The Value of Serum Ferritin in the Prediction of Iron Deficiency in Patients with Lymphoid Neoplasms

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Abstract

Background:- anemia is a well-known complication of patients with lymphoid neoplasm and many factors are involved in its pathogenesis including defective iron utilization and diagnosing iron deficiency in the context of malignancy or chronic illness can be very challenging as many of the parameters used are acute phase markers and can be misinterpreted in the presence of chronic illness .bone marrow iron is the gold standard mean for assessing marrow iron but it is an invasive and a laborious procedure ,on the other hand serum ferritin has been used as the most relevant indicator of iron deficiency in the general population but it has the drawback of being increased in the presence of an acute response.

Objectives:- evaluate the ability of serum ferritin at different cutoff levels and in combination with other parameters to identify iron deficiency in patients with lymphoid neoplasms.

Patients and methods:- 39 anemic patients with different types of lymphoid neoplasm attending the hematology unit in Baghdad hospital were enrolled in the study ,exclusion criteria included history of blood transfusion and iron therapy in the past 2 months .for all patients bone marrow iron study was carried out and accordingly patients were classified into 2 groups the iron depleted group (with absent marrow iron) and the iron replete group (with present iron stores).from all patients peripheral blood was taken for the evaluation of complete blood picture and ESR and serum ferritin by enzyme Linked immunosorbant assay method (ELISA).

Results:- the mean serum ferritin for the iron depleted group was 78.31ng/dl while for the iron replete group it was 202.77ng/dl there was a highly significant difference for serum ferittin between the 2 groups (p value .005),a less significant difference was found also for the mean cell volume(p value .05) while the other parameters showed no difference, the lowest cutoff level for serum ferittin that could accurately identify iron deficiency with a high sensitivity 96% and a high specifity 80% was 66.7ng/dl . Logistic regression analysis was done for serum ferritin and ESR to test for the effect of these parameters on the predictive power of bone marrow iron stores. The logistic equation was:

Log (p)=0.163 + .0621*ESR -0.026*ferritin

Using this equation with ferritin and ESR gave a prediction power of about 74% to identify the iron bone marrow store however using ferittin alone gave a 69% prediction power.

Conclusion: - serum ferritin at a cutoff value of 66.7ng/dl can be used with high accuracy for the identification of iron deficiency in patients with lymphoid neoplasm and can be used as a suitable alternative for bone marrow iron, correction of serum ferritin for the acute phase response did not enhance much its predictive power in identifying iron deficiency in those patients.

Key words: - ferritin, ESR, lymphoid neoplasm, marrow iron stores, anemia.

الخلاصة

الهدف :- يهدف البحث الى تقييم امكانية استخدام تحليل مستوى الفيرتين في الدم لوحده او بالشتراك مع عوامل اخرى لتقييم حالة فقر الدم الناجم عن نقص مستوى الحديد لدى المرضى المصابين باورام الجهاز اللمفاويز

طريقة الدراسة:- تم اجراء البحث في الفترة بين كانون الاول/2011 و تشرين الاول/2012 في قسم امراض الدم في المختبرات التعليمية/مدينة الطب-بغداد و شملت الدراسة 39مريض مصابين باورام الجهاز اللمفاوي بمختلف انواعه حيث تم سحب نخاع العظم للمرضى المشمولين و صبغ العينات الزجاجية بصبغات خاصة لكشف الحديد و في نفس الوقت تم اجراء تحليل مستوى الفيرتين في الدم للمرضى باستخدام تقنية مقايسة المناعة المرتبطة بالانزيم (ELISA) بالاضافة الى تحليل عداد الدم و تحليل قابلية ترسب كريات الدم الحمر.

النتائج و مناقشتها :- اظهرت الدراسة ان بالامكان استخدام فحص مستوى الفيرتين لتمييز فقر الدم الناجم عن نقص الحديد لدى المرضى المصابين باورام الجهاز اللمفاوي و لكن بحد ادنى اعلى من المتعارف عليه لدى الاشخاص الاصحاء و ان استخدام الفحص بالاشتراك مع الفحوص الاخرى لم يضف شيئا الى قابلية الفحص لوحده في تمييز هذه الحالات و ان استخدام تحليل مستوى الفيرتين في الدم حسب الحد الدنى المذكور يمكنه ان يميز و بدقة عالية حالات فقر الدم الناجمة عن نقص الحديد لدى المرضى المصابين باورام الجهاز اللمفاوي و يمكنه ان يغني عن فحص نخاع العظم لتحديد هذه الحالات .

Introduction

Iron deficiency anemia and anemia of chronic disorders are both considered to be the most important causes of anemia worldwide[1,2] .anemia of chronic diseases commonly develops in subjects with acute and chronic immune activation such as patients with infection, malignancy, or autoimmune diseases with functional iron deficiency being a major contributing factor [3] .the detection of iron deficiency in the presence of chronic illness has long been a diagnostic challenge taking into account the frequency of the problem and the impact it carries on the patient management.

Anemia is a common feature in lymphoid neoplasm even before patients have received cytotoxic therapy and even when there is no bone marrow involvement. The pathogenesis of this anemia is multifactorial [4]. And anemia has been used in the Binet classification [5] and in the Durie and Salmon classification [6].

Most authors agree that apart from determination of iron stores in Prussian blue stained bone marrow smears no single test on peripheral blood can differentiate accurately iron deficient patients in the presence of an acute phase response .although iron staining of bone marrow aspirates is the most sensitive but the procedure remains an invasive one and difficult to perform[7] .For long serum ferritin was considered as the most reliable indicator of bone marrow iron stores in non-complicated anemic patients[8] , however being an acute phase reactant protein it loses the predictive power in the presence of chronic inflammation ,malignancy or chronic infection[9]. Therefore many attempts have been made to improve the predictive ability of ferittin in the presence of chronic illness .some authors rely on combining ferritin with MCV, RDW, and other RBC indices [10] Others rely on combining ferittin with other markers of inflammation such as ESR, CRP, fibrinogen [11] while some prefer to establish new cut off values for ferritin in selected group of patients [12]

In the present study we try to evaluate the ability of serum ferritin in predicting bone marrow iron state in patients with lymphoid neoplasm by using different cut off level, or in combination with other RBC indices and with acute phase reaction marker, we tried to use the most simple and available markers attempting to obtain a simple mean that can be useful in places were advanced tests are not always available.

Aim of the study: is to evaluate the usefulness of serum ferritin for the

prediction of bone marrow iron status at different cutoff values and after correction for acute phase reaction in patients with lymphoid neoplasm.

Patients and methods

44 patients attending the hematology ward in Baghdad teaching hospital for the period between December 2011 till September 2012 were included in the study. Patients were diagnosed to have lymphoid neoplasm by tissue biopsy or by bone marrow biopsy, 6 patients were diagnosed with Hodgkin's lymphoma, 4 patients with chronic lymphocytic lymphoma, 3 patients with acute lymphoblasic leukemia and the rest were diagnosed as non-Hodgkin lymphoma. Bone marrow aspirate was done either at diagnosis or as a part of the staging system. All the patients were anemic with hemoglobin level below 13 g/dl for males and 12g/dl for females according to the WHO criteria [13]. Patients who gave history of blood transfusion or of iron treatment in the last 3 months were excluded from the study.

From each patient peripheral blood was taken in 2 tubes ,EDTA blood for estimation of hematological parameters(using ABBOT automated hematology analyzer)and ESR(determined by the Westergren method) ,and a plane tube for Serum which was frozen at -70 for future evaluation of serum ferritin.

Bone marrow aspirate was done for each patient and smears were examined for fragments and those with more than 7 fragments were chosen for iron staining by Prussian blue stain [14].

Bone marrow iron stained smears were examined independently by 3 hematopathologists unaware of the results of ferritin or the hematological parameters of the patient. On the basis of the bone marrow examination, the patients were divided into 2 subgroups, those with iron (iron replete group) and those without iron (iron depleted group) as shown in figure 1.

Ferritin was estimated by Enzyme linked immunosorbent assay method using (BioCheck ferritin enzyme immunoassay Kit) and according to the manufacture instructions.

Statistical analysis:

Statistical analysis was performed using. Logistic regression, student t-test and ROC curve (receiver operating characteristic). Calculations were performed using SPSS version 12. P values less than 0.05 were considered statistically significant.

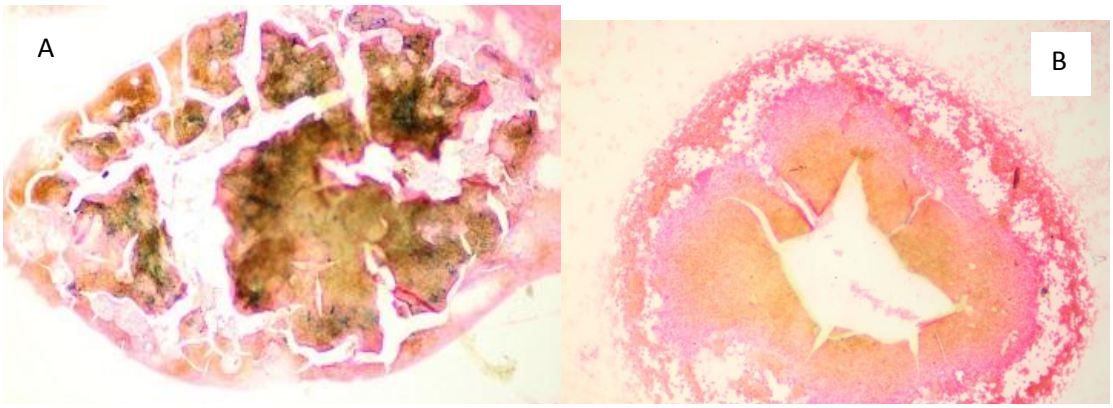

Figure 1: (A) Iron depleted bone marrow fragment (B) a marrow fragment positively stained for iron (×10, Prussian blue stain).

Results

Fourty four patients were initially enrolled in the study. Five patients were later on excluded because they did not meet the requirement of 7 bone marrow fragments and therefore could not be accurately assessed for the presence or absence of

bone marrow iron. Ultimately 39 patients were included in the study 21 were males, 18 were females, the mean age was 39.60 years.

Those 39 patients were then classified into 2 groups based on bone marrow examination, the iron depleted group (n=10) and the iron replete group (n=29). The mean hemoglobin level for the iron replete group was 9.4 g/dl while for the iron deplete group it was 8.8g/dl. The mean ferritin level for the iron depleted group was 78.33ng/dl while for the iron replete group it was 202.77ng/dl .When the 2 groups were compared using the T test a highly significant difference was found for Ferritin while a less significant result was seen for the MCV, the other parameters (Hb, PCV, MCH,MCHC and the ESR) showed no significant difference (table 1). The distribution of serum ferritin for the study groups is shown in figure 1.

Table 1. distribution of laboratory parameters of iron depleted and iron replete patients compared by T test.

parameter	Iron-deficient patients, n = 10	Iron-replet patients, n = 29	P value
Hb	8.8± 1.4	9.4± 1.9	0.367
PCV	28.7± 5.3	31.8± 5.9	0.150
MCV	76.73±6.59	81.51±6.60	0.050*
MCH	26.34±1.85	27.47±2.28	0.165
MCHC	32.74±0.97	33.61±1.31	0.061
Ferritin	78.31±96.13	202.77±120.96	0.005*
ESR	49.1±29.97	59.72±30.03	0.34

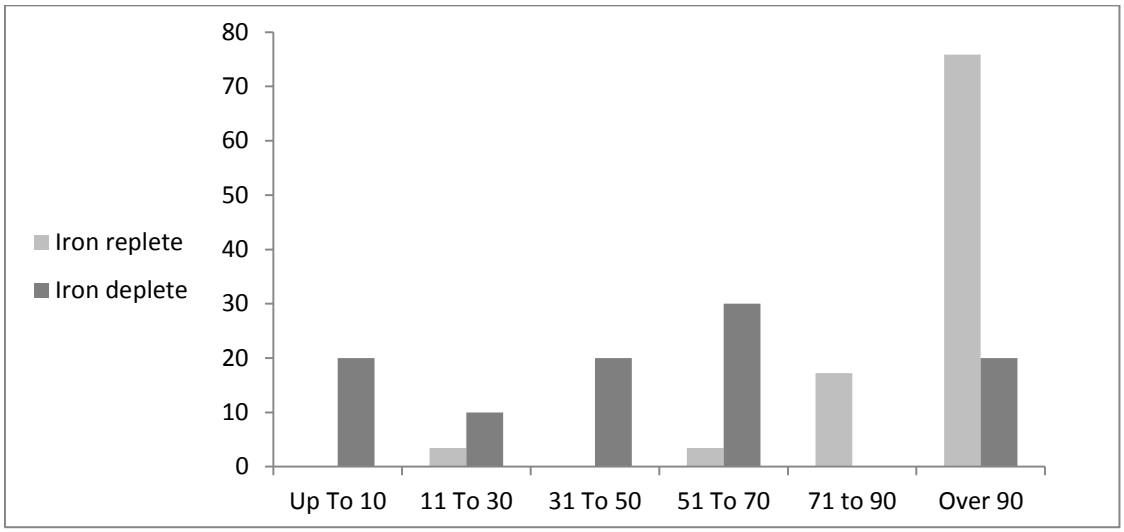

Figure 1: The distribution of serum ferritin in the iron depleted and iron replete group .

In an attempt to find a cut off value for serum ferritin suitable for patients with lymphoid neoplasm at which most of the iron depleted patients can be identified with a high sensitivity and specifity Receiver Operating Characteristic curves (ROC curves) for different cut off points of ferritin was done. ROC curves have shown that at a cutoff point of 66.7ng/dl there was a 96% sensitivity and 80% specifity, the area under the curve was 0.858. The positive predictive power (PPV) was 93% and the negative predictive power was 88 %. (fig 2)

Since serum ferritin level is elevated in many inflammatory conditions along with other acute phase proteins. In order to use ferritin as a predictor of bone marrow iron store, it must be corrected for other components of acute phase reactant.

Logistic regression analysis was done for serum ferritin and ESR to test for the effect of these parameters on the predictive power of bone marrow iron stores. The logistic equation was:

Log(p)=0.163 + .0621*ESR -0.026* ferritin

Using this equation with ferritin and ESR gave a prediction power of about 74% to identify the iron bone marrow store however using ferittin alone gave a 69% prediction power so correction for the ESR added only a modest elevation to ferritin predictive power.

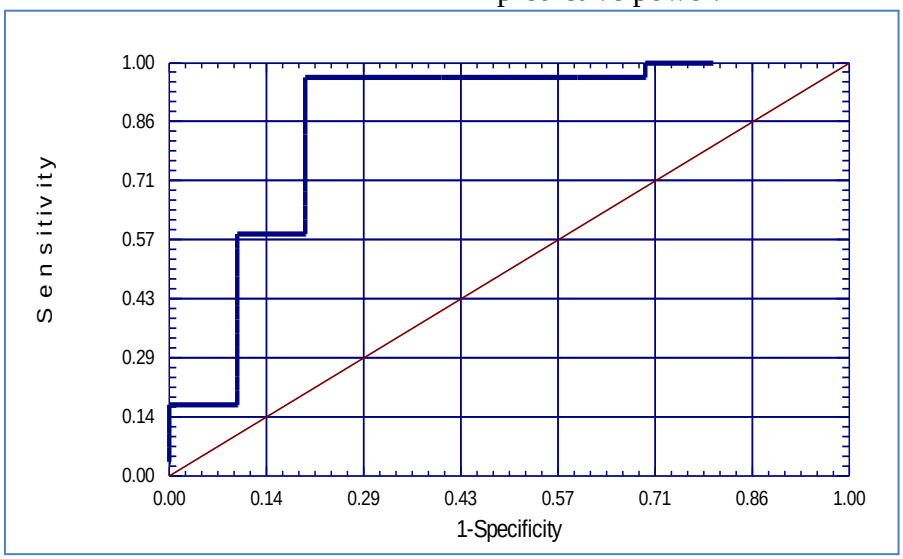

Figure 2: ROC curve of serum ferritin at different cut off levels in the identification of bone marrow iron status.

Discussion

Anemia is a serious and a well known complication for patients with lymphoid neoplasm and there are several factors that can contribute to its development including abnormal iron utilization, low erythropoietin level, hemolysis and bone marrow involvement [15]. Many studies have emphasized the importance of anemia as a prognostic factor in patients with lymphoma and lymphoroliferative disorders [4]. Identification of the type of anemia can definitely influence the management and prognosis of such patients, identifying iron deficiency anemia in those patients without relying on the bone marrow iron study is challenging since many of the iron markers are influenced by the acute phase body response.

in this study we attempt to analyze the ability of serum ferritin alone and in combination with other parameters to discriminate iron deficiency in such patients, the results has shown that ferritin was significantly different between the iron depleted and iron replete groups and it also showed that ferritin at a cutoff level of 66,7ng/dl can identify patients with depleted stores with a high sensitivity and a well accepted specifity in fact ferritin at this cutoff level showed a 93% positive predictive power and 88% negative predictive power, in the general population ferritin less than 12ng/ml is used for diagnosis of iron deficiency which is too low for patients with malignant conditions .many studies has identified a higher cutoff value for ferritin when identifying iron deficiency in the setting of chronic infections or inflammation like the study of M.Kortu etal who used cutoff point of 30ng/dl to identify iron deficiency in patients with tuberculosis [16] , and the study by S. Kurer etal [17] who used a cutoff level 40ng/dl for patients with rheumatic diseases. In another study by KH Ong etal [18] the cutoff value was 60ng/dl for patients in acute care hospitals. Another study by J.Coenen [11] has used a

cut off level of less than 70ng/dl to identify iron deficiency in patients with chronic inflammatory conditions.

In our study combining ferritin with ESR in an attempt to correct the acute phase response did not improve much the predictive power of ferritin as it only modestly raised it from 69% to 74% which remains far below the ability of ferritin at a cutoff value of 66.7ng/dl in identifying iron deficient patients although these results disagree with other studies like the study of David L. Witte [10] and the study of A. Oyekunle [19] who both showed that ferritin correction using ESR or C reactive protein or fibrinogen can further enhance its discrimination power but still this matter is debatable as other authors have shown that correction for those acute phase proteins either did not add anything [11] or gave a slightly better accuracy [15] and using a higher cutoff level seems to be a more helpful alternative.

In conclusion our study has shown that in patients with lymphoid neoplasm a cutoff value for serum ferritin of 66.7 can predict with very high accuracy patients with iron depletion without the necessity to perform a bone marrow iron study and that correcting serum ferritin for other acute phase reactions namely the ESR did not improve much its predictive capacity still another study including a higher number of patients and measuring other iron parameters and other acute phase proteins can further endorse the use of ferritin in combination with other parameters to identify iron deficiency in patients with lymphoid neoplasm.

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