Peripheral Blood Smear Eosinophil Differential Count Comparison In Patients With Skabies And After One Week Recovered From Scabies

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Abstrak

Skabies adalah infeksi kulit yang disebabkan infestasi dan sensitisasi tungau sarcoptes scabiei, yang mudah menular dari orang ke orang. Respons imun berperan penting menyebabkan komplikasi sekunder dan morbiditas pada individu dengan skabies. Eosinophil penting dalam pertahanan terhadap parasit namun juga berperan menyebabkan disfungsi dan kerusakan jaringan pada penyakit – penyakit alergi. Penelitian ini bertuuan membandingkan hitung jenis eosinophil darah tepi pasien – pasien saat menderita skabies dan setelah 1 minggu sembuh dari skabies. Penelitian ini menggunakan metode analitik komparatif dengan rancangan kohort. Besar sampel 30 subjek dihitung dengan menggunakan rumus besar sampel penelitian analitik kategorik – numerik. Analisis data menggunakan Uji Wilcoxon karena data tidak terdistribusi normal. Hasil penelitian ini menunjukkan mean hitung jenis eosinophil darah tepi pasien – pasien saat menderita skabies 3,6% (3% - 5%) sedangkan setelah 1 minggu sembuh dari skabies 2,37% (2% - 3%). Terdapat perbedaan yang signifikan antara hitung jenis eosinophil darah tepi pasien – pasien saat menderita skabies dan setelah 1 minggu sembuh dari skabies dengan *p value* <0.05.

Kata kunci: hitung jenis; skabies; eosinophil; darah tepi; kohort

Abstract

Scabies is a skin infection disease caused by infestation and sensitization of the mite Sarcoptes scabiei, easily transmitted from person to person. Immune response has a crucial role in secondary complications and morbidity of individuals with scabies. Eosinophil is essential in defense against parasites but also contribute to tissue dysfunction and damage in allergic diseases. The purpose of this study was to compare the peripheral blood eosinophils differential count of patients while suffering from scabies and one week after recovered from scabies. This is an analytic comparative study with a cohort design. Sample size of 30 subjects calculated with sample size formula for paired categorical-numeric analytic research. Wilcoxon test was performed for data analysis due to the data not normally distributed. The result of this study showed the mean peripheral blood differential eosinophil count of patients while suffering from scabies was 2,37%(2% - 3%). There was signicant difference in the peripheral blood differential eosinophils count between patients while suffering from scabies and one week after recovered from scabies with a p value <0.05.

Keyword: differential count; scabies; eosinophil; peripheral blood; cohort

Introduction

World Health Organization (WHO) in 2017 declared scabies as one of Neglected Tropical Disease (NTD) in need of large scale intervention. Prevalence of scabies is estimated between 0,2 % up to 71% with 5%-10% prevalence among children. This disease affects 200 million people world wide with the highest number of infestation in tropical nations of high population density limited healthcare access (1). In Indonesia, according to the data released by the Indonesian Ministry oh Health in 2018, the prevalence of scabies was 6% of the total Indonesian population (2).

Secondary complications and morbidity of scabies are often considered as the direct consequences of scabies infestation. However, these complications are more accurately correspond with the massive itch reaction caused by immune responds towards the mite and the direct interaction between complement inhibitor produced by the mite and the complement system of the skin which facilitate bacterial growth. Moreover, scratching may cause defects in the skin epidermal barrier (3).

Various native effector cells that detected as immune respond towards *S. scabiei* mite are eosinophils, mast cells, basophils, neutrophils, dendritic cells and makrophages. Eosinophil is a key player in the defence against parasite but it also contributes in tissue dysfunction and disruption in allergic diseases (4).

Parasitic infection is the most common cause of eosinophilia worldwide. eosinophil is known to produce *eosinophil extracellular traps* (EETs) and releases *toxic protein granule* in the scabies skin lesion which contribute in clearing this pathogen. The *epithelial barrier* disruption and epidermal proliferation that resembles atopic dermatitis and psoriasis may also be found in scabies. Eosinophil may also play a role in the occurrence of bullous scabies in several case reports. In addition, it is thought that the intense itching in scabies is caused by eosinophil (5).

Based on the above, we feel it is necessary to conduct a study on the differences in peripheral blood smear eosinophil counts among patients when they are suffering from scabies and after one week of recovery from scabies.

Method

The method of this study is paired comparative analytical study with cohort design that compares peripheral blood eosinophil counts in patients while suffering from scabies and after 1 week of recovery from scabies. The research was carried out from June 2022 to March 2023 at Pusat Kesehatan Masyarakat Kecamatan Singkil Utara (North Singkil Public Health Center), Aceh Singkil Regency. Blood samples were collected and examined at Pusat Kesehatan Masyarakat Singkil Utara laboratory, Aceh Singkil Regency. Samples were taken twice in the patient group when they were suffering from scabies and after 1 week of recovery from scabies. Sampling was only carried out once in the control group.

The sample size in this study was determined using the paired numerical – categorical analytical study sample size formula, and determined a sample size of 30 people in the patient group and the same number in the control group with a total sample size of 60 research subjects (6).

The patient group subjects were obtained from patients who sought treatment at Pusat Kesehatan Masyarakat Singkil Utara and were diagnosed with scabies by finding 2 of the 4 cardinal signs, namely; itching that worsened at night, the infection affects people in groups, the lesions are in the form of straight lines tunnels or curves with an average length of 1 cm and at the end of the tunnel papules or vesicles were found, and the discovery of mites (7).

The control group subjects were individuals who did not present abnormalities nor other diseases that could affect the results of the peripheral blood eosinophil count in the initial examination.

The independent variables in this research are; currently suffering from scabies, after 1 week of recovery from scabies, and controls (healthy individuals). Meanwhile, the dependent variable in this study is the peripheral blood eosinophil count.

Peripheral blood smear eosinophil differential count is the relative number of eosinophils in the peripheral blood smear leukocyte differential count. differentiating the types of leukocytes was done visually using a microscope on a peripheral blood smear made on a glass slide with Giemsa staining, in this case the number of a type of leukocyte is expressed in (%) of 100 leukocytes (all types). Eosinophils are recognized by their similar shape and size to neutrophils, but the cytoplasm is filled with large red granules, and the granules are the same size. The normal reference value for eosinophil count is 1 - 3% (8).

Peripheral blood smears were taken by researcher in the clinic room of the Pusat Kesehatan Masyarakat Kecamatan Singkil Utara. Fixation, staining and leucocyte differential counting were carried out by analysts at Pusat Kesehatan Masyarakat Singkil Utara laboratory. Mann – Whitney U test was used to compare eosinophil counts between the control group and the patient group while suffering from and after 1 week of recovery from scabies because the data was not normally distributed. Meanwhile, the Wilcoxon test was used to compare the peripheral blood smear eosinophil differential count between patient groups while suffering from scabies and after 1 week of recovery from scabies because the data was not normally distributed (9).

Result

The characteristics of the control group and patients suffering from scabies in this study did not differ much. Most of the research subjects were < 20 years old, 25 subjects (83.3%) in the control group and 23 subjects (76.7%) in the patient group. In both groups, the largest proportion was high school educational status, in the control group there were 11 subjects (37%) while in the group of patients suffering from scabies there were 10 subjects (33%). In both groups, the majority were students, namely in the control group 18 subjects (60%) while in the group of patients suffering from scabies 21 subjects (70%). Based on gender, the control group was equal between men and women, namely 15 subjects each (50%), while in the group of patients suffering from scabies, the majority were men, namely 21 subjects (70%).

Table 1. Minimum, maximum, and *mean* result of peripheral blood smear eosinophil counts in patients while suffering from scabies, peripheral blood smear eosinophil counts patients 1 week after recovery from scabies, and peripheral blood smear eosinophil counts of healthy subjects in the control group.

Group	Minimum (%)	Maximum (%)	Mean (%)
Peripheral blood smear eosinophil counts in patients while suffering from scabies	3	5	3,60
peripheral blood smear eosinophil counts patients 1 week after recovery from scabies	2	3	2,37
peripheral blood smear eosinophil counts of healthy subjects in the control group	1	3	1,70

The minimum peripheral blood smear eosinophil counts in patients suffering from scabies was 3%, the maximum count was 5% with an average count of 3.60%. The minimum peripheral blood smear eosinophil counts for patients after 1 week of recovery

from scabies was 2 %, the maximum count was 3% with an average value of 2.37%. The minimum peripheral blood smear eosinophil counts for healthy people in the control group was 1%, the maximum count was 3% with an average count of 1.70%.

Table 2. Peripheral blood smear eosinophil counts comparison result using Mann – Whitney U Test between control group and patients while suffering from scabies.

Test	Statistics ^a
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	Eosinophil
Mann-Whitney U	13.000
Wilcoxon W	478.000
Ζ	-6.675
Asymp. Sig. (2-tailed)	.000

a. Grouping Variable: group

Result of the Mann – Whitney U test comparing the peripheral blood eosinophil count between the control group and the group of patients while suffering from scabies was significant with p-value of 0.000, less than <0.05, which means there was a significant difference between the peripheral blood smear eosinophil counts of the control group and the group of patients while suffering from scabies.

Table 3. Wilcoxon Test result comparing peripheral blood smear eosinophil counts between patients while suffering from scabies and after 1 week recovery from scabies.

Test Statistics ^a		
	Peripheral blood smear eosinophil count patients while suffering from scabies -	
	Peripheral blood smear eosinophil count patients after 1 week recovery from scabies	
Ζ	$-4,500^{b}$	
Asymp. Sig.	,000	
(2-tailed)		
a. Wilcoxon Sigr	ned Ranks Test	
b. Based on nego	ative ranks.	

Wilcoxon test result comparing peripheral blood smear eosinophil counts between patients suffering from scabies and after 1 week of recovery from scabies showed a significant p-value of 0.000, less than < 0.05, which means there was a significant difference between peripheral blood smear eosinophil counts of patients while suffering from scabies and the same patients after recovered from scabies for 1 week.

Table 4. Mann–Whitney U Test result comparing peripheral blood smear eosinophil counts between control group and patients after 1 week recovery from scabies.

Test Statistics^a

	Eosinophil
Mann-Whitney U	210.500
Wilcoxon W	675.500
Ζ	-4.041
Asymp. Sig. (2-tailed)	.000

a. Grouping Variable: Group

The result Mann – Whitney U Test comparing peripheral blood smear eosinophil counts between the control group and the patient group after 1 week of recovery from scabies showed significant p-value of 0.000, less than <0.05, which means there was significant difference between peripheral blood smear eosinophil counts of the control group and the patient group after 1 week recovery from scabies.

Discussion

The demographic characteristics in this study between the group of patients suffering from scabies and the control group of healthy people were relatively similar except for gender. The demographic make ups was in accordance with the general characteristics of patients seeking treatment at Pusat Kesehatan Masyarakat Singkil Utara in terms of age, education level and occupation. The proportion of gender was balanced in the control group because the control group sampling was carried out first and deliberately balanced between male and female, whereas the group of patients suffering from scabies was dominated by males.

There was no research that shows that gender can influence the results of peripheral blood smear eosinophil counts in patients with scabies. Although several literatures that examined eosinophil levels in patients with asthma, chronic obstructive pulmonary disease, and asthma - COPD overlap found that the average eosinophil levels of patients with these diseases were higher in male patients compared to female patients (10).

The diagnosis of scabies is usually made based on clinical symptoms. There is no standard laboratory test to diagnose scabies. Antigen and antibody immunoassays have been evaluated with suboptimal results, and neither is widely used (11).

Atypical clinical features of scabies such as chronic excoriation and skin eczema are often found in patients with long-standing infestation. Subtle cases are difficult to diagnose, especially if topical or systemic steroids have been given to mask the itching and inflammation of scabies. Accurate diagnosis of scabies is very important to prevent more serious consequences, for example the spread of infestation and severe infections that can be life-threatening (12).

Peripheral blood smear eosinophil count is not a routine examination for scabies. The role of eosinophils in scabies infection has attracted the interest of many researchers looking for a relationship between eosinophils and scabies infection, but none of these studies have examined peripheral blood smear eosinophil counts. The peripheral blood smear leukocyte differential count to assess eosinophils is a relatively simple examination, because it only requires Giemsa staining and a microscope thus can easily be carried out in first-level health facilities such as public health centers (Pusat Kesehatan Masyarakat).

The result of peripheral blood smear eosinophil counts in this study for patients while suffering from scabies were significantly higher compared to the group of healthy subjects as control. This is in line with a prospective study in Australia that examined absolute eosinophil levels in patients with crusted scabies finding eosinophilia in 53 of 88 cases (60%) with 33 of 88 cases having eosinophil levels of more than 1.0 $\times 10^6$ mmol/L (13).

In this study, the peripheral blood smear eosinophil count of patients while were suffering from scabies was significantly higher than the peripheral blood eosinophil count when the patients had recovered from scabies for 1 week, which had reached normal values, although when compared with peripheral blood smear eosinophils count in the control group of healthy people were significantly higher. This means the decrease in peripheral blood eosinophil counts is corresponds with clinical improvement in patients with scabies.

A case report in 2020 described hypereosinophilia in a 3 month old baby with skin lesions of papules, pustules, accompanied by eosinophilia (absolute eosinophil count

15,253 cells/µL) which was initially treated with 2 times administrations of whole body permethrin 5% that resulted no improvement for 3 weeks. The baby was then treated with sulfur precipitation ointment for 3 consecutive nights and repeated the following week. White blood cell level, including eosinophils returned to normal after 2 months and remained normal at examination 4 months later. This case report shows that the diagnosis of scabies should be considered in the presence of hypereosinophilia and atypical skin lesions, even though the patient had been treated. Eosinophilic leukemoid reaction indicates an immune system reaction to fight mite infestation and peripheral eosinophilia is an important sign of suspected scabies (14).

A case report of an 80-year-old woman in Japan who was initially diagnosed with papuloerythroderma and hypereosinophilia of 5100 cells/mm³. The patient was suspected of suffering from chronic eosinophilic leukemia, but no FIP1L1-PDGFRA gene fusion was found on peripheral blood examination. On examination of the crusts on the soles of the feet, mites and eggs were found, so the patient was then diagnosed as suffering from crusted scabies. In this case the improvement in skin symptoms after treatment was in line with the eosinophil count from peripheral blood examination. The case report also suggests checking the cause of eosinophilia if it is found along with the deckchair sign which is a skin symptom of papyloerythroderma (15).

Conclusion and recommendation

The peripheral blood smear eosinophil count in patients suffering from scabies was higher than in healthy people, and decreased significantly after 1 week of recovery from scabies. Based on this, the researchers concluded that peripheral blood smear eosinophil count is a simple test that can help health workers in first-level health facilities such as public health centers (Pusat Kesehatan Masyarakat) to diagnose and assess recovery from scabies.

The researcher suggests for further broader research on scabies regarding factors other than eosinophils. In addition, more inclusive diagnostic criteria are needed to avoid underdiagnoses of scabies.

References

- 1. World Health Organization. Scabies [internet]. (Updated 23 May 2023; cited 8 August 2023). Available from: <u>https://www.who.int/news-room/fact-sheets/detail/scabies</u>
- Kementerian Kesehatan Republik Indonesia. Profil Kesehatan Indonesia 2018 [internet]. (Updated June 2019; cited 8 August 2023). Available from: <u>https://www.kemkes.go.id/downloads/resources/download/pusdatin/profil-kesehatan-indonesia/profil-kesehatan-indonesia-2018.pdf</u>
- 3. Ständer S, Ständer S. Itch in Scabies—What Do We Know?. Front. Med. 2021;8:628392. doi: 10.3389/fmed.2021.628392
- 4. Trasia R F, Sari I P. Respon imun hospes terhadap Sarcoptes scabiei. J Biomedika Kesehat. 2020; 3(4):183-192 DOI: 10.18051
- Radonjic-Hoesli, S. Brüggen, M. Feldmeyer, L. Simon, H. Simon, D. Eosinophils in skin diseases Semin Immunopathol. 2021;43:393–409 <u>https://doi.org/10.1007/s00281-021-00868-7</u>
- 6. Dahlan M S. Besar Sampel dan Cara Pengambilan Sampel dalam Penelitian Kedokteran dan Kesehatan. Jakarta. Salemba Medika. 2010.
- Putri, Y Y. Astuti, R D I. Bharata, T. Karakteristik Tanda Kardinal Penyakit Skabies Pada Santri di Pesantren. Jurnal Integrasi Kesehatan dan Sains. 2020; 2(2): 126 – 129. DOI: <u>https://doi.org/10.29313/jiks.v2i2.5716</u>
- 8. Nugraha, G. Badrawi, I. Pedoman Teknik Pemeriksaan Laboratorium Klinik Untuk Mahasiswa Teknologi Laboratorium Medik. Trans Info Media Jakarta. 2018.
- University Of Bristol. Which test? [internet]. (updated 18 March 2023; cited 8 August 2023). Available from: https://www.bristol.ac.uk/medicalschool/media/rms/red/which_test.html
- Luo, J. Chen, H. Feng, Y. Chen, Y. Lei, X. Guo, S. et al. Blood Eosinophil Endotypes across Asthma and Chronic Obstructive Pulmonary Disease (COPD). 2022. https://doi.org/10.1155/2022/9656278
- Chandler, D J. Fuller, L C. A Review of Scabies: An Infestation More Than Skin Deep. Dermatology. 2019; 235:79–90. doi: <u>10.1159/000495290</u>
- Herwanto, N. Mappasing, H. Widiantari, S. Indranarum, T. Hidayati, A N. Sawitri, Ervianti, E. Murtiastutik, D. Martodihardjo, S. Scabies incognito Dermatology Reports; 11(s1):8083. <u>https://doi.org/10.4081/dr.2019.8083</u>
- 13. Hasan, T. Krause, V L. James, C. Currie, B J. Crusted Scabies; a 2-year prospective study from the Northern Territory of Australia. PLoS Negl Trop Dis. 2020; 14(12): e0008994. https://doi.org/ 10.1371/journal.pntd.0008994
- Sanchez-Flores, X. Cancel-Artau, K J. Figueroa, L. Eosinophilia with leukemoid reaction secondary to Sarcoptes scabiei JAAD Case Reports. 2021; 8:13-5. <u>https://doi.org/10.1016/j.jdcr.2020.12.002</u>
- 15. Nakamura K. A Case of Papuloerythroderma Secondary to Crusted Scabies. J Cutan Immunol Allergy. 2019;2:174–175.