(3) D. K. Lawrence and E. W. Gill, *Mol. Pharmacol.*, **11**, 595 (1975).
(4) I. Tamir, D. Lichtenberg, and R. Mechoulam, in "Nuclear Magnetic Resonance Spectroscopy in Molecular Biology," B. Pullman, Ed., D. Riedel, Dordene, Holland 1978, p. 405.
(5) N. O. Petersen and S. I. Chan, *Biochemistry*, **16**, 2657 (1977).
(6) D. Lichtenberg, N. O. Petersen, J. L. Girardet, M. Kainosho, P. A. Kroon, C. H. A. Seiter, G. W. Feignenson, and S. I. Chan, *Biochim. Biophys. Acta*, **382**, 10 (1975).
(7) T. E. Thompson and C. Huang, in "Physiology of Membrane Disorders," T. E. Andreali, J. F. Hoffman, and D. D. Fanesti, Eds., Plenum, London and New York, N.Y., 1978 p. 27.
(8) Y. Barenholz, S. Amselem, and D. Lichtenberg, *FEBS Lett.*, **99**, 210 (1979).
(9) R. Mechoulam and H. Edery, in "Marijuana," R. Mechoulam, Ed., Academic, New York, N.Y. and London, 1973, p. 118.
(10) M. F. Brown, G. P. Miliganich, and E. A. Dratz, *Biochemistry*, **16**, 2640 (1977).
(11) P. L. Harris and E. R. Thornton, *J. Am. Chem. Soc.*, **100**, 6737 (1978).
(12) H. Wennerstrom, *Chem. Phys. Lett.*, **18**, 41 (1973).
(13) C. F. Schmidt, D. Lichtenberg, and T. E. Thompson, *Biochemistry*, **20**, 4732 (1981).

Ilana Tamir ˣ
Department of Natural Products
School of Pharmacy
Hebrew University
Jerusalem 91000, Israel

Dov Lichtenberg
Department of Pharmacology
School of Pharmacy
Hebrew University
Jerusalem 91000, Israel

Received March 5, 1982.
Accepted for publication October 14, 1982.
This work was supported in part by the Israel center for psychobiology (Charles E. Smith family foundation).
Professor R. Mechoulam is acknowledged for his interest and the gift of the cannabinoids.

Experimental Evidence for Concentration-Dependent Plasma Protein Binding Effects on the Apparent Half-Lives of Restrictively Cleared Drugs

Keyphrases □ Plasma protein binding—concentration dependent, effects on the apparent half-lives of restrictively cleared drugs □ Pharmacokinetics—experimental evidence for concentration-dependent plasma protein binding effects on the apparent half-lives of restrictively cleared drugs □ Disopyramide—experimental evidence for concentration-dependent plasma protein binding effects on the apparent half-lives of restrictively cleared drugs

To the Editor:

Several early publications have addressed the influence of concentration-dependent (nonlinear) plasma protein binding on the pharmacokinetics of drugs which undergo restrictive clearance (elimination is proportional to free, unbound drug in serum or plasma) (1–3). Relatively simple models which excluded the influence of tissue binding were used to simulate plasma concentrations of free drug (C_f) and of total (free plus protein bound) drug (C_t). More recently, these earlier studies were extended (4) to include models which consider tissue binding and drugs which undergo nonrestrictive clearance (elimination is proportional to total drug in plasma). It was reported that log concentration–time plots of the elimination phase of C_f and C_t for drugs which undergo restrictive clearance may be linear, concave, or convex, depending on the extent to which drugs are bound to plasma and tissue protein (4).

Experimental data verifying the results of these simulation studies in humans are difficult to obtain because of the absence of a model drug which demonstrates concentration-dependent plasma protein binding at plasma concentrations achieved following the administration of safe, therapeutic doses and the absence of an analytical method sensitive enough to accurately measure plasma concentrations for extended time periods following the administration of safe doses of the drug. This is because the behavior of the decay of plasma concentration–time plots may require a wide concentration range to be expressed fully.

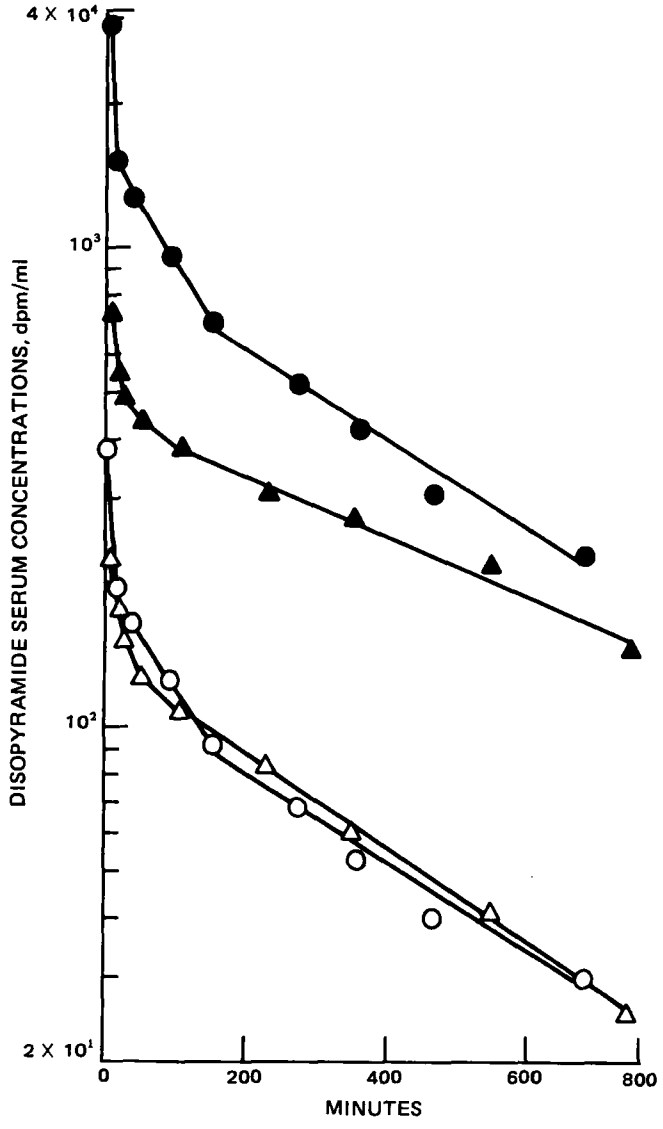

Figure 1—*Free (○,△) and total (●,▲) serum concentrations of [¹⁴C]disopyramide at various times following the administration of carbon 14 alone (○,●) and the simultaneous administration of [¹⁴C]disopyramide and the oral dose (△,▲). In each case a linear regression line was drawn through the last 5 serum concentration–time points.*

0022-3549/83/0400-0461$01.00/0

Table I—Pharmacokinetic Parameters of [^{14}C]Disopyramide

	Total (Bound + Free)				Free		
	Dose dpm	Dose[a], $AUC_{0\to\infty}$ ml/min	$t_{1/2}$[b], hr	V_{ss}[c], liters	Dose[a], $AUC_{0\to\infty}$ ml/min	$t_{1/2}$[b], hr	V_{ss}[c], liters
Tracer alone	2.09×10^7	42.7	5.3	18.0	329	5.3	138
Tracer plus oral dose	1.93×10^7	62.8	7.9	41.3	308	5.0	121

[a] *AUC* refers to the area under the [^{14}C]disopyramide concentration time curve calculated by trapezoidal rule. [b] The parameter $t_{1/2}$ refers to half-life and was calculated by: $t_{1/2} = 0.693/\lambda_z$, where λ_z is the apparent terminal slope of the serum concentration–time plot. [c] The parameter V_{ss} refers to volume of distribution at steady state.

Recent studies show that α-1-acid glycoprotein accounts for most of the plasma protein binding of disopyramide in humans (5, 6) and that plasma protein binding is concentration-dependent following the administration of therapeutic doses to patients (7) and normal subjects (6, 8). Furthermore, the clearance of disopyramide is restrictive (6–8), and pharmacodynamic activity is proportional to C_f (6). The systemic availability of disopyramide has been compared in a small group of patients and normal subjects by the administration of a 150-mg oral dose followed by an intravenous injection of 10 μCi of [^{14}C]disopyramide (specific activity = 4.259 mCi/mmole) (9) 1 hr later. At a later time, one of the normal subjects received a second tracer dose of [^{14}C]disopyramide alone. Prior to each dosing plasma was collected, and the binding of disopyramide was characterized as previously described (6).

Figure 1 compares the serum concentrations of [^{14}C]-disopyramide following the simultaneous administration of tracer and the oral dose with those following administration of just the tracer dose. Values of C_t as determined by liquid chromatography (10) following the simultaneous administration of tracer and oral disopyramide ranged between 2.47 (7.26×10^{-6} *M*) and 0.98 (2.89×10^{-6} *M*) μg/ml, respectively. The corresponding free fractions (*fu*) of the drug in plasma ranged between 0.32 and 0.18, respectively. Values of C_t, which were calculated from the specific activity, ranged between 9.4×10^{-9} and 4.0×10^{-7} *M* following administration of the tracer dose alone. The *fu* at these C_t values was 0.13 and was concentration independent. Table I compares the clearances of free ($Div/AUC^{free}_{0\to\infty}$) and total ($Div/AUC^{tot}_{0\to\infty}$) disopyramide concentrations, their respective half-lives ($t_{1/2}$), and steady-state volumes of distribution (V_{ss}). These data show that the pharmacokinetics of disopyramide calculated based on total serum concentrations are drug concentration dependent, while those based on free disopyramide are concentration independent at the concentration studied (300-fold range). The data also show that the $t_{1/2}$ of C_t is prolonged as compared with the $t_{1/2}$ calculated from C_f values when the plasma protein binding of disopyramide is drug concentration dependent. These data indicate further that as C_f and C_t decrease as a consequence of elimination following the simultaneous administration of the tracer and the oral dose, *fu* decreases to a limiting value, at which time the decay in C_f and C_t are parallel. Thus, the decay in C_t is convex over the drug concentration range achieved in this study.

It was reported (4) that in the absence of tissue binding, the decay in C_t–time data is convex when the ratio of the binding capacity of the drug-binding protein to its dissociation constant is low (low and high ratios of 9 and 100, respectively, were reported) or when tissue binding is moderate to extensive (tissue free fractions of 0.5 and 0.1, respectively, were reported). The ratio of the binding constants characterizing the interaction between disopyramide and α-1-acid glycoprotein in the serum of the subject was ~7.0, and the tissue free fraction was ~0.05 (11). Thus, the convex curvature in the terminal disopyramide C_t–time plot is consistent with previous findings (4) and provides at least some experimental support for the simulation studies described therein. Employment of tracer dose techniques provides another method (12) of examining the influence of concentration-dependent plasma protein binding on the pharmacokinetics of drugs.

(1) E. Kruger-Theimer, W. Diller, and P. Bünger, *Antimicrob. Agents Chemother.*, **5**, 183 (1965).

(2) B. K. Martin, *Nature (London)*, **1965**, 959.

(3) J. J. Coffey, F. J. Bullock, and P. T. Schoenemann, *J. Pharm. Sci.*, **60**, 1623 (1971).

(4) P. J. McNamara, G. Levy, and M. Gibaldi, *J. Pharmacokinet. Biopharm.* **7**, 195 (1979).

(5) J. J. Lima and L. B. Salzer, *Biochem. Pharmacol.*, **30**, 2633 (1981).

(6) J. J. Lima, H. Boudoulas, and M. Blanford, *J. Pharmacol. Exp. Ther.*, **219**, 741 (1981).

(7) P. J. Meffin, E. W. Roberts, R. A. Winkle, S. Harapat, F. A. Peters, and D. C. Harrison, *J. Pharmacokinet. Biopharm.*, **7**, 29 (1979).

(8) D. B. Haughey and J. J. Lima, *Biopharma Drug Disp.*, in press.

(9) D. B. Haughey, C. V. Leier, and J. J. Lima, *Clin. Pharmacol. Ther.*, **31**, 233 (1982).

(10) J. J. Lima, *Clin. Chem.*, **25**, 405 (1979).

(11) M. Gibaldi, G. Levy, and P. J. McNamara, *Clin. Pharmacol. Ther.*, **24**, I (1978).

(12) P. J. McNamara, J. T. Slattery, M. Gibaldi, and G. Levy, *J. Pharmacokinet. Biopharm.*, **7**, 397 (1979).

John J. Lima

Division of Pharmacy Practice
College of Pharmacy and
Division of Cardiology
College of Medicine
The Ohio State University
Columbus, OH 43210

Received March 1, 1982.

Accepted for publication October 28, 1982.

Supported by Grant GM-28420-01 from the National Institute of General Medical Sciences, National Institutes of Health.