detailed information about their physical properties, we might well find that Equation 7-53 was not really applicable to them. Therefore, in the next chapter we must turn to a less thorough and explicit, but more generally useful, analysis of the kinetics of transfer of substances between biological compartments.

EXERCISES. CHAPTER 7

Exercise 1

In textbooks of physiology it is commonly stated that in aqueous media carbon dioxide diffuses about 20 times as rapidly as oxygen. For example, Carlson, in the text by Ruch and Fulton (22), pages 796 to 797, states: "The intrinsic rate of diffusion of any substance is a function of its solubility, its molecular weight, and the permeability of the medium. Although a larger molecule than O_2, CO_2 is so highly soluble in the body fluids that it diffuses through the tissue 20 to 30 times as rapidly as O_2 does." What is the meaning of this statement?

Exercise 2

Forster (38) states, "Since the diffusion constant in air is about 1 million times that in saline . . . diffusion through 1 million microns (1 meter) of perfectly still gas would only demand a pressure difference equal to that normally associated with gas exchange across the pulmonary membrane." (It is assumed that the pulmonary membrane is 1 μ thick.) Is this statement justified?

Exercise 3

A. V. Hill (56) calculated the time needed for a sheet of muscle 1.0 mm. thick exposed to a constant concentration of oxygen on one side to attain by diffusion alone various average fractions of the equilibrium concentration of oxygen, the average being taken throughout the entire muscle. It was assumed that no oxygen was consumed by the tissue. For the diffusivity of oxygen in muscle he used the value $D_{O_2} = 4.5 \times 10^{-4}$ cm.^{2} per minute. Hill calculated that under these circumstances it would require 5 min. to reach an average of 53.4 per cent of the equilibrium concentration of oxygen throughout the tissue. Is this of the correct order of magnitude?

Exercise 4

Prove the validity of the statement made in the text (Section 7-4) that "over a considerable range of values of F_{eq} the time calculated for any particular value of D_S and any particular distance is roughly inversely proportional to the square of the fraction of equilibrium not yet attained, *i.e.*, inversely proportional to $(1 - F_{eq})^2$."

8

TRANSFER OF SUBSTANCES BETWEEN BIOLOGICAL COMPARTMENTS. GENERAL KINETICS

8-1. The Need for a More General Analysis of Transfer between Compartments

In Chapter 7 we undertook a detailed analysis of a particular mechanism —simple diffusion—by which a solute passes from one compartment to another. But it is often desirable to study the transfer of a drug, or a metabolite, or a radioactive isotope from one compartment to another without being concerned about the precise mechanism of transfer. For example, in the system discussed in Section 7-8 at the end of Chapter 7, if we knew Q_{tot} and if we were to measure the concentration of S in serial samples withdrawn from W and Z during the approach to equilibrium, we would be able to estimate V_W, V_Z, and $R_{Z/W}$. We could also calculate an exponential rate constant which would give us a very useful measure of the rate of approach to equilibrium, but would tell us nothing at all about the characteristics of the membrane. Indeed, the same kind of exponential approach to equilibrium can be caused by many processes other than simple passive diffusion (see Section 8-3). Therefore, it is desirable to undertake a more general analysis of the kinetics of transfer between compartments without reference to any particular mechanism of transfer. This analysis will then be applicable to a wide variety of problems.

8-2. Diagrams and Symbols for the Description of Transfer between Compartments

Figure 8-1 illustrates the kind of diagram and the symbols which will be used in the subsequent discussion. Each separate compartment is designated by a different capital letter and is represented in the diagram by a rectangle.* Each compartment is characterized by its volume V_A,

* The use of small rectangles for small compartments, large rectangles for large compartments often makes it easier to visualize the system. An even more elaborate

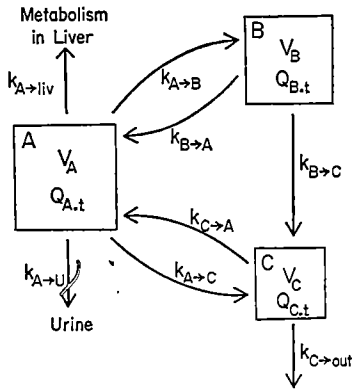

FIG. 8-1. Diagram of a Three-Compartment System

Each compartment is represented by a rectangle. Each pathway for transfer of solute is represented by a unidirectional arrow labeled with the appropriate rate constant. Once such a diagram has been drawn, the differential equations for the rate of change of the quantity of solute in each compartment can be written down by inspection (Section 8-6).

V_B, etc., and by the quantity of solute, $Q_{A.t}$, $Q_{B.t}$, etc., present in it at any instant of time. Each pathway by which the solute, S, moves out of or into a compartment is represented by an *arrow* pointing in the direction of movement. The quantity of S per unit of time which is moving out of a compartment by a particular pathway at any instant is assumed to be proportional to the quantity of S present in the compartment at that time (see Section 8-3). The *proportion* of S lost from the compartment along the pathway per unit of time can therefore be represented by an exponential rate constant, k, of exactly the sort discussed at length in Chapter 6. Each pathway has its own rate constant which is distinguished by a subscript denoting first the origin and then the destination of the pathway. For example, in Figure 8-1 the pathway which represents the renal excretion of some of the S in Compartment A is characterized by the rate constant $k_{A\to U}$; "$k_{A\to U}$" means "the proportion of the S in A which is being transferred *from A to* the urine per unit of time." Similarly, "$k_{B\to A}$" means "the proportion of the S in B which is being transferred *from B to A* per unit

pictorial device is to draw a cube in perspective for each compartment. The volume of the cube is made proportional to the volume of the compartment. Moreover, the thickness of an arrow representing a given pathway can be made proportional to the rate of transfer along that pathway. Examples of such diagrams may be found in Reference 88. For ordinary purposes, no such elaborations are needed.

of time"; "$k_{C\to out}$" means "the proportion of the S in C which is being transferred per unit of time *from C to* somewhere outside the system."

Now when the rate of transfer of S from Compartment A to Compartment B is proportional to the quantity of S present in A, the rate of transfer of S *by that particular pathway* will be given by the equation

$$(dQ/dt)_{A\to B} = k_{A\to B}Q_{A.t} \qquad (8\text{-}1)$$

where

$(dQ/dt)_{A\to B}$ = the rate at which S is being transferred along the pathway from A to B at time t

$Q_{A.t}$ = the quantity of S in Compartment A at time t

8-3. Simplifying Assumptions

To simplify the mathematical description of transfer between compartments we shall make the following assumptions:

1. We shall assume that the size, *i.e.*, the volume, of each compartment remains constant. This means that any equation for the *quantity* of S in a given compartment as a function of time can be converted to an equation for the *concentration* of S in the compartment as a function of time by dividing both sides of the equation by the volume of the compartment.
2. We shall assume that each compartment is well stirred, so that any S entering the compartment is instantaneously distributed throughout the entire compartment. The importance of this assumption has already been discussed (Section 7-3).
3. We shall assume that the proportional rate of transfer along each pathway remains constant so that the resulting change in quantity or concentration is exponential. In other words, we assume that the rate constants, k, are indeed constant. (Occasionally we may want to deal with a pathway through which a *constant quantity* of S per unit time is passing, but we shall characterize such a pathway not by a rate constant k, but by a symbol such as $\dot{Q}_{A\to out}$ which would mean quantity of S transferred out of Compartment A per unit of time.)

When these assumptions are not true for a particular system, the equations to be derived cannot properly be applied to that system. But fortunately, the assumptions are sufficiently valid for many of the biological systems in which we shall be interested. In particular, Assumption 3 is not as restrictive as it might seem, for a remarkable number of processes do transfer solutes at a rate which is proportional to concentration. Exponential change is the rule, not the exception! We have already seen that simple diffusion through a membrane between two well-stirred compart-

ments causes an exponential change in concentration. Similarly, the excretion of a solute solely by filtration through the glomeruli of the kidney will cause exponential disappearance if the glomerular filtration rate remains constant. But it is not such "passive" processes alone which account for exponential changes in concentration. An active process which removes S from a compartment may equally well produce exponential change *provided that the rate of removal is limited by the rate at which S is supplied to the process, and not by the capacity of the process itself.* For example, suppose that 1 out of every 5 molecules of S brought to the liver by a constant hepatic blood flow is destroyed enzymatically by the liver cells regardless of the concentration of S in the blood. Then the process is *supply-limited** and S will disappear exponentially. But if the enzyme system is so fully saturated with S that it is destroying S as fast as it can, then the quantity of S destroyed per unit of time will be constant regardless of the concentration of S in the blood and of the rate of blood flow. Such a process is *capacity-limited.** Many active processes responsible for the transport or removal of solutes become capacity-limited if they are supplied with substrate at a high enough rate. A familiar example is the glucosuria which occurs when the amount of glucose filtered per minute through the glomeruli of the kidney exceeds the reabsorptive capacity, *i.e.*, the "transfer maximum," of the renal tubular cells for glucose.

8-4. The Concept of Clearance

Because so many different processes may be responsible for removing S from a given compartment, it would be convenient to have some very general way of expressing the overall effectiveness of *any* such process. The *absolute rate of removal*, expressed as quantity of S removed per unit of time, would be suitable only when the process is capacity-limited, for otherwise it would be as dependent upon the concentration of S as upon the effectiveness of the process of removal. The *proportional rate of removal*, expressed by the rate constant, k, is also unsuitable as a general term because it is constant only so long as the rate of removal of S from the compartment remains proportional to the quantity of S in the compartment. Furthermore, the magnitude of the rate constant depends as much upon the volume of the compartment as it does upon the effectiveness of the process of removal. In contrast, the *clearance* depends only upon the

* Because of their broader meaning, the terms "supply-limited" and "capacity-limited" are used here in preference to the corresponding terms "substrate-limited" and "enzyme-limited." The capacity of some transport mechanisms may well be limited by how much of a nonenzymatic "carrier substance" is available for combination with S rather than by the amount of enzyme needed to form (or to split) the carrier-S complex.

overall effectiveness of removal, and can be used to characterize any process of removal whether it be constant or changing, capacity-limited or supply-limited.

The clearance of S from Compartment A by the pathway leading to B may be defined as the volume of A from which S is, in effect, completely removed, *i.e.*, "cleared," per unit of time via that pathway. It may equally well be defined as the *rate of removal of S from A* via the pathway to B *per unit of concentration in A*:

$$(\dot{V}\mathrm{cl})_{A\to B.t} = (dQ/dt)_{A\to B}/C_{A.t} \tag{8-2}$$

where

$(\dot{V}\mathrm{cl})_{A\to B.t}$ = the volume of W being cleared of S per unit of time by the pathway from A to B at time t

Equation 8-2 is a completely general definition of clearance, valid (with appropriate changes in the subscripts) for any substance cleared from any compartment along any pathway at any time. It is important to realize that Equation 8-2 is for clearance via any single *unidirectional pathway*. It will represent *net* removal of S from Compartment A only when there is no movement of S along any return pathway from B to A. But in practice, the term "clearance" is rarely used except for the irreversible removal of S from a compartment by unidirectional pathways of metabolism, storage, or excretion, and it is in this more restricted sense that the term will ordinarily be employed.

Notice that clearance has the dimensions of flow, *i.e.*, of volume per unit of time. It is sometimes called a "virtual flow." Indeed as far as the removal of S from A is concerned, the effect of a clearance of 15 ml. of A per minute is exactly the same as the effect of washing S out of Compartment A by continuous dilution with a real flow of 15 ml. per minute through the compartment (Table 9-1). Now by definition

$$C_{A.t} = Q_{A.t}/V_A \tag{8-3}$$

Combining Equations 8-2 and 8-3,

$$(\dot{V}\mathrm{cl})_{A\to B.t} = \frac{(dQ/dt)_{A\to B}}{Q_{A.t}/V_A} \tag{8-4}$$

Rearranging terms,

$$(dQ/dt)_{A\to B} = \frac{(\dot{V}\mathrm{cl})_{A\to B.t}}{V_A}\, Q_{A.t} \tag{8-4a}$$

By Assumption 1, Section 8-3, V_A is constant. Suppose that the clearance is also constant. Then the rate constant $k_{A\to B}$ in Equation 8-1 and the

constant $(\dot{V}\text{cl})_{A\to B}/V_A$ in Equation 8-4a are identical:

$$k_{A\to B} = (\dot{V}\text{cl})_{A\to B}/V_A \tag{8-5}$$

Thus when the volume of the compartment being cleared is constant (Assumption 1), the assumption that the proportional rate of transfer is constant (Assumption 3) is equivalent to assuming that the clearance is constant, the *rate constant being the ratio of the clearance to the compartment volume*. This makes sense. We have learned to think of a rate constant as a proportion per unit of time, and the ratio in Equation 8-5 is simply the proportion of the total volume of Compartment A which is cleared of S per unit of time via the pathway from A to B.

8-5. The Concept of Volume of Distribution

If clearance is a "virtual flow," the volume of distribution of a substance, S, is a "virtual volume." Suppose that the total quantity of S in the body, Q_{tot}, has had time to reach distribution equilibrium throughout all of the compartments which it can enter. Suppose that the concentration of S in some part of one of these compartments (usually blood plasma) can be measured. Let us call the equilibrium concentration in this reference fluid $C_{\text{ref.eq}}$. Then the volume of distribution, $(V\text{dist})$, of S is simply

$$(V\text{dist}) = Q_{\text{tot}}/C_{\text{ref.eq}} \tag{8-6}$$

Notice that the concept of volume of distribution is an *equilibrium* concept.

If the distribution of S is through a *single* compartment, say Compartment A, the term "volume of distribution in Compartment A" is synonymous with the term "volume of Compartment A" as defined in Section 7-3. This is still in keeping with the idea that volume of distribution can be defined only for distribution equilibrium, because the S in any single compartment is *always* supposed to be at distribution equilibrium throughout that compartment (Assumption 2, Section 8-3). Thus the term "initial volume of distribution" simply means the volume of the first compartment through which Q_{tot} is apparently distributed at time zero.

An example of a volume of distribution has already been given in Equation 7-54 which is really a specific example of Equation 8-6. The problem of how to measure the volume of distribution of S throughout a system of compartments will be discussed later (Section 8-11).

8-6. Differential Equations for Transfer between Compartments

Given a system of compartments and pathways such as the one depicted in Figure 8-1, nothing is easier than to write down a differential equation for the rate of change of the quantity of S in each compartment. Equation 8-1 shows that the rate at which S traverses any particular pathway is the

product of the rate constant for that pathway and the quantity of S in the compartment from which the pathway is coming. Obviously the *total* change in any compartment is the algebraic sum of all of the individual increments and decrements caused by transfer of S along all pathways leading to and from the compartment. For example, in Compartment A of Figure 8-1 there are two pathways (from B and from C) *adding* S to A, and four pathways (to B, to C, to the liver, and to the kidneys) *subtracting* S from A. At any instant of time, t, the quantity of S in A is therefore changing at the rate given by the following equation:

$$dQ_A/dt = k_{B\to A}Q_{B.t} + k_{C\to A}Q_{C.t} - k_{A\to B}Q_{A.t} - k_{A\to C}Q_{A.t} - k_{A\to \text{liv}}Q_{A.t} - k_{A\to U}Q_{A.t} \tag{8-7}$$

Similarly,

$$dQ_B/dt = k_{A\to B}Q_{A.t} - k_{B\to A}Q_{B.t} - k_{B\to C}Q_{B.t} \tag{8-8}$$

$$dQ_C/dt = k_{B\to C}Q_{B.t} + k_{A\to C}Q_{A.t} - k_{C\to A}Q_{C.t} - k_{C\to \text{out}}Q_{C.t} \tag{8-9}$$

Since all rate constants for pathways leading *away* from a given compartment are multiplied by the quantity of S in that compartment, it is convenient to have a single symbol to designate their sum. We will therefore define k_A to mean the sum of all the rate constants for pathways leading *away from* Compartment A. Accordingly, Equations 8-7, 8-8, and 8-9 can be rewritten:

$$dQ_A/dt = k_{B\to A}Q_{B.t} + k_{C\to A}Q_{C.t} - k_AQ_{A.t} \tag{8-10}$$

$$dQ_B/dt = k_{A\to B}Q_{A.t} - k_BQ_{B.t} \tag{8-11}$$

$$dQ_C/dt = k_{A\to C}Q_{A.t} + k_{B\to C}Q_{B.t} - k_CQ_{C.t} \tag{8-12}$$

Equations 8-10, 8-11, and 8-12 are a set of simultaneous differential equations which completely describe the behavior of S in the system at any instant of time. But as usual, the differential equations must be solved, *i.e.*, integrated, before we can use them to find the actual quantity of S in a given compartment at a specific time. Using an analog computer (Section 3-3) to obtain particular solutions of the set of differential equations not only saves much time and effort but may be the only practical way to deal with really complex systems of compartments. Obtaining a general analytical solution even for a two- or three-compartment system requires a knowledge of calculus beyond what is being assumed for this book. Nevertheless, we shall be able to solve one or two elementary problems completely, and we shall also be able to analyze certain more complex systems by using a general solution worked out by others (97). In the following sections, three examples are given in full. Others are suggested

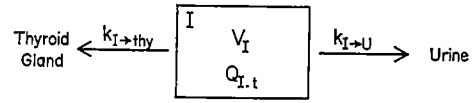

FIG. 8-2. Two Pathways of Loss from a Single Compartment

In the period immediately after the administration of a tracer dose of radioactive iodide, its behavior can often be described adequately by this very simple model (Section 8-7).

as exercises at the end of the chapter. In fact, the reader is strongly advised to work Exercise 1 himself *before* proceeding to the next few paragraphs.

8-7. Irreversible Loss via Several Pathways from a Single Compartment (Fig. 8-2)

Consider the removal of a single tracer dose of radioactive iodide, I^{131}, from the iodide compartment, I, by accumulation in the thyroid gland, thy, and by excretion in the urine, U. We will assume that during the period of interest all of the iodide which enters the thyroid gland is stored there as organic iodine and that none is returned to the iodide compartment or secreted as hormone. (This assumption is often, but by no means always, justified by the actual behavior of a tracer dose of I^{131}.)

Initial conditions: at $t = 0$, $Q_{I.0} = Q_{\text{tot}}$, $Q_{\text{thy}.0} = 0$, $Q_{U.0} = 0$

By inspection of Figure 8-2, the differential equation for Q_I is

$$\begin{aligned} dQ_I/dt &= -k_{I\to\text{thy}}Q_{I.t} - k_{I\to U}Q_{I.t} \\ &= -(k_{I\to\text{thy}} + k_{I\to U})Q_{I.t} = -k_I Q_{I.t} \end{aligned} \tag{8-13}$$

Integrating Equation 8-13,

$$Q_{I.t} = Q_{I.0}e^{-k_I t} = Q_{\text{tot}}e^{-k_I t} \tag{8-14}$$

Equation 8-14 shows that even though the I^{131} is leaving Compartment I by two separate pathways, its rate of disappearance from I is controlled by a single exponential term for which the rate constant is the *sum* of the rate constants of the two efferent pathways. Two leaks "are equal to one leak of larger size" (101).

Equation 8-14 enables us to calculate how much of the tracer dose remains in the iodide compartment at any time. According to Figure 8-2, the iodide which has left the iodide compartment must either have accumulated in the thyroid gland or have been excreted in the urine. But how much goes to each? It is obvious from Equation 8-13 that at any particular *instant* of time, the fraction of the total change due to thyroid accumulation, $F_{\text{thy/tot}}$, is

$$F_{\text{thy/tot}} = k_{I\to\text{thy}}Q_{I.t}/k_I Q_{I.t} = k_{I\to\text{thy}}/k_I \tag{8-15}$$

But since the k's are constant, this fraction is also constant and represents the fraction of $Q_{\text{tot}} - Q_{I.t}$ (*i.e.*, the quantity lost from I between time zero and time t) which has accumulated in the thyroid gland. Hence,

$$Q_{\text{thy}.t} = F_{\text{thy/tot}}(Q_{\text{tot}} - Q_{I.t}) = \frac{k_{I\to\text{thy}}}{k_I}(Q_{\text{tot}} - Q_{I.t}) \tag{8-16}$$

Substituting in Equation 8-16 the value of $Q_{I.t}$ from Equation 8-14,

$$Q_{\text{thy}.t} = \frac{k_{I\to\text{thy}}}{k_I}Q_{\text{tot}}(1 - e^{-k_I t}) \tag{8-17}$$

Similarly, if $Q_{U.t}$ represents the cumulative excretion of I^{131} in the urine between time zero and time t,

$$Q_{U.t} = \frac{k_{I\to U}}{k_I}Q_{\text{tot}}(1 - e^{-k_I t}) \tag{8-18}$$

By combining Equations 8-15 and 8-17 we obtain

$$(Q_{\text{thy}.t} - F_{\text{thy/tot}}Q_{\text{tot}}) = (Q_{\text{thy}.0} - F_{\text{thy/tot}}Q_{\text{tot}})e^{-k_I t} \tag{8-19}$$

where, according to the initial conditions, $Q_{\text{thy}.0} = 0$. Equation 8-18 may be rearranged in like manner. Equation 8-19 makes it clear that the quantity of I^{131} in the thyroid gland is rising toward the asymptote $F_{\text{thy/tot}}Q_{\text{tot}}$ at a rate determined by k_I, the *sum* of the rate constants for transfer of I^{131} to thyroid *and* to urine. Similarly the rate at which the cumulative excretion of I^{131} in the urine approaches its asymptote depends not just on the rate constant $k_{I\to U}$ for urinary excretion but upon the *sum* of the two rate constants.

This important conclusion may be generalized as follows: Suppose that an amount Q_{tot} of S is placed into Compartment A at time zero. Suppose that S is then *irreversibly* lost from A by several routes (to B, to C, to D, etc.), each characterized by its own rate constant. Then *the cumulative loss by any single route, say A to B, approaches an asymptote equal to* $(k_{A\to B}/k_A)Q_{\text{tot}}$ *with a half-time which is determined not by* $k_{A\to B}$ *alone but by* k_A, *the sum of all of the rate constants.* This means that the straight lines obtained by plotting ln $Q_{A.t}$ against time, ln $(Q_{B.\text{asymp}} - Q_{B.t})$ against time ln $(Q_{C.\text{asymp}} - Q_{C.t})$ against time, etc., will all be parallel with a slope of $-k_A = -(k_{A\to B} + k_{A\to C} + \cdots)$ (see Exercise 2).

8-8. Equilibration by Exchange in a Closed Two-Compartment System (Fig. 8-3)

A specific example of how equilibrium was approached by diffusion between two compartments was considered in detail in Chapter 7. We shall now derive more general equations for equilibration between two compartments, equations which will not depend upon the assumption of

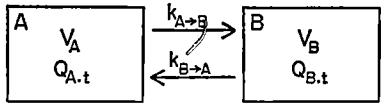

FIG. 8-3. A Closed Two-Compartment System

If a solute is introduced at time zero into either Compartment A or Compartment B, it approaches equilibrium throughout the entire system at the single exponential rate specified by the sum of the two rate constants (Section 8-8).

any particular mechanism of transfer. In the diffusion problem, we dealt only with *net* transfer. In the general case, it is easier to consider the transfer of solute in both directions.

Consider the approach to equilibrium when a solute, S, is added to Compartment A of the system depicted in Figure 8-3. By setting appropriate boundary conditions we will take into account the possibility that there may already be some S present in the two compartments at time zero.

$$\text{Initial conditions: at } t = 0, \qquad Q_A = Q_{A.0}, \qquad Q_B = Q_{B.0}$$

The differential equation for Compartment A is

$$dQ_A/dt = k_{B\to A}Q_{B.t} - k_{A\to B}Q_{A.t} \tag{8-20}$$

The differential equation for Compartment B is similar but with opposite signs. Since the system is a closed system (*i.e.*, one with no outlets), conservation of S requires that

$$Q_{tot} = Q_{A.t} + Q_{B.t} \tag{8-21}$$

Combining Equations 8-20 and 8-21 with the elimination of $Q_{B.t}$,

$$dQ_A/dt = k_{B\to A}Q_{tot} - (k_{A\to B} + k_{B\to A})Q_{A.t} \tag{8-22}$$

or, separating the variables prior to integration,

$$dQ_A/[k_{B\to A}Q_{tot} - (k_{A\to B} + k_{B\to A})Q_{A.t}] = dt \tag{8-22a}$$

Equation 8-22a may now be integrated in exactly the same way as Equation 7-44, $k_{B\to A}Q_{tot}$ corresponding to k_6 and $(k_{A\to B} + k_{B\to A})$ corresponding to k_5. Therefore, we need not repeat the intermediate steps but can simply copy the result given in Equation 7-48a:

$$\left(Q_{A.t} - \frac{k_{B\to A}}{k_{A\to B} + k_{B\to A}}Q_{tot}\right) = \left(Q_{A.0} - \frac{k_{B\to A}}{k_{A\to B} + k_{B\to A}}Q_{tot}\right) \exp\left[-(k_{A\to B} + k_{B\to A})t\right] \tag{8-23}$$

From Equation 8-23 it is evident that $Q_{A.t}$ is approaching its asymptote,

$k_{B\to A}Q_{tot}/(k_{A\to B} + k_{B\to A})$, at a rate determined by the sum of the two rate constants. We can easily prove that this asymptote is indeed $Q_{A.\infty}$, the equilibrium value which is being approached as time approaches infinity. For, by definition, at equilibrium as much S must move per unit of time from A to B as from B to A so that the net change in Q_A as given by Equation 8-22 will be zero. Then Equation 8-22 becomes

$$(k_{A\to B} + k_{B\to A})Q_{A.\infty} = k_{B\to A}Q_{tot} \tag{8-24}$$

Solving Equation 8-24 for $Q_{A.\infty}$,

$$Q_{A.\infty} = k_{B\to A}Q_{tot}/(k_{A\to B} + k_{B\to A}) \tag{8-24a}$$

which is indeed the asymptote of $Q_{A.t}$ in Equation 8-23.

Since Equation 8-23 is a general equation for equilibration in a closed two-compartment system, it should include Equation 7-52 for equilibration by diffusion as a special case. Notice that in Equation 7-52 the asymptote is the product of Q_{tot} and a ratio of volumes, whereas in Equation 8-23 the asymptote is the product of Q_{tot} and a ratio of rate constants. By using Equation 8-5 it is not difficult to prove that these ratios are equal. It is also possible to show that the exponent in Equation 7-52 corresponds to the exponent in Equation 8-23, though the reasoning is a bit more subtle (see Exercise 8).

8-9. A General Solution of the Two-Compartment Problem

The two previous examples have been chosen for their simplicity. Because of such special constraints as unidirectional transport, or absence of any pathways leading out of the system, the solutions contained only a single exponential term. But where no such constraints are imposed, general solutions soon become uncomfortably complex as the number of compartments increases. For the sake of simplicity, let us assume that at time zero a known amount, Q_{tot}, of S is given as a single dose into Compartment A, and that there is initially no S in any of the other compartments. Suppose that we have a system of N compartments, each having pathways both to and from every other compartment as well as a pathway leading out of the system. Then there would be altogether N^2 different pathways, each with its own rate constant. Each compartment will also have a volume, so that such a system of N compartments might have as many as $N^2 + N$ arbitrary constants or parameters. If we assume that all of these parameters are known, it will still be necessary to solve a set of N simultaneous differential equations, one for each compartment. Even with a three-compartment system, the resulting integrated equations are too complex to discuss here, although an explicit solution for the three-compartment system can be obtained (97). However, the explicit equations

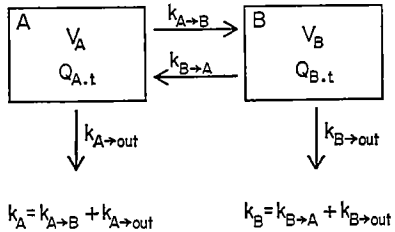

FIG. 8-4. An Open Two-Compartment System

A general solution of the differential equations for this system leads to somewhat complicated expressions which contain two exponential terms (Section 8-9).

for the generalized two-compartment system of Figure 8-4 are not as formidable, and will be found useful for the solution of a number of different problems. These equations have been obtained by simplification of the general solution for the three-compartment system published by Skinner *et al.* (97). The equations, which will be given here without derivation, are for the fraction of the total dose in Compartment A, $F_{A.t}$, and the fraction of the total dose in Compartment B, $F_{B.t}$, at any time, t:

$$F_{A.t} = \left\{\left(\frac{k_A - k_B + Z}{2Z}\right) \exp\left[-\tfrac{1}{2}(k_A + k_B + Z)t\right]\right\} + \left\{\left(1 - \frac{k_A - k_B + Z}{2Z}\right) \exp\left[-\tfrac{1}{2}(k_A + k_B - Z)t\right]\right\} \tag{8-25}$$

$$F_{B.t} = \frac{k_{A\to B}}{Z}\left\{\exp\left[-\tfrac{1}{2}(k_A + k_B - Z)t\right] - \exp\left[-\tfrac{1}{2}(k_A + k_B + Z)t\right]\right\} \tag{8-26}$$

where

$$Z = \sqrt{(k_A - k_B)^2 + 4k_{A\to B}k_{B\to A}} \tag{8-27}$$

Initial conditions: at $t = 0$, $\quad F_{A.0} = 1$, $\quad F_{B.0} = 0$

These equations can readily be converted into equations for the *concentration* of S in the two compartments at any time, t, by multiplying the fraction by the total dose divided by the volume of the compartment:

$$C_{A.t} = Q_{A.t}/V_A = Q_{\text{tot}}F_{A.t}/V_A \tag{8-28}$$

and

$$C_{B.t} = Q_{B.t}/V_B = Q_{\text{tot}}F_{B.t}/V_B \tag{8-29}$$

It is well worth noting that *both* of the exponential terms in Equation

8-25 are influenced by *all* of the rate constants, and hence by all of the individual processes of transfer illustrated in Figure 8-4. We are thus warned once more that when a multiple exponential equation is fitted empirically to biological data, in general we have no right to identify a particular term with a particular process occurring at the corresponding exponential rate.

8-10. Calculation of Rate Constants and Compartment Volumes from Experimental Observations

In the discussion thus far it has been assumed that the rate constants and the compartment volumes were known, so that the problem was to find a set of equations which would enable one to predict the distribution of S in the system as a function of time. But in practice, it is far more common to observe the concentration of S in one or more compartments as a function of time, and to try to deduce the parameters of the system from the observed behavior of S. For this purpose it is of the utmost value to have some *a priori* knowledge about how many compartments there are and what pathways interlink them. Otherwise, the calculated values are not likely to mean very much physiologically.

Example

There is substantial evidence that in man and in the dog, creatinine is distributed throughout two compartments, of which the first (which we shall call A) includes blood plasma, whereas the second (which we shall call B) is not clearly identified but presumably includes at least some "intracellular fluid." It is not possible to obtain samples for analysis from Compartment B. Exogenous creatinine leaves the body only via the urine. The behavior of creatinine may accordingly be represented by the model shown in Figure 8-5. The parameters in the following example have been taken from the discussion by Dominguez in *Medical Physics* (33).

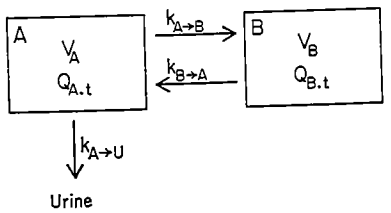

FIG. 8-5. A Model System for the Behavior of Creatinine in the Mammalian Body

The several parameters of this model system can be estimated from experimental observations as described in Section 8-10.

Ten grams of creatinine were administered intravenously at time zero to a human subject. Thereafter, the concentration of creatinine in plasma, corrected for endogenous creatinine, was measured at various times. The concentration was plotted against time on semilog graph paper, and the data were analyzed as described in Section 6-14. The points could be fitted by the following equation:

$$C_{A.t} = 0.38e^{-1.65t} + 0.18e^{-0.182t} \tag{8-30}$$

where

$C_{A.t}$ = the concentration of creatinine in plasma (regarded as a sample of Compartment A) in grams per liter
t = the time in hours

At time zero, Equation 8-30 gives

$$C_{A.0} = 0.38 + 0.18 = 0.56 \text{ gm./L.} \tag{8-31}$$

Since the dose administered, Q_{tot}, was 10 gm., the initial volume of distribution (taken as a measure of the volume of Compartment A) was

$$V_A = Q_{\text{tot}}/C_{A.0} = 10.0/0.56 = 17.9 \text{ L.} \tag{8-32}$$

Multiplying both sides of Equation 8-30 by $V_A/Q_{\text{tot}} = 1.79$ to convert it to $F_{A.t}$,

$$F_{A.t} = 0.68e^{-1.65t} + 0.32e^{-0.182t} \tag{8-33}$$

Equation 8-33 is a particular example of Equation 8-25. We can therefore equate the *numerical* values in Equation 8-33 to the corresponding *algebraic* values of Equation 8-25:

$$\tfrac{1}{2}(k_A + k_B + Z) = 1.65 \tag{8-34}$$

$$\tfrac{1}{2}(k_A + k_B - Z) = 0.182 \tag{8-35}$$

$$(k_A - k_B + Z)/2Z = 0.68 \tag{8-36}$$

Solving Equations 8-34 and 8-35 simultaneously for Z, we get $Z = 1.468$. Placing this value in Equation 8-36 and either 8-34 or 8-35 and solving the resulting equations simultaneously, we obtain $k_A = 1.180$ and $k_B = k_{B\to A} = 0.652$. Now by Equation 8-27

$$Z^2 = (k_A - k_B)^2 + 4k_{A\to B}k_{B\to A} \tag{8-27a}$$

Substituting our known values in Equation 8-27a, we find $k_{A\to B} = 0.719$, and, by subtracting this value from k_A, $k_{A\to U} = 0.461$. We have thus been able to calculate values for all of the rate constants

as well as for the volume of Compartment A:

$$k_{A\to B} = 0.719 \text{ per hour}$$

$$k_{A\to U} = 0.461 \text{ per hour}$$

$$k_{B\to A} = 0.652 \text{ per hour}$$

$$V_A = 17.9 \text{ L.}$$

We can therefore write a specific equation for $F_{B.t}$ in the form of Equation 8-26:

$$F_{B.t} = 0.49(e^{-0.182t} - e^{-1.65t}) \tag{8-37}$$

We can also multiply Equation 8-37 by Q_{tot} to obtain an equation for the quantity of creatinine in B at any time. But to complete our characterization of the system what we really need is an estimate of V_B, the volume of Compartment B. If we could get even a single sample of B for analysis, we could measure $C_{B.t}$. Since we can calculate $Q_{B.t}$, we could then estimate V_B as $Q_{B.t}/C_{B.t}$. But B is not open to sampling, and we therefore seem to be stuck!

Actually, the problem is not as hopeless as it seems. It is true that we cannot calculate the volume of B with reference to the concentration of creatinine in some portion of B itself. But it is quite easy to calculate the volume of distribution of creatinine in B with reference to its concentration in A as measured in samples of plasma. By definition (Equation 8-6), this will be

$$(V\text{dist})_{B_A} = Q_{B.\text{eq}}/C_{A.\text{eq}} \tag{8-38}$$

where

$(V\text{dist})_{B_A}$ = the volume of distribution in B with reference to the concentration in A

Also by definition (Equation 7-50),

$$R_{B/A} = C_{B.\text{eq}}/C_{A.\text{eq}} \tag{8-39}$$

Combining Equations 8-38 and 8-39 with the elimination of $C_{A.\text{eq}}$,

$$(V\text{dist})_{B_A} = R_{B/A}Q_{B.\text{eq}}/C_{B.\text{eq}} \tag{8-40}$$

or, since $Q_{B.\text{eq}}/C_{B.\text{eq}} = V_B$,

$$(V\text{dist})_{B_A} = R_{B/A}V_B \tag{8-41}$$

Now at equilibrium, the rate of transfer from A to B must be exactly equal to the rate of transfer from B to A. Hence,

$$k_{A\to B}Q_{A.\text{eq}} = k_{B\to A}Q_{B.\text{eq}} \tag{8-42}$$

or, since $Q_{A.eq} = C_{A.eq}V_A$, and $Q_{B.eq} = C_{B.eq}V_B$,

$$k_{A\to B}C_{A.eq}V_A = k_{B\to A}C_{B.eq}V_B \tag{8-43}$$

Combining Equations 8-39 and 8-43 with the elimination of $C_{B.eq}$, solving for $V_BR_{B/A}$, and including Equation 8-41 in the result,

$$(V\text{dist})_{B_A} = V_BR_{B/A} = (k_{A\to B}/k_{B\to A})V_A \tag{8-44}$$

Since the three quantities on the right of Equation 8-44 have already been evaluated, $(V\text{dist})_{B_A}$ may be calculated as 19.7 L. The total volume of distribution of creatinine in the entire system, calculated with reference to its concentration in A, is simply the sum of V_A and $(V\text{dist})_{B_A}$ as given by Equation 8-44:

$$(V\text{dist}) = V_A + (V\text{dist})_{B_A} = V_A\left(\frac{k_{A\to B} + k_{B\to A}}{k_{B\to A}}\right) \tag{8-45}$$

For this example, the total volume of distribution is 37.6 L.

The example just given shows that in a two-compartment system whose general arrangement is known, careful analysis of the changes in concentration in one of the two compartments coupled with knowledge of the boundary conditions allows one to estimate the rate constants and volumes which characterize the system. Skinner and his collaborators (97) discuss methods by which the parameters of even more complex three-compartment systems can be calculated. However, in interpreting the results of any such analysis one must bear clearly in mind the considerable uncertainties of fitting observed data by exponential equations (see Chap. 6). Whenever possible the calculations should be checked, supplemented, or even partially replaced by additional methods of studying the system. For instance, in the preceding example it would be highly desirable to collect samples of urine as well as of blood plasma and to calculate the renal plasma clearance of creatinine therefrom in the time-honored manner:

$$(\dot{V}\text{cl})_{A\to U} = \bar{\dot{Q}}_{U.t_{1\text{-}2}}/\bar{C}_{A.t_{1\text{-}2}} \tag{8-46}$$

where

$\bar{\dot{Q}}_{U.t_{1\text{-}2}}$ = the mean rate of excretion of creatinine in the urine between time 1 and time 2

$\bar{C}_{A.t_{1\text{-}2}}$ = the mean concentration (properly, the logarithmic mean concentration) of creatinine in the plasma between time 1 and time 2

This value for plasma clearance could then be compared with the value indirectly calculated by the relationship given in Equation 8-5:

$$(\dot{V}\text{cl})_{A\to U} = k_{A\to U}V_A \tag{8-47}$$

For the preceding example, $k_{A\to U}V_A$ = 8.25 L. of plasma per hour, or 138 ml. of plasma per minute, a value which is in reasonably good agreement with values for the renal plasma clearance of creatinine obtained by direct measurement in man.

8-11. The Measurement of Volume of Distribution

The term "volume of distribution of S" has already been defined as the volume of solution which, if it had a uniform concentration equal to $C_{ref.eq}$, would contain the same total amount of S as is distributed about the entire system at equilibrium, *i.e.*, when there is no net transfer of S between compartments. $C_{ref.eq}$ is the equilibrium concentration in some well-defined portion of the system, usually blood plasma. We must now see how the volume of distribution of S can be measured.

In a closed system where S is neither metabolized, nor excreted, nor hidden away in some storage depot, the problem is simple. A known dose of S, Q_{tot}, is administered at time zero, and the concentration of S in plasma is studied as a function of time until enough data have been gathered to define the equilibrium concentration being approached asymptotically. The volume of distribution of S in the whole system, $(V\text{dist})$, with reference to its equilibrium concentration in plasma, $C_{P.eq}$, is then given by the following variant of Equation 7-54:

$$(V\text{dist}) = Q_{tot.eq}/C_{P.eq} \tag{8-48}$$

where

$Q_{tot.eq}$ = the quantity distributed throughout the entire system at equilibrium (in this instance, equal to Q_{tot})

Usually, however, S is lost more or less rapidly by irreversible pathways of metabolism, excretion, and storage, so that a steady state of equilibrium is not approached with the passage of time after a single dose. Nevertheless, it may be possible to approximate a steady state of equilibrium by infusing S intravenously at a constant rate for a long period of time. For this method to be valid, all of the irreversible loss of S must be from the same compartment as the one into which S is being infused. Consider the system in Figure 8-6. Compartments B, C, and D have no outlet except via Compartment A. During continuous infusion of S at a constant rate of $\dot{Q}_{in\to A}$, S will accumulate in the several compartments of the system until the rate of loss, $k_{A\to out}Q_A$, equals the rate of infusion. In theory, this would occur only at infinite time. But in practice, if interchange between A, B, C, and D is reasonably rapid, equilibrium will be approached sufficiently closely in a finite time, *e.g.*, a few hours. At equilibrium Equation 8-48 can be used if $Q_{tot.eq}$ can be estimated, for example, by measuring S in serial samples of urine collected after abruptly discontinuing the in-

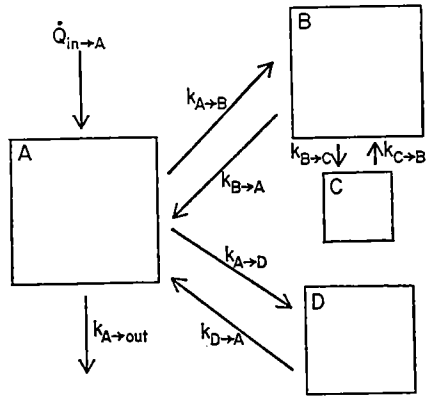

FIG. 8-6. A System of Compartments Which *Will* Approach Equilibrium with Each Other during Continuous Infusion of the Solute at a Constant Rate into Compartment A

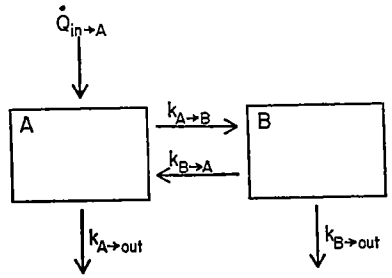

FIG. 8-7. A System of Compartments Which *Will Not* Approach Equilibrium with Each Other during Continuous Infusion of the Solute at a Constant Rate into Compartment A

fusion. This method can *not* be used for any system such as the one depicted in Figure 8-7 because part of the S infused into A is being lost from B. If S is continuously infused at a rate of $\dot{Q}_{in\to A}$, this system, like the previous one, will approach a steady state at which the rate of infusion is just equaled by the total rate of loss, $k_{A\to out}Q_A + k_{B\to out}Q_B$. At this steady state, it is obvious that $k_{A\to B}Q_A$ must *exceed* $k_{B\to A}Q_B$ by an amount equal to $k_{B\to out}Q_B$, whereas distribution equilibrium between A and B can be present only when $k_{A\to B}Q_A$ *equals* $k_{B\to A}Q_B$. Hence, even though the system approaches a *steady state*, it does not approach a state of distribution *equilibrium*, and Equation 8-48 cannot properly be applied.

A third method of estimating the volume of distribution is to add up

the volumes calculated individually for the different compartments. A detailed example for a two-compartment system was discussed above (Equation 8-45).

8-12. Biased Methods of Estimating Volume of Distribution

There are still other methods, which must be employed with great caution because they are biased in the direction of overestimating the volume of distribution. They may therefore give entirely erroneous results if used without proper appreciation of their limitations. Consider again the system in Figure 8-5, in which the only route of loss is from Compartment A to the urine, U. Suppose first that $k_{A\to U}$ is zero. Then the system would be simply a closed two-compartment system approaching both diffusion equilibrium and a steady state with the passage of time. Now suppose that $k_{A\to U}$ is greater than zero, but very much less than $k_{A\to B}$ and $k_{B\to A}$ which control the rate of distribution between A and B. Then the approach of A and B to equilibrium with each other will be scarcely influenced by the slow rate of loss of S in the urine. Therefore, after enough time for distribution has elapsed, *i.e.*, after the first exponential term in Equation 8-25 has become negligibly small, A and B will behave practically like a single compartment, which we shall call $(A + B)$, with a volume equal to $V_A + (V\text{dist})_{B_A}$ and a concentration equal to $C_{A.t}$. We can derive an equation for the concentration in this quasi-singular compartment by dropping the now negligibly small first term from Equation 8-25, and rewriting the equation for concentration as suggested by Equation 8-28:

$$C_{(A+B).t} = (Q_{tot}/V_A)\left(\frac{Z + k_B - k_A}{2Z}\right)\exp\left[-\tfrac{1}{2}(k_A + k_B - Z)t\right] \quad (8\text{-}49)$$

By Equation 8-49 the concentration assumed to be present in the "$A + B$" compartment at time zero will be

$$C_{(A+B).0} = (Q_{tot}/V_A)\left(\frac{Z + k_B - k_A}{2Z}\right) \quad (8\text{-}50)$$

This concentration can be estimated by plotting the observed concentration in Compartment A against time on semilogarithmic graph paper, and extrapolating the straight line for the second exponential back to its intercept at time zero. Then by the relation given in Equation 8-32,

$$V_{(A+B)} = (V\text{dist})_{\text{intercept}} = Q_{tot}/C_{(A+B).0} \quad (8\text{-}51)$$

where

$(V\text{dist})_{\text{intercept}}$ = the volume of distribution estimated from the *intercept* at time zero

Since both Q_{tot} and $C_{(A+B).0}$ are known, $(V\text{dist})_{\text{intercept}}$ can be calculated.

It is important for us to know how much this estimate differs from the true volume of distribution (Vdist) which, by Equation 8-45, is $V_A(k_{A\to B} + k_{B\to A})/k_{B\to A}$. Combining Equations 8-50 and 8-51 with the elimination of $C_{(A+B).0}$,

$$(V\text{dist})_{\text{intercept}} = 2ZV_A/(Z + k_B - k_A) \tag{8-52}$$

The ratio of $(V\text{dist})_{\text{intercept}}$ to $(V\text{dist})$ is

$$\frac{(V\text{dist})_{\text{intercept}}}{V\text{dist}} = \frac{2Zk_{B\to A}}{(Z + k_B - k_A)(k_{A\to B} + k_{B\to A})} \tag{8-53}$$

If $k_{A\to U}$ is so very small that we can assume $k_A = k_{A\to B}$, this ratio reduces to unity, *i.e.*, $(V\text{dist})_{\text{intercept}}$ equals the true $(V\text{dist})$. But when $k_{A\to U}$ amounts to an appreciable fraction of $k_{A\to B}$, the ratio exceeds unity. $(V\text{dist})_{\text{intercept}}$ then *overestimates the true volume of distribution by an amount which is impossible to calculate unless we already know all of the rate constants* characterizing the system! But if we know the rate constants we should use Equation 8-45. In Figure 8-8, a particular set of physiologically reasonable values has been used to illustrate how preposterously large $(V\text{dist})_{\text{intercept}}$ can be when the renal plasma clearance is even moderately rapid.

A second method, which also overestimates $(V\text{dist})$ when $k_{A\to U}$ is appreciably large, depends upon the relationship expressed by Equation 8-5; namely, that the rate constant for a given pathway is equal to the clearance by that pathway divided by the volume of the compartment being cleared. At present we are assuming that $(A + B)$ behaves as a single compartment being cleared by a single pathway. Hence,

$$k_{(A+B)\to U} = (\dot{V}\text{cl})/(V\text{dist})_{\text{slope}} \tag{8-54}$$

or,

$$(V\text{dist})_{\text{slope}} = (\dot{V}\text{cl})/k_{(A+B)\to U}$$

where

$(V\text{dist})_{\text{slope}}$ = the volume of distribution calculated from the rate constant for loss of S in urine and from the renal clearance of S

$k_{(A+B)\to U}$ = the rate constant for the disappearance of S from $(A + B)$ estimated from the *slope* of the straight line for the second exponential obtained when $C_{A.t}$ is plotted on semilog graph paper against time

Here, too, $(V\text{dist})_{\text{slope}}$ is most nearly correct when $k_{A\to U}$ is very small. Unfortunately, when $k_{A\to U}$ is negligibly small, so also is $k_{(A+B)\to U}$! Hence,

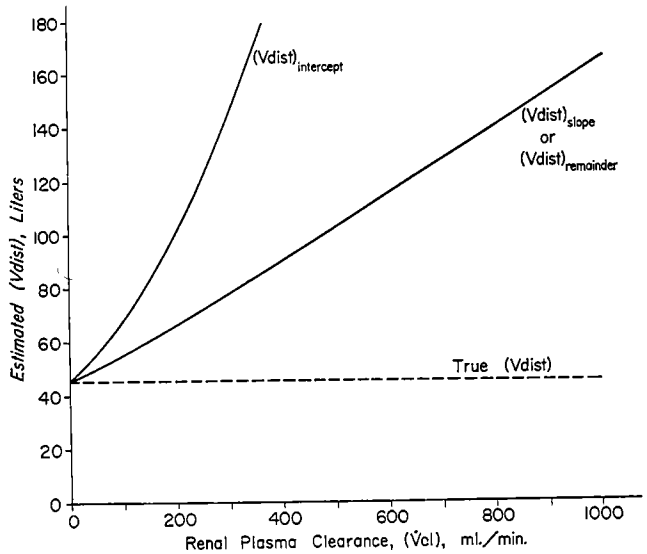

FIG. 8-8. Biased Estimates of the Volume of Distribution

The three biased estimates described in the text have been calculated for various rates of renal clearance with the following parameters for the model system of Figure 8-5: V_A = 15 L., V_B = 30 L., $k_{A\to B}$ = 0.9 per hour, $k_{B\to A}$ = 0.45 per hour. Except when the renal plasma clearance is very small (so that the two compartments behave practically as a single compartment after distribution between them is complete) all three methods grossly overestimate the true volume of distribution, estimation from the intercept (Equation 8-51) being particularly misleading (see also Figs. 8-9 and 8-10).

even when it is possible to measure a small renal plasma clearance accurately, it will probably not be possible to measure $k_{(A+B)\to U}$ with any precision. The method therefore suffers from the curious defect of being least accurate when least biased. This is too bad because $(V\text{dist})_{\text{slope}}$ is a less biased estimate of the true volume of distribution than is $(V\text{dist})_{\text{intercept}}$ (Fig. 8-8).

A third method of approximation yields results which are theoretically (*i.e.*, aside from errors of measurement) identical with the results given by the previous method. Suppose that at some time, t, the first exponential of Equation 8-25 has become negligibly small. Then

$$(V\text{dist})_{\text{remainder}} = (Q_{\text{tot}} - Q_{U.t})/C_{A.t} \tag{8-55}$$

where

$(V\text{dist})_{\text{remainder}}$ = the volume of distribution calculated from the quantity of S *remaining* in Compartments A and B at time t

$Q_{U.t}$ = the quantity of S which has been excreted between time zero and time t

Since this method does not depend upon estimating a very small slope, it is likely to give much more accurate results than the previous method, and should be, in practice, the least objectionable of the three approximate methods described above. But none of these methods has much to recommend it, and, in the author's opinion, there is little justification for regarding the volumes so calculated as equivalent to true volumes of distribution unless the final rate of decrease of the concentration of S in plasma is very small. The fallacy of treating two separate compartments as a single compartment when loss of S is rapid is further illustrated by Figures 8-9 and 8-10 which are worth careful study.

8-13. After Intravenous Injection, Why Is Not the Initial Volume of Distribution Always Equal to the Plasma Volume?

When a single dose of S is injected rapidly into a vein, surely the first "compartment" which it enters is the blood plasma. If samples of blood are taken early enough, and frequently enough, and the concentration of S in plasma is plotted against time, should not the intercept of the curve at time zero always indicate plasma volume? The answer to this perfectly logical question is simply that for most small molecules the blood plasma does not behave like a separate compartment. Mixing in plasma is not instantaneous. Indeed, when a dye which binds almost completely to plasma protein is injected intravenously it appears as a "hump" of concentration in the arterial blood, thus providing one method of measuring cardiac output (Section 9-5). There are often smaller subsequent "humps" during the first few recirculations of blood before mixing is complete. Moreover, with small molecules, filtration and diffusion out of capillary beds into the surrounding tissue fluids is extremely rapid (83), so that by the time concentration in plasma has become reasonably uniform, the solute has already penetrated into a much larger volume. It is this larger volume, including blood plasma, which constitutes the apparent initial volume of distribution for many small solutes such as creatinine. Only with large molecules, or substances firmly bound to large molecules, can one identify a separate plasma compartment by analyzing the early part of the curve of plasma concentration *versus* time. But since both molecular size and extent of binding to plasma protein vary from substance to substance over very wide ranges, we must expect to find some intermediate

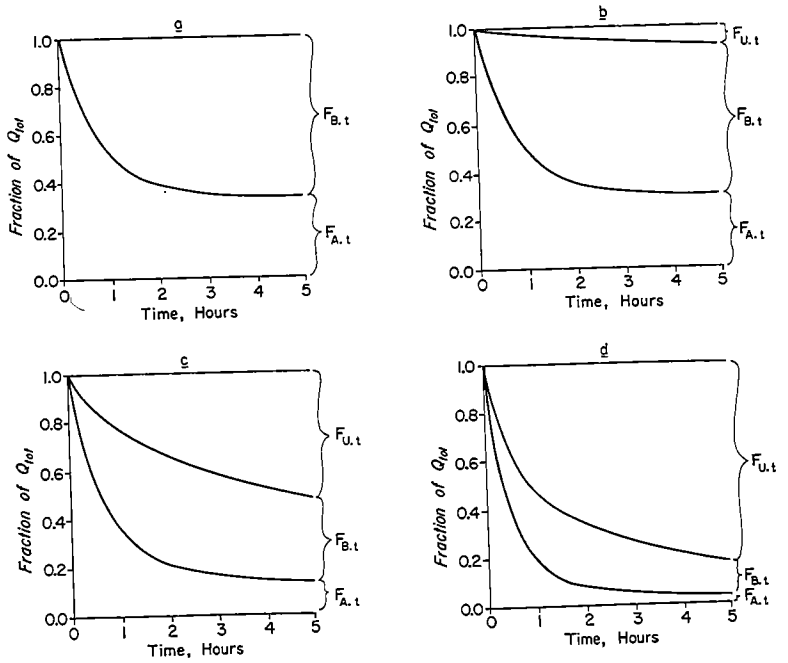

FIG. 8-9. How the Distribution of Solute between Two Compartments Is Influenced by Rate of Clearance

At time zero, Q_{tot} of solute was injected as a single dose into Compartment A of the system of Figure 8-5 with the same parameters as for Figure 8-8. The fractions of this total quantity remaining in Compartment A, $F_{A.t}$, remaining in Compartment B, $F_{B.t}$, or already excreted in the urine, $F_{U.t}$, are plotted as functions of time for clearances of 0 ml. per minute (Graph a), 10 ml. per minute (Graph b), 100 ml. per minute (Graph c) and 300 ml. per minute (Graph d).

With zero clearance (Graph a), the system is, in fact, a closed two-compartment system approaching a steady state of equilibrium with $F_{A.t} = \frac{1}{3}$ and $F_{B.t} = \frac{2}{3}$ at a proportional rate of $k_{A\to B} + k_{B\to A} = 1.35$ per hour, corresponding to a half-time of 0.513 hr. Equilibrium has been achieved, for all practical purposes, by the end of 4 or 5 hr. With a clearance of only 10 ml. per minute, Compartments A and B still come close to equilibrium with each other (Graph b), but with a clearance of 100 ml. per minute (Graph c) or 300 ml per minute (Graph d) a steady state of diffusion equilibrium between A and B is never approached. For example, when the rate of clearance is 300 ml. per minute, less than $\frac{1}{6}$ of the total solute remaining in the body at 5 hr. is present in Compartment A instead of the $\frac{1}{3}$ which would be present at distribution equilibrium (see also Fig. 8-10).

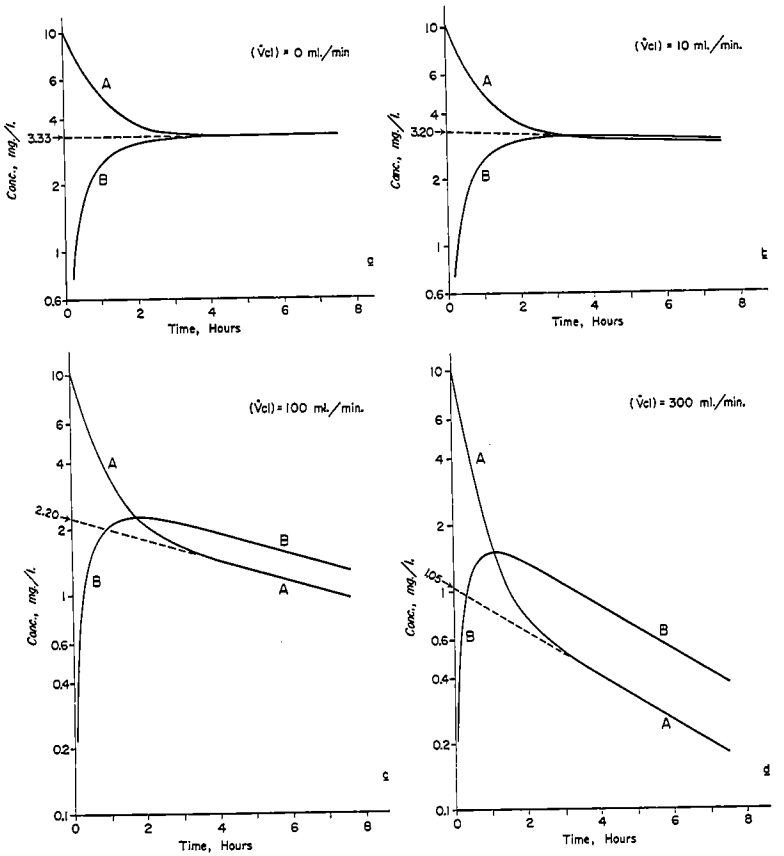

FIG. 8-10. How the Concentration of Solute in the Two Compartments of Figure 8-5 Is Influenced by Rate of Clearance

The parameters of the system are the ones listed in the legend for Figure 8-8 and also assumed for Figure 8-9. A single dose of 150 mg. of solute was placed in Compartment A at time zero. The four rates of clearance are the same as for Figure 8-9. With zero clearance (*top left*) the concentration in A falls, and the concentration in B rises, so that both approach the same equilibrium concentration, 3.33 mg. per liter, with a half-time of 0.513 hr. The true volume of distribution is thus equal to the total dose, 150 mg., divided by the equilibrium concentration, 3.33 mg. per liter, *i.e.*, 45 L. But when solute is being cleared from Compartment A at an appreciable rate, a steady state of equilibrium between A and B is not approached with the passage of time after a single dose. Notice that the concentration in B keeps increasing as long as there is a concentration gradient for diffusion of solute from A to B, *i.e.*, as long as the concentration in A exceeds the concentration in B. Notice also that at some instant of time A and B are momentarily in equilibrium with each other

compounds whose apparent initial volume of distribution corresponds neither to plasma volume, nor to "extracellular fluid volume," nor to any other recognizable entity. Indeed the greatest circumspection must be exercised in attempting to identify the volume of distribution of any substance with a particular body fluid.

An extensive table of volumes of distribution is given by Dominguez (33).

EXERCISES. CHAPTER 8

Exercise 1

In each of the following problems, write a differential equation for the rate of change of the quantity of X in Compartment A, dQ_A/dt. Then solve, *i.e.*, integrate, this equation so as to obtain an explicit expression for $Q_{A,t}$, the quantity of X in A at any time t. You will find it helpful to draw a diagram of compartments and pathways for each problem. You should also check each equation for $Q_{A,t}$ by seeing whether it makes sense when $t = 0$ and as $t \to \infty$.

A. Loss of X at a constant absolute rate of $\dot{Q}_{A\to out}$ mg. per minute from a single compartment, A.

B. Loss of X at a constant proportional rate of $k_{A\to out}$ per minute from a single compartment, A.

C. Continuous infusion of X into A at a constant rate $\dot{Q}_{in\to A}$ starting at time zero (no X present in A at time zero).

D. Same as "C" above but with the addition of a pathway for loss of X from A into the urine at a constant proportional rate, $k_{A\to U}$.

E. Same as "D" above, but with the addition of another route of loss at a constant proportional rate, $k_{A\to liv}$, representing metabolic transformation of X in the liver.

F. Same as "D" above, but with the addition of another route of loss at a constant absolute rate, $\dot{Q}_{A\to out}$.

(equal concentrations) so that there is no net diffusion between them. It is precisely at this point that the concentration in B achieves its maximum. (See the discussion of precursor-product relationships in Section 9-8.) As clearance of solute from Compartment A continues, the diffusion gradient is reversed, so that the concentration in B, though now decreasing, becomes, and remains, higher than in A. Instead of approaching a steady state of equilibrium, A and B approach a state in which their concentrations decline at the same exponential rate (*parallel straight lines* on the semi-log plot of this figure). But as the clearance increases, the intercept of the straight line for Compartment A on the time-zero axis becomes more and more misleading as an index of the volume of distribution. For example, when the clearance is 300 ml. per minute (*lower right*) the "volume of distribution" calculated from the intercept for Compartment A at time zero is 150/1.05 = 143 L. whereas the true value is only 45 L. (Fig. 8-8).

G. At time zero, Q_{tot} of X is present in Compartment B, and no X is in Compartment A. X is transferred *irreversibly* from B to A at a constant proportional rate, $k_{B \to A}$.

Exercise 2

In Section 8-7, it was assumed that the initial behavior of a tracer dose of radioactive iodide (I^{131}) given to a normal subject could be analyzed according to the model shown in Figure 8-2. Accepting this assumption, estimate from the data tabulated below:

A. The half-time for disappearance of iodide from the plasma.
B. The proportion of the tracer dose ultimately accumulated by the thyroid gland.
C. The plasma clearance of iodide by the thyroid gland if the renal plasma clearance was 34 ml. per minute.

(Time zero is the time of administration of the tracer dose of I^{131}.)

Urine Sample Collected between			Per Cent of Tracer Dose in Sample	Cumulative Per Cent
0.0	and	2.13 hr.	17.9	17.9
2.13		4.22	13.5	31.4
4.22		6.25	11.1	42.5
6.25		8.13	7.7	50.2
8.13		24.00	31.6	81.8
24.00		48.00	8.3	90.1

Exercise 3

Consider a closed system consisting of the two compartments, A and B. At time zero, Q_{tot} mg. of S was instantaneously dissolved in Compartment A. The quantity of S in A, $Q_{A.t}$, decreased with time as indicated below:

Time (min.)	10	20	30	40	50	70
$Q_{A.t}$ (mg.)	59	43	35	29	25	22

From these data, estimate

Q_{tot}, $Q_{A.\infty}$, $Q_{B.\infty}$, $k_{A \to B}$, $k_{B \to A}$, and

the half-time for equilibration.

Exercise 4. Accumulation of a drug given repeatedly

Suppose that a single intravenous dose, Q_D, of a drug is instantaneously distributed throughout a single compartment, producing an immediate

peak plasma concentration of C_0. After t hours, this concentration has declined exponentially to C_t. Let F be the fraction remaining at time t, so that (assuming the volume of the compartment remains constant) $F = C_t/C_0$. What will the *maximum* peak plasma concentration just after a single dose ultimately be if the same single dose is administered every t hours for a very long (theoretically, infinite) time? Express the ma peak concentration, C_{max}, in terms of C_0 and F.

Exercise 5

A substance not metabolized in the body is infused at a consta of 85 mg. per minute. The amount of the substance excreted in th per minute at various times after starting the continuous infusion i by the following tabulation:

Time of Infusion *hr.*	Urinary Excretion *mg./min.*
0	0
0.5	25
1.0	40
1.5	41
2.0	46
2.5	52.7
3.0	56.8
3.5	56.5
4.0	58.5

At 4.0 hr. the blood plasma contained 83.6 mg. per cent of the subs

Estimate: the biological half-life of the substance.
the volume of distribution of the substance.
the renal plasma clearance of the substance.
the extrarenal plasma clearance of the substance.

Exercise 6

Dominguez *et al.* (34) studied the fate of exogenous creatinine in the dog. The following data are taken from an experiment in which 6.66 gm. of creatinine were injected intravenously into a 20.2-kg. dog at time zero, and the concentration of creatinine in plasma was subsequently determined. Evidence was obtained that practically all of the injected creatinine was excreted unchanged by the kidneys. The data have been corrected for the small amount of endogenous creatinine present, so that the values given are for the exogenous creatinine only.

Time of Blood Collection (t)	Plasma Creatinine ($C_{A.t}$)
min.	*mg./100 ml.*
12	81.2
17½	70.2
34	42.2
47	41.1
61½	31.6
92½	24.2
122½	20.5
182½	16.9
242	12.5
302	8.6
363½	7.7
422½	6.6
484½	5.0

Assume that these data are in accord with the two-compartment model depicted in Figure 8-5, and that they may therefore be fitted by a double exponential of the general form:

$$C_{A.t} = G_1 e^{-k_1 t} + G_2 e^{-k_2 t}$$

where

$C_{A.t}$ = the concentration of creatinine in plasma at time t

G_1, G_2, k_1 and k_2 are parameters to be estimated from the data.

A. Estimate the parameters of the double exponential equation.

B. Estimate $k_{A\to B}$, $k_{A\to U}$, $k_{B\to A}$, V_A, $(V\text{dist})_{B_A}$, $(V\text{dist})$, and $(\dot{V}\text{cl})$.

Exercise 7

Prove that when both $k_{A\to\text{out}}$ and $k_{B\to\text{out}}$ are zero, so that the two-compartment system of Figure 8-4 is closed, Equation 8-25 (multiplied by Q_{tot} to convert it to an equation for the quantity of solute in A at time t) reduces to Equation 8-23.

Exercise 8

Prove that Equation 7-52 for equilibration by diffusion in a closed two-compartment system is a special case of Equation 8-23, the general equation for equilibration in a closed two-compartment system.

9

FURTHER KINETIC PROBLEMS. FLUID FLOW, METABOLIC TRANSFORMATIONS

9-1. Transport by Fluid Flow

In a number of important problems a solute is carried into or out of a compartment by an actual flow of fluid through the compartment. Provided the underlying assumptions remain valid, the equations previously developed for the kinetics of distribution can be applied equally well to problems of actual flow merely by substituting flow, $\dot{V}$, for clearance, $(\dot{V}\text{cl})$. For example, Equation 8-5 which gives the fundamental relationship between rate constant, compartment volume, and clearance becomes

$$k_{W\to X} = \dot{V}_{W\to X}/V_W \tag{9-1}$$

where

$\dot{V}_{W\to X}$ = the flow (*i.e.*, volume of fluid per unit time) from W to X

Briefly, the underlying assumptions are that the compartment volumes remain constant, that mixing within each compartment is instantaneous, and that the clearance, now the actual fluid flow, by each pathway remains constant. In addition, we have usually assumed hitherto that a single dose, Q_{tot}, of a solute, S, was introduced at time zero into Compartment W. We shall now see that by restricting consideration to a single compartment we can deal with certain problems in which the concentration of S in the inflow is variable and in which mixing is not necessarily instantaneous. However, we shall retain the assumptions that the compartment volumes and the flows are constant.

In the majority of physiological problems, the fluid which is flowing is blood. Very often the objective is to estimate blood flow from observations of how the concentration of some solute in the blood going to and coming from a particular region varies with time as the region adds solute to or removes solute from the bloodstream. The solution of such problems de-