Clinical Research

Erythrocyte and iron profiles in soil-transmitted helminth-infected children in a rural area in Banten, Indonesia
Saleha Sungkar¹, Ninik Sukartini², Aulia Wirastuti², Dian Wahyu Tanjungsari³

ABSTRACT

BACKGROUND Soil-transmitted helminth (STH) infection, frequently caused by Ascaris lumbricoides, Trichuris trichiura, and hookworms (Ancylostoma duodenale and Necator americanus), often gives rise to malabsorption of nutrients that form hemoglobin (Hb) thereby causing anemia. This study aimed to know the difference in erythrocyte profile in children with and without STH infections.

METHODS This cross-sectional study involved 205 children from two Panimbang Jaya elementary schools in Pandeglang, Banten, Indonesia. Blood and stool samples were collected from a previous study conducted from November 2021 to May 2022. Erythrocyte parameters were Hb concentration, erythrocyte count, mean corpuscular volume (MCV), mean corpuscular Hb (MCH), and red cell distribution width (RDW). The iron profile included serum iron, total iron-binding capacity (TIBC), and ferritin. Worm infestation was detected by direct stool microscopical examination. Statistical analysis was performed using SPSS software version 20.

RESULTS The prevalence of STH infection in Pandeglang was 44.4%, primarily characterized by mild intensity STH infection (79%). The identified STH species were A. lumbricoides, T. trichiura, and combination of both. The median differences between erythrocyte count, MCV, and MCH, and the mean differences of TIBC and serum iron were not statistically significant (p = 0.388, 0.098, and 0.057, and p = 0.304 and 0.455). However, children with STH infection had lower Hb (12.57 versus 12.95 g/dl) and ferritin (19.60 versus 30.57 µg/dl) levels but higher RDW (13.20 versus 13.10%).

CONCLUSIONS A high prevalence of STH infection was identified among schoolchildren, but their erythrocyte profiles were similar regardless of STH infection status.

KEYWORDS anemia, Ascaris lumbricoides, children, erythrocytes, Trichuris trichiura

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Studies have reported an association between anemia and worm infestation in children; however, most studies have only analyzed anemia based on the patient’s Hb levels. This study aimed to determine the differences in erythrocyte profiles between school-aged children with and without STH infections living in a rural area of Indonesia.

METHODS

This cross-sectional study was conducted at two Panimbang Jaya elementary schools in Pandeglang, Banten, Indonesia. Blood and stool examinations were conducted using previous sample collections. The samples were obtained from November 2021 to May 2022.

Children aged 6–12 years (in grades 1–6) willing to participate (with parental permission) were included in this study. Children who were severely ill or had high fever were excluded from the study. Verbal and written informed consents were obtained from the children’s guardians prior to data collection. The feces were collected in 10-ml plastic pots that were collected the next day. Venous blood samples of 0.5–3 ml were also collected.

Hematological examinations, including the Hb level, erythrocyte count, mean corpuscular volume (MCV), mean corpuscular Hb (MCH), and RDW, were performed using a Sysmex XN-3000 hematology analyzer (Sysmex Europe, Germany). The patients’ iron status (serum iron, TIBC, and ferritin levels) was examined using an Abbott Architect c8000 (Abbott, US). Anemia was defined based on the World Health Organization Hb cut-off level of <11.5 g/dl for ages 5–11 years and <12 g/dl for ages ≥12 years.4 Iron deficiency was defined as a serum ferritin level <15 ng/ml and/or a serum iron level <30 μg/dl with a TIBC >410 μg/dl.4 IDA was defined as anemia with low ferritin levels based on the hallmark of absolute iron deficiency.7 The feces were examined macroscopically (color and consistency) and microscopically using lugol and eosin wet mount staining. To minimize human error, the microscopic examinations were performed by two technicians. The Kato-Katz method was also used to count eggs to determine the intensity of infection.8 The examination was conducted at the Department of Clinical Pathology, Cipto Mangunkusumo Hospital.

Variables are presented as percentages and blood parameters as mean (standard deviation) or median (min–max). The data were analyzed using the unpaired t-test and Mann–Whitney U test. All statistical analyses were conducted using SPSS software version 20.0 (IBM Corp., USA). The α was set at 0.05, and 95% confidence intervals were determined. Statistical significance was set at p<0.05. Ethical approval was obtained from the Ethics Committee of the Faculty of Medicine, Universitas Indonesia, with approval number (No: 295/UN2.F1/ETIK/PPM.00.02/2019).

RESULTS

Of 205 children included in this study, 114 had STH infection. Thirty-eight children were infected with A. lumbricoides, 31 with T. trichiura, and 45 with both STHs. Most STH infections were detected at mild intensity (79%), and 21% of the study population had high intensity of infections. Of the 26 children with anemia, 16 had STH infection, and 25% had IDA. Iron deficiency was identified in 28 children (31%) with STH infection, while the Hb, RDW-coefficient of variation, and ferritin levels were slightly higher in children without STH infection. No other parameters were different between children with and without STH infections. Characteristics of all participants and blood parameters are shown in Table 1.

DISCUSSION

In this study, the prevalence of STH infection was 44.4% and higher in females. This may be secondary to open defecation on the beach due to a lack of fecal disposal facilities or bathing, washing, and toilet facilities, which are risk factors for the transmission of worm infections. In addition, low economic levels lead to low purchasing power for clean water, further increasing the risk of worm transmission.

A total of 18% of children with intestinal worm infections had anemia in this study. However, Sungkar et al9 reported a higher prevalence of anemia (71.3%) in children with worm infections in Perobatang, Sumba, East Nusa Tenggara. The previous study identified a hookworm infection rate of 11%; therefore, the proportion of patients with intestinal worm infection and anemia was higher.9 Hookworms cause the greatest blood loss among all STHs. However, no hookworm infections or infections of severe intensity were observed in this study.
A. lumbricoides was the most common species identified in this study. However, in a previous study conducted in Ende, East Nusa Tenggara, T. trichiura was the most common infection (20.6%). The difference in worm species proportions may be due to geographical conditions, humidity, soil properties, adequate bathing, washing, and toilet facilities, and clean and healthy living behaviors of the participants. Hookworms require sandy, loose soil that is shielded from direct sunlight. The absence of hookworms in the present study may be due to unsuitable environmental conditions for hookworms to thrive in Panimbang Jaya village. In addition, low intensity parasite infection, early or chronic phases of infection, or improvement in students’ personal hygiene may account for the absence of hookworms in this study.

Based on the high prevalence of worm infections (>20%), annual mass treatment of school-aged children is necessary. Community health centers can provide treatment by providing worm medicine to children at schools. Triple doses of albendazole or mebendazole can treat T. trichuria infections and have comparable efficacies. However, mebendazole is associated with more severe side effects than albendazole, including generalized edema and the release of worms through the mouth. Therefore, the government advises the use of triple-dose albendazole for mass treatment as it tends to be safer.

The average Hb levels of the infected and non-infected groups in this study were within the reference range, possibly due to the mild intensity of the worm infections. However, the Hb levels and RDW were significantly different between the groups. The difference in the Hb levels was not accompanied by a significant difference in the number of erythrocytes, MCV, or MCH values, possibly due to reduced Hb formation in patients with infection due to a lack of iron. Hb is the main component of erythrocytes. Therefore, the number of erythrocytes cannot be used to indicate normal Hb levels. Additionally, parameters such as MCV and MCH, which describe the volume and morphology of erythrocytes, do not always indicate an individual’s anemia status. The median RDW value of children with intestinal worm infection was significantly higher than that of children without infection in this study.

### Table 1. Characteristics and hematological profiles of the participants

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Infected (N = 91)</th>
<th>Not infected (N = 114)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>School grades, n (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>14 (50)</td>
<td>14 (50)</td>
<td></td>
</tr>
<tr>
<td>II</td>
<td>10 (48)</td>
<td>11 (52)</td>
<td></td>
</tr>
<tr>
<td>III</td>
<td>14 (52)</td>
<td>13 (48)</td>
<td></td>
</tr>
<tr>
<td>IV</td>
<td>11 (41)</td>
<td>16 (59)</td>
<td></td>
</tr>
<tr>
<td>V</td>
<td>19 (38)</td>
<td>31 (62)</td>
<td></td>
</tr>
<tr>
<td>VI</td>
<td>23 (44)</td>
<td>29 (59)</td>
<td></td>
</tr>
<tr>
<td>Sex, n (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>35 (40)</td>
<td>52 (60)</td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>56 (47)</td>
<td>62 (53)</td>
<td></td>
</tr>
<tr>
<td>Anemia, n (%)</td>
<td>16 (18)</td>
<td>10 (9)</td>
<td>0.06</td>
</tr>
<tr>
<td>Hb level (g/dl), mean (SD)</td>
<td>12.57 (0.88)</td>
<td>12.95 (0.90)</td>
<td>0.001*</td>
</tr>
<tr>
<td>Erythrocyte count (×10⁶/µl), median (min–max)</td>
<td>4.95 (3.85–6.55)</td>
<td>5.00 (4.03–6.56)</td>
<td>0.388'</td>
</tr>
<tr>
<td>MCV (fl), median (min–max)</td>
<td>77.90 (57.70–91.20)</td>
<td>78.70 (59.70–89.60)</td>
<td>0.098'</td>
</tr>
<tr>
<td>MCH (pg), median (min–max)</td>
<td>25.60 (17.60–30.40)</td>
<td>26.20 (19.20–30.00)</td>
<td>0.057'</td>
</tr>
<tr>
<td>RDW-CV (%), median (min–max)</td>
<td>13.20 (11.70–18.90)</td>
<td>13.10 (11.60–18.60)</td>
<td>0.038'</td>
</tr>
<tr>
<td>Serum iron (µg/dl), mean (SD)</td>
<td>81.28 (24.90)</td>
<td>83.91 (25.22)</td>
<td>0.455*</td>
</tr>
<tr>
<td>TIBC (µg), mean (SD)</td>
<td>336.36 (51.85)</td>
<td>329.52 (43.21)</td>
<td>0.304*</td>
</tr>
<tr>
<td>Ferritin (µg/dl), median (min–max)</td>
<td>19.60 (4.85–81.16)</td>
<td>30.57 (8.51–117.20)</td>
<td>&lt;0.001'</td>
</tr>
</tbody>
</table>

Hb=hemoglobin; MCH=mean corpuscular hemoglobin; MCV=mean corpuscular volume; RDW-CV=red cell distribution width-coefficient of variation; SD=standard deviation; TIBC=total iron-biding capacity
*Unpaired t-test; †Mann–Whitney U test
These results align with research by Sazawal et al., who reported that RDW was the first hematological parameter to increase in an iron-depleted state, even before anemia occurred. A higher RDW indicates a wider size variation of the erythrocytes. RDW increases if the value is greater than 14.5%. However, the present study did not observe an increased RDW in children with worm infections, possibly due to the mild and moderate intensities of the infections in this study. Therefore, the blood loss was not overwhelming and did not trigger a significant increase in erythropoiesis, and the RBC size did not vary significantly.

The ferritin levels were significantly different between the groups in this study, though the TIBC and serum iron levels were not. Darlan et al. reported no differences in the serum iron levels or TIBC between children with and without parasitic infections. High ferritin levels have been observed in children with STH infection. Darlan et al. hypothesized that good nutritional and immunity statuses might protect against parasites. Moreover, serum iron and TIBC change during stage II of iron deficiency. The circulating iron levels bound to transferrin are influenced by the timing of sample collection, intestinal absorption, and vitamin C intake. Ideally, the iron level assessment should be performed under fasting conditions for accurate results. In the present study, the children did not fast, which led to inaccurate high serum iron levels. Collecting blood samples after iron-rich food intake might also have contributed to this high result. Moreover, there was no difference in TIBC between the groups in this study, which is consistent with the results reported by Darlan et al. TIBC represents the total capacity of transferrin to bind to iron. High TIBC indicates an increased need for iron due to iron deficiency.

Limitations of this study included the unequal random distribution of variables, such as class, grade, and sex. Therefore, the results of the statistical analyses were not as expected. In conclusion, the prevalence of STH infection in schoolchildren in Pandeglang was high (44.4%). Lower Hb levels and higher RDW values were observed in children with worm infection than in those without worm infection, which may contribute to the development of anemia if the infection remains untreated. The high incidence of STH infection in schoolchildren living in rural areas underscores the need for health promotion and programs to decrease the prevalence and eliminate the disease in high-risk populations, including schoolchildren.

**Conflict of Interest**
Saleha Sungkar is the editorial board member but was not involved in the review or decision-making process of the article.

**Acknowledgment**
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