and it is possible that this, plus other serum factors, might account for the persistence of "protoplasts" in the kidney.

Host-parasite relationships of "protoplasts" are not well defined. The availability of an experimental model of infection in which "protoplasts" occur during the natural history of the treated disease will now permit study of this important problem.

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27 December 1963

Induction of Papillary Growths in the Heart

Abstract. *Papillary growths appeared on the atrial endocardium of dogs at sites where adjacent surfaces presumably came into contact with a reciprocating motion. It is suggested that the resulting friction stimulates the growth of papillae which then act as pedunculated ballbearings, reducing the friction.*

While villi, papillary growths, and papillomas are ubiquitous structures in animals, such growths have been induced previously only by using chemical carcinogens or following specific viral infection (*1*). In our experiments circumscribed overgrowths of surface layers appeared in the course of studies designed to evaluate the role of physical forces in the differentiation of vascular and related tissues (*2, 3*).

In nine anesthetized thoracotomized dogs, the left atrial appendage was invaginated to form an epicardial sheath. A 4-ml Silastic (*4*) ball was placed into this sheath and held in place by a suture. This procedure introduced an endocardium-lined mass into the atrium. The animals were killed 10 to 202 days after surgery.

All the growths were found in the left atrium. At 10 days a marked fibroblastic proliferation was seen on the endocardial surface of the mass. In the other eight animals killed 31 to 202 days after surgery, round, slightly raised cauliflower-like growths about 5 mm in diameter were present on the endocardial surfaces of the intra-atrial mass (Fig. 1). Each of these growths was paired with one on the adjacent parietal atrial wall or on the mitral valve. Two or three pairs of such growths were found in the left atrium in each of the animals. The central portion of some of the growth on the atrial wall was saucerized, as if wear had taken place. Some of the masses were deep brown; others were lighter in color. The microscopic structure in both types was similar. The growths were richly vascularized, endothelium-covered excrescences consisting of numerous papillae, some of which were branched (see cover). The endothelial cells on the tips of the papillae were hyperplastic (Fig. 2), and contained vesicles which were periodic acid-Schiff positive and stained with alcian-blue dye. Occasional polymorphonuclear and mast cells, elastin, and small quantities of hemosiderin were present in the stroma of the stalks.

In previous studies at this laboratory, it has been suggested that specific mechanical forces induce definitive changes in the walls of blood vessels. As examples, collagen appears in tissues subjected to tensile forces, elastin at sites of a high rate of change of tension (*5*), cartilage in regions of high rates of change of compression (*3*), and increased cross-sectional areas of blood vessels at sites of increased hydrodynamic drag (*6*).

The motion of the heart probably produced a to-and-fro movement and sliding contact of the intra-atrial mass with adjacent atrial and valvular surfaces. The appearance of papillary growths at such sites suggests that frictional forces may contribute to the genesis of these growths.

A number of data appear to support this hypothesis. For example, the papillomatous proliferation of the intestinal mucosa in ulcerative colitis suggests that frictional action of apposing colonic surfaces may stimulate the formation

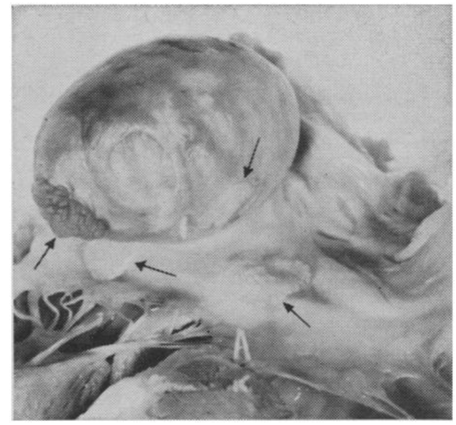

Fig. 1. Growths in the heart of a dog killed 7 months after surgery. The elliptical body is the endocardium lined intra-atrial mass. The mitral valve is at the lower left. Arrows point to the cauliflower-like growths, two on the intra-atrial mass, one on the parietal atrial wall (right), and one on a mitral leaflet.

of these growths. A ring of hypertrophic villi is usually present on the margin of peptic ulcers at which increased frictional forces may be operative. Papillomatous nodules occur on the vocal cords at sites which strike each other irregularly and haphazardly (*7*). Similar mechanisms during the inflammatory stage of rheumatic fever may contribute to the characteristic papillary proliferations on heart valves. Skin tags (*8*) (papilloma molle) are commonly found on the anterolateral surfaces of the neck and trunk at sites where collars and other clothing may produce chronic reciprocating frictional forces.

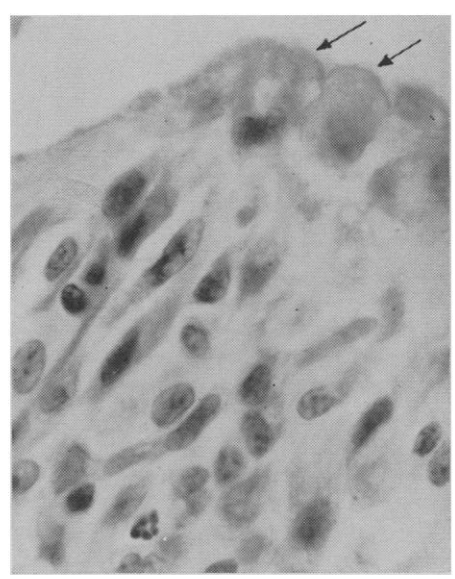

Fig. 2. Contact surface of a papilloma. Two large endothelial cells are situated above the fibroblastic growth (hematoxylin eosin, × 900).

The presence of alcian-blue staining intracytoplasmic vesicles at the contact surfaces of the papillary growths may be interpreted as a stage in the secretion of "lubricants."

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15 November 1963

Starch Formation Induced by a Plant Parasitic Nematode

Abstract. *This is the first report that a nematode causes plant tissues to produce starch. The formation of starch granules in the syncytial cells of several plant species is induced by* Nacobbus batatiformis. *The starch, which is associated with the feeding of the nematode, appears within a few days after the animal has become situated in the plant roots and diminishes in amount during nematode reproduction. The starch granules show the usual optical properties.*

Nacobbus batatiformis Thorne and Schuster (*1*), the Nebraska root-galling nematode, induces the formation of starch granules in plants. Such granules are not found in normal roots of certain plant species and varieties. Stimulation of starch formation in plant tissues infected by parasitic nematodes has not been reported previously. Owens and Novotny (*2*), who studied *Meloidogyne incognita* in tomato and cucumber roots, stated that "comparative chemical analyses show that . . . in the galls . . . starch disappeared." We found a decrease of starch in the vicinity of *Pratylenchus scribneri* in potato tubers and in the vicinity of *Heterodera trifolii* in roots of *Trifolium repens*. Starch formation was induced during the early stages of development of *Heterodera trifolii*. Starch was not detected in the walls of giant cells of soybean (*3*) and tomato (*4*) infected with *Meloidogyne* species. We have not found starch in giant cells of *Beta vulgaris* infected with *Meloidogyne incognita, Meloidogyne javanica, Meloidogyne hapla,* or *Heterodera schachtii*. Nor does *Pratylenchus penetrans* invoke obvious starch production in roots of wheat and *Chrysanthemum* species. DuCharme (*5*) noted a disappearance of starch around the lesions in grapefruit roots induced by *Radopholus similis*. Kostoff and Kendall (*6*) noted a starch accumulation in Nicotiana roots infected with root-knot nematodes but photographs showed no starch in giant cells or in their immediate vicinity.

The syncytium that arises within the gall adjacent to the anterior portion of *N. batatiformis* is more translucent in appearance than the other cells (*7*). It may reach 3 mm in length and contain hundreds of cells. Starch can be demonstrated within syncytial cells in thin sections (10 μ) of galls by staining (Fig. 1*A*) with Johansen's quadruple stain (*8*) and can be identified by the use of crossed polaroids (Fig. 1*B*). In general, starch grains are more numerous and larger near the anterior of the nematode and decrease in size and number away from the feeding area. Some syncytial cells are completely filled with starch granules, whereas others contain a few small or large granules, or some of various sizes, even when such cells are equidistant from the nematode's stylet. Because the starch granules are located only near the anterior, their formation is probably associated with feeding.

Starch granules are formed soon after the nematode infects a root and before the syncytium is formed. The granules first appear at the time that the nuclei and nucleoli enlarge and before hypertrophy of the cortical and epidermal cells. Initially, the granules are located around the nucleus and later are scattered throughout the cell. The high concentration of starch in the syncytium indicates that a considerable amount of nutrients are drawn to this area. *Nacobbus batatiformis* apparently utilizes the starch after transforming it into a soluble substance, since the amount of starch decreases during nematode reproduction.

The starch granules induced by *N. batatiformis* do not appear to differ in optical properties from those that occur naturally in other plant tissues. The granules have conspicuous concentric layering around the hilum when observed under oil immersion with bright light or polarized light. Birefringent crosses typical of starch are evident when the granules are viewed through crossed polaroids (Fig. 1*B*). In the root galls of sugar beets, spinach, and common purslane, the granules, which are usually spherical, range from 1 to 30 μ in diameter; most of them measure from 5 to 10 μ, and the average diameter is 11 μ.

In roots grown in organ culture, males as well as females of *N. batatiformis* are capable of inducing the formation of galls and their associated

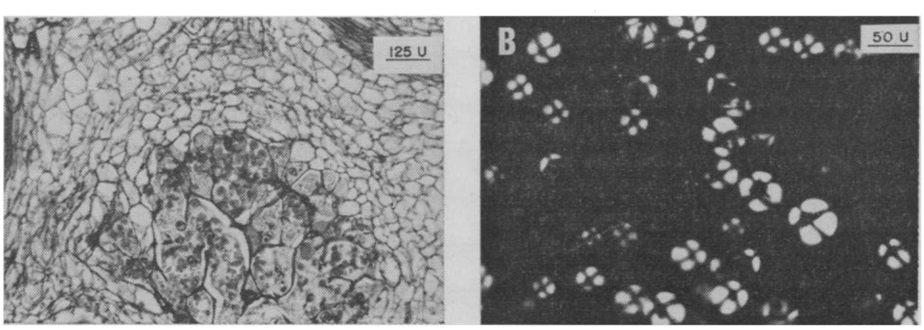

Fig. 1. Portions of a syncytium from *Beta vulgaris* showing the presence of starch granules induced by *N. batatiformus*. *A*, Stained with Johansen's quadruple stain and photographed through bright light (part of female nematode is in lower left-hand corner); *B*, photographed through crossed polaroids.