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Atherogenesis and Thrombogenesis in the Rat.* (30232)

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The relationship between atherosclerosis and thrombosis has been much investigated in recent years. Generally thrombosis has been regarded as a phenomenon secondary to atherosclerosis. Other possibilities have also been considered; namely, a) that atheromatous lesions may originate as mural thrombi and, b) that the 2 processes are independent of each other. The latter hypothesis in particular has been supported by recent publications claiming that certain diets high in saturated fats induce intravascular thrombosis in the rat with little structural changes in the vascular endothelium(1). Another report claimed the independent production of atherosclerosis and thrombosis by diets differing in fat composition(2). In view of such reports, our experience with similar diets in induction of cardiovascular lesions in the rat is presented here.

Materials and methods. Male Wistar albino rats weighing about 300 g were used. The diets given were the variations of the original diet used by Fillios and associates(3) and later modified by Thomas and Hartroft (1). The composition of these diets is given in Table I. The composition of the diet in Group I is similar to the diet reported to cause intravascular thrombosis and visceral infarction but little structural changes(1,2). In Groups II and III corn oil was substi-

TABLE I. Composition of Diets in the Various Groups in Percentage by Weight.

Ingredients	Group I	Group II	Group III
Cholesterol	5.0	5.0	5.0
Propylthiouracil	.3	.3	.3
Sodium cholate	2.0	2.0	2.0
Casein	20.0	20.0	20.0
Salt mixture	4.0	4.0	4.0
Vitamin mixture	2.0	2.0	2.0
Alphacel	6.0	6.0	6.0
Choline chloride	.2	.2	.2
Sugar	20.5	20.5	52.5
Butter	40.0	—	—
Corn oil	—	40.0	8.0

tuted for 40% butter at a level of 40% and 8% respectively. These diets are similar to the ones reported to cause atherosclerosis (2,3).

Pathology. Complete autopsies were done on all animals as soon as possible after death. Details of pathological methods employed have been published(4). Since few lesions were found in those animals that were maintained on the diet for less than 100 days, the analysis of the pathological results is based on the findings in animals that came to the autopsy after 100 days or more of the experimental period. Each animal was housed in an individual cage and fed *ad lib*.

Results and discussion. In the early weeks of the experiment there was loss of weight and a high incidence of mortality especially in the group receiving 40% butter. During this early period the animals that came to autopsy did not show significant intimal disease but rather showed the presence often of thrombus-like masses in cardiac chambers

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TABLE II. Incidence of Various Cardiovascular Lesions in all Animals in the 3 Groups Autopsied After 100 or More Days on Diet.

Diet group	40% butter	40% corn oil	8% corn oil
Total No.	31	18	37
Avg duration on diet (days)	128 (100-208)	162 (122-225)	228 (111-423)
Atheromata			
Aorta	9 (29%)	7 (39%)	15 (41%)
Coronary	6 (19%)	5 (28%)	14 (37%)
Thrombus like masses			
In cardiac chambers and great vessels	7 (23%)	4 (22%)	5 (14%)
In coronaries	1	0	1
Infarcts			
Heart	3	1	1

TABLE III. Incidence of Various Cardiovascular Lesions in Animals in the 3 Groups Maintained on the Diets for a Comparable Period and Autopsied After 100 or More Experimental Days.

Diet group	40% butter	40% corn oil	8% corn oil
Total No.	13	13	13
Avg duration on diet (days)	148 (125-208)	149 (117-171)	148 (111-199)
Atheromata			
Aorta	4	5	5
Coronary	5	5	4
Thrombus like masses			
In cardiac chambers and great vessels	7	3	4
In coronaries	1	0	1
Infarcts			
Heart	3	1	1

and great vessels. The animals that survived for over 100 days generally showed better defined structural changes including atheromatous lesions and occasional myocardial infarcts.

The incidence and site of various cardiovascular lesions in the 3 groups appear in Tables II and III. Table II includes the lesions in all the animals in the 3 groups that survived the diet for more than 100 days. It is noted that the *nature* of the lesions encountered in all groups was similar in that essentially 3 types of lesions were encountered in all groups; namely, atheroma, thrombus-like mass, and infarct. Moreover, the histopathological structure of the lesions was the same in all groups and details were similar to those published elsewhere(4).

The *incidence* and *severity* of the atheromatous lesions in any particular group seemed to be largely related to the length of time the animals had survived a given diet rather than the type of added fat. Length of sur-

vival of animals on such a severe diet regimen is variable and unpredictable. Suppression of thyroid activity by thiouracil and possibly toxicity by other ingredients in these diets together with a lessened nutritional status may make these animals vulnerable to environmental factors. Animals receiving 8% corn oil survived longer on the diet and had higher incidence of atheromatous lesions (Table II). Once this time factor was taken into account (Table III) and only the animals in the 3 groups that survived the diet for a comparable period of time were compared, we formed the impression that the animals receiving 40% butter developed equally severe atheromatous lesions. This is at variance with reports that this particular diet produces thrombosis with little structural changes(1,2). A shorter experimental period in the studies of these investigators could possibly explain the paucity of atheromatous lesions found. Certainly our experience indicates that if the animals survive on

the diet long enough, severe vascular lesions develop.

Thrombus-like masses were found in the cardiac chambers in all groups but appeared to be more numerous and pronounced in the animals receiving 40% butter. Occasionally such material was also found in the coronary vessels. These masses had a high content of lipids and a low content of fibrin, platelets, and leukocytes. They generally lay free of the endothelium, only occasionally having a point of adherence to the vascular endothelium or to an atheromatous plaque. For this reason, unlike other investigators(1,2), we are not satisfied that these thrombus-like masses were formed ante-mortem. They could have been simply a sequel to agonal clotting changes or occurred possibly post-mortem. This view is strengthened by the fact that these masses are often much too extensive (filling the cardiac chambers and aorta) to have been compatible with survival for any length of time. Furthermore we have not found them in animals that were exsanguinated and then sacrificed. It must be emphasized, however, that these masses were not seen in chow-fed control animals dying "spontaneously" during the course of the experiment. Such deaths are usually attributable to pulmonary infection. Since we have found the fibrinolytic activity of the blood to be higher in chow-fed animals compared to those fed 40% butter(4), the possibility occurred that in chow-fed animals normal post-mortem clots are lysed, whereas in animals on high fat diets no lysis occurs and thus thrombus-like masses are seen.

The incidence of visceral infarction may have been greater in animals receiving 40% butter compared to those receiving corn oil. Since the numbers are small, however, the statistical significance is uncertain. More-

over, we have not been able to ascertain that arterial occlusion, whether by thrombus or atheroma, was the cause of these infarcts and as such, unlike other investigators, we have reservations as to the correspondence of these infarcts to those found in man.

Summary and conclusions. Three groups of rats were given diets containing cholesterol, sodium cholate and propylthiouracil. In addition the diets contained a fat that consisted of 40% butter in Group I, 40% corn oil in Group II and 8% corn oil in Group III. All groups showed atheromatous lesions and thrombus-like masses in cardiac chambers and in great vessels. Infarcts were present in a small number of animals in each group. Incidence and severity of atheromatous lesions varied but this variation was mainly related to length of survival on the diet rather than the type of added fat. There was no isolated development of thrombosis related to the type of fat in the diet as suggested by some investigators. Moreover, reservations were expressed regarding the exact nature of the thrombus-like masses and infarcts and their relevance to atherosclerosis, thrombosis, and infarction in man.

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