the tyrosine transaminase to activity of the homogentisate oxidase in the liver. This type of alkaptonuria is therefore the result of an adaptive increase in relative rate of homogentisate formation, in contrast to the relative decrease in rate of homogentisate degradation believed to be responsible for hereditary alkaptonuria in man.

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Role of Potassium in Regulation of Coronary Blood Flow.* (23521)

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Katz and Lindner(1) have shown that small increments in the concentration of potassium (K) in defibrinated blood perfusing the fibrillating dog heart result in coronary vasodilation, whereas large increments of K produce vasoconstriction. Dawes(2) observed a similar relationship between blood K concentration and changes in arterial flow in the perfused hind limbs of cats and dogs. Since small increases in K levels decrease vascular resistance and since K is released from actively contracting muscle(3,4), Dawes(2) suggested that the K ion originating in the muscle may be responsible for the increased flow which accompanies muscular contraction. The present study was undertaken to determine to what extent the coronary vasodilation, which occurs during increased metabolic activity of the myocardium, could be accounted for by the release of K from muscle cells.

Methods. Ten dogs weighing 14.6 to 20.7 kg were used in these experiments. The animals were anesthetized with sodium pentobarbital (30 mg/kg) and the chest was opened in the fourth left intercostal space. Lung inflation was maintained by positive pressure artificial respiration and heparin (180 mg) was given to prevent blood clotting. The coronary sinus was cannulated via the right atrium and the venous outflow directed through rubber tubing into the right external jugular vein. The left coronary artery was cannulated with an Eckstein cannula(5) and received blood from the right common carotid artery via a pump perfusion system which maintained a constant perfusion pressure independent of aortic pressure. Mean coronary blood flow (CBF) was measured by an optically recording rotameter and mean perfusion pressure (PP) and phasic aortic pressure (AP) were recorded by modified Gregg manometers. In the first set of experiments several concentrations of KCl were infused proximal to the rotameter (to insure complete mixing of blood and infusate) at a constant rate of 0.93 cc per minute. Concentrations of KCl were used which were calculated to increase the animal's serum K level by 1.5, 3.0 and 6.0 meq/l. Recordings of CBF, AP, and PP were taken 2 min-

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TABLE I. Effect of Intracoronary Administration of Potassium on Coronary Blood Flow.

Exp. No.	Infused K^+, meq/min.	Coronary artery K^+, meq/l		Coronary sinus K^+, meq/l		Coronary blood flow, cc/100 g perfused heart/min.		% change CBF*
		Control	Exp.	Control	Exp.	Control	Exp.	
1	.13	2.56	4.23			100.5	114.1	+13.5
4	.13	3.28	4.37	3.29	3.89	72.0	71.8	− .3
6	.13	3.05	4.39	2.87	3.70	54.8	54.6	− .4
8	.13	2.50	5.34			102.3	110.4	+ 7.9
7	.13	3.39	5.40	3.26	4.80	41.6	58.0	+39.4
6	.26		5.80		4.80	49.4	59.9	+21.3
8	.26	3.35	6.20			106.3	116.5	+ 9.6
1	.26		7.48			94.0	100.7	+ 7.1
7	.26		7.50		6.10	62.8	59.9	− 4.6
2	.26	3.72	8.33			59.0	71.1	+20.5
1	.52		8.96			86.7	104.4	+20.4
10	.52	3.60	9.10	3.55	8.70	128.5	152.7	+18.8
6	.52		9.50		9.10	50.1	67.9	+35.5
8	.52	3.00	10.00			101.7	114.3	+12.4
4	.39		10.10		9.67	67.7	74.2	+ 9.6
2	.52		12.10			56.8	71.9	+26.6

* Plus (+) indicates an increase and minus (−) a decrease.

utes after the start of the infusion. At this time flows were stabilized at a new level. Arterial blood samples were drawn from the tubing just proximal to the inflow cannula, and coronary sinus blood samples were taken simultaneously from the tubing distal to the coronary sinus cannula. K concentrations were determined by flame photometry. A second recording of CBF, AP, and PP was taken immediately after the blood samples were collected. Measurements reported here represent the average of the records taken before and after the blood collection. The perfusion with KCl was then stopped and the CBF allowed to return to the pre-perfusion level. Control records of CBF, AP and PP and control samples of coronary arterial and venous blood were taken prior to each infusion of K. Control infusion of 0.9% NaCl at a rate of 0.93 cc/min. was without significant effect on CBF.

In the second set of experiments, CBF was increased by methods which augment myocardial metabolism and by asphyxia. The rate of cardiac metabolism was elevated by 1) intracoronary administration of 2,4-dinitrophenol (1.3-2.7 mg/min.) and epinephrine (9.3-18.6 μg/min.) and 2) constriction of the aorta distal to the left subclavian artery (to increase cardiac work). Asphyxia was induced by temporarily stopping the respirator. Recording of CBF, PP, AP and blood sampling were conducted as described above. After each experiment the perfused portion of the heart was stained, excised and weighed, and CBF has been expressed as flow per 100 g of perfused heart muscle.

Results. In the first group of experiments (Table I) a constant infusion of KCl produced a measurable increase in CBF in 13 of 16 experiments, with an average increase of 17.7%. The rise in flow was not related to the concentration of K added to the perfusate, nor to the final plasma K concentration. No vasoconstriction was observed even when plasma K levels reached 12.1 meq/l. The infusions of KCl produced no significant alteration in heart rate or arterial pressure.

In the second group of experiments, infusion of 2,4-dinitrophenol or epinephrine, production of asphyxia, or increased arterial resistance resulted in moderate to marked increases in CBF (Table II). The average increase in CBF with asphyxia was 60.5%, with epinephrine 63.6%, and with DNP 35.4%. The results with acute hypertension were variable: a 14% increase occurred in one animal and no significant change occurred in two other dogs. Although flow was significantly increased by several different procedures, no significant changes in coronary sinus plasma K concentrations or in K arteriovenous differences were observed.

Discussion. Intracoronary administration

TABLE II. Effect of Increasing Coronary Blood Flow on Coronary Sinus Blood Potassium Levels.

Exp. No.	Method of increasing CBF	Coronary artery K^+ conc., meq/l		Coronary sinus K^+ conc., meq/l		C.B.F., cc/100 g perfused heart/min.		% change CBF	Change K^+ A-V differences, meq/l
		Control	Exp.	Control	Exp.	Control	Exp.		
4	Asphyxia	3.66	3.25	4.11	3.87	67.2	130.2	+ 93.8	+.17
3	"	3.33	3.46	3.26	3.13	97.4	112.9	+ 15.9	+.26
6	"	3.50	3.48	4.10	4.00	92.2	125.4	+ 36.0	+.08
11	"	2.57	4.47	2.48	3.73	84.5	116.0	+ 96.4	+.83
11	Acute hypertension	2.45	2.58	2.46	2.61	80.5	77.9	- 3.2	+.02
11	*Idem*	2.81	2.89	2.69	2.81	74.0	75.5	+ 2.0	-.04
4	"	3.79	3.61	3.83	3.79	60.1	72.4	+ 20.4	+.14
	Dinitrophenol								
11	(1.3 mg/min.)	2.56	2.46	2.66	2.58	79.7	100.6	+ 26.2	-.12
11	(2.7 ")	3.00	2.52	2.78	2.61	67.2	108.5	+ 61.5	-.31
7	(1.5 ")	3.09	3.09	2.91	3.28	63.5	67.2	+ 5.8	+.35
7	(3.7 ")		3.10		3.16	63.5	94.5	+ 48.8	
10	(2.7 ")	3.55	3.20	3.50	3.15	137.5	185.4	+ 34.8	.00
	Epinephrine								
9	(9.3 μg/min.)	2.60	2.50	2.60	2.60	68.8	110.6	+ 60.6	-.10
11	(18.6 ")	2.84	2.69	2.73	2.20	63.2	130.1	+104.4	+.38
11	(9.3 ")	2.62	2.79	2.50	2.21	79.0	115.3	+ 45.9	+.46
10	(18.6 ")	2.50	2.85	3.50	2.45	109.5	156.5	+ 43.4	+.14

Plus (+) indicates an increase and minus (-) a decrease.

of KCl at rates which elevated plasma K from 1 to 8 meq/l produced small to moderate increases in CBF. Although these results confirm those of Katz and Lindner(1) and of Dawes(2), it is difficult to make quantitative comparisons of their data with ours because they did not measure plasma K concentrations and made almost all of their observations on blood flow following single rapid injections of KCl. With high concentrations of K these investigators(1,2) observed vasoconstriction. The concentration of K that must be reached in the plasma to induce vasoconstriction cannot be accurately determined from their data, but it must be in excess of 12 meq/l, since we observed only vasodilation at this, or lower, concentrations.

Although the heart beating *in situ* is more physiological than a fibrillating heart preparation, flow may conceivably be altered by changes in extravascular compression which would accompany changes in heart rate or force of contraction. However, it is unlikely that extravascular compression was significantly altered in the present study since heart rate and aortic pressure were not affected by the KCl infusion.

Procedures which increase CBF by increasing the oxygen requirements of the heart or by decreasing the oxygen supply to the myocardium were not associated with K release from the myocardium. In fact, with one exception (Exp. 11—asphyxia), negligible changes in K A-V differences occurred and most of these were in a positive rather than negative direction. In the few instances in which the potassium A-V difference decreased, the change was due to a fall in arterial rather than a rise in coronary sinus K concentration. It is possible that the present methods were not sensitive enough to detect the release of K from the heart. If this is true then the concentration of K reached in the vicinity of the arterioles is too small to produce changes in arterial resistance since concentrations as high as 12 meq/l failed to produce changes in blood flow of the magnitude seen with increases in myocardial metabolism or asphyxia.

These observations do not support the hypothesis of Dawes(2) that K release from muscle is responsible for the reduction in vascular resistance associated with increased muscular activity. The nature of the metabolite(s) which operate to adjust blood flow to metabolic requirements remains obscure.

Summary. Intracoronary infusion of KCl sufficient to elevate coronary arterial plasma K concentration to levels of from 4.23 to 12.10 meq/l produced increases in CBF averaging

17.7%. The changes in CBF produced by KCl infusion did not parallel the changes in plasma K concentration. Infusion of 2,4-dinitrophenol or epinephrine, asphyxia, or increased aortic pressure (all factors which are known to increase myocardial oxygen consumption and coronary blood flow) did not result in the release of K from the myocardium. Therefore, it appears unlikely that K release from active myocardium is responsible for adjustment in coronary resistance which accompanies changes in metabolic activity of the myocardium.

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Reactivity of Sulfhydryl and Disulfide Groups in Serum of Man.* (23522)

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Sulfhydryl-containing compounds such as cysteine and cysteinamine have been shown to be very effective in protecting against the lethal effects of ionizing radiations(1-3). As one approach to understanding the mechanism underlying this protection we have measured in human serum the levels of sulfhydryl (–SH) and disulfide (–S–S–) and their reaction to ultraviolet (UV) light, detergent, and electrolysis.

Materials and methods. The serum used in these studies was freshly prepared from the blood of young (20-25 years) and older (40-50 years) human subjects and from various types (metastatic carcinoma of the throat and rectum) of cancer patients from the Ghent University Clinic. The sulfhydryl and disulfide determinations were accomplished using one of the amperometric methods described by Kolthoff *et al.*(4). The –SH determination was carried out in an ammonia buffer (0.1 M NH_4OH, 0.3 M NH_4NO_3); after deaeration of the buffer solution with purified nitrogen(5) the serum was added and titrated with a 0.002 or 0.001 M solution of $AgNO_3$ at –0.30 volt, *vs.* a standard calomel electrode. The total volume was 20 ml. The –S–S– determination was carried out in ammoniacal medium in the presence of sulfite (0.1 M NH_4OH, 0.1 M NH_4NO_3, 0.2 M Na_2SO_3). The $AgNO_3$ solution and the potential were identical to that used in the –SH determination. The nitrogen content of the serum was determined by an amperometric method which makes it possible to analyze very minute quantities of serum (down to 0.02 ml) in 15 minutes(6). UV irradiation. A UV lamp (type 16200, Hanovia Chemical and Manufacturing Co., Newark, New Jersey) was used to irradiate a quartz cell with a diameter of 1.27 cm and a length of 8.40 cm. The distance from the cell to the lamp was 6 cm. An aliquot of serum, diluted 1:16 with air-free phosphate buffer (0.1 M Na_2HPO_4, 0.01 M NaH_2PO_4; pH 7.29), was pipetted into the cell, which was closed hermetically after purified nitrogen had been passed over the sample(4). A current of cold air was blown over the cell in order to avoid a rise of temperature during the experiment; after 2 hours of irradiation the temperature rose 1°C. Aluminum foil was used as a reflector behind the cell. Anionic Detergent. To test the effect of an anionic detergent upon –SH and –S–S– groups, 0.5 ml of serum was placed in 20 ml

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