

# Quantiferon Assay versus Tuberculin Skin Test in Detection of Latent TB Among Patients on Haemodialysis

Ibrahim M. Shalan<sup>1</sup>, Haitham A. Azeem<sup>2</sup> and kabil T. H.<sup>3</sup>

Departments of chest<sup>1</sup>, internal Medicine<sup>2</sup> and microbiology and immunology<sup>3</sup>  
Faculty of Medicine Al-Azhar University (Assiut).

Accepted 20<sup>th</sup> May, 2013

## Abstract

**Background:** Patients with end-stage renal disease (ESRD) and Mycobacterium tuberculosis infection pose a high risk of developing active TB disease. It is therefore important to detect latent TB infection (LTBI) to offer treatment and prevent progression to TB disease.

**Objective:** to compare the performance of Quantiferon-TB Gold test (QFT-G) in the diagnosis of LTBI with tuberculin skin test (TST) among patients on haemodialysis.

**Patients and methods:** sixty patients on hemodialysis at Al-Azhar and Assiut university hospitals were subjected to TST and QFT-G. The agreement between both tests was evaluated with respect to age, sex, BCG vaccination and history of contact to a known TB case.

**Results:** The percentage of positive TST results and positive QFT-G test results was 45% & 31.7% respectively. The overall agreement between both tests was 44/60 (73.3%) and it was statistically significant (P=0.001). BCG vaccination had no effect on either TST or QFT-G results. Positive TST and QFT-G results were 90% and 100 % respectively in patients with history of contact to known TB cases. Both tests were not significantly related to age or sex.

**Conclusions:** Both QFT-G and TST may be complementary to each other in the diagnosis of LTBI in hemodialysis patients.

**Keywords:**Quantiferon TB gold test- TST- latent TB- haemodialysis.

## Introduction

End stage renal disease (ESRD) patients are at increased risk of latent tuberculosis infection (LTBI) compared to the general population. LTBI is more likely to progress to TB infection in ESRD. Investigations from several countries have shown that the increased risk of TB among patients on long-term dialysis is 6.9 to 52.5 times higher than the rate in the general population (Hussein et al, 2002). Although guidelines recommend screening these patients for LTBI, the tuberculin skin test (TST) is believed to be insensitive in ESRD patients and TST negativity rate is high. False-negative TST of ESRD patients might be due to the immune compromised condition (Taggart et al, 2004; Hayriye et al, 2011). In addition, the TST might be falsely positive in persons with a history of previous nontuberculous mycobacterium (NTM) infection or BCG vaccination (Wang et al, 2002) Immune response to mycobacterial infections is primarily a cell-mediated immune response to M. tuberculosis antigens. When the T cells are stimulated by purified protein derivative (PPD) from M. tuberculosis, they produce the cytokine gamma interferon (IFN- $\gamma$ ), which allows macrophages to kill the intracellular pathogen (Luis et al, 2012). The Quantiferon-TB assay detects the in vitro cell-mediated immune response to M. tuberculosis infection

by measuring IFN- $\gamma$  in whole blood that was incubated with M tuberculosis PPD. An enzyme-linked immunosorbent assay (ELISA) detects the amount of IFN- $\gamma$  produced by the T cells (WHO, 2009).

The enhanced QFT-G test assay uses early secretory antigenic 6 kDa (ESAT-6) and culture filtrate protein 10 (CFP-10), The T SPOT-TB assay, uses ESAT6 and CFP10, and detects INF  $\gamma$ - producing T cells in peripheral blood mononuclear cells (PBMCs) based on ELISPOT (Anderson and Doherty, 2005).

## Aim of the work

The aim of this study is to compare the performance of Quantiferon-TB Gold test in the diagnosis of latent tuberculosis infection with tuberculin skin test among patients undergoing hemodialysis.

## Patients and Methods

This study was conducted on sixty patients undergoing hemodialysis at Al-Azhar and Assiut university hospitals during the period from *January 2011 to September 2012*. All the patients were subjected to:

-*Detailed history taking* particularly history of contact to a known tuberculous patient and history of BCG vaccination documented by BCG scar.

## -Full clinical examination.

**Tuberculin skin test (TST):** TST was performed by intradermal injection of 0.1 ml of PPD (5 units) into the skin of the forearm. The test was then read 48 to 72 hours later. The presence of induration was determined by touch. The diameter of the induration was measured transversely. Erythema is not considered. The size of induration was measured in millimeters, 10 mm or more was considered as positive results (CDC, 2006). QuantiFERON TB Gold test (QFT-G): The QFT-G system uses three specialized blood collection tubes that contain antigens representing certain M. tuberculosis proteins (ESAT-6, CFP-10 and TB 7.7) as well as positive (Mitogen) and negative (Nil) controls. The Mitogen tube can be used with the QFT-G test as a positive control. This may be especially warranted where there is doubt as to the individual's immune status, so it is specially used in Immune compromised patients (CDC, 2006). The following steps were performed according to package insert of QuantiFERON®-TB Gold (In-Tube Method), manufactured by Cellestis Limited (Australian public company):

a) **Incubation of whole blood with antigen:** 1 ml of blood was taken by venipuncture directly into each of the collection tubes. Shaking the tubes vigorously for 5 seconds (10 times) was done to ensure that the entire inner surface of the tube has been coated with blood. Tubes were labeled appropriately. The tubes were then transferred to a 37°C incubator as soon as possible within 12 hours of collection. Incubation of the blood for 16-24 hours, after which plasma was separated and tested for the presence of IFN-gamma produced in response to the peptide antigen.

b) **Measurement of IFN- $\gamma$  by ELISA:** Following 16 to 24 hours incubation, the tubes were centrifuged at 5000 rpm for 10-15 minutes and plasma was collected. The concentration of IFN-gamma was determined by enzyme linked immune sorbent assay (ELISA). The QFT-G ELISA uses a recombinant human IFN-gamma standard, which has been assayed against a reference IFN- $\gamma$  preparation. Prior to assay, plasma was mixed to ensure that IFN- $\gamma$  is evenly distributed throughout the sample. 50 $\mu$ L of freshly prepared working strength conjugate was added to the required ELISA wells using a multichannel pipette. 50 $\mu$ L of test plasma samples was added to appropriate wells using a multichannel pipette. Finally 50 $\mu$ L of each of the four standards were added to standard wells. Mixing the conjugate and plasma/standards thoroughly was done using a microplate shaker (Sorin, Biomedica) for 1 minute. Then the plate was covered with lid and

incubated at room temperature (22 °C  $\pm$  5 °C) for 120 minutes, without direct exposure to sunlight. The wells were washed with 400  $\mu$ L of working strength wash buffer for at least 6 cycles by automated plate washer (Stat-Fax 2600). 100  $\mu$ L of enzyme substrate solution to each well were added and mixed thoroughly by a microplate shaker. The plate was covered with a lid and incubated at room temperature for 30 minutes without exposure to direct sunlight. Then 50  $\mu$ L of enzyme stopping solution was added to each well and mixed. The plate was finally read at 450 nm using reference filter at 630 nm (Stat-Fax 2100) (Gerogianni et al, 2008).

c) **Interpretation of results:** The software performed a Quality control assessment of the assay and generated a standard curve for reading a test result for each subject. A positive result suggests that M. Tuberculosis is likely; a negative result suggests that infection is unlikely.

**Statistical methods:** Statistical Package for Social Science (SPSS) program version 16 was used for analysis of data. Data were presented as number, (percent) and mean + SD. Chi-square ( $\chi^2$ ) test was used to compare qualitative data. Significant correlation between different variables was determined using Pearson's correlation coefficient. The agreement between TST and QFT-G test results was assessed using Kappa coefficient.

## Results

**Table (1): Demographic Characteristics of the studied hemodialysis patients**

Demographic data	Patients group(N=60)
<b>SEX (NO. %)</b>	
MALE	33 (55%)
FEMALE	27 (45%)
<b>Age (years)</b>	
Rang (years) (mean $\pm$ SD)	(39-70) 55.17 $\pm$ 14.87
<b>History of BCG vaccinations (No. %):</b>	
Yes	40 (66.7%)
No	20 (33.3%)
<b>History of contact to T.B patient (no.%)</b>	9 (15%)
<b>Duration of haemodialysis (months) (mean <math>\pm</math>SD)</b>	36 $\pm$ 15

**Table (2): Results of TST and QFT-G in the studied patients**

Results	Tuberculin Skin-test (TST)	Quantiferon-TB Gold Test (QFT-G)
Positive	27 (45%)	19 (31.7%)
negative	33 (55%)	41 (68.3%)

**Table (3): Results of TST and QFT-G in the studied hemodialysis patients with respect to age**

Test		No. (%)	Age (Mean ± SD)	P value
TST	Positive	27 (45%)	52.6 ± 16.1	0.65
	Negative	33 (55%)	55.7 ± 14.2	
QFT-G	Positive	19 (31.7%)	55.9 ± 12.1	0.52
	Negative	41 (68.3%)	53.8 ± 16.7	

**Table (4): Results of positive TST and QFT-G in in the studied hemodialysis patients with respect to gender**

Gender	TST Positive (n=27)	QFT-GPositive (n=19)
Female	12/27 (44.4%)	10/27 (37%)
Male	15/33 (45.4%)	9/33 (27.2%)
P value	0.32	0.05

**Table (5): Results of positive TST and QFT-G in in the studied hemodialysis patients with respect to BCG vaccination**

BCG	TST Positive (n=27)	QFT-GPositive (n=19)
Non vaccinated (n=20)	8 (40%)	6 (30%)
Vaccinated (n=40)	19 (47.5%)	13 (32.5%)
P value	0.22	0.55

**Table (6): Results of positive TST and QFT-G in in the studied hemodialysis patients with respect to history of contact to known TB cases**

History of contact to TB	TST Positive (n=27)	QFT-G Positive (n=19)
No (n=51)	19 (37.25%)	10 (19.6%)
Yes (n=9)	8 (88.9%)	9 (100%)
P value	0.001	0.01

**Table (7): Agreement of TST and QFT-G in in the studied hemodialysis patients**

Test		TST (induration cut-off 10 mm)		
		Positive	Negative	Total
QFT-G	Positive	15 (25%)	4 (6.7%)	19 (31.7%)
	Negative	12(20%)	29 (48.3%)	41 (68.3%)
	Total	27(45%)	33 (55%)	60 (100%)
Kappa (k) P value		Agreement (15 + 29)/60 = 73.3% (**? = 0.44) p= 0.001.		

\*\*k = moderate agreement

**Table (8): Agreement of TST and QFT-G Test in BCG non-vaccinated hemodialysis patients**

Test		TST (induration cut-off 10 mm)		
		Positive	Negative	Total
QFT-G	Positive	5 (25%)	1 (5%)	6 (30%)
	Negative	3(15%)	11(55%)	14 (70%)
	Total	8(40%)	12 (60%)	20 (100%)
Kappa (k) P value		Agreement (5 + 11)/20= 80% (**? = 0.56) p= 0.001.		

**Table (8): Agreement of TST and QFT-G Test in BCG non-vaccinated hemodialysis patients**

Test		TST (induration cut-off 10 mm)		
		Positive	Negative	Total
QFT-G	Positive	10(25%)	3 (7.5%)	13 (32.5%)
	Negative	9(22.5%)	18(45%)	27 (67.5%)
	Total	19(47.5%)	21 (52.5%)	40 (100%)
Kappa (k) P value		Agreement (5 + 18)/40= 70% (**? = 0.37) p= 0.001.		

\*k = fair agreement

## Discussion

In the current study on 60 hemodialysis patients, the percentage of positive TST results (induration cut off 10 mm) and positive QFT-G test results was found to be 45% & 31.7% respectively (table 2). Our results are in accordance with Emin et al, (2011) who reported in their study that the positivity of QFT-G was 39.6%, however TST positivity differed according to cut off value. Therefore, the sensitivity of the QFT- G for the diagnosis of latent tuberculosis infection might be lower than that of the TST among studied groups. However Inoue et al., (2009) reported that the sensitivity and specificity of the QFT in hemodialysis patients was 100% and 89.7% respectively. In the present study the overall agreement between the QFT-G and the TST in hemodialysis patients was 44/60 (73.3%) and it was found to be statistically significant with moderate agreement (k=0.44 & p=0.001), (Table 7). These results were found to be in accordance with many researchers as Hayriye et al, (2011) who found moderate agreement between the TST and QFT-G assay results when TST induration is above 10 mm and Emin et al, (2011) who found fair to moderate agreements between QFT-G test and TST in either a TST cut off value of 5 mm and 10 mm in cases with renal failure receiving HD.

In previous studies determining the performances of QFT-G test versus the TST for detecting LTBI in ESRD cas-es, the agreement between these tests were variable. However, the agreement was not better than a fair result. In a recent study from Turkey, Seyhan et al, (2010) reported an overall agreement of 65% (concordance k=0.26) between the QFT-G and the TST at a cut off value of 10 mm. Lee et al, (2009) reported the overall agreement between QFT-G and TST as fair (k=0.34) when the TST cut off was defined as 5 mm and fair (k=0.25) when it was defined as 10 mm. They concluded that the QFT-G was a more accurate method for identifying those truly infected with Mycobacterium tuberculosis, even in BCG-vaccinated individuals. Also, the agreement between TST and QFT-G was poor (k=0.16, p=0.116) in Triverio et al, (2009) study.

They defined TST positive if an induration diameter was >5 mm and found that QFT-G was twice as effective at detecting probable LTBI (46%) as TST (25%). Another study by Winthrop et al, (2008) tested 100 patients with ESRD who received hemodialysis and they reported that QFT-G and T-SPOT-TB might offer a better method for detecting TB infection in ESRD patients. In our study the rate of BCG vaccination was 66.7% (table 1), similar result were reported by Emin et al, (2011). However previous studies revealed a BCG vaccination rate between 77.6% and 90%

(Ozdemir et al, 2007; Soysal et al, 2008; Elbek et al, 2009). In the present study the overall agreement between the QFT-G and the TST in BCG non vaccinated hemodialysis patients was 16/20 (80%) and it was found to be statistically significant with moderate agreement (k=0.56 & p=0.001), (Table 8). As regard to BCG vaccination the agreement between the two tests was fair (28/40, 70%, k=0.37 & p=0.003) (9). These results were found to be in accordance to Emin et al, (2011) who found in their study on 64 BCG vaccinated cases with renal failure receiving HD that the agreements between QFT-G test and TST in either a TST cut off value of 5 mm and 10 mm to be moderate and fair (k=0.432 & k= 0.229 respectively).

In non-vaccinated 29 cases, the overall agreement between the two tests were both fair (k=0.322, & k=0.228, respectively) for TST cut off value of 5 mm and 10 mm respectively. In our study we found that, BCG vaccination had no effect on either TST or QFT- G results in hemodialysis patients. Emin et al, (2011) found in their study that BCG vaccination had no effect on TST, however QFT- G was found to be significantly more positive in BCG non vaccinated cases. Hayriye and coworkers (2011) reported a history of BCG vaccination in (80%) of cases, however no statistically significant difference was detected in the TST and QFT-G test results between vaccinated and unvaccinated patients (P value =0.221 ; 0.454 respectively).

This might be explained by the fact that, in their study group, only 2 cases were positive by QFT-G: one of them was BCG vaccinated and the other one was not (Hayriye et al, 2011). In addition, Franken et al, (2007) found no significant relation between BCG vaccinated patients and TST and QFT-G test results. In the current study positive TST and QFT-G results were found to be 90% and 100 % respectively in hemodialysis patients with history of contact to known TB cases (table 6). Connell et al, (2011) compared the performance of TST and QFT-G test in a total of 75 subjects who were contacts of active tuberculous patients and found that concordant results between the two tests existed as patients with positive TST had positive QFT-G test and patients with negative TST had negative QFT-G. In our study both QFT-G and TST results were not significantly related to age. However our results were found to be in contrary to Emin et al, (2011); Porsa et al, (2006) who stated that elder patients were associated with positive QFT-G result. Passalent et al, (2007) found that in patients with end-stage renal disease undergoing dialysis, a positive T-SPOT.TB was strongly associated with age. Tegbru et al, (2006) compared TST conversion and reactivity among adults with and without HIV in total of 1286 study subjects, they found that the age was not significantly different between the studied

groups. Also Vinton et al, (2009) found that positive TST result was associated with older age.

As regards sex distribution, in the present study there was no statistically significant relation between the sex and results of TST or QFT (p value 0.032 and 0.05 respectively) (table 4). Similarly, Emin et al, (2011) found none significant relation between the two tests regarding sex. On the other hand, our results disagree with Tegbaru et al, (2006), who found more positive results in males than females. This difference may be attributed to the limited number of patients in our study group (60 patients) compared to their study which was performed on 1286 individuals. There is no diagnostic gold standard for LTBI. So, we could not assess the sensitivity and specificity of QFT-G for this disease state.

The only diagnostic standard for LTBI is the eventual development of active TB, and the predictive value for progression of TB can be ascertained only through longitudinal cohort studies that follow clinical outcomes of tested individuals. In the study reported by Diel et al, (2011), they showed that QFT-G is a more accurate indicator of the presence of LTBI than the TST and provides at least the same sensitivity for detecting individuals who will progress to active TB. Also, high sensitivity and specificity of QFT-G tests in active tuberculosis disease is reported. In a meta-analysis done by Pai and O'Brien, (2008) revealed that the pooled sensitivity was 78% (CI, 73% to 82%) for QFT-G test, and 70% (CI, 63% to 78%) for QFT-G test In-Tube. The pooled specificity for both QuantiFERON tests was 99% among non-BCG-vaccinated participants (CI, 98% to 100%) and 96% (CI, 94% to 98%) among BCG-vaccinated participants. They concluded that the Interferon gamma release assays (IGRAs), especially QFT-G test and QFT-G test In-Tube, have excellent specificity that is unaffected by BCG vaccination.

## Conclusions

QFT-G test shows moderate agreement with TST (73.3%) so the two tests may be complementary to each other in the diagnosis of latent tuberculosis infection in hemodialysis patients. Therefore QFT-G could be used in screening of LTBI simultaneously with TST; but its relatively high cost and need for laboratory infrastructure is a limitation of its usage.

## Recommendations

Hemodialysis patients should be screened routinely for tuberculosis and latent tuberculosis infection. Further prospective studies are required to determine whether the QFT-G is more sensitive than the TST in HD patients in LTBI. We are in need for a gold standard test for detection of latent TB infection.

## References

- Anderson P and Doherty TM (2005): The success and failure of BCG: implications for a novel tuberculosis vaccine. *Nat. Rev. Microbiol.* 3: 656-662.
- Centers for Disease Control and Prevention (2006): Emergence of Mycobacterium tuberculosis with extensive resistance to second-line drugs worldwide, 2000. *MWR* 55(11): 301-305.
- Connell DW, Singanayagam A, Charif R, George PM, Molyneux PL, McCrudden C, Magtoto M, Harden E, Seneviratne SL, Duncan ND, Kon OM (2011): A comparison between interferon gamma release assays and the tuberculin skin test in the contact tracing of patients with chronic kidney disease. *Thorax*, 66:729-730.
- Diel R, Loddenkemper R, Niemann S, Meywald-Walter K, Nienhaus A. (2011): Negative and positive predictive value of a whole-blood interferon- $\gamma$  release assay for developing active tuberculosis: an update. *Am J Respir Crit Care Med.* Jan 1; 183(1):88-95.
- Elbek O, Uyar M, Aydin N, Bórekçi S, Bayram N, Bayram H, Dikensoy O (2009): Increased risk of tuberculosis in patients treated with antitumor necrosis factor alpha. *Clin Rheumatol*; 28(4):421-6.
- Emin Maden, T. Taha Bekci, Recep Kesli, Huseyin Atalay, Turgut Teke, Yalcin Solak, Suleyman Turk, Kursat Uzun, Ramazan Koylu (2011): Evaluation of performance of quantiferon assay and TST in end stage renal disease patients receiving hemodialysis. *New Microbiologica*, 34, 2-356,
- Franken WP, Timmermans JF, Prins C, Slietman EJ, Dreverman J, Bruins H, van Dissel JT, Arend SM. (2007): Comparison of Mantoux and QuantiFERON TB Gold tests for diagnosis of latent tuberculosis infection in Army personnel. *Clin Vaccine Immunol.* Apr; 14(4):477-80.
- Gerogianni I, Papala M, Klapsa D, et al. (2008): Whole-blood interferon- $\gamma$  assay for the diagnosis of tuberculosis infection in an unselected Greek population. *Respirol.* 13: 270-274.
- Hayriye Sayarlioglu, Mustafa Gol, Canan Eren Dagli, Ekrem Dogan, Murat Şahin, Mehmet Ali Uğur, Nurhan Koksak, Mehmet, Sayarlioglu, Mumtaz Kerim Tahta (2011): Quantiferon-TB Gold test for screening latent tuberculosis infection in hemodialysis patients. *Tuberkulozvetoraksdergisi*; 59(2): 105-110
- Hussein MM, Mooij MJ, Roujouleh H (2002): Tuberculosis and chronic renal disease. *Semin Dial*; 16: 38-44.
- Inoue T, Nakamura T, Katsuma A, Masumoto S, Minami E, Katagiri D, et al. (2009): The value of QFT-G in the diagnosis of tuberculosis among dialysis patients. *Nephrol Dial Transplant*; 24: 2252-7.
- Lee SS, Chou KJ, Su IJ, Chen YS, Fang HC, Huang TS, et al. (2009): High prevalence of latent TB infection in patients in end-stage renal disease on hemodialysis: comparison of QFT-G, ELISPOT, and tuberculin skin test. *Infection*; 37: 96-102.
- Luis Anibarro<sup>1</sup>, Matilde Trigo<sup>2</sup>, Diana Feijoó, Mónica Ríos, Luisa Palomares, Alberto Pena<sup>1</sup>, Marta Núñez, Carlos Villaverde and África González - Fernández (2012): Value of the tuberculin skin testing and of an interferon-gamma release assay in haemodialysis patients after exposure to M. tuberculosis Anibarro et al. *BMC Infectious Diseases*, 12:195
- Ozdemir D, Annakkaya AN, Tarhan G, Sencan I, Cesur S, Balbay O, Guclu E. (2007): Comparison of the tuberculin skin test and the quantiferon test for latent Mycobacterium tuberculosis infections in health care workers in Turkey. *Jpn J Infect Dis. May*; 60(2-3):102-5.
- Pai M, O'Brien R. (2008): New diagnostics for latent and active tuberculosis: state of the art and future prospects. *Semin Respir Crit Care Med.* Oct; 29(5):560-8.

16. Passalent L, Khan K, Richardson R, Wang J, Dedier H, Gardam M. (2006): Detecting latent tuberculosis infection in hemodialysis patients: a head-to-head comparison of the T-SPOT. TB test, tuberculin skin test, and an expert physician panel. *Clin J Am SocNephrol*; 2:68-73.
17. Porsa E, Cheng L, Seale M, et al. (2006): Comparison of a New ESAT-6 /CFP-10 Peptide-Based Gamma Interferon Assay and a Tuberculin Skin Test for Tuberculosis Screening in a Moderate-Risk Population. *Clin. and Vacc. Imm.* 53-58.
18. Seyhan EC, Sökücü S, Altın S, Günlüoğlu G, Trablus S, Yılmaz D, Koksalan OK, Issever H. (2010): Comparison of the QFT-G In-Tube test with the tuberculin skin test for detecting latent tuberculosis infection in hemodialysis patients. *Transpl Infect Disease* 12(2):98-105.
19. Soysal A, Bakir M, Millington KA, Deeks JJ, Efee S, Aslan Y, Dosanjh DP, Lalvani A. (2008): Prognostic value of a T-cell-based, interferon-gamma biomarker in children with tuberculosis contact. *Ann Intern Med.* Dec 2; 149(11):777-87.
20. Taggart EW, Hill HR, Ruegner RG et al. (2004): Evaluation of an in vitro assay for gamma interferon production in response to *Mycobacterium tuberculosis* infections. *ClinDiagn. Lab. Immunol.*, 11 (6):1089-1093.
21. Tegbaru B, Wolday D, Messele T, Legesse M, Mekonnen Y, Miedema F, van Baarle D. (2006): Tuberculin skin test conversion and reactivity rates among adults with and without human immunodeficiency virus in urban settings in Ethiopia. *Clin Vaccine Immunol.* Jul; 13(7):784-9
22. Triverio PA, Bridevaux PO, Roux-Lombard P, Niksic L, Rochat T, Martin PY, et al. (2009): Interferon-gamma release assays versus TST for detection of latent tuberculosis in chronic haemodialysis patients. *Nephrol Dial Transplant*; 24: 1952-6.
23. Vinton, P., et al. (2009): "Comparison of QuantiFERON-TB Gold In-Tube Test and tuberculin skin test for identification of latent *Mycobacterium tuberculosis* infection in healthcare staff and association between positive test results and known risk factors for infection." *Infection Control & Hospital Epidemiology* 30.3 215-21.
24. Wang L, Turner MO, Elwood RK, Schulzer M, FitzGerald JM. (2002): A meta-analysis of the effect of Bacille-Calmette-Guerin vaccination on tuberculin skin test measurements. *Thorax*; 57: 804-9.
25. Winthrop KL, Nyendak M, Calvet H, Oh P, Lo M, Swarbrick G, et al. (2008): Interferon-gamma release assays for diagnosing *mycobacterium tuberculosis* infection in renal dialysis patients. *Clin J Am SocNephrol*; 3: 1357-63.
26. World Health Organization, editor. *Global tuberculosis control (2009): surveillance, planning, financing.* Geneva: World Health Organization. p. 1-303.