Evaluation of the Lung Uptake of Iodine-131 HIPDM by the Single-Pass Multiple Indicator Dilution Technique in a Rabbit Model

Douglas N. Abrams, S.F. Paul Man, and Antoine A. Noujaim

Department of Nuclear Medicine, Health Sciences Centre, Winnipeg; and Faculty of Medicine and Faculty of Pharmacy and Pharmaceutical Sciences, University of Alberta, Edmonton, Alberta, Canada

The first-pass lung uptake of [^{131}I]HIPDM was studied in a rabbit model using a conventional single-pass multiple indicator dilution (SPMID) technique with [^{99m}Tc]HSA as the intravascular marker. Lung extraction of [^{131}I]HIPDM determined by the SPMID method was saturable, decreasing from 89.5 ± 5.8% (mean ± s.d., n = 4) at 4 nmol kg^{-1} to 63.8 ± 6.2% at 1.5 × 10^4 nmol kg^{-1} relative to the intravascular tracer. The first-pass extraction of [^{131}I]HIPDM derived from gamma-camera emission imaging data correlated well (R = 0.834) with simultaneously derived SPMID data. The extraction efficiencies were 94.4 ± 3.3 and 87.9 ± 3.2 at 200 nmol kg^{-1} and 68.0 ± 6.2 and 64.0 ± 10.7 at 1 × 10^4 nmol kg^{-1} (n = 4) for the SPMID and gamma-camera methods, respectively. Analysis of the lung arterial effluent blood indicated that no appreciable de-iodination or metabolism of [^{131}I]HIPDM, within the limits of detection, occurred during the first pulmonary transit.

Radionuclide studies have been used to study regional lung function for almost three decades and have contributed much to the understanding of normal and abnormal lung function. However, little information concerning the metabolic integrity of the lung can be obtained using conventional ventilation-perfusion imaging, gallium scans, or labeled blood cell studies (*1*). The importance of the lung as a metabolic organ has gained recognition recently with its role as a metabolic regulator of biogenic amines, prostaglandins, and vasoactive hormones (*2*). It has been suggested that the altered uptake of vasoactive or other lung avid substances might provide a sensitive indicator for pulmonary endothelial damage (*3*).

One of the methods for the study of the metabolic disposition of drugs by the lung is the multiple indicator dilution technique in either an isolated perfused lung or an intact animal. This method is, however, an invasive technique with limited clinical applicability. The use of gamma-camera emission imaging to determine

the metabolic integrity of the pulmonary system has been investigated. The uptake and washout of carbon-11 (^{11}C) -octyl amine was measured in mice (*4*), rabbits (*5*) and humans (*6*) while the uptake of [^{11}C]chlorpromazine (*7*) in normal lungs has been compared with that in patients with chronic obstructive lung disease. More recently, N-isopropyl-*p*-iodoamphetamine (IMP) (*8*) has been evaluated as a potential radiopharmaceutical for use in assessing the metabolic capacity of the lung and was shown to be extracted by the lung by a saturable binding system (*9*).

N,N,N'-trimethyl-N'- (2-hydroxy-3-methyl-5-iodobenzyl)-1,3-propandiamine (HIPDM) (*10*) was originally synthesized as a "pH shift" reagent for brain perfusion imaging. However the initial tissue distribution work indicated that iodine-131 ([^{131}I]HIPDM) also had a strong affinity for the lung (*11*). It has also been proposed for studying pulmonary metabolic function and lung uptake and clearance of HIPDM has been reported (*12–14*).

We wished to further assess the kinetics of uptake of [^{131}I]HIPDM by the lung using the in vivo single-pass multiple indicator dilution (SPMID) technique. In addition, we wished to modify this technique to make it more applicable to gamma camera imaging; we there-

fore correlated these extraction values with gamma-camera derived retention values.

MATERIALS AND METHODS

Radiochemicals and Instrumentation

The [^{131}I]HIPDM and technetium-99m human serum albumin ([^{99m}Tc]HSA) were employed*. The radiochemical purity of the radioiodinated HIPDM was monitored by radio thin layer chromatography (radio-TLC) on silica-gel (Whatman MK6F) using ethanol:ethyl acetate 1:1 by volume as the eluting solvent (Rf HIPDM = 0.05, Rf iodide = 0.85). Some de-iodination of the [^{131}I]HIPDM did occur upon standing therefore the free radioiodide was removed prior to use by elution of the HIPDM through a DEAE-cellulose column (1.0 × 0.4 cm i.d.) with 0.005M lithium acetate buffer. Final radiochemical purity of [^{131}I]HIPDM used was always >95%. The radiochemical purity of the [^{99m}Tc]HSA was >99% and it was used without further purification.

Gamma-emitting radionuclides were counted with a Beckman Gamma-8000 gamma-counter. All samples were decay corrected where necessary. The reverse-phase high pressure liquid chromatographic (RP-HPLC) separations were performed on a Tracor 900 series chromatographic system and a Radial Pac C8 column (Waters) using tetrahydrofuran: 0.1M TRIS buffer (pH = 8.1):acetonitrile 1:5:10 by volume, as the solvent (iodide Rt-void volume, N'-demethyl HIPDM Rt = 4.5 min, HIPDM Rt = 6.0 min). The column effluent was monitored for ^{131}I with a Flo-One DR radioactive flow detector (0.5 ml cell volume) in liquid scintillation (LS) counting mode using Aquasol 2 (4 ml min^{-1}) LS scintillant. Liquid scintillation samples were counted on a Beckman LS9000 LS counter. Gamma images were obtained on a Pho-Gamma IV Digital PDP 11/34 camera system operating under Gamma 11 software.

Lung Extraction of HIPDM

New Zealand White rabbits (3.5–4.5 kg) under general anesthesia were used for the study. The rabbits were anesthetized with a mixture of allobarbital (64 mg ml^{-1}), urethane (255 mg ml^{-1}) and monomethyl urea (360 mg ml^{-1}). The animals were tracheostomized after a midline incision and catheters (PE 190) were inserted into the right atrium and the ascending aorta through an external jugular vein and an internal carotid artery respectively. A second arterial line in either a femoral artery or a carotid artery was connected to a Statham pressure transducer for continuous monitoring of the blood pressure (normal range 80–140 mmHg) on a Gould recorder 2400S. In addition, cardiac output calculated from the reference curve measurements (350–600 ml min^{-1}), arterial blood gases (pCO$_2$ 18 – 28, pO$_2$ 100–250, ventilated with supplemental O$_2$) and rectal temperature (36.5–38.5 C) were monitored throughout the experiments.

Technetium-99m HSA was used as the intravascular tracer since it does not leave the pulmonary circulation during a single pass. Three successive measurements of the pulmonary extraction of [^{131}I]HIPDM at increasing concentrations were obtained at 45-min intervals by injecting a bolus (1.0 ml) of a mixture of [^{131}I]HIPDM and [^{99m}Tc]HSA into the right atrial line and sampling the arterial blood leaving the lung through the ascending aortic line. The arterial canula was attached to

a peristaltic pump which transferred the blood at a rate of 20 ml min^{-1} to a fraction collector (Pharmacia Frac 100) which collected at a rate of 0.6 sec per sample. The blood samples were collected in 1.5 ml Eppendorf centrifuge tubes containing 1.0 ml of heparinized (1,000 IU l^{-1}) normal saline kept at ice-bath temperature. First appearance of the indicators in the blood occurred after at least ten samples had been collected. Accordingly, aliquots (5 μl) of the injection sample were added to the first five collection tubes as reference samples and were analyzed with the effluent samples. All samples were weighed and analyzed for ^{131}I and ^{99m}Tc activity by gamma-spectroscopy using standard dual-label crossover techniques.

The extraction of [^{131}I]iodide by the lung was also determined similarly in the same animals following the [^{131}I] HIPDM experiments.

In a second series of experiments, dynamic gamma-camera images of [^{131}I]HIPDM uptake and release by the lung were acquired (64 × 64 matrices; 20% ^{131}I window; medium-energy, parallel hole collimator) simultaneously during the SPMID measurements. Images were obtained at four frames sec^{-1} for 60 sec and at 60 sec intervals thereafter for 30 min. Crossover of ^{99m}Tc into the ^{131}I window was negligible under these conditions.

Data Analysis

The fractional concentrations of [^{99m}Tc]HSA and [^{131}I] HIPDM were calculated as the ratio of the concentration of [^{99m}Tc]HSA and [^{131}I]HIPDM (in nmol) in each arterial blood sample to the total amount of [^{99m}Tc]HSA and [^{131}I]HIPDM (in nmol) injected. These values were used to generate the fractional concentration versus time curves and to calculate the percentage of removal (instantaneous extraction ratio, E) of [^{131}I]HIPDM at each time point (15) using the equation:

$$E(t) = 1 - (Cp(t)/Cr(t)) \tag{1}$$

where Cp and Cr are the fractional concentrations of the permeable ([^{131}I]HIPDM) and reference ([^{99m}Tc]HSA) tracers, respectively. The cumulative extraction of [^{131}I]HIPDM within any interval during the first pass was calculated with the equation:

$$E(t)dt = \int_0^t (Cr(t) - Cp(t))dt/(Cr(t)dt) \tag{2}$$

The extraction values are reported as a % instantaneous extraction at peak, % cumulative extraction to peak and % cumulative extraction to first pass (Table 1).

The gamma-camera data were analyzed by generating background corrected time-activity curves for regions of interest (ROIs) for the lungs excluding the heart and normalizing the data to peak values (7). The peak value of each curve was determined by fitting the data to a gaussian curve. The descending portion of the curve was smoothed by least squares regression analysis. The gamma-camera derived retention values were calculated from the ratio of the lung activity at the end of one pulmonary transit to the lung activity at peak. The time for one pulmonary transit was calculated from the reference curve of the SPMID data. The time point at which the lung retention of [^{131}I]HIPDM was determined was taken as the time at which peak lung activity occurred plus the length of time required for one pulmonary transit.

TABLE 1
Dose Effect on Lung Extraction of HIPDM Calculated from SPMID Measurements

Norminal HIPDM concentration (nmol kg⁻¹)		Instantaneous peak extraction E(t)	Extraction to peak E(t)dt	Extraction to first pass E(t)dt
4	mean	89.6	89.5	88.4
	± s.d.	5.5	5.8	6.0
50	mean	91.5	91.9	89.2
	± s.d.	0.5	1.3	7.4
150	mean	91.8	92.2	88.4
	± s.d.	3.7	3.5	8.1
500	mean	80.9	80.6	78.2
	± s.d.	2.7	4.7	5.1
10,000	mean	66.2	69.5	62.0
	± s.d.	3.3	6.9	7.0
15,000	mean	53.2	63.8	21.2
	± s.d.	8.4	6.2	22.4
40,000	mean	51.2	51.5	33.8
	± s.d.	11.3	20.0	14.7

De-iodination of [^{131}I]HIPDM

The extent of [^{131}I]HIPDM de-iodination occurring in the lung during the first pass was studied in three animals. Blood samples obtained as described for the SPMID experiments were collected in 1.0 ml of an ice cold solution of heparin (1,000 IU l⁻¹) and HIPDM (10 μM) in normal saline. The samples were counted, weighed, and centrifuged. One ml of the supernatant was transferred to an Amberlite IR-120 cation-exchange column (1.0 × 1.0 cm i.d.). The pellet was diluted with 1.0 ml distilled water, vortexed to lyse the RBCs and transferred quantitatively with distilled water to a cation-exchange column as above. The columns were eluted with 8.0 ml of an aqueous solution of sodium iodide and sodium thiosulfate (1% by volume). The ^{131}I radioactivity in the eluate and that remaining on the resin was determined.

Free [^{131}I]iodide was eluted from the column with 99.8% efficiency and [^{131}I]HIPDM was retained with 98.4% efficiency with this technique. Aliquots of the injection sample were analyzed along with the effluent blood samples to serve as controls for each experiment.

In one rabbit, blood samples for RP-HPLC analysis were treated similarly to the samples analyzed by the ion-exchange method up to the separation step. Each sample was then extracted with 3 by 3.0 ml volumes of methanol and the combined extracts were dried under a nitrogen atmosphere with gentle heating. The dried samples were stored at −80°C until analysis, at which time each sample was dissolved in 1.5 ml RP-HPLC solvent and 0.5 ml of this extract was analyzed by RP-HPLC directly. A second 0.5 ml aliquot was analyzed directly by LSC to determine the total amount of ^{131}I injected. The column effluent was monitored for uv absorbance at 260 nm and for radioactivity with a flow through detector. The effluent was also collected at 30-sec intervals and counted by LSC.

RESULTS

In the SPMID experiments, the pulmonary extraction of progressively increasing doses of [^{131}I]HIPDM

was determined (Table 1). The instantaneous extraction at peak (E(t)) was calculated using Eq. (1) and the integrated extracted (E(t)dt) to peak and extraction to first pass was calculated using Eq. (2). Data derived from animals with abnormal physiological data were omitted from the calculations.

Figure 1 is a plot of the fractional concentrations of [^{131}I]HIPDM (at different doses) and [^{99m}Tc]HSA recovered in the arterial blood versus sample time. Figure 1 was obtained from data from one rabbit following three successive injections of [^{131}I]HIPDM in progressively increasing doses. The [^{99m}Tc]HSA reference curve (▲) serves as an indicator that is not metabolized or extracted by the lung and is confined to the vascular space under these experimental conditions. Therefore
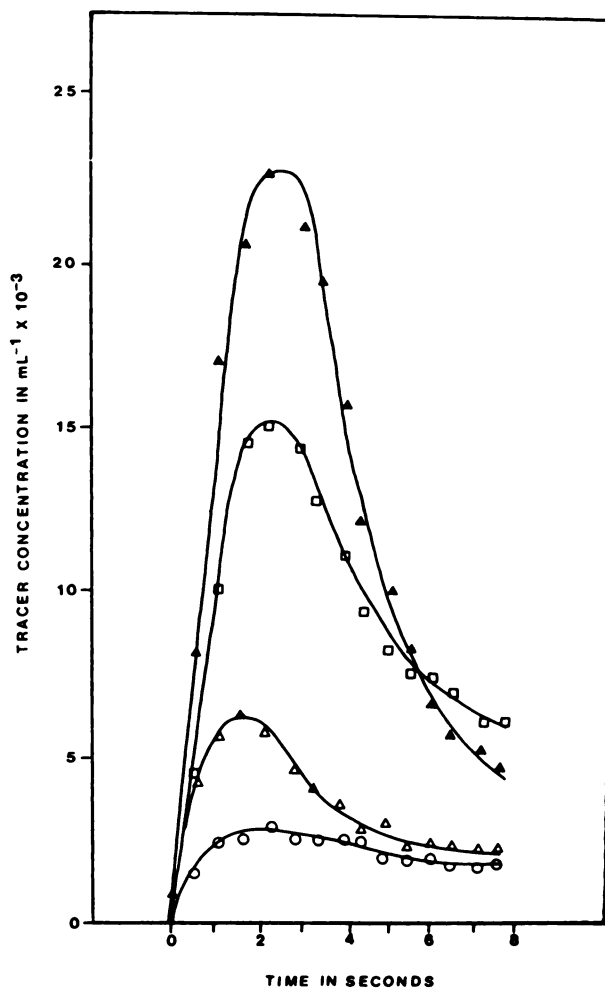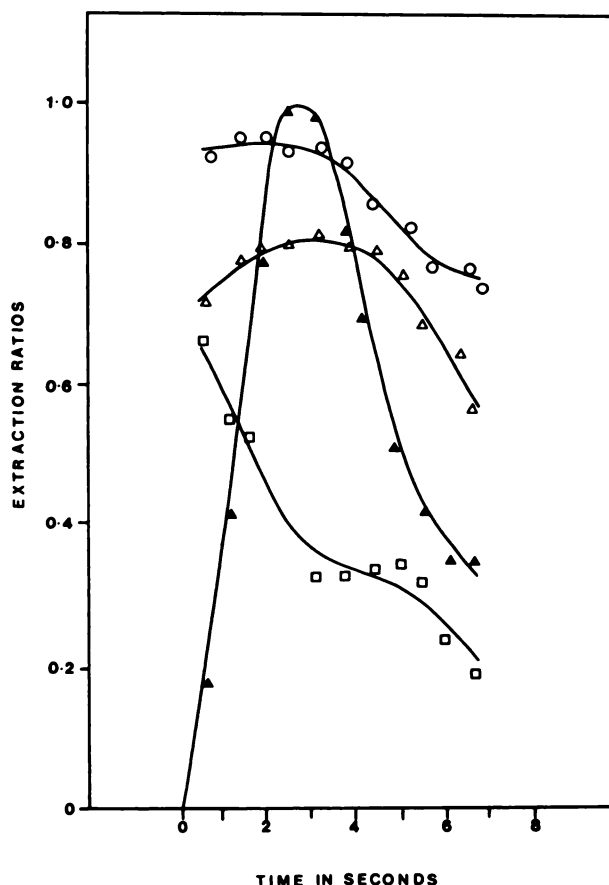

FIGURE 1
Pulmonary extraction of [^{131}I]HIPDM derived from the single-pass multiple indicator dilution experiments. The data are expressed as the total dose of [^{131}I]HIPDM injected which was recovered in the blood following a single pass through the lung. Different doses of [^{131}I]HIPDM are compared. (O) 4 nmol/kg, (Δ) 600 nmol/kg, and (☐) 30,000 nmol/kg [^{131}I]HIPDM. [^{99m}Tc]HSA (▲) was injected simultaneously with each dose of [^{131}I]HIPDM and was used as the reference marker to which the recovery of HIPDM was normalized.

this curve represents complete recovery of the injected material. The normalized concentration curves of [^{131}I]HIPDM were not congruent with the [^{99m}Tc]HSA reference curves since only a fraction of the injected [^{131}I]HIPDM was recovered in the effluent blood due to selective extraction by the lung. The area under the [^{131}I]HIPDM curves was proportional to the lung extraction efficiency which in turn was a function of the injected dose.

The instantaneous extraction of [^{131}I]HIPDM was calculated by comparing the areas under each curve and assuming 100% recovery of the [^{99m}Tc]HSA. The shape of the curves was dose dependent, as the area under each curve increased as the injected dose increased. This indicated the presence of a saturable component in the uptake process. The descending portion of the [^{131}I]HIPDM curve was complicated by return of this compound into the circulation and resulted in the crossover of the descending portions of the [^{131}I]HIPDM and [^{99m}Tc]HSA curves observed at the higher doses of HIPDM.

The effect of [^{131}I]HIPDM return from the lung may be more readily illustrated by Figure 2, in which the instantaneous extraction ratios, E(t), of [^{131}I]HIPDM are plotted versus time. The associated [^{99m}Tc]HSA reference curve (arbitrary units) was included to put timing into perspective. The E(t) curves for the lower doses (4 nmol and 600 nmol/kg) were relatively constant early in time until return of [^{131}I]HIPDM from the lung (presumably the endothelial cells) into the blood became great enough to decrease the calculated extraction. The very early rise in the E(t) curves has been attributed to the inhomogeneity or nonuniformity of the capillary transit time of the blood in the lung (15). Thus, the [^{131}I]HIPDM appearing in the early effluent has had a shorter residence time in the lung capillaries than [^{131}I]HIPDM appearing later and this would result in a lower extraction efficiency. For the higher doses (30,000 nmol/kg) the E(t) curve tended to decrease during the transit of [^{131}I]HIPDM through the lung due to early saturation of the uptake process and return of [^{131}I]HIPDM becoming more prominent earlier in the curve.

The % E(t) in Table 1 supported the dose dependency of the lung extraction of [^{131}I]HIPDM at higher injected doses. Table 1 is presented as the % instantaneous extraction of [^{131}I]HIPDM at the curve peak and also as the % integrated extraction from the initial appearance of [^{131}I]HIPDM in the effluent blood to peak and to end of first pass. The contribution of [^{131}I]HIPDM return from the lung to the [^{131}I]HIPDM concentration in the blood was considered negligible in the instantaneous peak and extraction to peak calculations. The lower extraction efficiencies calculated up to the end of first pass for the higher doses of [^{131}I]HIPDM, were due to return of [^{131}I]HIPDM from the lung to the blood.

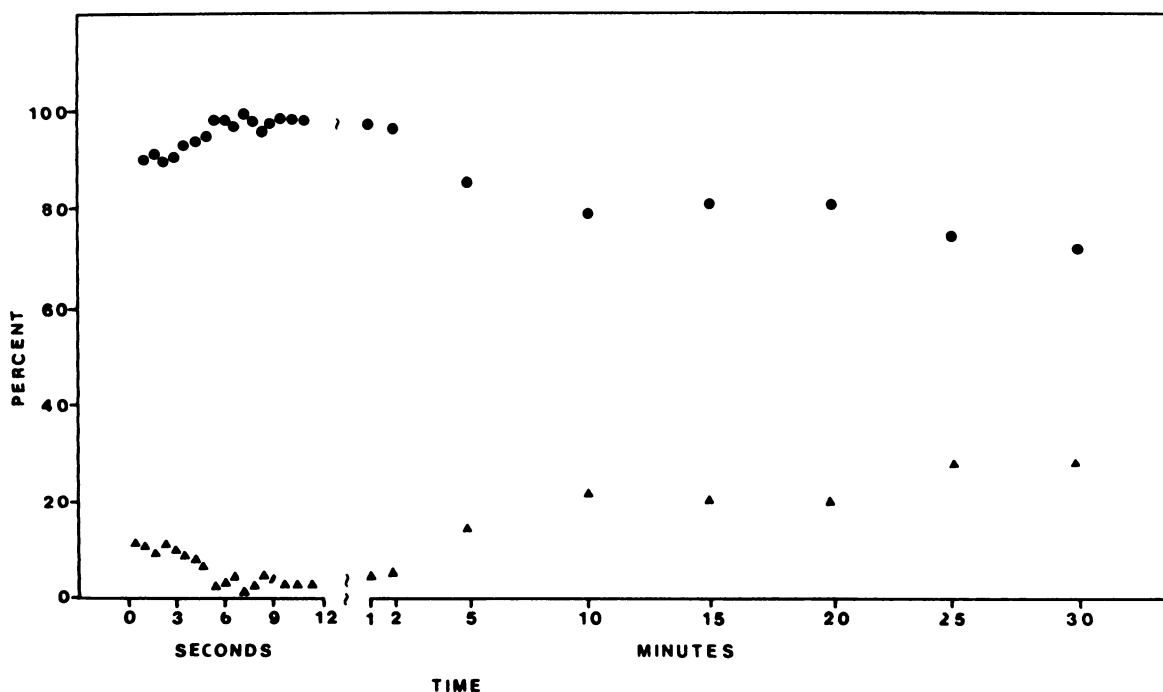

FIGURE 2
Pulmonary extraction of [^{131}I]HIPDM derived from the single-pass multiple indicator dilution experiments. The data are expressed as a ratio of the fractional concentration of [^{131}I]HIPDM to [^{99m}Tc]HSA recovered in each sample following a single pass through the lung. Different doses of [^{131}I]HIPDM are compared. (O) 4 nmol/kg, (△) 600 nmol/kg, and (□) 30,000 nmol/kg [^{131}I]HIPDM. The [^{99m}Tc]HSA was injected simultaneously with each dose of [^{131}I]HIPDM and was used as the reference marker. The [^{99m}Tc]HSA (▲) curve is included to put timing into perspective.

The chemical form of ^{131}I leaving the lung during the first transpulmonary pass was analyzed by two methods. A cation exchange resin was used to separate free [^{131}I] iodide from [^{131}I]HIPDM to estimate the extent of de-iodination occurring during the first pass and thereafter (Fig. 3). The data are expressed as the fraction (%) of total blood ^{131}I identified as either free iodide or intact HIPDM. No appreciable de-iodination occurred during the first pass but free iodide became evident within 5 min postinjection.

The high concentration of [^{131}I]iodide present early in Figure 3 data has been attributed to the presence of free [^{131}I]iodide (<5%) in the [^{131}I]HIPDM preparation and the difference in the extraction of [^{131}I]iodide and [^{131}I]HIPDM by the lung. Therefore at early transit times the [^{131}I]HIPDM was extracted with >90% efficiency (Table 1) while the free [^{131}I]iodide was extracted

FIGURE 3

Chromatographic analysis of the ^{131}I-labeled compounds leaving the lung in the blood immediately following a single pass of [^{131}I]HIPDM through the lung. The data are expressed as the fraction (%) of the total blood ^{131}I co-chromatographing with either free iodide (▲) or intact HIPDM (●).

with 17.8 ± 3.9% efficiency (data not included). This produces a higher free [^{131}I]iodide to [^{131}I]HIPDM ratio than can be corrected for by subtraction of the free [^{131}I] iodide found in the injection standards. This ratio decreases as the contribution of [^{131}I]HIPDM return from the lung increases. This trend reverses itself at later time periods (1–30 min after injection) as the fraction of [^{131}I]HIPDM steadily decreases with a concomitant rise in free [^{131}I]iodide in the blood.

The ion-exchange experiments allowed the extent of de-iodination occurring in the lung during the first pass to be estimated but did not differentiate between [^{131}I] HIPDM and metabolites retaining both ^{131}I and the amino functionality. Therefore, a RP-HPLC method for separating [^{131}I]HIPDM from [^{131}I]iodide and other potential metabolites was developed. The sensitivity of the RP-HPLC technique was ~1.5% of the total effluent radioactivity. The pattern of radioactive components identified in the blood using RP-HPLC was virtually identical to that observed using the cation-exchange resin (data not presented).

The differences in lung extraction efficiency of different doses of HIPDM determined using the single pass multiple indicator dilution method were also demonstrated with the dynamic gamma-camera imaging of the first pass of [^{131}I]HIPDM through the lung. The pulmonary retention (R(t)) of progressively increasing doses of [^{131}I]HIPDM was determined from the dynamic gamma-camera derived data and compared to
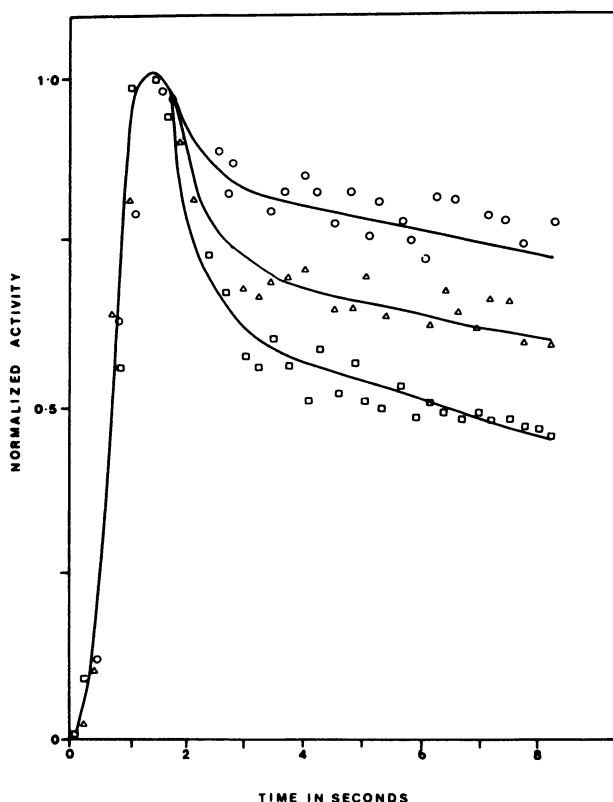
SPMID data obtained from the same injections (Table 2). The data given in Table 2 indicated that the two methods were directly comparable and have a correlation coefficient of 0.894 (n = 11). Figure 4 is a plot of the whole lung activity normalized to peak lung activity versus time for three doses of HIPDM (4.0 to 3 × 10^4 nmol kg^{-1}) injected sequentially in the same rabbit. For comparison, these figures were derived from the same experiments as the SPMID data presented in Figures 1 and 2. The retention values after one pulmonary transit of 75.6, 63.2, and 49.7 correlate well (R = 0.988) with the SPMID (extraction to peak) values of 92, 79, and 34, but do not demonstrate the large spread of the SPMID data.

DISCUSSION

The selection of HIPDM as a suitable tracer for the study of the metabolic function of the lung was based

TABLE 2

Comparison of SPMID Calculated Lung Extraction and Gamma-Camera Derived Lung Retention of HIPDM

Nominal HIPDM concentration (nmol kg^{-1})		Instantaneous peak extraction E(t)	Extraction to peak E(t)dt	Gamma-camera derived retention R(t)
200	mean	94.4	95.0	87.9
	± s.d.	3.3	3.1	3.2
10,000	mean	66.2	69.5	64.0
	± s.d.	3.3	6.9	10.7

FIGURE 4
Pulmonary retention of [131I]HIPDM derived from dynamic gamma-camera images. The data are expressed as time-activity curves generated for the lungs during a single pass of a bolus dose of [131I]HIPDM through the lungs. Different doses of [131I]HIPDM are compared. (○) 4 nmol/kg, (△) 600 nmol/kg, and (□) 30,000 nmol/kg [131I]HIPDM. The data were normalized to the peak value which was calculated by fitting the data to a Gaussian curve.

on the potential for HIPDM to be a substrate for the monoamine (16) or polyamine (17) transport mechanisms in the lung; the potential for HIPDM to be a substrate for one or more of the monoamine, diamine, and polyamine oxidase enzyme systems thought to be present in the human lung; and the high uptake of [123I] HIPDM demonstrated in rat (11) and human (12-14) lungs.

Pistolesi et al. (12-14) have studied the blood and lung clearance of [123I]HIPDM in man using gamma-camera imaging but did not study the rapid uptake phase immediately following injection. We investigated the uptake of HIPDM further by first determining whether a saturable uptake mechanism for HIPDM could be demonstrated using a first-pass multiple indicator dilution technique and secondly, by deriving similar lung extraction data using gamma-camera emission analysis. The single pass multiple indicator dilution method for determining the lung extraction of [131I] HIPDM required the injection of two tracers simultaneously, knowledge of the mass of each tracer injected

and the fraction of each tracer recovered in the arterial effluent blood. From this data, the lung extraction of [131I]HIPDM was calculated as the difference between the fractional recoveries of the vascular tracer ([99mTc] HSA) and [131I]HIPDM. Derivation of the lung retention of [131I]HIPDM from the gamma-camera data required a noninvasive method for calculating the fractional concentrations of [131I]HIPDM remaining in the lungs. This was solved by normalizing the data to the peak count rate in the region of interest (ROI) (7). This calculation makes the assumption that if a small volume bolus is injected quickly, all of the injected [131I]HIPDM will have entered the lung ROI before any has left. This peak value therefore represents the total injected mass.

Analysis of the blood samples obtained from the SPMID extraction studies revealed that a significant (45–50%) proportion of the 131I injected as [131I]HIPDM was present in the RBC pellet. Preliminary studies (data not presented) indicated that the rate and the extent of RBC uptake of [131I]HIPDM at 0°C was similar to that observed at ambient temperature. This suggested that the RBC uptake was mostly a physical process since a mediated uptake mechanism would be expected to be much less active at 0°C. Although the affinity of [131I] HIPDM for RBCs was substantial, it did not seem to compete efficiently with the lung uptake of [131I] HIPDM which was ~90% at lower doses (Table 1).

Since the metabolites of [131I]HIPDM are not known, the separation of N'-demethyl HIPDM (the chemical precursor to HIPDM) from [131I]HIPDM was used to define the necessary separation conditions. N'-demethyl HIPDM was selected because it is a possible metabolite of HIPDM and it is more polar than HIPDM. Most metabolites of aromatic amines are more polar than the parent substrate. The separation of these two compounds implies the ability to separate other more polar metabolites of HIPDM using these solvents and separation parameters. Under these conditions no radioactive components other than [131I]iodide and [131I] HIPDM were observed in either the plasma or RBC fractions of the arterial effluent blood. Any component comprising 1.5% of the total effluent activity would have been detected. The pattern of radioactive components identified by RP-HPLC was virtually identical to that observed using the cation-exchange resin.

The chromatographic analysis of the blood samples leaving the lung indicated that little metabolism of [131I] HIPDM occurred during the first pass through the lung. A recent article by Rao et al. (18) suggests that rabbit lungs do not contain diamine oxidase. This lack of DAO activity may be partially responsible for the low rate of metabolism observed. However, HIPDM is a potential substrate for other amine oxidase enzymes thought to be present in the rabbit lung. Some deiodination was observed to occur well after the first pulmonary transit.

The results suggest that [^{131}I]HIPDM was efficiently extracted by the lung at low doses and that this extraction efficiency was dose related. However, relatively large doses of HIPDM were required to demonstrate the saturability of the extraction process in comparison to other substrates such as serotonin and norepinephrine (16). In addition, doses in the range of 3×10^4 nmol kg^{-1} were found toxic in a number of rabbits and the majority of experiments used a maximum of 1×10^4 nmol kg^{-1}. The high doses necessary to decrease the lung extraction efficiency of HIPDM may be due in part to the high lipophilicity of HIPDM resulting in a large degree of nonspecific uptake by the large surface area of the lung. This may explain the lack of substantial difference in lung extraction of HIPDM in the low dose range between 4 and 500 nmol kg^{-1} observed in Table 1.

The SPMID methodology is a well established technique for estimating the role of the lung in the extraction and metabolism of circulating vasoactive amines (15). Therefore, the data suggest that the proposed gamma-camera analysis of the first pass of [^{131}I]HIPDM through the lung would be an estimate of the lung extraction efficiency of HIPDM. If a correlation between the extraction efficiency of the lung for HIPDM and lung function can be established, the method could be useful in the early detection of lung dysfunction. However, the dose of HIPDM (10,000 nmol, 4.35 mg) necessary to demonstrate saturability of the lung extraction process was relatively close to doses (30,000 nmol, 13 mg) shown to be toxic to rabbits. In similar experiments in dogs, we have injected up to 4.5 mg (10,300 nmol) without signs of toxicity. Kung et al. (11) injected up to 3.6 mg (8,275 nmol) in rats without any reported toxicity. The maximum dose that would have been injected using the kit method developed by Kung et al. was 1 mg (2,300 nmol). To our knowledge, no toxic effects resulting from HIPDM administration have been reported. In view of the toxicity of HIPDM observed in rabbits, it would be necessary to demonstrate a correlation between lung extraction efficiency and lung function at nontoxic concentrations of HIPDM.

NOTE

ACKNOWLEDGMENTS

REFERENCES

1. Sullivan DC. Radionuclide studies in lung disease. Sem Respir Med 1983; 5:31–42.
2. Mellins RB. Metabolic functions of the lung and their clinical relevance. Am J Roentgenol 1982; 138:999–1009.
3. Gillis CN, Pitt BR. The fate of circulating amines within the pulmonary circulation. Ann Rev Physiol 1982; 44:269–281.
4. Gallagher BM, Fowler JS, MacGregor RR, et al. Evaluation of radiorespirometry for the determination of monoamine oxidase activity in vivo utilizing ^{11}C-octylamine as a substrate. Biochem Pharmacol 1977; 26:1917–1922.
5. Fowler JS, Gallagher BM, MacGregor RR, et al. Radiopharmaceuticals. XIX. ^{11}C-labeled octylamine, a potential diagnostic agent for lung structure and function. J Nucl Med 1976; 17:752–754.
6. Gallagher BM, Christman D, Fowler JS, et al. Radioisotope scintigraphy for the study of the dynamics of amine regulation by the human lung. Chest 1977; 71(suppl 2):282–284.
7. Syrota A, Pascal O, Crouzel M, et al. Pulmonary extraction of C-11 chlorpromazine, measured by residue detection in man. J Nucl Med 1981; 22:145.
8. Rahimian J, Glass EC, Touya JJ, et al. Measurement of metabolic extraction of tracers in the lung using a multiple indicator dilution technique. J Nucl Med 1984; 25:31–37.
9. Touya JJ, Rahimian J, Grubbs DE, et al. A noninvasive procedure for in vivo assay of a lung amine endothelial receptor. J Nucl Med 1985; 26:1302–1307.
10. Tramposch KM, Kung HF, Blau M. Radioiodine-labeled N,N-dimethyl-N'-(2-hydroxy-3-alkyl-5-iodobenzyl)-1,3-propanediamines for brain perfusion imaging. J Med Chem 1983; 26:121.
11. Kung HF, Tramposch KM, Blau M. A new brain perfusion imaging agent: I-123 HIPDM: N,N'-trimethyl-N'- (2-hydroxy-3-methyl-5-iodobenzyl) - 1,3-propanediamine. J Med Chem 1983; 24:66.
12. Pistolesi M, Fazio F, Marini G, et al. Lung distribution and kinetics of I-123 HIPDM: a potential agent for early detection of lung cellular injury [Abstract]. J Nucl Med 1983; 24:P71.
13. Pistolesi M, Fazio F, Marini G, et al. Lung uptake of ^{123}I-HIPDM in man: an index of lung metabolic function. J Nucl Med All Sci 1983; 27:180.
14. Pistolesi M, Miniati M, Ghelarducci L, et al. Lung release of HIPDM: a new index of lung dysfunction for clinical and experimental studies [Abstract]. J Nucl Med 1985; 26:P14.
15. Rickaby DA, Linehan JH, Bronikowski TA, et al. Kinetics of serontonin uptake in the dog lung. J Appl Physiol: Respir Environ Exercise Physiol 1981; 51:405–414.

16. Catravas JD, Gillis CN. Single-pass removal of ^{14}C-5-hydroxytryptamine and ^3H-norepinephrine by rabbit lung, *in vivo*: kinetics and site of removal. *J Pharmacol Exp Ther* 1983; 224:28-33.

17. Smith LL, Wyatt I, Cohen GM. The accumulation of diamines and polyamines into rat lung slices. *Biochem Pharmacol* 1982; 31:3029-3033.

18. Rao SB, Ras KSP, Mehendale HM. The absence of diamine oxidase activity from rabbit and rat lungs. *Biochem J* 1986; 234:733-736.