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Improved testing for the pathogenic fungi *Aspergillus fumigatus*

Statement of the Problem: *Aspergillus fumigatus* (AF) is a major cause of invasive aspergillosis (IA) in immunocompromised people with mortality rates as high as 70-90% for patients in the most at-risk group. *A. fumigatus* spores are readily transported by air and are of particular concern in areas where decomposition occurs, e.g., compost sites, farms and waste sites and in hospitals, where the number of immunosuppressed patients is rapidly increasing. The current protocol for measuring *A. fumigatus* in bioaerosols is outdated and relies heavily on laboratory staff accurately identifying closely related species and may substantially underestimate the actual numbers of live *A. fumigatus* in the air. Molecular methods such as quantitative PCR (qPCR) have been used to estimate the numbers of *A. fumigatus* in bioaerosols. However, this method does not quantify active fungi. The purpose of this study is to provide an improved method of quantifying viable *A. fumigatus* emitted in bioaerosols.

Methodology & Theoretical Orientation: RNA extraction efficiency from *A. fumigatus* Af293 was examined using different bead-beating techniques followed by TRIzol extraction. Primers were designed to target the 1-3 β-glucan synthase catalytic sub unit, fksP. Standards were prepared using RT-qPCR products of RNA from *A. fumigatus* Af293. The specificity of the assay was tested against other species of *Aspergillus*, *Neosartorya fischeri* and *Penicillium glabrum* and environmental isolates collected during the study.

Findings: Primer specificity was complicated due to the close relationship with *N. fischeri*, a food-borne fungus phenotypically related to *A. fumigatus*. However, a two-step reverse transcriptase-qPCR (RT-qPCR) assay was designed using the SYBR Green technology.

Conclusion & Significance: Effective treatment of IA depends on the causative agent. Rapidly identifying the most common cause of IA will lead to more targeted and effective treatment and in addition will aid us in identifying contamination in hospital clean rooms.

Biography

Ceri L Gwyther has been working in environmental science since 1998. She has received her MSc in Conservation and Land Management and PhD in Bioreduction (the biodegradation of animal carcasses in a sealed unit containing mesophilic, aerated water by utilizing the intrinsic microbial communities) which offered the perfect opportunity of blending environmental science with microbiology. She pursued her career as a Project Officer in Microbiology. Her current research is focused on the implications of bioaerosols on human health.

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