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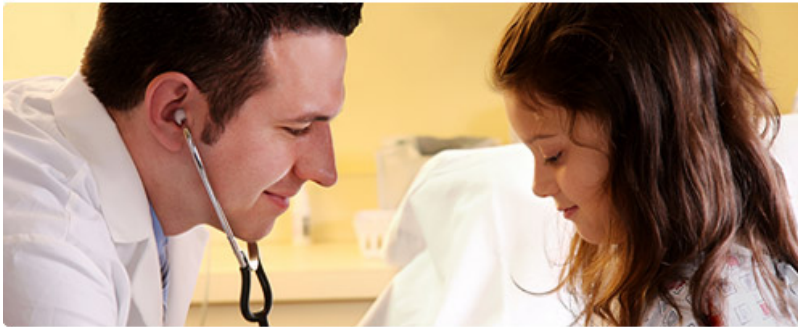
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Nutritional Status and Blood Profile amongst Patient with Child and Maternal Leprosy in Endemic and Non-Endemic Area of Indonesia

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Abstract

Leprosy remains endemic in several countries, where the disease is still considered as a health burden. The development of the disease is determined by several factors, amongst which that play significant roles are close household contact and impaired immunity. Maternal index case significantly associated with leprosy case in children and nutritional status plays a pivotal role in shaping the immune response against the bacteria *Mycobacterium leprae*. Thus, this paper aims to evaluate the association between nutritional status and leprosy, especially in maternal and child leprosy. The study was conducted in Tuban, Indonesia. We found significant differences in haemoglobin, red blood cells, and haematocrit levels in subjects with maternal leprosy in the group of child without leprosy and leprosy mother compared to the control group. The difference in haemoglobin and haematocrit levels are also associated with child leprosy in the group of child with leprosy and mother with leprosy. In addition, although no significant association on BMI was observed, we found that the child whose mother contracted leprosy has a lower BMI compared to the other groups.

Keywords: Leprosy; Nutritional status; Blood Profile; Endemic; Non-endemic

Introduction

Leprosy or Hansen's disease is a long-term infection caused by the bacteria *Mycobacterium leprae*.¹ The disease remains endemic in several countries despite the elimination program that has been arranged by the World Health Organization.² In Indonesia, one of the endemic countries that accounts for most of leprosy cases after Brazil and India, 17,439 new cases of leprosy were

detected in 2019. Amongst those, leprosy in children add up to 2,009 (11.52%) cases.² Due to the slow replicative nature of the *M. leprae* and thereby the long incubation period, leprosy is more common in adults than in children.³ Nevertheless, the high incidence of leprosy amongst paediatric population should prompt a warning of an unconfined disease transmission in the community.³

The most influencing factor in leprosy dissemination is close household contact with a person who contracts the disease.⁴ However, most persons who have been exposed to the bacteria would not contract the disease and the bacteria is thought to have low virulence.⁵ The causal relevance between risk factors and the development of the leprosy remains unclear due to the difficulty in

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evaluating and analysing the temporal relationship of the associated risk factors and the onset of the disease.⁶

Impairment of the immune response is known to augment the development of leprosy. Involved factors include variabilities in the gene that regulates inflammatory response and innate immunity;⁷ as well as non-genetic factors such as overall nutritional status and micronutrient intake.⁸ In addition, maternal index case was also associated with increased risk of nutritional deficiency in child.⁹

Previous findings showed the important relationship between nutritional status and leprosy. However, there remains a gap in the causal relationship. It is also stated that close household contacts with leprosy patient was amongst the most determining risk factor to develop leprosy and the position of women and their role within family increases the risk of leprosy transmission to their child. Therefore, this paper aims to evaluate the nutritional status amongst patients with leprosy, especially in maternal and child leprosy.

Material and Methods

Cross sectional design is used in this study. The study was done in endemic area and non-endemic area of Tuban regency, East Java, Indonesia in March 2020. Tuban regency is considered one of the leprosy pocket area in the country. In 2018, 173 new cases were detected in the regency, in which 5.81% of the total cases occurred in pediatric population and 43.35% cases affecting women.¹⁰

The population of this study include a pair of mother and her child who lives in endemic and non-endemic area. The inclusion criteria for subject with leprosy was those with confirmed diagnosis of leprosy and aged between 5-18 years old for children; whilst the excluded were those with poor general condition, any leprosy reaction, and diagnosed with inflammatory disorder including allergy, autoimmune, or infectious disease other than leprosy. Subject with pregnancy was also excluded. All of the subjects were given informed consent and have agreed to it. The ethical clearance for this study was approved by the Health Research Committee of Dr Soetomo General Hospital, Surabaya (1664/KEPK/XI/2019).

Subjects were divided into 5 groups (Figure 1). Group A was child with leprosy and mother without leprosy in an endemic area; group B was child without leprosy and mother with leprosy in an endemic area; group C were child with leprosy and mother with leprosy in an endemic area; group D was child without leprosy and mother without leprosy in an endemic area; group E was child without leprosy and mother without leprosy in non-endemic area (control group).

Leprosy cases were selected from the registry data of local primary health centre's. Thereafter, to confirm the diagnosis, each subject underwent clinical examination done by a dermatologist. Subjects also underwent skin smear that then examined microscopically with acid-fast staining by the laboratory professional from Tropical Disease Centre of Airlangga University.

Blood sample was taken from the subjects and underwent complete blood count including haemoglobin, red blood cells, white blood cells, platelets, haematocrit. Serum zinc level and albumin level were also measured. The equipment used to collect data were tourniquet, 3 mL syringe, vacutainer tube, alcohol swab, and adhesive plaster. Samples were collected in EDTA tube for haematological parameter and serum-separating tube for albumin. Haematological parameter were measured using Sysmex XN haematology analyser. Albumin were measured using Dimension EXL (Siemens). Zinc was measured using atomic absorption spectrophotometric method.

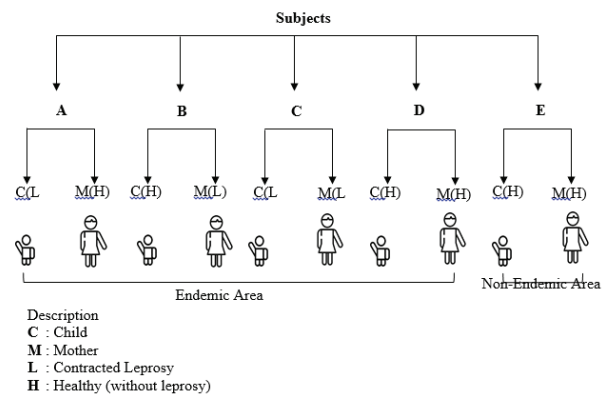


Figure 1. Schematic Figure of Group Allocation

Data were analysed using SPSS® software. Normality of the data distribution were assessed by Kolmogorov-Smirnov and Shapiro-Wilk tests. Thereafter, one-way analysis of variance (ANOVA) was used to analyse the

intergroup comparison and the p -value of <0.05 was considered as statistically significant.

Results and Discussion

Data were collected from 82 subjects, in which they were a pair comprised of mother and child subjects; 41 subjects for each of the age group. The subjects' characteristics from the child-aged subject and the mother are presented in Table 1 and Table 2, respectively.

Table 1. Characteristics of the child-aged subject. Data are presented as mean (standard deviation) or as a proportion (%)

	Group A (n= 10)	Group B (n= 10)	Group C (n= 2)	Group D (n= 11)	Group E (n= 8)	Reference range¹¹⁻¹⁵
Age, years	16.60 (\pm 2.63)	10.70 (\pm 4.76)	18.50 (\pm 0.70)	12.90 (\pm 4.32)	11.19 (\pm 3.71)	
BMI*	18.29 (\pm 3.86)	17.97 (\pm 3.31)	22.50 (\pm 1.83)	20.60 (\pm 5.70)	17.16 (\pm 4.10)	18 - 23
Blood Parameters						
Haemoglobin, g/dl	12.48 (\pm 2.86)	12.78 (\pm 0.98)	16.75 (\pm 0.35)	13.67 (\pm 1.11)	13.00 (\pm 0.35)	12.0 – 15.0
RBC* count, $\times 10^{12}/\mu\text{l}$	5.00 (\pm 0.86)	4.92 (\pm 0.60)	5.70 (\pm 0.28)	5.00 (\pm 0.35)	5.01 (\pm 0.54)	4.0 – 5.40
WBC* count, $\times 10^9/\mu\text{l}$	8.34 (\pm 2.42)	7.95 (\pm 3.10)	6.50 (\pm 0.56)	7.60 (\pm 2.21)	7.86 (\pm 2.19)	4.5 – 13.5
Platelet count, $\times 10^9/\mu\text{l}$	335.000 (\pm 68.39)	397.000 (\pm 76.51)	188.000 (\pm 131.52)	335.910 (\pm 76.87)	353.000 (\pm 0.35)	150 – 450
Haematocrit, %	38.52 (\pm 7.38)	37.90 (\pm 3.51)	49.00 (\pm 1.63)	40.75 (\pm 3.82)	39.05 (\pm 0.35)	35 – 49
Albumin, g/ dl	3.89 (\pm 0.31)	3.95 (\pm 0.86)	4.15 (\pm 0.35)	3.99 (\pm 0.13)	4.00 (\pm 0.10)	3.4 – 5.4
Zinc, $\mu\text{g}/\text{ml}$	4.11 (\pm 1.73)	5.00 (\pm 0.86)	5.43 (\pm 0.49)	3.83 (\pm 1.90)	3.26 (\pm 2.01)	6 – 8

*BMI= body mass index; RBC= red blood cells; WBC= white blood cells

Low BMI was observed in group B (child without leprosy and mother with leprosy in an endemic area) and group E (the control group; child without leprosy and mother without leprosy in non-endemic area). All parameters from the parameters blood count i.e. haemoglobin, RBC, WBC, platelet, and haematocrit showed normal value in all of the child-aged groups. However, low zinc serum concentration was observed in all of the groups (Table 1).

For the mother subjects, high BMI was observed amongst group A (child with leprosy and mother without leprosy in an endemic area), group B, and group D (child without leprosy and mother without leprosy in an endemic area). No abnormality was observed in all of the blood parameters, including the albumin level. Nevertheless, all of the groups showed marked low zinc serum concentration (Table 2).

Table 2.Characteristics of the mother subjects. Data are presented as mean (standard deviation) or as a proportion (%)

	Group A (n= 10)	Group B (n= 10)	Group C (n= 2)	Group D (n= 11)	Group E (n= 8)	Reference range11–15
Age, years	46.50 (± 6.48)	39.70 (± 7.36)	46.50 (± 2.12)	41.09 (± 11.27)	36.25 (± 6.62)	
BMI	25.25 (± 6.77)	24.42 (± 4.76)	22.43 (± 3.32)	25.42 (±4.54)	22.06 (± 1.80)	18 - 23
Blood Parameters						
Haemoglobin, g/dl	13.45 (± 1.16)	12.67 (± 1.19)	13.30 (± 1.69)	12.87 (± 1.34)	13.68 (± 0.49)	11.0 – 14.7
RBC count, x 1012/ µl	4.83 (± 0.52)	4.35 (± 0.51)	4.80 (± 0.28)	4.66 (± 0.48)	4.85 (± 0.33)	3.69 – 5.46
WBC count, x 109/ µl	8.51 (± 3.27)	8.74 (± 3.10)	7.35 (± 3.32)	7.94 (± 2.21)	7.57 (± 2.19)	3.37 - 10
Platelet count, x 109/ µl	299.500 (± 61.330)	302.400 (± 87.057)	371.500 (± 10.606)	326.909 (± 72.042)	297.125 (± 54.120)	150 - 450
Haematocrit, %	40.55 (± 3.64)	37.80 (± 2.68)	40.65 (± 6.15)	39.00 (± 3.82)	41.31 (± 2.20)	35.2 – 46.7
Albumin, g/ dl	3.85 (± 0.13)	3.85 (± 0.19)	3.95 (± 2.37)	3.83 (± 0.26)	3.90 (± 0.13)	3.4 – 5.0
Zinc, µg/ ml	4.46 (± 1.84)	3.99 (± 2.22)	3.95 (± 2.37)	4.15 (± 1.92)	4.09 (± 2.31)	8-12

The results of the statistical analysis amongst the child subjects showed a statistically significant difference on the haemoglobin between group C (child with leprosy and mother with leprosy in an endemic area) and group E. Significant difference was also observed on haematocrit parameter between group C and group E. For the mother

subjects, significant differences were observed between group B (child without leprosy and mother with leprosy in an endemic area) and group E on the haemoglobin ($p=0.038$), RBC ($p=0.029$), and haematocrit ($p=0.009$). No notable difference was observed on BMI parameters in any of the groups.

Table 3. Statistical analysis of the intergroup comparison of the child subjects

Parameters	Group Comparison				
	A : E	B : E	C : E	D : E	(A+B+C+D) : E
BMI	$p= 0.557$	$p= 0.762$	$p= 0.121$	$p= 0.165$	$p= 0.269$
Haemoglobin	$p= 0.619$	$p= 0.555$	$p<0.001$	$p= 0.119$	$p= 0.754$
RBC	$p= 0.648$	$p= 0.740$	$p= 0.132$	$p= 0.987$	$p= 0.879$
WBC	$p= 0.671$	$p= 0.792$	$p= 0.428$	$p=0.801$	$p= 0.906$
Platelet	$p= 0.535$	$p= 0.283$	$p= 0.065$	$p= 0.656$	$p= 0.783$
Haematocrit	$p= 0.849$	$p= 0.440$	$p= 0.001$	$p= 0.282$	$p= 0.743$
Albumin	$p= 0.355$	$p= 0.441$	$p=0.270$	$p= 0.874$	$p= 0.505$
Zinc	$p= 0.186$	$p= 0.481$	$p= 0.186$	$p= 0.541$	$p= 0.273$

Table 4. Statistical analysis of the inter-group comparison of the mother subjects

Parameters	Group Comparison				
	A : E	B : E	C : E	D : E	(A+B+C+D) : E
BMI	$p= 0.215$	$p= 0.156$	$p= 0.826$	$p= 0.065$	$p= 0.127$
Haemoglobin	$p= 0.557$	$p= 0.762$	$p= 0.121$	$p= 0.165$	$p= 0.269$
RBC	$p= 0.926$	$p= 0.029$	$p= 0.850$	$p= 0.361$	$p= 0.252$
WBC	$p= 0.467$	$p= 0.346$	$p= 0.880$	$p=0.554$	$p= 0.440$
Platelet	$p= 0.933$	$p= 0.883$	$p= 0.101$	$p= 0.340$	$p= 0.542$
Haematocrit	$p= 0.611$	$p= 0.009$	$p= 0.787$	$p= 0.187$	$p= 0.138$
Albumin	$p= 0.425$	$p= 0.545$	$p=0.627$	$p= 0.533$	$p= 0.596$
Zinc	$p= 0.344$	$p= 0.706$	$p= 0.940$	$p= 0.506$	$p= 0.718$

In this study, the BMI amongst child subjects showed no significant difference compared to the control groups. However, previous study found that BMI is associated with leprosy as lower BMI increase

the chance of contracting leprosy, especially amongst women.¹⁶In addition, a study done in an endemic area of leprosy in India found that 3%–48% child with leprosy suffer from malnutrition.¹⁷ Lower BMI generally

reflects poor nutritional status and this condition could lead to impairment of the immunity, as nutrients are needed in the regulation of immune response, especially in children.¹⁸

The insignificant difference on BMI parameter in our study could be influenced by other factors. We observed that subjects in the group E (control group) have the lowest BMI compared to the other groups. Leprosy patients in Indonesia obtain a logistic help from the government, which include ferrous sulphate, antioxidants, and other vitamins. In contrast, the subjects without leprosy from the control group (group E) did not receive any logistic help from the government.¹⁹

We also observed that children from mother with leprosy (group B) tend to have a lower BMI compared to other groups. Our finding is in line with previous evidence that observed that female index case significantly associated with nutritional status of the child in the same household.⁹ In addition, three groups of the mother subjects (group A, B and D) showed a higher mean of BMI, implying an overweight state. Previous study also found that an overweight state is common amongst leprosy patients, affecting 61% population with leprosy.²⁰ The relationship between overweight and leprosy is thought to be multifactorial. It is affected by sedentary lifestyle and activity or work limitation amongst leprosy patients, as well as unhealthy dietary intake that resulted in excessive caloric intake.²⁰

In this study, we found a significant difference on haemoglobin parameter both in the child (group C) and mother subjects (group B) compared to the control groups ($p < 0.001$ and $p = 0.038$; for group C and group B, respectively). This finding is in accordance with previous study, in which leprosy patient has a significantly lower haemoglobin levels compared to the healthy control groups.²¹ Low haemoglobin level could reflect a state of micronutrient deficiency, mainly iron deficiency.²²

Although the subjects in this study did not have an anemia, current evidence showed that patient with leprosy tend to have a lower serum iron concentration.²³ Low iron serum level may be observed in the chronic disease such as leprosy, that result from impaired iron-transport protein metabolism. Proinflammatory cytokines, mainly interferon- γ (IFN- γ) in leprosy, could stimulate the production of divalent metal transporter-1

(DMT-1) thereby increasing intracellular iron storage. The high concentration of the cytokine could also lead to decrease in Ferroportin-1 (Fpn-1) that serves as iron transporter from the reticuloendothelial cells to the erythroid progenitors. Both of the process lead to reduced iron utilization in erythroid cell lines and a decrease in haemoglobin synthesis.²⁴

Haemoglobin concentration amongst the child subjects, however, was observed to be lower in the control group compared to the group C (child with leprosy and mother with leprosy in an endemic area). This finding could be influenced by several factors. First factor is that subjects with leprosy (including those within group C) get logistics aid from the government as stated earlier. In addition, the higher hemoglobin level in group C could also be caused by the fewer participants in the group C.

The RBC and hematocrit of the mother subjects showed notable difference ($p = 0.029$ and $p = 0.009$, respectively) between group B and the control group. We also found a difference ($p = 0.001$) in child subjects between group C and the control group. Prior evidence found that leprosy patients receiving multi drug therapy (MDT) which included dapsone have a marked decrease in RBC concentration and in their hematocrit level.²⁵ Dapsone, one of the drugs used in MDT regimen is known to cause hemolytic anemia, particularly in the patient with glucose-6-phosphate dehydrogenase (G6PD) deficiency. The hemolysis of the red cells involve an oxidative stress induced by the hydroxylamine compound derived from the drug.²⁶ In addition, the decrease in the RBC amongst leprosy patient could be caused by the high levels of proinflammatory cytokines such as tumor necrosis factor- α (TNF- α) and IFN- γ . Both cytokines are able to inhibit erythropoiesis, in which TNF- α acts via inhibition of erythropoietin receptor and thereby leading to proliferation inhibition of the erythroid progenitors; and IFN- γ acts through induction of nitric oxide production that lead to apoptosis of the erythroid cell lines.²⁴ Regardless the evidence, in this study we also observed that the RBC and hematocrit in the group C was higher compared to the control group that could be attributed to the reason we mentioned earlier (i.e. logistic help from the government and fewer participants).

We found no significant difference regarding zinc between any of the group compared to the control group. Previous study found that zinc level amongst leprosy patients, especially lepromatous leprosy patients, is significantly lower compared to the general population.²⁷ Zinc is known to play a crucial role in immune response. Zinc deficiency could lead to poor Th1 response that impair immune response against intracellular pathogen including *M. leprae*.⁶ Regardless the unremarkable difference in the plasma zinc concentration amongst different groups in this study, we found that all of the groups, both in child and mother subjects, have a mean concentration of zinc below the normal reference range. The exact prevalence of zinc deficiency in Indonesia is unknown, however, two studies in school-aged children in Semarang, Indonesia (number of participants were 70 and 40) found that all of the subjects had a serum zinc concentration below the level recommended by the International Zinc Nutrition Consultative Group (IZiNCG) for developing country.²⁸

There is no notable difference in albumin level amongst any of the group compared to the control. In contrast, prior evidence showed that there is a depletion of plasma albumin concentration in patient with leprosy.²⁹ However, apart from the inadequate macronutritional intake, depletion in albumin level mostly caused by the inflammatory process of the disease. In leprosy, chronic inflammation depletes albumin level by decreasing its synthesis rate and increasing protein catabolism.³⁰ In other words, in regards to temporal aspect, albumin deficiency reflects a complication of the disease rather than a risk factor, especially when there is no malnutrition observed. This is supported by the fact that the subjects did not have a low BMI.

Conclusion

The difference in haemoglobin, RBC, and haematocrit levels are associated with maternal leprosy amongst child without leprosy and mother with leprosy. The difference in haemoglobin and haematocrit level are also associated with child leprosy in the same group. In addition, although no significant association on BMI were observed, we found that the child whose mother contracted leprosy has a lower BMI compared to the other groups. However, there is a limitation to this study due to the sample size of the study. Therefore, further

study with larger number of subjects are needed to confirm our findings.

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