Role of QuantiFERON-TB gold test in monitoring treatment of presumed ocular tuberculosis

Md. Mezbahul Alam[1], Tanmay R. Shuvo[1], Rahmat -E- khuda[2], SM Shafiul Bari Rasel[3], Ummay kawsar[4], Kabir hossain[1], Farjana S. Shimu[1], Ershadul H. Rahat[1], Atiqul Haque[1], Kamrunnahar[1], Arfin Akter[1]

Abstract

Background: *Tuberculosis (TB) is a major global health problem, with an estimated 9.6 million new cases and 1.5 million deaths per year. It represents one of the main causes of mortality and morbidity in the world. Recent research has focused on the use of interferon gamma (IFN-g) release assays (IGRAs) as a biomarker of treatment success. Animal and human studies have shown a relationship between the Mycobacterium tuberculosis (MTB) bacillary load and the magnitude of IFN-g responses to MTB antigen. It has therefore been postulated that a decrease in the magnitude of IFN-g responses to MTB-specific peptides measured by IGRA can be used as a biomarker of cure. To assess the utility of IGRAs for this purpose, a number of studies have investigated the kinetics of IGRA responses during the treatment of TB. We did a retrospective review of response to treatment of presumed ocular TB using IGRA.*

Method: *We retrospectively reviewed thirty nine cases diagnosed with presumed ocular tuberculosis successfully treated with a complete course of ATT and a minimum follow-up of at least 6 months following completion of ATT. in addition we had a quantiFERON-TB gold test (QFT-G test) done prior to starting ATT. ATT consisted of isoniazid (INH) 5 mg/kg/day, rifampicin (RIF) 600 mg/day, ethambutol 15 mg/kg/ day, and pyrazinamide (PYZ) 25 mg/kg/day for initial 2 months, followed by INH + RIF for 7 months. Successful response was indicated by absence of recurrences of inflammation following ATT. The study period was between January 2019 and December 2021.*

Result: *Thirty nine patients with suspected tubercular uveitis who underwent QFT-G test were analyzed retrogradely. Among them seventeen (43%) had QFT-G positive. Of them 10 were male and 7 were female. After 6 months following completion of ATT course repeat QFT-G test become negative in 4 cases. Among them 3 were vasculitis retinae and 1 was serpiginous like choroiditis. The mean age of QFT-G positive patients was 35.7years (range 12-51years). Male –female ratio was 1:1.42.*

Conclusion: *IFN-γ concentrations may offer some value in monitoring treatment response among OTB patients as our findings also show a significant decrease in IGRA values after 6 months of ATT conclusion, although most of our patients continued to be IGRA positive. Given the complexity of the immune response to TB, it may not be surprising that the measurement of a single cytokine does not provide sufficient discrimination to assess response to treatment. Studies investigating the potential of other novel biomarkers or combination of biomarkers with improved sensitivity and specificity are urgently needed to accurately determine the potential of immunoassays to assess the response to anti-tuberculous treatment.*

Keywords: *QFT-G test, presumed ocular TB.*

Introduction

Tuberculosis (TB) is a major global health problem, with an estimated 9.6 million new cases and 1.5 million deaths per year.[1]It represents one of the main causes of mortality and morbidity in the world [1]. Tuberculosis is characterized by pulmonary and extrapulmonary manifestations

1. National Institute of Ophthalmology and Hospital, Dhaka.
2. Dept of eye, Rajshahi Medical College and Hospital, Rajshahi
3. Sheikh Fazilatunnesa Mujib Eye Hospital and Training Institute, Gopalgonj
4. Dept of ophthalmology, MH. Samorita medical college, Dhaka

Address of Correspondence: Dr. Mezbahul Alam, Associate Professor, Department of Vitreo-retina, National Institute of Ophthalmology, Sher-e-banglanagar, Dhaka, Email: mejbah1549@yahoo.com

which may involve the skin, eye, and nervous, cardiovascular, gastrointestinal, or genitourinary systems.[2]The main manifestation of ocular TB is uveitis. Prevalence of TB uveitis (TBU) is estimated between 9% and 11% in endemic countries and between 1% and 6% in nonendemic countries.[3-6]Intraocular TB is rare in active lung TB, while it is more frequent in patients with advanced tubercular lesions or extrapulmonary forms.[7]Trauma, immunosuppression, and malnutrition are predisposing factors for development of intraocular TB.[8] Pathogenesis of ocular involvement in TB is controversial. Two possible pathophysiological mechanisms have been proposed to explain the inflammatory reaction caused by Mycobacterium tuberculosis (MTB): hematogenous spread with direct invasion of MTB in the eye[9,10]and delayed hypersensitivity reaction secondary to MTB located anywhere on the body.[10,11] The immune-mediated mechanism of inflammation and the paucibacillary nature of OTB make confirmatory techniques, such as MTB culture, acid-fast bacilli smear, or polymerase chain reaction (PCR) from ocular samples, have poor sensitivity (20–30%).[9,12] Currently, there is no definitive non-invasive method to confirm OTB and its diagnosis is mainly presumptive. Nonetheless, diagnosis accuracy has increased in the last years with the introduction of interferon-gamma release assays (IGRAs).[13] This powerful test can measure in vitro the production of interferon-gamma (IFN-ã) released by T-cells in peripheral blood, in response to MTB antigens ESAT-6, CFP-10, and TB 7.7. Unlike the tuberculosis skin test (TST), IGRA test is not affected by Bacillus Calmette–Guérin (BCG) vaccination and most non-tuberculous Mycobacterium infection.[13] It has been especially useful to detect latent tuberculosis infection (LTBI) in settings where BCG vaccination is mandatory, such as in Bangladesh.[14] In pulmonary and LTBI, IGRA test has shown specificity between 91% and 99% and sensitivity between 89% and 91%.[15,16] In OTB, these values are not well established, although values between 80% and 85% had been reported for sensitivity and specificity, respectively, in a BCG-vaccinated and nonendemic population.[13,15]

A biomarker to indicate successful tuberculosis (TB) treatment would be a major advance for the management and control of TB globally. Recent research has focused on the use of interferon gamma (IFN-g) release assays (IGRAs) as a biomarker of treatment success. Animal and human studies have shown a relationship between the Mycobacterium tuberculosis (MTB) bacillary load and the magnitude of IFN-g responses to MTB antigens.[12,17] It has therefore been postulated that a decrease in the magnitude of IFN-g responses to MTB-specific peptides measured by IGRA can be used as a biomarker of cure.[18]To assess the utility of IGRAs for this purpose, a number of studies have investigated the kinetics of IGRA responses during the treatment of TB. We did a retrospective review of response to treatment of presumed ocular TB using IGRA.

Method

We retrospectively reviewed thirty nine cases diagnosed with presumed ocular tuberculosis successfully treated with a complete course of ATT and a minimum follow-up of at least 6 months following completion of ATT; in addition we had a quantiFERON-TB gold test (QFT-G test) done prior to starting ATT. The diagnosis of presumed ocular tuberculosis was made on the criteria by Gupta et al.[19] ATT consisted of isoniazid (INH) 5 mg/kg/day, rifampicin (RIF) 600 mg/day, ethambutol 15 mg/kg/ day, and pyrazinamide (PYZ) 25 mg/kg/day for initial 2 months, followed by INH + RIF for 7 months. Successful response was indicated by absence of recurrences of inflammation following ATT. The study period was between January 2019 and December 2021. Demographic, clinical, and laboratory data were collected. Demographic data included age and sex. Clinical data included history of systemic tuberculosis, usage of oral steroids in the last 6 months prior to starting ATT, and the type of uveitis. Laboratory data prior to starting ATT included results of erythrocyte sedimentation rate (ESR), Mantoux test, serum angiotensin converting enzyme, liver function tests and QFT-G test. Computed tomography (CT) of the thoraxwas done in selective cases. Patients receiving ATT were re-tested for QFT-G test at 6

months after ATT completion. Patients were excluded if they had indeterminate IGRA result, daily prednisone use 10 mg/day or any immunosuppressant treatment if taken 3 months before testing for IGRA. The study was conducted as per the Declaration of Helsinki and an approval to collect retrospective data was obtained from our hospital review board. All statistical analysis were done using SPSS software. Descriptive analysis was performed and independent variables were analyzed

Interferon-gamma release assay The Cellestis Quantiferonw(R) TB Gold IFN-ã release assay (IGRA) was performed according to the manufacturer's instructions (Qiagen, Valencia, CA, USA). The plasma concentration of IFN-ã was determined to be 'negative', 'positive' or 'intermediate' by the manufacturer's software (cut-off of 0.35 IU/ml. For repeat IGRA in treated patients, reversions were defined as baseline IFN-γ > 0.35 IU/mL.

Result

Thirty nine patients with suspected tubercular uveitis who underwent QFT-G test were analyzed retrogradely. Among them seventeen (43%) had QFT-G positive. Of them 10 were male and 7 were female. After 6 months following completion of ATT course repeat QFT-G test become negative in 4 cases. Among them 3 were vasculitis retinae and 1 was serpiginous like choroiditis. The mean age of QFT-G positive patients was 35.7years(range 12-51years). Male –female ratio was 1:1.42. The pattern of uveitis among QFT-G test positive patients are given below. (Table1.1)

Discussion

The search for a TB biomarker that reflects the true stage of disease, from non-infected to latently infected to active disease, to successful vs. unsuccessful treatment, to cure vs. relapse, continues to be elusive despite much research in areas ranging from microbiology to radiology to gene expression profiles. The M. tuberculosis-specific CD4þ Th1 cell response is crucial to the immunological response to M. tuberculosis infection, as this recruits and activates innate immune cells and produces cytokines such as IFN-ã. [20] The importance of IFN-ã is demonstrated by the susceptibility to mycobacterial infections of those with innate or acquired impaired IFN-ã mediated immunity.[21,22] Because IFN-ã production from Tcells increases in response to increased TB antigenic burden, a decline in IFN-ã concentrations may signal a successful treatment response. As IFN-ã expression in response to tuberculous infection is easily measured using the QuantiFERON-TB Gold or TSPOT.TB IGRA kits, many studies have used these kits to assess whether changes in IFN-ã levels correlate with treatment response.

The prognostic use of IGRA tests as marker for response to therapy is not established and data are conflicting, performed in various endemic settings and with different IGRAs and upper and lower cut-off levels. Most studies have been performed on patients treated for active TB.[13,23-26]Pai et al. show persistence of QFT-TB responses during treatment.[24] In contrast, Katiyar et al. found that only 48% were positive by the same assay after 6

Different types of uveitis and QFT-G test (table1.1)

Pattern of uveitis (N=17)	QFT-G test +ve before treatment	QFT-G test +ve after treatment
Anterior Uveitis	1	1
Intermediate Uveitis	2	2
Pan Uveitis	1	1
Scleritis	2	2
Vasculitis retinae	3	-
Serpigenous like choroiditis (SLC)	3	2
Multifocal choroiditis (MFC)	5	5
Choroidal abscess	1	1

months.[25] Also studies of TSPOT.TB are conflicting as Dheda et al. report 81% with negative test in late phase therapy of patients with active TB.[26] The corresponding numbers in Ribeiro et al's study is only 10%.[13] The large variation between studies in the IGRA reversion rate at the end of treatment (between 0 and 72%) suggests that measuring categorical changes in IGRA does not offer a reliable method for monitoring anti-tuberculous treatment in either active or latent TB.[27]

In our study, the QFT-G results were not affected by intake of oral steroids. This is concurrent with existing reports that QFT results are not affected by systemic steroids.[28-30] Most of our patients continued to be IGRA positive with reduction of the IFN-ã values after 6 months of ATT completion. In our study the mean drop in IGRA value to 1.419 post treatment,in agreement with previous results.[30] Lee et al.[15] have shown in their series of cases with active tuberculosis that less than half exhibited a QFT-G reversion even after successful response with ATT. But they did see decreasing IFN-ã levels with time in their cases. Komiya et al.[31] showed that reversion to a negative QFT-G result was closely associated with the magnitude of the IFN-ã response prior to treatment and increasing age in active TB. The fact that most patients continued to be IGRA positive after 6 months of ATT conclusion could be explained by the long mean time between ocular symptoms onset and ATT initiation.[32] A TB infection that persisted for a long time before being cleared with ATT could have triggered a robust and durable immunological memory that leads to persistent IGRA immunoreactivity.[33,34] The study has limitations. This is a retrospective study and we have included only those cases of presumed ocular tuberculosis that had a QFT-G test done prior to starting ATT. QFT-G is an expensive test. This limits the increased use of this test in our patient population. Thus, the sample size of the study group is small in comparison to the larger number of cases of presumed ocular tuberculosis we routinely see. Peppleet al.[35] have discussed the challenges in interpretation of a positive QFT-G result in uveitis in nonendemic countries with a low pretest probability of the disease. In our study, we do not have controls to assess the likelihood ratio of a positive QFT-G, which is the third limitation of this study. Nevertheless, this paper reviews the results of QFT-G in our patient population (high TB endemic country) of presumed ocular tuberculosis treated successfully with ATT.

Conclusion

IFN-ã concentrations may offer some value in monitoring treatment response among OTB patients as our findings also show a significant decrease in IGRA values after 6 months of ATT conclusion, although most of our patients continued to be IGRA positive. Given the complexity of the immune response to TB, it may not be surprising that the measurement of a single cytokine does not provide sufficient discrimination to assess response to treatment. Studies investigating the potential of other novel biomarkers or combination of biomarkers with improved sensitivity and specificity are urgently needed to accurately determine the potential of immunoassays to assess the response to anti-tuberculous treatment.

References

1. World Health Organization. Global tuberculosis report, 2015. WHO/HTM/TB/2015.22. Geneva, Switzerland: WHO, 2015.
2. A. R. Kee, J. J. Gonzalez-Lopez, A. Al-Hity et al., "Anti-tubercular therapy for intraocular tuberculosis: a systematic review and meta-analysis," Survey of Ophthalmology, vol. 61, no. 5, pp. 628–653, 2016.
3. M. Ishihara and S. Ohno, "Ocular tuberculosis," Nippon Rinsho, vol. 56, pp. 3157–3161, 1998.
4. V. Gupta, A. Gupta, and N. A. Rao, "Intraocular tuberculosisan update," Survey of Ophthalmology, vol. 52, no. 6, pp. 561–587, 2007.
5. N. J. Cutrufello, P. C. Karakousis, J. Fishler, and T. A. Albini, "Intraocular tuberculosis," Ocular Immunology and Inflammation, vol. 18, no. 4, pp. 281–291, 2010.
6. M. Ang and S.-P. Chee, "Controversies in ocular

tuberculosis," British Journal of Ophthalmology, vol. 101, no. 1, pp. 6–9, 2017.

7. J. Biswas and S. S. Badrinath, "Ocular morbidity in patients with active systemic tuberculosis," International Ophthalmology, vol. 19, no. 5, pp. 293–298, 1995.
8. F. I. Shakarchi, "Ocular tuberculosis: current perspectives," Clinical Ophthalmology, vol. 9, pp. 2223–2227, 2015.
9. Chegou N N, Heyckendorf J, Walzl G, Lange C, Ruhwald M. Beyond the IFN-gamma horizon: biomarkers for immunodiagnosis of infection with Mycobacterium tuberculosis. EurRespir J 2014; 43: 1472–1486.
10. Pai M, Denkinger C M, Kik S V, et al. Gamma interferon release assays for detection of Mycobacterium tuberculosis infection. ClinMicrobiol Rev 2014; 27: 3–20.
11. Sauzullo I, Mengoni F, Lichtner M, et al. In vivo and in vitro effects of antituberculosis treatment on mycobacterial interferon-gamma T-cell response. PLOS ONE 2009; 4: e5187.
12. Carrara S, Vincenti D, Petrosillo N, Amicosante M, Girardi E, Goletti D. Use of a T cell-based assay for monitoring efficacy of antituberculosis therapy. Clin Infect Dis 2004; 38: 754–756.
13. Ribeiro S, Dooley K, Hackman J, et al. T-SPOT.TB responses during treatment of pulmonary tuberculosis. BMC Infect Dis 2009; 9: 23.
14. Syed Manzoor Ahmed Hanifi, Mizanur Rahman. BCG vaccination in Bangladesh: should it be given at birth or given along with pentavalent? International Journal of Epidemiology, 2020, 1–4
15. Lee S W, Lee C T, Yim J J. Serial interferon-gamma release assays during treatment of active tuberculosis in young adults. BMC Infect Dis 2010; 10: 300.
16. Denkinger C M, Pai M, Patel M, Menzies D. Gamma interferon release assay for monitoring of treatment response for active tuberculosis: an explosion in the spaghetti factory. J ClinMicrobiol 2013; 51: 607–610.
17. Pathan AA, Wilkinson KA, Klenerman P, McShane H, Davidson RN, Pasvol G, et al. Direct ex vivo analysis of antigen-specific IFN-gammasecreting CD4 T cells in Mycobacterium tuberculosis-infected individuals: associations with clinical disease state and effect of treatment. J Immunol 2001;167:5217e25.
18. Lalvani A. Counting antigen-specific T cells: a new approach for monitoring response to tuberculosis treatment? Clin Infect Dis 2004;38:757e9.
19. Gupta V, Gupta A, Rao NA. Intraocular tuberculosis—an update. SurvOphthalmol. 2007;52:561–587.
20. Prezzemolo T, Guggino G, La Manna M P, Di Liberto D, Dieli F, Caccamo N. Functional signatures of human CD4 and CD8 T_cell responses to Mycobacterium tuberculosis. Front Immunol 2014; 5: 180.
21. Remus N, Reichenbach J, Picard C, et al. Impaired interferon gamma-mediated immunity and susceptibility to mycobacterial infection in childhood. Pediatr Res 2001; 50: 8–13.
22. Xie Y L, Rosen L B, Sereti I, et al. Severe paradoxical reaction during treatment of disseminated tuberculosis in a patient with neutralizing anti-IFNc autoantibodies. Clin Infect Dis 2016; 62: 770–773.
23. Vekemans J, Lienhardt C, Sillah JS, et al: Tuberculosis contacts but not patients have higher gamma interferon responses to ESAT-6 than do community controls in The Gambia. Infect Immun 2001, 69:6554-7.
24. Pai M, Joshi R, Bandyopadhyay M, et al: Sensitivity of a whole-blood interferon-gamma assay among patients with pulmonary tuberculosis and variations in T-cell responses during anti-tuberculosis treatment. Infection 2007, 35:98-103.
25. Katiyar SK, Sampath A, Bihari S, Mamtani M, Kulkarni H: Use of the QuantiFERON-TB Gold In-Tube (R) test to monitor treatment efficacy in active pulmonary tuberculosis. International Journal of Tuberculosis and Lung Disease 2008, 12:1146-52.
26. Dheda K, Pooran A, Pai M, et al: Interpretation of Mycobacterium tuberculosis antigen-specific IFN-gamma release assays (T-SPOT.TB) and factors that may modulate test results. Journal of Infection 2007, 55:169-73.
27. Vanessa C, Yu H, Christel Z, Tom C, Nigel C: Interferon gamma release assays for monitoring the response to treatment for tuberculosis: A systematic review. Tuberculosis 95.2015; 639-650.

28. Sudarshan S, Ganesh SK, Balu G, et al. Utility of QuantiFERON-TB Gold test in diagnosis and management of suspected tubercular uveitis in India. IntOphthalmol. 2012;32:217–223.

29. Shovman O, Anouk M, Vinnitsky N, et al. QuantiFERONTB Gold in the identification of latent tuberculosis infection in rheumatoid arthritis: a pilot study. Int J Tuberc Lung Dis. 2009;13:1427–1432.

30. Babu K, Bhat SS, Philips M, Subbakrishna DK. Review of results of QuantiFERON TB gold test in presumed ocular tuberculosis in a South Indian patient population. OculImmunolInflamm. 2016 Oct;24(5):498–502. doi: 10.3109/ 09273948.2015.1010094.

31. Komiya K, Ariga H, Naga H, et al. Reversion rates of Quantiferon TB gold test are related to pre treatment IFN-g levels. J Infect. 2011;63:48–53.

32.Y. Fernández-Zamora et al. Role of Interferon-gamma release assay for the diagnosis and clinical follow up in ocular tuberculosis.OculImmunolInflamm. 2022 jan; 001–008.

33. Behr MA, Edelstein PH, Ramakrishnan L. Revisiting the timetable of tuberculosis. BMJ. 2018 Aug 23;362:k2738. doi: 10.1136/bmj. k2738.

34. Pourakbari B, Mamishi S, Benvari S, et al. Can interferon-ã release assays be useful for monitoring the response to anti-tuberculosis treatment?: a systematic review and meta-analysis. Arch ImmunolTherExp (Warsz). 2020 Feb 3;68(1):4. doi: 10.1007/s00005-020- 00568-4.

35. Pepple KL, Gelder RV, Forooghian, F. Caveats about QuantiFERON-TB gold testing for uveitis. Am J Ophthalmol. 2014;157:752–753.