MITOSIS IN TRYPANOSOMA CRUZI: STUDIES OF THE INVOLVEMENT OF COHESIN, HELICASES, POLO-LIKE KINASE AND PROTEASOME

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Mitosis in *Trypanosoma cruzi* is characterized by unusual features: chromatin fibers do not form visible chromosomes and the nuclear membrane persists during the process. Since few are known about the functions of structural and regulatory proteins in trypanosomatids, we have started our studies by identifying and cloning the corresponding genes in *T. cruzi*. In this way, we have cloned the four subunits of the cohesin complex that maintains chromatid cohesion from chromosome duplication until anaphase. The subunit TcSCC1 was expressed in *E. coli* and polyclonal antibody against it was obtained. Western blot analysis of unsyncronized and hydroxyurea syncronized cultures of epimastigotes showed that this cohesin subunit is more abundant after 2-4 hour of hydroxyurea removal and is involved in mitosis. Two smaller bands were also identified, suggesting that TcSCC1 is being cleaved by separase which is indirectly regulated by proteasome. The regulatory enzyme TcECO1, an acetylase required for cohesion establishment, lacks the zinc finger domain, suggesting a different function in this family. Two homologues in *T. cruzi* of the yeast unique CHL1p, a helicase that binds to Eco1, were cloned and expressed in *E. coli*, as well as TcECO1 and TcPLK, a polo-like kinase involved in the exit of the majority of cohesin from the chromosomes in the transition prometaphase-metaphase.