## From vessel to tissue: the travels of *Trypanosoma cruzi*

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The protozoan *T. cruzi* is the etiological agent of Chagas' disease of major medical significance throughout South to Central America. The disease was discovered by Carlos Chagas 100 years ago. In 1909, the Brazilian physician described the parasite in the gut of an insect belonging to the Reduviidae family. This discovery is one of the few examples in the literature where the disease, the parasite, the insect vector and the cell cycle were described in a single manuscript. Modern description of the complex dixenic life cycle of the parasite is basically the same, as originally published. Different isolates from *T. cruzi* have demonstrated that the parasite populations are extremely polymorphic. This polymorphism has been correlated with the pathology observed in different clinical forms and distinct geographic areas. Indeed, isoenzyme and riboprinting analyses, rRNA promoter activity, mini-exon gene sequencing and microsatellite markers have provided evidence that *T. cruzi* is very heterogeneous. This polymorphism has been correlated with the pathology observed in different clinical forms and distinct geographic areas: in Brazil, the asymptomatic form predominates (60-70%), followed by the cardiac (20-30%) and digestive forms (8-10%); in Chile, the digestive form predominates, whereas in Argentina digestive symptoms correspond to only 3.5% of the total chagasic patients. These differences are due to genetic variations of the parasite and to physiological and genetic conditions of the host, but the contribution of each is a matter of debate. Whichever are the differences among clones and hosts in the development of the disease in humans it should be stressed that *T. cruzi* must express at the surface ligands that recognize specific host receptors allowing adhesion to the extracellular matrix macromolecules and, finally, to cells. In the beginning of the 1980s our laboratory described the existence of a group of closely related glycoproteins, originally called Tc-85, which were specifically expressed at the surface of the infective trypomastigote stage. Monoclonal antibodies against these glycoproteins were able to inhibit partially the interiorization of parasites into host cells. Further studies by several groups detected a gene superfamily of which trans-sialidase is a member. This group was called the gp85/trans-sialidase gene superfamily. The existence of this family was entirely demonstrated by the TriTryp effort showing the existence of approximately 700 genes and similar number of pseudogenes with high degree of homology, performing 1-2% of the parasite genome. These genes express the corresponding glycoproteins at the parasite surface with a half-life of 3.5 hours after which they are shed in membrane vesicles. One of the cloned members of the gp85/trans-sialidase superfamily has an affinity to laminin, but not to gelatin or fibronectin and it was found that the binding site is mainly located at the N-terminal sequence. Another clone produced a protein with higher affinity for fibronectin in comparison to laminin. An independent approach using the SELEX method provided RNA aptamers with strong affinity to trypomastigotes that were displaced by fibronectin, laminin, heparan sulfate and thrombospondin, respectively, showing expression of ligands with specific receptor affinities on the surface of the parasite. These aptamers, notably the one displaced by laminin, were able to inhibit partially the entrance of trypomastigotes into epithelial cells. The whole superfamily possesses a constant peptide sequence that is invariant among members, located at the carboxyterminus, VTV*X*NVFLYNR, which is called FLY domain, for short. This peptide binds to epithelial cells in a saturable manner. An attempt to identify the receptor of that sequence on the cell surface led to cytokeratin-18 (CK-18), although its existence on the membrane is debated. The binding of the peptide promotes dephosphorylation of CK-18 that leads to cytoskeleton reorganization and activation of the ERK1/2 signaling cascade. As a result, there is an increase of parasite entry into epithelial cells. From the site of insect bite where parasites reside almost undetected by the immune system (stealth parasites) until reaching lymphonodes or the arterial circulation, the trypanosomes will eventually have to cross the arterial walls and fall into the extracellular matrix. *T. cruzi* invades almost any cell, including experimentally enucleated cells, but does not invade red blood cells. This promiscuity needs, at least, ligands with different specificities and equally specific receptors.

If one imagines a sequential series of bindings and detachments it may require also, as one of the possible mechanisms, hydrolysis by protein and carbohydrate hydrolases. Thus, the parasite could be freed of the successive haptotactic interactions, allowing its migration through the basal lamina and the extracellular milieu. The literature is rich in data involving many molecules at the trypomastigote surface that bind to fibronectin, laminin, heparan sulfate, heparin, thrombospondin-1, collagen, sialic acid, TGFß and bradikynin receptors, among others. Certainly, *T. cruzi* developed many different paths to progress inside a vertebrate to reach the host cells of the target tissues. Financial support: FAPESP and CNPq