the synthesis or assembly of the molecules of compact myelin. Blakemore et al. reported that protein and lipid analyses of myelin from affected white substance were not abnormal except for some lowering of total protein content. Internodal myelin and much of that surrounding the paranodes appears normal. Some circumferential thinning of myelin sheaths has been described, but no lesions of the inner or outer tongues were reported. The longest focal lesions are well short of the length of any internode thus emphasizing their paranodal character.

Although Blakemore et al. reported some specialized contact points between processes and axon, it may be that the profuse process proliferation interferes with the establishment of the normal array of axoglial tight junctions. This may breach the integrity of the axoglial space and cause conduction abnormalities. Fluid distending interlamellar and axodinal spaces may account for the marginal vacuolation seen in many foci. Some eventual disruption of formed compact myelin in the affected paranodes could explain the luxol fast blue-positive granules seen in many foci.

The large cells, called hypertrophic oligodendrocytes by Blakemore et al. resemble the immature oligodendrocytes of active myelination. Perhaps an inability to switch off the proliferation of fine processes explains the continuing cytologic picture of high metabolic activity shown by these cells.

The shape and dimensions of the focal lesions are related to the three dimensional anatomy of white substance. Peters et al. have observed that nodes of Ranvier frequently appear in groups in transverse sections and propose that each group reflects sheaths formed by the ultimate processes of one of the several major processes of an oligodendrocyte. Each focal lesion is thus a clump of affected paranodes. There are no large confluent lesions; each group of abnormal paranodes remains discrete.

The relatively late onset and progressive nature of the disease may reflect the slow persistent proliferation of fine processes with consequent failure to adequately ensheath the growing axon and to establish normal axoglial tight junctions.

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Multihormonal Islet Cell Carcinoma in a Greater Bushbaby (Galago crassicaudatus argentatus)


Few tumors involving the pancreas of nonhuman primates have been described. We report an islet cell tumor in a prosimian producing multiple polypeptide hormones.

An adult (11 years old) female Greater Bushbaby had been maintained in the Vanderbilt University Animal Facility for 9 months prior to her death. The animal had been born at the Oregon Regional Primate Center and was part of a breeding colony. During that time she had not been subjected to any experimental manipulations and had become pregnant twice, delivering one liveborn and one stillborn infant at 19 months of age and 7 years of age. No experimental studies had been done while at Vanderbilt. Other than a cataract of the right eye, the animal appeared normal prior to finding it dead in its cage. The only lesion noted at necropsy involved a 2.5 cm nodule in the cranial lobe of the pancreas. The mass was well encapsulated and smooth. Cut surface revealed partial lobulation with the color and consistency resembling that of a normal pancreas.

Pancreas was fixed in neutral buffered 10% formalin and paraffin-embedded; 5 μm sections were stained with hematoxylin and eosin (HE) or placed on gelatin coated slides which were heated at 56 C for 30 minutes. After deparaffinization in two changes of xylene at 20 minutes each, sections were cleared in three changes of absolute ethanol for 5 minutes each followed by an endogenous peroxidase quench in 0.3% H2O2 methanol for 30 minutes. Sections were then hydrated through graded ethanol and placed in phosphate buffered saline (pH 7.2). Control sections included nontumor area of pancreas from the involved animal, as well as normal human pancreas for the demonstration of insulin, glucagon, somatostatin, and bovine pancreatic polypeptide. Human adrenal medulla was used as a control for neuron specific

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enolase, pyloric antrum of stomach for gastrin, human islet cell tumor for substance P, and human medullary carcinoma for calcitonin. Tissue sections were then incubated for 10 minutes in 10% normal swine serum. (Normal swine serum, normal rabbit serum, swine anti-rabbit, and rabbit peroxidase anti-peroxidase from DAKO Corporation, 22 North Milpas Street, Santa Barbara, CA 93103.) Excess serum was shaken off. Optimal dilutions of primary antisera (rabbit anti-glucagon [1:500], rabbit anti-somatostatin [1:100], guinea pig anti-insulin [1:1,000], rabbit anti-gastrin [1:2,000] and rabbit anti-neuron specific enolase [1:1,600] from DAKO Corporation, Santa Barbara, CA; rabbit anti-bovine pancreatic polypeptide [1:500] from Lilly; rabbit anti-calcitonin [1:1,000], rabbit anti-VIP [1:1,000], and rabbit anti-substance P [1:800] from Immuno Nuclear Corporation, Stillwater, MN) were applied and incubated overnight at room temperature in a humid chamber. Substitution of normal rabbit serum for the primary antisera served as a negative control. Slides were rinsed with phosphate buffered saline and washed in three changes of phosphate buffered saline for 8 minutes each. Swine anti-rabbit diluted 1:20 was applied for 30 minutes followed by rinsing as described above. Rabbit peroxidase anti-peroxidase diluted 1:50 was applied for 30 minutes.

Fig. 1. Tumor composed of sheets of neoplastic cells subdivided by a fine fibrovascular stroma. Scattered mitotic figures are present (HE).

Fig. 2. Aggregates and individual neoplastic cells staining intensely for insulin and sometimes obscuring nuclear detail.

Slides were placed in Tris-HCl buffer (pH 7.6) for 10 minutes. Diaminobenzidine substrate (5 mg) (DAB, Sigma D9015) and 10 microliters of 5% H$_2$O$_2$ in 10 ml of Tris-HCl buffer, was applied for 10 minutes, followed by 5 minute washes in phosphate buffered saline and distilled water. After a hematoxylin counterstain, slides were dehydrated through graded ethanol and xylene. Cover slips were mounted with synthetic mounting media.

The pancreatic nodule seen grossly was composed of sheets of cells having round to oval, basophilic stippled nuclei often with indistinct nuclei (Fig. 1). Cells frequently had moderate amounts of lightly staining granular cytoplasm although some cells had a somewhat vacuolated cytoplasm. Rows of neoplastic cells were occasionally separated by narrow spaces to form vascular-like channels. A moderate collagenous stroma forming partial lobules gave rise to a delicate capillary network which was infrequently surrounded by palisading neoplastic cells. Mitotic figures were numerous and sometimes bizarre. Although condensed stroma encapsulated much of the tumor, capsular invasion by neoplastic cells was present in several areas. Lymphocytes were well dispersed throughout the capsule. Only equivocal vascular in-
vasion by neoplastic cells was evident. Based on gross and histological findings, the tumor was classified as an islet cell carcinoma.

Additional histologic findings were confined to the liver and kidney. Hepatic portal triads often contained a mixed inflammatory infiltrate consisting of lymphocytes, plasma cells, and hemosiderin-laden macrophages; occasional neutrophils were noted as well. Large numbers of lymphocytes were located in interstitium and fat adjacent to the renal pelvis. A few small aggregates of lymphocytes and plasma cells were evident in renal cortical interstitium and were frequently periglomerular in distribution.

Immunohistochemical staining of tumor and adjacent normal pancreatic islets revealed a positive reaction of antibody to insulin, glucagon, somatostatin, neuron specific enolase, and pancreatic polypeptide. Tumor and normal pancreatic islets were negative to calcitonin, gastrin, substance P, and vasoactive intestinal polypeptide. Specifically, reactivity of normal islets adjacent to the tumor occurred in approximately 75% of islet cells for insulin, 15–20% for glucagon, 50% for neuron specific enolase, 0–5% for somatostatin and 5–20% of islet cells for pancreatic polypeptide. The tumor contained several large aggregates of tumor cells which were positive for insulin (Fig. 2). Positive reaction consisted of a brown granular cytoplasmic stain in affected cells. There were numbers of scattered glucagon-positive tumor cells as well as reactivity in the area of fibrous stroma which contained duct-like structures. Neuron specific enolase was present in scattered tumor cells with a lighter, less granular cytoplasmic appearance. Scattered tumor cells were also positive for pancreatic peptide.

Pancreatic islet cell tumors can release both eutopic and ectopic polypeptides. Eutopic refers to humoral substances native to the islets while ectopic refers to islet cell tumours elaborating polypeptides not normally native to the adult pancreas. Eutopic release includes four polypeptides; glucagon from the alpha cells, insulin from the beta cells, somatostatin from the delta cells, human pancreatic polypeptide from the F (D), or PP) cells, and the amine serotonin from the enterochromaffin cells. Ectopic secreting polypeptides of pancreatic tumors include peptides of the gastrin family (gastrin, cholecystokinin), peptides of the secretin family (secretin, vasoactive intestinal peptide), substance P, neurotensin, leu-enkephalin, alpha and beta endorphin, motilin, calcitonin, ACTH, parathyroid hormone and vasopressin and growth hormone releasing factor. The clinical recognition, pathology, and treatment of these various tumors in humans has been reviewed.

Regardless of the underlying cause of multihormonal tumors, it is now possible to identify the potentially functional nature of the tumor by quantitative assays of polypeptide content and to determine the cellular nature by immunoperoxidase and immunofluorescence techniques.

References

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