Failure of Pregnancy and Estrogen to Improve Collateral Circulation in Nonischemic Hearts of Dogs.* (29811)

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A number of natural and synthetic estrogens have been shown to decrease serum cholesterol (1). It is also known that there is a low incidence of coronary disease in premenopausal women, who usually have lower serum cholesterol than do postmenopausal women. These facts coupled with the generally accepted relationship between elevated serum cholesterol and arteriosclerosis has prompted the use of estrogen in patients who survive myocardial infarction in an attempt to prevent further attacks. Evidence, which may be open to question(2), suggests that conjugated equine estrogens are effective(3,4); ethinyl estradiol is not(5). The fact that administration of Premarin® does not universally decrease serum cholesterol(4) suggests that other factors may operate to diminish the incidence of myocardial infarction in these patients.

The finding in early postpartum mice and in those treated with ethinyl estradiol alone or in combination with progesterone(6,7) of an increase in the diameter of the pericardiacomediastinal arteries which anastomose with the coronary vessels, taken together with the vascular changes observed during pregnancy, suggest that an improvement of the coronary collateral circulation might be induced by administration of estrogen. This report describes experiments designed to test this hypothesis in dogs.

Methods. Mongrel dogs weighing 15 to 17.5 kg were given daily I.V. injections of a conjugated equine estrogen, Premarin®,‡ in doses of 8 and 16 mg/dog for the first and second weeks respectively, and 20 mg daily thereafter for periods ranging from 6 to 21 weeks. The animals were then anesthetized with 30 mg of morphine sulfate/dog and about 20 mg/kg of pentobarbital. Positive pressure respiration was given through an intratracheal tube. The left chest was opened in the fifth intercostal space. The left external jugular vein, left common carotid artery and circumflex coronary artery were isolated. Heparin (100 to 150 mg) was given intravenously.

The left common carotid artery was cannulated to supply blood to the cannulated circumflex artery. Retrograde circumflex blood flow and mean peripheral circumflex pressure were measured while mean aortic pressure was maintained at 100 mm Hg by adjusting an aortic clamp and bleeding or reinfusing blood *via* the cannulated left external jugular vein as previously described(8).

After these measurements electrocardiograms were recorded from lead AVR before and during occlusion of circumflex arterial inflow. The changes were compared with preocclusion records and graded from + to +++. Flattening or T-wave inversion without S-T depression was assigned a value of +; S-T segment depressions of less than 2 mm were graded ++; S-T depressions greater than 2 mm were rated +++. After maximum changes had occurred, the animals were killed by bleeding.

Dilute India ink was injected into the circumflex artery after it was flushed with normal saline. The remainder of the common left coronary and right coronary arteries was not perfused. The circumflex was again briefly flushed with saline after which the heart was removed to a warm saline bath, injected with barium gelatin mixture at 45°C, chilled and opened by the Schlesinger technique(9). Roentgenograms were made and examined for evidence of presence of barium in the surrounding vessels. The India ink stained areas were isolated and weighed. On dogs 9 and 10 only the data from the injection studies were obtained because of their deaths after 20 and 21 weeks of treatment respectively.

The results of the measurements of collateral function were compared with dogs se-

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TABLE I. Statistical Comparison Between Control and Experimental Dogs.

Dogs		Original dog wt (kg)	Final dog wt (kg)	Heart wt (g)	Wt of circumflex area (g)	MPCP* (mm Hg)	Retrograde flow (cc/min)
Control	No.	17		17	17	17	17
	Range	12.3–19.9		90.5–209	31.5–66.5	7–21	.4–7.4
	Avg	16.0		143.0	49.8	14.1	3.0
	S.D.	2.4		30.0	8.7	4.2	1.9
Experimental—treated with Premarin(R)	No.	10	9	10	10	8	8
	Range	15.0–17.5	14.1–18.9	108.5–167.5	40 –65.5	11–21	1.5–7.6
	Avg	16.3	16.6	137.9	51.5	15.3	4.7
	S.D.	.8	1.5	18.6	9.8	3.4	2.6
Experimental—early postpartum	No.	2		2	2	2	2
	Range	11.4; 11.4		61.4; 87	27.7; 28	10–11	2.6–3.2
	Avg	11.4		77.2	27.9	10.5	2.9
	S.D.						

* Mean peripheral coronary pressure.

lected on the basis of similarity of body weight from the previous experiments of Eckstein and Leighninger(10).

Two early post partum dogs were also studied in the manner described above.

Results. The data from these experiments are presented in Tables I and II and compared with the control animals. Peripheral circumflex pressures and retrograde flows are not significantly different from the controls. In both cases $P > .05$. Likewise, the ECG changes following circumflex occlusion are similar in the two groups. Comparisons of roentgenograms following barium gelatin injection showed essentially similar degrees of collateral development in the two groups.

A plot (not shown) of retrograde flow *vs* duration of treatment indicates that these two variables are unrelated.

Discussion. These studies indicate that neither pregnancy nor treatment of dogs with large doses of estrogen promotes the development of collateral vessels as assessed by measurements of retrograde blood flow, peripheral coronary pressure, ECG changes subsequent to arterial occlusion and the evaluation of roentgenograms following intracoronary injections of barium gelatin. The considerable mammary enlargement in the treated females and the slight enlargement in the males indicate that the dose of estrogen was sufficient to produce physiological effects.

TABLE II. Results of Electrocardiograms and Roentgenograms.

Dogs	ECG changes following circumflex occlusion	Collaterals by Ba gelatin injection
Control	1+	1 slight
	1++	12 none
	10+++	
Total	12	13
Experimental—Treated with Premarin(R)	1++	1 slight
	7+++	9 none
Total	8	10
Experimental—Early postpartum	1+++	1 none
Total	1	1

The size of the pericardiophrenic and mediastinal arteries (homologues of the pericardiaco-mediastinal artery in mice) was not determined in these experiments; however, the marked ECG changes seen following circumflex arterial occlusion indicate the lack of significant development of extracoronary anastomoses. The enlargement of the pericardiaco-mediastinal arteries found by Zweens(6,7) may possibly be specifically related to the mammary enlargement rather than to the coronary collateral circulation.

Even though the measurements of retrograde flow, peripheral coronary pressure, and roentgenograms may fail to show the full extent of collateral function(11), the fact that the ECG reveals evidence of severe ischemia in most of these experimental animals following circumflex arterial occlusion is strong evidence against other collateral function undetected by these methods.

Although the results of these studies on normal dog hearts are clear, they do not necessarily apply to the hearts of patients where

hypoxia and intercoronary arterial pressure gradients may exist to provide more favorable conditions for collateral growth.

Summary. The effect of pregnancy and administered estrogen on the coronary collateral circulation of otherwise normal dogs was investigated. The data show that neither pregnancy nor estrogen given for 21 weeks resulted in significant alteration from controls in peripheral coronary occlusion, or in the roentgenographic appearance of the coronary arterial vasculature following injection with a barium gelatin mixture.

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Adrenal Steroid Secretion in Rabbits Following Prolonged ACTH Administration.* (29812)

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The principal secretory products of the adrenal cortex of adult mammals are corticosterone and cortisol. Bush has shown that the relative quantity of these compounds varies from species to species(1). He and other workers have demonstrated that the rabbit is predominantly a corticosterone producer, the corticosterone/cortisol ratio (B_K/F_K) being approximately 20:1(1,2,3).

Based on the observation that administration of corticosterone to rabbits had no effect on the lymphoid tissue whereas administration of ACTH had a pronounced effect, Kass *et al* investigated the secretion of the rabbit adrenal cortex following a 7-28-day-period of porcine ACTH administration(2). This treatment converted the rabbit into a predominantly cortisol producer.

In a recent review by Saba *et al*, ACTH is said to have two effects on adrenal secretion (4). The first effect is seen as an immediate rise in the output of the adrenal steroids without a change in their ratios. The second effect, after prolonged ACTH administration manifests itself as an alteration in the ratio of the adrenal products. To our knowledge this long term effect of ACTH administration is attributed primarily to the work of Kass and coworkers and has never been confirmed. The concept that prolonged ACTH administration changes the ratio of steroid output by the adrenal cortex is of sufficient importance that a repetition of the experiments seemed warranted.

Methods. Male rabbits weighing 2.7-3.5 kg were maintained on Purina rabbit pellets and had water *ad libitum*. The animals were divided into 3 groups. Porcine ACTH was given to Group I for 15 days and to Group II for 30 days. The porcine ACTH† was suspended in sesame oil and was administered twice daily subcutaneously to both

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