Clinical Workshop

S. G. HERSHEY, M.D., *Editor*

A Safer Method for Measuring Body Fluid Compartments in Patients

DAVID R. COOK, M.D.,* STEPHEN J. GALLA, M.D.,†
WILLIAM S. GUALTIERE, PH.D.‡

In the management of critically-ill patients, measurements of body fluid compartments and correction of volume aberrations can be essential. Measurement of these compartments is based on the principle of tracer dilution. Various groups[1-3] have described isotope dilution techniques for simultaneous measurement of plasma volume, extracellular fluid volume, and erythrocyte mass, using ^{131}iodine, ^{35}sulfur, and ^{51}chromium, respectively. Moore[4] and others[5] have measured total body water with tritium. Plasma volume alone has been estimated accurately with Evan's blue dye.[6]

Our interest in measuring fluid compartments resulted from our concern with the unnecessarily high doses of radioactivity currently used: ^{131}iodine, 10 microcuries; ^{35}sulfur, 70–100 microcuries; tritium, 1.0 millicuries; ^{51}chromium, 25 microcuries. Although this represents a total body radiation dose of approximately 0.402 rems, the testicular and thyroid exposure from the radioactive sulfur and iodine is 2.925 rems.[7] Because liquid scintillation spectrometry has provided accuracy and increased sensitivity in the measurement of the weak beta emitters, we chose to evaluate the measurement of fluid compartments at considerably reduced activity levels.

METHODS

Experimental Procedure. Thirty-eight male college athletes, 18–23 years of age, attending the University of Pittsburgh, volunteered for the study. After they and their parents were informed of the nature, purpose, and potential risks involved, the written consent of both was obtained. The subjects maintained their usual dietary habits on the day preceding the test and avoided strenuous physical activity. On the morning of the study, in the fasting state, they remained supine for 30 minutes prior to administration of the tracer. Evan's blue dye, 25 mg, sodium sulfate-^{35}S, 10 microcuries, and tritiated water, 0.5 millicurie, were injected intravenously. Postinjection blood samples were obtained from the opposite arm at 15, 20, 25, 120, and 180 minutes.

Analytical Methods. Blood samples collected 20 minutes postinjection were used for determination of Evan's blue dye concentration and microhematocrit. The plasma level of Evan's blue dye was measured spectrophotometrically at 620 mμ. Blanks and standard solutions were prepared using plasma collected from a preinjection sample. An 0.5-ml aliquot of plasma was dissolved in a 15-ml mixture of dioxane–0.7 per cent PPO–0.007 per cent dimethyl-POPOP. All samples were counted for 10,000 cpm or 100 minutes by liquid scintillation spectrometry using a double-labeling technique. Separate channels were used for

* Teaching Fellow in Anesthesiology.
† Associate Research Professor of Anesthesiology.
‡ Assistant Professor of Education.
Received from the Departments of Anesthesiology and Physical Education, Schools of Medicine and Education, University of Pittsburgh, Pittsburgh, Pennsylvania 15213. Supported in part by Grant GM 13965, National Institutes of Health, Bethesda, Maryland, and Grant RT-14, Social and Rehabilitation Service, Department of Health, Education and Welfare, Washington, D. C. Presented at the Annual Meeting of the American Society of Anesthesiologists, Washington, D. C., October 23, 1968.

TABLE 1. Calculation of Body Fluid Compartments

Plasma volume (ml)	$= \frac{\text{Evan's blue dye injected, mg}}{\text{Evan's blue dye in plasma, mg/ml}}$
Blood volume (ml)	$= \frac{\text{Plasma volume, ml} \times 100}{(100 - \text{per cent hematocrit} \times 0.91)}$
Total body water (ml)	$= \frac{\text{Tritium injected, dpm}}{\text{Tritium in plasma, dpm/ml}} \times \text{PWC}^*$
Extracellular fluid (ml)	$= \frac{^{35}\text{sulfur injected, dpm} \times \text{PWC} \times \text{GD}^\dagger}{^{35}\text{sulfur in plasma, dpm/ml}}$
Intracellular water	= Total body water − extracellular water

* PWC = Plasma water concentration.
† GD = Gibbs–Donnan equilibrium constant.

counting ^{35}sulfur and tritium, and a third channel was used for external ^{226}radium standard to correct for quenching.

Calculations. Sample calculations are shown in table 1. The Gibbs–Donnan equilibrium constant was calculated to be 0.91 and referred to the total volume of sulfate distribution.[8] Since the plasma disappearance curve of ^{35}sulfur reflects the loss of isotope into the urine and intracellular space, the zero time extrapolation was used in the calculations. Tritium activity at three hours was used as the plasma equilibration value. All data were corrected to reflect plasma water content, which was measured at 91 per cent.

RESULTS

Optimal counting efficiency for tritium and ^{35}sulfur was obtained when the ratio of their plasma counts was approximately 20:1. This was accomplished by adjusting the ratio of their activities in the dose to approximately 50:1. Table 2 compares our results with those obtained by Moore and his associates.[4]

DISCUSSION

Our reasons for using college athletes to validate our methods were twofold: (1) an investigator in the Department of Physical Education was interested in making these measurements on various athletic groups; and (2) athletes constituted a healthy and relatively homogeneous group.

Numerous investigators[9-13] have made simultaneous measurements of plasma volume in man using Evan's blue dye and radioiodinated serum albumin (RISA). When the plasma activity of RISA was corrected for losses in glassware, the correlation between the two methods was good,[12] recognizing that different characteristics of the plasma volume were measured.[9, 13] The plasma volume–hematocrit method has been shown to reflect total blood volume measurements accurately when large-vessel macrohematocrit is corrected for trapped plasma. Since the amount of plasma trapped by the microhematocrit technique is

TABLE 2. Comparison of Measurements of Human Body Fluid Compartments Obtained by Various Investigators*

	Moore, F. D., *et al.*	Present Authors
Plasma volume (per cent of body weight)	4.5	5.3
Blood volume (per cent of body weight)	6.8	9.0
Extracellular fluid space (per cent of body weight)	23.9	22.8
Total body water (per cent of body weight)	58.9†	57.7
Intracellular water / Total body water	0.59	0.60

* All numbers are mean values. Standard errors did not exceed 3.2 per cent of mean.
† Age group 16–30 years.

negligible,[14] our data were not corrected. Venous hematocrit was corrected to whole-body hematocrit by multiplying by 0.91.[15]

As seen in table 2, our results for men in the 18–23-year age group compare favorably with Moore's data.[4] Variations of our values for plasma and blood volume from Moore's are probably due to differences between the populations under investigation.[16] Our subjects were predominantly highly-trained college athletes, studied at the peak of their training season, whereas Moore's were not.

Although the dose necessary for teratogenicity has not been determined, ^{35}sulfate concentrates heavily in the testes following redistribution from the extracellular fluid space. Consequently, our dose of 10 microcuries per subject represents a considerable reduction in potential hazard. Since ^{131}iodine tends to concentrate in the thyroid gland, need for it was eliminated by using Evan's blue dye, which gave satisfactory results without producing a cyanotic skin color. Although of lesser consequence, the reduction of the tritium dose to 0.5 millicuries per subject is desirable.

Summary

Body fluid compartment volumes were measured in young men during the month of March, 1968, at the peak of their athletic season. Our results for the group compare favorably with Moore's data for men in the same age group. An Evan's blue–microhematocrit method was used for measuring blood volume. Total body water and extracellular fluid space were measured with tritium and ^{35}sulfur using a double-labeling liquid scintillation technique. Reductions in ^{35}sulfur and tritium doses were achieved without sacrificing accuracy. Elimination of ^{131}iodine and ^{51}chromium and reductions in ^{35}sulfur and tritium doses lowered total body exposure to radiation by 72 per cent without sacrificing accuracy or reproducibility. Recent advances in liquid scintillation counting have made such procedures feasible.

The authors express their appreciation to Dr. C. C. Li for his assistance with the experimental design and statistical analyses.

References

1. Albert, S. N., Shibuya, J., Economopoulos, B., Radice, A., Culvo, N., Varrone, E. V., and Albert, C. A.: Simultaneous measurement of erythrocyte, plasma and extracellular fluid volumes with radioactive tracers, Anesthesiology 29: 908, 1968.
2. Albert, S. N., Hirsch, E. F., Economopoulos, B., and Albert, C. A.: Triple tracer technique for measuring red blood cell, plasma and extracellular fluid volume, J. Nucl. Med. 9: 19, 1968.
3. Shires, T., Williams, J., and Brown, F.: Simultaneous measurement of plasma volume, extracellular fluid volume, and red cell mass in man utilizing I^{131}, $S^{35}O_4$, and Cr^{51}, J. Lab. Clin. Med. 55: 776, 1960.
4. Moore, F. D., Olesen, K. H., McMurrey, J. D., Parker, H. V., Ball, M. R., and Boyden, C. M.: The Body Cell Mass and its Supporting Environment, Body Composition in Health and Disease. Philadelphia–London, W. B. Saunders Company, 1963.
5. Shapira, J., and Perkins, W. H.: Liquid scintillation counting of aqueous solutions of carbon-14 and tritium, Science 131: 414, 1960.
6. Gregersen, M. I., and Rawson, R. A.: Blood volume, Physiol. Rev. 39: 307, 1959.
7. Vennart, J., and Minski, M.: Radiation doses from administered radio-nuclides, Brit. J. Radiol. 35: 372, 1962.
8. Ryan, R. J., Pascal, L. R., Inoye, T., and Bernstein, L.: Experiences with radiosulfate in the estimation of physiologic extracellular water in healthy and abnormal man, J. Clin. Invest. 35: 1119, 1956.
9. Freinkel, N., Schreiner, G. E., and Athens, J. W.: Simultaneous distribution of T-1824 and I^{131}-labeled human serum albumin in man, J. Clin. Invest. 32: 138, 1953.
10. Zipf, R. E., Webber, J. M., and Grove, G. R.: A comparison of routine plasma volume determination methods using radioiodinated human serum albumin and Evans blue dye (T-1824), J. Lab. Clin. Med. 45: 800, 1955.
11. Schultz, A. L., Hammarsten, J. F., Heller, B. I., and Ebert, R. V.: A critical comparison of the T-1824 dye and iodinated albumin methods for plasma volume measurement, J. Clin. Invest. 32: 107, 1953.
12. Reeve, E. B., and Franks, J. J.: Errors in plasma volume measurement from adsorption losses of albumin-I^{131}, Proc. Soc. Exp. Biol. Med. 93: 299, 1956.
13. Remington, J. W., and Baker, C. H.: Evaluation of blood volume measurement techniques, Circ. Res. 9: 60, 1961.
14. Leonard, P. J., Blackman, V., and Jones, K. W.: Total body water, extracellular fluid, plasma volume and red cell mass in healthy East Africans, Clin. Sci. 29: 427, 1965.
15. Nadler, S. B., Hidalgo, J. U., and Block, T.: Prediction of blood volume in normal human subjects, Surgery 51: 224, 1962.
16. Kjellberg, S. R., Rudhe, U., and Sjöstrand, T.: Increases of the amount of hemoglobin and blood volume in connection with physical training, Acta Physiol. Scand. 19: 146, 1949.