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Neoplasms of the liver

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Primary neoplasms of the liver are composed of cells that resemble the normal constituent cells of the liver. Hepatocellular carcinoma, in which the tumor cells resemble hepatocytes, is the most frequent primary liver tumor, and is highly associated with chronic viral hepatitis and cirrhosis of any cause. Benign tumors, such as hepatocellular adenoma in a noncirrhotic liver or a large, dysplastic nodule in a cirrhotic liver, must be distinguished from well-differentiated hepatocellular carcinoma. Cholangiocarcinoma, a primary adenocarcinoma that arises from a bile duct, is second in frequency. It is associated with inflammatory disorders and malformations of the ducts, but most cases are of unknown etiology. Cholangiocarcinoma resembles adenocarcinomas arising in other tissues, so a definitive diagnosis relies on the exclusion of an extrahepatic primary and distinction from benign biliary lesions.
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A basic principle of pathology is that a neoplasm usually differentiates in the manner of cells that are normally present in the tissue in which the neoplasm arises. Thus, primary neoplasms and tumor-like lesions that occur in the liver usually resemble the major constituent cells of the liver, namely hepatocytes, biliary epithelial cells, endothelial cells, or combinations of these with various mesenchymal cells. A simple classification, based on principal cell type, is shown in Table 1.

In evaluating a tumor in the liver, it is important to remember that metastases are by far the most frequent type of liver tumor, outnumbering primary hepatic malignancies, by as much as 30 to 1, especially in patients without underlying liver disease. Lung, breast, colon, and pancreas are the most common primary sites of hepatic metastases (Figure 1), but malignant tumors from almost any site can at times metastasize to the liver. Melanomas, neuroblastomas, and carcinomas of the gastrointestinal tract, biliary tract, pancreas, lung, and breast often metastasize to liver, while cancers of the head and neck and sarcomas of any site seldom do. In patients with cirrhosis, however, primary liver cancer, especially hepatocellular carcinoma, is relatively more frequent,[1–3] outnumbering metastases by more than 3 to 1 (Figure 1). Among primary liver tumors that come to clinical attention, over three-fourths are hepatocellular carcinoma (HCC), while the second most common primary malignancy, cholangiocarcinoma (CC) accounts for 8% (Figure 2). Therefore, this discussion will focus on these two tumors and their differential diagnosis.

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Hepatocellular carcinoma

One of the most striking features of HCC is the wide variation in its incidence in different parts of the world.[1,4] Overall, HCC is the fifth most common malignancy among men and the eighth most common among women worldwide, but East Asia and Subsaharan Africa have by far the greatest number of cases, and in the countries of those regions HCC is among the leading causes of death. In areas of low incidence, such as the US, carcinoma of the liver (primarily HCC) accounts for only 2.3% of cancer deaths in recent years with an annual incidence of approximately 4.1 per 100 000. It is currently the seventh leading cause of cancer deaths in the US, but the incidence has been rising, more than doubling between 1975 and 1998.[5] In general, regions of the world with a high incidence of HCC are those that have a high prevalence of chronic hepatitis B infection,[6] but even within these areas there is geographic variability; for example, in western Africa the country of Gambia has nearly five times the incidence of Nigeria, suggesting that environmental co-factors such as aflatoxin exposure may also be important. Hepatitis C infection is

Table 1 Abbreviated classification of primary neoplasms and tumor-like lesions of the liver

Benign	*Malignant*
Hepatocellular	*Hepatocellular*
Hepatocellular adenoma	Hepatocellular carcinoma
Focal nodular hyperplasia	Fibrolamellar hepatocellular carcinoma
Dysplastic nodule	Combined hepatocellular-cholangiocarcinoma
	Carcinosarcoma
	Hepatoblastoma
Biliary	*Biliary*
Bile duct cyst	Biliary cystadenocarcinoma
Ciliated foregut cyst	Cholangiocarcinoma
Von Meyenburg complex	
Peribiliary gland hamartoma	
Biliary papillomatosis	
Biliary cystadenoma	
Vascular	*Vascular*
Hemangioma	Angiosarcoma
Infantile hemangioendothelioma	Epithelioid hemangioendothelioma
Other	*Other*
Angiomyolipoma	Primary lymphomas
Mesenchymal hamartoma	Other sarcomas and rare tumors

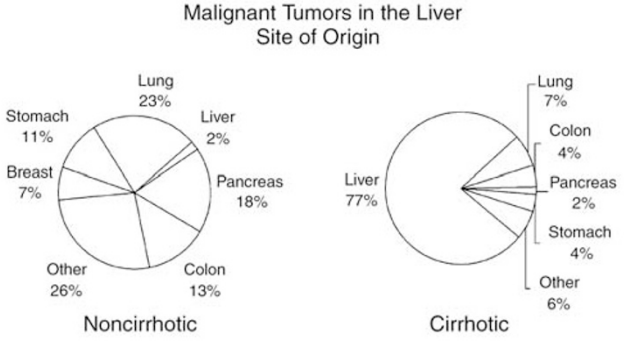

Figure 1 Primary site of origin of malignant tumors found in the liver at autopsy. Left side shows relative frequency in noncirrhotic livers. Right side shows relative frequency in persons with cirrhosis.

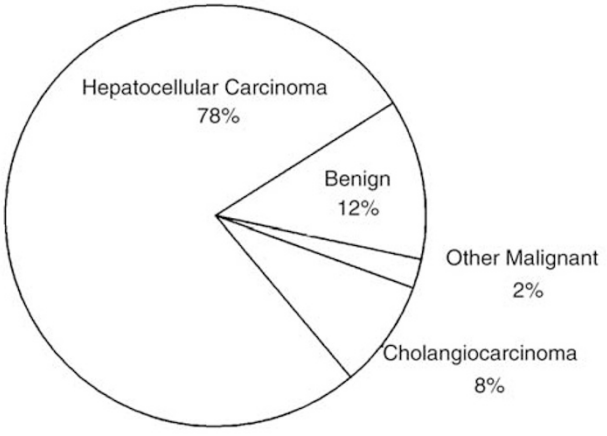

Figure 2 Relative frequency of primary liver tumors in the US.

associated with many cases in countries such as Japan where the prevalence of hepatitis B is intermediate but the incidence of HCC is relatively high.

The incidence of HCC generally increases with age, although there are geographic differences.[4] In Europe and the US the peak age-specific incidence is in the seventh decade, while in Qidong province in China where the incidence is the highest in the world, the peak is in the fifth decade. In South Africa, the average age of patients with HCC is 35 years, and 40% are 30 or younger, whereas in Taiwan (another area of high incidence), the majority of patients are 40–60 years old with a peak incidence in the eighth decade. Nevertheless, HCC can occur in younger individuals and even young children. Regardless of geographic location, HCC occurs more frequently in men than women, with male:female ratios in various countries ranging from 2:1 to 5:1. The precise reason is not known, but it has been shown that many tumors have androgen receptors,[7] raising the possibility that androgens may promote tumor development and growth. There is also a male predominance in risk factors, such as chronic viral hepatitis, alcoholism and smoking, which undoubtedly also play a role.

Risk Factors for HCC and Suspected Etiologies

Cirrhosis is the greatest single risk factor. In various populations, anywhere from 45 to 90% of HCC occurs in the setting of underlying cirrhosis. Cirrhosis of any etiology may be complicated by HCC, but chronic viral hepatitis, especially hepatitis B but also hepatitis C is generally considered responsible for the majority of cases of HCC in the world with up to 100-fold increased risk in patients with chronic viral hepatitis.[6,8,9] There is considerable evidence for a direct effect of the virus on the hepatocyte genome, and also evidence for carcinogenesis as an indirect result of the cycle of inflammation, necrosis and regeneration that occurs in the setting of chronic hepatitis. Aflatoxin B1 contamination of food is common in parts of the world with a high incidence of HCC, and there is evidence that it may be synergistic with hepatitis B in the etiology of HCC in highly endemic areas through the production of mutations in the p53 gene. There is very little evidence that alcohol is directly carcinogenic or genotoxic, but alcoholic cirrhosis is the single most frequent risk factor in the US and many Western countries. Heavy alcohol consumption is associated with a six-fold increase in risk of HCC in cirrhotic patients.[10] Diabetes mellitus is associated with a two to three-fold increase in risk of HCC, regardless of other risk factors, while obesity is associated with a two to four-fold increased risk, both by causing steatohepatitis and cirrhosis.[11] Cases of HCC have occurred in a number of metabolic diseases, but

the strongest associations are with genetic hemochromatosis (GH), tyrosinemia and alpha-1-antitrypsin deficiency. Other metabolic diseases associated with HCC include porphyria cutanea tarda, other forms of porphyria, hypercitrullinemia, fructosemia, Wilson's disease, Byler's disease and Alagille's syndrome. There are many reported cases of HCC in patients taking both anabolic and contraceptive steroids, but the evidence for a direct etiologic effect is less convincing than with other risk factors.[1]

Treatment and Prognosis

It is important for pathologists to have an understanding of the clinical implications of their diagnoses. For HCC, most patients worldwide present with advanced disease and survive for only a short time. For patients diagnosed early, however, treatment of HCC has advanced considerably in the past decade, and cure rates of 35% or higher can be obtained for patients with small tumors detected while still asymptomatic. In general, these are patients with known chronic liver disease and cirrhosis, living in developed countries and receiving careful surveillance, usually with semiannual or quarterly screening with serum alpha-fetoprotein levels and ultrasound examination to detect tumor development.

Many forms of curative and palliative therapy have been attempted with varying success, and optimal treatment strategies are still controversial.[12,13] Surgical resection works best for small tumors in patients with no underlying liver disease. For patients with multiple small tumors and compensated cirrhosis, liver transplantation offers the possibility of curing the underlying liver disease as well as the tumor. As transplantation is not readily available for many patients, percutaneous ablation of the tumor has become the treatment of choice for early but unresectable tumors. Injection of ethanol directly into the HCC under ultrasound guidance causes necrosis of the tumor and surrounding liver tissue. Several types of thermal ablation have been used similarly, with a device inserted into the tumor and radiofrequency waves, microwaves, laser, or cryoablation are used to destroy the tumor and a rim of surrounding nontumor tissue. Of these, ethanol injection and radiofrequency ablation have become the most widely used and have been shown to cure small lesions and prolong survival as a temporary treatment for patients awaiting definitive therapy by resection or transplantation. Radiation, chemotherapy, and hormonal therapy have proven of little benefit in patients with HCC. As HCC receives its blood supply from the hepatic artery rather than the portal vein, angiographic embolization of the artery has been used to produce tumor necrosis and prolong survival. The embolic material may be combined with lipiodol and antineoplastic drugs such as doxorubicin, and this chemoembolization may provide further therapeutic efficacy. Current recommendations for therapy of HCC are surgical resection for patients who are asymptomatic with a single nodule and no evidence of portal hypertension; transplantation or ablation for asymptomatic patients with portal hypertension and single tumors or no more than three nodules all <3 cm in diameter; palliative therapy with chemoembolization for patients with good functional status but large multinodular tumors; and symptomatic treatment for those with advanced or end-stage disease.

Diagnosis of HCC

Although the diagnosis can be fairly certain on clinical grounds in some patients, for example someone with known cirrhosis, a large hepatic mass and a very high serum alpha-fetoprotein, definitive diagnosis usually is made on examination of a biopsy or resection specimen. The biopsy must show that the lesion is malignant and has evidence of hepatocellular differentiation. Microscopically the cells of HCC resemble normal liver cells to a variable extent, depending on the degree of differentiation.[1] Nuclei are usually prominent with prominent nucleoli and a high nuclear:cytoplasmic ratio, and there is usually some hyperchromatism and nuclear irregularity, although this is quite variable. The tumor cells usually have distinct cell membranes and a moderate amount of eosinophilic, finely granular cytoplasm. They may contain a variety of cellular products, mimicking normal and pathologic liver cell function. Bile canaliculi are nearly always present between cells and can often be seen by light microscopy (Figure 3) and demonstrated by immunostaining with polyclonal antiserum to CEA due to the presence of cross-reacting biliary glycoprotein I (Figure 4). Bile pigment may be found in tumor cells or in dilated canaliculi in about 50% of tumors (Figure 5). Cytoplasmic fat is

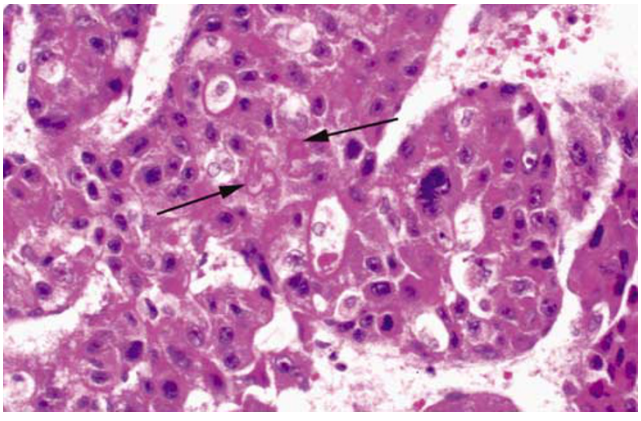

Figure 3 Hepatocellular carcinoma, characterized by large cells with eosinophilic cytoplasm, large nuclei and prominent nucleoli. Intercellular bile canaliculi are present (arrows).

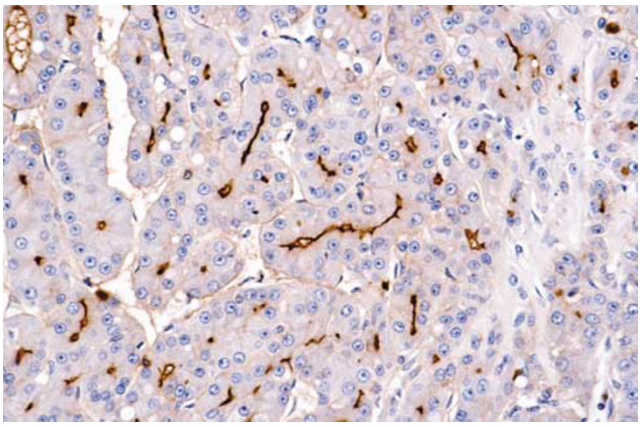

Figure 4 Hepatocellular carcinoma. Immunostain with polyclonal antiserum to CEA demonstrates the bile canaliculi in the tumor due to the presence of cross-reacting biliary glycoprotein I.

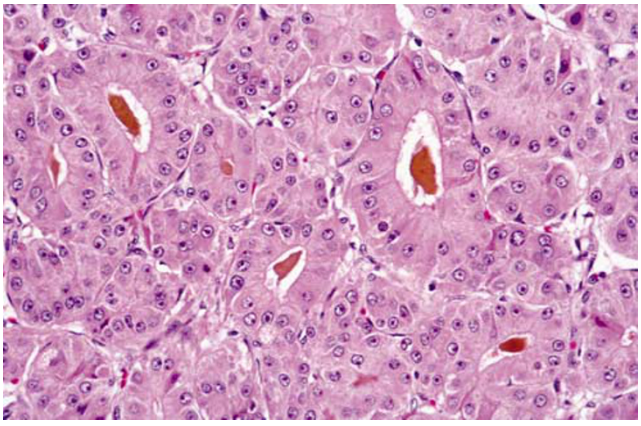

Figure 5 Hepatocellular carcinoma, bile production. Bile is present in dilated canaliculi in the lined by tumor cells.

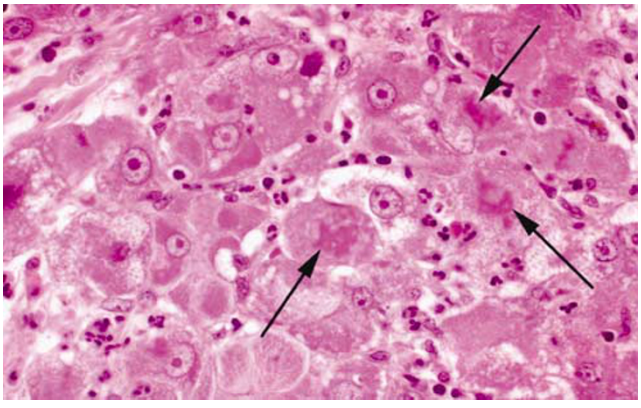

Figure 6 Hepatocellular carcinoma. Cytoplasmic Mallory bodies, identical to those of alcoholic liver disease, are present in cells of this tumor (arrows).

present in at least some tumor cells in about two-thirds of tumors and abundant in about 10%. Large amounts of cytoplasmic fat and/or glycogen can cause the cytoplasm to appear white in routine sections, producing a clear cell appearance. Cytoplasmic Mallory bodies are present in approximately 20% of tumors (Figure 6) and hyaline globules in another 20%, sometimes representing alpha-1-antitrypsin storage, while fibrinogen and other plasma proteins can be demonstrated by immunostains and sometimes form lightly eosinophilic ground-glass-like cytoplasmic inclusions.

The cells of HCC generally try to grow in ways that mimic the cell plates of normal liver, producing well-recognized growth patterns.[1] Most often the tumor grows in a *trabecular* pattern with thickened cords of cells separated by vascular sinusoids, mimicking the cell plates and sinusoids of normal liver (Figure 7). Rapid growth of the tumor cells causes the plates to become thickened and contorted, producing the trabeculae that are surrounded by endothelial cells which phenotypically resemble capillary endothelium (CD34 positive, Figure 8) rather than normal hepatic sinusoidal endothelium (CD34 negative). The centers of the trabeculae may contain very dilated canaliculi, producing a *pseudoglandular* pattern (Figure 9), or the trabeculae may grow together, compressing the sinusoids and forming sheets of tumor cells, producing a *compact* pattern (Figure 10). Connective tissue stroma is typically sparse, and reticulin fibers are absent or reduced, being found only at the periphery of trabeculae. The lack of a desmoplastic stroma is a helpful diagnostic clue and explains why, in contrast to most other malignant epithelial neoplasms, HCC is soft, with a few exceptions. Rarely, tumors with cytologic and phenotypic features of otherwise typical HCC will produce abundant stroma, producing the *scirrhous* pattern of HCC. There is also a subset of tumors that has been named *fibrolamellar* HCC, in which the cells have abundant granular eosinophilic cytoplasm as well as abundant stroma composed of lamellae of collagen (Figure 11). This subset, which differs from other types of HCC in clinical features and prognosis, is the only type with special significance.[14,15] It typically occurs in young people (mean age 23), with approximately equal numbers of males and females, and no association with chronic liver disease, cirrhosis, or any other known risk factor. Fibrolamellar HCC tends to be slow-growing compared to other types, frequently surgically respectable, with a possible better prognosis than other types and 50–70% 5-year survival rates, compared to 5% for other types (particularly in comparison to HCC arising in cirrhosis).

Immunostains may be used to study HCC and to distinguish HCC from other malignancies, especially intrahepatic cholangiocarcinoma and metastatic adenocarcinoma.[1,16] As the cells of HCC attempt to mimic normal liver cells, they may produce any of the cellular products that can be found in hepatocytes both in health and in disease, and if present, these are readily demonstrated by immunostaining. Many of these can also be found in

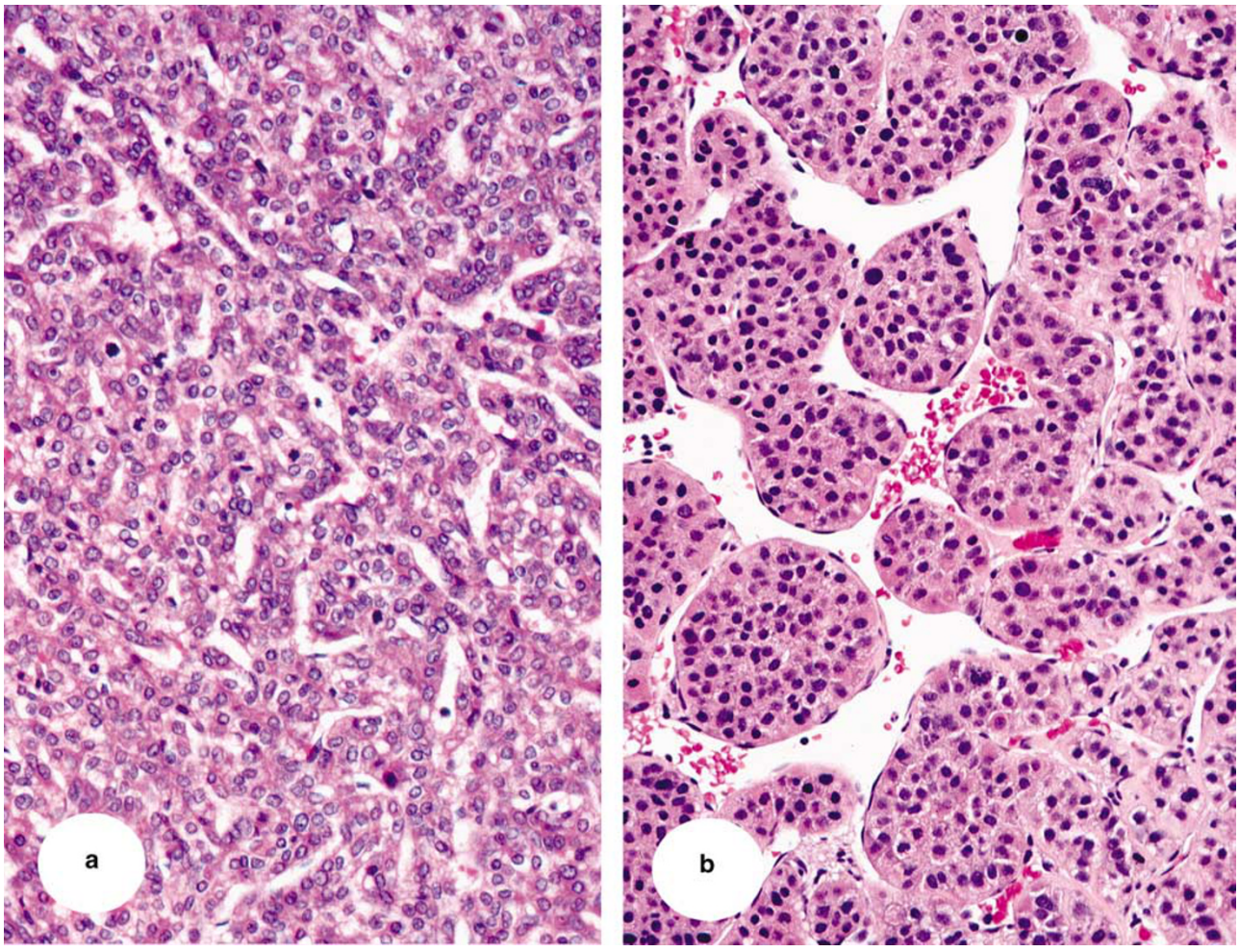

Figure 7 Hepatocellular carcinoma, trabecular growth patterns, with cords of tumor cells separated by sinusoidal spaces that area lined by endothelial cells. (**a**) Microtrabecular pattern with trabeculae that are three to five cells thick. (**b**) Macrotrabecular pattern composed of trabeculae that are ten or more cells thick.

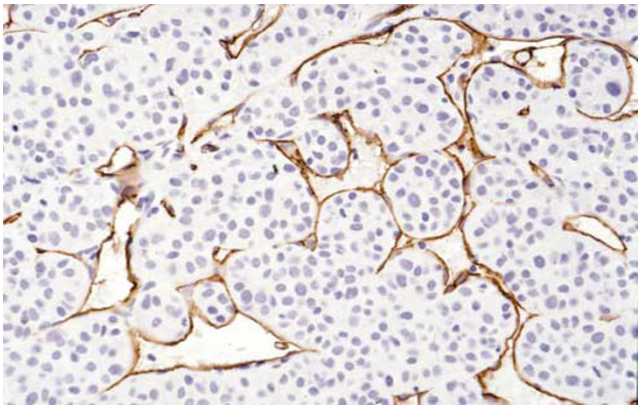

Figure 8 Hepatocellular carcinoma, trabecular pattern. Immunostain for CD34 shows endothelial cells surrounding the trabeculae.

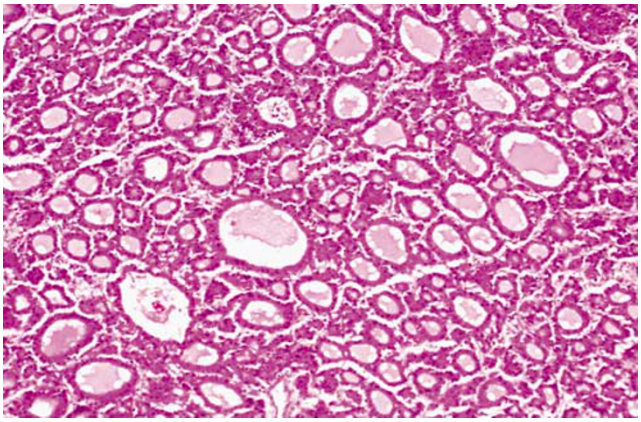

Figure 9 Hepatocellular carcinoma, pseudoglandular growth pattern. There is a dilated canaliculus in the center of the trabeculae so that in cross section each appears to be a gland-like space lined by hepatocyte-like tumor cells.

tumors other than HCC, and so are of little use in differential diagnosis. There are no stains that can consistently distinguish well-differentiated HCC from benign lesions, such as hepatocellular adenoma or dysplastic nodules, and similarly, no single stain can always distinguish poorly differentiated HCC from poorly differentiated cholangiocarcinoma

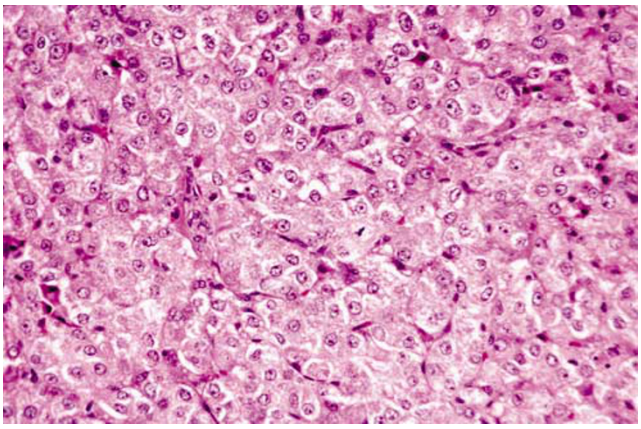

Figure 10 Hepatocellular carcinoma, compact growth pattern. The trabeculae are pressed together with inconspicuous sinusoids, making the tumor appear compact.

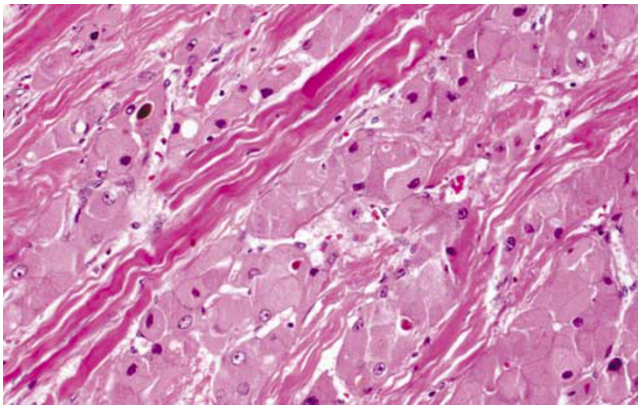

Figure 11 Hepatocellular carcinoma, fibrolamellar type. The tumor cells are large and polygonal with abundant, granular, eosinophilic cytoplasm, and there is a fibrous stroma arranged in parallel lamellae, hence the name fibrolamellar.

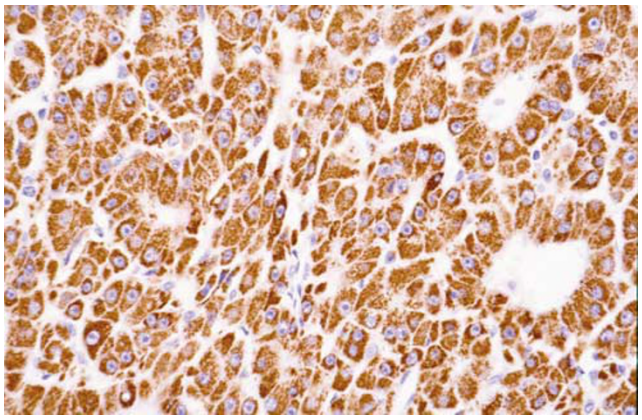

Figure 12 Hepatocellular carcinoma, positive HepPar-1 stain. The tumor cells have a coarsely granular staining pattern.

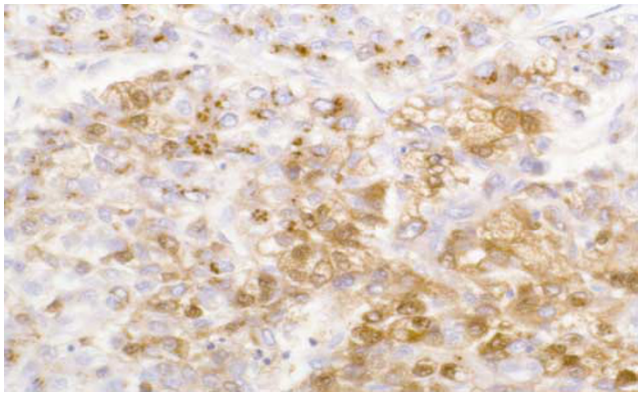

Figure 13 Hepatocellular carcinoma, alpha-fetoprotein stain. Although the tumor is poorly differentiated, there is positive cytoplasmic staining, proving hepatocellular differentiation.

or metastatic adenocarcinoma. However, selected immunostains, taken in the context of other morphologic features, can be very helpful in establishing the diagnosis of HCC in difficult cases. *Polyclonal antiserum to carcinoembryonic antigen (pCEA)* is useful in demonstrating bile canaliculi, both in normal liver and in HCC, due to the presence of a CEA-like cross-reactive substance called biliary glycoprotein I (Figure 4). *HepPar-1 (Hepatocyte Paraffin 1)* is a monoclonal antibody that reacts with an epitope of liver mitochondria, with a typical granular pattern in most liver specimens.[17,18] It also sometimes reacts with renal tubules and intestinal epithelium as well as with intestinal metaplasia in the stomach and esophagus, so it is not completely specific for hepatocytes. It produces positive staining in approximately 90% of cases of HCC (Figure 12) and only 4% of other tumors, including some cholangiocarcinomas and metastatic adenocarcinomas from the stomach and other sites, but when used in the context of morphology, clinical setting and other stains, HepPar-1 is very useful in distinguishing HCC from other malignancies. Approximately 10% of HCC are negative for HepPar-1, and the degree of positive staining varies from case to case; some tumors have a very patchy distribution of positive cells, which can be easily missed in a small biopsy. *Alpha-fetoprotein (AFP)* is frequently elevated in the serum of patients with HCC, even when the tumor is negative by immunostaining, and in most published series it has been found in no more than 50% of tumors (Figure 13), so in general it is less useful than pCEA for diagnostic purposes. *Cytokeratins* are sometimes suggested as a means of tumor classification, especially cytokeratin (CK) 7 and 20, but there are many cases where the tumors display aberrant keratin expression, so staining patterns must be interpreted with caution. CK7 is present in about 50% of HCC, while CK20 can be found in about 20% of tumors, so cytokeratin profiles are not very useful in the diagnosis of HCC. Staining with a pancytokeratin cocktail (AE1/AE3) usually produces negative or weak staining. *CD34* is present and demonstrable in endothelial

cells of large blood vessels and most capillary beds throughout the body with the exception of normal hepatic sinusoidal endothelium. The endothelial cells that surround the trabeculae of HCC are usually positive for CD34 (Figure 8), whereas benign hepatocellular lesions typically have CD34 positive sinusoids only in areas that receive increased arterial blood, so that cirrhotic nodules tend to be positive only around the periphery and focal nodular hyperplasia in sinusoids near the fibrous septa. Consequently, diffuse, regular CD34 positivity of sinusoids can be helpful in distinguishing a cirrhotic nodule from a well-differentiated hepatocellular carcinoma. Staining in hepatocellular adenomas is variable, so that a positive stain does not necessarily indicate malignancy.[16,19]

Differential Diagnosis

A frequent problem in differential diagnosis is in distinguishing poorly differentiated HCC from other malignancies, especially metastases but also from poorly differentiated cholangiocarcinoma (Table 2). A large cell carcinoma with eosinophilic cytoplasm, prominent nuclei and nucleoli may well be HCC but that diagnosis should not be made without definite evidence of hepatocellular differentiation. Bile, canaliculi recognizable on H&E or CEA stain, a positive stain for AFP, a granular staining pattern with HepPar-1, or a trabecular growth pattern allows a diagnosis of HCC with more or less certainty, except for the very rare cases of liver metastasis from a gastrointestinal adenocarcinoma with hepatoid features. If no such evidence of hepatocellular differentiation is found, the tumor is more likely to be metastatic than primary in the liver, especially in the absence of cirrhosis. Cholangiocarcinomas and metastatic adenocarcinomas typically have a desmoplastic stroma, in contrast to HCC, and so a tumor with abundant stroma is almost always an adenocarcinoma, with the exception of the rare fibrolamellar type and even rarer scirrhous type of HCC. Metastatic melanomas (especially amelanotic) as well as carcinoids and some more poorly differentiated neuroendocrine tumors and also renal cell carcinomas may also be confused with HCC, as these all typically have large tumor cells with abundant cytoplasm. If definite features of HCC are not present, these should all be considered and evaluated with a battery of immunostains (S-100 and HMB45 for melanoma, chromogranin and synaptophysin for carcinoids and neuroendocrine tumors, leu-M1 and RCC antigen for renal cell carcinoma).

Table 2 Differential diagnosis of hepatocellular carcinoma from adenocarcinoma, either metastatic or primary intrahepatic cholangiocarcinoma

	Hepatocellular carcinoma	*Adenocarcinoma (metastatic or primary cholangiocarcinoma)*
Stroma	None	Desmoplastic
Growth patterns	Trabecular	
	Pseudoglandular	Tubulo-glandular
	Compact	Papillary
	Fibrolamellar	Solid
	Mixed	Mucinous
Bile	50%	None
Canaliculi	Present	None
Mucin	None	Present or absent
Fat	>50%	None
Mallory bodies	20%	None
Immunostains		
CEA (polyclonal)	Canalicular	Diffuse
HepPar-1	90%	Negative
Alpha-fetoprotein	50%	Negative
Pancytokeratin	Weak or negative	Positive
CK7	50%	Positive
CK8	Usually positive	70%
CK20	20%	40%

Two types of benign lesions enter into the differential diagnosis of HCC (Table 3), hepatocellular adenoma in a noncirrhotic liver, and dysplastic nodules in the setting of cirrhosis.[1,20,21] Hepatocellular adenoma (HCA) typically develops in women in the reproductive age group (15–45 years), nearly always associated with oral contraceptive steroid use. Cases of HCA have been reported rarely in men, children, and women not taking oral contraceptives, but at least some of these are probably misdiagnoses. As hepatocellular adenomas nearly always occur in long-term users of oral contraceptives, any cases outside this setting are highly suspect, and may actually be a different benign lesion such as FNH or a well-differentiated hepatocellular carcinoma. Patients with HCA usually come to medical attention when symptoms develop, with an abdominal mass (25–35%), chronic or episodic abdominal pain (20–25%), and acute abdominal pain due to hemorrhage into the tumor or into the peritoneal cavity (30–40%) frequent modes of presentation. The lesions arise in noncirrhotic livers and may become quite large, up to 30 cm in diameter, although most are 5–15 cm. Microscopically, they are composed of benign hepatocytes arranged in sheets and cords without acinar architecture with cells that are usually larger and paler than nontumor hepatocytes in the surrounding tissue due to increased cytoplasmic glycogen and/or fat (Figure 14). The nuclei of the tumor cells are typically uniform and regular, the nuclear-cytoplasmic ratio is low, and mitoses are almost never seen. A well-developed reticulin framework is usually present in the tumor. The sinusoids, with flattened endothelial lining cells, are usually compressed, thus contributing to the sheet-like appearance. Sometimes the sinusoids are dilated, a finding which can be mistaken for peliosis hepatis. Bile ducts are not found in HCA, but ductules and progenitor cells may be present. The presence of dilated sinusoids and ductules has

caused some tumors to be classified as a telangiectatic variant of focal nodular hyperplasia, but molecular studies have shown these to be a variant of HCA.[22]

Dysplastic nodule (DN) is the term used for a benign lesion that can be confused with HCC.[20,21,23] This encompasses lesions previously called 'adenomatous hyperplasia', 'macroregenerative nodule', and a variety of less popular terms. These are nodules in a cirrhotic liver that are macroscopically distinct from the surrounding cirrhotic nodules. They are usually larger than surrounding nodules and may be detected by imaging studies. They may also differ in color or texture and may bulge from the surface of the liver. Histologic examination is required to distinguish a DN from a small HCC, and they are further classified as low-grade or high-grade, based on morphologic features. Low-grade dysplastic nodules contain portal areas within the nodule, and are composed of liver cells that are minimally abnormal. The nuclear/cytoplasmic ratio is normal or slightly increased, but nuclear atypia is minimal, and there are no mitoses. Steatosis may be present and there may be Mallory bodies. Iron may be increased or decreased compared to the surrounding cirrhotic liver. High-grade dysplastic nodules are characterized by small cell change, which refers to clusters of cells with features that suggest increased cellular proliferation—plates more than two cells thick, pseudogland formation, cytoplasmic basophilia, higher than normal nuclear/cytoplasmic ratio, nuclear hyperchromasia or an irregular nuclear contour. These features often confined to one or more foci within the nodule, giving the appearance 'nodule-in-nodule' formation (Figure 15). It is generally accepted that high-grade DN are precursors of HCC based on several lines of evidence, including morphologic features intermediate between low grade nodules and HCC, the presence of foci of HCC in otherwise high-grade

Table 3 Differential diagnosis of well differentiated hepatocellular carcinoma from benign hepatocellular nodules

	Hepatocellular carcinoma	*Hepatocellular adenoma*	*Dysplastic nodule*
Cirrhosis in surrounding liver	80%	0%	100%
Portal areas	Absent	Absent	Present
Growth patterns	Trabecular Pseudoglandular Compact	Sheet-like	2-cell thick plates Nodule-in-nodule (high-grade)
Nuclear:cytoplasmic ratio	High	Low	Low
Nuclear pleomorphism	Present or absent	Absent	Absent
Nucleoli	Frequent	Absent	Absent
Mitoses	Occasional	Absent	Absent
Vascular invasion	Frequent	Absent	Absent
Stains			
Reticulin	Decreased	Present	Present
CD34	Diffuse	Variable	Periseptal

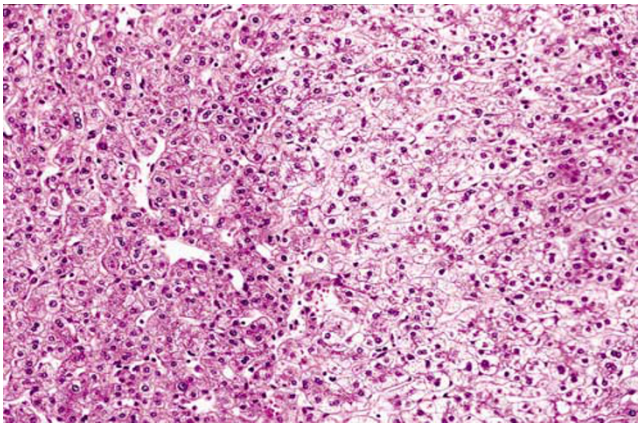

Figure 14 Hepatocellular adenoma. The unencapsulated tumor on the right side of the picture has abundant, pale cytoplasm compared to the nontumor hepatocytes on the left, due to the presence of large amounts of glycogen.

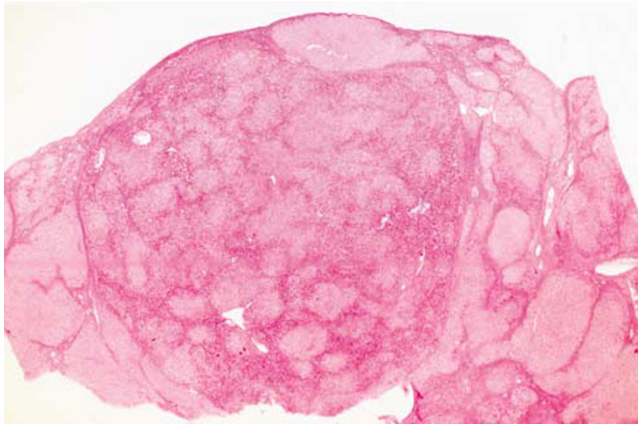

Figure 15 High-grade dysplastic nodule in a cirrhotic liver. The large nodule contains portal areas and cytologically benign hepatocytes, but although they are not surrounded by fibrosis, there are numerous smaller nodules within the large nodule.

dysplastic nodules, and follow-up showing progression to malignancy in a few cases. In some cases it may be impossible to tell a high-grade DN from well-differentiated HCC, especially in needle biopsies. High nuclear:cytoplasmic ratio of the tumor cells is the most reliable sign of malignancy, even when the nuclei are not atypical, and this in combination with trabecular or pseudoglandular growth patterns distinguishes well-differentiated HCC from benign hepatocellular lesions.

Cholangiocarcinoma

Cholangiocarcinoma (CC) is an adenocarcinoma arising from a bile duct. Distinction is often made between those that arise within the liver (intrahepatic or peripheral CC), those that arise at the confluence of the right and left hepatic ducts (hilar CC) and those that arise between the ampulla of Vater and the hepatic hilum (extrahepatic CC). This is justified, since the clinical presentations differ. Hilar cholangiocarcinomas, often called 'Klatskin tumors', produce obstructive jaundice very early in their course and are typically small when they come to clinical attention.[24] Tumors of the extrahepatic ducts may be somewhat larger, but they still present while relatively small. Tumors within the liver, however, may become quite large before they are detected. Histologically, however, the features are very similar among tumors arising in any of the sites, and there is close resemblance to ductal adenocarcinoma of the pancreas.

The incidence of intrahepatic cholangiocarcinoma (ICC), based on evidence from national health statistics and surveys of cancer diagnoses, is reported to be rising in several parts of the world, including North America, Europe, Australia, and Japan, whereas extrahepatic CC has declined slightly.[25–28] In the US, the SEER (Surveillance Epidemiology and End Results) program, which gathers data from population-based tumor registries in 11 parts of the US, found a nearly three-fold increase in the diagnosis of ICC between 1975 and 1999.[27] No cause is apparent, and it remains to be determined whether this is a true increase or merely a change in diagnostic trends brought on by advances in imaging techniques. It is estimated that CC accounts for approximately 3% of all gastrointestinal cancers worldwide and that ICC comprises 10 to 20% of all primary liver cancer.[28] In the US, it is estimated that there are approximately 5000 CC per year, 40% intrahepatic, 7% hilar, and the remainder extrahepatic. In most series, CC is more frequent in men than women (60–70%), and while occasional cases occur in young people, the mean age is in the seventh decade. CC spreads by direct invasion of surrounding tissue and by infiltrating blood vessels and lymphatics. Extrahepatic metastases are found at autopsy in about three-fourths of patients. The median survival is approximately 6 months, with 5-year survival of $<5\%$ for intrahepatic and 10–15% for extrahepatic CC. Surgical resection offers the only hope of cure, but many of the patients present with unresectable tumors. Of those deemed operative candidates, resection with clear margins results in five year survival of 20–40% for extrahepatic and 29–36% for ICC.[29]

Etiology

Possible etiologic factors are much less well established than for HCC, and most cases have no known etiology, but it is clear that disorders that cause chronic inflammation of the biliary tract are associated with an increased incidence of CC.[29] Most cases of ICC arise in a noncirrhotic liver, but there does appear to be an increased risk in cirrhosis, and in some series, patients with cirrhosis due to hepatitis C have approximately 1000-fold increase in risk of ICC compared to the general population.[30] Parasitic infections of the biliary tract by Clonorchis sinensis and Opisthorchis viverrini cause chronic inflammation with adenomatous hyperplasia of the biliary epithelium.[28] These parasites are endemic in Southeast Asia and appear to account for the very high incidence of CC in Thailand and adjacent countries. Hepatolithiasis or the formation of stones in the biliary tract, often with recurrent pyogenic cholangitis, is also much more common in East Asia than in other parts of the world and may account for the relatively high incidence of CC in Japan, where biliary parasites are not highly prevalent. Adenocarcinomas can arise in solitary cysts of the liver, and ICC has been reported in congenital hepatic fibrosis, Caroli's disease, von Meyenburg complexes and polycystic liver disease.[31] Overall, approximately 3% of patients with various forms of cystic dilatations of the bile ducts, including choledochal cyst, will develop CC. Primary sclerosing cholangitis, with or without associated inflammatory bowel disease, predisposes to the development of CC in about 12% of patients.[32] Other reported associations with less definite pathogenetic significance include alcohol consumption, anabolic and contraceptive steroid use, genetic hemochromatosis, alpha-1-antitrypsin deficiency, extrahepatic biliary atresia, and Thorotrast exposure.[1]

Histopathology

Peripheral ICCs may be quite large when discovered, while tumors that occur near the hepatic hilum are usually small and come to clinical attention by causing obstructive jaundice. Histologically, CC may resemble any adenocarcinoma of extrahepatic origin, so that a confident diagnosis depends on the exclusion of other primary sites. The tumor cells are most often arranged in tubules and glands (Figure 16), which may be cribriform, but they can also form nests, solid cords or papillary structures. There is

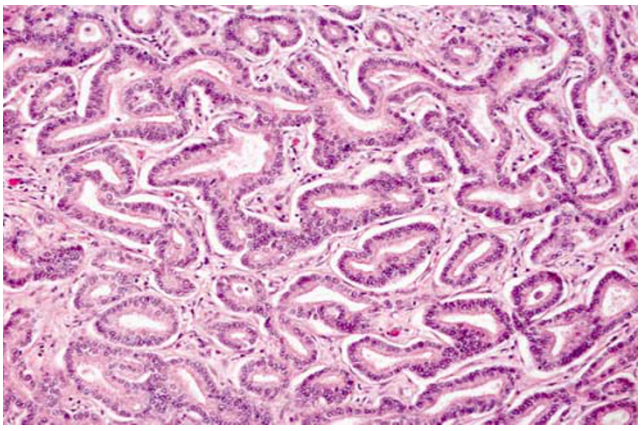

Figure 16 Cholangiocarcinoma. This well-differentiated tumor is composed of cells that resemble biliary epithelium growing in tubules and acini in a desmoplastic stroma.

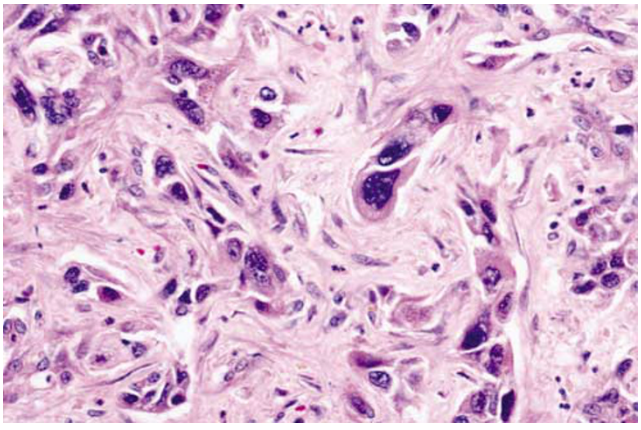

Figure 17 Cholangiocarcinoma. This poorly differentiated tumor is composed largely of infiltrating single cells in a desmoplastic stroma.

typically a fibrous stroma, in contrast to most hepatocellular carcinomas. Most CCs tend to be well-differentiated although poorly differentiated tumors are not infrequent (Figure 17). The well-differentiated CCs are composed of columnar to cuboidal epithelial cells with a moderate amount of clear or slightly granular, lightly eosinophilic cytoplasm. Nuclei are usually small, and typically lack the prominent, eosinophilic nucleoli of cells of hepatocellular carcinoma. The tumor cells can grow along sinusoids and spread intravascularly throughout the liver. Mucin, which may be demonstrable with special stains (Figure 18), is seldom abundant, but occasional tumors are mucinous with copious cytoplasmic and extracellular mucin. Other uncommon variants include mucinous, adenosquamous, clear cell carcinomas and spindle cell or sarcomatoid carcinomas. Numerous antigens have been found in CC, but none specifically can be used to distinguish CC from metastatic adenocarcinoma.

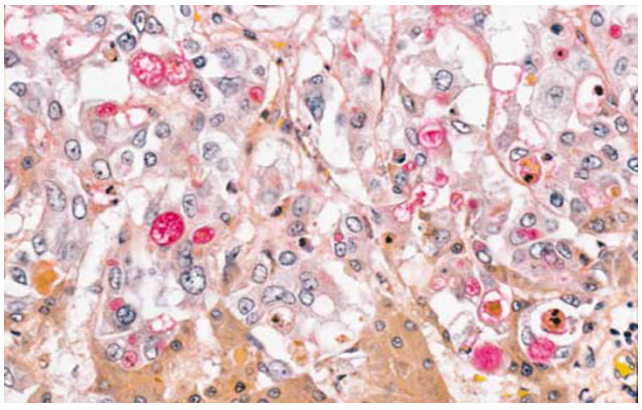

Figure 18 Cholangiocarcinoma, positive mucicarmine stain. This poorly differentiated tumor has intracellular mucin in a number of tumor cells.

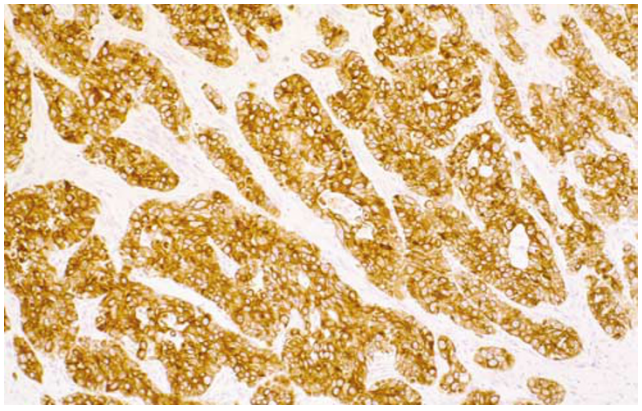

Figure 19 Cholangiocarcinoma, positive stain for pancytokeratin. The gland-forming tumor cells are strongly and diffusely positive.

Differential Diagnosis

Metastatic adenocarcinoma, especially from foregut derived tissues (lung, esophagus, stomach, pancreas) is histologically indistinguishable from CC, and must be differentiated on clinical grounds. Hepatocellular carcinoma may grow in a pseudoglandular pattern, but unlike the true glands of CC, there is no mucin production by the cells lining the pseudoglands. Bile may be present in the pseudogland lumen, which is actually a dilated bile canaliculus that can be demonstrated immunohistochemically by a polyclonal CEA immunostain. Cells of CC show cytoplasmic staining for CEA and are usually strongly positive for cytokeratin (Figure 19) but negative for HepPar-1 (hepatocyte antigen).

Benign biliary lesions can be mistaken for CC, especially when there is inflammation, producing reactive atypia. Peribiliary glands, which are found around the major ducts can be quite atypical when there is inflammation and benign reactive ductular proliferation following inflammation and/or scarring can be extensive and sometimes cytologically atypical. Two types of hamartomatous lesions, the

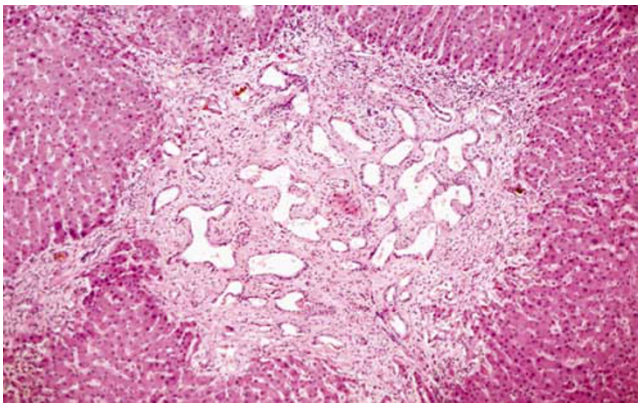

Figure 20 Von Meyenburg complex or biliary microhamartoma. The lesion is composed of irregular duct-like structures lined by low cuboidal epithelium in a fibrous stroma adjacent to a portal area.

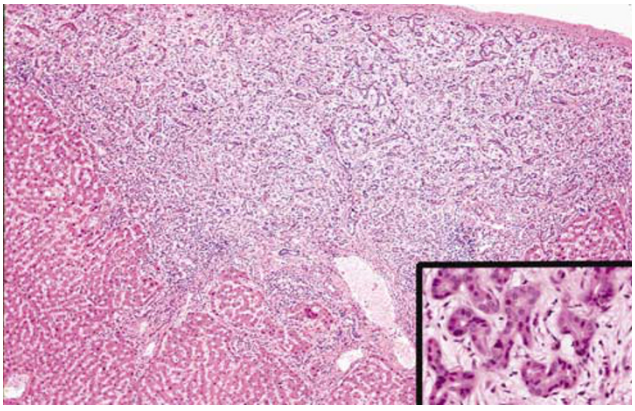

Figure 21 Peribiliary gland hamartoma (also called bile duct adenoma). The lesion is subcapsular and well-circumscribed. It is composed of small acini and tubules in an inflamed fibrous stroma. Inset shows the benign glands at higher magnification.

von Meyenburg complex (biliary microhamartoma) and peribiliary gland hamartomas (bile duct adenomas) can have considerable cytologic atypia when inflamed. Von Meyenburg complexes[33] are typically adjacent to a portal area and consist of a fibrous stroma that contains irregularly shaped duct-like structures lined by a low cuboidal epithelium (Figure 20). The ducts are usually somewhat dilated, often U-shaped or branching, and they may contain proteinaceous or bile-stained secretions. The peribiliary gland hamartoma,[34] also called bile duct adenoma, is a small, well-circumscribed mass composed of small acini and tubules set in fibrous stroma that may be loose and scanty, or dense and hyalinized (Figure 21). There is usually some inflammation, which occasionally is considerable, and normal portal tracts are often identified in the lesion. Histologic features that are helpful in distinguishing well-differentiated CC from the various benign biliary lesions are nuclear size variation and irregularity, mitoses, and cribriform growth pattern.

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