Efficacy and safety of recombinant human interleukin -2 in the adjuvant treatment of relapsing infiltrative pulmonary tuberculosis

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Abstract: Objective: To study the efficacy and safety of recombinant human interleukin-2 in the adjuvant treatment of relapsing infiltrative pulmonary tuberculosis. **Methods:** 198patients with reactivation tuberculosis were randomly divided into the test group and the control group. The dosing regimen was anti-tubercular agents and recombinant human interleukin-2 in the test group, and only the anti-tubercular agents in the control group. Conventional dose of anti-tubercular agents according to the clinical application of recombinant human interleukin-2 is 200 000 units. **Results:** The rate of sputum negative conversion and rate of focus absorption in the test group was higher than that in the control group ($P<0.05$). Although the rate of cavity closure in the test group is higher than the control group. Compared with the two groups the differences were not statistically significant ($P>0.05$). The ratio of CD4 and CD4/CD8 in the test group increased significantly at the third, and eighth months after the treatment, and it had the statistical significance compared with the control group ($P<0.05$). The level of soluble interleukin-2 receptors in the test group decreased significantly, and it had significantly differences compared with the control group ($P<0.05$). No serious adverse events were reported in the two groups. **Conclusions:** Recombinant human interleukin-2 is safe and effective as adjuvant treatment of relapsing infiltrative pulmonary tuberculosis**.**
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Pulmonary tuberculosis is a kind of infectious disease through respiratory tract, it has a serious effect on the daily live and health of human. This disease can cause the immunity of patients decreased. In recent years, there has been a high resistance situation, especially the re-treatment and severe pulmonary tuberculosis [1-2]. The re-treatment refers to the patients who received chemotherapy in the past period or received chemotherapy more than once and would receive once again, including the patients of unsuccessful initial treatment; the patients with sputum and sputum positive conversion after the full course of treatment in regular; the patients with irregular chemotherapy more than 1month; the patients with chronic bacteria row, and so on. At that moment, the patients have been formed into drug-resistant tuberculosis in vivo, so the effect of re-treatment is less apparent than the initial treatment [3-4]. Therefore, it is imperative to seek for the effective treatment of pulmonary tuberculosis drugs and methods. Study shows that adding immunomodulator may enhance the efficacy of re-treatment tuberculosis on the base of the traditional chemotherapy [5-6]. Recombinant interleukin-2 is the immunomodulatory drugs with the adjuvant therapy to tuberculosis. It can increase the cell immunity response level, promote the proliferation, differentiation of T lymphocytes, enhance the immune cell activity and the function of anti-tuberculosis drugs, thereby promoting the improvement of disease [7-8]. The study investigated the clinical efficacy of recombinant human interleukin-2 in the treatment of pulmonary tuberculosis, explored the role and the appropriate dosage with the recombinant interleukin-2 in the treatment of retreated pulmonary tuberculosis, evaluate the safety of the recombinant interleukin-2, and provided the basis for the popularization and application.

1. Objects and Methods

1.1 Objects

Choose patients with relapsing infiltrative pulmonary tuberculosis in the General Hospital of Yima Coal Industry Group from July, 2000 to April, 2002.

Diagnostic Criteria: According to the Chinese Medical Association tuberculosis branch made the diagnostic criteria for pulmonary tuberculosis and classification for the pulmonary tuberculosis [9].

Inclusion Criteria: (1) Diagnosed as relapsing infiltrative pulmonary tuberculosis by the bacteriology and X-ray. (2) All selected cases were sputum bacterium positive or negative pulmonary tuberculosis: the criteria of sputum bacterium positive pulmonary tuberculosis is a smear++, or bacterial

culture positive through the criteria identification except the non-mycobacterium tuberculosis. Bacterial culture positive needs to have a drug sensitive test. The criteria of sputum bacterium negative pulmonary tuberculosis is bacterial culture negative, but it must observe the following three conditions: (a) Specific toxic symptoms: fever, night sweats, fatigue, loss of appetite, loss of weight and so on, with the symptoms of cough, chest pain, hemoptysis, and so on. (b) Rapid red cell sedimentation rate. (c) A chest X ray manifestation is consistent with the characteristics of tuberculosis. (3) Selected cases were confirmed pulmonary infiltrates by the chest radiograph, with or without cavities. (4) Aged from 18 to 65, Type 2 diabetes fasting blood glucose was controlled below 8.3mmol/L. (5) Without using any immune agents in the past 3 months. (6) The cases received treatment according to the scheme，with completed follow-up data .

Exclusion Criteria: (1) The patients combined with extrapulmonary tuberculosis. (2) Pregnant and nursing women. (3) HIV positive. (4) The patients with severe disease of heart, liver, kidney. (5) The patients have history of mental disease or epilepsy. (6) patients are allergic to the biological preparation, especially the interleukin-2.

1.2 Drug Source

Experimental drugs (IL-$_2$, named Yuan Cexin) were provided by the Beijing Yuance Pharmaceutical Co., Ltd. and the lot number was 000115A.

1.3 Testing Plan

In order to ensure the comparability for the analysis of clinical testing design, the patients in hospital were randomly divided into the test group and the control group by the number of admission. The dosing regimen of the test group: 2HL$_2$AK（E）Z（TH）V+IL-$_2$/1HL$_2$Z （TH） V+IL-$_2$/5HL$_2$V； The dosing regimen of the control group: 2HL$_2$AK（E）Z（TH）V/1HL$_2$Z(TH)V+ /5HL$_2$V. Among them, H is Isoniazid, L is Rifapentine, AK is Amikacin, E is Ethambutol, Z is Pyrazinamide , TH is Protionamide, V is Levofloxacin. Anti-tubercular agents was the conventional dose according to clinical application, the dose of IL-2 is 200 000 units.

1.4 Main Outcome Measures

1.4.1 Examination of the Sputum

Before and after 1, 2, 3, 8 months treatment, Sputum examination for acid fast bacteria three times a month, Sputum mycobacterium tuberculosis culture once a month (bacterial culture positive needs to add the drug sensitive test). Drug sensitive tests adopted the absolute concentration method. Observing the situation of the sputum conversion, sputum negative conversion no more relapsed for 2nd consecutive month as the criteria. Patients with sputum negative had “no positive” as the criteria.

1.4.2 X-Ray Examination

Take the chest radiography in the 1, 2, 3, 8 months before and after the treatment, and if it is necessary, the patient needs to have the examination of Tomography. (1) Focus assessment. Apparent absorption: focus absorption≥1/2 Primary lesions; Absorption: focus absorption<1/2 Primary lesions; invariance: the focus had no apparent change; Deterioration, the focus expanded or disseminated. (2) Cavity evaluation. Closure: closed or blocked; narrow, cavity narrow≥1/2 original cavity diameter; Invariance: cavity narrowed or expanded<1/2 original cavity diameter; Deterioration, cavity expanded>1/2 original cavity diameter.

1.4.3 Immunological Examination

Before treatment, or at the end of the third or eighth month of treatment, do the following: (1) Adopting the APPAP bridging method, to measure the CD4， CD4/CD8 according to the manufacturers’ instructions. The CD4/CD8 is low or high before treatment, it is effective that the CD4/CD8 recovers to the normal and the ratio increases after treatment, no change before and after treatment is invalid. (2) NK cell assay: it is effective that the result is lower than that of normal before the NK cell treatment and recover to the normal after the NK cell treatment. (3) Soluble receptor of interleukin-2 determination: it is effective that after the treatment the result is lower than that before the treatment.

1.4.4 Other Inspections

Before the treatment and in the course of treatment, having the inspections of blood routine, urine routine, renal function, liver function, blood biochemical, and the electrolyte once a month.

1.4.5 Adverse Drug Reaction Evaluation

A variety of adverse drug reaction and the changes of the injection of recombinant human interleukin-2 with local swelling, tenderness, induration, limitation needed to be observed during the treatment.

1.5 Data Processing

χ^2 test was used to measure the comparison of rate, t test was used to measure the comparison of immunological data.

2. Results

2.1 The General Information of Patients

For the test group and the control group, each group had 99 cases, there were two patients lost to follow-up in the test group, the control group had 8 patients lost to follow-up. 97cases involved in the result analysis in the test group, and 91cases in the control group. There were 37males and 60 females in the test group, the average age was 45.8±8.2, 74 cases had 1-2 lung fields, 16 cases had 3-4 lung fields. 7cases had 5-6 lung fields. 69 cases had cavity. There were 40 males and 51 females in the control group, the average age

was 43.3±8.0, 67 cases had 1-2 lung fields, 24 cases had 3-4 lung fields, 74 cases had cavity. The gender, age, lesion of lung field, and the cavity number of two groups were approximate. Therefore, it was comparable between the two groups.

2.2 The Situation of Sputum Negative Conversion

In the test group, the rate of sputum smear negative conversion was 22.3%，34.9%，60.7%，79.5% after the treatment for 1, 2, 3, 8 months. In the control group, the rate of sputum smear negative conversion was 21.8%，30.2%，35.3%，55.6%. It was obvious that the rate in the test group was higher than that in the control group during the period of the third month and the eighth months (P<0.05). See figure 1.

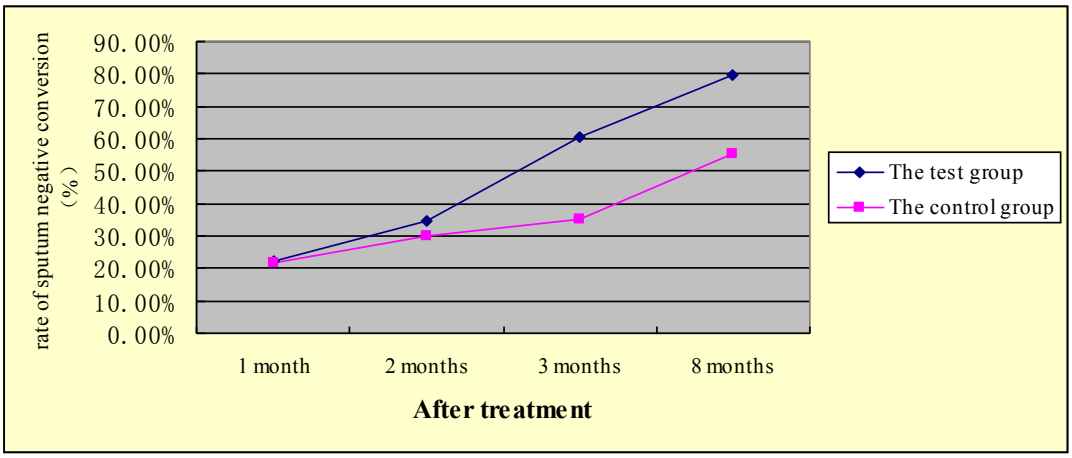

Figure 1 The situation of sputum negative conversion after the treatment for 8 months

2.3 The focus absorption situation

All the cases had lesions, in the test group, the rate of the focus absorption is 26.8%，46.2% after the treatment for 3 and 8 months. In the control group, the rate is16.4%, 28.4%, it is obvious that the rate of the focus absorption in the test group is higher than in the control group after the treatment for 8 months（P<0.05). See figure 2.

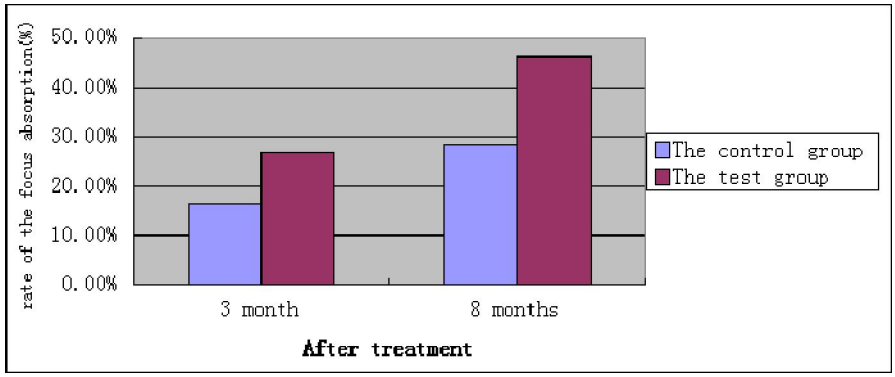

Figure 2 The rate of the focus absorption situation

2.4 Cavity Closure Situation

The rate of cavity closure was higher in the test group after treatment for 2 months, 3months and 8 months, but the two groups didn't have the statistical significance (P>0.05), see table 1. In addition, 1 case in the test group (1.4%) and 3cases in the control group(4.0%) appeared the expansion of cavity at the end of the treatment, the two groups didn't have the statistical significance difference.

Table 1 The cavity closure situation in the test group and the control group (n/%)

Groups	n	2 months	3 months	8 months
The test group	69	1/1.4	7/10.1	27/39.1
The control group	74	2/2.7	2/2.7	18/24.3
P		1.000	0.089	0.145

2.5 Immunological Evaluation

97 cases of the test group and 91 cases of the control group had the determination of T lymphocyte subsets before and after the treatment. Compared with the control group, the ratio of CD4, CD4/CD8 had improved significantly at the third month and the eighth month in the test group, which had the statistical significance($P<0.05$). But the ratio of CD4, CD4/CD8 of the control group had not apparent changes before and after the treatment. Interleukin-2 soluber receptor in test group decreased apparently at the third month and the eighth month, compared with the test group, they had no significant difference. NK cells in the test group and the control group had no significant difference. See table 2.

Table 2 The test results of two groups about patients' immunological indexes (x±s)

groups	time	CD4	NK cells	CD4/CD8	Interleukin -2 soluble receptor
The test group	Before treatment	33.0±2.5	34.7±3.9	1.05±0.14	1041.5±217
	treatment of 3months	35.3±2.6*▲	33.3±3.8	1.07±0.13*▲	819.9±419.0*▲
	treatment of 8months	37.3±2.2*▲	32.9±2.2	1.17±0.16*▲	717.4±432.8*▲
The control group	Before treatment	33.8±1.6	33.1±2.4	1.02±0.10	1030.2±206.5
	treatment of 3months	33.4±2.3	33.6±3.8	0.96±0.53	974.7±258.0
	treatment of 8months	34.7±4.3	33.7±2.5	1.07±0.48	827.8±493.1

Notes: Compared with that before treatment, *P<0.05; Compared with the control group ▲P<0.05

2.6 Drug adverse reaction

There were 4 cases in the test group and 7 cases in the control group, having adverse drug reactions caused by anti-tuberculosis drugs. A total of 18 cases of adverse reactions were caused by the interleukin-2 in the test group. 10 cases appeared the symptoms of local swelling and induration, because of the injection in 1-3days (1 patient had the symptom of tenderness), 1 case lasted 20 days, and the rest cases lasted 7-10 days, 2 cases just had the symptoms of feverish, 6 cases had a fever with chills, which disappeared and had no influence after the different symptomatic treatments.

3 Discussion

Pulmonary tuberculosis is a kind of diseases with a low immune function, especially the severe patients of re-treatment and refractoriness had the significant symptoms. Therefore, it is of important significance for patients to improve the treatment of tuberculosis and the quality of life by improving the immunological status of the body [10-11]. At present, the treatment of tuberculosis advocates comprehensive treatment, one of the important things is the application of immunopotentiator. The basic and clinical research proved that recombinant human interleukin-2 can enhance human' immune function and disease-resistant ability, and promote the immunopotentiator of lesion absorption [12-13].

The research applied recombinant human interleukin-2 in the adjuvant therapy of re-treatment pulmonary tuberculosis patients with sputum smear in 3, 8 months of negative conversion rate which was significantly higher than that in the control group. The treatment of 8 months the focus absorption rate in the test group was significantly higher than that in the control group, the differences between the two groups were not significant ($P<0.05$), the rate of cavity closure in the test group was higher than that in the control group during the treatment of 2, 3, 8 months, but there were no statistical significance between the two groups($P>0.05$). The results showed that pulmonary tuberculosis patients were significantly improved in the aspects of the sputum mycobacterium tuberculosis negative conversion, lesion improvement, after using the recombinant human interleukin -2 combined with the anti-tuberculosis drugs.

In most cases, the patients infected with Mycobacterium tuberculosis had no symptoms and infectivity. However, when the response of immune was in disorder, the reactivation of potential infection can occur, and then developed the active pulmonary tuberculosis. The body can acquire the immunity against Mycobacterium tuberculosis, which was mainly provided by the cell-mediated immunity of T lymphocyte. $CD4^+$, $CD8^+$ T lymphocytes not only play a major role in the development of tuberculosis, but also become the wide research issues for many people at present[14-18]. Cellular immune function of patients with tuberculosis will decline in different degrees, improving the patients' immune systems in the course of chemotherapy, it is of great significance to improve the rate of re-treatment and reduce recurrence to pulmonary tuberculosis. The study adopted to treatment prescription of anti-tuberculosis drugs combined with recombinant human interleukin-2, the results showed that the ratio of CD4, CD4/CD8 in the test group were significantly increased at the third months and the eighth months, compared with the control group, they had the statistical significance($P<0.05$), but there were no significant differences before and after treatment. Soluble interleukin-2 receptors in test group were significantly lower at the third months and eighth months than that before treatment, compared with the

control group, they had the differences (P<0.05). This study was consistent with the reports of many domestic scholars.

Above all, on the basis of anti-tubercular drugs, combining with the recombinant human interleukin-2 can play the role of adjuvant therapy to the re-treatment of pulmonary tuberculosis patients, which can accelerate the sputum negative conversion, promote the focus absorption, and improve the immune function. The clinical efficacy was quite well, and had no obvious adverse reaction. It was proved that the recombinant human interleukin-2 with small doses of 200 000 units was the safe and reliable biological agent, which was well worthy popularizing in clinical application.

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