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Interferon-gamma release assay versus tuberculin test in diagnosis of latent tuberculosis infection among healthcare workers, Sana'a city Yemen

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ABSTRACT
Background: Latent tuberculosis infection (LTBI) is the presence of Mycobacterium tuberculosis in the body without evidence of tuberculosis (TB) disease. Healthcare workers (HCWs) are at occupational risk of contracting & transmitting TB. Therefore, identifying of persons with LTBI is an essential TB prevention & control strategy. The aim of this study was to evaluate the performance of interferon-gamma release assay (IGRA) in diagnosis of LTBI among HCWs, in comparison to tuberculin skin test (TST); as well as identifying risk factors fortest positivity.
Methods: A cross-sectional study was conducted in 2015 on 90 HCWs tested with TST and IGRA. All Participants were subjected to a questionnaire to assess their risk factors. Differences between categorical variables were analyzed by X2. Kappa coefficient used to detect the agreement between TST and IGRA. Also, accuracy of both tests was calculated. Results: Of the 90 HCWs; 54.4% were females and 45.6% were males with an age ranged between 20-60 years. The participants were classified as 38.9% nurses, 34.4% physicians, 16.7% technicians and 10% were administrators. The highest prevalence of LTBI was found in female 14.3% & 28.6% by TST and IGRA, respectively. In general, the positivity by both tests increased with increasing age. 27.8% of HCWs were Bacille Calmette-Guérin (BCG) vaccinated and most of the positivity occurs among vaccinated HCWs. The crude prevalence for LTBI was 12.2% by TST and 20% by IGRA; overall agreement between TST and IGRA results was poor (k= 0.39).
Conclusion: Most HCWs with LTBI were females, nurses, older age, BCG vaccinated and in a group of profession of close contact with TB patients'. The higher positivity was found by IGRA than by TST with poor agreement between them.
Superior specificity was observed with IGRA; whereas TST had a higher sensitivity.

CONTENTS

Introduction:

Tuberculosis (TB) is a serious public health problem that is responsible for about 1.7 million deaths each year. The current trends suggest that TB will still be among the 10 leading causes of global disease burden in the year 2020; with an estimation that one-third of the world's population latently infected with TB 1, and therefore potentially at future risk of developing active disease. Latent tuberculosis infection (LTBI) is considered to be given when the immunological tests are positive and active TB is ruled out by way of X-ray bronchoalveolar lavage or sputum-smear microcopy2.

Working in healthcare is a well-known risk factor for TB. Even in countries with low TB incidence, the risk of TB in healthcare workers (HCWs) is elevated for a wide range of tasks in healthcare, and the prevention of nosocomial infection of HCW remains as a challenge 3. Improvement of diagnostic methods for LTBI is an important step towards the goal of tuberculosis elimination, as laid out by the WHO Stop TB strategy 4. In Yemen, the identification of LTBI relies on the tuberculin skin test (TST) which is based on a type IV delayed hyper sensitivity reaction that occurs when those infected with Mycobacteria tuberculosis (M.tuberculosis) are exposed intradermally to standardized mycobacterial antigenic components. The immune response To M.tuberculosisis highly dependent up interferon-gamma (IFN-γ) production by macrophage and antigen-specific T cells 2, 5. However, TST has many limitations in terms of its potential and the cross-reactivity with the other strains of mycobacteria and antigens present in Bacille Calmette-Guérin (BCG) vaccination 6. Recently, interferon gamma release assays (IGRAs) have provided an additional tool to identify suspected cases of TB. As with TST, IGRAs measure the cell-mediated immune response to antigens specific for M. tuberculosis, therefore, both TST and IGRAs do not measure the presence of viable M. tuberculosis but merely the immunological footprints left behind by recent infection 7.

Aimes of the study

The primary objective of this study was to assess the performance of IGRA in diagnosis of LTBI in comparison to TST among HCWs in Sana'a, Yemen. Secondly, to identify the risk of infection associated with TST and IGRA positivity among these HCWs.

Subjects and methods

A cross-sectional study of 90 HCWs was carried out in the Typical Police Hospital, Al-Thawra Hospital and the National TB Center in Sana'a city from January to March 2015. Variables were collected at the individual levels included sex, age, occupation. Each participant in this study was examined for the presence of BCG scar. All individuals were informed of the nature of the study and agreed to participate.

About 5 ml of blood were collected from each participant and serum were separated and stored at -20 C0 for later analysis by DRG IFN-γ ELISA kit; which is a solid phase enzyme amplified sensitivity immunoassay performed on micro titer plate. The TST was performed by injecting of 0.1 ml of tuberculin into the skin in the lower part of the arm of each participiant. The test was read within 48-72 hours, according to manufacture information. TST induration ≥ 10 mm was usedas a cutoff point 8.

SPSS software for windows,version 20 (SPSS Inc, Chicago, IL, USA) was used for statistical analysis. The chi square (X2) test was used to compare the frequencies of positive test results among different groups of study recruits. Results were reported as odd ratios (OR) with confidence intervals (CI). Statistical significance was set to p-value <0.05. Test concordance was calculated using chance corrected proportional agreement rate; kappa (κ) coefficients, with κ>0.75 defined as excellent agreement, κ<0.4 as poor agreement, and κ between 0.4 and 0.75 as fair to good agreement 9. Additionally, sensitivity, specificity, false positive and negative, positive predictive values (PPV) and negative predictive values (NPV) were calculated

Results

This study included 90 HCWs; 54.4% were females and

45.6% were males. The participants' age ranged between 20-60 years. Most of the selected individuals were in age group 30-39 years (31.1%), followed by age group 20-29 years (28.9%) while age group 50-60 years were only 13.3%.

The distribution of HCWs according to profession in our study; 38.9% was nurses, followed by physicians (34.4%), technician (16.7%) and administrators (10%). Of the 90 HCWs only 25 (27.8 %) have been vaccinated against TB (Table 1).

Table 1: Description of the study population

Variables	No.
	41(45.6%) 49 (54.4%)
Age group (years) 20-29 30-39 40-49 50-60	 26 (28.9%) 28 (31.1%) 24 (26.7%) 12 (13.3%)
Profession Administrator Technician Physician Nurse	 09 (10%) 15 (16.7%) 31 (34.4%) 35 (38.9%)
BCG vaccination NO Yes	 65 (72.2%) 25 (27.8%)
Total	90 (100%)

The higher positive rate was found in female 14.3% and 28.6% by TST & IGRA, respectively. The age specific rates were highly varied for different age groups (ranged from 7.7-16.7% by TST and from 3.8-50% by IGRA).

Generally, both tests tended to have higher positive results with increasing age.

As regard profession, the highest rate of TST positive occurred among nurses (14.3%) followed by physicians (12.9%) however, the highest rate by IGRA occurred among technicians and administrators (33.3%). Among the vaccinated HCWs; the positivity of IGRA (56%) was higher than those with TST (36%). Similarly, the positive results (6.2%) by IGRA were higher than that by TST (3.1%) among unvaccinated HCWs. Moreover, most of the positivity results 81.8% by TST and 77.8% by IGRA were found among vaccinated HCWs (Table 2).

Table 2: Prevalence of LTBI among health care workers using IGRA and TST

Variables	TST Positivity No.	IGRA Positivity No.
Sex (n) Male (41) Female (49)	 4 (9.8%) 7(14.3%)	 4 (9.8%) 14(28.6%)
Age group (years) (n) 20-29 (26) 30-39 (28) 40-49 (24) 50-60 (12)	 2 (7.7%) 3 (10.7%) 4 (16.7%) 2 (16.7%)	 1(3.8%) 5(17.8%) 6(25%) 6(50%)
Profession (n) Administrator (9) Technician (15) Physician (31) Nurse (35)	 1 (11.1%) 1 (6.7%) 4 (12.9%) 5 (14.3%)	 3(33.3%) 5(33.3%) 4(12.9%) 6(17.1%)
BCG vaccination (n) NO (65) Yes (25)	 2 (3.1%) 9 (36%)	 4 (6.2%) 14 (56%)
Total	11 (12.2%)	18 (20%)

The association of TST and IGRA with risk factors summarized in Table 3. When sex was considered, there was association of positivity with female in which associated OR is equal 1.54 & 3.7 times by TST and IGRA, respectively; being significant (p=0.02) only in IGRA. When the positive results of both tests were analyzed using age group, the significant association was obtained only for a positive IGRA with the oldest group (p=0.005). As an indicator of experience, the highest positivity by TST and IGRA found in the older age group; 16.7% & 33.3% (OR=1.96 & 4.0), respectively; being significant (p=0.011) only in IGRA. As regard BCG vaccination, the positivity of both tests among vaccinated HCW was higher than among unvaccinated and the difference was statistically significant (p= 0.000) by both tests.

Table 3: Risk factors of latent tuberculosis infection among health care workers

Character	TST				IGRA			
	OR	CI	χ^2	*P*	OR	CI	χ^2	*P*
Sex (n)								
Male (41)	Ref				Ref			
Female (49)	1.54	0.42-5.69	0.43	0.37	3.7	1.1-12.33	4.94	0.023
Age group (years)								
(N)	Ref				Ref			
20-29 (26)	0.81	0.15-3.79	0.09	0.76	0.82	0.22-2.9	0.12	0.73
30-39(28)	1.7	0.4-7.4	0.25	0.61	1.55	1.3-2.4	0.51	0.005
40-49(24)	1.5	0.2-9.5			5.5		7.8	
50-60(12)								
Profession (n)								
Administrator(9)	Ref							
Technician(15)	0.46	0.02-4.1	0.52	0.47	2.4	0.6-9.5	2.09	0.15
Physician(31)		0.24-4.7			0.48	0.12-1.8	1.5	0.22
Nurse (35)	1.11	0.3-5.65	0.02	0.88		0.22-2.5	0.29	0.58
	1.41		0.23	0.63	0.74			
BCG vaccination (n)								
Negative(65)	Ref				Ref			
Positive (25)	0.56	.011-.287	18.2	0.00	.052	0.014.186	28.0	0.000

OR: odds ratio ≥ 1 at risk; CI: Confidence intervals; X2: Chi-square ≥ 3.9 (significant);P: Probability value ≤ 0.05 (significant)

Total concordant results of the two tests were 83.3%,predominantly in those with negative results 90.7%, however, only 9.3% were positive by both tests. The total discordant results between the two tests occurred in 16.7% of the HCWs, most of them as a combination of TSTnegative/IGRA positive 73.3%; while the combination of TSTpositive/IGRAnegative was only 26.7%. Overall agreement between the assays was poor (k= 0.39, Table 4).

Table 4: Overall agreement between TST & IGRA

κ:

Concordant results (75, 83.3%)	Positive results (7, 9.3%) Negative results (68, 90.7 %)
Discordant results (15, 16.7%)	TST neg./IGRA pos. (11,73.3%) *VS* TST pos./IGRA neg. (4, 26.7%)
k	0.39

kappa coefficients; κ: κ>0.75 defined as excellent agreement, κ<0.4 as poor agreement, and κ between 0.4 and 0.75 as

fair to good agreement

Estimation of the accuracy of the tests for diagnosis of LTBI are shown in Table 5. As regard TST test, the sensitivity, specificity, were 63.6%, 86%, while for IGRA
38.8%, 94.4%, respectively.

Table 5: The accuracy of TST and IGRA

Test	Sensitivity	Specificity	False pos.	False neg.	PPV	NPV
TST	63.6%	86%	13.9%	36.4%	38.9%	94.4%
IGRA	38.9%	94.4%	5.5%	61%	63.6%	86.1%

PPV: positive predictive values, NPV: negative predictive values.

Discussion

This study investigated the performance of IGRA for a diagnosis of LTBI in comparison to TST (≥10mm) among HCWs. We found that the crude prevalence of LTBI in HCWs by TST was 12.2% while by IGRA was 20%. Our results were comparable to previous studies 10, 11 in Europe and Saudi Arabia that reported the prevalence of LTBI among HCWs by TST to be 12.8% and 11%, respectively and among French HCWs by IGRA being 22.2% 12. In addition, the prevalence of LTBI among HCWs in our study is higher than that of general population in Yemen (2-4%) 13. Our result was confirmed by many studies 14-16 which suggest that nosocomial transmission of TB is an important occupational problem among HCWs. On the other hand, lower rate of IGRA (7.2%) was reported by a study in a hospital for pulmonary diseases in Germany 17. Females had a higher rate of TST and IGRA positive results (14.3% & 28.6%, respectively) but the association was only significant with IGRA (p=0.023, Table 2). Our result is similar to previous studies in which females showed a higher prevalence of LTBI than males 18-20. While other studies showed the prevalence of LTBI not to be associated with sex 11, 16, 21. As regard the age, our results showed an association between age and the positive results of both tests, however the association was only significant by IGRA (p=0.005).

This was in agreement with a study reporting higher positive results with increasing age 16. Moreover, German and Japanese studies showed the association between age and LTBI with IGRA but not with TST 17, 22, 23. The immunologic interpretation of this observation is not clear; however, it may be due difficulties to apply the tuberculin correctly into the aging skin or by decreasing mobility of T lymphocytes to the forearm 22. In accordance with our results, a German study using Quanti-FERON-TB Gold In tube assay reported that the lowest prevalence in HCWs occurred among younger age group 17.

Surprisingly, in our study profession was not associated with both assays. Nurses had the higher positivity with TST (14.3 %), while technicians and administrators had the higher positivity (33.3 %) with IGRA without any significant. In agreement with our result, Torres

et al. 21 did not find any association of profession with positivity of TST nor IGRA. However, other studies reported that the nurses are at high risk of developing TB than other HCWs due to possibility of nosocomial transmission 3, 15, 24-26.

Regarding BCG vaccination, most of positivity of both assay; 23 (92%) of participants were BCG vaccinated. The significant association of positive LTBI with the previous BCG vaccination (p=0.000) in our results are consistent with many works that reported people with BCG vaccination to have a higher prevalence of LTBI 6, 21, 23, 25,27-29. A study reported that 20-25% of those receiving school-age BCG boosters would have a persistently positive TST at least 10 years later 30, and the repeated BCG vaccination increased the probability of TST results higher than 10 mm 21. Importantly, the declining of TST over years after vaccination cannot be measured accurately as a result of different types of BCG strains that administered and genetic variability of the individual 31. On comparing both TST & IGRA, our results found the higher positivity of LTBI among vaccinated HCWs were

56% by IGRA as compared to 36% by TST. Recently, a study reported that a recent BCG vaccination or a repeated BCG vaccination in general decreased the probability of IGRAs positive and they suggest that this may be due to a protective effect of BCG vaccination 21. However, in our study we did not have data about the number of doses nor time of BCG vaccination. Overall agreement between TST and IGRA was poor (k=0.39) which is in agreement with several studies 32, 14, 23, 33. In contrast, other studies showed good to excellent agreement between the two assays 23, 34-35. An earlier study by Pttumartly et al. using quantification assay showed a correlation between quantitative IGRA and the size of skin test indurations 36. Another study suggests that a positive QFT-GIT result may be a better indicator of LTBI than a positive TST result 37. According to this, quantifying the IFN-γ response may predict the future risk of developing TB. Concerning discordant results the proportion of TSTnegative/IGRA positive is more than TSTpositive/ IGRAnegative (11 vs. 4) of all HCWs tested. Our findings are consistent with a number of studies 12, 21 which reported that the overall positivity of LTBI by IGRAs is higher than that by TST. Nienhause et al. suggested that these combinations partly explained by decreasing sensitivity of the TST with age and the BCG vaccination 23. Although our study and other studies found that the prevalence of LTBI by both methods was statistically significance with BCG vaccination, however, there was no clear trend that TSTpositive/IGRAsnegative discordant result, increased with an increasing proportion of BCGvaccination 38. Interestingly, one study conducted in a city with a high TB incidence, high LTBI prevalence and where BCG is routinely administered, reported that the discordance between the two assay was high (24%), and BCG did not account for this discordance. Another study suggested that TSTpositive/IGRA negative results might be explained by resolved or old TB infection 39. However, other previous studies observed that only the combination of TSTpositive/IGRAnegative HCW develop active TB 27, 40. In addition, discrepancies encountered in our study might be due to the natural host variability and the different assay conditions.

In this study, we also assess the accuracy of the positivity of both tests in terms of sensitivity, specificity, NPV and PPV. Importantly, for any new test to replace the TST should show a higher specificity, especially in subjects with confounding factors, such as BCG vaccination, and at least a similar sensitivity. Our result showed that the specificity of IGRA (94.4%) was higher than that of TST (86%); however, its sensitivity (38.8%) was lower than that of TST (63.6%). This finding is consistent with many studies that compared TST and IGRA and reported the specificity of IGRA to be higher than that of TST 41-43. As we know the immune response to TB is complex and the decreased specificity is due to the non-specific immune response to a heterogeneity mixture of antigens

that are present in the mycobacterial cell wall 44, as well as due to BCG vaccination based on purified protein derivative 45. False positive of TST was 13.9% and 5.5% for IGRA while false negative 36.4%, 61%, respectively. False positive reactivity may be explained due to tuberculin strains such as BCG, inter-observer variability in reading 33, while the under laying immunosupression may lead to false negative results. In our study, NPV of TST (94.4%) was higher than that of IGRA (86.1%) while the PPV of IGRA (63.3%) is higher than that of TST (38.9%). Unfortunately, the lack of gold standard diagnosis method for LTBI makes the determination of accuracy for IGRA and TST difficult. However, an alternative way to evaluate the sensitivity in this context is through comparison of the risk of developing active tuberculosis in subjects with an increased risk of progression from LTBI to active TB 46.

Conclusion

A High positivity rate of LTBI was found by IGRA than by TST among HCW in Sana'a city, Yemen. IGRA results are more associated with risk factors than TST. TST and IGRA were significantly influenced by previous BCG vaccination, with poor overall agreement between the two assays. IGRA had asuperior specificity than TST; whereas TST had a higher sensitivity.

Recommendations

Additional studies are needed with quantities estimation of IGRA in line with study ofprogression disease.

References

[1] Ahmed N, Hasnain S. "Molecular epidemiology of tuberculosis in India: Moving forward with a systems biology approach".Tuberculosis 2011; 91 (5):407-3.

[2] Mathema B, Kurepina N E, Bifani P J, et al. Molecular Epidemiology of Tuberculosis: Current Insights. Clin Micro boil Rev2006;19: 658-85.

[3] Kumar V, Abbas AK, Fausto N, Mitchell RN. Robbins Basic Pathology. 8th ed. Saunders Elsevier, 2007. p. 516-22.

[4] WHO (2011). "Global Tuberculosis Control 2011". World Health Organization. Retrieved 15 April 2012.

[5] Young DB, Perkins MD, Duncan K & Barry CE. Confronting the scientific obstacles to global control of TB. J ClinInvest. 2008;118(4):1525-65.

[6] Marsh BJ, San Vicente J, von Reyn CF. Utility of dual skin tests to evaluate tuberculin skin test reactors of 10 to 14 mm in health care workers. Infect Control Hosp Epidemiol 2003; 24:821-4.

[7] Lalvani A, Pathan AA, McShane H et al. Rapid detection of Mycobacterium tuberculosis infection by enumeration of antigen-specific T cells. Am J Respir Crit Care Med 2001;163, 824-8.

[8] Roth VR, Garrett DO, Laserson KF, Laserson KF, Starling CE, Kritski AL, Medeiros EA, Binkin N, Jarvis WR. A multicenter evaluation of tuberculin skin test positivity and conversion among health care workers inBrazilian hospitals. Int J Tuberc Lung Dis. 2005;9:1335-42.

[9] Kang YA, Lee HW, Yoon HI, Cho B, Han SK, Shim YS, et al. Discrepancy between the tuberculin skin test and the wholeblood IFN-γ assay for the diagnosis of latent TB infection in an intermediate TB-burden country. JAMA 2005;293:2756-61.

[10] Nienhaus A, Schablon A, Loddenkemper R, Hauer B, Wolf N, Diel R. Prevalence of latent tuberculosis infection in health care workers in geriatric care. Pneumologie. 2007; 61(9):613-6.

[11] Abbas M, AlHamdan N, FialaFA,et al. Prevalence of Latent TB among Health Care Workers in Four Major Tertiary Care Hospitals in Riyadh, Saudi Arabia. J Egypt Public Health Assoc. 2010; 85(1):2-10.

[12] Nienhaus A, Gariepy PK, Trouve C, Lhaumet C, Toureau J, & Peters C. Tuberculosis screening at the Sainte-Anne Hospital in Paris-Results of first and second IGRA. J OccupMedToxicol. 2014;9:24.

[13] National Tuberculosis Program (NTP) (2006). The annual report of the National Tuberculosis Program of Yemen.

[14] Naidoo S, Jinabhai CC. TB in health care workers in KwaZulu-Natal, South Africa. Int J Tuberc Lung Dis 2006; 10(6):676-82.

[15] He GX, van den Hof S, van der Werf MJ, Wang GJ, Ma SW,et al. Infection control and the burden of tuberculosis infection and disease in health care workers in china: a cross-sectional study. BMC Infect Dis2010;10(1): 313.

[16] Jo KW, Hong Y, Park JS, Bae IG, Eom JS, Lee SR, et al. Prevalence of Latent TB Infection among Health Care Workers in South Korea: A Multicenter Study Tuberc Respir Dis. 2013;75:18-24.

[17] Schablon A, Beckmann G, Harling M, Diel R, Nienhaus A. Prevalence of latent tuberculosis infection among health care workers in a

hospital for pulmonary diseases. J Occup Med Toxicol. 2009;4:1-7.

[18] Lawn SD, Zumla AI. "Tuberculosis". Lance 2011; 378 (9785):57-72. doi:10.1016/S0140-6736 (10) 62173-3.

[19] Möller M, Hoal, EG. "Current findings, challenges and novel approaches in human genetic susceptibility to tuberculosis". Tuberculosis 2010; 90 (2):71-83.

[20] Klimuk D, Hurevich H, Harries AD, Babrukevich A, Kremer K, Van den Bergh R, et al. Tuberculosis in health care workers in Belarus. Public Health Action 2014; 4(3): S29–S33.

[21] Torres Costa J, Silva R, Ringshausen FC and Nienhaus A. Screening for tuberculosis and prediction of disease in Portuguese health care workers. J Occup Med Toxicol 2011; 6,19.

[22] Harada N, Nakajima Y, Higuchi K, Sekiya Y, Rothel J, et al. Screening for TB infection using whole-blood IFN-γ and Mantoux testing among Japanese health care workers. Infection control and hospital epidemiology 2006; 27(5): 442-8.

[23] Nienhaus A, SchablonA, Le Bâcle C, Siano B, Diel R. Evaluation of the IFN-γ release assay in health care workers. Int Arch Occup Enviro Health 2006;81:295-300.

[24] Claassens M, Schalkwyk C, Toit E, Roest E, Lombard C, Enarson D, et al. Tuberculosis in Health care Workers and Infection Control Measures at Primary Health care Facilities in South Africa. PLOS One 2013;8 (10): e76272

[25] Baussano I, Nunn P, Williams B, Pivetta E, BugianiM, et al. Tuberculosis among health care workers. Emerg Infect Dis 2011; 17(3): 488-94.

[26] Buregyeya E, Nuwaha F, Wanyenze RK, Mitchell EM, Criel B et al. Utilization of HIV and Tuberculosis Services by Health Care Workers in Uganda: Implications for Occupational Health Policies and Implementation. PLOS ONE 2012; 7(10):e46069.

[27] Tripodi D, Brunet-Courtois B, Nael V et al. Evaluation of the tuberculin skin test and the IFN-γ release assay for TB screening in French health care workers. J Occup Med Toxicol. 2009;4, 30

[28] Larcher C, Frizzera E, Pretto P, Lang M, Sonnleitner N, Huemer HP. Immunosurveillance for Mycobacterium tuberculosis of health care personnel in a third level care hospital. Med Lav 2012; 103(1), 26-36.

[29] Freeman JT, Marshall RJ, Newton S et al. Screening for Mycobacterium tuberculosis infection among health care workers in New Zealand: prospective comparison between the tuberculin skin test and the Quanti-FERONTB Gold In-Tube assay. NZMed J. 2012; 125(1349), 21-9.

[30] Menzies D. What does tuberculin reactivity after BCG vaccination tell us? Clin Infect Dis 2000; 31:S71-4.

[31] Diagnostic Standards and Classification of Tuberculosis in Adults and Children. This official statement of the American Thoracic Society and the Centers for Disease Control and Prevention was adopted by the ATS Board of Directors, This statement was endorsed by the Council of the Infectious Disease Society of America, September 1999. Am J RespirCrit Care Med. 2000;161:1376-95.

[32] Pai M, Denkinger CM, Kik SV, et al. Gamma interferon release assays for detection of Mycobacterium tuberculosis infection. Clin Microbiol Rev. 2014;27(1):3-20.

[33] Schluger NW, Burzynski J. Recent advances in testing for latent TB. Chest. 2010;138:1456-63.

[34] Katial RK, Hershey J, Purohit-Seth E, Belisle JT, Brennan PJ, Spencer JS, et al. Cell-Mediated Immune Response to Tuberculosis Antigens: Comparison of Skin Testing and Measurement of In Vitro Gamma Interferon Production in Whole-Blood Culture. Clin Diag Lab Immunol 2001; 8(2):339-45.

[35] Diel B, Loddenkemper R, Meywald-Walter K, Gottscholk R, and Nienhaus A. Comparative Performance of Tuberculin Skin Test, QuantiFERON-TB-Gold In Tube Assay, and T-Spot. TB test in Contact Investigations for Tuberculosis. Chest 2009; 135:1010-8.

[36] Pottumarthy S, Morris A, Harrison A, and Wells V. Evaluation of the tuberculin IFN-γ assay: potential to replace the Mantoux skin test. J. Clin. Microbiol 1999;37:3229-32.

[37] He GX, Zhao YL, Jiang GL, Liu YH, Xia H, Wang SF, Wang LX, Borgdorff MW, van der Werf MJ, van den Hof S. Prevalence of tuberculosis drug resistance in 10 provinces of China. BMC Infectious Diseases 2008;8:166.

[38] Nienhaus A, Ringshausen FC, Costa JT, Schablon A & Tripodi D. IFN-γ release assay versus tuberculin skin test for monitoring TB infection in healthcare workers. Expert Review of Anti-infective Therapy. 2013;11:1, 37-48.

[39] Pai M, Kalantri S, Menzies D. Discordance between tuberculin skin test and IFN-γ assays. Int J Tuberc Lung Dis 2006;10(8):942-4.

[40] Diel R, Loddenkemper R, Niemann S, MeywaldWalter K, Nienhaus A. Negative and positive predictive value of a whole-blood IFN-γ release assay for developing active tuberculosis: an update. Am J Respir Crit Care Med 2011; 183(1):88-95.

[41] Pai M, Zwerling A, Menzies D. Systematic review: Tcell-based assays for the diagnosis of latent tuberculosis infection: an update. Ann. Intern. Med. 2008;149(3),177-84.

[42] Diel R, Loddenkemper R, Nienhaus A. Evidence-based comparison of commercial IFN-γ release assays for detecting active TB: a meta-analysis. Chest 2010; 137(4):952-68.
[43] Sester M, Sotgiu G, Lange C et al. IFN-γ release assays for the diagnosis of active tuberculosis: a systematic review and meta-analysis. Eur. Respir J 2011;37 (1):100-11.
[44] Lalvani A, Millington KA. T cell-based diagnosis of childhood tuberculosis infection. Curr Opin Infect Dis 2007; 2:264-71.
[45] Lalvani A, Pareek M: Interferon gamma release assays: principles and practice. Enferm Infec Microbiol Clin 2010; 28:245-52.
[46] Mack U, Migliori GB, Sester M, et al. LTBI: latent tuberculosis infection or lasting immune responses to M.TB? ATBNET consensus statement. Eur Respir J2009; 33:956-73.