Original Article

Diagnostic Value of T-cell Interferon-γ Release Assays on Synovial Fluid for Articular Tuberculosis: A Pilot Study

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Abstract

Background: Tuberculosis (TB) remains a major global public health challenge. Articular TB is an important form of extrapulmonary tuberculosis, and its diagnosis is difficult because of the low sensitivity of traditional methods. The aim of this study was to analyze the diagnostic value of T-SPOT.TB on synovial fluid for the diagnosis of articular TB.

Methods: Patients with suspected articular TB were enrolled consecutively between August 2011 and December 2015. T-SPOT.TB was performed on both synovial fluid mononuclear cells (SFMCs) and peripheral blood mononuclear cells (PBMCs). The final diagnosis of articular TB was independent of the T-SPOT.TB result. The diagnostic sensitivity, specificity, predictive value, and likelihood ratio of T-SPOT.TB on SFMCs and PBMCs were analyzed.

Results: Twenty patients with suspected articular TB were enrolled. Six were diagnosed with articular TB, and 14 patients were diagnosed with other diseases. Sensitivity and specificity were 83% and 86% for T-SPOT.TB on SFMCs, and 67% and 69% for T-SPOT.TB on PBMCs, respectively. The positive predictive value (PPV) and negative predictive value (NPV) of T-SPOT.TB on SFMCs were 71% and 92%, respectively. The PPV and NPV were 50% and 82% for T-SPOT.TB on PBMCs.

Conclusion: Sensitivity, specificity, and NPV of T-SPOT.TB on SFMCs appeared higher than that on PBMCs, indicating that T-SPOT.TB on SFMCs might be a rapid and accurate diagnostic test for articular TB.

Key words: Articular Tuberculosis; Diagnosis; Interferon-γ Release Assays; T-SPOT.TB; Sensitivity; Specificity; Synovial Fluid

Introduction

Tuberculosis (TB) remains a leading infectious disease in the world. Worldwide 9.6 million people are estimated to have developed TB in 2014, and China accounted for 10% of the total TB cases.[1] Pulmonary TB is given the most attention for its public health relevance, however, extrapulmonary tuberculosis (EPTB) such as articular TB is also important. Approximately, 2.2% to 4.7% of all tuberculous cases involve the skeletal system.[2] If osteoarticular TB is diagnosed and treated at an early stage, approximately 90% to 95% of patients can achieve full recovery with near normal function.[3] Therefore, timely diagnosing is important. In articular TB, since the clinical specimens obtained from relatively inaccessible sites are often paucibacillary, the sensitivity of traditional diagnostic tests such as smear and culture is low.[4]

Interferon-γ (IFN-γ)-releasing assays (IGRAs) have recently shown promising results in diagnosing active pulmonary or EPTB.[4,5] T-SPOT.TB on serous effusion and cerebral spinal

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fluid have a higher diagnostic value for tuberculous serositis and tuberculous meningitis.[6,7] However, the diagnostic value of T-SPOT.TB for articular TB was rarely reported, and most of the reports were about T-SPOT.TB on peripheral blood. Only one case report was about T-SPOT.TB on synovial fluid for the diagnosis of articular TB up to now.[8]

Methods

Study population

Patients with clinical suspected articular TB at our institution were consecutively recruited between August 2011 and December 2015. This study was reviewed and approved by the Institutional Review Board at our institution and waiver of consent was granted because this was a retrospective study. All the patients enrolled in the study were given T-SPOT.TB test on synovial fluid, and all patients were given T-SPOT.TB test on peripheral blood except one.

Clinical information was extracted from patients' medical recordings by two researchers, who also reviewed patients' treatment and discharge diagnosis. The diagnosis was made independent of the T-SPOT.TB results, if the two researchers have different opinions of the diagnosis, a third researcher was referred to for confirmation. All patients were given HIV test. Synovial fluid was obtained by joint cavity puncture and the following tests were performed: routine cell counting, microscopy (Gram-stain, acid-fast *Bacilli* stain), bacterial culture, *Mycobacterium tuberculosis* (MTB) culture (liquid culture method, BD MGIT960), fungal culture, and TB polymerase chain reaction (PCR) (Roche Amplicor). Heparinized samples of venous blood (4 ml) and of synovial fluid (4 ml) were obtained and processed for detecting specific T-cell responses to RD1 encoded antigens by T-SPOT.TB (Oxford Immunotec., Abingdon, UK).

Diagnosis of articular tuberculosis

Based on previous publications,[9,10] patients were classified as having confirmed TB if clinical specimens were positive for MTB on culture or by a PCR assay. Patients were classified as having probable TB if they responded to anti-TB therapy and histologic examination of biopsy samples showed caseating granulomas associated with radiographic findings consistent with osteoarticular TB. Patients were classified as having "not active TB" if another diagnosis was made or if there was a clinical improvement without anti-TB therapy. Patients were classified as having possible TB if they did not fulfill the above criteria but active TB could not be excluded.

T-SPOT.TB on synovial fluid and peripheral blood

Four ml of synovial fluid was collected from each patient and was performed within 6 h after collection by laboratory staff blinded to patients' clinical data. T-SPOT.TB utilized AIM-V (GIBCO™ AIM-V Medium Liquid, Invitrogen, USA) as a negative control, phytohaemagglutinin (PHA) as positive control, and ESAT-6 and CFP-10 as specific antigens, respectively. Synovial fluid mononuclear cells (SFMCs) were separated by Ficoll-Hypaque gradient centrifugation and plated (2.5×10^5 per well) on a plate pre-coated with an antibody against IFN-γ. After incubation 16–18 h at 37°C in 5% carbon dioxide, plate wells were washed and incubated with a conjugate against the antibody used and an enzyme substrate. Spot-forming cells (SFCs) that represented antigen-specific T-cells secreting IFN-γ were counted with an automated enzyme-linked immunospot (ELISPOT) reader (AID-iSpot, Strassberg, Germany). A positive response was defined as six or more SFCs in the target well. The background number of spots in the negative control well for SFMCs should be less than ten spots. When the cell counts in synovial fluid could not harvest 2.5×10^5 cells per well, we used the ratio between 2.5×10^5, the target number and the actual number to adjust the result. Four ml of peripheral blood was also collected from each patient except one and RD-1 ELISPOT assay protocol for peripheral blood mononuclear cells (PBMCs) was same with that for SFMCs.

Statistical analysis

Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), likelihood ratio positive (LR^+), and likelihood ratio negative (LR^-) were calculated to evaluate the diagnostic performance of T-SPOT.TB on SFMCs and PBMCs. Means were used for data of normal distribution, while median and interquartile range (IQR) were used for data that were not normally distributed. Means and medians were compared using Student's *t*-test or Wilcoxon test as appropriate. Positive proportions were compared using Pearson's Chi-square test. 95% confidence intervals (*CI*s) were estimated according to the binomial distribution. Significance was inferred for $P < 0.05$ and statistical analysis was performed by SPSS 16.0 (SPSS Inc., Chicago, IL, USA).

Results

Twenty patients with suspected articular TB were enrolled. Six patients were diagnosed with articular TB, including one patient with confirmed articular TB (positive for an MTB PCR assay), one patient with probable articular TB, and four patients with possible articular TB. Fourteen patients were diagnosed with non-TB arthritis, including three patients with common bacterial infection, two patients with synovitis, two patients with spondyloarthropathy (ankylosing spondylitis and undifferentiated spondyloarthropathy), and one each patient with undifferentiated arthritis, reactive arthritis, gout, femoral head necrosis, juvenile idiopathic arthritis, Behcet disease, and sarcoidosis [Tables 1-3]. All the six patients with articular TB had single joint involved (knee joint), among the 14 patients with non-TB arthritis, seven had single joint involved (knee and hip joint), and seven had more than one joint involved. Thirteen patients (five with TB and eight with non-TB) had acid-fast *Bacilli* stain of synovial fluid and all were negative. Nine patients (three with TB and six with non-TB) had MTB culture and all were negative. Twenty patients had T-SPOT.TB on synovial fluid, 19 patients had T-SPOT.TB on

Table 1: Demographic and clinical characteristics of the patients

Characteristics	Articular TB (n=6)	Non-TB arthritis (n=14)
Age (years), median (IQR)	41 (27–48)	47 (28–56)
Gender, *n*		
Male	2	5
Female	4	9
Duration (month), median (IQR)	17 (8–63)	12 (2–52)
HIV (+), *n*	0	0
Underlying diseases, *n*	2	4
Corticosteroids/immunosuppressant use, *n*	3	7
Lung TB, *n*	2	1
Previous history of TB, *n*	0	2
Radiography evidence of previous TB, *n*	0	1
Symptoms		
Fever, *n* (%)	4	6
Peak temperature (°C), median (IQR)	38.5 (37.6–39.5)	38.2 (37.9–39.1)
Redness of the involved joint, *n*	1	3
Swelling of the involved joint, *n*	6	13
Increased skin temperature around the involved joint	1	3
Pain of the involved joint	6	13
Movement disorder of the involved joint	5	8
Joints involved, *n*		
Knee	6	11
Ankle	0	2
Hip	0	1
Blood examination, median (IQR)		
Leukocytes ($\times10^9$/L)	7.55 (5.31–11.04)	7.61 (5.23–8.67)
Lymphocytes ($\times10^9$/L)	1.39 (1.14–1.81)	1.67 (1.05–2.39)
ESR (mm/h)	42 (33–88)	64 (11–83)
hsCRP (mg/L)	60.26 (16.80–84.89)	20.32 (3.42–45.95)

Duration: The course of the disease before definitive diagnosis was made. ESR: Erythrocyte sediment rate, normal range: male >15 mm/h, female >20 mm/h; hsCRP: Hypersensitive C-reactive protein, normal range: >3 mg/L; IQR: Interquartile range; TB: Tuberculosis.

peripheral blood at the same time. All the six patients with articular TB received anti-TB drugs; five patients also had arthroscopy operations. The median follow-up time was 2 months (IQR 2–33 months) with significant improvement in all cases.

Sensitivity and specificity of T-SPOT.TB on synovial fluid mononuclear cells and peripheral blood mononuclear cells

Among the six patients with articular TB, T-SPOT.TB on SFMCs was positive in five patients, giving a sensitivity of 83% (95% *CI*: 0.62–1.00), and T-SPOT.TB on PBMCs was positive in four patients, giving a sensitivity of 67% (95% *CI*: 0.24–0.94). T-SPOT.TB was negative on both SFMCs and PBMCs in one patient who was diagnosed as possible TB (the patients did not fulfill the criteria of confirmed and probable TB, but active TB could not be excluded) [Table 4].

Among the 14 patients with non-TB arthritis, T-SPOT.TB on SFMCs was negative in 12 patients, giving a specificity of 86% (95% *CI*: 0.56–0.97), and T-SPOT.TB on PBMCs (performed in 13 patients) was negative in nine patients, giving a specificity of 69% (95% *CI*: 0.39–0.90). Four patients with non-TB arthritis, who had a previous history of TB, Behcet disease, gout, and synovitis, respectively, had positive results with T-SPOT.TB on SFMCs and/or PBMCs.

Predictive value and likelihood ratio of T-SPOT.TB on synovial fluid mononuclear cells and peripheral blood mononuclear cells

Among the seven patients with positive T-SPOT.TB on SFMCs, five were diagnosed with articular TB, giving the PPV of 71%. Among the eight patients with positive T-SPOT.TB on PBMCs, four were diagnosed with articular TB, giving the PPV of 50%.

Among the 13 patients with negative T-SPOT.TB on SFMCs, 12 were diagnosed with non-TB arthritis, giving the NPV of 92%. Among the 11 patients with negative T-SPOT.TB on PBMCs, nine were diagnosed with non-TB arthritis, giving the NPV of 82%.

The positive likelihood ratio (PLR) and negative likelihood ratio (NLR) of T-SPOT.TB on SFMCs were 5.80 (95% *CI*: 1.54–22.10) and 0.19 (95% *CI*: 0.03–1.19), respectively. The PLR and NLR of T-SPOT.TB on PBMCs were 2.17 (95% *CI*: 0.80–5.84) and 0.48 (95% *CI*: 0.15–1.60), respectively [Table 4].

Table 2: Clinical characteristics of articular tuberculosis (n = 6)

Case	Age (years)/ gender	Underlying diseases	Previous history of TB	Clinical symptoms of joints	Duration (months)	Other sites of TB	Articular X-ray/MRI/CT	T-SPOT.TB (SFCs/10^6 MC)		Classification of TB diagnosis
								PBMC	Synovial fluid	
1	54/female	SLE	No	Swelling and pain of right knee, with movement disorder	6	Lung	Multiple cartilage thinning of right knee, hematocele in right knee articular cavity and suprapatellar bursa	120	620	Confirmed
2	29/female	No	No	Swelling and pain of right knee, with movement disorder	60	Lung	Diffused synovium lesions in right knee with multiple bone erosions, effusion of right knee	1700	2000	Probable
3	44/female	No	No	Redness, swelling, pain and increased skin temperature of right knee, with movement disorder	72	No	Diffused and irregular synovium thickening of right knee, effusion of right knee	2092	272	Possible
4	46/male	No	No	Swelling and pain of left knee, with movement disorder	10	No	Normal	0	296	Possible
5	19/female	RA	No	Swelling and pain of right knee, with movement disorder	24	No	Narrowing of right knee joint space with effusion, synovium thickening, and bone erosions	64	80	Possible
6	37/male	No	No	Swelling and pain of right knee	9	No	Effusion of right knee cavity	0	0	Possible

Case	Examination of synovial fluid						Histopathology of synovium of the involved joint	Anti-TB Treatment	Outcome
	Puncture site	Appearance	Leukocyte	Erythrocyte	Acid-fast *Bacilli* stain	Fast mycobacterium culture			
1	Right knee	Yellowish-brown and muddy	Full view	15-20/HPF	Negative	–	–	Yes	Improved
2	Right knee	–	–	–	–	–	Epithelioid cell granulomas, fibrinoid necrosis, chronic inflammation of synovium, acid-fast *Bacilli* stain negative	Yes	Improved
3	Right knee	Yellow and muddy	8-10/HPF	10-15/HPF	Negative	–	Chronic inflammation, with proliferation of fibrous tissue and hyaline degeneration, acid-fast *Bacilli* stain negative	Yes	Improved
4	Right knee	Light yellow and muddy	10-20/HPF	20-30/HPF	Negative	Negative	Acute and chronic inflammation, with proliferation of fibrous tissue and vessels, acid-fast *Bacilli* stain negative	Yes	Improved
5	Right knee	–	–	–	Negative	Negative	Chronic inflammation, with proliferation of lymphoid tissue and small vessels, acid-fast *Bacilli* stain negative	Yes	Improved
6	Right knee	–	–	–	Negative	Negative	Acute and chronic inflammation, exudation of necrosis were observed	Yes	Improved

SLE: Systemic lupus erythematosus; RA: Rheumatic arthritis; HPF: High power field; –: Not available; MC: Mononuclear cells; MRI: Magnetic resonance imaging; CT: Computed tomography; TB: Tuberculosis; SFCs: Spot-forming cells.

Compared with T-SPOT.TB on synovial fluid alone, the combination of T-SPOT.TB on synovial fluid and PBMCs appeared to have higher specificity, PPV, and LR^+ but have no advantage on sensitivity, NPV, or LR^-.

Comparison of frequencies of RD1 antigen-specific interferon-γ secreting T-cells on synovial fluid and peripheral blood in articular tuberculosis patients

The frequencies of cells responding to ESAT-6 and CFP-10 in synovial fluid for articular TB patients were not statistically different from those observed in peripheral blood.

Discussion

Our data showed that it had higher sensitivity (83%), specificity (86%), and NPV (92%) compared to commonly used tests. Mycobacterial osteomyelitis and arthritis are the third most common infection of EPTB after pleural and lymphatic TB worldwide.[11] In China, EPTB accounts

Table 3: Clinical characteristics of non-TB arthritis (n = 14)

Case	Age, years/ gender	Underlying diseases	Previous history of TB	Clinical symptoms of the joint	Duration (months)	Other sites of TB	Articular X-ray/ MRI/CT	T-SPOT.TB (SFCs/10^6 MC)		Diagnosis
								PBMC	Synovial fluid	
1	29/female	SLE	No	Pain of both hips, with movement disorder	48	No	Necrosis of femoral head	0	0	Multiple arthritis with bacterial infection
2	59/female	No	No	Pain of left knee	12	No	Narrowing of left knee joint space	0	0	Left knee arthritis with bacterial infection
3	24/female	SLE	No	Swelling of multiple joints, with movement limitations	0.3	No	Effusion of both knee cavities	0	24	Left knee arthritis with bacterial infection
4	40/female	No	No	Redness, swelling, and increased skin temperature of left knee	24	No	Effusion of left knee cavity	24	0	Synovitis of left knee
5	49/male	No	Yes	Swelling and pain of right knee, with movement disorder	12	No	Effusion of right knee cavity	332	0	Synovitis of right knee
6	13/male	No	No	Swelling and pain of both ankles, with movement disorder	24	No	Abnormal signals in bones of feet and articular capsule	/	0	AS
7	56/female	No	No	Swelling and pain of left knee, with movement disorder	108	No	Effusion of left knee cavity	0	0	USpA
8	56/female	No	No	Swelling of both knees, with movement disorder	2	No	Degeneration of both knees	0	0	Undifferentiated arthritis
9	8/female	No	No	Pain of both hips and knees	66	No	Osteoporosis of both knees	0	0	Juvenile idiopathic arthritis
10	67/male	No	No	Swelling and pain of both knees, wrists, and elbows, with movement disorder	2	No	Normal	24	0	Gout
11	38/male	No	No	Wandering pain of knees, redness and increased temperature of skin	7	No	Effusion of left knee cavity	0	0	Reactive arthritis of left knee
12	48/male	RA	Yes	Pain of right hip, with movement disorder	5	Lung	Necrosis of right femoral head	0	0	Necrosis of right femoral head
13	51/female	Cirrhosis after hepatitis B	No	Redness, pain, and increased skin temperature of left knee	84	No	Synovitis of left knee, swelling of soft tissue in suprapatellar bursa	596	42	Behcet disease
14	51/female	No	No	Swelling of left knee	2	No	Subluxation of patella in left knee	0	0	Sarcoidosis

Case	Examination of synovial fluid						Histopathology of synovium of the involved joint	Outcome
	Puncture site	Appearance	Leukocyte	Erythrocyte	Acid-fast *Bacilli* stain	Fast mycobacterium culture		
1	Left knee	/	/	/	Negative	Negative	/	Improved
2	Left knee	/	/	/	/	/	Necrosis, inflammatory exudate, acute purulent inflammation of synovium, multi nuclear giant cells were observed	Improved
3	Left knee	Yellow and muddy, with clot	Large amount	20–30/HPF	Negative	Negative	Chronic inflammation, fibrinoid exudation was observed	Improved
4	Left knee	Yellow and thick	8–14/HPF	2–5/HPF	/	Negative	Chronic inflammation, proliferation of small vessels in stroma	Improved
5	Right knee	Yellow and muddy, with clot	Large amount	5–10/HPF	/	/	Acute and chronic inflammation, partially papillary hyperplasia, swelling in stroma and proliferation of small vessels	Improved

Contd...

Table 3: Contd...

Case	Examination of synovial fluid						Histopathology of synovium of the involved joint	Outcome
	Puncture site	Appearance	Leukocyte	Erythrocyte	Acid-fast *Bacilli* stain	Fast mycobacterium culture		
6	Left ankle	/	/	/	Negative	/	Acute and chronic inflammation of synovium, acid-fast *Bacilli* stain negative	Improved
7	Left knee	Yellow and muddy	Full view	Full view	Negative	Negative	Acute and chronic inflammation, erosion on the surface of synovium	Improved
8	Right knee	Yellow and muddy	Large amount	2–5/HPF	Negative	/	/	Improved
9	Right knee	Bloody	0–4/HPF	Full view	/	/	/	Improved
10	Left knee	/	/	/	Negative	/	/	Improved
11	Left knee	Orange and muddy, with clot	Full view	Large amount	/	/	Chronic inflammation, proliferation of fiber and vessels	Improved
12	Right hip	/	/	/	/	/	Acute and chronic inflammation, with regression and calcification, inflammatory and fibrinoid exudate were observed	Improved
13	Left knee	Yellow and muddy	Full view	Large amount	Negative	Negative	/	Improved
14	Left knee	/	/	/	Negative	Negative	/	Improved

USpA: Undifferentiated spondyloarthropathy; AS: Ankylosing spondylitis; MC: Mononuclear cells; HPF: High power field; SLE: Systemic lupus erythematosus; SFCs: Spot-forming cells; MRI: Magnetic resonance imaging; CT: Computed tomography; TB: Tuberculosis; PBMC: Peripheral blood mononuclear cell; /: Not available.

Table 4: Single and combined diagnostic parameters of T-SPOT.TB on SFMCs and PBMCs

T-SPOT.TB on	95% *CI*					
	Sensitivity	Specificity	PPV	NPV	LR^+	LR^-
SFMC	0.83 (0.36–0.99)	0.86 (0.56–0.97)	0.71 (0.30–0.95)	0.92 (0.62–0.99)	5.80 (1.54–22.10)	0.19 (0.03–1.19)
PBMC	0.67 (0.24–0.94)	0.69 (0.39–0.90)	0.50 (0.15–0.83)	0.82 (0.48–0.97)	2.17 (0.80–5.84)	0.48 (0.15–1.60)
PBMC or SFMC (parallel)	0.83 (0.36–0.99)	0.62 (0.32–0.85)	0.50 (0.20–0.80)	0.89 (0.51–0.99)	2.17 (1.00–4.70)	0.27 (0.04–1.78)
PBMC and SFMC (serial)	0.67 (0.24–0.94)	0.93 (0.64–1.00)	0.80 (0.30–0.99)	0.87 (0.58–0.98)	9.33 (1.30–67.03)	0.36 (0.11–1.13)

PPV: Positive predictive value; NPV: Negative predictive value; LR^+: Likelihood ratio positive; LR^-: Likelihood ratio negative; SFMCs: Synovial fluid mononuclear cells; PBMCs: Peripheral blood mononuclear cells; TB: Tuberculosis; *CI*: Confidence interval.

for about 10–20% of all cases of active TB with 19.9% being bone and joint TB.[12] Spine, hip and sacroiliac joint, knees, ribs, and shoulder are the sites commonly involved. Consistent with this pattern, our study showed TB of the knee joint was the most often involved sites.

Given that difficulty in identifying the organism and the fact that the clinical symptoms are nonspecific with an insidious onset that can mimic other joint diseases like rheumatoid arthritis and osteoarthritis, the diagnosis of osteoarticular TB is often delayed.[13] In the previous study among Chinese patients, the mean time from symptom onset to diagnosis of bone and joint TB was 13.16 months (range from 0.5 to 96 months, median delay was 7 months).[14] In our study, the median duration from onset of articular symptoms to definite diagnosis was 17 months (IQR 8–63 months), suggesting a huge need for improvement. Traditional diagnostic methods such as acid-fast stains of the joint fluid are positive in only 20% to 25% of those examined.[15] Despite that fact that 90% to 95% of cases would achieve full recovery with a nearly full joint function if diagnosed and treated early. Therefore, it is necessary to develop a fast diagnostic method for articular TB.[3]

IGRAs became a new diagnostic method in recent years, sensitivity and specificity of T-SPOT.TB on peripheral blood varied from different studies. In our study, the sensitivity and specificity of T-SPOT.TB on peripheral blood for articular TB was 67% and 69%, respectively. In a study by Fan *et al*.,[16] 92 patients with confirmed osteoarticular TB and 64 patients without active osteoarticular TB were analyzed, the sensitivity and specificity for T-SPOT. TB assay on peripheral blood were 93.5% (86/92) and 78.1% (50/64), respectively. In another study of 28 patients with confirmed osteoarticular TB and 38 patients with non-TB arthritis,[9] the sensitivity and specificity of T-SPOT.TB on peripheral blood was 100% (95% *CI*: 0.88–1.00) and 55% (95% *CI*: 0.40–0.70), respectively. Our study showed that the PPV and NPV of T-SPOT.TB on peripheral blood for articular TB were 50% and 82%, respectively. According to Cho

et al.,[9] the PPV and NPV of T-SPOT.TB on peripheral blood for articular TB were 62% and 100%, respectively. Multivariate analysis revealed that chronic forms of EPTB were independently associated with higher sensitivity of blood T-SPOT.TB test ($P = 0.007$).

In the report about T-SPOT.TB on synovial fluid for the diagnosis of articular TB, the numbers of MTB specific T-cells, as determined by ELISPOT, were 2-fold to 6-fold higher in synovial fluid than in blood.[8] Our patients with confirmed and probable articular TB also had higher frequencies of T-cells in synovial fluid than in blood, which may due to the accumulation of MTB-specific T-cell in synovial fluid results from selectin-mediated migration and a local proliferation.[17]

Among the four patients who had positive T-SPOT.TB on SFMCs and/or PBMCs but with non-TB arthritis, one had previous TB, which may have led to latent TB. One had Behcet disease, which was reported to have some associations with TB, Pervin *et al*. showed that mapped T-cell epitopes of heat shock protein (HSP) in patients with BD by stimulating T cells with overlapping synthetic peptides derived from gene sequences of MTBHSP. MTBHSP displayed molecular mimicry to human HSP, resulting in an immunologic cross-reaction and the subsequent development of BD.[18] As China is an area with relatively high TB prevalence, many people may have latent TB infection (LTBI) that could result in positive T-SPOT.TB. The limitation of IGRAs is that it cannot differentiate active TB and LTBI. Therefore, compared to peripheral blood, the diagnostic value of T-SPOT.TB on synovial fluid without the influence of LTBI is of great importance. One patient with possible articular TB had negative T-SPOT.TB on both SFMCs and PBMCs, he had no underlying diseases and normal leukocytes and lymphocytes, the negative T-SPOT.TB result on both may be due to the failed lymphocyte compartmentalization, migration, and activation to RD1 peptides.[19]

All the six patients with articular TB took anti-TB drugs, and three patients also had operations. The mainstay treatment of articular TB is multidrug antituberculous chemotherapy (for 12–18 months) and active-assisted nonweight bearing exercises of the involved joint throughout the period of healing. Operative intervention is required when the patient is not responding after 4–5 months of chemotherapy (synovectomy and debridement), or if the therapeutic outcome is not satisfactory, such as excisional arthroplasty for the hip or the elbow. Joint replacement may be considered if the disease has remained inactive for 10 years or more.[3]

Limitations

There are several limitations of our study. First, the sample size was small and only a few patients had confirmed TB, and this was a retrospective study, which might overestimate or underestimate the diagnostic value of T-SPOT.TB on synovial fluid. However, examination of T-SPOT.TB on synovial fluid has not been previously reported and rarely performed in clinical settings. Our study was exploratory rather than confirmatory. Our data during the past 5 years indicated the potential that T-SPOT.TB on synovial fluid can improve the diagnosis of articular TB and call for a larger study to verify this finding. Second, this study was done in a single center; therefore, the results in this group of patients might not be representative for other areas.

In summary, this study showed that T-SPOT.TB on synovial fluid can potentially improve the diagnosis of articular TB. The sensitivity, specificity, and NPV appeared high. Patients may have T-SPOT.TB on synovial fluid together with clinical findings and other tests if possible to assist diagnosis of articular TB.

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Conflicts of interest

There are no conflicts of interest.

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