

## Original Research Article

# HAEMATOLOGICAL PROFILE AND MENTZER INDEX IN ADULT PATIENTS PRESENTING WITH MICROCYTIC ANEMIA: A CROSS-SECTIONAL OBSERVATIONAL STUDY

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Received : 13/09/2025  
Received in revised form : 28/10/2025  
Accepted : 17/11/2025

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DOI: 10.70034/ijmedph.2025.4.266

Source of Support: Nil,  
Conflict of Interest: None declared

Int J Med Pub Health  
2025; 15 (4); 1486-1491

## ABSTRACT

**Background:** Anemia remains a significant public health concern globally, particularly in developing countries like India, where underdiagnosis in adults contributes to morbidity. Differentiating iron deficiency anemia (IDA) from  $\beta$ -thalassemia trait (BTT)—the two major causes of microcytic anemia—is essential due to their differing management strategies. The Mentzer Index (MCV/RBC count) has emerged as a potential screening tool, with values  $<13$  suggesting BTT and  $>13$  suggesting IDA.

**Materials and Methods:** A retrospective, cross-sectional study was conducted over one year in the Department of Pathology at a tertiary care hospital. A total of 120 adult patients ( $>18$  years) with microcytic anemia and complete laboratory data were included. Hematological indices, iron studies, and hemoglobin electrophoresis results were analyzed. Patients were categorized into BTT or IDA based on confirmatory tests, and Mentzer Index was calculated for all. Statistical analysis was performed using SPSS v26.0, with  $p < 0.05$  considered significant.

**Results:** Out of 120 patients, 111 (92.5%) were diagnosed with IDA and 9 (7.5%) with BTT. A Mentzer Index  $>13$  correctly classified 108 IDA cases, while a value  $<13$  identified 8 of 9 BTT cases. BTT patients had significantly higher RBC counts ( $5.6 \pm 0.7$  vs  $3.8 \pm 0.6 \times 10^6 / \mu\text{L}$ ), lower MCV ( $64.2 \pm 4.9$  vs  $72.8 \pm 7.5$  fL), lower RDW ( $13.8 \pm 1.1\%$  vs  $17.6 \pm 2.2\%$ ), and normal iron studies. The Mentzer Index demonstrated 88.9% sensitivity, 97.3% specificity, 72.7% positive predictive value (PPV), 99.1% negative predictive value (NPV), and 96.7% overall diagnostic accuracy for BTT detection.

**Conclusion:** The Mentzer Index offers a simple, cost-effective, and reliable screening tool to differentiate BTT from IDA in adults with microcytic anemia. Its high specificity and NPV make it particularly effective in ruling out BTT. However, confirmatory iron studies and hemoglobin electrophoresis remain essential for definitive diagnosis.

**Keywords:** Anemia, Iron Deficiency, Beta-Thalassemia Trait, Mentzer Index, Microcytic Anemia.

## INTRODUCTION

Anemia is a common yet significant global health problem that affects over two billion individuals worldwide and contributing significantly to morbidity and decreased quality of life. It is more common in developing countries including India.<sup>[1]</sup>

Among adults the burden of anemia is often underrecognized. Consequences of such an unrecognised anemia range from reduced physical performance and cognitive function to adverse maternal and perinatal outcomes in women. The World Health Organization (WHO) defines anemia in adults as hemoglobin (Hb) levels  $<13.0$  g/dL in

men and  $<12.0$  g/dL in non-pregnant women. These thresholds corresponds to diminished capacity of the blood to carry oxygen effectively. In low-resource settings anemia frequently not only remains undiagnosed but also misclassified due to the absence of accessible diagnostic tools.<sup>[2]</sup>

The etiology of anemia in adults is multifactorial and include nutritional deficiencies, chronic inflammatory diseases, malignancies, renal insufficiency and inherited hematological disorders. Among this iron deficiency anemia (IDA) is the most prevalent subtype globally, particularly in women of reproductive age and in individuals with poor dietary intake, chronic gastrointestinal blood loss, or malabsorption syndromes. In regions with high carrier rates, beta-thalassemia trait (BTT) remains a significant differential diagnosis for microcytic anemia. Differentiating between iron deficiency anemia and beta thalassemia trait is important not only because of different therapeutic approaches but also because misdiagnosis can lead to inappropriate treatment. For instance, prescription of empirical iron supplementation in cases of BTT may lead to iron overload with long-term administration.<sup>[3]</sup>

Confirmatory tests for diagnosis of thalassemia include hemoglobin electrophoresis and high-performance liquid chromatography (HPLC). Though diagnostic, these tests are not universally accessible or affordable particularly in rural or resource-limited healthcare settings. Moreover, routine use of such advanced diagnostics in every patient presenting with microcytic anemia is often not practical because of availability and affordability issues. Therefore there is need for simple, cost-effective screening test that can help differentiate IDA from BTT with reasonable accuracy. One such tool is the Mentzer Index which can easily be calculated by dividing the mean corpuscular volume (MCV) by the red blood cell (RBC) count.<sup>[4]</sup>

The Mentzer index was first Proposed by William C. Mentzer in 1973. It is based on the observation that in IDA the MCV tends to be low due to small erythrocytes while the RBC count is also decreased due to impaired erythropoiesis. In contrast BTT is characterized by microcytosis with a normal or even elevated RBC count as a result of ineffective erythropoiesis and compensatory marrow hyperactivity.<sup>[5]</sup> A Mentzer Index value more than 13 is highly suggestive of IDA while a value less than 13 favors diagnosis of BTT. This cutoff value has been validated across multiple pediatric and some adult cohorts. Its ease of calculation from a standard complete blood count (CBC) makes it a potentially indispensable component of anemia evaluation in outpatient and inpatient settings.<sup>[6]</sup>

In recent years, the utility of the Mentzer Index has been extensively explored in pediatric populations, where thalassemia trait screening is often part of national health policies. However, adult populations are underrepresented in research concerning the diagnostic utility of red cell indices. Therefore, this study aims to bridge this knowledge gap by

evaluating the hematological profile, iron status and Mentzer Index of adult patients presenting with microcytic anemia in a tertiary care setting.

## MATERIALS AND METHODS

This was a retrospective, cross-sectional observational study conducted in the Department of Pathology, NRI Institute of Medical Sciences, Visakhapatnam. The duration of study was one year extending from October 2024 to September 2025. A total of 120 adult patients aged 18 years and above and presenting with microcytic anemia were included in this study on the basis of a predefined inclusion and exclusion criteria. Sample size estimation was performed based on prior prevalence rates of iron deficiency anemia and beta-thalassemia trait in similar settings, assuming a power of 80% and a confidence level of 95%. The calculated minimum sample size was 100 patients hence 120 cases were included to improve statistical validity and account for potential exclusions. Since it was purely observational study and all patient data was anonymised, Ethical approval was waived from the Institutes ethics committee.

The study included patients who had CBC reports showing anemia, defined per WHO criteria (Hb  $<13.0$  g/dL in males and  $<12.0$  g/dL in females). Patients only with microcytic anemia were included. For each patient, detailed hematological indices were analysed from the laboratory database. These included parameters such as hemoglobin concentration, mean corpuscular volume (MCV), red blood cell (RBC) count, mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), red cell distribution width (RDW) and hematocrit. The Mentzer Index was calculated by the formula: MCV (fL)/RBC count (million/ $\mu$ L). Based on the calculated Mentzer Index patients were classified into two groups: values  $<13$  suggestive of beta-thalassemia trait (BTT) and values  $>13$  suggestive of anemia due to iron deficiency (IDA).

Subsequently the confirmatory investigations were reviewed for these patients. These included serum ferritin, serum iron, total iron-binding capacity (TIBC), transferrin saturation, and hemoglobin electrophoresis. Cases with Mentzer Index suggestive of BTT were correlated with hemoglobin electrophoresis findings to identify the presence of elevated HbA2 ( $>3.5\%$ ) indicative of thalassemia trait. Conversely cases with indices suggestive of IDA, iron studies were used to confirm the diagnosis with iron deficiency being defined by low serum ferritin ( $<15$  ng/mL in females,  $<30$  ng/mL in males) or low transferrin saturation ( $<16\%$ ). Patients found to have mixed pictures or secondary causes such as chronic kidney disease, chronic infections or malignancy were excluded from the study to ensure diagnostic clarity.

Statistical analysis was performed using SPSS version 26.0 (IBM Corp., Armonk, NY). Descriptive statistics were used to present demographic and hematological data. Continuous variables were depicted as mean  $\pm$  standard deviation (SD). Categorical variables were expressed as frequency and percentages. Comparative analysis between the IDA and BTT groups was done by using Chi-square test for categorical variables. Unpaired t-test was used for continuous variables. A p-value less than 0.05 was considered statistically significant.

#### Inclusion Criteria

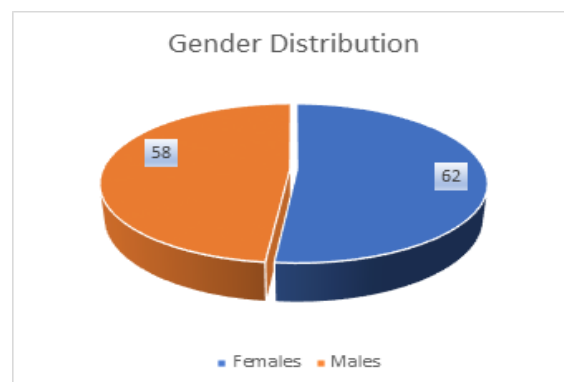
- Adult patients above 18 years of age.
- Hemoglobin levels  $<13.0$  g/dL in males and  $<12.0$  g/dL in females fulfilling WHO criteria for anemia.
- Microcytic hypochromic picture on peripheral smear.
- Availability of complete blood count (CBC) reports.
- Availability of confirmatory tests including either iron studies or hemoglobin electrophoresis.

#### Exclusion Criteria

- Patients below 18 years of age.
- Patients with chronic renal failure, malignancy, autoimmune hemolytic anemia, or aplastic anemia.
- Patients with a history of blood transfusion within the past 3 months.
- Incomplete laboratory records or missing confirmatory investigations (iron studies or Hb electrophoresis).
- Patients with evidence of dual pathology or anemia of chronic disease without iron or thalassemia-specific findings.

## RESULTS

The analysis of the gender distribution of the studied cases showed that females constituted a slightly higher proportion, with 62 cases (51.7%), compared to males who accounted for 58 cases (48.3%). There was slight preponderance of females with a M:F ratio of 0.93:1 [Figure 1].



**Figure 1: Gender Distribution of Studied cases.**

The analysis of the age group distribution of the studied cases showed that the most commonly affected age group in both females and males was 31–45 years. This was followed by the 18–30 year age group. The above 45 year group accounted for 16 cases (25.8%) in females and 16 cases (27.6%) in males. The mean age of both the groups was found to be comparable, with females having a mean age of  $34.8 \pm 10.2$  years and males  $35.6 \pm 9.8$  years, with no statistically significant difference ( $P=0.61$ ) [Table 1].

**Table 1: Gender wise distribution of studied cases.**

Age Group	Females (n = 62)	Males (n = 58)	Total (n = 120)	p-value
18–30 years	20 (32.3%)	20 (34.5%)	40 (33.3%)	0.61
31–45 years	26 (41.9%)	22 (37.9%)	48 (40.0%)	
>45 years	16 (25.8%)	16 (27.6%)	32 (26.7%)	
Total	62 (51.7%)	58 (48.3%)	120 (100.0%)	
Mean age $\pm$ SD	$34.8 \pm 10.2$	$35.6 \pm 9.8$	—	

The analysis of hematological parameters between patients diagnosed with  $\beta$ -thalassemia trait (BTT) and those with iron deficiency anemia (IDA) revealed several statistically significant differences. The mean red blood cell (RBC) count was notably higher in the BTT group ( $5.6 \pm 0.7 \times 10^6$  / $\mu$ L) compared to the IDA group ( $3.8 \pm 0.6 \times 10^6$  / $\mu$ L), with a highly significant p-value ( $<0.001$ ). The mean corpuscular volume (MCV) was significantly lower in BTT cases ( $64.2 \pm 4.9$  fL) than in IDA cases ( $72.8 \pm 7.5$  fL), with a p-

value of 0.004. Similarly, the mean corpuscular hemoglobin (MCH) was lower in BTT ( $18.9 \pm 1.8$  pg) versus IDA ( $22.3 \pm 2.3$  pg), and this difference was statistically significant ( $p=0.002$ ). Red cell distribution width (RDW) was lower in BTT ( $13.8 \pm 1.1$ %) compared to IDA ( $17.6 \pm 2.2$ %), with a p-value  $<0.001$ . Although the mean hemoglobin level was slightly higher in BTT ( $10.1 \pm 1.1$  g/dL) than in IDA ( $9.4 \pm 1.2$  g/dL), this difference was not statistically significant ( $p=0.071$ ) [Table 2].

**Table 2: Comparison of Hematological Parameters Between  $\beta$ -Thalassemia Trait and Iron Deficiency Anemia**

Parameter	BTT (n=9) Mean $\pm$ SD	IDA (n=111) Mean $\pm$ SD	p-value
Hemoglobin (g/dL)	$10.1 \pm 1.1$	$9.4 \pm 1.2$	0.071
RBC ( $10^6$ / $\mu$ L)	$5.6 \pm 0.7$	$3.8 \pm 0.6$	$<0.001^*$
MCV (fL)	$64.2 \pm 4.9$	$72.8 \pm 7.5$	$0.004^*$
MCH (pg)	$18.9 \pm 1.8$	$22.3 \pm 2.3$	$0.002^*$
RDW (%)	$13.8 \pm 1.1$	$17.6 \pm 2.2$	$<0.001^*$

Serum iron levels were considerably higher in the BTT group ( $82.4 \pm 13.7 \mu\text{g/dL}$ ) compared to the IDA group ( $38.5 \pm 11.3 \mu\text{g/dL}$ ), with a p-value  $<0.001$ . Similarly, serum ferritin was markedly elevated in BTT cases ( $72.1 \pm 14.9 \text{ ng/mL}$ ) versus IDA ( $11.2 \pm 4.7 \text{ ng/mL}$ ) ( $p<0.001$ ). Total iron-binding capacity

(TIBC) was significantly lower in BTT patients ( $295.4 \pm 48.2 \mu\text{g/dL}$ ) compared to those with IDA ( $410.1 \pm 52.0 \mu\text{g/dL}$ ), with a p-value  $<0.001$ . Transferrin saturation followed the same trend, being higher in BTT cases ( $28.6 \pm 6.5\%$ ) than in IDA cases ( $9.3 \pm 3.1\%$ ) ( $p<0.001$ ) [Table 3].

**Table 3: Comparison of Iron-Study Parameters Between  $\beta$ -Thalassemia Trait and Iron Deficiency Anemia**

Parameter	BTT (n=9) Mean $\pm$ SD	IDA (n=111) Mean $\pm$ SD	p-value
Serum Iron ( $\mu\text{g/dL}$ )	$82.4 \pm 13.7$	$38.5 \pm 11.3$	$<0.001^*$
Serum Ferritin ( $\text{ng/mL}$ )	$72.1 \pm 14.9$	$11.2 \pm 4.7$	$<0.001^*$
TIBC ( $\mu\text{g/dL}$ )	$295.4 \pm 48.2$	$410.1 \pm 52.0$	$<0.001^*$
Transferrin Saturation (%)	$28.6 \pm 6.5$	$9.3 \pm 3.1$	$<0.001^*$

The analysis of the diagnostic distribution based on the Mentzer Index classification showed that among the 120 studied cases, the majority were diagnosed with iron deficiency anemia (IDA), comprising 111 cases. Of these, 108 cases had a Mentzer Index  $>13$ , while only 3 cases had an index  $<13$ . Conversely,

beta-thalassemia trait (BTT) was identified in 9 cases, of which 8 had a Mentzer Index  $<13$  and just 1 case had an index  $>13$ . This indicates that a Mentzer Index  $<13$  was highly suggestive of BTT, while a value  $>13$  was strongly associated with IDA. [Table 4].

**Table 4: Mentzer Index in cases of BTT and IDA.**

Diagnosis	Mentzer Index $<13$	Mentzer Index $>13$	Total
BTT	8	1	9
IDA	3	108	111
Total	11	109	120

The analysis of the diagnostic performance of the Mentzer Index in differentiating beta-thalassemia trait (BTT) from iron deficiency anemia (IDA) showed a high overall diagnostic accuracy of 96.7%. The sensitivity of the index was 88.9%, indicating that it correctly identified the majority of true BTT cases. The specificity was even higher at 97.3%, reflecting its strong ability to correctly exclude non-

BTT (i.e., IDA) cases. The positive predictive value (PPV) was 72.7%, suggesting that nearly three-quarters of cases with a Mentzer Index  $<13$  truly had BTT. Meanwhile, the negative predictive value (NPV) was exceptionally high at 99.1%. These results confirm that the Mentzer Index is a highly reliable screening tool, especially effective in ruling out BTT when the index is  $>13$  [Table 5].

**Table 5: Diagnostic Performance of the Mentzer Index ( $<13$ ) for Identifying  $\beta$ -Thalassemia Trait.**

Diagnostic Metric	Value
Sensitivity	88.9%
Specificity	97.3%
Positive Predictive Value	72.7%
Negative Predictive Value	99.1%
Overall Diagnostic Accuracy	96.7%

## DISCUSSION

In the present cross sectional study of adult patients with microcytic anemia, we observed that the majority were female (51.7%) and clustered in the 31–45 year age group (40.0%). This demographic distribution aligns with the known higher prevalence of iron deficiency anemia (IDA) among women of reproductive and perimenopausal age. Our mean age (near 35 years) is slightly older than many pediatric centric studies of microcytic anemia—but of course our focus was adults. The small group identified with  $\beta$  thalassemia trait (BTT) (n=9) showed a higher mean RBC count, lower MCV and lower RDW compared to IDA cases. These findings mirror the classic hematologic pattern described by earlier authors. In BTT, microcytosis with relatively preserved RBC count and narrower anisocytosis (lower RDW) compared with IDA where RBC

production is reduced and RDW widened. For example, Tabassum S et al reported that the Mentzer index (MI) differentiated IDA and BTT with acceptable accuracy.<sup>[7]</sup> Similar utility of MI was also reported by the authors such as Zafar M et al and Bhattacharya S et al.<sup>[8,9]</sup>

When comparing specific hematologic indices, we found that RBC count in BTT ( $5.6 \times 10^6/\mu\text{L}$ ) was significantly higher than in IDA ( $3.8 \times 10^6/\mu\text{L}$ ) and the mean MCV in BTT was 64.2 fL compared to 72.8 fL in IDA ( $p=0.004$ ). The RDW in BTT (13.8%) versus IDA (17.6%) differed highly significantly ( $p<0.001$ ). These results are consistent with previous literature. For instance, A Sharma and colleagues found significantly lower MCV and RDW in thalassemia trait compared to IDA.<sup>[10]</sup> However, the mean hemoglobin difference (10.1 vs 9.4 g/dL) did not reach statistical significance ( $p=0.071$ ) in our study. Some earlier investigations reported a clearer



hemoglobin separation—though often in pediatric cohorts or in mixed etiology populations. The non-significant hemoglobin difference in our sample may reflect the relatively small BTT subgroup (n=9), or the fact that adult BTT carriers may present with mild anemia overlapping with IDA. Similar haematological profiles in IDA and BTT have also been reported by the Jameel T et al and J. Odhwani et al.<sup>[11,12]</sup>

Turning to iron study parameters, we observed that serum ferritin, serum iron, TIBC and transferrin saturation all showed statistically significant differentiation between BTT and IDA ( $p<0.001$ ). For example, mean ferritin in BTT was 72.1 ng/mL vs 11.2 ng/mL in IDA, and mean transferrin saturation was 28.6 % vs 9.3 %. These findings align with the expected pathophysiology of these 2 types of anemia. In BTT, iron stores are typically normal (unless there is co-existent iron deficiency) whereas in IDA iron stores are depleted. In prior studies, the combination of red cell indices plus iron studies has been emphasized. For instance, A Sundh et al reported that indices alone may be less reliable without iron status and hemoglobin- electrophoresis confirmation.<sup>[13]</sup> In our study the integration of iron studies adds internal validity to our diagnostic separation of IDA vs BTT. The diagnostic performance of the Mentzer Index in this adult study yielded an overall accuracy of 96.7 %, sensitivity 88.9 %, specificity 97.3 %, PPV 72.7 % and NPV 99.1 % for identifying BTT (using  $MI < 13$ ). These results appear quite strong and support the utility of MI as a screening tool in resource- limited settings. They compare favourably with other literature: for example, the retrospective study by Saxena S et al found MI sensitivity of ~89 %, specificity ~87.9 % across all ages.<sup>[14]</sup> On the other hand, studies in pediatric populations and other geographic regions have reported lower sensitivities or specificities: Al Qarni et al in Saudi children found MI sensitivity ~74 %, specificity ~63 % for BTT.<sup>[15]</sup> Given these variations, our adult- based data in an Indian tertiary care centre appear encouraging. Nonetheless, we must remain cautious: the BTT subgroup in our study is small (n=9) and so the precision of sensitivity/PPV estimates is limited. Also, the high NPV suggests MI is especially useful to rule out BTT when  $MI > 13$ —but the lower PPV (72.7 %) means that  $MI < 13$  should not be used as definitive diagnosis of BTT without confirmatory testing.

From a broader perspective, our findings have several implications. First, in adult patients presenting with microcytic anemia in low- resource or rural settings (such as many centres in India), calculation of MI from a standard CBC may provide a low- cost, rapid screening step to help triage which patients merit more advanced testing (e.g., HPLC for Hb A<sub>2</sub> ). This may reduce empirical iron supplementation in unsuspected BTT carriers, thereby mitigating potential risk of iron overload over time. Second, our data reinforce that red cell indices (RBC count, MCV, RDW) retain discriminative power in adults as

well as in paediatric cohorts. Third, however, the limitations must be acknowledged: the small BTT sample, single- centre design, retrospective observational nature, and the fact that patients with mixed pathologies (e.g., combined IDA+BTT) were excluded, limit generalizability. Future studies should include larger adult cohorts, multi centre populations, and also evaluate MI cut offs tailored to adults in India (and perhaps adjusted for co-existing iron deficiency or inflammatory states). Finally, while MI is useful, it should not substitute for confirmatory iron studies and hemoglobin electrophoresis; rather, it serves as a screening/triage tool.

## CONCLUSION

The Mentzer Index proved highly effective as a screening test, offering strong specificity and an excellent negative predictive value for excluding  $\beta$ -thalassemia trait in adults with microcytic anemia. Its simplicity, negligible cost, and ease of calculation make it particularly useful in resource-limited settings where advanced diagnostics are not readily accessible. Nonetheless, its moderate positive predictive value reinforces that it should guide preliminary triage rather than establish a definitive diagnosis. Confirmatory iron studies and hemoglobin electrophoresis remain essential, and larger adult cohorts are needed to further validate its performance.

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