

Vascular Pathology in Rabbits Following Administration of Foreign Protein.

ROBERT A. FOX AND LLOYD R. JONES.

From the Departments of Pathology and Bacteriology, St. Louis University School of Medicine, St. Louis.

During the past 9 months 3 lots of albino male and female rabbits (31 in all), weighing 1 to 3 kg each, were injected parenterally with horse serum. Each animal received one, 2, or 3 large doses (10 ml/kg) of serum administered either intravenously, intraperitoneally, or subcutaneously, and one or more smaller doses (1 ml), intravenously. Most of the animals were tested for skin hypersensitivity on one or more occasions. The total number of doses, the time interval between them, the sequence of large and small doses, and the time of skin testing were arbitrarily chosen, varied considerably, and seemed to bear no significant relationship to the observed results. For this reason a detailed record of each animal is deemed unnecessary. Those rabbits which did not die of anaphylaxis were sacrificed at arbitrary intervals following the last injection of serum. An autopsy was performed on all animals, and sections from a variety of organs were examined microscopically. Of the group of 31 rabbits, 20 exhibited pathologic changes of sufficient intensity to be considered significant. Seven showed changes which were considered to be only minimum in character and the remaining 4 were negative.

The significant histopathologic changes, which were limited almost exclusively to the heart, consisted primarily of mild vascular and perivascular alterations of the smaller branches of the coronary arteries. A few animals showed mild infiltration of leucocytes (mostly eosinophils) in the myocardium. Vascular and perivascular lesions were occasionally noted in the liver, lung, testis, kidney, and mesentery.

The vascular changes observed in the heart were characterized by intimal hyperplasia and proliferation of adventitial cells. The latter were frequently interspersed with, or sur-

rounded by, lymphocytes and mononuclear cells, with an occasional polymorphonuclear neutrophil. Fibrinoid degeneration of the intima and media was noted infrequently, and necrosis was not observed. The mononuclear cells varied in size from medium to large, and often were multinucleated. They frequently occupied a paravascular position, and, intermingled with lymphocytes, adventitial cells and a few polymorphonuclear neutrophils, vaguely resembled Aschoff bodies. The vascular lesions in which the cells were more circumferentially placed, resembled those often described as "rheumatic arteritis."

Klinge¹ and Vaubel² described similar vascular lesions in rabbit hearts following subcutaneous and intravenous injections of horse serum. Vaubel varied the size of the dose from small (0.005 ml) to large (30.0 ml), and observed alterations only in those animals which received 9 or more 2 to 10 ml injections. He was impressed by the similarity of the lesions to those of periarteritis nodosa, thromboangiitis obliterans, and to those noted in the necrotic portion of an Arthus skin reaction, and above all, to those of "rheumatic arteritis." He referred to the reaction as an "hyperergic arteritis." Klinge, on the other hand, was interested more in the pathologic changes in joints than in the changes which he observed in the heart muscle and valves. It was his opinion that the morphologic changes in the "blood vessel connective tissue apparatus" (perivascular tissue) and musculature (myocardium) closely resembled those observed in "human rheumatism" (rheumatic fever?). Recently Rich and Gregory observed similar lesions following intravenous injections of

¹ Klinge, F., *Beitr. z. Path. Anat. u. z. allegem. Path.*, 1929, **83**, 185.

² Vaubel, E., *Beitr. z. Path. Anat. u. z. allegem. Path.*, 1932, **89**, 374.

horse serum. The latter authors felt they had produced experimental periarteritis nodosa,³ and "lesions with the basic characteristics of rheumatic carditis."⁴

On the basis of the following observations: (1) general well-being of the animals throughout the course of the experiment, (2) vascular lesions mainly in the heart, (3) general preservation of vascular architecture, (4) absence of nodular lesions in arteries, (5) absence of vascular necrosis, and (6) paucity of polymorphonuclear neutrophiles, and mild character of the vascular reaction in general, the lesions which we observed were considered to be unlike those of periarteritis nodosa in man. On the other hand, they correspond rather to those reported by Vaubel¹ as "rheumatic arteritis" and by Rich and Gregory⁴ as "rheumatic carditis."

Certain immunological aspects of these experimental animals were studied in relation to the development of the vascular abnormalities. Of 23 rabbits, skin-tested during the course of the experimental work, all manifested positive reactions, as judged by inflammation, swelling, erythema, and induration at the site of injection. The more severe reactions in this group were characterized further by the development of grossly recognizable necrosis. This feature was observed in 13 of the animals, and these exhibited vascular alterations in the heart. This suggests that the degree of hypersensitivity requisite for the development of a necrotic type of skin reaction may be correlated in some manner with the capacity to develop vascular changes in the viscera. It is to be noted, however, that vascular reactions were also observed in 8 rabbits of the 10 which did not manifest the necrotic type of skin reaction. In this group the last skin test was made 8 to 17 days prior to necropsy, and while this interval does not differ materially from that in the previous group, it is recognized that the precise chronological relationship obtaining between marked skin hypersensitivity and the vascular allergic

response has not been investigated. Furthermore, the potentiality for the necrotic type of skin reaction, as well as the vascular alteration itself, may be transitory in character and easily missed.

Twelve (39%) of the 31 rabbits developed the serum sickness reaction⁵ on the ears, following a large parenteral dose of horse serum. A possible correlation between this reaction and pathologic changes in blood vessels is suggested, since all of the animals which developed the former were also found to have the latter.

Anaphylactic distress (mild or severe) was noted in 9 of 29 rabbits receiving two or more, spaced, parenteral injections of the antigen, and was not correlated with the development of morphologic vascular changes.

A correlation is known to exist between the capacity of the rabbit to develop the Arthus phenomenon upon intradermal administration of homologous antigen, and the humoral antibody level.⁶ In our experiments it was noted that the capacity to develop a necrotic type of skin reaction was related to the development of pathologic changes in blood vessels; therefore, it is probable that further investigation will reveal quantitative correspondence also between humoral antibody levels and vascular changes.

Some of our rabbits exhibited vascular abnormalities following a single large dose of foreign serum, as was also noted by Rich and Gregory.³ Added interest attaches to this observation because of its possible importance in relation to the clinical use of large doses of antisera and to the contemplated use of heterologous plasma, or its fractions, in the treatment of shock, etc., in man.

Summary. Mild vascular and perivascular alterations of the smaller branches of the coronary arteries were observed in a majority of rabbits following the parenteral injection of horse serum. No rabbit which elicited a necrotic-type of skin reaction failed to show vascular lesions.

³ Rich, A. R., and Gregory, J. E., *Bull. Johns Hop. Hosp.*, 1943, **72**, 65.

⁴ Rich, A. R., and Gregory, J. E., *Bull. Johns Hop. Hosp.*, 1943, **73**, 239.

⁵ Fleisher, M. S., and Jones, L., *J. Exp. Med.*, 1931, **54**, 597.

⁶ Cannon, P. R., and Marshall, C. E., *J. Immunol.*, 1941, **40**, 127.