Anemia of Chronic Renal Failure

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Historical Background

In 1836 Richard Bright first commented on the pallor of patients with poor kidney function. The combination of pale mucous membranes and yellow-brown skin, sometimes darker on the face, may lead the examining physician to suspect the diagnosis of chronic renal failure (CRF). As documented by Erslev and others [1, 2], the degree of anemia is approximately proportional to the level of renal dysfunction as measured by blood urea nitrogen (BUN), serum creatinine, or creatinine clearance (Ccr). The worse the glomerular filtration rate (GFR), the lower the hematocrit (HCT) will be (Table 1). A plot of HCT versus serum creatinine is shown in Figure 1. When renal function is < 15% of normal, the HCT is almost always in the 20's. In the study of Hakim and Lazarus, the mean HCT for patients with a serum creatinine of 5 – 10 mg/dL was 29%, and for those with a serum creatinine > 10 mg/dL, it was 26% [3]. The HCT rarely falls < 20% due to uremia alone. If such a patient is encountered, other causes of anemia, such as iron deficiency, microangiopathic hemolysis, or multiple myeloma as a cause of chronic renal failure (CRF), should be sought.

Table 1. Equations Relating Renal Function and Hematocrit

HCT = –2.1 serum creatinine + 44, p < 0.001 (both sexes)
HCT = –2.52 serum creatinine + 45.9, p < 0.0001 (men)
HCT = –1.36 serum creatinine + 37.7, p < 0.0001 (women)
HCT = 0.48 Ccr – 0.0031 $(Ccr)^2$ + 26.0, p < 0.0001 (men)
HCT = 0.48 Ccr – 0.0031 $(Ccr)^2$ + 21.5, p < 0.0001 (women)

HCT = hematocrit (%), serum creatinine (mg/dL), Ccr = creatinine clearance (mL/min), Data taken from reference [2]

Physiology of Erythropoiesis

In 1950 Reissman showed that the hypoxic rat caused enhanced erythropoiesis in its parabiotic, non-hypoxic partner [4]. Then Erslev in 1953 demonstrated a hormone in anemic animals which stimulated erythropoiesis when injected into non-anemic recipients [5]. This factor did not affect white blood cells or platelets. Four years later Jacobson and colleagues reported that the kidneys must play a key role in the release of this hormone, erythropoietin (EPO) [6]. It then became accepted that the kidneys sense an oxygen deficit and respond by increasing EPO production. Under normal conditions, a plasma EPO

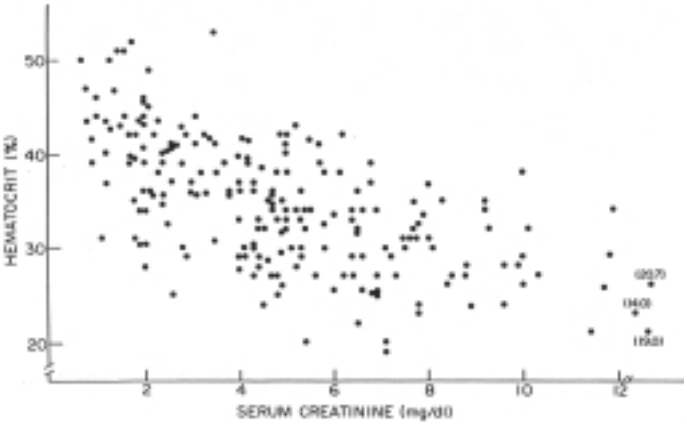

Figure 1. Concentrations of serum creatinine and hematocrits of individual patients from the Nashville Department of Veterans Affairs Medical Center (VA) Renal Clinic are plotted.

level of 8 – 18 mu/mL will maintain a baseline rate of erythrocyte synthesis, an adequate oxygen delivery to the kidneys, and thereby continued EPO synthesis [7]. In severe anemia (HCT < 20%) the plasma level of EPO may increase as much as 100-fold in an attempt to restore the erythrocyte mass to normal. Thus there is a hormonal feedback loop involving the kidneys which controls erythrocyte production.

In 1977 Miyake et al. purified EPO from human urine (from patients with aplastic anemia) [8]. However, the use of EPO as a therapeutic agent was severely hampered by the small quantities which could be extracted. By 1985 the human EPO gene had been isolated, cloned, and expressed in mammalian cell lines by 2 groups [9, 10]. Biologically active human EPO could now be produced in large quantities.

EPO is a heavily glycosylated protein consisting of a single strand of 165 amino acids with an equal amount of carbohydrates. Its molecular weight is 30,500 daltons [7]. The gene coding for EPO is found on human chromosome 7 and consists of 5 exons and 4 introns. A downstream enhancer is sensitive to hypoxia. An hypoxia-inducible factor (HIF-1) binds to this enhancer and acts as a transcription factor [7].

Koury et al. first demonstrated that EPO messenger RNA (mRNA) was localized to a small fraction of renal cortical interstitial cells near the base of proximal tubular cells [11]. The rate of EPO production correlated not only with HCT but also with the number of renal interstitial cells expressing EPO mRNA. In mild anemia EPO positive cells are present in small groups in the inner cortex. In maximum anemia (HCT < 15%), cells throughout the cortex possess EPO mRNA but still compose only 7% of the total interstitial cells. The mechanism by which this recruitment occurs is unknown.

EPO can also be produced by hepatocytes and other non-renal tissues in small quantities. However, this supply of EPO to the body is inadequate to restore normal hematopoiesis in the absence of normal renal parenchyma.

The erythrocyte progenitor cells which are the main targets for EPO are the mature burst-forming units-erythroid (BFU-Es) and colony-forming units-erythroid (CFU-Es) [7]. The EPO receptor on these cells is a 55,000 dalton member of the cytokine receptor superfamily. In the absence of EPO, these cells undergo apoptosis. In the presence of adequate quantities of EPO, CFU-Es are transformed into erythroblasts, then to reticulocytes, and finally to mature erythrocytes. Thus

the effect of EPO is not one of enhancing the proliferation of erythrocyte precursors, but rather of inhibiting apoptosis of these same cells.

Causes of Anemia Related to Chronic Renal Failure (CRF)

Anemia begins to be manifest when renal function (Ccr) is < 40% of normal, approximately when the serum creatinine is above 2 – 3 mg/dL. The erythrocytes of patients with CRF are normocytic and normochromic. Reticulocytes are diminished for the degree of anemia. Iron, folate and vitamin B_{12} stores are usually normal.

Decreased erythrocyte survival in the uremic state has been documented by 3 techniques: ^{51}Cr or ^{14}C-cyanate tagged red cells or by carbon monoxide exhalation [12]. In general, the hemolysis is mild and red cell survival is approximately 50% of normal. Erythrocytes transfused into uremic patients have a similarly shortened life span, while uremic red cells survive for a normal period when transfused into normal subjects [13]. Therefore, the cause of this hemolysis is deemed to be extracorpuscular. Neither maintenance hemodialysis nor chronic peritoneal dialysis (PD) improves red cell survival. However, this form of hemolysis is not a major reason for the anemia of CRF since a normal kidney-marrow feedback system would easily compensate for it.

There is also evidence that the uremic state inhibits production of erythroid stem cells and their offspring [13]. Among the purported inhibitors accumulating in uremic serum are spermine, spermidine and parathyroid hormone (PTH). Uremic animals have been shown by some investigators to have a subnormal response to exogenous EPO. Pharmacologic doses of EPO are sometimes required in dialysis-dependent anemic humans. Other studies have been unable to document any inhibition of erythropoiesis in CRF. Marrow inhibition is at best of minor significance in the pathogenesis of uremic anemia.

The most important cause of anemia in CRF patients is a deficient production of EPO in response to diminished renal oxygen delivery [7, 12, 13]. Serum levels of EPO are much less in uremia than in other types of anemia where there is normal renal function. As long as the kidneys were not surgically removed, a stable HCT in the 20's could be maintained in patients during the era prior to EPO therapy. Following total nephrectomy, all dialysis patients became transfusion-dependent. In our experience, an average of 2 units of red cell transfusions were required per month to maintain the HCT at 20% in anephric patients. Nothing else worked. Androgens had no effect. Iron was not indicated since iron stores were either normal or increased in anephrics. Before EPO therapy, only a successful renal transplant could correct transfusion-dependent anemia in an anephric patient.

Aggravating Factors

Problems that decrease the response of the marrow to endogenous or exogenous EPO can worsen uremic anemia (Table 2). Proinflammatory cytokines such tumor necrosis factor α (TNFα), and interleukins 1 and 6 (IL-1, IL-6) inhibit the effect of EPO on the bone marrow [7]. This may occur in inflammatory

Table 2. Causes of Anemia Related to Chronic Renal Failure

A. Primary Causes
1. Underproduction of EPO
2. Shortened erythrocyte survival due to uremia
3. Inhibitors of erythropoiesis accumulating in uremia

B. Aggravating Causes
1. Factors hampering EPO production or effect*
   a. Inflammatory states
   b. Infections
   c. Nephrectomy
   d. Aluminum accumulation
   e. Neoplasia
   f. ACE inhibitors
2. Deficiency states
   a. Iron
   b. Folate/B_{12}
   c. Malnutrition
3. Other hemolytic states
   a. Oxidant drugs/toxins
   b. Microangiopathic hemolysis (e.g. accompanying malignant hypertension)
   c. Hypersplenism
   d. Dialysis-related factors (hypotonic dialysate, hyperthermia, copper loading,etc.)
4. Hemorrhage/blood loss
   a. GI Tract (platelet defects may contribute)
   b. Dialysis-related blood losses
      (1) During vascular access puncture and needle removal
      (2) Into dialyzer and blood lines
      (3) For laboratory tests
5. Diseases causing both anemia and renal failure
   a. Sickle hemoglobinopathies
   b. Scleroderma
   c. Systemic Lupus Erythematosus
   d. Lead intoxication
   e.Paraprotein disorders
6. Hyperparathyroidism

*See Table 4 about factors causing rHu-EPO resistance

states (e.g. rheumatoid arthritis), during infections, or with malignant neoplasms. Although aluminum accumulation has decreased as a problem in CRF patients lately because of better purification of dialysate water and less use of aluminum-containing phosphate binders, aluminum overload can cause EPO resistance or total insensitivity. The mechanism may involve an interference with iron metabolism. Following the advent of EPO therapy, iron deficiency has become a major therapeutic challenge. This will be covered later in this chapter. Either vitamin B_{12} or folate deficiency may occasionally contribute to defective erythropoiesis in CRF patients, but both are rare due to dietary counsel and multivitamin supplements.

Hemodialysis is accompanied by a peculiar set of problems, some of which were never seen before dialysis became widespread. Hemolysis may be caused by overheated dialysate (51°C) and by hypotonic dialysate when concentrate is allowed to run out. Contamination of dialysate with chloramines, copper, zinc, formaldehyde or sodium hypochlorite (bleach) may also cause hemolysis. Hemolysis due to shearing of red cells has been seen when dialyzer blood is pumped at high speeds through small needles or cannulas. Treatment with hemodialysis leads to massive whole blood losses but in small increments. It has been estimated that with each hemodialysis 15 – 25 mL of blood is lost into the dialyzer, on to the gauze pads used to control bleeding and in the blood lines despite careful rinsing. This blood loss is further aggravated by blood withdrawn for laboratory tests. When summed over the typical 156 dialyses each patient receives every year, over 3 L of whole blood have been removed. It is no wonder that iron deficiency is rampant in dialysis units.

Of minor importance are the following factors. GI bleeding from gastritis, duodenitis,

peptic ulcers or colonic lesions may be enhanced by heparin used during hemodialysis or by uremic platelet dysfunction. The latter is almost never a problem by itself. Drugs may contribute to hemolysis if they are oxidants or blunt the response to EPO (angiotensin-converting enzyme (ACE) inhibitors). Hypersplenism and hyperparathyroidism rarely contribute to uremic anemia. One final category, which is sometimes forgotten by clinicians, concerns diseases which cause both renal failure and anemia, but not necessarily by the aforementioned mechanisms. Among these are sickle hemoglobinopathies, lead intoxication, systemic lupus erythematosus (SLE), scleroderma, and paraproteinemias such as multiple myeloma. In these diseases, anemia out of proportion to the level of renal dysfunction may be a clue to the correct diagnosis.

Table 3. Therapy of Uremic Anemia in the Pre-EPO Era

1. Correction of nutritional deficits
   a. Iron
   b. Folate - vitamin B_{12}
   c. Protein-calorie
2. Provision of adequate dialysis
3. Androgens
4. Avoidance of total nephrectomy
5. Red cell transfusions
6. Rarely indicated
   a. Splenectomy
   b. Parathyroidectomy

Therapy of Uremic Anemia in the Pre-EPO Era

Maintenance hemodialysis became available in many centers in the U. S. in the late 1960's. Widespread use of hemodialysis followed over the next 20 years. Initially, most dialysis patients were young and had disease limited to the kidneys (glomerulonephritis, polycystic kidney disease (PKD)). Individuals with systemic illnesses, such as diabetes mellitus (DM), were usually not dialyzed. Hemodialysis was employed as a temporizing measure until the patient could receive a renal allograft. A large percentage of maintenance hemodialysis patients were able to be transplanted in the late 1960's and early 1970's. Transplantation corrected uremic anemia and occasionally was accompanied by erythrocytosis.

Anemia was considered to be a minor part of the uremic syndrome in these young patients. Levels of HCT in the 13 – 19% range were tolerated if the patient was receiving all available therapy (short of transfusion) and if the patient could function at that low HCT. Red cell transfusions were withheld because of the risk of sensitization to future allografts and the risk of viral hepatitis. However, red cell transfusions were still widely used out of necessity for very low HCT values, for acute blood loss or for symptoms attributable to anemia which could not be corrected by less toxic therapies. As dialysis practice widened in the 1970's and early 1980's, older and sicker patients were being dialyzed. Very low levels of HCT could not be tolerated by these patients, and transfusion usage increased.

Standard therapy of anemia began with the provision of adequate protein-calorie nutrition, folate, vitamin B_{12}, and iron (Table 3). Both of these vitamins were known to be dialyzable. The usual criteria for iron deficiency in non-uremic subjects (serum iron, total iron binding capacity, and ferritin) were

found to be too stringent when applied to dialysis patients, resulting in a change in the guidelines toward higher values. For example, a serum ferritin of less than 20 ng/mL would be indicative of iron deficiency in a normal subject, whereas a value of 100 ng/mL might be too low in a dialysis patient.

Most patients were hemodialyzed for 4 hours on cellulosic dialyzers using blood flows of 200 – 250 mL/min and dialysate flow of 500 mL/min. This would be substandard in many patients by today's ideas of dialysis adequacy. However, it was typical to see an increase in HCT averaging about 5 points in the first few months following dialysis initiation if the patient was not anephric. Some patients, typically those with PKD, normalized their HCT levels with dialysis, iron and vitamins alone. Those few patients who had to be totally nephrectomized and thus lacked endogenous EPO could not be managed this way and had to be regularly transfused.

Another helpful adjunct to uremic anemia therapy at that time was the use of androgens [14]. Androgens somehow augment the efficacy of EPO. Therefore, androgens do not work in anephric patients unless EPO is supplemented. Although both oral and parenteral forms of synthetic androgens were used in some dialysis units, we found them to be too toxic to the liver. Testosterone enanthate (1 – 4 mg/kg) as a once weekly injection had few side effects and would increase the HCT by up to 5 points in the average patient with native kidneys intact. However, the response was slow and not all patients were helped. In our center in the late 1970's, the mean HCT was 26% in 30 "nephric" patients receiving testosterone and an average of 0.1 units of packed red cells per month (very few patients were transfused). The mean HCT in surgically anephric patients (N = 6) was 20%, and they received an average of 2.1 units of packed red cells per month.

Although rarely indicated and controversial in success rate, both splenectomy and parathyroidectomy have been advocated as helpful in certain patients. In the former, excessive erythrocyte destruction by the spleen should be carefully documented prior to surgery. Reversal of increased marrow fibrosis is cited as the reason for the effect of parathyroidectomy on uremic anemia. We have not seen it in our patient population.

The EPO Era

Introduction

Since deficiency of EPO is the major factor in uremic anemia, provision of EPO to affected patients would be the ideal treatment. After the EPO gene was cloned in 1985, it was transfected into Chinese hamster ovary (CHO) cells enabling the production of EPO in large quantities [10]. The purified protein is immunologically and biologically identical to human urinary EPO. It is prepared for administration to patients in a buffered saline solution containing 0.25% human serum albumin. Vials (1 mL) of 2000, 3000, 4000 and 10,000 U/mL are available.

Clinical Trials

In late 1986 and early 1987 the first studies of the use of recombinant human EPO (rHuEPO) on a large scale (10 dialysis patients in the U. K. and 25 dialysis patients in the U. S.) were published [15, 16]. Almost all patients responded with an increase in HCT, a cessa-

tion of need for blood transfusions, and a general improvement in the sense of well-being and exercise tolerance. In the U. S. study, intravenous doses of 1.5, 5.0, 15, 50, 150, and 500 U/kg body weight of rHu-EPO were given thrice weekly at the end of dialysis. Baseline HCTs were 19 – 22%. At the 1.5 and 5.0 U doses, no changes in reticulocytes or HCT were observed. Beginning at 15 U/kg thrice weekly, there was a dose response affecting both maximum HCT and the rate of rise of HCT. At 15 U/kg thrice weekly a HCT of 24% was achieved in 12 weeks; at 50 U/kg thrice weekly the HCT rose to 40% in 11 weeks; and at 500 U/kg thrice weekly the HCT normalized to 42% in 6 weeks. Seventeen patients wound up receiving 25 – 100 U/kg thrice weekly as maintenance therapy for 3 – 7 months in order to maintain HCTs of 35 – 40%. The adverse effects were minimal, chiefly exacerbation of hypertension in 24% and hyperkalemia. Hyperkalemia occurred because 2 patients felt so much better that they saw no need to continue a renal failure diet.

These phase I – II trials were followed by a phase III study involving 333 U. S. hemodialysis patients beginning in the fall of 1986 [17]. All had a HCT < 30%. rHu-EPO in a dose of 300 U/kg thrice weekly was given intravenously to the initial patients. This dose was lowered to 150 U/kg thrice weekly shortly after the study began. Once a target HCT of 35% was achieved, the rHu-EPO dose was adjusted to a maintenance dose required to keep the HCT at 32 – 38%. The response rate was 97.4%. An elevated reticulocyte count occurred in the first 2 weeks, and an increased HCT was seen within 2 – 6 weeks. Erythrocyte transfusions were eliminated in all patients within 2 months of starting rHu-EPO. The median maintenance rHu-EPO dose was 75 U/kg thrice weekly. Adverse effects included iron deficiency (43%), exacerbation or new appearance of hypertension (35%), seizures (5.4%), and myalgias (5%). The rate of vascular access clotting was no different (0.5 clotting events/patient/year) than in dialysis patients not receiving rHu-EPO. No patient developed antibodies to rHu-EPO. The 9 patients out of 333 who failed rHu-EPO therapy had other reasons for anemia including myelofibrosis, chronic infection, and hemorrhage.

Taking everything into account, the results of this large and expensive undertaking were overwhelmingly positive. Nearly all patients could normalize their HCTs, whether or not they had residual renal tissue. Improvements in the quality of life were remarkable. The drug was not only effective, if iron stores were maintained, but was also safe. The main thing to look out for was the exacerbation of hypertension.

rHu-EPO Pharmacokinetics and Dosing

Similar to other parenteral medications, subcutaneous (SC) dosing of rHu-EPO is followed by lower peaks and higher troughs than with intravenous (IV) administration. Following an IV dose, there is rapid disappearance ($t^{1/2}$ = 6.8 hours), and low rHu-EPO concentrations are seen at 24 – 48 hours [18]. With subcutaneous (sc) administration, therapeutic levels may be seen for 72 – 96 hours. As little as 40 U/kg thrice weekly or 60 U/kg twice weekly of SC rHu-EPO can maintain plasma EPO levels within the probable therapeutic range (30 – 100 mu/mL) (see Figure 2). Thrice weekly intravenous (IV) doses of 40 U/kg result in subtherapeutic EPO concentrations during a significant portion of the time. Therefore, there is a greater economy with the SC route of administration. On the average, 25 – 40% less rHu-EPO is required during SC

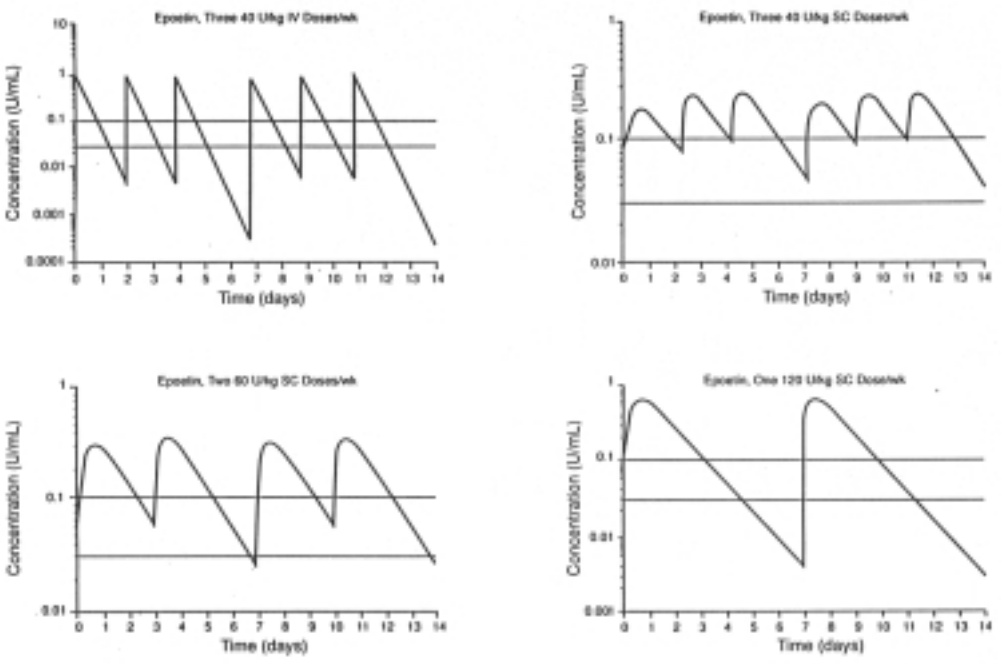

Figure 2. Concentration time simulations for differing strategies at a constant erythropoietin dose of 120 U/kg/week. With permission from Besarab et al. [18].

compared to IV dosing to obtain the same effect. The rate of rise in HCT (points/week) is linearly related to the logarithm of the rHu-EPO dose (U/kg thrice weekly). The steepest part of the curve, and therefore the most efficient incremental response, oc curs between a dose of 40 and 150 U/kg thrice weekly (Figure 3). Above a dose of 150 U/kg thrice weekly the increase in HCT is less steep.

Patients should be replete in iron, folate, and vitamin B_{12} before commencing rHu-EPO therapy. Percent transferrin saturation (TSAT) is the most cost-effective test [19]. The initial recommended dose is 50 – 100 U/kg thrice weekly, either SC (preferably) or IV. We round off to the nearest 1000 U. If after 6 weeks, the patient's HCT has not increased by 5 – 6 points or the desired HCT of approximately 35% has not been achieved, then the dose may be increased in increments of 25%

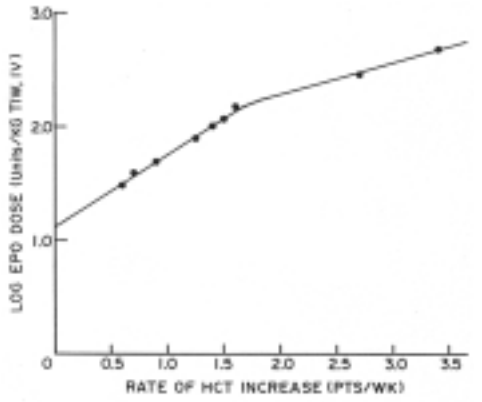

Figure 3. The logarithm of the rHu-EPO dose is plotted against the rate of increase of HCT. Note the change in slope above a dose of 150 U/kg IV 3 times per week (log = 2.18). Data are replotted from reference [25].

of the maintenance dose or 25 U/kg. An interval of 4 weeks should elapse between any dose adjustment thereafter since the erythropoietic response may not maximize for 2 – 6 weeks. If the HCT exceeds 36%, rHu-EPO should not be discontinued since this shuts off erythropoiesis. We prefer to decrease the rHu-EPO dose by 25%. Again, the time to equilibrate with the new lower dose will be 2 – 6 weeks.

Intraperitoneal (IP) administration of rHu-EPO has been tried in chronic PD patients. The response is hampered by the slow peritoneal transport of proteins of this molecular weight (30,500 daltons) into the circulation which may take more than 12 hours. The patient, of course, also needs to be dialyzed. In our opinion IP rHu-EPO should only be used as a last resort after the failure of SC dosing. IP administration should be in a small volume (40 mL) injected into the dry peritoneal cavity followed by a 10 mL saline flush [20]. Overnight undialyzed dwell times approaching 12 hours would lead to maximum absorption and potential TIW dosing in quantities similar to the SC route. When SC rHu-EPO dosing is used in PD or CCPD patients, they require less rHu-EPO on average than hemodialysis patients. Although the cause of this difference is unclear, potential reasons include less iron deficiency, blood losses, and laboratory testing than in hemodialysis patients.

rHu-EPO Resistance

Some patients require large doses of rHu-EPO (≥ 300 U/kg thrice weekly) to restore erythropoiesis, or do not respond at all. Table 4 lists most of the common reasons for this resistance [21]. However, it is not always possible to find the cause in an individual refractory patient. A careful search for iron deficiency in either an initially refractory patient or in one who becomes unresponsive during maintenance rHu-EPO therapy is a good starting point.

Table 4. Causes of rHu-EPO Resistance

1. Iron deficiency
2. Inflammatory states a. Infections b. Rheumatoid arthritis and other connective tissue diseases
3. Inadequate dialysis
4. Deficiency of folate/vitamin B_{12}
5. Aluminum toxicity
6. Marrow fibrosis due to hyperparathyroidism
7. Hemolysis
8. Hemorrhage
9. Hemoglobinopathy
10. Anemia of chronic disease
11.Drug interactions and toxicities

There is no perfect test of iron stores in a patient with CRF. A serum ferritin concentration < 50 ng/mL almost always reflects iron depletion, and a value of < 100 ng/mL may indicate deficiency. Ferritin is an acute phase reactant and may be elevated in the face of low iron stores if there is concomitant infection, inflammation, liver disease or neoplasia. The per cent transferrin saturation (TSAT) may be a useful adjunct to the ferritin. A value < 20% usually indicates iron depletion. In 52 patients with a HCT less than 25% during rHu-EPO therapy, 60% had a serum ferritin < 100 ng/mL, a TSAT < 20% or both [22]. Many of these patients had been prescribed oral iron tablets. Others use a TSAT cutoff of < 25% to define an iron-responsive group of anemic

dialysis patients [23] (See Figure 4). Recent data from Wave 1 of the Dialysis Morbidity and Mortality Study (USRDS 1996) suggest that despite use of rHu-EPO in over 80 – 90% of dialysis patients, 48% had a HCT < 30% [54]. A total of 54% had TSAT < 20%; almost 25% had TSAT < 10%; and 36% had serum ferritin < 100 ng/mL. These data suggest that iron deficiency partly explains the failure to reach the target HCT of > 30%.

Most hemodialysis patients and some PD patients will require oral iron to maximize the response to rHu-EPO (see Table 5 for a list of oral iron preparations). Table 6 lists helpful hints to assure compliance with oral iron therapy. Patients must be educated that iron deficiency is the most common cause of a suboptimal response to oral iron therapy. Attempts to give iron on an empty stomach have generally failed in our hands due to GI side effects. Medications that decrease gastric acidity (e.g. cimetidine), phytates (green, leafy vegetables) and tannates (tea, coffee, red wine) may also diminish iron absorption.

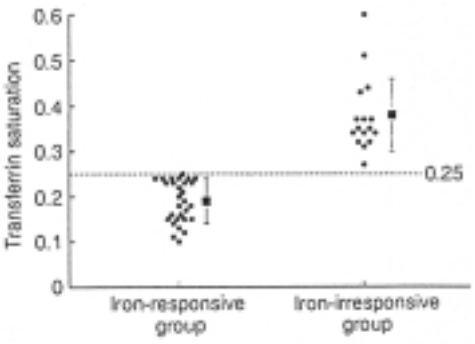

Figure 4. Serum transferrin saturation cut-off value between iron-responsive and iron-unresponsive anemic dialysis patients. With permission from Tarng et al. [23]

Some patients will be noncompliant with oral iron (suggested dose: ≥ 200 mg of elemental iron/day), and this will blunt their responsiveness to rHu-EPO. Other patients will be unable to replete iron stores (e. g. a serum ferritin > 100 ng/mL) on oral therapy [25]. Parenteral iron dextran is a key option in these groups. It is formulated in 2 mL ampules containing 50 mg of iron/mL. We give 100 mg into the venous line over 5 minutes at the end of dialysis. Ten doses are given over 10 dialyses for a total of 1000 mg per course. A course may be repeated if indications of iron deficiency persist. For home hemodialysis (HD)

Table 5. Oral Iron Preparations

Preparation	Trade Name	Elemental Iron/Tablet	Daily Dose (To supply 200 mg elemental iron) Total
Ferrous sulfate	Feosol	65 mg	TID (195 mg)
Ferrous fumarate	Tabron	100 mg	BID (200 mg)
Ferrous fumarate	Chromagen	66 mg	TID (198 mg)
Iron-polysaccharide complex	Niferex-150	150 mg	one tab Nif-150
	Niferex W/C	50 mg	Plus one tab Nif. W/C=200 mg
	Niferex elixir	100 mg/5 mL	5 mL BID=200 mg

Table 6. Factors Aiding Compliance with Oral Iron. Preparations and Maximizing Intake of Iron

1. Patient education
2. Increased dosage per day
   a. Smaller, more frequent doses
   b. Bedtime doses
3. Administration by dialysis nurses of at least 2 doses per hemodialysis
4. Alternative iron formulations for variety and less GI side effects
5. Pharmacy monitoring of refills
6. High iron food
7. Separation of the ingestion of phosphate binders from the time of oral iron dosing; e. g. iron before meals and binders after meals

and PD patients with absolute iron deficiency, 1g of iron dextran (in 500 mL saline) or 500 mg (in 250 mL saline) can be administered in the home training unit after a test dose of 25 mg iron. A test dose of 0.5 mL (25 mg of iron) is given before each course followed by observation to avert anaphylactoid reactions or anaphylaxis. However, even test doses can cause anaphylactoid reactions in 1% of patients and result in 40% of all anaphylactoid reactions [26]. The incidence of anaphylaxis is about 1 in 1000 doses, so dialysis units must be prepared to handle such an emergency. Adverse reactions occur in about 5% of patients, but most are not serious [26]. An acute large joint arthritis can be caused by the iron dextran itself. Itching and wheezing are the most common adverse events.

Chronic low doses (25 – 200 mg/week) of parenteral iron dextran have been given routinely to hemodialysis patients in the U. S. and Europe as an alternative to oral iron therapy [27]. Benefits have included an increase in mean HCT, a decrease in rHu-EPO dose by 33 – 46%, and an overall cost savings. The potential risk of iron overload when such doses are given over a long period of time has not been assessed. Another factor in need of study is the use of supplemental androgen therapy to decrease the rHu-EPO dose and resultant expense.

rHu-EPO resistance may also be caused by undiagnosed or untreated infections. Our first patient with rHu-EPO resistance had a staphylococcal diskitis and vertebral osteomyelitis. His neck pain was attributed to osteoarthritis, which was also present, and there was no fever or leukocytosis. rHu-EPO resistance led to a search for a reason and to the correct diagnosis. Patients with active non-infectious inflammatory states such as rheumatoid arthritis may require higher doses of rHu-EPO. Other reasons for rHu-EPO resistance are summarized in Table 4. Folate deficiency may develop in HD patients who either restrict their protein intake (because dialysis loss of folate exceeds dietary intake), or who require phenytoin therapy. The presence of red cell macrocytosis suggests folate deficiency, if iron overload is excluded. Oral folic acid therapy corrects or prevents this complication. However, most dialysis patients ingest enough dietary folate to remain in positive folate balance.

Benefits of rHu-EPO

Quality of Life

Almost everyone involved in the initial rHu-EPO studies has an anecdote to tell about this subjective variable. One of my patients, a 42 year old farmer, was forced to sell his hogs and cattle when he went on dialysis because of poor exercise tolerance. He required 4 units of packed red cells per month to maintain a HCT of 20 – 22%. Within a few months of his participating in the Phase III rHu-EPO trial,

he had a HCT of 35 – 38% and needed no transfusions. He was able to go back to full-time farming, a rigorous occupation.

Beusterien and colleagues studied 484 dialysis patients who had not been previously treated with rHu-EPO [28]. All patients were assessed by 6 scales taken from the Medical Outcomes Study 36-Item Health Survey at baseline and 49 – 180 days later. Despite a rise in HCT from only 25.5 to 29.9% at follow-up, significant improvements occurred in physical and social functioning, vitality, health status, mental health, and mental component summary score. Activity items showing better scores were looking after the home, social life, interests/hobbies, and sexual satisfaction. The amount of change in HCT was a significant predictor of quality-of-life improvements. Others have reported increases in energy and activity levels, functional ability, appetite, taste for food, cold tolerance, and sexual function. Sleep patterns have also improved in patients receiving rHu-EPO with less insomnia at night and fewer naps during the day.

Cardiorespiratory Benefits

MacDougall et al. studied 10 hemodialysis patients by maximum exercise testing, pulmonary function tests, echocardiography, chest roentgenography, and rheological assessment over 12 months as they initiated rHu-EPO therapy [29]. After 2 months of rHu-EPO, significant increases in exercise time, maximum oxygen consumption, and anaerobic threshold were seen. There was a substantial decrease in exercise-induced cardiac ischemia and left ventricular mass (by echocardiography and chest roentgenogram). Other hemodynamic changes seen in rHu-EPO-treated patients are increased venous tone, peripheral vascular resistance (PVR), blood viscosity and tissue oxygenation [25]. Decreased left ventricular end-diastolic diameter and left atrial diameter have accompanied the diminished left ventricular mass. Overall, there is improved ventricular performance due to increased oxygen delivery. Hemodialysis-related hypotension is less of a problem in rHu-EPO-treated patients.

Reduction in Transfusions

Prior to rHu-EPO therapy, approximately 50% of hemodialysis patients were transfused each year [30]. A mean of 10 units was given annually to each transfused patient. The risks of this practice include hypersensitivity reactions, viral hepatitis, human immunodeficiency virus (HIV), sensitization to HLA antigens, iron overload, and a variety of less common complications. Following a response to rHu-EPO therapy, the need for transfusions is nearly abolished. In the U. S. Phase III trial, 333 patients had received 1030 units of red cells in the 6 months prior to starting rHu-EPO therapy [17]. During the next year, only rare patients were transfused for blood losses at surgery or for medical complications. By avoiding blood transfusions, the rHu-EPO-treated dialysis patient is a better candidate to receive a renal transplant because of less viral hepatitis and lower amounts of panel-reactive HLA antibodies.

Enhanced Cerebral Function

Poor cognitive function is a part of the uremic syndrome and is helped by the initiation of an adequate maintenance dialysis regimen. rHu-EPO therapy has shown measurable benefits even above adequate dialysis. Grimm et al. found objective improvement in several tests of brain function following rHu-EPO therapy [31]. As the HCT increased from 22.7

to 30.6%, auditory event-related potentials and multimodality stimulus-related evoked potentials improved. Marsh and colleagues also found improvement in event related potentials and 4 neuropsychological tests following 3 – 12 months of rHu-EPO therapy [32]. During this investigation, HCT values increased from 23.7 to 36.5%. Thus, improvement in anemia was directly linked to better cerebral function.

Miscellaneous

Although standard blood coagulation tests such as prothrombin time (PT), partial thromboplastin time (PTT), and fibrinogen concentration are unchanged following treatment with rHu-EPO, mild increases in platelet count and platelet aggregation as well as decreased bleeding time have been observed [25]. The higher HCT may force more platelets toward the periphery of blood vessels and promote hemostasis in response to injury.

Immune function has benefited from rHu-EPO therapy in 3 areas: phagocytosis/chemotaxis, antibody responses to vaccines, and cell-mediated immunity [25]. Some of these changes may be related to a better nutritional status.

Adverse Effects of rHu-EPO

Hypertension

During the first 3 months of the U. S. Phase III trial of rHu-EPO, 35% of patients developed an increase in diastolic blood pressure of ≥ 10 mm Hg or needed increased doses of antihypertensive medications [17]. Subsequent multicenter studies in Europe, West Germany, and Japan have demonstrated a similar incidence. This phenomenon has not been seen in normal subjects, HIV positive patients, patients with advanced multiple myeloma, or rheumatoid arthritics with normal renal function treated with rHu-EPO. Risk factors for the development of rHu-EPO-induced hypertension besides CRF are high rHu-EPO dosage, IV administration, rapid correction of anemia and severe anemia before rHu-EPO therapy (HCT < 20%). Postulated mechanisms include increased blood viscosity, loss of hypoxic vasodilatation, heightened endothelin production with imbalance of local endothelial factors – endothelium derived relaxing factor (EDRF) and endothelin 1 –, and increased free calcium in vascular smooth muscle cells. In practical terms, the development of hypertension post-rHu-EPO can be prevented or ameliorated by achieving good control of blood pressure prior to beginning therapy via medication and by maintaining low interdialytic weight gains. Blood pressure should be monitored not only during dialysis but also on non-dialysis days. Adjustments in fluid weight and dosages of antihypertensive drugs are almost always able to prevent serious hypertension in rHu-EPO-treated patients. This is especially important in the first two months of therapy. With this approach, there is no difference in severe hypertension (diastolic pressure > 110 mm Hg) in rHu-EPO-treated vs. control subjects [7].

Iron Deficiency

Careful monitoring of iron stores in all patients receiving rHu-EPO is mandatory. During the Phase III rHu-EPO trial, 43% of patients became iron deficient despite oral iron [17]. Serum ferritin and TSAT values should be obtained monthly during the 3 month initiation phase and then at least quarterly thereafter. If the patient is receiving parenteral iron,

the tests should be drawn 2 – 4 weeks after the last dose of iron dextran since false elevations are seen before that time.

Seizures

Seizures were seen in the Phase I – II rHu-EPO trials and were attributable to hypertensive encephalopathy in nearly all cases [15, 16]. Patients had a prodrome of headache and visual blurring. Unlike hypertensive encephalopathy, papilledema is often absent. In Phase III of the U.S. multicenter study, seizures were reported in 5.4% of patients receiving rHu-EPO. This was not significantly different from the incidence (4 – 6%) in the control dialysis-dependent population [17]. Although there is no evidence suggesting rHu-EPO is epileptogenic, uncontrolled hypertension resulting in changes in cerebral perfusion is undoubtedly a major factor. Good blood pressure control as in the Canadian Multi-centre Trial has resulted in no excess appearance of seizures in the rHu-EPO-treated group versus controls. Therefore, seizures can be avoided during rHu-EPO therapy by careful attention to the maintenance of normal blood pressures by medication and avoidance of hypervolemia.

Vascular Access Clotting

Hemodialysis patients regularly clot their vascular accesses, especially if they are synthetic – polytetrafluoroethylene graft (PTFE). Native arteriovenous fistulas are less at risk. Although a control group was not included in the U.S. Phase III trial, the incidence of access thrombosis seen (0.5 events per patient year) was similar to non-treated patients [17]. Subsequent investigations have shown a higher incidence (up to 3-fold) of PTFE fistula clotting in rHu-EPO-treated versus control patients. This is not a reason to withhold rHu-EPO therapy. Patients who appear to have this problem can be managed with careful attention to vascular access performance (venous pressure, recirculation) and intervention by radiologists or surgeons prior to complete thrombosis. Antiplatelet drugs may be a helpful adjunct. Sreedhara et al. found dipyridamole to be beneficial in patients with new PTFE grafts [33]. However, aspirin did not improve the risk of thrombosis in these grafts. Neither dipyridamole nor aspirin had any beneficial effects in patients with prior thrombosis of PTFE grafts. Dialyzer clotting has been successfully managed by modest increases in heparin dosages.

Less Efficient Hemodialysis

This was a theoretical problem because higher HCT values would result in a smaller plasma volumes. Most studies saw either small or no changes in BUN, serum creatinine, serum potassium, and serum phosphate following initiation of rHu-EPO therapy. The few instances of severe hyperkalemia were more related to dietary indiscretions than to inefficient hemodialysis. In this era of frequent monitoring of dialysis adequacy by urea kinetics, any loss of dialysis efficiency due to rHu-EPO will be measurable and can be compensated for by alterations in dialysis parameters, such as blood flow and type of dialyzer employed.

Summary

Monitoring for adverse effects of rHu-EPO should be done on a routine basis in all treated patients. Attention to achieving normal blood pressures and adequate iron stores is of para-

mount importance. Fistulas need to be monitored for signs of impending thrombosis such as increased percent recirculation and higher venous pressures (VDP). Decreased hemodialysis access flow as measured by duplex ultrasonography has been shown to be a significant predictor of future HD access thrombosis and may be a more promising technique compared to VDP monitoring. Dialysis adequacy (urea reduction ratio > 70% or *Kt/V* > 1.4) should be measured on a regular basis, and the dialysis prescription adjusted accordingly. Fortunately, antibodies to rHu-EPO continue to be an extremely rare occurrence.

Table 7. Indications for rHu-EPO in Pre-dialysis Patients with Anemia

1. Left ventricular hypertrophy
2. Coronary artery disease
3. Upcoming elective major surgery
4. HIV nephropathy
5. Students
6. Job holders and at-home mothers with small children
7. Transfusion dependence
8. Living-related transplant candidates

rHu-EPO in the Pre-dialysis Patient

Pre-dialysis patients (Ccr of 10 – 30 mL/min) with HCT values < 30% are candidates for rHu-EPO therapy. Similar guidelines are followed as in dialysis patients. Blood pressure must be normalized and iron stores replenished prior to starting rHu-EPO therapy and monitored during treatment. Initial studies using 50, 100, or 150 U/kg IV thrice weekly have shown good response rates and few adverse events [34]. Target HCTs were 33 – 36%. Progression of renal insufficiency was not increased by receiving rHu-EPO, in the setting of blood pressure control and avoidance of rapid increases in Hct (> 4% increase in 4 weeks). Subsequently, others have demonstrated efficacious use of SC rHu-EPO in doses of 75 – 150 U/kg thrice weekly or even 75 – 150 U/kg once weekly [35]. Patients in whom predialysis rHu-EPO is particularly recommended are listed in Table 7.

A beneficial aspect of predialysis rHu-EPO is its potential for reducing left ventricular hypertrophy (LVH). The adverse effects of LVH are well documented in the dialysis population including the relative risk of death that it confers [36]. Furthermore, once established, LVH rarely regresses in dialysis patients. Levin and colleagues recently noted that the prevalence of LVH in a predialysis population was 38.9%, and this percentage increased with progressive renal failure [37]. LVH was present in 26.7%, 30.8%, and 45.2% of patients with Ccr > 50 mL/min, 25 – 49 mL/min, and < 25 mL/min, respectively. Logistic regression analysis revealed that age, Ccr, hemoglobin, and systolic blood pressure were significantly different between those patients with and without LVH. The last two, as modifiable risk factors, could potentially be controlled to lessen the consequences of LVH in the dialysis population.

rHu-EPO and Renal Transplantation

A successful renal transplant results in a peak serum EPO level of 100 mu/mL or more within the first week after grafting even if the

patient is still oliguric [38]. This is followed by a falling serum EPO concentration as graft excretory function improves. A second EPO peak is seen from day 20 to day 50. HCT values return to normal range in the first 1 – 2 months, and serum EPO levels are normal by 2 months postgrafting. About 10 – 15% of renal transplant recipients develop erythrocytosis (HCT > 51%), usually within the first 2 years [39]. Factors other than EPO may be involved, and the condition rarely spontaneously resolves. Therapy consists of phlebotomy, ACE inhibitors, and consideration for native nephrectomy.

It is controversial as to whether and when rHu-EPO therapy should cease in a dialysis patient who receives a renal allograft. Because endogenous EPO production picks up rapidly in non-rejecting renal transplants, prudence would dictate that rHu-EPO therapy be stopped either at the time of grafting or within the first 2 weeks thereafter. Most episodes of acute rejection are brief enough not to require reinstitution of rHu-EPO. However, in the chronically failing allograft, anemia will recur in a similar manner as in native kidney failure. If patients with chronic renal insufficiency from graft disease or rejection become anemic, rHu-EPO therapy should be resumed following guidelines similar to those for predialysis patients.

Conclusions

The past 10 years have seen a revolutionary treatment of the anemia of CRF evolve and gain broad acceptance. rHu-EPO therapy has been remarkably effective in correcting anemia and returning most patients to a better state of health. With careful management few adverse events are observed. The overall benefits to patients are almost as great as those of maintenance dialysis itself. While recognizing this remarkable progress, it is not sufficient to stop there.

As of late 1995 in the U.S., 90% of hemodialysis patients and 60% of peritoneal dialysis patients were receiving rHu-EPO therapy. The mean HCT was 31.4%, and the median HCT was 32% [40]. At a minimum cost of $10 per 1000 units and a mean dose of 5000 units thrice weekly, $7800 would be spent per patient per year on just rHu-EPO. It is not unreasonable to assume that 200,000 CRF patients in the U.S. are now being treated with rHu-EPO for an overall annual cost of $ 1.56 billion. These figures only consider the expense of rHu-EPO itself. The development of a cheaper method of treating the anemia of CRF is of paramount importance. Areas of clinical research which may help alleviate the problem somewhat include determining the optimum HCT range with the lowest risk to benefit ratio, finding the best method of achieving and maintaining normal iron stores in the presence of rHu-EPO therapy, and the optimization of rHu-EPO-sparing adjuncts such as androgens. Patient and physician education programs still have a long way to go. SC administration of rHu-EPO is not being used enough. In 1993, 92.5% of rHu-EPO doses were given IV and only 7.5% were SC [24]. Since SC rHu-EPO is about 40% more effective, switching IV to SC dosing could result in a large cost saving. Finally, new laboratory investigations may provide cheaper, small molecular weight peptide mimetics of rHu-EPO [41]. Such molecules have been discovered and may possibly be developed into pharmaceuticals. Competition may also lower the price of the established drug.

References

[1] *Erslev AJ* 1983 Anemia of chronic renal failure. In: Williams WJ, Beutler E, Erslev AJ, Lichtman MA (eds): Hematology. McGraw-Hill, New York, pp 417-425

[2] *Howard AD, Moore J, Welch PG, Gouge SF* 1989 Analysis of the quantitative relationship between anemia and chronic renal failure. Am J Med Sci *297:* 309-313

[3] *Hakim RM, Lazarus JM* 1988 Biochemical parameters in chronic renal failure. Am J Kidney Dis *11:* 238-247

[4] *Reissman KR* 1950 Studies on the mechanism of erythropoietic stimulation in parabiotic rats during hypoxia. Blood *5:* 372-380

[5] *Erslev AJ* 1953 Humoral regulation of red cell production. Blood *8:* 349-357

[6] *Jacobson LO, Goldwasser LF, Fried W, Plzak L* 1957 Role of the kidney in erythropoiesis. Nature *179:* 633-634

[7] *Erslev AJ, Besarab A* 1997 Erythropoietin in the pathogenesis and treatment of the anemia of chronic renal failure. Kidney Int *51:* 622-630

[8] *Miyake T, Kung CKH, Goldwasser E* 1977 Purification of human erythropoietin. J Biol Chem *252:* 5558-5564

[9] *Jacobs K, Shoemaker C, Rudersdorf R, Neill SD, Kaufman RJ, Mufson A, Seehra J, Jones SS, Hewick R, Fritsch EF, Kawakita M, Shimizu T, Miyake T* 1985 Isolation and characterization of genomic and cDNA clones of human erythropoietin. Nature *313:* 806-810

[10] *Lin FK, Suggs S, Lin CH, Browne JK, Smalling R, Egrie JC, Chen KK, Fox GM, Martin F, Stabinsky Z, Badrawi SM, Lai PH, Goldwasser E* 1985 Cloning and expression of the human erythropoietin gene. Proc Natl Acad Sci *82:* 7580-7584

[11] *Koury ST, Koury MJ, Bondurant MC, Caro J, Graber SE* 1989 Quantitation of erythropoietin-producing cells in kidneys of mice by in situ hybridization: correlation with hematocrit, renal erythropoietin mRNA, and serum erythropoietin concentration. Blood *74:* 645-651

[12] *Eschbach JW, Adamson JW* 1985 Anemia of end-stage renal disease (ESRD). Kidney Int *28:* 1-5

[13] *Besarab A, Girone JF, Erslev A, Caro J* 1989 Recent developments in the anemia of chronic renal failure. Sem Dial *2:* 87-97

[14] *Hendler ED, Goffinet JA, Ross S, Longnecker RE, Bakovic V* 1974 Controlled study of androgen therapy in anemia of patients on maintenance hemodialysis. N Engl J Med *291:* 1046-1051

[15] *Winearls CG, Oliver DO, Pippard MJ, Reid C, Downing MR, Cotes PM* 1986 Effect of human erythropoietin derived from recombinant DNA on the anaemia of patients maintained by chronic haemodialysis. Lancet *2:* 1175-1178

[16] *Eschbach JW, Egrie JC, Downing MR, Browne JK, Adamson JW* 1987 Correction of the anemia of end-stage renal disease with recombinant human erythropoietin. N Engl J Med *316:* 73-78

[17] *Eschbach JW, Abdulhadi MH, Browne JK, Delano BG, Downing MR, Egrie JC, Evans RW, Friedman EA, Graber SE, Haley NR, Korbet S, Krantz SB, Lundin AP, Nissenson AR, Ogden DA, Paganini EP, Rader B, Rutsky EA, Stivelman J, Stone WJ, Teschan P, Van Stone JC, Van Wyck DB, Zuckerman K, Adamson JW* 1989 Recombinant human erythropoietin in anemic patients with end-stage renal disease. Results of a phase III multicenter clinical trial. Ann Intern Med *111:* 992-1000

[18] *Besarab A, Flaharty KK, Erslev AJ, McCrea JB, Vlasses PH, Medina F, Caro J, Morris E* 1992 Clinical pharmacology and economics of human erythropoietin in end-stage renal disease: the case for subcutaneous administration. J Am Soc Nephrol *2:* 1405-1416

[19] *Hutchison FN, Jones WJ* 1997 A cost-effectiveness analysis of anemia screening before erythropoietin in patients with end-stage renal disease. Am J Kidney Dis *29:* 651-657

[20] *Bargman JM, Jones JE, Petro JM* 1992 The pharmacokinetics of intraperitoneal erythropoietin administered undiluted or diluted in dialysate. Perit Dial Int *12:* 369-372

[21] *Muirhead N, Bargman J, Burgess E, Jindal KK, Levin A, Nolin L, Parfrey P* 1995 Evidence-based recommendations for the clinical use of recombinant human erythropoietin. Am J Kidney Dis *26:* (suppl 1) S1-S24

[22] *Eschbach J, Fishbane S, Wish JB* 1997 Re-evaluating iron management in renal failure. Triclinica, New York

[23] *Tarng DC, Chen TW, Huang TP* 1995 Iron metabolism indices for early prediction of the response and resistance to erythropoietin therapy in maintenance hemodialysis patients. Am J Nephrol *15:* 230-237

[24] *United States Renal Data System* 1996 Annual Data Report. The USRDS dialysis morbidity and mortality study (wave 1). Am J Kidney Dis *28:* (suppl 2) S58-S78

[25] *Thompson JR* 1994 Treatment of anemia in dialysis subjects. In: Henrich WL (ed.): Principles and Practice of Dialysis. Williams and Wilkins, Baltimore, pp 297-327.

[26] *Fishbane S, Ungureanu VD, Maesaka JK, Kaupke CJ, Lim V, Wish J* 1996 The safety of intravenous iron dextron in hemodialysis patients. Am J Kidney Dis *28:* 529-534

[27] *Fishbane S, Frei GL, Maesaka J* 1995 Reduction in recombinant human erythropoietin doses by the use of chronic intravenous iron supplementation. Am J Kidney Dis *26:* 41-46

[28] *Beusterien KM, Nissenson AR, Port FK, Kelly M, Steinwald B, Ware JE* 1996 The effects of recombinant human erythropoietin on functional health and well-being in chronic dialysis patients. J Am Soc Nephrol *7:* 763-773

[29] *MacDougal IC, Lewis NP, Saunders MJ, Cochlin DL, Davies ME, Hutton RD, Fox KAA, Coles GA, Williams JD* 1990 Long-term cardiorespiratory effects of amelioration of renal anaemia by erythropoietin. Lancet *335:* 489-493

[30] *Crowley JP, Nealey TA, Metzger J, Pono L, Chazan JA* 1987 Transfusion and long-term hemodialysis. Arch Intern Med *147:* 1925-1928

[31] *Grimm G. Stockenhuber F, Schneeweiss B, Madl C, Zeitlhofer J, Schneider B* 1990 Improvement of brain function in hemodialysis patients treated with erythropoietin. Kidney Int *38:* 480-486

[32] *Marsh JT, Brown WS, Wolcott D, Carr CR, Harper R, Schweitzer SV, Nissenson AR* 1991 rHuEPO treatment improves brain and cognitive function of anemic dialysis patients. Kidney Int *39:* 155-163

[33] *Sreedhara R, Himmelfarb J, Lazarus JM, Hakim RM* 1994 Anti-platelet therapy in graft thrombosis: Results of a prospective, randomized, double blind study. Kidney Int *45:* 1477-1483

[34] *Stone WJ, Graber SE, Krantz SB, Dessypris EN, O'Neil VL, Olsen NJ, Pincus TP* 1988 Treatment of the anemia of predialysis patients with recombinant human erythropoietin: a randomized placebo-controlled trial. Am J Med Sci *296:* 171-179

[35] *Zappacosta AR, Perras ST, Bell A* 1991 Weekly subcutaneous recombinant human erythropoietin corrects anemia of progressive renal failure. Am J Med *91:* 229-232

[36] *Silberberg JS, Barre PE, Prichard SS, Sniderman AD* 1989 Impact of left ventricular hypertrophy on survival in end-stage renal disease. Kidney Int *36:* 286-290

[37] *Levin A, Singer J, Thompson CR, Ross H, Lewis M* 1996 Prevalent left ventricular hypertrophy in the predialysis population: identifying opportunities for intervention. Am J Kidney Dis *27:* 347-354

[38] *Ward HJ* 1990 Implications of recombinant erythropoietin therapy for renal transplantation. Am J Nephrol *10:* (suppl 2) 44-52

[39] *Gaston RS, Julian BA, Curtis JJ* 1994 Posttransplant erythrocytosis: an enigma revisited. Am J Kidney Dis *24:* 1-11

[40] *United States Renal Data System;* 1997 Annual Data Report. Treatment modalities for ESRD patients. Am J Kidney Dis *30:* (suppl 1) S54-S66

[41] *Wrighton NC, Farrell FX, Chang R, Kashyap AK, Barbone FP, Mulcahy LS, Johnson DL, Barrett RW, Jolliffe LK, Dower WJ* 1996 Small peptides as potent mimetics of the protein hormone erythropoietin. Science *273:* 458-463