DETERMINATION OF MYOCARDIAL BLOOD FLOW IN THE ANESTHETIZED DOG AFTER A BOLUS OF ^{84}Rb

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A method using double coincidence counting devices and ^{84}Rb to measure myocardial blood flow in experimental animals and in man has been developed in this laboratory (*1–4*). Initially myocardial blood flow was determined by constant intravenous infusion of ^{84}Rb. During 7-min test periods, the rate of myocardial uptake was measured by precordial external coincidence detectors. A disadvantage of constant infusion is that rapid changes in flow cannot be assessed. If the method is accurate, rapid intravenous injection of a bolus of ^{84}Rb, as carried out by Donato (*5*) and by Knoebel and McHenry (*6,7*), lets one measure myocardial blood flow during a short time (less than 90 sec). In addition, the calculations of myocardial clearances are easy, and a comparison with flow measured according to the direct Fick principle is more readily accomplished.

Reports in the literature have not yet clearly answered the principal question: Is the myocardial clearance of ^{84}Rb obtained with coincidence counters after a bolus injection of the isotope a true measure of myocardial blood flow? In this paper we report experiments in a large series of dogs in which this question is examined. In addition, our study deals with the effects of two cardiotonic drugs (isoproterenol and norepinephrine) on the myocardial clearance of ^{84}Rb in the anesthetized dog.

METHODS

The principle of this method using a single bolus injection of ^{84}Rb is based on the observations of Love (*8*) and Sapirstein (*9,10*). In 1954 Love tried to quantitate the exchange of rubidium in organs in comparison to potassium after injections of ^{86}Rb and ^{42}K. The myocardial uptake of ^{86}Rb was measured over a long period of time. Sapirstein in 1956 and 1958 injected a single bolus of ^{42}K or ^{86}Rb into a large group of animals. The animals were killed at different intervals up to 120 sec after injection, and the activity of each organ and the carcass was measured. During this time interval, the organ uptake of both isotopes was a constant percentage of the total-body uptake. Brain, however, showed its maximum isotope content earlier than 9 sec following injection and declined precipitously thereafter (*10*).

Sapirstein deduced that the organ extraction ratio of rubidium must be equal to the total-body extraction ratio during the period of observation. Consequently, it follows that myocardial uptake is related to body uptake as organ blood flow is related to cardiac output. Thus

$$\frac{U_H(t)}{U_B(t)} = \frac{MBF}{CO} \qquad (1)$$

in which $U_H(t)$ is total myocardial uptake of ^{84}Rb following the injection (cpm) and $U_B(t)$ is total-body uptake of ^{84}Rb following injection (cpm)—which equals the amount of ^{84}Rb injected. MBF is total myocardial blood flow (cc/min) and CO equals cardiac output (cc/min). Solving for coronary blood flow,

$$MBF = \frac{U_H(t)\ CO}{U_B(t)}. \qquad (2)$$

Cardiac output is calculated by the Stewart-Hamilton formula (*11*); the result is

$$CO = \frac{U_B(t)}{\int_0^\infty A_1(t)dt} \qquad (3)$$

in which $\int_0^\infty A_1(t)dt$ is the integrated concentration of ^{84}Rb during the primary arterial circulation (counts/cc). Substituting Eq. 3 for cardiac output in Eq. 2, one gets

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$$MBF = \frac{U_H(t)}{\int_0^\infty A_1(t)dt} = MCL \qquad (4)$$

in which MCL is myocardial clearance.

Consequently, myocardial blood flow (MBF) equals myocardial uptake of ^{84}Rb divided by the integrated arterial concentration prior to recirculation; this is identical with myocardial clearance (MCL) of ^{84}Rb in cc/min.

The validity of this assumption can now be tested by comparing myocardial clearance as defined by Eq. 4 to myocardial blood flow as determined by the Fick principle (*12*) which is given by

$$MBF = \frac{U_H(t)}{\int_0^t A(t)dt - \int_0^t V(t)dt} \qquad (5)$$

in which $\int_0^t A(t)dt$ represents the integrated arterial concentration of ^{84}Rb during a defined period of time (counts/cc) and $\int_0^t V(t)dt$ represents the integrated concentration of ^{84}Rb in coronary sinus blood, determined for the same period of time and expressed in counts/cc.

In 1964 Donato (*5*) developed a method for measuring myocardial blood flow with a single bolus injection of ^{86}Rb or ^{42}K; his method is based on the findings of Love and Sapirstein mentioned previously. A major disadvantage of Donato's method was that activity of blood in the cardiac chambers interfered with the measurement of the uptake of the isotope by the heart muscle, necessitating the injection of a second isotope, ^{131}I-albumin, to determine intracavitary activity (*13,14*). In addition, the detection of radiation from ^{86}Rb necessitates complicated collimation, making the separation of the activity of the heart muscle from that of surrounding structures difficult. In contrast, ^{84}Rb, a positron emitter, lets one isolate myocardial activity with the double coincidence counting system developed in this laboratory (*1*). McHenry and Knoebel (*6*) in 1967 combined the rapid bolus technique of Donato with the use of the double coincidence counting technique.

^{84}Rb decays by positron emission 19% of the time. The positron travels 1–2 mm inside the body before annihilation with an electron, producing two 0.51-MeV gamma photons directed 180 deg apart. The half-life of ^{84}Rb is 33 days which is convenient for clinical and experimental use. The detection system (CO-INSITRON, American Science and Engineering, Inc., Cambridge, Mass.) consists of pairs of coincidence-counting crystals placed anteriorly and posteriorly to the chest 180 deg apart. One pair of sodium iodide scintillation detectors (4 in. in dia) is aligned anterior and posterior to the heart (H). A second pair of detectors (2 in. in dia) is placed anterior and posterior to the right chest (B). The activity seen by this pair of detectors (B) is electronically subtracted from the heart and chest activity (H); this results in the isolation of myocardial activity alone (H-B). For a count to be registered, a gamma photon pair emitted at 180 deg must strike the anterior and posterior detectors almost simultaneously (within 1 μsec). Hence, only activity which is present between two corresponding detectors will be recorded. Thus, the major advantage of the double coincidence counting technique is that without elaborate collimation it is possible to define the radiation from the heart muscle independent of surrounding structures.

The other advantages of the double coincidence counting system and ^{84}Rb are that: (1) the field of view of a detection system is precisely defined; (2) background counting rates arising from natural radioactivity, cosmic-ray activity and radioactive contaminants in the room are negligible; and (3) for equal injected activities of rubidium and identical detector fields of view, the coincidence method using the ^{84}Rb gives five times the counting rate that ^{86}Rb would yield using the standard single technique.

Healthy mongrel dogs weighing 10–26 kg were anesthetized with sodium pentobarbital (30 mg/kg). The animals were ventilated with room air through endotracheal tubes. They were placed in the left-lateral position so that the outline of the heart (previously drawn on the chest wall during fluoroscopy) could be positioned between the 4-in. pair of coincidence-counting detectors. The "background" (B) detectors were not used in dogs because of the left-lateral position of the animal during the test and the shape of its chest. Natural background was determined before the first injection of ^{84}Rb. The residual background activity was obtained before subsequent injections.

The bolus of ^{84}Rb (0.4 μCi/lb) was injected into the inferior vena cava through a catheter inserted into a femoral vein. Myocardial activity was recorded for 4½ min following each injection. However, only the precordial activity between 90 and 270 sec was used to determine the myocardial uptake. As shown later, the output of the detector pair remained constant for that period of time. This illustrates that the initial distribution of ^{84}Rb into the organs is complete 90 sec after injection. Therefore, it is possible after 90 sec to measure the accumulated myocardial activity from the bolus injection. Precordial counts representing myocardial uptake were summed at 3-sec intervals and recorded as a direct printout. The

sum of these counts from 90 to 270 sec (3 min) was converted to average activity per minute.

Arterial blood was withdrawn from the descending aorta through a catheter introduced through a femoral artery. This blood was circulated through a polyethylene coil in a well counter and reinfused into the distal femoral artery by a Roller-Pump (Model 3500, Sarns, Inc., Ann Arbor, Mich.) at a constant flow of 40 cc/min. From the time of injection until 120 sec, the activity of the arterial blood detected in the well counter was printed at 3-sec intervals by the same counting apparatus. To determine the myocardial blood flow according to the Fick principle (Eq. 5), the integrated arterial concentration of ^{84}Rb ($\int_0^{120} A(t)dt$) over a period of 2 min after injection was used. This period was chosen because arterial and venous activity became constant and equal during this time, letting us determine the arteriovenous difference following the slug injection of ^{84}Rb.

To determine coronary venous activity, a catheter was directed, under fluoroscopic control, from the external jugular vein into the coronary sinus. Coronary sinus blood was collected by gravity in a flask over the same 2-min period after ^{84}Rb injection. Three 1-cc samples were then pipetted from this flask into test tubes. The activity in each aliquot was later determined in the well counter used to register activity in arterial blood.

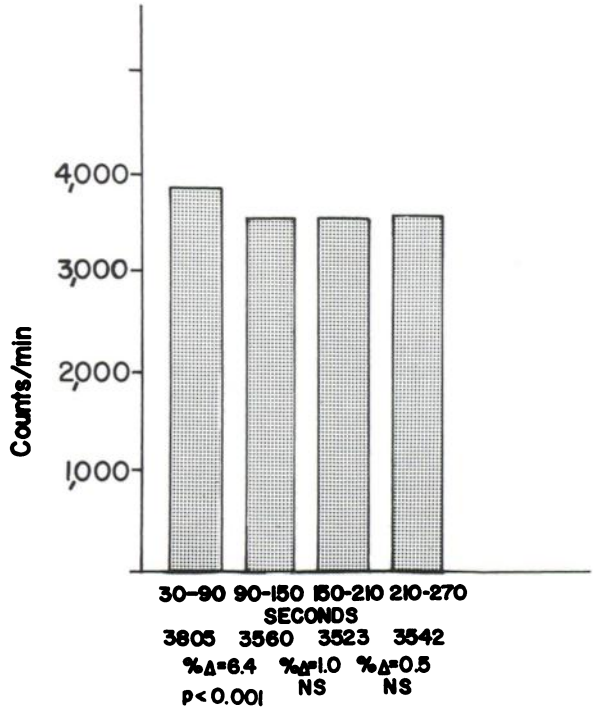

FIG. 1. Constancy of uptake. Each bar represents the average counting rate (cpm) in 30 dogs beginning 30 sec after slug injection of ^{84}Rb.

TABLE 1. MYOCARDIAL UPTAKE FOLLOWING INJECTION OF ^{84}Rb BOLUS

Experiment No.	Counting rate (cpm) 30–90 (sec)	90–150 (sec)	150–210 (sec)	210–270 (sec)
50	5,831	5,752	5,734	5,758
51	3,933	3,742	3,785	3,896
52	2,464	2,542	2,531	2,601
53	3,334	2,840	2,784	2,830
54	3,395	3,376	3,438	3,396
55	3,327	3,214	3,239	3,117
56	3,733	3,657	3,675	3,686
57	4,511	4,337	4,335	4,274
58	3,399	3,107	3,007	3,074
59	4,938	4,698	4,456	4,538
60	5,671	4,921	4,824	4,855
61	3,735	3,403	3,320	3,083
62	2,290	2,088	2,090	2,263
63	2,916	2,891	2,866	2,930
64	3,016	2,910	2,865	2,901
65	5,442	4,839	4,802	4,683
66	6,252	5,696	5,518	5,487
67	3,396	3,164	3,166	3,299
68	3,130	3,064	3,035	3,053
69	4,713	4,263	4,035	4,095
70	4,548	4,014	3,799	3,886
71	4,935	4,923	5,050	4,900
72	3,582	3,346	3,249	3,365
73	3,387	2,881	2,927	2,840
74	3,744	3,507	3,465	3,479
75	3,198	2,939	2,965	2,981
76	2,650	2,400	2,474	2,480
77	3,115	2,959	2,802	2,921
78	2,067	2,047	2,122	2,213
79	3,487	3,273	3,317	3,368
Average	3,805	3,560	3,523	3,542
	$\Delta = 245$	$\Delta = 37$	$\Delta = 19$	
	$\%\Delta = 6.4$	$\%\Delta = 1.0$	$\%\Delta = 0.5$	
	$P < 0.001$	N.S.	N.S.	

To obtain $\int_0^{120} V(t)dt$ (Eq. 5), the activity of this coronary venous blood was corrected for the different geometry of the test tube compared to the coil by a conversion factor. This conversion factor was obtained by relating the activity of an aqueous solution of ^{84}Rb in the test tube to that in the coil.

To obtain $\int_0^{\infty} A_1(t)dt$ (Eq. 4) for the myocardial clearance of ^{84}Rb, the arterial concentrations were summed from the onset of the appearance to the beginning of recirculation. The downslope was plotted on semilog paper and the area under the curve was calculated. The numerator $U_H(t)$ of Eq. 4 is identical to that of Eq. 5.

Another conversion factor was used to relate the activity obtained from the well counter to precordial counts. The determination of this coefficient has been described previously (*3*). To compare the myocardial uptake measured *in vivo* with the uptake of the isolated and empty heart, the hearts of

TABLE 2. COMPARISON OF MYOCARDIAL BLOOD FLOW AND ^{84}Rb CLEARANCE DETERMINED BY FIRST AND SECOND BOLUS IN THE SAME ANIMAL 20 MIN APART

Experiment No.	Weight (kg)	Flow (cc/min)		Clearance (cc/min)	
		Bolus I	Bolus II	Bolus I	Bolus II
3	19.1	138.0	157.7	130.1	160.4
4	15.9	121.0	94.0	142.2	113.0
5	14.1	135.4	84.0	137.7	93.4
6	16.3	240.0	228.0	206.7	189.2
8	15.9	169.6	129.3	137.8	135.9
9	14.5	141.4	105.1	149.9	127.5
10	18.6	267.5	223.0	231.1	231.8
11	25.9	362.0	290.0	401.0	386.9
12	21.3	259.9	215.0	286.0	249.0
13	18.6	173.0	198.6	174.6	190.9
14	13.6	177.8	169.3	178.2	165.3
Average	17.6	198.7	172.2	197.8	185.8
SE		±22.5	±12.8	±24.9	±24.7
		$P < 0.1$ N.S.		$P < 0.2$ N.S.	

TABLE 3. COMPARISON OF MYOCARDIAL BLOOD FLOW AND ^{84}Rb CLEARANCE

Experiment No.	Weight (kg)	Flow (cc/min)	Clearance (cc/min)	Difference (cc/min)	Difference (%)
1	20.0	136.5	149.8	13.3	9.7
2	15.9	112.7	107.3	−5.4	−4.8
3	19.1	138.0	130.1	−7.9	−5.7
4	15.9	121.0	142.2	21.2	17.5
5	14.1	135.4	137.7	2.3	1.7
6	16.3	240.0	206.7	−33.3	−13.9
7	16.8	200.4	220.5	20.1	10.0
8	15.9	169.6	137.8	−31.8	−18.7
9	14.5	141.4	149.9	8.5	6.0
10	18.6	267.5	231.1	−36.4	−13.6
11	25.9	362.0	401.0	39.0	10.7
12	21.3	259.9	286.0	26.1	10.0
13	18.6	173.0	174.6	1.6	0.9
14	13.6	177.8	178.2	0.4	0.2
15	17.3	187.9	160.4	−27.5	−14.6
16	13.2	105.2	118.1	12.9	12.3
17	20.4	171.1	230.6	59.5	34.8
18	15.0	160.7	164.1	3.4	2.1
19	11.8	91.7	133.0	41.3	45.0
20	15.9	147.4	156.3	8.9	6.0
21	15.4	114.9	125.3	10.4	9.1
22	10.0	119.2	137.7	18.5	15.5
23	13.1	150.8	150.7	−0.1	−0.1
24	24.0	171.6	199.5	27.9	16.3
25	18.4	161.4	173.2	11.8	7.3
26	13.8	144.7	162.5	17.8	12.3
27	15.2	146.1	147.5	1.4	1.0
28	18.1	98.1	136.4	38.3	39.0
29	16.8	226.2	190.1	−36.1	−16.0
30	16.3	173.0	187.6	14.6	8.4
31	20.0	129.8	134.8	5.0	3.8
32	18.1	133.8	149.4	15.6	11.6
33	17.7	112.2	118.2	6.0	5.3
34	25.9	200.4	254.9	54.5	27.2
35	16.1	109.7	129.2	19.5	17.0
36	16.1	167.2	166.4	0.8	0.5
37	22.2	246.7	221.6	−25.1	−10.2
38	17.2	136.3	162.8	26.5	19.5
39	19.1	165.0	160.6	−4.4	−2.7
40	14.7	198.7	202.1	3.4	1.7
41	21.3	189.4	194.1	4.7	2.5
42	24.0	273.5	312.6	39.1	14.3
43	20.2	243.6	275.2	31.6	13.0
44	15.2	169.4	167.0	−2.4	−1.4
Average	17.5	170.0	179.0	9.0	6.6
SE		±8.4	±8.7		
				$P < 0.02$	

15 animals were excised and counted at the end of the experiments.

To test the reproducibility of the method, a second bolus was injected in 10 animals 15–20 min after the first injection. The stability of the relationship between clearance and flow was evaluated during infusion of vasopressor amines. In these experiments, isoproterenol (1 μg/kg/min) and norepinephrine (0.25, 0.5 and 1 μg/kg/min) were infused intravenously. The second bolus of ^{84}Rb was injected when the blood pressure had become stable. This usually occurred from 3 to 6 min after beginning infusion of each drug.

Central aortic (phasic and mean) pressure was measured through a catheter placed in the ascending aorta and recorded on a photographic strip chart (Model DR-8 Recorder, Electronics for Medicine, White Plains, N.Y.). In a few experiments cardiac output was also calculated by the same isotopic injection method (Eq. 3), substituting the activity of the injected ^{84}Rb for body uptake.

RESULTS

Myocardial uptake. Table 1 and Fig. 1 show that from 90 to 270 sec after the injection of ^{84}Rb, the myocardial uptake of ^{84}Rb remained constant. This period is used for measuring myocardial flow and clearance.

Reproducibility of results. Paired determinations of resting flow and clearance were performed 20 min apart in 10 animals to establish the reproducibility of the method. Table 2 shows that there is no statistically significant difference between dual determinations of flow ($P < 0.1$) and clearance ($P < 0.2$). This is in agreement with the data of Knoebel and McHenry (7).

Table 3 and Fig. 2 give myocardial flow and clearance in 44 dogs at rest. The average flow is 170.0 ± 8.4 cc/min, and the average clearance is 179.0 ± 8.7 cc/min. It can be seen that over a wide range of flow (91–362 cc/min/whole heart), the mean difference between flow and clearance is 9.0 cc/min (6.6%), which is significant ($P < 0.02$).

The regression line relating clearance (MCL) to flow (MBF) is seen in Fig. 3. The estimating equation is: MCL = 15.44 + 0.96 MBF. The essential features of the regression line are: (1) the relationship between flow and clearance is linear (correlation coefficient, r = 0.92 ± 0.06); (2) this linear relationship is significant at a level less than 0.1% as shown by analysis of variances (F-value = 237.5); (3) the standard error of the estimate is ±22.7; and (4) the regression coefficient is 0.96 ± 0.06 or nearly one.

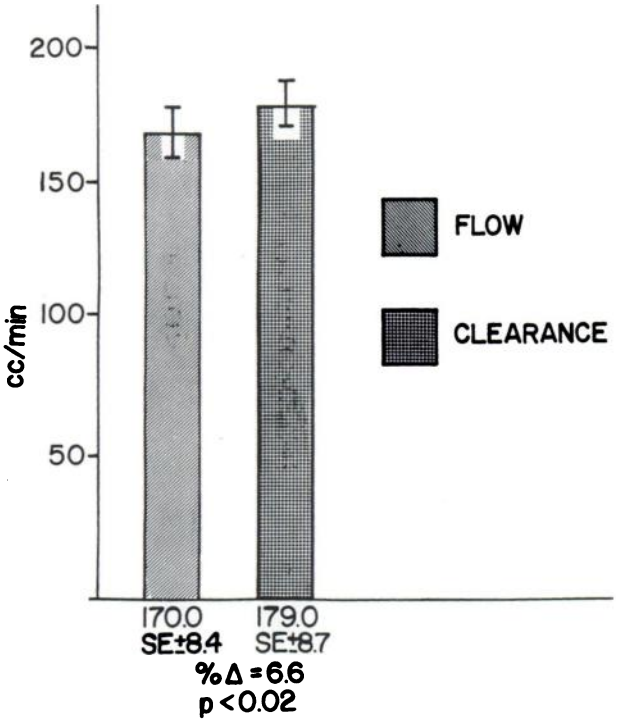

FIG. 2. Flow and clearance in 44 anesthetized dogs. Bars represent average myocardial blood flow and clearance of ^{84}Rb.

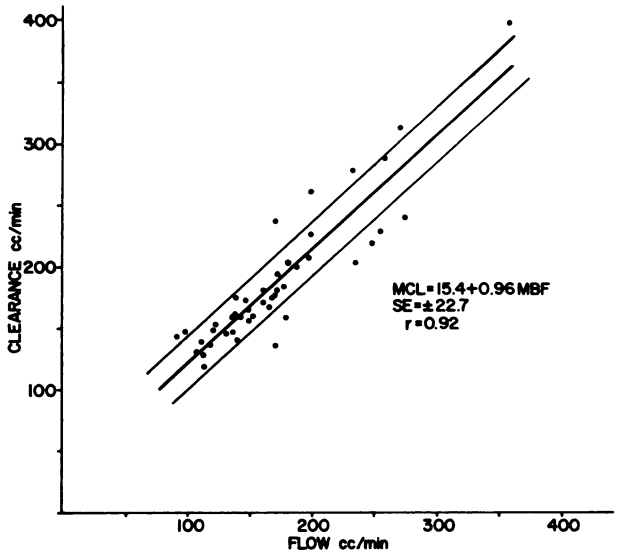

FIG. 3. Regression line relating myocardial clearance of ^{84}Rb to myocardial blood flow in 44 anesthetized dogs. Thin lines enclose standard error.

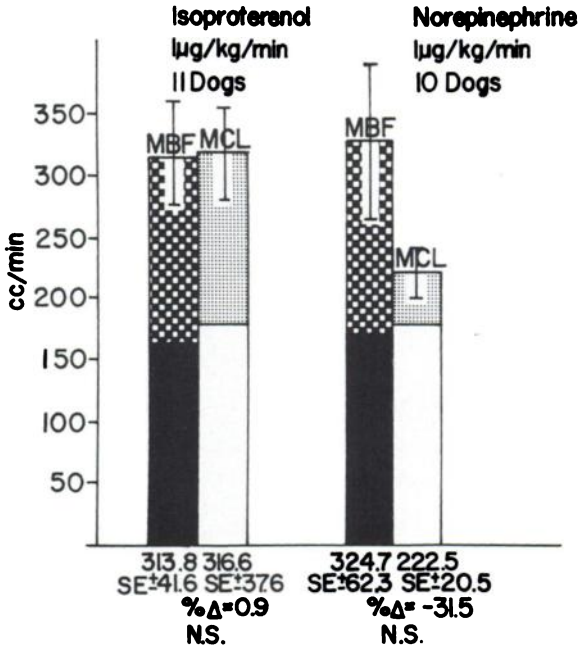

FIG. 4. Flow and clearance during isoproterenol and norepinephrine infusion. Left-hand bar of each pair is myocardial blood flow (MBF); right-hand bar is myocardial clearance of ^{84}Rb (MCL). Solid black and white lower portions are resting values and hatched upper portions are increases.

Effects of isoproterenol and norepinephrine infusions. The effect of these drugs on myocardial blood flow and clearance of ^{84}Rb are illustrated in Tables 4 and 5 and Fig. 4. It may be seen that both drugs at a dose of 1 μg/kg/min cause an increase in myocardial blood flow of about 100%. During isoproterenol infusion (Table 4), flow and clearance show a proportional increase; however, during norepinephrine infusion (Table 5), the increase in flow was considerably greater compared to clearance. Thus the mean difference between flow and clearance during isoproterenol infusion is only 3.2%, while the mean difference during norepinephrine infusion is −19.8% (Fig. 4). The regression line obtained in the normal population of 44 dogs is also used for Fig. 5. It can be seen that the results during the infusion of isoproterenol (1.0 μg/kg/min) and norepinephrine (0.25 and 0.5 μg/kg/min) are within the standard error of that regression line. In contrast, values obtained during infusion of 1.0 μg/kg/min norepinephrine lie outside and below the standard error because myocardial clearance fails to increase proportional to flow.

TABLE 4. EFFECT OF ISOPROTERENOL INFUSION ON MYOCARDIAL BLOOD FLOW

Experiment No.	Weight (kg)	Dose (μg/kg/min)	Before infusion				During infusion				Increase (%)	
			Flow (cc/min)	Clearance (cc/min)	Clearance-flow (Δ)	Clearance-flow (%Δ)	Flow (cc/min)	Clearance (cc/min)	Clearance-flow (Δ)	Clearance-flow (%Δ)	Flow	Clearance
2	15.9	1.0	112.7	107.3	−5.4	−4.8	262.3	280.8	18.5	7.1	132 7	161.7
15	17.3	1.0	187.9	160.4	−27.5	−14.6	318.2	282.5	−35.7	−11.2	69.3	76.1
16	13.2	1.0	105.2	118.1	12.9	12.3	160.5	189.2	28.7	17.9	52.6	60.2
17	20.4	1.0	171.1	230.6	59.5	34.8	469.3	446.2	−23.0	−4.9	174.2	93.5
18	15.0	1.0	160.7	164.1	3.4	2.1	414.2	420.0	5.8	1.4	157.7	155.9
19	11.8	1.0	91.7	133.0	41.3	45.0	192.2	223.0	30.8	16.0	109.5	67.6
20	15.9	1.0	147.4	156.3	8.9	6.0	228.5	214.9	−13.6	−6.0	55.0	37.4
22	10.0	1.0	119.2	137.7	18.5	15.5	150.2	163.7	13.5	9.0	26.0	18.9
42	24.0	1.0	273.5	312.6	39.1	14.3	597.6	566.2	−31.4	−5.2	118.5	81.1
43	20.2	1.0	243.6	275.2	31.6	13.0	338.3	384.7	46.4	13.7	38.9	39.8
44	15.2	1.0	169.4	167.0	−2.4	−1.4	320.2	311.9	−8.4	−2.6	89.0	86.8
Average	16.3	1.0	162.0	178.4	16.4	11.1	313.8	316.6	2.8	3.2	93.0	79.9
SE			±17.2	±19.9			±41.6	±37.6				
					N.S.				N.S.			

DISCUSSION

Experiments were performed to test the validity of a method for measuring myocardial blood flow in which an injection of a single bolus of a positron emitter, ^{84}Rb, was used. In addition, the effects of isoproterenol and norepinephrine on myocardial blood flow and clearance were studied. In this laboratory the infusion of ^{84}Rb, rather than a bolus injection, has been used for a number of years to measure myocardial clearance. The infusion technique has the disadvantage that the determination of myocardial clearance requires a prolonged steady state (7-min periods) and that the calculations are complicated and time consuming. The procedure used in this paper is based on the method of Donato (*5*), using the assumption of Sapirstein (*10*) that for a certain period of time (in dogs from 20 to 120 sec) a bolus of rubidium is distributed to the individual organs in proportion to the fraction of the cardiac output perfusing that organ.

If this is correct, then during a limited period of time following a bolus injection of the isotope, clearance and flow should be identical. Before this technique can be used, however, it is necessary to demonstrate that the activity of blood in the cardiac chambers does not interfere with the measurement

TABLE 5. EFFECT OF NOREPINEPHRINE INFUSION ON MYOCARDIAL BLOOD FLOW

Experiment No.	Weight (kg)	Dose (μg/kg/min)	Before infusion				During infusion				Increase (%)	
			Flow (cc/min)	Clearance (cc/min)	Clearance-flow (Δ)	Clearance-flow (%Δ)	Flow (cc/min)	Clearance (cc/min)	Clearance-flow (Δ)	Clearance-flow (%Δ)	Flow	Clearance
21	15.4	0.25	114.9	125.3	10.4	9.1	141.7	149.0	7.3	5.2	23.3	18.9
23	13.2	0.5	150.8	150.7	−0.1	−0.06	232.2	220.4	−11.8	−5.1	54.0	46.3
25	18.4	0.5	161.4	173.2	11.8	7.3	222.7	230.1	7.4	3.3	28.0	23.2
26	13.8	0.5	144.7	162.5	17.8	12.3	159.9	186.7	26.8	16.7	10.5	14.9
27	15.2	0.5	146.1	147.5	1.4	1.0	157.3	172.2	14.9	9.5	7.7	16.7
28	18.2	1.0	98.1	136.4	38.3	39.0	139.1	179.7	40.6	29.2	41.8	31.7
29	16.9	1.0	226.2	190.1	−36.1	−16.0	309.6	253.2	−56.4	−18.2	36.9	33.2
31	20.0	1.0	129.8	134.8	5.0	3.8	174.4	171.3	−3.1	−1.7	34.4	27.1
33	17.7	1.0	112.2	118.2	6.0	5.3	206.6	161.7	−44.9	−21.7	84.1	36.8
40	14.7	1.0	198.7	202.1	3.4	1.7	392.9	269.1	−123.8	−31.5	97.7	33.2
41	21.3	1.0	189.4	194.1	4.7	2.5	225.2	200.5	−24.7	−11.0	18.9	3.3
42	24.0	1.0	273.5	312.6	39.1	14.3	355.1	308.8	−46.3	−13.0	29.8	−1.2
43	20.2	1.0	243.6	275.2	31.6	13.0	466.3	323.1	−143.2	−30.7	91.4	17.4
44	15.2	1.0	169.4	167.0	−2.4	−1.4	796.1	226.3	−569.8	−71.6	369.9	35.5
45	19.1	1.0	56.5	60.6	4.1	7.3	181.5	131.1	−50.4	−27.7	221.2	116.3
Average	18.7	1.0	169.7	179.1	9.4	6.9	324.7	222.5	−101.9	−19.8	102.6	33.3
SE			±22.0	±23.5			±62.3	±20.5				
					N.S.				N.S.			

AND ^{84}Rb CLEARANCE

Mean aortic pressure (mmHg)			Heart rate (beat/min)		
Before	During	De-crease (%)	Before	During	In-crease (%)
76	53	—30.3	133	190	47.4
96	83	—13.5	146	206	41.1
60	43	—29.2	122	164	34.4
95	69	—27.4	163	205	25.8
99	79	—20.2	152	215	41.5
94	49	—47.9	166	230	38.6
87	74	—15.5	152	216	41.6
85	62	—27.0	130	183	40.7
115	71	—38.3	200	226	13.0
108	66	—38.5	152	140	—5.3
97	61	—37.5	170	169	—0.6
92	64	—28.9	153	195	28.9

of the myocardial uptake during the time of measurement. To demonstrate that the radioactivity of blood in the cardiac chambers using coincidence-counting detectors does not significantly alter precordial counts, a series of experiments were carried out in the isolated, nonbeating dog heart. These data have been previously published (*4*). In these tests, the activity of intracavitary blood was maintained equal to that after prolonged intravenous infusion of ^{84}Rb. The activity of blood in the cardiac chambers did not significantly interfere with the precordial counts. This is because the sensitivity of the detection system of the well counter is about 160 times that of the paired coincidence-counting detectors. Therefore, the activity of the blood can be maintained so low that it plays only an insignificant role in the total counts from the coincidence-counting detectors (*1,15*). McHenry and Knoebel also stated that the contribution of intracavitary blood to the external myocardial counts is minimal (2.3%) from 120 to 150 sec (*6*).

Another fact to be ascertained is whether myocardial uptake is constant for the period of its measurement (90 to 270 sec). During that period, myocardial uptake reflects the previously accumulated amount of isotope in the heart muscle. Table 1 and Fig. 1 show that from 90 to 270 sec after the ^{84}Rb injection, myocardial uptake remains constant. The value for myocardial ^{84}Rb uptake for the period from 30 to 90 sec is only 6.4% higher than during the following 60 sec (Table 1 and Fig. 1). Therefore, the initial uptake of ^{84}Rb in the heart muscle is almost completed 30 sec after the bolus injection. Thus a steady state of at least 30 sec, but not exceeding 90 sec, is necessary. Love and coworkers (*8,16*) using punch biopsy of heart muscle found that the relatively high isotope value of heart muscle compared to that in blood is not due to continuous gain of the isotope, but rather to "lag" in the loss of the isotope from the myocardium. Sapirstein (*9,10*), Levy and Oliveira (*17*) and McHenry and Knoebel (*6*) also demonstrated constancy of the myocardial uptake for a limited period of time.

It is likely that the myocardial uptake in animals as determined in our experiments is too high. This is illustrated in Table 6, in which the uptake of the

AND ^{84}Rb CLEARANCE

Mean aortic pressure (mmHg)			Heart rate (beat/min)		
Before	During	In-crease (%)	Before	During	In-crease (%)
109	112	2.8	106	120	13.2
60	71	18.3	156	174	11.6
85	96	12.9	79	147	86.1
64	145	126.6	151	102	—32.4
106	110	3.8	187	187	0
57	61	7.0	142	165	16.2
49	57	16.3	181	178	—1.7
86	211	144.0	174	132	—24.1
105	159	51.5	151	74	—51.0
96	115	19.8	146	153	4.8
115	173	50.4	200	82	—59.0
108	173	61.0	152	115	—24.3
97	161	65.3	170	82	—51.7
39	100	156.4	110	103	—6.4
84	134	63.5	159	120	—21.9

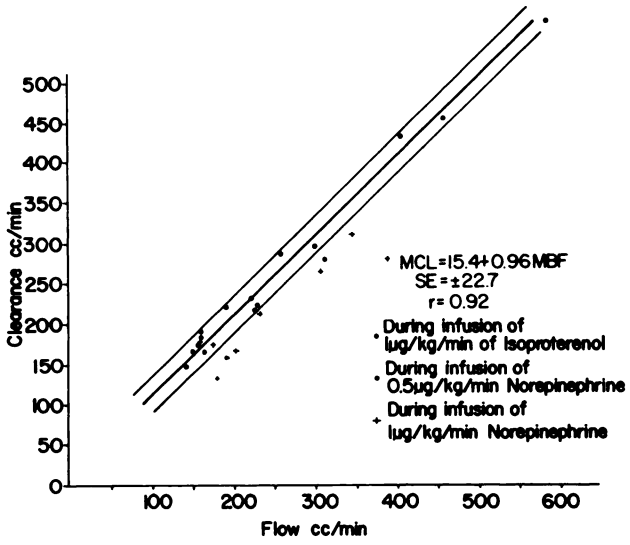

FIG. 5. Effect of isoproterenol and norepinephrine on relationship between clearance and flow. Values of myocardial clearance of ^{84}Rb and myocardial blood flow are plotted with same regression line as in Fig. 3.

TABLE 6. COMPARISON BETWEEN PRECORDIAL ACTIVITY (HEART IN VIVO) AND ACTIVITY OF EXCISED AND DRAINED HEART (IN VITRO)

Experiment No.	In vivo (cpm)	In vitro (cpm)
35	6,447	4,546
36	7,860	7,831
37	9,478	7,234
38	10,345	10,447
39	13,957	11,895
40	9,069	3,916
41	10,287	5,119
42	12,693	5,969
43	12,969	5,967
44	17,735	15,468
45	8,287	4,121
46	9,524	5,928
47	13,202	10,899
48	8,500	7,239
49	8,449	6,919
Average	10,587	7,567
Difference (%)	28.5	
	$P < 0.001$	

heart *in vivo* is compared to that of the same heart removed from the body. The uptake of the isolated heart with blood-free chambers was 28.5% lower than that of the heart *in vivo*. This discrepancy is due in part to adjacent thoracic activity counted by the large 4-in. detectors which were designed for the heart size of adult man. In addition, the pair of 2-in. detectors, devised for subtraction of thorax "background" could not be used in the dog because of the anatomical arrangement of the canine thorax. Consequently, the values for myocardial uptake in the dog as obtained in this series are consistently too high. This, however, does not defeat the main purpose of these experiments in which clearance is compared to flow. Since the same value for uptake is used for both clearance and flow, comparison between these two determinations remains justified.

Another fact which needs verification is the reproducibility of the results under identical experimental conditions. Table 2 shows that there is no statistically significant difference between paired studies carried out 20 min apart. Knoebel and McHenry (7) reported similar results.

The comparison between myocardial clearance and flow in 44 animals shows that over a wide range of flow (91–362 cc/min/whole heart) the average difference is only 6.6%; this, however, is statistically significant. The regression line relating clearance to flow is linear, and the regression coefficient is nearly one. Consequently, clearance is flow dependent over a wide range, but clearance is consistently higher by a small, but significant, quantity. This disparity may be caused either by a comparative reduction of the amount of rubidium in the brain as postulated by Sapirstein (*10*) or by uptake of the isotope in the lungs as demonstrated by Clarke and Rushmer (*18*) and Sheppard and coworkers (*19*).

One may object to the validity of the conclusion drawn from a comparison of clearance to flow since the same isotope and the same detection system were used. In general, the direct Fick principle, which is used to measure total myocardial blood flow in our experiments, is a valid yardstick. Clearance represents nutritional flow. Since the principle underlying the measurement of myocardial clearance of rubidium is different from that entailed in the measurement of flow, comparisons are valid; as a matter of fact, the use of an identical isotope and detection system is a definite advantage since the experimental errors cancel each other out. In the nitrous oxide method, a steady state of at least 4 min is essential and the coronary sinus must be incubated (*20*). Other procedures are dependent upon direct injections in the left ventricle (*21,22*) or in the coronary artery (*23,24*). There is at the present time no simple method which offers the direct and quantitative measurement of all values necessary for the calculation of myocardial flow in man by the Fick principle (Eq. 5). In animals electromagnetic flow meters do not permit the measurement of total myocardial blood flow since it is extremely difficult to place a probe around the left common coronary artery.

The finding that clearance is primarily flow dependent is substantiated by the results obtained during the infusion of isoproterenol. During infusion of that drug, a proportional increase of clearance and flow is observed. However, this is not the case during the infusion of a large dose of norepinephrine (1.0 μg/kg/min). Here the increase in clearance lags behind that in flow with a mean difference of almost 20%. This is in line with the observations of many workers that the extraction ratio diminishes as flow increases (*4,25–30*). Under these circumstances, clearance is not flow dependent only since the percentage of myocardial extraction diminishes with increasing flow, impairing the basic principle of the method. The relation of myocardial extraction of rubidium to total-body extraction, and the relation of myocardial uptake of rubidium to myocardial blood flow is altered as the extraction ratio changes. The method employing constant infusion of the isotope, previously used in this laboratory, does not have this disadvantage because the calculation of flow is obtained by extrapolation of the myocardial clearance to time zero. At that time, the extraction ratio is theoretically one, provided the myocardium is extracting all of the isotope at this time (*4*).

Although the data show that clearance is primarily flow dependent, it is not claimed here that flow is the only factor influencing clearance. It has been determined that velocity of flow (*31*), relationship of permeability to surface area of capillaries (*32–34*) and intramyocardial shunts (*29*) also may influence the myocardial extraction ratio of rubidium and consequently its clearance. But the data show that under the experimental condition present in this report these factors are of no major importance in influencing the clearance-to-flow dependency.

SUMMARY

The myocardial blood flow in the anesthetized dog using the coincidence-counting system in connection with a bolus injection of the isotope ^{84}Rb was investigated. The validity of the method was tested by comparing myocardial blood flow with myocardial clearance of ^{84}Rb; flow was calculated with the Fick principle and clearance was obtained by dividing precordial uptake of the isotope by the integrated arterial concentration of the first circulation.

The average coronary blood flow in 44 dogs was 170.0 cc/min/whole heart; the average myocardial clearance was 179.0 cc/min/whole heart. The difference was 6.6% and statistically significant ($P < 0.02$). It could be demonstrated, however, that over a wide range of flows (91–362 cc/min/whole heart) there is a linear and direct relationship between myocardial blood flow and clearance in the resting dog.

The effects of isoproterenol and norepinephrine on both myocardial blood flow and clearance of ^{84}Rb were examined. Isoproterenol did not change the relationship between clearance and flow, but a high dose of norepinephrine caused a proportional greater increase in flow than in clearance. It is likely that this is due to an alteration of the myocardial extraction ratio of rubidium. The results demonstrate that myocardial clearance of ^{84}Rb is primarily flow dependent under these experimental conditions and that the factors which can alter clearance were not a major influence.

The advantages and disadvantages of the bolus method as compared to the constant infusion method were discussed.

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