TITLE: DIFFERENT POPULATIONS OF *PNEUMOCYSTIS JIROVECI* IN BRONCHOALVEOLAR LAVAGE FROM HIV- INDIVIDUALS ASSOCIATED WITH INDICATORS OF IMMUNOSUPPRESSION AND PATHOGENICITY IN UBERABA-MINAS GERAIS - BRAZIL

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ABSTRACT:

Pneumocystis jiroveci cannot be cultured in vitro, and the Polymerase Chain Reaction (PCR) has a greater potential for its detection than standard morphological techniques, especially in cases with low microorganism load such as in human immunocompetent hosts. Some authors showed that Nested PCR was more sensitive in the detection of *P. jiroveci* DNA, while others demonstrated that a single round PCR was the most sensitive. In addition, the Nested PCR should be avoided in the laboratory diagnostic routine since too much manipulation of the sample increases the risk of contamination. In this work, we evaluated the occurrence of P. *jiroveci* in 30 bronchoalveolar lavages (BAL) of HIV and HIVs (HIV-) samples sent to the Department of Surgical Pathology from the Clinic Hospital of the Federal University of Triângulo Mineiro (UFTM), Uberaba, Minas Gerais, Brazil. The data found were associated with clinical manifestations of infected individuals. The samples were centrifuged, stained with toluidine and silver for morphological studies, and diluted v/v with Guanidine-EDTA for PCR. After DNA extraction with phenol-chloroform, the detection of *P. jiroveci* was performed by single-round PCR with LSU rRNA primers PAZ102 and PAZ102-H. Electrophoresis of the amplified products was performed on 6% polyacrylamide gels. Amplification of *P. jiroveci* DNA occurred in 23.3% (7/30) of the samples, which was not evidenced by cytological staining. Three different amplification profiles were observed as single bands in the regions of 346 bp or 400 bp, and double bands of 250 bp and 400 bp. The presence of the fungus in HIV-positive individuals was related to isolated or associated low immune response indicators in 71.4% (5/7) of the cases: 60.0% candidiasis, 20.0% neoplasia, 20.0% use of dexamethasone, and 20.0% alcoholism. Isolated or associated pulmonary alterations (pleural effusion, pneumonia, RX opacity) occurred in 100% (7/7) of the cases; Pulmonary symptoms in 42.8% (3/7); Hematological changes in 42.8% (3/7): 100% (3/3) anemia, 66.7% (2/3) increase in leukocytes, platelet loss, prothrombin and thromboplastin. PCR showed greater sensitivity than the staining methods and variability of genetic profiles suggestive of reinfections or the presence different *P. jiroveci* genotypes in the studied region. However, the presence of immunosuppression and pathogenicity indicators in most of the cases suggests recent reactivation of latent infections.

Keywords: *P. jiroveci*, HIV negative patients, PCR , Pathogenicity

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