Anemia in Elderly Patients: An Emerging Problem for the 21st Century

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Anemia is a significant problem in elderly patients. Although many anemic elderly patients can be diagnosed with nutritional deficiency, anemia of chronic inflammation or comorbid diseases that explain their decreased hematocrit, the etiology of anemia in a significant fraction remains obscure. There is evidence to suggest that the hematopoietic stem cell displays increasing erythropoietin (EPO) resistance with age. EPO levels rise in elderly, nonanemic patients, and it is hypothesized that there is an interplay between this rising demand for EPO and the decreasing ability of the aging kidney to produce adequate hormone to meet that need. There is further considerable evidence that aging is associated with increased proinflammatory cytokine expression and that many of these cytokines can contribute to EPO insensitivity. Consequently, genetic variation in the expression of these proinflammatory cytokines may influence the development of anemia in elderly patients, both through induction of hepcidin expression (anemia of inflammation) and through cytokine suppression of erythroid colony formation. The impact of inflammatory mediators, EPO insensitivity, and other factors that may act on the hematopoietic stem cell to decrease erythropoiesis are under active study and should serve to elucidate the pathophysiology of this important cause of morbidity and mortality in elderly individuals. A better understanding of the pathophysiology of anemia in elderly patients should provide critical entry points for interventions that will improve survival and quality of life in the aging population.

Anemia of any degree is recognized as a significant independent contributor to morbidity, mortality, and frailty in elderly patients.[1,2] Although anemia has often been considered a normal consequence of aging, the pathophysiology of such an age-related decline in erythrocyte production is obscure, and efforts to understand anemia in elderly individuals have become a major target of research interest. Recent studies suggest that, although anemia likely arises in part from the cumulative effect of age-related comorbidities and physical decline, there are still age-specific changes in the hematopoietic system that influence red blood cell production. Understanding of these changes could have important diagnostic and therapeutic implications for addressing this common problem. Because the number of elderly individuals is expected to reach unprecedented levels in the 21st century, anemia represents an emerging global health problem negatively impacting quality of life in a significant proportion of the elderly population and requiring an ever-greater allocation of health care resources.

Epidemiology of Anemia in Elderly Patients

In the Third National Health and Nutrition Examination Survey (NHANES III) study, the incidence of anemia in men and women older than age 65 was 11% and 10%, respectively.[3] The prevalence of anemia rose rapidly after the age of 50, approaching a rate greater than 20% in those individuals aged 85 years or older. It is estimated that more than 3 million Americans aged 65 years and older are anemic. Of the anemic patients, one-third were identified to have nutritional deficiency, one-third were diagnosed on the basis of iron studies to have anemia of inflammation, and one-third were diagnosed with "unexplained" anemia.[3] A wide ethnic disparity was also noted, with non-Hispanic blacks having a rate of anemia that was three times that of non-Hispanic whites; a finding that is consistent with the results seen in other studies.[4,5] The prevalence of anemia and the relationship between anemia subtypes and mortality were examined in the Women's Health and Aging Study I, a cohort of 688 severely disabled, community-dwelling women aged 65 years or older. This analysis similarly revealed that one-third of anemia remained unexplained, and found that the anemia of inflammation and anemia of chronic renal disease was associated with significantly increased mortality, with unexplained anemia associated with a trend toward higher mortality that did not reach statistical significance.[6] More recent follow-up analysis of anemia and mortality in NHANES III reveals that race and ethnicity appear to be important determinants of hemoglobin concentration, and various anemia subtypes appear to be associated with differential risks of mortality.[7] In the wake of these studies, several important questions remain to be answered:

1. Why is the rate of anemia so much higher in non-Hispanic blacks? The well-documented differences in blood counts seen in African Americans have caused some researchers to suggest that the stated normal range for hemoglobin, hematocrit, and mean corpuscular volume (MCV) in the African-American population should be modified. Beutler and West[4] examined these norms in the Kaiser Permanente records and presented data on 1491 African-American individuals, compared with more than 31,000 white subjects. They report that these parameters were all lower in blacks than in age-matched whites, whereas the serum ferritin was higher. After correcting for the incidence of α-thalassemia and iron deficiency, the differences were smaller, but still statistically significant. Of note, the difference in men was quite small, with a mean close to 14.5 g/dL in both African Americans and white patients. It seems unlikely to explain all of the differences in anemia incidence among black patients, since, in the Beutler study, after correcting for α-thalassemia and iron deficiency, the percentages of anemia in

African-Americans was still strikingly higher than in whites (11.26 in blacks vs 3.83 for whites). Dr. Beutler initially recommended against adjusting the norms for determining anemia, given the broad implications and complex variables that go into these parameters, but rather recommended greater consciousness of these considerations in practicing clinical judgment.[4] In a subsequent study, however, he evaluates the World Health Organization (WHO) classification of anemia and recommends that the lower limit of normal for black men above and below the age of 60 be placed at 12.9 and 12.9, respectively (vs 13 as defined in the WHO standards), and that the equivalent for white men be placed at 13.2 and 13.7, respectively.[8] To extend these observations, it is important to determine whether the morbidity and mortality associated with mild anemia using either set of norms is the same in the black population as in the white population. Furthermore, if one can determine the cause of unexplained anemia, it may provide insight into the particular genetic variables that influence the increased incidence of anemia in the non-white population.

2. What is the role of chronic inflammation in the etiology of unexplained anemia? There is strong evidence that many markers of inflammation, including tumor necrosis factor-α (TNFα) and interleukin (IL)-6, are increased in the elderly population, regardless of health status.[9,10] However, studies detailing evidence for a chronic proinflammatory state contributing to the pathogenesis of unexplained anemia in the elderly population have been few in number and limited by small sample sizes, lack of correlation with hepcidin and EPO levels, and reliance on measurements of peripheral blood cytokine levels as determinants of the presence of a chronic inflammatory state. Genetic variations determining overall inflammatory responsiveness may, in fact, be better markers of disease susceptibility than peripheral cytokine levels,[11] particularly when examining the impact of proinflammatory states on the bone marrow, a specialized compartment in which local exposure to inflammatory mediators is likely distinct from what is detectable by plasma assays. Therefore, larger studies correlating genotypic variation in proinflammatory genes with peripheral blood cytokine levels and with established biomarkers of inflammation will be required to determine whether chronic inflammation plays a role in the pathogenesis of unexplained anemia in the elderly population.

3. How much of the anemia of aging can be explained by comorbidity? The high frequency of comorbid conditions in elderly patients confounds the establishment of the etiology of an independent tendency for reduction in hematocrit with age. Although there appears to be a component of age-related anemia, even in healthy individuals, the incidence is much higher in patients with comorbid disease.[12] We postulate that the tendency toward a proinflammatory state in the elderly population predisposes them to accentuated debility in the face of comorbid disease. We further propose that genetic factors determining the level of proinflammatory markers contribute to the likelihood of developing features of frailty in response to other stressors.

Age-Related Expression of Inflammatory Markers, Anemia, and Frailty

Aging and the development of age-related comorbidities has been associated with chronically increased levels of proinflammatory cytokines, such as TNFα, IL-6, IL-1ß, macrophage migration inhibitory factor (MIF), and acute phase proteins.[9,10,13] It is unclear whether this chronic inflammatory state reflects primary age-related immune dysregulation or a systemic response to the presence of comorbid conditions. Some studies have suggested that, in normal healthy elderly individuals, the circulating levels of inflammatory cytokines are not elevated, whereas others have suggested that these markers are elevated even in the absence of comorbid disease. In the large InCHIANTI (Invecchiare in Chianti, aging in the Chianti area) study of more than 1300 individuals, levels of IL-6, IL-1 receptor antagonist, IL-18, C-reactive protein, and fibrinogen were all elevated in patients over age 65, although when adjusted for cardiovascular risk factors and morbidity, the elevation was small.[10] Other studies, particularly from the infectious diseases literature, have noted the limitations of the blood compartment in assessing periodic, but pathogenically relevant, increases in proinflammatory cytokines.[14] Taken together, there exists some debate as to whether assaying cytokine levels in the peripheral blood is an accurate method for estimating chronic inflammatory states. This is likely to be especially true in examining the impact of proinflammatory states on the bone marrow, which is a specialized compartment that may have local exposure to inflammatory mediators that is distinct from what is detectable by plasma assays. Furthermore, cytokine levels may be variable and subject to transient perturbations in the setting of acute stresses and comorbidities. Therefore, studies linking cytokine promoter polymorphisms to the development of various inflammatory diseases suggest that genetic variations that determine overall inflammatory responsiveness may be better markers of disease susceptibility than peripheral cytokine levels. Consequently, the assessment of functional polymorphisms in candidate genes offers a worthwhile investigative approach to assessing clinical outcomes.

Chronically increased levels of proinflammatory cytokines—such as IL-6, TNFα, IL-1ß, and MIF—have been associated with the development of a number of age-related comorbidities. MIF is a cytokine with broad immune-activating properties that is secreted by macrophages and T cells, and increased in a host of inflammatory disorders. It acts on macrophages to induce release of many proinflammatory mediators, such as IL-6, and serves as the upstream regulator of TNFα. Furthermore, both MIF and TNFα have been shown to impair erythroid colony formation, and MIF has been implicated in the pathogenesis of anemia associated with malaria.[15] The TNFα, IL-6, MIF, and IL-1ß genes have functional polymorphisms that affect the level of cytokine expression.[11,16] TNFα polymorphisms have been linked to susceptibility to severe malarial anemia[17] and lepromatous leprosy,[18] and may predict the response to anti-TNF treatment in rheumatoid arthritis.[19] High-activity MIF alleles have been associated with inflammatory arthritis,[20] inflammatory bowel disease,[21] and sarcoidosis.[22] IL-6 polymorphisms appear to influence the phenotypic expression of a wide variety of both benign and malignant disorders.[23]

Anemia and inflammation are strongly associated with, and may contribute to, the development of "frailty," a poorly defined syndrome of the elderly population associated with weight loss, impaired mobility, generalized weakness, and poor balance.[24] Some studies have suggested that elevated proinflammatory markers are associated with development of frailty.[24] Furthermore, anemia is associated with an increase in nearly all markers of frailty in elderly populations, suggesting that there may be a link between the pathogenesis of the two syndromes.[25] For example, IL-6 elevation has been associated with frailty,[25] and IL-6 is also the main inducer of hepcidin secretion, thus providing a potential common pathway for the development of anemia in that setting. Therefore, genetic variation in the level of cytokine expression may have an impact on the frequency with which older individuals develop certain subtypes of anemia and other manifestations of frailty.

Anemia of Inflammation

Anemia of inflammation (AI) has historically been termed the "anemia of chronic disease" and is most commonly seen in association with infection, rheumatologic disorders, malignancy, and other chronic illnesses. On a biochemical level, it is classically characterized by low serum iron and low iron binding capacity in the setting of an elevated serum ferritin. Although the etiology of classical AI has been attributed to decreased red cell survival, disordered iron-limited erythropoiesis, and progressive EPO resistance of erythroid progenitors, the relative role and interplay of these three mechanisms in the development of anemia remain unknown, as are the potential common pathways that may link them.

Our understanding of AI has been transformed by the discovery of the antimicrobial peptide hepcidin.[26] Hepcidin is a peptide synthesized by the liver that is a key regulator of iron metabolism. It has been demonstrated to inhibit intestinal absorption of iron and to block release of iron from macrophages. Overexpression of hepcidin in transgenic mice results in perinatal mortality from iron deficiency unless the mice are salvaged with intravenous iron,[27] whereas mice in which the hepcidin gene is ablated develop severe iron overload[28] Patients with AI, as diagnosed by elevated ferritin and low iron and iron binding capacity, have been demonstrated to have elevated levels of hepcidin. Furthermore, patients with transfusional iron overload have been shown to express elevated hepcidin levels. The NHANES III study preceded the identification of hepcidin as an important factor in the development of anemia, and therefore its potential role in the etiology of unexplained anemia was not assessed.

Regulation of hepcidin synthesis is complex and includes a number of inflammatory-mediated cellular pathways. Hepcidin is an acute phase reactant potently induced by IL-6, and hepcidin is implicated in mediating iron-limited erythropoiesis in patients with acute and chronic inflammatory states.[26,29] A recent analysis of a subgroup of participants in the InCHIANTI study examined the association between urinary hepcidin levels, proinflammatory markers, and anemia, and found that although IL-6 and C-reactive protein were associated with anemia and low iron status, they were not associated with higher urinary hepcidin, leading the authors of the study to postulate that increased hepcidin synthesis occurs only in situations of overt inflammation.[30] As with all disease association studies, these findings will need to be confirmed in larger cohorts of patients and will also benefit from further analysis using an assay of serum hepcidin. However, the observations do support our hypothesis that anemia may be mediated through hepcidin-independent proinflammatory pathways, such as TNFα.

Leptin—an adipokine associated with inflammation, body fat mass, and energy metabolism—has also been recently shown to induce hepcidin via JAK2/STAT3 signaling, potentially linking obesity to inflammation and iron homeostasis.[31] Multiple polymorphic alleles within the leptin gene influence leptin expression,[32] and low leptin levels have been associated with impaired EPO responsiveness of erythroid progenitors and the syndrome of frailty in the elderly population.[33] Hepcidin is also downregulated by hypoxia, and recent studies reveal that AI patients who respond to high-dose EPO therapy have concomitant decreases in hepcidin.[34] However, it remains to be determined whether hypoxia-inducible factor 1α, the body's primary hypoxia sensor, directly suppresses hepcidin synthesis.

A growing body of evidence links low vitamin D levels with poor outcomes associated with cardiovascular disease and various cancer diagnoses. As vitamin D supplementation among the population is becoming more and more commonplace in general clinical practice, our group examined the correlation of low vitamin D levels with anemia in phases 1 and 2 of NHANES III (1988–1994). Anemia was defined as a hemoglobin < 13g/dL and < 12g/dL for men and women, respectively. Anemia subtypes (nutritional, AI, unexplained anemia, chronic renal disease) were defined using phase 2 data as described.[3] In keeping with generally accepted guidelines, we classified vitamin D deficiency as vitamin D_3 levels of < 20 ng/dL. Statistical comparisons were performed using logistic regression models that adjusted for the characteristics used for sampling (age, sex, race/ethnicity). In phases 1 and 2 of NHANES III, we examined the association between anemia and vitamin D levels in men and women older than age 60 years ($n = 5100$) and found that vitamin D deficiency was associated with anemia independent of age, sex, race/ethnicity, with the odds for anemia being increased approximately 60% in the presence of vitamin D deficiency (odds ratio [OR] 1.6; 95% CI [1.37;1.95]; $P < .05$). Using phase 2 data, we next examined the prevalence of vitamin D deficiency in anemia subtypes in men and women older than age 60 years ($n = 2657$) and found that, among those with anemia, vitamin D deficiency was most prevalent among those with AI. The risk of AI was significantly increased in vitamin D-deficient versus nondeficient participants (OR 1.85; 95% CI [1.64;2.07]; $P < .05$). These are the first population-based studies demonstrating an association between vitamin D deficiency and anemia, particularly AI, in an older adult cohort (T. Perlstein and G. Vanasse, manuscript submitted, 2010) and provide compelling evidence that vitamin D deficiency may be a previously unrecognized contributor to the development of anemia in relatively healthy older individuals and is particularly prevalent among those with AI. The potential efficacy of vitamin D in ameliorating inflammatory anemia in elderly patients and the physiologic mechanisms by which vitamin D may abrogate anemia remain to be studied.

Unexplained Anemia in the Elderly Population

The pathophysiology of unexplained anemia in elderly patients is poorly understood, and it remains primarily a diagnosis of exclusion. Although myelodysplasia and other uncommon causes of anemia may potentially explain a portion of those with unexplained anemia, their combined contribution is felt to be relatively low. The impact of vitamin deficiency, beyond iron, vitamin B_{12}, and folate deficiencies, or the potential impact of subclinical renal disease on the development of unexplained anemia remains poorly studied. We postulate that overexpression of proinflammatory cytokines is an important determinant of unexplained anemia in elderly patients, and that they induce anemia by suppression of erythroid colony formation (MIF/TNFα/IL-1ß) on the one hand and impairment of iron utilization (IL-6/hepcidin) on the other. We suggest that the classic anemia of inflammation is at one end of the spectrum of the two inflammatory pathways, but that patients may also develop clinically apparent anemia in a proinflammatory state without having the classic iron abnormalities described for the anemia of inflammation. In our analysis of the association of vitamin D deficiency with anemia in older individuals in NHANES III, we did not find a correlation between low vitamin D levels and unexplained anemia. Ferrucci et al[35] used data collected from a representative sample of elderly individuals enrolled in the InCHIANTI study to examine the association between proinflammatory mediators and elderly subjects with unexplained anemia. In this study, 42 of 124 anemic individuals met the criteria for unexplained anemia. The

unexplained anemia cohort was found to have low serum EPO levels, low levels of proinflammatory markers, and low lymphocyte counts, thus leading the authors to conclude that the blunted EPO response associated with unexplained anemia was not associated with increased levels of inflammation.[35] The main limitations of this are its small sample size and by its sole reliance on measurements of serum levels of proinflammatory mediators (IL-6, TNFα, and C-reactive protein) used to define the presence of a chronic inflammatory state.

EPO and Anemia in the Elderly Population

EPO is the major cytokine influencing red blood cell development and is induced in the setting of anemia through an oxygen-sensing mechanism. Impaired EPO responsiveness of the hematopoietic stem cell has been implicated in the pathophysiology of anemia in elderly patietns.[36] The Baltimore Longitudinal Study on Aging demonstrated that EPO levels rose with age in healthy, nonanemic individuals, and that the slope of the rise was greater for individuals without diabetes or hypertension.[37] Those with anemia also had a lower slope of rise, suggesting that anemia reflected a failure of a normal compensatory rise in EPO levels with age. Low EPO levels have been preferentially associated with unexplained anemia in the elderly population,[35] but the mechanism for this inadequate EPO response remains to be determined, and the findings of this study need to be confirmed in larger cohorts of elderly patients. Taken together, this inappropriately reduced EPO response suggests progressive EPO resistance of the hematopoietic stem cell in the face of aging. Whether this reflects inflammatory cytokine-mediated impairment of normal EPO-dependent cellular pathways or other complex, age-related comorbidities and decreases in renal function that blunt the EPO response to anemia—or a combination of the two—remains to be determined. In some patients, this is primarily manifested as EPO insufficiency; in others, there is sufficient hepcidin expression to induce the classic features of AI. Supporting this hypothesis is the observation that the concomitant administration of EPO and intravenous iron has shown some success in ameliorating certain subgroups of patients with inflammation-associated anemia.[38] An alternative and as-yet untested hypothesis is that red blood cell life span may be shorter in older individuals, resulting in a compensatory rise in EPO levels in response to increased red blood cell turnover.

Anemia and Human Immunodeficiency Virus Disease

As the population of those infected with the human immunodeficiency virus (HIV) continues to age, it provides us with an opportunity to study the impact of a chronic proinflammatory state on an aging population. Anemia is the most common hematologic morbidity of HIV infection, and is associated with decreased survival, increased progression of disease, and decreased quality of life. As the HIV^+ population ages, it is at risk for anemia related to HIV, as well as the anemia of aging. The etiology of anemia in the setting of HIV infection is multifactorial and includes opportunistic infections, decreased EPO levels, effects on the kinetics of hematopoietic cell differentiation, nutritional deficiency, and associated malignancy and medications.[39]

To better study anemia in an aging HIV population, our group is currently involved in analyzing anemia within The Veterans Aging Cohort Study (VACS), an observational study of 3213 veterans with, and 3161 demographically similar veterans without, HIV infection. More than 75% of VACS subjects are black or Latino, and the median age is 54 years. The VACS cohort is designed for the study of an aging population. Within the HIV population, patients older than age 50 are considered "older," and, as we have hypothesized, they are likely to show the trends seen in older patients earlier than their HIV^- counterparts. Also, an age-stratified population like VACS should allow us to study whether the effects of aging on the development of anemia reflect a spectrum of changes that develop over the patients' lifetime. We have performed a preliminary study of anemia and survival in relation to MCV in 6870 HIV-infected men within the Veterans Administration (VA) system using data from the centralized VA records. Anemia was defined by the WHO standard as a hemoglobin <13 g/dL, and characterized by MCV as microcytic (< 80 fL), normocytic (81–98 fL), or macrocytic (> 98 fL). Anemia was present in 35% of HIV^+ men, with the following distribution: microcytic, 11%; normocytic, 74%; and macrocytic, 15%. In a multivariate Cox proportional hazards model of survival, adjusted for age, hepatitis C, log viral load, CD4 count, HAART (Highly Active Antiretroviral Therapy) therapy, diabetes, and liver disease, the hazard ratio was 1.5 for microcytic anemia, 1.6 for normocytic anemia, and 2.5 for macrocytic anemia, with macrocytic anemia being an independent predictor of decreased survival ($P < .05$) (G.V. and N.B., manuscript in preparation). Further studies examining the role of inflammation in the development of anemia subtypes in VACS are presently underway.

Conclusions

Anemia is a significant problem in elderly patients. Although many anemic elderly patients can be diagnosed with nutritional deficiency, anemia of chronic inflammation or comorbid diseases that explain their decreased hematocrit, the etiology of anemia in a significant fraction remains obscure. The impact of inflammatory mediators, EPO insensitivity, or other factors that may act on the hematopoietic stem cell to decrease erythropoiesis are under active study and should serve to elucidate the pathophysiology of this important cause of morbidity and mortality in elderly individuals. A better understanding of the pathophysiology of anemia in the elderly population should provide critical entry points for interventions that will improve survival and quality of life in the aging population.

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