Giardia duodenalis: AXENIZATION AND CHARACTERIZATION OF THREE ISOLATES FROM BRAZIL, EMPLOYING BIOLOGICAL, BIOCHEMICAL, IMMUNOLOGICAL AND MOLECULAR PARAMETERS

This study reports the isolation, axenization and characterization of three human isolates of Giardia duodenalis from Belo Horizonte, State of Minas Gerais, Brazil. The isolates were axenized from cysts obtained from the feces of symptomatic (BHRA93) and asymptomatic (BHRF92 and BHLF93) patients. The reference strain Portland-1 and one clone of each isolate were included in the study. Biological, immunological, biochemical and molecular parameters were used for characterizing the isolates. The growth curves were very similar for all the isolates in spite of Portland-1 strain having reached the exponential growth phase earlier. All the isolates were able to infect Holtzman neonatal rats and higher numbers of trophozoites in the duodenum were obtained from BHRF92 and Portland-1 isolates. Antigenic differences among the isolates were observed by SDS-PAGE, immunoblotting and indirect immunofluorescent test. Greater antigenic heterogeneity was observed between Portland-1 and Brazilian isolates. Several protein bands ranging from 15 to 200 kDa were identified in the SDS-PAGE. All the isolates induced the production of anti-Giardia specific antibodies in rabbits immunized with antigenic extract of the trophozoites. Reactivity of the anti-sera was greater with the homologous antigen than with the heterologous ones. Portland-1 and BHRA93, both symptomatic isolates induced higher titers of antibodies sera. The isolates were grouped into three zymodemes by means of isoenzyme analysis obtained with malate dehydrogenase (MDH), malic enzyme (ME), phosphoglucomutase (PGM) and glucose phosphate isomerase (GPI) enzymes. BHRA93 and BHRF92 isolates presented the same profile, which differed from the BHLF93 isolate in the mobility of MDH enzyme. The Portland-1 strain represented the third isoenzyme profile. The RAPD analysis divided the isolates into two main groups, one represented by the Portland-1 strain and the other represented by the Brazilian isolates, which, in turn, were divided into two subgroups, the first including BHRA93 and BHLF93 isolates and the second the BHRF92 isolate. The PCR revealed that all the isolates studied, the Brazilian ones and the Portland-1 strain, belong to the genotype A. The data obtained with all the parameters used for characterizing the isolates detected little difference between the parental isolates and the clones.

The results showed a great heterogeneity between the Portland-1 strain and the Brazilian isolates, although the latter presented some individual polymorphism. The degree of similarity or differences presented by Brazilian isolates varied according to the technique used for the analysis. It was not possible to correlate the differences with the clinical characteristics of the isolates.

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