Evaluation of [Iodine-125] N,N,N′-Trimethyl-N′-[2-Hydroxy-3-Methyl-5-Iodobenzyl]-1,3-Propanediamine Lung Uptake Using an Isolated-Perfused Lung Model

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Lung uptake of N,N,N′-trimethyl-N′-[2-Hydroxy-3-methyl-5-iodobenzyl]-1,3-propanediamine (HIPDM) has been reported, but the mechanism of this process has not yet been established. Thus, single-pass [^{125}I]HIPDM accumulation was studied in rat lungs perfused with a Krebs-Ringer bicarbonate buffer containing 4.5% bovine albumin. Iodine-125 HIPDM lung accumulation was monitored by the percent of extraction per gram of lung tissue. Iodine-125 HIPDM lung uptake did not appear to occur by simple diffusion. As the time of perfusion was increased from 2 to 15 min, the rate of uptake of 2 μM [^{125}I]HIPDM decreased by 40%. During a 2-min perfusion, 98.6% ± 6.7 (n = 8) extraction was observed with 2 μM [^{125}I] HIPDM, but only 38% ± 2.0 (n = 3) was extracted when the [^{125}I]HIPDM concentration was 1 mM. The addition of 1 mM chlorpromazine, propranolol or imipramine also decreased [^{125}I]HIPDM lung uptake to 43.0% ± 1.5, 51.4% ± 2.2, and 49.8% ± 0.8, respectively, (each n = 4 − 6, $p < 0.001$). Cold (4°C) had little effect on pulmonary accumulation (77.7% ± 7.4, n = 5, $p < 0.01$), and the addition of ouabain or the use of sodium-free medium had no effect. Thus, pulmonary [^{125}I]HIPDM accumulation does not appear to occur by sodium-dependent active transport. Rather, its uptake appears to be similar to the uptake of other basic amines, such as propranolol and imipramine, which are known to bind by physico-chemical interactions to pulmonary endothelial cell membranes and reflect pulmonary vascular surface area.

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In the past 10 years, numerous reviews have stressed the function of the lungs as a metabolic organ that is capable of activating or inactivating various circulating hormones and mediators (*1–6*). Thus, compounds as different as peptides, amines, nucleotides, and lipids are activated, degraded, and/or taken up by the lungs. These phenomena often are quantitatively important enough to create large arteriovenous differences in the concentrations of some circulating bioactive substances. The lung uptake of biogenic amines such as serotonin and norepinephrine involves an energy-requiring sodium-dependent active transport system located on the endothelial cell membrane (*7–10*). In contrast, the pulmonary accumulation of basic lipophilic amines such as imipramine and propranolol appears to result mainly from saturable and specific physico-chemical binding on endothelial cell membranes (*11–14*).

The pulmonary extractions of radiolabeled serotonin and propranolol have been proposed for assessment of pulmonary endothelial cell function and vascular surface area, respectively (*15–17*). Unfortunately, such measurements have limited clinical utility because they require carbon-14- (^{14}C) and tritium-labeled compounds. In addition, the procedures are invasive (arterial catheters are needed for measurements). However, because of the large number of lung disorders that involve the endothelial cell, it is important to develop compounds that permit numerous sequential measurements.

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Touya et al. (*18*) have suggested that [^{123}I] N-isopropyl-p-iodoamphetamine (IMP) may be useful for in vivo assessment of a lung amine endothelial receptor. We wished to evaluate this possibility further in a model system using an iodine-125-(^{125}I) labeled amine. Iodine-125 IMP was not available to us. However, radioiodinated N,N,N′-trimethyl-N′-[2-hydroxy-3-methyl-5-iodobenzyl]-1,3-propanediamine (HIPDM) is a similar compound that has been used as a brain-perfusion imaging agent (*19*), and is proposed for pancreas imaging as well (*20*). High lung uptake has been reported also (*19,20*). Thus, HIPDM was labeled with [^{125}I] and used as our test compound.

The present study was undertaken to investigate the mechanism and extent of [^{125}I]HIPDM accumulation by studying the behavior of [^{125}I]HIPDM in isolated perfused rat lungs. This allowed us to evaluate the pulmonary accumulation of HIPDM in the presence of many of the same inhibitors used to study the behavior of ^{14}C or tritiated compounds by previous investigators (*7,8,13,14*).

MATERIALS AND METHODS

Perfusate Media

The perfusion medium (*7*) consisted of Krebs-Ringer bicarbonate buffer, containing 5m*M* glucose and 4.5% bovine serum albumin, Cohn Fraction V, prepared daily. This solution was equilibrated with 5% CO_2 and 95% O_2. The pH of the albumin solution was adjusted by the addition of 1*N* NaOH so that the final pH was 7.4. During all experiments, the medium remained constantly equilibrated with the gas mixture and control pH measurements were done. To study the effect of sodium-free medium on [^{125}I]HIPDM accumulation, NaCl was replaced by isotonic sucrose (0.25 m*M*) and $NaHCO_3$ by Tris (25 m*M*), the pH of the final solution being adjusted to 7.4 by adding 1*N* HCl. The sodium concentration in the sodium-free medium was ~1 m*M*. Appropriate amounts of unlabeled HIPDM were added when high concentrations were desired.

Iodine-125 was used because its long half-life permitted numerous experiments. Radioiodination of HIPDM (0.456 mCi/ml, 0.5 mg/ml) and thin layer chromatography were carried out according to the method developed by Kung et al. (*20*). The iodination yields always were >95%. In preliminary experiments, we found that perfusion of [^{125}I] sodium iodide resulted in <3% lung extraction during a 2-min perfusion. Therefore, because <3% of the already small amount of free radioiodide was extracted by the lung, no correction for free [^{125}I] sodium iodide was made in any subsequent experiments.

Perfusion System

The perfusion system (Fig. 1) used for measuring amine uptake was the same as described previously in detail by Junod

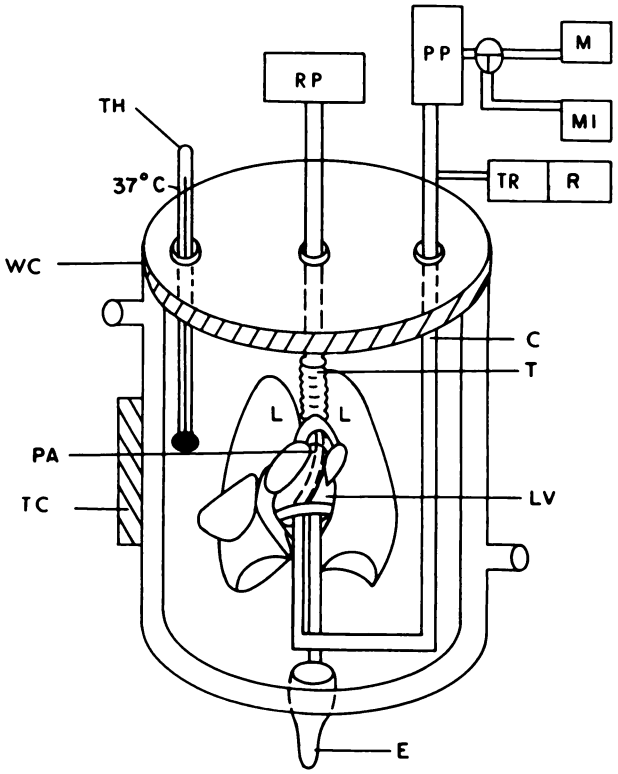

FIGURE 1
Schematic diagram of apparatus for perfusion of isolated lung. Peristaltic pump (PP) reservoir for the standard medium (M) and the medium with the labeled amine (MI). The transducer (TR) and recorder (R) are connected to the pulmonary artery inflow catheter (C) which goes through the right ventricle inside the pulmonary artery (PA). Through the left ventricle (LV), the medium goes directly to the bottom of the water-jacketed chamber (WC) and is collected at the exit port (E). The lungs (L) are ventilated by a respiratory pump (RP) connected to the trachea (T). The chamber is kept at 37°C using a temperature controller (TC).

(*7*). After anesthesia with pentobarbital injected intraperitoneally (50 mg/kg), a tracheostomy was performed on each adult pathogen-free male Sprague-Dawley rat (n = 72, 180–240 g). The animals were then mechanically ventilated with a humidified gas mixture of 95% O_2 and 5% CO_2. The end-expiratory pressure was set to 2 cm H_2O, the frequency was 60 breaths/min and the tidal volume 1 ml/100 g of body weight. The thorax was opened and a loose ligature was placed around the pulmonary artery and the aorta. Right and left ventricular incisions were made and a cannula was inserted immediately into the pulmonary artery through the right ventricle. The cannula was secured by tying the ligature while the perfusion began. The lungs then were isolated (i.e., dissected free from the thorax), perfused at the flow rate of 10 ml/min and placed in a closed water-jacketed chamber maintained at 37°C and saturated with water vapor. During each lung perfusion, the mean pulmonary artery perfusion pressure at constant flow rate was continuously monitored with a pressure transducer and recorded with a direct-writing oscil-

lograph. The zero reference for the perfusion pressure was determined at the beginning of each experiment by maintaining the experimental flow rate of perfusate with the lung out of the circuit.

The perfusion system consisted of two independent sets of tubing from separate reservoirs maintained at 37°C. A three-way stopcock was provided in the circuit so that the lung could be perfused from either reservoir without interruption of flow. The lungs were perfused with the standard medium described above (first reservoir) for 10 min (equilibration period). For each set of saturation experiments, a solution containing various concentrations (range: 0.03 μM–1 mM) of labeled amine (second reservoir) was infused for 2 min.

For experiments in the presence of inhibitors, 2 μM of [^{125}I] HIPDM was added to the appropriate test solution. Like previous works by Junod and others (*7–14*), we studied the effect of cold (4°C), sodium-free media and ouabain on [^{125}I] HIPDM accumulation. Cold nonspecifically reduces the activity of enzyme systems (*9*). The absence of sodium deactivates the sodium-dependent ATPase system for intracellular transport, and the chemical ouabain is well-known to "poison" the sodium, potassium-dependent ATPase system (*21*). Thus, all these inhibitors were used to assess the degree to which [^{125}I] HIPDM pulmonary accumulation depended on the energy-requiring sodium-dependent active transport.

To study the degree to which [^{125}I]HIPDM accumulation competed with other amines (whose accumulation mechanisms are more well understood), we infused [^{125}I]HIPDM (2 μM) along with varying concentrations of chlorpromazine, imipramine, and propranolol. These experiments also were designed to simulate those previously performed by other investigators (*7,8,13,14*) with tritiated or ^{14}C-labeled amines.

After the infusion was completed, the lungs were dissected free from the trachea and other tissues, and the intravascular fluid was eliminated as the lungs (lobe by lobe) were blotted dry. Then the lungs were weighed and counted. To evaluate the possibility of fluid accumulation during the experiment, we determined a percent lung dry-to-wet weight ratio at the end of each experiment. The dry-to-wet weight ratio approach has been used by others (*22,23*) and is considered as an accurate control parameter when the isolated-perfused lung model is used. In preliminary results, this ratio was found to be 19.4% ± 1.2 (mean ± s.d., n = 11). Only 8 experiments demonstrated a ratio below 17.0% (mean-2 s.d.), and they were excluded from further analysis.

Analytical Procedure

The radioactive content of inflow samples (1 ml) and the whole lungs were measured in a Na(I) well-type scintillation counter (counting of X and gamma emissions 28–35 keV of ^{125}I, 60-day half-life). The [^{125}I]HIPDM uptake in the lung was expressed as: (a) the tissue/medium ratio (T/M), specifically: cpm.g^{-1} lung tissue/cpm.ml^{-1} inflow; (b) the total [^{125}I] HIPDM lung uptake (μmoles/g lung tissue); and (c) the percent extraction per gram of lung tissue.

Results were expressed as the arithmetic mean ± 1 s.d. The t-test was applied to determine the significance of differences between two means. One-way analysis of variance was applied to determine the significance of the variations of T/M among the different doses of [^{125}I]HIPDM. Statistical significance was defined as $p < 0.05$.

RESULTS

Effect of Duration of Perfusion on the Pulmonary Uptake of HIPDM

Expressed as the rate of uptake (T/M.min^{-1}, μmol/g or % uptake/g of perfusion with 2 μM [^{125}I]HIPDM), the effect of duration of perfusion on pulmonary accumulation was significant (Table 1); the rate of uptake decreasing by 40% as the time of perfusion increased from 2 to 15 min. A 2-min perfusion with [^{125}I]HIPDM was used in all subsequent experiments.

Effect of Substrate Concentration on the Pulmonary Uptake of HIPDM

Increasing the [^{125}I]HIPDM concentration from 0.03 μM to 1 mM did not result in a linear increase of [^{125}I] HIPDM lung uptake (Table 2). A significant decrease in uptake appeared at a concentration of 100 μM (Fig. 2) but no plateau was obtained. From the 98.6% ± 6.7, (n = 8) lung extraction in the control experiment (2 μM), a decrease to 38% ± 2.0, (n = 3, $p < 0.001$) was observed with a 1 mM concentration of [^{125}I]HIPDM. Using these data and the Michaelis-Menten kinetic equation, the apparent Michaelis-Menten constant (K_m) and the apparent maximum initial velocity (Vmax) were estimated to be K_m = 611 μM and V_{max} = 6.1 μmol/g lung/min.

Effect of Specific Perfusates on Pulmonary Uptake of HIPDM

In control experiments, we found a 98.6% ± 6.7, (n = 8) lung extraction of [^{125}I]HIPDM, which decreased to 77.7% ± 7.4, (n = 5, $p < 0.01$) when the perfusate temperature was 4°C. Therefore, a slight inhibitory effect of low temperature on the pulmonary accumulation of [^{125}I]HIPDM did exist (Table 3). On the other hand, replacement of sodium in the perfusion-medium by sucrose, as well as addition of 1 mM ouabain in the medium had no effect (Table 3).

TABLE 1
Effect of Time of Perfusion of 2μM [^{125}I]HIPDM on Its Accumulation by Isolated Rat Lungs

Time of perfusion (min)	n	T/M/min ± s.d.	Percent extraction/g lung ± s.d.	μmol/g lung ± s.d.
2	8	9.86 ± 0.67	98.6 ± 6.7	0.040 ± 0.003
6	4	8.94 ± 0.14*	89.4 ± 1.4*	0.107 ± 0.002†
15	3	5.94 ± 0.39†	59.4 ± 3.9†	0.179 ± 0.012†

*$p < 0.05$.
†$p < 0.001$.

TABLE 2
Effect of Various [^{125}I]HIPDM Concentrations (μM) on Its Accumulation by Isolated Rat Lungs

[^{125}I]HIPDM (μM)	n	T/M ± s.d.	% Extraction /g lung ± s.d.	μmol/g lung ± s.d.
0.03	6	18.8 ± 1.0	94.1 ± 5.0	0.001 ± 0.0002
1.2	5	19.3 ± 1.9	96.6 ± 9.3	0.023 ± 0.002
2.0	8	19.7 ± 1.2	98.6 ± 6.7	0.040 ± 0.003
10.0	6	19.4 ± 2.6	97.2 ± 12.9	0.194 ± 0.026
100.0	4	17.2 ± 1.0*	86.0 ± 5.7*	1.720 ± 0.114*
1000.0	3	7.6 ± 0.3†	38.0 ± 2.0†	7.592 ± 0.403†

*$p < 0.01$.
†$p < 0.001$.

TABLE 3
Effect of Various Experimental Conditions on the Accumulation of 2 μM HIPDM by Isolated Rat Lungs

Experimental conditions	n	T/M ± s.d.	% Extraction /g lung ± s.d.	μmol/g lung ± s.d.
(2μM) 37°C	8	19.7 ± 1.2	98.6 ± 6.7	0.040 ± 0.003
(2μM) 4°C	5	17.3 ± 1.0*	77.7 ± 7.4*	0.031 ± 0.003*
(1mM) 37°C	3	7.6 ± 0.3†	38.0 ± 2.0†	7.592 ± 0.403†
Sodium-free	5	18.5 ± 1.7	92.4 ± 9.9	0.037 ± 0.004
Chlorpromazine				
0.1 mM	4	20.7 ± 2.1	99.5 ± 10.4	0.044 ± 0.004
1.0 mM	4	8.6 ± 0.3†	43.0 ± 1.5†	0.017 ± 0.001†
Propanolol				
1.0 mM	6	10.3 ± 0.4†	51.4 ± 2.2†	0.021 ± 0.001†
Imipramine				
1.0 mM	4	10.0 ± 0.1†	49.8 ± 0.8†	0.020 ± 0.001†
Ouabain				
1.0 mM	5	19.5 ± 1.4	97.5 ± 7.7	0.039 ± 0.003

*$p < 0.01$.
†$p < 0.001$.

Effect of Drugs on Pulmonary Uptake of HIPDM

Table 3 shows the inhibitory effects of high concentrations of lipophilic basic amines (imipramine, propranolol and chlorpromazine). These effects were similar to the effect of carrier HIPDM itself. The 98.6% ± 6.7, (n = 8) lung extraction of [^{125}I]HIPDM in control experiments decreased to 43.0% ± 1.5, (n = 4, $p < 0.001$) with addition of 1 mM of chlorpromazine in the medium, to 51.4% ± 2.2, (n = 6, $p < 0.001$) with addition of 1 mM propranolol, to 49.8% ± 0.8, (n = 4, $p < 0.001$) with addition of 1 mM imipramine and to 38.0% ± 2.0, (n = 3, $p < 0.001$) with addition of 1 mM unlabeled HIPDM.

DISCUSSION

The isolated-perfused lung model has not been used previously to characterize *mechanisms* of lung extraction of gamma-emitting radionuclides, but is considered by others (*22,23*) as the appropriate tool to study intermediary metabolism and drug uptake in the lung (*7,8, 13,14*). It can be considered as a two-compartment model, i.e., the pulmonary intravascular and extravascular compartments. Thus, we evaluated the nature of the removal of [^{125}I]HIPDM from the intravascular compartment. This removal was characterized by the T/M ratio. The numerator (T) represented the total radioactivity of the lungs at the end of each experiment, and M was the concentration of [^{125}I]HIPDM in the (intravascular) perfusate.

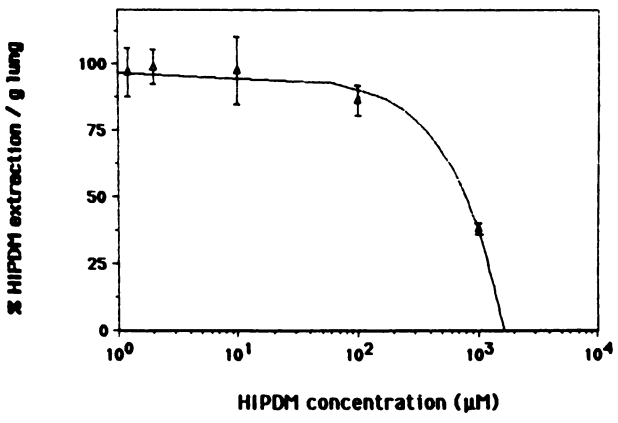

FIGURE 2
Relationship between the HIPDM concentration (μM) in the perfusion medium and the percent of HIPDM extraction per gram of lungs. Because the HIPDM concentrations range between 0.01 μM and 1 mM, a logarithmic scale was used for the x-axis.

Extraction of circulating substances by the lung could occur by passive diffusion through the capillary wall, by facilitated diffusion (carrier-dependent), by an active transport through the cell (energy-dependent) or by binding to the endothelial cell membrane (without transport). Basic lipophilic amines share common characteristics of lung accumulation that are markedly different from the uptake of biogenic amines. A sodium-dependent, carrier-mediated transport has been described for the biogenic amines (serotonin and norepinephrine), which are metabolized by monoamineoxidase within endothelial cells (*7–10,21,24*). On the other hand, the mechanisms of lung extraction of basic lipophilic amines such as chlorcyclizine, methadone, imipramine, propranolol, and amphetamine are saturable with specific binding-sites and different affinities (*11–14*). The two processes of accumulation of biogenic amines and basic amines also present different kinetic characteristics i.e., the K_m and V_{max} of serotonin are considerably lower than the apparent K_m and V_{max} for the accumulation of imipramine (6.2 μM against ~200 μM for the K_m, and 19 mμmol/g/min against 2 μmol/g/min for the V_{max}) (*7,13*).

The 40% decrease in rate of [^{125}I]HIPDM lung uptake over time and the 60% decrease in [^{125}I]HIPDM lung uptake when the medium [^{125}I]HIPDM concentration

is increased from 2 to 1,000 μM provide evidence for the presence of a saturable lung uptake process. This trend toward saturation strongly suggests that passive diffusion is not the uptake mechanism. These results compare favorably with the 39% decrease in rate of imipramine uptake between 3 and 12 min of perfusion shown by Junod (*13*) and with the 25% decrease in rate of propranolol uptake between 2 and 10 min found by Dollery and Junod (*14*). Although increasing the [^{125}I] HIPDM concentration did not result in a plateau in uptake; this may be able to be explained by the large capacity for endothelial cell binding, similar to propranolol (*14*). It strongly suggests that there is *specific* mechanism of lung removal.

Low temperature inhibits enzymatic processes (*9*) and ouabain inhibits potassium-activated processes (*21*). If an enzymatic process were involved in the removal of [^{125}I]HIPDM, we should expect a dramatic decrease of its extraction with low temperature. The slight decrease in [^{125}I]HIPDM extraction seen in the presence of cold also has been reported for propranolol (*14*), where it is postulated to be related to cold-induced reduction in the avidity of certain high-affinity sites of binding. The absence of effect of sodium-free medium, ouabain and the slight effect of low temperature are strong evidence against an active process involving a sodium-dependent carrier-mediated transport such as the one described for serotonin (*7,9,10,24*) or norepinephrine (*8,9,21*).

The magnitudes of the imipramine, propranolol or chlorpromazine inhibition of [^{125}I]HIPDM accumulation are similar to the results found for propranolol inhibition with chlorpromazine or imipramine (*14*). In addition, the estimated kinetic constants of the [^{125}I] HIPDM accumulation compared favorably with the kinetic constants reported for imipramine accumulation i.e., 611 μM against ~200 μM for the K_m, and 6.1 μmol/g/min against 2 μmol/g/min for the V_{max}) (*13*). The fact that these basic amines also inhibit [^{125}I] HIPDM lung uptake strongly suggests that passive diffusion is not the uptake mechanism.

Our findings suggest that the pulmonary uptake of [^{125}I]HIPDM is not passive diffusion or an energy requiring sodium-dependent, carrier-mediated transport, but that it exists a specific saturable process. These results do not differentiate between a facilitated diffusion process (carrier-dependent) or only specific binding on endothelial cell membranes. The similarities in uptake kinetics of basic lipophilic amines, as well as their cross-inhibition, suggest that the [^{125}I]HIPDM mechanism of lung uptake is similar to that of imipramine (*13*) or propranolol (*14*). Although the exact pharmacological nature of this binding process and the location of the binding sites still are unknown, it has been suggested that the sites are phospholipids on endothelial cell membranes (*13,14,25*). Our results suggest that [^{125}I]HIPDM lung uptake might occur by the same process.

Using a scintigraphic method, Touya et al. (*18*) showed that the process of IMP lung uptake was saturable and has a large capacity (amount of IMP bound at saturation estimated to be 30 mg). In addition, Akber et al (*26*) found propranolol to decrease the lung uptake of IMP in a dose-related ratio and they suggested that propranolol competes with IMP for the same lung endothelial binding sites. Their results agree with the finding of Anderson et al. (*12*) who, using an isolated-perfused lung model, demonstrated that amphetamine, a similar molecule to IMP or HIPDM, is removed by the lung through a saturable process that is not an active transport, energy-requiring or sodium-dependent (no effect of ouabain). Our findings with [^{125}I]HIPDM are similar to these IMP results, and suggest that the mechanisms of IMP and HIPDM pulmonary accumulation are similar.

The results of this study suggest ways in which ^{123}I-labeled HIPDM or IMP extraction might be used for evaluation of lung disorders in patients. Dargent et al. (*16*) and Morel et al. (*17*) studied patients undergoing surgery with extracorporeal circulation and patients with adult respiratory distress syndrome using the standard triple-indicator dilution technique ([^{3}H] propranolol, indocyanine green and [^{14}C] serotonin). They concluded that serotonin lung extraction is related to lung endothelial cell function and that propranolol reflects the pulmonary circulation available to the blood-pool. The propranolol-like uptake mechanism of [^{125}I]HIPDM suggests that a decrease in its uptake will be related to decreases in the number of binding sites available in the pulmonary vascular bed. Indicator-dilution techniques have been used to study the effect of pulmonary artery occlusion or shock lung on the pulmonary removal of propranolol. Pang et al. (*27*) showed that propranolol extraction depended on the surface area of pulmonary endothelium to which the drug was exposed when partial occlusion of the pulmonary circulation was made. The isolated-perfused lung model will be an important tool to evaluate variations of HIPDM or IMP extraction related to lung disorders in which a loss of vascular surface area is suspected. Furthermore, the model is easy to use, inexpensive to set up, and yields reproducible results in a controlled experimental setting. Such lung model work could provide a focal point for development of new pharmaceuticals for in vivo imaging of the nonrespiratory functions of the lung.

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REFERENCES

1. Philpot RM, Anderson MW, Eling TE. Uptake, accumulation and metabolism of chemicals by the lung. In: Backhle YS, Vane JR, eds. Metabolic functions of the lung. New-York; Marcel Decker, 1977; 4:123–171.
2. Datta H, Stubes WA, Alberti KGMM. Substrate utilization by the lung. In: Metabolic activities of the lung. Ciba foundation symposium. *Exerpta Medica.* 1982; 85–104.
3. Hechtman HB, Shepro D. Lung metabolism and systemic organ function. *Circ Shock* 1982; 9:457–467.
4. Said SI. Metabolic functions of the pulmonary circulation. *Circ Res* 1982; 50:326–333.
5. Block ER. Early metabolic changes in response to lung injury: extrapolation from animals to humans. *J Toxicol Environ Health* 1984; 13:369–386.
6. Junod AF. 5-hydroxytryptamine and other amines in the lung. In: Handbook of physiology. Section 3: The respiratory system, volume 1. Circulation and nonrespiratory functions. pp. 337–349. Edited by Fishman AP, Fisher AB, eds. Bethesda, Maryland. American Physiological Society, 1985.
7. Junod AF. Uptake, metabolism and efflux of ^{14}C-5-hydroxy tryptamine in isolated perfused rat lungs. *J Pharmacol Exp Ther* 1972; 183:341–355.
8. Nicholas TE, Strum JM, Angelo LS, et al. Site and mechanism of uptake of H^3-1-norepinephrine by isolated perfused rat lungs. *Circ Res* 1974; 35:670–680.
9. Iwasawa Y, Gillis CN, Aghajanian G. Hypothermic inhibition of 5-hydroxythryptamine and norepinephrine uptake by lung: cellular location of amines after uptake. *J Pharmacol Exp Ther* 1973; 186:498–507.
10. Pickett RD, Anderson MW, Orton TC, et al. The pharmacodynamics of 5-hydroxytryptamine uptake and metabolism by the isolated perfused rabbit lung. *J Pharmacol Exp Ther* 1975; 194:545–553.
11. Orton TC, Anderson MW, Pickett RD, et al. Xenobiotic accumulation and metabolism by isolated perfused rabbit lungs. *J Pharmacol Exp Ther* 1973; 186:482–497.
12. Anderson MW, Orton TC, Pickett RD, et al. Accumulation of amines in the isolated perfused rabbit lung. *J Pharmacol Exp Ther* 1974; 189:456–466.
13. Junod AF. Accumulation of ^{14}C-imipramine in isolated perfused rat lungs. *J Pharmacol Exp Ther* 1972; 183:182–187.
14. Dollery CT, Junod AF. Concentration of (+/−)-propranolol in isolated, perfused lungs of rat. *Br J Pharmacol* 1976; 57:67–71.
15. Gillis CN, Cronau LH, Mandel S, et al. Indicator dilution measurement of 5-hydroxytryptamine clearance by the lung. *J Appl Physiol* 1979; 46:1178–1183.
16. Dargent F, Neidhart P, Bachmann M, et al. Simultaneous measurement of serotonine and propranolol pulmonary extraction in patients after extracorporeal circulation and surgery. *Am Rev Respir Dis* 1985; 131:242–245.
17. Morel DR, Dargent F, Bachmann M, et al. Pulmonary extraction of serotonine and propanolol in patients with adult respiratory distress syndrome. *Am Rev Respir Dis* 1985; 132:479–484.
18. 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.
19. Kung HF, Tramposh KM, Blau M. A new brain perfusion imaging agent: (I-123) HIPDM: N,N,N′-trimethyl-N′-(2-hydroxy-3-methyl-5-iodobenzyl)-1,3-propanediamine. *J Nucl Med* 1983; 24:66–72.
20. Yamamoto K, Som P, Srivastava S, et al. Pancreas accumulation of radioiodinated HIPDM in mice and rats. *J Nucl Med* 1985; 26:765–769.
21. Bogdanski DE, Brodie BB. The effects of inorganic ions on the storage and uptake of ^{3}H-norepinephrine by rat hearts slices. *J Pharmacol Exp Ther* 1969; 165:181–189.
22. Woods HF, Meredith A, Tucker GT, et al. Methods for the study of lung metabolism. In: Metabolic activities of the lung. Ciba foundation symposium. *Excerpta Medica* 1982; 61–83.
23. Watkins CA, Rannels DE. In situ perfusion of rat lungs: stability and effects of oxygen tension. *J Appl Physiol* 1979; 47:325–329.
24. Kjellstrom T, Ahlman H, Dahlstrom A, et al. The uptake of 5-hydroxytryptamine in endothelial cells cultured from the pulmonary artery in rats. *Acta Physiol Scand* 1984; 120:243–250.
25. Huunan-Seppala A. Binding of propanolol and chlorpromazine by mitochondrial membranes. *Acta Chem Scand* 1972; 26:2713–2733.
26. Akber SF, Glass EC, Bennett LR. Scintigraphic demonstration and quantification of pharmacological inhibition of pulmonary uptake of N-isopropyl-1-123-p-iodoamphetamine by propranolol [Abstract]. *J Nucl Med* 1985; 26:P14.
27. Pang JA, Blackburn JP, Butland RJA, et al. Propanolol uptake by dog lung: effect of pulmonary artery occlusion and shock lung. *J Appl Physiol* 1982; 52:393–402.