

# Multigene typing of *Giardia Duodenalis* isolated from tuberculosis and non-tuberculosis subjects

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## Abstract

*Giardia duodenalis* is a cryptic protozoan, which has eight assemblages (A-H). Assemblages A and B are the main genotypes reported from humans with probable anthroponotic and zoonotic transmission. The current study aimed to characterize *G. duodenalis* assemblages in tuberculosis (TB) patients and healthy subjects using multilocus genotyping (MLG). Thirty *Giardia*-positive stool samples, which were obtained from TB patients and healthy subjects were included in the study. After total DNA extraction, three  $\beta$ -giardin (*bg*), triosephosphate isomerase (*tpi*), glutamate dehydrogenase (*gdh*) genes were amplified and sequenced. Obtained sequences were compared to the GenBank database to characterize assemblages. Phylogenetic analysis using Maximum Likelihood (ML) and Tamura 3-parameter was performed for each gene. From 30 *Giardia*-positive subjects, 17 (57%) and 13 (43%) were from healthy and TB-infected subjects, respectively. There was no significant co-existence of *Giardia* and tuberculosis ( $P$ -value = 0.051). In addition, 14 (46.7%) and 16 (53.3%) of *Giardia* isolates were from asymptomatic and symptomatic subjects, respectively. PCR amplification was successful in 25 single samples (83.3%) consisted of 20 for *tpi*, 15 for *bg*, and 13 for *gdh* genes. Accordingly, 13/25 (52%) and 8/25 (32%) belonged to assemblage A and assemblages B, respectively, whereas 4/25 (16%) were either assemblage A or B with different genes at the same time. Significant correlation between assemblages and TB, age, and symptoms was not seen. The phylogenetic analyses represented no separation based on TB and gastrointestinal symptoms. Assemblage A was the predominant genotype in samples. The high frequency of assemblage AII indicated importance of anthroponotic transmission of *Giardia* in both healthy and TB patients. In addition, considering the exclusive reports of sub-assemblage AIII in wild ruminants, the presence of AIII in the current study have to be carefully interpreted. The inconsistency between the assemblage results of either *bg* or *gdh* loci with *tpi* gene signifies the insufficiency of single gene analysis and the necessity for MLG in molecular epidemiology of *G. duodenalis*.