Pancreatic Ectasia in Uremic Macaques

RODERICK T. BRONSON, DVM, WILLIAM STRAUSS, BS, and WENDY WHEELER, BS

From Harvard Medical School, New England Regional Primate Research Center, Southborough, Massachusetts

Pancreatic ectasia (PE) is a common incidental finding in people dying from uremia. It has been described as dilatation of acini, inspissation of secretions, and proliferation of ductal cells. PE occurred in 17 macaques, 11 of which were known to have been uremic. The lesion was studied by light and electron microscopy and histochemistry and by construction of a three-dimensional model of a dilated acinar ductal system from serial semithick Epon sections. Atrophic acinar cells interspersed with clumps of centroacinar cells lined all portions of the system. There was no evidence of ductular proliferation. Fibrillar material was present in the dilated acinar lumens and associated with epithelial cells and leukocytes, but no blockage of the system was demonstrated. The lesion is similar to those induced by a variety of experimental procedures and to the pancreatic lesions of cystic fibrosis. (Am J Pathol 1982, 106: 342–347)

PANCREATIC ECTASIA (PE) in man is characterized by focal or diffuse dilatation of acinar and ductular lumens, inspissation of secretions, and mild acute inflammation of the interstitium.[1-7] This lesion has been observed at autopsy in patients dying from uremia,[3] typhus,[3] carcinoma with cachexia,[4] ulcerative colitis,[6,7] and a variety of other diseases.[1,4] Lesions that are similar in some respects were observed in experimental pancreatic duct occlusion,[8-11] certain experimental intoxications,[12-14] and cystic fibrosis.[15-19] The pathogenesis and clinical significance of PE have not been established, since it is essentially a postmortem diagnosis. Such mechanisms as dehydration[3,4] and vagal stimulation leading to ductal cell proliferation[5] have been suggested as important factors in its pathogenesis. Possible long-term sequelae are fibrosis or lipomatosis.[4,5]

PE is known to occur in obese macaques (*Macaca*) dying from a disease called fatal fasting syndrome.[20] It is characterized by weight loss and fatty change of liver and proximal convoluted renal tubular epithelium. Weight loss is secondary to anorexia, which may be induced by other diseases, by wounds from fighting, or simply by being recaged with unfamiliar monkeys. About one third of the cases with renal fatty change die with uremia, perhaps due to degeneration of lipid laden proximal renal epithelial cells. Nearly all uremic monkeys with the syndrome have PE; a few have focal pancreatic necrosis. Nearly all in which pancreatic disease is found have generalized fat necrosis.[20]

This is a report of the histopathology, three-dimensional histology, and ultrastructure of PE in monkeys. The lesion will be compared with various other pancreatic lesions, including cystic fibrosis of the pancreas, which has been reported once in a single rhesus monkey.[21]

Materials and Methods

Ectatic pancreases from 14 mature female rhesus monkeys (*M mulatta*) and three mature crab-eater macaques (*M fascicularis*) were studied. The cases were assembled during retrospective, experimental, and clinical studies of fatal fasting syndrome[20] with which all animals died after 2 to 4 weeks illness. Five cases had no significant lesion apart from weight loss, fatty changes, PE, and fat necrosis. Five had skin lacerations and bruises but no internal injuries; seven had a variety of other lesions—colitis, generalized Herpes B infection, suppurative gingivitis, pyothorax, endometriosis, early bronchopneumonia, and uterine necrosis. The mean blood urea nitrogen (BUN) in 10 cases, measured within 2 days of death,

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Address reprint requests to Roderick T. Bronson, DVM, Tufts Veterinary Diagnostic Laboratory, State Laboratory Institute, 305 South Street, Jamaica Plain, MA 02130.

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was 215 ± 80 mg/dl. The BUN of one monkey was 27 mg/dl, but creatinine was 12.1 mg/dl. Serum chemistry screens of five cases revealed no other outstanding abnormalities apart from elevated BUN, creatinine, and phosphorus. No clinical studies were performed in the 6 other monkeys with ectasia. Two other monkeys in the earlier study[20] were uremic but had no ectasia.

Tissues removed from most of these monkeys at death or sacrifice were fixed by immersion in 10% buffered formalin. Paraffin sections from all cases were stained with hematoxylin and eosin (H&E). A few sections were also stained with periodic acid–Schiff (PAS), alcian blue, and mucicarmine stains. Sampling of pancreases varied in extent. In 12 cases a single portion of pancreas was available for study. It usually included a cross-section of the organ at the approximate level of the bile duct and included the duodenum. In 5 cases three to five pieces of pancreas randomly taken from unspecified portions of the organ were available.

A portion of pancreas from one monkey with diffuse ectasia was diced and fixed immediately after death in 3% glutaraldehyde in 0.1 M phosphate buffer. The tissues were postfixed in 1% osmium tetroxide, stained *en bloc* with 2.0% uranyl acetate, and embedded in epoxy resin. One block was trimmed for serial semithick sectioning with glass knives. Sections were stained with toluidine blue. Each section was photographed at $\times$ *105; the prints were magnified to a total magnification of* $\times$656. The mean thickness of sections, 1.77 μ, was estimated by dividing the mean diameter of 36 pancreatic acinar nuclei by the mean number of serial sections required to section through 13 nuclei.

A three-dimensional model of one acinar system was constructed of dental wax. A lumen was selected in the first photomicrograph, and its outline was traced onto tracing paper. The tracing was cut out and superimposed over dental wax of approximately the thickness of a section as magnified. The wax outline was cut out. The same lumen appearing in the next section was similarly traced in wax, positioned over and melted to the first wax outline after checking the orientation relative to other lumens. The process was repeated until a wax model equivalent to 103 μ in height of original tissue was constructed.

Two stained semithick sections, previously mounted on glass and photographed for construction of the model, were selected for ultrastructural study. Trimmed Epon block faces were glued directly on the glass-mounted semithick sections with cyanoacrylate ester glue. The block was wedged off the slide with a razor blade and was thin-sectioned. Thin sections were stained with 0.1% lead citrate.

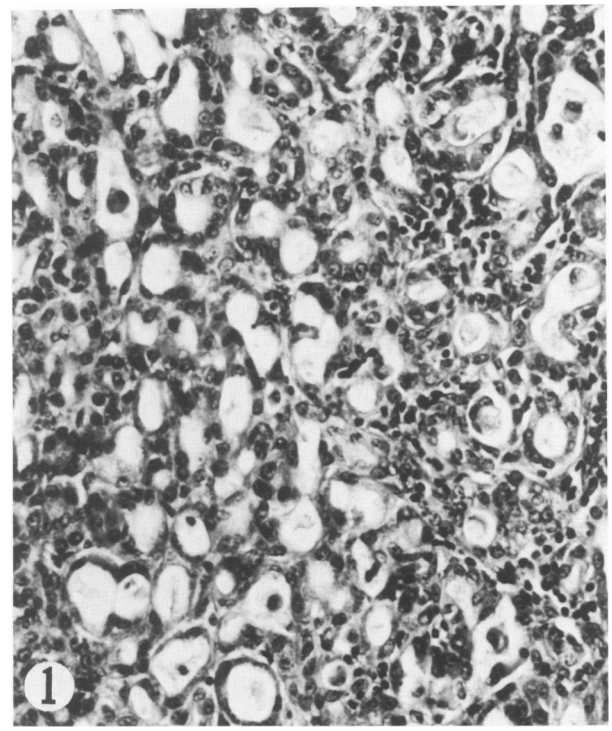

Figure 1—Most acini are dilated; a few contain leukocytes. The acinar cells are atrophic. There is mild interstitial inflammation. (H&E, ×216)

Results

In the H&E sections of some pancreases whole lobules or portions of lobules with ectasia were scattered among normal lobules. In other pancreases all lobules were affected. The abnormal ones were composed of dilated acini usually lined by cells with pale cytoplasm and little or no zymogen. Some dilated lumens contained a floccular eosinophilic material and occasional neutrophils and macrophages (Figure 1). Often there was mild to moderate acute inflammation and/or fibrosis in the intralobular and perilobular connective tissue. Some lobules, particularly in two cases of longer standing uremia, had more severe fibrosis and only poorly formed acini (Figure 2). The interlobular ducts in all cases were undilated and had normal epithelium. A few contained pink material. The islets were never affected. One pancreas had lipomatosis in addition to ectasia.

In toluidine blue stained Epon sections the nature of the epithelial cells lining ectatic acini was more evident. Most had dark-staining cytoplasm with a few or many dark blue granules, interpreted as zymogen

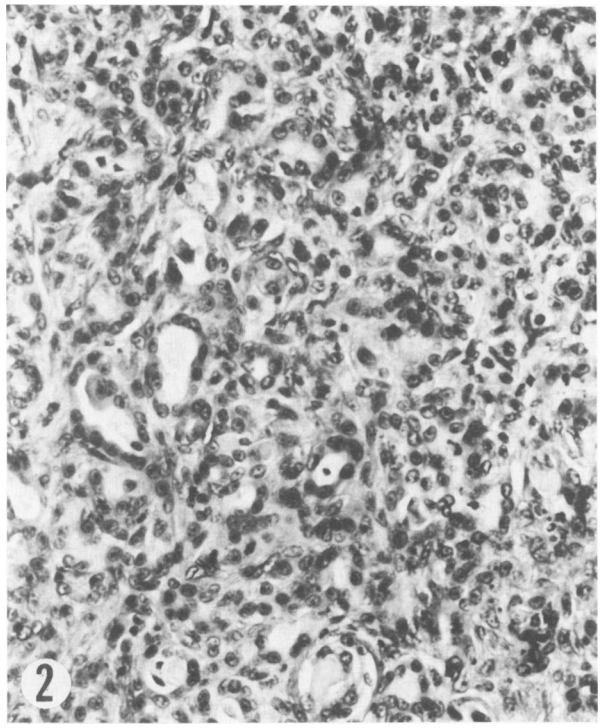

Figure 2—Pancreas from the monkey in which uremia developed after experimental regrouping.[20] Some acini are ectatic. Much parenchyma is composed of cells with pale cytoplasm and little or no apparent zymogen arranged in indistinct acini. The number of fibroblasts and the amount of collagen in the interstitium are increased. (H&E, ×216)

granules, in the apical cytoplasm. These cells, believed to be atrophic acinar cells, contrasted to pale-staining cells without granules believed to be centroacinar cells. The material in the lumens stained faintly with toluidine blue. It was usually clumped and did not fill the entire lumen.

Construction and study of the three-dimensional model showed that the ectatic pancreatic lobule was composed of branching tubules with dilated outpouchings lined by atrophic acinar cells (Figure 3). Centroacinar cells were clustered in small patches, usually at points where lumens branched. No lumen contiguous with those depicted in the model appeared to exit from it except at the bottom. This implied that the model represented the distal end of the ramifications of a ductular system. Intraluminal material and occasional inflammatory cells were present throughout the system, which was not blocked at any point.

By electron microscopy the distinction between the two cell types lining the lumen was clear. The acinar cells had relatively large amounts of rough endoplasmic reticulum, zymogen granules, and autophagic vacuoles (Figure 4). The centroacinar cells were electron-lucent and contained few cytoplasmic organelles. Sometimes they had a few uniformly sized small dark granules in the cytoplasm. The nuclei of centroacinar cells had less clumped chromatin than those of acinar cells. Some of the material in the lumens consisted of loosely arranged filamentous material (Figure 5). This was also present in cytoplasm of macrophages (Figure 6), acinar cells, and centroacinar cells (Figure 7).

The histochemical study showed that the intraluminal material was faintly to moderately PAS-positive, occasionally alcian-blue–positive at pH 2.5, and mucicarmine-negative.

Discussion

By light microscopy pancreatic ectasia of macaques is similar to that of man.[1-7] This study clarifies some points raised by earlier investigators, who had only paraffin-embedded tissue to study. It has been argued that pancreatic ectasia involves proliferation of ductular epithelial cells.[1-7] To the contrary, this study shows that most cells lining ectatic acini are acinar cells[22-25] that have undergone atrophy. Pancreatic ectasia, then, is principally acinar ectasia, and no other structural derangements are present, at least in early lesions. Later, acute inflammation and fibrosis may supervene, causing greater destruction of lobular architecture.

The ectatic changes in acini facilitated a three-di-

Figure 3—Wax model of the lumen of an ectatic acinar system. The black patches signify those areas of the lumen lined by centroacinar cells. Pale patches throughout the system in both the blind pouches and the tubules are lined by acinar cells. As depicted here, the secretions drained toward the bottom of the system; no open lumen was contiguous with the system except at the bottom. (×788)

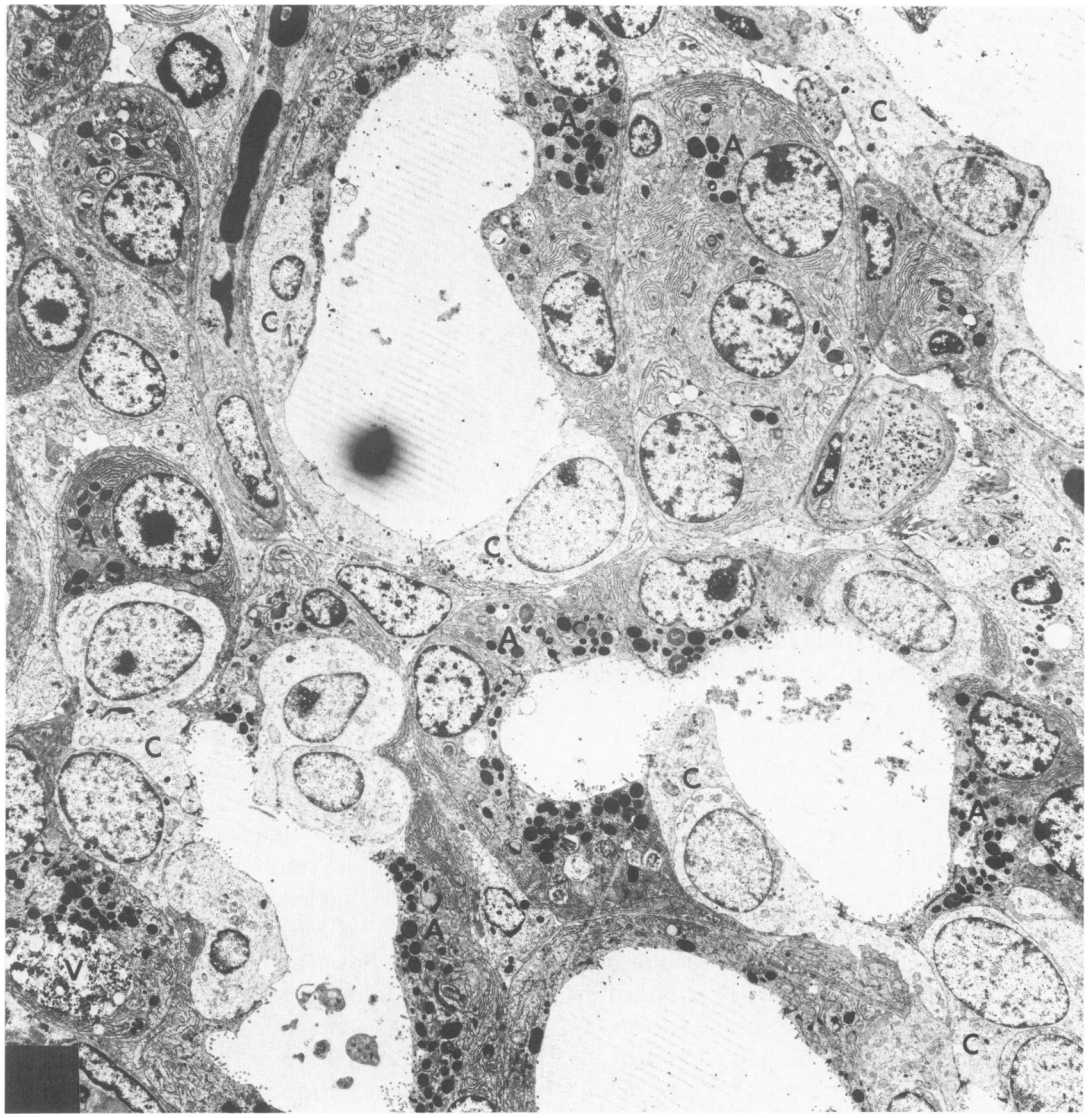

Figure 4 – Thin section of ectatic pancreas. Centroacinar (*C*) and acinar (*A*) cells are clearly differentiated. Acinar cells have endoplasmic reticulum, a few zymogen granules, and occasional autophagic vacuoles (*V*). Centroacinar cells have few organelles but sometimes have small dark cytoplasmic granules. (× 2000)

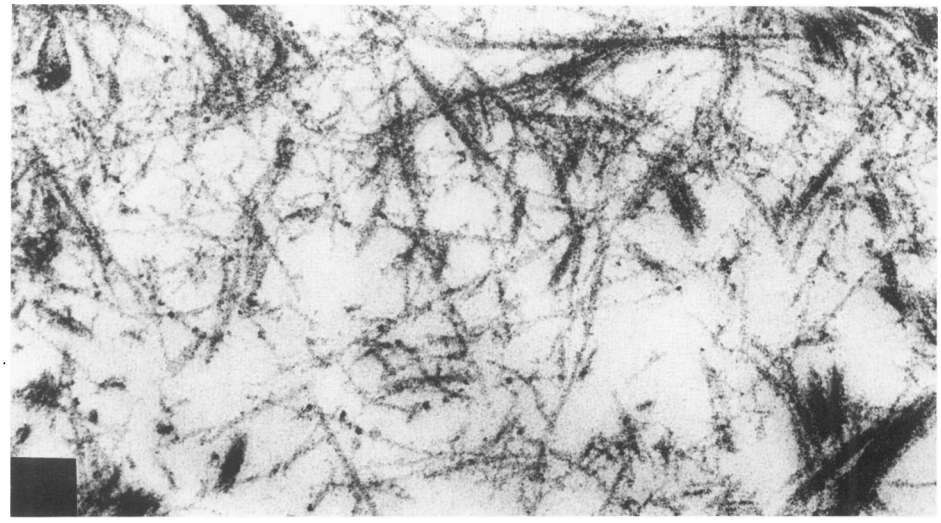

Figure 5 – Loosely arranged fibrillar material present in some lumens. (× 100,000)

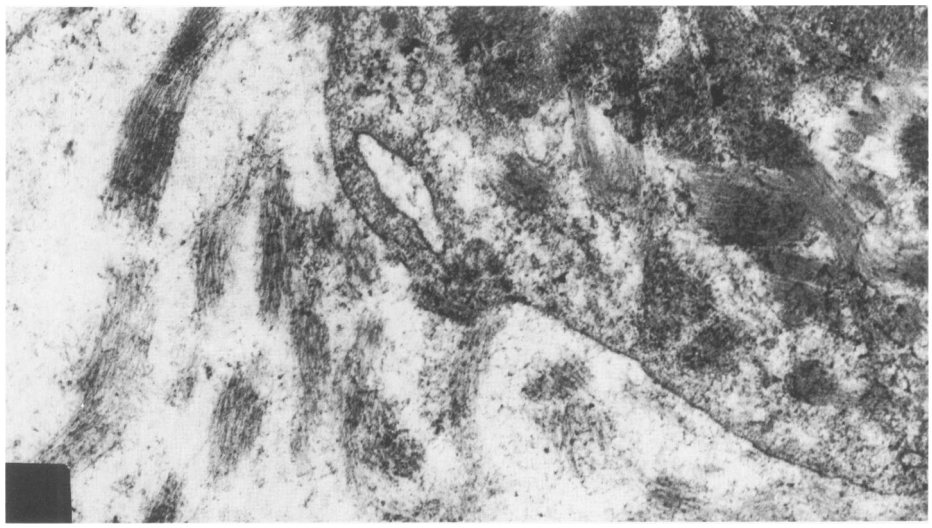

Figure 6—Sheaves of fibrillar material within and around a macrophage. (×40,000)

mensional study of lobular architecture, since the lumens could be readily traced from section to section. Such a study would have been much more difficult to perform on normal pancreases. Indeed, only two other similar studies have been attempted. One involved three-dimensional reconstruction of the ductular system of a human pancreas with polycystic disease.[26] The acinar architecture was not studied. The other was a study of the normal exocrine pancreas of the rat, which showed that in that species the pancreas is not arranged in the grape cluster fashion implied by the term "acinar."[27] Rather it is arranged in anastomosing cords. The macaque pancreas, too, is arranged as a complex of tubules lined predominantly by acinar cells. But blind pouches equivalent to classic "acini" are present, and anastomosis is not a prominent architectural feature. Interestingly, too, centroacinar cells are not located only in contiguity with ductular cells, as expected,[22-24] but are arranged as small islands of cells surrounded by acinar cells.

This study sheds light on the pathogenesis of PE. It is clear that a secretory material fills or partially fills the dilated lumens. In the small part of the pancreas examined, no discrete point of blockage of intraluminal flow of secretory material was observed. The lobular distribution of ectasia in many cases suggests that the point of blockage might be at the level of the interlobular duct. Indeed, experimental pancreatic duct ligation does result in ectasia,[8-11] and acinar cells develop autophagic vacuoles[10] similar to those observed here. No evidence of such blockage was observed here, however.

It seems more likely that ectasia results from an abnormality of pancreatic secretions which become inspissated throughout the ductular system. It has been argued that dehydration and inadequate nutrition are causative in some unknown way, since they are commonly associated with the diseases in which ectasia occurs.[1-7] It is reasonable to assume that pancreatic secretions devoid of a large serous component because of dehydration might inspissate.

The fibrillar material observed ultrastructurally in

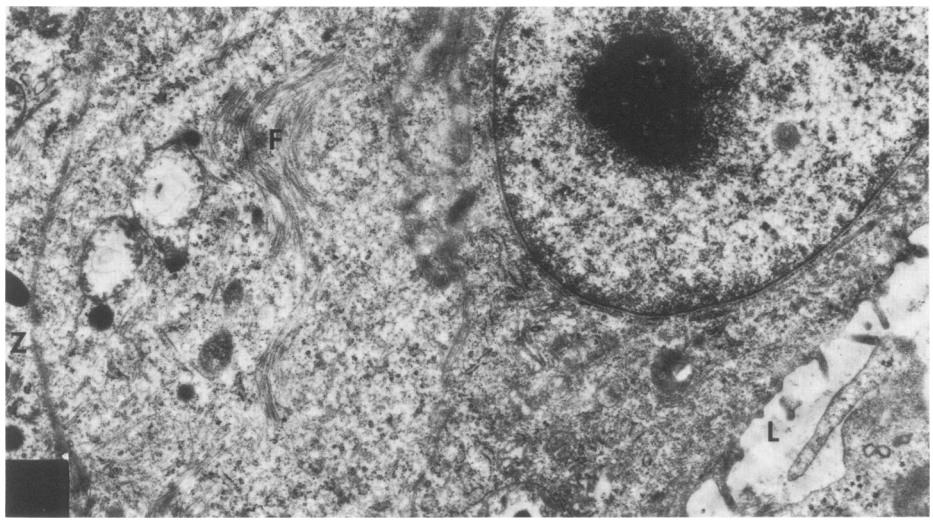

Figure 7—Fibrillar material in the cytoplasm of an acinar cell. A portion of the lumen (*L*) is to the right. The acinar cell to the left has zymogen granules (*Z*). Mitochondria (*M*) are artifactually exploded. (×12,000)

the dilated lumens, macrophages, and acinar and centroacinar cells has been observed in a variety of experimental pancreatic diseases. These have included intoxications affecting the pancreas[13,14] and experimental duct occlusion.[8] It has been argued that the material may account for ectasia observed in various pancreatic diseases, including cystic fibrosis.[14]

Histochemical studies of the intraluminal material in pancreatic ectasia were reported in earlier studies.[5,28] In man as in the monkeys, stains for acid mucopolysaccharide stained the material only weakly in some but not all lumens. The material in PE has been compared with that of cystic fibrosis,[28] in which a variable increase in the amount of acid mucopolysaccharide has been reported.[16,19,29] The inspissated secretion in PE and cystic fibrosis may thus be similar in some histochemical aspects,[28] but important histologic differences should not be overlooked. It is characteristic of cystic fibrosis and not of PE that the intraluminal material forms concentric and crystalline concretions.[16-19] Perhaps the secretions in PE would be similar if the disease progressed for as long a time as cystic fibrosis.

The association of PE with mild to moderate acute pancreatic inflammation and fat necrosis is important. Since inflammation is not a constant finding, it can be assumed to occur in parallel with or secondary to PE. Whether the presumed long-term sequelae to PE, fibrosis or lipomatosis,[4,5] only occur when a pancreas with PE becomes inflammed is unknown. In any case, PE should not be construed as an entirely benign incidental postmorten finding.

The similarities between what may have been cystic fibrosis in a young rhesus monkey[21] and of PE of macaques and between the two diseases of man suggests that future studies of PE in rhesus monkeys might be productive in the understanding of cystic fibrosis. Moreover, the association of PE with uremia suggests ways of reproducing the disease experimentally.

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