Proceedings
of the
Society
for
Experimental Biology and Medicine

VOLUME 60 NOVEMBER, 1945 NUMBER 2

SECTION MEETINGS

IOWA
State University of Iowa October 23, 1945
PACIFIC COAST
Stanford University Medical School October 24, 1945

15130

Calculation of the Rate of Absorption of Exogenous Creatinine.

RAFAEL DOMINGUEZ AND ELIZABETH POMERENE.
From the Division of Laboratories and Research, St. Luke's Hospital, Cleveland, Ohio.

The purpose of this paper is to calculate the rate of absorption of a substance from the changes in the concentration of the substance in the blood.

Because the rate of absorption is in a large measure independent of the chemical changes which the substance may undergo after absorption, we shall study the absorption of a substance which is not metabolized in the body—creatinine. (To avoid repetition, the word creatinine will be used to mean exogenous creatinine, unless otherwise stipulated). The conclusion that creatinine is not metabolized is based on previous work showing that (1) after intravenous injection, about 96% of creatinine is recovered in the urine in the first 24 hours and (2) after oral administration to a patient with an ileal fistula, the amount of creatinine recovered in the urine was about equal to the difference between the quantity given and the amount collected from the fistula.[1] If this conclusion is valid, it follows that the incomplete recovery of creatinine in oral experiments (a fact ascertained by all investigators) is due to incomplete absorption from the intestinal tract. In other words, the amount of creatinine recovered after ingestion is actually the amount absorbed.

After ingestion, the concentration of creatinine in the plasma rises to a maximum and then falls. The rate of this change in concentration depends, in general, on three other rates: the rate of absorption from the intestine, the rate of excretion by the kidney, and the rate of diffusion from the plasma into the other body fluids. If the body fluids are in diffusion equilibrium during absorption, the rate of diffusion may be eliminated from further consideration. Because of this simplification we shall limit this paper to experiments in which the assumption of diffusion equilibrium can be justified. The method of calculation when the rate of diffusion must be considered will be presented in another paper.

In the first few minutes of absorption the concentration of creatinine in some parts of the portal system is undoubtedly higher than

[1] Dominguez, R., and Pomerene, E., PROC. SOC. EXP. BIOL. AND MED., 1945, **58**, 26.

in others and especially higher than in the systemic veins. This initial difference, however, becomes negligible after a short time because of the circulation of the blood and because of the dilution of the blood from the portal system with the larger volume of blood from the venæ cavæ. Inasmuch as the absorption of creatinine usually lasts several hours, we shall neglect the transient initial inequalities in concentration.

With the foregoing considerations in view, the absorption of creatinine may be formulated as follows: Let q be the amount of creatinine which is present in the body at a given time, an amount which, by assumption, is evenly distributed in the body fluids. The rate of change in the amount of creatinine q is equal to the difference between the rate of absorption, ρ, and the rate of excretion, η, that is,

$$\frac{dq}{dt} = \rho - \eta \qquad (1)$$

The amount q is equal to the product of creatinine concentration in the plasma, ξ, and the volume of distribution, V. Since in the conditions of the experiment the volume V remains constant, equation 1 becomes

$$V\frac{d\xi}{dt} = \rho - \eta \qquad (2)$$

or, after transposition,

$$\rho = V\frac{d\xi}{dt} + \eta \qquad (3)$$

Equation 3 means that, at any given time, the rate of absorption is equal to the product of the volume of distribution and the rate of change of the plasma concentration, plus the rate of excretion.

The rate of excretion, η, is, on an average, proportional to the plasma concentration, ξ, that is,

$$\eta = A\xi \qquad (4)$$

where A is a constant.[2]

With this value of η, equation 3 becomes

$$\rho = V\frac{d\xi}{dt} + A\xi \qquad (5)$$

After absorption ends (that is, $\rho = 0$ in equation 5), the creatinine concentration in the plasma falls according to the equation[3]

$$\xi = a\,e^{-\alpha t} \qquad (6)$$

where a is a constant, e is the base of natural logarithms and α represents the relation

$$\alpha = \frac{A}{V} \qquad (7)$$

Equation 6 has been experimentally verified.[2]

The calculation of the rate of absorption ρ (equation 5), consists of the following steps. (1) The rate of excretion is plotted *vs.* the plasma concentration. The slope of the line is the constant A. (2) The logarithms of the plasma creatinine concentrations are plotted *vs.* time and the constants of equation 6 are computed. The slope of the linear descending part of the graph is the constant α. The plasma data can be increased in number by dividing the rates of excretion of creatinine by the constant A and treating these data as plasma data. For this step, the endogenous blanks are subtracted from both the plasma concentration of total creatinine and the rate of excretion of total creatinine. (3) A smooth curve is drawn through or fitted to the early plasma creatinine data and is joined smoothly to the exponential curve $a\,e^{-\alpha t}$ of step 2. This curve should begin at zero with the slope zero because, at zero time, both the rate of absorption and the rate of excretion are zero. Hence, by equation 3, the slope $d\xi/dt$ of the plasma curve is zero at zero time. (4) A sufficient number of values of ξ and $d\xi/dt$ are calculated from the fitted curve. (5) the volume V is calculated by equation 7. (6) With these values, the operations indicated in the right hand side of equation 5 are performed. The curve of the rate of absorption is then plotted *vs.* time.[4]

[2] Dominguez, R., and Pomerene, E., *J. Biol. Chem.*, 1934, **104**, 449.

[3] Dominguez, R., PROC. SOC. EXP. BIOL. AND MED., 1934, **31**, 1146.

[4] For further details the following books may be consulted: Lipka, J., *Graphical and Mechanical*

TABLE I.

Protocol of Experiments on Subject G.N.B.

Breakfast at 7:45 a.m.; creatinine, 8 g in water, taken by mouth at 9:17 a.m.; lunch at 11:50 a.m.; dinner at 5:00 p.m.

Urine			Blood	
Interval of collection	Vol., cc	Creatinine conc. mg/100 cc	Time	Creatinine conc., mg/100 cc of oxalated plasma
9:09 a.m.-10:27 a.m.	79	671	10:22 a.m.	7.8
10:27 " -11:29 "	70	1669	11:24 "	11.9
11:29 " -12:35 p.m.	71	1744	12:29 p.m.	9.1
12:35 p.m.- 1.37 "	43	1745	1:31 "	6.3
1:37 " - 2:40 "	73	869	2:34 "	4.8
2:40 " - 4:14 "	308	213	4:04 "	3.3
4:14 " - 5:55 "	76.5	807	5:51 "	3.0
5:55 " - 7:09 "	60	561	7:04 "	2.8
7:09 " - 9:20 a.m. the following morning	1113	184		
9:20 a.m.-10:26 a.m.	22.5	316		

Ileal fistula		
Interval of collection	Vol. cc	Conc. of chromogenic substances mg creatinine per 100 cc
9:10 a.m.-10:29 a.m.	318	4.2
10:29 " -11:30 "	185	34.2
11:30 " -12:36 p.m.	132.5	1013
12:36 " - 1:38 "	137	160
1:38 " - 2:41 "	310	94
2:41 " - 4:10 "	300	12.5
4:10 " - 5:56 "	298	11.3
5:56 " - 7:10 "	220	6.8

The amount absorbed can be obtained by calculating the area under the graph of the rate of absorption. The amount excreted can be determined, first, by calculating the area under the plasma creatinine curve and then by multiplying the area thus found by the constant A. If the excretion data are regular and the constant A applies to the whole curve, the calculated amount excreted will be close to the amount actually excreted. If the excretion data are irregular and the constant A is not well determined, an adequate value of A can be obtained by dividing the amount actually found by the area under the plasma creatinine curve. This step follows from the integration of equation 4.

Computation, John Wiley and Sons, Inc., New York, 1918; Scarborough, J. B., *Numerical Mathematical Analysis*, The Johns Hopkins Press, Baltimore, 1930; Sherwood, J. K., and Reed, C. E., *Applied Mathematics in Chemical Engineering*, McGraw-Hill Book Company, Inc., New York, 1939.

Since the calculated amount absorbed is equal to the calculated amount excreted, the equality of the calculated amounts serves as a check of the numerical work.

With respect to diffusion equilibrium, it follows from the meaning of the exponent a (equation 7) that if A and a are constant, the volume of distribution V is also constant. This constant value of V holds for the descending part of the plasma creatinine curve, the part from which a is calculated (equation 6). A subsidiary calculation is necessary to test whether the same value of V holds also for the first part of the plasma creatinine curve. If it does, the assumption of diffusion equilibrium holds.

All these steps will be illustrated by several examples.

Subject G.N.B. White male, age 24, weight 54 kg, height 179 cm. This man had a permanent ileostomy for a chronic ulcerative colitis.[1] The protocol of the experiment is

presented in Table I. All the analyses were made according to the methods of Folin[5] and Folin and Wu.[6] The average endogenous creatinine in the plasma was 0.74 mg per 100 cc and the average rate of excretion of endogenous creatinine was 1.0 mg per min. The relation of the rate of excretion of total creatinine to the total plasma creatinine after the maximum concentration is shown in Fig. 1. By

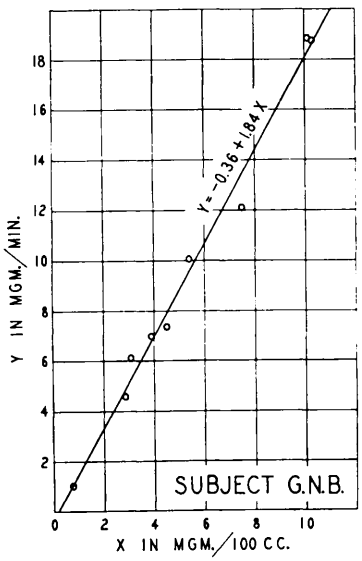

FIG. 1.

Relation between the rate of excretion of total creatinine y and the plasma concentration of total creatinine x from the data of Subject G.N.B. after the maximum creatinine in the plasma (Table I).

the method of averages, the slope of the line, constant A, is 1.841. After subtraction of the endogenous blanks, the rates of excretion were multiplied by 1/1.841 and all the data were treated as plasma creatinine. The plot is shown in Fig. 2. A smooth curve was drawn through the points from 0 to 4.2 hr. The line $ln\ \xi = ln\ 17.89 - 0.259\ t$ was fitted to the points from 4.2 to 9.25 hr by the method of averages. The whole curve can be seen in Fig. 2. From a magnified plot of this curve, values and differences of the concentration ξ were tabulated from 0 to 2.5 hr at intervals of 0.1 hr and from this table the values of the

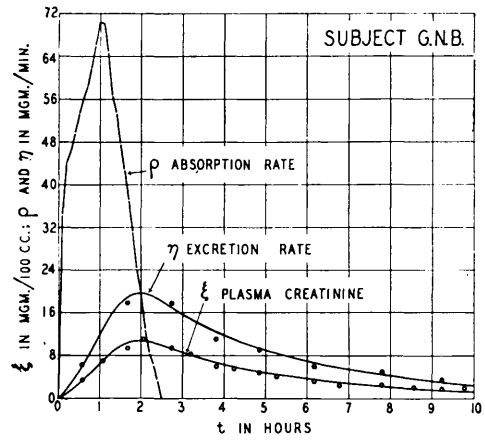

FIG. 2.

Curves of the plasma creatinine ξ, of the rate of excretion η, and of the rate of absorption ρ from the data of Subject G.N.B. (Table I). The closed circles represent the average rate of creatinine excretion at the middle of the intervals of collection and the open circles represent the concentrations of plasma creatinine.

derivative $d\xi/dt$ were computed. The volume V, corrected for units, is $V = \frac{1.841}{0.259} \times 60 \times 10^2$ cc $= 42.6 \times 10^3$ cc. The rate of absorption was then calculated by equation 5 and plotted (Fig. 2). The irregularities in the graph are undoubtedly due to uncertainties in the tabulated values of ξ.

The test for diffusion equilibrium is as follows. The term-by-term integration of equation 5, from 0 to t_1, is

$$P_1 = V\xi_1 + H_1$$
$$(\rho_1 = V\xi_1 + \eta_1)$$

Where P_1 is the amount absorbed in the time t_1 and H_1 is the amount excreted in the same time. This equation simply means that the amount absorbed in a given interval (P_1) is equal to the amount present in the body fluids at the end of this interval ($V\xi_1$) plus the amount excreted in the same interval (H_1). The calculation is carried out at cumulative intervals as shown in Table II. It is seen in this table that the cumulative amounts absorbed have the mean value 5.92 g from 2.2 to 9.87 hr and that the variations in the calculated amount absorbed lie within 2.2% of the mean. The only constant quantity in this calculation is the volume V, a volume which was

[5] Folin, O., *J. Biol. Chem.*, 1914, **17**, 469.

[6] Folin, O., and Wu, H., *J. Biol. Chem.*, 1919, **38**, 81.

TABLE II.
Cumulative Amount of Creatinine Absorbed by Subject G.N.B.

(1) Time t hr	(2) Plasma creatinine (from graph) ξ mg/100 cc	(3) Volume of distribution V liters	(4) Amt of creatinine present in body fluids at time t $V\xi$ g	(5) Amt excreted in time t (observed) g	(6) Amt absorbed in time t (4) + (5) g
1.17	7.8	42.6	3.32	0.44	3.76
2.20	10.5		4.47	1.55	6.02
3.30	7.8		3.32	2.72	6.04
4.33	5.8		2.47	3.41	5.88
5.38	4.4		1.87	3.98	5.85
6.95	3.0		1.28	4.54	5.82
8.63	1.9		.81	5.06	5.87
9.87	1.4		.60	5.32	5.92

The mean of the last 7 figures of column 6 is 5.91 g.

obtained from the part of the plasma data in which equilibrium can be assumed. Since the use of this value of V in the first part of the experiment does not lead to any internal inconsistency, the assumption of diffusion equilibrium is justified.

The amount absorbed, calculated by Simpson's rule, is 6.13 g. The amount excreted, calculated by numerical integration from 0 to 4.2 hr and by straight integration from 4.2 to 25 hr, is 5.93 g. The amount actually excreted from 0 to 25 hours, from the data of Table I, is 6.53 g. Inasmuch as the calculation of the rate of absorption was based on the rates of excretion of the first 10 hours only, some adjustment is needed to take care of the amount excreted after 10 hours. But since this adjustment does not change the shape of the early part of the plasma creatinine curve, we have preferred to present the results without adjustment. We regret that, for reasons already given,[1] this experiment could not be repeated.

It should be noticed that (1) the maximum rate of absorption (Fig. 2) corresponds to the first point of inflexion of the plasma creatinine curve and (2) at the maximum plasma creatinine, the rate of absorption is equal to the rate of excretion. At the time of the maximum plasma creatinine, however, 5.89 g has already been absorbed.

In this subject we have 3 estimates of the amount absorbed: (1) by the difference between the quantity ingested and the amount collected from the fistula, 8 g — 1.96 g = 6.04

TABLE III.
Protocol of Experiment on Subject S.
Breakfast at 7:45 a.m.; creatinine, 8 g in water, taken by mouth at 9:07 a.m.; lunch at 12:30 p.m.; dinner at 5:30 p.m.

Urine			Blood	
Interval of collection	Vol., cc	Creatinine conc., mg/100 cc	Time	Creatinine conc., mg/100 cc oxalated plasma
8:05 a.m.- 9:04 a.m.	23.0	260	9:06 a.m.	.8
9:04 " - 9:39 "	61.5	368	9:41½ "	8.2
9:39 " -10:10 "	103.5	520	10:13 "	11.0
10:10 " -11:08 "	78.0	1337	11:11½ "	11.2
11:08 " -12:09 p.m.	152.0	656	12:13 p.m.	9.1
12:09 p.m.- 1.07 "	73.0	912	1:12 "	6.4
1:07 " - 2:11 "	52.5	945	2:15 "	5.7
2:11 " - 3:07 "	44.5	863	3:11 "	4.4
3:07 " - 4:09 "	53.5	652	4:11 "	3.3
4:09 " - 5:08 "	61.0	521	5:13 "	3.0
5:08 " - 9:19 a.m. the following morning	780.0	239	9:24 a.m.	.88
9:19 a.m.-10:20 a.m.	55.0	116		

g, (2) by the amount excreted in 25 hr and the assumption that ingested creatinine is not transformed in the body, 6.53 g, and (3) by the method of this paper, 6.13 g. The first estimate is independent of any observations on plasma or urine. The second estimate requires no knowledge of the rate of excretion and is of course independent of the plasma creatinine concentration. The third estimate depends only on the rates (equation 5) and is independent of the amount excreted after, say, 10 hr. These 3 estimates represent 76, 82, and 77% of the quantity ingested. With this approximation we can say that the amount excreted is equal to the amount absorbed, or, in other words, that ingested creatinine is not transformed in the body.

Subject S. White male, age 29, weight 56.2 kg, height 163.2 cm. The protocol of the experiment is given in Table III. The endogenous creatinine excretion of several control periods was, on an average, 1.0 mg per minute. The endogenous plasma creatinine was 0.8 mg/100 cc. The relation between the rate of excretion and the plasma concentration of total creatinine is seen in Fig. 3.

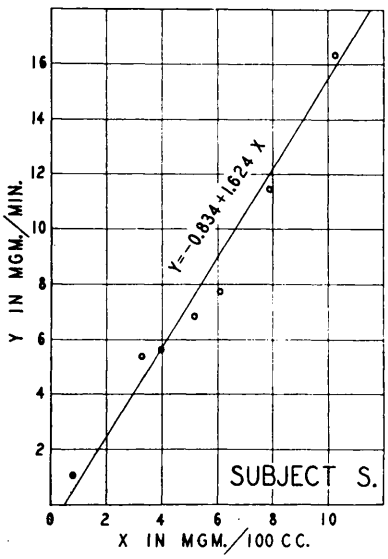

FIG. 3.

Relation between the rate of excretion of total creatinine y and the plasma concentration of total creatinine x from the data of Subject S. after the maximum creatinine in the plasma (Table III). The line with slope 1.624 (see text) passes through the centroid of the points shown in the graph.

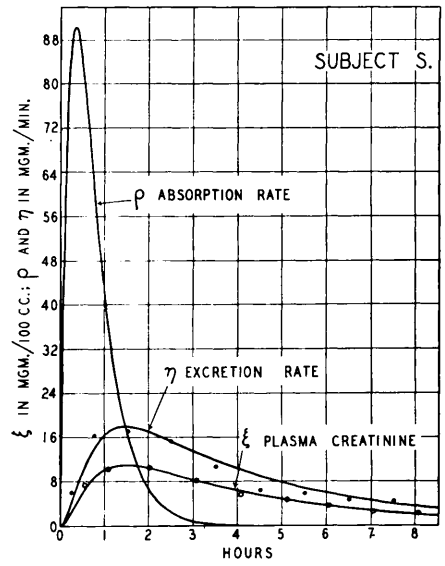

FIG. 4.

Curves of the plasma creatinine ξ (equation 8), of the rate of excretion η (equation 4), and of the rate of absorption ρ (equation 9) from the data of Subject S. (Table III). The closed circles represent the average rate of creatinine excretion at the middle of the intervals of collection and the open circles represent the concentrations of plasma creatinine. The values of the constants of the equations are shown in Table IV.

Instead of drawing a smooth curve through the points from 0 to 3 hr, as in the first example, a mathematical equation was fitted to all the points. The equation is

$$\xi = a e^{-at} - c e^{-\gamma t} + d e^{-\delta t} \qquad (8)$$

where the first exponential fits the data from 3 to 8 hr, the second fits the residuals of the first exponential and the data from 1 to 3 hr, and the third exponential forces the curve to start at zero with the slope zero.

The equations for the latter conditions are

$$a - c + d = 0$$
$$a\alpha - c\gamma + d\delta = 0$$

The constants d and δ are computed from these equations as soon as the other 4 constants are determined.

The values of the constants are given in Table IV and the curve of the plasma creatinine is reproduced in Fig. 4.

The rate of absorption ρ is derived by per-

TABLE IV.

Constants of Equations 4, 8, and 9, and Quantities of Creatinine Ingested, Excreted, and Absorbed.

Subject and exp. No.	Constants of Equation 4	Constants of Equation 8					Constants of Equation 9	Vol. of distribution, V	Quantity ingested	Amt excreted in stated time		Calculated amount absorbed,
	A	a	c	d	α	γ	δ	p			hr	g	g
	cc/min $\times 10^2$	mg/cc $\times 10^{-2}$				1/hr		mg/min	liters	g			
GNB*	1.84	20.8			0.296				42.6	8	25.2	6.52	6.13
S	1.62	19.4	38.6	19.2	0.278	2.04	3.8	396	35.0	8	25.2	5.43	5.42
E3†	1.81	6.0	6.7	0.7	0.286	1.09	8.0	33.8	38.0	3	8.3	1.41	1.64
E4	1.99	18.3	25.5	7.2	0.286	1.69	5.3	248	41.7	8	6.7	4.87	5.99
E5	1.98	10.8	15.0	4.2	0.286	1.69	5.3	145	41.5	5	10.2	3.27	3.53
E6	1.94	11.0	15.4	4.4	0.286	1.69	5.2	146	40.7	5	8.6	3.13	3.52
E8	1.67	12.0	17.4	5.4	0.286	1.64	4.6	137	35.0	5	7.2	2.73	3.26

* This subject had a permanent ileal fistula.[1] During this experiment 1.96 g of creatinine was recovered from the fistula (Table I).
† Data on Subject E from Dominguez, R., and Pomerene, E., *J. Biol. Chem.*, 1934, **104**, 449.

forming on equation 8 the operations indicated in equation 5. The result is

$$\rho = p(e^{-\gamma t} - e^{-\delta t}) \qquad (9)$$

where $p = \frac{Ac}{a}(\gamma - a) = \frac{Ad}{a}(\delta - a)$.

The value of A for the descending part of the curves is, by the method of averages, 1.49. The value of A obtained by dividing the amount actually excreted in 25.2 hr by the area under the plasma creatinine curve is $A = 5.43/3.352 = 1.62$ (Fig. 3). This result indicates that the value of A was greater than the mean before the maximum plasma creatinine and smaller than the mean after the maximum. With $A = 1.62$, the curve of the rate of excretion (equation 4) and the curve of the rate of absorption (equation 9) are reproduced in Fig. 4. The amount absorbed is

$$\int_0^\infty \rho\, dt = 60 \times 396\left(\frac{1}{2.04} - \frac{1}{3.82}\right) = 5.42 \text{ g},$$

a good check of the numerical work (Table IV).

The maximum rate of absorption can be shown to occur at the time of the first point of inflexion of the plasma creatinine curve (Fig. 4). The rate of absorption is equal to the rate of excretion at the time of the maximum plasma creatinine, but by this time 4.905 g has already been absorbed.

The test for diffusion equilibrium is presented in Table V. The calculated amount absorbed reaches the value 5.52 g, its greatest value, 3.02 hr after ingestion (Table V) and this value is within 2% of the total amount excreted in 25.2 hr (Table IV). From 3 to 8 hr the amount absorbed remains stationary (Table V), except for small variations due to irregularities in excretion. As in the preceding example, the assumption of diffusion equilibrium is justified.

Subject E. White female, age 26, weight 45 kg, height 161 cm. The data of 6 experiments on this subject have been published elsewhere,[2] but in that publication only the data of the descending part of the curves were utilized. We have found that equation 8, which fits

TABLE V.
Cumulative Amount of Creatinine Absorbed by Subject S.

(1) Time t hr	(2) Plasma creatinine (from graph) ξ mg/100 cc	(3) Vol. of distribution V liters	(4) Amt of creatinine present in body fluids at time t $V\xi$ g	(5) Amt excreted in time t (observed) g	(6) Amt absorbed in time t (4) + (5) g
.525	6.0	35.0	2.10	0.19	2.29
1.04	10.3		3.60	0.70	4.30
2.01	10.5		3.68	1.68	5.36
3.02	8.3		2.90	2.62	5.52
3.99	6.3		2.20	3.22	5.43
5.06	4.7		1.64	3.66	5.30
5.99	3.6		1.26	3.98	5.24
7.02	2.7		.94	4.27	5.22
8.01	2.1		.74	4.53	5.26

The mean value of the last 6 figures of column 6 is 5.33 g.

the data of subject S, fits also the data of subject E.

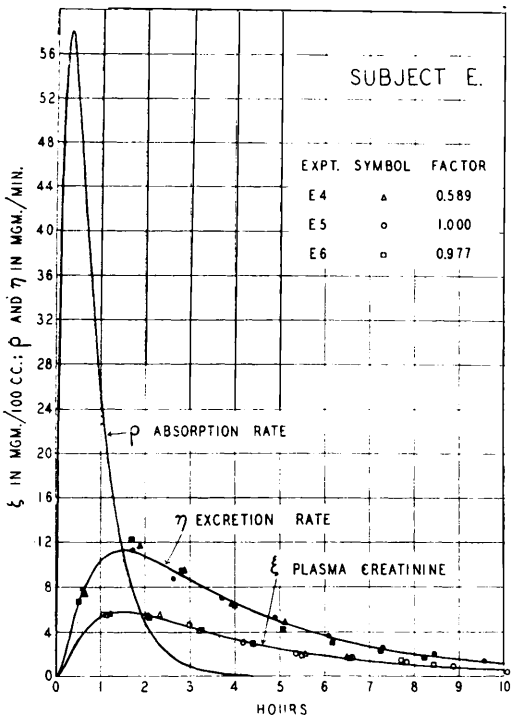

FIG. 5.

Curves of the plasma creatinine ξ (equation 8), of the rate of excretion η (equation 4), and of the rate of absorption ρ (equation 9) of Subject E. The data of three experiments have been reduced to one by means of the factors shown in this table. The values of the constants of the equations are shown in Table IV. The closed symbols represent the average rates of creatinine excretion at the middle of the intervals of collection and the open symbols represent the concentrations of plasma creatinine. (Data of Dominguez, R., and Pomerene, E., *J. Biol. Chem.*, 1934, **104**, 449.)

Of the 6 experiments on subject E, 5 form a homogeneous set with the common value $a = 0.286$. Furthermore, experiments E4, E5, and E6 can be reduced to one—as shown in Fig. 5—by means of the factors 0.589, 1, and 0.977, respectively. The other two experiments, E8 and E3, must be computed separately. The values of the constants in these five experiments, calculated by the present method, are given in Table IV. In order to make use of all the data and at the same time satisfy the amounts excreted before, as well as after the maximum plasma creatinine, a new value of A was calculated by dividing the amount actually excreted by the area under the plasma creatinine curve (Table IV).

The curves corresponding to the grouped data of experiments E4, E5, and E6 are reproduced in Fig. 5.

Discussion. The method we have described is applicable to substances which are in diffusion equilibrium during absorption, are not utilized in the body and are almost entirely excreted by the kidneys. It is also applicable to substances which are partially utilized and whose rate of utilization is proportional to the concentration of the substance in the plasma, such as xylose.[7,8]

With suitable modifications the method can be extended to more complicated problems in absorption.

[7] Dominguez, R., and Pomerene, E., *J. Clin. Invest.*, 1934, **13**, 753.

[8] Dominguez, R., Goldblatt, H., and Pomerene, E., *Am. J. Physiol.*, 1937, **119**, 429.

Unlike other methods used in physiology,[9] the present method determines the instantaneous rate of absorption. It can be used repeatedly in the same animal and is probably the only method that can be used in man. It throws a different light on the interpretation of the cumulative average absorption as calculated by Cori's method.[10] Thus, in the experiment on Subject G.N.B., the amount absorbed in 1 hr is 3.06 g, in 2 hr, 5.89 g, and in 3 hr, 6.13 g. Cori's coefficients are $3.06/1 = 3.06$, $5.89/2 = 2.95$, and $6.13/3 = 2.04$ g per hr. According to the usual interpretation, the rate of absorption of creatinine would have been considered constant in the first 2 hours. A glance at Fig. 2 shows that this conclusion would have been erroneous. In other words, the cumulative average amount absorbed per hour (Cori's coefficient) should not be considered the same as the rate of absorption.

Summary and Conclusions. For an inert substance in diffusion equilibrium, the rate of absorption at any time is shown to be equal to the rate of excretion plus the product of the volume of distribution and the rate of change in the concentration of the substance in the plasma. Two solutions of this equation are described and are applied to the absorption of creatinine in 3 human beings.

9 McGee, H. E., *Physiol. Rev.*, 1930, **10**, 473.

10 Cori, C. F., *J. Biol. Chem.*, 1925, **66**, 691.

15131

Effect of Pyridoxine on Granulopenia Caused by Thiouracil.*

ELLA H. FISHBERG AND JEFFERSON VORZIMER.

From the Biochemical Laboratory, Beth Israel Hospital, New York City.

In the course of an investigation of the effect of thiouracil on the basal metabolic rate of 96 hyperthyroid subjects[1] we encountered a marked fall in the granulocytic cells of 20% of these patients among which were 4 cases of true agranulocytosis. It has been noted by Bilter, Schiro and Spies[2] that intravenous injection of pyridoxine into 3 pellagrins with pernicious anemia resulted in an increase in the white cell count and that this increase took place principally in the granulocytic series. Cantor and Scott[3] have administered pyridoxine hydrochloride intravenously in daily doses of 125-200 mg to 3 cases of agranulocytosis of varied origin and have noted a marked and rapid return to a normal granulocyte count in each instance.

Although thiouracil reduced the B.M.R. in our hyperthyroid subjects at an average rate of 1 unit % per day, with parallel amelioration of all accompanying signs and symptoms of thyrotoxicosis, it had to be withdrawn forthwith after a rapid or precipitate fall in the white count. We therefore used pyridoxine in the hope of lowering the incidence of these hematological accidents.

To 8 hyperthyroid subjects receiving the usual dose of thiouracil 200 mg of pyridoxine were given daily by mouth. Fig. 1 shows the total leucocyte count for 8-week periods prior (open symbols), and during (solid symbols) pyridoxine administration. The average white counts were increased between 1500-2000 cells per cu mm.

Fig. 2 shows the leucocyte count of a subject receiving a 0.2 g maintenance dosage of thiouracil, in whom owing to the destructive action of thiouracil on the bone marrow, the granulocyte count dropped from 3700 to 400 cells per cu mm within 2 days. Four cc of a 5% solution of pyridoxine hydrochloride (200 mg) were injected and resulted in an increase

* Thiouracil and pyridoxine supplied by Dr. Staunton Hardy of Lederle Lab.

1 Fishberg, E. H., and Vorzimer, J., *J. Am. Med. Assn.*, 1945, **128**, 915.

2 Vilter, R. W., Schiro, H. S., and Spies, T. D., *Nature*, 1940, **145**, 388.

3 Cantor, M. M., and Scott, J. W., *Science*, 1944, **100**, 545.