53. Tuberculin skin tests, interferon-gamma release assays and beyond

P294
The specificity of a new skin test – Diaskintest (recombinant protein CFP10-ESAT6) in patients with sarcoidosis and non-tuberculous pulmonary diseases
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Background: DIASKINTEST (DST) is a recombinant protein combination of CFP10-ESAT6 for intradermal injection and does not cause delayed-type hypersensitivity reactions in the BCG vaccinated. Objectives and methods: We have used DST intradermally in a dose of 0.2 mg in 0.1 ml in 179 patients aged 18-69 yrs.: 62 patients with morphologically identified sarcoidosis, 11 active tuberculosis (TB) patients and 106 patients with other nonspecific inflammatory lung diseases. In 72 hours we measured the diameter of induration and hyperemia in mm. Positive response is evaluated as any size induration, negative - no induration and hyperemia
Results: Among sarcoidosis patients we saw no reaction to DST - the negative results were in all 62 cases, and specificity of DST in this group was 100% [95%CI 99.9%-100%]. In 106 patients with nontuberculous inflammatory lung diseases we had 8 positive result (2 patients with asthma, 2 with nonspecific pneumonia, 2 with lung cancer, 2 with COPD), specificity of the method in this group was 92.5% [95%CI 87.4-97.5%]. 2 of 11 TB patients had negative response to DST (1 patient with TB and hepatitis C and 1 with TB and alcoholism), so specificity is 81.8% for TB cases [95%CI 58.9%-100%]. In our previous research we had similar results using IFN-g release assay (IGRA) in response of blood cells to ESAT-6 ex vivo, but in the clinical practice we see that the IGRA is more laborious method.
Conclusion: DST has high specificity in sarcoidosis patients and in patients with other nonspecific inflammatory lung diseases. DST may be used as an additional test for the differential diagnosis with TB in unclear cases.

P295
DIASKINTEST as a screening method at the mass child health examination for tuberculosis in Russia
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The Russian Federation has developed a new efficient method for the detection of initial tuberculosis presentation, DIASKINTEST, which is a combination of two antigens, present in the virulent strains of Koch’s bacilli and absent in the BCG vaccine strain and strains of other nonpathogenic mycobacteria. The purpose of research is the study of the efficient usage of current diagnostic program as an alternative to the open patch test with PPD-L (PPD test with 2 TE) in conditions of screening programs for tuberculosis. Children and teenagers, school and college students are divided into two groups by the method of total single-step survey. The first group comprises of children (aged 7-14) in the quantity of 816 individuals, the second group comprises of teenagers (15-17 years) in the quantity of 422 individuals. Over 44% of children and teenagers had a positive take to PPD-L. All children (in the quantity of 1238 individuals) were examined via DIASKINTEST: Only 4% of them revealed positive reactions to it. 7 patients showed the activity of local tuberculous changes during the X-ray examination. Thus, detection of local tuberculous at the examination of all schoolchildren and teenagers with the help of DIASKINTEST made 0.56%.
Conclusion: The implementation of a new method of tuberculosis diagnostics as a screening method of examination of children and teenagers in the medical practice, which possesses a high specificity owing to the use of secretory proteins ESAT-6 and CFP-10 and its easy test version will allow to enhance the efficiency of tuberculous infection diagnosis, reduce unnecessary expenses and improve the overall epidemic situation on tuberculosis.

P296
Differential diagnosis of outpatient pneumonia and infiltrative pulmonary tuberculosis by the Diaskintest²
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The purpose of this study was to investigate the possibility of differential diagnosis of infiltrative pulmonary tuberculosis and pneumonia with the use of traditional diagnostic set minimum and the Diaskintest in pulmonary center. The study involved 86 patients with suspected tuberculosis. Of these, 10 patients gave a positive result on the Diaskintest, and then activity of tuberculous process was confirmed in a tuberculous dispensary. The rest of the inclusion group and the of a comparison group with the classical outpatient pneumonia (52 patients) was negative result on the Diaskintest, antigenic and antibacterial test had a positive effect. It is concluded that the Diaskintest may be a marker to identify active tuberculosis in the differential diagnosis of outpatient pneumonia.

P297
The role of two stage tuberculin skin test in screening of household contacts
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Rationale: The role of two stage tuberculin skin test (TST) of persons who have had close contact with a case of smear-or culture positive pulmonary tuberculosis (TB) phenomenon may occur and therefore screening with TST in a two stage approach may not be ideal [1].
Methods: We conducted a prospective study including consecutive household contacts of patients with active PTB between Jan 2010 and Jan 2011. All contacts below the age of 35 years of age were interviewed and screened by TST. If the TST was below 6mm, contacts were asked to return for a second TST six weeks later. TST conversion was defined as a TST of greater than 6mm or a 5mm increase of the initial TST. Interferon Gamma Receptor Assay (IGRA), (T-spot test) was performed on those that had TST conversion to evaluate the booster phenomenon.
Results: Out of 589 adult and paediatric contacts screened who had an initial TST of less than 6mm, 189 contacts did not attend for a second TST. A total of 406 had TST performed. 344 contacts attended and did not show TST conversion and were therefore discharged. 68 contacts underwent TST conversion of whom 43 had an IGRA. Out of 43 contacts, 6 had a positive IGRA, 7 out of 43 were indeterminate and 30 were negative. Indeterminate and negative IGRA results were more common in children than in adults.
Conclusion: A two stage approach of TST may cause a booster effect and therefore should be confirmed by IGRA.
Reference:
P298

Variables responsible for tuberculosis positivity of household contacts of sputum positive TB cases

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In high TB endemic countries such as India, chemoprophylaxis is routinely offered only to contacts below 6 years of age. The possibility however remains that the index case may have transmitted the infection to other family members before the diagnosis of the disease. We evaluated this in the context of Indian patients. We did a cross sectional study of household contacts of the sputum positive TB patients diagnosed from our center that had no other index case at home present in or out of the past. The contacts were tested for tuberculosis infection using 5 T-SPOT PPID (Purified Protein Derivative) immediately after the diagnosis of the index case. An induration of more than 10 mm was taken as a positive result. A total of 179 household contacts of 50 index cases were evaluated. 85/59 (47.48%) were tuberculosis positive. 6/23 (26.09%) contacts below 6 years of age were infected. 31/44 spouses (70.45%) were tuberculin positive as against 39/87 (44.82%) first degree relatives and 15/48 (31.25%) second degree relatives. 61/112 (54.46%) contacts sharing same bedroom were infected as against 13/57 (23.28%) not sharing the bedroom. The prevalence of infection of contacts correlated with the grade of sputum positivity of the index case. The variables that did not show any statistically significant correlation were presence of lung cavity, hemoptysis, literacy level and gender. This study suggests the need for offering chemoprophylaxis to the contacts sharing bedroom including spouses in addition to contact children below 6 years of age.

P299

Tuberculin skin test size and risk of tuberculosis: A 12-year follow-up of contacts of TB cases

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Background: There are no long-term cohort studies assessing the risk of developing active TB according to the tuberculin skin test size in contacts of TB cases who did not receive treatment for latent TB infection (LTBI). The objective of this study was to determine if TST size at baseline predicts risk of TB disease.

Objective: To assess the risk of TB – according to the tuberculin skin test (TST) size in contacts of active TB cases who did not receive LTBI treatment.

Methods: A retrospective cohort study of contacts of active TB cases recorded in British Columbia, Canada. We estimated the risk at 12-year rate of developing TB for infected and uninfected contacts – according to tuberculin skin test size - using incidence rates. Contacts with HIV infection or with an indeterminate TST were excluded.

Results: Among 26,542 contacts, 180 individuals developed TB (tuberculosis rate 678/100,000). Non-infected Household contacts (tuberculin skin test size 0-4 mm) had a TB rate of 1,014/100,000 whereas those with a TST of 5-9 mm a TB rate of 2,162/100,000; and those with 10-14 mm a rate of 4,478/100,000. Non-infected Close non-household contacts had a TB rate of 222/100,000; those with a TST of 5-9 mm a TB rate of 296/100,000 and those with 10-14 mm a rate of 1,821/100,000.

Non-infected Casual contacts had a TB rate of 83/100,000; those with a TST of 5-9 mm a TB rate of 204/100,000 and those with 10-14 mm a rate of 860/100,000.

Conclusions: The risk of TB increases with TST size in all contacts; especially with close contacts. TB risk is high for all household contacts, including those considered non-infected. The risk of TB in non-household contacts is significant only when the TST is >10 mm.

P300

Comparative study of the usefulness of TST and three interferon-gamma release assays (IGRAs) for the differential diagnosis of pulmonary tuberculosis

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Objective: We compared the usefulness of tuberculin skin test (TST) and three interferon-gamma release assays (IGRAs) for the differential diagnosis of pulmonary tuberculosis (TB).

Methods: The subjects were 70 patients who required the differentiation of pulmonary TB clinically. The final clinical diagnosis of pulmonary TB in 22 patients and non-pulmonary TB in 48 patients was established by clinical specimens and according to the revaluation.

Results: In 22 patients with pulmonary TB, the positive response rate was 60% on TST, 80% on QFT-2G, 85% on QFT-3G and 95% on T-SPOT.TB. In 48 patients with non-pulmonary TB disease, the positive response rate was 47% on TST, 9% on QFT-2G, 9% on QFT-3G, 13% on T-SPOT.TB. Indeterminate results on three IGRAs were recognized in one patient each on QFT-2G and QFT-3G among patients with pulmonary TB in three patients on QFT-2G and two patients on QFT-3G among patients with non-pulmonary TB disease. However, there were no indeterminate results on T-SPOT.TB in either patient group. Patients with false-negative or indeterminate results on IGRAs had severe underlying diseases or were receiving immunosuppressive treatments.

Conclusions: T-SPOT.TB provided the best positive response rate for patients with pulmonary TB among three IGRAS, although T-SPOT.TB may have problems in the specificity of diagnosing TB disease. Therefore, we think it is important to perform T-SPOT.TB in combination with QFT to elevate the sensitivity of the diagnosis of TB disease based on the findings in this study.

P301

Comparison between tuberculin skin testing and QuantiFERON Gold-InTube for detection of latent tuberculosis in rheumatoid arthritis patients in a TB endemic population

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Introduction: The purified protein derivative (PPD) skin test is the only widely used method which detects latent tuberculosis infection (LTBI) and is dependent on a normal T cell function. In rheumatoid arthritis (RA) the T cell function is altered, which may result in an inability to develop an adequate PPD reaction. T-SPOT.TB (QuantiFERON Gold-InTube) are now available alternatives to tuberculin skin testing (TST) to detect LTBI.

Aims: This study compared QuantiFERON-TB Gold In-Tube (QFT-IT) with the TST for the detection of latent tuberculosis infection among patients with RA in an area with a high prevalence of tuberculosis (138/1000), where BCG vaccination is mandatory.

Methods: A prospective study of patients with received the TST and an IGRA, the QFT-IT, at Pneumonology Department, Clinical Hospital "Dr.V Babes", Timisoara, from July 2009-December 2010.

Results: Of 194 patients who underwent TST and QFT-IT testing, 101 (52%) had RA. Significantly fewer controls than RA patients had positive TST results (26.7% vs. 65.6%, p < 0.001), whereas the percentage of positive QFT-IT results was comparable for both groups (44.5% vs. 59.1%, p = 0.054). Indeterminate QFT-IT results were found in two (1.9%) RA patients. QFT-IT has 2-times greater sensitivity than TST, but positive QFT results in older RA patients compared with controls was reduced (40% vs 71%).

Conclusion: In a TB endemic population, the TST-IT assay seemed to be a more accurate test for detection of LTBI in RA patients compared with the TST, and may potentially improve the targeting of prophylactic therapy before treatment with anti-TNF agents.

P302

IGRA and TST predictive value in immunocompetent close contacts

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Background: Limited data is available on the predictive values of interferon-g release assays (IGRAs) and tuberculin skin test (TST) for progression to active tuberculosis (TB).

Objective: To access the positive and negative predictive values (PPV and NPV) of IGRA and TST for progression to TB disease, in recently exposed immunocompetents of positive smear sputum TB patients who did not do preventive treatment.

Methodology: Retrospective cohort study of close contacts of smear-positive TB cases screened for TB from January 2007 to December 2009 – follow up until February 2011. Screening included: symptom inquiry, TST and IGRA (to confirm a positive TST >10 mm), chest radiography – negative results were re-evaluated 8-12 weeks later.

Results: 106 contacts were studied. Considering only those who did not do preventive therapy (79%) both tested positive for TST and 19 tested positive for IGRA (54 were already positive for TST and 16 were positive for IGRA in the first evaluation). Three contacts developed active TB: one positive for TST and IGRA at first evaluation (refused preventive treatment) and two (TST+IGRA-) missed revaluation. In the first evaluation, TST presented a PPV of 5.7% and a NPV of 100%; IGRA presented a PPV of 5.3% and a NPV of 96.1%. In the second evaluation, both TST and IGRA had a PPV of 0% and a NPV of 100%.

Conclusions: TST and IGRA had a better PPV for progression to TB in the initial evaluation (after index case diagnosis). A negative IGRA in the first evaluation could not exclude progression to TB.

P303

QFT-GIT vs TST in diagnosis of latent tuberculosis infection (LTBI) in Poland

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Introduction: Poland is characterized by medium prevalence of tuberculosis (61.0/100 000) in 2009. The diagnosis of LTBI is difficult due to obligatory multiple reevaluation in the past as well as low specificity of tuberculin skin test. So far IGRA tests were rarely done in Poland.

Abstract printing supported by Chiesi Visit Chiesi at Stand D.30
Aim: To assess the prevalence of LTBI in the risk groups: 1. homeless, 2. close contacts, 3. casual contacts, 4. nursing home pensioners, 5. random population subjects from Krakow (controls) by QFT-GIT and TST, to compare the agreement and kappa of these tests at 5, 10, 15 mm of TST cut-off and to establish the best TST cut-off for the diagnosis of LTBI.

Material and methods: From July 2007 to October 2009 QFT-GIT test was performed in 785 subjects: in group 1 (n = 150); 2 (n = 171); 3 (n = 163); 4 (n = 152); 5 (n = 149) and TST was carried out in: 129, 156, 147 and 121 subjects respectively. In each group the agreement and kappa coefficient between QFT-GIT and TST at 5, 10, 15 mm of TST diameter was analyzed.

Results: We observed high prevalence of LTBI in relevant studied groups: 37% 27%, 25%, 21% and 23% by QFT-GIT and by TST 56%, 47%, 48%, 43%, 48% respectively. Agreement and kappa at 5, 10, 15 mm of TST cut-off in each group were: in group 1: 58.7% (0.26); 60.7% (0.34); 75.4% (0.65); 2: 55.8% (0.20); 73.0% (0.29); 73.4% (0.32); 3: 53.7% (0.19); 75.0% (0.30); 76.1% (0.40); 4: 64.0% (0.29); 78.8% (0.36); 79.0% (0.45); and in group 5: 55.8% (0.20); 60.9% (0.24); 70.0% (0.23).

Conclusions: TST was characterized by lower diagnostic value in Polish population vaccinated and revaccinated with BCG but TST cut-off 15 mm and more should be considered for diagnosis of LTBI because of highest agreement with QFT-GIT.
T-TSPOT.TB assay were done. Diagnosis of latent TB infection was based on the T-TSPOT.TB assay, after excluding active disease. Window prophylaxis was offered if the initial test was negative and stopped if the repeat T-SOT.TB assay done after 8–10 weeks of last exposure was negative. 36 (76.6%) the contacts were tested negative by the T-SOT.TB assay. Their median CD4 count was 343 cells/μl and 44.4% of them were on anti-retroviral therapy. 8 contacts had positive T-SOT.TB assay result, 2 of whom had active TB (yield of 4.3%). The indeterminate rate was 2.1%. 3 contacts completed preventive treatment while 14 contacts had window prophylaxis. During the mean follow up of 19 months (median of 21.5 months, range of 10 to 25 months), one of the T-SOT.TB assay negative contacts, whose CD4 count was 4 cells/μl, developed active TB 11 months after completing screening.

Except in cases with extremely low CD4 count, the T-SOT.TB assay appears to have a good negative predictive value for progression to active disease in HIV positive TB contacts in Singapore.

### P307

**Quantiferon and Mantoux results by Nationality**

<table>
<thead>
<tr>
<th>Nationality</th>
<th>Number</th>
<th>Positive (%)</th>
<th>Negative (%)</th>
<th>Mantoux reaction mm (range)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nepal</td>
<td>44</td>
<td>32 (73)</td>
<td>12 (27)</td>
<td>11 (6–21)</td>
</tr>
<tr>
<td>African</td>
<td>65</td>
<td>47 (72)</td>
<td>18 (28)</td>
<td>12 (6–22)</td>
</tr>
<tr>
<td>British</td>
<td>10</td>
<td>6 (60)</td>
<td>4 (40)</td>
<td>10.5 (6–15)</td>
</tr>
<tr>
<td>Other</td>
<td>6</td>
<td>4 (66)</td>
<td>2 (33)</td>
<td>10.5 (6–15)</td>
</tr>
<tr>
<td>Total</td>
<td>125</td>
<td>89 (71)</td>
<td>36 (29)</td>
<td></td>
</tr>
</tbody>
</table>

A positive QuantiFERON test and a Mantoux reaction of ≥15mm was assessed using a Chi-squared test. 153 cases were identified. 28 were excluded due to insufficient data, leaving a sample of 125. All cases were male. Median age was 21.5 years (range 17–53). For analysis recruits were divided into 4 areas (see table 1).

<table>
<thead>
<tr>
<th>Area</th>
<th>QFT+</th>
<th>QFT–</th>
<th>QFT+</th>
<th>QFT–</th>
</tr>
</thead>
<tbody>
<tr>
<td>East Europe</td>
<td>20</td>
<td>30</td>
<td>20</td>
<td>30</td>
</tr>
<tr>
<td>Romania</td>
<td>15</td>
<td>8</td>
<td>15</td>
<td>8</td>
</tr>
<tr>
<td>Kosovo</td>
<td>1</td>
<td>7</td>
<td>1</td>
<td>7</td>
</tr>
<tr>
<td>Bulgaria</td>
<td>1</td>
<td>5</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>Latin America</td>
<td>0</td>
<td>4</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>Asia</td>
<td>22</td>
<td>30</td>
<td>20</td>
<td>30</td>
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<tr>
<td>Afghanistan</td>
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<td>15</td>
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<tr>
<td>Iran</td>
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<td>0</td>
<td>5</td>
</tr>
<tr>
<td>Pakistan</td>
<td>0</td>
<td>4</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>Others</td>
<td>2</td>
<td>6</td>
<td>2</td>
<td>6</td>
</tr>
</tbody>
</table>

### P309

**Evaluating the use of the interferon-γ response to mycobacterium tuberculosis (MTB) specific antigens to diagnose latent tuberculosis infection in patients with chronic inflammatory joint and skin diseases**

**Abstract**

**Objective:** The treatment of chronic inflammatory diseases has been transformed with targeted biologic therapies. Patients receiving this treatment are at increased risk of reactivating latent tuberculosis infection (LTBI). Tuberculin skin test (TST) has been the gold standard for detecting latent LTBI, but may be difficult to interpret in patients receiving immunosuppressive therapies. In vitro interferon-gamma-release assays (IGRA) are an alternative.

**Aims:** Evaluate and compare the effectiveness of IGRA (QFR-TB Gold In Tube assay [QFR] and T-SOT.TB assay [TSTB]) against TST in a cohort of patients due to commence biological therapies.

**Methods:** A prospective cross sectional study was conducted at an inner London tertiary referral centre. Patients were screened for LTBI and a TST was performed. Venous blood samples were obtained for QFR and TSTB.

**Results:** 102 patients were included, aged between 18 and 83 years. A total of 42 patients were receiving at least one immunosuppressive therapy. A TST result was available in 84 patients. The overall agreement between the QFR vs TSTB excluding the indeterminate results was 69 out 78 (k=0.572), between QFR vs TST was 62 out of 79 (k=0.241) and between TSTB vs TST was 58 out of 72 (k=0.304). 13 patients received chemoprophylaxis for presumed LTBI.

**Conclusion:** Our study showed variation in results obtained from TST and IGRA's when used in our population, some of whom where immune compromised. For patients where interpretation of the results may prove challenging, LTBI screening tests may be best used in combination.
some nationalities and in Pts with TST>15 mm, partly decreasing the economic advantage of its use to reduce PTs. Better information on QFT role could be obtained extending this program to HS with TST ≤9 mm or age≥36 yrs but QFT technicalities and health-care budget cuts do not allow this wider on-field application.

P311
Practical use of quantiferon test in Norway’s largest TB-clinic

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Introduction: Since November 2008, Norway’s largest TB-clinic (Diagnosestasjonen, Oslo University Hospital, Ullevål) has implemented Quantiferon-TB GOLD In-Tube test (QFT) as a supplement to tuberculin skin test (TST).

Aim & objectives: The aim of our study was to evaluate clinical use and consequences of QFT results for the patient.

Methods & design: Patients with conclusive QFT result between November 2008 and December 2009, were included in this retrospective cross-sectional study.

Results: 415 patients were included and categorized by reason for referral: Immigrants (31.1%), School children (49.6%), Immunomodulation (7.5%), TB-Contacts (10.6%), Other (1.2%). Only 14 (3.4%) patients had negative TST (Mantoux<6mm). 124 (29.9%) patients had positive Quantiferon test. While 70 (56.9%) Immigrants had positive QFT; 33 (16%) of the School Children had positive QFT result. A multivariate analysis showed that the independent risk factors of a positive QFT test were higher age, reported TB-exposure, high-incidence TB region of birth and immigrants. QFT positivity was associated with age group >35 (OR 3.5) compared to age group<14, reported TB-exposure (OR 4.5), and high-incidence TB region of birth (OR 3.84). QFT result was the single most important factor for determining clinical outcome: 116 patients (94.3%) of patients with positive QFT tests were selected for either preventive therapy or follow-up, versus 11 (3.8%) of patients with negative tests.

Conclusions: QFT result was the single most influencing factor on the clinicians decision making. Though QFT has reduced numbers of patients given treatment and follow-up, it is important to keep in mind the limited sensitivity of QFT in certain patient groups when interpreting the QFT results.