BRIEF COMMUNICATION

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Differences in Clinical Manifestations according to the Positivity of Interferon-γ Assay in Patients with Intestinal Tuberculosis

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Intestinal tuberculosis (ITB) remains prevalent in Asia. An interferon-γ assay (QuantiFERON-TB gold test [QFT]) is considered to be an effective supplementary tool for diagnosing ITB. We retrospectively analyzed the clinical features of ITB patients based on the initial results of QFT. A total of 109 patients with ITB were enrolled, and 82 patients (75.2%) showed positive QFT results. In the QFT-positive group, the mean age (44.1±12.0 years) was significantly higher than that in the QFT-negative group (37.0±14.8, p=0.0096). Abdominal pain (p=0.006) and diarrhea (p=0.030) were more frequent in the QFT-negative group. Further, C-reactive protein (CRP) levels were significantly higher in the QFT-negative group (6.4±9.9 mg/dL) than in the QFT-positive group (1.3±2.3, p<0.001). Multivariate analysis confirmed that younger age (p=0.016), diarrhea (p=0.042), and high levels of CRP (p=0.029) were independent predictors of QFT-negative results in patients with ITB. These results suggest that prior exposure to TB, reflected by QFT positivity, may cause mild inflammation in patients with ITB. **(Gut Liver 2016;10:649-652)**

Key Words: Intestinal tuberculosis; Interferon-gamma assay; C-reactive protein

INTRODUCTION

Tuberculosis (TB) remains prevalent worldwide with considerable morbidity and mortality rates.[1] Currently, one-third of the world's population is infected with TB, demonstrating its resurgence.[1] Intestinal tuberculosis (ITB) is a form of extrapulmonary TB that involves the gastrointestinal tracts. ITB is generally rare, accounting for 1% to 3% of TB cases,[2] however, the incidence of ITB in both developing and developed countries has increased steadily in recent 20 years.[3] It is important to differentiate ITB from inflammatory bowel disease (IBD), especially Crohn's disease (CD) in Asian countries, including China, Taiwan, India, and Korea, because of the increasing prevalence of CD with a still-high prevalence of ITB.[4-7] However, it is sometimes challenging to distinguish these two diseases because of their similar clinical manifestations and examination results.[7,8]

Recently, an interferon-γ release assay (IGRA) that measures the release of interferon-γ (IFN) after stimulation *in vitro* by *Mycobacterium tuberculosis*-specific antigens has been used as an aid in diagnosing both latent TB infection (LTBI) and active TB disease.[9-11] IFN-γ plays an important role in control of cell-mediated immune reactivity to *M. tuberculosis*. There are two IGRAs, QuantiFERON-TB gold test (QFT) (Cellestis Ltd., Carnegie, Australia) and T-SPOT.TB (TSPOT) (Oxford Immunotec Ltd., Abingdon, UK) tests available and their results are comparable. IGRA also has been utilized in the differential diagnosis of ITB and CD.[12-14] In our previous study, the positive rate for the QFT in patients with ITB was significantly higher than it was in patients with CD (67% vs 9%, p<0.001) and we suggested that the QFT is a limited but useful diagnostic aid in the diagnosis of ITB.[12]

However, the clinical implication of the results of QFT in patients with ITB remains unknown. Only two-thirds of ITB patients showed positive QFT results, with the remainder showing negative results. Therefore, we evaluated the clinical differences of ITB patients according to the results of QFT.

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MATERIALS AND METHODS

From April 2007 to July 2013, 109 consecutive patients with ITB were enrolled retrospectively from four hospitals in Korea. They all performed the QFT at the time of ITB diagnosis. We used the Korean diagnostic guideline of ITB, which classifies the ITB into definite or probable ITB.[15] Definite diagnosis of ITB was made on histopathologic findings or culture results.[15] In patients suspected of having ITB, those with the characteristic endoscopic findings of ITB or positive QFT results, but without definite evidence of ITB, empirical anti-TB therapy was tried for 2 to 3 months. The probable diagnosis of ITB was made when they showed marked improvement in the clinical and endoscopic findings to empirical anti-TB therapy.[15] We compared the clinical parameters between the two groups according to the results of QFT including clinical symptoms, extents of colonoscopic involvement, laboratory findings, and existence of extraintestinal TB. This study was approved by the ethics committees of each institution.

The QFT (Cellestis Ltd.) test was performed in two stages according to the manufacturer's instructions. A positive response value of 0.35 IU/mL was used as the cutoff for the QFT test. If the results showed indeterminate, we regarded the data as negative. Comparisons between the two groups were made using Student t-test, Pearson chi-square test, and Fisher exact test. The multivariate analysis was done using logistic regression analysis. Two tailed p-values below 0.05 were considered statistically significant. We conducted statistical analysis using PASW Statistics 17.0 (SPSS Inc., Chicago, IL, USA).

Table 1. Baseline Characteristics of Patients with Intestinal Tuberculosis

Characteristic	No. of patients (n=109)
Male sex	55 (50.5)
Age, yr	42.6±14
Definite diagnosis (n=26)	
Caseating granuloma	6 (23.1)
Tissue AFB positive	4 (15.4)
Tissue culture positive	16 (61.5)
Positivity of QFT	82 (75.2)
Prior active TB history	15 (13.8)
Extraintestinal TB	29 (26.6)
Underwent surgery	6 (5.5)
BCG vaccination	89 (81.7)
Underlying disease	11 (10.1)

Data are presented as the number (%) or mean±SD. Extraintestinal TB is defined as TB involved outside of the gastrointestinal tract.
AFB, acid fast bacilli; QFT, QuantiFERON-TB gold test; TB, tuberculosis; BCG, Calmette-Guérin bacillus vaccine.

RESULTS

A total of 109 patients (mean age, 42.6±14 years; male 55, female 54) were enrolled retrospectively in this study. According to the guideline for ITB diagnosis,[15] 26 (23.9%) had definite diagnosis of ITB and 83 (76.1%) had probable diagnosis. Table 1 shows the detailed characteristics of enrolled patients.

When the 109 patients separated into two groups according to the QFT results at the time of ITB diagnosis, 82 patients (75.2%) showed positive QFT results, whereas 27 (24.8%) showed negative results. Two patients showed indeterminate results and we grouped these two patients as QFT-negative group. The mean age at the time of diagnosis was 44.1±12 years in the QFT-positive group, which is significantly higher than the age of QFT-negative group (37.0±14.8 years, p=0.0096). In addition, in the QFT-negative group, patients complained significantly more

Table 2. Clinical Parameters of the Patients with Intestinal Tuberculosis Based on the Initial Results of QFT

Parameter	QFT-positive (n=82)	QFT-negative (n=27)	p-value
Age, yr	44.1±12.0	37.0±14.8	0.0096
Male sex	40 (48.8)	15 (55.6)	0.471
Clinical manifestation			
Abdominal pain	27 (33.0)	17 (63.0)	0.006
Diarrhea	14 (17.1)	10 (37.0)	0.030
Hematochezia	9 (11.0)	5 (18.5)	0.329
Fever	4 (4.9)	3 (11.1)	1.000
Weight loss	8 (9.8)	4 (14.8)	0.102
Nonspecific	34 (41.5)	5 (18.5)	0.031
Prior active TB history	12 (14.6)	3 (11.1)	0.645
Underlying disease	9 (11.0)	2 (7.4)	0.072
Colonoscopic involvement			0.198
Ileocecal area	28 (34.1)	7 (25.9)	
Less than 4 segments	24 (29.3)	4 (14.8)	
More than 4 segments	30 (36.6)	16 (59.3)	
Laboratory finding			
Initial hemoglobin, g/dL	13.07±1.93	13.62±2.14	0.209
Initial WBC, /mm^3	6,387±1,963	9,287±4,999	<0.001
Initial ESR, mm/hr	30.1±21.7	31.8±27.6	0.987
Initial CRP, mg/dL	1.3±2.3	6.4±9.9	<0.001
Initial albumin, g/dL	4.16±0.43	3.96±0.56	0.181
Granuloma in colonic tissue	35 (42.7)	9 (33.3)	0.095
Extraintestinal TB	20 (24.4)	9 (33.3)	0.592

Data are expressed as the mean±SD or number (%). Underlying disease means the accompanying liver cirrhosis, diabetes mellitus or malignancy. Extraintestinal TB is defined as TB involved outside of the gastrointestinal tract. Nonspecific symptoms are defined as the symptoms not presumed to be related to intestinal tuberculosis.
QFT, QuantiFERON-TB gold test; TB, tuberculosis; WBC, white blood cell; ESR, erythrocyte sedimentation rate; CRP, C-reactive protein.

Table 3. Logistic Regression Analysis with Intestinal Tuberculosis Based on the Initial Negative Results of QFT

	OR	95% CI	p-value
Age, yr	0.947	0.90–0.99	0.016
Abdominal pain	3.896	0.78–19.29	0.096
Diarrhea	3.936	1.05–14.70	0.042
Initial WBC, /mm^3	1.000	1.00–1.00	0.091
Initial CRP, mg/dL	1.146	0.914–1.001	0.029

QFT, QuantiFERON-TB gold test; OR, odds ratio; CI, confidence interval; WBC, white blood cell; CRP, C-reactive protein.
Significance at p<0.05 by logistic regression analysis.

about the symptoms of ITB, such as abdominal pain (p=0.006) and diarrhea (p=0.030), as compared to the QFT-positive group. However, there was no difference in gender (p=0.471), the frequency of accompanying extra-intestinal TB (p=0.592), extents of colonoscopic involvement (p=0.198), and the presence of granulomas in colonic tissues (p=0.095) (Table 2). Eleven patients had the underlying diseases such as liver cirrhosis, diabetes mellitus or malignancy. Nine patients were grouped in the QFT-positive group (11%) and two patients were QFT-negative group (7.4%), without statically significance (p=0.072) (Table 2). Laboratory tests revealed that C-reactive protein (CRP) levels at the time of ITB diagnosis were significantly lower in the QFT-positive group (1.3±2.3 mg/dL) as compared to the QFT-negative group (6.4±9.9 mg/dL, p<0.001). White blood cell count was also significantly lower in the QFT-positive group (6,387±1,963 /mm^3) than QFT-negative group (9,287±4,999/mm^3, p<0.001). However, no difference was found between groups regarding the levels of erythrocyte sedimentation rate (p=0.987), hemoglobin (p=0.209), and albumin (p=0.181) (Table 2). Multivariate analysis revealed that the younger age (OR, 0.947; p=0.016), diarrhea (OR, 3.936; p=0.042), and high level of CRP (OR, 1.146; p=0.029) were the independent predictors of QFT-negative results in patients with ITB (Table 3).

DISCUSSION

Although current ITB patients show mild clinical manifestations, such as lower surgery rate as compared to patients in the 1970s to 1990s,[16] the prevalence of ITB remains high and troublesome in Asian countries.[17] A recent meta-analysis confirmed that IGRA has good sensitivity (81%) and specificity (85%) for the diagnosis of ITB in Asia. The authors suggested that the overall accuracy of IGRA was relatively high for the diagnosis of ITB.[13] Another meta-analysis also suggested that IGRA has good specificity (87%) for accurate differential diagnosis of ITB from CD.[18] In this study, QFT positivity was seen in 82 patients with ITB (75.2%), which is similar to our previous study (67%).[12] Considering these results,[12-14,18] we can conclude that IGRA is a useful tool for the diagnosis of ITB.

However, although QFT is a good diagnostic tool for ITB, not all patients with ITB show positive QFT results. In this study, only 75.2% of ITB patients showed the positive QFT results. To our knowledge, this is the first report evaluating clinical manifestations according to the results of QFT in patients with ITB.

Interestingly, the mean age of QFT-positive group was higher than that of QFT-negative group. In addition, the clinical manifestations were milder in the QFT-positive group considering the low frequency of diarrhea and the findings concerning CRP levels. These results suggested that QFT-positive patients had later-onset development of ITB and had more mild inflammatory disease.

Indeed, most people infected by *M. tuberculosis* do not develop active TB with clinical symptoms. Initial *M. tuberculosis* infection might be eliminated or remain in an inactive and non-infectious state, which is called LTBI. Among them, 2% to 5% of patients may develop active TB. IGRA has been used most frequently to detect LTBI. We suppose that ITB also has the same sequence. Although there is no exact data on the incidence and prevalence of ITB in Korea, older individuals are more likely to have LTBI as compared to younger people. That is, among patients with LTBI, the state in which the bacterial replication is maintained at a subclinical level by the immune system, some patients develop to ITB as a result of several factors, such as malnutrition or immune dysfunction. Therefore, we suppose that many cases of ITB develop from LTBI and the possibility that prior exposure to TB may cause mild inflammation in patients with QFT-positive ITB.

QFT has high specificity for the diagnosis of TB because it examines the *M. tuberculosis* specific antigen (ESAT-6 or CFP-10), leading to the lack of cross-reaction with most non-tuberculous mycobacteria (NTM) or Calmette-Gue´rin bacillus vaccine (BCG). Accordingly, a recent meta-analysis reported that IGRA has high specificity for the diagnosis of LTBI particularly in populations that have received BCG vaccinations.[19] In this study, most ITB patients (81.7%) had a history of BCG vaccination according to national policy of Korea.

Our study has some limitations. First, we used a small sample size, as we enrolled ITB patients who had undergone QFT at the time of ITB diagnosis. In clinical practice, use of QFT as an aid for the diagnosis of ITB has been popular since 2009 in Korea. Therefore, the small study population is attributed to the lack of usage of QFT in the diagnosis of ITB before 2009. In addition, ITB patients without QFT test were excluded in this study and there could be selection bias. Second, this study was conducted retrospectively, as we were curious as to why some ITB patients showed negative QFT results. A future prospective study will provide more detailed information about the IGRA-negative ITB patients. Also, *in vitro* experiments on the mechanism underlying the sensitization of T-cells and clinical manifestations of ITB will give us an understanding of the pathophysiology of ITB. Third, we did not consider the factors which can affect on

the QFT results including the time of blood draw, blood volume, transportation temperature, or incubation time.[11]

In conclusion, QFT is a useful diagnostic tool for the diagnosis of ITB. The QFT-positive group showed mild inflammatory status considering less frequent clinical symptoms and lower levels of CRP as compared to the QFT-negative group. We suggest that prior exposure to TB may cause mild inflammation in patients with ITB.

CONFLICTS OF INTEREST

No conflict of interest relevant to this article was reported.

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