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TITLE PAGE

QuantiFERON-TB Gold and TST are both useful for latent TB screening in autoimmune diseases

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ABSTRACT

Screening for active tuberculosis (TB) and latent TB (LTBI) is mandatory prior to initiation of TNFα inhibitors therapy. However, no agreement exists on the best strategy for detecting LTBI in this population.

The aim of this study was to analyze the performance of tuberculin skin test (TST) and QuantiFERON®-TB Gold In Tube (QFT) on LTBI detection in subjects with immunomediated inflammatory diseases (IMID).

TST and QFT were prospectively performed in 398 consecutive IMID subjects, 310 (78%) on immunosuppressive therapy and only 16 (4%) Bacillus Calmette-Guérin (BCG)-vaccinated.

Indeterminate results to QFT were found in 5 (1.2%). Overall 74/393 subjects (19%) were TST+ and 52 (13%) QFT+. Concordance between TST and QFT results was good (87.7%) (kappa 0.55): 13 were QFT+/TST- and 35 QFT-/TST+. By multivariate analysis both tests were associated significantly with older age. Just TST was associated with BCG vaccination and radiological lesions of past TB. Use of immunosuppressive drugs differently modulated QFT or TST scoring.

Use of the QFT test, as a screening tool for LTBI among IMID subjects, is feasible. Until further data will elucidate discordant TST/QFT results, a strategy of simultaneous TST and QFT testing in a low prevalence BCG-vaccinated population, should maximize potentials of LTBI diagnosis.

Abstract word count: 200

Key words: Interferon-γ release assay, latent tuberculosis infection, tuberculin test, rheumatic diseases, TNFα inhibitors.

INTRODUCTION

One-third of the world's population is believed to harbor latent tuberculosis infection (LTBI) [1], defined as presence of quiescent mycobacteria. Approximately 10% of immunocompetent persons with LTBI will ever develop tuberculosis (TB). In countries with a low incidence of TB, such as Italy, the tracing and targeted treatment of LTBI subjects constitutes a major pillar of TB control [2].

Aging of the population and increased use of immunosuppressive treatments, that represent risk factors for progression to active TB, highlight the need for additional strategies to maintain and improve TB control [3].

Tumor necrosis factor α (TNFα) inhibitors are approved for the treatment of immune-mediated inflammatory diseases (IMID) and provide clinical benefit. However, TNFα inhibitors showed an increased risk of serious life-threatening infections, including reactivation of TB infection in patients previously exposed to mycobacteria [4]. Thus, screening for active TB and LTBI has become mandatory prior to initiation of TNFα inhibitors therapy [5, 6]. However, to date, no agreement exists on the best strategy for detecting LTBI in this population, and tuberculin skin test (TST) remains largely used [7-10].

TST attempts to measure cell-mediated immunity in the form of a delayed-type hypersensitivity response to purified protein derivative (PPD). However, TST lacks specificity in populations with high Bacillus Calmette-Guérin (BCG) coverage and non-tuberculous mycobacteria (NTM) exposure. Moreover, TST may have a higher probability to have false negative results in IMID patients than in general population, because of the immune disregulation linked to the disease itself or due to immunosuppressive drugs use [11].

Recently, two new assays have been developed to identify LTBI, QuantiFERON-TB Gold (QFT) and T-SPOT *TB*. They measure antigen-specific Interferon-γ (IFN-γ) secretion by peripheral blood CD4+ T lymphocytes in response to *in vitro* stimulation with early secreted antigenic target (ESAT-6) and culture filtrate (CFP-10) proteins [12, 13] which are more specific for *M. tuberculosis* infection detection than PPD, as they are not shared with BCG substrains or with most environmental mycobacteria [13, 14].

Several unresolved issues remain on the potential clinical use of IFN-γ release assays (IGRAs) and one area of controversy is whether they can be used in immunosuppressed patients as IMID subjects in addition or in alternative to TST. The lack of a gold standard for LTBI diagnosis has hampered the assessment of diagnostic accuracy of IGRAs, and the comparison of these new tests with TST. Therefore it has been suggested that test for LTBI diagnosis can be validated by analyzing the association of positive results with risk factors for TB, and different tests can be compared by evaluating differences in their association with risk factors [15, 16]. We took this approach to compare performance of QuantiFERON TB-Gold In Tube (QFT) (Cellestis Limited, Carnegie, Australia), and TST for the detection of LTBI among IMID subjects coming from a low endemic TB country. Moreover we analyzed the impact of different classes of drugs on the response to these tests.

METHODS

Study population

We enrolled 398 consecutive subjects, with rheumatic diseases (rheumatoid arthritis, psoriasic arthritis, ankylosing spondylitis) or other immunomediated chronic diseases, that required use of biologic drugs (infliximab, etanercept, adalimumab, rituximab). The subjects were recruited at two rheumatological outpatient clinics of

two Florence hospitals, Italy (Careggi University Hospital and Nuovo Ospedale San Giovanni di Dio Hospital), from May 2006 to December 2007 and fulfilled published criteria permitting use of a biologic drugs [6].

Demographic information, data on BCG vaccination, treatment regimens on the last 3 months and risk factors for LTBI were collected by a standardized patients interview by a researcher who was not aware of the results of TST or QFT. We classified treatments in systemic corticosteroids, conventional disease-modifying antirheumatic drugs (DMARDs) (including methotrexate, azathioprine, cyclosporine, leflunomide, cyclophosphamide, hydroxychloroquine) and TNFα inhibitors (infliximab, etanercept or adalimumab).

All subjects underwent chest x-ray. The following characteristics were considered risk factors for LTBI: birth or residence for ≥6 months in a high TB prevalence country (>20 cases per 100'000 inhabitants); history of household TB contact; health care worker who work at facilities where patients with tuberculosis are treated.

Risk factor for progression to active TB were considered: a medically confirmed history of active TB; chest x-ray findings suggestive of tuberculosis history (nodules, fibrotic scars, calcified granulomas, or basal pleural thickening); HIV-positivity; body weight lower than 10% of ideal body weight; presence of comorbidity (diabetes mellitus, silicosis, chronic renal failure/hemodialysis, neoplastic or hematologic diseases); prolonged therapy with corticosteroids (more than 4 weeks) or other immunosuppressive therapy; injection-drug use; previous gastrectomy or jejunoileal bypass [17].

QFT and TST

Blood samples for QFT were obtained immediately before the TST performance. Within 2 to 6 hours of blood draw, the tubes were incubated at 37°C. The same biologist, who was unaware of subjects' characteristics and results of the TST, performed all whole blood IFN-γ ELISAs according to the manufacturer's instructions. Immediately after draw, all participants received by a trained physician an injection of 5 units of PPD (Biocine Test-PPD; Chiron, Siena, Italy). The transverse diameter of induration was measured in millimeters 72 hours later with ballpen method. TST induration was interpreted relative to risk in accordance with published guidelines [5, 18].

Statistical Analysis

Information from the questionnaires, TST and QFT results were entered into Microsoft Excel 2003 software. We assessed the concordance between QFT and TST by computing the κ statistics, with a κ-value of >0.75 representing excellent agreement beyond chance, 0.40-0.75 representing fair to good agreement beyond chance, and <0.40 representing poor agreement beyond chance [19].

Standard univariate analysis was performed to analyze the association of TST and QFT results to TB risk factor, the type of IMID and to the kind of immunosuppressive therapy.

TB risk factors associated with either QFT or TST results in univariate analysis were entered in two separate logistic regression models in which the outcomes were QFT or TST results. In these models we also entered use of systemic corticosteroids, conventional DMARDs and TNFα inhibitors therapy as independent categorical variables.

Finally, to compare the overall diagnostic performance of the two tests, we evaluated whether the association between number of risk factors for LTBI, selected *a priori* according to their prognostic relevance [20], and the odds ratio for testing

positive varied according to the kind of test. To this purpose we used Generalized Estimating Equations (GEEs) to perform a logistic regression that accounted for the association between results of QFT and TST performed on the same subject. Statistical analysis was performed with SPSS 15.0 for Windows (SPSS Inc., Chicago, Ill, USA) and STATA 9.2 (Stata, College Station, TX, USA).

RESULTS

Studied population

A total of 398 consecutive subjects with IMID were enrolled (Figure 1). Indeterminate results to QFT were found in 5/398 (1.2%) subjects. No association was found between risk factors for TB and immune suppression (data not shown) and these samples were excluded for further analysis.

Demographic and clinical features of the 393 remaining subjects are reported in Table 1. Our population was characterized by a high median age (54 years), a low prevalence of BCG-vaccination (4.1%) and a small number of subjects coming from countries at high endemia of TB (4.6%). None of enrolled subjects was found to have active TB.

TST and QFT results

Concordance between TST and QFT

Among the 393 subjects analyzed by both TST and QFT tests, 52 resulted positive to QFT and 74 to TST (p=0.4). Among them 306 [77.8%, 95% Confidence Interval (CI) 73.4-81.9] were concordant in their negative results and 39 (10%, 95% CI 7.1-13.3) in their positive results, for an overall concordance of 87.8% (κ= 0,55, p <0.0001, 95% CI 0.44 to 0.66).

TST+/QFT- discordant results were found in 35/393 (8.9%, 95% CI 6.3-12.2) of subjects while TST-/QFT+ results were found in 13/393 (3.3%, 95% CI 1.8-5.6).

Several factors were evaluated to understand their impact on the discordant QFT/TST data obtained (history of TB exposure, sex, age, history of BCG vaccination, underlying rheumatic disease, etc.). Among them, a history of previous BCG vaccination was associated with discordant QFT-/TST+ results (p=0.004) whereas use of TNFα inhibitors with a discordant QFT+/TST- results (p=0.03).

Association with risk factors for LTBI and type of IMID

By univariate analysis both, TST and QFT positivity were significantly associated with older age and history of close contacts with sputum positive TB patients. However, only TST positivity was associated with BCG vaccination, radiological lesions suggestive of past TB and origin from countries at high endemia of TB whereas only QFT positivity was significantly associated with male sex (Table 1). These associations were confirmed by multivariate analysis (Table 2). No associations between type of IMID and results of TST and QFT were found in univariate analysis (Table 1).

Impact of immunosuppressive therapy

In univariate analysis, regarding to treatment, the proportion of positive scoring was lower for both tests for treated patients (with the exception of those treated with DMARDs or DMARDs associated with steroids) compared to the proportion of positive results in patients who were not on treatment. A significant association, however, was found only between treatment with steroids and TST results (Table 1).

By multivariate analysis, considering the impact of each drug class, the use of DMARDs was not associated with test scoring while use of steroids was associated

with a lower probability of a QFT- or TST-positive scoring (OR=0.4, CI 0.2-09; OR=0.3, CI 0.2-0.6, respectively) (Table 2). Moreover, treatment including TNFα inhibitors significantly decreases the positive outcome of TST (OR=0.3, CI 0.1-0.6) without affecting QFT results.

Comparison of the diagnostic perfomance of TST and QFT

In absence of a gold standard for LTBI diagnosis, we compared the diagnostic performance of the two tests by evaluating the association of the odds of testing positive with the presence of one or more risk factors for LTBI (age older than 50 years, chest X-ray suggestive of past TB, close contact of patients with sputum positive TB and birth/residence in a country with high incidence of TB) (Table 3). The results indicate a clear-cut trend for an increase in odds ratios with increasing number of risk factors for both tests, and show no significant difference between the two tests. Moreover, we computed the prevalence of LTBI in our cohort using different combinations of the two tests used (Table 4). This analysis approach suggests a better single performance of TST than QFT, although the combined used of both tests seems to be the best approach to maximize the sensitivity of the screening.

DISCUSSION

Immunosuppressed subjects are one of the most important targets for the screening of TB infection because of the high risk of progression to active TB. Despite its widely recognized limitations, the TST remains the standard method for identifying TB infection before TNFα inhibitors therapy, due to the lacking of sufficient data on IGRA use in this population [7-10]. To date there were limited data describing

performance of IGRA for detecting LTBI in IMID subjects awaiting TNFα inhibitors therapy, of whom only six reporting direct comparison with the TST (Table 5) [21-28]. Moreover, the published results were not easily comparable, depending on the small number of enrolled subjects, high rate of BCG vaccination in the studied population, not homogeneous type of underlying diseases considered, and kind of IGRA used (Table 5) [21-28].

To our knowledge, this is the largest study comparing the results of an IGRA to TST for TB screening in IMID subjects coming from a low endemic TB country (only 4.6% subjects coming from an high TB prevalence country) and with low BCG vaccination rate (4.1%).

The first relevant finding of our work was a high positive rate at both TST and QFT testing (18.8% and 13%, respectively). This result was probably due to a high median age of IMID population (55.7% aged more than 50 years old), most likely exposed to *M. tuberculosis* infection in young age when Italy was a high TB prevalence country. Therefore, we confirmed the need for TB screening in IMID subjects also in an actually low TB prevalence country such Italy.

Our study showed a lower rate (1.2%) of indeterminate results at QFT testing compared to the other previous published studies utilizing the same test (QuantiFERON-TB Gold In Tube) [21, 23, 24, 27], where the rate of indeterminate results ranged between 1.5 and 10.3% (Table 5). Our finding confirms the feasibility of QFT in the IMID population, often undergoing at LTBI screening already strongly pre-treated - 79% in our population - with immunomodulating drugs (steroids, DMARDs, TNFα inhibitors).

A fair concordance between QFT and TST (87.5% with κ= 0.55, p <0.0001, 95% CI 0.43-0.67) was observed, higher than reported in the previous studies in which the concordance ranged from 51.7% to 82.8% (Table 5). Probably this is due

to the lower rate of BCG-vaccinated subjects that led to a reduction of the rate of TST+/QFT- discordant results compared to the literature, with the only exception of the report by Ponce de Leon *et al*. [27]. This interpretation is supported by authors showing a significant association between TST+/IGRA- results and BCG vaccination status by the multivariate analysis [23, 28]. Our multivariate analysis confirmed this association although our population accounted for only 16 BCG-vaccinated subjects. Therefore these discordant results most likely represent TST associated to a TB infection. The alternative possibility of a past sensitization to NTM antigens present in the TST and not in the QFT assay is difficult to ascertain.

Regarding the 13 TST-/QTF+ discordant subjects (3.3%), since the high specificity of QFT for *M. tuberculosis* infection, we may speculate that TST missed these LTBI diagnoses. Probably, the two main reasons to explain this finding are: first, we did not perform a two-step TST which would have maximized the TST sensitivity, but this strategy would have required 4 visits instead of 2 for every subject. Second, all TST-/QTF+ discordant subjects were under treatment with immunosuppresive drugs, and 6 of 13 with TNFα inhibitors which may explain the negative results of TST, considering the negative impact of steroid and TNFα inhibitors on *M. tuberculosis*-specific responses [29] and on TST performance (Table 2). Moreover no associations between any LTBI tests and kind of IMID were found (Table 1). Our results confirm those reported by Sellam and colleagues and it is consistent with the lacking of relationship with TB infection risk [22].

Regarding the association of TST and QFT with risk factors for TB infection our data showed a good association for both tests with known risk factors for TB in Italy, such as older age and history of close contacts of sputum positive TB patients. However, TST seems to be more strictly associated with TB infection, considering that only this test was associated significantly with other known risk factors as origin

from countries at high endemia of TB and radiological lesions suggestive for TB history.

Establishing sensitivity and specificity for *M. tuberculosis* infection is difficult due to the absence of a "gold standard" for the diagnosis of LTBI. Thus an approach to index test validation that goes beyond the diagnostic accuracy paradigm is needed for an alternative evaluation process in the absence of a gold standard. In some studies the association of positive results with risk factors for TB has been analyzed, and different tests has been compared by evaluating differences in their association with risk factors [15, 16, 20]. We used this approach to assess the accuracy of the TST and QFT in detecting LTBI, comparing their results to a gradient of risk factors for LTBI selected *a priori* according to their prognostic relevance. Our findings showed a clear-cut trend for an increase in odds with growing number of risk factors for LTBI, confirming that both, TST and QFT are predictive of LTBI without difference in estimation ability between the two tests (Table 3). Based on previous studies that showed a reduced performance of TST in immunosuppressed RA subjects [12, 27], we analyzed the impact of different classes of drugs on the response to TST and QFT tests. By multivariate analysis, we found that steroids significantly affected both QFT- and TST-positive outcome (OR=0.4 and 0.3, respectively), while treatment including TNFα inhibitors significantly decreases only the positive outcome of TST (OR=0.3). This result disagrees with that reported by Matulis and colleagues. However, these authors did not evaluate the association with TST results (TST was not performed simultaneously with QFT in that study) [23]. They explained their result with the TNFα inhibitors decrease the activation of CD4+ T lymphocytes by mycobacterial antigens. However, our results seems to be more consistent with the crucial role that TNFα plays in recluting macrophages in tuberculin-induced delayed-type hypersensitivity that should justify the reduced response to TST in our

population of whom 24.2% were already exposed to TNFα inhibitors at the time of screening [29].

In conclusion, our study confirm that QFT, as a screening tool for LTBI among IMID subjects awaiting TNFα inhibitors therapy, is feasible, due to the low incidence of indeterminate results and the strict association with risk factor for TB of this test. However, our findings do not support the recent published UK national guidelines on TB suggesting the use of IGRA as a confirmatory test in TST-positive subjects [30]. Based on these data in a country at low prevalence of BCG-vaccination like Italy, this approach would have had the risk of missing subjects with LTBI. On the other hand, our data suggest the potential use of QFT, as primary test may offer the advantage of being less impaired by the treatment with TNFα inhibitors compared to TST. In this context, large-scale cohort studies are needed to estimate risk for progression to active disease in persons who had TST and/or IGRA positive results, with particular interest on the risk for disease in persons with discordant reactions. Until further data are available on the implication of discordant TST/QFT results, a strategy of simultaneous TST and QFT testing in population at low prevalence of BCG vaccination, should maximize the sensitivity for LTBI diagnosis.

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Figure 1. Flow diagram of the study population. QFT = QuantiFERON-TB Gold In-Tube; TST = tuberculin skin testing.

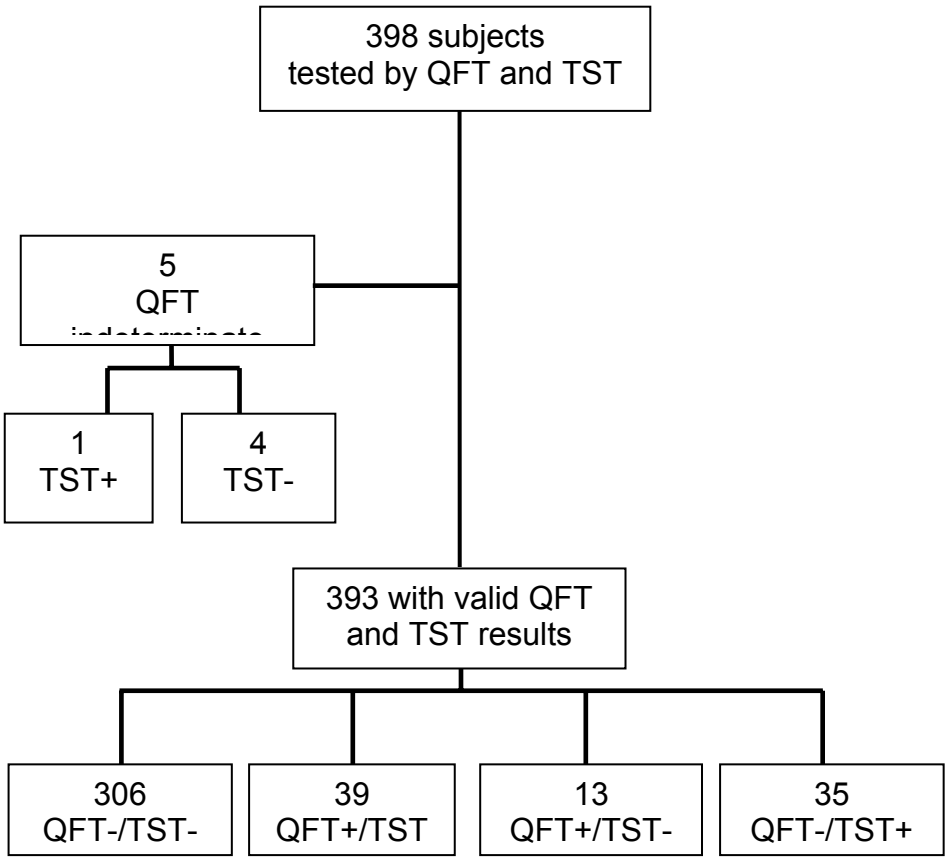

Table 1. Associations of demographic, epidemiological, clinical characteristics and therapy with QFT- and TST-positive results in subjects with reumathic diseases (univariate analysis).

	Total N. (%)	**QFT-positive N. (%)**	**p***	**TST-positive N. (%)**	**p***
Total	393 (100)	52 (13.2)		74 (18.8)	
Gender			0.03		0.09
Male	137 (34.9)	25 (18.2)		32 (23.4)	
Female	256 (65.1)	27 (10.5)		42 (16.4)	
BCG vaccinated			0.1		0.009
Yes	16 (4.1)	0		7 (9.5)	
No	377 (95.9)	52 (100.0)		67 (90.5)	
Birth/residence in countries at high prevalence of TB			0.7		0.05
Yes	18 (4.6)	2 (3.8)		7 (9.5)	
No	375 (95.4)	50 (96.2)		67 (90.5)	
Age in years			0.001		0.03
≤ 29	45 (11.5)	1 (2.2)		2 (4.4)	
30 – 49	129 (32.8)	12 (9.3)		25 (19.4)	
50 - 69	167 (42.5)	25 (15)		35 (21.0)	
> 70	52 (13.2)	14 (26.9)		12 (23.1)	
Chest X-ray patterns			0.08		0.009
No lesions of past TB	80 (20.3)	6 (7.5)		14 (17.5)	
Lesions of past TB	41 (10.5)	9 (22.0)		15 (36.5)	
Other patterns	272 (69.2)	37 (13.5)		45 (16.5)	
Close contacts of patients with sputum positive TB			0.02		0.02
Yes	7 (1.8)	3 (5.7)		4 (5.4)	
No	386 (98.2)	49 (94.3)		70 (94.6)	
Diagnosis			0.3		0.4
Rheumatoid arthritis	201 (51.1)	27 (13.4)		35 (17.4)	
Psoriasic arthritis	94 (23.9)	23 (24.5)		16 (17)	
Ankilosing spondilytis	58 (14.8)	5 (8.6)		11 (19)	
Other †	40 (10.2)	4 (10)		5 (12.5)	
Therapy					
No therapy (in last 3 months)	83 (21.1)	13 (25)	---	22 (29.7)	---
DMARDs	75 (19.1)	12 (23.1)	0.9	19 (25.7)	0.8
DMARDs + steroids	106 (27)	13 (25)	0.5	22 (29.7)	0.3
Steroids	34 (8.7)	2 (3.8)	0.1	1 (1.3)	0.004
TNFα inhibitors	30 (7.6)	6 (11.5)	0.2	4 (5.4)	0.1
TNFα inhibitors + DMARDs	25 (6.4)	4 (7.6)	0.9	4 (5.4)	0.2
TNFα inhibitors + steroids	4 (1)	0	0.3	0	0.2
TNFα inhibitors + DMARDs + steroids	36 (9.2)	2 (3.8)	0.1	2 (2.7)	0.009

Definition of abbreviations: QFT: QuantiFERON-TB Gold In Tube, TST: tuberculin skin test, TNFα: Tumor Necrosis Factor α, DMARD: disease-modifying antirheumatic drugs, TB: tuberculosis, BCG: Bacillus Calmette-Guérin

* By chi-square test or Fisher's exact test as appropriate; for therapy regimens p are derived from comparison of test results in patients with each single regimen and those of untreated patients.

† Includes uveitis, Crohn disease, juvenile chronic arthritis, undifferentiated spondyloarthropathy (three each); Behçet's disease, Still's disease, entesitis, temporal arteritis, sclerodermia, seronegative oligoarthritis, seronegative spondyloarthritis, ulcerative colitis, (two each); systemic lupus erythematosus, systemic sclerosis, erythema nodosum, Wegener's granulomatosis, polymyalgic syndrome, sarcoidosis, reactive arthritis, psoriasis, myopathy of unclear origin, familial mediterranean fever, sacroileitis, seronegative spondyloarthritis with Castleman's disease (one each).

Table 2. Risk factors associated with QFT- and TST-positivity in subjects with rheumatic diseases: multivariate analysis.

	QFT-positive		TST-positive	
	Odds ratio (95% CI)	**P value**	**Odds ratio (95% CI)**	**P value**
Male	2.1 (1.1-4.2)	0.01	1.65 (0.9-2.9)	0.08
BCG vaccinated	NA*	NA*	3.8 (1.0-13.9)	0.04
Age in years				
≤29	reference		reference	
30-49	2.8 (0.3-23.4)	0.3	8.6 (1.0-69.7)	0.04
50-69	6.8 (0.8-53.0)	0.06	14.7 (1.8-117.2)	0.01
>70	15.9 (1.9-132.0)	0.01	13.2 (1.5-112.7)	0.01
Chest X-ray patterns				
No lesions of past TB	reference		reference	
Lesions of past TB	1.3 (0.5-3.2)	0.5	3.0 (1.3-6.8)	0.005
Other patterns	0.4 (0.1-1.2)	0.1	1.6 (0.7-3.4)	0.1
Immunosuppressive therapy including				
Steroids	0.4 (0.2-0.9)	0.03	0.3 (0.2-0.6)	0.002
DMARD	1.1 (0.5-2.3)	0.7	1.7 (0.9-3.4)	0.09
TNFα inhibitors	0.9 (0.4-2)	0.8	0.3 (0.1-0.6)	0.004
Close contacts of patients with sputum positive TB	5.3 (1-28)	0.04	6.5 (1.1-36.9)	0.03
Birth/residence in countries at high prevalence of TB	1.1 (0.2-5.8)	0.8	3.7 (1.1-12.3)	0.03

Definition of abbreviations: QFT: QuantiFERON-TB Gold In Tube; TST: tuberculin skin test; TNFα: Tumor Necrosis Factor α; DMARD: disease-modifying antirheumatic drugs; TB: tuberculosis; CI: confidence interval; BCG: Bacillus Calmette-Guérin.

* NA: not available.

Table 3. Association between number of risk factors for TB infection and QFT- and TST-positivity

	QFT-positive		TST-positive		P value†
	Odds ratio (95% CI)	**P value**	**Odds ratio (95% CI)**	**P value**	
Number of risk factors*					
0	1		1		
1	3.30 (1.48 – 7.39)	0.004	2.57 (1.35-4.90)	0.004	0.730
≥ 2	5.71 (2.13-15.31)	0.001	5.35 (2.34-12.20)	<0.001	

Definition of abbreviations: QFT: QuantiFERON-TB Gold In Tube, TST: tuberculin skin test, CI: confidence interval, BCG: Bacillus Calmette-Guérin.

* Age ≥50 years; Chest X-ray suggestive of a past TB; Close contact of patients with sputum positive TB; Birth/residence in a country with high incidence of TB.

† p-value for the conjoint hypothesis of no difference in the estimates of the two models.

Table 4. LTBI prevalence estimate using different approaches

Test considered	Estimated rate (95% CI)
Only TST +	18.8% (14.9-22.7)
Only QFT +	13.2% (9.8-16.5)
QFT+ and TST+	9.9% (6.9-12.8)
QFT+ among TST+	9.9% (6.9-12.8)
At least one test +	22.1% (18-26.2)

Definition of abbreviations: CI: Confidence Interval; TST: Tuberculin Skin Test; QFT: QuantiFERON-TB Gold In Tube.

Study, year	Country	IGRA kind	Participants n°	BCG %	Indeterminate %	κ	Concordants results		Discordants resullts	
							TST+/ IGRA+ N (%)	TST-/ IGRA- N (%)	TST-/ IGRA+ N (%)	TST+/ IGRA- N (%)
Cobanoglu *et al.*, 2007	Turkey	QFT-G IT	68 cases 38 controls	100	10.3 5.3	0.14 -0.05	8 (13.1) 0	23 (37.7) 23 (63.8)	1 (1.6) 1 (2.8)	29 (47.5) 12 (33.3)
Sellam *et al.*[†], 2007	French	EliSPOT*	35 cases 33 controls	100	0	---	na[†]	na[†]	na[†]	na[†]
Matulis *et al.*[‡], 2008	Swiss	QFT-G IT	142	83	6	0.16	10 (7)	60 (44.7)	5 (3.5)	34 (25.4)
Pratt *et al.*, 2006	UK	QFT-G IT	101	78.5	9.9	---	na[§]	na[§]	na[§]	na[§]
Kobashi *et al.*[‖], 2007	Japan	QFT-G	252	60.3	12.6	0.29	30 (13.6)	120 (54.5)	6 (2.8)	64 (29.1)
Dinser *et al.*, 2008	Germany	Flow cytometric assay	97	5.1	0	0.31	6 (6.2)	74 (76.3)	10 (10.3)	7 (7.2)
Ponce de Leon *et al.*, 2008	Peru	QFT-G IT	101 cases 93 controls	80.2 80.6	1.9 0	0.37 0.55	21 (20.8) 16 (17.2)	50 (49.5) 61 (65.6)	24 (23.8) 5 (5.4)	6 (5.9) 11 (11.8)
Vassiloupolos *et al.*, 2008	Greece	T SPOT TB	70	40	0	0.38	12 (17.1)	39 (55.7)	4 (5.7)	15 (21.5)
Bocchino *et al.*, 2008	Italy	QFT-G IT T SPOT TB	69 69	2.8 2.8	2.8 5.8	0.57 0.48	14 (20.9) 12 (18.5)	41 (61.2) 39 (60)	8 (11.9) 9 (13.8)	4 (6) 5 (7.7)
Greenberg *et al.*, 2008	USA	QFT-G	61 cases 42 controls	27.8 23.3	11.5 2.4	---	na**	na**	na**	na**
Our data	Italy	QFT-G IT	398	4.1	1.5	0.55	39 (10)	306 (77.8)	13 (3.3)	35 (8.9)

Table 5. Concordance of IGRA and TST for the screening of TB infection in IMID population

Definition of abbreviations: IGRA: Interferon-γ release assay; QFT-G: QuantiFERON-TB Gold (CFP-10 and ESAT-6); QFT-G IT: QuantiFERON-TB Gold In Tube (CFP-10, ESAT-6 and TB-7); BCG: Bacillus Calmette-Guérin.

* Home made EliSPOT with antigens CFP-10 and ESAT-6.

† No direct comparison between two test.

‡ TST analyzed retrospectively. The median time between performing the TST and the QFT assay was 102 days (range 7–184).

§ na = not available (TST not done)

‖ Not homogeneous population including different underlying diseases (74 subjects had malignant diseases, 72 were undergoing immunosuppressive treatment with steroids and/or TNFα inhibitors, 52 had diabetes mellitus, 50 had chronic renal failure, 4 had HIV infection).

** No direct comparison between two tests.