Online journal ISSN - 1676-5680
GMR Genetics and Molecular Research
EVOLUTION AND TECHNOLOGY

Thesis Abstract

Analysis of minicircle sequences of kDNA obtained from clinical samples (lesions and scars) of patients with American cutaneous leishmaniasis in Pernambuco State, Brazil

Eduardo Henrique Gomes Rodrigues

2006. Pós-Graduação em Biologia Celular e Molecular, Instituto Oswaldo Cruz - IOC, Fundação Oswaldo Cruz - FIOCRUZ, Rio de Janeiro, RJ, Brasil. Ph.D. thesis. 123p. Orienting Professors: Drs. Frederico Guilherme Coutinho Abath and Octavio Fernandes da Silva Filho.

Some species of the genus *Leishmania* are causative agents of leishmaniasis, which is an important public health concern. American cutaneous leishmaniasis (ACL), caused by *Leishmania* (*Viannia*) *braziliensis*, is characterized by cutaneous lesions that heal spontaneously or after specific treatment. In the present study, *in vitro* and *in silico* approaches were performed to analyze the mitochondrial genome. Two hundred and ninety complete *Leishmania* sp kDNA minicircles obtained from clinical samples (lesions and scars) of patients with ACL were studied. The present study demonstrates multiple alignment of minicircle sequences, where size polymorphisms (ranging from 518-797 bp) were present, as well as sequence polymorphisms, indicating significant heterogeneity of classes, particularly in minicircles amplified from active cutaneous lesions. The minicircle sequences obtained from scars were grouped into one cluster, indicating some degree of homogeneity, possibly due to clonal selection. The compositional analysis showed A + T of 70%. Palindromes were mapped and identified in two clones, showing that the variable region of minicircles was richer in A + T than was the conserved region. Direct repeated alternate motifs composed of TA, AT, TT, and AA represented 55% of these sequences. Fifty-five polymorphic microsatellites were mapped in both the conserved and variable regions of the minicircles. The mapping of palindromes, direct alternate repeats and microsatellites was not differentially present, suggesting that they represent common structural features of this molecule. Specific motifs were identified and mapped in the conserved and variable regions of kDNA minicircles, and classified according to frequency as pertaining to scar minicircles or lesion minicircles. For the same motif, the frequency was higher in the scar minicircles. In conclusion, some molecular features seem to be common to all minicircles. On the other hand, the classes of minicircles obtained from scars are more similar, as judged by the multiple alignment and cladogram analyses, possibly due to the predominance of specific motifs. In future

study, it would be interesting to identify precisely the regions responsible for the genetic differences between minicircles obtained from scars and cutaneous lesions. The regions encoding gRNA deserve particular interest, as different types of gRNA may be essential to the survival of *Leishmania* in the biological context of healed tissue or cutaneous lesions. However, the possible biological consequences of our findings for persistent *Leishmania* need further investigations aimed at a better understanding of this complex issue.