

Abstract

Pneumocystis jirovecii is an opportunistic fungi which is classified in Ascomycota phylum. This fungus is colonized in alveolar epithelium of patients with impaired cell-mediated immunity like as AIDS, chronic pulmonary disease, lung malignancy and TB then causes *Pneumocystosis* or intensifies the basic pulmonary disease due to co-infection. Pathogenicity of this fungus is associated with inflammation, obstruction and malignancies of respiratory ways. In this cross sectional study we used two methods of Nested-PCR and Methenamine silver stain to investigate the frequency of presence and colonization of *Pneumocystis jirovecii* in 100 BAL specimens of TB positive patients compared to 100 BAL of TB negative (with unknown pulmonary disorders). The mt-Lsu rRNA Gene (Large sub unit of mitochondrial ribonucleic acid) is used to be amplified as target in Nested-PCR. This mitochondrial gene has high identical sensitivity with more specificity and gave more positive results in clinical samples. The sequencing approach is also used to more sub phylogenic studies and *P.jirovecii* confirmation in PCR products. RFLP and CSGE techniques applied on DHPS gene to discriminate eventual sulfa drug resistance. And single-base mismatches. High false positives were reported using GMS staining (13.5%) While using Nested-PCR, 5 positive cases were reported from the tuberculosis group Which is relatively lower than that of people with AIDS. No reports of co- infection were found in TB negative patients Using enzyme AccI in RFLP method all five clinical isolates in the position 165 have mutations, while using HaeIII restriction enzymes, four clinical isolates except IS³ have mutation at position 171 However, by CSGE on DHPS fragments, Heteroduplex bands were able to detect single-base mutation in four isolates mentioned with the exception of the third isolate.