Rheumatoid factor and anti-cyclic citrullinated peptide (anti-CCP) antibodies with hepatitis B and hepatitis C infection: Review

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A – research concept and design; B – collection and/or assembly of data; C – data analysis and interpretation;
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Abstract

Background. Viruses are common and are involved in the etiology of idiopathic rheumatological diseases. Hepatitis B virus (HBV), a member of the family *Hepadnaviridae* and hepatitis C virus (HCV), play an important role in the undetermined etiology of arthritis. The clinical manifestations of hepatitis B and C show similarities with various diseases, such as rheumatic diseases. Anti-cyclic citrullinated peptide (anti-CCP) is a specific serological marker for rheumatoid arthritis.

Objectives. The aim of this study was to analyze anti-CCP and rheumatoid factor (RF) levels in patients with a hepatitis B and C infection.

Material and methods. Forty-four patients with hepatitis B, 43 patients with hepatitis C, 25 patients with rheumatoid arthritis, and 46 healthy control serums and their RF and anti-CCP levels were compared. RF was measured by the nephelometer, which detects IgM-RF. Anti-CCP was measured using enzyme-linked immunosorbent assay (ELISA) that is included in the second-generation anti-CCP antibody assays (anti-CCP2).

Results. The anti-CCP positivity levels were 20.5%, 32.5%, 72.4% and 10.9% for HBV, HCV and RA groups and healthy control group, respectively. When the groups were compared based on their RF positivity and anti-CCP positivity while the values for HBV and HCV group and healthy control group were the same, in RA group there is a significant difference to the rest of the groups ($p < 0.01$).

Conclusions. Anti-CCP may be positive for HBV and HCV as well, but it is a sensitive and specific immunological marker for RA diagnosis, especially in high-titres.

Key words: rheumatoid arthritis (RA), hepatitis, anti-CCP

Anti-citrullinated protein antibodies (ACPA) and RF are autoantibodies present in the majority of rheumatoid arthritis patients. Among these, anti-cyclic citrullinated peptide (anti-CCP) antibodies are widely known to be an important diagnostic and prognostic tool due to their high specificity. It has been determined that anti-CCP has a sensitivity level between 69.6% and 77.5% and a specificity between 87.8% and 96.4% in RA diagnosis.[4] anti-CCP and RF are important indicators in RA diagnosis, but they can also be positive for various infections and connective tissue diseases.[5–9] Anti-CCP and rheumatoid factor (RF) may be traced in some viral infections just as in hepatitis B (HBV) and hepatitis C (HCV) infections respectively.[14]

The diagnosis of chronic HBV and HCV infections is based on the HBsAg and anti-HCV positivity persisting for more than 6 months.[2] Certain characteristics of chronic HBV and HCV infections are similar to those of rheumatic and renal diseases. Any of the symptoms of chronic HBV and HCV infections such as arthralgia, peripheral arthritis or laboratory findings like elevated acute phase reactants including C-reactive protein (CRP), erythrocyte sedimentation rate (ESR) and positivity of RF are important markers for the diagnosis of inflammatory rheumatic diseases.[3] The aim of this study was to evaluate the rate of anti-CCP and RF positivity in hepatitis B and C infection.

Material and methods

The study subjects consisted of 44 patients with hepatitis B (male/female: 18/26), 43 patients with hepatitis C (male/female: 10/33), 25 patients with rheumatoid arthritis (male/female: 9/16) and 46 healthy individuals as the control group (male/female: 18/28). All patients with RA fulfilled the 1987 ACR criteria for RA.[10] The diagnosis of chronic HBV infection is based on the persistence of the hepatitis B surface antigen (HBsAg) for more than 6 months and patients were determined to be carriers of inactive HBV (level of HBV DNA below 2000 IU/mL and serum levels of ALT that remained normal). Chronic hepatitis C infection is defined by the presence of anti-HCV and HCV-RNA positivity for at least 6 months. Serum levels of ALT remained normal in all patients with HCV group. No patient received antiviral therapy in both HBV and HCV groups. No specific musculoskeletal symptoms and findings in all patients carrying chronic phase hepatitis B and chronic hepatitis C have been defined yet. Serum samples were obtained from venous blood, frozen and stored at -80° for future analysis. Anti-CCP and RF were studied on these samples. RF was measured by the Nephelometer, which detects IgM-RF and the normal range was between 0 and 20 IU/mL. Anti-CCP was measured using enzyme-linked immunosorbent (ELISA), which is included in the second generation anti-CCP antibody assays (anti-CCP2) and manufacturer's cut-off for positivity was ≤ 25 U/mL. Anti-CCP levels were classified in titre as low, moderate and high for values 25–50 U/mL, 50–75 U/mL and 75 U/mL respectively. All participants were informed on the study, and the procedures complied with the Declaration of Helsinki and institutional guidelines. The Ethics Committee of Gaziantep University approved the study, and informed consent was obtained from all patients.

As for the statistical analysis, the Statistical Package for Social Sciences (SPSS) was used to analyze the data. A one-way ANOVA test was used for multiple comparisons. In addition, the χ^2 and Fisher's exact tests were used for categorical variables, and the Tukey test was used to compare mean values. P values < 0.05 were considered as statistically significant.

Table 1. Demographic features of all study groups

Patients	Chronic hepatitis B n = 44	Chronic hepatitis C n = 43	Rheumatoid arthritis n = 25	Healthy control n = 46	p-value
Age (years)	34.2 ± 7.4	34.3 ± 6.0	38.4 ± 7.8	33.9 ± 7.2	0.079
Sex (male/female)	18/26	10/33	9/16	18/28	0.068

Table 2. Laboratory findings of study population

Study groups	Levels of RF	RF positivity	Levels of anti-CCP	Anti-CCP positivity
Hepatitis B	14.5 ± 11.0	5 (11.4%)	22.1 ± 23.7	9 (20.5%)
Hepatitis C	16.0 ± 11.0	7 (16.3%)	28.7 ± 24.3	14 (32.5%)
Rheumatoid arthritis	60.4 ± 82.5	15 (60%)	141.1 ± 137.4	18 (72.4%)
Healthy control	7.8 ± 4.1	1 (2.2%)	18.7 ± 4.4	5 (10.9%)
p-value	< 0.001	< 0.001	< 0.001	< 0.001

RF – rheumatoid factor; anti-CCP – anticyclic citrullinated peptid.

Results

The average ages in the study groups for chronic hepatitis B, chronic hepatitis C, RA groups and healthy control group were 34.2 ± 7.4, 34.3 ± 6.0, 38.4 ± 7.8 and 33.9 ± 7.2 respectively. There were no meaningful differences of age or gender in the groups (p = 0.079, p = 0.068 respectively). Demographic characteristics of the patients and healthy controls are presented in Table 1.

Table 3. Comparisons of anti-CCP positivity levels of groups

Anti-CCP positivity	Hepatitis B (%)	Hepatitis C (%)	Rheumatoid arthritis (%)	Healthy (%)	p-value
Low level positivity	11.4	20.9	8.0	10.9	0.373
Moderate level positivity	6.8	9.3	8.4	0	0.248
High level positivity	2.3	2.3	56	0	0.00

anti-CCP – anticyclic citrullinated peptid; anti-CCP 25–50 U/mL low; 50–75 U/mL moderate; above 75 U/mL high titre positivity.

Table 4. Published reports of the prevalence of anti-CCP antibodies in HBV-infected patients

Studies	HBV		RA	
	n	anti-CCP positivity (%, no)	n	anti-CCP positivity (%, no)
Sang-il Lee 2007 *16	176	0.6%, (1)	**	**
Lim MK 2009 *17	240	4.5%, (11)	10	9% (90)
Zhou RF 2012 *22	280	5.7%, (15)	15	13% (86.7)
Our study	44	20.5%, (9)	25	18% (72)

*reference no; **not studied.

HBV and HCV groups and healthy control group have not presented a statistically significant difference between their RF and anti-CCP positivity. However, RA group had higher RF and anti-CCP levels and displayed a striking difference to the rest of the groups (p < 0.001 for each). Group laboratory results are presented in Table 2. A comparison of anti-CCP positivity levels of groups (Table 3) presented that hepatitis B, hepatitis C and healthy controls had lower levels of positivity (11.4%, 20.9%, 10.9% respectively). However, RA group showed mostly high levels of anti-CCP positivity (56%).

Discussion

In this study, we have analyzed anti-CCP positivity in asymptomatic chronic hepatitis B and C patients. We determined the anti-CCP positivity to be 20.5% and 32.5% in HBV and HCV patients respectively. However, most of these were low-level positivity.

Autoantibodies are widely used in rheumatoid disease diagnostics. Antinuclear antibody (ANA), RF and anti-CCP are among the most commonly used autoantibodies in clinic practice. RF has been used in RA diagnostics since 1970s and is amongst 1987 ACR criteria and 2010 classification criteria.[36]

According to the literature, RF positivity changes between 17.5–42.7% in HBV patients and between 9.7–54% in HCV patients. A correlation between RF, arthralgia and arthritis has been determined.[16,17,19,20,22,24,25]

Our study also resulted in 11.4% RF levels in HBV patients and 16.3% for HCV patients, results not falling far from previous studies.

RF can also be induced in chronic infections (such as osteomyelitis, tuberculosis and subacute bacterial endocarditis) and acute infections besides chronic hepatitis. We had determined in a previous study that Brucellosis patients with arthritis had an RF level of 20%. In addition to those, RF could also be positive in rheumatoid diseases such as Sjögren's Syndrome, systemic lupus erythematosus, cryoglobulinemia and in healthy individuals.[37,38]

Anti-CCP was first used in the 2000s as a RA diagnosis tool and took its place amongst classification criteria in 2010. Previous studies show anti-CCP levels of HBV patients as between 0–13.9% (Table 4), and between 0–33% in HCV patients. Similarly, there has been a correlation detected between arthritis and anti-CCP positivity in these patients. Riccio et al. and Bassyouni et al. have determined 33% and 20%, respectively, of anti-CCP positivity in HCV patients with musculoskeletal symptoms.[19,29] In asymptomatic HCV patients, Liu Feng-Cheng et al. found 5.2% and Orge et al. 4.9% of anti-CCP positivity.[14,19,20,24,25] We determined a 20.5% anti-CCP positivity in HBV and a 32.5% in HCV in our study. Our study showed higher Anti-CCP positivity than previous studies, but significant part of them in low titers (11.4% in HBV, 20.9% in HCV in low titer).

Anti-CCP antibodies can be positive in other infections like tuberculosis (37%) and Lyme disease (2%) besides chronic hepatitis. As much as it is a sensitive and specific marker in RA diagnosis, it still can be found positive in systemic lupus erythematosus (8–17%), Sjögren's Syndrome (3–7.5%), scleroderma (5–10.6%), ulcerative colitis (3%), fibromyalgia (3%) and polymyositis or dermatomyositis (14%).[5,7–9,14,34]

There are some limitations to our study. First of all, none of the patients had symptoms. An evaluation of symptomatic hepatitis patients could have provided more detailed information. Secondly, patients were not observed over a long period of time, so there was no data about long-term consequences in our findings.

In conclusion, we determined that RF and anti-CCP positivity is between 11.4% and 20.5% in HBV patients and between 16.3% and 32.5% in HCV patients. Therefore, chronic hepatitis should be considered a possibility particularly in patients with positive anti-CCP antibodies and low RF titer.

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