International Journal of Infectious Diseases 15 (2011) e594–e600

Contents lists available at ScienceDirect

International Journal of Infectious Diseases

journal homepage: www.elsevier.com/locate/ijid

Review

Adjunctive therapy with interferon-gamma for the treatment of pulmonary tuberculosis: a systematic review

X.-F. Gao [a,*], Z.-W. Yang [b], J. Li [a]

[a] Department of Preventive Medicine, North Sichuan Medical College, Nanchong, Sichuan 637007, China
[b] Morphometric Research Laboratory, North Sichuan Medical College, Nanchong, Sichuan, China

ARTICLE INFO

Article history:
Received 24 September 2010
Received in revised form 3 April 2011
Accepted 3 May 2011

Corresponding Editor: William Cameron, Ottawa, Canada.

Keywords:
Pulmonary tuberculosis
Interferon-γ
Systematic review

SUMMARY

Objective: To evaluate the efficacy and safety of adjunctive therapy using interferon-gamma (IFN-γ; an immunomodulator) for the treatment of pulmonary tuberculosis (TB).
Methods: We conducted a systematic review of controlled clinical trials that compared anti-TB drugs in combination with IFN-γ with the same anti-TB drugs alone for the treatment of pulmonary TB.
Results: Nine trials were identified, with IFN-γ being aerosolized or administered subcutaneously in one trial, aerosolized only in five trials, and administered intramuscularly in three trials. The methodology quality of all trials was rated 'C'. Meta-analysis of the trials with aerosolized IFN-γ showed statistical benefits on sputum negative conversion and chest radiograph: the pooled relative risk (RR) for conversion was 1.97 (95% confidence interval (CI) 1.20–3.24, $p = 0.008$) after 1 month of treatment, 1.74 (95% CI 1.30–2.34, $p = 0.0002$) after 2 months of treatment, 1.53 (95% CI 1.16–2.01, $p = 0.003$) after 3 months of treatment, 1.57 (95% CI 1.20–2.06, $p = 0.001$) after 6 months of treatment, and 1.55 (95% CI 1.17–2.05, $p = 0.002$) at the end of treatment; the pooled RR for the chest radiograph was 1.38 (95% CI 1.10–1.17, $p = 0.006$) at the end of treatment. For intramuscularly administered IFN-γ, meta-analysis of three trials showed its significant improvement on sputum negative conversion after 2 months of treatment. A randomized controlled trial with aerosolized and subcutaneously administered IFN-γ reported significant reductions in the symptoms of fever, wheeze, and night sweats in the IFN-γ-treated groups compared with the control group after 1 month of treatment. No patients discontinued treatment because of adverse effects caused by IFN-γ.
Conclusion: Adjuvant therapy using IFN-γ, especially by aerosol, might be beneficial to TB patients, but large randomized controlled trials are needed for further evaluation of its efficacy and safety considering the quality of the trials analyzed.

1. Introduction

Tuberculosis (TB) is a major cause of illness and death, and the worldwide epidemic of HIV/AIDS and emergence of multidrug-resistant TB (MDR-TB), or even extensively drug-resistant TB (XDR-TB), are exacerbating this global problem.[1,2] Recent global estimates showed 9.27 million new cases of TB and 1.77 million deaths from TB in 2007.[3]

The incidence and progression of TB is closely related to the host immune condition. The immune response to the tubercle bacilli mainly takes the classic form of delayed-type hypersensitivity, which is thought to be mediated by a cooperative interaction between T-lymphocytes and macrophages, depending upon the interplay of cytokines produced by a variety of mononuclear cells.[4] The progression of TB may be associated with cellular immunodeficiency.[5] Conventional anti-tubercular chemotherapy combined with immunotherapeutic modalities could therefore be a promising new approach for the treatment of TB. As indicated in a meta-analysis, the use of *Mycobacterium vaccae* as an immunomodulator is helpful for patients with recurrent pulmonary TB (PTB).[6]

Interferon-gamma (IFN-γ), made by immune cells, is a highly pleiotropic cytokine with immunomodulatory antimicrobial, antiproliferative, and antifibrotic activities that also modulates the production or activities of several cytokines and chemokines.[7–9] Early animal models and pharmacokinetic and pharmacodynamic studies suggested that adjunctive therapy with IFN-γ may favor recovery from PTB.[10–12] Some uncontrolled studies in humans also showed that the adjunctive therapy could improve TB progression and was well tolerated by MDR-TB patients.[13,14] However, there were uncontrolled studies showing that the adjunctive therapy did

* Corresponding author. Tel.: +86 0817 2242774.
E-mail address: xfgao_nsmc@163.com (X.-F. Gao).

1201-9712/$36.00 – see front matter
doi:10.1016/j.ijid.2011.05.002

not improve the sputum culture conversion of refractory or advanced MDR-TB.[15,16] Hence, adjunctive immunotherapy with IFN-γ needs to be evaluated in controlled trials. Some controlled clinical trials with adjunctive IFN-γ have recently been completed, and we therefore conducted a systematic review of the trials on the efficacy and safety of adjunctive IFN-γ therapy for TB, in order to provide timely evidence-based information for TB physicians.

2. Methods

2.1. Selection criteria

We included controlled clinical trials that met the following criteria: (1) Participants. Eligible participants were patients with drug-susceptible or drug-resistant PTB confirmed by sputum smear and/or culture. (2) Intervention. Only trials that directly compared IFN-γ plus anti-TB drugs with the same anti-TB drugs alone were included. (3) Outcome measures. The primary outcome was sputum negative conversion (sputum smear and/or *Mycobacterium tuberculosis* culture) at a specific number of months after therapy. The secondary outcomes included chest radiographic improvement and (severe) adverse events. Chest radiographic improvement was defined as a decrease in the extent of lesions in the lungs, or a >50% decrease in the cavity size at a specific number of months after treatment. Severe adverse events, including severe hepatotoxicity, were defined as serious adverse drug reactions that finally led to hospitalization of the patient or discontinuation of treatment, and were determined by the authors based on symptoms, physical signs, and/or laboratory examination.[17,18] Other outcomes included biochemical variables (e.g., serum interleukin (IL)-4, IFN-γ) reflecting immune function, and bacteriological relapse after completion of treatment. (4) Language of articles. We limited languages to English and Chinese in this study.

2.2. Search strategy and study selection

We searched published information in Medline (1966 to April 2010), the Cochrane Central Register of Controlled Trials (CENTRAL, Cochrane Library issue 1, 2010), the Chinese Biomedical Literature Database (CBM, 1978 to April 2010), and the VIP Database (1989 to May 2010), and unpublished information in the WANFANG Database (up to May 2010) and at 'ClinicalTrials.gov'. CBM, VIP, and WANFANG are widely used databases in China, which extensively index articles published in journals and conference proceedings in Chinese. We searched these databases using the search strategy of '(tuberculosis OR TB) AND (interferon OR IFN)'. We also scanned the reference lists of all relevant articles for the selection of controlled clinical trials. Figure 1 shows the process by which articles were selected.

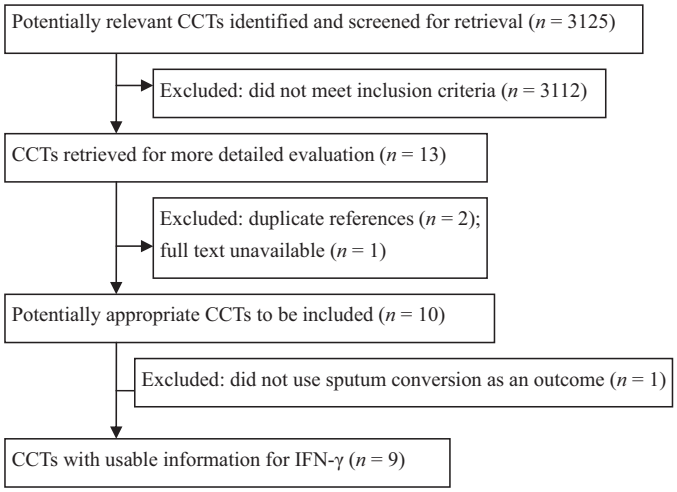

Figure 1. Results of the search for controlled clinical trials (CCTs) in the systematic review.

2.3. Review procedure

Two reviewers independently selected the trials for inclusion, assessed the quality of the trials, and extracted data in the form of a predefined table. Discrepancies were resolved through discussion or by the third party. We contacted the authors of the trials for missing information by telephone. For trials published in Chinese journals, we inquired, using pre-designed questions, about their method of allocating participants, to judge whether the trial was randomized or not. As the trial by Dawson et al.[19] compared chemotherapy combined with aerosolized or subcutaneously administered IFN-γ with chemotherapy alone, we separately analyzed the comparison between chemotherapy combined with aerosolized IFN-γ and chemotherapy alone, and the comparison between chemotherapy combined with subcutaneously administered IFN-γ and chemotherapy alone.

The methodology quality of each trial was evaluated according to the following five predefined items: randomization, allocation concealment, blinding, reporting loss to follow-up or withdrawal, and comparability of baseline. Each item was rated as A, B, or C (Table 1) in accordance with the Cochrane handbook.[20] When all five items were rated A, the trial was considered high quality (overall grade A); when one or more items were rated C, the trial

Table 1
Methodology quality of the trials included in this review

Trial	Randomization[a]	Allocation concealment[b]	Blinding[c]	Reporting loss of follow-up/withdrawal[d]	Comparability of baseline[e]	Overall grade[f]
Dawson et al., 2009[19]	A	B	C	A	A	C
Yang et al., 2009[21]	A	C	C	A	A	C
Zhang et al., 2009[22]	C[g]	C	C	C	B	C
Shi et al., 2008[23]	C	C	C	C	A	C
Li et al., 2008[24]	C	C	C	A	A	C
Wang et al., 2006[25]	C	C	C	B	A	C
Xu, 2006[26]	C	C	C	C	A	C
Liang, 2003[27]	C	C	C	A	A	C
Yao and Liu, 2003[28]	C[g]	C	C	C	A	C

[a] A = adequate, B = unclear (randomization was reported, but the method not described), C = inadequate (quasi-randomization or non-randomization).
[b] A = adequate, B = unclear (not mentioned), C = clearly inadequate concealment/not used.
[c] A = adequate, B = unclear (not mentioned), C = not used ('open-label' or 'unmasked' trials).
[d] A = adequate reporting (including numbers and causes), B = partly reported, C = not reported.
[e] A = comparable (at least including age and sex), B = unclear, C = not comparable.
[f] A = low risk of bias, B = moderate risk of bias, C = high risk of bias.
[g] Temporarily regarded as non-randomized trials due to our failure to contact the authors.

Table 2
Anti-tuberculosis drug regimens and interferon-γ therapy of the trials in this review

Trial, year (country), reference	Trial participants	Anti-TB drug regimens in control and treatment groups[a]	IFN-γ immunotherapy in the treatment group	Follow-up time (months)
Dawson et al., 2009 (South Africa)[19]	Drug-sensitive, cavitary, culture-positive PTB	2HRZE/4HR	Human recombinant IFN-γ1b: 2 MU/dose, aerosol or subcutaneous injection, three times weekly for 4 months	12
Yang et al., 2009 (China)[21]	Multidrug-resistant PTB	3SpPaZETh/9SpPaE	Human recombinant IFN-γ: 1 MU/dose, aerosol, three times weekly for 2 months	12
Zhang et al., 2009 (China)[22]	Previously treated, smear- or culture- positive PTB	2LPaThV/?	IFN-γ: 5 MU/dose, aerosol, three times weekly for 2 months	2
Shi et al., 2008 (China)[23]	Multidrug-resistant PTB	3HPEZAkhV/9EZV	Human recombinant IFN-γ: 2 MU/dose, aerosol, three times weekly for 6 months	6
Li et al., 2008 (China)[24]	Previously untreated, smear- or culture- positive PTB	2HRZE/4HR	Human recombinant IFN-γ: 1 MU/dose, intramuscular injection, three times weekly for 2 months	2
Wang et al., 2006 (China)[25]	Previously untreated, smear- or culture- positive PTB	2SHRZ/4HR(E) or 2HREZ/4HR(E)	Human recombinant IFN-γ: 1 MU/dose, intramuscular injection, three times weekly for 2 months	6
Xu, 2006 (China)[26]	Multidrug-resistant PTB	3AkPaThVL/15PaThVL	IFN-γ: 5 MU/dose, aerosol, three times weekly for 3 months	3
Liang, 2003 (China)[27]	Previously treated, smear- or culture-positive PTB	$2H_3R_3Z_3S_3E_3/6H_3R_3E_3$	IFN-γ: 1 MU/dose, intramuscular injection, three times weekly for 2 months	8
Yao and Liu, 2003 (China)[28]	Multidrug-resistant PTB	3KmPaZEO/6PaEO	Human recombinant IFN-γ: 1 MU/dose, aerosol, three times weekly for 3 months	9

TB, tuberculosis; PTB, pulmonary tuberculosis; IFN-γ, interferon-gamma; MU, million units.
[a] Numbers before the letters indicate the duration of the treatment phase in months; subscript numbers indicate the number of times the drug was taken each week; H, isoniazid; R, rifampin; Z, pyrazinamide; E, ethambutol; S, streptomycin; P, *para*-aminosalicylic acid; Akh, amikacin hydrochloride; Ak, amikacin; Sp, sparfloxacin; V, levofloxacin; Th, prothionamide; Km, kanamycin; Pa, pasiniazide; O, ofloxacin; L, rifapentine; ?, without report of drugs used in the continuation phase.

was considered low quality (overall grade C); and when one or more items were rated B and no items were rated C, the trial was considered of moderate quality (overall grade B), see Table 1.

2.4. Statistical analysis

According to the intention-to-treat principle, we performed a meta-analysis of the trials to explore the effects of IFN-γ. Treatment effects were summarized as the relative risk (RR) for binary variables and as the weighted mean difference (WMD) for numerical variables. The *Z*-test was used for the pooled effect size, and the statistical significance of difference was set at $p \leq 0.05$. We also calculated the 95% confidence interval (CI) of the pooled effect size. The heterogeneity of treatment effects among trials was detected using the Chi-square test and I^2 statistics. Statistical heterogeneity was set at $p \leq 0.1$. When statistical heterogeneity was not detected, the pooled effect was calculated using a fixed effect model; when heterogeneity was detected, we tried to find the sources of heterogeneity and then used a random effects model to calculate the pooled effect when it was still appropriate to combine trials. All analyses were performed using RevMan 4.2 software (The Cochrane Collaboration, Oxford, UK).

3. Results

3.1. Trials included

Nine trials, reported between 1998 and 2009, were identified in this study.[19,21–28] In our literature search, we temporarily excluded one relevant trial (clinical trial identifier: NCT00001407) because of our failure to obtain the data.[29] Eight of the included trials were conducted in China[21–28] and one in South Africa.[19] Three trials were conducted in patients with drug-sensitive or previously untreated, smear- or culture-positive PTB,[19,24,25] four in patients with multidrug-resistant PTB,[21,23,26,28] and two in patients with previously treated, culture-positive PTB (Table 2).[22,27] Anti-TB drug regimens varied in the trials. The IFN-γ intervention was given three times weekly, but its administration route and dose varied in the trials. One trial used aerosolized and subcutaneously administered IFN-γ at 2 million units (MU) per dose,[19] five used only aerosolized IFN-γ at 1, 2, or 5 MU per dose,[21–23,26,28] and the other three used intramuscularly administered IFN-γ at 1 MU per dose .[24,25,27] The IFN-γ interventions lasted 2–4 months in eight trials[19,21,22,24–28] and 6 months in one trial.[23] All trials were open-labeled (Table 1). According to our telephone interview, two trials[19,21] were randomized, two trials[23,25] were quasi-randomized because of the alternate allocation of TB patients, three[24,26,27] were non-randomized because of the allocation of TB patients based on patient opinion, and in two this was unclear because of our failure to contact the authors.[22,28] The latter two trials[22,28] were temporarily regarded as non-randomized trials as the trials were conducted in class 2 (county-level) hospitals where authentic randomized trials were rarely carried out.[30]

3.2. Sputum negative conversion rates

As chemotherapy combined with aerosolized or subcutaneously administered IFN-γ were simultaneously compared with chemotherapy alone in one included trial,[19] we did not calculate the total effect of the outcomes. Instead, we performed a subgroup analysis according to the IFN-γ administration route.

Of the nine trials evaluating therapy with IFN-γ, two reported sputum smear negative conversion rates after 1 month of treatment,[19,26] five after 2 months of treatment,[22,24–27] three after 3 months of treatment,[21,26,28] four after 6 months of treatment,[21,23,25,28] and four at completion of chemotherapy.[21,25,27,28]

One randomized controlled trial (RCT)[21] with aerosolized IFN-γ reported higher smear conversion rates in the IFN-γ-treated group compared with the control group after 3 or 6 months of treatment or at the completion of chemotherapy, although there were no statistically significant differences between the IFN-γ-treated and the control groups at these time-points. However, another RCT[19]

Table 3
Sputum smear negative conversion-related results of the trials included in this review

Administration route	Trial design	Sputum negative conversion	Trial	Treatment *n*/*N*	Control *n*/*N*	RR (95% CI)	*p*-Value[a]	*p*-Value[b]
Aerosol	RCT	After 1 month of treatment	Dawson et al., 2009[19]	20[c]/32	11[c]/30	1.70 (0.99–2.93)	-	0.05
		After 3 months of treatment	Yang et al., 2009[21]	18/30	13/30	1.38 (0.84–2.29)	-	0.20
		After 6 months of treatment	Yang et al., 2009[21]	21/30	15/30	1.40 (0.91–2.15)	-	0.12
		At completion of treatment	Yang et al., 2009[21]	23/30	16/30	1.44 (0.97–2.12)		0.07
	Non-RCT	After 1 month of treatment	Xu, 2006[26]	10/23	3/20	2.90 (0.92–9.09)	-	0.07
		After 2 months of treatment	Zhang et al., 2009[22]	32/35	21/35	1.52 (1.14–2.03)		
			Xu, 2006[26]	15/23	5/20	2.61 (1.15–5.90)		
			Subtotal	47/58	26/55	1.74 (1.30–2.34)	0.18	0.0002
		After 3 months of treatment	Xu, 2006[26]	19/23	10/20	1.65 (1.03–2.66)		
			Yao and Liu, 2003[28]	22/32	15/34	1.56 (1.00–2.43)		
			Subtotal	41/55	25/54	1.60 (1.15–2.21)	0.86	0.005
		After 6 months of treatment	Shi et al., 2008[23]	9/11	4/11	2.25 (0.98–5.16)		
			Yao and Liu, 2003[28]	25/32	17/34	1.56 (1.07–2.29)		
			Subtotal	34/43	21/45	1.70(1.20 – 2.41)	0.43	0.003
		At completion of treatment	Yao and Liu, 2003[28]	25/32	16/34	1.66 (1.11–2.48)	-	0.01
Intramuscular injection	Non-RCT	After 2 months of treatment	Wang et al., 2006[25]	42/63	45/73	1.08 (0.84–1.39)		
			Li et al., 2008[24]	31/40	22/40	1.41 (1.02–1.95)		
			Liang, 2003[27]	28/36	19/35	1.43 (1.01–2.03)		
			Subtotal	101/139	86/148	1.25 (1.05–1.49)	0.31	0.01
		After 6 months of treatment	Wang et al., 2006[25]	63/63	70/73	1.04 (0.99–1.09)	-	0.08
		At completion of treatment	Wang et al., 2006[25]	63/63	70/73	1.04 (0.99–1.09)		
			Liang, 2003[27]	33/36	29/35	1.11 (0.92–1.32)		
			Subtotal	96/99	99/108	1.06 (0.99–1.13)	0.37	0.07

RCT, randomized controlled trial; RR, relative risk; CI, confidence interval.
[a] *p*-Value of Chi-square test for heterogeneity.
[b] *p*-Value of *Z*-test for total effect.
[c] Estimated value.

with aerosolized IFN-γ found a significantly higher smear conversion rate in the IFN-γ-treated group compared with the control group after 1 month of treatment. Similarly, meta-analysis of four non-RCTs also showed a statistical effect on the rate after 2, 3, and 6 months of treatment and at the completion of chemotherapy (Table 3). When we pooled the data of the RCTs and non-RCTs with aerosolized IFN-γ, there was no statistical heterogeneity among the trials on this outcome after 1, 2, 3, or 6 months of treatment or at the end of treatment. The pooled RR was 1.97 (95% CI 1.20–3.24, $p = 0.008$) after 1 month of treatment, 1.74 (95% CI 1.30–2.34, $p = 0.0002$) after 2 months of treatment, 1.53 (95% CI 1.16–2.01, $p = 0.003$) after 3 months of treatment, 1.57 (95% CI 1.20–2.06, $p = 0.001$) after 6 months of treatment, and 1.55 (95% CI 1.17–2.05, $p = 0.002$) at the completion of chemotherapy. When we restricted the analysis to trials in MDR-TB patients,[21,23,28] the pooled RR was 1.57 (95% CI 1.20–2.06, $p = 0.001$) after 6 months of treatment and 1.55 (95% CI 1.17–2.05, $p = 0.002$) at the end of treatment. As shown by these pooled RRs with their 95% CIs, combined use of aerosolized IFN-γ improved sputum smear negative conversion at these time-points.

For trials with intramuscular administration of IFN-γ, meta-analysis showed a statistical effect on the sputum smear conversion after 2 months of treatment (Table 3). The only trial with subcutaneously administered IFN-γ reported no statistically

Table 4
Chest radiographic improvement-related results of the trials included in this review

Administration route	Trial design	Radiographic improvement	Trial	Treatment *n*/*N*	Control *n*/*N*	RR (95% CI)	*p*-Value[a]	*p*-Value[b]
Aerosol	RCT	At completion of treatment	Yang et al., 2009[21]	26/30	21/30	1.24 (0.94–1.63)	-	0.13
	Non-RCT	After 2 months of treatment	Zhang et al., 2009[22]	32/35	21/35	1.52 (1.14–2.03)		
			Xu, 2006[26]	13/23	5/20	2.26 (0.98–5.23)		
			Subtotal	45/58	26/55	1.67 (1.24–2.25)	0.34	0.0006
		After 3 months of treatment	Yao and Liu, 2003[28]	15/32	8/34	1.99 (0.98–4.05)	-	0.06
		After 6 months of treatment	Shi et al., 2008[23]	10/11	5/11	2.00 (1.02–3.92)		
			Yao and Liu, 2003[28]	20/32	16/34	1.33 (0.85–2.08)		
			Subtotal	30/43	21/45	1.49 (1.03–2.16)	0.32	0.03
		At completion of treatment	Yao and Liu, 2003[28]	25/32	17/34	1.56 (1.07–2.29)	-	0.02
Intramuscular injection	Non-RCT	After 2 months of treatment	Li et al., 2008[24]	38/40	36/40	1.06 (0.93–1.20)		
			Wang et al., 2006[25]	63/63	69/73	1.06 (1.00–1.12)		
			Liang, 2003[27]	25/36	16/35	1.52 (1.00–2.31)		
			Subtotal	126/139	121/148	1.10 (0.94–1.28)	0.03	0.23
		After 6 months of treatment	Wang et al., 2006[25]	63/63	72/73	1.01 (0.99–1.04)	-	0.32
		At completion of treatment	Wang et al., 2006[25]	63/63	72/73	1.01 (0.99–1.04)		
			Liang, 2003[27]	33/36	26/35	1.23 (0.99–1.54)		
			Subtotal	96/99	98/108	1.11 (0.68–1.82)	<0.00001	0.67

RCT, randomized controlled trial; RR, relative risk; CI, confidence interval.
[a] *p*-Value of Chi-square test for heterogeneity.
[b] *p*-Value of *Z*-test for total effect.

significant difference between the IFN-γ-treated group and the control group in the rate.[19]

3.3. Chest radiographic improvement

One trial[19] reported chest radiographic improvement in the cavity size of PTB, and eight trials provided the outcome in improvement rate. Five trials[22,24-27] reported chest radiographic improvement rates after 2 months of treatment, one trial[28] after 3 months of treatment, three trials[23,25,28] after 6 months of treatment and four trials[21,25,27,28] at the end of treatment.

One RCT[19] with aerosolized IFN-γ showed no statistically significant differences in the cavity size between the IFN-γ-treated and the control groups after 4 months of treatment (cavity in mm 18 ± 17 vs. 20 ± 16, $p = 0.65$). Similarly, another RCT reported no statistically significant differences in radiographic improvement rates between the groups at the completion of chemotherapy. Nonetheless, meta-analysis of four non-RCTs with aerosolized IFN-γ showed statistical improvement of chest radiograph after 2 and 6 months of treatment, and at the completion of chemotherapy (Table 4). When we pooled the data of the randomized and non-randomized trials on the rate at the completion of chemotherapy, there was no statistical heterogeneity between the trials and the pooled RR was 1.38 (95% CI 1.10–1.17, $p = 0.006$).

For trials with intramuscular administration of IFN-γ, the three non-RCTs showed no statistical effect on the rate after 2 or 6 months of treatment or at the completion of treatment (Table 4). The only RCT[19] with subcutaneously administered IFN-γ showed no statistically significant differences in the cavity size between the IFN-γ-treated and the control groups after 4 months of treatment (cavity size in mm 29 ± 24 vs. 20 ± 16, $p = 0.10$).

3.4. Biochemical variables reflecting immune function

One RCT with aerosolized IFN-γ reported CD4 T cells counts and CD4/CD8 ratios, which were higher in the IFN-γ-treated group after 3 months of treatment compared with the control group.[21] The other RCT with aerosolized or subcutaneously administered IFN-γ, instead of providing concrete data, reported levels of inflammatory cytokines in 24-h bronchoalveolar lavage supernatants with figures and found a significant decline in the levels of these cytokines from baseline to 4 months only in the aerosolized IFN-γ group.[19]

Two of three trials with intramuscularly administered IFN-γ measured serum IFN-γ and IL-4 levels after 2 months of treatment,[24,25] with no significant heterogeneity being detected in serum IFN-γ levels between the trials: the pooled WMD was 0.04 (95% CI 0.01–0.08; $p = 0.01$). Significant heterogeneity between trials was detected in serum IL-4 levels: the pooled WMD was −0.03 (95% CI −0.12–0.06; $p = 0.53$).

3.5. Adverse effects

Four of six trials with aerosolized IFN-γ provided data on the total adverse effects.[21–23,26] There was no statistically significant heterogeneity among trials ($I^2 = 0\%$, $p = 0.65$), and the pooled RR was 0.89 (95% CI 0.59–1.35, $p = 0.59$) (Table 5). With respect to severe adverse effects, one RCT[21] with aerosolized IFN-γ reported that only one patient in the IFN-γ-treated group stopped treatment because of a severe photosensitivity reaction caused by sparfloxacin. Another RCT[19] with aerosolized or subcutaneously administered IFN-γ reported that five patients did not complete the trial due to serious adverse events, including one patient in the aerosolized IFN-γ group, three patients in the subcutaneously administered IFN-γ group, and one patient in the control group. Nonetheless, these severe adverse events were not thought to be related to the IFN-γ and/or anti-TB regimens. The other seven trials all reported no discontinuation of treatment because of severe adverse effects. No deaths associated with IFN-γ and/or anti-TB regimens were reported in any trial.

3.6. Bacteriological relapse

Data on bacteriological relapse were not available for eight trials because the trials were not followed-up after completion of chemotherapy. The remaining trial, an RCT,[19] reported no relapse within 6 months after treatment completion. Besides the above-mentioned outcome, the RCT reported patient respiratory symptoms and found a significant reduction in the prevalence of fever, wheeze, and night sweats in the aerosolized and subcutaneously administered IFN-γ groups compared with the control group after 1 month of treatment.

4. Discussion

This systematic review identified controlled clinical trials with adjunctive IFN-γ therapy for PTB, and a meta-analysis was performed according to the route of administration of IFN-γ. All trials included in this study did not have remarkably large sample sizes, which made it difficult to find small- to moderate-sized effects of IFN-γ from a single trial; meta-analysis as used in this study could make up for this disadvantage to some degree, since the analysis is a method of obtaining a sufficient sample size to conclusively detect a small treatment effect by combining trials, even when no single trial has an adequate sample size. Our meta-analysis demonstrated that aerosolized IFN-γ (three times weekly) had statistical benefits on sputum negative conversion and chest radiographic improvement, and that intramuscular administration of IFN-γ had a statistical effect on sputum negative conversion. The findings suggest that IFN-γ, especially by aerosol, could be beneficial in PTB.

This is the first systematic review of controlled clinical trials with IFN-γ for TB. A recent published study, by reviewing early experimental studies and uncontrolled studies of IFN-γ, established that IFN-γ plays an essential role in acquired protective immunity against pathogenic mycobacteria and other intracellular pathogens.[31] Our systematic review of nine available controlled clinical trials, showed a positive effect of IFN-γ on TB from the clinical point of view, and a more precise estimate of the

Table 5
Adverse effects-related results of the trials with aerosolized interferon-gamma

Trial design	Trial	Treatment *n*/*N*	Control *n*/*N*	RR (95% CI)	*p*-Value[a]	*p*-Value[b]
RCT	Yang et al., 2009[21]	8/30	7/30	1.14 (0.47–2.75)		
Non-RCT	Zhang et al., 2009[22]	10/35	15/35	0.67 (0.35–1.28)		
	Shi et al., 2008[23]	7/11	6/11	1.17 (0.58–2.35)		
	Xu, 2006[26]	2/23	2/20	0.87 (0.13–5.62)		
	Total	27/99	30/96	0.89 (0.59–1.35)	0.65	0.59

RCT, randomized controlled trial; RR, relative risk; CI, confidence interval.
[a] *p*-Value of Chi-square test for heterogeneity.
[b] *p*-Value of *Z*-test for total effect.

magnitude of the effect, and found a statistical effect on chest radiograph only in patients given aerosolized IFN-γ, but not for intramuscularly or subcutaneously administered IFN-γ. Interestingly, in a randomized controlled trial with aerosolized IFN-α – a type I interferon – given for 2 months as an adjuvant in 20 drug-sensitive TB patients, Giosuè et al. also found improvements in the high-resolution computed tomography scores only in the IFN-α-treated group.[32] In contrast with the negative effects of intramuscularly administered IFN-γ on chest radiograph, a pilot study of 6 months of intramuscular administration of IFN-γ as adjuvant at 1 MU per dose to standard chemotherapy in eight MDR-TB patients, showed sputum conversion within 3 months and marked improvement of chest radiograph.[13] However, this was not a controlled trial, and the dosage and duration of IFN-γ given in the trial were different from those given in the trials included in our study. Intramuscular administration of IFN-γ was given at 1 MU per dose three times weekly for 2 months in the included trials. No trials directly compared the effects of aerosolized and intramuscularly administered IFN-γ on clinical outcomes. One trial directly compared aerosolized and subcutaneously administered IFN-γ in PTB patients, finding that only aerosolized IFN-γ, not subcutaneously administered IFN-γ, was effective in clearing the sputum of *M. tuberculosis* and reducing the spontaneous release of inflammatory cytokines in 24-h bronchoalveolar lavage supernatants.[19]

It is worth noting that in patients with MDR-TB, the adjunctive use of aerosolized IFN-γ appeared to improve sputum negative conversion during the whole course of treatment, and chest radiographs after 2 or 6 months of treatment. These findings were similar to the results in one uncontrolled study, which, with aerosolized IFN-γ as adjuvant at 5 MU three times weekly for 1 month in five pulmonary MDR-TB patients, showed sputum negative conversion and cavitary lesion reduction in all five patients.[14] A similar study by Koh et al.,[16] with aerosolized IFN-γ as adjuvant at 2 MU three times weekly in six refractory pulmonary MDR-TB patients for 6 months, reported radiological improvement in five patients. Considering that the number of MDR-TB patients continues to increase worldwide and that only about 60% of patients are disease-free,[33] adjunctive therapy using IFN-γ via aerosol might be a beneficial choice for the early treatment of MDR-TB.

There are several limitations with regard to the results obtained in this study. The quality of the trials included in the study was low; this could lead to bias. The trials were not blinded, for example, which might have influenced the original evaluation of the outcomes. Both randomized and non-randomized trials were included in the study. Non-RCTs compared with RCTs may tend to exaggerate treatment effects.[34,35] Therefore we separately reviewed the results of RCTs and non-RCTs; statistical heterogeneity, which might result from the difference in the allocation methods for pooling the data of RCTs and non-RCTs, was not found in any outcome. Similarly, statistical heterogeneity in the doses of aerosolized IFN-γ was not found either. The optimal dose of IFN-γ via aerosol for TB remains uncertain. In patients with MDR-TB, aerosol of 5 MU showed advantages in sputum conversion, radiograph, and body weight.[14] In healthy volunteers, IFN-γ was inhaled in single doses of up to 20 MU and multiple doses of 5 MU. Adverse events were limited to uncommon transient cough following administration; also, one subject had a transient decrease in white blood cell count.[36–38] In patients with mild to moderate cystic fibrosis lung disease, higher rates of adverse events were seen in patients receiving higher doses of aerosolized IFN-γ.[39] In our study, the dose range of 1 MU to 5 MU for aerosolized IFN-γ was well tolerated by the TB patients. In addition, only one trial provided information on relapse, which is not enough to assess the long-term benefits of IFN-γ. Therefore, additional studies are required to carefully consider the dose, long-term benefits, and length of treatment of aerosolized IFN-γ in the future.

In conclusion, our systematic review suggests that adjunctive therapy with IFN-γ, especially by aerosol, might be useful and acceptable for the treatment of PTB. Due to the limited number of RCTs identified in this study, large RCTs need to be conducted and the systematic review needs to be renewed in the future to further evaluate the efficacy and safety of this adjunctive therapy.

Acknowledgements

This study was carried out with support of a grant (09ZA034) from the Education Department of Sichuan Province, China. The sponsor was not involved in the research procedure or in the writing of the manuscript.

Conflict of interest: None.

References

1. Centers for Disease Control Prevention. Emergence of *Mycobacterium tuberculosis* with extensive resistance to second-line drugs—worldwide, 2000–2004. *MMWR Morb Mortal Wkly Rep* 2006;**55**:301–5.
2. Shah NS, Pratt R, Armstrong L, Robison V, Castro KG, Cegielski JP. Extensively drug-resistant tuberculosis in the United States, 1993–2007. *JAMA* 2008;**300**:2153–60.
3. World Health Organization. WHO report 2009. Global tuberculosis control: epidemiology, strategy, financing. WHO/HTM/TB/2009.411.Geneva, Switzerland: WHO; 2009. Available at: http://www.who.int/tb/publications/global_report/2009/en/index.html (accessed).
4. Edwards D, Kirkpatrick CH. The immunology of mycobacterial diseases. *Am Rev Respir Dis* 1986;**134**:1062–71.
5. Palma Beltrán O, Eiguchi K, Kantor IN, Eisele E, Barrera L, Perez Escalante L. Immunological evaluation of patients with pulmonary tuberculosis in Buenos Aires (abstract). *Am Rev Respir Dis* 1990;**141**:A799.
6. Fan M, Chen X, Wang K, Mao H, Feng Y, Tang X, et al. Adjuvant effect of *Mycobacterium vaccae* on treatment of recurrent treated pulmonary tuberculosis: a meta-analysis. *Chinese J Evid-based Med* 2007;**7**:449–55.
7. Schroder K, Hertzog PJ, Ravasi T, Hume DA. Interferon-gamma: an overview of signals, mechanisms, and functions. *J Leukoc Biol* 2004;**75**:163–89.
8. Farrar MA, Schreiber RD. The molecular cell biology of interferon-gamma and its receptor. *Annu Rev Immunol* 1993;**11**:571–611.
9. Boehm U, Klamp T, Groot M, Howard JC. Cellular responses to interferon-γ. *Annu Rev Immunol* 1997;**15**:749–95.
10. Cooper AM, Dalton DK, Stewart TA, Griffin JP, Russell DG, Orme IM. Disseminated tuberculosis in interferon-gamma gene-disrupted mice. *J Exp Med* 1993;**178**:2243–7.
11. Flynn JL, Chan J, Triebold KJ, Dalton DK, Stewart TA, Bloom BR. An essential role for interferon-gamma in resistance to *Mycobacterium tuberculosis* infection. *J Exp Med* 1993;**178**:2249–54.
12. Khor M, Lowrie DB, Coates AR, Mitchison DA. Recombinant interferon-gamma and chemotherapy with isoniazid and rifampicin in experimental tuberculosis. *Br J Exp Pathol* 1986;**67**:587–96.
13. Suárez-Méndez R, García-García I, Fernández-Olivera N, Valdés-Quintana M, Milanés-Virelles MT, Carbonell D, et al. Adjuvant interferon gamma in patients with drug-resistant pulmonary tuberculosis: a pilot study. *BMC Infect Dis* 2004;**4**:44.
14. Condos R, Rom WN, Schluger NW. Treatment of multidrug-resistant pulmonary tuberculosis with interferon-gamma via aerosol. *Lancet* 1997;**349**:1513–5.
15. Park SK, Cho S, Lee IH, Jeon DS, Hong SH, Smego Jr RA, et al. Subcutaneously administered interferon-gamma for the treatment of multidrug-resistant pulmonary tuberculosis. *Int J Infect Dis* 2007;**11**:434–40.
16. Koh WJ, Kwon OJ, Suh GY, Chung MP, Kim H, Lee NY, et al. Six-month therapy with aerosolized interferon-γ for refractory multidrug-resistant pulmonary tuberculosis. *J Korean Med Sci* 2004;**19**:167–71.
17. Gao XF, Li J, Yang ZW, Li YP. Rifapentine vs. rifampicin for the treatment of pulmonary tuberculosis: a systematic review. *Int J Tuberc Lung Dis* 2009;**13**:810–9.
18. Ena J, Valls V. Short-course therapy with rifampin plus isoniazid, compared with standard therapy with isoniazid, for latent tuberculosis infection: a meta-analysis. *Clin Infect Dis* 2005;**40**:670–6.
19. Dawson R, Condos R, Tse D, Huie ML, Ress S, Tseng C, et al. Immunomodulation with recombinant interferon-γ 1b in pulmonary tuberculosis. *PLoS One* 2009;**4**:e6984. doi: 10.1371/journal.pone.006984.
20. Alderson P, Green S, Higgins JP. Cochrane reviewers' handbook 4.2.2. March 2004 update. The Cochrane Library. Oxford, UK: The Cochrane Collaboration; 2004.
21. Yang Z, Wang L, Guo L, Lin M, Chen X. A short-term efficacy on aerosolizing inhalation recombinant human gamma interferon in multidrug-resistant pulmonary tuberculosis. *J Chinese Anti-tuberc Assoc* 2009;**31**:660–3.

22. Zhang YQ, He CW, Li HQ, Zhao HL, Li BJ. Effects of aerosolized interferon-γ in previously treated patients with smear-positive pulmonary tuberculosis. *Medical Recapitulate* 2009;**15**:306–7.
23. Shi JW, Yuan Y, Gu HY. Effects of aerosolized interferon-γ in 11 patients with multidrug-resistant pulmonary tuberculosis. *J Clin Pulmonary Med* 2008;**13**:1653–4.
24. Li D, Du DB, Qiu SQ, Meng QH, Luo SZ, Yuan RJ. The early effectiveness and cellular immune function of interferon-γ combined with *Mycobacterium vaccae* for previously untreated pulmonary tuberculosis. *J Chinese Anti-tuberc Assoc* 2008;**30**:460–1.
25. Wang L, Wang X, Cheng Q, Cai Y, Xiao H. The effect of the interferon-γ injected by intramuscular combined with regular anti-tuberculosis regimen used in pulmonary tuberculosis. *J Chinese Anti-tuberc Assoc* 2006;**28**: 77–9.
26. Xu JY. Effects of interferon-γ in the adjunctive treatment of multidrug-resistant tuberculosis. *Chinese J Clin Med Res* 2006;**152**:34.
27. Liang WC. Effect of interferon-γ for the treatment of previously treated and smear-positive pulmonary tuberculosis. *Chinese J Clin Med Res* 2003;**101**: .3–5.
28. Yao YX, Liu CY. Effects of interferon for the treatment of multidrug-resistant tuberculosis. *J Chinese Anti-tuberc Assoc* 2003;**25**:43–4.
29. National Institutes of Health Clinical Center. Interferon Gamma for Drug Resistant Tuberculosis. http://www.clinicaltrial.gov/ct2/show/NCT00001407?term=NCT00001407. March 3, 2008.
30. Wu T, Li Y, Bian Z, Liu G, Moher D. Randomized trials published in some Chinese journals: how many are randomized? *Trials* 2009;**10**:46.
31. Reljic R. IFN-gamma therapy of tuberculosis and related infections. *J Interferon Cytokine Res* 2007;**27**:353–64.
32. Giosuè S, Casarini M, Alemanno L, Galluccio G, Mattia P, Pedicelli G, et al. Effects of aerosolized interferon-alpha in patients with pulmonary tuberculosis. *Am J Respir Crit Care Med* 1998;**158**:1156–62.
33. World Health Organization. Multidrug and extensively drug-resistant TB (M/XDR-TB): 2010 global report on surveillance and response. WHO/HTM/TB/2010.3. Geneva: WHO; 2010. Available at: http://whqlibdoc.who.int/publications/2010/9789241599191_eng.pdf (accessed).
34. Moher D, Pham B, Jones A, Cook DJ, Jadad AR, Moher M, et al. Does quality of reports of randomised trials affect estimates of intervention efficacy reported in meta-analyses? *Lancet* 1998;**352**:609–13.
35. Schulz KF, Chalmers I, Hayes RJ, Altman DG. Empirical evidence of bias. Dimensions of methodological quality associated with estimates of treatment effects in controlled trials. *JAMA* 1995;**273**:408–12.
36. Jaffe HA, Buhl R, Mastrangeli A, Holroyd KJ, Saltini C, Czerski D, et al. Organ specific cytokine therapy. Local activation of mononuclear phagocytes by delivery of an aerosol of recombinant interferon-γ to the human lung. *J Clin Invest* 1991;**88**:297–302.
37. Martin RJ, Boguniewicz M, Henson JE, Celniker AC, Williams M, Giorno RC, et al. The effects of inhaled interferon gamma in normal human airways. *Am Rev Respir Dis* 1993;**148**:1677–82.
38. Virgolini I, Kurtaran A, Leimer M, Smith-Jones P, Agis H, Angelberger P, et al. Inhalation scintigraphy with iodine-123-labeled interferon gamma-1b: pulmonary deposition and dose escalation study in healthy volunteers. *J Nucl Med* 1997;**38**:1475–81.
39. Moss RB, Mayer-Hamblett N, Wagener J, Daines C, Hale K, Ahrens R, et al. Randomized, double-blind, placebo-controlled, dose-escalating study of aerosolized interferon gamma-1b in patients with mild to moderate cystic fibrosis lung disease. *Pediatr Pulmonol* 2005;**39**:209–18.