(2) D. M. Tennent, H. Siegel, M. E. Zanetti, G. W. Kuron, W. H. Ott, and J. F. Wolf, *J. Lipid Res.*, **1**, 469(1960).
(3) T. B. Van Itallie and S. A. Hashim, *Med. Clin. N. Amer.*, **47**, 629(1963).
(4) D. Berkowitz, *Amer. J. Cardiol.*, **12**, 834(1963).
(5) S. A. Hashim and T. B. Van Itallie, *J. Amer. Chem. Soc.*, **192**, 289(1965).
(6) M. E. Zanetti and D. M. Tennent, *Proc. Soc. Exp. Biol. Med.*, **112**, 991(1963).
(7) J. B. Carey, Jr., *J. Clin. Invest.*, **40**, 1028(1961).
(8) W. H. Johns and T. R. Bates, *J. Pharm. Sci.*, **58**, 179(1969).
(9) *Ibid.*, **59**, 329(1970).
(10) *Ibid.*, **59**, 788(1970).
(11) D. G. Gallo, K. R. Bailey, and A. L. Sheffner, *Proc. Soc. Exp. Biol. Med.*, **120**, 60(1965).
(12) K. D. G. Edwards, *Med. J. Aust.*, **2**, 925(1965).
(13) K. D. G. Edwards and M. McCredie, *ibid.*, **4**, 534(1967).
(14) F. Bergman, P. A. Heedman, and W. Vander Linden, *Acta Endocrinol.*, **53**, 256(1966).
(15) R. C. Northcutt, J. N. Stiel, J. W. Hollifield, and E. G. Stant, Jr., *J. Amer. Med. Ass.*, **208**, 1857(1969).
(16) R. Saral and V. L. Spratt, *Arch. Int. Pharmacodyn. Ther.*, **167**, 10(1967).
(17) "Encyclopedia of Industrial Chemical Analysis," vol. 5, S. Snell and J. Hilton, Eds., Interscience, New York, N. Y., 1967, p. 517.
(18) W. Godtfredsen, K. Roholt, and L. Tybring, *Lancet*, **1962**, 928.
(19) I. Langmuir, *J. Amer. Chem. Soc.*, **38**, 2221(1916).
(20) F. Helfferich, "Ion Exchange," McGraw-Hill, New York, N. Y., 1962, p. 166.

ACKNOWLEDGMENTS AND ADDRESSES

Received August 11, 1971, from the *Division of Pharmaceutics, School of Pharmacy, University of Connecticut, Storrs, CT 06268*
Accepted for publication January 17, 1972.
Presented in part to the Basic Pharmaceutics Section, APhA Academy of Pharmaceutical Sciences, San Francisco meeting, March 1971.
Supported in part by Grant 076 from the University of Connecticut Research Foundation, Storrs, CT 06268, and by a National Institutes of Health Fellowship (FO1-GM-45,855).
* Present address: Bristol Laboratories, East Syracuse, N. Y.
▲ To whom inquiries should be directed.

Drug–Cholestyramine Interactions II: Influence of Cholestyramine on GI Absorption of Sodium Fusidate

WILLIAM H. JOHNS* and THEODORE R. BATES▲

Abstract □ The results of a previously reported *in vitro* study provided evidence that the hypocholesterolemic agent, cholestyramine, was capable of strongly and rapidly interacting with the antibiotic, sodium fusidate. Based on these findings, the influence of this pharmacologically important anionic exchange resin on the *in vivo* absorption pattern of sodium fusidate was studied in the rat. Serum antibiotic levels were determined microbiologically as a function of time following oral administration of the antibiotic alone and in the presence of the resin. Concurrent administration of the resin yielded statistically significant reductions in serum drug levels at all experimental time intervals. Peak serum levels of fusidate were found to decrease by 33–77% of control values as the resin-to-drug dose ratio administered was varied from 0.14:1 to 0.72:1. At dose ratios of greater than 1:1 (resin–drug) but far less than the ratio of the average therapeutic, single dose of each drug (*i.e.*, 10:1, cholestyramine–fusidate), there were no detectable serum antibiotic levels. This latter observation indicated that the presence of the insoluble resin in the GI tract totally prevented drug absorption. The time interval between the oral administration of the resin and antibiotic was found also to influence peak serum antibiotic levels.

Keyphrases □ Cholestyramine effect—GI absorption of sodium fusidate, rats □ Sodium fusidate, GI absorption—effect of cholestyramine, rats □ Drug–anionic exchange resin interactions—effect of cholestyramine on GI absorption of sodium fusidate, rats □ Absorption, GI, sodium fusidate—effect of cholestyramine, rats

The water-insoluble, anionic exchange resin, cholestyramine, lowers serum cholesterol levels by binding bile salt anions in the small intestine (1–7). The reduction in bile salt concentration decreases the intestinal absorption of exogenous cholesterol and lipids and increases the hepatic metabolism of endogenous (serum) cholesterol into additional bile salts, which are subsequently bound by the resin. The resin is, therefore, an important therapeutic agent for the treatment of biliary cirrhosis and those conditions normally associated with high blood cholesterol and lipid levels (*e.g.*, atherosclerosis and thrombotic vascular disease).

Since anionic drugs may be concurrently administered during chronic cholestyramine therapy, the possibility exists that a drug–cholestyramine interaction could occur within the GI tract. Such an interaction is of considerable importance clinically because it might result in a decrease in the rate and/or extent of drug absorption and, hence, the onset and/or intensity of drug activity. However, only a limited number of studies have been reported in the literature pertaining to this potential drug–resin therapeutic incompatibility (8–13). These investigations are informative, but their experimental protocols preclude any rigorous interpretation of the *in vivo* absorption data. In addition, no attempts were made by these investigators to determine the influence of relative times of administration of the test drug(s) and cholestyramine on the absorption characteristics of the drug.

It was previously demonstrated *in vitro* (14) that cholestyramine possesses a marked affinity for the steroidal antibiotic, sodium fusidate. This investigation

further revealed that the interaction occurred at a rapid rate and was influenced by a number of physicochemical parameters. It was, therefore, important to determine whether this *direct* interaction could occur *in vivo* and whether it would affect significantly the absorption of fusidate. To accomplish this aim, *in vivo* experiments were conducted to determine the effect of various oral doses of the resin, administered at various time intervals, on the rate and extent of GI absorption of sodium fusidate in the rat.

EXPERIMENTAL

Materials—Sodium fusidate[1] was protected from light at all times and stored under vacuum desiccation until used. Pharmaceutical grade cholestyramine[2] was used as received. The compositions and methods of preparation of the Bacto Antibiotic (Penassay) Media No. 1[3] and the Bacto Penassay Broth Media[3] used in the turbidimetric, microbiological assay procedure are the same as that described in the Difco Manual (15). The medium employed for the cup-plate assay was that suggested by E. R. Squibb (16).

The test organism employed in the turbidimetric assay of fusidate-containing serum samples was *Staphylococcus aureus*[4] (ATCC 6538P); for the cup-plate method, *Corynebacterium xerosis*[1] (SC 3638) was used. Both organisms were received in a lyophilized form and activated by procedures furnished by the suppliers. The rats used in this investigation were healthy, adult, male albino rats of the Sprague-Dawley strain[5] and were maintained on laboratory chow[6] for a 2-week period prior to use. They weighed 300–400 g. at the time of an experiment.

***In Vivo* Absorption Studies**—The rats were fasted for 20 hr. prior to and during the course of the absorption experiments. Water was allowed *ad libitum*.

In studies designed to determine the effect of concurrent oral administration of resin and drug on the absorption pattern of sodium fusidate, the rats were weighed, lightly anesthetized with ether, and dosed with either an aqueous solution of sodium fusidate (500 mg./kg.) and a 0.25% methylcellulose suspension of cholestyramine (214.5 mg./kg.) or with the drug solution and the resin-free methylcellulose suspension vehicle. Administration of the test preparations was facilitated by the use of a curved, bulb-tipped oral dosing needle attached to a 2.0-ml. syringe. The total dosing volume was held constant at 5.0 ml./kg. Following the administration of the dose, the animals were allowed to recover full motor control while being simultaneously checked for any evidence of dose rejection. At selected time intervals, the animals were sacrificed by decapitation, blood samples were collected, and the serum obtained therefrom was frozen until microbiologically assayed for drug content (see *Assay Procedure*).

An identical experimental protocol was followed in the studies involving the effect of cholestyramine dosage levels on the peak serum antibiotic concentrations, except that an oral dose of either 71.5, 214.5, or 357.5 mg./kg. of the resin was administered concurrently with 500 mg./kg. of the antibiotic.

By employing a dosage level of 500 mg./kg. of sodium fusidate and 214.5 mg./kg. of cholestyramine, the influence of time of administration of the resin on the peak antibiotic serum levels was also determined. Dosing intervals of 0, 1, and 2 hr. between the oral administration of the resin and sodium fusidate were investigated using the previously described procedures.

In all *in vivo* absorption studies, at least four animals were sacrificed at each time interval.

Assay Procedure—The microbiological, turbidimetric procedure of Snell and Hilton (17) for the determination of sodium fusidate in pure aqueous media was modified[7] to make it suitable for serum antibiotic determinations. The test organism employed was *S. aureus*, ATCC 6538P. The fusidate-containing serum samples were diluted fivefold with a protein precipitant solution composed of a 1:1 v/v mixture of acetone and Sorenson's pH 7.0 phosphate buffer. The protein-precipitated samples were chilled slightly and subsequently centrifuged at 3000 r.p.m. for 5 min. The chilling step was found to aid significantly in the clean separation of the aqueous phase from the precipitated protein. A 0.5-ml. aliquot of the supernate was then transferred to test tubes containing a 10.0-ml. quantity of a diluted bacterial suspension in Bacto Penassay broth (15). The tubes were sealed, shaken, and incubated at 37° for exactly 10 hr. Subsequently, a 1.0-ml. quantity of a 12% v/v formaldehyde solution was added to each tube and the contents were thoroughly mixed. The addition of formaldehyde served to inhibit further growth of the test organism and thus stabilized the samples. The samples were then allowed to remain at room temperature for 1 hr. prior to measuring the turbidity of the samples at 440 nm.

Blanks containing no drug, but consisting of serum processed in an identical manner, were also prepared. One blank was incubated for 10 hr. at 37°, while the other was immediately stabilized by the addition of formaldehyde. The former blank, which represented the maximum growth of the test organism, and the latter blank, which represented only background bacterial growth, were used to calibrate the colorimeter[8] prior to the measurement of the percent transmittance (% T) of fusidate-containing samples.

The % T values for a series of protein-precipitated, fusidate-containing samples, treated in the manner already outlined, were found to yield a straight line when plotted *versus* the logarithm of the concentration of sodium fusidate (0.10–2.0 mcg./ml.) present. A second calibration plot for fusidate in pure aqueous samples (*i.e.*, devoid of serum) was found to be identical to that obtained in the presence of serum and indicates that serum does not interfere with this assay procedure. It was also established that the acetone–buffer protein precipitant does not affect the growth of the test organism.

On each assay day, known samples were concurrently assayed with unknown samples, and a new calibration plot constructed. At least three determinations were performed on each unknown fusidate-containing serum sample.

As a check on the accuracy of this microbiological assay procedure, five rats were dosed orally with 500 mg./kg. of sodium fusidate and the 3-hr. serum samples were assayed by both the turbidimetric and standard cup-plate (16) assay methods. *C. xerosis*

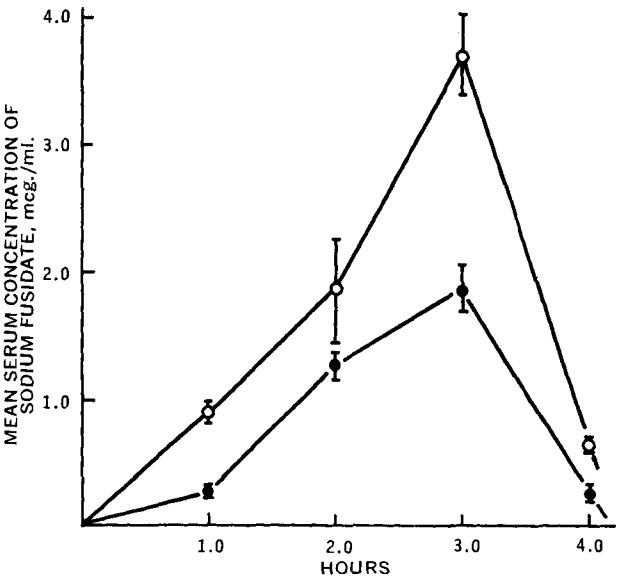

Figure 1—*Serum concentration of sodium fusidate after its oral administration alone (500 mg./kg.) (○) and concurrently with cholestyramine (214.5 mg./kg.) (●). Each data point represents the mean of at least four animals. Vertical bars represent 95% confidence intervals.*

[1] Generously supplied by E. R. Squibb and Sons, New Brunswick, N. J., Batch B14.

[2] Generously supplied by Merck & Co., Inc., Rahway, N. J. (particle size: 100% <100 mesh, 80% <200 mesh).

[3] Obtained from Difco Laboratories, Detroit, Mich.

[4] Obtained from American Type Culture Collections, Rockville, Md.

[5] Obtained from Blue Spruce Farms, Altmont, N. Y.

[6] Purina Lab Chow.

[7] A more detailed description of the procedure can be obtained upon request.

[8] Bausch & Lomb Spectronic 20.

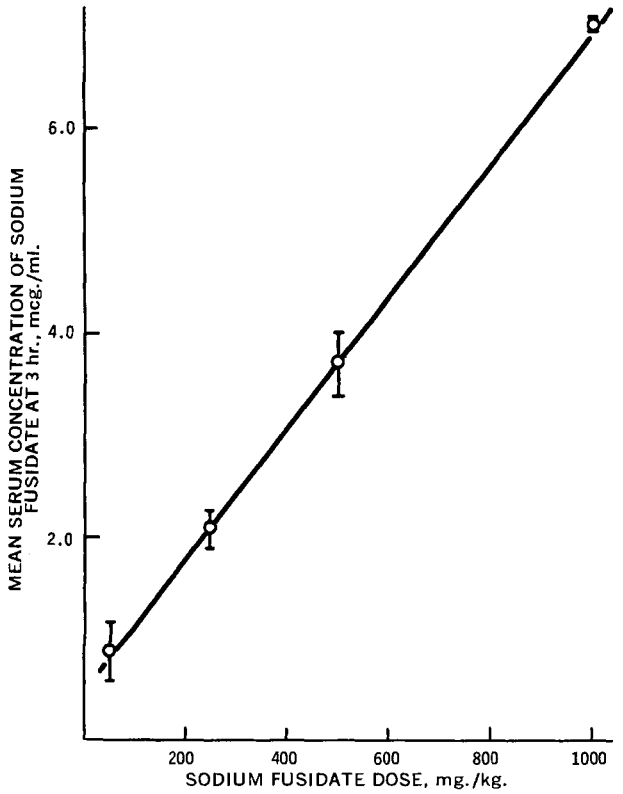

Figure 2—*Relationship between 3-hr. antibiotic serum levels and dose of sodium fusidate orally administered. Each data point represents the mean of at least four animals. Vertical bars represent 95% confidence intervals.*

was employed as the test organism in the latter method. This experiment demonstrated that: (*a*) the assay results obtained with the cup-plate procedure were quite variable as compared to the turbidimetric assay employed in the *in vivo* studies, but (*b*) the serum concentrations determined by the two assay procedures were comparable. As a result of the greater reproducibility and simplicity of the turbidimetric, microbiological method, it was selected as the method of choice.

RESULTS AND DISCUSSION

Effect of Cholestyramine on Fusidate Absorption—The usual oral therapeutic dose of sodium fusidate for human use is 500 mg., administered three or four times daily. On the basis that an average human adult weighs approximately 70 kg., a dose corresponding to 0.014 of the normal single human dose (7.14 mg./kg.) was orally administered to rats, and serum samples were collected as a function of time for 5 hr. However, microbiological analysis of these samples detected no biologically active drug. Upon subsequent dosage level studies, it was established that oral doses equal to or greater than 50 mg./kg. were required to obtain measurable and reproducible blood levels of active drug. In view of the potential reductions of serum drug levels upon coadministration of cholestyramine, it was necessary to employ a sodium fusidate dose of 500 mg./kg. to ensure that assayable blood levels were also obtained in the presence of the resin.

Clinically, cholestyramine is administered over a dosage range of from 5 to 30 g. daily, usually in three divided doses. The average human single-dose ratio of cholestyramine to sodium fusidate would, therefore, be 5 g. to 500 mg. To maintain this 10:1 w/w dose ratio in the rat experiments, an oral cholestyramine dose of 5 g./kg. would have been necessary. However, such a high resin dose presented technical problems in the oral dosing of the animals with a suspension dosage form of the resin. Therefore, it was decided to reduce the dosage level of cholestyramine, rationalizing that if a lower resin-to-antiobiotic dose ratio produced significant reductions in serum antibiotic levels, then the normal ratio would

Table I—Effect of Concurrent Oral Administration of Sodium Fusidate (500 mg./kg.) and Cholestyramine (214.5 mg./kg.) on the Serum Concentrations (mcg./ml.) of Antibiotic as a Function of Time

Hours	Dosage Regimen	Mean Serum Concentration, mcg./ml.	*SE*	Level of Significance[a]	Reduction, %
1	Fusidate	0.892	0.017		
				$p < 0.001$	69.4
	Fusidate + resin	0.273	0.011		
2	Fusidate	1.868	0.131		
				$p < 0.005$	31.5
	Fusidate + resin	1.280[b]	0.042		
3	Fusidate	3.713	0.101		
				$p < 0.001$	49.2
	Fusidate + resin	1.886	0.060		
4	Fusidate	0.668	0.016		
				$p < 0.001$	54.9
	Fusidate + resin	0.301	0.012		

[a] Level of significance determined by Student's *t* test. [b] Represents the mean serum concentration of six animals; all other values represent the mean of four animals.

produce further reductions in drug absorption. For this portion of the investigation, the resin dose administered orally to rats was $^1/_{70}$th of the average *daily* human dose (214.5 mg./kg.). The dose ratio of resin (214.5 mg./kg.) to fusidate (500 mg./kg.), concurrently administered, was, therefore, approximately 0.43:1. By using these doses, it was possible to subject both the control (fusidate alone) and test (fusidate–cholestyramine) blood level data to appropriate biopharmaceutical interpretation.

At selected time intervals following oral administration of fusidate alone (500 mg./kg.) or concurrently with cholestyramine (214.5 mg./kg.), the animals were sacrificed, blood samples were collected, and the serum obtained therefrom was subjected to microbiological assay for active drug content. The results in Table I represent the mean serum antibiotic concentrations of at least four rats at each time interval. It may be noted from the magnitude of the standard errors (*SE*) that, both in the absence and presence of coadministered resin, there exists a very low order of animal-to-animal variation in fusidate serum concentrations, suggesting that the antibiotic is rather uniformly absorbed from the GI tract of the test animals. At each time interval studied, the mean serum antibiotic levels were markedly reduced when cholestyramine was coadministered with sodium fusidate as compared with the cholestyramine-free control animals.

Figure 1 depicts the results shown in Table I. The vertical line associated with each data point represents the 95% confidence interval and demonstrates further that statistically significant differences exist between the two dosage regimens. The area under a typical blood level *versus* time curve (*AUC*) may be used as a measure of the extent of drug absorption across the GI mucosa (18). The following two pharmacokinetic equations relate the *AUC*, from $t = 0$ to $t = 4$ hr., in the presence $[(AUC)_p]$ and absence $[(AUC)_a]$ of cholestyramine to the amount of fusidate ultimately

Table II—Effect of Dose of Cholestyramine on the Serum Concentrations (mcg./ml.) of Sodium Fusidate (500 mg./kg.) at 3 hr. after Concurrent Administration

Resin Dose, mg./kg.	Mean Serum Concentration, mcg./ml.[a]	*SE*	Level of Significance[b]
0.0	3.713	0.101	$p < 0.001$
71.5	2.496	0.089	$p < 0.001$
214.5	1.886	0.060	$p < 0.001$
357.5	0.838	0.076	

[a] Represents the mean serum concentrations of four animals per dosage level. [b] Level of significance determined by Student's *t* test.

Table III—Effect of Time of Administration of Cholestyramine (214.5 mg./kg.) on the Serum Concentrations (mcg./ml.) of Sodium Fusidate at 3 hr. after Antibiotic Administration (500 mg./kg.)

Time between Resin and Antibiotic Administration, hr.	Mean Serum Concentration, mcg./ml.[a]	*SE*	Level of Significance[b]
0	1.886	0.060	
1	2.956	0.067	
2	3.323	0.038	$p < 0.001$
Infinity[c]	3.713	0.101	$p < 0.05$

[a] Represents the mean serum concentrations of four animals per time interval. [b] Level of significance determined by Student's *t* test. [c] Control animals having never received a dose of cholestyramine.

absorbed in 4 hr. under these two experimental conditions:

$$(AUC)_p = \frac{f_p D_o}{V_d K_E} \qquad \text{(Eq. 1)}$$

$$(AUC)_a = \frac{f_a D_o}{V_d K_E} \qquad \text{(Eq. 2)}$$

where V_d is the apparent volume of distribution of the drug in the body; K_E is the apparent first-order, overall elimination rate constant for the drug; D_o is the dose of sodium fusidate orally administered to the test animals; and f_p and f_a are the fractions of the dose of sodium fusidate absorbed from the GI tract in the presence and absence of cholestyramine, respectively. If it is assumed that the pharmacokinetic parameters K_E and V_d are only dependent on the nature of the drug and not the test conditions, then Eq. 2 can be divided into Eq. 1 to yield an expression for the relative biological availability (*RBA*) at 4 hr. of sodium fusidate in the presence of cholestyramine (Eq. 3):

$$\% \; RBA = \frac{(AUC)_p}{(AUC)_a} \times 100 = \frac{f_p}{f_a} \times 100 \qquad \text{(Eq. 3)}$$

Using the trapezoidal rule, the *AUC*, from $t = 0$ to $t = 4$ hr., for the two plots shown in Fig. 1 was measured and the % *RBA* was calculated to be 52.7%. This means that upon concurrent administration of cholestyramine (214.5 mg./kg.), there is a 47.3% reduction in the total amount of fusidate absorbed in 4 hr. from a 500-mg./kg. dose. It is important to bear in mind that this significant reduction occurred with a resin-to-drug dose ratio of only 0.43:1 and that the human dose ratio is 10:1.

When sodium fusidate alone was orally administered to the experimental animals over the dosage range of 50–1000 mg./kg., a linear relationship was observed between the 3-hr. serum antibiotic levels and the dose administered (Fig. 2). Further investigations should provide the type of information necessary to explain the *apparent* nonzero intercept displayed by this graph. Nevertheless, this direct proportionality over the 20-fold dosage range examined suggests that the antibiotic is being absorbed by an apparent first-order, passive-diffusion mechanism. With the aid of this linear plot, it is possible to present an alternate interpretation of the effect of concurrent resin administration on the amount of drug absorbed. The mean serum concentration of about 1.9 mcg./ml. obtained 3 hr. following coadministration of drug and resin (Table I) corresponds to an *apparent* oral antibiotic dose of 208 mg./kg. That is, the presence of the resin (214.5 mg./kg.) causes a 500-mg./kg. oral dose of sodium fusidate to appear as if only a 208-mg./kg. dose had been administered.

Influence of Cholestyramine Dosage Levels on Peak Antibiotic Serum Levels—To establish the effect of varying concurrent doses of cholestyramine on the peak serum antibiotic levels, experiments were conducted in which sodium fusidate (500 mg./kg.) was coadministered with either 71.5-, 214.5-, or 357.5-mg./kg. doses of cholestyramine. These cholestyramine doses correspond to cholestyramine–fusidate dose ratios of approximately 0.14:1, 0.43:1, and 0.72:1, respectively. Serum concentrations of drug were determined 3 hr. after drug administration. It is apparent from an examination of the data listed in Table II that as the resin dose is increased from 0 to 357.5 mg./kg., there is a progressive and significant decrease in the magnitude of the peak serum antibiotic levels and, hence, in the amount of drug in the body. The percent reductions in the control serum concentrations were 33, 49, and 77% upon coadministration of resin doses of 71.5, 214.5, and 357.5 mg./kg., respectively. These studies provide additional evidence to support the fact that sodium fusidate is capable of strongly interacting *in vivo* with cholestyramine, even at low resin-to-drug dose ratios and in the presence of competing physiologic anions. At resin-to-drug ratios greater than 1:1 but much less than the human dose ratio of 10:1, *no serum levels of active antibiotic were detectable.* This latter observation suggests that the presence of the resin in the GI tract completely prevented the absorption of the antibiotic.

Influence of Time of Resin Administration on Peak Drug Serum Levels—As a general rule, with a substance (*e.g.*, cholestyramine) whose presence in the fluids of the GI tract interferes with the normal GI absorption of a therapeutic agent, it would be logical to avoid concurrent administration. Therefore, the objective of this portion of the investigation was to test the effect of time of administration of resin on the absorption rate of fusidate. For these studies, animals were predosed with resin (214.5 mg./kg.) at 0, 1, and 2 hr. prior to the administration of the antibiotic (500 mg./kg.), and the peak serum drug levels were subsequently determined. The results of this study, together with the antibiotic serum level obtained after the administration of sodium fusidate alone (*i.e.*, animals having never received a dose of resin), are presented in Table III. At dosing time intervals of 0 and 1 hr., control serum antibiotic levels are reduced by 49 and 20%, respectively. This indicates that if sodium fusidate is administered 1 hr. following the administration of cholestyramine, there would still be significant quantities of resin present in the GI tract capable of interacting with the drug. The relatively small, but statistically significant, reduction observed at the 2-hr. dosing interval (*i.e.*, 10%) suggests that the adsorbate has become physically separated from the adsorbent in the GI tract of the rat. However, at higher resin-to-drug dose ratios the resin might produce a more pronounced inhibitory effect on the absorption of fusidate at this dosing interval.

Quantitative extrapolation of these *in vivo* findings in rats to those expected in humans should be done with extreme care. Unlike humans, the rat does not possess a gallbladder for bile storage. As a result, bile salt-containing bile would continuously flow into the small intestinal lumen of the rat; in the intestinal tract of humans, high bile salt concentrations are only intermittently present. Therefore, the potential competitive effects of bile salt anions on the *in vivo* binding of sodium fusidate to cholestyramine may occur to a different extent in the rat as compared to man. With this in mind and the fact that a resin-to-drug single-dose ratio of 10:1 would be administered to man, it is reasonable to propose that upon concurrent oral administration of sodium fusidate and cholestyramine to man, marked reductions in both rate and extent of drug absorption would occur. In addition, more than 2 hr. between the oral administration of the resin and the antibiotic may be required in humans before the effect of cholestyramine on the absorption pattern of sodium fusidate becomes of no therapeutic significance. In this connection, single and multiple oral dose studies are currently being conducted to ascertain the influence of cholestyramine on the absorption of sodium fusidate in man.

REFERENCES

(1) S. S. Bergen, T. B. Van Itallie, D. M. Tennent, and W. N. Sebrell, *Circulation*, **20,** 981(1959).

(2) D. M. Tennent, H. Siegel, M. E. Zanetti, G. W. Kuron, W. H. Ott, and J. F. Wolf, *J. Lipid Res.*, **1,** 469(1960).

(3) T. B. Van Itallie and S. A. Hashim, *Med. Clin. N. Amer.*, **47,** 629(1963).

(4) D. Berkowitz, *Amer. J. Cardiol.*, **12,** 834(1963).

(5) S. A. Hashim and T. B. Van Itallie, *J. Amer. Chem. Soc.*, **192,** 289(1965).

(6) M. E. Zanetti and D. M. Tennent, *Proc. Soc. Exp. Biol. Med.*, **112,** 991(1963).

(7) J. B. Carey, Jr., *J. Clin. Invest.*, **40,** 1028(1961).

(8) D. G. Gallo, K. R. Bailey, and A. L. Sheffner, *Proc. Soc. Exp. Biol. Med.*, **120,** 60(1965).

(9) K. D. G. Edwards, *Med. J. Aust.*, **2,** 925(1965).

(10) K. D. G. Edwards and M. McCredie, *ibid.*, **4,** 534(1967).

(11) F. Bergman, P. A. Heedman, and W. Vander Linden, *Acta Endocrinol.*, **53**, 256(1966).
(12) R. C. Northcutt, J. N. Stiel, J. W. Hollifield, and E. G. Stant, Jr., *J. Amer. Med. Ass.*, **208**, 1857(1969).
(13) R. Saral and V. L. Spratt, *Arch. Int. Pharmacodyn. Ther.*, **167**, 10(1967).
(14) W. H. Johns and T. R. Bates, *J. Pharm. Sci.*, **61**, 730(1972).
(15) "Difco Manual," 9th ed., Difco Labs., Detroit, Mich., 1953, pp. 203–206.
(16) T. B. Platt and W. E. Brown, E. R. Squibb and Sons, New Brunswick, N. J., personal communication.
(17) "Encyclopedia of Industrial Chemical Analysis," vol. 5, S. Snell and J. Hilton, Eds., Interscience, New York, N. Y., 1967, pp. 513, 514.
(18) G. A. Portman, in "Current Concepts in the Pharmaceutical Sciences: Biopharmaceutics," J. Swarbrick, Ed., Lea & Febiger, Philadelphia, Pa., 1970, p. 18 *ff*.

ACKNOWLEDGMENTS AND ADDRESSES

Received August 11, 1971, from the *Division of Pharmaceutics, School of Pharmacy, University of Connecticut, Storrs, CT 06268*
Accepted for publication January 17, 1972.
Presented in part to the Basic Pharmaceutics Section, APhA Academy of Pharmaceutical Sciences, San Francisco meeting, March 1971.
Supported in part by Grant 076 from the University of Connecticut Research Foundation, Storrs, CT 06268, and by a National Institutes of Health Fellowship (FO1-GM-45,855).
* Present address: Bristol Laboratories, East Syracuse, N. Y.
▲ To whom inquiries should be directed.

Physiological Disposition of Fenoprofen in Man II: Plasma and Urine Pharmacokinetics after Oral and Intravenous Administration

ALAN RUBIN▲, BRUCE E. RODDA, PATRICIA WARRICK, ANTHONY S. RIDOLFO, and CHARLES M. GRUBER, Jr.

Abstract □ Two studies of *dl*-2-(3-phenoxyphenyl)propionic acid or fenoprofen are described. In these studies, the pharmacokinetic parameters of fenoprofen administered orally and intravenously were compared first and then urine and plasma kinetics were compared. The results indicate that: (*a*) fenoprofen is rapidly and efficiently absorbed from the GI tract; (*b*) fenoprofen is extensively metabolized, and it and its metabolites are rapidly eliminated from the body by the kidneys; and (*c*) good agreement exists between plasma and urinary kinetics.

Keyphrases □ Fenoprofen—urinary and plasma kinetics compared after oral and intravenous administration, major urinary metabolites identified, man □ *dl*-2-(3-Phenoxyphenyl)propionic acid—urinary and plasma kinetics compared after oral and intravenous administration, major urinary metabolites identified, man □ Urinary kinetics, fenoprofen—compared to plasma kinetics, major metabolites identified, man □ Fenoprofen glucuronide—major urinary metabolite of fenoprofen, man □ 4′-Hydroxyfenoprofen glucuronide—major urinary metabolite of fenoprofen, man

Results of initial pharmacokinetic studies of *dl*-2-(3-phenoxyphenyl)propionic acid or fenoprofen were reported previously (1). In that study, fenoprofen was administered orally to human subjects as the sodium or calcium salt, and a rapid appearance of fenoprofen in plasma was observed. The plasma disposition curve was compatible with a two-compartment open model. However, a significant peripheral ("tissue") compartment was not detected in that case, and a one-compartment model satisfactorily described the plasma kinetics of orally administered fenoprofen. In preliminary studies wherein sodium fenoprofen was administered intravenously, a two-compartment model seemed more appropriate than a one-compartment model.

This paper reports the results of two studies undertaken to extend the comparison of the pharmacokinetic parameters of fenoprofen administered by both the oral and intravenous routes and to provide kinetic information about the urinary excretion pattern of fenoprofen and its metabolites in man.

EXPERIMENTAL

Subjects—Two study designs were used. For each study, four male subjects[1] were admitted to the clinical research ward and examined as described in an earlier publication (1). The volunteers were between the ages of 22 and 31 years, ranging in weight from 65.8 to 69.0 kg. (145 to 152 lb.) and in height from 170.2 to 185.4 cm. (5 ft. 7 in. to 6 ft. 1 in.). Five subjects were Caucasian, and three (A.C., R.W., and M.B.) were Negro. Informed consent was obtained from each subject before participation in a study.

Study 1: Oral and Intravenous Administration—*Design*—Orally administered doses of 250 mg. fenoprofen[2] were assigned to each of four subjects according to the experimental design used previously (1). One week after the oral crossover was completed, all subjects were given 250 mg. fenoprofen intravenously. All medications were administered at 6:00 a.m. after an overnight fast; food was withheld for an additional 2 hr. After receiving the dose, the men were unrestricted in movement and position. Smoking and water consumption were permitted. The subjects were observed carefully during the tests, and no adverse effects were observed.

Sodium fenoprofen for oral administration was formulated in a single capsule; calcium fenoprofen was formulated in two cap-

[1] From the Indiana Reformatory.
[2] All data are expressed in terms of the free acid (fenoprofen); corrections have been made for molecular weight differences.