



Correlation of the biomarker in saliva with the oral tuberculosis in extrapulmonary tuberculosis patients

Nanda Rachmad Putra Gofur^{1*}, Aisyah Rachmadani Putri Gofur², Soesilaningtyas³, Rizki Nur Rachman Putra Gofur⁴

¹ Department of Health, Faculty of Vocational Studies, Universitas Airlangga, Surabaya, Indonesia

² Faculty of Dental Medicine, Universitas Airlangga, Surabaya, Indonesia

³ Department of Dental Nursing, Poltekkes Kemenkes, Surabaya, Indonesia

⁴ Faculty of Medicine, Universitas Airlangga, Surabaya, Indonesia

Abstract

Introduction: Tuberculosis or TB is one of the infectious diseases caused by bacterium called *Mycobacterium tuberculosis*. It can damage several organs in the body. There are more cases among men than women, Men with aged ≥ 15 years accounted for 56% of the people who had developed TB in 2019, and women accounted for 32%. The TB infection has close correlation with oral cavity and can be seen through the clinical appearance and often associated with the immune system. Individuals infected with TB have different levels of salivary marker proteins.

Objective: To analyze the correlation of the biomarker saliva with the oral tuberculosis in extrapulmonary tuberculosis patients.

Discussion: Saliva has been reported in other disease, including leukaemia, oral cancer, oral lichen planus and several periodontal diseases. Saliva also can be used as the diagnostic sample in biomarker of TB. IL-2 is an important immunomodulatory cytokine produced by several cell types including activated T cells, dendritic cells, and NK cells and is essential both for the immune response to many infectious diseases and for the maintenance of tolerance. IL-6 has previously been shown to be produced at higher levels in TB patients.

Conclusion: Several biomarkers in saliva has been used as a tool to do the early detection of oral tuberculosis due to the easy sampling method and the high level of host markers in saliva showed that these markers may be more reliably as a sample in early detection of oral tuberculosis.

Keywords: oral tuberculosis, saliva biomarker, extrapulmonary tuberculosis

Introduction

Tuberculosis or TB is one of the infectious diseases which often occur in people who lives in the tropical country. This disease caused by bacterium called *Mycobacterium tuberculosis*. It can damage several organs in the body, but most often occur in lung or called pulmonary TB. TB has an involvement with the lymph node such as parotid lymph node, and often associated with the oral cavity or called extrapulmonary TB (Zhang *et al.*, 2018) [10]. This disease can spread when people with TB resulting an aerosol borne into the air, for example when coughing, talking, eating or singing. This infection can be found in anyone, but most often found in adults. According to WHO in 2019, there are more cases among men than women, Men with aged ≥ 15 years accounted for 56% of the people who had developed TB in 2019, and women accounted for 32%. Among all those were infected, the 8,2% were living with HIV. In the world, it is estimated that 10 million people infected with TB. Globally, 7.1 million people with TB were reported to have been newly diagnosed and notified in 2019, up from 7.0 million in 2018 and a large increase from 6.4 million in 2017 and 5.7–5.8 million annually in the period 2009–2012 (World Health Organization, 2020) [8].

The extrapulmonary TB which associated with oral cavity can be determined as an oral TB. The TB infection has close correlation with oral cavity and can be seen through the clinical appearance and often associated with the immune system which has found in

oral mucous and saliva. Saliva, has a role as a cleaning agent in the oral cavity, but it also acts as an antibacterial. In saliva there are marker proteins that can be used to diagnose diseases in the oral cavity. Individuals infected with TB have different levels of salivary marker proteins. The marker protein is also useful as an indicator to determine the immune condition in the individual's oral cavity (Phalane *et al.*, 2013) [7].

Clinical appearance in oral cavity of tuberculosis is including in secondary tuberculosis. These lesions of secondary tuberculosis may occur at any oral mucosa but the most common sites are gingiva and the tongue. The oral manifestation in TB also can be followed by the form of any periodontal problems. The oral manifestation sometimes not only found in secondary stage of TB but also in primary stage. Due to that, the early diagnosis is needed to determine the best treatment plan of TB in other sites of the body which may be an uncommon appearance so that the prognosis would be good (Pateria, 2011) [6].

Epidemiology

Tuberculosis has spread all over the world. This disease is more common in people living in developing countries. This disease is more susceptible to the population belonging to young adults. In addition, this disease can be easily transmitted to workers who are around tuberculosis patients or who are in frequent contact

with tuberculosis patients. In addition to HIV, TB can also cause high mortality. Patients with HIV are also susceptible to TB infection. TB bacteria can also easily infect people who have a weakened immune system. This infection can spread through aerosol particles that come from people with TB which are then inhaled by other people. It is known that 1/3 of the world's population has been infected with latent TB. However, this latent infection can turn into an active one. WHO stated that around 9 million people were infected with TB in 2013. Deaths from TB infection reached 2.5% of all deaths in the world in 20034. The prevalence of this disease depends on virulence, innate immune system and individual susceptibility (Fogel, 2015) [1].

Diagnosis

TB diagnosis can be made through anamnesis to obtain a general history of the patient, physical examination, tuberculin skin test, lung radiology examination, and Mantoux test. In children, the tuberculin test has proven to be the most useful way to find out whether they are or have been infected with *Mycobacterium tuberculosis*. The medical history includes several questions about the patient's data or identity, main symptoms, history of systemic disease, past history of oral disease, medical history, family history, and other history related to the patient's sociodemography. On physical examination, the patient's general condition often found pale conjunctiva in the eyes and/or pale skin due to anemia, high body temperature (fever), chronic productive cough lasting more than 2 weeks, night sweats, and weight loss. In primary tuberculosis, the patient's physical examination often does not reveal any abnormalities. While on extraoral examination found enlarged lymph nodes (Kurniawati, Mertaniasih and Agil, 2017) [3].

Saliva Biomarker

A marker protein or biomarker is an objectively measured and evaluated indicator of a normal biological process, a pathogenic process, or a pharmacological response to a therapeutic intervention. Therefore, biomarkers are used as markers in the body that are able to provide information that can explain pathological and physiological conditions in the human body. There are various biomarkers that can be used to describe conditions in the oral cavity. These biomarkers can be seen through saliva, mucosal tissue or oral fluids such as gingival cravicular fluid (GCF) in the oral cavity. These biomarkers can explain the current state of the oral cavity defense (Yoshizawa *et al.*, 2013) [9].

These marker proteins include interferon- γ (IFN)- γ , interleukin (IL)-1 β , IL-1 α , IL-2, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-12(p70), IL-13, IL-15, IL-17, soluble IL-2 receptor alpha (sIL-2R α), interferon inducible protein (IP)-10, tumor necrosis factor (TNF)- α , epidermal growth factor (EGF), monocyte chemotactic protein (MCP)-1, macrophage inflammatory protein (MIP)-1 β , soluble CD40 ligand (sCD40L), transforming growth factor (TGF)- α , vascular endothelial growth factor (VEGF), C-reactive protein (CRP), matrix metalloproteinase (MMP)-2, and MMP-9. This marker protein can be seen through several methods, namely ELISA (Enzyme-linked immunosorbent assay), PCR (Polymerase Chain Reaction) and Luminex Multiplex Immunoassay (Phalane *et al.*, 2013; Jacobs *et al.*, 2016; Kwan *et al.*, 2020) [7, 5, 4].

Management

The management of tuberculous lesions in the oral cavity is the same as for systemic tuberculosis. Currently, the most effective treatment requires a combination of four drugs namely isoniazid, rifampin, pyrazinamide, and ethambutol given daily for the first two months, followed by an additional four months with only two drugs namely isoniazid and rifampin (Khan, 2015) [2].

Discussion

From several studies, saliva has been used widely as one of the biomarkers to determine the early detection of infectious disease. Saliva has been reported in other disease, including leukaemia, oral cancer, oral lichen planus and several periodontal diseases. Saliva has been used as an HIV rapid test sample due to the marker which can detected the immune system. There are many advantages to use saliva in any other early detection. In several studies has been reported that saliva also can be used as the diagnostic sample in biomarker of TB. The high level of host markers in saliva showed that these markers may be more reliably measured even with lateral flow devices, which are usually easy to perform and suitable even for remote settings, but often have low detection limits. Although diagnostic tests based on markers of salivary inflammation may be less specific because levels of these markers may also be high in other diseases, markers may be useful when combined with clinical information. Such a test would be very useful in the field of TB diagnostics, given the difficulties associated with diagnosing TB disease when insufficient sputum is obtained and when there is an inability to expel sputum as is obtained in young children and in cases of extrapulmonary TB (Yoshizawa *et al.*, 2013) [9].

IL-2 is an important immunomodulatory cytokine produced by several cell types including activated T cells, dendritic cells, and NK cells and is essential both for the immune response to many infectious diseases and for the maintenance of tolerance. IL-2 has been investigated extensively especially in T-cell-based studies and has shown potential as a diagnostic marker for TB disease. IL-6 is a pleiotropic cytokine and has diverse effects on the regulation of immune responses, inflammation, oncogenesis, and hematopoiesis. IL-6 has previously been shown to be produced at higher levels in TB patients and was the only marker showing potential as a diagnostic biomarker in both serum and saliva samples (Phalane *et al.*, 2013) [7]. Fractalkins are induced in endothelial cells and antigen-presenting cells in the presence of various TH1-supporting signals including IFN- γ , CD40L, and TNF- α and are inhibited in the presence of IL-4/IL-13. MIP-1 β is mainly produced by macrophages, dendritic cells, NK cells, and T cells. It has been investigated as a diagnostic biomarker in T cell-based studies, but the potential shown in several adult studies was not confirmed in children. In the present study, MIP-1 β showed potential in the diagnosis of TB disease in saliva but not in serum. CRP, SAA, and SAP are acute phase proteins and are secreted during the acute phase of inflammation where they function as opsonins or in cell recruitment to sites of inflammation. These nonspecific markers of inflammation are known to be predominantly produced by the liver and have been studied in many diseases. EGF, basic fibroblast growth factor, insulin, and members of the insulin-like growth factor family have been detected in the salivary glands (Jacobs *et al.*, 2016; Phalane *et al.*, 2013) [7, 5].

Conclusion

Saliva is one of the samples which can be used to determine the immune system which occur in oral tuberculosis. Several biomarkers in saliva have been used as a tool to do the early detection of oral tuberculosis due to the easy sampling method and the high level of host markers in saliva showed that these markers may be more reliably as a sample in early detection of oral tuberculosis.

References

1. Fogel N. 'Tuberculosis: A disease without boundaries', *Tuberculosis*. Elsevier Ltd,2015;95(5):527-531. doi: 10.1016/j.tube.2015.05.017.
2. Khan M. 'Oral manifestations of Tuberculosis: The role of the dentist', *South African Dental Journal*,2015;70(10):434-435.
3. Kurniawati A, Mertaniasih NM, Agil M. 'Clinical Manifestation of Oral Tuberculosis', *UNEJ e-Proceeding*, 2017, 127-131.
4. Kwan PKW. *et al.* 'Gene expression responses to anti-tuberculous drugs in a whole blood model', *BMC Microbiology*. *BMC Microbiology*,2020;20(1):1-9. doi: 10.1186/s12866-020-01766-y.
5. Jacobs R, Maasdorp E, Malherbe S, Loxton AG, Stanley K, van der Spuy G, *et al.* Diagnostic potential of novel salivary host biomarkers as candidates for the immunological diagnosis of tuberculosis disease and monitoring of tuberculosis treatment response. *PLoS ONE*,2016;11(8):1-13. <https://doi.org/10.1371/journal.pone.0160546>
6. Pateria K. Oral Lesions of Tuberculosis- An Overview. *Journal of Orofacial & Health Sciences*, 2011, 2(2). <https://doi.org/10.5602/2229-3624.51>
7. Phalane KG, Kriel M, Loxton AG, Menezes A, Stanley K, van der Spuy *et al.* Differential expression of host biomarkers in saliva and serum samples from individuals with suspected pulmonary tuberculosis. *Mediators of Inflammation*, 2013. <https://doi.org/10.1155/2013/981984>
8. World Health Organization, *Global Tuberculosis Report*, 2020.
9. Yoshizawa JM, Schafer CA, Schafer JJ, Farrell JJ, Paster BJ, Wong DTW. Salivary biomarkers: Toward future clinical and diagnostic utilities. *Clinical Microbiology Reviews*, 2013;26(4):781–791. <https://doi.org/10.1128/CMR.00021-13>
10. Zhang D, Li X, Xiong H, Yang C, Lv F, Huang X, *et al.* Tuberculosis of the parotid lymph nodes: Clinical and imaging features. *Infection and Drug Resistance*,2018;11:1795-1805. <https://doi.org/10.2147/IDR.S164993>