Original Research Paper

Pathology

UTILITY OF RETICULOCYTE HEMOGLOBIN CONTENT IN THE DIAGNOSIS OF ANEMIA

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ABSTRACT

Background: Anemia is a major public health concern across the world and Iron deficiency is the most frequent cause of anemia especially in developing countries like India. Evaluation of serum ferritin is very important to know the iron status of patients and diagnosis of Iron deficiency anemia(IDA). But as serum ferritin is an acute phase reactant, it may increase in inflammatory conditions also. Automated cell counters form an integral part of modern-day hematology. Automated reticulocyte counts not only provide accuracy and enhanced precision but they also accurately quantify the content of mRNA and cellular indices including cell volume, Hemoglobin concentration and Hemoglobin content. In this study, the potential use of newer reticulocyte parameter – Reticulocyte hemoglobin content(Ret-Hb) in the diagnosis of anemia has been assessed. Material and Methods: This is a retrospective study which included 185 EDTA blood samples of adult anemia patients at Dr. Pinnamaneni Siddhartha Institute of Medical Sciences & RF, Chinna avutapally from January 2022 to December 2022.Results: Among the 185 adult anemia patients studied, there was female preponderance. Most of the cases were microcytic hypochromic anemia followed by normocytic normochromic anemia. Ret-Hb showed good sensitivity of 90.45% in detecting anemia in patients with low MCV. Ret-Hb levels were low even in patients with low Hb and normal MCV. Thus, Ret-Hb can detect iron deficiency anemia earlier than traditional markers like MCV. Conclusion: Ret-Hb, like MCV provides results in a very short period, does not incur additional costs, can be measured simultaneously in automatic blood count devices and it will detect iron deficiency and iron deficiency anemia earlier even in patients with normal MCV. The present study showed that we can use Ret-Hb in detecting iron deficiency/ iron deficiency anemia.

KEYWORDS : Anemia, Iron deficiency anemia, Hemoglobin, MCV, Reticulocyte hemoglobin content.

INTRODUCTION

Anemia is a significant worldwide health problem. Approximately one-third of the world's population suffers from anemia, half of which is caused by iron deficiency (ID)[1,2]. Traditional indices of body iron such as serum ferritin(SF), serum iron, total iron binding capacity and transferrin saturation(TSAT) are affected by other factors (inflammatory state, diurnal variation, diet/malnutrition, decreased liver function) and iron metabolism. New reticulocyte parameters are provided by automated hematology analysers for the assessment of iron status. Reticulocyte hemoglobin content(Ret-Hb) is a newer reticulocyte parameter that provides information at cellular level about iron availability for erythropoiesis in individual reticulocytes. Reticulocyte hemoglobin content(Ret-Hb) is not affected by any factors other than those participating in iron metabolism, as long as there is no abnormality in Hb synthesis[3]. Reticulocytes are immature red blood cells released into the blood two to three days following hematopoiesis and become mature red blood cells one to two days later. In the bone marrow, iron is taken up from the blood during hematopoiesis and then binds to heme protein to form hemoglobin (Hb).The hemoglobin content of reticulocytes(Ret-Hb) is considered to reflect Hb synthesis potential and the iron levels used in hematopoiesis[4]. Since the normal lifespan of these cells is between one to two days in peripheral blood, Ret-Hb is a good indicator of the availability of iron and can point to early iron deficient erythropoiesis. In contrast to iron deficiency anemia caused by inadequate iron stores, the anemia of chronic disease is associated with decreased iron availability despite abundant stores[5,6,7].The hemoglobin content of reticulocytes (Ret-Hb) may assist in differentiating between these forms of anemia[8,9] and shows a lower degree of within patient variability than ferritin or other serum indicators of iron status[10]. The cut-off level of 28pg for Ret-Hb was suggested after a prospective study[11].Ret-Hb is an effective and widely available Hb synthesis status indicator that permits ID/IDA diagnosis and monitoring in all age groups with or without underlying diseases, including beta-thalassemia.

MATERIALS AND METHODS

This is a retrospective study which was performed on 185 adult anemia patients who came to our hospital between January 2022 to December 2022. Adult patients(age between16yrs to 70yrs) who were diagnosed with anemia as per the WHO reference range - Pregnant women (Hb <11g/dl), Non-pregnant women (15 years and above - Hb <12g/dl) and Men (15 years and above-Hb<13g/dl) were included in the study. Blood samples were run in the automated hematology analyzer. Hemoglobin, red cell indices (MCV) and Ret-Hb values obtained were analyzed.

STATISTICAL ANALYSIS & RESULTS

Data was analyzed using statistical package for social sciences (SPSS) version 20. Descriptive statistical methods were used to calculate the percentages, sensitivity, specificity and PPV of Ret-Hb.

Table 1: showing severity of anemia and corresponding MCV and Ret-Hb values:

Hb(g/dl)	MCV low (<80fl)	MCV Normal (80-100fl)	Ret-Hb low (<28pg)	Ret-Hb Normal (>28pg)
10-10.9	0(0%)	3(1.6%)	2(1.1%)	1(0.6%)
7-10	26(14%)	2(1.05%)	25(13.5%)	3(1.7%)
<7	152(82.2%)	2(1.05%)	140(75.6%)	14(7.5%)
Total	178(96.2%)	7(3.7%)	167(90.2%)	18(9.8%)

Table 2: Showing analysis of Ret-Hb and MCV:

Ret-Hb	MCV Low (<80fl)	MCV Normal (80-100fl)	Total
LOW(<28pg)	161(87%)	6 (3.2%)	167 (90.2%)
NORMAL(>28pg)	17 (9.2%)	1 (0.54%)	18 (9.7%)
TOTAL	178 (96.2%)	7 (3.78%)	185 (100%)

Table 3: Showing Anlaysis of Ret-Hb among patients with different MCV values:

Ret-Hb	MCV low 50-60fl	MCV low 60-70fl	MCV low 70-80fl	MCV Normal 80-100fl	Total
LOW (<28pg)	63(37.7%)	58 (34.7%)	40 (23.9%)	6(3.2%)	167 (90.2%)
NORMAL (>28pg)	5 (27.7%)	5 (27.7%)	7 (38.8%)	1(0.54%)	18 (9.7%)
TOTAL	68(36.7%)	63(34%)	47 (25.4%)	7(3.78%)	185 (100%)

DISCUSSION:

Of the total 185 anemic patients analyzed, 156 were females and 29 were males. Age range was between 16years to 70years. Hemoglobin range was between 4.1-11.5g/dl with the mean hemoglobin of 8.5± 1.76g/dl. Among the total 185 anemic patients, 178(96.2%) patients had low MCV <80fl and 7 (3.78%) patients had normal MCV 80-100fl. This is more when compared to study by Nova stoffman et al[12] . In that study, a total of 381 patients were included, anemia was diagnosed in 63 patients (16.5%) and low MCV was found in 170(44.6%) patients. The more percentage in the present study could be due to more common nutritional and iron deficiency in developing countries like India. In this study, 167 patients had low Ret-Hb and 18 patients had normal Ret-Hb. Out of 178 anemic patients with low MCV,161(90.44%) patients had low Ret-Hb with a sensitivity of 90.45%, with a PPV of 96.41%. This is comparable to study by Alan E et al[13] where the overall sensitivity is 60.7%. In the present study, 6(85.7%) patients showed low Ret-Hb and 1 (0.54%) patient showed normal Ret-Hb out of 7 patients with low Hb and normal MCV indicating that Ret-Hb can detect anemia earlier than MCV. In our study, 178 patients were with low MCV. Among the 47 patients with MCV between 70-80fl, 63 patients with MCV between 60-70fl and 68 patients with MCV between 50-60fl, low Ret-Hb was shown by 40(23.9%)patients, 58(34.7%) patients and 63(37.7%) patients respectively. Even in patients with low hemoglobin and normal MCV, Ret-Hb was fallen. The fall in Ret-Hb may precede the development of microcytes, showing Ret-Hb could detect iron deficiency anemia earlier than traditional markers like MCV, and in the present study Ret-Hb showed good sensitivity of 90.45% in detecting anemia in patients with low MCV.

CONCLUSION

Measurement of Ret-Hb is rapid, convenient, cost-effective and can be measured simultaneously in automatic blood count devices. It will detect iron deficiency and iron deficiency anemia earlier even in patients with normal MCV. It has an excellent diagnostic sensitivity and specificity. Thus Ret-Hb is a potential marker in the diagnosis of iron deficiency anemia.

LIMITATIONS

It was a single-center, retrospective study with a relatively small sample size.

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