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Single-Dose Pharmacokinetics, Safety, and Tolerability of Albinterferon Alfa-2b in Subjects with End-Stage Renal Disease on Hemodialysis Compared to Those in Matched Healthy Volunteers[▽]

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Albinterferon alfa-2b (albIFN) is being developed, in combination with ribavirin, for the treatment of hepatitis C virus infection. This study was designed to evaluate the pharmacokinetics, safety, and tolerability of a 900-μg dose of albIFN administered as a single subcutaneous injection in end-stage renal disease (ESRD) patients on hemodialysis and matched healthy volunteers (by age [±5 years], weight [±5 kg], and gender). The maximum concentration in plasma (C_{max}) and the area under the concentration-time curve from time zero to infinity ($AUC_{0-\infty}$) were 42.8 ± 14.0 ng/ml and 16,414 ± 4,203 ng · h/ml, respectively, for healthy volunteers, while the C_{max} and $AUC_{0-\infty}$ were 49.9 ± 20.9 ng/ml and 18,919 ± 8,008 ng · h/ml, respectively, for ESRD patients. The geometric least-squares mean ratios were 1.15 (90% confidence interval [CI], 0.78, 1.68) for C_{max} and 1.11 (90% CI, 0.83, 1.48) for $AUC_{0-\infty}$. Adverse events were as expected for an interferon (e.g., flu-like symptoms), with the main laboratory adverse event being a decline in total white blood cell count, which was specifically related to a decline in the neutrophil count. This effect was somewhat greater in the ESRD patients, with the maximal decreases in neutrophil counts from those at the baseline being $(-2.6 \pm 0.32) \times 10^9$ and $(-2.19 \pm 0.58) \times 10^9$ cells/liter for the ESRD patients and the healthy volunteers, respectively. This study indicates no significant effect of renal failure on the pharmacokinetics of albIFN. Safety and tolerability were as expected for an interferon.

Hepatitis C virus (HCV) is an important cause of chronic liver disease. The World Heath Organization estimates that approximately 3% of the world's population, or as many as 170 million persons worldwide, are infected with HCV (7). Almost 4 million people in the United States, 8.9 million people in Europe, and approximately 0.25 million people in Canada and Australia have antibodies to HCV, indicating ongoing or previous infection with the virus. The prevalence of HCV infection in some countries in Africa, the eastern Mediterranean, Southeast Asia, and the western Pacific (when prevalence data are available) is higher (ranging from 21 million to 62 million people) than that in some countries in North America and Europe (7). Chronic HCV infection is the cause of cirrhosis and hepatocellular carcinoma in a significant proportion of subjects with these diseases.

An interferon, usually pegylated (PEG) modified, plus oral ribavirin is the standard of care for the treatment of chronic HCV. The treatment duration depends on the HCV genotype and is generally 24 to 48 weeks, with the individual being assessed for a sustained viral response at 6 months after the end of treatment. Ribavirin, secondary to toxicity, is contraindicated in individuals with a creatinine clearance (CL_{CR}) of less than 50 ml/min, leaving no treatment or monotherapy with an interferon as the only clinical option in this patient group. A significant percentage of subjects with end-stage renal disease (ESRD) have HCV infection, for which physicians may want to consider treatment with albinterferon alfa-2b (albIFN) (2).

Albinterferon alfa-2b (albIFN) is a novel genetic fusion protein composed of the mature form of recombinant human albumin (rHA) and the mature form of recombinant interferon alfa-2b (rIFN-α2b) developed for the treatment of patients with chronic HCV infection. Alpha interferon (IFN-α) is a cytokine that plays a key role in immune regulation via activation of a cascade of intracellular pathways and has antiviral, immunomodulatory, and antiproliferative effects. IFN-α has been the mainstay of anti-HCV therapy for the past decade. albIFN has high antiviral activity, has safety and tolerability similar to those of the standard of care, and has a prolonged elimination half-life ($t_{1/2}$) which maintains drug concentrations above the 90% effective concentration over prolonged dosing intervals (6). Three phase 2 trials and two phase 3 trials of albIFN in IFN treatment-experienced and treatment-naïve subjects with chronic HCV infection have been completed to evaluate efficacy and safety (6). Animal studies indicate that albinterferon has no significant renal clearance (Novartis and Human Genome Sciences, data on file), and therefore, the pharmacokinetics in humans with ESRD on hemodialysis are not expected to be different from those in matched healthy volunteers. In contrast, the clearance of pegylated interferon alfa-2b is reduced by approximately 50% in patients with a creatinine clearance of less than 50 ml/min (1). For pegylated interferon alfa-2a, a modest 25% to 45% higher level of exposure is seen in subjects undergoing hemodialysis (4). The present study evaluated a dose of 900 μg

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TABLE 1. albIFN arithmetic mean pharmacokinetic parameters following a single subcutaneous dose of 900 μg albIFN

Subject	Parameter	T_{max} (h)[a]	C_{max} (ng/ml)	AUC_{0-last} (ng · h/ml)[c]	$AUC_{0-\infty}$ (ng · h/ml)	$t_{1/2}$ (h)	CL/F (ml/h)
Healthy volunteers (n = 8)	Mean	168 (48–169)	42.8 ± 14.0	16,151 ± 4,134	16,414 ± 4,203	133 ± 26	57.6 ± 13.1
	CV (%)		32.6	25.6	25.6	19.6	22.7
Hemodialysis patients (n = 8)	Mean	96 (47–169)	49.9 ± 20.9[b]	18,530 ± 7,741	18,919 ± 8,008	146 ± 34	53.4 ± 16.6
	CV (%)		44.0	41.8	42.3	23.7	31.1

[a] Data represent median (min–max).
[b] The value is 50.9 ± 22.4 ng/ml when the data for subject 5105 are excluded.
[c] AUC to the last detectable concentration time point.

albinterferon, as it was the lower of the bimonthly doses evaluated in phase 3 studies and allowed an adequate safety margin in the event of an unexpected increase in the level of exposure to albIFN in subjects with ESRD (8).

MATERIALS AND METHODS

Study design. This was an open-label, single-subcutaneous-dose study conducted to explore the pharmacokinetic safety and tolerability of albIFN in HCV-negative patients with ESRD on hemodialysis compared to those in healthy matched volunteers (subjects matched by age [±5 years]), weight [±5 kg], and gender). Eight dialysis patients and eight healthy volunteers who met the eligibility criteria, after a screening period of no more than 30 days, received a single injection of study medication. Within each matched pair of ESRD subject and healthy volunteer, one of the following injection sites was selected: abdomen or upper thigh. ESRD patients received study medication within 2 h of the completion of their dialysis, while it was administered at time zero to the matched healthy volunteers. Pharmacokinetics were evaluated for up to 6 weeks following the dose. Safety assessments were required during and for up to 2 weeks following completion of the pharmacokinetic sampling (details are in "Safety and tolerability assessments"). The main inclusion criteria consisted of the following: male or female of nonchildbearing potential) between 18 and 75 years of age, inclusive; no reported history of treatment for depression or a history of intolerance or allergic reactions to alpha interferons; alanine aminotransferase (ALT) and aspartate aminotransferase (AST) concentrations ≤3 times the upper limit of normal; and bilirubin concentrations ≤1.5 times the upper limit of normal. Patients with ESRD were required to be on a stable hemodialysis program (defined as above 1.2 dialyzer clearance of urea · dialysis time/volume of distribution of urea within the past 4 weeks without a significant change in the past 3 months) and to present with a hemoglobin concentration of >10 g/dl, a white blood cell (WBC) count within the range of from 3,500 to 11,000/μl, and a platelet count of between 100,000/μl and 600,000/μl prior to study drug administration. Healthy volunteers were required to have a creatinine clearance, as calculated by the Cockcroft-Gault formula, of >70 ml/min and to present with hemoglobin concentrations, white blood cell counts, and platelet counts in the normal range prior to study medication administration.

The main exclusion criteria consisted of the following: a positive hepatitis B virus surface antigen (HBsAg) or hepatitis C virus test result and significant acute illness within 2 weeks prior to initial dosing.

This single-center study was conducted at CRS Clinical Research Services, Kiel, Germany, in accordance with good clinical practice (GCP) guidelines and the Helsinki Declaration. The patient informed-consent form was approved by the local ethics committee (Ethikkommission der Aerztekammer Schleswig-Holstein, Segeberg, Germany) and health authorities.

albIFN was manufactured in the United States and was provided to Novartis by Human Genome Sciences, Inc., Rockville, MD.

Serum concentrations of albinterferon. Serum concentrations of albIFN were measured using a validated enzyme-linked immunosorbent assay (ELISA) method. The ELISA measured the interferon moiety of albIFN using an anti-human IFN-α capture/anti-human IFN-α detection sandwich format. The lower limit of quantitation for the assay was 0.53 ng/ml, and the upper limit of quantitation was 50 ng/ml. The precision (coefficient of variation [CV]) and accuracy (bias) were 3.9 to 6.9% and 96.8 to 98.6%, respectively.

Pharmacokinetic analysis. Samples were collected for pharmacokinetic analysis predosing and at 3 (48 h) 5 (96 h), 8, 15, 22, 29, 36, and 43 days postdosing. The following pharmacokinetic parameters were determined using noncompartmental method(s): the area under the concentration time-curve (AUC) from time zero to time t (AUC_{0-t}), the area under the concentration-time curve from time zero to infinity ($AUC_{0-\infty}$), the maximum concentration in plasma (C_{max}), the time to C_{max} (T_{max}), $t_{1/2}$, and clearance (CL/F). Biofluid concentrations were expressed in ng/ml. All concentrations below the limit of quantification or missing data were labeled as such in the concentration data listings. Concentrations below the limit of quantification were treated as zero in the summary statistics for concentration data only. They were not considered for calculation of pharmacokinetic parameters (with the exception of the samples obtained predosing).

Safety and tolerability assessments. The following measurements were collected during this trial: vital signs and body measurements, electrocardiograph readings, and measurements from standard clinical laboratory evaluations (hematology, blood chemistry, and, for the healthy volunteer population, urinalysis). Information on serious adverse events, adverse events, and concomitant medications were collected throughout the trial.

Statistical analysis. Descriptive statistics of the pharmacokinetic parameters included mean, standard deviation (SD), CV, minimum (min), and maximum (max). When a geometric mean was presented, it is stated as such. Since T_{max} was generally evaluated by a nonparametric method, median values and ranges were given for this parameter.

AUC_{0-t}, $AUC_{0-\infty}$, and C_{max} were compared between subjects on hemodialysis and healthy volunteers. Log-transformed values of the pharmacokinetic variables were analyzed by mixed-model analysis of variance with a fixed effect for disease state (patients on dialysis or healthy volunteers) and a random effect for the matching pair. The two-sided 90% confidence interval (CI) for the estimated ratio of the value for the dialysis patients/value for healthy volunteers for mean AUC_{0-t}, $AUC_{0-\infty}$, and C_{max} were calculated. The ratios of geometric means and their confidence intervals were obtained by backtransforming the estimate.

Descriptive statistics were provided for all safety and tolerability assessments.

RESULTS

Subject disposition. Healthy volunteers were matched to the ESRD patients; thus, their demographics were highly similar. An equal number of women and men enrolled in this study, and for the hemodialysis subjects and healthy volunteers, the average ages were 43.9 ± 11.1 years and 44.1 ± 10.2 years, respectively, and the average weights were 84.5 ±13.3 kg and 84.5 ± 11.3 kg, respectively.

Pharmacokinetics. Data from all subjects were used in the pharmacokinetic parameter calculations. The scheduled determination (~C_{max}) at 96 h was delayed by ~18 h in one subject. Thus, the C_{max} from this subject (subject 5105) was included only in the descriptive statistics of pharmacokinetic parameters. The arithmetic mean values of albIFN pharmacokinetic parameters in ESRD patients on hemodialysis and matched healthy volunteers are listed in Table 1. The mean concentration-time profiles are shown in Fig. 1. All subjects with evaluable pharmacokinetic data, except C_{max} in subject 5105, were included in the inference analysis, and the results are presented in Table 2. On the basis of the geometric least-squares mean ratios for both C_{max} and AUC including the value of 1 (i.e., no effect) in the analysis described above and the small

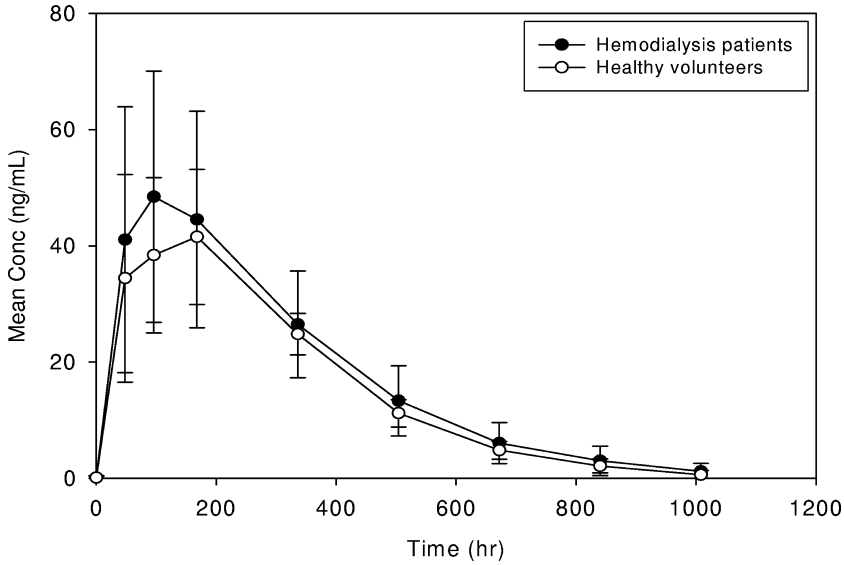

FIG. 1. albIFN concentration (mean ± SD)-time profile following a single subcutaneous dose of 900 μg.

magnitude of the effect (11% for AUC), there is no evidence of a systematic difference in pharmacokinetic parameters between the two groups.

Safety and tolerability. Adverse events by treatment group and incidence are presented in Table 3. There were no serious adverse reactions in the study. The most common adverse events were similar to those observed with interferon administration. Generally, the adverse events observed were flu-like symptoms, and there was no clear difference between the two treatment groups. The adverse event severity was mild to moderate except in three subjects, in whom it was graded as severe. One hemodialysis patient presented with a decrease of the leukocyte count on study day 8. The nadir was observed on day 8 (1.8×10^9 cells/liter). The counts returned to within normal limits (4×10^9 to 10×10^9 cells/liter) by the end of the study visit (4.3×10^9 cells/liter). One healthy volunteer presented with chills on study day 1, and one healthy volunteer presented with flu-like symptoms and nausea/vomiting on study day 1. Pharmacological intervention was provided to the healthy volunteers. No intervention was required for the hemodialysis patient. All events resolved on follow-up.

Total WBC counts declined in both groups after a single dose of albIFN. This reduction was specifically related to a decline in the neutrophil count, which is a known adverse effect of interferons. The absolute baseline neutrophil counts were $3.84 \times 10^9 \pm 0.53 \times 10^9$ cells/liter in the healthy volunteers and $4.80 \times 10^9 \pm 0.50 \times 10^9$ cells/liter in the ESRD patients. The maximal decrease in neutrophil count from the baseline was somewhat higher for the ESRD patients than for the healthy volunteers at $(-2.6 \pm 0.32) \times 10^9$ and $(-2.19 \pm 0.58) \times 10^9$ cells/liter, respectively (Fig. 2).

DISCUSSION

This study examined the effect of renal function on the pharmacokinetics of albinterferon alfa-2b in ESRD patients on hemodialysis compared to those in matched healthy volunteers.

The healthy volunteers were well matched to the ESRD patients by age, weight, and gender, minimizing potential confounding variables in the evaluation of the comparative pharmacokinetics. The main findings of this single-dose study were that albIFN was safe and that there were no significant differences in the values of the pharmacokinetic parameters between hemodialysis patients and matched healthy volunteers. The 1.11 (90% CI, 0.83, 1.48) ratio for $AUC_{0-\infty}$, which indi-

TABLE 2. Statistical analysis of pharmacokinetic parameters

Parameter	Subject group	No. of subjects	Geometric least-squares mean	Geometric least-squares mean ratio for hemodialysis patients/ healthy volunteers	90% confidence interval
$AUC_{0-\infty}$ (ng · h/ml)	Hemodialysis patients	8	17,759	1.11	0.83, 1.48
	Healthy volunteers	8	15,998		
AUC_{0-last} (ng · h/ml)	Hemodialysis patients	8	17,422	1.11	0.83, 1.48
	Healthy volunteers	8	15,737		
C_{max} (ng/ml)	Hemodialysis patients	7	47.0	1.15	0.78, 1.68
	Healthy volunteers	8	40.9		

TABLE 3. Summary of adverse events by treatment group

Body system	Preferred term	No. (%) of subjects	
		Hemodialysis patients (n = 8)	Healthy volunteers (n = 8)
Any body system	Total	8 (100)	7 (87.5)
Blood and lymphatic system	Pancytopenia	1 (12.5)	0 (0)
Ear and labyrinth disorders	Vertigo	0 (0)	1 (12.5)
GI[a] disorder	Nausea	0 (0)	2 (25)
	Vomiting	0 (0)	2 (25)
General disorders/administration site conditions	Chills	0 (0)	1 (12.5)
	Feeling cold	0 (0)	1 (12.5)
	Influenza like illness	6 (75)	2 (25)
	Injection site erythema	0 (0)	2 (25)
	Pyrexia	0 (0)	1 (25)
Infections and infestations	Nasopharyngitis	1 (12.5)	0 (0)
	Oral herpes	0 (0)	2 (25)
Investigations	ALT increased	2 (25)	0 (0)
	AST increased	1 (12.5)	0 (0)
	GGT[b] increased	1 (12.5)	0 (0)
	WBC decreased	1 (12.5)	0 (0)
	Platelet count decreased	1 (12.5)	0 (0)
Muscloskeletal and connective tissue disorders	Back pain	0 (0)	1 (12.5)
	Musculoskeletal pain	0 (0)	1 (12.5)
	Pain in extremities	0 (0)	1 (12.5)
Nervous system disorder	Headache	0 (0)	3 (37.5)
Skin and subcutaneous tissue disorder	Skin fissures	0 (0)	1 (12.5)

[a] GI, gastrointestinal.
[b] GGT, gamma-glutamyltransferase.

cates an 11% higher exposure than that for the healthy volunteers, is within the intersubject variability of albIFN and is not considered clinically relevant. The safety and laboratory findings were consistent with the differences in the two patient populations. The main interferon-related adverse effects were influenza-like symptoms after the dose and a decline in the total WBC count as a result of a decline in the neutrophil count; the effect was somewhat greater in the ESRD patients, with the

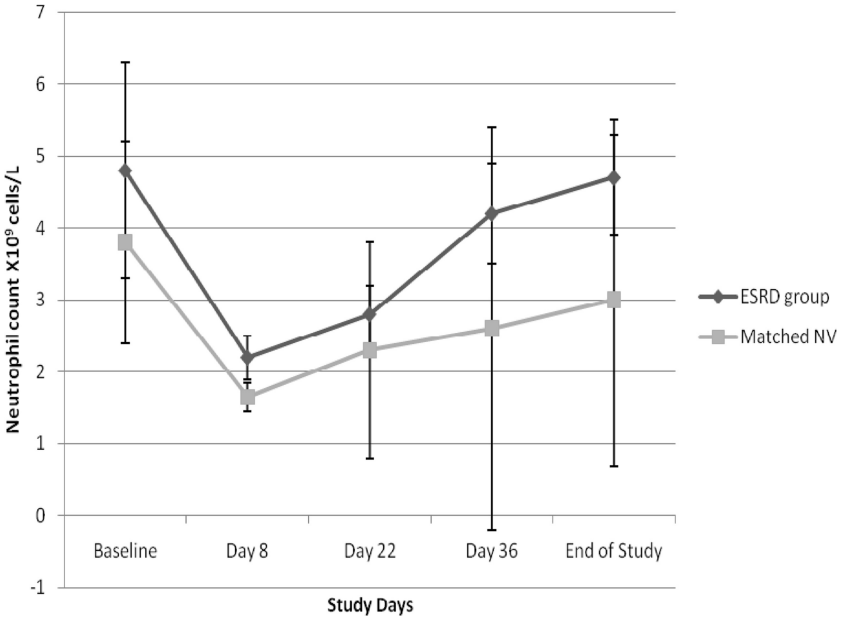

FIG. 2. Absolute neutrophil counts in the matched healthy volunteers (NV) and hemodialyzed ESRD patients.

maximal decreases in neutrophil count from that at the baseline being $(-2.6 \pm 0.32) \times 10^9$ cells/liter and $(-2.19 \pm 0.58) \times 10^9$ cells/liter for the ESRD patients and the healthy volunteers, respectively. Bone marrow suppression is a known effect of all interferons, with effects on neutrophil count occurring earlier than effects on hemoglobin or platelets. The 900-μg dose used as a single dose in this trial, when given every 2 weeks with ribavirin on the basis of the subject's weight, was not different from pegylated interferon alfa-2a in inducing neutropenia (8). There was a modest decline in the hemoglobin concentration (~0.6 to 0.7 g/liter) over the study in both groups, consistent with the blood-drawing requirements (130 ml) of the study, although an effect from albIFN cannot be ruled out.

A preclinical study was performed in a nephrectomized rat model to assess the effect of renal function on the elimination of several interferons, including albIFN, prior to the initiation of this clinical study (Novartis and Human Genome Sciences, data on file). Rodents (age, 8 weeks) underwent bilateral nephrectomy or a sham procedure. Upon recovery, study drug (albIFN, recombinant interferon-alfa-2b [Intron A], PEG-IFN-α2b [PEG-Intron], or peginterferon alfa-2a [Pegasys]) was administered intravenously via the tail vein. Blood samples were collected for up to 68.75 h after study drug administration (68.75 h for albIFN and PEG IFN, 54 h for peginterferon alfa-2a, and 24 h for recombinant albIFN-α2b). The result of this study indicated little change in albIFN (<3%) or PEG IFN-α2a (7%) clearance in nephrectomized rats, while IFN-α2b and PEG IFN-α2b clearances were 5- to 10-fold slower in nephrectomized rats than in the control animals. These results indicated that renal clearance contributes more to the elimination of smaller proteins like recombinant albIFN for injection (19.3 kDa) and PEG IFN-α2b (31 kDa) and little to larger proteins like peginterferon alfa-2a (60 kDa) and albIFN (85.7 kDa). This is consistent with data indicating that small therapeutic proteins are thought to be filtered by the glomeruli in the kidney and reabsorbed into the proximal tubules, where degradation occurs (3), while the modified (pegylated) interferons are generally eliminated by plasma and tissue proteases with minor renal excretion. The clinical findings reported in the literature and from this study are also consistent with the animal data, in that the impacts of renal function are in the same rank order with the molecular size. The overall effects of ESRD on the pharmacokinetics of the interferons in humans are somewhat greater than those found in the nephrectomized rat model, and this may be a reflection of the short sampling time (up to 68.75 h) compared to the half-lives of some of the agents, such as albIFN and peginterferon alfa-2a, in humans, which are in the range of 6 to 8 days.

albIFN has a large molecular size of 85.7 kDa. It is not expected to be cleared by the use of hemodialysis since the smaller protein albumin is not cleared and hemodialysis poorly clears even smaller proteins such as β_2-microglobulin (molecular mass, ~11 kDa) (5). The results from this study confirmed that albIFN pharmacokinetics are not altered by severe renal impairment, with modest 11 and 15% differences in $AUC_{0-\infty}$ and C_{max}, respectively, being detected. In contrast, the values of the PEG IFN-α2b pharmacokinetic parameters (AUC, C_{max}, and $t_{1/2}$) were reported to increase progressively as CL_{CR} declined (1). In patients with CL_{CR}s of <30 ml/min, PEG IFN-α2b AUCs and C_{max}s were increased 90% compared with those for healthy controls, while its half-life was increased by up to 40% over that in healthy controls. In the case of PEG IFN alfa-2a, it has been shown that the C_{max} and CL/*F* in subjects with stable chronic renal impairment are comparable to those in patients with normal renal function, while the CL/*F* of PEG IFN alfa-2a was 25% to 45% lower in patients on hemodialysis than in subjects who had normal kidney function (7). The approved labeling for PEG IFN-α2b and peginterferon alfa-2a reflects these findings, with dose reductions of 50% and 25% being recommended for each drug, respectively, for patients with severe renal impairment.

No dose adjustment is recommended for albinterferon in renally impaired patients on the basis of the results of this study. The pharmacokinetics of albIFN were not affected by ESRD in subjects who were on a stable hemodialysis program. The clinical study results are consistent with the prediction for molecular size clearance on dialysis and the nephrotomized rat model. The safety profiles were as expected for an interferon, with influenza-like symptoms and neutropenia being observed.

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