

Budget-friendly protocol for TR34/L98H and TR46/Y121FT289A mutation detection in *Aspergillus* section *Fumigati* isolates

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Introduction

Aspergillus section *Fumigati* is one of the most common sections, in the environment [1].

It has been found in different occupational environments, such as sawmills and waste sorting [1,2].

Its cryptic species show intrinsic resistance to several antifungals [3].

Resistance in *A. fumigatus* is emerging due to selective pressure caused by the prolonged use of azoles.

It is often associated with mutations in the Cyp51A gene [3].

The fungal priority pathogens list (WHO), includes *A. fumigatus* with critical priority [4].



Further analysis to identify potential resistance mechanisms and mutations is needed.

Objective

This evaluation aims to offer a protocol for mutation detection in *Aspergillus* section *Fumigati* isolates, It will contribute for the development of guidance that can support future occupational exposure assessments.

Methodology

Hypotheses were determined based on the advantages and disadvantages of each suggested method, including its cost.

	Whole genome sequencing:	Sequencing (x2) of all isolates:	Sequencing (x2) of resistant isolates:	Sequencing + ddPCR:	Incubation + RT-PCR:
	All information with one analysis	All the necessary information	Information regarding the section and the presence of TR and point mutations	Information regarding the presence of TR and point mutations	All the necessary information
	Additional unnecessary information for this analysis		Loss of information regarding point mutations in the CYP51A	Cryptic species not identified	Loss of information regarding point mutations in the Cyp51A

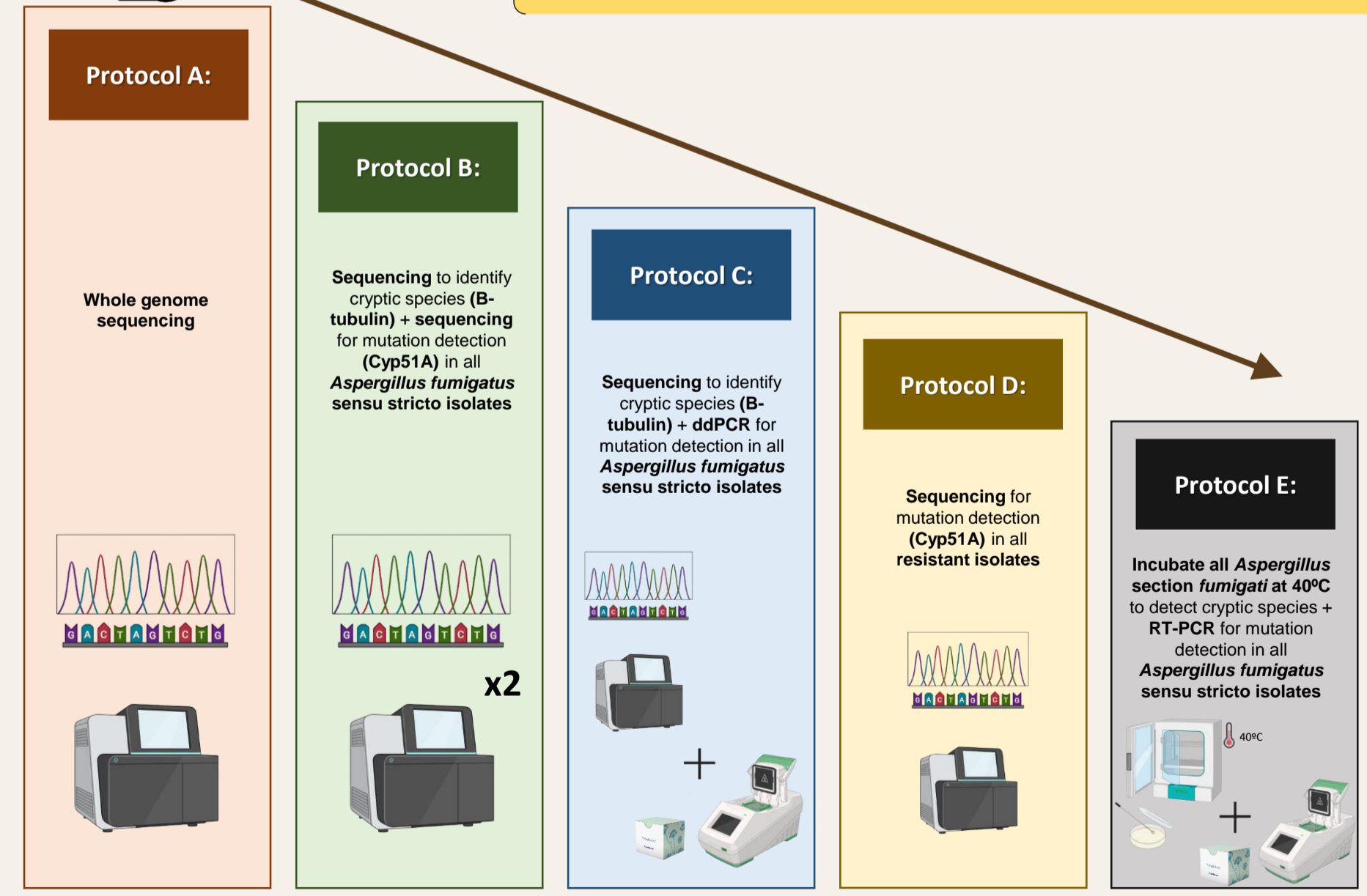
*Tandem repeat

Results and discussion

- Azole resistance is mostly caused by particular mutations in CYP51A [5].
- Wild-type CYP51A-resistant isolates question the effectiveness of the available methods [5].
- Whole-genome sequencing is becoming increasingly common to address these issues [5].



- 1st – Perform susceptibility testing through the EUCAST 9.4 method (microdilutions)
- 2nd – Identify which are *Aspergillus* section *Fumigati* cryptic species through sequencing or thermotolerance
- 3rd – Select one of the following protocols for mutation detection



Conclusions

This study allowed determining several ways to detect mutation in *Aspergillus* section *Fumigati* isolates. It provided the necessary tools to perform an accurate occupational exposure assessment to *Aspergillus* section *Fumigati* and allowed a more detailed risk assessment while overcoming cost issues at the same time.

References

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