

New Constructed PstS-1(285-374):CFP10 Fusion Protein to detect *Mycobacterium tuberculosis* Infection

Leonardo S de Araujo, Fernanda C Q Mello, Nidai B M da Silva, Janaina A M Leung, Silvia M A Machado, Isabela G Sardella, Scarlet T M Mota, Iza C A Pina, Theolis C B Bessa, and Maria Helena F Saad

Laboratory of Cellular Microbiology, Oswaldo Cruz Institute, Fiocruz, Federal University of Rio de Janeiro, Clementino Fraga Filho Hospital, Rio de Janeiro, RJ; Gonçalo Moniz Research Center - Fiocruz/Salvador, BA, Brazil

The PstS1 antigen is highly immunogenic, especially when combined with CFP10 during both latent and active TB infection. In the present study, a selected *pstS1* peptide gene was cloned, fused with CFP10, and expressed in *Escherichia coli*. The cloned peptide is described to induce better proliferation *in vitro* in TB infection. The product [PstS-1(285-374):CFP10] was compared with the recombinant fused RD1 (region of deletion 1) protein (ESAT-6:CFP10) in detecting *Mycobacterium tuberculosis* infection in 108 recent contacts (rCt) of pulmonary tuberculosis (TB) cases from Rio de Janeiro, considering a positive tuberculin skin test (TST) to be the baseline. All contacts were clinically followed for up to 1 year, and 87% of the tuberculin skin test-positive (TSTpositive) patients accepted preventative treatment. Additionally, a comparative analysis was performed in a pilot group of 9 TB patients and 13 rCt from a different TB endemic area, Salvador, Bahia. For these individuals the result of a commercial IGRA (Quantiferon Gold In-Tube®, QFT) was positive for all TB and in 4/13 rCt. The release of gamma interferon (IFN- γ) in 22-h whole-blood stimulation assays primed with each antigen was determined. Results showed that contact-TSTpositive detection was slightly increased (23/54) when comparing the IFN- γ response to PstS-1(285-374):CFP10 in the 22-h assay to the level seen with the RD1 protein (18/54). Two of 12 untreated TSTpositive contacts progressed to active TB and were concordantly positive in all assays. In the TSTnegative group, similarly lower numbers (<5/48) of responders were achieved for both antigens. Of 7/9 TB and 5/13 rCt responded to PstS-1(285-374):CFP10, of which 2 rCt were QFT+ and 3 QFT-. Our results show that PstS-1(285-374):CFP10 slightly increased recent TB contact positivity and detected active disease progression, suggesting its potential application as a TB infection marker, adding positivity to QFT.

Key words: IGRA, IFN- γ , LTBI, *M. tuberculosis*, PstS-1(285-374):CFP10
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