Pharmacokinetics of recombinant human insulin-like growth factor-1 in dialysis patients

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Pharmacokinetics of recombinant human insulin-like growth factor-1 in dialysis patients. Six maintenance hemodialysis (MHD), six continuous ambulatory peritoneal dialysis (CAPD) and six normal adults underwent pharmacokinetic studies of insulin-like growth factor-1 (IGF-1). Each subject received two separate subcutaneous injections of recombinant human IGF-1 (rhIGF-1) (50 or 100 μg/kg) in random order separated by 7 to 21 days. Two different responses were observed. With the 50 μg/kg dose, serum IGF-1 levels and the pharmacokinetic parameters were not different between the three groups. With the 100 μg/kg dose, peak serum IGF-1 concentrations were significantly greater in the MHD and CAPD patients than in normals. However, by 12 to 14 hours after injection, serum IGF-1 was not different in the three groups. Although the T_max area under the curve and serum clearance of IGF-1 were similar in the three groups, the half-life and volume of distribution of rhIGF-1 was significantly decreased in both MHD and CAPD patients. These data indicate that IGF-1 pharmacokinetics are abnormal in maintenance dialysis patients.

Insulin-like growth factor-1 (IGF-1) is a compound with broad anabolic and antihyperglycemic actions that has a molecular weight of 7650 kD [1, 2]. Synthesis of IGF-1, which occurs in tissues throughout the body, is stimulated by growth hormone and is suppressed by poor nutrition or severe catabolic stress. With the development of recombinant DNA biosynthesis technology, recombinant human IGF-1 (rhIGF-1) has been administered experimentally to humans with a variety of clinical conditions as a potential therapeutic agent [3]. Malnourished patients with chronic renal failure have been given rhIGF-1 to improve their protein balance [4]. Although the treatment of chronic renal failure patients with rhIGF-1 will probably increase, there are virtually no data on the serum concentrations or clearance rates of IGF-1 in such patients during treatment. Because IGF-1 is present in normal urine [5], the healthy kidney degrades many peptides [6] and there appear to be abnormally high concentrations of serum IGF binding proteins in renal failure [7, 8]. Hence, it is possible that the pharmacokinetics of rhIGF-1 are altered in renal failure.

We therefore carried out a pharmacokinetic study of the response to two different doses of rhIGF-1 in patients with chronic renal failure. Patients undergoing either maintenance hemodialysis (MHD) or continuous ambulatory peritoneal dialysis (CAPD) were studied. Normal adults of similar age were evaluated for comparison. The results of these studies indicate that the pharmacokinetics of rhIGF-1 are altered in renal failure.

Methods

Patients

Studies were carried out in six patients receiving MHD, six patients undergoing CAPD and six normal adults. Exclusion criteria included pregnancy, insulin and noninsulin dependent diabetes mellitus, infection, vasculitis, autoimmune diseases, presence of malignancy, obesity or malnutrition (relative body weight >120% or <80%), edema, proteinuria greater than 3 g per day, alcoholism, other recreational drug use, and heart, lung or liver failure. Subjects were not included if they received any catabolic or cytotoxic medications during the preceding six months.

Characteristics of the patients are shown in Table 1. Except for the serum urea and creatinine concentrations, there were no differences between the three groups for any parameter in this table. Normal volunteers had a serum creatinine in the normal range and no proteinuria on dipstick examination of the urine. Patients received hemodialysis for 3.5 to 4.5 hours thrice weekly with high-flux polysulfone membrane dialyzers and bicarbonate containing dialysate. CAPD patients underwent four to five exchanges daily with 1.5 to 2.0 liter of dialysate per exchange that contained 1.5 to 4.25% dextrose and 7.0 mg/dl of calcium. MHD patients had undergone hemodialysis for 46.6 ± 17.6 (SEM) months prior to the study. CAPD patients received peritoneal dialysis for 12.8 ± 4.0 months before the study. During the course of this protocol, one hemodialysis patient transferred to CAPD therapy because of problems with his vascular access. He was restudied after four months of treatment with CAPD. This protocol was approved by the Harbor-UCLA Medical Center Human Subjects Committee. Informed written consent was obtained from all subjects.

Protocol design

All subjects were admitted to the Clinical Research Center (CRC) at Harbor-UCLA Medical Center on two occasions separated by 7 to 21 days to receive either 50 or 100 μg/kg body weight of rhIGF-1. Each individual received a different dose of rhIGF-1 on each CRC admission; the order of administration of the two doses was determined randomly. MHD patients entered the CRC immediately after a routine hemodialysis treatment and did not receive another dialysis treatment until after the 24-hour blood
RhIGF-1 was obtained in vials containing 9.8 mg of pure dry recombinant human IGF-1 and 0.3 mg of sulfoxide (a gift from Ciba-Geigy Corp., courtesy of Dr. H.P. Guler), and stored at 4°C. For preparation of the rhIGF-1 for administration, 0.98 ml normal saline was slowly injected into the vial. The vial was then gently rotated along its long axis, to avoid bubble formation, for about 15 seconds. The rhIGF-1 solution was then rapidly transferred into an insulin syringe to be injected subcutaneously. The maximum time between dilution and injection of rhIGF-1 did not exceed 20 minutes.

Assays

Serum total IGF-1 concentration was measured in duplicate by radioimmunoassay (RIA) after acid-ethanol extraction according to Daughaday, Mariz and Blethen [10], except that the specimen were centrifuged for 20 minutes at 4°C after acid-ethanol extraction. A polyclonal rabbit antibody from the NIH (courtesy of Dr. Underwood) was used. Intra- and interassay variabilities were, respectively, 7% and 14%. A comparison in our laboratory of the methods of acid-ethanol extraction with the cryoprecipitation technique [11] indicated that the two methods gave similar results, although the acid-ethanol extraction technique yielded slightly higher values (by 13.8%). Yves LeBouc, M.D. (Hôpital Armand-Trousseau, Paris, France) remeasured IGF-1 in a number of serum specimens from the normal subjects and hemodialysis patients at four different time points using the acid chromatography technique [12]. The results of the two sets of measurements were quantitatively similar. There was also a good correlation between the two sets of serum IGF-1 measurements (N = 39, r = 0.72, P < 0.001).

Pharmacokinetic analyses

For the kinetic analyses, it was assumed that the injected dose was completely absorbed. \( C_{\text{max}} \) is the observed maximal serum concentration of IGF-1, and \( T_{\text{max}} \) is the time from injection until the appearance of \( C_{\text{max}} \) for each individual subject. The serum IGF-1 concentrations were fitted to a one-compartment open model using a weighted, non-linear least-squares regression computer analysis program (ADAPT; Drs. David D’Argenio and Alan Schumitzky, Laboratory of Applied Pharmacokinetics, University of Southern California) [13]. The pharmacokinetic parameters \( K_a \) (absorption rate constant), \( V_d \) (apparent volume of distribution), and \( K_e \) (elimination rate constant) were thus calculated. The assay error pattern was linear, and weights were given to the data points corresponding to the inverse variance of the assay weighting. We examined a two-compartment model and found no improvement in fit; therefore, we used the simpler one-compartment model. In order to calculate the pharmacokinetic parameters, the baseline value for IGF-1 was subtracted from each measured serum IGF-1 concentration. To obtain the baseline IGF-1 value, the two serum IGF-1 levels measured before the rhIGF-1 injection were averaged. The serum IGF-1 half-life was calculated by the formula:

\[
\text{Half-life} = 0.693 / K_e
\]
The area under the curve (AUC) for exogenous IGF-1 was calculated from 0 to infinity by using the equation:

\[ AUC = \text{Dose/\text{Kel} \cdot Vd} \]

Clearance of serum IGF-1 (SC1) after the subcutaneous injection of rhIGF-1 was calculated as follows:

\[ SC1 = \text{Dose/AUC} \]

The endogenous production rate of IGF-1 was estimated for each patient in the three groups from the following equation:

\[ P = SC1 \cdot S \]

where \( P \) is the assumed endogenous IGF-1 production rate (mg/day) as previously described [14]. SC1 is calculated from the one-compartment model (see above) (liter/day), and \( S \) is the baseline serum concentration of IGF-1 (µg/liter). This calculation is based on the following assumptions: (1) Baseline IGF-1 levels reflect the endogenous serum IGF-1 value for the entire 24 hour period, as shown by Hizuka et al [15]. (2) There are no relatively large, slowly exchanging compartments of IGF-1 in which both synthesis and degradation are occurring. (3) \( P \) and SC1 do not change after the load of rhIGF-1.

### Statistical analysis

Values are given as the mean ± standard error of the mean (SEM). A one-way analysis of variance (ANOVA), with adjustment of \( P \) values for multiple comparisons, was performed to compare the overall mean levels between groups. If the ANOVA indicated a significant trend among groups, the Fisher exact test was used to test for differences between individual groups. Similar statistical analyses were used to compare the post-injection serum concentrations with the baseline values within each group. To determine if there was a dose effect, a one-way analysis of variance with repeated measures was performed to compare the results from the two doses within the same group. Statistical significance was taken as \( P < 0.05 \).

### Results

The serum IGF-1 levels are shown in Figures 1 and 2 and Table 2. Figures 1 and 2 show the average values at a given time, whereas Table 2 indicates the average maximum serum IGF-1 levels (\( C_{\text{max}} \)), which did not always occur at the same time. Baseline serum IGF-1 concentrations were not different between groups before the injection of either dose of rhIGF-1. After each subcutaneous injection of rhIGF-1, there was a rapid and highly significant rise in serum IGF-1 in each group of subjects. The \( C_{\text{max}} \) and the increment in serum IGF-1 levels were significantly greater with the 100 µg rhIGF-1/kg dose as compared to the 50 µg rhIGF-1/kg dose for each group of subjects (\( P < 0.05 \); Table 2).

With the 50 µg/kg dose, serum IGF-1 increased by 386 ± 54, 416 ± 56 and 331 ± 54 µg/liter above baseline in the MHD, CAPD and normal subjects, respectively. Among the three groups of subjects given 50 µg rhIGF-1/kg, there was no difference in either the increment or in the absolute concentrations of serum IGF-1 (Fig. 1, Table 2). Twenty-four hours after injection of rhIGF-1, the serum IGF-1 remained significantly greater than baseline only in the CAPD and normal subjects, by 61% and 43%, respectively (\( P < 0.05 \) for each group).

After the 100 µg/kg dose, the serum IGF-1 values increased more in the dialysis patients than in the normal subjects (Fig. 2, Table 2). Serum IGF-1 increased by a maximum of 810 ± 73, 887 ± 124 and 471 ± 80 µg/liter above baseline in the MHD, CAPD and normal subjects, respectively (MHD vs. normal \( P < 0.05 \), CAPD vs. normal \( P < 0.025 \)). With 100 µg rhIGF-1/kg, serum IGF-1 was significantly greater in the MHD patients than in normals at 90, 120, and 180 minutes. In CAPD patients, serum IGF-1 was greater than in normal subjects at 120, 180 and 240 minutes after injection. Serum IGF-1 levels 24 hours after injection were almost identical in the three groups, and were 69%, 132% and 114% greater than baseline in the MHD, CAPD and normal subjects, respectively (\( P < 0.05 \) for MHD, \( P < 0.025 \) for CAPD and normals).

The \( T_{\text{max}} \) did not differ with either the 50 µg/kg or 100 µg/kg dose among the three groups (Table 2). The half-life of rhIGF-1
was significantly shorter in the MHD and CAPD patients as compared to normals after the 100 μg/kg dose (P < 0.005; Table 2). There was no difference in the AUC or serum clearance of IGF-1 with either dose among the three groups. After the 50 μg/kg rhIGF-1 dose, the volume of distribution (Vd) of IGF-1 was not different in the three groups (Table 2, Fig. 3). In the MHD and the CAPD patients, the Vd was essentially unchanged after the 100 μg/kg dose as compared to the 50 μg/kg dose. In the normals, the Vd also did not rise significantly with the 100 μg/kg dose. However, the absolute values for Vd tended to rise more in the normal individuals, and the Vd with the 100 μg/kg dose was reduced by more than 50% in both the MHD and CAPD patients as compared to normals (P < 0.005 for MHD or CAPD vs. normal; Fig. 3).

The estimated endogenous production of IGF-1 (P) is shown in Table 3. In general, the estimated P rates for the two doses of rhIGF-1 within each group were similar. This was particularly true for the CAPD patients and normal subjects, where P with the higher rhIGF-1 dose was 10% and 4% different, respectively, from P with the lower dose. The estimated P was not different between the three groups at either dose of rhIGF-1, although the mean P value for each group varied from 34 to 61 μg/kg/day.

**Discussion**

We studied the pharmacokinetics of rhIGF-1 because of the growing interest in both the use of growth hormone and IGF-1 for the treatment of patients with renal failure as well as other clinical disorders [4, 16–20]. Growth hormone stimulates the synthesis and release of IGF-1 which mediates most, if not all, of the anabolic effects of growth hormone [1]. Growth hormone has been used successfully for the treatment of impaired growth in children with chronic renal failure [21–23], and both growth hormone and rhIGF-1 have been used experimentally to treat malnutrition. Currently, there are no published data on the pharmacokinetics of rhIGF-1 in patients with chronic renal failure. Since, in renal failure, serum concentrations of some IGF-1
binding proteins are abnormal [24–27] and the contribution of the kidneys to the synthesis, degradation and urinary excretion of IGF-1 should be reduced, it would not be unexpected for the metabolic fate of IGF-1 to be altered.

In all three groups when the dose of rhIGF-1 was doubled, there was a significant increase in the maximum serum IGF-1 concentration and the maximum increment in serum IGF-1 above baseline (Δmax; Table 2). The AUC tended to increase with the higher dose, although this rise was significant only in the CAPD patients. When the rhIGF-1 dose was doubled in the normal subjects, the rise in Cmax and Δmax was disproportionately small. Cmax rose by only 28% and Δmax by 42%. These findings are similar to the results following injection of 50 and 100 μg/kg or 40 and 80 μg/kg of rhIGF-1 in healthy adults by Wilton and associates, Hizuka and coworkers, and Strong and Raskin [14, 15, 28]. These investigators also noted a disproportionately smaller rise in Cmax and Δmax with the higher rhIGF-1 dose. In contrast, in the MHD and CAPD patients, in the present study when the dose of rhIGF-1 was doubled, the Cmax and Δmax also increased by about 100%. Indeed, the Cmax and Δmax with the 100 μg/kg dose of rhIGF-1 were significantly greater in both the MHD and CAPD patients as compared to the normal subjects.

Perhaps the most striking observation from this study was that the MHD and CAPD patients, as compared to the normal adults, tended to have a shorter half-life and a reduced Vd. These differences were statistically significant with the higher dose of rhIGF-1 (100 μg/kg; Table 2, Fig. 3). The cause for the smaller Vd in the dialysis patients could be due to elevated serum IGF-1 binding proteins (IGFBP) and binding protein fragments. Several studies have described either increased serum concentrations of IGFBP and binding proteins fragments [7, 25, 26] or an increased serum binding capacity for IGF-1 [7, 8, 29] in patients with chronic renal failure. Serum IGFBP-1, IGFBP-2, IGFBP-4 and IGFBP-6 are increased two- or threefold in adults with advanced renal failure [27, 30]. Serum IGFBP-3, the most abundant serum IGFBP, is variously reported to be normal or increased in chronic renal failure [24, 31], whereas serum concentrations of the large ternary IGFBP-3 complex which normally is responsible for most of the serum binding of IGF-1 [32] appears to be reduced in chronic renal failure [24]. These observations provide indirect evidence in support of the hypothesis that increased serum IGF-1 binding is a cause for the reduced Vd in dialysis patients.

The mechanism for the reduced serum IGF-1 half-life in the dialysis patients is also uncertain. Some plasma IGF-1 binding proteins have shorter half-lives [32]. If these binding proteins are increased in renal failure patients, it might lead to a shorter IGF-1 half-life.

In spite of the almost total absence of renal function in the dialysis patients, it was somewhat surprising that their serum IGF-1 half-life was not increased. The kidney is a major site for the degradation of peptides, including peptide hormones [33]. Hence, it would not be unexpected for the kidneys to be a major site for the catabolism of rhIGF-1. There are currently no data on the metabolism of IGF-1 by the kidney. The normal urinary IGF-1 excretion appears to be quite low relative to the total amount injected in the present study or estimated to be synthesized endogenously (see below) and is roughly only about 0.1 to 2.0 μg/day in adult men and women [5, 34]. Hizuka et al reported that in normal men, urinary IGF-1 excretion increased from a baseline level of 0.1 μg/day to 1.4 μg/day after the subcutaneous injection of 100 μg rhIGF-1/kg body wt [5]. The present results indicate that the loss of the metabolic activity of the kidney do not substantially increase the half-life or decrease the SC1 of IGF-1 in renal failure patients. These findings suggest that the normal kidneys do not play a major role in the total body catabolism or clearance of IGF-1. It should be noted that the foregoing considerations assume that the bioavailability of subcutaneously injected rhIGF-1 is 100%, a finding that Kabi Pharmacia is reported to have found [14].

Wilton and coworkers have published the only other pharmacokinetic analysis of IGF-1 in normal humans [14]. These authors gave subjects a subcutaneous injection in the thigh of 40 and 80 μg rhIGF-1/kg body wt. Although the SC1 for IGF-1 was similar to the present results, the Cmax was lower and the Tmax, half-life, and Vd were greater than in the normals in the present study. Subcutaneous injection of insulin is associated with variable absorption kinetics, depending on the location of injection [35]. There is a greater lag time and higher Tmax for 125I-insulin when it is injected subcutaneously in the thigh as compared to the abdomen [35]. If a similar phenomenon exists for rhIGF-1, this could explain the discrepant findings between the present study and that of Wilton et al [14].

Our estimate of the assumed endogenous production of IGF-1 is about 49 μg/kg/day (3.1 mg/day) in normal adults and roughly similar in the MHD (61 μg/kg/day) and CAPD (34.5 μg/kg/day) patients (Table 3). Our data for normal adults are similar to that reported by Wilton et al, 38 μg/kg/day (3 mg/day) [14]. These data indicate that the two rhIGF-1 doses employed in the present study, 50 and 100 μg/kg body wt, are roughly similar to our daily assumed endogenous production of IGF-1 in the normal and dialysis subjects.

The results of the present study indicate that, although the pharmacokinetics of rhIGF-1 following a single subcutaneous injection are different in MHD and CAPD patients as compared to normal adults, these differences are not marked, particularly at lower (that is, 50 μg/kg body wt) doses of rhIGF-1. With the 100 μg/kg dose, serum IGF-1 concentrations are greater in MHD or CAPD patients than in normal individuals for the first four hours, but by 10 to 12 hours after injection of rhIGF-1, the serum levels are similar. Also, for each dose of rhIGF-1, over the 24 hour period of observation, the AUC and, hence, exposure to the compound was similar in the normal and dialysis patients. If higher peak serum IGF-1 levels are hazardous, it may be preferable to give chronic renal failure patients smaller, more frequent injections of rhIGF-1 to prevent abnormally high serum IGF-1 concentrations. On the other hand, the shorter IGF-1 half-life in renal failure suggests that IGF-1 is not more likely to accumulate with extended treatment.

It is not known whether the pharmacokinetics of IGF-1 will change in the dialysis subjects with multiple doses. RhIGF-1 injections are known to increase serum concentrations of some IGF-1 binding proteins—particularly IGF binding protein 2 [36]. A rise in serum binding proteins could alter IGF-1 pharmacokinetics. However, in three studies of normal adults given daily subcutaneous injections of rhIGF-1 for seven days, most pharmacokinetic parameters did not change dramatically [5, 14, 28]. It is possible that the rate of endogenous IGF-1 synthesis or catabolism or the rise in serum IGF-1 binding proteins in response to repeated administration of rhIGF-1 may be different in MHD or
CAPD patients than in normal adults. Further studies will be necessary to answer this question.

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References

11. BLUM WF, BREEHER BH: Radioimmunoassays for IGFs and IGFBPs, Growth Reg 4:51–52, 1994


