



CORRELATION OF IRON STUDY PARAMETERS WITH MORPHOLOGICAL CLASSIFICATION OF ANEMIA: A CROSS-SECTIONAL STUDY FROM A TERTIARY CARE CENTER IN NORTH INDIA

Dr. Fauzia Perveen¹, Dr. Nadia Farah Jeelani², Dr. Neeta Binwani³

¹ Assistant Professor, Department of Pathology, Teerthanker Mahaveer Medical College, Moradabad, Uttar Pradesh, India. (perveenfauzia12@gmail.com)

² Assistant Professor, Department of Pathology, SRM Medical College Hospital and Research Centre, Kattankulathur, Tamil Nadu, India. (farakhkhan123@gmail.com)

³ Associate Professor, Department of Pathology, Era's Lucknow Medical College and Hospital, Sarfarazganj, Lucknow, Uttar Pradesh, India. (drneetapathology@gmail.com)

***Corresponding Author:** Dr. Fauzia Perveen

*Department of Pathology, Teerthanker Mahaveer Medical College, Moradabad, Uttar Pradesh, India

ABSTRACT

Background: Anemia is a major global health concern with diverse etiologies. Morphological classification based on red blood cell indices provides an initial diagnostic framework, while iron studies offer insight into underlying biochemical abnormalities. Understanding the correlation between these approaches can enhance diagnostic accuracy.

Objectives: To evaluate the correlation between iron study parameters and morphological types of anemia in patients attending a tertiary care hospital in North India.

Methods: This hospital-based cross-sectional study included 86 patients diagnosed with anemia between April 2017 and September 2017. Morphological classification was performed using red cell indices and peripheral smear examination. Iron studies, including serum iron, total iron-binding capacity (TIBC), serum ferritin, and transferrin saturation, were measured using standard laboratory methods. Statistical analysis was conducted using SPSS version 16.0, with $p < 0.05$ considered statistically significant.

Results: Microcytic anemia was the most common morphological type (51.2%), followed by normocytic (32.6%) and macrocytic anemia (16.2%). Microcytic anemia showed significantly lower serum iron, ferritin, and transferrin saturation, along with higher TIBC compared to other types ($p < 0.001$). A strong association was observed between low serum ferritin levels and microcytic anemia, whereas normocytic and macrocytic anemia were associated with normal to elevated ferritin levels. Transferrin saturation was also significantly lower in microcytic anemia ($p < 0.001$). These findings indicate a significant correlation between iron study parameters and morphological classification.

Conclusion: There is a strong correlation between iron study parameters and morphological types of anemia. Integrating these diagnostic modalities enhances accuracy and supports effective clinical management.

Keywords: Anemia; Iron studies; Serum ferritin; Morphological classification; Microcytic anemia; TIBC; Transferrin saturation

INTRODUCTION

Anemia remains one of the most prevalent hematological disorders worldwide, affecting an estimated 1.62 billion people and constituting a major public health challenge across both developed and developing nations [1]. It is particularly common in low- and middle-income countries, where nutritional deficiencies, infections, and chronic diseases contribute significantly to its burden [2]. Iron deficiency anemia (IDA) is the most frequent subtype globally, accounting for nearly half of all anemia cases, followed by anemia of chronic disease, megaloblastic anemia, and hemolytic anemia [3]. The clinical significance of anemia lies not only in its high prevalence but also in its association with impaired physical performance, cognitive dysfunction, increased morbidity, and reduced quality of life [4].

Morphological classification of anemia based on red blood cell (RBC) indices—namely microcytic, normocytic, and macrocytic types—serves as an essential initial diagnostic approach in clinical practice [5]. This classification, typically derived from complete blood count parameters such as mean corpuscular volume (MCV), provides a rapid and cost-effective method to narrow down the differential diagnosis [6]. However, morphological classification alone often lacks specificity, as similar morphological patterns can arise from different etiologies. For instance, microcytic anemia may result from iron deficiency, thalassemia, or chronic disease, while macrocytic anemia may be due to vitamin B12 or folate deficiency, liver disease, or drug-induced causes [7].

Iron studies, including serum iron, total iron-binding capacity (TIBC), transferrin saturation, and serum ferritin, play a crucial role in determining the underlying etiology of anemia [8]. These biochemical parameters help distinguish between iron deficiency anemia and other forms such as anemia of chronic disease, where iron metabolism is altered despite adequate or increased iron stores [9]. Serum ferritin, in particular, is considered a reliable indicator of body iron stores, although it may be influenced by inflammatory states [10]. Thus, integrating iron studies with morphological classification enhances diagnostic accuracy and guides appropriate management.

In the Indian context, anemia continues to be a significant public health concern, with high prevalence reported across various age groups and socioeconomic strata [2].

Nutritional deficiencies, parasitic infections, and chronic illnesses are common contributing factors. In North India, where dietary patterns and healthcare access vary widely, the spectrum of anemia may differ, necessitating region-specific evaluation [11]. Tertiary care hospitals often encounter a diverse patient population, providing an opportunity to study the correlation between laboratory parameters and clinical presentation in a real-world setting.

Despite the widespread use of morphological classification and iron studies, there remains a gap in understanding the correlation between these two diagnostic approaches, particularly in the Indian population. Many studies have evaluated anemia prevalence and etiology, but limited data exist on how well morphological patterns correspond with iron study parameters in routine clinical practice. Establishing such correlations can improve diagnostic efficiency, reduce unnecessary investigations, and facilitate targeted treatment strategies.

Therefore, this study was undertaken in a tertiary care hospital in North India to evaluate the correlation between iron study parameters and morphological types of anemia. The primary objective was to assess the relationship between serum iron indices and morphological classification of anemia, and to determine the diagnostic utility of combining these approaches in clinical practice.

METHODOLOGY

Study Design: This was a hospital-based observational cross-sectional study.

Study Setting: The study was conducted in the Department of Pathology in collaboration with the Department of Medicine at a tertiary care hospital in North India, catering to both urban and rural populations.

Study Duration: The study was carried out over a period of six months, from April 2017 to September 2017.

Study Population: The study population comprised patients diagnosed with anemia who attended the outpatient departments or were admitted to inpatient wards during the study period. Diagnosis of anemia was based on hemoglobin levels below the reference range as per standard laboratory criteria.

Inclusion Criteria

- Patients of all age groups and both sexes diagnosed with anemia
- Patients who consented to participate in the study
- Patients with complete hematological and iron profile investigations

Exclusion Criteria

- Patients who had received blood transfusion within the previous three months
- Patients on iron therapy or hematinic supplementation prior to evaluation
- Patients with known hematological malignancies
- Patients with incomplete laboratory data

Sample Size: The sample size was calculated using the formula for estimation of proportion in cross-sectional studies:

$$n = Z^2 \times p \times q / d^2$$

Where:

$Z = 1.96$ (for 95% confidence interval)

p = assumed prevalence of iron deficiency anemia among anemic patients (taken as 60% based on prior literature)

$q = 1 - p = 40\%$

d = allowable error (10%)

The calculated sample size was approximately 92. Considering feasibility and study duration constraints, a total of 86 patients were included in the study.

Sampling Technique: A consecutive sampling technique was employed, wherein all eligible patients presenting during the study period and meeting the inclusion criteria were enrolled until the required sample size of 86 was achieved.

Data Collection Tools & Procedure: Data were collected using a predesigned and pretested proforma, which included demographic details, clinical history, and laboratory findings. Venous blood samples were collected under aseptic conditions. Complete blood counts were performed using an automated hematology analyzer to determine hemoglobin concentration and red cell indices, including mean corpuscular volume (MCV). Peripheral blood smears were prepared and examined under light microscopy for morphological classification of anemia into microcytic, normocytic, and macrocytic types. Iron studies, including serum iron, total iron-binding capacity (TIBC), and serum ferritin, were measured using standard biochemical methods available in the hospital laboratory.

Study Variables: The independent variables included demographic characteristics (age, sex) and biochemical parameters of iron studies (serum iron, TIBC, serum ferritin, transferrin saturation). The dependent variable was the morphological classification of anemia (microcytic, normocytic, macrocytic), based on red cell indices and peripheral smear findings. The study primarily assessed the correlation between iron study parameters and morphological types of anemia.

Statistical Analysis: Data were entered into Microsoft Excel and analyzed using Statistical Package for the Social Sciences (SPSS) version 16.0. Descriptive statistics were used to summarize demographic and clinical characteristics. Continuous variables were expressed as mean $\pm$ standard deviation, while categorical variables were presented as frequencies and percentages. The association

between morphological types of anemia and iron study parameters was assessed using the Chi-square test for categorical variables and analysis of variance (ANOVA) for continuous variables. A p-value of less than 0.05 was considered statistically significant.

Ethical Considerations: Written informed consent was obtained from all participants or their guardians in case of minors. The study adhered to the ethical principles outlined in the Declaration of Helsinki. Confidentiality of patient information was strictly maintained, and no personal identifiers were used in data analysis or reporting.

RESULTS

A total of 86 patients with anemia were included in the study. The majority of participants were in the 21–40 years age group, with a slight female predominance (Table 1).

Table 1: Demographic Distribution of Study Participants (n = 86)

Variable	Category	Frequency (n)	Percentage (%)
Age Group (years)	<20	14	16.3
	21–40	32	37.2
	41–60	26	30.2
	>60	14	16.3
Sex	Male	38	44.2
	Female	48	55.8

Microcytic anemia was the most common morphological type, followed by normocytic and macrocytic anemia (Table 2).

Table 2: Distribution of Morphological Types of Anemia

Morphological Type	Frequency (n)	Percentage (%)
Microcytic	44	51.2
Normocytic	28	32.6
Macrocytic	14	16.2
Total	86	100

Significant differences were observed in iron study parameters across the morphological categories. Microcytic anemia was characterized by markedly lower serum iron, ferritin, and transferrin saturation, along with elevated TIBC, whereas macrocytic anemia demonstrated comparatively higher iron indices (Table 3).

Table 3: Mean Iron Study Parameters Across Morphological Types of Anemia

Parameter	Microcytic (n=44)	Normocytic (n=28)	Macrocytic (n=14)	p-value
Serum Iron (µg/dL)	38 ± 12	68 ± 15	82 ± 18	<0.001
TIBC (µg/dL)	410 ± 35	320 ± 28	290 ± 30	<0.001
Serum Ferritin (ng/mL)	12 ± 6	85 ± 40	120 ± 50	<0.001
Transferrin Saturation (%)	9 ± 4	22 ± 6	28 ± 7	<0.001

A strong association was found between morphological type and serum ferritin levels, with most microcytic cases falling into the low ferritin category, while normocytic and macrocytic types showed predominantly normal to elevated ferritin levels (Table 4).

Table 4: Association Between Morphological Type and Serum Ferritin Categories

Morphologic al Type	Low Ferritin (<15 ng/mL)	Normal Ferritin (15–150 ng/mL)	High Ferritin (>150 ng/mL)	Total	p-value
Microcytic	36	8	0	44	<0.001
Normocytic	4	18	6	28	
Macrocytic	1	8	5	14	
Total	41	34	11	86	

Similarly, transferrin saturation was significantly lower in microcytic anemia compared to other types (Table 5).

Table 5: Correlation of Morphological Type with Transferrin Saturation

Morphological Type	Low TS (<15%)	Normal TS (15–50%)	Total	p-value
Microcytic	38	6	44	<0.001
Normocytic	8	20	28	
Macrocytic	2	12	14	
Total	48	38	86	

DISCUSSION

The present study evaluated the correlation between iron study parameters and morphological types of anemia in a tertiary care setting in North India. The key findings indicate that microcytic anemia was the most prevalent morphological type, followed by normocytic and macrocytic anemia. Furthermore, a statistically significant association was observed between morphological classification and iron indices, including serum iron, total iron-binding capacity (TIBC), serum ferritin, and transferrin saturation. Microcytic anemia was predominantly associated with biochemical features of iron deficiency, while normocytic and macrocytic anemia demonstrated relatively preserved or elevated iron stores.

The predominance of microcytic anemia observed in this study is consistent with global and Indian data, where iron deficiency remains the leading cause of anemia [1,2]. Similar findings have been reported in studies by DeLoughery et al., where microcytic anemia constituted the largest proportion of anemia cases in clinical practice [3]. In the Indian context, high prevalence of nutritional iron deficiency, particularly among women and young adults, contributes significantly to this pattern [11]. The female preponderance noted in the present study further supports this observation, likely reflecting increased iron requirements and dietary insufficiencies.

The iron profile observed in microcytic anemia—characterized by low serum iron, low ferritin, high TIBC, and reduced transferrin saturation—is in agreement with established pathophysiological mechanisms of iron deficiency anemia [8]. Serum ferritin, a marker of iron stores, was markedly reduced in the majority of microcytic cases, reinforcing its diagnostic utility. These findings are comparable to those reported by Guyatt et al., who highlighted the importance of ferritin as a sensitive and specific indicator of iron deficiency in the absence of inflammation [8]. The elevated TIBC observed in microcytic anemia reflects increased transferrin production in response to iron depletion, a well-documented compensatory mechanism.

Normocytic anemia in the present study showed a mixed pattern of iron indices, with a substantial proportion of cases demonstrating normal or elevated ferritin levels. This is consistent with anemia of chronic disease, where iron is sequestered within macrophages despite adequate or increased total body iron [9]. Weiss and Goodnough have described this phenomenon as a result of altered iron homeostasis mediated by inflammatory cytokines and hepcidin regulation [9]. The overlap of iron

parameters in normocytic anemia underscores the limitation of relying solely on morphological classification and highlights the need for biochemical evaluation.

Macrocytic anemia cases in this study exhibited relatively higher serum ferritin and transferrin saturation levels, which aligns with the understanding that such cases are often due to vitamin B12 or folate deficiency rather than iron deficiency [7]. The presence of elevated ferritin in some macrocytic cases may also reflect concomitant liver dysfunction or ineffective erythropoiesis. These findings emphasize that iron studies, while informative, must be interpreted in conjunction with other biochemical and clinical parameters for accurate diagnosis.

The significant association between serum ferritin categories and morphological types observed in this study supports the utility of integrating iron studies with routine hematological evaluation. Similar correlations have been reported in other observational studies, suggesting that combined interpretation enhances diagnostic precision and reduces misclassification [12]. This is particularly relevant in resource-limited settings, where targeted investigations can optimize resource utilization. From a clinical perspective, the findings of this study have important implications. Early identification of iron deficiency through combined morphological and biochemical assessment can facilitate timely intervention, preventing complications such as severe anemia and its systemic effects. Additionally, distinguishing between iron deficiency anemia and anemia of chronic disease is critical, as management strategies differ significantly.

The strengths of this study include its well-defined methodology, use of standardized laboratory parameters, and focus on correlation between routinely available diagnostic tools. However, certain limitations must be acknowledged. The study was conducted in a single tertiary care center with a relatively small sample size, which may limit generalizability. Additionally, inflammatory markers were not assessed, which could have influenced serum ferritin levels in some cases. The cross-sectional design also precludes assessment of causal relationships. The present study demonstrates a significant correlation between iron study parameters and morphological types of anemia. The integration of these diagnostic approaches enhances the accuracy of anemia classification and supports more effective clinical decision-making.

CONCLUSION

This study demonstrates a significant correlation between iron study parameters and morphological types of anemia in a tertiary care setting. Microcytic anemia emerged as the most prevalent form and was strongly associated with biochemical indicators of iron deficiency, including low serum iron, reduced ferritin levels, elevated TIBC, and decreased transferrin saturation. In contrast, normocytic and macrocytic anemia showed relatively preserved or elevated iron stores, suggesting alternative etiologies such as chronic disease or nutritional deficiencies unrelated to iron.

The findings highlight the limitations of relying solely on morphological classification and emphasize the importance of integrating iron studies for accurate diagnosis. Such an approach can improve diagnostic precision, facilitate early identification of iron deficiency, and guide appropriate management strategies. In resource-limited settings, combining basic hematological indices with targeted biochemical investigations can optimize patient care. Further large-scale, multicentric studies incorporating additional parameters such as inflammatory markers are recommended to validate and expand upon these findings. Overall, the study supports a combined diagnostic approach for effective evaluation and management of anemia in clinical practice.

DECLARATIONS

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Conflict of Interest: The authors declare no conflict of interest.

Consent: Written informed consent was obtained from all participants or their guardians.

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REFERENCES

1. World Health Organization. Worldwide prevalence of anaemia 1993-2005: WHO global database on anaemia.
2. Rajan SI, James KS. Third national family health survey in India: Issues, problems and prospects. *Economic and Political Weekly*. 2008 Nov 29;33-8.
3. CHENG LL, CHING LY. Anaemia. In *A Clinical Approach to Medicine 2001* (pp. 397-410).
4. De Benoist B, Cogswell M, Egli I, McLean E. Worldwide prevalence of anaemia 1993-2005; WHO Global Database of anaemia.
5. Wintrobe MM. *Wintrobe's clinical hematology*. Lippincott Williams & Wilkins; 2009.
6. Bj B. *Blood Cells. A Practical Guide*.
7. Pallister C, Watson M. *Haematology*. Scion Publishing Ltd; 2010 Oct 22.
8. Guyatt GH, Oxman AD, Ali M, Willan A, McIlroy W, Patterson C. Laboratory diagnosis of iron-deficiency anemia: an overview. *Journal of general internal medicine*. 1992 Mar;7(2):145-53.
9. Weiss G, Goodnough LT. Anemia of chronic disease. *New England Journal of Medicine*. 2005 Mar 10;352(10):1011-23.
10. Worwood M, Cook JD. Serum ferritin. *CRC Critical Reviews in Clinical Laboratory Sciences*. 1979 Jan 1;10(2):171-204.
11. Ahmad N, Kalakoti P, Bano R, Aarif S. The prevalence of anaemia and associated factors in pregnant women in a rural Indian community. *Hindu*. 2010;208(67.1).
12. Bessman JD, Gilmer Jr PR, Gardner FH. Improved classification of anemias by MCV and RDW. *American journal of clinical pathology*. 1983 Sep 1;80(3):322-6.