Evaluation of TNF-α cytokine production in patients with tuberculosis compared to healthy people

Evaluierung der TNF-α-Zytokinproduktion bei Patienten mit Tuberkulose im Vergleich zu gesunden Menschen

Abstract

Background: Mycobacterium tuberculosis (TB) is one of the most important causes of human mortality. Approximately one-third of the world’s population is infected with TB and 5–10% of them develop the active form of the disease. Cytokines play a major role in the host defense process against Mycobacterium infections. Among these cytokines, tumor necrosis factor alpha (TNF-α) has a prominent role in the defense of and pathological responses to tuberculosis.

Materials and methods: A case-control study was carried out from May 2016 to June 2017. 45 patients with diagnosis of tuberculosis (smear and positive culture) were included as case group and 45 healthy subjects as control group. The serum levels of TNF-α were determined with the enzyme-linked immunosorbent assay (ELISA) method.

Results: The concentration of TNF-α in patients with TB was significantly higher than in the control group (P<0.05). However, the difference was only significant in the age groups 20–30 and 50–60 years; in the age groups 30–40, 40–50 and 50–70 years, the difference was not significant, although certain trends were apparent.

Discussion and conclusion: Since the level of serum TNF-α is higher in patients with pulmonary tuberculosis than in individuals without it, the measurement of TNF-α levels can be useful as a probable marker for the diagnosis of tuberculosis.

Keywords: mycobacterium tuberculosis, tumor necrosis factor

Zusammenfassung

Hintergrund: Mycobacterium tuberculosis (TB) ist eine der wichtigsten Ursachen der menschlichen Sterblichkeit. Ungefähr 1/3 der Weltbevölkerung ist mit TB infiziert; davon erkranken 5–10%. Zytokine spielen eine wichtige Rolle im Abwehrprozess gegen Mycobacterium-Infektionen. Unter den Zytokinen besitzt der Tumornekrosefaktor (TNF-α) eine herausragende Rolle bei der Abwehr und bei pathologischen Reaktionen auf Tuberkulose.

Material und Methode: Die Fall-Kontroll-Studie wurde vom Mai 2016 bis Juni 2017 durchgeführt. Es wurden 45 Patienten mit der Diagnose Tuberkulose (Abstrich und positive Kultur) als Fallgruppe und 45 gesunde Probanden als Kontrollgruppe untersucht. Die Serumspiegel von TNF-α wurden mittels ELISA bestimmt.

Ergebnisse: In der Fallgruppe war die Konzentration von TNF-α signifikant höher als in der Kontrollgruppe (P<0,05). Der Unterschied war jedoch nur in den Altersgruppen 20–30 und 50–60 Jahre signifikant; in den Altersgruppen 30–40, 40–50 und 50–70 Jahre war die Differenz nur tendenziell.

Diskussion und Schlussfolgerung: Da bei Patienten mit Lungentuberkulose der TNF-α Serumspiegels erhöht ist, kann seine Bestimmung als möglicher zusätzlicher Marker für die Diagnose der Tuberkulose nützlich sein.
Introduction

*Mycobacterium tuberculosis* (TB) is one of the most important causes of mortality in humans. According to the World Health Organization report in 2017, tuberculosis is responsible for 1.7 million deaths every year [1]. About one-third of the world population is infected, but only about 10% of these develop active tuberculosis [2]. The causative agent is the intracellular bacterial pathogen itself; its antigens have the ability to stimulate the production of cytokines via the mononuclear phagocyte system. Studies show that susceptibility to the disease varies among individuals, e.g., not everyone who is exposed to this bacterium becomes infected with TB [3]. The course of the disease also differs from person to person. These differences can be due to host factors and the genetic sensitivity of various individuals [3]. In this disease, cellular immunity and cytokines are intermediaries in the immune system and inflammatory responses [4], [5]. It seems that the acquired immune response in the pathway of T-helper lymphocytes can limit and stop bacterial growth. These cells secrete cytokines, such as interferon gamma and TNF-α, and stimulate macrophages to further produce more active oxygen and free radicals. As a result, cells are better able to kill microbes, and more microbial antigens are delivered to T cells [6], [7]. T-helper 2-type cytokines such as IL-4, IL-5, IL-10, IL-13 influence the course of the disease [8]. Tumor Necrosis Factor-α is a cytokine produced by many types of cells, such as macrophages and monocytes. TNF-α exists in two forms; a type II transmembrane protein, and a mature soluble protein. However, this cytokine is initially produced as a membrane protein of 212 amino acids [9]. TNF-α binding to cell surface receptors TNFRp55 (TNF-R1, CD120a or p55/60) and TNF Rp75 (αR2, CD120b and p75/80) triggers an intracellular signaling cascade. The Rp55 receptor is expressed by most tissues, and TNF-α terraform solution is mainly attached to Rp55 receptor, although activated by both types of TNF-α. The membrane associated form mostly binds to TNF Rp75, which is specific to the immune system cells [10]. Awareness of the specific functions and the activation of TNF-α receptors considerably help to understand the inhibitory effects of TNF-α on some diseases [11]. The purpose of this study was to determine the importance of TNF-α in protecting against tuberculosis and investigate the rate of production of this cytokine in patients with tuberculosis as compared to the control group.

Materials and methods

Study design

A case-control study was conducted from June 2016 to July 2017. The case group consisted of 45 patients with a definite tuberculosis diagnosis (with smear and positive culture), and 45 healthy subjects were selected as the control group.
with pulmonary tuberculosis. Fatima et al. [15] studied the serum level of TNF-α in patients with tuberculosis. The results of their study, which were similar to those of the present study, showed a significant increase in serum levels of TNF-α in the patient group compared to the control group (P<0.001). Other studies have shown the importance of TNF-α in patients with tuberculosis [16], [17], [18]. The results of these studies showed that the serum levels of TNF-α decreased significantly with treatment. Nakya et al. [19] studied serum levels of TNF-α in patients with active pulmonary tuberculosis compared to a control group. The results of that study showed that the serum level of TNF-α increased significantly in the patient group compared to the control group, which agrees
Table 1: Analysis of TNF-α serum levels by age groups

| Bonferroni multiple comparisons | Difference | Tukey | 95% CI of diff |
|----------------------------------|------------|-------|---------------|
| 20–30 vs. 30–40                  |            |       |               |
| Patient                          | −1.957     | 1.574 | −5.859 to 1.946 |
| Control                          | 0.5962     | 0.4568| −3.499 to 4.692 |
| 20–30 vs. 40–50                  |            |       |               |
| Patient                          | −1.273     | 0.8490| −5.980 to 3.433 |
| Control                          | 0.9214     | 0.6130| −3.796 to 5.638 |
| 20–30 vs. 50–60                  |            |       |               |
| Patient                          | 1.560      | 0.8160| −4.440 to 7.560 |
| Control                          | −0.1000    | 0.06376| −5.022 to 4.822 |
| 20–30 vs. 60–70                  |            |       |               |
| Patient                          | −2.522     | 1.987 | −6.504 to 1.460 |
| Control                          | 0.5833     | 0.2967| −6.754 to 5.587 |
| 30–40 vs. 40–50                  |            |       |               |
| Patient                          | 0.6833     | 0.4706| −3.874 to 5.240 |
| Control                          | 0.3253     | 0.2389| −3.948 to 4.598 |
| 30–40 vs. 50–60                  |            |       |               |
| Patient                          | 3.517      | 1.876 | −2.367 to 9.400 |
| Control                          | −0.6962    | 0.4857| −5.194 to 3.802 |
| 30–40 vs. 60–70                  |            |       |               |
| Patient                          | −0.5652    | 0.4662| −4.370 to 3.239 |
| Control                          | −1.179     | 0.6341| −7.017 to 4.658 |
| 40–50 vs. 50–60                  |            |       |               |
| Patient                          | 2.833      | 1.380 | −3.611 to 9.278 |
| Control                          | −1.021     | 0.6322| −6.092 to 4.049 |
| 40–50 vs. 60–70                  |            |       |               |
| Patient                          | −1.248     | 0.8470| −5.874 to 3.377 |
| Control                          | −1.505     | 0.7508| −7.794 to 4.785 |
| 50–60 vs. 60–70                  |            |       |               |
| Patient                          | −4.082     | 2.158 | −10.02 to 1.855 |
| Control                          | −0.4833    | 0.2354| −6.928 to 5.961 |

Conclusion

The findings of this study show that cytokines can be used as a possible marker for the diagnosis of tuberculosis. Measuring the level of cytokines can be used to supplement the diagnostic tests for Mycobacterium. More studies are needed to confirm these findings.

Notes

Competing interests

The authors declare that they have no competing interests.
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