

Results 233 children were followed for a median of 2.0 (1.7–2.3) years. The median (IQR) age was 8.0 (5.0–12.2) years. 71 children isolated PA during the study period. The incidence rate (95% CI) of new PA infections was 0.15 (0.10–0.22) cases per patient year for those that had never previously isolated PA and 0.19 (0.13–0.27) cases per patient year for those that had been free from PA for >12 months. This rate varied between 0.08 (0.04–0.18) and 0.28 (0.14–0.49) cases per patient year at the 8 recruiting centres. 42% of children were asymptomatic at the time of PA acquisition. The median (IQR) number of antibiotic courses per patient year varied between the centres: 0.6 (0.2–1.3) to 3.6 (3.1–4.2) for oral and 0.0 (0–0) to 0.4 (0–1.2) for intravenous.

Conclusions This is the first prospective study to report the incidence of new PA infection in a large cohort of children with CF, considered to be free of PA airway infection. Incidence rate was higher in children who had isolated PA previously. The variation between centres is not easily explained and needs further investigation.

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P198 NEW APPROACHES TO THE CULTURE OF MYCOBACTERIUM ABSCESSUS COMPLEX FROM PATIENTS WITH CYSTIC FIBROSIS

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Introduction *M. abscessus* complex (Mab) are Rapid Growing Mycobacteria (RGM) that can cause severe infection. Prevalence is increasing and a recent study using whole genome sequencing showed cross infection between Cystic Fibrosis patients. Frequent surveillance for Mab infection may allow earlier diagnosis and prevent spread.

Automated broth (e.g. MGIT), is a sensitive rapid method for mycobacterial culture. Decontamination is needed to kill other bacteria and yeasts before culturing CF sputa in MGIT, but decontamination may reduce Mab numbers.

Other possibilities include chlorhexidine decontamination which yields more Mab but is incompatible with MGIT. Some mycobacteria grow directly from sputum on *Burkholderia cepacia* selective agar (Bcc) after extended incubation, without prior decontamination.

The aim of this study was to improve Mab culture from CF sputum.

Methods We compared MGIT culture of CF sputa with extended incubation of Bcc used in the routine laboratory. We compared growth of 30 known Mab on 3 formulations of Bcc and 2 Middlebrooke selective agars. We took 12 sputa from 9 CF patients with Mab infection and compared MGIT with culture on selective agars or chlorhexidine decontamination followed by culture onto non selective agar. Mycobacteria were identified by the National Mycobacterium Reference Laboratory and an in house PCR.

Results Eighteen of 515 CF sputa grew RGM (9 on Bcc agar and MGIT, 3 MGIT alone, 4 Bcc alone and 2 on Bcc with no MGIT culture). Contamination with other bacteria and fungi made it extremely difficult to see RGM on the routine Bcc.

Thirty sequenced *M. abscessus abscessus*, *M. bolletii* and *M. massiliense* all grew on the 3 commercial Bcc and 2 Middlebrooke agars.

One Bcc and one Middlebrooke agar successfully cultured RGM from all 12 sputa with fewest contaminants. Chlorhexidine decontamination and blood agar was effective but labour-intensive. Only 8 of 12 MGIT cultures grew RGM.

There was no difference in time to positive culture between agar and MGIT.

Conclusion Culture onto selective agar may be more sensitive than MGIT. It is quantitative and provides pure culture for identification, typing and susceptibility testing.

This may be a sensitive cost-effective way to screen sputa from patients at risk.

P199 MOLECULAR ANALYSIS DEMONSTRATES SHARED STRAINS OF MYCOBACTERIUM ABSCESSUS ISOLATES IN CYSTIC FIBROSIS PATIENTS ATTENDING A SINGLE CENTRE

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Introduction and objectives The *Mycobacterium abscessus* complex is an emerging group of pathogens in cystic fibrosis (CF), which may cause cross infection. The aim of this study was to determine whether CF patients infected with *M. abscessus* attending a single UK adult CF centre harboured unique or shared strains of *M. abscessus*.

Methods Isolates were from 12 patients attending a single adult CF centre, who yielded one or more positive sputum cultures for *M. abscessus* complex during the period January 2010 to August 2013. Isolates were identified to subspecies level using *hsp65-rpoB* concatenated sequence cluster analysis. Variable Number Tandem Repeat (VNTR) analysis was used to compare these isolates and determine whether two or more patients were infected with the same strain.

Results 11 isolates were identified as *M. abscessus sensu lato*. VNTR analysis demonstrated 2 clusters of 6 and 2 patients carrying the same strains of *M. abscessus sensu lato*, both

Abstract P199 Table 1 Mycobacterium abscessus cluster sequence analysis and Variable Number Tandem Repeat profiling results

Patient	<i>M. abscessus</i> subspecies	VNTR Profile
1	<i>abscessus</i>	2+, 5+, 3, 6, 2, 5, 1+, 2+, -
2	<i>abscessus</i>	2+, 5+, 3, 6, 2, 5, 1+, 2+, -
3	<i>abscessus</i>	2+, 5+, 3, 6, 2, 5, 1+, 2+, -
4	<i>abscessus</i>	2+, 5+, 3, 6, 2, 5, 1+, 2+, -
5	<i>abscessus</i>	2+, 5+, 3, 6, 2, 5, 1+, 2+, -
6	<i>abscessus</i>	2+, 5+, 3, 6, 2, 5, 1+, 2+, -
7	<i>abscessus</i>	3+, 4+, 3, -, 4, 3+, 2+, 2+, 2
8	<i>abscessus</i>	3+, 4+, 3, 2, 4, 3+, 2+, 2+, 2
9	<i>abscessus</i>	2+, -, 3, 4, 2, 5, 1+, 2+, -
10	<i>abscessus</i>	-, 5+, 2, 2+, 4, 5, 1+, 3, 2
11	<i>abscessus</i>	1+, 4+, 2, 2, 4, 3+, 2+, 2+, 2
12	<i>boletii</i>	2+, -, 5, 5, 4, 3+, 1+, 2, 1+

Corrections

P198 - withdrawn

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