Simple, low-cost micro-culture method for rapid diagnosis of mucormycosis in murine model

Hamid Badali[1], Afsane Vaezi[1], Mehdi Fakhar[2]

1. **Mazandaran University of Medical Sciences, Sari Iran, Department of Medical Mycology / Invasive Fungi Research Center (IFRC), Sari, Iran**
1. **Molecular and Cell Biology Research Center, Mazandaran University of Medical Sciences, Sari, Iran**

Mazandaran University of Medical Sciences

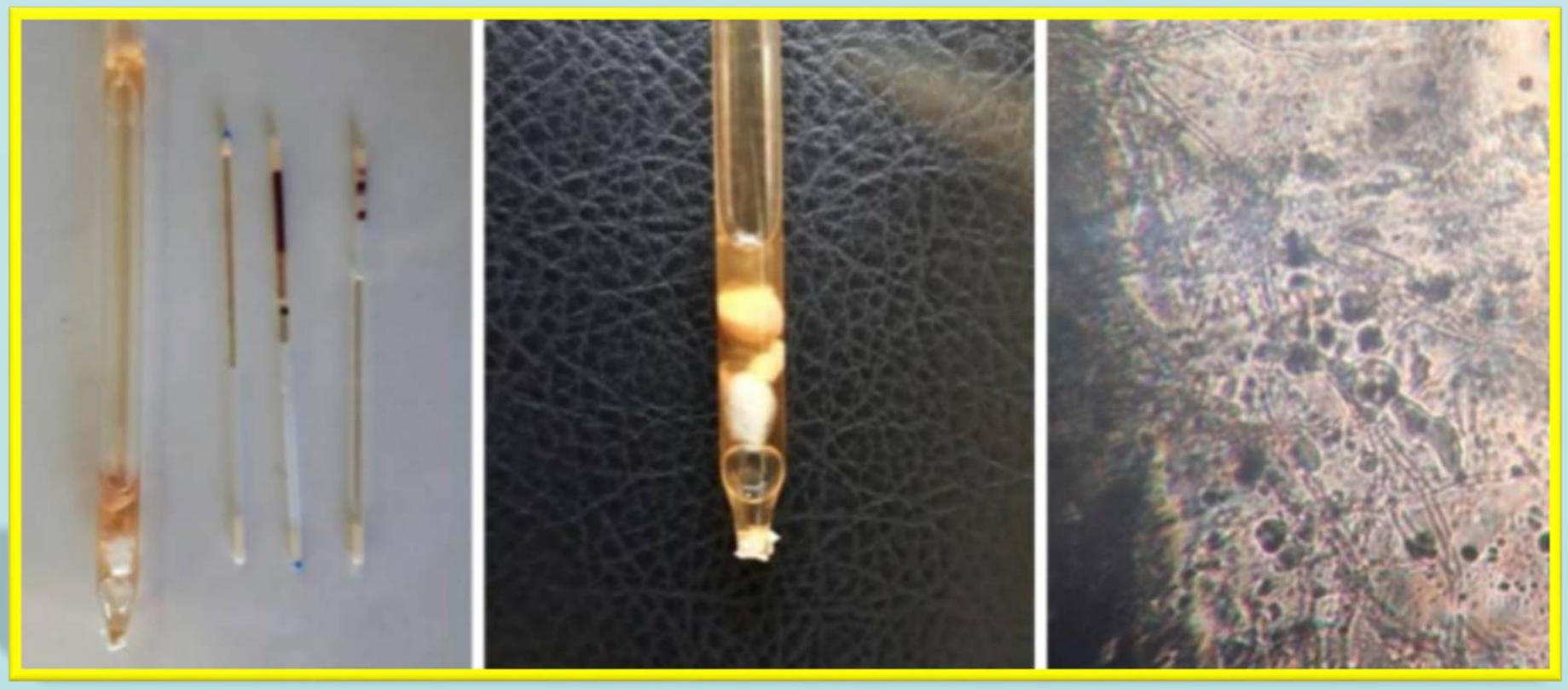
ISHAM
INTERNATIONAL SOCIETY FOR HUMAN AND ANIMAL MYCOLOGY
AMSTERDAM
30 JUNE – 4 JULY 2018

Objectives:

Mucormycosis is a life threatening invasive fungal infection caused by mucoralean fungi and delay in diagnosis and treatment usually results to high mortality rates. This study aimed to describe a simple, low cost micro-culture method for rapid diagnosis of mucormycosis in murine model.

Methods:

Infection by reference strain (*Rhizopus oryzae*), isolated from cerebral mucormycosis was induced in three groups which consist of five immunocompetent mice with three different inoculums (1×10^4, 1×10^5 and 1×10^6 CFU/ml) in a volume of 0.2 ml into the lateral tail vein. Animals were euthanized daily, at day 3 to 7 post infection. Homogenized tissues (brain and kidney) and blood samples were cultured on SDA and diphasic blood-culture bottle and incubated at 35°C. Subsequently, histopathology and molecular assay have performed for confirmation. Micro-culture sampling was adjusted by non-heparinized glass capillary tube with 50-70 µl of RPMI 1640 medium. Homogenized tissue and blood samples were inoculated into capillary tubes and sealed with paraffin wax and incubated at 35 °C. After 24 h, direct examination were performed using invert microscope.



Results:

21 out of 75 (28%) blood samples and 14 of 15 (93.3%) brain and kidney of each samples showed positive micro-culture results. From 25 micro-culture blood samples for each group, 5, 9 and 7 were positive with the inoculum sizes of 1×10^4, 1×10^5 and 1×10^6 CFU/ mouse, respectively. Neither positive blood culture, nor PCR were observed. However, PCR and tissue culture were positive for 25 of 30 (83.3%) and 27 of 30 (90%), respectively.

Conclusion:

Use of micro-culture as a simple, rapid, and reliable method has the potential role to become a valuable surrogate assay for early diagnosis of mucormycosis. Further validation is required to confirm the clinical utility of this method.