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The Efficacy of Interferon-α in the Treatment of Multidrug Resistant Tuberculosis

Davood Mansoori [1], Sasan Tavana [2], Mehdi Mirsaeidi [1], Mojgan Yazdanpanah [1],Hamid Sohrabpour [2]

[1]Department of Infectious Diseases, NRITLD, [2]Department of Pulmonary Medicine, Labaffi Nejad Hospital, Shaheed Beheshti University of Medical Sciences and Health Services, TEHRAN-IRAN

ABSTRACT

Background: *Tuberculosis is a major cause of infectious disease mortality all over the world. Multidrug resistant tuberculosis (MDR-TB) is a major problem in the management of tuberculosis. With recent advances in understanding the immunopathogenesis of tuberculosis, the use of various cytokine therapies has been suggested. The objective of this study was to evaluate the efficacy of parenteral INF– for treating MDR-TB patients.*

Materials and Methods: *To conduct the study, 12 MDR- TB patients hospitalised in the clinical mycobacteriology ward of Massih Daneshvari hospital were selected randomly between October 2000 and March 2001. All had chest involvement in radiography, so they were smear and culture positive on two occasions. All had at least resistance to isoniazid and rifampin in antibiogram. They were divided in two groups. One group received INF- ; (3,000,000U, three times a week, subcutaneously) in addition to anti-TB drugs, and the other group received only anti -TB medications as control group.*

Results: *Results indicate that the mean (±SD) degree of sputum smear positivity at the beginning of therapy was 2.4±0.89 in the case group and 2±0.89 in the control group which showed no significant difference (p= 0.132). Also, at the beginning of our study, there was no significant difference in the degree of sputum culture positivity between the two groups. At the end of the 8th week, all cases became smear and culture negative, but all control subjects remained smear and culture positive (p= 0.017). At the end of the 6th month; however, only two cases remained smear negative , one remained culture negative and the rest became positive. All control subjects had positive culture results (p= 0.693).*

Conclusion: *We conclude that cytokines have at least temporary effect on disease remission and can be used as adjunctive therapy.* ***(Tanaffos 2002;1(3):29-34)***

Key words*: Multidrug-resistant tuberculosis, Anti-tuberculosis drugs, Interferon-*

INTRODUCTION

Tuberculosis is a major cause of infectious disease mortality around the world (l).

Multidrug resistant tuberculosis (MDR- TB) , at the same time, is important as a major obstacle to control the infection. Multidrug resistant bacillus is defined as a bacillus resistant to at least two drugs; many experts believe that resistance to isoniazid and rifampin is necessary for the definition to be completed (2).

Patients' non-compliance with proper and regular usage of drugs, as well as the lack of knowledge, precision and interest of physicians for treating

Correspondence to: Mirsaeidi M
Telefax: +98-21-2296362
E-mail address: mmirsaeidi@nritld.ac.ir

tuberculosis are the most important factors that increase the incidence of multidrug resistance TB.

Other factors to be mentioned are single drug regimens, inadequate anti-TB medication in the primary regimen, not using the proper drug dose, inconsistent usage or quitting of the drug, and finally insufficient absorption of the drugs (3); all these factors increase antibiotic resistance of tubercle bacilli.

The degree of resistance varies in different countries. In our country, the prevalence of resistance is rising. In a study conducted by Mansoori et al. 27 patients were evaluated during one-year period who had an average period of 3.4 times anti-TB treatment which were mainly incomplete, showing inadequate response due to patients' non-compliance (4).

With increasing advances in mycobacteriology, the immunopathogenesis of the disease is being much more elucidated with time. All the clinical manifestations of tuberculosis is attributable to cellular immune response to tubercle bacilli which manifests as monocyte, macrophage, lymphocyte and polymorphonuclear leukocyte (PMNs) infiltration in the lesion.

The immune response begins after sensitization of lymphocytes with mycobacterium tuberculosis antigens and the release of various cytokines. These cytokines act as messengers of the immune system and play a fundamental role in the regulation of immune response. IFN-α is one of these cytokines, which is secreted by macrophages primarily in response to bacterial and viral presentation (5).

The usage of cytokines as stimulators of the immune system is a new method in treating intracellular infections. Different treatment results have been observed with IL-2, IFN-γ, IFN– α, IL - 12, GM- CSF, and other agents being used in various studies on the subject (6).

Current treatment modalities, for MDR-TB; however, consist of second line anti- TB medications and surgery, having many complications without the desired effect (7).

Cytokine therapy has not yet found its specific ground in treating MDR-TB patients. In this study, we examine the effect of parenteral IFN-α in MDR-TB patients who have had a history of unresponsiveness to adequate treatment with various anti-TB regimens in order to evaluate the efficacy of this cytokine in treating MDR-TB.

MATERIALS AND METHODS

This study was conducted in a period of time between November 2000 and March 2001, on 12 MDR-TB patients in Massih Daneshvari clinical mycobacteriology ward. The subjects were randomly selected and the inclusion criteria were as follows: Age>14, negative serology for HIV, absence of other secondary immunocompromise states such as malignancy, diabetes, inflammatory connective tissue disorders, cirrhosis, renal insufficiency, and lack of steroidal and immunosuppressive drugs usage. All the subjects had clinical and radiological manifestations of pulmonary TB and had at least two positive sputum smears and a positive sputum culture with at least resistance to isoniazid and rifampin.

They were all treated specifically for MDR-TB at least for six months with standard regimens of our centre with standard doses, and remained smear and culture positive. The drug regimens consisted of at least five drugs including quinolones. The subjects were divided randomly into two groups of six subjects.

Control subjects received standard anti-mycobacterial treatment, and cases received $3x10^6$ Us of interferon A (α_2b), manufactured by Schering Plough company in France, three times a week as subcutaneous injections for 8 weeks (total dose of $72xl0^6$ Us.) in addition to anti-mycobacterial regimen. Response to therapy was evaluated according to sputum and culture colony results (with available standards) every other week in the first 8 weeks and once 6 months after the initiation of the study. The side effects of the drug were recorded and in the case of serious complications the patient was

excluded. Results were analyzed by using chi-square test.

RESULTS

One subject from the case group was excluded due to severe skin necrosis associated with injection; hence, the study ended up with 11 patients. The mean age (±SD) of the case group was 44.4±17.9 years (range, 22-59 years), and the mean age (±SD) for the control group was 43.5±15 years (range,17-63 years). The difference was not statistically significant (p = 0.86). All subjects were male due to the lack of MDR female patients at the time of study. The mean time (±SD) elapsed between the presence of clinical symptoms and the beginning of the study was 3±1.41years in the case group, and 1.36 ±0. 95 years in the control group. Considering drug side effects, one subject had severe skin necrosis and four subjects had symptoms of mild arthralgia and myalgia; furthermore, there were flu-like symptoms among all subjects receiving IFN–α. Usual side effects of anti-TB medications were totally absent.

At the beginning of the study the mean degree (±SD) of sputum smear positivity was 2.4±0.89 in the case group and 2±0.89 in the control group (p=0.32) (Table1).

Table 1. Frequencies of the degree of sputum smear positivity in the study of the efficacy of INF-α in multiple drug resistant TB patients

The degree of sputum smear positivity	Case	Control	Total
	freq. (%)	freq. (%)	freq. (%)
Negative	0 (0)	0 (0)	0 (0)
+ 1	1 (20)	2 (33)	3 (27)
+ 2	1 (20)	2 (33)	3 (27)
+ 3	3 (60)	2 (34)	5 (46)
+ 4	-	-	-

The mean degree (±SD) of sputum culture positivity at the beginning of the study was 2.61 ±1.4 and 2.3±1.21,in the case and the control group, respectively. There was no significant difference between two groups (p= 0.387) (Table 2).

Table 2. Frequencies of the degree of sputum culture positivity in the study of the efficacy of INF-α in multiple drug resistant TB patients

The degree of sputum culture positivity	Case	Control	Total
	freq. (%)	freq. (%)	freq.(%)
<+1	1 (20)	-	1 (9)
+1	-	2 (33)	2 (18)
+2	-	1 (17)	1 (9)
+3	3 (60)	2 (33)	5 (46)
+ 4	1 (20)	1 (17)	2 (18)

After 8 weeks, all five subjects of the case group became sputum smear negative; the control group remained smear positive and the difference was significant (p = 0.012). Evaluation of smear results after 6 months showed two smear negative subjects in the case group; however, in the control group, all were smear positive (p = 0.132) which was not significantly different (Table3).

Table 3. Frequencies of the degree of sputum smear positivity at the end of 8th week and 6th month

The degree of sputum smear positivity	8th week		6th month	
	case	Control	case	control
	freq. (%)	freq. (%)	freq. (%)	freq. (%)
Negative	5 (100)	0 (0)	2 (40)	-
+ 1	-	3 (50)	1 (20)	-
+ 2	-	2 (33)	2 (40)	4 (66)
+ 3	-	1 (17)	-	2 (44)
+ 4	-	-	-	-

Considering the degree of sputum culture positivity at the end of 8th week, all cases were culture negative and all 6 control subjects had positive cultures (p= 0.017).

After 6 months; however, only one subject from the

case group (20%) remained sputum culture negative and the rest had positive results. All control subjects had positive results, which showed no significant difference (p = 0.0693)(Table 4).

Table 4. Frequencies of the degree of sputum culture positivity in the 8th week and in the 6th month

The degree of sputum culture positivity	8th week		6th month	
	Case freq.(%)	Control freq. (%)	case Freq. (%)	control freq. (%)
Negative	6 (100)	-	1 (20)	-
+ 1	-	1 (16)	1 (20)	1 (16)
+ 2	-	3 (50)	1 (20)	2 (34)
+ 3	-	1 (17)	2 (40)	3 (50)
+ 4	-	1 (17)	-	-

DISCUSSION

This is the first published study in Iran, regarding the therapeutic effects of IFN-α as an adjunctive treatment of MDR-TB. IFN-α is secreted by fibroblasts and leukocytes, and it also stimulates thyrosin kinase enzymes activity such as Tyk2, Jakl, which in turn cause the assembly of the IFN-α, and stimulated gene factors3(ISGF3) complex. This complex includes STAT-1, STAT- 2, and Pr-P48 (4). IFN-α also enhances the expression of P2 portion of IL-12 receptor, which results in T-lymphocyte hyperesponsiveness to IL-2. IFN-α stimulates Th_l lymphocytes to secrete cytokines and increases NK lymphocyte response to cytokines that shift Th_0 toward Th_l. It appears that IFN-α either directly enhances the death of mycobacteria or poses its antimycobacterial effect by regulation of IFN-γ or IL-12 production (8).

A study by Giosues et al. in which IFN-α was used for treating drug sensitive pulmonary tuberculosis, has proved the efficacy of the inhalant form of IFN-α ($3x10^6$ units three times a week for 8 weeks) for treatment in conjunction with anti-TB drugs. In this study patients were divided into two groups and the control group received only anti-TB medications, while the case group received inhalant IFN-α along with anti-TB medications. In the case group, improvement of clinical and radiological symptoms, conversion of sputum smears, and decreased number of cells and inflammatory cytokines in broncho-alveolar lavage fluid were much more prominent (9).

Palmero et al. treated MDR-TB patients with subcutaneous IFN-α (3×10^6 units three times a week for 12 weeks)(10). Of 5 subjects being studied, 2 cases had converted smears and cultures, and one became smear negative but remained culture positive. The remaining two case's sputum smear and culture remained positive after 30 months of follow-up. In this study, which is the second experiment regarding the efficacy of parenteral IFN-α, the results are similar to Palmero's experiment. Although the duration of injection is 4 weeks less than what conducted by Palmero, the level of efficacy is considerable. In our study 20% of patients (one case) had negative smear and culture results at the end of the 6th month, which continued to be negative 15 months after the beginning of study.

In different studies elsewhere, other biologic agents such as IFN - γ, IL-12, GM- CSF and IL-2 have also been used for treating MDR-TB. In one study the efficacy of subcutaneous IL-2 in treating MDR-TB was estimated to be approximately 60% based on lowered number of bacilli on smear, and smear conversion (11). Inhalant IFN-γ (500 μg three times weekly for 4 weeks) administered to five MDR-TB patients resulted in negative smear results after one month; however, all the patient's culture remained positive, and they became smear positive again after cessation of IFN-α treatment (7).

This problem was present in both Palmero's study and ours. Of all cases who had smear and culture conversion due to treatment with IFN- α, only two and one had a negative smear and culture, respectively at the end of the sixth month. It appears that this problem is true for other cytokines too.

Bermudez et al. showed that both IFN-α and IFN- γ

increased the concentration of macrolides especially azithromycin in macrophages, which might justify the effectiveness of regimens containing these cytokines (12). In our study; however, concomitant use of macrolides with IFN-α was not experimented, and we can not relate this phenomenon to the efficacy of IFN-α.

In general, experiments on treatment modalities with cytokines for mycobacterial infections, especially tuberculosis is rising, although it is not fully developed.

Nowadays, it is difficult to draw a definite conclusion regarding the usage of cytokines based on limited studies on the subject. It may be stated that cytokines have a special role as an adjuvant therapy in MDR-TB patients, although more expanded experiments and follow-up is needed to determine the dosage and duration of treatment. Considering the cost and desirable primary efficacy, IFN-α appears to have a better effect as an adjuvant in treating MDR-TB patients.

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