CENTER FOR DRUG EVALUATION AND RESEARCH

Application Number NDA 21-108

CLINICAL PHARMACOLOGY and BIOPHARMACEUTICS REVIEW(S)

MAY 1 2000

Clinical Pharmacology/Biopharmaceutics Review

NDA: 21-108 **SUBMISSION DATE: 9/1/99**

PRODUCT: 0.02% Tretinoin Emollient Cream (RENOVA®)

SPONSOR: Johnson and Johnson Consumer Companies, Inc.
Skillman, NJ 08558

REVIEWER: Tapash K. Ghosh, Ph.D.

Review of a NDA

I. BACKGROUND

Tretinoin (all-*trans*-retinoic acid) is an oxidation product in the physiological pathway of retinol metabolism, which executes retinol (vitamin A) - dependent functions, including embryonic development, the maintenance of epidermal differentiation, testicular function, and regulation and differentiation of many different cell types. Tretinoin is used in the US in topical preparations for the treatment of acne vulgaris at concentrations of 0.1%, 0.05%, 0.025% and 0.01%, and for the mitigation of fine wrinkles, mottled hyperpigmentation and tactile roughness resulting from chronic sun exposure at concentrations of 0.05%. As of the date of this application, tretinoin emollient cream 0.02% (TEC II) is not marketed anywhere in the world.

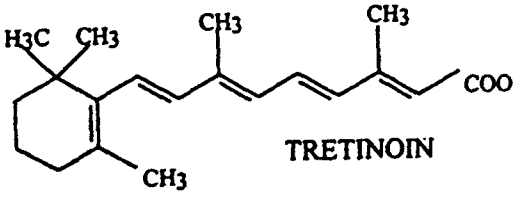

Fig 1: The Structure of All-*Trans*-Retinoic Acid

What is the purpose of this NDA?

In this application (NDA 21-108), the sponsor seeks approval of a 0.02% concentration of tretinoin for the treatment of photodamaged facial skin. The proposed clinical regimen of the new formulation is the application once daily of 250-500 mg of 0.02% cream (50-100 µg of tretinoin). The currently marketed RENOVA ® (tretinoin emollient cream or TEC) 0.05% was approved under NDA 19-963. This formulation is also referred to as TEC IA. RENOVA 0.02% refers to the tretinoin emollient cream formulation designated as TEC II. The TEC II formulation was derived from the currently marketed RETIN-A ® formulation, and was reportedly developed to overcome the observed freezing instability and viscosity reduction which occurred during manufacture and at accelerated temperatures. In addition, substitutions in composition were made to

increase storage and use stability, reduce comedogenic potential, and decrease facial stinging. As per the sponsor, the new formulation (TEC II) with the lower concentration should be better tolerated; this expectation is confirmed by comparison of cumulative irritation trials, which demonstrate greater irritation scores with RENOVA 0.05% as compared to RENOVA 0.02%. The incidence of skin and subcutaneous tissue adverse events was 29% in the pivotal trials of RENOVA 0.02% as compared with 86% in the pivotal trials of RENOVA 0.05% (NDA 19-963).The pivotal trials for RENOVA 0.05% were conducted in subjects from 30 to 50 years of age with mild to moderate photodamage. The pivotal trials for RENOVA 0.02% were conducted in subjects from 45-70 years of age who exhibited moderate to severe photodamage. This NDA (21-108) also provides support for extending eligibility to older patients and to patients with more severe photodamage at baseline.

II. RECOMMENDATION

Based on the results from the in-vivo studies, an insignificant increase in the endogenous level of tretinoin from its baseline (≈ 2 ng/mL) occurs from the topically applied 0.05% RETIN-A, RENOVA (TEC IA), and TEC II cream formulations in healthy volunteers. No pharmacokinetic study has been conducted with the proposed 0.02% TEC II formulation. The sponsor, however, conducted an in-vitro release study where the slopes for 0.05% and 0.02% tretinoin creams were 0.7562 $\mu g/h^{1/2}$ and 0.2713 $\mu g/h^{1/2}$ respectively. The results indicate that the release rate of tretinoin is proportional to its concentration in the formulation.

In view of all the in-vivo results obtained with 0.05% formulations coupled with the in-vitro release study, significant (i.e., detectable) systemic absorption is quite unlikely from the proposed formulation with 0.02% all-trans tretinoic acid. The application is acceptable from biopharmaceutics point of view.

TABLE OF CONTENTS

III. PHARMACOKINETICS

What do we know about Pharmacokinetics of Tretinoin?

Detailed analysis of the pharmacokinetic behavior of tretinoin in humans has been restricted by both low plasma concentrations reached after an oral dose and the absence of an intravenous form which would avoid the variability associated with oral drug administration. Consequently, most pharmacokinetic data are derived from studies following oral administration, and the estimates of apparent volume (Vd) and clearance are all corrected for the unknown fraction of the drug systemically available (Bioavailability, F). Long term tretinoin administration appears likely to lower peak concentrations and total systemic exposure (AUC_∞) in studies in mice, monkeys and human patients. Tretinoin pharmacokinetic parameters after single and multiple doses (which is much higher than 0.02% topical dose) in humans are summarized in Tables I and II respectively. Increased oxidation via the CYP enzyme system appears to account, at least in part, for the observed reduction in plasma drug concentration and development of resistance after multiple dosing (specially seen in patients with acute promyelocytic leukaemia, APL) (Table II).

Table I. Mean pharmacokinetic parameters (± SD or range where available) of tretinoin in humans after a single dose

Reference	No. of patients	Diagnosis	Dose (mg/m^2)	C_{max} (μg/L)	t_{max} (h)	AUC_∞ (μg/L • h)	CL/F ($L/h/m^2$)	$t_{½z}$ (min)
Lefebvre et al.[18]	15	APL	45	590 ± 700	1.9	630	71.4	39 ± 19
Muindi et al.[49]	13	APL	45	346 ± 266	2.2 ± 1.0	682 ± 500	65.9 ± 48.3	48 ± 6
	10		45			499 ± 200	90.1 ± 36.1	
Muindi et al.[50]	20	APL	45	279 ± 243	2.2 ± 1.0	603 ± 442	74.6 ± 54.7	48 ± 12
	47	Solid tumours	45	191 ± 137	3.3 ± 1.4	454 ± 419	99.1 ± 91.4	48 ± 18
Smith et al.[51]	2	Paediatric tumours	22.5		1.7[a]	387 ± 84	58.1 ± 12.6	58 (52-66)
	8	Paediatric tumours	30		2.0[a]	339 ± 141	88.5 ± 36.8	45 (36-64)
	8	Paediatric tumours	40		3.0[a]	1005 ± 630	39.8 ± 24.9	40 (27-77)
Lee et al.[52]	1	Solid tumours	60			3800	15.8	
	2	Solid tumours	175	1038 ± 541		2365 ± 163	74.0 ± 5.1	
	1	Solid tumours	200	1396		3570	56.0	
Castaigne et al.[53]	5	APL	25	1630		733	34.1	
	8	Paediatric tumours	40		3.0[a]	1005 ± 630	39.8 ± 24.9	40 (27-77)
Smith et al.[54]	3		17			168	101.2	
Lee et al.[55]	7	Solid tumours	45			1355 ± 1479 (median 1147)	33.2 ± 36.2	
Adamson et al.[56]	8	Kaposi's sarcoma	40	330 ± 60		725 ± 130	55.2 ± 9.9	
Regazzi (unpublished observations)	13	CML	40	219 ± 131	2.6 ± 0.9	568 ± 440	70.4 ± 54.5	66 ± 2
	3	MDS	22.5	75 ± 42	3.3 ± 2.3	190 ± 84	118.4 ± 52.1	54 ± 4
	8	APL	45	321 ± 204	2.6 ± 0.7	673 ± 313	66.8 ± 31.1	50 ± 10

a Median.

Abbreviations: APL = acute promyelocytic leukaemia; AUC_∞ = area under the concentration-time curve; C_{max} = maximum plasma concentration; CL/F = total body clearance divided by bioavailability; CML = chronic myelogenous leukaemia; MDS = myelodysplasticsyndrome; t_{max} = time to reach maximum drug concentration; $t_{½z}$ = elimination half-life associated with the terminal slope λ_z of the semilogarithmic concentration-time curve.

Ref Regazzi *et al.*, "Clinical Pharmacokinetics of Tretinoin", *Clin. Pharmacokinet.* 1997 May: 32(5) 382-402.

Table II. Change in the pharmacokinetic parameters of tretinoin in humans after multiple doses

Reference	No. of patients	Diagnosis	Treatment regimen	Daily dosage (mg/m^2)	Pharmacokinetic study			
					dose (mg/m^2)	C_{max} ($\mu g/L$)	AUC_{∞} ($\mu g/L \cdot h$)	AUC_{∞} reduction or increase vs day 1 (%)
Smith et al.[51]	7	Paediatric tumours	Continuous therapy	45-60-90	22.5-30-45			
day 1							447 ± 237	
day 28							<90 ± 150	↓80
Muindi et al.[49]	7	APL	Continuous therapy	45	45			
day 1						294 ± 89	537 ± 191	
after 2-6wk						138 ± 139	248 ± 135	↓54
Lee et al.[52]		Solid tumours	Continuous therapy					
day 1	1			60	60	742	3800	
after 10.5mo						293	1590	↓58
day 1	2			175	175	1038 ± 541	2365 ± 162	
after 2.1 ± 0.1mo						155 ± 18	800 ± 240	↓66
day 1	1			200	200	890	3570	
after 2.1mo						152	1396	↓61
Muindi et al.[50]		APL		45	45			
day 1	10						499 ± 200	
at relapse	6						244 ± 145	↓51
Adamson et al.[56]	8	Kaposi's sarcoma	7 days on/ 7 days off	40	40			
day 1						330 ± 60	725 ± 130	
day 7							90 ± 20	↓88
day 15 (day 1 wk 3)							885 ± 195	↑22
day 71 (day 1 wk 11)							640 ± 130	↓12
Lee et al.[55]		Solid tumours	14 days on/ 7 days off	90	45			
day 1	6						1355 ± 1479	
day 14	4						308 ± 476	↓77
Adamson et al.[57]	7	Refractory cancer	3 days on/ 4 days off	90	30			
day 1						330 ± 90	500 ± 75[a]	
day 3						60 ± 30	130 ± 120	↓74
day 29 (day 1 wk 4)						300 ± 120	570 ± 95	↑14
Toma et al.[58]	1	Kaposi's sarcoma	3 days on/ 4 days off	45	22.5			
day 1						55.7	91	
day 3						9.6	20	↓78
day 8 (day 1 wk 2)						53.6	115	↑26
day 22 (day 1 wk 4)						27.0	75	↓18
day 52 (day 3 wk 8)						52.3	62	↓32
Regazzi (unpublished observations)	13	CML	7 days on/ 7 days off	80	40			
day 1						219 ± 131	567 ± 440	
day 7						71 ± 58	192 ± 169	↓66
day 15 (day 1 wk 3)						203 ± 145	545 ± 395	↑4

Table II. Contd

Reference	No. of patients	Diagnosis	Treatment regimen	Daily dosage (mg/m^2)	Pharmacokinetic study			
					dose (mg/m^2)	C_{max} (µg/L)	AUC_{∞} (µg/L • h)	AUC_{∞} reduction or increase vs day 1 (%)
	6	APL	2wk on/ 10wk off	45	45			
day 1						321 ± 204	673 ± 313	
day 14						131 ± 94	245 ± 201	↓64

a Standard error.

Abbreviations and symbols: APL = acute promyelocytic leukaemia; $\%AUC_{\infty}$ = change in area under the concentration-time curve; C_{max} = maximum drug plasma concentration; CML = chronic myelogenous leukaemia; wk = week(s); ↓ indicates decrease; ↑ indicates increase.

Ref Regazzi *et al.*, "Clinical Pharmacokinetics of Tretinoin", *Clin. Pharmacokinet.* 1997 May: 32(5) 382-402.

IV. METABOLISM

Does Tretinoin get metabolized in Skin?

Metabolism of tretinoin (all-trans tretinoic acid, RA) occurs via a cytochrome P-450 mediated enzyme system in tissues such as liver, trachea, intestine, and rodent skin. The primary product formed is 4-OH RA which is further transformed to 4-oxo RA. Topical administration of RA to skin of 4-day old rats increases RA metabolism fourfold. In order to investigate the effect of a pharmacological dose of RA on the level of RA in adult human skin and on cytochrome P-450 activity, a single topical dose of of 0.1% RA cream or cream vehicle was applied to adult human skin and kept under occlusion. After 96 h, vehicle and RA-treated sites were tested for their capacity to metabolize RA and were analyzed for RA and RA metabolite content. Treated areas were removed by a keratome and microsomal fraction was isolated from each biopsy. In vitro incubation of ^{3}H-RA with microsomes from in vivo RA treated sites resulted in a 4.5 fold increase ($P = 0.0001$, $n= 13$) in its transformation to 4-OH RA in comparison to in vitro incubations with microsomes from in vivo cream alone treated sites. To determine whether metabolites might play a significant role in retinoid effects observed following topical applications of RA, treated sites were analyzed for the presence of RA and RA metabolites through tape stripping. The high levels of extracted 13-cis-RA and 4-OH RA suggests increased metabolism of RA within living layers of epidermis and the effects observed clinically after topical application of pharmacological doses of RA may be due in part to RA and 13-cis-RA as well as 4-OH RA.

V. FORMULATION

How does TEC II differ from TEC IA?

RENOVA (TEC IA) 0.05% is a water-in-oil formulation whereas RENOVA (TEC II) 0.02% is an oil-in-water formulation. The following table describes the line by line comparison of TEC IA and TEC II formulations:

Formula	TEC IA	TEC II
Ingredients	%w/w	%w/w
Tretinoin, USP		
Butylated Hydroxytoluene (—), NF		
Citric Acid, Monohydrate, USP		
Edetate Disodium. Dihydrate, USP		
Quaternium-15 (		
Propylparaben, NF		
Methyl Paraben, NF		
Xanthan Gum , NF		
Dimethicone 50 CS		
Stearoxytrimethylsilane + Stearyl Alcohol		
PEG-45 Dodecyl Glycol Copolymer		
Methoxy PEG-22 Dodecyl Glycol Copolymer		
Sorbitol Solution, USP		
Hydroxyoctacosanyl Hydroxysteartae		
Light Mineral Oil, NF		
Stearath-2		
Benzyl Alcohol, NF		
Stearyl Alcohol, NF		
Cetyl Alcohol, NF		
Stearic Acid , NF		
Steareth-20		
Caprylic/Capric Triglyceride		
Water, Purified, USP		

VI. SUMMARY OF SUPPORTIVE IN-VIVO STUDIES

Did the Sponsor provide adequate supportive in-vivo study results?

The sponsor provided results from two supportive in-vivo studies from the previous NDA (19-963) for 0.05% RENOVA. Study I described percutaneous absorption of tretinoin under single and multiple applications up to 28 days whereas Study II described percutaneous absorption of tretinoin under single and multiple applications up to one year. Studies III and IV are continuation of Study I where the sponsor evaluated the effect of single and multiple applications of 0.05% tretinoin on the endogeneous concentration of tretinoin and its major metabolites using samples from Study I. They did not perform any pharmacokinetic study with new 0.02% tretinoin formulation. The summary of the studies described below is summarized in Table X.

Study I (Dec 14, 89): The Percutaneous Absorption of ^{3}H - All-Trans-Retinoic Acid (^{3}H-Tretinoin) in Human Volunteers (Report # DMR-1415)

The purpose of this study was to determine and compare the percutaneous absorption of ^{3}H – tretinoin (—— 8203) from each of three different cream formulations (RETIN-A 0.05%, TEC-I 0.05% and TEC-II 0.05%) following single and repeated applications to normal male subjects.

Each of the twenty-one subjects (seven for each formulations) received topically a single dose of ^{3}H -tretinoin (100 μCi; 50 μg) in 100 mg of cream. The remaining twenty-one subjects received the same radioactive dose following administration of the assigned formulation (non-radioactive) for the prior 28 days (repeat). Blood was collected at various intervals up to 72 hr after dosing with ^{3}H - tretinoin. Urine and feces were collected for 7 days after dosing. Plasma, urine and fecal (following combustion) radioactivity levels were determined by ——————————At 10 hours following drug administration, the patients washed their faces. The radioactive contents of the facial wipes and washes as well as the rubber gloves and weighing paper (used in the dispensing of the dose) were also determined. The results are summarized in Tables III and IV:

Table III: Recovery of Administered Radioactivity from Biological And Non-biological Materials

	Retin-A		TEC-I		TEC-II	
% Radioactivity recovered in	Single	Repeat	Single	Repeat	Single	Repeat
Urine	1.22±0.26	1.47±0.44	1.40±0.60	1.11±0.22	1.36±0.28	0.94±0.30
Feces	0.63±0.33	0.66±0.32	0.58±0.19	0.61±0.17	0.66±0.17	0.44±0.10
Total Percutaneous Absorption	**1.85±0.58**	**2.13±0.66**	**1.98±0.78**	**1.72±0.26**	**2.02±0.40**	**1.38±0.37**
Dispensing materials	24.1±4.1	26.5±3.6	14.1±5.8	11.4±3.1	16.9±9.1	15.7±3.0
Facial wipes and washes	44.8±29.9	56.2±14.7	41.2±18.6	57.9±18.3	34.8±17.3	62.2±10.4
Total Recovery	**70.75**	**84.83**	**57.28**	**71.02**	**53.72**	**79.28**

Table IV: Peak-Plasma Concentration Following Topical Administration of ^{3}H-Tretinoin To Human Subjects

	Treatment Group					
C_{max} (pg-eq/mL)	Retin-A		TEC-I		TEC-II	
	Single	Repeat	Single	Repeat	Single	Repeat
Range						
Mean±SD	19.46±5.24	21.10±6.52	18.73±11.14	18.83±1.94	16.85±5.19	12.18±9.30
%CV	26.93	30.89	59.47	10.33	30.79	76.39

The data obtained in this study indicate that tretinoin is minimally absorbed following topical administration, and that the low percutaneous absorption is unaffected by either the specific cream formulation or the dosage regimen (single versus multiple application).

Study II (July 31, 1990): An open label study to compare the percutaneous absorption of tretinoin following single-dose and long-term administration (Report # DM – 90339)

The study was designed to compare the percutaneous absorption of ^{3}H - tretinoin after a single dose and after long-term (at least 12 months) administration.

The study was an open label, single site phase I study in nine volunteers (three female/two male single dose, and three female/one male with long-term tretinoin exposure). The nine subjects were divided into two treatment groups. Four subjects had received at least twelve months pretreatment with 0.05% **RETIN-A®** cream prior to receiving the ^{3}H - tretinoin treatment. The other five subjects had not been pretreated and were of similar age and sex to the subjects in the pretreated group. Each subject received topically a single dose of ^{3}H-tretinoin (100 μCi; 50 μg) in 100 mg of cream. Blood was collected at various intervals up to 72 hr after dosing with ^{3}H tretinoin. Urine and feces were collected for seven days after application. All samples, including application and wash materials, were assayed for total radioactivity by [redacted]. The actual dose applied and the extent of percutaneous absorption were determined from the data.

The actual applied dose for each subject was obtained by subtracting the radioactivity measured in the dispensing/application materials from the corresponding total dose of radioactivity. Since each subject received a slightly different dose, all measured radioactivity values were corrected to a dose of 35 μg for statistical comparisons.

Mean plasma concentration vs. time profiles for the long-term and single dose applications are illustrated in Figure 2. From these data, the pharmacokinetic parameters are summarized in Table V.

Table V: Pharmacokinetic Parameters in the Long-Term and Single-Dose Application Groups

PK-parameters	Single Dose	Long-term
$AUC_{(0-\infty)}$ (pg-eq.h/mL) (mean±SD)	626.50±193.50	511.40±137.40)
C_{max} (pg-eq/mL) (mean ±SD)	12.59±3.47	10.46±1.11
T_{max} (h) (mean±SD)	16.00±7.48	17.50±7.55

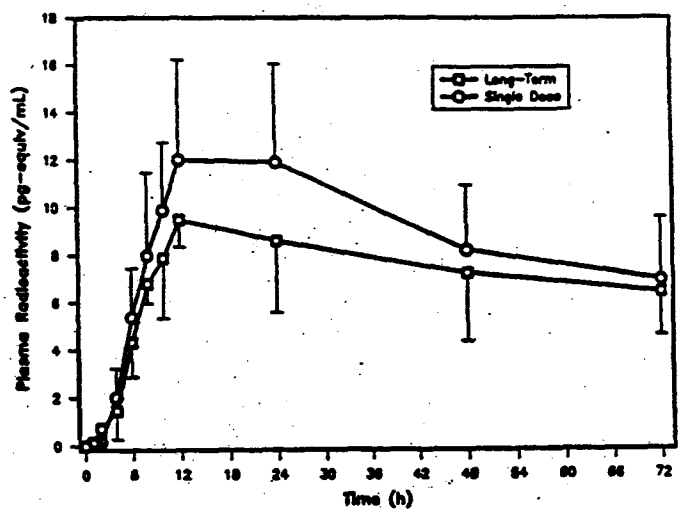

Fig 2: Mean Plasma Concentration vs. Time Curves Following the Long-Term and Single Dose Application of 0.05% RETIN-A®

The mean cumulative excretions of urinary and fecal radioactivity are plotted for each treatment group in Figures 3 and 4 respectively. The total extent of percutaneous absorption (the sum of the urine and fecal elimination) is summarized in Table VI.

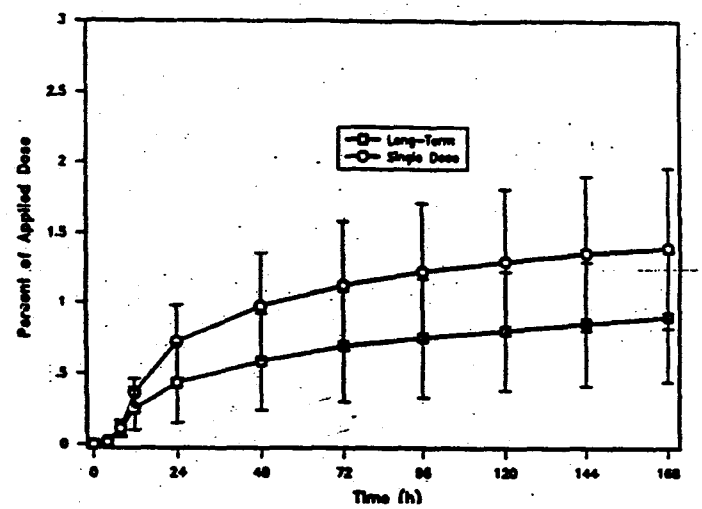

Fig 3: Cumulative Percent of Radioactivity Excreted in Urine vs. Time Curves Following the Long-Term and Single Dose Application of 0.05% RETIN-A®

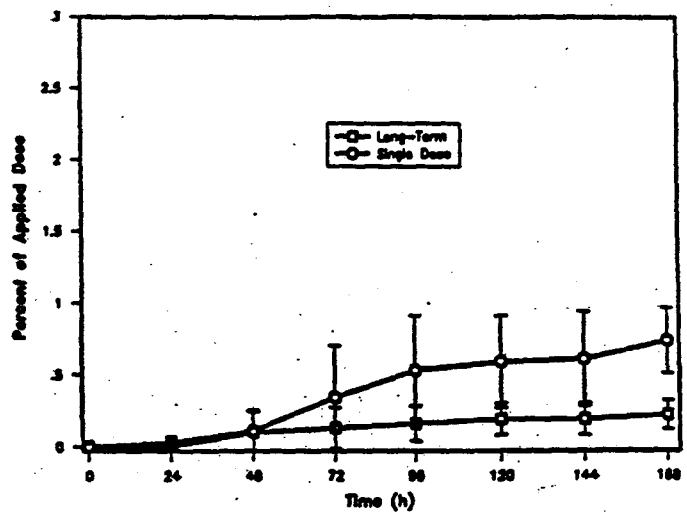

Fig 4: Cumulative Percent of Radioactivity Excreted in Feces vs. Time Curves Following the Long-Term and Single Dose Application of 0.05% RETIN-A®

Table VI: Cumulative Absorption of Radioactivity in the Single Dose and Long Term Applications Group

Cumulative Radioactivity eliminated in (%)	Treatment Type	
	Single Dose	Long-term
Urine	1.39±0.56	0.90±0.46
Feces	0.74±0.23	0.22±0.10
Total Percutaneous Absorption	**2.13±0.65**	**1.11±0.40**

Plasma radioactivity concentrations were consistently low for both treatment groups. The total cumulative absorption for the long-term group was significantly lower than that of the single dose group although the magnitude of the difference was small. The data obtained in this study indicate that trtetinoin is minimally absorbed after topical administration and that the extent of absorption does not increase after long-term administration. The peak plasma radioactivity concentrations obtained in this study are at least 100-fold lower than known endogenous concentration (2-4 ng/mL), suggesting that the potential for adverse systemic effects due to the percutaneous absorption of tretinoin would be negligible.

Study III (Aug 13, 1991): Determination of Tretinoic Acid, 13-cis-4-oxo-Retinoic Acid and All-trans-4-oxo-Retinoic Acid concentrations in Human Volunteers Following Administration of RETIN-A and RENOVA for 29 Days (Report DM 91020)

The present study was conducted to determine whether endogenous concentrations of tretinoin and its metabolites were altered following single or repeated topical application of two 0.05% cream formulations (RETIN-A and RENOVA).

In this study, an extremely sensitive and accurate HPLC method of quantitation was used to determine the concentration of parent drug (All-trans-retinoic acid) and its major metabolites 13-cis-Retinoic Acid, 13-cis-4-oxo-Retinoic Acid and all-trans-4-oxo-Retinoic Acid in the plasma samples collected **previously** from fourteen subjects (seven/formulation) treated topically with a single dose of ^{3}H-tretinoin (100 μCi; 50 μg) in 100 mg of cream following administration of the assigned formulation (non-radioactive) for the prior 28 days (Study I, Report #DMR 1415). The results are summarized in Tables VII and VIII.

Table VII: Tretinoin Plasma Concentration at Various Time Points in Patients Receiving Single and Multiple Topical Applications of 0.05% RENOVA®

Time (hrs)	Tretinoin Concentration (ng/mL)	
	Single (mean ± SD)	Multiple (mean ± SD)
0	2.68±0.46	2.57±0.75
12	Not measured	2.79±0.77
24	2.54±0.33	2.90±0.86
48	2.66±0.80	2.65±0.85
72	3.14±0.68	2.87±0.70

Table VIII: Tretinoin Plasma Concentration at Various Time Points in Patients Receiving Single and Multiple Topical Applications of 0.05% RETIN-A®

Time (hrs)	Tretinoin Concentration (ng/mL)	
	Single (mean ± SD)	Multiple (mean ± SD)
0	2.74±0.57	2.36±0.71
12	Not measured	2.32±0.76
24	2.91±0.81	2.60±1.10
48	3.10±0.56	2.45±0.60
72	2.45±1.16	2.96±0.85

The results indicate that endogenous plasma concentrations of tretinoin and its metabolites were unchanged after receiving a single application or after multiple topical treatments of either 0.05% RENOVA ® or 0.05% RETIN-A ®. The difference between the systemic concentrations of exogenous material (radioactivity) and the endogenous tretinoin concentrations is at least 100 fold, as shown in Figures 5 and 6 respectively.

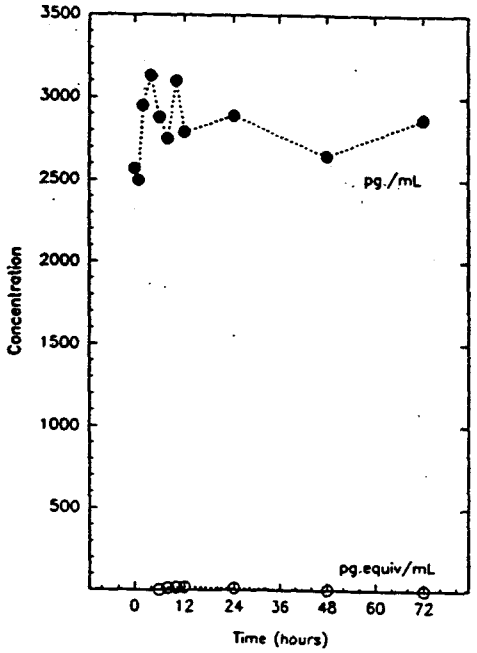

Fig 5: Mean Plasma Concentrations in Subjects Treated for 28 Days with RENOVA® (•-----• Tretinoin, pg/mL) and Subjects Pretreated for 28 Days with RENOVA Followed By A Single Dose of ^{3}H-RENOVA (o-----o Total Radioactivity, pg-equivalent/mL)

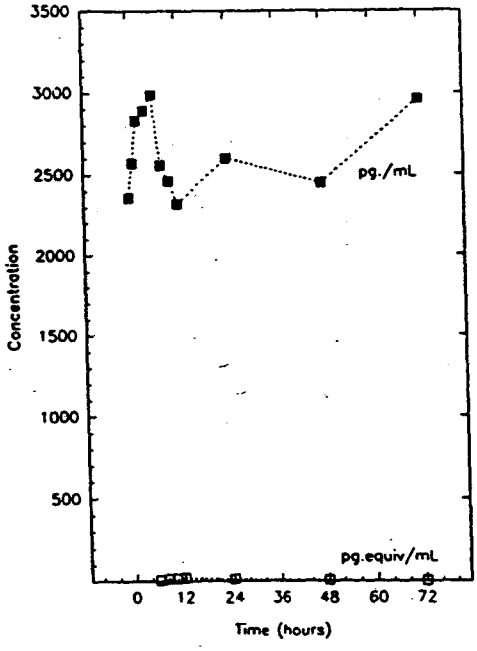

Fig 6: Mean Plasma Concentrations in Subjects Treated for 28 Days with RETIN-A® (•-----• Tretinoin, pg/mL) and Subjects Pretreated for 28 Days with RETIN-A Followed By A Single Dose of ^{3}H-RETIN-A (o-----o Total Radioactivity, pg-equivalent/mL)

Plasma concentrations of the metabolites were generally non-measurable. The endogenous plasma concentrations of 13-cis-RA were below the quantitation limit of ———— in almost all subjects. Only three subjects, at scattered time points, had any measurable plasma concentrations on day 29, which ranged from ———— For the cis-4-oxo-RA metabolite, measurable concentrations of this endogenous compound were found in several subjects and these ranged from ———— In none of the subjects was any of the trans-4-oxo-RA metabolite above the limit of quantitation of ————

The results obtained in this study demonstrate that the range of endogenous concentrations of tretinoin, 13-cis-retinoic acid, 13-cis-4-oxo-RA, and all-trans-4-oxo-RA was unchanged after single or multiple application of 0.05% tretinoin in RENOVA® or RETIN-A formulations.

Study IV (Feb 15, 1993): Determination of All-trans-Retinoic Acid Concentrations in Human Volunteers Following Administration of 0.05% TEC II for 29 Days (Report: DM-91020-A)

As an addendum to the previous study (DM-91020), the present study was conducted to determine whether endogenous concentrations of tretinoin and its metabolites were altered following single or repeated topical application of 0.05% TEC II. Even though minimal absorption was demonstrated in the previous study with RENOVA and RETIN-A, that study did not address the influence of topical administration on endogenous concentrations of tretinoin from TEC II formulation.

Seven subjects received topically a single dose of ^{3}H - tretinoin (100 μCi; 50 μg) in 100 mg of cream. An additional seven subjects received the same radioactive dose following administration of the assigned formulation (non-radioactive) for the prior 28 days. Blood was collected at various intervals up to 72 hr after dosing with ^{3}H tretinoin. Plasma concentrations of intact tretinoin were determined using a validated HPLC assay which included ———— The results are summarized in Table IX.

Table IX: Tretinoin Plasma Concentration at Various Time Points in Patients Receiving Single and Multiple Topical Applications of 0.05% TEC II

Time (hrs)	Tretinoin Concentration (ng/mL)	
	Single (mean ± SD)	Multiple (mean ± SD)
0	2.00±2.87	3.26±2.14
12	Not measured	3.71±2.66
24	0.00±0.00	3.34±3.56
48	0.73±1.80	4.46±2.69
72	1.43±2.65	6.70±3.63

To determine whether there was a difference between mean plasma concentrations after single and multiple dosing, analysis of covariance was performed with concentration as

the dependent variable, dose as a factor, and time as a covariate. To examine any change in baseline concentrations of tretinoin after repetitive dosing, the mean plasma concentrations at 0 h of single dosing, 0 h on day 29 of multiple dosing, and 12 h on day 29 of multiple dosing were compared by means of two-way analysis of variance. The 12 h concentration data were also included for comparison, as a previous study showed that it was the time to mean peak concentration of total radioactivity after as single radiolabelled dose. Statistical analyses were performed using SAS procedures.

Endogenous plasma concentrations of tretinoin were unchanged after receiving a single application or after repeated topical applications of TEC II. Due to the limit in assay sensitivity ——— only few subjects at scattered time points had measurable concentrations of tretinoin on day 1. Owing to the insufficiency of single-dose data, statistical analysis using time as the covariate to compare the single- and multiple-dose data was not performed. The values (0 h, day1; 0 h, day 29; 12 h, day 29) were not statistically different from the two-way analysis of variance (P=0.33).

The data obtained in this study demonstrate that the range of endogenous concentrations of tretinoin was unchanged after a single application or after a repeated daily topical applications of 0.05% tretinoin in TEC II. The range of baseline tretinoin concentration in these subjects (< 2 to 11.84) is in good agreement with previously reported literature values of 1 to 7 ng/mL for healthy, untreated subjects (Fig. 7).

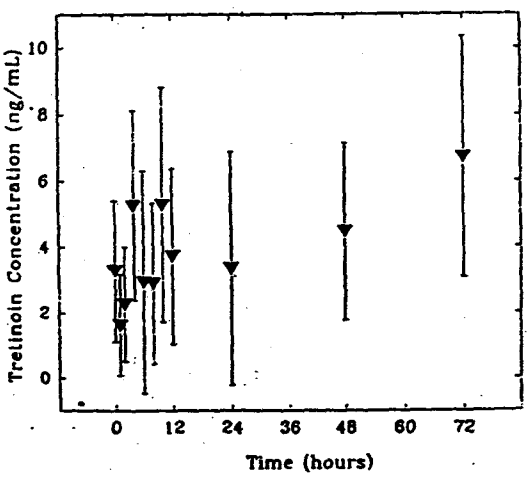

Fig. 7: Day 29 Mean Plasma Concentrations in Subjects After Repeated Topical Applications of 0.05% TEC II

The results of this study is not unexpected as previous analysis of these samples for radioactivity had shown that systemic absorption of exogenous tretinoin from TEC II was minimal, ranging from 1.4 – 2.2% of the applied dose and peak plasma radioactivity concentrations (Cmax) ranged from ——— at 10-12 h postdose. In comparison, the mean tretinoin plasma concentration at 12 h postdose on day 29 was 3.71 ± 2.66 ng/mL. The difference between the systemic concentrations of exogenous drug (plasma radioactivity) and the endogenous tretinoin concentrations is at least 100 fold.

Similar results have been previously reported for the RETIN-A® and RENOVA® formulation. It is of note that in a previous percutaneous absorption study (Study II), in which male and female subjects had been on daily RETIN-A® treatment for over one year, the extent of absorption was significantly lower for the long-term group, and no sex related differences in absorption were observed. Plasma radioactivity concentrations in that study showed a similar trend, i.e., total drug-derived material was more than 100-fold lower than the literature-reported endogenous tretinoin concentrations. Collectively, absorption from topically applied tretinoin was minimal and unlikely to contribute to the overall body pool of tretinoin and metabolites.

VII. SUMMARY OF IN-VITRO STUDY

Was the design of the In-Vitro study all right to address the effect of loading concentration on tretinoin release?

The sponsor adequately designed in-vitro release study using artificial membrane to show dose proportionality on the release of tretinoin from the same formulation. However, human cadaver skin could have been a better membrane for mimicking real life situation.

Study V: (Feb 19, 1999) In Vitro Release Studies of Tretinoin From TEC II Cream Formulations Using ——— Apparatus

This study was conducted to assess the effect of concentration, process and manufacturing site on the release rate of tretinoin from TEC II formulations.

The study of interest from biopharmaceutics point of view was to compare the effect of drug concentration (0.05% and 0.02% tretinoin) on the release of tretinoin from TEC II formulation.

In-Vitro Methodology:

A standard 10 mL ——— of ——— design with a ——— was used. Using an ——— template to produce a layer of uniform thickness and diameter, the formulation ——— was applied to the surface of the ——— ——— mounted on the ——— under occluded condition. The ——— was filled with receptor solution ——— ———) maintained at 32°C by a ——— At predetermined time intervals, the receptor medium was removed. ——— Initial solubility experiment confirmed maintenance of ——— under this condition. Tretinoin content in the receptor phase was analyzed by a validated ——— method.

——— Method Validation:
The ———method is based on the separation of Tretinoin from other components in the sample on a ——— column. The separation is performed ——— and an

[redacted] is used at [redacted] Quantitation is accomplished by external standardization.

System Suitability and Specificity: The method was validated for system suitability and specificity. Retention time of tretinoin is about [redacted] and the method is specific to distinguish between tretinoin, 13-cis-RA and 11-cis-RA at [redacted] tretinoin concentration.

Linearity: The linearity of the [redacted] method was determined by analyzing eight different tretinoin concentrations ranging from [redacted]

[redacted]

Recovery: The percent recovery was calculated by determining the ratio of the amount found divided by the amount [redacted] The values of percent recovery are within [redacted] which are acceptable.

Within Assay Precision and Ruggedness: The repeatability was tested by using [redacted] determinations of [redacted] different tretinoin concentrations by a single analyst using the same liquid chromatograph with the same column. The % RSD from determination to determination is less than [redacted]. Analysis of the same solutions by a second analyst using different liquid chromatograph and different column confirmed the ruggedness of the method.

Limit of Quantitation (LOQ): The value of [redacted] was assigned as the LOQ based on the [redacted] ratio produced by tretinoin solution at various concentrations.

Stability of Tretinoin in Receptor Solution: The stability of two concentrations of tretinoin [redacted] in the receptor solution at [redacted] was evaluated. Tretinoin is stable for [redacted] days at both [redacted]

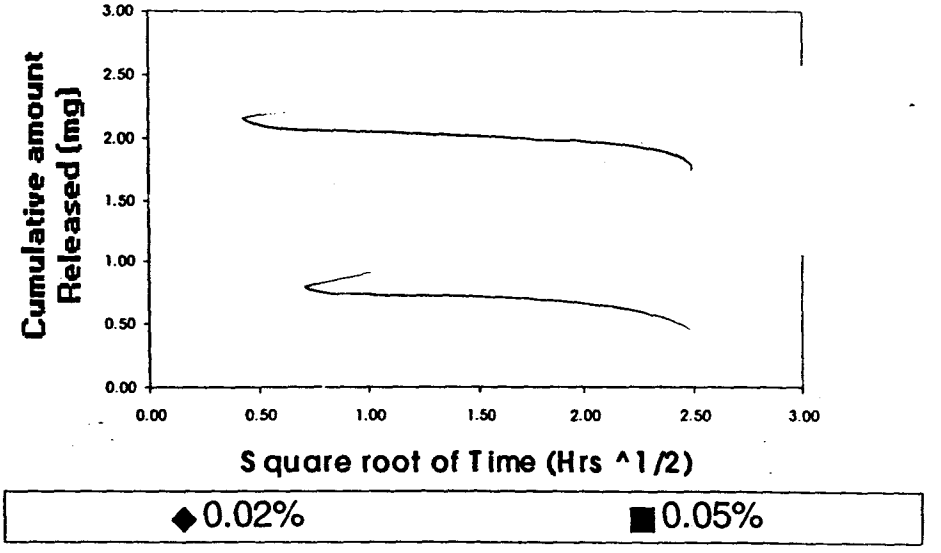

Figure 8: Release of Tretinoin From 0.02% and 0.05% TEC II Cream Formulations

Results:

The release profiles evaluating the effect of concentration (0.05% and 0.02% tretinoin) are shown in Figure 8. The slopes for 0.05% and 0.02% tretinoin are [redacted] and [redacted] respectively. The results indicate that the release rate of tretinoin is proportional to its concentration in the formulation.

VIII. CONCLUSIONS

A summary of the pertinent studies related to this application is described in the following Table X:

Table X: Summary of Supporting Studies Arranged in Chronological Order

Study#	Signed out Date	Report#	Title	Conclusion
I	12/14/89	DMR-1415	The Percutaneous Absorption of ^{3}H - All-Trans-Retinoic Acid (^{3}H-Tretinoin) in Human Volunteers	The sponsor showed through radioactive study that percutaneous absorption of tretinoin was minimal from all three (RETIN-A, TEC IA, TEC II) formulations at 0.05% concentration both under single and multiple applications (28 days)
II	7/31/90	DM-90339	An open label study to compare the percutaneous absorption of tretinoin following single-dose and long-term administration	The sponsor showed through radioactive study that percutaneous absorption of tretinoin was minimal from RETIN-A formulation at 0.05% concentration both under single and long-term applications (1 year)
III	8/13/91	DM-91020	Determination of Tretinoic Acid, 13-cis-4-oxo-Retinoic Acid and All-trans-4-oxo-Retinoic Acid concentrations in Human Volunteers Following Administration of RETIN-A and RENOVA for 29 Days	Through sensitive HPLC method of quantitation on previously collected samples, the sponsor showed that endogenous concentration of tretinoin and its major metabolites remain unchanged after single and multiple applications of 0.05% tretinoin in RETIN-A and RENOVA (TEC IA) formulations.
IV	2/15/93	DM-91020-A	Determination of Tretinoic Acid, 13-cis-4-oxo-Retinoic Acid and All-trans-4-oxo-Retinoic Acid concentrations in Human Volunteers Following Administration of TEC II (0.05%) for 29 Days	Through sensitive HPLC method of quantitation on previously collected samples, the sponsor showed that endogenous concentration of tretinoin and its major metabolites remain unchanged after single and multiple applications of 0.05% tretinoin in TEC II formulation.
V	2/19/99		In Vitro Release Studies of Tretinoin From TEC II Cream Formulations Using [redacted] Apparatus	The only new study conducted by sponsor under this NDA. The results indicate that the release rate of tretinoin is proportional to its concentration in the formulation.

The contents of this submission is based on an agreement between the Division of Dermatologic and Dental Drug Products and the sponsor where it was agreed that an integrated summary of clinical pharmacokinetics data and in-vitro release data would suffice for this NDA.

No significant increase beyond endogenous levels of tretinoin were seen in these studies or would be expected from the topical application of the proposed formulation. The data obtained in the clinical studies, and those discussed in the nonclinical pharmacokinetic section were used to develop a physiologically based pharmacokinetic model. The model was used to estimate maternal and fetal plasma concentrations of tretinoin and its metabolites in a theoretical abuse situation, i.e., after excessive application to face, lower arms, chest and neck and assuming exaggerated absorption of 10%. This model demonstrated that the systemic concentrations of tretinoin and potentially toxic metabolites achieved under such conditions remained several orders of magnitude below endogenous concentration and minimally teratogenic dose of retinoic acid. However, as the conclusion drawn from this model did not bear any implication on the approvability of this NDA, fitting of the clinical and non-clinical data in the proposed model and its predictability of systemic toxicity in the real life situation was not reviewed critically.

IX. COMMENTS:

- Studies III (DM 91020) and IV (DM 91020A) are continuation of the study I (DMR 1415). Final report of the study I (DMR 1415) was signed out on December 14, 1989 whereas studies III and IV were concluded in 1991 and 1993 respectively. The sponsor did not address the issue of the stability of tretinoin in the blood samples collected in or prior to 1989 and analyzed between 1991 and 1993. In future, stability of actives after long term storage should be provided in the submission.

- [redacted] **(This Comment is not to be conveyed to the sponsor)**

- Literature on oral tretinoin as well as supportive studies submitted herewith on topical administration showed a decrease in systemic AUC of tretinoin after multiple-day therapy by both routes of administration. This reduction in AUC after oral administration is to a large extent due to CYP P- 450 mediated metabolism at liver and intestinal wall. Metabolism of tretinoin after topical administration in rodent as well as human skin is reported in the literature. Therefore, reduction in AUC as observed in Study II (Report # DM-90339) may be due to reduced percutaneous absorption (measured indirectly from urine and feces) and/or increased metabolism of tretinoin by induced CYP P-450 system at skin after long-term therapy. This result raises concern on efficacy of topical 0.02% tretinoin therapy after long-term treatment.

X: LABELING COMMENTS:

The following labeling changes should be incorporated in the label:

[ ]

[/S/]
Tapash K. Ghosh, Ph.D. 3/28/2000
Pharmacokineticist
DPE III

Team Leader: E. Dennis Bashaw, Pharm.D. /S/ 5/1/00

CC: NDA 21-108 (Orig)
HFD-540/Div File
HFD-540/CSO/Cintron
HFD-880(Bashaw/Ghosh)
HFD-880 (Lazor)
HFD-344 (Viswanathan)
CDR ATTN: B. Murphy

APPEARS THIS WAY
ON ORIGINAL