## Abstracts of Papers Presented at the Society of Nuclear Medicine, Central Chapter Meeting, February 1, 1964

### *Instrumentation for the Detection of Iodine-125.* Robert N. Beck, B.S. (Argonne Cancer Research Hospital[1], The University of Chicago, Chicago, Illinois)

Iodine-125 decays by electron capture producing a Te-125 atom with an orbital electron vacancy and a nucleus with 35.4 kev excess energy. Filling of the electron vacancy is accompanied by x-ray emission (predominantly $K\alpha$ and $K\beta$ with energies of 27.4 and 31.2 kev). This process competes with the emission of Auger electrons. The 35.4 kev gamma is largely converted in the K shell providing another x-ray or Auger electrons and lower energy x-rays.

All of these emissions can be detected with suitably prepared photographic emulsions, while the more energetic gamma and K x-rays are most conveniently detected with thin window Geiger tubes, gas flow counters, scintillation detectors, etc. In diagnostic scanning procedures, scintillation detectors are used to determine the distribution of $I^{125}$ by detecting the K x-rays and 35.4 kev gamma which appear in a single photopeak. These detectors consist of a focused collimator, NaI(Tl) crystal and photomultiplier. Since a half value layer in lead is approximately 0.001″ for these photons, focused collimators can be constructed having very thin septa without loss of collimator resolution due to septum penetration. Such collimators are 2 to 3 times more efficient than their counterparts, designed for $I^{131}$. Similarly, for $I^{125}$, very high photopeak crystal efficiency is realized with thin crystals, with the additional advantage of a low background count rate. Some further increase in detector efficiency is achieved by providing the crystal with a beryllium window and a minimum of internal packing material, instead of the usual aluminum container. Good energy resolution requires the use of stable, low noise photomultiplier, amplifier and pulse height analyzer.

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[1]Operated by the University of Chicago for the United States Atomic Energy Commission.

### *The Radiochemistry and Production of Iodine-125.* W. E. Erlebach, Ph.D. (Atomic Energy of Canada, Ltd., Ottawa, Canada.)

Although the production of iodine-125 by the neutron irradiation of natural xenon was first described in 1951 it was not until almost a decade later that investigations showed that a hundred-fold reduction in cost could be obtained by production in a nuclear reactor rather than in a particle accelerator. At this same time the clear cut advantages of iodine-125 over iodine-131 in diagnostic applications and a method of production, separating, and purification were published. In the production of multi-curie batches of iodine-125, xenon gas is irradiated in a flux exceeding $10^{14}$ neutrons/cm$^2$/sec at pressures up to 5000 psi. The isotope xenon-124 which has an abundance of 0.096 percent is converted to xenon-125 which decays to the 57.4-day iodine-125. Some of the iodine-125 captures neutrons to produce the undesirable contaminant 13.3-day iodine-126. Optimum production is obtained by irradiating for one to two months and by allowing the iodine-126 to decay to an acceptable level.

The iodine is separated from the xenon by low temperature fractionation. Subsequent separation of the iodine from radio-cesiums produced during the irradiation of xenon is accomplished by ion exchange. Experiments are described which show the effectiveness of this separation.

Measurements made to ensure the purity of the product are described. These include the determination of iodine-126, cesium-137, total solids, pH, reducing agents and trace elements. The methods include gamma spectroscopy and spectrographic analysis.

Although the iodine-125 is prepared as iodide in a basic sulfite solution a combination of radiation and dissolved air in the solution rapidly oxidizes the sulfite to sulfate. This change is followed by irradiation induced reactions of the iodide whch lead to various oxidation states of iodine. A method of reconverting these states to the iodide is discussed.

### *Iodine-125 Production Studies at Oak Ridge National Laboratory.* P. S. Baker (Oak Ridge National Laboratory, Oak Ridge, Tennessee)

Since the announcement by the U. S. Atomic Energy Commission on September 20, 1963 that it (explicitly ORNL) was withdrawing from routine production of $I^{125}$, ORNL has concentrated its efforts on the AEC directive to "continue its research leading toward a large-scale production technology of $I^{125}$ which would then be made available to industry."

The ORNL research and development program has centered around an investigation of the batch-type vs loop-type experiments, the economics of normal vs enriched Xe-125 target, and a comparison of the advantages of short irradiations with no time for decay of $I^{126}$ as compared to longer irradiations with sufficient storage to allow the $I^{126}$ to decay to acceptable levels.

Unit full recovery cost for 2-curie batches of $I^{125}$ containing $< 15\%$ $I^{126}$ and made by irradiation of 55-cc cans of normal xenon at 100 psi is $\sim$ \$1.00/mc. Increasing the pressure to only 400 psi (pressures of $>1000$ psi are feasible) would lower the cost to $\sim$ \$0.30/mc, and the use of larger cans would lower the cost still further. With a circulating pressurized loop using enriched Xe-124, the cost could be reduced to $<$ \$0.05/mc.

Measurements are also being made to determine whether or not there is a nutron activation resonance peak which is sufficiently significant to be a major concern in the production of $I^{125}$.

### *The Assay and Dosimetry of Iodine-125.* Paul V. Harper, M.D. and Charles Lipscomb, M.D. (The University of Chicago and Argonne Cancer Research Hospital, Chicago, Illinois)

Iodine-125 emits $1.45 \pm 0.05$ photons per disintegration in the 27.3 – 35.4 kev energy range. Since these photons are detectable with virtually 100% efficiency using a commercially available thin crystal of NaI (Tl) with a 5 mil beryllium window, it is relatively simple to count a sample of $I^{125}$ under conditions of well defined geometry, and to determine the absolute disintegration rate. A more sophisticated approach is possible which eliminates much of the uncertainty in the decay scheme parameters and requires only a well crystal. Since $I^{125}$ decays by electron capture followed by a $\gamma$ transition, photons emitted by those two processes are in coincidence, and are summed in the crystal so that when two coincident pho-

TABLE I

AVERAGE DOSE IN RADS PER $\mu$C DESTROYED/GM

| *Organ* | $I^{131}$ $\beta$ | $I^{131}$ $\gamma$ | *Total* | $I^{125}$ *β-like* | $I^{125}$ *γ-like* | *Total* |
|---|---|---|---|---|---|---|
| 30 gm. Thyroid | 110 | 10 | 120 | 92 | 28 | 120 |
| 300 gm. Kidney | 110 | 23 | 133 | 92 | 66 | 158 |
| 1500 gm. Liver | 110 | 42 | 152 | 92 | 113 | 205 |
| 70 Kg Total Body | 110 | 90 | 200 | 92 | 180 | 272 |

tons interact in the crystal simultaneously, they appear as a single pulse in a photopeak at 55 kev instead of at 27 kev. To data thus obtained, the formulation for the conventional coincidence assay may be applied; viz,

$$\text{Activity} = \frac{(\text{total photons detected})^2}{4 \times (\text{total coincidences})}$$

to a very close approximation. Note that each coincidence corresponds to two photons detected. Most detectors have adequate resolution to separate the principal and coincidence photopeaks, and the spectrum may be displayed with a single or multi-channel analyzer or by using a simple integral bias curve. Results from these two assay methods agree within experimental error.

The radiations from $I^{125}$ fall into two groups, the high energy group of K and $\gamma$ photons which have an I $\gamma$ of 1.28 r/hr at 1 cm, and a HVL (wide beam) of 2.5 cm in tissue; and the low energy $\beta$-like group which is absorbed near the point of origin depositing 21.0 kev/disintegration. Approximate calculations based on the above parameters using the indicated assumptions are shown below in comparison with $I^{131}$. The lower energy dissipation rate of $I^{125}$ more than compensates for its longer physical half-life in all actual applications considered. Calculations for the $\beta$-like radiations present no problem, but the estimation of $\bar{g}$ for the x-rays is complicated by the attenuation and is not covered by the formulations of the standard reference works. For the thyroid, kidney and liver $\mu$ eff. = 0.277 is assumed, and the dosage calculated with an analogue computer. Spherical models are assumed for the organs. Estimated doses to other organs may be derived by interpolation from the values in Table I given the appropriate biological parameters. The total body calculation assumes complete absorption of the emitted energy.

TABLE II

$I^{131}$ AND $I^{125}$ COMPARATIVE RADIATION DOSAGE (APPROXIMATE)

| *Procedure* | $I^{131}$ *Initial Dose Rate* rad/hr | $I^{131}$ *Total Dose* rad | $I^{125}$ *Initial Dose Rate* rad/hr | $I^{125}$ *Total Dose* rad | *Dose* $I^{-131}/I^{-125}$ |
|---|---|---|---|---|---|
| | Teff = 6.9 d | | Teff = 27 d | | |
| Radioiodine Scan<br>50 $\mu$c<br>60% uptake<br>T biol = 50 days | 0.43 | 103 | .046 | 43 | 2.4 |
| Renogram<br>15 $\mu$c<br>T biol = 45 min. | Teff = 45 min.<br>0.025 | 0.027 | Teff = 45 min.<br>0.0028 | 0.0030 | 9.0 |
| Rose Bengal Scan<br>200 $\mu$c<br>T biol = 2 hrs. | Teff = 2 hrs.<br>0.074 | 0.210 | Teff = 2 hrs.<br>0.014 | 0.040 | 5.3 |
| Blood Volume Tagged Albumin<br>5 $\mu$c<br>T biol = 17 days | Teff = 5.4 d<br>0.00005 | 0.010 | Teff = 13 d<br>0.0000096 | 0.003 | 3.3 |

### *$I^{125}$ As A Source of X-rays.* JOHN R. CAMERON, PH.D. (University of Wisconsin, Madison, Wisconsin)

Iodine-125 is a nearly monochromatic source of 27.4 kev photons. 75% of the photons emitted by the isotope are at this energy. The remainder are emitted with energies of 31 or 35.4 kev. The 27.4 kev energy corresponds well to the effective kev of conventional diagnostic x-ray devices. At this energy, the linear absorption coefficients of soft tissue and of bone differ by about a factor of 10. Concentrated sources of $I^{125}$ have been used to make diagnostic x-rays. The monochromaticity of the isotope can be improved by the use of an Sn filter. A .06 mm thick filter of Sn will improve the monochromaticity to better than 95%. The resulting beam of 27.4 kev photons has been used to measure the bone mineral content *in vivo* by a direct absorption technique.[1] The good monochromaticity and constant intensity of the emitted beam make the $I^{125}$ source superior to conventional x-ray sources in this application.

[1]Cameron, J. R., and Sorenson, J.: *Science* **142**, 230-232 (Oct. 11, 1963).

### *The Application of $I^{125}$ to the Direct Photographic Visualization of the Thyroid.* GLENN CLAYTON, B.S., JOSEPH KENSKI, THEODORE FIELDS, M.S., ERVIN KAPLAN, M.D. (Veterans Administration Hospital, Physics Section—Radioisotope Service Hines, Illinois)

This investigation was begun in the hope that observation of the thyroid might be feasible by radiographic techniques wherein an elaborate mechanical scanning system would not be necessary.

The properties of x-ray film, professional photographic films, and Polaroid films were investigated. Exposures were made by direct contact with a commercial phantom. With the no-screen type of x-ray film an image appeared in 30 minutes with a 100 microcurie phantom, or 3000 microcurie-minutes. Two negatives were exposed simultaneously, as in a dental packet. The two superimposed negatives should have given an image in 1500 microcurie-minutes. This, however, proved to be below the threshold of the film. Screen-type film and x-ray intensifier screens were tried, without significant improvement. With the intensifier screens, however, a visible thyroid image was produced in 1000 microcurie-minutes on Polaroid 3000 film.

A comparison of $I^{125}$ and $I^{131}$ effects on the intensifier screen and Polaroid film will be shown. Other measurements indicated that the intensifier screen absorbs 4/5 of the $I^{125}$ radiation for conversion into light.

A more successful technique has been the use of a thin thallium-activated sodium iodide layer and Polaroid 3000 speed film. This arrangement produced an image in 700 microcurie-minutes. Polaroid film, rated at 10,000 speed, required between 200 and 300 microcurie-minutes for an image. This image detail showed up some imperfections in our sodium iodide layer, but there is a good indication that a grown and cut, optically clear sodium iodide crystal might well render much better resolution and detail than we have been able to obtain thus far.

The sodium iodide layer employed consisted of pulverized commercial crystal. The particles were suspended in a silicone fluid, 0.1 inch thick, 3 x 4 inches between two glass slide plates. It is somewhat self-absorbent to its own emitted light. By exposing our film on the same side of our NaI as the source, we were able to reduce some of the self-absorption. These conditions then produced a reasonable image in 120 microcurie-minutes.

Attempts were made to increase the sensitivity and contrast by electronic display. A television test-pattern type of generator was used with a thyroid film. The signal is connected directly to the video amplifier of a commercial TV receiver. Results will be discussed.

### *Pair-Labeled Antitumor Antibodies.* YASUO YAGI, PH.D. AND DAVID PRESSMAN, PH.D., (Roswell Park Memorial Institute, Buffalo, New York)

Antibodies against a particular organ have been shown to localize preferentially into the same organ when injected intravenously. Such localization *in vivo* of antibody can be determined readily by the use of radioactive iodine as a label. Iodine-131 was used in the past.

With normal organs, the localization values are rather constant from animal to animal, therefore the comparison between antiserum globulin and normal serum globulin can be made easily by determining average values for two separate groups of animals. However, in search for tumor localizing antibodies, we have found that the localization in tumor is extremely variable even for normal serum globulin. The amount of localization from antibody preparation above the localization of control normal globulin can be determined precisely by the pair labeling technique. Antiserum globulin was labeled with iodine-131 and normal serum globulin with iodine-125 (or *vice versa*). The two were mixed, injected into tumor-bearing animals, and the amounts of iodine-131 and iodine-125 in the tumors and other organs of the injected animals were determined with a double channel gamma-ray spectrometer. A computer program was developed to simplify the calculation of the results. Two antisera such as antihepatoma and antinormal liver, can be compared closely in the same manner.

The method was applied to studies of antibodies against the N-fluorenyl-acetamide- induced rat hepatoma *in vivo* as well as *in vitro*. The results indicate that the rabbit antihepatoma serum contains some antibodies capable of localizing preferentially in tumor as well as those which also localize in normal liver. Such information would have been impossible to get without use of this technique, since the localization of control protein or antibody is unpredictably variable in different tumors of the same individual or even in different parts of individual tumors depending on vascularity, stage of development, etc. The technique should find particularly important application in studies of human tumors in the future. Iodine-125 has completely displaced the short-lived isotopes, iodine-130 or iodine-133, which had been used in the past as a pair with iodine-131. Although these short-lived isotopes were difficult to handle because of their rapid decay, a triad labeling method with $I^{131}$, $I^{130}$ and $I^{133}$ was used in our laboratory. Now the more practical triod of $I^{125}$, $I^{130}$ and $I^{131}$ is being developed.

Iodine-125 found another important use in studies of antitumor antibodies. Because of its low energy Auger electron, it was used successfully for radioautography of tumor sections from animals injected with iodine-125 labeled antibody or control proteins. Thus, the sites of antibody localization in tumor could be determined at cellular level. The method is much more sensitive than fluorescent antibody technique. Moreover, the tissue sections are obtained by the regular histological technique (paraffin section) and are more suitable for histological examinations from frozen sections.

### *Electron Microscopic Autoradiography of Bacteria Labeled With Iodine-125.* CARL G. HARFORD, M.D. (Washington University School of Medicine, St. Louis, Missouri)

In recent years, most autoradiography of tissue sections or cultured cells has employed compounds labeled with tritium. One advantage of tritium is the low energy of the electrons it emits so that electrons that travel in directions that are not perpendicular to the plane of the section are apt to be absorbed before they reach the layer of emulsion. Iodine-125 has a similar advantage in that its Auger and internal conversion electrons have low energies. Moreover, the x-rays and gamma rays emitted by iodine-125 do not affect the emulsion in autoradiographic preparations. At the present time, methods can be used in which autoradiography can be carried out under the electron microscope and we have used such techniques to determine whether autoradiography with iodine-125 would be comparable to that with tritium. This work was done with me by Doctor Nobuko Kuhn.

Bacteria were used because they are small biological objects. They were labeled with iodine-125, fixed and sectioned for electron microscopy. The sections were coated with Ilford L4 emulsion by a technique devised by Caro in which the emulsion is taken up on a loop and allowed to gel before it is applied to the section. Bacteria labeled with tritiated thymidine were studied for comparison. In addition to an Eastman fine grain developer, we employed a developer devised by Caro which contains paraphenylene diamine and produces small rod-like grains. Counts were done to determine the percentage of grains that were over bacteria, and we found that preparations of iodine-125 and tritium were similar in that 86 percent of grains from iodine-125 were over bacteria while 71 percent of those from tritium were over bacteria.

This work indicates an accuracy of localization by iodine-125 that is at least as good as that of tritium, and we think that iodine-125 could be used in a variety of autoradiographic experiments.

Reference: Kuhn, N. O. and Harford, C. G.: *Science* 141:355, 1963.

### *The Use of Iodine-125 in Double Isotope Labeling Techniques for Metabolic Studies.* LESTER M. LEVY, M.D. (Long Island Jewish Hospital, New Hyde Park, Long Island, New York)

The availability of two isotopes of iodine, $I^{125}$ and $I^{131}$, which can be measured readily and accurately simultaneously in the same sample now makes possible a great variety of studies in the evaluation of metabolic processes.

As an example, a precursor can be tagged with one isotope and an end-product tagged with the other; both injected and followed over a period of time and the metabolic fate of both can be determined simultaneously under the same condition. If certain assumptions hold, the conversion of precursor to end-product can be quantitated.

Another example exists in the assay of metabolically interesting substances. A tracer tagged with one of the isotopes is introduced into the sample and an extraction procedure is performed. Extraction need not be quantitative since the tracer permits calculation of yield. A second substance used in the actual assay is tagged with the second isotope and the assay is carried out uncomplicated by the presence of the first or yield indicating isotope.

An illustration of this will be presented in a new method for assay of thyroxine. Biologic fluid tagged with thyroxine-I-125 (Volk) is extracted with absolute ethanol which gives an incomplete yield but is very convenient. This extraction is expoesd to triiodothyronine $I^{131}$ tagged standard pooled serum and this is passed through a molecular sieve, Sephadex G25, which simultaneously separates protein and protein bound compound from smaller molecules and also adsorbs free aromatic amino acids such as thyroxine and triiodothyronine. This permits the determination of a percentage of both thyroxine ($I^{125}$) and triiodothyronine ($I^{131}$) which are bound to the specific binding proteins of the standard serum. Standards containing known amounts of thyroxine are run simultaneously and a curve constructed. From this the amount of thyroxine in the unknown biologic fluid can be measured. This procedure has been applied to several sera from patients with thyroidal diseases. The results were compared to clinical tests and chemical protein bound iodide $I^{127}$ determinations and the method, while still preliminary, appears to afford interesting insights and new research leads to some of these disease entities.

### *The use of Iodine-125 for Determination of Transfer Rates of Proteins.* W. R. BRUCE, M.D., AND D. MOUNT, M.D. (The Ontario Cancer Institute, Toronto, Ontario, Canada)

A technique for measuring the volume of plasma and the permeability of blood vessels to proteins in a specimen of rabbit skin *in vivo* has been developed by utilizing the unique properties of $I^{125}$. Recordings of the radioactivity of a defined volume of skin were made following the injection of $I^{125}$ labelled rabbit serum albumin. Autoradiographic studies of the distribution of the isotope suggested a two compartment model for interpreting the tracings. An application of this model yielded normal values of: plasma volume 0.07 ml/gm tissue, permeability 0.8 gms albumin transferred hr/gm albumin within the vessels.

This technique has been used to examine changes of these parameters following a large acute local x-ray dose. Large easily measurable changes in both parameters were observed.

### *The Utilization of Iodine-125 for Thyroid Scanning.* N. DAVID CHARKES, M.D. (Albert Einstein Medical Center, Philadelphia, Pennsylvania)

Iodine-125 has been employed successfully for thyroid scanning at Albert Einstein Medical Center, Northern Division, and at present is used in preference to $I^{131}$ for this purpose.

To date 275 patients have been scanned with $I^{125}$. In 31 patients, $I^{131}$ scans were performed for comparison. $I^{125}$ scanning advantages stem from the greater resolution obtainable from collimating the 27.4 kev Tellurium Kα-X-ray in comparison with the 364 kev $I^{131}$ gamma photon.

Comparison of pyramidal lobes photoscanned with both isotopes consistently revealed improved ability to demonstrate this anatomic variant with $I^{125}$. The pyramidal lobe was visualized in 7 of 72 patients without thyroid disease (9.7%) and in 14 of 37 patients with diffuse toxic goiter (38%). Eighteen of 74 patients with "cold" nodules had demonstrable pyramidal lobes (24.3%), but only 3 of 31 with functioning nodules (9.7%) and 2 of 21 with multinodular (colloid) goiter (9.5%), suggesting a possible association of neoplasia with the visualization of this anatomic variant. The incidence of demonstrable pyramidal lobes among 10 patients with thyroid carcinoma was 40% (4 of 10 patients).

In some patients, cold nodules were better visualized with $I^{125}$. In no cases were $I^{131}$ scans of cold nodules superior.

Because of marked attenuation of the 27.4 kev X-ray by overlying bone, $I^{125}$ scans of substernal nodules were inferior to those produced by $I^{131}$.

$I^{125}$ may be used for thyroid scanning in conjunction with $I^{131}$ uptake studies. A pulse height analyzer is not needed to perform the uptake.

A particular advantage of $I^{125}$ is its use with $I^{131}$ in evaluation of the suppressive action of l-triiodothyronine (T-3, Cytomel®) on functioning (warm or hot) thyroid nodules. Twenty-six such nodules were studied. In two of these patients, both euthyroid, unsuspected autonomous adenomas were found. In two patients with thyrotoxicosis, dominant nodules were shown to be non-suppressible hyperplastic tissue in a diffuse toxic goiter. By using both isotopes, the study can be completed in five days, and results can be quantitated. Previously, when $I^{131}$ was used exclusively, either an excessive radiation dose was given or the study had to be extended over weeks or months, and the degree of suppression could not be quantitated.

$I^{125}$ can also be used in conjunction with $I^{131}$ in studies of TSH responsiveness.

Since these studies can be carried out with a lower radiation dose to the thyroid, $I^{125}$ is preferable to $I^{131}$ for routine thyroid scanning.

## *Clinical Application of $I^{125}$-Labeled Albumin.*[1] JOHN A. WILLIAMS, M.D.[2] (Harvard Medical School and Beth Israel Hospital, Boston, Massachusetts)

Because of its unusual radiation characteristics iodine-125 has a number of advantages over iodine-131 as an albumin label for measurements of plasma (and blood) volume. Parallel studies have revealed that $I^{125}$-albumin and $I^{131}$ albumin have the same *in vivo* distribution space, and that prepared tracer doses of each appear to retain their chemical integrity and biologic acceptability for at least three radiation half-lives. For $I^{131}$-albumin this means 24 days, while for $I^{125}$-albumin preliminary data indicate that the useful shelf-life may be in excess of 180 days. Tracer doses of $I^{125}$-albumin prepared from 6 months old stock solutions (stored at 4 degrees Centigrade) have yielded the same value for plasma volume as fresh doses of $I^{131}$-albumin.

The high efficiency with which $I^{125}$ can be counted *in vitro* by thin-cased sodium iodide detectors permits studies of plasma volume with smaller amounts of injected radioactivity (0.5 to 2.5 microcuries) than are generally necessary with $I^{131}$-albumin (1 to 5 microcuries), assuming equal sample volumes and counting times for the two tracer studies. Irradiation dosage to the patient when $I^{125}$-albumin is used is further reduced by the fact that most of the $I^{125}$ released from its protein bonding in the course of normal catabolic processes is excreted before it has a chance to decay, whereas most of an injected dose of $I^{131}$ (as $I^{131}$-albumin) decays *in vivo*.

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[1]This study was aided by a grant from the United States Public Health Service, National Heart Institute (H5547).

[2]Associate in Surgery, Harvard Medical School; Associate Visiting Surgeon and Assoc. in Surg. Research, Beth Israel Hosp., Boston.

Simultaneous determinations of red cell volume (RV) with Cr-51-tagged RBC and plasma volume (PV) with $I^{125}$-albumin can be made without a spectrometer. Measured tracer doses of Cr-51-RBC (10-50 micro-curies) and $I^{125}$-albumin are injected intravenously, and "postmix" samples of blood are drawn 10 to 20 minutes later. In an instrument such as the Volemetron, calibrated to yield direct readings of dilution volumes for both $Cr^{51}$ and $I^{125}$, stainless steel sleeves (35 mils thick) interposed between the whole blood samples and the scintillation crystals prevent the soft (0.027 mev) photons from the $I^{125}$ present in the plasma from influencing the assay of Cr-51-RBC dilution. Without the sleeves, determination of the $I^{125}$-albumin dilution can be made directly on samples of plasma from the whole blood specimens. Absorption of some of the soft $I^{125}$ radiations occurs within the plasma itself (self-absorption), and counting efficiency will, therefore, vary to a certain extent depending on sample geometry. Rigid standardization of sample tube dimensions as well as an exceptionally stable high voltage supply and discriminator level, are essential for accurate, reproducible plasma volume measurements with this tracer. An additional requirement is that the plasma contain no $Cr^{51}$, which, if present, would give rise to spuriously high plasma counting rates. To obviate this the $Cr^{51}$-RBC doses used must be carefully prepared by repeated saline rinsings to remove all $Cr^{51}$ not firmly bound within the red cells. Hemolysis, a potential source of contamination of plasma with $Cr^{51}$, can be avoided by careful handling of the blood specimens. If hemolysis does occur, the contribution of the contaminating $Cr^{51}$ to the plasma specific activity can be determined by counting the plasma sample with and without an interposed metal absorber.

## *Iodine-125 Labeled Diiodofluorescein in the Diagnosis of Intraocular Tumor*[1]

**FRANK W. NEWELL, M.D., (Department of Surgery, Ophthalmology University of Chicago, Illinois)**

The use of radioactive isotopes as an ancillary tool in the diagnosis of intraocular and intraorbital tumors is of unusual value because of the inability, in many instances, to secure tissue for histologic study without destroying the eye. Phosphorus-32 has been widely used in the study of ocular lesions but because of the limited penetration of beta particles it is of value mainly in lesions of the anterior portion of the eye where an accuracy of approximately 95 percent is obtained, provided the eye has not been the site of recent surgery. Additionally, small tumors of the iris may give rise to false negatives. Gamma emitters have not been of value in ocular diagnosis because the radioactivity arising from intracranial circulation obscures any special concentration in the eye.

Iodine-125, with its limited half-tissue depth, is thus of considerable theoretic value as an isotope to be used as an adjunct for the diagnosis of intraocular tumors. Radioactive iodine-125 conjugated to diiodofluorescein, which was first described by Moore as a diagnostic aid in brain tumors, using an iodine-131 label, has been used. It has been administered in a dosage of 5 microcuries per kilogram of body weight (sp. activity 1 mc/17 mg). Counts are carried out eight hours later over each eye and a 20 per cent difference between the two eyes is considered significant.

In 24 patients with clinically nonmalignant lesions (which were not studied histologically) there were no false positive tests and the maximum difference between the two eyes was 13 percent. In 22 patients with histologically confirmed intraocular and intraorbital lesions there were six false negatives and no false positives. This is a diagnostic accuracy of 73 per cent, roughly comparable to that seen in all types of brain tumors, using the same compound with an iodine-131 label. Four out of 13 patients with malignant melanoma of the choroid had false negatives. In two of these patients chromatography of the diiodofluorescein after the test indicated that all of the radioactive label was not bound to the compound. One of four patients with retinoblastomas had a false negative.

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[1]This investigation has been supported in part by Research Grant NB 03216 and by Program Project Grant B-3358 from the National Institute of Neurological Diseases and Blindness of the National Institutes of Health, Public Health Service.

It should be emphasized that when the questionable lesion is in the anterior segment of the eye the use of phosphorus-32 is superior to iodine-125. This, however, appears to be the isotope of choice in posterior lesions. There are a number of other agents to which iodine-125 may be conjugated and may prove to be more satisfactory than diiodofluorescein.

### *The Use of Iodine-125 in Diagnostic Urology.* CHESTER C. WINTER, M.D. (The Ohio State University Hospitals, Columbus, Ohio)

Sodium ortho-iodo-hippurate (Hippuran®), secreted almost exclusively by the renal tubules, serves to measure renal blood flow as well.(1) Its iodine atom is exchangeable with either $I^{131}$ or $I^{125}$, making it a choice radioactive test agent in diagnostic urology.(2) The practical advantages of $I^{125}$ over $I^{131}$ are: 1) longer shelf-life, 2) greater safety to personnel and patient, 3) applicability of less bulky and more portable equipment, and 4) greater directionality of its photons. These advantages are directly attributable to the low voltages of the x- and gamma rays of $I^{125}$ (27 to 35 kev) and absence of beta particle emission.(3)

The urologic procedures beneficially utilizing Hippuran-$I^{125}$ as the testing medium are: 1) the radioisotope renogram, (4, 9) 2) the radioisotope blood-kidney clearance test, (5) 3) the radioisotope uroflometry test, (6) 4) the radioisotope bladder residual test, (6) 5) the radioisotope vesicoureteral reflux test, (7) 6) the radioisotope individual and total renal excretion tests, (8) 7) the radioisotope individual and total renal clearance tests, (9) and 8) the radioisotope kidney roentgenogram. (10)

Standard renography equipment is modified when $I^{125}$ is used by the reduction of the thickness of the scintillation crystal to 2 mm. Collimators are of brass instead of lead with marked reduction in their weight and size. Transistorized scintillation counters and ratemeters are additional features that make the equipment easily portable and less bulky. These fortitous improvements make renography more adaptable to seriously ill patients confined to bed, and to small children.

The attractive features of $I^{125}$ are lessened somewhat when small scintillation crystals and multiplier phototubes are used with a resultant increase in "electronic noise" in the absence of a discriminator. Likewise, an increased background count rate is due to the use of less dense collimators. And finally, there is more difficulty encountered with voltage regulation of the ratemeter. Application of optimal crystal and probe size and circuitry should reduce these problems. The equipment modifications for $I^{125}$ increase the cost, but reduced isotope shipping charges and a less frequent purchase schedule are compensating factors. It appears that difficulty in producing uncontaminated strong point sources of $I^{125}$ has daleyed the widespread use of this radioisotope in roentgenography.

The renogram measures vascular capacity, tubular function and evacuation ability of the kidney. Renography is useful as a screening test, especially for renal hypertension, and is valuable in serial appraisals of renal disorders and during surgical procedures on the kidney. (11) While renography is in progress, a third probe placed against the chest makes possible the production of a curve that records the isotope level in the systemic blood and serves to measure total renal function; this procedure is a useful adjunct to renography of the individual kidney.

At the conclusion of a renogram there is generally enough radioactive urine in the bladder to give a high count rate so that radioisotope uroflometry can be carried out. With the patient sitting or standing, a probe is placed posteriorly over the bladder and the subject is instructed to void. The recorder speed is increased to six inches per minute and maximal as well as average flow rates are depicted and computed. The test also reveals bladder residual.

The vesico-ureteral reflux test is carried out by placing a scintillation probe over each kidney with the patient lying supine. A small dose of hippuran-$I^{125}$ is placed in the bladder which is then filled with saline. A continuous recording will detect any isotope reaching the kidney. The standard roentgenographic method for this purpose has the disadvantages of intermittent films and a larger dosage of irradiation.

The radioisotope renal excretion test is performed by the intravenous inoculation of a known amount of Hippuran-$I^{125}$. The percentage of the inoculum collected in the bladder

during a period of time is an index of total renal function. If urine is collected from each kidney through ureteral catheters, an individual renal test is made available. When a continuous infusion of Hippuran-$I^{125}$ is given to maintain a steady and low blood level, renal clearance studies indicative of renal blood flow or tubular functional capacity can be computed.

Kidney roentgenograms are made with $I^{125}$ by placing a strong point source on one side of the kidney and a small malleable cassette containing a non-screen film on the other side. A 2- or 3-second exposure produces satisfactory pictures of kidney stones during surgery.

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