Association of Seroconversion Status with Outcome in Admitted Covid-19 Patients

Fatima Kanani[1], Sabiha Anis[2], Bushra Kaleem[3] and Saba Jamal[4]

[1]*Chemical Pathology Section, Department of Pathology, Indus Hospital & Health Network, Karachi, Pakistan*
[2]*Immunology Section, Department of Pathology, Indus Hospital & Health Network, Karachi, Pakistan*
[3]*Indus Hospital Research Center, Indus Hospital & Health Network, Karachi, Pakistan*
[4]*Indus Blood Center, Indus Hospital & Health Network, Karachi, Pakistan*

ABSTRACT

Objective: To determine the association between seroconversion status and outcome in admitted COVID-19 patients and compare inflammatory markers amongst them.

Study Design: Single cohort observational study.

Place and Duration of study: Indus Hospital and Health Network between 10th May and 10th July 2020.

Methodology: All admitted patients were tested serially for anti-COVID-IgM and IgG until their sera showed positive results. This was continued until their expiry or discharge. Those patients who remained negative for both anti-COVID-19-IgG and IgM were labeled as non-seroconverts. Demographics, comorbidities, inflammatory marker levels and outcome (alive/expired) were compared between seroconverts and non-seroconverts.

Results: In 224 admitted patients, the median seroconversion time of IgM and IgG was six and seven days in survivors and non-survivors respectively. Expired patients displayed higher levels of procalcitonin (maximum), C-reactive protein, and Interleukin-6 (baseline and maximum). Of 34 non-seroconverts, 17 (50%) expired. Non-seroconverts significantly failed to develop fever and had lower levels of ferritin, CRP, and LDH.

Conclusion: Non-seroconversion in hospitalised COVID-19 infected patients indicated muted immune and acute phase response and was associated with poor outcomes. Hence these patients need to be carefully evaluated and managed.

Key Words: *Antibody response, Corticosteroids, Immunosuppression, SARS-Cov-2, Seroconversion.*

How to cite this article: Kanani F, Anis S, Kaleem B, Jamal S. Association of Seroconversion Status with Outcome in Admitted Covid-19 Patients. *J Coll Physicians Surg Pak* 2022; **32(07)**:874-879.

INTRODUCTION

The immune regulation in *Sars-Cov-2* infection has remained a focal point of research since the beginning of the pandemic. The longitudinal pattern of the natural antibodies in COVID-19 is variable and largely unknown for different grades of disease severity with certain studies reporting two weeks for maximum seroconversion[1] while others reporting earlier appearance in acutely sick patients.[2]

The immune system plays an important role in the pathogenicity of the disease. The rapid replication of *SARS-Cov-2* triggers inflammatory responses leading to recruitment of macrophages and monocytes. This results in the release of cytokines and chemokines responsible for cytokine storms.[3]

Correspondence to: Dr. Fatima Kanani, Department of Chemical Pathology, Indus Hospital and Health Network, Karachi, Pakistan
E-mail: fatima.kanani@tih.org.pk
Received: January 04, 2022; Revised: May 14, 2022; Accepted: May 19, 2022
DOI: https://doi.org/10.29271/jcpsp.2022.07.874

Numerous inflammatory markers help in identifying disease severity. Markers such as procalcitonin (PCT), serum ferritin, C-reactive protein (CRP), lactate dehydrogenase (LDH), and interleukin-6 (IL-6) have been found to be associated with a high risk of severe COVID-19.[4]

Although many studies have been published on the biomarker levels in COVID-19, there are relatively few which have evaluated their association with seroconversion and outcome (survival status). The present study aimed at evaluating the seroconversion times and the levels of various inflammatory markers in admitted COVID-19 patients to see their association with the disease outcome.

METHODOLOGY

All patients with positive *SARS Cov-2* PCR admitted in the COVID-19 unit of The Indus Hospital, Karachi between 10th May and 10th July 2020, irrespective of age or gender, were included in the study. Patients whose PCRs were not done or were observed to be negative were excluded. This single cohort study was conducted in accordance with the Declaration of Helsinki (2013 version) and approved by the institutional ethics board of the study centre (IRD_IRB_2020_07_010). Stored

serum samples from these patients were used to perform the serological tests. Serial patient samples till discharge or expiry were tested for both IgM and IgG -anti- *SARS Cov-2* antibodies using *SARS Cov-2* IgM IgG Antibody Assay kit (Zybio), a rapid test based on colloidal gold immunochromatographic method previously validated in our lab[5]. Electronic medical records of the patients were searched to ascertain days of onset of symptoms, comorbidities, levels of inflammatory markers, presence of anti- *SARS Cov-2* antibodies, and patient outcome. CRP (cutoff value: <05mg/L), and LDH (reference range: 125-220 U/L) were performed on Alinity c, serum ferritin (reference range: 10-250 ng/ml) by chemiluminescent microparticle immunoassay on Alinity i while PCT (reference range: <0.046 ng/ml) and IL-6 (reference range: <7.0 pg/ml) were performed on electrochemiluminescent technique on Roche e411. All admitted patients were tested serially for anti-COVID-IgM and IgG until their sera showed positive results (seroconverts). This was continued until their expiry or discharge (outcome). Those patients who remained negative for both anti-COVID-19-IgG and IgM were labeled as non-seroconverts. The impact of all the variables was observed on seroconversion as well as the outcomes *i.e*. survival status.

The normality of continuous variables such as days of age, admission, and levels of inflammatory markers was evaluated using the Shapiro-Wilk test. As the variables were not normally distributed, they were displayed by medians with quartiles while categorical parameters were displayed by percentages and frequencies number. Pearson's chi-square test was performed for categorical data. Mann-Whitney test was performed for multiple comparisons of continuous data. Age over 60 years, presence of at least one co-morbidity [Hypertension (HTN), Diabetes Mellitus (DM), Ischaemic Heart Disease (IHD), Chronic Obstructive Pulmonary Disease (COPD), End-Stage Renal Disease (ESRD), Chronic Liver Disease (CLD), Human Immunodeficiency Virus (HIV) infections, malignancy for at least 6 months)] and inflammatory markers i.e. LDH, CRP, PCT, ferritin, and IL-6 were the analysed variables. The statistical analysis was performed on SPSS version 26.0. A p-value of <0.05 was considered statistically significant.

RESULTS

The total number of study participants was 224. There were 151 (67.4%) survivors and 73 (32.6%) non-survivors. Baseline characteristics of study participants are shown in Table I. There was a preponderance of males as compared to female patients in both the groups (76.2/23.8% and 75.3/24.7%). There was no significant difference in the clinical features, days of hospitalisation, and co-morbidities between the survivors and non-survivors.

There was a median (IQR) time of 8 (5.0-12.5) days between admission and expiry in the non-survivors. Fever and cough were more common in the expired patients compared to survivors (68.5% and 53.4% *vs*. 64.2% and 45.7%). Diabetes mellitus was the most frequent co-morbidity and was more frequent in non-survivors (50.7 % *vs.* 43.7%).

Amongst the inflammatory biomarkers, maximum values of PCT were significantly higher in non-survivors, while baseline and maximum levels of CRP and IL-6 were also relatively higher in non-survivors as compared to survivors (Table I). The median seroconversion time for both IgM and IgG was 6 days (IQR: 3.0 – 9.0 days) in survivors and 7 days (IQR: 4.0-11.0 days) in non-survivors (Table I). In all, 34 patients did not show seroconversion during their hospital stay. Of note, 23.3% of non-survivors compared to 11.3% of survivors did not seroconvert.

A comparison of baseline inflammatory marker levels was done between seroconverts and non-seroconverts alone as well as on the basis of their survival status (Table II). Significantly, 50% of the non-seroconverts expired compared to 29.5% of seroconverts.

Non-seroconverts had fewer comorbidities and failed to develop fever more often than the seroconverts amongst both the survivors (p-value: 0.008) and non-survivors (Table II). They also had significantly lower LDH and CRP levels than the seroconverts in both survivors and non-survivors. Moreover, median ferritin values amongst non-seroconverting survivors were also significantly lower. However, median levels of PCT and IL-6 were higher in non-seroconverts for both survivors and non-survivors. High CRP levels at baseline carried 2.4 times more chances of death in patients (Table III).

DISCUSSION

This study took place during the first wave of the pandemic when the knowledge about disease pathophysiology, host immune responses, the efficacy of the pharmacological agents in use, and successful management protocols were all in the evolving phase. It was conducted on admitted patients who required hospital care and could not be managed at home or daycare facilities. These factors account for the relatively high mortality rate of almost 33% seen in this cohort.

The male to female ratio in the present study was 3.15:1. The median (IQR) age of the expired patients was 60.0 years (48.5 – 65.0 years). However, age did not impact neither non-seroconversion nor mortality. These figures were also similar within the survivors' group unlike those quoted in some other studies.[6] The reason for this was most likely the fact that our entire study cohort consisted of moderately to critically ill patients.

COVID-19 serology was performed serially on these 224 admitted patients and seroconversion was observed in 84.8% of the study cohort. Most studies on seroconversion after COVID-19 infection or exposure have been done in healthcare workers while few are community-based. A large-scale study in Pakistan reported a seroprevalence of ~7% in the general population with symptomatic or asymptomatic COVID-19 infection[7] while other studies reported seroprevalence between 36%-40% in health care workers or in a specific community.[8,9] There are limited studies that have assessed the antibody immune response in moderate to severe COVID-19 infected patients, as well as the outcome in these patients in relation to seroconversion.

Table I: Baseline characteristics, antibody, and inflammatory marker profile of study participants based on survival status.

Variables	Expired (n=73)	Alive (n=151)	p-value
Age, years [Median (IQR)]†	60.0 (48.5 – 65.0)	57.0 (45.0 – 65.0)	0.845
Gender [M/F, n (%)]‡	55 (75.3)/18 (24.7)	115 (76.2)/36 (23.8)	>0.99
Days of admission, [Median (IQR)]†	8.0 (5.0 – 12.5)	7.0 (5.0 – 9.0)	0.119
Symptoms			
Fever (n=147), n (%)‡	50 (68.5)	97 (64.2)	0.552
SOB (n=143), n (%)‡	43 (58.9)	100 (66.2)	0.302
Cough (n=108), n (%)‡	39 (56.5)	69 (46.6)	0.192
Diarrhea (12), n (%)‡	3 (4.1)	9 (6.0)	0.755
Vomiting (n=11), n (%)‡	4 (5.5)	7 (4.6)	0.752
Fatigue (n=11), n (%)‡	2 (2.7)	9 (6.0)	0.510
Co-morbidities			
Diabetes Mellitus (n=103), n (%)‡	37 (50.7)	66 (43.7)	0.391
Hypertension (n=94), n (%)‡	31 (42.5)	63 (41.7)	>0.99
Ischemic Heart Disease (n=12), n (%)‡	4 (5.5)	8 (5.3)	>0.99
End-stage Renal Disease, (n=11), n (%)‡	3 (4.1)	8 (5.3)	>0.99
Chronic Obstructive Pulmonary Disease (n=11), n (%)‡	3 (4.1)	8 (5.3)	>0.99
No comorbidity (n=78), n (%)‡	28 (38.4)	50 (33.1)	0.457
Initial antibody appearance			
IgM/IgG (n=120), n (%)‡	35 (47.9)	85 (56.3)	0.240
IgG (n=69), n (%)‡	21 (28.8)	48 (31.8)	0.758
IgM (n=1), n (%)‡	0	1 (0.7)	>0.99
No seroconversion (n=34) ‡	17 (23.3)	17 (11.3)	0.019*
Time interval between symptom onset and seroconversion (days)			
IgM, days [Median (IQR)]†	7.0 (4.0 – 11.0)	6.0 (3.0 – 9.0)	0.118
IgG, days [Median (IQR)]†	7.0 (4.0 – 11.0)	6.0 (3.0 – 9.0)	0.315
IgM/IgG, days [Median (IQR)]†	7.0 (4.0 – 11.0)	6.0 (3.0 – 9.0)	0.120
Inflammatory markers			
BL Ferritin, ng/ml [Median (IQR)] †	726.2 (224.8 – 1635.2)	884.9 (409.2 – 1675.6)	0.189
Max. Ferritin, ng/ml [Median (IQR)]†	1542.2 (351.3 – 1838.1)	1349.4 (500.3 – 1716.7)	0.568
BL CRP, mg/L [Median (IQR)]†	130.5 (54.6 – 258.8)	119.0 (71.6 – 254.2)	0.289
Max CRP, mg/L [Median (IQR)]†	161.5 (60.9 – 256.9)	112.2 (87.0 – 229.9)	0.944
BL LDH, IU/L [Median (IQR)]†	421.0 (378.5 – 804.5)	473.0 (403.0 – 780.0)	0.778
Max LDH, IU/L [Median (IQR)]†	431.0 (327.5 – 769.5)	577.0 (421.0 – 713.0)	0.255
BL IL-6, pg/ml [Median (IQR)] †	40.3 (21.5 – 93.2)	25.5 (15.2 – 73.9)	0.803
Max IL-6, pg/ml [Median (IQR)] †	68.7 (15.5 – 836.5)	60.6 (16.7 – 184.8)	0.797
BL PCT, ng/ml [Median (IQR)] †	0.87 (0.14 – 1.85)	0.30 (0.12 – 0.58)	0.576
Max PCT, ng/ml [Median (IQR)]†	3.5 (0.14 – 7.6)	0.22 (0.09 – 0.98)	0.024*

*†Mann-Whitney U-test; ‡ Pearson's Chi-square test; *statistically significant.*

Chughtai *et al*. conducted a study on 75 admitted COVID-19 patients and reported 68% seroconversion by day 7 which increased to 100% by day 21.[10] However, the presence of comorbidities and non-survivors were not included. Aziz *et al*. studied the association of seroconversion with disease severity but did not discuss the outcome of these patients.[11] This is the first study in Pakistan, to the best of our knowledge, which has analyzed anti-COVID-19 antibody response in relation to other inflammatory markers and the outcome in admitted patients.

Our study showed median seroconversion of IgM and IgG at 6 -7 days' post symptoms onset in the cohort which was sooner than that reported in certain studies,[12,13] but concordant with others.[14] IgM and IgG paralleled each other closely during their entire course and there was no difference in their seroconversion time between the survivor and non-survivor groups. In fact, in almost 50% of the patients (120 out of 224), the initial test at hospital admission demonstrated the presence of both IgM and IgG. A postulate for this phenomenon could be that as all of these patients had moderate to severe disease, their antibody response was rapid and robust. Several researchers have also shown that immune response and seroconversion are earlier in severely symptomatic patients compared to milder or asymptomatic ones who tend to develop antibodies late *i.e*. up to several weeks post positive COVID-19 PCR.[15] In a study of 1000 COVID-19 patients from India, seroconversion was found to be associated with days after the symptom onset, disease severity, and presence of co-morbidity.[13]

The serial biomarker levels performed during the hospital stay of these patients were analysed. The baseline levels of the markers were in accordance with other studies.[16,17] Procalcitonin levels were significantly higher in non-survivors. This indicates that the patients may have succumbed due to secondary infections due to immunocompromised state. The baseline and maximum median (IQR) levels of IL-6 and CRP too were relatively higher in non-survivors compared to survivors. In fact, high CRP levels had 2.4 times higher odds of mortality. Of note, there was no significant difference in the known co-morbidities between the two groups.

Table II: Comparison of various factors between Seroconverts versus Non-seroconverts (individually and based on survival status).

	No seroconversion (n = 34) n (%)	Seroconversion (190) n (%)	p-value	Alive			Expired		
				No Seroconversion (n=17)	Seroconversion (n =134)	p-value	No Seroconversion (n=17)	Seroconversion (n=56)	p-value
Age [Median (IQR)]†	50.0 (36.0 – 65.0)	58.0 (45.8 – 65.0)	0.042*	40.0 (34.0 – 61.0)	58.0 (45.0 - 65.0)	0.008*	52.0 (42.0 – 65.5)	60.0 (50.0 – 64.8)	0.758
Outcomes									
Expired [n (%)]‡	17 (50.0)	56 (29.5)	0.028*	-	-		-	-	
Alive [n (%)]‡	17 (50.0)	134 (70.5)		-	-		-	-	
Presence of co-morbidities									
Yes [n (%)]‡	18 (52.9)	128 (67.4)	0.119	9 (52.9)	92 (68.7)	0.092	9 (52.9)	36 (64.3)	0.156
No [n (%)]‡	16 (47.1)	62 (32.6)		8 (47.1)	42 (31.3)		8 (47.1)	20 (35.7)	
Fever									
Present [n (%)]‡	16 (47.1)	131 (68.9)	0.013*	6 (35.3)	91 (67.9)	0.008*	10 (58.8)	40 (71.4)	0.142
Absent [n (%)]‡	18 (52.9)	59 (31.1)		11 (64.7)	43 (32.1)		7 (41.2)	16 (28.6)	
Serum Ferritin (ng/ml)									
Median (IQR)†	704.8 (358.1 – 1675.6)	991.9 (471.5 – 1675.6)	0.205	581.8 (325.1 – 1675.6 -)	1048.6 (492.1 – 1675.6)	0.072	908.9 (353.8 – 1616.3)	768.5 (414.5 – 1655.4)	0.951
<427.8‡	13 (38.2)	43 (22.6)	0.291	8 (47.1)	29 (21.6)	0.013*	5 (29.4)	14 (25.0)	0.133
427.8 - 927.8‡	7 (20.6)	49 (25.8)		3 (17.6)	31 (23.1)		4 (23.5)	18 (32.1)	
927.9 - 1675.6‡	14 (41.2)	96 (50.5)		6 (35.3)	72 (53.7)		8 (47.1)	24 (42.9)	
>1675.6‡	0	2 (1.1)		0	2 (1.5)		0	0	
Lactate Dehydrogenase (IU/L)									
Median (IQR)†	319.0 (258.3 – 631.8)	504.5 (405.3 – 691.5)	0.004*	316.0 (241.5 – 912.0)	487.0 (404.5 – 673.5)	0.111	322.0 (266.5 – 561.0)	522.0 (405.0 – 764.0)	0.017*
<380.8‡	19 (55.9)	35 (19.0)	0.000*	9 (52.9)	24 (18.6)	0.027*	10 (58.8)	11 (20.0)	0.006*
380.8 - 479.5‡	4 (11.8)	51 (27.7)		2 (11.8)	38 (29.5)		2 (11.8)	13 (23.6)	
480.0 - 686.5‡	3 (8.8)	52 (28.3)		1 (5.9)	37 (28.7)		2 (11.8)	15 (27.3)	
>686.5‡	8 (23.5)	46 (25.0)		5 (29.4)	30 (23.3)		3 (17.6)	16 (29.1)	
Procalcitonin (ng/ml)									
Median (IQR)†	0.57 (0.21 – 2.09)	0.32 (0.12 – 1.03)	0.049*	0.54 (0.28 – 5.75)	0.30 (0.12- 1.02)	0.046*	0.78 (0.16 – 2.09)	0.37 (0.09 – 1.30)	0.493
<0.129‡	4 (12.5)	52 (28.4)	0.200	1 (6.3)	37 (28.7)	0.018*	3 (18.8)	15 (27.8)	0.063
0.129 - 0.343‡	7 (21.9)	45 (24.6)		4 (25.0)	34 (26.4)		3 (18.8)	11 (20.4)	
0.344 - 1.28‡	10 (31.3)	42 (23.0)		6 (37.5)	28 (21.7)		4 (25.0)	14 (25.9)	
>1.28‡	11 (34.4)	44 (24.0)		5 (31.3)	30 (23.3)		6 (37.5)	14 (25.9)	
Interleukin-6(pg/ml)									
Median (IQR)†	32.4 (14.9 – 362.0)	28.5 (11.5 – 79.3)	0.401	585.2 (6.8 – 3971.3)	28.5 (11.6 – 79.1)	0.395	32.4 (19.9 – 56.4)	29.6 (11.1 – 89.5)	0.754
<12.2‡	2 (10.5)	37 (26.6)	0.326	2 (25.0)	26 (26.8)	0.101	0	11 (26.2)	0.099
12.2 - 29.4‡	7 (36.8)	33 (23.7)		2 (25.0)	23 (23.7)		5 (45.5)	10 (23.8)	
29.5 - 84.2‡	4 (21.1)	36 (25.9)		0	25 (25.8)		4 (36.4)	11 (26.2)	
>84.2‡	6 (31.6)	33 (23.7)		4 (50.0)	23 (23.7)		2 (18.2)	10 (23.8)	
C-Reactive Protein (mg/L)									
Median (IQR)†	85.8 (48.6 – 147.6)	145.4 (76.3 – 223.9)	0.001*	81.8 (52.2 – 136.7)	133.5 (65.4 – 205.2)	0.015*	85.9 (27.2 – 159.4)	149.4 (77.1 – 229.9)	0.032*
<66.0‡	15 (45.5)	41 (21.6)	0.002*	8 (47.1)	27 (20.1)	0.008*	7 (43.8)	14 (25.0)	0.011*
66.0 - 132.2‡	7 (21.2)	49 (25.8)		2 (11.8)	35 (26.1)		5 (31.3)	14 (25.0)	
132.3 - 208.4‡	10 (30.3)	46 (24.2)		6 (35.3)	29 (21.6)		4 (25.0)	17 (30.4)	
>208.4‡	1 (3.0)	54 (28.4)		1 (5.9)	43 (32.1)		0	11 (19.6)	

*†Mann-Whitney U-test; ‡ Pearson's Chi-square test; *statistically significant.*

The studies that have addressed the absence of antibody response in patients with moderate to severe infections are limited and showed a frequency of 8.8% to 25%.[16,17] They either showed a favorable response in those who did not seroconvert[16] or a variable outcome.[17] In this study, a total of 34 patients (15%) did not seroconvert till their death or discharge. There was no significant difference in the co-morbidities or gender between the two groups, although non-seroconverts were relatively younger than seroconverts. More importantly, a significantly greater percentage of patients who did not seroconvert succumbed to COVID-19 infection (50%) compared to those who were seroconverted (29.3%). Non-seroconverts relatively failed to develop fever and had lesser inflammatory response compared to seroconverts, with lower CRP and LDH levels, which has also been shown previously.[18] Several explanations are possible for the blunted humoral response in non-seroconverts. The immune response in these patients might be targeted against other antigens and be T cells mediated. Another plausible reason is that the antigenic exposure is limited to the mucosal surface of the respiratory tract in these patients and does not trigger the humoral response. Some researchers have shown that patients with higher viral loads and low cycle threshold values show delayed viral clearance and fail to seroconvert.[18] Gallais *et al*. suggest that after exposure to COVID-19, subjects can mount a virus-specific T-cell response without seroconversion in a study based on COVID-19 contacts.[19] In a Spanish study with a mean age of cohort more than 65 years, the muted immune response was noted in patients with the severe disease which were also related to immunodeficiency in a subset of patients.[16] Thakkar *et al*. did a study in COVID-19 infected patients with malignancies and found that 18% patients with hematological malignancies and 5.5% with solid tumors did not mount an anti-COVID antibody response.[20] They also observed that these patients had high morbidity and mortality. Moreover, they also found an association between anti-CD20, CART cell and corticosteroid therapy and an absence of seroconversion. A meta-analysis reported variable and controversial anti-COVID-19 responses in patients with different severity of diseases. However, the authors also concluded that heightened immune response was usually correlated to severe disease, but at the same time, patients with immunodeficiency and with muted immune response had a poor outcome.[3]

Table III: Odds Ratio for Mortality.

Variables	Odds Ratio	p-value	95% Confidence interval
Age			
>60 years	0.919	0.772	0.520 – 1.624
Gender			
Male	1.045	0.893	0.545 – 2.003
Comorbids			
>=1 comorbidities	1.257	0.440	0.703 – 2.247
Fever			
Present	0.826	0.530	0.456 - 1.499
Ferritin (ng/ml)			
427.8 - 927.8	0.794	0.556	0.367 – 1.714
927.9 - 1675.6	1.252	0.523	0.628 – 2.494
>1675.6	1.000		
CRP (mg/L)			
66.0 - 132.2	1.168	0.693	0.539 – 2.533
132.3 - 208.4	1.000	1.000	0.465 – 2.149
>208.4	2.4	0.044*	1.022 – 5.637
LDH (IU/L)			
380.8 - 479.5	1.697	0.199	0.757 – 3.804
480.0 - 686.5	1.422	0.383	0.645 – 3.139
>686.6	1.172	0.690	0.536 – 2.562
PCT (ng/ml)			
0.129 - 0.343	1.286	0.553	0.560 – 2.950
0.344 - 1.28	0.895	0.785	0.402 – 1.992
>1.28	0.829	0.640	0.378 – 1.818
IL-6 (pg/ml)			
12.2 - 29.4	0.655	0.381	0.254 – 1.687
29.5 - 84.2	0.655	0.381	0.254 – 1.687
>84.2	0.884	0.804	0.334 – 2.341

The present study patient cohort neither had malignancies nor were they on immunosuppressive drugs prior to getting COVID-19 infection. As per the study center's management guidelines, all moderate to severely infected patients received intravenous steroids. Taken together, the genetic susceptibility and differential response to steroid therapy may be a reason of high morbidity and mortality in our patients who did not seroconvert, however, this needs to be explored further.

This study had a few limitations. The patient cohort all belonged to the moderate to a severely critical group. Secondly, quantitative antibody levels were not performed. Patients were only followed till their discharge from the hospital for their immunological and inflammatory status. A larger study is required to ascertain the factors responsible for non-seroconversion in severe diseases.

CONCLUSION

Seroconversion status can act as a marker for modifying and personalising the treatment options in COVID-19 by helping in the judicial use of immunosuppressive drugs including corticosteroids. This is also supported by the fact that in this study PCT levels were significantly high in the expired patients and relatively higher in non-seroconverts suggesting a high probability of bacterial infections in them secondarily to immunosuppression.

ETHICAL APPROVAL:

The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013) and was approved by the institutional ethics board of The Indus Hospital (IRD-IRB-2020-07-010).

PATIENTS' CONSENT:

Due to the retrospective nature of the study and the use of stored and de-identified samples, patients' consent was not required.

COMPETING INTEREST:

The authors declared no competing interest.

AUTHORS' CONTRIBUTIONS:

FK: Contributed to the concept, and design of the study, acquisition of data, drafting the article, and critically reviewing for intellectual content.
SA: Contributed to drafting the article, analysis and critically reviewing for intellectual content. BK: Contributed to analysis, interpretation of data and drafting of the manuscript.
SJ: Contributed to critically reviewing for intellectual content.
All authors approved the final version of the manuscript to be published.

REFERENCES

1. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, *et al*. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet* 2020; **395(10223)**:497-506. doi: 10.1016/S0140-6736(20)30183-5. doi: 10.1016/S0140-6736(20)30183-5.
2. Chvatal-Medina M, Mendez-Cortina Y, Patino PJ, Velilla PA, Rugeles MT. Antibody responses in COVID-19: A review. *Front Immunol* 2021; **12**:633184. doi: 10.3389/fimmu.2021.633184.
3. Schietzel S, Anderegg M, Limacher A, Born A, Horn MP, Maurer B, *et al*. Humoral and cellular immune responses on *SARS-CoV-2* vaccines in patients with anti-CD20 therapies: A systematic review and meta-analysis of 1342 patients. *RMD Open* 2022; **8(1)**:e002036. doi: 10.1136/rmdopen-2021-002036.
4. Qin C, Zhou L, Hu Z, Zhang S, Yang S, Tao Y, *et al*. Dysregulation of immune response in patients with Coronavirus 2019 (COVID-19) in Wuhan, China. *Clin Infect Dis* 2020; **71(15)**:762-8. doi: 10.1093/cid/ciaa248.
5. Kanani F, Jamal S, Khowaja S, Kaleem B, Anis S, Iftikhar S, *et al*. Sensitivities and specificities of Anti-*SARS CoV2* detection kits-comparison and agreement between fifteen assays. *Jpn J Infect Dis* 2021; JJID. 2021.031. doi: 10.7883/yoken.JJID.2021.031.
6. Chen T, Wu D, Chen H, Yan W, Yang D, Chen G, *et al*. Clinical characteristics of 113 deceased patients with coronavirus disease 2019: Retrospective study. *BMJ* 2020; **368**:m1091. doi: 10.1136/bmj.m1091.
7. Ahmad AM, Shahzad K, Masood M, Umar M, Abbasi F, Hafeez A. COVID-19 seroprevalence in Pakistan: A cross-sectional study. *BMJ Open* 2022; **12(4)**:e055381. doi: 10.1136/bmjopen-2021-055381.
8. Abbas S, Raza A, Iftikhar A, Khan A, Khan S, Yusuf MA. Seroprevalence of *SARS-CoV-2* antibodies among health care personnel at a health care system in Pakistan. *Asia Pac J Public Health* 2021; **33(5)**:632-4. doi: 10.1177/10105395211011010.
9. Zaidi S, Rizwan F, Riaz Q, Siddiqui A, Khawaja S, Imam M, *et al*. Seroprevalence of anti-*SARS-CoV-2* antibodies in resi-

dents of Karachi — challenges in acquiring herd immunity for COVID 19. *J Public Health* 2021; **43(1)**:3-8. doi: 10.1093/pubmed/fdaa170.

10. Chughtai OR, Batool H, Khan MD, Ashraf S. Seroconversion in newly diagnosed cases of coronavirus disease. *J Coll Physicians Surg Pak* 2020; **30(8)**:801-4. doi: 10.29271/jcpsp.2020.08.801.
11. Aziz H, Fatima S, Mahmood H, Muhammad S, Saeed MA, Khurshid S, *et al*. Antibody response to *SARS-CoV-2* in relation to the contributing factors in COVID-19 patients. *Viral Immunol* 2022; **35(2)**:142-9. doi: 10.1089/vim.2021.0097.
12. Zhao J, Yuan Q, Wang H, Liu W, Liao X, Su Y, *et al*. Antibody responses to S*ARS-CoV-2* in patients of novel coronavirus disease 2019. *Clin Infect Dis* 2020; **71(16)**:2027-34. doi: 10.1093/cid/ciaa344.
13. Yadav AK, Ghosh S, Kotwal A, Kaushik S, Bobdey S, Sahu R, *et al*. Seroconversion among COVID-19 patients admitted in a dedicated COVID hospital: A longitudinal prospective study of 1000 patients. *Med J Armed Forces India* 2021; **77**:S379-S84. doi: 10.1016/j.mjafi.2021.06.007.
14. Zhang G, Nie S, Zhang Z, Zhang Z. Longitudinal change of severe acute respiratory syndrome coronavirus 2 antibodies in patients with coronavirus disease 2019. *J Infect Dis* 2020; **222(2)**:183-8. doi: 10.1093/infdis/jiaa229.
15. Kong WH, Zhao R, Zhou JB, Wang F, Kong DG, Sun JB, *et al*. Serologic response to *SARS-CoV-2* in COVID-19 patients with different severity. *Virol Sin* 2020; **35(6)**:752-7. doi: 10.1007/s12250-020-00270-x.
16. Jurado A, Martin MC, Abad-Molina C, Orduna A, Martinez A, Ocana E, *et al*. COVID-19: Age, Interleukin-6, C-reactive protein, and lymphocytes as key clues from a multicentre retrospective study. *Immun Ageing* 2020; **17**:22. doi: 10.1186/s12979-020-00194-w.
17. Shah C, Grando DJ, Rainess RA, Ayad L, Gobran E, Benson P, *et al*. Factors associated with increased mortality in hospitalised COVID-19 patients. *Ann Med Surg* 2020; **60**:308-13. doi: 10.1016/j.amsu.2020.10.071.
18. Masia M, Telenti G, Fernandez M, Garcia JA, Agullo V, Padilla S, *et al*. *SARS-CoV-2* seroconversion and viral clearance in patients hospitalised with COVID-19: Viral load predicts antibody response. *Open Forum Infect Dis* 2021; **8(2)**:1-8. doi: 10.1093/ofid/ofab005.
19. Gallais F, Velay A, Nazon C, Wendling MJ, Partisani M, Sibilia J, *et al*. Intrafamilial Exposure to *SARS-CoV-2* associated with cellular immune response without seroconversion, France. *Emerg Infect Dis* 2021; **27(1)**:113-21. doi: 10.3201/eid2701.203611.
20. Thakkar A, Pradhan K, Jindal S, Cui Z, Rockwell B, Shah AP, *et al*. Patterns of seroconversion for *SARS-CoV2*-IgG in patients with malignant disease and association with anticancer therapy. *Nat Cancer* 2021; **2(4)**:392-9. doi: 10.1038/s43018-021-00191-y.