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Research Articles

In Vivo Pharmacodynamic Evaluation of Oral Dosage Forms by Whole Body Liquid Scintillometry

By GERALD HECHT*, JOHN E. CHRISTIAN, and GILBERT S. BANKER

A large volume liquid scintillation detector was used to determine rates of excretion, absorption, and intercompartmental clearance in ambulatory dogs. A γ-emitting test substance was administered intravenously, orally in aqueous solution, and orally in sustained and delayed-release dosage forms. Unanesthetized female dogs were partially restricted, catheterized, and fasted prior to dosing. Where applicable, plots of log per cent whole body retention as a function of time were resolved into linear components following apparent first-order kinetics. The rate constants of these components were calculated and compared. Sustained-release forms were prepared which exhibited zero-order release characteristics *in vitro* and *in vivo*, and the characteristics of an enteric coated dosage form were compared after *in vitro* and *in vivo* testing. The procedure provided such parameters as absorption, excretion, release, and the effect of formulation techniques and formula variation on the biological availability of the test substance employed without the necessity of excreta and blood sampling and analysis.

THE PROBLEM of evaluating the biological availability and pharmacodynamic properties of any drug, new or old, in a new dosage form, is of increasing importance due to the greater potency and specificity of drugs, the greater complexity of dosage forms, and the increasing demands of the drug laws and new drug application requirements.

Several authors (1–9) have attempted to standardize the *in vitro* testing of various oral dosage forms, not necessarily as a means of simulating the characteristics which would be expected *in vivo*, but rather as a means of insuring control and correlation with data collected *in vivo*.

Many methods for the *in vivo* evaluation of oral dosage forms have been devised, but undoubtedly, those supplying the most pertinent information are concerned with the evaluation of absorption,

Received March 21, 1966, from the Bionucleonics and Industrial Pharmacy Departments, School of Pharmacy and Pharmacal Sciences, Purdue University, Lafayette, Ind.
Accepted for publication April 8, 1966.
Presented to the Medicinal Chemistry Section, A.PH.A. Academy of Pharmaceutical Sciences, Dallas meeting, April 1966.
This investigation was supported by grant 5FL-GM-21,017-02 from the Institute of General Medical Sciences, U. S. Public Health Service, Bethesda, Md.
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blood levels, blood clearance, and excretion rates. Heimlich *et al.* (10, 11) used urinary excretion for the calculation of the half-lives and excretion constants for phenylpropanolamine hydrochloride and trimeprazine in sustained-release dosage forms, and Wagner and Nelson (12–14) used per cent absorbed *versus* time plots to elucidate kinetic models for drug absorption.

Large volume liquid scintillation counters have been employed to measure the parameters of whole body retention and excretion of selected γ-emitting radionuclides in humans (15–20). These procedures involve administering a dose of a γ-emitting test substance and determining the whole body radioactivity at zero time and at succeeding time intervals thereafter. Data of this type have been heretofore primarily expressed as whole body retention and have not been specifically correlated to urinary excretion or rate changes in blood levels for the purpose of describing the pharmacodynamics of a drug or a dosage form. For the system studied, whole body activity appears to be an effective method of picturing total compartmentalization and absorption-distribution-excretion kinetics.

In this study, a model system was devised wherein a γ-emitting radionuclide (sodium ^{131}I-orthoiodohippurate)[1] was administered intravenously and orally in various dosage forms, for the purpose of studying by whole body liquid scintillometry the parameters of absorption, excretion, and intercompartmental clearance of the radionuclide and the effect of variations in formula composition on these parameters.

The method described in this study would be applicable to the investigation of (*a*) any drug or test substance which could be labeled with a γ-emitting radionuclide to illustrate drug pharmacodynamics, (*b*) the factors affecting dosage form release mechanisms and properties *in vivo* according to dosage form design and the physicochemical properties of the radionuclide compound, (*c*) drugs which alter the sodium and potassium content of the body.

EXPERIMENTAL

General Considerations.—Since the evaluation of oral dosage forms was of primary interest in this study, the length of time over which whole body radioactivity determinations are normally made in such an investigation (12 hr. or longer) provides certain requirements of the γ-emitting test substance. These requirements may be listed as follows. (*a*) The test substance must not be concentrated or bound in specific tissues of the body such as the thyroid gland or liver. (*b*) The test substance should be completely eliminated by the kidneys. (*c*) The excretion rate must be rapid enough to clear a major portion of the material from the body in 3 or 4 hr. or less. (*d*) The material should not be degraded in the body to release a radioactive metabolite which would confuse interpretation of the data.

Instrumental Methods.—Whole body activity was determined in the 4 π large volume liquid scintillation counter at Purdue University (21).[2] The instrument was calibrated to differentially count the γ-emissions of ^{131}-iodine having an energy of 0.364 mev. Dosages were adjusted to provide count rates of 150,000 counts per minute or less in order to avoid having to make data corrections for coincidence loss which were found to become significant at about 250,000 counts per minute.

The *in vitro* evaluation of the release of the test material from sustained and delayed-release oral dosage forms, and the evaluation of whole blood disappearance of the test substance, was conducted in a 2-in. Harshaw[3] NaI(Tl) well crystal and appropriate electronics, calibrated to differentially count the γ-emissions of ^{131}-iodine having an energy of 0.364 mev.

In vitro release tests were conducted on a rotating bottle apparatus, as described by Souder and Ellenbogen (9), at 36–38°, and at a speed of 40–45 r.p.m.

Animal Methodology.—Seven adult dogs were utilized in this study. The dogs averaged from 8–10 Kg. in weight, and from 2 to an estimated 8 years of age. The dogs were from two groups of animals, the first group consisting of three mixed breed female animals, ranging in estimated age from 2–8 years of age, and the second group consisting of four female AKC registered beagles ranging from 2–2.5 years of age. All animals were immunized for rabies, distemper, and hepatitis, and were wormed. A maximum of four animals were used in any single experiment. On the day before an experiment was conducted, each participating animal was removed from its run and was placed in a metabolism cage. All food and water were withheld for a period of 12 hr. prior to initiation of each experimental procedure.

Catheterization of the female animals to facilitate repeated bladder emptying immediately prior to each whole body radioactivity determination was accomplished using a No. 14 French scale Bardex[4] indwelling catheter, which was fixed in place immediately prior to dosing and removed immediately after the experimental period. To prevent removal of the catheter by the animal, partial restriction was necessary. To accomplish this restriction, each animal was fitted with a "girdle" formed from pieces of polyethylene sheet 1/8 in. thick, 7–8 in. wide, and as long as necessary to completely surround the thoracic and abdominal areas. The girdle was secured by a leather strip which passed in front of the forelimbs and across the chest.

The animals were dosed immediately after bladder drainage. Immediately prior to the determination of whole body activity at each subsequent interval, the bladder was drained, and rinsed with 50 ml.

[1] Hippuran-^{131}I, Volk Radiochemical Co., Chicago, Ill.

[2] The Sinco-P was converted from 2 to 4 π geometry prior to the initiation of this experimentation.

[3] Harshaw Chemical Co., Cleveland, Ohio.

[4] Davol Rubber Co., Providence, R. I.

of sterile sodium chloride injection U.S.P. The urine so collected, the rinse, and washings from the collection graduate were placed in 1-L. polyethylene bottles,[5] brought to 1-L. volume with water, and placed in the geometrical center of the whole body scintillation counter for determination of cumulative excretion values.

So that the animals would not move about unduly to produce geometry errors while in the whole body counter, they were placed in a cylindrical fiber drum[6] of 8-gal. capacity. All counts collected on a single animal at each time interval were averaged and corrected for background, decay, and variation of counter efficiency. Each urine sample was similarly treated, with the exception that decay corrections were unnecessary since the activity of a urine phantom containing an amount of test substance identical to that administered the animals was determined each time a urine sample was counted.

A time span of not less than 4 days was allowed between subsequent uses of an individual animal.

Mass Absorption Effects.—In the initial experimentation it was necessary to determine the effects of the variability of whole body mass absorption, caused by variations in geometry and body mass surrounding the gastrointestinal tract of the animal on the reproducibility of whole body count rates as measured in the Sinco-P. An insoluble sealed source of ^{131}-iodine was prepared by imbedding an ^{131}I Radiocap[7] in an epoxy resin polymer.[8] Each sealed source was tested *in vitro* in simulated gastric and intestinal juices for absence of leach-out prior to animal dosing. Periodic determinations of whole body radioactivity were made over a 24 hr. period in the whole body counter for the animals administered the sealed sources. In all cases the sealed ^{131}I source was recovered intact from the fecal material, and the urine and feces were monitored and found to be free of radioactivity.

Dosage Regimen.—The animals used were fasted, restricted, and catheterized as described earlier. After bladder drainage and rinsing, the following dosage regimen was followed. (*a*) A dose of 1.0 ml. of a sterile aqueous solution of sodium ^{131}I-*o*-iodohippurate containing 0.14 μc./ml. was injected into each of four animals *via* the cephalic vein. (*b*) A solution containing 10 mg. of nonlabeled sodium-*o*-iodohippurate[9] and approximately 0.1 μc. of sodium ^{131}I-*o*-iodohippurate was administered orally to each of four animals. In regimens 1 and 2 whole body radioactivity was determined at zero time and at scheduled intervals thereafter. Values for whole body retention and cumulative per cent excretion were plotted as a function of time. (*c*) A solution containing 10 mg. of nonlabeled sodium-*o*-iodohippurate, and from 15–22 μc. sodium ^{131}I-*o*-iodohippurate was administered orally to four animals. At scheduled intervals following this oral administration, 1 ml. of blood was removed from the cephalic vein and analyzed for per cent of administered dose per milliliter in the Harshaw NaI(Tl) well crystal described earlier. (*d*) Ten milligrams of nonlabeled sodium *o*-iodohippurate

[5] Plax Corp., Hartford, Conn.
[6] Continental Can Co., Inc., New York, N. Y.
[7] Abbott Laboratories, Oak Ridge, Tenn.
[8] Armstrong Products Co., Inc., Warsaw, Ind.
[9] Mallinckrodt Chemical Works, St. Louis, Mo.

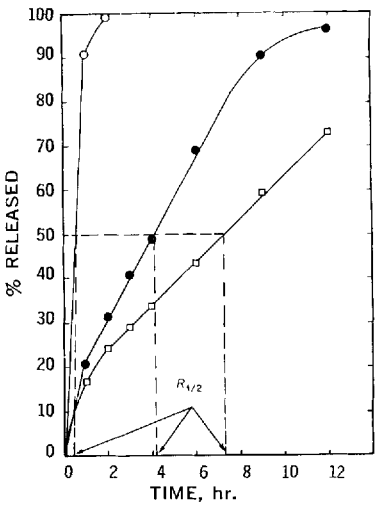


Fig. 1.—*In vitro* release, release half-times, and rate constants of sodium ^{131}I-*o*-iodohippurate contained in sustained-release oral dosage forms. Key: □, 50% polymer, $R_{1/2}$ 7.3 hr., k_0 0.082%/min.; ●, 30% polymer, $R_{1/2}$ 4.2 hr., k_0 0.168%/min.; ○, 10% polymer.

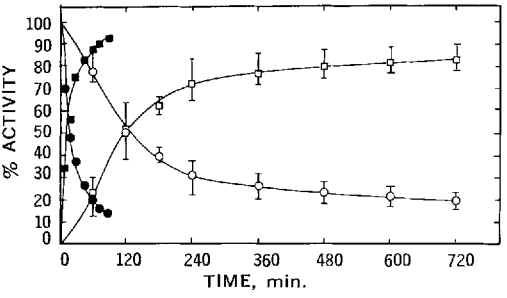


Fig. 2.—Comparison of per cent whole body retention and cumulative per cent excretion for sodium ^{131}I-*o*-iodohippurate administered orally and intravenously in aqueous solution. Key: ○, oral whole body retention; □, oral cumulative excretion; ●, intravenous whole body retention; ■, intravenous cumulative excretion.

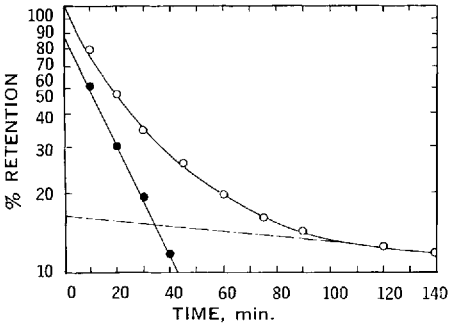


Fig. 3.—Whole body retention of sodium ^{131}I-*o*-iodohippurate administered intravenously in aqueous solution. Key: ○, whole body retention; ●, fast component; — —, slow component.

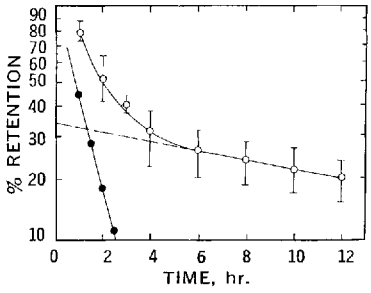


Fig. 4.—Whole body retention of sodium ^{131}I-*o*-iodohippurate administered orally in aqueous solution. Key: O, whole body retention; ●, fast component; — —, slow component.

and approximately 0.1 μc. sodium ^{131}I-*o*-iodohippurate were administered to four animals in each of two sustained-release dosage forms which exhibited zero-order release kinetics *in vitro*. (*e*) Ten milligrams of nonlabeled sodium *o*-iodohippurate and approximately 0.1 μc. of sodium ^{131}I-*o*-iodohippurate was administered to four animals in an enteric coated delayed-release dosage form.

Preparation of the Solid Dosage Form.—Sustained-release oral dosage forms of sodium ^{131}I-*o*-iodohippurate containing 0.1 μc. of ^{131}I per tablet were prepared. Three groups in all, each weighing 400 mg., were prepared containing 10, 30, and 50%, respectively, of carboxypolymethylene polymer.[10]

Care was taken to compress the tablets to the same thickness and hardness.

The *in vitro* release characteristics of these three formulas are shown in Fig. 1, with the release half times ($R_{1/2}$) and zero-order rate constants (k_0) for the 30 and the 50% formulas.

Four-hundred-milligram tablets, each containing 10 mg. of nonlabeled sodium *o*-iodohippurate and approximately 0.1 μc. of sodium ^{131}I-*o*-iodohippurate were prepared and coated with a mixture of cellulose acetate phthalate,[11] diethylphthalate,[11] and acetone. These tablets were designed to release their active principle in a delayed manner in the weakly acid or alkaline reaction of the proximal small intestine, but not in the strongly acid stomach.

When tested *in vitro* on the rotating bottle apparatus at 36–38°, the tablets prepared in this manner were seen to release less than 5% of the radioactive principle after 2.5-hr. exposure to simulated gastric juice U.S.P. without pepsin, and 100% of the radioactive principle within 15 min. after exposure to simulated intestinal fluid U.S.P. without pancreatin.

[10] Carbopol 934, B. F. Goodrich Co., Cleveland, Ohio.
[11] Eastman Organic Chemicals, Rochester, N. Y.

RESULTS AND ANALYSIS

Statistical evaluation of the data accumulated on corrected whole body activity determined intermittently during 24 hr. following administration of a sealed source of iodine-^{131}I showed the individual determinations to be within two standard deviations of the mean. These limits represent an average of plus or minus 5.45% of the mean, indicating minimal influence of mass absorption.

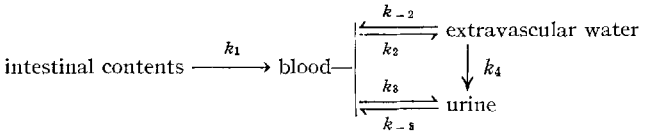
The average results of whole body retention, and cumulative excretion, of ^{131}I-*o*-iodohippurate after intravenous and oral administration in aqueous solution, are compared in Fig. 2 and plots of log per cent whole body retention as a function of time are presented and resolved into apparent first-order processes by the method of Sapirstein *et al.* (22) and Tauxe *et al.* (26) as shown in Figs. 3 and 4.

A simplified compartmental analysis of sodium ^{131}I-*o*-iodohippurate after oral administration is shown in Scheme I.

In Fig. 3, the plot for intravenous administration, the fast component A (Table I), is considered to be k_3, and the slow component B, k', a combination of k^{-2} and k_4 (the constant of tubular secretion). Figure 4 for oral administration is analyzed in the same way, designating the fast component A, k_3, and the slow component B, K' a combination of k'_{-2} and k_4.

Table I compares the apparent first-order rate constants and biological half-lives (T_B) derived from the linear components in Figs. 3 and 4.

Figure 5 shows the correlation of observed and theoretical whole body retention of sodium ^{131}I-*o*-



Scheme I

iodohippurate administered intravenously and orally in aqueous solution. The theoretical whole body retention is derived by subtracting the cumulative per cent excreted from 100%. If the theoretical per cent whole body retention perfectly matched the observed per cent whole body retention, the straight line shown in Fig. 5 would connect such points in complete agreement. The extent of departure of the points in Fig. 5 is a reflection of the departure of the observed data from the theoretical data. The data for intravenous administration can be seen to depart somewhat from complete agreement, while the data of oral administration exhibit excellent correlation.

Figure 6 illustrates the cumulative excretion, theoretical whole body retention, and whole blood disappearance following oral administration of sodium ^{131}I-*o*-iodohippurate in aqueous solution. The data can be seen to be extrapolated to the 12-hr. value for use in Fig. 7. Figure 7 is a plot of the log per cent whole body retention, and log of the per cent of the dose in the blood as a function of time. The rate constants derived from the linear components of this figure are shown in Table II. The fast component of the theoretical whole body retention curve is represented by k_3 and the slow

TABLE I.—APPARENT FIRST-ORDER RATE CONSTANTS AND BIOLOGICAL HALF-LIVES OF THE LINEAR COMPONENTS OF FIGS. 3 AND 4

Parameter	Intravenous Components A	Intravenous Components B	Oral Components A	Oral Components B
k_3	0.048 min.$^{-1}$	...	0.014 min.$^{-1}$	...
K'^a	...	0.0026 min.$^{-1}$	...	0.0011 min.$^{-1}$
T_B	14.4 min.	4.43 hr.	49.5 min.	10.5 hr.

[a] $K' = k_{-2} + k_4$.

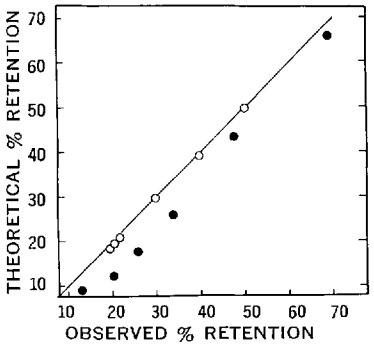


Fig. 5.—Correlation of observed and theoretical whole body retention of sodium ^{131}I-*o*-iodohippurate administered orally and intravenously in aqueous solution. Key: ○, oral; ●, intravenous.

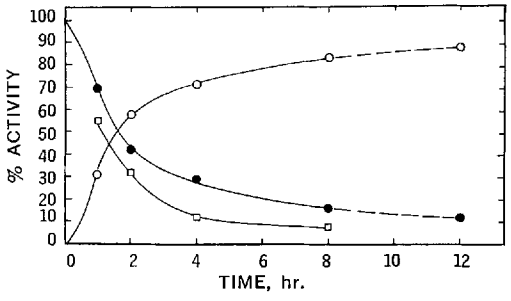


Fig. 6.—Cumulative excretion, theoretical whole body retention, and whole blood disappearance following oral administration of sodium ^{131}I-*o*-iodohippurate in aqueous solution. Key: ○, cumulative excretion; ●, theoretical retention; □, amount of dose in blood (%/ml. × 10^4).

component, $K'(k_{-2} + k_4)$. The fast component of the whole blood disappearance curve is felt to represent a macroscopic rate expressed by the constants $k_2 + k_3$. The slow component is represented by the constant, k_{-2}.

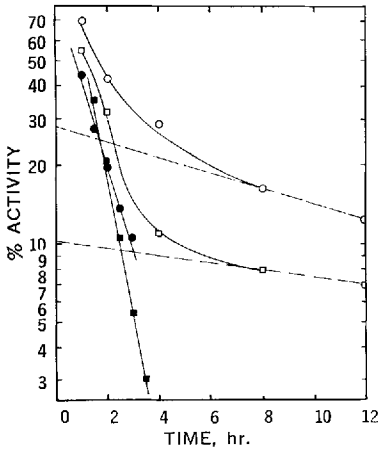


Fig. 7.—Theoretical whole body retention and whole blood disappearance of sodium ^{131}I-*o*-iodohippurate following oral administration in aqueous solution. Key: ○, whole body retention; ●, fast component; — —, slow component; □, amount of dose in blood (%/ml. × 10^4); ■, fast component; ——, slow component.

Figure 8 shows the correlation of whole body retention and whole blood disappearance. The deviation from perfect correlation noted in this figure is felt to be a reflection of the fact that whole blood disappearance is faster than excretion.

Figure 9 is a comparison of the whole body retention *versus* time plots from sodium ^{131}I-*o*-iodohippurate contained in the two sustained-release tablet formulations which exhibited two different zero-order release rates *in vitro* and the curve for sodium ^{131}I-*o*-iodohippurate in aqueous solution, and Table III presents the rate constants and half times for these systems, compared to those derived from Fig. 1. Figure 10 shows the comparison of the whole body retention *versus* time plots for sodium ^{131}I-*o*-iodohippurate contained in an enteric coated delayed-release tablet.

TABLE II.—COMPARISON OF APPARENT FIRST-ORDER RATE CONSTANTS OF WHOLE BLOOD DISAPPEARANCE AND WHOLE BODY RETENTION OF SODIUM ^{131}I-*o*-IODOHIPPURATE ADMINISTERED ORALLY IN AQUEOUS SOLUTION

	k_3	k_{-2}	K'^a	$k_2 + k_3$
Whole blood disappearance	...	0.0072 min.$^{-1}$		0.0204 min.$^{-1}$
Theoretical whole body retention	0.0120 min.$^{-1}$		0.00112 min.$^{-1}$	...
Biological half-life	58 min.	96 min.	12.5 hr.	30 min.

[a] $K' = k_{-2} + k_4$.

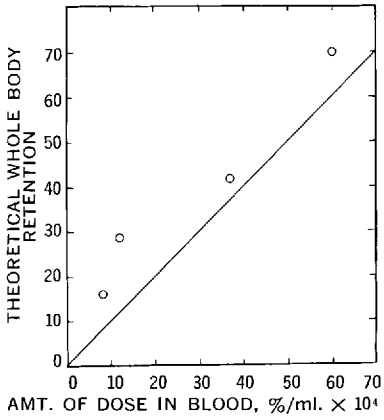


Fig. 8.—Correlation of theoretical whole body retention with the per cent of dose per milliliter of blood for sodium ^{131}I-*o*-iodohippurate administered orally in aqueous solution.

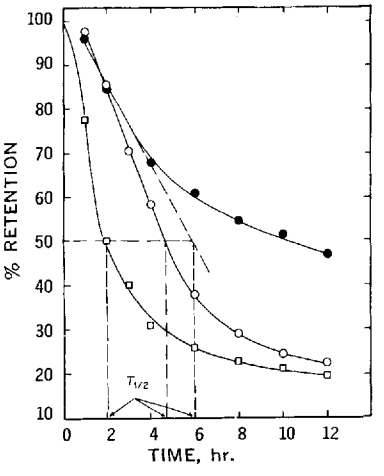


Fig. 9.—*In vivo* comparison of sodium ^{131}I-*o*-iodohippurate administered orally in aqueous and in two sustained-release dosage forms. Key: ○, 30% carboxypolymethylene polymer; ●, 50% carboxypolymethylene polymer; □, aqueous solution.

DISCUSSION

Sodium ^{131}I-*o*-iodohippurate was found to satisfactorily meet most of the requirements for a γ-emitting test substance outlined earlier. It does not concentrate in the thyroid gland or liver. It is at least 90% excreted by the kidneys.

Mathematical treatments of the excretion of substances excreted both by glomerular filtration and tubular secretion are given by Sapirstein *et al.* (22) for creatinine and by Conn *et al.* for sodium-*o*-iodohippurate.

Benzoic acid is detoxified in man by conjugation with glycine to form hippuric acid and by conjugation with glucuronic acid to form benzoyl glucuronide. Analogously, ^{131}I-orthoiodobenzoic acid forms the similar corresponding metabolites, one of which is ^{131}I-*o*-iodohippuric acid (23). Thus, it was felt that if a test substance were supplied to the body as a metabolite, degradation of the test substance would be reduced and interpretation of accumulated data would be simplified.

TABLE III.—COMPARISON OF THE *In Vivo* AND *In Vitro* ZERO-ORDER RATE CONSTANTS, RELEASE HALF TIME, AND BIOLOGICAL HALF-LIVES OF SODIUM ^{131}I-*o*-IODOHIPPURATE IN SUSTAINED-RELEASE DOSAGE FORMS

	30% Carboxypolymethylene Polymer	50% Carboxypolymethylene Polymer
In vitro k_0	0.168%/min.	0.082%/min.
In vivo k_0	0.227%/min.	0.147%/min.
Apparent $T_{1/2}$[a]	4.65 hr.	6.0 hr.
T_B[b]	3.60 hr.	5.7 hr.
$R_{1/2}$[c]	4.20 hr.	7.3 hr.

[a] Determined from 50% whole body retention values in Fig. 9. [b] Calculated from the expression for the half life of a zero-order process. $T_{1/2} = 1/2\ Co/k_0$. [c] Determined from 50% released values in Fig. 1.

It is possible that when administered intravenously, a certain amount of sodium ^{131}I-*o*-iodobenzoylglycine may be converted to the glucuronide, and further it is possible that on oral administration the iodobenzoic acid moiety may be liberated from its glycine conjugate by the effect of intestinal enzymes, then absorbed and detoxified by conjugation with glucuronic acid or glycine.

Regardless of the metabolic pathway followed by sodium ^{131}I-*o*-iodohippurate on oral administration, it has been observed that reproducible values for the fast and slow components of a plot of log per cent whole body retention as a function of time are readily obtainable.

It will be noted from Table II that the rate constant (k_3) for the fast component upon intravenous administration of sodium ^{131}I-*o*-iodohippurate was 0.048 min.$^{-1}$, while the k_3 value for sodium ^{131}I-*o*-iodohippurate administered orally in aqueous solution was 0.0143 min.$^{-1}$. If the k_3 obtained after intravenous administration is considered to be the renal excretion rate constant, then the k_3 obtained after oral administration, which is approximately one-third of the value of the intravenous k_3, must be the rate constant for some rate-limiting process in the passage of sodium ^{131}I-*o*-iodohippurate from the gastrointestinal tract to the urine. This rate-limiting step is hypothesized to be the intestinal absorption rate of sodium ^{131}I-orthoiodohippurate. This hypothesis is supported, but not confirmed, by the fact that the whole body clearance becomes a zero-order process when sodium ^{131}I-*o*-iodohippurate is administered orally in a sustained-release dosage form which exhibits zero-order release kinetics on *in vitro* testing.

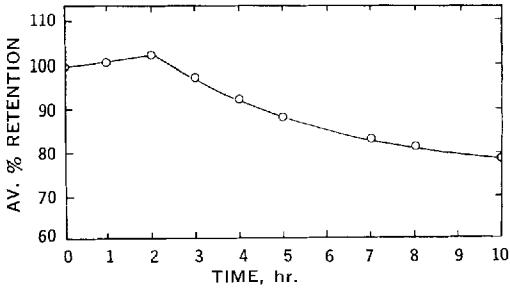


Fig. 10.—*In vivo* whole body retention of sodium ^{131}I-*o*-iodohippurate administered orally in a delayed-release dosage form.

It will be noted from Fig. 9 that contrary to the characteristics observed for the sustained-action dosage forms *in vitro* in Fig. 1, the zero-order elimination begins to deviate from linearity at 4–5 hr. after dosage. This deviation may be due to decreased biological availability of sodium ^{131}I-*o*-iodohippurate. This may occur due to enzymatic degradation of the compound to form one which is not readily absorbed, due to liberation of free ^{131}-iodine which would concentrate in the thyroid gland, or more likely may occur due to the pH of the environment being such that the compound is ionized and thus not readily absorbable. This phenomenon may also be observed from Fig. 10 where only about 22% of the administered dose was excreted over a 10-hr. period.

It has been demonstrated that the property of biological availability of a test substance, a parameter most important in dosage design and formulation, can be readily evaluated by the whole body monitoring of the γ-emissions of the test substance. An insight into *in vivo* release rates from sustained and delayed-release oral dosage forms is gained along with the effect of these dosage forms on the biological availability of the test substance in the body. Furthermore, specific information can be gained on the effect of the dosage form on absorption, excretion, and distribution of the test substance in the body. In addition, in the test substance under study, whole body retention of the radionuclide could be correlated very well to the excretion rate of the drug orally administered and fairly well to drug intravenously administered. The whole blood disappearance rate of the radionuclide could also be correlated to whole body retention as determined by the whole body scintillometry method described.

The added advantage of simplicity of animal and sample handling is also evident. Since urine samples in the larger animals must be accumulated on the short term basis by catheterization, the excretion of the test substance can be evaluated after urine drainage in as little as 3 min. without the necessity for further urinalysis. This is accomplished by determining the per cent whole body retention in a large volume liquid scintillation detector. Sinco-P, the whole body scintillation counter at Purdue University, was used in this study, but smaller and less expensive units are available which will readily accommodate a dog the size of a 20-lb. beagle (24, 25).

SUMMARY AND CONCLUSIONS

The use of the large volume liquid scintillation counter at Purdue University was investigated for its applicability to the evaluation of the pharmacodynamic properties of selected drugs and oral dosage forms. Use of data accumulated on the whole body retention of ^{131}I-orthoiodohippurate sodium was illustrated. Semilog plots of per cent whole body retention as a function of time were constructed and analyzed on the basis of first-order kinetics. Data obtained from *in vivo* analysis were compared with *in vitro* data for sodium ^{131}I-*o*-iodohippurate in sustained and delayed-release oral dosage forms. These studies indicated the following.

1. A large volume liquid scintillation counter is useful in obtaining information on the pharmacodynamic properties of a test substance in oral dosage form.

2. Whole body retention data may be readily correlated to cumulative excretion rates and blood disappearance rates.

3. Meaningful rate constants are obtainable from semilogarithmic plots of whole body retention as a function of time.

4. The biological availability of a test substance may be readily determined from whole body retention data.

5. Rates of release of a test substance in sustained or delayed-release dosage forms may be obtained *in vivo* without the necessity of blood and urine analysis.

6. Sodium ^{131}I-*o*-iodohippurate was found to be a suitable γ-emitting radioactive test substance.

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