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Effect of Ethyl Cellulose in a Medium-Chain Triglyceride on the Bioavailability of Ceftizoxime

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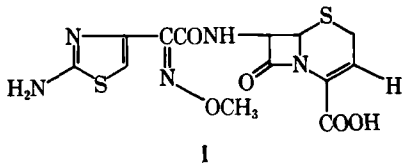
Received February 22, 1982 from the *Research Laboratories, Fujisawa Pharmaceutical Co., Ltd., Osaka 532, Japan.* Accepted for publication July 16, 1982.

Abstract □ The oral bioavailability of new formulations of ceftizoxime sodium was investigated in animals and humans. In rats, one of the formulations tested showed significant improvement, with a urinary excretion of 47.7% (0–24 hr). Good results were obtained also in dogs. In humans, the mean peak serum level was 3.6 μg/ml at 3.3 hr postadministration for formulation 10. The average ceftizoxime AUC at 0–8 hr was 17.3 μg hr/ml and urinary excretion of ceftizoxime was 9.6% (0–24 hr). The concentrations in the serum exceeded the minimum inhibitory concentrations for most of the commonly encountered bacterial pathogens.

Keyphrases □ Bioavailability—oral ceftizoxime in rats, dogs, and humans, effect of ethyl cellulose in medium-chain triglyceride □ Ceftizoxime—bioavailability of oral formulations in rats, dogs, and humans, effect of ethyl cellulose in medium-chain triglyceride □ Ethyl cellulose—effect with medium-chain triglyceride on the oral bioavailability of ceftizoxime in rats, dogs, and humans □ Triglyceride, medium-chain—effect with ethyl cellulose on the oral bioavailability of ceftizoxime in rats, dogs, and humans

Ceftizoxime (I) is a new cephalosporin antibiotic which is active against both Gram-positive and Gram-negative bacteria.



I

The activity of ceftizoxime *in vitro* has been confirmed (1), and the reports on its clinical efficiency are numerous (2). The metabolism and pharmacokinetics of this drug have also been described (3). Ceftizoxime is administered parenterally for effective systemic action since it is poorly absorbed from the GI tract[1]. The purpose of this investigation was to determine the oral bioavailability of ceftizoxime after the administration of its sodium salt to animals and humans in new formulations produced with a combination of medium-chain triglyceride and ethyl cellulose as additives.

Attempts to improve the oral bioavailability of poorly absorbed drugs by devising different pharmaceutical formulations have been reported (4–6). Similarly, our efforts have been directed to the development of new oral formulations of ceftizoxime. We have systematically studied a number of hydrophilic and hydrophobic vehicles, surfactants, and nonsurfactants as additives and found that a combination of ethyl cellulose and a medium-chain triglyceride enhanced the oral absorption of ceftizoxime in rats, dogs, and humans.

EXPERIMENTAL

Materials—Ceftizoxime sodium[2] was prepared as described in the patent (7). Commercially available ethyl cellulose[3], polyethylene glycol 400[4], a medium-chain triglyceride[5], olive oil[4], and ethyl alcohol[4] were used in the suspensions.

Formulations—Formulations containing various amounts of ethyl cellulose were tested. A medium-chain triglyceride (50 ml) and ethyl cellulose (500 mg) dissolved in 1 ml of ethyl alcohol were mixed with stirring. The ethyl alcohol was removed under reduced pressure. Ceftizoxime sodium (5 g-potency) was then dispersed in the resulting vehicle to give the formulation of ceftizoxime used. Similar formulations containing olive oil or polyethylene glycol 400 instead of the medium-chain triglyceride were also investigated.

Absorption Studies—*Rats*—Six-week-old male Sprague-Dawley rats, weighing 160–230 g, were fasted for 18 hr. The rats were given the ceftizoxime sodium formulations (Table I) using a gastric tube at a dose equivalent to 20 mg/kg. Urine was collected for 24 hr, stored at −20°, and

[1] Unpublished data.

[2] Epocelin; Fujisawa Pharmaceutical Co., Ltd. Osaka, Japan.
[3] Ethocel, 10, 45, and 100 cps; Dow Chemical Co.
[4] Hayashi Pure Chemical Industries, Ltd., Japan.
[5] Miglyol 812; Dynamit Nobel Co.

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Table I—Effect of Vehicle on the Urinary Excretion of Ceftizoxime after Oral Dosing in Rats with Ceftizoxime Sodium in Different Vehicles [a]

Vehicle	Cumulative Urinary Excretion (0–24 hr), % of dose [b]	
Polyethylene glycol 400	7.1	(5)
A medium-chain triglyceride	16.2	(3)
Olive oil	9.7	(3)
A medium-chain [c] triglyceride + ethyl cellulose	37.7	(9)
Olive oil [c] + ethyl cellulose	24.6	(5)
Water	10.3	(5)

[a] At a dose of 20 mg-potency/kg. [b] Values represent the mean; sample size is in parentheses. [c] Ethyl cellulose concentration in the vehicle ~1% (w/v); viscosity of ethyl cellulose was 100 cps.

assayed within 24 hr. The amount of ceftizoxime excreted was determined by high-performance liquid chromatography (HPLC) using a standard curve.

Dogs—The formulations used in the dog experiments are shown in Table II. Male beagle dogs weighing 8.0–13.2 kg were used, with four dogs per group. The animals were fasted overnight and the drug formulations (in hard gelatin capsules) were given in doses equivalent to 40 mg ceftizoxime/kg. Urine was collected for 24 hr; 5-ml venous blood samples were drawn at 0.5, 1, 2, 4, 6, and 8 hr postadministration and centrifuged to obtain the serum. All serum and urine samples were stored at −20° and assayed within 24 hr.

Humans—Formulations 3 and 10 (Table II) were used in the human experiments. Six healthy males, 33–47 years of age and 49.5–67.5 kg, participated in the study after informed written consent was obtained. No other drugs were taken during the investigation period. Prestudy physical examinations and laboratory parameters for all subjects were normal.

Experimental Design—A two-way crossover design was used in the human studies. The two formulations (3 and 10) were given orally to the six volunteers, at an interval of 2 weeks. The volunteers drank 200 ml of water immediately after ingesting the drug. All volunteers fasted overnight before dosing and were given regular meals 2 hr after dosing. Venous blood samples (5 ml) for assay of serum concentration of ceftizoxime were drawn 0.5, 1, 2, 4, 6, 8, and 24 hr postdose. Urine samples were collected at 2-hr intervals from 0 to 8 hr and for the interval 8–24 hr postdose. All serum and urine samples were stored at −20° and assayed within 24 hr.

Assay of Ceftizoxime—The concentration of ceftizoxime in the serum and urine was determined by HPLC[6]. Samples (5 μl) were injected directly into the HPLC equipped with a 254-nm detector and a stainless steel column (30 cm × 4-mm i.d.) packed with μ-Bondapak C_{18}[7]. The mobile phase was 0.6% potassium phosphate–0.2% sodium phosphate–acetonitrile (1:1:0.22). The system was operated at 2.0 ml/min with a column pressure of 2000 psi. Sample chromatograms are shown in Fig. 1.

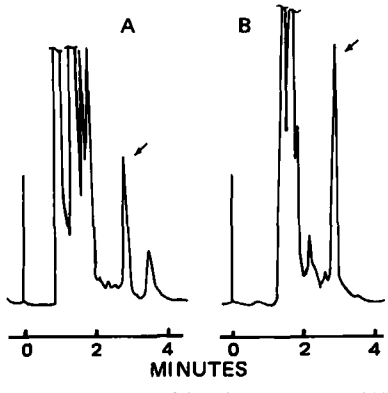

Figure 1—*Chromatogram of ceftizoxime in serum (A) and urine (B) after oral dosing with ceftizoxime sodium. The retention time of the ceftizoxime peak (indicated by the arrow) is 2.8 min in serum and 3.6 min in urine.*

[6] Model ALC/GPC 204 (model 6000A pump, U6-K, and 400 detector); Waters Associates, Milford, Mass.

[7] Waters Associates, Milford, Mass.

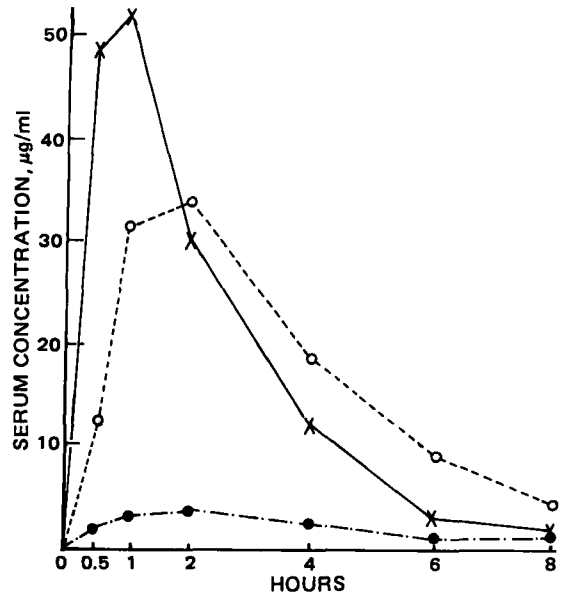

Figure 2—*Time plots of mean serum concentration of ceftizoxime in four dogs after oral dosing with formulations 3 (○) and 10 (×), and with a water vehicle (●) as the control. Dogs received ceftizoxime sodium (at a dose of 40 mg-potency/kg) in a hard gelatin capsule.*

Standard Curves—The reproducibility and linearity of standard curves generated from rat, dog, and human serum and urine containing various amounts of ceftizoxime sodium were studied. The standard curves represent triplicate analyses and were prepared over concentration ranges of 50–600 μg/ml for urine and 2–10 μg/ml for serum. In serum, the least-squares regression line had a slope of 5.324, intercept of −0.143, and correlation coefficient of 0.99996; the limit of detection is 0.2 μg/ml. In urine, the least-squares regression line had a slope of 0.152, intercept of 1.369, and correlation coefficient of 0.999969; the limit of detection is 5 μg/ml.

RESULTS AND DISCUSSION

Table I shows the urinary excretion of ceftizoxime in rats as the percentage of dose after oral administration of ceftizoxime sodium suspension in some typical vehicles. It was found that 10.3% of the dose was excreted when the drug was given in a simple water suspension (control). The urinary excretion of ceftizoxime was somewhat lower when polyethylene glycol 400 or olive oil was used; the medium-chain triglyceride slightly increased the drug bioavailability as evidenced by the urinary excretion rate of 16.2%. It was found, however, that coadministration of ethyl cellulose with ceftizoxime sodium in the medium-chain triglyceride significantly increased the drug absorption: 37.7% of the drug was excreted. The coadministration of ethyl cellulose in olive oil also produced considerably higher urinary excretion (24.6%). However, ethyl cellulose

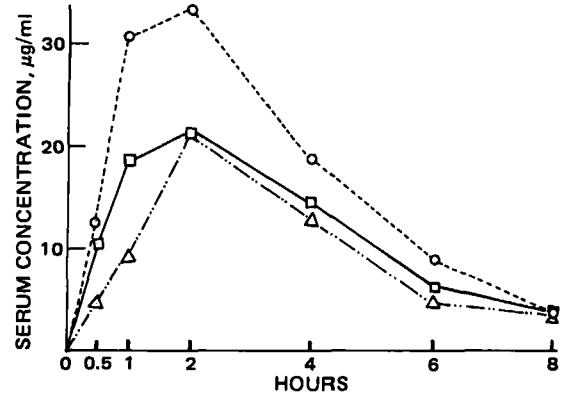

Figure 3—*Time plots of mean serum concentration of ceftizoxime in four dogs after oral dosing with formulations 3 (○), 8 (□), and 9 (△). Dogs received ceftizoxime sodium (at a dose of 40 mg-potency/kg) in a hard gelatin capsule.*

Table II—Oily Formulations for Oral Ceftizoxime Sodium

Formulation	Ceftizoxime Sodium, mg-potency	A Medium-Chain Triglyceride, ml	Ethyl Cellulose[a], mg	Ceftizoxime Sodium in Vehicle, % (w/v)	Ethyl Cellulose in a Medium-Chain Triglyceride, % (w/v)
2	500	10	100 (100 cps)	5.0	1.0
3	500	5.0	50 (100 cps)	10.0	1.0
4	500	2.5	25 (100 cps)	20.0	1.0
5	500	1.0	10 (100 cps)	50.0	1.0
8	500	5.0	50 (10 cps)	10.0	1.0
9	500	5.0	50 (45 cps)	10.0	1.0
10	500	5.0	200 (100 cps)	10.0	4.0

[a] Viscosity of ethyl cellulose in parentheses.

Table III—Effect of Ethyl Cellulose in a Medium-Chain Triglyceride on the Urinary Excretion of Ceftizoxime After Oral Dosing with Ceftizoxime Sodium in Rats[a]

Ethyl Cellulose per ml of Medium-Chain Triglyceride, mg[b]	Concentration of Ethyl Cellulose in the Vehicle, % (w/v)	Cumulative Urinary Excretion (0–24 hr), % of dose[c]
2	0.2	25.5 ± 8.9 (3)
4	0.4	36.5 ± 7.1 (3)
6	0.6	47.7 ± 3.4 (3)
8	0.8	40.7 ± 8.0 (3)
10	1.0	37.7 ± 1.9 (9)

[a] At a dose of 20 mg-potency/kg, suspended in a medium-chain triglyceride. [b] Viscosity of ethyl cellulose was 100 cps. [c] Mean ±*SE*; sample size in parentheses.

did not affect the drug absorption when given in polyethylene glycol 400.

The data in Table III indicate that the effect of ethyl cellulose in the medium-chain triglyceride on the absorption of ceftizoxime in rats was dependent on the concentration of ethyl cellulose used. Ethyl cellulose in the range of 2–10 mg/ml significantly increased the drug absorption. The maximum effect was observed at ~0.6 % (w/v) ethyl cellulose. When ceftizoxime sodium in aqueous solution was administered intravenously to rats at a dose of 20 mg-potency/kg, the urinary excretion of ceftizoxime at 0–24 hr was 79.8% of the given dose[1]. The urinary excretions of 47.7% after oral dosing indicates that ~60% of the administered drug was absorbed.

The experiments in dogs were designed on the basis of these data. The formulations used in the dog experiments are shown in Table II. Figure 2 plots the serum concentrations of ceftizoxime in dogs as a function of time postdose with formulations 3 and 10. The maximum serum concentration of ceftizoxime was significantly higher for formulation 10 than formulation 3; but when the areas under the curve (AUC), calculated by trapezoidal rule, are compared, the differences between the bioavailabilities of formulations 3 and 10 are very small.

The bioavailability studies in dogs, using ethyl cellulose of three different viscosities (10, 45, and 100 cps), demonstrate that ethyl cellulose with a viscosity of 100 cps resulted in higher serum concentrations of

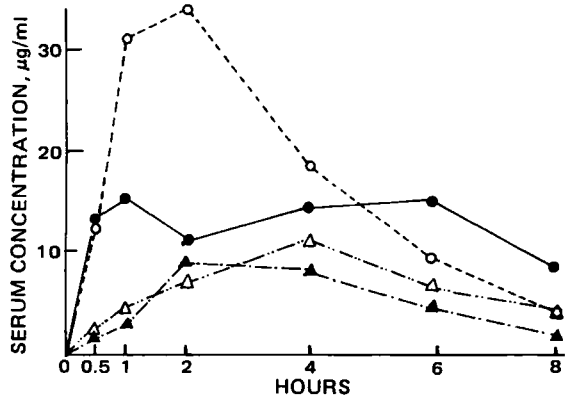

Figure 4—*Time plots of mean serum concentration of ceftizoxime in four dogs after oral dosing with formulations 2 (●), 3 (○), 4 (△), and 5 (▲). Dogs received ceftizoxime sodium (at a dose of 40 mg-potency/kg) in a hard gelatin capsule.*

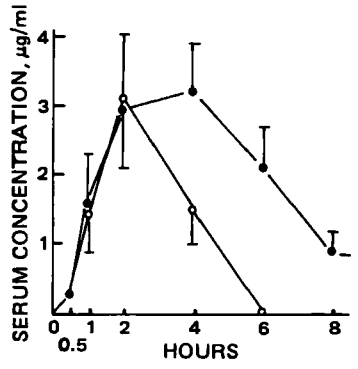

Figure 5—*Time plots of mean serum concentration of ceftizoxime in six human volunteers after oral dosing with formulations 3 (○) and 10 (●). The dose was 500 mg-potency/kg. The data points represent the mean values; standard errors are represented by the bars around each point.*

ceftizoxime than ethyl cellulose additives with viscosities of 10 and 45 cps (formulations 8 and 9) (Fig. 3).

The effect of the vehicle volume on the oral absorption of ceftizoxime was also investigated. Figure 4 shows the bioavailabilities (serum concentrations) in dogs for 5, 10, 20, and 50% (w/v) suspensions of ceftizoxime sodium in the medium-chain triglyceride containing ethyl cellulose (formulations 2, 3, 4, and 5, respectively). The oral absorption of ceftizoxime was shown to be dependent on the volume of the vehicle. The greatest absorption was observed with the 10% suspension, the next

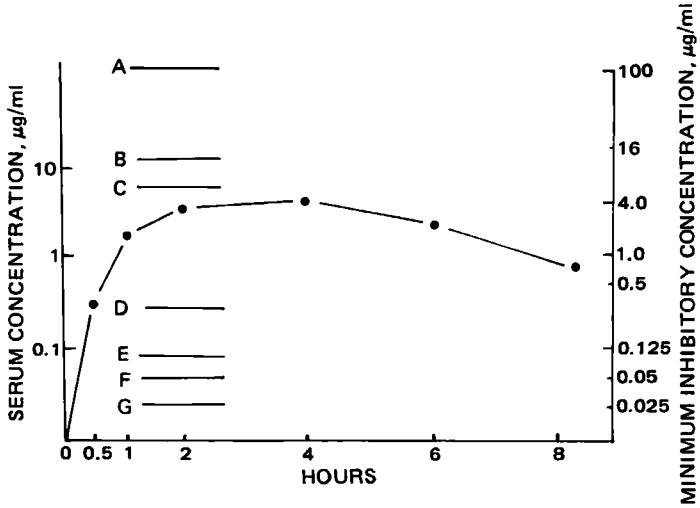

Figure 6—*Mean serum concentration of ceftizoxime (●) in six human volunteers after oral dosing with formulation 10. The lines indicate the minimum inhibitory concentrations for various bacterial pathogens. Key: (A)* Pseudomonas aeruginosa *IAM-1095; (B)* Escherichia coli *35 and* Enterobacter cloacae *1; (C)* Staphylococcus aureus *209p JC-1; (D)* E. coli *28; (E)* Proteus rettgeri *14; (F)* Proteus vulgaris *IAM-1025 and* Streptococcus pneumoniae *III; and (G)* Klebsiella pneumoniae *NCTC-418,* Shigella flexneri *la EW-8,* Proteus mirabilis *1, and* Salmonella typhi *T-287.*

Table IV—Serum Concentrations of Ceftizoxime in Human Volunteers after Oral Dosing with Formulations 3 and 10 [a]

Formulation	Subject	Age, years	Weight, kg	Serum Concentrations of Ceftizoxime, μg/ml [b] 0.5 hr	1 hr	2 hr	4 hr	6 hr	8 hr	C_{max}, μg/ml	T_{max}, hr	AUC, μg hr/ml
	A	33	66.0	0.6	2.9	3.4	2.1	0.1	—[c]	3.4	2.0	12.0
	B	46	49.5	0.1	0.2	0.3	0.2	—	—	0.3	2.0	1.1
3	C	45	66.0	—	1.0	3.7	2.1	—	—	3.7	2.0	10.5
	D	47	54.5	—	—	0.6	—	—	—	0.6	2.0	0.9
	E	38	54.5	—	2.0	6.1	3.0	—	—	6.1	2.0	16.7
	F	35	53.5	1.0	2.5	4.4	1.6	—	—	4.4	2.0	12.2
Mean				0.3	1.4	3.1	1.5			3.1	2.0	8.9
SE				±0.2	±0.5	±0.9	±0.5			±0.9	±0.0	±2.6
	A	33	67.0	—	0.9	1.7	4.9	4.2	2.2	4.9	4.0	23.6
	B	46	50.0	—	—	0.1	0.5	0.1	—	0.5	4.0	1.4
10	C	45	67.5	—	0.7	3.2	3.9	2.2	1.4	3.9	4.0	18.9
	D	47	54.5	—	0.7	3.2	2.3	1.1	0.2	3.2	2.0	12.3
	E	38	55.0	—	4.6	5.7	4.4	3.1	0.9	5.7	2.0	27.9
	F	35	54.0	1.5	2.7	3.3	3.4	2.1	0.9	3.4	4.0	19.6
Mean				0.3	1.6	2.9	3.2	2.1	0.9	3.6	3.3	17.3
SE				±0.3	±0.7	±0.8	±0.7	±0.6	±0.3	±0.7	±0.4	±3.8

[a] At a dose of 500 mg-potency/person. [b] At 24 hr no ceftizoxime could be detected. [c] — not detected.

Table V—Urinary Excretion of Ceftizoxime in Human Volunteers after Oral Dosing with Formulations 3 and 10 [a]

Formulation	Subject	Age, Years	Weight, kg	Urinary excretion at time interval in hr, % 0–2	2–4	4–6	6–8	8–24	Cumulation % (0–24 hr)
	A	33	66.0	4.4	3.1	1.3	0.2	—[b]	8.9
	B	46	49.5	0.3	0.3	0.3	0.1	—	1.0
3	C	45	66.0	0.6	2.7	0.7	—	—	4.0
	D	47	54.5	—	0.2	0.2	0.1	—	0.5
	E	38	54.5	1.2	3.6	2.1	0.2	—	7.0
	F	35	53.5	2.5	2.0	0.6	0.1	—	5.2
Mean				1.5	2.0	0.9	0.1	—	4.4
SE				±0.7	±0.6	±0.3	±0.0	—	±1.3
	A	33	67.0	2.2	6.0	4.8	3.4	1.4	17.8
	B	46	50.0	0.1	0.2	0.2	0.1	—	0.5
10	C	45	67.5	1.2	5.2	4.1	1.3	0.1	12.0
	D	47	54.5	1.3	5.7	1.2	1.3	0.2	9.6
	E	38	55.0	2.6	4.6	2.7	0.8	0.2	10.8
	F	35	54.0	3.0	2.1	1.5	0.2	—	6.8
Mean				1.7	4.0	2.4	1.2	0.3	9.6
SE				±0.4	±0.9	±0.7	±0.5	±0.2	±2.3

[a] At a dose of 500 mg-potency/person. [b] — not detected.

greatest with 5% suspension. The mean AUC of the serum concentration–time curve for the 10% suspension was a little larger than that for the 5% suspension, which resulted in a lower maximum serum concentration with a longer duration. The 20 and 50% suspensions tended to produce smaller C_{max}.

Since the animal studies described above suggest that similar effects may occur in humans, a study of the effect of the two formulations that gave the highest serum concentrations in dogs was deemed appropriate. The serum concentrations and bioavailability parameters of ceftizoxime in human volunteers after oral dosing with formulations 3 and 10 are given in Table IV. The time courses of mean serum concentrations of ceftizoxime are shown graphically in Fig. 5. After oral dosing with formulation 3, the serum concentrations of ceftizoxime peaked at 2.0 hr and then declined rather sharply up to 6 hr. The mean (±*SE*) peak concentration (C_{max}) of ceftizoxime was 3.1 ± 0.9 μg/ml (range, 0.3–6.1 μg/ml) and the mean (±*SE*) concentration was 1.5 ± 0.5 μg/ml (range, 0.2–3.0 μg/ml) at 4 hr. After oral dosing with formulation 10, the serum concentrations peaked at 3.3 hr and then declined. The mean C_{max} was 3.6 ± 0.7 μg/ml (range, 0.5–5.7 μg/ml) and the mean concentrations were 2.1 ± 0.6 (range, 0.1–4.2 μg/ml) and 0.9 ± 0.3 μg/ml (range, 0.2–2.2 μg/ml) at 6 and 8 hr, respectively. Formulation 10 gave a slightly higher C_{max} (not statistically significant) and a longer T_{max} (statistically significant at $0.01 < p < 0.05$) than formulation 3. The mean AUC (±*SE*) for the serum concentration–time curve was 8.9 ± 2.6 μg hr/ml (range, 0.9–16.7 μg hr/ml) for formulation 3 and 17.3 ± 3.8 μg hr/ml (range, 1.4–27.9 μg hr/ml) for formulation 10. Thus, formulation 10 gave an AUC double that of formulation 3 (statistically significant at $p < 0.01$).

The urinary excretion of ceftizoxime in human volunteers after dosing with formulations 3 and 10 is given in Table V. The mean (±*SE*) cumulative urinary excretions of ceftizoxime for 0–24 hr were 4.4 ± 1.3% of the dose (range, 0.5–8.9%) for formulation 3 and 9.6 ± 2.3% (range, 0.5–17.8%) for formulation 10. Formulation 10 resulted in a cumulative urinary excretion double that of formulation 3 (statistically significant at $0.01 < p < 0.05$), thus confirming the AUC values.

As shown in Fig. 6, the concentrations determined in the serum over a period of 8 hr after a single dose of 500 mg-potency of ceftizoxime sodium (formulation 10) were found to exceed the therapeutically active level (minimum inhibitory concentration *in vitro*) against many Gram-negative organisms, including strains of *Escherichia coli*, *Klebsiella*, and *Proteus* (1, 2).

In conclusion, when compared with an aqueous solution of ceftizoxime sodium, formulations containing ethyl cellulose improved oral absorption of ceftizoxime. Formulation 10, which contained the higher additive concentration gave a larger ceftizoxime AUC value and higher urinary excretion than formulation 3. The concentration in the serum exceeded the minimum inhibitory concentration for normally encountered bacterial pathogens.

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COMMUNICATIONS

Correlation Between the Psychotropic Potency of Cannabinoids and Their Effect on the ^{1}H-NMR Spectra of Model Membranes

Keyphrases □ Membranes—model, correlation between psychotropic potency of cannabinoids and their effect on the ^{1}H-NMR spectra □ Cannabinoids—correlation between psychotropic potency and their effect on the ^{1}H-NMR spectra of model membranes □ Cholesterol—model membranes, correlation between the psychotropic potency of cannabinoids and their effect on the ^{1}H-NMR spectra □ Phosphatidylcholine—model membrane, correlation between the psychotropic potency of cannabinoids and their effect on the ^{1}H-NMR spectra

To the Editor:

Little is known of the mode of psychotropic action of Δ^1-tetrahydrocannabinol (I), the major active component of hashish (1). It has been suggested that this drug, as well as other psychotropic cannabinoids, exerts its psychotropic effect through a nonspecific interaction with lipid constituents of nerve cell membranes (2). To investigate such nonspecific interactions, phospholipid vesicles (liposomes) have been used as model membranes. These models represent an oversimplification of the complex biological membranes. However, the possibility of manipulating their composition and size permits deduction of the relative importance of the various structural and compositional factors of the membrane in determining the interactions of membranes with drugs. For the effect of a drug on the physical properties of a model membrane to be regarded as relevant to the mode of action of the drug, a correlation must be established between the effect of various derivatives of the drug and their potency on the membrane.

For psychotropically active cannabinoids, electron spin resonance measurements showed that these drugs reduce the order within the bilayer (3) when introduced into vesicles composed of phosphatidylcholine and cholesterol. The disordering effect of five different cannabinoids correlated qualitatively with their psychotropic potency. The psychotropically nonactive drug, cannabidiol (VI), increased the order parameter within the bilayer (3). Since electron spin resonance, as well as several other physicochemical techniques, involve the use of external probes, which might alter the bilayer properties, we have recently studied the effect of the cannabinoids, I and VI, on the ^{1}H-NMR spectra of lipid vesicles (4). The conclusions of this research were similar to those obtained from the electron spin resonance study: addition of small amounts of I (1 mole percent) to vesicles composed of egg phosphatidylcholine and cholesterol (2:1) caused narrowing of the apparent linewidth of the phospholipid methylene groups signal, whereas the chemically similar, nonactive compound, VI, had the opposite effect.

The purpose of the present work was twofold. First, we wanted to establish a correlation between the potency of various cannabinoids[1] and the effect of small quantities (1 mole percent) of these drugs on the apparent linewidth of the NMR resonance of the phospholipid hydrocarbon chain protons in model membranes composed of egg phosphatidylcholine and cholesterol. Second, in a previous work we showed that in the absence of cholesterol in the model membranes, small amounts of I do not produce any significant change in the linewidth of the ^{1}H-NMR resonances. Therefore, we found it of interest to study the role of cholesterol in the interactions of I with the model membranes. These interactions most likely are not due to the existence of specific interactions of I with cholesterol, since there is no evidence for such interactions in the ^{1}H-NMR spectrum of a mixture of these two components in chloroform (4). One alternative explanation was that the lack of a fluidizing effect in the absence of cholesterol is due to the disrupted packing in the highly curved small unilamellar vesicles (5–7). More specifically, vesicles formed by sonification of mixtures of phosphatidylcholine and cholesterol are larger and less curved than those made of the pure phospholipid by the same method. This may cause the difference between the effect of I on vesicles with and without cholesterol. To investigate this possibility, we studied the effect of cannabinoids on the larger vesicles (~500-Å diameter) made of pure phosphatidylcholine by the French press method (8). The results of this study, in conjunction with the dependence of the fluidizing effect of I on the cholesterol content in the vesicles, indicate that cholesterol is important in the interaction of this drug with model membranes.

The model membranes were prepared as follows: solutions of phosphatidylcholine[2] and cholesterol[3] in chloro-

[1] All cannabinoids used in this study (Table I) were a gift of Professor R. Mechoulam of the Natural Products Laboratory, Hebrew University.

[2] Egg yolk phosphatidylcholine (lecithin, grade 1) was purchased from Makor Chemicals (Jerusalem). It was at least 99% pure, giving one spot on a TLC plate at a 1-μmole loading. It was used without further purification.

[3] Cholesterol (Merck) was recrystalized from ethanol.

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