EFFECT OF ETHANOL ON ULTRASTRUCTURE OF THE PANCREATIC ACINI

A. E. Podol'skii

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The histological and ultrastructural changes in the acinar parenchyma of the pancreas produced by the action of ethanol were studied in experiments on Wistar albino rats. Lipid inclusions discovered by the author in the cytoplasm of the acinar cells and changes in the mitochondria were regarded as functional. The opinion of several workers that alcohol has a cytotoxic action on the pancreas is not confirmed by the author. The results of this investigation question the leading role of alcohol in the etiology of acute pancreatitis.

KEY WORDS: *alcohol; acinar cell; pancreas; pathogenesis; pancreatitis.*

In 1878 Friedreich [8] described the "pancreas of the alcoholic." Since that time alcohol has been mentioned increasingly often in the special literature among the etiological factors of pancreatitis. Lefas [10] and Poggenpohl [12] observed a correlation between pancreatitis and alcoholism in clinical practice. According to statistics accumulated by clinicians [4, 8, 11] acute pancreatitis coexisted with alcoholism in 11-68% of cases. Attempts to reproduce alcoholic pancreatitis experimentally have proved unsuccessful [5, 7, 9, 13, 14]. Suggestions that disturbances of metabolism and of external secretion were linked with damage to the pancreas have not been confirmed morphologically [5, 7, 8, 13, 14, 17]. Some light has been shed on the problem by electron-microscopic studies [6, 16] which have revealed foci of cytoplasmic destruction in the acinar, centroacinar, and duct cells.

The problem of the level of the structural formations of the gland at which the changes lying at the basis of the disturbed secretion or destruction of the parenchyma in alcoholic poisoning take place still remains unanswered. Yet the need for a solution to this problem at the present time is urgent.

EXPERIMENTAL METHOD

Experiments were carried out on 25 Wistar albino rats weighing from 120 to 150 g. Twenty experimental rats received a balanced diet but instead of water they were given a 20% solution of ethanol. Five rats served as the control. The animals were decapitated after brief (from 15 min to 24 h) and prolonged (from 1 to 14 days) consumption of ethanol. The pancreas was studied after intervals of 15, 30, 60, and 90 min, 2 and 3 h, and 1, 3, 7, and 14 days. For histological investigation sections were stained with hematoxylin–eosin, with picrofuchsin by Van Gieson's method, by Foot's method, with orcein, Sudan III, and Sudan Black B. Zymogen granules were revealed by Mallory's reaction and RNA by Brachet's method. The objects for electron-microscopic examination were embedded in Epon 812 and ultrathin sections cut on the LKB-111 ultratome were studied in the UEMV-100K electron microscope.

EXPERIMENTAL RESULTS

Macroscopically the pancreas of the experimental animals was indistinguishable from the control at all times of the investigation. Histological and histochemical investigations revealed no destructive changes in the acinar cells, the acini, or the exocrine apparatus of the gland as a whole.

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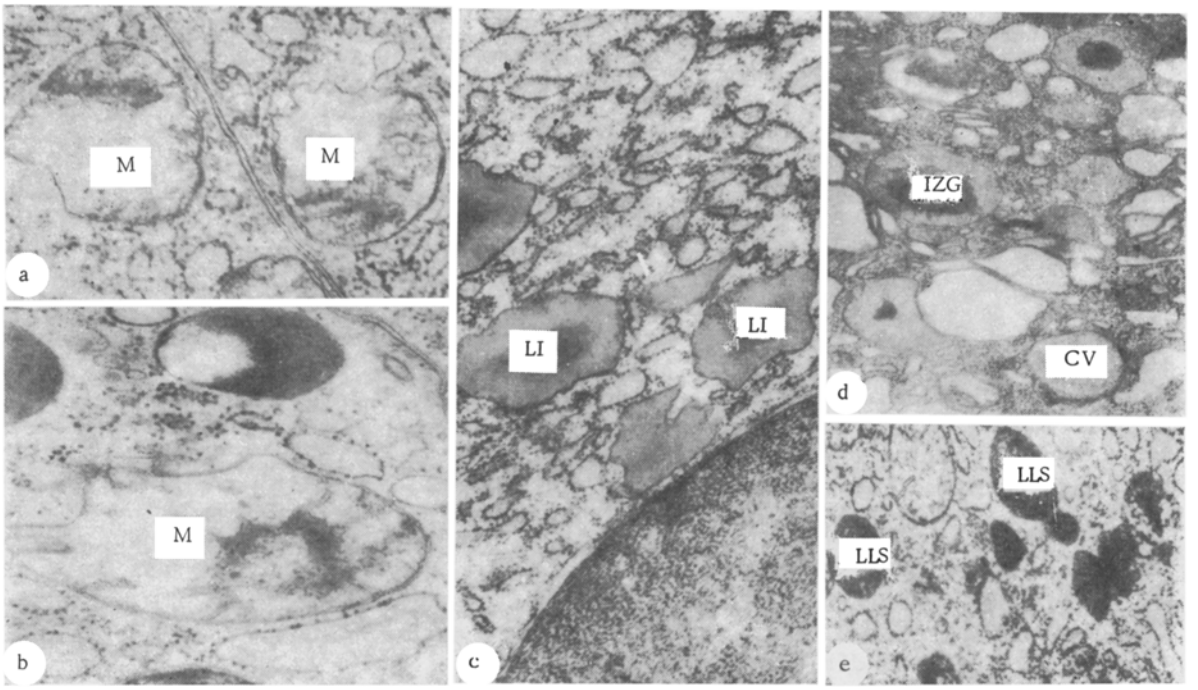

Fig. 1. Ultrastructure of cytoplasmic organelles of pancreatic acinar cells in alcohol loading. a and b) Thinning and evagination of membrane of mitochondria (M) and focal lysis of cristae (22,000×); c) lipid inclusions (LI) in cytoplasm of basal zones of acinar cells (18,000×); d) condensation vacuoles (CV) and immature (IZG) and mature zymogen granules at periphery of hypertrophied Golgi complex (16,000×); e) lysosome-like structures (LLS) in cytoplasm of acinar cells (16,000×).

Electron-microscopically, 15-30 min after consumption of ethanol the changes observed included clearing of the cytoplasm of the endothelial cells of the blood capillaries, numerous pinocytotic vesicles in the cytoplasm, and a few ribosomes and mitochondria with a dense, osmiophilic matrix and clearly defined cristae. The interendothelial junctions were dense and osmiophilic. The basement membrane of the capillaries and acini was well outlined. The pericapillary spaces were not dilated. The plasma membrane of the acinar cells was osmiophilic and clearly defined throughout its length. Many free ribosomes were present in the cytoplasm of the acinar cells, and the cisterns of the rough endoplasmic reticulum were moderately dilated. The mitochondria contained a clear matrix and foci of lysis of the cristae. The nuclei had uniformly distributed chromatin in the nucleoplasm. In the apical zones of the cytoplasm there were solitary mature granules of zymogen, immature granules, and condensation vacuoles. The lumen of the secretory capillaries was filled with osmiophilic contents. In the duct cells there were lipid inclusions in the apical zones of the cytoplasm.

The number of macro- and micropinocytotic vesicles and ribosomes in the cytoplasm of the endothelial cells was increased after 60-90 min. Dilatation of the interendothelial spaces and fenestration of the capillary walls were observed. Vesicular dilatation of the cisterns of the ergastoplasm, lipid inclusions, and numerous ribosomes were present in the cytoplasm of the basal zones of the acinar cells. Small mitochondria with focal clearing of the matrix were situated on the lateral surface of the plasma membrane. In some mitochondria the cristae were disarranged and shortened and most of them showed lysis. The mitochondrial membrane was thinner than normal, gave off flask-shaped evaginations, and was reduced to a single membrane, defective in places. Where the membrane was thin cristae were absent. In the region of the hypertrophied Golgi complex there were condensation vacuoles, immature and mature zymogen granules, and many free ribosomes. In the apical zones of the cytoplasm there were a few mature and immature zymogen granules, vacuolar structures, and cystic dilatations of the cisterns of the rough endoplasmic reticulum. The number of zymogen granules in the acinar cells of the acini varied, indicating an asynchronous secretory cycle. In the duct cells there were tiny and large vesicles, fibrillary structures, mitochondria, and ribosomes. The ducts contained weakly osmiophilic contents (Fig. 1). The structure of the capillaries and the pericapillary space 2-3 h after administration of ethanol differed only a little from that at the previous time.

Between 24 h and 14 days after taking the alcohol the histological appearance and ultrastructure of the acinar cells and of the ducts and blood capillaries showed no pathological changes.

The ultrastructure of the acinar cells and the contents of the secretory capillaries as described above during the first 30 min of the experiment reflect the excretory phase of the secretory cycle which commenced immediately after administration of the ethanol. However, in the pancreas the clear division of the acinar cells into "dark" and "light" still persisted, which is evidence of continuation of the asynchronous (physiological) rhythm. It is therefore possible to speak of stimulation of secretion in the acinar parenchyma of the pancreas. The reasons for this focal arrangement have not yet been explained. It may be connected with the extra production of secretin by the duodenal mucosa, on which alcohol, according to the testimony of Baylis and Starling [3], has a stimulating effect. The presence of a few lipid droplets in the cytoplasm of the acinar cells may be connected with an increase in fatty acid synthesis under the influence of alcohol or may be the result of mobilization of intracellular energy reserves in response to the extra section of digestive enzymes. No tendency was observed for the number and size of the lipid droplets to increase if the experiment was prolonged. The changes observed in the shape and in the membrane of the mitochondria were in all probability functional, for in the late stages of the experiment after repeated administration of alcohol their ultrastructure was indistinguishable in every respect from the control. Disturbance of the integrity of the inner mitochondrial membrane and defects in the outer membrane reflect a state of increased functional activity, as was observed previously under conditions of intensified digestion [1] or after administration of the corresponding drugs. The fact that only single mitochondria were altered also confirms the functional genesis of these changes. Unlike Darle et al. [6], the present writer is not inclined to regard the presence of lysosomes in the cytoplasm of the acinar cells as a manifestation of local cytoplasmic destruction. This opinion is supported by two factors: First, in none of the experiments were disorganization and destruction of the intracellular structures found, and second, the structure of the lysosomes differed considerably from the structure of the classical autophagosomes, the presence of which can be taken as evidence of destructive changes in the cytoplasm. Consequently, changes in the ultrastructure of the acinar cells observed in response to alcohol loading, described above, were functional and cannot be regarded as destructive. Considering these experimental results, it can tentatively be suggested that the importance of pure alcohol in the pathogenesis of acute pancreatitis is relatively small.

LITERATURE CITED

1. N. K. Permyakov, A. E. Podol'skii, and G. P. Titova, Ultrastructural Analysis of the Secretory Cycle of the Pancreas [in Russian], Moscow (1973).
2. A. A. Shelagurov, Diseases of the Pancreas [in Russian], Moscow (1970).
3. W. M. Baylis and E. H. Starling, J. Physiol. (London), 28, 325 (1902).
4. E. Clark, Am. J. Dig. Dis., 9, 426 (1942).
5. J. Cueto, N. Tajen, and B. Zimmerman, Surgery, 62, 159 (1967).
6. N. Darle, R. Ekholm, and J. Edlung, Gastroenterology, 58, 62 (1970).
7. D. A. Dreiling, A. Richman, and N. F. Fradkin, Gastroenterology, 20, 636 (1952).
8. N. Friedreich, in: Cyclopedia of the Practice of Medicine, Vol. 8, New York (1878), p. 601.
9. H. D. Jonowity and M. Bayer, Ann. Intern. Med., 74, 444 (1971).
10. E. Lefas, Arch. Gen. Med., 3, 539 (1900).
11. W. K. Myers and C. S. Keefer, New Engl. J. Med., 210, 376 (1934).
12. Poggenpohl, cited by D. A. Dreiling et al. [7].
13. V. M. Sardesai and J. M. Orten, J. Nutr., 96, 241 (1968).
14. H. Schapiro, L. D. Wruble, and L. G. Britt, Surgery, 60, 1108 (1966).
15. W. Symmons, Dublin J. Med. Sci., No. 143, 244 (1917).
16. F. Tasso, J. Clop, H. Sarles, et al., Pathol. Biol., 15, 733 (1967).
17. B. E. Walton, H. Schapiro, and E. D. Woodward, Ann. Surg., 28, 443 (1962).