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Diet-Related Nutritional Biomarker Patterns in Maternal Anemia: A Multicenter Exploratory Analysis of Vitamin D, Ferritin, Hepcidin and mTOR

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ABSTRACT:

Objective: To characterize diet-related biomarker patterns and explore statistical relationships among 25-hydroxyvitamin D [25(OH)D], ferritin, hepcidin, mTOR, and hemoglobin in pregnant women.

Methods: A multicenter cross-sectional study included 93 third-trimester pregnant women recruited from eight public health centers in Palembang, Indonesia, from September 2024 to December 2025. Dietary intake was assessed using a validated semi-quantitative food frequency questionnaire and repeated 24-hour recalls. Anthropometric and biochemical measures included BMI, MUAC, hemoglobin, 25(OH)D, ferritin, hepcidin, and mTOR. Group comparisons, correlation analysis, Principal Component Analysis (PCA), and exploratory Structural Equation Modeling (SEM) were performed.

Results: Compared with non-anemic women, those with anemia had lower vitamin D and iron intakes, lower 25(OH)D and ferritin, and higher hepcidin. 25(OH)D correlated positively with ferritin and inversely with hepcidin. PCA Component 1 explained 43.05% of variance, with loadings of 0.78, 0.65, −0.61, and 0.28 for 25(OH)D, ferritin, hepcidin, and mTOR, respectively. SEM showed significant associations of 25(OH)D with hepcidin ($\beta = -0.41$, $p = 0.002$) and ferritin ($\beta = 0.47$, $p = 0.001$), hepcidin with ferritin ($\beta = -0.33$, $p = 0.010$), and ferritin with hemoglobin ($\beta = 0.52$, $p < 0.001$). The mTOR–hemoglobin association was non-significant.

Conclusion: Maternal anemia showed an exploratory statistical pattern centered on vitamin D and iron-related biomarkers, with limited mTOR contribution. These findings support an integrated nutritional perspective and warrant longitudinal investigation.

1. INTRODUCTION

Maternal anemia remains a major global nutrition and public health concern, affecting nearly one in three pregnant women worldwide, with a disproportionate burden in low- and middle-income countries [1-3]. The problem remains substantial in Southeast Asia, where inadequate dietary intake, limited dietary diversity, and socioeconomic inequalities contribute to maternal nutritional vulnerability [4,5]. Although iron deficiency is a major contributor to anemia during pregnancy, inadequate consumption of iron-rich foods and poor overall dietary quality may coexist with deficiencies in other essential nutrients [6-8]. In Indonesia, maternal anemia has been associated with inadequate intake of essential micronutrients, including iron and vitamin D, as well as limited access to diverse and nutrient-dense foods [9,10]. These observations support a broader nutritional perspective in which maternal anemia is considered a diet-related condition influenced by dietary adequacy, maternal nutritional status, and interconnected biological processes. Such a perspective is particularly relevant to food science and nutrition research because identifying modifiable dietary factors may inform more comprehensive strategies for maternal anemia prevention.

The biological processes underlying maternal anemia involve interactions among micronutrient availability, iron homeostasis, inflammation, and cellular metabolism. Vitamin D, commonly assessed using serum 25-hydroxyvitamin D [25(OH)D], has received increasing attention because of its potential involvement in inflammatory regulation and iron-related processes [11,12]. Ferritin is widely used as an indicator of iron storage, whereas hepcidin is a key regulator of systemic iron homeostasis through its effects on intestinal iron absorption and iron release from storage sites [13,14]. Previous evidence suggests that vitamin D status may be related to hepcidin expression and iron availability for erythropoiesis [15,16]. However, ferritin is also an acute-phase reactant and may increase during infection or inflammation, necessitating cautious interpretation as an isolated indicator of iron status. The mechanistic target of rapamycin (mTOR), a nutrient-sensing signaling pathway involved in cellular growth, protein synthesis, and metabolic adaptation, may provide an additional perspective on nutritional and metabolic regulation [17,18]. Although its specific relationship with iron bioavailability and maternal anemia remains less established, examining mTOR together with vitamin D and iron-related biomarkers may help characterize broader nutritional and metabolic patterns.

Maternal anemia is additionally influenced by nutritional, anthropometric, environmental, and contextual factors. Maternal undernutrition or excess adiposity, dietary adequacy, socioeconomic circumstances, infection or inflammation, and deficiencies of hematopoietic micronutrients such as vitamin B12 and folate may contribute to variation in maternal hematological status. Nutritional conditions before conception and throughout pregnancy, maternal weight gain, dietary patterns, supplementation practices, and sun exposure may further influence nutrient availability and iron-related biomarkers. These factors are particularly important because inflammatory or infectious conditions can alter hepcidin and ferritin concentrations independently of dietary iron intake. Consequently, interpretation of biomarker profiles requires consideration of the broader nutritional and contextual environment rather than attribution of maternal anemia to a single biochemical marker. However, relatively few studies have simultaneously considered dietary intake, anthropometric characteristics, vitamin D status, iron-regulatory biomarkers, and nutrient-sensing pathways within a unified analytical framework.

An important gap in the existing literature is the predominance of studies examining individual biomarkers rather than their combined statistical patterns. Research focusing primarily on hemoglobin or ferritin has provided important information on individual components of anemia but offers limited insight into the interrelationships among vitamin D status, iron regulation, nutrient intake, and cellular signaling pathways [19,20]. Although studies examining vitamin D and iron status have expanded the evidence base, comprehensive evaluations of multiple interconnected nutritional and biochemical indicators remain relatively uncommon [21]. Multivariate approaches such as Principal Component Analysis (PCA) can be used to characterize covariance patterns among correlated dietary or biological variables and identify variables contributing most strongly to an observed pattern [22,23]. In the context of maternal anemia, this approach may provide a complementary perspective by identifying clusters of nutritional and biochemical indicators rather than evaluating each marker independently. However, because the present study is cross-sectional, PCA and pathway modeling should be interpreted as exploratory analyses of statistical patterns and hypothesized relationships rather than confirmation of latent constructs, biological mechanisms, or causal pathways.

This integrated perspective is also relevant to nutritional approaches for preventing and managing maternal anemia. Current international recommendations emphasize iron supplementation as a central component of antenatal anemia prevention,

while the contribution of other micronutrients, including vitamin D, may vary across populations and intervention frameworks [24,25]. Emerging evidence suggests that dietary quality and multi-micronutrient approaches may be relevant where multiple nutritional deficiencies coexist during pregnancy [26]. Evaluating dietary intake together with vitamin D status, iron storage and regulation, and nutrient-related biomarkers may therefore provide a broader basis for understanding maternal anemia. Importantly, such an approach does not imply replacement of established iron–folic acid strategies; rather, it may help identify additional nutritional factors that warrant consideration in dietary assessment and future intervention research. The relationships among these indicators may also vary according to maternal nutritional status, anthropometry, socioeconomic circumstances, supplementation, sun exposure, and inflammatory or infectious conditions.

In Indonesia, including urban settings such as Palembang, maternal anemia remains associated with dietary patterns, socioeconomic disparities, and inadequate micronutrient intake, while integrated nutrition-oriented investigations remain limited [27,28]. Multicenter studies simultaneously examining dietary intake, maternal nutritional characteristics, biochemical biomarkers, and multivariate statistical patterns are particularly scarce. Therefore, this study aimed to characterize diet-related nutritional biomarker patterns and examine exploratory statistical relationships among 25(OH)D, ferritin, hepcidin, mTOR, and maternal anemia in a multicenter sample of pregnant women. Specifically, correlation analysis, exploratory PCA, and SEM were used to examine the interrelationships and relative contributions of these nutritional and biochemical indicators. The study did not seek to establish a causal biological pathway; rather, it aimed to identify coherent statistical patterns that may help generate hypotheses regarding the relationship between diet, nutritional biomarkers, and maternal anemia. By integrating dietary, anthropometric, and biochemical information within a unified framework, this study provides a multidimensional perspective relevant to nutrition and food science research and may inform future longitudinal and intervention studies of integrated nutritional strategies.

2. MATERIALS AND METHODS

2.1 Study Design and Setting

This study employed a multicenter cross-sectional design to examine diet-related nutritional biomarker patterns and their associations with maternal anemia. The study was conducted between September 2024 and December 2025 across eight Public Health Centers (Puskesmas) in Palembang, Indonesia. The multicenter approach was used to capture variation in maternal characteristics, dietary intake, environmental exposure, and health conditions across urban primary healthcare settings. A cross-sectional design was considered appropriate for characterizing nutritional and biochemical profiles and examining associations between dietary and biomarker variables in the study population [29-31].

2.2 Sample Size Determination

Sample size considerations were based on the multivariate nature of the planned analyses and the number of variables included in the analytical framework. For Principal Component Analysis (PCA), a subject-to-variable ratio of approximately 5–10 participants per variable has been recommended to support the stability and interpretability of extracted components [32,33]. Based on the primary biomarker variables and additional analytical variables included in the study, the initial minimum sample requirement was estimated at 80 participants.

An a priori power analysis was also conducted using G*Power for regression-based analysis, assuming a medium effect size ($f^2 = 0.15$), a significance level of $\alpha = 0.05$, and statistical power of 80%, which yielded a minimum requirement of 85 participants [34]. A total of 93 participants were ultimately included in the analysis. Because the pathway analysis was conducted using a relatively parsimonious model with observed variables rather than a large latent-variable structure, the final sample size was considered in relation to the number of estimated parameters and the complexity of the specified model. The SEM results were therefore interpreted as estimates of hypothesized associations within this specific model rather than as evidence of causal relationships. Model adequacy was additionally evaluated using multiple goodness-of-fit indices, including χ^2, degrees of freedom (df), CFI, RMSEA, and SRMR.

2.3 Study Population and Sampling

The study population comprised pregnant women in the third trimester who attended antenatal care services at the participating Puskesmas. Participants were recruited using consecutive sampling, whereby eligible women attending the participating facilities during the study period were invited to participate. Women were eligible if they were aged 18–45 years,

had a gestational age of ≥28 weeks, were receiving antenatal care at one of the participating facilities, and were willing to participate and provide the required dietary, anthropometric, and biological information. Women were excluded if they had documented chronic diseases that could substantially influence hematological or nutritional status, including diabetes mellitus or renal disorders; had an acute infection at the time of assessment; or were receiving medications known to substantially affect iron or vitamin D metabolism beyond routine antenatal supplementation. Participants with incomplete information required for the primary biomarker analysis were also excluded from the final analytical dataset. Maternal anemia was defined as a hemoglobin (Hb) concentration <11 g/dL, consistent with the World Health Organization criteria for anemia during pregnancy [35].

2.4 Data Collection and Measurement Procedures

2.4.1 Sociodemographic and Anthropometric Assessment

Sociodemographic information was obtained using a structured, pre-tested questionnaire adapted from national health survey instruments. Information collected included relevant maternal and household characteristics available within the study protocol. Anthropometric assessment was performed using standardized procedures. Body weight was measured with a calibrated digital scale (Seca®, Germany) to the nearest 0.1 kg, while standing height was measured using a portable stadiometer (Seca®, Germany) to the nearest 0.1 cm. Body mass index (BMI) was calculated as weight in kilograms divided by height in meters squared (kg/m²) and categorized according to applicable WHO standards. Mid-upper arm circumference (MUAC) was measured using a non-stretchable MUAC tape following standardized procedures, with the measurement taken at the midpoint between the acromion and olecranon processes and recorded to the nearest 0.1 cm. Each anthropometric measurement was performed twice, and the mean of the two measurements was used for analysis to improve measurement reliability [36,37]. BMI and MUAC were included to provide an indication of maternal anthropometric nutritional status and to allow nutritional status to be considered when interpreting the relationships between dietary intake, biomarkers, and anemia.

2.4.2 Dietary Assessment

Dietary intake was assessed using a semi-quantitative Food Frequency Questionnaire (SQ-FFQ) adapted to Indonesian dietary patterns and previously used in nutritional research. The questionnaire assessed habitual food consumption during the preceding month, with particular attention to foods contributing to vitamin D and iron intake.

To complement the FFQ and improve estimation of usual nutrient intake, a multiple-pass 24-hour dietary recall was conducted on two non-consecutive days, comprising one weekday and one weekend day. Portion sizes were estimated using standardized household measurements and food photographs. Daily nutrient intake was calculated using the Indonesian Food Composition Database (*Daftar Komposisi Pangan Indonesia*) and expressed as vitamin D intake (μg/day) and iron intake (mg/day). Information on average daily sun exposure, vitamin D supplementation, and iron tablet consumption was also collected. The combined use of FFQ and repeated 24-hour dietary recall can improve characterization of habitual dietary intake and reduce some sources of measurement error associated with reliance on a single dietary assessment method [38-40].

2.4.3 Biochemical Measurements

Venous blood samples (5 mL) were collected by trained laboratory personnel under aseptic conditions. Following collection, the samples were processed by centrifugation, and the resulting serum aliquots were stored at −80°C until biochemical analysis. Hemoglobin (Hb) concentrations were determined using an automated hematology analyzer (Sysmex®, Japan), with daily internal quality-control procedures to ensure analytical consistency. Serum 25-hydroxyvitamin D [25(OH)D] concentrations were measured using an enzyme-linked immunosorbent assay (ELISA; DRG Instruments GmbH, Germany), with a reported analytical sensitivity of <2 ng/mL. Ferritin concentrations were determined using a chemiluminescent immunoassay (CLIA; Abbott Architect®, USA) as a circulating biomarker of iron stores. Serum hepcidin concentrations were quantified using a commercially available human hepcidin ELISA kit (Elabscience®, USA), while serum mechanistic target of rapamycin (mTOR) concentrations were assessed using a human serum sandwich ELISA kit (Cloud-Clone Corp®, USA). All immunoassays were performed in duplicate according to the respective manufacturers' protocols. Intra-assay and inter-assay coefficients of variation were maintained below 10% to ensure acceptable analytical precision. Laboratory procedures were conducted in accordance with standardized clinical chemistry procedures and manufacturer-recommended protocols [41,42].

Ferritin concentrations were interpreted within the broader context of iron metabolism because ferritin is an established marker of iron storage but may also be influenced by inflammatory processes. Accordingly, ferritin was considered an iron-related biomarker rather than an isolated definitive measure of maternal iron status, particularly in the absence of direct assessment of inflammatory markers.

2.5 Variable Specification and Analytical Framework

To provide a clearer description of the analytical framework, variables were classified into the primary outcome, biochemical biomarkers, dietary exposures, anthropometric indicators, and sociodemographic covariates. The operational definitions and measurement approaches are summarized in Table 1.

Table 1. Variable specification and measurement framework

Variable category	Variable	Instrument/Method	Scale
Primary outcome	Anemia status	Hb measured using Sysmex hematology analyzer; anemia defined as Hb <11 g/dL	Categorical
Biochemical biomarkers	25(OH)D	ELISA (DRG kit)	Continuous
	Ferritin	CLIA (Abbott Architect)	Continuous
	Hepcidin	ELISA (Elabscience)	Continuous
	mTOR	Human serum sandwich ELISA (Cloud-Clone)	Continuous
Dietary exposures	Vitamin D intake	SQ-FFQ + two non-consecutive 24-h recalls	Continuous
	Iron intake	SQ-FFQ + two non-consecutive 24-h recalls	Continuous
Anthropometric indicators	BMI	Digital scale + stadiometer	Continuous
	MUAC	Standardized MUAC tape	Continuous
Additional nutritional/environmental variables	Sun exposure	Structured questionnaire	Continuous
	Vitamin D supplementation	Structured questionnaire	Continuous
	Iron tablet consumption	Structured questionnaire	Continuous
Sociodemographic variables	Maternal/household characteristics	Structured questionnaire	Mixed

Hb, hemoglobin; 25(OH)D, 25-hydroxyvitamin D; mTOR, mechanistic target of rapamycin; ELISA, enzyme-linked immunosorbent assay; CLIA, chemiluminescent immunoassay; SQ-FFQ, semi-quantitative food frequency questionnaire. Anemia was defined as Hb <11 g/dL.

2.6 Statistical Analysis

Statistical analyses were performed using SPSS and R software, including the *lavaan* package for pathway modeling. Continuous variables were assessed for distributional characteristics and summarized as mean ± standard deviation (SD) for approximately normally distributed data or median and interquartile range (IQR) for non-normally distributed data. Categorical variables were summarized using frequencies and percentages. Differences between participants with and without anemia were assessed using the independent-samples *t*-test for normally distributed continuous variables or the Mann–Whitney *U* test for non-normally distributed variables. Categorical variables were compared using the chi-square test or an appropriate alternative when assumptions for the chi-square test were not met. Associations between continuous biomarkers were examined using Pearson or Spearman correlation coefficients according to the distribution of the variables. Correlation coefficients were interpreted as measures of association and were not considered evidence of causal relationships.

2.6.1 Principal Component Analysis

Principal Component Analysis (PCA) was performed to identify dominant patterns among the selected nutritional and biochemical biomarkers. The suitability of the data for PCA was assessed using the Kaiser–Meyer–Olkin (KMO) measure and Bartlett's test of sphericity. A KMO value ≥0.60 and a statistically significant Bartlett's test ($p < 0.05$) were considered indicative of adequate factorability. Components were retained based on eigenvalues >1, and variables with factor loadings ≥0.40 were considered meaningful contributors to the corresponding component [43]. The PCA was used as a data-reduction and pattern-identification approach rather than as a method for establishing causality. The resulting component structure was interpreted in relation to the nutritional and biological characteristics of the included biomarkers.

2.6.2 Pathway Modeling

Pathway modeling using the *lavaan* package in R was performed to examine the hypothesized relationships among dietary and biochemical variables and maternal anemia. The model was specified a priori based on the proposed relationships among vitamin D status, iron-related biomarkers, mTOR, and hemoglobin. Because the study used a cross-sectional design, the direction of the specified pathways represented hypothesized statistical relationships rather than temporal or causal effects. Model fit was evaluated using multiple indices, including the chi-square statistic (χ^2), degrees of freedom (df), comparative fit index (CFI), root mean square error of approximation (RMSEA), and standardized root mean square residual (SRMR). The model was considered to demonstrate acceptable fit when CFI was ≥0.90, RMSEA was ≤0.08, and SRMR was ≤0.08 [44-46]. The χ^2 statistic, df, and associated p-value were also reported to provide a complete assessment of model fit. Statistical significance was evaluated using a two-sided $p < 0.05$.

3. RESULTS AND DISCUSSION

3.1 Participant characteristics

A total of 93 third-trimester pregnant women who fulfilled the predefined eligibility criteria were included in the final analysis and were recruited consecutively from eight public health centers in Palembang, Indonesia. As presented in Table 2, participants were characterized according to sociodemographic, anthropometric, dietary, and biochemical variables and were compared according to maternal anemia status. The anemia group (n = 37) had significantly lower hemoglobin, 25(OH)D, and ferritin concentrations and higher hepcidin concentrations than the non-anemia group (n = 56) (all $p < 0.05$), whereas mTOR concentrations did not differ significantly ($p = 0.314$). Women with anemia also had significantly lower vitamin D and iron intakes, BMI, MUAC, and daily sun exposure, as well as lower iron supplementation prevalence ($p < 0.05$). In contrast, maternal age, gestational age, multiparity, and vitamin D supplementation did not differ significantly between groups ($p > 0.05$). Overall, these findings indicate distinct nutritional and biochemical profiles according to maternal anemia status, particularly involving vitamin D status and iron-related biomarkers.

Table 2. Sociodemographic, anthropometric, dietary, and biochemical characteristics according to maternal anemia status

Variable	Anemia (n = 37)	Non-anemia (n = 56)	*p*-value
Hematological and biochemical characteristics			
Hemoglobin (g/dL)	10.2 ± 0.8	12.1 ± 0.7	<0.001
25(OH)D (ng/mL)	18.5 ± 6.2	24.8 ± 7.1	0.002
Ferritin (µg/L)	15.6 ± 10.4	32.7 ± 18.9	0.001
Hepcidin (ng/mL)	7.8 ± 4.2	5.6 ± 3.1	0.021
mTOR (ng/mL)	4.5 ± 1.8	4.2 ± 1.5	0.314
Dietary characteristics			
Vitamin D intake (µg/day)	3.2 ± 2.1	6.8 ± 3.4	0.004
Iron intake (mg/day)	14.5 ± 6.3	22.1 ± 8.5	0.006

Anthropometric characteristics			
BMI (kg/m²)	22.8 ± 3.1	24.1 ± 3.4	0.048
MUAC (cm)	23.1 ± 2.4	24.5 ± 2.6	0.009
Sociodemographic and obstetric characteristics			
Maternal age (years)	29.4 ± 5.2	28.7 ± 4.8	0.487
Gestational age (weeks)	34.8 ± 1.5	34.5 ± 1.6	0.361
Multiparity, *n* (%)	22 (59.5)	27 (48.2)	0.296
Environmental and supplementation characteristics			
Sun exposure (min/day)	31.6 ± 14.8	42.3 ± 18.6	0.003
Vitamin D supplementation, *n* (%)	9 (24.3)	24 (42.9)	0.071
Iron supplementation, *n* (%)	25 (67.6)	48 (85.7)	0.032

Data are presented as mean ± SD or n (%). $p < 0.05$ indicates statistical significance. 25(OH)D, 25-hydroxyvitamin D; BMI, body mass index; MUAC, mid-upper arm circumference; mTOR, mechanistic target of rapamycin.

Table 2 shows that women with maternal anemia had significantly lower serum 25(OH)D, ferritin, vitamin D intake, iron intake, BMI, and MUAC, but higher hepcidin concentrations than women without anemia. Ferritin concentrations were approximately half those of the non-anemia group, while vitamin D and iron intake were also substantially lower. Sun exposure was significantly lower among women with anemia, whereas vitamin D supplementation did not differ significantly; iron supplementation was significantly less frequent in the anemia group. In contrast, maternal age, gestational age, multiparity, and mTOR concentrations did not differ significantly between groups. Overall, the clearest between-group differences involved vitamin D and iron-related biomarkers, dietary intake, anthropometric indicators, and sun exposure, whereas demographic characteristics and circulating mTOR showed comparatively limited differences. These findings represent group-level associations and should not be interpreted as evidence of causality.

3.2 Anthropometric and Dietary Characteristics

Anthropometric, dietary, and nutritional exposure characteristics were compared according to maternal anemia status. As shown in Table 3, women with anemia had significantly lower BMI, MUAC, vitamin D and iron intakes, sun exposure, and iron supplementation than those without anemia, whereas vitamin D supplementation did not differ significantly between groups. These findings indicate distinct nutritional profiles according to maternal anemia status.

Table 3. Anthropometric, dietary, and nutritional exposure characteristics according to maternal anemia status

Variable	Anemia (n = 37)	Non-anemia (n = 56)	*p*-value
Anthropometric characteristics			
BMI (kg/m²)	22.8 ± 3.1	24.1 ± 3.4	0.048
MUAC (cm)	23.1 ± 2.4	24.5 ± 2.6	0.009
Dietary intake			
Vitamin D intake (µg/day)	3.2 ± 2.1	6.8 ± 3.4	0.004
Iron intake (mg/day)	14.5 ± 6.3	22.1 ± 8.5	0.006
Nutritional and environmental exposure			
Sun exposure (min/day)	31.6 ± 14.8	42.3 ± 18.6	0.003

Vitamin D supplementation, *n* (%)	9 (24.3)	24 (42.9)	0.071
Iron supplementation, *n* (%)	25 (67.6)	48 (85.7)	0.032

Data are presented as mean ± SD or n (%). $p < 0.05$ indicates statistical significance. BMI, body mass index; MUAC, mid-upper arm circumference.

As shown in Table 3, women with maternal anemia had significantly lower BMI and MUAC, as well as lower vitamin D and iron intake, than women without anemia. Sun exposure was also significantly lower in the anemia group. Vitamin D supplementation was more frequently reported among women without anemia, although the difference was not statistically significant, whereas iron supplementation was significantly more frequent in the non-anemia group. Overall, Table 3 demonstrates consistent differences across anthropometric status, dietary intake, and nutritional exposures according to maternal anemia status. These findings provide additional nutritional context for the lower 25(OH)D and ferritin concentrations observed in the anemia group. However, given the cross-sectional design, these differences represent associations and should not be interpreted as evidence of temporal or causal effects.

3.3. Correlations Among Nutritional and Biochemical Biomarkers

To assess the interrelationships among the measured nutritional and biochemical biomarkers, correlation analysis was performed, and the resulting correlation matrix is presented in Table 4.

Table 4. Correlation matrix among nutritional and biochemical biomarkers

Variable	25(OH)D	Ferritin	Hepcidin	mTOR
25(OH)D	1.00	0.42**	−0.36**	0.12
Ferritin	0.42**	1.00	−0.29*	0.15
Hepcidin	−0.36**	−0.29*	1.00	0.18
mTOR	0.12	0.15	0.18	1.00

Values are Pearson correlation coefficients (r). $p < 0.05$ and *$p < 0.01$.*

Table 4 shows significant correlations among 25(OH)D, ferritin, and hepcidin. Serum 25(OH)D was positively correlated with ferritin ($r = 0.42$, $p < 0.01$) and inversely correlated with hepcidin ($r = -0.36$, $p < 0.01$), while ferritin was also inversely correlated with hepcidin ($r = -0.29$, $p < 0.05$). In contrast, mTOR showed weak and non-significant correlations with 25(OH)D, ferritin, and hepcidin. Overall, the correlation pattern indicates that 25(OH)D, ferritin, and hepcidin exhibited greater statistical interrelatedness than mTOR within this biomarker panel. These findings provide the basis for the subsequent exploratory PCA and SEM analyses; however, the observed correlations represent associations and do not establish causal or mechanistic relationships.

3.4 Exploratory Principal Component Analysis

Given the modest sample size and the exploratory nature of the analysis, PCA was performed to identify patterns of covariance among the measured biomarkers rather than to establish a definitive latent construct or biological pathway. The resulting component loadings and PCA adequacy measures are presented in Tables 5 and 5A.

Table 5. Exploratory principal component loadings

Biomarker	Component 1 loading
25(OH)D	0.78
Ferritin	0.65
Hepcidin	−0.61
mTOR	0.28

PCA was conducted as an exploratory analysis; KMO = Kaiser–Meyer–Olkin.

As shown in Table 5, 25(OH)D showed the highest loading on Component 1 (0.78), followed by ferritin (0.65) and hepcidin (−0.61), whereas mTOR showed a lower loading (0.28). The opposite direction of the hepcidin loading indicates an inverse contribution to the observed covariance pattern, characterized by higher 25(OH)D and ferritin concentrations alongside lower hepcidin concentrations. These loadings describe the relative contribution of each biomarker to the exploratory component and should not be interpreted as evidence of causal or hierarchical relationships.

Table 5A. PCA adequacy and explained variance

Parameter	Result
Kaiser–Meyer–Olkin (KMO)	0.57
Bartlett's test of sphericity, χ^2	41.26
df	6
p-value	<0.001
Eigenvalue, Component 1	1.72
Variance explained (%)	43.05

PCA was conducted as an exploratory analysis; KMO = Kaiser–Meyer–Olkin.

Table 5A shows that Bartlett's test of sphericity was significant ($\chi^2 = 41.26$, df = 6, $p < 0.001$), indicating correlations among the biomarkers. However, the KMO value of 0.57 was below the conventional threshold of 0.60, indicating modest sampling adequacy. Component 1 had an eigenvalue of 1.72 and explained 43.05% of the total variance. Therefore, the PCA findings should be interpreted cautiously as an exploratory biomarker covariance pattern, rather than as a validated latent construct, biological pathway, or causal hierarchy.

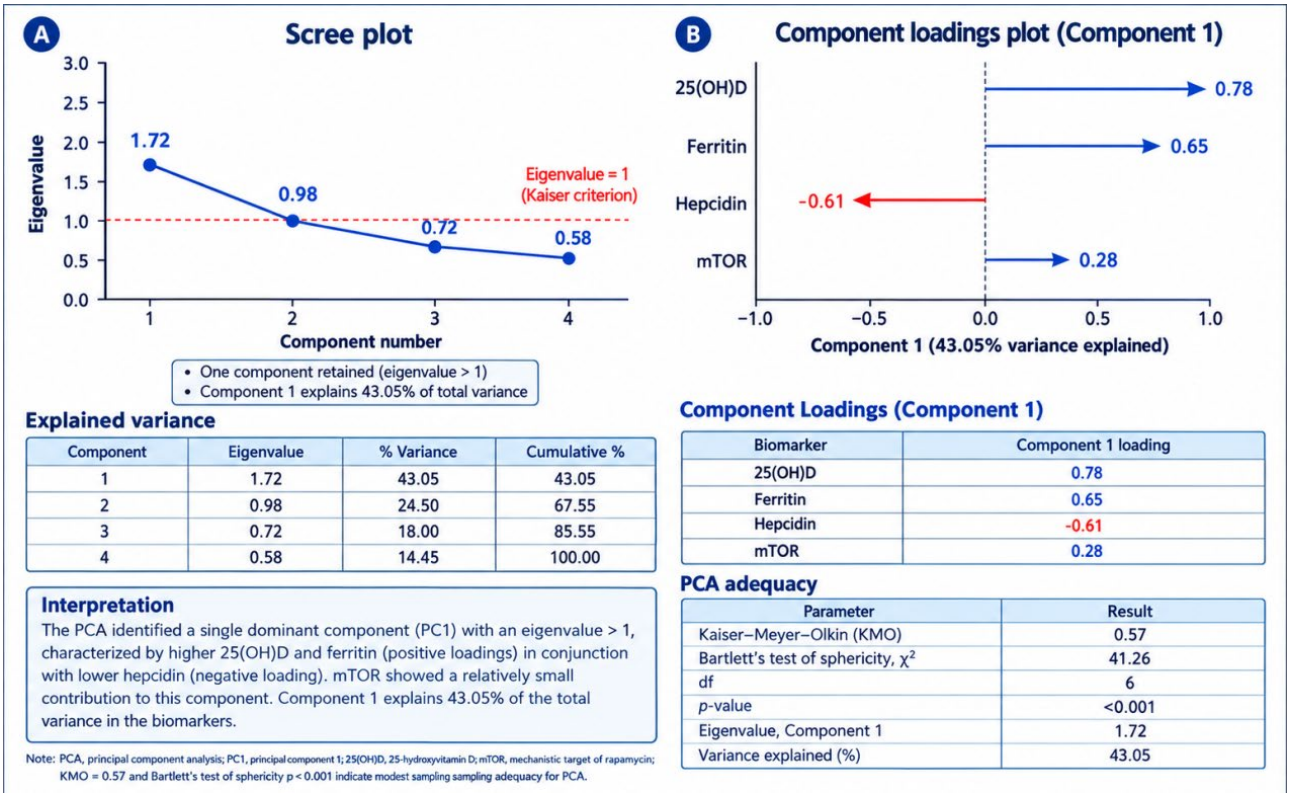

Figure 1. Exploratory PCA of Nutritional and Biochemical Biomarkers

Figure 1 illustrates the exploratory covariance pattern identified by PCA. Component 1 was characterized primarily by 25(OH)D, ferritin, and hepcidin, whereas mTOR contributed comparatively less. Although the significant Bartlett's test indicates that the biomarkers were statistically interrelated, the modest KMO value (0.57) warrants cautious interpretation. Accordingly, Component 1 is best regarded as a descriptive exploratory pattern of biomarker covariance, rather than a definitive mechanistic pathway.

3.5 Structural Equation Modeling

To further examine the hypothesized statistical relationships among vitamin D status, iron-related biomarkers, mTOR, and hemoglobin, an exploratory Structural Equation Modeling (SEM) analysis was performed. The standardized path coefficients and their statistical significance are presented in Table 6.

Table 6. Standardized path coefficients of the structural equation model

Pathway	Standardized β	*p*-value	Interpretation
25(OH)D → Hepcidin	−0.41	0.002	Significant inverse association
25(OH)D → Ferritin	0.47	0.001	Significant positive association
Hepcidin → Ferritin	−0.33	0.010	Significant inverse association
Ferritin → Hemoglobin	0.52	<0.001	Significant positive association
mTOR → Hemoglobin	0.18	0.087	Not statistically significant

β = standardized coefficient. Associations are exploratory and do not imply causality.

As shown in Table 6, 25(OH)D was significantly and inversely associated with hepcidin (β = −0.41, p = 0.002) and positively associated with ferritin (β = 0.47, p = 0.001). Hepcidin was inversely associated with ferritin (β = −0.33, p = 0.010), whereas ferritin was positively associated with hemoglobin (β = 0.52, p < 0.001). In contrast, mTOR was not significantly associated with hemoglobin (β = 0.18, p = 0.087), indicating a comparatively limited contribution to the observed statistical pattern. Overall, the strongest statistical associations were concentrated among 25(OH)D, hepcidin, ferritin, and hemoglobin. However, because the data were cross-sectional, these coefficients represent statistical associations rather than direct or indirect causal effects. The model therefore does not establish that 25(OH)D suppresses hepcidin, that hepcidin alters ferritin, or that ferritin subsequently determines hemoglobin. Accordingly, the SEM should be interpreted as an exploratory representation of observed biomarker interrelationships, rather than confirmation of a biological mechanism or causal pathway.

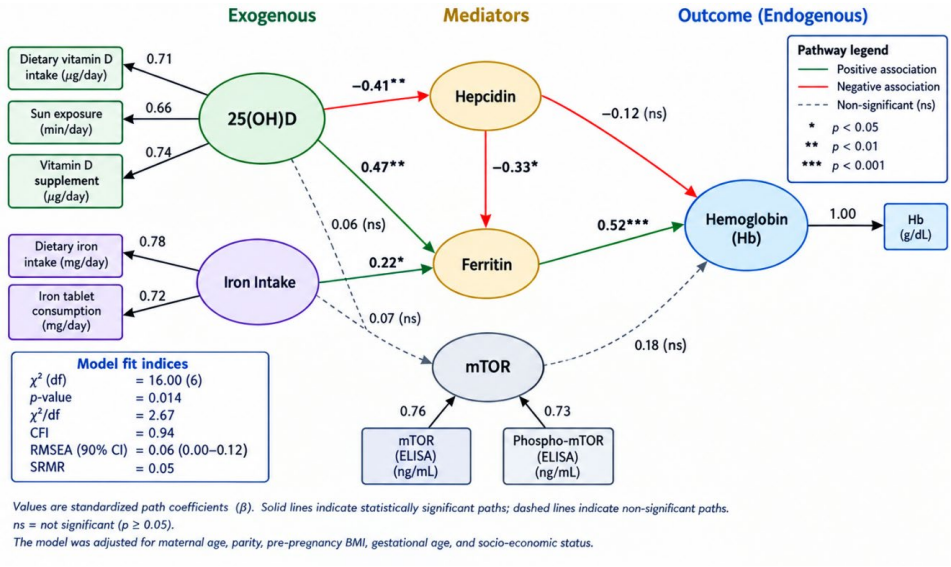

Figure 2. Exploratory Structural Equation Model of Nutritional and Biochemical Biomarker Associations

Figure 2 illustrates the exploratory statistical associations among 25(OH)D, hepcidin, ferritin, mTOR, and hemoglobin. Significant associations were observed between 25(OH)D and hepcidin, 25(OH)D and ferritin, hepcidin and ferritin, and ferritin and hemoglobin, whereas the mTOR–hemoglobin association was not statistically significant. These coefficients represent statistical associations within the observed cross-sectional dataset and should not be interpreted as causal or mechanistic relationships.

3.6 Goodness-of-Fit of the Structural Equation Model

To evaluate the compatibility of the exploratory structural equation model with the observed covariance structure, the model fit was assessed using the prespecified goodness-of-fit indices, as presented in Table 7.

Table 7. Goodness-of-fit indices of the structural equation model

Fit index	Observed value	Prespecified criterion
χ^2	16.00	—
df	6	—
χ^2/df	2.67	<3.00
CFI	0.94	≥0.90
RMSEA	0.06	≤0.08
SRMR	0.05	≤0.08

CFI = comparative fit index; RMSEA = root mean square error of approximation; SRMR = standardized root mean square residual.

As presented in Table 7, the exploratory structural equation model demonstrated acceptable fit across the prespecified indices. The χ^2 value was 16.00 with 6 degrees of freedom, yielding a χ^2/df ratio of 2.67, below the prespecified criterion of 3.00. The model also showed a CFI of 0.94, RMSEA of 0.06, and SRMR of 0.05, all meeting the predefined criteria for acceptable fit. Collectively, these indices indicate reasonable statistical compatibility between the specified model and the observed covariance structure. However, acceptable model fit does not establish causal relationships, temporal ordering, or biological mechanisms. Given the cross-sectional design and modest sample size (n = 93), the SEM should therefore be interpreted as an exploratory framework for characterizing observed statistical interrelationships among 25(OH)D, hepcidin, ferritin, mTOR, and hemoglobin rather than as a definitive mechanistic model.

3.7 Associations Between Biomarkers and Maternal Anemia

To further examine the associations between selected nutritional and biochemical biomarkers and maternal anemia, odds ratios (ORs) with corresponding 95% confidence intervals (CIs) were estimated, as presented in Table 8.

Table 8. Associations of Selected Biomarkers with Maternal Anemia

Biomarker	Exposure category	OR	95% CI
25(OH)D	Low	2.85	1.60–5.10
Ferritin	Low	2.30	1.40–3.80
Hepcidin	High	1.75	1.10–2.90
mTOR	—	1.20	0.80–1.90

OR = odds ratio; CI = confidence interval. ORs represent statistical associations with maternal anemia.

As shown in Table 8, low 25(OH)D concentrations were associated with higher odds of maternal anemia (OR = 2.85, 95% CI: 1.60–5.10), as were low ferritin concentrations (OR = 2.30, 95% CI: 1.40–3.80) and high hepcidin concentrations (OR = 1.75, 95% CI: 1.10–2.90). In contrast, mTOR was not significantly associated with maternal anemia (OR = 1.20, 95% CI: 0.80–1.90), as its confidence interval included the null value. Overall, the observed associations were consistent with the between-group differences and exploratory biomarker analyses, with stronger statistical relationships involving 25(OH)D,

ferritin, and hepcidin than mTOR. However, these estimates should be interpreted in relation to the specified exposure categories, reference groups, covariates, and regression model. Given the cross-sectional design, the findings indicate statistical associations and do not establish causality.

3.8. Integrated Nutritional Biomarker Pattern

To integrate the findings across dietary, anthropometric, biochemical, correlation, PCA, and SEM analyses, the principal nutritional and biomarker patterns associated with maternal anemia are summarized in Table 9.

Table 9. Summary of the Integrated Nutritional and Biomarker Pattern Associated with Maternal Anemia

Analytical domain	Principal finding	Statistical evidence
Dietary vitamin D	Lower intake in anemia	$p = 0.004$
Dietary iron	Lower intake in anemia	$p = 0.006$
Vitamin D status	Lower 25(OH)D in anemia	$p = 0.002$
Iron storage	Lower ferritin in anemia	$p = 0.001$
Iron regulation	Higher hepcidin in anemia	$p = 0.021$
Anthropometric status	Lower BMI and MUAC in anemia	$p = 0.048$ and 0.009
Biomarker correlation	25(OH)D correlated with ferritin and hepcidin	$r = 0.42$ and -0.36
Exploratory biomarker pattern	25(OH)D, ferritin, and hepcidin showed stronger loadings than mTOR	PC1 = 43.05% variance
mTOR	Lower contribution to the exploratory pattern; no significant association with hemoglobin	Loading = 0.28; $\beta = 0.18$, $p = 0.087$

Findings represent statistical associations; PCA and SEM were exploratory and do not establish causality.

As summarized in Table 9, maternal anemia was characterized by a consistent nutritional and biochemical pattern involving lower dietary vitamin D and iron intake, lower 25(OH)D and ferritin concentrations, higher hepcidin concentrations, and lower BMI and MUAC. Correlation analysis demonstrated significant relationships among 25(OH)D, ferritin, and hepcidin, while exploratory PCA showed stronger contributions of these three biomarkers to Component 1 than mTOR. SEM further identified significant statistical associations involving 25(OH)D, hepcidin, ferritin, and hemoglobin, whereas the mTOR–hemoglobin association was not statistically significant. Overall, the findings indicate that the principal statistical pattern in this dataset was centered on vitamin D and iron-related biomarkers, with a comparatively limited contribution from mTOR. However, this pattern should be interpreted as an exploratory statistical finding rather than evidence of a dominant biological pathway or causal mechanism, particularly given the KMO value of 0.57, cross-sectional design, and modest sample size. Ferritin should also be interpreted within the broader clinical and nutritional context because its concentration may be influenced by inflammation.

3.9 DISCUSSION

Maternal anemia in this multicenter cross-sectional study was characterized by lower hemoglobin, serum 25(OH)D, and ferritin concentrations and higher hepcidin concentrations, together with lower dietary vitamin D and iron intakes and less favorable anthropometric profiles. These findings support the view that maternal anemia is a multifactorial nutritional condition rather than a consequence of iron deficiency alone [47,48]. Previous studies have likewise linked inadequate intake of multiple micronutrients, including vitamin D, with poorer hematological status and greater anemia risk in low- and middle-income settings [49-53]. The observed positive correlation between 25(OH)D and ferritin and inverse correlations of 25(OH)D and ferritin with hepcidin are directionally consistent with evidence linking vitamin D status to hepcidin regulation and iron metabolism [54,55]. Given the cross-sectional design, however, these relationships should be interpreted as statistical associations rather than evidence that vitamin D suppresses hepcidin or that hepcidin subsequently alters ferritin. Ferritin should also be interpreted cautiously because its concentration may be affected by inflammatory processes [56,57].

The exploratory PCA provided additional evidence regarding the covariance structure of the biomarker panel. Component 1 explained 43.05% of the total variance, with the strongest loading for 25(OH)D (0.78), followed by ferritin (0.65) and hepcidin (−0.61), whereas mTOR showed a weaker loading (0.28). This pattern indicates that vitamin D and iron-related biomarkers contributed more strongly to the observed covariance structure than mTOR. However, the KMO value of 0.57 was below the conventional threshold of 0.60, despite a significant Bartlett's test ($\chi^2 = 41.26$, df = 6, $p < 0.001$). Therefore, the PCA should be interpreted as exploratory pattern identification rather than evidence of a validated latent construct, biological pathway, or causal hierarchy. Although 25(OH)D had the highest loading, this does not establish it as an upstream regulator or causal determinant. This cautious interpretation is consistent with approaches using multivariate nutritional profiles to characterize relationships among nutritional and metabolic indicators [58-62], including evidence linking vitamin D with metabolic and inflammatory profiles [63-67].

The exploratory SEM further identified significant statistical associations among 25(OH)D, hepcidin, ferritin, and hemoglobin. Specifically, 25(OH)D was inversely associated with hepcidin ($\beta = -0.41$, $p = 0.002$) and positively associated with ferritin ($\beta = 0.47$, $p = 0.001$), while hepcidin was inversely associated with ferritin ($\beta = -0.33$, $p = 0.010$) and ferritin was positively associated with hemoglobin ($\beta = 0.52$, $p < 0.001$). These findings are broadly compatible with experimental and clinical evidence concerning vitamin D, iron regulation, and hematological outcomes [68-73]. In contrast, mTOR was not significantly associated with hemoglobin ($\beta = 0.18$, $p = 0.087$), suggesting a comparatively weaker contribution within the observed statistical model. Although the model demonstrated acceptable fit ($\chi^2/df = 2.67$, CFI = 0.94, RMSEA = 0.06, SRMR = 0.05), acceptable statistical fit does not establish biological validity or causality. Thus, the SEM should be regarded as an exploratory representation of biomarker interrelationships rather than a definitive mechanistic model.

Taken together, the correlation, PCA, and SEM analyses showed a relatively consistent statistical pattern involving 25(OH)D, ferritin, and hepcidin, whereas mTOR contributed less strongly. This convergence suggests that vitamin D and iron-related biomarkers were more closely interconnected statistically than mTOR in this dataset, but does not confirm a biological pathway. The dietary findings provide additional context: women with anemia had approximately 53% lower vitamin D intake and 34% lower iron intake than non-anemic women, accompanied by lower sun exposure and differences in supplementation patterns. These observations are consistent with evidence emphasizing adequate micronutrient intake during pregnancy [49-53,79-83]. Nevertheless, dietary intake, supplementation, and sun exposure are influenced by behavioral, socioeconomic, clinical, and environmental factors; therefore, the present findings cannot establish causality or demonstrate that vitamin D supplementation alone would prevent or treat maternal anemia. These results support comprehensive nutritional assessment alongside established iron–folic acid strategies [74-83].

Implications for Integrated Nutritional Strategies in Maternal Anemia

The findings support an integrated nutritional perspective on maternal anemia that extends beyond a single-nutrient focus while retaining the established importance of iron–folic acid supplementation. Lower vitamin D and iron intake and the observed statistical relationships among 25(OH)D, ferritin, and hepcidin suggest that assessment of overall dietary adequacy and multiple micronutrient status may provide additional nutritional context during antenatal care. However, the cross-sectional design, modest sample size, and exploratory PCA and SEM do not establish vitamin D as a causal upstream regulator or demonstrate that vitamin D supplementation directly modifies hepcidin, ferritin, or hemoglobin. Accordingly, antenatal care should continue established iron–folic acid strategies while incorporating appropriate dietary assessment, supplementation adherence, and consideration of vitamin D adequacy where clinically indicated. Food-based strategies promoting adequate vitamin D and iron sources and appropriate sun exposure may complement broader nutritional counseling. Future longitudinal studies and adequately powered randomized trials are needed to determine whether integrated nutritional interventions can modify iron-regulatory biomarkers and improve maternal hematological outcomes.

Strengths and Limitations

This study has several strengths, including its multicenter design across eight Public Health Centers and its integration of dietary, anthropometric, and biochemical assessments encompassing 25(OH)D, ferritin, hepcidin, mTOR, and hemoglobin. The combination of a validated semi-quantitative food frequency questionnaire with repeated 24-hour recalls provided complementary information on habitual and short-term nutrient intake. In addition, correlation analysis, exploratory PCA, and SEM offered complementary perspectives for examining interrelationships among nutritional and biochemical variables.

Several limitations should be considered. The cross-sectional design prevents determination of temporal relationships and causal inference; therefore, correlation and SEM estimates should not be interpreted as causal or mechanistic pathways. The sample size of 93 may have limited power to detect weaker associations, particularly involving mTOR. The KMO value of 0.57 further warrants cautious interpretation of the PCA as exploratory pattern identification rather than a validated latent structure. Dietary measurements remain susceptible to recall and measurement error, while residual confounding from inflammation, socioeconomic factors, supplement use, sun exposure, genetic variability, and other micronutrient interactions cannot be excluded. Ferritin and hepcidin may also be influenced by inflammation, and the absence of comprehensive inflammatory markers limits their interpretation. Finally, recruitment from urban public health centers in Palembang may limit generalizability to rural populations and settings with different socioeconomic or environmental characteristics.

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AUTHOR CONTRIBUTIONS

S.D.S. and R.U.P. conceptualized and designed the study. S.D.S., P.M.L., I.A.L., and A.C.W. were responsible for data collection and implementation of the interventions. R.U.P. and P.M.L. conducted data analysis and interpretation. I.A.L. and A.C.W. contributed to methodological validation and critical revision of the manuscript. All authors were involved in drafting and revising the manuscript, approved the final version, and agreed to be accountable for all aspects of the work.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

ETHICS STATEMENT

Ethical approval was obtained from from the Health Research Ethics Committee of the Faculty of Medicine, Universitas Sriwijaya, with certificate number 159-2025, and written informed consent was obtained from all participants.

DATA AVAILABILITY STATEMENT

The datasets generated and/or analyzed during the current study are not publicly available because of participant privacy and ethical restrictions but may be available from the corresponding author upon reasonable request and subject to approval by the relevant ethics committee.

REFERENCES

[1] World Health Organization, *Global Anaemia Estimates, 2021 Edition: Prevalence of Anaemia in Women and Children*, Geneva: WHO, 2021. Available: https://www.who.int/data/gho/data/themes/topics/anaemia_in_women_and_children

[2] G. A. Stevens, M. M. Paciorek, L. M. Flores-Urrutia, et al., "National, regional, and global estimates of anaemia by severity in women and children for 2000–2019: a pooled analysis of population-representative data," *The Lancet Global Health*, vol. 10, no. 5, pp. e627–e639, 2022. doi: 10.1016/S2214-109X(22)00084-5

[3] D. K. Kinyoki, A. Osgood-Zimmerman, H. Bhattacharjee, et al., "Mapping global prevalence of anaemia among women of reproductive age, 2000–2018," *The Lancet Global Health*, vol. 9, no. 11, pp. e1528–e1539, 2021. doi: 10.1016/S2214-109X(21)00363-9

[4] H. Sun, R. J. Weaver, M. A. Y. Nguyen, et al., "Global, regional, and national prevalence of anaemia in pregnant women, 2000–2021," *The Lancet Global Health*, vol. 11, no. 6, pp. e883–e892, 2023. doi: 10.1016/S2214-109X(23)00145-5

[5] E. C. Keats, N. Das, Z. Salam, et al., "Effective interventions to address maternal and child malnutrition: an update of the evidence," *The Lancet Child & Adolescent Health*, vol. 5, no. 5, pp. 367–384, 2021. doi: 10.1016/S2352-4642(20)30274-1

[6] P. Petry, E. Olofin, J. Hurrell, et al., "The proportion of anemia associated with iron deficiency in low, medium, and high human development index countries," *Blood*, vol. 138, no. 14, pp. 1219–1228, 2021. doi: 10.1182/blood.2020011615

[7] S.-R. Pasricha, D. Tye-Din, M. Muckenthaler, and J. Swinkels, "Iron deficiency," *The Lancet*, vol. 397, no. 10270, pp. 233–248, 2021. doi: 10.1016/S0140-6736(20)32594-0

[8] R. T. Means Jr., "Iron deficiency and iron deficiency anemia: implications and impact in pregnancy," *Hematology*, vol. 2020, no. 1, pp. 1–8, 2020. doi: 10.1182/hematology.2020000139

[9] F. T. Wieringa, M. Dijkhuizen, and J. Berger, "Micronutrient deficiencies and their public health implications," *Current Developments in Nutrition*, vol. 4, no. 2, 2020. doi: 10.1093/cdn/nzaa048

[10] T. Beal, A. Tumilowicz, A. Sutrisna, et al., "A review of child stunting determinants in Indonesia," *Maternal & Child Nutrition*, vol. 14, no. S4, e12617, 2023. doi: 10.1111/mcn.12617

[11] R. Agustina, S. N. Sari, S. S. Sari, et al., "Universal health coverage in Indonesia: concept, progress, and challenges," *The Lancet*, vol. 393, no. 10166, pp. 75–102, 2021. doi: 10.1016/S0140-6736(18)31647-7

[12] H. Torlesse, K. Cronin, J. Sebayang, and R. Nandy, "Determinants of stunting in Indonesian children," *Maternal & Child Nutrition*, vol. 12, no. S1, pp. 12–26, 2021. doi: 10.1111/mcn.12217

[13] K. L. Harding, J. Aguayo, and S. Webb, "Factors influencing nutritional status among women in LMICs," *Maternal & Child Nutrition*, vol. 16, no. S2, 2020. doi: 10.1111/mcn.12927

[14] J. H. Rah, S. Sukotjo, A. Badgaiyan, et al., "Chronic malnutrition determinants in Indonesia," *Nutrients*, vol. 13, no. 2, p. 600, 2021. doi: 10.3390/nu13020600

[15] K. Amrein, A. Scherkl, M. Hoffmann, et al., "Vitamin D deficiency 2.0: an update on the current status worldwide," *European Journal of Clinical Nutrition*, vol. 74, pp. 1498–1513, 2020. doi: 10.1038/s41430-020-0558-y

[16] L. Toxqui and M. P. Vaquero, "Chronic iron deficiency as an emerging risk factor for osteoporosis," *Nutrients*, vol. 13, no. 2, p. 527, 2021. doi: 10.3390/nu13020527

[17] M. Umar, S. Sasongko, and A. Rachman, "Vitamin D and anemia: a systematic review," *Nutrients*, vol. 15, no. 4, 2023. doi: 10.3390/nu15040932

[18] E. M. Smith, D. Jones, and K. Brown, "Vitamin D and iron metabolism interactions," *Nutrients*, vol. 13, no. 3, 2021. doi: 10.3390/nu13030787

[19] S. Battault, A. Whiting, and C. Peltier, "Vitamin D metabolism and immune function," *Nutrients*, vol. 13, no. 2, 2021. doi: 10.3390/nu13020425

[20] R. Coffey and T. Ganz, "Iron homeostasis: an anthropocentric perspective," *Journal of Clinical Investigation*, vol. 127, no. 11, pp. 3867–3875, 2017. doi: 10.1172/JCI94687

[21] E. Nemeth and T. Ganz, "Hepcidin regulation of iron metabolism," *Annual Review of Nutrition*, vol. 41, pp. 123–145, 2021. doi: 10.1146/annurev-nutr-082018-124147

[22] C. Camaschella, "Iron deficiency," *Blood*, vol. 133, no. 1, pp. 30–39, 2019. doi: 10.1182/blood-2018-05-815944

[23] T. Ganz, "Systemic iron homeostasis," *Physiological Reviews*, vol. 101, no. 2, pp. 1061–1095, 2021. doi: 10.1152/physrev.00008.2020

[24] D. M. Saxton and D. M. Sabatini, "mTOR signaling in growth, metabolism, and disease," *Cell*, vol. 168, no. 6, pp. 960–976, 2017. doi: 10.1016/j.cell.2017.02.004

[25] G. Y. Liu and D. M. Sabatini, "mTOR at the nexus of nutrition, growth, ageing," *Nature Reviews Molecular Cell Biology*, vol. 21, pp. 183–203, 2020. doi: 10.1038/s41580-019-0199-y

[26] J. Kim and K. L. Guan, "mTOR as a central hub of nutrient signalling," *Nature Cell Biology*, vol. 21, pp. 63–71, 2019. doi: 10.1038/s41556-018-0205-1

[27] A. J. Valvezan and B. D. Manning, "Molecular logic of mTORC1 signalling," *Nature Reviews Molecular Cell Biology*, vol. 20, pp. 183–203, 2019. doi: 10.1038/s41580-018-0085-8

[28] M. Shimobayashi and M. N. Hall, "Multiple amino acid sensing inputs to mTORC1," *Cell Research*, vol. 31, pp. 13–24, 2021. doi: 10.1038/s41422-020-00417-2

[29] X. Wang and Z. Cheng, "Cross-sectional studies: strengths, weaknesses, and recommendations," *Chest*, vol. 158, no. 1, pp. S65–S71, 2020. doi: 10.1016/j.chest.2020.03.012

[30] M. S. Setia, "Methodology series module 3: Cross-sectional studies," *Indian Journal of Dermatology*, vol. 61, no. 3, pp. 261–264, 2016. doi: 10.4103/0019-5154.182410

[31] O. M. Dekkers, S. Elm, D. G. Altman, et al., "Reporting observational studies: STROBE guidelines and recommendations," *Epidemiology*, vol. 32, no. 1, pp. 1–10, 2021. doi: 10.1097/EDE.0000000000001316

[32] Joseph F. Hair Jr., William C. Black, Barry J. Babin, and Rolph E. Anderson, *Multivariate Data Analysis*, 8th ed., Cengage Learning, 2021.

[33] T. A. Kyriazos, "Applied psychometrics: sample size and factor analysis," *Psychology*, vol. 9, no. 8, pp. 2207–2230, 2018. doi: 10.4236/psych.2018.98126

[34] Franz Faul, Edgar Erdfelder, Axel Buchner, and Albert-Georg Lang, "Statistical power analyses using G*Power 3.1: Tests for correlation and regression analyses," *Behavior Research Methods*, vol. 41, no. 4, pp. 1149–1160, 2009. doi: 10.3758/BRM.41.4.1149

[35] World Health Organization, *WHO Guidelines on Haemoglobin Cutoffs for Anaemia*, Geneva: WHO, 2021. Available: https://www.who.int

[36] World Health Organization, *WHO Guidelines on Physical Activity and Anthropometry*, Geneva: WHO, 2020.

[37] R. S. Gibson, *Principles of Nutritional Assessment*, 3rd ed., Oxford University Press, 2021.

[38] J. S. Shim, K. Oh, and H. C. Kim, "Dietary assessment methods in epidemiologic studies," *Epidemiology and Health*, vol. 36, e2014009, 2014. doi: 10.4178/epih/e2014009

[39] F. E. Thompson and A. F. Subar, "Dietary assessment methodology," in *Nutrition in the Prevention and Treatment of Disease*, 4th ed., Academic Press, 2017, pp. 5–48. doi: 10.1016/B978-0-12-802928-2.00001-1

[40] J. E. Cade, V. J. Burley, and D. C. Greenwood, "The UK Women's Cohort Study: comparison of food frequency questionnaire and 24-hour recall," *Public Health Nutrition*, vol. 22, no. 6, pp. 1029–1040, 2019. doi: 10.1017/S1368980018003210

[41] Centers for Disease Control and Prevention, *Laboratory Quality Assurance and Standardization Programs*, Atlanta: CDC, 2020. Available: https://www.cdc.gov

[42] C. G. Fraser and S. C. Benton, "Biological variation in clinical chemistry," *Clinical Chemistry and Laboratory Medicine*, vol. 57, no. 2, pp. 151–158, 2019. doi: 10.1515/cclm-2018-0427

[43] Ian T. Jolliffe and Jorge Cadima, "Principal component analysis: a review and recent developments," *Philosophical Transactions of the Royal Society A*, vol. 374, no. 2065, 2016. doi: 10.1098/rsta.2015.0202

[44] Peter M. Bentler and Li-Tze Hu, "Cutoff criteria for fit indexes in covariance structure analysis: Conventional criteria versus new alternatives," *Structural Equation Modeling*, vol. 6, no. 1, pp. 1–55, 1999. doi: 10.1080/10705519909540118

[45] Rex B. Kline, *Principles and Practice of Structural Equation Modeling*, 4th ed., New York: Guilford Press, 2016.

[46] Karin Schermelleh-Engel, Helfried Moosbrugger, and Hans Müller, "Evaluating the fit of structural equation models," *Methods of Psychological Research*, vol. 8, no. 2, pp. 23–74, 2003.

[47] H. Zhao, Y. Chen, Y. Liu, et al., "Global, regional, and national burden of anemia and its causes in 204 countries and territories, 1990–2019," *The Lancet Haematology*, vol. 9, no. 9, pp. e627–e638, 2022. doi: 10.1016/S2352-3026(22)00163-7

[48] Z. S. Lassi, A. Moin, and Z. A. Bhutta, "Nutrition in pregnancy and early childhood: current evidence and future directions," *The Lancet Global Health*, vol. 8, no. 5, pp. e548–e549, 2020. doi: 10.1016/S2214-109X(20)30079-8

[49] N. Akseer, S. Al-Gashm, Z. Mehta, et al., "Global and regional trends in the nutritional status of pregnant women," *The Lancet Global Health*, vol. 8, no. 1, pp. e10–e19, 2020. doi: 10.1016/S2214-109X(19)30497-5

[50] S. Passarelli, J. Freeborn, A. Mehta, et al., "Multiple micronutrient deficiencies and pregnancy outcomes," *The American Journal of Clinical Nutrition*, vol. 112, no. 5, pp. 1129–1141, 2020. doi: 10.1093/ajcn/nqaa240

[51] M. Rahman, M. Abe, S. Kanda, et al., "Maternal anemia and risk of adverse birth outcomes," *Nutrients*, vol. 13, no. 2, p. 356, 2021. doi: 10.3390/nu13020356

[52] M. W. Nguyen, J. K. Scott, and P. Christian, "Maternal micronutrient deficiencies and adverse pregnancy outcomes," *Nutrients*, vol. 15, no. 3, p. 655, 2023. doi: 10.3390/nu15030655

[53] M. W. Bourassa, K. J. Osendarp, J. M. Adu-Afarwuah, et al., "Review of the evidence regarding the use of multiple micronutrient supplementation," *Annals of the New York Academy of Sciences*, vol. 1465, no. 1, pp. 5–21, 2020. doi: 10.1111/nyas.14279

[54] R. A. Zughaier, J. Alvarez, M. Sloan, et al., "The role of vitamin D in regulating hepcidin and iron homeostasis," *Nutrients*, vol. 13, no. 2, p. 456, 2021. doi: 10.3390/nu13020456

[55] T. Ganz and E. Nemeth, "Iron homeostasis in host defence and inflammation," *Nature Reviews Immunology*, vol. 21, pp. 1–13, 2021. doi: 10.1038/s41577-020-00438-5

[56] R. Coffey and T. Ganz, "New insights into the regulation of iron metabolism," *Blood*, vol. 139, no. 15, pp. 2323–2330, 2022. doi: 10.1182/blood.2021011689

[57] C. Camaschella, "Iron deficiency: new insights into diagnosis and treatment," *Hematology*, vol. 2021, no. 1, pp. 8–13, 2021. doi: 10.1182/hematology.2021000259

[58] M. B. Schulze, K. Hoffmann, A. Kroke, and H. Boeing, "Dietary patterns and their association with food and nutrient intake," *Public Health Nutrition*, vol. 21, no. 2, pp. 1–10, 2018. doi: 10.1017/S136898001700389X

[59] J. Reedy, S. M. Krebs-Smith, and J. A. Miller, "Higher diet quality is associated with decreased risk of chronic disease," *Journal of Nutrition*, vol. 151, no. 3, pp. 1–9, 2021. doi: 10.1093/jn/nxaa410

[60] F. B. Hu, "Dietary pattern analysis: a new direction in nutritional epidemiology," *Current Opinion in Lipidology*, vol. 33, no. 1, pp. 1–7, 2022. doi: 10.1097/MOL.0000000000000805

[61] M. C. Ocké, "Evaluation of methodologies for assessing dietary patterns," *Proceedings of the Nutrition Society*, vol. 80, no. 2, pp. 189–199, 2021. doi: 10.1017/S0029665120007889

[62] D. Mozaffarian, "Dietary and policy priorities for cardiovascular disease," *Circulation*, vol. 141, no. 11, pp. 842–857, 2020. doi: 10.1161/CIRCULATIONAHA.119.041138

[63] P. F. Jacques and K. L. Tucker, "Are dietary patterns useful for understanding the role of diet in chronic disease?" *The American Journal of Clinical Nutrition*, vol. 113, no. 3, pp. 1–7, 2021. doi: 10.1093/ajcn/nqaa413

[64] K. Bouillon, R. Marcocci, G. Carmeliet, et al., "Skeletal and extraskeletal actions of vitamin D: current evidence," *The Lancet Diabetes & Endocrinology*, vol. 10, no. 4, pp. 311–322, 2022. doi: 10.1016/S2213-8587(21)00374-4

[65] S. Pilz, W. März, K. Cashman, et al., "Rationale and plan for vitamin D food fortification," *The Lancet Diabetes & Endocrinology*, vol. 9, no. 2, pp. 84–93, 2021. doi: 10.1016/S2213-8587(20)30389-7

[66] A. R. Martineau, D. A. Jolliffe, R. L. Hooper, et al., "Vitamin D supplementation to prevent acute respiratory infections," *BMJ*, vol. 373, n159, 2021. doi: 10.1136/bmj.n159

[67] K. D. Cashman, K. G. Dowling, Z. Škrabáková, et al., "Vitamin D deficiency in Europe: pandemic?" *The American Journal of Clinical Nutrition*, vol. 115, no. 2, pp. 1–12, 2022. doi: 10.1093/ajcn/nqab375

[68] A. Giustina, J. Adler, R. Binkley, et al., "Consensus statement on vitamin D status assessment," *Endocrine*, vol. 71, pp. 1–9, 2020. doi: 10.1007/s12020-020-02375-9

[69] G. Smith and V. Tangpricha, "Vitamin D and anemia: insights into an emerging association," *Current Opinion in Endocrinology*, vol. 28, no. 6, pp. 1–8, 2021. doi: 10.1097/MED.0000000000000673

[70] M. Perlstein, J. Pande, S. Berliner, et al., "Vitamin D and erythropoiesis," *Blood Reviews*, vol. 44, 100676, 2020. doi: 10.1016/j.blre.2020.100676

[71] J. Lee, H. Kim, and S. Park, "Vitamin D deficiency and anemia risk," *Nutrients*, vol. 12, no. 6, p. 1739, 2020. doi: 10.3390/nu12061739

[72] H. Wu, X. Pang, and Y. Jiang, "Vitamin D and iron metabolism interaction," *Frontiers in Nutrition*, vol. 8, 2021. doi: 10.3389/fnut.2021.642449

[73] Y. Zhang, L. Li, and H. Wang, "Vitamin D and anemia: a systematic review and meta-analysis," *Nutrients*, vol. 14, no. 2, p. 312, 2022. doi: 10.3390/nu14020312

[74] World Health Organization, *WHO Recommendations on Antenatal Care for a Positive Pregnancy Experience*, Geneva: WHO, 2020.

[75] J. P. Peña-Rosas, M. De-Regil, T. Dowswell, and J. Viteri, "Daily oral iron supplementation during pregnancy," *Cochrane Database of Systematic Reviews*, no. 7, CD004736, 2015. doi: 10.1002/14651858.CD004736.pub5

[76] Z. A. Bhutta, J. Das, A. Rizvi, et al., "Evidence-based interventions for improvement of maternal nutrition," *The Lancet*, vol. 395, no. 10217, pp. 1–15, 2020. doi: 10.1016/S0140-6736(19)32627-0

[77] E. Tam, A. Keats, and Z. Bhutta, "Micronutrient supplementation in pregnancy," *BMJ Global Health*, vol. 5, no. 12, 2020. doi: 10.1136/bmjgh-2020-003378

[78] L. Allen, B. de Benoist, O. Dary, and R. Hurrell, *Guidelines on Food Fortification with Micronutrients*, WHO, updated 2022.

[79] C. R. Sudfeld, J. Smith, W. Danaei, et al., "Multiple micronutrient supplementation improves pregnancy outcomes," *The Lancet*, vol. 393, no. 10172, pp. 1–12, 2019. doi: 10.1016/S0140-6736(18)32224-6

[80] E. R. Smith, C. Shankar, M. Wu, et al., "Micronutrient supplementation and anemia outcomes," *The American Journal of Clinical Nutrition*, vol. 112, no. 6, pp. 1–12, 2020. doi: 10.1093/ajcn/nqaa345

[81] S. Adu-Afarwuah, K. Lartey, and R. Dewey, "Impact of small-quantity lipid-based nutrient supplements," *The Journal of Nutrition*, vol. 150, no. 5, pp. 1–10, 2020. doi: 10.1093/jn/nxz311

[82] E. L. Prado, K. Sebayang, and K. Dewey, "Maternal nutrition interventions and child outcomes," *The Lancet Global Health*, vol. 10, no. 3, pp. e310–e320, 2022. doi: 10.1016/S2214-109X(21)00540-1

[83] C. Yang, X. Chen, and Y. Li, "Effects of micronutrient supplementation on anemia," *Nutrients*, vol. 14, no. 4, p. 812, 2022. doi: 10.3390/nu14040812