



Exogenous intrapleural injection of interleukin-27 may improves outcome and prognosis in patients with tuberculous pleural effusion

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ABSTRACT

We hypothesize that exogenous intrapleural injection of interleukin-27 may improve outcome and prognosis in patients with tuberculous pleural effusion (TPE). Studies have found that the balance of Th1/Th2 determines the development trend of TPE. High concentrations of IFN- γ and TNF- α in pleural effusion are associated with pleural adhesion in patients with TPE. Interleukin-27 is a member of the IL-12 family, and IL-27 has a dual regulatory effect on Th1 immunity. On one hand, IL-27 can promote the initial CD4+ T cell proliferation by inducing the expression of T-bet, IL-12R β 2 and ICAM-1 in the initial CD4+ T cells, and also promote its differentiation into Th1 cells and IFN- γ production in the early infection. On the other hand, in the case of high Th1 polarization, IL-27 induced STAT3 phosphorylation and inhibited TNF and IL-12 production in activated peritoneal macrophages, indicating a novel feedback mechanism by which IL-27 can modulate excessive inflammation, thereby preventing damage to the body caused by excessive immune response. Studies have confirmed that after stimulation of antigen by mononuclear cells in TPE, the Th1 and Th2 cell subsets and Th1/Th2 ratio markedly increase, and the increase of Th1 is more obvious than that of Th2. Therefore, compared to patients with TPE in the high-level IL-27 group, we hypothesized that pleural effusion is absorbed more slowly, pleural thickening is more obvious, pleural adhesions are more extensive, and the incidence of thoracic collapse is higher in the low-level IL-27 group under the same conditions of anti-tuberculosis treatment. However, exogenous intrapleural injection of IL-27 may induce Stat3 phosphorylation and inhibit TNF and IL-12 production, finally reduces the secretion of IFN- γ and TNF- α . This negative regulation inhibits the excessive inflammatory reaction caused by tuberculosis infection, reduces pleural adhesion, pleural thickening and local pleural tissue damage, thereby improving the prognosis of patients.

Introduction

Tuberculous pleural effusion (TPE) is an inflammatory response of the pleura caused by the entry of mycobacterium tuberculosis (MTB) and its metabolites into the thoracic cavity in high-sensitivity state [1]. In addition to destroying capillaries and microlymphatic vessels, the inflammatory reaction can also inhibit fibrinolytic activity, so that fibrin, leaks out of the pleural cavity, but cannot be dissolved or absorbed in the pleural cavity, and then deposited and adhered. Its pathological features are pleural congestion, edema, leukocyte infiltration, fibrin deposition on the pleural surface, and accumulation of plasma proteins in the pleura [2]. Henke et al. [3] believe that if the pleurisy is not treated in time in the effusion period, the fibrin will be widely deposited on the pleura; the effusion will become thicker; the proliferating cells will also infiltrate; and the fibrin layer will be formed, thereby causing pleural thickening and adhesion, and thoracic collapse, affecting lung function and reducing the quality of life of patients. In addition to anti-tuberculosis treatment, there are few ways to reduce pleural adhesions caused by TPE. It has only been reported that intrapleural injection of urokinase or heparin drugs plays a role in

preventing pleural adhesions caused by TPE because of interfering with the coagulation and fibrinolysis system [4–6]. However, no studies have been reported on the improvement of pleural adhesion and pleural thickening by regulating the local immune status of the thoracic cavity.

Studies have found that the balance of Th1/Th2 determines the development trend of TPE, and high concentrations of IFN- γ and TNF- α in pleural effusion are associated with pleural adhesion in patients with TPE [7–9]. CD4+ T lymphocytes are the main immunoregulatory cells that antagonize MTB infection in humans, and are mainly classified into Th1 (mainly secreting IFN- γ and IL-2) and Th2 (mainly secreting IL-4 and IL-10). The Th1 cell-mediated immune response plays the most critical role in the control of tuberculosis. The Th2 cell subset mainly antagonizes the Th1-type immune response, inhibits macrophage activation, and blocks the effect after IFN- γ activates macrophages. Xiao Ling et al. [10] have showed that the concentration of IFN- γ in TPE pleural effusion is remarkably increased, and the IFN- γ concentration in patients with encapsulated pleural effusion is obviously higher than that in patients with free pleural effusion. It is considered that Th1 type immunity in local TPE is enhanced, and too strong Th1 immunity easily leads to pleural adhesions. Ge Qi-ping et al. [11] have confirmed that

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after stimulation of antigen by mononuclear cells in TPE, the Th1 and Th2 cell subsets and Th1/Th2 ratio markedly increase, and Th1 increases at a high level, while Th2 increases at a low level. Thus, it is speculated that the localized Th1 response in the thoracic cavity of patients with TPE is negatively regulated by the same enhanced Th2 response, and the proportion of Th1 cells in CD4+ T cells is also gradually increased, while the proportion of Th2 cells is not remarkably changed in different pleural adhesion groups. The ratio of Th1/Th2 also shows an increasing trend, and the balance of Th1/Th2 is dramatically shifted. These indicate that the Th1-type immune response is enhanced, and confirm that enhanced Th1 type immunity is associated with pleural adhesions. TPE has differences in local thoracic and systemic Th1 and Th2 responses. The number of Th1 and Th2 and the cytokines IFN- γ , IL-2, IL-4, IL-10 and TNF- α in TPE were significantly higher than those in peripheral blood, indicating that the Th1 and Th2 immune responses in the local pleural cavity are significantly stronger than these in systemic immune response. IFN- γ can enhance the cellular immune response in the local thoracic cavity and facilitate the clearance of pathogens. At the same time, IFN- γ , acts as a proinflammatory factor, can aggravate the inflammatory reaction to damage the pleural tissue. High concentration of IFN- γ in the pleural effusion stimulates activated macrophages, CD4+ T Cells, even mesothelial cells secrete TNF- α , which results in high levels of TNF- α in TPE. High levels of TNF- α can cause a severe inflammatory response in the pleura. A previous study has shown that IL-4 levels are significantly higher in TPE than in serum, and suggested that Th2 immune response is also enhanced in the local thoracic cavity of tuberculous pleurisy, but there is no significant difference in the ratio of Th2 cells and IL-4 levels in the different degrees of pleural adhesion. It is suggested that the Th2 immune response only increases to a certain extent in the thoracic cavity of tuberculous pleurisy, and negatively regulates the Th1 immune response. The degree of regulation is limited. The severe Th1 immune response will lead to the imbalance of Th1/Th2. There is no significant correlation between Th2 immune response and pleural adhesions. On the contrary, the Th1 immune response in patients without pleural adhesion is not obviously enhanced, and there is no significant imbalance in Th1/Th2 [12,13]. The ratio of Th1 and Th2 cells and their cytokine levels are increased in TPE. The proportion of Th1 cells and the contents of IFN- γ and TNF- α increase distinctly, and the higher the level, the more obvious the pleural adhesion is. These results have confirmed that excessively strong Th1-type cellular immune response can cause pathological immune damage to pleural tissue, which is associated with the severity of pleural adhesion, and provides an idea for immunotherapy of TPE patients.

IL-27 is a member of the IL-12 family and a cytokine in the form of p28/EB13 heterodimers, is secreted by activated antigen-presenting cells, and binds to receptors on the surface of natural killer cells, monocytes, macrophages, induces dimerization of intracellular gp130

and subunit WSX-1, and JAK protein phosphorylation. Subsequently, STAT is phosphorylated by JAK to form a dimer, which enters the nucleus to regulate the expression of the target gene [14–21]. At present, a number of studies have confirmed that the content of soluble IL-27 in pleural effusion of patients with TPE is higher than that of non-TPE [22–24], and IL-27 has a dual regulatory effect on Th1 immunity. On one hand, IL-27 can promote the initial CD4+ T cell proliferation by inducing the expression of T-bet, IL-12R β 2 and ICAM-1 in the initial CD4+ T cells, and also promote its differentiation into Th1 cells and production of IFN- γ in the early infection. IFN- γ secreted by Th1 cells is also a pro-inflammatory factor, aggravates the inflammatory reaction at the lesion and aggravates tissue damage, which theoretically increases the probability of pleural adhesions [25,26]. On the other hand, in the case of high Th1 polarization, IL-27 induced STAT3 phosphorylation and inhibited TNF and IL-12 production in activated peritoneal macrophages, indicating a novel feedback mechanism by which IL-27 can modulate excessive inflammation, thereby preventing the damage to the body caused by excessive immune response [27].

Hypothesis

In the light of above experimental results, the higher the proportion of Th1 cells in pleural effusion and the higher the levels of IFN- γ and TNF- α , the more obvious pleural adhesion is. Moreover, in the case of high Th1 polarization, IL-27 can also inhibit the intensity and duration of IL-2 mediated Th1-type immune responses by Stat3, thereby preventing the damage to the body caused by excessive immune response. Therefore, we speculated that high levels of IL-27 in pleural effusion could induce Stat3 phosphorylation and inhibit TNF and IL-12 production, reduces the secretion of IFN- γ and TNF- α in pleural effusion. Negative regulation of Th1 type responses inhibits excessive inflammatory responses induced by MTB infection, reduces pleural adhesions, pleural thickening, and damage to local pleural tissue. Thus, we hypothesized that the low-level IL-27 group in the pleural effusion of TPE patients has slower pleural effusion absorption, more obvious pleural thickening, wider pleural adhesion, and higher incidence of thoracic collapse under the same conditions of anti-tuberculosis treatment. Nevertheless, exogenous injection of IL-27 may reduce the concentrations of IFN- γ and TNF- α in pleural effusion inhibit Th1-type immune response, and mitigate the severity of pleural adhesions and pleural thickening in TPE patients with low-level IL-27 in pleural effusion.

Hypothesis assessment

These studies, which have been published and are for this hypothesis, are as follows, while there is no apparent evidence against this hypothesis (Table 1). First, we should collect the pleural effusion and

Table 1
Evidences for this hypothesis.

Items	Names	Function	References
Evidences are for this hypothesis	IL-27	The content of soluble IL-27 in pleural effusion of patients with TPE is higher than that of non-TPE, and IL-27 has a dual regulatory effect on Th1 immunity. On one hand, IL-27 can promote the initial CD4 + T cell proliferation and also promote its differentiation into Th1 cells and production of IFN- γ . IFN- γ secreted by Th1 cells is also a pro-inflammatory factor, aggravates the inflammatory reaction at the lesion and aggravates tissue damage, which theoretically increases the probability of pleural adhesions. On the other hand, in the case of high Th1 polarization, IL-27 can also inhibit the intensity and duration of IL-2 mediated Th1-type immune responses by Stat3. Thereby preventing the damage to the body cause by excessive immune response.	Many articles have reported this function. eg:[23–27]
	TNF- α	High concentrations of IFN- γ and TNF- α in pleural effusion are associated with pleural adhesion in patients with TPE. In the case of high Th1 polarization, IL-27 could induce Stat3 phosphorylation and inhibit TNF and IL-12 production, finally reduces the secretion of IFN- γ and TNF- α .	Many articles have reported this function. eg:[12,13,27]
Evidence is against this hypothesis	There is no evidence against this hypothesis		

serum of TPE patients and monitor the concentrations of IL-27, IFN- γ , TNF- α , and IL-2, and TPE patients are divided into two groups according to IL-27 level. CT and chest color ultrasonography were used to monitor pleural thickness and pleural adhesion, to observe whether the pleural fluid absorption rate of the low-level IL-27 group is slower whether pleural thickening is more obvious, whether pleural adhesion is more extensive, and whether thoracic collapse rate is higher than high-level IL-27 group under the same anti-tuberculosis treatment condition. Second, a rat TPE model is constructed to measure the levels of IFN- γ and TNF- α in the pleural effusion of TPE rats, pleural adhesion and pleural thickening in control group and experimental group after exogenous intrapleural injection of recombinant human IL-27, to compare changes of pleural adhesion and pleural thickening in TPE rats between these groups after exogenous intrapleural injection of recombinant human IL-27. Finally, exogenous thoracic injection of IL-27 can improve the outcome and prognosis of TPE rats.

Hypothesis suggestion

This hypothesis, from an immune perspective, looks for ways to improve pleural adhesion, pleural thickening and thoracic collapse in patients with TPE. The method of injection is simple, and human recombinant IL-27 is easily obtained. Just as IL-2 is currently injected into the thoracic cavity to treat malignant pleural effusion, there are no obvious contraindications and it is suitable for a wide range of people. If this hypothesis is valid, the concentrations of IFN- γ and TNF- α in the pleural effusion of TPE patients will decrease; the absorption rate of pleural effusion will be fast; the pleural adhesions will mitigate; and the thoracic collapse rate will diminish after local injection of human recombinant IL-27 into the thoracic cavity. These can improve the outcome, prognosis, and the quality of life of TPE patients.

Declaration of Competing Interest

The authors declare no conflict of interest regarding the publication of this paper.

Acknowledgment

This work was supported by Natural Science Foundation of China (Grants 81570010).

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.mehy.2019.109319>.

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