DIET-INDUCED HYPERTENSION AND CARDIOVASCULAR LESIONS IN MICE:

The effects on the coronary arteries of feeding a normal diet to mice previously fed a high-fat diet have been described. Following the first week of "recovery" (feeding a normal diet) there was a high incidence of medial hyalinization and a mural and perivascular infiltration of inflammatory cells. After two and three weeks of recovery a perivascular fibrosis and subendothelial hyperplasia of smooth muscle had developed. Medial circular smooth muscle was also hyperplastic. These lesions resemble those of experimentally induced hypertension. Previous studies have shown that the arterial system of mice is relatively resistant to damage produced by atypical diets. In the coronary arteries of mice restricted to the high-fat diet for as long as 78 weeks, lesions were limited in severity and incidence. Arterial lesions of rabbits fed an atherogenic diet are enhanced when the animals are returned to a normal diet. In swine and dogs, alternating periods of starvation and refeeding produce hypertension and mural vascular lesions.

This report considers the relationship between hypertension and the arterial lesions produced in mice fed a normal diet following seven weeks of restriction to a high-fat, hypolipotropic diet.

MATERIALS AND METHODS

Animals. Young adult nulliparous mice (8-12 weeks of age ranging in weight from 28-32 gm.) of the Taconic Swiss (TS) stock were used. The mice were divided into two groups, the recovery animals and the experimental controls. Animals in the recovery group received the experimental diet for seven weeks and were then fed a normal diet (Purina Laboratory chow) for a period of one to three weeks. Representative animals were killed after one, two, or three weeks of recovery. The experimental controls were fed the experimental diet for periods of 7-10 weeks and were killed at intervals of seven, eight, nine, and ten weeks. Blood pressure and hematocrit determinations were made prior to death by cervical compression. Blood pressure and hematocrit levels were also determined in normal mice receiving no treatment and maintained on Purina Laboratory chow.

* N.I.H. Predoctoral Trainee in the Anatomical Sciences (5 T01 GM00287).
** Associate Professor of Anatomy.
† Professor of Anatomy.
‡ Supported by grants 5 T01 GM 00287 and HE 04052 from the National Institutes of Health.

Received for publication 11 June 1970.
Diets. The basic experimental diet has been used in other studies. Here, a relatively unsaturated fat, cod liver oil, has been substituted for lard, its more saturated counterpart. The composition of this diet is as follows:

| Ingredient                      | gm/100 |
|---------------------------------|--------|
| Sucrose                         | 57.5   |
| Cod liver oil                   | 28.0   |
| Casein (vitamin free)           | 8.0    |
| Salt mixture no. 2 (U.S.P. XIII)| 4.0    |
| L-cystine                       | 0.5    |
| Vitamin mixture                 | 2.0    |

The vitamin and salt mixtures are considered adequate and have been previously described.10 The diet was not supplemented with a lipotropic material and is obviously high in fat and carbohydrate and low in protein. Feeding the diet and housing the mice have also been described.10 To limit oxidation of fats and fat-soluble vitamins, the experimental diet was prepared twice weekly and refrigerated with small amounts of the diet being added to the feeders as needed.

Histological methods. These procedures have been described earlier.7 At least twelve complete frontal sections of each heart were studied. In addition to the hearts, samples of kidney, lung, liver, spleen, pancreas, aorta, and adrenals were also studied.

Blood pressure determinations. The method has been described in detail.10 The mice were anesthetized with intraperitoneal sodium pentobarbital, 0.06 mg. per gram of body weight. The anesthetized mice (after 10 minutes at room temperature) were placed in a warming box for nine minutes at 35° C. prior to the pressure readings. An auscultatory microphone (Carter Electronics) was attached to the mouse's tail distal to an occlusion cuff. The microphone, picking up the Korotkoff sounds, was monitored through attachments to a Grass recorder and oscilloscope. The occlusion cuff was attached to a Tyco aneroid manometer for reading blood pressure.

Hematocrit determinations. Venous blood was obtained from the orbital sinus by standard heparinized microhematocrit capillary tubes. The blood was spun four minutes at approximately 12,000 r.p.m. on an Adams Autocrit centrifuge.

Statistics. Statistical analyses were calculated by the "Student test."10

OBSERVATIONS

Blood pressure. Systolic blood pressures were determined in two separate groups, 10 and 15 mice each, of non-dietary controls at three-week intervals. The mean systolic pressures in these groups were 86.1 and 85.6 mm Hg. When these two groups were pooled, the mean pressure was 85.8 mm Hg (Fig. 1). After receiving the experimental, high-fat, low-protein, diet for seven weeks the mean systolic pressure decreased to 52.1 mm Hg (p < 0.001). Animals fed this atypical diet (dietary controls) for as long as 10 weeks continued to be hypotensive. The highest mean in this group was 59.6 mm Hg at nine weeks.

Figs. 1-3. Graphs plotting systolic pressure (mm Hg), hematocrit (%) and weight change (%) against time (in weeks). The dotted line represents weeks on the experimental or deficient diet and the solid line weeks on the normal or recovery diet.
Following a period of one week of recovery (return to a normal diet) preceded by one of eating the experimental diet for seven weeks, there was a sharp rise (56.9 mm Hg) in mean systolic pressure from 52.1 to 109.0 mm Hg (p<0.001). Pressures had returned to approximately normal values after two weeks of recovery. There was no significant difference (p>0.5) in this group at 83.1 mm Hg as compared to non-dietary controls. Although the pressure continued to fall during the third week of recovery, it appeared to be stabilizing. Again, there was no significant difference between this group at 78.0 mm Hg as compared to non-dietary controls (p>0.5).

**Hemocrit determination.** The results of hemocrit determinations are illustrated in Figure 2. The mean initial hemocrit in normal or non-dietary controls was 46.3%. Following five weeks of feeding the experimental diet, the mean hemocrit had decreased to 24% but the range was wide. By the seventh week the mean had declined to 9.8% with a very restricted range. Hematocrits remained below 15% in mice which had been continued on the experimental diet for as long as 10 weeks.

The hemocrits increased rapidly in mice returned to a diet of laboratory chow. At the end of one week of recovery the hemocrits had risen to a mean value of 41.8%, slightly below the normal value of 46.5%. After two and three weeks the hemocrits seemed to be leveling off at values of 45.1% and 46.6% respectively.

**Weight changes.** Initially all mice weighed between 28 and 32 gm. (mean 29.4 gm.). Weight change was highly variable in animals fed the experimental diet (Fig. 3). There was a gradual loss of weight averaging —26.3% at seven weeks. Mice remaining on the high-fat, low-protein, and hypolipotropic diet after seven weeks continued to lose weight. After one week of recovery the mean weight change was —11.7% and at three weeks had risen to +16.2%.

**Arterial lesions.** A complete description of lesions identical to those observed here has been reported. In this earlier study the experimental diet contained lard as fat rather than cod liver oil.

In mice fed the experimental diet only (Table 1) for periods ranging from 7-10 weeks the only change observed was a low incidence of restricted mural hyalinization of the intramyocardial branches of the coronary arteries (Fig. 4). Although these hyalin deposits were usually focal, the affected portions of the vessel were swollen and all layers of the arterial wall were frequently involved. Necrotic medial smooth muscle was observed occasionally in the more severely damaged vessels. Only one vessel in this group contained a site of leukocytic response and this appeared to be a reaction to an adjacent focus of myocardial necrosis.
TABLE 1. INCIDENCE (%) OF ARTERIAL LESIONS IN MICE FED EXPERIMENTAL DIET

| Experimental regimen | Received experimental diet only for 7-10 weeks | Received plus seven days recovery on normal diet* | Received plus fourteen days recovery on normal diet* | Received plus twenty-one days recovery on normal diet* |
|----------------------|---------------------------------------------|-----------------------------------------------|-----------------------------------------------------|------------------------------------------------------|
| Number of mice       | 75                                          | 20                                             | 15                                                  | 18                                                   |
| Lesions:             |                                             |                                               |                                                     |                                                      |
| Hyalinized and/or necrotic arteries | 17%                                      | 52%                                          | 73%                                                 | 33%                                                  |
| Polyarteritis (with and without mural hyalinization) | 1%                                       | 24%                                          | 80%                                                 | 17%                                                  |
| Perivascular fibrosis and smooth muscle hyperplasia | 0%                                        | 47%                                          | 67%                                                 | 50%                                                  |
| * Purina Laboratory Chow. |

After one week of recovery, the incidence of mural hyalinization in arteries had increased considerably, as to involvement of hearts as well as incidence in the group (Table 1). Although medial hyalinization remained the most frequent lesion, but with an increased incidence (from 17% to 52%), an acute arteritis had developed in many vessels and involved all layers of the arterial wall and extended into the adjacent myocardium. This inflammatory exudate was cellular, fibrinous, or both (Fig. 5). The cellular response was chiefly of mononuclear leukocytes. Hyaline substance had been completely removed from some of the more advanced arterial lesions.

Following the acute arteritis a condition or stage characterized by adventitial and periadventitial fibrosis with an apparent hyperplasia of smooth muscle of the media and intima was seen. This response, not seen in mice fed only the experimental diet, was observed in 47% of the mice after one week of recovery. Hyalinization and arteritis reached their peaks during the second week of recovery with respective incidences of 73% and 80%. The perivascular fibrosis and smooth muscle hyperplasia had attained an incidence of 67% by the second week and continued to be the predominant changes after three weeks (Table 1). The earliest evidence of perivascular fibrosis was the replacement of leukocytes by fibroblasts within the adventitia. The degree of fibroblastic proliferation differed from vessel to vessel and was associated with varying amounts of collagenesis. Smooth muscle hyperplasia began in the medias of recovery vessels although occasionally a
few longitudinally oriented fibers could be located within the intima, between the endothelium and a fragmented internal elastic membrane.

By three weeks of recovery the circularly oriented smooth muscle of the media was several layers thick. The longitudinally oriented smooth muscle of the intima had also thickened, constricting the lumen of many vessels. In some vessels the proliferation was even and concentric while in others the hyperplasia was unevenly distributed. This resulted in several protruding thickenings or cushions creating an eccentrically located and/or stellate shaped lumen (Fig. 6). The intimal thickenings were also characterized by an intimal fibrosis composed of reticular, collagenous, and elastic fibers (Fig. 7). By the third week of recovery the incidences of hyalinization and arteritis had decreased.

Other organs. Although extensive fatty changes and ceroid deposition occurred within the livers of these animals, it is not the purpose of this study to report these findings. In addition to the coronary arteries, similar vascular lesions were also seen in the hepatic arteries of recovery mice. Occasionally vascular changes, typical of recovery were seen in small arteries of the superior mediastinum. The kidneys showed no vascular changes and inflammation was infrequent. The adrenals were apparently normal with varying amounts of ceroid in the zona reticularis. None of the other organs studied, lung, spleen, pancreas and aorta, demonstrated vascular alterations. The intestines and mesenteric vessels were not studied.

DISCUSSION

In this study mice were fed a normal diet of Purina Laboratory chow following restriction to an experimental (high fat, low protein, and hypolipotrophic) diet for a period of seven weeks. Mice fed the experimental diet developed hypotension. Similar diets with added thiouracil produce hypotension in rats.* Prolonged periods of malnutrition or deprivation of food also result in hypotension.** Furthermore, hypotension has been shown to produce arterial lesions.*** An attempt was made to prevent a possible toxic effect of dietary lipid oxidation. This toxicity, if present, will not produce the recovery lesions by itself as they are not seen in animals fed the experimental diet only as long as ten weeks.

During the first week of recovery (eating the normal diet) the systolic pressures rose rapidly to levels above that of controls. Although the degree of absolute hypertension was not too great (109.0 mm Hg), the relative hypertension was statistically significant with an increase of 56.9 mm Hg. The earliest coronary artery lesions were also seen at this time, thus representing a maximum of one week for development of hypertension and the
Fig. 4. (Figures 4-7 show intramyocardial branches of coronary arteries of mice). Heart from mouse fed the experimental diet only for seven weeks. Arrow indicates a portion of the vessel wall that is swollen and hyalinized. Endothelial cells are swollen but intact. PAS and H. ×200.

Fig. 5. Heart from mouse fed a normal diet for one week after seven weeks on the experimental diet. Acute arteritis involving all layers of the vessel wall and extending into the adjacent myocardium is demonstrated. The exudate consists of both cellular and fibrinous components. Many of the endothelial cells are vacuolated and the internal elastic membrane is fragmented. PAS and H. ×200.

Fig. 6. Heart from mouse fed a normal diet for three weeks after seven weeks on the experimental diet. The size of the arterial lumen has been severely reduced by hyperplasia of smooth muscle. PAS and H. ×400.

Fig. 7. Heart from mouse fed a normal diet for three weeks after seven weeks on the experimental diet. The reticular pattern surrounding the intimal smooth muscle and adventitial and periadventitia fibrosis are demonstrated. Reticulum plus PAS method. ×400.
vascular lesions. According to Wilson and Byrom,\textsuperscript{39} the severity of the arterial lesions associated with hypertension is dependent upon the acuteness of the onset of hypertension rather than the height or duration. Here it was not possible to determine whether the hypertension contributed to or was a sequela of the vascular lesions. It seems reasonable to assume that the hypertension was primary since it was transient while the lesions were progressive.

Studies in man have shown that severe hypertension develops upon return to a normal diet following prolonged periods of malnutrition.\textsuperscript{40,44} In dogs and swine, alternating periods of starvation and refeeding produce hypertension and hypertensive vascular changes.\textsuperscript{45-47} Studies in rabbits, blood pressures not recorded, have shown that cholesterol atherosclerosis is enhanced and the cytomorphology of the lesions altered by returning the animals to a normal diet.\textsuperscript{48-54} In rats, atherosclerosis is also accelerated by concurrent hypertension.\textsuperscript{55,56,57} Again, the actual cause and effect relationship was not determined. The results of the present study demonstrate an episode of hypertension related to the development of arterial lesions but they do not explain the etiology of the hypertension.

Selye\textsuperscript{58} states that chronic undernourishment with protein-deficient diets is stressful, but the stress reaction is unusual in that there is no adrenal stimulation. Handler and Bernheim\textsuperscript{59} reported a diminished ACTH production in rats on low protein diets. The severe stress of dietary regimen such as the one used has been mentioned by several investigators.\textsuperscript{60,61} Possibly, the hypertension associated with refeeding is due to stress and mediated via the adrenals. However, no adrenal changes were observed in mice used in this study.

Mural hyalinization and fibrinoid degeneration, as shown in the recovery lesions, are characteristic of hypertensive vascular injury.\textsuperscript{62} The severe mural and periarterial reaction is not so pronounced in man\textsuperscript{62} but is characteristic of hypertensive disease in rats.\textsuperscript{63} Medial smooth muscle hyperplasia and hypertrophy is a well known response to the increased intraluminal pressure of hypertension.\textsuperscript{64} Cellular hyperplasia internal to the internal elastic membrane, identified here as smooth muscle, is typical in many forms of vascular injury including acute, degenerative or malignant hypertension.\textsuperscript{65-67} The intimal foam cells characteristic of atherosclerosis are also considered as smooth muscle derivatives and/or smooth muscle-like cells.\textsuperscript{68-70} The difference in their appearance and the hyperplastic cells observed here may be dependent upon the presence of a hyperlipemia during or following the proliferative phase.\textsuperscript{71} Hyperlipemia could not be substantiated in mice fed the experimental diet used here.\textsuperscript{72} However, the mice were fasted prior to blood sampling.
SUMMARY

Mice fed a high-fat, low-protein, hypolipotropic diet for 7-10 weeks lost body weight (−26.3%), became progressively anemic (Hct. 9.8%) and hypotensive (mean pressure 52.1 mm Hg compared to 85.8 mm Hg in controls). A low incidence (17%) of mural hyalinization and/or fibrinoid necrosis of the coronary arteries occurred in this group.

A second group of mice received the experimental diet (as above) for seven weeks, were then fed an adequate diet of commercial laboratory chow for one to three weeks before being killed. During the first week of recovery (feeding of the normal diet) hematocrits returned to normal values (41.8%) and a transient hypertension developed, reaching its peak (109 mm Hg) after one week and returning to normal or subnormal levels (78.0 mm Hg) by three weeks. A high incidence (52%) of damage to the coronary arteries, well correlated with the hypertensive episode, also appeared during the first week. This damage consisted chiefly of medial hyalinization that progressed rapidly to an acute arteritis. After two to three weeks the predominant lesions were medial and intimal hyperplasia of smooth muscle and periarterial fibrosis.

REFERENCES

1. Ashburn, A. D., Smith-Vaniz, G. T., Wilson, J. L., and Williams, W. L.: Changes in coronary arteries of mice fed a high-fat, low-protein diet followed by a normal diet. Anat. Rec., 1969, 165, 379-389.
2. Skelton, E. R.: Development of hypertension and cardiovascular-renal lesions during adrenal regeneration in the rat. Proc. Soc. Exp. Biol. (N.Y.), 1955, 90, 342-346.
3. Masson, G. M. C., McCormak, L. J., Dustan, H. P., and Corcoran, A. C.: Hypertensive vascular disease as a consequence of increased arterial pressure. (Quantitative study in rats with hydralazine-treated renal hypertension.) Amer. J. Path., 1958, 34, 817-834.
4. Crane, W. A. J. and Dutta, L. P.: The utilization of tritiated thymidine for deoxyribonucleic acid synthesis by the lesions of experimental hypertension in rats. J. Path. Bact., 1963, 86, 83-97.
5. McGee, W. G. and Ashworth, C. T.: Fine structure of chronic hypertensive arteriopathy in the human kidney. Arch. Path., 1963, 71, 96-102.
6. Crane, W. A. J. and Ingle, D. J.: Tritiated thymidine uptake in rat hypertension. Arch. Path., 1964, 78, 209-221.
7. Spiro, D., Lattes, R. G., and Wiener, J.: The cellular pathology of experimental hypertension. I. Hyperplastic arteriolar sclerosis. Amer. J. Path., 1965, 47, 19-49.
8. Koletsky, S., Rivera-Velez, J. M., and Pritchard, W. H.: Production of hypertension and vascular disease by angiotensin. Arch. Path., 1966, 82, 99-106.
9. Clarkson, T. B.: Atherosclerosis—spontaneous and induced. In, Advances in Lipid Research, by Rodolfo Paoletti and David Kritchevsky (Eds.). New York and London, Academic Press, 1963, pp. 211-252, Vol. I.
10. Ball, C. R., Williams, W. L., and Collum, J. M.: Cardiovascular lesions in Swiss mice fed a high fat, low protein diet with and without betaine supplementation. Anat. Rec., 1963, 145, 49-60.
11. Thomas, H. M., Williams, W. L., and Clower, B. R.: Cardiac lesions in C mice, result of choline-deficient and choline-supplemented diets. Arch. Path., 1968, 85, 532-538.
12. Constantinides, P., Booth, J., and Carlson, G.: Production of advanced cholesterol atherosclerosis in the rabbit. Arch. Path., 1960, 70, 712-724.

13. Buck, R. C.: Lesions in the rabbit aorta produced by feeding a high cholesterol diet followed by a normal diet. An electron microscopic study. Brit. J. exp. Path., 1962, 43, 236-240.

14. Imai, H., Lee, K. T., Pastori, S., Panfilio, E., Florentin, R., and Thomas, W. A.: Atherosclerosis in rabbits. Architectural and subcellular alterations of smooth muscle cells of aortas in response to hyperlipemia. Exp. molec. Path., 1966, 5, 273-310.

15. Wilhelmj, C. M., Meyers, V. W., and McCarthy, H. H.: Diastolic hypertension produced by high fat diets and dietary stresses. Proc. Soc. Exp. Biol. (N.Y.), 1957, 95, 801-804.

16. Smith, G. S., Smith, J. L., Mameesh, M. S., Simon, J., and Johnson, B. C.: Hypertension and cardiovascular abnormalities in starved-refed swine. J. Nutr., 1964, 82, 173-182.

17. Hembrough, F. B. and Link, R. P.: Capacitance changes in the arterial system of swine induced by starvation and refeeding. Proc. Soc. Exp. Biol. (N.Y.), 1968, 122, 1055-1061.

18. Clarke, T. D., Ashburn, A. D., and Williams, W. L.: Cortisone-induced hypertension and cardiovascular lesions in mice. Amer. J. Anat., 1968, 123, 429-440.

19. Stone, S. H.: Method of obtaining venous blood from the orbital sinus of the rat or mouse. Science, 1954, 119, 100.

20. Sorg, D. A. and Buckner, B.: A simple method of obtaining venous blood from small laboratory animals. Proc. Soc. Exp. Biol. (N.Y.), 1964, 115, 1131-1132.

21. Snedecor, G. W.: Statistical Methods. Ames, Iowa, The Iowa State University Press, 1956, p. 45.

22. Dustin, P., Jr.: Arteriolar hyalinosis. In, International Review of Experimental Pathology, by G. W. Richter and M. A. Epstein (Eds.). New York and London, Academic Press, 1962, pp. 73-138, vol. 1.

23. Karsner, H. T.: Acute inflammations of arteries. In, American Lecture Series: American Lectures in Pathology, by Paul R. Cannon (Ed.). Springfield, Ill., Charles C. Thomas, 1947, No. 6, pp. 4-37.

24. Koletsky, S., Roland, C., and Rivera-Velez, J. M.: Rapid acceleration of atherosclerosis in hypertensive rats on high fat diet. Exp. molec. Path., 1968, 9, 322-338.

25. Burger, G. C. E., Sandstead, H. R., and Drummond, J.: Starvation in Western Holland. Lancet, 1945, 2, 282-283.

26. Spies, T. D. and Stone, R. E.: Hypotension associated with nutritive failure. Proc. Soc. Exp. Biol. (N.Y.), 1949, 72, 368.

27. Keys, A., Brozek, J., Henschel, A., Mickelsen, O., and Taylor, H. L.: The Biology of Human Starvation, Vol. I. Minneapolis, University of Minnesota Press, 1950, pp. 607-634.

28. Duff, G. L.: In, Symposium on Atherosclerosis, Publication No. 338 of the National Academy of Sciences and the National Research Council U.S.A., Washington, D.C., 1954, p. 33.

29. Williams, G.: Experimental studies in arterial ligation. J. Path. Bact., 1956, 72, 569-574.

30. Buck, R. C.: Intimal thickening after ligation of arteries, an electron-microscopic study. Circulat. Res., 1961, 9, 418-426.

31. de Faria, J. L.: Histopathological changes in the coronary arteries following shock or hypotension in man. Their relations to thrombosis and atherogenesis. Path. Microbiol. (Basel), 1963, 26, 385-398.

32. Wilson, C. and Byrom, F. B.: Renal changes in malignant hypertension. Experimental Pathology, Lancet, 1939, I, 130-139.

33. Brozek, J., Chapman, C. B., and Keys, A.: Drastic food restriction; effect on cardiovascular dynamics in normotensive and hypertensive conditions. J. Amer. med. Ass., 1948, 137, 1569-1574.

34. Itahara, K., Fukuchi, S., Fujibayashi, T., and Yamaguchi, M.: Frequency of essential hypertension and the influence of environmental condition thereon. Tohoku J. exp. Med., 1955, 61, 231-244.
35. Deming, Q. B., Mosback, E. H., Bevans, M., Daly, M. M., Abell, L. L., Martin, E., Brun, L. M., Halpern, E., and Kaplan, R.: Blood pressure, cholesterol content of serum and tissues, and atherogenesis in the rat. J. exp. Med., 1958, 107, 581-598.

36. Eades, C. H., Jr., Phillips, G. E., Blaustein, A., Hsu, I. C., and Solberg, V. B.: Coronary atherosclerosis. I. Contrast of DOCA hypertension with renal hypertension in the etiology of coronary atherosclerosis in the adult male wistar rat. Angiologica (Basel), 1965, 2, 61-76.

37. Selye, H.: The Physiology and Pathology of Exposure to Stress. Montreal, Canada, Acta, Inc., Medical Publishers, 1950, pp. 47, 324.

38. Handler, P. and Bernheim, F.: Effect of choline deficiency on ACTH production and on hypertension of subtotaly nephrectomized rats. Amer. J. Physiol., 1950, 162, 375-378.

39. Wilhelmj, C. M., Gunderson, D. E., Shuput-Meyers, D., and McCarthy, H. H.: The effect of diet in the blood pressure and heart rate of normal dogs. Animal fat. Amer. J. dig. Dis., 1955, 22, 219-227.

40. Wilhelmj, C. M., Shuput-Meyers, D., and McCarthy, H. H.: Prolonged diastolic hypertension of dietary origin. Exp. Med. Surg., 1956, 14, 286-298.

41. Heptinstall, R. H.: Malignant hypertension: A study of fifty-one cases. J. Path. Bact., 1953, 65, 423-439.

42. Cromartie, W. J.: Arteritis in rats with experimental renal hypertension. Amer. J. med. Sci., 1943, 206, 66-75.

43. Baumgartner, H. R. and Studer, A.: Controlled over-dilatation of the abdominal aorta in normo and hypercholesteremic rabbits. Path. Microbiol. (Basel), 1963, 26, 129-148.

44. Geer, J. C., McGill, H. C., Jr., and Strong, J. P.: The fine structure of human atherosclerotic lesions. Amer. J. Path., 1961, 38, 263-287.

45. Haust, M. D., Balis, J. U., and More, R. H.: Electron microscopic study of intimal lipid accumulations in human aorta and their pathogenesis. (Abstract). Circulation, 1962, 26, 656.

46. Constantinides, P.: Experimental Atherosclerosis. New York, Elsevier Publishing Co, 1965, p. 26.

47. Ball, C. R.: Hematologic studies of mice fed a thrombogenic diet. Arch. Path., 1968, 85, 547-553.