

Useful Algorithms for Histopathological Diagnosis of Liver Disease Based on Patterns of Liver Damage

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Abstract

Previous articles have reviewed the most frequent liver pathologies from the morphological point of view and looked at the importance of adequate correlation for obtaining better understanding between clinicians and pathologists. The next exercise is directed toward histopathological diagnosis of some of the liver diseases for which biopsies are performed most frequently. It takes into account algorithms based on patterns of liver damage.

Keywords

Liver biopsy, algorithms, patterns, portal infiltrates, granulomas, duct reaction, cholangiolar proliferation, lobular injury, pigments, fibrosis, cirrhosis, steatosis, hepatic mass.

PATTERNS OF LIVER DAMAGE

A simple description of what is observed on a microscope slide with a slice of a liver biopsy mounted on it can have very limited diagnostic value. The pathologist must ensure optimal management of the biopsy, proper fixation, and proper processing and must understand additional histochemistry, immunohistochemistry and molecular studies that can add value to the study to finally identify and analyze the type of liver injury present in the tissue sample and to finally make a diagnosis. Fitting the morphological findings of a lesion within one of the patterns of liver damage, always correlated with clinical information, is key to diagnosis.

The liver's response to injury is limited, no matter what cause triggers the damage. It should be remembered that morphological changes are summarized into cellular edema (ballooning), steatosis, cholestasis, necrosis and/or apoptosis, inflammation, regenerative changes and architecture alteration with or without fibrosis. These characteristics, presented in isolation or several simultaneously, give rise to

patterns of liver damage which are distinctly morphological. Each encompasses several differential diagnoses. Analyzed in an algorithmic way, they guide diagnosis. Used together with the clinical history, they can achieve very accurate diagnoses.

Most Important Patterns

- Pattern I, cellular infiltrates
- Pattern II, ductal reaction
- Pattern III, minimal or almost normal changes
- Pattern IV, injury or lobular injury
- Pattern V, steatosis
- Pattern VI, fibrosis
- Pattern VII, presence of masses

The first steps are to identify whether or not there are any alterations of normal lobular architecture and to determine the existence of portal spaces with presence of the normal structures of the portal triad, central vein and uniformly distributed hepatocellular trabeculae (Figure 1).

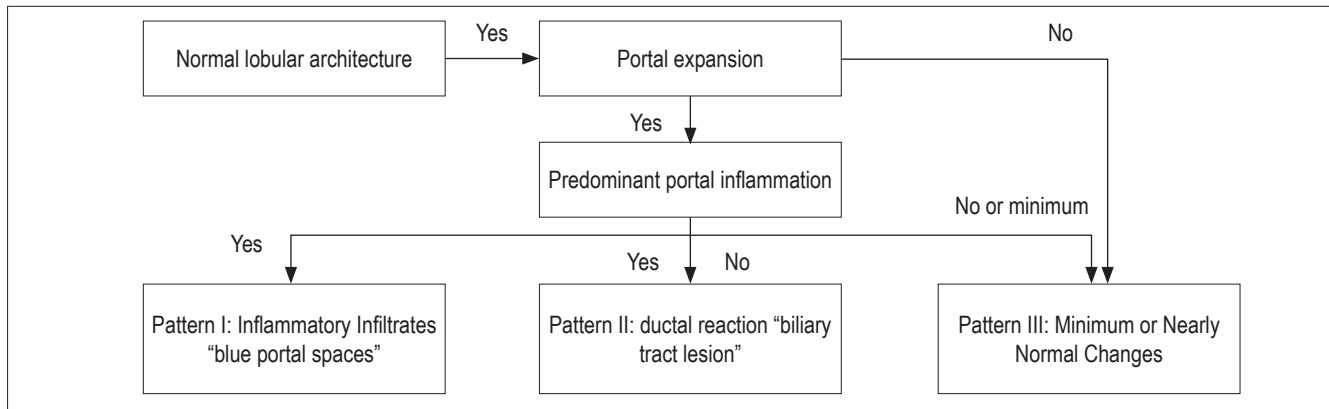


Figure 1. Algorithm of normal lobular architecture which is useful when the architecture of the hepatic lobule is normal.

PATTERN I: INFLAMMATORY INFILTRATES

One of the most frequent findings in hepatic pathology is the presence of portal inflammatory infiltrates which are observed in a wide variety of liver diseases that mainly include necroinflammatory diseases, such as viral hepatitis, autoimmune and drug induced hepatitis. They are also found in chronic cholestatic diseases such as primary biliary cholangitis, primary sclerosing cholangitis, metabolic diseases such as Wilson's disease, α 1 antitrypsin deficiency and neoplastic conditions, such as lymphoma infiltration, leukemia and post-transplant lymphoproliferative syndrome (Figure 2A).

After identifying the inflammatory infiltrates pattern of portal expansion, it is important to consider other keys for histopathological diagnosis such as the type of infiltrate, the pattern of inflammation of the limiting plaque or lobule, association with necrosis and presence of fibrosis. Some of these characteristics were discussed above and will be summarized below.

Inflammation of Portal Lymphocytes

This is observed mainly in chronic hepatitis B and C, acute hepatitis caused by hepatotropic viruses, non-hepatotropic viruses and drug toxicity (Figure 2B) (1, 2).

Chronic hepatitis C

- Inflammation of portal lymphocytes with lymphoid aggregates or follicles with germinal centers
- Interface hepatitis
- Lobular hepatitis with rows of lymphocytes
- Hepatocellular necrosis and/or apoptosis

- Macrovesicular steatosis
- Fibrosis depending on the stage

Hepatitis crónica B

- Inflamación portal linfocitaria
- Hepatitis de interfase
- Hepatitis lobulillar
- Necrosis y/o apoptosis hepatocelular
- Células en vidrio esmerilado

Chronic hepatitis B

- Inflammation of portal lymphocytes
- Interface hepatitis
- Lobular hepatitis
- Hepatocellular necrosis and/or apoptosis
- Ground glass cell appearance

Portal inflammation with plasma cells

Portal inflammation with a predominance of plasmocytes occurs in autoimmune hepatitis, primary biliary cholangitis, primary sclerosing cholangitis, IgG4 disease, transplant pathologies, recurrent diseases such as autoimmune hepatitis and biliary cholangitis, and late cellular rejection rich in plasma cells (Figure 2C). (3-6)

Autoimmune hepatitis

- Portal infiltrate with predominance of very dense plasma cells
- Interface hepatitis
- Formations of hepatic liver cell rosettes
- Lobular hepatitis
- Hyaline globules in Kupffer cells

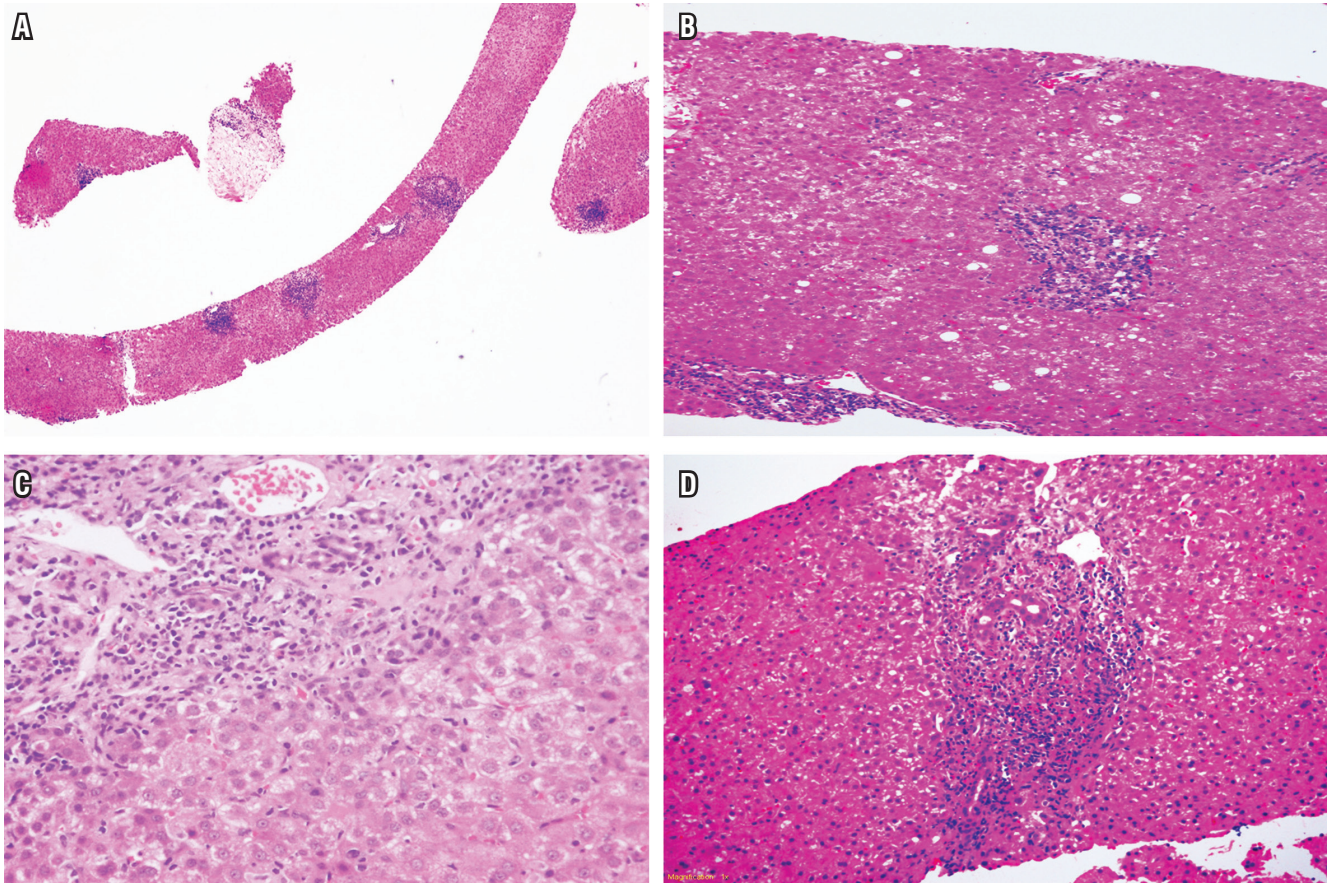


Figure 2. Pattern I, portal inflammation. A. H & E 4x Portal nodule inflammation. B. H & E 10x HCV, portal inflammation with lymphocytes. Fat vacuoles visible in lobule. C. H & E 40x Portal and periportal inflammation with plasma cells in autoimmune hepatitis. D. H & E 20x. Portal inflammation with heterogeneous infiltration of lymphocytes, neutrophils and eosinophils frequently found in acute cellular rejection.

Portal inflammation with eosinophils

There are numerous liver diseases in which eosinophils are present, but when they are predominant they do not necessarily imply toxicity or drug hypersensitivity. Also, systemic conditions with eosinophilia, eosinophilic gastroenteritis or infrequent parasitic diseases such as schistosomiasis of the portal system can produce eosinophilic abscesses. The differential diagnosis also includes some tumors and pseudotumors that compromise the liver such as Langerhans cell histiocytosis, inflammatory pseudo-tumors, myofibroblasts tumors, and post-transplant acute cellular rejection (Figure 2D). (7, 8)

Mixed portal inflammation with lymphocytes, plasma cells and neutrophils or polymorphonuclear eosinophils

This condition is characteristic of acute cellular rejection, Hodgkin's disease, viral hepatitis and drug-induced hepatitis.

Inflammation with granulomas

The prevalence of granulomas ranges from 2.4% to 15%, of which more than 60% is secondary to systemic processes, one third to primary hepatic conditions while 6% to 10% are idiopathic. (7, 9, 10)

Granulomas can be located in the portal region, in the parenchyma, or in both. The location and characteristics of granulomas can guide diagnosis. Table 1 summarizes some of these characteristics.

PATTERN II: DUCTAL REACTIONS

Previously, the term ductal or cholangiolar proliferation was used to refer to the proliferation of small bile ducts in the periphery of the portal tracts, but this pattern not only includes the proliferation of cholangioles, but also changes in the portal stroma, at the interface and in the parenchyma. The combination of these elements helps define diseases that

Table 1. Characterization of hepatic granulomas

Etiology	Characteristics of Granulomas	Location
Autoimmune diseases		
Primary biliary cholangitis (PBC 25% -50%)	Epithelioid cells without caseous necrosis but with multinuclear cells with inclusions	Portal location that is periductal in relation to florid ductal lesion. Lobular granulomas are rare.
Sarcoidosis (12%-30%)	Large, sometimes coalescing fibrotic granuloma (peripheral fibrosis of the granuloma)	Predominantly portal and periportal locations
Infectious		
Mycobacterium tuberculosis (90% in miliary tuberculosis)	Epithelioid cells and giant cells with ring of lymphocytes and caseous necrosis. Tests positive for acid-alcohol resistant bacilli through culture or PCR	Portal and/or lobular
<i>Mycobacterium avium-intracellulare</i> (AIDS, 50% are disseminated)	Accumulations of foamy macrophages. Tests positive for acid-alcohol resistant bacilli.	Portal and/or lobular
Lepromatous leprosy (60%)	Foamy macrophage aggregates. Tests positive for acid-alcohol resistant bacilli	Portal and/or lobular
Tuberculoid leprosy (20%)	Foamy macrophage aggregates with giant cells. Rarely tests positive for acid-alcohol resistant bacilli	Portal and/or lobular
Endemic histoplasmosis in immunocompromised patients	Macrophages, lymphocytes and central epithelioid cells. May have caseous necrosis. Tests positive for histoplasma capsulatum.	Portal and/or lobular
Listeriosis in newborns and immunocompromised patients	Microabscesses with small granulomas	Lobular, occasionally portal
Rickettsiae or Q Fever	Granulomas with fibrin ring or surrounding fat vacuole	Lobular
Schistosomiasis	Granulomas rich in eosinophils, with presence of fibrosis surrounding the eggs of the schistosomes	Periportal and sinusoidal lobules portal
HCV (1% -10%) of new onset or after treatment with interferon	Small epithelioid cell granulomas without necrosis	Any location
Medications or chemicals (10%)	Usually accompanied by other lesions, such as cholestasis, steatosis or hepatitis.	Any location, most frequently portal accompanied by moderate to marked lobular inflammation
Malignancies		
Hodgkin's and non-Hodgkin's lymphomas, renal cell carcinoma	Granulomas without necrosis Granulomas with fibrin ring	Any location
Idiopathic (3% -37%)	Granulomas without necrosis, usually small	Any location
Lipogranulomas	Histiocytes and macrophages surrounding triglyceride vacuoles. Small and without necrosis	Lobular, associated with hepatic steatosis
Foreign body granulomas of talc, silicone, or oil after surgical or radiological procedures	Inclusions of foreign material in the cytoplasm, refractive and visible with polarized light.	Portal, venous

are characterized by prominent ductal reactions. It should be remembered that hepatic progenitor cells react to damage to bile ducts and hepatocytes and that interface biliary metaplasia of hepatocytes represents a regenerative process. When there is an important ductal reaction, it is related, in the first instance, to acute or chronic biliary tract diseases. (11, 12)

Prominent Ductal Reaction

Acute or chronic biliary tract blockage due to stones, parasites, fibrosis, or masses does not usually require a

biopsy because the clinical and/or imaging diagnosis is usually clear. Blockages are consistently found in post-transplantation biopsies and during autopsies. Other causes of ductal reactions are ascending cholangitis, slow cholangitis, acute hepatitis with submassive or massive necrosis, and chronic cholestatic diseases such as primary biliary cholangitis and primary sclerosing cholangitis. Biliary ductal reaction is defined as the proliferation of bile ducts accompanied by neutrophils and is divided into three types: Type I is associated with biliary obstructions, Type II is related to active hepatitis, and Type III is asso-

ciated with massive hepatic necrosis. This terminology is only useful for guiding diagnosis of possible etiologies (Figure 3). (13-15)

Acute biliary obstruction

- Proliferation of ducts that appear to anastomose with small lights
- Marked edema of the surrounding stroma
- Variable neutrophil infiltrate
- Variable hepatocellular and ductal cholestasis
- Biliary infarction in severe obstructions

Ascending cholangitis

- Proliferation of cholangiocytes with presence of intraluminal neutrophils
- Acute inflammation of the ductal epithelium
- Dilatation of the duct with attenuation of the epithelium
- Variable hepatocanalicular cholestasis
- Periportal biliary infarctions are common

Slow cholangitis

Slow cholangitis occurs in patients with impending sepsis or sepsis, obstruction of the biliary tract or as the result

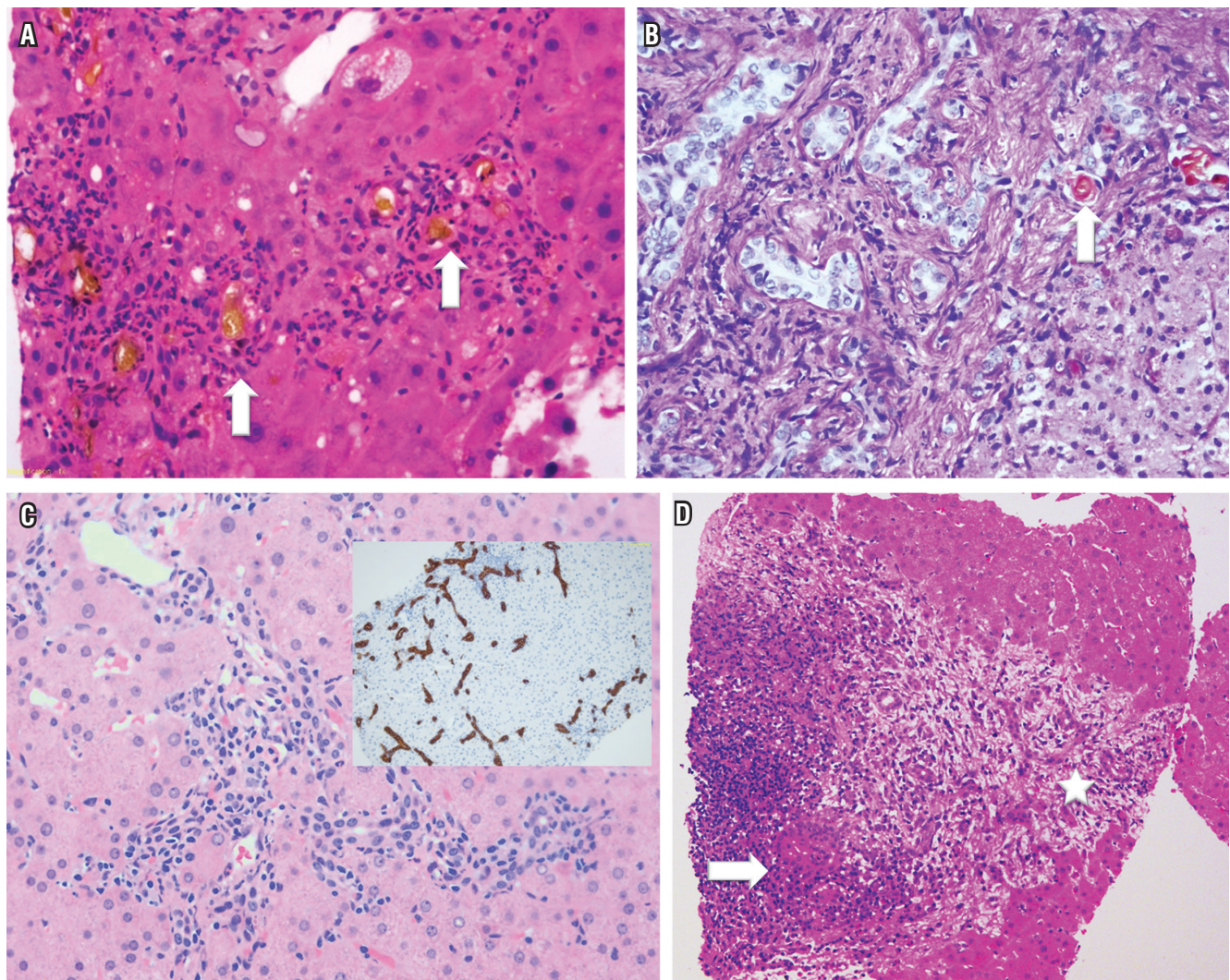


Figure 3. Pattern II, ductal reactions A. H & E 40X. Prominent ductal reaction in acute cholestasis. H & E 20X. Inflammation with neutrophils can be observed in the portal space and biliary plugs. B. PAS 40X. The proliferation of intralobular bile ducts is more evident. White arrows identify some of the bile plugs present in the microphotographs. C. H & E 20X. Ductal reaction prominent in chronic cholestasis. Portal space with marked proliferation of intralobular bile ducts, absence of bile pigment. The immunohistochemical study for cytokeratin 7 in the box showing brown ductal proliferation corroborates this (left). D. H & E 20x. Prominent ductal reaction in chronic primary biliary cholangitis. Observe the ductal proliferation (asterisk) and inflammation of the ductal lesion (arrow).

of endotoxins. It is considered a medical emergency and is characterized by ductal proliferation with dilated ducts filled with bile (ductal cholestasis), bile concretions in the portal space or interface zone and minimal inflammation.

Acute Hepatitis with Massive or Submassive Necrosis

- Prominent ductal reaction due to activation of the stem cells for regeneration of parenchyma and restoration of its function
- Minimal edema
- Mild inflammation, usually due to neutrophils
- Macrophages with PAS material which tests positive for diastase

Chronic Obstruction and Chronic Cholestasis

- Cholangiolar proliferation
- Minimal edema
- Slight inflammation due to lymphocytes
- Effect of clear halo in periportal or septal region
- Hepatobiliary xanthomatosis (cholestasis)
- Periportal copper deposit
- Periductal fibrosis

PATTERN III: NEARLY NORMAL WITH MINIMAL CHANGES

About 5% of biopsies have no specific changes but may have minimal portal or lobular inflammatory infiltrates. There is no evidence of fibrosis or structural changes. In 80% of these patients, there are persistent anomalies in liver function tests and 20% have unexplained ascites which requires a biopsy.

It is possible to define an etiology for approximately half of the patients who have adequate clinical histories and para-clinical studies. These include hepatic compromises due to systemic autoimmune conditions such as systemic lupus erythematosus and rheumatoid arthritis (15%), autoimmune hepatitis, primary biliary cholangitis (10%), metabolic syndrome (10%), chronic inflammatory conditions of the gastrointestinal tract (5%), hepatic ischemia (1% -5%) and non-cirrhotic portal hypertension (1%). Many of these pathologies are heterogeneous and focal including chronic cholestatic diseases and fatty liver disease. Others have periods of latency and inactivity during their clinical courses.

In some cases, especially in entities that cause non-cirrhotic portal hypertension, vascular changes are subtle and focal. Sinusoidal dilation, dilated portal vessels or those with fibrosis can easily pass unnoticed. In 25% of cases, functional abnormalities will normalize and in another 25% they will persist for many years. Table 2 summarizes probable diagnoses and describes morphological changes found when specific lesions are not identified in the biopsy and when a liver biopsy appears to be normal or almost normal (Figure 4). (7)

PATTERN IV: LOBULAR DAMAGE

Lobular Inflammation and Damage

Figure 5 shows an algorithm for normal lobular architecture and portal expansion requiring search for lobular injury or steatosis.

When a lobule is viewed under low magnification, the lobule looks restless. This is due either to occupation by inflammatory cells or because of necrosis which would indicate an injury or lobular damage. There are multiple

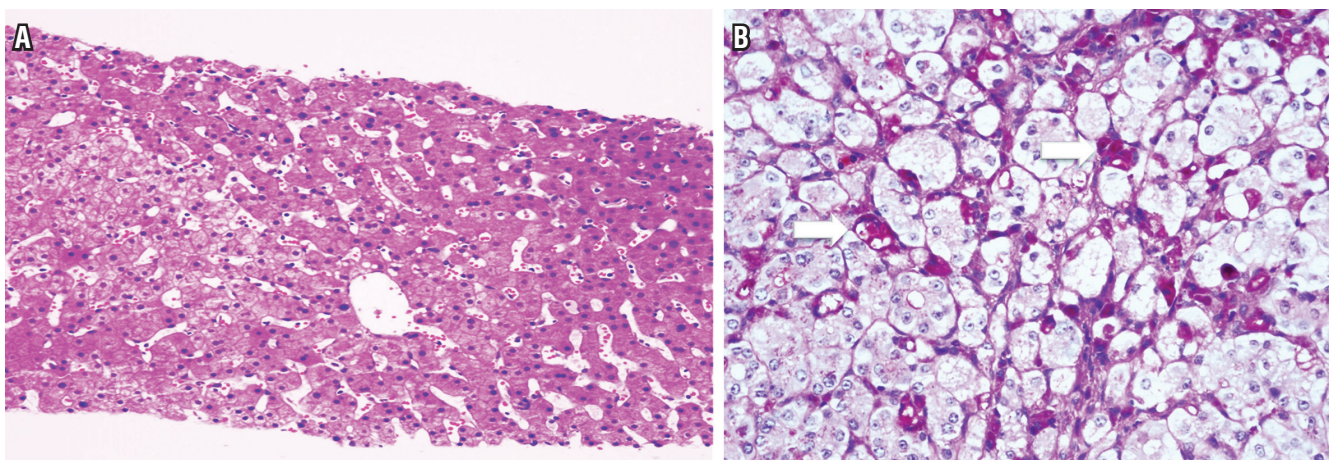


Figure 4. Pattern III, sinusoidal changes. A. H & E 10x shows sinusoidal dilatation in zones 2 and 3 without evidence of inflammation in a patient with non-cirrhotic portal hypertension indicating possible obstruction of the venous flow. B. PAS diastase 40x. Presence of PAS positive material resistant to digestion with diastase in Kupffer cells. In cases of reactive hepatitis, this is sometimes the only finding observed.

Table 2. Characteristics of Pattern III, where minimal changes are observed or is a biopsy that seems normal

Location	Associated Conditions	Morphological Changes
Sinusoidal Changes	<p>Hepatitis in resolution: idiosyncratic drug effect, self-limiting viral hepatitis</p> <p>Sinusoidal dilation: hypercoagulability syndromes and venous flow abnormalities (polycythemia, Waldenström macroglobulinemia, multiple myeloma, medications, autoimmune diseases, hepatic ischemia)</p>	<p>Minimum lobular and/or portal inflammation. Pigmented macrophage accumulations, PAS-diestase staining positive. Regenerative changes with binucleation and discrete thickening of the hepatocyte trabeculae. No sign of chronicity.</p> <p>Subtle changes, minimal or no inflammation, varying degrees of dilation and sinusoidal congestion mainly in zone 3, fibrosis of the central vein which may be very imperceptible.</p>
Changes of Hepatocytes	<p>α1-antitrypsin deficiency</p> <p>Glycogenic hepatopathy (diabetic patients)</p> <p>Megamitochondria typical of NASH, chronic cholestatic diseases, medications, mitochondrial metabolism defects, urea cycle failures and other metabolic defects</p> <p>Ground glass cytoplasmic changes due to medications or immunosuppression</p> <p>Cytoplasmic induction due to proliferation of the endoplasmic reticulum or medications</p> <p>Minimal soft cholestasis due to medications, recurrent benign cholestasis, hypothyroidism and hyperthyroidism, intrahepatic cholestasis of pregnancy</p>	<p>PAS-diestase positive blood cells in periportal locations, sometimes very small and poorly formed.</p> <p>Discreet increases of hepatocyte size, pale-looking cytoplasm</p> <p>Normal architecture, no inflammation. Small rounded inclusions of intense pink color in the cytoplasm of the hepatocytes (negative PAS)</p> <p>Amphiphilic inclusions occupying cytoplasm of hepatocytes</p> <p>Hepatocytes with changed cytoplasm coloration, amphiphilic appearance or 2-color hepatocytes made more evident by PAS</p> <p>Minimal focal intracanalicular cholestasis which requires careful and careful observation</p>

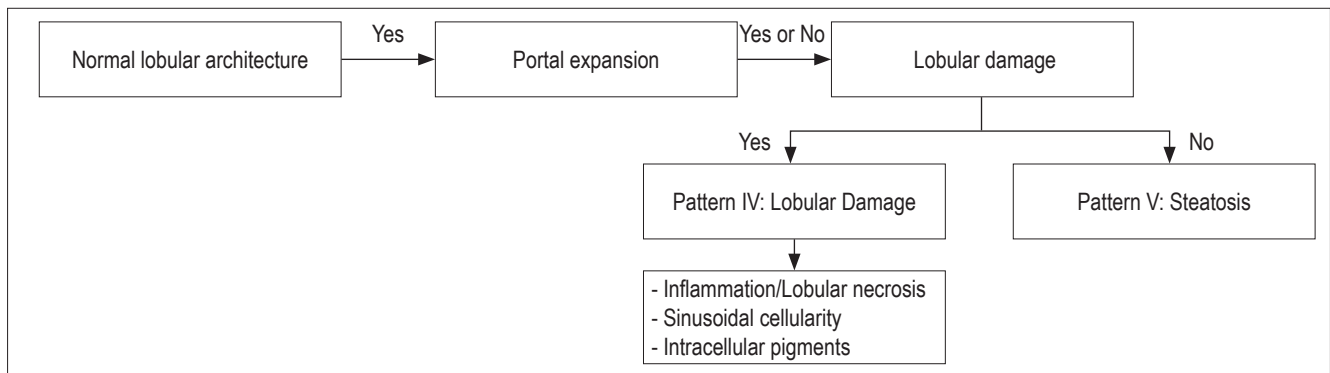


Figure 5. Algorithm for samples from normal lobular architecture with portal expansion and presence of lobular injury or steatosis. This is useful for the analysis of a biopsy without architectural changes but with portal inflammation and lobular alterations.

possibilities which require detailed study with greater magnification to find additional keys to guide later diagnosis together with the medical history.

Some examples of this pattern are acute hepatitis; subacute hepatitis; non-hepatotropic hepatitis such as cytomegalovirus, herpes virus, adenovirus, Epstein-Barr and hepatitis E; compromises due to systemic diseases such as Celiac disease, lymphomas, leukemia, hypervitaminosis A and when pigments of lipofuscin, copper or iron are found. (16-20)

Acute Hepatitis

A biopsy is normally not indicated for acute hepatitis except in rare cases such as when there is doubt or error in the clinical diagnosis and when, despite a well-established diagnosis, the clinician needs more accurate information on the stage or severity of the disease, and following a transplant. Moderate and severe cases of acute hepatitis show prominent lobular inflammation, mild lymphocytic portal inflammation, hepatocellular edema (ballooning), isolated

necrosis or apoptosis dispersed in the lobule, acidophilic bodies, accumulations of macrophages with material that is PAS-diastase positive and hepatocanalicular cholestasis (Figure 6).

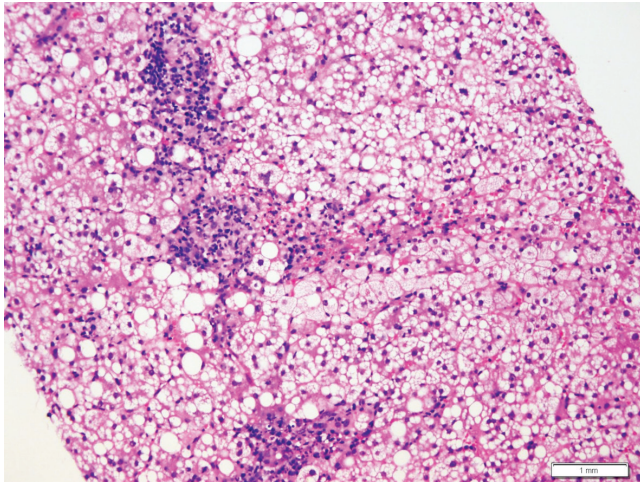


Figure 6. Pattern IV, lobular inflammation and injury. Acute hepatitis shows significant cellular edema, lobular and portal inflammation and occasional acidophilic bodies.

Non-Hepatotropic Viral Hepatitis

Cytomegalovirus (CMV)

CMV occurs in immunosuppressed patients and presents with portal and lobular inflammation with lymphocytes, neutrophils, microabscesses of polymorphonuclear neutrophils located in the lobule, and viral inclusions of CMV without zonal distribution.

Herpes Simplex

Hepatitis secondary to infection with herpes simplex virus is similar to CMV hepatitis, is rare, and is seen in immunosuppressed patients. It produces inflammation and scattered patchy necrosis in the lobule and viral or nuclear amphiphilic or eosinophilic inclusions with a peripheral ring due to chromatin margination.

Adenovirus

Adenovirus infections are rare and usually fatal and in immunosuppressed patients. The virus is similar to herpes virus, and infections present large areas of parenchymal involvement with necrosis but without zonal distribution, lymphocytic inflammation at the extremities of the necrosis zones and presence of adenovirus inclusions.

Sinusoidal Cells

An increase of cells in the sinusoidal space can be a subtle finding that requires analysis with high magnification.

When sinusoidal cellularity is very noticeable, one must think of a serious infection or neoplasia.

Celiac disease, Epstein-Barr hepatitis, lymphoma, leukemia, extramedullary hematopoiesis, hypervitaminosis A and lipidosis of stellate cells are the most frequent reasons for increased sinusoidal cellularity.

Celiac Disease

Up to 30% of patients at the time of diagnosis of Celiac disease have compromised livers. Findings range from normal appearance to lobular hepatitis and changes suggestive of autoimmune diseases such as autoimmune hepatitis, biliary cirrhosis and primary sclerosing cholangitis associated, with or without fibrosis. (19) Sinusoidal lymphoid inflammatory cells, scattered apoptotic cells, mild portal inflammation of lymphocytes, changes in fat and nodular regenerative hyperplasia are likely.

Epstein-Barr Hepatitis

Epstein-Barr hepatitis is most frequent among immunosuppressed patients, but it can also occur in apparently healthy young individuals. (20, 21) It is characterized by numerous sinusoidal lymphoid inflammatory cells simulating a predominantly lobular mononucleosis-like syndrome, small epithelioid cell granulomas with fibrin rings, absence of apoptosis or very occasional and dispersed apoptosis, mild lymphocyte portal inflammation and is positive for localized hybridization.

Lymphoma and Leukemia

Most case of hepatitis due to lymphoma and leukemia are secondary. They present atypical sinusoidal lymphoid and/or portal cells. Immunohistochemical studies and/or flow cytometry are essential for establishing the phenotype.

Extramedullary Hematopoiesis

Extramedullary hematopoiesis is a compensatory mechanism through which blood cells are produced outside the bone marrow when production in the bone marrow is unable to maintain the needs of the organism. Extramedullary hematopoiesis can be observed in a variety of hematological disorders such as myeloproliferative diseases, chronic anemia in neonates and children with giant hemocytes and other pathologies (22, 23). Normoblasts, megakaryocytes, evidence of myelopoiesis, and erythropoiesis in the sinusoidal space are commonly found (Figure 7).

Hypervitaminosis A or Lipidosis of Stellate Cells

In normal livers, vitamin A is stored in stellate liver cells which activate and acquire a phenotype similar to myofibroblasts which produce the extracellular matrix. Toxicity or hypervitaminosis can be serious and can even lead to

cirrhosis. (24, 25) Hypervitaminosis A is characterized by prominent stellate cells in sinusoids in Dissé's space, crescent-shaped cells with dark nuclei which appear like an image of a starry sky due to expansion of lipid vacuoles, proliferation of myofibroblasts, and subsinusoidal fibrosis.

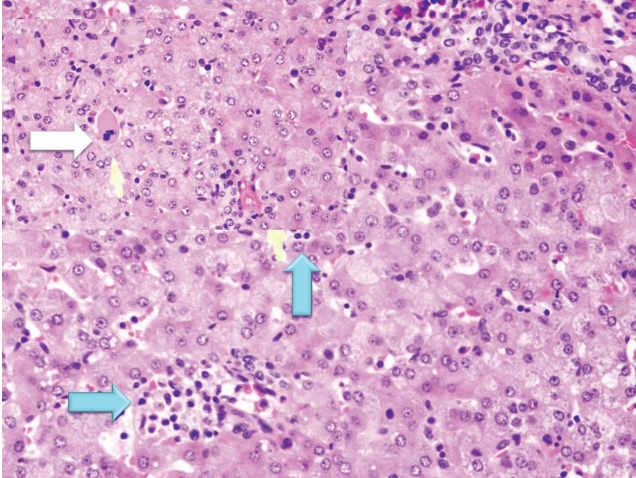


Figure 7. Pattern IV, sinusoidal cellularity. H & E 40x. Increased sinusoidal cellularity with normoblasts (blue arrows) and megakaryocytes (white arrow). The example is from a patient with myeloproliferative syndrome.

Intracellular Pigments

Intracellular pigments are most frequent in the cytoplasm of hepatocytes. They are fine or coarse granular aggregates that acquire various colorations and that may require spe-

cial histochemistry studies to determine the etiology. The most frequent are deposits of lipofuscin, iron, copper or bile. (Figure 8)

Lipofuscin

Lipofuscin is a normal constituent of adult livers, especially during the fifth and sixth decades of life. It is also deposited in chronic liver diseases such as HCV and primary biliary cirrhosis. (7) It is most prevalent in gold and brown colored granular pigment deposits and is distributed in perivenular zones 2 and 3 or in the area between the zones. It occurs near the bile or canalicular canaliculi in the cytoplasm of hepatocytes.

Siderosis (Iron Deposits)

Siderosis is observed in primary conditions such as hereditary and non-hereditary hemochromatosis but can also result from transfusions, hemodialysis, and diet due to increased iron intake. Liver diseases such as alcoholic and nonalcoholic steatohepatitis and hepatitis C can also cause siderosis. (7, 26) It is characterized by refractile gold or brown colored granular pigment deposits in periportal areas. In hereditary hemochromatosis it is also identified throughout the lobule. Located in the canalicular areas of hepatocytes in hereditary forms, excess iron is also deposited in the Kupffer cells, endothelium, stromal cells and ductal epithelia. In siderosis due to transfusions or diet, iron deposits are located in the Kupffer cells. Iron reacts with Prussian blue.

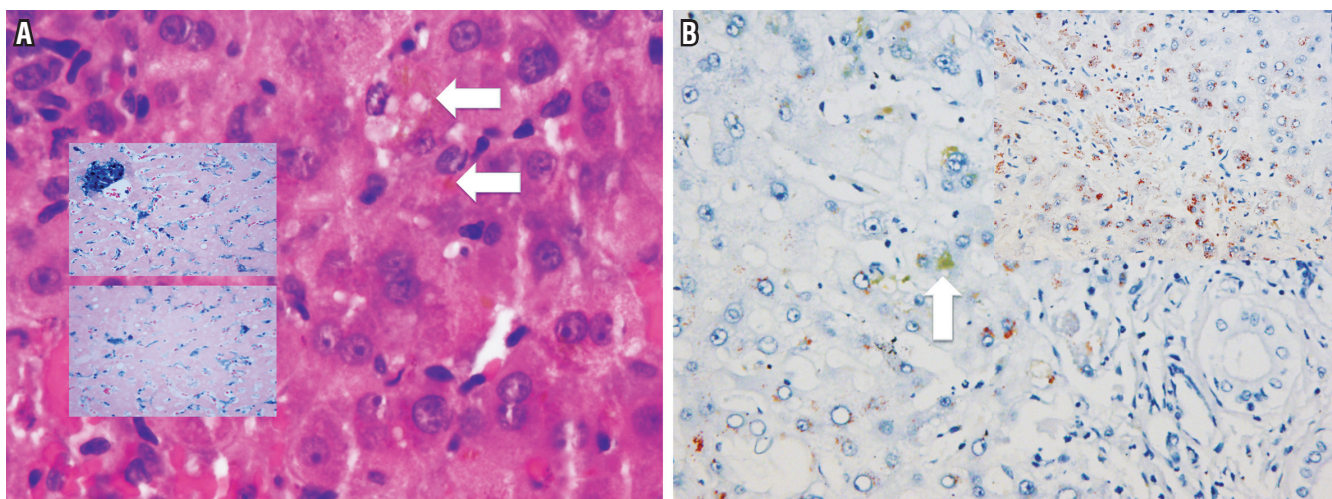


Figure 8. Cellular pigments. A. H & E 20x. Boxes show Prussian blue coloration for 40x iron. Presence of brown pigment, sometimes evident, as indicated by the white arrow, but not evident in other cells. The severe siderosis in blue is identified in the cell marked with the arrow and is demonstrated in Kupffer cells and hepatocyte cytoplasm. B. 40x histochemistry study with orcein stain shows yellow copper deposits in chronic cholestatic disease. In addition, greenish bile pigment (white arrow) is observed.

Copper and/or copper binding serum proteins

Copper and/or copper binding serum proteins are found due to interference with excretion in chronic cholestatic diseases and due to defects in excretion in hereditary conditions such as Wilson’s disease. (7, 27) They appear as deposits of grayish pigment in the cytoplasm. Copper is shown with yellow brick granules in rhodamine, orcein and Victoria blue histochemical studies. It can be observed indirectly from deposits of its fixative protein shown with fuscine aldehyde staining. Frequently, PAS diastase is positive. In chronic cholestatic diseases, deposits are located in the periportal area or at the interface.

PATTERN V: STEATOSIS

Steatosis is fat accumulation and is commonly observed in biopsies. A useful algorithm to guide diagnosis is to observe

whether or not balloon cells and Mallory-Denk bodies are present (Figure 9). This subject was extensively discussed in an earlier article. (28)

Figure 10 shows the algorithm used for this pattern.

PATTERN VI: FIBROSIS

Fibrosis is the end result of the vast majority of liver diseases and is not a pattern of early damage. Determining the stage, amount and type of fibrosis is an indispensable component of the analysis of a biopsy. It should always be evaluated using histochemistry studies such as Masson’s trichrome, the most commonly used, and Sirius red, less frequently used. The pattern of fibrosis provides the keys for diagnosis and subclassification, helps to establish the severity of the disease, and correlates directly with the severity of portal hypertension (Figure 11). (7, 29)

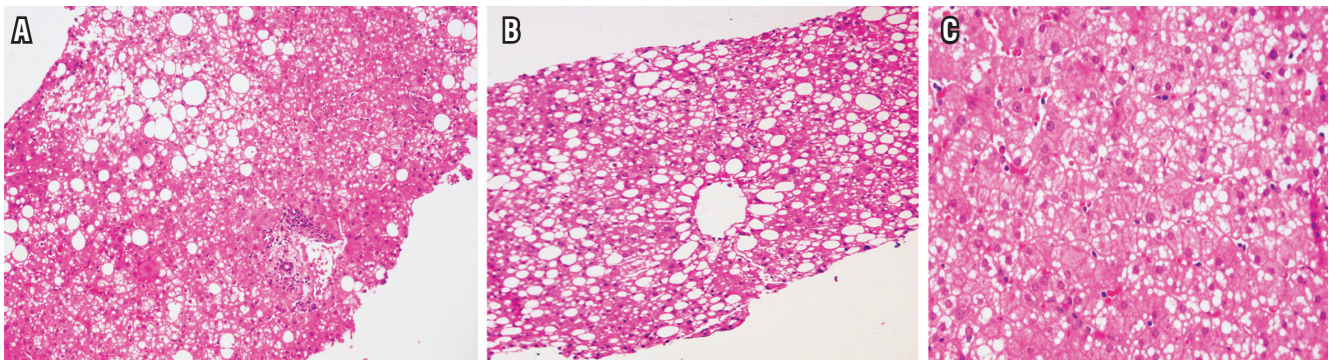


Figure 9. Pattern V, steatosis. A. H & E 20x. Steatosis with ballooning can be observed in alcoholic and non-alcoholic steatohepatitis (NASH). B. H & E 20x. Macrovacuole steatosis without ballooning. C. H & E 40x. Microvacuole steatosis without ballooning.

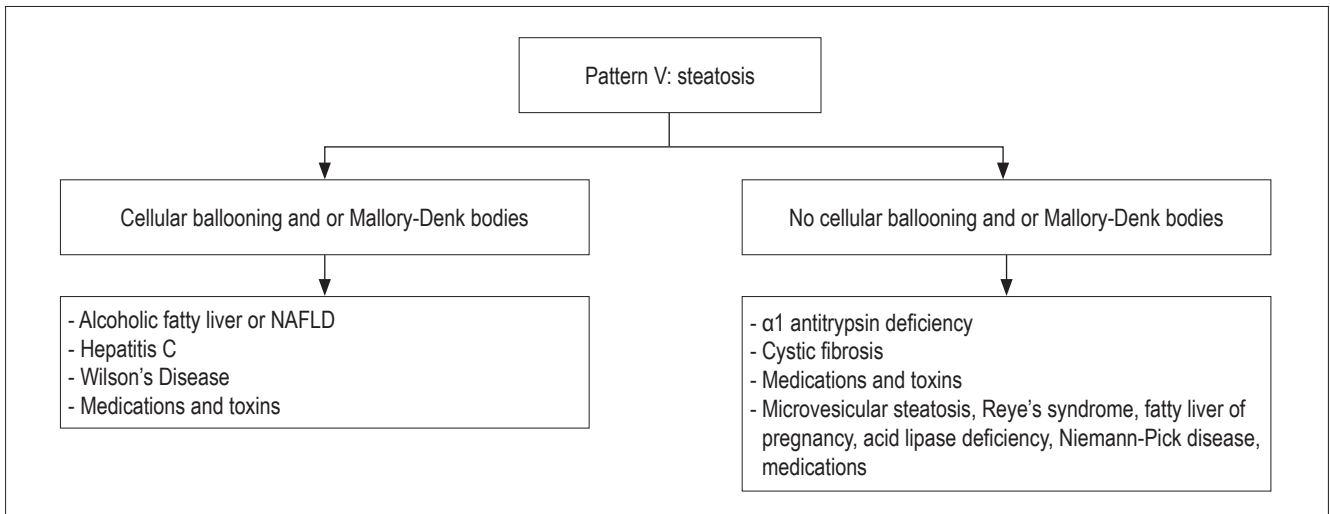


Figure 10. Steatosis analysis algorithm.

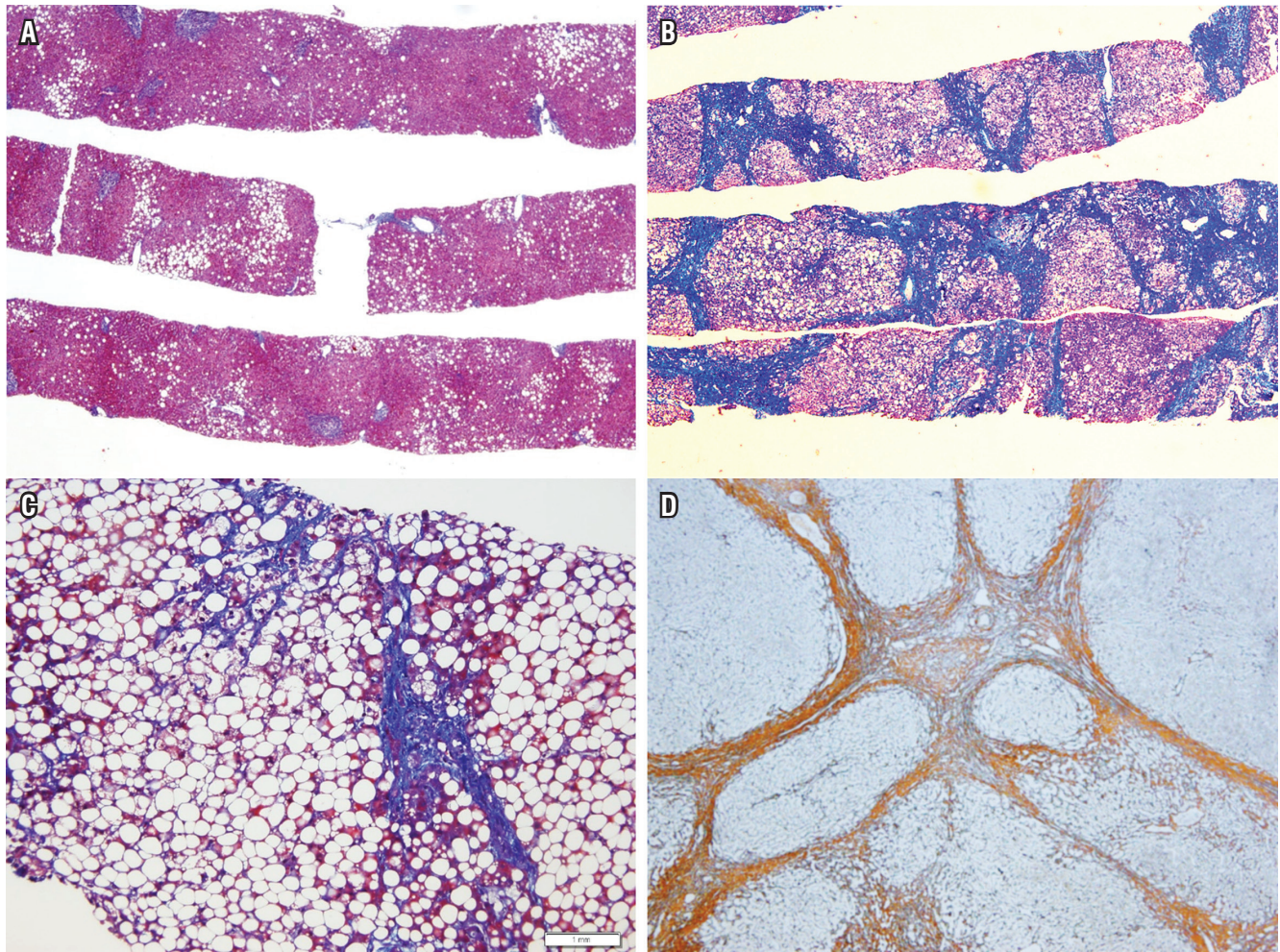


Figure 11. Pattern VI, fibrosis. It is essential to use special stains to establish fibrosis. The most commonly used is Masson's trichrome (MT). A. MT 4x. Steatohepatitis without fibrosis, Stage 0. B. MT 4x. Steatohepatitis with marked fibrosis, nodule formation Stage 4. C. MT 20x. Steatohepatitis with severe steatosis, subsinusoidal fibrosis and portal fibrosis, Stage 2. D. Reticulum staining is also useful for establishing fibrosis. This picture shows an established cirrhosis.

Figure 12 shows the algorithm used when to find alterations of the lobular architecture.

Frequently, fibrosis is overestimated in subcapsular biopsies. A thickness of up to 5 mm in the capsule is considered normal, with more fibrous tissue being observed in the portal spaces and around the central veins, including the presence of some fibrous partitions. In addition, the amount of fibrous tissue also depends on the size of the portal spaces and is directly proportional to the size of the structures of the portal triad. Also, longitudinal cuts that cross the length of the biopsy can suggest fibrous walls or bridges that do not really exist.

Fragmentation of percutaneous liver biopsies is common and increases with the progression of fibrosis from early to advanced stages. Specimens with significant fibrosis or

cirrhosis are the most frequently and most extensively fragmented of all, but fragmentation is not always easy to identify when there are no fibrous edges. A rounded appearance with impaired lobular architecture (absence of central vein or portal triad structures) or fragmented reticular weave suggest that possible cirrhosis, despite the absence of true nodule formation. (30)

Deposits of young collagen (light blue with Masson's trichrome) are especially common in acute hepatitis with necrosis of the recent parenchyma but can be over-interpreted which leads to diagnostic errors. The collapse of the reticular weft helps establish the true nature of the lesion. To determine if fibrosis corresponds to the deposit of mature collagen, check for loss of the reticular weft and presence of cholangiolar proliferation in areas of necrosis and

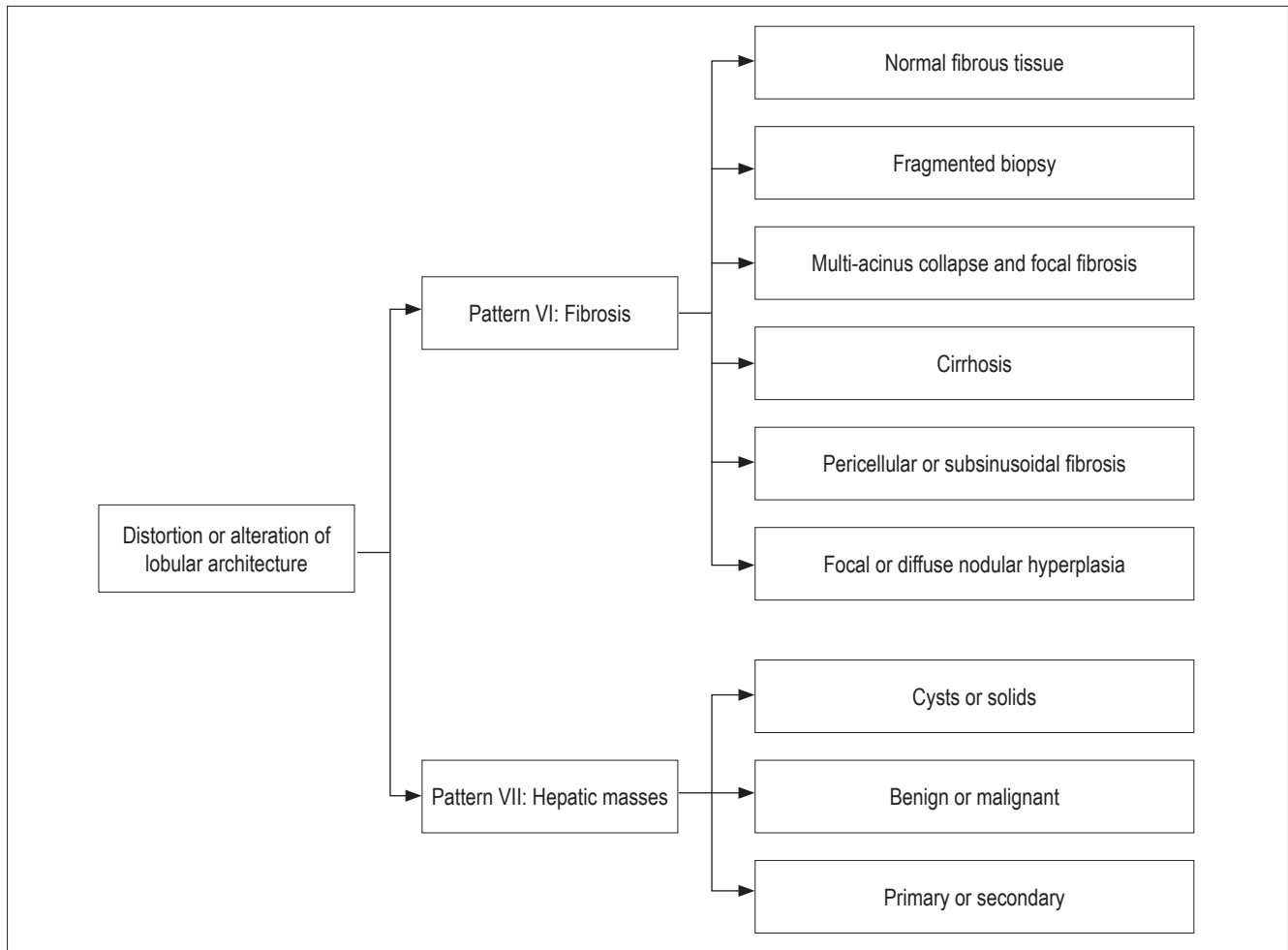


Figure 12. Algorithm of architectural alteration which is useful when there is alteration of the architecture due to either fibrosis or the presence of a mass.

parenchyma with absence of its normal structures, because this will indicate if it is really fibrosis in bridges with incomplete or complete partitions (cirrhosis).

Pericellular or subsinusoidal fibrosis is characteristically observed in zone 3 in nonalcoholic and alcoholic steatohepatitis. In the latter it can progress to occlusion of the central vein. It also occurs in diseases that cause obstruction to chronic venous flow such as sinus veno-occlusive disease and Budd Chiari syndrome and metabolic diseases such as Wilson's disease, tyrosinemia, and galactosemia, and in infectious diseases such as leishmaniasis and congenital syphilis. Portal space-centered fibrosis occurs in chronic liver disease of any etiology but especially in viral hepatitis B and C and in autoimmune hepatitis. It begins irregularly in the portal space, extends into periportal spaces, and later develops portal to portal partitions and finally, develops portal to center partitions until regenerative liver nodules

are formed. The term "early cirrhosis" is used when nodularity is not complete. (29)

PATTERN VII: MASSES

The algorithm (Figure 13) used to determine whether liver masses are cystic or solid mass involves observing the type of epithelial lining and/or the cells from which tumor proliferation originates (Figure 14). Additional histochemistry and immunohistochemistry may be useful. In addition, clinical and imaging correlation is fundamental for establishing whether it is a primary or secondary mass. (30, 31)

CONCLUSION

Liver pathology is extensive and complex. When the study of a liver biopsy is to be done and morphological findings

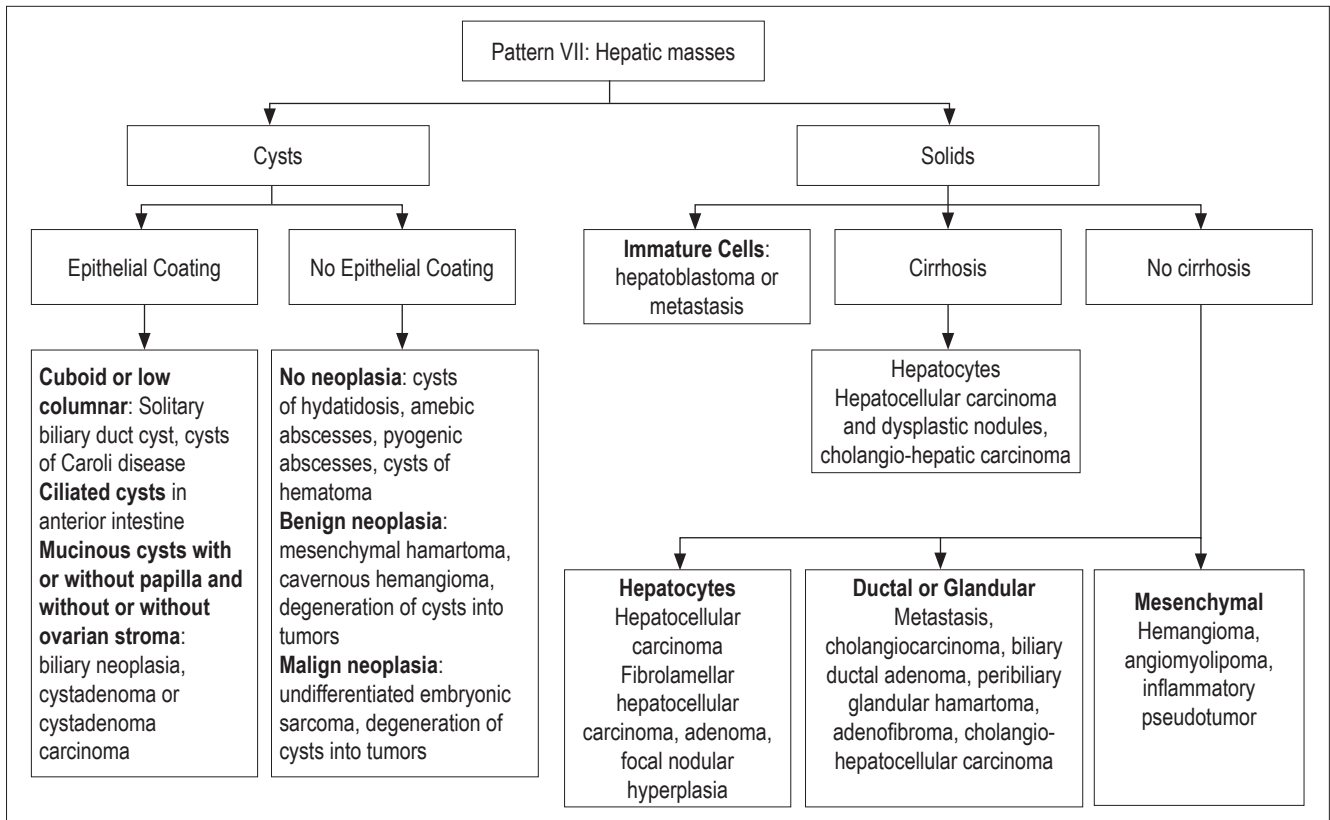


Figure 13. Algorithm for analysis of hepatic masses.

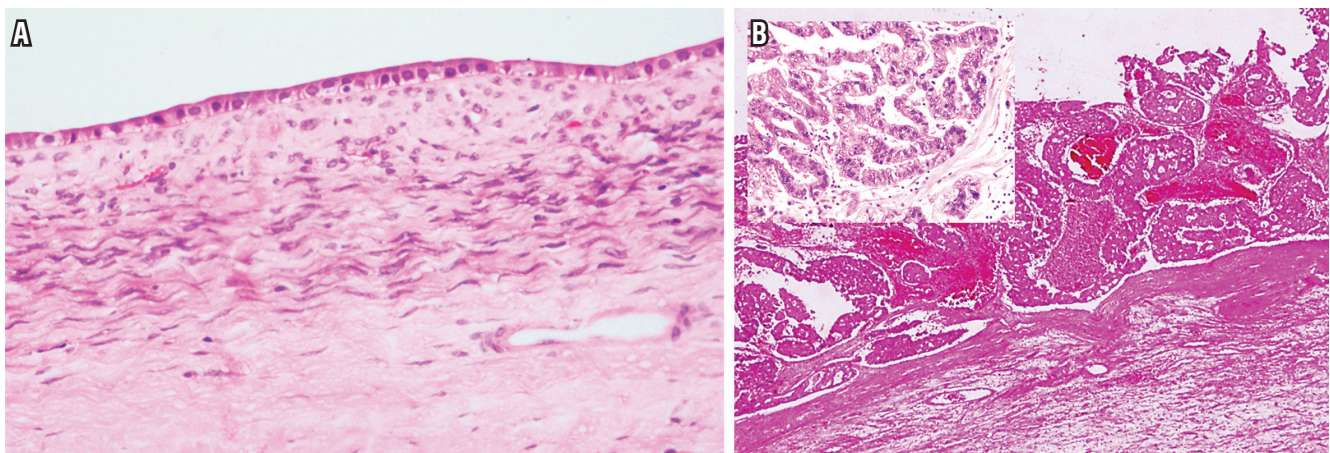


Figure 14. Pattern VII, masses. A. H & E 20X. Cystic mass with epithelial lining. Biliary cystadenoma. Wall coated by a single layer of low cuboid cells without atypia. B. Malignant cystic mass with epithelial lining. The example is from cystadenocarcinoma. Cystic wall coated by complex epithelium, with multiple layers of epithelial cells forming glandular structures, irregular pseudo-papilla with nuclear pleomorphism, hyperchromasia and mitosis.

are analyzed, it is very helpful to rely on algorithms that provide a simpler diagnostic path. Nevertheless, only proper correlation between the histopathology and the complete clinical history can achieve optimum results.

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