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Research Article

The Safety of Co-Administration of Antibiotics with Lopinavir/Ritonavir for COVID-19 Patients: A Retrospective Comparative Single-Center Study in Makkah

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Abstract

Background: SARS-CoV-2 triggered the COVID-19 pandemic, which resulted in millions of illnesses and deaths globally. A complex secondary respiratory bacterial infection is often associated with COVID-19. We administer antibiotics for COVID-19-related bacterial infections. The side effects and toxicities of selected antibacterial medicines for treating bacterial infections in COVID-19 patients need to be evaluated, as the safety of administering these drugs in COVID-19 is un-known. Widespread use during the pandemic raises concerns about side effects and drug interactions, highlighting the need for careful evaluation.

Methods: A total of 130 patients admitted to King Faisal Hospital in Makkah, Saudi Arabia, were included in this study. All patients received antiviral therapy with lopinavir and ritonavir, alongside antibacterial treatments, including azithromycin, vancomycin, and levofloxacin. Key laboratory parameters such as white blood cell count, hemoglobin, creatine kinase, liver enzymes, serum creatinine, blood urea nitrogen, lactate dehydrogenase, albumin, and bilirubin were measured upon admission and reassessed after 2–3 weeks of hospitalization.

Results: The administration of azithromycin led to increases in white blood cell count, liver enzymes, alkaline phosphatase, blood urea nitrogen, and bilirubin levels, while reducing hemoglobin and albumin levels. Levofloxacin significantly de-creased albumin and hemoglobin levels but caused no notable changes in other parameters. Vancomycin slightly reduced hemoglobin and albumin levels while in-creasing liver enzymes, serum creatinine, and blood urea nitrogen. Co-administration of lopinavir/ritonavir with antibiotics markedly amplified these side effects across the tested parameters.

Conclusion: Azithromycin, when combined with lopinavir and ritonavir to treat secondary pulmonary bacterial infections in COVID-19 patients, may heighten the risk of renal and hepatotoxicity. Daily administration of vancomycin posed a moderate risk of nephrotoxicity, whereas levofloxacin demonstrated the lowest overall risk among the antibiotics studied. These findings emphasize the need for careful antibiotic selection to reduce renal and hepatic risks in future pandemics.

INTRODUCTION

The severe acute respiratory syndrome coronavirus 2 (SARS - CoV - 2) led to the current pandemic of COVID - 19 disease, and millions of cases and deaths were reported worldwide. COVID-19 infection is transmitted mainly via the contaminated droplets that carry SARS - CoV - 2, produced by coughing or sneezing of an infected individual (Sharma et al., 2020).

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COVID-19 infection could lead to respiratory inflammation that is accompanied by an elevation in the inflammatory cytokines such as interleukin-1 β (IL-1 β), interleukin-6 (IL-6), and tumor necrosis factor-α (TNF-α). Moreover, some common symptoms were reported, such as fever, cough, muscle pain, diarrhea, respiratory difficulties, and loss of smell and taste (Costela -Ruiz et al., 2020). The complicated secondary respiratory bacterial infection is commonly associated with COVID-19 (Langford et al., 2020).

Various materials, including steroids, plasminogen, hydroxychloroquine, heparin, and ACE2, have been identified as promising candidates for treating SARS-CoV-2-related disorders when delivered through inhalation and nano formulation (Abdellatif et al., 2021). Clinical trials of many drugs were conducted for treating COVID-19, such as the antiviral therapy with lopinavir and ritonavir combination. Lopinavir/ritonavir combination is preciously used as the firstline treatment of the human immune deficiency virus (HIV). However, due to its higher burden and increased toxicity, it is no longer recommended to treat HIV (Chandwani & Shuter, 2008). The most common adverse effect of the lopinavir/ritonavir combination is moderate or severe diarrhea (Wegzyn et al., 2012), while Other common adverse effects include abdominal pain, vomiting, and rash. Additionally, Lopinavir/ritonavir combination could raise hepatic enzymes and may lead to hyperlipidemia (Bethesda, 2017). Lopinavir and ritonavir can cause several side effects, including gastrointestinal issues (nausea, vomiting, diarrhea), liver toxicity (elevated liver enzymes), cardiovascular problems (QT prolongation, elevated cholesterol), pancreatitis, and fat redistribution. They also have significant drug interactions due to their effect on the CYP3A4 enzyme. Regular monitoring is required to manage these risks (Al- Hadidi et al., 2021). In addition, due to their cardiotoxic effect it is preferred to avoid administering lopinavir/ritonavir combination to patients with a preexisting cardiac disease (Talasaz et al., 2020).

Bacterial infections are commonly associated with viral respiratory tract infections such as influenza (Morris et al., 2017). It is believed to be one of the most important causes of morbidity and mortality, necessitating antibacterial therapy intervention (Klein et al., 2016). Similarly, many studies reported the incidence of bacterial co-infections in patients infected with SARS - CoV - 2 (Langford et al., 2020). Although antibacterial agents are ineffective in treating COVID-19, they are prescribed in patients with suspected or documented COVID-19 for various reasons: (1) difficulty in excluding the presence of current bacterial co-infection; (2) the possible secondary bacterial infection during the illness. Previous data showed increased morbidity and mortality due to secondary bacterial infections during the influenza pandemics. Therefore, several guidelines suggest using empirical antibacterial agents for patients with severe COVID-19 (Alhazzani et al., 2020). Antibiotics like azithromycin, vancomycin, and levofloxacin are used in COVID-19 patients to treat bacterial co-infections but must be used cautiously. Risks include QT prolongation (azithromycin, levofloxacin), kidney toxicity (vancomycin), and antibiotic resistance. Safety depends on patient-specific factors and careful monitoring (Al- Hadidi et al., 2021).

However, this suggestion raises apprehensions about the overuse of antibacterial agents, which may lead to subsequent adverse effects and the development of bacterial resistance. Understanding the damaging effects of the overuse of empirical antibacterial agents for treating acute respiratory bacterial co-infection is crucial to minimizing the possible negative consequences. Besides, this information could significantly set the guidelines for practical antibiotic administration for patients with COVID-19. The secondary bacterial infections associated with COVID-19 could be caused by gram-positive bacteria, including methicillin-resistant Staphylococcus aureus (MRSA) and Enterococci species. These common nosocomial infections could cause ventilator-associated pneumonia (Sharifipour et al., 2020), necessitating vancomycin treatment. The most serious adverse effect of vancomycin is its ability to induce kidney nephron-tubular damage. For this reason, monitoring plasma levels of vancomycin must be carried out to ensure that the drug is clinically effective and to avoid nephrotoxicity. Levofloxacin is a fluoroquinolone antibacterial agent commonly used to treat nosocomial pneumonia and community-acquired pneumonia (Izadi et al., 2018). It is also used to treat acute urinary tract infection, bronchitis, acute prostatitis, and rhinosinusitis (Podder & Sadiq, 2019).

Combining lopinavir and ritonavir with antibiotics can significantly increase the side effects, particularly affecting blood parameters such as liver enzymes, kidney function, and electrolyte levels. This combination increases the risk of gastrointestinal issues, liver toxicity, and cardiovascular problems, requiring careful monitoring of blood tests and dosage adjustments (Purwati et al., 2021). However, the benefits of preventing bacterial infections in vulnerable COVID-19 patients may outweigh these risks. Secondary bacterial infections can complicate the clinical course of COVID-19, potentially leading to worse outcomes. Therefore, careful risk-benefit analysis is necessary, with close monitoring to manage side effects while ensuring effective treatment of bacterial infections (Ye et al., 2020).

The safety of combining lopinavir/ritonavir with antibiotics in COVID-19 treatment raises significant concerns due to potential adverse effects and drug interactions. Additionally, widespread use during the pandemic heightens the risk of toxicity and underscores the need for careful monitoring and judicious prescription practices to ensure patient safety.

Because the lack of data about the safety of administer-

ing antibacterial agents to manage accompanied bacterial infections in COVID-19 patients is insufficient, our retrospective study aimed to evaluate the adverse drug effects and toxicities of some selected antibacterial agents given for treating bacterial infections in COVID-19 patients. It was performed by comparing these toxicities in groups of patients when co - administered with the lopinavir/ritonavir. Based on the guidelines for clinical management of patients with COVID-19 published by the Saudi ministry of health (www.moh.gov.sa), several drugs could be prescribed to COVID-19 patients according to the severity of the disease and the presence of a preexisting complication.

MATERIALS AND METHODS

Population and Study Sample

This study was held at King Faisal hospital (Makkah, Saudi Arabia). According to data obtained from the Saudi ministry of health, about 88,804 cases were reported in Makkah province till February 2021, about 86,060 cases were successfully treated, and 2,402 deaths were recorded (www.moh.gov.sa). Many of the recovered cases were treated in King Faisal hospital. The study explored the hospital records of COVID-19 patients admitted between March 2020 and January 2021.

Demographic and clinical characteristics

The investigated data included age, sex, body mass index.

(BMI), admission to intensive care unit (ICU), intubation, deaths, presence of combined comorbidities such as diabetes, cardiac disease, and hypertension in an existing secondary bacterial infection. Moreover, we studied the treatment regimen given to COVID-19 patients, including the selected antibacterial agent such as azithromycin, levofloxacin, vancomycin, and antiviral therapy. Finally, we check the occurrence of possible adverse effects, focusing on the adverse reactions caused by antibacterial agents and their potential role in worsening the symptoms of COVID-19.

130 eligible patients were enrolled in the study (Table 1). All patients were diagnosed with COVID-19 with respiratory tract infection and were admitted to King Faisal hospital (Makkah, Saudi Arabia) between March 2020 and February 2021. The age of patients ranged from 20 to 90 years, with an average age of 53.59 years. Most patients (45.38%) were between 41 and 60 years. 30% were females, and 70% were males. Regarding body mass index, 44.62% had normal body mass index, 36.92% were overweight, and 18.46% were obese. Most patients were admitted to the intensive care unit (83.85%).

Concomitant chronic diseases such as diabetes, hypertension, and cardiac diseases were identified in 72.31% of patients. 37.69 % of patients with chronic disease were hypertensive, 64.62 % were diabetic, and 17.69 % had cardiac disease. 32.31% of patients suffered from two or more chronic conditions (Table 1).

Sample Size and Selection of Sample

According to the sample size calculator (https://www.calculator.net/sample-size-calculator.html). The data was collected from 130 patients to avoid any misfit data. Records of patients with an age of more than 20 years old were included in the study.

Data Collection

After admission to the hospital, a full report about the physical and clinical manifestations of the patient was collected in an investigation form approved by the Sau-di ministry of health. The common COVID-19 symptoms were checked and reported in addition to the presence of any other signs or risk factors (diabetes, cardiac disease, hypertension, and secondary bacterial infection). Furthermore, a daily checkup is performed and recorded in contact tracing form, and the performance of a periodic examination and blood tests every other day. To differentiate between the clinical manifestation caused by COVID-19 and that caused by the continuous administration of antibacterial or antiviral drugs or their combination, we studied the patients' contact trac- ing form to determine whether the clinical manifestation appeared after drug administration or not.

We collected and tabulated data from medical records or patients' files who were admitted to the King Faisal hospital from March 2020 to January 2021. The age of the patient, sex, weight, high, smoking, presence of other complications (chronic disease such as hypertension, diabetes, and cardiovascular disease), laboratory tests such as white blood cells (WBC) count, hemoglobin (HGB), creatinine kinase (CK), aspartate transaminase (AST), alanine transaminase (ALT) alkaline phosphate (ALP), serum creatinine (SerCr), blood urea nitrogen (BUN), lactate dehydrogenase (LDH), albumin (ALB), and bilirubin (BIL). Data were taken after admission to the hospital and after 2-3 weeks. After completing data collection, we analyzed the safety of the administration of antibacterial agents to manage secondary bacterial infections in COVID-19 patients by investigating whether the antibacterial agents were in-volved in worsening the case and their involvement in the appearance of other people symptoms or adverse effects.

Inclusions and excluded criteria

A total of 130 patients diagnosed with COVID-19, confirmed via RT-PCR, were included in this study. All participants received antiviral therapy with lopinavir and ritonavir, along with antibacterial treatments comprising azithromycin, vancomycin, or levofloxacin. For the analysis, patients presenting with abnormal baseline laboratory values after admission were excluded, while those with normal values were included. The study focused on adults aged 20 years or older with complete medical records and baseline and follow-up

Table 1: Demographic and clinical characteristics

		Count	Percentage
Age	20 -40	26	20.00
	41 -60	59	45.38
	> 60	45	34.62
Sex	Males	91	70
	Females	39	30
BMI	Normal	58	44.62
	Overweight	48	36.92
	Obese	24	18.46
ICU admission	Admission to ICU	21	16.15
	No admission	109	83.85
Chronic Diseases	One chronic disease	52	40.00
	Two chronic diseases	19	14.62
	Three chronic diseases	23	17.69
	No chronic disease	36	27.69
	+ve cases	-ve cases	Percentage of +ve cases
Hypertension	49	81	37.69%
Diabetes	84	46	64.62%
Cardiac disease	23	107	17.69%

Table 2: Excluded and included patients

Parameter	Included patients	Excluded patients	Total
WBC	83	47	130
HGB	112	18	130
CK	130	0	130
ALT	120	10	130
AST	119	11	130
ALP	119	11	130
SerCr	116	14	130
BUN	130	0	130
LDH	130	0	130
ALB	130	0	130
BIL	126	4	130

laboratory data.

Exclusion criteria encompassed patients with preexisting chronic liver disease or renal failure, those who received antibiotics or antivirals outside the study protocol, pregnant or breastfeeding women, and patients with incomplete medical records or missing data. Additionally, individuals discharged or transferred to other facilities within 2–3 weeks of admission were excluded from the study.

Table 2 summarizes the number of included and excluded patients for the investigation of each parameter.

Data analysis

Data was collected and recorded in a datasheet using Microsoft Excel software (Redmond, Washington, USA). Data analysis was performed using GraphPad Prism 9 software (San Diego, California, USA) and IBM statistics SPSS software (Armond, New York, USA). For qualitative analysis, chi-squared test, Inde-pendent-samples t-test, and ANOVA were applied. The Cox regression model will be used in this study to determine mortality risk. Hazard ratio calculation is the ratio of the risk of outcome (adverse event or complication) in one group/risk of outcome in another group, occurring at a given interval. Pearson's correlation test was performed to study the correlation be-tween different parameters at specific periods.

Ethics and Human Subjects Issues

The study was conducted following the guidelines of the National Statement on Ethical Conduct in Human Research and after the acceptance of the Makkah Directorate ethics committee (H-02-K-076-0621-517). We prioritized respect for the dignity of research participants and took every precaution to ensure that they would not be harmed in any way during our research. Furthermore, we ensured that their privacy was protected. The research data was adequately protected against disclosure. We were careful not to mislead or exaggerate regarding the study's goals and purposes, and we considered the participants' right to remain anonymous. All potential conflicts of interest, funding sources, and affiliations were disclosed. Transparent and honest communication on the research was maintained, and we made sure that no biased data or misleading information was presented.

RESULTS

Effect of daily administration of antibiotics on WBC count

Eighty-three patients of 130 were included in this test whose WBC count was within the normal range. Patients with WBC count above the normal range (11 x1000/microliter) were excluded. Daily administration of one antibiotic, azithromycin or levofloxacin, led to a significant increase in WBC count after 2-3 weeks compared to the WBC count after admission (Fig. 1a, 1b). administration of vancomycin showed no significant elevation in WBC count (Fig. 1c). Similar results of WBC elevation were observed after concurrent administration of two or more antibiotics for 2- 3 weeks (Fig. 1d). The effect of single or multiple antibiotics therapies on WBC count after 3 weeks of treatment showed no significant difference (Fig. 1e). Additionally, there was no significant difference in WBC count levels between patients who received either levofloxacin, azithromycin, or vancomycin alone for 2-3 weeks. In diabetic and hypertensive patients, administering these antibiotics led to a significant elevation in the WBC count after 2-3 weeks of treatment (Fig. 1g,1h). Similar results were observed in patients who have no or multiple chronic diseases (Fig. 1i, 1j)

Effect of daily administration of antibiotics on hemoglobin level

Eighteen patients were excluded from this test as they were admitted with HGB levels below normal (less than 11 gm/dl). This study included 112 patients with normal HGB levels (11-17 gm/dl). Daily administration of a single antibacterial agent (azithromycin, levofloxacin, or vancomycin). In addition, lopinavir and ritonavir antiviral drugs significantly decreased HGB levels after 2-3 weeks of treatment but were still within the normal range (Fig. 2a, 2b, 2c). Similarly, comparing the HGB levels after admission to the hospital and HGB levels after 2-3 weeks of treatment with two or more different antibacterial agents in addition to lopinavir and ritonavir antivirals showed a marked de-crease in HGB levels (10.85 ± 0.43 gm/dl) slightly be-low the normal range (Fig. 2d). There was no significant difference between the lowering effect of single or multiple antibacterial agents' therapy on the HGB level (Fig. 2e). Similarly, there was no significant difference between the lowering effect of individual administration of azithromycin, vancomycin, or levofloxacin after 2-3 weeks (Fig. 2f). The HGB levels were significantly reduced in diabetic patients who received either azithromycin, levofloxacin, vancomycin, or combinations (Fig. 2g). Similar results were observed in patients with no or multiple chronic diseases (Fig. 2i, j). Surprisingly, there was no significant decline in HGB levels in patients with hypertension after antibiotics administration (Fig. 2h).

Effect of daily administration of antibiotics on Creatinine kinase levels

The normal range of creatinine kinase is between 55-170 U/L in males and 30-135 U/L in females. A high percentage of patients admitted to the hospital had creatinine kinase levels above the normal range (53 patients). It was reported that creatinine kinase is commonly elevated after COVID-19 infection (Orsucci et al., 2021). Therefore, all patients were included in this test. The creatinine kinase levels among the test group treated with a single antibacterial agent were 193.66 ± 20.62 U/L after hospital admission. Treatment with lopinavir and ritonavir in addition to antibiotic monotherapy for 2-3 weeks showed no significant elevation in creatinine kinase levels (Fig. 3a, 3b, 3c, 3f).

On the other hand, the test group that received antiviral agents and multiple antibacterial drugs had a creatinine kinase level of 229.55 ± 31.37 U/L, and the creatinine kinase level was not significantly elevated after antibiotics combined therapy for 2-3 weeks to reach (Fig. 3d). Moreover, there was no significant difference in creatinine kinase levels between both test groups after 2-3 weeks of the administration of antibiotics (Fig. 3e). Similarly, there was no significant difference in creatinine kinase levels after 2-3 weeks of antibiotics therapy in diabetic patients, antihypertensive patients, or patients with no or multiple chronic diseases (Fig. 3g-3j).

Effect of daily administration of antibiotics on alanine transaminase (ALT) and aspartate transaminase (AST) levels

One hundred twenty patients were included in studying the effect of antibacterial administration on ALT and AST levels in COVID-19 patients with secondary respiratory tract infection. These patients had a normal level of ALT and AST, 7-55 U/L and 10-40 U/L, respectively. Patients admitted to the hospital with elevated ALT and AST levels were excluded from this test. Treatment with azithromycin combined with ritonavir and lopinavir antivirals resulted in a significant elevation in ALT (Fig. 5a) and AST (Fig. 5a) within 2-3 weeks of treatment. Interestingly, the administration of levofloxacin or vancomycin did not result in significant elevation in ALT (Fig. 4b, 4c) or AST (Fig. 5b, 5c). However, the administration of multiple antibiotics with antiviral drugs led to a significant increase in ALT (Fig. 4d) and (Fig. 5d) levels compared to normal values. There was a significant difference in ALT and AST levels between patients who received a single antibiotic therapy and those who received multiple antibiotic therapies after 2-3 weeks of treatment (Fig. 4e, 5e). Patients who received various antibiotics had higher ALT levels and AST than patients treated with a single antibacterial agent. Comparing ALT and AST levels between patients who received either azithromycin, vancomycin, or levofloxacin showed no significant difference in their levels after 2-3 weeks of treatment

(Fig. 4f, 5f). Chronic diseases such as diabetes, hypertension, or multiple chronic diseases showed a marked elevation in ALT (Fig. 4g, 4h, 4j) and AST (Fig. 5g, 5h, 5j). Additionally, the administration of single antibiotic or multiple antibiotics in combination with ritonavir and lopinavir in COVID-19 patients with secondary respiratory bacterial infection led to a significant elevation in ALT and AST levels (Fig. 4i, 5i).

Effect of daily administration of antibiotics on alkaline phosphatase (ALP) levels

Eleven patients were excluded from this test because they were admitted to the hospital with high ALP levels (the normal level is 44-147 IU/L). One hundred nineteen patients were included in this test. Daily administration of azithromycin (not vancomycin or levofloxacin) significantly increased the ALP levels but remained within the normal range (Fig. 6a, 6b, 6c). Similarly, multiple antibiotic therapies significantly elevated ALP levels after 2-3 weeks (Fig. 6d). There was no significant difference between single or multiple anti-biotics therapies on ALP levels after 2-3 weeks of treatment (Fig. 6e, 6f). ALP levels were significantly elevated in patients with diabetes or multiple chronic diseases or even in patients with no chronic diseases after 2-3 weeks of treatment with antibacterial agents. However, there was no significant elevation in ALP levels in hypertensive patients compared to its levels after admission (Fig. 6g, 6h, 6i, 6j).

Effect of daily administration of antibiotics on serum creatinine (SerCr) levels

Fourteen patients were excluded from this test as they were admitted to the hospital with SerCr levels above the normal range (110 μmol/L). Administration of vancomycin (not azithromycin or levofloxacin) led to a significant elevation in SerCr levels but still within the normal range (Fig. 7a, 7b, 7c). On the other hand, administering two or more antibiotics showed a marked increase in SerCr levels above the normal levels (Fig. 7d). Patients who received multiple antibiotics for 2-3 weeks of treatment showed a significant elevation in SerCr levels compared with patients who received a single antibiotic (Fig. 7e). Comparing SerCr levels be-tween patients who received either azithromycin, levofloxacin, or vancomycin for 2-3 weeks showed no significant difference (Fig. 7f). Administration of anti-bacterial to patients with no chronic disease, diabetic patients, or multiple chronic diseases led to a consider-able elevation in SerCr levels after 2-3 weeks of treatment. However, SerCr levels were not significantly elevated in hypertensive patients (Fig. 7g, 7h, 7i, 7j).

Effect of daily administration of antibiotics on blood urea nitrogen (BUN) levels

All 130 patients were involved in this test as they had normal BUN levels after admission to the hospital (be-low 20 mg/dL). Although the administration of azithromycin, vancomycin, or multiple antibiotics led to a significant elevation in BUN levels (Fig. 8a, 8c, 8d), the levels were within the normal range. However, there was no substantial change in BUN levels after levofloxacin administration (Fig. 8b).

There was a significant difference in BUN levels between patients who received single antibiotic therapy and those who received multiple antibiotics after 2-3 weeks of treatment (Fig. 8e). Comparing BUN levels between patients who received either azithromycin, levofloxacin, or vancomycin for 2-3 weeks showed no significant difference (Fig. 8f).

BUN levels were not significantly elevated in hypertensive patients after 2-3 weeks of treatment with single or multiple antibacterial agents (Fig. 8h). On the other hand, a significant increase in BUN levels was observed in diabetic patients, patients with various chronic diseases, or patients with no chronic disease (Fig. 8g, 8i, 8j).

Effect of daily administration of antibiotics on Lactate dehydrogenase (LDH) levels

The normal range of LDH levels is 140-280 U/L. Most patients (107 patients) admitted to the hospital had an increased LDH level. Therefore, all patients were included in this test, where it was previously reported that COVID-19 infection is commonly associated with elevated LDH levels (Hariyanto et al., 2021). Patients treated with a single antibiotic of azithromycin, vancomycin, or levofloxacin combined with ritonavir and lopinavir had a high LDH level after admission. After administration of the antibiotics, the levels of LDH were not significantly increased (Fig. 9a, 9b, 9c, 9f). Patients who received multiple antibiotics were admitted to the hospital with an elevated LDH level, which was a significant increase after treating these patients with various antibiotics (Fig. 9d). Furthermore, there was a significant difference in LDH levels between patients who received a single antibiotic and patients who received various antibiotics after 2-3 weeks of treatment (Fig. 9e). There was no significant elevation in LDH levels either in the absence or presence of chronic diseases (Fig. 9g, 9h, 9i, 9j).

Effect of daily administration of antibiotics on albumin level

Hypoalbuminemia is a common symptom after COVID 19 and could be used to diagnose the infection. In our study, 116 patients had low albumin levels (below 34 g/dL) after admission to the hospital. Treatment with a single or multiple antibiotics led to a significant decrease in albumin levels in both treatment protocols (Fig. 10a, 9b, 10c, 10d). There was no significant difference in albumin levels between patients treated with a single antibiotic or patients treated with multiple antibiotics (Fig. 10e). However, there was a substantial difference in albumin levels in patients who received azithromycin for 2-3 weeks compared to those treated with vancomycin for the same period (Fig. 10f). Ad-

Table 3: Variables in the Equation

	B	SE	Wald	df	Sig.	Exp(B)
BMI	-.052	.019	7.555	1	.006	.949
WBC	-.011	.025	.200	1	.654	.989
HGB	-.026	.047	.299	1	.585	.974
Creatinine Kinase	.000	.000	.855	1	.355	1.000
ALP	.004	.001	9.635	1	.002	1.004
LDH	.000	.000	1.127	1	.288	1.000
ALT	.000	.000	1.408	1	.235	1.000
AST	.000	.000	1.040	1	.308	1.000
BIL	-.005	.005	.928	1	.335	.995
ALB	.101	.029	11.936	1	<.001	1.106
SerCr	-.001	.001	1.227	1	.268	.999
BUN	.033	.014	5.578	1	.018	1.033

ditionally, albumin levels were significantly reduced in patients with chronic diseases (Fig. 10g, 10h, 10i, 10j).

Effect of daily administration of antibiotics on bilirubin levels

In this test, 4 patients were excluded as they were admitted to the hospital with elevated BIL levels (more than 20 mg/dL). One hundred twenty-six patients were included. Multiple therapies or single therapy with azithromycin and the daily administration of ritonavir and lopinavir to COVID-19 patients with secondary respiratory tract infection showed a significant increase in BIL levels (Fig. 11a, 11d). There was no significant elevation in BIL in patients treated with levofloxacin or vancomycin (Fig. 11b, 11c). The BIL level was not significantly changed in patients who received multiple antibiotic therapies than in patients who received a single antibacterial agent for 2-3 weeks. However, both groups' BIL levels remained within the normal range (Fig. 11e, 11f). Administration of single or multiple antibiotics to patients with diabetes, hypertension, or numerous chronic diseases showed a significant elevation after 2-3 weeks of treatment. Additionally, similar results were observed in patients with no reported chronic disease (Fig. 11g, 11h, 11i, 11j). SerCr , BIL, LDH, or ALP levels and ALB levels.

The correlation between the administration period of antibiotics and biomarkers of chronic diseases

The correlation between the administration period of a single antibiotic or multiple antibiotics and other parameters showed that patients treated for less period had no chronic diseases (Fig. 12). Additionally, there is a weak negative relationship between the treatment period and other parameters such as WBC and LDH, AST, SerCr , and BUN levels. The relationship between the total treatment period with antibiotics and ALP was a moderate negative relationship. The increase in age showed a strong negative relationship with creatinine kinase levels and a reasonable negative relationship with LDH levels. The increase in BMI is usually associated with the presence of chronic disease. However, there was no observed relationship between BMI and other parameters except creatinine kinase, ALT, and AST levels, where a weak negative relationship was observed. A moderate positive relationship exists between one or more chronic diseases and WBC and BIL levels. The presence of chronic disease was associated with a mild increase in creatinine kinase and ALP levels in patients with COVID-19 and secondary bacterial infection treated with single or multiple antibiotics. A strong negative relationship was observed between WBC and HGB or ALB levels. Similarly, there was a strong positive relationship between WBC and ALP, LDH, BIL, SerCr , or BUN.

Moreover, it was observed that the decrease in HGB levels in patients with COVID-19 and secondary bacterial infection had a strong negative relationship with ALP, LDH, ALT, AST, BIL, SerCr , or BUN. There was a moderate positive relationship between creatinine and LDH levels, while the association of creatinine kinase levels with BIL and SerCr was weakly positive. LDH, ALT, AST, BIL, SerCr , and BUN showed a moderate to strong positive relationship toward ALP. Regarding the relationship between LDH levels and other variables, there was a strong positive relationship between BIL, SerCr , ALT, and AST levels. The increase in ALT or AST levels could be associated with an increased BIL level. Furthermore, there was a positive, strong relationship between BIL levels and SerCr or BUN levels. Interestingly, there is a strong negative relationship between ALB levels and BUN,

Survival Analysis

93% of patients included in our study died during the follow-up, while 6.9% of patients were still alive at the end of the study. No cases were reported with missing value.

The regression coefficient (B), which could predict the hazard of death, showed that CK, LDH, ALT, and AST had zero regression coefficient. BMI, WBC, HGB, BIL, and SerCr levels had a negative coefficient indicating their negative relationship to the hazard for the terminal event (patients' death) (Table 3).

Lower variables (SerCr and BIL levels) are associated with less survival time. On the other hand, ALP, ALB, and BUN levels had a positive coefficient indicating their positive relationship to the hazard of the terminal event (patients' death) (Table 3).

Higher variables (BUN levels) are associated with less survival time. Similarly, BMI, WBC, HGB, BIL, and SerCr variables were related to negative regression slopes (Exp(B)) value, while ALP, ALB, and BUN variables were associated with positive regression slopes. BMI, ALP, ALB, and BUN variables showed a significant effect on the survival time of patients, where elevation in these variables is associated with lower survival time.

Most patients died within 2-3 weeks of admission to the hospital, represented in the rapid decline of cum survival data between 14-19 days after entry (Fig. 13a).

Additionally, the hazard function at a mean of covariates was elevated along the study time (Fig. 13b).

DISCUSSION

The pandemic of COVID-19 in late 2019, caused by the new SARS-CoV-2, has resulted in a significant and long-term rise in hospitalizations of patients (Lin et al., 2020). Acute infection with SARS-CoV-2 led to a wide range of illnesses ranging from minor upper respiratory tract infections to life-threatening pneumonia. Many patients had secondary bacterial infections after hospital admission (Ortiz-Prado et al., 2020). Because COVID-19 patients often require lengthy hospitalization and prolonged antiviral and antibacterial therapies, serious adverse effects could be developed. Therefore, we aimed to investigate the risks of protracted administration of azithromycin, levofloxacin, and vancomycin in line with antiviral drugs, ritonavir, and lopinavir to treat the secondary respiratory tract infection in COVID-19 patients.

In our study, 55% of patients were obese or overweight. Obesity is a common health problem in Saudi Arabia. A survey conducted in 2016 showed that the percentage of obese children in Saudi Arabia was comparable to that of children in the United States of America, considering that America is one of the most obese countries (Brahim et al., 2016). Furthermore, data obtained from National Nutrition Survey confirmed that obesity reached 34% in some Saudi provinces (Al Othaimeen et al., 2007). Increased body weight and chronic diseases such as hypertension, diabetes, and cardiac diseases could increase the risk of respiratory tract infection and worsen the symptoms associated with COVID-19 infection (Zhou et al., 2021). This could explain the high percentage of overweight or obese patients and patients with chronic diseases with serious respiratory tract infections. COVID-19 infection is usually associated with an elevation in WBC count, as previously reported (Popov et al., 2020). Additional administration of the antibacterial agent, especially azithromycin, increased the risk of WBC count elevation. Moreover, azithromycin is believed to accumulate in WBC and improve its expression (Matzneller et al., 2013). The weak effect of levofloxacin on WBC count in COVID-19 patients with secondary bacterial infection could be attributed to its inhibitory effect on WBC expression (Wang et al., 2016). The presence of chronic disease could increase the risk of WBC elevation where raised WBC count was reported in patients with type 2 diabetes (Gkrania-Klotsas et al., 2010). Therefore, the follow-up of WBC count should be continually performed, especially in diabetic patients and after azithromycin administration.

Severe cases of COVID-19 could be associated with lower HGB levels (Taneri et al., 2020). Although azithromycin and vancomycin do not reduce HGB levels (Bloch et al., 2020; Dalla Costa et al., 1998; Hart et al., 2020), our study showed an opposite observation, where patients who received azithromycin showed a significant decrease in HGB levels after 2-3 weeks of treatment. However, levofloxacin is reported to induce autoimmune hemolytic anemia and reduce HGB expression (Oh et al., 2003). Anemia is commonly associated with diabetes due to the high burden of chronic renal disease on these patients (Thomas, 2007). Therefore, antibacterial administration to diabetic patients increases the risk of anemia .

Elevated creatinine kinase levels are considered an initial presentation of COVID-19, observed in our study (Chan et al., 2020). However, treatment with azithromycin, vancomycin, or levofloxacin did not cause a significant elevation in CK levels after 2-3 weeks of treatment. In our study, diabetic or hypertensive patients treated with antibacterial and antiviral agents showed no significant elevation in CK levels. However, diabetes could alter the plasma concentration of CK in some patients, as previously reported (Schwertner et al., 1979).

It was previously reported that the incidence of increasing serum aminotransferase plasma levels in hospitalized patients with COVID-19 ranged from 14% to 53%. Although these patients did not previously report any hepatic problems, hospitalized patients with COVID-19 had Abnormal liver enzymes, and increased serum levels of ALT, AST, and LDH (Ramachandran et al., 2020). For this reason, it is essential to follow up the administration of some hepatotoxic antibacterial and antiviral agents when administered to COVID-19 patients. Azithromycin-induced hepatic injury was reported after 1-3 weeks of treatment. Although most patients are completely recovered after treatment cessation, some patients have persistent hepatic damage and severe cutaneous responses, which may lead to death or the requirement of liver transplantation (Martinez et al., 2015). Diabetes may be associated with increased oxidative stress and steatosis without affecting liver enzymes (Nannipieri et al., 2005).

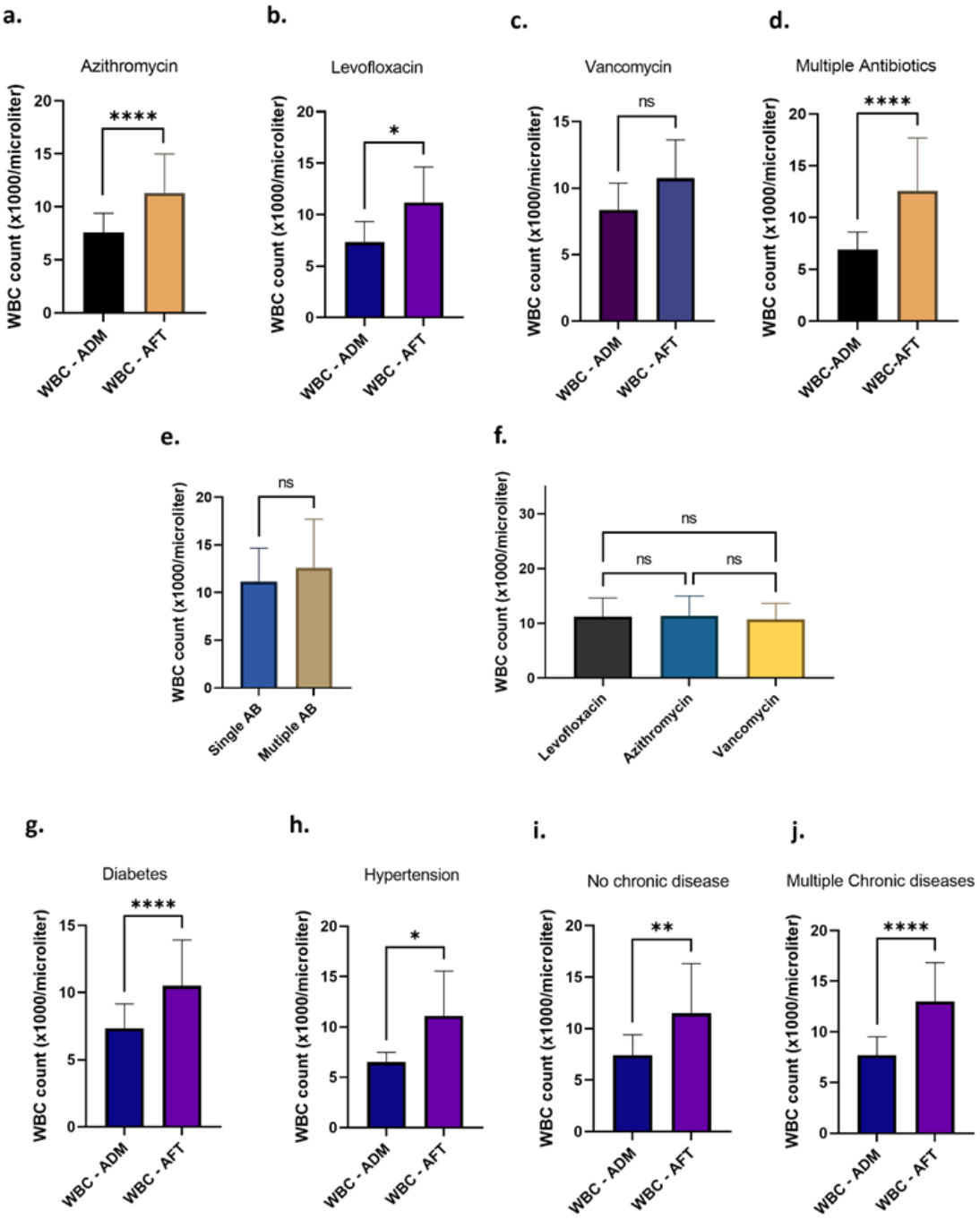

Figure 1: Effect of daily administration of antibiotics with lopinavir and ritonavir on WBC count

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** WBC count in patients who received azithromycin **(n = 39).b.** WBC count in patients who received levofloxacin **(n = 8). c.** WBC count in patients who received vancomycin **(n = 10). d.** WBC count in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on WBC count after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on WBC count after 2-3 weeks of treatment. **g.** WBC count in diabetic patients who received antibacterial therapy **(n = 26).h.** WBC count in hypertensive patients who received antibacterial therapy **(n = 5). i.** WBC count in patients with no chronic disease and received antibacterial therapy **(n = 21). j.** WBC count in patients with multiple chronic diseases and received antibacterial therapy **(n = 31).**Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ****p 0.0001 = WBC count after 2-3 weeks (WBC – AFT) vs. WBC count after admission (WBC – ADM); **p 0.01 = WBC-AFT vs WBC-ADM; ns = non-significant.

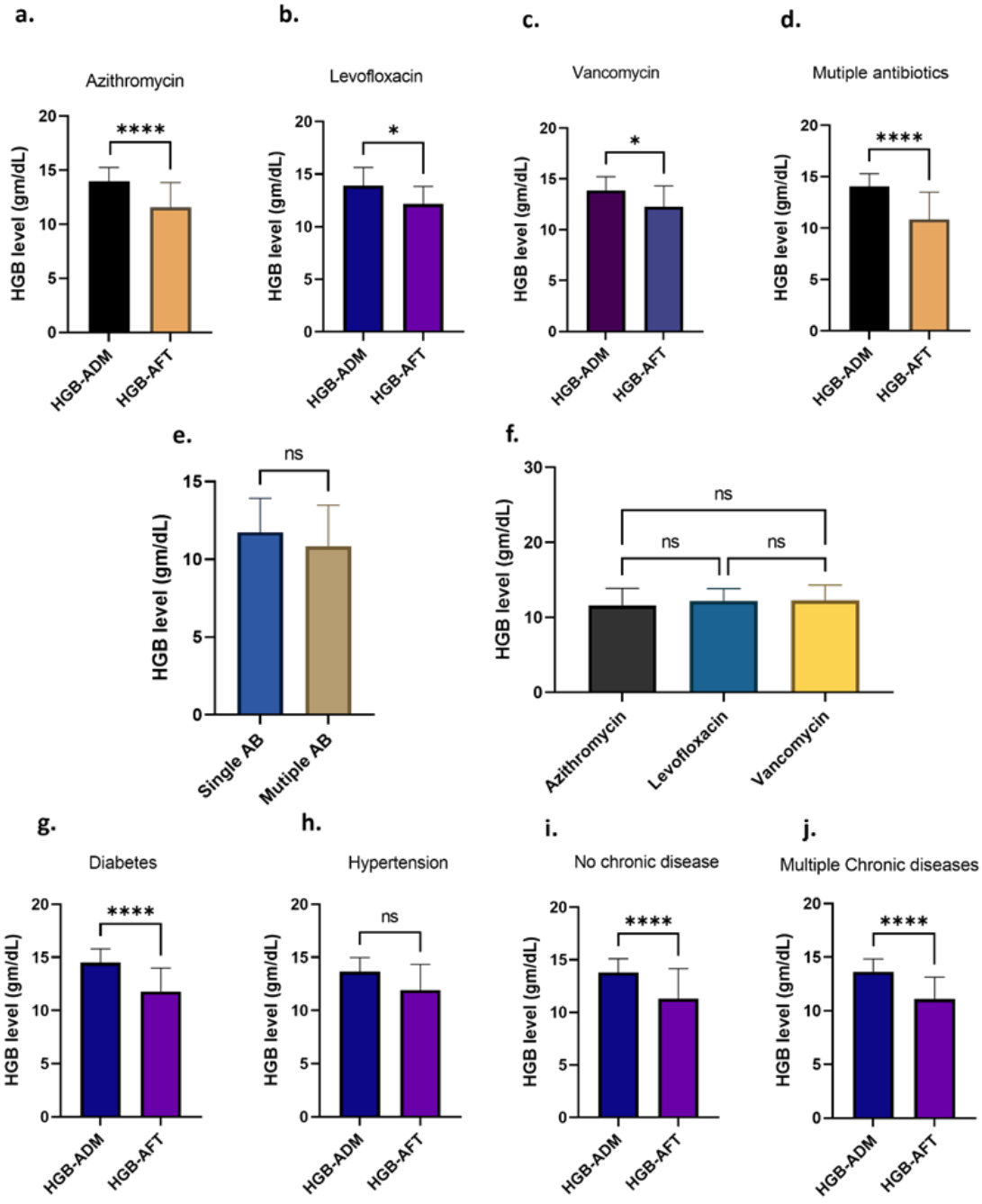

Figure 2: Effect of daily administration of antibiotics with lopinavir and ritonavir on hemoglobin levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** HGB levels in patients who received azithromycin **(n = 56). b.** HGB levels in patients who received levofloxacin **(n = 9).c.** HGB levels in patients who received vancomycin **(n = 10). d.** HGB levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on HGB levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on HGB levels after 2-3 weeks of treatment. **g.** HGB levels in diabetic patients who received antibacterial therapy **(n = 38).h.** HGB levels in hypertensive patients who received antibacterial therapy **(n = 5). i.** HGB levels in patients with no chronic disease and received antibacterial therapy **(n = 31). j.** HGB levels in patients with multiple chronic diseases received antibacterial therapy **(n = 38).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ****p 0.0001 = HGB levels after 2-3 weeks (HGB– AFT) vs. HGB levels after admission (HGB– ADM); *p 0.05 = HGB-AFT vs HGB-ADM; ns = non-significant.

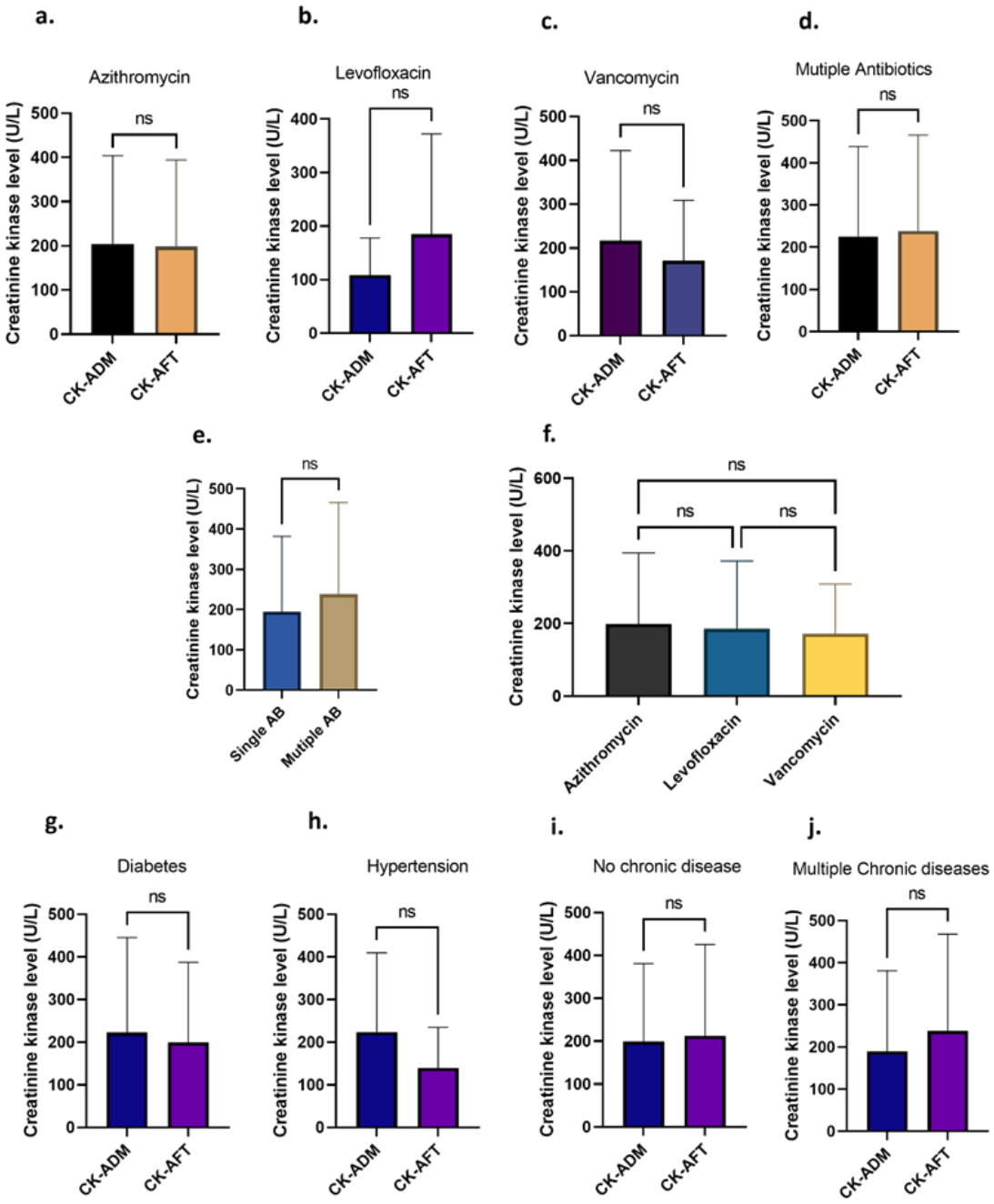

Figure 3: Effect of daily administration of antibiotics with lopinavir and ritonavir on creati-nine kinase (CK) levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with ritonavir and lopinavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** CK levels in patients who received azithromycin **(n = 63).b.** CK levels in patients who received levofloxacin **(n = 11). c.** CK levels in patients who received vancomycin **(n = 11). d.** CK levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on CK levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on CK levels after 2-3 weeks of treatment. **g.** CK levels in diabetic patients who received antibacterial therapy **(n = 45).h.** CK levels in hypertensive patients who received antibacterial therapy **(n = 7). i.** CK levels in patients with no chronic disease received antibacterial therapy **(n = 36). j.** CK levels in patients with multiple chronic diseases received antibacterial therapy **(n = 42).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ****p 0.0001 = CK levels after 2-3 weeks (CK–AFT) vs. CK levels after admission (CK–ADM); *p 0.05 = CK-AFT vs CK-ADM; ns = non-significant.

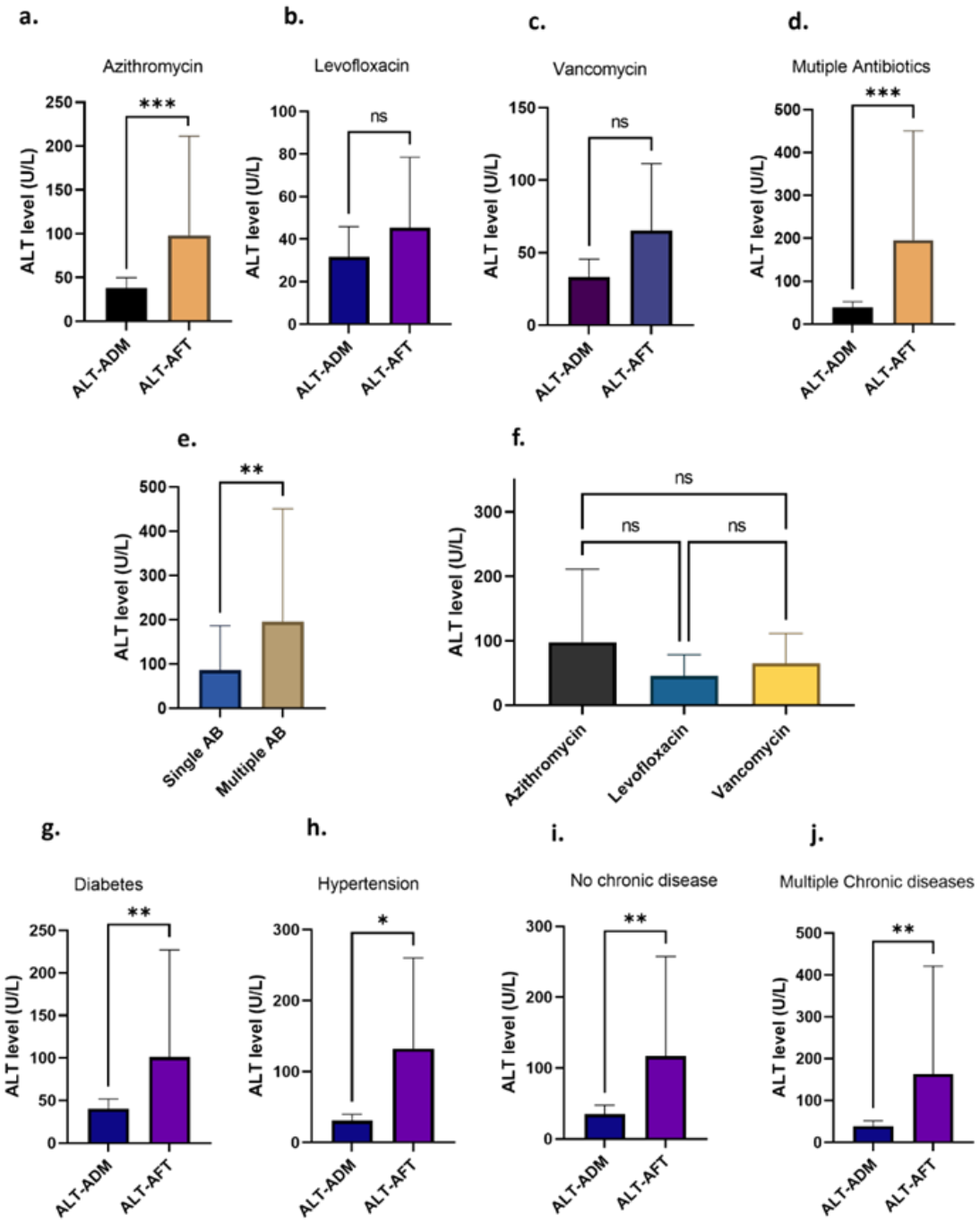

Figure 4: Effect of daily administration of antibiotics with lopinavir and ritonavir on alanine transaminase (ALT) levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** ALT levels in patients who received azithromycin **(n = 56). b.** ALT levels in patients who received levofloxacin **(n = 11). c.** ALT levels in patients who received vancomycin **(n = 10). d.** ALT levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on ALT levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on ALT levels after 2-3 weeks of treatment. **g.** ALT levels in diabetic patients who received antibacterial therapy **(n = 43).h.** ALT levels in hypertensive patients who received antibacterial therapy **(n = 7). i.** ALT levels in patients with no chronic disease and received antibacterial therapy **(n = 32). j.** ALT levels in patients with multiple chronic diseases received antibacterial therapy **(n = 38).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ***p 0.001 = ALT levels after 2-3 weeks (ALT–AFT) vs. ALT levels after admission (ALT–ADM); **p 0.01 = ALT-AFT vs ALT-ADM; *p 0.05 = ALT-AFT vs ALT-ADM; ns = non-significant.

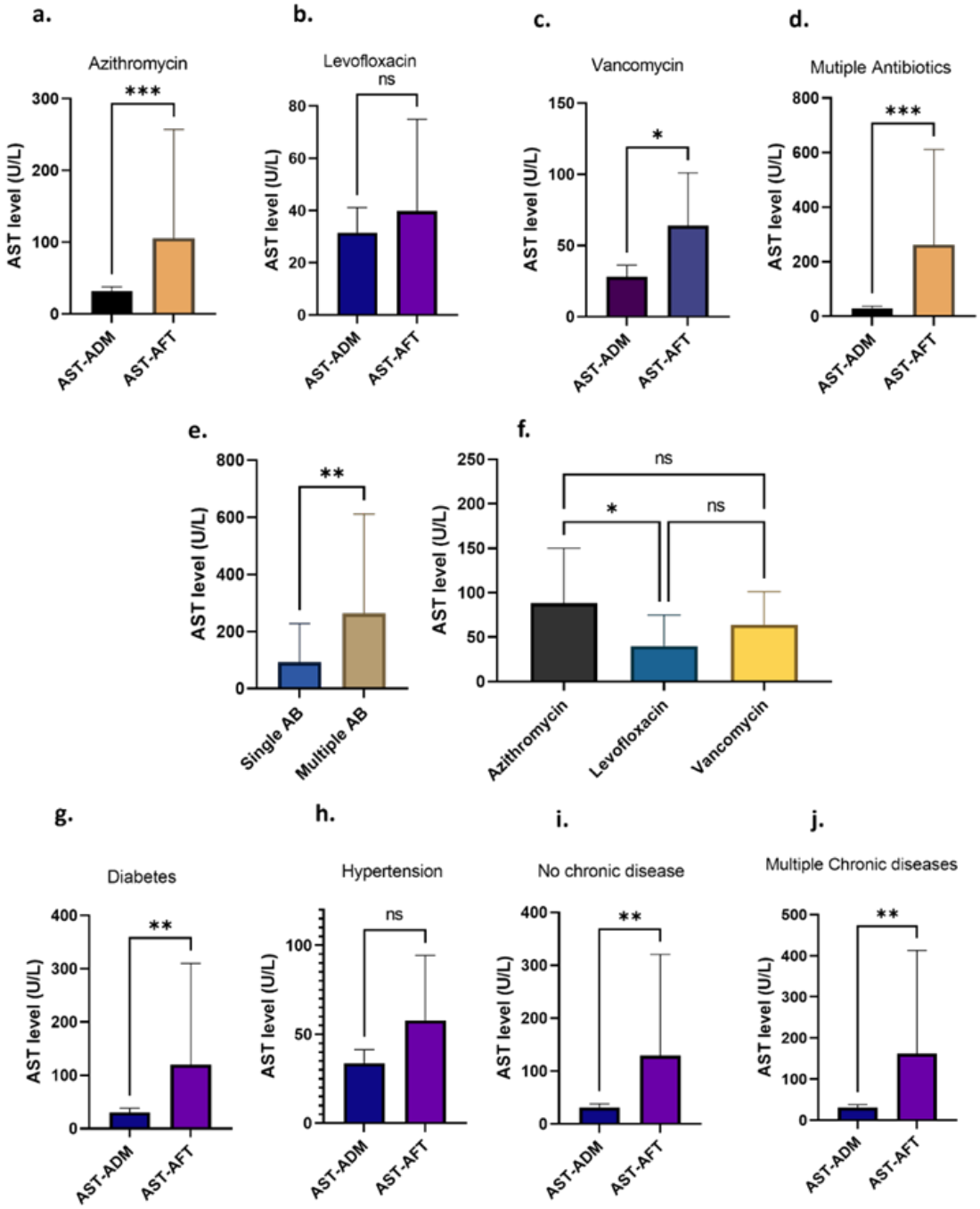

Figure 5: Effect of daily administration of antibiotics with lopinavir and ritonavir on aspartate transaminase (AST) levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** AST levels in patients who received azithromycin **(n = 60). b.** AST levels in patients who received levofloxacin **(n = 10). c.** AST levels in patients who received vancomycin **(n = 9). d.** AST levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on AST levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on AST levels after 2-3 weeks of treatment. **g.** AST levels in diabetic patients who received antibacterial therapy **(n = 43).h.** AST levels in hypertensive patients who received antibacterial therapy **(n = 6). i.** AST levels in patients with no chronic disease and received antibacterial therapy **(n = 33). j.** AST levels in patients with multiple chronic diseases and received antibacterial therapy **(n = 38).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ***p 0.001 = AST levels after 2-3 weeks (AST–AFT) vs. AST levels after admission (AST–ADM); **p 0.01 = AST-AFT vs AST-ADM; *p 0.05 = AST-AFT vs AST-ADM; ns = non-significant.

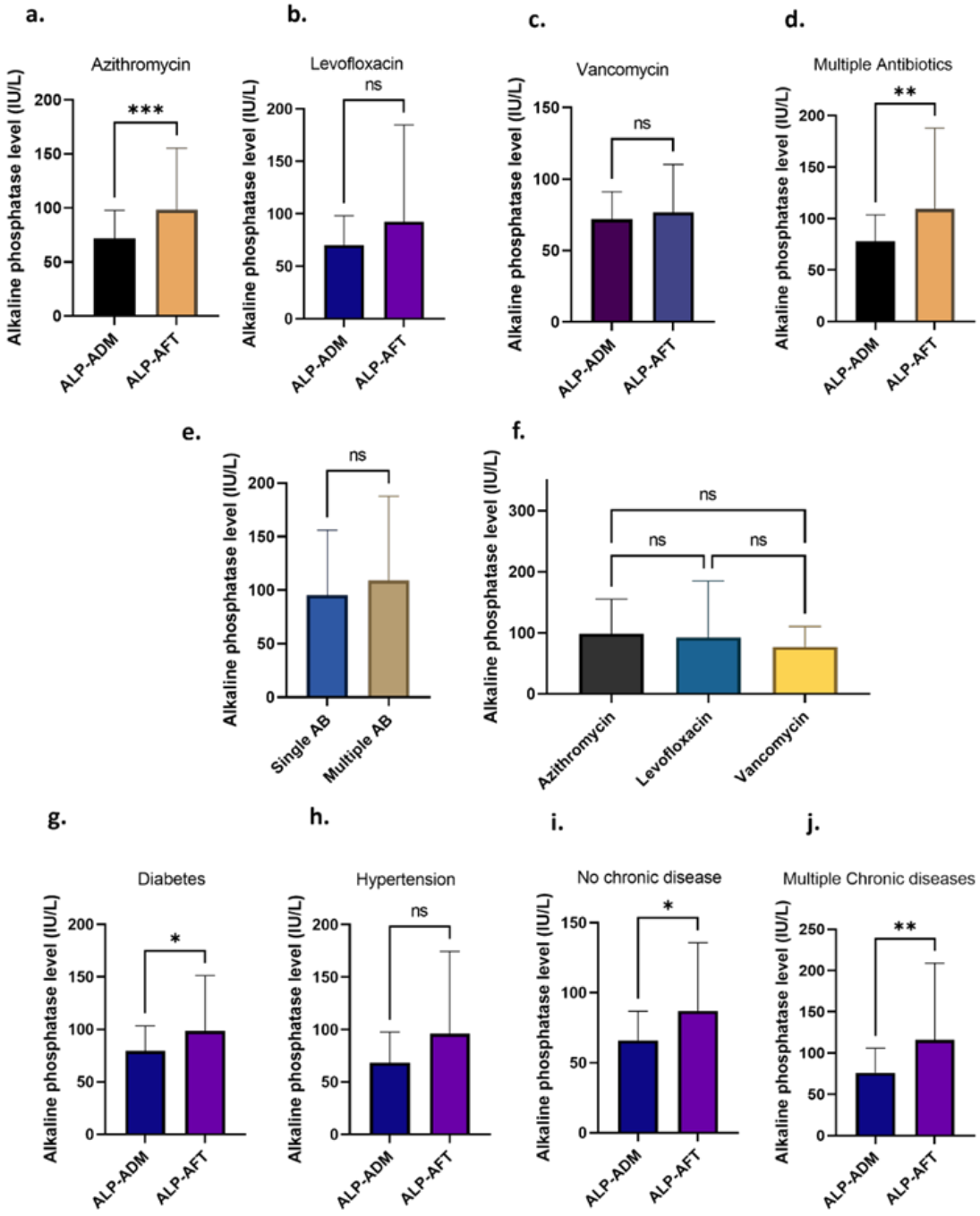

Figure 6: Effect of daily administration of antibiotics with lopinavir and ritonavir on alkaline phosphatase (ALP) levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** ALP levels in patients who received azithromycin **(n = 57). b.** ALP levels in patients who received levofloxacin **(n = 11). c.** ALP levels in patients who received vancomycin **(n = 8). d.** ALP levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on ALP levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on ALP levels after 2-3 weeks of treatment. **g.** ALP levels in diabetic patients who received antibacterial therapy **(n = 40). h.** ALP levels in hypertensive patients who received antibacterial therapy **(n = 5). i.** ALP levels in patients with no chronic disease and received antibacterial therapy **(n = 35). j.** ALP levels in patients with multiple chronic diseases and received antibacterial therapy **(n = 38).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ***p 0.001 = ALP levels after 2-3 weeks (ALP–AFT) vs. ALP levels after admission (ALP–ADM); **p 0.01 = ALP-AFT vs ALP-ADM; *p 0.05 = ALP-AFT vs ALP-ADM; ns = non-significant.

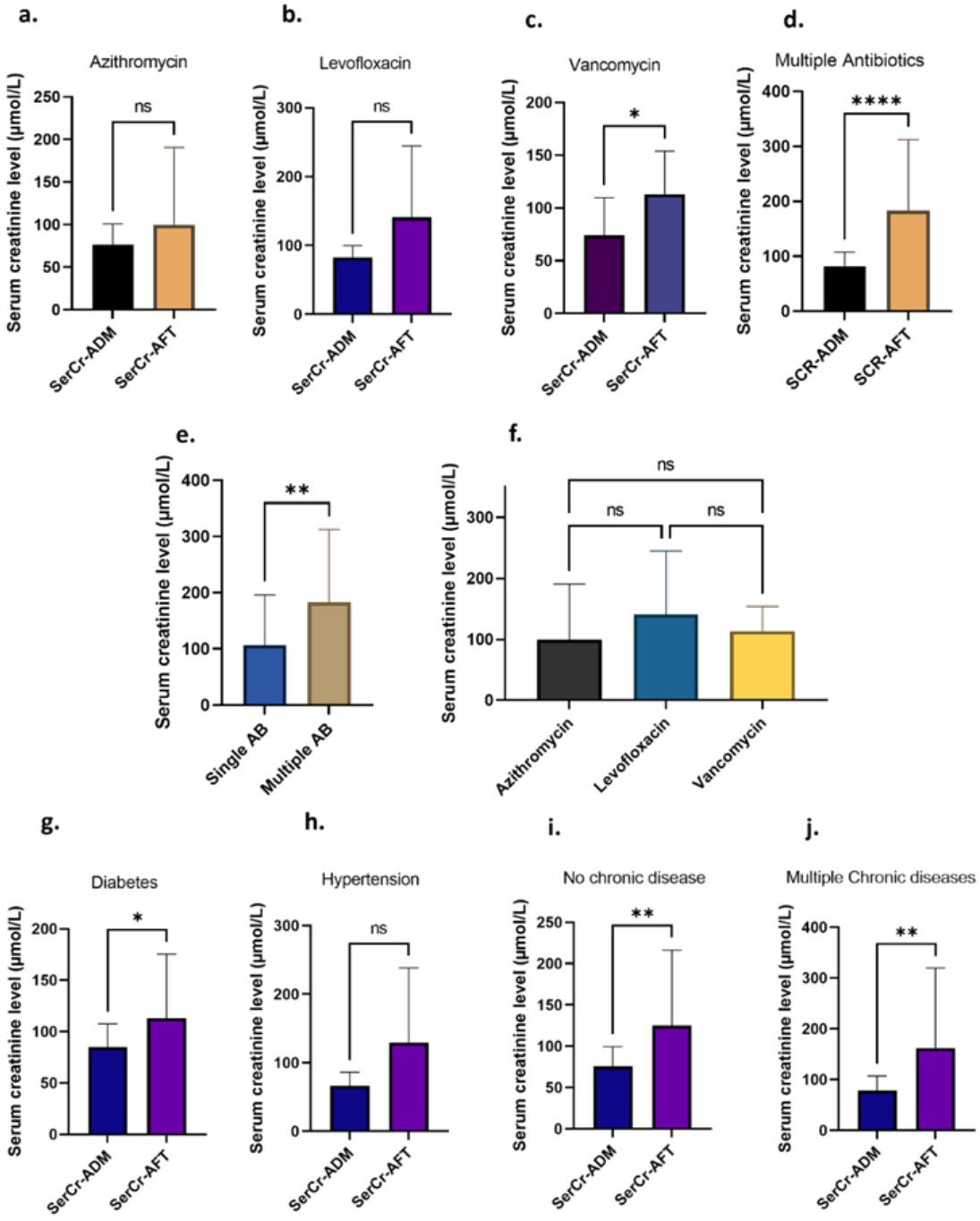

Figure 7: Effect of daily administration of antibiotics with lopinavir and ritonavir on serum creatinine (SerCr) levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** SerCr levels in patients who received azithromycin **(n = 57). b.** SerCr levels in patients who received levofloxacin **(n = 11). c.** SerCr levels in patients who received vancomycin **(n = 9). d.** SerCr levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on SerCr levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on SerCr levels after 2-3 weeks of treatment. **g.** SerCr levels in diabetic patients who received antibacterial therapy **(n = 38).h.** SerCr levels in hypertensive patients who received antibacterial therapy **(n = 7). i.** SerCr levels in patients with no chronic disease received antibacterial therapy **(n = 34). j.** SerCr levels in patients with multiple chronic diseases and received antibacterial therapy **(n = 37).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ****p 0.0001 = SerCr levels after 2-3 weeks (SerCr –AFT) vs. SerCr levels after admission (SerCr –ADM); **p 0.01 = SerCr -AFT vs SerCr -ADM; *p 0.05 = SerCr -AFT vs SerCr -ADM; ns = non-significant.

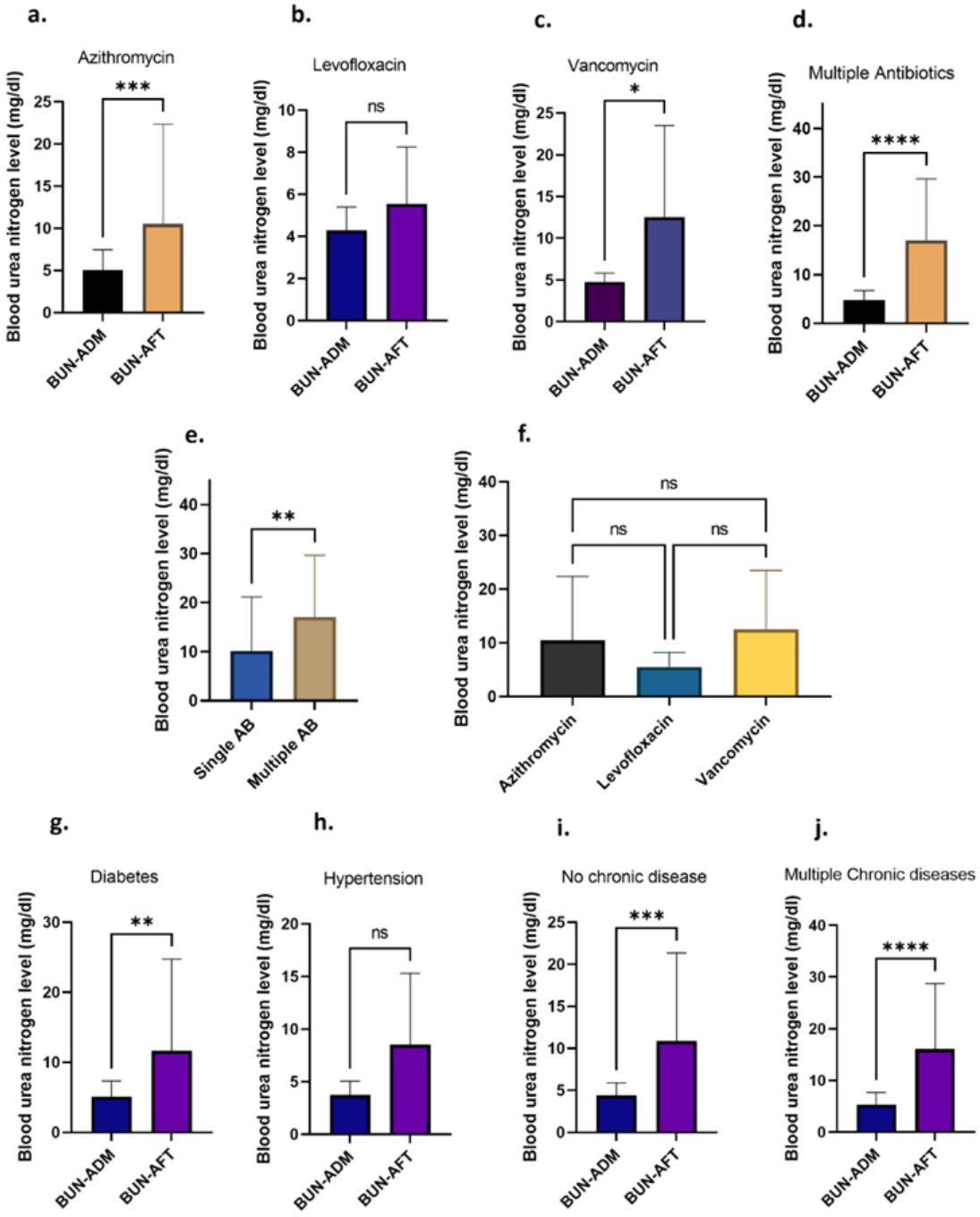

Figure 8: Effect of daily administration of antibiotics with lopinavir and ritonavir on blood urea nitrogen (BUN) levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** BUN levels in patients who received azithromycin **(n = 63). b.** BUN levels in patients who received levofloxacin **(n = 11). c.** BUN levels in patients who received vancomycin **(n = 11). d.** BUN levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on BUN levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on BUN levels after 2-3 weeks of treatment. **g.** BUN levels in diabetic patients who received antibacterial therapy **(n = 45).h.** BUN levels in hypertensive patients who received antibacterial therapy **(n = 7). i.** BUN levels in patients with no chronic disease received antibacterial therapy **(n = 36). j.** BUN levels in patients with multiple chronic diseases received antibacterial therapy **(n = 42).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ****p 0.0001 = BUN levels after 2-3 weeks (BUN–AFT) vs. BUN levels after admission (BUN–ADM); ***p 0.001 = BUN-AFT vs BUN-ADM; **p 0.01 = BUN-AFT vs BUN-ADM; *p 0.05 = BUN-AFT vs BUN-ADM; ns = non-significant.

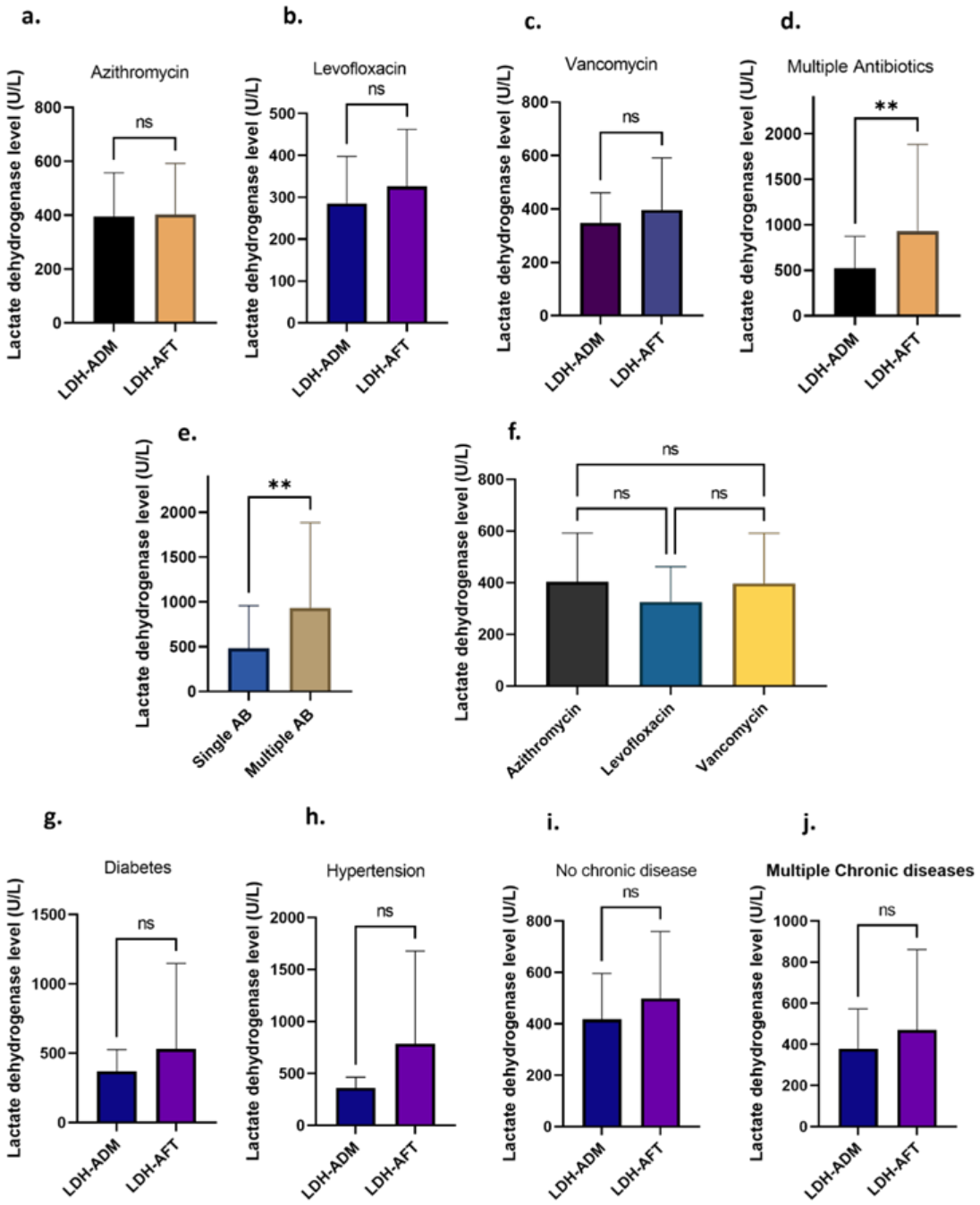

Figure 9: Effect of daily administration of antibiotics with lopinavir and ritonavir on lactate dehydrogenase (LDH) levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** LDH levels in patients who received azithromycin **(n = 63). b.** LDH levels in patients who received levofloxacin **(n = 11). c.** LDH levels in patients who received vancomycin **(n = 11). d.** LDH levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on LDH levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on LDH levels after 2-3 weeks of treatment. **g.** LDH levels in diabetic patients who received antibacterial therapy **(n = 45).h.** LDH levels in hypertensive patients who received antibacterial therapy **(n = 7). i.** LDH levels in patients with no chronic disease received antibacterial therapy **(n = 36). j.** LDH levels in patients with multiple chronic diseases and received antibacterial therapy **(n = 42).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. **p 0.01 = LDH levels after 2-3 weeks (LDH–AFT) vs. LDH levels after admission (LDH–ADM); ns = non-significant.

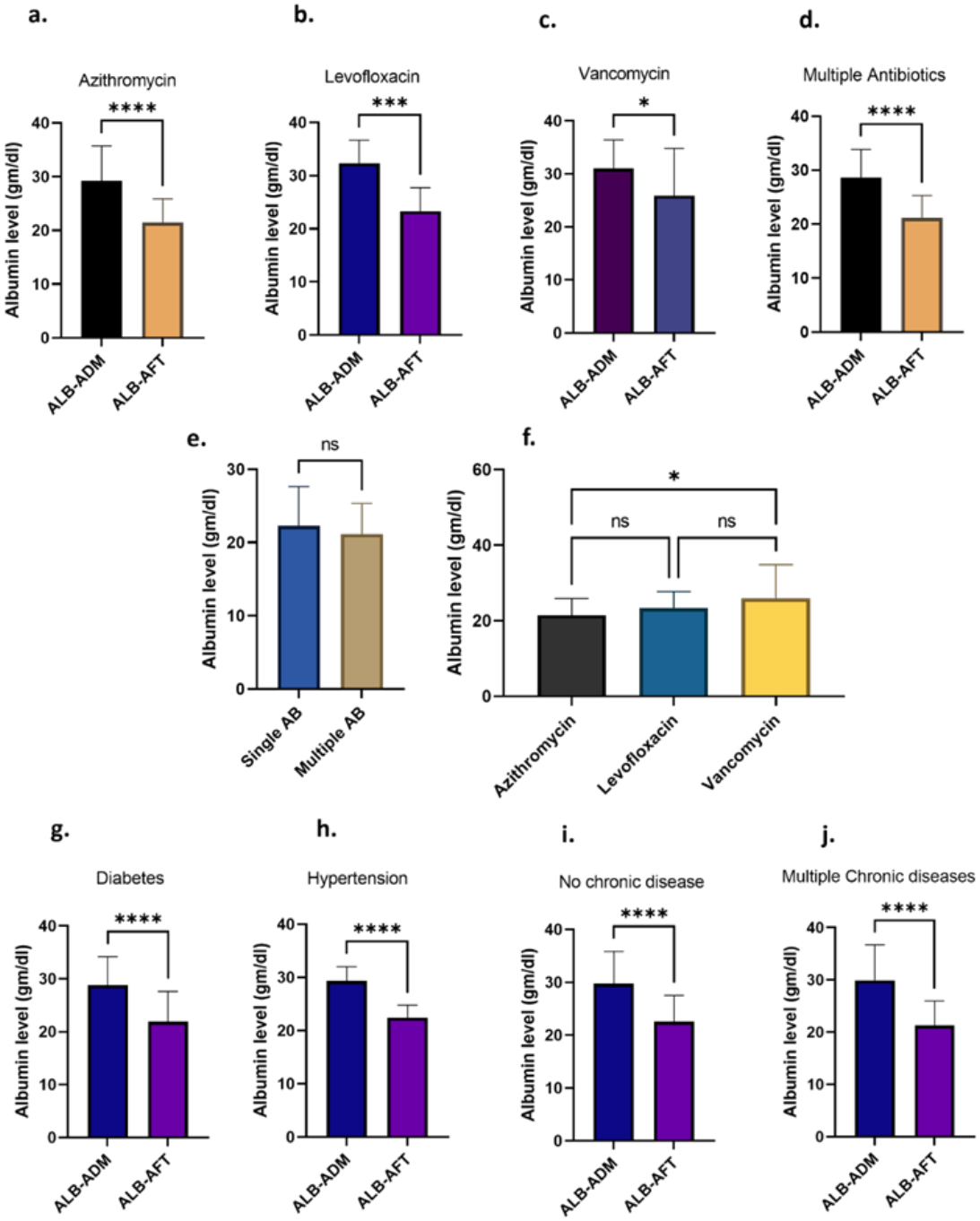

Figure 10: Effect of daily administration of antibiotics with lopinavir and ritonavir on albumin (ALB) levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** ALB levels in patients who received azithromycin **(n = 63). b.** ALB levels in patients who received levofloxacin **(n = 11). c.** ALB levels in patients who received vancomycin **(n = 11). d.** ALB levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on ALB levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on ALB levels after 2-3 weeks of treatment. **g.** ALB levels in diabetic patients who received antibacterial therapy **(n = 45). h.** ALB levels in hypertensive patients who received antibacterial therapy **(n = 7). i.** ALB levels in patients with no chronic disease and received antibacterial therapy **(n = 36). j.** ALB levels in patients with multiple chronic diseases received antibacterial therapy **(n = 42).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ****p 0.0001 = ALB levels after 2-3 weeks (ALB–AFT) vs. ALB levels after admission (ALB–ADM); ***p 0.001 = ALB-AFT vs ALB-ADM; *p 0.05 = ALB-AFT vs ALB-ADM; ns = non-significant.

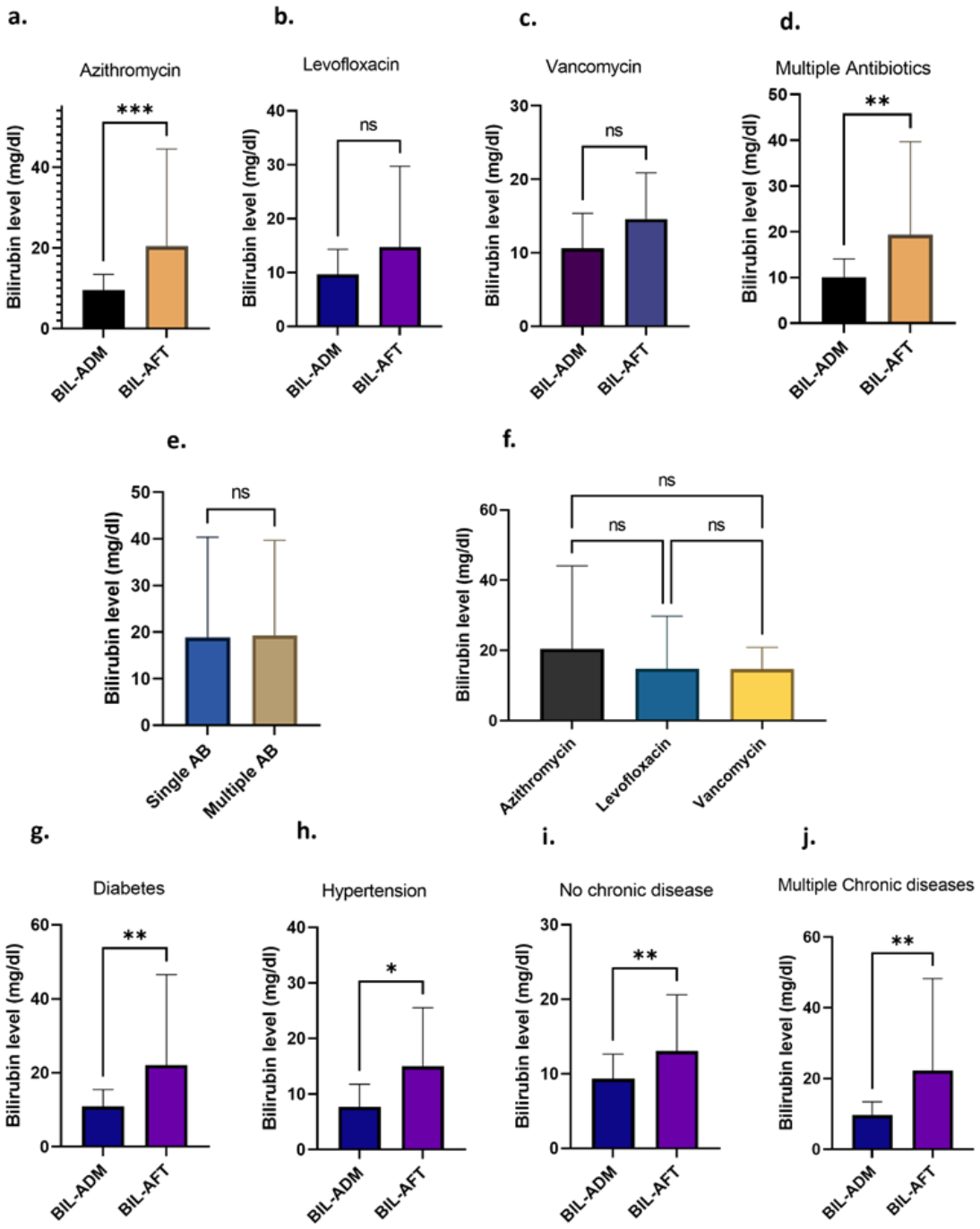

Figure 11: Effect of daily administration of antibiotics with lopinavir and ritonavir on Bilirubin (BIL) levels

Graphical presentation of the possible effect of daily administration of azithromycin, vancomycin, or levofloxacin with lopinavir and ritonavir to patients with COVID-19 and secondary bacterial infection for 2-3 weeks on **a.** BIL levels in patients who received azithromycin **(n = 61). b.** BIL levels in patients who received levofloxacin **(n = 11). c.** BIL levels in patients who received vancomycin **(n = 11). d.** BIL levels in patients who received multiple antibiotics **(n = 27). e.** comparison between the effect of single antibiotic therapy and multiple antibiotic therapies on BIL levels after 2-3 weeks of treatment. **f.**, compare the effect of different antibiotics on BIL levels after 2-3 weeks of treatment. **g.** BIL levels in diabetic patients who received antibacterial therapy **(n = 43).h.** BIL levels in hypertensive patients who received antibacterial therapy **(n = 7). i.** BIL levels in patients with no chronic disease received antibacterial therapy **(n = 36). j.** BIL levels in patients with multiple chronic diseases received antibacterial therapy **(n = 40).** Data are represented as mean ± SD. Statistical analysis was performed using paired and unpaired T-tests and one-way ANOVA. ***p 0.001 = BIL levels after 2-3 weeks (BIL–AFT) vs. BIL levels after admission (BIL–ADM); **p 0.01 = BIL-AFT vs BIL-ADM; *p 0.05 = BIL-AFT vs BIL-ADM; ns = non-significant.

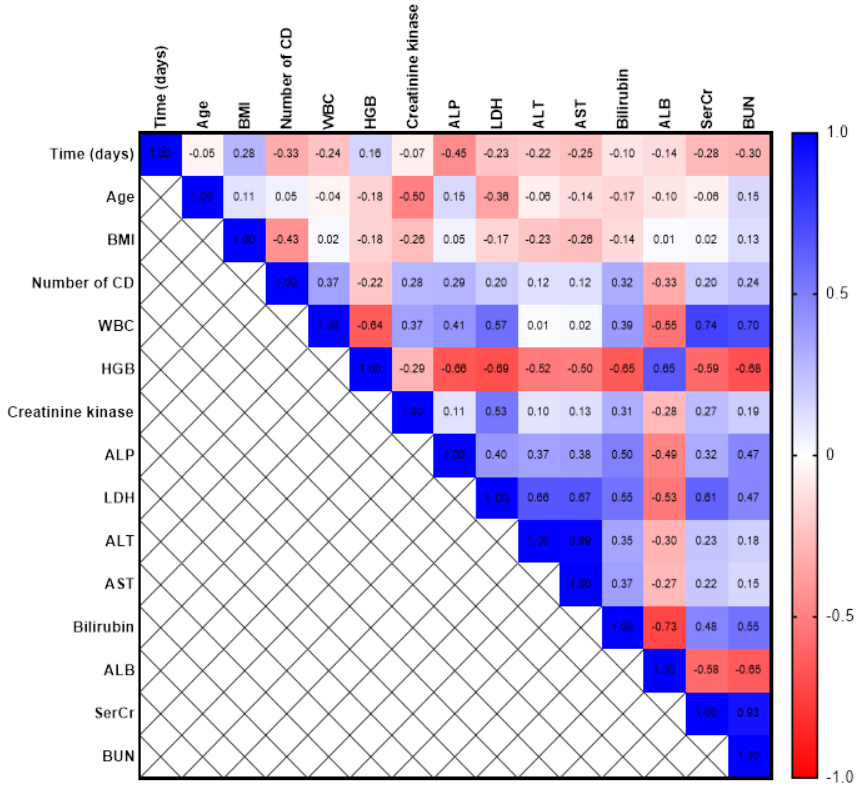

Figure 12: The correlation between the administration period of antibiotics with lopinavir and ritonavir and biomarkers of chronic diseases. The analysis of the relationship between different variable was performed by Pearson's correlation analysis. correlation value of r = 0 to 0.19 would be a negli-gible correlation, r = 0.2 to 0.29 would be a weak positive correlation, r = 0.3 to 0.39 would be a moderate positive correlation, r = 0.4 to 0.69 would be a strong positive correlation, r = 0.7 or higher would be a very strong positive correlation. r = 0 to - 0.19 would be a negligible correla-tion, r = - 0.2 to - 0.29 would be a weak negative correlation, r = - 0.3 to - 0.39 would be a mod-erate negative correlation, r = - 0.4 to - 0.69 would be a strong negative correlation, r = - 0.7 or higher would be a very strong negative correlation.

Similarly, hypertension could lead to a mild elevation in hepatic enzymes due to an increased risk of fatty liver formation (Donati et al., 2004). Therefore, administering azithromycin to diabetic or hypertensive patients could increase the risk of hepatic enzyme elevation. ALP is a cholangiocyte-related enzyme that remarkably increases after COVID-19 infection (Wu et al., 2020). ALP is distributed in the bile duct, bone, intestine, and kidneys. Increased ALP activity is usually associated with liver diseases.

Additionally, increased ALP levels may reflect pathological disorders in bone, kidneys, and endocrine systems (Fernandez & Kidney, 2007). Azithromycin-induced hepatic toxicity could lead to an elevation in ALP levels (Lockwood et al., 2010). Furthermore, diabetic patients had significantly higher ALP levels than normal healthy patients, which could be due to diabetic complications in those patients (Tibi et al., 1988). Decreased ALB levels are believed to indicate damage to synthetic hepatic function. Howerwev could indicate COVID-19 severity where pulmonary exudates and normal metabolism impairment associated with COVID-19 infection could lead to abnormal ALB distribution (Wu et al., 2020). It was previously reported that azithromycin does not reduce ALB levels after long-term administration (Alshehri et al., 2020; Breitschwerdt et al., 1999). However, in our study, ALB levels were significantly reduced after azithromycin, vancomycin, and levofloxacin treatment. This means that the reported decrease in ALB could be due to COVID-19 infection, and it is not related to the adverse effects of antibacterial therapy. Therefore, the possible risk of antibacterial administration to COVID-19 patients with a secondary bacterial infection on ALB levels needs further investigation. BIL levels increase in patients with severe or non-severe COVID-19 disease (Paliogiannis & Zinellu , 2020). In our study, only azithromycin led to an elevation in plasma levels of BIL, which could be due to its induced hepatotoxicity (Baciewicz et al., 2005). Previous studies confirmed that high levels of BIL cold be protectant against diabetes (Cheriyath et al., 2010), and it does not have any role in hypertension (Takeda et al., 2010). Therefore, the reported increase in BIL levels in patients with the chronic disease could be attributed to the hepatotoxic effects of azithromycin that was administered daily to these patients.

The prevalence of renal impairment in hospitalized

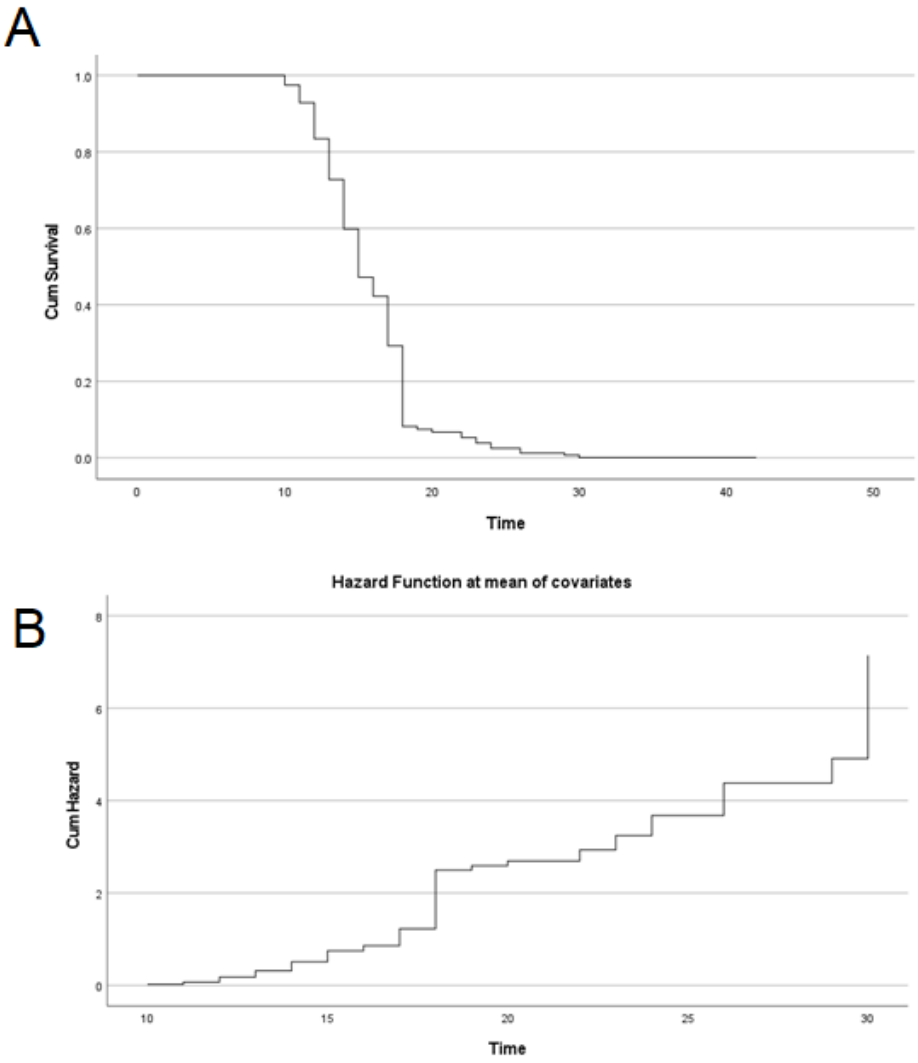

Figure 13: Survival function at a mean of covariates a) and hazard function at a mean of covari-ates b)

COVID-19 patients was high. In a study performed on 710 COVID patients, 44% were presented with proteinuria, 26% had hematuria , and acute kidney injury occurred in 3.2% of patients. SerCr levels were elevated in these patients (Cheng et al., 2020). It was also reported that COVID-19 patients who developed acute kidney disease had high levels of SerCr (10% increase) and BUN (14% increase). Additionally, they have a higher mortality risk (Li et al., 2020). Azithromycin and levofloxacin show a low risk of nephrotoxicity, which explains their weak effect on SerCr levels (Inage et al., 1992; Jeong et al., 2019). Vancomycin is a known nephrotoxic glycopeptide antibacterial agent (Rybak et al., 1990) as it causes interstitial nephritis and acute renal failure. Its administration could increase the risk of renal disorders in COVID-19 patients. In a previously reported case, administering vancomycin to treat pneumonia in COVID-19 patients led to an elevation in SerCr and BUN levels (Sise et al., 2020). In another study, 3 patients with COVID-19 showed a peak in SerCr and BUN after administering vancomycin (Na et al., 2020). The acute renal injury occurred after vancomycin intake, which points to the possible risk of its administration inducing acute renal injury. In people with diabetes, SerCr and BUN are commonly elevated, which could be related to increased blood glucose levels and diabetic complications (Shrestha et al., 2008). Therefore, administering vancomycin to diabetic patients with COVID-19 could increase the risk of acute renal failure.

Prolonged antibiotic use (azithromycin, levofloxacin, and vancomycin) with antivirals for COVID-19 secondary bacterial infections was discussed. These therapies were needed due to pandemic hospitalizations, demonstrating their clinical relevance. This study's findings help optimize treatment protocols, especially for high-risk groups like diabetics, the elderly, and those with preexisting organ dysfunctions, through tailored monitoring strategies.

Diabetics, who are prone to renal and hepatic complications, must have their kidney and liver function monitored regularly. Regularly checking liver enzymes and serum creatinine levels can detect toxicity early. This is

especially important for azithromycin, which has high hepatotoxicity, and vancomycin, which has moderate nephrotoxicity.

Baseline evaluations and scheduled follow-ups to monitor hemoglobin , albumin, and electrolyte levels may reduce adverse outcomes in elderly and comorbid patients. In drug-tolerant populations, levofloxacin may be preferred due to its lower risk.

In order to avoid complications and resistance, these findings suggest that COVID-19 antibiotic use should be more prudent. Antibiotic duration, dose adjustments for patients with compromised organ function, and alternative treatment integration should be clearly stated in protocols.

This study illuminates the safety concerns of these therapies and provides a framework for developing targeted monitoring strategies to improve patient out-comes, especially for vulnerable groups in future pandemics.

CONCLUSION AND RECOMMENDATIONS

COVID-19 is usually associated with secondary pulmonary infection, necessitating antibacterial agents' administration. Azithromycin, levofloxacin, and vancomycin are commonly administrated individually or combined with lopinavir and ritonavir. Our findings showed that administration of azithromycin increased white blood cell count, liver enzymes, alkaline phosphates, blood urea nitrogen, and bilirubin levels and reduced hemoglobin and albumin levels leading to an increased risk of hepatic and renal toxicities. Moreover, administering vancomycin slightly reduced hemoglobin and albumin levels and slightly increased liver enzymes, serum creatinine, and blood urea nitrogen levels, indicating a high risk of hepatotoxicity or nephrotoxicity. Levofloxacin showed the lowest risk in the liver and kidneys, slightly reducing albumin and hemoglobin levels and showing no significant changes in other parameters. Lopinavir and ritonavir with antibiotics increase side effects, especially blood parameters, but preventing bacterial infections in vulnerable COVID-19 patients is more important. Especially in critically ill patients, preventing secondary bacterial infections like sepsis or pneumonia can be beneficial. Hazards like liver damage, gastrointestinal issues, and cardiovascular complications require careful monitoring and tailored treatment. To avoid bacterial infections and manage side effects, these combinations should weigh the urgency of preventing infections against the risk of harm.

Clinical monitoring guidelines should be refined to manage and mitigate the risks associated with using these antibacterial agents. Special attention should be given to diabetic and hypertensive patients due to their higher susceptibility to drug-induced hepatic and renal complications

It is suggested that having a similar study should be expanded in multi hospitals to improve patient safety. Another similar research may also be needed to compare the effect of antibiotics agents on people infected with the COVID- 19 with antivirals agents is applied to different age groups and special cases, including pregnant women. Finally, it is suggested that using antimicrobial agent guidelines in COVID -19 with bacterial co-infection minimizes the risk of side effects.

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DECLARATIONS

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