# PHARMACOKINETICS[1]

By John G. Wagner
*Medical Research Division, The Upjohn Company, Kalamazoo, Michigan*



"It will be at once admitted that the medical practitioner ought to be acquainted with the strength of the various compounds which he applies as remedial agents, and that he ought, if possible, be able to regulate their potency"—John Snow, 1847.

"Upon closer examination, it is usually found that the black boxes of biology are actually various hues of grey"—Grove C. Nooney, 1966.

The word pharmacokinetics means the application of kinetics to *pharmakon*, the Greek word for drugs and poisons. Material reviewed here concerns the kinetics of absorption, distribution, metabolism, and excretion of drugs, poisons, and some endogenous substances. The purpose of pharmacokinetics is to study the time course of drug and metabolite concentrations and amounts in different tissues and excreta, and to construct suitable models to interpret such data. The relationship between pharmacokinetics and pharmacologic effect is not covered in this review.

The past ten years of feverish activity in the field of pharmacokinetics was preceded by a century of relative calmness. The writings of John Snow (1) are analogous to the taproot of pharmacokinetics. The contributions of many authors to the pharmacokinetics of the inhalation anesthetics are noteworthy and were reviewed by Butler (2). The birth of modern pharmacokinetics, both from practical and theoretical standpoints, occurred during the years 1937–1948 (3–7).

With some exceptions, the review of Nelson (8) and the book by Rescigno & Segre (9) are considered to cover the literature to 1961. Hence, this review is principally concerned with literature of the period 1961–1966.

## The Analysis of Pharmacokinetic Data

*Philosophy of modeling.*—Major contributors (10–18) in the application of mathematical models to physiological systems and pharmacokinetic data have developed a philosophy of modeling. Their articles should be read by all who elaborate models from blood level and urinary excretion data. These authors, who contributed much of the mathematics, were frequently more cautious than some of the later investigators who applied the mathematics to particular data.

*Parameter spaces and confidence intervals.*—Estimates of pharmacokinetic parameters obtained graphically should be considered preliminary estimates

[1] The survey of literature pertaining to this review was concluded in November, 1966. Some papers cited were published in 1967.

only, since they may usually be refined by application of a digital computer. Additional information about the derived parameters, such as the nature of surfaces or spaces and confidence intervals, may be obtained simultaneously along with the least squares estimates of the parameters. At the Gordon Research Conference on Biomathematics in 1966, Berman discussed the theory of model building, placing emphasis on the tests required to lead one to the "best model." The closeness of fit, as measured by residuals, is a strong point; but he cautioned that one must look for trends in areas of poor fit to test the adequacy of the model. The digital computer is a most useful tool in investigation of the error map. If the calculated parameter has converged into a "steep-sided error hole," Berman judges the parameter useful. However, if the parameter resides in "a plane or in a shallow hole," the parameter should be eliminated. Details of least squares nonlinear estimation and exploration of the parameter spaces are given by Booth & Peterson (19). Papers concerned with nonlinear regression, confidence regions, applications of digital computers and related subject matter have been written by several authors (20–53). Many companies and institutions are developing digital computer software for both simulation and data-fitting models. With these and high speed digital computers at their disposal, the ability to interpret pharmacokinetic data will expand greatly in the next few years.

*Deterministic and stochastic models.*—Most pharmacokinetic models are deterministic in nature. They assume that results are decided by antecedent causes which may be inherited or environmental. There is no particular emphasis on discrete events, and transformations are followed only in bulk. Such models give the expectation of a smooth concentration or amount-time course. Deviations and fluctuations are usually attributed to experimental error. Parameters, such as rate constants and volumes, are assumed to remain constant over the entire experimental period.

Bartholomay (54) and other investigators have proposed stochastic models for chemical reactions and for enzymatic catalysis, in particular. These provide a mathematical model sufficiently complete to account for random fluctuations and the differentiation between so-called "reproducible" (small fluctuations) and "irreproducible" (large scale) fluctuations. Ideally, the smooth concentration-time course of the deterministic model may be thought of as the central tendency of a stochastic process, i.e. the stochastic model is "consistent in the mean" with the deterministic model.

*Estimation of absorption rate.*—Hellman et al. (55) showed that dietary cholesterol is largely absorbed through the lymphatics of man and that the major portion is esterified during the absorptive process. Schedl & Clifton studied small intestinal absorption of various steroids in rats (56) by tying inlet and exit cannulas into the duodenum and terminal ileum, respectively, then infusing the steroid solution with an infusion pump. In man they (57, 58) have studied absorption of L-methionine and various steroids with the method of steady-state perfusion through a transintestinal tube using so-called nonabsorbable indicators to follow volume changes. Berkowitz et al.

(59) studied rates of stomach emptying and fat absorption following administration of fatty meals containing triolein-$^{131}$I by externally counting six abdominal areas. The method may also be used to evaluate the effectiveness of antimotility medications on gastric emptying and intestinal transit times. A similar method was used by Lutwak & Shapiro (60) to study calcium absorption in man. Sund & Schou (61) measured absorption rates following subcutaneous and intramuscular administration of various labeled compounds by excising tissues at various times, post administration.

Absorption rates may also be estimated from blood concentration and urinary excretion data. The method of Wagner & Nelson (62) is applicable to both blood (serum, plasma) concentration data and, in modified form, to measurement of urinary excretion of unchanged drug. This method leads to a plot of amount, or per cent, of drug absorbed per milliliter of the apparent volume of distribution against time. Such plots may then be resolved to determine the kinetics of absorption (63–65). Nelson (66) extended the method to allow estimation from measurement of metabolite concentration in blood; this method, however, is restricted to cases where little or no unchanged drug is eliminated via the urine. A more generalized form of the equation was given by Riegelman & Ballard (67). Scholer (68) also reported a method for determination of absorption rates by integration of rates of appearance and disappearance. Levy & Miller (69) showed that the time of onset of a suitable pharmacologic response under conditions where a constant drug concentration gradient is maintained across the absorbing membranes reflects absorption rate. Garrett et al. (70) estimated the rate constant for absorption by using an analogue computer to fit a line through serum concentrations observed following oral administration, after the other rate constants of the system had been estimated from studies involving intravenous administration in the normal and nephrectomized dog. The integral equation approach was elaborated by Stephenson (71) and was applied to estimation of absorption rates of $^{28}$Mg by Silverman & Burgen (72) and of $^{47}$Ca by Birge et al. (73). This method relates the plasma concentration curves following both intravenous and oral administration of the same substance to the same subject at different times. Some of the pitfalls of this method are as follows. If the two routes are used at different times, the intrasubject variation in the rate parameters, which can be quite large (65, 74, 75), is ignored; the large errors may not be obvious, but they exist. There is no reason to expect *a priori* that the drug is handled the same after intravenous and oral administration. Finally, the rate and interval of the intravenous injection probably greatly affects the shape of the plasma concentration curve obtained by this route, and the assumption that this curve is the resultant of a "spike" dose is most likely invalid in many practical situations. The rate constant for absorption may sometimes be estimated by plotting residuals on Cartesian or semilogarithmic paper as illustrated by Wagner (35, 74). However, if the model contains reversible transfers between compartments, the rate parameters taken from the curves are just intermediate computing parameters

and cannot be directly associated with one or more rate constants of the model as was practiced by Hecht et al. (76); for details of another practical example, see Wagner et al. (77); for the equations, see Frost & Pearson (78).

*Estimation of efficiency of absorption.*—An equation for calculating per cent absorption from data collected in perfusion or intubation studies in which a nonabsorbable indicator is used was given by Borgström et al. (79). A study of experimental errors in estimation of $^{59}Fe$ absorption by means of whole body counting and fecal recovery techniques was reported by Lindell et al. (80). Simultaneous administration of an oral dose of $^{60}Co$-labeled vitamin $B_{12}$ and an intravenous dose of $^{57}Co$-labeled vitamins $B_{12}$, followed by external counting above the liver, was shown by Weisberg & Glass (81) to give absorption results which correlated well with a previous, more time-consuming method. Similarly, absorption of thyroxine was estimated by Hays (82) by administering an oral dose of $^{125}I$-labeled thyroxine, an intravenous dose of $^{131}I$-labeled thyroxine, and calculating the ratio of $^{125}I/^{131}I$ in subsequent serum samples. Smith (83) showed that net magnesium absorption, expressed as a percentage of intake, correlated linearly with transit time to the distal ileum. Nelson et al. (84) showed that the amount of the metabolite of tolbutamide excreted in the urine, and its excretion rate, correlated with the surface area of tolbutamide in the dosage form administered. The results of these two experiments are not unexpected since, as the latter indicated, available surface area and solubility of a poorly soluble drug controls rate of dissolution, and ultimately, absorption rate. In addition, the amount absorbed is a sum of the products of the absorption rates and the times over which the rates were operative. Transit time in the gastrointestinal tract may become extremely important in those cases where absorption of the drug is limited to a specific segment of the tract (85).

The equation where $F$ is the fraction of the dose $D$ which is absorbed, $V$

$$FD = VK\int_0^\infty C(t)dt \qquad 1.$$

is the apparent volume of distribution, $K$ is the first order rate constant for overall loss of drug from the body, and the integral is the area under the plasma or serum concentration curve of unchanged drug from time of administration to infinite time, was given in modified form by Wagner et al. (86), and in the above form by Wagner (85). Equation 1 is implicit in the work of Teorell (5) and in the clearance studies of Schück & Šmahel (87). The equation indicates that the area under the plasma concentration curve will be directly proportional to the fraction absorbed, if the dose and the plasma clearance, $VK$, are constant; and the area will be directly proportional to the dose administered, if the fraction absorbed and the plasma clearance are constant. Unfortunately, Gladtke (88) erroneously translated the amount absorbed, $FD$, as the rate of absorption. Comparison of the areas under the calcium serum concentration curve was used by Neipmann (89) as an index of relative absorption of calcium. Wagner & Nelson (64) presented seven

methods of estimating relative absorption from blood concentration and urinary excretion data. An additional method, reported by Wagner (90), proved useful for estimating relative absorption of a drug in a series of clinical studies in which blood levels are measured after single or multiple doses or both. Evidence that these equations are valid has been reported by Dost & Gladtke (91), Middleton et al. (92), Gladtke (88), Dost (93), Diller (94), and Wagner et al. (74, 77, 90).

*Estimation of volume of distribution.*—Dominguez (95) defined volume of distribution as the volume of body fluids which holds the substance in solution at the same concentration as the plasma. Swintosky et al. (96) estimated the volume of distribution of sulfaethidole by dividing the calculated amount of drug in the body three hours post administration by the blood concentration measured at the same time. Czaczkes & Kleeman (97) estimated the volume of distribution of antidiuretic hormone by dividing the infusion rate (mass/time) by the product of the equilibrium state plasma concentration and the first order rate constant for elimination; the latter was estimated from the down slope after the infusion had ceased. A modification of this method, which does not require continuing the infusion until the equilibrium state has been reached, was reported by Wagner & Alway (98). Volume of distribution may also be estimated by dividing the renal clearance of the compound by the rate constant for renal excretion of the compound (99). Renal clearance is equivalent to the slope of the straight line when the excretion rate of the compound is plotted against the total plasma or serum concentration of drug at the midpoints of the excretion intervals. Such a linear relationship has been reported for penicillins G and V (100), tetracycline (101), salicylate (102), chloramphenicol (74), and digitoxin (103, 104). Chulski et al. (101) showed that this method provided a more plausible volume of distribution of tetracycline than had been reported by other authors, who used the extrapolation method discussed below. Smith et al. (105) estimated the volume of distribution of 5-methylpyrazole-3-carboxylic acid by dividing the amount of compound excreted in the urine in infinite time by the product of the area under the plasma concentration curve from zero to infinite time and the rate constant for elimination which was estimated from the terminal plasma concentation data. In this case, the entire dose was excreted in the urine.

Krüger-Thiemer (106) criticized the equations and models of Wagner (85, 107). The basis of the criticism is the lack of cognizance of protein binding of the drug in estimating the volume of distribution. As an alternative, Krüger-Thiemer (106) suggests that for the description of the distribution of all drugs of low molecular weight, the body should be divided into only three invariable, real volumes: plasma water, interstitial water, and intracellular water. It would be desirable if the real world were as Krüger-Thiemer hypothesizes. However, such is not the case. Few drugs and tracer substances are ever distributed into exactly the same fluids and tissues of the body; hence, there are no real volumes which are applicable to all drugs. Thus, the prac-

tice of assigning numerical values to two or three volumes of distribution, on the basis of body weight or other such criterion, as Krüger-Thiemer does (106), is incongruous and not in keeping with known facts relating to drug distribution. The literature on this subject is vast and space allows only limited documentation (77, 108–113).

It is common practice to plot the plasma or serum concentration of a compound, observed following rapid intravenous injection, on semilogarithmic graph paper, draw a straight line through the terminal linear segment, extrapolate the line back to time zero, and divide the intercept obtained into the dose administered to estimate the apparent volume of distribution, pool size, or space. The practice is almost universal (113–117), but complete documentation is impossible here. If the model which applies to the particular system involves a single compartment, and the initial non-linearity of the semilogarithmic plot is assumed to be attributable to mixing, then the estimate of the volume of distribution made by this extrapolation method would be very close to that expected on the basis of the appropriate mathematical expression. However, Wagner & Northam (117) showed that if the two-compartment open model applies to the data, then the volume estimated by the extrapolation method is always an overestimate of the true volume, and the error depends upon the relative values of the ratios of the rate constants and the two volumes of the system. If the extrapolation method is applied to data obtained after administration of a compound by any route other than intravenously, the number obtained is not a volume of distribution at all, but is a complex function of many variables (117). The above discussion should explain why the extrapolation method and the constant rate infusion method failed to yield the same estimate of the distribution volume of antidiuretic hormone (87). Factors involved in blood volume measurement were discussed by Albert et al. (118).

Volume of distribution is model-dependent as Dominguez (95) indicated. It is feasible to interpret the same set of data in terms of two or more compartment models and to obtain different estimates of the volume of distribution. It is appropriate to define the volume of distribution, $V_i$, of a given compartment of a model as the quotient of the total amount of drug in the compartment, divided by the concentration of the total drug in the compartment, where both the amount and concentration are estimated and measured, respectively, at the same time. For a system of $n$ compartments, the total volume of distribution, $V$, is then

$$\sum_{i=1}^{n} V_i \quad \text{where} \quad i = 1, 2, \cdots, n \qquad 2.$$

*Estimation of other rate constants and clearances.*—Samachson (119) introduced the useful concept of gastrointestinal clearance in reporting results of a study in which $^{85}Sr$ and $^{45}Ca$ were given intravenously to human patients. He defined gastrointestinal clearance as the ratio of the amount of isotope secreted into the gastrointestinal tract and not reabsorbed in a given time to

the average plasma concentration of the isotope in the same time interval. He showed that the numerator is the true fecal excretion in the given time and is equivalent to the urinary excretion in the same time multiplied by the ratio of the gastrointestinal clearance to the renal clearance. Also, the gastrointestinal clearance is equal to the product of the renal clearance and the ratio of the total amount ultimately excreted in the feces to the total amount ultimately excreted in the urine. The method circumvents the problem of the lengthy and variable delay in fecal excretion.

By plotting ratios of certain parameters, Cavalieri & Searle (120) were able to estimate the rate constant for loss from compartment 2, namely $k_2 + k_3$, for the system

$$1 \underset{k_2}{\overset{k_1}{\rightleftarrows}} 2 \xrightarrow{k_3} \qquad 3.$$

They applied the method to $^{131}$I-labeled thyroxine, where compartment 1 represents the plasma and compartment 2 represents the extravascular space of the liver. It appears that the method may be applicable to data derived from transintestinal intubation studies in man and perfusion studies in animals; in these cases, the compartments would be redefined in such a way that compartment 1 would be the gastrointestinal contents and compartment 2 would be the blood and other fluids of distribution. Hence, $k_1$ would be the rate constant for efflux from the intestine to blood; $k_2$ the rate constant for influx from blood to gastrointestinal contents; and $k_3$ would represent the rate constant for elimination except for that portion of drug going back into the gastrointestinal tract.

Bloom & Nelp (121) reported a discrepancy between the plasma half life of tritiated digoxin and the so-called biological half life. The discrepancy is probably an artifact since considerable digoxin-$^3$H is excreted in feces; and because of the time lag in fecal excretion, the authors were not plotting the correct quantities to obtain the so-called biological half life.

Wagner (122) described a method for estimating the rate constants for absorption, metabolism, and elimination from the time course of urinary excretion of a metabolite when there is an independent estimate of the rate constant for renal excretion of the metabolite. The method applies to a model in which there is a catenary chain with parallel paths branching from the compartment representing drug in blood and other fluids of distribution.

*Compartment models.*—Most pharmacokinetic models have been elaborated by joining together, in appropriate fashion, a series of unit processes. A unit process may represent the movement of a species from one location through a membrane to another location, or it may represent a biotransformation, a metabolic process, or a hydrolytic reaction. Each unit process may be considered to be composed of two compartments which have one or two rate parameters associated with them.

Practically all pharmacokinetic models are composed of five basic types of unit processes. These are: (*a*) the conversion of a chemical species to another chemical species by an enzymatic reaction or a hydrolytic reaction

catalyzed by a chemical constituent of the body; (*b*) transfer of a species across a barrier or membrane where the volume of the compartment on one side of the barrier may be ignored; (*c*) transfer of a species across a barrier or membrane where the volumes of the compartments on both sides of the membrane must be considered; (*d*) transfer of a species from inside the body to the urine, feces, sweat, or expired air, all of which are considered to be outside the body; and (*e*) a system composed of free, or nonprotein-bound, species and the corresponding protein-bound species.

The approximation arrived at by considering that the Michaelis constant is much greater than the substrate concentration, when the enzyme responsible for a biotransformation is far below saturation by the substrate, is considered valid in most enzymatic biotransformations of drugs and their metabolites (9); hence, processes of type (*a*) above may usually be regarded as obeying first order kinetics. It is possible, however, for the kinetics to be zero order or second order. Teorell (5) discussed the absorption of a drug from the fluids of the gastrointestinal tract, through the gastrointestinal barrier into the blood, as an example of a type (*b*) unit process. In this case, the volume of the fluid in the gastrointestinal tract may be ignored, and the more complicated differential equation for the process may be replaced by a simple first order rate equation in which the variable is the amount of drug. Wagner & Nelson (64) showed that when such an equation is integrated, one of the terms should be the amount absorbed, $FD$, where $F$ is the fraction of the dose, $D$, which is absorbed, and not the dose. The two-compartment open system, discussed by several authors (117, 123, 124), incorporates an example of unit process (*c*) above. The most common example of a type (*d*) unit process is urinary excretion of a drug or metabolite. Renal excretion may usually be represented by a first order rate constant, but there are reasons why this may not be valid at all plasma concentrations. The papers of Krüger-Thiemer et al. (106, 125, 126) provide adequate examples of unit processes of type (*e*).

Pharmacokinetics has inherited a vast knowledge of schematic and mathematical models which have been applied to the interpretation of data arising from experiments using tracer or isotopic compounds (9, 11–13, 16–18, 31, 33, 112, 127–147). This knowledge is applicable not only to data arising from the administration of isotopically-labeled drugs, but also to blood concentrations and amounts of drug excreted in the urine which are measured by chemical and microbiological assay methods. Literature (25, 26, 29, 37, 78, 117, 123, 124, 148–163) on compartment models from the fields of biophysics, chemistry, medicine, pharmacology, and physiology is applicable also to pharmacokinetic modeling. Compartment models for drugs and their metabolites have been elaborated by Teorell (5), Nelson (8, 164–169), Rescigno & Segre (9), Campbell, et al. (170), Garrett and co-workers (70, 171–173), Stelmach et al. (174), Riggs (175), Wiegand & Taylor (176, 177), Nodine et al. (33), Levy (178, 179). Von Wittenan & Yeary (180) Bray et al. (181), Elliott (182), Portmann, Moore and co-workers (183, 184). Martin

(185), Krüger-Thiemer (34, 106, 126, 186), Hecht et al. (76), Smith et al. (105), Wagner and co-workers (35, 63, 64, 74, 85, 98, 107, 117, 187) and others. Space limitation does not allow classification or discussion of these.

If the model which applies to a particular set of data contains a pair of model rate constants indicating a reversible transfer between two compartments, or if there is a cyclic structure formed from three or more compartments, or there is a catenary chain with parallel paths, then some or all of the observable rate parameters which may be estimated from the data are not equivalent to the model rate constants, but rather are complicated functions of them.

*Blood levels and urinary excretion following multiple doses of drug.*—Usually therapy with a drug involves the administration of multiple doses and not just a single dose. Koppanyi & Avery (188) pointed out that all drugs are cumulative provided the rate of administration exceeds the total rate of elimination. Some indices of drug accumulation were given by Wagner (189). One index is the ratio of the average amount of drug circulating in the body, and lost from the body, during a dosage interval at the equilibrium state, to the amount of drug absorbed, and lost from the body in infinite time, after a single dose of the drug. The index is based on the assumption that if $D$ is the single dose of drug, then $D$ is given at the beginning of each uniform dosage interval. If the dose becomes a variable, then the equation (189) to estimate the extent of drug accumulation is revised. Krüger-Thiemer's (125, 190) concept of drug accumulation implies that drug accumulation is associated not with the final state, but only with the transition between the initial and final states.

Earlier literature (7, 149, 192–194) contains some mathematical descriptions of drug accumulation curves. More recent literature (74, 98, 125, 191, 195–202) provides additional equations useful for prediction of blood concentrations and urinary excretion as a function of time following multiple doses of drug. Accurate prediction in such cases would require constancy of all pharmacokinetic parameters, such as efficiency and rate of absorption, volume of distribution, and rate constant for elimination. In light of the known inter- and intra-subject variation of such parameters (35, 65, 75, 77, 178, 182, 203), one would not expect an accurate prediction on an individual subject basis, but a reasonably accurate prediction for the average of a panel of subjects is possible (74, 98, 202).

Wagner et al. (191) showed that the area under the blood concentration curve during a dosage interval at the steady state will be equal to the total area under the blood concentration curve following a single dose, providing certain conditions are satisfied. These workers supplied a simple equation to estimate the average blood concentration at the steady state; since the rate constant for absorption is not included in this equation, the latter has more utility than equations which attempt to predict the entire blood concentration-time course at the steady state. Modifications of this equation (191) and Equation 1 of this review, given by Wagner (35, 74, 90), allow estimation

of: (*a*) relative contributions of half life, and the ratio of the fraction of the dose absorbed to the volume of distribution in causing variability of blood levels; (*b*) relative absorption of a drug in a series of clinical studies; and (*c*) a method to show relative constancy of the fraction of the dose absorbed, the volume of distribution, and the rate constant for elimination which is based on plotting the average blood concentration at the equilibrium state against the dose of drug in mg/kg; for an example of the latter, see Wagner et al. (77).

Although blood levels and urinary excretion are usually measured following single doses of drug, a number of investigators (104, 204–215) made such measurements after multiple doses of drug. Wagner et al. (77) pointed out the advantages of defining blood levels during a dosage interval at the equilibrium state, rather than attempting to measure the peaks and nadirs after various doses. The reviewer agrees with Sugarman & Rosen (210) that in order to run a valid comparison of dosage forms that contain the same drug, but which release the drug at different rates, it is necessary to establish a steady state between input and output before measuring blood levels and urinary excretion as a basis of comparison. The urinary excretion study of Sugarman & Rosen (210), and the serum concentration study of Wagner et al. (77) are examples. The variability of equilibrium state serum concentrations of salicylate following administration of aspirin in two different dosage forms (215), and the variability of equilibrium state serum concentrations of indoxole following administration of indoxole in four different dosage forms (77) are worthy of note. The equation of Wagner et al. (191) also indicates that the average serum or plasma concentration at the steady state will be the same following equal doses of drug in a readily available form and in a sustained or prolonged action form, providing the efficiency of absorption is the same from the two dosage forms. The data of Sugarman & Rosen (210) and Green (215) support this mathematical prediction. These considerations suggest that the concept of the blood levels of drug, following administration of a sustained action dosage form, skirting between the level needed for biological activity and some level below the side effect level is not scientifically sound. This concept, illustrated with blood levels expected after single doses (216), falls down when one recognizes the individual subject variability of blood levels at the equilibrium state, and the expectation of the same average blood level independent of dosage form under the conditions stated above.

*Real and potential errors in the analysis of data.*—Gibaldi & Kanig (217) discussed errors in the use of polynomial approximation of urinary excretion rates. Wagner (218) discussed errors in plotting pharmacokinetic data and causes of curvature in semilogarithmic plots. Absorption rates of tolbutamide as reported by Nelson et al. (220) are in error since (*a*) the assay used was non-specific as shown by Thomas & Ikeda (219), (*b*) polynomial approximations were used, and (*c*) the wrong equation was used (66). The practice (97, 221–223) of drawing two or three linear segments through data points on semilogarithmic plots, without estimating residuals, does not conform to

theory. Preliminary estimation of the absorption rate constant by the method of Moore et al. (224) is really only valid when the ratio of the absorption rate constant to the loss rate constant is equal to two, in light of the equation of Teorrell (5). Differences in the apparent volumes of distribution of salicylic acid and sodium salicylate reported by Borzelleca & Lowenthal (225) depended only on inappropriate use of a digital computer program. Curvature in the semilogarithmic plots of Cameron et al. (226) following injection of bilirubin-$^{14}C$ in monkeys suggests a more complicated model than the authors employed. The analysis made by Fincher et al. (227) appears inappropriate in light of the large body of theory which should have guided the analysis. The apparent enhancement of absorption of tetracycline by chymotrypsin, reported by Seneca & Peer (228), depended mainly on the authors' method of averaging individual percentage changes, which gives great weight to extreme values. There were no significant differences in average serum concentrations of the antibiotic when it was administered alone or with chymotrypsin. The data of MacDonald et al. (229) support this viewpoint. The apparent volume of distribution of salicyluric acid calculated by Levy (178) may be in error since he misinterpreted the data of Schachter & Manis (230).

Several reports (178, 182, 231–234) claim that man has a limited capacity for the synthesis of salicylurate from salicylate and glycine, and that the kinetics of elimination of salicylate change to parallel zero order and first order paths above some critical salicylate concentration in the body after doses of aspirin of about 1.5 g or more. Evidence presented by Nelson et al. (232) for rats at doses above 20 mg/kg is very convincing. However, the reviewer believes that the kinetics of elimination of salicylate, following oral doses of 1.5 to 4 g of aspirin to adult man, are first order at all times post administration for the following reasons. First, the data of Levy (178) indicate that following ingestion of 1.5 and 2 g of aspirin in two subjects, the percentage of salicylurate excreted in each urine collection from two to 39 hours remained essentially constant and independent of time for each subject. Secondly, Wagner (235) has shown that approximately 50 per cent of a metabolite excreted in the urine may be excreted at such rates that the cumulative urinary excretion plot would appear to be nearly linear when the model is a catenary chain with parallel paths involving only first order rate constants. In addition, when excretion rate rather than formation rate of a metabolite is used to prepare a 'Lineweaver-Burk' plot (236), based on the kinetics of Michaelis & Menton (237), as practiced by Levy (231–233), the resulting plots are artifacts and do not have the significance attributed to them.

## Factors Altering The Kinetics and Magnitude of Pharmacokinetic Parameters

*The variability of pharmacokinetic parameters.*—The variation of serum or plasma concentrations at a given time or urinary excretion during a given

interval of time is such that, assuming a normal distribution, the coefficient of variation (per cent standard deviation) usually ranges from 25 to 75 per cent. The variation of estimated pharmacokinetic parameters, such as lag time, half life, area under the serum or plasma concentration-time curve, rate constant for absorption, rate constant for elimination, apparent volume of distribution, and renal clearance, is usually in the same range. The data of Juncher & Raaschov (238) yielded coefficients of variation of the areas of 46.5, 40.9, 43.1, and 49.0 per cent for potassium penicillin V, calcium penicillin V, penicillin V acid, and sodium penicillin G, respectively. Both inter-subject variation and intra-subject variation are important. Levy & Hollister (239) reported that in 12 subjects the rate constants for salicylate elimination obtained in two tests differed by less than 20 per cent for any one individual, while somewhat greater differences were found in five other subjects. Levy & Hollister (65) showed that coefficients of variation calculated from serum salicylate concentrations and per cent absorbed values were generally the same, except for the greater homogeneity of the absorption values when absorption was near completion. They also reported that the half life of meprobamate in 12 healthy human volunteers varied from 6.2 to 16.6 hrs, with a mean of 11.3 hrs, following administration of a single dose of 800 mg of meprobamate (240). Wagner (74) showed that the intra-subject variation of lincomycin serum concentrations as well as the areas under the lincomycin serum concentration curves was somewhat less than the inter-subject variation. Credit for recognition of such variability should not go to recent authors. Marshall et al. (4), in 1938, pointed out that absorption from the gastrointestinal tract may be a very important factor in toxicity, and that with large doses of both sulfanilamide and acetylsulfanilamide, the variation in absorption may explain the variability in the toxic reaction of animals to large doses. These authors (4) also were probably the first to point out that the efficiency of absorption of poorly water-soluble compounds, such as sulfanilamide and acetylsulfanilamide, decreases as the dose is increased–a fact that is still not well recognized in toxicology. Brodie (203) pointed out that a drug may be inactivated at rates which vary by 500 per cent or more among different individuals.

Ordinarily, panels of 5 to 20 subjects are used in pharmacokinetic studies. The assumption is usually made that the measurements or estimated parameters are normally distributed when the standard deviation or coefficient of variation is estimated. Wagner (35) presented data on larger numbers of subjects which indicated this assumption may not always be valid. Half lives of novobiocin estimated from serum concentrations observed in 58 adult volunteers were found to be log-normally distributed–i.e. the logarithms of the half lives were normally distributed. Half lives of lincomycin were also estimated from serum concentrations observed in a different group of 58 adult volunteers. Although one could not reject the hypothesis that the lincomycin half lives were normally distributed, the value of Chi-Square was lower when the logarithms of the half lives were used than when the half

lives were used. The possibility of bimodal and trimodal distributions also exist.

*Drug interactions.*—Brodie (241) summarized mechanisms of drug interaction when drugs are either given in combination or are present in the body at the same time. Mechanisms which lead to a reduction in concentration of drug at receptor sites include: (*a*) one drug may interfere with the absorption of another drug; (*b*) one drug may alter stomach-emptying rate and, therefore, alter absorption rate of another drug; (*c*) alteration of plasma pH, such as by inhalation of carbon dioxide, will alter transfer rates of an acidic or basic drug across internal membranes and, hence, change tissue concentrations; (*d*) a urinary acidifier or alkalinizer, such as ammonium chloride and sodium bicarbonate, respectively, will change the excretion rate of acidic and basic drugs; (*e*) one drug may block transport, or compete for the transport mechanism with another drug, in the active tubular secretory pathway in the kidney tubules; (*f*) a diuretic may alter the urinary excretion rate of another drug by altering urine flow rate; (*g*) one drug may displace another drug from its protein-bound complex and, hence, alter plasma and tissue concentrations; a drug may also displace an endogenous compound which is protein-bound; (*h*) chronic administration of a drug may stimulate its own rate of metabolic transformation by enzymes in liver microsomes; and one drug may lead to enhanced metabolism of a wide variety of other drugs; evidence suggests that in these cases, the drug increases enzyme activity by inducing the synthesis of more enzyme protein; hence, the use of the phrase "enzyme induction."

Examples of this group of mechanisms are as follows. Bleifeld & Gehrmann (242) reported that *p*-aminosalicylic acid interfered with absorption of vitamin $B_{12}$. Berkowitz et al. (59) showed that antimotility medications delayed and slowed fat absorption. Portnoff et al. (243) and Beckett & Rowland (244) illustrated the dependency of urinary excretion rate on urine pH. Probenecid apparently blocks or inhibits active tubular secretion of penicillins and some other drugs (245); the pharmacokinetics of this effect was discussed by Levy (246). Kostenbauder (247) has found that the half life of one drug may be increased by a factor of two to three times when another drug is present in the body in sufficient amounts at the same time. This effect appears to be attributable to competition for excretion by the active tubular transport system, and acids were found to interfere with bases, and vice versa. Increasing urine flow rate may increase renal clearance (245), but usually the effect is not marked. Sulfonamides can alter serum protein binding of penicillins in man, but the effect has little practical significance (248). Reviews have been written on the subject of enzyme stimulation and inhibition in the metabolism of drugs (249–251). The future will reveal how important this phenomenon is in therapy with drugs in man. It is too early to make an assessment at this time.

*Pharmacogenetics.*—Several examples of genetically determined, individual differences of drug metabolism in man are well established. The differ-

ences have one of three causes: (*a*) alterations of the properties of an esterase; (*b*) a failure involving acetylation; and (*c*) several conditions affecting the conjugation of drugs with glucuronic acid. All of the examples seem to involve alterations of enzyme activity in the liver. The first fully documented observation of hereditary control of drug metabolism was that of atropinesterase in rabbits. The hereditary alterations affecting esterase activity in man were first noted in studies of individual differences in the response to succinylcholine, which is widely used as a muscle relaxant during anesthesia and for anticonvulsive therapy (252). Evans & White (253) reported that sulfamethazine acetylation is polymorphic in human populations, and that the isoniazid inactivator polymorphism is due to the same acetylation phenomenon. Studies performed with human liver homogenates revealed that the polymorphism lies in the activity of the acetyltransferase enzyme, which transfers acetyl groups from acetylcoenzyme A to the acceptor drug molecule. Hydralazine is also an acceptor molecule which is subject to the same polymorphic acetylation. Andersen (254) reported that with respect to isonicotinic acid hydrazide in man, the rate constant for renal excretion of acetylated compound, and the rate constant for other forms of elimination were normally distributed; whereas the rate constant for acetylation was bimodally distributed.

Nelson (255) pointed out that in pharmacogenetic studies, decisions as to the existence of polymorphism is often based on the distribution of blood concentrations of the drug measured at some arbitrary time following administration to a large number of subjects, but that this procedure may yield results that are artifacts. Simulations performed by the reviewer with a digital computer and simple model tend to confirm this prediction of Nelson. Using $\hat{B} = (A^\circ)(k_1/k_1\text{-}k_2)\ (e^{-k_2 t}\text{-}e^{-k_1 t})$, for a given $A^\circ$, when $k_1$ and $k_2$ were normally distributed, values of $\hat{B}$ at $t=4$ were normally distributed, but values of $\hat{B}$ at $t=9$ were not normally distributed. Other simulations produced similar results. Nelson suggested that half life was the appropriate parameter to use for histograms.

Porter (256) reviewed the evidence that certain drug reactions are caused by variations in metabolic patterns that are genetically determined. The books of Meier (257) and Kalow (258) constitute extensive reviews of pharmacogenetics.

*The effect of urinary pH and flow rate on urinary excretion and metabolism of acidic and basic drugs.*—When the urine pH is high, basic drugs are excreted slowly and more extensive metabolism occurs. When the urine pH is low, basic drugs are excreted more rapidly and metabolism is less extensive. With acidic drugs, the relationships are reversed (243, 244, 259–264). The tubular epithelium of the distal convoluted tubules is selectively permeable or more permeable to the unionized lipid soluble molecule than to the poorly lipid soluble anion or cation (259). The $pK_a$ of the compound and the pH and volume of the tubular fluid determine the concentration of un-ionized molecules in the tubular fluid; the rate of reabsorption from tubular fluid back

into the circulation depends on this concentration and on the partition characteristics of the molecules. There is probably a limiting ratio of concentration of molecules in the tubular and peritubular fluids below which reabsorption is neglible, since water loading can eliminate volume-dependent fluctuations in urinary excretion rate of drug (264). The implications of the above factors in experimental design were discussed by Beckett (264).

There is an effect of posture on urinary pH, electrolyte excretion, and urinary flow rate (265, 266), and there is a diurnal variation of both urinary pH and flow rate in man (264). Dreaming sleep in man causes changes in urine volume and osmolality (267). Careful controls and sequential studies in the same subjects would be necessary to distinguish these effects from the effect of alteration of pH at the absorption site, such as in the studies of Gibaldi & Kanig (268, 269) with salicylate and creatinine.

The biological response to amphetamine was shown to be a function of urinary pH by Smart & Turner (270); hence, pharmacokinetic considerations have been correlated with biological response. Also, the intensity and duration of the primary pharmacologic action and of side effects of a drug may depend on the time of day the dose is given. In the case of intoxication following overdosage, acid diuresis in the case of a basic drug, and alkaline diuresis in the case of an acidic drug, are indicated (264).

Several authors (126, 271–273) have published equations which give the renal clearance of a drug as a function of the many variables involved. Wagner (273) reviewed these and suggested equations for excretion rate, corrected renal clearance, and uncorrected renal clearance of exogenous compounds which are not actively reabsorbed. Butler (99) pointed out that a drug may be largely eliminated in a few minutes, or it may persist in the body for years. In the termination of action of a drug, the relative importance of metabolic inactivation and elimination of unchanged drug will be determined by the relative rates of the processes.

*Protein binding.*—The significance of protein binding of drugs, particularly of sulfonamides and antibiotics, and its relationship to therapeutic activity and pharmacokinetics, is poorly understood. The excellent reviews of Goldstein (274) and Thorp (275) delineate the problem. The fact that plots of the logarithm of the total plasma or serum concentration of a drug against time are linear in the post-absorptive region for drugs that are protein-bound suggests: (*a*) that the plasma contains a constant proportion of the total drug in the body in the region where linearity exists, and (*b*) that the instantaneous rate of drug loss is directly proportional to the concentration of total drug and not to the concentration of free drug. The latter suggests that the protein-bound drug acts as if it were not protein bound with respect to metabolism by body enzymes and excretion by the active secretory pathway in the kidney. For the protein-drug interaction,

$$\text{Drug} + \text{Protein} \underset{k_2}{\overset{k_1}{\rightleftarrows}} \text{Drug-Protein Complex}; \quad \text{where} \quad K = k_1/k_2$$

Martin (185, 276) indicated that the binding of a model drug to plasma proteins only has an appreciable effect on pharmacokinetics if $K > 1 \times 10^4$. Two considerations largely invalidate the theorizing of Martin. First, for most drug protein interactions, reported $K$ values are in the range $10^2$ to $10^5$, and the majority of values are below $1 \times 10^4$. Hence, Martin's model curves (276) for hypothetical drugs with $K$ values of $10^6$ and $10^7$ do not apply to most real drugs. Secondly, Thorp (275) pointed out that the constants, $K$ (the association constant), $N$ (the number of binding sites per protein molecule), and $B$ (the fraction of bound drug) are determined at equilibrium, and this tends to obscure the kinetic nature of the drug-protein interaction. The rate constants, $k_1$ and $k_2$, have seldom been determined. Where they have been determined, the debinding constant $k_2$ was of the order of $10^6$ $hr^{-1}$ corresponding to a half life of $7 \times 10^{-6}$ hrs. Hence, the rate of dissociation of a drug-protein complex will have little or no effect on the overall kinetics in the body. Burns et al. (277) reported linear plots over a range of plasma concentrations of 10 to 80 $\mu$g/ml when the logarithm of the total plasma concentration of phenylbutazone was plotted against time; but Martin's theory (276) would not predict this, since phenylbutazone has a $K$ of $1.25 \times 10^5$ (275). Such plots may have curvature which is obscured by experimental error. Simulation studies with computers may throw light on this problem.

It is generally accepted that the antibacterial activity of serum, containing a sulfonamide or antibiotic, depends on the concentration of free drug (125, 248, 274, 275, 278–280), but there is some disagreement (281–283). One of the best correlations published related activity to total plasma or serum concentration (282). Robinson & Sutherland (279) reported no significant variation in the extent of binding of benzylpenicillin and cloxacillin between different human subjects, and that the extent of binding of several antibiotics was largely independent of concentration of antibiotic below 100 $\mu$g/ml Von Wittenau & Delahunt (284) pointed out that the antibiotic concentration in interstitial fluid is largely determined by the equilibrium of drug between tissues and interstitial fluid, since a relatively small percentage of the administered dose is bound to serum proteins, even if the serum protein binding is high.

Krüger-Thiemer et al. (125, 280) have done some excellent work devising dosage schedules of antibacterial agents, based on maintaining minimum concentrations of free drug. The reviewer believes that future studies should be oriented towards gaining more insight into the significance of protein binding with respect to both therapeutic activity and pharmacokinetics, rather than fixation to standard formulas which are not necessarily supported adequately by scientific facts and appropriate correlations.

*Physiochemical factors.*—The physiochemical properties of both the drug and the dosage form in which the drug is administered are important in pharmacokinetics. Mechanisms of drug absorption and physiological transport of drugs have been reviewed by Schanker (285, 286). These reviews emphasize the importance of lipid/water partition coefficient, $pK_a$, and lipid

solubility in gastrointestinal absorption, drug distribution, and renal excretion of drugs. Gutman et al. (287) reported an inverse relationship between $pK_a$ and rate of renal excretion of phenylbutazone analogues in man and the dog. A change in $pK_a$ of only two units changed the plasma half life of the compounds from one to 72 hr. Such correlations in a series of analogues offer the medicinal chemist tremendous opportunities for improving the usefulness of his compounds, but the phenomenon seems to be studied very little. Gray & Ingelfinger (288) showed that the enzymatic hydrolytic step is not rate-limiting in the overall process of sucrose absorption in man, when sucrose is infused in solution in the intestinal tract. Wagner (85), Nelson (289), and others have pointed out that the absorption rate of a drug is often a function of the time needed for the drug to dissolve in the fluid at the site of absorption. Both the rate at which a given drug is absorbed and the efficiency of absorption may differ markedly when the type or nature of the dosage form in which the drug is administered is altered. Such effects are not necessarily of a low order of magnitude. Wagner et al. (77) reported that when indoxole was administered to human volunteers in an emulsion, serum concentrations were increased tenfold at a low dose and twentyfold at a high dose as compared with results obtained when the drug was administered as a very finely divided powder in a capsule. The new field of *biopharmaceutics* (85) encompasses the study of the relationship between the nature and intensity of the biological effects observed in animals and man, and the physiochemical and pharmaceutic properties of the drug, and the dosage form in which the drug is administered. One implication of biopharmaceutical knowledge is that poorly soluble drug candidates should probably be administered in divided dosage schedules in toxicological studies rather than in single large doses once a day. Another implication is that drug candidates should be administered in more than one dosage form during the early research and development phase (74). The study of drug dosage regimens and formulation design is greatly facilitated by use of the analogue computer (85, 173, 290, 291), but simulations are also quite feasible with a digital computer (35). With the development of new programs for the digital computer, the reviewer believes the digital computer will far surpass the analogue computer in this area in the years to come.

Levy and co-workers (292–294) studied the effect of complex formation on drug absorption in rats and goldfish. They reported that modification of barbiturate absorption by the surfactant polysorbate 80 represents the net effect of enhanced absorption and decreased thermodynamic activity of the drug due to micellar complexation. Their studies with acidic dyes suggested that the apparent lipid/water partition coefficient did not reflect the intestinal absorption characteristics of the dye complexes investigated. When salicylic acid was complexed separately with polysorbate 60 and caffeine, absorption rate in rats was modified. Japanese scientists (295–301) have performed some excellent investigations that tend to elucidate effect of dosage forms and their composition on gastrointestinal and rectal absorption of

sulfonamides, the mechanism of absorption of sulfonamides, and the effect of complexation and micelle formation on rectal absorption. Levy & Jusko (302) studied the effect of viscosity on drug absorption using ethanol and salicylic acid as model drugs. They claimed their results provided an explanation for the initial lag time encountered in absorption studies with salicylic acid and certain other drugs, and that the results rationalize the use of zero time shifts in the analysis of pharmacokinetic data.

Sorby (303, 304) showed that promazine was adsorbed by attapulgite and charcoal *in vitro*. Administration of promazine with the adsorbents resulted in decreased absorption rates and decreased total absorption in man, as judged by urinary excretion, compared with administration of promazine alone. Wagner (74) reported that the effect of the antidiarrheal preparation Kaopectate® on the absorption of the antibiotic lincomycin hydrochloride depended upon the time relationship of administration of the antidiarrheal preparation and the antibiotic. The phenomenon of reduced absorption of lincomycin hydrochloride when administered with the antidiarrheal preparation was correlated with *in vitro* studies in which the antibiotic was absorbed by the clays present in the antidiarrheal remedy.

O'Reilly et al. (305) studied the relationship between the *in vitro* dissolution kinetics and the *in vivo* intestinal absorption characteristics of tablet preparations of the coumarin anticoagulant, warfarin. The composition of the dissolution medium had a significant qualitative and quantitative effect on dissolution kinetics *in vitro*. In man, the kinetics of absorption were sometimes zero order or first order. The variation in kinetics was attributed to variation in pH of gastric fluids. Smith et al. (306) reported that an average of 2.23 times as much of the principal urinary metabolite of medroxyprogesterone acetate was excreted in eight hours after ingesting tablets prepared from micronized medroxyprogesterone acetate than was excreted following ingestion of tablets prepared from non-micronized steroid. The effects of particle size on dissolution and gastrointestinal absorption rates of other drugs were reviewed by Levy (307). Correlation of *in vitro* dissolution kinetics and rate and efficiency of gastrointestinal absorption were reviewed by Wagner (63) and Levy (308).

*Dose.*—The kinetics of absorption may change with increase in dose if a drug is absorbed at low doses by a saturable active transport mechanism. Most drugs are absorbed by a passive diffusion mechanism (285, 286), but some vitamins and drugs, which are analogues of endogenous substances, are absorbed at low doses by means of an active transport mechanism. For example, Spencer et al. (309) reported that the antitumor agents, azaserine (O-diazacetyl-L-serine) and DON (6-diazo-5-oxo-L-norleucine), which are glutamine analogues, behave like amino acids so far as transport by the hamster small intestine *in vitro* is concerned. Schanker et al. (310) reported that numerous purine and purine-like compounds inhibit the active transport of uracil across the intestinal wall *in vitro*, and presented evidence that suggests that purines and pyrimidines compete for a common transport process. Dietschy

& Carter (311) presented evidence that 5,5-dimethyl-2,4-oxazolidinedione is actively transported in everted gut sacs of rats. The uphill transport of the sulfonamide sulfisomezole was demonstrated by the everted sac technique, but it was suggested that this was explicable on the basis of the pH-partition hypothesis (296). Levy & Jusko (312) reported that the urinary recovery of riboflavin as a function of dose after oral administration to fasted normal subjects shows that the process responsible for the absorption of this vitamin is saturable. The saturation effect was not evident when riboflavin doses as high as 30 mg were administered after a meal.

Change in the efficiency of absorption, and possibly in the nature of the kinetics of absorption, of poorly water soluble substances with increase in dose has already been discussed. However, Wagner (235) has pointed out that apparent linearity of a cumulative urinary excretion plot may lead to the conclusion that constant rate (zero order) absorption is operative, when, in fact, all rate constants of the system are first order rate constants.

The kinetics of metabolism will be apparent first order when the concentration of drug is far below saturation of the enzyme by the substrate, and, specifically, when the drug concentration is very much less than the Michaelis constant (9, 178). When the enzyme is saturated by drug, the reaction becomes zero order. In an intermediate range of drug concentration, one may expect the reaction to be second order. Examples of zero order elimination at certain dose levels, due to saturation of the metabolizing enzyme, are alcohol in man (313) and the conversion of salicylate to salicylurate in rats (232). Constant excretion rate of the metabolite hippuric acid, following administration of high doses of benzoic acid in the rabbit, was reported by Bray et al. (181); the data suggested that the availability of glycine for conjugation controlled hippuric acid formation. Similarly, Levy & Matsuzawa (314) showed that man has a limited capacity for salicylamide sulfate formation, and their data suggested that the limiting factor was the availability of sulfate for conjugation.

There are several other reports of change in the nature of kinetics, or changes in the magnitude of elimination rates, with increase in dose, which are less understood. Following constant rate intravenous infusions and single intravenous injections of a fat emulsion in man, Hallberg (315) showed that at high concentrations of plasma triglyceride, elimination was zero order, but elimination became first order at low concentrations. Similar results were reported for dogs (316). The elimination of heparin was reported to be first order in man and the dog by Olson et al. (317), but the half life increased with increase in dose. In man, following doses of 100, 200, and 400 IU kg, the half lives were 56, 96, and 152 min, respectively. Dayton et al. (318) reported that in two subjects the plasma level decay rate of probenecid was faster following 0.5 g than following 2 g doses intravenously; this implied more rapid metabolism of probenecid at the lower dose. They reviewed the literature and cited streptomycin, dicoumarol, biscoumarate, amethopterin, and diphenylhydantoin as drugs which exhibited the same phenomenon of dose depen-

dence of first order disappearance rate from the plasma. Following high doses of phenylbutazone (204), the protein binding capacity of plasma proteins is exceeded (275) and, because of the larger fraction of free drug, the drug is cleared more rapidly from the body. There are other possible causes of dose dependence of half life. If the two-compartment open model (117, 124) applies, then a decrease in the distribution rate constant with increase in dose could cause the slope of the log blood level, time line to change with dose even though the true rate constant for loss from the body was independent of dose. Distribution of a drug may be increased as the dose is raised so that the drug reaches sites following high doses which are not reached following low doses; the rate of metabolism may be greater or less at one site than at another. Also a drug's metabolism or urinary excretion could be inhibited by itself or by one of its metabolites. In such a case, dose dependence of clearance rate would be expected.

*Route and method of administration.*—There are relatively large differences in the per cent of the dose which is metabolized and the per cent which is excreted unchanged in the urine, when neostigmine methylsulfate is administered intramuscularly as compared with orally (319–321). Similar results were reported by Cohn et al. (322) for benzquinamide; however, differences in efficiencies of absorption by the two routes could explain the results as well as the hypothesis presented by the authors.

Sullivan & Miller (323) reported that dosage of 5-fluoro-2′-deoxyuridine (5-FUDR), when given in 24 hr infusions in the treatment of human cancer, need be only 1/30 to 1/60 of that given as a single injection. Clarkson et al. (324) found that both 5-FUDR and 5-fluorouracil (5-FU) were degraded more completely when administered by continuous infusion than in a single injection. Also, very high local concentrations could be achieved by infusion into small arteries and by regional perfusion.

The assumption that a drug is handled the same following oral, intramuscular, and intravenous administration, without supporting evidence, may lead to erroneous conclusions.

*Diseased states.*—Fluid deposits such as are prevalent in patients with ascites alter distribution time and volume of distribution (114). Patients with impaired liver or renal function would be expected to clear drugs from the body more slowly if significant amounts of the drug are metabolized or excreted via the urine, respectively. Examples were given by Reinarz (325), Bates et al. (326), Bulger et al. (327) and Kutt et al. (328). However, Klipstein & Lindenbaum (329) reported more rapid clearance of folic acid than normal in several patients with chronic liver disease. Although Ueda et al. (330) reported longer half lives of tolbutamide in cirrhotic and renal patients than in normal subjects and diabetics, Nelson (331) claimed half lives of the drug were within normal range in patients with hepatic dysfunction. Results reported by Lindberg et al. (332) indicated that if renal excretion of chloramphenicol is impaired, then the drug normally excreted in the urine is metabolized.

*Enterohepatic circulation.*—Williams et al. (110) reviewed the influence of enterohepatic circulation on the toxicity of drugs. From the kinetic standpoint enterohepatic circulation may be very important. If a drug or metabolite or both are excreted in significant amounts in the feces, complete elucidation of the kinetics would require determination of whether the material in the feces was partly or wholly unabsorbed drug, drug which arose from biliary excretion, or drug which arose from secretion through the intestinal wall. Williams et al. (110) and Glazko (110) pointed out that enterohepatic circulation may be responsible for prolonged retention of certain drugs and metabolites in the body. Enterohepatic circulation may cause secondary peaks, shoulders, or oscillations in blood concentration, time curves (333, 334). These are usually most evident in the initial part of the curve following intravenous administration (334), but following oral administration may occur at later times particularly following food ingestion when the gall bladder evacuates (77, 333). If enterohepatic circulation is prominent, the compartment model should have a reversible transfer between the compartments representing blood and the absorption sites. For such models, the true rate constant for elimination cannot be estimated from the terminal blood concentration data (77).

*Miscellaneous factors.*—A number of other factors may contribute to variation in observed measurements such as blood concentrations or amounts excreted in the urine and in derived kinetic parameters. Wagner (74) reviewed the importance of many of these in pharmacokinetic studies and cited several examples.



## LITERATURE CITED


1. Snow, J., *London Med. Gaz.*, **39,** 498, 539 (1847); **41,** 893 (1848); **42,** 330 (1848); **46,** 749 (1850)
2. Butler, R. A. *Brit. J. Anaesthesia*, **36,** 193–99 (1964)
3. Marshall, E. K., Jr., Emerson, K., Jr., Cutting, W. C., *J. Pharmacol. Exptl. Therap.*, **61,** 191–95, 196–204 (1937)
4. Marshall, E. K., Jr., Cutting, W. C., Emerson, K., Jr., *J. Am. Med. Assoc.*, **110,** 252–57 (1938)
5. Teorell, T., *Arch. Intern. Pharmacodyn.*, **57,** 205–25, 226–40 (1937)
6. Stewart, J. D., Rourke, G. M., Allen, J. G., *J. Am. Med. Assoc.*, **110,** 1885–87 (1938)
7. Boxer, G. E., Jelinek, V. C., Tompsett, R., DuBois, R., Edison, A. O., *J. Pharmacol. Exptl. Therap.*, **92,** 226–35 (1948)
8. Nelson, E., *J. Pharm. Sci.*, **50,** 181–92 (1961)
9. Rescigno, A., Segre, G., *Drug and Tracer Kinetics* (Blaisdell Publ. Co., Waltham, Mass., 209 pp., 1966)
10. Fein, L., *Simulation*, 5, V–VII (1965)
11. Cornfield, J., Steinfeld, J., Greenhouse, S. W., *Biometrics*, **16,** 212–34 (1960)
12. Solomon, A. K., *Advan. Biol. Med. Phys.*, **3,** 65–97 (1953)
13. Berman, M., Schoenfeld, R., *J. Appl. Phys.*, **27,** 1361–70 (1956)
14. Ličko, V., *Bull. Math. Biophys.*, **27,** Suppl. 1, 15–19 (1965)
15. Nooney, G. C., *J. Chronic Diseases*, **19,** 325–32 (1966)
16. Sheppard, C. W., Householder, A. S., *J. Appl. Phys.*, **22,** 510–20 (1951)
17. Reiner, J. M., *Arch. Biochem. Biophys.*, **46,** 53–79 (1953)
18. Bellman, R., Roth, R. S., *J. Theoret. Biol.*, **11,** 168–76 (1966)
19. Booth, G. W., Peterson, T. I., *Nonlinear Estimation (IBM-Princeton)*, IBM 704 Program WL NLI, Rev. 1960 (Mathematics & Applications Dept., IBM, Inc., New York, N.Y.)
20. Turner, M. E., *Biometrics*, **19,** 183–88 (1963)

21. Schlender, B., Krüger-Thiemer, E., *Arzneimittel-Forsch.*, **12**, 992–1001 (1962)
22. Turner, M. E., Monroe, R. J., Homer, L. D., *Biometrics*, **19**, 406–28 (1963)
23. Haljok, C. A., *Instr. Control Systems*, **36**, 147 (1963)
24. Anon., *Simulation*, **2**, 38–41 (1964)
25. Berman, M., Shahn, E., Weiss, M. F., *Biophys. J.*, **2**, 275–87 (1962)
26. Berman, M., Weiss, M. F., Shahn, E., *Biophys. J.*, **2**, 289–316 (1962)
27. Perl, W., *Intern. J. Appl. Radiation Isotopes*, **8**, 211–22 (1960)
28. Savitzky, A., Golay, M. J. E., *Anal. Chem.*, **36**, 1627–39 (1964)
29. Stark, L., Sandberg, A. A., Stanten, S., Willis, P. A., Dickson, J. F., *Ann. N.Y. Acad. Sci.*, **115**, 738–62 (1964) Art. 2
30. Bradham, G. B., Bennett, L. R., Dehaven, F. C., Deland, E. C., Wolf, M. B., Maloney, J. V., *Surg. Gynecol. Obstet.*, **119**, 1062–68 (1964)
31. Hazelrig, J. B., *Mayo Clinic Proc.*, **39**, 841–48 (1964)
32. Wiegand, R. G., Sanders, P. G., *J. Pharmacol. Exptl. Therap.*, **146**, 271–75 (1964)
33. Nodine, J. H., Platt, J. M., Carranza, J., Dykyj, R., Mapp, Y., *Intern. J. Appl. Radiation Isotopes*, **15**, 263–68 (1964)
34. Krüger-Thiemer, E., *Antibiot. Chemotherapia*, **12**, 253–69 (1964)
35. Wagner, J. G., *Clin. Pharmacol. Therap.*, **8**, 201–18 (1967)
36. Bellman, R., Kalaba, R., *Proc. Natl. Acad. Sci. U.S.*, **54**, 1293–96 (1965)
37. Garfinkel, D., Ching, S. W., Adelman, M., Clark, P., *Ann. N.Y. Acad. Sci.*, **128**, 1054–68 (1966)
38. Beauchamp, J. J., Cornell, R. G., *Technometrics*, **8**, 319–26 (1966)
39. Box, G. E. P., *Ann. N.Y. Acad. Sci.*, **86**, 792–816 (1960) Art. 3
40. Box, G. E. P., Draper, N. R., *Biometrika*, **52**, 355–65 (1965)
41. Hartley, H. O., *Technometrics*, **3**, 269–80 (1961)
42. Hartley, H. O., Booker, A., *Ann. Math. Statistics*, **36**, 638–50 (1965)
43. Lipton, S., McGilchrist, C., *Biometrics*, **19**, 144–51 (1963)
44. Matz, A. W., *Appl. Statistics*, **13**, 118–27 (1964)
45. Myhill, J., Wadsworth, G. P., Brownell, G. L., *Biophys. J.*, **5**, 89–107 (1965)
46. Tornheim, L., *Technometrics*, **5**, 513–14 (1963)
47. Hartley, H. O., *Biometrika*, **51**, 347–53 (1964)
48. Guttman, I., Meeter, D. A., *Technometrics*, **7**, 623–37 (1965)
49. Box, G. E. P., Hunter, W. G., *Technometrics*, **4**, 301–18 (1962)
50. Box, G. E. P., Lucas, H. L., *Biometrika*, **46**, 77–90 (1959)
51. Box, G. E. P., Hunter, W. G., *Technometrics*, **7**, 23–42 (1965)
52. Hunter, W. G., Reiner, A. M., *Technometrics*, **7**, 307–23 (1965)
53. Box, G. E. P., Coutie, G. A., *Proc. Inst. Elect. Engrs. (London) Pt. B.*, **103**, Suppl. 1, 100–7 (1956)
54. Bartholomay, A. F., *Stochastic Models in Medicine and Biology*, Proc. Symp. Mathematics Res. Center, U. S. Army, Univ. Wis., Madison, Wis., June, 1963, **P. 105–12**
55. Hellman, L., Frazell, E. L., Rosenfeld, R. S., *J. Clin. Invest.*, **39**, 1288–94 (1960)
56. Schedl, H. P., Clifton, J. A., *Gastroenterology*, **41**, 491–99 (1961)
57. Schedl, H. P., Clifton, J. A., *J. Lab. Clin. Med.*, **62**, 1011–12 (1963)
58. Schedl, H. P., Clifton, J. A., Nokes, G., *J. Clin. Endocrinol. Metab.*, **24**, 224–26 (1964)
59. Berkowitz, D., Shapiro, J., Shapiro, B., *J. Am. Med. Assoc.*, **187**, 133–35 (1964)
60. Lutwak, L., Shapiro, J. R., *Science*, **144**, 1155–57 (1964)
61. Sund, R. B., Schou, J., *Acta Pharmacol. Toxicol.*, **21**, 313–25, 339–46 (1964)
62. Wagner, J. G., Nelson, E., *J. Pharm. Sci.*, **52**, 610–11 (1963)
63. Wagner, J. G., *The Case for In Vitro Rate of Dissolution as a Criterion of Rate of Absorption and/or Availability of Drug for Absorption*, 5th Natl. Ind. Pharm. Res. Conf., Land O'Lakes, Wis., 1963
64. Wagner, J. G., Nelson, E., *J. Pharm. Sci.*, **53**, 1392–1403 (1964)
65. Levy, G., Hollister, L. E., *J. Pharm. Sci.*, **53**, 1446–52 (1964)
66. Nelson, E., *J. Pharm. Sci.*, **54**, 1075–76 (1965)
67. Riegelman, S., Ballard, B. E. (Personal Communication to Nelson, E., Aug. 9, 1965)
68. Scholer, J. F., *Am. J. Digest. Diseases*, **7**, 43–49 (1962)
69. Levy, G., Miller, K. E., *J. Pharm. Sci.*, **53**, 1301–5 (1964)
70. Garrett, E. R., Thomas, R. C., Wallach, D. P., Alway, C. D., *J.*

*Pharmacol. Exptl. Therap.*, **130**, 106–18 (1960)
71. Stephenson, J. L., *Bull. Math. Biophys.*, **22**, 1–17, 113–38 (1960)
72. Silverman, M., Burgen, A. S., *J. Appl. Physiol.*, **16**, 911–13 (1961)
73. Birge, S. J., Jr., Peck, W. A., Berman, M., *Clin. Res.*, **XIV**, 347 (1966)
74. Wagner, J. G., *Can. J. Pharm. Sci.*, **1**, 55–68 (1966)
75. Levy, G., Hollister, L. E., *J. Pharm. Sci.*, **54**, 1121–25 (1965)
76. Hecht, G., Christian, J. E., Banker, G. S., *J. Pharm. Sci.*, **55**, 678–84 (1966)
77. Wagner, J. G., Gerard, E. S., Kaiser, D. G., *Clin. Pharmacol. Therap.*, **7**, 610–19 (1966)
78. Frost, A. A., Pearson, R. G., *Kinetics and Mechanism*, 2nd Ed. (Wiley & Sons, New York, 405 pp., 1962)
79. Borgström, B., Lundh, G., Hofmann A., *Gastroenterology*, **45**, 229–38 (1963)
80. Lindell, B., Strandberg, O., Reizenstein, P., *Phys. Med. Biol.*, **9**, 189–201 (1964)
81. Weisberg, H., Glass, G. B. J., *J. Lab. Clin Med.*, **68**, 163–72 (1966)
82. Hays, M. T., *Clin. Res.*, **XIV**, 281 (1966)
83. Smith, R. H., *Nature*, **198**, 161–62 (1963)
84. Nelson, E., Long, S., Wagner, J. G., *J. Pharm. Sci.*, **53**, 1224–27 (1964)
85. Wagner, J. G., *J. Pharm. Sci.*, **50**, 359–87 (1961)
86. Wagner, J. G., Carpenter, O. S., Collins, E. J., *J. Pharmacol. Exptl. Therap.*, **129**, 101–7 (1960)
87. Schück, O., Šmahel, O., *Antibiot. Med. Clin. Therapy*, **5**, 98–103 (1958)
88. Gladtke, E., *Antibiot. Chemotherapia*, **12**, 159–69 (1964)
89. Neipmann, W., *Klin. Wochschr.*, **39**, 1064–70 (1961)
90. Wagner, J. G., *J. Pharm. Sci.*, **56**, 652–53 (1967)
91. Dost, F. H., Gladtke, E., *Z. Klin. Chem.*, **1**, 14–18 (1963)
92. Middleton, E. J., Nagy, E., Morrison, A. B., *New Engl. J. Med.*, **274**, 136–39 (1966)
93. Dost, F. H., *Antibiot. Chemotherapia*, **12**, 149–58 (1964)
94. Diller, W., *Antibiot. Chemotherapia*, **12**, 85–94 (1964)
95. Dominguez, R., Kinetics of Elimination, Absorption and Volume of Distribution in the Organism, *Med. Physics*, **II**, 476–89 (1950)
96. Swintosky, J. V., Robinson, M. J., Foltz, E. L., *J. Am. Pharm. Assoc. Sci. Ed.*, **46**, 403–11 (1957)
97. Czaczkes, J. W., Kleeman, C. R., *J. Clin. Invest.*, **43**, 1649–58 (1964)
98. Wagner, J. G., Alway, C. D., *Nature*, **201**, 1101–3 (1964)
99. Butler, T. C., *Federation Proc.*, **17**, 1158–62 (1958)
100. Heatley, N. G., *Antibiot. Med. Clin. Therapy*, **2**, 33–41 (1956)
101. Chulski, T., Johnson, R. H., Schlagel, C. A., Wagner, J. G., *Nature*, **198**, 450–53 (1963)
102. Nelson, E., Levy, G., *Nature*, **197**, 1269–70 (1963)
103. Leach, C. N., Jr., Peterson, R. E., Lukas, D. S., *Clin. Res.*, **XIV**, 254 (1966)
104. Lukas, D. S., Peterson, R. E., *J. Clin. Invest.*, **45**, 782–95 (1966)
105. Smith, D. L., Wagner, J. G., Gerritsen, G. C., *J. Pharm. Sci.*, **56**, 1150–57 (1967)
106. Krüger-Thiemer, E., *Arzneimittel-Forsch.*, **14**, 1332–43 (1964)
107. Wagner, J. G., *Antibiot. Chemotherapia*, **12**, 53–84 (1964)
108. Rall, D. P., Zubrod, C. G., *Ann. Rev. Pharmacol.*, **2**, 109-28 (1962)
109. Burgen, A. S. V., *J. Cellular Comp. Physiol.*, **48**, 113–38 (1956)
110. Williams, R. T., Millburn, P., Smith, R. L., *Ann. N. Y. Acad. Sci.*, **123**, 110–24 (1965)
111. Grady, J. E., Stern, K. F., *Antimicrobial Agents Chemotherapy*, 201–5 (1965)
112. Matthews, C. M. E., *Phys. Med. Biol.*, **2**, 36-53 (1957)
113. Rosenthal, R. K., Bayles, T. B., Fremont-Smith, K., *Arthritis Rheumat.*, **7**, 103–9 (1964)
114. Soberman, R., Brodie, B. B., Levy, B. B., Axelrod, J., Hollander, V., Steele, J. M., *J. Biol. Chem.*, **179**, 31–42 (1949)
115. Stokes, J. M., Ter-Pogossian, M. M., *J. Am. Med. Assoc.*, **187**, 20–23 (1964)
116. Fozzard, H. A., *Am J. Physiol.*, **206**, 309–12 (1964)
117. Wagner, J. G., Northam, J. I., *J. Pharm. Sci.*, **56**, 529–31 (1967)
118. Albert, S. N., Gravel, Y., Turmel, Y., Albert, C. A., *Anesthesia Analgesia, Current Res.*, **44**, 805–13 (1965)
119. Samachson, J., *Radiation Res.*, **27**, 64–74 (1966)
120. Cavalieri, R. R., Searle, G. L., *J. Clin. Invest.*, **45**, 939–49 (1966)
121. Bloom, P. M., Nelp, W. B., Tuell,

S. H., *Am. J. Med. Sci.*, **251**, 133–44 (1966)
122. Wagner, J. G., *J. Pharm. Sci.*, **56**, 489–94 (1967)
123. Shore, M. L., *J. Appl. Physiol.*, **16**, 771–82 (1961)
124. Sapirstein, L. A., Vidt, D. G., Mandel, M. J., Hanusek, G., *Am. J. Physiol.*, **181**, 330–36 (1955)
125. Krüger-Thiemer, E., Bünger, P., *Chemotherapia*, **10**, 61–73, 129–44 (1965–66)
126. Krüger-Thiemer, E., Bünger, P., Evaluation of the Risk of Crystalluria with Sulfa Drugs, *Experimental Studies and Clinical Experience-The Assessment of Risk, Vol. VI, Proc. European Soc. Study Drug Toxicity*, Stockholm, Sweden, June, 1965 (Reprinted from Excerpta Med. Intern. Congr. Ser. No. 97, 185–207)
127. Tait, J. F., Little, B., Tait, S. A. S., Flood, C., *J. Clin. Invest.*, **41**, 2093–2100 (1962)
128. Rutherford, E. R., Chadwick, J., Ellis, C. D., *Radiation from Radioactive Substances* (Cambridge Univ. Press, London, 588 pp., 1951)
129. Whipple, Harold E., Ed., *Multicompartment Analysis of Tracer Experiments, Ann. N. Y. Acad. Sci.*, **108**, 1–338 (1963 Art. 1)
130. Tait, J. F., Tait, S. A. S., Little, B., Laumas, K. R., *J. Clin. Invest.*, **40**, 72–80 (1961)
131. Robertson, J. S., *Physiol. Rev.*, **37**, 133–54 (1957)
132. Russell, J. A., *Perspectives Biol. Med.*, **1**, 138–73 (1958)
133. Fukuda, N., Sugito, M., *J. Theoret. Biol.*, **1**, 440–59 (1961)
134. Samachson, J., *J. Appl. Physiol.*, **18**, 824–28 (1963)
135. Peterson, R. E., *Recent Progr. Hormone Res.*, **XV**, 231–74 (1959)
136. Gurpide, E., Mann, J., Lieberman, S., *J. Clin. Endocrinol. Metab.*, **23**, 1155–76 (1963)
137. Lax, L. C., Wrenshall, G. A., *Nucleonics*, **11**, 18–20 (1953)
138. DeHertogh, R., Hoet, J. J., Materazzi, F., Ekka, E., *Acta Endocrinol.*, **47**, 165–76 (1964)
139. Sharney, L., Wasserman, L. R., Gevirtz, N. R., Schwartz, L., Tendler, D., *Am. J. Med. Electronics*, **4**, 95–99 (1965)
140. Cartwright, G. E., Athens, J. W., Wintrobe, M. M., *Blood*, **24**, 780–801 (1964)
141. Turco, G. L., Ghemi, F., Molino, G., Segre, G., *J. Lab. Clin. Med.*, **67**, 983–93 (1966)
142. Campbell, R. M., Cuthbertson, D. P., Matthews, C. M., McFarlane, A. S., *Intern. J. Appl. Radiation Isotopes*, **1**, 66–84 (1956)
143. Gellhorn, A., Merrell, M., Rankin, R. M., *Am. J. Physiol.*, **142**, 407–27 (1944)
144. Solomon, A. K., *J. Clin. Invest.*, **28**, 1297–1307 (1949)
145. Cohn, W. E., Brues, A. M., *J. Gen. Physiol.*, **28**, 449–61 (1945)
146. Wrenshall, G. A., *Can. J. Biochem. Physiol.*, **33**, 909–25, 940–47 (1955)
147. Hart, H. E., *Bull. Math. Biophys.*, **17**, 87–94 (1957)
148. Wendel, O. W., *Phys. Biol. Med.*, **5**, 411–30 (1961)
149. Jokipii, S. G., Turpenein, O., *J. Clin. Invest.*, **33**, 452–58 (1954)
150. Cohn, S. H., Lippincott, S. W., Gusmano, E. A., Robertson, J. S., *Radiation Res.*, **19**, 104–19 (1963)
151. Bronner, F., Richelle, L. J., Saville, P. D., Nicholas, J. A., Cobb, J. R., *J. Clin. Invest.*, **42**, 898–905 (1963)
152. Swain, C. G., *J. Am. Chem. Soc.*, **66**, 1696–1700 (1944)
153. Solomon, A. K., Compartmental Methods of Kinetic Analysis, *Mineral Metabolism*, 119–67, Vol. I, Pt. A (Comar, C. L., Bronner, F., Eds., Academic Press, New York, 386 pp., 1963)
154. Rescigno, A., *Biochim. Biophys. Acta*, **21**, 111–16 (1956)
155. Berman, M., *J. Theoret. Biol.*, **4**, 229–36 (1963)
156. Ackerman, E., Rosevear, J. W., McGurkin, W. F., *Phys. Med. Biol.*, **9**, 203–13 (1964)
157. Ackerman, E., Gatewood, L., Rosevear, J., Molnar, G., *Bull. Math. Biophys.*, **27**, 21–37 Special Issuc (1965)
158. Conn, R. B., Sabo, A. J., Landes, D., Ho, J. Y. L., *Nature*, **203**, 143–46 (1964)
159. Shaney, L., Wasserman, L. R., Gevirtz, N. R., *Am. J. Med. Electronics*, **3**, 249–60 (1964)
160. Leitner, F., *Bull. Math. Biophys.*, **27**, 431–34 (1965)
161. Segre, G., *Bull. Math. Biophys.*, **27**, 49–63 (1965)
162. Apter, J. T., *J. Am. Med. Assoc.* **195**, 931–34 (1966)
163. Moses, V., Lonberg-Holm, K. K., *J. Theoret. Biol.*, **10**, 336–55 (1966)
164. Nelson, E. *J. Am. Pharm. Assoc. Sci. Ed.*, **48**, 96–103 (1959)

165. Nelson E. *J. Am. Pharm. Assoc. Sci. Ed.*, **49**, 54–56 (1960)
166. Nelson, E., O'Reilly, I., *J. Pharmacol. Exptl. Therap.*, **129**, 368–72 (1960)
167. Nelson, E., *J. Theoret. Biol.*, **2**, 193–200 (1962)
168. Nelson, E., Morioka, T., *J. Pharm. Sci.*, **52**, 864–68 (1963)
169. Nelson, E., *Antibiot. Chemotherapia*, **12**, 29–40 (1964)
170. Campbell, J. A., Nelson, E., Chapman, D. G., *Can. Med. Assoc. J.*, **81**, 15–20 (1959)
171. Garrett, E. R., Johnston, R. L., Collins, E. J., *J. Pharm. Sci.*, **52**, 668–78 (1963)
172. Garrett, E. R., *Antibiot. Chemotherapia*, **12**, 227–42 (1964)
173. Garrett, E. R., Lambert, H. J., *J. Pharm. Sci.*, **55**, 626–34 (1966)
174. Stelmach, H., Robinson, J. R., Eriksen, S. P., *J. Pharm. Sci.*, **54**, 1453–58 (1965)
175. Riggs, D. S., *The Mathematical Approach to Physiological Problems* (Williams & Wilkins, Baltimore, 445 pp., 1963)
176. Wiegand, R. G., Taylor, J. D., *Biochem. Pharmacol.*, **3**, 256–63 (1960)
177. Taylor, J. D., Wiegand, R. G., *Clin. Pharmacol. Therap.*, **3**, 464–72 (1962)
178. Levy, G., *J. Pharm. Sci.*, **54**, 959–67 (1965)
179. Levy, G., *J. Pharm. Sci.*, **55**, 989 (1966)
180. Von Wittenau, M. S., Yeary, R., *J. Pharmacol. Exptl. Therap.*, **140**, 258–66 (1963)
181. Bray, H. G., Thorpe, W. V., White, K., *Biochem. J.*, **48**, 88–96 (1951)
182. Elliott, H. C., *Proc. Soc. Exptl. Biol. Med.*, **121**, 861–64 (1966)
183. Portmann, G. A., McChesney, E. W., Stander, H., Moore, W. E., *J. Pharm. Sci.*, **55**, 59–62 (1966)
184. Moore, W. E., Portmann, G. A., Stander, H., McChesney, E. W., *J. Pharm. Sci.*, **54**, 36–41 (1965)
185. Martin, B. K., *Nature*, **207**, 959–60 (1965)
186. Krüger-Thiemer, E., *Klin. Wochschr.*, **38**, 514–20 (1960)
187. Carpenter, O. S., Northam, J. I., Wagner, J. G., Use of Kinetic Models in the Study of Antibiotic Absorption, Metabolism and Excretion, *Antibiotics–Advances in Research, Production and Clinical Use*, 165–72 (Herold, M., Gabriel, Z., Eds., Butterworth, London, 778 pp., 1966)
188. Koppanyi, T., Avery, M. A., *Clin. Pharmacol. Therap.*, **7**, 250–70 (1966)
189. Wagner, J. G., *J. Clin. Pharmacol. & J. New Drugs*, **7**, 84–88 (1967)
190. Krüger-Thiemer, E., *J. Am. Pharm. Assoc. Sci. Ed.*, **49**, 311–13 (1960)
191. Wagner, J. G., Northam, J. I., Alway, C. D., Carpenter, O. S., *Nature*, **207**, 1301–2 (1965)
192. Widmark, E., Tandberg, J., *Biochem. Z.*, **147**, 358–69 (1924)
193. Druchrey, H., Küpfmüller, K., *Dosis und Wirkung. Beiträge Zur theoretischen Pharmakologie* (Pharmazie, 8. Beiheft, 1. Erg. Bd., 135 pp., 1949)
194. Dost, F. H., *Der Blutspiegel. Kinetik der Konzenstrationsablöufe in der Kreislaufflüssigkeit* (Thieme, Leipzig, 362 pp., 1953)
195. Nelson, E., *J. Theoret. Biol.*, **2**, 193–200 (1962)
196. Wiegand, R. G., Buddenhagen, J. D., Endicott, C. J., *J. Pharm. Sci.*, **52**, 268–73 (1963)
197. Wagner, J. G., *Kinetic Analysis of Blood Levels*, Natl. Ind. Pharm. Res. Conf., 6th, Land O'Lakes, Wis., June, 1964
198. Brusenback, R., Wiegand, R., *Simulation of Multiple Dose Drug Kinetics*, Midwestern Simulation Council Mtn., Northwestern Technological Inst., Northwestern Univ., Evanston, Ill., October, 1963
199. Engberg-Pedersen, H., Mørch, P., Tybring, L., *Brit. J. Pharmacol.*, **23**, 1–13 (1964)
200. Krüger-Thiemer, E., *Application of Kinetic Analysis in Clinical Pharmacology*, Postgrad. Course, Animal & Clinical Pharmacologic Techniques in Drug Evaluation, Hahnemann Med. Coll. & Hosp. and Am. Therap. Soc., Philadelphia, January, 1966
201. Kwan, K. C., Zirrith, M. P., Costello, R. A., Marcus, A. D., *A Mathematical Treatment of Repetitive Dose Kinetics*, Acad. Pharm. Sci., Dallas, Texas, April, 1964
202. McChesney, E. W., Portmann, G. A., Koss, R. F., *J. Pharm. Sci.*, **56**, 594–99 (1967)
203. Brodie, B. B., *Clin. Pharmacol. Therap.*, **3**, 374–80 (1962)
204. Burns, J. J., Rose, R. K., Chenkin, T., Goldman, A., Schulert, A., Brodie, B. B., *J. Pharmacol. Exptl. Therap.*, **109**, 346–57 (1953)
205. Swintosky, J. V., Bondi, A., Jr., Robin-

son, M. J., *J. Am. Pharm. Assoc. Sci. Ed.*, **47**, 753–56 (1958)
206. Krüger-Thiemer, E., Büinger, P., *Arzneimittel-Forsch.*, **11**, 867–74 (1961)
207. Forney, R. B., Hughes, F. W., *Clin. Pharmacol. Therap.*, **4**, 619–21 (1963)
208. Sutherland, J. M., *Antimicrobial Therapy of the Newborn Infant*, Intern. Congr. Chemotherapy, Proc., 3rd, Stuttgart, July, 1963 (1964)
209. Cummings, A. J., Martin, B. K., *Biochem. Pharmacol.*, **13**, 767–76 (1964)
210. Sugerman, A. A., Rosen, E., *Clin. Pharmacol. Therap.*, **5**, 561–68 (1964)
211. Huang, C. L., Ruskin, B. H., *J. Nervous Mental Disease*, **139**, 381–86 (1964)
212. Rosen, E., Tannenbaum, P., Ellison, T., Free, S. M., Crosley, A. P., *J. Am. Med. Assoc.*, **194**, 1203–5 (1965)
213. Mellinger, T. J., Mellinger, E. M., Smith, W. T., *Clin. Pharmacol. Therap.*, **6**, 486–91 (1965)
214. Bell, S. A., Berdick, M., Holliday, W. M., *J. New Drugs*, **6**, 284–93 (1966)
215. Green, D. M., *J. New Drugs*, **6**, 294–304 (1966)
216. Hermelin, V. M., *Drug Cosmetic Ind.*, **94**, 36–37, 90 (1964)
217. Gibaldi, M., Kanig, J. L., *J. Pharm. Sci.*, **52**, 306–8 (1963)
218. Wagner, J. G., *J. Pharm. Sci.*, **52**, 1097–1101 (1963)
219. Thomas, R. C., Ikeda, G. J., *J. Med. Chem.*, **9**, 507–10 (1966)
220. Nelson, E., Knoechel, E. L., Hamlin, W. E., Wagner, J. G., *J. Pharm. Sci.*, **51**, 509–14 (1962)
221. Wallach, D. P., Wagner, J. G., *J. Pharmacol. Exptl. Therap.*, **124**, 16–24 (1958)
222. Bolinger, R. E., Morris, J. H., McKnight, F. G., Diederich, D. A., *New Engl. J. Med.*, **270**, 767–70 (1964)
223. Doluisio, J. T., Swintosky, J. V., *Am. J. Pharm.*, **137**, 175–93 (1965)
224. Moore, W. E., Portmann, G. A., Stander, H., McChesney, E. W., *J. Pharm. Sci.*, **54**, 36–41 (1965)
225. Borzelleca, J. F., Lowenthal, W., *J. Pharm. Sci.*, **55**, 151–54 (1966)
226. Cameron, J. L., Pulaski, E. J., Abei, T., Iber, F. L., *Ann. Surg.*, **163**, 330–38 (1966)
227. Fincher, J. H., Adams, J. G., Beal, H. M., *J. Pharm. Sci.*, **54**, 704–8 (1965)
228. Seneca, H., Peer, P., *Antimicrobial Agents Chemotherapy*, 657–61 (1963)
229. MacDonald, H., Place, V., Diermeier, H., Dornbush, A., Forbes, M., Kulkarni, S., *Antimicrobial Agents Chemotherapy*, 173–78 (1964)
230. Schachter, D., Manis, J., *J. Clin. Invest.*, **37**, 800–7 (1958)
231. Levy, G., *J. Pharm. Sci.*, **54**, 496 (1965)
232. Nelson, E., Hanano, M., Levy, G., *J. Pharmacol. Exptl. Therap.*, **153**, 159–66 (1966)
233. Levy, G., Amsel, L. P., *Biochem. Pharmacol.*, **15**, 1033–38 (1966)
234. Cummings, A. J., Martin, B. K., Renton, R., *Brit. J. Pharmacol.*, **26**, 461–67 (1966)
235. Wagner, J. G., *J. Pharm. Sci.*, **56**, 586–94 (1967)
236. Lineweaver, H., Burk, D., *J. Am. Chem. Soc.*, **56**, 658–66 (1934)
237. Michaelis, L., Menton, M. L., *Biochem. Z.*, **49**, 333–69 (1913)
238. Juncher, H., Raaschov, F., *Antibiot. Med. Clin. Therapy*, **4**, 497–507 (1957)
239. Levy, G., Hollister, L. E., *Brit. Med. J.*, **II**, 286–88 (1964)
240. Hollister, L. E., Levy, G., *Chemotherapia*, **9**, 20–24 (1964)
241. Brodie, B. B. (Paper read by Gillette, J.), *Theoretical Aspects of Drug Interaction*, Postgrad. Course, Animal & Clinical Pharmacologic Techniques in Drug Evaluation, Hahnemann Med. Coll. & Hosp., Am. Therap. Soc., Philadelphia, January 1966
242. Bleifeld, W., Gehrmann, G., *Ger. Med. Monthly*, **11**, 12-14 (1966)
243. Portnoff, J. B., Swintosky, J. V., Kostenbauder, H. B., *J. Pharm. Sci.*, **50**, 890 (1961)
244. Beckett, A. H., Rowland, M., *Nature*, **204**, 1203–4 (1964)
245. Smith, H. W., *The Kidney: Structure and Function in Health and Disease* (Oxford Univ. Press, New York, 1049 pp., 1951)
246. Levy, G., *Am. J. Med. Sci.*, **250**, 174–76 (1965)
247. Kostenbauder, H. B., *Drug Combinations-Studies of Modification of Biological Half-Lives of Drugs in Human Subjects*, Seminar presented at The Upjohn Co., Kalamazoo, Mich., January, 1966
248. Kunin, C. M., *Clin. Pharmacol. Therap.*, **7**, 166–79, 180–88 (1966)
249. Burns, J. J., Conney, A. H., Koster,

R., *Ann. N. Y. Acad. Sci.*, **104**, 881–93 (1963) Art. 3
250. Conney, A. H., Schneidman, K., Jacobson, M., Kuntzman, R., *Ann. N. Y. Acad. Sci.*, **123**, 98–109 (1965)
251. Burns, J. J., Conney, A. H., *Proc. Roy. Soc. Med.*, **58**, 955–60 (1965)
252. Kalow, W., *Ann. N. Y. Acad. Sci.*, **104**, 894–904 (1963) Art. 3
253. Evans, D. A. P., White, T. A., *J. Lab. Clin. Med.*, **63**, 394–403 (1964)
254. Andersen, C. E., *Acta Physiol. Scand.*, **59**, *Suppl.* 213, 11 (1963)
255. Nelson, E., *J. Theoret. Biol.*, **5**, 493–96 (1963)
256. Porter, I. H., *Tox. Appl. Pharmacol.*, **6**, 499–511 (1964)
257. Meier, H., *Experimental Pharmacogenetics-Physiopathology of Heredity and Pharmacologic Responses* (Academic Press, New York, 213 pp., 1963)
258. Kalow, W., *Pharmacogenetics* (Saunders, Philadelphia, 231 pp., 1965)
259. Milne, M. D., Schribner, B. N., Crawford, M. A., *Am. J. Med.*, **24**, 709–29 (1958)
260. Asatoor, A. M., Galman, B. R., Johnson, J. R., Milne, M. D., *Brit. J. Pharmacol.*, **24**, 293–300 (1965)
261. Beckett, A. H., Wilkinson, G. R., *J. Pharm. Pharmacol.*, **17**, 256–57 (1965)
262. Beckett, A. H., Rowland, M., *J. Pharm. Pharmacol.*, **17**, 628–39 (1965)
263. Orloff, J., Berliner, R. W., *J. Clin. Invest.*, **35**, 223–35 (1956)
264. Beckett, A. H., *Dansk. Tidsskr. Farm.*, **40**, 197–223 (1966)
265. Steinmetz, P. R., Bank, N., *J. Clin. Invest.*, **42**, 1142–49 (1963)
266. Steinmetz, P. R., Eisinger, R. P., *Metabolism*, **15**, 76–87 (1966)
267. Mandell, A. J., Chaffey, B., Brill, P., Mandell, M. P., Rodnick, J., Rubin, R. T., Sheff, R., *Science*, **151**, 1588–60 (1966)
268. Gibaldi, M., *An Investigation of the Gastrointestinal Absorption and Pharmacokinetics of Selected Compounds in Man* (Doctoral thesis, Columbia Univ., New York, 1965, Dissertation Abstracts, **26**, 64–9185, #2, August, 1965)
269. Gibaldi, M., Kanig, J. L., *Arch. Intern. Pharmacodyn.*, **161**, 343–58 (1966)
270. Smart, J. V., Turner, P., *Brit. J. Pharmacol.*, **26**, 468–72 (1966)
271. Koizumi, T., Arita, T., Kakemi, K., *Chem. Pharm. Bull. (Tokyo)*, **12**, 428–32 (1964)
272. Conn, R. B., Sabo, A. J., Landes, D., Ho., J. Y. L., *Nature*, **203**, 143-46 (1964)
273. Wagner, J. G., *J. Clin. Pharmacol. & J. New Drugs*, **7**, 89–92 (1967)
274. Goldstein, A., *Pharmacol. Rev.*, **1**, 102–65 (1949)
275. Thorp, J. M., The Influence of Plasma Proteins in the Action of Drugs, *Absorption and Distribution of Drugs*, 64–76 (Binns, T. B., Dodds, C., Eds., Williams & Wilkins, Baltimore, Md., 270 pp., 1964)
276. Martin, B. K., *Nature*, **207**, 274–76 (1965)
277. Burns, J. J., Rose, R. K., Chenkin, T., Goldman, A., Schulert, A., Brodie, B. B., *J. Pharmacol. Exptl. Therap.*, **109**, 346–57 (1953)
278. Scholtan, V. W., *Arzneimittel-Forsch.*, **13**, 347–60 (1963)
279. Robinson, G. N., Sutherland, R., *Brit. J. Pharmacol.*, **25**, 638–50 (1965)
280. Krüger-Thiemer, E., Diller, W., Bünger, P., *Antimicrobial Agents Chemotherapy*, 183–91 (1965)
281. Madsen, S. T., Øvsthus, Ø., Bøe, J., *Acta Med. Scand.*, **173**, 707–17 (1963)
282. Kaplan, K., Chew, W. H., Weinstein, L., *Clin. Pharmacol. Therap.*, **6**, 316–20 (1965)
283. Batterman, R. C., Tauber, L. F., Bell, M. E., *Current Therap. Res.*, **8**, 75–84 (1966)
284. Von Wittenau, M. S., Delahunt, C. S., *J. Pharmacol. Exptl. Therap.*, **152**, 164–69 (1966)
285. Schanker, L. S., *Ann. Rev. Pharmacol.*, **1**, 29–44 (1961)
286. Schanker, L. S., *Advan. Drug Res.*, —71–106 (1964)
287. Gutman, A. B., Dayton, P. G., Yü, T. F., Berger, L., Chen, W., Sicam, L. E., Burns, J. J., *Am. J. Med.*, **29**, 1017–33 (1960)
288. Gray, G. M., Ingelfinger, F. J., *J. Clin Invest.*, **45**, 388–98 (1966)
289. Nelson, E., *Clin. Pharmacol. Therap.*, **3**, 673–81 (1962)
290. Stelmach, H., Robinson, J. R., Eriksen, S. P., *J. Pharm. Sci.*, **54**, 1453–58 (1965)
291. Ballard, B. E., Goyan, J. E., *Med. & Biol. Engng.*, **4**, 483–90 (1966)
292. Levy, G., Reuning, R. H., *J. Pharm. Sci.*, **53**, 1471–75 (1964)
293. Levy, G., Matsuzawa, T., *J. Pharm. Sci.*, **54**, 1003–6 (1965)
294. Levy, G., Miller, K. E., Reuning, R.

H., *J. Pharm. Sci.*, **55**, 394–98 (1966)
295. Kakemi, K., Arita, T., Sezaki, H., Sugimoto, I., *Arch. Pract. Pharm.*, **25**, 19–22 (1965)
296. Nogami, H., Hanano, M., Watanabe, J., *Chem. Pharm. Bull. (Tokyo)*, **12**, 1465–71 (1964)
297. Nogami, H., Hanano, M., Aruga, M., *Chem. Pharm. Bull. (Tokyo)*, **14**, 166–73 (1966)
298. Koizumi, T., Arita, T., Kakemi, K., *Chem. Pharm. Bull. (Tokyo)*, **12**, 413–20, 421–27 (1964)
299. Kakemi, K., Arita, T., Koizumi, T., *Arch. Pract. Pharm.*, **25**, 22–25 (1965)
300. Kakemi, K., Arita, T., Muranishi, S., *Chem. Pharm. Bull. (Tokyo)*, **13**, 969–75, 976–85 (1965)
301. Yamada, H., Yamamoto, R., *Chem. Pharm. Bull. (Tokyo)*, **13**, 1279–84 (1965)
302. Levy, G., Jusko, W. J., *J. Pharm. Sci.*, **54**, 219–25 (1965)
303. Sorby, D. L., *J. Pharm. Sci.*, **54**, 677–83 (1965)
304. Sorby, D. L., Lin, G., *J. Pharm. Sci.*, **55**, 504–10 (1966)
305. O'Reilly, R. A., Nelson, E., Levy, G., *J. Pharm. Sci.*, **55**, 435–37 (1966)
306. Smith, D. L., Pulliam, A. L., Forist, A. A., *J. Pharm. Sci.*, **55**, 398–403 (1966)
307. Levy, G., *Am. J. Pharm.*, **135**, 78–92 (1963)
308. Levy, G., *Correlation of Gastrointestinal Absorption and In Vitro Dissolution Kinetics of Drugs Contained in Solid Oral Dosage Forms*, E. Nelson Memorial Symp. Biopharmaceutics, 113th Ann. Meeting Am. Pharm. Assoc., Dallas, Texas, 1966
309. Spencer, B. P., Bow, T. M., Markulis, M. A., *Biochem. Pharmacol.*, **12**, 599 (1963)
310. Schanker, L. S., Jeffrey, J. J., Tocco, D. J., *Biochem. Pharmacol.*, **12**, 1047–53 (1963)
311. Dietschy, J. M., Carter, N. W., *Science*, **150**, 1294–1296 (1965); **152**, 799 (1966)
312. Levy, G., Jusko, W. J., *J. Pharm. Sci.*, **55**, 285–89 (1966)
313. Lundquist, F., Wolthers, H., *Acta Pharmacol. Toxicol.*, **14**, 265–89 (1958)
314. Levy, G., Matsuzawa, T., *J. Pharm. Sci.*, **55**, 222–23 (1966)
315. Hallberg, D., *Acta Physiol. Scand.*, **64**, 299–305, 306–13 (1965)
316. Carlson, L. A., Hallberg, D., *Acta Physiol. Scand.*, **59**, 52–61 (1963)
317. Olsson, P., Lagergren, H., Stig, E., *Acta Med. Scand.*, **173**, 619–30 (1963)
318. Dayton, P. G., Yü, T. F., Chen, W., Berger, L., West, L. A., Gutman, A. B., *J. Pharmacol. Exptl. Therap.*, **140**, 278–86 (1963)
319. Nowell, P. T., Scott, C. A., Wilson, A., *Brit. J. Pharmacol.*, **18**, 617–24 (1962)
320. Scott, C. A., Nowell, P. T., Wilson, A., *J. Pharm. Pharmacol.*, **14**, 31T–33T (1962)
321. Roberts, J. B., Thomas, B. H., Wilson, A., *Nature*, **200**, 1330 (1963)
322. Cohn, B., Brahen, L. S., Wiseman, E. H., Pinson, R., *Current Therap. Res.*, **5**, 301–4 (1963)
323. Sullivan, R. D., Miller, E., *Cancer Res.*, **25**, 1025–31 (1965)
324. Clarkson, B., O'Connor, A., Winston, L., Hutchinson, D., *Clin. Pharmacol. Therap.*, **5**, 581–610 (1964)
325. Reinarz, J. A., *Antimicrobial Agents Chemotherapy*, 232–38 (1965)
326. Bates, J. H., Schultz, J. C., Abernathy, R. S., *Clin. Res.*, **13**, 77 (1965)
327. Bulger, R. J., Lindholm, D. D., Murray, J. S., Kirby, W. M. M., *J. Am. Med. Assoc.*, **187**, 319–22 (1964)
328. Kutt, H., Winters, W., Scherman, R., McDowell, F., *Arch. Neurol.*, **11**, 649–56 (1964)
329. Klipstein, F. A., Lindenbaum, J., *Clin. Res.*, **12**, 210 (1964)
330. Ueda, H., Sakurni, T., Ota, M., Nakajima, A., Kamii, K., Maezawa, H., *Diabetes*, **12**, 414–19 (1963)
331. Nelson, E., *Am. J. Med. Sci.*, **248**, 657–59 (1964)
332. Lindberg, A. A., Nilsson, L. H., Bucht, H., Kallings, L. O., *Brit. Med. J.*, **2**, 724–28 (1966)
333. Fujii, R., Konno, M., Matuzaki, M., Takeshita, M., Uno, S., *J. Antibiotics (Tokyo), Ser. B.*, **16**, 371–77 (1963)
334. Turco, G. L., de Filippi, P., Prinetti, V., Segre, G., *Clin. Pharmacol. Therap.*, **7**, 603–9 (1966)

# CONTENTS