In-house real time PCR for the diagnosis and prognostication of invasive fungal infections in a tertiary care cancer hospital

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Introduction: Invasive fungal infections (IFI) have emerged as an important cause of morbidity and mortality in cancer patients. Aggressive chemotherapeutic protocols for treatment resulting in prolonged and profound neutropenia, are the most important contributory factors. Patients with hematological malignancies and those undergoing bone marrow transplantation are at high risk of invasive mycoses and an increase in morbidity and mortality. Blood culture lacks the sensitivity but with the availability of molecular techniques, the diagnosis of systemic fungal infections has significantly improved.

Objectives: To evaluate an in-house real-time PCR for the diagnosis of IFI. To correlate the results of PCR with the EORTC classification of invasive fungal infections (IFI).

Methods: 3 ml of whole blood is collected from patients with suspected invasive fungal infections. Extraction is performed and DNA is detected using SYBR green PCR. The panfungal PCR using primers NL1 and 260R targeting a region of the ribosomal gene followed by species specific hybridization with probes for Candida species as well as Aspergillus species.

Results: A total of 80 in patients were included in the study from August 2015 to December 2015 at Tata Memorial Hospital. 52 patients had haematological malignancies and 28 patients belonged to the surgical disease management group (DMG). They were classified by the EORTC criteria as proven, possible and probable cases of IFI of the 80 patients, 49 were positive for yeast DNA and 3 were positive for Aspergillus DNA.

Discussion: Fungal infections, in neutropenic patients with malignancies do not show characteristic signs and symptoms, making accurate diagnosis difficult. Early recognition is crucial, as the progression of invasive disease from detection to death is typically less than 14 days. Empirical treatment with antifungal agents is initiated in high-risk patients with suspected fungal infection. This is associated with high toxicity and high cost.

Conclusions: The SYBR green real time PCR was useful and sensitive indicator for the detection of fungal DNA. The SYBR Green PCR is found to be a reproducible assay and it is validated for patients with Candidemia.

Biography
Prashant Mule has completed his MD in Microbiology from the Department of Microbiology, Tata Memorial Hospital, Mumbai, India in 2016. Presently, he is working as a Senior Resident in the Department of Microbiology. His areas of interests are Mycology, Molecular Microbiology, Virology and prevention of health care associated infections. He has worked on the evaluation of in house real time PCR for the diagnosis and prognostication of invasive fungal infections in a tertiary care cancer institute in Mumbai.

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