Introduction

The infection of the liver caused by hepatotropic virus such as hepatitis C virus (HCV) poses an important global epidemiological problem. A number of infected people is estimated to at 170 mln worldwide [4, 20]. 80% of cases result in chronic infection, which can be asymptomatic for many years, but it can result in liver cirrhosis and hepatocellular carcinoma [11]. Diagnosis of HCV infection is based on serological tests and the detection of virus genetic material. The determination of the severity and the stage of the disease is based on liver biopsy which has been considered as a golden standard for many years. The major disadvantage of this method is its invasiveness, which is a main reason for searching for other diagnostic tools of liver diseases. We selected serum Alanine Aminotransferase activity (ALT) and C-reactive protein (CRP) as biochemical markers of impaired liver function. In order to evaluate the correlation between serum ALT activity, serum CRP and degree of inflammatory activity in the liver, we examined 37 liver biopsy specimens from young adult patients with chronic hepatitis C. Serum CRP levels and ALT activity were determined at the time of liver biopsy in all patients. The activity of ALT correlated with the grade of liver disease (Pearson’s correlation coefficient = 0.378) and macrovesicular steatosis (Pearson’s correlation coefficient = 0.498). We didn’t found any correlation between serum CRP levels and activity and stage of liver disease. This results suggest that ALT activity, in opposition to serum CRP concentration, is a suitable biochemical marker of progression of liver disease in chronic hepatitis C.

Key words: ALT, CRP, hepatitis C virus, inflammation, liver fibrosis

Comparative study of the grade and stage of chronic hepatitis C with CRP and ALT activity in young adult patients

Paulina Godzik1, Kazimierz Madaliński1, Bożena Walewska-Zielecka1, Joanna Cielecka-Kuszyk1, Joanna Jabłońska2, Wadim Kapulkin1

1 Department of Virology
National Institute of Public Health, National Institute of Hygiene
Warsaw, Poland

2 Department of Hepatology and Acquired Immunodeficiency Syndroms
Institute of Infectious and Parasitic Diseases, Warsaw Medical University
Warsaw, Poland

Abstract

The aim of this study was to analyze histological changes in the liver and to determine the role of Alanine Aminotransferase activity (ALT) and C-reactive protein (CRP) as biochemical markers of impaired liver function. In order to evaluate the correlation between serum ALT activity, serum CRP and degree of inflammatory activity in the liver, we examined 37 liver biopsy specimens from young adult patients with chronic hepatitis C. Serum CRP levels and ALT activity were determined at the time of liver biopsy in all patients. The activity of ALT correlated with the grade of liver disease (Pearson’s correlation coefficient = 0.378) and macrovesicular steatosis (Pearson’s correlation coefficient = 0.498). We didn’t found any correlation between serum CRP levels and activity and stage of liver disease. This results suggest that ALT activity, in opposition to serum CRP concentration, is a suitable biochemical marker of progression of liver disease in chronic hepatitis C.

Key words: ALT, CRP, hepatitis C virus, inflammation, liver fibrosis

Address for correspondence
Paulina Godzik
Department of Virology
National Institute of Public Health, National Institute of Hygiene
Chocimska 24
00-791 Warsaw, Poland
Phone: +48 22 542 13 37
E-mail: pgodzik@pzh.gov.pl
Materials and methods

Patients
Total number of 37 patients (18 male, 19 female, mean age 20.5; range 15-25 yrs) with chronic hepatitis C virus infection confirmed by serological (elevated serum ALT, anti-HCV antibodies) and molecular (HCV-RNA) criteria underwent percutaneous liver biopsy. Serum CRP and ALT activity were examined at the time of liver biopsy in all patients. Informed consent for the biopsy was granted from patients themselves. All liver biopsies were performed for the first time, prior to the antiviral treatment. Patients with alcohol abuse and metabolic syndrome were excluded from the study.

Examination of needle liver biopsy specimens
Specimens taken by a blind biopsy with 1.6 mm needle were fixed in 4% buffered formalin and routinely processed into paraffin. Tissue sections 4 microns thick, containing at least 10 portal spaces were stained with H&E, impregnated with silver by the Gomori method for reticulin fibres and stained by chromotrope 2R and aniline blue for collagen fibres. Examination of inflammatory activity and the stage of fibrosis was performed according to criteria proposed by Desmet et al. in 1994 [6]. All histological features as piecemeal necrosis (interface hepatitis), lobular inflammation, focal necrosis, portal inflammation were finally scored using Histological Activity Index (HAI) with eighteen points scale to assess the grade of the disease (inflammatory activity in the lobules and portal tracts, hepatocellular damage) as: minimal: 1-3 points; mild: 4-8 points; moderate: 9-12 points; severe: 13-18 points. The stage of the disease was assessed using five points scale as: 0 – no fibrosis, normal connective tissue; 1 – portal fibrosis, fibrous portal expansion; 2 – periportal fibrosis and portal-portal septa; 3 – septal fibrosis, fibrous septa with architectural distortion; 4 – cirrhosis.

Serum sample examination
Samples of sera were tested for the presence of anti-HCV by the immunoenzymatic method (Abbott, Chicago, USA). Positive results were confirmed by RIBA HCV 3.0 (Chiron).

The ALT activity and CRP were tested at the time of the liver biopsy. The ALT activity (normal value < 40 U/l) was evaluated in 37 serum samples, collected from patients with chronic hepatitis C by the reflectance spectrophotometry method (The VITROS Chemistry System). The level of CRP (normal value < 10 mg/l) in 37 serum samples was evaluated by the immunonephelometric method (Beckman Array Analyzer).

Statistical analyses
Correlations between CRP levels and inflammatory activity, CRP and stage of liver fibrosis, CRP and liver steatosis, CRP and ALT were evaluated by calculation of the Pearson’s correlation coefficient.

Results
The age of the patients ranged from 15 to 25 years old. The ALT activity ranged from 30 to 727 U/l (mean 126 U/l, normal value < 40) (Fig. 1) and correlated with the grade of the disease (Pearson’s correlation coefficient = 0.378) and with hepatocellular macrovesicular steatosis (Pearson’s correlation coefficient = 0.498). CRP level ranged from 0.9 mg/l to 9.9 mg/l (mean 2.5, normal value < 10 mg/l). We found no correlation between serum CRP concentration and both activity and stage of liver disease.

The inflammatory activity (Fig. 2) described as minimal with characteristic minimal portal inflammation, no interface hepatitis and occasional spotty hepatocellular necrosis was observed in 5 patients (13%); mild portal inflammation with mild piecemeal necrosis, little hepatocellular damage in 20 patients (54%); moderate portal inflammation, piecemeal necrosis and lobular inflammation with noticeable hepatocellular changes in 8 patients (22%); severe inflammation with severe piecemeal and lobular necrosis in 4 patients (11%). The histological stage of the disease was 0 in 2 patients (no fibrosis, 5%), minimal in 0 patients, mild in 24 patients (portal fibrosis, periportal fibrosis with portal-portal septa, 65%), moderate in 8 patients (septal fibrosis, architectural distortion, 22%) and severe in 3 patients (cirrhosis, 8%) (Fig. 3). Hepatocellular macrovesicular steatosis was present in 23 patients (62%) and correlated with the degree of inflammation (Pearson’s correlation coefficient = 0.374).
Discussion

Previous studies showed, that the best predictors of the progression of the disease are age at the first liver biopsy, degree of ALT elevation, inflammation in liver histology and hepatic siderosis [5, 12, 13, 17]. In the present study we added the examination of CRP level and the activity of ALT, biochemical marker of impaired liver function which correlated with the degree of inflammation, liver cell necrosis and macrovesicular steatosis in comparison with histological changes in the liver.

CRP is considered as a marker of inflammatory state. CRP concentration increases rapidly during inflammation. Infection with HCV causes inflammation and liver disease, which could supposedly cause increased CRP serum level. Some authors proved that there is a correlation between CRP serum level and virus infection [2, 10, 14]. Noursadeghi et al showed that CRP values were higher in HIV-positive patients than in general population [14]. This findings were confirmed by Lau et al in study of 513 HIV-infected men. HIV-positive patients had a significant increase in CRP level, which was associated with HIV disease progression [14]. However, it was not a marker of the intensity of histological changes in the liver caused by hepatitits C infection. The extension of inflammatory infiltrates and hepatocellular changes (steatosis and spotty necrosis) correlated with age of the patients and ALT activity but did not show any association with fibrosis.

This study confirms that serum ATL activity is a good marker of the grade of chronic hepatitis C in young adult patients. We found positive correlation between serum ALT levels and activity of liver disease. CRP production in the liver cells is not affected in the natural course of hepatitis C.

It is possible that massive necrosis of hepatocytes, caused by immunological reactions against HCV antigens, may result in lack of increase in CRP level. CRP is synthesized mainly by hepatocytes and necrosis of these cells might lead to decreased CRP production. Serum CRP concentration during inflammatory activity of the liver may not increase, because of hepatocyte damage. Our previous study showed, however, that patients with simultaneous chronic hepatitis C and bacterial infection or rheumatoid arthritis, had elevated CRP serum concentration (not published). These results suggest that CRP production is not blocked by hepatocyte necrosis. In fact, extrahepatic synthesis of CRP has been reported in neurons, monocytes and lymphocytes, but the mechanism regulating its production is unknown [9].

Therefore, level of serum CRP isn’t useful diagnostic tool in liver disease in young patients with chronic hepatitis C.

These findings prompted us to determine the correlation between CRP serum levels, ALT activity and HAI score in young adult patients with chronic hepatitis C. In the whole group of patients evaluated in this study CRP serum levels were below upper normal value (<10mg/l) and there was no correlation between CRP concentration and activity and stage of liver disease. These results suggest that CRP production is not associated with progression of liver damage in chronic hepatitis C.

Acknowledgements

We thank Dr Pawel Gorynski, Dept. Medical Statistics, National Institute of Public Health, Warsaw, for performing the statistical analysis.

This work was supported by grant PBZ-KBN 119/P05/2005 from the Committee for Scientific Research, Poland.
References