Reactions of the Isolated Surviving Coronary Artery to Epinephrine, Acetylcholine and Histamine.* (17709)

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During the course of an investigation into the physiologic behavior of the vascular system in rheumatic fever and other disease states, it became necessary to investigate the responses of normal vessels exposed to certain vasoconstrictor and vasodilator drugs. The effects of epinephrine, acetylcholine and histamine were studied in surviving coronary arteries from man, pig, ox and sheep, employing an angioplethysmokymographic technique. The results are presented in this paper.

Experimental procedure. The angioplethysmokymographic technique was developed in collaboration with Doctor Jerome T. Syverton(1). In this procedure an isolated surviving blood vessel is perfused and a continuous record is made of the changes in the volume of the blood vessel. A segment of artery (approximately 2 cm in length) is dissected out and mounted with minimum trauma in a plethysmographic chamber as shown in Fig. 1. The plethysmograph is filled with Tyrode's solution and the stopcock is turned so that any variation in the volume of the vessel causes a change in the position of the meniscus of the fluid in the horizontal pipette. The artery is perfused with Tyrode's solution using a perfusion system as illustrated in Fig. 1. The inflow pressure is maintained at 100 cm of water and the outflow pressure at 30 cm of water. The needle at the outlet limits the flow through the artery to 40 cc per minute. Test doses of drugs are injected directly into the perfusate through the modified T-tube in the perfusion system. These test doses flow through the artery in 15 to 20 seconds. The perfusate is oxygenated with 5% carbon dioxide in oxygen, thus providing oxygen for the artery and maintaining the reaction of the perfusate at pH 7.2 to pH 7.4. The perfusion system and plethysmographic chamber are suspended in a constant temperature bath (37.5°C±0.01°C). A photographic record of the volume changes is made at 15 second intervals on a drum camera over a period of several hours. Vasoconstriction is measured by diminution in the fluid volume, and vasodilatation by its increment. Of the two, the former is the more readily obtained measurement; whereas the latter must be dependent upon the initial state of vasomotor activity exhibited by the arterial segment. Dale and Richards(2) have pointed out that it is impossible to demonstrate vasodilatation in vessels already dilated. It follows that in

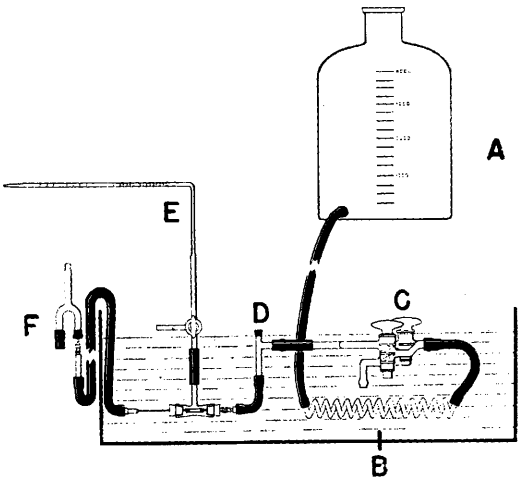

FIG. 1.
Perfusion System. A. Perfusion bottle. B. Warming coil. C. Manifold. D. Modified T-tube for injecting test doses into perfusate. E. Plethysmograph consisting of a 0.2 ml pipette connected to the specimen chamber by a 3-way stopcock. F. Outlet line. B, C, D, E (specimen chamber), and part of F are mounted in a constant temperature bath (37.5°C ± 0.01°C).

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order to determine vasodilatation, it is necessary that a vessel wall exert some degree of pressure upon the contents of the vessel. There has been some discrepancy between vessels in this respect, particularly in that the human vessels used appeared to possess or to retain this capacity to a lesser degree than did vessels obtained from experimental animals. This may be due to an inherent difference between the coronary arteries of man and those of other species, but may equally well reflect the fact that the human vessels were obtained from subjects who had died of disease while those of the pig, sheep and ox were obtained from slaughtered healthy animals. It will not, in consequence, be possible to draw too close a parallel between the behavior of human coronary arteries and those of experimental animals in respect to vasodilatation. Such differences between the behavior of arteries are for the present considered to be related more to the degree of viability of the individual vessel (or alternatively to the metabolic changes associated with vascular death) than to peculiar differences in their mode of reaction to the same stimulus.

After the artery is mounted in the plethysmographic chamber perfusion is carried out until the artery reaches a constant volume before the test drugs are injected. This usually requires about 2 hours. The baseline obtained is fairly constant for a given artery for a further period of several hours. The volume level assumed by the artery depends in part upon the pressure exerted intrinsically by the arterial wall upon its fluid contents. A satisfactory demonstration of the capacity to dilate of a given specimen was to test the artery with a known vasodilator drug, such as theophylline ethylene-diamine.

Results. 1. *Effect of epinephrine.* It has been shown previously(3) that epinephrine causes constriction of most of the peripheral arteries and probably causes dilatation of the coronary arteries(3,4,5,6). The effect of l-epinephrine (synthetic) was tested in 32 coronary arteries from different species in 54 experiments. The results in tabular form are recorded in Table I.

TABLE I.
Reaction of Isolated Coronary Arteries to Epinephrine.

Species	Man	Pig	Ox	Sheep	Totals
Vasoconstriction	18	17	0	1	36
Vasodilatation	1	11	6	0	18
Total	19	28	6	1	54

TABLE II.
Reaction of Isolated Coronary Arteries to Acetylcholine.

Species	Man	Pig	Ox	Sheep	Totals
Vasoconstriction	17	8	0	1	26
Vasodilatation	0	0	0	0	0
Total	17	8	0	1	26

TABLE III.
Reaction of Isolated Coronary Arteries to Histamine.

Species	Man	Pig	Ox	Sheep	Totals
Vasoconstriction	27	40	8	1	76
Vasodilatation	0	0	0	0	0
Total	27	40	8	1	76

2. *Effect of acetylcholine.* Acetylcholine is a dilator of the peripheral arteries(3) but its effect on the coronary arteries has not been precisely demonstrated by previous experiments. Acetylcholine was used 26 times on 18 coronary arteries, with the results shown in Table II.

3. *Effect of histamine.* Best and McHenry (7) have reviewed the data on histamine including the action of histamine upon the coronary arteries. They quote several investigators, using isolated rings and strips of coronary arteries, as having found constriction to be caused by histamine in man, dog and ox. Anrep(5), however, found that histamine produced coronary vasodilatation in perfusion experiments upon the human heart.

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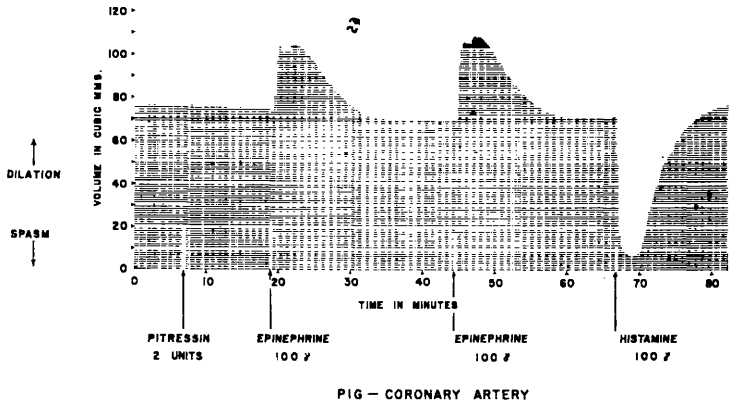

FIG. 2.
Record of the reactions of the coronary artery of a slaughtered healthy pig.

Essex *et al.*(6) demonstrated increased coronary flow following histamine injection in the unanaesthetized dog.

In the present series of experiments 76 observations have been made on 32 coronary arteries. The results are tabulated in Table III.

4. *Sample experiments.* The following are the individual records of 2 typical experiments. The dosages of the drugs employed are noted on Fig. 2 and 3 and throughout the series of experiments varied from 0.1 γ to 100.0 γ. These dosages give concentrations of between 1 part per 100,000 to 1 part per 100,000,000 under the experimental conditions employed.

Exp. 522: Fig. 2. This experiment records the reactions of the coronary artery of a healthy pig. The specimen was mounted 2 hours after the animal had been slaughtered. Pitressin caused no reaction. Epinephrine caused vasodilatation on 2 occasions. Histamine produced vasoconstriction.

Exp. 588: Fig. 3. This coronary artery† was derived from a 23-year-old white female who had succumbed to Hodgkin's Disease. It was removed at autopsy and mounted in the plethysmograph 2 hours postmortem. Again, in these experiments histamine and acetylcholine both produced vasoconstriction. Epinephrine in similar doses produced slight

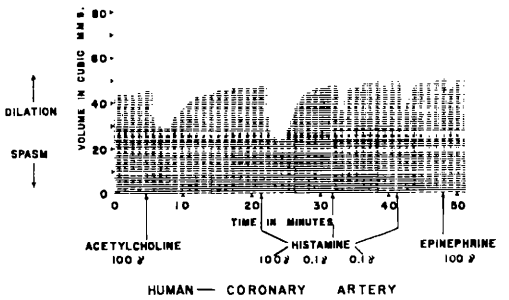

FIG. 3.
Record of the reactions of the coronary artery of a 23-year-old white female who died of Hodgkin's disease.

vasoconstriction, much smaller than that produced by histamine and acetylcholine.

Discussion. The responses of the coronary arteries to epinephrine were of two types and appeared to depend on the degree of vasomotor activity exhibited by the segment under test. Those arteries which were nearly completely dilated (as determined by their failure to respond to known vasodilators) responded to epinephrine by a slight, transient vasoconstriction as in Fig. 3. Those arteries which were not as completely dilated always responded to epinephrine by dilation as in Fig. 2. Certain of the pig arteries were completely dilated at the beginning of the experiment and would show only constriction to epinephrine; later the vessel wall would assume a certain degree of tone and under these conditions epinephrine produced only dilation of the vessel. In these specimens in which both constriction and dilation were recorded the

† All human tissues studied were supplied through the Department of Pathology, Rochester School of Medicine and Dentistry. Their cooperation in this study is hereby acknowledged.

constriction recorded in the fully dilated specimen seemed to correspond closely in size, shape and timing to the notching of the curve of dilation as in Fig. 2. None of the human coronary arteries ever assumed any significant degree of tone. Variations in dosages of test drugs employed varied the magnitude and duration of the reaction produced; it never reversed the response to a given drug (as from constriction to dilation or vice versa). Acetylcholine in varying doses uniformly produced constriction of the coronary arteries examined. Histamine likewise produced only vasoconstriction of the coronary arteries examined; this effect was uniform regardless of variation in the dosage employed (Fig. 3). It is felt that this technic will prove of value in assessing the effect of drugs and hormones upon the coronary and other arteries and that different responses may be obtained in vessels from subjects suffering from different pathological conditions.

Summary. 1. The method of employing angioplethysmokymography to evaluate the responses of coronary arteries to several drugs is described.

2. Epinephrine did not produce a uniform response in the coronary arteries examined. The relation of this inconsistency to the state of vasodilation of the specimen artery is discussed.

3. Acetylcholine was found to produce consistent coronary vasoconstriction in man, pig and sheep.

4. Histamine uniformly produces vasconstriction of the coronary arteries of man, pig, ox and sheep.

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Release of Histamine During Hemolytic Reactions in the Blood of Rabbits. (17710)

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It has been shown in different species that histamine is released from the cells of one or several organs during anaphylactic reactions(1-6). Special circumstances pertain in the blood. Most of the histamine contained in blood is held in the white cell layer which separates between the plasma and the red cells when unclotted blood is centrifuged(7,8). This bound or fixed histamine is released by comparatively mild trauma such as clotting or agitation. During anaphylactic reactions in drawn blood, Katz has found that histamine is released from the cells into the plasma(9). In Katz's experiments, rabbits were sensitized by several intravenous injections of egg white. Later when the antigen was added *in vitro* to heparinized samples of the blood the histamine content of the plasma increased 100 to 600% above that of plasma of control samples of the same blood to which no antigen had been added. Studies by Dragstedt and his co-workers(10,11) and Rose and Browne(12) have confirmed and

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