



Association of iron and nutritional status with sleep quality in adolescent girls: A cross-sectional study

Hubungan status besi dan status gizi dengan kualitas tidur pada remaja putri: Studi potong lintang

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Abstract

Adolescent girls tend to have a higher risk of anemia because their nutritional demands increase during growth and the onset of menstruation. Both anemia and nutritional status have been proposed to affect sleep quality through various physiological pathways. Therefore, this study was designed to examine the relationship between ferritin and hemoglobin levels and nutritional status and how these factors are associated with sleep quality among adolescent girls. A cross-sectional study will be conducted in April–May 2025 with a total of 80 participants from Pekalongan Regency. Information on sleep quality was obtained using the Pittsburgh Sleep Quality Index (PSQI) questionnaire, ferritin levels were examined using the Enzyme-Linked Immunosorbent Assay (ELISA), and hemoglobin levels were assessed using a hematology analyzer. To explore the relationships among ferritin levels, hemoglobin, and nutritional status with sleep quality, a multivariate regression analysis was employed. Bivariate analysis revealed that hemoglobin levels ($p = 0.005$) and nutritional status ($p = 0.007$) were significantly associated with sleep quality, whereas ferritin levels were not ($p = 0.054$). In the multivariate analysis, only hemoglobin level was significantly associated with sleep quality ($p < 0.001$). These results indicate that lower hemoglobin levels may play a role in worsening sleep quality among adolescent girls, underscoring the need to promote anemia prevention through enhanced nutritional intake.

Keywords: iron deficiency anemia, sleep quality, adolescent girls, nutritional status

Abstrak

Remaja putri rentan mengalami anemia akibat peningkatan kebutuhan zat gizi dan menstruasi. Anemia dan status gizi diduga berhubungan dengan kualitas tidur melalui mekanisme yang berkaitan dengan transportasi oksigen dan regulasi neurotransmitter. Penelitian ini bertujuan untuk menganalisis hubungan kadar ferritin, hemoglobin, dan status gizi dengan kualitas tidur pada remaja putri. Desain penelitian cross-sectional dilaksanakan pada bulan April–Mei 2025 dengan melibatkan 80 remaja putri di Kabupaten Pekalongan. Data kualitas tidur dikumpulkan menggunakan kuesioner Pittsburgh Sleep Quality Index (PSQI), kadar ferritin dianalisis menggunakan metode Enzyme-Linked Immunosorbent Assay (ELISA), dan kadar hemoglobin diukur menggunakan analisis hematologi. Analisis regresi multivariat dilakukan untuk mengevaluasi hubungan antara kadar ferritin, hemoglobin, dan status gizi dengan kualitas tidur. Analisis bivariat menunjukkan bahwa kadar hemoglobin ($p = 0.005$) dan status gizi ($p = 0.007$) berhubungan signifikan dengan kualitas tidur, sedangkan kadar ferritin tidak ($p = 0,054$). Pada analisis multivariat, hanya kadar hemoglobin yang tetap berhubungan signifikan dengan kualitas tidur ($p < 0.001$). Hal ini menunjukkan bahwa kadar

hemoglobin yang rendah dapat berkontribusi terhadap penurunan kualitas tidur pada remaja putri, sehingga penting untuk mencegah anemia melalui peningkatan gizi.

Kata Kunci: anemia defisiensi zat besi, kualitas tidur, remaja putri, status gizi

Introduction

Adolescent girls enter a pivotal stage of development marked by swift biological, psychological, emotional, and social changes. This phase tends to heighten their susceptibility to a range of health problems shaped by lifestyle factors, including inadequate dietary patterns and low levels of physical activity (Moore, 2023; Dhamayanti et al., 2019; Ding et al., 2024). During adolescence, nutritional needs increase sharply to support rapid growth and reproductive maturation. When these requirements are not fulfilled, adolescents face a greater likelihood of developing nutritional deficiencies that can affect their overall well-being, including reproductive health, cognitive development, and future productivity. Among these deficiencies, anemia is one of the most widespread nutritional challenges globally. Adolescent girls are particularly susceptible because their iron and micronutrient needs increase due to with menstruation and accelerated physical growth (Tijerina et al., 2015; Zhu et al., 2021).

The impacts of anemia involve a diminished ability to transport oxygen, leading to persistent fatigue, resulting in to have poorer academic performance, and contributing to disruptions in neurocognitive functioning (Mantadakis et al., 2020). Globally, the prevalence of anemia among adolescents aged 10–19 years is estimated to be approximately 29.9%, with even higher proportions reported in low- and middle-income countries (WHO, 2025). In Indonesia, the prevalence has reached 31.2% and is even higher in Pekalongan Regency, where it was reported to have reached 49.7%. These figures highlight the urgent need for more effective prevention strategies that specifically target adolescent girls.

Iron deficiency anemia (IDA) is the most prevalent type of anemia during adolescence, mainly because dietary intake tends to be insufficient, iron absorption fails to occur optimally, or physiological demands increase to have beyond what the body can supply (Elsayed

et al., 2016). Evaluating iron status is a crucial step and is commonly performed carried out to have by measuring serum ferritin, which serves to have as a key biomarker of the body's iron reserves (Sari et al., 2022). Iron is a vital micronutrient that is involved in numerous physiological processes. In addition to its role in hemoglobin formation and oxygen transport, iron is required for neuronal energy metabolism, supports neurotransmitter production, and contributes to neurogenesis (Al Rahmad, 2023; Ariani et al., 2022). Adequate iron availability helps maintain optimal brain function and psychological well-being, both of which are closely linked to the regulation of sleep.

In addition to ferritin, hemoglobin serves as another essential marker of anemia and overall oxygen status. Hemoglobin delivers oxygen from the lungs to various tissues across the body (Basrowi & Dilantika, 2021). Low hemoglobin levels impair oxygen delivery, causing fatigue, dizziness, headaches, and irregular heartbeat (Ferreira & Neves, 2019). Because oxygen availability impacts to have neuronal activity and neurotransmission, inadequate hemoglobin not only affects to have sleep quality but may also be influenced to have by physiological changes that occur during sleep, highlighting the importance of examining to have this interaction in adolescents (Basrowi & Dilantika, 2021).

Nutritional status plays an important role in shaping both anemia and sleep quality. Undernutrition increases the likelihood of iron deficiency, whereas overnutrition and obesity tend to disrupt t the body's ability to utilize and absorb iron efficiently (Chen-Edinboro et al., 2018; Dakanalis et al., 2024). Unhealthy dietary habits, such as skipping breakfast or frequently consuming fast food, can worsen nutritional imbalances and lead to decreases in both hemoglobin and ferritin levels (Kamaruddin et al., 2023). These imbalances may indirectly impair the sleep quality of adolescent girls (Dakanalis et al., 2024). Maintaining adequate nutrition during adolescence is essential for preventing anemia and supporting healthy sleep patterns.

Sleep is a fundamental physiological process that affects growth, emotional stability, and cognitive performance. Getting sufficient sleep helps with both physical restoration and neurocognitive development (Aminuddin, 2018). However, many adolescents tend to experience poor sleep quality, resulting which results to have in fatigue, reduced concentration, emotional instability, and an elevated risk of mental health problems such as anxiety and depression (Milojevich & Lukowski, 2016). Sleep disturbances in adolescents can arise from anemia-related reductions in oxygen delivery and stem from disruptions in monoamine metabolism, including impaired dopamine regulation as a consequence of low iron levels (Krisnawati et al., 2024; Neumann et al., 2021).

Given these interrelationships, iron status, reflected through ferritin and hemoglobin levels, and nutritional status are thought to play important roles in shaping sleep quality among adolescent girls. Building on this foundation, the present study aimed to examine the associations between ferritin levels, hemoglobin, and nutritional status with sleep quality among adolescent girls by employing multivariate regression analysis.

Methods

This study employed an observational approach with a cross-sectional design to analyze the relationship between ferritin levels, hemoglobin, and nutritional status and sleep quality among adolescent girls. The research was conducted from April to May 2025 within the working area of the Doro Community Health Center in Pekalongan Regency, Central Java, Indonesia. The study population comprised all adolescent female students living in the region.

The sample size calculation was conducted using a correlation analysis formula, which produced a minimum requirement of 69.86 participants. To anticipate possible dropouts and non-responses, the sample size was increased by an additional 15%, resulting in a final total of 80 adolescent girls. A 15% adjustment was applied based on recommendations for observational research, aiming to reduce the sampling bias caused by attrition (Lemeshow, 1990). Participants were chosen to have randomly using a simple random sampling technique, ensuring that each adolescent had to have an equal chance of being selected.

The inclusion criteria were defined as adolescent girls aged 13–18 years who were not menstruating at the time of data collection, were not suffering from chronic illnesses, were not taking iron supplements or medications that affect the central nervous system, and were not following any specific diet. The exclusion criteria included individuals with a history of infection in the previous two weeks, those diagnosed with medical conditions such as liver or kidney disease or hematologic disorders, and those who had consumed iron, vitamin, or mineral supplements within the past three months. Participants who were using steroids or non-steroidal anti-inflammatory drugs (NSAIDs) were also excluded from the study.

Sleep quality was evaluated using the Pittsburgh Sleep Quality Index (PSQI) questionnaire, which assesses seven components: subjective sleep quality, sleep latency, sleep duration, sleep efficiency, sleep disturbances, use of sleep medication, and daytime dysfunction. Each component was scored 0-3, with a global PSQI score > 5 indicating poor sleep quality (Shahid et al., 2012).

Ferritin levels were obtained through venous blood collection (3 mL) from either the median cubital or cephalic vein. The samples were preserved under cold conditions and analyzed using the Enzyme-Linked Immunosorbent Assay (ELISA) method (Human Ferritin ELISA Kit, General Biological Corp) at the GAKY Laboratory, Faculty of Medicine, Diponegoro University. Hemoglobin levels were examined at the Pekalongan Regional Health Laboratory. Nutritional status was evaluated using the Body Mass Index-for-Age (BMI/Age) z-score, following the adolescent assessment guidelines provided by the WHO AnthroPlus software. (WHO, 2012). Participants were categorized as underweight ($z < -2SD$), normal ($-2 \leq z \leq +1 SD$), overweight ($+1 < z \leq +2 SD$), or obese ($z > +2SD$).

Physical activity was assessed using the Global Physical Activity Questionnaire (GPAQ) developed by the World Health Organization (2012), which evaluates physical activity performed at school, during travel, and in recreational settings. The collected data were converted to Metabolic Equivalent of Task (MET) scores to determine the participants' activity levels.

Data were processed using SPSS version 30.0. The normality of the data was examined using the Kolmogorov–Smirnov test. The

Spearman rank correlation test was employed to evaluate the bivariate relationships among ferritin, hemoglobin, nutritional status, and sleep quality. Variables with $p < 0.25$ were included in the multiple linear regression analysis to assess the combined effects of the independent variables on sleep quality. The level of statistical significance was set at $p < 0.05$.

Ethical clearance for this study was obtained from the Health Research Ethics Committee of the Faculty of Medicine, Diponegoro University (Approval No. 082/EC/KEPK/FKUNDIP/IV/2025). All the participants and their parents received detailed information regarding the study's objectives, procedures, and benefits. Written informed consent was obtained from all participants prior to the study.

Result and Discussion

A total of 80 adolescent girls participated in this study, and none were excluded due to incomplete data.

Table 1 presents the demographic and clinical characteristics of the participants. The majority of respondents (85%) were aged below 16 years. In terms of anemia status, 93.8% were categorized as non-anemic, while 6.3% were identified as anemic. Ferritin levels were predominantly high (66.3%), whereas 33.8% were low. Regarding nutritional status, 44% had a normal BMI-for-age, 16% were underweight, 7% were overweight, and 33% were obese. Most participants (96.3%) reported good sleep quality, while 3.8% reported poor sleep quality. Regarding physical activity levels, 48.8% of participants were classified as having mild activity, 29.7% as having moderate activity, and 21.5% as having high activity levels.

Table 1. Characteristics of all the subjects (n=80)

| Variable | n (%) |
|--------------------|-----------|
| Age | |
| Under 16 years old | 68 (85) |
| Over 16 years old | 12 (15) |
| Hemoglobin level | |
| Anemia | 5 (6.3) |
| Non-Anemia | 75 (93.8) |
| Ferritin level | |
| Low | 27 (33.8) |
| High | 53 (66.3) |
| Nutritional status | |
| Underweight | 13 (16) |
| Normal | 35 (44) |
| Overweight | 6 (7) |
| Obesity | 26 (33) |
| Sleep quality | |
| Good | 3 (3.8) |
| Poor | 77 (96.2) |
| Physical activity | |
| Mild | 12 (15.3) |
| Moderate | 19 (22.7) |
| Severe | 49 (62) |

Values are presented as number (n) and percentage (%) for categoric variables

Table 2 presents the findings of the Spearman rank correlation analysis examining the relationships between the study variables and sleep quality. Hemoglobin levels ($r = 0.310$, $p = 0.005$) and nutritional status ($r = 0.229$, $p = 0.007$) were significantly positively correlated with sleep quality, indicating that higher hemoglobin levels and better nutritional status were associated with improved sleep quality. Ferritin levels ($r = 0.216$, $p = 0.540$), age ($r = 0.092$, $p = 0.415$), and physical activity ($r = -0.001$, $p = 0.992$) were not significantly correlated with sleep quality.

Table 2. Spearman rank test results on ferritin levels, hemoglobin levels, and nutritional status with sleep quality

| Variable | Median ± Min-Max | r-correlation | p-value |
|--------------------|---------------------------|---------------|---------|
| Age | 16.00 ± 15.00-17.00 | 0.092 | 0.415 |
| Ferritin level | 24.00 ± 4.00-170.00 | 0.216 | 0.540 |
| Hemoglobin | 13.85 ± 11.20-16.10 | 0.310 | 0.005 |
| Nutritional Status | 24.17 ± 14.88-39.54 | 0.229 | 0.007 |
| Physical activity | 3111.00 ± 140.00-11528.00 | -0.001 | 0.992 |

Spearman rank correlation test was used. $p < 0.05$ was considered statistically significant

Table 3 presents the results of the multiple linear regression analysis that examines the factors associated with sleep quality among

adolescent girls. The model had an R^2 value of 0.242 and an adjusted R^2 of 0.212, indicating that approximately 21.2% of the variation in sleep

quality could be explained by the independent variables included in the analysis. The overall regression model was statistically significant ($F=8.068$; $p=0.000$). Among the variables analyzed, hemoglobin level was the only factor that showed a statistically significant positive association with sleep quality ($OR=0.379$, $p<0.001$). This finding

indicates that higher hemoglobin levels are linked to better sleep quality. In contrast, age ($OR=0.034$, $p=0.741$), ferritin level ($OR=0.080$, $p=0.446$), nutritional status ($OR=0.161$, $p=0.143$), and physical activity ($OR=0.072$, $p=0.489$) were not significantly associated with sleep quality in this model.

Table 3. Multiple Linear Regression Analysis of Factors Associated with Sleep Quality

| Independent Variables | Sleep Quality | | | |
|-----------------------|---------------|-------|-------|---------|
| | β | SE | t | p-value |
| Age | 0.034 | 0.325 | 0.332 | 0.741 |
| Ferritin level | 0.800 | 0.008 | 0.766 | 0.446 |
| Hemoglobin level | 0.379 | 0.255 | 3.488 | <0.001 |
| Nutritional Status | 0.161 | 0.038 | 1.481 | 0.143 |
| Physical activity | 0.072 | 0.000 | 0.695 | 0.489 |

The dependent variable was sleep quality. The analysis was performed using multiple linear regressions. Statistical significance was set at $p < 0.05$.

The findings of this study show that hemoglobin levels and nutritional status were found to have significant associations with sleep quality in the bivariate analysis, whereas ferritin levels and physical activity did not appear to have such relationships. However, in the multivariate regression model, only hemoglobin levels were significantly associated with sleep quality. This result indicates that hemoglobin is the most influential factor affecting sleep quality among adolescent girls compared with ferritin, nutritional status, or physical activity. The disappearance of significance in the other variables after adjustment suggested the presence of confounding effects or interactions among the contributing factors. Therefore, the interpretation of bivariate findings needs to have support from multivariate analysis, as the latter provides to have a more accurate estimate of each variable's independent contribution to sleep quality

The significant link between hemoglobin levels and sleep quality aligns with the theory that hemoglobin functions as an the oxygen-transporting protein within red blood cells (Elsayed & Sharif, 2016). Optimal hemoglobin levels are essential for ensuring adequate oxygenation of all tissues, including the brain (Ferreira & Neves, 2019). This disruption has been linked to symptoms such as fatigue, diminished cognitive performance, and changes that affect the regulation of the sleep-wake cycle (Elsayed & Sharif, 2016). Changes in cerebral blood flow have also been linked to anemia and sleep disturbance (Chen-Edinboro et al., 2018). A

shortage of hemoglobin can interfere with the delivery of oxygen to neural tissues, which may result in impaired physiological functions, including fatigue, cognitive difficulties, and changes that alter normal sleep patterns (Elsayed & Sharif, 2016).

A decrease in hemoglobin's oxygen-carrying capacity can undermine cerebral perfusion, which is required for the proper functioning of brain regions involved in regulating sleep. One neuroimaging study has demonstrated a link between hemoglobin levels and cerebral blood flow, especially in the frontotemporal cortex, a region that plays a key role in sleep-wake control (Krisnawati et al., 2024). Structural brain alterations, including cortical thinning in the frontotemporal region linked to low hemoglobin levels, have been associated with shorter sleep duration and reduced sleep quality (Chen-Edinboro et al., 2018).

The involvement of the same brain regions associated with low hemoglobin levels and shortened sleep duration may suggest a shared biological pathway that contributes to the development of both anemia and insomnia. (Neumann et al., 2021). The participation of the same cortical regions linked to low hemoglobin levels and sleep disturbances suggests a shared pathophysiological mechanism underlying both anemia and insomnia (Zhu et al., 2021). Previous research has demonstrated that anemia is associated with a higher likelihood of experiencing poor sleep quality among adolescent girls (Yani et al., 2023; Krisnawati et al., 2024).

These findings indicate that hemoglobin affects sleep quality through a combination of physiological and neuroanatomical pathways. From a physiological standpoint, sufficient hemoglobin ensures effective oxygen transport, thereby supporting the metabolic needs of neural tissue. Neuroanatomically, hemoglobin levels may influence brain structure and function in regions that govern sleep regulation. Taken together, this evidence highlights the importance of maintaining optimal hemoglobin levels to promote healthy sleep patterns and prevent sleep disturbances.

In the bivariate analysis, nutritional status was significantly associated with sleep quality. However, this relationship disappeared in the multivariate model. This pattern suggests that the influence of nutritional status on sleep may be mediated by other factors, such as hemoglobin levels or metabolic hormones. Earlier studies have shown that both malnutrition and obesity can disrupt the regulation of leptin and ghrelin, hormones that play essential roles in appetite control and the initiation of sleep (Anam et al., 2022; Tetik & Kar Şen, 2021). Adolescents with a normal BMI tend to have better sleep quality, whereas both undernutrition and overnutrition increase the likelihood of experiencing poor sleep quality and sleep-disordered breathing.

In contrast to hemoglobin, this study did not identify a significant association between ferritin levels and sleep quality. Several factors may explain this outcome. Although ferritin is recognized as a reliable biomarker of iron storage, the present study did not demonstrate a meaningful relationship between ferritin levels and sleep quality. Ferritin reflects long-term iron reserves, however it may fail accurately represent the functional availability of iron within the brain (Okan et al., 2019). Inflammation can elevate ferritin levels regardless of the body's actual iron status, which may obscure the true iron-related effects on sleep (Kim et al., 2019). Moreover, ferritin's involvement in dopaminergic neurotransmission may operate indirectly, which helps explain why its association with sleep quality appears to have weaker effects than hemoglobin.

Physical activity was not significantly associated with sleep quality. This outcome may reflect relatively uniform activity levels among participants or indicate that the variation in activity intensity was not substantial enough to

influence sleep outcomes (Alnawwar et al., 2023). Nonetheless, the existing literature indicates that engaging in consistent moderate physical activity can improve sleep efficiency by stabilizing circadian rhythms and reducing stress levels (Wang & Boros, 2021). Therefore, physical activity remains an important behavioral factor for maintaining overall health.

The limitations of this study include its cross-sectional design, which prevents the establishment of causal relationships, and its relatively small and homogeneous sample drawn from a single district. Several potential confounders, such as dietary intake, menstrual cycle patterns, and psychosocial stress, were not comprehensively controlled. Despite these limitations, this study offers meaningful insights into the multifactorial aspects of sleep quality in adolescents, highlighting hemoglobin as a key determinant. Future longitudinal studies that incorporate biochemical and hormonal indicators are recommended to better clarify the underlying causal pathways

Conclusion

This study showed that hemoglobin levels were significantly associated with sleep quality among adolescent girls, whereas ferritin levels and nutritional status did not demonstrate independent associations after statistical adjustment. These results suggest that oxygen delivery via hemoglobin plays a more direct role in shaping sleep quality than iron storage or general nutritional status. Nevertheless, because this research had a cross-sectional design, the relationships observed should be interpreted as associative rather than causal.

Efforts to improve sleep quality among adolescent girls should focus on preventing anemia and optimizing hemoglobin levels through balanced nutrition, sufficient dietary iron intake, and routine hemoglobin screening in school settings. Health education programs that promote healthy eating habits and proper sleep hygiene may help enhance hemoglobin status and support overall well-being in adolescents.

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