

The Significance of Serum Transferrin Receptor Levels in the Diagnosis of the Coexistence of Anemia of Chronic Disease and Iron Deficiency Anemia

Kronik Hastalık Anemisi ve Demir Eksikliği Anemisi Birlikteliğinin Tanısında Serum Transferrin Reseptör Düzeyinin Önemi

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Abstract

Objective: Iron deficiency anemia is the most common cause of microcytic anemia throughout the world. Ferritin levels are good indicators of iron stores; however, levels may increase irrespective of iron stores in cases of chronic disease. Therefore, it is difficult to diagnose iron deficiency anemia coexisting with anemia of chronic disease.

Materials and Methods: To determine the level of transferrin receptor in subjects, 30 patients with iron deficiency anemia, 30 patients with anemia of chronic disease and 30 patients with both diseases were included in the study.

Results: Mean serum transferrin receptor levels were 5.99 ± 2.98 mg/L in the iron deficiency anemia group, 1.90 ± 1.15 mg/L in the anemia of chronic disease group and 3.07 ± 0.90 mg/L in the combination group. Comparing groups with each other revealed significant differences ($p < 0.05$).

Conclusion: It is concluded that the assessment of serum transferrin receptor levels is a useful method for the diagnosis of iron deficiency anemia in patients.

Key Words: Anemia of chronic disease, Iron deficiency anemia, Serum transferrin receptor level

Özet

Amaç: Demir eksikliği anemisi tüm dünyada en sık görülen mikrositer anemi sebebidir. Ferritin düzeyi demir depolarının iyi bir göstergesi olmasına rağmen kronik hastalık durumlarında demir depolarından bağımsız olarak yükselebilmektedir. Bu yüzden kronik hastalık anemisiyle birlikte olan demir eksikliği anemisinin tanısını koymak zordur.

Gereç ve Yöntem: Transferrin reseptör düzeyinin yararını değerlendirmek amacıyla demir eksikliği anemisi, kronik hastalık anemisi ve kombine olmak üzere her gruptan 30'ar hasta çalışmaya alındı.

Bulgular: Ortalama serum transferrin reseptör düzeyleri demir eksikliği anemisinde 5.99 ± 2.98 mg/L, kronik hastalık anemisinde 1.90 ± 1.15 mg/L ve kombine olan grupta ise 3.07 ± 0.90 mg/L olarak bulundu. Gruplar birbirleri ile karşılaştırıldığında aralarında anlamlı farklılık saptandı ($p < 0.05$).

Sonuç: Hastalarda demir eksikliği anemisini tespit etmede, serum transferrin reseptör düzeyinin değerlendirilmesinin oldukça faydalı bir yöntem olduğu sonucuna varılmıştır.

Anahtar Kelimeler: Demir eksikliği anemisi, Kronik hastalık anemisi, Serum transferrin reseptör düzeyi

Introduction

Anemia of chronic disease (ACD) is the second most common cause of microcytic anemia after the iron deficiency anemia (IDA). The level of ferritin is a good indicator of iron stores; however, it usually increased at the anemia of chronic disease.

Therefore, it is difficult to diagnose IDA coexisting with ACD. If these anemias are overlapped and can not be diagnosed by routine examination, bone marrow aspiration smear stained with Prussian blue could be used to show iron storage [1].

ACD is a normochromic-normocytic or hypochromic microcytic anemia. Hemoglobin (Hb) is usually around 9-11

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g/dl. If the Hb is below 8 g/dl, other factors deeping the anemia should be investigated [2, 3]. Ferritin is the indicator of IDA. If serum ferritin level is below 30 ng/ ml, this favors the presence of IDA; but if serum ferritin level is above 200 ng/ ml, IDA have been ruled out [4]. The etiology of ACD includes long term infections, malignancies, autoimmune diseases, solid organ transplantations, chronic renal insufficiency and inflammation.

Recently, serum transferrin receptor (STfr) level has been considered as an important examination to reflect iron deficiency. STfr is a transmembrane protein, consisting of two polypeptide chains bound by disulfide bonds. It weighs 95 kD. The STfr level reflects the transferrin receptor level on the cell surface; and increase of this molecule shows intracellular iron deficiency. However, STfr level isn't affected at the anemia of chronic disease [5].

The golden standard to diagnose the coexistence of IDA and ACD is the assessment of iron stores on the bone marrow aspiration. Since it is invasive and expensive method, it is difficult to be used routinely. Recent studies have shown that IDA and ACD can be differentiated with STfr level [6]. This study aims to assess the benefits of STfr level in differential diagnosis of iron deficiency anemia, anemia of chronic disease and coexistence of these anemias.

Materials and Methods

This study included 90 patients whose hemoglobin (Hb) values were 12.5 g/dl or lower and Mean Corpuscular Volume (MCV) values were a maximum of 95 fl. Patients were divided into the following three groups containing 30 subjects each:

Group 1. Iron deficiency anemia (IDA): The diagnosis for iron deficiency anemia was made by evaluating Hb, hematocrit (hct), MCV, Red cell Distribution Width (RDW), serum iron level (sFe), serum iron binding capacity (sTBC), ferritin level and peripheral smear (PY). Patients whose ferritin levels were <30 ng/ml and MCV values were <80 fl and who had symptoms of hypochromic microcytic anemia were included in this group.

Group 2. Anemia of chronic disease (ACD): Patients who had anemia and were diagnosed with chronic infection, collagen tissue disease, or fever of unknown origin for at least one month. In addition, patients with ferritin levels >200 ng/ ml and high sedimentation and CRP values were included in this group.

Group 3. Coexistence of iron deficiency anemia and anemia of chronic disease (IDA+ACD): Patients who had been diagnosed with anemia of chronic disease were selected. Bone marrow aspiration was performed on patients with ferritin levels of 30-200 ng/ml, MCV values <80 fl, and high sedimentation and CRP values. Patients with negative iron stores

in bone marrow were included in this group. Patients who met the following criteria were excluded: received oral or parenteral iron treatments and blood transfusions in the last 3 months; suffered from diseases such as renal, hepatic, endocrinological, tumoral and myeloproliferative disease; and had megaloblastic anemia caused by folic acid or B12 deficiency.

After overnight fasting, 2 ml venous blood was collected from the enrolled patients for hemogram analysis. Hb, Hct, MCV, and RDW levels were examined. Four ml venous blood was collected in plastic tubes for serum ferritin, sFe, sTBC, STfr levels, biochemistry, and CRP, and 2 ml venous blood was collected in citrate tubes for sedimentation.

Complete blood counts were evaluated by using a Coulter LH 750 (Beckman Coulter, Miami, FL, USA) automated blood counter. The serum iron level and serum iron binding capacity were studied with a calorimetric method using an Olympus autoanalyzer. The serum ferritin was assessed with the chemiluminescence method using an Architect autoanalyzer.

CRP was determined using the nephelometric method, while sedimentation was measured with the Westergren method. Peripheral smears were assessed with Giemsa stain and serum transferrin receptors were assessed using the nephelometric method using a Dade Behring kit.

Normal values were designated as the following: Hb 12.5-15.3 g/dl, Hct 42-50%, MCV 80-95 fl, RDW 11.5-14.5, serum iron 25-156 µg/dl, serum iron binding capacity 25-425 µg/dl, ferritin 30-200 ng/ml, CRP 5 mg/L, sedimentation 0-12 mm/h, and serum transferrin receptor level 0.83-1.76 mg/L. Bone marrow aspiration was performed only on patients in group 3 with chronic disease and ferritin values of 30-200 ng/ml to determine whether they had iron deficiencies or not. Prussian blue was used for iron staining. One-way ANOVA was used in the statistical studies. For statistical values, the post hoc LSD test was used to evaluate the significance status between groups. For all statistical analyses, $p < 0.05$ was considered statistically significant. Pearson correlation analysis was used for correlation analysis of numerical values.

Results

There were no statistically significant differences between age and gender in all three groups ($p > 0.05$) (Table 1). When the three groups were compared with each other, no statistically significant differences were determined for Hb, hematocrit, MCV and RDW values ($p > 0.05$). Serum ferritin and transferrin receptor values were significantly different ($p < 0.05$) (Table 2).

Iron values were significantly different ($p < 0.05$) when comparing Group 1 and Group 2. However, there were no significant differences when comparing Group 1 and Group 3, and Group 2 and Group 3 ($p > 0.05$). When Group 1 was com-

pared with Group 2 and Group 3, the total iron binding capacity levels were found to be significantly different ($p < 0.001$). However, no significant differences were noted when Group 2 and Group 3 were compared with each other ($p > 0.05$).

Age and serum transferrin receptor levels were compared and no correlation was found ($p > 0.05$). Hemoglobin levels and serum transferrin receptor levels were compared between groups. In Group 1, a statistically significant correlation was found between Hb and serum transferrin receptor ($p < 0.05$; $r = -0.519$), although there was no such correlation in Group 2 and Group 3 ($p > 0.05$).

When MCV and serum transferrin receptor levels were compared for all patients, a negative correlation was found ($p < 0.01$; $r = -0.313$). Similarly, when serum transferrin receptor levels and ferritin were compared, a negative correlation was also observed ($p < 0.001$; $r = -0.445$). Comparing serum iron and serum transferrin receptor, there was no significant correlation ($p > 0.05$), and when RDW values were compared with serum transferrin receptor levels, a positive correlation was observed ($p < 0.01$; $r = 0.335$).

Discussion

When evaluating iron deficiency, assessing iron stores in the bone marrow is the gold standard. However, as different stains are used for bone marrow iron storage, this method is considered to be subjective. Measurement of sTfR levels is good indicator for the differentiation of these two anemias [7]; the limit value of sTfR levels is 5 mg/L when differentiating iron deficiency from ACD [8]. Inhibition of bacteria and tumor cell growth by keeping iron in the reticuloendothelial system in patients with ACD is important; thus, iron treatment contains risks [9, 10].

Table 1. The age and gender properties of the groups

	Group 1 (IDA)	Group 2 (ACD)	Group 3 (IDA+ACD)	P value
Number of patients (n)	30	30	30	
Male/Female	14/16	11/19	13/17	$p > 0.05$
Age-mean (range)	45±18.35 (20-84)	43.4±16.96 (22-81)	45.3±16.69 (20-75)	$p > 0.05$

Anemia becomes worse and symptoms increase when ACD coexists with IDA. The basic mechanism in ACD involves the directing of iron from circulation to the reticuloendothelial system and restricting erythropoiesis. Inflammatory processes affect the intake of iron, and production of hepcidin, a modulator in iron deficiency, prevents the release of iron from macrophages [11]. Thus, serum hepcidin levels can also assist in determining whether iron deficiency is present in patients with ACD [9].

A study conducted by Allen et al. showed no statistical significance when comparing sTfR level with age, gender and race ($p > 0.05$) [12]. In our study, age and gender factors were not statistically significant, which is in agreement with the literature. In defining the correct diagnosis for IDA, one study affirmed that measuring sTfR and ferritin levels together was more valuable than measurement of sTfR levels alone [13]. Similar studies reported that the accuracy of sTfR and sTfR/SF (serum ferritin) rates and the Log (sTfR/SF) to reflect iron deficiency were as high as 83% and 99%, respectively [14]. sTfR/SF rates are also used when IDA coexists with ACD [15].

Serum sTfR levels and the TfR-F index in IDA and anemia of chronic disease (IDA + ACD) were reported to be remarkably higher compared to a healthy control group and an iron-treated group with rheumatoid arthritis (RA). These tests were reported as a non-invasive, cost-effective method to indicate iron deficiency in anemia patients with RA when ferritin levels are above 60 µg/L [16]. There are several reports revealing that sTfR levels were significantly highest in the IDA group [17, 18], and that the IDA and ACD coexistence group had higher levels compared to the ACD group alone [17].

As a result of using serum transferrin receptor (sTfR) and sTfR/ferritin indices to differentiate IDA, ACD and combined anemia (IDA + ACD) in children, the sTfR level was found to be a very useful indicator [19].

During our study, a negative correlation was identified between the serum transferrin receptor and Hb in Group 1 (IDA) ($p < 0.05$; $r = -0.519$), although in Group 3 (IDA+ACD) and group 2 (ACD), no correlation was identified ($p > 0.05$). The relationship between anemia of chronic disease and iron deficiency anemia is in agreement with the literature. Although no significant correlation was identified in Group 3 (IDA + ACD), other studies have reported a negative correlation, which may be related to the number of patients evaluated.

Table 2. Comparing of serum ferritin and transferrin receptor values (mean)

	Group 1	Group 2	Group 3	Group 1-2	Group 1-3	Group 2-3
Ferritin ng/ml	11.53±7.77 (1.7-28)	516±239.09 (192-1272)	93.3±52.78 (29.8-200)	$p < 0.001$	$p < 0.05$	$p < 0.001$
STfr* mg/L	5.99±2.98 (1.82-13.5)	1.90±1.15 (0.77-5.48)	3.07±0.90 (1.73-6.15)	$p < 0.001$	$p < 0.001$	$p < 0.05$

*Serum transferrin receptor level

A study of 114 cases compared serum transferrin receptor level and serum iron levels, and identified a negative correlation ($p < 0.001$; $r = -0.502$) [10]. However, we didn't found any significant correlation between those factors ($p > 0.05$). Serum iron is not a reliable parameter, because it depends on variables such as satiety and diurnal rhythm. Therefore, it might be considered insignificant in our study.

This study has shown that the sTfR level was not affected by age or gender and is a useful parameter to identify combined anemia (IDA+ACD). The sTfR level does not increase in anemia of chronic disease and can be alternatively used to demonstrate iron presence in bone marrow in order to identify iron deficiency in ACD cases. When high sTfR levels are observed in cases of anemia of chronic disease and iron deficiency anemia, administering iron treatment may be an appropriate approach.

Conflict of interest statement: The authors declare that they have no conflict of interest to the publication of this article.

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