SPECIFICITY STUDIES ON SERINE PROTEASES FROM *LEISHMANIA AMAZONENSIS*: SUBSTRATES, INHIBITORS AND DISCOVERY OF NEW ENZYMES

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Leishmaniasis is a disease caused by trypanosomatidae protozoa, endemic in tropical and subtropical regions of the world. The high-cost, toxicity and resistance issues associated with the current treatment turns necessary the discovery of new drugs. Serine proteases (SP) are important targets for immunoprophylaxis and chemotherapy due to their involvement in host-parasite interaction. The goal of this work is improving our knowledge about SP of *L. amazonensis* (LA) to design specific inhibitors. Recently, our group has identified three SP: LSP-I, II and III in LA fractions. We have obtained affinity chromatography fractions of the aqueous- (LAAE) and detergent-soluble cellular extracts (LADE) and from the culture supernatant (LACS) of LA. Zymography indicated gelatin-degrading SP other than LSPs in LADE (75kDa), LAAE (116kDa) and LACS (73 and 80kDa). From gel filtration of LACS we have isolated a new oligopeptidase (120kDa) that was not described in literature yet. All the fractions were active over synthetic substrates carrying arginine in P1. We are now testing substrates with different residues in P1-P4. To find inhibitors for these enzymes we are screening a panel of peptides where the affinity constants for the most active will be analyzed using NMR and enzyme kinetics. These data will provide the design of new peptidomimetics for pursuing the future development of new antileishmanial drugs.

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