MOLECULAR IDENTIFICATION OF *PARACOCCIDIOIDES* SPECIES ISOLATED FROM PATIENTS AND ENVIRONMENT OF THE STATES OF SÃO PAULO AND MINAS GERAIS, BRAZIL.

COCIO, T.A.*1; NASCIMENTO, E.1; VON ZESKA KRESS, M.R.2; BAGAGLI, E.3; MARTINEZ, R.1

¹ FACULDADE DE MEDICINA DE RIBEIRÃO PRETO – USP (FMRP/USP) – DEPRTAMENTO DE CLÍNICA MÉDICA – AV. BANDEIRANTES, 3900 – MONTE ALEGRE - CEP: 14049-900 – RIBEIRÃO PRETO/SP/BRASIL. ² FACULDADE DE CIÊNCIAS FARMACÊUTICAS DE RIBEIRÃO PRETO – USP (FCFRP/USP) - DEPARTAMENTO DE ANÁLISES CLÍNICAS, TOXOCOLÓGICA E BROMATOLÓGICAS – AV. DO CAFÉ, S/N – MONTE ALEGRE - CEP: 14040-020 – RIBEIRÃO PRETO/SP/BRASIL. ³ INSTITUTO DE BIOCIÊNCIAS DE BOTUCATU - UNESP – CAMPUS DE BOTUCATU – DEPARTAMENTO DE MICROBIOLOGIA E IMUNOLOGIA – RUA DR. PLÍNIO PINTO E SILVA, S/N – DISTRITO DE RUBIÃO JÚNIOR - CEP: 18618-691 - BOTUCATU/ SP/ BRASIL.

Paracoccidioidomycosis (PCM) has two etiological agents, Paracoccidioides brasiliensis and P. lutzii. Based on phylogenetic and geographical differences, P. brasiliensis was characterized into four subgroups, S1, PS2, PS3 and PS4. P. lutzii was characterized as a new species by phylogenetic and micromorphological differences existing among the etiological agents of PCM. In this study, clinical and environmental isolates of *Paracoccidioides* spp. were genotyped in order to obtain knowledge of the geographic distribution of their species and subgroups in southeastern Brazil. Isolates were obtained from 14 patients (54% with chronic form and 46% with acute/subacute form), 12 from Ribeirão Preto-São Paulo State (1 with AIDS) and 2 from southern of Minas Gerais State, and from soil (n=1) and armadillos (n=2) of the Ibiá - MG city. To confirm the genus of the clinical and environmental isolates of Paracoccidioides spp., the genomic DNA was submitted to Polymerase Chain Reaction (PCR) for the amplification of the GP43 coding gene. The identification of P. brasiliensis complex the subgroups and P. lutzii species were performed by PCR - RFLP (Polymerase Chain Reaction - Restriction Fragment Length Polymorphism) method with the amplification of the α - Tubulin - coding gene with the double digestion with the restriction endonucleases, Bcll and Mspl. The DNA fragments were checked on 2% agarose gel electrophoresis. The clinical (14) and environmental (3) isolates were identified into the subgroup S1 of the P. brasiliensis complex. The geographical distribution of P. brasiliensis (S1) predominates in the south and southeast of the country. Thus, the results confirmed the prevalence of P. brasiliensis - S1 subgroup in the southeastern region of Brazil, covering environmental and clinical isolates and independently of the clínical form of paracoccidioidomycosis.

Keywords: Paracoccidioides spp., Genotyping, PCR-RFLP

Financial Support: UPDT/PN-DST/AIDS – MINISTÉRIO DA SAÚDE, BRASIL; FUNDAÇÃO DE APOIO AO ENSINO, PESQUISA E ASSISTÊNCIA DO HOSPITAL DAS CLÍNICAS DA FACULDADE DE MEDICINA DE RIBEIRÃO PRETO DA UNIVERSIDADE DE SÃO PAULO - FAEPA E COORDENAÇÃO DE APERFEIÇOAMENTO DE PESSOAL DE NÍVEL SUPERIOR - CAPES.