CHIMERIC PROTEINS OF THE TRIOSEPHOSPHATE ISOMERASE: EFFECT OF SULFHYDRYL AGENTS ON THE ACTIVITY OF CHIMERAS OF TRIOSEPHOSPHATE ISOMERASE FROM TWO RELATED PARASITES.

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Trypanosoma cruzi and *T. brucei* are two related parasites that cause Chagas disease and Sleeping Sickness in South America and Africa, respectively. Homodimeric triosephosphate isomerases (TIM) from *T. cruzi* (TcTIM) and *T. brucei* (TbTIM) have 73% of identity in their amino acid sequence and have remarkably similar X-ray structures. In both TIMs, an interface cysteine (Cys14) is surrounded by loop 3 of the adjacent subunit. Nonetheless, the reactivity of Cys 14 of TcTIM to thiol reagents like methylmethane thiosulfonate (MMTS) and dithionitrobenzoic acid (DTNB) is 70-fold higher than that of Cys 14 of TbTIM. To explore what causes that difference, we divided the aminoacid sequence of TIM into eight regions, including one beta sheet, the adjoining loop and the alpha helix, and constructed and characterized kinetically six chimeric proteins with different regions of TcTIM and TbTIM. Exploring the susceptibility of these enzymes to the action of MMTS and DTNB, we found that a chimera that possesses regions 1-4 of TcTIM (residues 1-119) and regions 5-8 of TbTIM (residues 120-250) showed a similar behavior to TcTIM, however, a chimera with regions 1-3 of TcTIM (residues 1-87) and regions 4-8 of TbTIM (residues 88-250) showed a similar pattern of susceptibility to TbTIM. This last chimeric enzyme showed an important decrease of activity at concentrations of 20 μM of both MMTS and DTNB. Thus, by focusing our study on the differences between TbTIM and TcTIM in region 4 (residues 88-119), we may know what causes the marked differences against sulfhydryl agents between these two TIMs.