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Contribution of a IFN-γ assay in contact tracing for tuberculosis in a low-incidence, high immigration area

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Summary

Aims of study: to analyse, in contacts exposed to smear+/culture + (S+/C+) or S-/C+ TB, most of whom are foreign-born: 1) correlation between T-SPOT.*TB* IFN-γ release assay (Oxford Immunotec, UK), TST and exposure scores; 2) agreement between T-SPOT.*TB* and TST in *Bacillus of Calmette-Guérin* (BCG) vaccinated and non-vaccinated subjects, and 3) impact of results of T-SPOT.*TB* on diagnosis and treatment for latent tuberculosis infection (LTBI).

Patients and methods: TST and T-SPOT.*TB* were performed in 295 contacts (74% foreign-born) 8–12 weeks after exposure. Contacts completed five exposure scores. Data were analysed according to most recent US (ATS/CDC), British (NICE) and Swiss guidelines.

Results: T-SPOT.*TB* was positive in 115 (39%) and indeterminate in 15 subjects (5.1%). Neither TST, nor T-SPOT.*TB* was significantly related to exposure scores or infectiousness of the index case. In multivariate analysis, incidence of TB in country of origin was the strongest predictor of result of TST or T-SPOT.*TB*. Agreement between TST and T-SPOT.*TB* (kappa: 0.19–0.27) was low but improved in non BCG-vaccinated subjects (kappa: 0.28–0.47). According to guidelines referred to, 10–24% of subjects screened were T-SPOT.*TB*+/TST–: the prognosis of this group is yet undetermined. Another 10–27% were T-SPOT.*TB*–/TST+: present guidelines recommend withholding treatment for LTBI in these subjects although longitudinal data are still scarce.

Conclusions: the lack of a relationship between T-SPOT.*TB* and exposure scores probably results from both the variability inherent to the design of this study (ie, multiple contact investigations, exposure in different settings) and limits in the performance of the IGRA tested. Longitudinal data are needed to clarify the risk of TB in T-SPOT.*TB*+/TST- individuals. Unreliability of diagnosis of LTBI in spite of the present use of IGRA in algorithms is illustrated by the wide variations in identification of LTBI according to different guidelines referred to.

Key words: latent tuberculosis infection; tuberculosis; interferon-γ release assays

This study was registered at www.clinicaltrials.gov: Title: Use of a Gamma-IFN assay in Contact Tracing for Tuberculosis in a Low-Incidence, High Immigration Area. Registration No.: NCT00557765.

This study was approved by the Ethics committee on clinical research of Geneva University Hospital (President: Professor O. Irion; Hôpital Cantonal Universitaire; 25, rue Micheli-du-Crest; 1211 Geneva 14. Switzerland).

Introduction

In Switzerland, as in most industrialised countries, there has been over the past 20 years a regular decline in the incidence of tuberculosis (TB) among native subjects, and a simultaneous increase in the proportion of foreign-born migrants among TB notifications. In fact, the number of TB cases in foreign-born subjects (64% of all TB cases in 2004) exceeds that of the Swiss-born since 1994 [1]. In the Geneva area where this study was performed, 84% of TB cases are foreign-born. Thus a high proportion of immigrants is to be expected among close contacts of TB cases, with a high rate of prior BCG vaccination, and also of previously acquired latent tuberculosis infection (LTBI).

Control of TB in industrialised countries is based on rapid detection of active TB, contact screening for detection of LTBI after exposure to TB, and screening of high risk groups. Detection of LTBI has relied until recently on the tuberculin skin test (TST). Although there is a substantial body of evidence linking results of the TST to subsequent risk of TB [2], TST lacks specificity [3], leading to potentially unnecessary treatments for LTBI. Over the past five years, two *in vitro* tests (IFN-γ release assays: IGRA) have become com-

mercially available in several European countries [4]. Their major contribution is that they do not yield false positive results in subjects with previous BCG vaccination, or infection by most environmental mycobacteria [5]. In patients with TB, their sensitivity equals or surpasses that of the TST [6]. However, because there is no gold standard for diagnosing LTBI, studies supporting the use of IGRA for detecting LTBI rely on circumstantial evidence, ie, studies showing that results of IGRA are better correlated with scores of exposure than TST [7–12]. Most of these studies describe situations occurring in an environment with a low probability of prior exposure to TB, in which intensity of exposure to index cases could be clearly quantified [7–10, 12]. Based on these data, IGRA have recently been introduced in the algorithms of several national guidelines for contact tracing procedures. ATS/CDC guidelines (2000) [3] recommended a 5 mm threshold value for TST; more recently, CDC recommendations stated that IGRA can replace the TST for the diagnosis of LTBI [13]. The 2006 British National Institute for Health and Clinical Excellence (NICE) guidelines [14] and the 2007 Swiss National Guidelines [15] both recommend, for immuno-competent adults, the "two-step procedure" (ie, confirming positive TST results by one of the IGRA assays). NICE guidelines recommend a 15 mm cut-off for TST in BCG-vaccinated individuals *vs* 5 mm in non-vaccinated subjects. Swiss guidelines recommend a 10 mm cut-off value for TST irrespective of BCG vaccination status [15].

This prospective study aimed to analyse the contribution of the T-SPOT.*TB* IGRA in routine contact-tracing, in a low-incidence area for TB, but with an important immigrant population (45% in 2008), and a very high proportion of foreign-born subjects among TB cases (84%), highlighting recent epidemiological trends of TB in Western Europe. More specifically, we wished to quantify: 1) the correlation between T-SPOT.*TB*, TST and exposure scores in this setting; 2) the agreement between T-SPOT.*TB* and TST in BCG vaccinated and non-vaccinated subjects, and 3) the impact of results of T-SPOT.*TB* on diagnosis and treatment for LTBI, according to Swiss, British and US guidelines.

Patients and methods

The Division of Pulmonary diseases (Geneva University Hospital) supervises all contact tracing procedures in the Geneva area (440,000 inhabitants, TB incidence of $20*10^{-5}$, of which 84% are foreign-born). Between October 1, 2004 and February 1, 2006, all subjects screened after contact with a case of culture confirmed pulmonary TB were prospectively invited to participate to the present study. Subjects with known prior TB were excluded.

The study protocol was accepted by the Ethics committee on clinical research of Geneva University Hospital. All subjects included provided written informed consent.

Eight to 12 weeks after last exposure to index case, contacts were interviewed by a research nurse, who recorded: age, social status, country of birth, history of prior TST testing, TB infection and BCG vaccination, presence of scars suggesting BCG, history of HIV infection or other cause of immuno-suppression, infectiousness of index case (smear + *vs* others) and 5 scores of exposure derived from previous publications (see Appendix 1: Scores 1–5) [10, 12, 16–18]. Scores focused either on environment in which contact occurred, duration of contact, or relationship between index case and contact.

A tuberculin skin test (TST) was performed, according to the Mantoux technique, using 2 units of purified protein derivative (PPD) RT 23 from Statens Serum Institute, Copenhagen, Denmark (bioequivalent to 5 units of the US PPD standard) and read after 72 hours by two experienced nurses (readings were averaged).

Blood sampling for determination of *M tuberculosis* specific IFN-γ secreting T-cells (T-SPOT.*TB*, Oxford Immunotec, Oxford, UK) and TST were performed simultaneously. Peripheral venous blood samples (8 ml) were processed by our laboratory within three hours. Peripheral blood mononuclear cells were separated by centrifugation, counted, re-suspended in a serum-free culture medium (AIM-V™, Gibco Invitrogen, Basel, Switzerland), and dispensed in wells pre-coated with an anti-IFN-γ antibody (2.5×10^5 cells per well). Plates were incubated overnight at 37 °C with 5% CO_2 in presence of medium alone (negative control), phyto-haemagglutinine (positive control), ESAT-6 or CFP-10 antigens. After incubation, wells were washed and developed with a conjugate against the antibody used and an enzyme substrate. Spot-forming units (SFUs) were counted with an automated ELISPOT reader (AID system, Strasberg, Germany). According to the manufacturer's recommendations, tests were considered as indeterminate: 1) if SFUs in the positive control were <20, or 2) if SFUs in the negative well exceeded 10 and both antigen wells had less than twice the number of SFUs of the negative well. Results were scored as positive if SFU count in either antigen well was >6 spots above SFUs of negative control. SFU values reported are SFUs of highest of ESAT-6 or CFP-10 wells – SFUs of negative control.

According to national and international guidelines, all subjects with a TST >5 mm and/or a positive T-SPOT.*TB* test were referred for clinical examination, and chest roentgenogram [3, 15]. When indicated, treatment for latent tuberculosis infection was initiated, according to Swiss guidelines [15].

Statistical analysis

Data are reported as mean ± standard deviation (SD) unless specified otherwise. Continuous variables were compared between groups by unpaired t-tests, and categorial variables, by chi-square test.

All statistical analyses took into account TST thresholds as specified in ATS/CDC (>5 mm), British NICE (>15 mm in BCG vaccinated subjects, if not: 5 mm) and Swiss (>10 mm) national guidelines.

Univariate logistic regression was used to test for association between results of T.SPOT.*TB* or TST (at different threshold levels) and either exposure scores (scores

1–5, see appendix 1), or infectiousness of index case (smear + *vs* others). Logistic regression was used to identify variables associated with either TST or T-SPOT.*TB* results (dependent variables) among the following covariates: age, gender, being Swiss or foreign-born, incidence of TB in country of origin, history of living or prolonged travel abroad, history of BCG vaccination, exposure scores, and infectiousness of index case. Models were selected using a backward procedure, guided by the analyst. To facilitate comparisons between models, we retained in our final analysis all variables which appeared in at least one of the empirically derived models. Results of multivariate logistic regression are expressed as odds ratio with 95% confidence interval (95%CI).

To measure agreement between T.SPOT.*TB* and TST, we reported the percentage of subjects in which results of TST and T.SPOT.*TB* agreed, and kappa coefficients for all subjects, and for subjects without BCG vaccination. Kappa values between 0.2 and 0.4 represent fair agreement between two tests; values between 0.4–0.6, moderate agreement, values above 0.6: substantial agreement [19].

For all tests, level of significance was set at $p < .05$.

Results

During the study period, among 319 subjects screened, 24 declined inclusion. Details of the study population (n = 295) and index cases as well as results of TST and T.SPOT.*TB* are listed in table 1. Most contacts (74%) and index cases (87%) were foreign-born. Incidence of TB in country of origin (cases per 10^5 inhabitants) was <50 in 201 subjects (68.1%), 50–99 per 10^5 inhabitants in 55 (18.6%) and >100 in 39 (13.2%). Among all subjects, 20 (6%) had at least one factor of immune suppression (diabetes: 8; cancer: 5; immunosuppressive treatment: 4; chronic renal failure: 1). No subject reported HIV infection.

Fifteen T.SPOT.*TB* tests (5.1%) were indeterminate, only one of which had a factor of immune suppression (diabetes). Neither age nor gender differed between subjects with indeterminate tests and other contacts.

No case of active TB was detected, either at initial screening, or after a follow-up of 2.1 ± 0.4 years (range: 1.1–2.7).

Agreement between results of TST or T-SPOT.*TB* and scores of exposure or infectiousness

We found no significant relationship between results of either TST, or T-SPOT.*TB* and infectiousness of index case (univariate logistic regression). Neither was there any significant relationship between exposure scores and results of TST, or T-SPOT.*TB*. Multiple logistic regression, with either TST or T-SPOT.*TB* as dependent variables, identified age, gender, history of BCG, and incidence of TB in country of origin (stratified as low: $<50*10^{-5}$; intermediate: $50–99*10^{-5}$; high: $>100*10^{-5}$ cases/year) as significant covariates (tab. 2). Exposure scores were not retained as significant covariates in regression models. Not surprisingly, having a TST >5 mm (but not >10 mm) was significantly associated with history of BCG vaccination. Analysing results of either antigen alone (ESAT-6 or CFP-10) as dependant variable did not improve relationship with exposure scores (results not shown).

Table 1
Study population, and index cases.

Number of contacts (% Male, % Female)	295 (52% M, 48% F)
Age (Mean ± SD, range, years)	40 ± 13 (16–83 years)
Foreign-born	218 (73.9%)
History of BCG or suggestive scar	238 (80.6%)
Index cases (% Male, % Female)	73 (46% M, 54% F)
Age (Mean ± SD, years)	45 ± 20
Foreign-born	63 (87%)
Number of contacts per index case	4.2
Contact exposed to:	
Cavitary TB, Smear +, Culture +, n (%)	105 (35.6%)
Non-cavitary TB, Smear +, Culture +, n (%)	168 (56.9%)
Pulmonary TB, Smear –, Culture +, n (%)	22 (7.5%)
Tuberculin skin test (TST) induration	
>5 mm, n (%)	173 (58.6%)
>10 mm, n (%)	148 (50.2%)
>15 mm, n (%)	61 (20.7%)
TST + according to NICE guidelines	74 (25.1%)
T.SPOT.*TB* positive	115 (39%)

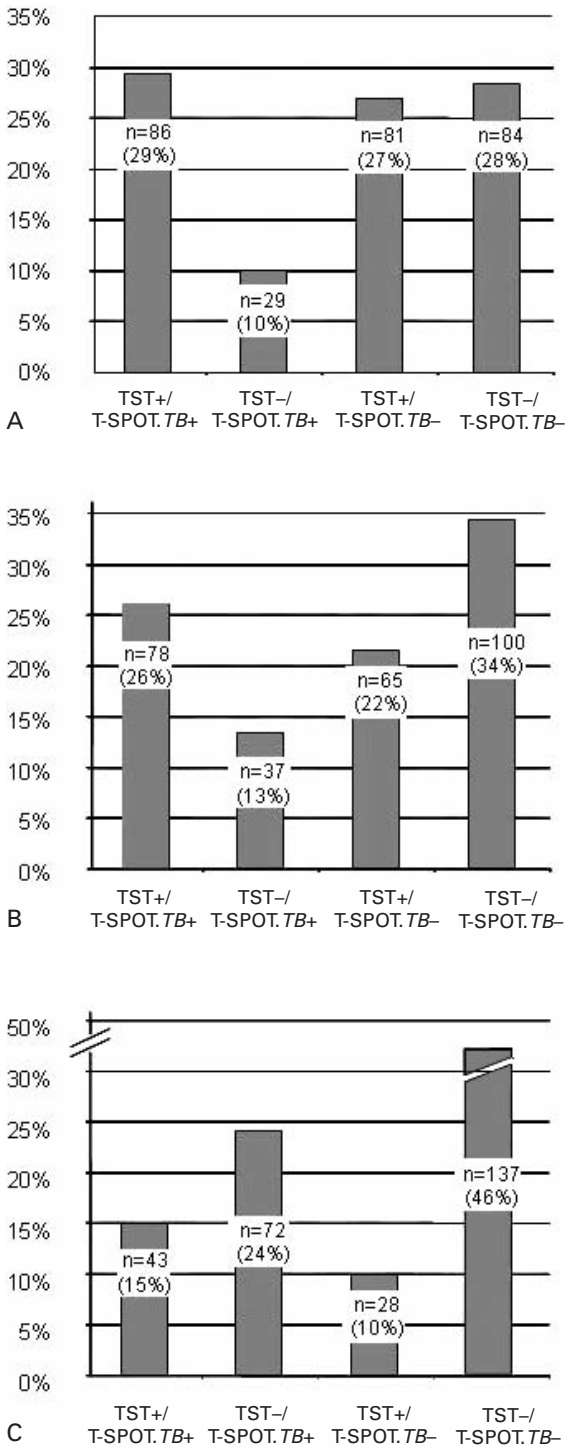

Figure 1

Agreement between TST and T-SPOT.*TB*.
A (top): results for a TST threshold of 5 mm (2000 ATS/CDC guidelines);
B (middle): results for a TST threshold of 10 mm (Swiss 2007 guidelines);
C (bottom): results according to British 2006 NICE guidelines (see text for details). Percentages refer to total population (n = 295), of which 15 had indeterminate results for T-SPOT.*TB*.

To test for effect of BCG, univariate logistic regressions were performed on unvaccinated subjects only (n = 51, results not shown): neither exposures scores, nor infectiousness of index case were significantly related to TST or T-SPOT.*TB*.

Agreement between TST and T-SPOT.*TB*

According to guidelines and TST thresholds referred to, T-SPOT.*TB* agreed with TST in 60.7–64.2% of results (fig. 1); 90% of discordant results occurred in BCG-vaccinated subjects. Kappa values are given in table 3. When excluding BCG-vaccinated contacts, κ values increased substantially. Conversely, changing the cut-off value for the T-SPOT.*TB* (SFUs of highest of ESAT-6 or CFP-10 wells – SFUs of negative control) to 4, 8 or 10 SFUs did not significantly improve κ values (data not shown). Indeterminate results were excluded from analysis when testing for agreement, which slightly improves percentages and kappa values reported in table 3.

Impact of T-SPOT.*TB* on indication for treating LTBI:

According to guidelines referred to, screening identified 10–24% T-SPOT.*TB*+/TST– individuals and 10–27% T-SPOT.*TB*–/TST+ subjects (fig. 1). Based on NICE 2006 guidelines, 43 (15%) of all subjects screened would receive treating for LTBI. By contrast, most recent CDC guidelines would lead to treating all T-SPOT.*TB* positive individuals (39% of all subjects screened) [20].

Distribution of results of T-SPOT.*TB* and TST

Distribution of TST results was bi-modal (not shown), as expected in contact investigations after exposure to TB [21, 22]. Conversely, distribution of T-SPOT.*TB* results (SFUs) was skewed, uni-modal, with no obvious threshold for discriminating between subjects with *vs* without LTBI (fig. 2). Average SFU count of negative controls was 1.3 ± 1.7 (upper 95%CI: 4.7). The cut-off suggested by the manufacturer (>6 SFU) was

Table 2

Results of multiple logistic regression with age (by 10 year increments), gender, BCG vaccination, and incidence of TB in country of origin (category scale: OR = 1 for subjects originating from a country with an incidence <50/10^5) as covariates, and, as dependent variables: TST at different thresholds, T.SPOT.*TB* and probable LTBI according to NICE British guidelines (see text for details). P values: *: <.05; **: <.01; #: <.005; ##: <.001. OR: Odds ratio; 95%CI: 95% confidence intervals.

Dependent variables	TST >5 mm		TST >10 mm		LTBI according to NICE guidelines		T-SPOT.*TB*	
Covariates	OR	95%CI	OR	95%CI	OR	95%CI	OR	95%CI
Age (by 10 year increments)	1.03	0.84–1.27	1.07	0.87–1.30	1.13	0.90–1.41	1.30*	1.06–1.60
Gender (Male *vs* Female)	2.07**	1.22–3.51	1.52	0.92–2.51	1.50	0.84–2.67	1.15	0.69–1.92
BCG vaccination	2.98#	1.39–6.41	1.91	0.90–4.05	0.42*	0.19–0.93	0.94	0.44–2.00
Incidence in country of origin								
50–99/10^5 *vs* <50/10^5	2.58#	1.26–5.27	2.22*	1.15–4.27	2.03	0.99–4.14	2.17*	1.13–415
>100/10^5 *vs* <50/10^5	3.67#	1.40–9.60	3.84#	1.61–9.20	3.82##	1.68–8.69	2.62*	1.18–5.82

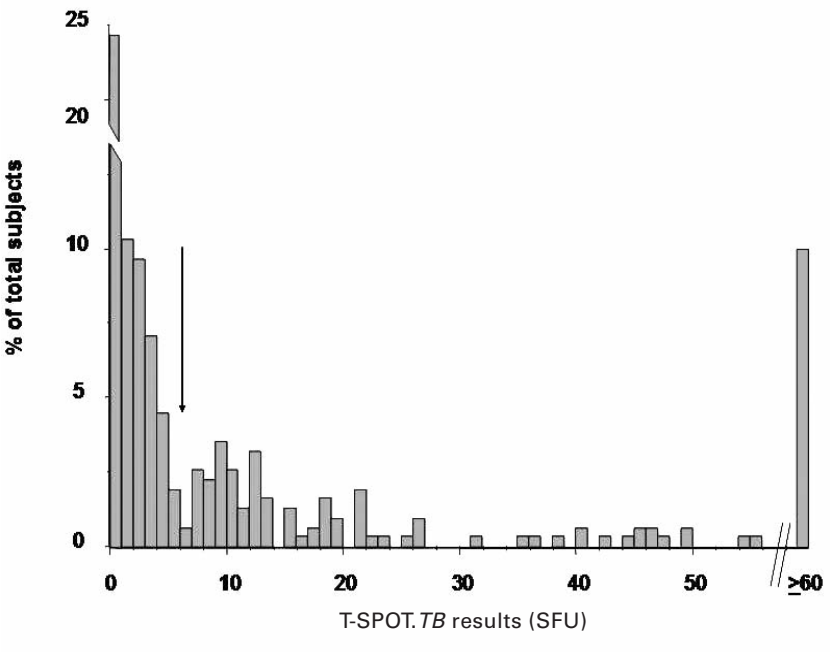

Figure 2

Histogram of distribution of results of T-SPOT.*TB* expressed as [Total SFUs – SFUs of negative control]. SFU: Spot forming units. Vertical arrow: threshold value specified by manufacturer (Oxford Immunotec, UK). X axis truncated to increase readability.

Table 3

Agreement between results of T.SPOT.*TB* and TST at different threshold values of TST in all contacts studied (n = 280, indeterminate results deleted), and in subjects without BCG vaccination (n = 51), expressed as percentage of concordant results and kappa coefficients. Kappa values between 0.2 and 0.4 represent fair agreement between 2 tests; values between 0.4–0.6, moderate agreement, values above 0.6: substantial agreement. *: 15 cases had indeterminate T-SPOT.*TB* results. **: For NICE guidelines, see text.

All subjects (n = 295)*	Concordant results: n (%)	Kappa value	95% CI
TST >5 mm	170 (60.7%)	0.24	0.14–0.33
TST >10 mm	178 (63.6%)	0.27	0.16–0.38
TST >15 mm	179 (63.9%)	0.19	0.09–0.30
TST + according to NICE guidelines**	180 (64.2%)	0.22	0.10–0.33
BCG non-vaccinated subjects (n = 51)			
TST >5 mm	40 (78.4%)	0.47	0.20–0.74
TST >10 mm	39 (76.5%)	0.41	0.14–0.68
TST >15 mm	40 (78.4%)	0.28	0.03–0.54
TST + according to NICE guidelines	40 (78.4%)	0.47	0.20–0.74

thus 3.3 SD above average SFUs of negative controls in this population.

SFU counts according to TST results in T-SPOT.*TB* subjects

Fig. 3 shows results of T-SPOT.*TB* expressed as SFUs: T-SPOT.*TB*+ subjects are grouped according to result of TST (> *vs* ≤5 mm). Median SFUs (n, range, p value for comparison between subjects with TST >5 vs ≤5 mm, unpaired t test) were: for T-SPOT.*TB*+/TST+ subjects: 25 (n = 93; 7–239); for T-SPOT.*TB*+/TST– subjects: 10 (n = 31; 7–143; p = 0.003).

Discussion

This prospective study, involving subjects recently exposed to an identified case of pulmonary TB, performed in the routine of a TB clinic over 18 months, led to the following observations. First, in an urban setting with a high immigrant population, after correction for incidence of TB in country of origin, age, gender and BCG vaccination, neither TST nor T-SPOT.*TB* were correlated with scores of exposure. Secondly, agreement between TST and T-SPOT.*TB* was low, and remained low when modifying threshold values for TST. Although agreement increased substantially when excluding BCG vaccinated individuals, there was an important residual discordance

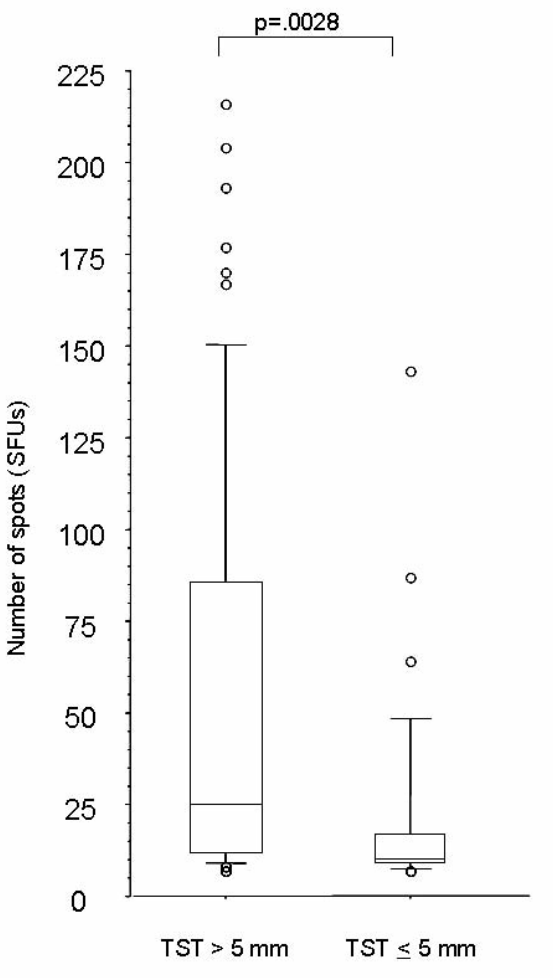

Figure 3

Box plots of results of T-SPOT.*TB* (SFUs) in T-SPOT.*TB*+ subjects according to TST results (the box showing median value, 25th and 75th percentiles, and the bars showing 5th and 95th percentiles). Level of significance (p value) shown for unpaired t-test comparing [T-SPOT.*TB*+/TST+] subjects with [T-SPOT.*TB*+/TST–] subjects.

between tests which remains unexplained. Thirdly, the simultaneous use of TST and T-SPOT.*TB* defined two sub-groups of patients: a group of TST+/T-SPOT.*TB*– individuals for whom present guidelines recommend withholding treatment for LTBI, and a group of TST–/T-SPOT.*TB*+ subjects for whom recommendations have yet to be harmonised. Noteworthy is the substantial variability of these groups according to guidelines referred to.

TST, T-SPOT.*TB* and exposure scores:

The absence of a significant relationship between TST or T-SPOT.*TB* and exposure scores, once corrected for incidence of TB in country of origin and age, was unexpected. Several hypotheses must be considered. The first is to question the validity of the exposure scores. However, all scores chosen had previously been shown to correlate with IGRA tests in different settings [10, 12, 16, 18]. Furthermore, scores included all items which are considered critical for risk of exposure: infectiousness of index case, relationship to index case, time and environment of exposure [11, 22]. The second hypothesis is that neither the TST nor the T-SPOT.*TB* could identify recent exposure to TB in this population, because of a very high "background noise" related to LTBI acquired earlier in life. Indeed, contacts screened were mostly foreign-born (74%: tab. 1). This hypothesis is supported by the multiple logistic regression analysis (tab. 2). However, restricting the analysis to subjects originating from low incidence countries only did not improve the relationship between TST or T-SPOT.*TB* and exposure scores (data not shown). The third explanation is that most previous studies had analysed contacts exposed to a single index case, thus eliminating a large proportion of the variability inherent to the present study (ie, infectiousness of index cases over time, effective exposure). The absence of any significant relationship between TST or T-SPOT.*TB* and exposure scores may thus result from the combination of the variability related to pooling data from multiple contact tracing procedures and less than optimal performances of both TST and T-SPOT.*TB*.

Agreement between TST and T-SPOT.*TB*:

Agreement between T-SPOT.*TB* and TST in our study was (tab. 3) within previously reported values (48–75%; κ: 0.16–0.51), and increased substantially when excluding BCG-vaccinated contacts (tab. 3) [5, 11, 12, 23–25]. Similar results were reported by Diel et al. and Ferrara et al. [5, 23]. Even in non-vaccinated individuals however, there remains – in this study and in previous reports – a high rate of discordant results (20–39%) for which there is no clear explanation [11, 12, 25]. In this study, immuno-suppression could not explain these observations, and infection by environmental mycobacteria is very unlikely in our area. Discordance may have resulted from suboptimal cut-off levels for either test. Indeed, small changes in cut-off values for IGRA have been shown to decrease level of discordance between TST and IGRA [25–27]. However, for the TST, choosing three different thresholds had little impact on levels of agreement (tab. 3). For the T-SPOT.*TB*, an optimal cut-off level was difficult to determine on the basis of the distribution of results, shown in figure 2. Different threshold values (4, 8, 10) did not improve *kappa* values. Discordant results therefore most probably reflect limitations of *both* tests in terms of sensitivity and specificity.

Practical implications of discordant TST and T-SPOT.*TB* results:
T-SPOT.*TB*–/TST+ subjects

As in previous studies, the T-SPOT.*TB* IGRA identified *probable* false positive results of the TST and thus avoided what present guidelines classify as unnecessary treatments for LTBI. This assumption is based on the high specificity of the IGRA tests (0.92, 95%CI: 0.88–0.95 for T-SPOT.*TB*) [6]. It must be stated, however, that, in the absence of any long-term follow-up data of T-SPOT.*TB*–/TST+ subjects, we do not presently know whether some of these subjects are in fact true positives, even if the likelihood of this state-

ment is low. Depending on guidelines referred to (fig. 1, A–C), 10–27% of subjects in this study had *probable* false positive TST results, the highest percentage being associated with ATS/CDC 2000 guidelines, which recommend the most sensitive cut-off level for the TST [3]. Previous reports have yielded presumptive false positive TST rates of 18–32% [11, 12].

T-SPOT.*TB*+/TST– subjects

This finding is more intriguing, and has been reported in 3.3–16% of subjects tested with the T-SPOT.*TB* [8, 9, 11, 12, 23, 28]. As shown in figure 1, the importance of this group varies widely according to guidelines referred to (10–24%). TST-based longitudinal studies would suggest that these subjects – if immuno-competent and young – are at low risk for developing TB [2]. Indeed, no treatment would be recommended for these subjects using the British or Swiss "two-step procedure". However, following the most recent CDC guidelines, in which IGRA tests can replace TST, these patients should receive treatment for LTBI.

TST-/T-SPOT.*TB*+ subjects have significantly lower SFU counts compared to TST+/T-SPOT.*TB*+ individuals, thus a lower IFN-γ production (fig. 3). IFN-γ levels have been correlated to activity of TB [29–32]. One hypothesis would be that these subjects have been infected several years earlier, and thus have a decreased IFN-γ response to *M. tuberculosis* specific antigens. An alternative explanation is that, in some contacts, the IGRA may be more sensitive than TST if no booster procedure is performed: repetition of the TST (which was not performed in this study) may decrease the observed difference between tests.

Subjects for whom treatment for LTBI would be recommended

Most recent CDC guidelines in which IGRA can replace TST, would lead to the highest rate of treatment for LTBI in this particular setting (all T-SPOT.*TB*+ individuals: 39%) [20]. Conversely, NICE guidelines would result in the lowest rate of treatment for LTBI (15%).

Study limitations

First, results of this study must be considered as clearly context-dependent, and cannot be extrapolated to low incidence areas with a lower immigrant population: the lack of a significant relationship between exposure scores and TST or the IGRA does not question the previously established strong correlations between IGRA results and gradients of exposure after recent infection [8–12]. Secondly, we did not compute a combined score related to contagiousness and circumstances of exposure as performed by Shams et al [11], in order to further explore a possible relationship between exposure scores and TST or the IGRA. Combining results of all scores as binary variables did not improve correlation between TST or the IGRA and exposure (data not shown). Thirdly, BCG vaccination status often relied on indirect evidence (suggestive scars): this may affect results of κ values in non-vaccinated subjects; results presented are however in close agreement with previous publications.

Conclusions

This study focused on contact tracing for LTBI in a population with a high proportion of immigrants, reflecting trends in the epidemiology of TB in several countries of Western Europe and thus differed from many previous studies of IGRA in contact tracing procedures [7, 8, 10, 12]. Variability inherent to the design of the study (ie, multiple contact investigations, exposure in different environments) and suboptimal performance of T-SPOT.*TB* probably explain the lack of significant relationship between IGRAs and exposure scores, which differs from previous publications. However, many questions regarding the diagnostic performance of the T-SPOT.*TB* remain unanswered. For instance, short and long term risk of TB in T-SPOT.*TB*+/TST– immuno-competent adults has yet to be established. Also, causes for the residual discordance between TST and T-SPOT.*TB* even in non vaccinated individuals warrant further investigation. Undoubtedly, IGRA are valuable contributions for the detection of LTBI, with improved diagnostic performances when compared to the TST. However, the striking differences in percentage of subjects for whom treatment for LTBI would be indicated according to guidelines referred too illustrates the remaining uncertainties as to identifying LTBI, in spite of having integrated IGRAs in the diagnostic strategies.

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Appendix 1

Scores used during interview of study subjects.

Score 1 (derived from: Lalvani A et al., 2001; [10]):

1. Category A contacts (close and prolonged exposure) had lived in the same household or shared their workplace office with their index case
2. Category B contacts (regular and intermittent exposure) had been in the same room as their index case at least once a week, for an estimated mean time longer than 1 hour/week, for at least 4 weeks before diagnosis or end of exposure
3. Category C contacts (casual intermittent exposure) had been in the same room as their index case at least once a week, for an estimated mean time of less than 1 hour/week, for at least 4 weeks before diagnosis or end of exposure
4. Category D contacts had worked or studied in the same institution as their index case, without any known contact with index case or other tuberculosis patient or had no significant contact with index case

Score 2 (derived from Bailey WC et al.; [16]):

Place of most significant exposure:

1. Home
2. Transportation means (bus, van, car, plane or train)
3. Leisure gathering spot (bar, poolhall)
4. Confined (work, group home, shelter, boarding house, jail, other institution)
5. No significant contact

Score 3 (derived from Bailey WC et al.; [16]) (*):

Size of environment in which contacts occurred with index case (from Bailey WC et al.; [16]):

1. Size of a vehicle or car
2. Size of a (bed)room
3. Size of a large room
4. Size of a house
5. Larger than a house

Score 4 (derived from Reichler et al. and Lankensjold et al. [18, 33]) (*):

Relationship to index case:

A Close family tie with index case (spouse, parent)
B Other relationship with index case (friend, acquaintance, neighbour, worker in household of index case)
C Professional or scholastic relationship (same workplace or school)

Score 5 (from Zellweger et al. [12]):

Total number of hours of exposure

1. <3 hours
2. 3–8 hours
3. >8 hours of total exposure

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