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Studies of Standard Liver Function and Hepatic Fibrosis in Rats with Obstructive Jaundice and Biliary Drainage*)

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ABSTRUCT

The authors prepared the obstructive jaundice model on which the external biliary drainage was operable, and tried to clarify the critical duration of obstruction from the aspects of the standard liver function and hepatic fibrosis, and the following results were obtained.

1) The T. B, Al-p, GOT and GPT levels in 1 to 4 weeks after the common bile duct obstruction was significantly high ($p<0.01$) as compared with the normal levels, but no definite differences were found between each period of obstruction. After the biliary drainage was performed for 5 days in each period of obstruction, the GOT level was significantly high ($p<0.05$) as compared with the normal level in 1, 3 and 4 weeks, but no significantly differences were noted in the T. B, Al-p and GPT levels in 1 to 4 weeks.

2) Classifying the severity of hepatic fibrosis into the scores from 0 to 3, the severity score of hepatic fibrosis indicated higher levels with the prolongation of the common bile duct obstruction, and significant differences were observed between 1 and 2 weeks ($p<0.05$), and between 3 and 4 weeks ($p<0.01$). The severity score of hepatic fibrosis after biliary drainage showed significantly lower levels ($p<0.05$) as compared with the common bile duct obstruction in 1 to 3 weeks. In 4 weeks, however, no significant difference was noted.

INTRODUCTION

In recent years, percutaneous transhepatic cholangiodrainage (PTCD) has been spreading, and the results of surgical therapy on obstructive jaundice have been improving. However, if serious jaundice persists for a long period, impairments are often observed not only in the liver, which is the main organ for the metabolism of a organism, but also in other organs, bringing about serious complication like acute gastric mucosal lesion (AGML), hepatic failure or renal failure. Accordingly, it is considered important to understand the pathological states of various organs accompanying the chronological changes of obstructive jaundice, and to find out the appropriate period for relief of the obstruction.

In the present study, the authors tried to clarify the critical duration of obstruction from the aspects of the standard liver function and hepatic fibrosis.

MATERIALS AND METHODS

1) *Animal procedures*

Seventy-one Wistar strain male rats were

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used for the study. Venous catheters were introduced into the common bile ducts of rats[2]. After closing the other ends of the catheters, the animals were bred for 1 to 4 weeks under constant temperature (the jaundice group). In 1 to 4 weeks during the period of obstruction, the catheters were opened for each week, and external biliary drainage was performed for 5 days (the drainage group).

2) *Determination of the standard liver function*

After fasting the animals for 24 hr, they were sacrified by decapitation and the blood was immediately collected. Serum total bilirubin (T. B) was determined by the modified method of Michaëlsson, Al-p by the method of Bessey, and transaminase (GOT, GPT) by the method of Wroblewski.

3) *Observation of hepatic fibrosis*

A piece of liver was fixed in 10% formaldehyde solution and embedded in paraffin. Subsequently, sections of liver were cut at 6 μ and stained with haematoxylin and eosin, and Azan-Mallory methods. In accordance with the degrees of fibrogenesis, the following scores were decided: the amount of fibers observed in the non-treated group (control) was made Score 0 (Fig. 1), the proliferation of fibers observed only in the portal area or around the central vein was made Score 1 (Fig. 2), when proliferation of fibers in the portal area extended into the liver parenchyma, or when proliferation of fibers established a shunt between the portal areas or between the portal area and the central vein, it was made Score 2 (Fig. 3), and pseudolobular formation was made Score 3 (Fig. 4). When 10 visual fields per 1 preparation (1 rat) were observed randomly, the severity score of hepatic fibrosis of each animal was denoted with the mean value.

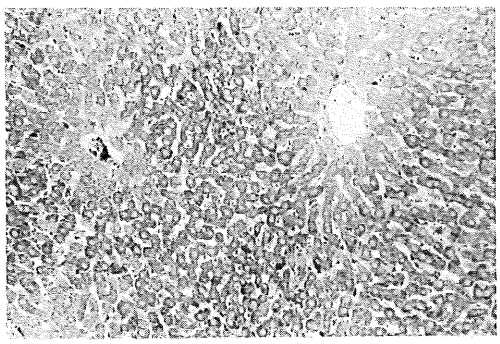

Fig. 1. Histological pictures of a normal rat liver (score 0). A little fiber can be seen around the portal area. Azan-Mallory stain, ×200

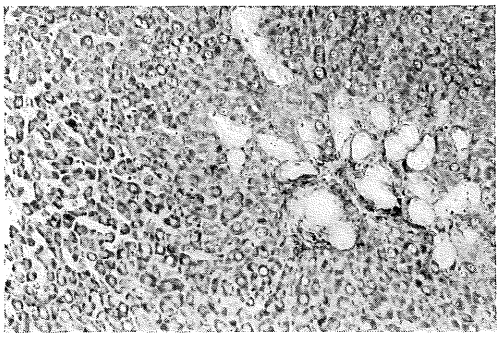

Fig. 2. Histological pictures of a rat liver in 2 weeks after the common bile duct obstruction (score 1). The proliferation of fibers can be seen around proliferating bile ductules. Azan-Mallory stain, ×215

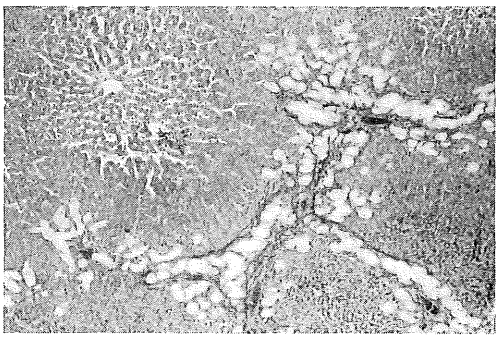

Fig. 3. Histological pictures of a rat liver in 4 weeks after the common bile duct obstruction (score 2). The proliferation of fibers establish a shunt between the portal areas. Azan-Mallory stain, ×100

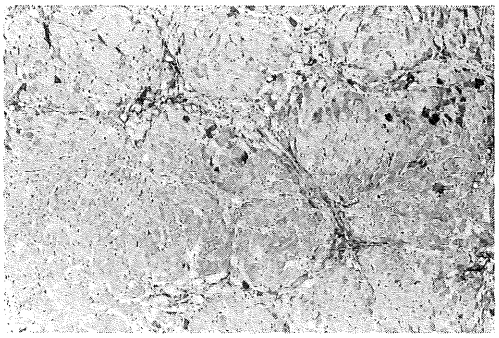

Fig. 4. Histological pictures of a rat liver on the 5th day after biliary drainage in 4 weeks after the common bile duct obstruction (score 3). Pseudolobular formation can be seen. Azan-Mallory stain, ×110

Student's t-test was employed to determine statistical significance.

RESULTS

1) *Changes in the standard liver function* (Fig. 5)

The T. B level in the jaundice group was significantly high ($p<0.01$) as compared with the control group (nomal rat) from 1 week, reaching the plateau level during 3 and 4 weeks, and each of Al-p, GOT and GPT levels were also significantly high ($p<0.01$) in 1 to 4 weeks.

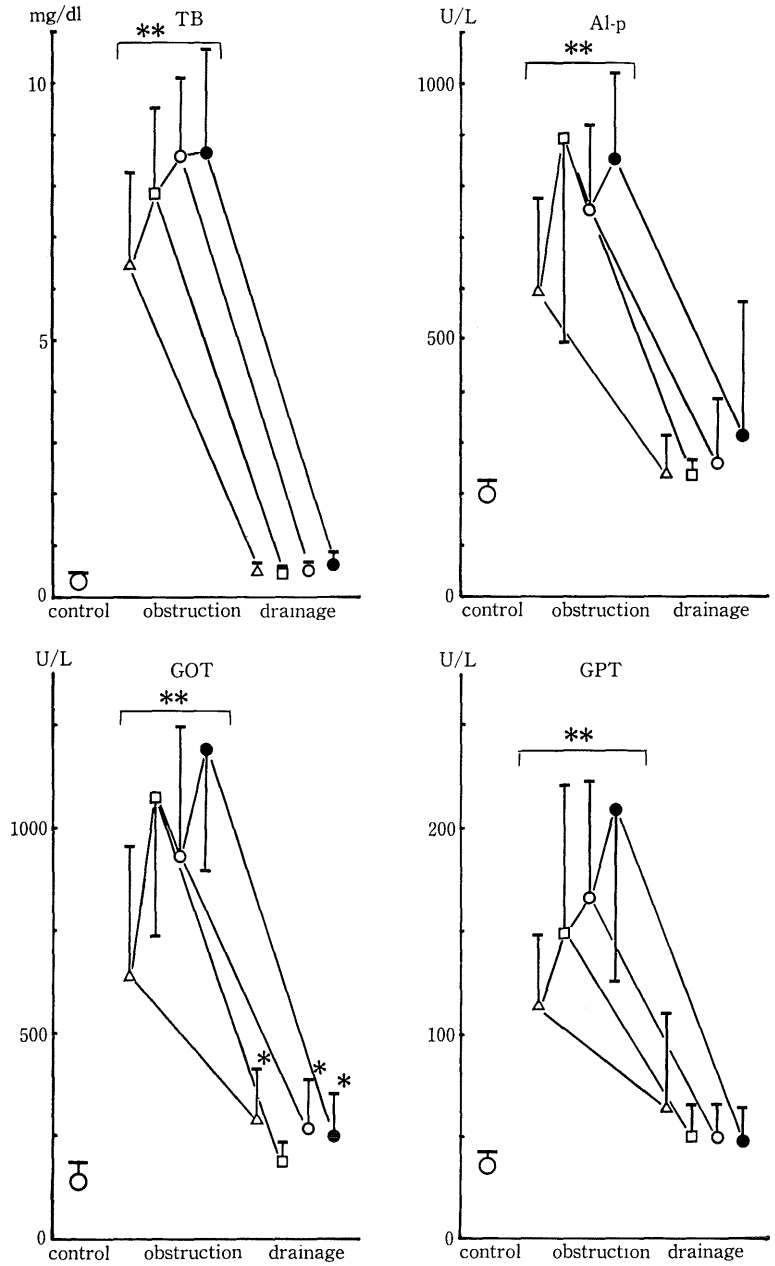

Fig. 5. Changes in the standard liver function test in rats with obstructive jaundice and levels on the 5th day after biliary drainage. △—△ 1 week, □—□ 2 weeks, ○—○ 3 weeks, ●—● 4 weeks * Significance of the difference between means of the control and each group value. (n=7, M±SD, *$p<0.05$, **$p<0.01$)

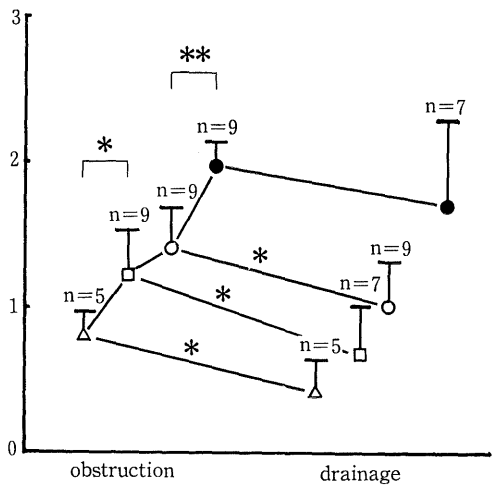

Fig. 6. Changes in the severity score of hepatic fibrosis in rats with obstructive jaundice and level on the 5th day after biliary drainage. △—△ 1 week, □—□ 2 weeks, ○—○ 3 weeks, ●—● 4 weeks
* Significance of the difference between means of the each group value. (M±SD, *$p<0.05$, **$p<0.01$)

In the drainage group, although the GOT level was significantly high as compared with the control group in 1, 3 and 4 weeks ($p<0.05$), no significant differences were noted in the T. B, Al-p and GPT levels as compared with the control group, in 1 to 4 weeks.

2) *Changes in the severity score of hepatic fibrosis* (Fig. 6)

The severity score of hepatic fibrosis in the jaundice group was 0.8±0.2 in 1 week, 1.2±0.3 in 2 weeks, 1.4±0.3 in 3 weeks, and 2.0±0.2 in 4 weeks, indicating higher levels with the prolongation of the obstruction, and siginificant differences were observed between 1 and 2 weeks ($p<0.05$), and between 3 and 4 weeks ($p<0.01$). The severity score of hepatic fibrosis in the drainage group was 0.4±0.2 in 1 week, 0.7±0.4 in 2 weeks, 1.0±0.3 in 3 weeks, and 1.7±0.6 in 4 weeks, showing significantly lower levels ($p<0.05$) as compared with the jaundice group, in 1 to 3 weeks. In 4 weeks, however, no significant difference was noted.

DISCUSSION

The authors prepared the obstructive jaundice model with rats on which the external biliary drainage was operable[2], and the standard liver function test was examined. It was found as a result that hyperbilirubinemia and liver dysfunction were noted in 1 week after the common bile duct obstruction, which persisted until 4 weeks, but no definite differences were found between each period of obstruction. It was further found that, after biliary drainage in each period of obstruction, a trend of the improvement of hyperbilirubinemia and liver dysfunction almost to the normal levels was recognized in any of the weeks. Consequently, it is considered difficult to foresee the critical duration of obstruction from the aspects of the standard liver function test.

In histological findings of liver after the common bile duct obstruction, marked bile stasis and proliferation of the distended bile ductules were observed chronologically. It is believed that hepatic fibrosis occurs in principle in three locations: 1) in the portal tracts, 2) around hepatocytes and 3) around proliferating bile dectules which are located in the portal tracts but also extend into the parenchyma[4]. It is well known that hepatic fibrosis per se and the resulting circulatory disturbance aggravate the liver dysfunction. It is presumed, therefore, that hepatic fibrosis may be an important factor contributing to liver parenchymal dysfunction in obstructive jaundice. In the findings by the authors, poliferation of fibers occured chiefly around the proliferating bile ductules, and in the high degree in 3 to 4 weeks after the common bile duct obstruction. Although the pseudolobules was formed partly, biliary cirrhosis[3,5] was not recognized within 4 weeks in rats with obstructive jaundice. According to the results obtained by the authors, however, the further persistence of the common bile duct obstruction may possibly develop into biliary cirrhosis.

The authors examined such chronological changes in hepatic fibrosis with the severity score[1]. It was found as a result that the severity score of hepatic fibrosis became higher chronologically after the common bile duct obstruction, and hepatic fibrosis made progress in accordance with the progress of obstructive jaundice, making marked development particularly in 3 to 4 weeks in rats with obstructive jaundice. These findings obtained by the authors are supported by the results obtained by Koyama et al.[3] who studied hepatic fibrosis

quantitatively.

On the other hand, the severity score up to 3 weeks in rats with obstructive jaundice was reduced after biliary drainage, while the score still remained high in 4 weeks. According to this results, in 4 weeks in rats with obstructive jaundice, serious liver parenchymal dysfunction not foreseeable by the standard liver function test may persist after biliary drainage.

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