ORIGINAL ARTICLE

Characterization of overlap syndrome between primary biliary cirrhosis and autoimmune hepatitis according to antimitochondrial antibodies status

Laurent ALRIC (1), Sophie THEBAULT (1), Janik SELVES (2), Jean-Marie PERON (3), Sanae MEJDOUBI (1), Françoise FORTENFANT (4), Jean-Pierre VINEL (3)

(1) Service de Médecine Interne, Fédération Digestive, Pavillon Dieulafoy ; (2) Service d'Anatomopathologie ; (3) Service d'Hépato-Gastroentérologie, Fédération digestive ; (4) Service d'Immunologie, CHU Purpan, Toulouse.

SUMMARY

Aims — Codification of variant forms between Primary Biliary Cirrhosis (PBC) and Autoimmune Hepatitis (AIH) has not been definitively standardized. The aim of this study was to compare among 102 consecutive patients, 2 subsets of overlap syndrome (OS, N=21) with and without antimitochondrial antibody (AMA) to two groups of patients with typical PBC (N=43) or AIH (N=38).

Methods — OS was defined by the presence in the same patient of at least 2 of 3 accepted criteria of PBC and AIH. Twelve patients with OS were AMA negative and 9 were AMA positive.

Results — A lower level of alanine transaminase (139±48 *vs* 269±154 IU/L, P<0.05) and a trend towards a higher level of alkaline phosphatase or gamma-glutamyl transpeptidase was observed in OS without AMA than in OS with AMA (693±200 *vs* 544±124 IU/L; 370±66 *vs* 241±77 IU/L, respectively). All AMA-negative patients with OS had antinuclear and/or antismooth muscle antibodies. OS without AMA differed from those with AMA in that they had more severe bile duct damage including destructive cholangitis (P<0.05), ductopenia (P<0.05), ductular hyperplasia (P<0.05) and a higher METAVIR fibrosis score (2.5±0.3 *vs* 1.3±0.3, P<0.05). The response to therapy was not different between PBC, AIH and OS.

Conclusions — According to the presence of AMA, 2 homogeneous subgroups of patients with overlap syndrome between PBC and AIH may be identified. AMA status affects clinical presentation and liver disease severity of OS.

RÉSUMÉ

Caractéristiques des formes frontières entre cirrhose biliaire primitive et hépatite auto-immune en fonction du statut pour les anticorps anti-mitochondries

Laurent ALRIC, Sophie THEBAULT, Janik SELVES, Jean-Marie PERON, Sanae MEJDOUBI, Françoise FORTENFANT, Jean-Pierre VINEL

(Gastroenterol Clin Biol 2007;31:11-16)

Objectifs — La codification des formes frontières entre cirrhose biliaire primitive (CBP) et hépatite auto-immune (HAI) n'est pas encore standardisée. Le but de cette étude était de comparer parmi 102 malades consécutifs, 2 sous-groupes de forme frontière (N = 21) avec et sans anticorps antimitochondries (AAM) avec 2 groupes de malades atteints d'une forme typique de CBP (N = 43) ou d'HAI (N = 38).

Méthode — Les formes frontières étaient définies par la présence chez le même malade d'au moins 2 des 3 critères classiques de CBP et d'HAI. Parmi les 21 malades avec une forme frontière, 12 n'avaient pas d'AAM et 9 étaient porteurs d'AAM.

Résultats — Un taux plus faible d'ALAT (139 ± 48 vs 269 ± 154 UI/L, P < 0,05) et une tendance à une activité plus élevée de phosphatases alcalines ou de gammaGT étaient observés pour les formes frontières sans AAM par rapport à celles avec AAM (693 ± 200 vs 544 ± 124 UI/L, 370 ± 66 vs 241 ± 77 UI/L, respectivement). Toutes les formes frontières sans AAM avaient des anticorps antinucléaires et/ ou des anticorps antimuscles lisses. Les formes frontières sans AAM étaient différentes de celles avec AAM et présentaient des lésions biliaires plus sévères incluant une cholangite destructive (P < 0,05), une hyperplasie ductulaire (P < 0,05) et un score de fibrose plus élevé (2,5 ± 0,3 vs 1,3 ± 0,3, P < 0,05). La réponse au traitement n'était pas différente entre les malades avec une forme frontière et ceux ayant une CBP ou une HAI.

Conclusions — En fonction de la présence d'AAM, 2 sous-groupes homogènes de malades avec une forme frontière entre CBP et HAI peuvent être identifiés. Le statut en AAM modifie la présentation clinique et la sévérité de la maladie hépatique des malades ayant une forme frontière.

Introduction

The diagnostic criteria of primary biliary cirrhosis (PBC) and autoimmune hepatitis (AIH) have been codified on the basis of clinical laboratory and histological findings [1, 2]. However, some patients exhibit overlapping features between PBC and AIH [4-11]. The codification of these variant forms has not been definitively standardized.

Reprints: L. ALRIC, Service de Médecine Interne, Fédération Digestive, Pavillon Dieulafoy, CHU Purpan, 31059 Toulouse cedex.

E-mail : ALRIC.L@chu-toulouse.fr

Serum antimitochondrial antibody (AMA) positivity is the most useful diagnostic test for PBC. The specificity and the sentivity of this marker are not absolute because it has been found in other auto-immune liver disease and 5 to 10% of patients with typical clinical and histological PBC are negative for AMA [2, 6]. To date the classification of AMA negative patients is still debated. Some authors [6, 10, 11], have used the term of autoimmune cholangitis for a variant form of PBC with clinical and pathological features of PBC but with negative AMA and positive antinuclear antibodies (ANA). Conversely, Micheletti et al. [4] as well as Czaja et al. [12] have described under the same term of autoimmune cholangitis, patients with negative serum AMA and auto-immune features overlapping between

PBC and AIH. On the other hand, Chazouilleres et al. [5] proposed to classify overlap syndrome (OS) as lying between PBC and AIH in patients with at least 2 of 3 accepted criteria for each disease [5]. Taking into account all these studies, overlap syndrome might therefore be a large entity including distinct subsets of patients not yet defined [13].

The aim of this study was to compare using the Chazouilleres et al. criteria [5], 2 subgroups of OS with and without AMA to two groups of patients with typical PBC or AIH. The response of the variant syndrome to immunosuppressive and ursodeoxycholic acid (UDCA) therapies was also evaluated.

Patients and methods

Study population

Between 1997 and 2003, a cohort of 102 consecutive patients referred for type 1 AIH, PBC or OS with a follow up for at least one year were studied. Patients with other causes of liver disease such as primary sclerosing cholangitis, viral hepatitis, exposure to hepatotoxic drugs or alcohol abuse were excluded. For each patient, standardized clinical and laboratory findings as well as liver histology were available at the time of initial evaluation. Ultrasonography of the liver and the biliary tract was normal in all patients.

The diagnostic criteria proprosed by Chazouilleres et al. [5] were:

— Group 1: 43 patients had a diagnosis of PBC on the following criteria: 1) serum alkaline phosphatase (AP) levels at least two times the upper limit of normal values; 2) a positive test for AMA; and 3) a liver biopsy specimen showing florid bile duct lesions.

— Group 2: 38 patients had a diagnosis of type 1 AIH on the following criteria: 1) serum alanine transaminase (ALT) levels at least five times the upper limit of normal values; 2) serum immunoglobulin G (IgG) levels at least two times the upper limit of normal values or a positive test for anti-smooth muscle antibodies (ASMA); and 3) a liver biopsy showing moderate or severe periportal or periseptal lymphocytic piecemal necrosis.

— Group 3: 21 patients were designed to have a PBC-AIH overlap syndrome. This diagnosis required the presence of at least two of three accepted criteria of PBC and AIH. Among them, 12 patients were AMA negative and 9 patients were AMA positive. Magnetic resonance cholangiography was performed in 20 patients. All of them had a normal cholangiography.

Immunoserological assessment

ASMA, ANA, AMA, ANCA were sought on murine section, Hep-2 cells and neutrophil spreading by indirect immunofluorescence. ASMA had specificity against smooth muscle actin confirmed with dot (DTek DIasorin, Stillwater USA). AMA negative samples were tested by westernblotting against liver mitochondrial antigens as previously described [14]. No patient found to be negative at immunofluorescence was positive with Werstern blotting analysis.

ABBREVIATIONS:

PBC : primary biliary cirrhosis
AIH : autoimmune hepatitis
AMA : antimitochondrial antibodies
OS : overlap syndrome
AP : alkaline phosphatase
ANA : antinuclear antibodies
UDCA : ursodeoxycholic acid
ASMA : antismooth muscle antibodies

HLA typing

Only 68 of the 102 patients were evaluated for class I (A1 and B8 loci) and class II (DR3 and DR4 loci) HLA using a standard microlymphocytotoxicity technique as previously described [15].

Histological assessment

All patients underwent an initial liver biopsy. A single pathologist with a special interest in liver disease read all the samples. Specimens were staged according to METAVIR score [16] and other international criteria used for AIH and PBC [1, 17].

Response to treatment

Because treatment strategy was standadized in our unit since 1999 only, the response of 33, 28 and 14 patients from group 1, group 2 and group 3 were analysed respectively:

— Group 1: PBC (N=33), patients received UDCA 15 mg/Kg/day.

— Group 2: AIH (N=28), all the patients were treated with an initial dose of 0.75 mg/Kg of prednisone daily during one month. When serum ALT was lower than twice the upper normal value, prednisone was progressively tapered by 2.5 mg per day every two weeks. Corticosteroid therapy was combined with azathioprine at a dose of 1mg/Kg per day.

— Group 3: OS (N=14), all the 14 patients were treated by UDCA and prednisone as previously described above. Azathioprine at a dose of 1 mg/Kg/day was added to UDCA and corticosteroid therapy.

Patients with normal or a decrease ≥50% of ALT, GGT and AP level after one year of treatment were considered to have complete or partial response, respectively.

Statistical analysis

Results were expressed as mean ± SD. Chi 2 method with Yates' correction and the Fisher's exact test were used to compare dichotomous variables. Differences in the means of continuous variables were assessed by the Student's paired t-test. The Mann-Whitney's test was used to compare nonparametric variables in independent samples. A P corrected (Pc) with Bonferroni's correction was used for HLA analysis [18].

Results

Clinical and laboratory findings at diagnosis

Of 102 patients, 21 (20.5%) could be classified as having an OS. No difference was observed between the 3 groups for the age at diagnosis (table I). In patients with OS a significantly ($P<0.05$) higher prevalence of male sex (38.1%) was observed as compared to PBC (6.9%). The sex ratio was not different between OS and AIH groups. No difference was observed between the 3 groups for clinical symptoms or associated immune diseases. The most frequent associated autoimmune diseases were sicca syndrome (25%, 14%, and 23%), Raynaud phenomenon (20%, 7%, and 9%), systemic sclerosis (15%, 0%, 9%) and thyroiditis (18%, 17%, and 19%) observed in PBC, AIH and OS, respectively. A significantly greater serum AST and ALT levels (table I) was observed in OS than in patients with PBC ($P<0.05$). Patients with OS were undistinguishable from those with PBC with regard to serum AP, gamma glutamyl-transpeptidase (GGT), bilirubin or gammaglobulins values. Conversely, patients with OS (table I) differed from patients with type 1 AIH by lower serum ALT and higher serum levels of AP, GGT and IgM