



Journal of

Cystic Fibrosis

The Official Journal of the European Cystic Fibrosis Society

Title:

Investigating *Pseudomonas aeruginosa* population structure and frequency of cross-infection in UK cystic fibrosis clinics - a reference laboratory perspective.

Lay Title:

Molecular fingerprinting of *Pseudomonas aeruginosa* from PWCF, and a UK reference laboratory investigation into frequency of cross-infection

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What was your research question?

As a reference laboratory we monitor *Pseudomonas aeruginosa* strain types among people with CF (PWCF). We wanted to investigate how common the Liverpool epidemic strain (LES) was compared with strains commonly found in the environment, and with those received from other hospital patients. We also aimed to evaluate any evidence for cross-infection within centres.

Why is this important?

PWCF, their families and CF clinical teams invest considerable effort in preventing crossinfection to limit the spread of strains such as LES. As a reference laboratory our role is to "fingerprint" *P. aeruginosa* isolates to identify strain types from PWCF submitted from hospitals as part of annual review. The results are sent back to the clinical teams who monitor any changes. Because we receive isolates from a lot of UK CF centres, we are fortunate in

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being able to generate a national "picture" which shows any changes in the frequency of certain strains and any new strains.

What did you do?

We performed molecular "fingerprinting" of *P. aeruginosa* from 2619 PWCF submitted by hospitals to our laboratory between 2017 and 2019. This was conducted using a DNA-based method which produces a unique 9-digit numerical code (i.e. profile) for every strain type. We compared these profiles within and between hospitals, and assessed how many PWCF had LES, how many had common strains found in the hospital environment or among other hospital patients (or both), and how many had their own unique strain. We also looked at the whole DNA content (genome) of four sets of closely related strain "clusters" from one hospital to assess evidence of cross-infection.

What did you find?

51% of 2619 PWCF carried either shared strains found relatively frequently in clinical and environmental settings, or transmissible strains such as LES, while 49% of PWCF had their own unique strain or one shared with a small number of other PWCF. The occurrence of LES was 9.5% and had not increased since our previous estimate of 10% in 1204 PWCF, published in 2013. Analysis of the whole genomes of representatives of four common shared strains from PWCF attending one centre found limited evidence of cross-infection. Hospital-specific strains shared between small numbers of PWCF were found in 11 of 12 centres, but recent new acquisitions were uncommon.

What does this mean and reasons for caution?

These data suggested that new acquisitions of LES and of other shared strains were relatively uncommon. Although some evidence for cross-infection was found, this mostly involved small numbers of individuals and was mainly "historical" with few examples of new infections, suggesting existing cross-infection measures are helping to limit transmission. However, although isolates were submitted to the reference laboratory from 21 of 25 adult CF centres and 20 of 26 paediatric centres, data from the remaining centres were not available.

What's next?

We will continue to monitor the prevalence of different *P. aeruginosa* strains, and to report to hospitals any new incidences of shared strains between PWCF. Increased remote testing and increased availability of CFTR modulators may affect the frequency of certain strains so

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we aim to evaluate these changes in the coming months by working with clinical colleagues and publishing our findings.

Original manuscript citation in PubMed

https://pubmed.ncbi.nlm.nih.gov/37271666/

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