Safety Issues in Iron Treatment in CKD

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Summary: Intravenous iron products are essential for the treatment of anemia in end-stage renal disease patients maintained on hemodialysis. Although proper use of these compounds is necessary for the prevention of iron deficiency, their indiscriminate use could potentially cause insidious adverse consequences. Iron overload can intensify the chronic kidney disease–associated oxidative stress, inflammation, and cardiovascular disease; increase the risk of infections; worsen the severity of type 2 diabetes; and exacerbate neurologic and cognitive dysfunction. These and other adverse effects largely are mediated by iron-catalyzed generation of reactive oxygen species. Unlike conventional oral iron products, the newly released iron-containing phosphate binder ferric citrate has been shown to increase iron stores in end-stage renal disease patients. Therefore, iron indices should be monitored in patients receiving this product. Two published studies have shown a high prevalence of hepatic iron loading among hemodialysis patients treated with erythropoiesis-stimulating agents and intravenous iron compounds. Given the potential risks related to iron treatment in this vulnerable population, studies to better understand safety are needed.
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Observational studies consistently have found an association between the severity of anemia and adverse outcomes in patients with chronic kidney disease (CKD). These findings prompted several randomized clinical trials in currently dialysis-independent and dialysis-dependent patients with CKD to test the hypothesis that correction of anemia may improve cardiovascular outcomes.[1–3] However, data analysis in those trials showed that patients assigned to the normal or near-normal hemoglobin targets experienced higher adverse outcomes than patients assigned to the lower hemoglobin targets. In the same studies, the subgroup of patients whose hemoglobin level could be normalized did considerably better.[1,4] In addition, the minority of end-stage renal disease (ESRD) patients who naturally maintain normal hemoglobin levels without requiring erythropoiesis-stimulating agents (ESAs) do not experience increased mortality.[5] Despite high doses of ESA and intravenous (IV) iron products, only a minority of patients randomized to the normal (21% in Correction of Anemia with Epoetin Alfa in Chronic Kidney Disease (CHOIR)) or near-normal hemoglobin (38% in Cardiovascular Reduction Early Anemia Treatment Epoetin β (CREATE)) groups achieved the target.[1–3] As a result of implementation of the bundling reimbursement system in the United States and the high cost of ESAs, the ESA dosing has decreased and IV iron has increased and ferritin levels have continued to increase. This trend could potentially be harmful because iron could[6] amplify oxidative stress and inflammation, which are constant features of CKD/ESRD.[7–10]

The potential role of high doses of ESA in the pathogenesis of cardiovascular disease (CVD), thromboembolic disorders, and other complications has been reviewed previously[11,12] and will not be addressed here. This article provides an overview of the potential safety concerns with use of iron preparations. At physiologic ranges, iron is protein-bound so that it is safely liganded and kept catalytically inactive. However, when improperly liganded, H_2O_2, which is generated by mitochondria and inflammatory cells, reacts with ferrous iron. This leads to conversion of Fe^{2+} to Fe^{3+} and generation of the hydroxyl radical $\cdot OH$, which is a highly reactive free radical. Superoxide, produced by mitochondria and mono-oxygenase enzymes, in turn, converts the ferric back to ferrous iron (Haber–Weiss reaction), which provides the fuel for continuous $\cdot OH$ production and perpetuation of oxidative stress. Because free or incompletely liganded iron is catalytically active and causes oxidative stress and tissue injury, absorption, transport, and storage of iron, its retrieval from storage sites and its uptake by cells are tightly regulated. On each occasion, iron is bound to proteins such as transferrin in the plasma and ferritin in hepatocytes and reticuloendothelial cells, hemoglobin in erythrocytes, and myoglobin in myocytes, preventing exposure to the redox active iron.

When administered intravenously the biologic safeguards for handling and regulating iron are partially bypassed. Numerous in vitro and in vivo studies have

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shown the ability of IV iron compounds to promote oxidative stress and cell injury/death in cultured renal proximal tubular epithelial cells and endothelial cells.[13,14] Studies in CKD rats have shown persistent oxidative stress in the aorta, myocardium, and other organs several weeks after IV iron administration.[9,15] IV iron products, evaluated in a relatively small number of patients with ESRD, have shown significant increases in biomarkers of oxidative stress including lipid, protein, and DNA oxidation products and inflammatory mediators.[16–18] In the presence of inflammation, the release of proteases and secretion of hydrogen ion by lysosomes results in dissociation of iron from the binding proteins, further enabling iron to catalyze formation of reactive oxygen species (ROS).

IRON AND THE CARDIOVASCULAR SYSTEM

There is emerging evidence supporting the role of iron overload in cardiovascular complications. A recent study comparing the effect of oral versus parental iron in the CKD population was terminated early because the group receiving IV iron experienced a 2.51-fold higher incidence of cardiovascular events ($P < .001$) and a two-fold higher incidence of hospitalization for heart failure in the IV compared with the oral iron-treated group.[19]

Studies conducted in animals and cultured cells have identified the mechanisms of adverse cardiovascular effects of excess iron. For example, in cultured human endothelial cells, exposure to IV iron products inhibited proliferation, induced apoptosis, and up-regulated monocyte adhesion.[13,20] Endothelial dysfunction and injury play a central part in the pathogenesis of atherosclerosis, thrombosis, and CVD. Oxidative stress induces endothelial dysfunction via ROS-mediated inactivation of nitric oxide, depletion of nitric oxide synthase co-factor (tetrahydrobiopterin), and accumulation of the nitric oxide synthase inhibitor asymmetric dimethylarginine. In fact, IV iron products significantly reduce acetylcholine-induced vasorelaxation in isolated artery rings.[13,21] Similar responses have been identified in normal human subjects.[20,22] Increased plasma asymmetric dimethylarginine levels potentially predict cardiovascular events in ESRD patients.[23,24]

Carotid artery media-intima thickness, a marker of atherosclerosis, may be associated with the annual cumulative dose of IV iron preparations in ESRD patients.[25,26] Also, the severity of atherosclerotic lesions has been shown to be greater in patients with carotid lesions who had received long-term IV iron preparations.[26] Rabbits fed a high-cholesterol diet accumulate iron in the atherosclerosis plaques,[27,28] and atherosclerotic lesions in apolipoprotein E–deficient mice contain significant amounts of iron.[28] Iron accumulation also is higher in abdominal aortic aneurysm walls compared with non–aortic aneurysm walls.[29] Interaction between iron and lipoproteins can lead to foam cell apoptosis and plaque instability, a process that can precipitate acute cardiovascular events.[30–32] Conversely, reducing iron availability by chelation therapy in animal models of carotid injury significantly inhibits intimal thickening and vascular smooth muscle cell proliferation.[33] These mechanisms by which iron may exacerbate atherosclerosis and contribute to the development of vascular calcification could be relevant for patients with CKD.[34]

CONTRIBUTION OF IRON OVERLOAD TO SYSTEMIC INFLAMMATION AND INFECTIONS

CKD results in simultaneous activation and deficiency of the immune system.[35] Activation of the immune system in this population is responsible for systemic inflammation, a driving force behind many CKD-associated complications including atherosclerosis, CVD, cachexia, anemia, and numerous other morbidities. The CKD-associated immune deficiency results in impaired response to vaccination, high incidence, increased severity, and poor outcome of infections.

The CKD/ESRD-induced systemic inflammation is caused by activation of the innate and adaptive immune responses and impairment of the anti-inflammatory regulatory factors, events that are mediated by the following: (1) accumulation of proinflammatory oxidized low-density lipoprotein coupled with the deficiency of high-density lipoprotein and its reduced anti-inflammatory capacity[36,37]; (2) disruption of the intestinal epithelial barrier structure and altered intestinal microbiome, which promote systemic inflammation by enabling influx of endotoxin and other noxious products in the systemic circulation[38]; (3) increased population and sustained activation of monocytes, which is marked by their increased basal expressions of integrins, Toll-like receptors 2 and 4, and spontaneous production of cytokine and reactive oxygen species[39,40]; (4) impaired inhibitory activity and a reduced population of regulatory T lymphocytes, which are essential for mitigating inflammation[41]; (5) spontaneous activation, degranulation, and increased basal production of ROS by circulating polymorphonuclear leukocytes (PMNs)[39]; (6) increased ROS production and chemokine expression by the cellular constituents of various tissues[10,41,42]; and (7) co-existing conditions such as autoimmune diseases and diabetes.

The main causes of CKD-associated immune deficiency include the following: (1) depletion of dendritic cells, which are the primary antigen-presenting cells[43]; (2) reduced $CD4^+$ helper T-lymphocyte/$CD8^+$ suppressor T-lymphocyte ratio and depletion of naive and central memory T lymphocytes[44]; (3) diffuse depletion of B lymphocytes, which contributes to impaired

antibody production[45,46]; (4) defective T-cell and natural killer cell proliferation[47,48]; and (5) depressed phagocytic capacity and increased apoptosis of the neutrophilic PMN.[49,50]

The immune system is affected adversely by both iron deficiency and iron overload. For example, iron deficiency can cause thymus atrophy and reduce T lymphocytes,[51] and iron overload can lead to immune deficiency and susceptibility to infections by several mechanisms. For example, iron overload has been shown to deplete helper $CD4^+$ T lymphocytes and increase expansion of $CD8^+CD28^-$ suppressor T cells.[52,53] Lymphocytes have a poor ability to sequester iron such that excess iron impairs proliferation and promotes apoptosis in these cells.[54] IV iron compounds may hinder lymphocytes owing to oxidative stress caused by iron-catalyzed ROS production. This assumption is supported by in vitro experiments that showed a time-dependent increase in intracellular ROS and diminished survival of $CD4^+$ T-cells after incubation in media containing therapeutic concentrations of IV iron preparations.[55] High doses of IV iron products also have been shown to impair PMN's phagocytic and bactericidal capacities.[56–59] Finally, iron preparations decrease whereas ESAs, which reduce iron stores, enhance the response to hepatitis B vaccination,[60] and iron overload increases the risk and severity of infections[61,62] by facilitating microbial growth and virulence.[63] In fact, a recent trial comparing oral with IV iron in CKD patients showed a 2.12-fold ($P < .006$) higher incidence of infections in the IV iron group.[19] A retrospective analysis of the US Renal Data System database including a large cohort of hemodialysis patients (~117,000 patients) comparing bolus versus maintenance dosing of IV iron showed a significantly greater risk of infection-related hospitalizations in the bolus IV iron–treated group.[64]

There is evidence that indiscriminate use of IV iron products intensify systemic inflammation: (1) high doses of IV iron compounds result in increased ROS generation, cytokine activation, and loss of mitochondrial membrane potential in the blood mononuclear cells in ESRD patients[65]; (2) in vivo iron loading results in the formation of proinflammatory M1 macrophages that sustain local inflammation and prevent the healing process by secreting high levels of proinflammatory cytokines (tumor necrosis factor-α, interleukin [IL]-1, IL-6, and IL-23) and generating large quantities of ROS, and nitrogen radicals[66–69]; and (3) ROS trigger inflammation by activating nuclear factor-κB directly or indirectly via formation of oxidized lipids and lipoproteins.

Taken together the available data provide convincing evidence for the presence of impaired adaptive immunity and inflammation in advanced CKD and the possibility of exacerbation by indiscriminate use of iron preparations.

POTENTIAL ADVERSE EFFECTS OF IV IRON ON THE LIVER

Liver is the major site of iron storage and hepatic tissue iron content closely correlates with total body iron stores.[70] There is increasing evidence that iron overload commonly occurs in ESRD patients receiving IV iron and ESA in compliance with the currently accepted guidelines that permit use of IV iron in dialysis patients with serum ferritin values as high as 500 to 900 μg/L. This is based on the assumption that increased ferritin might not represent iron overload. However, a number of recent studies have documented a high prevalence of iron overload in hemodialysis patients receiving standard anemia treatment with ESAs and IV iron preparations. By using a superconducting quantum interference device to measure the nonheme iron content of the liver, Canavese et al[71] found iron overload in 70% of their patients, the majority of whom (70%) had serum ferritin values less than 500 μg/L. They further showed that serum ferritin values exceeding 340 μg/L were associated with iron overload. Similarly Ferrari et al[72] and Rostoker et al[73] recently showed a dramatic increase of liver iron contents approaching those found in hemochromatosis with routine administration of IV iron preparations in hemodialysis patients. It thus is clear that iron overload commonly occurs in ESRD patients treated with ESAs and IV iron according to the currently accepted guidelines. Whether this causes any damage or injury to the liver is unclear.

It should be noted that mildly increased and even normal levels of iron in the liver can be damaging when accompanied by other hepatotoxic agents/conditions such as alcohol, drugs, or viral hepatitis.[61] By intensifying the pathogenicity of microorganisms, impairing the macrophage and lymphocyte function, and stimulating fibrogenic pathways, excess iron can intensify liver injury. Chronic viral hepatitis caused by hepatitis C and B viruses is quite common among dialysis populations worldwide, particularly in endemic regions. There is irrefutable evidence that an increased iron burden can intensify and iron depletion by phlebotomy can ameliorate hepatitis and modify the response to interferon therapy.[63,74–75] In addition, the stainable iron level in hepatocytes and portal tract cells is a predictor of progression and clinical outcomes in advanced chronic hepatitis C.[76] Therefore, caution should be exercised in the use of IV iron in patients with liver disease.

EFFECTS OF IRON ON DIABETES

Type 2 diabetes is the leading cause of CKD worldwide. As described in a review by Swaminathan et al,[77] overt and subtle iron overload plays a part in the

pathogenesis and progression of diabetes. Overt iron overload of all etiologies (eg, hereditary hemochromatosis, transfusion-induced hemosiderosis, excess dietary iron, porphyria cutanea tarda, and mitochondrial iron overload [Fredrich's ataxia]) results in an increased risk of type 2 diabetes. Iron overload induces insulin deficiency by promoting apoptosis of pancreatic β cells. Pancreatic β cells are highly susceptible to oxidative stress because of a strict reliance on mitochondrial glucose metabolism, their limited antioxidant defense capacity,[78] and their high capacity for iron uptake via divalent metal transporters.[79] In addition to frank iron overload, subtle increases in dietary heme content (red meat) and a modest increase of body iron stores are associated with insulin resistance,[80,81] metabolic syndrome,[82,83] and gestational diabetes.[84] There is evidence that reducing body iron stores with bloodletting or blood donation ameliorates insulin resistance and improves glycemic control in patients with type 2 diabetes.[85,86] In fact, iron deficiency improves insulin sensitivity and decreases the risk of diabetes.[87]

Protein glycation plays a major part in the pathogenesis of renal and vascular complications of diabetes by sustaining the catalytic activity of transition metals,[88] events that can participate in the pathogenesis of oxidative stress. Iron and other transition metals facilitate hyperglycemia-induced protein glycation as evidenced by the reduction of glycated hemoglobin with iron chelation in diabetic animals.[89] There is preliminary evidence suggesting the contribution of iron to the progression of diabetic nephropathy.[90] Plasma NTBI (non transferrin bound iron) concentration commonly is increased in diabetic patients and has been implicated in the pathogenesis of vascular complications.[91] In fact, preliminary studies have shown a marked reduction in proteinuria with iron chelation therapy in patients with diabetic nephropathy.[92]

COGNITION AND NEUROLOGIC DISORDERS

Advanced CKD frequently is associated with peripheral neuropathy, sleep disorder, depression, dysregulation of neurohormonal systems, cognitive dysfunction, and even frank encephalopathy with severe uncontrolled uremia. Cognitive impairment is a common but underdiagnosed neurologic consequence of advanced CKD and ESRD.[93] Cognitive impairment in CKD is mediated in part by endocrine disorders, uremic toxicity, psychosocial stress, medications, and dialysis-induced hypotension.[94–96] Kell[97] identified oxidative stress as the common mediator of brain injury in Alzheimer's disease, Parkinson's disease, Friedreich's ataxia, amyotrophic lateral sclerosis, multiple sclerosis, age-related macular degeneration, and prion diseases. In many instances catalytically active iron (and sometimes copper) accumulates in the affected brain tissues via binding to the abnormal disease-specific ligands. Relentless production of hydroxyl radical by the catalytically active iron plays a role in the progression of these neurologic disorders. This assumption is supported by the salutary effect of iron chelation therapy in human beings or experimental animal models of these diseases.[98] Induction of CKD with subtotal nephrectomy in experimental animals results in diffuse oxidative and nitrosative stress and massive accumulation of nitrotyrosine (a byproduct of ROS interaction with nitric oxide) in the white and grey matters of the brain.[98] It is of interest that administration of erythropoietin, which shifts tissue iron stores to erythrocytes for hemoglobin synthesis, has been shown to improve cognitive function and neurohormonal abnormalities in dialysis patients.[99,100] The salutary effects of anemia treatment with erythropoietin on ESRD-associated cognitive dysfunction have been attributed to the correction of anemia and the direct trophic action of this hormone on the brain. In addition, the reduction in tissue iron burden may be an equally important factor in this case.

A MORE CONSERVATIVE APPROACH TO IRON TREATMENT FOR PATIENTS ON DIALYSIS

As noted earlier, carefully conducted investigations have shown a high prevalence of hepatic iron overload in hemodialysis populations receiving IV iron and ESA products according to the accepted anemia treatment guidelines in Western countries. Compared with Western countries, the Japanese guidelines for prescription of IV iron in dialysis patients are far more conservative and recommend IV iron only when the transferrin percentage saturation is less than 20% and the ferritin level is less than 100 ng/mL.[101] Despite this more measured approach, outcomes of Japanese dialysis patients are as good or better than their American counterparts. Given the potential safety issues with aggressive IV iron treatment, and the lack of well-powered studies to examine safety, a more conservative approach to iron therapy should be considered in the United States.

CONCLUSIONS

Although proper use of IV iron products is critical for the prevention of iron deficiency in hemodialysis patients, their indiscriminate use could result in adverse consequences. Studies have shown extensive liver iron content in hemodialysis patients receiving ESA and IV iron according to the accepted guidelines. In recent years, the mean serum ferritin level in the United States has increased to more than 700 ng/mL. Given the potential deleterious effects of iron overload in

hemodialysis patients, revision of the anemia treatment guidelines and practice is needed.

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