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Comparison of simplified score with the revised original score for the diagnosis of autoimmune hepatitis: A new or a complementary diagnostic score?

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ABSTRACT

Background and aims: The International Autoimmune Hepatitis Group developed a simplified score for autoimmune hepatitis. We assessed this "new scoring system" and compared it with the International Autoimmune Hepatitis Group original revised score.
Methods: 502 patients were evaluated namely, 428 had liver diseases of various etiology [hepatitis B ($n=109$), hepatitis C ($n=100$), hepatitis D ($n=4$), alcoholic liver disease ($n=28$), non-alcoholic fatty liver disease ($n=55$), autoimmune cholestatic diseases ($n=77$), liver disorders of undefined origin ($n=32$) and miscellaneous hepatic disorders ($n=23$)], 13 had autoimmune hepatitis/overlap syndromes, 18 had autoimmune hepatitis/concurrent with other liver diseases and 43 had autoimmune hepatitis.
Results: The specificity of the simplified score was similar to that of the revised score (97% vs. 97.9%). The sensitivity in unmasking autoimmune hepatitis in autoimmune hepatitis/overlap syndromes was also similar in both systems (53.8% and 61.5%). However, the sensitivity for autoimmune hepatitis diagnosis in autoimmune hepatitis patients with concurrent liver disorders was lower by the new score ($p=0.001$). Liver biopsy proved to be the only independent factor for unmasking autoimmune hepatitis component among patients ($p=0.003$).
Conclusion: The simplified score is a reliable and simple tool for excluding autoimmune hepatitis. However, both systems cannot unmask autoimmune hepatitis component efficiently in autoimmune hepatitis patients with concurrent autoimmune or non-autoimmune liver diseases. This study also strongly reiterates the importance of liver biopsy in the work-up of patients.

1. Introduction

Autoimmune hepatitis (AIH) is an inflammatory condition of unknown aetiology that can affect patients of all ages, sexes and races, characterized by interface hepatitis on liver biopsy, hypergammaglobulinaemia, autoantibody detection and good responsiveness to immunosuppressive therapy [1–5]. The heterogeneity of its clinical presentation and the absence of specific markers applicable for all patients make sometimes the correct and timely diagnosis difficult [6,7]. Furthermore, the diagnosis and consequently the management are more complicated in overlap syndromes such as AIH/primary biliary cirrhosis (PBC) or AIH/primary sclerosing cholangitis (PSC) as well as in cases with coincidence of AIH and other liver diseases such as chronic viral hepatitis (CVH), non-alcoholic fatty liver disease (NAFLD) and alcoholic liver disease.

The diagnostic criteria for AIH have been codified by the International Autoimmune Hepatitis Group (IAIHG) [8], in an attempt to provide uniform and standard criteria for the diagnosis of relatively homogenous groups of AIH patients for both clinical and research purposes. However, a major limitation of the revised scoring system concerning the diagnosis of AIH was its complexity [8]. In order to resolve these difficulties, the IAIHG developed a simplified scoring system based on four components [9]. This simplified score showed a high degree of sensitivity (88%) and specificity (97%) [9,10]. It was acknowledged however in that report [9] that these recommendations would require further validation by prospective evaluation, especially in patients with characteristics of two autoimmune liver diseases. In this context, Czaja [10] compared the performance parameters of both scoring systems in a large series of patients diagnosed with several liver disorders, but did not include "overlap" patients, while Yeoman et al. [11] analyzed a large cohort

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of patients with AIH and non-AIH aetiologies including patients with AIH/overlap syndromes but no cases with concurrent AIH and chronic viral hepatitis, NAFLD or alcoholic liver disease. Very recently, Muratori et al. [12] also validated the simplified diagnostic criteria for AIH in Italian patients with AIH, PBC, chronic hepatitis C (CHC), including a fair number of hepatitis C virus (HCV)-infected patients positive for liver kidney microsomal antibodies (anti-LKM) and patients with AIH/PBC overlap syndrome.

We have recently reported [13] in a large cohort of patients the effectiveness of AIH revised score in patients with AIH/PBC, AIH/PSC overlap syndromes and in patients with coincidence of AIH with other liver diseases. Accordingly, the aim of the present study was to assess the applicability of the "new simplified score" [9] for AIH diagnosis in the same study population with heterogeneous groups of chronic liver diseases ($n = 490$) after the addition of 12 (total number of patients included: 502) "difficult" cases (5 HCV-positive/anti-LKM-positive and 7 with concurrence of AIH and any kind of liver disorder) and compare the results with those obtained by the IAIHG revised score published in 1999 [8]. Special attention has been paid in the group of patients with AIH associated or not with liver disorders of autoimmune origin, as both scoring systems have not been originally validated in such group of patients [14].

2. Materials and methods

This retrospective study included 459 patients with liver diseases of various etiologies and 43 controls with documented AIH followed in our centre over a 5-year period (1999–2004). The medical records of patients were systemically reviewed with respect to the data required to complete the "new simplified scoring system" using data at the time of first visit in the outpatient clinic or first admission of each patient in our department. In our previous study [13] the IAIHG codified scoring system was estimated for these patients at the same time points. All patients were evaluated in a uniform fashion and diagnosis was assigned according to established diagnostic criteria [2–4,6,8,15–23] by two independent experienced investigators (G.N.D. and K.Z.) as we described in detail in our previous report [13]. The human research review committee of the University of Thessaly, Medical School approved the study protocol.

In brief, the diagnosis of chronic hepatitis C was based on clinical, laboratory and histologic evaluations [13,17]. Actually, all CHC patients included in the study met the following criteria: (a) serological evidence of chronic hepatitis C virus (HCV) infection as determined by the detection of antibodies to HCV (anti-HCV), using a third-generation enzyme immunoassay at least twice within 6 months and (b) active virus replication as defined by HCV RNA detection using a polymerase chain reaction. The diagnosis of chronic hepatitis B (CHB) and hepatitis D (CHD) was based on clinical, laboratory and histologic evaluations as described previously [17,23] and according to EASL International Consensus Conference on Hepatitis B [18]. Patients with PBC met the following criteria: elevated cholestatic enzymes, liver histology with PBC lesions and positivity for antimitochondrial antibodies (AMA, positive titre $\geq$ 1:40) detected by indirect immunofluorescence (IIFL) on in-house rodent tissue substrates and confirmed by a competitive enzyme linked immunosorbent assay (ELISA) with reference to antibody directed against the E2 subunit of pyruvate dehydrogenase or of the branched chain keto acid dehydrogenase and by Western blotting using rat liver mitochondrial fractions following published protocols [2–4,13,19–21]. The diagnosis of PSC was based on biochemical or clinical signs of cholestasis, compatible liver histology, repeatedly AMA negativity by IIFL, Western blot or ELISA and/or typical findings on endoscopic retrograde cholangiopancreatography (ERCP) or magnetic resonance cholangiography (MRCP) [20,21]. The diagnosis of NAFLD was based on the presence of metabolic syndrome, exclusion of other causes of chronic liver disease (viral, autoimmune, drug and toxin induced) including alcohol abuse, and compatible liver histology [22]; while the presence of alcoholic liver disease was documented on the grounds of history of increased alcohol consumption and compatible laboratory and histological lesions. Patients with liver disorders of undefined origin ($n = 32$) consisted of subjects with no confirmed diagnosis, in spite of a thorough clinical and laboratory investigation including liver biopsy and immunoserological assessment. Patients with miscellaneous hepatic disorders ($n = 23$) included Wilson's disease ($n = 3$), secondary haemochromatosis ($n = 5$), Budd-Chiari syndrome ($n = 2$), drug-induced hepatitis ($n = 5$), benign liver tumours ($n = 5$) and one of each of the following conditions: a_1-antithrypsin deficiency, benign cholestasis of pregnancy and Gilbert's syndrome ($n = 3$). The diagnosis of AIH was based on the revised descriptive criteria reported by the IAIHG in 1999 [8], while patients with AIH/overlap syndromes fulfilled the criteria for AIH diagnosis as well as those for the diagnosis of either PBC or PSC. Specifically, AIH/PBC and AIH/PSC overlap syndromes were defined by the coexistence of AIH and PBC or PSC diagnosed either simultaneously or consecutively according to previously described criteria [15,24,25]. Actually, patients fulfilled at least two of the three criteria for each disease. AIH criteria: (1) positivity for autoantibodies (antinuclear, ANA; smooth muscle antibodies, SMA; or anti-LKM); (2) elevated serum IgG levels and (3) liver biopsy showing moderate or severe periportal or periseptal lymphocytic piecemeal necrosis; PBC criteria: (1) cholestatic biochemical profile; (2) positivity for AMA and (3) histological lesions compatible with PBC; PSC criteria: (1) cholestatic biochemical profile; (2) specific cholangiographic features on ERCP or MRCP characteristic of PSC and (3) histological changes characteristic of PSC. The diagnosis of coincidental existence of AIH and CVH, NAFLD or alcoholic liver disease was based on the descriptive criteria for AIH diagnosis [8] along with those for chronic viral hepatitis (CVH), NAFLD and alcoholic liver disease as mentioned above [16,20]. In particular, the diagnosis of cases with AIH and concurrent CVH was based on the criteria described in detail by us previously [16,17,20,23]. In brief, all these cases had serological and virological evidence of HBV, HCV or HDV infections, while they had increased levels of aminotransferases with normal cholestatic enzymes, diffuse hypergammaglobulinaemia, detectable ANA, SMA or anti-LKM in high titres, seronegativity for AMA, absence of other aetiological factors such as, alcohol consumption or use of known hepatotoxic drugs and a liver biopsy with characteristic lesions of AIH [8,13].

Accordingly, based on their clinicopathological features, the patients of the study ($n = 502$) were divided into three groups:

(i) The "*other*" group consisted of 428 patients with diverse liver diseases excluding AIH. In more detail, 109 had CHB; 100 had CHC (10 patients positive for anti-LKM); 4 had CHD; 28 had alcoholic liver disease; 55 had NAFLD; 51 had PBC; 26 had PSC (in total, 77 patients with autoimmune cholestatic liver diseases, ACLD); 32 suffered from liver disorders of undefined origin and 23 had miscellaneous hepatic disorders as described above.

(ii) The "*combined*" group consisted of 31 patients with AIH associated with any kind of liver disorders, including 10 patients with AIH/PBC, 3 with AIH/PSC (in total, 13 patients with AIH/overlap syndromes), 4 with AIH/CHB, 4 with AIH/CHC and 2 with AIH/CHD. Six patients suffered from concurrent AIH/NAFLD and 2 from AIH/alcoholic liver disease (in total, 18 patients had coincidence of AIH with viral, NAFLD or alcoholic liver disease).

(iii) The "*control*" group consisted of 43 patients with documented AIH (disease control group).

Table 1
Characteristics of the study population.

	"Other" group $n=428$ (%)	"Combined" group $n=31$* (%)	AIH/overlap syndromes $n=13$ (%)	AIH and other liver diseases $n=18$ (%)	"Control" group $n=43$ (%)
Age (mean ± SD, years)	51.4 ± 22.5	53.5 ± 18.5	50.6 ± 21.2	54.8 ± 16.1	52.3 ± 15.7
Sex (F/M)	234/194 (54.7/45.3)	18/13 (58.1/41.9)	8/5 (61.5/38.5)	10/8 (55.6/44.4)	33/10 (76.7/23.3)
Alcohol abuse > 60 g/day (yes/no)	98/330 (22.9/77.1)	3/28 (9.7/90.3)	0/13 (0/100)	3/15 (16.7/83.3)	1/42 (2.3/97.7)
Drug use (yes/no)	25/403 (5.8/94.2)	0/31 (0/100)	0/13 (0/100)	0/18 (0/100)	1/42 (2.3/97.7)
Other autoimmune diseases (yes/no)	24/404 (5.6/94.4)	7/24 (22.6/77.4)	5/8 (38.5/61.5)	2/16 (11.1/88.9)	18/25 (41.9/58.1)
Serum globulin or IgG above normal (pos/neg; pos = score > 1)	204/224 (47.7/52.3)	21/10 (67.7/32.3)	9/4 (69.2/30.8)	12/6 (66.7/33.3)	34/9 (79.1/20.9)
ANA, SMA or anti-LKM (pos/neg; positive titre ≥ 1:40)	340/88 (79.4/20.6)	30/1 (96.8/3.2)	12/1 (92.3/7.7)	18/0 (100/0)	43/0 (100/0)
AMA (pos/neg; positive titre ≥ 1:40)	49/379 (11.4/88.6)	4/27 (12.9/87.1)	4/9 (30.8/69.2)	0/18 (0/100)	0/43 (0/100)
Anti-SLA/LP (pos/neg)	0/428 (0/100)	0/31 (0/100)	0/13 (0/100)	0/18 (0/100)	5/38 (11.6/88.4)
Hepatitis viral markers (pos/neg)	213/215 (49.8/50/2)	10/21 (32.3/67.7)	0/13 (0/100)	10/8 (55.6/44.4)	0/43 (0/100)
Histology (pos/neg)	61/367 (14.3/85.7)	18/13 (58.1/41.9)	5/8 (38.5/61.5)	13/5 (72.2/27.8)	35/8 (81.4/18.6)
Aggregate simplified score (mean ± SD)	3.4 ± 1.3	5 ± 1	5.4 ± 1.2	4.8 ± 0.9	6.6 ± 1.1

Abbreviations are same as in text; F = female; M = male.

* Combined group ($n=31$) with coincidence of AIH and any kind of liver disorder is divided to patients with AIH/overlap syndromes ($n=13$) and patients with coincidence of AIH and other liver diseases ($n=18$); for statistical significances see Section 3.3.

The new simplified scoring system assigns a score for each of the following parameters: serum autoantibodies, serum immunoglobulin G (IgG) level, liver histology and absence of viral hepatitis [9]. According to our previous publications, non-organ specific autoantibodies (ANA, SMA, and anti-LKM) were detected by IIFL on HEp-2 cells and in-house rat liver–kidney–stomach cryostat sections using standard protocols and accepting titers ≥1:40 as positive in all cases [2,3,26–28]. Commercially available ELISA was used for the detection of anti-SLA autoantibodies according to the manufacturer's instruction. Liver tissue specimen of adequate size was obtained from each patient by needle biopsy and the histological findings were interpreted blindly by an experienced hepatopathologist (G.K.K.). A diagnosis of "definite" AIH requires an aggregate score equal to or greater than 7, while a score equal to 6 denotes "probable" AIH [9]. In order to compare each parameter of the simplified scoring system, patients were divided into two groups [positive (pos)/negative (neg)] as follows: autoantibodies (pos: ANA, SMA, LKM titer ≥ 1:40 and/or positivity for SLA; neg: no detection of autoantibodies), IgG (pos: above the upper normal limit; neg: lower than the upper normal limit), liver histology [pos: either typical or compatible with AIH according to Hennes et al. [9]; neg: atypical [9]], viral hepatitis (pos: absence of viral markers/neg: presence of viral markers).

2.1. Statistical analysis

Results are expressed as mean ± SD. Data were analyzed by x^2 (two-by-two with Yate's correction), Fisher's exact test, Kruskal–Wallis test and Wilcoxon–Mann–Whitney test where applicable. The parameters that were significant in the univariate analysis entered a binary logistic regression model, in order to identify independent risk factors. p-Values < 0.05 were considered statistically significant.

3. Results

The demographic, clinical, biochemical and histological features required for the calculation of the two scoring systems in the study population are shown in Table 1.

Table 2
Specificity of the revised and the simplified scoring system in patients with diverse liver disorders other than AIH ($n=428$).

Liver diseases	N	Specificity of the revised scoring system	Specificity of the simplified scoring system	p
HBV	109	108/109 (99.1%)	109/109 (100%)†	1.000
HCV	100	98/100 (98%)	100/100 (100%)†	0.497
HDV	4	4/4 (100%)	4/4 (100%)	1.000
Alcoholic liver disease	28	28/28 (100%)	24/28 (85.7%)	0.111
NAFLD	55	53/55 (96.4%)	52/55 (94.5%)	1.000
PBC	51	50/51 (98%)	48/51 (94.1%)	0.617
PSC	26	24/26 (92.3%)	23/26 (88.5%)	1.000
Undefined	32	31/32 (96.9%)	32/32 (100%)	1.000
Miscellaneous	23	23/23 (100%)	23/23 (100%)	1.000
Overall	428	419/428 (97.9%)	415/428 (97%)	0.564

Abbreviations are same as in the text; N = number of patients studied in each group.

† Specificity was significantly higher for HBV, or HCV patients compared to alcoholic liver disease ($p<0.01$), PSC ($p<0.01$), NAFLD ($p<0.05$), and PBC patients ($p<0.05$).

Table 3
Sensitivity of the revised and the simplified scoring system in patients with AIH associated with any kind of liver diseases ("combined" group, $n = 31$).

	N	Diagnosis by Scoring system				Sensitivity of the revised scoring system	Sensitivity of the simplified scoring system	p
		Definite AIH		Probable AIH				
		Revised	Simplified	Revised	Simplified			
AIH/overlap	13	2	1	6	6	8 (61.5%)	7 (53.8%)	
AIH/PBC	10	2	1	5	6	7 (70%)	7 (70%)	1.000
AIH/PSC	3	0	0	1	0	1 (33.3%)	0 (0%)	
AIH/viral	10	0	0	8	1	8 (80%)	1 (10%)	
AIH/HBV	4	0	0	3	0	3 (75%)	0 (0%)	
AIH/HCV	4	0	0	3	1	3 (75%)	1 (25%)	0.001
AIH/HDV	2	0	0	2	0	2 (100%)	0 (0%)	
AIH/NAFLD	6	0	1	4	0	4 (66.7%)	1 (16.7%)	
AIH/alcoholic liver disease	2	0	0	1	0	1 (50%)	0 (0%)	
Overall	31	2	2	19	7	21/31 (67.7%)	9/31 (29%)	0.005

Abbreviations are same as in the text. N = number of patients studied in each group.

3.1. Specificity and sensitivity of the revised original and the simplified score

Table 2 shows the specificity of both scoring systems in each liver disease and overall in the group of patients with liver disorders other than AIH ($n = 428$). In fact, the overall specificity as well as the specificity for each liver disease (true negative/true negative+false positive) of the new simplified scoring system for excluding AIH in this group of patients ($n = 428$) was similar compared with that obtained by using the IAIHG revised score (not statistically significant differences; Table 2). However, 13 out of 428 patients had an AIH simplified score equal to 6 (probable AIH), while none had a score equal to or greater than 7 (definite AIH) [9]. This subgroup of patients consisted of 4 patients with alcoholic liver disease, 3 with NAFLD and 6 with ACLD (3 PBC and 3 PSC). All of these patients achieved 2 points for more than 1.1×IgG increase, presence of autoantibodies characteristic of AIH and absence of markers of hepatitis virus infections, while none took points for a compatible or typical AIH liver biopsy. Of note, none of the 10 HCVpos/anti-LKMpos patients achieved a simplified score for probable or definite AIH though the IAIHG revised score failed to rule out AIH in one patient (100% vs. 90%; $p = 1.000$).

The sensitivity (true positive/true positive+false negative) of the new simplified scoring system for detecting AIH in association with any kind of other liver disease ("*combined*" group; $n = 31$) was 29%, while the revised scoring system achieved a sensitivity of 67.7% ($p = 0.005$) (Table 3). This difference was mainly due to the group of AIH patients with coincident viral, alcoholic or fatty liver disease (Table 3). Only 9 patients (7 with AIH/PBC, 1 with AIH/CHC and 1 with AIH/NAFLD) achieved an aggregate simplified score ≥6. The remaining 22 patients (3 with AIH/PBC, 3 with AIH/PSC, 4 with AIH/CHB, 3 with AIH/CHC, 2 with AIH/CHD, 5 with AIH/NAFLD and 2 with AIH/alcoholic liver disease) had an AIH simplified score less than 6. Of these 22 false negative patients, 10 (45.5%) had normal levels of IgG, 1 (4.5%) had a negative score for the presence of autoantibodies, 9 (40.9%) presented with concurrent viral hepatitis and only 13 (59.1%) had a liver biopsy compatible or typical for AIH. Similar sensitivity was revealed from both scoring systems in detecting AIH in patients with AIH/PBC or AIH/PSC overlap syndromes (61.5% vs. 53.8%; not statistically significant difference; Table 3).

3.2. Parameters associated with increased AIH simplified score in patients with chronic liver diseases ("other" group; n = 428)

The univariate analysis showed that increased IgG levels and absence of viral hepatitis markers were significantly more frequent in patients with liver diseases other than AIH who achieved a simplified score equal to six ($n = 13$) compared to those ($n = 415$) having a negative score (100% vs. 46%, $p < 0.001$ and 100% vs. 48.7%, $p = 0.001$, respectively). However, the application of the binary logistic regression analysis demonstrated that none of these parameters could be considered as an independent factor associated with a probable simplified AIH score in non-AIH patients with chronic liver diseases.

3.3. Comparison between groups

Table 1 shows the comparison between subgroups of the study population. A liver biopsy compatible or typical for AIH was significantly more frequent in patients with AIH/overlap syndromes and AIH with concurrent other liver diseases ("*combined*" group, $n = 31$) compared to those with chronic liver diseases other than AIH (18/31; 58.1% vs. 61/428; 14.3%, $p < 0.001$, Table 1).

After subdivision of the "*combined*" group ($n = 31$) into the subgroup of patients with AIH/overlap syndromes ($n = 13$) and the patients with coexistence of AIH and other liver diseases ($n = 18$) and comparison of the parameters of the simplified scoring system with the "other" group of patients ($n = 428$), we found that (Table 1): (a) patients with AIH/overlap syndromes had significantly lower prevalence of viral hepatitis markers compared to the group of patients with chronic liver diseases other than AIH (0/13; 0% vs. 213/428; 49.8%, $p = 0.001$), and those with AIH concurrent with other liver diseases(0/13; 0% vs. 10/18; 55.6%, $p = 0.001$), and (b) patients with AIH concurrent with other liver diseases had significantly more frequently a liver biopsy compatible or typical with AIH (13/18; 72.2%) compared to patients with other liver diseases(61/428; 14.3%, $p < 0.001$).

Patients with AIH/overlap syndromes and coexistence of AIH with other liver diseases ("*combined*" group; $n = 31$) had higher prevalence of viral hepatitis markers (10/31; 32.3% vs. 0/43; 0%, $p < 0.001$) and higher frequency of an atypical liver biopsy (13/31; 41.9% vs. 8/43; 18.6%, $p = 0.05$) compared with patients suffering from AIH ("*control*" group; $n = 43$; Table 1). In addition, after subdivision of the "*combined*" group ($n = 31$) into the two subgroups of patients (AIH/overlap syndromes, $n = 13$ and AIH/other liver diseases, $n = 18$) and comparison of the parameters of the simplified score with the group of patients with AIH ("*control*"; $n = 43$), we found that (Table 1): (a) patients with AIH/overlap syndromes had significantly lower possibility of having a liver biopsy compatible or typical for AIH (5/13; 38.5% vs. 35/43; 81.4%; $p = 0.005$) compared to AIH patients and (b) patients with coincidence of AIH and other liver disease had significantly higher prevalence of viral hepatitis markers (10/18; 55.6% vs. 0/43; 0%, $p < 0.001$) compared to AIH patients.

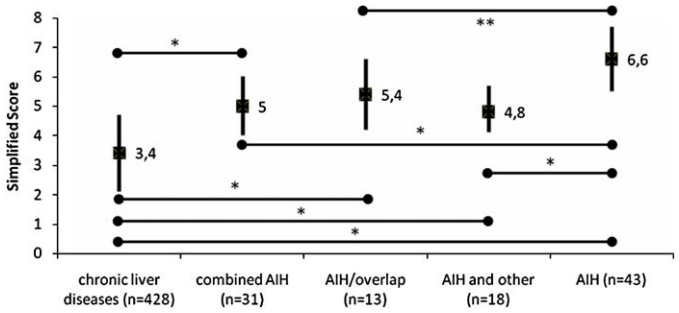

Fig. 1. Mean ± SD simplified score in each subgroup of patients; $^{*}p < 0.001$, $^{**}p = 0.003$.

However, the binary logistic regression analysis showed that a typical or compatible liver biopsy for AIH was the only independent factor that was able to differentiate the presence of isolated AIH from either the AIH/overlap or the coexistence of AIH with other liver disorders ($p = 0.003$).

3.4. Aggregate scores of AIH in the groups of patients

The mean simplified AIH score was significantly different among the four groups of the study ($p < 0.001$, Kruskal–Wallis test) (Fig. 1). The score in the "*combined*" group of patients with coexistence of AIH and any kind of liver disease ($n = 31$) was significantly higher (5 ± 1) compared to that found in the group of patients with liver diseases other than AIH (3.4 ± 1.3; $n = 428$; $p < 0.001$), but significantly lower than the simplified score observed in the "*control*" group of patients with "pure" AIH (5 ± 1; $n = 31$ vs. 6.6 ± 1.1; $n = 43$, $p < 0.001$). The simplified score in the "other" group was significantly lower than the score calculated in the "control" group (3.4 ± 1.3; $n = 428$ vs. 6.6 ± 1.1; $n = 43$, $p < 0.001$). The AIH simplified score for each subgroup of patients, AIH/overlap (5.4 ± 1.2; $n = 13$) and patients with coincidence of AIH and other liver disease (4.8 ± 0.9; $n = 18$) was significantly higher compared to that found in the group of patients with chronic liver diseases (3.4 ± 1.3; $n = 428$, $p < 0.001$) and significantly lower than the AIH simplified score observed in the group of patients with AIH (6.6 ± 1.1; $n = 43$, $p = 0.003$ and $p < 0.001$, respectively).

4. Discussion

In order to evaluate the simplified scoring system, we applied it to a large cohort of patients suffering from heterogeneous groups of chronic liver diseases ($n = 502$) including overlap syndromes and patients with both AIH and other liver diseases. However, it is well known in medicine that diagnostic scoring systems are by their nature not designed for all. The reason to evaluate them in this way is because non-specialists may choose to apply the scoring systems anyway, and therefore highlighting pitfalls is important. In this context, we found that the new simplified criteria are equally reliable to exclude AIH in patients with several liver diseases as the original IAIHG scoring system (specificity 97% vs. 97.9%, respectively). This high specificity of the new score has also been reported by two tertiary referral centres recently [11,12]. Actually in our study, only 13 out of 428 patients (3%) with a definite liver disorder were falsely evaluated as probable AIH by the new simplified score. Of note however, none of these 13 patients had compatible liver biopsy for AIH indicating that liver biopsy is an essential diagnostic procedure for excluding AIH [4,6,8–10,13,29]. Indeed, separate analysis between subgroups identified liver biopsy as the only independent factor for distinguishing patients with AIH/overlap syndromes or AIH with concomitant liver diseases from patients suffering from liver diseases other than AIH.

In addition, in our study the simplified score had very high specificity for the exclusion of probable or definite AIH (100%) concerning the special subgroup of chronic HCV patients positive for anti-LKM autoantibodies. In concordance, Muratori et al. [12] have described in a series of 38 HCV-positive/anti-LKM-positive patients, 6 with a diagnosis of probable AIH (specificity of the simplified score for probable AIH: 84.2%) but none with a diagnosis of definite disease (specificity of the simplified score for definite AIH: 100%). In these patients, it is mandatory to exclude AIH, because treatment with interferon could unmask or provoke autoimmune hepatic reactions or even "true" AIH, especially in patients with a probable simplified score; in this case a liver biopsy and a reconsideration of the prominent liver disorder – though difficult – should be performed [27,30].

The sensitivity of the simplified scoring system to detect AIH as a component of the overlap syndromes though low (53.8%) was similar to that obtained by the IAIHG revised score (61.5%). Muratori et al. [12] demonstrated that the simplified score was rather efficacious even for the detection of AIH component in 15 patients with AIH/PBC overlap syndromes (14/15 of patients with simplified score ≥ 6; sensitivity 93.3% vs. 70% in our AIH/PBC series; Table 3). However, this difference was not statistically significant ($p = 0.267$) due to the low number of AIH/PBC cases analyzed in both of the abovementioned studies. On the contrary, the new scoring system was able to detect only 2 out of 18 patients with coincidence of AIH with non-autoimmune liver diseases, while the IAIHG score identified 13 of these 18 patients; thus the simplified score cannot detect for example, patients with coexistence of AIH and CVH. The latter could be a problem for areas with high endemicity of CVH infections like for instance the Mediterranean basin, where the possibility of suffering from both diseases (AIH and CVH) cannot be excluded [31–38]. In a previous study of the IAIHG the simplified score was found 97% specific and 88% sensitive [9], while recently, Czaja [10] demonstrated that the simplified score system was better at excluding the diagnosis in diseases with concurrent immune manifestations (90% specificity, 95% sensitivity), but both studies did not include "overlap" patients. These findings along with the findings of the present study suggest that the new simplified score is a reliable diagnostic tool for excluding diagnosis of AIH in patients with several distinct liver disorders achieving a specificity ranging from 90% to 97%, but as also the original IAIHG revised score, it is rather inconvenient to detect AIH concurrence with other liver diseases, of autoimmune origin or not.

Diagnosis of AIH is always challenging, and especially in patients with concurrent autoimmune or non-autoimmune liver diseases, as early and correct diagnosis might be helpful for the final outcome of the patients. However, early diagnosis may be difficult because the clinical picture is heterogeneous and there is no specific test applicable for all patients [7,37]. Standardized diagnostic criteria have not been promulgated; experiences between institutions have not been compared; natural history for each variant form remains uncertain; and treatment algorithms have not been validated. The presence of liver biopsy as an independent discriminative factor in almost all of our comparisons underlines its overall significance in the diagnosis of the AIH component in the individual case. Thus, it must be noticed that liver biopsy should be mandatory in patients with liver disorders in whom there is evidence of autoimmunity and absence of evidence of HBV or HCV markers. Therefore, the cooperation between clinicians and expertise hepatopathologists in an attempt to unmask, or perhaps provoke, the presence of AIH not only in the typical "pure" cases of AIH but also in difficult cases of concurrence with any kind of liver disorder seems mandatory.

In conclusion, we demonstrated that the simplified scoring system has similar specificity for excluding AIH in patients with several liver diseases compared to the former IAIHG revised scoring system. Though there were some differences, both scoring systems cannot

differentiate AIH patients efficiently with concurrent autoimmune or non-autoimmune liver disorders. In these "difficult" cases, the discrepancies between the scoring systems can be attributed to the points awarded or subtracted for features graded in the revised original scoring system but not in the simplified score [8–13]. The revised scoring system might be useful in the evaluation of "difficult" cases with few or atypical features of AIH after exclusion of other diagnoses, while the simplified scoring system – simpler in its determination – can be used in aetiologically distinctive cases with concurrent immune trappings in order to exclude AIH [8–13]. Therefore, it seems rational that each system has different and even complementary roles, and the simplified score may be an expansion rather than a revision of the current diagnostic armamentarium [8–13]. Nevertheless, in any case, this study strongly shows that as there is no single test for AIH, histology is very important where there is any doubt about the diagnosis, and in patients where there may appear to be more than one disease, histology is the best arbiter and guide of treatment.

Conflict of interest statement

None declared.

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