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The Effect of Iron Supplementation and Anthelmintic Administration on Hemoglobin Levels in Adolescent Girls at SMP Advent 2 Medan

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ABSTRACT

Adolescent girls may experience low hemoglobin because of inadequate iron availability and intestinal helminth infection. This study evaluated hemoglobin changes after iron supplementation and anthelmintic administration, individually and in combination, among girls at SMP Advent 2 Medan, Indonesia. A quasi-experimental, four-group pretest–posttest study was conducted from August to October 2026. Forty girls were allocated to combined iron–folic acid and albendazole, iron–folic acid alone, albendazole alone, or control groups, with 10 participants each. Supplementation provided 60 mg elemental iron and 0.4 mg folic acid twice weekly for three months; albendazole was administered as a single 400 mg dose on day 1. Hemoglobin was measured using HemoCue. Paired t-tests, Welch ANOVA, and Games–Howell comparisons were performed. Five participants (12.5%) were anemic at baseline. Mean hemoglobin changes were 1.43 ± 0.71 g/dL with combined treatment, 1.12 ± 1.29 g/dL with iron alone, 0.03 ± 0.34 g/dL with albendazole alone, and −0.21 ± 0.16 g/dL in controls. Changes differed overall (Welch $F(3, 17.071) = 18.962$; $p < 0.001$). Combined treatment exceeded albendazole alone and control (both $p < 0.001$), while iron alone exceeded control ($p = 0.040$). Combined treatment did not differ significantly from iron alone (difference, 0.31 g/dL; 95% CI, −1.04 to 1.66; $p = 0.908$). Iron-containing regimens improved hemoglobin, but adding albendazole was not demonstrated to provide an additional benefit. Interpretation is limited by the small, predominantly nonanemic sample and unconfirmed helminth and iron-deficiency status.

Keywords: hemoglobin, adolescent girls, iron supplementation, folic acid, albendazole, anemia

INTRODUCTION

Hemoglobin carries oxygen through reversible binding to the iron-containing heme groups in its globin subunits. Normal adult hemoglobin predominantly contains two alpha and two beta chains, each with a heme group. Adequate iron availability and coordinated erythropoiesis are essential to hemoglobin synthesis and tissue oxygen delivery [1], [2], [3]. Reduced hemoglobin can cause fatigue and limit physical capacity, affecting adolescents' health and daily activities [4].

Adolescent girls are vulnerable to anemia because rapid growth increases nutrient requirements and menstruation can add recurrent blood loss. The World Health Organization identifies menstruating adolescent girls among the populations particularly affected by anemia [5]. Dietary intake may fail to meet this increased demand, especially when the quantity or bioavailability of iron is insufficient [6], [7]. However, anemia has multiple causes, and a low hemoglobin concentration alone does not establish iron deficiency; the contribution of nutritional deficiency, blood loss, inflammation, and inherited disorders requires consideration [8].

Iron tablets provide an accessible means of increasing the iron available for erythropoiesis. Reviews focused on adolescent girls have reported improvements in hemoglobin and iron status after supplementation, although the response varies with the regimen and participants' initial condition [9], [10]. A meta-analysis of 129 randomized trials also supports the hematological benefits of oral iron while emphasizing differences related to supplementation schedule, duration, dose, and accompanying nutrients [11]. In school programs, consistent consumption is important because access to tablets does not necessarily translate into sustained intake.

Intestinal helminth infection provides a second plausible pathway to impaired hematological status. Soil-transmitted helminths may reduce nutrient availability, and hookworm infection can cause chronic intestinal blood loss [12], [13]. In Indonesia, helminth control has been addressed through public-health regulations, including the Ministry of Health's 2017 regulation [14]. A systematic review and meta-analysis found lower hemoglobin levels and a higher risk of anemia among children with soil-transmitted helminth

infections, supporting attention to infection when nutritional interventions are considered [15].

Anthelmintic treatment targets parasitic infection rather than directly supplying the substrates required for hemoglobin synthesis. Albendazole, a benzimidazole anthelmintic, disrupts parasite microtubule function and energy metabolism [16], [17]. Combining deworming with iron supplementation could therefore address both inadequate iron availability and infection-related losses when these conditions coexist [13], [18]. Whether this complementary biological rationale produces an additional measurable hemoglobin benefit must be established through comparisons with iron supplementation alone.

This study compared hemoglobin changes among adolescent girls at SMP Advent 2 Medan receiving iron–folic acid plus albendazole, iron–folic acid alone, albendazole alone, or no study intervention. It assessed within-group changes and between-group differences, particularly whether adding albendazole improved the response to iron supplementation.

RESEARCH METHODS

Study design and participants

A quantitative, quasi-experimental study with a four-group pretest–posttest design was conducted at SMP Advent 2 Medan, Indonesia, from August to October 2026. The school's eligible female-student population comprised 40 adolescents. Total population sampling was used, and all 40 participants were included. After recruitment, participants were allocated to four groups of 10 using simple random allocation by lottery or random numbers, as specified in the study protocol.

Eligibility required female students aged 12–15 years who attended the school, were willing to participate, and had permission from a parent or guardian. Exclusion criteria were a history of hematological or chronic disease, current use of iron supplements or other medication affecting hemoglobin, and failure to complete the intervention sequence. The analyzed participants were aged 12–14 years. Age, school grade, intervention group, and hemoglobin measurements were recorded for analysis.

Intervention procedures

Baseline hemoglobin was measured before treatment. The combined group received tablets containing 60 mg elemental iron and 0.4 mg folic acid twice weekly for three consecutive months, together with a single oral dose of albendazole 400 mg on the first intervention day. The iron-only group received the same iron–folic acid regimen without albendazole. The albendazole-only group received a single 400 mg dose on day 1. Controls received neither study medication during follow-up. These regimens are summarized in Table 1. The intermittent iron schedule corresponds to the adolescent regimen in the IDAI recommendation cited in the study background [19].

Table 1. Intervention regimens over the three-month study period

Group	n	Iron–folic acid supplementation	Albendazole
Combined	10	60 mg elemental iron + 0.4 mg folic acid, twice weekly for 3 months	400 mg orally once, day 1
Iron only	10	Same iron–folic acid regimen	None
Albendazole only	10	None	400 mg orally once, day 1
Control	10	None	None

"Iron only" denotes iron–folic acid supplementation without albendazole. All groups underwent baseline and follow-up hemoglobin measurement.

Medication administration was recorded on observation sheets to support adherence monitoring. After the intervention period, hemoglobin was measured again using the same method and device. No numerical adherence rate, treatment-related adverse-event count, or additional hematological endpoint was available for analysis. Consequently, the results assess the observed hemoglobin response rather than comparative tolerability or changes in iron stores.

Hemoglobin measurement and outcomes

Hemoglobin concentration was measured with a HemoCue device and expressed in g/dL. The principal outcome was the individual change in hemoglobin, calculated as post-intervention minus baseline concentration. Positive values indicated an increase. Baseline anemia was classified using hemoglobin <12.0 g/dL, whereas concentrations ≥12.0 g/dL were categorized as nonanemic. Hemoglobin interpretation was informed by the laboratory and hematology references used in the study background [20], [21]. This classification describes hemoglobin status and does not identify the cause of anemia.

Statistical analysis

Data were edited, numerically coded, entered, and checked for inconsistencies before analysis in SPSS. Continuous variables were summarized as mean ± standard deviation, and categorical variables as frequencies and percentages. Shapiro–Wilk tests assessed the distribution of hemoglobin change within each group. Paired t-tests evaluated within-group changes. Because the mean-based Levene test indicated unequal variances, Welch analysis of variance was used for the overall comparison of changes, followed by Games–Howell pairwise comparisons. Mean differences and 95% confidence intervals were taken from the corresponding statistical output. Tests were two-sided, with $p < 0.05$ considered statistically significant.

Ethical considerations

The study protocol was approved by the Health Research Ethics Committee (Komisi Etik Penelitian Kesehatan, KEPK), Universitas Prima Indonesia, Medan, Indonesia (Approval No. 076/KEPK/UNPRI/VII/2026, dated 22 July 2026). The approval was valid from 22 July 2026 to 22 July 2027.

The Faculty of Medicine and Dentistry, Universitas Prima Indonesia, issued a formal request for permission to conduct the study to the principal of SMP Advent 2 Medan (Letter No. 028/SPH/F1.1.E/UNPRI/VII/2026, dated 24 July 2026). Participation required the adolescent's willingness to take part and permission from a parent or guardian. Participant names and identifying information are omitted; results are presented as group summaries.

RESULT AND DISCUSSION

Participant characteristics

The analysis included 40 adolescent girls, with 10 participants in each group. Fifteen participants (37.5%) were aged 13 years, 14 (35.0%) were aged 14 years, and 11 (27.5%) were aged 12 years. Grade VII accounted for 16 participants (40.0%), followed by grade VIII with 14 (35.0%) and grade IX with 10 (25.0%). At baseline, five participants (12.5%) had hemoglobin <12.0 g/dL, while 35 (87.5%) were nonanemic (Table 2). Thus, enrollment was not restricted to girls with anemia.

Table 2. Participant characteristics at enrollment

Characteristic	Category	n (%)
Age	12 years	11 (27.5)
	13 years	15 (37.5)
	14 years	14 (35.0)
School grade	VII	16 (40.0)
	VIII	14 (35.0)
	IX	10 (25.0)
Baseline hemoglobin status	Anemic (<12.0 g/dL)	5 (12.5)
	Nonanemic (≥12.0 g/dL)	35 (87.5)

N = 40. Each intervention group contained 10 participants (25.0% of the sample).

Changes within intervention groups

Mean baseline hemoglobin ranged from 12.88 ± 1.60 g/dL in the combined group to 14.45 ± 1.43 g/dL in the albendazole-only group. The combined group increased to 14.31 ± 1.09 g/dL, corresponding to a mean change of 1.43 ± 0.71 g/dL (95% CI, 0.92 to 1.94; p < 0.001). The iron-only group increased from 13.78 ± 2.61 to 14.90 ± 1.55 g/dL, a mean change of 1.12 ± 1.29 g/dL (95% CI, 0.20 to 2.04; p = 0.022).

Hemoglobin was essentially unchanged after albendazole alone: 14.45 ± 1.43 g/dL at baseline and 14.48 ± 1.26 g/dL at follow-up, with a mean change of 0.03 ± 0.34 g/dL (95% CI, −0.21 to 0.27; p = 0.787). Controls decreased from 14.16 ± 1.74 to 13.95 ± 1.68 g/dL, a mean change of −0.21 ± 0.16 g/dL (95% CI, −0.32 to −0.10; p = 0.002). Table 3 summarizes the measurements and paired comparisons. These tests describe change within each group and do not themselves establish differences between interventions.

Table 3. Hemoglobin concentrations and within-group changes

Group	Baseline (g/dL)	Follow-up (g/dL)	Change (g/dL)	95% CI for mean change	Paired p-value
Combined	12.88 ± 1.60	14.31 ± 1.09	1.43 ± 0.71	0.92 to 1.94	<0.001
Iron only	13.78 ± 2.61	14.90 ± 1.55	1.12 ± 1.29	0.20 to 2.04	0.022
Albendazole only	14.45 ± 1.43	14.48 ± 1.26	0.03 ± 0.34	−0.21 to 0.27	0.787
Control	14.16 ± 1.74	13.95 ± 1.68	−0.21 ± 0.16	−0.32 to −0.10	0.002

Values are mean ± SD; n = 10 per group. Change = follow-up minus baseline hemoglobin. CI, confidence interval. Paired t-tests have 9 degrees of freedom. Confidence intervals use the post-minus-pre direction.

Comparisons between groups

Shapiro–Wilk p-values for hemoglobin change were 0.563, 0.165, 0.344, and 0.609 in the combined, iron-only, albendazole-only, and control groups, respectively. The mean-based Levene statistic was 5.421 (p = 0.004), indicating unequal variances. Welch ANOVA identified an overall difference in mean change between groups, F(3, 17.071) = 18.962, p < 0.001.

Games–Howell comparisons showed greater increases with the combined regimen than with albendazole alone (difference, 1.40 g/dL; 95% CI, 0.67 to 2.13; p < 0.001) or control (1.64 g/dL; 95% CI, 0.93 to 2.35; p < 0.001). Iron supplementation alone also differed from control (1.33 g/dL; 95% CI, 0.06 to 2.60; p = 0.040). The combined and iron-only groups did not differ significantly (0.31 g/dL; 95% CI, −1.04 to 1.66; p = 0.908). Neither iron versus albendazole nor albendazole versus control reached statistical significance (Table 4).

Table 4. Games–Howell comparisons of mean hemoglobin changes

Comparison	Difference (g/dL)	95% CI (g/dL)	p-value
Combined vs. iron only	0.31	−1.04 to 1.66	0.908
Combined vs. albendazole only	1.40	0.67 to 2.13	<0.001
Combined vs. control	1.64	0.93 to 2.35	<0.001
Iron only vs. albendazole only	1.09	−0.19 to 2.37	0.103
Iron only vs. control	1.33	0.06 to 2.60	0.040
Albendazole only vs. control	0.24	−0.11 to 0.59	0.231

Difference = mean change in the first group minus mean change in the second. Confidence intervals and p-values are from Games–Howell comparisons. Overall Welch ANOVA: $F(3, 17.071) = 18.962$, $p < 0.001$.

Principal findings and biological interpretation

Iron-containing regimens were associated with increased hemoglobin over three months. The combined regimen produced the largest numerical increase, followed by iron alone, whereas albendazole alone produced little average change. The variance-robust analysis supports differences between combined treatment and both albendazole alone and control, and between iron alone and control. However, adding albendazole did not demonstrate an additional benefit over iron alone.

The response in the iron-containing groups is biologically plausible. Iron transported to erythroid precursors supports heme synthesis and the assembly of functional hemoglobin. When iron supply is inadequate, erythropoiesis is constrained; increasing iron availability can improve hemoglobin production [8], [22]. Nevertheless, hemoglobin alone is not a specific marker of iron deficiency. Clinical hematology distinguishes anemia from its underlying cause through additional laboratory and clinical assessment [23]. In this study, neither baseline iron depletion nor replenishment of iron stores was demonstrated, so the findings should be described as changes in hemoglobin rather than confirmed correction of iron-deficiency anemia.

Dietary conditions and consistent intake may also influence the observed response. Iron absorption varies with the form of dietary iron, physiological iron requirements, and accompanying foods; enhancers and inhibitors can alter its bioavailability [24]. The study recorded intervention administration, but did not provide quantitative adherence or dietary data. These unmeasured factors may help explain differences in response among participants. Both iron-containing regimens also included folic acid, meaning that their effects cannot be attributed to elemental iron independently of the administered formulation.

Comparison with supplementation evidence

The findings are consistent with the direction of the adolescent supplementation evidence summarized by Silitonga et al. [9] and Nanda et al. [10]. Those reviews support improved hemoglobin after iron supplementation, while the present study extends the comparison to separate iron-containing and anthelmintic regimens within one school. Direct comparisons of effect magnitude require caution because the reviews included different baseline anemia profiles and supplementation schedules. In particular, the review by Nanda et al. focused on adolescents with anemia, whereas only 12.5% of participants in the present study were anemic at enrollment.

Andersen et al. synthesized 129 randomized trials and reported hematological benefits of oral iron across heterogeneous supplementation approaches [11]. Their findings support the biological rationale for the present response, but do not establish that the specific twice-weekly regimen used here is optimal. The current study did not compare alternative doses, frequencies, or durations. Its findings therefore concern the implemented three-month school-based regimen and should not be extended to untested schedules or to all adolescent populations.

Deworming and the combined regimen

The minimal change after albendazole alone warrants interpretation in relation to infection status. Anthelmintics can remove a cause of ongoing blood loss when susceptible intestinal helminths are present, but do not directly replace depleted iron [12], [13]. The meta-analysis by Alnaz et al. linked helminth infection, particularly hookworm and multiple infections, with lower hemoglobin and increased anemia risk [15]. Without confirmation of the presence, species, or intensity of infection in this study, the extent to which such a pathway was active among participants remains unknown.

The deworming literature also contains differing population-level findings. Girum and Wasie reported benefits for anemia-related outcomes in schoolchildren [25], whereas the Cochrane review by Taylor-Robinson et al. found no consistent hemoglobin benefit from routine public-health deworming across children living in endemic areas [26]. The lack of a detectable albendazole effect here is compatible with variability in the infection burden and nutritional characteristics of treated populations. It does not establish that treatment of confirmed helminth infection is ineffective.

The combined regimen exceeded albendazole alone and control, but these comparisons cannot isolate the contribution of albendazole because the combined group also received iron–folic acid. The relevant incremental comparison is combined treatment versus iron supplementation alone. Its 95% confidence interval crossed zero and allowed both a lower and a higher mean response with combination treatment. Thus, the nonsignificant result provides neither evidence of superiority nor proof of equivalent effectiveness. The study also did not establish a synergistic interaction between the interventions.

Clinical relevance and study limitations

The predominance of nonanemic participants is central to clinical interpretation. A rise in average hemoglobin across a mixed population does not establish the same treatment effect among girls with confirmed anemia. Baseline hemoglobin was also lower in the combined group than in the other groups, creating more potential for an increase. Change-score comparisons do not fully resolve the influence of this initial imbalance, and no baseline-adjusted treatment estimate was reported. The small decline in controls reached statistical significance, but its cause cannot be assigned to menstruation, diet, or infection because these factors were not measured in sufficient detail.

Several limitations restrict generalization. The sample comprised 40 students from one school, with only 10 in each group, resulting in imprecise pairwise estimates. The protocol used total population sampling and described simple random allocation, but allocation concealment and blinding were not documented. Menstrual blood loss, nutritional status, dietary intake, and confirmed helminth infection were not characterized. Ferritin and other iron-status measurements were unavailable, and quantitative adherence and adverse-event data were not reported. These gaps limit causal interpretation, assessment of anemia mechanisms, and comparisons of safety or treatment acceptability.

Future studies should recruit larger samples and characterize baseline anemia, iron status, and parasitic infection before comparing the regimens. Follow-up assessment of adherence, menstrual losses, diet, and nutritional status would improve interpretation of individual responses. Baseline-adjusted analyses and more precise confidence intervals would help determine whether adding an anthelmintic offers a clinically useful benefit beyond iron–folic acid alone. School-based education on nutrition and personal hygiene remains relevant to the broader prevention approach described in the study, but the present findings do not justify universal combined treatment solely to obtain a greater hemoglobin increase.

CONCLUSION

Hemoglobin increased after three months of iron–folic acid supplementation, alone or combined with albendazole. Overall changes differed significantly across groups. Combined treatment exceeded albendazole alone and control, and iron alone exceeded control. Albendazole alone did not differ significantly from control. Combination treatment was not demonstrated to be superior to iron alone. Interpretation should account for the small, predominantly nonanemic sample, baseline differences, and unconfirmed helminth and iron-deficiency status.

Further school-based studies should assess anemia etiology, confirm helminth infection, and monitor adherence alongside hemoglobin. Larger comparative studies are needed before an added hematological benefit of combining albendazole with iron–folic acid can be established.

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