Predictive Role of Acute Phase Reactants in the Response to Therapy in Patients with Chronic Hepatitis C Virus Infection

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Background/Aims: Biochemical parameters and acute-phase proteins (APPs) may provide complementary data in patients with chronic hepatitis C (CHC). We aimed to evaluate the predictive role of APPs in the response to antiviral therapy. Methods: Forty-five patients underwent antiviral therapy. Serum ferritin, C-reactive protein (CRP), transferrin, albumin, alpha-1 acid glycoprotein (A1AG), and alpha-2 macroglobulin (A2MG) levels were examined at the initial evaluation and at the 4th, 12th, and 48th weeks. HCV RNA levels were examined at the initial evaluation and at the 12th and 48th weeks. Results: Ferritin, transferrin, A1AG, and A2MG levels were significantly higher in the patient group (p<0.05). CRP, ferritin, A1AG, and A2MG levels were significantly increased from baseline to the 4th week (p<0.05). The responders and nonresponders to antiviral therapy had insignificantly but remarkably different levels of CRP, ferritin, transferrin, A1AG, A2MG, and alanine aminotransferase (ALT) both at the initial evaluation and at the 12th week. Conclusions: Variations in ferritin, A1AG, A2MG, albumin, CRP, and transferrin levels are not alternatives to virological and biochemical parameters for predicting an early response to therapy in patients with CHC. However, the investigation of ALT levels and hepatitis C virus RNA in combination with acute-phase reactants may provide supplementary data for evaluating responses to antiviral therapy. (Gut Liver 2013;7:82-88)

Key Words: Hepatitis C; Acute-phase proteins; Hepatitis C virus RNA

INTRODUCTION

Chronic hepatitis C (CHC) virus infection; one of the most frequent cause of cirrhosis and hepatocellular carcinoma (HCC), is a public health problem for all races and has become the most common cause of death associated with liver disease.¹ It is suggested that 300 million people is infected by hepatitis C virus (HCV) worldwide and the prevalence of anti-HCV positivity in Turkish population is 1.8%.² A majority of patients infected with HCV do not spontaneously clear the virus and become chronically infected. More than 80% of all HCV infected patients will develop chronic hepatitis, and in 20% of this patients will lead to liver cirrhosis.³

CHC, associated with liver cell necrosis, inflammation, regeneration, fibrosis, and cirrhosis has been recognized as an important risk factor for the development of HCC.⁴ Accumulation of iron and serum iron levels are strongly correlated with hepatic fibrosis and inflammatory activity. The presence of increased hepatic iron, which is present in 30% to 40% of patients with CHC, has been linked to more severe liver disease and poorer response to interferon (IFN) monotherapy.⁵ The clinical course of HCV infection is influenced with iron storage which leads to hepatic dysfunction and macrophage activation.⁶ Serum ferritin levels represents the severity of hepatic fibrosis independent from the liver iron accumulation.⁷ Alpha-2 macroglobulin (A2MG), an acute-phase proteins (APP), is synthesized in hepatocytes and stellate cells.⁸ A2MG inhibits the catabolism of matrix proteins and fibrotic process and both hepatic and serum levels of alpha-1 acid glycoprotein (A1AG) increases in fibrogenic environment.⁹

The aim of this study was to evaluate the predictive role of...
APPs in response to antiviral therapy.

MATERIALS AND METHODS

This study was conducted between May 2004 and May 2005 in Department of Gastroenterology, Gaziantep University Faculty of Medicine. Forty-five patients with CHC virus infection and 30 healthy individuals were enrolled. Inclusion criteria for patients group were; elevated alanine aminotransferase (ALT) levels for more than 6 months, anti-HCV positivity and HCV RNA level exceeding 200 IU/ml and moderate to severe necroinflammatory activity and fibrosis stage of 2 to 6/6. Entire patients were in genotype 1b and none of participants had a history of antiviral therapy. Control group consisted of healthy subjects with negative anti HCV. Patients and controls with concomitant disorders like congestive heart failure, chronic renal failure, psychosis, pregnancy, hematological disorders, decompensated liver cirrhosis, and combined hepatitis C+B or C+B+D virus infection were exclusion.

Diagnosis of CHC is confirmed by biochemical, serological, and histopathological examination. Modified Knodell scoring system was used for histopathological assessment. Patients with CHC received pegIFN-a-2a 180 μg/wk subcutaneously, plus oral ribavirin at a dose of 1,000 to 1,200 mg/day. HCV-RNA levels were examined at the 12th week of treatment. Treatment was continued if HCV-RNA was negative or a decrease of 2 log or more in HCV RNA level was achieved. Nonresponders were described as patients with a decrease less than 2 log in HCV RNA levels at the 12th week. Also patients with positive HCV RNA at the 24th week were accepted as nonresponders; treatment was continued up to 48th week for patients with negative HCV RNA at the 24th week.10-12

Total blood count and biochemical analysis were carried out in every outpatient clinic visit. Serum ferritin, C reactive protein (CRP), transferrin, albumin, A1AG, and A2MG levels were examined at the 4th, 12th, and 48th weeks. Serum HCV RNA levels were quantitatively analyzed at 12th, 24th, and 48th weeks.

Biochemical parameters were spectrometrically examined by Modular System Otoanalyser (Roche Diagnostic, Basel, Switzerland). Ferritin levels were assessed by Roche E-170 Modular System Electro Chemiluminecance. Serologic parameters including anti-HCV, HBsAg, anti-HBs, and autoantibodies (antinuclear antibody, smooth muscle antibodies, liver-kidney microsome type 1) were analyzed by ELISA method. PCR hybridization was used to determine HCV RNA levels. CRP, A1AG, A2MG, transferrin levels were analyzed nephelometrically by Nephelometer 100 analyser (Dade Behring, Deerfield, IL, USA).

In accordance with Declaration of Helsinki, patients were informed and informed constant was obtained.

Statistical analysis

Student’s t-test was used to calculate mean values. Grouped data were evaluated for significance using chi-squared test, and Fisher’s exact test. A p-value of smaller than 0.05 was considered to be statistically significant difference between the compared groups. Correlation of data was performed by Pearson’s test.

RESULTS

Table 1 indicates the demographic features and biochemical parameters of patients and control group. Ferritin, transferrin, A1AG, and A2MG levels were significantly higher in patients group (p<0.05) (Table 2). Difference was nonsignificant between groups in terms of CRP and albumin levels.

CRP levels were significantly elevated from initial evaluation

Table 1. Clinical and Demographic Features of Patients and Control Group

| Parameter                      | Patients (n=45) | Control group (n=30) |
|-------------------------------|----------------|---------------------|
| Gender, female/male           | 26/19          | 19/11               |
| Mean age                      | 46.1±6.1       | 45.2±3.1            |
| Body mass index, kg/m²        | 25.7±2.7       | 24.5±3.4            |
| ALT, U/L                      | 66.6±38.6      | 33.0±4.1            |
| AST, U/L                      | 60.8±34.8      | 36.2±3.4            |
| GGT, U/L                      | 55.4±43        | 45±10               |
| ALP, U/L                      | 203.9±41.4     | 170.2±25.4          |
| Total bilirubin, mg/dL        | 0.59±0.14      | 0.4±0.2             |
| WBC, 10³/µL                   | 6.9±1.4        | 5.4±1.2             |
| Hemoglobin, g/dL              | 13.2±1.3       | 13.0±1.1            |
| Platelet, 10³/µL              | 255±155        | 250±110             |
| Prothrombine time, sec        | 14.3±0.3       | -                   |
| HAI                           | 6±2.2          | -                   |
| Fibrosis                      | 2.1±1.0        | -                   |
| HCV RNA, IU/ml                | 1,007,598±1,438,867 | -            |

Data are presented as mean±SD. ALT, alanin aminotransferase; AST, aspartat aminotransferase; GGT, gamma glutamyl transferase; ALP, alkaline phosphatase; WBC, white blood cell; HAI, histological activity index; HCV RNA, hepatitis C virus RNA.

Table 2. Acute Phase Proteins Level of Patients and Control Group

| Parameter         | Patients group | Control group | p-value |
|-------------------|----------------|---------------|--------|
| CRP, mg/L         | 4.05±2.3       | 3.3±0.6       | >0.05  |
| Ferritin, ng/mL   | 95.4±93.9      | 32.9±19.3     | 0.01   |
| Transferrin, mg/ml| 2.8±0.5        | 2.2±0.6       | <0.01  |
| A1AG, mg/ml       | 0.7±0.3        | 0.6±0.2       | 0.017  |
| A2MG, mg/ml       | 2.7±0.9        | 1.2±0.5       | <0.01  |
| Albumin, g/dL     | 4.3±0.4        | 4.1±0.4       | >0.05  |

Data are presented as mean±SD. CRP, C-reactive protein; A1AG, alpha-1 acid glycoprotein; A2MG, alpha-2 macroglobulin.
to 4th and 12th week of therapy (p<0.05). However, significant elevation did not remain stable at the 48th week. Ferritin levels were significantly decreased from 4th to 12th week and to 48th week; however, it was still significantly higher than baseline point (p<0.05). A1AG levels were significantly increased between 4th, 12th, and 48th week (p<0.05). A2MG levels were significantly decreased from baseline point to 4th and to 12th week (p<0.05); however, significant decrease did not remain stable between baseline point and 48th week. Transferrin levels were nonsignificantly decreased from baseline point to 4th week; however, there was a nonsignificant elevation of transferrin levels between baseline point and 12th week. Significant decrease in transferrin levels from baseline evaluation to 48th week was observed (p<0.05). Difference of albumin levels between baseline point and 4th, 12th, and 48th week were nonsignificant. Reduction of ALT levels between baseline point and 4th, 12th, and 48th week were significant (p<0.05). Reduction of HCV RNA levels between baseline point and 12th and 48th week were also significant (p<0.05) (Table 3).

CRP, ferritin, transferrin, A1AG, A2MG, and ALT levels of responders (n=37) and nonresponders (n=8) to treatment were nonsignificantly different (p>0.05). Albumin and HCV RNA levels were decreased remarkably but nonsignificantly (p=0.054 vs p=0.055, respectively) (Table 4).

There was no significant difference between CRP, ferritin, transferrin, A1AG, A2MG, albumin, and ALT levels of responders (n=37) and nonresponders (n=8) at 12th week (p>0.05). However, HCV RNA levels were significantly different (p<0.01) (Table 5).

There was a positive correlation between HCV RNA and CRP, ferritin, A2MG, albumin levels at the 12th week; however, it was statistically nonsignificant (r=0.01, r=0.27, r=0.19, r=0.08,

### Table 3. Mean Variations of Acute Phase Proteins and ALT Levels at Initial Point, 4th, 12th, and 48th Week and HCV RNA Levels at Initial Point, 12th and 48th Week of Patients Group

| Parameter | Initial | 4th wk | 12th wk | 48th wk | p-value | 4th wk | 12th wk | 48th wk |
|-----------|---------|--------|---------|---------|---------|--------|---------|---------|
| CRP, mg/L | 4.1±2.3 | 6.8±6.5 | 6.2±5.5 | 4.8±2.6 | 0.005   | 0.014  | >0.05   |
| Ferritin, ng/mL | 95.5±93.9 | 311.9±302.9 | 242.1±218.2 | 182.3±134.1 | 0.000 | 0.001 | 0.002 |
| Transferrin, mg/mL | 2.8±0.5 | 2.6±0.7 | 2.9±0.6 | 2.5±0.5 | >0.05 | >0.05 | 0.044 |
| A1AG, mg/mL | 0.8±0.3 | 1.1±0.5 | 1.1±0.5 | 1.5±0.8 | 0.001 | 0.003 | 0.000 |
| A2MG, mg/mL | 2.8±0.9 | 2.1±0.8 | 2.2±0.9 | 2.5±0.7 | 0.085 | 0.074 | >0.05 |
| Albumin, g/dL | 4.3±0.5 | 4.2±0.5 | 4.2±0.4 | 4.3±0.6 | >0.05 | >0.05 | >0.05 |
| ALT, U/L | 66.6±38.7 | 40.9±24.1 | 31.7±16.5 | 29.7±16.5 | 0.001 | 0.005 | 0.012 |
| HCV RNA, IU/mL | 1,007,598.0±1,438,867.9 | 312,741.9±950,001.9 | 209.1±49.1 | 950,001.9 | |

Data are presented as mean±SD.

ALT, alanine aminotransferase; HCV RNA, hepatitis C virus RNA; CRP, C-reactive protein; A1AG, alpha-1 acid glycoprotein; A2MG, alpha-2 macroglobulin.

### Table 4. Mean Acute Phase Proteins, ALT, and HCV RNA Levels of Responders and Nonresponders to Treatment at Initial Point

| Parameter | Responders (n=37) | Nonresponders (n=8) | p-value |
|-----------|-------------------|---------------------|---------|
| CRP, mg/L | 4.4±0.7 | 5.0±3.7 | >0.05 |
| Ferritin, ng/mL | 92.8±96.8 | 108±84.1 | >0.05 |
| Transferrin, mg/mL | 2.8±0.5 | 2.6±0.55 | >0.05 |
| A1AG, mg/mL | 0.78±0.25 | 0.75±0.45 | >0.05 |
| A2MG, mg/mL | 2.6±0.9 | 3.2±0.5 | >0.05 |
| Albumin, g/dL | 4.3±0.4 | 4.0±0.7 | 0.054 |
| ALT, U/L | 67.4±40 | 62.8±33.3 | >0.05 |
| HCV RNA, IU/mL | 816,862.2±1,331,726.1 | 1,889,750±1,676,629 | 0.055 |

Data are presented as mean±SD.

ALT, alanine aminotransferase; HCV RNA, hepatitis C virus RNA; CRP, C-reactive protein; A1AG, alpha-1 acid glycoprotein; A2MG, alpha-2 macroglobulin.

### Table 5. Acute Phase Proteins, ALT, and HCV RNA Levels of Responders and Nonresponders to Treatment at 12th Week

| Parameter | Responders (n=37) | Nonresponders (n=8) | p-value |
|-----------|-------------------|---------------------|---------|
| CRP, mg/L | 5.9±4.8 | 5.4±2.4 | >0.05 |
| Ferritin, ng/mL | 374.3±385.4 | 213.4±156.7 | 0.58 |
| Transferrin, mg/mL | 2.8±0.5 | 2.9±0.6 | >0.05 |
| A1AG, mg/mL | 0.9±0.3 | 1.1±0.4 | >0.05 |
| A2MG, mg/mL | 2.7±1.9 | 2.1±0.6 | >0.05 |
| Albumin, g/dL | 4.2±0.6 | 4.3±0.3 | >0.05 |
| ALT, U/L | 39.1±23.8 | 53.4±23.1 | >0.05 |
| HCV RNA, IU/mL | 1,240.6±5,995 | 1,862,750±1,492,880 | <0.01 |

Data are presented as mean±SD.

ALT, alanine aminotransferase; HCV RNA, hepatitis C virus RNA; CRP, C-reactive protein; A1AG, alpha-1 acid glycoprotein; A2MG, alpha-2 macroglobulin.
respectively; p>0.05). ALT and CRP, ferritin, transferrin, A2MG levels were positively but nonsignificantly correlated (r=0.04, r=0.1, r=0.13, r=0.00, respectively; p>0.05). HCV RNA and transferrin, A1AG were negative and nonsignificantly correlated (r=0.22, r=0.125, respectively; p>0.05). ALT and A1AG levels were negatively and nonsignificantly correlated (r=0.18, p>0.05). ALT and albumin levels were negatively but significantly correlated (r=-0.37, p<0.05).

HCV RNA and CRP, transferrin, A2MG levels at the 48th week were positively but nonsignificantly correlated (r=0.11, r=0.03, r=0.33, respectively; p>0.05). ALT and CRP, transferrin, A1AG, albumin levels were negatively and nonsignificantly correlated (r=-0.16, r=-0.05, r=-0.21, r=-0.29, respectively; p>0.05). HCV RNA and ferritin, A1AG levels were negative and nonsignificantly correlated (r=-0.02, r=-0.17, respectively; p>0.05). ALT and ferritin levels were positively and significantly correlated (r=0.53, p<0.05). ALT and A2MG levels were positively and nonsignificantly correlated (r=0.05, p>0.05). HCV RNA and albumin levels were negatively but significantly correlated (r=-0.75, p<0.05).

**DISCUSSION**

Acute phase reactants (APRs) plays vital role on the regulation of inflammation and repairment of tissue damage. Synthesis and catabolism of APR is regulated by interleukin (IL-1, IL-6, IL-11), tumor necrosis factor (TNF), leukemia inhibitor factor, and oncostatin M. Tissue damage is associated with acceleration of positive APR (carnoloplasmin, C3, A1AG, A2MG, alpha-1 antitrypsin, alpha-1 antichymotrypsin, fibrinogen, haptoglobin, CRP, and serum amyloid A protein) or deceleration of negative APR (prealbumin, albumin, transferrin, and alpha-2 glycoprotein) synthesis. Tsui et al. found HCV seropositivity to be associated with lower lipids, CRP and fibrinogen levels, and higher levels of IL-6 and TNF-α.

Liver biopsy is an invasive procedure that has been associated with complications; morbidity risk of 0.3% to 0.6% and mortality risk of 0.05%. Bourliere et al. combined noninvasive procedures like Fibroscan and Hepascore (Echosens, Paris, France), and concluded that diagnostic accuracy was increased and necessity of liver biopsy was diminished.

HCV RNA and ALT are recent virologic and biochemical parameters to evaluate response to treatment in CHC. Early virologic response is determined by negative HCV RNA or reduction of 2 log by PCR analysis at the 12th week of therapy. APR may provide supplementary data as a cost-effective and feasible parameter in patients with CHC and may predict the response to therapy.

Patients with chronic viral hepatitis have increased levels of IL-1 and TNF-α; and elevated levels of cytokines stimulate the production of CRP. However, exact role of CRP on the progression of hepatitis is still unknown and remains to be determined. Shima et al. compared CRP levels of patients with chronic hepatitis B (CHB) or CHC and mentioned correlation between CRP and histologic activity in patients with CHB. Significant correlation exists between serum ALT and CRP levels and also between disease progression and CRP in patients with CHB. However, it was nonsignificant in patients with CHC. In contrast to patients with nonA-nonB hepatitis, Atono et al. determined significantly higher levels of CRP in patients with acute hepatitis A or B. Huang et al. determined significantly higher high-sensitivity CRP (hsCRP) levels in a study conducted on 95 patients with CHC and 95 healthy controls, and also achieved significant decrease in hsCRP level following to treatment with pegylated IFN and ribavirin. Biró et al. examined the effect of IFN-α-2b therapy on APRs. They observed elevated complement proteins (C-9 and C1-INH) subsequent to antiviral therapy for 3 months. Increment in C-9 and C1-INH was strongly correlated with virological response. In our study, CRP levels were similar between patients with CHC and healthy control group however decrease in CRP levels between initial evaluation and 4th and 12th week was significant (p=0.005 and p=0.014, respectively).

Alcoholic patients with CHC have diminished response to IFN therapy and significantly higher levels of CRP than nonalcoholic patients with CHC subsequent to IFN therapy. Lapinski carried out a study in 20 patients with CHC that underwent to IFN-α-2a therapy and determined no significant difference between CRP and albumin levels at the 2nd and 12th week. We observed no significant difference between responders and nonresponders in terms of CRP levels at baseline evaluation and 12th week. Elevation of CRP was significant between baseline evaluation and 4th or 12th week; however, difference between baseline evaluation and 48th week was nonsignificant. In contrast to previous reports, IFN use is considered to be the cause of elevation of CRP levels.

Ferritin levels were significantly higher in patients with CHC than healthy control group; similar to published data. Stam et al. emphasized that IFN therapy is related with increase of synthesis and secretion of ferritin. Exact mechanism of elevation of ferritin levels subsequent to IFN therapy is unknown; however, it is possibly related with liver tissue damage. Some other recent reports mentioned that combined IFN and ribavirin therapy is associated with decrease in the levels of serum iron, ferritin, transferrin, and increase amount of transferrin receptor. Our patients exhibit significantly higher ferritin levels at 4th, 12th, and 48th week. Early virologic response was evaluated at the 12th week and ferritin levels of responders were nonsignificantly higher than nonresponders. No significant correlation was observed between HCV RNA and ferritin levels. Elevated ferritin level is considered to be related with IFN therapy.

Nonresponders to IFN therapy have high levels of iron saturation. Hepatic fibrogenesis is stimulated by both liver tissue damage by viral infection induced oxygen radicals and increased liver iron storage. In a study from Turkey, 21 and
19 patients underwent to IFN+desferrioxamine and IFN alone, respectively. Ferritin and HCV RNA levels were significantly lower and histologic activity was better in the former group. Serum iron, ferritin levels and transferrin saturation were significantly higher in African-Americans with CHC and response to IFN therapy was significantly poorer in this group of patients. In contrast to relation of liver iron concentration and response to therapy, elevated level of ferritin is associated with poorer response to therapy (combined ribavirin and pegIFN therapy). Significant relation exist between serum iron level and hepatic fibrosis in patients with CHC. Serum ferritin level indicates the severity of hepatic fibrosis; independent from liver iron deposition. Serum iron level and liver iron storage are extremely high in IFN-nonresponders. Excessive iron load leads to poorer response to IFN however improves by iron chelation. Ferritin levels of our patients that response to IFN were nonsignificantly lower than nonresponders. The presence of increased hepatic iron, which is present in 30% to 40% of patients with CHC, has been linked to more severe liver disease and poorer response to IFN monotherapy. Iron depletion by phlebotomy has been shown to be associated with decreased incidence of decompensation and HCC.

Serum transferrin levels exhibit hepatic iron load and related with poorer response to IFN therapy. Kalabay et al.23 performed a study in 40 patients with CHC that underwent to IFN-α-2b therapy. Patients responsive to treatment had significantly lower levels of transferrin than unresponsive group; however, CRP levels were similar between two groups. Patients with CHC usually higher levels of transferrin saturation. Iron load influences the natural course of HCV infection by leading to hepatic dysfunction and macrophage activation. Combined IFN and ribavirin therapy decreases serum iron, ferritin, and transferrin levels and increases the density of transferrin receptors. Transferrin levels of our patients significantly decreased from baseline point to 48th week. No significant difference was observed between transferrin levels of responders and nonresponders at the 12th week of therapy. This may be related to the fact that responders had higher transferrin levels at the initial evaluation.

A2MG, haptoglobin, gammaglutamyl transferase (GGT) and total bilirubin are serum predictors of hepatic fibrosis. A2MG is an APR; produced by hepatocytes and stellate cells, inhibits catabolism of matrix proteins and hepatic fibrosis. Fibrogenesis and cytokine induced hepatocyte growth factor stimulates the synthesis of A2MG while inhibiting the synthesis of haptoglobin. Patel et al.27 determined high levels of A2MG in patients with moderate to severe fibrosis. Patients with chronic persistant hepatitis and cirrhosis have increased density of lymphocytes and fibroblasts in liver tissue and higher levels of serum IgG and A2MG. Poynard et al. stated that serum markers like A2MG, haptoglobin, GGT, and total bilirubin may have a predictive role on the progression of fibrosis and virologic response in patients with CHC without possible harmful effects of invasive methods such as liver biopsy. Ho et al. carried out a study to assess the predictive role of noninvasive serologic biomarkers such as A2MG, vitamin D binding protein (VDBP) and apolipoprotein AI (ApoAI). They revealed that the serum concentration of A2MG significantly increased from mild to advanced fibrosis however the protein levels of VDBP and ApoAI were significantly higher in normal or mild fibrosis, when compared to those in advanced fibrosis (both p<0.01). A2MG levels were significantly higher in our patients with CHC than healthy control group. Nonresponders had nonsignificantly higher levels of A2MG than responders. A2MG levels of our patients significantly decreased at the 4th and 12th week of treatment; however, it did not remain stable at the 48th week.

Serum albumin level and thrombocyte count indicate the severity of liver damage. Thrombocyte count lower than 140,000/mm³, AST/ALT >1, globulin/albumin >1 are considered to be the predictors of progression to cirrhosis. Volchkova et al. reported that patients with acute viral hepatitis have low levels of albumin, prealbumin, transferrin and high levels of haptoglobin. However, patients with CHC had low levels of haptoglobin as well as the other markers. No significant difference was observed in terms of albumin levels at the 4th, 12th, and 48th week. Nonresponders had nonsignificantly but remarkably lower albumin levels than responders at the baseline evaluation (p=0.054). Negative correlation exist between HCV RNA and albumin levels at the 48th week and between ALT and albumin levels at the 12th week. Low albumin levels at the baseline evaluation is associated with poorer response to therapy.

A1AG, an internal membrane protein related with collagen overproduction, is synthesized by neutrophils, lymphocytes, monocytes, and hepatocytes. Stam et al. established elevated A1AG levels at the 4th week and at the 6th month of IFN-α-2b therapy. Nonresponders had higher A1AG levels at the 12th week when compared to responders. There was a significant increase in A1AG levels during treatment period however nonresponders exhibit remarkably higher increase in A1AG levels. Patients with CHC have increased levels of A1AG; however, elevation of A1AG subsequent to antiviral therapy is considered to associated with effect of IFN.

The present study has some limitations. First, sample size in our study was relatively low. Statistical significance may be enhanced by increasing sample size. Second limitation of the study was that ALT and HCV RNA are globally accepted parameters in predicting response to treatment in patients with CHC. However combining ALT and HCV RNA with APP may be more accurate to predict response to antiviral therapy. Third, patients in our study have different duration of CHC. Heterogenity in the duration of CHC may have significant impact on response to treatment. Also racial factors may play important in response to antiviral therapy. Ioannou et al. mentioned the effect of race
and stated African-American patients with CHC exhibit poorer response to IFN therapy when compared to patients from other origin.

In conclusion, variations of ferritin, A2MG, albumin, CRP, transferrin, and A1AG levels would not be an alternative to virologic and biochemical parameters as a predictor of early response to therapy in patients with CHC. However these parameters may provide supplementary data in predicting response to treatment. Further studies with large number of individuals are required to establish these data.

**CONFLICTS OF INTEREST**

No potential conflict of interest relevant to this article was reported.

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