Determination of molecular targets in the 18s ribosomal RNA gene for detecting Apicomplexan parasites of medical interest.

Pinheiro, S. N; ANDRADE-NETO, V. F. KRETTLI; Lanza, D.C.F.

Universidade Federal do Rio Grande do Norte, Departamento de Bioquímica, Laboratório de Biologia Molecular Aplicada - LAPLIC

INTRODUCTION: Studies about primer designing in the conserved regions of the eukaryotic 18S RNA gene (18S rDNA) has been effective in approaches, which aimed a distended coverage of species in diverse environments. Nevertheless this gene has a potential for the identification of a restrict number of species that has not been explored yet. Here we make a proposal to allow the identification of Apicomplexa of medical interest, by creating a new strategy which facilitates the diagnosis. **OBJECTIVES**: Develop a molecular system for Apicomplexa identification based on the analysis of specific conserved and variable regions of the 18S RNA gene. **MATERIAL AND METHODS**: Primers targeting the 18S RNA gene were designed flanking specific regions selected by searching for similarity through Blast tool and by alignments from Clustal Ω and Geneious sofltwares. Its specificity and sensibility were tested *in silico* and *in vitro using* AUTO DIMER and Primer BLAST. **RESULTS AND DISCUSSION:** A new set of primers flanking conserved and variable regions of the 18S RNA gene were predicted and designed, and their coverage efficiency was confirmed amplifying different samples containing DNA of some different Apicomplexan genera. Computational predictions appoint that the set of primers developed can identify, specifically, 45 species from eight different Apicomplexan genera. **CONCLUSION:** These primers can be used as a semi nested-PCR, generating 166 amplicons with distinct sequences, that can be used to discriminate 45 Apicomplexan species, demonstrating that is also possible to use the 18SrRNA gene to detect specific protozoan, in a restrictive approach that not aim the screening of a large number of species.

Key Words: Primer designing, Plasmodium, Eukaryotic ssu RNA