Despite discrepancies in experimental findings, it would appear that the initial antibody produced in poikilotherms constitutes a rapidly sedimenting one with $\gamma_1$-globulin electrophoretic mobility, and therefore, is the one most commonly observed. Even though $\gamma_2$-globulins are observed, they appear to lack specific antibody activity when specific activity has been found in the $\gamma_1$-globulins(12,13). Uhr *et al*(14) report that in the frog and goldfish, antibody activity is associated with $\gamma_2$-globulins. As has been pointed out by workers in this field, the known variations in the immune response to different antigens, antigen dosages, and time of immunization are factors that may contribute to the differences observed. Our finding of at least 2, and possibly 3, electrophoretic components in the alligator with specific antibody activity may reflect an evolutionary trend, but, it may also reflect the injection schedule followed.

*Summary and conclusions.* Alligators can be actively immunized with hemocyanin, a protein antigen, and the circulating antibody consists of at least 2 kinds of immunoglobulin that differ in electrophoretic mobility and perhaps size. Results indicate that alligators, in contrast to other reptiles, may possess a primitive type of immunological memory. Although the production of circulating antibody by the alligator was not unexpected, the nature of the immune response and of the anti-KLH immunoglobulins in the alligator appear to differ enough from that reported for other poikilotherms to warrant further study, especially from an evolutionary standpoint.

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## Atherosclerosis in Denervated and Intact Carotid Arteries in Rabbits.* (31762)

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Evidence indicating that the autonomic nervous system, both through the adrenal medulla and directly through its innervation of adipose tissue, is involved with lipid metabolism, chiefly the mobilization of free fatty acids (FFA), has been well reviewed(1-3). Stimulation of nerves innervating adipose tissue has been shown to increase FFA release (4). The process of atherosclerosis occurs in vascular channels which possess a rich adrenergic innervation. Do these sympathetic nerves exert any appreciable influence on the

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TABLE I. Summary of Data on Aortic and Carotid Atherosclerosis and Other Observations.

| Animal No. | Operation (side) | Miotic pupil (side) | Weeks on diet | Degree of atherosclerosis* | | | Serum cholesterol, mg % | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | Aorta | Carotids Left | Carotids Right | 2 wk | 17 wk | 35 wk |
| 39 | None | None | 0 | 0 | 0 | 0 | — | — | — |
| 19 | L | L | 0 | 0 | 0 | 0 | — | — | — |
| 40 | None | None | 0 | 5 | 0 | 0 | — | — | — |
| 9 | L | L | 19 | 10 | 0 | 0 | 80 | 1025 | — |
| 10 | R | R | 40 | 10 | 0 | 0 | 380 | 830 | 895 |
| 12 | R | R | 20 | 20 | 5 | 5 | 130 | 1010 | — |
| 14 | None | None | 20 | 40 | 5 | 5 | — | — | — |
| 24 | R | R | 30 | 40 | 0 | 0 | 70 | 1120 | 1100 |
| 37 | L | L | 39 | 50 | 5 | 0 | 84 | 1710 | 930 |
| 7 | L | L | 44 | 55 | 5 | 5 | 130 | 975 | 920 |
| 31 | R | R | 18 | 60 | 0 | 0 | 60 | 1440 | — |
| 4 | L | L | 39 | 55 | 25 | 35 | 82 | 945 | 630 |
| 21 | L | L | 53 | 55 | 20 | 10 | 120 | 830 | 960 |
| 5 | R | R | 19 | 60 | 10 | 10 | 175 | 1250 | — |
| 34 | R | R | 39 | 75 | 50 | 25 | 42 | 990 | 695 |
| 17 | R | R | 53 | 75 | 15 | 15 | 190 | 990 | 1025 |
| 28 | L | L | 54 | 65 | 15 | 5 | 44 | 1135 | 1040 |
| 20 | R | R | 54 | 80 | 10 | 20 | 70 | 750 | 735 |
| 27 | L | L | 40 | 90 | 10 | 10 | 170 | 1150 | 1170 |
| 33 | R | R | 54 | 90 | 15 | 15 | 44 | 1185 | 830 |
| 29 | L | L | 54 | 100 | 25 | 25 | 180 | 1665 | 895 |
| 25 | L | L | 54 | 100 | 75 | 25 | 140 | 1410 | 1280 |

* % of surface area demonstrating sudanophilia.

dynamic equilibrium of lipid deposition and mobilization occurring in these arterial vessels? Does their stimulation result, as it does in adipose tissue, in the mobilization of FFA? Would denervation of arterial channels impede lipid turnover and result in a net increase in deposition? If the answers to these questions were affirmative, this would be of importance in the basic understanding of the pathogenesis of atherosclerosis. In addition, it would be of potential clinical significance as the surgical procedure of sympathectomy is still widely practiced and might conceivably result in accelerated atherosclerosis in denervated vascular channels.

This study was undertaken to evaluate what effect, if any, sympathetic denervation of a blood vessel, one carotid artery, would have on the rate of atherosclerosis in that vessel as compared with its neurally intact partner. In addition, atheromatous deposition in the contralateral intact and ipsilateral adrenergically denervated iris and cornea was studied.

*Methods.* Sympathetic denervation of one carotid artery, 10 left and 9 right, was accomplished in rabbits by exposing the common carotid artery in the neck and excising the stellate, superior, middle and inferior cervicle ganglia as well as stripping off all visible nerves coursing to the carotid artery. The completeness of denervation was confirmed by the postoperative presence of a markedly miotic pupil as well as dilatation of the arterioles in the ipsilateral ear. In all animals, the pupil remained miotic throughout the duration of the study.

After the animals had recovered from the operation, 19 were given a diet of Purina rabbit chow to which was added 2% of corn oil and 0.3% of cholesterol. Three animals, 1 operated and 2 unoperated, received laboratory chow without additions.

Serum cholesterol determinations(6) were made at intervals. Five animals were sacrificed approximately 20 weeks after the feeding period was started, one at 30 weeks when it appeared in poor condition, and the remainder after 40 to 54 weeks on the diet. The heart, aorta, and carotids were removed, washed, fixed in formalin, and stained with Sudan IV in alcohol. The area of the aorta and carotid surfaces covered by lipid deposits was estimated. The relative amounts of lipid deposited in the cornea and iris of each eye were also recorded.

*Results.* A brief summary of the results is presented in Table I. As expected, none of

the control animals developed a significant amount of atherosclerosis and their corneas and irises remained free of lipid deposits. All animals fed cholesterol developed some degree of atherosclerosis of the aorta. The degree of involvement varied greatly and was not clearly dependent upon the length of time the diet was fed or the level of hypercholesterolemia which developed. Plaques apparently formed first and were most extensive at the ostia of vessels leaving the abdominal aorta and at its bifurcation with the iliac vessels. The extent of atherosclerosis in the aorta was always substantially greater than that in the carotids. In the carotid arteries the atheromatous plaques were most common at the bifurcation of the common carotid into its internal and external branches. No consistent difference in the degree of atherosclerosis in the denervated and neurally intact carotid vessels was observed. Similarly, there were large individual differences in the extent of lipid deposition in the corneas and irises but no apparent difference between the intact and denervated sides.

Many of the animals which had received the diet for long periods of time developed fatty cirrhosis. Overall weight gains were not impressive. Although all of the animals gained weight originally, a decrease from the maximum weight achieved was common after 6 to 8 months on the diet.

*Discussion.* The level of dietary cholesterol chosen for these studies was lower than that which has been used by many investigators. It seemed likely that if denervation did influence the deposition of lipid, this would be more evident if massive doses were not used. Hypercholesterolemia developed at very different rates and to different extents in the individual animals but by 17 weeks the serum cholesterol levels approached 1,000 mg% or more in all animals (Table I). Serum cholesterol levels at any particular time were poor predictors of the degree of atherosclerosis observed at autopsy. The levels of serum cholesterol at 35 weeks, which were approximately the maximum levels reached, have been plotted against the extent of atherosclerosis observed in the aorta in Fig. 1. The data suggest that the animals may have represented 2, perhaps 3, different populations which varied in susceptibility to plaque formation. Under the conditions of the study where the plaques developed over rather long periods, it might be expected that individual difference in susceptibility might have an optimum opportunity to express itself.

Extreme variability in the animals was evident in the extent of atherosclerosis which developed in the aorta, carotids, and eyes. The carotids were, however, uniformly less affected than the aorta, particularly the abdominal aorta. Although it may be argued that regeneration of the sympathetic nerves to the carotids might have occurred during the months following surgery, the pupils remained miotic throughout the study. Since there is no indication of any consistent asymmetry in the extent of lipid deposition in the innervated and denervated carotids, it appears highly unlikely that neural innervation plays a significant role in the pathogenesis of atherosclerosis in these structures.

The relative resistance of the carotid vessels to lipid deposition as compared to the aorta is consistent with the findings in man by Fisher *et al*(5) who noted that in 178 autopsies the degree of atherosclerosis and calcium deposition was greater in the aorta than in the carotid arteries. They also found both carotid arteries to be equally involved and most extensive at the carotid bifurcation.

*Summary.* The effect of sympathetic denervation of one carotid artery, iris and cornea on the rate of atherosclerosis in these structures as compared to its neurally intact partner was studied in 19 male albino rabbits fed a diet containing 0.3% of cholesterol. Animals were sacrificed after receiving the diet for 18 to 54 weeks. The levels of serum cholesterol rose at markedly different rates and to different degrees. Similarly, the extent of atherosclerosis showed large individual differences not clearly related to the degree of hypercholesterolemia. The carotid vessels were less affected than the aorta and no consistent asymmetry in lipid deposition in the carotid vessels, iris or cornea was observed following unilateral adrenergic denervation.

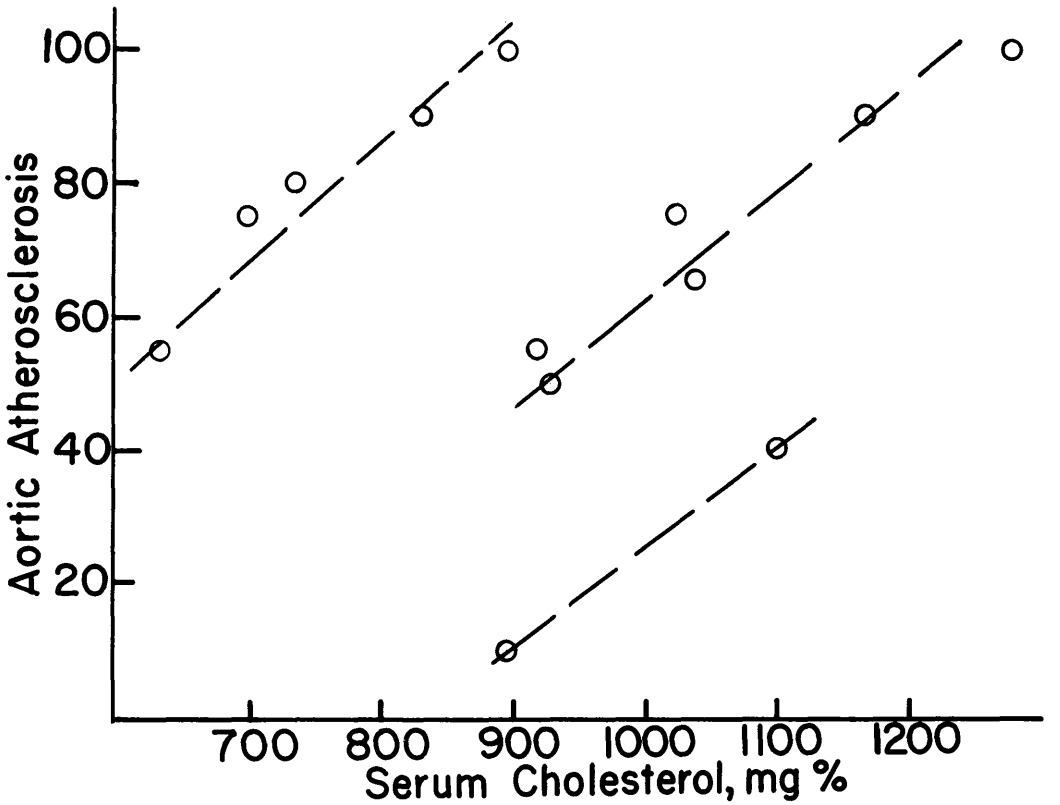


FIG. 1. Serum cholesterol level after 35 weeks on the diet *vs* degree of aortic atherosclerosis. The data suggest that the rabbits may represent 3 different populations of varying degrees of susceptibility.

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## Interaction of the Lathyrogen Beta-Aminopropionitrile (BAPN) with a Copper-Containing Amine Oxidase.* (31763)

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Lathyrogens appear to exert their effect on connective tissues by inhibiting the production of new covalent crosslinks in collagen and elastin(1,2,3). The excess soluble collagen which accumulates in the tissues of experimental animals after administration of lathyrogens seems to be intrinsically normal, but is deficient in aldehyde content(4,8). Substances which are thought to be the covalent interchain crosslinks of elastin and the intra-

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