Detecting latent tuberculosis infection during anti-tumour necrosis factor therapy

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Abstract

Background

There is little information regarding the reliability of repeat tuberculin skin tests (TSTs) and interferon gamma release assays (IGRAs) in detecting latent tuberculosis infection (LTBI) in people on anti-tumour necrosis factor (TNF) medication.

Methods

We conducted a prospective, observational study of patients referred to the Saskatoon Tuberculosis (TB) Clinic prior to starting anti-TNF medication. A chest x-ray (CXR), 2-step TST and IGRA (QuantiFERON-TB Gold In-Tube Method) were performed at baseline. Those patients with a baseline TST ≥10 mm and/or a positive IGRA were followed with a clinic visit, CXR, TST and IGRA at 3 and 6 months after starting anti-TNF medication.

Results

Of 106 potential patients, 91 consented to participate. Twenty-six patients had a positive (≥10 mm) TST or IGRA at baseline; twelve started and stayed on anti-TNF medication through the 6-month follow-up and completed both planned follow-up visits. The baseline mean TST measurement for the 12 participants was 13.9 mm (SD 11.4), increasing to a mean of 16.8 mm (SD 9.3) post-booster. At 3 months post-anti-TNF initiation, there was an overall decrease in TST measurement (mean=10.0 mm; SD 9.3; p=0.013), with measurements <5 mm in 3 of the 12 patients. By the 6-month TST, a response recovery was observed with a mean TST measurement of 14.5 mm (SD 7.7), with 11/12 ≥5 mm. The IGRA was unchanged throughout the study period in all patients. The overall agreement between TST and IGRA was poor (kappa coefficient = 0.180, p=0.020).

Conclusion

We demonstrated a transient but significant decrease in TST response in the first six months of anti-TNF therapy.

Key words

anti-tumour necrosis factor, anti-TNF, TB, latent tuberculosis, interferon gamma assays, tuberculin skin test, Mantoux test, TST, Quantiferon® TB-Gold

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Received on November 26, 2010; accepted in revised form on April 18, 2011.

Funding: This research.was funded by the Royal University Hospital Foundation.

Competing interests: none declared.

Introduction

Latent tuberculosis infections (LTBI) have historically been diagnosed primarily through utilisation of the tuberculin skin test (TST) (1). Limitations of this testing method and research demonstrating the role of interferon-gamma in the immune response to *M. tuberculosis* infection have led to the recent development of interferon-gamma release assays (IGRAs). These assays rely on previously sensitised T-cells producing interferon-gamma when re-exposed to certain mycobacterial antigens (2). Since the first IGRA (QuantiFERON-TB) was approved for use, a third generation version of that assay (QuantiFERON-TB Gold In-Tube Method), has been released (2). Interpretive criteria for the IGRAs have not been uniform in the published literature, and calculation of accuracy of the IGRAs and the TST is challenging due to the lack of a reference standard for diagnosing LTBI (3). Poor agreement between the TST and IGRAs has been reported (4).

Anti-tumour necrosis factor (TNF) biological agents are widely employed in several inflammatory rheumatologic syndromes as well as other disorders such as inflammatory bowel disease (5-8). Anti-TNF agents have been associated with both increased progression to tuberculosis disease (TB) in TST positive patients and increased risk of acquiring a new TB infection (9). The overall risk has been reported to be higher with the monoclonal antibody anti-TNF agents (infliximab and adalimumab) compared to the soluble receptor anti-TNF agent (etanercept) (10, 11). Infliximab has also been associated with a shorter time interval from therapy initiation to TB onset as well as a higher risk of disseminated and extra-pulmonary TB when compared to etanercept (10, 12, 13).

There is limited documentation of the reliability of repeated TST and IGRAs for detection of LTBI in patients utilising anti-TNF agents (4, 14-16). The majority of previous studies investigating results of TST and IGRA before and during anti-TNF therapy did not employ a standard 2-step TST at baseline (4, 14-16). There has been little evaluation of the TST earlier than six months following anti-TNF therapy initiation. Recognising a propensity for LTBI reactivation early in the course of anti-TNF therapy (10), we conducted a prospective study to observe the effect of these biologic agents on TST and IGRA results during the first six months of anti-TNF therapy in patients with positive baseline results.

Methods

We conducted a prospective observational study of patients starting anti-TNF therapy. All patients referred to the Saskatoon TB clinic between May 2007 and August 2008 for evaluation prior to anti-TNF treatment were asked to participate. Written informed consent was obtained from all subjects; those patients unwilling or unable to consent were excluded. The protocol and consent form were approved by the Biomedical Ethics Committee at the University of Saskatchewan. All patients were assessed by a TB physician. Height and weight were recorded, and demographic information was collected including age, date of birth, birthplace, ethnicity (self-reported), and occupation. A medical history was obtained including indication for anti-TNF medication, duration of illness, current and recent medications, and TB risk factors. All patients received a chest x-ray and a two-step TST (using the Mantoux technique as outlined in the Canadian Tuberculosis Standards 6th ed.) (1), and blood was drawn for the QuantiFERON®-TB Gold In-Tube (QFT-GIT) assay (Cellestis Inc., Valencia, USA). The TSTs were performed by staff at the Saskatoon TB clinic or at rural Public Health Centers in Saskatchewan with experience in the administration and interpretation of the TST. For the majority of patients, blood for QFT-GIT assay was drawn and the TST was planted at the same visit. In the case of the two-step TST, this would have occurred with the first step. Due to scheduling and to minimise patient travel, these tests occasionally occurred on separate dates.

Patients with negative baseline TST and QFT-GIT results did not have further follow-up within the study. Patients with a baseline two-step TST ≥10 mm and/or a positive baseline QFT-GIT

result were reviewed by the TB clinic approximately three and six months after starting anti-TNF medication. At the three and six month clinic visits, patients were asked about current and recent medications, whether they had remained on the anti-TNF medication, and whether they had symptoms of TB. A repeat one-step TST and chest x-ray were performed, and blood was drawn for repeat QFT-GIT testing. Only those patients that underwent both TST and QFT-GIT at baseline, three and six months, and remained on anti-TNF medication for the six month duration of the study (evaluable group) were included in the analysis.

Statistical Analysis

The sample size estimation was based on an alpha of 0.05, a beta of 0.20, with a clinically significant effect size of 5 mm and a standard deviation of the difference between repeated TST test measurements of 5 mm (based on initial pilot data) providing a minimum sample size requirement of 8 subjects.

SPSS v.12.0 was employed for data entry and analysis. Frequencies and means were calculated. Paired *t*-tests were employed for comparison of within subject continuous data. Independent samples two-tailed *t*-tests were used for comparison of non-paired continuous data. Repeated-measures ANOVA was used for comparison of longitudinal continuous data. Chi-square testing and Fisher's exact test were employed for categorical data comparisons. A kappa coefficient was used to evaluate the agreement between the TST and QFT-GIT (17).

Results

Of 106 patients eligible to be screened for participation in this study, 91 consented to participate. Of these 91 patients who were screened by QFT-GIT and TST, 20 had only a positive (≥10 mm) TST, two had only a positive QFT-GIT, and four had positive results for both. There were four patients with 'indeterminant' QFT-GIT results: one declined further participation in the study, one was not followed, and the other two had negative QFT-GIT results upon repeat testing. All four had 0 mm TSTs. The overall agreement between baseline TST and QFT-GIT was poor (kappa coefficient = 0.180, p=0.020). Of the 26 patients with positive screening, 14 were initiated and maintained on anti-TNF therapy for the six month study period. Of the remaining patients, seven did not start anti-TNF therapy, one was lost to follow-up, and four started but received anti-TNF therapy for less than three months. No patients were identified as having active TB as the reason for failure to initiate or continue anti-TNF therapy. None of the patients followed in this study received TB prophylaxis after informed discussion, choosing close follow-up over chemical prophylaxis. Chest x-ray findings were negative for TB in all patients and were unchanged throughout the study period, and no patients reported or demonstrated signs or symptoms of TB infection at any of the follow-up visits.

Of the 14 patients on anti-TNF therapy studied over the six month time period, twelve patients met criteria for inclusion in the Evaluable Group as two patients had only two of the three TSTs performed.

Baseline demographic information on the Evaluable Group is outlined in Table I. The TST and QFT-GIT results are presented aggregate in Table II and individually in Table III.

The baseline mean TST measurement for the 12 participants in the Evaluable Group was 13.9 mm (SD 11.4); this increased to a mean of 16.8 mm (SD 9.3) post-booster. Two patients had initial TST measurements of zero and required a booster TST (Table III). The post-booster baseline TST values will be used for the remainder of the discussion.

The second TST was performed approximately 3 months post-anti-TNF initiation (mean 3.4 months; range 3–6). Comparison of the baseline and three month TST results revealed an overall decrease in TST measurement from 16.8 mm (SD 9.3; range 10–45 mm) to 10.0 mm (SD 6.3; range 0–18 mm). This decrease was statistically significant p = 0.013 (95% CI: 1.77, 11.89). Additionally, 3 of 12 patients had three month post-TNF initiation TST measurements which had dropped below 5 mm.

Table I. Baseline demographic information for the evaluable group.

n.	12
Mean age (years)	55.7 (SD 15.1; range 31-81)
Gender	
Male	5
Female	7
BMI	28.9 (SD 3.7; range 23.1-34.6)
BCG	
Yes	5
No	4
Uncertain	3
Ethnicity	
Caucasian	8
Indian	2
Metis	1
Other	1
Anti-TNF	
Infliximab	4
Etanercept	5
Adalimumab	3
Disease	
Rheumatoid arthritis	8
Psoriatic arthritis/psoriasis	3
Ankylosing spondylitis	1

Table II. Aggregate Tuberculin skin test and QuantiFERON-TB Gold In-Tube results.

	Baseline TST (baseline + booster)	3 months^	6 months#
TST (mm)	16.8 (SD 9.3; range 10-45)	10.0 (SD 6.3; range 0-18	14.5 (SD 7.7; range 0-25)
≥10 mm (n)	12	8	10
≥5 mm (n)	12	9	11
QuantiFERON-TB Gold (In-Tube Method) Interferon			
Positive	2	2	2
Negative	10	10	10
Agreement (TST ≥5 mm, interferon +) n	2/12	2/9	2/11

^Mean interval between baseline and 3 month testing time 3.4 months (SD 0.84; range 3-6 months);
#Mean interval between 3 month and 6 month testing time 4.1 months (SD 1.67; range 3-9 months).

Table III. Individual Tuberculin Skin Test and QuantiFERON-TB Gold In-Tube results.

Gender	Age (years)	Type of anti-TNF	TST n.1 (mm)	TST n.1+boost (mm)	3 month TST (mm)	Actual n. months between visits	6 month TST (mm)	Actual n. months between visits	QuantiFERON-TB Gold In-Tube result
male	56	etanercept	16	16	15	3	11	9	negative
male	57	infliximab	0	10	0	3	0	4	negative
female	65	adalimumab	18	18	12	3	10	5	negative
male	31	etanercept	15	15	10	4	14	4	negative
female	57	adalimumab	14	14	5	6	8	3	negative
male	56	etanercept	45	45	8	3	17	3	negative
female	32	etanercept	9	20	15	4	25	5	positive
female	63	infliximab	12	12	0	3	10	3	negative
female	38	etanercept	11	11	10	3	25	3	negative
female	70	adalimumab	0	14	4	3	14	3	negative
female	62	infliximab	15	15	15	3	15	3	negative
male	81	infliximab	12	12	16	3	25	4	positive

Interestingly, at the third visit a mean of 4.1 (range 3–9) months after the three-month assessment, a response recovery was observed with a mean TST measurement of 14.5 mm (SD 7.7); only one patient had a TST response which persisted at <5 mm. Figure 1 illustrates the evolution of the TST measurements over the study period. Employing repeated-measures ANOVA, significant within-subject differences in TST measurements were observed over time (p=0.048).

The QFT-GIT status of all patients was unchanged throughout the study. No patient's QFT-GIT results became negative or converted over the course of the study.

Discussion

We observed variation in the TST response during 6 month follow-up of patients receiving anti-TNF therapy after positive baseline two-step TST and/or QFT-GIT results. Specifically, we recorded a significant decrease from baseline in the magnitude of the TST reaction at the three month time point. This decrease was sufficient to drop the response to <5 mm in a quarter of the Evaluable Group, demonstrating the importance of completing TST screening prior to initiation of anti-TNF therapy. The Canadian Tuberculosis Standards suggests TST results of 5-9 mm are significant in patients on immune suppression (1). The TST measurements were significantly larger and reverted to ≥5mm in 2 of 3 patients at the 6-month evaluation compared to 3 months. Reversion of the TST reaction has been recognised as more common in particular subsets of patients, including those who are older, those who demonstrate the booster phenomenon, and those with a more modest magnitude of TST reaction (18). In our study, two of the three patients whose TST results became <5 mm at the first follow-up visit point had demonstrated the boosting phenomenon at baseline. However, the subsequent recovery of the TST reaction by the second follow-up in the majority of our study patients as outlined in Table III is unexplained and unlikely to be attributable to a boosting phenomenon. The underlying mechanism for the observed variation in TST response is unclear and merits further investigation.

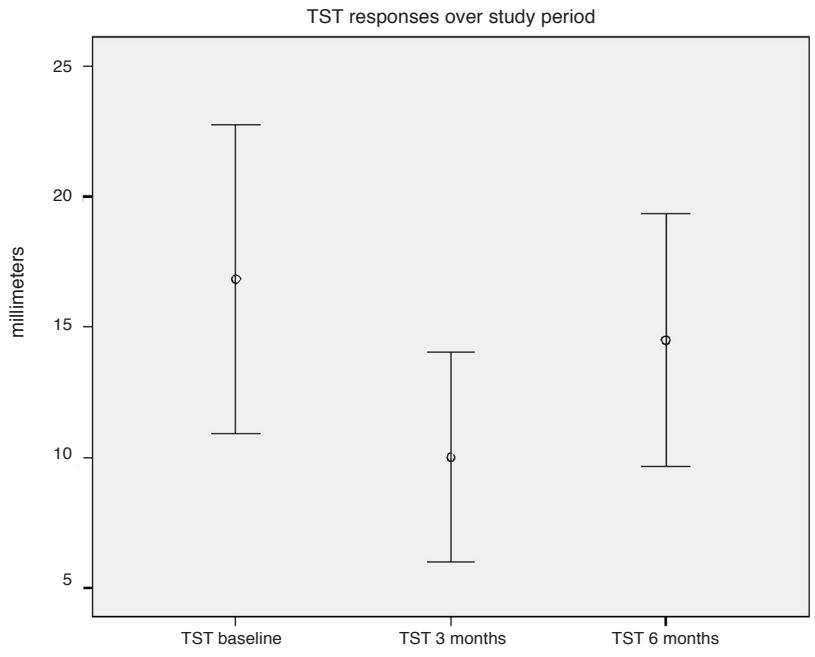

Fig. 1.

Our results are in contrast to other reports that indicated the TST was not affected by infliximab (14, 19). Although Joven and colleagues did report that one of their 13 patients with a positive TST did revert to a negative reaction after infliximab, overall the TST measurement increased after initiation of infliximab. Hatemi *et al*. reported no significant difference overall between baseline and repeat TST in patients receiving infliximab, they did observe that of their nine patients with an initially positive TST, the TST reaction decreased in seven patients. Differences in observations may be in part related to the timing of the repeat assessments.Our baseline and first

follow-up measures were done a mean of 3.4 months apart, an earlier time interval than employed in these previous reports (14, 19). We acknowledge that some our 3 and 6 month evaluations occurred outside of these time windows. This is a result of the infrequency of the TB clinics and the remote location of some of the patients. Due to our small overall numbers, these patients were included in the Evaluable Group. The exact timing of their visits are documented in Table III.

The potential for immunomodulators to affect IGRA results is a consideration. Papay *et al*. (20) recently reported a significantly lower rate of IGRA positivity in inflammatory bowel disease patients on immunomodulators compared to those who were not. Interestingly, of the 8.7% of their study population who been exposed to infliximab prior to screening, none had a positive IGRA (20). These findings support close clinical follow up of patients with indeterminant IGRA results in this susceptible population. In our Evaluable Group, immunomodulatory baseline medications included prednisone 5-10 mg/day in three patients, methotrexate in two patients, leflunomide in 3 patients at the time of baseline testing and within the three months prior to baseline testing in another patient, hydroxychloroquine in two patients and anakinra in one patient.

Due to the small study population, subgroup comparisons based on gender, age or specific anti-TNF therapy would have limited value. However, comparing PPD measurements in the five patients receiving etanercept (anti-TNF receptors) and the seven receiving monoclonal antibody forms of anti-TNF therapy (infliximab and adalimumab) revealed that the patients receiving the monoclonal antibody therapy were more likely to have a significant drop in PPD measurement at three months post-initiation (from baseline of 13.57 (SD 2.57) to a second measurement of 7.43, (SD 6.83, p=0.034; 95% CI: 0.63, 11.65) than those on etanercept (p=0.185). By the third timepoint measurement there was a recovery in PPD response such that there was no significant difference when compared to baseline measurements for either therapeutic group. Few in this group of patients initiating anti-TNF therapy had a positive QFT-GIT result, and no changes in the QFT-GIT results were evident during the six month study period. There was poor agreement between TST and QFT-GIT results, consistent with the findings of previous investigators (4).

Our observed decrease in TST response during the first six months of anti-TNF therapy support heightened clinical monitoring for TB symptoms and signs and careful interpretation of LTBI screening tests in this population.

Acknowledgements

We would like to thank the patients involved in this project for their participation and interest. We are grateful to Drs Hoeppner, Ward and Fenton for seeing study patients in their TB clinics. Dr Hoeppner was also integral to the study design. We would also like to thank Mrs Evelyn McLeod for her administrative assistance, Mrs Karen Mochoruk for her laboratory assistance, the nurses at the TB clinic for performing the TSTs, and Dr Lilian Urroz and Mrs Xia Liu for their research assistance and phlebotomy skills. We are grateful to the Royal University Hospital Foundation for funding this research.

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