280. Risk assessment for tuberculosis

P2649

Within-subject variability of tuberculosis immune responses in health care workers

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Background: Although IFN-γ release assays (IGRAs) are increasingly used for periodic tuberculosis (TB) screening of health care workers (HCWs), data regarding the interpretation of IGRA results in serial testing is scare.
Objective: To evaluate the within-subject variability of two commercial IGRAs, QuantiFERON®-TB Gold In-Tube (QFT) and T-SPOT®.*TB* (T-SPOT).
Methods: Thirty-four immunocompetent German HCWs (age 42±10 yrs, 79% female) without recent TB exposure or other individual risk factors were repeatedly tested with both IGRAs in weekly intervals over a four week period.
Results: According to the manufacturers' predefined dichotomous cut-offs 10 (29.4%) and 3 (8.8%) of the 34 subjects had divergent overall trends with the QFT and the T-SPOT, respectively ($p<0.001$). The QFT showed 4 unstable conversions (11.8%) and 6 reversions (17.6%), while the T-SPOT showed no conversion and 3 reversions (8.8%). The proportion of concordantly negative IGRA results increased over time. Changes of ±70.4% (QFT) and ±31.2% (T-SPOT) from the mean log IFN-γ response accounted for 95% of the within-subject variability, respectively.

IGRA positivity, discordance and agreement

Visit (day)	Subjects n	Positive QFT n (%)	Positive T-SPOT n (%)	Discordant IGRAs n (%)	Agreement Kappa
1 (0)	34	11 (32.4)	9 (26.5)	8 (23.5)	0.44
2 (7)	34	11 (32.4)	9 (26.5)	6 (17.6)	0.58
3 (14)	29	6 (17.6)	6 (17.6)	2 (5.9)	0.79
4 (21)	30	4 (11.8)	4 (11.8)	2 (5.9)	0.71
5 (28)	26	4 (11.8)	3 (8.8)	1 (2.9)	0.84
Total	153	36 (21.2)	31 (18.2)	19 (12.4)	0.64

Conclusions: We observed considerable variability with both IGRAs and a tendency towards regression of IGRA results over a 4-week period, which should be considered when interpreting repeated IGRA results.

P2650

Conversion and reversion rates in serial examination in German healthcare worker with the interferon-gamma release assay

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Background: Data concerning conversion and reversion rates in serial testing of healthcare workers (HCWs) is rare. There is no consensus to date on how to define and interpret IGRA conversions and reversions. We, therefore analyzed conversion and reversion rates when conducting serial testing of HCWs.
Methods: The study population comprises 426 HCWs from German hospitals, all of whom participated in routine occupational safety and health tuberculosis (TB) screening between January 2007 and October 2010. The QuantiFERON-TB® Gold In-Tube (QFT) assay was used. Different definitions for conversion und reversion were used and risk- related rates calculated.
Results: The first and second QFTs were positive in 8.7% (n=37) of the HCWs,

each. The highest conversion and reversion rates of 3.8% (n=16) and 43.4% (n=23) respectively were observed with the least stringent definition of negative to positive. An uncertainty zone of <0.2 to >0.7 IU/ml gave the lowest conversion rate of 0.6% and a reversion rate of 19.4%. With regarded to the different workplaces, we observed a conversion rate of 5.8% in the high risk group, of 4.1% in the moderate risk group and 1.4% in the low risk group from. The highest reversion rate was found in the low risk group (8.5%). However, the differences were not statistically significant.
Conclusion: Our data suggests that HCWs working on wards with a high risk of TB-infection have higher rates of conversions compared to the low risk group and the use of an uncertainty zone of <0.2 to >0.7 IU/ml around the cut- off when conducting serial testing with the QFT.

P2651
Specificity and negative predictive value of the QuantiFERON-TB®-Gold In Tube testing on trainees in healthcare settings
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Evidence is growing of high sensitivity and specificity of the interferon-gamma release assays (IGRA) for the detection of latent TB infection. A further question is that of the reliability of their negative results. A few studies have evaluated the outcome of Healthcare workers and trainees with negative IGRA results over time. We analysed the specificity and the negative predictive value (NVP) of the Quantiferon-Gold In-Tube (QFT) in trainees in healthcare settings.
Methods: A cohort of trainees at the Vivantes healthcare training institute in Berlin established between October 2008 and July 2010 was tested with the QFT at the beginning and after the first year of training.
Results: The study population comprised 194 trainees. Two trainees were excluded from the specificity analysis: one due to indeterminate QFT-IT result and one following a history of TB. Of the 192 trainees at baseline, 153 were still in training. At the beginning of training two trainees were QFT positive (1%). The specificity was estimated as 99%. 151 trainees showed persistently negative QFT results. One trainee was QFT positive during the follow-up. One conversion occurred in one case and one reversion occurred. The NVP in this healthy trainee group was 99.3%.
Conclusion: Our data confirms the findings of a recent meta-analysis (1). This analysis determined a specificity of 99.4% in individuals belonging to low risk groups. The high specificity and the good NPV support the use of QFT for serial testing of HCWs.
Reference:
[1] Diel R, et al. Interferon-γ release assays for the diagnosis of latent Mycobacterium tuberculosis infection: a systematic review and meta-analysis Eur Respir J 2011 37:88-99.

P2652
Prevalence of LTBI using IGRA and TST in a cohort of health professional trainees from India
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Objectives: Estimate the prevalence of LTBI among health professional trainees at a referral hospital in India, using Tuberculin skin test (TST) and Quantiferon TB Gold In-tube, (QFT).
Methods: From November 2009 to February 2011, students in health professional programs (except medical and nursing students) were approached for consent. In addition to a detailed questionnaire on TB exposure, participants underwent TST (10 mm) and the QFT-GIT (0.35 IU/ml).
Results: 164 students completed testing. Mean age was 21.5 yrs, 48.8% were female and 59.15% had BCG. Mean time in health care was 11.5 months, and 21.6% recalled contact with PTB cases. Seventy-nine (48.2%, 95%CI: 40.3-56.1%) were positive by TST, and 38 (23.2%, 95% CI: 16.9-30.4%) were positive by QFT. In a cohort of nursing students from the same institution, prevalence was estimated at 40.3% (TST) and 17.04% (QFT), thus lower than the health professional cohort. Possible explanations include a higher proportion of male students compared with nursing, also present cohort were less likely to own a car or house as compared with nursing students, suggesting lower SES. Multivariate logistic regression showed age was associated with QFT positivity but not TST (OR=1.24, 95%CI: 1.01-1.52). Length of time in health care, family income, direct contact with TB all showed no association with either test. Participating in sputum collection and/or processing showed a trend towards QFT positivity (but not TST positivity), although this did not reach statistical significance (OR=1.1 (95%CI: 0.89-1.4).
Conclusions: LTBI is common among health professional trainees in India, however, risk factors appear to be better correlated with QFT.

P2653
Evaluation of latent tuberculosis infection in health care workers by QuantiFERON-TB Gold test and tuberculin skin test
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Introduction: It is estimated one third of the world population is infected by mycobacterium tuberculosis (TB) and the risk factor of TB infection is contacting with TB patients.
Aim: The aim of this study was to evaluate the parameters affecting the tuberculin skin test and QuantiFERON-TB Gold tests and the results of both tests used in health care workers for confirming latent tuberculosis infection (LTBI).
Method: 94 health care workers who work in Yedikule Chest Diseases and Surgery Hosital participated to this study.The demographic characteristics of cases, working hours and the number of BCG scar were recorded.Tuberculin skin test were applied to all patients and QuantiFERON levels were measured
Results: The TST and QTF-G positivities had similar dispersion in gender groups and work types.There were a statistically significant relation between the levels of QTF with work times and ages of all participants. It was determined that there was a statistically significant relationship between positivity of QTF with TST psitivities and TST enduration diameters.
Conclusion: In this study the findings indicated that QTF is very usefull in determining and following LTBI as a confirming test for healthcare workers who have close and longterm contact with tuberculosis patients and whose TST tests were positive.

P2654
Serial testing using IGRAs in a cohort of Indian nursing students
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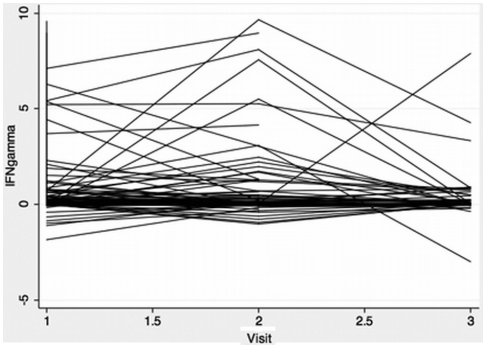
Objectives: Evaluate how continuous Interferon gamma response varies across and within individuals over time in nursing students at a tertiary care hospital in Southern India upon annual screening for LTBI using QFT-GIT.
Methods: Students were approached to participate in a longitudinal study. In addition to history, clinical log books provided detailed information on potential TB exposure prior to baseline, and between annual testing. Students underwent QFT-GIT testing in 2008, 2009 and 2010.
Results: 125 nursing students completed QFT testing at 2-3 points for a total of 311 observations. We present a trajectory plot of continuous IFN-gamma across time for each participant.



There exists a high degree of correlation among observations from the same individual (ICC=0.55). To account for the correlation within individuals over time, we used linear mixed models to estimate a subject specific random effects model. We found IFN-gamma results to be negatively associated with successive visits (-0.09) suggesting responses may decrease over time within individuals, independant of exposure, although this effect was small and not statistically significant. We also identified a small random effect for student (variance = 1.76) after accounting for known LTBI risk factors and TB exposure, suggesting there may be unknown factors contributing to differences in baseline IFN-gamma response across students.

P2655
Tuberculosis screening program using the QuantiFERON-TB Gold test and chest computed tomography for healthcare workers accidentally exposed to patients with tuberculosis
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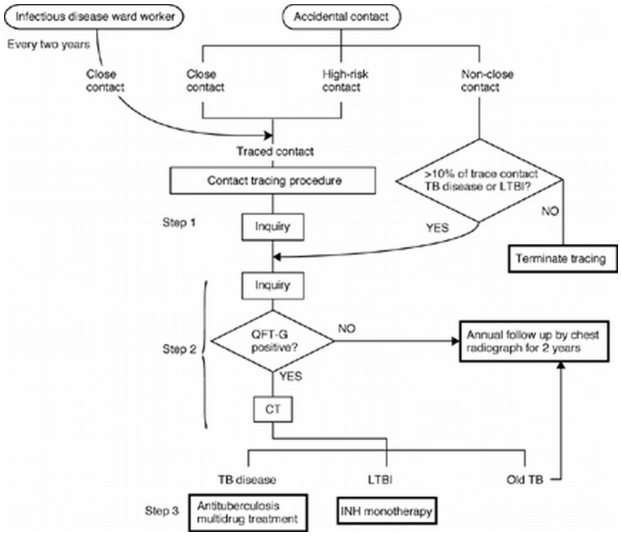
Health-care associated transmission of TB is a serious issue. Health care workers (HCWs) have been reported to show high incidences of TB. Periodical screenings and as-needed screenings for HCWs are important. We integrated chest CTs and

the QFT-G test in our TB screening program for HCWs. First, contacts were tested using the QFT-G test. Second, the HCWs positive for the QFT-G test were investigated by CT and classified as having TB disease, LTBI, or old TB. Finally, the HCWs with TB disease were treated with a multi-drug regimen, while HCWs with LTBI were treated with INH monotherapy.



Between April 2005 and April 2010, 11 patients who had not been diagnosed with TB disease were found to have TB disease during hospitalization. A total of 512 close contacts and high-risk contacts were identified, who underwent the TB screening program. Out of them, 34 (6.64%) showed positive result for the QFT-G test, whereas 478 (93.36%) showed negative results. Of the 34 QFT-G positive HCWs, 4 had CT findings compatible with TB disease and received multi-drug treatment. The chest CT for 24 showed no findings suggestive of TB disease; these HCWs received INH for 6 months. All HCWs who received treatment completed their regimens without any adverse effects. The TB screening program integrating CT and the QFT-G test was safe and feasible (*J Hosp Infect* in press).

P2656

The additional diagnostic value of interferon gamma release assay to the tuberculin skin test in Greek health care workers (a preliminary study)

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Introduction: Health care workers (HCWs) are at increased risk of Mycobacterium tuberculosis infection. The tuberculin skin test (TST) can be positive (among others) after vaccination with bacille Calmette-Guérin (BCG) creating confusion in countries where this is still compulsory, like Greece. Interferon gamma release assays (IGRAs) are used to solve this problem since they are more specific than TST and usually do not become positive after BCG vaccination.
Aims and objectives: This is a preliminary study aiming to evaluate the additional diagnostic value of Quanti-FERON®-TB Gold In-Tube (QFT-GIT) to the tuberculin skin test (TST) in Greek HCWs.
Methods: A cross-sectional study of TST and QFT-GIT was carried out among 275 immuno-competent HCWs at a 900-bed Greek tertiary referral University Hospital. TST was performed on all participants, while QFT-GIT was performed in all subjects with TST $\geq$10 mm.
Results: Among the 275 study subjects, 124 (45.1%) had TST indurations of $\geq$10 mm, 96 of them were BCG vaccinated. None had clinical or radiologic evidence of active tuberculosis. From these 124 HCWs, 69 (age 20-67, mean 48,2) agreed to have the QFT-GIT test which was positive in only 9/69 (13%).
Conclusion: Based on IGRA's results, in the majority of Greek HCWs of our study, positive TST was not due to latent tuberculosis infection but to previous BCG vaccination or other unidentified reasons, like exposure to non-tuberculous mycobacteria. This is in agreement with a previous study of our Department in a younger age group of Greek Army recruits (S. Katsenos et al. Int J Tuberc Lung Dis 14(5):545–550).

P2657

Concordance between IGRA and TST in a cohort of health professional trainees from India

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Objectives: To estimate the concordance between the Tuberculin skin test (TST) and Quantiferon TB Gold In-tube, (QFT) among health professional trainees at a referral hospital in India, and to evaluate risk factors associated with discordant results.
Methods: From November 2009 to February 2011, students registered in various health professional programs (with the exception of medical and nursing students) were approached to participate. In addition to a questionnaire on TB exposure, participants underwent TST (10 mm cutoff) and the QFT-GIT (0.35 IU/ml cut off).
Results: 164 students completed. Mean age was 21.5 yrs (Range: 17-34), 48.8% were female and 59.15% had BCG scars. Mean time in health care was 11.5 months, and 21.6% recalled direct contact with PTB. Prevalence of LTBI by TST or QFT was 48.2% and 23.2% respectively. Agreement between tests was 71.3% (kappa=0.415). The predominant discordance was TST +/QFT- (43/164, 26.2%). Using multivariate logistic regression we evaluated whether discordant results were associated with any particular risk factors, including: age, sex, education, family income, time in health care setting, days spent in high risk wards, performing high risk procedures, pre-existing medical illness, BCG, and known TB exposure. No factors were associated with discordant results, however, age was associated with concordant positives (OR=1.27, 95%CI: 1.03-1.59), and higher family income was protective (OR=0.67, 95%CI:0.45-0.99).
Conclusions: There was fair to weak agreement between TST and QFT in this population. Concordant positives were associated with older age, and lower family income. Discordant results were not associated with any known risk factors.

P2658

IGRA testing correlation with clinical and laboratory parameters in patients with tuberculosis

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Background: IGRA test (Quantiferon-TB-Gold in-tube test, further referred to as QFT) is based on ELISA method for interferon gamma evaluation created by antigen specific T-lymfocytes. QFT has become a part of complex tuberculosis diagnosis.
Aims and objectives: QFT outcomes comparison with other diagnostic methods in patients with tuberculosis and assessment of malnutrition and immunity system influence on QFT outcomes.
Methods: In a group of 62 patients treated in our clinic for tuberculosis in 2010, we compared the QFT results with tuberculin skin test (TST), PCR, smear and culture. The effect of malnutrition and lymfopenia on possible false QFT negativity was studied as well. Non-parametric Mann-Whitney rank test was used for comparing the mean TST value with QFT and in other parameters, Chi-square test was used in frequency tables.
Results: Statistically significant correlation between QFT positivity and TST magnitude, PCR positivity and culture positivity was demonstrated. Statistical significance has not been proven neither between QFT outcomes and microscopic diagnosis nor the effect of malnutrition or lymfopenia on possible false QFT negativity was proven.

P2659

Useful of Quantiferon G in managing solitary pulmonary nodule of less than 3 cm

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Introduction: Development of new tools for the rapid diagnosis of tuberculosis is a priority. There are few data from high burden countries on the utility of Quantiferon G for the rapid diagnosis of solitary pulmonary nodule.
Material and methods: We evaluated the utility of Quantiferon G in 30 patients with solitary pulmonary nodule which was less than 3 cm. All were Thai people, and all got BCG vaccination. Bronchoscopy with Autofluorescense was done, bronchial washing was sent for AFB stain, TB culture, PCR-TB and cytology. Tuberculin test was also performed.
Results: Tuberculosis test was positive in 16 patients (53%), Quantiferon G was positive in 19 patients (63%)and PCR-TB was positive in 2 patients (6.7%). Those with positive Quantiferon G were treated with anti tuberculosis drugs for 9 months, all showed satisfactory response. Among those with negative Quantiferon G,1 year follow up was provided; one turned to be Mycobacterium avium intracellulare, another one developed adenocarcinoma.
Conclusion: Quantiferon G may provide additional useful tool in making decision in managing solitary pulmonary nodule in countries with high incidence of tuberculosis.

P2660

T spot TB – Changing trends and resource implications for trusts

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Introduction: The diagnosis of TB has been revolutionised by Gamma interferon testing with the advantage of being more specific and sensitive in detecting both active and latent TB. East Kent Hospitals NHS Trust ia a lage trust serving a population of 700,000. This study outlines our experience and in particular focuses on the resource implications as usage of biological agents increases.
Methods: All samples assayed over a twelve month period were included in the study. Data, including the demographics of the patients, indication for the test and the result, was collated. The data was analysed.
Results: A total of 491 samples were sent for analysis within the study period. There was an equal gender distribution (M:F – 221:270) and the median age was 53.02.

Table 1. Reason for requesting T spot T

TB dagnosis	TB Screening	Pre -biologics	Reson not specified	Total
73	77	156	185	491
14.87%	15.68%	31.77%	37.68%	100%

Table 2. Result of T spot T by indication

Indication	Non-reactive	Reactive	Indeterminate	Unknown	Total
TB Diagnosis	50 (68.5%)	17 (23.3%)	6 (8.2%)	0 (0%)	73 (100%)
TB screening	50 (64.9%)	21 (27.3%)	6 (7.8%)	0 (0%)	77 (100%)
Pre-biologics	146 (93.6%)	8 (1.3%)	2 (5.1%)	0 (0%)	156 (100%)
Not specified	126 (68.1%)	29 (15.7%)	13 (7%)	17 (9.2%)	185 (100%)

Conclusion: T spot TB provides a highly sensitive and specific way of detecting active or latent TB. Almost a third of the requests are to assess TB status prior to the introduction of biological agents. This will have a major impact on resources with the increased usage of biologics, something that needs to be borne in mind whilst developing this service.

P2661

Clinical utility of the interferon-gamma for the diagnosis of active pulmonary tuberculosis

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Introduction: A rapid diagnosis of TB is crucial not only for patients, but also for TB control in the community. Currently, T-cell based interferon gamma release assays (IGRA) are acknowledged as the best methods available for the screening of latent tuberculosis infection (LTBI) and also as aid for the diagnosis of active tuberculosis (TB). The performance of these diagnostic tests has not been evaluated in Serbia.
Aim: To compare the sensitivity of QuantiFERON-TB Gold In-tube test (QFT), tuberculin skin test (TST) and acid-fast staining of sputa in patients with culture confirmed active pulmonary TB (PTB).
Methods: The sensitivities were evaluated in 70 HIV negative patients with culture confirmed M. tuberculosis infection. The sputum culture result was used as a gold standard. TST results were analysed at 5, 10 and 15mm cut-offs. QFT test was interpreted following the manufacturer's criteria.
Results: Sensitivities of the TST using a 5, 10 and 15mm of cut-offs were low: 55,7%, 51,4%, 45,7% respectively. Sensitivity of QFT (68,6%) was higher than that for all the TST sensitivities. The overall agreement between TST (all cut-offs) and QFT was poor. Sensitivity of the acid-fast staining of sputa was 75,7%. Naither detected statistical significant difference between sensitivity of QFT and the acid-fast staining of sputa, nor between of QFT and TST≥5 and TST ≥10 mm, but detected between sensitivity of QFT and TST≥15 mm (p=0,01).
Conclusion: Although findings have revealed a generally low sensitivity of the QFT, our opinion is that QFT can be used as adjunct diagnostic technique for active TB disease.

P2662

Unfavorable factors associated with false negative results of interferon-gamma release assay for tuberculosis

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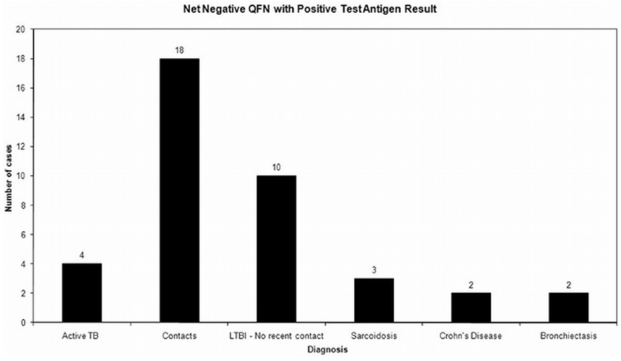
Imperfect sensitivity of interferon-gamma release assay (IGRA) is a potential problem to detect tuberculous infection. We comprehensively investigated factors that can lead to false negativity of IGRA, regarding active tuberculosis as a surrogate. In total, 543 patients with new smear-positive pulmonary tuberculosis were tested. At the time of diagnosis, peripheral blood was collected and IGRA (QuantiFERON-TB Gold In-Tube®, Cellestis, Victoria, Australia) was performed. Clinical and epidemiological information of the host and pathogen was collected together. Factors negatively influencing IGRA results were evaluated using logistic regression model.
Age ≥ 80 years old, body mass index <18.5 and the extensive infiltrates on chest X-ray showed significant associations with IGRA negativity (OR = 8.47 [95% CI, 1.53–46.84], 3.46 [95% CI, 1.14–10.49], and 3.07 [95% CI, 1.18–7.97], respectively), whereas HIV co-infection was rather associated with indeterminate results (OR = 23.30 [95% CI, 4.28–126.82]). Having HLA-DRB1*0701 allele was also associated with IGRA negativity; the number of the HLA allele that the patients possess correlated inversely with concentrations of interferon-gamma induced by *M. tuberculosis*-specific antigens (heterozygotes; 2.08 IU/ml [interquartile range, 0.79–4.64] and homozygotes; 0.15 IU/ml [0.06–0.26], respectively; $P = 0.0064$)
A variety of factors including an HLA allele affect false negativity of IGRA. Assessment of these factors in tested populations would contribute to a secure and reliable interpretation of IGRA results.

P2663

The influence of a high negative control on the interpretation of QuantiFERON Gold in Tube (QFN) results

William Ricketts, Graham Bothamley. *Respiratory Medicine, Homerton University Hospital, London, United Kingdom*

Introduction: The QFN result is a composite of a nil control, a mitogen positive control and a test sample (TB Ag) utilising ESAT-6, CFP-10 and TB 7.7 antigens. The result is defined as TB Ag minus nil control (positive >0.35 IU/ml). A strongly positive nil control (>8 IU/ml) or failure of the positive control are defined as indeterminate results. We were concerned that high values obtained after antigen stimulation might be significant.
Method: All patients tested by QFT-GIT between 31/3/10 and 28/1/11 (n=651) had the raw data from their test reviewed. Clinical diagnosis was examined in all those with a TB Ag result >0.35 IU/ml with a nil control high enough to bring the net result to <0.35 IU/ml or an indeterminate result.
Results: There were 39 (6%) instances of a net negative result despite a response to antigen, including four cases of active TB (4.7% of TB cases). Nil control results ranged from 0.1-3.07 IU/ml, with TB Ag levels up to 1.52 IU/ml.



Eight with a negative mitogen response included two cases of active TB, one of whom also had HIV. No cases were defined as indeterminate by virtue of a strongly positive negative control.
Conclusion: Reporting of a net result without a breakdown leaves clinicians vulnerable to missing cases of active TB. Sarcoidosis, Crohn's disease and bronchiectasis, all diseases associated with polyclonal B cell activation, also appear to show spontaneous interferon-gamma release.

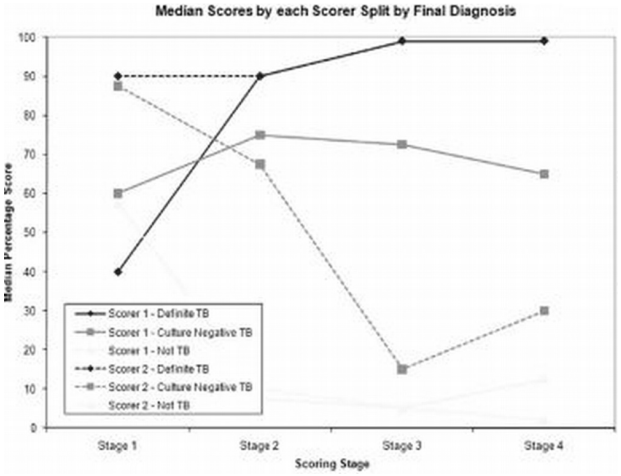
P2664

Clinical history and IGRA is unreliable in the diagnosis of tuberculosis

William Ricketts, Graham Bothamley. *Respiratory Medicine, Homerton University Hospital, London, United Kingdom*

Aim: To monitor the stages in the diagnosis of tuberculosis (TB) and assess whether early interferon-γ responses can be helpful.
Method: Site – a TB clinic in a high incidence (58 per 100,000) area of London. 25 patients with suspected TB were followed through their diagnostic process. Patients were scored using a modified UCSD Clinical Suspicion of TB tool by two clinicians blinded to the patient's identity and each other's scores. Estimated risk of TB was made at four stages in the diagnostic process: with clinical data, TST,

HIV and IGRA (QuantiFERON Gold in Tube) results; after radiology; sputum smear; and blood tests. A decision as to whether to offer treatment was made at stage two and at each stage thereafter.
Results: Clinical history with IGRA was associated with a wide variation in estimated probability of TB (IQR 15-85%). As the diagnostic process progressed, accuracy increased (see figure) and inter-scorer variation decreased. Diagnosis of culture-negative TB was especially problematic. Despite differences between the two scorers there was no difference in the number of patients treated correctly after radiology (69% vs 54% TB; 92% vs 92% not-TB), sputum smear (67% vs 44% TB; 83% vs 92% not-TB) and other blood tests (77% vs 46% TB; 92% vs 92%not-TB).



Conclusion: Clinical history even with IGRA results is unreliable in the diagnosis of TB. New tests for culture-negative TB would be valuable.

P2665
Specificity of IRGAs in Japan
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Objective: The specificity of IGRAs with the population being tested. The specificity of T-SPOT®.*TB* (T-SPOT) is generally reported to be lower than that of QuantiFERON®-TB Gold In-Tube (QFT-GIT). It is likely that studies carried out in developing countries include individuals with latent tuberculosis infection (LTBI). Therefore, we compared the specificities of T-SPOT and QFT-GIT in selected healthy subjects in Japan.
Subjects and methods: Blood samples were taken for both T-SPOT and QFT-GIT from university students who were selected according to stringent inclusion criteria and performed according to the manufacturer's instructions. Blood samples for T-SPOT were kept overnight at room temperature and treated the next day with T-Cell *Xtend* before PBMCs preparation. Blood samples for QFT-GIT were incubated within 16 hours after blood collection.
Results: University students with very low risk factors for TB infection were selected for the study. Of 111subgects, one was positive in both assays and 110 were negative by T-SPOT and 96 were negative by QFT-GIT. No results were borderline by T-SPOT (between 5 and 7 spots, USA criteria) and 14 were borderline by QFT-GIT (between 0.1 and 0.35 IU/ml, Japanese criteria). There was no indeterminate result.
Conclusion: Although several meta-analyses showed that QFT-GIT has the higher specificity compared to T-SPOT, our result demonstrate that the specificity of both assays is equally high. This implies that subjects for specificity studies should be strictly selected, especially as there is no gold standard for LTBI. If studies do not meet these criteria, they should not be included meta-analyses.

P2666
Do kinetics of interferon-gamma responses predict TB relapse?
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Evidence that quantitative T-cell interferon-gamma responses to *M. tuberculosis*-specific antigens may reflect mycobacterial burden has raised interest in these responses as biomarkers for treatment outcomes.
A cohort of 263 HIV-negative, culture-positive pulmonary TB patients enrolled between August 2005 and May 2007 who completed treatment and who were tested with QuantiFERON-Gold In-tube® (QFT-IT) and T-SPOT.*TB*® assays pre- and post-treatment were matched with the national TB registry for any re-notification as at December 31 2010.
Four patients relapsed with culture-positive, pansensitive pulmonary TB (relapse rate 1.5%). The time to relapse was 11, 17, 35 and 44 months. All were male. At the time of the first TB episode, their mean age was 45.4 years old, two were diabetic, two were cigarette smokers; all were baseline sputum AFB smear-positive; two had cavitary disease. All received directly-observed, 6-month chemotherapy with four first-line drugs in the intensive phase, followed by thrice weekly Rifampicin and Isoniazid in the continuation phase. All had one negative sputum culture (done once only) at two months of treatment.
All four patients were IGRA positive at baseline. The QFT-IT was negative at end of treatment and 6 months post treatment-completion in three and four patients respectively. All remained T-SPOT positive at treatment completion, while two reverted their T-SPOT results 6 months thereafter. Compared to the whole treatment cohort, a lower proportion of relapsed patients tested IGRA positive at 6 months post-treatment completion (0% vs 46.3% for QFT-IT; 50% vs 79.2% for T-SPOT). Our observations suggest that the kinetics of interferon-gamma responses are not useful for predicting disease relapse.

P2667
Necessity of re-centrifugation of QuantiFERON®-TB Gold In-Tube samples after plasma harvest
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Objective: In the course of research of QuantiFERON®-TB Gold In-Tube (QFT-GIT), we found that re-centrifugation of plasma samples from blood collection tubes decreased IFN-γ values. Thus, we examined the reasons for this phenomenon.
Subjects and methods: Blood samples were taken for QFT-GIT, incubated and centrifuged after incubation to separate plasma and blood cells. Plasma samples were harvested using pipettes and divided into two tubes. One tube was re-centrifuged and the other was not. After re-centrifugation, IFN-γ values of both samples were measured according to manufacturer's instructions.
Results: Most of re-centrifuged samples showed lower IFN-γ values compared with non re-centrifuged samples. We noticed that debris on the surface of separation gel was included in plasma samples after harvest, and when debris was added in ELISA plates, higher IFN-γ values were produced. Microscopic observations showed that debris included many live lymphocytes and these live lymphocytes adhered on plate wells even after intensive washing.
Conclusion: These observations suggested that live lymphocytes adhered on ELISA plate well may produce non-specific reaction, which result in higher IFN-γ values. Therefore, in order to obtain accurate results, QFT-GIT plasma samples after harvesting from QFT-GIT blood collection tubes should be re-centrifuged to spin down live lymphocytes.

P2668
Accuracy of IFN-γ and IP-10 detection for diagnosis of tuberculosis in children
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Objective: Evaluate IP-10 detection for latent tuberculosis infection (LTBI) and active tuberculosis (TB) in children, comparing the results with IFN-γ detection.
Material and methods: IFN-γ released was determined by Quantiferon-TB Gold In Tube (QFN). IP-10 was retrospectively detected in supernatants by an in-house ELISA and analyzed using preset cut offs for positive (6.4ng/ml) and indeterminate (3.5ng/ml) IP-10 test result (Ruhwald, Latorre in prep).
Results: 45 pediatric patients were classified in 3 groups. *Group 1:* 10 children diagnosed with active TB, with a *Mycobacterium tuberculosis* positive culture or active clinical TB. *Group 2:* 15 children enrolled during LTBI screening studies, with a positive QFN. *Group 3:* 20 healthy control children, with a negative QFN. Sensitivities of both IFN-γ and IP-10 assays were 50% (Group 1). Specificity of IP-10 detection was 100% (Group 3). Combining both cytokines the sensitivity improved to 60%, without a compromise of the specificity. Percentage of positive IP-10 responders among children from group 2 was 66.7%. Two children with negative IP-10 assays from this group had QFN positive borderline results. Global concordance between assays was 82% (κ=0.624). IP-10 released after specific antigens stimulation in active TB patients was significantly higher than in healthy controls (p=0.007), this was not seen for IFN-γ (p=0.199).
Conclusions: IFN-γ and IP-10 sensitivity is low, but combination of both cytokines increased sensitivity without a compromise of the specificity. Lowering IP-10 and IFN-γ cut offs could improve the sensitivity in children.
Concordance between both assays is good.
IP-10 could be an alternative marker for LTBI and active TB in children.