ORIGINAL ARTICLE

Risk Assessment of Tuberculosis in Immunocompromised Patients
A TBNET Study

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Abstract

Rationale: In the absence of active tuberculosis, a positive tuberculin skin test (TST) or interferon-γ release assay (IGRA) result defines latent infection with *Mycobacterium tuberculosis*, although test results may vary depending on immunodeficiency.

Objectives: This study compared the performance of TST and IGRAs in five different groups of immunocompromised patients, and evaluated their ability to identify those at risk for development of tuberculosis.

Methods: Immunocompromised patients with HIV infection, chronic renal failure, rheumatoid arthritis, solid-organ or stem-cell transplantation, and healthy control subjects were evaluated head-to-head by the TST, QuantiFERON-TB-Gold in-tube test (ELISA), and T-SPOT.*TB* test (enzyme-linked immunospot) at 17 centers in 11 European countries. Development of tuberculosis was assessed during follow-up.

Measurements and Main Results: Frequencies of positive test results varied from 8.7 to 15.9% in HIV infection (n = 768), 25.3 to 30.6% in chronic renal failure (n = 270), 25.0% to 37.2% in rheumatoid arthritis (n = 199), 9.0 to 20.0% in solid-organ transplant recipients (n = 197), 0% to 5.8% in stem-cell transplant recipients (n = 103), and 11.2 to 15.2% in immunocompetent control subjects (n = 211). Eleven patients (10 with HIV infection and one solid-organ transplant recipient) developed tuberculosis during a median follow-up of 1.8 (interquartile range, 0.2–3.0) years. Six of the 11 patients had a negative or indeterminate test result in all three tests at the time of screening. Tuberculosis incidence was generally low, but higher in HIV-infected individuals with a positive TST (3.25 cases per 100 person-years) than with a positive ELISA (1.31 cases per 100 person-years) or enzyme-linked immunospot result (1.78 cases per 100 person-years). No cases of tuberculosis occurred in patients who received preventive chemotherapy.

Conclusions: Among immunocompromised patients evaluated in this study, progression toward tuberculosis was highest in HIV-infected individuals and was poorly predicted by TST or IGRAs. Clinical trial registered with www.clinicaltrials.gov (NCT 00707317).

Keywords: interferon-γ release assays; immunocompromised; TBNET; tuberculin-skin test; tuberculosis

At a Glance Commentary

Scientific Knowledge on the Subject: Patients with immunodeficiencies are particularly vulnerable to progression from latent infection with *Mycobacterium tuberculosis* to active disease. Tuberculin skin test and interferon-γ release assays are recommended for risk assessment for the future development of tuberculosis, but knowledge of their performance in immunocompromised patients is limited.

What This Study Adds to the Field: Tuberculin skin test and interferon-γ release assay responses vary substantially among different groups of patients with immunodeficiencies and are poor predictors for the development of tuberculosis in immunocompromised patients.

Tuberculosis is a leading cause of morbidity and mortality worldwide, and is the 10th cause of all recorded deaths (1). In the absence of an effective vaccine, tuberculosis prevention relies on early case finding, infection control measures, and preventive chemotherapy of individuals latently infected with *Mycobacterium tuberculosis* (2). In clinical practice, latent infection with *M. tuberculosis* (LTBI) is defined by the presence of an adaptive immune response against antigens of *M. tuberculosis* in individuals without evidence of active tuberculosis, and is either determined *in vivo* by the tuberculin skin test (TST) or *ex vivo* by interferon-γ release assays (IGRAs) (3). At present, two IGRAs are commercially available: the QuantiFERON-TB-Gold in-tube test (Qiagen, Hilden, Germany), an ELISA; and the T-SPOT.*TB* test (Oxford Immunotec, Abingdon, UK), an enzyme-linked immunospot assay (ELISPOT). A positive TST or IGRA result is assumed to identify individuals with the highest risk of progression to tuberculosis (4). In most cases in patients from low-incidence countries, tuberculosis is caused by reactivation of LTBI, and could have been avoided by preventive chemotherapy following targeted TST or IGRA testing (5). However, apart from individuals from defined tuberculosis risk groups, immunodiagnosis for LTBI and preventive chemotherapy is not generally recommended, because the positive predictive value of the TST and IGRAs for progression toward tuberculosis in the general population is very low (4, 6), especially in the absence of prior *M. tuberculosis* exposure, and preventive chemotherapy is not efficacious outside of risk groups (7).

The risk for tuberculosis is directly related to exposure with *M. tuberculosis* and patients with immunodeficiencies are particularly vulnerable to progression from LTBI to active disease (8, 9). These include HIV-infected individuals, transplant recipients, patients with chronic renal failure, or patients undergoing tumor necrosis factor antagonist therapies. In general, the risk for tuberculosis in immunocompromised persons is influenced by the underlying immunologic mechanisms and degree of immunodeficiency, and by the duration of and temporal relationship with previous *M. tuberculosis* exposure (8, 9). Because timing of exposure outside of contact tracing is generally unknown, a positive immunodiagnostic test result is considered as a proxy of prior *M. tuberculosis* exposure. In addition, the results of immunodiagnostic testing may depend on the nature and extent of the immunodeficiency and may not equally predict the risk for tuberculosis among different groups of immunocompromised patients. Because a positive test in the clinical setting is a finding on which a clinician needs to act, increased knowledge of performance and limitations of the currently available immunodiagnostic tests and their ability to predict tuberculosis in immunocompromised patients is needed.

In previous studies, the evaluation of immunodiagnostic tests for LTBI was mostly restricted to individual patients groups (10–13) or individual assays (14–19). Our study included five different groups of

immunocompromised patients from 17 centers in 11 European countries, and tested all three immunodiagnostic tests in parallel. The two main objectives were to characterize the performance of three immunodiagnostic tests for LTBI in a patient population with various etiologies of immunodeficiency, and to assess the risk of developing tuberculosis in patients with a positive immunodiagnostic test at the time of screening.

Methods

Recruitment of Study Population

Study participants were recruited from 17 European healthcare facilities; 18 years of age and older; and diagnosed with HIV infection, chronic renal failure, rheumatoid arthritis, solid-organ transplantation, or stem-cell transplantation. All local ethics committees approved the study and written informed consent was obtained from all individuals. Patients were recruited from June 1, 2008 to May 31, 2011 in a consecutive manner as part of their routine care, and followed up for the development of tuberculosis. We additionally included adult control patients at the same facilities from noninfectious disease departments who did not have an immunocompromizing clinical condition, and had a low risk for *M. tuberculosis* exposure.

Study Design and Data Collection

The study included a cross-sectional part where data on demographic and clinical parameters, and data related to the risk of *M. tuberculosis* exposure were obtained through a structured questionnaire. In addition, all participants had the three separate tests for LTBI administered (3). Experimental and clinical data were recorded electronically and transmitted to the coordinating center, where they were assessed for inconsistencies or missing entries. In the prospective part of the study, follow-up information on the occurrence of tuberculosis after initial testing was actively sought and collected by personal patient contact by the treating physician.

Technical Procedures, Data Sources, and Exposure Variables

The TST according to the Mantoux technique (20) and the two IGRAs (the ELISPOT, T-SPOT.*TB*; and the ELISA, QuantiFERON-TB-Gold in-tube) were performed as described in the online supplement. The laboratory technicians were fully masked to the *M. tuberculosis* exposure status of the participants. The TST could be performed a maximum of 30 days before the IGRAs.

M. tuberculosis exposure was defined by a reported history of either exposure to *M. tuberculosis*, active tuberculosis, tuberculosis treatment, LTBI, or chemoprophylaxis for LTBI, or being a resident in a high tuberculosis incidence country for at least 1 year. For stratified analyses of subgroups, CD4 cell count was dichotomized at 200 cells per microliter (21). Duration of dialysis was dichotomized at 5 years, and the time after solid-organ transplantation at 1 year. In addition, a drug score was determined as specified in the METHODS section of the online supplement to quantify the level of immunosuppression in transplant recipients. In patients with rheumatoid arthritis, severity was classified according to the disease activity score (DAS; group I, <3.3; group II, 3.3–5.1; group III, >5.1) (22).

Statistical Analysis

The agreement between the proportions of positive tests was assessed by Cochran-Q test for matched samples (23). Agreement between tests was assessed by the Kappa statistics (24). The association between test results and *M. tuberculosis* exposure was assessed by logistic regression in which confounding was deemed present if the relationship between test result and exposure changed by more than 10% after inclusion of an exploratory variable. The risk of tuberculosis by initial test results was assessed using the Kaplan-Meier estimate in survival analysis assessing patient follow-up data at the earliest date of the last clinical assessment, diagnosis of tuberculosis, death, or end of follow-up (either 1, 2, or 5 yr after testing). Only patients with at least 30 days of follow-up were included in the analysis to consider very early tuberculosis as a screening failure rather than a valid case of incident tuberculosis.

Results

Study Population

A total of 1,537 patients with immunocompromizing medical conditions (Figure 1) and 211 immunocompetent control subjects were enrolled. Causes of immunodeficiency included chronic HIV infection (n = 768), chronic renal failure (n = 270), rheumatoid arthritis (n = 199), solid-organ transplantation (n = 197; 134 renal, 41 lung, 17 liver, 4 renal-pancreas, and 1 renal-liver transplants), and stem-cell transplantation (n = 103) (Table 1). Information on the distribution of patients by country is given in Table E1 in the online supplement.

Cross-Sectional Study: Indeterminate and Positive Test Results

TST, ELISA, and ELISPOT test results were available from cross-sectional analysis of 1,495 (97.3%), 1,532 (99.7%), and 1,503 (97.8%) patients, and 211 (100%), 208 (98.6%), and 209 (99.1%) control subjects, respectively. The percentages of

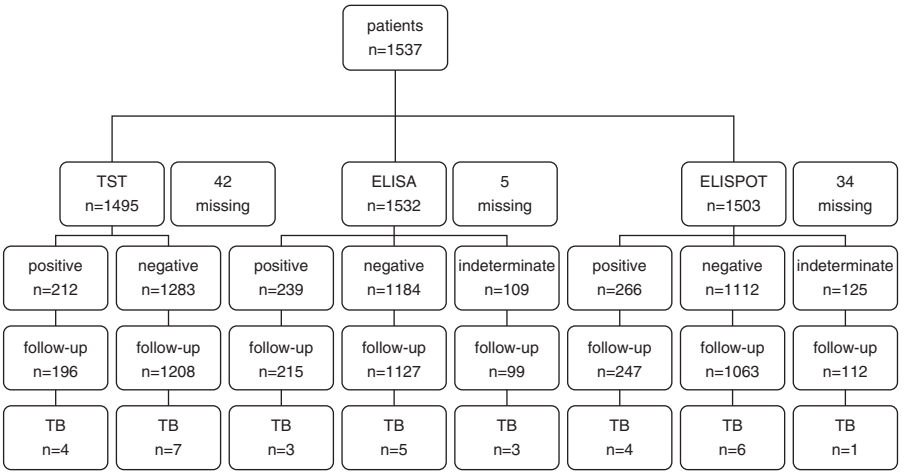

Figure 1. Flow chart of test results and cases of active tuberculosis (TB) in patients included in the study. All patients with at least 30 days of follow-up were included to assess development of TB on follow-up. ELISPOT = enzyme-linked immunospot assay; TB = tuberculosis; TST = tuberculin skin test.

Table 1. Demographic Characteristics of Patients and Control Subjects

	HIV n = 768		CRF n = 270		RA n = 199		SOT n = 197		SCT n = 103		Control Subjects n = 211	
	n	%	n	%	n	%	n	%	n	%	n	%
Female	203	26.4	97	35.9	154	77.4	83	42.1	41	39.8	134	63.5
Age (median, IQR)	40.8	33.5–48.4	62.5	49.0–73.3	55.7	45.4–63.6	56.5	46.4–63.9	57.1	44.8–64.6	23.3	20.1–43.8
White	709	92.3	261	96.7	192	96.5	191	97.0	97	94.2	208	98.6
Immigrant	101	13.2	42	15.6	14	7.0	22	11.2	12	11.7	6	2.8
Years since immigration (median, IQR)	9.0	3.0–16.0	22.5	11.5–38.5	20.0	8.0–30.0	38.5	26.0–42.0	33.0	21.5–40.5	27.0	23.0–32.0
Mycobacterium tuberculosis exposure*	187	24.3	66	24.4	87	43.7	66	33.5	24	23.3	0	0.0
History of exposure to *M. tuberculosis*	44	5.7	28	10.4	19	9.6	31	15.7	3	2.9	0	0.0
History of active TB	62	8.1	19	7.0	9	4.5	11	5.6	2	1.9	0	0.0
History of TB treatment	58	7.6	13	4.8	6	3.0	6	3.1	1	1.0	0	0.0
History of LTBI	13	1.7	1	0.4	20	10.1	31	15.7	0	0.0	0	0.0
History of LTBI chemoprophylaxis	8	1.0	1	0.4	14	7.0	6	3.1	18	17.5	0	0.0
>1 yr in high TB–incidence country	94	12.2	28	10.4	55	27.6	10	5.1	3	2.9	0	0.0
Valid results in all three tests†	635	82.4	245	90.7	164	82.4	145	73.6	69	67.0	197	93.4

Definition of abbreviations: CRF = chronic renal failure; HIV = HIV-infected patients; IQR = interquartile range; LTBI = latent infection with *M. tuberculosis*; RA = rheumatoid arthritis; SCT = stem-cell transplantation; SOT = solid-organ transplantation; TB = tuberculosis.
*Self-reported evidence of prior "*M. tuberculosis* exposure" defined by a reported history of either exposure to *M. tuberculosis*, active tuberculosis, tuberculosis treatment, LTBI or chemoprophylaxis for LTBI, or being at least for 1 year a resident in a high TB–incidence country; immigrants were only included in the "*M. tuberculosis* exposure group" if patients had lived in high TB–prevalence countries for more than 1 year before immigration.
†Valid results include positive and negative tests and exclude indeterminates.

indeterminate and positive test results are shown in Figures 2A and 2B, respectively. The highest percentage of indeterminate results in the ELISA was observed among solid-organ and stem-cell transplant recipients (Figure 2A; 20.3 and 20.4%, respectively), whereas indeterminate results in the ELISPOT assay were most frequently observed in HIV-infected patients and stem-cell transplant recipients (11.2 and 14.6%, respectively). Indeterminate test results of IGRAs in these patient groups were related to the extent of immunosuppression (*see* Table E2). The percentage of indeterminate results among patients with chronic renal failure or rheumatoid arthritis was only marginally different from that observed in healthy control subjects (Figure 2A). Overall, failure to adequately react toward the positive control stimulus was the cause of indeterminate results in 85.0% of ELISA and 50.4% of ELISPOT samples, whereas excess reactivity in the negative control was the reason for indeterminate results in all other samples.

Percentages of positive test results among all patients with valid results in all three assays showed substantial differences among the groups (Figure 2B). In general, positive tests were most frequently observed in patients with chronic renal failure (25.3–30.6%) and rheumatoid arthritis (25.0–37.2%), which contrasted with results from HIV-infected patients and solid-organ transplant recipients, respectively, where only 8.7–15.9% and 9.0–20% of tests were positive. The percentage of positive tests in HIV-infected patients was lowest in patients with low CD4 T-cell counts, whereas the effect of the level of immunodeficiency in other groups was less evident (*see* Table E3). When comparing the results of the three tests in each group, the percentage of positive test results among HIV-infected patients and solid-organ transplant recipients was lower when using the TST as compared with ELISA and ELISPOT ($P < 0.001$ for HIV-infection; $P = 0.002$ for solid-organ transplant recipients). Among stem-cell transplant recipients, positive tests were generally less frequent and were only observed using the IGRAs. The percentage of positive TST results was significantly higher as compared with that of IGRAs in patients with rheumatoid arthritis ($P = 0.003$), whereas no difference between the tests was found in patients with chronic renal failure ($P = 0.121$) (Figure 2B).

Cross-Sectional Study: Between-Test Agreement and Association with Exposure

When comparing results of the three different tests, the agreement between the two IGRAs was higher than that between either IGRA and the TST (*see* Table E3). Agreement between ELISA and ELISPOT was substantial in patients with chronic renal failure (K = 0.65) and rheumatoid arthritis (K = 0.77), and moderate in HIV-infected individuals (K = 0.50), and solid-organ (K = 0.58) and stem-cell transplant recipients (K = 0.41).

The association between self-reported evidence of prior *M. tuberculosis* exposure and test result was not confounded by age, sex, or group-specific characteristics of immunosuppression for any of the three tests, with the exception of patients with rheumatoid arthritis in which the DAS score confounded this association (*see* Table E4). Moderate associations of positive IGRA results with exposure were found in HIV-infected patients, which reached statistical significance for the ELISPOT (odds ratio [OR], 2.0; 95% confidence interval [CI], 1.2–3.4), and near significance for the ELISA (OR, 1.6; 95% CI, 0.9–3.1). In patients after solid-organ transplantation,

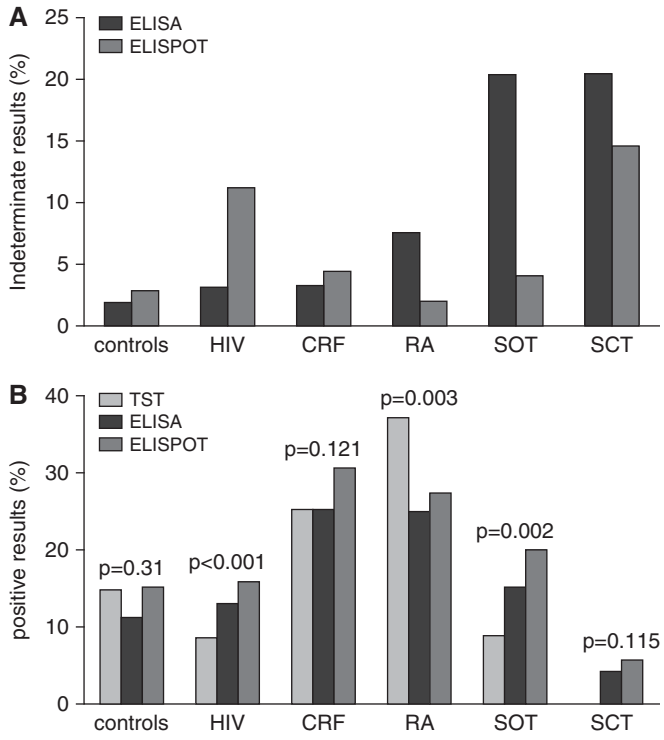

Figure 2. Results of immune-based testing in immunocompromised patients and control subjects. (*A*) The percentage of indeterminate results in the ELISA and the enzyme-linked immunospot (ELISPOT) assay and (*B*) the percentage of positive test results among patients with valid results in all three tests was quantified. CRF = chronic renal failure; RA = rheumatoid arthritis; SCT = stem-cell transplantation; SOT = solid-organ transplantation; TST = tuberculin skin test.

a positive ELISA was significantly associated with exposure (OR, 2.2; 95% CI, 1.0–4.6). A significant association of each of the three tests with exposure existed for patients with rheumatoid arthritis. After adjusting for DAS score, the OR was 2.7 (95% CI, 1.4–5.1) for TST, 4.4 (95% CI, 2.1–9.4) for ELISA, and 4.9 (95% CI, 2.4–10.2) for ELISPOT. Conversely, although the percentage of positive test results in patients with chronic renal failure and rheumatoid arthritis was similarly high (Figure 2B), a positive TST or IGRA in patients with chronic renal failure was not associated with evidence of prior *M. tuberculosis* exposure.

When analyzing the number of positive tests in the three assays in relation to exposure status, the percentage of cases with three negative tests decreased along a gradient of likely exposure, and was highest in individuals nonexposed to *M. tuberculosis* (*see* Table E5). Likewise, the percentage of individuals with three positive tests increased with increasing exposure and ranged from 4.9% in nonexposed to 13.4% in cases where exposure was likely highest. Of note, this also held true for cases where only the two IGRAs were positive. In contrast, the percentage of individuals with only one positive test was rather similar across the groups (*see* Table E5).

Prospective Study: Incidence of Tuberculosis

Of the 1,537 patients, a total of 1,464 (95.3%) had an assessment of tuberculosis on follow-up and were included in the prospective part of the study. Eighteen patients were excluded because of a follow-up of less than 30 days. The number of patients with either a positive or negative test result included in the analysis for tuberculosis incidence was 1,404 for the TST, 1,342 for the ELISA, and 1,310 for the ELISPOT. A total of 11 patients developed active tuberculosis in the median follow-up time of 1.8 years (interquartile range [IQR], 0.2–3.0). Of these, 10 patients were HIV-infected and one was a solid-organ transplant recipient (Table 2). No cases of tuberculosis were observed among patients with chronic renal failure, rheumatoid arthritis, stem-cell transplantation, or control subjects. All patients who developed tuberculosis were born in a medium- or high-prevalence country for tuberculosis and had not received preventive chemotherapy after testing. Among HIV-infected patients, viral load was detectable in all cases (median, 20,593 copies per microliter; IQR, 82.5–49,819 copies per microliter), and median CD4 T-cell counts were 302 per microliter (IQR, 196–370 per microliter) (Table 2). Six out of the 11 patients either had a negative or indeterminate test result at the time of testing and only two had positive test results in all three assays (Table 2).

The incidence of tuberculosis in HIV-infected patients with a positive test was generally higher as compared with a negative test (*see* Table E6). Two years after testing, the incidence was higher in patients with a positive TST result (3.26 cases per 100 person-years) as compared with the ELISPOT (1.80 per 100 person-years) and the ELISA (1.37 per 100 person-years) (*see* Table E6). Similar results were obtained after 1 and 5 years of follow-up (*see* Table E6).

Finally, the influence of preventive chemotherapy administered after testing on progression to tuberculosis was analyzed for all patients after 1, 2, and 5 years (Table 3). Information on preventive chemotherapy by patient group and country is given in Table E1. Among patients, 21.3% (47 of 196) of TST positives, 26.0% (56 of 215) of ELISA positives, and 24.0% (54 of 247) of ELISPOT positives received preventive chemotherapy. No case of tuberculosis occurred in these individuals. Among patients who had not received preventive chemotherapy, the incidence of tuberculosis in individuals with a negative test was expectedly low (0.15, 0.11, and 0.17 per 100 person-years after 2 yr for TST, ELISA, and ELISPOT, respectively). In contrast, the incidence of tuberculosis after a positive TST, ELISA, or ELISPOT was 1.15, 0.71, and 0.88 per 100 person-years after 2 years, respectively. Similar results were obtained after 1 and 5 years of follow-up (Table 3). Taken together, although the incidence of tuberculosis was higher in patients with positive test results, IGRAs were not superior in identifying patients at risk for developing tuberculosis when compared with the TST.

Discussion

We have evaluated currently available immunodiagnostic tests for the identification of LTBI and their value in

Table 2. Characteristics of Patients Developing Active Tuberculosis on Follow-up

	Country of Birth/Actual Residence	Sex	TB (*mo after Testing*)	TB Type	TB Diagnosis	TST (*mm*)	ELISPOT (SFC)	ELISA (*U/ml*)	Chemoprevention		HIV		HIV	CD4
									Offered	Done	CDC	HAART	Load	T Cells/μl
HIV	Argentina/Italy	Male	1.1	Pulmonary	Clinical*	Positive (20)	Positive (18)	Positive (2.7)	No	No	C2	No	25,285	492
HIV	Bulgaria/Bulgaria	Male	1.3	Extrapulmonary	Culture confirmed	Positive (12)	Positive (16)	Positive (0.39)	No	No	C3	Yes	83,176	245
HIV	Portugal/Portugal	Male	2.0	Pulmonary	Culture confirmed	Positive (5)	Positive (125)	Negative (0.15)	Yes	No	A3	No	38,700	22
HIV	Portugal/Portugal	Female	2.5	Pulmonary	Clinical*	Negative (0)	Negative (0)	Negative (0)	No	No	A2	No	15,900	371
HIV	Portugal/Portugal	Male	8.4	Both	Culture confirmed	Negative (0)	Negative (1)	Indeterminate (0.13)	Yes	No	C3	Yes	120,000	50
HIV	Portugal/Portugal	Female	21.8	Both	Culture confirmed	Negative (0)	Negative (4)	Indeterminate (0)	Yes	No	A1	No	38,200	333
SOT	Croatia/Germany	Female	25.2	Extrapulmonary	PCR confirmed	Negative (0)	Negative (0)	Indeterminate (0)	No	No	NA	NA	NA	Unknown
HIV	Portugal/Portugal	Male	33.6	Both	Culture confirmed	Negative (0)	Negative (1)	Negative (0.09)	No	No	A2	No	<50	369
HIV	Eritrea/Germany	Male	35.4	Pulmonary	Clinical*	Negative (0)	Negative (0)	Negative (0.12)	No	No	C3	Yes	60	271
HIV	Argentina/Italy	Male	52.5	Pulmonary	Culture confirmed	Positive (10)	Indeterminate (49)†	Positive (0.7)	No	No	A2	No	10,071	263
HIV	Eritrea/Germany	Male	56.7	Pulmonary	Culture confirmed	Negative (0)	Positive (9)	Negative (0.09)	No	No	A3	Yes	90	354

Definition of abbreviations: ELISPOT = enzyme-linked immunospot assay; HAART = highly active antiretroviral therapy; PCR = polymerase chain reaction; SFC = spot forming cells; SOT = solid-organ transplantation; TB = tuberculosis; TST = tuberculin skin test.
"HIV CDC" refers to the classification by the United States Centers for Disease Control and Prevention (CDC).
*The diagnosis of clinical tuberculosis was based on signs and symptoms of active tuberculosis and response to antituberculosis therapy.
†Indeterminate because of 22 SFC in the Nil control.

Table 3. Follow-up for Active Tuberculosis Depending on Test Result and Prophylaxis 1, 2, and 5 Years after Testing for Latent Infection with *Mycobacterium tuberculosis*

	Test Result*	Prophylaxis†	N	1 yr			2 yr			5 yr		
				PY at Risk	TB Cases	Incidence‡	PY at Risk	TB Cases	Incidence‡	PY at Risk	TB Cases	Incidence‡
TST	Negative	No	1,133	1084.2	2	0.18 (0.05–0.74)	1951.5	3	0.15 (0.05–0.48)	3076.4	7	0.23 (0.11–0.48)
	Negative	Yes	75	73.0	0	0	110.1	0	0	131.2	0	0
	Positive	No	149	144.5	3	2.08 (0.67–6.44)	261.5	3	1.15 (0.37–3.56)	382.4	4	1.05 (0.39–2.79)
	Positive	Yes	47	46.7	0	0	82.8	0	0	105.0	0	0
ELISA	Negative	No	1,079	1036.6	2	0.19 (0.05–0.77)	1864.4	2	0.11 (0.03–0.44)	2895.2	5	0.17 (0.07–0.41)
	Negative	Yes	48	46.6	0	0	77.8	0	0	95.1	0	0
	Positive	No	159	151.8	2	1.32 (0.33–5.27)	282.4	2	0.71 (0.18–2.83)	462.4	3	0.65 (0.21–2.01)
	Positive	Yes	56	55.4	0	0	85.6	0	0	100.5	0	0
ELISPOT	Negative	No	1,024	976.2	2	0.20 (0.05–0.82)	1752.8	3	0.17 (0.06–0.53)	2718.3	6	0.22 (0.10–0.49)
	Negative	Yes	39	37.3	0	0	64.2	0	0	84.7	0	0
	Positive	No	193	183.3	3	1.64 (0.53–5.07)	342.0	3	0.88 (0.28–2.72)	556.7	4	0.72 (0.27–1.91)
	Positive	Yes	54	53.9	0	0	86.3	0	0	103.7	0	0

Definition of abbreviations: ELISPOT = enzyme-linked immunospot assay; PY = person-year; TB = tuberculosis; TST = tuberculin skin test.
*This analysis includes all patients with valid test results (positive or negative), whereas indeterminate test results were not considered in this analysis.
†Prophylaxis refers to treatment administered after the TST/interferon-γ release assays test performed in this study.
‡Incidence is given per 100 PY; the rates refer to the cumulative rates after 1, 2, and 5 years after testing for latent infection with *M. tuberculosis*.

assessing the risk for progression to active tuberculosis in a large cohort of patients with different etiologies of immunodeficiencies in Europe. As most important findings we identified substantial differences in the frequencies of positive *M. tuberculosis*–specific immune responses among patients with HIV infection, rheumatoid arthritis, chronic renal failure, and solid-organ or stem-cell transplants, whereas less prominent variations were observed between the TST, the QuantiFERON-TB-Gold in-tube, and the T-SPOT.*TB* test within these groups. Of note, among immunocompromised patients, the risk for the development of tuberculosis was clearly highest in HIV-infected individuals.

The strength of this large prospective cohort study is that all three immunodiagnostic tests were evaluated in parallel in patients from five groups of different etiologies of immunodeficiencies, which allowed for a direct comparison of results within one study. Unlike skin testing, IGRAs are less influenced by immunosuppression in HIV-infected individuals and in patients after solid-organ or stem-cell transplantation. Similarities in test performance in these two patient groups may result from the fact that both immunodeficiencies primarily affect T cells, and lower percentages of positive test results appear to be caused by either an HIV-related overall decrease in CD4 T-cell counts (11, 25, 26) or by a drug-induced inhibition of T-cell functionality (27). In contrast, higher percentages of positive test results were found in patients with chronic renal failure or rheumatoid arthritis where immunodeficiency is multifactorial and not primarily acting on T cells (28). Patients with rheumatoid arthritis were striking in that the highest percentage of positive test results were found with the TST. However, because between-test agreement was higher among IGRAs, and both IGRAs showed a strong association with prior *M. tuberculosis* exposure, the TST seems to be less specific and seems to measure different subpopulations of patients.

The proportion of indeterminate IGRA results was expectedly lowest in immunocompetent control subjects, whereas up to 20% of indeterminate results were found in patients. Subgroup analyses indicated that a higher percentage was associated with markers of immunodeficiency, such as CD4 T-cell counts less than 200 cells per microliter, or recent transplantation attendant with higher levels of immunosuppression with multiple drugs in solid-organ and stem-cell transplant recipients. The findings in HIV-infected patients are in line with previous reports suggesting that HIV-positive individuals with less than 200 $CD4^+$ T cells per microliter have impaired IGRA response (10, 11, 17, 18, 26). The proportion of indeterminate results in this study was lowest among patients with rheumatoid arthritis and renal failure, which was generally in line with indeterminate result rates in other studies in these patient groups (12, 29–34). Together with the highest percentage of positive test results, this suggests that immunodiagnostic assays in these two patient groups are least affected by the underlying etiology of immunodeficiency.

Tuberculosis incidence rates found after positive tests are in line with estimates from systematic reviews of predictive value of IGRAs for incident active tuberculosis, which have predominantly included nonimmunocompromised individuals during contact tracing (6, 35). Up to now, only a few studies exist that have specifically analyzed progression in immunocompromised patients, and most studies have only used one IGRA without comparison with TST (36). Most studies did not report incidence rates, but the percentage of tuberculosis cases among untreated patients with positive IGRA results ranged from 7 to 20% in HIV-infected patients (17, 19, 37), 5.6% in patients after renal transplantation (15), and 2.6% in patients before tumor necrosis factor antagonist therapy of which only a small number had rheumatoid arthritis (38). In our multicenter European study, incident cases of tuberculosis were lower and were almost exclusively found among HIV-infected patients. Most of these patients did not receive antiretroviral therapy and those who did still had detectable levels of viral replication confirming that antiretroviral therapy alone reduces the risk for the development of tuberculosis in HIV-infected patients substantially (39, 40). Although positive test results were generally more frequently observed with IGRAs as compared with the TST, progression to tuberculosis also occurred in patients with a negative result in any of the three tests. In half of the tuberculosis cases, LTBI was not detectable by any of the tests at the time of screening and positive results in all three tests were only observed in 2 of 11 patients who subsequently developed tuberculosis. Results from the current study further show that a higher percentage of positive test results in a given group of immunocompromised patients does not indicate a substantially higher risk for progression to tuberculosis. This is illustrated by the fact that no cases of active tuberculosis were observed among patients with renal failure or patients with rheumatoid arthritis not receiving preventive chemotherapy, although these were the patient groups with the highest percentage of positive test results. In contrast, the risk of developing tuberculosis was highest for patients with HIV infection, although the frequency of positive test results was substantially lower. These findings indicate that in immunocompromised patients, none of the three tests is sufficient to assess the risk of progression to tuberculosis.

A recent study in military recruits indicated that individuals where all three tests were positive had a higher epidemiologic risk of prior infection, whereas individuals with only one positive test were suggested to be likely false-positive (41). In line with these findings, the percentage of individuals with positive test results in all three assays or with two positive IGRAs in our study was also highest in cases with highest likelihood of exposure. The fact that the percentage of patients where only one test was positive was less strikingly associated with exposure variables may be considered as a hint toward false-positive results, but this may also be influenced by variable effects of immunodeficiency on immune reactivity *in vivo* and *in vitro*.

It is unclear which test should be preferred for immunodiagnostic testing. When cross-sectionally comparing test results obtained by IGRAs and TST, IGRAs generally had a higher rate of positivity and were more strongly associated with *M. tuberculosis* exposure. However, results from the longitudinal part of this study may suggest superiority of the TST for risk assessment in HIV-infected patients. However, given the considerably high percentage of positive tests and the low number of tuberculosis cases on follow-up, there was no striking difference between

the tests and neither IGRA nor TST was adequately able to predict those at risk. In addition, although progression rates were low in patients with negative test results, physicians caring for immunocompromised patients must be aware that a negative result with any of the currently available tests does not rule out the future risk of developing tuberculosis.

The results from this study should have implications for refining future screening policies in immunocompromised patients in countries with low tuberculosis incidence. The low progression rates among test-positive individuals emphasize the limitations of current recommendations (7, 42) to screen all patients with chronic renal failure or with rheumatoid arthritis for LTBI in the absence of additional risk factors for tuberculosis. Because at least 50% of HIV-infected patients who developed tuberculosis had negative test results at the time of screening, both the positive and negative predictive values of any of the immunodiagnostic tests were very poor in low-incidence countries. Because development of tuberculosis was almost exclusively observed in patients with ongoing HIV replication, the data are consistent with lower tuberculosis rates associated with effective antiretroviral therapy, and it may be suggested that HIV-infected patients with positive immunodiagnostic test results do not have an increased risk of developing tuberculosis when HIV-replication is suppressed to undetectable levels by antiretroviral therapy. This emphasizes that the effect of antiretroviral therapy needs to be carefully studied and addressed when future policies for tuberculosis prevention are made for HIV-infected patients in countries of low tuberculosis prevalence.

The study has some limitations, of which most are inherent to the nonrandomized observational study design. The low number of cases with tuberculosis may be confounded by the fact that patients with the highest presumed risk for the development of tuberculosis were potentially more likely to have received preventive chemotherapy. Although formal randomization of receiving preventive therapy is ethically questionable, techniques to adjust for this treatment-by-indication bias, such as inverse probability weighting (43), could not be reliably used given the small proportion of patients receiving preventive therapy and the limited information of confounding variables. In addition, as in the setting of contact tracing, comparisons of progression rates toward tuberculosis among different groups of immunocompromised patients would be least biased if patients were harmonized for presumptive exposure, and if exogenous reinfection rates were either absent or identical. However, in this observational study, the time of exposure is largely unknown, which is caused by the fact that risk assessment in immunocompromised patients in a clinical routine setting is not primarily guided by recent contact. One of Comstock's studies in military recruits showed a high predictive value of a positive TST in the first year, whereas rates were increasing over 5 years of follow-up in those with baseline negative tests (44). In our study, we did not perform serial testing or specifically collect information on reexposure after testing. Therefore, we could not exclude that infection with *M. tuberculosis* occurred after screening, although incidence rates for tuberculosis were similar in individuals with HIV infection at the different time points of follow-up. Finally, this study was not powered to identify regional differences in the role of different assays to assess development of tuberculosis.

In conclusion, the performance of TST and IGRAs differs among patients with various etiologies of immunodeficiency. Immunocompromised patients at risk for developing tuberculosis are poorly identified by both TST and IGRAs. Better methods and biomarkers are urgently needed to specifically target preventive chemotherapy in immunocompromised individuals who would otherwise develop tuberculosis in the future. ■

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References

1. Lozano R, Naghavi M, Foreman K, Lim S, Shibuya K, Aboyans V, Abraham J, Adair T, Aggarwal R, Ahn SY, *et al*. Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the Global Burden of Disease Study 2010. *Lancet* 2012;380:2095–2128.
2. Erkens CG, Kamphorst M, Abubakar I, Bothamley GH, Chemtob D, Haas W, Migliori GB, Rieder HL, Zellweger JP, Lange C. Tuberculosis contact investigation in low prevalence countries: a European consensus. *Eur Respir J* 2010;36:925–949.
3. Mack U, Migliori GB, Sester M, Rieder HL, Ehlers S, Goletti D, Bossink A, Magdorf K, Hölscher C, Kampmann B, *et al*.; C. Lange; TBNET. LTBI: latent tuberculosis infection or lasting immune responses to M. tuberculosis? A TBNET consensus statement. *Eur Respir J* 2009;33: 956–973.
4. Diel R, Goletti D, Ferrara G, Bothamley G, Cirillo D, Kampmann B, Lange C, Losi M, Markova R, Migliori GB, *et al*. Interferon-γ release assays for the diagnosis of latent *Mycobacterium tuberculosis* infection: a systematic review and meta-analysis. *Eur Respir J* 2011;37:88–99.
5. International Union Against Tuberculosis Committee on Prophylaxis. Efficacy of various durations of isoniazid preventive therapy for tuberculosis: five years of follow-up in the IUAT trial. *Bull World Health Organ* 1982;60:555–564.
6. Rangaka MX, Wilkinson KA, Glynn JR, Ling D, Menzies D, Mwansa-Kambafwile J, Fielding K, Wilkinson RJ, Pai M. Predictive value of

interferon-γ release assays for incident active tuberculosis: a systematic review and meta-analysis. *Lancet Infect Dis* 2012;12:45–55.
7. Mazurek GH, Jereb J, Vernon A, LoBue P, Goldberg S, Castro K; IGRA Expert Committee; Centers for Disease Control and Prevention (CDC). Updated guidelines for using interferon gamma release assays to detect *Mycobacterium tuberculosis* infection—United States, 2010. *MMWR Recomm Rep* 2010;59:1–25.
8. Sester M, Bumbacea D, Duarte R, Lange C. Tuberculosis in the immunocompromised host. *Eur Respir Monogr* 2012;58:230–241.
9. Horsburgh CR Jr, Rubin EJ. Clinical practice. Latent tuberculosis infection in the United States. *N Engl J Med* 2011;364:1441–1448.
10. Luetkemeyer AF, Charlebois ED, Flores LL, Bangsberg DR, Deeks SG, Martin JN, Havlir DV. Comparison of an interferon-gamma release assay with tuberculin skin testing in HIV-infected individuals. *Am J Respir Crit Care Med* 2007;175:737–742.
11. Leidl L, Mayanja-Kizza H, Sotgiu G, Baseke J, Ernst M, Hirsch C, Goletti D, Toossi Z, Lange C. Relationship of immunodiagnostic assays for tuberculosis and numbers of circulating CD4+ T-cells in HIV infection. *Eur Respir J* 2010;35:619–626.
12. Triverio PA, Bridevaux PO, Roux-Lombard P, Niksic L, Rochat T, Martin PY, Saudan P, Janssens JP. Interferon-gamma release assays versus tuberculin skin testing for detection of latent tuberculosis in chronic haemodialysis patients. *Nephrol Dial Transplant* 2009;24:1952–1956.
13. Hadaya K, Bridevaux PO, Roux-Lombard P, Delort A, Saudan P, Martin PY, Janssens JP. Contribution of interferon-γ release assays (IGRAs) to the diagnosis of latent tuberculosis infection after renal transplantation. *Transplantation* 2013;95:1485–1490.
14. Bartalesi F, Vicidomini S, Goletti D, Fiorelli C, Fiori G, Melchiorre D, Tortoli E, Mantella A, Benucci M, Girardi E, *et al*. QuantiFERON-TB Gold and the TST are both useful for latent tuberculosis infection screening in autoimmune diseases. *Eur Respir J* 2009;33:586–593.
15. Kim SH, Lee SO, Park JB, Park IA, Park SJ, Yun SC, Jung JH, Kim YH, Kim SC, Choi SH, *et al*. A prospective longitudinal study evaluating the usefulness of a T-cell-based assay for latent tuberculosis infection in kidney transplant recipients. *Am J Transplant* 2011;11:1927–1935.
16. Manuel O, Humar A, Preiksaitis J, Doucette K, Shokoples S, Peleg AY, Cobos I, Kumar D. Comparison of quantiferon-TB gold with tuberculin skin test for detecting latent tuberculosis infection prior to liver transplantation. *Am J Transplant* 2007;7:2797–2801.
17. Aichelburg MC, Rieger A, Breitenecker F, Pfistershammer K, Tittes J, Eltz S, Aichelburg AC, Stingl G, Makristathis A, Kohrgruber N. Detection and prediction of active tuberculosis disease by a whole-blood interferon-gamma release assay in HIV-1-infected individuals. *Clin Infect Dis* 2009;48:954–962.
18. Brock I, Ruhwald M, Lundgren B, Westh H, Mathiesen LR, Ravn P. Latent tuberculosis in HIV positive, diagnosed by the *M. tuberculosis* specific interferon-gamma test. *Respir Res* 2006;7:56.
19. Kim YJ, Kim SI, Kim YR, Wie SH, Park YJ, Kang MW. Predictive value of interferon-γ ELISPOT assay in HIV 1-infected patients in an intermediate tuberculosis-endemic area. *AIDS Res Hum Retroviruses* 2012;28:1038–1043.
20. Mantoux C. L'intradermo-reaction de la tuberculine et son interpretation clinique. *Presse Med* 1910;2:10–13.
21. Centers for Disease Control (CDC). Revision of the CDC surveillance case definition for acquired immunodeficiency syndrome. Council of State and Territorial Epidemiologists; AIDS Program, Center for Infectious Diseases. *MMWR Morb Mortal Wkly Rep* 1987;36:1S–15S.
22. Wells G, Becker JC, Teng J, Dougados M, Schiff M, Smolen J, Aletaha D, van Riel PL. Validation of the 28-joint Disease Activity Score (DAS28) and European League Against Rheumatism response criteria based on C-reactive protein against disease progression in patients with rheumatoid arthritis, and comparison with the DAS28 based on erythrocyte sedimentation rate. *Ann Rheum Dis* 2009;68:954–960.
23. Cochran WG. The comparison of percentages in matched samples. *Biometrika* 1950;37:256–266.
24. Landis JR, Koch GG. The measurement of observer agreement for categorical data. *Biometrics* 1977;33:159–174.
25. Elzi L, Schlegel M, Weber R, Hirschel B, Cavassini M, Schmid P, Bernasconi E, Rickenbach M, Furrer H; Swiss HIV Cohort Study. Reducing tuberculosis incidence by tuberculin skin testing, preventive treatment, and antiretroviral therapy in an area of low tuberculosis transmission. *Clin Infect Dis* 2007;44:94–102.
26. Aabye MG, Ravn P, PrayGod G, Jeremiah K, Mugomela A, Jepsen M, Faurholt D, Range N, Friis H, Changalucha J, *et al*. The impact of HIV infection and CD4 cell count on the performance of an interferon gamma release assay in patients with pulmonary tuberculosis. *PLoS ONE* 2009;4:e4220.
27. Sester U, Wilkens H, van Bentum K, Singh M, Sybrecht GW, Schäfers HJ, Sester M. Impaired detection of *Mycobacterium tuberculosis* immunity in patients using high levels of immunosuppressive drugs. *Eur Respir J* 2009;34:702–710.
28. Bumbacea D, Arend SM, Eyuboglu F, Fishman JA, Goletti D, Ison MG, Jones CE, Kampmann B, Kotton CN, Lange C, *et al*. The risk of tuberculosis in transplant candidates and recipients: a TBNET consensus statement. *Eur Respir J* 2012;40:990–1013.
29. Ponce de Leon D, Acevedo-Vasquez E, Alvizuri S, Gutierrez C, Cucho M, Alfaro J, Perich R, Sanchez-Torres A, Pastor C, Sanchez-Schwartz C, *et al*. Comparison of an interferon-gamma assay with tuberculin skin testing for detection of tuberculosis (TB) infection in patients with rheumatoid arthritis in a TB-endemic population. *J Rheumatol* 2008;35:776–781.
30. Chen DY, Shen GH, Chen YM, Chen HH, Hsieh CW, Lan JL. Biphasic emergence of active tuberculosis in rheumatoid arthritis patients receiving TNFα inhibitors: the utility of IFNγ assay. *Ann Rheum Dis* 2012;71:231–237.
31. Inanc N, Aydin SZ, Karakurt S, Atagunduz P, Yavuz S, Direskeneli H. Agreement between Quantiferon-TB gold test and tuberculin skin test in the identification of latent tuberculosis infection in patients with rheumatoid arthritis and ankylosing spondylitis. *J Rheumatol* 2009;36:2675–2681.
32. Greenberg JD, Reddy SM, Schloss SG, Kurucz OS, Bartlett SJ, Abramson SB, Bingham CO III. Comparison of an in vitro tuberculosis interferon-gamma assay with delayed-type hypersensitivity testing for detection of latent *Mycobacterium tuberculosis*: a pilot study in rheumatoid arthritis. *J Rheumatol* 2008;35:770–775.
33. Passalent L, Khan K, Richardson R, Wang J, Dedier H, Gardam M. 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 Soc Nephrol* 2007;2:68–73.
34. Winthrop KL, Nyendak M, Calvet H, Oh P, Lo M, Swarbrick G, Johnson C, Lewinsohn DA, Lewinsohn DM, Mazurek GH. Interferon-gamma release assays for diagnosing *Mycobacterium tuberculosis* infection in renal dialysis patients. *Clin J Am Soc Nephrol* 2008;3:1357–1363.
35. Diel R, Loddenkemper R, Nienhaus A. Predictive value of interferon-gamma release assays and tuberculin skin testing for predicting progression from latent TB infection to disease state: a meta-analysis. *Chest* 2012;142:63–75.
36. Chee CB, Sester M, Zhang W, Lange C. Diagnosis and treatment of latent infection with *Mycobacterium tuberculosis*. *Respirology* 2013;18:205–216.
37. Soborg C, Ruhwald M, Andersen PH, Ravn P. 6-year follow-up of 522 HIV-positive individuals screened for *Mycobacterium tuberculosis* infection in Denmark. *Eur Respir J* 2014;44:540–543.
38. Jung YJ, Lyu J, Yoo B, Lee CK, Kim YG, Yang SK, Byeon JS, Kim KJ, Ye BD, Lee KH, *et al*. Combined use of a TST and the T-SPOT.TB assay for latent tuberculosis infection diagnosis before anti-TNF-α treatment. *Int J Tuberc Lung Dis* 2012;16:1300–1306.
39. Lawn SD, Badri M, Wood R. Tuberculosis among HIV-infected patients receiving HAART: long term incidence and risk factors in a South African cohort. *AIDS* 2005;19:2109–2116.
40. Karo B, Haas W, Kollan C, Gunsenheimer-Bartmeyer B, Hamouda O, Fiebig L; German ClinSurv HIV Study Group. Tuberculosis among people living with HIV/AIDS in the German ClinSurv HIV Cohort: long-term incidence and risk factors. *BMC Infect Dis* 2014;14:148.
41. Mancuso JD, Mazurek GH, Tribble D, Olsen C, Aronson NE, Geiter L, Goodwin D, Keep LW. Discordance among commercially available diagnostics for latent tuberculosis infection. *Am J Respir Crit Care Med* 2012;185:427–434.
42. European Centre for Disease Prevention and Control. Use of interferon-gamma release assays in support of TB diagnosis. Stockholm: ECDC; 2011.
43. Hermans SM, Manabe YC, Kiragga AN, Hoepelman AI, Lange JM, van Leth F. Risk of tuberculosis after antiretroviral treatment initiation: a comparison between efavirenz and nevirapine using inverse probability weighting. *Antivir Ther* 2013;18:615–622.
44. Comstock GW, Edwards LB, Livesay VT. Tuberculosis morbidity in the U.S. Navy: its distribution and decline. *Am Rev Respir Dis* 1974;110: 572–580.