Anemia Management and the Delay of Chronic Renal Failure Progression

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Abstract. Interstitial fibrosis plays a key role in the progression of chronic kidney diseases. Analysis of the biologic effects of erythropoietin and of the pathophysiology of interstitial fibrosis suggest that treatment with epoetin may slow the progression of chronic kidney disease, both by decreasing interstitial fibrosis and by protecting against its consequences. The results of two small prospective studies and of a retrospective one also suggest that treatment with epoetin may have such protective effects.

End-stage renal disease is a major health problem resulting in considerable increase of morbidity and mortality, in decrease of quality of life, and in heavy costs from renal replacement therapies. Furthermore, for the past decade, the incidence of ESRD has been relentlessly increasing at an annual rate of about 6 to 8% in most European countries, and even more rapidly in the United States. Slowing the progression of renal failure thus appears to be a major therapeutic challenge. Besides treatment of the underlying disease whenever possible, the main therapeutic tools that are available to slow the progression of renal failure are optimal control of BP, use of angiotensin converting enzyme (ACE) inhibitors or of angiotensin II receptor blockers (ARB), and protein restriction (reviewed in 1, 2). The efficacy of these therapies is however limited, and there is a need for additional treatments. Among the therapeutic interventions that could possibly slow the progression of CKD is correction of anemia through administration of epoetin.

Mechanisms of Progression of Chronic Renal Failure

Regardless of the underlying renal disease, progression of CKD leads to a common histologic endpoint, the “end-stage kidney,” that is characterized by nonfunctional sclerotic glomeruli, atrophied tubules, and a fibrotic interstitium. Nephron destruction is not only a direct consequence of the underlying renal disease, but it is also due to progressive glomerulosclerosis secondary to nephron reduction, and to tubular damage associated with interstitial fibrosis (Figure 1).

Glomerulosclerosis

Various experimental studies in rats have demonstrated that reduction in nephron number favors the occurrence of glomerulosclerosis (reviewed in 3). Histological and physiologic studies of rats undergoing subtotal nephrectomy have shown that nephron loss increases capillary pressure and flow in the remaining glomeruli and induces hypertrophy of these glomeruli, which in turn leads to podocyte injury and to overproduction of extracellular matrix by mesangial cells (reviewed in 4–6). Experimental data suggest that PDGF and TGF-β play an important role in this process (7,8). Furthermore, they show that the occurrence of glomerulosclerosis can be prevented by the use of ACE inhibitors or by a low protein diet (reviewed in 9). In humans, sequential studies of few patients with an important reduction of their renal mass have also shown that a decrease in nephron number can also be responsible for the development of glomerulosclerosis (10,11).

Interstitial Fibrosis

The notion that interstitial fibrosis plays an important role in the progression of CKD is mostly supported by two kinds of observations. First, for many renal diseases, there is a striking correlation between renal function at the time of biopsy and severity of interstitial fibrosis on renal biopsy (reviewed in 12). Second, the extent of interstitial fibrosis is the best histologic prognostic marker of most renal diseases (reviewed in 13). Studies of human renal biopsies and of experimental models of CKD have shown that one of the most striking features associated with interstitial fibrosis is the presence of so-called “atubular glomeruli,” which consist of non sclerotic glomeruli that are not linked to a tubule (14,15). This suggests that interstitial fibrosis decreases nephron number by inducing tubular lesions that lead to tubular destruction, with formation of nonfunctional atubular glomeruli. Analyses of renal biopsies obtained from patients with renal failure and from animals with various experimental renal diseases have also shown that, besides tubular destruction, interstitial fibrosis is associated with a decrease in the number of peritubular capillaries (reviewed in 16), and that there is a good inverse correlation between the severity of interstitial fibrosis and the number of peritubular capillaries as well as between renal function and the number of interstitial capillaries (12,17,18). Thus, it is likely that the destruction of peritubular capillaries links interstitial...
fibrosis and tubular destruction (Figure 1). The combination of an overproduction of thrombospondin-1 by tubular and interstitial cells, and of a loss of VEGF synthesis by tubular cells could play a key role in this process (reviewed in 16). However, the ischemic lesions of tubular cells are probably due not only to a destruction of peritubular capillaries, but also to an interposition of extracellular matrix between capillaries and tubules, which decreases oxygen delivery to tubular cells. Furthermore, they are worsened by an increased consumption of oxygen by the remaining tubular cells, and this is likely to be one of the mechanisms by which proteinuria or high protein diet accelerate the progression of CKD (19). Finally, obstruction of peritubular capillaries may also increase glomerular capillary pressure, and thus enhance glomerular damage (dotted line). Proteinuria enhances tubular dysfunction and, thus, interstitial fibrosis (dotted line).

The Effects of Epoetin Therapy
It is well known that correcting anemia with epoetin increases oxygen delivery to tissues and thus reduces hypoxia. Nevertheless, this treatment may have other beneficial effects, including protection against oxidative stress and apoptosis (Figure 2).

The links between oxidative stress and anemia come mostly from the fact that erythrocytes represent a major antioxidant component of the blood (reviewed in 32). Their antioxidant effects are mediated through the glutathione system, enzymes such as superoxide dismutase or catalase, and cellular proteins that are devoid of enzymatic activity but can react with reactive oxygen species, such as low-molecular weight proteins of the erythrocyte membrane, vitamin E or coenzyme Q. Furthermore, erythrocytes can regenerate consumed redox equivalents through the pentose phosphate pathway, and through reduction of oxidized glutathione by glutathione reductase. In addition to increasing the number of red blood cells, epoetin may also reduce oxidative stress by increasing the antioxidant potential of erythrocytes. Experimental data have shown that the binding of epoetin to its receptor activates NF-κB, which in turn enhances the expression of genes encoding proteins, such as superoxide dismutase or glutathione, that have antioxidant properties (33).

The ability of erythropoietin to promote survival of red blood cells via the binding to its receptor has been clearly established for a long time. It appears to be mostly mediated by an overexpression of anti-apoptotic proteins such as Bcl-xL and Bcl-2, but also X-IAP or c-IAP, and by the activation of PKB/Atk that phosphorylate proapoptotic molecules such as Bad and caspase 9 (34,35). Recent in vivo data have shown that the anti-apoptotic effects of epoetin can also be observed with other cell types expressing the erythropoietin receptor, such as neuronal cells, and that systemic administration of epoetin can dramatically decrease the neurologic consequences of various
experimental injuries (36–39). In vitro, epoetin has also been shown to protect endothelial cells or vascular smooth muscle cells against apoptosis (40,41). One can thus speculate that this effect also exists for other cells that express the erythropoietin receptor, such as proximal tubular cells and renal endothelial cells (42). Furthermore, in vitro and in vivo experiments suggest that erythropoietin may also have proangiogenic properties (43,44).

**Potential Effects of Epoetin Treatment on the Progression of Chronic Kidney Disease**

As previously outlined, hypoxia of tubular cells appears to be the main link between interstitial fibrosis and tubular destruction (reviewed in 45). Thus, correcting anemia with epoetin should increase oxygen delivery to tubular cells, decrease tubular damage, and ultimately protect against nephron loss induced by tubular injury. Furthermore, because hypoxia stimulates the production of extracellular matrix by tubular cells and by renal interstitial fibroblasts, as well as the release of profibrotic cytokines such as TGF-β, decreasing hypoxia should slow the rate of extracellular matrix accumulation (24,31). Part of the beneficial effects of epoetin on hypoxia may also be mediated through its proangiogenic properties (43,44), that will oppose the decrease in the number of interstitial capillaries.

Besides its effects on hypoxia, treatment with epoetin could also have beneficial effects on the progression of CKD through a reduction of oxidative stress. In cases of chronic reduction in nephron number, oxygen consumption by the remaining nephrons increases, which leads to increased production of reactive oxygen species (reviewed in 19). This oxidative stress probably enhances not only tubular damage but also interstitial inflammation and fibrosis. Experimental data have shown that oxidative stress stimulates the production of extracellular matrix by fibroblasts. For example, in vitro, noncytolytic doses of hydrogen peroxide stimulate collagen synthesis by renal fibroblastic cells, as well as TGF-β production (29). Similarly, lipid peroxidation products, which are produced in increased amounts in response to oxidative stress, upregulate collagen production by fibroblasts (27,28). Furthermore, in vitro, reactive oxygen species can activate NF-κB in proximal tubular cells, and thus the release of proinflammatory molecules such as MCP-1 (26) The physiologic relevance of these in vitro effects is suggested by experiments showing that, in vivo, treatment of rats with antioxidants can protect against the development of interstitial fibrosis, whereas deprivation of antioxidants has opposite effects (29,30).

The anti-apoptotic effects of epoetin could also be beneficial for the progression of CKD, because apoptosis has been implicated in the progressive loss of tubular cells observed during CKD (46). For example, apoptosis appears to play an important role in the progression of tubular lesions observed in rats submitted to subtotal nephrectomy or to experimental anti-glomerular basement membrane nephritis (47,48). The mechanisms underlying the increased apoptosis of tubular cells are still poorly understood, but reactive oxygen species could play a role in this process (47,48). Thus, treatment with epoetin may have beneficial effects not only by protecting tubular cells against apoptosis, but also by decreasing the production of reactive oxygen species.

**Clinical Studies Suggest That Epoetin May Slow the Progression of Renal Failure**

The fact that correcting anemia does not accelerate the progression of CKD has been shown by different clinical studies performed in the early 1990s, and it is no longer questioned. Nevertheless, the question of whether it slows the progression of renal failure remains unanswered. So far, two prospective clinical studies including a relatively large number of patients and a retrospective study support this hypothesis (49–51).

The first study included 83 patients with severely impaired renal function (mean GFR 10 ml/min), and severe anemia (mean hematocrit 26.8%) (49). After a 2-mo stabilization period, 40 patients were randomly assigned not to receive epoetin and 43 to receive epoetin for their hematocrit levels to reach 35%. The patients were followed for 48 wk. No beneficial effect of epoetin could be demonstrated by simply comparing renal survival or GFR (GFR) decrease between the two groups of patients. Nevertheless, when the data were analyzed after the hematocrit levels of the patients in the epoetin group had reached target values (i.e., after week 16), the rate of GFR decline was three times slower in the treated group than in the control group (−0.13 ± 0.35 ml/min/mo versus −0.39 ± 0.65 ml/min/mo, P = 0.05).

The second study included 73 patients with anemia (mean hematocrit 27.4%) and severe renal failure (mean creatinine clearance 18.2 ml/min) (50). After an 8-wk stabilization period, the patients were randomly assigned to receive or not to receive epoetin. Thirty one patients were left untreated; 42 patients received epoetin to increase their hematocrit levels to 33 to 35%. During the 36-wk follow-up period, creatinine doubled in about 52% of patients in the treated group, and in more than 90% of patients in the control group (P < 0.0005). Furthermore, although 64% of patients in the control group required dialysis, only 33% of those in the epoetin group needed to start dialysis (P < 0.005).

More recently, a retrospective and noncontrolled study also suggested that epoetin treatment may slow the progression of renal failure (51). In this study, the authors compared 20 patients with chronic renal failure who were treated with epoetin with 43 patients who had a similar degree of renal failure but who were less anemic and thus did not receive epoetin. The rate of decline of creatinine clearance did not change over time in the control group, whereas, in the treated group, it was significantly slower after epoetin treatment had been started (−0.36 ± 0.16 ml/min per 1.73 m² per month versus −0.26 ± 0.15 ml/min per 1.73 m² per month; P < 0.05).

**Conclusion**

Different experimental data support the hypothesis that correction of anemia with epoetin may slow the rate of progres-
sion of renal failure, and few small clinical studies also suggest the importance of testing this hypothesis. The definite answer should come from a large clinical trial that is much awaited.

References

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