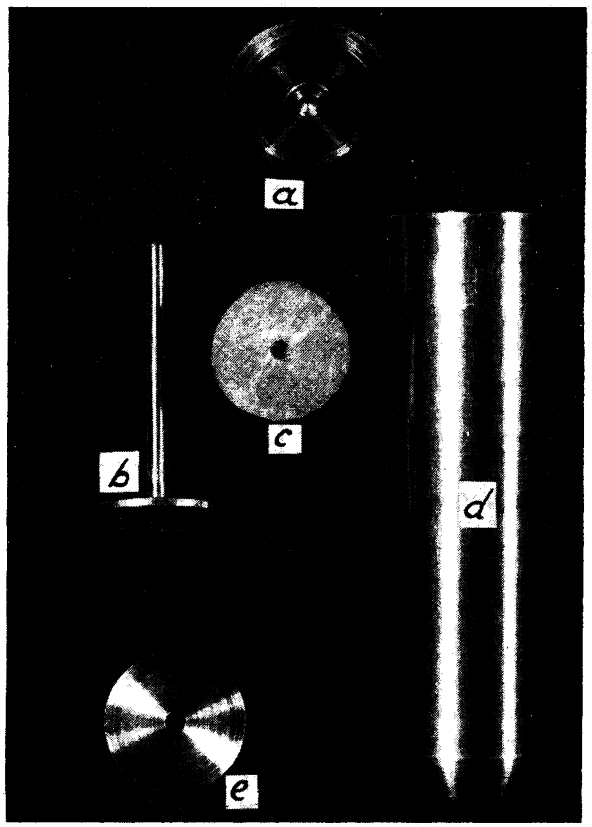

FIG. 2. Centrifugal tissue mincer disassembled.

mincing, screening, and centrifuging in a single step requiring less than 10 min, instead of more than 1 hr as under the older method. It fits the rotor of a Sorvall SS-1 superspeed angle centrifuge and is operated at 13,000 rpm at an R.C.F. of about 20,000 × G. The stainless steel construction allows dry heat sterilization and is not objectionable on chemical grounds. Since the rotor carries 8 tubes, 4 properly balanced mincing units may be run at the same time.

In practice, the tube (Fig. 2, *d*) containing only the stemmed screen (Fig. 2, *b*), is dry-sterilized in a cotton-plugged Pyrex tube; cap and piston (Fig. 2, *a* and *e*) are sterilized in a deep Petri dish. The device may be chilled before loading 4 chick embryos (or other tissues to be minced) into space *A*. The piston is picked up with sterile forceps at its upper end and slipped over the stem into the tube, which is then capped. After critical counterbalancing with a steel tube containing finest birdshot in the opposite tube-depression of the previously chilled rotor, the centrifuge is gradually brought to top speed, and shut off after 3 to 4 min at 13,000 rpm. The mincing tube is then removed, opened under sterile precautions, and with a pair of flamed pliers the screen-piston assembly is grasped by the stem and pulled out slowly to avoid a vacuum.[1] Space *B* now contains a steep bank of tightly packed dissociated tissue and about 4 ml of clear, concentrated embryo juice. This amount is more than double the yield per embryo that may be expected after mincing with curved scissors.

The dimensions in inches of the unit shown in Figs. 1 and 2 are: length without cap, 3.895; OD, 1.125; ID, space *A*, 0.939; ID, space *B*, 0.760; distance from top to shelf, 2.000; the piston diam, 0.938; piston height, 0.750; screen thickness, 0.060; screen diam, 0.938; screen stem diam, 0.125; hole through piston to accommodate stem, 0.126; cap thickness, 0.160; thickness of cap lip, 0.042; diam of cap recess, 0.937. Holes in screen are drilled with a No. 60 to 69 drill and closely spaced in concentric circles.

A disk of stainless steel mesh (Fig. 2, *c*), 80–90 wires/inch (gauge #37 B & S), with openings measuring 150–165 μ square, may be inserted above the screen for very fine mincing. This is not recommended for making embryo extract, but is useful in preparing uniform tumor suspensions.

[1] An improvement in design, since this description has gone to press, eliminates formation of a vacuum by replacing the stem (Fig. 1) with stainless tubing which opens into space *B* below the screen disk.

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A Simple Procedure for Determination of the Approximate Lymph Space[1]

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In the course of studies of permeability changes caused by irradiation it was discovered that whatever macromolecular or corpuscular substance was injected intravenously, it disappeared faster from the circulation of x-rayed animals than from that of normals (*1*). Subsequent studies led to the observation that the substance which disappeared from the blood entered the tissue space (*2*). In order to determine the magnitude of this change, it became essential to measure the space.[3, 4]

Data on the tissue space are few and discrepant (*3*). The main difficulty in the estimation of the magnitude of this space is caused by differences in composition of the lymph in different parts of the

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[2] The authors gratefully acknowledge the constructive criticism of this manuscript by W. S. Wilde, C. W. Sheppard, A. W. Kimball, and A. S. Householder and the help of J. Z. Hearon and W. S. Wilde in the formulation of equations (*a*) and (*b*).
[3] The term "lymph" is used in the broad sense, including the fluid in the interstitial spaces and lymphatics. These two spaces combined are conventionally called tissue space.
[4] The term "space" is used instead of volume for theoretical reasons, but it is certain that the values for the two are close if not identical.

body (*4*), great variations in the composition of the lymph with the rate of flow, and other factors (*3*).

When homologous albumin tagged with I^{131} is introduced into the circulation, some of it leaves the blood stream and enters the lymph as illustrated in Fig. 1.

The average rate of disappearance of the tagged albumin from the blood is about 26%/hr during the first half hour, 19%/hr during the second half hour, 6%/hr during the second hour, and slower thereafter[5] (*5*) until equilibrium is reached between blood and lymph (*6,7*). An equation for this disappearance rate has been formulated by Wasserman and Mayerson (*6*). Equilibration time is variable, but it is nearly complete at 5–12 hr after injection. The measures of the radioactivity of lymph and blood plasma after equilibration time parallel the albumin concentration of fluid in these compartments. The decline in radioactivity after about 24 hr is parallel in the two compartments and due largely to metabolic destruction of albumin.

After correction for loss due to metabolism the total amount of activity (tagged albumin) which disappears from the circulation during equilibration is proportional to the mass of albumin in the lymph.

The blood plasma volume is measured from the plasma radioactivity about 5 min after injection:

$$V_p = \frac{R^*}{R_p^{(o)}},$$

where V_p = plasma volume in ml; R^* = total radioactivity injected; and $R_p^{(o)}$ = radioactivity per ml of plasma measured 5–10 min after injection.

In calculating the tissue space, equations (*a*) and (*b*) were derived (*8*). These equations are similar to those of Gellhorn, Merrell, and Rankin (*9*). (For an extensive mathematical analysis of the problem in general, see Sheppard and Householder [*10*] and the review by Solomon [*11*]).

The transfer equations between plasma and lymph may be formulated as follows (*8*):

$$(a)\quad \frac{dR_p^{(t)}}{dt} = \frac{hA}{V_p}\left(\frac{R_l^{(t)}}{C_l} - \frac{R_p^{(t)}}{C_p}\right) - k_p R_p^{(t)}$$

$$(b)\quad \frac{dR_l^{(t)}}{dt} = \frac{hA}{V_l}\left(\frac{R_p^{(t)}}{C_p} - \frac{R_l^{(t)}}{C_l}\right) - k_l R_l^{(t)}$$

where $R_p^{(t)}$ = radioactivity/ml plasma measured at time, t; $R_l^{(t)}$ = radioactivity/ml of lymph measured at time, t; C_p = concentration of albumin in plasma; C_l = concentration of albumin in lymph; h = interchange between the two compartments; A = common area of the two compartments; V_p = volume of the plasma; V_l = volume of the lymph; k_p = exponential rate of plasma albumin metabolism; and k_l = exponential rate of lymph albumin metabolism.

These equations could be expressed more explicitly in terms of specific activities, mean exchange rates and albumin mass (*10*). Since the present study is concerned with volume and reliance is placed more on radioisotopic than on chemical determinations, we prefer the above formulation. In any event the final special equation (*c*), used for calculation, would not be affected. No correction for radioactive decay is necessary since all readings were taken at the same time. All equations are formulated for first approximation for an idealized two-compartment system under the conditions of a dynamic equilibrium. The number, sites, and character of the subcompartments remain to be identified.

[5] These are averages on a total of 8–11 supposedly normal dogs studied.

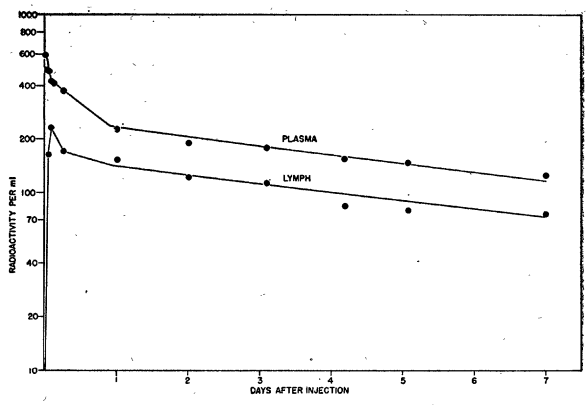

FIG. 1. Radioactivity (cps/ml) of plasma and lymph of a dog after intravenous injection of I^{131}-labeled canine albumin.

Assuming that the volumes of blood and lymph remained constant with time and that the metabolism rates in the two compartments were equal, the two differential equations may be reduced to the following form:

$$(c)\quad R^* e^{-kt} = V_p R_p^{(t)} + V_l R_l^{(t)},$$

where $k_l = k_p = k$, and R^* is the total injected radioactivity. This equation holds true after there is uniform mixing of the tagged albumin in and between lymph and blood plasma.

In order to determine the tissue space, measurements are made of the radioactivity per ml in each compartment any time after equilibrium has been established. The total activity injected and the plasma volume are known. This leaves two unknowns in equation (*c*): the metabolism rate and the tissue space. The former can be estimated from the slope of the linear portions of the activity curves (Fig. 1). If τ_1 and τ_2 are two times after equilibrium, we may estimate k as follows:

$$-k = \frac{\ln R_p^{(\tau_1)} - \ln R_p^{(\tau_2)}}{\tau_1 - \tau_2}.$$

We may then solve equation (*c*) for the remaining unknown V_l.

The accuracy of the value obtained depends mainly on how uniform the albumin concentration is in the various lymphatic channels and interstitial fluids. Equality or closeness of k_p and k_l is a reasonable assumption; in any event the values in question are small. The ratio of lymph/blood albumin concentration is variable from dog to dog but not to a great extent. In normal dogs examined by us the ratio of albumin concentration in the thoracic lymph to that in

the plasma ranged between 0.5 to 0.7. If the lymph radioactivity is not measured, $R_l^{(t)}$ can be considered as being approximately 0.6 $R_p^{(t)}$, for periods after equilibration, and accordingly the tissue space can be estimated in normal animals without tapping lymphatics, with an accuracy of about ±20%.

There are numerous reports in the literature (reviewed by Drinker [3]) on the albumin concentration of the lymph in different parts of the body, and the Bollman (12) type of cannulation enables a crude estimation of the relative quantity of lymph in different parts of the body.

The tissue space of any part of the body can also be estimated as follows: The plasma volume of an organ or given part of the body is determined by injection of I^{131}-tagged albumin, and the organ activity is measured after the mixing time of 5–10 min (13). Similar determinations of organ activity are made after equilibration. These measurements may be made with ease with the aid of the gamma ionization chamber (14). The organ activity after equilibration is due to the combined albumin activity in the blood and tissue space of that organ. Thus, the following values are obtained: (*a*) plasma volume, (*b*) organ plasma volume, (*c*) combined organ radioactivity in plasma plus lymph, and (*d*) activity per unit volume of plasma. The unknown is (*e*), activity per unit volume of lymph, which is a function of the albumin concentration of the lymph. The protein concentration of the lymph of different organs or parts of the body is known in the literature (3). Using these values, the tissue space (volume) can be calculated in a manner similar to that outlined above.

A more precise determination can be made by actually measuring the lymph activity and by remeasuring blood plasma and organ plasma volumes after equilibration with a different isotope—e.g., C^{14}.

These considerations probably apply to all mammals. Under pathological conditions resulting in increased permeability, the ratio of albumin concentration in the lymph to that in the plasma may approach unity, thus setting an upper limit for the lymph space. In our experiments the lowest conversion factor (ratio of albumin concentration in lymph to plasma) was 0.5. It is possible that with some pathological states the conversion factor is even lower.

At present I^{131}-tagged albumin (15) is the best for this purpose. Albumin naturally labeled with C^{14} would be preferable.

The approximate tissue space (volume of lymph) can be estimated by measuring the quantity of labeled albumin per unit volume of blood plasma after mixing time, and of blood and lymph after equilibration time. After allowance for metabolism the loss in blood activity during equilibration time is a function of the albumin mass in the lymph. In normal dogs and some other species, a conversion factor of 0.6 can be used if lymph activity is not measured. The loss in plasma activity multiplied by this conversion value (ratio of albumin concentration between lymph and plasma) is proportional to the tissue space (lymph volume).

Mathematically this may be expressed as follows:

$$R^* e^{-kt} = R_p^{(t)} (V_p + 0.6 V_l).$$

The conversion factor is raised with heightened permeability, the theoretical maximum being unity. This simple procedure enables approximation of the lymph volume without sampling lymph.

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A Multirange Recording and Control System for Electrical Measurements

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In studying the effect of nuclear reactor irradiation on the electrical properties of semiconductors, it is desirable to record automatically the temperature and resistivity during irradiation as a function of exposure time. One records (1) the output voltage of a thermocouple, (2) the potential drop across a fixed portion of the sample, and (3) the current as determined by the potential drop across a standard resistor.

The change in resistance of the sample under bombardment may be of the order of a few per cent over time periods of several days, as in the case of low resistivity P-type Ge, or it may be several orders of magnitude in a few hours as in the case of Si and Cu_2O: thus resistances from 0.001 ohm to 100 megohms have to be measured and recorded directly to within ±0.1% accuracy. The temperature may vary from −78° C to 50° C in a single experiment. In addition, it is frequently desirable to expose two or more samples simultaneously for comparison purposes. Some samples—e. g., rectifiers—are non-ohmic, in which case current or voltage has to be held constant at predetermined values.

A device fulfilling these requirements has been constructed and is now in use at this laboratory. The in-