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Differential diagnosis of tuberculosis, sarcoidosis, and anthracosis by CD8, CD3, and CD4 levels by flow-cytometry

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ABSTRACT

Background: Determination of a desirable diagnostic tests is an issue of importance especially to differentiate between tuberculosis) TB(, sarcoidosis, and anthracosis. The purpose of the present study was to determine the differential diagnosis of tuberculosis, sarcoidosis and anthracosis with CD8, CD3, and CD4 by flow-cytometry.

Materials and Methods: In this descriptive cross-sectional comparative survey, 40 consecutive patients attending to Masih Daneshvari Hospital in Tehran were enrolled and CD4/CD8 ratio, CD8, CD3, and CD4 were determined by flow-cytometry and compared across patients with three diseases including tuberculosis, sarcoidosis, and anthracosis.

Results: The results demonstrated that CD4 was significantly higher in anthracosis cases ($P<0.007$) and the CD8 was significantly higher in patients with TB ($P<0.008$).

Conclusion: It was attained ultimately that CD4 and CD8 levels could be a desirable diagnostic markers for anthracosis and TB, respectively.

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INTRODUCTION

Sarcoidosis is a systemic disease characterized by the presence of non-caseating granulomas in affected organs, with the lung being the major diseased organ in more than 90% of patients [1,2]. Particularly, studies on BAL fluid in pulmonary sarcoidosis have demonstrated that sarcoid granuloma formation in the lung is preceded by an influx of mononuclear cells into the alveoli [1]. The presence of activated alveolar macrophages and CD4 (helper/inducer) T-lymphocytes were shown as markers of alveolitis [3,4]. Moreover, the increased numbers of BAL CD4 T cells are considered a hallmark of pulmonary sarcoidosis [5,6]. The percentages of CD4 and CD8 T-cells in BAL correlate properly with IHC methods in sarcoidosis patients [7]. Therefore, the BAL lymphocytosis, low or normal granulocytes, and the increase in CD4/CD8 ratio appears in pulmonary sarcoidosis are not disease-specific and refined flow cytometric tests could be required to assist the diagnosis [8-10].

Apparently, the organs involved in the disease include the respiratory system (In 90% of patients), visual system, and the immune system, which could be seen in patients in active phase [11]. It also causes high mortality in individuals who are particularly aged 55 years and older [12]. Therefore, the adoption of methods for early diagnosis and reduction of disease severity can play a significant role in reducing the burden of disease. Respiratory complications in patients with sarcoidosis could increase the respiratory tract allergens, particularly in allergens [13]. Anthracosis refers to pneumoconiosis due to coal dust and in the lung caused by the deposition of carbon particles, silica, quartz, etc. In the mucosa, submucosa and inside macrophages, which appear as black lesions in the bronchoscopy view [14,15]. The aforementioned patients are more prone to respiratory infections, such as TB which the rates are 27% to 75% [16]. Therefore, adopting strategies for timely diagnosis and treatment and reducing the severity of the disease can be effective in reducing the burden of disease. Tuberculosis infection is caused by Mycobacterium tuberculosis. Among the most common infectious diseases, especially in countries, such as Iran. This organism could grow in different environments and thus the possibility of transferring it to others could be quite high. Eloquently, its prevalence varies from 3.7 to 9.6 % [17,18]. However, this incidence in different regions and in those with underlying diseases and conditions varies; moreover, the importance of each area should be considered [11,12]. TB as a global emergency, requires assessment comprehensively. In addition to the failure of TB control programs in most developing countries, in recent years it has been attained that the treatment of TB is a multidimensional issue that includes complications, efficacy, compliance of patients, and drug resistance complications [17-19]. According to the above-mentioned methods, in order to reduce the proportion of TB is quite important in Iran. In fact, the number of patients with TB has increased globally, as well as an increase in antibiotic resistance rates, which has also been reported in children [20,21]. In this regard, the primary step toward the initiation of treatment is to diagnose the bacteria appropriately; therefore, the clinicians can make the proper action and optimize the treatment process. Diagnostic markers, such as CD8, CD3, and CD4 are expected to be able to distinguish between these three diseases by flow cytometry. For this reason, mentioning the importance of reading and to reduce the dimensions of the case, in this study, the differential tuberculosis, sarcoidosis and anthracosis using diagnostic markers CD8, CD3, and CD4 could be analyzed by using flow cytometry.

MATERIALS and METHODS

In this observational descriptive cross-sectional study, 40 BAL (Broncho alveolar lavage) samples from patients suspected of having tuberculosis (n = 10), sarcoidosis (n = 20), and anthracosis (n = 10) who had been referred to Masih Daneshvari Hospital, evaluated and CD8, CD3, and CD4 determined by using flow cytometry and were compared in the three types of the disease.

The inclusion criteria were the following: cases with tuberculosis, anthrax, and sarcoidosis; on the other hand, the exclusion criteria included a reluctance to continue the collaboration by the patient. In this study, the information was kept confidential and the participation in this research was voluntarily. Eventually, after gathering the required information from all subjects, we analyzed the data in which we used SPSS version 24 software.

Statistical tests which were used in this context included chi-square tests and analysis of variance (ANOVA) and the level of significance for the interpretation of the correlations between variables was 0.05.

RESULTS

According to the data, 20% in the tuberculosis group, 30% in the anthracosis group, and 50% in the sarcoidosis group were male (P>0.05).

The mean age was significantly lower in patients with sarcoidosis than in other diseases (P=0.004), the mean age in the TB group was 56.7, in the anthracosis group was 62.1, and in the sarcoidosis group it was 47.5 years.

As shown in Table 1, the mean CD3 and CD4 / CD8 ratio did not indicate significant differences between the three groups (P>0.05) but the average CD4 significantly in anthracosis (P<0.007) and the mean of CD8 was significantly in the tuberculosis group (P<0.008) were higher than the other two groups. Distribution of diagnostic markers based on age and gender groups showed no significant difference (p>0.05).

Table1. Distribution of diagnostic markers in three groups

	CD3	CD4	CD8	CD4/CD8
TB	39.2 ± 28.8	14.1± 11.9	25.1 ± 21.3	1 ± 0.8
Anthracosis	29.7 ± 17.6	23.8± 13.1	21.8 ± 12.6	1.8 ± 1.7
Sarcoidosis	13.4 ± 14.9	10.1 ± 8.2	8.5 ± 10.9	1.9 ± 1.5

DISCUSSION

We compared tuberculosis, sarcoidosis, and anthracosis by using diagnostic markers CD8, CD3, and CD4 analyzed by flow cytometry. The outcomes showed that the mean CD3 and CD4 / CD8 markers did not change significantly between the three groups. However, the CD4 mean was significantly higher in the anthracosis groups and the CD8 mean was significantly higher in TB groups.

In a study by Morteza et al. in Iran, the results were published in 2015, It was announced that CD8, CD4, CD56 can distinguish lung cancer from non-cancerous tissues. CD8 and CD4 also could be useful in differentiating sarcoidosis. In addition, CD4 could be useful in differentiating tuberculosis [22]. However, in this study regarding TB, it is useful in differentiation of CD8 marker. A study was conducted as a cross-sectional analytical study by Barcelos et al. in Brazil, and the results were published in 2006. By studying 20 patients with tuberculosis which was announced that CD19, CD4, and CD8 cell differentiation and also predict response in treatment can be helpful for patients [23] which is similar to our research results. In a study, a review by Sweiss et al. in the United States, published in 2010. By studying 28 patients with sarcoidosis was indicated that CD19, CD4, and CD8 could be helpful in differentiating patients with severe sarcoidosis [24]. In fact, in the case of sarcoidosis in our study was not quite helpful. In an observational study, as a cross-sectional analytical study, which was done by Kojima et al in Japan which the results published in 2012. By studying 38 patients with sarcoidosis the ration of CD4 to CD8 was helpful in differentiating between patients with sarcoidosis [25]. However, there was no significant relationship in the mentioned study with our study. In a cross-sectional analytical study conducted by Rodrigues et al. In Brazil, the results were published in 2002. By studying 61 patients with suspected tuberculosis, it was indicated that CD38, CD4, and CD8 cell could be useful in differentiating patients with high detection efficiency is 90% [26]. Transparently, in our outcomes, CD8 was the most effective for tuberculosis, which could be due to the greater involvement of cellular immunity.

Conclusion

In summary, the CD4 marker could be used to identify anthracotic and CD8 markers for diagnosis of tuberculosis which they have a proper diagnostic performance. Therefore, for laboratory diagnostic methods, they could be used to distinguish the two markers. Eventually, it is recommended that further studies be conducted to confirm the findings of this study with a higher sample size and multi-centered, and furthermore. Further studies on other diagnostic methods should be considered as well.

Indeed, it is suggested to plan effective methods to reduce the number of tuberculosis, anthracosis, and sarcoidosis in the community by identifying and reducing the effective factors in this field.

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