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## **ACCEPTED MANUSCRIPT**

# Assessment of nuclear and mitochondrial genes in precise identification and analysis of genetic polymorphisms for the evaluation of *Leishmania* parasites

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#### ABSTRACT

The polymorphism and genetic diversity of *Leishmania* genus has status under discussion depending on many items such as nuclear and/or mitochondrial genes, molecular tools, *Leishmania* species, geographical origin, condition of micro-environment of *Leishmania* parasites and isolation of *Leishmania* from clinical samples, reservoir host and vectors. The genetic variation of *Leishmania* species (*L. major*, *L. tropica*, *L. tarentolae*, *L. mexicana*, *L. infantum*) were analyzed and compared using mitochondrial (COII and *Cyt b*) and nuclear (*nagt*, ITS-rDNA and *HSP70*) genes. The role of each enzymatic (COII, *Cyt b* and *nagt*) or housekeeping (ITS-rDNA, *HSP70*) gene was employed for accurate identification of *Leishmania* parasites. After DNA extractions and amplifying of native, natural and reference strains of *Leishmania* parasites, polymerase chain reaction (PCR) products were sequenced and evaluation of genetic proximity and phylogenetic analysis were performed using MEGA6 and DnaSP5 software. Among the 72 sequences of the five genes, the number of polymorphic sites was significantly lower as compared to the monomorphic sites. Of the 72 sequences, 54 new haplotypes (five genes) of *Leishmania* species were submitted in GenBank (Access number:

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