etiology is essential for a rapid diagnosis and treatment. [14]

The infection is very challenging to treat due to resistance of *Fusarium* spp. to many antifungals.[15]

Fusarium species are cosmopolitan soil saprobes and facultative plant pathogens that can cause infection or toxicosis in humans and animals. [16]

Mycotic keratitis is a mycosis of the cornea and can be caused by a wide variety of fungi. Keratitis due to filamentous fungi (*Fusarium, Aspergillus*, phaeohyphomycetes and *Scedosporium apiospermum*) most commonly occurs after trauma although previous use of corticosteroids and contact lens wear are gaining importance as risk factors. Mycotic keratitis can also be caused by yeasts; in this case, there is usually some systemic or local (ocular) defect. [14]

Although *Pseudomonas aeruginosa* has been reported as the most common cause of contact lens associated keratitis, outbreaks have been reported with both *Fusarium* and *Acanthamoeba* species. [17,18]

The definitive diagnosis of fusariosis requires the isolation of *Fusarium* species from clinical specimens (blood, skin, sinuses, lungs, other).

Culture identification is important because of the histopathological similarities between *Fusarium* and other members of the hyalohyphophomycosis family and the different susceptibilities of these pathogens to antifungal agents.[19]

It is important to establish that they are truly acting as pathogens, for example, by ensuring that there is evidence (such as microscopy) of tissue penetration.

Fusarium species recorded as causes of human disease include *F. chlamydosporum, F. moniliforme, F. nivale, F. oxysporum, F.proliferatum, F. solani* and *F. verticillioides* amongst others.[16,20]

IN RECENT NEWS

Fungal Meningitis Outbreak Associated with Procedures Performed under Epidural Anesthesia in Matamoros, Mexico

CDC, the Mexican Ministry of Health, and U.S. state and local health departments are responding to a multinational outbreak of fungal meningitis.

As of June 23, 2023 there were 169 persons under investigation, 16 suspected cases, 10 probable cases, 9 confirmed cases, and 6 deaths.

Molecular testing has detected fungal signals consistent with *Fusarium solani* species complex from the CSF of some patients involved in this outbreak.

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