Simultaneous Measurement in Dogs of Plasma Volume With I$^{131}$ Human Albumin and T-1824 With Comparisons of Their Long Term Disappearance From the Plasma

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When used for measurement of plasma volume, bovine plasma proteins (1) and dog hemoglobin (2) have the same initial volume distribution as the dye, T-1824, which in the bloodstream combines preferentially with albumin (3).

In regard to simultaneous comparisons in dogs of plasma volume measured with T-1824 and human albumin labeled with the I$^{131}$ isotope, there are conflicting reports (cf. 4-7). We here show results confirming those studies in man and dog in which agreement was found (6-9). It will be seen also that although loss rates of T-1824 and I$^{131}$-human albumin are similar during the first hour, the long term losses of these two substances are distinctly different in dogs, as previously reported by Wasserman and Mayerson (10).

METHODS

A sample of jugular blood was drawn, and then T-1824 and I$^{131}$-albumin were injected through the same needle from separate syringes. In the following hour five jugular blood samples were removed, and for each the hematocrit and the plasma levels of protein, T-1824 and I$^{131}$ were determined. T-1824 was measured by extraction (11) and I$^{131}$ with a scintillation counter. By using Sear's method (12), which obviates the use of pipettes, a small constant volume, 0.3-0.4 ml, was forced into a coil of polyethylene tubing fixed near the face of a thallium-activated NaI crystal. In each experiment the I$^{131}$-albumin in ampoules from Abbott Laboratories was diluted 100-200 times in 0.9% NaCl. This was immediately used for injection and for making standard dilutions as follows: three or four 1.00-ml portions were transferred to 25, 50 and 100 ml flasks and made to volume with 1% detergent (Alconox). The activities of plasma, standards and syringe wash were measured. A total of more than 8,000 counts were always recorded and the counting rates corrected for background and blank activities.

The computation of plasma volume was performed by plotting the counts per minute against the time after injection and extrapolating linearly to the moment of injection. The counting rate at zero time was divided into the mean counting rate of the standards; the resulting quotient was multiplied by both the dilution factor of the standards and the volume injected. The latter, 9.86-10.17 ml, had been corrected for the volume of I$^{131}$-albumin left in the calibrated syringe after injection. This was obtained by rinsing the syringe with 1% Alconox and then counting the activity in this volume.

In 5 experiments the loss of T-1824 and I$^{131}$-human albumin was followed for a period of 2 weeks. Here it was sometimes necessary to extract the dye from 2-3 ml plasma samples, and the lower activity required a longer counting period.

RESULTS AND DISCUSSION

Table I shows the results of 13 experiments on 12 normal dogs of the sex and condition given in columns 1-4. The percentage loss of T-1824 and I$^{131}$-albumin in the hour following the injections is obviously similar for both substances (cf. columns 5 and 6).

Simultaneous determinations of plasma volume are listed together in columns 7 and 8, and the percentage difference between the measurements with T-1824 and I$^{131}$-human albumin is given in column 9. The mean percentage difference is only $-0.51\%$, and it is indicated statistically that this difference is not significant (table 1, footnote).

In figure 1 the long term loss of T-1824 and I$^{131}$ human albumin are plotted as percentage of initial levels. In spite of considerable differences in initial levels, the relative loss of each substance is so similar for a series of five dogs that the data from all experiments are combined as shown. Although in the first hour the loss of I$^{131}$-albumin is similar to that of T-1824 (table 1 and figure 1), it is apparent that the

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subsequent removal of 131-human albumin occurs far more slowly. It may be seen that by the third day the plasma levels of T-1824 are less than 3% of the original levels. At this time, when the dye has practically disappeared from the plasma, about 30% of the 131 is still present and on the 14th day about 10%.

The primary advantage of using a substance such as 131-albumin for measuring plasma volume presumably lies in the fact that the label is attached prior to the injection, whereas with T-1824 the combination occurs in the plasma stream. The present experiments show that, practically speaking, substances outside the plasma do not compete effectively with albumin for the dye. Otherwise, the volume distribution of dye would exceed that of 131-albumin.

Simultaneous tests in dogs with T-1824 and 131-human albumin have been reported by Gibson et al. (6), Crispell et al. (7) and Krieger et al. (4). The first two groups reported on four and three dogs respectively and concluded that the results agreed. However, Krieger et al. (4) from 10 experiments indicated that the plasma volume measured with dye is higher than with 131-albumin. Neglecting the possibility of constant errors in methods employed by the various groups of investigators, such as might occur with the use of a single sample procedure (cf. 4, 5, 8), the combined results of 30 experi-
ments (17 from the literature and 13 reported here) show an absence of any significant difference in the simultaneous measurements of plasma volume in dogs. (Mean difference = -1.86%, S.D. = 8.22, t = 1.24. Therefore P > 0.20). The same conclusion has recently been arrived at by Schultz et al. (8) for the numerous simultaneous measurements on man (5, 7-9). Good agreement with T-1824 was also found in dogs for 10 experiments with bovine albumin, 2 with bovine globulin and 4 with the polysaccharide S III (1), and more recently for 10 experiments with dog hemoglobin (2).

In spite of the large differences in characteristic loss rates of these substances, ranging from nearly zero with S III (1) to 23 per cent for dog hemoglobin (2), their initial volume distributions all agree with that of T-1824. We regard the above comparative evidence, albeit indirect, as adequate proof that the dye method measures the plasma volume.

SUMMARY

In dogs, the simultaneous determinations of plasma volume with 131-human albumin and with T-1824 agree on the average to within 0.5%.

The loss rates during the first hour after injection are the same, but judging from the plasma levels measured over a period of 2 weeks, the 131-human albumin is retained longer than T-1824.

REFERENCES