

In conclusion, biofilm production is an important virulence factor in infections caused by *Candida* species, bearing in mind that infections caused by non-albicans species have increased recently. Therefore, preventive measures such as the use of antimicrobial coating of biomaterials should be applied in order to prevent infections caused by *Candida* species.

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Lack of isolation of *Pseudomonas aeruginosa* associated with agricultural practices: relevance to patients with cystic fibrosis

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Cystic fibrosis (CF) is the most common inherited disease in persons originating from a white and European background and has a genetic carriage rate of one in 20 persons and an incidence of one in 2500 live births.¹ It is an autosomal recessive condition whereby two alleles carrying a polymorphism in the CF transmembrane conductance regulator (CFTR) gene phenotypically manifest the disease state through a variety of multiorgan problems, and is associated with a pharmacological dysfunction to regulate chloride ion secretion across cell membranes.

The most common complication of CF is the recurrence of chronic chest infections usually caused by bacterial pathogens.⁴ Cystic fibrosis patients continue to suffer from recurrent and chronic respiratory tract infections and most

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Table 1. Description of air sampling procedures for the isolation of *Pseudomonas aeruginosa*, including aerial sampling locations and volumes of air sampled.

Location description	Start location (coordinates)	End Location (coordinates)	Volume of air sampled (L)	Presence of <i>P. aeruginosa</i>
Rural motorway	Moirá (N54°29'.250";W006°15'.900")	Portadown (N54°41'.700";W006°43'.400")	18,750	Negative
Farmland	Loughgall (N54°27'.273";W009°3'.21")	Moy (N54°54'.200";W006°69'.400")	17,250	Negative
Farmland	Moy (N54°54'.200";W006°69'.400")	Caledon (N54°21'.820";W006°50'.940")	18,750	Negative
Farmland	Caledon (N54°21'.820";W006°50'.940")	Emyvale, Co. Monaghan (N54°34'.557";W006°95'.167")	22,500	Negative
Rural road	Ballygawley (N54°27'.250";W007°01'.220")	Dungannon (N54°28'.500";W006°46'.530")	12,750	Negative
Rural-urban motorway	Dungannon (N54°28'.500";W006°46'.530")	Belfast (N54°35'.060;W005°57'.340")	33,750	Negative

of their morbidity and mortality is due to such infections throughout their life.⁵ These infections are usually dominated by Gram-negative organisms, especially by the pseudomonads, particularly *Pseudomonas aeruginosa*. Once acquired, this organism is responsible for the establishment of bacterial biofilms in the lung, which makes its eradication extremely difficult and often generally impossible, leading to a chronic state of colonisation, infection and inflammation. Thus, a thorough understanding of the environmental sources of this organism is important to help judge the risk of its potential environmental acquisition for non-colonised CF patients.

Among CF patients and their families, there is great interest and anxiety about where such patients may acquire this bacterium from the environment and frequently patients query whether this organism can be acquired from the air or when they visit the countryside. Given that Northern Ireland is largely rural with an agrarian-based economy, this study aims to examine airborne and waterborne contamination of *P. aeruginosa*, associated with common local agricultural practices, to assess the airborne and waterborne contamination of this organism, particularly

in the spring when agricultural activities are intensive.

The bacteriological quality of outdoor air for the presence of *P. aeruginosa* was examined during the period May–July 2008. Outdoor air was sampled immediately adjacent to where aerial slurry spreading activities were taking place. An electrical slit air sampler (CF Casella, London, England) was employed, in accordance with the manufacturer's instructions, powered by a small portable petrol electrical generator. After thorough decontamination of the air slit sampler with absolute ethanol, sampling was carried out while traversing the rural countryside by vehicle immediately adjacent to where the slurry was falling (Table 1). For bacteriological examination, agar plates (8 cm diameter; Sterilin) of *Pseudomonas* Isolation Agar (PIA; Oxoid CM0559 + SR0102) were employed. Following air sampling, plates were incubated for five days at 37°C, after which any colonies were confirmed morphologically by Gram stain. Where presumptive colonies of *P. aeruginosa* were indicated, colonies were confirmed by a specific polymerase chain reaction (PCR), as described previously.²

The bacteriological quality of surface waters from 12 surface waterways was examined for the presence of

Table 2. Description of results and sampling locations for the isolation of *Pseudomonas aeruginosa* from 12 environmental surface waters.

Sample number	Watercourse	Temperature	Grid coordinates	Presence of <i>Pseudomonas</i> spp.	GenBank Accession Number submitted
1	Lower River Bann	21°C	N54°45'.257";W006°27'.725"	–	
2	River Moyola	20°C	N54°47'.921";W006°36'.492"	–	
3	Lissan Water	20°C	N54°46'.901";W006°48'.655"	+ (<i>P. tolaasii</i>)	GQ221034
4	Lough Fea	18°C	N54°43'.836";W006°49'.546"	–	
5	Unnamed stream	18°C	N54°46'.297";W006°50'.635"	+ (<i>P. fluorescens</i>)	GQ214543
6	Unnamed stream	18°C	N54°44'.671";W006°55'.340"	+ (<i>Pseudomonas</i> sp.)	GQ214544
7	Unnamed stream	17°C	N54°43'.896";W006°57'.491"	+ (<i>Pseudomonas</i> sp.)	GQ214545
8	Unnamed stream	19°C	N54°42'.455";W007°02'.876"	+ (<i>Pseudomonas</i> sp.)	GQ214546
9	Unnamed stream	17°C	N54°41'.225";W007°05'.217"	–	
10	Owenreagh River	20°C	N54°41'.064";W007°05'.897"	+ (<i>Pseudomonas</i> sp.)	GQ214547
11	Upper River Bann	20°C	N54°28'.003";W006°25'.652"	+ (<i>Pseudomonas</i> sp.)	GQ214548
12	River Kells	18°C	N54°48'.486";W006°13'.982"	+ (<i>P. fluorescens</i>)	GQ214549

P. aeruginosa (Table 2). Rural waterways were sampled at random and all sampling took place within an area that represented prime agricultural grazing land for sheep and cattle. Sampling at each site was performed by aseptically obtaining 100 mL water in a sterile disposable plastic universal container (Sterilin). All samples were processed within 6 h of sampling. Individual water samples (100 µL) were inoculated on PIA medium and spread uniformly on the surface of the agar with the aid of an L-shaped spreader, and the plates were incubated at 37°C for 72 h. Presumptive positive isolates were further characterised by sequence analysis of their near complete 16S rDNA gene locus.

P. aeruginosa was not detected in any air sampled, nor was it found in any surface water examined. However, eight surface waters were positive for other *Pseudomonas* species, including *P. fluorescens* ($n=2$) and *P. tolaasii* ($n=1$). Five other waters sources were positive for other species of *Pseudomonas* (Table 2). All resulting 16S rDNA sequences relating to these organisms have now been deposited in GenBank and their accession numbers are given in Table 2.

Water is a well-documented environmental source of this organism;³ however, there are a limited number of published studies describing its presence in water-related activities associated with agriculture and agricultural practices. Many CF patients and their families are concerned to avoid the acquisition of this organism in the lower airways and hence try to avoid high-risk situations/environments where transmission may occur, including swimming, contact with other CF patients infected with *P. aeruginosa* and avoidance of spas and jacuzzis. As a result, many CF patients ask their clinicians and nurses about the risk from recreational use of water in rural areas, where agriculture is the most important factor, as well as the avoidance of inhaling air contaminated with the smell of animal slurry during periods of high slurry spreading activity.

The inability to isolate this organism from intensive agricultural environments is surprising and suggests that the organism may not be as ubiquitous as thought originally. However, given that other species of *Pseudomonas* were cultured from environmental surface waters, CF patients should still consider water and associated work or recreational activities as high risk, where such water may be contaminated with several species of *Pseudomonas*.

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Congee: a cause of gross but transient elevation in plasma creatinine concentration

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A previously healthy 14-month-old boy was admitted to hospital with diarrhoea. Clinical examination was unremarkable. Apart from a markedly elevated plasma creatinine level (222 µmol/L; Roche Modular Analytics, Indianapolis, USA), routine blood tests, including plasma urea, were normal. By the next day his plasma creatinine had spontaneously normalised to 39 µmol/L (Table 1). He was discharged after the diarrhoea settled and subsequently referred for exclusion of renal pathology.

When seen three months later, there was no abnormality on examination but plasma creatinine concentration was 214 µmol/L (Beckman Coulter Synchron, Fullerton, USA) which when repeated four days later was normal (45 µmol/L). Other biochemical and haematological parameters were normal, as were ultrasonography of the renal system and a dimercaptosuccinic acid renal scan. As the creatinine assay based on the Jaffé reaction is subject to interference,¹ creatinine was re-analysed using the creatininase-creatinase enzymatic method (Vitros Chemistry system, Ortho-Clinical Diagnostics, Rochester, USA), which confirmed the elevated plasma creatinine and provided no evidence for assay interference (Table 1). Urinary toxicology screen revealed no abnormality.

On further specific questioning, the parents denied giving any herbs, health products, traditional medicine or drugs to the child but stated that the child had been given pork congee (concentrated meat broth) cooked from 500 g lean pork prior to each hospital visit. Therefore, plasma creatinine (high-performance liquid chromatography [HPLC]-tandem MS) was measured and found to be 424 µmol/L and 167 µmol/L (reference interval: 17–109 µmol/L) in the plasma samples with a creatinine of 214 µmol/L and 45 µmol/L (Table 1).

Dietary creatinine and creatine affect plasma creatinine concentrations. The effect of plasma creatinine interference in the Jaffé creatinine assay is small.² *In vivo* conversion of creatine to creatinine occurs at an average rate of 1.6% daily,³ and therefore normally only makes a small contribution to circulating creatinine concentration. Jacobsen *et al.*⁴ and Mayersohn *et al.*⁵ reported that plasma creatinine levels increase by 50% one and a half to three and a half hours after ingestion of a cooked meat meal. As raw meat ingestion does not give similar effect, the authors suggest that creatinine is produced from creatine during cooking and ingested from the cooked meat.

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