

Stage-specific surface antigens during the morphogenesis of Trypanosoma cruzi : developmentally regulated expression of a glycosyl-phosphatidylinositol anchored glycoprotein of amastigotes

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During its life cycle, T.\_cruzi assumes a variety of differentiation forms, each displaying specific antigenic polypeptides (1,2). Slender trypomastigotes, from blood or cell culture, undergo extracellularly morphological rearrangements in which the parasites become gradually broader and transform into amastigotes. By scanning electron microscopy we have observed a progressive internalization of the flagellum and reorganization of the cell shape in a helical fashion, in parasites undergoing transformation. After 48 hours of extracellular incubation the parasite population consisted exclusively of amastigotes with a short protruding flagellum. The morphological changes were associated with the expression of different surface antigens defined by monoclonal antibodies : the trypomastigote-specific antigens Ssp-1 (a 100-150 KDa glycoprotein), Ssp-2 (a 70 KDa glycoprotein), Ssp-3 (undefined), and Ssp-4, an amastigote-specific surface antigen (3).

Ssp-4 is an acidic, mannose containing 70-84 KDa glycoprotein which homogeneously covers the whole surface of amastigotes in a manner suggestive of a densely packed coat. It is expressed in maximal amounts by recently transformed amastigotes, being progressively shed during their intra- or extracellular development.

Ssp-4 can be metabolically labeled with myristic acid and is converted into a hydrophilic form by treatment with the

glycan-specific phospholipase C of T. brucei (4), or following lysis of the parasites in non-ionic detergents. The hydrophilic form of Ssp-4 is recognized by antibodies to the cross-reactive determinant (CRD) of the variant surface glycoprotein of African trypanosomes. This property, together with the data mentioned above, indicates that Ssp-4 is anchored to the membrane by a glycosyl-phosphatidylinositol (GPI) moiety.

We were able to demonstrate that T. cruzi contains an endogenous phospholipase C and that most of the Ssp-4 shed during amastigote development is hydrophilic, does not contain myristic acid and reacts with anti-CRD. These observations provide strong evidence that phospholipase C mediates the release of this GPI-anchored glycoprotein under physiological conditions, as the parasite undergoes differentiation (5).

Since amastigotes expressing Ssp-4 are present in the bloodstream of infected mice (3), there is a possibility that soluble Ssp-4 is also being generated in vivo. Sera from chagasic patients was shown, by immunoprecipitation and immuno-radiometric assays, to contain antibodies to Ssp-4.

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