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Red cell distribution width's role in differentiating iron deficiency anemia from other hypochromic microcytic anemias

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Shno Ibrahim Hussein[1]* Abbas Abdulkadir Rabaty[2]

Abstract

Background and objective: The red cell distribution width is suggested to be a more sensitive indicator for microcytic hypochromic anemia. Therefore, this study aimed to determine the role of red cell distribution width in the diagnosis of iron deficiency anemia from other causes of hypochromic microcytic anemia.

Methods: This cross-sectional study involved the children patients who attended Rapareen Teaching Hospital in Erbil city in 2019 and were diagnosed with hypochromic microcytic anemia.

Results: The red cell distribution width was determined in a group of 70 children with iron deficiency anemia and 30 cases with a non-iron deficiency (other hypochromic microcytic anemias). Patients with a higher socio-demographic status were more likely to have iron deficiency anemia than those with low socio-demographic status; 82.61% vs. 76.60%, respectively. The patients with symptoms were more likely to be diagnosed with iron deficiency anemia (P = 0.024). The mean red cell distribution width value was 14.38%, 15.73%, and18.02% among mild, moderate, and severely anemic children (P <0.001). Increasing red blood cells (r=-0.271), hemoglobin (r=-0.454), serum iron (r=-0.601), and serum ferritin (r=-0.560) lead to decrease red cell distribution width. The sensitivity, specificity, positive predictive value, negative predictive value, and accuracy of red cell distribution width in diagnosing iron deficiency anemia in children patients were 77.14%, 63.33%, 83.08%, 54.29%, and 73.0%, respectively.

Conclusions: This study showed that red cell distribution width has good sensitivity and specificity in diagnosing iron deficiency anemia.

Keywords: RDW; IDA; RBC indices; Microcytic anemia; Serum iron.

Introduction

Red blood cells (RBC) are responsible for hemoglobin levels. Deficits in their production, increased destruction, or loss through bleeding are three main mechanisms by which anemia occurs.[1] Anemia resulting from a lack of sufficient iron to synthesize hemoglobin is the most common hematological disease in infants and children. It has been estimated that 30% of the global population suffers from iron deficiency anemia (IDA).[2] The highest level of hemoglobin is in the first three days of life (18.5 gm/dl), which decreases gradually until six months of age when it becomes 11.5 gm/dl then starts to increase again until reaching 12.5 gm/dl between 2 and 6 years old age.[3] Anemia in children between 6-59 months old can be divided into mild (10-10.9 gm/dl), moderate (7-9.9 gm/dl), and severe (< 7 gm/dl).[3,4]

Iron deficiency is the most commonly seen micronutrient nutritional deficiency among children and has contributed significantly to an increase in morbidity and mortality. The most severe consequence of iron deficiency seen is IDA.[5] Iron has a vital role in many biologic functions, including energy production, respiration, and cell proliferation. IDA is the end-stage result of the lack of iron in the body resulting from inadequate iron intake, increased iron

[1] Rapareen Pediatric Hospital, Erbil, Iraq.
[2] Department of Pediatrics, College of Medicine, Hawler Medical University, Erbil, Iraq.
* Correspondence: shnoibrahim5@gmail.com

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loss, or excessive iron requirements.[6] As a consequence, erythropoiesis becomes insufficient to fulfill the body's physiologic needs.[1] Complete blood count (CBC) is used for primary evaluation of anemia. A CBC can help determine the mean corpuscular volume (MCV), which measures the average size of RBCs, and mean corpuscular hemoglobin concentration, which measures the concentration of hemoglobin (Hb) in a given amount of packed RBCs.[7]

Red cell distribution width (RDW) measures the variation in red blood cell volume or red blood size. Its normal range is 12.8 ±1.2%.[8] RDW is suggested to be a more sensitive indicator to establish the possible parameter for microcytic hypochromic anemia.[9] RDW can give the idea of early changes in RBC, which is accompanied IDA.[10,11] It is done as part of CBC. Its value is usually beneficial for several cases such as anemias, especially of microcytic hypochromic types, diabetes mellitus, heart diseases, liver diseases, and cancers.[12,13] High levels of RDW may indicate a deficiency of nutrients such as folate, vitamin B12, and iron. A normal level of RDW with low MCV may be found in chronic kidney diseases. A normal RDW with high MCV may indicate aplastic anemia.[14]

Various diagnostic approaches are important to rule out diseases with adequate accuracy. Bone marrow studies are invasive methods, and serum ferritin, serum transferrin, and serum iron are relatively expensive. RDW, alongside other red cell indices, are part of routine blood counts in laboratories using automated hematology analyzers. Some of these tests do not have high sensitivity and specificity; therefore, patients have to undergo further evaluation for the diagnosis of IDA.[15]

Several previous studies have reported the controversial role of RDW in the diagnosis of IDA.[16,17] These studies did not report the conclusive word on using RDW in the diagnosis of IDA. Therefore, this study aimed to determine the role of RDW in the diagnosis of IDA from other causes of hypochromic microcytic anemia.

Methods

In this cross-sectional study, patients with IDA and non-IDA who attended Rapareen Teaching Hospital in Erbil city were screened physically and clinically for the eligibility criteria. They were referred to the hospital laboratory for investigations.

The included patients were children aged between 1 and 5 years old who were diagnosed with microcytic anemia; IDA, and non-IDA. The following exclusion criteria were applied to the patients; patients who did not complete iron study; children who received iron therapy over the past month, or blood transfusion over the past three months; patients who had an acute infection during data collection; patients with IDA along with concomitant thalassemia trait or disease; and children diagnosed with a chronic disorder, lead poisoning or sideroblastic anemia.

The diagnoses of the anemia types were established for microcytic (MCV <75fl) anemia or hemoglobin <11g/dL; and IDA: serum iron <25µg/dL and ferritin < 12ng/ml.[18] Red cell distribution width measures the variation in red blood cell volume (Normal range: 12.8±1.2%).[8] An anonymous questionnaire was presented to the patients' patents or accompanied persons who provided the following information. The information was collected in two sections in a pre-designed questionnaire. The questionnaire was composed of three domains: demographic and socioeconomic data, nutritional habits and presenting complaints, and hematological parameters.

Part A- Demographic and socioeconomic data: Part A included general information, including age, weight, gender, residency (urban, rural), socioeconomic state, and clinical presentation.[19]

Part B- Nutritional habits and clinical presentation: This part included the feeding -related information of patients. The information included feeding in the first

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six months categorized as breastfeeding, bottle-feeding, mixed feeding, type of child's current and main feeding, and drinking tea. The current feeding of patients was recorded as ordinary family food, breastfeeding, commercial infant formula, and adult milk feeding. The following sign and symptoms were collected, pallor, loss of appetite, poor weight gain, irritability, and pica.

Part C - hematological parameters: It included the results of medical investigations, including information on RBC, HB, MCV, MCH, serum ferritin, serum iron, and IDA. The patients were determined as microcytic (MCV <75fl) anemia (hemoglobin <11g/dL), IDA (serum iron <25µg/dL and ferritin <12ng/ml),[18] and RDW (normal range are 12.8±1.2%)[8]. Venous blood samples (10 mL) were collected using a disposable syringe after 8 to 9 h of fasting; 4 mL of blood was placed in a lavender top tube containing EDTA. The automated hematology analyzer (mythic 22- orphee- Geneva/ Switzerland) was used for evaluating complete blood count. Serum ferritin and serum iron level were measured by an automated chemistry analyzer (Roche Cobas C 311- Hitachi - Tokyo/ Japan).

The descriptive purposes of the study were determined in number and percentage, including the prevalence of IDA and other hypochromic microcytic anemias, including thalassemia trait or disease, anemia of chronic disorder, lead poisoning, and sideroblastic anemia. The general information was presented in the form of mean and standard deviation (SD) or number and percentage. Independent t-test and one-way ANOVA tests were performed to compare socioeconomic status, feeding patterns, clinical manifestations, and hematological parameters between two groups. The bivariate correlation was performed to find out the correlation of RDW with hematological parameters. The sensitivity and specificity of RDW in the diagnosis of IDA in children were determined in percentage. The significant level was determined in a *P* value of less than 0.05. The statistical calculations were performed by the statistical package for the social sciences (SPSS, version 25; IBM Corp; USA).

The ethical approval of the present study was obtained from the ethical committee of the Kurdistan Board of Medical Specialties (KBMS). The confidentiality of the personal information of the patients was protected by the anonymity of participants. The written informed consent was obtained from the patient's parents before participating in the study.

Results

The mean age and weight of the patients were 2.33 (SD: 1.28 years) and 13.37 (SD: 3.93 Kg), respectively. The patients were males (56.0%) and females (44.0%). Most of them live in urban areas (72.0%). In the current study, out of 100 blood samples tested, 70 were diagnosed with IDA, and 30 samples were non-iron deficiency. In the socioeconomic status, there was a significant difference between both groups (*P* = 0.015).Thepercentageof IDA was 82.6% among those with high socioeconomic status (SES), 50% among those with moderate SES, and 76.6% among those with low SES, as shown in Table 1.

The highest percentage of IDA was among the breastfeeding group (79.31%), and the lowest percentage (62.5%) was among the mixed feeding group. In the bottle feeding group, the percentage was more than half (68.09%). Statistically, there were no significant differences (*P* = 0.383) in the first six months of feeding. The clinical symptoms of IDA as compared with other hypochromic microcytic anemia were pallor (100%, 83.33%), loss of appetite (78.57%, 46.67%), poor weight (72.86%, 20.00%), irritability (25.71%, 6.67%), and pica (35.71%, 3.33%), respectively, as revealed in Table 2.

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Table 1 Types of anemia according to the socioeconomic status and feeding behavior

Patient characteristics (100)	Categories	No of patients	IDA		Other hypochromic anemia		*P* value
			No.	(%)	No.	(%)	
Socioeconomic state	Low	47	36	(76.60)	11	(23.40)	**0.015**
	Moderate	30	15	(50.00)	15	(50.00)	
	High	23	19	(82.61)	4	(17.39)	
Feeding (First 6 months)	Breastfeeding	29	23	(79.31)	6	(20.69)	0.383
	Bottle feeding	47	32	(68.09)	15	(31.91)	
	Mixed Feeding	24	15	(62.50)	9	(37.50)	
Feeding (Current Feeding)	Ordinary Family food	64	44	(68.75)	20	(31.25)	0.633
	Breast Milk	6	4	(66.67)	2	(33.33)	
	Commercial Infant Formula	9	8	(88.89)	1	(11.11)	
	Adult Milk	21	14	(66.67)	7	(33.33)	
Feeding (Drinking Tea)	No	58	40	(68.97)	18	(31.03)	0.791
	Yes	42	30	(71.43)	12	(28.57)	

Pearson Chi-squared test was performed for statistical analyses.
The bold numbers show a significant association.

Table 2 Comparison of symptoms between patients with iron deficiency anemia and other hypochromic anemias

Symptoms	IDA (n=70)		Other hypochromic anemia (n=30)		*P* value
Pallor					**<0.001**
Yes	70	100	25	83.33	
No	0	0.00	5	16.67	
Loss of Appetite					**0.002**
Yes	55	78.57	14	46.67	
No	15	21.43	16	53.33	
Poor Weight					**<0.001**
Yes	51	72.86	6	20.00	
No	19	27.14	24	80.00	
Irritability					**0.029**
Yes	18	25.71	2	6.67	
No	52	74.29	28	93.33	
Pica					**0.001**
Yes	25	35.71	1	3.33	
No	45	64.29	29	96.67	

Pearson Chi-squared test was performed for statistical analyses.
The bold numbers show a significant association

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The means of RBC, HB, RDW, MCV, MCH, serum ferritin, and serum iron were 4.59 $10^{\times 12}$/L, 9.41 g/dl, 15.32%, 61.03 fl, 20.36 pg, 16.91 ng/ml, and 31.02 mg/dl, respectively. All of the patients included in this study were anemic and microcytic, as shown in Table 3.

In the RDW comparison between IDA and non-IDA groups, the mean RDW scores were significantly higher in the IDA group (16.40) than in the non-IDA group (12.82) in independent t-test (*P* <0.001). In addition, the RDW means were increasing with the severity of IDA (mild, moderate, and severe anemia were 14.38%, 15.73% and 18.02%, respectively).

The width of red cell distribution was decreased with increasing RBC count (r=-0.271; *P* = 0.013), Hb (r=-0.454; *P* <0.001), MCH (r=-0.291, p=0.008), serum iron (r=-0.601, *P* <0.001), and serum ferritin (r=-0.560, *P* <0.001), as presented in Table 4.

The sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and accuracy of RDW in the diagnosis of IDA in children patients were 77.14%, 63.33%, 83.08%, 54.29%, and 73.0%, respectively (Table 5)

Table 3 Hematological parameters of the studied groups

Parameter (n=100)	**Statistics Mean (SD)**			
	All patients	**IDA**	**Other microcytic anemia**	***P* value**
RBC count	4.59 (0.62)	4.46 (0.61)	4.83 (0.69)	0.015[a]
Hb	9.41 (1.25)	9.10 (1.33)	10.10 (0.65)	<0.001[a]
PLT count	335.56 (76.27)	340.13 (73.18)	324.90 (83.35)	0.390[a]
RDW (Normal values: 12.8±1.2%) (CV)	15.32 (2.44)	16.40 (1.91)	12.82 (1.53)	<0.001[a]
MCV	61.03 (5.42)	60.42 (5.85)	62.36 (4.09)	0.066[a]
MCH	20.36 (2.07)	19.84 (2.08)	21.48 (1.55)	<0.001[a]
Serum ferritin	16.91 (15.22)	7.67 (2.74)	39.21 (8.00)	<0.001[a]
Serum iron	31.02 (24.24)	16.29 (4.59)	65.40 (14.47)	<0.001[a]
RDW category (IDA group)				
Mild		14.38 (2.38)		<0.001[b]
Moderate		15.73 (2.16)		
Severe		18.02 (0.71)		

[a] An independent t-test and [b]ANOVA one-way were performed for statistical analyses.

Table 4 Correlation of RDW with hematological parameters

		Correlations					
		RBC	**HB**	**MCV**	**MCH**	**S.iron**	**S.ferritin**
RDW	The correlation coefficient (r)	-0.271	-0.454	-0.136	-0.291	-0.601	-0.560
	P-Value	**0.013**	**<0.001**	0.221	**0.008**	**<0.001**	**<0.001**

Table 5 Sensitivity and specificity of RDW in the diagnosis of IDA in children

	TP	**TN**	**FP**	**FN**	**Sensitivity**	**Specificity**	**PPV**	**NPV**	**Accuracy**
RDW	54	19	11	16	77.14%	63.33%	83.08%	54.29%	73.0%

TP: True positive; **TN**: True negative; **FP**: False positive; and **FN**: False negative
PPV: Positive predictive value; **NPV**: negative predictive value

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Discussion

Values of RDW are one of the RBC indices that reflect the degree of anisocytosis of red blood cells. It has been used to differentiate between patients with IDA and thalassemia trait for decades.[20] In the present study, 70% were diagnosed with IDA, and 30% of samples with non-iron deficiency. This agreed with a study conducted in India,[21] which showed that out of 100 cases, 79% were found to have IDA, and 21% were non-iron deficient anemia. The results above showed a significant difference in a low socioeconomic state between IDA and other hypochromic microcytic anemia (76.60% and 23.40%), respectively. This is comparable to the previous studies,[20,22] which revealed a protective effect of high IDA prevalence in a representative pediatric population in Korea and India. The onset of IDA can be affected by several factors, which include age, sex, physiological, pathological, and nutritional state. However, illiteracy and low standard of living are the main causes of anemia among children. The clinical features of IDA and hypochromic microcytic anemia are similar to those of other types of anemia, although there are specific symptoms attributed to iron deficiency. These agreed with Llanos et al. and Wang, who reported that iron imbalance in IDA is affected by several mechanisms, which included reduced absorption, increased losses, and/or increased requirements. Often such factors can coexist, requiring complex management of the consequent anemia.[23,24] Also, Fareeq documented a positive effect of IDA on physical growth in children, especially during the first two years of life during which growth is fast.[25]

The means of RDW in IDA and other microcytic anemia were 16.40 and 12.82, respectively. These agreed with two other studies that showed overall average RDW value of 18.31 ±1.75 and 17.3±2.5 in IDA, respectively.[16,26] It was stated that RDW is useful in the early detection of IDA. Also, our result has good sensitivity, specificity, and predictive values in the diagnosis of IDA in children. The width of red cell distribution was decreased with increasing RBC, Hb, MCH, serum iron, and serum ferritin. This agreed with Jassim, who documented that RBC count, Hb, Hct, MCH, and MCHC were significantly lower, whereas RDW was significantly higher in IDA subjects.[27] Similarly, the RDW means were increased with the severity of IDA. This result was concordant with Choudhary et al. in that there was a progressive increase in RDW with the severity of anemia, and there was also a progressive decrease in MCV with an increase in the severity of anemia.[5] In addition, another study reported the inverse association between RDW and the hemoglobin value in the IDA group. However, this kind of association was not found in the control group.[17] Similar findings were reported in other studies.[9,20,28-31] It has been seen that anisocytosis occurs, where the erythrocytes produced are of smaller than average size and have a large size variation due to inadequate iron supply. The morphology and function of erythrocytes at a molecular level have been known to be disturbed due to IDA. Therefore, an increase in RDW values may occur in IDA, allowing early detection of ID before the reduction in MCV occurs.[28]

The evaluation of microcytic anemias and/or microcytosis is an expensive process. To differentiate these diseases from each other, a set of investigations is required. Complete blood counts, serum iron, serum ferritin, TIBC, Hb electrophoresis are performed for the definitive diagnosis.[5,15,29] The RBC indices are cost-effective parameters to detect anemias.[30] However, in patients with microcytic anemias, RBC shows similar results, such as low hemoglobin and MCV. Red cell distribution width is a better index than RBC to differentiate IDA from other causes of microcytosis; since it is abnormal in the development of IDA.[31] Similar findings were reported elsewhere.[20,32]

Conclusion

RDW can be used as a tool for the diagnosis of IDA in a large number of samples at major hospitals to reduce time consumption because of the high prevalence of IDA and expensive costs of specialized tests and adopting cost-effective, accurate and efficient measures is required. Red cell distribution width has good sensitivity and specificity for IDA screening diagnosis amongst patients with the hypochromic microcytic anemia.

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Competing interests

None declared.

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