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scRNA sequencing of circulating peripheral blood mononuclear cells from people with cystic fibrosis and nontuberculous mycobacteria pulmonary disease

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Background: People colonized with *Mycobacterium abscessus*, such as people with cystic fibrosis (PwCF), are at risk of nontuberculous mycobacterial pulmonary disease (NTM-PD) and have heterogeneous clinical outcomes. New reliable biomarkers, such as immune signatures, are among the unmet needs for better clinical management of NTM-PD.
Methods: We collected blood samples from PwCF with no history of NTM detection and clinically stable (CF), with NTM-positive isolates and clinically stable (CF-NTM), and with NTM-positive isolates and a clinical diagnosis for risk of pulmonary disease (CF-NTM-PD). We isolated peripheral blood mononuclear cells (PBMCs) and plasma to perform scRNA-sequencing and Luminex assay.
Results: We performed scRNAseq on 10 PBMC samples, identifying 10 cellular immune populations according to expression levels of canonical markers. We performed differentially expressed gene (DEG) analysis in the three groups in each cell population to identify transcriptomic signatures characterizing CF-NTM-PD. We compared DEGs in samples from people at risk of pulmonary disease (CF-NTM-PD) with those from the other two groups (CF, CF-NTM) and identified unique RNA transcriptomic profiles activated in monocytes that were specifically upregulated in the CD14+ monocyte cluster of CF-NTM-PD.
Conclusions: Our scRNA-sequencing data suggest that monocyte responses are present in PwCF witath risk of NTM-PD.
Acknowledgements: This work was supported by FFC#7/2022 and FFC#20/2020 from the Fondazione Italiana per la ricerca sulla Fibrosi Cistica."

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Exploring the diversity and function of the RTA1-like protein family in *Aspergillus fumigatus*

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Background: The common pathogenic mold *Aspergillus fumigatus* is responsible for most tissue-damaging, invasive pulmonary aspergillosis cases. *Aspergillus* is also the main fungus cultured in the airways of people with cystic fibrosis (CF), a progressive genetic disease [1, 2]. The widespread presence of *A. fumigatus* and its role in conditions such as invasive pulmonary aspergillosis and CF is a significant global burden, with these conditions collectively leading to more than 500,000 deaths worldwide annually [3]. The RTA1-like protein family is common in fungi, particularly *A. fumigatus*, which harbors more than 20 RTA1 gene copies. These proteins have multiple transmembrane regions and are unique to fungi. RTA1 transcripts are significantly upregulated when fungi encounter several cellular stressors. Although some RTA1 genes in yeast have been examined, the function of genes in filamentous fungi is unclear [4, 5].
Methods: Using a combination of large-scale genome-wide analysis and gene expression data, we examined RTA1 proteins in *A. fumigatus* and identified more than 800 in aspergilli, with 25 copies in the *A. fumigatus* strain A1163. Despite sharing core features, these proteins exhibit significant sequence variability, indicating specialized roles. Gene expression data revealed distinct regulatory networks for RTA1 proteins, suggesting responses to specific environmental cues.
Results: We generated a deletion library of all *A. fumigatus* RTA1 mutants and screened for their susceptibility to various clinically relevant stressors alongside the WT. We uncovered that multiple RTA1 genes may have a crucial role in responding to clinically relevant stressors.
Conclusions: Treating *A. fumigatus* infection is challenging in CF, with rising antifungal resistance and lack of new therapeutics to target this organism. Complementation of mutants of RTA1-encoding genes in *A. fumigatus*, coupled with transcriptomics studies, will provide further insight into the biological roles and therapeutic potential of targeting these fungal-specific, stress-responsive proteins.
Acknowledgements: SKD is supported by grant K22AI174009 from the National Institute of Allergy and Infectious Diseases.

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Diagnostic target product profiles for managing infections and exacerbations in cystic fibrosis

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Background: The Cystic Fibrosis Antimicrobial Resistance Syndicate (CF Trust, Medicines Discovery Catapult, LifeArc) set out to deliver a suite of diagnostic target product profiles (TPPs) to address unmet needs in managing infections and pulmonary exacerbations in cystic fibrosis (CF). Diagnostic TPPs describe the required characteristics for tests to address a specific need and can act as an instruction list for developers.
Methods: The TPPs were developed by engaging with people with CF and their multidisciplinary care teams in focus groups to understand current and evolving unmet diagnostic needs. Discussions focused on areas including early detection of pulmonary exacerbations, sampling considerations and alternatives to sputum, changes in symptoms of infections for people on modulators, and utility of antimicrobial susceptibility in infection management. Several high-level TPPs were outlined, each addressing a different unmet need. The TPP characteristics were refined through extensive stakeholder engagement, a series of surveys, and a virtual symposium.