Serum Iron, Total Iron Binding Capacity and Transferrin Saturation Levels in Leprosy Patients before Multi-Drug Therapy – World Health Organization (MDT-WHO) Compared with Healthy Control Group

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ABSTRACT Leprosy is an infectious disease caused by bacteria Mycobacterium leprae. This study aimed at investigating the Serum Iron, Total Iron Binding Capacity and Transferrin Saturation Levels on the leprosy patients compared with the healthy control group. This study is a continuation of previous research that has been studied Hemoglobin and Ferritin Serum Level on Leprosy Patient before Multi-Drug Therapy compared with Healthy Control Group. The study was conducted in Dr Wahidin Sudirohusodo Hospital and other hospitals in Makassar City from March to May 2018. The study used the analytic observational method with the case-control study design. Samples were the leprosy patients and the control group of 18 - 52 years old who came to be treated to the dermatovenereology of Dr Wahidin Sudirohusodo Hospital and other hospitals in Makassar City. As many as 40 samples consisted of 20 samples of the leprosy patients and 20 samples of the control group. This study shows that the iron and transferrin saturation levels in patients with leprosy are significantly lower compared to the healthy population. Future studies with larger samples are needed to confirm this relationship. Iron supplementation might be considered in leprosy patients with anaemia.

KEYWORDS leprosy, anaemia, iron, transferrin saturation

Introduction

Leprosy or Hansen's disease is a chronic granulomatous bacterial infection that develops slowly and is caused by the obligate intracellular bacteria Mycobacterium leprae. It mainly affects the peripheral nerves and may subsequently invade the skin, oral mucosa, upper respiratory tract, reticuloendothelial system, eyes, muscles, bones and testes except for the central nervous system. [1]

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Dermatoneurological signs and symptoms are the primary manifestations that fall in the spectrum of two stable poles (tuberculoid and lepromatous). For practical purposes, leprosy is divided into paucibacillary and multibacillary types. Transmission between humans is thought to be the primary mode of transmission, where the upper airway of the multibacillary patient (lepromatous and borderline) is the main route of bacteria, and into the upper airway, especially the nasal mucosa through contaminated droplets. The morphology of this germ is straight pleomorphic with both ends of round with a length of 1-8 microns and width of 0.2 to 0.5 microns, is acid-resistant, rod-shaped and gram-positive, usually in groups and there are scattered one-on-one, life in cells, especially tissues with cold temperatures such as skin, nasal mucosa, peripheral nerves, especially Schwann cells.[2]

All bacteria require a purine base from the nucleotides to make nucleic acids and also for oxidative metabolism. Unlike

other mycobacteria, M. leprae cannot synthesise and may obtain it from a host cell. Mycobacteria also require iron that they extract and procure it from the host through chelation with mycobactin. [3] Iron is indispensable in various metabolic processes such as oxygen transport, DNA synthesis and transport of electrons as essential nutrients. Iron is also imperative as an energy cofactor in mitochondrial respiration, proliferation and activation of T lymphocytes, B lymphocytes, and Natural Killer cells.

Intake of iron from host tissue plays a vital role in the development of pathogenic bacteria in vivo. The limited amounts of iron in the host trigger bacteria to take iron reserves from transferrin and lactoferrin, or ferritin. Mycobacteria produces an iron-binding molecule (siderophore) called mycobactin, exochelin, and carboxymycobactin, which then stores it in the form of bacterioferritin. Gram-positive mycobacteria tend to acquire iron-bound home rather than ferritin, in which heme is the largest iron-binding protein in the body (80%). Mycobacteria can prevent phagosome maturation, thus providing an appropriate environment for the bacteria to grow. [4]

Reduced iron reserves will decrease CMI functions such as delayed hypersensitivity, lymphocyte proliferation due to antigen stimulation, natural killer cytotoxicity and others. The presence of iron at the cellular level is required in the differentiation and proliferation of Th-1 and Th-2. Th-1 is more sensitive to antitransferin receptor antibody, with the result being a barrier to DNA synthesis. Th-2 is relatively more resistant to the administration of these antibodies. It is therefore suspected that Th-1-mediated function is more sensitive to iron hemostasis in the body. [5]

Chronic infectious diseases such as leprosy, in the development of the disease, can cause changes in the process of formation of erythrocytes (erythropoiesis) and the occurrence of anaemia which is called anaemia due to inflammation or better known as chronic anaemia disease. The clinical picture of chronic disease anaemia, often asymptomatic and covered by the clinical symptoms of the underlying disease, requires further evaluation. [6]

The most common anaemia in leprosy patients is mild to moderate anaemia, with normochromic to hypochromic normocytic characteristics in both tuberculoid and lepromatous leprosy patients. [7]

Clinical characteristics of anaemia, chronic disease is mild-moderate anaemia (Hb 7-11 g / dL), with normochromic to hypochromic microcytic normocytic morphology. Some patients may have severe anaemia. In chronic anaemia disease, there is a decrease in the number of reticulocytes and serum erythropoietin levels; Serum iron concentration can be normal until it decreases, total iron binding capacity (TIBC) decreases and serum ferritin can be normal until it rises. [5]

As far as the author's knowledge there are no studies have reported serum iron levels (Serum Iron, SI), total iron binding capacity (Total Iron Binding Capacity, TIBC) and transferrin saturation (Transferrin Saturation) in leprosy patients before WHO MDT-therapy, then compare with healthy control. For monitoring and assessing the results of the analysis, lepers were selected who met the criteria as samples according to WHO criteria and measured serum iron levels (Serum Iron, SI), total iron binding capacity (TIBC) and transferrin saturation (Transferrin Saturation) before therapy.

MATERIAL AND METHODS

Time and Place

The study was conducted from March to May 2018 and took place in the Dermatovenereology Department of Dr Wahidin Sudirohusodo Hospital, Hasanuddin University Hospital, Tadjuddin Khalid Hospital and other networking hospitals in Makassar, Indonesia.

Design Study and Variable

This is an analytic observational study with a case-control design. The independent variable is leprosy patients pre-MDT-WHO, and the dependent variables are the serum iron, Total Iron Binding Capacity (TIBC) and Transferrin Saturation.

Population and Sample

Previously untreated male and female patients with leprosy ageing from 16-60 years old who were willing to participate in this study by signing the informed consent form were included in the leprosy group. Leprosy patient who was pregnant had bleeding or experiencing other types of skin infections were excluded. Healthy participants ageing 16-60 years old who were willing to join the study were taken as the control group. Those with anaemia, Diabetes Mellitus, or history of leprosy in close relatives and family were excluded from the control group.

Data Collection

A blood specimen was collected to determine the Serum Iron, Total Iron Binding Capacity and Transferrin Saturation using ELFA technique. The data was then analysed and reported in the results.

Data Analysis

The collected data was presented in the form of tables and graphics. Data analysis was performed using SPSS version 22.0. Independent T-test and Mann Whitney test were done with a p-value of <0.05 considered as significant.

RESULTS

A total of 40 subjects (20 leprosies and 20 control) were enrolled in this study. Table 1 shows the baseline characteristics of both groups. There was no difference regarding sex and age in leprosy and control groups. Table 2 shows the comparison of iron level in both groups. Analysis using Mann-Whitney test indicates that the iron level in patients with leprosy was significantly lower compared to the control group ($p=0.000$). The TIBC (Total Iron Binding Capacity) in the leprosy group was found to be higher compared to the control group (Table 3). This difference, however, was not statistically significant ($p>0.05$). Subjects in the leprosy group showed a significantly lower transferrin saturation level compared to the control group ($p=0.000$).

DISSCUSION:

Leprosy is a chronic infection with bacteria Mycobacterium leprae as the infectious agent which mainly invades the skin and peripheral nerves. Other organs, such as the upper respiratory tract, reticuloendothelial and musculoskeletal system, eye, and testis may also be affected. Leprosy has a broad clinical spectrum associated with the immunologic response from the host. [8] Various chronic immunologic processes may affect the erythropoiesis process and lead to a specific type of anaemia

Table 1 Demographic Data

	Group		Total	p-value
	Leprosy	Control		
Sex				
Female	7(35%)	7(35%)	14(35%)	1.000
Male	13(65%)	13(65%)	26(65%)	1.000
Total	20(100%)	20(100%)	40(100%)	1.000
Age				
$\leq$ 20 years	2(10%)	2(10%)	4(10%)	1.000
21-30 years	6(30%)	6(30%)	12(30%)	1.000
31-40 years	6(30%)	6(30%)	12(30%)	1.000
>40 years	6(30%)	6(30%)	12(30%)	1.000
Total	20(100%)	20(100%)	40(100%)	1.000

Table 2 Iron Level in Both Groups

Group	n	Mean	SD	p-value
Leprosy	20	62,17	31,42	0,000
Control	20	106,10	26,87	

called anaemia of chronic disease. This study aims to explore the mechanism of anaemia in patients with leprosy by studying and comparing the hematologic profile and measuring the iron level in previously untreated leprosy patients and healthy controls.

Our data shows that the iron level in the leprosy group (62.10) was lower compared to the control group (106.10), which was found to be statistically significant ($p<0.01$). Iron is an essential element in homeostasis which functions to deliver oxygen, DNA synthesis, and energy metabolism. A defect in iron metabolism plays a major role in the pathogenesis of anaemia in chronic disease as a result of the effect of pro-inflammatory cytokines. It was found that TNF-α, IL-1, and IL-6 led to hypoferremia due to increased iron storage in the macrophage and ferritin synthesis. TNF- α will also inhibit iron release from macrophage and iron intake from erythroid cells. IL-6 induces the heptidin synthesis, a pleiotropic protein which is involved in iron metabolism. Heptidin impairs iron absorption in the intestine and iron release from the macrophage using ferroportin degradation. IL-10 upregulates the expression of transferrin receptor, resulting in increased transferrin uptake by monocytes. Also, other pro-inflammatory cytokines such as IFN- γ stimulates the formation of nitric oxide which in turn increases the activity of the iron regulatory protein (IRP)-1, a compound that controls ferritin formation in macrophage and transferrin receptor synthesis. Patients with anaemia of chronic disease are in a catabolic state that is characterised by low plasma albumin and transferrin levels. Impaired iron absorption and distribution were found, leading to shorter erythrocyte lifespan in such patients. [9]

The data in our study did not show any significant TIBC level difference between both groups, although the TIBC level was found to be higher in the leprosy group (318.35 compared to 312.02). A persistent iron deficiency leads to a depletion in the iron storage and impairs erythropoiesis. Consequently, there will be an abnormality in the morphology of erythrocyte; although clinically anaemia has not occurred. This condition is termed iron-deficient erythropoiesis. In this phase, a decrease in transferrin saturation and elevation in TIBC will typically be found. [4]

Table 3 TIBC Level Based on Group

Group	n	Mean	SD	p
Leprosy	20	318,35	74,04	0,925
Control	20	312,02	45,79	

Table 4 Transferrin Saturation Level Based on Group

Group	n	Mean	SD	p
Leprosy	20	0,20	0,08	0,000
Control	20	0,35	0,10	

Table 4 shows that the transferrin saturation level in leprosy group was significantly lower compared to the control group. An in vivo study showed that iron uptake from the host plays a pivotal role in the development and defence towards bacterial invasion. The limited amount of free iron in the human body causes to an increased need for bacteria towards iron, prompting them to take it from transferrin and lactoferrin, or ferritin. The bacteria M. leprae produces iron-binding molecule (siderophores) called mycobactin, excohelin, and carboxymycobactin, and stores them in the form of bacterioferritin. [4]

To the best of our knowledge, this is the first study to assess the iron profile in leprosy patients shown by the serum iron, TIBC, and transferrin saturation levels compared to the normal control group. Future studies should be conducted with larger sample size and consider possible comorbid diseases that might alter the iron profile. The result of this study may provide baseline data for recommending iron supplementation in leprosy patients with anaemia.

CONCLUSION:

This study shows that the iron and transferrin saturation levels in patients with leprosy are significantly lower compared to the healthy population. Future studies with larger samples are needed to confirm this relationship. Iron supplementation might be considered in leprosy patients with anaemia.

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Authors' Statements

Competing Interests

The authors declare no conflict of interest.

References

1. Kumar B, Kar HK. IAL Textbook of Leprosy: Jaypee Brothers, Medical Publishers Pvt. Limited; 2015.

2. Corrêa RdGCF, Aquino DMCd, Caldas AdJM, Amaral DKCR, França FS, Mesquita ERRBP-L. Epidemiological, clinical, and operational aspects of leprosy patients assisted at a referral service in the state of Maranhão, Brazil. Rev Soc Bras Med Trop. 2012;45:89-94.

3. Bryceson A, Pfaltzgraff RE. Leprosy: Churchill Livingstone; 1990.

4. Schaible UE, Kaufmann SH. Iron and microbial infection. Nature Reviews Microbiology. 2004;2(12):946.

5. Weiss G. Iron and immunity: a double-edged sword. Eur J Clin Invest. 2002;32 Suppl 1:70-8.

6. Goldsmith L, Katz S, Gilchrest B, Paller A, Leffell D, Wolff K. Fitzpatrick's Dermatology in General Medicine, Eighth Edition, 2 Volume set: McGraw-Hill Education; 2012.

7. Sen R, Yadav SS, Singh U, Sehgal P, Dixit VB. Patterns of erythropoiesis and anaemia in leprosy. Lepr Rev. 1991;62(2):158-70.

8. Jarduli LR, Sell AM, Reis PG, Sippert EÂ, Ayo CM, Mazini PS, et al. Role of HLA, KIR, MICA, and cytokines genes in leprosy. BioMed research international. 2013;2013.

9. Spivak JL. Iron and the anemia of chronic disease. Oncology (Williston Park). 2002;16(9 Suppl 10):25-33.