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Association of serum transferrin receptor concentration with markers of inflammation in Zimbabwean children☆

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Abstract

Background: Clinical studies have shown that degree of erythropoiesis, the hypoxic response, and iron status each independently influences transferrin receptor concentration, but there is conflicting information regarding the effect of inflammation on transferrin receptor expression.
Subjects and methods: Levels of hemoglobin, reticulocytes, serum ferritin, transferrin receptors and inflammatory markers (C-reactive protein, interleukin-6 and neutrophils) were determined in 208 Zimbabwean children ≤5 years attending well-child clinics in a non-malaria transmission, non-hookworm area.
Results: In linear regression models among 147 children with ferritin >10 ng/mL that adjusted for erythropoiesis with $\log_{10}$ reticulocyte count, the hypoxic response with hemoglobin concentration and iron status with $\log_{10}$ ferritin concentration, positive correlations were found between $\log_{10}$ transferrin receptor concentration and $\log_{10}$ C-reactive protein concentration ($P=0.012$), $\log_{10}$ interleukin-6 concentration ($P=0.011$) and $\log_{10}$ neutrophil count ($P=0.013$). These models predict that, with a baseline transferrin receptor concentration in the upper normal range of 8.0 mg/L and holding hemoglobin concentration and reticulocyte count constant, an increase from 1 to 10 mg/L in C-reactive protein is associated with a rise of 1.6 mg/L in transferrin receptor (95% C.I. 0.3–3.0 mg/L), an increase from 0.5-to-5.0 pg/mL in interleukin-6 with a rise of 1.9 mg/L (0.4–3.7 mg/L), and an increase from 2000 to 20,000/μL in neutrophil count with a rise of 3.6 mg/L (0.7–7.5 mg/L).
Conclusion: Our results suggest that inflammation leads to an increase in circulating transferrin receptor concentration that is independent of the degree of erythropoiesis, the hypoxic response and iron status.

Keywords: Transferrin receptors; Inflammation; Iron status; Children

1. Introduction

Transferrin receptors are expressed on the surface of all cells of the human body, except mature red blood cells, for the purpose of acquiring iron from plasma transferrin [1]. At the cellular level, the expression of transferrin receptor protein is coordinately regulated with the expression of ferritin protein through the interaction of iron-responsive proteins with an iron regulatory element on the 5′-untranslated region of ferritin mRNA and iron regulatory elements on the 3′-untranslated region of transferrin receptor mRNA [2]. Transferrin receptor expression is increased and ferritin expression is decreased with

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low cytosolic iron concentration, while the opposite occurs with elevated cytosolic iron level [2]. In addition to the regulation of translation of transferrin receptor by cellular iron status, the transcription of transferrin receptor is regulated by oxygen status [3]. Hypoxia leads to stabilization of hypoxia-inducible factor-1α, the principal transcriptional regulator of genes that respond to hypoxia including the transferrin receptor gene [4]. Hypoxia-inducible factor-1α then translocates to the nucleus where it binds to a hypoxia-inducible factor-responsive element on the promoter region of the transferrin receptor gene [5].

Soluble transferrin receptor present in plasma is a truncated fragment of the membrane transferrin receptor and the concentration reflects tissue expression [6]. At least three factors are important in regulating transferrin receptor expression at the level of the organism. First, the degree of erythropoiesis influences the total amount of transferrin receptors expressed in the body and the concentration of transferrin receptors in the plasma, with increased erythropoiesis being associated with increased plasma transferrin receptor concentration [7]. Second, body iron status regulates transferrin receptor expression in the organism as a whole: iron deficiency is associated with increased plasma transferrin receptor levels [8,9] and elevated iron stores are associated with reduced plasma transferrin receptor concentration [10]. Third, a general upregulation of the hypoxic response in the human body leads to increased plasma transferrin receptor levels after adjustment for degree of erythropoiesis and iron status [11].

There is conflicting information in the literature regarding the effect of inflammation on transferrin receptor expression. At the cellular level, treatment of murine macrophage cell lines with interferon-λ and lipopolysaccharide reportedly leads to lower transferrin receptor mRNA levels [12,13]. At the systemic level, some studies show no difference between normal subjects and patients with acute or chronic inflammation [8,9,14,15]. However, other studies have reported increases in transferrin receptor concentration during inflammation, possibly due to inflammation-associated iron-limited erythropoiesis in the absence of systemic iron deficiency [16–20]. Studies in African children with malaria infection have consistently shown increased serum transferrin receptor concentrations [21,22], but this finding can plausibly be explained by enhanced erythropoiesis associated with this hemolytic disorder [21,23]. In the present study, we examined the relationship between markers of inflammation and serum transferrin receptor concentration among African children in an area where malaria transmission and hookworm infestations are low to absent ([24], Nathoo PK, unpublished observations).

Table 1
Laboratory data of 208 Zimbabwean children

Age, months	24±15
Female sex, number (%)	114 (54)
Wbc, $\times 10^3/\mu L$	8.9±3.8 [a]
Neutrophils, $\times 10^3/\mu L$	2.02 (1.28–2.93) [a]
Lymphocytes, $\times 10^3/\mu L$	5.68 (4.48–7.13) [a]
Monocytes, $\times 10^3/\mu L$	0.19 (0.09–0.37) [a]
Rbc, $\times 10^6/\mu L$	4.35±0.50 [a]
Hemoglobin, g/dL	10.8±1.2 [a]
Mean corpuscular volume, fL	74±7 [a]
Reticulocyte count/μL	30320 (20313–42098) [b]
C-reactive protein, mg/L	0.7 (0.2–2.6)
Interleukin-6, pg/mL	1.12 (0.68–2.26) [c]
Serum ferritin, μg/L	18 (10–31)
Serum transferrin receptor, mg/L	6.8 (5.5–8.9)
Transferrin receptor/$\log_{10}$ ferritin index	5.3 (4.0–8.6)

Values are median and interquartile range except for age, white blood cell count, red blood cell count, hemoglobin concentration and mean corpuscular volume (mean±SD).
[a] $n=196$.
[b] $n=191$.
[c] $n=175$.

Table 2
Spearman correlations of serum transferrin receptor with markers of iron status, erythropoiesis, hypoxia and inflammation

	n	*R*	*P*
A. All participants			
Serum ferritin	208	−0.384	<0.001
Absolute reticulocyte count	191	0.263	<0.001
Hemoglobin	198	−0.340	<0.001
Mean corpuscular volume	198	−0.481	<0.001
C-reactive protein	208	0.174	0.012
Interleukin-6	175	0.202	0.008
Absolute neutrophil count	196	0.137	0.057
B. Participants with serum ferritin>10 ng/ml			
Serum ferritin	147	0.033	0.690
Absolute reticulocyte count	134	0.229	0.008
Hemoglobin	140	−0.145	0.087
Mean corpuscular volume	140	−0.319	<0.001
C-reactive protein	147	0.223	0.007
Interleukin-6	123	0.231	0.011
Absolute neutrophil count	139	0.183	0.031

2. Subjects and methods

2.1. Study subjects

Two hundred and eight apparently healthy children 3–60 months of age attending well-child clinics in Harare, Zimbabwe were studied. The Institutional Review Boards of the Medical Research Council of Zimbabwe and Howard University approved the study and written permission from the mothers or guardians was obtained. Five milliliters of peripheral blood were drawn from each child during the morning into two vacutainer tubes, one containing K_3-EDTA and one with no anticoagulant.

2.2. Analysis of blood samples

Complete blood counts were performed on an automated analyzer (Sysmex, Norderstedt, Germany) and leukocyte differential counts were done manually. Reticulocyte counts were determined by microscopic examination of supravital stained peripheral blood smears and malaria parasitemia by microscopic examination of thin blood smears stained by Giemsa. Serum concentrations of ferritin and transferrin receptors were measured with enzyme immunoassay kits (Ramco Laboratories, Stafford, TX) and the transferrin receptor/$\log_{10}$ ferritin index was calculated. The reference range for serum transferrin