

## CHAPTER 1

# The background to hepatocellular carcinoma and the liver

It is well known that more than 80% of hepatocellular carcinoma (HCC) occurs in hepatitis virus-related liver cirrhosis and chronic hepatitis; HCC in a healthy liver is rare [1]. Although HCC is most commonly associated with viral hepatitis infection, there are other liver diseases such as alcohol abuse [2–5], hemochromatosis [6,7], parasitic infestation [8], aflatoxin [9,10],  $\alpha_1$ -antitrypsin deficiency [11] and Thorotrast [12] where HCC can be expected to develop. More recently, nonalcoholic steatohepatitis (NASH) has also been spotlighted as a high-risk factor [13]. In particular, patients with hepatitis B virus (HBV)- or hepatitis C virus (HCV)-related liver cirrhosis and chronic hepatitis have been regarded as the most high-risk population for HCC development, and as a result have been carefully monitored. While the association of HBV-related liver cirrhosis is more common in African and Asian countries, HCV-related cirrhosis is more common in Western countries and Japan [14–18]. Over the past decade or so, the morphologic characteristics of chronic liver diseases induced by different hepatitis viruses (B and C) have been shown to vary significantly [19–23].

### Hepatitis virus-related liver cirrhosis

#### Morphologic differences between hepatitis B virus-related and hepatitis C virus-related cirrhosis associated with HCC

The differences in hepatitis B virus-related cirrhosis (type B cirrhosis) and hepatitis C virus-related

cirrhosis (type C cirrhosis) associated with HCC are best appreciated in surgically resected cases. Through studying resected HCCs, it has been discovered that there are three major differences that distinguish type B and type C cirrhosis:

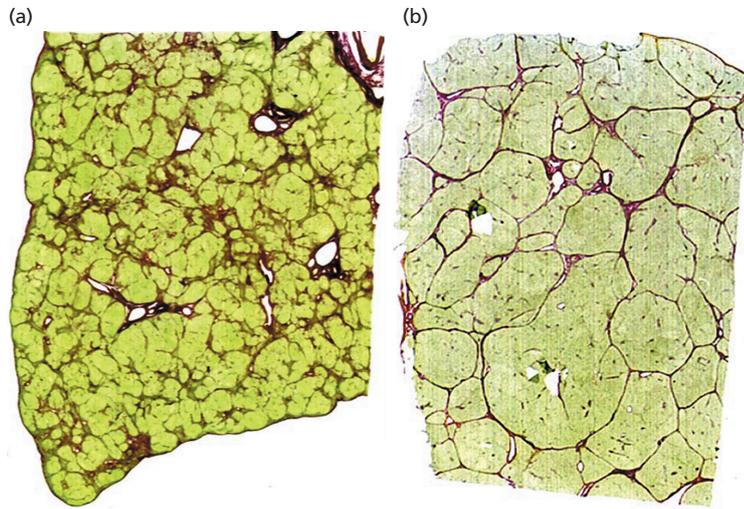
- the size of regenerative nodules
- the degree of fibrosis
- the degree of active inflammation [23]

In autopsy cases, the differences between the two types are less accentuated because the terminal events such as parenchymal extinction due to portal vein thrombosis and pronounced artery–portal vein shunts frequently modify the characteristics of type B and type C cirrhosis.

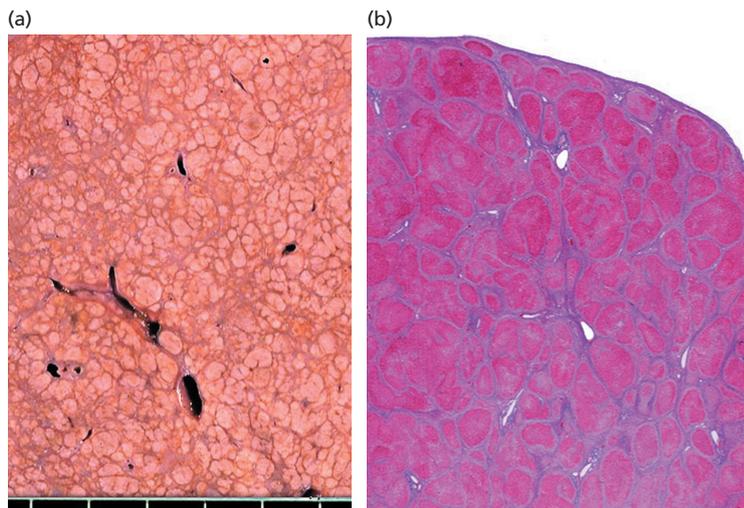
#### Regenerative nodules

The predominant morphologic difference is the size of regenerative nodules. Type C cirrhosis tends to have the features of micronodular cirrhosis, which more resembles alcoholic cirrhosis: the regenerative nodules are about 2–3 mm in diameter, significantly smaller than nodules of type B cirrhosis and irregular in shape (Fig. 1.1a). In contrast, the regenerative nodules of type B cirrhosis are mostly 7–10 mm in diameter and are more regular in shape (Fig. 1.1b). The histometric analysis of surgically resected cases of liver cirrhosis associated with HCC proves that there is a significant difference in the size of regenerative nodules between type B and type C cirrhosis. The mean size of regenerative nodules found in type B cirrhosis was  $40.5 \pm 22.7 \text{ mm}^2$ , and was approximately three times greater than that of type C

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**Fig. 1.1** Difference in size of regenerative nodules between type B and type C cirrhosis. (a) Regenerative nodules in type C cirrhosis are irregular in shape and measure about 2–3 mm. (b) Regenerative nodules in type B cirrhosis are regular in shape and measure about 5–10 mm. (Surgical cases; reticulin stain.)



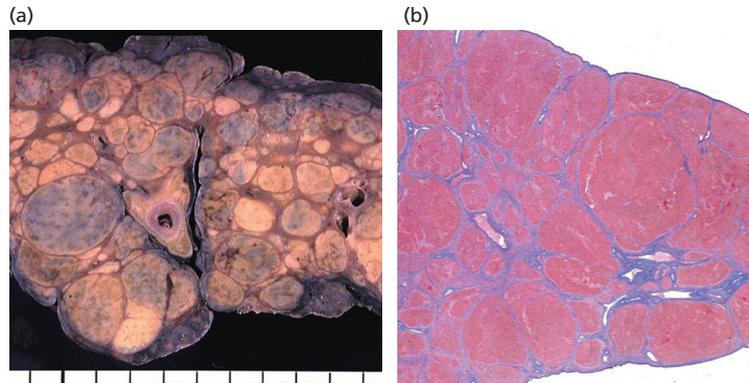
**Fig. 1.2** Autopsy case of type C cirrhosis. (a) Regenerative nodules measure about 2–3 mm and are irregular in shape. (b) Fibrous septa are wider than those of surgical cases. (Azan–Mallory stain.)

cirrhosis,  $16.9 \pm 10.8 \text{ mm}^2$ . These morphologic differences are more evident in surgically resected HCC cases, in which associated liver cirrhosis is in its relatively early stage and liver function is well preserved, compared with those taken from autopsy or explant livers of advanced cirrhotic livers. However, significant differences in the size and shape of regenerative nodules are also observed in many autopsy cases (Figs 1.2 and 1.3). By contrast, these differences are less apparent in advanced liver cirrhosis as a result of parenchymal extinction, which brings about varying degrees of intrahepatic circula-

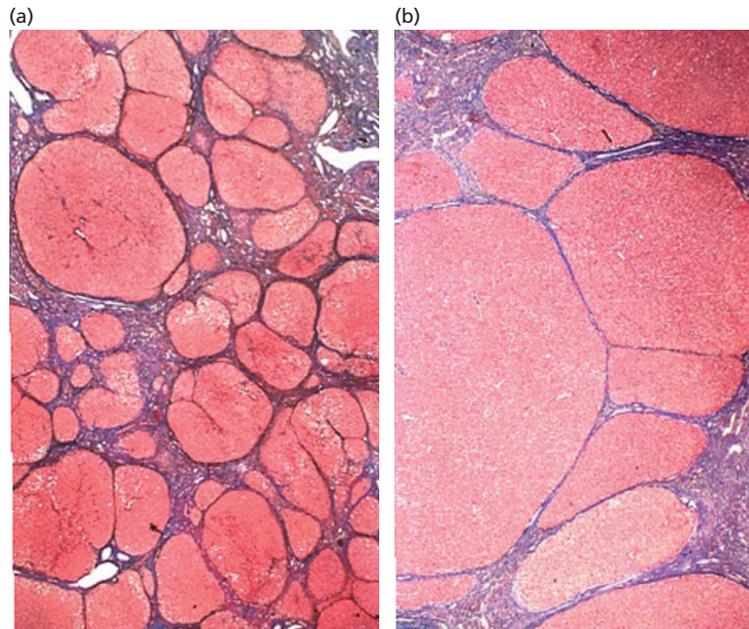
tory disturbance, which in turn modifies the typical morphologic features of type B and C cirrhosis. Many cases of advanced liver cirrhosis tend to show the features of “mixed macro- and micronodular cirrhosis,” in which small and large regenerative nodules are intermixed, although in general terms large regenerative nodules are more dominant in type B cirrhosis and small regenerative nodules are dominant in type C cirrhosis (Fig. 1.4).

Active regenerative features of hepatocytes are more remarkable in type B cirrhosis than in type C. This difference may be explained by the persistent

**Fig. 1.3** Autopsy case of type B cirrhosis. (a) Gross appearance: large regenerative nodules, measuring about 1–2 cm, are observed. (b) Fibrous septa are relatively regular compared with those of type C cirrhosis. (Azan–Mallory stain.)



**Fig. 1.4** Mixed macro- and micronodular cirrhosis in autopsy cases. (a) Small regenerative nodules and wider fibrous septa are more dominant in type C cirrhosis. (b) Large regenerative nodules and thin fibrous septa are dominant in type B cirrhosis. (Azan–Mallory stain.)



active inflammation in the portal areas seen in type C cirrhosis; this inflammatory reaction remains inactive in type B cirrhosis. In other words, persistent active inflammation in type C cirrhosis may disturb active hepatocyte regeneration and therefore does not permit large regenerative nodules to be formed as seen in type B cirrhosis. Meanwhile, strong regenerative activity after subsidence of active inflammation following seroconversion leads to the formation of large regenerative nodules in type B cirrhosis.

Another characteristic difference in the regenerative nodules is the frequent presence in type B cirrhosis of large cell dysplasia (LCD) of Anthony [24] (see Chapter 2, Fig. 2.3). LCD was found in 52.3% of type B cirrhosis bearing HCC but in only 8.1% of type C cirrhosis and in only 3.9% of cirrhotic liver without HCC. Thus, LCD seems to be closely related to the association of HCC, in the context of type B cirrhosis bearing HCC. Some investigators have reported that LCD is directly related to hepatocarcinogenesis, but its role in hepatocarcinogenesis is

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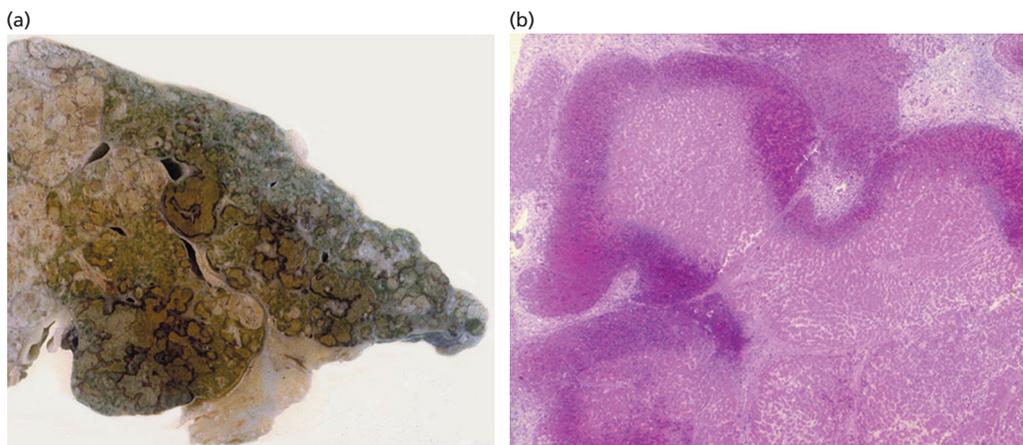
doubtful, and LCD tends to be interpreted as a secondary event following HCC development [25,26]. At present, small cell dysplasia (SCD), which is characterized by small dysplastic cells that have a tendency to form a small round focus, has attracted more attention with its possible relationship to hepatocarcinogenesis [27] (see Chapter 2, Fig. 2.2). Shibata et al. [28] reported irregular regeneration as slight or severe, based on histologic expression of pleomorphism, anisocytosis, bulging, and maplike distribution of hepatocytes in chronic hepatitis or liver cirrhosis (see Chapter 2, Fig. 2.4). They stressed that irregular regeneration is a risk factor for HCC and recommended that liver biopsy be performed in patients with HCV-related chronic liver diseases to detect the risk of developing HCC.

In occasional HCC cases associated with liver cirrhosis, a sudden marked elevation (up to more than 1000 IU/L) of serum alanine aminotransferase (ALT) and aspartate aminotransferase (AMT) is noticed shortly after massive hemorrhage from esophageal varices or elsewhere in the gastrointestinal tract, and the patients die in a short period [29]. In the author's institute, we have experienced 12 such cases (2.7%) among the 439 consecutive autopsy cases. In those cases, the liver shows coagulative necrosis of regenerative nodules clearly demarcated by hemorrhagic rims representing a

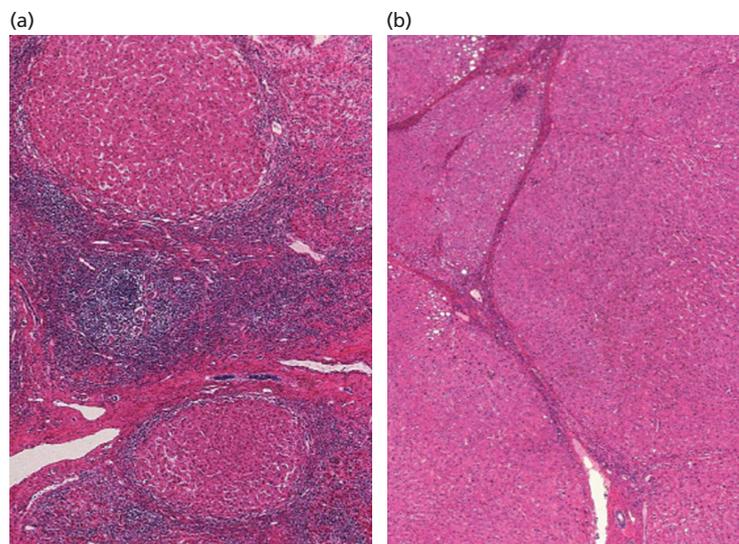
macular pattern (Fig. 1.5). Such a characteristic necrosis of the regenerative nodules is localized in certain areas of noncancerous liver parenchyma, and varying degrees of tumor thrombi in the portal veins are present in all cases. It is presumed that systemic hypotension after hemorrhage and reduced portal blood supply may cause ischemic necrosis of the regenerative nodules.

#### Fibrous septa

Fibrosis tends to be accentuated in the vicinity of HCC, but the difference between type B and C is characterized by the hepatic tissue away from the tumor. Here the fibrous septa are different depending on whether it is type B or type C cirrhosis. In type B, the fibrous septa are relatively thin and regular in shape whereas they appear broad and irregularly shaped in type C cirrhosis (Figs 1.2, 1.3, and 1.4). Histometric analysis showed that the mean ratio of fibrosis area to the parenchyma area was  $22.9 \pm 12.0\%$  (SD) in type C cirrhosis and  $14.5 \pm 5.9\%$  (SD) in type B cirrhosis – the former is twice as great as the latter. Moreover, the broad and irregular fibrous septa in type C are caused by active inflammation, whereas the thin and regular septa in type B cirrhosis are formed by active regeneration of the hepatocytes without active inflammation.



**Fig. 1.5** (a) Ischemic necrosis of regenerative nodules in the left lobe of type C cirrhosis bearing massive hepatocellular carcinoma (HCC) in the right lobe. Necrotic regenerative nodules are demarcated by hemorrhagic rims showing a macular appearance. (b) Histologically, the regenerative nodules show coagulative necrosis and are surrounded by hemorrhage.



**Fig. 1.6** (a) Persistent active inflammation with lymphoid follicle and piecemeal necrosis in the fibrous septa in type C cirrhosis. (b) Inactive inflammatory reaction in type B cirrhosis. (Surgical cases.)

### Inflammation

In about 80% of type C cirrhosis bearing HCC, severe lymphocyte infiltration with frequent lymphoid follicles and a piecemeal necrosis along the fibrous septa is present, but it is observed in only 29% of type B cirrhosis, mostly HBe-antigen positive, and active inflammation is lessened in most cases of type B cirrhosis with seroconversion (Fig. 1.6). In about 90% of type C cirrhosis, focal necrosis and/or single cell necrosis (acidophilic body) and ballooning of the hepatocytes indicating significant necroinflammation are also observed. Continuous active inflammatory features are well reflected in the biochemical data, and most of the patients with type C cirrhosis have fluctuations in the levels of ALT and AMT. One possible reason for such a marked difference in the inflammatory reaction between the two groups is the continuous replication of HCV in the hepatocytes and the host's intense immune reaction in type C cirrhosis. Indeed, it has been reported that the number of virus copies in serum is low but gradually increases along with the progression of disease in hepatitis C patients [30]. It is also reported that the replication of HBV could decrease in type B patients because of seroconversion, and therefore this leads to a reduction in the inflammatory reaction [31].

### Persistent active inflammation and hepatocarcinogenesis

It has been suggested that the presence of a continuous active inflammatory reaction is one of the major causes of the high prevalence of HCC in type C cirrhosis. Sato et al. [32] studied the risk of HCC in 101 patients with cirrhosis by analyzing the mean serum levels of ALT for 15 years. They found that the mean serum level of ALT was significantly higher in 25 patients with type C cirrhosis who developed HCC than the patients who did not develop HCC, but interestingly this was not the case with patients studied in the type B cirrhosis group. In this group, patients who developed HCC and those who did not had similar mean serum ALT levels. Tarao et al. [33] reported the association between high serum ALT levels, more rapid development and higher rate of incidence of HCC in patients with type C cirrhosis. They followed up 93 patients with type C cirrhosis for 5 years for the development of HCC by dividing them into three groups: group A included 33 patients with annual average serum ALT levels that were persistently over 80 IU/L, group B included 41 patients with annual average serum ALT levels that were persistently below 80 IU/L, and group C included 19 unclassified patients. They found that 27 patients (81.8%) of

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group A developed HCC, 17 (63.0%) of whom had multiple nodules. In contrast, only 12 patients (29.3%) of group B developed HCC and only one of them had multiple nodules. Similar results have been reported by others [34–37]. These results suggest a close correlation between hepatocarcinogenesis and sustained necroinflammation in chronic hepatitis C and type C cirrhosis.

### Chronological evolution of the morphologic type of liver cirrhosis associated with HCC

In Japan, HCV infection has been widespread throughout the country since the early 1950s, and the incidence of HCC has seen a sharp increase since the mid-1970s [37,38]. According to a nationwide survey of HCC by the Liver Cancer Study Group of Japan, in the 10-year period from 1968 to 1977, 40.7% of HCC cases were positive for hepatitis B surface antigen (HBsAg), compared with 17.8% in the period from 1990 to 1991. Now, the HBsAg-positive rate in HCC is only about 10% and more than 70% of HCC cases are HCV related. Although the precise reason for such a remarkable increase in HCV infection is unclear, there are some suspected infection routes, such as intravenous drug abuse, which began in the 1950s, tainted blood products obtained from professional blood donors in the early 1960s, and so on [38,39]. In addition to this, a nationwide vaccination program for children to protect against HBV has been implemented since the 1980s, thereby reducing the incidence of new HBV infection.

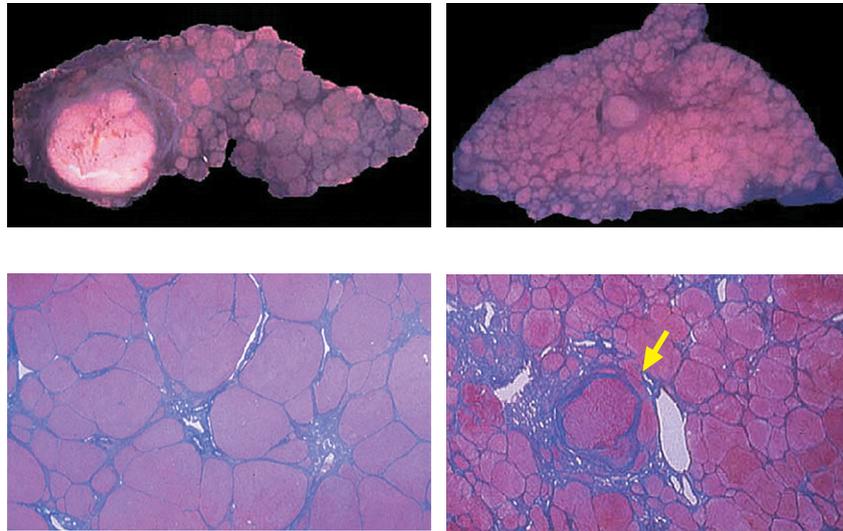
Such a remarkable chronological trend in hepatitis virus infection pattern in Japan is clearly reflected in the morphology of liver cirrhosis bearing HCC. In short, the prevalence of macronodular cirrhosis characteristic for HBV-related cirrhosis plummeted, and micronodular cirrhosis, which is characteristic for HCV-related cirrhosis and used to be infrequent, has become more common over the past two decades. In a comparison of 122 autopsy cases of HCC associated with liver cirrhosis from the period 1970 to 1975 (i.e. before the start of the steep increase in HCC) and 74 autopsy cases from 1989 to 2004 (i.e. after the start of the steep increase of HCC), macronodular cirrhosis (mostly HBsAg

positive) was associated in 39% of the former series and in 12% of the latter series, and 68% of HCCs in the latter series were associated with micronodular cirrhosis (mostly anti-HCV antibody positive). In 1960, 51.2% of a large autopsy series of HCCs were associated with macronodular cirrhosis in Japan. Thus, from the point of view of pathologic features, liver cirrhosis associated with HCC reflects well the chronological evolution of the infection pattern of hepatitis viruses from HBV to HCV predominance over the past four decades in Japan.

### Comparison of HCCs with type B cirrhosis and HCCs with type C cirrhosis

Although the morphology of HCC *per se* is not significantly different between type B and type C cirrhosis, the age of patients and tumor size are significantly different in surgically resected cases. In a comparison of 35 consecutively resected HCCs associated with type B cirrhosis and 100 cases with type C cirrhosis, the mean age of HCC patients with type B cirrhosis was  $50.6 \pm 9.9$  years, and that of the HCCs with type C cirrhosis was  $64.3 \pm 7.3$  years. The HCV-related patients were on average more than 10 years older than the HBV-related patients. This same age difference has been reported worldwide. Such an age difference could be explained by the fact that perinatal or mother-to-infant infections are the commonest means of transmission among HBV carriers whereas HCV is mainly transmitted by blood transfusion, dialysis, intravenous drug abuse, and so on, events that normally happen later in life. Another point to note is that HCC normally appears earlier in HBV patients than in HCV patients. In addition, a different mechanism of hepatocarcinogenesis between HBV and HCV could be attributable.

The tumor size ranges from 1.5 to 11.5 cm with a mean of  $4.5 \pm 2.7$  cm in HCCs associated with type B cirrhosis, and from 0.7 to 15.1 cm with a mean of  $3.1 \pm 1.9$  cm in those with type C cirrhosis. The mean tumor size of HCC with type B cirrhosis is significantly greater than that of HCC with type C cirrhosis ( $P < 0.05$ ) (Fig. 1.7). The prevalence of tumors  $< 3.0$  cm in diameter is 65% in HCCs associated with type C cirrhosis, but only 28% in cases with type B cirrhosis. The difference in tumor size



**Fig. 1.7** Comparison of HCCs in type B cirrhosis (left) and in type C cirrhosis (right). Minute HCC measuring 1 cm in diameter is easily recognizable in type C micronodular cirrhosis. It is predicted that such a minute tumor may be concealed by larger regenerative nodules in type B macronodular cirrhosis. (Surgical cases.)

between HBV-related and HCV-related cases can be explained by the following factors. Patients with type C cirrhosis tend to visit their physicians more frequently than those with type B cirrhosis because of a more rapid deterioration of liver functions due to the presence of persistent necroinflammation, and have more chance of undergoing an imaging examination. By contrast, patients with type B cirrhosis may visit their physicians less frequently because of inactive inflammatory activity due to seroconversion in many of them. Accordingly, HCC should be detected at an earlier stage in patients with type C cirrhosis.

On periodic screening of cirrhotic patients using ultrasonography, minute tumors can be more easily detected in type C cirrhosis, consisting of small regenerative nodules, than type B cirrhosis, characterized by large regenerative nodules. In fact, it has been reported that intrahepatic echograms of patients with type B cirrhosis show irregular hypoechoic masses surrounded by hyperechoic rings designated as a “meshwork pattern,” which reflects the large regenerative nodules [40]. It is presumed that this meshwork pattern is prone to conceal the presence of minute HCC in type B cirrhosis and it seems to be

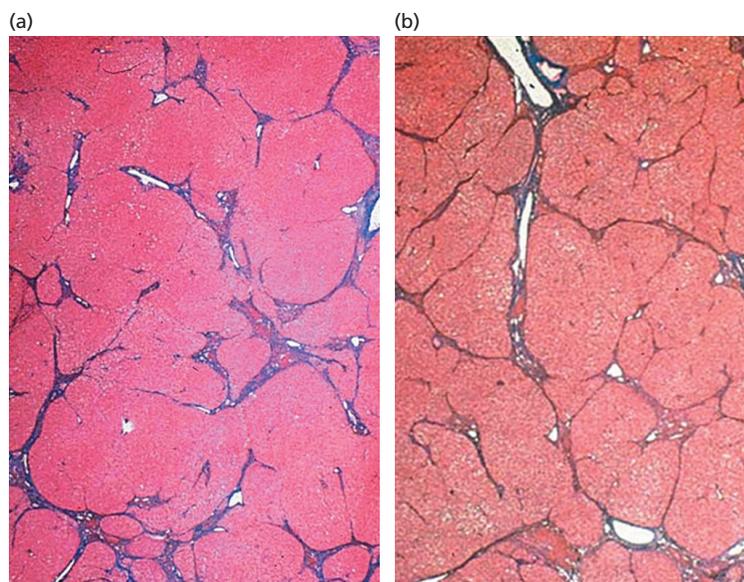
one of the causes of the significantly larger tumor size in surgically resected cases associated with type B cirrhosis.

## Chronic viral hepatitis

### Frequency of chronic viral hepatitis in HCC

Chronic viral hepatitis has a broad morphology ranging from cases with little fibrotic change to cases with advanced fibrosis showing a tendency for lobular distortion (Fig. 1.8). Clinically, a certain proportion of chronic hepatitis seems to be diagnosed as liver cirrhosis due to such morphologic variation. Approximately 20% of surgically resected HCCs in the author's institute were preoperatively misdiagnosed as HCC with cirrhosis, when in fact they were HCC with chronic hepatitis regardless of virus type. By contrast, relatively few resected HCC cases that had been diagnosed with associated chronic hepatitis were interpreted morphologically as liver cirrhosis. Thus, there is a certain discrepancy in the frequency of the association of liver cirrhosis and/or chronic hepatitis in HCC among clinicians and pathologists in these surgical cases. The accuracy of diagnosis of

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**Fig. 1.8** Fibrosis in chronic hepatitis C. (a) Moderate fibrosis without lobular distortion. (b) Advanced fibrosis showing a tendency of lobular distortion. Preoperatively, both cases were diagnosed as liver cirrhosis. (Surgical cases; Azan–Mallory stain.)

associated hepatic lesions may be improved by needle biopsy, but there is still a problem with diagnostic biopsy of chronic hepatitis C, in which fibrotic change is often not uniformly distributed in the liver [19–22].

It has been reported that the frequency of association of chronic hepatitis in HCC is around a few to 25%. In general, HBV-related HCC tends to occur in the stage of chronic hepatitis more frequently than HCV-related HCC. Among the surgically resected HCCs in the author's institute, chronic hepatitis is associated in 43% of HBV-related HCCs and in 30% of HCV-related cases. The frequency of the association of chronic hepatitis in the surgical cases is higher than that of HCCs treated nonsurgically. This can be explained by the fact that relatively small HCCs associated with liver cirrhosis tend to be treated by local ablation therapy whereas cases with chronic hepatitis are more frequently sent for surgery because of milder liver dysfunction.

#### Comparison of HCC with chronic hepatitis B and HCC with chronic hepatitis C

Among surgically resected HCCs associated with chronic viral hepatitis, the mean age is  $53.8 \pm 9.9$  years in HBV-related cases and  $62.5 \pm 6.3$  years in HCV-related cases. Hence, the mean age of HBV-

related HCCs is significantly lower than HCV-related cases as well as those associated with liver cirrhosis. Histologically, about 70% of HCV-related chronic hepatitis cases bearing HCC demonstrate the features of severe chronic hepatitis (active necroinflammation) and moderate to severe fibrosis, whereas the others represent mild chronic hepatitis with mild fibrosis according to a new classification by Desmet et al. [41]. By contrast, only 17% of cases of HBV-related chronic hepatitis bearing HCC represent moderate to severe chronic hepatitis with mild to moderate fibrosis, the others representing mild chronic hepatitis with mild fibrosis. Thus, active inflammation is much more predominant in HCV-related cases as well as in HCCs associated with type C liver cirrhosis.

In a comparison of 94 HCCs with chronic hepatitis representing moderate fibrosis and 156 HCCs with liver cirrhosis among the surgically resected cases in the author's institute, the frequency of small HCC (<2 cm in diameter) is 21.3% in the cases with chronic hepatitis and 33.3% in those with liver cirrhosis. On the other hand, the frequency of advanced HCC (>10 cm in diameter) is 10.8% in the cases with chronic hepatitis and only 2.6% in cases with liver cirrhosis. Thus, HCCs in patients with chronic hepatitis are detected at a more advanced stage than those with liver cirrhosis. In particular,

in chronic hepatitis B the tumors tend to be much larger than in those with chronic hepatitis C. A possible reason for this higher frequency of advanced HCCs in chronic hepatitis patients, particularly in HBV-related cases, is the lack of attention given to HCC development in patients with chronic hepatitis compared with cirrhotic patients. In chronic hepatitis B, inflammatory activity is much milder and the patients tend to have fewer and milder subjective symptoms than those with chronic hepatitis C. Also the trend seems to be that patients with chronic hepatitis B visit their physician less often than those patients who have liver cirrhosis. Morphologically, the most striking difference is found in the gross appearance of small HCCs between chronic hepatitis with mild fibrosis and liver cirrhosis. In the cirrhotic liver, approximately one-third of small HCCs <2 cm in diameter are vaguely nodular in type and consist of very well-differentiated cancerous tissues that are often difficult to distinguish from high-grade dysplastic nodules [42] (see Chapter 3). In contrast, small HCCs in chronic hepatitis with mild fibrosis are found as a distinctly nodular tumor (Fig. 1.9), and the author has not seen small HCC of the vaguely nodular type in any of 14 small HCCs with chronic hepatitis with mild fibrosis. It is suggested that such a difference in gross appearance

may be attributed to a different process of hepatocarcinogenesis in chronic hepatitis with mild fibrosis. Namely, it has been proposed that many HCCs in cirrhotic liver occur via dysplastic nodules in a stepwise process of hepatocarcinogenesis, whereas dysplastic nodules are extremely rare or absent in chronic hepatitis with mild fibrosis and normal liver. In chronic hepatitis with mild fibrosis, many HCCs may be *de novo* cancers without a multistep process of hepatocarcinogenesis.

The majority of small HCCs in the early stages with liver cirrhosis are well-differentiated tumors, as are many small HCCs at an early stage with chronic hepatitis. However, in a comparison of histologic grade of small HCCs <2 cm in diameter with liver cirrhosis or with chronic hepatitis, the prevalence of moderately or poorly differentiated HCC tends to be higher in those with chronic hepatitis. The prevalence of well-differentiated HCC is 50.0% in cases with chronic hepatitis, which is significantly lower than the 75.5% in those with liver cirrhosis ( $P < 0.05$ ). In addition, the prevalence of poorly differentiated cancer is 10.0% in cases with chronic hepatitis, but is only 3.8% in those with liver cirrhosis. Thus, early-stage HCCs occurring in chronic hepatitis with mild fibrosis tend to be less differentiated than those associated with liver cirrhosis. It is predicted that the dedifferentiation process, from well-differentiated to moderately or poorly differentiated cancer, may progress more rapidly in the early stage of HCCs occurring in chronic hepatitis than in those in liver cirrhosis.



**Fig. 1.9** Small HCC in chronic hepatitis. Minute tumor measuring 1.0 cm in diameter shows a distinct nodular appearance although it is not encapsulated. Note the hemorrhage corresponding to the biopsy site in the tumor.

## Other liver diseases

### Alcoholic cirrhosis

The etiologic relationship between HCC and heavy alcohol drinking has been a matter of controversy for many years. Recently, the involvement of hepatitis viruses has been given prominence even in HCCs associated with alcoholic cirrhosis, and it has been suggested that alcohol alone may not play a significant role in hepatocarcinogenesis [43–46]. Brechot and colleagues [43] compared the presence of serologic markers of HBV infection with the presence of the viral DNA in the livers of 20 HCC patients with alcoholic cirrhosis, and found that 9 of

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the 16 tested had serologic markers of HBV infection, but all 20 had HBV DNA integrated into the genome of the neoplastic liver cells. They suggested that HBV plays a part in the pathogenesis of HCC in alcoholics. Bruix et al. [46] studied the presence of HCV antibody in 96 HCC patients in Spain and found that the prevalence of anti-HCV was significantly higher in HCC patients with alcoholic cirrhosis (76%) than in patients with alcoholic cirrhosis alone (38.7%). They also described how HCV infection may have a role in the pathogenesis of HCC, even in patients with chronic alcoholic disease. A high prevalence of anti-HCV in HCC patients with alcoholic cirrhosis has also been reported by Colombo et al. in Italy [47]. By contrast, Walter et al. [48] reported that there was no molecular evidence for a contribution of HBV infection to the development of HCC in alcoholic liver disease based on the study of Southern blot analyses of DNA extracted from HCC tissues with chronic alcohol intake.

In Japan, alcoholic cirrhosis is rather rare, and HCC with alcoholic cirrhosis is extremely rare. Among 750 autopsy cases of HCC in the author's institute, there was no case of HCC associated with typical alcoholic cirrhosis even in patients with a history of alcohol abuse. It used to be reported that HBsAg-negative liver cirrhosis bearing HCC in patients with a heavy drinking habit was morphologically characterized by a mixture of relatively large and small regenerative nodules, and that the small regenerative nodules reflected alcohol abuse. It was also suggested that macronodular cirrhosis in alcoholics may be related to increased lifespan of the patient, which allows conversion of micronodular cirrhosis into a macronodular one [49]. However, it has been clarified that there is a high prevalence of involvement of hepatitis virus in HCCs among alcohol abusers in Japan as well.

### Primary biliary cirrhosis

It is known that the prevalence of HCC is higher among the patients with primary biliary cirrhosis (PBC), especially in advanced stages [50–52]. Jones et al. [50] examined 667 cases of PBC and found 16 HCCs (5.9%), and all of them developed in stage III and IV disease. Nijhawan et al. [51] surveyed 162 cases of PBC at the Mayo Clinic from 1976 to 1985

and found 15 hepatobiliary malignancies including 12 HCCs and three cholangiocarcinomas. They reported that hepatobiliary malignancies had a relative risk of 46 ( $P < 0.0001$ ) for women and 55 ( $P < 0.0001$ ) in men with a remarkably increased risk for the development of HCC. In Japan likewise, HCC is increasingly regarded as a complication of the cirrhotic stage of PBC. Nakanuma et al. [52] surveyed 319 autopsy cases of PBC in the Annual Registry of Pathological Autopsy Cases in Japan from 1977 to 1987 and found 13 HCC cases (4.1%). Although an increasing tendency of HCC in PBC has been suggested, negative data have been also reported [53,54].

### Schistosoma infestation

In chronic schistosomiasis, characteristic fibrosis, including "septal fibrosis" and "pipe-stem fibrosis," is observed in the liver in varying degrees (Fig. 1.10).



**Fig. 1.10** Schistosomal cirrhosis bearing HCC. Liver surface shows a tortoise-shell shape and the portal trunks with tumor thrombus show pipe-stem fibrosis in the cut surface.



Although HCC has been reported to have a geographical prevalence similar to that of schistosomiasis, an etiologic relationship between the two diseases has been controversial. In chronic schistosomiasis japonica, calcified egg nodules are observed in various organs such as liver, lung, intestine, lymph node and so on, but a high incidence of cancer is found only in the liver. In a comparison of cancer incidence in various organs among 229 autopsy cases of chronic schistosomiasis japonica and 4657 controls in the author's institute, the incidence of HCC was four times higher than in controls [55,56]. However, anti-HCV was positive in about 88% of HCCs with chronic schistosomiasis japonica in a formerly endemic area of Japan [57]. Accordingly, the high incidence of HCC in chronic schistosomiasis seems to be related to infection by hepatitis B or C viruses, and it is presumed that schistosomiasis may play a synergistic role in hepatocarcinogenesis.

#### **Aflatoxin B<sub>1</sub>**

Aflatoxins are metabolites produced by the mold *Aspergillus flavus*, and one of the metabolites, aflatoxin B<sub>1</sub>, is well known as an etiologic factor in human hepatocarcinogenesis. The molds grow on grains and food products in humid tropical and subtropical regions, and epidemiologic study and food analysis have also confirmed a high contamination rate of aflatoxin B<sub>1</sub> (approximately 60%) in those regions [58,59]. Thus, the involvement of aflatoxin B<sub>1</sub> in hepatocarcinogenesis is geographically limited. However, because the regions of high aflatoxin contamination are also highly endemic for hepatitis B virus, it cannot be discounted that aflatoxin B<sub>1</sub> contamination and hepatitis B virus infection may play a synergistic role in hepatocarcinogenesis in tropical and subtropical regions.

#### **Hemochromatosis**

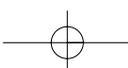
In genetic hemochromatosis, an excessive and progressive iron overload in the hepatocytes eventually leads to micronodular cirrhosis with fairly uniform distribution of iron in the regenerative nodules. It is widely known that HCC is a common complication in genetic hemochromatosis, and that the majority of HCCs are confined to the established cirrhosis [60–62]. Among 850 autopsy cases of HCC at the

author's institute, there was only one case of genetic hemochromatosis with micronodular cirrhosis and associated small HCC.

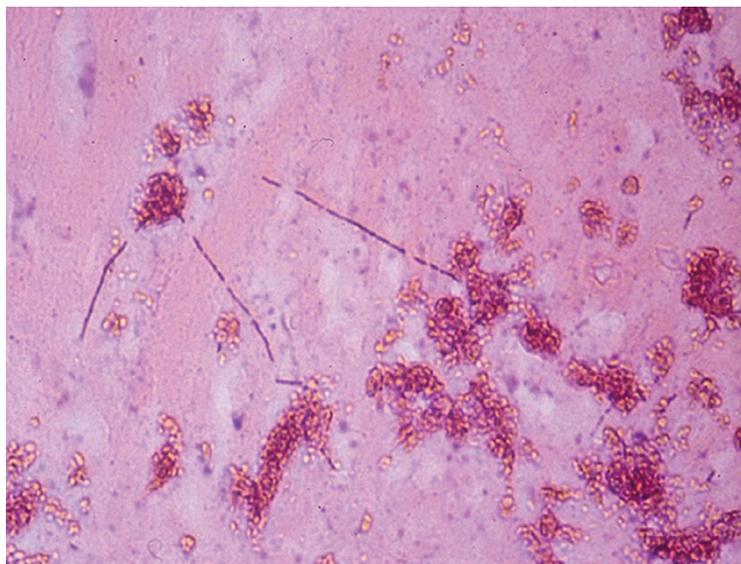
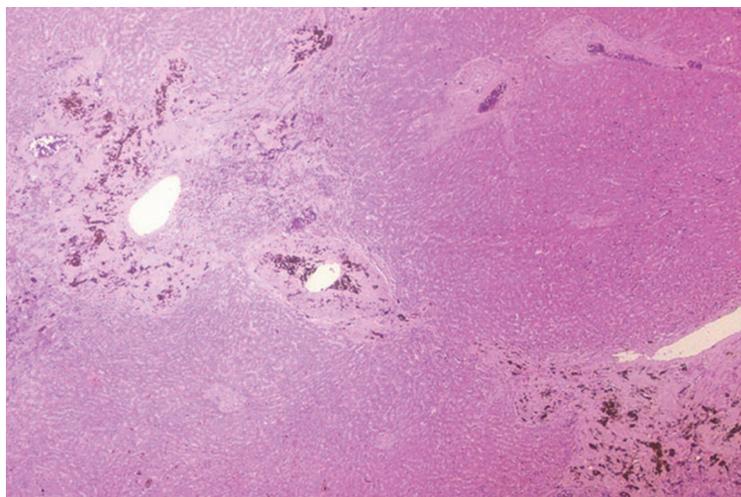
#### **Thorotrast**

Thorotrast, a stabilized 25% colloidal solution of thorium dioxide, was utilized in many countries from the 1930s to the 1940s as a contrast medium for various roentgenographic examinations. This was despite the high risk of carcinogenicity of Thorotrast, particularly in the liver, which had been warned about even at the time of its introduction. Thorotrast was widely used because it lacked acute toxicity and provided excellent radiographic results. In Japan, Thorotrast was used mainly to examine injured soldiers of the old imperial regime. Malignant tumors were detected in many of these soldiers 20–40 years after they were injected with Thorotrast [63]. Varying degrees of irregular fibrosis were observed in the liver, and autoradiography revealed numerous alpha tracts radiating from Thorotrast particles in liver specimens even 30 years after the Thorotrast injection (Fig. 1.11).

Among Thorotrast-induced hepatic malignancies, cholangiocarcinoma and angiosarcoma are the most frequent, whereas HCC is relatively infrequent. The author and colleagues [12] surveyed 102 autopsy cases of Thorotrast-induced hepatic malignancies collected from all over Japan; they consisted of 44 cases (43.1%) of cholangiocarcinoma, 39 cases (38.2%) of angiosarcoma, 16 cases (15.7%) of HCC, 2 cases of combined cholangiocarcinoma and HCC, and 1 case with a combination of all three malignancies. Similar frequencies of hepatic malignancies have been seen in various countries, and therefore it can be assumed that Thorotrast has a specific carcinogenic mechanism. Although it is suggested that Thorotrast may be responsible for certain rare cases of HCC where the hepatitis virus is not involved, no definite scientific data are available to prove it.



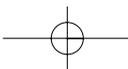
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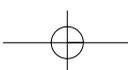
**Fig. 1.11** Liver fibrosis due to Thorotrast. Thorotrast granules (dark-brown pigments) are deposited in the portal tracts with fibrous extension. Autoradiography demonstrates alpha-rays radiating from Thorotrast granules even more than 30 years after the injection. (Autopsy case.)

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