Original Article

Serum Anti-SARS-CoV-2 IgG in Chronic Hepatitis B Virus Positive Patients

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Abstract:

Background: Advanced age and comorbidities such as diabetes mellitus, hypertension, chronic obstructive pulmonary disease or cardiovascular diseases are proven risk factors in COVID-19. In COVID-19, pre-existing liver illnesses could be a substantial risk factor for hospitalization and severity. Infection with the hepatitis B virus (HBV) is one of the most significant and common health disorders. **Objective:** To estimate serum anti-SARS-CoV-2 IgG in chronic Hepatitis B virus positive patients. **Methodology:** This cross-sectional analytical study was conducted in the Department of Biochemistry, Dhaka Medical College, Dhaka, during the period of January 2021 to December 2021. A total of 63 subjects were selected from Hepatology Department of DMCH and BSMMU as per selection criteria. Among them, 23 were chronic HBV positive patients (3 with COVID-19 and 20 without COVID-19) (group A) and the rest 40 were non HBV cases (20 with COVID-19 and 20 without COVID-19) (group B). Blood sample was collected for the measurement of anti-SARS-CoV-2 IgG. Biochemical analysis was performed by chemiluminescent microparticle immunoassay (CMIA) in the Department of Biochemistry, BSMMU, Dhaka. **Result:** In this study, the mean age of group A was 47.26 ± 11.24 years and group B was 44.53 ± 10.17 years. Males were predominant in both the groups. In group A, male was 65.2% & female was 34.8% and in group B, male was 60% & female was 40%. The median value of serum anti-SARS-CoV-2 IgG level was 1110.70 in group A and 417.85 in group B ($p>0.05$). The study showed frequency of positivity of serum anti-SARS-CoV-2 IgG level in COVID-19 positive cases was 95.7% and in COVID-19 negative cases it was 90%. In this study, the median value of serum anti-SARS-CoV-2 IgG level significantly higher in COVID-19 positive (1063.20) than in COVID-19 negative (119.00) in non-HBV cases. **Conclusion:** Serum anti-SARS-CoV-2 IgG level was non-significantly higher in chronic HBV positive patients than non-HBV patients.

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Introduction:

Corona virus disease 2019 (COVID-19) is a pandemic disease declared by the World Health Organization (WHO) on 11th march, 2020, which is caused by severe acute respiratory syndrome corona virus 2 (SARS-CoV-2). The virus was first identified as the cause of an outbreak of pneumonia of unknown cause in Wuhan city, Hubei province, China in December 2019.[1] At October 2020, more than 3,90,00,000 cases of SARS- CoV-2 have been detected worldwide, leading to near 11,00,000 deaths.[2] In Bangladesh, COVID-19 infections are being reported from the Directorate General of Health Service on daily basis. So far, around 4,10,988 cases with 5,966 deaths upto 2 November, 2020.[1]

The global risk assessment by the WHO is quite high, and community transmission is occurring in many countries, but how easily the virus transmits between people is unknown.[1] The respiratory and gastrointestinal symptoms of COVID-19 have been recorded in several, including fever, cough, dyspnea, vomiting, diarrhea, and abdominal pain.[3] SARS-CoV-2 is an enveloped corona virus having a genomic sequence that is 80%

identical to SARS-CoV-1 and 96.2 percent similar to bat corona virus RaTG13.[4] It belongs to the Sarbeco virus subgenus of the Coronaviridae family and is the seventh corona virus known to infect humans, the transmission of which is person to person through fomites and respiratory droplets. It is a positive stranded RNA virus with an estimated genome size equal to 29.9 kb.[2] SARS-CoV-2 is transmitted primarily through infected respiratory droplets, with viral infection occurring through direct or indirect contact with nasal, conjunctival or oral mucosa. Contact pattern, environment, infectiousness of the host, and socioeconomic factors all influence transmission risk.[4]

COVID-19 is a self-limiting infection. However, fatality rates of 3-7 percent have been reported in rare circumstances. Organ failure, particularly acute respiratory distress syndrome, is caused by excessive production of pro-inflammatory cytokines. COVID-19 has demonstrated risk factors such as co-morbidities and advanced age.[5] SARS-CoV-2 interacts to the host cell receptor ACE2. Fever, headaches, and respiratory symptoms are common indications of active viral replication and release in the lungs. The locations of infection and symptoms may be explained by the distribution of ACE2 receptors in different tissues.[4] The ACE2 receptor is expressed on a variety of cell types, including hepatocytes and cholangiocytes, and SARS-CoV-2 has a strong affinity for it. Upregulation of ACE2 receptors in the liver may contribute to abnormal liver enzyme activity reported in COVID-19 patients.[3]

Knowledge of the pathogenesis of SARS-CoV-2 is still accumulating. Besides pulmonary injury, other organs and cell types are damaged during the illness. Upto 60% patients suffer from liver damage and chronic Hepatitis B virus infection was shown to be independent risk factors for disease progression to acute respiratory distress syndrome, in case of SARS patients.[6] Hepatitis B virus is a hepadnaviridae family double-stranded DNA virus that is enveloped and includes a viral DNA genome within its core. This virus infection is one of the most dangerous and widespread health concerns, affecting about 2 billion people globally. Based on an intergroup difference of more than 8% in the entire nucleotide sequence, the virus includes at least eight primary genotypes (A to H). It is transmitted parenterally through percutaneous or permucosal exposure to infected blood or body fluids, which might be visible or not.[7] Chronic hepatitis B is liver inflammation caused by the hepatitis B virus that lasts longer than 6 months.[8]

Against SARS-CoV-2, the human adaptive immune system usually mounts a unique response, including the generation of specific IgM, IgG, and IgA antibodies. IgM levels start to rise at 5 days after infection and then level off around 10 days. IgG is delayed at first, but after about 10 days, it surpasses IgM concentration. Antibody response varies depending on the severity of illness.[9] Antibody testing of representative populations, including asymptomatic individuals, can reveal the true spread of COVID-19. Antibody tests tend to demonstrate a higher population-wide prevalence of SARS-CoV-2 than RT-PCR tests.[9] The immune response to SARS-CoV-2 infection in chronic HBV-positive patients is still unknown.

Methods:

This cross-sectional analytical study was conducted in the Department of Biochemistry, Dhaka Medical College, Dhaka from January 2021 to December 2021 over a period of one year. A total of 63 chronic HBV positive patients with or without history of diagnosed COVID-19 infection, selected from Hepatology Department of DMCH and BSMMU. Patients were divided into two groups: Group-A consisted with chronic HBV positive patients, with or without history of COVID-19 infection and Group-B consisted with non HBV patients, with or without history of COVID-19 infection (based on positive RT-PCR & / HRCT findings of lung diagnosed within 3 to 12 weeks from the time of sample collection). Patients with malignancy, pregnancy, HCV positive and COVID-19 vaccinated cases were excluded from this study.

Twenty-three of them were chronic HBV positive patients (3 with COVID-19 and 20 without COVID-19) and the rest 40 subjects were non-HBV patients (20 with COVID-19 and 20 without COVID-19). Purpose and procedure of the study were elaborately explained to each of the study subjects and informed written consent was taken from them. Data collection procedure was including direct history taking, physical examination and laboratory procedure. Venous blood was collected and serum anti-SARS-CoV-2 IgG was estimated by chemiluminescent microparticle immunoassay (CMIA) technology. Data was recorded in a pre-designed data collection sheet.

Collection and Preservation of blood samples:

After all aseptic precaution, 5 ml of venous blood sample was collected from median cubital vein from each study subject with a EDTA containing vacuum tube. A clear serum was separated after centrifuging

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the blood at 3000 rpm for10 minutes. The separated serum was stored at -20C and then used for the measurement of anti-SARS-CoV- 2 IgG. Biochemical analysis was performed by chemiluminescent microparticle immunoassay (CMIA) technology, in the Department of Biochemistry, BSMMU, Dhaka. Data was recorded in a pre-designed data collection sheet.

After meticulous checking and rechecking, all the data were recorded in a predesigned data collection sheet. All analysis was done using the SPSS 22.0 (Statistical package for social science). Continuous variables were expressed as mean ± SD and were compared between subjects by unpaired student's t-test in case of normal distribution or median or IQR compare by Mann-Whitney U-test in case of skewed distribution. Categorical variables were compared using Chi-square test or Fisher's exact test as appropriate and were presented as absolute frequencies with percentages. A value of 'p' < 0.05 was considered as level of significance.

Results:

Table I: Demographic profile of the study subjects according to age (n=63)

	Group A (n=23) n (%)	Group B (n=40) n (%)	p-value
Age (years)	47.26 ± 11.24	44.53 ± 10.17	[b]0.326
Gender			
Male	15 (65.2)	24 (60.0)	[a]0.681
Female	8 (34.8)	16 (40.0)	

aChi-Square test and bUnpaired t test was done to measure the level of significance The mean age of group A was (47.26 ± 11.24) and group B was (44.53± 10.17), difference was not statistically significant (p > 0.05) between these two groups. In group A, 65.2% male and 34.8% female and in group B, 60% male and 40% female. The gender difference was not statistically significant (p > 0.05) between these two groups (Table 1).

Table II: Co-morbidities of the study subjects (n=63)

Co-morbidity	Group A (n=23) n (%)	Group B (n=40) n (%)	p-value
Diabetes mellitus	4 (17.4)	6 (15.0)	1.000
Hypertension	3 (13.0)	6 (15.0)	1.000
CKD	0 (0.0)	1 (2.5)	1.000
COPD	0 (0.0)	1 (2.5)	1.000

Fisher's Exact test was done to measure the level of significance

In group A, DM (17.4%), HTN (13%) and in group B, DM (15%), HTN (15%), CKD (2.5%), COPD (2.5%). The result was not statistically significant (p >0.05) between these two groups (Table 2).

Table III: Comparison of Serum Anti-SARS-CoV-2 IgG level between chronic and non-chronic HBV patients (n=63)

Serum Anti-SARS-CoV-2 IgG (AU/ML)	Group A (n=23)	Group B (n=40)	p-value
Median	1110.70	417.85	0.232
IQR	250.3 – 2323.4	104.9 – 2805.9	

Mann-Whitney U test was done to measure the level of significance

The median value of group A was 1110.70 and group B was 417.85. The result was not statistically significant (p > 0.05) between these two groups (Table 3).

Table IV: Comparison of Serum Anti-SARS-CoV-2 IgG between COVID positive and COVID negative in Group A (n=23)

	COVID+ve (n=3)	COVID– ve(n=20)	p-value
Median	840.60	1213.7	0.763
IQR	254.8 – 840.60	155.5 – 2633.5	

Mann-Whitney U test was done to measure the level of significance

The median value of COVID-19 positive was 840.60 and COVID-19 negative was 1213.7. The result was not statistically significant (p > 0.05) in group A (Table 4).

Table V: Comparison of Serum Anti-SARS-CoV-2 IgG between COVID positive and COVID negative in Group B (n=40)

	COVID+ve (n=20)	COVID–ve (n=20)	p-value
Median	1063.20	119.00	0.020
IQR	240.4 – 7586.0	76.9 – 1002.0	

Mann-Whitney U test was done to measure the level of significance

The median value of COVID-19 positive was1063.20 and COVID-19 negative was 119.00. The result was statistically significant (p<0.05) in group B (Table 5).

Table VI: Frequency of positivity of antibody in COVID positive and COVID negative patients (n=63)

	COVID +ve (n=23)	COVID -ve (n=40)	p-value
Positive (>50)	22 (95.7)	36 (90.0)	0.644
Negative (≤50)	1 (4.3)	4 (10.0)	

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Fisher's Exact test was done to measure the level of significance

In COVID-19 positive (95.7% positive, 4.3% negative) and in COVID-19negative (90% positive, 10% negative). The result was not statistically significant (p >0.05) (Table 6).

Table VII: Frequency of positivity of antibody in HBV positive and HBV negative patients (N=63)

	HBV +ve (n=23)	HBV- ve (n=40)	p-value
Positive (>50)	21 (91.3)	37 (92.5)	1.000
Negative (≤50)	2 (8.7)	3 (7.5)	

Fisher's Exact test was done to measure the level of significance

In chronic HBV positive patients, 91.3% positive and 8.7% negative. In non HBV patients, 92.5% positive and 7.5 % negative. The result was not statistically significant (p >0.05).

Discussion:

In this present study, the mean age of group A and group B were (47.26 ± 11.24) years and (44.53 ± 10.17) years respectively. The difference was not statistically significant between these two groups (p >0.05). Ahmed et al. also found the mean age of the patients (42 ± 14.5) which was almost similar to this study.[10] In group A, 15 (65.2%) were male and 8 (34.8 %) were female. In group B 24 (60%) were male and 16 (40 %) were female. Male gender was predominant in both groups. No significant difference was found between the groups (p>0.05). Liu et al. also found 66.7% male in their study, which was almost similar to this study.[6]

According to the co-morbidities of the study subjects, DM was 17.4% and HTN was 13 % in group A and DM was 15%, HTN was 15%, CKD was 2.5% and COPD was 2.5% in group B. Zádori et al. found DM (14%) and HTN (13.1%) as co-morbidities in their study.[11] In this study, the median value of serum anti-SARS-CoV-2 IgG level in group A (1110.70) and in group B (417.85). The result was not statistically significant (p>0.05). There was no significant difference of serum anti-SARS-CoV-2 IgG level between chronic HBV positive patients and non-HBV patients.

In HBV positive cases, the median value of serum anti-SARS-CoV-2 IgG level in COVID-19 positive was 840.60 and in COVID-19 negative was 1213.7. The result was not significant statistically (p>0.05). In HBV negative cases, the median value of serum anti-SARS-CoV-2 IgG level in COVID-19 positive was 1063.20 and in COVID-19 negative was 119.00 in group B. The result was significant statistically (p <0.05). Ahmed et al. also found that the antibody level of IgG in participants with RT-PCR positive cases, significantly higher.[10] In the present study, the frequency of positivity of serum anti-SARS-CoV-2 IgG in COVID-19 positive (positive 95.7% and negative 4.3%) and in COVID-19 negative (positive 90% and negative 10%) within both groups has found. There was no significant difference of positivity of serum anti-SARS-CoV-2 IgG between COVID-19 positive and COVID-19 negative patients. This may be due to several mild/asymptomatic/recovered cases that were never tested using RT-PCR for various reasons or because of the ineffectiveness of RT-PCR in detecting previous infections. In this study, the frequency of positivity of serum anti-SARS-CoV-2 IgG in HBV positive was 91.3% and in HBV negative was 92.5% (p >0.05). There was no significant difference of positivity of serum anti- SARS-CoV-2 IgG between HBV positive and HBV negative patients.

Conclusion:

In this present study, the median value of serum anti-SARS-CoV-2 IgG was 1110.70 in chronic HBV positive patient and 417.85 in non HBV patient. But the difference was not statistically significant. The study also showed positivity of serum anti-SARS-CoV-2 IgG level in COVID-19 positive (positive 95.7% and negative 4.3%) and in COVID-19 negative (positive 90% and negative 10%) patients, which was not significant. The median value of serum anti-SARS-CoV-2 IgG in COVID-19 positive was 1063.20 and 119.00 in COVID-19 negative cases among HBV negative cases.

References:

1. National Guideline on Clinical Management of COVID-19 2020. Mohfw.gov.bd. 2021. Available at: http://www.mohfw.gov.bd/index.php?option= com_docman & task = doc_down load & gid = 2242 Viewed on 17/06/2021.

2. Canedo-Marroquin G, Saavedra F, Andrade CA, Berrios RV, Rodriguez-Guilarte L, Opazo MC, et al. SARS-CoV-2: immune response elicited by infection and development of vaccines and treatments. Frontiers in immunology. 2020:3259.

DOI: https://doi.org/10.3329/jcomcta.v28i1.75714

3. Aldhaleei WA, Alnuaimi A, Bhagavathula AS. COVID-19 induced hepatitis B virus reactivation: a novel case from the United Arab Emirates. Cureus. 2020 Jun 15;12(6).

4. Cevik M, Kuppalli K, Kindrachuk J, Peiris M. Virology, transmission, and pathogenesis of SARS-CoV-2. bmj. 2020 Oct 23;371.

5. Hegyi PJ, Váncsa S, Ocskay K, Dembrovszky F, Kiss S, Farkas N, et al. Metabolic associated fatty liver disease is associated with an increased risk of severe COVID-19: a systematic review with meta-analysis. Frontiers in medicine. 2021:277.

6. Liu X, Wang J, Xu X, Liao G, Chen Y, Hu CH. Patterns of IgG and IgM antibody response in COVID-19 patients. Emerging microbes & infections. 2020 Jan 1;9(1):1269-74.

7. Liaw YF, Chu CM. Hepatitis B virus infection. The lancet. 2009 Feb 14;373 (9663):582-92.

8. Kumar S. Hepatitis B, chronic 2021. Available at: https://www.msdmanuals.com/ home/ liver- and-gallbladder-disorders/hepatitis/hepatitis-b-chronic. Viewed on 02/05/2021

9. Vogl T, Leviatan S, Segal E. SARS-CoV-2 antibody testing for estimating COVID-19 prevalence in the population. Cell Reports Medicine. 2021 Feb 16;2 (2):100191.

10. Ahmed ZB, Razu MH, Akter F, Rabby M, Islam R, Karmaker P, et al. Seropositivity of SARS-CoV-2 IgG Antibody among People in Dhaka City during the Prevaccination Period. BioMed Research International. 2022 Jan 27;2022.

11. Zádori N, Váncsa S, Farkas N, Hegyi P, Erőss B. The negative impact of comorbidities on the disease course of COVID-19. Intensive care medicine. 2020 Sep;46(9):1784-6.

DOI: https://doi.org/10.3329/jcomcta.v28i1.75714