Clinical Pharmacology Review of Peg-interferon alfa-2a (Ro25-8310, PEGASYS)

Sponsor: Hoffmann-La Roche
Indication: Chronic Hepatitis C, 180 ug, sc QW for 48 weeks
BLA 103964

Bioavailability and Bioequivalence Studies

1. Protocol NP 15537, A bioavailability study of Ro 25-8310 administered subcutaneously and intravenously to healthy subjects, RR N-181142. June 25, 1999
2. Protocol NP 15762, A relative bioavailability study of pegylated---- interferon α-2a administered subcutaneously via needle injection and ---------- to healthy volunteers. RR N-181460. April 15, 2000
3. Protocol NP 15985, A study of the feasibility of using the ---------- ----- for subcutaneous injection, N-181606. 2000
4. Protocol NP 15989, A bioequivalence and relative bioavailability study of PEG-IFN administered s.c. via needle injection and ---------- to various sites in healthy subjects, N-181607. May 5, 2000

Dose Escalation Study

5. Protocol NP 15330, Single-dose safety/tolerability and pharmacokinetics/pharmacodynamics following administration of ascending subcutaneous doses of PEG-interferon alfa-2a and Roferon-A to healthy subjects. March 15, 1999.

Clinical Studies

6. Protocol NP 15538, Pharmacokinetic study of Ro 25-8310 in male and female healthy subjects following subcutaneous administration. RR N-181141. April 16, 1999.
7. Protocol NP 15580, A single center open label, parallel design, 12 young and 12 elderly healthy subjects to assess the effect of age on Ro 25-8310 pharmacokinetics following subcutaneous administration. RR N-181144. April 7, 2000.
8. Protocol NP15579, Pharmacokinetic study of Ro 25-8310 in compromised renal function patients following subcutaneous administration. RR N-181145. March 17, 2000
9. Protocol JP 15722, Phase I clinical study of Ro 25-8310 (PEG-interferon alfa-2a) in healthy volunteers. N-181609. April 10, 2000.
10. Protocol NV 15489, A phase II, open-label randomized, multicenter, ascending-dose study, evaluating the safety and efficacy of peg-interferon alfa-2a vs interferon alfa-2a in the Rx of noncirrhotic patient with chronic hepatitis C. N-181405. February 14, 2000.
11. Protocol NV 15495, A phase II/III Open-label, randomized multicenter, parallel-group study evaluating the safety and efficacy of peg-interferon a-2a (Ro 25-8310) vs Roferon-A in the treatment of patients w/chronic hepatitis C with cirrhosis. RR N-181406. February 29, 2000.
12. Protocol NV 15496, A phase III open-label, randomized, multicenter, parallel dose efficacy and safety study comparing PEG-IFN alfa-2a to a standard regimen of Roferon-A in the treatment of patients with chronic hepatitis C. RR N-181408. April 10, 2000.
13. Protocol NV 15497, A phase III open-label, randomized, multicenter, parallel-group efficacy and safety study comparing PEG-IFN alfa-2a to an induction regimen of Roferon-A in the treatment of patients with chronic hepatitis C. RR N-181410. 2000.
14.Protocol NP15581, Effect of Ro 25-8310 on cytochrome P450s (1A2, 2C9, 2C19, 2D6 and 3A4) in healthy subjects. RR N-181143 February 15, 2000.
15.Protocol NP16569, A comparability study of Peginterferon alfa-2a (PEG-IFN) of clinical trial material and the commercial product administered subcutaneously via needle injection in healthy subjects. RRN-1007463 March 29, 2002.

16. Protocol NR16081, A pharmacokinetic/pharmacodynamic ascending dose study to evaluate the safety of a single dose of Pegasys in end-stage renal disease patients undergoing hemodialysis. RR 1003975 April 1, 2002.

Studies Review

1. Protocol NP 15537, A bioavailability study of Ro 25-8310 administered subcutaneously and intravenously to healthy subjects, RR N-181142. June 25, 1999

The purpose of this study was to evaluate the absolute bioavailability of PEG-IFN (Ro 25-8310) given subcutaneously as compared to the intravenous route of administration. The study was conducted as an open-label, randomized, two-way crossover study with a 35 day washout period between administrations of the drug. Twenty healthy male subjects were given 90 ug in 1 ml iv and 180 ug in 2 ml sc on separate occasions. The drug used in this study was from batch C 187376-02.

Pharmacokinetic endpoints were estimates by a model independent approach. An analysis of variance procedure was used to determine F values and the 90% confidence interval for F.

Blood samples for pharmacokinetics and pharmacodynamics measures were collected at predose (0), 0.5, 1, 3, 5, 8, 12, 24, 48, 72, 84, 96, 120, 144, and 168 hours after injection. Serum concentrations of PEG-IFN were determined using a ----------. Pharmacokinetic endpoints were computed with standard techniques and the slope of the terminal log-linear phase of the serum profile was determined by a least squares linear regression from which t1/2 was calculated by dividing 0.693 by the slope.

In anticipation of febrile responses, two 500 mg tablets of paracetamol were permitted and administered at 2, 6, and 10 hours post-dosing and as required, but not exceeding 4 g in any 24 hour period.

Pharmacokinetic analysis from period 1 indicated that the absolute bioavailability of PEG-IFN was 61%. Only the serum values from the first period were used in determining bioavailability as the results of the second period were invalid. Serum concentrations, including Cmax, and AUC for both the iv and sc routes were significantly smaller during the second as compared to the first period for unknown reasons. Furthermore, intersubject variability was much larger in period 2. Five subjects withdrew from the study during or at the completion of period 1. Three subjects' failure to return after their fist injection, one subject refused further treatment, and one subject was withdrawn with transient grade 4 neutropenia. Of the 18 subjects given an iv dose PEG-IFN, 10 occurred in period 1 and 8 in period 2. Of the 17 subjects given a sc dose, 10 occurred in period 1 and 7 in period 2. The pharmacokinetic data are summarized in the table below.

Pharmacokinetic endpoints	90 ug iv, N=10	180 ug sc, N=10
Tmax, h	NA	78 ± 27
Cmax , ng/ml	ND	14.± 2 2.5
T1/2, h	68 ± 31	77 ± 45
AUC168, ug-h/ml	1.4 ± 0.4	1.7 ± 0.5
Vd, L	4.9 ± 0.8 (1)	8 ± 3
Cl, ml/h	60 ± 25 (3)	82 ± 38 (4)

Table of pharmacokinetics from bioavailability study for period 1. NA= not applicable; ND = not determined;(1) Vz; (2) Vx/F; (3) Cl; (4) Cl/F. Values are mean ± SD.

The adverse event profile was that expected of interferons. Influenza-like illness and headache were the most frequently reported adverse event. Other adverse effects included nausea, fatigue, fever, myalgia, neutropenia, lowered WBC and local injection site reaction. The overall incidence of adverse events was lower during period 2 of the study.

2. Protocol NP 15762, A relative bioavailability study of pegylated--- interferon -2a administered subcutaneously via needle injection and ---------- to healthy volunteers. RR N-181460. April 15, 2000.

This was an open-label, single dose study designed in a randomized manner using parallel groups to investigate the relative bioavailability of PEG-IFN (Ro 25-8310) given sc via needle injection or ----------. Pharmacokinetic endpoints were determined from individual profiles of serum concentrations after single iv dose of 90 ug, single sc needle injection of 180 ug and a ----------. The study enrolled 54 healthy male subjects. Lots---------- for 90 ug/ml and ---------- for 180 ug/ml were used. ----------. Various pharmacokinetic endpoints were assessed including serum concentration at 30 minutes (C30), AUC to 168h, apparent Vd associated with the terminal phase, predicted Vd at steady-state, Vd of the central compartment, clearance and relative bioavailability. Additionally, safety parameters were measured including the pain upon injection.

Serial blood samples for the quantification of serum concentrations of PEG-IFN were collected at predose (0 time), 0.5, 1, 3, 5, 8, 12, 24, 48, 72, 84, 96, 120, 144, 168, 192, 216, 240, and 312 hours following injection. Serum levels of PEG-IFN were determined using a ----------. For iv dosing, Vd associated with the terminal phase was calculated as Vz=dose-iv/(terminal slope AUCinf); Vc as dose-iv/C30; Vss as (dose-iv/AUCinf MRT-iv), clearance as dose-iv/AUCinf. For sc dosing the apparent volume of distribution was computed and the apparent total body clearance. Absolute bioavailability was determined as (AUCsc/AUCiv)(dose-iv/dose-sc). Standard means of computing pharmacokinetic endpoints were used in the analysis of the study results. The results are summarized in the table below.

Pharmacokinetic endpoint	90 ug iv, N=18	180 ug, sc, needle, N=15	----------
Tmax, h	NA	102 ± 49	109 ± 43
C30, ng/ml	16 ± 5	NA	NA
Cmax, ng/ml	NA	9 ± 4	10 ± 3
AUC to 168h, ug-h/ml	.7 ± .3	1 ± .5	1 ± .3
T1/2, h	55 ± 31	73 ± 44	79 ± 95
Vc, l	6 ± 2	NA	NA
Vss, l	9 ± 5	NA	NA
Vz, l	NA	13 ± 8	9 ± 5
Cl, ml/h	126 ± 66	NA	NA
Cl/F, ml/h	NA	142 ± 80	124 ± 55

Table of pharmacokinetics comparing iv and sc by either needle ----------. Means ± SD reported.

The absolute bioavailability of PEG-IFN after a single needle sc injection was approximately 80%. The
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4. Protocol NP 15989, A bioequivalence and relative bioavailability study of PEG-IFN administered sc via needle injection and ---------- to various sites in healthy subjects, N-181607. May 5, 2000.

The bioequivalence of PEG-IFN was assessed using needle injection and ---------- in healthy subjects. The sites of subcutaneous injection were the abdomen and thigh. The study involved 173 healthy male and female subjects and was designed as an open-label, single-dose, multi-center using randomized, parallel groups. Females composed 60.7% and males 39.3% of the study population. The protocol specified that 168 subjects were to be enrolled; subjects were added as needed to replace treatment failures in accordance with the planned group sizes and so compensated for any failed injections of the ----------. The primary comparison for this study was that between ---------- and needle injections in the abdomen. Therefore, a sample size of 56 subjects per treatment group for this comparison was planned and based on an estimate of 40% inter-subject coefficient of variation for AUC to demonstrate equivalence within 80 to 125% of the reference mean with a power of 80% at P< or equal to 0.05. A single 1ml sc injection of PEG-IFN was made with a conventional needle and compared to a single sc injection of PEG-IFN from a ----------. The --------- was the ----------.

Doses in both groups were 180 ug of PEG-IFN (Ro 25-8310, Lot --------- for the conventional injection and Lot ----------, however, the concentration of the injected formulations were different. In the conventional injection the strength of the dosing solution was 180 ug/ml ---------- Safety and pharmacodynamic data in terms of serum 2',5'-OAS activity and serum neopterin concentrations were also collected during the course of the study. In anticipation of febrile responses, two 500 mg tablets of paracetamol were permitted and administered at 2, 6, and 10 hours post-dosing and subsequently as required, but not exceeding 4 g in any 24 hour period. Blood samples for pharmacokinetics and pharmacodynamics measures were collected at predose (0), 0.5, 1, 3, 5, 8, 12, 24, 48, 72, 84, 96, 120, 144, 168, 192, 216, 240 and 312 hours after injection. Serum concentrations of PEG-IFN were determined using a ----------. 2',5'-OAS activity was determined using a ------. Plasma neopterin levels were assessed with a ----------. Pharmacokinetic endpoints were computed with standard techniques and the slope of the terminal log-linear phase of the serum profile was determined by a least squares linear regression from which t1/2 was calculated by dividing 0.693 by the slope.

Subjects were divided into 4 groups. Group A (N = 28 subjects) was given a single dose by needle injection to the thigh; group B (N = 56 subjects) by needle injection to the abdomen; group C (32 subjects) [

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Pharmacokinetic results were evaluated as Cmax, Tmax, AUC to last sample, t1/2, apparent total body clearance (Cl), and apparent volume of distribution (Vd) associated with the terminal phase of elimination. Based on pk parameters after sc injection in the ---------- and needle injections are not comparable. These results are summarized below. A relative bioavailability of 84% was found when the AUCs of needle injections in the thigh vs needle injections in the abdomen were compared. Following needle injection in the abdomen, mean serum levels of PEG-IFN were higher compared to those following needle injection in the thigh. Pharmacodynamic differences were noted due to the different sites and means of injection as noted in the table below.

Group	Cmax, ng/ml	AUC-last, ng-h/ml
Abdomen, ----------(group D-S)	11.2	1604
Abdomen, needle injection (group B)	14.3	1902

Table of pharmacokinetic endpoints reported as geometric square means for needle vs successful needle-free injections in the abdomen.

Group	Cmax, ng/ml	AUC, ng-h/ml
Abdomen, ---------- (group D-S) vs needle injection (group B)	0.79	0.84
Abdomen, pk comparability ratio	0.66, 0.94	0.66, 1.08

Table of the ratio of geometric least square means and 90% CI for pharmacokinetic endpoints of needle vs ----------

Group	Cmax, ng/ml	AUC-last, ng-h/ml
Thigh, ---------- (group C-S)	11.2	1522
Thigh, needle injection (group A)	13.1	1603

Table of pharmacokinetic endpoints reported as geometric square means for needle vs ---------- in the thigh

Group	AUC, ng-h/ml
Relative bioavailability of thigh, ---------- w/successful injection (group C-S) vs needle injection (group A)	0.95
Thigh, pk comparability ratio	0.68, 1.32

Table of the ratio of geometric least square means and 90% CI for pharmacokinetic endpoints of needle vs ---------- in the thigh.

Pd parameter	Needle, thigh	----------, thigh	Needle, abdomen	----------, abdomen
Tmax-OAS, h	48 – 312	48 -312	48 - 312	24 - 312
2',5'-OAS, pM/h	769 – 3959	1008 - 7062	2123 - 1089	291- 5106
Tmax, neopterin	48 – 144	24 - 144	24 – 120	24 - 96
Neopterin, nM	7 – 19	6 - 19	4 – 20	5 – 22

Table of pharmacodynamic parameters. Values are the range.

The incidence of adverse effects was highest in group D with successful injections at 98% and followed by group C with successful injections at 93%, group B at 88% and group A at 79%. Thus, groups of individuals given ---------- experienced a slightly higher rate of adverse events as compared to needle injection. The most frequently reported adverse events were headache, influenza-like symptoms, back pain, myalgia and injection site reaction. Adverse event profiles were similar across all treatment groups. The highest incidence of abnormally low neutrophil counts was associated with needle injections in the abdomen.

5. Protocol NP 15330, Single-dose safety/tolerability and pharmacokinetics/pharmacodynamics following administration of ascending subcutaneous doses of PEG-interferon alfa-2a and Roferon-A to healthy subjects. March 15, 1999.

The purpose of the this study was investigate the pharmacokinetics and pharmacodynamics of various single, ascending doses of PEG-IFN (45, 135 and 270 ug protein) given sc to healthy subjects relative to a standard dose of IFNα-2a. Safety and tolerability was also assessed in these subjects to PEG-IFN. The study was conducted in 2 parts. In the first part, a subject was given a single sc dose of 45, 135 or 270 ug of PEG-IFN (Ro 25-8310) or 3 MIU (15 ug) of IFNα-2a. The site of injection was the upper arm. The study design in part 1 was a double-blind, randomized, dose escalation scheme with unequal subjects per dose group. Five subjects were given 45 ug, 32 subjects 135 ug and 16 subjects 270 ug of PEG-IFN; 34 subjects were given 3 MIU of IFNa-2a. In part 2 , five subjects were given a single dose of 18 MIU (90 ug) of IFNα-2a in an open-label manner. Subjects in these investigations were healthy males between the ages of 18 to 45 years of age. All 92 subjects were evaluated for safety, 80 for pharmacodynamics and 77 for pharmacokinetics.

Ro 25-8310 was used from batch number ---------- for the 45 ug/ml dosage form and ---------- for the 90 ug/ml dosage form. For IFN-2a batch number ---------- were used for the 3 MIU dosage form; -------- was used for the 18 MIU dosage form.

Blood samples were collected at 0 (predose), 3, 6, 12, 24, 48, 72, 96, 120, 144, 168, and 192 hours after injection for pharmacokinetics. Similar time points were used for pharmacodynamics with the addition of two more time points at 240 and 264 hours after injection. Serum levels of IFN were determined with an ------ method and those for PEG-IFN with an ----------. Pharmacokinetic results were evaluated as Cmax, Tmax, AUC to last sample, t1/2, apparent total body clearance (Cl), and apparent volume of distribution (Vd) associated with the terminal phase of elimination. The slope of terminal log-linear portion of serum profile was determined by least square regression and the terminal phase t1/2 was calculated by dividing 0.693 by the calculated slope.

The pharmacokinetics of PEG-IFN were examined over the range of doses specified in the protocol. Due to limitations of the analytical assay, only the two highest doses of PEG-IFN (135 ug and 270 ug) were analyzable. Although Tmax's were similar, other pharmacokinetic endpoints did not appear to be proportionate over the two doses. Half-life was decreased and clearance increased. Both Cmax and AUC do not appear proportionate to dose. The pharmacokinetic basis for these results are unclear. Clearance was more than 10 times lower and t1/2 was larger for PEG-IFN as compared to IFN-2a. Cmax was achieved between 12 and 144 hours after injection of PEG-IFN as compared to 3 to 12 hours for IFNa-2a. In terms of pharmacodynamics as reflected by 2',5'-OAS activity, levels following PEG-IFN were more sustained as compared to IFN-2a. The pharmacokinetic results are summarized in the table below.

Pharmacokinetic endpoint	Ro 25-8310		IFNa-2a	
	135 ug, N=24	270 ug, N=14	3 MIU, N=34	18 MIU=5
Tmax, h	76 ± 34	73 ± 28	10 ± 3	11 ± 3
Cmax, ng/ml or U/ml	7.9 ± 6.1	10.0 ± 5.0	13.4 ± 3.7	57.7 ± 12.3
T1/2, h	104 ± 65	69 ± 49	9 ± 6	8 ± 3
AUC, ng-h/ml or U-h/ml	809 ± 424	1063 ± 706	153 ± 67	884 ± 154
Clearance, ml/h	128 ± 51	279 ± 176	11836 ± 3238	16824 ± 3521

Table of pharmacokinetic endpoints for PEG-IFN and IFNa-2a. Mean ± SD.

The adverse event profile for PEG-IFN and IFN-2a were similar. The most frequent adverse events were influenza-like illness, headache, fatigue and bruising. The most frequently reported laboratory abnormalities were decreases in neutrophils and slight increases in ALT.

6. Protocol NP 15538, Pharmacokinetic study of Ro 25-8310 in male and female healthy subjects following subcutaneous administration. RR N-181141. April 16, 1999.

The purpose of this study was to assess the effect of gender on the pharmacokinetics of PEG-IFN (Ro 25-8310) after sc injection in healthy subjects. This was an open-label, nonrandomized, single dose study that was conducted at a single site. Lot ---------- of PEG-IFN was used. Twenty-four subjects (12/sex) were enrolled and given a dose of 180 ug of PEG-IFN. Pharmacokinetic measures were collected and safety assessed during the course of the study. Adverse events due to PEG-IFN were as expected and included influenza-like illness, headache, nasal congestion and bruising at the site of injection. Paracetamol was administered prophylactically at 2, 6, and 10 hours post-dosing and subsequently as required..

Blood samples were collected at 0 (predose), 3, 6, 12, 24, 48, 72, 96, 120, 144, 168, and 264 hours after injection for pharmacokinetics. Similar time points were used for pharmacodynamic measures with the addition of two more time points at 240 and 264 hours after injection. Serum levels of IFN were determined with an ------ method and those for PEG-IFN with an ----------. Pharmacokinetic results were evaluated as Cmax, Tmax, AUC to last sample, t1/2, apparent total body clearance (Cl), and apparent volume of distribution (Vd) associated with the terminal phase of elimination. The slope of terminal log-linear portion of the serum profile was determined by least square regression and the terminal phase t1/2 was calculated by dividing 0.693 by the calculated slope.

Peak levels of PEG-IFN between 12 to 14 ng/ml were observed at approximately 80 hours after injection. The t1/2 was approximately 51 hours. No gender specific changes in the pharmacokinetics of PEG-IFN were found. The pharmacokinetic results are summarized in the table below.

Pharmacokinetic endpoints	Female, N=12	Male, N=12
Tmax, h	84 ± 26	81 ± 27
Cmax, ng/ml	12 ± 3	14 ± 4
AUC264, ug-h/ml	1.7 ± .8	1.7 ± .8
Vd, l	6 ± 4	7 ± 3
Cl, ml/h	98 ± 42	100 ± 42
T1/2, h	51 ± 39	52 ± 29

Table of pharmacokinetics values comparing the results from female and male subjects. Mean ±SD

7. Protocol NP 15580, A single center open label, parallel design, 12 young and 12 elderly healthy subjects to assess the effect of age on Ro 25-8310 pharmacokinetics following subcutaneous administration. RR N-181144. April 7, 2000.

The purpose of this study was to assess the effects of age on the pharmacokinetics and pharmacodynamics of PEG-IFN (Ro 25-8310) after sc administration in healthy, male subjects. Twenty-four subjects were enrolled in this study of which 12 were between the ages of 18 to 25 and 12 were greater than 60 years of age. In the younger aged group the mean age was 22 years of age with a range of 19 to 25. In the older aged group the mean was 66.7 years of age with a range of 62 to 77. All subjects were male Caucasians.

Subjects were given a single dose of 180 ug in 1 ml sc in an open-label, single-center, non-randomized design. Lot C190447 was used in the study. Besides the serum levels of PEG-IFN, 2',5'-OAS activity was measured as a pharmacodynamic parameter. During the course of the study, safety endpoints were determined. Pharmacokinetic endpoints were reported as AUC, Cmax, Tmax, terminal elimination t1/2, apparent volume of distribution associated with the terminal phase and apparent total body clearance. Pharmacokinetic endpoints were computed with standard techniques and the slope of the terminal log-linear phase of the serum profile was determined by a least squares linear regression techniques from which t1/2 was calculated by dividing 0.693 by the slope.

Blood samples were drawn over 336 hours. Blood samples were collected predose (0 hours), 3, 6, 12, 24, 48, 72, 96, 120, 168, 264 and 336 hours post-dosing. Serum concentrations of PEG-IFN were determined using a ----------. 2',5'-OAS activity was determined using a ---------.

After sc administration, absorption appeared to be delayed in the older aged group; furthermore, peak levels of PEG-IFN were observed to be lower but more potent in the older group of individuals. Thus, their AUCs were higher. This finding is consistent with a lower measured clearance in the older group. In spite of a higher AUC level in the older group, maximal OAS activity was lower. The pharmacokinetic and pharmacodynamic data are summarized in the tables below. The pharmacokinetic data were not evaluated for all subjects. For subjects 21678/0003 and 21678/0006 in the younger group and subject 21678/0011 in the older group exhibited serum levels greater than 1 ng/ml and were excluded from their analysis.

Pharmacokinetic endpoint	Subjects 18 to 25 years (N=10)	Subjects >60 years (N=11)
Tmax, h	82 ± 23 (48 to 120)	115 ± 36 (72 to 169)
Cmax, ng/ml	10.3 ± 4 (2.4 to 14.2)	9.1 ± 3.7 (2.4 to 17.9)
AUC 0-336h, ng-h/ml	1471 ± 868 (363 to 3089)	1810 ± 938 (363 to 3306)
T1/2, h	61 37 ± (15 to 135)	110 ± 73 (15 to 218)
Vd, L	13 ± 11 (2 to 42)	14 ± 7 (2 to 30)
Cl, ml/h	183 ± 145 (45 to 478)	132 ± 122 (45 to 359)

Table of pharmacokinetic endpoints comparing younger and older individuals. Mean ± SD and range are cited.

Pharmacodynamic endpoint	Subjects 18 to 25 years (N=12)	Subjects >60 years (N=12)
Tmax, h	68 ± 23 (24 to 97)	130 ± 73 (24 to 337)
OASmax, nM/h	2.1 ± 0.8 (0.9 to 3.7)	1.5 ± 6 (1.0 to 3.2)

Table of pharmacodynamic endpoints comparing younger and older individuals. Mean ± SD and range are cited.

A comparison of pharmacokinetic and pharmacodynamic endpoints indicates that in spite of an increase in exposure to PEG-IFN in terms of AUC, biological activity in terms of 2',5'-OAS activity is lower in individuals >60 years of age and as compared to individuals between 18 to 25 years of age.

The most frequently observed adverse event for both groups was mild rash around the site of injection. Additionally other adverse events occurred including influenza-like illness, headache, dizziness, back pain, myalgia and fever. With the exception of influenza-like illness, and depressed WBC count the adverse event rate was approximately the same in the two aged groups. The incidence of influenza-like illness was higher among younger subjects with a value of 25% vs that of older individuals with an event rate of 8%. Depressed WBC counts occurred in 33% of the younger vs 8% of the older subjects.

In anticipation of febrile responses, two 500 mg tablets of paracetamol could be administered at 2, 6, and 10 hours post-dosing and subsequently required, but not exceeding 4 g in any 24 hour period.

8. Protocol NP15579, Pharmacokinetic study of Ro 25-8310 in patients with impaired renal function and healthy subjects following subcutaneous administration. RR N-181145. March 17, 2000.

The purpose of this study was to assess the effect of renal impairment on the pharmacokinetics and pharmacodynamics of PEG-IFN (Ro 25-8310) after sc injection. Lots ---------- (90 ug) was used for the study. The study was designed as a nonrandomized, single-center, single dose study in healthy subjects and patients with chronic renal impairment. Group 1 was composed of healthy subjects with no impaired renal function. Group 2 of individuals had no to minimal renal impairment that was defined as creatinine clearance <100 ml/min but >80 ml/min. Group 3 was defined as individuals having a creatinine clearance between < or equal to 80 ml/min but >60 ml/min. Groups 4 and 5 were composed of patients with increasingly severe renal impairment. For group 4 creatinine clearance was < or equal to 60 ml/min and >40 ml/min. Lastly, individuals in group 5 exhibited creatinine clearance < or equal to 40 ml/min but >20 ml/min.. Each group was composed of 6 individuals given a dose of 90 ug of PEG-IFN. The study population was composed of males with a mean age of 38.8 years.

Pharmacokinetics were reported as AUC, Cmax, Tmax, terminal elimination t1/2, apparent volume of distribution associated with the terminal phase and apparent total body clearance. Pharmacokinetic endpoints were computed with standard techniques and the slope of the terminal log-linear phase of the serum profile was determined by a least squares linear regression technique from which t1/2 was calculated by dividing 0.693 by the slope. The serum levels of PEG-IFN were determined using a ----------. 2',5'-OAS activity was determined using a--------.

Because of the anticipated febrile response to PEG-IFN, paracetamol was given as a pretreatment at 2, 6, and 10 hours post-dosing and as required. The protocol was amended to collect additional blood samples for groups 4 and 5 at 384, 432 and 504 hours after dosing. Due to additional sampling times for groups 4 and 5, their calculated t1/2's are not comparable to those of groups 1, 2 and 3.

The absorption, distribution and clearance of PEG-IFN was not significantly different between groups of subjects with varying degrees of renal failure. However, clearance does appear to be negatively influenced by renal function in spite of lack of change in AUC. This may have resulted from the differences in the

number and timing of serum samples used in the pharmacokinetic analysis as well as the small number of subjects in group 5. The pharmacokinetic results are cited in the table below.

Pharmacokinetic endpoints	Group 1, N=5	Group 2, N=4	Group 3, N=5	Group 4, N=6	Group 5, N=3
Tmax, h	81± 36	102 ± 23	115 ± 36	96 ± 22	104 ± 14
Cmax, ng/ml	6 ± 2	5 ± 2	5 ± 2	5 ± 0.3	5 ± 2
AUC336, ng-h/ml	1001 ± 681	773 ± 418	820 ± 313	857 ± 183	950 ± 219
T1/2, h	76 ± 23	69 ± 44	103 ± 36	107 ± 40	117 ± 68
Vd, ml/h	11± 4	12 ± 6	15 ± 5	13 ± 3	13 ± 6
Cl, ml/h	118 ± 68	158 ± 136	107 ± 53	95 ± 35	80 ± 22

Table of pharmacokinetic endpoints in subjects with varying degrees of renal impairment. Mean ± SD.

As regards PEG-IFN induced OAS activity, individuals in group 5 experienced a substantially reduced response as compared to the other groups. The pharmacodynamic results are cited in the table below.

Pharmacodynamic endpoints	Group 1, N=6	Group 2, N=6	Group 3, N=6	Group 4, N=6	Group 5, N=6
Tmax, h	144 ± 95	92 ± 44	108 ± 50	136 ± 39	196 ± 115
OASmax, nM/h	2.3 ± 0.9	2.8 ± 1	2.4 ± 1.1	2.9 ± 1.6	1.2 ± 0.3

Table of pharmacodynamic measures related to OAS activity in subjects with renal impairment. Mean ± SD.

The overall incidence of adverse effects was higher in subjects with greater renal impairment although the spectrum of adverse events was similar across all groups. The overall incidence of adverse effects by group are as follows: 50% - groups 1 and 2, 83% group 3, 100% group 4 and 67% group 5.

9. Protocol JP 15722, Phase I clinical study of Ro 25-8310 (PEG-interferon alfa-2a) in healthy volunteers. N-181609. April 10, 2000.

The objective of this study was to determine the pharmacokinetics, pharmacodyanmics and safety of PEG-IFN after a single sc dose in healthy male, Japanese subjects between the ages of 20 to 35 years. Lot numbers ------ also designated as ---------- for the 90 ug/ml, ---------also designated ----------for 180 ug/ml and --------- also designated -------- for 270 ug/ml dosages were used in the study. The study was designed with escalating doses of 90, 180 or 270 ug (N=12/dose) of Ro 25-8310. The study incorporated an open-label, nonrandomized design within a single center. The site of injection was the forearm. Blood sampling for pharmacokinetics and pharmacodynamics occurred at before dosing,(0), 8, 24, 48, 72, 84, 96, 120, 144, 168, 192, 264 and 336 hours after injection.

Serum levels of PEG-IFN were used to determine various pharmacokinetic endpoints using standard methods. These endpoints included the following: Cmax, Tmax, AUC, t1/2 associated with the terminal phase of elimination, apparent volume of distribution associated with the terminal elimination phase and apparent clearance. . Pharmacokinetic endpoints were computed with standard techniques and the slope of the terminal log-linear phase of the serum profile was determined by a least squares linear regression from which t1/2 was calculated by dividing 0.693 by the slope.

Serum concentrations of PEG-IFN were determined using a ----------. 2',5'-OAS activity was determined using a -------. Pharmacodyanmics were determined through an analysis of 2',5'-OAS activity. Safety was also assessed.
For individual pharmacokinetic profiles, Tmax occurred between 48 to 72 hours after dosing for all doses. Additionally, Cmax and AUC were proportional to dose and no change in t1/2 was observed. Clearance appeared to increase and subsequently decrease; however, this finding was not statistically significant. When creatinine clearance was used to adjust AUCinf/Dose, values were found to be more consistent with each other. The results of the study are summarized below.

Pharmacokinetic endpoint	90 ug, N=11	180 ug, N=11	270 ug, N=12
Tmax, h	72 ± 18	71± 37	73 ± 41

Cmax, ng/ml	7 ± 3	11± 4	20 ± 9
T1/2, h	40 ± 16 (1)	32 ± 31	43 ± 28
Clearance	126 ± 102 (1)	212 ± 197	110 ± 64
AUC 0 to 336 h, ng-h/ml	858 ± 453	1380 ± 884	2920 ± 1400

Table of pharmacokinetic endpoints for doses of 90, 180 and 270 ug in Japanese males, Mean ± SD (1).N=10

Dose, ug, N=12/group	2',5'-OAS activity
90	5.5 ± 15
180	64 ±17
270	84 ± 54

Table of OAS activity following various doses of PEG-IFN in Japanese men. Mean ± SD.

The most frequently observed adverse event for both groups was mild rash around the site of injection. Additionally other adverse events occurred including influenza-like illness, headache, dizziness, back pain, myalgia and fever. Mild dose dependent decreases in WBC were observed (incidence of 50%, 67% and 67% in accordance with dose), neutrophils also decreased in the two highest groups tested with an incidence of 17% in the 180 ug and 50% in 270 ug groups. Platelets were transiently lowered with an incidence of 25%, 33% and 75% in accordance with increasing doses.

10. Protocol NV 15489, A phase II, open-label randomized, multicenter, ascending-dose study, evaluating the safety and efficacy of peg-interferon alfa-2a vs interferon alfa-2a in the treatment of noncirrhotic patient with chronic hepatitis C. N-181405. February 14, 2000

The primary purpose of the study was to compare the safety, tolerability and efficacy of 4 doses of PEG-IFN when injected sc once weekly with that of 3 MIU of IFN injected iv 3 times weekly. The treatment period for this study was 48 weeks, and was followed by 24 weeks without treatment to determine the nature of any sustained response. In addition to the primary objective, the pharmacokinetics of PEG-IFN in patients with chronic hepatitis C without cirrhosis was investigated. Using a randomized, open-label, ascending dose design, 4 doses sc of PEG-IFN (45, 90, 180 and 270 ug) were studied and compared to a dose 3 MIU of IFN tiw given iv.

Samples of blood for pharmacokinetic analysis were taken during the course of the study. On day 1 of week 1, samples were taken pre-dosing (0), 8, 24, 48, 72, 84, 96, 120, 144 and 168 hours after dosing. At 4 week intervals from week 4 to week 44, blood samples were taken immediately before dosing to determine trough levels. Beginning the 48th week, blood was taken pre-dose and 8, 24, 72, 84, 96, 120, 144, 168, and 264 hours after injection of PEG-IFN. Blood samples were taken from 43 patients. In the 45 ug dosing group were 7 patients; in the 90 ug group were 11 patients; in the 180 ug group 11 patients and in the 270 ug group were 14 patients. Serum concentrations of PEG-IFN were determined with a quantitative ---------. Serum for anti-interferon antibodies was collected 1 month after the last dose. Patients were considered evaluable for the development of anti-interferon antibodies if they were treated for 60 days, if they demonstrated no pretreatment antibodies and pre- and post-treatment data were available. Evaluable patients were scored as anti-body positive if their post-treatment serum samples were more than 100 INU/ml as measured by a bioassay. (INU = interferon neutralizing units) Seven patients developed neutralizing antibodies of interferon. Five patients (4.4%) were treated with PEG-IFN and 2 patients (7.2%) with IFN.

Serum levels of PEG-IFN were used to determine various pharmacokinetic endpoints using standard methods. These endpoints included the following: Cmax, Tmax, AUC, t1/2 associated with the terminal phase of elimination, apparent volume of distribution associated with the terminal elimination phase and apparent clearance. Pharmacokinetic endpoints were computed with standard techniques and the slope of the terminal log-linear phase of the serum profile was determined by a least squares linear regression technique from which t1/2 was calculated by dividing 0.693 by the slope.

Average steady-state trough concentrations (Css, min) were determined from selected trough values. Only samples from patients receiving the same 4 previous doses and were collected between 144 and 192 hours after the last preceding dose. The average steady-state concentrations (Css, average) were computed using the following formula: (Css, max – Css, min)/ln (Css, max/Css, min) where Css, max was the observed maximum concentration after week 48.

The material used in this study for PEG-IFN (Ro 25-8310) was lot ---------- (45 ug),---- ------- (90 ug), ---------- and ------ (180 ug) and --------- (270 ug). The volume of injection was 1 ml for all doses of PEG-IFN.

The following pharmacokinetic endpoints were determined: Cmax, Tmax, t1/2, AUC to 168 hours, apparent total body clearance (Cl), apparent volume of distribution associated with the terminal phase (Vd) and steady-state serum concentrations (Css). The pharmacokinetic findings for single and multiple doses are summarized in the tables below. The pharmacokinetics of PEG-IFN in patients with chronic hepatitis C appear similar to those in healthy subjects. Exposure to PEG-IFN is proportional to dose and drug accumulation occurred in a predictable manner. Steady-state serum levels were achieved after 6 to 8 weeks of multiple dosing. The peak-to-trough ratio after multiple doses of PEG-IFN was between 1.5 to 2.0.

Pharmacokinetc endpoints	45 ug, N=3	90 ug, N=9	180 ug, N=8	270 ug, N=7
Tmax, h	72 ± 63 (24 to 143)	62 ± 27 (24 to 97)	85 ± 40 (24 to 144)	79 ± 26 (47 to 120)
Cmax, ng/ml	5 ± 4 (2 to 9)	10 ± 4 (4 to 16)	9 ± 5 (2 to 17)	19 ± 7 (11 to 29)
AUC168, ug-h/ml	.5 ± 4 (.2 to 1)	1.2 ±.6 (.5 to 2)	1.2 ±.7 (.1 to 2)	2.3 ±.9 (1.4 to 3.7)
T1/2, h	61 ± 24 (36 to 84)	71 ± 44 (23 to 128)	115 ± 104 (37 to 341)	168 ± 70 (71 to 251
Vd, l	8 ± 4 (4 to 12)	6 ± 5 (2 to 18)	22 ± 23 (8 to 73)	12 ± 5 (7 to 20)
Cl, ml/h	115 ± 105 (32 to 233)	74 ±54 (28 to 191)	291 ± 483 (18 to 1372)	55 ± 22 (33 to 88)

Table of pharmacokinetics after a single dose of PEG-IFN. Mean ± SD with range indicated.

Pharmacokinetic endpoints	45 ug, N=4	90 ug, N=9	180 ug, N=6	270 ug, N=9
Tmax, h	38 ± 28 (8 to 72)	67 ± 23 (25 to 96)	75 ± 48 (8 to 21)	69 ± 48 (0 to 167)
Css, max, ng/ml	7 ± 4 (1 to 12)	19 ± 12 (9 to 48)	29 ± 10 (18 to 45)	46 ± 17 (14 to 71)
Css, min, ng/ml	4 ± 3 (.2 to 7.6)	13 ± 10 (2 to 36)	15 ± 9 (.5 to 28)	31± 10 (19 to 52)
AUC168, ug-h/ml	.9 ± 7 (.08 to 1.8)	2.6 ± 1.5 (1.1 to 6.3)	4.1 ± 1.5 (2 to 6)	6 ± 2 (2 to 9)
T1/2, h	102 ± 59 (48 to 156)	187 ± 102 (42 to 366)	176 ± 119 (35 to 323)	164 ± 55 (95 to 23
Vd, l	15 ± 16 (4 to 39)	11 ± 10 (3 to 35)	11± 8 (2 to 20)	11± 6 (4 to 24)
Cl, ml/h	174 ± 262 (25 to 566)	43 ± 20 (14 to 78)	45 ± 26 (18 to 87)	51± 38 (30 to 142
Css, average, ng/ml	5 ± 4 (1 to 10)	16 ± 11 (5 to 42)	22 ± 8 (9 to 29)	43 ± 9 (34 to 59)

Table of pharmacokinetics after multiple doses of PEG-IFN. Mean ± SD with range indicated.

Accumulation indices	45, ug N=4	90 ug, N=9	180 ug, N=8	270 ug, N=7
Based on AUC168	1.8 to 1.9	1.2 to 2.9	1.9 to 9.2	2.4 to 2.5
Based on Cmax	1.4 to 1.5	1.1 to 6.1	1.8 to 6.1	2.3 to 4.2

Table of accumulation indices presented as the range of values.

11. Protocol NV 15495, A phase II/III Open-label, randomized multicenter, parallel-group study evaluating the safety and efficacy of peg-interferon a-2a (Ro 25-8310) vs Roferon-A in the treatment of patients with chronic hepatitis C with cirrhosis. RR N-181406. February 29, 2000

The primary purpose of the study was to compare the safety, tolerability and efficacy of 2 doses of PEG-IFN (90 and 180 ug) when injected sc once weekly with that of 3 MIU of IFN injected sc 3 times weekly. The treatment period for this study was 48 weeks, and was followed by 24 weeks without treatment to determine the nature of any sustained response. In addition to the primary objective, the pharmacokinetics of PEG-IFN in patients with chronic hepatitis C with cirrhosis or transitional cirrhosis was investigated after single and repeated doses. Using a randomized, open-label, parallel dose design, 2 doses sc of PEG-IFN (90 and 180 ug) were studied and compared to a dose 3 MIU of IFN tiw sc.

Samples of blood for pharmacokinetic analysis were taken during the course of the study. On day 1 of week 1, samples were taken pre-dosing (0), 8, 24, 48, 72, 84, 96, 120, 144 and 168 hours after dosing. At 4 week

intervals from week 4 to week 44, blood samples were taken immediately before dosing to determine trough levels. Beginning the 48th week, blood was taken pre-dose and 8, 24, 48, 72, 96, 120, 144, 168 and 264 hours after injection of PEG-IFN. Blood samples were taken for pharmacokinetic analysis from twelve patients (7 subjects for 90 ug and 5 for the 180 ug doses of PEG-IFN). Serum concentrations of PEG-IFN were determined with a quantitative ----------. Serum for anti-interferon antibodies were collected prior to dosing and 2 months after the last dose. Patients were considered evaluable for the development of anti-interferon antibodies if they were treated for 60 days, if they demonstrated no pretreatment antibodies and pre- and post-treatment data were available. Evaluable patients were scored as anti-body positive if their post-treatment serum samples were more than 100 INU/ml as measured by a bioassay. (INU = interferon neutralizing units). In the group given IFN 11% developed anti-interferon antibodies as compared to 4% in the group given 90 ug PEG-IFN or 4% in the group given 180 ug of PEG-IFN.

The material used in this study for PEG-IFN (Ro 25-8310) were lots [
] (180 ug). The volume of injection was 1 ml for all doses of PEG-IFN.

Serum levels of PEG-IFN were used to determine various pharmacokinetic endpoints using standard methods. These endpoints included the following: Cmax, Tmax, AUC, t1/2 associated with the terminal phase of elimination, apparent volume of distribution associated with the terminal elimination phase and apparent clearance. Pharmacokinetic endpoints were computed with standard techniques and the slope of the terminal log-linear phase of the serum profile was determined by a least squares linear regression technique from which t1/2 was calculated by dividing 0.693 by the slope.

Average steady-state trough concentrations (Css, min) were determined from selected trough values. The average steady-state concentrations (Css, average) were computed using the following formula: (Css, max – Css, min)/ln (Css, max/Css, min) where Css, max was the observed maximum concentration after week 48. The following pharmacokinetic endpoints were determined: Cmax, Tmax, t1/2, AUC168, apparent total body clearance (Cl), apparent volume of distribution associated with the terminal phase (Vd) and steady-state serum concentrations (Css). The pharmacokinetics of PEG-IFN in patients with chronic hepatitis C with cirrhosis or transition to cirrhosis appears similar to those in healthy subjects. Peak serum levels were achieved between 55 and 75 hours after dosing. Exposure to PEG-IFN is proportional to dose and drug accumulation occurred in a predictable manner. Steady-state serum levels were achieved after 6 to 8 weeks of multiple dosing. The accumulation was typically 2 to 3 fold over the treatment interval. The peak-to-trough ratio for the PEG-IFN was 1.6 to 1.7 for both doses after week 48. The pharmacokinetic findings for single and multiple doses are summarized in the tables below.

Pharmacokinetic endpoints	90 ug, N=7	180 ug, N=5
Tmax, h	72 ± 39 (24 to 144)	57 ± 40 (24 to 120)
Cmax, ng/ml	5 ± 3 (1 to 9)	9 ± 6 (3 to 19)
AUC to 168h, ug-h/ml	.6 ±.3 (.1 to 1)	1.2 ± .9 (.02 to 2.7)
T1/2, h	62 ±15 (42 to 86)	51 ± 35 (21 to 89)
Vd, l	19 ± 27 (6 to 75)	18 ± 21 (5 to 49)
Cl, ml/h	204 ± 252 (54 to 715)	223 ±118 (113 to 382)

Table of pharmacokinetic values for patients given PEG-IFN with hepatitis C and cirrhosis given a single dose. Values are means ±SD and ranges. One patient had a clearance of 715 ml/h; all other exhibited clearances between 54 and 144 ml/h.

Pharmacokinetic endpoint	90 ug PEG-IFN, N=5
Tmax, h	50± 30 (9 to 72)
Css-max, ng/ml	11± 1.5 (9 to 13)
Css-min, ng/ml	7 ± 2 (6 to 11)
Peak to trough ratio	1.5 ±.4 (1 to 2)
AUC to 168h, ug-h/ml	1.5 ±.3 (1.3 to 1.9)
T1/2, h	121± 57 (71 to 198)

Vd, l	10 ± 4 (7 to 16)
Clearance, ml/h	61 ± 12 (47 to 71)
Css-average, ng/ml	9 ± 2 (8 to 11)

Table of pharmacokinetics for 90 ug dose of PEG-IFN in patients with hepatitis and cirrhosis after repeated dosing. Values are cited as means ± SD and the ranges. As only 2 patients given 180 ug had evaluable data, their results are not presented.

12. Protocol NV 15496, A phase III open-label, randomized, multicenter, parallel dose efficacy and safety study comparing PEG-IFN alfa-2a to a standard regimen of Roferon-A in the treatment of patients with chronic hepatitis C. RR N-181408. April 10, 2000.

The primary purpose of the study was to compare the safety, tolerability and efficacy of 2 doses of PEG-IFN when injected sc once weekly with that of 3 MIU of IFN injected iv 3 times weekly. The treatment period for this study was 48 weeks, and was followed by 24 weeks without treatment to determine the nature of any sustained response. In addition to the primary objective, the pharmacokinetics of PEG-IFN in patients with chronic hepatitis C without cirrhosis was investigated. Using a randomized, open-label, parallel dose design, 2 doses sc of PEG-IFN (135 and 180 ug) were studied and compared to a dose 3 MIU of IFN tiw iv.

Samples of blood for pharmacokinetic analysis were taken during the course of the study. On day 1 of week 1, samples were taken pre-dosing (0), 8, 24, 48, 72, 84, 96, 120, 144 and 168 hours after dosing. At 4 week intervals from week 4 to week 44, blood samples were taken immediately before dosing to determine trough levels. Beginning the 48th week, blood was taken pre-dose and 8, 24, 48, 72, 96, 120, 144, 168 and 264 hours after injection of PEG-IFN. Blood samples were taken for pharmacokinetic analysis from 39 patients (20 subject for 135 ug and 19 for the 180 ug dose of PEG-IFN). Serum concentrations of PEG-IFN were determined with a quantitative ----------. Serum for anti-interferon antibodies were collected prior to dosing and 2 months after the last dose. Patients were considered evaluable for the development of anti-interferon antibodies if they were treated for 60 days, if they demonstrated no pretreatment antibodies and pre- and post-treatment data were available. Evaluable patients were scored as anti-body positive if their post-treatment serum samples were more than 100 INU/ml as measured by a bioassay. (INU = interferon neutralizing units). In the group given IFN 16% developed anti-interferon antibodies as compared to 4% in the group given 135 ug PEG-IFN or 1% in the group given 180 ug of PEG-IFN.

Serum levels of PEG-IFN were used to determine various pharmacokinetic endpoints using standard methods. These endpoints included the following: Cmax, Tmax, AUC, t1/2 associated with the terminal phase of elimination, apparent volume of distribution associated with the terminal elimination phase and apparent clearance. Pharmacokinetic endpoints were computed with standard techniques and the slope of the terminal log-linear phase of the serum profile was determined by a least squares linear regression technique from which t1/2 was calculated by dividing 0.693 by the slope.

Average steady-state trough concentrations (Css, min) were determined from selected trough values. The average steady-state concentrations (Css, average) were computed using the following formula: (Css, max – Css, min)/ln (Css, max/Css, min) where Css, max was the observed maximum concentration after week 48.

The material used in this study for PEG-IFN (Ro 25-8310) were lots[----------(180 ug). The volume of injection was 1 ml for all doses of PEG-IFN.

The following pharmacokinetic endpoints were determined: Cmax, Tmax, t1/2, AUC168, apparent total body clearance (Cl), apparent volume of distribution associated with the terminal phase (Vd) and steady-state serum concentrations (Css). The pharmacokinetic findings for single and multiple doses are summarized in the tables below. The pharmacokinetics of PEG-IFN in patients with chronic hepatitis C appear similar to those in healthy subjects. Exposure to PEG-IFN is proportional to dose and drug accumulation occurred in a predictable manner. The accumulation was 2 to 3 fold over the treatment interval Steady-state serum levels

were achieved after 6 to 8 weeks of multiple dosing.. The peak-to-trough ratio for the PEG-IFN was 1.6 to 1.7 for both doses after week 48.

Pharmacokinetic endpoints	135 ug, N=19	180 ug, N=14)
Tmax, h	86 ± 41 (47 to 167)	80 ± 28 (23 to 119)
Cmax, ng/ml	10 ± 4 (3 to 20)	15 ± 4 (7 to 22)
AUC168, ug-h/ml	1.1 ± .5 (.4 to 2.1)	1.8 ± .6 (.8 to 2.6)
T1/2, h	81 ± 61 (12 – 277)	83 ± 59 (8 to 178)
Vd, l	8 ± 3 (5 to 17)	7 ± 4 (1 to 20)
Cl, ml/h	110 ± 86 (40 346)	83 ± 50 (33 to 186)

Table of pharmacokinetics after a single dose of PEG-IFN. Values are presented as the mean ±SD and range.

Pharmacokinetic endpoints	135 ug, N=16	180 ug, N=16
Tmax, h	60 ± 51 (8 to 194)	45 ± 36 (0 to 97)
Css, max, ng/ml	20 ± 7 (9 to 35)	26 ± 9 (10 to 40)
Css, min (ng/ml)	14 ± 7 (5 to 32)	16 ± 6 (4 to 28)
AUC168, ug-h/ml	2.7 ± 1 (1.1 to 4.9)	3.3 ± 1 (1.2 to 4.8)
T1/2, h	137 ±5 0 (60 to 235)	221 ± 143 (92 to 594)
Vd, l	11± 6 (5 to 28)	19 ± 13 (5 to 48)
Cl, ml/h	57 ± 24 (27 to 118)	60 ± 25 (37 to 142)
Css, average, ng/ml	17± 6 (7 to 29)	21 ± 6.3 (6 to 31)

Table of pharmacokinetics after multiple doses of PEG-IFN. Values are presented as the mean SD and range.

13. Protocol NV 15497, A phase III open-label, randomized, multicenter, parallel-group efficacy and safety study comparing PEG-IFN alfa-2a to an induction regimen of Roferon-A in the treatment of patients with chronic hepatitis C. RR N-181410. 2000.

The primary purpose of the study was to compare the safety, tolerability and efficacy of a 180 ug doses of PEG-IFN when injected sc once weekly in comparison to a regimen of IFN. The treatment period for this study was 48 weeks, and was followed by 24 weeks without treatment to determine the nature of any sustained response. In addition to the primary objective, the pharmacokinetics of PEG-IFN in patients with chronic hepatitis C was investigated. The percentage of patients with cirrhosis or transition to cirrhosis was set as a maximum of 20%. Adverse events as reported in this study were similar to those recognized for IFN and PEG-IFN.

Samples of blood for pharmacokinetic analysis were taken during the course of the study. On day 1 of week 1, samples were taken pre-dosing, 8, 24, 48, 72, 84, 96, 120, 144 and 168 hours after dosing. At 4 week intervals from week 4 to week 44, blood samples were taken immediately before dosing to determine trough levels. Beginning the 48th week, blood was taken pre-dose and 8, 24, 48, 72, 96, 120, 144, 168 and 264 hours after injection of PEG-IFN. Serum concentrations of PEG-IFN were determined with a quantitative sandwich immunoassay. Serum for anti-interferon antibodies were collected prior to dosing and week 56 of the study (about 2 months after the last dose). Patients were considered evaluable for the development of anti-interferon antibodies if they were treated for 60 days, if they demonstrated no pretreatment antibodies and pre- and post-treatment data were available. Evaluable patients were scored as anti-body positive if their post-treatment serum samples were equal to or more than 100 INU/ml as measured by a bioassay (INU = interferon neutralizing units). In the group given IFN 17% developed anti-interferon antibodies as compared to 2% in the group given 135 ug PEG-IFN or 1% in the group given 180 ug of PEG-IFN.

Lots ---------- of PEG-IFN (Ro 25-8310) were used in this study.

Serum levels of PEG-IFN were used to determine various pharmacokinetic endpoints using standard methods. These endpoints included the following: Cmax, Tmax, AUC to 168 h, t1/2 associated with the terminal phase of elimination, apparent volume of distribution associated with the terminal elimination phase and apparent clearance. Pharmacokinetic endpoints were computed with standard techniques and the slope

of the terminal log-linear phase of the serum profile was determined by a least squares linear regression technique from which t1/2 was calculated by dividing 0.693 by the slope. An analysis to characterize the pharmacokinetics of PEG-IFN over the entire 48 weeks was performed.

Average steady-state trough concentrations (Css, min) were determined from selected trough values. Only samples from patients who received the same 4 previous doses and were collected between 144 and 192 hours after the last preceding dose were considered steady-state trough concentrations. Only trough samples that were within 80% to 125% of the mean trough concentrations were included in the estimation of Css, min to ensure that steady-state was achieved. The average steady-state concentrations (Css, average) were computed using the following formula: (Css, max – Css, min)/ln (Css, max/Css, min) where Css, max was the observed maximum concentration after week 48.

The pharmacokinetics of PEG-IFN were measured after a single dose in sixteen patients given a dose of 180 ug sc. PEG-IFN was absorbed with a Tmax of approximately 100 h and half-life averaged 137 h in the patient population. Accumulation of serum levels occurred over the initial 5 to 7 weeks of treatment and averaged 2.4 if based on AUC to 168 h or 2.9 if based on Cmax. In one patient (20982/2885), neutralizing antibodies were found with a titer of 282 INU/ml, but no alterations in the patient's pharmacokinetics were reported. The pharmacokinetics of PEG-IFN in patients with chronic hepatitis C appear similar to those in healthy subjects. Exposure to PEG-IFN is proportional to dose and drug accumulation occurred in a predictable manner. The pharmacokinetic findings for single and multiple doses are summarized in the tables below.

Pharmacokinetic endpoints	PEG-IFN, 180 ug
Tmax, h	105 ± 62 (29 to 226)
Cmax, ng/ml	11 ± 4 (5 to 21)
AUC168, ug-h/ml	1.4 ± .6 (.5 to 2.8)
T1/2, h	137 ± 62 (42 to 231)
Vd, l	10 ± 5 (5 to 25)
Cl, ml/h	56 ± 17 (27 to 90)

Table of pharmacokinetics for PEG-IFN after a single dose in patients with chronic hepatitis C. Values are the means ±SD as well as the range.

The pharmacokinetics of PEG-IFN were measured after multiple dose of PEG-IFN in sixteen patients given a dose of 180 ug over the 48 week treatment period.

Pharmacokinetic endpoints	PEG-IFN, 180 ug
Css, min, ng/ml	17 ± 7 (7 to 33)
Css, max, ng/ml	24 ± 9 (15 to 49)
Css, average, ng/ml	21 ± 7 (11 to 40)
Peak to Trough ratio	1.4 ± .4 (1.1 to 2.4)
AUC168, ug-h/ml	3.6 ± 1.2 (2 to 6.9)
Cl, ml/min	56 ± 18 (26 to 90)

Table of pharmacokinetics for PEG-IFN after multiple doses. Values are means ± SD as wells as the range.

Pharmacokinetic basis for R	Accumulation indices (R)
AUC168	2.4 ±.7 (1.3 to 3.7)
Cmax	2.9 ± 1.3 (1.3 to 5.8)

Table of accumulation indices based on either AUC or Cmax. Values presented as means ± SD and range.

14. Protocol NP15581, Effect of Ro 25-8310 on cytochrome P450s (1A2, 2C9, 2C19, 2D6 and 3A4) in healthy subjects. RR N-181143 February 15, 2000

The purpose of this study was to investigate the influence of PEG-IFN (Ro 25-8310) on drug metabolism mediated by P450 cytochrome system in vivo in healthy subjects. Lot ---------- (180 ug./ml) was used as the source of PEG-IFN. This study was an open-label, single-center study that was conducted in healthy subjects known to be extensive 2D6 and 2C19 metabolizers. The study was divided into 3 parts: first

administration of probe drugs, second was PEG-IFN administration, and third was administration of probe drugs. Probe drugs were given orally and were as followed: theophylline (125 mg, probe for CYP1A2), tolbutaminde (500 mg, probe for 2C9), mephenytoin (100 mg, probe for 2C19), debrisoquine (10 mg, probe for 2D6) and dapsone (100 mg, probe for 3A4). On day 1, five probe drugs were administered to identify individuals who were extensive drug metabolizers. Of the 16 subjects who began the study, 15 proceeded on the parts 2 and 3. Serial blood samples were taken at pre-dose (0), 0.5, 1, 2, 4, 6, 8, 12, 24, 48, 72 and 120 hours after dosing. Urine samples were collected pre-dose and from 0 to 4 hours and 4 to 8 hours post-dosing. Individuals identified as 2D6 and 2C19 extensive metabolizers were enrolled in the study to investigate the interaction of PEG-IFN on drug metabolism. PEG-IFN was then administered sc once weekly 4 times prior to re-administration of the probe drugs. Pharmacokinetics of the probe drugs were used to assess the potential for drug-drug interactions. Blood and urine samples were taken at the same time points as were used previously. Adverse events were monitored throughout the study and did not reveal qualitative or quantitative changes in the incidence or intensity of PEG-IFN toxicities.
Pharmacokinetics were reported as AUC, Cmax, Tmax, terminal elimination t1/2, apparent volume of distribution associated with the terminal phase and apparent total body clearance. Pharmacokinetic endpoints were computed with standard techniques and the slope of the terminal log-linear phase of the serum profile was determined by a least squares linear regression technique from which t1/2 was calculated by dividing 0.693 by the slope.

The study population was solely composed of Caucasian males with an approximate age of 34 years. One subject withdraw for adverse effects due to PEG-IFN.

Because of the anticipated febrile response to PEG-IFN, paracetamol was given as a pretreatment at 2, 6, and 10 hours post-dosing and as required.

A statistically significant interaction between theophylline as a metabolic probe for CYP1A2 and PEG-IFN was found based on AUC measurement. The AUC extrapolated to infinity of oral theophylline was increased by 24% when compared to baseline values after multiple once-weekly dosing with PEG-IFN. Cmax values for theophyilline were slightly increased by exposure to PEG-IFN. T1/2 was were generally increased after exposure to PEG-IFN. In 9 of 14 subjects, oral clearance of theophyilline was decreased by 15% to 60% after PEG-IFN. A range of responses were observed to the change of theophyilline metabolism upon re-testing with the probe drugs. In fact, two individuals demonstrated a 26% to 50% increase in metabolism. The relative ratio of the geometric means of AUC extrapolated to infinity for PEG-IFN treated individuals relative to baseline was 1.24 with a 90% confidence interval of 1.05 to 1.47. No evidence of an interaction with CYP2C9, CYP2C19, CYP2D6, and CYP3A4 systems. No statistically significant differences were observed with tolbutamide, mephenytoin, debrisoqunine or dapsone pharmacokinetics or metabolism.

The pharmacokinetic changes for theophyilline are summarized in the table below.

Pharmacokinetic endpoint	Baseline, N=14	After PEG-IFN, N=14
AUCinf, ug-h/ml (1)	43.6	54.2
AUClast, ug-h/ml (1)	38.7	47.1
Cmax, ug/ml (1)	37.6	40.5
T1/2, h (2)	9 ± 4	13 ± 8
Clearance, L/h (2)	3 ± 0.9	2.5 ± 1.2
Vd, L (2)	35 ± 9	39 ± 10

Table of pharmacokinetics for theophyilline presented as geometric mean (1) or mean ± SD.

15. Protocol NP16569, A comparability study of Peginterferon alfa-2a (PEG-IFN) of clinical trial material and the commercial product administered subcutaneously via needle injection in healthy subjects. RRN-1007463 March 29, 2002.

The pharmacokinetic comparability of the investigational article (PEG-IFN) used during phase 3 (lot

----- was compared to material intended for marketing (lot ------) in an open-label study, single dose, randomized two-way crossover study. In this study healthy male and females subjects were given a SC injection of 180 ug/ml in the abdomen. The study consisted of 2 periods of 312 hours each that were separated by a washout period of 4 to 6 weeks. Blood samples were collected on days 1 through 14 of each study period; blood samples for pharmacokinetics were taken predose, 0.5, 1, 3, 5, 8, 12, 24, 48, 72, 84, 96, 120, 144, 168, 192, 216, 240, and 312 hours postdose. Samples for pharmacodynamics were taken predose, 8, 24, 48, 96, 120, 168, 312 hours postdose. Of the 160 subjects enrolled, 156 were dosed and 138 completed the crossover study. Pharmacodynamic markers and safety parameters were also studied. The pharmacokinetic parameters of PEG-IFN were determined through the use of model-independent approach. Pharmacokinetic comparability based on AUClast and Cmax were computed using a 90% confidence interval for the ratio of the means. The region considered as establishing pharmacokinetic comparability was 0.8 to 1.25. Pharmacokinetic values were adjusted for the actual dose injected. An initial assessment of pharmacokinetic comparability was made using 66 subjects. The findings from this group established pharmacokinetic comparability with the CI for Cmax being 0.93 to 1.08 and that for AUClast being 0.88 to 1.03. The results of the final study group is reported below:

	Cmax, ng/ml	AUClast, ng-h/ml
----------, N = 76	1.09	1850
Vial, N = 76	1.08	1941

Table of pharmacokinetic values for subjects completing the crossover study.

Ratio of A/B	1.01	0.95
90% confidence interval	0.93, 1.08	0.88, 1.03

Table of pharmacokinetic comparability for crossover study.

Assessment of pharmacodynamic measures as 2',5'-OAS were consistent with the findings based on pharamcokinetic comparability.

The overall assessment for safety did not reveal any differences based on whether clinical lots or commercial lots were used.

16. Protocol NR16081, A pharmacokinetic/pharmacodynamic ascending dose study to evaluate the safety of a single dose of Pegasys in end-stage renal disease patients undergoing hemodialysis. RR 1003975 April 1, 2002

The primary objective of this clinical study was to determine the pharmacokinetics of PEG-IFN in patient with end stage renal disease undergoing hemodialysis. The pharmacodynamics and safety of PEG-IFN were also assessed. Patients were given a single dose in an open-label, nonrandomized, parallel-group dose escalation study. Four single ascending doses of PEG-IFN (45, 90, 135, and 180 ug) were given to each group of 6 planned patients in the right side of the abdomen. Blood samples for pharmacokinetic evaluation was taken predose, 3, 6, 12, 24, 48, 72, 168, 240, 336, 456 and 600 postdose. Pharmacokinetic results are summarized below:

	45 ug	90	135	180
Cmax, ng/ml	3.8 (1.74)	8.9 (4.87)	13.1 (5.10)	18.1 (5.40)
AUC168, ng-h/ml	463 (204)	1024 (549)	1667 (708)	2215 (648)
T1/2, h	75 (13)	85 (28)	103 (31)	90 (22)
Cl, ml/h	67 (30)	69 (29)	61 (35)	50 (17)

Table of pharmacokinetic endpoints by noncomparental analysis.

A 25% to 45% reduction in total body clearance of PEG-IFN was observed in patients with end stage renal disease.

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