EFFECT OF LONG TERM ALCOHOL FEEDING ON THE PANCREAS IN RAT

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Summary

To elucidate the pathophysiological process of alcoholic pancreatitis, chronic alcohol intoxication was made in Wistar rats on balanced diet giving 20% ethanol freely for 60 weeks. The control rats received water. Histological picture of the pancreas, hormonal activity in the mucosa of upper digestive tract and the nature of pancreatic juice were examined in every 15th week. The results were as follows. 1) No histological changes were noted in the pancreas of control group. In the ethanol group, morphological abnormalities of the pancreas appeared after 30 weeks. Of the histological findings, the changes on the ductal system such as dilatation of pancreatic duct, plug formation in the ductal lumen and periductal fibrosis were significant. 2) The long term ethanol administration tended to decrease the amounts of gastrin, secretin and cholecystokinin contained in the gastrointestinal mucosa. 3) Regardless of the histological changes of the pancreas, almost no changes were noted in the bicarbonate and protein concentration during the experimental period of 60 weeks.

From the above results, a mechanism obstructing pancreatic ductal system is considered to be important in the pancreatic lesions by alcohol rather than a mechanism of stimulating pancreatic exocrine secretion.

Key Words: chronic alcohol intoxication, experimental pancreatitis, gastrointestinal hormones, pancreatic juice, plug formation.

Introduction

The long standing alcoholic abuse causes pancreatitis. This is a well recognized fact and a generally accepted theory. During 13 years, 1963 through 1975, there were 73 cases of chronic pancreatitis surveyed by us and alcoholic intake was thought to be involved as a causative factor in approximately 45% of them. The majority of these cases belonged to chronic relapsing pancreatitis and pancreatic lithiasis was found in 90% of them. The mechanism involved in the pancreatic damage due to alcohol has been not fully understood as yet. It was generally thought, by early investigators, that alcohol ingestion induced stimulation of the exocrine secretion of the pancreas and simultaneously created partial obstruction of the pancreatic ductal system. At the same time it was found that serum gastrin and secretin increased after alcohol ingestion. It was also reported that chronic consumption of alcohol changed the
nature of the pancreatic juice, creating intraductal plug, and therefore created obstruction to the pancreatic out-flow.

The purpose of the present investigation is to observe the effects of long term alcohol administration especially in regard to the polypeptide hormone contents of the gastrointestinal mucosa, composition of the pancreatic juice, and histological finding of the pancreas.

Materials and Methods

Male Wistar strain rats weighing 100–200 g were subjected to the experiment. For 60 weeks, 134 rats were given 20% ethanol instead of water, 86 rats were given water and all rats were maintained on a balanced diet. At the initiation of experiment and thereafter periodically in every fifth weeks, body weight was measured. In each 15th weeks, 10 rats from both ethanol and control group were separated. Five rats were subjected to the histological study and determination of the polypeptide hormone contained in the gastrointestinal mucosa.

No diet was given to these rats 24 hours prior to the sacrifice. In the ethanol group, ethanol administration was discontinued and water was given instead of ethanol. The rats were killed by a blow on the neck. The pancreas, liver, ventral part of glandular stomach and oral part of the duodenum were removed and placed in Zencker-Formalin solution for the subsequent histological examination.

Dorsal part of glandular stomach, anal part of the duodenum and 20 cm oral jejunum were used for the assay of gastrointestinal hormones. The mucosal surface of these organs was cleaned and ablated from the tunica muscularis as evenly as possible. The mucosal membrane of the stomach was placed into a small test tube of polyethylene containing 1.0 ml of physiological saline and that of the upper small intestine was placed into the test tube containing 2.7 ml of 0.01 N HCl. The specimens were immediately heated in a boiling water bath for 3 minutes in order to inactivate the proteolytic enzymes. After heating 0.3 ml of 1.0 N HCl was added to the intestinal specimen. All the specimens were preserved at −20°C until they were extracted. On the occasion of extraction, the froze tissue specimens were thawed, and well homogenized in the test tubes of polyethylene.

When gastrin was extracted, the homogenates were once more heated in a boiling water bath for 30 minutes by adding 1.0 ml of 0.01 M PBS solution, centrifuged at 12,000 rpm at 4°C for 30 minutes, and the supernatants were used for the immunoassay. When secretin and cholecystokinin (CCK) were extracted, the homogenates were centrifuged at 10,000 rpm at 4°C for 15 minutes, and the supernatants were subjected to the bioassay of both hormones. Respective sediments were well dried in a desiccator, and were weighed to 0.01 mg.

The immunological activity of gastrin was determined with the CIS G-RIA kit*. The mean of values estimated in duplicate was devided by the dry weight of sediment obtained after extraction and expresseed in ng/mg. The biological activity of secretin and CCK was measured by the method of Lov4)-Tachibana5) using rat with the pancreatic fistula and by the method of Ljungberg6) using gallbladder of the guinea pig, respectively. The value estimated by both methods was also devided by the dry weight of sediment, and expressed in ng of

* The ahtiserum used in this kit had a high specificity. The percentage of cross-reactions of this antiserum with respect to the immunoreactive gastrin species and circulating substances are calculated as follows:

Gastrin 1-17: 100%
Gastrin 2-17: 73%
Big gastrin: 98%
Gastrin-like pentapeptide: 3.5%
cholecystokinin (CCK): 3.5%