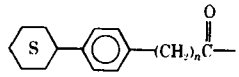
Table II—Prednisolone C_{21}-Esters of



n	Melting Point
0	242°
1	202–203°
2	190–191°
3	160–162°

para-substituted phenyl and substituted alkyl moieties and the sum of ΔH_s contributions of each functional group present in the R moiety in the ester. Some important ΔH_s values used in our study are shown in Table I (2). A straightforward additivity rule was applied in calculating ΔH_s contributions from the R group. For instance, for the *p*-benzamidophenyl ester of dinoprostone, the ΔH_s contribution of the phenolic moiety is the sum of 2 × 10.0 for phenyl, 6.5 for >C=O, and 5.6 for —NH—, giving 32.1 kcal/mole.

The relationship between the melting points of 22 dinoprostone C_1-esters and 25 dinoprost C_1-esters and the ΔH_s contribution of the R groups is shown in Fig. 1. A least-squares analysis resulted in a slope of 3.04°/kcal/mole of ΔH_s and an intercept, the physically meaningless melting point of the prostaglandins without a proton at C_1, of 26.7°. Although the melting points of dinoprostone (63.4°) and dinoprost (~35°) are rather removed, visual inspection of the plot gives no indication of two separate relationships. The correlation coefficient was 0.851, which is surprisingly good considering all of the assumptions and approximations involved.

The reasonably good correlation observed appears to reflect the validity of our basic assumption that the prostaglandin group contribution to the melting point is constant. Both the E and F series of prostaglandins have substantial intramolecular interactions between two alkyl chains in the solid state (4) and the C_1-substituents probably do not disturb this basic molecular conformation, resulting in a rather constant crystal packing arrangement of the esters.

Of the C_1-esters prepared, about 20% of the liquid esters are predicted to be solids. Since it was difficult to crystallize many of the esters (up to 2 weeks of manipulation was sometimes required), many of these liquid esters possibly could be crystallized under proper conditions. Although we attribute the deviations observed largely to assumptions in the analysis, they could originate from polymorph formation. All of the esters were carefully purified by silica gel column chromatography, and it is unlikely that the deviations were due to impurities.

The present analysis applies mainly to terminally substituted prodrugs such as the prostaglandin C_1-esters. Application of the additivity principle may not hold for other prodrugs where the modification is at an internal position. Under these conditions, the assumption that ΔS_f is constant is not warranted due to a significantly higher entropy contribution (mainly rotational) brought about by modification at an internal position. This situation is illustrated by the *decrease* in melting points observed in some prodrug series using internal homologation (Table II). This decrease probably is due to an additional increase in entropy, which would lower T_m (Eq. 1). Therefore, our group contribution approach is not applicable in this case.

Cautious application of the treatment to terminally substituted prodrugs of relatively constant size should be of great use in analyzing the influence of structure on melting points. In these special prodrug series, the melting points may now allow *a priori* estimates of the influence of structure on other related physical properties.

(1) W. Morozowich, T. O. Oesterling, W. L. Miller, C. F. Lawson, J. R. Weeks, R. G. Stehle, and S. L. Douglas, *J. Pharm. Sci.*, **68**, 833 (1979).

(2) A. Bondi, "Physical Properties of Molecular Crystals, Liquids, and Glasses," Wiley, New York, N.Y., 1968, pp. 139–196, 452–488.

(3) "Handbook of Chemistry and Physics," 49th ed., R. C. Weast, Ed., Chemical Rubber Co., Cleveland, Ohio, 1968, p. C-686.

(4) J. W. Edmonds and W. L. Daux, *Prostaglandins*, **5**, 275 (1974). *J. Am. Chem. Soc.*, **97**, 413 (1975) and references cited therein.

(5) G. Carenini, M. Carissimi, R. D'Ambrosio, and R. Ravenna, *Farmaco Ed. Sci.*, **30**, 167 (1975).

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Noncompartmental Determination of the Steady-State Volume of Distribution

Keyphrases □ Volume of distribution—steady state, noncompartmental determination, calculations □ Pharmacokinetics—steady-state volume of distribution, noncompartmental determination, calculations

To the Editor:

Up to the present time, the use in pharmacokinetics of the steady-state volume of distribution (Vd_{ss}) has been limited to specific compartmental mammillary models. However, mathematical methods usually associated in medicine with the use of indicator dilution curves to determine cardiac output and with the determination of mean residence time of endogenous substances following radiolabeled tracer injection permit Vd_{ss} determination without the assumption of a specific compartment model (or its analog, the assumption of a specific number of exponential functions).

Riggs (1) introduced the use of an overall volume of distribution term, Vd_{ss}, defined specifically with respect to the two-compartment open model, with elimination taking place from the central compartment:

$$Vd_{ss} = \left(1 + \frac{k_{12}}{k_{21}}\right) V_1 \qquad \text{(Eq. 1)}$$

The steady-state volume of distribution, as defined by Riggs (1), equals the total quantity of drug in the body divided by the concentration in the reference region of the central compartment when these measurements are taken when the tissue compartment contains the maximum amount of drug. Riegelman *et al.* (2) presented an exten-

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sive discussion of the steady-state volume of distribution and the advantages of using this term when describing multicompartment distribution of drug in the body. They preferred the use of steady-state volume of distribution because this term (Eq. 1) is independent of the elimination rate constant. However, Benet and Ronfeld (3) pointed out that Vd_{ss} is independent of the elimination rate constant only when elimination occurs exclusively from the central compartment.

Gibaldi (4) showed that under steady-state conditions, as in a zero-order infusion, Vd_{ss} may be calculated without establishing the appropriate model or determining the distribution and elimination rate constants if plasma concentrations are measured from time zero until steady state is reached and then following termination of the infusion until no drug remains in the body. Once again, this determination is only valid when elimination occurs exclusively from the central compartment of a multicompartmental model (3). In practice, Vd_{ss} determinations in the literature (at least up to 1977) have been based on calculations involving microconstants and the volume of the central compartment such as is given in Eq. 1 for a two-compartment model.

Wagner (5) and van Ginneken (6) showed that Vd_{ss} may be determined directly from the coefficients and exponents of a polyexponential equation fit to the plasma concentration time data:

$$Vd_{ss} = \frac{\text{dose} \sum \left(\frac{A_i}{\lambda_i^2}\right)}{\left(\sum \frac{A_i}{\lambda_i}\right)^2} \qquad \text{(Eq. 2)}$$

where A_i defines the coefficients and λ_i defines the exponents in a polyexponential equation such as Eq. 3, an example for data requiring three exponential terms to describe the plasma concentration–time curve:

$$C_p = A_1 e^{-\lambda_1 t} + A_2 e^{-\lambda_2 t} + A_3 e^{-\lambda_3 t} \qquad \text{(Eq. 3)}$$

Wagner (5) stated that the Vd_{ss} value calculated using Eq. 2 applies only to the general model where elimination occurs exclusively from the central compartment. He suggested: "However, because of the dilemma of not knowing from which compartment there are exit rate constants to outside the body, use of Equation 2 will at least make all authors homogeneous in their approach." However, when the distribution constants for the model are determined as was done previously for calculations of Vd_{ss} (2) or when the coefficients and exponentials of a polyexponential equation are utilized (5, 6), the investigator is still required to propose some model and to determine a number of constants and coefficients either by graphical means or through the use of a nonlinear least-squares computer program. We describe a noncompartmental method of determining Vd_{ss} utilizing area under the curve determinations exclusively.

In indicator dilution studies, basic mathematical methodology is based on the Stewart–Hamilton theorems for flow and volume (7, 8). Perl and Samuel (9) generalized these theorems to the case of multiple input channels with partially labeled input in a study of the input rate and traced mass of body cholesterol. More recently, Oppenheimer *et al.* (10, 11) adapted these principles to facilitate calculation of conventionally used parameters in the study of iodothyronine metabolism and distribution. In these endocrinology studies (9, 10), the basic parameter determined using noncompartmental analysis is the mean residence time, $\bar{t}$, following an intravenous bolus dose:

$$\bar{t} = \frac{\int_0^\infty tC_p\,dt}{\int_0^\infty C_p\,dt} = \frac{AUMC_{0\to\infty}}{AUC_{0\to\infty}} \qquad \text{(Eq. 4)}$$

The denominator of Eq. 4 is the familiar (in pharmacokinetic terms) area under the plasma concentration–time curve (AUC) from time zero to infinity. The numerator is the area under the first moment of the plasma curve ($AUMC$), *i.e.*, the area under the curve of the product of time, t, and plasma concentration, C_p, from time zero to infinity.

Oppenheimer *et al.* (10) defined the mean residence time in terms of a rate constant, k, designated as the fraction of the total pool of iodothyronine irreversibly removed per unit of time:

$$k_{ss} = 1/\bar{t} \qquad \text{(Eq. 5)}$$

We have added a subscript to this rate constant to indicate that it is a constant *defined at steady state*. This k_{ss} should not be confused with either the rate constant describing elimination from the plasma compartment of a pharmacokinetic model or the rate constant describing the terminal plasma disappearance curve in a multicompartment model.

At steady state, the product of the rate constant k_{ss} and the volume Vd_{ss} may be defined as the total body clearance:

$$\text{total body clearance} = k_{ss} Vd_{ss} \qquad \text{(Eq. 6)}$$

since clearance may be defined as a product of the rate constant describing the total drug fraction irreversibly removed from the body as a function of time, multiplied by the volume of distribution on which that rate constant is operating. That is, Eq. 6 is identical to other definitions of total body clearance such as $k_{10}V_1$ and βVd_β (5). The total body clearance is a model-independent term and may be calculated for an intravenous injection as dose divided by the area under the plasma concentration–time curve from time zero to infinity. Substituting this quotient into Eq. 6, substituting the area relations for k_{ss} defined in Eq. 4 and 5, and rearranging yield the noncompartmental method for determining the steady-state volume of distribution:

$$Vd_{ss} = \frac{\text{dose}\left[\int_0^\infty tC_p\,dt\right]}{\left[\int_0^\infty C_p\,dt\right]^2} = \frac{\text{dose}[AUMC_{0\to\infty}]}{[AUC_{0\to\infty}]^2} \qquad \text{(Eq. 7)}$$

This calculation will be valid only when the underlying assumptions discussed by Perl and Samuel (9) are valid. In terms of pharmacokinetic analysis, the two most important of these assumptions are: (*a*) the system must respond linearly, *i.e.*, follow first-order kinetics, and (*b*) the exit of tracer or drug from the body must be directly from the measured site, *i.e.*, the plasma or central compartment

Table I—Sample Noncompartmental Vd_{ss} Calculation

Hours	C_p, μg/ml	tC_p, μg hr/ml	$\int_0^t C_p\,dt$ $[AUC_{0\to t}]$, μg hr/ml	$\int_0^t tC_p\,dt$ $[AUMC_{0\to t}]$, μg hr²/ml
0.0	100.00	0		
0.0833	78.90	6.57	7.40[a]	0.3[a]
0.1667	65.01	10.83	13.4	1.0
0.25	55.86	13.97	18.4	2.1
0.5	43.14	21.57	30.6	6.5
0.75	39.28	29.46	40.9	12.9
1.0	37.92	37.92	50.5	21.3
1.5	36.83	55.24	69.2	44.6
2.0	36.08	72.16	87.4	76.5
3.0	34.68	104.04	122.8	164.6
5.0	32.04	160.22	189.5	428.8
7.0	29.61	207.27	251.2	796.3
9.0	27.36	246.23	308.1	1249.8
12.0	24.30	291.61	385.6	2056.6
18.0	19.17	345.07	515.6	3982.9
24.0	15.12	362.97	618.5	6107.0
36.0	9.41	338.84	763.7	10360.0
48.0	5.86	281.18	855.3	14080.1
72.0	2.27	163.36	947.8	19372.2
96.0	0.88	84.37	985.6	22344.9
120.0	0.34	40.85	1000.2	23847.5
∞	0.0	0.0	1008.8	25099.1

[a] All integrals approximated by using the linear trapezoidal rule. Vd_{ss} = (dose $[AUMC_{0\to\infty}]/[AUC_{0\to\infty}]^2$) = 12,332 ml.

in a pharmacokinetic analysis. However, these are the same assumptions necessary to calculate Vd_{ss} by the methods described previously (Eqs. 1 and 2).

Wagner (5) compared Vd_{ss} calculation by Eq. 2 with Eq. 1 by simulating a bolus intravenous injection of a drug following two-compartment kinetics with elimination exclusively from the central compartment. Equation 8 contains the coefficients and exponentials utilized by Wagner to describe plasma concentrations following an intravenous 500-mg bolus dose (5):

$$C_p = 60.9545e^{-5.0605t} + 39.0459e^{-0.03952t} \qquad \text{(Eq. 8)}$$

Using Eq. 8, we calculated plasma concentration values over a 120-hr period (Table I). Column 1 indicates time in hours; column 2 is plasma concentration in micrograms per milliliter; column 3 is the product of columns 1 and 2. The linear trapezoid rule was utilized to calculate areas under the curve. Column 4 tabulates area under the plasma concentration *versus* time curve, and column 5 tabulates the values under the moment curve. The area under the plasma concentration–time curve from $C_{p_{last}}$ until time infinity was estimated using:

$$\int_{t_{last}}^{\infty} C_p\,dt = \frac{C_{p_{last}}}{\lambda_2} \qquad \text{(Eq. 9)}$$

where λ_2 is the terminal rate constant, *i.e.*, $-0.03952\ hr^{-1}$ in Eq. 8. Likewise, the area under the concentration–moment curve from $C_{p_{last}}$ to time infinity is given by Eq. 10 (11):

$$\int_{t_{last}}^{\infty} tC_p\,dt = \frac{t_{last}C_{p_{last}}}{\lambda_2} + \frac{C_{p_{last}}}{(\lambda_2)^2} \qquad \text{(Eq. 10)}$$

By using the coefficients and exponents of Eq. 8 to calculate AUC (*i.e.*, $A_1/\lambda_1 + A_2/\lambda_2$) and $AUMC$ (*i.e.*, $A_1/\lambda_1^2 + A_2/\lambda_2^2$), values of 1000.0 and 25,002.5 are obtained (5). The difference between these values and the summations in columns 4 and 5 of Table I results from the errors inherent in estimating areas using the linear trapezoid rule. If the logarithmic trapezoid rule (12, 13), Eq. 11[1], is used to estimate AUC, then a value of 1000.5 is obtained:

$$AUC_{t_1\to t_2} = \frac{(y_1 - y_2)\,\Delta t}{\ln (y_1/y_2)} \qquad \text{(Eq. 11)}$$

When the time infinity values for AUC and $AUMC$ from Table I are substituted into Eq. 7 with a dose of 500,000 μg, Vd_{ss} is calculated to be 12,332 ml, which is ~1.3% less than the actual 12,500-ml value. (Use of the logarithmic AUC calculation results in a value of 12,537 for Vd_{ss}.) These Vd_{ss} calculations were carried out using simple area under the curve determinations, independent of the assignment of a particular compartment model or a multiexponential computer data fit. Unfortunately, if plasma level measurements are not carried out to characterize accurately the area determinations or the terminal rate constant (as necessary in Eqs. 9 and 10), this simple method to determine Vd_{ss} is just as inaccurate as the classical methods under those conditions. A future publication will discuss the theoretical basis for the method as well as an error analysis. We have been utilizing this method in our recent pharmacokinetic work (14–16).

(1) D. S. Riggs, "The Mathematical Approach to Physiological Problems," Williams & Wilkins, Baltimore, Md., 1963, chaps. 7 and 8.

(2) S. Riegelman, J. C. K. Loo, and M. Rowland, *J. Pharm. Sci.*, **57**, 128 (1968).

(3) L. Z. Benet and R. A. Ronfeld, *ibid.*, **58**, 639 (1969).

(4) M. Gibaldi, *ibid.*, **58**, 327 (1969).

(5) J. G. Wagner, *J. Pharmacokinet. Biopharm.*, **4**, 443 (1976).

(6) C. A. M. van Ginneken, "Pharmacokinetics of Antipyretic and Anti-Inflammatory Analgesics," Stichting Studentenpers, Nijmegen, The Netherlands, 1976.

(7) G. N. Stewart, *Am. J. Physiol.*, **58**, 20 (1921).

[1] This equation is only appropriate for decreasing plasma concentrations with time. When calculating AUC for curves that increase to a maximum, the linear trapezoid rule should be used for concentrations up to the peak. Again, Eq. 11 is just an approximation since theoretically the logarithmic trapezoid rule should only give a correct area estimation during the terminal log-linear portion of the plasma–time curve.

(8) W. F. Hamilton, J. W. Moore, J. M. Kinsman, and R. G. Spurling, *ibid.*, **99**, 534 (1931).
(9) W. Perl and P. Samuel, *Circ. Res.*, **25**, 191 (1969).
(10) J. H. Oppenheimer, J. L. Schwartz, and M. I. Surks, *J. Clin. Endocrinol. Metab.*, **41**, 319 (1975).
(11) *Ibid.*, **41**, 1172 (1975).
(12) K. C. Yeh and K. C. Kwan, *J. Pharmacokinet. Biopharm.*, **6**, 79 (1978).
(13) W. L. Chiou, *ibid.*, **6**, 539 (1978).
(14) N. S. Aziz, J. G. Gambertoglio, E. T. Lin, H. Grausz, and L. Z. Benet, *ibid.*, **6**, 153 (1978).
(15) J. Ham, R. D. Miller, L. Z. Benet, R. S. Matteo, and L. L. Roderick, *Anesthesiology*, **49**, 324 (1978).
(16) R. Platzer, R. L. Galeazzi, G. Karlaganis, and J. Bircher, *Eur. J. Clin. Pharmacol.*, **14**, 293 (1978).

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BOOKS

REVIEWS

Methods in Pharmacology, Vol. 4B, Renal Pharmacology. Edited by MANUEL MARTINEZ-MALDONADO. Plenum, 227 W. 17th St., New York, NY 10011. 1978. 403 pp. 15 × 25 cm. Price $39.50.

This volume aptly fills a gap in the literature. Although the book was apparently intended for researchers in this area, each topic is treated such that even pharmacologists and physiologists not active in renal research can gain a better understanding of and appreciation for the technology discussed. In addition to the clear treatment of each topic, all chapters are well referenced. This book should be made available to all students of pharmacology, physiology, and medicine.

The first chapter deals with morphological methods for studying the kidney, including fixation methods, light microscopy, and EM procedures. The second and third chapters deal with renal dynamics, including clearance methods and intrarenal blood flow measurements. Chapter 4 deals with micropuncture techniques, while the fifth chapter is concerned with microanalytical methods for analyzing samples obtained by the methods discussed in the previous chapter.

In Chapter 6, methods measuring glomerular dynamics are described, and mathematical models of glomerular ultrafiltration are discussed.

Chapters 7 and 8 provide detailed descriptions of the apparatus used in microelectrode studies; Chapter 7 has to do with electrical potential measurement, and Chapter 8 concerns pH measurement. Chapter 9 is especially interesting, providing methods for dissection and perfusion of isolated tubules.

Chapters 10–12 deal with renal biochemistry: Na–K-ATPase, adenylate cyclase, and intermediary metabolism. The last three chapters describe the methodology used in studying isolated perfused kidney in the dog (*in vitro* and *in vivo*) and the rat (*in vitro*).

The effectiveness of the book would have been greatly enhanced by including with each chapter a brief description of several experiments using the methods described. Including examplary experimental drug application protocols and some typical data obtained by the methods would have provided the pharmacologist reader with a much clearer understanding of how the method might be applied.

In addition, it would probably be helpful for many readers if the names and addresses of manufacturers of critical equipment and supplies had been provided with each chapter.

I have personally experienced a need for such information in my previous experience with other volumes of *Methods in Pharmacology*.

These two shortcomings are the only major problems I see in this volume, and I view it generally as a very informative and useful contribution to the scientific literature.

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Cocaine: 1977 National Institute on Drug Abuse (Research Monograph Series 13). Edited by R. C. PETERSON and R. C. STILLMAN. Superintendent of Documents, U.S. Government Printing Office, Washington, DC 20402. Price $3 (paper cover).

The National Institute on Drug Abuse (NIDA) has been publishing monographs on specific substances of abuse. The monograph on cocaine follows a pattern similar to earlier monographs. There are 10 chapters by 13 authors with 350 references dating from 1884 to 1977. There is an adequate author and subject index.

The information in the book could be classified according to history, pharmacology and toxicology, abuse, use, behavioral aspects, and legal aspects.

This book and the others in the series are useful to anyone involved in the area of substance abuse. Their rather low price makes the entire series inviting.

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NOTICES

Disposition of Toxic Drugs and Chemicals in Man. Vol. 1. Centrally-Acting Drugs. By RANDALL C. BASELT. Biomedical Publications, P.O. Box 368, Canton, CT 06019. 1978. 306 pp. 15 × 23 cm. Price $22.50.

Handbook of Experimental Pharmacology, Vol. 51. Uric Acid. By WILLIAM N. KELLEY and IRWIN M. WEINER. Springer-Verlag New York Inc., 175 Fifth Ave., New York, NY 10010. 1978. 639 pp. 16 × 24 cm. Price $159.50

IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans. Some N-Nitroso Compounds. (Vol. 17). International Agency for Research on Cancer, distributed by World Health Organization. 1978. 165 pp. 17 × 24 cm. Price $25.00; 50 fr.

IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans. Vol. 18. Polychlorinated Biphenyls and Polybrominated Biphenyls. International Agency for Research on Cancer, distributed by World Health Organization. 1978. 140 pp. 18 × 24 cm. Price 20 fr.

The International Challenge of Drug Abuse: NIDA Research Monograph 19. By ROBERT C. PETERSEN. National Institute on Drug Abuse, Division of Research, 5600 Fishers Lane, Rockville, MD 20857. 1978. 349 pp. 14 × 23 cm.

Methods in Cancer Research, Vol. XVII. Cancer Drug Development, Part B. Edited by VINCENT T. DeVITA, Jr., and HARRIS BUSCH. Academic, 111 Fifth Ave., New York, NY 10003. 1979. 356 pp. 15 × 23 cm. Price $37.00.