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Title: Intracellular ESAT-6: A new biomarker of mycobacterium tuberculosis infection?

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Body: Introduction: Early secreted antigenic target 6 (ESAT-6) is a virulent factor of Mycobacterium tuberculosis (M.TB.) infection. Thus, the identification of intracellular (i/c) ESAT-6 antigen in host cells would be a direct marker of TB infection. Recently, we have developed a method to detect i/cESAT-6 by flow cytometry. The aim of this study is to investigate the expression of i/c ESAT-6 in the host cells of individuals with TB infection. Methods: The expression of i/cESAT-6 was examined in the blood of 41 active TB patients, in 33 close contacts with an index TB case and in 43 medically immunosuppressed patients under treatment with biological agents. Furthermore, i/cESAT-6 was tested in the sputum smear of 12 active TB patients. Results: The i/c ESAT-6 was positively detected in the blood of 90% of active TB patients, in 57% of close contacts and in 49% of immunosuppressed patients. In the 12 culture positive TB sputum specimens, sputum smear was Ziehl-Nielsen positive in 8 out of 12 and negative in 4 out of 12, while i/cESAT-6 in sputum cells was detected in all cases. Conclusion: In this study we showed that i/cESAT-6 is detected in the peripheral blood mononuclear cells and in sputum cells of individuals with TB infection. As this is a direct marker of M.TB. infection, further studies are needed in order to define exactly its diagnostic performance in TB infection and its discriminatory capacity between immunological memory and true infection.