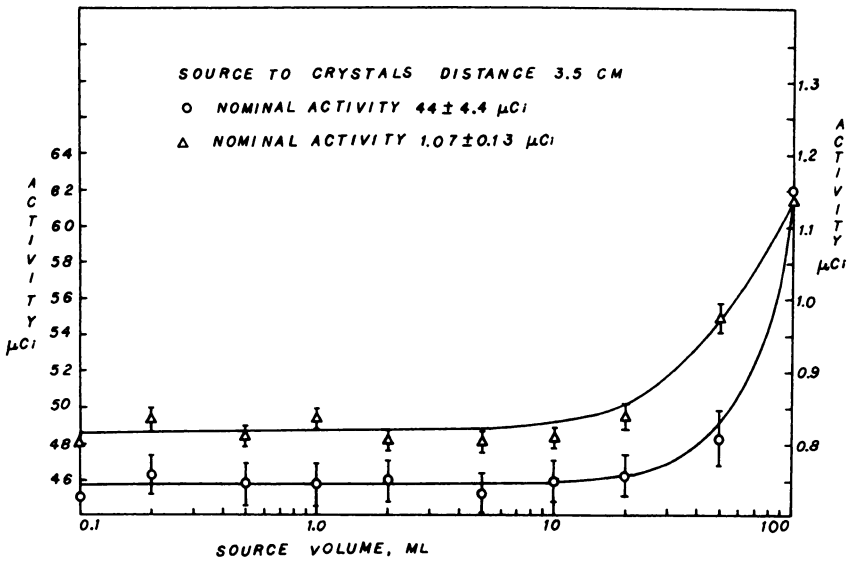

FIG. 2. Assays of two activities for various source volumes.

SCD. The 1.10-μCi line plateaus at about 6 cm and the 11.3-μCi graph plateaus at nearly 12 cm SCD. The 10% increase in the assayed activity, when the SCD is increased from 2 to 6 cm for the 1.10-μCi source and from 2 to 12 cm for the 11.3-μCi source is due entirely to the effect of the iodine x-ray escape, produced by the γ-ray photons' impinging on the crystal surface at closer and closer to 90° as the SCD is increased. The larger SCDs reduce the probability of the iodine x-ray escape through the edges of the crystal, and N_γ in the numerator of Eq. 1 becomes progressively larger than it would have been if the escape probability were constant. This effect is well understood and is discussed in detail by various authors (*3,4*). The 117-μCi source graph in Fig. 1 exhibits a shape that does not agree with the trend set by the two weaker sources. The counting rates produced by the strong source exceed the capability of the electronic circuits to handle them without significant losses. At excessive count rates, the deadtime will decrease the coincidence rate N_c faster than the corresponding decrease in N_x or N_γ of Eq. 1 and thus higher values for activity will be indicated. When the SCD is sufficiently large, this graph also levels out well within the nominal value of 117 ± 11.7 μCi. The conclusion drawn from Fig. 1 is that one must choose: (A) a geometry such that the probability of iodine x-ray escape is minimized (γ-ray crystal); and (B) distances such that the resulting counting rates can be handled adequately by the circuits used.

We next explored the effect of the source volume on the measured activity. Figure 2 shows that the assay is independent of the source volume from 0.1 to about 20 ml, when the SCD is 3.5 cm. Both sources examined show a marked rapid increase in the calculated value for the activity as the volume is increased beyond 20 ml. This effect is more pronounced for the low-activity source. This trend is also well understood and arises because any γ-ray that normally would not interact with either crystal can be Compton-scattered into the x-ray crystal by the large water volume at the extended source. If its energy falls within the x-ray spectrometer window, it will be counted as an x-ray. This increases N_x in Eq. 1 and results in a higher calculated activity. Thus, Fig. 2 shows that for an accurate assay one must use a source volume that minimizes this effect.

The overall percent error in this assaying procedure, when the SCD, the volume, and strength of the source are judiciously chosen, will depend only on the counting statistics and the dilution method, if any. In the data presented in Figs. 1 and 2 the percent error varies between 2 and 7%.

In conclusion, it can be stated that an accurate reliable method for ^{123}I assays is available through the coincidence-counting technique. The only drawback of this method is that it requires additional electronic hardware beyond that required by the single-crystal NaI counter.

PANTELIS MPANIAS
DANIEL A. GOLLNICK
WAI-NANG P. LEE
MOSES A. GREENFIELD
University of California Medical Center
Los Angeles, California

REFERENCES

1. HUDSON FR, GLASS HI, WATERS SL: The assay of iodine-123. *J Nucl Med* 17: 220–222, 1976

2. HERMAN MW, SPIEGLER P, KOONTZ R, GREENFIELD MA: Coincidence counting of ^{123}I in pediatric thyroid studies. *Radiology* 113: 455–458, 1974

3. MCGOWAN FK: Measure of K-shell internal conversion coefficients with a coincidence scintillation spectrometer. *Phys Rev* 93: 163–172, 1954

4. AXEL P: Escape peak correction to gamma ray intensity measurements made with NaI crystals. In *Brookhaven National Laboratory Report No. 271,* 1953

Reply

It has been encouraging to find considerable interest shown in our original note on ^{123}I coincidence summing (*1*). An analogy may be drawn between the effects in ^{123}I and in ^{125}I, and for some years coincidence counting has been used for the accurate assay of ^{125}I (*2*). It is interesting to note how practical use is being made of coincidence summing in ^{125}I (*3*) and we wonder whether this technique may be usefully applied to other summing nuclides. It would appear, however, as Mpanias et al. point out, that the method calls for a rather more complex technique that might not be

ideally suited for a routine user interested perhaps only in the clinical application rather than the counting method.

F. R. HUDSON
H. I. GLASS
S. L. WATERS
Hammersmith Hospital
London, England

REFERENCES

1. HUDSON FR, GLASS HI, WATERS SL: The assay of iodine-123. *J Nucl Med* 17: 220–222, 1976

2. ELDRIDGE JS, CROWTHER D: Absolute determination of ^{125}In in clinical applications. *Nucleonics* 22: 56–59, 1964

3. HERMAN MW, SPIEGLER P, KOONTZ R, et al.: Coincidence counting of ^{123}I in pediatric thyroid studies. *Radiology* 113: 455–458, 1974

Radiolabeling of Red Blood Cells

We have read with interest the article by Ryo and associates (*1*) and wish to commend the authors on their undertaking. Nevertheless, we are not in complete agreement with some of the methods and results presented in the article.

Their binding efficiencies employing the stannous glucoheptonate (SnGh) method (Method B[1]) for labeling RBCs with ^{99m}Tc (*2*) are not in agreement with our earlier published results, nor with our current work employing a modified, abbreviated method. The modified procedure requires only 7% of the reported amount of SnGh (equivalent to 4 μg of stannous ion and 14 mg of sodium glucoheptonate), eliminates EDTA, and requires only one saline wash. This reduces the preparation time without altering the binding efficiency or biologic behavior of the technetium-tagged RBCs. By either of our own labeling methods, binding efficiencies for human RBCs are greater than 95%, compared to the 90.1% average reported by Ryo et al.

It is difficult to understand the proposed advantage cited by these authors for a prolonged intravascular retention of technetium-labeled RBCs beyond 2 hr, since all imaging procedures can be completed easily within that time span. However, in order to reconfirm our results and form a basis for comparison with the authors' blood-disappearance studies by Method B, RBC-disappearance studies with ^{99m}Tc and ^{51}Cr were performed in two additional dogs. For one animal the technetium labeling was performed according to our published Method B, and in the second animal the RBCs were labeled by the abbreviated method above. All injected cells had a binding efficiency greater than 98%. Blood-disappearance results are shown in Table 1. In all whole-blood samples, more than 95% of the technetium or chromium radioactivity was associated with the RBCs. Chromium-51-RBC counts, expressed as a percent of the 15-min sample, indicate no sample less than 90% for the times shown. Comparing these results with those reported by Ryo et al. in their Tables 1 and 2 (*1*), technetium-tagged RBCs prepared in our laboratory by Method B show an intravascular retention in dogs more prolonged than those prepared by the authors when they employed the same method. In fact, our results indicate a comparable if not "superior" biologic behavior for our technetium-labeled RBCs over those prepared by the authors' Method A. The reason for the discrepancy between the results is not fully understood. However, the authors do not indicate the binding efficiency of the technetium-labeled RBCs reinfused into the dogs, nor do they fully explain the broad range in biologic half-life (5–35 hr) obtained in patients when employing technetium-tagged RBCs labeled by Method A.

TABLE 1. WHOLE-BLOOD ^{99m}Tc RADIOACTIVITY IN DOGS AFTER INJECTION OF ^{99m}Tc-LABELED AUTOLOGOUS RED BLOOD CELLS

Time after injection (hr)	SnGh method (Method B)	Modified SnGh method
½	97%	97%
1	92%	97%
2	88%	91%
24	65%	67%

Technetium-99m radioactivity present in the whole-blood sample is expressed as a percentage of the radioactivity in the 15-min sample.

We feel that technical details may be critical. Ryo et al. imply that the SnGh RBC labeling technique (Method B) was employed unmodified; yet the Unitag system, which was used, has not been commercially available since January 2, 1974. We now employ a plastic syringe method for labeling RBCs, with minimal agitation. When other labeling procedures are employed, errors could result from:

1. Incompatibilities among the reagents, red blood cells, and the labeling container.
2. Insufficient capacity of the labeling vessel to contain the 20 ml of saline for adequate washing of the RBCs. This would result in inefficient removal of the excess stannous ion and lead to poor binding efficiency.
3. Differences in ACD formulation that would result in changes in pH or dextrose concentrations, which could be damaging to the RBCs.
4. Excessive agitation.

In addition to this, we noted that at times the authors employed 5% dextrose solutions as the suspending agent for the labeled RBCs. It is well known that 5% dextrose is incompatible with RBCs, causing aggregation of cells. Moreover, RBCs incubated in 5% dextrose solutions attain a high intracellular concentration of dextrose and these cells lyse on reinjection (*3*). These factors may play a role in the shortened and irregular biologic behavior of the authors' labeled RBCs.

Observations relating to this work are: (A) poor reproducibility and low labeling yields (not seen with Method B in our laboratory); (B) poor reproducibility of in vivo data (not seen with Method B in our laboratory); and (C) Method A is time-consuming, requires excessive handling of radioactive washes, and reagents are not commercially available in sterile pyrogen-free form.

All factors considered, the authors' claim that their method "appears to be the best for labeling RBCs. . . ." seems somewhat premature.

R. F. GUTKOWSKI
H. J. DWORKIN
W. C. PORTER
H. ROHWER, Jr.
William Beaumont Hospital
Royal Oak, Michigan

REFERENCES

1. RYO UY, MOHAMMADZADEH AA, SIDDIQUI A, et al.: Evaluation of labeling procedures and in vivo stability of ^{99m}Tc-red blood cells. *J Nucl Med* 17: 133–136, 1976