The prevalence and diversity of fungi in respiratory samples of cystic fibrosis patients – a Dutch, nationwide, prospective, multicentre study

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Introduction

- Progressive lung injury in cystic fibrosis (CF) patients leads to chronic colonization with bacteria and fungi.
- At present there is no clear understanding of the clinical significance of fungal growth in respiratory samples and whether treatment should be advised.
- Fungal colonization is obtained from the environment which necessitates locally performed epidemiology studies.

Objective

- To investigate the prevalence and diversity of fungi in Dutch CF patients
- To assess azole resistance in *Aspergillus fumigatus*

Material and Methods

- All fungal isolates cultured in respiratory specimens of CF patients from 5 CF centers were collected over a 3 year period.
- Samples were inoculated on Sabouraud dextrose agar (SDA) and Medium B+.
- Molecular identifications were performed in one centre using Amplified Fragment Length Polymorphism (AFLP) fingerprinting, rDNA PCR and ITS, calmodulin and β -tubulin sequencing.
- Azole resistance was assessed for all *A. fumigatus* using a qPCR assay followed by phenotypic confirmation.

Results

- In 699 patients fungi were recovered, corresponding with 3,787 cultured fungal species.
- In total 107 different fungal species were identified

Prevalence of cultured fungi in respiratory samples of Dutch CF patients

Year	Prevalence			
	Year 1	Year 2	Year 3	Mean
<i>Aspergillus fumigatus</i>	30.0%	31,6%	32,8%	31,7%
<i>Aspergillus</i> spp.	7,0%	3,7%	6,6%	5,6%
<i>Penicillium</i> spp.	16,6%	11,6%	11,2%	12,6%
<i>Scedosporium</i> complex species	2,1%	2,8%	7,8%	4,5%
<i>Cladosporium</i> spp.	1,5%	1,1%	0,8%	1,1%
<i>Exophiala dermatitidis</i>	1,0%	1,1%	1,3%	1,1%
Any fungi	42.3%	40.7%	39.6%	40.7%

Azole resistance of *Aspergillus fumigatus* in Dutch CF patients

Resistance genotype	Year 1	Year 2	Year 3	Mean
TR34/L98H	9 (4,1%)	26 (7,1%)	16 (4,1%)	5,2%
TR46/T289A/Y121F	4 (1,8%)	6 (1,6%)	4 (1,0%)	1,4%
M220	0,0%	1 (0,3%)	1 (0,3%)	0,2%
G54W	0,0%	1 (0,3%)	1 (0,3%)	0,2%
Total resistance	5,9%	9,3%	5,6%	7,1%

*Year 1, 09/03/2010–31/3/2011; Year 2, 01/04/2011–31/3/2012; Year 3, 01/04/2012–27/3/2013

Conclusions

- This is the first Dutch multicentre study analyzing the diversity and prevalence of fungi, in respiratory samples of CF patients.
- A vast diversity of filamentous fungi was demonstrated, dominated by *Aspergillus* and *Penicillium* species.
- Because of a mean resistance rate of 7.1% we recommend susceptibility testing of *A. fumigatus* isolates in CF patients with azole-based antifungal treatment.