

Laboratory Medicine Care Division

Mycology

A. fumigatus - Molecular Detection of Antifungal Resistance

QIAGEN PyroMark Advanced 24 assay to assess azole antifungal resistance in the *Aspergillus fumigatus* azole target *cyp51A*.

Also known as *Aspergillus fumigatus cyp51A* pyrosequencing assay, *Aspergillus fumigatus* azole antifungal resistance testing by pyrosequencing, molecular resistance testing of *Aspergillus fumigatus*

General information

Turnaround time: Performed on demand – TAT 2-4 weeks, depending on the sample.

Sample type/container:

For primary samples, see “**Aspergillus species PCR (Quantitative)**” for a description of sample type and containers (<https://mft.nhs.uk/the-trust/other-departments/laboratory-medicine/mycology/a-z-of-mycology-tests/#Aspergillus-PCR-on-respiratory-samples>).

All primary samples are initially processed by *Aspergillus* species PCR (Quantitative) to determine suitability (>3000 copies/ml 18S rDNA as the minimum cutoff). Therefore, both the **Aspergillus species PCR (Quantitative)** and **A. fumigatus - Molecular Detection of Antifungal Resistance** tests must be requested simultaneously.

Cultures of *Aspergillus fumigatus* may also be tested. For isolate testing, use standard isolate transportation containers (slope, vial). See **Identification and susceptibility testing of yeasts and moulds** for a description of sample type and containers (<https://mft.nhs.uk/the-trust/other-departments/laboratory-medicine/mycology/a-z-of-mycology-tests/#yeasts-and-moulds>).

Laboratory Information

Biological interval/clinical decision values:

Fifteen sites within the *cyp51A* gene of *Aspergillus fumigatus* associated with triazole-antifungal resistance are screened for.

Results are reported as a list of triazole resistance-associated polymorphisms detected, with an interpretation of the expected susceptibilities for itraconazole, voriconazole and posaconazole, according to published literature.

When *cyp51A* gene polymorphisms are not detected, the results are reported as ‘no polymorphisms detected’.

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The test requires minimum 3000 copies/ml of *Aspergillus fumigatus* 18S rDNA. If the copy number is insufficient to amplify the *cyp51A* gene, the DNA amplification will fail. It will also fail in case of non-*Aspergillus fumigatus* species. When the DNA amplification fails, the result is reported as 'DNA amplification not successful'.

Clinical Information

No specific time of optimal collection.

The results need to be taken in context of the clinical condition of the patient and other fungal diagnostic test results. Mixed infections of susceptible and resistant strains do occur.

Please contact MRCM for further information.

References:

Novak-Frazer L, Anees-Hill SP, Hassan D, Masania R, Moore CB, Richardson MD, Denning DW, Rautemaa-Richardson R. Deciphering *Aspergillus fumigatus* *cyp51A*-mediated triazole resistance by pyrosequencing of respiratory specimens. J Antimicrob Chemother. 2020 Dec 1;75(12):3501-3509. doi: 10.1093/jac/dkaa357.

van der Torre MH, Novak-Frazer L, Rautemaa-Richardson R. Detecting Azole-Antifungal Resistance in *Aspergillus fumigatus* by Pyrosequencing. J Fungi (Basel). 2020 Jan 10;6(1):12. doi: 10.3390/jof6010012.

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