Original Article

IL-32 is a Novel Regulator for Increasing Caspase-1 Gene Expression as a Proinflammatory in Early SARS-CoV-2 Infection

Rand J. Sattar, Hula Y. Fadhil[1]

Department of Biotechnology, Department of Biology, College of Science, University of Baghdad, Iraq, [1]Molecular Virology and Bioinformatics, Department of Biology, College of Science, University of Baghdad, Baghdad, Iraq

Abstract

Objective: The aim of this study is to determine whether the levels of caspase-1 and IL-32 gene expression are associated with the severity and progression of COVID-19 and its correlation with other clinical and biological markers of infection. **Materials and Methods:** About 100 patients with coronavirus infectious disease (COVID-19) and 58 healthy individuals were enrolled to assess the gene expression of interleukin-32 (IL-32) and caspase-1 by reverse transcription-quantitative polymerase chain reaction (RT-qPCR), with some emphasis on clinical and biological markers of disease like c-reactive protein, chemokine (CCL-2), and tumor protein (P53). **Results:** The finding showed that CCL-2 and P53 levels in patients showed a significant decrease (185 ng/L and 24.5 ng/mL) than in the control group (229 ng/L and 30 ng/mL) ($P = 0.001$). The ratio of relative IL-32 and caspase-1 gene expression in COVID-19 patients compared to healthy control was 1.5 and 1.2, respectively. Although the $2^{-\Delta\Delta Ct}$ of IL-32 and caspase-1 expression showed no significant variation between the age group (≤45 and > 45 years), sex, and severity in patients and controls, nonstatistical increasing $2^{-\Delta\Delta Ct}$ means of IL-32 and caspase-1 in mild to moderate (17.1 and 13) than in severe (15 and 12.7) and critical (13.3 and 11.6) patients, respectively. The findings showed the CCL-2 levels were significantly correlated with the increase of IL-32 levels ($P < 0.05$) and a significant decrease in the P53 level with caspase-1 increased expression ($P < 0.01$). **Conclusion:** It is concluded that gene expression of IL-32 was a novel regulated caspase-1 increase expression in COVID-19 patients, especially CRP-positive patients. Moreover, the gene expression of IL-32 and caspase-1 as a biomarker may have a role in CCL-2 and P53 regulation to reduce COVID-19 risk.

Keywords: Caspase-1, CCL-2, c-reactive protein, IL-32, P53

Introduction

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is the cause of novel coronavirus disease (COVID-19). In the last two years, SARS-CoV-2 has infected millions of people worldwide in different waves, resulting in the deaths of many people. The evidence showed that host immune responses to SARS-CoV-2 play a pivotal role in the pathogenesis and clinical manifestations of COVID-19.

IL-32 is a key modulator in the pathogenesis of various clinical conditions and is mostly induced by IL-8. IL-32 modulates key inflammatory pathways (including TNF-α, IL-6, and IL-1β) and contributes to the pathogenesis of inflammatory diseases. For example, IL-32γ has antiviral effects against influenza A virus (IAV), human immunodeficiency virus (HIV), herpes simplex virus 2 (HSV-2), and vesicular stomatitis virus (VSV).[1-4]

Caspase-1 often contains highly conserved protein domains, such as caspase-associated recruitment domains (CARDS) and death effector domains (DEDs). Caspase-1 has been functionally classified according to its involvement in apoptosis or inflammation. Apoptosis is an immunologically silent and coordinated nonlytic process for dismantling and removing damaged, infected,

Access this article online

Quick Response Code:

Website: https://journals.lww.com/mjby

DOI: 10.4103/MJBL.MJBL_976_23

Address for correspondence: Dr. Rand J. Sattar, Department of Biotechnology, Department of Biology, College of Science, University of Baghdad, Baghdad 00964, Iraq.
E-mail: randjabbar363@gmail.com

Submission: 12-Jul-2023 **Accepted:** 16-Dec-2023 **Published:** 30-Sep-2025

How to cite this article: Sattar RJ, Fadhil HY. IL-32 is a novel regulator for increasing caspase-1 gene expression as a proinflammatory in early SARS-CoV-2 infection. Med J Babylon 2025;22:752-9.

and aging cells. Host cellular apoptosis is thought to be a common viral infection response mechanism for restricting viral expansion A panel of cytokines emerged as predictive of disease severity. The IL-8 – IL-32 axis could be involved in the pathogenesis of COVID-19 and may be associated with acute lung damage. In addition to inducing antiviral immune responses, SARS-CoV-2 can also cause dysregulated inflammatory responses characterized by the noticeable release of proinflammatory mediators in COVID-19 patients.

Among these proinflammatory mediators, chemokines are considered a subset of cytokines that are involved in the chemotaxis process to recruit immune and nonimmune cells to the site of inflammation and infection. Researchers have shown that monocyte chemoattractant protein-1 (MCP-1/CCL-2) and its receptor (CCR2) are involved in the recruitment of monocytes and the infiltration of these cells into the lungs of patients with COVID-19. CCL-2 belongs to the group of CC chemokines and is also known as monocyte chemoattractant protein-1 (MCP-1) due to its involvement in monocyte recruitment. It can bind to the CC chemokine receptor type 2 (CCR2, CD192), triggering various downstream signaling pathways.[5-8] Furthermore, studies suggest that hyperinflammation is associated with the severity of COVID-19 and is characterized by upregulation of inflammatory cytokines and chemokines such as IL-6, IL-10, tumor necrosis factor-alpha (TNF-α), IP-10, CCL-2, and CCL-3, leading to CRS. The effects of these cytokines on disease severity and the mechanisms involved in their role in disease development remain to be identified.

The aim of the study focused on determining whether levels of caspase-1 and IL-32 gene expression are associated with the severity and progression of COVID-19 and its correlation with other clinical and biological markers of infection.

Material and Methods

Study participants

The Ethics Committee of the Iraqi Ministry of Health and Environment approved the study. and all participants gave written informed consent. This study included 100 nasal swabs and blood samples with 58 healthy controls. Patients were referred to Al-Shifa Medical Center at Baghdad Teaching Hospital/Medical City during the period from February 28 to April 30, 2022. The nasal swabs were collected in clean, labeled, and screw-capped tubes (that contain viral transport media (VTM) for maintaining viral viability during transportation to a laboratory for diagnosis) from patients who suffered from upper and/or lower respiratory tract symptoms 10 days after the onset of symptoms. The specimens (swab and serum after separation) were stored at −70°C until the time of ELISA test. Patients and controls were stratified by age group, gender (male and female), and severity (mild, moderate, and severe). In addition, the healthy control sample included age- and gender-matched volunteers who donated blood and worked in healthcare services. They had no neurological or autoimmune disorders.

Blood sample collection

Five milliliters of blood were collected from each participant in an EDTA and gel tube. The tubes were left at room temperature (20–25°C) for 30 min and then centrifuged (the gel tube) for 15 min at 4°C to separate serum. The serum is divided into two parts and frozen at −20°C until assessment of the required test immunoassay of CCL-2 and P53. While the EDTA tube was added, triazole was used for RNA extraction.

Real-time quantitative PCR for gene expression

RNA was extracted from EDTA blood using QIA amp Viral RNA Mini Kit (Qiagen, Germany) and the manufacturer's instructions were followed. The complementary DNA (cDNA) synthesis was performed using the protocol within EasyScript® One-Step gDNA-deleted and complementary DNA generation Super-Mix (Transgen, China). RT-PCR tubes containing RNA (3–5 µL), 1 µL anchored oligo (dT) primer (0.5µg/µL) with 1 µL random primer (0.1µg/µL) and then incubated in a thermocycler at 65°C for 5 min and 4°C for 5 min. After incubation, it was added to 10 µL EX reaction mixture; 1 µL gDNA remover, 1 µL Easy Script®RT/RI enzyme mix and 3 µL RNase-free water to complete a final volume of 20 µL. Incubation in a thermocycler at 25°C for 10 min; followed by 42°C for 30 min and 85°C for 5 s. Amplification of cDNA was performed with specific primers and TransStart® Green qPCRSuper Mix Kit (Transgene, China) (cat no. ID52904). The source of all primers used in this study was Macrogen® (Korea). The *IL-32* gene (Forward-5-TGGCGGCTTATTATGAGGAGC-3 and Reverse-5-CTCGGCACCGTAATCCATCTC-3),[9] *caspase-1* gene (Forward-5-TTTCCGCAAGGTTCGATTTTCA-3 and Reverse-5-GGCATCTGCGCTCTACCATC-3),[10] and β-actin (Housekeeping genes) (Forward-5-CTCCATCCTGGCCTCGCTGT-3 and Reverse-5-GCTGTCACCTTCACCGTTCC-3).[11] The reaction containing master mix SybrGreen (10 µL), 1 µL of each primer, 3 µL of cDNA, and 5 µL of nuclease-free water. Initial denaturation of amplification included: 94°C temperature for 30 min, followed by 40 cycles for 94°C (5 s), 58°C (15 s), and 72°C (20 s). Finally, the dissociation included: temperature 55–95°C temperature, 1-min time for one cycle.

Immunoassay of CCL-2 and P53

CCL-2 and P53 were determined in serum using an enzyme-linked immunosorbent assay (ELISA) kit manufactured by SunLong Biotech, China (catalog no. SL3350Hu)

following the manufacturer's instructions. The detection range of the P53 assay was 0.5–200 ng/mL with a sensitivity of 0.32 ng/mL while the detection range of the CCl-2 assay was 5–1800 ng/L with a sensitivity of 3.09 ng/L.

Statistical analysis

The continuous variable was given as mean or median and standard deviation or interquartile range, and significant differences were assessed using a Mann–Whitney *U* test. Categorical variables were given as numbers and percentages, and significant differences were assessed using Fisher exact test or Pearson Chi-square test. Logistic regression analysis was applied to calculate odds ratio (OR) and 95% CI; in this analysis, patients were distributed as low- and high-production groups according to a median of biomarkers studied (≤ and > mean, respectively), and the control group was the reference category. Spearman's rank-order correlation was performed to analyze the correlation between biomarkers in COVID-19 and healthy control. A probability (*P*) value ≤ 0.05 was taken as statistically significant. The statistical package IBM SPSS Statistics 25.0 (IBM Corp., Armonk, NY) and GraphPad Prism version 8.0.0 (San Diego, CA) were used to perform these analyses.

Results

Levels of CCL-2 and P53

This study finding showed that CCl-2 and P53 levels had highly significant differences in patients (185 ng/L and 24.5 ng/mL) than control (229 ng/L and 30 ng/mL) (P = 0.001), meaning that the infection was down-regulating to induce inflammation with the highest viral load lower than their levels in healthy individuals. More cases of mild–moderate were observed with low levels of CCL-2 and P53, while these were reported in severe and critical with high levels. Significant variation was shown in severe and critical with CCL-2 (OR = 3.45 and 3.23; P = 0.008 and 0.019, respectively), but no significant differences between stratum with p53. Thus, CCL-2 is an effecter factor for disease progression.

Moreover, the results obtained in the current study revealed that there was a nonsignificant difference in the amount of CCL-2 between patients and the control groups, the median CCL-2 levels for patients aged ≤45 and >45 are 181 ng/mL (interquartile range IQR: 157.1–261.6) and 185 ng/mL (IQR: 157 – 236.7), respectively. For healthy controls, the median levels are 278.5 ng/mL (IQR: 210.8 – 350.5) for those ≤45 and 224.8 ng/mL (IQR: 196.9–309.4) for those >45. In the case of gender, results revealed a correlation between the male and female levels of CCL-2 183 ng/mL (IQR: 155.6–238) and 228.8 ng/mL (IQR: 185–312.4), respectively. This indicates that the levels of CCL-2 in female patients were higher than in males, but there are no significant differences in CCL-2 levels between males and females. Furthermore, for severity, there are three categories mild–moderate, severe, and critical. The median CCL-2 levels for patients with mild–moderate cases was 173 ng/mL (IQR: 157.4–236.9), while for severe and critical patients, the median levels were 177 ng/mL (IQR: 157.1–197) and 196.9 ng/mL (IQR: 160.1–273.5), respectively, and there are no significant differences in CCL-2 levels between the severity groups. Regarding, the p53 levels in patients aged 45 and younger were 22.8 ng/mL (interquartile range (IQR) 12.2–27.1) while in patients aged over 45, they were slightly higher at 24.6 ng/mL (IQR 9.8–27.4); however, the P value was not significant (P = 0.8). Among males, the median level of p53 rise to 25.7 ng/mL (IQR 17–28.9) compared to females who had a median level of 22.3 ng/mL (IQR 16.4–26.7); but, this increase was not significant (P = 0.23). In terms of disease severity, the median levels of p53 were lower in mild–moderate and severe, meanwhile revealing not significant (P = 0.248) rise in critical cases 22.3 (IQR7.2–27.1), 22.8 (IQR 17.6–26.8), and 26.7 (IQR 19.3–28.9) ng/mL, respectively.

Expression of IL-32 gene in qRT-PCR

The folding expression ($2^{-\Delta\Delta Ct}$) of *IL-32 and CASP1*mRNA was increased by 16.6 and 12.5 folds in total COVID-19 patients, respectively, but such increase was more significant in *IL-32* expression than *CASP1* ($P < 0.001$ vs. $P < 0.05$) upregulated the expression [Figure 1]. That is why we find the ratio of relative *IL-32* and *CASP1* gene expression in COVID-19 patients versus healthy control was 1.5 and 1.2, respectively.

Although the $2^{-\Delta\Delta Ct}$ means of gene expression showed no significant variation between the age group variation (≤45 and >45 years), sex, and severity in patients and controls, the results showed nonstatistical increasing $2-^{\Delta\Delta Ct}$ means of caspase-1 in male (13.5) than female (11.9) patients, also the *IL-32* and *caspase-1* expression increased in mild–moderate (17.1 and 13) than in severe (15 and 12.7) and critical (13.3 and 11.6) patients, respectively [Table 1].

Multinomial logistic regression of IL-32 and caspase-1 in COVID-19 patients

A multinomial logistic regression model in which the dependent variable contains three or more potential values. Table 2 shows that IL-32 and caspase-1 were protective factors and significantly increased severity when their values increased during infection (OR = 1.57 and 1.05; 95% CI = 1.18−1.98 and 1.0–1.98; respectively). Furthermore, there is no significant effect of age and sex on expression levels ($P > 0.05$).

Correlation between *IL-32* and *caspase-1* gene expression along with CCL-2, and P53 levels

Figure 2 explains Spearman's rank correlation coefficient (rs) between *IL-32* and *CCL-2* expression in patients. The findings showed the CCL-2 levels were significantly

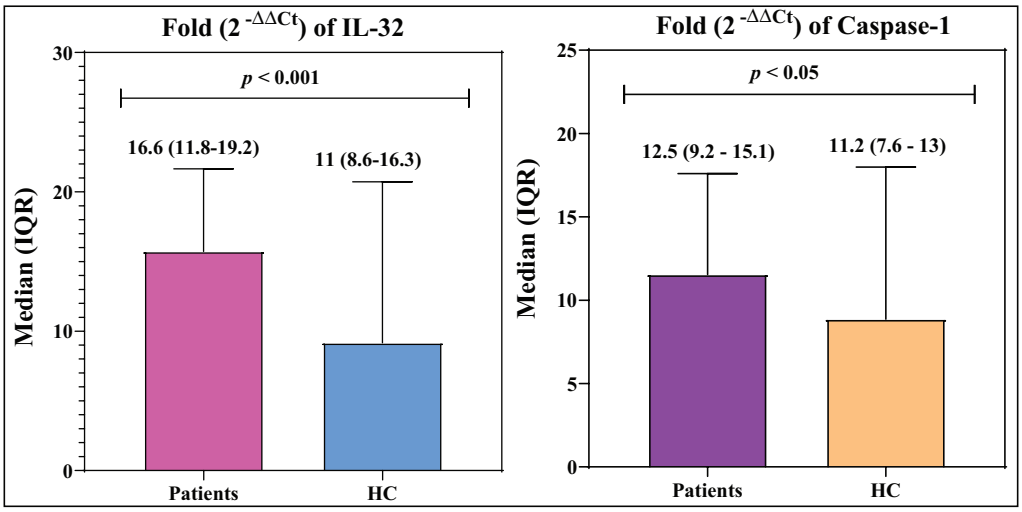

Figure 1: Fold rate of IL-32 and caspase-1 in COVID-19 patient and healthy control

Table 1: Median levels of IL-32 and caspase-1 fold stratified according to characteristics of COVID-19 patients and healthy controls

Character	Fold ($2^{-\Delta\Delta Ct}$) IL-32 median (IQR)		Fold ($2^{-\Delta\Delta Ct}$) caspase-1 median (IQR)	
	Patient (n = 100)	Control (n = 58)	Patient (n = 100)	Control (n = 58)
Age group				
≤45	15.9 (10.4–18.9)	11.2 (9.6–16.9)	12.95 (9.1–15.3)	11.4 (8.4–12.3)
>45	14.4 (9.8–18.7)	10.6 (7.3–16.6)	12.05 (9.6–14.9)	10.9 (7.6–13.9)
P value	P = 0.448	P = 0.367	P = 0.707	P = 0.599
Sex				
Male	15.4 (9.2–17.8)	10.2(14.2–18.6)	13.5 (9.7–15.4)	11.6 (7.5–15)
Female	15.5 (10.7–18.9)	11.1 (9.5–14.8)	11.9 (7.7–14.7)	11.1(10.1–11.6)
P value	P = 0.455	P = 0.521	P = 0.178	P = 0.501
Severity				
Mild–moderate	17.1 (10.1–19.1)	**NA**	13 (8.9–15.3)	**NA**
Severe	15 (10–18.6)		12.7 (9.7–15.4)	
Critical	13.3 (9–16.6)		11.6 (8.3–14.6)	
P value	P = 0.244		P = 0.671	

IQR: interquartile range, NA: not applicable, p: Kruskal–Wallis test and Mann–Whitney U test probability

Table 2: Logistic regression analysis of IL-32 and caspase-1fold in CIOVID-19 patient versus HC

Analysis model	OR (95%CI); IL-32	P value	OR (95% CI); Caspase-1	P value
I (unadjusted)	1.57 (1.18–1.98)	**0.004**	1.05 (1.0–1.98)	**0.041**
II (age-adjusted)	1.03 (0.99–0.08)	0.118	0.91 (0.81–1.01)	0.071
III (age and sex-adjusted)	1.13 (0.57–2.24)	0.736	1.3 (0.29–6.01)	0.726

The reference category is the control group; OR, Odds ratio, CI, confidence interval, p: probability (significant P value is indicated in bold)

correlated with the increase of IL-32 levels ($P < 0.05$) making that prognostic indicator for reducing the development of infection but, when caspase-1 expression increased, the CCL-2 level slightly nonsignificant increased ($P = 0.117$). On the other hand, found conversely no significant relationship between the IL-32 expression and the P53 level ($P = 0.69$) but, there is a significant decrease in the P53 level with caspase-1 increased expression ($P < 0.01$). The results observed that IL-32 is a novel regulator for significantly correlated and increasing caspase-1 gene expression ($P < 0.01$).

Discussion

This monocentric study examined blood parameters and cytokine levels of a population of patients admitted to Alshafaa we evaluated serum concentration of IL-32 in a cohort of COVID-19 patients. Overexpression of some

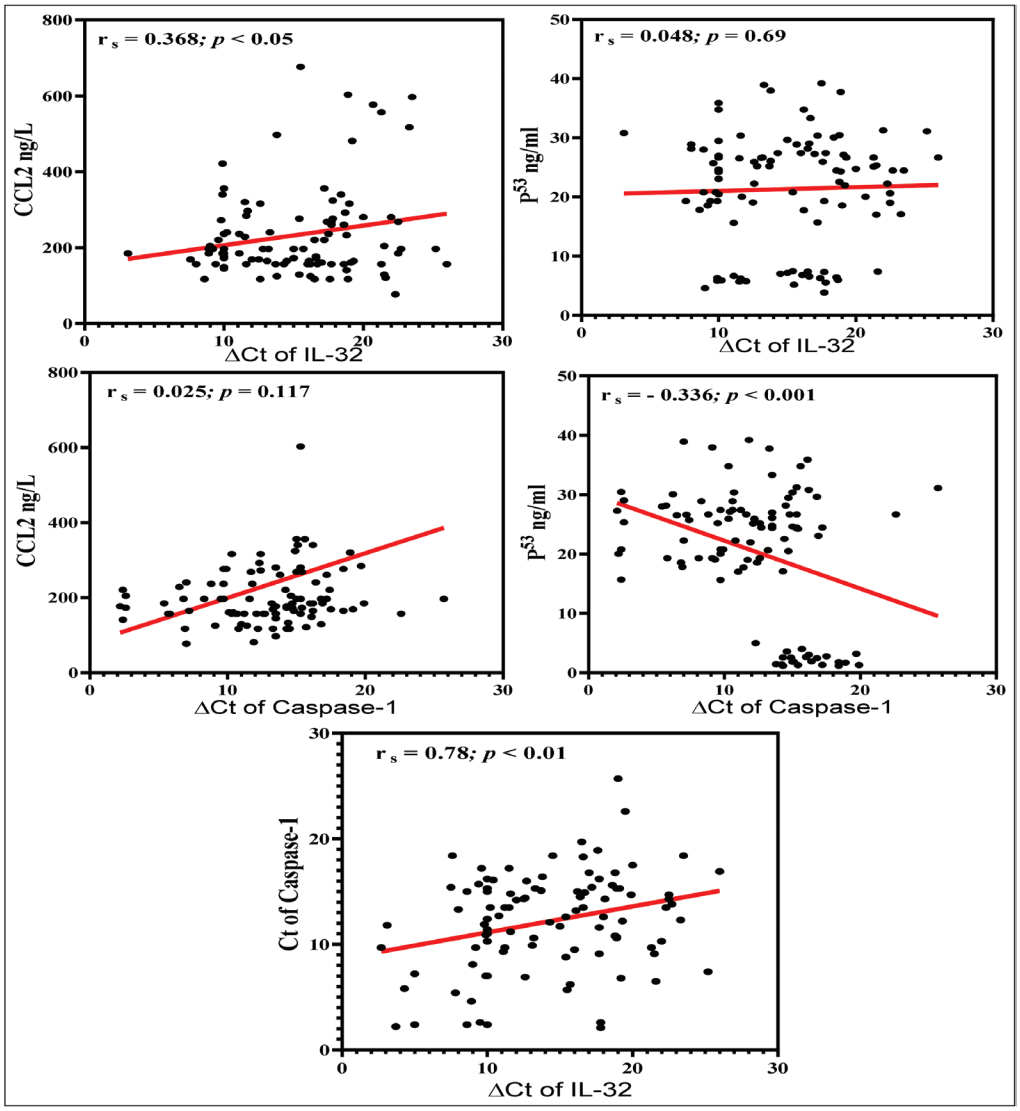

Figure 2: Scatter plot Spearman rank-order correlation coefficient (rs) for analysis between CCL-2 and P53 levels along with *IL-32* and *caspase-1* gene expression in COVID-19 patients

isoforms of IL-32 in our results, back to IL-32 plays a role in the downregulation of inflammatory responses.[12] In addition, COVID-19, as an invasive infectious disease, is largely related to defect(s) in immune system functions. There are several studies pointing to an imbalance in the immune system which plays a fundamental role in the pathogenesis, severity, and outcome of COVID-19. The present study was therefore focused on investigating how CCL-2, and P53 levels along with *IL-32* and *caspase-1* gene expression in COVID-19 patients.

Regarding that IL-32 stimulates immune cells to produce proinflammatory cytokines associated with the pathogenesis of COVID-19,[13] upregulation of *IL-32* gene expression was observed in COVID-19 patients compared with healthy, with highly significant. Other results revealed that severe COVID-19 patients had lower levels of IL-32 than non-severe COVID-19 cases, Furthermore, it is speculated that IL-32 along with other cytokines may contribute to the increased occurrence of atherosclerosis observed in COVID-19 patients. This finding could be associated with the antiviral impacts of IL-32, which participate in improving proinflammatory cytokines through activating signaling pathways of MAPK and NF-κβ and thereby control viral infections.

Viral infection stimulates COX-2 expression, followed by the prostaglandin E2 (PGE2) accumulation in the human lung. COX-2 was shown to regulate dsRNA-triggered IL-32 production, which revealed that COX-2 is an upstream regulatory factor of IL-32. IL-32 promoter activity was increased by the overexpression of COX-2.[14]

Activation of caspase-1 resulted in an increased IL-32 and thymic stromal lymphopoietin (TSLP) production.[15,16] Indeed, activated caspase-1 promoted COX-2-dependent inflammatory reactions, which further increased IL-32

production.[17] IL-32, a proinflammatory cytokine with four isoforms, cleavage by PRTN3 propagates cytokine activity and triggers IL-1β, TNF-α, IL-6, and chemokines.[18] ELANE, CTSG, and PRTN3 can cleave pro-IL-1β to bioactive IL-1β.[19] Caspase-1/interleukin-1 converting enzyme (ICE) cleaves proteins like precursors of the inflammatory cytokine's interleukin 1β and interleukin 18 into their mature biologically active forms[20,21] and CTSG regulates Caspase-1. In our study, the correlation between IL-32 and Caspase-1 direct reaction. The present study showed differences among age groups and study groups (recovered COVID-19 and healthy). These differences suggest that age is a major fatality with older people having the highest likelihood of becoming infected with COVID-19.[22] Lower ACE-2 production in the nasal mucosa of kids may well be attributable to poor SARS-CoV-2 vulnerability, resulting in little or no COVID-19 disease in kids.[23] COVID-19 and all-cause fatalities above the age of 40, when practically all COVID-19 deaths happen, can be described and analyzed using an exponential rise with age approach. Furthermore, COVID-19 fatality is more age-dependent than all-cause fatality, and male has a larger fatality compared to females, which is low obvious as they get older.[24] Incidence reports were recently published, confirming an increase in illness prevalence in men over the age of 60.[25] Older individuals (defined as people over 65 years old) have an increased chance of COVID-19-related incidence and fatality than other age groups, hence they have been given priority for COVID-19 immunization.[26] The occurrence of infected elderly people has a higher risk of COVID-19 infection, which can lead to a weakened immune system, chronic illnesses, malnutrition, increased ACE-2 expression, and organ dysfunction.

The percentage of immunocompromised people in a population is linked to the age structure of that population, therefore age appears to be a major risk factor for COVID-19 severity and prognosis. The present study showed that female patients were higher than males and these results mismatched with results[27] that showed a higher prevalence of COVID-19 infection in males (61%) than in females (39%) Despite the fact that men versus women had the same occurrence, men with COVID-19 are more likely to have infections and die, regardless of their age.[28] The male gender is appearing as a significant risk element for serious COVID-19 and much worse consequences. Relation to data from China, males made up 54.3–57.3% of hospital admissions and 61.1% of ICU patients.[29] Males scored 62% within hospital fatalities in Wuhan, related to results.[27] Men account for approximately two-thirds of fatalities, with more serious symptoms and lower health outcomes, according to preliminary reports from Wuhan encompassing more than 40,000 cases.[30] Previous studies showed females' innate immune responses were found to be more acute and powerful than males'. Nonetheless, males have high levels of proinflammatory cytokines such as the IL-6 receptor that is strongly expressed within lung tissues, indicating that males are more sensitive to cytokines storm, which can contribute to COVID-19 worsening.[31] The Korean Society of Infectious Diseases' 2020 study, however, found that women may be more susceptible to COVID-19.

These gender-specific disparities in COVID-19 incidence between studies are presumably the result of a number of biological, social, and economic factors, among others.[32]

In the case of disease severity most patients included in the study were suffering from severe (0.07), critical (0.03), and mild to moderate (0.07) infection, respectively.

In our study, a decrease in the level of P53 indicates induced deficient levels of IFN-I,[33-35] which is likely to lead to uninhibited viral replication and damage to the immune system. Low-level IFN responses may be a means by which coronaviruses evade immunity. The lack of adequate IFN response may be due to the decrease of P53 hydrolyzed by the coronavirus papain-like proteases (PLPs). PLPs are a class of cysteine proteases that inhibit innate immunity by stabilizing the binding of MDM2 and P53 to cause the ubiquitination of P53.[36] The SARS-unique domain (SUD) and papain-like proteases (PLPs) interact with cell E3 ubiquitin ligase and CHY zinc-finger domain-containing 1 (RCHY1) to facilitate their activities. SUD and PLPs target P53 with E3 ubiquitin ligase RCHY1 to degrade P53. The degradation of P53 decreases the level of earthly IFN.[37] In other words, coronaviruses can evade the host's natural immune defenses by degrading P53 through their own proteins. the role of this chemokine is critical in inflammation because following the ligation of CCL-2 to CCR2 and activation of signaling pathways, migration of monocytes initiated that play a protective role in the clearance of pathogen microorganisms.[38-40] In our study, the level of CCL-2 was lower than is according to variants because the time of sample collection was during the coronavirus pandemic Omicron, so the results correspond with the rate of CCL-2-mediated inflammatory responses in the Omicron variant being lower in mild and moderate forms of the disease, with the severity of the disease, these responses can increase at the level of the Delta variant and lead to severe symptoms in COVID-19 patients.[41] Collectively, CCL-2 concentrations can vary significantly depending on SARS-CoV-2 variants, which implies that viral protein mutations affect immune responses at the cellular and molecular levels.

Furthermore, the current study reported a highly significant correlation between caspase-1 and IL-32. Activated caspase-1 promoted COX-2-dependent inflammatory reactions, which further increased IL-32 production. In addition, the correlation between CCL-2 and P53 with IL-32 and caspase-1 of CCL-2 levels was lower, that is, according to variants because the time of

sample collection was during the coronavirus pandemic Omicron led to a decrease in the level of IFN-I that indicate induce deficient levels of P53. On the other hand, the elevated proinflammatory cytokine interleukin (IL)-32 and caspase-1againste immune response during viral infection lead to a decrease in CCL-2 and P53 levels. Increase in immune response leading to fibrosis necrosis of pulmonary cells.[42,43]

Conclusion

Gene expression of IL-32 was a novel regulated caspase-1 increase expression in COVID-19 patients, especially in CRP-positive patients. Moreover, the gene expression of IL-32 and caspase-1 as a biomarker may have a role in CCL-2 and P53 regulation to reduce the risk of COVID-19.

Conflict of interest

All the authors certify that they have no conflict of interest to disclose in relation to the subject matter or materials discussed in the present study.

Financial support and sponsorship

Nil.

References

1. Alhussien TAA, Fadhil HY. Analysis of mutations in conserved and susceptible regions across the whole genome sequencing analysis for SARS-CoV-2 in Iraqi patients. Iraqi J Sci 2023 special issue:56-64.
2. Cho KS, Park SH, Joo SH, Kim S-H, Shin CY. The effects of IL-32 on the inflammatory activation of cultured rat primary astrocytes. Biochem Biophys Res Commun 2010;402:48-53.
3. Kang J-W, Park YS, Lee DH, Kim MS, Bak Y, Ham SY, *et al*. Interaction network mapping among IL-32 isoforms. Biochimie 2014;101:248-51.
4. Ribeiro-Dias F, Saar Gomes R, de Lima Silva LL, Dos Santos JC, Joosten LAB. Interleukin 32: A novel player in the control of infectious diseases. J Leukoc Biol 2017;101:39-52.
5. Vakilian A, *et al*. CCL2/CCR2 signaling pathway in glioblastoma multiforme. Neurochem Int 2017;103:1-7.
6. Moadab F, Khorramdelazad H, Abbasifard M, Khorramdelazad H, Abbasifard M. Role of CCL2/CCR2 axis in the immunopathogenesis of rheumatoid arthritis: Latest evidence and therapeutic approaches. Life Sci 2021;269:119034.
7. Taghavi Y, Hassanshahi G, Kounis NG, Koniari I, Khorramdelazad H. Monocyte chemoattractant protein-1 (MCP-1/CCL2) in diabetic retinopathy: Latest evidence and clinical considerations. J Cell Commun Signaling 2019;13:451-62.
8. Tay MZ, Poh CM, Rénia L, MacAry PA, Ng LFP. The trinity of COVID-19: Immunity, inflammation and intervention. Nat Rev Immunol 2020;20:363-74.
9. Yao Q, Wang B, Jia X, Li Q, Yao W, Zhang J-a. Increased human interleukin-32 expression is related to disease activity of graves' disease. Front Endocrinol 2019;10.
10. Sun Y, Guo Y. Expression of caspase-1 in breast cancer tissues and its effects on cell proliferation, apoptosis and invasion. Oncol Lett 2018;15:6431-5.
11. Huang Y, Qi Y, Ma Y, He R, Ji Y, Sun Z, *et al*. The expression of interleukin-32 is activated by human cytomegalovirus infection and down regulated by Hcmv-MiR-UL112-1. Virol J 2013;10:51.
12. Kim S-H, Han S-Y, Azam T, Yoon D-Y, Dinarello CA. Interleukin-32: A cytokine and inducer of TNFα. Immunity 2005;22:131-42.
13. Bergantini L, d'Alessandro M, Cameli P, Otranto A, Luzzi S, Bianchi F, *et al*. Cytokine profiles in the detection of severe lung involvement in hospitalized patients with COVID-19: The IL-8/IL-32 axis. Cytokine 2022;151:155804.
14. Zhou Y, Zhu Y. Important role of the IL-32 inflammatory network in the host response against viral infection. Viruses 2015;7:3116-29.
15. Jeong H-J, Shin S-Y, Oh H-A, Kim M-H, Cho J-S, Kim H-M. IL-32 up-regulation is associated with inflammatory cytokine production in allergic rhinitis. J Pathol 2011;224:553-63.
16. Jeong H-J, Nam S-Y, Oh H-A, Han N-R, Kim Y-S, Moon P-D, *et al*. Interleukin-32-induced thymic stromal lymphopoietin plays a critical role in macrophage differentiation through the activation of caspase-1 in vitro. Arthritis Res Therapy 2012;14:R259-12.
17. Cunha TM, Talbot J, Pinto LG, Vieira SM, Souza GR, Guerrero AT, *et al*. Caspase-1 is involved in the genesis of inflammatory hypernociception by contributing to peripheral IL-1β maturation. Mol Pain 2010;6:1744-8069.
18. Guma M, *et al*. Caspase 1–independent activation of interleukin-1β in neutrophil- predominant inflammation. Arthritis Rheumat 2009;60:3642-50.
19. Luksch H, Romanowski MJ, Chara O, Tüngler V, Caffarena ER, Heymann MC, *et al*. Naturally occurring genetic variants of human caspase-1 differ considerably in structure and the ability to activate interleukin-1β. Hum Mutat 2013;34:122-31.
20. Srinivasan L, Ahlbrand S, Briken V. Interaction of *Mycobacterium tuberculosis* with host cell death pathways. Cold Spring Harbor Perspect Med 2014;4:a022459.
21. Meduri GU, Headley S, Kohler G, Stentz F, Tolley E, Umberger R, *et al*. Persistent elevation of inflammatory cytokines predicts a poor outcome in ARDS: Plasma IL-1β and IL-6 levels are consistent and efficient predictors of outcome over time. Chest 1995;107:1062-73.
22. Qifang I, *et al*. Epidemiology and Transmission of COVID-19 in Shenzhen China: Analysis of 391 cases and 1,286 of their close contacts. medrxiv 2020. Special issue.
23. Hussein TAA, Fadhil HY. Impact of inflammatory markers, dread diseases and cycle threshold (Ct) Values in COVID-19 progression. Bionatura 2023. Special issue.
24. Bauer P, Brugger J, König F, Posch M. An international comparison of age and sex dependency of COVID-19 deaths in 2020: A descriptive analysis. Sci Rep 2021;11:19143.
25. Robert Koch Institute. *Coronavirus* disease 2019 (COVID-19): Daily situation report of the Robert Koch Institute. 2020.
26. Whiteman A, *et al*. Demographic and social factors associated with COVID-19 vaccination initiation among adults aged ≥ 65 years—United States, December 14, 2020–April 10, 2021. Morb Mortal Wkly Rep 2021;70:725.
27. Zhou F, Yu T, Du R, Fan G, Liu Y, Liu Z, *et al*. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: A retrospective cohort study. Lancet (London, England) 2020;395:1054-62.
28. Jin J-M, *et al*. Higher severity and mortality in male patients with COVID-19 independent of age and susceptibility. 2020
29. Wang D, Hu B, Hu C, Zhu F, Liu X, Zhang J, *et al*. Clinical characteristics of 138 hospitalized patients with 2019 novel coronavirus–infected pneumonia in Wuhan, China. JAMA 2020;323:1061-9.
30. Wu C, Chen X, Cai Y, Xia J'an, Zhou X, Xu S, *et al*. Risk factors associated with acute respiratory distress syndrome and death in patients with coronavirus disease 2019 pneumonia in Wuhan, China. JAMA Internal Med 2020;180:934-43.
31. Gemmati D, Bramanti B, Serino ML, Secchiero P, Zauli G, Tisato V. COVID-19 and individual genetic susceptibility/receptivity: Role of ACE1/ACE2 genes, immunity, inflammation and coagulation might the double X-chromosome in females be protective against SARS-CoV-2 compared to the single X-chromosome in males? Int J Mol Sci 2020;21:3474.

32. Korean Society of Epidemiology, *et al*. Report on the epidemiological features of coronavirus disease 2019 (COVID-19) outbreak in the Republic of Korea from January 19 to March 2, 2020. J Korean Med Sci 2020;35:10.
33. Cheung CY, Poon LLM, Ng IHY, Luk W, Sia S-F, Wu MHS, *et al*. Cytokine responses in severe acute respiratory syndrome coronavirus-infected macrophages in vitro: Possible relevance to pathogenesis. J Virol 2005;79:7819-26.
34. Zhao X-N, You Y, Cui X-M, Gao H-X, Wang G-L, Zhang S-B, *et al*. Single-cell immune profiling reveals distinct immune response in asymptomatic COVID-19 patients. Signal Transduction Targeted Therapy 2021;6:342.
35. Major J, Crotta S, Llorian M, McCabe TM, Gad HH, Priestnall SL, *et al*. Type I and III interferons disrupt lung epithelial repair during recovery from viral infection. Science 2020;369: 712-7.
36. Harford JB, Kim SS, Pirollo KF, Chang EH. TP53 gene therapy as a potential treatment for patients with COVID-19. Viruses 2022;14:739.
37. Ma-Lauer Y, Carbajo-Lozoya J, Hein MY, Müller MA, Deng W, Lei J, *et al*. p53 down-regulates SARS coronavirus replication and is targeted by the SARS-unique domain and PLpro via E3 ubiquitin ligase RCHY1. Proc Natl Acad Sci USA 2016;113: E5192-201.
38. Yoshimura T, Takahashi M. IFN-γ-mediated survival enables human neutrophils to produce MCP-1/CCL2 in response to activation by TLR ligands. J Immunol 2007;179:1942-9.
39. Dessing MC, van der Sluijs KF, Florquin S, van der Poll T. Monocyte chemoattractant protein 1 contributes to an adequate immune response in influenza pneumonia. Clin Immunol 2007;125:328-36.
40. Yoshimura T, Galligan C, Takahashi M, Chen K, Liu M, Tessarollo L, *et al*. Non-myeloid cells are major contributors to innate immune responses via production of monocyte chemoattractant protein-1/ CCL2. Front Immunol 2014;4:482.
41. Korobova ZR, Arsentieva NA, Liubimova NE, Dedkov VG, Gladkikh AS, Sharova AA, *et al*. A Comparative Study of the Plasma Chemokine Profile in COVID-19 Patients Infected with Different SARS-CoV-2 Variants. Int J Mol Sci 2022;23:9058.
42. Jawad DH, Mezher MN. Interleukin-6 and Biomarkers Predict Outcomes in People with Diabetes and COVID-19 Infection. Med J Babylon 2024;21:S33-8.
43. Alfadhel SM, Thamer NA. Inflammatory Cytokines—The Link between Coronavirus Disease and Abortion: A Case-Control Study. Med J Babylon 2024;21:993-8.