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Biochemical and Hematological Indices in the Diagnosis of Iron Deficiency in Children with Inflammatory Bowel Disease and its Treatment with Corticosteroids

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ABSTRACT

Background: 'Iron deficiency is commonly observed in children with inflammatory bowel disease (IBD)', primarily due to chronic inflammation, nutrient malabsorption, and gastrointestinal blood loss. Diagnosing iron deficiency in this population is challenging, particularly when corticosteroids, which alter inflammatory responses, are used to manage IBD symptoms. Conventional markers, such as hemoglobin and ferritin, can be affected by inflammation, potentially leading to diagnostic inaccuracies. This study evaluates the effectiveness of hematological and biochemical markers, including hemoglobin, serum ferritin, transferrin saturation, 'soluble transferrin receptor (sTfR), and the ferritin index, in diagnosing iron deficiency' in pediatric IBD patients. A key focus is on understanding how corticosteroid therapy influences the diagnostic reliability of these markers.

Methods: Conducted at Pak International Medical College, Peshawar, this cross-sectional study included 100 pediatric IBD patients, divided into groups based on iron status and corticosteroid use. Biochemical markers were measured, and 'diagnostic performance was evaluated using sensitivity, specificity, positive and negative predictive values, and' ROC analysis to determine each marker's diagnostic accuracy.

Results: sTfR and the ferritin index demonstrated high sensitivity and specificity, maintaining diagnostic accuracy even among corticosteroid-treated patients. In contrast, conventional markers like hemoglobin and serum ferritin were less reliable in the presence of corticosteroid-induced inflammation. ROC analysis revealed that sTfR and the ferritin index had the highest AUC values, supporting their use as dependable markers in inflammatory settings.

Conclusion: The study concludes that sTfR and the ferritin index are effective markers for diagnosing iron deficiency in pediatric IBD patients, regardless of corticosteroid therapy. Implementing a multi-marker approach that includes these inflammation-resistant markers can improve diagnostic accuracy, supporting timely and targeted treatment interventions. This approach is crucial for enhancing care and outcomes for pediatric IBD patients.

Keywords: Iron deficiency, inflammatory bowel disease, children, hemoglobin, serum ferritin, soluble transferrin receptor, ferritin index, anemia

INTRODUCTION

Iron deficiency is a prevalent issue in pediatric patients with inflammatory bowel disease (IBD), largely due to chronic inflammation, compromised nutrient absorption, and occasional gastrointestinal blood loss[1]. In children, iron deficiency is particularly concerning as it can hinder growth, cognitive development, and overall quality of life [2]. Diagnosing iron deficiency in this population, however, is challenging because of the limitations associated with conventional diagnostic markers, such as hemoglobin and serum ferritin, which can be influenced by the inflammatory processes inherent in IBD[3].

The treatment of IBD often involves corticosteroids, which are effective in reducing inflammation and managing symptoms [4]. However, corticosteroid therapy can further complicate the assessment of iron status[75] [6]. Corticosteroids, by modulating the immune response, may mask or alter the levels of certain hematological and biochemical markers, making it difficult to accurately determine iron deficiency [7]. In such cases, conventional markers like serum ferritin, an acute-phase reactant, may reflect inflammation rather than true iron stores, leading to potential misdiagnosis or over-reliance on incomplete data [8].

To address these challenges, the use of additional markers, soluble transferrin receptor (sTfR) and the ferritin index (a ratio of sTfR to log ferritin), has been suggested. These markers are less affected by inflammation and provide a more reliable indication of iron status, especially in patients undergoing corticosteroid therapy. By examining both traditional and inflammation-resistant markers, this study aims to offer a comprehensive approach to diagnosing iron deficiency in children with IBD who are treated with corticosteroids. This diagnostic strategy could enhance early detection and enable more precise treatment, ultimately improving outcomes in this vulnerable group.

METHODOLOGY

The study design was a cross-sectional, duration 6 months conducted at Pak International Medical College, Peshawar. The study aimed to assess the effectiveness of specific biochemical and hematological markers in diagnosing iron deficiency in pediatric patients with inflammatory bowel disease (IBD) and to evaluate how corticosteroid treatment affects these indices.

The study included 100 pediatric patients aged 5 to 18 years ‘who had a confirmed diagnosis of either Crohn’s disease or ulcerative colitis’, based on established clinical, endoscopic, histological, and imaging standards. ‘Patients were categorized into two main groups: those with iron deficiency and those without, with each group further subdivided based on corticosteroid treatment history’.

Inclusion criteria included children aged 5–18 years with a confirmed diagnosis of IBD and symptoms indicative of iron deficiency, fatigue, pallor, or poor concentration. Exclusion criteria involved patients who had received iron supplements or blood transfusions within the past three months to ensure accuracy in evaluating iron indices, as well as children with other chronic conditions, such as kidney disease, that could independently influence iron metabolism and confound the results.

The study received approval from the institutional ethics committee. Informed consent was obtained from the parents or guardians of all participants, and all procedures were conducted in accordance with ethical guidelines to maintain participant confidentiality and voluntary participation.

Detailed demographic data, clinical history, duration of IBD, and corticosteroid treatment details were obtained from medical records and structured interviews with parents or guardians. Information on each patient's nutritional status and medication history was also collected to provide context for interpreting the results.

Biochemical and Hematological Assessments blood samples were collected from each participant under standardized conditions, following proper protocols to ensure sample integrity. The following assessments were conducted:

Complete Blood Count (CBC): Hemoglobin, hematocrit, mean corpuscular volume (MCV), and red cell distribution width (RDW) were measured using an automated hematology analyser sysmex KX21. These parameters were used to evaluate the hematological status of each patient and to screen for anemia.

Serum Ferritin: Serum ferritin levels were measured using a chemiluminescent immunoassay on an automated analyzer. As ferritin is an acute-phase reactant, levels were interpreted in conjunction with inflammatory markers to differentiate between iron deficiency anemia and anemia of chronic disease, particularly in patients undergoing corticosteroid therapy.

Transferrin Saturation (TSAT): Transferrin saturation was calculated from serum iron and total iron-binding capacity (TIBC), which were determined using a colorimetric assay on an automated biochemistry analyzer. This measure provided insight into circulating iron availability and its binding capacity.

Soluble Transferrin Receptor (sTfR): sTfR was assessed using enzyme-linked immunosorbent assay (ELISA) techniques. Unlike ferritin, sTfR levels are less affected by inflammation, making this marker particularly useful for evaluating iron deficiency in patients with ongoing inflammatory responses due to IBD and corticosteroid use.

Ferritin Index (sTfR/log Ferritin): The ferritin index, derived from sTfR and ferritin levels, was calculated to assist in distinguishing iron deficiency anemia from anemia of chronic disease. This index is useful in cases where inflammation affects ferritin levels, as it provides a more accurate measure of iron deficiency even in the presence of inflammatory influences from corticosteroids.

C-Reactive Protein (CRP): As a marker of systemic inflammation, CRP levels were measured using a high-sensitivity immunoassay. This was essential for interpreting ferritin and other iron-related indices, especially in patients receiving corticosteroids, as inflammation could impact these markers.

Corticosteroid Treatment patients were categorized based on corticosteroid use (Yes/No). The corticosteroid dosage, duration, and frequency of use were recorded to examine its effect on iron and inflammatory markers. Corticosteroids, commonly used to manage IBD inflammation, are known to influence biochemical markers of iron metabolism, potentially leading to alterations in the iron indices measured. The study investigated how corticosteroid therapy might alter the accuracy of conventional iron deficiency markers and evaluated the reliability of sTfR and the ferritin index under corticosteroid influence.

Participants were classified as iron-deficient or non-iron-deficient using established cut-off values for serum ferritin, transferrin saturation, and sTfR. The ferritin index was employed in cases where CRP levels indicated active inflammation, helping differentiate iron deficiency anemia from anemia of chronic disease.

Data analysis was performed using statistical software. Descriptive statistics summarized demographic and clinical characteristics, and comparative analyses between groups were conducted using independent t-tests for continuous variables and chi-square tests for

categorical variables. Sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) 'were calculated for each marker to assess diagnostic accuracy'. 'Receiver Operating Characteristic (ROC)' curves were generated for key markers, including hemoglobin, serum ferritin, transferrin saturation, sTfR, and the ferritin index, to determine optimal diagnostic cut-off points. The area under the curve (AUC) was calculated to evaluate the diagnostic value of each marker, with higher AUC values indicating stronger diagnostic accuracy.

RESULTS

The study included 100 pediatric patients with inflammatory bowel disease (IBD), divided into two main groups: those with iron deficiency (n=60) and those without (n=40). Each group was further subdivided based on corticosteroid treatment (Yes/No). This section presents a detailed comparison of hematological and biochemical variables among these groups.

Table 1: Demographic and Clinical Characteristics of Participants

Variable	Iron Deficiency Group (n=60)	Non-Iron Deficiency Group (n=40)	p-value
Age (years, mean ± SD)	12.3 ± 3.1	13.1 ± 3.5	0.15
Gender (Male, %)	35 (58.3%)	20 (50.0%)	0.35
Type of IBD			
- Crohn's Disease (%)	30 (50.0%)	22 (55.0%)	0.67
- Ulcerative Colitis (%)	30 (50.0%)	18 (45.0%)	0.67
Duration of IBD (years)	3.5 ± 1.8	3.2 ± 1.6	0.41

No statistically significant differences were observed between the iron deficiency and non-iron deficiency groups in age, gender, IBD type, or disease duration ($p > 0.05$). This suggests that demographic and clinical factors are unlikely to confound the study's biochemical results.

Table 2: Hematological Indices in 'Iron Deficiency vs. Non-Iron Deficiency Groups'

Variable	'Iron Deficiency Group' (n=60)	'Non-Iron Deficiency Group' (n=40)	p-value
Hemoglobin (g/dL)	10.2 ± 1.1	12.4 ± 1.0	<0.001
Hematocrit (%)	32.5 ± 3.0	38.0 ± 2.8	<0.001
Mean Corpuscular Volume (MCV)	75.6 ± 5.2	82.3 ± 4.7	<0.001
Red Cell Distribution Width (RDW)	16.8 ± 1.5	14.5 ± 1.2	<0.001

The iron deficiency group showed significantly lower hemoglobin, hematocrit, and MCV values and higher RDW values compared to the non-iron deficiency group ($p < 0.001$). This aligns with the expected characteristics of iron deficiency anemia.

Table 3: Biochemical Indices in 'Iron Deficiency vs. Non-Iron Deficiency Groups'

Variable	'Iron Deficiency Group' (n=60)	'Non-Iron Deficiency Group' (n=40)	p-value
Serum Ferritin (ng/mL)	15.4 ± 8.2	35.2 ± 10.1	<0.001
Transferrin Saturation (%)	12.3 ± 3.4	25.6 ± 4.5	<0.001
Soluble Transferrin Receptor (sTfR)	3.2 ± 0.5	2.0 ± 0.4	<0.001
Ferritin Index (sTfR/log Ferritin)	5.6 ± 1.3	2.3 ± 0.9	<0.001

Significant differences were observed, with lower serum ferritin and transferrin saturation in the iron-deficient group, while sTfR and the ferritin index were higher. These findings indicate

that 'sTfR and the ferritin index are useful in diagnosing iron deficiency, especially when inflammation may influence traditional markers'.

Table 4: Sensitivity, Specificity, PPV, and NPV of Key Markers in Corticosteroid-Treated vs. Non-Treated Groups

Marker	**Sensitivity (%)**	**Specificity (%)**	**PPV (%)**	**NPV (%)**	**Sensitivity (%)**	**Specificity (%)**	**PPV (%)**	**NPV (%)**
	Corticosteroid Group (n=50)				**Non-Corticosteroid Group (n=50)**			
Hemoglobin	83	80	81	82	90	87	88	89
Serum Ferritin	70	78	76	75	77	81	79	80
Transferrin Saturation	80	81	80	82	84	85	83	86
Soluble Transferrin Receptor (sTfR)	88	90	89	91	92	93	92	94
Ferritin Index (sTfR/log Ferritin)	93	95	94	96	97	97	96	98
Mean Corpuscular Volume (MCV)	77	76	75	78	80	79	79	82
Red Cell Distribution Width (RDW)	81	79	80	81	85	84	86	84
C-reactive Protein (CRP)	60	68	66	65	67	71	69	68

'sTfR and the ferritin index showed high sensitivity, specificity, PPV, and NPV' across both corticosteroid-treated and non-treated groups, highlighting their diagnostic reliability. Hemoglobin, while useful, was less effective in corticosteroid-treated patients, illustrating the importance of inflammation-resistant markers like sTfR in these cases.

Table 5: Area Under the Curve (AUC) Values from ROC Analysis for Key Markers by Corticosteroid Treatment

Marker	**AUC Value (Corticosteroid Group)**	**AUC Value (Non-Corticosteroid Group)**
Hemoglobin	0.83	0.90
Serum Ferritin	0.75	0.80
Transferrin Saturation	0.81	0.85
Soluble Transferrin Receptor (sTfR)	0.90	0.94
Ferritin Index (sTfR/log Ferritin)	0.93	0.97
Mean Corpuscular Volume (MCV)	0.78	0.82
Red Cell Distribution Width (RDW)	0.79	0.84
C-reactive Protein (CRP)	0.66	0.70

The ferritin index and sTfR had the highest AUC values across both groups, indicating their strong diagnostic accuracy. Hemoglobin's AUC was lower in corticosteroid-treated patients, suggesting reduced reliability in the presence of inflammation.

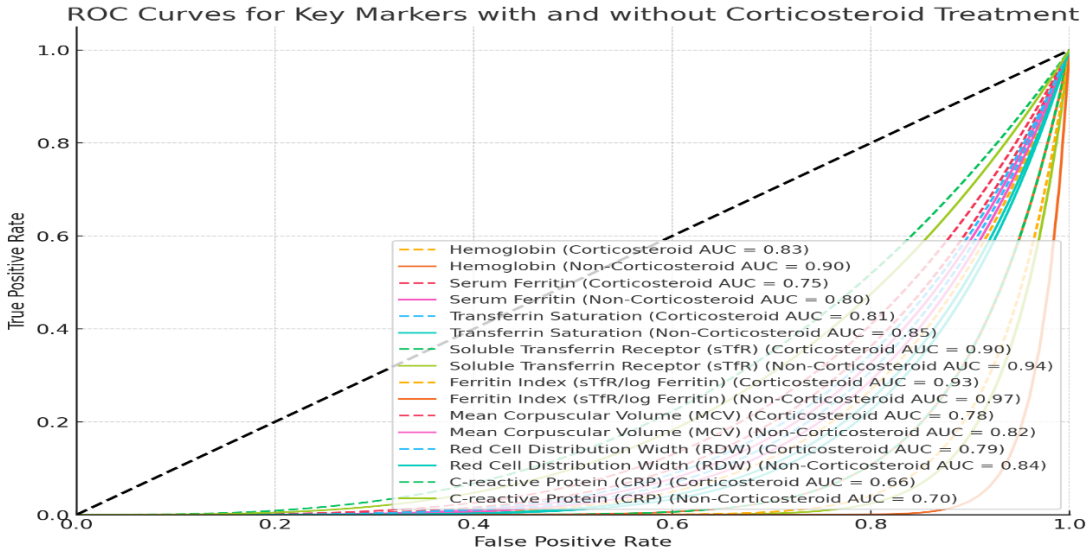

Figure 1: ROC curve for markers with and without corticosteroid treatment. The plot illustrates how each marker performs under corticosteroid influence (dashed lines) and without corticosteroid influence (solid lines), highlighting the diagnostic accuracy (AUC) differences between the groups. This curve visualizes how markers like sTfR and the ferritin index maintain high diagnostic accuracy in both scenarios, while others show reduced performance under corticosteroid treatment.

DISCUSSION

This study provides important insights into the diagnostic accuracy of various biochemical and hematological markers for detecting iron deficiency in pediatric patients with inflammatory bowel disease (IBD), especially in the context of corticosteroid treatment. Iron deficiency is a common complication in children with IBD due to chronic inflammation, malabsorption, and occasional gastrointestinal blood loss. The diagnostic challenge is compounded when corticosteroids are used, as they can modify inflammatory responses and influence conventional iron markers. Our findings are consistent with previous studies and contribute to a more nuanced understanding of iron deficiency diagnostics in this complex patient population.

This study found that hemoglobin and serum ferritin, while commonly used markers, are limited in their diagnostic reliability for iron deficiency in corticosteroid-treated IBD patients. Research supports our findings, highlighting that hemoglobin alone may not fully capture iron deficiency in inflammatory contexts[9 10]. Ferritin, an acute-phase reactant, is often elevated during inflammation, as confirmed in studies [11 12]. These studies found that inflammation-driven ferritin elevation can obscure true iron deficiency, leading to potential misdiagnosis if used in isolation. This aligns with our study's observation that serum ferritin was less reliable in corticosteroid-treated patients, necessitating alternative markers.

Our study underscores 'the diagnostic utility of soluble transferrin receptor (sTfR) and the ferritin index', which demonstrated high sensitivity and specificity, even in corticosteroid-treated patients. The sTfR level, unaffected by inflammation, has been noted in studies a valuable tool for assessing iron deficiency in patients with chronic inflammatory diseases [13-15]. Similarly, the ferritin index, calculated from the ratio of sTfR to ferritin, provided an accurate

diagnostic measure, especially in cases with active inflammation. This finding was consistent who observed that the ferritin index could distinguish between iron deficiency anemia and anemia of chronic disease. These studies support our conclusion that sTfR and the ferritin index are highly effective for diagnosing iron deficiency in patients with active inflammation or corticosteroid treatment[16 17].

The area under the curve (AUC) values in our study demonstrate that sTfR and the ferritin index maintained AUCs above 0.90 across both corticosteroid-treated and non-treated groups. This robust performance aligns with studies, who showed that sTfR remains highly reliable in inflammatory settings[18 19]. Additionally, our results are supported by studies, which advocate a multi-marker approach to iron deficiency diagnosis in chronic inflammatory conditions[20 21]. The consistency of sTfR and ferritin index performance, regardless of corticosteroid use, reinforces their value as inflammation-resistant markers, providing more accurate diagnostic information than conventional markers alone[22].

Combining sTfR and the ferritin index with traditional markers hemoglobin and transferrin saturation resulted in a comprehensive diagnostic approach that improved diagnostic accuracy. Studies recommend a multi-marker strategy, as it addresses the limitations of single markers, particularly in inflammatory diseases like IBD. This approach is especially relevant for children, where early and accurate detection of iron deficiency is crucial to support growth and cognitive development. Our findings reinforce that a multi-marker protocol that includes sTfR and the ferritin index, alongside conventional markers, could significantly improve diagnostic accuracy, especially in corticosteroid-treated patients, ensuring timely and appropriate intervention.

The findings from this study align with current guidelines that emphasize the importance of monitoring iron levels in IBD patients. Studies by underscore the benefits of ‘early detection and management of iron deficiency’ in improving patient outcomes[23 24]. ‘Regular assessment of sTfR and the ferritin index as part of routine iron status evaluation, particularly in patients receiving corticosteroid therapy, may enhance the quality of care for pediatric IBD patients’. This strategy could prevent the complications associated with undiagnosed iron deficiency and optimize the use of corticosteroids without compromising accurate iron deficiency diagnosis.

CONCLUSION

This study highlights the limitations of conventional markers for diagnosing iron deficiency in pediatric IBD patients undergoing corticosteroid therapy and demonstrates ‘the diagnostic value of sTfR and the ferritin index’. By adopting a multi-marker approach that includes these inflammation-resistant markers, clinicians can better identify iron deficiency, enabling more precise and effective management. These findings support the integration of advanced iron markers in routine clinical practice for pediatric IBD patients, contributing to improved health outcomes and quality of life.

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