FLUORESCENCE HISTOCHEMICAL STUDY OF THE PANCREAS IN THE CAT

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Summary

The exocrine and endocrine pancreas was investigated according to the fluorescence histochemical method of Falck and Hillarp. 1) Green fluorescent adrenergic fibers were regularly seen associated with arteries and arterioles in the exocrine pancreas. 2) Cholinergic fibers as shown by cholinesterase activity, were also found in the parenchyma of pancreas. 3) Yellow fluorescent cells scattered in the exocrine pancreas and localized to a population of pancreatic islet cells with a characteristic distribution at the islet periphery were found. 4) By the fluorescence microscopic observation, inter-or intralobular pancreatic ducts, involving the zymogen granules, can also be seen after treatment with HCL vapor. 5) Yellow fluorescent cells, ß-cells containing insulin, remained at the islet periphery. At present, the above mentioned yellow fluorescent cells are identified as containing HPP (Human pancreatic polypeptide) according to the immunofluorescence technique.

With the use of the Falck and Hillarp histochemical technique ethionine induced pancreatitis in cats has been investigated. 1) After seven days of ethionine (5 mg/kg BW oral ad.) treatment, pancreas showed histochemical changes such as hemorrhage, fat necrosis, destruction of acinar cells and degranulation of zymogen from the parenchyma of pancreas. 2) Oral administration of ethionine for ten days induced severe degranulation, rupture of vessels, especially of veins and venules and later influenced arteries or arterioles. 3) Necrosis and fibrosis began to appear in the spaces between the cellular debris and marked pancreatic atrophy could be found. 4) The destruction of Islets of Langerhans can be found in the ethionine induced pancreatic parenchyma. On the other hand, an increased number of Islets of Langerhans was also observed in the site of lobule. 5) The presented finding may also suggest that the duration of administration of ethionine is more important factor than graded doses of ethionine in the production of ethionine induced pancreatitis in cats.

Key Words: Inter-or Intralobular pancreatic ducts, HPP. (Human pancreatic polypeptide), Ethionine induced pancreatitis, Zymogen granules in the ruptured blood vessels, Degranulation and necrosis of acinar cells

Introduction

Pancreatic surgery today is a vast field for the treatment of pancreatic diseases such as acute hemorrhagic pancreatitis, chronic pancreatitis and cancer of pancreas. Although pancreatitis has been extensively studied, still many problems concerning this disease remain unsolved. This is mainly due to the fact that pancreatitis rather seldom exists as a distinct entity but is often superimposed on and overshadowed by other significant disorders such as gallbladder disease, diabetes mellitus,
fibrocystic disease of pancreas and pancreatic tumor, benign and malignant. A profound knowledge of all pancreatic diseases is, however, necessary since almost all these above mentioned disorders are difficult to diagnose because of the hidden position and large reserve function of the pancreas and because they appear in a wide variety of clinical forms ranging from a catastrophic "acute abdomen" to a silent growth or advance of a malignancy. In addition, the pancreas has many functions and furthermore consists of two anatomically and functionally separate units, i.e. an exocrine portion consisting of the pancreatic acinar cells and ductal system, involving the major ducts of Wirsung and Santorini and also another unit, the endocrine portion consisting of the islets of Langerhans. The islets are built up by different types of cells which produce at least two hormones, insulin from the β-cells and glucagon from the α-cells.

The functional and physiological significance of these are discussed elsewhere. Of the above mentioned details, the purpose of the present study is to identify or to define more clearly the changes in the function of the islet cells, and the acinar cells respectively, and of the ductal system in the pathogenesis of the clinical manifestation of acute pancreatitis in human through experiences gained from the experimental pancreatitis induced by ethionine in animal. Moreover, it is also the purpose to assist the clinical diagnosis of the pancreatic disorders in human by a basic study of the pancreatic adrenergic and cholinergic innervation, the type of endocrine monoamines, and the exocrine and endocrine functions according to the fluorescence histochemical methods of Falck and Hillarp 1,2 (1961) (Flack and Torp 3 1961; Falck 1962; Falck et al. 1962; Corrodi 4 and Hillarp 1963, 1964).

Part I

"Fluorescence Histochemical Study of the Pancreas, Especially, Innervation, the Exocrine and Endocrine in the Cat"

Basal Experiments:

Material and Methods.

Thirty healthy adult cats were used for the fluorescence histochemical experiments. Food and water was freely available up to the time of sacrifice. Each cat was anesthetized by intraperitoneal injection of sodium pentobarbital in a dose of 30 mg/kg BW. Specimens from various regions (head, body and tail) of the pancreas were dissected. These small pieces of the pancreas were immediately frozen to the temperature of liquid nitrogen in a mixture of propane and propylene or acetone-dry ice solution.

After freeze drying, the specimens were treated with formaldehyde gas according to the procedure of Falck and Hillarp, embedded in paraffin wax and sectioned at 6–10 μ thickness. Also, tissue specimens were taken from non-treated cats (as control) in the way described above, and from L-DOPA and/or L-5HTP pre-treated cats who were sacrificed 30–60 min after intraperitoneal injection of the drug.

The specimens taken were then processed (see Falck and Owman 5 1965) for fluorescence microscopic demonstration of certain aryethylamines according to the formaldehyde condensation technique of Falck and Hillarp, i.e., the sections were mounted in Entellan (Merck) or xylene and examined in a fluorescence microscope.

The sections or examined sections were then exposed for 1–15 min to HCL-vapour generated from 2 ml concentrate acid in a closed vessel (about 70–80 ml vol.) and re-examined for acinar cells and fluorescent cells. The borohydride reaction 6, 7 was applied to formaldehyde-treated sections before and after