The General Practice Guide to Autoimmune Diseases

Edited by Y. Shoenfeld and P. L. Meroni

EASi™
European Autoimmunity
Standardisation Initiative


ThermoFisher
SCIENTIFIC

The General Practice Guide to Autoimmune Diseases

Edited by Y. Shoenfeld and P. L. Meroni

Pabst Science Publishers
Lengerich, Berlin, Bremen, Miami,
Riga, Viernheim, Wien, Zagreb

Bibliographic information published by Deutsche Nationalbibliothek
The Deutsche Nationalbibliothek lists this publication in the Deutsche Nationalbibliografie; detailed bibliographic data is available in the Internet at <http://dnb.ddb.de>.

http://www.pabst-publishers.de

Printing: MercedesDruck, Berlin
Typesetting: Hilmar Schlegel, Berlin
Cover: Agentur für zeitgemäße Kommunikation Kaner Thompson
www.kanerthompson.de

ISBN 978-3-89967-770-6

Autoimmune hepatitis

Reinhild Klein

1 Introduction

Autoimmune hepatitis (AIH) is a chronic, progressive disease which occurs in children and adults of all ages and affects mainly females. Characteristic features are the fluctuating spontaneous course, histologically determined interface hepatitis, as well as the hypergammaglobulinaemia of immunoglobulin G (IgG) type, and the presence of circulating autoantibodies. AIH occurs all over the world with varying incidence and prevalence. Its prevalence is estimated to range between 50 and 200 cases per million in Western Europe and North America among the Caucasian population. In this group, AIH accounts for up to 20 % of cases of chronic hepatitis. In countries in which viral hepatitis B and C are endemic, such as in Asia and Africa, the incidence of AIH seems to be significantly lower [1, 2].

The pathogenesis of AIH is unknown. Loss of tolerance against hepatic tissue is presumed and an underlying genetic predisposition has been suggested. Anti-inflammatory/immunosuppressive treatment induces remission but long-term maintenance therapy is often required. Liver transplantation is generally successful in patients with decompensated cirrhosis unresponsive to or intolerant of medical therapy. Overall, long-term survival and average life expectancy of adequately treated patients are excellent and estimated to be comparable with those of the normal population.

2 Diagnostic criteria

The clinical presentation of AIH is very heterogeneous ranging from asymptomatic disease to severe icteric hepatitis, and even fulminant hepatitis which may require liver transplantation. In 1992, the International Autoimmune Hepatitis Group recommended a scoring system for the diagnosis of AIH to allow reliable diagnosis of the disease, and this was further updated in 1999 (Table 1a) [1–3]. The clinical relevance of this scoring system as well as other, more simplified systems (Table 1b) developed in the interim [3] is, however, still a matter of debate.

Table 1. a) International diagnostic criteria for the diagnosis of AIH [1–3].

Parameter		Score
gender	female	+2
	male	0
serum biochemistry		
	ratio of elevation of serum alkaline phosphatase vs. aminotransferase	
	> 3.0	-2
	1.5-3	+2
	< 1.5	+1
	< 1.0	0
total serum globulin, -globulin or IgG		
	Times upper normal limit	
	> 2.0	+3
	1.5-2.0	+2
	1.0-1.5	+1
	< 1.0	0
autoantibodies (titres by immunofluorescence on rodent tissues)		
	ANA, SMA or LKM-1	
	> 1:80	+3
	1:80	+2
	1:40	+1
	< 1:40	0
	antimitochondrial antibody	
	positive	-4
	negative	0
hepatitis viral markers	negative	+3
	positive	-3

other aetiological factors	
History of drug use	
yes	-4
no	+1
Alcohol (average consumption)	
< 25 g/day	+2
> 60 g/day	-2
Genetic factors: HLA DR3 or DR4	+1
Other autoimmune diseases	+2
Response to therapy	
complete	+2
relapse	+3
liver histology	
Interface hepatitis	+3
Predominant lymphoplasmacytic infiltrate	+1
Rosetting of liver cells	+1
None of the above	-5
Biliary changes	-3
Other changes	-3
Seropositivity for other defined autoantibodies	+2

3 Diagnostic measurement for experts

Autoantibodies are one of the distinguishing features of AIH. The discovery of autoantibodies directed against different cellular targets, including nuclear, cytosolic and microsomal antigens has allowed a suggested subclassification of AIH based on the presence of three specific autoantibody profiles — although there is little evidence to support a role for these antibodies in pathogenesis (Table 2).

According to this approach, AIH type 1 is characterized by the presence of antinuclear antibodies (ANA) and/or anti-smooth muscle antibodies (SMA) directed against F-actin. AIH type 2 is characterized by anti-liver-kidney microsomal antibodies (LKM-1) reacting with cytochrome P450 2D6, and AIH type 3 by autoantibodies against a soluble liver/liver-pancreas antigen (SLA/LP) currently identified as UGA suppressor serine tRNA-protein complex. Although there are some clinical/biochemical differences between these subgroups (Table 2) [4], this serological classification has no implications with respect to therapeutic regimes and is, therefore, still controversial.

An initial liver biopsy for confirmation of diagnosis and for grading and staging is desirable. Biopsies are also helpful for the observation of which degree aminotransferase activities in serum reflect an inflammatory activity in the liver, which is not closely related in all cases. The histological appearance of AIH is the same as that of chronic hepatitis of other aetiology, and although certain changes are characteristic, no findings are specific for AIH [1, 2]. The inflammatory component is characterised by a mononuclear cell infiltrate, which invades the limiting plate surrounding the portal triad and permeates the surrounding parenchyma (periportal infiltrate; piecemeal necrosis; interface hepatitis) and beyond (lobular hepatitis). Eosinophils are frequently present. Fibrosis is present in all but the milder forms of AIH. With distortion of the hepatic lobule and the appearance of regenerative nodules, the result is cirrhosis.

A genetic predisposition is viewed as a prerequisite of AIH, and first degree relatives of AIH-patients are at high risk of also developing AIH or another au-

Table 1. b) Simplified diagnostic criteria for AIH according to [3].

Parameter	Cutoff value	Points
ANA or SMA	$\geq$ 1: 40	1
	$\geq$ 1: 80	
Or LKM	$\geq$ 1: 40	2*)
Or SLA/LP	Positive	
IgG	> upper limit of normal	1
	> 1.1 × upper limit of normal	2
Histology (evidence of hepatitis is required)	Compatible with AIH	1
	Typical for AIH	2
Absence of viral hepatitis	yes	2

Interpretation of aggregate points: $\geq$ 6 points: probable AIH; $\geq$ 7 points: definite AIH
*) proceed by adding points achieved for all autoantibodies; the maximum is 2 points.

toimmune disease [1, 2]. However, the genetic background of AIH does not follow a Mendelian pattern, and a conclusive role for a single genetic locus capable of explaining the aetiology of AIH has not been identified. Association of HLA A1, Cw7, B8 and DR3 as well as DR4 with AIH and other autoimmune diseases has been conclusively demonstrated in a number of studies. Studies from Europe and the U.S. have identified DRB1*0301 and DRB1*0401 as susceptibility alleles, and DRB1*1501 as a resistance allele. However, immunogenetic findings appear to not apply universally and it has been noted that significant geographic differences exist. While in Caucasian patients those with HLA DR3 and DR4 are independently susceptible to autoimmune hepatitis, DR4 is predominant in Japanese patients, and there are no Japanese patients with DR3.

4 Requirements for family practitioners

The clinical presentation of AIH is very heterogeneous ranging from asymptomatic disease to severe icteric hepatitis, and even fulminant hepatitis which may require liver transplantation.

Patients may present with non-specific symptoms of varying severity, such as fatigue, lethargy, malaise, anorexia, nausea, amenorrhoea, abdominal pain, and itching. Arthralgia is quite common. Physical examination may be without pathological findings, but may also reveal hepatomegaly, splenomegaly, jaundice, and signs and symptoms of chronic liver disease. Other autoimmune diseases such as Hashimoto's thyroiditis, ulcerative colitis, type 1 diabetes, rheumatoid arthritis, and coeliac disease as well as other autoimmune liver disorders (primary biliary cirrhosis, primary sclerosing cholangitis) have been described as being associated with AIH.

It is important to identify and treat AIH at its earliest stages, because untreated patients with mild disease progress to cirrhosis within 15 years. If untreated, severe AIH has a very high mortality rate of up to 50 % 3–5 years after diagnosis. Immunosuppressive therapy with corticosteroids, usually in combination with azathioprine is considered the gold standard to induce and maintain remission. Moreover, response to immunosuppressive therapy confirms the diagnosis of AIH. The therapeutic goal should be complete normalization of transaminases because progression to liver cirrhosis may occur in patients with residual inflammatory activity within the liver. However, side-effects of therapy must be taken into consideration. Although some patients remain in remission after drug treatment is withdrawn, most require long-term maintenance therapy. It has been proposed that patients should be in stable remission for at least 4 years before withdrawal of immunosuppressive therapy can be considered.

AIH is not a contraindication to pregnancy. However, patients should be in remission under a maintenance therapy of 5 mg steroids per day. Steroid dose should be increased shortly after delivery because a relapse can occur.

Table 2. Classification and characteristics of autoimmune hepatitis (according to [2, 4]).

Variable/parameter		**AIH type 1**		**AIH type 2**	**AIH type 3**
		ANA positive	anti-actin positive	anti-LKM1 positive	anti-SLA/LP positive
		number patients analysed			
		167	218	40	175
sex	ratio f:m	2.6 : 1	1.9 : 1	2.3 : 1	5.3 : 1
age	mean	52.5	45.3	29.6	46.2
	range	11-88	2-83	5-73	8-86
laboratory parameters at presentation					
AST (normal < 20 IU)	mean	143	137	123	86
ALT (normal < 20 IU)	mean	149	169	125	110
IgG (normal < 1.800 mg/dl)	mean	3.103	2.864	1.998	2.697

5 Follow up

Clinical observations and expectations

Up to 30 % of adult patients already have histological features of cirrhosis at diagnosis. However, the presence of cirrhosis seems not to influence 10-year survival (90 %) and those patients require a similarly aggressive treatment strategy as patients without cirrhosis [1, 2, 5].

histopathologic features at presentation				
cirrhosis (%)	25	15	30	6
chronic active hepatitis/ acute hepatitis (%)	61	70	63	68
chronic persistent hepatitis (%)	13	15	8	17
geographic variation	worldwide	worldwide	worldwide; rare in North America	worldwide
association with other autoimmune diseases	common	common	common	common
clinical severity	broad range	broad range	generally severe	broad range
treatment failure	infrequent	infrequent	frequent	infrequent
relapse after drug withdrawal	frequent	frequent	frequent	frequent
need for long-term maintenance	variable	variable	approximately 100%	variable

In children, about 50 % have cirrhosis at the time of diagnosis. Long-term follow-up reveals that only a few children can completely stop all treatment and about 70 % of children receive long-term treatment. Most of these patients relapse when treatment is discontinued, or if the dose of immunosuppressive drugs is

reduced. About 15 % of patients develop chronic liver failure and are transplanted before the age of 18 years.

The aim of treatment is the induction of remission, i. e. a complete normalization of all inflammatory parameters including histology indices. This can be achieved in 65–75 % of patients after 24 months of treatment. Relapse is characterised by an increase of aminotransferase levels and IgG immunoglobulins and occurs in 50 % of patients within six months of treatment withdrawal and in 80 % after 3 years. It is associated with progression to cirrhosis in 38 % and liver failure in 14 %.

Blood tests

During treatment laboratory parameters should be assessed twice a year on patients who are asymptomatic and in remission, in patients with clinical symptoms every three months, and in patients showing inflammatory activity (elevated transaminases, IgG globulins) at least every two weeks until remission.

Autoantibody titres can decrease during immunosuppressive therapy and can even completely disappear in patients in remission. Increase of autoantibody activity together with an increase in IgG globulins in those patients may then indicate aggravation of the disease. However, there are also patients in whom autoantibodies persist despite adequate treatment.

6 Management

Independent of the clinically or immunoserologically defined type of AIH, standard treatment is implemented with prednisone (or prednisolone) alone or in combination with azathioprine (Table 3). Both strategies are equally effective. However, depending on a variety of definitions of response, success rates are only in the range of 65–70 %, which leaves a significant number of patients in need of other standard treatment [1, 2, 5]. Adults with cirrhosis at the time of initial biopsy and children, particularly those with AIH type 2, rarely stay in remission when treatment is withdrawn and will almost certainly require life-long maintenance therapy.

No firm guidelines exist for decisions regarding withdrawal of medications because histological changes may lag biochemical responses and a quiescent histological appearance and normal biochemical findings while patients are still receiving therapy, are not necessarily predictive of continued remission once therapy is withdrawn. Therapy is usually administered over a course of at least 2 years. The decision between monotherapy and combination therapy is guided by the side effects of steroid therapy. Cosmetic side effects (Cushing's syndrome), in particular, decrease patient compliance. Serious complications such as steroid diabetes, osteopenia, aseptic bone necrosis, psychiatric symptoms, hypertension and

cataract-formation must be anticipated in long-term treatment, especially when the steroid dose cannot be tapered down to 5 mg per day. Azathioprine can be used to decrease the dose of prednisone [5] but it bears a theoretical risk of teratogenicity. In addition, abdominal discomfort, nausea, cholestatic hepatitis, rashes and leucopenia may be encountered. Toxicity and/or intolerance to azathioprine and its metabolite 6-mercaptopurine can occur and depends upon mutations in the thiopurine methyltransferase genes. Dose reduction is aimed at finding the individually appropriate maintenance dose. Usually, a maintenance dose of prednisone or prednisolone ranges between 10 and 2.5 mg and of azathioprine between 50 and 100 mg per day (Table 3). The use of budesonide is, to date, only recommended for patients with mild inflammation or patients in remission.

Table 3. Standard treatment of autoimmune hepatitis in adults.

Regimen	Single-drug therapy	Combination therapy
Initial	prednisone or prednisolone 20–60 mg/day	prednisone or prednisolone 15–30 mg/day, azathioprine 50–100 mg/day
Maintenance	prednisone or prednisolone 5–15 mg/day	prednisone or prednisolone 15–30 mg/day, azathioprine 50–100 mg/day

Treatment failure is characterised by a progression of clinical, serological and histological parameters during standard therapy and is seen in about 10 % of patients. In these patients the diagnosis of AIH must be carefully reconsidered. Alternative immunosuppressive therapies have been proposed, mainly on the basis of small series or case reports. These have included cyclosporine, tacrolimus, methotrexate, cyclophosphamide, ursodiol, and mycophenolate mofetil (MMF) [5].

AIH patients who develop decompensated cirrhosis may require liver transplantation. There is no single indicator or predictor for the necessity of liver transplantation. The 5-year survival is up to 92 % and the rate of recurrence of AIH after transplantation ranges between 10 and 35 %.

7 Diagnostic test

Autoantibodies are one of the most important diagnostic markers in AIH (Fig. 1), although there is little evidence to support a role for these antibodies in pathogenesis.

AIH type 1 is a classical type, so-called lupoid hepatitis. It is associated with ANA and/or SMA which react with F-actin (Fig. 1A, B). These should be detected by immunofluorescence testing on cryostat sections and not by cell culture

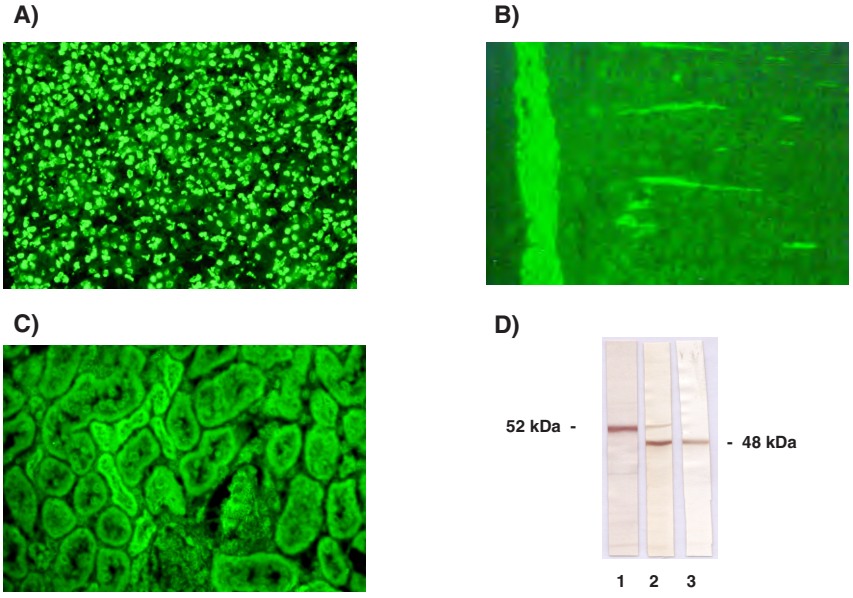

Figure 1. Demonstration of AIH-related autoantibodies by immunofluorescence test (IFT) (A–C) and Western blotting (D). A) Demonstration of antinuclear antibodies (ANA) on rat liver. B) Demonstration of antibodies to smooth muscle antigens with anti-actin specificity on rat stomach showing the typical staining of smooth muscle cells and interparietal cell fibers characteristic for anti-actin. C) Demonstration of antibodies to liver-kidney microsomes (LKM) on rat kidney showing the typical coarse granular cytoplasmic staining of tubules. D) Demonstration of anti-SLA/LP antibodies by Western blotting revealing the typical determinants at 52 and 48 kDa (anti-SLA/LP antibodies cannot be detected by IFT!). Three patterns can be observed: 1: sera reacting only with the 52 kDa band, 2: sera reacting with both, the 52 and the 48 kDa bands, and 3: sera reacting only with the 48 kDa band.

slides (for instance Hep2 cells) because the latter tests frequently detect naturally occurring ANA with no clinical relevance e. g. in patients with infectious or drug-induced disorders. Antibody titres may decrease during therapy.

Anti-liver/kidney microsome-1 (LKM-1) and anti-liver cytosol-1 (LC-1) antibodies occurring alone or together characterise AIH type 2 (Fig. 1C). Anti-LKM-1 antibodies are directed against cytochrome P450 2D6. About 10 % of patients belong to this group of AIH, and these are mainly children. Anti-LKM antibodies have also been found in patients with hepatitis B or C or drug-induced hepatitis, but these are directed against either other microsomal antigens or against epitopes of Cyp 2D6 other than the anti-LKM-1 antibodies in AIH type 2.

In contrast to ANA, SMA, or anti-LKM which can also be found in other (liver) disorders, the antibodies to soluble liver/liver-pancreas antigen (SLA/LP) are con-

fined to AIH and have not been found in any other liver disease [4]. They occur in about 30 % of AIH patients and can be associated with ANA or anti-actin. In about 10 % of patients, however, they occur without any other relevant autoantibody and may, therefore, comprise a separate serological group (AIH type 3). However, the antibodies cannot be detected by IFT but only by Enzyme Linked Immunosorbent Assay (Elisa), radioimmunoassay or Western blotting (Fig. 1D). Again, the antibodies can disappear during immunosuppressive therapy. The antigen involved has been identified as human suppressor serine tRNA associated protein, a co-translocation factor which incorporates seleno-cystein in human cells.

Moreover, antibodies to the asialoglycoprotein receptor protein (ASGPR), a membrane protein of hepatocytes, have been described which seem to correlate with disease activity and prognosis.

Antibodies to neutrophils showing an atypical perinuclear staining (pANCA) have also been detected in AIH, and in those patients an association of AIH with primary sclerosing cholangitis and/or ulcerative colitis must be considered.

However, it needs to be clearly stated that patients with AIH exist, in whom currently known autoantibodies are completely undetectable.

The presence of antimitochondrial antibodies in AIH is strongly indicative for an overlap syndrome with primary biliary cirrhosis.

One characteristic laboratory feature of AIH is the elevation of serum immunoglobulins, in particular IgG.

8 Testing methods

The benefits and limitations of diagnostic laboratory tests are discussed in the chapter 'primary biliary cirrhosis'.

Using IFT on cryostat sections from rodents the diagnosis of AIH can be made in about 90 % of patients (Fig. 2). Only anti-SLA/LP antibodies cannot be detected by this method. For their measurement, complement fixation, radioimmunoassay and Western blotting have been applied and shown to be the most reliable methods. Since identification of the antigen on a molecular level, Elisas using recombinant antigens are also available.

It is important to state that the demonstration of antinuclear antibodies in a chronic liver disorder is not diagnostic for AIH — especially when IFT on cell cultures (for instance Hep2 cells) is used instead of cryostat sections. In IFT on cell cultures, naturally occurring ANA induced during infectious or toxic processes are quite frequently observed. Furthermore, the ANA should be differentiated. For instance, antibodies to nuclear dots (sp100), nuclear membrane (gp210) or centromeres are rather more indicative of primary biliary cirrhosis than AIH.

For serological diagnosis, several assays are commercially available using recombinant antigens. However, antinuclear antibodies and antibodies to actin can-

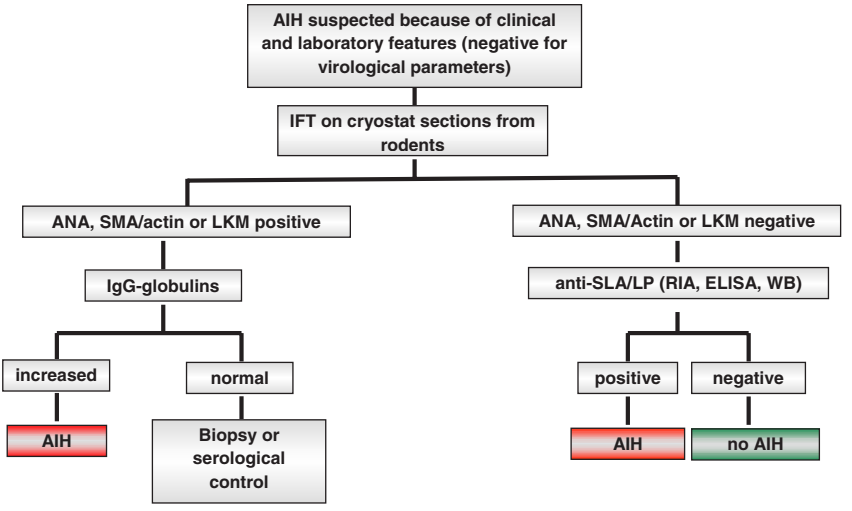

Figure 2. Flow chart for the serological diagnosis of autoimmune hepatitis.

not be reliably detected with those tests. The serological diagnosis of AIH should be, therefore, always proven by specialised laboratories.

References

[1] Czaja AJ, Manns MP. Advances in the diagnosis, pathogenesis, and management of autoimmune hepatitis. Gastroenterology 2010; 139: 58–72.
[2] Krawitt EL. Autoimmune hepatitis. N Engl J Med 2006; 354: 54–66.
[3] Hennes EM, Zeniya M, Czaja AJ, et al. Simplified criteria for the diagnosis of autoimmune hepatitis. Hepatology 2008; 48: 169–76.
[4] Stechemesser E, Klein R, Berg PA. Characterization and clinical relevance of liver-pancreas antibodies in autoimmune hepatitis. Hepatology 1993; 18: 1–9.
[5] Jothimani D, Cramp ME, Mitchell JD, Cross TJS. Treatment of autoimmune hepatitis: a review of current and evolving therapies. J Gastroenterol Hepatol 2011; 26: 619–27.