

Invited Review

The morphological changes of exocrine pancreas in chronic pancreatitis

**N. Ashizawa¹, M. Niigaki¹, N. Hamamoto², M. Niigaki², T. Kaji²,
T. Katsube², S. Sato², H. Endoh², K. Hidaka², M. Watanabe² and Y. Kinoshita²**

¹Department of Gastroenterology, Shimane Prefectural Central Hospital, Izumo, Shimane and

²Department of Internal Medicine II, Shimane Medical University, Izumo, Shimane, Japan

Summary. The following changes were found by either light or electron microscopic observation of the pancreas in spontaneously developed chronic pancreatitis models (WBN/Kob rats, spontaneously hypertensive rats, and rats with common bile-pancreatic duct stones) and in experimental models of chronic pancreatitis (alcoholic pancreatitis, ischemic pancreatitis, and obstructive pancreatitis): 1) the units of lobules, which were constituted by acinar cell deletion, ductular proliferation, and fibrosis; and 2) tortuous or helical ductal channels of pancreatic ducts with periductal fibrosis, which had many crater-like depressions and very long cilia in their inner surface. These are considered to be the results of obstructive pancreatitis, which are caused by the reactions of defensive factors against the increase of pancreatic duct pressure, including the apoptosis of acinar cells, the hyperplasia and hypertrophy of duct cells, a tighter junctional complex of duct cells, and periductal fibrosis.

Key words: Chronic pancreatitis, Animal models, Pancreatic duct system, Morphology

Introduction

The pathogenesis of chronic pancreatitis is still unknown, and the correlation between acute pancreatitis and chronic pancreatitis is also unclear. In order to elucidate the pathogenesis of chronic pancreatitis, it is necessary to study the characteristics of clinical findings in patients with chronic pancreatitis and the morphological characteristics of exocrine pancreas in normal and chronic pancreatitis. However, since morphological examination of pancreatic tissue in the early stages of human chronic pancreatitis is impossible, animal models of chronic pancreatitis are usually used in order to study the changes during the onset and advancement of

chronic pancreatitis. This article reviews the normal anatomy of exocrine pancreas and its morphological changes in chronic pancreatitis, and also discusses the pathogenesis of chronic pancreatitis.

Clinical findings in patients with chronic pancreatitis

A typical symptom in patients with chronic pancreatitis is repeated abdominal or back pain with an increased serum level of pancreatic enzyme (pancreatic pain), but in practice they have various clinical findings. In the decompensated stage, pancreatic pain, from which patients suffer in the compensated stage, decreases gradually and pancreatic malabsorption and diabetes mellitus occurs. Some patients suffer from continued pancreatic pain, while others have no history of pancreatic pain even in the decompensated stage (asymptomatic chronic pancreatitis). The characteristics of clinical findings in chronic pancreatitis take a variety of clinical courses.

Clinical examinations in patients with chronic pancreatitis

The greatest majority of the etiologies in chronic pancreatitis, as well as in acute pancreatitis, is occupied by alcohol abuse and biliary stones. It has been reported that the main pancreatic duct pressure is increased in patients with alcoholic, gallstone-associated, and idiopathic chronic pancreatitis (Okazaki et al., 1988). Furthermore, irregular dilated pancreatic ducts (bead-like deformity) are found in endoscopic retrograde pancreatography of patients with chronic pancreatitis. Therefore, it is suggested that the increase in pancreatic duct pressure is the most important factor contributing to the onset or advancement of chronic pancreatitis.

Anatomy of normal exocrine pancreas

Light microscopic (LM) examination

The exocrine pancreas consists of acini, pancreatic

Offprint requests to: N. Ashizawa, Department of Gastroenterology, Shimane Prefectural Central Hospital, 116 Imaichi-cho, Izumo 693-8555, Japan. Fax: +81-853-21-2975



Fig. 1. Scanning electron microscopic view of the inner surface of the interlobular duct in a conventionally-fixed Wistar rat's pancreatic tissue. The smooth surface is covered with short microvilli, which are dense along the epithelial cell boundaries and form a polygonal pattern, and each epithelial cell has a short cilium-like process (arrows). Original magnification: $\times 6,000$; Bar: $4 \mu\text{m}$. Reproduced from Ashizawa N. et al. (1991) Scanning electron microscopic observations of three-dimensional structure of the rat pancreatic duct. *Pancreas* 6, 542-550, with permission of the publisher.

ducts, blood vessels, and connective and fatty tissue. The acini occupy most of the lobule, and the intercalated ducts constitute a small percentage of pancreatic tissue volume. The acinus consists of acinar cells and centroacinar cells, which have the same structure as the intercalated duct cell. By reconstructing the pancreatic ducts in serial sections of a normal human pancreas, it was reported that the interlobular ducts were cylindrical and branched like a tree (Homma et al., 1979; Hirabayashi, 1987; Sabusawa et al., 1980). Watanabe et al. (1997) demonstrated that the pancreatic lobule was a glandular lobule dominated by a single intralobular duct. On the basis of the reconstruction of pancreatic tissue, Bockman (1976, 1978) and Akao et al. (1986) reported that the ductules and acini were arranged in a complex curving and branching system of tubules, which anastomose and end blindly (anastomosing tubular arrangement of exocrine pancreas), whereas Takahashi (1984) claimed that there was no anastomosis among the intercalated ducts and acini. However, using only LM examinations, it is impossible to demonstrate the three-dimensional structure of the intra-acinar secretory canaliculus and peripheral intercalated duct, since their lumina are too small (less than $1 \mu\text{m}$ in diameter) to be recognized by LM. We intend to demonstrate the three-dimensional structure of intra-acinar secretory canaliculi by an electron microscopic reconstruction of a $0.4 \mu\text{m}$ -thick serial section.

Scanning electron microscopic (SEM) examination of conventionally-fixed pancreatic tissue

The inner surfaces of the interlobular ducts are smooth and covered with short microvilli, which are



granules in the acinus. $\times 5,000$, Bar: $5 \mu\text{m}$.



Fig. 2. Scanning electron microscopic views of the intercalated ducts in conventionally-fixed Wistar rats' pancreatic tissue. **a.** Note the crater-like depressions (arrows) and short microvilli in the inner surface. $\times 13,000$, Bar: $1 \mu\text{m}$. Reproduced from Ashizawa N. et al. (1995) Scanning electron microscopic observation of pancreatic ducts in WBN/Kob rats. *Pancreas* 11, 389-395, with permission of the publisher. **b.** The intercalated duct close to the acinus possesses a thin wall and small lumen (large arrow, approximately $1 \mu\text{m}$ diameter). Spheres, arranged in the vicinity of the central lumen (small arrow), are zymogen

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dense along the epithelial boundaries, and each epithelial cell has a single cilium-like process (Fig. 1) (Ashizawa et al., 1991). The cilium-like process becomes shorter in proportion to the diameter of the pancreatic ducts (Ashizawa et al., 1995; Hidaka et al., 1995).

The intercalated ducts alone have crater-like depressions (Fig. 2a). The intercalated ducts close to the acini (Fig. 2b) possess a thin wall and small lumina, as small as the central lumina. When examined with LM, it is difficult to detect the small intercalated duct (approximately 1 μm in diameter) in the lobules, which are crowded with large acinar cells.

The central lumen and intercellular secretory canaliculus are termed the intra-acinar secretory canaliculus, since both of them have the same structure.

Fig. 3. Scanning electron microscopic views of the intra-acinar secretory canaliculi in conventionally-fixed Wistar rats' pancreatic tissue. The face is cracked along the cell boundaries of the acinus. Note the central lumina (large arrows) running straight through the center of the acinus, and the intercellular secretory canaliculi (small arrows) extending from the central lumen close to the cell base. x 2,000, Bar: 15 μm . Reproduced from Ashizawa N. et al. (1991) Scanning electron microscopic observations of three-dimensional structure of the rat pancreatic duct. *Pancreas* 6, 542-550, with permission of the publisher.

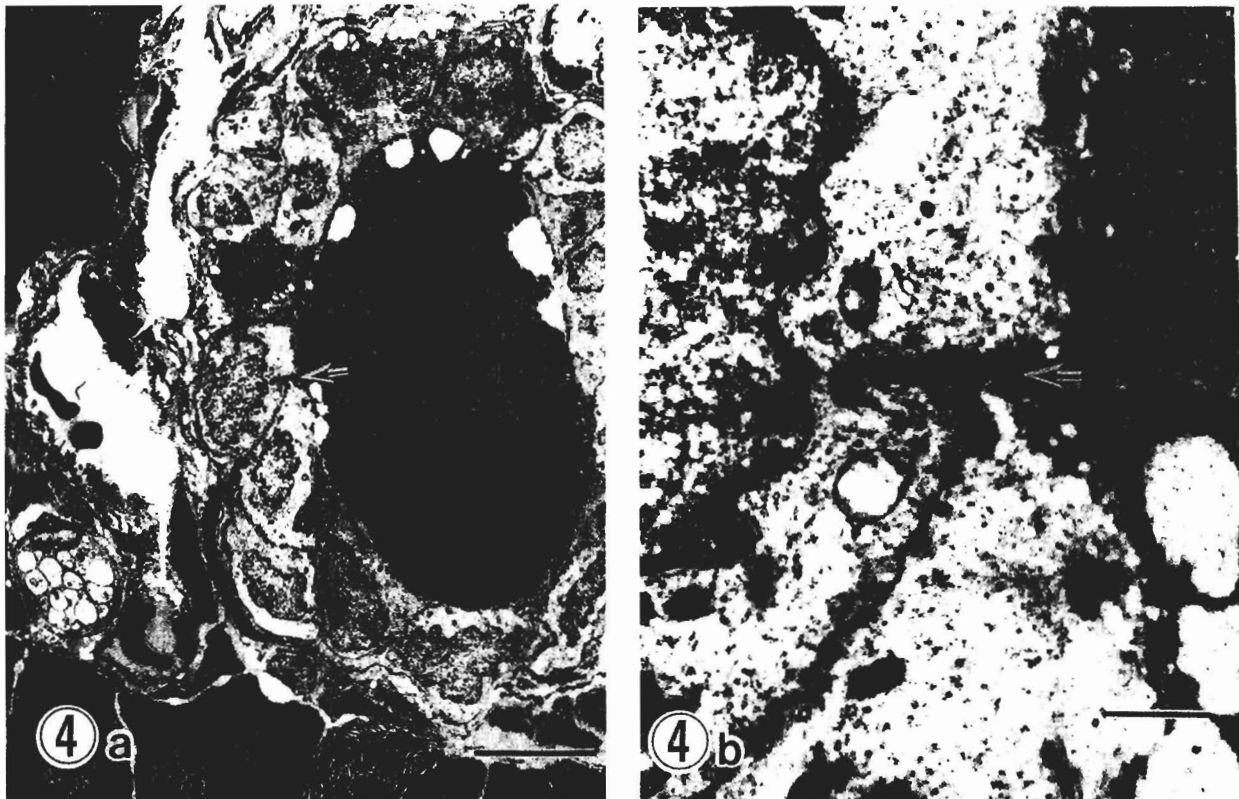


Fig. 4. Transmission electron microscopic views of the interlobular ducts in the Wistar rats. **a.** The arrow points to a cilium. x 1,500, Bar: 10 μm . **b.** Higher magnification of the cilium, which is seen to contain microtubules (arrow). N: nucleus. x 12,000, Bar: 1 μm . Reproduced from Endoh H. and Ashizawa N. (1997). Changes in the inner surface of the pancreatic duct in rats with common bile-pancreatic duct stones. *J. Jpn. Panc. Soc.* 12, 471-479, with permission of the publisher.

However, it seems reasonable to define the central lumen as a canaliculus formed over the three apical surfaces of the intra-acinar cells (acinar cells and centroacinar cells)

and the intercellular secretory canaliculus as a canaliculus between two intra-acinar cells. The intercellular secretory canaliculus extends from the central lumen through the intercellular space to the basement membrane of the acinus (Fig. 3). The termination of the intercellular secretory canaliculi are close to the basement membranes and the intervals between them are 1-4 μm (Takahashi, 1984). This portion probably has the highest pancreatic duct pressure, and is therefore the most liable to leak pancreatic juice out of the pancreatic duct system.

Transmission electron microscopic (TEM) examination of pancreatic tissue

The interlobular duct (Fig. 4) and the intercalated duct (Fig. 5) have cilia containing microtubules and microvilli containing microfilaments. Therefore, the cilium-like process in the inner surface of pancreatic duct presented by a SEM examination was demonstrated to be the cilium. However, the inner structure of the cilium in the pancreatic duct cell is different from that in the ciliated cell (e.g., uterine tube or bronchial epithelium). Most cross sections of the cilia of ciliated



Fig. 5. Transmission electron microscopic view of the longitudinal section of the intercalated duct in the Wistar rats. Note the many microvilli and very long cilia (arrows). x 5,000, Bar: 2 μm . Reproduced from Endoh H. and Ashizawa N. (1997). Changes in the inner surface of the pancreatic duct in rats with common bile-pancreatic duct stones. *J. Jpn. Panc. Soc.* 12, 471-479, with permission of the publisher.

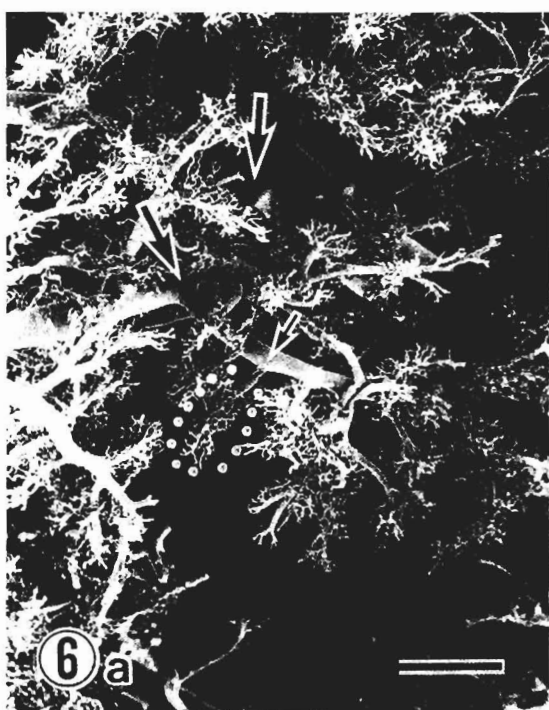


Fig. 6. Scanning electron microscopic views of the corrosion casts from the Wistar rats' pancreatic duct. **a.** Large arrows indicate the interlobular ducts. Small arrows indicate the intralobular ducts. White dots mark the territory of a lobule. x 50, Bar: 300 μm . **b.** Higher magnification of the territory marked by white dots in a. An intralobular duct (arrow) branches from an interlobular duct. Intercalated ducts, which branch off from an intralobular duct, wind and fork into two branches. x 250, Bar: 100 μm . Reproduced from Ashizawa N. et al. (1991) Scanning electron microscopic observations of three-

cells have eleven microtubules, whereas the number of microtubules in the cilium of a pancreatic duct cell is nine at its basal portion and decreases at its distal portion (Hidaka et al., 1995). The cilium of a pancreatic duct cell may have sensory functions rather than motor functions, since Bockman et al. (1986) pointed out that the microtubular pattern in the cilium of a pancreatic duct cell resembled that of the cilium modified for chemoreceptor. Therefore, the cilium of a pancreatic duct cell may not contribute to the transportation of pancreatic juice.

The bile canaliculus, which is very similar to the intra-acinar secretory canaliculus, was revealed to open and close repeatedly by time-lapse cinemicrophotography (Oshio and Phillips, 1981), and this rhythmical contraction was demonstrated to cause unidirectional bile flow (Watanabe et al., 1991). In the hepatocytes, actinfilaments (microfilaments) form a network lying beneath the bile canaliculus surface membrane, which converge mainly to the zonula adherens (intermediate junction) and zonula occludens (tight junction), and the axis of the microvillus. Parts of those actinfilaments connect with each other (Ishii et al., 1991). Bauduin et al. (1975) demonstrated by TEM observation of the pancreatic central lumina that treatment with cytochalasin B, which inhibits actin polymerization, caused the disappearance of microvilli and a dilatation of the lumina. Therefore, actinfilaments beneath the luminal membrane of the pancreatic duct system, which affect the density or the height of microvilli, may contribute to the transportation of pancreatic juice.

Smooth muscle bundles and connective tissue are found around the pancreatic ducts (Egerbacher and Boeck, 1997). The smooth muscle cell coat of the main pancreatic duct in cats is arranged in longitudinally

directed spirals (Garrett et al., 1973). The rhythmical contraction of periductal smooth muscle may contribute to the transportation of pancreatic juice.

SEM examination of corrosion cast of pancreatic duct

We made corrosion casts of Wistar rats' pancreatic ducts according to the method originally described by Murakami et al. (1984). SEM examination of our casts demonstrated that the pancreatic duct system had a tree-like shape and every pancreatic ductal lumen was almost cylindrical (Fig. 6) (Ashizawa et al., 1991). A comparative study between corrosion casts and conventionally-fixed pancreatic tissue by SEM (Ashizawa et al., 1991) demonstrated that our corrosion casts reliably reproduced the lumina of the pancreatic ducts (interlobular ducts to intercalated ducts). White dots in Fig. 6 indicate the territory of a lobule dominated by a single intralobular duct, which is consistent with the results obtained from the reconstruction of pancreatic ducts in serial sections of a normal human pancreas (see above).

Pathological findings in patients with chronic pancreatitis

Chronic pancreatitis is not a disease that uniformly affects the whole pancreas. In histological examinations of each pancreatic tissue specimen obtained from surgery or autopsy of patients with chronic pancreatitis, massive fibrosis, infiltration of inflammatory cells, deletion of acini, ductular proliferation, and replacement with adipose tissue are found. The lesions containing these findings are often contiguous to the almost intact lobules. Furthermore, Comfort et al. (1946) reported that the histological changes of acute pancreatitis were often found in the pancreatic tissue of patients with chronic pancreatitis. Pancreatic tissue in chronic pancreatitis is thought to be the aggregation of segments with a distinct pathological finding.

Morphological changes of exocrine pancreas in spontaneously developed chronic pancreatitis models

WBN/Kob rat

The male WBN/Kob rat is an animal model that spontaneously develops diabetes mellitus associated with massive fibrosis of the pancreas, causing not only exocrine but also endocrine pancreatic insufficiency (Nakama et al., 1985; Tsuchitani et al., 1985). In WBN/Kob rats, increased fibrous tissue and exocrine insufficiency are found after 3 months of age, whereas glycosuria and hyperglycemia develop after 9 months of age (Mori et al., 1990; Ohashi et al., 1990). Moreover, it was reported that WBN/Kob rats had irregular and dilated pancreatic ducts after 2 months of age, which are similar to the pancreatographic findings characteristic of

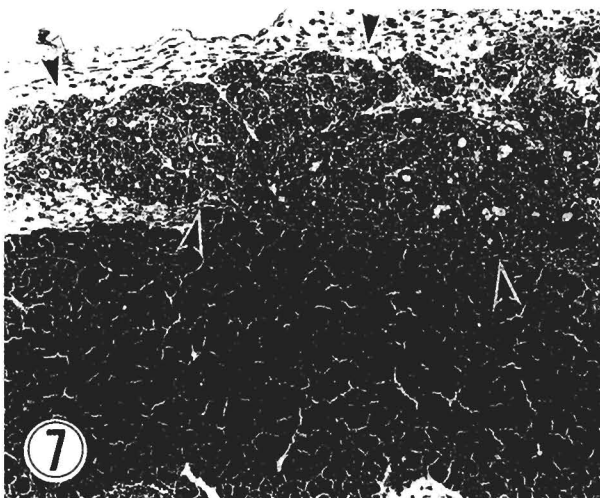


Fig. 7. Light microscopic view of the pancreatic tissue in a 3-month-old WBN/Kob rat. The chronic pancreatitis-like lesion (arrowheads), which contains ductular proliferation and dilatation of the central lumina, is contiguous to the almost intact lobules. Hematoxylin-eosin stain. x 100

human chronic pancreatitis (Ohashi et al., 1990). Therefore, the WBN/Kob rat has recently gained attention as an animal model of chronic pancreatitis.



Fig. 8. Scanning electron microscopic view of a corrosion cast from a 3-month-old WBN/Kob rat's pancreatic duct. Note the tortuous ductal channel of the pancreatic duct (arrows). $\times 50$, Bar: $300\ \mu\text{m}$. Reproduced from Ashizawa N. et al. (1995) Scanning electron microscopic observation of pancreatic ducts in WBN/Kob rats. *Pancreas* 11, 389-395, with permission of the publisher.

We observed the WBN/Kob rats' pancreatic tissue using LM and TEM, and their pancreatic ductal lumina using SEM (Ashizawa et al., 1995; Hidaka et al., 1995). After 3 months, hemorrhaging, an increase in fibrous tissue, and ductular proliferation were found in some regions. The location of these pancreatic lesions corresponded to the units of lobules, and were contiguous to the almost intact lobules (Fig. 7). The corrosion casts of the pancreatic ducts, from the interlobular channels (Fig. 8). After 14 months, the helical form and luminal blebs of the ductal cast material were found (Fig. 9), and the inner surfaces of the interlobular ducts had very long cilia and crater-like depressions (Fig. 10). The bead-like structure of the interlobular ducts seen in pancreatography (Ohashi et al., 1990) or in LM observation of pancreatic tissue (Fig. 11) results from the two-dimensional observation of the helical or tortuous pancreatic ducts. The crater-like depressions in the inner surface of the pancreatic ducts correspond to the luminal blebs of the ductal cast material.

Spontaneously hypertensive rat (animal model of chronic ischemic pancreatitis)

In the pancreatic tissue of spontaneously hypertensive rats (SHR) and stroke prone spontaneously hypertensive rats (SHRSP) over 12 weeks of age, chronic pancreatitis-like lesions, which include deletion of acini, ductular proliferation, and fibrosis, are sporadically dispersed. Onizuka et al. (1994) reported that these lesions were close to the sclerotic arterioles with a marked narrowing lumen. Tsunoda et al. (1994) demonstrated that administration of angiotensin-converting-enzyme inhibitor to SHRSP decreased the chronic pancreatitis-like lesions and the arteriosclerotic

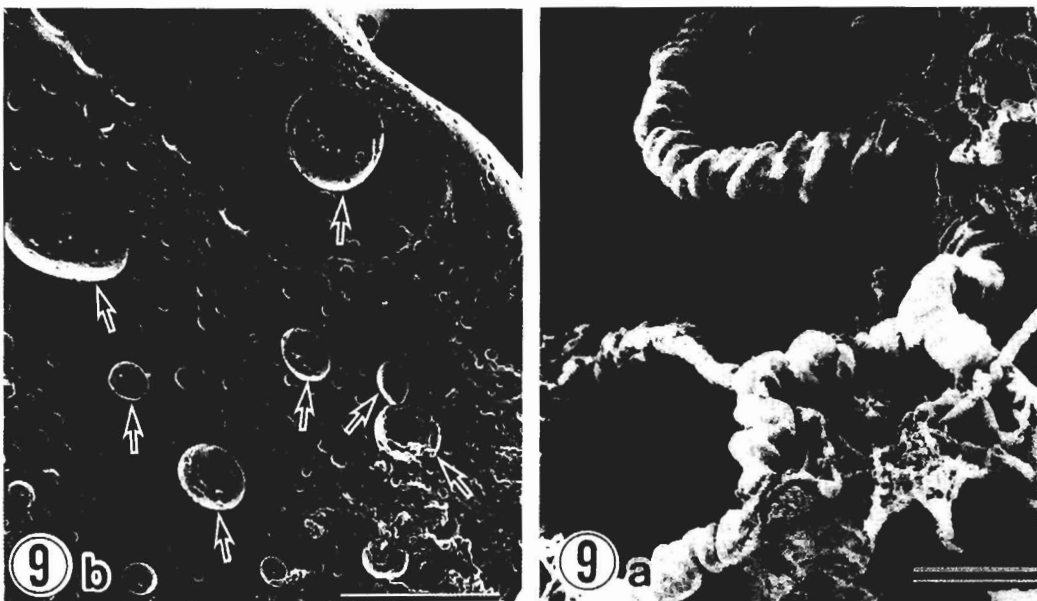


Fig. 9. Scanning electron microscopic view of a corrosion cast from a 14-month-old WBN/Kob rat's pancreatic duct. **a.** Note the helically-formed pancreatic duct. $\times 150$, Bar: $200\ \mu\text{m}$. Reproduced from Ashizawa N. et al. (1995) Scanning electron microscopic observation of pancreatic ducts in WBN/Kob rats. *Pancreas* 11, 389-395, with permission of the publisher. **b.** Higher magnification of the helically-formed pancreatic duct. Note the luminal blebs of the ductal cast material (arrows). $\times 2,000$, Bar: $10\ \mu\text{m}$.

change in the pancreas. Therefore, these rats are considered to be suitable experimental animal models of chronic ischemic pancreatitis. The location of these pancreatic lesions corresponded to the units of lobules, and were clearly demarcated from the surrounding intact lobules (Fig. 12a). Some interlobular ducts had a bead-like deformity (Fig. 12b).

The large interlobular ducts had long cilia and crater-like depressions. The corrosion casts of the pancreatic ducts, from the interlobular ducts to the intercalated ducts, showed tortuous ductal channels and luminal blebs (Fig. 13).

Rats with common bile-pancreatic duct stones (animal models of obstructive pancreatitis)

We found four Wistar rats with markedly dilated common bile-pancreatic ducts that contained soft stones (Endoh and Ashizawa, 1997). In LM examinations of these rats' pancreatic tissue, the interlobular ducts were markedly dilated (Fig. 14a), but deletion of acini, dilatation of pancreatic ducts or central lumina, and ductular proliferation was found in the intralobular regions of only a few lobules (Fig. 14b). In measurements on the TEM photographs of these rats' pancreatic tissue, no clear changes were found in the diameter or density of microvilli in the intercalated ducts and the central lumina (Fig. 15). In SEM examinations, the interlobular ducts had many crater-like depressions and long cilia. These findings suggest that common bile-

pancreatic duct stones in these rats caused an increase in pancreatic duct pressure, which in turn induced luminal dilatation, crater-like depressions, and the elongation of cilia in the inner surfaces of interlobular ducts. However, they apparently had no effect on the morphology of the intralobular region where pancreatic duct pressure is naturally high.

Morphological changes of exocrine pancreas in the experimental models of chronic pancreatitis

Alcoholic pancreatitis

Sarles et al. (1971) reported that in more than 50% of the rats investigated in their study, a 20% ethanol-intake for 20-30 months induced pancreatic lesions very similar to the incipient lesions of human chronic pancreatitis. These lesions corresponded to units of lobules, which contained deletion of acini, ductular proliferation, replacement with adipose tissue, and an increase in fibrous tissue, and were clearly demarcated from the surrounding almost intact lobules. However, other studies (Nagata et al., 1977; Nagasaki, 1978; Matsuno et al., 1983) have reported difficulty in inducing typical chronic pancreatitis in rats through long-term ethanol-administration.

Using TEM, dilatation of the central lumina and a decrease in the density of microvilli on their inner surfaces has been demonstrated in long-term alcohol-intake rats. These changes were believed to correspond to the initial morphological changes of chronic alcoholic pancreatitis (Sarles et al., 1971; Homma et al., 1976; Kakizaki et al., 1987).

We also studied the inner surface morphology of the

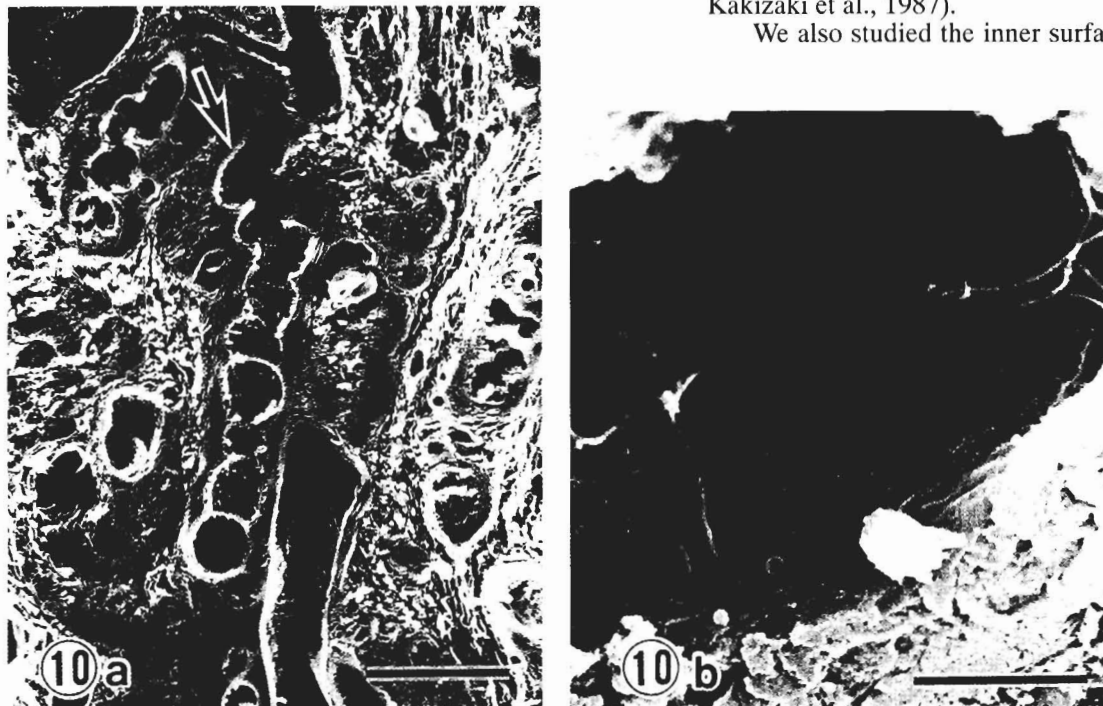


Fig. 10. Scanning electron microscopic views of interlobular ducts in a conventionally-fixed 14-month-old WBN/Kob rat's pancreatic tissue. **a.** Note the helically-formed lumen of the interlobular duct (arrow). x 200, Bar: 100 μ m. **b.** Higher magnification of the inner surface of the helically-formed pancreatic duct. Note the many crater-like depressions and long cilia. x 5,000, Bar: 6 μ m. Reproduced from Ashizawa N. et al. (1995) Scanning electron microscopic

pancreatic ducts in ethanol-intake rats, which were administered 20% ethanol in place of drinking water, for a maximum of six months. We could find no apparent change by SEM examination of the corrosion casts of pancreatic ducts or LM examination of pancreatic tissue, and could not find long cilia and crater-like depressions in SEM examination of the inner surfaces of the interlobular ducts. In the TEM examination of pancreatic tissue, the diameter of the central lumina varied and each central lumen contained a widely varying density of microvilli between the ethanol-intake and control (water-intake) rats and within both groups of rats (Fig. 16). Varied density of microvilli and the varying diameters in the central lumina may result from the rhythmical contraction of the central lumen (see above). However, it

is impossible to estimate the luminal diameter and the density of microvilli in the pancreatic central lumen by some TEM photographs of pancreatic tissue. Measurement is required. In rats administered ethanol for more than three months, duct cells with atrophic microvilli were sporadically found in the inner surfaces of some interlobular ducts (Fig. 17). This finding may indicate an injury of the pancreatic duct cells due to the alcohol administration.

Ischemic pancreatitis

Uchida (1987) observed chronic pancreatitis-like lesions, which corresponded to the units of lobules and were clearly demarcated from the surrounding intact lobules, in rabbits with experimental pancreatic arterial strictures. These lesions contained a deletion of acini, ductular proliferation, dilatation of interlobular ducts and fibrosis, and were also found in rats which had had an injection of microspheres into pancreatic artery (Freiburghaus et al., 1995) and SHR (spontaneously developed chronic ischemic pancreatitis model, see above). Such ductal changes are very similar to those seen in human obstructive pancreatitis and the experimental ligation of rats' pancreatic ducts (see next paragraph). Therefore, chronic ischemic pancreatitis may not be a simple ischemic change (i.e., infarction) of pancreatic tissue, and may be induced by the stasis of pancreatic juice caused by ischemia of the pancreatic tissue.

Obstructive pancreatitis

In rats and mice, ligation of the pancreatic duct induces mild acute edematous pancreatitis and atrophy of pancreatic tissue, which includes deletion of acini, ductular proliferation and fibrosis (Boquist and Edstroem, 1970; Pound and Walker, 1981; Walker, 1987; Walker et al., 1992; Abe and Watanabe, 1995). Atrophy

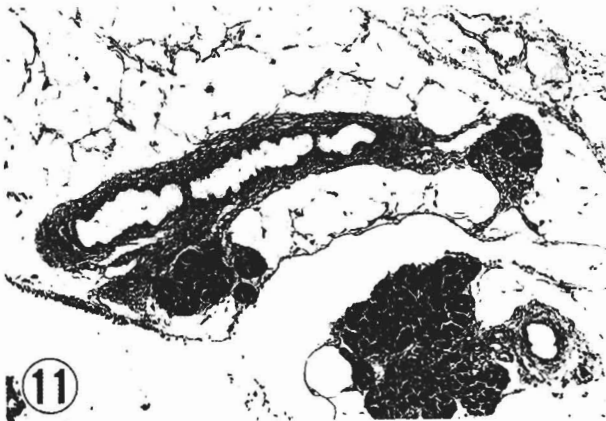


Fig. 11. Light microscopic view of the pancreatic tissue in a 14-month-old WBN/Kob rat. Note the bead-like deformity of the interlobular duct and replacement with adipose tissue. Hematoxylin-eosin stain. x 100. Reproduced from Ashizawa N. et al. (1995) Scanning electron microscopic observation of pancreatic ducts in WBN/Kob rats. *Pancreas* 11, 389-395, with permission of the publisher.

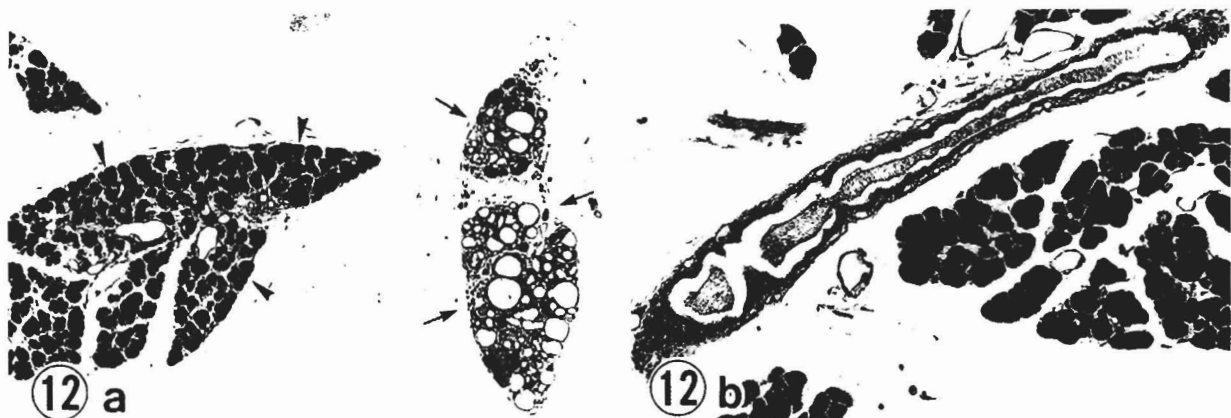


Fig. 12. Light microscopic views of pancreatic tissue in a 12-week-old stroke prone spontaneously hypertensive rat. **a.** Chronic pancreatitis-like lesion (arrows), which includes ductular proliferation, is clearly demarcated from the surrounding intact lobule (arrowheads). x 100. **b.** Note the bead-like deformity of the interlobular duct. x 200. Reproduced from Ashizawa N. et al. (1997). Scanning electron microscope examination of pancreatic ducts in stroke prone spontaneously hypertensive rats (SHRSP). *Int. J. Pancreatol.* 22, 51-57, with permission of the publisher.

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of pancreatic tissue due to deletion of acini results from the apoptosis of acinar cells (Walker, 1987; Walker et al., 1992; Abe and Watanabe, 1995; Doi et al., 1997). On the other hand, bile and pancreatic duct ligation in opossums induces severe acute necrotizing pancreatitis (Senninger

et al., 1986; Lerch et al., 1992, 1993). Kaiser et al. (1995) demonstrated by using experimental bile and pancreatic duct ligation that very little necrosis but a high degree of apoptosis was found in rat pancreatic tissue with mild pancreatitis, and marked necrosis but

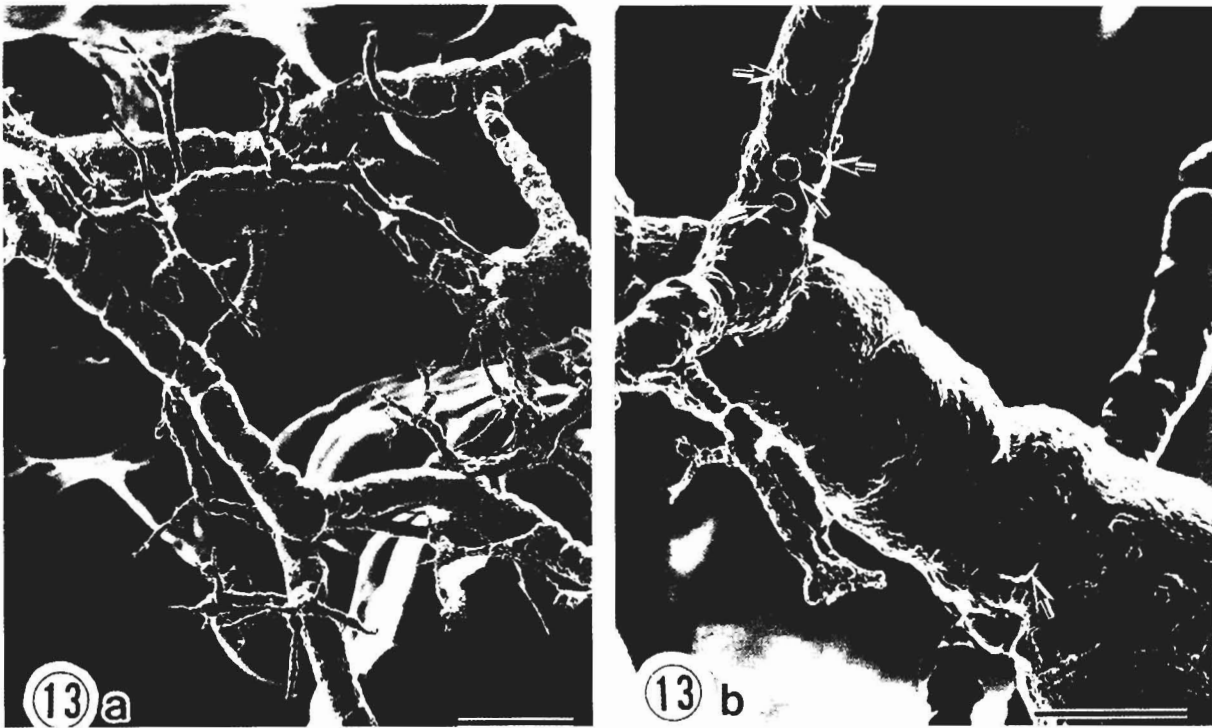


Fig. 13. Scanning electron microscopic view of a corrosion cast from a 12-week-old stroke prone spontaneously hypertensive rat's pancreatic duct. **a.** Corrosion casts show tortuous ductal channels. x 150, Bar: 100 μ m. **b.** Higher magnification of a. Note the luminal blebs of the ductal cast material (arrows). x 1,000, Bar: 20 μ m. Reproduced from Ashizawa N. et al. (1997). Scanning electron microscope examination of pancreatic ducts in stroke prone spontaneously hypertensive rats (SHRSP). *Int. J. Pancreatol.* 22, 51-57, with permission of the publisher.

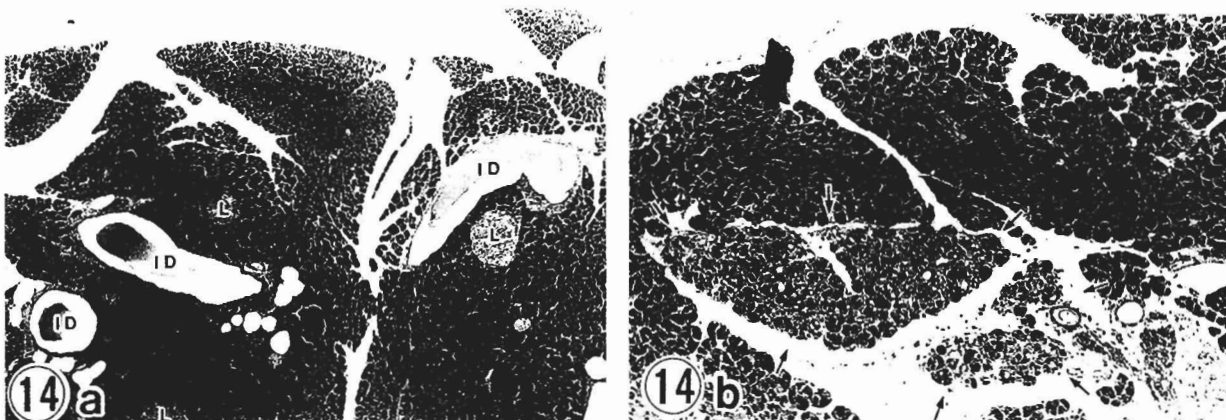


Fig. 14. Light microscopic view of the pancreatic tissue in a 14-month-old rat with common bile-pancreatic duct stones (hematoxylin-eosin stain). **a.** Note the markedly dilated interlobular ducts with mild periductal fibrosis. There was no apparent change in the intralobular regions. ID: interlobular ducts; L: Langerhans' islet. x 40. **b.** The chronic pancreatitis-like lesion (arrows), which includes ductular proliferation and dilatation of the central lumina, is demarcated from the surrounding, almost intact, lobules. x 100. Reproduced from Endoh H. and Ashizawa N. (1997). Changes in the inner surface of the pancreatic duct in rats with common bile-pancreatic duct stones. *J. Jpn. Panc. Soc.* 12, 471-479, with permission of the publisher.

very little apoptosis in the opossum tissue with severe pancreatitis. The degree of severity of acute pancreatitis is inversely related to the degree of apoptosis. These results suggest that massive apoptosis of acinar cells in the early stage of obstructive pancreatitis prevents acinar cells from necrosis and minimizes the severity of acute pancreatitis. Walker et al. (1992) reported that the ductular proliferation resulted not only from the disappearance of acinar cells but also from the hyperplasia and hypertrophy of intercalated duct cells. Wada et al. (1997) demonstrated by immunohistochemical study that proliferating cell nuclear antigen (PCNA: cell cycle-related protein) and B-cell leukemia-2 (Bcl-2: antiapoptosis protein) expression appeared in ductular cells forming the tubular complex from the early stages after the ligation of the rats' pancreatic duct, and increased significantly when acinar cells were diminishing. Therefore, it is considered that pancreatic duct ligation induces the apoptosis of acinar cells, whereas it induces proliferation of ductular cells while evading apoptosis of the ductular cells.

Our SEM observations on the lumina of rats' interlobular ducts after the ligation of bile and pancreatic ducts revealed tortuous or helical ductal channels with long cilia and crater-like depressions in their inner surfaces (unpublished). In a normal pancreas, the cilium becomes shorter in proportion to the diameter of the pancreatic duct, and crater-like depressions are found only in the intercalated ducts (see above). Therefore, it is suggested that longer cilia and more crater-like depressions are found in proportion to the pancreatic duct pressure, since the pancreatic duct pressure is considered to be decreased in proportion to the diameter of the pancreatic duct.

Pathogenesis of chronic pancreatitis

Inflammation is a reaction of the defensive factors against the offensive factors. We propose that the temporarily finished reaction of the defensive factors against the offensive factors in exocrine pancreas should be defined as acute pancreatitis and the continuous

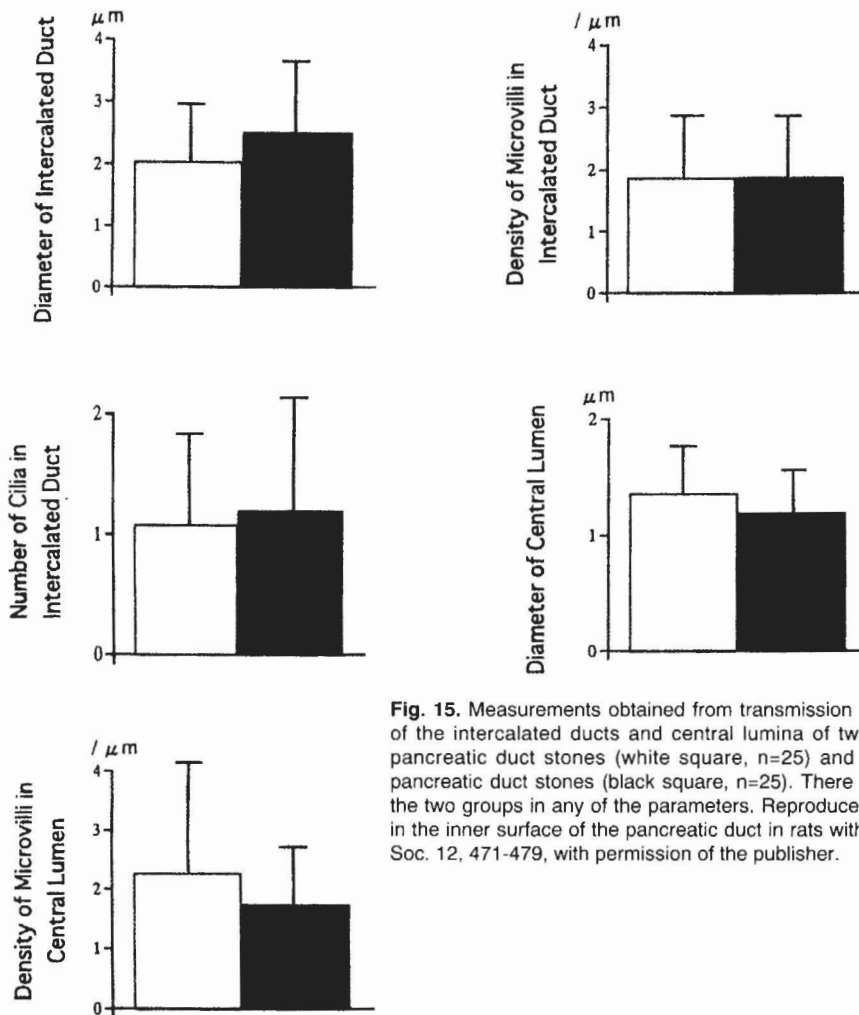


Fig. 15. Measurements obtained from transmission electron microscopic photographs of cross sections of the intercalated ducts and central lumina of two 14-month-old Wistar rats without common bile-pancreatic duct stones (white square, n=25) and two 14-month-old Wistar rats with common bile-pancreatic duct stones (black square, n=25). There were no statistically significant differences between the two groups in any of the parameters. Reproduced from Endoh H. and Ashizawa N. (1997). Changes in the inner surface of the pancreatic duct in rats with common bile-pancreatic duct stones. *J. Jpn. Panc. Soc.* 12, 471-479, with permission of the publisher.

Morphology of chronic pancreatitis

reaction as true chronic pancreatitis. In fact, the pancreatic tissue in patients with chronic pancreatitis consists of parts of true chronic pancreatitis, those of acute pancreatitis, and those of scar as a consequence of acute pancreatitis.

The degree of severity in acute pancreatitis depends on the delay of the expression of defensive factors against the offensive factor. A prompt and adequate expression of defensive factors against offensive factors will evade severe acute pancreatitis, and a delayed and inadequate expression of defensive factors will induce severe acute pancreatitis. The offensive factors include an increase of pancreatic duct pressure, activation of proteases in the pancreatic tissue (reflux of bile or duodenal juice to the pancreatic duct, colocalization of lysosome and zymogen granule, etc.), and toxicity of the duct cell or acinar cell (ischemia, infection, etc.). An increase of pancreatic duct pressure alone, which can be a continuous offensive factor, can cause true chronic pancreatitis subsequent to acute pancreatitis. Therefore, true chronic pancreatitis corresponds to asymptomatic obstructive pancreatitis. The defensive factors include apoptosis of acinar cells, hyperplasia and hypertrophy of duct cells, a tighter junctional complex of duct cells, periductal fibrosis, protease inhibitors, a mucous barrier on the ductal inner surfaces, various immunoreactions, etc.

The pancreas, which has abundant proteases, is a very dangerous organ. Inflammation with necrosis of the acinar cells causes a scattering of activated proteases on the surrounding tissue. During the increase of pancreatic duct pressure, the most dangerous portion anatomically

is the interval between the end of intercellular secretory canaliculus and the acinar basement membrane. This portion has the highest pancreatic duct pressure and the narrowest interval between the inner surface of pancreatic duct system and the basement membrane, and pancreatic juice is liable to leak out of the pancreatic duct system to the interstitial tissue or blood vessels. Therefore, the most important defensive factor against increased pancreatic duct pressure must be the apoptosis of acinar cells, which causes the cessation of synthesizing proteases, the deletion of the most dangerous portions, and the decrease of zymogen enzyme. It is suggested that the pancreatic duct will stand up to the increased pancreatic duct pressure by the hyperplasia and hypertrophy of duct cells, the tighter junctional complex of duct cells, and the periductal fibrosis. Hypertrophy or the tighter junctional complex of duct cells may induce many crater-like depressions and long cilia in the inner surface of the pancreatic duct due to the changes in the cytoskeleton of the duct cells. Hyperplasia or hypertrophy of the duct cells and an increase in periductal fibrosis or smooth muscle may cause tortuous or helical ductal channels in the pancreatic ducts, since they are originally arranged in a helical pattern. An adequate expression of these defensive factors against the continuous offensive factor (increased pancreatic duct pressure) induces true chronic pancreatitis subsequent to mild acute pancreatitis (Fig. 18d), which is thought to be accompanied by no apparent symptoms (asymptomatic chronic pancreatitis). Causes for the increase of pancreatic duct pressure include: 1) mechanical stenosis or occlusion of the

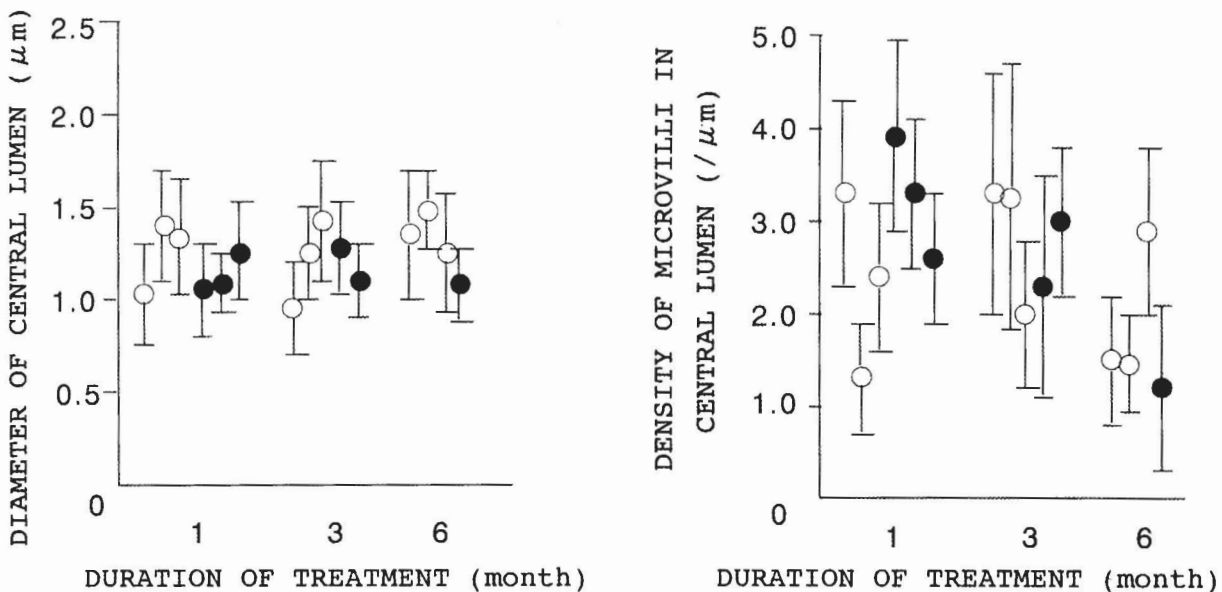
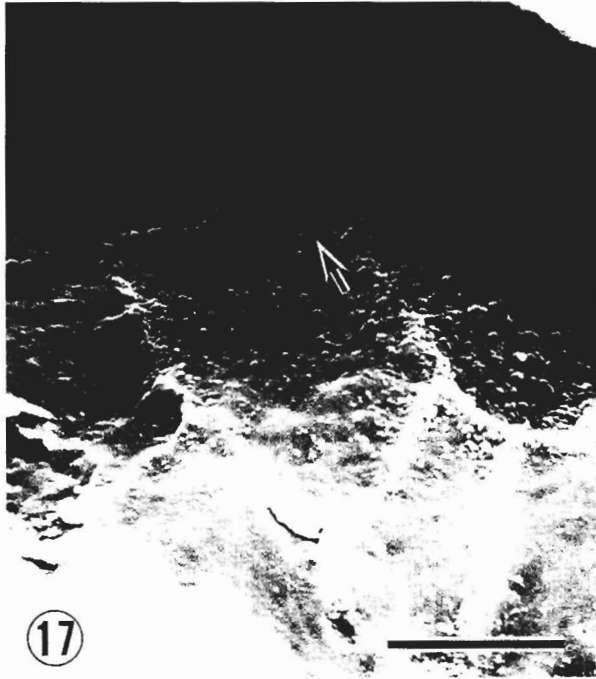


Fig. 16. The results of measurement in 25 pancreatic central lumina on the transmission electron microscopic photographs in each rat. black circle: ethanol-intake rat, white circle: control (water-intake) rat. Value represents mean \pm SD (n=25) in each rat. **a.** Diameter of central lumina in each rat. There is no apparent difference between either group of rats. **b.** Density of microvilli in cross sections of central lumina in each rat. Each central lumen contained a widely varying density of microvilli between the ethanol-intake and control (water-intake) rats and within both groups of rats



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Fig. 17. Scanning electron microscopic view of the interlobular duct's inner surface in the 5-month-old (treated for three months) ethanol-intake rat's conventionally-fixed pancreatic tissue. There are some duct cells with atrophic microvilli. Arrow indicates cilium. x 5,000, Bar: 5 μm.

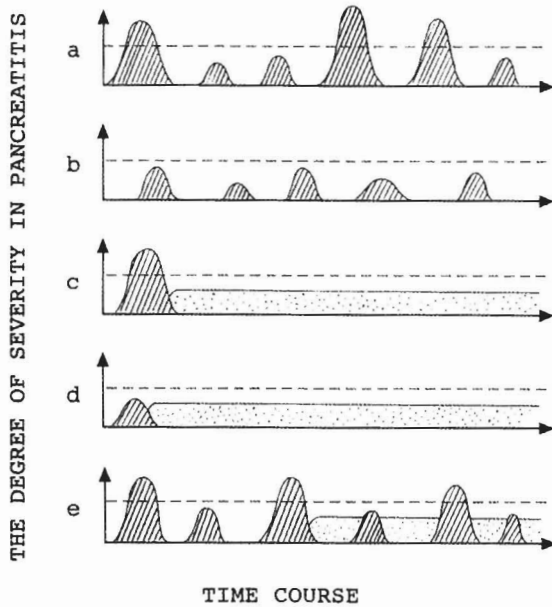


Fig. 18. The schema of various clinical courses of chronic pancreatitis. The areas filled with oblique lines express the severity of acute pancreatitis. The areas filled with dots express the true chronic pancreatitis (asymptomatic obstructive pancreatitis). In the severity of pancreatitis, the degree below a broken line means the level at which patients have no apparent symptoms.

pancreatic duct due to tumor, fibrosis, biliary stone, pancreatic stone, protein plug, etc; 2) sphincter of Oddi dysfunction; 3) change in the characteristics (hyperviscosity) of pancreatic juice due to injury or metaplasia of the duct (or acinar) cells; 4) hypersecretion of pancreatic juice; and 5) an inability of the pancreatic duct to transport pancreatic juice (injury to the periluminal actin filaments or periductal smooth muscle cells). Therefore, the changes associated with acute pancreatitis, which is caused by the offensive factor except for the increase in pancreatic duct pressure, can cause an increase in pancreatic duct pressure thereby inducing true chronic pancreatitis (asymptomatic obstructive pancreatitis). This sequence is compatible with the conclusion of Sarles et al. (1993) that acute pancreatitis is not a cause of chronic pancreatitis in the absence of residual duct strictures (the increase in pancreatic duct pressure).

The concept of a necrosis-fibrosis sequence proposed by Comfort et al. (1946) saying that recurrent attacks are the basis for the development of chronic pancreatitis is supported by some reports (Kloepfel and

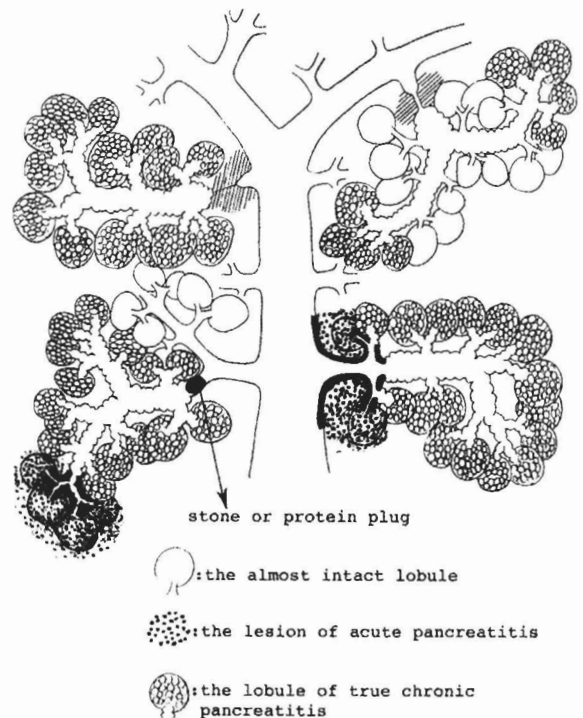


Fig. 19. The schema of the pathological changes of pancreatic tissue with so-called chronic pancreatitis. The areas filled with oblique lines indicate scars accompanied by stenosis or occlusion of interlobular ducts. Thick lines indicate the portions of the pancreatic duct cells injured by acute pancreatitis. The increase in pancreatic duct pressure due to the disturbance of the pancreatic juice flow induces the morphological changes of acute pancreatitis or true chronic pancreatitis (asymptomatic obstructive pancreatitis) subsequent to mild acute pancreatitis in the distal lobules from the disturbed point. With the increase of pancreatic duct pressure, more peripheral lobules are liable to be affected, since those have higher pancreatic duct pressure.

Maillet, 1991, 1993; Ammann and Muellhaupt, 1994; Ammann et al., 1996). Scars from repeated episodes of acute pancreatitis, which include asymptomatic acute pancreatitis, may contribute to the advancement of fibrosis in chronic pancreatitis (a or b in Fig. 18). However, in fact, the pancreatic tissue of patients with typical chronic pancreatitis consists of not only the parts of acute pancreatitis or scars but also those of true chronic pancreatitis (asymptomatic obstructive pancreatitis). In conclusion, the multiformity of the acute pancreatitis-true chronic pancreatitis sequence or ratio causes various clinical and pathological findings (Figs. 18, 19).

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