Journal of Inflammation Research

Dovepress
open access to scientific and medical research

Open Access Full Text Article

ORIGINAL RESEARCH

Altered Liver Enzyme Markers in Patients with Asymptomatic, and Mild Omicron Infection: A Retrospective Study

Xi Cao[1], Yong-Li Xie[2,3], Jian-Ying Yi[1], Zhi-Li Liu[4], Meng Han[1], Ji-hui Duan[1], Qiang Gao[1], Hong Mu[1], Chun-lei Zhou[1]

[1]Department of Clinical Laboratory, Tianjin First Central Hospital, Tianjin, People's Republic of China; [2]Department of Clinical Laboratory, Tianjin Stomatological Hospital, School of Medicine, Nankai University, Tianjin, People's Republic of China; [3]Tianjin Key Laboratory of Oral and Maxillofacial Function Reconstruction, Tianjin, People's Republic of China; [4]Department of Clinical Laboratory, the Third Central Hospital, Tianjin, People's Republic of China

Correspondence: Hong Mu; Chun-lei Zhou, Department of Clinical Laboratory, Tianjin First Central Hospital, Tianjin, People's Republic of China, Email mutjyzxyy@163.com; tj_zcl@hotmail.com

Purpose: The emergence of the SARS-CoV-2 Omicron variant has posed a significant global public health challenge. Elucidating the laboratory profiles of individuals infected with this variant is crucial for assessing organ damage. This study aimed to investigate the variations in liver function tests and their correlation with demographic characteristics and inflammatory markers in patients with early Omicron variant infections.

Patients and Methods: A retrospective cohort study was conducted on 1133 mild or asymptomatic COVID-19 cases at Tianjin First Central Hospital. Data on age, gender, body mass index (BMI), and serum markers were collected and analyzed. Statistical analyses were performed using SPSS software, version 24.0.

Results: Abnormal liver function parameters, including aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), gamma-glutamyl transferase (GGT), and total bilirubin (TBIL), were observed in 314 (27.71%) patients. "Hepatocellular type" was identified in 56 (4.94%) patients, "cholestatic type" in 185 (16.33%) patients, and "mixed type" in 73 (6.44%) patients. In the mixed group, we observed a pronounced elevation in the levels of ALT, AST, and GGT. Moreover, the hepatocellular group exhibited a statistically significant increase in AST and ALT concentrations relative to both the normal and cholestatic groups. Notably, the cholestatic group demonstrated a substantial increment in ALP levels. Males had a significantly higher prevalence of "abnormal liver enzyme markers" compared to females. Patients with "abnormal liver enzyme markers" exhibited significantly decreased immunoglobulin G (IgG) levels and elevated levels of inflammatory markers, including procalcitonin (PCT), interleukin-6 (IL6), as well as C-reactive protein (CRP) compared to normal group. Logistic regression analysis revealed that male gender and PCT levels were significantly associated with the risk of abnormal liver enzyme markers. Patients in hepatocellular group were likely accompanied with high CRP levels, whereas those in the cholestatic type were associated with high IL6 levels.

Conclusion: Early Omicron infection might cause liver stress response. Elevated liver enzyme marker levels were correlated with age, gender, inflammatory factors, and IgG.

Keywords: Omicron variant, "abnormal liver enzyme markers", male, inflammatory markers

Introduction

The coronavirus disease 2019 (COVID-19), initially identified in Wuhan, China, had engulfed the globe in a pandemic. Most COVID-19 patients present with typical respiratory symptoms. However, liver impairments are often developed in patients with COVID-19, liver damage ranges from mild asymptomatic elevation of liver enzymes to severe liver injury. With the rapid mutation of viral strains, the Omicron (B.1.1.529) replaced the Delta (B.1.617.2) globally and became virtually the predominant strain in most countries in December 2021.[1] On January 8, 2022, the first local case of Omicron

Received: 17 May 2024
Accepted: 27 September 2024
Published: 30 September 2024

(B.1.1.529) infection on the Chinese mainland was reported in Tianjin after which Omicron (B.1.1.529) swiftly spread throughout the country.

According to guidelines provided by the National Health Commission of China, patients with moderate and severe COVID-19 typically exhibit more serious respiratory symptoms or dyspnea, and chest imaging reveals characteristic changes indicative of pneumonia. Mild patients primarily exhibit clinical symptoms such as fever, slight fatigue, and disorders of smell and taste. In CT scans of the lungs, there are no characteristic interstitial changes typically seen in COVID-19 infections. In previous studies, hepatic impairment had been characterized by the presence of elevated liver enzyme levels. An elevation in AST and/or ALT was a prevalent indicator observed in patients with moderate to severe COVID-19. Conversely, an increase in GGT and/or ALP constituted a less frequent phenomenon, typically noted in the later phases of the illness.[2] The Omicron variant of SARS-CoV-2 had accumulated a higher number of mutations in its spike protein compared to other strains. This enhanced transmissibility, infectivity, and immune escape capacity, making it easier for patients to experience repeated infections.[3] Nevertheless, the pathogenicity of the infection appeared to be attenuated, with the majority of infected individuals exhibiting asymptomatic or mild clinical presentations. As the novel coronavirus coexisted with humans for a long time at present, the state of liver function in asymptomatic or mild cases remains an area of inquiry, and it remains to be elucidated whether such individuals are susceptible to hepatic injury or the development of hepatitis.

In the early stages of our study, we found that patients infected with the Omicron variant presented with lower cycle threshold (Ct) values of nucleic acid tests in nasopharyngeal swab specimens, indicating a higher viral load. High concentrations of the virus may elicit an immune protective response, leading to the release of a large number of inflammatory factors like CRP, IL-6 and so on.[4] Moreover, there was evidence of direct viral tropism for ACE2 receptors in both hepatocytes and cholangiocytes.[5,6] This direct COVID-19 viral targeting of liver cells could contribute to the observed liver dysfunction and injury in patients, especially considering the importance of the ACE2 receptor in the pathogenesis of the disease.

To date, the majority of current investigations focus on liver status in patients with moderate to severe COVID-19, many of whom have pre-existing chronic conditions or require long-term medication, suggesting that liver injury in these cases may arise from multifactorial causes. However, few studies have comprehensively analyzed liver enzymes and their correlation with inflammatory markers and demographic features in patients with early-stage Omicron infection. Serum liver enzyme markers (AST, ALT, GGT, ALP, and TBIL) and liver functional markers (APTT, PT, TT, DD, and FIB) offer considerable clinical utility for rapid and precise assessment of the patient's liver status. In order to fill this gap in relevant knowledge, we conducted a retrospective study that evaluated serum liver enzyme markers adjusted for demographic characteristics and other relevant markers, in a cohort of asymptomatic or mild patients with Omicron infection.

Materials and Methods

Participants and Data Source

The study was conducted at Tianjin First Central Hospital in Tianjin, China, from March 2022 to May 2023. The inclusion criteria were as follows: (1) patients with initial diagnosis of Omicron variant infection, (2) asymptomatic or mild COVID-19 cases as confirmed, (3) requirement for nasopharyngeal swab testing at least twice during hospitalization with a minimum interval of 24 hours, and (4) availability of admission laboratory results for AST, ALT, ALP, GGT, and TBIL. Exclusion criteria included: (1) patients with hepatic and/or renal impairment, (2) patients suffering from severe inflammatory conditions such as chronic pulmonary disease, gastrointestinal disorders, immunological diseases, etc., (3) patients presenting with cerebral infarction or cardiovascular diseases at admission, and (4) patients with incomplete data. In recognition of the fact that apart from liver disorders, various other pre-existing conditions—such as cardiopulmonary diseases, immunological disorders, gastrointestinal pathologies, among others—exert a considerable influence on inflammatory mediators, our study cohort was meticulously selected to include only patients infected with the Omicron variant who were devoid of underlying diseases. A total of 1133 COVID-19 patients were enrolled, consisting of 572 asymptomatic and 561 mild cases. Data collection involved recording clinical symptoms at admission, laboratory

findings, and nucleic acid test results from the COVID-19 rehabilitation ward at Tianjin First Central Hospital. The diagnosis of asymptomatic or mild cases followed guidelines provided by the National Health Commission of China. Asymptomatic patients were characterized by the absence of respiratory symptoms and pulmonary pathological changes, with only a positive nucleic acid test result. Mild patients primarily exhibit clinical symptoms such as fever, slight fatigue, and disorders of smell and taste. In chest computed tomography (CT) scans of the lungs, there are no characteristic interstitial changes typically seen in COVID-19 infections. The diagnosis and grouping of patients depended on clinical infectious disease doctors. Demographic data, laboratory findings, and CT results were procured from electronic medical records for all inpatients and reviewed by a team of specialized physicians. None of the patients showed radiological signs of pneumonia. We had reviewed the electronic medical records of all enrolled patients and confirmed that upon admission, none had a history of long-term medication use and alcohol abuse. The reference ranges for serum "liver enzyme markers" were defined as follows: ALT: 0–50 U/L; AST: 17–59 U/L; ALP: 38–126 U/L; GGT: 12–58 U/L; TBIL: 3–22 μmol/L. "Abnormal liver enzyme markers" was classified as: ALT > 50 U/L; AST > 59 U/L; ALP > 126 U/L; GGT > 58 U/L; TBIL > 22 μmol/L. As COVID-19 is a novel infectious disease, classifications of liver injury are not yet fully delineated. Consequently, this study categorized liver injury into hepatocellular, cholestatic, or mixed types. "Hepatocellular group" was identified by an elevation in ALT and/or AST above the upper limit of normal (ULN). "Cholestatic group" was indicated by increased ALP, GGT, and/or TBIL beyond ULN. "Mixed group" was diagnosed when patients exhibited both hepatocellular (ALT/AST above ULN) and cholestatic (ALP, GGT, and/or TBIL above ULN) characteristics. The BMI was calculated using the formula: Weight (kg)/Height² (m²).

Serum immunoglobulin G (IgG) and IgM antibodies against SARS-CoV-2 proteins were measured utilizing magnetic particle chemiluminescence assays (Biology and Science, China). C-reactive protein (CRP) levels were determined via latex-enhanced immunoturbidimetric assays (Mindray, China). Dry chemical colorimetric methods (VITROS, America) were employed for the evaluation of AST, ALT, ALP, GGT, and TBIL. PCT and IL6 were quantitatively assessed using electrochemiluminescence immunoassays (Ren Mai, China). Activated partial thromboplastin time (APTT), thrombin time (TT), prothrombin time (PT), fibrinogen (FIB), and D-dimer (DD) were quantitatively assessed using Automated Coagulation Analyzer (CP3000, Japan). To reduce the likelihood of false-negative results, nasopharyngeal swabs were collected daily from all patients. Each patient's specimen was tested with commercial SARS-CoV-2 kits from two separate manufacturers (Sheng Xiang/Bo Jie, China).

In accordance with the China Technical Guidelines for Laboratory Testing for COVID-19, a Ct value of 40 or below was deemed positive for SARS-CoV-2. The date of diagnosis was established as the day on which the initial specimen yielded a positive result for SARS-CoV-2 via reverse transcription polymerase chain reaction (RT-PCR). Discharge criteria were based on World Health Organization (WHO) recommendations, which stipulate two consecutive negative PCR results from nasopharyngeal swabs, collected with a minimum interval of 24 hours, in the absence of clinical symptoms.

Statistical Analysis

Statistical analysis was conducted using SPSS 24.0 for Windows software (SPSS, Chicago, IL). Normally distributed data were presented as the mean ± standard deviation. Between-group comparisons of variables with a normal distribution and homogeneous variances were performed using the *t*-test. Non-normally distributed data were expressed as the median (upper quartile and lower quartile), and the non-parametric Mann–Whitney *U*-test was used for between-group comparisons of non-normally distributed or heterogeneously variant data. Categorical characteristics were described in counts and percentages (%), with the chi-squared test used for categorical variables. Risk factors were analyzed through univariate logistic regression, providing the P value and odds ratio (OR), accompanied by its 95% confidence interval (CI). Significance was determined at $P < 0.05$, and a 95% CI for OR; an $OR > 1$ indicated an increased risk of the event, while an $OR < 1$ indicated a decreased risk. A P value of < 0.05 was considered statistically significant, and $P < 0.001$ was considered highly statistically significant.

Results

In this retrospective analysis, 1133 COVID-19 patients, comprising 572 asymptomatic and 561 mild cases, were enrolled to assess their liver enzyme levels, including ALT, AST, ALP, GGT, and TBIL. The male gender accounted for

Table 1 Laboratory Characteristics of COVID-19 Patients (mean ± SD)

Variables	Asymptomatic Patients	Mild Patients	P value
Cases, n	572	561	/
Male, n (%)	294(51.40%)	269(47.95%)	0.135
Age (yr)	33.76±0.71	33.60±0.65	0.237
High (cm)	166.19±0.57	167.12±0.49	0.218
Weight (kg)	67.56±0.80	66.47±0.69	0.362
ALT (U/L)	27.46±1.53	26.07±1.05	0.236
AST (U/L)	27.93±1.10	25.62±0.55	0.201
GGT (U/L)	27.69±1.36	28.32±1.29	0.916
ALP (U/L)	90.59±2.71	88.35±1.93	0.116
TBIL (umol/L)	11.17±0.29	11.57±0.29	0.854
IgG (g/L)	19.90±2.11	26.67±2.42	**0.023**
IgM (g/L)	0.62±0.21	0.47±0.08	0.457
PCT (ng/mL)	0.063±0.006	0.055±0.003	0.353
IL6 (U/mL)	11.48±0.38	11.94±0.44	0.452
CRP (mg/L)	6.33±0.39	7.26±0.41	0.072
APTT(s)	33.36±0.21	33.66±0.24	0.243
TT (s)	18.28±0.058	18.26±0.040	0.731
PT (s)	13.15±0.081	13.62±0.17	0.052
PT (%)	93.15±0.93	90.60±0.89	0.066
FIB (g/L)	2.75±0.030	2.79±0.030	0.265
DD (mg/L FEU)	0.41±0.040	0.44±0.030	0.515

Notes: ALT: 0–50 U/L, AST: 17–59 U/L, ALP: 38–126 U/L, GGT: 12–58 U/L, TBIL: 3–22 umol/L, PCT: 0–0.046 ng/mL, IL6: 0–7 pg/mL, CRP: 0–10 mg/L, APTT: 24–39s, TT: 14–21s, PT: 11–15s, PT (%): 80–120, FIB: 2–4 g/L DD: 0.03–0.55 mg/L FEU. A p-value below 0.05 was considered statistically significant. Statistically significant results are bold.

approximately 51.40% of the asymptomatic group and 47.95% of the mild group, with mean ages of 33.76 ± 0.71 and 33.60 ± 0.65 years, respectively (Table 1). Demographic and clinical characteristics were presented in Table 1, highlighting a significant discrepancy in IgG levels between asymptomatic and mild patients, with the latter demonstrating elevated levels (19.90 ± 2.11 vs 26.67 ± 2.42, $P < 0.05$).

Among the 314 patients exhibiting "elevated liver enzyme levels, hepatocellular type", characterized by ALT and/or AST levels exceeding the ULN, was observed in 56 cases (17.83%). In contrast, "cholestatic type", delineated by increased levels of ALP, GGT, and/or TBIL beyond ULN, was detected in 185 patients (58.92%), while a "mixed type", featuring both hepatocellular and cholestatic features, was identified in 73 patients (23.25%) (Table 2). In the hepatocellular group, elevations of AST (>2× ULN) and ALT (>2× ULN) were recorded in 1 (1.79%) and 6 (10.71%) patients, respectively (Table 3). Within the cholestatic group, elevated levels of GGT (>2× ULN), ALP (>2× ULN), and TBIL (>2× ULN) were observed in 4 (2.16%), 31 (16.77%), and 3 (1.62%) patients, respectively (Table 3). The mixed group exhibited a higher prevalence of marked elevations (>2× ULN) in AST (13.70%), ALT (28.77%), and GGT (23.29%), whereas no cases presented with elevated ALP or TBIL levels at this threshold (0.00% for both) (Table 3). We conducted

Table 2 Comparison of Demographic Parameters and Serum Markers for Normal, Hepatocyte, Cholestatic, and Mixed Groups

Variables	No liver Damage	Hepatocyte	Cholestasis	Mixed	P value
Cases	819	56	185	73	/
Male	331 (40.42%)$^{\#\Delta\&}$	39(69.64%)	135(72.97%)	58(79.45%)	**<0.001**
Age	35.33±0.51	36.19±1.86	24.62±1.47$^{*\#\&}$	34.88±1.51	**<0.001**
BMI	23.82±0.15$^{\#\&}$	26.25±0.55	22.44±0.51$^{\#\&}$	26.60±0.58	**<0.001**
IgG	26.64±2.04$^{\#\Delta\&}$	20.55±6.29	16.57±3.40	13.95±4.58	**0.046**
IgM	0.63±1.49	0.36±0.20	0.32±0.10	0.25±0.06	0.632
PCT	0.052±0.004$^{\#\Delta\&}$	0.071±0.007	0.077±0.008	0.078±0.007	**<0.001**
IL6	10.38±0.33	11.37±1.06	13.47±0.79$^{*\#}$	14.26±1.08$^{*\#}$	**0.005**
CRP	5.76±0.34$^{\#\&}$	9.00±1.17	7.93±0.66	9.37±0.95	0.360
APTT	33.48±0.18	34.16±0.70	33.99±0.44	32.20±0.55	0.143
TT	18.26±0.04	18.11±0.13	18.43±0.07	18.23±0.11	0.319
PT	13.35±0.11	13.34±0.16	13.01±0.28	12.66±0.18	0.204
PT (%)	91.01±0.72	91.15±2.39	95.10±1.80	96.18±2.56	0.578
FIB	2.78±0.02	2.72±0.09	2.08±0.05	2.80±0.09	0.346
DD	0.42±0.03	0.40±0.06	0.47±0.07	0.29±0.03	0.738

Notes: Compare with No liver damage:*P-values<0.05; Compare with Hepatocyte damage:$^{\#}$ P-values<0.05; Compare with Cholestasis damage:$^{\Delta}$P-values<0.05; Compare with Mixed damage:$^{\&}$P-values<0.05. A p-value below 0.05 was considered statistically significant. Statistically significant results are bold.

Table 3 Liver Enzymes Distribution Range in Patients with Hepatocyte, Cholestatic, and Mixed Type

Variables	Hepatocyte	Cholestasis	Mixed
ALT (50–100 U/L)	50 (89.29%)	0 (0.00%)	48 (65.75%)
ALT (>100 U/L)	6 (10.71%)	0 (0.00%)	21 (28.77%)
AST (59–118 U/L)	8 (14.29%)	0 (0.00%)	21 (28.77%)
AST (>118 U/L)	1 (1.79%)	0 (0.00%)	10 (13.70%)
GGT (58–116 U/L)	0 (0.00%)	32 (17.30%)	43 (58.90%)
GGT (>116 U/L)	0 (0.00%)	4 (2.16%)	17 (23.29%)
ALP (126–252 U/L)	0 (0.00%)	77 (41.62%)	8 (10.96%)
ALP (>252 U/L)	0 (0.00%)	31 (16.77%)	0 (0.00%)
TBIL (22–44 umol/L)	0 (0.00%)	45 (24.32%)	18 (24.66%)
TBIL (>44 umol/L)	0 (0.00%)	3 (1.62%)	0 (0.00%)

a comparative analysis of the mean concentrations of liver enzymes across the four designated groups. The results revealed that the levels of ALT, AST, and GGT in the mixed group were notably elevated compared to those observed in the other three groups (Figure 1). Furthermore, the hepatocellular group exhibited significantly higher AST and ALT

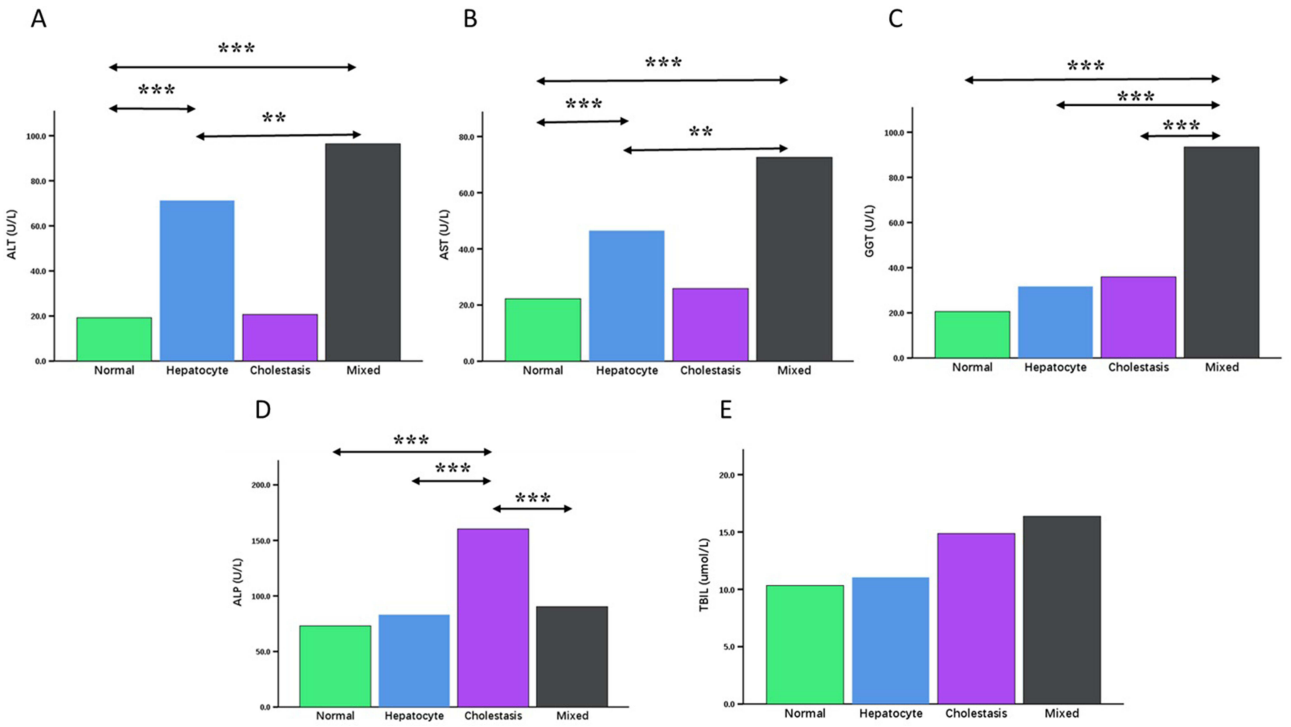

Figure 1 Comparison of the mean levels of liver enzymes in four groups (Normal, Hepatocyte, Cholestasis and Mixed groups). (**A**) ALT, (**B**) AST, (**C**) GGT, (**D**) ALP and (**E**) TBIL. **P-values<0.01; ***P-values<0.001.

levels relative to both the normal and cholestatic groups (Figure 1). In addition, the ALP level in the cholestatic group was significantly greater than that observed in the remaining three groups (Figure 1).

Table 2 illustrated statistically significant differences ($P < 0.05$) in gender distribution, age, BMI, IgG levels, CRP, IL-6, and PCT across the four groups (normal, hepatocellular, cholestatic, and mixed). Pairwise comparisons demonstrated that, compared to the normal group, the male proportion and PCT levels were significantly increased ($P < 0.05$) in the other three groups. Conversely, IgG levels in these three groups were significantly lower ($P < 0.05$) than in the normal group. Within the cholestatic group, ages were significantly lower compared to the hepatocellular and mixed groups. Furthermore, IL-6 levels in the cholestatic and mixed groups were significantly higher than those in the normal and hepatocellular groups. CRP levels in the hepatocellular and mixed groups were significantly elevated compared to the normal group. Notably, there were no significant differences observed in the coagulation parameters, including APTT, TT, PT, FIB, and DD, among the four groups.

Subsequent to the descriptive analyses, logistic regression modeling was conducted to assess the predictive value of various parameters in determining the risk of "abnormal liver enzyme markers". The objective was to delineate the principal contributors to elevated hepatic enzymes. The findings indicated that the female gender was a protective factor (OR = 0.296, 0.251, 0.175, respectively), with statistically significant protective effects ($P < 0.05$). Conversely, PCT emerged as a prominent risk factor across all types (OR = 3.847, 50.817, 7.497, respectively), with each OR achieving statistical significance ($P < 0.05$) (Table 4). Advanced age, however, was inversely associated with the likelihood of "cholestatic type". IL-6 was determined to be a significant risk factor specific to "cholestatic type" (OR = 1.428), whereas the CRP was significantly predictive of "hepatocellular type" (OR = 1.416). In the instance of "mixed type", male gender, elevated BMI, PCT, CRP, and IL-6 all surfaced as independent risk factors (Table 4).

Discussion

In the current study, we found that during the early stages of Omicron infection, a small percentage of individuals with asymptomatic or mild COVID-19 by the Omicron variant had elevated liver enzyme levels. These biomarkers demonstrated a positive correlation with age, male gender, elevated inflammatory factors (PCT, IL-6, and CRP), and a negative

Table 4 Logistic Regression Analysis of Risk Factors for Hepatocyte, Cholestatic, or Mixed Type Comparing with Normal Group

Variables		Regression Coefficient	Wald Value	OR (95% CI)	P value	AUROC
Hepatocyte damage	Gender (female)	−1.219	16.584	0.296 (0.164–0.531)	**<0.001**	0.646
	BMI	0.121	11.587	1.129 (1.053–1.211)	**<0.001**	0.685
	IgG	−0.003	0.754	0.997 (0.990–1.004)	0.385	0.496
	PCT	1.347	2.436	3.847 (0.709–20.882)	**0.049**	0.709
	IL6	0.216	1.104	1.114 (1.033–1.208)	0.144	0.600
	CRP	0.117	1.373	1.416 (1.321–1.614)	**0.032**	0.689
Cholestasis	Gender (female)	−1.381	58.760	0.251 (0.176–0.358)	**<0.001**	0.663
	Age	−0.056	67.515	0.900 (0.844–0.979)	**<0.001**	0.715
	IgG	−0.005	4.438	0.995 (0.991–1.000)	**0.035**	0.467
	PCT	3.928	10.146	50.817 (4.532–569.845)	**<0.001**	0.674
	IL6	0.428	11.782	1.428 (1.310–1.642)	**0.001**	0.682
	CRP	0.122	4.455	1.060 (1.002–1.122)	0.116	0.596
Mixed	Gender (female)	−1.741	34.049	0.175 (0.098–0.315)	**<0.001**	0.695
	BMI	0.144	19.385	1.155 (1.083–1.231)	**<0.001**	0.700
	IgG	−0.007	3.202	0.993 (0.985–1.001)	0.074	0.578
	PCT	2.014	4.331	7.497 (1.125–49.980)	**0.037**	0.741
	IL6	0.528	9.632	1.328 (1.200–1.711)	**0.033**	0.687
	CRP	0.322	5.411	1.267 (1.111–1.401)	**0.041**	0.668

Notes: Logistic regression analysis was adjusted for age, gender, BMI, IgG, PCT, IL-6, and CRP. A p-value below 0.05 was considered statistically significant. Statistically significant results are bold.

correlation with IgG antibody titers. Our data implied that even among asymptomatic or mild Omicron-infected individuals, there may exist a potential risk of subclinical injury in liver. In comparison to the Beta or Delta variants, individuals experiencing initial Omicron variant infection demonstrated higher viral loads and an increased susceptibility to reinfection due to the variant's enhanced ability to evade pre-existing immunity.[7] This suggested that Omicron variant infections might persist for extended durations. Prior research had corroborated these observations, implying that hepatocellular injury may be directly attributed to viral infection.[8] Multiple lines of evidence had indicated that ACE2 receptors, the primary entry points for SARS-CoV-2, are expressed at notably higher levels in bile duct cells (60%).[9,10] The virus binds to ACE2 receptors in bile duct cells via its spike proteins, facilitating direct invasion of target cells and subsequent liver injury.[11] Hence, elevated liver enzymes in COVID-19 might be associated with high SARS-CoV-2 viral loads and prolonged viral shedding during the early stages of Omicron infection. Our findings revealed a pronounced elevation of liver enzymes in males across all three types. This heightened susceptibility in males may be explained by the higher expression of ACE2 receptors, as previously reported in a Wuhan study.[12] Gender differences in COVID-19 severity had been documented, with males more likely to experience severe disease than females.[13] The protective effect of estrogen in females may contribute to their reduced risk of liver injury.[14] Of the 314 patients with elevated liver enzyme levels, the majority exhibited a cholestatic type. In a retrospective study by Ji et al[15] involving 202 COVID-19 patients, the predominant form of liver function impairment was parenchymal type (elevated AST and/or ALT), with only

2.6% exhibiting cholestatic impairment. Also previous clinical investigations suggested that elevations in GGT and ALP were less common and typically occurred in the later stages of COVID-19, whereas moderate increases in AST, ALT, and TBIL were frequent manifestations of the disease.[16,17] Contrarily, our study revealed that increments in GGT, ALP, and TBIL were prevalent among patients with early Omicron variant infection, and the cholestatic group was characterized primarily by elevation in ALP levels. The elevation of these enzymes, which serve as surrogate markers for bile duct injury,[18] indicates that the early Omicron infection may induce subclinical cholangiocyte damage through a multifactorial and intricate pathogenic process, differing from earlier strains of SARS-CoV-2. Our results underscored the variable and complex hepatobiliary manifestations of the Omicron variant, which may be strain-specific.

Upon analyzing the changes in liver enzyme levels and inflammatory markers, we observed that patients with abnormal liver enzyme levels exhibited significantly elevated values of inflammatory factors, including PCT, IL-6, and CRP, across varying degrees ($P < 0.05$). This may be attributed to stress response happening in the liver, characterized by the release of major acute-phase cytokines in response to Omicron infection. PCT levels were significantly increased in all patients with abnormal liver enzyme levels relative to those with normal levels. Patients in the mixed and hepatocellular injury groups showed elevated CRP levels, whereas those in the mixed and cholestatic injury groups had higher IL-6 levels, respectively. CRP is a key pro-inflammatory marker that rises during COVID-19 infection stage, increased CRP levels in asymptomatic or mild patients indicate the acute phase reaction of the immune system.[19] Our study showed that a raised CRP levels in patients who showed elevated ALT or AST, suggesting temporary inflammation of hepatocytes exists. Interestingly, the relative level of IL-6 was higher in the cholestatic and mixed groups. Previous study put forward that a biphasic pattern characterized by initial transaminase elevations followed by cholestatic changes in liver enzymes in COVID-19 infection.[20] Moreover, the time of elevation and half-life of different inflammatory factors (PCT, IL-6, and CRP) vary during inflammation. These may account for the differences in levels of inflammatory factors (PCT, IL-6, and CRP) among patients in different groups.

The increases in PCT, CRP, and IL-6 levels signified "stress response" to the liver as a consequence of the inflammatory response triggered by Omicron infection. These findings suggested that "elevated liver enzyme levels" caused by the Omicron strain were likely associated with the high viral load at the initial stage of infection. Previous studies showed that lower level of IgG in patients who have recovered from COVID-19 may lead to be re-infected.[21] After the outbreak of the pandemic, the vast majority of patients enrolled in the study had received one or more doses of the COVID-19 vaccine. In our study, the average levels of IgG in patients with abnormal liver enzyme indices were lower than that in the normal control group. IgG antibodies may provide protection to organs by neutralizing a portion of the virus, binding to the viral S-protein, and interfering with its interaction with the ACE2 receptor.[22] We speculated that patients with higher IgG antibody titers might clear the virus more rapidly.

Liver injury associated with SARS-CoV-2 infection is likely multifactorial, encompassing mechanisms such as the direct cytopathic effects of the virus, an exaggerated systemic immune response, vascular damage, coagulopathy, and drug-induced hepatotoxicity.[23] In the present investigation, all participants were devoid of underlying diseases and had no history of prolonged medication use "and alcohol abuse". Additionally, there was no appreciable distinction in coagulation markers between the cohort with "elevated liver enzyme levels" and normal group. Our findings showed the "occurrence of slightly elevated liver enzymes" in the early stages of Omicron infection. The observations suggested that the elevated levels of liver enzymes were likely a consequence of an inflammatory response triggered by high viral loads. This inflammation may induce a stress response in the liver, as evidenced by the increased enzyme levels. However, the data imply that despite these alterations, the overall liver function of the patients remains intact. This suggested that the liver was responding to the viral challenge without significant compromise to its functional capacity.

Logistic regression analysis in the current study revealed that inflammatory factors—CRP, IL6, and PCT—were all significantly associated with an increased risk of "elevated liver enzyme levels", with PCT showing the strongest correlation. In a cohort study of 148 patients hospitalized in Shanghai, China, by Fan et al, elevated hepatic enzymes were more prevalent in males and were frequently accompanied by increased levels of CRP and PCT,[24] in alignment with our findings. Omicron infection has been linked to cytokine released, and this proinflammatory state significantly contributes to the early stress response happening in liver, which was supported by evidence of early elevations in

AST, ALT, and cholestatic enzymes (GGT and ALP) during the infection. We supposed it may be mediated by inflammation, as proinflammatory cytokines can induce hepatocellular cholestasis by downregulating hepatobiliary uptake and excretory systems.[25] The modest elevations in liver enzyme levels will normalize rapidly in most individuals; however, patients exhibiting hepatocellular (ALT and/or AST > 2×ULN), cholestatic (GGT, ALP, and/or total bilirubin > 2×ULN), or mixed-pattern altered liver enzyme at admission may face a certain degree of risk for subclinical liver damage.

This retrospective investigation is subject to several limitations. Primarily, it is based on data from a single center, which may not be fully representative of the broader patient population, and is constrained by incomplete patient histories. Secondly, due to the challenges associated with emergency measurements, data such as the presence of underlying diseases were self-reported, potentially introducing recall biases. Thirdly, the study is limited by the availability of relevant research, and our focus is solely on regular markers, with all serum samples collected in the early stages of hospitalization. More specific indicators of liver function (like ASTm, ALT2, CER, and so on) were not included in the assessment. Lastly, there is no follow-up of patient liver ultrasound and liver tissue biopsy results to assess liver function comprehensively. Future studies should conduct a comprehensive analysis of additional test results, and long-term follow-up studies on Omicron patients to explore the specific impacts on the liver function.

Conclusion

Our findings suggested that even among asymptomatic or mild patients with Omicron infection, there might be an occurrence of abnormally elevated levels of liver enzyme markers, signifying a certain degree of stress response happening in the liver. This abnormality is correlated with increased levels of inflammatory biomarkers (PCT, IL-6, and CRP). Male gender and higher age were also identified as contributing risk factors. We believed that this knowledge will play a crucial role in developing early intervention strategies to effectively address these risks. Also, prospective long-term follow-up studies are necessary to investigate the potential long-term consequences of Omicron infection.

Data Sharing Statement

The raw data supporting the conclusions of this article will be made available by the authors under reasonable circumstances.

Ethics Approval and Informed Consent

This study received approval from the Medical Ethics Committee of Tianjin First Central Hospital (Ethics Committee archiving No. 2022N052KY) and adhered to the principles outlined in the Declaration of Helsinki. A waiver of informed consent was granted by the ethics committee due to a retrospective design of identified data and a minimal risk involved. The data presented in this study do not allow for the identification of individuals. We are committed to ensuring the privacy and confidentiality of the subjects, or their anonymity, throughout the entire research process.

Acknowledgments

The authors thank the Tianjin First Central Hospital for the research support.

Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising, or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

Funding

This work was supported by the Tianjin Key Medical Discipline (Specialty) Construction Project (No. TJYXZDXK-015A) and Tianjin Key Science and Technology Project of Tianjin Science and Technology Bureau (21ZXJBSY00040).

Disclosure

The author(s) report no conflicts of interest in this work.

References

1. Beraud G, Bouetard L, Civljak R, et al. Impact of vaccination on the presence and severity of symptoms in hospitalized patients with an infection of the Omicron variant (B.1.1.529) of the SARS-CoV-2 (subvariant BA.1). *Clin Microbiol Infect*. 2023;29(5):642–650. doi:10.1016/j.cmi.2022.12.020
2. Papagiouvanni I, Kotoulas SC, Pataka A, et al. COVID-19 and liver injury: an ongoing challenge. *World J Gastroenterol*. 2023;29(2):257–271. doi:10.3748/wjg.v29.i2.257
3. Ali AM, Tofiq AM, Rostam HM, Ali KM, Tawfeeq HM. Disease severity and efficacy of homologous vaccination among patients infected with SARS-CoV-2 Delta or Omicron VOCs, compared to unvaccinated using main biomarkers. *J Med Virol*. 2022;94(12):5867–5876. doi:10.1002/jmv.28098
4. Ali AM, Rostam HM, Fatah MH, Noori CM, Ali KM, Tawfeeq HM. Serum troponin, D-dimer, and CRP level in severe coronavirus (COVID-19) patients. *Immun Inflamm Dis*. 2022;10(3):e582. doi:10.1002/iid3.582
5. Bertolini A, van de Peppel IP, Bodewes F, et al. Abnormal liver function tests in patients with COVID-19: relevance and potential pathogenesis. *Hepatology*. 2020;72(5):1864–1872. doi:10.1002/hep.31480
6. Li D, Ding X, Xie M, Tian D, Xia L. COVID-19-associated liver injury: from bedside to bench. *J Gastroenterol*. 2021;56(3):218–230. doi:10.1007/s00535-021-01760-9
7. Li T, Han M, Wang J, Zhou C, Mu H. Clinical characteristics and risks of the convalescent COVID-19 patients with re-detectable positive RNA test: a 430 patients with Omicron infected cross-sectional survey in Tianjin, China. *J Infect Public Health*. 2022;15(12):1409–1414. doi:10.1016/j.jiph.2022.11.011
8. Zhang ZJ, Che TL, Wang T, et al. Epidemiological features of COVID-19 patients with prolonged incubation period and its implications for controlling the epidemics in China. *BMC Public Health*. 2021;21(1):2239. doi:10.1186/s12889-021-12337-9
9. Alqahtani SA, Schattenberg JM. Liver injury in COVID-19: the current evidence. *United Eur Gastroenterol J*. 2020;8(5):509–519. doi:10.1177/2050640620924157
10. Paizis G, Tikellis C, Cooper ME, et al. Chronic liver injury in rats and humans upregulates the novel enzyme angiotensin converting enzyme 2. *Gut*. 2005;54(12):1790–1796. doi:10.1136/gut.2004.062398
11. Hoffmann M, Kleine-Weber H, Schroeder S, et al. SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor. *Cell*. 2020;181(2):271–280. doi:10.1016/j.cell.2020.02.052
12. Zhao S, Lin Q, Ran J, et al. Preliminary estimation of the basic reproduction number of novel coronavirus (2019-nCoV) in China, from 2019 to 2020: a data-driven analysis in the early phase of the outbreak. *Int J Infect Dis*. 2020;92:214–217. doi:10.1016/j.ijid.2020.01.050
13. Karlberg J, Chong DS, Lai WY. Do men have a higher case fatality rate of severe acute respiratory syndrome than women do? *Am J Epidemiol*. 2004;159(3):229–231. doi:10.1093/aje/kwh056
14. Klein SL, Flanagan KL. Sex differences in immune responses. *Nat Rev Immunol*. 2016;16(10):626–638. doi:10.1038/nri.2016.90
15. Ji D, Qin E, Xu J, et al. Non-alcoholic fatty liver diseases in patients with COVID-19: a retrospective study. *J Hepatol*. 2020;73(2):451–453. doi:10.1016/j.jhep.2020.03.044
16. Chen T, Wu D, Chen H, et al. Clinical characteristics of 113 deceased patients with coronavirus disease 2019: retrospective study. *BMJ*. 2020;368:m1091. doi:10.1136/bmj.m1091
17. Weber S, Hellmuth JC, Scherer C, Muenchhoff M, Mayerle J, Gerbes AL. Liver function test abnormalities at hospital admission are associated with severe course of SARS-CoV-2 infection: a prospective cohort study. *Gut*. 2021;70(10):1925–1932. doi:10.1136/gutjnl-2020-323800
18. Iheanacho CO, Enechukwu OH. COVID-19-associated liver injury, role of drug therapy and management: a review. *Egypt Liver J*. 2022;12(1):66. doi:10.1186/s43066-022-00230-y
19. Ali KM, Ali AM, Atta PM, Mahmood KI, Rostam HM. A study on the side effects caused by the Pfizer/BioNTech COVID-19 vaccine: focus on IgG antibodies and serological biomarkers. *Cent Eur J Immunol*. 2024;49(1):2–10. doi:10.5114/ceji.2024.136382
20. Bernal-Monterde V, Casas-Deza D, Letona-Gimenez L, et al. SARS-CoV-2 infection induces a dual response in liver function tests: association with mortality during hospitalization. *Biomedicines*. 2020;8(9):328. doi:10.3390/biomedicines8090328
21. Ali AM, Ali KM, Fatah MH, Tawfeeq HM, Rostam HM. SARS-CoV-2 reinfection in patients negative for immunoglobulin G following recovery from COVID-19. *New Microbes New Infect*. 2021;43:100926. doi:10.1016/j.nmni.2021.100926
22. Marconato M, Abela IA, Hauser A, et al. Antibodies from convalescent plasma promote SARS-CoV-2 clearance in individuals with and without endogenous antibody response. *J Clin Invest*. 2022;132(12). doi:10.1172/JCI158190
23. Dufour JF, Marjot T, Becchetti C, Tilg H. COVID-19 and liver disease. *Gut*. 2022;71(11):2350–2362. doi:10.1136/gutjnl-2021-326792
24. Fan Z, Chen L, Li J, et al. Clinical features of COVID-19-related liver functional abnormality. *Clin Gastroenterol Hepatol*. 2020;18(7):1561–1566. doi:10.1016/j.cgh.2020.04.002
25. Shafran N, Issachar A, Shochat T, Shafran IH, Bursztyn M, Shlomai A. Abnormal liver tests in patients with SARS-CoV-2 or influenza - prognostic similarities and temporal disparities. *JHEP Rep*. 2021;3(3):100258. doi:10.1016/j.jhepr.2021.100258