Autoimmune hepatitis—is histology conclusive?

Andrea Beer^, Hans Peter Dienes

Department of Pathology, Medical University of Vienna, Waehringer Guertel 18-20, 1090 Vienna, Austria
Contributions: (I) Conception and design: Both authors; (II) Administrative support: None; (III) Provision of study materials or patients: None; (IV) Collection and assembly of data: Both authors; (V) Data analysis and interpretation: None; (VI) Manuscript writing: Both authors; (VII) Final approval of manuscript: Both authors.
Correspondence to: Hans Peter Dienes. Salzwiesengasse 40/3, 1140 Vienna, Austria. Email: hp.dienes@gmail.com.

Abstract: Liver biopsy is an essential and necessary element in the diagnosis and management of autoimmune hepatitis, and is of very special importance in the scoring system for diagnosis. Histopathology shows moderate to severe inflammatory infiltrates with abundant plasma cells in the enlarged portal tracts with interface hepatitis and moderate to severe necroinflammatory lesions in the lobules with lymphoplasmacytic reaction. Regeneration develops with rosette formation and regenerative nodules. One important issue is the differentiation between acute onset of autoimmune hepatitis and a flare up of chronic disease; this cannot be diagnosed in some cases clinically and therefore requires a biopsy to evaluate the stage of the disease. There are some variants of the disease with cholestatic features such as autoantibody negative autoimmune hepatitis and giant cell hepatitis as well as overlap syndromes with primary biliary cholangitis and primary sclerosing cholangitis. Clinically, three types of autoimmune hepatitis are differentiated according to autoantibody formation and the clinical picture, however, histopathologically there is no difference between these three types. Differential diagnosis of autoimmune hepatitis includes drug-induced liver injury with minocycline, alpha methyldopa, nitrofurantoin and checkpoint inhibitors such as infliximab. Wilson´s disease is also an important differential diagnosis especially in young adults. A liver biopsy is mandatory to confirm the diagnosis of autoimmune hepatitis but histopathology alone is not conclusive.

Keywords: Differential diagnosis of hepatitis; histopathology of the liver; immune mediated liver disease; liver cirrhosis; overlap syndromes

Autoimmune hepatitis (AIH) is a relatively rare and heterogeneous disease and may thus render the diagnosis difficult. Liver biopsy is an essential and mandatory element in the diagnosis and management of autoimmune hepatitis and is, owing to its special importance, included in the scoring system for diagnosis (1-6). Since individual, serological and clinical features are not specific for the diagnosis of AIH, evaluation of the liver biopsy helps to exclude other potential causes of liver disease and to identify variant syndromes. Additionally, the biopsy gives information about disease severity and the stage of fibrosis,

and assists in the evaluation of the response to therapy. A normal liver histology is a prerequisite for the withdrawal of therapy. Furthermore, histology is necessary to differentiate between a flare up in chronic disease and acute onset AIH; differentiation can be very difficult clinically.

Histopathology of AIH

According to the AASLD and EASL (4,6) the consensus of histopathology of AIH includes several components: (I) moderate to severe inflammatory infiltrations, which are

---

^ ORCID: 0000-0002-0726-1233.
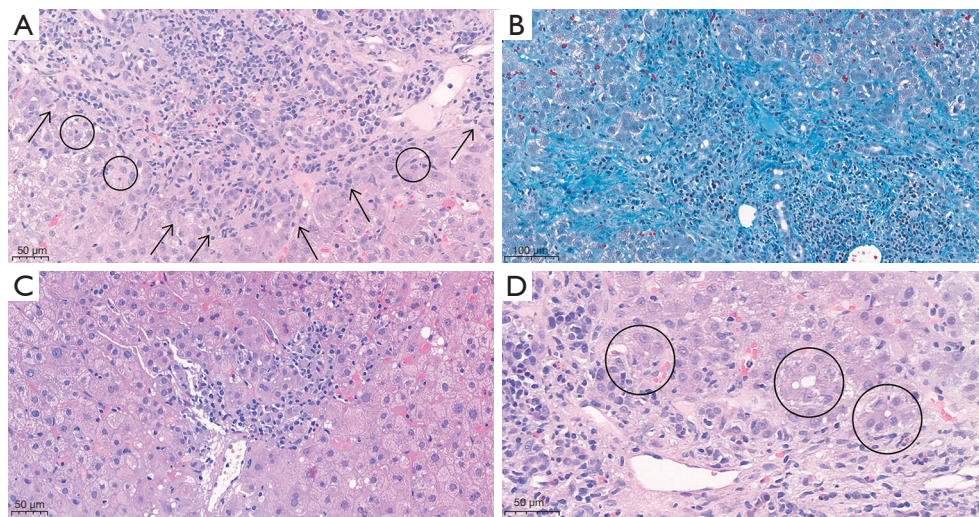
Figure 1 Histopathologic features of autoimmune hepatitis. (A, hematoxylin and eosin) Enlarged, densely infiltrated portal tract with interface hepatitis. The infiltrate consists mainly of lymphoplasmacytic cells; there are many hepatocellular necroses (arrows) and emperipolesis with lymphocytes (circles). (B, chromotrope aniline blue) Fibrosis starts with formation of collagen fibers in the portal tracts as is shown by staining with chromotrope aniline blue. (C, hematoxylin and eosin) Focal necrosis in the lobule close to zone 3 with several clusters of plasma cells. (D, hematoxylin and eosin) Regeneration of parenchyma with rosetting of hepatocytes in the periportal area (circles).

mostly lymphoplasmacytic, in the enlarged portal tracts with moderate to severe interface hepatitis; (II) moderate to severe necroinflammatory lesions in the lobules, sometimes accompanied by confluent necrosis (7,8) and a quantitative extent of plasma cells.

Ad 1: The portal tracts exhibit dense lymphoplasmacytic infiltrates (*Figure 1A*). Plasma cells are often seen in clusters with more than five cells. Neutrophilic or eosinophilic granulocytes may occur. About 25% of cases show features of a reactive cholangitis that are mostly focal, but are almost never associated with a destruction of the bile ducts as in primary biliary cholangitis (PBC). The infiltrates extend into the lobules with moderate to severe interface hepatitis (*Figure 1A*) and with necroses and loss of hepatocytes displaying ballooning and apoptosis. Fibrosis starts in the portal tracts and may progress to formation of septa (*Figure 1B*) and finally to cirrhosis when there is no response to therapy.

Ad 2: In the lobules, moderate to severe necroinflammatory lesions (*Figure 1C*) occur mainly in the periportal area with intense lymphoplasmacytic reaction and a conspicuous activation of Kupffer cells. The Kupffer cells proliferate and are activated and may contain typical Kupffer cell hyaline globules (9). Emperipolesis of lymphocytes (*Figure 1A*) is a characteristic but not specific phenomenon of AIH, that is that lymphocytes penetrate hepatocytes, i.e., T-effector cells form

an immunologic synapse with hepatocytes as their targets (10). The lymphocytes that are encroached on the cytoplasm of the hepatocytes may kill those. In severe acute AIH or an acute flare up of chronic disease confluent necrosis in zone 3 of the lobule may occur (*Figure 1C*), rendering the differentiation from ischemic or drug-induced lesions difficult. As a reaction to the necroses and a sign of the regenerative process, so called rosettes (*Figure 1D*) will form, especially in the periportal area where a ductular reaction and regenerative nodules may develop. However, neither emperipolesis nor rosettes are specific for AIH, because they can also be found in other types of hepatitis. In the areas of necroinflammatory lesions a typical endotheliitis of the central veins can be observed. According to some authors (11), results of copper and cytokeratin 7 staining can help to confirm the diagnosis.

Fibrosis and new collagen formation usually starts in the portal tracts (*Figure 1B*). With progression of the disease, septa may develop—mostly portal to portal, but occasionally also portal to central septa can be seen. In the further course of the disease, regenerative nodules with distortion of the architecture will develop, resulting in cirrhosis in cases where the disease cannot be treated effectively. The presence or absence of fibrosis does not specifically help in the diagnosis of AIH, but it provides significant information on the disease course. It is important to assess the amount of the fibrosis as it determines the stage of the disease which

should be scored according to Ishak's staging (0-no fibrosis to 6-complete cirrhosis) (12).

Histopathology of AIH may therefore prove difficult to interpret and it is recommended to send the biopsy to an experienced hepatopathologist as an extramural consultant (13).

Variant forms of AIH (14,15)

There is no histological difference between the clinical subtypes of AIH type 1 [autoantibodies positive for antinuclear antibody (ANA) and anti-smooth muscle antibodies (SMA)] and type 2 [positive for liver/kidney microsome (LKM) and liver cytosol antibody type 1 (LC1)] or type 3 [positive for soluble liver antigen/liver pancreas (SLA/LP) antibodies clinically as type 1] (1,9,10). Some of the patients also show antimitochondrial antibodies (AMA), however, this is not associated with PBC-like cholangitis (16).

Syncytial giant cell hepatitis is an uncommon pattern of liver injury defined by giant cell transformation of hepatocytes with each giant cell having more than 5 nuclei and a transformation of hepatocytes to giant cells in more than 10%. In children, this type of hepatitis is not uncommon, but in adults (post-infantile giant cell hepatitis, PIGCH) it is rather infrequent and is supposed to have an autoimmune background with the presence of autoantibodies (17).

There is also a subgroup of patients with AIH (about 15%) who lack autoantibodies, are showing high levels of immunoglobulins, a typical histology of AIH, a typical genetic background in the HLA system and response to therapy. The histology is not different from patients with a typical spectrum of autoantibodies (18).

Acute onset AIH versus flare up in chronic AIH

Acute onset AIH can present as a fulminant hepatitis with extensive hepatocyte necrosis and marked inflammatory changes. The necrosis is often panacinar with zone 3 predominance but can also be quite patchy. Acute onset AIH can be seen when the biopsy is taken within the first three months of onset of symptoms and an elevation of laboratory parameters such as bilirubin and transaminases (19-21). It has to be distinguished from a flare up in a chronic course, which is sometimes difficult to discriminate clinically. In this case, histology proves important: The flare up in a chronic course shows fibrosis to a different degree with features of regeneration in the form of regenerative nodules, and distortion of the lobular architecture. Over

time, scars may form in areas of collapse thus yielding a very heterogeneous pattern for the cirrhosis.

Overlap syndromes of AIH with PBC and PSC

Overlap syndrome of AIH and PBC (4,22-24): The diagnosis should be made only when the two concurrent diseases can be established and meet the criteria for each disease both clinically and histologically. According to AASLD guidelines the criteria are the following: serum alkaline phosphatase (AP) level at least two times the upper limit of normal or serum gamma GT at least five times the upper limit of normal, presence of AMA and florid bile duct lesions (two out of three criteria are necessary), moreover, serum alanine aminotransferase (ALT) at least five times the upper limit of normal and IgG at least two times the upper limit of normal or the presence of SMA (6). Histology shows the typical bile duct lesions with the destruction of ducts and granuloma formation on the one hand and a moderate to severe hepatitis in the lobules that exceeds the accompanying hepatitis in PBC, on the other hand.

Overlap of AIH and primary sclerosing cholangitis (PSC) (25,26): This incidence is less frequent than that of AIH and PBC. It may occur in children presenting with a so called autoimmune cholangitis but is more frequent in adults. The criteria in the AASLD guidelines are as follows: typical features of AIH, absence of AMA, evidence of large duct PSC by imaging or evidence of small-duct PSC based on onion skinning periductal fibrosis (6). Usually, AIH precedes PSC which is diagnosed when AIH responds to therapy but a cholestatic syndrome develops. Histology will demonstrate a typical hepatitis pattern and later on the bile ducts will show cholangitis with periductular fibrosis and a thickening of the duct membranes.

Autoimmune sclerosing cholangitis: This disease is more common in children and young adults and may be missed initially because the biopsy may show changes of AIH in the absence of features strongly suggesting the presence of biliary tract disease.

Small duct PSC is defined as liver biopsy findings consistent with PSC but with normal appearance of the bile ducts at cholangiography. A subset of these cases can also show overlapping histologic features with AIH, but require convincing evidence of both diseases.

Differential diagnosis

Viral hepatitis has, of course, to be excluded by serological

testing, but especially hepatitis C may concur with AIH. In these cases the patients are positive for HCV but also have autoantibodies. After successful treatment for the virus the patients show characteristic clinical and histologic features of AIH (27,28). Drug induced liver injury may mimic AIH (29), especially with drugs such as minocycline, alpha methyldopa, nitrofurantoin and checkpoint inhibitors such as infliximab when taken for longer periods of time. Therefore, a drug reaction has to be excluded carefully by clinical evaluation. Another disease may present as a hepatitis that is histologically indistinguishable from AIH: Wilson's disease. Especially in young adults this is an important differential diagnosis. Although it is possible to perform staining for copper in the tissue—rhodanine staining—this technique is not absolutely reliable. Consequently, the absolute copper concentration must be determined either by other techniques in the tissue or clinically by 24-hour urinary copper excretion.

In conclusion

A liver biopsy in AIH is necessary and mandatory to confirm the diagnosis, to determine the activity and severity of the disease and to exclude other diseases. Histopathology of AIH is typical but does not show pathognomonic features and is therefore not conclusive for the disease; thus, a definitive diagnosis of AIH by histology alone is not possible and should be based on the simplified criteria (3) that include presence of autoantibodies, elevation of IgG and histopathology graded as unlikely (0 points), possible (1 point) or likely (2 points).

Acknowledgments

Footnote

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

References

1. Mieli-Vergani G, Vergani D, Czaja AJ, et al. Autoimmune Hepatitis. Nat Rev Dis Primers 2018;4:18017.
2. Alvarez F, Berg PA, Bianchi FB, et al. International Autoimmune Hepatitis Group Report: review of criteria for diagnosis of autoimmune hepatitis. J Hepatol 1999;31:929-38.
3. Hennes EM, Zeniya M, Czaja AJ, et al. Simplified criteria for the diagnosis of autoimmune hepatitis. Hepatology 2008;48:169-76.
4. European Association for the Study of the Liver. EASL Clinical Practice Guidelines: Autoimmune hepatitis. J Hepatol 2015;63:971-1004.
5. Tanaka A. Autoimmune Hepatitis: 2019 Update. Gut Liver 2020;14:430-8.
6. Mack CL, Adams D, Assis DN, et al. Diagnosis and Management of Autoimmune Hepatitis in Adults and Children: 2019 Practice Guidance and Guidelines From the American Association for the Study of Liver Diseases. Hepatology 2020;72:671-722.
7. Dienes HP, Popper H, Manns MP, et al. Histologic features in autoimmune hepatitis. Z Gastroenterol 1989;27:325-30.
8. Terracciano LM, Patzina RA, Lehmann FS, et al. A spectrum of histopathologic findings in autoimmune liver disease. Am J Clin Pathol 2000;114:705-11.

9. Gurung A, Assis DN, McCarty TR, et al. Histologic features of autoimmune hepatitis: a critical appraisal. Hum Pathol 2018;82:51-60.

10. Dienes HP, Autschbach F, Gerber MA. Ultrastructural lesion in autoimmune hepatitis and steps of the immune response in liver tissue. Semin Liver Dis 1991;11:197-204.

11. Balitzer D, Shafizadeh N, Peters MG, et al. Autoimmune hepatitis: review of histologic features included in the simplified criteria proposed by the international autoimmune hepatitis group and proposal for new histologic criteria. Mod Pathol 2017;30:773-83.

12. Ishak K, Baptista A, Bianchi L, et al. Histological grading and staging of chronic hepatitis. J Hepatol 1995;22:696-9.

13. Torbenson MS, Arnold CA, Graham RP, et al. Identification of key challenges in liver pathology: data from a multicenter study of extramural consults. Hum Pathol 2019;87:75-82.

14. Czaja AJ. The variant forms of autoimmune hepatitis. Ann Intern Med 1996;125:588-98.

15. Bogdanos DP, Mieli-Vergani G, Vergani D. Autoantibodies and their antigens in autoimmune hepatitis. Semin Liver Dis 2009;29:241-53.

16. O'Brien C, Joshi S, Feld JJ, et al. Long-term follow-up of antimitochondrial antibody-positive autoimmune hepatitis. Hepatology 2008;48:550-6.

17. Protzer U, Dienes HP, Bianchi L, et al. Post-infantile giant cell hepatitis in patients with primary sclerosing cholangitis and autoimmune hepatitis. Liver 1996;16:274-82.

18. Lüth S, Herkel J, Kanzler S, et al. Serologic markers compared with liver biopsy for monitoring disease activity in autoimmune hepatitis. J Clin Gastroenterol 2008;42:926-30.

19. Weiler-Normann C, Lohse AW. Acute autoimmune hepatitis: many open questions. J Hepatol 2014;61:727-9.

20. Shen Y, Lu C, Men R, et al. Clinical and Pathological Characteristics of Autoimmune Hepatitis with Acute Presentation. Can J Gastroenterol Hepatol 2018;2018:3513206.

21. Nguyen Canh H, Harada K, Ouchi H, et al.; Intractable Liver and Biliary Diseases Study Group of Japan. Acute presentation of autoimmune hepatitis: a multicentre study with detailed histological evaluation in a large cohort of patients. J Clin Pathol 2017;70:961-9.

22. Boberg KM, Chapman RW, Hirschfield GM, et al. Overlap syndromes: the International Autoimmune Hepatitis Group (IAIHG) position statement on a controversial issue. J Hepatol 2011;54:374-85.

23. Dienes HP, Erberich H, Dries V, et al. Autoimmune hepatitis and overlap syndromes. Clin Liver Dis 2002;6:349-62, vi.

24. Lohse AW, zum Büschenfelde KH, Franz B, et al. Characterization of the overlap syndrome of primary biliary cirrhosis (PBC) and autoimmune hepatitis: evidence for it being a hepatitic form of PBC in genetically susceptible individuals. Hepatology 1999;29:1078-84.

25. Gohlke F, Lohse AW, Dienes HP, et al. Evidence for an overlap syndrome of autoimmune hepatitis and primary sclerosing cholangitis. J Hepatol 1996;24:699-705.

26. Abdalian R, Dhar P, Jhaveri K, et al. Prevalence of sclerosing cholangitis in adults with autoimmune hepatitis: evaluating the role of routine magnetic resonance imaging. Hepatology 2008;47:949-57.

27. López Couceiro L, Gómez Domínguez E, Muñoz Gómez R, et al. Healing of autoimmune hepatitis associated with hepatitis C virus infection treated with direct-acting antivirals. Rev Esp Enferm Dig 2019;111:159-61.

28. Simoes CC, Saldarriaga OA, Utay NS, et al. Direct-Acting Antiviral Treatment of Patients with Hepatitis C Resolves Serologic and Histopathologic Features of Autoimmune Hepatitis. Hepatol Commun 2019;3:1113-23.

29. Czaja AJ. Drug-induced autoimmune-like hepatitis. Dig Dis Sci 2011;56:958-76.