P0045
Poster Session I
How to improve fungal diagnosis

HIGH PREVALENCE OF PNEUMOCYSTIS JIROVECII IN BRONCHOALVEOLAR LAVAGE (BAL) SAMPLES USING A MULTIPLEX-NESTED PCR.

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OBJETIVES: Identify the carriage rate and clinical significance of *Pneumocystis jirovecii* (PJ) using a novel homemade multiplex-nested PCR with an internal positive/inhibition control in BAL samples.

METHODS: From March to November 2013, all BAL samples submitted to the Clinical Microbiology Lab were studied. A total of 112 BAL samples from 111 different patients were processed for DNA extraction using InstaGene Matrix (Bio-Rad). A multiplex-nested PCR was performed based on the method described by Wakefield et al**.** (Lancet. 1991 Jun 8;337) . The universal region 23S rRNA was used as internal control. PCR products were analysed by agarose gel electrophoresis. Direct immunofluorescence (DI) (Bio-Rad) was used as the diagnostic method.

RESULTS: 20 (17.85%) patients were PJ positive, 75 negative, and 16 PCR-inhibited. Only one of the positive patients had a *Pneumocystis jirovecii* pneumonia (PJP) diagnosed by conventional methods (DI). Ten patients were diagnosed of chronic lung disease (3, COPD; 3, silicosis; 2, cystic fibrosis, and 2, asthma). Nine of them were on corticosteroid-treatment. Pulmonary radiographic abnormalities related with PJP were identified in 10 patients, other radiographic anomalies not related with PJP were identified in 6 patients, and in 4 patients none radiographic anomalies were identified, so they were considered to be PJ asymptomatic carriers (9%).

CONCLUSIONS: There is substantial number of patients not diagnosed of PJP by DI, but diagnosed on the basis of clinical or radiologic findings. We found a high rate of PJ infection among patients diagnosed of a chronic lung disease on corticosteroid treatment. Asymptomatic PJ carriers might constitute a reservoir that may lead to acute PJP in a susceptible host. The multiplex-nested PCR approach could be helpful in the microbiology diagnosis of PJ infections.