Investigation of antimicrobial and anticoagulant effects of trypsin inhibitor from Caesalpinia ferrea var. cearensis

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Abstract—The species belonging to the Fabaceae family are abundant in the Brazilian region. They have great relevance because they are a source of plant proteins, and for its diversity of biomolecules with industrial potential and pharmaceutical applications. Among these are inhibitors, which can act in the regulation of endogenous proteinases and in the defense of plants against the attack of insects and microorganisms. This study reports the purification steps, partial characterization, in vitro effects of CfTI against pathogenic microorganisms, and hemostasis tests. Caesalpinia ferrea Trypsin Inhibitor (CfTI) was purified and partially characterized by standardized protocols for other inhibitors. Total protein was determined according to Bradford and the values were 4.7 mg/mL and 1.4 mg/mL in the crude extract and fraction eluted from affinity chromatography, respectively. CfTI reduced 96% on trypsin activity at 0.25 μ g but did not inhibit chymotrypsin. Additionally, the inhibitor kept 85% of its activity up to 60 °C and about 90% in pH from 2 to 9. The electrophoresis on SDS-PAGE revealed only one band with molecular mass of approximately 18 kDa. CfTI prolonged aPTT up to 2.45 times, suggesting in vitro anticoagulant effect. CfTI was also tested with strain of pathogenic microorganisms, including bacteria and yeast, however, there was no growth inhibition. The data suggests that CfTI belongs to the Kunitz family with potential anticoagulant effect.

Keywords-kunitz inhibitor; trypsin; blood coagulation