Mechanistic Insights Into Anti-angiogenic Activity of *Trypanosoma cruzi* Protein 21 and Its Potential Impact on Chagasic Cardiomyopathy Onset

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Introduction: Chagas disease, caused by the parasite *Trypanosoma cruzi*, is an important cause of Chronic Chagasic Cardiomyopathy (CCC). The prospection of innovative therapeutic agents against CCC is a major task. The recombinant form of P21 (rP21), a secreted *T. cruzi* protein involved in host cell invasion and on progression of chronic inflammatory processes have been studied as a potential novel therapeutic target. **Objectives:** Our present work aimed to verify the impact of rP21 on blood vessels formation in vitro and investigate the antiangiogenic activity triggered by rP21. **Material and Methods:** tEnd cells were treated with different concentrations of rP21 or bacterial extract and viability and cellular adhesion were evaluated by MTT and angiogenesis inhibition by Matrigel tube formation assay. To verify the rP21-proteolytic activity on ECM components, fibrinogen, matrigel and fibronectin was incubated with rP21 or not. The substrate digestion was analyzed by SDS-PAGE. The accumulation and distribution of F-actin was determined by Phalloidin staining using ImageJ software. tEnd cells were incubated with rP21 and the total RNA was extracted using RiboZol and cDNA was synthesized by cDNA Reverse Transcription kit and analyzed by real-time PCR. **Results and Discussion:** We observed that rP21 did not alter cell viability and adhesion, but strongly inhibited vessel formation. Tube formation assay showed that angiogenesis inhibition was dependent of the CXCR4-rP21 binding. Moreover, we found that rP21 significantly increased F-actin levels and this protein was able to modulate expression of genes related to angiogenesis and actin cytoskeleton. However, rP21 showed no significant activity on the matrix components. Thus, the anti-angiogenic activity of rP21 may be due to a cascade of events triggered by CXCR4-rP21 interaction. **Conclusion:** Angiogenesis inhibition by rP21 depends on its binding to CXCR4 receptor and on the modulation of gene expression of both cytoskeleton and anti-angiogenic molecules.

Key words: *Trypanosoma cruzi*; CCC; angiogenesis; actin cytoskeleton; rP21; CXCR4 receptor; endothelial cell

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