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Title: Latent tuberculosis infection is associated with increased Tregs frequencies in the BAL of healthy contact persons

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Body: Background Only a minority of individuals exposed to Mycobacterium tuberculosis (MTB) develops a positive Interferon- γ release assay (IGRA) from peripheral blood mononuclear cells (PBMC). Most contact persons do not develop a systemic immune response after inhalation of MTB aerosols. Pulmonary immune mechanisms preventing a systemic immune response in these subjects are incompletely understood. Objective To evaluate local and systemic immune responses in healthy MTB contacts. Methods Recruitment of MTB exposed health care workers (HCWs) and very close private house hold contacts (HHCs). Flow cytometry of bronchoalveolar lavage (BAL) cells and PBMC for immunophenotyping. IGRA testing of blood and BAL cells. Results 35 HCWs and 15 HHCs were recruited. Regulatory Tregs (CD4+CD25+CD127-) frequencies were increased in all contacts with latent MTB infection (PBMC IGRA positive; n=15) compared to contacts with a negative PBMC IGRA (n=25) with a median 2.12%, IQR 1.63-3.01 versus 0.68%; 0.32-0.96, respectively (p<0.0001). No immunophenotypic differences were seen in PBMC between IGRA positive and IGRA negative subjects (9.6%; 5.9-10.1 versus 7.7%; 4.6-11.3; p=0.47). In 5 of the 25 IGRA negative subjects, the BAL IGRA gave a positive result, possibly indicating incipient tuberculosis. Conclusion In close MTB contacts with LTBI, Tregs are increased in BAL but not systemically when compared to contacts that remain IGRA negative. The functional role of Tregs requires further investigation.