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TITLE PAGE

TITLE: "Liver dysfunction and fibrosis as predictors of biochemical response to autoimmune hepatitis treatment."

SHORT TITLE: "Predictors of response to autoimmune hepatitis treatment."

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ABSTRACT

Aims: AIH (AIH) is a disease that presents itself in various forms, ranging from aminotransferase asymptomatic alteration, acute hepatitis to decompensated cirrhosis. Few studies have evaluated the predictive criteria for response to treatment.

Methods: A cross-sectional analytical study examined patients with AIH who were seen in the hepatology clinic of a university hospital between January 2013 and July 2015.

Results: A total of 36 patients were included. The mean age was 44.7 ± 14.3 years, 22.2% male, and 19.2% of patients presented liver failure. Patients with significant fibrosis had lower mean aminotransferases and bilirubins and higher mean prothrombine activity (PA) than those with insignificant fibrosis. Most patients (94.5%) underwent treatment with azathioprine and prednisone. When comparing individuals who exhibited biochemical response (ALT and AST <55 U/L in the sixth month of treatment) to those without biochemical response, it was observed that those with biochemical response presented minor proportion of patients with significant fibrosis, and these patients had a higher proportion of liver failure in initial presentation and lower mean PA. Furthermore, it was observed that the lower the PA on admission, the lower ALT levels ($r = 0.682$, $p = .020$) and AST ($r = 0.431$, $p = .040$) in the sixth month of treatment.

Conclusions: Individuals with liver dysfunction and elevated aminotransferases show insignificant fibrosis histologically. Patients who open the AIH patients who initially present liver failure and insignificant fibrosis are more likely to develop biochemical response to treatment.

INTRODUCTION

Autoimmune hepatitis (AIH) is a chronic inflammation of the liver with unknown cause. Some research suggests that its pathogenesis is mediated by environmental factors, failures in immune tolerance mechanisms and genetic predisposition [1]. It is most common in women, in the proportion of 3.6:1 [2] and affects different ethnicities [3] and all age groups, with incidence peaks between 10 and 30 years old and the second peak between 30 to 50 years old [4]. The avearage annual incidence of AIH in the Northern European Caucasian population is from 0.9 to 1.9 per 100,000 people, and prevalence from 10.7 to 16.9 per 100,000 people [5, 6].

AIH presents itself clinically with non-specific symptoms such as nausea, abdominal pain, weakness, arthralgia, but it can also be totally asymptomatic [7]. It is known that AIH causes an abnormal increase in serum levels of aminotransferases, autoantibodies, and changes in the concentration of serum immunoglobulin G (IgG). Serological markers of AIH are antinuclear antibodies (ANA), smooth muscle (SMA) and antibody against the microsomal fraction of liver and kidney type 1 (LKM1)[8]. Among the histological findings of AIH are hepatitis interface, necrosis and lobular inflammation, plasma cell infiltration and presence of rosettes [9].

The diagnosis of AIH is clinical and made in the presence of a set of characteristics involving clinical condition, the absence of other causes of liver disease (alcohol, medication, viruses, hemochromatosis, Wilson's disease), coexistence of autoimmune diseases, increased serum globulins, histological features and response to treatment [10]. To facilitate the diagnosis, diagnostic criteria were created in 1993 [11], subsequently revised [12]. Recently, in 2008, a Simplified Score to facilitate the application of the criteria was proposed [13], and

demonstrated greater specificity and accuracy in the diagnosis of AIH than the original score (90% vs. 73% and 92% vs. 72%, respectively) [14].

Treatment AIH is based on the use of corticosteroids and immunosuppressive. The main form of treatment is the use of prednisone alone or in combination with azathioprine [15]. The objective of treatment is to induce clinical remission, normalization of liver biochemistry, reduce histological inflammation and increase survival of patients [16]. In general, the prognosis of patients who respond to immunosuppressive treatment is good, they use low doses of medication and have good life quality [17]. However, after discontinuing the medication, about 70% of patients present recurrence of the disease [18].

Considering the scarcity of studies evaluating AIH treatment response, the aim of this study is to describe the clinical, biochemical and histological characteristics of patients with AIH in southern Brazil, identify the characteristics associated with the presence of significant histological fibrosis and assess these patients treatment response.

METHODS

Cross-sectional study, conducted through medical record review, which evaluated AIH adults carriers assisted at the Gastroenterology and Hepatology Clinic of the University Hospital Polydoro Ernani de São Thiago at the Federal University of Santa Catarina (HU/UFSC), between January 2013 and July 2015. Those with insufficient registration of clinical and laboratory data in the medical records, overlap syndrome or uncertain diagnosis were excluded.

Individuals included in the study were analyzed according to their clinical, laboratory, histological and therapeutic response characteristics. All data were collected from records contained in medical records and transferred to the statistical spreadsheet program *Statistical Package for the Social Science* (SPSS); version 17.0 (Chicago; Illinois; EUA). The following

variables were studied: age; gender; skin color; liver failure; aspartate aminotransferase (AST); alanina aminotransferase (ALT); alkaline phosphatase (ALP); gamma-glutamyl transferase (GGT); direct bilirubin (DB); serum albumin; prothrombine activity (PA)ç globulins; gammaglobulins; surface antigen of hepatitis B virus (HBsAg); core antibody against the hepatitis B virus (anti-HBc); antibody against the surface antigen of hepatitis B virus (anti-HBs); antibody against the Hepatitis C virus (anti-HCV); antinuclear antibodies (ANA), smooth muscle (SMA) and the antibody against the microsomal fraction of liver and kidney type 1 (LKM1). Liver failure was defined as PA less than 40%. ALT and AST were analyzed by Clinical Chemistry System Dimension, with reagents ALT Flex® and AST Flex®, temperature at 37^{o}C. ALP, GGT, DB and albumin were also analyzed by Clinical Chemistry Dimension System ®, temperature at 37^{o}C. The reagent used for ALP was ALPI Flex® and for DB DBI Flex® was used. The PA was analyzed with RecombiPlasTin 2G® kit. For detection ANA, SMA, LKM1 and EMA, we used indirect immunofluorescence (IIF). Hepatic biochemical tests AST; ALT; ALP and GGT were expressed in absolute numbers and were evaluated at different times of treatment: Pre-treatment, 1 month, 3 months, 6 months, 12 months, 24 months and 48 months after treatment initiation. Biochemical Response to treatment was defined as ALT and AST values lower than 55 U/L in the sixth month of treatment.

Regarding liver biopsy, the following histological features were observed: significant fibrosis, defined as portal fibrous expansion with portal-portal and central-portal septa and/or complete nodes (definite cirrhosis).; significant periportal inflammatory activity as defined in piecemeal necrosis that can be discreet or present in large areas of many portal spaces; and significant parenchymal inflammatory activity defined as focal necrosis of hepatocytes surrounded by lymphohistiocytic aggregates in numerous sites with or without confluent necrosis, which can be extensive or multiple. Duplicate of pathology tests performed at the

University Hospital were requested. For AIH diagnostic classification the revised criteria of the International Score and Simplified Score were used [12, 13].

Statistical Analysis

Continuous variables were described as central tendency and dispersion measures; whereas categorical variables were described in absolute numbers and proportions. Continuous variables were compared using the Student t test or Mann-Whitney. Categorical variables were compared using the Fisher's exact or chi-square test when appropriate. The comparative analysis of biochemical variables before and after treatment was performed by paired t test. Bivariate analysis was performed to identify the characteristics associated with significant fibrosis and biochemical response. Spearman correlation analysis was performed to identify whether the aminotransferases at the sixth month of treatment were correlated with PA admission. Values of P lower than 0.05 were considered statistically significant. All tests were bicaudal and performed by SPSS; version 17.0 (SPSS; Chicago; Illinois; EUA).

RESULTS

Case by case analysis

Forty-five patients with AIH, assisted during the study period, were evaluated for inclusion, and 9 were excluded from the study: 5 individuals because they presented insufficient data , 3 patients were excluded because they were diagnosed with overlap syndrome and one for not having definite diagnosis of AIH.

The study included 36 patients with a mean age, standard deviation and median of 44.7 ± 14.3 (46.5) years old, 22.2% were men, and 97.1% declared themselves white. About one-fifth of patients (19.2%) exhibited liver failure in the disease initial presentation. No patient exhibited positive serology for hepatitis viruses B and C.

Among 36 patients, 24 patients (66.7%) were classified by the International Score as probable diagnosis and 6 patients (16.7%) had definitive diagnosis. According to the Simplified Score, 15 patients (16.7%) had probable diagnosis and 10 patients (27.8%) had definitive diagnosis.

Regarding the laboratory characteristics, subjects showed mean, standard deviation and median: ALT of 383.8 ± 368.6 (276.0) U/L; AST of 476.4 ± 505.6 (320.0) U/L; Direct Bilirubin of 4.3 ± 5.1 (1.7) mg/dL; albumin of 3.2 ± 0.8 (3.5) g/dL; PA of 57.6 ± 21.9 (60.0) and gamma globulins of 3.1 ± 4.6 (1.9) g/dL. Clinical and laboratory characteristics of patients included are shown in Table 1.

Regarding antibodies, 21 (58.3%) had positive ANA with titles ranging from 1:80 to 1:1280, 80% with speckled pattern. Among the 19 whose ANA standard was available, 15 (78.9%) had the fine speckled pattern. SMA was present in 55.6%, 5.6% were LKM-1 positive. Some patients had more than one positive autoantibody. The distribution of autoantibodies is shown in Figure 1.

Thirty-one patients underwent liver biopsy and 11 (35.5%) had cirrhosis (Table 2). Significant fibrosis was observed in 77.4%. About half the sample (48.4%) had interface hepatitis, 51.6% exhibited plasma cells and 16.1% rosettes.

Characteristics associated with the presence of significant fibrosis

When comparing individuals with significant fibrosis to those with insignificant fibrosis it has been observed that patients with significant fibrosis had lower mean ALT (295.9 ± 240.5 vs. 599.4 ± 351.6 U/L; P = 0.038), AST (315.2 ± 341.9 vs. 893.8 ± 640.2 U/L; P = 0.039), and bilirubin (2.9 ± 3.0 vs.8.3 ± 7.5 mg/dL; P = 0.013), and highest PA mean (65.2 ± 21.0 vs. 40.5 ± 19.3%; P = 0.020). There was no difference in relation to age gender, race, liver failure, AntiHBc (+),ANA, SMA, LKM1, ALP/ALT ≥ 1.5 ratio, ALP, albumin, PA and gammaglobulins.

Analysis of treatment

Most patients (94.5%) were subjected to treatment with azathioprine and prednisone, and only two patients received monotherapy with Prednisone. The development of hepatic biochemical tests can be seen in Figure 2, which shows a substantial reduction in ALT, AST, ALP and GGT levels 48 months after beginning the treatment.

Characteristics associated with biochemical response at six months of treatment

When comparing individuals who have levels of ALT and AST lower than 55 U/L in the sixth month of treatment to those without biochemical response it has been observed that patients with biochemical response had lower PA means (41.0 ± 22.5% vs. 66.4 ± 16.2%; P = 0.003) and a minor proportion of patients with significant fibrosis (53.8% vs. 88.2%; P = 0.049). Among the individuals who had biochemical response at six months of treatment, there was a higher proportion of liver failure in the disease initial presentation (44.4% vs. 5.9%; P = 0.034). There was no difference regarding age, gender, skin color, positive ANA, ANA title, positive SMA, SMA title, positive LKM1, and gamma globulins values, DB, albumin and PA. There was no difference regarding the presence of significant periportal inflammatory activity and significant parenchymal activity.

In Spearman correlation analysis (Figure 3) it is observed that the lower the PA on admission, the lower the ALT levels (r = 0.682; P = 0.020) and AST (r = 0.431; P = 0.040) in the sixth month of treatment (Table 3).

DISCUSSION

The characteristics of this sample are similar to those previously described for people with AIH: mean age of 40-50 years old, and prevalence of females between 70-80% [2].

Although AIH is present in all ethnic groups, the sample showed a prevalence of white skin color for patients with AIH in accordance with described by Ngu, New Zealand [19]. Noticeably, it was observed that almost 100% of patients reported themselves Caucasians, especially in a country of admixture such as Brazil. Possibly because in southern Brazil, where this study took place, colonization is predominantly of European origin and most of the population is white [20].

In this sample, 66.7% of patients were classified by the International Score as probable diagnosis and 16.7% had definitive diagnosis. On the other hand, according to the Simplified Score, 16.7% had probable diagnosis and 27.8% had definitive diagnosis. In other studies, in Mexico, the frequency of definitive diagnosis of AIH by Simplified Score and International Score was 41% and 40%, respectively. For probable diagnosis, 59% and 29%, respectively. The International Score exhibited a sensitivity of 95% and specificity of 90% and the simplified score showed a lower sensitivity (65%), but greater specificity (100%) [21]. In a study conducted by Qiu, in China, the definitive diagnosis according to International Score and simplified Score frequency found were 66% and 61%, respectively. Probable AIH 34% and 29%, respectively [22]. In general, the score is useful in cases where the clinical presentation and laboratory results are clearly typical AIH, as in borderline cases the scores are not sufficient to provide certainty at clinical diagnostic [23]. In the present study, it was observed that a smaller proportion of patients were classified as having AIH by the Simplified Score than by the International Score, possibly because 27.8% of subjects did not undergo liver biopsy at diagnosis due to bleeding disorders, and the biopsy has greater importance to diagnose by the Simplified Score which involves fewer variables. Although performing liver biopsy is recommended for all patients with AIH, histological findings have little impact on the conduct and patients progress [24]. Unfortunately, transjugular liver biopsy is not

available in this service to be performed in patients with coagulopathy, but this situation reflects the reality of many public reference hospitals in the world.

In the United States of America (USA) ANA is the most prevalent antibody (78%) followed by SMA (70%), similar to that described in our study (58% and 56% respectively), but in Brazil ratios are inverse, showing SMA antibody as most prevalent (87%) followed by ANA (57%). Interestingly, the proportion of ANA described in Brazil (57%) is equal to that found in our study (58%). When evaluating the antibodies present in isolation, the ratios remains and SMA is the most common antibody in Brazil (44%), while the ANA is the most prevalent antibody alone in the USA (31%), similarly described in our study (33%).

The prevalence of ANA in our sample features a higher prevalence of AIH Type 1. Gupta et al. report that the type 1 AIH is the main type of AIH, and represents 80% of cases (25), similar to the proportions shown in this study, of 86%. AIH has a preponderance in females compared to males in a ratio of 4:1 for type 1 AIH and 10:1 for type 2 AIH [25].

Different from Cançado et al.[26] describing that the presence of isolated ANA is a less aggressive disease marker, when comparing individuals with significant fibrosis to the others, there was no difference between the proportion of individuals with positive ANA among individuals with insignificant fibrosis and those with significant fibrosis (60.9% vs. 50.0%; P = 0.689).

Regarding the histological characteristics of the sample, 31 patients underwent liver biopsy, 35.5% showed cirrhosis and 77.4% showed significant fibrosis. In a study conducted by Ferreira, in Brazil, in a pediatric population, 76.9% patients had cirrhosis at diagnosis [27]. Differently, a cohort study in Denmark, which evaluated 1318 patients with biopsy, only 8.3% had fibrosis or necrosis [28], and in Alaska 44.8% have moderate to severe fibrosis (3 or 4)[29].

Although our study have included a large majority of white skin individuals, ethnicity appears to influence the different degrees of fibrosis. The presence of cirrhosis in North America is predominantly in black patients compared to the white ones (56.7% and 37.5%) [30]. Moreover, 85% of African American descents have cirrhosis on the initial liver biopsy compared to 38% white individuals [31].

When comparing individuals with significant fibrosis to those with insignificant fibrosis it was found that patients with significant fibrosis showed lower ALT, AST, bilirubin mean and higher PA mean.

The aminotransferases are sensitive indicators of hepatocellular damage [32]. High levels of aminotransferases, more than 10 times greater the normal limit, at the disease presentation suggest acute presentation [33]. The finding that patients with significant fibrosis present lower ALT, AST, bilirubin levels and higher PA levels probably reflect the records of acute presentation with liver dysfunction, typical of AIH. Al-Chalabi et al., similarly to the found in the present study, observed that in patients with AIH, AST levels 10 times greater than the normal limit at the presentation were associated with a lower risk of cirrhosis and better long term evolution [34].

Immunosuppressive therapy decreases the symptoms, improvemes hepatic chemistry tests and reduces histological the activity in 80% of patients [35]. The normalization of both aminotransferases (ALT / AST) is the overall goal of treatment; otherwise the progression of the disease cannot be avoided [36]. Notably, normalization of serum aminotransferase levels do not necessarily indicate histological normalization [37]. However, a small subset of patients have second biopsy and the time between the first and second biopsy was not standardized, which led us not to analyze this data in study.

Sixty-seven percent of patients improved with treatment and have normal or near normal liver tests [35]. Less than half of our sample (39.4%) had biochemical response at six

months of treatment. A systematic review published by Lamers et al. demonstrated remission in 10 to 53% of patients receiving different treatment schedules [38]. Usually the response to treatment, in order that consider the suspension of the treatment, is evaluated in one or two years [39], but not all patients in our study had a complete examination from this period. Mehndiratta et al. evaluated the response to treatment similarly to our study, at 6 months of treatment, in 52 patients and 10% had suboptimal response [40]. It is not well established in the literature how long the biochemical response is awaited in order to consider new biopsy and exchange of immunosuppressive therapy [38]. Individuals with cirrhosis typically do not tolerate immunosuppressive suspension because the risk of hepatic decompensation is high.

When assessing the response to treatment, we observed that a greater proportion of subjects with insignificant fibrosis on liver biopsy and liver failure (PA less than 40%) on admission, and those with lower mean PA have higher biochemical response rates (ALT and AST less than 55 U/L) in the sixth month of treatment. Likewise, the smaller the value of PA on admission, the lower the ALT and AST values in the sixth month of treatment. Thus, we found that the more severe the initial presentation and less significant initial fibrosis, the greater the chance of the individual to respond to immunosuppressive treatment.

Roberts et al. evaluated 128 AIH patients, 29% with cirrhosis at admission and observed that the presence of cirrhosis did not influence the frequency of remission, recurrence after discontinuing the drug, or treatment failure [41]. Nikias et al. evaluated 26 patients and compared them according to the initial presentation: acute or chronic. Remission, recurrence, treatment failure, progression to cirrhosis and death from liver failure occurred with similar frequencies in patients with acute and chronic presentations [42]. Moreover, Kessler et al. observed findings similar to those described in our study. They evaluated 30 patients: 10 with acute presentation and 20 with chronic disease. Patients with acute presentation differ significantly regarding encephalopathy, levels of albumin and bilirubin levels. In histological

evaluation, those with acute presentation had significantly less fibrosis [43]. Oliveira et al. evaluated 131 patients and separated individuals with acute presentation (n = 80) from those with acute presentation without fibrosis to histology (n = 6) and observed that the genuinely acute cases had liver function tests more preserved and with a better biochemical response to treatment [44], which partly differs from this study, possibly for not have performed such distinction. This information on genuinely acute cases require confirmation because of the small number of patients studied and the definition used.

A national population-based study in Denmark showed that male gender and cirrhosis were adverse prognostic factors [28]. Abe et al. evaluated 15 patients with AIH and compared them with individuals with hepatitis of other etiologies. They observed, unlike our study, that patients with high levels of bilirubin did not respond to corticosteroid therapy [45]. However, in our study only two patients make use of prednisone alone. The others were subjected to therapy combined with azathioprine.

Some limitations to this study should be mentioned: although the total number of patients included in the cohort was representative of the general population, we examined possible associations between certain factors, significant fibrosis, and biochemical response, with considerable reduction in the number of subjects with certain combinations of variables and consequently, reducing the power of statistical tests. Although it is not a strict rule, it is recommended to keep a minimum of ten events per variable in the logistic regression analysis. This recommendation is based on studies showing increased bias and variability, unreliable coverage of confidence intervals, and problems with the convergence model when the number of events per variable descended below 10 [46-48]. For this reason, these results need to be confirmed in a larger set of patients. In addition, the study design was cross-sectional with retrospective collection of data and did not include a longitudinal follow-up; yet the population described in this study and the results presented are comparable to previously

published data. And lastly, the fact that we have assessed the predictors of histological response for not having a second biopsy of all patients as mentioned previously. Still, identify to the admission factors predictive of biochemical response to treatment are of great value to conduct the patient with AIH. New studies are needed to evaluate the identification of liver failure on admission may, precociously, determine therapeutic choices and the evolution of the patient, in the long term.

This study therefore concludes that individuals with liver dysfunction and elevated aminotransferases do not have significant liver fibrosis on histology. Moreover, AIH patients who initially present liver failure and and insignificant fibrosis have a greater chance of presenting biochemical response to treatment.

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Figure 1. Distribution of the positivity of autoantibodies in 36 autoimmune hepatitis patients.

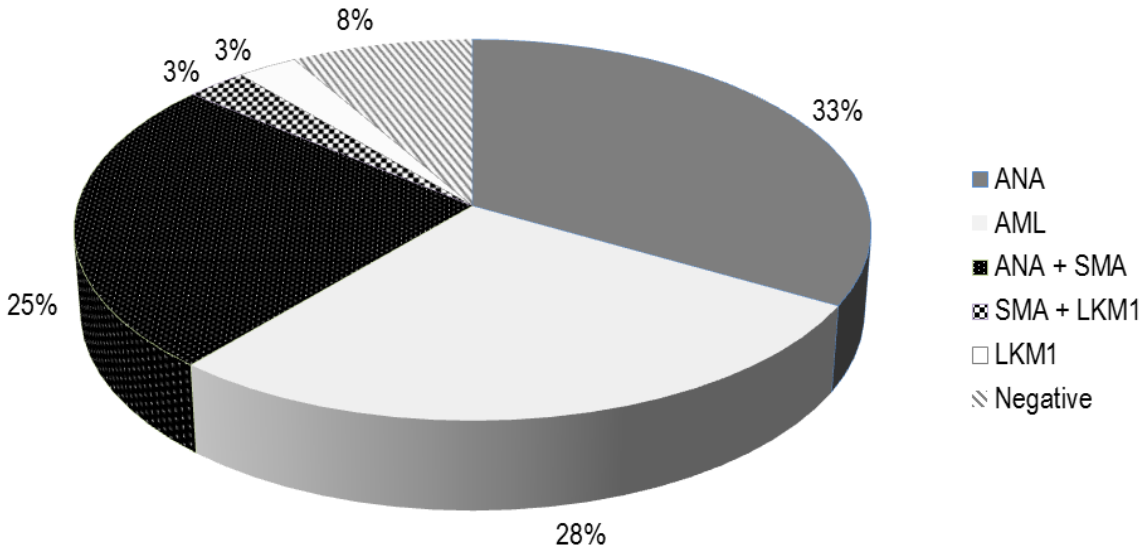

Figure 2. Evolution of hepatic biochemical tests with autoimmune hepatitis treatment.

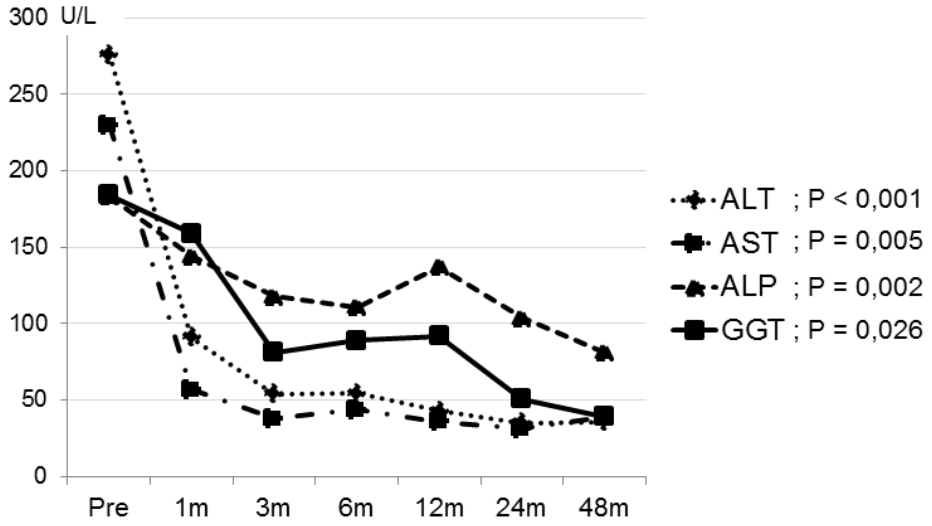

Figure 3. Correlation between the admission prothrombin activity and the aminotransferases after 6 months of treatment

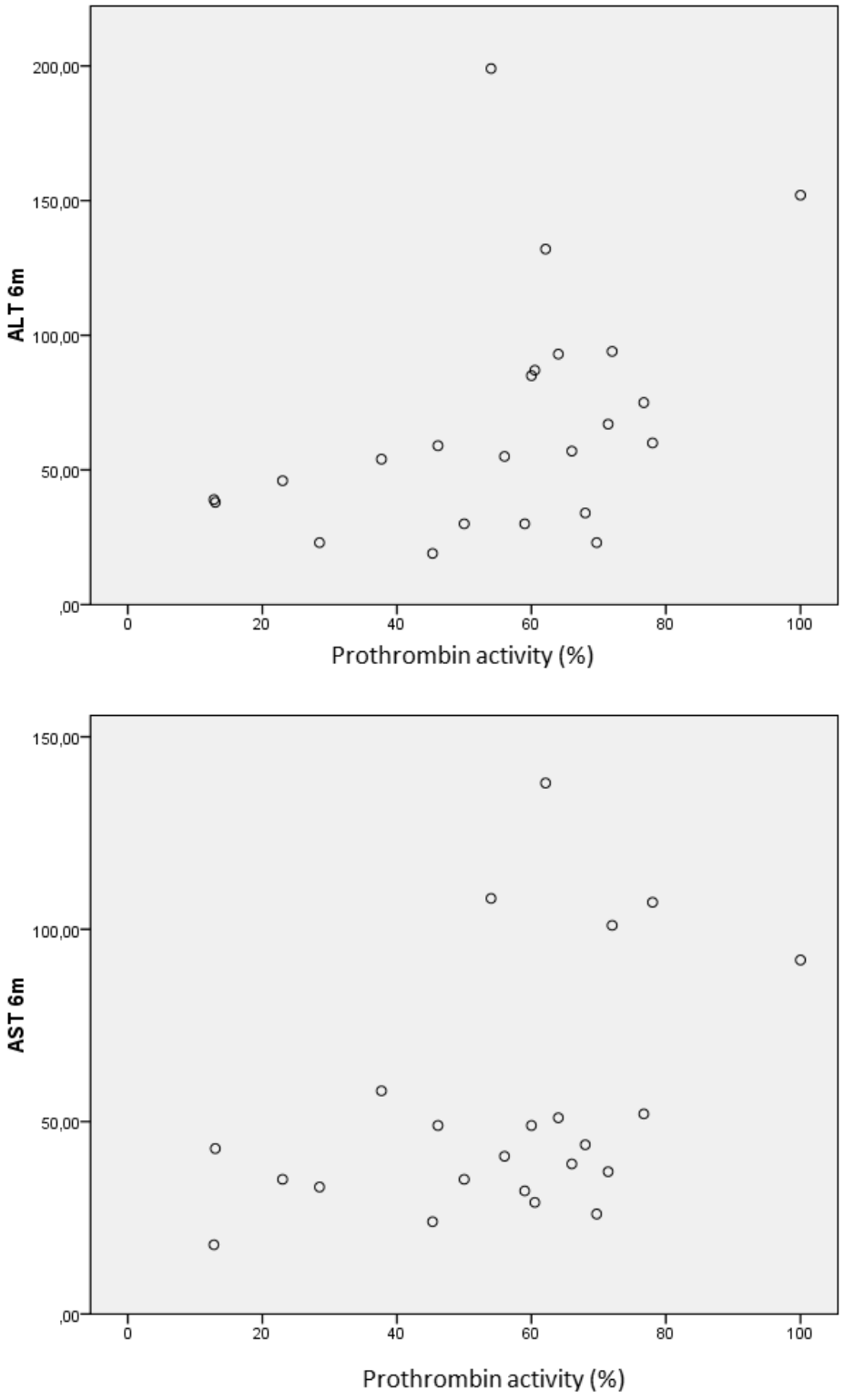

Table 1. Clinical and laboratory pre-treatment characteristics of 36 patients with autoimmune hepatitis.

Characteristics	n	%	Average ± standard deviation	Median
Male	22.2	8		
White skin	97.1	22		
Age			44.7 ± 14.3	46.5
AST basal (U/L)			476.4 ± 505.6	230.0
ALT basal (U/L)			383.8 ± 368.6	276.0
ALP basal (U/L)			204.4 ± 118.8	183.5
GGT basal (U/L)			254.7 ± 281.0	184.0
Direct bilirubin (mg/dL)			4.3 ± 5.1	1.7
PA (%)			57.6 ± 21.9	60.0
Albumin (g/dL)			3.2 ± 0.8	3.5
Gamma globulin (g/dL)			3.1 ± 4.6	1.9

AST: aspartate aminotransferase; ALT: alanine aminotransferase; ALP: alkaline phosphatase; GGT: gamma-glutamyl transferase; PA: prothrombin time activity.

Table 2. Clinical and laboratory characteristics associated with the presence of significant fibrosis on liver biopsy in 31 autoimmune hepatitis patients

Characteristics	**Total**	**Significant fibrosis N= 23**	**Insignificant fibrosis N = 8**	***P***
Age (years)*	44.7 ±14.3 (46.5)	48.0 ± 12.5 (47.0)	42.0 ± 15.6 (44.5)	0.311[t]
Male, n (%)	7 (22.6)	6 (26.1)	1 (12.5)	0.642 [f]
White skin, n (%)	30 (96.8)	22 (95.7)	8 (100.0)	1.000 [f]
AntiHBc (+), n (%)	5 (16.1)	4 (17.4)	1 (12.5)	1.000 [f]
ALP, n (%)	18 (58.1)	14 (60.9)	4 (50.0)	0.689 [f]
SMA, n (%)	20 (64,5)	15 (65.2)	5 (62.5)	1.000 [f]
LKM1, n (%)	1 (3.2)	1 (4.3)	0 (0.0)	1.000 [f]
ALP/ALT ≥ 1,5	7 (31.8)	6 (37.5)	1 (16.7)	0.616 [f]
AST (U/L)*	476.4 ±505.6 (230.0)	315.2 ± 341.9 (130.0)	893.8 ± 640.2 (1150.0)	**0.039**[t]
ALT (U/L)*	383.8 ± 368.6(276.0)	295.9 ± 240.5 (206.0)	599.4 ± 351.6 (515.0)	**0.038**[t]
ALP (U/L)*	204.4 ± 118.8 (183.5)	211.1 ± 144.4 (183.0)	217.2 ± 58.0 (206.0)	0.922[t]
GGT (U/L)*	254.7 ± 281.0 (184.0)	285.9 ± 321.1 (213.0)	224.0 ± 226.5 (115.0)	0.458[m]
Direct bilirubin (mg/dL)[*]	4.3 ± 5.1 (1.7)	2.9 ±3.0 (1.2)	8.3 ± 7.5 (6.4)	**0.013**[t]
Albumin (g/dL)*	3.2 ± 0.8 (3.5)	3.3 ± 1.0 (3.6)	2.9 ± 0.5 (2.8)	0.378[t]
PA (%)*	57.6 ± 21.9 (60.3)	65.2 ± 21.0 (66.2)	40.5 ± 19.3 (45.7)	**0.020** [t]
Gammaglobulins (g/dL)*	3.1 ± 4.6 (1.9)	3.8 ±6.9 (1.9)	2.2 ±1.0 (1.9)	0.913[m]

Anti HBc: antibody against the core antigen of hepatitis B virus; ANA: antinuclear antibody; SMA: anti-smooth muscle antibody; Anti-LKM1: antibody against the microsomal fraction of liver and kidney type 1; AST: aspartate aminotransferase; ALT: alanine aminotransferase; ALP: alkaline phosphatase; GGT: gamma-glutamyl transferase; PA: prothrombin time activity.

Table 3. Clinical and laboratory characteristics associated with biochemical response† of 33 patients with autoimmune hepatitis in the sixth month of treatment

Characteristics	Biochemical response N= 13	No response N = 20	*P*
Age (years)*	43.0 ± 13.3 (43.0)	46.4 ± 15.1 (48.0)	0.513 [t]
Male, n (%)	6 (30.0)	6 (30.0)	0.431 [f]
White skin, n (%)	13 (100.0)	19 (95.0)	1.000 [f]
Liver failure, n (%)	4 (44.4%)	1 (5.9)	**0.034** [f]
ANA, n (%)	7 (53.8)	13 (65.0)	0.522 [q]
ANA ≥1:320, n (%)	5 (71.4)	5 (38.5)	0.350 [f]
SMA, n (%)	8 (61.5)	12 (60.0)	0.930 [q]
SMA ≥ 1:320, n (%)	2 (25.0)	4 (33.3)	1.000 [f]
LKM1, n (%)	0 (0.0)	1 (5.3)	1.000 [f]
Gammaglobulins (g/dL)*	2.3 ± 0.9 (2.0)	3.8 ± 6.0 (1.9)	0.753 [m]
Direct bilirubin (mg/dL)*	6.5 ± 6.7 (5.6)	2.9 ± 3.6 (1.1)	0.082 [t]
Albumin (g/dL)*	2.3 ±1.3 (2.6)	3.4 ± 0.7 (3.5)	0.192 [t]
PA (%)*	41.0 ± 22.5 (45.3)	66.4 ±16.2 (64.0)	**0.003** [t]
Significant fibrosis, n (%)	7 (53.8%)	15 (88.2)	**0.049** [f]
Significant periportal activity, n (%)	8 (80.0)	11 (100.0)	0.214 [f]
Significant parenchymal activity (n%)	5 (45.5)	8 (66.7)	0.414 [f]

†Biochemical response: ALT and AST < 55 U/L in the sixth month of treatment; AST: aspartate aminotransferase; ALT: alanine aminotransferase; PA: prothrombin activity time; Significant fibrosis: portal fibrous expansion with portal-portal and central-portal septa and/or complete nodes (definite cirrhosis).; Significant periportal inflammatory activity: in piecemeal necrosis that can be discreet or present in large areas of many portal spaces; Significant parenchymal inflammatory activity: focal necrosis of hepatocytes surrounded by lymphohistiocytic aggregates in numerous sites with or without confluent necrosis, which can be extensive or multiple. *Average ± standard deviation (Median); Student's *t* [t]test; [m]Mann-Whitney, chi square [q]Test; Fisher exact [f]Test

Table 4. Clinical and laboratory characteristics associated with biochemical response † of 33 patients with autoimmune hepatitis in the sixth month of treatment

Characteristics	Biochemical response N= 13	No response N = 20	*P*
Age (years)*	43.0 ± 13.3 (43.0)	46.4 ± 15.1 (48.0)	0.513 [t]
Male, n (%)	6 (30.0)	6 (30.0)	0.431 [f]
White skin, n (%)	13 (100.0)	19 (95.0)	1.000 [f]
Liver failure, n (%)	4 (44.4%)	1 (5.9)	**0.034** [f]
ANA, n (%)	7 (53.8)	13 (65.0)	0.522 [q]
ANA ≥1:320, n (%)	5 (71.4)	5 (38.5)	0.350 [f]
SMA, n (%)	8 (61.5)	12 (60.0)	0.930 [q]
SMA ≥ 1:320, n (%)	2 (25.0)	4 (33.3)	1.000 [f]
LKM1, n (%)	0 (0.0)	1 (5.3)	1.000 [f]
Gammaglobulins (g/dL)*	2.3 ± 0.9 (2.0)	3.8 ± 6.0 (1.9)	0.753 [m]
Direct bilirubin (mg/dL)*	6.5 ± 6.7 (5.6)	2.9 ± 3.6 (1.1)	0.082 [t]
Albumin (g/dL)*	2.3 ±1.3 (2.6)	3.4 ± 0.7 (3.5)	0.192 [t]
PA (%)*	41.0 ± 22.5 (45.3)	66.4 ±16.2 (64.0)	**0.003** [t]
Significant fibrosis, n (%)	7 (53.8%)	15 (88.2)	**0.049** [f]
Significant periportal activity, n (%)	8 (80.0)	11 (100.0)	0.214 [f]
Significant parenchymal activity, n (%)	5 (45.5)	8 (66.7)	0.414 [f]

† Biochemical response = ALT and AST <55 U/ L in the sixth month of treatment; AST = aspartate aminotransferase; ALT = alanine aminotransferase; PA = prothrombin activity time; significant fibrosis = portal fibrous expansion with septa door-door and/or door-center and/or complete nodes; periportal inflammatory activity = significant necrosis on piecemeal that can be discreet or present in large areas of many portal spaces; significant parenchymal inflammatory activity = focal necrosis of hepatocytes surrounded by lymphohistiocytic aggregates in numerous sites with or without confluent necrosis, which can be extensive or multiple. *Average ± standard deviation (Median); Student's *t* [t]test; [m]Mann-Whitney, chi square [q]Test; Fisher exact [f]Test